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Life Science Journal

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Construction and Characterization of a Bacterial Artificial Chromosome Library from the Huoyan Goose

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Abstract: Huoyan goose is a famous Chinese local breed for its egg productivity. In this study, a bacterial artificial chromosome library of the Huoyan goose was successfully established using the Hind III site of the vector pBeloBAC11, comprising of 115, 200 clones arrayed in 543, 384 well microplates, with an average insert size of 102 kb and the content of the library was 11.4 genome equivalents, which yielded a theoretical probability of 99.93% for isolating a particular DNA sequence. BAC clones of the library were stable in the bacterial host for at least 100 passages.

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Keywords: Bacterial artificial chromosome library; Huoyan goose; genomic DNA

1. Introduction

Bacterial artificial chromosome (BAC) was one of the favorite vectors for cloning genomic DNA due to its large capacity, high clone stability and low DNA chimerism (Shizuya, 1992), bacterial artificial chromosome library, which could be also used in the study of comparative genomics (Jung, 2006), map position cloning (Quiniou, 2007), physical mapping (Tao, 2001; Zhebentayeva, 2008) and large-scale genome sequencing. Furthermore, single chromosome segregation technique has made the construction of specific chromosome become true (Gingrich, 1996).

The Huoyan goose was listed as one of the 78 nationally protected domestic animals by the Chinese government in the year of 2000. It was used for both meat and egg production, and was celebrated for its egg productivity during the winter. It has a genome size of approximate 1200 Mb/C. The establishment and characterization of Chinese Huoyan goose BAC library, our aim is not only to preserve this nationally protected breed resource at genomic level, but also supply valuable materials for positional cloning or genomic sequencing studies. Moreover, following the completion of whole genome sequencing of human and several model species, there was an ever-increasing demand for BAC libraries from species that were phylogenetic and/or biomedical importance.

2. Material and Methods

The Wing vein blood cells of Huoyan goose were embedded in agarose microbeads to prepare high molecular weight (HMW) genomic DNA (Ouyang, 2010). Quality of the prepared DNA was evaluated by pulse field gel electrophoresis (PFGE).

Genomic DNA was partially digested by three restriction enzymes Hind III, EcoRI and BamH I to determine the optimal partial digestion condition. The number of restriction enzyme units ranged from 15 to 70 units. Partially digested genomic DNA was subjected to size selection by two rounds of PFGE.

Size-selected genomic DNA was ligated to 30 ng prepared pBeloBAC11 in a molar ratio of 5:1. Ligation product was used to transform 20 µl E. coli Electro MAX DH10BTM cell suspension (Invitrogen) by electroporation. White colonies were picked with a Genetix robot (Genetix, New Milton, UK) in 384-well plates (Corning) containing LB freezing medium and incubated overnight at 37 °C. The whole library was stored at -80 °C.

Each of 400 randomly selected clones was incubated overnight in LB medium followed by an alkaline lysis procedure. The DNA pellet was dissolved in 50 µl TE and digested with 0.2 U Not I (New England Biolabs), followed by PFGE and photographed.

Clones were randomly picked from the library and incubated, primary cultures were considered as generation 0, Then the cultures were diluted 106 fold in 25 ml LB and incubated overnight. The process was repeated for 5 consecutive days, representing 100 generations. Samples were taken for analysis by Hind III digestion.

3. Results

An important aspect in large insert library construction is to gain HMW genomic DNA. Pulsed-field gel electrophoresis showed that the Huoyan goose genomic DNA was not degraded during preparation (Figure 1a).

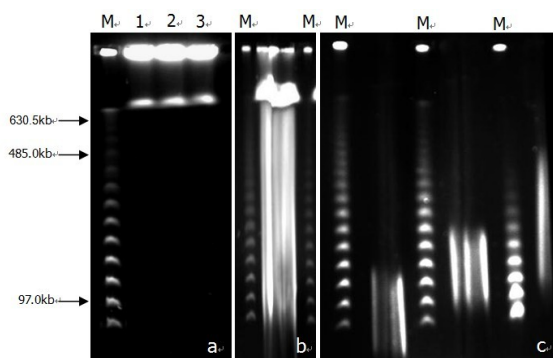


Figure 1. The preparation of HMW DNA from Huoyan goose detected by PFGE
 a, HMW genomic DNA, lanes 1-3 were DNA samples; b, PFGE result of recovered fragment by first recovery after partial digested genomic DNA; c, Second recovery after partial digested genomic DNA. M was Lambda Ladder PFG Marker.

It was necessary to determine the optimal conditions to generate the maximum concentration of DNA within the desired size range (Nilmalgoda, 2003). In this study, HMW genomic DNA was optimally digested with Hind III spreaded at 100-500 kb, then they were excised from the gel slices, the piece was cut into three pieces with sizes 100-200 kb, 200-300 kb and 300-500 kb, they were subjected to a second PFGE, excised, recovered, and dialyzed (Figure 1 b and c).

The recovered HMW DNA was quantified with λ DNA as control and its concentration was larger than 6.25 ng/ μ l. Desalting of the ligation reactions before transformation was found to improve transformation frequency two-fold, and thus was used frequently. In this study, 10% PEG8000 was used for concentration and we also got higher transformation efficiency (Osoegawa, 1998). The ligation solution should be used as soon as possible, or the large fragment DNA would degrade and aggregate.

Partially digested DNA was ligated to vector DNA, 9 ligations and 89 transformations were performed to complete the construction of the Huoyan goose BAC library. In total, 115, 200 clones were picked, deposited in 543 384-well microplates and stored at -80°C .

The percentage of non-insert clones was 0.8% among the 400 selected clones. Inserts varied from 48 to 150 kb with an average size of 102kb, corresponding to 11.4 genome equivalents, yielding a probability of 99.93% of isolating a particular DNA sequence.

The stability of the BAC library was analyzed by continuously incubating 5 random clones for 5 days. The result demonstrated that the restriction patterns of the 5 days were identical and the clones of the library were stable in the bacterial host for at least 100 generations (Figure 2).

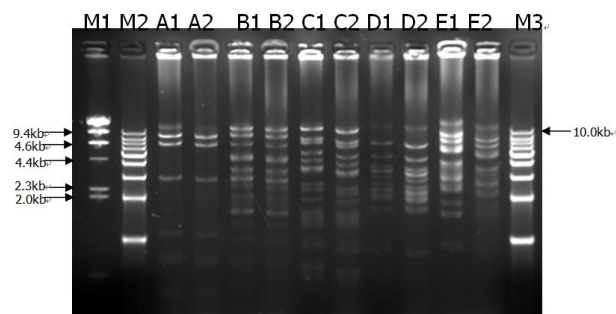


Figure 2. The stability detection of BAC library. M:1kb Marker; A1, A2, B1, B2, C1, C2, D1, D2, E1, E2 was digested by Hind III.

4. Discussions

An important aspect in large insert library construction is to gain HMW genomic DNA. During the preliminary preparation of goose mega-base-size DNA, a series of varied restriction digestions were performed using three restriction enzymes Hind III, EcoR I and BamH I. This was necessary to determine the optimal conditions to generate the maximum concentration of DNA within the desired size range (100-500 kb). In this study, high molecular weight genomic DNA (HMW) DNA was partially digested by three restrictive enzymes Hind III, EcoR I and BamH I. The results showed that HMW DNA was optimally digested with Hind III spreaded at 100-500 kb, so Hind III was the best. The gradient digestion showed that 40 U/ μ l Hind III was suitable.

Frijters used two enzymes in constructing a Lettuce BAC library to reduce chances of cloning bias owing to non-uniform distribution of restriction sites (Frijters, 1997). Similarly, Nilmalgoda constructed a hexaploid wheat library using the enzymes Hind III and BamH I (Nilmalgoda, 2003). Size distributions of randomly selected Hind III and BamH I clones were illustrated. Although the average insert size of the BamH I clones was slightly smaller than that of the Hind III clones, there had no significant difference. In our experiments, it showed that Hind III was the best for preparation of goose mega-base-size DNA. The most critical factor in achieving appropriate digestion was time which should not extend the digestion time longer than 40 min.

Pulsed-field gel electrophoresis was carried out using a double size selection strategy (Woo et al., 1994) in order to fractionate the goose HMW DNA. Previously studies have shown that two rounds of size selection eliminated small DNA fragments co-migrating with the selected range in the first pulsed-field fractionation (Cai, 1995). This strategy resulted in an increase in insert sizes and a more adqulis insert size distribution. However, the second sizes election obviously decreased the ligation and transformation efficiency (Wang, 2001). Similar losses after repeated size selection were also reported by Choi (Choi, 2000) and Nilmalgoda (Nilmalgoda, 2003). Nevertheless, O'Sullivan reported that single-step size selection was used to construct BAC library with good results (O'Sullivan, 2001). In our experiments, we have observed the insert size loss also, and insufficient DNA was obtained after the second pulsed-field fractionation to allow efficient ligation. Therefore, one-round purified DNA fragments were used to construct 80% of the Huoyan goose library, and double size selection procedure was used to remove the small DNA fragments usually trapped in the coils of the HMW DNAs.

The general parameters for assessing the quality of large insert libraries are the average insert size, the stability and integrity of inserts, the level of contamination by organelle DNA, the genome representation, and the potential of the library to provide a clone of interest (Moulet, 1999). In our study, the average insert size of the Huoyan Goose library was estimated as 102kb, which was a promising quality for the use of this library in large-scale sequencing and gene-screening projects. Size of insertion element in libraries varies with different investigation purposes. For large scale sequencing, the larger the insert size is, the better. If we want to clone a modicus gene, smaller insert size is enough. Likewise, the content of the libraries differs for different study purposes. When used for gene cloning and sequencing, higher genomic coverage was required (Zhang, 2000).

The BAC library we constructed from a female Huoyan goose consisted 115, 200 clones with an average insert size of 102 kb. Excluding the 0.6% empty clones, the coverage of this library was 11.4 genome equivalents. It was as demonstrated that the Huoyan goose genetic materials could be conserved by constructing and storing their BAC libraries at -80°C for a long term storage.

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The impact of corporate social responsibility on organizational performance

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Abstract: The purpose of this paper is to investigate the effects of corporate social responsibility (CSR) on firm performance. Data were collected via a questionnaire survey of star-rated hotels in Iran and a total of 100 valid responses were received. The hypothesis was tested by employing structural equation modelling with a maximum likelihood estimation option. It was found that CSR could enhance performance. This paper is one of the first to examine the effects of CSR on business performance. The empirical evidence from Iran adds to the existing literature on the respective importance of CSR. The main limitations include the use of cross-sectional data, the subjective measurement of performance and the uniqueness of the research setting (Iran).

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Keywords: Corporate social responsibility, organizational performance, Iran

1. Introduction

In today's changing business world, companies cannot be measured on profits alone. The external environment can play a major role in the perceived value and success of an organization. Managers should be concerned about social responsibilities, since it gives the company a right to exist based on their responsiveness to the external environment. Several outside stakeholders may influence the development of an organization's mission and goals. The modern era of corporate social responsibility (CSR) started from 1953 with Bowen's publication (Bowen, 2003), "Social Responsibilities of Businessman," according to Carroll (1979). Such CSR issues carried to tourism as a form of sustainability and have been investigated in the literature for the past several decades (Holden, 2000). In recent years, the significance of CSR for tourism-related industries has further increased (Kang, Lee, & Huh, 2010).

Corporate social responsibility is an important issue in contemporary international debates. In the past two decades, CSR appears to have become more ubiquitous and perceived as being relevant to corporations all over the world (Aras and Crowther, 2008). Moreover the link between CSR and business performance has become largely unquestioned. There have been various studies undertaken to investigate this important issue. Consequently much of the previous research regarding CSR deals with this issue and with the problems in the development of standards for managing and reporting such indeterminate activity. CSR is problematic as it is often perceived that there is a dichotomy between CSR activity and financial performance with one

being deleterious to the other and corporations having an imperative to pursue shareholder value. According to Morrisons (2005), a leading British supermarket chain, CSR is "about understanding and managing the relationship between our trading operations and the economy, environment and communities within which we operate". Morrisons claim that its CSR focus is on "managing the social, ethical and environmental issues that are material to our commercial performance, through a programme of continuous improvement". Corporate social responsibility (CSR) is one of the roles performed by the business world. This program is also aimed at encouraging business entities to run their activities ethically, minimizing bad effects on communities and the environment so that, ultimately, they can continue to carry on gaining economic benefit as their objective (Lesmana, 2007).

Corporate social responsibility has witnessed an ongoing debate not only among management theorists but also among industry practitioners. Sloan (1964) argues that "the strategic aim of a business is to earn a return on capital, and if in any particular case the return in the long run is not satisfactory, then the deficiency should be corrected or the activity abandoned for a more favourable one". Friedman (1962) also asserted that "the business of business is business and the only social responsibility is to increase profit". CSR can also be interpreted from the legitimacy perspective as firms' engage in socially responsible activities in an attempt to gain, improve or maintain legitimacy (Moir, 2001). Many studies have described the performance of socially responsible investments, with somewhat different results. Orlitzky et al. (2003) performed a meta-analysis of 52 studies

in search for the relationship between corporate social performance and corporate financial performance. The results confirm that socially responsible investing pays off. The relationship is strongest for the social dimension within corporate social performance. When isolating the environmental responsibility we come to the same conclusion but to a lesser extent. The key factor that initiates CSR is stakeholders' expectations that an investment decision should generate not only financial profit, but should also take into account the social and environmental aspect so that community welfare can be improved (Commission of the European Communities, 2001; both cited in Malovics et al., 2007). Some definitions state that CSR is the ethical, responsible and integrated business implementation applied to all operations (McWilliams and Siegel, 2001, cited in Wan-Jan, 2006; Business for Social Responsibility, 2003; cited in Jamali, 2006). Presently, however, the ideal definition if CSR is given by The World Business Council for Sustainable Development (2001): a business commitment that contributes to sustainable economic development through team work with employees and their representatives, their families, and local and public communities, to improve the quality of life by means of beneficial ways both for the business itself and for development (Jamali, 2006). Senior management faces a wide range of demands for Corporate Social Responsibility actions from different segments of society. Issues surrounding environmental quality, employee rights, community development, and diversity management all make claims on the attention of the contemporary business manager. In its broadest sense, CSR refers to "the firm's consideration of, and response to, issues beyond the narrow economic, technical, and legal requirements of the firm... (to) accomplish social benefits along with the traditional economic gains which the firms seeks.

2. Research background and hypothesis

2.1 Corporate social responsibility

CSR is defined as a "voluntary corporate commitment to exceed the explicit and implicit obligations imposed on a company by society's expectations of conventional corporate behavior" (Falck & Hebllich, 2007). For corporations, the adoption of CSR strategies can enhance their relationships with multiple stakeholders. Therefore, it is necessary to communicate CSR activities and use effective relationship management to satisfy stakeholders' expectations and achieve the expected goals of CSR initiatives (Clarke, 2000; Podnar, 2008). CSR has been defined as the duty of the organisation to respect individuals' rights and promote human welfare in its operations (Manakkalathil and Rudolf, 1995; Oppewal et al., 2006). Carroll and Buchholz (2000) stated that CSR encompasses the economic,

legal, ethical, and philanthropic expectations placed on organisations by society at a given point in time. CSR is a complex and diverse term representing business standards focusing on the long-term nature of business itself and increasing value for shareholders. Corporate social responsibility is not just an abstract concept but a business standard that we follow in our everyday activities. The observance of CSR principles is a prerogative for all company employees and not only for top management. This particular style of working gratifies and benefits all parties – the government, employees and their family members, consumers, local communities and shareholders. CSR refers to a company's voluntary activities "that appear to further some social good, beyond the interests of the firm and that which is required by law" (McWilliams & Siegel, 2001). Examples include the adoption of advanced human resource management programs, the reduction of environmentally hazardous substances, philanthropic activities, the production of products integrating social attributes, and support for local businesses (McWilliams & Siegel, 2001).

Corporate social responsibility means at a basic level – that "corporate activity should be motivated in part by a concern for the welfare of some non-owners, and by an underlying commitment to basic principles such as integrity, fairness and respect for persons" (Donaldson, 2005). In a general sense, social responsibility is management's acceptance of the obligation to consider profit, consumer satisfaction, and societal well-being of equal value in evaluating the firm's performance. It is the recognition that business must be concerned with the qualitative dimensions of consumer, employee, and societal benefits, as well as the quantitative measures of sales and profits, by which business performance is traditionally measured. Businesses may exercise social responsibility because such behavior is required by law, because it enhances the company's image, or because management believes it is the ethical course of action (Kurtz and Boone, 2008). CSR calls for corporations to take their social responsibilities as seriously as they pursue their economic objectives, and this applies to air transportation industries as to any other. There are four components in CSR; economic, legal, ethical and philanthropic responsibilities and these can be viewed as a process by which managers identify and accommodate the interests of those affected by their organization's actions. Isolating the appropriate CSR programs is difficult because there are multiple factors, such as program risk, corporate goals and limited CSR resources, at play. Corporate social responsibility is a form of management defined by the ethical and transparent relationship between a company and all the groups with which it relates, by the establishment

of corporate goals compatible with the sustainable development of society, preserving its environmental and cultural resources for the future generations, respecting diversity, and promoting the reduction of social inequality.

2.2 *organizational Performance*

Performance can be viewed in many aspects and connotations depend on the application. Derek, Torrington and Laura (1995) attributed performance as bottom line profit, doing better than competitors, maximum organization effectiveness and achieving specific organization objectives. In fact, Laitinen (2002) defined performance as the ability of an object to produce results in a dimension determined a priori, in relations to a target. OP is an indicator which measures how well an enterprise achieves their objectives (Hamon, 2003). Ho (2008) defined OP in terms of how well an organization accomplishes its objectives. Schermerhorn et al. (2002) point out that performance refers to the quality and quantity of individual or group work achievement. Delaney and Huselid (1996) suggest two ways to assess OP and market performance.

2.3 *Corporate social responsibility and organizational Performance*

Corporate social responsibility looks at how firms treat their stakeholders. One key stakeholder group that is frequently overlooked is the firm's shareholders. All too often, the corporate social responsibility literature focuses on customers, employees, and the natural environment, but rarely on shareholders. Corporate Social Responsibility is a term describing a company's obligations to be accountable to all of its stakeholders in all its operations and activities. Socially responsible companies consider the full scope of their impact on communities and the environment when making decisions, balancing the needs of stakeholders with their need to make a profit.

Many firms started reporting about their ethical, social and environmental conduct. And in marketing, being green and social is positioned as a relevant product and firm characteristic. In academic research, CSR has become a topic of interest too. Many studies investigate the connection between financial and social performance (see Lockett, Moon, and Visser, 2006). Numerous theoretical views on the link between financial and social performance are put forward (for an overview see Allouche and Laroche, 2006). Furthermore, a large number of empirical studies investigate the relationship between social and financial performance (see Orlitzky, Schmidt and Rynes 2003). Freeman and Liedtka (1991) presented seven reasons why the concept of social responsibility is often abandoned. One of the reasons given was, "Corporate social responsibility (CSR) promotes

incompetence by leading managers to involve themselves in areas beyond their expertise that is, repairing society's ill" (Freeman and Liedtka, 1991). Bauer et al. (2002) investigated the performance of international ethical mutual funds, corrected for investment style. The results show no significant difference in risk-adjusted returns between ethical and conventional funds for the period 1990-2001. Kneader et al. (2001) investigated the financial performance of 40 international ethical funds and 40 international non-ethical funds against their benchmark. The results show no statistical difference between their performances. They found that ethical funds have lower risk in comparison to their non-ethical counterparts. The cross-sectional analysis indicates that the risk-adjusted returns are not significantly related to the size, age or ethical status of the fund.

Hypothesis: CSR has a positive impact on organizational performance.

3. Research methodology

3.1 *Data collection*

To test our model, we collect data from hotel companies in Iran. The Iranian hotel industry provided a suitable context to test the hypotheses for the following reasons. To collect the data, a questionnaire together with a covering letter and a stamped return envelope was mailed to 200 hotels. With a cut-off date five weeks after the mailing, 100 completed questionnaires were received, resulting in a response rate of 50 per cent. The complete set of items for all the scales used in this study together with their sources is provided in the Appendix. All these scales (except for the performance scale which is measured by seven point scales) are measured using five point Likert type scales, ranging from "not at all" to "very much". The performance instrument is composed of three items asking the respondents to assess their companies' results against their major competitors in terms of sales growth, return on equity (ROE) and overall performance. Our operationalization of the CSR construct is based on the scale developed by Maignan et al. (1999). As the original scale is composed of 29 items, we used five dimensions for CSR. We obtained confirmatory factor analysis by SPSS software.

4. Analysis and results

In Table 1, we show the measurement properties of the variables and in Table 2 the correlations. All measures return a composite reliability in excess of the threshold value of 0.60 recommended by Bagozzi and Yi (1988). In sum, the results of the measurement analysis process indicate that the purified scales show adequate evidence of convergent validity. Construct reliability was evaluated using the procedure suggested by Fornell and Larcker (1981), including examining the parameter estimates and their

associated t values and assessing the average variance extracted for each construct (Anderson and Gerbing,

1988). Discriminant validity was assessed in a two-step process.

Variable/item	Mean	Standard deviation	Factor loading	Variance extracted (%)	Composite reliability
CSR					0.794
Item 1	3.255	0.765	0.854	0.698	
Item 2	3.564	0.875	0.912	0.656	
Item 3	3.214	0.998	0.759	0.719	
Item 4	3.659	1.021	0.851	0.515	
Item 5	3.587	0.855	0.912	0.645	
Performance					0.816
Overall	3.278	1.025	0.752	0.583	
ROE	3.651	1.135	0.698	0.714	
Sales	3.264	1.125	0.843	0.641	
H (CSR performance)	Loading (standardised)	Significant	Fit index		
	0.64	P<0.001	X ² =34.2	df=37	RMSEA=0.000 CFI=1.000 NFI=0.84

Notes: Item numbers refer to the item list in Appendix; all parameter estimates are significant at the p < 0.001 level; CSR = corporate social responsibility

Variables	Grow	ROE	Overall	CSR5	CSR4	CSR3	CSR2	CSR1
Grow	1.00							
ROE	0.18	1.00						
Overall	0.42	0.31	1.00					
CSR5	0.21	0.37	0.29	1.00				
CSR4	0.52	0.28	0.42	0.27	1.00			
CSR3	0.42	0.34	0.27	0.52	0.61	1.00		
CSR2	0.36	0.36	0.19	0.38	0.28	0.34	1.00	
CSR1	0.29	0.27	0.38	0.51	0.37	0.30	0.48	1.00

Notes: Grow = sales growth; ROE = return on equity; Overall = overall performance; CSR (1-5) = items 1-5 of corporate social responsibility scale. All correlations are significant at the p < 0.001 level except for the following. The correlation for CSR2- Overall is significant at the p < 0.05 level.

The hypothesis was tested by employing the structural equation modelling with a maximum likelihood estimation option. The results show that the effects of CSR (b = 0.57, p = 0.000) on performance is highly significant, hence the support H1. More specifically, the chi-square is 39.1 with 41 degrees of freedom and significant. The RMSEA-value is 0.000, and the NFI and CFI values are 0.84 and 1.000, respectively. The result shows that CSR could enhance performance.

5. Conclusion

The purpose of this paper is to investigate the effects of corporate social responsibility (CSR) on firm performance. The result shows that CSR could enhance performance. Hotel executives and managers may develop their overall CSR investments around community and product related issues, rather than employee relations, to maximize benefits of such

investments over both short- and long-terms. Managers in the hotel industry, in contrast, may focus their CSR initiatives on employee relations and product issues for the longterm, not the short-term. Consistent with Lee and Park (2009), the present results suggest that casino executives and managers may find a way to minimize their CSR investments in all dimensions because those investments do not appear to impact their firm's performance at all. Freeman and Liedtka (1991) presented seven reasons why the concept of social responsibility is often abandoned. One of the reasons given was, "Corporate social responsibility (CSR) promotes incompetence by leading managers to involve themselves in areas beyond their expertise that is, repairing society's ill" (Freeman and Liedtka, 1991). Bauer et al. (2002) investigated the performance of international ethical mutual funds, corrected for investment style. The

results show no significant difference in risk-adjusted returns between ethical and conventional funds for the period 1990-2001. This study has some limitations. The first limitation is that limitation is cross-sectional data were used in this study. Consequently, the time sequence of the relationships between CSR and organizational performance cannot be determined unambiguously. In addition, since this study only investigates Iranian hotels, hence, the findings and conclusions drawn from this research are representative of the Iranian hotels, and the findings may not generalize to other geographic regions or cultures. Future studies can also examine the proposed relationships in other countries. Future research might consider using objective performance measures such as stock returns to examine the framework we developed here.

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Appendix. Measurement scales

Corporate social responsibility (Maignan et al., 1999)

1. We continually improve the quality of our products and services
2. Our company seeks to comply with all laws regulating hiring and employees benefits
3. We are recognized as a trustworthy company
4. Our sales persons and employees are required to provide full and accurate information to all customers
5. We give active support to programs with good social causes

Organizational performance (Jaworski and Kohli, 1993)

How do you rate your company's performance in relation to your main competitors on a seven point scale

1. Our sales growth is ____
2. Our return on equity is ____
3. Our overall performance is ____

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Study of the behaviors of Prophets from the viewpoints of the Holy Koran and the Holy Bible

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Abstract: Making use of their different behaviors including honesty, firmness, eloquence, diction and their other behaviors as well as their different methods such as relinquishment, waiting, dispute, emotional and other methods, the prophets succeeded in leading the deeply ignorant and enslaved man towards perfection and humane peaks so that he/she know his/her responsibility under any circumstances. Under any circumstances, heavenly prophets fulfilled their religious responsibility, but they were never infatuated by terrestrial and natural beauties. They also underlined vindication and its presentation.

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Keywords: Honesty, Firmness, non-expectance, eloquence, relinquishment, expectation, dispute, emotional

Preamble

The philosophy behind the call to prophethood (prophetic mission) is to enlighten and lead people and to propagate religion, as mass evolution owes a lot to this issue and in case of the religious and social issue being removed, society will get involved in chaos and be dominated by barbarization and ferocity.

Given the common principles of heavenly religions, each and every prophet has his own specification for invitation and propagation and underlined a sensitive issue of promulgation regarding his era's special conditions and society. The holy Koran mentions some of them indeed. For instance, in the "She'ab's Invitation", economic invitation has been more emphasized as plunder and violence were common in financial affairs.

O my people! Give full measure and full weight in justice, and wrong not people in respect of their goods. And do not evil in the earth, causing corruption (Hud 85).

And in Saleh's call, campaign against secularism, aristocratism and welfare and conceit arising therefrom was underlined:

Build ye on every high place a monument for vain delight? And seek ye out strongholds, that haply ye may last for ever? And if ye seize by force, seize ye as tyrants? (Ash-Shuara 128-130)

And in Moses's invitation, political and social reforms and campaign against arrogance and unfair exploitation and power, wealth, deception, magic and other hubs were stressed.

Lo! Pharaoh exalted himself in the earth and made its people castes. A trive among them he suppressed, killing their sons and sparing their women. Lo! He was of those who work corruption (Al-Qasas 4)

In Joseph's invitation, divine management and in Solomon's invitation, policy and ruling, in David's invitation, destiny and in Jesus invitation, asceticism were proposed. Actually, Prophets' History, itself, depicts history of propagation and we are better off saying prophets themselves are founders of propagation and conductors of it.

Importance of propagation in verses of the Koran:

The Holy Koran attaches special importance to propagations, deems happiness, bliss and felicity of the Islamic society contingent upon endurance and execution of propagation (Al-Imran 11), deems compliance with this important responsibility one of the social necessities (Al-Asr 1-3) and considers this as a humanistic and belief advantage (At-Taubah 71). Now, we deal with some verses in this regard:

Mankind were one community, and Allah sent (unto them) Prophets as bearers of good tidings and as warners and revealed therewith the Scripture with truth that might judge between mankind concerning that wherein they differed. And only those unto whom (the Scripture) was given differed concerning it, after clear proofs had come unto them, through one of another. And Allah by His will guided those who believe unto the truth that concerning which they differed. Allah guideth whom He will unto a straight path (Al-Baqarah 213). And verily we have raised in every nation a messenger, (proclaiming): Serve Allah and shun false gods. Then some of them (there were) whom Allah guided, and some of them (there were) upon whom error had just hold. Do but travel in the land and see the nature of the consequences for the deniers! (An-Nahl 36) The importance of propagating, guiding, and teaching people is so great that the Almighty orders not to forget infidels in this regard. Whenever they are willing to hear the truths, you shall accept their request in complete safety and convey the divine truths to them. There are some verses in this regard as below: (Al-anfal 24, At-tobah 6).

Importance of Propagation in the Infallibles' Speeches

The value and importance of propagation have been emphasized in the Infallibles' speeches as well. Now, we present some examples in this regard: 1. According to Sermon 54 of Nahjolbalagheh, in Safein War, Imam Ali abstained from war in inasmuch as he was accused of fear of war!

“On the day of Kheibar War, Islamic troops were disappointed about conquest and victory, the former and the latter with defeat and escape from the Jewish scared Muslim fighters, Muhammad (Peace be upon him), the Prophet of Allah, said that, the next day, I will entrust the flag of Islam to a man who loves the Lord and His Prophet and the Lord and His Prophet love him too, he is the one who never escape war. And finally, he gave me the flag and before my departure, he said the God has chosen you as a unique man and wisheth you the best” Imam Ali said (Muslim Ibn Hajaj, 1957, Ibn Abil Hadid, 1959 a, Koleini, 1984, Majlesi, 1983 a, Mohammad, Hor-e-Ameli, 1989, Tabari, 1974 a). This saying conveys that propagation, guidance of people was of very great importance, and it even takes priority over Jihad in normal conditions.

2. After killing some of Imam Ali's troops, the warmongers of Jamal War were becoming ready to fight Imam, Imam Ali took the Holy Koran in his hand and said to his disciples that is there anyone among you to take the Holy Koran from me and frighten these misled and deceived ones from war? And if they cut his right hand, he takes the Holy Koran in his left hand? (Tabari 1974 b, Ibn Abi Hadid 1959 b, Majlesi 1983 b) A young man took delivery of the Holy Koran and fulfilled his duty until his martyrdom... From this historical event, we come to the conclusion that propagation and guidance of people are of such high importance that Imam Ali preached to his enemy before the war and for this purpose he let one of his loyal disciples to get killed.

Christianity and Propagation

On prophets, Isaiah, 8:9/6, says:

Also I heard the voice of the Lord, saying, Whom shall I send, and who will go for us? Then said I, Here am I; send me. And he said, Go, and tell this people, Hear ye indeed, but understand not; and see ye indeed, but perceive not.

Also EPHESIANS 4/11 says:

“And he gave some, apostles; and some, prophets; and some, evangelists; and some, pastors and teachers”

Christianity and the status of missioner:

All divine religions pay heed to propagation based on identity, guidance, advice, realism and missioner enjoys a very high position. Then he called his twelve disciples together, and gave them power and authority over all devils, and to cure diseases (Luke 1-2/9). And he sent them to preach the kingdom of God, and to heal the sick.

Also on the acceptance of missioner, says:

He that heareth you heareth me; and he that despiseth you despiseth me; and he that despiseth me despiseth him that sent me (Luke 16/10). There are some other examples in this regard (Luke 21/10, Jacob 20/5).

It can be observed that the Bible's logic on the missioner and messenger is the same as that of the Holy Koran:

The messenger believeth in that which hath been revealed unto him from his Lord and (so do) the believers. Each one believeth in Allah and His angels and His scriptures and His messengers. We make no distinction between any of His messengers and they say: We hear, and we obey. (Grant us) Thy forgiveness, our lord. Unto thee is the journeying (Al-Baqarah 285).

Personal Specifications of Prophets according to the Bible and the Koran:

Personal specifications of prophets are recounted as follows:

1. Prophets' honesty

Prophets' most effective propagation secret, that is, honesty and trueness, constitutes the survival secret of the Infallible Imams. For example, in the Holy Koran, while addressing Muhammad (Peace be upon him), the God says remember Moses who was a chosen and sincere Prophet:

And make mention in the Scripture of Moses. Lo! He was chosen and he was a messenger (of Allah), a Prophet (Maryam 51). For the sake of his obeying the God and for his servitude before the God, Moses attained a situation that he was expiated and cleansed by the God and verily if someone reaches such a position he/she will be immunized against pollution and obliquity. Whereas Satan, with all its perseverance to mislead the Lord's slaves, confesses that it is not capable of misleading and beguiling the sincere ones (Makarem Shirazi 1995 a). This is reflected in Verses 82 & 83 of Surah Sad:

He said: Then, by Thy might, I surely will beguile them every one,

Save Thy single-minded (sincere) slaves among them. There are many verses in this regard (Az-Zumar, 11, 14, Yusuf 24, Sad 45 & 46).

The same issue was observed in the Holy Bible:

And Moses made haste, and bowed his head toward the earth, and worshipped (Exodus 8/34).

So now it was not you that sent me hither, but God: and he hath made me a father to Pharaoh, and lord of all his house, and a ruler throughout all the land of Egypt (Genesis 8/45).

There are a plethora of examples in this regard, which are stated as below (Exodus 1/20, Genesis 16/41, 8/45, 3, 4/17, 17/17, 25/26, 15/35). Thus, as stated earlier and according to Koran and the Bible, we come to the conclusion that all prophets worship the God honestly and sincerely and their objective is to acquire the God's consent.

2. Prophets' firmness

In order to promulgate the religion, prophets bore a plethora of pains, hardships and troubles, and they were firm and constant and trusted in God to achieve this holy objective. The Holy Koran of

Muhammad gently called the believers to confide in the God against their tribe's opposition:

And lower thy wing (in Kindness) unto those believers who follow thee. And if they (thy kinsfolk) disobey thee, say: Lo! I am innocent of what they do. And put thy trust in the Mighty, the Merciful (Ash-SHuara 215-217).

And the Almighty says to the Prophet of Islam:

So tread thou the straight path as thou art commanded, and those who turn (unto Allah) with thee, and transgress not. Lo! He is Seer of What ye do (Hud 112).

The Heavenly Prophets' patience and fortitude is remarkably stated by Koran and the Holy Prophet of Islam is ordered thereby to be firm and patient like them.

Then have patience (O Muhammad) even as the stout of heart among the messengers (of old) had patience, and seek not to hasten on the doom of them. On the day when they see that which they are promised (it will seem to them) as though they had tarried but an hour of daylight. A clear message. Shall all be destroyed save evil living folk? (Al-Ahqaf 35)

Noah's patience and fortitude was mentioned in Koran. Noah, the great prophet of the God lived to be 950 according to Al-Ankabut 14, during these 950 years, Noah called his nation to theism and the worship of the God.

He said: My Lord! Lo! I have called unto my people night and day.

But all my calling doth but add to their repugnance;

And Lo! Whenever I call unto them that Thou mayest pardon them they thrust their fingers in their ears and cover themselves with their garments and persist (in their refusal) and magnify themselves in pride.

And Lo! I have called unto them aloud,

And Lo! I have made public proclamation unto them, and I have appealed to them in private (Nuh 5-9).

Noah made use of each and every method and stage of invitation and according to some interpreters, in order to pierce into the hearts of the stubborn and selfish people, Noah pursued his call through three methods as below:

1. Hidden call, which faced four reactions (thrusting their fingers into their ears, covering themselves with their garments and persisting in their refusal and blasphemy).
2. Solemn and public call
3. A combination of hidden and public calls

Noah incurred many losses in order to fulfill his mission, but his nation rioted and sinned and sought to annoy him and as they didn't accept his call, the Almighty stopped Sky bounty, that is, rain for 40 or 70 years and made their wives infertile so as to punish them (See Kashani 1957, Fakhreddin Rhazi 2000, Moghniyeh 1997, Balkhi 2003). Finally, all

problems and obstacles notwithstanding Noah fulfilled his mission and made his every endeavor. During his prophecy, a small group including 80 accepted his call and believed in him (Makarem Shirazi 1995 b).

There are other verses about this issue such as Al-Fath 29, Yunus 71).

The same has been regarded in the Holy Bible:

So I took them out of the land of Egypt and brought them into the wilderness.

"I gave them My statutes and informed them of My ordinances, by which, if a man observes them, he will live (Jacob 10,11/5). Noah is one of the people who believe in the God. When the God said to him that horror awaits unbelievers. He believed the God's saying. Despite the fact that there was no sign of storm, he built a ship and saved his ship.

By faith Noah, being warned of God of things not seen as yet, moved with fear, prepared an ark to the saving of his house; by the which he condemned the world, and became heir of the righteousness which is by faith (Hebrews 7/11).

There are many examples in this regard as below (Hebrews 24-27, 8-10/11, Romans 1-5/15, Jacob 10,11/5, 1 Peter 18-23/2). Therefore, as observed and according to the verses of the Holy Koran and Holy Bible, we come to the conclusion that, relying on their patience and resistance and confiding in the Almighty, the prophets tolerated all the hardships in their way.

3. Prophets had no expectations

Prophets expected nothing material from people and fulfilled their duties free of charge.

Muhammad (Peace be upon him) was one of the prophets that expected nothing material in return for the great responsibility of propagation of Islam as the holy Koran said:

Those are they whom Allah guideth, so follow their guidance. Say (O Muhammad, unto Mankind): I ask of you no fee for it. Lo! It is naught but a Reminder of (His) creatures (Al-An'am 90).

It should be noted that if the prophets were looking for material benefits in their propagation of religion and promulgation of prophecy is like a firm, people abhor religion (see Tabresi, 1993 a).

The other point is that, when prophets propagated the religion of Islam:

1. People become optimistic about such a call
2. Such a call will fructify sooner.
3. Prophets are immune from any libel (see Tabatabaei, 1997).

Therefore, all prophets could not continue their ways freely and away from any accusation and correct any deviation in people if they were materially in need and dependant. And People find out that the prophets have no mundane need and incur no worldly losses. The prophets have no material objective.

The same point is reflected in the Holy Bible as well. And he said, Lay not thine hand upon the lad, neither do thou anything unto him: for now I know that thou fearest God, seeing thou hast not withheld thy son, thine only son from me (Genesis 12/22). And Joseph said unto them, Fear not: for am I in the place of God?

But as for you, ye thought evil against me; but God meant it unto good, to bring to pass, as it is this day, to save much people alive (Ibid 19,20/50). There are some other examples in this regard (Genesis 14/43 and Exodus 17/2).

As stated earlier, according to the verses of the Holy Koran and the Holy Bible, we, therefore, come to the conclusion that the prophets expected nothing from people in return for propagating religion and they knew that their gratuities and bounties are with the Almighty.

4. Prophets' Eloquence

Eloquence and expression have direct relation with the prophets' propagation and progress in their objective. With their desirable expression, the prophets were able to propagate Islam and were successful indeed.

Moses is one of the prophets whose life, from the beginning until the end, includes examples, aphorism, promises, warnings and preaching. At the beginning of his prophecy, when he was sent by the Lord to propagate the Religion of God for the pharaohic, he felt that:

1. A prophet should be eloquent to fulfill the call to theism, to dispute, overcome the opponent and remove ambiguities. But Moses has a tongue tie or stammering and it was hard for him.
2. A prophet should have largeness, and therefore he asks the God for largeness so that his tolerance increases.
3. A prophet should be popular, but Moses had killed a man in order to help an Israelite man and therefore he stirred up people's hatred (see Gonabadi 1988).

(Moses) said: My Lord! Relieve my mind
And ease my task for me
And loose a knot from my tongue
That they may understand my saying
Appoint for me a henchman from my folk,
Aaron, my brother
Confirm my strength with him
And let him share my task (Ta-ha 25-32).

The same is reflected in the Holy Bible as below:

And Moses said unto the LORD, O my Lord, I am not eloquent, neither heretofore, nor since thou hast spoken unto thy servant: but I am slow of speech, and of a slow tongue.

And the LORD said unto him, Who hath made man's mouth? or who maketh the dumb, or deaf, or the seeing, or the blind? have not I the LORD?

Now therefore go, and I will be with thy mouth, and teach thee what thou shalt say.

And he said, O my Lord, send, I pray thee, by the hand of him whom thou wilt send.

And the anger of the LORD was kindled against Moses, and he said, Is not Aaron the Levite thy brother? I know that he can speak well. And also, behold, he cometh forth to meet thee: and when he seeth thee, he will be glad in his heart.

And thou shalt speak unto him, and put words in his mouth: and I will be with thy mouth, and with his mouth, and will teach you what ye shall do (Exodus 10-15/4).

As stated above and according to the verses of the Holy Koran and the Holy Bible, we come to the conclusion that the prophets' eloquence and expression are substantial condition for propagating the religion.

Prophets' propagation methods

The philosophy behind prophetic mission and heavenly envoys and the final objective of their books are to teach and guide people and to execute the propagation. In their call of people to Islam, the prophets made use of different methods in order to awaken man's asleep and dusty conscience and temperament and to remove the obstacles in the way of growth and reality-seeking and to lead people to realize truths and knowledge on reality. The heavenly prophets as the God's true missionaries made use of different propagation methods some of which are expressed here as below:

1. Relinquishment (Homami, 2006).

He openly turned away from me (Ragheb Esfahani, 2011).

There are many verses in this regard some of which are expressed as below:

O Joseph! Turn away from this, and thou, (O woman), ask forgiveness for thy sin. Lo! Thou art of the sinful (Yusuf 29).

(It was said) O Abraham! Forsake this! Lo! Thy Lord's commandment hath gone forth, and Lo! There cometh unto them a doom which cannot be repelled (Hud 76)

Those are they, the secrets of whose hearts Allah Knoweth. So oppose them and admonish them, and address them in plain terms about their souls (An-Nisa 63).

Then, withdraw (O Muhammad) from him who fleeth from Our remembrance and desireth but the life of the world (An-Najm 29).

Follow that which is inspired in thee from thy Lord; there is no God save Him; and turn away from idolaters (Al-An'am 106).

Turning away from heathens is three types:

- a) Turning away from heathen's belief, which is considered to be polytheism:

And verily it hath been revealed unto thee as unto those before thee (saying): If thou ascribe a partner

to Allah thy work will fail and thou indeed wilt be among the losers (Az-Zumar 65).

b) To relinquish the call of the heathens as they are not capable of being guided and do not comply with your call.

So let them chat and play until they meet their Day which they are promised (Al-Ma'arej 42).

So let them flounder (in their talk) and play until they meet the Day which they are promised (Az-Zukhruf 83).

c) Relinquishment from communication and calmness towards them, as there is no hope with them accompanying you and those, who say relinquishment and turning away from them betoken fighting with them and that this nullifies At-Taubah Verse 36, are wrong (Teyeb, 1991 a).

Lo! The number of months with Allah is twelve months by Allah's ordinance in the day that He created the heavens and the earth. Four of them are sacred: that is the right religion. So wrong not yourself in them. And wage war on all the idolaters as they are waging war on all of you. And know that Allah is with those who keep their duty (unto Him) (At-Taubah 36).

Al-an'am 107 is used here and turning away as stipulated in Verse 106 implies the second meaning that is relinquishment from call.

The same has been indicated in the Holy Bible:

Perverse disputings of men of corrupt minds, and destitute of the truth, supposing that gain is godliness: from such withdraw thyself (1 Timothy 5/6).

Therefore, as stated earlier and considering the verses of the Holy Koran and the Holy Bible, we come to the conclusion that the prophets made use of relinquishment and turning away suitably and as appropriate.

2. Expectancy (Homami, 2006)

Here, expectancy means to delay something (Ragheb Esfahani, 2011)

There are many verses in this regard such as:

(The messengers) said: O Lot! Lo! We are messengers of thy Lord; they shall not reach thee. So travel with thy people in a part of the night, and let not one of you turn round (all) save thy wife. Lo! That which smiteth will smite her (also). Lo! Their tryst is (for) the morning. Is not the morning night? (Hud 81)

He said: Terror and wrath from your Lord have already fallen on you. Would ye wrangle with me over names, which ye have named, ye and your fathers, for which no warrant from Allah hath been revealed? Then await (the consequence), Lo! I (also) am of those awaiting (it) (Al-Araf 71).

Wait they, indeed, for nothing less than that the angels should come unto them, or thy Lord should come, or these should come one of the portents from thy Lord (Al-An'am 158).

<<Expectance and expectancy means to see by eyes or by inner eyes and spiritual (unworldly) heart.

Ultimately, an issue is interpreted as expectancy if it bears actuality. Like what the Almighty says:

Look, therefore, at the prints of Allah's mercy (in creation); how He quickeneth the earth after her death. Lo! He verily is the Quickener of the Dead, and He is Able to do all things (Ar-rum 50).

And an issue is interpreted as expectation if it is dated. For instance, the Almighty's promise worked and befell soon and the infidels and idolaters were subjugated and the Muslims defeated them and they were annihilated.

And those who disbelieved said unto their messengers: Verily we will drive you out from our land unless ye return to our religion. Then their Lord inspired them, (saying): Verily we shall destroy the wrongdoers,

And verily we shall make you to dwell in the land after them. This is for him who feareth My Majesty and feareth My threats (Ebrahim 13, 14). >> (Tayeb 1991 b)

There are many other verses in this regard (Hud 39, 122, Al-Qasas 35).

The same has been observed in the Holy Bible:

Behold, tomorrow about this time I will cause it to rain a very grievous hail, such as hath not been in Egypt since the foundation thereof even until now (Exodus 18/9).

Else, if thou refuse to let my people go, behold, tomorrow will I bring the locusts into thy coast:

And they shall cover the face of the earth, that one cannot be able to see the earth: and they shall eat the residue of that which is escaped, which remaineth unto you from the hail, and shall eat every tree which groweth for you out of the field: (Ibid 4,5/10)

There are many other instances in the Holy Bible in this regard (Mathew 22,23/27, Job 18,1/24, EZEKIEL 27,28.12 and Revelation 12/22).

Therefore, as stated earlier and given the verses of the Holy Koran and the Holy Bible, we come to the conclusion that the prophets made use of the propagation method of expectation to call people of their times and carried out their call and warnings and fulfilled their duties thereby.

3. Dispute (Homami, 2006)

According to Mofradat-e-Raqeb, dispute is utterance in the form of debate to defeat other party; in other words, it means to rush in order to overcome enemy (Ragheb Esfahani, 2011).

Dispute means:

1. To defeat adverse power and to lead it to the right way through reasoning (Tabresi, 1993 b)
2. To bring reason against reason
3. Tenacious dissent that is to say to throw a rope or twist it.
4. A fierce man, it also means to cut an opponent down (Ibid 1993)

There are many verses in this regard, some of which are as below:

Bethink thee of him who has an argument with Abraham about his Lord, because Allah had given him the Kingdom; how, when Abraham said: My Lord is He who giveth life and causeth death, he answered: I give life and cause death. Abraham said: Lo! Allah causeth the sun to rise in the East, so do thou cause it to come up from the west. Thus was the disbeliever abashed. And Allah guideth not wrong doing folk (Al-Baqarah 258).

He said: O my people! Bethink you, if I rely on a clear proof from my lord and there hath come unto me a mercy from His presence, and it hath been made obscure to you, can we compel you to accept it when ye are averse thereto?

And O my people! I ask of you no wealth therefore. My reward is the concern only of Allah, and I am not going to thrust away those who believe Lo! They have to meet their Lord but I see you a folk that are ignorant.

And, O my people! Who would deliver me from Allah if I thrust them away? Will ye not then reflect?

I say not unto you: "I have the treasures of Allah" nor "I have knowledge of the Unseen", nor say I: "Lo! I am an angel! Nor say I unto those whom your eyes scorn that Allah will not give them good Allah Knoweth best what is their hearts Lo! Then indeed I should be of the wrong doers.

They said: O Noah! Thou hast disputed with us and multiplied disputation with us; now bring upon Us that where with thou threatenest us, if thou art of the truthful.

He said: Only Allah will bring it upon you if He will, and ye can by no means escape

My counsel will not profit you if I were minded to advise you, if Allah's will is to keep you astray. He is your lord and unto Him ye will be brought back.

Or say they (again) He hath invented it? If I have inverted it, upon me be my crimes, but I am innocent of (all) that ye commit (Hud 28-35).

Say: Is there of your partners (whom ye ascribe unto Allah) one that produceth it? Say: Allah produceth creation, then reproduceth it. How then, are ye misled!

Say: Is there of your partners (whom ye ascribe unto Allah) one that leadeth to the Truth? Say: Allah leadeth to the Truth. Is he who leadeth to the Truth more observing that He should be followed, or he who findeth not the way unless he (himself) be guided. What aileth you? How judge ye? (Yunus 34, 35)

There are some other verses in this regard (Al-An'am 76-81, Ash-Shu'ara 15-31, Al-Imran 61, Yunus 38).

The same has been observed in the Holy Bible:

Behold now, I have ordered my cause; I know that I shall be justified.

Who is he that will plead with me? for now, if I hold my tongue, I shall give up the ghost (Job 18, 19/13).

There are some other samples in this regard (Exodus 7/17, Isaiah 18/1, 1 kings 16-22/3).

4. Emotional Method (Homami, 2006)

Emotional method is another method used by the prophets for their call.

There are many verses in this regard:

O my father! Lo! I fear lest a punishment from the Beneficent overtake thee so that thou become a comrade of the devil (Maryam 45).

Go, both of you, unto Pharaoh. Lo! He hath transgressed (the bounds).

And speak unto him a gentle word, that peradventure he may heed or fear (Ta-Ha 43, 44).

And know that the messenger of Allah is among you. If he were to obey you in much of the government, ye would surely be in trouble; but Allah hath endeared the faith to you and hath beautified it in your hearts, and hath made disbelief, lewdness, and rebellion hateful unto you. Such are they who are rightly guided (Al-Hujurat 7).

There are some other verses in this regard. (Ash-shuara 107, Al-Araf 62, An-Nazi'at 17, 18, Maryam 42).

The same has been observed in the Holy Bible:

And Moses said, Behold, I go out from thee, and I will intreat the LORD that the swarms of flies may depart from Pharaoh, from his servants, and from his people, tomorrow: but let not Pharaoh deal deceitfully any more in not letting the people go to sacrifice to the LORD (Exodus 29/8).

Now therefore fear ye not: I will nourish you, and your little ones. And he comforted them, and spake kindly unto them (Genesis 21/50).

There are some other examples in this regard (JEREMIAH 36/48, Exodus 11, 13/32).

Considering the above and given the verses of the Holy Koran and the Holy Bible, we come to the conclusion that the holy prophets have made use of emotions, kindness and sensation in their propagation methods.

Conclusion

The heavenly prophets were sent all over the world throughout history. They fulfilled their conspicuous roles in the guidance and education of people. Each prophet completed his predecessor and proof of him. According to Al-Hadid 25:

"We verily sent Our messengers with clear proofs, and revealed with them the Scripture and the Balance, that mankind may observe right measure; and He revealed iron, wherein is mighty power and (many) uses for mankind, and that Allah may know him who helpeth Him and His messengers, though unseen. Lo! Allah is Strong, Almighty".

It can be said that all prophets looked for a unique goal and each prophet's teaching and propagation method was commensurate with the time and place, thought and culture of the people. From among the divine prophets, Noah, Abraham, Moses and Jesus put at people's disposal some

books including individual, social orders and regulations and legal and ethical obligations commensurate with the conditions of time and environment, but parts of these books were lost or diverted literally and immaterially with the passing of time. For instance, the story of Death of Moses (Peace be upon him)(Deuteronomy 4-8/34), vilification of them, such as accusing David (Peace be upon him) of adultery with a married woman (2 Samuel 2-5/11), accusing the Prophet Lut of drinking alcohol and adultery with a married woman (Genesis 30-38/19) and the like. Meanwhile, according to Christians, the present Bible is not the book the Almighty revealed to Jesus as it contains reports attributed to some disciples of his. In addition to recommendation of drinking alcohol, drinking is considered as a miracle of the Jesus (John 2).

Yet, what was said above does not mean that the whole Bible is diverted. And as expressed earlier, the Holy Koran conforms to the Holy Bible in many respects. We therefore come to the conclusion that the Heavenly Prophets have been sent to guide man; and according to the Holy Koran, they complement each other in behaviors and propagation methods. And according to the Holy Bible and the Holy Koran, the prophets rose from deprived societies and poor families and after passing through meandrous ways and facing very sad events, they were able to create movements in their societies and created deep effects on man's soul and the human civilization and the environment around them to the extent that sometimes they created dazzling and brilliant breakthroughs for men. The lives of the Jesus and Muhammad (Peace be upon him) are two examples and men bow in respect before the revolutions created according to these two prophets' teachings. I hope that, one day in the future, man will make use of the prophets' achievements and deliver him from ignorance, aberrance, selfishness etc.

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The Role of Glucose–Insulin–Potassium on ST Resolution in Acute Myocardial Infarction; A randomized clinical trial

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Abstract:Background: The effects of glucose–insulin–potassium in the treatment of ST segment elevation myocardial infarction are controversial. We aimed to evaluate the effect of this solution on ST segment resolution which is an early noninvasive marker of coronary reperfusion. Methods: From September 2008 to July 2009, this randomized clinical trial enrolled 74 patients who had myocardial infarction that was treated with 25% glucose, 50 unit of soluble insulin per liter, and 80 milliequivalents of potassium chloride per liter at 1 ml/kg/hour (case group) or normal saline (control group) as adjunct to thrombolytic therapy in Sabzevar, Iran. ST-segment resolution was defined as complete (>70%), partial (30% to 70%), or none (<30%) and absent (<50%) or present (≥50%). Results: No difference was present between groups in ST-segment resolution (p=0.8). There was no difference in the rate of complete ST-segment resolution between groups at 120 min (P=NS). Left ventricular ejection fraction was significantly higher in patients who had complete ST-segment elevation resolution (44.6± 7.4%) than others (40.9± 9.2 % and 35.0± 9.2%). Conclusion: ST-segment resolution was similar in groups. A more degree of ST-segment elevation resolution was correlated with better ejection fraction.

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1. Introduction

The ST-segment resolution on electrocardiogram (ECG) has been shown to be a simple, indicator of reperfusion (1) (2) (3) (4), which is the most effective treatment in acute myocardial infarction(5). However, reperfusion itself may cause reperfusion myocardial injury and affect infarct size. Therefore, next step is limiting myocardial infarct size in order to improve clinical outcomes in patients with myocardial infarction(6). One of these strategies is treatment with glucose–insulin–potassium (GIK) which can shift myocardial free fatty acid metabolism towards glucose metabolism(7), but the effects of glucose–insulin–potassium in the treatment of ST segment elevation myocardial infarction (STEMI) are controversial. Fath-Ordoubadi et al published a meta-analysis of GIK treatment and showed that GIK infusion could reduce in-hospital mortality (8), while in some studies, no benefit effects of GIK were observed(9) (10) (11).

However, studies about GIK effects are ongoing; the effects on ST-segment resolution have been used in a few studies. Hence we investigated the effect of administration of GIK on single-lead ST-segment analysis before and 120 minutes after

thrombolytic therapy. In addition we used a different thrombolytic in this study (streptokinase) which is not investigated in previous studies.

2. Materials and Methods

This is a part of a study which evaluated the effects of GIK on STEMI. As soon as the university ethic committee approved the research, recruitment began. From September 2008 to July 2009, 74 consecutive STEMI patients were randomized to normal saline (N = 39) or GIK infusion (N = 35). Two ECGs in the GIK group were not valid to include in the study. All consecutive patients who had ST elevation myocardial infarction according to AHA guideline, when admitted to the Coronary Care Unit of the Vaseie general hospital (Sabzevar, Iran) were recruited (12) (13). Details of study design and population are described elsewhere(14)

Patient with hypotension, congestive heart failure, creatinine >2.0 mg/dL and anemia (hemoglobin <11 g/dL), were excluded. Further exclusion criteria were unwillingness to participate. Informed consent was obtained from the participants or their legal guardians.

The study was randomized, prospective, and triple blinded. On admission, patients were randomized via block randomization and assigned to the respective group by a closed envelope system. In A group, patients received high-dose GIK infusion (25% glucose, 50 IU of soluble insulin per liter, and 80 mmol of potassium chloride per liter at 1 ml/kg/hour) as an adjunct to thrombolytic therapy (1.5 MU of streptokinase/30 to 60 minutes) [GIK group]. In B group 1 L normal saline at 60 mL/h infused in addition to thrombolytic therapy (1.5 MU of streptokinase/30 to 60 minutes) [control group]. Neither the patients nor the researcher and investigator assessing the ECGs, nor the statistical consultant knew which group called A and which one called B group (triple blinded). GIK administered in the hospital that initiated the reperfusion therapy. All other medication was standard as ACC/AHA guidelines. (13)

In each patient, total CK and CK-MB level were measured on admission, 16 and 24 hours thereafter. In all patients, plasma concentrations of glucose and potassium were determined before and at 6 hour after administering therapy. Two-dimensional echocardiography was obtained in all patients after 72 hours of admission.

A standard 12-lead electrocardiogram with a speed of 25 mm/s and amplitude of 10 mm/mV were obtained before and within 120 minutes of starting treatment. ST-segment elevation was investigated by a single observer who was blinded to study groups with lens-intensified calipers. The single lead with the most prominent ST-segment elevation before treatment was identified, and ST-segment elevation was measured at 20 ms after the J point with the PR segment as reference baseline. The percent resolution of ST-segment deviation from baseline to 120 was calculated and categorized: complete (>70%) STRs, partial (30% to 70%) STRs, and no (<30%) STRs [5]. Another classification was performed as present ST-segment resolution (STRs \geq 50%) or absent ST-segment resolution (STRs <50%). A similar approach was used in recent studies (15) (16) (17). Patients were followed clinically for 7 days.

Data were analyzed through the Statistical Package for the Social Sciences (SPSS, version 15). Continuous variables were expressed as mean \pm SD and were compared using independent t tests and repeated measure ANOVA. Categorical data were analyzed by use of chi-square or Fisher exact test. The relation between ST segment resolution and left ventricular ejection fraction was made with 2-way analysis of variance. Assuming a power of 80% and $\alpha=5\%$, comparing ST segment resolution could be demonstrated with a sample size of 32 patients in each group. A p value <0.05 was considered to be statistically significant.

