Comparative study between available used Newcastle Vaccines in market on broiler chickens

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Abstract: For studying the effect of 3 common Newcastle disease (ND) vaccines (La Sota, Avenew and Vetapest) in the Egyptian broiler chicken flocks; 200, one day old commercial broiler chicks in four equal groups reared on commercial ration. Groups 1, 2, and 3 were received La Sota, Avenew and Vitapest ND vaccine; respectively; while birds of group 4 was kept as negative control. At 36 days old (10 days after last vaccination as vaccination design start at 5th days of age with Hitchener B1 vaccine then at 16th days of age 1st vaccination takes place according regmin of each groups and 2nd vaccination takes place at day 26 of age as postering per each groups) all groups were challenged 10 days after last vaccination with 10^7 EID_{50} /ml of velogenic ND virus. Results of **HI test** revealed that all vaccines produce positive protective antibody titer start to increase after 1st vaccination as the highest was Vetapest (10.25 avarage) followed by Lasota (8.25 avarege) then Avenew (6.5 avarege) then continue after 2nd vaccination the highest was Vetapest (10.5 averge) followed by Lasota (10.125 avarege) then Avenew (9.875 avarege), while after challenge the titer slightly decreased amonge all vaccinated groups the lowest was Lasota (7.125 avarege) then followed by Avenew (7.5 averge) while the highest was Vetapest (7.75 avarege). All challenged vaccinated groups showed 100% protection against20% in control negative group. The histopathologically changes were recorded in Avenew vaccinated birds as the tracheal sections appeared normal, while mild infiltration of the submucosa with mononuclear cells was detected in some examined sections. Liver in most examined sections revealed normal hepatocytes with normal organization ,but few examined sections showed minute focal area of hepatocellular necrosis infiltrated by mononuclear cells. Proventriculus showed normal mucosa with normal proventricular glands with no inflammatory reactions. On the other hand the two other vaccinated groups showed focal area of deciliation with moderate infiltration of the submucosa with mononuclear cells and submucosal edema, while liver showed thickening of the perihepatic capsule as well as congestion of the blood vessels with focal area of hepatocellular necrosis infiltrated by mononuclear cells, there was portal edema and hyperplasia of epithelial lining bile duct. Proventriculus showed infiltration of the mucosa with mononuclear cells . After challenge with virulent virus the condition become more sever in all examined tissue samples as trachea showed necrosis of the mucus gland with massive infiltration of the submucosa with mononuclear cells while liver showed different types of necrosis, as sporadic cell necrosis with pyknotic nuclei , centrilobular necrosis of hepatocytes and Proventriculus showed heavy infiltration of the mucosa with large number of inflammatory cells mostly heterophils and mononuclear cells as well as submucosal edema. Histopathological examination revealed that mild lesion in Avenew as well as moderate in both La Sota and Vetapest. These lesions become more sever following challenged with field virus as in case of Avenew.

It could be concluded that the use of live vaccine could protect the birds from clinical signs when challenged with field virus with occurrence of microscopic lesions which was milder in Avinew. This finding may be depending on origin of the strain either respiratory or enteric one

[Kh. M. ELbayoumi; Eman R.Hassan; K.M.Mahgoup; Zeinab M. S. Amin Girh; and Hoda M. Mekky. **Comparative study between available used Newcastle Vaccines in market on broiler chickens**. *Report and Opinion*. 2012;4(4):46-51]. (ISSN: 1553-9873). http://www.sciencepub.net/report. 9

Key words: ND live vaccines, HI test, Histopathology, challenge test.

Introduction:

Newcastle disease is highly contagious, septcaemic, fatal and destructive disease which attacks chiefly chickens and turkeys ***(Cole and Hult, 1961)***, occasionally human being ***(Chang, 1981),*** ducks ***(Higgins, 1971)*** and even wild birds maybe also infected with the virus. The disease transmitted by either inhalation or ingestion ***(Alexander, 1988)*** the disease cause great economic losses due to high mortality, reduction of weight gain and drop in egg production ***(Alexander, et al., 1985; Leslie, 2000 and Musa et al., 2010)***. Histopathological changes following NDV infections are related to the virulent pathotypes ***(McFerran and McCracken, 1988)*** and may occur in various organs during course of infection ***(Cheville et al., 1972*** *and* ***Stevens et al., 1976)*** not only virulent strains causing infection but also lentogenic strains may cause apparent disease ***Beach,(1944)*** also ***Hitchner and Johnson(1948)*** found that infections caused by viruses of the lentogenic pathotype may cause mild or inapparent respiratory disease (Hitchner's form), also those

lentogenic strains may infect the gut causing no obvious disease ***McFerran and McCracken,(1988)*** which known by Asymptomatic – enteric form ,some live commercial vaccines are of this pathotype.

Many serological tests are useful tool for assaying the antibody responses of commercial broiler chickens vaccinated against ND such as HI test and ELISA test ***Snyder et al., (1983)*** and ***Cevelic- Cabrilo et al., (1993).***

Prevention and control of the disease depend mainly on strict hygienic measures and proper vaccination programme, taken in consideration the fact of ND vaccination usually protects the birds from more serious symptoms but virus replication and shedding may still occurs ***(Guittet et al., 1993*** *and* ***Alexander et al., 1999) .***

Therefore; the present work was planned to compare the effect of 3 out of the most widely used vaccines (La Sota, Avenew and Vetapest) on broiler chicken using HI test as a model of serological monitoring and histopathological examination to find microscopical changes in organs of vaccinated chickens.

2. Material and Methods:

Experimental birds:

Two hundred (200), one day old, Ross fed commercial ration and reared under strict hygienic measures.

Vaccinal viruses:

1. La Sota vaccine: batch no. A5147, 1000 doses, and titer of $10^{7.5}$ EID_{50} /dose.
2. Avenew vaccine: batch no. L374462, 1000 doses, and titer of $10^{7.5}$ EID_{50} /dose.
3. Vitapest vaccine: batch No. 1806Z3U1B, 1000 doses, and titer of $10^{7.5}$ EID_{50} /dose.

Virulent NDV virus:

Used for challenge the chicks throughout the experiments was a locally field isolate, velogenic visrotropic Newcastle disease virus (VVNDV). It was isolated and identified by ***Sheble and Reda (1976)***. It had an infective titer of 10 EID**50**/ml kindly supplied by doctor khaled Mohamed mahgoub, National Research Center.

Estimation of virus infectivity:

Infectivity of used vaccines and challenge virus was done according to ***Anon (1971)*** and the embryo infected dose 50 (EID_{50}) was calculated according to ***Reed and Muench (1938)***.

Haemagglutinating antigen:

It was prepared according to methods of ***Allan et al. (1973).***

Serum samples for HI test:

Prepared from blood samples collected from wing vein.

Chicken red blood cells:

Red blood cells (RBCs) from susceptible adult birds were collected on 4% sodium citrate as anticoagulant. The RBCs were washed three times with phosphate buffered saline (PBS) at PH 7.0 – 7.2.

Physiological saline:

Prepared and autoclaved according to ***Cruickshank (1975)*** then stored at 4˚c till use.

Tissue samples for histopathology:

Tissue specimens from trachea, liver; and proventriculus of experimentally infected, vaccinated and control chicks were fixed in 10% neutral formalin solution for histopathological examination.

Formalin saline solution:

10% formalin in saline was used for preservation of the collected tissue specimens for histopathological examination.

Haemagglutination inhibition (HI) test:

The test was carried out according to the standard procedure described by ***Majiyagbe and Hitchner (1977)*** the end point were estimated according to scheme described by ***Kaleta and Siegmann (1971).***

Histopathological studies:

Samples were collected and preserved in 10% natural formalin. The specimens were processed, stained by Hematoxylin-Eosine (H&E) stain which was prepared according to ***Culling (1973)*** and examined microscopically for any evidence of histopathological changes.