4. Results

74 Patients were enrolled into the study. There was no significant difference in basal data between two study groups. The time from chest pain onset to initiation of streptokinase was 4.66 ± 3.81 hours. There was no significant difference in this interval between two study groups ($P=0.16$). Glucose level at Baseline was 149 ± 61 mg/dL in the control and 168 ± 83 mg/dL in the GIK group ($P=0.25$). Mean glucose level over 6 hours was higher in the GIK group (212 ± 139 mg/dL) than control group (142 ± 46 mg/dL) ($P=0.006$).

No difference was present between two groups in ST-segment elevation resolution ($p=0.8$). Ejection Fraction (39% G vs 41% C, $p=0.34$) was similar between the two groups.

In GIK and control group, complete resolution was registered in 8 patients (25%) vs 18 patients (46.2%), partial in 12 (37.5%) vs 14 (35.9%), and absent in 12 (37.5%) vs 7 (17.9%), respectively ($P=NS$). A resolution $\geq 50\%$ was observed in 56.3% of patients in GIK group, and in 71.8% of control group ($P=0.17$).

Left ventricular EF was significantly higher in patients who had complete ST-segment elevation resolution ($44.6 \pm 7.4\%$) than in those who had partial resolution ($40.9 \pm 9.2\%$), and in patients who had absent resolution ($35.0 \pm 9.2\%$), ($p=0.003$).

There was no significant difference between groups regarding mean admission serum CK, peak CK, peak CK-MB levels and early peak of CK within 16 hour.

3. Discussion:

According to our data GIK therapy in patients who receive streptokinase, offers no effect on ST-segment resolution, which is a sign of successful recanalization (18).

The simplest approach to analyze ST-segment resolution is to classify ST-segment elevation in different groups and to categorize the patient in these groups. One of these classifications is ST-segment resolution with the 50% cutoff as present or absent (15) (16) (19). We also used the other common classification with 70% and 30% thresholds, as complete, partial, or absent ST-segment elevation resolution, (20). We found GIK ineffective according to both classification.

Previous studies have investigated the effect of GIK on ST-segment elevation resolution in animals. GIK has decreased ST-segment elevation in dogs with induced myocardial ischemia (21). Result of differences between animal studies versus our study can be due to thinner myocardium in animals which is more resistant to ischemia after myocardial infarction than in humans with thick myocardium. One study showed that GIK can decrease in free fatty acid

concentration and promotion of glycolysis. Therefore an ischemic cell needs less oxygen and produces less toxic intermediates of free fatty acid metabolism (7). However, while reperfusion therapy with streptokinase can rescue the ischemic myocardium effectively, treatment with GIK has less value. In a post hoc analysis, GIK results in a higher number of patients with partial or complete ST-segment elevation resolution but they suggested more prospective trial (22). In 32 ST-elevation MI patients treated with reteplase and alternately assigned to either GIK or saline with potassium chloride, GIK failed to decrease ST-segment elevation (23), which is corresponding to our study with enough sampling. Therefore at least at the present time, there is no role for GIK as adjunctive treatment with thrombolytic drugs.

In our study, ST-segment elevation resolution was related to a favorable outcome in EF according to both classification of ST segment resolution, which is corresponding to previous studies (15) (24) (25). We suggest the use of electrocardiogram to estimate left ventricular function. Or, in minimum ST-segment evaluation can be used to categorize outcome as acceptable or unacceptable.

We should mention some limitations of this study. We used easy-to-use method to analyze ST-segment resolution and using all ECG leads could have produced different results. However, this method has been used and validated in previous studies. (16) (26) (27) (28) And, recent studies failed to demonstrate advantages of more complex measurements. (26) (27). However, one of the advantages of this method is early decision making because we can evaluate it shortly after treatment. As far as there is no data about functional status before infarction, we could not recognize functional recovery, and we cannot exclude some degree of functional impairment in a patient with normal left ventricular EF.

In conclusion, in patients with STEMI, according to ECG finding, there is no role for GIK as adjunctive treatment with streptokinase, but a more degree of ST-segment elevation resolution remains a valuable predictor of effective reperfusion and left ventricular EF.

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Conflict of interest: None declared

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Evaluation of 8-hydroxyquinoline physiological effect and Genotoxicity on *Paramisgurnus dabryanus* using hepatase activity and comet assay

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Abstract: With economic development and industrialization, vast genotoxic chemicals were produced and distributed in the environment. As an important industrial raw material, 8-hydroxyquinoline (8-HOQ) has been used in a wide variety of industrial circle. Therefore, human beings and other organisms, especially aquatic organisms might be exposed to these drugs and would have health risks. This study was a preliminary step to evaluate the toxicity effect of 8-hydroxyquinoline (8-HOQ) on *Paramisgurnus dabryanus* through using the methods of acute toxicity test, hepatase activity and comet assay. The results indicated that 8-HOQ had obvious toxicity effect on *Paramisgurnus dabryanus*. With the increase of the treatment-concentration and -time of 8-HOQ, the hepatic GPT and GOT activity of *Paramisgurnus dabryanus* were decreased obviously. Meanwhile, three comet parameters of hepatocyte were increased significantly, and there was significant difference between control group and each treatment group ($p < 0.05$). These results suggest that 8-HOQ may become toxic chemical contaminant in environment and threaten aquatic and other organism health.

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Keywords *Paramisgurnus dabryanus*, 8-hydroxyquinoline, GOT, GPT, comet assay

1. Introduction

With economic development and industrialization, vast genotoxic chemicals were produced and distributed in the environment. 8-Hydroxyquinoline is a white to off-white crystal or crystalline powder that is insoluble in water or ether and freely soluble in ethanol, acetone, chloroform, benzene, and aqueous mineral acids (Elena et al., 2009). As an important industrial raw material, 8-hydroxyquinoline (8-HOQ) has been used in a wide variety of fields such as preservative, antimicrobial, healing drugs (medicine), agricultural pesticides and dyestuffs (Ling et al., 2009). Compared with aliphatic series and aromatic series compounds, 8-HOQ which is a nitrogen heterocyclic compound has a very much lower performance of biodegradation (Gros et al., 2009). Therefore, 8-HOQ and its derivatives have huge potential of bioaccumulation and persistence in the environment and would lead a major worldwide (water) environmental contamination. In addition, some previous studies indicate that the quinoline of 8-HOQ parent compound and its derivatives have obvious toxicity effect on organisms (Shen et al., 1999; Volkova et al., 2007; Gary et al., 2008; Hsu et al., 2008; Carolina et al., 2010; Siddharth et al., 2011). However, there is limited information available on organisms exposure to 8-hydroxyquinoline.

Therefore, the toxicity study about 8-HOQ is needed.

The long-term accumulation of toxic substances in animal body would cause grievous injury in hepatic tissue, and then would cause animal hepatase activity changes. Among all hepatase, glutamic-pyruvic transaminase (GPT) and glutamic-oxalacetic transaminase (GOT) are important aminotransferase broadly existing in animal mitochondria. Under normal circumstances, the aminotransferases have higher activity in liver cells. However, when organism is in toxication, the activity of liver aminotransferases would decrease and the blood serum aminotransferases activation would increase (Saravanan et al., 2012). Therefore, the enzymological parameters of liver and blood serum in tested fish were assessed by GOT and GPT activity assays in this study. In addition, the comet assay is a simple visual technique to measure DNA damage in cells (Singh et al., 1988). Comet assay is a quick, simple, reliable, and sensitive technique to detect and measure genetic damage in almost any type of eukaryotic cell, by using a small number of cells (Eşref et al., 2010). Therefore, this technique has been widely used for various *in vitro* and *in vivo* studies to monitor the effects of DNA-damaging agents in several fishes and other organisms (Capriglione et al., 2011; Piperakis et al., 2006; Cheryl et al., 2012). Then, in this study, the genetic

toxicity effect of 8-HOQ on *Paramisgurnus dabryanus* was also assessed by comet assay.

Paramisgurnus dabryanus, which belongs to *Paramisgurnus*, *Cypriniformes*, and *Cobitidae*, is widely distributed freshwater fish and can be found in Chinese water bodies. Furthermore, with the advantages of short life cycles, easy to catch and quick responses to environmental disturbances in an integrated and continuous manner, *P. dabryanus* is used in toxicity tests of chemicals usually. Earlier studies also had confirmed that *P. dabryanus* was the ideal test organism for assessing the toxicological effects of pollutants (Zhang et al., 2008; Seok-Ki et al., 2010). However, to our knowledge, the information of 8-HOQ-induced biotoxicity in *P. dabryanus* is limited. Thus convenient and valid biomonitoring methodology is essential for evaluating possible health risks for fish and other organism health due to 8-HOQ contaminant. The aim of this study is to evaluate the water environmental risk of 8-HOQ contamination for fish populations, and propose the comet assay applied to fish as a biomonitoring methodology to evaluate 8-HOQ genotoxicity.

2. Materials and methods

2.1. Tested fish and major chemicals

Adult fish (*P. dabryanus*) (16.4 ± 3.2 g) were collected from wetlands in the old course of Yellow River, Yanjin (Henan, China), and were bred in the exposure of tap water under laboratory conditions. The 8-hydroxylquinoline was purchased from the Nanjing Jiancheng Bioengineering Institute (China), and its purity was more than 99%. All other chemicals used were of analytical grade.

2.2. Acute toxicity on *P. dabryanus*

Acute toxicity test was carried out by using the Spearman-Kärber method with some modification to obtain a 50% lethal concentration (LC_{50}) of 8-HOQ in *P. dabryanus* after 24, 48 and 96 h of exposure (Kärber, 1931; Li et al., 2012). Sixty adult fish were randomly divided into 6 groups (10 fish in each group), out of which 5 groups served as HOQ-treatment group and another as a control. The treatment groups were exposed to the 8-HOQ solution at the concentration of 14.37, 21.48, 32.23, 48.34 and 72.58 $mg\ l^{-1}$ for 96 h, respectively. And the control group was placed in aerated tap water for the same period. Each test-group was all placed in plastic aquaria (6 L in volume), exchanged with its same concentration 8-HOQ solution after exposure of 24 h and no food was provided during the treatment-time. Each test was conducted in duplicate. The dead and surviving fish were recorded in each group during the exposure period. The LC_{50} of 8-HOQ on *P. dabryanus* after 24 h, 48 h and 96 h of exposure were calculated using SPSS version 13 with the amended Spearman-Kärber method.

2.3. Hepatic transaminase activity assays

By the result of acute toxicity, ninety adult *P.*

dabryanus were randomly divided into five groups (3 parallels in each group, 6 fish in each parallel), out of which four groups served as 8-HOQ-treatment group and another as a control. The treatment groups were exposed to the 8-HOQ solution at the concentration of 3.629, 7.258, 10.887 and 14.516 $mg\ l^{-1}$ for 6 days. Its treatment process was the same as the acute toxicity test. After 2, 4, and 6 d of exposure, three fish of each group (1 fish in each parallel) were all anaesthetized with 100 $mg\ l^{-1}$ MS-222 (Tricaine) and then dissected. Samples (blood serum and liver) were collected for the hepatic amino acid transaminase (GPT/GOT) activity assays. GPT/ GOT were determined using the Diagnostic Reagent Kits, purchased from the Nanjing Jiancheng Bioengineering Institute (China), according to the manufacturer's instructions. The results of these enzymatic assays were given in units of enzymatic activity per milligram of protein ($U/mg\ prot$), whereas 1 U of GOT/GPT was defined as a change of 0.001 in the absorbance of NADH/min (Yin et al., 2011).

2.4. Comet assay

The experimental design of comet assay was similar to the enzymes activity assay test. Three fish of each group (1 fish in each parallel) were anaesthetized with 100 $mg\ l^{-1}$ MS-222 (Tricaine) and followed by the severing of the spinal cord of tail stern after 6, 24, 48 and 96 h of exposure, respectively. Then, the livers were dissected from the tested fish quickly, placed in PBS (pH 7.5), kept on ice and gently cut up with scissors. After isolated hepatocytes were released as a suspension, the cell number and cell viability were measured with the performance as described by Mitchelmore et al. (Mitchelmore et al., 1998), with some modifications. Cells were typically > 80% viable with $1 \times 10^6 \sim 1 \times 10^8$ cells isolated per liver.

The single cell gel electrophoresis was performed as described by Singh et al. and Piperakis et al. (Singh et al., 1988; Piperakis et al., 2006) with minor modification. After cell lysis (2.5M NaCl, 10 mM Tris, 100 mM Na_2EDTA , 1% Triton X-100, 10% DMSO, 1% Na sarcosinate, pH 10.0, ≥ 1 h), DNA was placed in an alkaline electrophoresis buffer (300 mM NaOH, 1 mM Na_2EDTA ; pH13, 4°C) for 10 min to allow the DNA to unwind, followed by electrophoresis in the same buffer for 20 min at 25 V, 300 mA at 4°C. Samples were stained with ethidium bromide (10 $\mu g/ml$, 10 min) and examined under fluorescent microscope (OLYMPUS BX60) or placed in a humid, dark box at 4°C until analysis (within 48 h). All slides were coded and the whole slide was scanned randomly. Two slides per specimen were prepared and fifty random cells per slide were analyzed and scored by using an image analysis package. Cells with damaged DNA appeared as comets (Fig 3), whose tail length, tail DNA% and tail moment ($TM = \text{tail DNA}\% \times \text{tail length}$) were assessed by using the CytoVision NT automatic image

analysis system. Then tested cells were categorised for four grade of damage (using Tail Moment) based on the criteria reported by Anderson et al. (Anderson et al., 1994) with minor modifications (grade of damage: zero or minimal 1-5, low damage 5–20, mid damage 20-60, and high damage > 60).

2.5. Statistical analyses

All data were expressed as mean \pm SD and analyzed by one-way analysis of variance. And the statistical significance was evaluated by SPSS version 17.0 (P values less than 0.05 were considered statistically significant) and the individual comparisons were obtained by Duncans' multiple range test.

3. Results

3.1. Acute toxicity of 8-HOQ on *Paramisgurnus dabryanus*

The acute toxicity data of 8-HOQ on adult *P. dabryanus* was described in table1. As was shown by the table1, the mortality (%) were increased with the increase of treatment-concentration and treatment-time of 8-HOQ. Additionally, the LC50 of 8-HOQ to *P. dabryanus* was decreased with the increase of treatment-time of 8-HOQ. The 24, 48 and 96 h LC₅₀ of 8-HOQ on adult loach were 69.70 \pm 3.29,

48.35 \pm 4.23 and 28.53 \pm 2.84 mg l⁻¹, which indicated that 8-HOQ had obvious toxicity to *P. dabryanus*.

Table 1. Mortality rate of adult *P. dabryanus* induced by 8-HOQ

8-HOQ (mg l ⁻¹)	Mortality (%)		
	24 h	48 h	96 h
14.37	0 \pm 0	0 \pm 0	10 \pm 0
21.48	0 \pm 0	10 \pm 0	20 \pm 0
32.23	10 \pm 0	25 \pm 5	35 \pm 5
48.34	25 \pm 5	45 \pm 5	85 \pm 5
72.58	45 \pm 5	70 \pm 0	100 \pm 0

3.2. GOT and GPT activity assay in hepatocyte and serum of adult fish

The Data of enzyme activity assays of 8-HOQ to adult *P. dabryanus* were described in Table 2. Subacute exposure to 8-HOQ decreased the liver GOT and GPT activity in tested fish, while the blood serum GOT and GPT activity increased compared with their control (Table 2). And there was significant difference (p<0.05) between each treatment group and its control group, except that the hepatic GPT and GOT activity of treatment-group 3.629 mg L⁻¹ of 2 d exposure (p > 0.05).

Table 2. GOT and GPT activity of hepatocyte and serum induced by 8-HOQ in *P. dabryanus*

8-HOQ (μg l ⁻¹)		2d		4d		6d	
		GPT (U mg·prot ⁻¹)	GOT (U mg·prot ⁻¹)	GPT (U mg·prot ⁻¹)	GOT (U mg·prot ⁻¹)	GPT (U mg·prot ⁻¹)	GOT (U mg·prot ⁻¹)
0	H	233.00 \pm 24.98	196.47 \pm 13.29	228.00 \pm 13.95	188.26 \pm 15.55	218.22 \pm 17.56	188.02 \pm 15.77
	S	19.79 \pm 3.37	18.46 \pm 2.24	20.19 \pm 3.34	19.24 \pm 3.06	21.26 \pm 3.44	20.97 \pm 2.09
3.629	H	210.76 \pm 10.59	170.61 \pm 11.36	175.01 \pm 10.82*	117.13 \pm 6.17*	172.24 \pm 13.75*	115.37 \pm 12.10*
	S	31.23 \pm 3.27*	29.91 \pm 2.59*	43.50 \pm 3.75*	35.79 \pm 4.81*	48.89 \pm 5.28**	47.46 \pm 4.69**
7.258	H	193.51 \pm 14.47*	144.05 \pm 12.93*	161.34 \pm 11.43*	101.76 \pm 6.22*	144.03 \pm 10.13*	97.45 \pm 8.27*
	S	53.65 \pm 6.47**	41.34 \pm 3.53*	62.27 \pm 7.86**	52.5 \pm 3.71**	70.68 \pm 6.24**	63.12 \pm 3.97**
10.887	H	181.52 \pm 10.64*	108.91 \pm 4.49	162.53 \pm 10.79*	95.54 \pm 7.69*	134.25 \pm 12.24*	84.89 \pm 3.97*
	S	66.28 \pm 6.42**	64.24 \pm 4.39**	79.56 \pm 8.26**	62.71 \pm 7.43**	88.91 \pm 6.42**	72.43 \pm 5.34**
14.516	H	157.22 \pm 10.70*	106.77 \pm 12.49*	145.39 \pm 11.74*	97.96 \pm 8.97*	159.22 \pm 12.98*	87.47 \pm 10.04*
	S	84.61 \pm 12.16**	80.22 \pm 9.01**	95.24 \pm 7.92**	86.69 \pm 5.36**	101.31 \pm 8.75**	91.62 \pm 10.32**

Note : H: the GOT and GPT activity induced by 8-HOQ in the hepatocyte of *P. dabryanus*; S: the GOT and GPT activity induced by HOQ in the serum of *P. dabryanus*. Values followed by "*" and "**" mean significant difference at 0.5 and 0.01 level, respectively. ()

3.3. Comet assay and data analysis

Comet scores of control and exposed groups were shown in Figure 1. Figure 2 was made on the basis of three comet parameters of Trailing rate (%), tail length (μm) and tail moment by SPSS version 17.0, which summarises the results for Comet assay of liver cells in tested fish. The SCGE assay showed an obvious increase in three comet parameters in exposed fish, where the genome damage was significantly different (P<0.05) from control fish (Fig. 2). The DNA damage documented by the comet assay test, i.e. increased tail length paralleled by a reduction in head size (Figs. 1, 2), increasing with

treating time. For example, there was no apparent difference (P > 0.05) in the liver trailing rates between control and the lowest concentration exposed groups (3.629 mg l⁻¹) after for 6 and 24 h exposure to 8-HOQ. However, the hepatocytes trailing rates between control and the lower concentration exposed groups (7.258 mg l⁻¹) after for 6 h exposures to 8-HOQ had a significant difference (P < 0.05), and had a extreme significant difference (P < 0.01) between the higher exposed groups (14.516 and 29.032 mg l⁻¹) and their control after for other all treatment-time (Fig. 2A). The results indicated that the trailing rates of *P. dabryanus* liver

cells were increasing with the increase of exposed concentration and exposed time, which showed an obvious dose-effect and time-effect relationships (Fig. 2A). In the meantime, the tail length and tail moment showed similar results (Fig. 2B and 2C).

According to comet classifications, the DNA damage of control fish liver cells all belonged to class 1 (Fig. 1A, TM < 5). However, there was different class DNA damage of exposed fish liver cells at four treatment-times. For example, there were low damage cells in the lower concentration

treatment-groups (3.629, 7.258 mg l⁻¹) by exposed 6 h (Fig 1B, class 2, TM: 5-20). The mid damage cells (class 3, Fig 1C, TM: 20-60) appeared in all treatment-groups by exposed 24 and 48 h for 8-HOQ. And the high damage cells (class 4, Tail moment > 60, Fig. 1D) were noted in the highest concentration treatment-groups (29.032 mg l⁻¹) by exposed 48 h and longer than it for 8-HOQ. The comet assay results showed that 8-HOQ can induce obvious genotoxicity to *P. dabryanus* liver, which also showed dose-effect and time-effect.

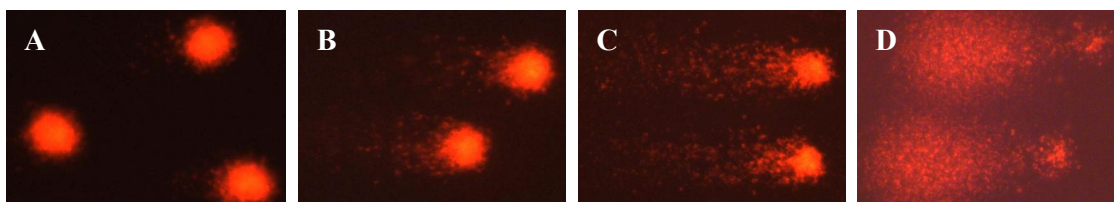


Figure 1. The Comet scale of *P. dabryanus* liver cells induced by 8-HOQ ($\times 400$), a four-class classification based on tail moment (TM) adopted from Anderson et al (Anderson et al. 1994)

Note: A: control cells (CC, TM < 5); B: low damage cells (LDC, TM 5-20); C: mid damage cells (MDC, TM 20-60); D: high damage cells (HDC, TM > 60)

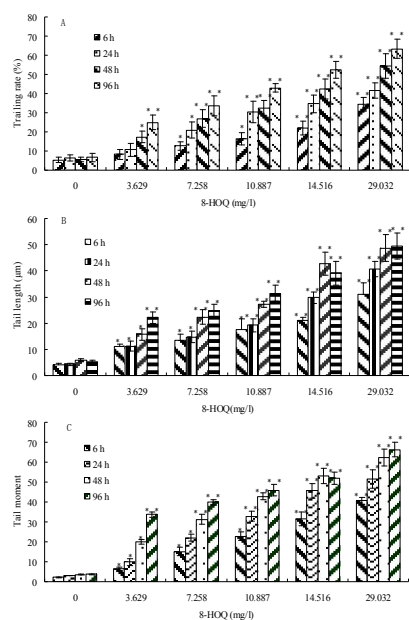


Figure 2. DNA damage effect of 8-HOQ on liver cells of *P. dabryanus*: A. Effect of 8-HOQ on the % trailing rate of tested fish liver cells; B. Effect of 8-HOQ on the tail length of tested fish liver cells; C. Effect of 8-HOQ on the Tail Moment of tested fish liver cells

Note : Values followed by “*” and “**” mean significant difference at 0.5 and 0.01 level, respectively.

4. Discussion

This study contributes significantly to the knowledge of 8-HOQ ecological consequences in the

aquatic environment. LC₅₀ can generally represent the degree of toxicity of toxicants, and thus it is the most important index that must be determined in an acute toxicity test (Li et al., 2012). In the present study, the acute toxicity data of 8-HOQ on adult *P. dabryanus* (Table 1) indicated that 8-HOQ had marked acute toxicity to loach. The 24 h, 48 h, and 96 h LC₅₀ of 8-HOQ on *P. dabryanus* were 69.7 ± 3.29 , 48.35 ± 4.23 and 28.53 ± 2.84 mg l⁻¹ respectively (table 1), which were lower compared with other environmental pollutants (Zhang et al., 2008; Seok-Ki et al., 2010). And tested fish (especially in the highest concentration 8-HOQ-treatment group, 72.58 mg l⁻¹) had showed typical toxic symptoms as follows, moving about and writhing quickly at the initial phase of test, slowing in reacting and losing balance gradually after several hours, then died finally. The bodies of dead fish were forniciform frequently and their liver were swelling and aterrimus, which is similar to the research results about the fish toxic symptom induced by heavy metal and others poisonous substances (Palanivelu et al., 2005; Nurullah et al., 2010; Vijaya et al., 2011; Li et al., 2012). The results of above suggested that 8-HOQ had strong toxicity effect to organisms, especial in aquatic organisms. Subsequent GPT/GOT activity assay and comet assay also showed that 8-HOQ could induce obvious physiological effect and genotoxicity in tested fish liver.

The liver or hepatopancreas, is the target of toxic chemicals usually. The results of our physiology toxicity test also indicate that liver is the major target organ of 8-HOQ to fish. In the process of participating in organism metabolic activity on poisonous substances, liver could be damaged in

varying degrees (Palanivelu et al., 2005). And the enzymatic activity changes of liver major enzymes also reflect the damage degree of animal liver. Under normal circumstances, aminotransferase is mainly in hepatic cell cytoplasm, only a few GPT and GOT are released to blood serum. When pathological changes or damage induced by foreign toxic substances take place in liver, the cytomembrane permeability of hepatic cell would change, a lot of GPT and GOT in liver would seep into blood plasma. Therefore, the activation of liver aminotransferases would decrease when organism is in toxication, meanwhile, the blood serum aminotransferases activation would increase (Yin et al., 2011). In this study, the enzyme content in loach liver was remarkably declined and showed concentration-effect and time-effect (Table 3). However, the enzyme level in blood serum was significantly promoted when compared with the control and had showed concentration-effect and time-effect also (Table 3). Those results suggested that the physiology toxicity induced by 8-HOQ maybe occur in the adult loach. This result was coincide with the results of others studies (Storelli et al., 2005; Tepe et al., 2008; Tigano et al., 2009; Yin et al., 2011; Saravanan et al., 2012).

The comet assay or single cell gel electrophoresis (SCGE) assay is a rapid, sensitive and relative simple method for detecting microscopically DNA damage at the level of individual cells. The main advantages of the Comet Assay include: (a) the collection of data at the level of the individual cell, (b) a small number of cells per sample (<10,000) is needed, (c) sensitivity for detecting DNA damage and (d) use of any eukaryote single cell population both in vitro and in vivo. Therefore, comet assay is one of the very widely used assays to toxic test, such as eco-genotoxicological studies and environmental monitoring, regulatory genotoxicological studies, and this assay has also been used to show protective effects of different dietary factors in chemo-preventive studies (Bichler et al., 2007; Cheryl et al., 2012, Eşref et al., 2010, Capriglione et al., 2011). In our present study, three parameters of trailing rate (%), tail length (µm) and tail moment were adopted for detecting the DNA damage effect of 8-HOQ on loach. The results indicated that 8-HOQ had obvious DNA damage effect to loach liver cells, and showed a clear dose-effect. Those results were also in accordance with that of enzyme activity assay, which illustrated that 8-HOQ could not only damage DNA, but also affect the normal physiological metabolic activity of liver in organism. This research result indicated the potential of 8-HOQ to damage fish cells, which was in accord with the similar findings in mammalian cells (Chang et al., 1991).

In conclusion, this study revealed that 8-HOQ could have obvious biotoxicity effect on *p. dabryanus*. And the liver of tested fish was a visceral target of 8-HOQ. 8-HOQ exposure could induce

significant physiology effect (change in hepatic enzyme activity) and genetic toxicity (DNA damage) in liver, which might be one of the possible mechanisms of 8-HOQ toxicity to aquatic animals. Therefore, this study provided some theoretical knowledge and guidance for safety application of 8-HOQ, and the prevention and control of 8-HOQ to environment pollution. In addition, the comet assay of DNA damage in loach liver cells adopted in this study offered a fast and sensitive method for monitoring water contamination.

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Preservation of Cultural Resources By Development of Tourism

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Abstract: In order to plan, develop, and manage tourism, economic, cultural-social, and cultural factors should be taken into account. Tourism bears positive and negative impacts on the environment and its own domain, and the environment may be effective on tourism procedure. The results of tourists' presence possess notable importance in the world, and at the micro level in Iran. This is notable in a way that the presence of tourists results in the emergence of positive and negative economic, cultural-social, environmental, infrastructural, and managerial impacts in the world and specifically in Iran. But what bears high significance in this paper is to study and evaluate the impacts of tourism in the present situation in Iran's territory which is considered a special condition. Iran is considered a country accepting tourists in the region due to its natural, historical, and man-made (ancient) attractions, and its special geographical and historical position. A country of such possessions and capabilities shouldn't be taken away of the economic outcomes because of political problems and cultural-resources worries, and on the other hand the international tourism market shouldn't deprive itself of Iran's tourism attractions.

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1. Introduction

Tourism has an obvious role in cultural convergence and stable peace in the region and the world. Tourism can create many undiscovered economic, commercial, cultural, and social resources and interests for countries. Developing tourism will lead to knowing the spiritual culture, changing economy and commerce, earning full money, making more job opportunities, and fading inappropriate and unreal images of the negative advertising done by other societies toward the host society. Tourism can revive the national pride, cultural manner, and protect the cultural and social heritage. Developing tourism helps build cultural and social self-confidence, develop cultural exchanges between knowing and understanding cultures, and create a dialogue of civilizations in the present time. At present, a mutual relationship between culture and tourism has emerged. Culture is thought to be a significant resource to develop tourism, and tourism has great share in cultural development. Although, there are a lot of materials on different impacts of tourism management in the relevant resources and the studies in this area, its cultural-social impacts have not been well introduced.

These impacts refer to the fact that tourism and traveling affect the society's value system, behavioral patterns, social structures, quality and style of living in the host society and those of the tourists', while the main focus of these studies are on the purposes of tourism, and the areas sending tourists and the tourists themselves have been paid less attention to [Richards G. and Wilson, J. (2007)].

Tourism: The word "Tourism" is taken from the word "Tour" which originates from the Latin word "Turns" meaning "Go around", "Go and come back between an origin and a destination", and "Orbit around", which entered Spanish from Greek and finally came into English. In Webster dictionary, tourism refers to a trip which is taken to a destination and is followed by a return to the residing location. In Longman dictionary, tourism is defined as a trip taken as a pastime for entertainment. In Larousse dictionary, tourism is discussed as trip for entertainment. However, the word "Tourism" has transformed from the individual meaning to a collective whole for travelling in the developed countries since the 19th century and specially the early 21st century, a time when fast means of transportation replaced the old ones [OECD (2009)].

In the social dimension, tourism is the interface between the normal lives of local residents and abnormal lives of the tourists [Peter E. Murphy.1983]. In March 1993, United Nations statistical commission accepted the definition of tourism presented by the World Tourism Organization. By this definition, tourism is "A set of activities done by people who travel to locations out of their workplace or residential area in order to rest and recreate or do other stuff, while not spending more than a year in those locations constantly." [McIntosh, R.W. ,Goeldner, C.R. and Ritchie, J.R.B. (1995)]

Culture: Cultural dimensions bear high significance while studying human and society behavior. Cultural

dimensions are also important in the international environment, because there are clearly cultural differences between countries. Culture can be defined Journal of American Science, 2012;8(4) as a set of thoughts, beliefs, values, wisdoms, and behaviors which compose the common base of every social action. However, this definition is not as comprehensive as it should be. Culture underlies all the actions and features of life [Enzensberger, Hans's Magnus.1994]. Anthony Burger defines culture like this, "Everything man does and doesn't do." [Sheldon, P. 2003]

A country's culture is crystallized and visualized on the stamps, the lifestyle of people, dressing code and make-up of people, every institute in that country, street sceneries, markets and buses ,patterns, general appearances and features of cities and villages, and any other factor relating that country (its main feature) or regarding the habits and costumes of the people [RICHARDS, G. and RAYMOND, C. (2000)].

2. Tourist reception

Tourist reception in a region is formed socially and culturally in order to revive habits and costumes, and tourist's visiting the societies leads to an experience and a memory. The type of host-tourist behavior in a region can be looked at in three perspectives:

1. The interactive perspective: This perspective visualizes the type of host-tourist behavior as an identifiable incident with a number of positive and negative outcomes so that it depends on the researcher's approach towards the righteous path of research and development to evaluate.
2. The negative perspective: The function-oriented perspective includes various social elements which may consider every change, specifically such as abnormalities, as a consequence of tourism.
3. The positive perspective: This perspective mainly considers the positive aspects of cultural changes which are made by tourists to reshape skills and costumes such as hygiene, discipline, etc. [Dogan, H. (1989)].

3. The impact of travel on the traveler

Even if people have little curiosity or cultural sensitivity, traveling puts them in contact with various environments. Even if they meet few local people, they will still obtain a set of experiences and touches. At least, they will learn something about the scenery and view of a place, local foods and drinks, and people's behavior. There are just a few people traveling with the purpose of discovery. These people are mostly interested in learning about various cultures. They may even become interested in research and study about a

country's culture such as history, art, language. Every passenger is impressed in a way. Their tastes, experiences, and perceptions get more global. Traveling helps the process of globalization. In fact, it helps the cross-culture interactions and exchanges which try to make people's lifestyles closer and more like [Tekin, A.1994].

The role of religion

Religious beliefs are of deepest emotions. In many countries, religious beliefs form the strongest factors affecting the way people deal with life. Wherever a religion gets strongly institutionalized, it can play a huge role in people's lives; in fact, there is a slight difference between religion and politics. The constitution accepts an official religion so that the connections become clearly obvious. Religion has always had a crucial role in augmenting the sense of national identity .Being English means being Christian, coming from Mali means being Muslim, being Balinese means being Indo. However, being English, Muslim, or Indo (not Christian) has now become the part of a new culture, and its importance has increased with the total change and development of national identity. Although it can be easily observed that visitors are the followers of a religion and the local people are the followers of another religion, there are some reasons for confrontation and clash. Different religions are expected to show more tolerance and feel greater responsibility for other religions. Thought exchange among religions can form a positive part of tourism alongside other cultural activities [WorldTourism Organization. 1999].

4. New tourism destination

Tourism directly affects the lives of people living in the tourism areas. Some impacts of tourism development on a new destination are reviewed in the following: -Tourism development creates new job opportunities.

This makes people leave their current jobs, because jobs relating to tourism are more promising, pleasant, and cleaner.

-Many women find the opportunity to work outside the house for the first time in their lives. This provides them with new skills, reliability, confidence, and financial power.

-There may not be enough people to fill jobs in the region, so development needs that foreign workers enter the country as a steady process. These workers bearing different cultural characteristics form relationships with local people, marry them, etc.

-Tourism increases people's incomes. They can buy things and do stuff that they were never be able to.

-Development increases government's power to improve educational and sanitary systems. A better

education level and healthier people present a different viewpoint of life. There will be a large gap between young people and the elderly.

-House-building will be improved, and different plans of housing will be implemented.

-Machines and technological systems will replace human labor force.

Tourists are customers who are willing to buy handicrafts and spend money on works of arts and living art stuff, so traditionally cultural activities are put in the process of commercialization [World Tourism Organization. 1999].

5. Contemptuous opinions about services

Services are considered in an undermining way in most cultures, and it's true at both national and individual levels. A country may look at tourism as a low priority. Politicians may prefer producing other stuff to presenting services to the visitors. They say, "We are not a serving nation." They may see tourism in a way that an individual is at other people's disposal and is always ready to follow foreigners' orders to serve them, so this matter may be considered contemptuous. Similarly, young people have evaded tourism jobs. Serving others is blamed. Nowadays, tourism has overcome this point of view.

However, tourism must save its own general appearance as a modern and international section which is able to present competitive and interesting job opportunities [Dwyer, L. 2003].

6. Extreme price changes

Extreme price changes mostly emerge when people are able to spend more money because of their net incomes. Given this buying power, prices are inflated to exploit tourists. This is a result of opportunism and greed. Tourists are badly punished. Some of them are definitely generous and patient, so they pay high prices without hesitation. But, some others resist this extortion. Tourists' unawareness of prices creates a potentially harmful situation. The lists of prices must be prepared and distributed.

Equally, two-price systems, in which local people pay a price and visitors pay another one, represents an new source of price fluctuation, and tourists are easily abused. Market mechanisms can be freed to function, but visitors must be informed of locally normal prices [Richards, G. (1993)].

1) Positive and negative consequences

-Enhancing visitors' ability to speak the local language.

-Host society's tendency to accept and learn other languages in order to communicate more with the tourists.

-Creating and reinforcing the sense among residents to know themselves.

-Helping local residents to obtain more knowledge of their local culture.

-Increasing cultural and national pride.

-Creating suitable situation for residents' active participation in introducing the culture of region to guests.

-Reviving and keeping local myths and folklores that might go into oblivion.

-Reviving local arts and costumes.

-Encouraging locally economic growth through cultural tourism and reviving historical-cultural works and buildings.

2) Positive social impacts

-Improving the quality of life.

-Increasing recreational and entertainment facilities.

-Improving the quality of urban services.

-Improving the quality of public services (health, police, etc.)

Generally, the consequences and vista of tourism can be sought out in differentiating and publicizing places, globalizing and internationalizing tastes and features alongside the opportunities for people's more creativity regarding the world [Hallinshead, keit (1999)].

3) Negative cultural-social impacts

-Increase in prostitution.

-Alcoholism growth.

-Increase in crimes.

-Formation of wrong local cultures.

7. Discussions

Tourism as a stable and continuous fact at the present time can present its abilities in a stable developing path in social structures through various symbols, and it can be studied by looking at communication system between the people of destination region and the tourists through different aspects of costumes and behaviors, productions of material and spiritual cultures, and also cultural social tendencies and states which are risen for the tourists spiritually in an aesthetic way, and as the existing reality in the society and social life of people. Generally, the dimensions of tourism's positive impacts can include the following items in the cultural-social experience framework.

First, if the interests of tourism are evenly distributed in the societies and regions of a country, the quality of people's lives will be improved. Second, developing tourism in consequence of exchanges made between local people and tourists, family's structure will be modernized. Third, domestic tourism may spread a kind of cultural-social integrity and convergence inside the country. Fourth, tourism can help the expansion and maintenance of cultural

heritage, museums, theaters, and other cultural facilities in the societies. Finally, tourism development may create opportunities for cultural exchanges between tourist and local people so that these two groups can get to know each other's cultures more and respect each other gradually.

Generally, the negative dimensions of tourism include the following issues in cultural-social terrain.

Economically, too high density of tourists may cause inflation of goods and services in some destinations, so the local residents become angry and hostile toward the tourist. First, cultural relic's destruction. If tourists visit a historically important place too much, it may become destroyed gradually. Second, cultural transformation, with particular attention to the fragility of cultural heritage, tourism development may cause damage to the authenticity of local cultural patterns. Third, cultural invasion. It is possible that local values turn to materials due to extreme efforts in making tourism appealing and dealing with cultural aspects over commercially in tourism destinations. Finally, corruption and social disorders. Tourism may intensify social pathologies such as alcoholism, addiction, crime, and prostitution.

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Absurd and Morality

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Abstract Absurd theater is an art which represents absurdity of life in the contemporary western countries as a fundamental objection against the absurd life the meaning and values of which are not the pre-given or natural affairs, but they are originated from human practice and the relation that he has given to the world. Meaninglessness of the life and world is the product of the action, practice and existential experience of the wise creatures who cannot give the meaning to the life and its world any more. This disability of the human being in giving meaning to the life is the climax of fall from all transcendental ascents which have shined as stars in the different periods of the mankind civilization. The morality is the most important ascent which has been experienced through catastrophes and disasters and gradually and step by step it has been threatened and become infirm. Seemingly, meaninglessness of life is product of weakening and probably destruction of morality basis. By domination of modernity, the ethic became independent and actually it lost the religion protection (lose of traditional basis of morality). In other words, the morality remained valid only by leaning on humanity and human reason. The statements by great thinkers as Marx, Nietzsche, Freud on the costly position of human as God's representative or kin as well as scientific advantages of the scientists as Copernicus, Coupler, Galileo, Darwin, Heisenberg and Einstein against the prevail understanding of human and his position in the existence, made the human's base and status unstable and eventually the world wars did the final stroke. Because of its disconnection from religion, the morality, which had only leaned on the human, became seriously unstable and weakening by these events and fall of human's base as a creature from the ancestry of animals (Darwin), slave of instincts (Freud), plaything of historical condition (Marx), restricted in his/her own perspective (Nietzsche)... (Lose of Modern morality basis). Absurdity of absurd theater in the structure and content, which is experienced from this structure, is represented. Absurdity is perceived not only from the totality of structure but the various parts of structure. The effete or outcast characters or both of them, the trivial subjects which are lack of great values of human, dialogues including the words with out communication and non-solidarity phrases, cyclic constructions which are without the current climax and fall of classic and neo-classic, absence of conflict between the opposite ideas and the characters who protect the ideas are the structural elements which generally express absurdity of human life as well as independently narration of this tragedy. Considering the great works including *Waiting for Godot* (Samuel Becket), *Rhino* (Eugene Ionesco), *Return to Home* (Harold Pinter) prove this condition.

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Key words: absurd theater, morality, value, meaning, modernity

1. Introduction

In the end of middle centuries, the western human gradually refused the autonomy of church in order to become-as Kant had wished-mature, which is, he/she himself/herself seeks and find episteme. This maturity was venerating the reason in one side and disdaining any reference in the other side. Reason had reached to the divine dignity and it intent to establish a paradise, which has been promised by the religious books in "the next world", on the earth. Francis Bacon and Descartes promised this "wrought – reason" paradise in the beginning of modernity. Independent from its religious basis, even moral affair was seeking by help of reason. Kant assumed the moral independent from any intrest, praised and authentic and he had designed firm reasoning in expression the

pure moral affair. The moral goodness and badness, according to Kant, is perceived by every body through the reason and we ought to try to live according to this perception. In other words, he expressed the morality of liberal and senior, but he had taken an important protector from morality, that is, religion. Although the morality is free and sublimated from any promise, undoubtedly most part of society contains the common people who are not familiar to the liberal eth morality and their good and evil deeds are not resulted from the rational understanding but for paradise or fear from hell. Destruction of the religion protection seriously destroys the morality base or leastwise abstinence from non-moral affair in them.

Although Kant destroyed the religious protection of morality as well as the leaning on the

vulgar, the background for this destruction had been provided before him. Corruption, oppression and ignorance of the church authorities had blemished the religion validity; therefore, it (religion) had not the competence of authority and preferentiality. Reading of the Catholic Church from Christian had been spread to the area of policy and political incompetence of and inefficiencies were not detachable from the religion capabilities; consequently, every thing was ascribed to the religion. Also, illegitimacy of the believers or custodians of religion were not detachable from the religion itself; therefore, it was recorded for religion. The vain champion of the church with scientific advantages (Galileo, Ceppler, Jerdano Brono...), the huge wealth of the clergymen who were promoting the ascetic life of Christ inconsistently, incompatibility of the church training and human instinct tendencies (as bachelorhood, disdain wealth and comfort...), torture and murder of innocent people by excuse of ancisation, the criminal and at the same time unhappy crusade wars, disability of the church in explaining and justification of the new advantages of sciences were the issues that determined the end of the church paradigm.

By scientific struggles, philosophical optimism, and protection of the artistic movements, modernity determined a condition in which all religious and superstitious promises in the people's belief... the ideal world or utopia and promised paradise is possible in this earthly world by use of reason. But exactly in the moment in which all modernity ideals were in stage of certainty, namely beginning of 20th century, the time which the reason could achieve the domination of human on the world and open the gates of sky to him, the world wars, poverty, corruption..., gad between the classes, domination of bureaucratic system and injustice rules of market and the work area persuaded people to believe that the new God (reason) in constructing paradise not only was unable but instead it has constructed a horrible hell.

2. Research Method and Research Consideration

In this research, which focuses on representation of morality absurdity in the absurd theater, studies three major works in the area of absurd dramatic literature, that is, Samuel Bucket's *Waiting for Godo*, Eugene Ionesco's *Rhino* and Harold Pinter's *Return to Home*. Consequently, the central thought of the works which is representation of the morality absurdity become clear. Some books have been referred to as the references for explaining the absurd, but it has been preferred to avoid the effect of the other critics and analyst for analyzing the plays.

3. Consideration and Discussion

The pioneer philosopher of modernity in the 17th, Descartes had said "I think then I exist." This statement, which announced the modern humanist domination, was carrying a great truth in itself. "Human's existence is his/her thought." Without thought, human is not a human. Two centuries after Descartes, the professional reason of human had made an instrument which has destroyed human existence in the shortest possible time. People in Hiroshima and Nagasaki by the product of the reason to which, in Descartes terms, human's existence was conditioned were destroyed as soon as possible.

Therefore, human's thought had determined his/her living not his/her existence. The thinking human had thought this living burden. Therefore, it had to say "I think then I existed."

This is the time of absurd emergence, a theatrical movement which was pioneered by its greatest precursors including Samuel Bucket, Eugene Ionesco, Jan Jene and Arthur Adamov. The early plays of Edward Albee and Harold Pinter also fall in the scope of this category... [1]. Moreover, we can find the similarities between the works by Chekhov, Sartre, Camus, Eanil and those of this movement. The most works of this movement are in French language (Bucket, Ionesco and Jene); therefore, it is originally a French movement. Art mirrors human and reveals the existence and essence of human in a non-conceptual experience and this ability is incomparable to the manifestations of human life. Absurd theater represents the western contemporary human realistically. The western human, after the spree of rejection the religion and belief in the god of reason, wondering and unconscious was proud of reason. He had not any base to which fight or by which rises against that condition. Being alive and no living was the story of the contemporary human from the perspective of absurd theater. Regarding the condition of emergence of absurd theater Bracket said: "theater was exceedingly influenced by the war horrors and possibility of its repetition. The thinkers of the period have planed the fundamental questions about human's responsibility and even his survival. The very themes of the plays are anxiety and feeling of sinfulness." [2]

This horror and anxiety captured the scene of theater. Theater was no more an entertainment for instruction or purification. It was, like others, captured by anxiety and its representation was horrible. Millions people had been victimized by the world wars because of only two atomic bombs. In the end of War II, thousands atomic bombs were prepared by different countries to target one another. As a result, if this new war (nuclear war), the scope of which was all over the world, was occurring, it would had annihilated human for ever.

About the setting and atmosphere of absurd work, Hans Leh Man has said: “the atmosphere in which absurdity theater is living has the political, philosophical and literary causes: the experience of savagery in the twentieth century, possibility of the end of history (Hiroshima), bureaucracies without meaning, political submission, an existentialist return on the individual and nihilism are in close interconnection.” [3]

This theater has been dismantled from what has been called as art mission or its capability since Plato to Nietzsche. It neither reveals divine truth (Plato) nor instructs people (Aristotle) or appears infinite idea (Hegel), it neither inaugurates a new way for life (Nietzsche) nor uncovers the existence (Heidegger). Power of contemporary theater has been judged by Peter Brook as: “there is not a living theater which answers to the real contemporary needs... theater do not satisfy any contemporary need, the people who used to go to theater, cannot get something from the plays; they can not achieve the least element for their routine future life... “. [4]

Optimism in the beginning of modernity was replaced by exceeding despair and disappointment. The hopelessly conditions had directed the life horizon toward a thick darkness; consequently, people were expecting the collapse and destruction of existence by outbreak of the destructive nuclear war. Expect of the worst condition, while nobody could do any thing, caused an exceptional experience. “When the atomic dust of Hiroshima and Nagasaki massacre settled down, when the horrible pictures of human burner’s kiln of Germany and ideological massacres by Russian were presented to the world, darkness and sorrow captured the theater scenes beyond the conventional notions.” [5]

This difficult experience gave the life a different meaning from what is in the philosophical or religious literature. There is always a way to escape from the crisis and catastrophe in the most pessimistic religious ideas. At least, religion promises the next world justly. The pessimistic philosophers also suggest some alternatives but all the alternatives seemed foolishly, ridiculous and deadlock in the new horrible experience. The lesson which the war, occupation and, in particular, explosion of the atomic bomb taught was too cruel that anybody accept to deceive itself or participate in the lies by others... Human wanted to demonstrate his/her own meaninglessness and nihilism in a nihilist world, and to question the thought habit and the most traditional and sacred beliefs and clearly understand the nothingness and vacuum to which humanity and human were running.” [6]

Absurd drama represents the horrible experience of life and the life accompanied by

horrible, anxiety, imminent waiting for catastrophe and nothingness. It is a condition in which a hero will not rise to change the unjust condition because human has lost the belief in the power to change the condition. In the condition in which the future is too much darker and ambiguous than the past, possibility of the fall of the sky roof on the head of the earth inhabitants, rejection of religious and non-religious belief, and the great victim is the morality. In this condition, everybody can do every thing because everything is legitimized or there is not any forbidden affair. Legitimized or illegitimated are the rules to order the society and facilitate possibility of life. When the life is unstable and existence is going to fall, there is not any a tomorrow in “the World” or “Area of the world” in which human suffers the vengeance of this world; therefore, every thing is legitimized. In other words, it is possible to keep the parents in the ash-bin (*End Game*), or betray the wife or husband in front of him/her... (*Return to Home*) or employ human as an animal (*Waiting for Godo*) or he was curious, when the Men became into animals, whether they the animal is one-horn or two-horn (*Rhino*)

The absurd artist writes against these conditions. He does not deliver a political manifest against the dominant conditions but, by revolution of the conditions and uncovering it for use, it exposes us to the terrible experience of the world which has been dismantled by our affair and its endurance is difficult and even impossible. Some affairs including “devastation of natural environment, the dangers of chemical bombs, atomic rubbish, developing the mass media in a way that the private identity is lost, change in the morality and eradication of any moral taboo... . [7]

Absurd artist is not a social reformist or political activist; therefore, it does not say us what we should or should not do to better the world. He reveals the world to us by the artist instruments, that is, what he has and the language by which he/she speaks. In other words, he/she clarifies the world to which we have gotten and we are unable to perceive its abnormality, irrationality or unfairness. But, the art through defamiliarization attracts our attention. The artistic instrument of the absurd artists is exerted so: “in their works, they put some elements which are in the common atmosphere and, instead of the cause and

effect relations, they met the turbulent structures which are their current dramatic subject. In this way, the feeling of nihilism from the things which is by combination of developed incompatible accidents is produced and it is resulted in a serious- ridiculous and ironic atmosphere". [8]

Waiting for Godo is a brilliant example of lose of the basis particularly the basis of morality. "Estragon" and "Vladimir" are the wandering effete whose existences are summarized in vain delay and binds as carrot and turnip involving in a shoe and hat. Pertinence of the morality life is a purposeful and transcendental- based life and human's sublime has a relationship to his/her transcendental desires. The two features are not observed in the life of "Estragon" and "Vladimir". If we do not assume Godo as the product and creation of the melancholically imaginations, it is an unfaithful promised character who enjoys his own unfaithfulness and Estragon and Vladimir's waiting for him. Moreover, a child who sometimes brings lie promise of Godo's coming in the future is also his accomplice or the object of his action in this amoral practice. If we assume Godo as Estragon and Vladimir's imagination, it is another symbol of the childish life against which Kant had reacted and expressed improvement at human's maturity as "struggle to achieve episteme for its own self."

"Lucky", in this work, is a human-animal which carries load and shows the way, it beats and kicks if it is necessary, it recites lectures, and it is a strange mixture of dog, donkey and peacock and eats the reminder of the master's (Putsoo) food delightfully. "Putsoo", who seems the master, is a pathetic creature that is blind and lame and the autonomy of his masterhood is only and only a consequence of automatic slavery of lucky. Lucky's slavery represents the amoral life by "Lucky" who practice slavery and "Putsoo" who enslaves as the lie promise (by Godo and the child) with Estragon and Vladimir's is also amoral. If there is not any rule that protect such victimized men- it is the result of absurdity of morality- why is the morality and social protection the source and basis of rule in each society. Society does not show any reaction to this status. Does not it understand this condition? Estragon and Vladimir also represent society individuals, who are at the same time victims and indifferent in comparison to the Lucky slavery and they reveal indifferent society against injustice, fall of morality.

"Rhino" reveals another kind of absurdity of life. In a horrible condition in which the Men become into the harsh and dangerous animals, the dominant concern of society is to know whether the harsh and dangerous animals are one-horn or two-horn, Asian or African. Society has lost its sensitiveness for the norms and values and it does not react in opposition to

their violation. Eventually, "Branje" is the only human who has been not metamorphosed into an animal. His stability is not because of a particular ability or a firm belief in a transcendental value as a hero but because of his disability has remained a human. He has tried seriously to become a human. Even he has tried the voice of animals, but, he has remained, against his tendency, a human.

Absurdity of life is in a way in which to be a human is not a prominent accomplishment but an unwanted and unwittingly pathetic weakness! Interestingly Branje's behavior was blaming by others before the attack of Rhino and epidemic of becoming into Rhino, particularly by John. On those times Branje was going to work in a drunken condition. His cloth was not clean and tidy and his face was not shaved every day. In the position of a morality teacher-the role which was played by the priests- John continuously reminds Branje's duties in the position of a normal citizen and Branje also listens shamefully, but John, interestingly, is among the first people who become into Rhino! Branje, who is disabling to believe in the imposed social norms, is the only one who does not transform into a Rhino. Possibly, "*Return to Home*" is the most horrible experience among the work against absurdity of the life. The woman has been reduced into the tool for sexual passions and the sensual are regardless her husband's brothers and father and amazingly every thing happens in front of the husband. The woman decides to leave her children and stay among the group. Because of the costs, the group decides for prostitution of the women in the opportunities (e. g when the men are at work). Her husband is a designer of this alternative.

Traditionally the parents are remembered by their good deeds, but the family mother, against tradition, is remembered as the present bride of the family. The family uncle declares that they have experienced, like this condition, a successful competition to achieve their brother's wife! The children are in a serious competition, which is sometimes resulted in beating, to their father to attract the family bride. The children treat their father inferiorly and abusively.

The family uncle reveals his job capabilities proudly. He recognizes himself as the favorite driver of his rich passengers because he knows when he should or should not speak. He knows what he should or should not say so that satisfies the passengers! As if, Lucky of "*Witting for Godo*" has been repeated in a more tangible and believable way. Both are the tools dismantled from human emotions and they merely do what they have been wanted or it has originally been scheduled for them. Evaluation of the role and function and comparison to the morality principles is

impossible because morality is no more a transcendental value.

4. Conclusion

Absurd drama reveals the contemporary western human as it is or it is possible to be and revelation of total fall taken place by the morality litter. The prominent works of absurd theater represent the lost leaning of human morality. The morality, which was propagated by morality independently from religion, lost its validity when mortality ideals reached to deadlocks and became a subordinate from the needs of "body-diagnosis" which are common between human and animals. By disconnection of religion and morality, in the beginning of modernity particularly in Kant's works, morality merely leaned on human. Scientific and philosophical advantages and historical experiences have destroyed the basis of human validity that recognized from gods ancestry in the ancient Greece and God's child in the Middle Ages. Destruction of human validity actually recorded the only leaning of morality which was followed by meaningless and aimless life and absurd brilliant works show us this regretful condition.

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Load Frequency Control by using a new controller

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Abstract: In an interconnected power system, as a power load demand varies randomly, both area frequency and tie-line power interchange also vary. The objectives of load frequency control (LFC) are to minimize the transient deviations in these variables (area frequency and tie-line power interchange) and to ensure their steady state errors to be zeros. When dealing with the LFC problem of power systems, unexpected external disturbances, parameter uncertainties and the model uncertainties of the power system pose big challenges for controller design. Active disturbance rejection control (ADRC), as an increasingly popular practical control technique, has the advantages of requiring little information from the plant model and being robust against disturbances and uncertainties. In this paper an optimal load frequency controller for two area interconnected power system is presented to quench the deviations in frequency and tie line power due to different load disturbances. In classical LFC problems, PI type controllers are used to control of system. But due to some disadvantages of the PI type controllers, the researches are toward finding a better control scheme. Although many different advanced method have been carried out to LFC problem, but the industries are willing to use simple PI controllers. In this regard, PID type controller is used for LFC in this paper.

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Keywords: Load Frequency Control, Multi Area Electric Power System, PI Controller, PID controller

1. Introduction

Power systems are used to convert natural energy into electric power. They transport electricity to factories and houses to satisfy all kinds of power needs. To optimize the performance of electrical equipment, it is important to ensure the quality of the electric power. It is well known that three-phase alternating current (AC) is generally used to transport the electricity. During the transportation, both the active power balance and the reactive power balance must be maintained between generating and utilizing the AC power. Those two balances correspond to two equilibrium points: frequency and voltage. When either of the two balances is broken and reset at a new level, the equilibrium points will float. A good quality of the electric power system requires both the frequency and voltage to remain at standard values during operation. For North America, the standard values for the frequency and voltage are *60 Hertz* and *120 Volts* respectively. However, the users of the electric power change the loads randomly and momentarily. It will be impossible to maintain the balances of both the active and reactive powers without control. As a result of the imbalance, the frequency and voltage levels will be varying with the change of the loads. Thus a control system is essential to cancel the effects of the random load changes and to keep the frequency and voltage at the standard values. Although the active power and reactive power have combined effects on the frequency and voltage, the control problem of the

frequency and voltage can be decoupled. The frequency is highly dependent on the active power while the voltage is highly dependent on the reactive power. Thus the control issue in power systems can be decoupled into two independent problems. One is about the active power and frequency control while the other is about the reactive power and voltage control. The active power and frequency control is referred to as load frequency control (LFC).

Many control strategies for Load Frequency Control in electric power systems have been proposed by researchers over the past decades. This extensive research is due to fact that LFC constitutes an important function of power system operation where the main objective is to regulate the output power of each generator at prescribed levels while keeping the frequency fluctuations within pre-specified limits. A unified tuning of PID load frequency controller for power systems via internal mode control has been proposed [1]. In this paper the tuning method is based on the two-degree-of-freedom (TDF) internal model control (IMC) design method and a PID approximation procedure. A new discrete-time sliding mode controller for load-frequency control in areas control of a power system has been presented [2]. In this paper full-state feedback is applied for LFC not only in control areas with thermal power plants but also in control areas with hydro power plants, in spite of their non minimum phase behaviors. To enable full-state feedback, a state estimation method based on fast sampling of

measured output variables has been applied. The applications of artificial neural network, genetic algorithms and optimal control to LFC have been reported in [3-5]. An adaptive decentralized load frequency control of multi-area power systems has been presented in [6]. Also the application of robust control methods for load frequency control problem has been presented in [7-8]. In this paper PI and PID type controllers are simulated and compared. The parameters of the proposed controllers are tuned by using Tabu search (TS).