3. Result and discussion

Results of **HI test** revealed that all vaccines produce positive protective antibody titer start to increase after 1st vaccination as the highest was Vetapest (10.25 avarage) followed by lasota (8.25 avarege) then Avenew (6.5 avarege) then continue after 2nd vaccination the highest was Vetapest (10.5 averge) followed by Lasota (10.125 avarege) then Avenew (9.875 avarege), while after challenge the titer slightly decreased amounge all vaccinated groups the lowest was lasota (7.125 avarege) then followed by Avenew (7.5 averge) while the highest was Vetapest (7.75 avarege. The recorded result of HI test revealed that all vaccines give positive titer increased after 1st and 2nd vaccination the highest was in group receive vetapest vaccine followed by La Sota and finally avenew and all of them give protective titer. After one week post challenge the titer slightly decreased in all groups these may due to neutralization of some protective antibody the obtained result was matched with ***Giambrone and closer (1990) and Madkour et al. (1992).*** Also all vaccinated chicks found to resist challenge with the

virulent virus 10 days after last vaccination. The obtained results are summarized in table (1).

Challenged vaccinated groups showed 100% protection while, the control group showed 40% protection.

Histopathological examination showed that all vaccines cause changes in examined tissue section (trachea, liver and proventriculus) but in various degrees as in case of Avenew trachea appeared with normal mucosa and submucosa. In some examined sections showed mild infiltration of the submucosa with mononuclear cells (fig. 1) the liver in most examined sections revealed normal hepatocytes with normal organization ,few examined sections showed minute focal area of hepatocellular necrosis infiltrated by mononuclear cells(fig. 2). Portal area revealed mild infiltration with mononuclear cells (fig.3) while proventriculus showed normal mucosa with normal proventricular glands with no inflammatory reactions demonstrated in these cases. On the other hand the two other vaccinated groups with La Sota and vetapest are nearly the same as examined tracheal section showed focal area of deciliation with moderate infiltration of the submucosa with mononuclear cells(fig.4) and submucosal edema, while liver showed thickening of the perihepatic capsule by faint pink edematous fluid as well as congestion of the blood vessels (fig. 6) with focal area of hepatocellular necrosis infiltrated by mononuclear cells(fig. 7), there was portal edema and hyperplasia of epithelial lining bile duct (fig. 8). Proventriculus showed infiltration of the mucosa with mononuclear cells (fig. 9).

After challenge with virulent virus the condition become more sever in all examined tissue samples as trachea showed necrosis of the mucus gland with massive infiltration of the submucosa with mononuclear cells (fig.10) while liver showed different types of necrosis, as sporadic cell necrosis with pyknotic nuclei (fig.11), centrilobular necrosis of hepatocytes (fig.12) and large area of hepatocellular necrosis that infiltrated by mononuclear cells. Portal area revealed hyperplasia of epithelial lining bile duct and formation of newly formed bile ductuoles as well as portal fiberoplasia (fig13). Proventriculus showed heavy infiltration of the mucosa with large number of inflammatory cells mostly heterophils and mononuclear cells as well as submucosal edema (fig.14 and 15).this result was matched with ***Brandly and Hanson (1967)*** who noticed that, in vaccinated birds against ND, the tissues and adjacent lymphoid aggregates of heart, liver and proventriculus occasionally show necrotizing and haemorrhagic lesions. Also they stated that among birds which survive challenge following vaccination a considerable birds show histopathological changes in various organs. Also ***Mohammadamin and Qubih (2011)*** found that there was less post – vaccination reaction with the enteric vaccinal strain instead of a respiratory vaccinal strain of ND which also was matched with our result of those of Avenew when compared with La Sota and Vetapest.

It could be concluded that the use of live vaccine could protect the birds from clinical signs when challenged with field virus with occurrences of microscopic lesions also live vaccine varing from each other according origin of the strain either respiratory or enteric one and both induce histopathological changes anyhow this need further investigation.