2. Plant model

Fig. 1 shows a two-control area power system which is considered as a test system. The state-space model of the system is as (1) [9].

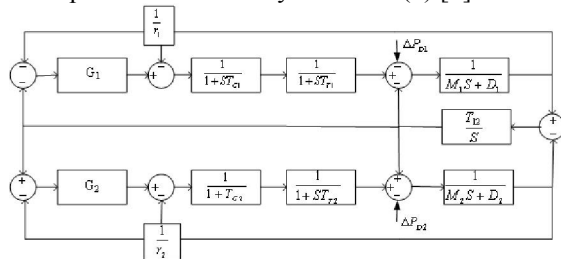


Figure 1: Two-area electric power system for LFC studies

$$\begin{cases} \dot{x} = Ax + Bu \\ y = Cx \end{cases} \quad (1)$$

Where:

$$u = [\Delta P_{D1}, \Delta P_{D2}, U_1, U_2]$$

$$y = [y_1, y_2] = [\Delta f_1, \Delta f_2, \Delta P_{tie}]$$

$$x = [\Delta P_{G1} \quad \Delta P_{T1} \quad \Delta f_1 \quad \Delta P_{tie} \quad \Delta P_{G2} \quad \Delta P_{T2} \quad \Delta f_2]$$

$$A = \begin{bmatrix} -\frac{1}{T_{G1}} & 0 & -\frac{1}{r_1 T_{G1}} & 0 & 0 & 0 & 0 \\ \frac{1}{T_{T1}} & -\frac{1}{T_{T1}} & 0 & 0 & 0 & 0 & 0 \\ 0 & \frac{1}{M_1} & -\frac{D_1}{M_1} & -\frac{1}{M_1} & 0 & 0 & 0 \\ 0 & 0 & T_{12} & 0 & 0 & 0 & -T_{12} \\ 0 & 0 & 0 & 0 & -\frac{1}{T_{G2}} & 0 & -\frac{1}{T_{T2}} \\ 0 & 0 & 0 & 0 & \frac{1}{T_{T2}} & -\frac{1}{T_{T2}} & 0 \\ 0 & 0 & 0 & \frac{1}{M_2} & 0 & \frac{1}{M_2} & -\frac{D_2}{M_2} \end{bmatrix}$$

$$B = \begin{bmatrix} 0 & 0 & \frac{1}{M_1} & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & \frac{1}{M_2} \\ \frac{1}{T_{G1}} & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & \frac{1}{T_{G2}} & 0 & 0 \end{bmatrix}$$

The parameters of model, defined as follow:

- Δ: Deviation from nominal value
- M=2H: Constant of inertia
- D: Damping constant
- R: Gain of speed droop feedback loop
- T_i: Turbine Time constant
- T_G: Governor Time constant
- G₁: First area controller
- G₂: Second area controller

The typical values of system parameters for nominal operation condition are as follow:

$$T_{T1} = T_{T2} = 0.03 \quad T_{G1} = T_{G2} = 0.08$$

$$T_{P1} = T_{P2} = 20 \quad R_1 = R_2 = 2.4$$

$$K_{P1} = K_{P2} = 120 \quad B_1 = B_2 = 0.425$$

$$K_1 = K_2 = 1; a_{12} = -1 \quad T_{12} = 0.545$$

Where, the footnote 1 indicates first area parameters and footnote 2 indicates second area parameters and the parameters of two areas are considered equal.

The objectives are Design G₁ and G₂ in Load Frequency Control (LFC). As referred before, many methods have been carried out to design these controllers so far. In this paper PID type controller is considered to control of system. A Meta heuristic optimization method named TS is used to tuning the proposed controllers.

3. Design methodology

The PID controller performance is evaluated on the proposed test system given in section 2. The parameters of the PID controllers are obtained using TS. In the next subsection a brief introduction about TS is presented.

3.1. Tabu search

Tabu search (TS) was first presented in its present form by Glover. Many computational experiments have shown that TS has now become an established optimization technique which can compete with almost all known techniques and which - by its flexibility - can beat many classical procedures. Up to now, there is no formal explanation of this good behavior. Recently, theoretical aspects of TS have been investigated [10]. The success with TS implies often that a serious effort of modeling be done from the beginning. In TS, iterative procedure plays an important role: for most optimization problems no procedure is known in general to get directly an "optimal" solution.

The general step of an iterative procedure consists in constructing from a current solution x_i a next solution x_j and in checking whether one should stop there or perform another step.

In other hand, a neighborhood $N(x_i)$ is defined for each feasible solution x_i , and the next solution x_j is searched among the solutions in $N(x_i)$.

In this part we summarize the discrete TS algorithm in four steps. Assume that X is a total search space and x is a solution point sample and $f(x)$ is cost function:

- 1- Choose $x \in X$ to start the process.
 - 2- Create a candidate list of non-Tabu moves in neighborhood. ($x_i, i=1,2,\dots,N$)
 - 3-Find $x_{winner} \in N(x)$ such that $f(x_{winner}) < f(x_i), i \neq winner$.
 - 4- Check the stopping criterion. If satisfied, exit the algorithm.
- If not, winner $x = x_{winner}$, update Tabu List and then go to step 2.

In order to exit from algorithm, there are several criterions that are considered in our research.

- 1- by determining a predetermined threshold: If the value of cost function was less, algorithm would be terminated.
- 2- Determination of specific number of iterations.
- 3- If the value of the cost was remained invariable or negligible change for several iterations, algorithm would be terminated.

A didactic presentation of TS and a series of applications have been collected in [10].

3.2. controller adjustment using TS

In this section the parameters of the proposed PID controllers are tuned using TS. In optimization methods, the first step is to define a performance index for optimal search. In this study the performance index is considered as (2). In fact, the performance index is the Integral of the Time multiplied Absolute value of the Error (ITAE).

$$ITAE = \int_0^t t|\Delta\omega_1|dt + \int_0^t t|\Delta\omega_2|dt \quad (2)$$

It is clear to understand that the controller with lower ITAE is better than the other controllers. To compute the optimum parameter values, a 10 % step change in ΔP_{D1} is assumed and the performance index is minimized using TS. It should be noted that TS algorithm is run several times and then optimal set of parameters is selected. The optimum values of the PID parameters are obtained using TS and summarized in the Table 1.

Table 1. Optimum parameters of PID controllers

	K_p	K_i	K_D
First area	13.43	45.71	1.04
Second area	6.22	70.23	1.10

In order to comparison and show effectiveness of the proposed method, PI type controller optimized by TS is incorporated for LFC. The optimum value of the PI controllers Parameters are obtained and summarized in the Table 2.

Table 2. Optimum values of K_p and K_i for PI controllers

	K_p	K_i
First area	11.7321	43.1291
Second area	5.5099	66.7739

4. Results and discussions

The results are carried out on the multi area test system with the proposed PID and PI controllers. Three operating conditions are considered for simulation as follows:

- i. Nominal operating condition
- ii. Heavy operating condition (20% changing parameters from their typical values)
- iii. Very heavy operating condition (40% changing parameters from their typical values)

In order to demonstrate the robustness of the proposed method, The *ITAE* is calculated following step change in the demand of first area (ΔP_{D1}) at all operating conditions (Nominal, Heavy and Very heavy) and results are listed at Table 3. Following step change, the PID controller has better performance than the PI controller at all operating conditions.

Table 3. 10% Step increase in demand of first area

	The calculated ITAE	
	PID	PI
Nominal condition	0.0025	0.0055
Heavy condition	0.0041	0.0681
Very heavy condition	0.0066	0.0112

Figure 2 shows $\Delta\omega_1$ at nominal, heavy and very heavy operating conditions following 10 % step change in the demand of first area (ΔP_{D1}). Each figure contains two plots as solid line for PID controller and dashed line for PI controller. It is seen that the PID controller has better performance than the other method at all operating conditions.

5. Conclusions

This paper presented the application of a new control scheme for LFC problem. PID type controller has been successfully carried out for LFC problem. The parameters of the proposed PID controller have been tuned by using TS. The proposed PID controller had significant priority rather than PI controller. The simulation results which have been carried out on a two-area electric power system showed the viability of PID controller. The PI

controller is the most commonly used controller in the industry and practical systems, therefore the paper's results can be used for the practical LFC systems.

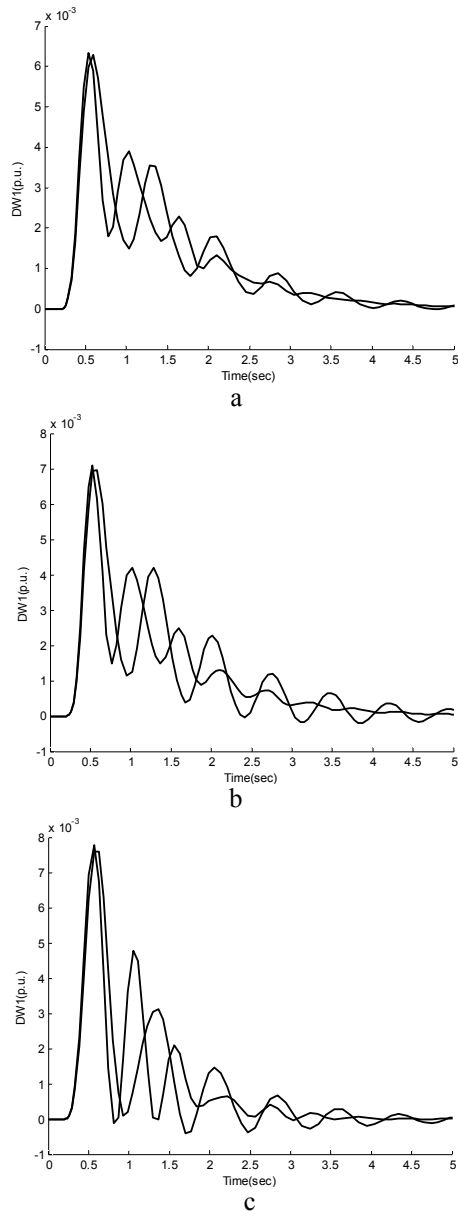


Figure 2: Dynamic response $\Delta\omega_1$ following step change in demand of first area (ΔP_{D1})
 a: Nominal b: Heavy c: Very heavy
 Solid (PID controller), Dashed (PI controller)

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Pharmacodynamics of gonadotrophin releasing hormone (Receptal[®]) and prostaglandine (Estrumate[®]) on ovarian activity, hematological picture and some steroid hormones of cows during summer season

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Abstract: Treatment of infertility and induction of estrous cycle in cows is usually based on **progestagens, prostaglandin and gonadotropin releasing hormone** or its analogues. gonadotropin-releasing hormone from hypothalamus a polypeptide hormone was found to regulate the secretion of luteinizing and follicle stimulating hormones. The present study aimed to study the pharmacodynamics of GnRH and PGF₂α (OvSynch protocol) treatment on cattle fertility and some biochemical and hormonal status during summer season. Fifteen cross breed cows (Baladi X Abundance), 9 primiparous and 6 pleuriparous cows, their ages (4.84±1.91) years, not exhibiting estrus signs for duration of 108±45.52 days, were treated with OvSynch protocol (GnRh-PGF₂α-GnRh). Cows were injected with 2.5ml Receptal[®] (GnRH) I/M. Seven days later, 2ml Estrumate[®] (PGF₂α) was injected I/M., followed 48 hours later with injection of 2nd dose of GnRH (2.5ml Receptal[®]), I/M., then timed artificial insemination (T.A.I.). Blood samples were collected from jugular vein along the experimental period. Complete blood picture, Serum progesterone and estradiol-17β were assayed. The response to treatments were determined using rectal palpation and ultrasonography (US). The results revealed that no marked changes were occurred in blood parameters (RBCs, MCV, HCT, HB, WBCs, LY% & GR%), after treating with the drugs, Receptal[®]-Estrumate[®]-Receptal[®] (OvSynch protocol). But it is clear that, these fluctuations in blood parameters lie within the normal blood value range of cattle. A high estradiol level (35.9±3.62pg/ml) was recorded at day (10) at fixed TAI and lowest one (21.03±0.79pg/ml) was detected at day (73). A higher P4 value (17.05±5.23ng/ml) was recorded at day (12) and a significantly lowest one (0.29±0.05ng/ml) was recorded at day (73). The ovulation rate was 72%, while the pregnancy rate was 40% in cows treated with GPG protocol. It is concluded that the hormones used in OvSynch protocol (GnRH, PGF₂α & GnRh), stimulated the ovarian activity of cows and can overcome summer infertility and the hormones used had no adverse effect on hematological parameters and/or metabolic status of treated animals.

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Keywords: GnRh, PGF₂α, OvSynch, Cows, Hormonal levels, Synchronization

1. Introduction

It was reported that a polypeptide namely, gonadotropin-releasing hormone from hypothalamus, was found to regulate the secretion of luteinizing and follicle stimulating hormones (Schally *et al.*, 1973).

In cycling cows, administration of GnRH or a derivative induces a gonadotropin surge (Chenault *et al.*, 2003) with peak LH within 2 to 3 h (Williams *et al.*, 1982) and alters the pattern of follicle growth (Thatcher *et al.*, 1989; Wolfenson *et al.*, 1994). Administration of GnRH induces a LH surge with similar maximum LH concentrations (McDougall *et al.*, 1995) but with approximately half the duration (Chenault *et al.*, 2003), when compared to the endogenous LH release during the normal estrous cycle at the time of ovulation (Rahe *et al.*, 1980; Chenault *et*

al., 2003). A single injection of GnRH or an agonist is sufficient to induce ovulation or atresia of a dominant follicle (Crowe *et al.*, 1993; Twagiramungu *et al.*, 1995). Several reports demonstrated that growing follicles greater than 10 mm in diameter ovulate after GnRH injection (Pursley *et al.*, 1997; Silcox *et al.*, 1995; Martinez *et al.*, 2000)

The Induction of ovulation of ovarian follicles was demonstrated in milked (Britt, *et al.*, 1974) and suckled cows (Schams *et al.*, 1977) following an injection of Gonadotropin-releasing hormone. GnRH induced effects is indirect (Chenault *et al.*, 2003) through their induced release of LH (Britt *et al.*, 1974) and follicle stimulating hormone (FSH) from anterior pituitary gland. Later, GnRH analogues and agonist

were developed, which were more potent than native GnRH (Thatcher *et al.*, 1989).

Synchronization of follicular waves and selection of new large follicle following GnRH at any stage of the estrous cycle was used as a tool to further develop estrous synchronization programs for fixed timed AI (Twagiramungu *et al.*, 1995).

To improve the estrus synchrony exogenous GnRH, which controls the developmental stage of the preovulatory follicle has been included with prostaglandin for synchronization of estrus in dairy cows (Stevenson and Pursley, 1994; Thatcher *et al.*, 2001). The random administration of GnRH during the estrous cycle results in LH release (Chenault *et al.*, 2003), causes ovulation or luteinization of large follicles present in the ovary, synchronizes the recruitment of a new follicular wave (Thatcher *et al.*, 1989; Martinez *et al.*, 2000), and equalizes follicle development waves (Twagiramungu *et al.*, 1995; Schmitt *et al.*, 1996). Subsequent administration of PGF₂ α induces the regression of an original or GnRH-induced CL, and allows final maturation of the synchronized dominant follicle (Schmitt *et al.*, 1996). Further, there is no apparent detrimental effect of GnRH on the responsiveness of GnRH-induced CL or spontaneous CL to prostaglandin (Twagiramungu *et al.*, 1995).

Recently, it is important that effective estrous synchronization protocols are developed in order to increase the use of A.I. In addition, estrous synchronization protocols should be designed to reduce time and labor inputs by limiting cattle handlings and reducing or eliminating estrus detection (Larson *et al.*, 2006).

The main purpose of this study was to study the pharmacodynamics of Receptal[®] (GnRH) and Estrumate[®] (PGF₂ α) injection on animal fertility, hematological and steroid hormone pattern and pregnancy rate in cows.

2. Material and Methods

The present study was conducted in private sector farms in Beni-Suef Governorate, Upper Egypt, during 2010 to August/2011.

1. Animals

This study was conducted on 15 crossbreed cows (Baladi \times Abundance). The age of the animals ranged from 3 to 8 years (4.84 \pm 1.91 years). These animals were reared under correct management system including feeding, housing, and veterinary medical care as well as recording system. The selected animals were free from any reproductive disorders. The body condition score (BCS) of these animals was recorded (2.7 \pm 0.57) and scaled according to Gordon (1996).

The general characteristics of the used animals and types of the applied protocols are presented in table (1)

Table 1: General reproductive and productive characteristics of cattle under experimentation.

VARIABLE		Cows
Breed		Crossbreed cows (Baladi \times Abundance)
Month of experiment		Summer (August)
Parity	Primiparous	9 (60%)
	Pleuriparous	6 (40%)
Average daily milk yield (kg)		3.5
(B.C.S.)		2.7 \pm 0.57
Interval from calving to 1 st estrus (days)		108 \pm 45.52
Age (year)		4.84 \pm 1.91

2. Chemicals

A. Estrumate[®] (Synthetic prostaglandin)

Each ml contains 263 μ g cloprostenol sodium (BP-vet.) equivalents to 250 μ g cloprostenol (Schering Plough, Essex Animal Health, and Germany).

B. Receptal[®] (Gonadotropin releasing hormone)

Each ml contains 0.0042 mg buserelin acetate equivalent to 0.004 mg buserelin, 10mg benzyl alcohol (Intervet International B.V. Boxmeer, Holland).

3. Semen: Bull semen no. 91, name Jiscar processed and packaged in mini straws (0.25 ml) at A.I. center, Beni-Seuf, Egypt.

Methods

1. Clinical examination

Ovarian findings: Clinical examination was performed according to Arthur *et al.* (1989). Rectal palpation was done for detection of the ovarian activities at the beginning of each protocol. Follicular consistency was examined rectally at the time of insemination and classified into turgid or fluctuating follicles.

Genital tract: Examination of the genital tract at the time of insemination was done as described by Arthur *et al.* (1989).

Estrus signs: Observations of the animals for signs of heat were done throughout the day from early morning to evening and classified into strong and weak estrous signs according to the intensity of nervous manifestation exhibited by cows at estrous.

2. Fertility indices

Rectal examination of the cows for pregnancy diagnosis was done 45-60 days and ultrasound examination at 30 days post insemination. The fertility indices were calculated as described by Grusenmeyer *et al.* (1992).

3. Blood sampling and serum preparation

Blood samples were collected from the jugular vein into two test tubes; one containing anticoagulant (2-3 drops Heparin) for hematological analysis and other test tube containing no anticoagulant for serum preparation for hormonal assay.

4. Hematological examination

The blood parameters were measured by automated Animal hematology analyzer (Animal Hematology analyzer, Model XF-9080).

5. OvSynch Protocols of synchronization of estrous (GPG)

Summary of the protocol procedure was carried out according to Mialot *et al.* (2003). On day (0) cows examined clinically per rectum, blood sampling and injection of 2.5ml Receptal® (GnRH) I/M. Seven days later, injection of 2ml Estrumate® (PGF₂α) I/M. After 48 hours, animals were injected I/M with 2nd dose (2.5ml) of GnRH. Timed artificial insemination (T.A.I.) 24 hours after the 2nd dose of GnRH. Pregnancy diagnosis by rectal palpation at day 73 of experiment for the inseminated cows (Fig.1).

Statistical analysis:

Probabilities of the different fertility levels were calculated as expansion of binomial distribution according to the following equation:(mean)

$$\bar{X} = \frac{\sum fx}{\sum f}, \quad S.D. = \sqrt{\frac{\sum fx^2}{n} - \bar{x}^2}$$

$$se = \frac{sd}{\sqrt{n}}$$

S.D= standard deviation; SE= standard error
n=number (Thirkettle, 1981).

The obtained probabilities were multiplied by 100% to obtain the probability %. Analysis of variance was done by calculation of the LSD using the PC-STAT (1985).

3. Results

1. Hematological Parameters.

The investigated blood parameters are illustrated in table (2).

Erythrocytic count: The results of the current study revealed that a significantly higher value (7.26±0.32x10¹²/L) was detected at day (7) in pregnant animals. Meanwhile, no significant differences in erythrocytic count were detected among other days of experiment in non pregnant animals.

Mean Cell Volume (MCV): A significantly higher value (48.75±0.09 FL) was recorded at day (7) in conceived (pregnant) animals compared with other days of experiment. It was also appeared that, in non pregnant animals a higher MCV value (46.42±0.67FL) was recorded at day 21 in comparison with that recorded at the rest of the experimental days.

Hematocrite value (HCT):The findings of the present study revealed no significant differences in HCT values among the different days of the experiment within the same group. However a significantly higher value (0.36±0.015 L/L) was detected at day 7 in pregnant cows compared with other values in non pregnant cows.

Hemoglobin (HB): A significantly lowest value (76.58±6.51 g/L) was recorded at day 31 of the experiment in pregnant cows compared with other days of experiment. On the other hand in non pregnant animals no clear significant differences in HB values were recorded among the different experimental days.

White Blood cells (WBCs) Parameters

WBCs count: The results of the current study revealed that, a significantly lowest value (6.45±0.58 x10⁹/L) was detected at day (10) compared with other days of the experiment in conceived /pregnant cows. Significantly higher values (10.36±0.61 and 9.39±0.54 x10⁹/L) were recorded at days (7 and 31) in pregnant cows compared with that (6.41±0.46 and 6.65±0.35 x10⁹/L) recorded at the same days in non pregnant cows. No significant differences were observed in WBCs count among different days of the experiment in non pregnant cows.

Lymphocyte percentages (LY %): In pregnant animal, our results revealed no significant differences among values of LY % at different days of experiment. Meanwhile in non pregnant animals, a significantly lower value (21.85±1.49) was recorded at day (10) compared with values that recorded at days 7 (36.19±1.46) and 31(36.7±1.41).

Granulocyte percentages (GR %): Our results proved that treatment had no significant effect on GR % at different days of experiment in pregnant animals. Meanwhile in non pregnant animals, a significantly highest value (72.25±1.61) was recorded at day 10 compared with that (55.80±1.58 and 56.99±1.47) recorded at days (7 and 31) respectively. Moreover no significant differences in GR % were detected between the pregnant and non pregnant cows.

Hormonal patterns :

Estradiol level

Although hormonal data presented in table (3) showed no significant ($P = 0.0295$) differences in estradiol level among the different experimental days, a high estradiol level (35.9±3.62 pg/ml) was recorded at day (10) at fixed TAI and lowest one (21.03±0.79 pg/ml) was detected at day (73).

Progesterone serum level

A highly significant ($P < 0.0001$) variation in (p4) levels among different days of the protocol was observed, with a higher value (17.05±5.23 ng/ml) was recorded at day (12). Meanwhile a significant lowest one (0.29±0.05ng/ml) was recorded at day (73).

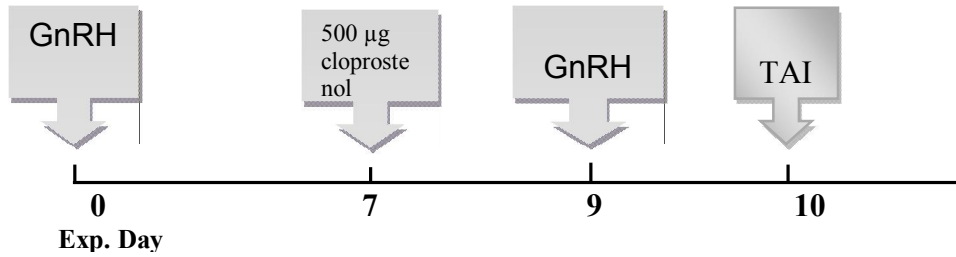


Table 2:- Some hematological parameters in cows subjected to Ovsynch protocol (GPG).

EXP. Day	Response to treatment	RBC $\times 10^{12}/L$ (Mean \pm SE)	MCV FL (Mean \pm SE)	HCT L/L (Mean \pm SE)	HB g/L (Mean \pm SE)	WBC $\times 10^9/L$ (Mean \pm SE)	LY% (Mean \pm SE)	GR% (Mean \pm SE)
0	Pregnant	6.25 \pm 0.31 ^a	42.6 \pm 1.82 ^a	0.28 \pm 0.021 ^{ab}	109.9 \pm 11.70 ^{bc}	10.316 \pm 0.72 ^b	26.51 \pm 2.90 ^{ab}	63.54 \pm 2.49 ^{abc}
7		7.27 \pm 0.32 ^b	48.75 \pm 0.09 ^b	0.36 \pm 0.015 ^b	125 \pm 12.41 ^{bc}	10.36 \pm 0.61 ^b	29.43 \pm 2.97 ^{bac}	64.13 \pm 3.17 ^{abc}
21		6.19 \pm 0.39 ^a	46.5 \pm 1.27 ^b	0.30 \pm 0.023 ^{ab}	127.62 \pm 14.64 ^c	6.46 \pm 0.58 ^a	29.27 \pm 1.58 ^{bac}	68.59 \pm 0.88 ^{cb}
31		6.92 \pm 0.24 ^a	43.08 \pm 0.90 ^c	0.30 \pm 0.013 ^{ab}	76.58 \pm 6.51 ^a	9.39 \pm 0.54 ^b	35.98 \pm 1.95 ^{cb}	57.95 \pm 2.06 ^{ab}
0	Non Pregnant	6.25 \pm 0.10 ^a	41.75 \pm 1.12 ^a	0.26 \pm 0.008 ^a	94.16 \pm 4.65 ^{ab}	8.85 \pm 0.81 ^{ab}	29.74 \pm 2.24 ^{bac}	64.03 \pm 2.72 ^{abc}
7		6.14 \pm 0.19 ^a	42.08 \pm 1.03 ^a	0.27 \pm 0.013 ^a	113.83 \pm 7.82 ^{bc}	6.41 \pm 0.46 ^a	36.19 \pm 1.46 ^{cb}	55.80 \pm 1.58 ^a
21		5.71 \pm 0.19 ^a	46.42 \pm 0.67 ^b	0.27 \pm 0.005 ^a	108.21 \pm 5.52 ^{bc}	6.77 \pm 0.40 ^a	21.85 \pm 1.49 ^a	72.25 \pm 1.61 ^c
31		6.013 \pm 0.33 ^a	44.61 \pm 0.99 ^{ba}	0.30 \pm 0.026 ^{ab}	93.5 \pm 8.55 ^{ab}	6.65 \pm 0.35 ^a	36.7 \pm 1.41 ^c	56.99 \pm 1.47 ^{ab}

SE = standard Error

Values (Means) in the same column with different letter are significantly different ($P < 0.05$).

0-day: 1st dose of GnRH; 7-day: PGF $_2\alpha$; 10-day: timed A.I.; 31-day:sampling.

Table 3: Serum hormonal concentration in cows subjected to Ovsynch – protocol (GPG)

Exp. Day	Treatment (activities)	n*(15)	Estradiol (pg/ml) means \pm SE	Progesterone (ng/ml) means \pm SE
0	GnRh (1 st dose)	15	31.07 \pm 3.79 ^a	1.86 \pm 0.44 ^a
4	Sampling		33.7 \pm 3.38 ^a	1.05 \pm 0.20 ^a
7	PGF $_2\alpha$		24.71 \pm 2.53 ^a	8.88 \pm 2.62 ^{ab}
9	GnRh 2 nd dose		-	-
10	TAI		35.9 \pm 3.62 ^a	1.17 \pm 0.29 ^a
12	Sampling		33.9 \pm 3.15 ^a	17.05 \pm 5.23 ^b
21	Sampling		32.58 \pm 4.82 ^a	2.7 \pm 0.53 ^a
31	Sampling		26.88 \pm 3.74 ^a	0.4 \pm 0.09 ^a
73	Pregnancy diagnosis		21.03 \pm 0.79 ^a	0.29 \pm 0.05 ^a

n=number of animals

SE = standard Error

Values (Means) in the same column with different alphabetical are significantly different ($P < 0.05$)

II. Main reproductive results of cows subjected to GPG (OvSynch protocol).

Table 4. Main reproductive results in cows subjected to OvSynch (GPG) protocol.

VARIABLE	Results	
Cyclicity before treatment n. (%)	35.75% normal cyclic	
	29.25% repeat breeder	
	35% non cyclic	
Ovulation rate	72%	
Rectal findings at time of A.I.	ovarian findings -follicular consistency	30% turgid follicles
		70% fluctuating follicles
	consistency of uterus	28% slightly tonic
		72% erected
Pregnancy rate % (n)	40% (6/15)	

4. Discussion

To synchronize the estrous cycle, ovarian activity is manipulated so that the time of ovulation can be predicted. This is achieved by (1) controlling the

luteal phase of the cycle through the administration of prostaglandins or progesterone analogues or (2) controlling follicle development and ovulation using different combinations of prostaglandins,

progesterone or gonadotrophin releasing hormone (GnRH).

Modern estrus synchronization protocols involve either lengthening or shortening the animal's estrous cycle to achieve synchrony. A variety of techniques are available for producers to utilize and all are based on several strategies of hormonal supplementation including progestin, PGF₂ α and gonadotropins (Odde and Holland, 1994; Ryan *et al.*, 1995).

The results of the present study revealed significant changes in some erythrocytic parameters including (RBCs count, MCV, HCT and HB) but these changes within the normal blood value of cattle according to Nemi (1986) and Victor *et al.* (2000).

The data of the current study revealed a significant increase in RBCs count and MCV, 7 days after injection of GnRh in group (2); however these elevations in RBCs and MCV within normal blood value range of cattle, meanwhile GnRh had no significant effect on HCT, HB, WBCs, lymphocyte and granulocyte. These finding might attributed to GnRh, where it had no significant effect on the metabolic and/or the healthy status of treated animals as reported by Victor *et al.* (2000).

Our results in cows revealed that the average animals that responded to the treatment was 72%, these finding were lower than those obtained by Pursley *et al.* (1997) whom found that the second GnRH injection induces ovulation in 87 to 100% of cows, which occurs 24 to 32 h after GnRH was administered.

The decrease in response to exogenous GnRH and PGF₂ was reported previously by many authors, Stevens *et al.* (1995) reported that administration of GnRH and prostaglandin simultaneously on Day 8 or 10 of estrous cycle does not improve the synchrony of estrus and ovulation (luteolysis in only 6 of 16 animals) because GnRH disrupts follicular dynamics and induces premature ovulation or delays the normal return to estrus. Birnie *et al.* (1997) treated heifers with GnRH injections every 24 or 48 h from Day 3 until Day 17 of estrous cycle and administered prostaglandin on Day 13 of estrous cycle to study the luteal response of GnRH treated animals to a physiological dose of prostaglandin. They observed the luteolytic activity by using ultrasonography only in seven of 16 animals in GnRH treated group. Birnie *et al.* (1997) reported that the luteolytic activity PGF₂ α analogue is reduced when it is administered in combination with the GnRH agonist. They reported that reduced luteolytic effect of prostaglandin when given simultaneously with GnRH may be due to a luteotropic protection of GnRH on the CL, thus preventing the usual cascade of oxytocin stimulation

and progesterone inhibition that occurs until completion of luteolysis. In cattle, administration of GnRH during the early or mid luteal phase causes an alteration of follicular distribution in the ovary by increasing the number of medium sized follicle and decreasing the number of large follicles by inducing luteinization and or atresia (McNatty *et al.*, 1981; Thatcher *et al.*, 1989; Guilbault, *et al.*, 1990). GnRH administered on Day 11 to 13 of estrous cycle alters the ovarian follicular dynamics (Skaggs *et al.*, 1986) since the dominant follicle either luteinizes (Thatcher *et al.*, 1989) or develops into a secondary CL following ovulation (Stevenson *et al.*, 1993). Wolfenson *et al.* (1994) studied the dynamics of follicular development by ultrasonography in cows following administration of a single dose of GnRH in the mid luteal phase (Day 12) of the estrous cycle. They reported the preovulatory follicles in cows following the injection of GnRH during the luteal phase were more homogeneous (belonging to the same follicular wave), more estrogen-active, probably due to preovulatory follicles being recruited and selected close to the time of estrus, and more dominant. GnRH induced ovulation or atresia of dominant follicle is followed by a new wave emergence within 3 to 4 d of treatment at any stage of estrous cycle (Twagiramungu *et al.*, 1995). Administration of GnRH induces a FSH increase at any stage of the estrous cycle (Ryan *et al.*, 1994). Thus, in cows treated with GnRH after the selection of a dominant follicle, gonadotropin surge is followed by a transient FSH increase, that is associated with the emergence of a new follicle wave. When GnRH treatment is applied before the selection of the dominant follicle, follicular growth is not affected (Ryan *et al.*, 1994).

In addition, the results of the current study revealed that overall pregnancy rate were 40%. These data are in agreement with the finding of Pursley *et al.* (1997 ; 1998), they found that pregnancy rates resulting from fixed time AI in lactating dairy cows vary from 32 to 45% following administration of Ovsynch.

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Effect of PGF₂α Double Injection on Hematological Picture, Hormonal Levels and Fertility of Cows During Summer Season

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Abstract: Heat stress has an adverse effect on animal fertility due to direct or indirect action on hypothalamic-hypophyseal axis and/or on gonadal hormones. The present study aimed to study the pharmacological action of prostaglandin F₂α injection on ovarian activity, blood picture, gonadal hormones and animal fertility during heat stress. Ten crossbred native cows (Baladi X Abundance), their ages 3-8 years, not showing estrus signs for a mean of 95.28±46.18 days (summer infertility) were treated with a regimen consisted of one or double doses of PGF₂α (2.5 ml Estrumate[®]/animal), the second dose was injected after 9- 11 days for cows not exhibited estrus post 1st dose. The blood samples were from jugular vein, before, during and after treatment. Complete blood picture, Serum progesterone and estradiol-17β were estimated. The response to treatments were determined using rectal palpation and ultrasonography (US). The results revealed that there is no significant differences in RBCs count at different days of the experiment in non pregnant cows, whereas, a significantly lower value (6.11±0.40 vs. 7.38±0.33) was recorded at day 7 in pregnant cows. **Mean Cell Volume (MCV)**, in pregnant cows was averaged (49.91 ±2.27 and 49.5 ± 1.99, *p*<0.05) at days 21 and 31, respectively, whereas, in non pregnant cows, a significantly higher value (60.5±1.45) was recorded only at day (7). **Hematocrite value (HCT)**, were significantly different (*p*<0.05) between pregnant and non pregnant animals. It recorded higher values (*p*<0.05) at days 7&31 in non pregnant animals than that reordered at any other day of the experiment in both pregnant and non pregnant cows. **Hemoglobin (HB):** A significantly lower HB value (104±3.58) was recorded at day (7) of pregnant cows, while the highest value (156.83±13.67) was detected at day 21. Concerning the non pregnant cows, a significantly highest HB value (157.25±8.27) was observed at day 7. However significant differences in HB values were recorded among pregnant and non pregnant animals at days 7, 21, and 31 of the experiment. **WBCs count:** In pregnant cows, a significantly higher WBCs count was recorded at day 0 (12.72±1.19) compared with values that recorded at the other days of the treatment or in non pregnant cows. **Granulocyte percentages (GR %):** A significantly higher values (63.17±0.19, 61.83±1.24) were detected at day 7 in pregnant and non pregnant cows, respectively. A significantly (*p*<0.001) highest level of estradiol was recorded at day 22 (36.64±4.26 pg/ml) and significantly lower one was measured at day 10 (18.61±0.23 pg/ml), whereas, Progesterone (P₄) was significantly decreased at day 10 (1.7±0.14/ml ng). The reproductive results in cows treated with double injection of PGF₂α indicated that the peak of response of cows ranged from 72 hrs to 96 hrs post inj. of 1st dose of PGF₂α, it also appeared the pregnancy rate was 70% (7/10). It is concluded that PGF₂α double injection (PG-PG) was effective for resuming animal fertility and ovarian activity during summer season without adverse effect on hematological parameters and/or metabolic status of treated animals.

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1. Introduction

In the early 1970s several workers pioneered the luteolytic effect of prostaglandin F₂α (PGF₂α) in cattle (Rowson *et al.*, 1972). Subsequent research efforts then attempted to improve the reproductive efficiency of dairy cattle by inducing estrus with PGF₂α (Seguin *et al.*, 1978; Plunkett *et al.*, 1984). Several studies demonstrated the capacity of PGF₂α

and its synthetic analogues, alfaprostol (Randel *et al.*, 1988; Tolleson and Randel, 1988; Randel *et al.*, 1996), cloprostenol (Cooper, 1974; Cooper and Rowson, 1975), fenprostalene (Martinez and Thibier, 1984; Stotts *et al.*, 1987), luprostioli (Plata *et al.*, 1989; Plata *et al.*, 1990) and tiaprost (Schams and Karg, 1982) to trigger the regression of a mature CL in the ovary, thus provoking and synchronizing estrus

(Stevenson *et al.*, 1989; Stevenson and Pursley, 1994). When PGF₂α was administered to cows with a functionally mature CL, 85 to 95% reached estrus within 7 d of treatment (Armstrong *et al.*, 1989; Folman *et al.*, 1990; Rosenberg *et al.*, 1990); 70 to 90% showing signs of estrus 3 to 5 d after treatment (Ferguson and Galligan, 1993).

Prostaglandin treatment in the early stage of estrous cycle (first 5 d) was found to be ineffective in causing a luteolytic response in cattle (Cooper and Rowson, 1975; Lauderdale, 1975). Consequently, a double protocol in which PGF₂α was given at a 7, 11 or 14 d interval was developed so that cows at a stage in the estrous cycle other than diestrus would have a functional CL when they received the second PGF₂α dose (Rosenberg *et al.*, 1990; Kristula *et al.*, 1992). However, several authors report the improved reproductive efficiency of cows detected to be in estrus after the second PGF₂α dose using the double regime in which PGF₂α doses are given 11 or 14 d apart (Ferguson and Galligan, 1993; Stevenson *et al.*, 2000). Further, an enhanced estrus response and normal fertility were reported when PGF₂α was given at the late, rather than early to middle stage of the luteal phase (Watts and Fuquay, 1985; Xu *et al.*, 1997). Thus, the 14 d interval double prostaglandin regimen seems to show an improved response over the 11 d protocol, since two treatments given 14 d apart ensures that most animals are in the late luteal stage (cycle Day 11 to 14) when they receive the second PGF₂α dose (Folman *et al.*, 1990; Rosenberg *et al.*, 1990).

Many researchers have noted normal or above normal fertility following synchronization of estrus with PGF₂α in cows (Lucy *et al.*, 1986; Wenzel, 1991). However, reduced conception rates due to variations in the time of ovulation have been noted after timed AI, either following single (Fetrow and Blanchard, 1987; Archbald *et al.*, 1992) or double (Waters and Ball, 1978; Stevenson *et al.*, 1987) prostaglandin administration, compared to AI at detected estrus. Reproductive performance in dairy cattle was also improved following double 14 d-PGF₂α treatment without assessing ovarian status when compared to a single dose based on detecting a CL by rectal palpation or by milk progesterone enzyme immunoassay (Heuwieser *et al.*, 1997).

Several studies have reported that the stage of estrous cycle at the time of prostaglandin greatly influences the conception rate in dairy cows. Armstrong (1988) reported that the conception rate among the cows treated on Day 13 (71 %) was significantly higher when compared to the cows treated on Day 8 (46 %). Larson and Ball (1992) reported that higher progesterone concentrations at the time of PGF₂α administration are associated with

delayed onset of estrus. It has also been reported that estrus was manifested in more percentage of cows (84 %) that had high progesterone concentrations, > 3.1 ng/mL, at the day of the last PGF₂α injection than did cows with low progesterone levels (56 %). Folman *et al.* (1990) found that cows conceiving to AI at induced estrus had higher progesterone levels during the preceding luteal phase than those not conceiving. However, Gyawu *et al.* (1991) showed that excessively long periods of high progesterone prior to insemination can suppress fertility. Therefore, the aim of the present work was to study the pharmacological action of double doses of PGF₂α on hemogram, ovarian response and some hormonal changes of mixed breed of cows.

2. Material and Methods

The present study was conducted in private sector farm in Beni-Suef Governorate, Upper Egypt, from 2010 to 2011.

1. Animals

This study was conducted on 10 crossbreed cows (Baladi×Abundance). The age of the animals ranged from 3 to 8 years and other parameters are tabulated in table(1). These animals were reared under correct management system including feeding, housing, and veterinary medical care as well as recording system. The selected animals were free from any reproductive disorders. The body condition score (BCS) of these animals was recorded and scaled according to **Gordon (1996)**.

Table 1: General reproductive and productive characteristics of cattle under experimentation.

VARIABLE		
Breed		Crossbreed cows (Baladi×Abundance)
Month of experiment		Summer (August)
Parity	Primiparous	3 (30%)
	Pleuriparous	7 (70%)
Average daily milk yield (kg)		3.5
(B.C.S.)		2.9±0.55
Interval from calving to 1 st estrus (days)		95.28±46.18
Age (year)		5.35±2.17

2. Chemicals

Estrumate® (Synthetic prostaglandin)

Each ml contains 263 µg cloprostenol sodium (**BP-vet.**) equivalents to 250 µg cloprostenol (**Schering Plough, Essex Animal Health, and Germany**).

3.Semen: Bull semen no. 91, name Jiscar processed and packaged in mini straws (0.25 ml) at A.I. center , Beni-Seuf , Egypt.

Methods

1. Clinical examination

Ovarian findings: Clinical examination was performed according to **Arthur et al.(1982)**. Rectal palpation was done for detection of the ovarian activities at the beginning of each protocol. Follicular consistency was examined rectally at the time of insemination and classified into turgid or fluctuating follicles.

Genital tract: Examination of the genital tract at the time of insemination was done as described by **Arthur et al.(1989)**.

Estrus signs: Observations of the animals for signs of heat were done throughout the day from early morning to evening as reported by **Boriek (2002)** and classified into strong and weak estrous signs according to the intensity of nervous manifestation exhibited by cows at estrous.

2.Fertility indices

Rectal examination of the cows for pregnancy diagnosis was done 45-60 days and ultrasound examination at 30 days post insemination. The fertility indices were calculated as described by **Grusenmeyer et al. (1992)**.

3.Blood sampling and serum preparation

Blood samples were collected from the jugular vein into two test tubes; one containing anticoagulant (2-3 drops Heparin) for hematological analysis and other test tube containing no anticoagulant for serum preparation for hormonal assay.

A-Hematological examination

The blood parameters were measured by automated Animal hematology analyzer (Animal Hematology analyzer, Model XF-9080).

B- Hormonal assay

The concentrations of Estradiol (E₃) and Progesterone (P₄) in serum were measured by Radioimmunoassay technique (RIA) using the diagnostic commercial kit (Estradiol Coat-A-Count kit) and (Progesterone Coat-A-Count RIA kit) respectively from Siemens Medical Solutions Diagnosis, USA according to **Hiroshi et al.(2008)**, then read by γ - Counter (Berthold, Model, MAG 312, Lab of Pharmacology, NRC).

4. Protocol of synchronization for estrous :Double injection of PGF_{2a}.

A summary of the experimental procedure was carried out according to **Stevenson and Phatak(2005)**.

At Day (0): all cows examined clinically per rectum, blood sampling and applying of the 1st dose of 2.5 ml Estrumate[®] (I/M). Insemination

of cows which detected in heat (2 to 5 days after the 1st dose of Estrumate[®]). At day (10) of experiment, injection of 2.5 ml Estrumate[®] (2nd dose) for the not inseminated cows. Two days after 2nd dose heat detection (HD) and insemination (AI) of cows were done. Pregnancy diagnosis by rectal palpation at day 72 of experiment for the inseminated cows. Blood sampling was performed at days 0, 4, 7, 10, 22, 32 and 72 for measuring some hematological parameters and hormones.

Statistical analysis:

Probabilities of the different fertility levels were calculated as expansion of binomial distribution according to the following equation:(mean)

$$\bar{X} = \frac{\sum fx}{\sum f} \quad , \quad S.D. = \sqrt{\frac{\sum fx^2}{n} - \bar{x}^2}$$

$$se = \frac{sd}{\sqrt{n}}$$

S.D= standard deviation se= standard error
n=number (**Thirkettle, 1981**).

The obtained probabilities were multiplied by 100% to obtain the probability %. Analysis of variance was done by calculation of the LSD using the **PC-STAT (1985)**.

3. Results

I. **Hematological Parameters.** The investigated blood parameters are presented in table (2).

Red Blood Cells (RBCs) Parameters

Erythrocytes count: Our results revealed no significant differences in RBCs count at different days of the experiment in non pregnant cows. Concerning the pregnant cows, a significantly lower value (6.11±0.40) was recorded at day 7 compared with that recorded at day 0 (7.38±0.33).

Mean Cell Volume (MCV):In pregnant cows, a significantly higher values (49.91 ±2.27) and (49.5 ± 1.99) were recorded at days 21 and 31 respectively compared with values at day 0 (44.46±1.78) and day 7 (41.5±7.07). In non pregnant cows, a significantly higher value (60.5±1.45) was recorded at day (7) compared with those recorded at any other day of the protocol.

Hematocrite value (HCT): Regarding the pregnant animals, a significantly lowest value (0.25±0.003) was detected at day (7) compared with that recorded at other days of the experiment. Meanwhile a significantly higher values (0.38±0.032) and (0.38±0.016) were recorded at days 21and 31, respectively. Moreover there were significant differences in (HCT) values between pregnant and non pregnant animals. In non pregnant animals the (HCT) values at days 7 and 31 were significantly

higher than that reordered at any other day of the experiment in both pregnant and non pregnant cows.

Hemoglobin (HB):In pregnant cows, a significantly lower HB value (104 ± 3.58) was recorded at day (7), while the highest value (156.83 ± 13.67) was detected at day 21. Concerning the non pregnant cows, a significantly highest HB value (157.25 ± 8.27) was observed at day 7. However significant differences in HB values were recorded among pregnant and non pregnant animals at days 7, 21, and 31 of the experiment.

White Blood Cells (WBCs) Parameters

WBCs count: In pregnant cows, a significantly higher WBCs count was recorded at day 0 (12.72 ± 1.19) compared with values that recorded at

the other days of the treatment. Regarding the non pregnant cows no significant differences in WBCs count were observed among the different experimental days.

Lymphocyte percentages (LY %): The results indicated that no clear significant differences among values at different days of experiment in both pregnant and non pregnant animals.

Granulocyte percentages (GR %):In pregnant cows, a significantly higher value (63.17 ± 0.19) was detected at day 7 compared with those recorded at the other days of the experiment. While in non pregnant cows a significantly highest value (61.83 ± 1.24) was recorded at day (7).

Table 2: Some hematological parameters in cows subjected to PGF₂α double injection regimen.

EXP. Day	Response to treat.	RBC $\times 10^{12}/L$ (Mean \pm SE)	MCV FL (Mean \pm SE)	HCT L/L (Mean \pm SE)	HB g/L (Mean \pm SE)	WBC $\times 10^9/L$ (Mean \pm SE)	LY% (Mean \pm SE)	GR% (Mean \pm SE)
0	Pregnant	7.38 ± 0.33^{ba}	44.46 ± 1.78^a	0.33 ± 0.025^b	133.8 ± 10.69^{bc}	12.72 ± 1.19^b	32.4 ± 3.29^a	61.58 ± 3.03^c
7		6.11 ± 0.40^a	41.5 ± 2.24^a	0.25 ± 0.003^a	104 ± 3.58^a	6.59 ± 0.47^a	40.6 ± 5.05^{ba}	63.18 ± 0.19^{cd}
21		7.67 ± 0.51^b	49.91 ± 2.27^b	0.38 ± 0.032^c	156.83 ± 13.67^d	9.12 ± 1.35^a	49.61 ± 19.29^b	50.9 ± 1.64^b
31		7.64 ± 0.29^a	49.5 ± 1.99^b	0.38 ± 0.016^c	118.5 ± 7.24^{ab}	8.05 ± 0.70^a	39.17 ± 3.67^{ba}	42.9 ± 1.82^a
0	Non Pregnant	6.45 ± 0.15^{ba}	46.5 ± 0.99^a	0.31 ± 0.013^b	117.16 ± 5.84^{ab}	8.83 ± 0.41^a	31.8 ± 1.33^a	61.83 ± 1.24^c
7		7.495 ± 0.13^{ba}	60.5 ± 1.45^c	0.46 ± 0.044^d	157.25 ± 8.27^d	9.06 ± 0.13^a	46.22 ± 2.05^{ba}	74.85 ± 1.34^d
21		6.35 ± 0.04^{ba}	47.5 ± 1.12^b	0.37 ± 0.025^c	141 ± 2.68^c	8.87 ± 0.83^a	45.9 ± 6.93^{ba}	57.17 ± 2.62^{cb}
31		7.39 ± 0.37^{ba}	51.16 ± 1.46^b	0.47 ± 0.041^d	149.66 ± 13.76^c	9.63 ± 0.71^a	32.93 ± 2.41^{ba}	61.53 ± 2.89^{cb}

SE = Standard Error

Values in the same column with different letter are significantly different ($P < 0.05$)

0-day: 1st dose of PGF₂ &; 7-day: sampling; 21-day: sampling; 31-day: sampling

Table (3): Serum hormonal levels in cows assigned to PGF₂α double inj. Protocol.

Exp. Day	Treatment (activities)	n* (10)	Estradiol (pg/ml) Mean \pm SE**	Progesterone (ng/ml) Mean \pm SE**
0	PGF ₂ α 1 st dose + sampling	10	29.7 ± 3.67^{ab}	2.06 ± 0.46^b
3	Heat detection + Insemination	5	-	-
4	Sampling	10	26.76 ± 3.47^{ab}	3.24 ± 0.89^b
5	Heat detection + Insemination	2	-	-
7	Sampling	10	18.72 ± 0.40^a	2.3 ± 0.58^b
10	Pgf ₂ α 2 nd dose + sampling	3	18.61 ± 0.23^a	1.7 ± 0.14^a
12	Heat detection + Insemination	3	-	-
22	Sampling	10	36.64 ± 4.26^b	2.10 ± 0.88^b
32	Sampling	10	19.40 ± 0.98^a	3.83 ± 1.01^b
72	Pregnancy diag. + sampling	10	19.78 ± 1.33^a	2.27 ± 0.15^b

n=number of animals

SE = standard Error

Values in the same column with different letter are significantly different ($P < 0.05$)

Table 4: Response and fertility results of cows subjected to PGF₂α double inj. protocol

VARIABLE	Results	
Cyclicity before treatment n. (%)	39.6% normal cyclic	
	20.4% repeat breeder	
	40% non cyclic	
Distribution of estrus RESPECTIVELY onset in cows treated with PGF ₂ α inj.	Post 1 st dose	20% after 120hrs (2/10) 50% after 72hrs (5/10)
	Post 2 nd dose	20% after 48hrs 2/10
Rectal findings at time of A.I.	ovarian findings	20% turgid follicles
	consistency	80% fluctuating follicles
	consistency of uterus	20% slightly tonic

Pregnancy rate % (n)	70% (7/10)	80% erected
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4. Discussion

To synchronize the estrous cycle, ovarian activity is manipulated so that the time of ovulation can be predicted. This is achieved by (1) controlling the luteal phase of the cycle through the administration of prostaglandins or progesterone analogues or (2) controlling follicle development and ovulation using different combinations of prostaglandins, progesterone or gonadotrophin releasing hormone (GnRH).

Modern estrus synchronization protocols involve either lengthening or shortening the animal's estrous cycle to achieve synchrony. A variety of techniques are available for producers to utilize and all are based on several strategies of hormonal supplementation including progestin, PGF₂α and gonadotropins (Odde and Holland, 1994; Ryan *et al.*, 1995).

The results of the present study revealed significant changes in some erythrocytic parameters including (RBCs count, MCV, HCT and HB) but these changes within the normal blood value of cattle according to Nemi (1986) and Victor *et al.* (2000).

Many protocols involving exogenous administration of PGF₂α have been developed and the normal treatment regimen for PGF₂α consists of two injections spaced 10 to 14 d apart (MARK, 2002).

The results of the current study revealed that, cows synchronized with PGF₂α varied in response to treatment according to stage of cycle at the beginning of experiment as reported by King *et al.* (1982); Tanabe and Hann (1984).who reported that, The main factors affecting synchrony when using PGF₂α treatment was the stage of the estrous cycle and the sensitivity of the CL to PGF₂α administration, it was greatest on d 10 of cycle, also MARK (2002) added the theory behind this treatment is at least one injection will be administered during the middle stage of the estrous cycle, and in theory, all cattle should be responsive to PGF₂α at this time. When cattle were injected with PGF₂α during d 5-16 of the estrous cycle a return to estrus was observed within 2 to 4 d.

Our results reveal that the peak of response of cows after 1st injection ranged from 2-5 days, where 70% of responsive animals exhibited estrus within that period and the rest of responsive animals come in heat 48hrs post 2nd dose of PGF₂α. These findings may be attributed to the type of follicular structure present at time of injection, such findings coincides with that obtained by De Rensis and Lopez (2007), who recorded that the effect of PGF₂α

administration is very similar to that observed in cattle and the time interval between PGF₂α treatment and the onset of estrus in buffaloes varies according to the stage of follicular development at the time of PGF₂α administration. Animals treated when follicles are in the pre-dominance stage (DF) of development display estrus 4-6 days later, whereas animals treated in the presence of a DF display estrus 23 days after PGF₂α administration. Thus, since the intervals between treatment, estrus, and ovulation vary after PGF₂α administration, a timed artificial insemination protocol cannot be applied. The author of this latter study reported that the mean interval from PGF₂α to estrus was 88hrs (range 48-144hrs), PGF₂α induced estrus and ovulation in about 60-80% of treated animals.

The results of our study revealed that 10% of treated animals not respond for the treatment, these results can be explained in the light of published report by Kojima *et al.* (2000) who indicated that, cattle that failed to respond to the 2nd PGF₂α injection had either a cystic follicle or early developing CL.

Our results revealed overall pregnancy rates was 70%, these results are higher than those found by De Rensis and Lopez (2007) who indicated that, pregnancy rates following prostaglandin treatment are 45-50% on average and appear to be similar to those obtained after natural estrus.

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Studying the Effect of rhBAFF & BAFF-R-Fc Fusion Protein on Lymphocytes & Platelets in Children with ITP

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Abstract: Introduction: B cell activating factor, a member of tumor necrosis factor family, is a crucial homeostatic cytokine for B cells. It has been shown to enhance the expression of CD19⁺ cells and mediate the maturation of autoreactive B cells. BAFF is elevated in several autoimmune diseases including immune thrombocytopenic purpura (ITP). Increased survival of CD8⁺ T cells also may promote the apoptosis of platelets through cytotoxic T lymphocyte-mediated platelet lysis. Blockade of BAFF receptor has demonstrated a clinical benefit in immunologic diseases. **Methods** PBMCs and platelets from 15 acute ITP patients and 15 healthy controls were cultured with rhBAFF or a combination of rhBAFF and BR3-Fc and then analyzed by flow cytometry for apoptosis of autologous platelets and/or CD19⁺, CD8⁺ and CD4⁺ cells. **Results** Blockade of BAFF receptor by BR3-Fc significantly increased the apoptosis of CD19⁺ cells in patients only and decreased the apoptosis of platelets in both patients and controls. Apoptotic CD8⁺ cells were significantly increased in patients only, following the addition of BR3-Fc. **Conclusion** These findings suggest that blockade of BAFF receptor (BR3-FC) could successfully correct the effects of BAFF by promoting the apoptosis of CD19⁺ and CD8⁺ cells and decreasing the apoptosis of platelets. Further research will indicate whether blocking BAFF-BR3 will have a therapeutic applicability in the management of ITP or not.

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1. Introduction

Immune thrombocytopenia (ITP) is an autoimmune disorder in which the patient's immune system is activated by platelet autoantigens resulting in immune-mediated platelet destruction and/or suppression of platelet production (1). Autoantibodies against platelet antigens are considered the diagnostic hallmark of ITP (2). The autoantibodies produced by autoreactive B cells against self-antigens, specifically immunoglobulin G (IgG) antibodies against glycoprotein IIb (GPIIb)/IIIa and/or GPIb/IX, are considered to play a crucial role (3). Opsonization by antibody not only accelerates platelet clearance but can also alter platelet function and interfere with platelet production (4). Although in most patients ITP is antibody mediated, the autoantibodies are under the control of T helper cells (Th1) and their cytokines. Abnormal T-cell responses drive the differentiation of autoreactive B-cell clones and autoantibody secretion (5). It is well known that decreased apoptosis of activated T lymphocytes play an important role in the pathogenesis of ITP (6). It was suggested that CD8⁺ cytotoxic T cells might be involved in the pathogenesis of ITP through cell-mediated destruction of platelets (7), and through suppression of megakaryocyte apoptosis leading to impaired platelet production (8).

B-cell activating factor (BAFF) was reported to belong to the tumor necrosis factor (TNF) family, and is thought to be critical for the maintenance of normal B-cell development, homeostasis, and autoreactivity (9) and T-cell costimulation (10,11). In addition, BAFF also augments certain Th1-associated inflammatory responses (12). BAFF is the natural ligand of three unusual tumor necrosis factor receptors termed BCMA, TACI and BR3/BAFF-R (13,14). BR3, identified as the crucial receptor for B-cell survival, is expressed on a wide range of B-cell subsets, including immature, transitional, mature, memory, and germinal center B cells, as well as on plasma cells (15). Signaling through BR3/BAFF-R and BCMA stimulates B lymphocytes to undergo proliferation and to counter their self destruction by apoptosis. Furthermore, BAFF binding to BR3 on T cells has been shown to costimulate T-cell proliferation both *in vitro* and *in vivo* (11).