Table (1): HI antibody titers post vaccination and challenge as well as protection rate in vaccinated and control chicken groups.

Group no	Vaccine	Mean HI titers			Challenge test
		Post vaccination		Post challenge	Protection rate
		1st Week	2nd week	1 week	
1	La Sota	8.25	10.125	7.125	100%
2	Avenew	6.5	9.875	7.50	100%
3	Vetapest	10.25	10.50	7.75	100%
4	Control -ve	5.32	4.875	3.122	20%

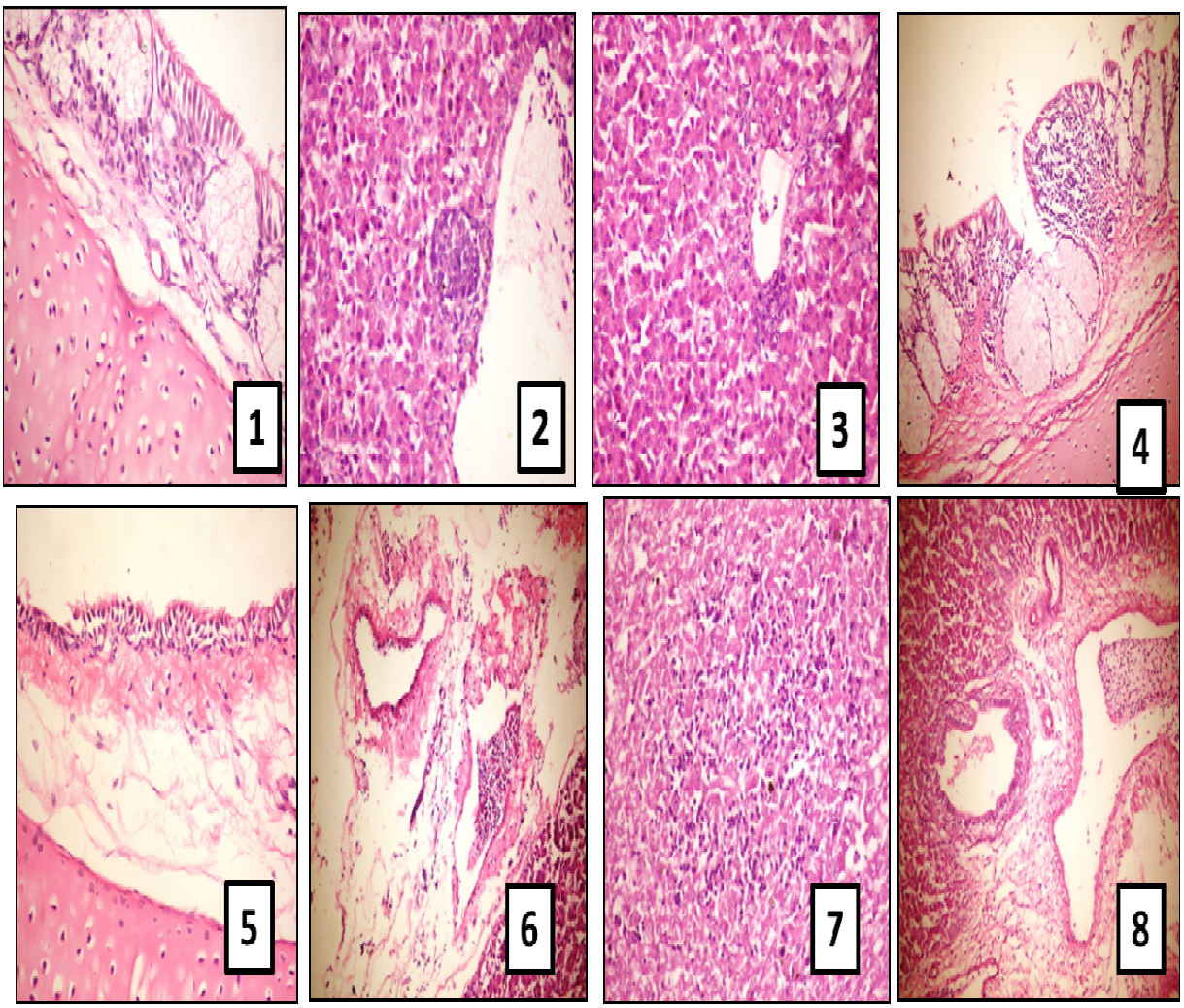

Fig.(1): trachea showed mild infiltration of the submucosa with mononuclear cells(H&E X400).
Fig.(2): liver showed minute focal area of hepatocellular necrosis infiltrated by mononuclear cells (H&E. X400).
Fig.(3): liver showed mild infiltration of portal area with mononuclear cells (H&E X400).
Fig.(4): trachea showed focal area of deciliation with infiltration of the submucosa with mononuclear cells (H&E X200).
Fig.(5): trachea showing submucosal edema (H&E X400).
Fig.(6): liver showing thickening of the perihepatic capsule with congestion of blood vessles and faint pink edematous fluid (H&E X200).
Fig.(7): l iver showing large focal area of hepatocellular necrosis infiltrated by mononuclear cells (H&E X400).
Fig.(8): liver showing portal edema and hyperplasia of epithelial lining bile duct (H&E X200).