Several lines of evidence suggested that BAFF may play an important role in autoimmunity. Autoantigen-binding B cells may have an increased dependence on the BAFF survival signal (16). In addition, elevated BAFF plasma level was observed in many patients with autoimmune diseases such as rheumatoid arthritis (RA) (17), systemic lupus erythematosus (SLE) (18), Sjögren syndrome (SS) (19), and multiple sclerosis (20). Inhibition of BAFF

signaling is a potentially therapeutic option for treatment of B cell-mediated autoimmune conditions.

The aim of the present study is to investigate the effect of rhBAFF and BR3-FC fusion protein on PBMCs and platelets from ITP patients as well as from normal controls, in a trial to elucidate the role of BAFF in the pathogenesis of ITP.

2. Patients and Methods:

Study population:

Fifteen newly diagnosed cases of acute childhood ITP (disease duration from 0-3 months) were selected from the pediatric wards of New Children's Cairo University Hospital during the time period from Sept 2011 to January 2012. They were diagnosed according to The British Committee for Standards in Hematology (21) and included 6 females and 9 males with a median age, 6 years and platelet count range; $9-30 \times 10^9/L$ with a median count, $18 \times 10^9/L$. Fifteen healthy children matched for age and sex were included in the study as a control group and included 7 females and 8 males, with a median age 6 years and platelet count range; $160-430 \times 10^9/L$, with a median count; $200 \times 10^9/L$. The control subjects were healthy children who presented to Kasr Al-Ainy hospital laboratory for routine checkup. Written informed consents were obtained from the parents of participating children before enrollment.

Exclusion criteria included the following:

Splenomegaly; lymphadenopathy; abnormalities of CBC other than isolated thrombocytopenia with or without mild microcytic hypochromic anemia due to bleeding; abnormal bone marrow picture other than increased and/or defective megakaryocytes; evidence of SLE; recent immunization; drug intake (heparin, sulphonamides, alcohol, quinine) and features of congenital syndromes e.g. absent radii, skeletal anomalies, ocular anomalies or hearing loss.

Preparation of PBMCs:

PBMCs were isolated from 2 ml sample of heparinized blood using 1.077 g/mL of Ficoll-Hypaque (Invitrogen, USA) by density gradient centrifugation method (2000 rpm for 20 minutes, $20^\circ C$) as described by Al-Janadi *et al.*, 1993 (22). The isolated PBMCs were then washed twice using sterile phosphate buffered saline then resuspended in 1ml RPMI 1640 and adjusted to 10^6 PBMCs/mL for cell culture.

Preparation of autologous platelets:

Autologous platelets were separated from 5–10 ml sample of heparinized blood, at day 1, by centrifugation at 600rpm for 10 minutes at $20^\circ C$. The platelet-rich plasma was separated and acid citrate dextrose (ACD) was added at a ratio one ACD to 9 PRP then recentrifuged at 2500 rpm for 15 minutes and the platelet pellet was washed once using a

prepared solution consisting of 0.5 ml ACD and 3 ml PBS and then resuspended in 1 ml RPMI (23).

The effect of rhBAFF and/or BR3-Fc on apoptosis of PBMCs (CD 19+, CD 4+ and CD 8+ cells) by flow cytometry:

PBMCs with autologous platelets were cultured at a ratio 1:10 in RPMI 1640 medium, supplemented with 10% FBS and penicillin-streptomycin 10000 IU/mL, in culture tubes with rhBAFF (R&D systems, USA) alone or with a combination of rhBAFF and BR3-Fc (R&D systems, USA) at $37^\circ C$ for 24 hrs. Culture tubes were prepared as follows:

Tube 1: contained PBMCs with autologous platelets cultured with 2 μL of rhBAFF, in RPMI medium supplemented with 100 μL FBS, giving a total volume of 1 ml in the culture tube.

Tube 2: contained PBMCs with autologous platelets cultured with 2 μL of rhBAFF and 10 μL of BR3-Fc, in RPMI medium supplemented with 100 μL FBS, giving a total volume of 1 ml in the culture tube. PBMCs were harvested after 24 hrs. For each tube to be analyzed; 3 tubes were prepared, where 100 μL of PBMCs were incubated with either: 20 μL of phycoerythrin (PE)-cyanin 5 (Cy5)-conjugated CD19, 20 μL of PE conjugated CD8 or 20 μL of PE-Cy 5-conjugated CD4 (Beckman Coulter Company, USA) for 30 minutes. Cells were then washed with PBS and incubated with 5 μL of fluorescein isothiocyanate-conjugated annexin 5 (Beckman Coulter Company, USA) for 15 mins. Cells were analyzed within 1 hr by fluorescence-activated cell sorting using Beckman Coulter (Coulter Epics XL).

The effect of rhBAFF and/or BR3-Fc on apoptosis of autologous platelets by flow cytometry:

PBMCs alone were cultured in RPMI 1640 medium supplemented with 10% FBS and penicillin-streptomycin 10000 IU/mL, in culture tubes with rhBAFF alone or with a combination of rhBAFF and BR3-Fc at $37^\circ C$ for 24 hrs. By the second day, autologous platelets were separated as described previously and incubated with the cultured PBMCs for 2 hrs. Culture tubes were prepared as follows:

Tube 3: contained PBMCs with autologous platelets cultured with 2 μL of rhBAFF, in RPMI medium supplemented with 100 μL FBS, giving a total volume of 1 ml in the culture tube.

Tube 4: contained PBMCs with autologous platelets cultured with 2 μL of rhBAFF and 10 μL of BR3-Fc, in RPMI medium supplemented with 100 μL FBS, giving a total volume of 1 ml in the culture tube.

Two hrs after addition of platelets, platelets were harvested. For each tube to be analyzed, 1 tube was prepared where 100 μL of platelets were incubated with 20 μL of PE-Cy5-conjugated CD41 (Beckman Coulter Company) for 30 mins. Cells were then washed with PBS and incubated with 5 μL of

fluorescein isothiocyanate-conjugated annexin 5 for 15 mins. Cells were analyzed within 1 hr by fluorescence activated cell sorting. Flow cytometry was adjusted with linear amplification of the forward and side scatter signals. Correlated data of 10,000 events per sample were acquired using dual expression with a live gate applied on CD8+/annexin+ve, CD4+/annexin+ve, CD19+/annexin+ve and CD41+/annexin+ve cells. Two-parameter histograms demonstrating cell staining were created and quadrant statistics were placed on the basis of cell staining.

Interpretation of results:

Results were expressed as a percentage of annexin V (apoptotic) cells with labelled monoclonal antibodies.

Statistical analysis

Descriptive data were expressed as mean plus or minus SD. Paired t-test was used in comparison of means for paired samples while the student's t-test was used in comparison of means for 2 independent groups (cases and controls). Statistical significance was determined by analysis of variance. All tests were performed by SPSS 13.0 system. A *P* value less than 0.05 was considered statistically significant.

3. Results:

The differences in the mean percent expression of annexin V positive T, B lymphocytes and platelets following incubation with rhBAFF / rhBAFF and BR3-Fc between ITP patients and normal healthy controls were statistically evaluated (**Table 1**).

Comparison between the percent expression of annexin V positive T, B lymphocytes and platelets before and after addition of BR3-Fc in both ITP patients and control group are shown in **Table 2**

Effects of rhBAFF and/or BR3-Fc on apoptosis of peripheral CD19⁺, CD4⁺, and CD8⁺ cells

After culture of PBMCs with rhBAFF, the following findings were detected:

The mean percent expression of annexin V positive CD4⁺ cells was significantly lower in the patients' group than the control group (*P* value <0.033). Addition of BR3-Fc in both patients and control groups lead to an increase in the annexin V positive CD4⁺ cells, however this increase was not statistically significant (**Figure 1**).

The mean percent expression of annexin V positive CD8⁺ cells was significantly lower in the patients' group than the control group (*P* value <0.001). There was a highly significant statistical increase in annexin V positive CD8⁺ T cells following addition of BR3-Fc in patients only (*P* <0.001), whereas no statistically significant difference was detected in the control group on adding BR3-Fc (**Figure 2**).

The mean percent expression of annexin V positive CD19⁺ cells was also lower in the patients' group than the control group but this decrease was not statistically significant and expression was increased after adding BR3-Fc, which caused a highly significant increase in annexin V positive CD19⁺ B cells in patients only (*P* <0.001) (**Figure 3**).

Effects of rhBAFF and/or BR3-Fc on apoptosis of autologous platelets

Apoptosis of platelets on addition of rhBAFF and rhBAFF with BR3-Fc in the control group was significantly higher than apoptosis of platelets in patients' group (*P* =0.006 & *P* =0.003) respectively. There was a significant statistical decrease in annexin V positive platelets following the addition of BR3-Fc in both patients and controls, this decrease was highly significant in the patient group (*P* <0.001) and it was significant in the control group (*P* <0.003) as seen in (**Figure 4**).

Figure 5 represents the apoptosis of platelets in 2 typical ITP patients where rhBAFF increased apoptosis of platelets in ITP patients while BR3-Fc corrected the effect of rhBAFF.

Table (1): Comparison between ITP patients and controls, regarding apoptosis (annexin V expression) on lymphocyte subsets and platelets

	Patients (n= 15)	Control (n= 15)	<i>P</i>	
CD4 rhBAFF	9.27 ± 3.77	11.85 ± 2.26	0.033	S
CD4 rhBAFF+BR-FC3	9.68 ± 4.13	13.03 ± 3.45	0.023	S
CD8 rhBAFF	8.06 ± 3.38	20.78 ± 10.2	<0.001	HS
CD8 rhBAFF+BR-FC3	11.24 ± 3.54	20.5 ± 9.04	0.002	S
CD19 rhBAFF	12.83 ± 4.13	16.96 ± 13.23	0.266	NS
CD19 rhBAFF+BR-FC3	16.25 ± 5.73	12.58 ± 11.76	0.290	NS
CD41 rhBAFF	11.96 ± 4.33	17.83 ± 6.26	0.006	S
CD41 rhBAFF+BR-FC3	8.69 ± 3.9	13.98 ± 4.95	0.003	S

*Statistical analysis was done using student t test

Table (2): Comparison between ITP patients and controls regarding apoptotic (annexin V) expression of lymphocyte subsets and platelets, before and after adding BR3-Fc

	CD4 rhBAFF	CD4 rhBAFF+BR3-Fc	P	
Patient (n=15)	9.27 ± 3.77	9.68 ± 4.13	0.389	NS
Control (n=15)	11.85 ± 2.26	13.03 ± 3.45	0.126	NS
	CD8 rhBAFF	CD8 rhBAFF+BR3-Fc	P	
Patient (n=15)	8.06 ± 3.38	11.24 ± 3.54	<0.001	HS
Control (n=15)	20.78 ± 10.2	20.5 ± 9.04	0.809	NS
	CD19 rhBAFF	CD19 rhBAFF+BR3-Fc	P	
Patient (n=15)	12.83 ± 4.13	16.25 ± 5.73	<0.001	HS
Control (n=15)	16.96 ± 13.23	12.58 ± 11.76	0.168	NS
	CD41 rhBAFF	CD41 rhBAFF+BR3-Fc	P	
Patient (n=15)	11.96 ± 4.33	8.69 ± 3.9	<0.001	HS
Control (n=15)	17.83 ± 6.26	13.98 ± 4.95	0.003	S

*Statistical analysis was done using paired t test

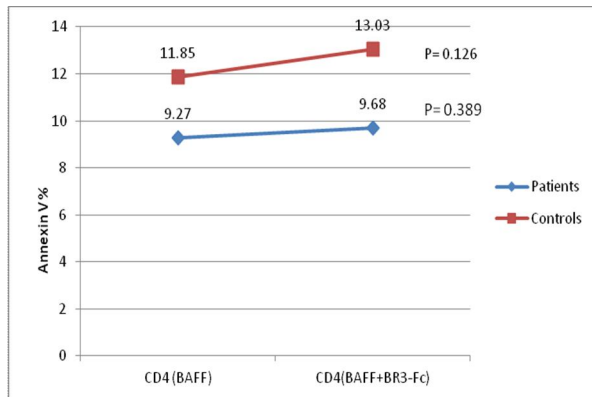


Figure (1): Annexin V positive CD4+ cell changes in patients and controls

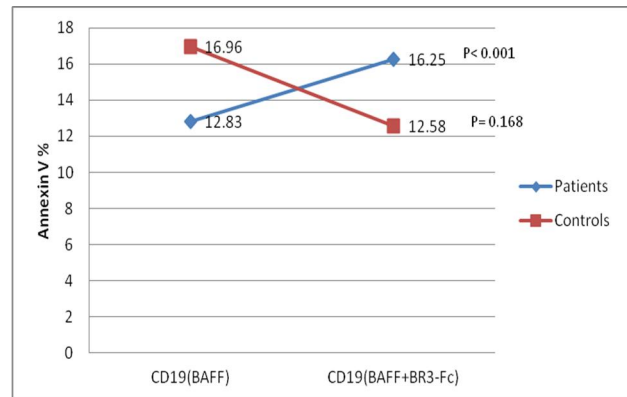


Figure (3): Annexin V positive CD19+ cell changes in patients and controls

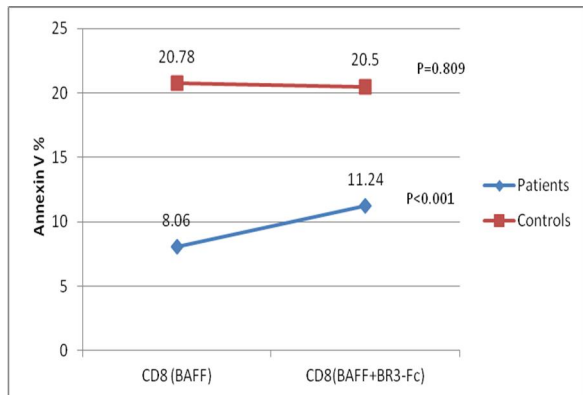


Figure (2): Annexin V positive CD8+ cell changes in patients and controls

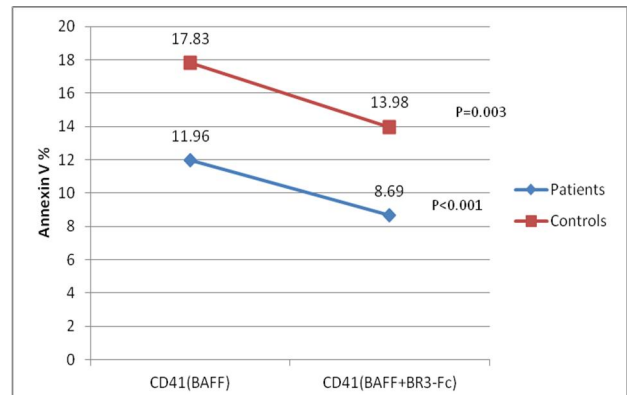
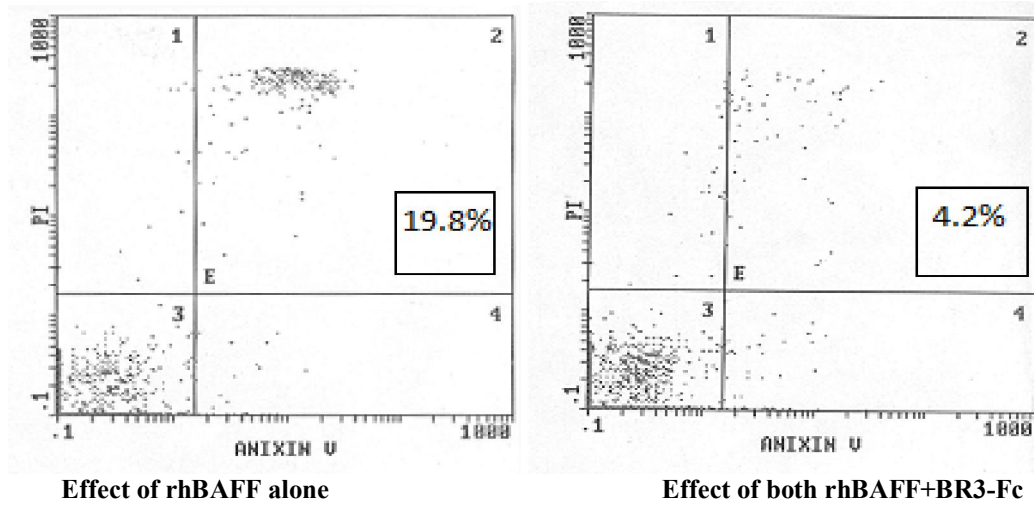


Figure (4): Platelet changes in patients and controls

Patient A :



Patient B :

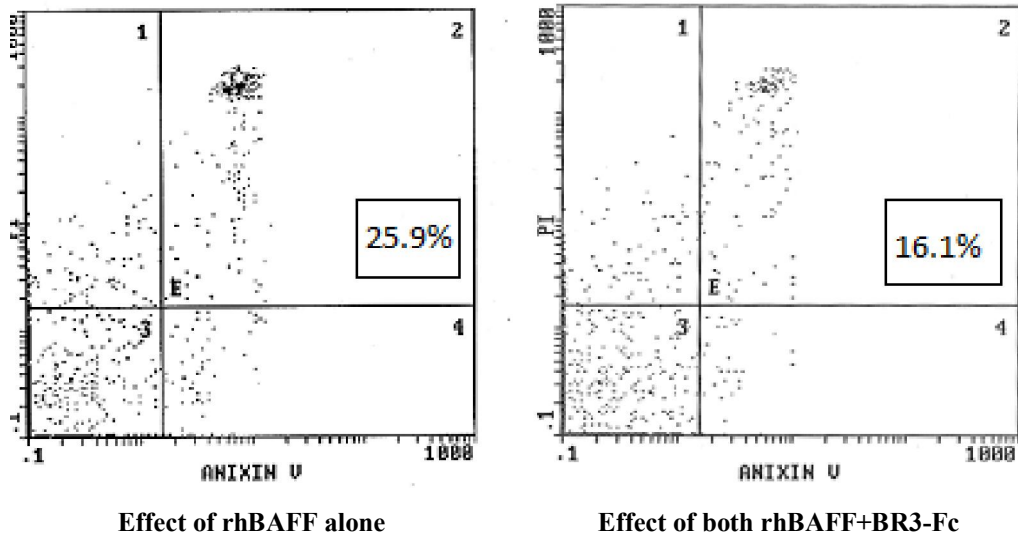


Figure (5): Flow-cytometric dot plots representing the effects of rhBAFF and BR3-Fc on apoptosis of platelets in 2 ITP patients (A & B) where rhBAFF promoted the apoptosis of platelets and BR3-Fc corrected the effects of rhBAFF.

4. Discussion:

ITP is an autoimmune disease manifested by immune-mediated platelet destruction and suppression of platelet production. Although several abnormalities involving the cellular mechanisms of immune modulation have been identified, development of autoantibodies against platelet glycoproteins remains central in the pathogenesis of ITP (4).

BAFF is a crucial homeostatic cytokine for B cells that is up-regulated during inflammation and links adaptive with innate immunity. BAFF has been shown to enhance the expression of CD19 cells and mediate the maturation of autoreactive B cells (24).

BAFF exerts its role through binding to its receptors. BAFF-R, a specific receptor for BAFF, is expressed on a wide range of B cell subsets and is the predominant receptor that mediates BAFF-dependent B cell signaling and plays critical roles in controlling peripheral B cell survival (25). Signaling through BR3/BAFF-R stimulates B lymphocytes to undergo proliferation and to counter their self destruction by apoptosis. Furthermore, BAFF binding to BR3 on T cells has been shown to costimulate T-cell proliferation both *in vitro* and *in vivo* (11).

BR3-Fc, a fully human fusion protein of the extracellular domain of human BAFF-R with the Fc of human IgG1, is a selective BAFF blockade that

could block the interaction of BAFF with all 3 of its receptors (26).

In a study by *Zhu et al., 2009*, the level of plasma BAFF and BAFF mRNA in ITP patients with active disease was found to be significantly higher than that in patients in remission and controls (26). It was also noted that high-dose dexamethasone (HD-DXM) has shown its clinical efficacy in ITP patients (27).

Wang et al., 2010 investigated the change of BAFF and regulatory T-cells before and after high-dose dexamethasone therapy and assessed the effect of BAFF on Treg cells in ITP. They concluded that HD-DXM might play a role in ITP treatment by down-regulating BAFF expression and up-regulating Treg cells number (28).

In the present study, we tried to elucidate the effect of rhBAFF on PBMNCs and platelets, and the effect of blocking BAFF-R on the survival of these cells. Fifteen patients diagnosed as acute ITP were selected for this study in addition to fifteen healthy subjects as a control group. Basic investigations for diagnosis of ITP were performed including complete blood picture and bone marrow aspiration. Culture of isolated lymphocytes and platelets was then done with rhBAFF or rhBAFF and BR3-Fc, and analysis of apoptotic cells was performed using dual expression flowcytometry.

After culture of PBMCs with rhBAFF, we found that the mean percent expression of annexin V positive CD4+ and CD8+ cells were significantly lower in the patients' group than the control group (P value = 0.033, P value = <0.001) respectively. The mean percent expression of annexin V positive CD19+ cells was also lower in the patients' group than the control group but this decrease was not significant statistically. In a study by *ZHU et al., 2009* they demonstrated that rhBAFF significantly decreased the annexin V percentage of CD19+ cells in ITP patients but not in controls, while it significantly decreased the annexin V percentage of CD8+ cells in both ITP patients and controls (ITP: 6.5%±3.2%, controls: 10.5%±2.7%, P <0.05). On the other hand they found no significant effect of rhBAFF on annexin V percentage of CD4+ cells in both ITP patients and controls (P <0.05) (26).

In this study, we demonstrated that the decrease in the apoptotic expression of different lymphocyte subsets from ITP cases was corrected after adding BR3-Fc, which caused an increase in annexin V positive CD4+ cells, however this increase was not statistically significant. On the other hand, there was a highly significant statistical increase in annexin V positive CD8+ T cells and CD19+ B cells following addition of BR3-Fc (P <0.001 in both). Our results were in agreement with *ZHU et al., 2009* study where

BR3-FC corrected the decrease in annexin V percentage of CD19+ cells caused by rhBAFF in 14 ITP patients. It also corrected the effect of rhBAFF on apoptosis of CD8+ cells only in ITP patients, while they stated that there was no significant effect for rhBAFF+ BR3-FC on annexin V percentage of CD4+ cells in both ITP patients and controls (26).

Concerning the annexin V percentage of CD41+ platelets, we detected a significant statistical decrease in annexin V positive platelets following the addition of BR3-Fc in both patients' group and control group, this decrease was highly significant in the patient's group and was moderately significant in the control group. *ZHU and his co-workers, 2009* demonstrated that rhBAFF significantly increased apoptosis of platelets in ITP patients but not in controls. Addition of BR3-Fc corrected the effect of rhBAFF on apoptosis of platelets. Further study showed that the apoptosis of platelets increased in ITP patients when autologous platelets were incubated with PBMNCs from the same patient after rhBAFF was added whereas the apoptosis of platelets did not increase without PBMNCs, indicating that BAFF may contribute to thrombocytopenia partially by cell-dependent platelet destruction in ITP (26).

Our study demonstrated that BR3-Fc could significantly promote the apoptosis of CD19+ cells in ITP and block the BAFF-mediated survival of B cells. In addition to promoting the apoptosis of CD8+ cells, BR3-Fc also inhibited the apoptosis of platelets. BAFF blockade could result in B-cell reduction in animal models (29,30). Recent clinical trials with BAFF blockade have shown clinical benefit in SLE and RA (31,32). These findings offer further support to the contention that blockade of BAFF signaling may be of therapeutic benefit in a variety of autoimmune diseases.

In summary, BAFF is elevated in ITP patients with active disease, and excessive BAFF may rescue autoreactive B and T cells from apoptosis. Increased survival of CD8+ T cells may promote the apoptosis of platelets through CTL-mediated platelet lysis. BR3-Fc, a selective BAFF blockade, could successfully correct the effects of rhBAFF by promoting the apoptosis of CD19+ and CD8+ cells and inhibiting apoptosis of platelets. Blockade of BAFF by BR3-Fc could be a promising therapeutic approach for ITP.

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Analysis of Genetic signature for some *Plectropomus* species based on some dominant DNA markersY. M. Saad^{1,2}; AbuZinadah, O. A. H.¹; El-Domyati, F. M.^{1,3} and Sabir, J. M.¹¹Dept. of Biol. Science, Fac. of Sciences, King Abdulaziz Univ., KSA.² Genetic Lab., National Institute of Oceanography and Fisheries (NIOF), Egypt.³ Dept. of Genetics, Faculty of Agric., Ain Shams Univ., Shobra El-Kheima, Egypt.yasser_saad19@yahoo.com

Abstract: Successful conservation of fish such as *Plectropomus* species depend on analysis of genetic signature and relations among fish species and subspecies. Knowledge about the genetic signature and structure of *Plectropomus* species needs to increase several times to enable us for conserve these economic fish in the future. The current study aims to develop and analysis of genetic signature for three *Plectropomus* species (*P.maculatus*, *P.leopardus* and *P.areolatus*) based on some dominant DNA markers (RAPD and ISSR). A total of 9 ISSR and 12 RAPD primers were used to develop some DNA markers and their loci were estimated. Most studied DNA markers were polymorphic. The overall gene diversity (h) based on RAPD data was higher than based on ISSR data. Data analysis showed that, the *P.areolatus* was distantly related from both *P.maculatus* and *P.leopardus*. RAPD and ISSR markers proved to be powerful methods to detect genetic signature for the applied *Plectropomus* species. The genetic markers which developed in this study will play a valuable role in monitoring the *Plectropomus* genetic resources in the future.

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Key words: *Plectropomus*, DNA polymorphism, RAPD and ISSR markers.

1. Introduction

Overfishing of *Plectropomus* species as high value marine food in the Red sea (Zhu *et al.*, 2005) makes them difficult to manage (FAO 2009).

Developing of genetic signature of such species will facilitate the analysis of fish genetic structure for management of such aquatic genetic resources in the future. In addition, the effective management of fisheries requires the ability to discriminate among fish stocks (Rico *et al.*, 1997) and/or species (Madhan *et al.*, 2011 and Rashed *et al.*, 2011).

Up to date, the application of DNA-based genetic analysis in *Plectropomus* species research, stock development and management is still not fully maximized. In addition, the level of speciation and evolutionary relationships between *Plectropomus* species remains unclear.

RAPD (Rashed *et al.*, 2011), SSR (Rashed *et al.*, 2009) and ISSR (Raina, *et al.* 2001) techniques should find major population genetics applications, notably in the field of genetic conservation, where molecular markers need to be developed at a reasonable cost.

The information revealed from the analysis of genetic polymorphism for fish species (Rashed *et al.*, 2011) and subspecies (Rashed *et al.*, 2008 and Saad *et al.*, 2009) is widely used in the management of aquatic genetic resources because management of aquatic genetic resources should ideally involve a continuum of activities (Eknath, 1994). These activities are documentation of genetic resources and the variety of

ecosystems; characterization to determine the genetic structure, evaluation to estimate economic potential and utilization in sustainable breeding schemes.

The current study aims to develop and analysis of genetic signature for some of *Plectropomus* species (*P.maculatus*, *P.leopardus* and *P.areolatus*) based on some dominant DNA markers (RAPD and ISSR).

2. Material and Methods**Fish Material:**

The fish samples (*Plectropomus maculatus*, *Plectropomus leopardus* and *Plectropomus areolatus*) were obtained from MBSPR project (funded from DSR, King Abdulaziz Univ., KSA during year of (2012). Jeddah province is the main source of these fish samples. A total of 20 individuals from each studied fish species were analyzed.

DNA extraction:

Small pieces of caudal fins (0.1g) were stored in 95% ethyl alcohol immediately. DNA was extracted and performed according to Hillis *et al.* (1996).

Genetic signature based on RAPD (Random Amplified Polymorphic DNA) markers:

Twelve RAPD primers (Operon Technologies) were tested for detection of applied fish genetic signature. The primer codes and sequences were presented in Table (1). PCR mixture, reaction conditions and product separation were carried out as described by Rashed *et al.* (2008) with some

modifications. Initial denaturation was for 4 min at 94°C; followed by 30 cycles of 40 s at 94°C, 40 s at 37°C, and 1 min at 72°C; with a final 15 min extension at 72°C.

Genetic signature based on ISSR (Inter Simple Sequence Repeat) markers:

A total of nine ISSR primers (Table 1) were originally tested and selected to develop genetic signature for the three applied fish genomes. PCR

mixture, reaction conditions and product separation were carried out as described by Raina, *et*

al. (2001) with minor modifications. The amplification was carried out in a 10 µl reaction volume containing 10 mM Tris-HCl (pH 9.0), 50 mM KCl, 1.5 mM MgCl₂, 0.1 mM each dNTP, 200 µM primer, 1 U Taq DNA polymerase, and 5 ng of genomic DNA. Initial denaturation was for 2 min at 94°C; followed by 30 cycles of 30 s at 94°C, 45 s at 44°C, and 2 min at 72°C; with a final 15 min extension at 72°C.

The amplification products in both cases (RAPD and ISSR) were size-separated by standard horizontal electrophoresis in 1.5% agarose (Sigma) gels and stained with ethidium bromide.

Table (1): Codes and sequences of RAPD and ISSR primers used in the study.

RAPD code	Sequence	ISSR code	Sequence
A01	5' CAG GCC CTT C 3'	7	5' [CT] ₈ RG 3'
A02	5' TGC CGA GCT G 3'	814	5' [CT] ₈ TC 3'
A03	5' AGT CAG CCA C 3'	844	5' [CT] ₈ RC 3'
B01	5' GTT TCG CTC C 3'	898	5' [CA] ₆ RY 3'
B02	5' TGA TCC CTG G 3'	899	5' [AG] ₇ YC 3'
B03	5' CAT CCC CCT G 3'	JOHN	5' [AG] ₇ YC 3'
C01	5' TTC GAG CCA G 3'	DAT	5' [GA] ₇ RG 3'
C02	5' GTG AGG CGT C 3'	TERRY	5' [GTG] ₄ RC 3'
C03	5' GGG GGT CTT T 3'	MAO	5' [CTC] ₅ RC 3'
D01	5' ACC GCG AAG G 3'		
D02	5' GGA CCC AAC C 3'		
D03	5' GTC GCC GTC A 3'		

Data analysis:

All gels were analyzed using Total Lab program V2.01. These data were analyzed as diploid data for Dominant markers by standard POPGENE program (version 1.32), for population genetic analysis (Yeh and Boyle,1997).

3.Results

I-Analysis of genetic signature based on RAPD markers:

The number of detected fragments (ranged from 6 to 19 bands), number of polymorphic fragments (ranged from 6 to 19 bands) and ranges of fragment sizes generated by each primer were scored (Table 2).

1-Genetic variations within each *Plectropomus* species:

Most of detected RAPD bands were polymorphic. The percentages of polymorphic fragments were 61.01%, 37.74% and 11.95% in *P. maculatus*, *P. leopardus* and *P. areolatus* respectively (Table 3).

Our results presented 23 species-specific RAPD markers. Four of them were detected in *Plectropomus maculatus* using primers B02, (321bp and 85bp), C01 (442bp) and D01 (229bp). Regarding *Plectropomus leopardus*, five specific RAPD markers were identified at molecular sizes 410 bp (primer A02), 194 bp (primer B03), 996 bp & 296 bp (primer

C01) and 221bp (primer D02). 14 RAPD markers were specific to *Plectropomus areolatus* at molecular sizes 610 bp, 404 bp, 200 bp (primer A01), 829 bp, 283 bp (primer A02), 430 bp, 232 bp (primer A03), 352 bp, 266 bp, 186 bp, 108bp (primer B01), 977bp, 106bp (primer B02) and 211bp (primer C01).

The mean of Actual number of alleles (na), Effective number of alleles (ne), Nei's gene diversity (h) and Shannon's Information index (I) within each applied fish species were calculated (Table 3). *Plectropomus maculatus* had the highest values for the previous estimates.

2-Genetic distance among applied *Plectropomus* species:

The genetic distance values among the applied fish species were calculated. The lengths among applied fish species, and nodes on the phylogenetic tree were presented in Table (4). The genetic distance values were 0.1685, 0.4265 and 0.399 between (*P. maculatus* and *P. leopardus*), (*P. maculatus* and *P. areolatus*) and (*P. leopardus* and *P. areolatus*) fish pairs, respectively (Table5). The dendrogram revealed genetic relationships among the applied fish species based on RAPD polymorphism was presented in Figure (1-a).

II- Analysis of genetic signature based on ISSR markers:

Generally, all studied loci were polymorphic. The molecular weight of each detected band was estimated. The ranges of these molecular sizes were presented in Table (Table 2).

All the nine studied loci, were informative in detecting the genetic signature of applied fish species (Table 2).

1-Genetic variations within applied fish species:

The actual number of alleles (na), effective number of alleles (ne) and Shannon's information index (I) were calculated within each estimated fish species and for all studied fish species for all primers. The mean of actual number of alleles (na), Effective number of alleles (ne), Nei's gene diversity (h) and Shannon's Information index (I) values were presented in Table (3). *P. areolatus* had the highest ne (1.146), h (0.087) and I (0.131) values relatively.

Table (2): Locus code, detected bands, polymorphic alleles (bands), and range of fragment generated by RAPD and ISSR markers.

RAPD				ISSR			
Code	DB	PB	R FS (Bb)	Code	DB	PB	R FS (Bp)
A01	17	16	924-122	7	32	32	1315-99
A02	15	15	859-114	814	23	23	1038-79
A03	14	14	775-114	844	17	17	570-120
B01	16	16	1005-108	JOHN	40	40	909-75
B02	9	9	977-106	899	12	12	785-111
B03	10	10	989-850	DAT	7	7	761-208
C01	12	12	1012-211	MANNY	40	40	874-67
C02	6	6	302-99	TERRY	27	27	524-99
C03	10	10	1100-234	MAO	25	25	855-109
D01	17	17	1356-135				
D02	19	19	656-155				
D03	14	14	1353-108				

DB= number of detected bands, PB= polymorphic bands and R FS= range of fragment size.

Table (3): Mean \pm SE of Actual number of alleles (na), Effective number of alleles (ne), Nei's gene diversity (h), Shannon's information index (I) and percentage of polymorphic loci (%PL) for each studied fish species.

	RAPD				ISSR			
	P.m	P.l	P.a	Total	P.m	P.l	P.a	Total
na	1.61	1.37	1.119	1.981	1.313	1.336	1.251	1.995
	± 0.48	± 0.486	± 0.322	± 0.136	± 0.46	± 0.47	± 0.4346	± 0.067
ne	1.35	1.23	1.085	1.46	1.127	1.128	1.146	1.302
	± 0.40	± 0.370	± 0.25	± 0.33	± 0.26	± 0.25	± 0.293	± 0.318
h	0.20	0.133	0.047	0.28	0.07	0.08	0.087	0.19
	± 0.20	± 0.197	± 0.135	± 0.164	± 0.14	± 0.14	± 0.16	± 0.16
I	0.302	0.198	0.0691	0.433	0.124	0.129	0.131	0.31
	± 0.29	± 0.28	± 0.193	± 0.216	± 0.217	± 0.214	± 0.237	± 0.219
%PL	61.01 %	37.74 %	11.95 %	98.11%	31.39 %	33.63 %	25.11 %	99.55

P.m=*P. maculatus*, P.l=*P. leopardus*. and P.a=*P. areolatus*

2-Genetic distance among applied fish species:

The genetic distance values among the applied fish species were calculated. The lengths among applied fish species, and nodes on the phylogenetic tree were presented in Table (4).

The genetic distance values were 0.1461, 0.3007 and 0.3729 between (*P. maculatus* and *P. leopardus*),

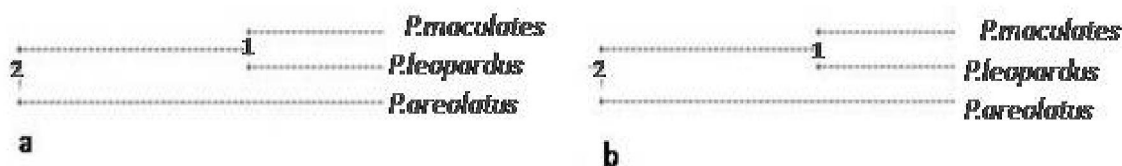
(*P. maculatus* and *P. areolatus*) and (*P. leopardus* and *P. areolatus*) fish pairs, respectively (Table 5). The dendrogram revealed genetic relationships among the applied fish species based on ISSR polymorphism was presented in Figure (1-b).

Table (4): The lengths among applied fish species and nodes on the phylogenetic tree (based on RAPD and ISSR)

Based on RAPD			Based on ISSR		
Between	And	Length	Between	And	Length
2	1	12.31	1	1	12.21
1	<i>P. maculatus</i>	7.94	<i>P. maculatus</i>	<i>P. maculatus</i>	8.42
1	<i>P. leopardus</i>	7.94	<i>P. leopardus</i>	<i>P. leopardus</i>	8.42
2	<i>P. areolatus</i>	20.25	<i>P. areolatus</i>	<i>P. areolatus</i>	20.62

Table (5): Genetic distance based on ISSR (lower half) and RAPD markers (upper half) among the three *Plectropomus* species

	<i>P.maculatus</i>	<i>P.leopardus</i>	<i>P.areolatus</i>
<i>P.maculatus</i>		0.1685	0.4265
<i>P.leopardus</i>	0.1461		0.3990
<i>P.areolatus</i>	0.3007	0.3729	

**Figure (1):** Reconstruction of phylogenetic relationships among the applied fish species based on RAPD (a) and ISSR (b) polymorphism.

4. Discussion

The capacity to conserve and to use natural resources wisely requires identifying taxa and variation at both the individual and/or population levels (Rashed *et al.*, 2008 and Saad *et al.*, 2011).

Conservation or good management of aquatic genetic resources should ideally involve a continuum of activities. These activities are documentation, characterization, evaluation and utilization of aquatic genetic resources (Eknath, 1994).

In the present study, three *Plectropomus* species (*P. maculatus*, *P. leopardus* and *P. areolatus*) were identified using two molecular techniques (RAPD and ISSR) to detect genetic signature of these economic fish. RAPD and ISSR were chosen because they are cheaper, simple, and fast techniques for detecting genetic polymorphism at a molecular level. In addition, just only one primer could obtain the different profiles for genomic analysis (Rina *et al.*, 2001 and Antunes, *et al.*, 2010).

In the present study, most detected DNA markers were polymorphic reflecting a rich allelic diversity in the applied fish species. So the primers of these loci are recommended to detect the genetic polymorphism and inferring the genetic signature for the applied *Plectropomus* species.

A comparison of RAPD and ISSR patterns in *Plectropomus* species samples, ISSR primers generate higher polymorphism (99.55%) than those generated by RAPD primers (98.11%). It could provide simple and convenient method to discriminate genetic variation of *Plectropomus* species.

It is necessary to estimate intra and inter population variations and phylogenetic relationships among fish genomes (Saad *et al.*, 2011) to help the breeder in designing suitable breeding programs for fish improving and/or conservation. In addition, this will be useful in detecting any genetic contamination in these fish genomes.

In the present study the *P. areolatus* was distantly related from both *P. maculatus*, *P. leopardus*. The genetic distance values among the applied fish species were calculated for determining genetic dissimilarity among them. Determining true genetic dissimilarity between fish individuals based on molecular marker analysis is a decisive point to cluster and analyzing diversity within and among fish species because different dissimilarity indices may yield conflicting outcomes.

Our results presented some species-specific RAPD and ISSR markers. These markers could be used in different two ways. In the first way these

markers will be used, as a species genetic signature. Regarding the second way, these markers are useful as marker assisted selection (Rashed *et al.*, 2009) in breeding (to develop local fish breeds) and restocking programs.

The species-specific PCR method could be potentially used by regulatory agencies as routine control assay for the commercial fish production.

PCR-based methods commonly used for fish species identification include PCR-sequencing, random amplified polymorphic DNA (RAPD), inter simple sequence repeat (ISSR). Those are simple, specific and sensate methods for genetic characterization (Asensio *et al.*, 2009).

Restocking and stock enhancement programs are now recognized as an important tool for the management of fishery resources. It is important, however, to have an adequate knowledge on the genetic population structure of both the released stock and the wild population before carrying out such programs (Pereira *et al.*, 2010). So, the genetic markers should be conducted to provide the information needed for a sound management of this an economic aquatic resources in wild fish stocks and/or farms (Saad *et al.*, 2011). In addition, development of local *Plectropomus* species breeds is important because many of them will be resilient to climatic stress and represent a unique source of genes for improving *Plectropomus* species production and conservation in the future.

Detection and analysis of genetic signature for *Plectropomus* species is an important for studying the fish genetic fragmentation. Fish fragmentation into subpopulations revealing groups that is genetically different from main source.

To date, very few studies have reported and/or used Nei's gene diversity and Shannon's based on RAPD and ISSR markers in *Plectropomus* species in the Red sea. The Nei's gene diversity (h) and Shannon's information index (I) estimates were calculated in the present study to detect genetic diversity within applied fish species. They were slightly different. Based on RAPD results, both of them were higher than in the case of results revealed from ISSR. Shannon's information index has general applications in ecology and is relatively insensitive to the skewing effects caused by the inability to detect heterozygous loci (Dawson *et al.*, 1995). To avoid the different approaches for its calculation we adopted the approach employed by Yeh and Boyle (1997).

Our results will be useful in studying the *Plectropomus* sub-species genetic structure because the genetic structure of *Plectropomus* sub-species and populations are not fully maximized and unclear. Generally, genetic diversity which is required for populations to be more adaptive with the environmental changes can be measured using an array

of molecular methods (Rashed *et al.*, 2008). The present study showed that, both RAPD and ISSR primers were informative in detecting species specific DNA markers.

Conclusion:

In our study, RAPD and ISSR markers proved to be powerful methods for the detection of genetic signature for *Plectropomus* species. The future of *Plectropomus* species conservation will rely on development of sound management techniques such as RAPD and ISSR to characterize and monitor *Plectropomus* genetic resources. This will increase our knowledge of the genetic characteristics of each fish genetic resource and to examine the effects of management practices on the gene pools of these fish species.

The detected of genetic signature using RAPD and ISSR techniques will assist in the definition of appropriate units for fish conservation thus providing a restricted focus for good management of *Plectropomus* genetic resources.

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The analysis of the neural electrophysiological examination on therapeutic effect of the complete carpal tunnel & palmar aponeurosis release(CTPAR) of carpal tunnel syndrome

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Abstract: Objective: To compare the therapeutic effect of the complete carpal tunnel & palmar aponeurosis release(CTPAR) of different severity carpal tunnel syndrome (CTS) patients using the neural electrophysiological examination before and 3 months after the surgery. Method: The median nerve sensory nerve conduction velocity (SCV), latency of motor nerve terminal (M-lat), sensory nerve active potential (SNAP) and the compound muscle action potentials of the opponens pollicis (compound muscle action potential, CMAP) from three groups of different lesion levels of 87 patients were measured before and 3 months after the CTPAR. The results were analyzed with SPSS 17.0. Result: In the mild CTS patients, the differences of the SCV and SNAP before and after 3 months of CTPAR were statistically significant ($P < 0.05$), while the differences of the M-lat and CMAP displayed no statistical significance ($P > 0.05$); In the moderate CTS patients, the SCV, M-lat, SNAP and CMAP improved significantly 3 months after the CTPAR, and the differences were statistically significant ($P < 0.01$); In the severe CTS patients, the differences of the SCV, M-lat, SNAP and CMAP before and after the CTPAR had no statistical significance ($P > 0.05$). Conclusion: The recoveries of the mild and moderate groups of CTS patients were very well after the CTPAR, while the recoveries of the severe patients were not ideal. As a result, if CTS patients are mild or moderate ones, and the effects of the conservative treatment are not ideal, the operation treatment should be performed as soon as possible to avoid sensory and motor dysfunction, which will affect the quality of life.

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Keywords: Carpal tunnel syndrome; Neural electrophysiological examination; Therapeutic effect

1. Introduction

Carpal tunnel syndrome(CTS) is one of the most common peripheral entrapment neuropathies[1], and the symptoms and signs are caused by an entrapment of the median nerve in the carpal tunnel at the wrist, which may be due to acute or chronic injury. It affects mainly middle aged men or women, often bilateral, the most common causes are tenosynovitis, synovitis and diabetes mellitus[2]. The early symptoms of CTS are the dull, tingling and numbness feeling of the thumb, index finger and middle finger, which often influence patient's life and work, so its early diagnosis and treatment is of great importance.

In our study, CTS patients were divided into three groups, mild, moderate and severe, according to the grading standard published by the United States of America electrophysiological diagnosis Association[3,4], and Combining with the clinical manifestation and the electrophysiological results[5-7]. The study was designed to observe the changes of SCV, M-lat, SNAP and CMAP before and after the complete carpal tunnel & palmar aponeurosis release

(CTPAR) [8] in CTS patients with different extent of lesions, and also to testify the great importance of CTS's early diagnosis and treatment.

2. Material and Methods

2.1 Subject

From March 2010 to March 2012, CTS patients who had a CTPAR in the department of surgery of the First Affiliated Hospital of Zhengzhou University were registered. A total of 87 individuals of both sexes with CTS voluntarily participated in the study. Informed consents were obtained from all participants. Inclusion criteria: CTS patients. Exclusion criteria: 1. Numbness and pain in the hand caused by other diseases, such as diabetic peripheral neuropathy, alcoholic peripheral neuropathy, uremic peripheral neuropathy and other peripheral neuropathies. 2. Central nervous system diseases. According to the grading standard published by the United States of America electrophysiological diagnosis Association, the clinical manifestation and the electrophysiological results, 87 CTS patients were divided into three groups, mild(20 patients), moderate (26 patients) and severe(41 patients). The grading

standard was listed as below(1)mild: The median nerve sensory nerve conduction velocity from digitus medius to wrist slowed down(<44 m / s),the M -lat of the median nerve from wrist to the midpiece of thenar eminence was normal,(2)moderate:The median nerve sensory nerve conduction velocity from digitus medius to wrist slowed down(<44 m / s),the M -lat of the median nerve from wrist to the midpiece of thenar eminence delayed(>4ms),(3) The median nerve sensory nerve conduction velocity was absent, the M -lat of the median nerve delayed or was absent.

28 males and 59 females were included.Age ranging from (27~74) years old, and the average age was 54.7 years old. Course of disease ranging from (1 month~2years) .

2.2 Methods

2.2.1 Electrophysiological detection method

The instrument was the Oxford Medelec Synergy nerve EMG / evoked potential instrument.The parameters were set as follows: the band-pass range was from 3Hz to 10kHz , the scanning speed was 3ms/Div ,the stimulus intensity was 0 ~100mA (adjustable),and the pulse width was 0.1~1.0ms.

The SCV, M-lat, SNAP and CMAP of all patients were measured and recorded before the surgery for the first time, then the patients were divided into three groups, and the SCV, M-lat, SNAP and CMAP of all patients were measured and recorded again 3 months after the CTPAR.

2.2.2Procuring the SCV, M-lat, SNAP and CMAP

When the patient's skin temperature was > 32 °C , he/she lay down on the bed with supine position, and the anterograde stimulation method[9] was adopted.

Procuring the SCV and SNAP:First, the thumb and middle finger dominated by the median nerve sensory fiber were stimulated respectively with a ring electrode, second, the distance from the stimulus point to the record point was measured with a tape rule, the amplitude of SNAP was also measured, third, the SCV was calculated.

Procuring the M-lat and CMAP:The median nerve was stimulated at the wrist with a saddle-shape electrode, then CMAP was recorded at opponens pollicis with another surface electrode, at last the median nerve M -lat and the amplitude of CMAP were measured.

2.2.3 Statistical analyses

The statistical analyses were performed using SPSS 17.0. The measurement datas were expressed as $\bar{x} \pm s$, and paired t-test was used, the difference was statistically significant (P<0.05).

3. Results

3.1 The SCV and M -lat of the median nerve

The SCV and M-lat of of the median nerve of each group before and 3 months after the CTPAR (Table 1). In the mild CTS patients, the difference of the SCV before and 3 months after the CTPAR was statistically significant(P<0.05),while the difference of the M-lat displayed no statistical significance(P>0.05); In the moderate CTS patients, the SCV and M-lat improved obviously 3 months after the CTPAR, and the differences were statistically significant (P<0.01); In the severe CTS patients, the differences of the SCV and M-lat before and after the CTPAR had no statistical significance (P>0.05).

3.2The SNAP and CMAP of the median nerve

The SNAP and CMAP of the median nerve of each group before and 3 months after the CTPAR(Table 2). In the mild CTS patients, the difference of the SNAP before and 3 months after the CTPAR was statistically significant(P<0.05),while the difference of the CMAP showed no statistical significance(P>0.05); In the moderate CTS patients, the SNAP and CMAP improved obviously 3 months after the CTPAR, and the differences were statistically significant(P<0.01); In the severe CTS patients, the differences of the SNAP and CMAP before and after the CTPAR had no statistical significance(P>0.05).

Table 1. SCV,M-lat before and after the CTPAR

Groups	Mild		Moderate		Severe	
	SCV (m/s)	M-lat (ms)	SCV (m/s)	M-lat (ms)	SCV (m/s)	M-lat (ms)
Before CTPAR	40.35±1.62	3.34±0.75	25.83±4.63	4.28±0.67	0*	6.11±0.98
After CTPAR	47.61±2.35	3.48±0.76	38.25±2.11	3.67±0.86	0*	5.98±1.37
P	<0.05	>0.05	<0.01	<0.01	>0.05	>0.05

※ : SCVs weren't evoked before and after the CTPAR in the severe patients group.

Table 2. SNAP, CMAP before and after the CTPAR

Groups	Mild		Moderate		Severe	
	SNAP (μv)	CMAP (mv)	SNAP (μv)	CMAP (mv)	SNAP (μv)	CMAP (mv)
Before CTPAR	3.16±0.96	4.57±1.45	2.25±1.23	2.91±1.78	0*	1.55±1.31
After CTPAR	3.65±1.14	4.62±1.42	3.12±1.45	3.78±1.62	0*	1.52±1.27
P	<0.05	>0.05	<0.01	<0.01	>0.05	>0.05

※: SNAPs weren't evoked before and after the CTPAR in the severe patients group.

4. Discussions

CTS is one of the most common upper extremity compressive neuropathies[10], and the incidence of CTS is increasing in recent years[11]. The disease occurs in the population who repetitively use their wrists, a significant proportion of patients have onset bilaterally successively, and the right side was more serious. The United States statistics showed an annual incidence of 506 cases per 100,000 in females and 139 cases per 100,000 in males[10]. CTS is closely related to the specificity of carpal tunnel anatomy. The carpal tunnel is located in the root of palm, and it is made of bone and ligament that together form the tunnel like structure, in which the median nerve passes through. Because the carpal tunnel structure arranges tightly, the gap is very limited, and the organization of carpal tunnel is relatively tough and lack of flexibility, consequently, acute or chronic carpal tunnel pressure increased induced by any reasons may cause the median nerve compression, ultimately resulting into CTS[5]. The typical symptoms of CTS are numbness and the sensory abnormalities of thumb, index finger and middle finger, which often exacerbated at night, therefore early diagnosis and treatment can avoid occurrence of disabilities[12].

Due to the patients' or doctors' reasons, as well as other reasons such as the limitations of CT and X ray, CTS was often confused with cervical vertebra diseases, peripheral neuritis and so on, however, neural electrophysiological examination can provide CTS an objective and effective diagnostic basis. Some reports showed that the sensitivity of CTS diagnosed by neural electrophysiological examination was high, reached from 80% to 92%, as a result, most clinicians make the final diagnosis rely on electrophysiological examination for the clinical doubtful CTS patients[13]. Currently, CTS electrophysiological diagnosis is mainly based on the median nerve motor and sensory latency delay at wrist, and that the sensory latency delays (or SCV steps down) is not only more sensitive than motor latency dose, but also the must condition to diagnose

CTS[3,14]. The electrophysiological examination of early CTS is that only median nerve SCV slows down and evoked potential amplitude decreases, that the M-lat prolongs and the clinical symptoms gradually worsen are considered as the disease progresses[15].

In the experiment, 87 CTS patients had a preoperative neurophysiological detection, the result showed that CTS occurred in female patients easily, with a female to male ratio of 2.11. The abnormality rate of the median nerve sensory conduction was 100%, and the abnormal form was that different degrees of sensory conduction velocity slowed down or / and sensory action potential amplitude decreased or sensory conduction was absent, while the rate of the median nerve M-lat prolonged and the opponens pollicis CMAP amplitude decreased was 65.5%, the rate of the median nerve M-lat absent was 11.5%. In 3 months follow-up after the CTPAR, the clinical symptoms of mild and moderate CTS patients improved significantly, and the neural electrophysiological examinations after the CTPAR were compared with the ones before the CTPAR, the result showed that SCV (mild and moderate), M-lat (moderate), SNAP (mild and moderate) and CMAP (moderate) improved obviously, the differences were statistically significant ($P < 0.01$ or $P < 0.05$). While the clinical symptoms of 68.5% severe CTS patients were unchanged or deteriorated after the CTPAR, and neural electrophysiological examinations showed that SCV, M-lat, SNAP and CMAP didn't improve obviously, the differences weren't statistically significant ($P > 0.05$).

From the information above, the neural electrophysiological examination was used to assess the therapeutic effect of the CTPAR and was helpful to judge the improvement of clinical symptoms after the surgery. Although the postoperative recovery was correlated to the severity of CTS, the therapeutic effects of mild and moderate CTS patients were good without considering the severe CTS patients, especially the muscle atrophy patients, whose therapeutic effects were slow and poor. On the whole, the operation treatment is safe and reliable for carpal tunnel syndrome patients, and it can alleviate

the distress of patients and prevent further neurological damage. Therefore when the conservative treatment is invalid or the effect of the conservative treatment is not ideal, the neural electrophysiological examination should be done to judge the severity of CTS patients. If the patients are mild or moderate ones, the operation treatment should be performed as soon as possible to avoid muscular atrophy resulting in sensory and motor dysfunction, which will affect the quality of life.

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Application of Optimal Homotopy Asymptotic Method to the Equal Width Wave and Burger Equations

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Abstract: In this paper, we use the optimal homotopy asymptotic method (OHAM) for approximate solutions of the Equal Width Wave equations (EWW) and Burger equations, respectively. For (EWW) equations the numerical results obtained are compared with the results obtained by using variational iteration method (VIM) and Adomian decomposition method (ADM), while for burger equation comparison is made with ADM. From the obtained results it is observed that the suggested method is explicit, effective, and very easy to use. [Islam S, Nawaz R, Arif M and Shah SIA, **Application of Optimal Homotopy Asymptotic Method to the Equal Width Wave and Burger Equations.** *Life Sci J* 2012;9(4):2380-2386] (ISSN:1097-8135). <http://www.lifesciencesite.com>. 353

Keywords: Optimal Homotopy Asymptotic Method, Burger Equation, Equal Width Wave Equation

1. Introduction

Mathematical modeling of different problems in various fields has been discussed using formal approaches [20-32], fuzzy logic [19] and Differential equations [1-3]. Differential equations play a vital role in modeling different problems in physics, biology, chemical reactions and in engineering sciences. The one dimension non-linear differential equation, which is similar to the one dimension Navier-Stokes equation without the stress term, was presented for the first time in a paper in 1940 from Burger. It is the model for the solution of Navier-Stokes equation and is applied to laminar and turbulence flows as well. The first theoretical solution of Burger equation was given by Cole [13] which is based on Fourier series analysis using the appropriate initial and boundary conditions. Another theoretical solution was given by Madsen and Sincovec [14], based on the "test and trial" method, using the appropriate initial and boundary conditions. The Burger equation can be used as a model for different problems of a fluid flow nature, where shocks or viscous dissipation is a major factor. It can be used as a model for any nonlinear wave transmission problem subject to dissipation [11-15].

The Equal Width Wave (EW) equation was suggested by Morrison et al., to use as a model for the simulation of one-dimensional wave propagation in nonlinear media with dispersion processes [17].

Marinca and Herisanuet al. introduced a new semi analytic method (OHAM) for approximate solution of nonlinear problems of thin film flow of a fourth grade fluid down a vertical cylinder. They used OHAM for understanding the behavior of nonlinear mechanical vibration of an electrical machine. By using this method they investigated solution of nonlinear equations arising in the study of

fourth grade fluid past a porous plate. The convergence criterion of proposed method is similar to that of HAM and HPM, but this method is more efficient and flexible [1-3]. The proposed method has been used by many researchers for solution of Ordinary and Partial Differential equations [5-10]. The objective of this paper is to show the effectiveness of OHAM for (EWW) and Burger equations. We consider the EWW equation, derived for long waves propagating in the positive x-direction which has the form

$$\frac{\partial u(x,t)}{\partial t} + u(x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^3 u(x,t)}{\partial x^2 \partial t} = 0 \quad (1)$$

And Burger equation of the form

$$\frac{\partial u(x,t)}{\partial t} + \alpha u \delta (x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^2 u(x,t)}{\partial x^2} = 0 \quad (2)$$

With initial condition given by

$$u(x,0) = 0.5 + 0.5 \tanh\left(\frac{-\alpha \delta}{2(\delta + 1)} x\right) \quad (3)$$

for all $0 \leq x \leq 1$ and $t \geq 0$. α and δ are parameters and $\delta > 0$.

The whole paper contains four sections. Each of them is analyzed as follows. The first section is the introduction. The fundamental theory of OHAM is given in the section 2.

In section 3 numerical solution of (EWW) equation is presented by OHAM and absolute errors are compared with NIM and ADM. In section 4 Comparisons are made between the results of the proposed method and ADM for Burger equation. In all cases the proposed method yields very encouraging results.

Here we start by describing the basic idea of OHAM, Consider the partial differential equation of the form:

$$\mathcal{L}(u(x,t)) + \mathcal{N}(u(x,t)) + g(x,t) = 0, \quad x \in \Omega \tag{4}$$

$$\mathcal{B}(u, \partial u / \partial t) = 0, \tag{5}$$

where \mathcal{L} is a linear operator and \mathcal{N} is nonlinear operator. \mathcal{B} is boundary operator, $u(x,t)$ is an unknown function, and x and t denote spatial and time variables, respectively, Ω is the problem domain and $g(x,t)$ is a known function.