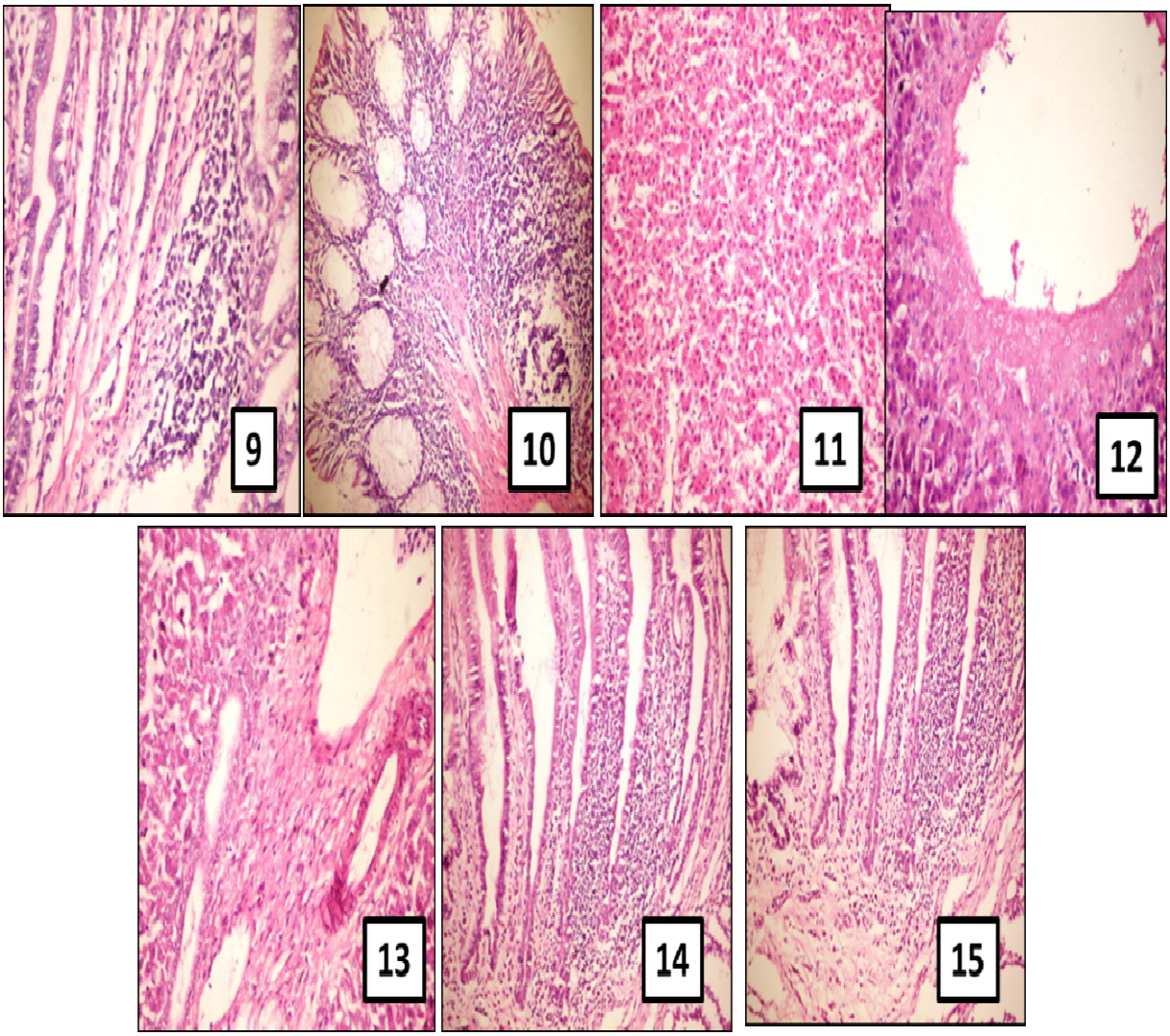

Fig.(9): proventriculus showing infiltration of the mucosa with mononuclear cells (H&E X400).
Fig.(10): trachea showing necrosis of the mucous gland(arrow) with massive infiltration of the mucosa with mononuclear cells(H&E X200).
Fig.(11): liver showing sporadic cell necrosis with pyknotic nuclei(H&E X400).
Fig.(12): liver showing centrilobular necrosis of hepatocytes(H&E X400).
Fig.(13): liver showing portal fibroplasia, oval cell hyperplasia and formation of newly formed bile ductuoles(H&E X400).
Fig.(14): proventriculus showing heavy infiltration of mucosa with inflammatory cells mostly heterophils and mononuclear cells(H&E X200).
Fig.(15): proventriculus showing heterophils and mononuclear cells infiltrating the mucosa as well as submucosal edema (H&E X200).

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References:

Alexander,D.J.(1988):Newcastle disease:methods of spread. In D.J. Alexander, (ed.) Newcastle Disease kluwer academic publishers: Boston MA, 256-272.

Alexander,D.J.; Manvell, R.J.; Banks, J.; Collins, M.S.; Parsons, G.; Cox, B.; Frost, K.M.; Speidel, E.C.; Ashman, S. and Aldous, E.W. (1999): Experimental assessment of the pathogenicity of

the Newcastle disease viruses from outbreaks in Great Britain in 1997 for chickens and turkeys and the protection afforded by vaccination. Avian pathol. 28:501-512.

Alexander, D.J.; Wilson, G.W.; Russell, P.H.; Lister, S.A. and Parsons, G. (1985): Newcastle disease outbreaks in fowl in Great Britain during 1984. Vet.Rec.,117:429-434.

Allan,W.H., Lancaster, J.E., and Toth,B.(1973): The production and use of ND vaccines P.53., F.A.O., Rome Italy 1115.

Anon, (1971): Methods of examining poultry biologics and for identifying and quantifying avian pathogens. Nat. Acad. of Sci., Washington, D. C.

Brandly C.A. and Hanson R.P.(1967): Diseases of pultry" 5th ed., 633-674.

BeachJ.R.(1944): Proc. Soc. 46 th Ann. Meet. U.S. Livestock Sanit. Ass. , 177-184.

Cevelic – Cabrilo, V.; Mazua,H.; Bidin,Z. and Ragland,W.(1993): Correlation of haemagglutination inhibation and enzyme linked immunosorbant assays for antibodies to Newcastle disease virus. Avian Pathol.(1992),21(3):509-512.

Chang, P.W.(1981): Newcastle disease. In G.W. Beran(ed.), CRC hand book series in zoonoses.section B:viral zoonoses volume II. CRC Press: Baton Raton, 261-274.