According to the basic idea of OHAM, one can construct the optimal homotopy

$$\begin{aligned} \psi(x,t;q) : \Omega \times [0,1] \rightarrow R \quad \text{which satisfies} \\ (1-q) \{ \mathcal{L}(\psi(x,t;q)) + g(x,t) \} = \\ \mathcal{H}(q) \left\{ \begin{aligned} &\mathcal{L}(\psi(x,t;q)) + \\ &\mathcal{N}(\psi(x,t;q)) + g(x,t) \end{aligned} \right\}, \end{aligned} \tag{6}$$

where $q \in [0,1]$ is an embedding parameter, $\mathcal{H}(q)$ is a nonzero auxiliary function for $q \neq 0$, $\mathcal{H}(0) = 0$. Eq (6) is called optimal homotopy equation. Clearly, we have:

$$q = 0 \Rightarrow \mathcal{H}(\psi(x,t;0), 0) = \mathcal{L}(\psi(x,t;0)) + g(x,t) = 0, \tag{7}$$

$$\begin{aligned} q = 1 \Rightarrow \mathcal{H}(\psi(x,t;1), 1) \\ = \mathcal{H}(1) \left\{ \begin{aligned} &\mathcal{L}(\psi(x,t;q)) + \\ &\mathcal{N}(\psi(x,t;q)) + g(x,t) \end{aligned} \right\} = 0. \end{aligned} \tag{8}$$

Clearly, when $q = 0$ and $q = 1$ it holds that $\psi(x,t;0) = u_0(x,t)$ and $\psi(x,t;1) = u(x,t)$ respectively. Thus, as q varies from 0 to 1, the solution $\psi(x,t;q)$ approaches from $u_0(x,t)$ to $u(x,t)$, where $u_0(x,t)$ is obtained from Eq (3) for $q = 0$

$$\mathcal{L}(u_0(x,t)) + g(x,t) = 0, \quad \mathcal{B}(u_0, \partial u_0 / \partial t) = 0. \tag{9}$$

Next, we choose auxiliary function $\mathcal{H}(q)$ in the form $\mathcal{H}(q) = qC_1 + q^2C_2 + \dots$ (10)

Here C_1, C_2, \dots are constants to be determined later. To get an approximate solution, we expand $\psi(x,t;q, C_i)$ in Taylor's series about q in the following manner,

$$\psi(x,t;q, C_i) = u_0(x,t) + \sum_{k=1}^{\infty} u_k(x,t; C_i) q^k, \tag{11}$$

$i = 1, 2, \dots$

Substituting Eq. (10) into Eq. (4) and equating the coefficient of like the powers of q , we obtain Zeroth order problem, given by Eq. (6), the first and second order problems are given by Eqs. (11-12)

respectively and the general governing equations for $u_k(x,t)$ are given by Eq. (13):

$$\mathcal{L}(u_1(x,t)) = C_1 \mathcal{N}_0(u_0(x,t)), \quad \mathcal{B}(u_1, \partial u_1 / \partial t) = 0 \tag{12}$$

$$\begin{aligned} \mathcal{L}(u_2(x,t)) - \mathcal{L}(u_1(x,t)) = C_2 \mathcal{N}_0(u_0(x,t)) + \\ C_1 \left[\begin{aligned} &\mathcal{L}(u_1(x,t)) + \\ &\mathcal{N}_1(u_0(x,t), u_1(x,t)) \end{aligned} \right], \end{aligned} \tag{13}$$

$$\begin{aligned} \mathcal{B}(u_2, \partial u_2 / \partial t) = 0 \\ \mathcal{L}(u_k(x,t)) - \mathcal{L}(u_{k-1}(x,t)) = \\ C_k \mathcal{N}_0(u_0(x,t)) + \sum_{i=1}^{k-1} C_i \left[\begin{aligned} &\mathcal{L}(u_{k-i}(x,t)) \\ &+ \mathcal{N}_{k-i} \left(\begin{aligned} &u_0(x,t), \\ &u_1(x,t), \dots, \\ &u_{k-i}(x,t) \end{aligned} \right) \end{aligned} \right], \end{aligned} \tag{14}$$

$$\mathcal{B}(u_k, \partial u_k / \partial t) = 0. \quad k = 2, 3, \dots,$$

where $\mathcal{N}_{k-i}(u_0(x,t), u_1(x,t), \dots, u_{k-i}(x,t))$ is the coefficient of q^{k-i} in the expansion of $\mathcal{N}(\psi(x,t;q))$ about the embedding parameter q .

$$\begin{aligned} \mathcal{N}(\psi(x,t;q, C_i)) = \mathcal{N}_0(u_0(x,t)) + \\ \sum_{k=1}^{\infty} \mathcal{N}_k(u_0, u_1, u_2, \dots, u_k) q^k. \end{aligned} \tag{15}$$

Here u_k for $k \geq 0$ are set of linear equations with the linear boundary conditions, which can be easily solved.

The convergence of the series in Eq. (10) depends upon the auxiliary constants C_1, C_2, \dots . If it is convergent at $q = 1$, one has:

$$\tilde{u}(x,t; C_i) = u_0(x,t) + \sum_{k=1}^{\infty} u_k(x,t; C_i). \tag{16}$$

Substituting Eq. (15) into Eq. (1), it results in the following expression for residual:

$$\mathcal{R}(x,t; C_i) = \mathcal{L}(\tilde{u}(x,t; C_i)) + g(x,t) + \mathcal{N}(\tilde{u}(x,t; C_i)). \tag{17}$$

If $\mathcal{R}(x,t; C_i) = 0$, then $\tilde{u}(x,t; C_i)$ will be the exact solution.

For computing the of auxiliary constants, $C_i, i = 1, 2, \dots, m$, there are many methods like Galerkin's Method, Ritz Method, Least Squares Method and Collocation Method to find the optimal values of $C_i, i = 1, 2, \dots, m$. One can apply the Method of Least Squares as under:

$$\mathcal{J}(C_i) = \int_0^t \int_{\Omega} \mathcal{R}^2(x,t, C_i) dx dt, \tag{18}$$

where R is the residual,

$$\mathcal{R}(x,t; C_i) = \mathcal{L}(\tilde{u}(x,t; C_i)) + g(x,t) + \mathcal{N}(\tilde{u}(x,t; C_i)) \text{ and}$$

$$\frac{\partial J}{\partial C_1} = \frac{\partial J}{\partial C_2} = \dots = \frac{\partial J}{\partial C_m} = 0. \tag{19}$$

The constants C_i can also be determined by another method as under:

$$\mathcal{R}(h_1; C_i) = \mathcal{R}(h_2; C_i) = \dots = \mathcal{R}(h_m; C_i) = 0, \tag{20}$$

$$i = 1, 2, \dots, m.$$

at any time t , where $h_i \in \Omega$. The convergence, depends upon constants C_1, C_2, \dots , can be optimally identified and minimized by Eq. (18).

3. Application of OHAM for (EWW) Equation.

Consider the (EWW) equation of the form (1)

$$\frac{\partial u(x,t)}{\partial t} + u(x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^3 u(x,t)}{\partial x^2 \partial t} = 0, \tag{21}$$

Subject to constant initial condition

$$u(x,0) = 3 \sec h^2 \left(\frac{x-15}{2} \right), \tag{22}$$

with exact solution given by

$$u(x,t) = 3 \sec h^2 \left(\frac{x-15-t}{2} \right), \tag{23}$$

Zeroth Order Problem

$$\frac{\partial u_0(x,t)}{\partial t} = 0,$$

Its solution is

$$u_0(x,t) = 3 \sec h^2(0.5(x-15)). \tag{24}$$

First Order Problem

$$\frac{\partial u_1(x,t)}{\partial t} - (1 + C_1) \frac{\partial u_0(x,t)}{\partial t} - \tag{25}$$

$$C_1 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x} + C_1 \frac{\partial^3 u_0(x,t)}{\partial x^2 \partial t} = 0.$$

$$u_1(x,0) = 0.$$

Its solution is

$$u_1(x,t,C_1) = -9C_1 t \operatorname{sech}^4(0.5(x-15)) \tag{26}$$

$$\tanh(0.5(x-15)).$$

Second Order Problem

$$\frac{\partial u_2(x,t)}{\partial t} - (1 + C_1) \frac{\partial u_1(x,t)}{\partial t} -$$

$$C_2 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x} + C_2 \frac{\partial^2 u_0(x,t)}{\partial x^2 \partial t} -$$

$$C_2 \frac{\partial u_0(x,t)}{\partial t} - C_1 u_1(x,t) \frac{\partial u_0(x,t)}{\partial x} -$$

$$C_1 u_0(x,t) \frac{\partial u_1(x,t)}{\partial x} + C_1 \frac{\partial^3 u_1(x,t)}{\partial x^2 \partial t} = 0.$$

$$u_2(x,0) = 0 \tag{27}$$

Its solution is

$$u_2(x,t,C_1) = -6.75 t C_1^2 \sec h^8(7.5 - 0.5x) \tag{28}$$

$$- 9C_1 t \operatorname{sech}^4(7.5 - 0.5x) \tanh(0.5(x-15))$$

$$- 9C_1^2 t \operatorname{sech}^4(7.5 - 0.5x) \tanh(0.5(x-15))$$

$$- 9C_2 t \operatorname{sech}^4(7.5 - 0.5x) \tanh(0.5(x-15))$$

$$- 31.5 C_1^2 t \operatorname{sech}^6(7.5 - 0.5x) \tanh^2(0.5(x-15))$$

$$+ 36 C_1^2 t \operatorname{sech}^4(7.5 - 0.5x) \tanh^2(0.5(x-15)).$$

Third Order Problem

$$\frac{\partial u_3(x,t)}{\partial t} - (1 + C_1) \frac{\partial u_2(x,t)}{\partial t} - C_3 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x}$$

$$+ C_3 \frac{\partial^3 u_0(x,t)}{\partial x^2 \partial t} - C_3 \frac{\partial u_0(x,t)}{\partial t} - C_2 u_1(x,t) \frac{\partial u_0(x,t)}{\partial x}$$

$$- C_2 u_0(x,t) \frac{\partial u_1(x,t)}{\partial x} + C_2 \frac{\partial^3 u_1(x,t)}{\partial x^2 \partial t} - C_2 \frac{\partial u_1(x,t)}{\partial t}$$

$$- C_1 u_2(x,t) \frac{\partial u_0(x,t)}{\partial x} - C_1 u_1(x,t) \frac{\partial u_1(x,t)}{\partial x} -$$

$$C_1 u_0(x,t) \frac{\partial u_2(x,t)}{\partial x} + C_1 \frac{\partial^3 u_2(x,t)}{\partial x^2 \partial t} = 0.$$

$$u_3(x,0) = 0 \tag{29}$$

Its solution $u_3(x,t,C_1,C_2,C_3)$ is obtained in the same manner. The third order approximate solution is of the form.

$$\tilde{u}(x,t,C_1,C_2) = u_0(x,t) + u_1(x,t,C_1) + \tag{30}$$

$$u_2(x,t,C_1,C_2) + u_3(x,t,C_1,C_2,C_3)$$

For the calculations of the constants C_1, C_2 and C_3 using the Method Least Squares we have computed that

$$C_1 = -2.683233 \text{B}47988335 \times 10^{-8},$$

$$C_2 = 3917.7906 \text{Q}532371, \text{ and}$$

$$C_3 = -32847.8147 \text{G}37734. \tag{31}$$

The 3rd order OHAM solution yields very encouraging results after comparing with 3rd order NIM and ADM solution [18].

Table 1.1: Comparison between the absolute error of the solution of EWW equation by He's variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and = 0.

t	VIM	ADM	OHAM
0.01	3.668×10 ⁻⁹	3.668×10 ⁻⁹	3.33195×10 ⁻⁹
0.02	7.333×10 ⁻⁹	7.334×10 ⁻⁹	6.66023×10 ⁻⁹
0.03	1.099×10 ⁻⁸	1.099×10 ⁻⁸	9.98485×10 ⁻⁹
0.04	1.465×10 ⁻⁸	1.465×10 ⁻⁸	1.33058×10 ⁻⁸
0.05	1.830×10 ⁻⁸	1.830×10 ⁻⁸	1.66231×10 ⁻⁸
0.1	3.652×10 ⁻⁸	3.652×10 ⁻⁸	3.31549×10 ⁻⁸

Table 1.2: Comparison between the absolute error of the solution of EWW equation by He’s variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and $x = 5$

t	VIM	ADM	OHAM
0.01	5.382×10^{-7}	5.429×10^{-7}	5.32925×10^{-7}
0.02	1.075×10^{-6}	1.085×10^{-6}	1.06531×10^{-6}
0.03	1.612×10^{-6}	1.627×10^{-6}	1.67319×10^{-6}
0.04	2.149×10^{-6}	2.168×10^{-6}	2.12844×10^{-6}
0.05	2.685×10^{-6}	2.709×10^{-6}	2.92244×10^{-6}
0.1	5.357×10^{-6}	5.405×10^{-6}	5.83132×10^{-6}

Table 1.3: Comparison between the absolute error of the solution of EWW equation by He’s variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and $x = 10$

t	NIM	ADM	OHAM
0.01	3.611×10^{-5}	4.852×10^{-5}	2.21357×10^{-5}
0.02	7.228×10^{-5}	9.698×10^{-5}	4.43255×10^{-5}
0.03	1.085×10^{-4}	1.453×10^{-4}	6.65695×10^{-5}
0.04	1.448×10^{-4}	1.936×10^{-4}	8.88676×10^{-5}
0.05	1.811×10^{-4}	2.418×10^{-4}	1.1122×10^{-4}
0.1	3.637×10^{-4}	4.819×10^{-4}	2.23787×10^{-4}

Table 1.4: Comparison between the absolute error of the solution of EWW equation by He’s variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and $x = 15$

t	NIM	ADM	OHAM
0.01	5.137×10^{-5}	6.000×10^{-6}	3.03829×10^{-7}
0.02	2.055×10^{-5}	2.400×10^{-5}	1.21531×10^{-6}
0.03	4.623×10^{-5}	5.400×10^{-5}	2.73445×10^{-6}
0.04	8.220×10^{-5}	9.600×10^{-5}	4.86123×10^{-6}
0.05	1.284×10^{-4}	1.500×10^{-4}	7.59564×10^{-6}
0.1	5.137×10^{-4}	6.000×10^{-4}	3.03816×10^{-5}

Table 1.5: Comparison between the absolute error of the solution of EWW equation by He’s variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and $x = 20$

t	NIM	ADM	OHAM
0.01	3.605×10^{-5}	4.860×10^{-5}	2.05582×10^{-5}
0.02	7.205×10^{-5}	9.728×10^{-5}	4.11931×10^{-5}
0.03	1.080×10^{-4}	1.460×10^{-4}	6.19047×10^{-5}
0.04	1.438×10^{-4}	1.948×10^{-4}	8.26931×10^{-5}
0.05	1.797×10^{-4}	2.437×10^{-4}	1.03558×10^{-4}
0.1	3.580×10^{-4}	4.894×10^{-4}	2.09041×10^{-4}

Table 1.6: Comparison between the absolute error of the solution of EWW equation by He’s variational iteration method (VIM) and Adomian decomposition methods (ADM) and optimal homotopy asymptotic method (OHAM) at various values of t and $x = 15$

t	NIM	ADM	OHAM
0.01	5.387×10^{-7}	5.434×10^{-7}	5.42223×10^{-7}
0.02	1.078×10^{-6}	1.087×10^{-6}	1.08499×10^{-6}
0.03	1.617×10^{-6}	1.632×10^{-6}	1.6283×10^{-6}
0.04	2.158×10^{-6}	2.177×10^{-6}	2.17216×10^{-6}
0.05	2.699×10^{-6}	2.722×10^{-6}	2.71657×10^{-6}
0.1	5.412×10^{-6}	5.459×10^{-6}	5.44683×10^{-6}

4. Application of OHAM for Burger Equation.

Let us consider Burger Equation of the form (2).

$$\frac{\partial u(x,t)}{\partial t} + \alpha u^\delta(x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^2 u(x,t)}{\partial x^2} = 0 \quad (32)$$

With initial condition given by

$$u(x,0) = 0.5 + 0.5 \tanh\left(\frac{-\alpha \delta}{2(\delta + 1)} x\right) \quad (33)$$

Case 1: when $\alpha = 1$ and $\delta = 1$

For $\alpha = 1$ and $\delta = 1$ the above equation takes the form

$$\frac{\partial u(x,t)}{\partial t} + u(x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^2 u(x,t)}{\partial x^2} = 0 \quad (34)$$

Subject to constant initial condition

$$u(x,0) = 0.5 - 0.5 \tanh(0.25x) \quad (35)$$

The exact solution of equation (24) with given condition is given by

$$u(x,t) = 0.5 - 0.5 \tanh[0.25(x - 0.5t)] \quad (36)$$

following the basic idea of OHAM presented in preceding Section we start with

$$L(\phi(x,t,q)) = \frac{\partial \phi(x,t,q)}{\partial t} \quad (37)$$

$$N(\phi(x,t,q)) = u(x,t) \frac{\partial u(x,t,q)}{\partial x} - \frac{\partial^2 u(x,t,q)}{\partial x^2} \quad (38)$$

The initial condition is:

$$\phi(x,0,q) = 0.5 - 0.5 \tanh[0.25(x - 0.5t)] \quad (39)$$

Zeroth Order Problem

$$\frac{\partial u_0(x,t)}{\partial t} = 0,$$

Its solution is

$$u_0(x,0) = 0.5 - 0.5 \tanh(0.25x) \quad (40)$$

First Order Problem

$$\frac{\partial u_1(x,t)}{\partial t} - (1+C_1) \frac{\partial u_0(x,t)}{\partial t} - C_1 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x} + C_1 \frac{\partial^2 u_0(x,t)}{\partial x^2} = 0$$

$$u_1(x,0) = 0. \quad (41)$$

Its solution is

$$u_1(x,t,C_1) = -t(0.0625C_1 \operatorname{sech}^2(0.25x)). \quad (42)$$

Second Order Problem

$$\frac{\partial u_2(x,t)}{\partial t} - (1+C_1) \frac{\partial u_1(x,t)}{\partial t} - C_2 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x} + C_2 \frac{\partial^2 u_0(x,t)}{\partial x^2} - C_2 \frac{\partial u_0(x,t)}{\partial t} - C_1 u_1(x,t) \frac{\partial u_0(x,t)}{\partial x} - C_1 u_0(x,t) \frac{\partial u_1(x,t)}{\partial x} + C_1 \frac{\partial^2 u_1(x,t)}{\partial x^2} = 0.$$

$$u_2(x,0) = 0 \quad (43)$$

Its solution is

$$u_2(x,t,C_1) = ((-0.0625C_1(1+C_1) - 0.0625C_2) t \sec^2(0.25x) + \sec^2(0.25x) (0.00195313C_1^2 t^2 \sinh(0.75x)) + \sec^4(0.25x)(0.00195313C_1^2 t^2 \tanh(0.75x))). \quad (44)$$

Third Order Problem

$$\frac{\partial u_3(x,t)}{\partial t} - (1+C_1) \frac{\partial u_2(x,t)}{\partial t} - C_3 u_0(x,t) \frac{\partial u_0(x,t)}{\partial x} + C_3 \frac{\partial^2 u_0(x,t)}{\partial x^2} - C_3 \frac{\partial u_0(x,t)}{\partial t} - C_2 u_1(x,t) \frac{\partial u_0(x,t)}{\partial x} - C_2 u_0(x,t) \frac{\partial u_1(x,t)}{\partial x} + C_2 \frac{\partial^2 u_1(x,t)}{\partial x^2} - C_2 \frac{\partial u_1(x,t)}{\partial t} - C_1 u_2(x,t) \frac{\partial u_0(x,t)}{\partial x} - C_1 u_1(x,t) \frac{\partial u_1(x,t)}{\partial x} + C_1 \frac{\partial^2 u_0(x,t)}{\partial x^2} = 0.$$

$$u_3(x,0) = 0 \quad (45)$$

Its solution is

$$u_3(x,t,C_1,C_2,C_3) = \left(\frac{1}{(1+e^{0.5x})^2} t \sec^2(0.25x) (-0.0625C_2 - 0.0625C_3 + C_1(-0.0625 + C_1(-0.125 - 0.015625t) + C_2(-0.125 - 0.015625t) - 0.000651042C_1^2(5.0718+t)(18.9282+t)) + ((288C_2 + 288C_3 + C_1(288 + 576C_2 + C_1(576 - 24t) - 24C_2t - 3C_1^2(-6.58301+t)(14.583+t) \cosh(0.25x) - (-96C_2 - 96C_3 + C_1(-96 - 192C_2 + C_1(-192 - 24t) - 24C_2t + 5C_1^2(-7.396+t)(2.596+t))) \sinh(0.25x) - 24C_2t + 5C_1^2(-7.396+t)(2.596+t))) \sinh(0.25x) - (-0.000651042 \cosh(0.75x) - 0.000651042 \sinh(0.75x))) \right). \quad (46)$$

Adding equations (40, 42, 44, 46) we obtain:

$$\tilde{u}(x,t,C_1,C_2) = u_0(x,t) + u_1(x,t,C_1) + u_2(x,t,C_1,C_2) + u_3(x,t,C_1,C_2,C_3) \quad (47)$$

For the calculations of the constants C_1, C_2 and C_3 using the Method Least Squares we have computed that

$$C_1 = -1.0000840 \quad 70827354,$$

$$C_2 = 9.46571385 \quad 1467995 \times 10^{-8}, \text{ and}$$

$$C_3 = -1.98940934 \quad 674601 \times 10^{-11}.$$

Putting the values of these constants into equation (34) the third order approximate solution using of OHAM is

$$u_3(x,t) = (0.5 - 5.26078452 \quad 47854 \times 10^{-6} t \operatorname{sech}^2(0.25x) + \frac{1}{(1+e^{0.5x})^2} (6.3600904 \quad 15451543 \times 10^{-9} + (1.3153067 \quad 008227217 \times 10^{-6} + 0.00065120 \quad 5881306229 \quad 2t) t + (1.2720180 \quad 853076355 \times 10^{-8} - 0.00260482 \quad 3525224917 \quad t^2) \cosh(0.5x) + (6.3600904 \quad 2653818 \times 10^{-9} + (-1.315306 \quad 7008227217 \times 10^{-6} + 0.00065120 \quad 5881306229 \quad 2t) t) \cosh(x) + (1.2720180 \quad 853076355 \times 10^{-8} - 0.00260482 \quad 3525224917 \quad t^2) \sinh(0.5x) + (6.3600904 \quad 2653818 \times 10^{-9} + (-1.315306 \quad 7008227217 \times 10^{-6} + 0.00065120 \quad 5881306229 \quad 2t) t) \sinh(x) - 0.5 \tanh(0.25x) + 0.00781381 \quad 3661895406 \quad t^2 \operatorname{sech}^2(0.25x) \tanh(0.25x) + \operatorname{sech}^2(0.25x) (0.0625052 \quad 5442670962 \quad t)). \quad (38)$$

The 3rd order OHAM solution yields very encouraging results after comparing with 4th order ADM solution [16]

Table 2.1: Comparison of absolute errors of 3rd order OHAM solution and 4th order ADM solution for Burger equation for $x = 0.1$ and $t \in [0, 2]$

t	Exact solution	Absolute Error ADM	Absolute Error OHAM
0.5	0.518741	6.34216×10^{-8}	5.32631×10^{-8}
1.0	0.549834	2.02886×10^{-6}	7.98928×10^{-8}
2.0	0.610639	6.42801×10^{-5}	3.2441×10^{-5}

Table 2: Comparison of absolute errors of 3rd order OHAM solution and 4th order ADM solution for Burger equation for $x = 0.5$ and $t \in [0, 2]$

t	Exact solution	Absolute Error ADM	Absolute Error OHAM
0.5	0.468791	5.66705×10^{-8}	5.82744×10^{-8}
1.0	0.50000	1.8471×10^{-6}	3.89112×10^{-6}
2.0	0.562177	6.06928×10^{-5}	7.40943×10^{-5}

Table 2.3 Absolute errors of 3rd order OHAM solution for various values of x and $t = 0.003$ and $t = 0.1$

x	Absolute Error for $t = 0.003$	Absolute Error for $t = 0.1$
-4	7.88258×10^{-15}	3.00377×10^{-10}
-2	8.54872×10^{-15}	2.023×10^{-9}
0	4.55191×10^{-15}	1.02224×10^{-10}
2	1.11022×10^{-15}	1.98526×10^{-9}
4	9.57567×10^{-16}	3.6574×10^{-10}

Table 2.4 Absolute errors of 3rd order OHAM solution for various values of x and $t = 0.5$ and $t = 0.1$

x	Absolute Error for $t = 0.5$	Absolute Error for $t = 1$
-4	1.86591×10^{-7}	2.68667×10^{-6}
-2	1.2447×10^{-6}	1.96202×10^{-5}
0	7.37402×10^{-8}	2.10382×10^{-6}
2	1.26475×10^{-6}	2.0391×10^{-5}
4	2.37125×10^{-7}	4.14971×10^{-6}

Case 2: when $\alpha = 1$ and $\delta = 2$

For $\alpha = 1$ and $\delta = 22$ equation (2) takes the form

$$\frac{\partial u(x,t)}{\partial t} + u^2(x,t) \frac{\partial u(x,t)}{\partial x} - \frac{\partial^2 u(x,t)}{\partial x^2} = 0 \tag{34}$$

Subject to constant initial condition

$$u(x,0) = 0.5 - 0.5 \tanh\left(\frac{x}{3}\right) \tag{35}$$

The exact solution of equation (24) with given condition is given by

$$u(x,t) = 0.5 - 0.5 \tanh\left[\frac{1}{4}\left(x - \frac{1}{3}t\right)\right], \tag{36}$$

Using same lines as above the third order approximate solution using OHAM is obtained and absolute errors for various values of x and t are given in table (2.5-2.6).

Table 2.5: Absolute error of the solution of Burger equation optimal homotopy asymptotic method (OHAM) at $x = 0.1$ and various values of t

t	Exact solution	OHAM solution	Absolute error
0.1	0.699207	0.699207	1.02602×10^{-11}
0.2	0.703168	0.703168	3.52315×10^{-9}
0.3	0.707107	0.707107	2.39034×10^{-8}
0.4	0.711024	0.711024	8.5422×10^{-8}
0.5	0.714919	0.714919	2.23819×10^{-7}
0.6	0.718791	0.718791	4.86729×10^{-7}
0.7	0.722639	0.722638	9.34107×10^{-7}
0.8	0.726464	0.726462	1.63865×10^{-6}
0.9	0.730263	0.73026	2.68617×10^{-6}
1.0	0.734037	0.734033	4.17601×10^{-6}

Table 2.6: Absolute error of the solution of Burger equation optimal homotopy asymptotic method (OHAM) at $x = 0.5$ and various values of t

t	Exact solution	OHAM solution	Absolute error
0.1	0.650264	0.650264	3.16105×10^{-10}
0.2	0.654428	0.654428	1.15677×10^{-9}
0.3	0.658578	0.658578	7.62659×10^{-10}
0.4	0.662715	0.662715	1.33499×10^{-8}
0.5	0.666837	0.666837	4.97044×10^{-8}
0.6	0.670944	0.670944	1.28683×10^{-7}
0.7	0.675035	0.675035	2.7546×10^{-7}
0.8	0.679109	0.679109	5.22082×10^{-7}
0.9	0.683166	0.683165	9.08017×10^{-7}
1.0	0.687205	0.687204	1.48069×10^{-6}

Conclusion

In this paper, the OHAM has been successfully implemented for the approximate solution of Burger and EWW Equations. For EWW equation the results obtained by OHAM are very consistent in comparison with ADM and VIM. For Burger equation the third order approximate solutions results of proposed method are very encouraging and agrees to the fourth order approximate solution by ADM.

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Finite Groups With At Most Nine Non T-Subgroups

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Abstract. In this short note, we extend the characterization of soluble groups by using the number of their non-T-subgroups and also we classify finite groups having exactly nine non-T-subgroups.

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Key words. Finite soluble groups, Normal subgroups, T-subgroups

1. Introduction

Group is an algebraic structure provides information about operations and relations satisfying certain algebraic conditions. Group and its algebraic properties provides many application in various fields like chemistry [8], Physics [9] and Computer sciences [10-23]. In this paper we concentrate on A group G is T- group when its every subnormal subgroup is normal. Thus T-groups are exactly those groups in which normality is a transitive relation. This structure has been studied by many authors such as Gaschutz [5], Zacher [4] and Robinson [6]. Robinson characterized finite soluble groups by T-groups. He proved that if all subgroups of a finite group are T-groups then G is soluble. For details we refer to Robinson [2].

Arif et al characterized finite soluble groups by number of non-T-subgroups. In [6] this has been proven that if a finite group G has at most 4 non-T-subgroups then the group will be soluble. It has also been shown there that finite groups having exactly five non-T-subgroups may not be soluble.

The purpose of this present note is to extend the characterization of finite groups up to 9 non-T-subgroups of group G . Thus we prove if G is a finite group all whose proper subgroups are T-groups except for $n = 6, 7, 8$, then G is soluble see Theorem 1. We also prove that if G is a finite group such that G contains exactly 9 non-T-subgroups then either G is soluble or G is isomorphic to $SL(2, 8)$ see Theorem 2.

Since our characterization is based on the number of non- T-subgroups so in Table 1 we present the details of the number of non-T-subgroups of each non-soluble group up to order 900 for the sake of completeness and for future use. We have used the following two GAP [3] functions to construct this

table. In Table 1, O denotes the order of non-soluble groups, N_1 denotes the number of non-soluble groups of the order given in column first and N_2 denotes the number of non-T-subgroups of each non-soluble subgroup of the group given in column second.

```
function( G ):= NrNonTSubgroups
local allsubgrps, i;
allsubgrps:=Subgroups( G );
for i in allsubgrps do
if IsSubnormal(G,i) then
if not IsNormal(G,i) then
return false;
break;
fi;
fi;
od;
return true;
end ;
function( G ):= NrNonTSubgroups
local allsubgrps;
allsubgrps:= Subgroups( G );
return Number(allsubgrps, t → not IsTGroup( t
));
end;
```

1. Preliminaries

The following results from [7] are needed.

- (1). If G is a finite simple group such that G has exactly n non-T-subgroups ($n \geq 1$), then G is isomorphic to a subgroup of S_n [7, Theorem 2.2].
- (2). Let G be a finite group with exactly n non-T-subgroups and suppose that any finite group with exactly m non-T-subgroups is soluble for $1 \leq m \leq n-1$ [7, Theorem 2.3].

Table 1. Number of non-T-subgroups of each non-soluble subgroup.

O	N_1	N_2
60	01	05
120	03	05,25,10
168	01	49
180	01	30
240	08	25,50,30,15,30,10,100,25
300	01	16
336	03	49,91,175
360	06	30,115,90,160,70,70
420	01	25
480	26	35,75,55,20,55,35,175,371,126,220,141,246,100,225,371,115,201,40,145,271,190,30,259,25,515,80
504	02	09,154
540	02	55,160
600	05	16,62,142,58,38
660	02	55,10
672	08	217,161,91,301,231,175,385,833
720	23	115,90,150,130,200,160,270,110,90,380,110,110,130,70,455,215,190,405,340,333,600,210,210
780	01	10
840	06	10,98,50,170,50,20
900	01	72

2. Main results

The following guarantees the solubility of a finite group having exactly n non-T-subgroups, $6 \leq n \leq 8$.

Theorem 1. *If G is a finite group all of whose proper subgroups are T-groups except for $n = 6, 7, 8$, then G is soluble.*

Proof. Let G be an insoluble group containing exactly n proper non T-subgroups, $L = \{H_1, H_2, \dots, H_n\}$. Clearly each H_i contains fewer than n proper non T-subgroups and so is soluble by the above result 2, no H_i can be normal. By the result 1, there is a homomorphism $f : G \rightarrow S_n$. Let K be the kernel of f . Then K cannot contain any of the H_i (since $K \leq \cap H_i$) and so K is soluble and in particular is a T-group. Then we must have G/K isomorphic to an insoluble subgroup of S_n . But S_6, S_7, S_8 has no non soluble subgroups which have 6, 7, 8 non T-subgroups respectively, a contradiction. Hence G is soluble for $n = 6, 7, 8$.

Theorem 2. *If G is a finite group such that G contains exactly 9 non T-subgroups. Then either G is soluble or G is isomorphic to $SL(2, 8)$.*

Proof. Suppose that there is an insoluble group with at most 9 non-T-subgroups and let G be a minimal counterexample. Since the property is preserved under taking subgroups and quotient groups, G must be simple. So, by result 1, G is isomorphic to a non soluble subgroup of S_9 and since $SL(2, 8)$ is the only non-soluble subgroup of S_9 which have 9 non-T-subgroups, therefore G is isomorphic to $SL(2, 8)$.

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Effect of Aquatic Pollution on Fish (Review)

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Abstract: Aquatic pollution is still a problem in many freshwater and marine environments as it causes negative effects for the health of the respective organisms. The present literature review of the effect of pollution on fish includes, water quality, pesticide, chemical miscellaneous and physical pollution. The present study was concluded that, there were inversely and reversely proportion relationship between pollution in aquaculture and the prevalence of external and internal parasitic infestation respectively. Control measures depend mainly on strict hygienic measure from the causative agent beside specific antipollution.

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Keywords: Aquatic pollution; fresh water; marine; environment; pesticide

1. Introduction

Fish plays an important role, not only in human food diets but also in animal and poultry rations. It is a palatable and easily digested food which is rich in vitamins, calcium, phosphorous and iodine. In Egypt, fish is considered as a cheap food article if compared with other foods of animal origin. The flesh of healthy fish is considered as a marker for the natural aquatic environment.

Factors which vary in freshwater environment, both in terms of the physical characteristic of rivers, fish farms, lakes and the chemical composition of water. Today, we are cushioned against the vagaries of our climate by central heating and perhaps air-conditioning as well as modern agricultural practices and food preservation techniques provide us with a constant year-round supply of food and a varied diet. We tend to forget that natural populations of animals and plants can undergo considerable changes in abundance from year to year in response to fluctuations in climate and in predator-prey relationships. Also, we tend to expect our environment to remain constant and to regard any deviations as the result of human interference. Too often we overlook the role of climatic changes, especially the frequency and seasonal pattern of rainfall, on the balance of aquatic animal and plant (**Farombi et al., 2007**). Heavy metals are natural trace components of the aquatic environment, but their levels have increased due to domestic, industrial mining and agricultural activities (**Kalay and Canli 2000**). Aquatic organisms such as fish accumulate metals to concentrations many

times higher than present in water (**Olaifa et al., 2004 and Noor El-Deen et al., 2010**).

Pollution is induced by human being reach, directly or indirectly through substances or energy (e.g. heat) into the marine environment (including estuaries) resulting in such deleterious effects as harm to living resources, hazards to human health, hindrance to marine activities including fishing, impairment of quality for use of seawater and reduction of amenities (**Gesamo, 1980**).

This definition, plants and animals that live sea are seen as a resource which needs to be conserved. If man's activities have a harmful effect on this, resource pollution has occurred. No such accepted definition exists for freshwater, but the general concept is transferable between the two environments. Man-made alterations to water-flow rates are understandably not included in this definition, but the general perception of pollution is that it is caused by substances (e.g. chemicals) and heated effluents (**Noor El-Din, 1997**).

Pollution as defined above is caused when the effect of the loading on the resource is unacceptable and the point at which the load of a chemical is sufficiently.

Water containing concentrations of chemicals which are below this stand are said to be contaminated a term used which indicates that the substances present in the water are at concentrations that are not harmful to the ecosystem. The use of this term can cause confusion because in other contexts, such as the purity of foodstuffs, this term is associated with harmfulness.

Aquatic pollution is still a problem in many freshwater and marine environments; it causes

negative effects for the health of the respective organisms (**Fent, 2007**). The number of studies dealing with effects of pollutants and concurrently occurring parasites is still relatively low (**Sures, 2007**). However the effect of environmental pollutants on fish parasites varies depending on the particular parasite and pollutant that interact (**Lafferty and Kuris, 1999**). Pollutants may affect the immune system of the fish either directly or by change water quality; that in turn may reduce the fish immunity to parasites (**Poulin, 1992**). Also, water pollution may accelerate the life cycle of the external parasites and promote their spread (**El-Seify et al, 2011**).

Osmoregulation

Fish differ from terrestrial organisms in that they have to maintain an osmotic equilibrium with the surrounding water. The body fluids in a freshwater fish have a much higher salt content than that of the surrounding water. Therefore, by simple osmosis, water is taken up into the body. As in humans, this water is filtered from the blood by the kidneys but in fish there is no resorption of water and a copious flow of urine is produced. Freshwater fish are in no danger of dehydration!

However, the kidneys retained much of the sodium and chloride from the urine in order to prevent undue loss of salt. Because fish skin is not waterproof, sodium and chloride escape from the body although the rate at which this occurs can be slowed up by the layer of mucus on the skin surface. This salt loss is restored by the active uptake of these elements via special cells in the gill (**Woo, 2006**).

Some natural factors that can modify the toxicity of chemical to fish.

Temperature: controls the rate of response to toxic concentrations, increases toxicity at low temperature? and affects the ionization of some sustenance (e.g. ammonia) and dissolved oxygen (DO), reduced DO causes increased toxicity and pathological changes in gills (**Robert, 2012**).

pH: controls the chemical species present (e.g. aluminium) or degree of ionization (e.g. ammonia). At the pH of the water falls below 5.5, so the harmful effects on fish begin to increase. These can be direct harmful effects on the ability of the fish to maintain their natural salt balance, especially in water with very low calcium content, or indirect effects such as that from a reduced food supply. There is a reduction in the productivity of natural waters at these low pH values because the recycling of nutrients is inhibited. Sudden increases in acidity to pH values below 4.5 may be lethal to fish; in general, the salmonid species are more resistant than other families of fish because

they are adapted to living in acid water (**Noor El-Din, 1997**).

Hardness (calcium carbonate): reduces the toxicity of divalent heavy metals. Humic and fulvic acid, forms low toxicity complexes with some metals (**Ahmed et al., 2010**).

Changes in both temperature and the DO content of the water will affect the resistance of fish to toxic chemicals. Also, changes in temperature, PH, water hardness and humic acids can affect their toxic state of some chemicals or compete with their uptake by fish (**Zdenka et al., 1993**).

Because toxic substances exert their harmful effects by interfering with normal chemical reactions within the body, the primary site of action will be within the cell. There are two ways by which such effects can be measured:

1) Direct measurements made on the concentration of various chemicals which can affect the vital processes within the cell, such as enzymes and the substrates on which they act. This can give an insight into the mode of toxic action; for example the inhibition of acetyl cholinesterase in nerve cells by organophosphorous insecticides prevents the normal transmission of electrical impulses.

2) Other reactions measured may be connected with the detoxification mechanism for a particular chemical. For example, metals such as copper, zinc and cadmium are transported in the blood throughout the body by protein molecules known as metallothioneins. Exposure of a fish to these metals in the water leads to an increase in the body levels of metallothioneins in order to remove them from the gills via the blood to sites where they can be safely stored or excreted. Therefore; metallothionein analysis can be used to determine whether fish have been exposed to higher levels of heavy metals in the environmental.

Another technique is the measurement of enzymes known as multifunctional oxygenases (MFOs) that are responsible for the breakdown of organic molecules within one of the major changes in gill structure that can reflect a change in water quality concerns the relative production of certain cell types present. For example, increases; in the abundance of the cells responsible for the excretion of mucus or the uptake of chloride (associated with the maintenance of osmoregulation) in relation to other types of epithelial cells. Another important use of histopathology is in the identification of abnormal cells (e.g. tumors in fish exposed carcinogenic chemicals).

Effect of some common heavy metals pollutants on fish:

Heavy metals (zinc, copper, lead, cadmium, etc) were recorded as major fish polluting chemicals in both developed and non developed countries (**Hamelink and Spacie, 1977**).

Sources

Probably the most important heavy metals sources was, and still in some countries, the waste arising from mining activities, such as mine drainage water. Effluent from tailings ponds (where waste crushed core is settled out) and drainage water from spoil heaps. These sources can continue to discharge heavy metals into watercourses long after the original mining activities have ceased. This problem was recognized in the last century and formed one of the topics for research on the effects of pollution on fish. Another important source is the industries that use these metals in various processes especially electroplating.

1- Zinc:

Because of the relatively high solubility of zinc compounds, this metal is widely detected in freshwater. Zinc as essential element for aquatic life: for example, it occurs in the enzyme carbonic anhydrase, catalyses the formation of carbonic acid from carbon dioxide in the blood. Small amount in the water or in the diet are therefore essential. It is also follows that the organisms will have an internal mechanisms to transport zinc around the body in order to manufacture such vital enzymes. When the zinc, in the water rises to a level where the amount entering the organism through the Gills exceeds the requirement for this metal. It was originally thought that the direct toxic action of zinc on fish was to precipitate the layer of mucus on the surface of the gill, causing suffocation. While this may be true for those species which produce a copious supply of mucus. The white precipitate observed on the gills of say rainbow trout is mainly composed of disintegrating epithelial cells which may be associated with the onset of mortality. Zinc may also cause a certain amount of tissue damage by reacting with protein and this could effected the respiratory efficiency as well as the osmoregulatory of function of the gills (**Andres et al., 2000 and Papagiannis et al., 2004**).

2- Copper

Good experimental data on copper toxicity are more difficult to obtained than for zinc. In hard water, copper precipitate out as a basic carbonate which is very slow to redissolve. It is difficult to prepare experimental solution in such waters because the colloidal precipitate which can be formed is not

acutely toxic and the amount of copper present in the toxic ionized form may be variable. Copper sulphate has been widely used in the past as an algacide in fish-bearing water, at concentrations which would be toxic if the metal was present in the toxic ionized form. In practice, no fish mortalities have been reported as a result of these operations, presumably because the copper was rapidly precipitate or complexed into much less toxic form. However most, if not all, of this inactive copper will ultimately enter sediment sinks where it may have limited bioavailability for organisms living there (**Figuro et al., 2006**).

The acute toxic action of copper seems to be similar to that of zinc. Also, lethal effects occurring at concentration down to 10% of the threshold LC_{50} . Growth rate of fish are affected at these concentration may be due to reduction in feeding rate or increased rate of activity (**Haemelink and Spacie 1977**).

As with zinc, the main environmental factor affecting the toxicity of copper is the calcium concentration of the water, again because of the competition, between these two ions for binding sites in the tissues of the gills and other organs. However, in contrast to zinc, salmonid species are not the most susceptible to copper toxicity; for example, perch may be three times as sensitive as rainbow trout to this metal (**Andres et al., 2000**). The reason for this difference in species sensitivity is not known and, it is possible that the toxic actions of copper and zinc are in some way slightly different.

3- Lead

Lead occurs naturally in the environment. However, most lead concentrations that are found in the environment are a result of human activities. Due to the application of lead in gasoline, an unnatural lead-cycle has consisted. In car engines lead is burned, so that lead salts (chlorines, bromines, oxides) will originated and enter the environment through the exhausts of cars. The larger particles will drop to the ground immediately and pollute soils or surface waters, the smaller particles will travel long distances through air and remain in the atmosphere. Part of this lead will fall back on earth when it is raining. This lead-cycle caused by human production is much more extended than the natural lead-cycle, and has caused lead pollution to be a worldwide issue (**Dowidar et al., 2001**).

Although lead has a high profile in human toxicology of much lesser importance for aquatic life. This is mainly due to a low solubility which limits its occurrence at significant concentration in all but very soft waters. Sub-lethal effects include the drinking of the trials of salmonid fish and his can be diagnostic of low levels of lead in the water. Some evidence of this

effect has been found in Fish Rivers receiving discharges from old lead mines in mid-wales (**Figuro et al., 2006**).

Lead has a tendency to accumulate in tissue and organs of exposed fish resulting in hepatic and renal dysfunction with growth retardation (**Haneef et al., 1998**). Consequently, it could induce alterations in hematological and serum biochemical parameters (**Gill 1991**) as well as pathological changes in most body organs (**Ghalab, 1997 and Mona S. Zaki et al., 2010**).

4- Mercury:

In animals, mercuric oxides cause inhibition of certain enzymes, which has several neurological effects. Next to the neurological effects vanadium can cause breathing disorders, paralyses and negative effects on the liver and kidneys. Mercuric and vanadium can cause harm to the reproductive system of male animals and rat and it is accumulated in the female placenta. Vanadium can be found in fishes and many other species. Mussels and crabs where mercuric and vanadium are strongly bioaccumulated, which can lead to concentrations of about 10^5 to 10^6 times greater than the concentrations that are found in seawater (**Figuro et al., 2006; Huang and Ghio, 2006; Nadal, 2007 and Zaki et al., 2011**). Mercury can be taken up by fish as the inorganic form but the main route is by absorption of mercury which has been methylated by bacterial Action in sediments. However, the standards set for safe level of mercury in fish for human consumption are lower than those that affect fish, so the importance of this metal to fish population is much reduced if the public health standards are met (**Worle et al, 2007**).

5- Cadmium:

Cadmium metal has a high profile in human toxicology where it has been transferred at harmful concentration concentrations through food chain. In the water, the main point source is effluents from electroplating works. Also, there are numerous diffuse inputs from the widespread use of this metal as well as a few areas where cadmium is leach from geological deposits. Cadmium is strongly adsorbed onto organic and inorganic particles in the water but, although it can form soluble complex with humic substance, the toxicity is not reduced as it is with copper (**Kargin and Coğun 1993 and Rashed, 2001**).

6- Other heavy metals

There is much less information on the toxicity to fish of other heavy metals such as aluminum, nickel, chromium and vanadium, mainly because of their lower importance as actual pollutants

in the fresh water environmental. Comprehensive reviews of the published data are available and the information should be assessed in the light of the comments which have already been made on potential source of error.

The toxicity of aluminium to fish is extremely complex as it can exist in many different chemical forms in water, depending on the pH, and these forms have differing toxicities. The most toxic chemical form is found within the range 5.2-5.8, and this may account for the reduced populations of fish found in waters with this range of acidity. As with other metals, the toxicity of aluminium is reduced when the calcium concentration of the water is increased, and added protection is also given by silicon salts (**Figuro et al., 2006**).

The correlation between the results of laboratory experiments and field observations on the effects of low pH and aluminium is made difficult because of the problems of controlling the acidity of test solutions to the required accuracy in the former, and the wide seasonal fluctuations in the latter which can depend on the frequency and extent of the rainfall, or, in some cases, the snow melt. It should be pointed out that these fluctuations in chemical conditions are more severe watercourses than in lakes where such changes are buffered by the large volume of water. However, effects on lake fish populations because of the feeder streams where they may breed can be subjected to intermittent harmful pulses of acidity (**Haemelink and Spacie 1977**).

The biological activities of the different types of vegetation in the catchment area may also be a factor in affecting the acidity of the run-off water. So the effect of acid rain on fisheries is still difficult to quantify, even though the main contributing factors to the problem have been identified and in some cases the concentration effect relationships have been accurately established. But the example of acidity serves to illustrate the difficulties in establishing the harmful effects of diffuse inputs of chemicals, in this case from the atmosphere via the soil of fisheries (**Fent, 2007**).

Effect of pollutions on fish reproduction:

Many studies found a change in behavior from the normal effect on reproduction including decrease of fertile eggs and time of reproduction interval due to herpidr and thermal effects of pollution on reproduction (**Jones and Reynolds, 1997**).

Effect of pollutions on fish parasites:

The aquatic pollution effects on parasitic infestations. The presence of pesticides, hydrocarbons, poly chlolorinated biphenyls, heavy

metals and sewage affected reversely on internal parasites and irreversibly on external parasites (**Blanar et al., 2009**).

Control

- 1- Prevent causative causes of pollution.
- 2- Strict hygienic measured.
- 3- Using specific antidotes.
- 4- The outhertoties notified the dangerous of pollution on health hazard.
- 5- Humic acid used as general antipollution.

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Synthesis and study of complexes of tetradentate Schiff base and bridging ligand of thiocyanate with transition metals of Fe, Cr and Co

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ABSTRACT: A group of combinations which have more importance in biological studies, biochemistry and also catalytic applications, are schiff base complexes. They have been considered by the chemists. In this research, Schiff base ligand was provided by means of 2,3-diaminopyridine and salicylaldehyde in absolute ethanol solvent with reflux method. Then the complexes of transition metals Fe, Cr and Co with the supplied ligand to being determined synthesized bridging thiocyanate and using FT-IR, cyclic voltammetry (CV) and UV-Vis spectroscopy. The information obtained from cyclic voltammograms of the complexes showed that the electrochemical behavior of the synthesized combinations indicates reversible the state of the complexes. Electronic spectra in the complexes accompanied by axial ligand with due attention to central electron giving ligand feature has decreased or increased transfers intensity which can be confirmed in complex formation.

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Keywords: Bridging thiocyanate, Schiff base, Transition metals, 2,3-Diaminopyridine and salicylaldehyde

INTRODUCTION

Schiff bases are very important in development of coordination chemistry. These compounds have a lot of uses. For example, dye industries, plastic, agriculture, biochemistry, physiology and providing liquid crystal [1,2]. Aliye Altundas and associates (2010) reported that pathogene structure in the field of medical has made in Cr(III), Zn(II) complex. All these elements have examined in order to antibacterial activity against pathogenic strains compounds like *Listeria monocytogenes* 4b (ATCC-19115), *Staphylococcus aureus* (ATCC25923), *Proteus* OX2 Wrah (ETS.40-A-4), *Escherichia coli* (ATCC-1280), *Salmonella typhi* H (NCTC-901.8394), *Pseudomonas putida* sp., *Brucella abortus* (A.99, UK-1995) RSKK-03026. *Sh. boydii* type 11 (Pasteur51.6), *Sh. boydii* type 16 (cHe 67.11), *Sh. boydii* type 6 (RSKK-96043), and antifungal activity against *Candida albicans* (Y-1200-NIH, Tokyo). Some of the compounds exhibited activity comparable to ampicillin ofloxacin, nystatin, kanamycin, sulphamethoxazol, amoxycillin, and chloroamphenicol [3]. The new Co(II), Cu(II), Ni(II) and Zn(II) complexes of

potentially N₂O₂ schiff base ligand [N,N'-bis(salicylaldehyde)-1,4-bis-(*o*-aminophenoxy)butane] (H₂L) prepared from 1,4-bis-(*o*-aminophenoxy)butane and salicylaldehyde in DMF. Obtained data from microanalysis, elemental analysis, magnetic measurements, ¹HNMR, ¹³CNMR, UV-Visible and IR spectrum indicated that they cause amplification of structure like as conductivity's measurement. In all complexes, H₂L functions as a tetradentate ligand [4]. Ismet kaya and associates (2008) reported that a new schiff base polymer has made as a compound (PHMPMDAP) by spectroscopy, elemental analysis and thermal analysis. This azomethine polymer, easily complexes with transitions elements Co(II), Zn(II), Fe(II), Cu(II) and Pb(II). By using the study of IR, UV-Vis phenol oxygen and safety nitrogen indicated that ligand has a configuration. Data Thermogravimetric analysis (TGA) indicates that polymer has as stability as monomer. Obtained polymer structure by using FT-IR, UV-Vis, ¹³CNMR, ¹HNMR has reinforced and detection has performed by TGA, SEC and solubility tests. Also,

electrical conductivities of PHMPMDAP and polymer-metal complexes are measured by four probe technique [5]. Novel zinc(II), copper(II), and cobalt(II) complexes of the Schiff base derived from 2-hydroxy-1-naphthaldehyde and D,L-selenomethionine were synthesized and characterized by elemental analysis, IR, electronic spectra, conductance measurements, magnetic measurements and powder XRD. The analytical data showed the composition of the metal complex to be $ML(H_2O)$, where L is the Schiff base ligand and $M = Co(II)$, $Cu(II)$ and $Zn(II)$. IR results confirmed the tridentate binding of the Schiff base ligand involving azomethine nitrogen, naphthol oxygen and carboxylato oxygen atoms. 1H NMR spectral data of lithium salt of the Schiff base ligand $[Li(HL)]$ and $ZnL(H_2O)$ agreed with the proposed structures. The conductivity values of complexes between 12.50 and $15.45 S\ cm^2\ mol^{-1}$ in DMF suggested the presence of non-electrolyte species. The powder XRD studies indicated that $Co(II)$ complex is amorphous, whereas $Cu(II)$ and $Zn(II)$ complexes are crystalline. The results of antibacterial and antifungal screening studies indicated that $Li(HL)$ and its metal complexes are active, but $CuL(H_2O)$ is most active among them [6].

MATERIALS AND METHODS

Ligand synthesis (2,3-diaminopyridine and salicylaldehyde)

In a 250 ml ballon we solved 1 mol 2,3-diaminopyridine (1 mmol and 0.109 g) in 30 ml absolute ethanol and refluxed it for half an hour. Then we added 2 mol salicylaldehyde (2 mmol and 0.2 cc) and refluxed it for 3 to 4 hours on a magnetic mixer and we continued refluxion until refluxion sediment formed. When a dark brown sediment formed we filtrated the sediment. To make sure that the sample is pure, we did TLC test during the experiment. First, we solved a little amount of the sample in ethanol and with a capillary tube we put the sample on TLC paper and we put it in hexane and ethyl acetate (1:1) solvent. Blemish went up in the solvent. Because it has capillary feature and it is made sure of sample purity by seeing just one blemish in the tank. Also sample electrical conduction in ethanol solvent with $10^{-3} M$ concentration was measured and it was $0.11\ \mu s/cm$ in $16\ ^\circ C$ figure (1).

The synthesized combination has the following formula:

Chemical formula: $C_{19}H_{15}N_3O_2$
Molecular weight: $317.33\ g.\ mol^{-1}$

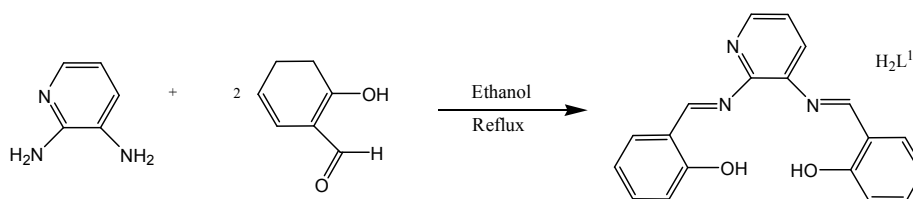


figure (1) Ligand 2,3-diaminopyridine and salicylaldehyde synthesis

Fe(III) chloride, Cr(III) chloride and Co(II) acetate synthesis respectively and separately with 2,3-diaminopyridine Schiff base ligand and salicylaldehyde with thiocyanate axial ligand

In a 250 ml ballon, we solved the ligand obtained (1 mmol and 0.31 g) in 30 ml absolute ethanol and they were refluxed for half an hour. Then to the same proportion of stoichiometry for the Fe

sample, iron salt (III) chloride (1 mmol and 0.27 g), for Cr sample, chromium salt (III) chloride (1 mmol and 0.26 g) and for Co sample, cobalt salt (II) acetate (1 mmol and 0.24 g) were added. It was refluxed on a magnetic mixer for an hour. Finally thiocyanate salt was added as axial ligand to the mixture of Fe, Cr and Co samples reactions respectively and separately in a stoichiometry proportion (6 mmol and 0.58 g). Refluxion

continued for 3 more hours until sediment formed. Samples sediment became brown after becoming dry. To make sure of the sample purity, during the experiment, TLC test was done. Sample melting point was about 205°C for Fe, 120 °C for Cr and 89 °C for Co. Complexes electric conduction in ethanol solvent with a 10^{-3} M concentration was measured and its amount was 0.96 $\mu\text{s}/\text{cm}$ for Fe, 0.12 $\mu\text{s}/\text{cm}$ for Cr and 0.11 $\mu\text{s}/\text{cm}$ for Co in 16 °C. Fe, Cr and Co samples in ethanol, acetonitrile,

methanol, DMF and DMSO can be easily solved figure (2,3,4).

The synthesized combinations has the following formula:

Chemical formula(Fe): $\text{C}_{20}\text{H}_{13}\text{N}_4\text{O}_2\text{SFe}$

Molecular weight(Fe): 429.25g. mol^{-1}

Chemical formula(Cr): $\text{C}_{20}\text{H}_{13}\text{N}_4\text{O}_2\text{SCr}$

Molecular weight(Cr): 425.40g. mol^{-1}

Chemical formula(Co): $\text{C}_{20}\text{H}_{13}\text{N}_4\text{O}_2\text{SCo}$

Molecular weight(Co): 432.34g. mol^{-1}

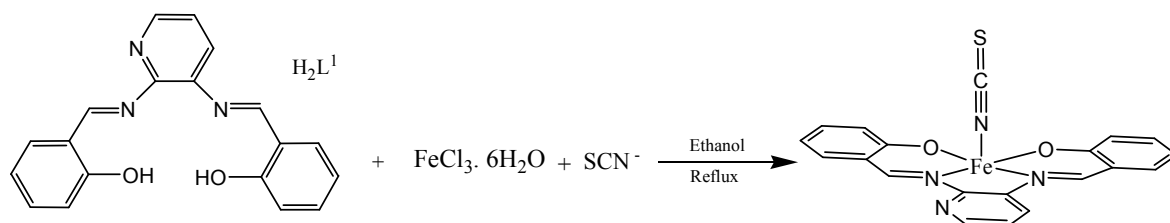


figure (2) H_2L^1 with bridging thiocyanate

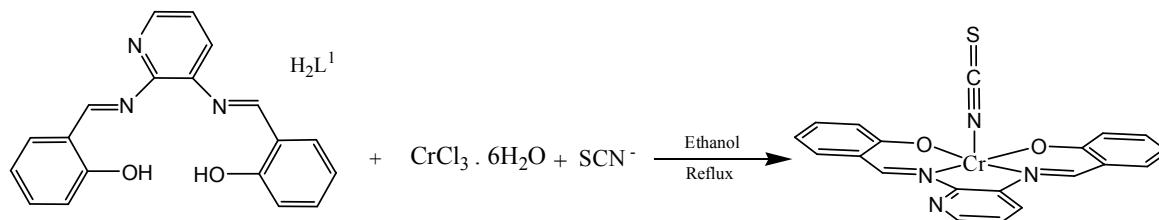


figure (3) H_2L^1 with bridging thiocyanate

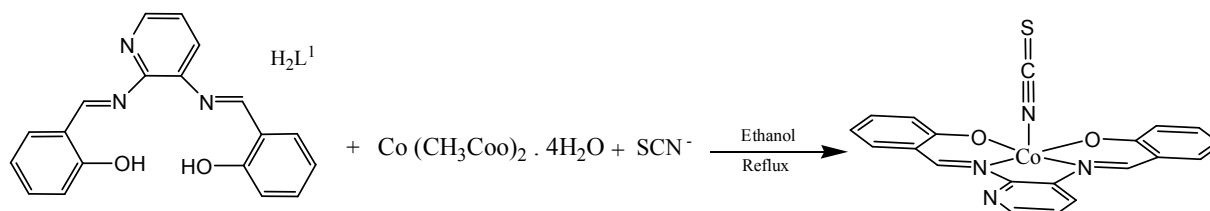


figure (4) H_2L^1 with bridging thiocyanate

RESULTS AND DISCUSSION

FT-IR spectra features of ligand combination

There is an absorption band in 3411.78 cm^{-1} range that is related to phenol OH stretch vibration. There are bands of aromatic stretch CH in 2925.44 cm^{-1} range and aromatic bending CH band

in 665.95 cm^{-1} . There is a strong absorption band in 1610.95 cm^{-1} that is related to stretch vibration of safety group C=N and an absorption band in 1397.06 cm^{-1} that is related to phenol C-O group figure (5).

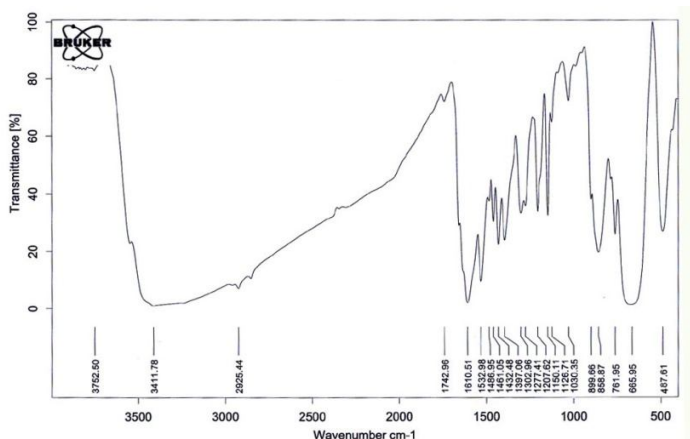


figure (5) FT-IR spectra ligand combination

Cyclic voltammery features of ligand combination

Synthesized ligand cyclic voltammogram had investigated with scanning rate 0.1 Vs^{-1} and in 1 to -2 volt range in DMSO solvent. In high range of diagram, ie, in oxidation part, there is a short peak in $E_a = -0.78\text{ V}$, also there is another peak in $E_a = 0.21\text{ V}$. In low range of diagram, ie, in the

reduction part, there is a short peak in $E_c = 0.19\text{ V}$ range and there are two peaks with low height respectively $E_c = -0.42\text{ V}$ and $E_c = -1.7\text{ V}$. With a comparison of oxidation and reduction peaks, it can be resulted that there is a peak in reduction region like as a peak in oxidation region. Thus, its electrochemical behavior shows reversibility of ligand figure (6).

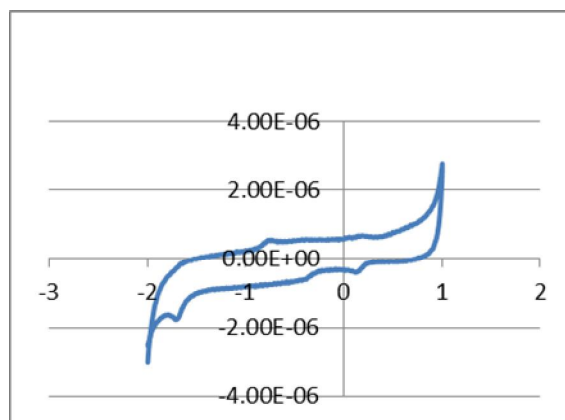


figure (6) Cyclic voltammogram ligand combination

UV-Vis spectra features of ligand combination

There are transitions in synthesized ligand around 200-280 nm range that is related to aromatic ring $\pi \rightarrow \pi^*$ transitions. 280-350 nm range is related to

$\pi \rightarrow \pi^*$ electron transition of safety bond system that it has a high peak. In 350-400 nm range, there are $n \rightarrow \pi^*$ transitions with a sharp and high peak that is related to nonbond electrons on nitrogen atoms of azomethine bond figure (7).

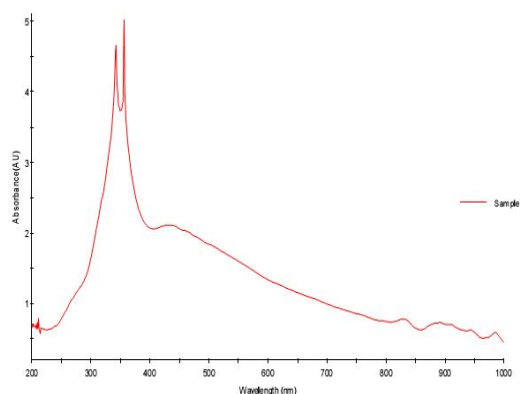


figure (7) UV-Vis spectra ligand combination

FT-IR spectra features of Fe(III) chloride with bridging thiocyanate combination

In FT-IR spectra, there is an iron complex that has displaced in absorption band with regard to free ligand that indicates coordination of central metal atom to electron donation atoms in ligand structure. In this complex, there is a C=N bond in

1617.37 cm^{-1} frequency that it indicates coordination of central metal to ligand. Absorption band that has obtained in 3411.02 cm^{-1} range is related to phenol OH stretch vibration. There is CH vibration in 684.61 cm^{-1} that is related to aromatic bending and also CH vibration that is in 2924.95 cm^{-1} indicates aromatic stretch. There is phenol C-O vibration in 1384.40 cm^{-1} range figure (8).

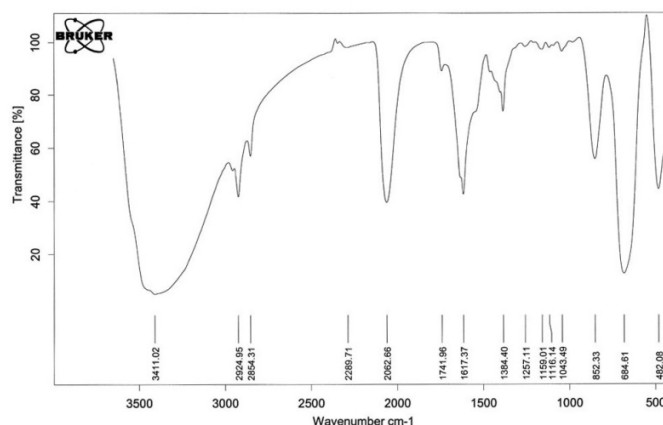


figure (8) FT-IR spectra Fe(III) chloride with bridging thiocyanate combination

Cyclic voltammetry features of Fe(III) chloride with bridging thiocyanate combination

Complex cyclic voltammogram Fe(III) that synthesized with scanning rate 0.1 Vs^{-1} had investigated in 0.9 to -2 volt range in DMSO solvent. In comparison of ligand diagram with obtained complex, there is a peak in $E_a = -0.25 \text{ V}$ range in oxidation region and also there is a rather high peak in $E_a = 0.76 \text{ V}$ range that has displaced with regard to a ligand peak. In low range of diagram, there are two peaks in $E_c = 0.1 \text{ V}$ and $E_c = -0.6 \text{ V}$ respectively that has displaced with regard

to a ligand peak in reduction range. With a comparison that obtained from oxidation and reduction of this diagram, it can be resulted that there are two peaks in oxidation range like as reduction range, thus its electrochemical behavior shows the reversibility of complex, ligand and metal are near together, for this reason a peak of oxidation range is rather full. Because of displacements that has taken both in oxidation and reduction ranges, it is an approval for formation of compound and supplied complex is an electroactive complex figure (9).

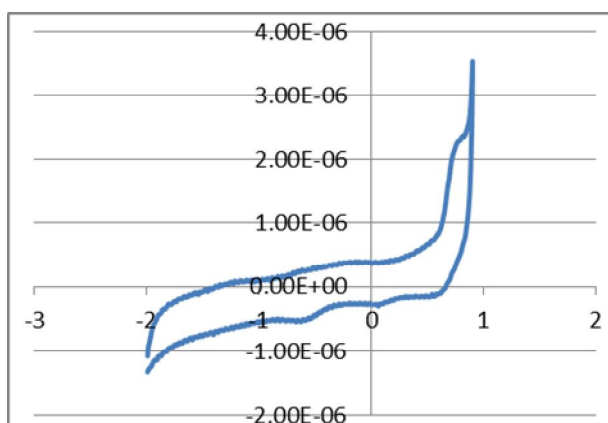


figure (9) Cyclic voltammogram Fe(III) chloride with bridging thiocyanate combination

UV-Vis spectra features of Fe(III) chloride with bridging thiocyanate combination

In this complex, there is an absorption band in 200-280 nm that is related to aromatic $\pi \rightarrow \pi^*$ transitions and there is a high peak in 280-400 nm that is related to $\pi \rightarrow \pi^*$ transitions of C=N bond. There is a peak in 400-450 nm that is related to $n \rightarrow \pi^*$ transition and also d-d complex transfers have appeared in 450-800 nm. Transfer rate has

influenced by axial ligand and electronical rate of this ligand to central metal that it is an approval for formation of this complex. Comparing electronic spectra of this combination with ligand spectra is an indicator of coordination of central metal into 2 nitrogen and phenol oxygen and the transfers in the above mentioned combination have moved towards red wavelength range figure (10).

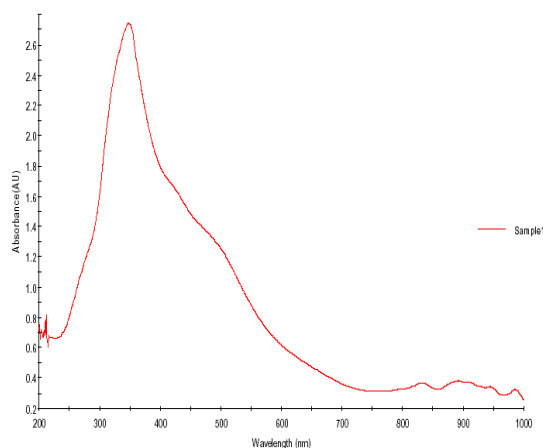


figure (10) UV-Vis spectra Fe(III) chloride with bridging thiocyanate combination

Cyclic voltammetry features of Cr(III) chloride with bridging thiocyanate combination

Synthesized complex cyclic voltammogram Cr(III) with 0.1 Vs^{-1} scanning rate and in 1.2 to -2 volt range had investigated in DMSO solvent. With a comparison of a ligand diagram that obtained with complex, there has been a peak in $E_a = -0.63 \text{ V}$ range and also there are two another peaks in $E_a = -0.12 \text{ V}$ and $E_a = 0.88 \text{ V}$ ranges that has displaced with regard to ligand peak. There are three peaks in $E_c = 0.18 \text{ V}$, $E_c = -0.61 \text{ V}$ and $E_c = -0.75 \text{ V}$ ranges in reduction part that has displaced with regard to a

ligand peak in reduction range. With a comparison that obtained from oxidation and reduction peaks in this diagram, it can be resulted that there are three peaks in oxidation range like as reduction range, thus its electrochemical behavior shows its reversibility of complex and ligand and metal are near together, for this reason, a peak in oxidation region is rather full. Because of displacements that has taken both in oxidation and reduction ranges, it is an approval for formation of compound and supplied complex is an electroactive complex figure (11).

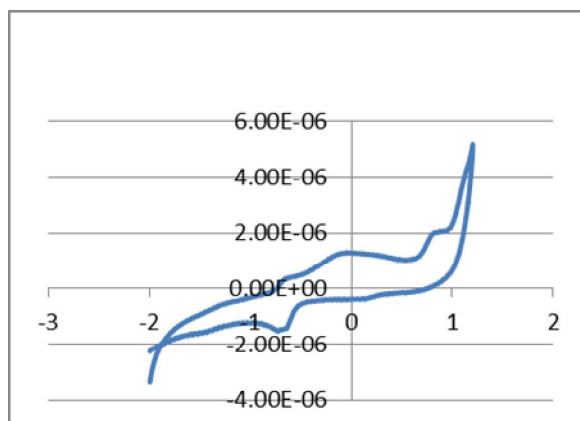


figure (11) Cyclic voltammogram Cr(III) chloride with bridging thiocyanate combination

UV-Vis spectra features of Cr(III) chloride with bridging thiocyanate combination

In this complex, there is an absorption band in 200-280 nm range that is related to aromatic $\pi \rightarrow \pi^*$ transitions and there is a high peak in 280-410 nm range that is related to $\pi \rightarrow \pi^*$ transitions of C=N bond. There is a peak in 410-450 nm range that is related to $n \rightarrow \pi^*$ transition and also d-d complex transfers have appeared in 450-800 nm. Transfer

rates has influenced by the presence of axial ligand and electron donation rate of this ligand to central metal that it is an approval for formation of this complex. Comparing electronic spectra of this combination with ligand spectra is an indicator of coordination of central metal into 2 nitrogen and phenol oxygen and the transfers in the above mentioned combination have moved towards red wavelength range figure (12).

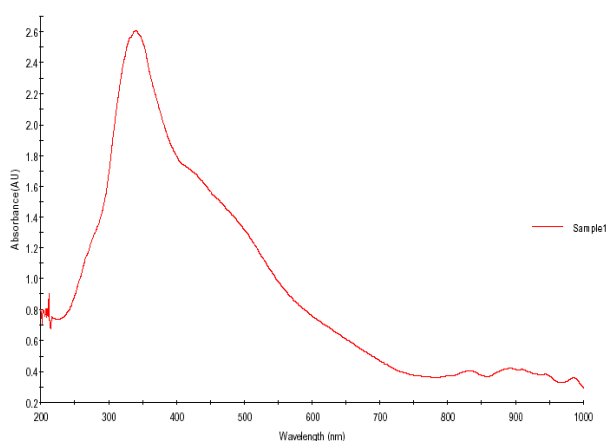


figure (12) UV-Vis spectra Cr(III) chloride with bridging thiocyanate combination

Cyclic voltammetry features of Co(II) acetate with bridging thiocyanate combination

Synthesized complex cyclic voltammogram Co(II) with scanning rate 0.1 Vs^{-1} had investigated in 1 to -2 volt range in DMSO solvent. With a comparison of ligand diagram, there is a peak in high range of diagram, ie, in oxidation part in $E_a = -0.24 \text{ V}$ and there is another peak in $E_a = 0.76 \text{ V}$ range that has displaced with regard to ligand peak, in low range of diagram, ie, in reduction part, there are two very small peaks in $E_c = 0.2 \text{ V}$, $E_c = -0.6 \text{ V}$ respectively that has displaced with regard to ligand peak in

reduction range. With a comparison of oxidation and reduction peaks that obtained from this diagram, it can be resulted that there is a peak in oxidation region like as reduction region, thus its electrochemical behavior shows reversibility of complex and ligand and metal are near together, for this reason, the peak of oxidation range is rather full. For the reason of displacements that has performed both in oxidation and reduction ranges, it is an approval for formation of compound and supplied complex is an electroactive complex figure (13).

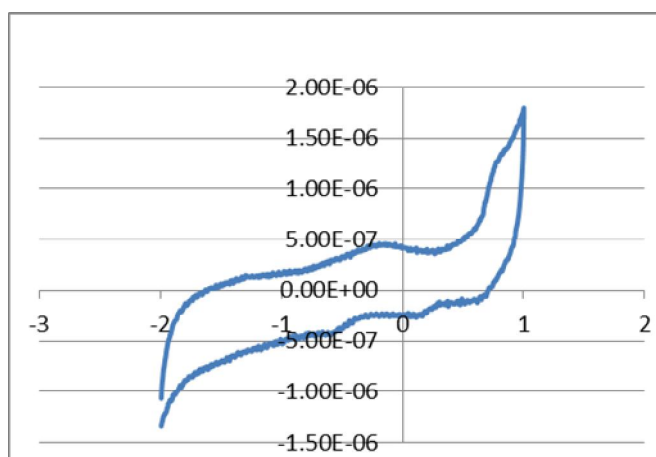


figure (13) Cyclic voltammogram Co(II) acetate with bridging thiocyanate combination

CONCLUSION

The results obtained from UV-Vis spectra, FT-IR and also the information gotten from synthesized complexes cyclic voltammetry can confirm the considered complex formation. In this research, quite new structures were obtained due to using thiocyanate bridging combination. UV-Vis spectra of the complexes which shows $\pi \rightarrow \pi^*$ transfers presence related to aromatic ring and absorbing band of $n \rightarrow \pi^*$ safety link and d-d transfers of the center of metals is a reason for Schiff base formation and these bands movement in electronic spectra of the complexes, It is an indicator of Schiff base coordination with metal. Cyclic voltammetry makes it possible to be able to get

information not only about electronic spectrum, but also about electronic features of the complexes and evaluate the possibility of their usage as a catalyst in oxidation–reduction reactions.

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An applicable modified probabilistic method for seismic hazard assessment in Northern Khorasan province, Iran

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Abstract: Iran plateau is a vast pressured region along mountain building belt of Alps-Himalayas. North Khorasan province where is studied in this research is located in Kope Dagh seismotectonic province. Kope Dagh seismotectonic province is a mountainous region which final phases of Alps have played a basic role in its present-day formation. This province is an active seismic region with mostly shallow earthquakes with less than 30 kilometers depth. Seismicity of the region indicates high activity in Northern Khorasan and Kope Dagh plate in the area of 56° to 59.30° E and 36° to 38° N. Regarding to the high seismicity and importance of Kope Dagh region because of the existence of Mozduran gas reservoir, which after refinement and desulphurization supplies fuel of Shirvan and Bojnourd cities, and due to economic and cultural importance, it is crucial to analyze seismic hazard in this region. It is important to note that however lots of studies have been done in north-east of Iran, but building codes modification needs more precisely seismic hazard analysis. Furthermore, previous studies have been applied free download softwares, which were provided before 2000, but the most important advantage of this study is applying professional industrial software, which has been written in 2009. This applied software can cover previous software weak points very well such as gridding potential sources, attention to the seismogenic zone and applying attenuation relationships directly. In the present study, modified probabilistic method has been applied instead of traditional one. For this purpose at first, a complete seismic catalogue of main earthquakes of the region provided. Then, map of potential sources prepare and compared to the previous studies. Finally, seismic parameters of the region have been determined for seismic hazard analysis. This study states the most PGA will be experienced in a northwest-central trend, which fits very well with Quchan-Robat and Esfarayen-Robatqarebil faults and matches with the expected trend based on tectonic evidences and previous statistical studies. In this research peak ground acceleration (PGA) will be experienced up to 0.07 gal during the expected earthquake with 63% probability of occurrence in 50 years in the area of Robatqarebil.

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Keywords: Seismic Hazard Analysis, Peak Ground Acceleration, EZ-FRISK Software, Northern Khorasan

Introduction

Among the areas prone to earthquake in Iran, Esfahan, Yazd, and the coastal plains of Khuzestan and Bushehr are the safest areas, while Azerbaijan (e.g., Tabriz, etc.) and Khorasan (e.g., Tabas, Bayaz Plain, etc.) are the most hazardous places. The studied area in the present research is located in active areas where are prone to destructive earthquakes ($M_s > 6$). The concerned zone in this study is located in longitude of 56° to 59° E and latitude of 36° to 38° N as a part of the Kope Dagh seismotectonic province, where several earthquakes have been happened. The required data in this study have been obtained from the seismic Network of Tehran University Geophysics Institute, as well as global seismic information web sites, such as ISC and USGS.

General Geology

Northern Khorasan Province, which is located in northeast of Iran, can be geologically divided into Kope Dagh and Binalood structural separate area parts.

- a) **Kope Dagh structural Area:** The north and northwest parts of Northern Khorasan Province can be considered as a part of the northern Iran pressure zone in Kope Dagh. Geographically and orographically point of view, Kope Dagh is part of the eastern continuation of the Alborz Mountains. Its structural and geological features are different from those of the surrounding areas.

Kope Dagh is a mountainous area whose present form affected by the late Alpien phase. Normally, anticlines form the heights, whereas synclines form the between-mountains plains. Moreover, Mozduran carbonate components (upper Jurassic) and Tiregan lower Cretaceous are the typical units forming the appearance of this area. The Shirvan-Bojnord plain in this province is one of the low areas in Kope Dagh. The Kope Dagh Area has been formed as a sedimentary plate in the upper Triassic, where different types of continuous marine sediments, from Jurassic to Oligocene, are settled on each other with the same slope, as thick as 6000 meters. The sediment in the Kope Dagh sedimentary area in the Jurassic era is similar to that of the Alborz. Nevertheless, from Cretaceous, it has obtained its own specific features. Its stratigraphical hierarchy consists, in a chronological order, of the structures of Kashafrood, Chamanbid, Mozduran, Shuriche, Tirgan, Sarcheshme, Sangan, Etamir, Abderaz, Abtalkh, Neizar, Kalat, Pesteligh, Chehelkaman, and Khangiran. Moreover, it is as ancient as the Jurassic and Neogene. The zone is rich in gas resources and some non-metallic minerals.

- b) **Binalood Structural Area:** This area is restricted between Tooran rigid plate and the mini-continent of central Iran. The south boundary is Mayamei fault or Shahrood fault, while its north-west boundary is Semnan fault. The zone is part of the Alborz, with individualistic geological features. Binalood geological unit is considered as the gradual zone between central Iran and the Alborz, since the sediment and Paleozoic features found in this zone are similar to those of the Alborz. The zone is rich at minerals, especially some metals, such as iron, and construction materials (Tchalenko, 1975).

In general, the main faults of the studied region, which earthquakes of this province occur due to them can be listed as:

1. **Main fault zone located in northeast (Kope Dagh):** it joins the north-west and Caspian Sea

seismic belt; moreover, the Turkmen earthquakes are related to this zone.

2. **Bokharden-Quchan fault zone:** it causes greatest earthquakes in northeast of Iran.
3. **Baghan-Garmab fault:** extended in a NW-SE direction, it starts from near the Turkmen boundary and continues up to the cities Shirvan, Faruj, and Quchan. Its length is 50 to 55 km, and located in Kope Dagh.
4. **Esfarayan fault:** it is classified as being among seismic potential sources, having blind rupturing and causing earthquakes without outcrop.
5. **Robat-e-ghare Bil fault:** it is one of the Quaternary faults.
6. **Bojnord fault:** it causes several earthquakes with Ms more than 5. It is on the south of the Bojnourd city.
7. **Jajarm fault:** it is considered as one of the potential seismic sources with inverse focal mechanism (Berberian et al, 1982).

Statistical study of historical and instrumental events (1900-2011)

These events, which commonly are based on historical documents, contain time interval between 943 to 1895. Figure 1 depicts number of occurrence to time diagram in the notified period. Time intervals without any seismic activity are signified from the diagrams. Iran's instrumental recording started at the end of 19th century with European seismograms. However, number of the stations well equipped by qualified recording instruments has been developed fast after 1963 and again there is a quality improvement of data after 1973. Figures 2 and 3 respectively show the number of events by time for time intervals of 1963 to 1974 and 1974 to 2011.

Seismic hazard assessment, probabilistic method

Probabilistic methods, which are applied to assess seismic hazard analysis, are provided by Cornell (1968).

It has the following steps:

- 1- Determining seismic potential sources
- 2- Obtaining seismic parameters
- 3- Estimation of event effects in the desired site
- 4- Seismic hazard assessment for the desired site

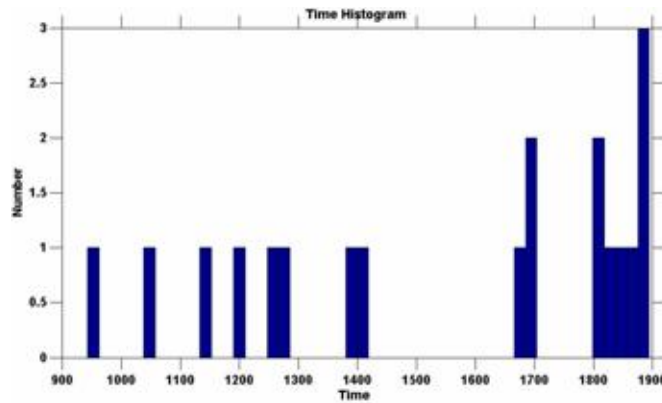


Figure1. Number of historical events by time

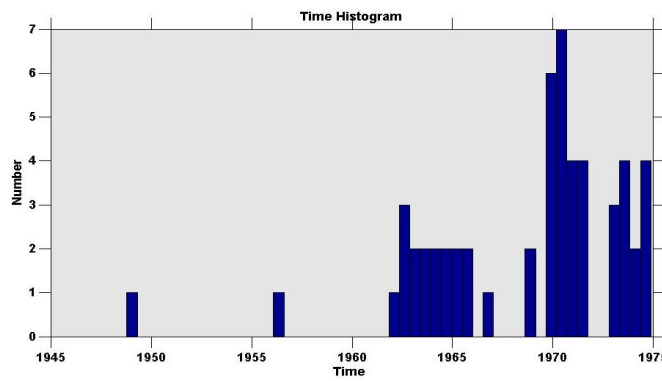


Figure2. Number of events by time (1963 to 1974)

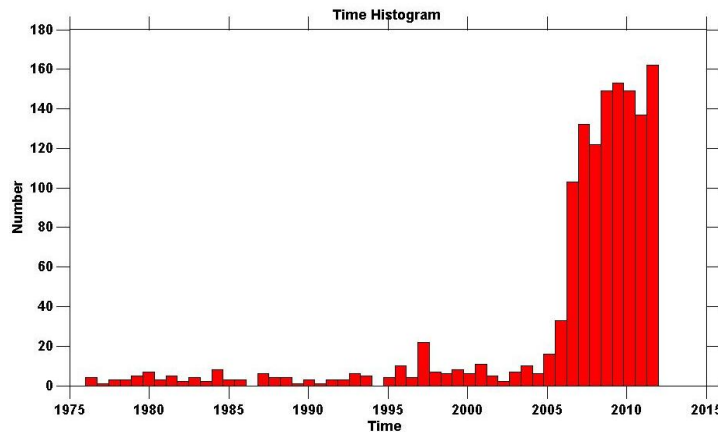


Figure3. Number of events by time (1974 to 2011)

Shi et al (1992) proposed a new method in hazard analysis, which can cover, lacks of the routine method, it is named modified probabilistic method with the following three essential steps which is showed in figure4.

- 1- Determining seismotectonic provinces and calculating seismic parameters in each province
- 2- Determining seismic potential sources , maximum magnitude (M max) for each potential source and calculation of special

- distribution function in desired magnitude interval for each source
- 3- Gridding the studied area, hazard analysis for each node by using proper attenuation

relationship and alternatively seismic zoning of the studied area.

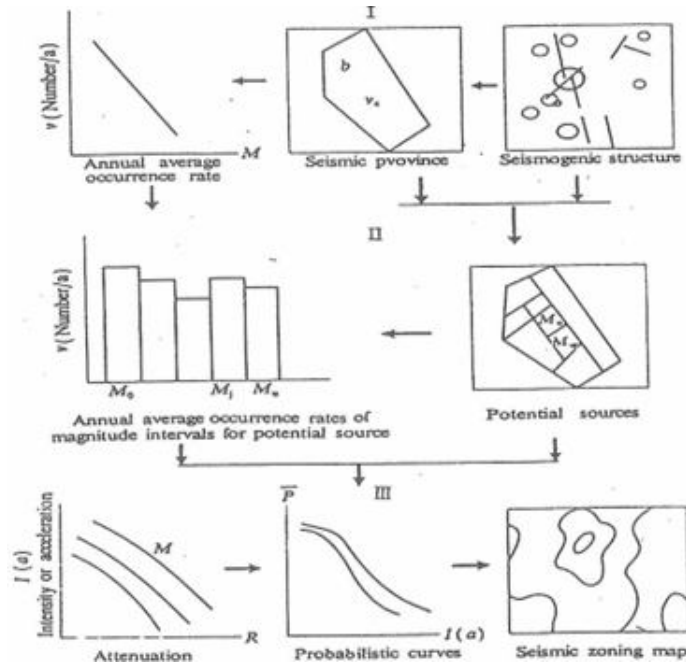


Figure4. Essential steps of modified probabilistic method (Shi et al, 1992)

Catalogue providing

The applied catalogue needs some process to be ready as an input for hazard assessment softwares, which needs the following steps.

- **Providing uniform catalogue:** To achieve a uniform catalogue a linear regression ($M_s = 1.14m_b - 0.883$) illustrates the relation between M_s and m_b in the studied area. These results are given in figure5.

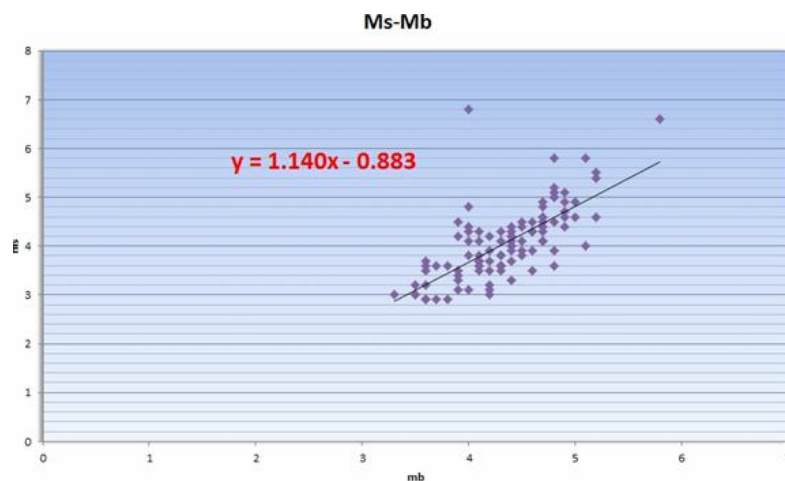


Figure5. Relationship between M_s and m_b

● **Depended events omitting:** Probabilistic seismic hazard method assume earthquakes occurrence as Poisson function. Therefore, events happen along faults or area potential sources randomly in time and space. Events location, occurrence time and their magnitude are independent from other events. Normal distribution function of probability is described as following:

$$p(x,t) = \frac{(\lambda t)^x \exp(-\lambda t)}{x!}$$

Where; λ is seismic parameter and t is desired time interval. Therefore, foreshocks and aftershocks, which are taken into account as depended events, cannot match with Poisson function and should be removed from the catalogue.

A time window and a spatial window is needed to distinguish depended events. Regarding to crucial rule of the windows dimension, Gardner and Knopoff (1974) windows has been used in the present paper.

● **Completeness calculation:** The used catalogue is belonged to long time interval, which causes heterogeneity in time and space. Step method has been applied to evaluate catalogue completeness by $n = N / \Delta N$. Where; n is average rate of events and N is cumulative number of events. In this method, calculated variance equals to $\sigma_n^2 = n / \Delta t$ and standard deviation, σ_n , is parallel to line $1 / \sqrt{\Delta t}$. In the present paper Mirzaii et al, (2000) completeness diagram is taken into account to apply Stepp's method. Table (1) shows complete beginning of catalogue completeness in different magnitude intervals for main seismotectonic provinces in Iran (Mirzaii, 1997)

Table (1). Completeness in different magnitude intervals for main seismotectonic provinces in Iran (Mirzaii, 1997)

6.0≤Ms	5.5≤Ms<6.0	5.0≤Ms<5.5	4.5≤Ms<5.0	4.0≤Ms<4.5	Seismotectonic province
1860	1900	1900	1945	1975	Alborz-Azarbayjan
1850	1904	1925	1963	1975	Kopeh-dagh
1860	1925	1944	1965	1975	Zagros
1900	1900	1955	1955	1975	Central Iran
1919	1919	1965	1965	1975	Makran

Potential seismic sources

In the present research potential sources determined by Mirzaii et al (2000) has been used but authors add some more sources regarding to the seismic activity of the area. Figure6 shows used potential sources in this study; all of them are area sources and diamonds indicate suggested potential sources by authors.

Seismic parameters

To describe seismic activity normally three parameters λ , β , M_{\max} should be determined, λ presents seismic activity rate which means number of earthquakes with magnitude equal or greater than threshold magnitude (M_0) in time period (T), b or β ($\beta = b \ln(10)$) indicates ratio of small earthquakes to

large ones. Table (2) contains calculated seismic parameters for each potential source.

Seismic hazard analysis and seismic zoning

Figure7 illustrates seismic zoning map of the studied area for 63% occurrence probability for return period of 50 years. It contains PGA contours of the area. This area will experience maximum PGA of 0.07 g and minimum PGA will be 0.012 in north east of the area.

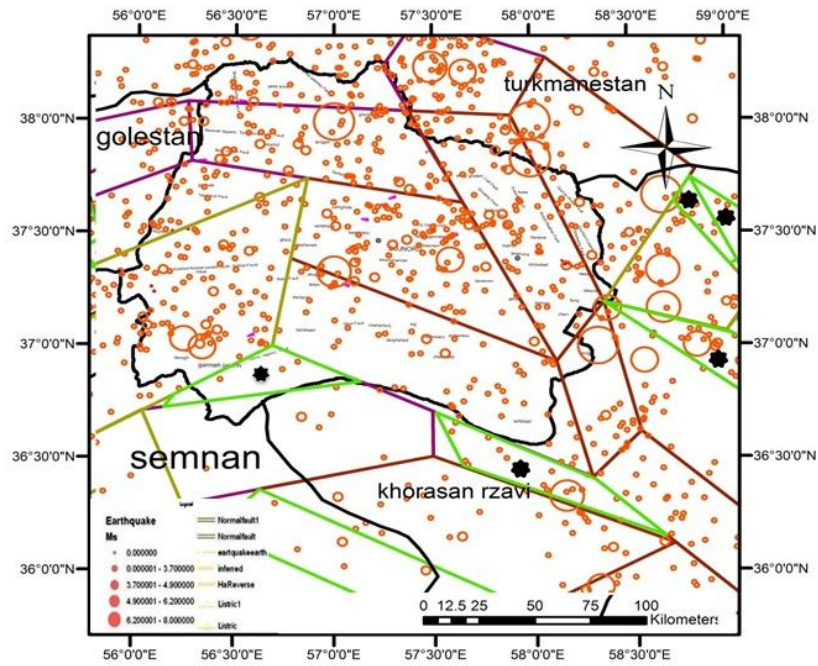


Figure6. seismic potential sources (Mirzaii etal, 2000)

Table (2). Seismic parameters in each potential source

seismic sources	number of earthquakes	LAMBDA	BETA	Mmax
1	37	0.22	2.62	4.5
2	72	0.46	2.62	7.6
3	79	0.19	2.62	6.6
4	7	0.04	2.62	3.4
5	134	0.80	2.62	5.8
6	66	0.43	2.62	4.8
7	104	0.64	2.62	7
8	76	0.39	2.62	4.9
9	57	0.35	2.62	7.2
10	10	0.17	2.62	4
11	5	0.15	2.62	2.6
12	8	0.15	2.62	4.4
13	54	0.33	2.62	4.3
14	15	0.10	2.62	7.1
15	33	0.17	2.62	6.9
16	5	0.06	2.62	2.7
17	29	0.22	2.62	5
18	42	0.31	2.62	7.6
19	35	0.20	2.62	4.6
20	1	0.06	2.62	4.4
21	1	0.04	2.62	2.3
22	19	0.11	2.62	5.3
23	16	0.15	2.62	5.2
24	4	0.06	2.62	4.5
25	1	0.06	2.62	4.5
26	12	0.09	2.62	4.5
27	0	0.09	2.62	0

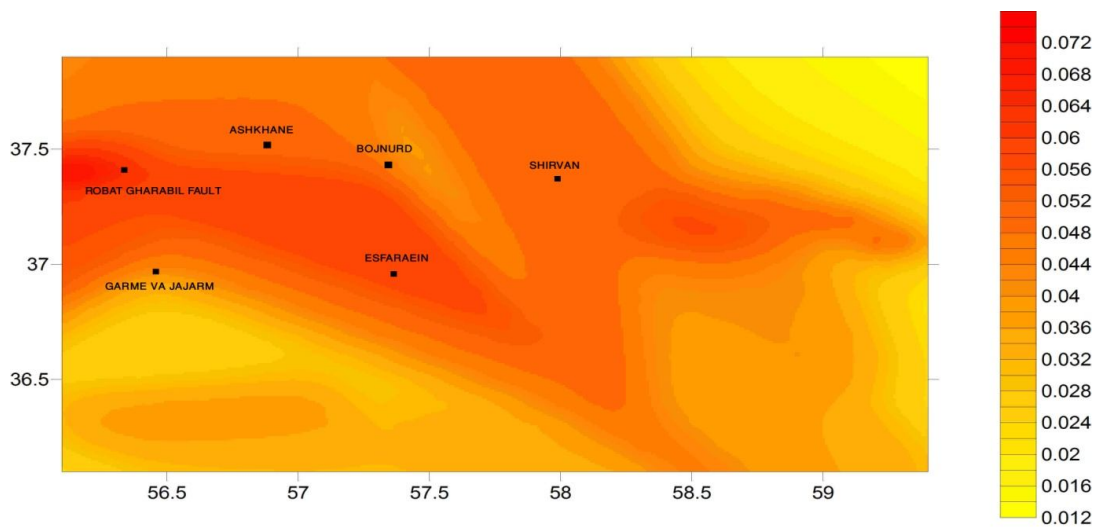


Figure7. Seismic zoning map for 63% occurrence probability in return period of 50 years

Conclusion and discussion

In the present study although different attenuation relations were taken into account the most precisely mapping obtained from Ambersays (1996) attenuation relations.

Seismic zoning map indicates that the greatest PGA will be experienced in northwest- central trend up to 0.07g, which has strong correlation with Robat Gharabi and Esfarayen faults therefore Esfarayen city, and Robatgharabil region are the most hazardous places in the studied area. Minimum PGA is estimated for south of Jajarm city up to 0.012g.

In this study area sources play role as seismic sources but seismogenic zone studied can guide us to plan volume sources, which can obtain more accurate results. Site effect should be considered in such studies to achieve risk assessment in the area.

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Information and communication technologies (ICT) in agriculture

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Abstract: Challenges which faced the early users of distance education are still with us today. Distance education is a method of education in which the learner is physically separated from the teacher and the institution sponsoring the instruction. It may be used on its own, or in conjunction with other forms of education, including face-to-face instruction. If distance education is to play a greater role in improving the quality of education, it will require expanded technology; more linkages between schools, higher education, and the private sector; and more teachers who use technology well. Teachers must be involved in planning the systems, trained to use the tools they provide, and given the flexibility to revise their teaching. Federal and state regulations will need revision to ensure a more flexible and effective use of technology. Connections have been established across geographic, instructional, and institutional boundaries which provide opportunities for collaboration and resource sharing among many groups. In the pooling of students and teachers, distance learning reconfigures the classroom which no longer is bounded by the physical space of the school, district, state or nation.

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Keywords: ICT, distance education

Introduction:

Trainers using these new technologies were successful educational programs to millions seek learning opportunities and thereby reach out to the educational spaces, training centers to expand. With the development of long-distance telephone system in the early twentieth century method of capacity and distance learning methods for students to access educational opportunities in the world increased. Translation. But until the invention of mobile tele conference ever in the 80 and 90 and the main role in the concept of distance education did not play. Telemetry system, allowing for teachers conference provided that without the slightest delay at a time when your students can listen to them talk and sometimes they see.

The background of distance education to mid-nineteenth century dates. Pioneers in America and Europe of the best distance learning technologies for training that day, took advantage. For example: mailing system for creating educational opportunities for those able to go to regular schools were not interested in science education, but had been used. Of course at that time most of those who took advantage of this type of Physically Handicapped facilities, women allowed to attend the classes along with men who did not have a. Location is N. There was a school; were. One of the pioneers in this field English personal name was Isaac Pitman. His short-term training through correspondence and the correspondence began in 1840 in England. Students were required to read the Bible a part of written questions and answers raised by Pittman to get a good score should return by mail.

But distance education in America and for the first time at the University of Illinois Veslin was implemented in 1874. In 1900, university education through correspondence, face became more public. National Association of Home Education in 1926 and led the establishment of distance education and related programs in universities and schools, and more important aspect to find drivers. Education in 1920 invented the radio and TV appearance in 1940 led to important new techniques in communications that the nature of the field of distance education also created dramatic changes.

Expansion of computer networks in the decade 1990 and connect millions of people through lines to the telephone networks made it possible to simply distance learning via computers and computer conferences around the world is possible (a) and Today with the development of control technology in science and technology around the world are.

Benefits of Distance Learning:

Benefits and opportunities that distance education provides, include:

- training a wide range of audiences.
- meet the needs of students and students who can not attend in place.
- Possible connection between students and students with cultures, beliefs and experiences are different.
- Benefiting from coaches and speakers who do not live in the country.

Remote educational tool:

distance learning tools and supplies various uses. These tools in four main courses are:

A - Audio Tools:

Audio tools include training such as two-way interactive telephone, video conference, shortwave radio and a strain of tools such as audio tape and radio.

B - Image tools:

including slides, films, video tapes and video conferences.

C - Data:

computers as electronic data are sent and received. Because the data word description for a wide range of educational tools is used.

Computer applications for distance education are varied and include the following:

- 1- Training to Computer Management.
- 2 - Computer Assisted Instruction.
- 3 - through PCs.
- 4 - e-mail, telegraph, computer conference and the World Wide Web simultaneously.

D - Print:

The main element of distance education programs, particularly in the exchange and delivery system information tools are considered.

Key factors in the process of distance education:

the process of remote training, the following factors contribute:

- Students:

Regardless of educational content, role and main element in the learning process students are responsible.

- Coaches and Teachers:

Success depends on a lot of educational activities the ability, skills and knowledge are the coaches and professors.

- Facilitators of communication:

Facilitator bases, as the bridge between students and mentors are. Must base expectations of teachers and educational needs of students and service coordination and communication to create.

- Support staff:

One of the important pillars of any development of distance education programs, by development group finds. Operational support staff such as student registration, copy and distribute their resources, order textbooks, security and copyright, and are responsible for the report.

- Management:

The group decision makers, builders and judges are considered to be educational and should be considered among the factors above, establish the correct relationship formation.

Types of Distance Education Programs:

There are two types of programs offered by distance education schools: synchronous learning programs and asynchronous learning programs. With synchronous learning, distance education students must log on to the school's website at a set time. Often, they interact with their peers and professors via group chats, web

seminars, video conferencing, and phone call-ins. With asynchronous learning, distance education students complete all coursework on their own time. They often learn via assignment sheets, message boards, email, pre-recorded video lectures, mp3s, and traditional mail correspondence.

Distance education began for the delivery of courses to students who live in remote areas. Over the years, though, this form of education has become the preferred method for learning outside of the classroom.

Distance Education is now undertaken by people with busy schedules, hectic lifestyles, special needs, and also those living in isolated areas. What's more, with such flexible learning options you can choose to study at any time and from any location you like.

There are a number of different **forms of distance education** and it's important to know which method you prefer:

- **Correspondence learning:** your course materials are printed and sent out to you by mail/courier. The advantages are that you have a printed set of reference materials, you can study anywhere and you are not reliant on a computer, you can learn for long periods of time.
- **eLearning:** your course materials are provided to you in multimedia format; that is, on CD/DVD. In this way you can choose to take your study materials within you and learn anywhere in the world with just a laptop.
- **Online learning:** no materials are sent to you and you do all your learning online. The limitation is that you need to be logged onto a computer (though you may be able to download and print some of your materials yourself, though this can cost you more in ink), there is a limit to how much you can absorb and do online, and most people's attention span on-screen is limited to 20 minutes (your eyes get tired after that).
- **Broadcast learning:** where you tune into a series of television, radio or Internet broadcasts (e.g. podcast, YouTube, etc.).
- **Teleconferencing:** where your lessons are conducted in real time through an Internet connection. Limitations are that streaming can be slow, connections can cause problems (students and teachers generally need to be computer literate) and there can be delays in talk-time, depending on software, hardware and connection capabilities.
- **Approaches to ICT diffusion:**
- The diffusion of innovations has been a focus of many research and scientific studies from diverse academic areas (Roger 2003). There were over 1500 diffusion oriented studies even during the 1950s and 1960s and research areas ranged from

anthropology, rural sociology, medical sociology to educational or mass media research (Harper 1989:111). As Charles Harper (1989:111) notices, research findings in these diverse areas have been “remarkably consistent and cumulative”.

- Diffusion theories suggest that there are several types of factors affecting the spread of innovations. Emphasizing different sets of factors, theoretical perspectives offer the ways how to analyze the dissemination of new technologies, ideas, reforms or products. The recent spread of information communication technologies in society has raised new aspects in diffusion research.
- According to Paul Attewell (1996:204), two main metaphors or images are prevalent in diffusion research. He classifies the diffusion theories into 2 main categories: (1) adopter studies, and (2) macro-diffusion theories.
- The first group implies theories which explain the patterns of innovation diffusion in relation to communication flows. The diffusion research focuses on adoption by individuals (or by single organizations) and investigates the impact of such factors as the nature of innovation, characteristics of adopters, diffusion networks and other. (Attewell 1996, Harper 1989).
- The most widespread theory of innovation diffusion is presented by Everett Rogers. According to this theory, diffusion is the process by which an innovation is communicated through certain channels over time among the members of a social system (Rogers 1983:5), thus the main 4 elements, which are identifiable in every diffusion research study, are (1) an innovation, (2) communication through certain channels, (3) time and (4) members of social system.
- The first element – innovation - is considered to be any idea, practice, or material artifact perceived to be new by the adopting organization or individual (Rogers 1983). In our case, we analyse ICT communication through certain channels among the members of rural communities.
- The potential adopters can find about new ideas just in case they are informed about them, thus the diffusion process implies the second element - communication through certain channels. According to Rogers (1983), innovations such as ICT can be transmitted to the receiver using 2 types of communication channels: (1) interpersonal channels, and (2) mass media channels.
- First type implies direct (e.g. face-to-face) communication between transmitter and receiver; and the second type includes governmental policies communicated through TV, radio, newspapers, etc. This existing discourse makes an impact on beliefs and attitudes of people toward ICT use and is one of the means for ICT diffusion among society members.
- The second group of theories - macro-diffusion theories – examines the diffusion of new technologies across entire populations, communities, society. Speed of adoption depends on such factors as population size of an area, the distance o that area from other centers of population (Attewell 1996:205).
- John Carey (1996) distinguishes marketplace factors as a separate group in diffusion research. This group includes pricing policy, replacement cycles. The price of innovation (new product, technology, service, etc.) has an important role in the process of adoption by the public. Generally new products are introduced at a high price, as early manufacturing is more expensive (due to the costs associated with the research and development of the product, low scale of production). As John Carey (1996) argues: a new technology has to find some early users who are able and willing to pay a high price for the product or service in order to achieve the economies of scale in manufacturing that can reduce the price for the general public.
- The mass production reduces the costs and the price of the product. Typical examples of such pricing policy are introduction of radio, black white and color TVs, telephone connection. The initial price of the new technology was very expensive for an average household and the technology was not widely used, but the decrease in price resulted in a wider adoption.
- But, as John Carey (1996) argues, the personal computer has followed a different pricing pattern: “rather than drop the price of personal computers, manufacturers have increased the capabilities of PCs each year”. Replacement cycles are also important. The growth of some technologies is linked to the purchase of other media. In this sense, replacement cycles for existing media can provide an important way to introduce new media. For example, in U.S. households, the average color TV is replaced after 8 years, the average telephone answering machine after 5 years and the average personal computer after 6 years (Carey 1996).
- Information technology is the core element analyzing the new, global, knowledge-based society. In today’s world the use of ICT becomes one of the most influential factors that determine both the present performance and the future conditions for the person. The Internet offers a variety of ways for interaction. Lelia Green

- (2001:197) distinguishes 3 ways of interaction: (1) information access and retrieval, (2) private interactive
- communication with individuals or small groups and (3) public interactions. But unequal opportunities to use the Internet eliminate this variety of interaction. When we talk about the impact of new information communication technologies on the society, we analyze mainly two aspects of impact– networked or socially excluded people.
 - Contemporary scientists have formulated the terms like information poor and information rich (Green 2001). An approach like this emphasizes the circumstances of people with access to minimal or large amounts of information. People who do not have or have limited access to information resources (non-haves of information or information poor) are in the social position lower than information rich. The policies based on the idea of fundamental equity are that all people should have “trouble-free access to information” and this will promote equality (Green 2001:105).
 - Of course, not everything depends on the access: “Access to technology does not necessarily lead to its use, and information does not necessarily fuel self-empowering activity” (Green 2001:105). As Lelia Green argues: access is a necessary, but by no means sufficient, condition of equitable participation. To talk simply in terms of equity of access ignores the fact that effective interaction in the information society requires high levels of motivation and sustained effort. Such keenness to interact with the technology of information cannot be assumed. Continuing motivation is perhaps the key determinant of successful participation – more important than access per se (Green 2001: 104). The diffusion of ICT and adoption in everyday life activities such as e-learning, ecommerce, e-banking, etc. are rather complicated phenomena, depending on various characteristics of an individual and a certain social system.
 - Considering the use of the Internet, it is obvious that socio-demographic characteristics determine a gap between different groups of the population. According to the data of a survey Digital Lithuania 2001, performed in the framework of a study Lithuanian Information Society, carried out by The Open Society Fund (Šaulauskas, 2001), the Internet and other information technologies are mainly used by young, educated, well paid and urban consumers. The statistical data of this survey showed that people at the age of 15 – 49, who have acquired higher education or live, or aim at living in Vilnius, Kaunas and other major cities of the country, and have high income are the most

involved in the processes of information society development (Šaulauskas, 2001).

- According to the statistical data, the lowest awareness of the processes and opportunities of information society development is among the Lithuanians over 60, who have acquired secondary or special secondary education, live in villages, rural centres or towns and have rather low income (Šaulauskas, 2001). It is obvious that different socio-demographic characteristics have determined a gap between different groups of the population. This can lead to the information gap, when one part of the population uses digital devices, while the other part of the population is in a digital divide. Thus the residents of rural communities are at the risk of being in a digital divide or even in a social exclusion.

Information Technology and its Components

Induction of IT as a strategic tool for agricultural development and welfare of rural requires that the necessary IT infrastructure is in place. The rapid changes and downward trend in prices in various components of IT makes it feasible to target at a large scale IT penetration into rural. Some of the broad factors to be noted with respect to various components of IT are listed below :

1. Input devices :

Radical improvements are witnessed with respect to the means of communication by human beings with computers such as key boards, mouse devices, scanners. The advent of touch screen monitors that allow users to give input to computers by touching on the appropriate location of the monitor has made it possible to develop user-friendly interface for farmers which is easy, intuitive, circumvents language barrier and at the same time provides a relaxed environment to the users. The present day digital cameras make it possible to capture and store good quality graphics and large video clips. The small size and low weight of these digital cameras, which are increasingly becoming affordable, open up the possibilities of providing computer based demonstration clips to educate the farmers.

2. Output devices :

Monitor screens, printers & plotters, data projectors support high resolution and good quality output. The quality of these output devices have the potential of generating renewed interest in the farmers in using IT based services. The light weight portable data projectors can be easily carried by the agricultural extension personnel for serving larger audience. Similarly, speakers can also be attached to the computers to incorporate voice based trainings for farmers.

3. Processors:

The processing speeds of computers have gone up. At present, Intel P-IV based processors @ 1.5 Ghz are available in the PC range which makes it possible to

undertake substantial processing of data at the client side.

4. Storage Devices :

40GB and even higher hard disk drives have become common in PC range of computers. This makes it possible to store substantial information at the local level which facilitates faster access. Similarly, high capacity floppy disk drives, CDs make it possible to transfer large volumes of data to locations which can not be connected to networks immediately. These storage devices are also used for backup of crucial data. As a precaution, many corporates store their backups at locations away from the place of work.

5. Software :

Various operating systems are available which act as interface between the user and the machine. The graphic user interface (GUI) has become an accepted prerequisite for end users. Microsoft's 'Windows' continues to be a favourite. Application softwares which can support complex user requirements are available. Of the shelf solutions for office automation packages, groupware applications, complex database solutions, communication products, solutions based on remote sensing & geographical information systems are available. In addition, solutions based on some or all of these are also readily available. The present downward trend in the IT industry provides an opportunity get customised application for any specific task developed at an affordable price. Rapid Application Development and Deployment (RADD) is a popular model for quick development and deployment of applications. Development environment itself is simplified with tools that quicken the pace of software specialists. Project management and monitoring software are available that facilitate efficient execution of large and complex applications that are required for rural

6. Networking devices :

The capacity of modems, used to convert the data from digital to analog and vice versa, which are popularly employed to use telephone lines have increased. Internal modems are available integrated into the computer so that they are not exposed to outside environment. The capacities of other networking devices such as routers have also gone up which makes it possible to create large networks with smooth data transmission.

7. Transmission Media:

The media through which the data transfer takes place has also undergone revolutionary change. Telephone lines are still the popular source although the reliability and low bandwidth are still major issues. High capacity cables, optical fibre, radio, wireless local loops, satellite transmission and various solutions based on a combination of these are already being used in many parts of the country.

8. Other accessories :

Uninterrupted Power Supply (UPS) devices are crucial to ensure the longevity of the IT equipment as well as provide backup mechanisms. The potential of solar power packs to provide a feasible solution to shortage of power in the rural areas needs to be exploited.

Conclusion:

Technology transports information, not people. Distances between teachers and students are bridged with an array of familiar technology as well as new information age equipment. What sets today's distance education efforts apart from previous efforts is the possibility of an interactive capacity that provides learner and teacher with needed feedback, including the opportunity to dialogue, clarify, or assess. Advances in digital compression technology may greatly expand the number of channels that can be sent over any transmission medium, doubling or even tripling channel capacity. Technologies for learning at a distance are also enlarging our definition of how students learn, where they learn, and who teaches them. No one technology is best for all situations and applications. Different technologies have different capabilities and limitations, and effective implementation will depend on matching technological capabilities to education needs.

Distance education places students and their instructors in separate locations using some form of technology to communicate and interact. The student may be located in the classroom, home, office or learning center. The instructor may be located in a media classroom, studio, office or home.

The student may receive information via satellite, microwave, or fiber optic cable, television (broadcast, cable or Instructional Television Fixed Services (ITFS), video cassette or disk, telephone - audio conferencing bridge or direct phone line, audio cassette, printed materials - text, study guide, or handout, computer - modem or floppy disk, and compressed video. Recent rapid development of technology has resulted in systems that are powerful, flexible, and increasingly affordable. The base of available information technology resources is increasing with dramatic speed. Much has been learned about connecting various forms of technology into systems, so that the ability to link systems is growing. Most distance learning systems are hybrids, combining several technologies, such as satellite, ITFS, microwave, cable, fiber optic, and computer connections.

Interactivity is accomplished via telephone (one-way video and two-way audio), two-way video or graphics interactivity, two-way computer hookups, two-way audio. Interactivity may be delayed but interaction provided by teacher telephone office hours when students can call or through time with on-site facilitators. Classes with large numbers of students have

a limited amount of interactivity. Much of the activity on computer networks is on a delayed basis as well. Possibilities for audio and visual interaction are increasingly wide.

In the earlier days of distance learning, it was most common to see distance learning used for rural students who were at a distance from an educational institution. The student might watch a telecourse on a television stations, read texts, mail in assignments and then travel to the local college to take an exam. This model is still in use, but as the technology has become more sophisticated and the cost of distance learning dropped as equipment prices dropped, the use of distance education has increased.

High front-end costs prevented an early widespread adoption of electronically mediated learning. Distance learning has been aggressively adopted in many areas because it can meet specific educational needs. As the concept of accountability became accepted and laws required certain courses in high school in order for students to be admitted to state colleges, telecommunications was examined as a way to provide student access to the required courses. Many rural school districts could not afford the special teachers to conduct required courses. Distance education met this need by providing courses in schools where teachers were not available or were too costly to provide for a few students. It also fulfilled a need for teacher training and staff development in locations where experts and resources were difficult to obtain. These systems link learner communities with each other and bring a wide array of experts and information to the classroom.

Challenges which faced the early users of distance education are still with us today. If distance education is to play a greater role in improving the quality of education, it will require expanded technology; more linkages between schools, higher education, and the private sector; and more teachers who use technology well. Teachers must be involved in planning the systems, trained to use the tools they provide, and given the flexibility to revise their teaching. Federal and state regulations will need revision to ensure a more flexible and effective use of technology. Connections have been established across geographic, instructional, and institutional boundaries which provide opportunities for collaboration and resource sharing among many groups. In the pooling of students and teachers, distance learning reconfigures the classroom which no longer is bounded by the physical space of the school, district, state or nation.

The key to success in distance learning is the teacher. If the teacher is good, the technology can become almost transparent. No technology can overcome poor teaching which is actually exacerbated in distance education applications. When skilled

teachers are involved, enthusiasm, expertise, and creative use of the media can enrich students beyond the four walls of their classroom.