Cheville,N.F., stone, H.; Riley, J. and Ritchie, A.E. (1972): Pathogenesis of virulent Newcastle disease in chickens.J Am Vet Med Assoc 161:169-179.

Cole,R.K. and Hult,F.B. (1961): Genetic differences in resistance to ND Avian Dis.,5:205-214.

Culling C.F.A.(1973): Handbook of histopathological and histochemical techniques, 3rd ed. Butter worths, London, Boston.

Cruickshank, R.; Duguid, J. P.; Marmion, and Swain, R. H. A. (1975): Medical Microbiology, 12th Ed., Vol. 11, Churchill Lingstone, Limited Edinburgh, London and NY.

Giambrone, J.J. and Closser, J. (1990): Effect of broiler vaccination on immunization of progeny against ND. Avian Dis.,34:114-119.

Guittet, M.; Le Coq, H.; Morin, M.; Jestin, V.and Bennejean, G. (1993): Distribution of Newcastle disease virus after challenge in tissues of vaccinated broilers.In Procc. of the Xth world Vet. Poult. Asso. Congress, Sydney,179.

Higgins, D.A.(1 971): Nine disease outbreaks associated with myxoviruses among ducks in Hong Kong.Trop Anim. Health Prod. 3:232-240.

Hitchner,S.B. and P.Johnson.(1948): Avirus of low virulence for immunizing fowls against Newcastle disease (Avian pneumoencephalitis). Vet. Med. 43:525-530.

Kaleta,E.F. and Siegmann,O.(1971): Comparative studies on the demonstration of haemagglutinating inhibiting and virus neutralizating antibodies after vaccination against NewCastle disease. Arch. Grefugelk, 35, 79.

Leslie, J.(2000): NewCastle disease: outbreak losses and control policy costs.Vet.Rec. 146:603-606.

Madkour,M.S.;Mohamed,R.T.;Nadia,M.H.;Khodier, M.H.;ElAssile,S.;Ensaf,K.H.,ELBordiney,F.;Afaf ,A.H. and Reda,I.M.(1992): Humoral and local immune response to different NewCastle disease vaccine.Vet.Med.J.,40:111-118.

Majiyagbe,K.A.,and S.B.Hitchner(1977): Antibody Response to strain combination of NDV,as measured by HI.,Av.Dis.21(4) 576-584.

McFerran,J.B. and R.M.McCracken(1988): NewCastle Disease. Kluwer Academic Publishers:Boston,MA,161-183.

Mohammadamin,O.G.;Qubih,T.S.(2011):Histopathology of virulent NewCastle disease virus in immune broiler chickens treated with IMBO. Iraqi J. of vet. Sci., 25:1,9(En)-13(En).

Musa, I.W.;Abdu, P.A.;Sackey, A.K.B.;Oladele, S.B.;Lawal, S.;Yakubu, I.U. (2010): Outbreak of velogenic viscerotropic NewCastle Disease in broilers. Internat. J. of poult. Sci., 9:12,1116-1119.26.

Reed, L. J. and Muench, H. (1938): Simple method of estimating 50 per cent end point. Am. J. Hyg. , 27: 493-499.

Sheble, A. and Reda, I.M.(1976): Cited by khafagy,A.K.(1977).thesisM.V.Sc.,Fac.Vet.Med., cairo univ.

Snyder,D.B.,W.W.Marquadt,E.T.Mallinson,andE.Russek.(1983): Rapaid serological profiling by enzyme-linked immunosorbent assay. I Measurment of antibody activity titer against NewCastle disease virus in asingle dilution. Avian Dis.27:161-170.

Stevens, J.G.; Nakamura, R.M.; Cook, M.L. and Wilczynski ,S.P. (1976): NewCastle disease as amodel for paramyxovirus – induced neurological syndromes: pathogenesis of the respiratory disease and preliminary character-ization of the ensuing encephalitis. Infect Immun 13:590-599.