The interchange of ideas requires different communication methods than in conventional classrooms: information technologies are predominantly visual media, rather than the textual and auditory environment of the conventional classroom, the affective content of mediated messages is muted compared to face-to-face interaction, and complex cognitive content can be conveyed more readily in electronic form because multiple representations of material (e.g., animations, text, verbal descriptions, and visual images) can be presented to give learners many ways of understanding the fundamental concept.

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Factors Facilitating Entrepreneurship and Self- Employment in Agriculture Sector

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Abstract: The main purpose of the study was to analyze factors which facilitate entrepreneurship and self- employment in agriculture sector. To achieve this, a surveying methodology was used. Statistical population of the study consisted of all agricultural students in Bu- Ali Sina University. Among them, 122 were selected randomly as samples. Data gathering tool was a structured questionnaire. The face validity of the questionnaire was verified by a panel of experts as well as reliability of the questionnaire was tested by Cronbach's Alpha coefficient and it was 87 percent. In the data analysis part of this study, factor analysis method was used in order to achieve key elements and factors. Results showed that factors which facilitate entrepreneurship and self- employment in agriculture sector could be classified into four categories. The four-factor presented by the factor analysis method explained 44.7% of the total variance. The first category of the factors was labeled as educational factor. This factor explained 15.13% of the total variance and was identified the most important element. At the end, some recommendations have been presented in regard to develop entrepreneurship and self- employment in agriculture sector.

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Keywords: Entrepreneurship, Agriculture Sector, Agricultural Student, Facilitating Factors.

Introduction

In recent years, a special attention is paid to entrepreneurship and entrepreneur in different countries and most scholars believe that entrepreneurship and entrepreneurs have a considerable role in economic growth and development especially in developing countries (Rahmati & et al, 2010). In such a way that entrepreneurship is considered one of the main factors in competitive markets and entrepreneur companies response the environmental challenges better and faster compared to other companies (Jeemens, 2008). Entrepreneurship

plays also an important role in social development of different societies. From the economic viewpoint, entrepreneurship referred to the production factors that assists the economic growth through discovering and creating new opportunities and from social viewpoint, contributes the improvement of social situations through increasing job or employment chances (Mohapatra & et al, 2007). The category of entrepreneurship in most developed countries and all developing ones has been considered as the most basic source of development (Macline & Richard, 2004). In agricultural sector, entrepreneurship

considered as a complimentary section in economy and job-creating activities due to response or react to the stagnancy and problems available in agricultural sector and also as a survival strategy for rural families (Petrin, 1994, Smits, 2004). Therefore, the development of entrepreneurship in agricultural sector can act as a crucial force for the development of this sector (Sharifi & et al, 2009). In general, entrepreneurship is a powerful tool for identifying opportunities, acquiring benefits, and removing such problems as unemployment, the lack of dynamic human resources, low efficiency, quality reduction of products and services, and economic stagnancy (Maclin & Richard, 2004). With regard to the importance and contribution of entrepreneurship, it is essential that entrepreneurship and entrepreneurs in all sectors including agricultural sector be supported and developed. Agricultural sector is one of the major economic sectors of the country that plays an important role in economic development of country due to its high potential and special properties. However, in this sector the entrepreneurship trend is slow and complex, due to natural and climatic conditions and undesirable quality of resources. It is also expected more limitations concern to employment in agriculture. The average cost of creating employment in agricultural sector is considerably lower than that of industry sector. From viewpoint of foreign exchange investment, employment in agriculture is considered a relative advantage for this sector especially in case of shortage of foreign exchange (Zahedi & Kazerooni, 2000). Thus creating employment in agricultural sector through entrepreneurship should be planned as a strategic policy by the planners and policy makers In Iran. Accordingly, this study was an attempt to investigate the facilitating factors in developing entrepreneurship and self-employment in agricultural sector.

Literature Review

The challenge of employment or the issue of unemployment is not only one of the most

important social problems in our country but also is considered the most important social challenge of the next few decades due to the rate of population growth in the country. In this vein, one of the basic and important tools for the governments can be the development of entrepreneurship. Agricultural sector as one of the important economic sectors can assist the policymakers take proper decisions against the challenge of employment in the country. In this regard, Noroozadeh and Mehrabiyeganeh (2006) state that the development of entrepreneurship in applied and scientific higher education centers of agricultural is one of the efficient methods in increasing efficiency of human resources in agricultural sector. In this area, numerous investigations have been conducted. Below some of the most important empirical studies will be dealt with.

Khosravipoor and his colleagues (2007) in a case study about identifying influential factors in empowering entrepreneurship for Agricultural Scientific- Applied Instruction Centers presented some important indices. These were necessity of changing instructional methods, necessity of changing and improving educational contents, necessity of equipping educational centers, necessity of changing the students' enrollment and selection, and necessity of improving the organization and management system of the Agricultural Scientific- Applied Instruction Centers. Yaghubi (2008) in his research found a set of ways in order to promote entrepreneurship in agriculture sector include an especial support of the entrepreneurs, eliminate bribe and lawlessness, attracting the participation of entrepreneurship in economic activities, giving more authority and facilities for entrepreneurship activities, creating competition in production and marketing and proving entrepreneurship insurance. The findings of Jahanian investigation (2010) showed that the most important solutions for developing entrepreneurship in technical and vocational centers of Islamic Azad universities include creating entrepreneurship culture, entrepreneurship management, developing entrepreneurship instruction, creating entrepreneurship structure, developing

continuous learning, providing financial sources for entrepreneurship, and developing entrepreneurship through creative and innovative human forces. Tybjee and Vickery (1998) mentioned that the development of entrepreneurship is affected by the trends and policies of government, social competitions and economic situation. Klapper et al (2004) believe that governmental and economical laws and instructions influence the individual's tendency toward entrepreneurship. Shane et al (2003) believe that the motivation of individuals is an important factor in making decision about entrepreneurship activities. They stated that motivation is like a road map which helps investigators to choose the right route and to make the correct decisions in entrepreneurship process. The study by Jones and English (2005) revealed that we could increase the knowledge, skills and self-confidence of student through entrepreneurship instruction and empower them toward creating jobs of their own. Peak and Marshall (2006) emphasized the importance of education as the most important human wealth and the results of their study showed that the higher level education has a great effect on the performance of an entrepreneurship institute. Urbano et al (2011) studied the social and cultural factors effecting the development of entrepreneurship in Spain and came to the conclusion that tendency toward entrepreneurship is one of the most important and influential factors in emerging entrepreneurship behavior.

Method

Present study aimed at to investigate the facilitating factors for entrepreneurship in agriculture sector through using a surveying methodology. In this method by sampling and studying selected samples from the target community, the distribution, frequencies and the relationship between variables have been studied. In this regard, the most common method for data collection is questionnaire. The accuracy of indices and variables in the questionnaire or face validity has been confirmed by a group of experts. In order to determine the reliability of the questionnaire, a pre-test process was used through filling the

questionnaires by 25 students and a Cronbach's Alpha test was used through SPSS software and it was 87% which shows an acceptable rate. The statistical population of this study was the agricultural students of Bu-Ali Sina University during 2011- 2012 academic year. A number of 122 samples were selected randomly and their attitudes towards factors facilitating entrepreneurship and self-employment in agriculture sector were reviewed. In this research both descriptive and analytic methods were used. In descriptive part, to describe the variables, such statistics as frequency, percentage, standard deviation, variance, and mean were used. In analysis section, factor analysis was used to access the key factors affecting the entrepreneurship.

Results

In descriptive part of the results, personal status of the respondents was described. From the results the respondents' age was 24 years. About the gender's status of the students, findings showed that 51% were female and 49% male. Most of the students were at undergraduate level and only four percent were studying at postgraduate. According to the results, 84% of the target students rated their ability to establish an independent business in a level of high or average. This shows that there is a high level of entrepreneurship's capacity among the students.

As mentioned later, a factor analysis method was used in order to classify factors facilitating the entrepreneurship in agriculture sector. To determine whether or not the variables are usable for factor analysis, Kaiser-Meyer-Olkin (KMO) and Bartlett's tests is usually used. The KMO test was used to measure the sampling adequacy and Bartlett's Test of sphericity to determine the identity of the correlation matrix. If the KMO value is less than 0.5 the data will not be suitable for factor analysis if the value is greater than 0.7 the correlations between data will be appropriate for factor analysis (Kalantari, 2003). In this study, the Kaiser-Meyer-Olkin value (KMO=0.765) indicated an

adequate sampling for the factor analysis and the result of Bartlett's test of Sphericity was significant ($p < .01$).

As shown in table 1, in the first round of the factor analysis, four factors were extracted.

these extracted factors had the highest amount of overlap (in terms of calculated variance). This means these factors can explain more than 44 % of the total discrepancy among the variables.

Table 1: Number of component, initial Eigen values, % of variance and cumulative percent

Factors	initial Eigen value	% variance	of cumulative percent
First factor	4.238	15.137	15.137
Second factor	3.321	11.861	26.997
Third factor	2.619	9.352	36.349
Fourth factor	2.338	8.351	44.701

Table (1) shows that the first factor (initial Eigen value =4.23) has the highest share and the fourth factor (initial Eigen value =2.33) has the lowest role in presenting variables. In general, four mentioned factors were able to explain 44.7 percent of the total variance. In table 2, the results from rotated matrix of the variables have been shown. The Eigen values, the numbers of extracted factors from rotated matrix, and each of variables have been also illustrated.

Table 2: The results from rotated matrix of the variables

Factor	Variable	Eigen value
Educational factor	Establishing growth and entrepreneurship centers in universities	0.518
	Creating skills in agriculture students	0.658
	Reducing costs of starting and opening new businesses	0.547
	Enhancing practical skills of agriculture students	0.632
	Increasing the number of practical courses	0.522
	Changing curricula and new methods of teaching	0.528
	Making the academic contents tailor to the labor market demands	0.671
	Having the ability and practical skills in agriculture	0.727
Financial and legal factors	Financial supporting of entrepreneurship in agricultural sector	0.534
	Providing bank's facilities for the agricultural graduates	0.594
	Reforming of legislation related to entrepreneurship in agriculture	0.566
	Removing administrative barriers of entrepreneurship	0.734
	Reforming tax legislation related to entrepreneurs	0.538
	Providing the necessary infrastructure in the agricultural sector	0.513
Infrastructural factors	increasing job opportunities in the public sector	0.502
	Access to facilities and financial resources for starting-up a new business	0.562
	Providing marketing facilities for agricultural products	0.611
	Introducing successful entrepreneurs to agricultural graduates	0.714
Cultural Factors	Media advertising about entrepreneurship	0.812
	Strengthening the entrepreneurial culture in the community	0.693
	Developing and strengthening Counseling entrepreneurship centers in the agricultural	0.614

First Factor-Educational factor: variables included in the first factor are as following: Establishing growth and entrepreneurship centers in universities, creating skills in agriculture students, reducing costs of starting, and opening new businesses, improving practical skills of agriculture students, increasing the number of practical courses and content appropriateness whit labor market demands and change in subjects plans and new method of teaching. Thus this factor was called educational factor and its specific amount was 4.23 which is greater than all other factors. This factor has the greatest effect and importance in expressing variables and in general, accounts for 15.13Percent of total variance of variables. Hence, in regard whit developing entrepreneurship in agriculture sector, educational factor more than any other factor functions as a facilitating factor. Findings of study by Heidari (2003), khosravipour et al (2007), Alambeigi et al (2009), Yaghoubi (2010), Tybjee and Vickery (1998), and Peak and Marshall (2006) confirmed the results of present study.

Second Factor- Financial and legal factors: variables included in the second factor are as following: The financial support of entrepreneurship in agricultural sector, provide banking facilities to the agricultural graduates, reform of legislation related to entrepreneurship in agriculture, reduction administrative barriers of entrepreneurship, reform tax legislation related to entrepreneurs and providing the necessary infrastructure in the agricultural sector. According to table 1, specific amount of this factor was 3.321. This factor accounts for 11.861 Percent of total variance of variables. Findings of study by Yaghoubi (2010), Jahanian (2010) and Klapper et al (2004) confirmed the results of present study.

Third factor- Infrastructural factors: variables included in the second factor are as following: Lack of job opportunities in the public sector, access to facilities and financial resources for starting a new business, facilitates marketing of agricultural products and introducing Successful entrepreneurs to agricultural graduates. This factor was called

Infrastructural factor and its specific amount was 2.61. In general, accounts for 9.35 Percent of total variance of variables. Findings of study by Yaghoubi (2010) confirmed the results of present study.

Fourth factor- Cultural Factors: variables included in the second factor are as following: Positive publicity in the media towards entrepreneurship, strengthen the entrepreneurial culture in the community and develop and strengthening counseling entrepreneurship centers in the agricultural. This factor was called **cultural factor** and its specific amount was 2.33. In general, accounts for 8.35 Percent of total variance of variables. Findings of study by Jones and English (2005), Jahanian (2010) and Urbano et al (2011) confirmed the results of present study.

Conclusion and recommendations

Graduates' unemployment in agriculture sector has become as one of the basic challenges in the agriculture sector and this problem has engaged the policymakers and all stakeholders' organizations in order to find proper solutions in this regard. In this vein, entrepreneurship is considered as one of the most important solutions that can be as an appropriate attempt to create new employments and therefore this troubleshoot must be as the highest priority in the governmental policies. In recent decades, the issue of entrepreneurship and its development has created a series of entrepreneurship development policies. In this study, the factors facilitating entrepreneurship in agriculture sector was analyzed. The results of this analysis showed that, in general, four factors include educational, financial and legal, infrastructural, and cultural factors were identified as the most important factors in facilitating entrepreneurship. Thus, it is recommended that easy access to credits, granting low-interest loans to the students and graduates of agriculture field are of priority in government policies in order to accelerate and develop entrepreneurship in agriculture sector. Also, revising available laws and infrastructures could pave the way for

easy arrival of students and graduates in the field entrepreneurship and self-employment. In addition, given the importance of practical skills and the weakness of these skills in the graduates of agriculture field, it is recommended that the teaching methods of practical and skilled subjects be revised and more interaction and cooperation be exercised among universities and other administrative organizations involved in this field. Also, while increasing practical courses and defining related objectives, desired presentation of practical courses should be accomplished.

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The role of information and communication technologies (ICT) in rural development

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Abstract: The concept of development of the rural, today, is not just project initiatives and governance; it is much more beyond that. This paper uncovers a whole plethora of ICT emergence as a technology of the new millennium. Against the backdrop of the ongoing ICT boom, this paper makes an attempt towards studying its applications and usage planning process and policy making for the rural communities focusing on how it helps in aligning the key factors and reduce the problems of alienation, fragmentation and dislocation of knowledge. Review of literature shows that intervention of information and communication technologies (ICT) in rural development initiatives are capable of development, but are not successful. Lack of community participation, absence of an integrated approach and non-inclusion of traditional knowledge systems (TKS) in the project designs are the major impediments. We therefore suggest a systems-based approach in the design of e-Governance projects, and brief some future directions. Sustained development using rural informatics is possible, only if ICT interventions are able to respond to the local needs and re-adjust as per the prevailing knowledge (Traditional Knowledge Systems- TKS) of the rural areas.

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Keywords: information and communication technologies (ICT), rural development

Introduction:

Direct or indirect application of ICT, in rural development sector has also been referred to as "Rural Informatics". Rural economies can be benefited from ICT by focusing on social production, social consumption and social services in the rural areas (Malhotra, 2001). The inculcation of a Citizen-to-Government (C2G) and Citizen-to-Citizen (C2C) interface would provide this link that would also lead to community participation in design and implementation of ICT interventions. This in return could promise better economic opportunities as well as social inclusion of rural people in the processes of governance. Such attributes in the social set up are essential prerequisites for good governance and rural development.

Globalization and technological changes, the processes in the past fifteen years have been quickly lead to a new global economy have been driven with the reinforced technology and fuel (energy) that by providing information and knowledge.

The global economy requires the kind of necessity and purpose of educational institutions. Since the current trend towards reducing incomplete information and access to accurate information is growing, other schools can not control time to transfer a set of prescribed information from teacher to student during a fixed time point are, but schools must to promote Culture of "Teaching for Learning For example, acquisition of knowledge and continuous learning skills which make possible during the individual's life. According to Alvin Toffler, illiterate in 21st century, who was not read and write but those who do not know

which fail to learn or remember are illiterate. (Jauhari, 2004).

Concerns about educational quality and educational opportunities with the necessity of developing those most vulnerable are the accumulation of globalization is symbiotic. Generally, "the changes of globalization in developing countries, on low-income groups, especially women and girls and" low skill workers, as well as all groups applying for and obtaining new skills to press. (Bellamy and Taylor, 1998).

In the rural context, development involves use of physical, financial and human resources for economic growth and social development of the rural economies (Burkey, 2000). The term rural development also represents improvement in quality of life of rural people in villages. As per Chambers (1983) "Rural Development is a strategy to enable a specific group of people, poor rural women and men, to gain for themselves and their children more of what they want and need." Singh (1999) defines Rural Development as "A process leading to sustainable improvement in the quality of life of rural people, especially the poor". The fact of the matter is that three quarters of the world's poor, about 900 million people are in rural areas, and the Millennium poverty target set by Millennium Development Goals (MDG), cannot be met unless the world addresses rural poverty. "Sustainable Rural Development can make a powerful contribution to four critical goals of: Poverty Reduction, Wider shared growth, Household, national, and global food security and Sustainable natural resource management" (World Bank, 1997). Hence worldwide there is a growing

emphasis on development of rural economy of the countries. Any improvement, in the social or economic status of rural areas would not just directly benefit rural poor but would also bring down the migration-pressures on cities and contribute by positive ripple effect in global stride towards development.

The process of development in a country is to be aided by its governance. The goal of governance "should be to develop capacities that are needed to realize development that gives priority to the poor, and creates needed opportunities for employment and other livelihoods" (The World Bank, 1992, UNDP, 1994). Increased number of poor, hungry or marginalized people in a country represents decrease in its quality of governance. To promote development, various studies have proposed governance in the contextual realities of each country, including veritable participation of citizens in the governmental decision-making process (Grindle, 2004; Evans and David, 2006). Several Institutions and experts accept Governance as a reflexive process, wherein policies, institutions, outcomes and analysis interact, to maximize the process of participatory development (UNDP, 1997; Ludden, 2005; Mehta, 2006).

Information and communication technologies (ICT), including radio and television and the newer digital technologies like computers and the Internet as potentially are introduced powerful tools and activators of educational reform and changes. different ICT, when properly applied can be developed to help access to education and the relationship between training and workshops to strengthen the increasingly digital, the quality of education also helped to create teaching and learning in an active process connected to real life high take. However, the experience of being raised by ICT in the classroom and other educational sites around the world during the last few decades proves that is not automatic fully realize the potential benefits of ICT training. (Guptaand et al, 2004)

Effective integration of ICT in the educational system is a complex process that involves not only technology but also involves educational and technical training, institutional readiness, teacher competencies and long-term investment. In fact the subject of such vital importance is that the technology to get the easiest part of it. Introduced ICT information and communication technologies are for this purpose, as a different set of tools and technology resources, used to information communicate, create, release, storage and management have been defined. These technologies are including computers, internet, broadcasting technologies (radio and television) and telephone. In recent years started a wave of intense public interest about how computers and the Internet can become a better control to the efficiency and effect of education at

all levels and in both formal and informal development. (Rogers and Shukla, 2001).

But nowadays, ICT is more than a technology. Although the old technologies such as telephone, radio and television, will be less attention in the past but were used as educational tools. For example, "radio and television are used for over forty years to open and distance education. In this regard, although print remains the most expensive method and therefore available, but in developed and developing countries is provided the most prominent mechanism.

ICT for developing countries, are associated a potential for increased availability and quality of training and development. ICT basis and attract a lot of knowledge and its acquisition, providing unprecedented opportunities for developing countries, adding and expanding educational systems, improve policy formulation and implementation of opportunities to expand scope of work and gives poor facilitation. One of the biggest hardships that the poor are bearing the other people, who live in the poorest countries, is the sense of isolation. Communication technologies such sensory loss, are guaranteed and also has been unthinkable facilitate access to knowledge through the ways that already. However, the reality of the digital divide (the gap between those who control access technology and those who do not have access) means that the introduction and integration of ICT are challenging at different levels and in various types of training, most commitments. Failure in this struggle to become more significant gap of knowledge and the deepening economic and social inequalities (UNDP, 1997; Ludden, 2005; Mehta, 2006).

How ICT can help developing access to education?

ICT is a potentially powerful tool for developing educational opportunities, whether formal or informal is for areas already "stated (rural and dispersed populations) ethnic minorities, women, girls, disabled and old people traditionally excluded from education groups because of cultural or social reasons are also all those financial reasons or time constraints can not register in educational centers. Any time, anywhere (defining feature ICT) capability in ICT is a passing of time and place. ICT, education or training with asynchronous features provide a time delay between education and its acceptance by students makes it possible.

ICT applications in education:

Organizations and educational policy planners should first of all about the desired educational outcomes (mentioned above) is straightforward. The broad objectives must choose different technologies used to go and how to apply the guidance to go. Potential of each technology varies according to how to use. Haddad and Draxler have been identified IT application in education at least five levels of:

1. present,
2. experimental proof,
3. practice and practice,
4. interaction,
5. collaboration

Each of the different ICT tools (print, audio cassette and video, broadcast radio and television, computer or Internet) may provide the most basic means and surfaces used to go to prove. Except for visual technology, practice and practice the maximum use of both technologies may be offered. Each of the different ICT (print, audio cassette and video, broadcast radio and television, computer or Internet) used to may provide the most basic means and surfaces. Except for visual technology, may be offered practice and practice the maximum use of both technologies. The other network computers and Internet, ICT interactive learning that are provided and they if only used for providing proof or go, was not realized can better their full potential (Jauhari, 2004)

Approaches to ICT diffusion:

The diffusion of innovations has been a focus of many research and scientific studies from diverse academic areas (Roger 2003). There were over 1500 diffusion oriented studies even during the 1950s and 1960s and research areas ranged from anthropology, rural sociology, medical sociology to educational or mass media research (Harper 1989:111). As Charles Harper (1989:111) notices, research findings in these diverse areas have been “remarkably consistent and cumulative”.

Diffusion theories suggest that there are several types of factors affecting the spread of innovations. Emphasizing different sets of factors, theoretical perspectives offer the ways how to analyze the dissemination of new technologies, ideas, reforms or products. The recent spread of information communication technologies in society has raised new aspects in diffusion research.

According to Paul Attewell (1996:204), two main metaphors or images are prevalent in diffusion research. He classifies the diffusion theories into 2 main categories: (1) adopter studies, and (2) macro-diffusion theories.

The first group implies theories which explain the patterns of innovation diffusion in relation to communication flows. The diffusion research focuses on adoption by individuals (or by single organizations) and investigates the impact of such factors as the nature of innovation, characteristics of adopters, diffusion networks and other. (Attewell 1996, Harper 1989).

The most widespread theory of innovation diffusion is presented by Everett Rogers. According to this theory, diffusion is the process by which an innovation is communicated through certain channels over time among the members of a social system (Rogers 1983:5),

thus the main 4 elements, which are identifiable in every diffusion research study, are (1) an innovation, (2) communication through certain channels, (3) time and (4) members of social system.

The first element – innovation - is considered to be any idea, practice, or material artifact perceived to be new by the adopting organization or individual (Rogers 1983). In our case, we analyse ICT communication through certain channels among the members of rural communities.

The potential adopters can find about new ideas just in case they are informed about them, thus the diffusion process implies the second element - communication through certain channels. According to Rogers (1983), innovations such as ICT can be transmitted to the receiver using 2 types of communication channels: (1) interpersonal channels, and (2) mass media channels.

First type implies direct (e.g. face-to-face) communication between transmitter and receiver; and the second type includes governmental policies communicated through TV, radio, newspapers, etc. This existing discourse makes an impact on beliefs and attitudes of people toward ICT use and is one of the means for ICT diffusion among society members.

The second group of theories - macro-diffusion theories – examines the diffusion of new technologies across entire populations, communities, society. Speed of adoption depends on such factors as population size of an area, the distance of that area from other centers of population (Attewell 1996:205).

John Carey (1996) distinguishes marketplace factors as a separate group in diffusion research. This group includes pricing policy, replacement cycles. The price of innovation (new product, technology, service, etc.) has an important role in the process of adoption by the public. Generally new products are introduced at a high price, as early manufacturing is more expensive (due to the costs associated with the research and development of the product, low scale of production). As John Carey (1996) argues: a new technology has to find some early users who are able and willing to pay a high price for the product or service in order to achieve the economies of scale in manufacturing that can reduce the price for the general public.

The mass production reduces the costs and the price of the product. Typical examples of such pricing policy are introduction of radio, black white and color TVs, telephone connection. The initial price of the new technology was very expensive for an average household and the technology was not widely used, but the decrease in price resulted in a wider adoption.

But, as John Carey (1996) argues, the personal computer has followed a different pricing pattern: “rather than drop the price of personal computers, manufacturers have increased the capabilities of PCs each year”. Replacement cycles are also important. The

growth of some technologies is linked to the purchase of other media. In this sense, replacement cycles for existing media can provide an important way to introduce new media. For example, in U.S. households, the average color TV is replaced after 8 years, the average telephone answering machine after 5 years and the average personal computer after 6 years (Carey 1996).

Information technology is the core element analyzing the new, global, knowledge-based society. In today's world the use of ICT becomes one of the most influential factors that determine both the present performance and the future conditions for the person. The Internet offers a variety of ways for interaction. Lelia Green (2001:197) distinguishes 3 ways of interaction: (1) information access and retrieval, (2) private interactive communication with individuals or small groups and (3) public interactions. But unequal opportunities to use the Internet eliminate this variety of interaction. When we talk about the impact of new information communication technologies on the society, we analyze mainly two aspects of impact– networked or socially excluded people.

Contemporary scientists have formulated the terms like information poor and information rich (Green 2001). An approach like this emphasizes the circumstances of people with access to minimal or large amounts of information. People who do not have or have limited access to information resources (non-haves of information or information poor) are in the social position lower than information rich. The policies based on the idea of fundamental equity are that all people should have “trouble-free access to information” and this will promote equality (Green 2001:105).

Of course, not everything depends on the access: “Access to technology does not necessarily lead to its use, and information does not necessarily fuel self-empowering activity” (Green 2001:105). As Lelia Green argues: access is a necessary, but by no means sufficient, condition of equitable participation. To talk simply in terms of equity of access ignores the fact that effective interaction in the information society requires high levels of motivation and sustained effort. Such keenness to interact with the technology of information cannot be assumed. Continuing motivation is perhaps the key determinant of successful participation – more important than access per se (Green 2001: 104). The diffusion of ICT and adoption in everyday life activities such as e-learning, ecommerce, e-banking, etc. are rather complicated phenomena, depending on various characteristics of an individual and a certain social system.

Considering the use of the Internet, it is obvious that socio-demographic characteristics determine a gap between different groups of the population. According to the data of a survey Digital Lithuania 2001,

performed in the framework of a study Lithuanian Information Society, carried out by The Open Society Fund (Šaulauskas, 2001), the Internet and other information technologies are mainly used by young, educated, well paid and urban consumers. The statistical data of this survey showed that people at the age of 15 – 49, who have acquired higher education or live, or aim at living in Vilnius, Kaunas and other major cities of the country, and have high income are the most involved in the processes of information society development (Šaulauskas, 2001).

According to the statistical data, the lowest awareness of the processes and opportunities of information society development is among the Lithuanians over 60, who have acquired secondary or special secondary education, live in villages, rural centres or towns and have rather low income (Šaulauskas, 2001). It is obvious that different socio-demographic characteristics have determined a gap between different groups of the population. This can lead to the information gap, when one part of the population uses digital devices, while the other part of the population is in a digital divide. Thus the residents of rural communities are at the risk of being in a digital divide or even in a social exclusion.

Problems faced is that:

- The population of the earth is burgeoning every minute and there is sufficient evidence of impending food crisis, especially in the developing countries even after attaining self-sufficiency.
- Even the major powers in the world are finding it difficult to balance the agricultural productivity with the environmental requirement and meet the expectation of the millions round the world.
- The politics and economics in any country and the world trade mechanism are now dependent on the balance of supply and demand of the food.
- Inefficient recording and storage of data in spite of huge data collection.
- Lack of timely forecasting of weather and agriculture productivity.

In the current scenario, the role of IT assumes great importance and only with proper integration of IT with agriculture, the problem of food crisis can be solved and the world can move towards a sustainable production.

Integration of IT with agriculture must be done with following main objectives in mind:

- Develop multi-level decision support models for synergising the natural resource system with economic and social imperatives.
- To develop indicators of sustainability for agricultural production system.

- Based on the above scientific assessment, suggest alternatives to conserve and improve the health of natural resource system.

Two fundamental steps exist in establishing an innovation as a valuable, readily used tool: diffusion and adoption. Both diffusion and adoption must occur in order for an innovation to successfully reach its target user and be implemented (Mahajan, et al.). First, diffusion, the process by which an innovation “is communicated through certain channels over time among the members of a social system” (Rogers), must occur. In this study, the StratSoy project was a major factor in the IT diffusion process in state soybean organizations. Other factors that influenced diffusion included the media, word-of-mouth, and experiences of friends, associates and family members.

In addition to individuals having access to a new technology, adoption must also occur, which means individuals accept the innovation as valuable and use it. Numerous factors could influence IT adoption and use in agricultural organizations and can be grouped into five categories: access to IT, demographic, IT training/education, trust, and time. It is possible for adoption factors to fit into more than one category.

In the case of IT, access to the technology means an individual must have access to a computer equipped with IT such as e-mail and access to the WWW. The category “access to IT” would not only include the use of a computer with IT ability, but would also include the ability to upgrade computer hardware and software to facilitate IT use. The price of needed computer equipment and the expense of Internet use are also related to access to IT. It is predicted that the higher the level of access to IT, the higher the level of IT use by an individual.

The demographic category includes adoption factors such as age, education level, gender, and income level. It is hypothesized that factors in the demographic category will not significantly influence IT adoption and use. Although previous literature suggests that IT use will be higher for younger, more educated individuals (Batte, et al.), 1997 survey results suggest that demographic factors have little influence on IT adoption and use. This may reflect that demographic factors may influence the decision to adopt a new technology, but once that decision to adopt is made, demographic factors may have little influence on use.

Another category of IT adoption factors is IT training/knowledge. This IT adoption factor can be measured with variables such as type of IT training, days of IT training, and the level of knowledge on IT use. It is hypothesized that as the quality and level of IT training increases, the use of IT will also likely increase.

An important factor influencing the adoption of any new technology is an individual's perception of that technology. It is hypothesized by this research that one

of the key perception aspects influencing the adoption of IT is the level of trust that the potential adopter has in the IT system and in those who use IT. Trust can be defined as “an individual’s optimistic expectation about the outcome of an event” (Hosmer 1995). There are different aspects of trust related to IT.

An individual must first trust that information technologies will work and that IT will be beneficial in accomplishing his/her goals and in completing his/her tasks. An individual must also trust that the information they obtain via IT is accurate and the information they send via IT will not be tampered with and privacy levels will be maintained.

Trust proves to be a difficult variable to measure. Factors included in the trust category include an individual’s perception of the ease of use of IT as well as the benefit of IT. In this study, trust is measured by variables such as helpfulness of IT for work-related communication, problem solving ability, and banking and shopping via the Internet. Some individuals, either due to their background or current environment, have a fear of IT and feel that it is difficult to use. It is hypothesized that an individual will use IT more if they have a positive perception or high trust level in IT.

The final IT adoption category proposed by this research is the passage of time. It is hypothesized that individuals will increase their use of IT over time, as access to IT becomes more commonplace. In this study, the same group of people were surveyed twice to evaluate their changes in IT use over time. Time was measured by establishing a dummy variable where each survey response from the 1997 survey was assigned a value of zero and each survey response from 1998 was assigned a value of one. Time-interaction variables were also created for each variable by multiplying the original variable by the time variable. For example, the “days of training” variable (tdays) was multiplied by the time variable and became the “timeinfluenced days of training” variable (tdayst).

Key factors in the process of distance education:

the process of remote training, the following factors contribute:

- Students:

Regardless of educational content, role and main element in the learning process students are responsible.

- Coaches and Teachers:

Success depends on a lot of educational activities the ability, skills and knowledge are the coaches and professors.

- Facilitators of communication:

Facilitator bases, as the bridge between students and mentors are. Must base expectations of teachers and educational needs of students and service coordination and communication to create.

- Support staff:

One of the important pillars of any development of distance education programs, by development group finds. Operational support staff such as student registration, copy and distribute their resources, order textbooks, security and copyright, and are responsible for the report.

- **Management:**

The group decision makers, builders and judges are considered to be educational and should be considered among the factors above, establish the correct relationship formation.

FORMS OF DISTANCE EDUCATION

In its original form, teachers using distance education traveled to remote sites and taught a class, or corresponded with students through mail, telephone, or fax machine. Individualized study has been a method of reaching the remote student for some time. Detailed course instructions are sent to the learner who performs the assigned tasks and returns the completed work to the teacher for evaluation and reassignment if necessary.

Technology has raised the quality of individualized distance instruction. The use of various forms of electronic media increases time effectiveness and improves the delivery of information. Video, audio, and computer-based applications may enhance the product received by the independent learner. Electronic delivery can occur using synchronous communication, in which class members participate at the same time, or asynchronous communication where participants are separated by time (Romiszowski, 1993).

Video/audio models of distance education include broadcast television, cable television, satellite, microwave, fiber optics, and audio graphics. The most widely used format is broadcast and cable television (Parrott, 1995). However, developments in satellite and fiber optic systems have produced other successful programs. The interactive capability of many of these networks has produced a distance classroom that is nearly identical to a regular classroom. Teachers and students can interact through both two-way video and one-way video with two-way audio systems. The recent development of Desktop Video Conferencing (DVC) which brings interactive video capability to the desktop computer, further enhances learning opportunities.

The linking of computer technology through the use of the Internet or CD-ROM with television transmission provides a potentially new dimension to distance education. This technique can link university professors to high school teachers, or to physically disabled students, in a distance setting (McLean, 1996).

Another form of interaction is the use of computer conferencing. This method utilizes asynchronous communication in such forms as an e-mail list group, an Internet discussion group, or other types of conferencing software. Asynchronous methods of communication are especially appealing to the learner

who has difficulty scheduling specific time- and place-bound course work.

Have you been wondering about Distance Education?

Distance Education implies the provision of educational services to students who are not physically present. Put more simply, its educational courses, whether short-term programs granting a specific certification or complete academic degrees, which are delivered online or via other media, like TV and VCR, CDs, audio tapes, or mailed print material.

What Distance Education is all about?

The main aspects of Distance Education are as follows-

1. Geography: The point of Distance Education is to educate despite geographical differences. Online education effectively abolishes geographical as well as time differences, allowing an unprecedented number of students from all over the world to study in an institution, at any time of day.

2. Why higher education and Why take it from Distance: The world of today requires advanced education. Nowadays, advanced degrees are the standard – Having more than one education is not extraordinary but sometimes a must. Foreign studies are also common.

With the increased penetration of the Internet, Distance Education, offering anything from individual classes to complete doctoral degrees online, is a natural development of modern educational processes and requirements.

3. Distance Education as we know it today: It is a web-based education developed using e-Learning software tools and other distance-communication means. With increasing reliance on information, the demand for better and faster education grew and brought about the Internet.

4. Who Uses Distance Education? More and more people acquire higher education. Even professionals with full-time careers acquire new degrees, without change of pace at work. Distance education is perfect for the Military and for social institutions of all kinds. Parents can now combine higher education with family. Foreign education has never been easier – all it takes now is to log on.

Information Technology and its Components

Induction of IT as a strategic tool for agricultural development and welfare of rural requires that the necessary IT infrastructure is in place. The rapid changes and downward trend in prices in various components of IT makes it feasible to target at a large scale IT penetration into rural. Some of the broad factors to be noted with respect to various components of IT are listed below :

1. Input devices :

Radical improvements are witnessed with respect to the means of communication by human beings with

computers such as key boards, mouse devices, scanners. The advent of touch screen monitors that allow users to give input to computers by touching on the appropriate location of the monitor has made it possible to develop user-friendly interface for farmers which is easy, intuitive, circumvents language barrier and at the same time provides a relaxed environment to the users. The present day digital cameras make it possible to capture and store good quality graphics and large video clips. The small size and low weight of these digital cameras, which are increasingly becoming affordable, open up the possibilities of providing computer based demonstration clips to educate the farmers.

2. Output devices :

Monitor screens, printers & plotters, data projectors support high resolution and good quality output. The quality of these output devices have the potential of generating renewed interest in the farmers in using IT based services. The light weight portable data projectors can be easily carried by the agricultural extension personnel for serving larger audience. Similarly, speakers can also be attached to the computers to incorporate voice based trainings for farmers.

3. Processors:

The processing speeds of computers have gone up. At present, Intel P-IV based processors @ 1.5 Ghz are available in the PC range which makes it possible to undertake substantial processing of data at the client side.

4. Storage Devices :

40GB and even higher hard disk drives have become common in PC range of computers. This makes it possible to store substantial information at the local level which facilitates faster access. Similarly, high capacity floppy disk drives, CDs make it possible to transfer large volumes of data to locations which can not be connected to networks immediately. These storage devices are also used for backup of crucial data. As a precaution, many corporates store their backups at locations away from the place of work.

5. Software :

Various operating systems are available which act as interface between the user and the machine. The graphic user interface (GUI) has become an accepted prerequisite for end users. Microsoft's 'Windows' continues to be a favourite. Application softwares which can support complex user requirements are available. Of the shelf solutions for office automation packages, groupware applications, complex database solutions, communication products, solutions based on remote sensing & geographical information systems are available. In addition, solutions based on some or all of these are also readily available. The present downward trend in the IT industry provides an opportunity get customised application for any specific task developed at an affordable price. Rapid Application Development

and Deployment (RADD) is a popular model for quick development and deployment of applications. Development environment itself is simplified with tools that quicken the pace of software specialists. Project management and monitoring software are available that facilitate efficient execution of large and complex applications that are required for rural

6. Networking devices :

The capacity of modems, used to convert the data from digital to analog and vice versa, which are popularly employed to use telephone lines have increased. Internal modems are available integrated into the computer so that they are not exposed to outside environment. The capacities of other networking devices such as routers have also gone up which makes it possible to create large networks with smooth data transmission.

7. Transmission Media

The media through which the data transfer takes place has also undergone revolutionary change. Telephone lines are still the popular source although the reliability and low bandwidth are still major issues. High capacity cables, optical fibre, radio, wireless local loops, satellite transmission and various solutions based on a combination of these are already being used in many parts of the country.

8. Other accessories :

Uninterrupted Power Supply (UPS) devices are crucial to ensure the longevity of the IT equipment as well as provide backup mechanisms. The potential of solar power packs to provide a feasible solution to shortage of power in the rural areas needs to be exploited.

Role of IT in Agriculture

In the context of agriculture, the potential of information technology (IT) can be assessed broadly under two heads : (a) as a tool for direct contribution to agricultural productivity and (b) as an indirect tool for empowering farmers to take informed and quality decisions which will have positive impact on the way agriculture and allied activities are conducted.

Precision farming, popular in developed countries, extensively uses IT to make direct contribution to agricultural productivity. The techniques of remote sensing using satellite technologies, geographical information systems, agronomy and soil sciences are used to increase the agricultural output. This approach is capital intensive and useful where large tracts of land are involved. Consequently it is more suitable for farming taken up on corporate lines.

The indirect benefits of IT in empowering farmer are significant and remains to be exploited. The farmer urgently requires timely and reliable sources of information inputs for taking decisions. At present, the farmer depends on trickling down of decision inputs from conventional sources which are slow and

unreliable. The changing environment faced by farmers makes information not merely useful, but necessary to remain competitive.

Results:

This paper is a multidisciplinary study of ICT initiatives for rural development. It emphasizes adoption of a more systematic approach for integrating Traditional Knowledge Systems (TKS) and ICT inputs to ensure sustainability of rural e-governance projects. The study of literature related to rural development and e-governance has indicated various issues impeding success of such initiatives. The main issues are lack of localization of content for rural communities and inadequate participation of rural communities in design of rural ICT initiatives. The study therefore suggests the use of the systems-approach to integrate the relevant TKS along with ICT initiatives in the design of e-governance systems for rural development. This participatory approach can lead to creation of more acceptable and sustainable e-governance projects.

Regardless of the wide differences in ICT access between rich and poor countries and between different groups in the country, there are concerns that challenge the application of ICT in education with the existing differences among the lines of economic, social, cultural, geographic and gender will be broader. Everyone equal opportunities in terms of suitability for participation are necessary, but access to various factors, either as users or as producers through their sources is difficult and heavy. Therefore, the primary differences enhance and even grow. Consequently, programmers' international education is faced with a difficult challenge and how to help solve the problem and its development.

Promoting ICT in education, when done without careful study, can lead to the marginalization of those with more favorable conditions are unknown. For example, "women compared with men, because of illiteracy, lack of higher education, lack of time and mobility and poverty, controlling access to ICT and fewer opportunities for training are relevant. Also, more boys than girls' access to computers at home and school are not strange to say that if more boys than girls are willing to work with computers. The report of the University Association of American Women is that "Although some girls have an important gender gap have been limited, but today's technology, technology club, and boys in public schools while its own problems and programs are settled girls use computers for word processing the brand". In an assessment in four African countries, the activities organized by World links remote international cooperation on projects between teachers and students in developing countries will promote, despite creating programs without regard to sex contacts, sexual inequalities remain Uganda and Ghana. In addition, while more girls than boys in

relation to academic performance and advanced communication skills program will enjoy more than boys, but they were unable to perform their technological skills were. A set of economic factors, organizational and cultural differences involved in the social.

"The high ratio of students to computers and politics, whoever came first, the first is used in accordance with the girls wanted it." Girls travel restrictions in the early hours of daily work and home responsibilities are that this will limit their access. Also because local patriarchal beliefs dominate the boys are in the computer lab environment. Including proposed measures to address this discrimination, strategies to encourage schools to create "fair use" in the computer labs and the holding of meetings and sexual sensibilities conductivity decreased defense duties after school girls. ICT provides access to only a small part of the action is created equal. Equal attention should also be applied to ensure the technology really "is used by learners and ways of how well their needs will cure.

An educational program that reinforced this approach shows the overall program is bilingual. The program seeks to establish technology learning centers for bilingual teachers, students, teachers, parents and community members. Technical teams from each center three students, two teachers and the director of the Center with at least one female student and a teacher are female.

Another example of a general approach to the application of ICT in education, radio education project Gobi Women of Mongolia, which seeks to provide professional and educational structure of women's favorite courses around the nomads and their opportunities for income generation.

It contains topics such as livestock rearing, family support (family planning, health, nutrition and health) to create income in the application of local raw materials and basic skills for the job is a new market.

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A Comparison between Hegelian and Lacanian Ideal Hero in Sophocles' Tragedy of *Antigone*

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Abstract: Hegel, the German philosopher, and Lacan, the French psychoanalyst, have a lot in common when asserting their viewpoints concerning an ideal hero they favor in tragedy. Not all the heroes having dominated the classical tragedies have been able to mesmerize Hegel and Lacan as remarkably as Antigone has. The hero they support is capable of true heroism resulting from intellectuality rather than ephemeral emotions. Indeed, Hegel assures that the real hero create harmony between his internal motives and external elements. Based on Hegel, the real hero always acts and reacts in accordance with the divine pathos having been implanted in his heart. The pathos is substantial in that it has been divinely issued by the gods. The Hegelian hero takes his nobility from the fact that he has merged his individuality with gods' universality in order to have their support. The real tragic hero, Hegel says, should stand firmly loyal to the god he has chosen to obey. Such loyalty is apparently appreciated by Zeus because the tragic heroes have erased their will just to dictate and preserve the equilibrium Zeus desires. In fact, they act consciously and voluntarily in the plays, though they know they may be paid by death. Like Hegel, Lacan admires Stability in action and loyalty to the aim when talking of his favorite tragic hero. To Lacan, the concept of *beautiful soul* can manifest in a hero who knowingly acts and bravely welcomes what he himself has given rise to. Lacanian tragic hero like Hegelian one knows from the beginning the consequences of his behavior; nevertheless, he never loses his heart when confronted with the reactions coming from his opponent forces. Antigone can best glorify the features Lacan and Hegel endeavor to attribute to real tragic hero. Though encountered with physical death and conceptual dissolution better known as *second death*, Antigone stands faithful to the aims she has agitated for.

Keywords: dialectical method, pathos, ethical substantiality of family and state, the thing, sublimation, Derrida's supplementation, the beautiful soul, the second death.

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Introduction:

Hegel has asserted that the essence of a real tragedy should be explored where the heroes are caught up between two choices equally righteous and defensible. He believes that a talented dramatist artfully puts his heroes under sophisticated circumstances where they have to make up their minds between alternatives equally imposing and coherent. The hero situated between equal options, Hegel declares, is more life-like and digestible in contrast to a hero manipulatively placed between black and white options. The playwright Hegel admires arranges his plot in a manner which pressures the heroes involved to meditate, deliberate and hesitate prior to taking any action owing to the fact the alternatives designed interminably resemble and negate each other before heroes' eyes.

Hegel emphasizes that the essence of the whole universe is based firmly on endless dualities which intermittently metamorphose what we adore into what

we abhor and vice versa. Such dualities, Hegel asserts, function contradictorily, and coexist independently from our consciousness and intellect. The alternatives that challenge the heroes' discretion involve contradictions owing to the fact that they originate and rise from each other.

Prior to examining Hegel's notion of tragedy, and the procedure he explores in search of his ideal tragic hero, it would be worth-reading to see how Hegel has been appealing to his dialectical method to get equipped in search of actual heroism. Dialectical method is one of the concepts Hegel has patented; it functions as a key to enter his philosophical system that has outshone all the achievements he has extended to us. Hegel initiates his adventure through reversing in his favor Spinoza's premise of *determination*. "Spinoza proves that determination is negation. He concludes that to determine a category, one must negate that the original category belongs to other surrounding categories (Stace, 2001, p. 42). To

determine that blue is blue, it necessarily must be negated that blue is green, or red, or white. Consequently every determination necessitates a sort of negation. Hegel's reversal of Spinoza's premise of determination eventuates in his own theory of *negation*. He emphatically asserts that "every negation is determination and it is absolutely the reversed path of Spinoza's recitation"(Inwood, 1992, p. 200) Hegel announces that not only is every determination a negation, but also every negation is a determination. The color of blue is not green, or red, or white or any other color known to us; accordingly, through these negations blue is determined to be nothing except blue.

Reversing Spinoza's premise of determination, Hegel then constructs his own hypothesis of *tremendous power of negation* through which "Hegel reminds in view of the fact that every determination is a negation, the positive aspect of every thing is concealed in its negative one"(Stace, 2001, p. 43). To recognize and appreciate a category, we need to have access to the categories which negate our initial one. Hegel in fact makes an attempt to help us realize how shaky and unstable are the borders we draw between opposing forces. Contradicting elements owe a lot to each other beyond our minds having been inclined to observe the universe pessimistically through binary oppositions. Contrasting elements need vitally each other to find existence and meaning. In reality, opposing forces are so intermingled that one hardly can recognize them when viewed in isolation. In the twentieth century, post-modernist approaches take advantages of Hegel's enterprise as Jacques Derrida represents his own *supplementary method* which connotes the instability, and simultaneously the spuriousness imposed upon ostensibly opposing elements.

The theory of negation endorsed, Hegel embarks upon advancing his dialectical method, one of his most fundamental achievements the understating of which equips us in exploring his philosophy. This method of deciphering the universe and the forces it includes shows in practice how fragile are the walls built between the opposing forces, and simultaneously decodes why human's mind gets puzzled when exposed to varied alternatives that make the procedure of decision-making an extremely agonizing task. Hegel believes that we should place our tragic heroes in such situations where they face up to contrasting points which basically originate from one single essence. Two forces equally righteous are put before the heroes who are bound to vindicate just one. Nevertheless, Hegel says only the characters capable of reasoning can make their path amid the seemingly chaotic forces.

The theory of *monism*, promoted by Spinoza, stresses that the whole universe should be deduced from a single, unique, perfect, and self-independent category which never undergoes any change or split in it. Owing to the theory that the initial category of the universe is innately perfect, it does not need anything or anyone to make it get proven or stabilized. However, Hegel believes that Spinoza has come to such results because he has used his *understanding* in deducing the source of the universe. Hegel, instead, advances his own logical procedure which "disturbs that 'freedom' to think for *oneself*. It disturbs our 'freedom' to stand above a topic and think 'about' it as seems rational to us" (Houlgate, 1991, p. 65)

Reasoning is a stage Hegel prescribes instead of understanding. Although Hegel does not reject Spinoza's *understanding*, he insistently believes that compared to understanding, reasoning is far more accurate and reliable. Reasoning is precious to Hegel because it generates his dialectical method that he frequently appeals to in deducing the origin of the universe. The dialectical method reminds that the initial category or origin of the universe, unlike Spinoza's theory, cannot remain endlessly unique and undivided, and it undergoes changes and divisions throughout its development. Hegel's dialectic involves three steps: (1) we take one or more concepts or categories as fixed, sharply defined and distinct from each other. We call it the stage of understanding. (2) When we reflect on such categories, we behold one or more contradictions emerge in them which give rise to dialectic proper or the dialectical or negative REASON. (3) The result we take is a new, higher category that embraces the earlier categories and resolves the contradiction emerged in them. Hegel names it the stage of speculation or positive reason (Inwood, 1992, p. 62).

Hegel's understanding sets the concept of *Being* as the initial category of the whole universe. Being is a source which sheds life on the whole universe and the categories it contains. Based on understanding, Being functions as an immediate category that does not need any pre-defined approvals; it creates and approves the existence of every thing we have in the universe. Then, through reasoning, Hegel extracts the concept of *Non-Being* from the heart of the Being. This stage in Hegel's words delineates the negative reasoning whereby the Non-Being emerges from the Being and contradicts its own creator. Then, Hegel introduces his concept of *Becoming* which resolves in itself the contradictions having occurred between Being and Non-Being.

An example can give a clearer picture of what Hegel means by the triple hierarchy he advises us to follow when pondering on the origin of the universe. Suppose we have an *old bilingual* dictionary. Then,

we begin to reduce the number of specifying features which our dictionary includes. When we ignore the *oldness* of our dictionary, what remains is a *bilingual* dictionary. We can proceed to eliminate the specifying features by erasing the *bilingualism* of the dictionary, and what, at last, remains is the dictionary devoid of color, material, thickness, oldness, and so on. Despite being deprived of any feature, the dictionary exists, and we still can say we have a dictionary. What Hegel strives to prove is the fact that Being is what we never can take from the dictionary. Being, as the origin of the whole universe, incarnates through the whole categories of the universe, and as W. T. Stace remarks "the feature of being is the only joint feature among the entire universal concepts" (ibid, p. 120). If we carry out the same procedure on the whole categories occupying our universe, what at last we encounter is the initial concept of Being.

Nevertheless, such a concept of Being virtually means Non-Being, what Hegel has made us notified of earlier. When something is cleansed of all the features it has previously had, though still keeping its being, it is ultimately invisible, and in practice conceptualizes Hegel's Non-Being. To set us free from such a duality, Hegel announces the third stage of Becoming. It is a sphere which resolves in itself both the Being and Non-Being. The Becoming puts an end to the contradiction and challenge which the Being and the Non-Being have between themselves; it temporarily sets peace and stability. The Becoming indicates a principle whereby apparently stable thoughts reveal their inherent instability, turning into new more complex thoughts, as the thought of being turns first into the thought of nothing, and then into the thought of becoming. This principle Hegel tells us, is the soul of all truly scientific knowledge (Houlgate, 1991). Sedgwick in his book titled *An Introduction to European Philosophy* confirms that "dialectical method is in general the principle of all motion, of all life, and of all activation in the actual world. Equally, it is also the soul of all genuinely scientific cognition. Life itself is so dialectical that all life involves change (p. 62).

The definition Hegel renders of tragedy is closely linked to his dialectical method owing to the fact that the hero is not entrapped between an ultimately wicked position, and an exaggeratedly benevolent option. In practice, the hero is exposed to a power which fosters its own enemy; in other words, what he is bound to choose gives rise to its own contradiction (Hegel, 1999, p. 93). The very relationship covering the concepts of Being and Non-Being dominates the whole categories offered to the tragic heroes; consequently, the hero finds himself enslaved as he feels he is free. The hero gets naturally flabbergasted when he is at the verge of taking steps throughout the

tragedy. Hegel has a deep contempt for the tragedies which offer their heroes unsophisticated conditions where a paragon of humanity is arrayed versus a diabolic antagonist, due to the fact the heroes can easily determine which to vindicate. To Hegel, the essence of true tragedy should be traced where the hero has to make up his mind between two equally righteous and imposing forces which identically lure the heroes to themselves. The battle never takes place between the bad and the good; in fact, two good forces are presented before the hero, and he should choose just one. "The essentially tragic fact is the self-division and intestinal warfare of the ethical substance, not so much the war of good with evil as the war of good with good" (Paulucci, 1975, p. 67). The Greek tragedies can best exemplify the ideal tragedy Hegel desires. Such tragedies represent a setting where opposing forces emerge from each other and then resolve their contrast to support Hegel's triple formula of the universal phenomena. The forces involved put equal pressure upon the heroes who wisely strive to determine which force they should vindicate. One power functions a thesis which gives rise to its enemy incarnated as the antithesis. However; this battle is followed by the synthesis that brings peace, and regulates the chaotic atmosphere the opposing forces create.

The forces presented in Greek tragedies, as well as the heroes summoned into the battlefield are not engaged with mundane issues. The tragic heroes, Hegel says, consciously try to be dealing with universal and divine issues at the price of erasing their own individuality. They act and react in a world immensely different from ours; it is meticulously monitored by the divine gods who seek their shares on the Earth. These gods endlessly quarrel with each other owing to the fact the domains of their affairs confront, and cause them to start fighting to stabilize their favorite orders (Rutherford, 2005, p. 64). One god affirms an issue, immediately another god feels threatened and begins to press back the god initiating the quarrel. The triangle formula of thesis, anti-thesis, and synthesis drawn by Hegel comes into mind where Zeus, the supreme god, enters to resolve the challenge occurred, and exerts peace and regularity on the sphere he is in charge of. The gods actualize their influences upon the earth through what Hegel names *ethical substantiality* which the heroes undertake to perform. The ethical substantiality highlights the divinity and spirituality which is attributable to the gods the Greeks used to worship. In fact, the heroes starring in the Greek tragedies wisely and knowingly have the inclination to make their personal wills identical to the ethical and divine decrees coming from gods. "The hero makes himself inseparably coalesce with particular aspect of ethical and substantive life,

contently and deliberately commits himself to that” (Paulucci, 1975, p. 47). Indeed, the heroes in tragedies, in accordance with their social and political roles, subordinate themselves to specific territories dominated by different gods.

The tragic heroes assuredly confide their will to the divine gods because such a sacrifice enables them to achieve two important goals. (Charlton, 1988, p. 43). First, the hero hereafter has the ultimate vindication of the god he has chosen to obey. He has realized that acting through his individual will can eventuate in eternal catastrophic curses from which he, and his descendants as well, will not find any escape. Coherently, he aptly concludes that he needs to make a compromise between his personal will and gods' divine orders. The heroes know well that the whole phenomena occurring in the universe find their justifications from the relationships the divine gods develop among themselves. They also know that Zeus himself, above all hands, meticulously observes the issues the gods are involved in, and equalizes the equilibrium of power among them. Adhering to what the gods will to do make possible on the earth the very equilibrium Zeus decrees. Consequently, the gods involved in tragedy have some agents who sustain their wills on the earth, and guarantee the balance among gods (Rutherford, 2005, p. 72). Hegel brings out his concept of *true action* that “consists of two parts: 1, what is in substance good and great, the divine actualized in the world, as the foundation of everything genuine and absolutely eternal in the make-up of an individual's character and aim; 2, self determination and freedom” (di Beistegui & Sparks, 1969, p. 46). Hegel's definition of true action reveals the second aspect of ethical substance which exacerbates the heroes from the probable retributions awaiting them in the nether world. In fact, the heroes “act in accordance with a specific character, a specific pathos. In such a case, there is no lack of decision and no choice (Paulucci, 1975, p. 70). The gods, who in the world after death have the right to judge the heroes' actions, feel responsible for the circumstances the heroes face, and vindicate them against divine punishments. What the Greek spirituality disapproves most is the selfish emphasis that ethical powers put on their rights. In fact, the ethical substances issued from gods as well as the heroes supporting them should succumb to the third powers that function as synthesis to resolve the clashes. Zeus above all observes the affairs among gods, and lastly this is he who says the last word, and reestablishes order throughout the universe.

The family and State in ancient Greek culture were not secular at all; in fact, they were responsible for some clashes recorded in tragedies among the gods. Apparently, they were two domains ultimately of the

gods' interests. Both of them had their own particular supporters among divine gods. The individuals who are in charge of the State and the family are assumed to make a compromise between their own individual wills and the gods' orders. However, the heroes enjoy the compromise made provided that the gods be given the upper hands. As long as the gods fight, their representative heroes on the Earth involve in conflicts. Nevertheless, reconciliation at last will bring the forces under a same sphere where peace and calmness dominate.

Hegel regards Antigone one of the greatest characters ever seen in tragedies because her tragedy fully puts the family and State against each other. “Antigone reverences the ties of blood-relationship, the gods of the nether world. Creon alone recognizes Zeus, the paramount power of public life and common wealth” (Kaufmann, 1992, p. 205). The family, as a thesis, gives life to its anti-thesis which incarnates into the State. Then, death enters synthetically to establish peace between these two clashing forces. In practice they build the society. The family fosters the State by supplying it with the members it has nourished, and in return the State takes the responsibility of defending the family against possible hazards, and provides the facilities the family needs to survive. The borders existing between these two categories are established by Greek divine gods. Zeus makes women supervise the family affairs, and commits the king to sustain the social affairs. Indeed, Hegel highlights that women control the family, and men dominate the State; an agreement that assures the equilibrium between two opposing sexes. The family is regarded as the realm of divinity, for the rules dominating it are self-conscious and generalized. “Ethicality is the divine or feminine law, that of the family as ‘natural ethical immediacy’ whose existence is sheer being substance” (di Beistegui & Sparks, 1969, p. 22). The family is devoid of political laws; it is the realm of Penates and obscure forces. The Family is a natural ethical community working in an unconscious way, and finds its divinity from the nether world. It also helps the pre-birth world incarnate on the Earth.

The State, the masculine world, is of self-conscious reality where human rules take the place of divine laws. Nevertheless; the State is not entirely cleaned of divinity because Zeus vindicates and stabilizes it; he taught mankind how to establish a State to exert laws on his society. Zeus and Apollo, the god of wisdom, do not care about the family-related issues as devotedly as the new gods like Eumenides, Dike, and Nemesis (Grimall, 1990, pp. 78-80). Zeus supports the State, while Eumenides and the gods of the nether world are guardians of emotional affairs in the family. The family and State undergo battles; the State emphasizes its rough and inflexible rules, whereas the

family defends its soft and affectionate affairs. Each of these two spheres challenges to establish its own laws, and tries to keep society's members under its dominance.

The tragedy of *Antigone* written by Sophocles reveals the battle between the family and the State. Creon, the head of the State and the supporter of social affairs, is in charge of the security of the whole kingdom. He has Zeus's and Apollo's support, he has to keep the society safe; he can not allow marginal affairs to jeopardize the society. To Creon, Eteocles is the defender of the State; consequently, he deserves a splendid burial ceremony which can glorify the state of those martyred for their nation. On the contrary; Polyneices has attacked the State, so he should not be buried respectfully. Creon is somehow righteous in his hostility to Polyneices, because he has to vindicate the very State that provides the nation security. If he let him be buried as gracefully as his brother, the insurgents would venture to risk the nation, assured that they will be celebrated as heroes as they die. Coherently, Creon does not let his individual feeling middle in the governmental affairs. On the other hand, Antigone, as a woman, defends the issues cherished in family territory, and preaches the gods of nether world who watch blood-relationships fostered in family. She apparently ignores Creon's governmental decree ordering the unrespectable burial of Polyneices. She is not capable of dividing her brothers into friend or enemy because the family does not basically particularize categories; it mainly generalizes them, so both brothers, regardless to what they have done to the State, deserve equally respectable burials. Such firm unwritten laws cause Antigone to risk her life by burying her brother, Polyneices.

*Antigone: But I bury him; and if I must die
I say that this crime is holy: I shall lie down
With him in death, and I shall be as dear to him as he
to me.
It is the dead.
Not the living, who make the longest demands;
We die forever... (Sophocles I, 55-65)*

Antigone also announces that what she does is not merely her own intention; in fact, the gods of nether world emphasize the rights of the dead and family relationship. She responds Creon's accusation and says:

*It was not God's proclamation. That final justice
That rules the world below makes no such laws.
Your edict, king, was strong,
But all your strength is weakness itself against
The immortal unrecorded laws of God.
They are not merely now: they were, and shall be*

Operative for ever, beyond man utterly. (II, 55-61)

Antigone reminds that the ethical substantiality of the nether world, to her, overweighs the rules of the State. These laws are unrecorded because they have not entered the symbolic and particularized realm of men. However, her sister Ismene is overtaken by the imposing doctrines of the State, and timidly escapes the responsibility she is expected to take for burying Polyneices. Apparently, she proves to lack the sense of obligation Antigone has:

Think how much more terrible than these

*Our own death would be if we should go against
Creon*

And do what he has forbidden! We are only women

We cannot fight with him, Antigone!

The law is strong; we must give in to the laws.

In this doing, and in worse, I beg the dead

To forgive me, but I am helpless: I must yield

*To those in authority. And I think it is dangerous
business*

To be always meddling. (II, 44-52).

Nevertheless; Creon is given the responsibility of maintaining and guarding the laws having been dictated by Zeus and Apollo who wish to see humans' community stable and strong. Creon is equally justified in stressing the laws he carries on. He should give Eteocles a respectable burial ceremony, while he should prevent Polyneices from being buried like a national hero. He is a traitor to the State, so he should be treated as he deserves.

*I have nothing but contempt for the kind of Governor
who is afraid for, whatever reason, to follow the
course that he knows is best for state; and as for the
man who sets private friendship above the public
welfare...and I need hardly remind you that I would
never have any dealings with the enemy of the people.
No one values friendship more highly than I; but we
must remember that friends made at the risk of
wrecking our ship are not real friends at all. (I, 17-
27).*

Apparently, we can trace the formula delineated by Hegel when talking about his dialectical method. The family initially plays its role as thesis; then, it gives birth to its anti-thesis, the State. Both Creon and Antigone are doing their duties designated by the Greek gods. However, the Hegelian triple formula still lacks its third side, the synthesis, which fully manifests through the different relationships family members organize among themselves; the inevitable death that terminates the presence of both the family and state.

To know properly how death functions as synthesis, the ways people within a family relate to each others should be taken into account. Hegel defines three kinds of relationship that people in a common family can generate. First, we have the husband-wife relationship, that Hegel regards purely natural and sensual. Hegel believes that the grave ethical substance that gods put in family cannot come from this relationship, for a man and his wife initially have based their connection on natural and sensual needs. The second relationship seen in a family emerges among the children and the parents. Such relationship to Hegel is still devoid of divinity owing to the fact that they are natural and unconscious. This relationship is contaminated because it has come out of sexual affairs, and it does not have a root in the so-called pre-birth ethical world. The children owe their lives to their parents, and the parents have their parenthoods from their children. The last but not the least significant relationship that Hegel highly values appears between sisters and brothers. Hegel believes that this relationship is ultimately purified, and does not contain any natural or sensual desires. Sisters and brothers do not need each others for their existences; they are independent beings that do not provide each other's sensual and natural needs (Paulucci, 1975, p. 238). Accordingly; the entire substantiality of the family aptly appertains to the connection created between sister and brother. For such substantial role, sisterhood functions as a confident receiver of gods' divine orders, and dislodges motherhood approximately out of substantiality of the family. She leads the army of femininity in its battle against the non-divine masculine world. Though the brother goes into the State's service, sister remains devoted to her brother, and never lets her pure, divine feelings get discolored during this imposed separation. The State strives to keep men aloof from the world of femininity, and to a large part it succeeds to. The affection the sister has to her brother is as strong as before, while being in State's service influences brother's feeling to his sister. Consequently, sisterhood is regarded by the State an everlasting menace plotting to undermine its totality.

Nevertheless, the disputes between the State and family are not everlasting. Death synthetically reconciles these opposing forces, and supplies a moment for subsiding hostilities. The brother's death enables the sister to express fully and publicly her uncontaminated ethical feelings. Mother and wife due to their natural and sensual feelings cannot be confident representatives for offering ethical divinity to the brother. In fact, if it were not for the relation between sister and brother, the whole system of family would remain permanently natural and unethical. The funerary might be regarded as the only moment when the sister is permitted officially to release her suppressed feelings. It is a particular moment that the State must recognize so that the divine ethicality can present itself on the Earth. If prevented, she undoubtedly shakes the bases of both the community and the State. Hegel highlights that if after some coincidences, the sister realizes that she can not bury her brother, and reveal her ethical substance, the femininity in body of sisterhood would destroy the whole organism of community; "the femininity will come to fight the masculinity for such a suppression"(di Beistegui & Sparks, 1969, p. 23). Antigone is prevented from her right to show her ethicality in her brother's funeral ceremony; consequently, she agitates against both the State and community. This anger gets so strong that it finally ruins even the household of Creon. Death reconciles the divine pre-birth world from which we originate and the domain of the State. Death solves both these spheres into itself because it spoils the body, and causes us to return to where we have come from. "Death is the ethical and natural given ground around which the two ethical principles (pre-birth and on-earth lives) commune" (ibid, p. 22). Death above all brings freedom and security for the heroes. In the battle between two equally justified forces, the one that dies, indeed, reconciles its own vindicating god. However, the one that has seemingly won the war cannot meet the god that supports it. The winner has to live on the Earth, and suffer the punishments executed by the Furies who guard the sanctity of all ethical forces on the Earth. Antigone dies while she has fulfilled her duty to the gods when living on the Earth. Creon, however, has to face the agonies exerted by the Furies. He insisted on his right, and totally violated what Antigone cherished. He knows well that his son's and wife's deaths are the outcomes of his spiteful behavior to Antigone.

Is Antigone the criminal, for transgressing 'the laws of the land' (that is, Creon's edict)? Or that Creon is, for going against the 'sworn justice of the gods' (a female's right to mourn her family)? Antigone ends up committing suicide and Creon finishes the play

broken-hearted, his son having joined Antigone in her suicide. Neither side can be said to 'conquer' the threat posed by the other.(Whitmarsh, 2004, p. 81)

Though both Antigone and Creon obey divine gods, Hegel cannot conceal his respect to Antigone. He believes that compared to Creon, Antigone reacts more heroically in facing her difficulties. Indeed, Hegel regards Antigone an ideal hero who aptly represents heroic features. The ideality of Antigone does not exclusively come from her fidelity to the divine orders which Greek gods issue; she bravely welcomes the whole probable outcomes resulting from her behavior. She accepts consciously to be in charge of whatever following her reaction to the State, although her individuality acts in accordance with the divine ethical pathos which is substantive (ibid, p. 84). Antigone heroically lets herself be blamed for whatever coming out of the compromise she has made with the gods; in other words, she takes the responsibility of the processes that the Greek gods in large part have given rise to. Antigone sincerely appreciates the fact that she has been defended by the divine gods, and knows well they have assured her salvation in both the earthly and after-death lives. She does not let duplicity contaminate her morality; she receives unwelcomed results as satisfactorily as sweet ones. She never accuses the gods of the catastrophic scenes occurring in the tragedy. Not only does she accept the conclusion of her actions, but she also opens her arms warmly to the curses her family members have suffered; she approves that her royal prestige accompanies the agonies undoing the house of king Oedipus. Besides, Antigone, as a Hegelian ideal hero, does not observe any difference between what we may consider as conscious or unconscious; she regards herself responsible for everything done, and her moral obligation does not allow her to escape the issues she herself has not directly had hand in. Antigone copies her father in welcoming penalties even though following from unconscious commitments. Oedipus feels he deserves to be cursed for the incestuous affairs he has unknowingly had with his own mother, and lends himself to punishment (Draper, 1980, p. 113).

In Greek tragedies the miseries are handed down from one generation to the next, and the members of a dynasty shares in their honors as well as misfortunes; they do not find themselves exonerated from what their ancestors have committed.

*Antigone: you have touched it at last: that bridled
Their crime infection of all our family!
O Oedipus father and brother!
Your marriage strikes from the grave to murder mine.*

*I have been a stranger here in my own land:
All my life
The blasphemy of my birth has followed me. (IV 37-44).*

Antigone acknowledges the truth of a misery that runs in her family, and confirms she has done her duty to her brother knowingly; she makes no attempt to escape the death proclaimed by Creon.

Antigone: Creon, what more do you want than my death?

Creon: Nothing. That gives me every thing

Antigone: Then I beg you kill me...

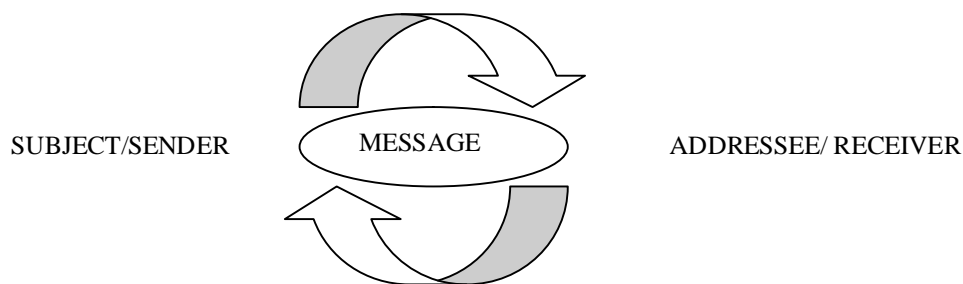
I should have praise and honor for what I have done. (II 91-97).

She publically announces her readiness to death, and declares her death brings honor and everlasting praise. Since faced by Creon's disagreements, she has predicted her death, and even her voluntary moving to death is confessed through the Chorus. However, such brave reaction towards destiny is seen by no means in Creon's personality. In fact, he loses his totality when confronted by the results his irresponsible behavior has generated. From Hegel's notion, he does not deserve to be counted a one-hundred-percent hero though acting based on divine orders of Zeus and Apollo, supporting gods of the State (Grimall, 1990, p. 58).

Passing approximately two centuries, Jacques Lacan, the French psychoanalyst repeats Hegel's definition of an ideal hero; nevertheless, he uses his own technique in search of the ideal hero. Lacanian ideal hero in large part represents the same features Hegel brings out when delineating the ideal hero; both Lacanian and Hegelian ideal heroes succeed heroically to sustain their integrities when they come across to the conclusions their performances have given rise to (Lemaire, 1994, pp. 61-2). Both heroes act consciously, and feel responsible for what they embark on doing.

There are few people in literature history not having been astonished by Antigone's brilliant performance in her tragedy. She knows well her actions do not mean anything but certain death; still, she peruses to fulfill what she thinks to be true, ignoring how others may react to her performance. Slavoj Zizek, the renowned Lacanian psychoanalyst, uses his mentor assumptions to present the public the shining soul that elevates Antigone to the state of an ideal hero. Zizek, in his book titled *enjoy your symptoms*, refers to what Lacan learned from Edgar Allen Poe's 'purloined letter'. He opens his debate by announcing that "a letter always arrives at its destination (Zizek, 1992, p. 12) . By this he means

that whenever a subject sends a letter, its addressee undoubtedly will receive, and answer it. The subject by his letter sends a message to his addressee, and it influences the receiver greatly enough to cause him to react. Zizek also insists that the letter unquestionably completes a circle, where the sender and the receiver organize its diameter (ibid, p. 12)



Lacan's formula

Such a circle is always guaranteed; the communication between the sender and receiver never cuts off. As the letter arrives, the receiver gets stimulated to answer it. The essence of the letter is important, and the sender can easily guess how his addressee might react; consequently, he is fully prepared for what the receiver will send him. However, Jacques Derrida reacts posing some coherent questions concerning the certainty of this alleged form of communication. He believes that this theory is totally illusionary, and it cannot get externalized. He asks: "So why does the letter always arrive at its destination? Why could it not sometimes, at least also fail to reach it? Isn't it always possible for a letter to go astray?" (ibid, p. 13). He truly mentions that we should accept the fact that there are many potential obstacles that prevent the letter from getting its exact address. Derrida's skeptical view regarding what Lacan assumes about the letter theory originates from his views concerning the arbitrary and shakable relationships between any signifier with its alleged signified. He is sure that the purloined letter theory is not an exception; the same chaotic atmosphere that dominates the world of the signifiers and the signifieds is felt in the assumption Lacan has made. To Derrida, the Lacanian letter functions as a signifier that wrongfully strives to ascertain a signified as its own permanent reference. Derrida believes that such a firm relationship is purely illusionary due to the fact that "the signified cannot orient or make permanent the

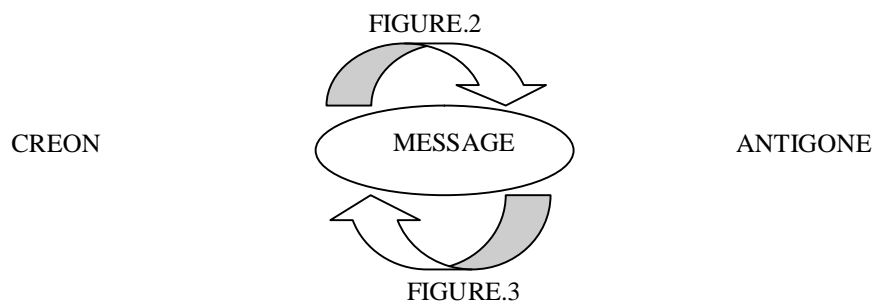
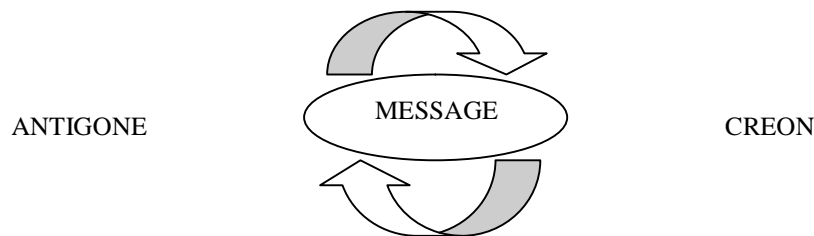
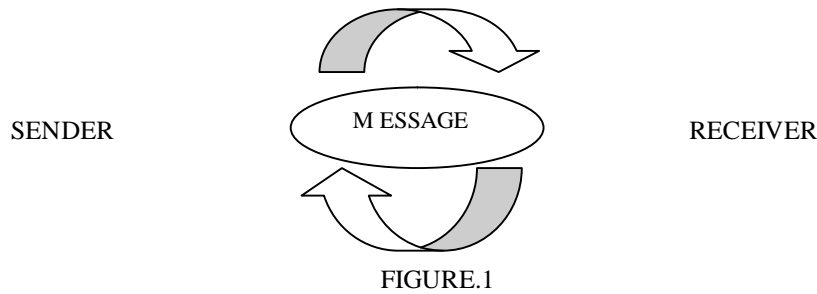
meaning of the signifier because the relationship between the signifier and the signified is both arbitrary and conventional" (Bressler, 2007, p. 119). The way we incline to perceive the relationship between the signifiers and the signifieds cannot be a trustworthy touchstone because what we regard as a firm signified for a signifier can itself be a signifier for something else that might possibly contradict the initial signifier, so we should not rely on these infirm and chaotic relations (Sedgwick, 2001, p. 175)

Despite the coherent objection made by Derrida, Lacan persists that his theory can bypass Derrida's assumption because the letter that in Derrida's words acts as a signifier does not have any pre-determined addressee or signified. Indeed, the letter does not have any clear addressee, and every person who receives it is its addressee. He adds that "a letter always arrives at its destination since its destination is wherever it arrives" (Zizek, 1992, p. 12); consequently everyone who intends to read this *addressless* letter is to be regarded as the receiver of that letter. Lacan surely has in his mind the German experience of *flaschenpost* where a man entrapped in an uninhabitable and remote island puts an SOS message into a bottle, and throws it into the ocean. The doomed man has not ascertained any person as his message receiver; everyone taking the bottle is truly the receiver. Zizek points out the significance of the purloined letter lies in the fact that the sender of a message knows fully the content of his letter; accordingly, he knows well what his receiver

might respond when encoding his message. Zizek confirms that “the sender always receives from the receiver his own message in reverse form, “the repressed always returns,” “the frame itself is always being framed by part of its content” (ibid, p. 12). The message sent causes an answer in the receiver and the answer is exactly the externalized meaning of the message having been sent; the receiver acts as a mirror reflecting the sender’s intention.

Such a discovery gives rise to Lacanian definition of *the beautiful soul*. Similar to Hegelian ideal hero, those who own beautiful souls take the responsibility of the entire consequences of their actions; they consciously know what they do, and bravely wait to face whatever may follow from their actions. They welcome the bad results as warmly as their own great

achievements. To possess the beautiful soul, the subject should not lose his totality when encountered with his action reflection; he has no right to say: “*but I didn’t mean that*”. He must be notified that he is taking what he has paid for. Lacan defines hero as the subject who fully assumes the consequences of his act, that is to say, who does not step aside when the arrow that he shot makes a full circle and flies back at him. Based on the assumptions made, Antigone can show herself as a hero equipped with the beautiful soul whereas Creon is introduced as an anti-hero who disintegrates as confronted with the echoes of his decision.



The figures shown above represent that Antigone knowingly sends her message to Creon, she is fully aware of the outcomes of her performance;

nevertheless, she never undergoes any hesitation when realizing that her behavior has ascertained her death. However, the figure focusing on Creon disgraces him

as an anti-hero who retreats when he comes across to the catastrophic results of his role in the play.

Antigone: You must decide whether you will help me or not.

Ismene: I do not understand you. Help you in what?

Antigone: I am going to bury him.

And some lines later Antigone adds that:

But I will bury him; and if I must die. (Sophocles 27-55)

Antigone assuredly proclaims her message of war though she knows it can mean death. She sends Creon her message and Creon as the receiver answers her back by sentencing her to death; her performance matches the formula Lacan supposes in dealing with the ideal hero.

Antigone: I knew I must die, even without your decree:

I am only mortal. And if I must die

Now, before my time to die,

Surely this is no hardship. (II 63-6).

Attempting to understand why Creon in Lacanian point of view does not deserve appreciation, we put him in trail by considering the figure (3) where he sends his message to Antigone in order to stimulate her to answer him. Unlike Antigone who welcomes all the consequences of her actions, Creon gets remorseful and surprised when he notices how Antigone reacts against his sent messages.

Creon: Polyneices, I say, is to have no burial: no man is to touch him or say the least prayer for him; he shall lie on the plain, unburied; and the birds and the scavenging dogs can do with whatever they like. This is my command... (I 35-7)

Here Creon recites his message one more, and Antigone makes herself the addressee of Creon's message. Creon has insulted the realm and laws of the dead, and Antigone considers her the vindicator of the divine realm of the dead and the family. The message sent by Creon has in itself some threats for the very domain sustained by Antigone. The message sent can be translated into the message of war and death; nevertheless, Antigone bravely rushes to receive the message. She externalizes what is hidden in Creon's message, and lends herself warmly to the jaws of her death. What makes the profound differences between Antigone and Creon is matter of the *beautiful soul*, which Creon lacks. Such a failure to possess the *beautiful soul* causes Creon to become massively fragile when encountered with the subsequences of his deeds.

Choragus: Go quickly: Free Antigone from her vault

And build a tomb for the body Polyneices

Creon: You would have me to do this?

Choragus: Creon, yes!

And it must be done at once: God moves

Swiftly to cancel the folly of stubborn men

Creon: It is hard to deny the heart! But I

Will do it: I will not fight with destiny

Choragus: You must go yourself. You cannot leave

It to Others.

Creon: I will go.

-Bring axes, servants:

Come on with me to the tomb. I bring her, I

Will set her free. (V 96-106).

Realizing the certainty of Teiresias's prediction, Creon changes his mind, and strives to survive Antigone from death. Such a retreat from the inflexible initial position descends Creon to an anti-hero who unwarrantedly tries to propagate him a real hero: a defender of divine laws. Unlike Hegel, Lacan shows no devotion to Creon: indeed, he believes it is unquestionably wrong if, like Hegel, we name Creon a hero who sustains divine laws. To him, the ethical equation delineated by Hegel in his philosophical notion concerning the puzzlement between two equally righteous powers, does not emerge convincing and defensible. Lacan insists that we not bother ourselves in recognizing the right from the wrong when asked about what passes between Antigone and Creon. The results are clear and unwavering: Antigone is double victimized; first her defense of divine laws is humiliated by an anti-hero incarnated in Creon, and second, she is put unjustly in the scale where her rightness is to be deduced when compared to Creon's. Lacan does not feel uncertain when he announces that "the behavior of Creon is marked by hamartia: he makes a mistake in judgment. Still more, he commits stupidity; Creon is an anti-hero, a secondary hero who wrongfully has been favored so far. (Harasym, 1998, p. 111). Indeed, Creon muddles in the affairs which are at the hands of the gods of the nether world.

Despite the dissention that Hegel and Lacan have in their debates on Creon, both glorify the shining beauty that elevates Antigone to a real hero. They use their systematic findings for shading light on the puzzles exposed to the heroes, and monitoring the paths the heroes choose to win their fame and reputation. Neither Hegel nor Lacan approves the dramatic characters who escape the consequences of their deeds. In fact, Antigone's brilliant performance paves the way for quenching the thirst of those who eagerly seek for real heroes. The realm of psychoanalysis in parallel with philosophy reveal how real heroes make their way amid the gloomy milieu they are involved. They rely on the talent of their reasoning to discover

what is true , what is the real essence of humanity, and the spring the causes them to flourish throughout history. Passing excellently the standards designed by Lacan and Hegel, Antigone amounts to the position which serves as a scale for weighing the honesty of every body claiming to have valor and heroism.

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Effect of Ill-Health on Rural Households' Welfare in Ondo State, Nigeria

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Abstract: This study assessed the health status and welfare of rural households in Ondo East and Ondo West Local Government Areas of Ondo State. The data were obtained through the use of structured questionnaires that were administered to farmers in two randomly selected Local Government Areas (LGAs) of the study area. Simple descriptive and regression methods were used to analyze the data. The common self reported types of sickness were tuberculosis (17.3%), whitlow (17.3%), guinea worm (5.5%), malaria (40.0%), pneumonia (2.7%), cholera (2.7%), diabetes (2.7%) and hookworm (1.8%). The study also revealed that, cost of treatment, cost of hired land and days of incapacitation have negative relationship with per capita income, while the imputed cost of family labour, cost of other inputs (such as fertilizer and seeds), family size and land area have positive relationship. The study recommended the need to raise national awareness of the incidence of diseases and their implication through information, health care facilities, encouraging government rural partnership, micro entrepreneurship and institution and industrial development.

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Introduction

Poverty refers to a situation where an individual or a group of individuals fails to attain a minimum level of welfare that is generally considered essential by the standard of a society (Aluko, 1975, Ajakaiye and Adeyeye, 2001). In many developing countries, poverty is concentrated among people with certain characteristics like low education, poor condition of health, unstable employment/unemployment, low status job, low and unstable income, poor housing condition, large families, absence of savings, constant struggle for survival and absence of material possession/investment (Sen, 1992). Past research efforts have closely addressed many of these key policy aspects, but in some African countries, just very little is known about the linkage between households' economic status and health status of the people. However, widespread poverty and emergence of many infectious diseases in Africa presuppose that research into the linkage between households' welfare and ill-health will be of tremendous assistance to policy makers in their efforts to improve people's standard of living.

Several approaches have been used to conceptualize households' welfare like the households' income, basic needs (which go beyond income to focus on deprivation of requirements for meeting basic human needs) and human capacity (which places some emphasis on the ability of people to enjoy long/healthy lives, to be literate and to participate freely in social/development activities in

their society). Also, economic analyses of health issues and well-being have focused on health status and health expenditures as inputs or means for achieving economic growth through its productivity effects (Lawson, 2004). This perspective presupposes that a healthy population is an indispensable engine for economic growth and development.

The classical economists' view of the relationship between health and economic development states that wealth leads to health with improving health as an output of the growth process. Therefore, considering the interconnected nature of health and welfare, incapacitation through sickness is likely to have an impact on the capacity of households to escape poverty (Wagstaff, 2002). Similarly, ill-health has other implications such as damaging traditional social support networks and increasing health care costs, all of which make breaking out of the cycle of poverty more difficult (Lawson, 2004). Kyegombe (2003) identified five main dimensions through which aspects of health/ill-health interact with other components of poverty. These are income poverty nutrition and health which makes the poor not to be on good nutrition; shelter and health which implies that the poor cannot afford good shelter; work environment and health implying that the poor cannot afford to work in good environment; income poverty and health care cost showing that the poor cannot afford to give himself good health treatment and ill health and erosive livelihood strategies implying that ill health can lead

to depletion of human assets by withdrawal of children from schools.

World Health Organization (WHO) noted that the 1.2 billion people in the world that were living in absolute poverty are those with high vulnerability to infectious diseases such as malaria, tuberculosis (TB) and HIV/AIDS (Tolhurst and Theobald, 2003). Essentially, the Millennium Development Goals (MDGs) relating to poverty reduction and health improvement are closely linked. This is because many aspects of poverty have a negative impact on health and on access to effective health services (DfID, 2003). Specifically it had also been submitted that while the economic burdens of infectious diseases like HIV/AIDS, malaria and TB are enormous, malaria alone slows the growth rate of African economy by 1.3 percent.

WHO and the United Nations (UN) have also reported that despite a tremendous growth in World's economic activities due to the penetration of transnational corporations and computerization, the world's poor have not so much benefited (Kent, 1998). It had been argued that the horrific disease burden of the poorest countries is a fundamental barrier to economic improvements of the world's poorest people. Consequently, although health is a valid end in itself, the main reason for seeking to improve the health of poor people is as an investment to facilitate household welfare. Therefore, World Health Organization (WHO) advocates a broad response to poverty and health that includes focus on specific diseases, efforts to promote pro-poor health systems and measures that address broader determinants of health initiatives, which promotes cross-sectoral actions in education and social protection, among others.

The objective of this study is to analyze the effect of inability to attend to normal activities due to ill-health on the welfare level of rural households in Nigeria. The research hypothesis (null) states that there is no significant relationship between the number of days farmers could not work due to ill-health and households' per capita income. In the remaining parts of the paper, materials and methods of analysis, results and discussions and conclusions are presented in that order.

Materials and Methods

The study area

The study was carried out in Ondo State, which was created on February 3, 1976. It has a total land area of 13,595.00sq/km with an estimated population of 3,460,877 people in 2006 (NBS, 2009). Ondo State lies between Longitude 4° 30' and 6° East of the Greenwich meridian 5° 45' and 8° 15' North of Equator. This means that the State lies entirely in the tropics. It is bounded in the North by Ekiti/Kogi

states, in the East by Edo state; in the West by Oyo and Ogun States; and in the South by the Atlantic Ocean. Ondo State has 18 local government areas. The climate is tropical with two distinct seasons; the rainy season and the dry season. The temperature throughout the year ranges from 21°C to 31°C while humidity is relatively high. The annual rainfall varies from 2,000 mm in the southern parts to 1,150 mm in the northern parts.

Sampling Methods

The research employed multi-stage random sampling technique. At the first stage, Ondo zone was randomly selected from the Agricultural Development Project (ADP) zones in the State. At the second stage, we randomly selected 2 Local Government Areas (LGAs) from Ondo zone. The 2 LGAs were Ondo East and Ondo West. The third stage involved division of the LGAs into their political wards. It was found that Ondo West has 12 wards and Ondo East has 10. In absence of household listing, we proceeded to select a total of 5 households from each of the wards. Therefore 60 households were sampled from Ondo West while 50 were interviewed from Ondo East.

Multiple Regression Analysis

Following Morduch and Sicular (2002), households' welfare can be assessed using the human capital theory, with the index of welfare being the per capita income. In this study, some functional specifications of equation 1 were estimated in order to determine the one that best fits the data. The implicit form of the estimated model is presented as:

$$Y_i = f(X_1, X_2, X_3, X_4, X_5, X_6, e_i) \quad 1$$

Where Y_i is the per capita income, X_1 is cost of treatment (₦), X_2 is cost of family labour (₦), X_3 is cost of hired labour (₦), X_4 is cost of other inputs (₦),

X_5 is number of days of incapacitation, X_6 is land area and e_i is the error term.

Results and Discussions

Rural households' socio-economic characteristics

Table 1 shows that about 75% of the respondents were male. Out of the respondents, 94.5% were married, 4.5% were divorced and 0.9% was single. Also, while only 1.8% of the respondents were between 21 and 28 years of age, those between 41 and 50 years constituted 44.5%. The minimum age was 30 years, while the maximum was 78 years. Average age was 49.51 years with coefficient of variation of 481.09 percent. The data also revealed that 6.4% of the farmers had no formal education, 57.3% had primary education, 30% had secondary education while only 6.3% had tertiary education. This implies that there is generally low level of education among the farmers.

Table 1: Frequency distribution of some socio-economic characteristics

Socio-economic characteristics	Frequency	Percent
<i>Age (Year)</i>		
21 – 30	2	1.8
31 – 40	20	18.2
41 – 50	49	44.5
51 – 60	23	20.9
61 – 70	13	11.8
≥ 70	3	2.7
<i>Sex</i>		
Male	82	74.5
Female	28	25.5
<i>Marital status</i>		
Single	1	0.9
Married	104	94.5
Divorced	5	4.5
<i>Educational status</i>		
No education	7	6.4
Nursery	1	0.9
Primary	62	56.4
Secondary	33	30.0
Tertiary	7	6.3
<i>Primary Occupation</i>		
Farming	101	91.8
Professional	9	8.2
<i>Cooperative membership</i>		
Cooperative membership	42	38.2
Non membership	68	61.8
<i>Household size</i>		
1 – 3	4	3.6
4 – 5	34	30.9
6 – 7	32	29.1
8 – 9	19	17.3
≥10	21	19.1

Also, 91.8% of the respondents interviewed had farming as their primary occupation, while 8.2% had other professions like tailoring, carpentry, brick-laying as their primary occupation but were also into farming as a secondary occupation. Many of the respondents (61.8%) did not belong to any cooperative society. The reason adduced for this included lack of full information about activities of cooperative societies and their non-willingness to take any unnecessary risk. Only 3.6% of the respondents had household size below 4 members. Average household size is 8.62 with standard deviation of 6.53. It implies that majority of the respondent raise large family size. This is because farmers often raise a team of family labour required to assist on their farms.

Health problems reported by the farmers

Table 2 shows the type of illness that farmers considered to be most prominent in their households. It showed that majority had malaria (40%). This is due to the dirty environment which serves as breeding space for mosquitoes. Also about 17.3% indicated tuberculosis as a common illness while another 17.3% had whitlow. These two sicknesses are as a result of the nature of their job [farming]. About 12.7% had cholera which is as a result of drinking unclean water and eating unclean food. Tuberculosis results when the farmers inhale dust in the course of performing their farming activities. Whitlow also results from infected hand injuries

Table 2: Most common types of sickness among rural households in Ondo State

Types of Sickness	Frequency	Percent
Tuberculosis	19	17.3
Whitlow	19	17.3
Guinea worm	6	5.5
Malaria	40	40.0
Pneumonia	3	2.7
Cholera	14	12.7
Diabetes	3	2.7
Hookworm	2	1.8
Total	110	100

Factors explaining households' welfare (per capita income)

Table 3 shows the coefficients of the estimated parameters in the regression equations. What we first did was to select the best functional form using the criteria of significance of the estimated parameters and coefficients of determination. At 5% level of significance, only days of incapacitation is significant in the linear functional form. Under the semi logarithm model, only the cost of family labour is significant at 5% level of significance. Under the double logarithm, five of the six variables are significant. Precisely, cost of family labour, cost of hired labour, cost of other inputs, days of incapacitation and land area are significant at least at 5% level of significance. With the exponential functional form, only two variables; cost of family labour and cost of hired labour are significant at 5% level of significance. Comparing the adjusted R square values of the four functions, the double logarithm has the highest value of 69.63% while the linear function has 58%, semi logarithm has 62% and the exponential has 13%. Considering the above statistics, we can rightly say that the double logarithm produced the best fit for the data and should be used for results interpretations.

Although statistically insignificant at 5 percent level of significance, the estimated parameters for the cost of treatment variable has negative coefficient. This implies that the cost incurred for treatment during sickness reduces the farmers' per capita income. This is because such income would have been invested for further production, instead of using it to settle hospital bills or for drugs during illness. The parameter of the cost of hired labour variable is statistically significant at 1 percent level and negatively signed. This implies that as the cost of hired labour increases, the per capita income of the farm households significantly reduces. This is due to the fact that the farmers, during illness will not want the work at the farm to stop and so will hire labourers to work for them. Cost incurred on these expenses significantly reduces the per capita income.

Family labour is positively signed and statistical significantly influence per capita income. This implies that as more members of the family participate in working on the family farm, the per capita income of the household will significantly increase. Cost of other inputs also has a positive coefficient and this implies that an increase in the use

of other inputs like fertilizers, chemical, seeds etc will lead to an increase in yield and this will lead to increase in households' per capita income. It can also be observed from the table that parameter for the days of incapacitation variable is negatively signed and it is statistically significant at 1 percent level. This implies that the research hypothesis has to be rejected. Therefore, as the number of days that farmers could not attend to normal farm activities increases, the per capita income significantly decreases. This can be explained from the viewpoint of scarcity of hired labour and timeliness of agricultural activities. When the farmer is not able to attend to normal farm activities, several forms of wastages reduce farm and non-farm incomes. Also, the parameter estimated for the land area variable is positively signed and statistically significant. This implies that as the land area increases, the per capita income of the farm household increases. This is a reflection of the relatively fertile status of the agricultural land in the study area. This may not be the case in a situation where the larger portion of the farm lands had been degraded.

Table 3: Determinants of per capita income

Function	Constant	Cost of treatment	Cost of family labour	Cost of hired labour	Cost of other inputs	Days of incapacitation	Land area	Adjusted R square
Linear	57677.27 (65300.08)	13.11481 (8.003329)***	25.45517 (22.10378)	19.13440 (13.13734)	-0.326139 (1.654710)***	1181.604 (559.8646)**	3043.351 (9723.547)	0.5585
Semi-logarithm	-39040.88 (269711.3)	14720.08 (29441.57)	8718.557 (4354.174)**	2435.217 (8295.520)	10723.50 (15304.13)	-1924.787 (969.8000)***	-15881.50 (42582.25)	0.6054
Double logarithm	5.031564 (1.195906)	-0.033309 (0.125034)	0.104280 (0.036576)*	-0.078763 (0.036098)**	0.558938 (0.077011)*	-0.042956 (0.012292)*	0.253095 (0.084871)*	0.6966
Exponential	10.35528 (0.355318)	5.50E -05 (4.36E -05)	0.000255 (0.000121)**	0.000164 (7.25E -05)**	1.06E -05 (9.07E -06)	0.004555 (0.003213)	0.077163 (0.052910)	0.2691

Standard errors are in the brackets, * = 1% level of significance, ** = 5% level of significance and *** = 10% level of significance

Conclusion and recommendation

This study revealed the health status of households in rural areas of Ondo state. The different types of diseases that are prevalent in the area were also identified and the nature of treatment adopted by the people was assessed. The number of days of incapacitation due to illness was also observed in the course of the study. The welfare of the households in the study area was assessed. Having established the important linkage between health status of rural farmers and their welfare status, some recommendation can be derived from the results. In a bid to combat health risk of rural households, national awareness of incidence of diseases and the impact on their income should be raised in rural areas through information dissemination, informal education and effective communication. This may be an important

factor given the low level of education of the respondents. Government should also ensure prompt and adequate provision of health facilities that is affordable to the rural dwellers. The health centre available in most of the rural areas is too far from them that they sometimes prefer to adopt self medication or native treatment. Most of the times, drugs are not available in the primary health centre and those that are available are not affordable. Access to inputs like chemicals, seeds and fertilizers should be enhanced because this is important for increasing farmers' income and their welfare. These inputs should be made available through subsidy channeled through the farmers' cooperative societies.

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A Proposed Model of Customer E-loyalty Measurement in Internet Banking

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Abstract: In recent years, Internet Banking (IB) applications are gaining popularity among retail banking customers. The long term success of these applications is however influenced by customer loyalty because it affects customers' perceptions about banks' innovative ability and customer caring intentions. This study has been developed for a proposed model of e-loyalty measurement for IB in Tejarat bank of Iran. For this purpose, after the comparative studies, both e-satisfaction and e-trust were identified as customer e-loyalty measurement model dimensions. This research is an applied field study and the method employed is descriptive and quantitative. The research sample is composed of 218 internet customers of Iranian Tejarat banks. The required data is gathered through stratified random sampling method using a researcher-made questionnaire with a 0.9406 Cronbach alpha coefficient. Also for the effectiveness of each model dimensions, it was used linear regression. The results introduced both major dimensions satisfaction with coefficient of 0.262 in both indicators of website design and customer service, and e-trust with coefficient of 0.591 in both indicators of fulfillment/ reliability and security/privacy. Finally, with emphasis on the effectiveness of these two dimensions were proposed approaches to improve the E-loyalty.

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Keywords: E-loyalty; E-satisfaction; E-trust; Internet Banking

1. Introduction

The evolutionary nature of Information technology and the vast application of internet in the last decade, has given a face lift to the banking industry in financial service distribution management (Maenpaa et al., 2008). One crucial instrument for developing E-commerce is the existence of E-Banking systems which can facilitate E-Commerce activities along with the world's fiscal and monetary systems (Durkin et al., 2008). E-banking includes conventional banking using the internet as a tool for giving bank services such as money transfer, bill payment as well as other new services like E-Payment. In today's highly competitive financial services market, it is essential that banks continue to develop their internet banking capabilities by identifying strategies to increase adoption, access, and usage of this low-cost channel (Yousafzai & Yani-de-Soriano, 2011). Most of the banks these days don't need more branches they can serve thousands of customers around the world through the internet. In order to attract customers and their satisfaction in this method, banks need to provide information security in their internet services. E-Banking supports E-Commerce applications such as immediate transactions and stock market activities through the internet (Lee, 2009) and plays an important role in electronic payments (Durkin et al., 2008). Moreover it increases customers' loyalty and satisfaction and

lowers both customers and banks costs by providing adequate information.

Besides all opportunities internet has provided, it also introduced new challenges in the field of competition to the banks and financial institutions all around the world. The internet banking services is seen as a high-risk service that contains several uncontrollable elements, any of which could lead to money and/or privacy loss (Lifen Zhao, 2010). E-Banking needs to deal with lots of problems. Some of them are the lack of technical infrastructure, culture and training, legal and judicial challenges in E-Banking, Lack of E-Customers loyalty, limited bank services, international sanctions, Managerial complications, lack of insurance for electronic payments and security issue (Abtahi and MeraatNia, 2008).

One important issue in E-Banking and E-Commerce is to maintain internet customers by increasing their E-Loyalty. Customers' loyalty is their powerful interest to the organization and directly affects the organization's profitability. Numerous studies have been done on customers' loyalty but most of them are concentrated on conventional banking. E-Loyalty however is an effective, key factor in E-banking success. Since the products offered to the customers of a bank are more or less standardized in nature, banks are feeling an increasing need to differentiate themselves from the

competitors on other criteria that can influence customer satisfaction and loyalty (Ganguli and Roy, 2011). “Floh, and Treiblmaier (2006) found that website quality and service quality of internet banks would have an effect on trust and satisfaction, which, in turn, influence customer loyalty. E-Loyalty can help customer maintenance as well as reducing the costs of attracting new customers and consequently help electronic companies and organizations’ profitability”. Therefore the lack of E-Loyalty is introduced as one of the important challenges in E-banking. A review on the literature can help us to recognize different dimensions of E- Loyalty.

“De Oliveira (2007) emphasized on the relationship between the quality of services and E-Loyalty in his study and introduced accessibility, user friendliness, privacy and security, Graphical interface, accountability and adequate responsiveness as effective factors in E-Loyalty. “Anderson and Srinivasan (2003) indicate that although E-Satisfaction has an impact on E-loyalty, this relationship is moderated by (a) consumers’ individual level factors and (b) firms’ business level factors”. Among consumer level factors, convenience motivation and purchase size were found to accentuate the impact of E-Satisfaction on E-loyalty. With respect to business level factors, both trust and perceived value, as developed by the company, significantly accentuate the impact of E-satisfaction on E-loyalty. “Xue, and Liang (2005) suggested service quality, trust, switching cost and staff loyalty as the customers’ E-Loyalty determining factors”. “Clark (2007) has introduced core offering, demographics, the marketplace, share of wallet, elasticity level and satisfaction as the six major factors that play key roles in influencing the loyalty and the commitment of customers”. According to “Devaraj et al. (2003) information content, transaction security, responsiveness, customer data recovery, visual attractions, download time, return process speed, ease of navigation, payment options and reputation are the factors which can affect E-loyalty and a business site’s success”.

In this study we have used the website design and customer service as the indicators of E-Satisfaction and fulfillment/ reliability and security/privacy as the indicators of E-Trust to measure customer E-loyalty for Tejarat bank of Iran. For this purpose after introducing E-Satisfaction and E-Trust and their effective indicators conceptual framework of the study is explained. Then we go through the research method including data gathering method and statistical tests. After that data analysis is done using our model and Finally we go through the concluding remarks.

2. Material and Methods

2.1 E-loyalty

According to “Oliver (1999) customers exhibit loyalty toward a brand or product when they insist on buying them despite other companies’ products”. So, E-loyalty is customers tendency to visit a certain business site due to the positive experiences they have had or their future expectations (Anderson and Srinivasan, 2003). When business competitions in electronic world started by just a few mouse clicks and customers became able to compare all the available options, E-Loyalty has become a concern for companies (Javad, 2009). E-Loyalty has been measured through various dimensions in different studies. In the present study E-Satisfaction and E-Trust are introduced as two effective dimensions of E-Loyalty.

2.2 E-Satisfaction

E-Satisfaction is defined as the customer’s pleasant sense due to his/her positive experience with an electronic business organization. Different studies have shown that customers may need more satisfaction and can easily move toward more convenient financial institutions despite their positive experience with the current bank or financial institutions. Therefore, E-Satisfaction can influence E-Loyalty in electronic business; E-Satisfaction in turn is influenced by website design and customer service (Kim, 2005).

Based on an in-depth review of relevant literature, we can say first that satisfaction in terms of previous interactions is considered a key antecedent of customer loyalty about an online financial services provider (Casaló et al., 2008).

Website design refers to all interactive components of the website with the customer such as user navigation, providing adequate information and customer order processes. Information content, information nature and the type of graphic are the other components of this indicator which can positively affect E-Satisfaction (Kim, 2005). Web site design quality showed positive direct effects on pleasure, arousal, and perceived information quality and on satisfaction (Young and Hyunjo, 2012). Furthermore measuring internet banking service’ user satisfaction through Technology Acceptance Model (TAM) shows that factors like website user friendliness, ease of access, and adequate information can also influence E-Satisfaction (Pikkarainen et al., 2004).

Customer service refers to responding to customers in the minimum time possible. A survey including 100 American retailers’ websites shows that lack of immediate response specifically for e-mails is a prevalent phenomenon amongst online product sellers (Kim, 2005). Overall customer service satisfaction and technical customer service

satisfaction dimensions fully mediated the relationships between customer service skills and customer loyalty (Amjad et al., 2011). So customer service is an effective indicator on E-Satisfaction.

2.3 E-Trust

E-Trust is defined as the degree of customer security in online transactions (Ribbink et al., 2004). According to “Byoungho et al. (2008) E-Trust is the customers’belief in an organization’s word which can assure them (i.e. credibility) that the organization would not misuse customers’ vulnerable situation”. Doing business through the internet always involves risk, the most important cause of which is the lack of trust between the organization and the customer.

This happens because they are not physically present at marketplace. In every mutual relationship trust is the first things’ first. As matter of fact, if an organization wants to succeed in providing electronic services, it needs to build trust in its relationship with customers in the first place. The lack of trust can be overcome with a better understanding of factors that can boost customers’ trust for e-banking (Yap et al., 2010). E-Trust itself is affected by the two indicators: fulfillment/ reliability and security/ privacy (Kim, 2005).

Fulfillment/ reliability is defined as receiving appropriate service in a specific time period and providing accurate information for inside website services in a way that fulfills customers’ expectations (Wolffingbarger and Gilly, 2003). One important thing which can result in E-trust is fulfillment/reliability that is receiving the right service at right time (Gummerus et al., 2008). Then different studies imply that fulfillment/reliability can influence E-Trust for an online retailer.

Security/privacy refers to website security in its interactions with customers, credit card payments and encrypting shared data. Guarantying privacy/security plays a crucial role in building E-Trust, since it can reduce customers’ concern about any kind of personal information abuse. Therefore if a degree of security is guaranteed and it can meet customers’ expectations, the customer would share a part of his/her personal information. There is growing concern regarding security issues and the use of information given online in terms of the privacy of personal information and the unintended uses of it (Roca et al., 2009).

Regarding dimensions and indicators mentioned above, our conceptual model is presented in figure 1.

Table 1. The results of KMO and Bartlet Test

KMO	Significance level (Bartlet)	Chi-square statistic	Error
0.896	0.000	3027.475	0.05

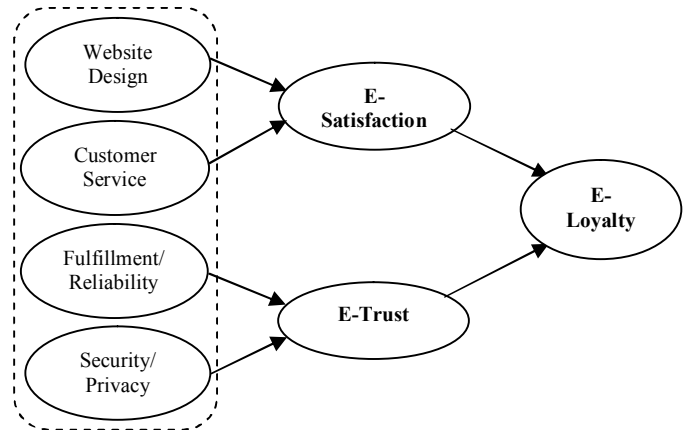


Figure 1. Conceptual Model

2.4 Research Method

This research is an applied field study and the method employed is descriptive and quantitative. Our statistical society includes all internet bank service customers of Iranian Tejarat Bank in the year 2011. The research sample is composed of 218 internet customers of the bank. The required data is gathered through stratified random sampling method using a researcher-made questionnaire with five demographic (gender, marital status, age, education and monthly income) and 25 specialized questions. Bank’s internet customers’ point of view about the factors which can influence E-Loyalty is gathered according to Likert scale using the bank’s website and customer e-mails.

In order to check the questionnaire validity, besides setting the questions according to the theoretical basis we took advantage of the opinions of the supervisor, the advisor and the experts in research and planning office of Tehran Tejarat bank. Moreover, the appropriate sample size, the required number of questions and correlation among them is checked using factor analysis and Kaiser-Meyer-Olkin (KMO) and Bartlet Tests. Table 1 contains the results.

As we can see KMO is greater than 0.05 which shows that the sample size is appropriate. The significance level of Bartlet test is equal to 0.000 which is smaller than 0.05 and approves the correlation among the variables and the factor analysis.

The questionnaire’s reliability is checked using Chronbach’s alpha. The results are represented in table 2.

Table 2. Reliability Coefficients

	Dimension	Chronbach’s alpha
1	E-Satisfaction	0.896
2	E-Trust	0.972
	All the questions	0.9406

As the table shows Chronbach’s alpha is greater than 0.7 which means that the questionnaire is reliable enough.

2.5 Data Analysis

In the present study data analysis is done using both descriptive and inferential statistics. First general characteristics of the subjects (gender, marital status, age, education and monthly income) are represented through descriptive statistics and frequencies. The distribution of our sample regarding demographic variables is illustrated in table 3.

Table 3. Sample Distribution Regarding Demographic Variables

Demographic Variable	Stratum	Frequency	Percentage
Gender	Male	131	65
	Female	69	35
Marital Status	Single	109	54.5
	Married	91	45.5
Age	21-25	56	28
	26-30	103	51.5
	31-35	19	9.5
	36-40	12	6
Education	41-50	10	5
	Diploma	14	7
	Post Diploma	18	9
	Bachelor’s	65	32.5
Monthly Salary	Master’s	99	49.5
	Doctoral	4	2
Monthly Salary	<100	21	10.5
	100-300	10	5
	300-500	68	34
	500-700	44	22
	700-1000	32	16
	>1000	25	12.5

As we can see in table 4, for the gender variable the highest percentage belongs to men with 65 per cent, for marital status the highest percentage belongs to singles with 54.5 percent, for the age variable the highest percentage belongs to 26-30 year olds with 51.5 per cent, for education the highest percentage belongs to MA degree graduates with 49.5 per cent, and finally for the monthly income the highest percentage belongs to the people whose income was between 300 to 500 thousand Toomans, with 34 per cent.

In order to test our hypotheses Pearson correlation test and to check the relation between dimensions and indicators linear regression is used. The multiple linear regressions’ validity is approved using different tests like Anova test (for linearity), Durbin-Watson test (for auto correlation of error terms), Kolmogorov-Smirnov test (for error terms’ normality) and finally Collinearity test. Then the regression equation is extracted from non-standard coefficients. We can use this equation in order to

forecast the dependent variable regarding explanatory variables. Finally according to standard variables the degree of each variable’s effectiveness is determined. Table 4 represents the relationship among dimensions, indicators and E-Loyalty.

Table 4. Spearman Correlation Test results for the hypotheses

Hypotheses	coefficient	Error	Result
1. E-Satisfaction has direct positive effect on E-Loyalty.	0.605	0.05	Relationship exists
2. E-Trust has direct significant effect on E-Loyalty.	0.680	0.05	Relationship exists
3. The website design has positive significant effect on E-Satisfaction	0.443	0.05	Relationship exists
4. Customer service has positive significant effect on E-Satisfaction.	0.456	0.05	Relationship exists
5. Fulfillment/Reliability has positive significant effect on E-Trust.	0.554	0.05	Relationship exists
6. Security/Privacy has positive significant effect on E-Trust.	0.566	0.05	Relationship exists

The results show that a significant relationship exists among explanatory and dependent variables.

Table 5 represents the degree to which each dimension and its indicators affect E-Loyalty.

Table 4. Regression results

Hypotheses	t-Statistics	The degree of effectiveness	Result
1. E-Satisfaction has direct positive effect on E-Loyalty.	4.53 7	%26.2	Hypothesis confirmed
2. E-Trust has direct significant effect on E-Loyalty.	10.2 6	%59.1	Hypothesis confirmed
3. The website design has positive significant effect on E-Satisfaction	6.89 4	%41.2	Hypothesis confirmed
4. Customer service has positive significant effect on E-Satisfaction.	6.04 3	%36.1	Hypothesis confirmed
5. Fulfillment/Reliability has positive significant effect on E-Trust.	8.45 5	%44.5	Hypothesis confirmed
6. Security/Privacy has positive significant effect on E-Trust.	8.47 5	%44.6	Hypothesis confirmed

As the table shows, all hypotheses are confirmed. Both E-Satisfaction and E-Trust have positive direct effect on E-Loyalty. In addition E-Trust with 0.591 coefficient and its indicator privacy/security with 0.446 coefficient have the most powerful effects on E-Loyalty.

3. Results

The appearance of electronic trade and market introduced new concepts like E-Satisfaction, E-Trust and E-Loyalty. E-Loyalty is a key element in internet banking success. In the virtual world banks need to be armed with the knowledge and new solutions to increase their customers' satisfaction, trust, security and therefore E-Loyalty. So, both the banks and the society can take advantage of recognizing the effective factors on E-Loyalty, while the lack of knowledge about these factors can bring about big challenges in the banks' way to success. The results of testing hypotheses indicate that E-Satisfaction and E-Trust both have positive significant effect on E-Loyalty.

Regarding that the website design is an effective factor on customers' E-Satisfaction, the system interface can be designed in a way that it can reduce the search costs for the customers and the time needed for processing information in order to increase this indicator's effect. This indicator is aligned with website design quality in Young & Hyunjoo's study (2012) and with Critical Success Factors (CSF) in Devaraj et al. study (2003). Customers may be dissatisfied with a company's service, before the organization or institute provides online services. In these cases it would be hard for the company to bring the customers back after providing the service. This finding is aligned with the customer service in Amjad et al. study (2011). Our results suggest that fulfillment/reliability is a key factor in building and retaining E-Trust. In other words, an customers must believe, the service they receive from an electronic organization would not waste their time and money. This indicator in the present study is aligned with reliability and accountability in the studies of De Oliveira (2007). Being sure about your privacy and security is another important determinant of E-Trust. The effect of this indicator on E-Trust can be empowered by reducing customers' concern about the exposure of his private information. This indicator is aligned with security in the studies of Roca et al. (2009) and De Oliveira (2007). The degree to which each dimension and its indicators can affect E-Loyalty is illustrated in figure 2.

4. Discussions

The results of the present study suggest many implications for the managers of different industries, business website designers and internet bankers. Regarding that more than 90 per cent of the

customers have academic education, more than 50 percent of them earn more than 5 and 10 million Rials, and the highest frequency belongs to 20-35 year olds, the banks need to focus on these groups. So, they have to have adequate plans to attract and retain them, provide them with up to date services, and use appropriate promotional and encouraging plans (such as free internet subscription) to improve E-Loyalty.

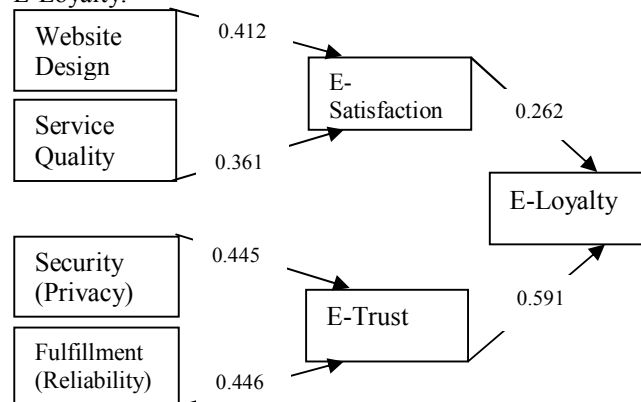


Figure 2. The effect of explanatory variables on the dependent variable

Regarding our variables and hypotheses, promoting the electronic service culture, providing useful information and preparing informative advertisement about the issue, taking user friendliness into accounting in designing internet services and flexibility can improve customer service indicator for Iranian Tejarat Bank. In addition, designing the website in a way that simplifies using the internet banking service for the customers, providing comprehensive, up to date and accurate information about the bank improves the website design indicator.

In order to improve fulfillment/reliability it would be helpful for Iranian Tejarat Bank to provide its customers with enough information about the technology used in internet banking. It can help the bank to assure its customers that the probability of any mistake would be the least possible.

Finally making customers believe that their privacy and personal information are protected as the conventional banking system as well as informing them about the protocols, mechanisms and the degree of security their password have would help Iranian Tejarat Bank to improve security/privacy.

Regarding the restriction of selected research domain as our main limitation we suggest future E-Loyalty researchers to focus on other sectors of the industry. Moreover, considering that customers have different understandings about the products and services of various industries, it is suggested to

classify the process development of E-Loyalty in several industries and evaluate customers' different understandings. Finally, since website design is an important indicator of E-Satisfaction, it is suggested to the future researchers to study more different aspects of this indicator.

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