Comparative studies between Gum Arabic recognized as a natural prebiotic and Bifidobacterium as probiotic as potential cure for experimental bacterial infection in mice

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Abstract: Gum Arabic as natural prebiotic is food ingredients that stimulate the growth of useful bacteria which lives in the large intestine of the human being or animal and beneficial to the digestive system, body's immunity, disposal of poisons, fats and excreta. Probiotics are beneficial bacteria that can be found in various foods, or in the form of dietary supplements. Common strains of probiotics are Lactobacillus and Bifidobacterium families of bacteria. So present study aimed to investigate the effect of Gum arabic as natural prebiotics in comparison with Bifidobacterium animalis sub sp. lactis (Bb12) as probiotic and combination of prebiotic and probiotic when administrated as food additive when challenged the experimental mice with salmonella typhimurium. The results indicated that there were symbiosis between prebiotic and probiotic, gum Arabic is good for health as natural prebiotic especially in the large dose, Bifidobacterium lactis Bb12 also good for health and immune status while the combination between gum Arabic as prebiotic and Bifidobacterium as probiotic (5 g GA+ Bb12) was better whether the combined application of PRO and PRE (symbiotics) has synergistic and additive significant effects, they give good results than each one alone and improved the health conditions, physiological and immune response; cellular and humeral for challenged mice with salmonella typhimurium.

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Key words: Gum Arabic; Bifidobacterium; phagocytic activity; lysozyme;AST; ALT; Uric acid; Creatinine.

Introduction:

Gum arabic, known as acacia gum, chaar gund, char goond, or meska, is a natural gum made of hardened sap taken from two species of the acacia tree; Senegalia senegal and Vachellia seyal. The gum is harvested commercially from wild trees throughout the Sahel from Senegal to Somalia, although it has been historically cultivated in Arabia and West Asia. It is a complex mixture of glycoproteins and polysaccharides. It was historically the source of the sugars arabinose and ribose, both of which were first discovered and isolated from it (Renard et al., 2006; Calame, et al.,2008).

Gum Arabic as Prebiotic is food ingredients that stimulate the growth of useful bacteria which lives in the large intestine of the human being or animal since birth and beneficial to the digestive system, body's immunity, disposal of poisons, fats and excreta (Da Silva et al.,2013). Besides it negates the effects of harmful bacteria thus protecting from the diseases, cancer, diabetes and obesity. It is pointed out that the gum-Arabic, extracted from Acacia Senegal trees which is more common in Sudan than anywhere in the world, is one of the natural sources of the prebiotics similar to breast milk and the processed inulin (Gerstenzang et al.,2007; Calame, et al.,2008). Although very few studies have tested the health effects of acacia fiber, there's some evidence that it may offer certain benefits Chloé and Fabien (2014).

The intestinal microbiota is essential for triggering local and systemic responses favorable to the host health. The intestinal microbiota is composed of about 100 trillion bacteria and encompasses more than 1,000 species (Kassinen et al., 2007; Fletcher et al., 2009; Garrett et al., 2010; Shinohara et al., 2010; Kamada et al.,2013). It plays an important role in protection against pathogenic microorganisms, development and homeostasis of immune cells, digestion of polysaccharides that is indigestible by human enzymes and fat metabolism, among other functions (Holmes et al., 2011; Da Silva et al.,2013).

Present study aimed to investigate the effect of Gum arabic as natural prebiotics in comparison with Bifidobacterium animalis subsp. lactis (Bb12) as probiotic and combination of prebiotic and probiotic when administrated as food additive when challenged the experimental mice with salmonella typhimurium.

Material and methods:

Experimental animals and design:

A total number of 100 white albino 6-weeks old male mice, they were prepared for the experiment and acclimated for 1 week. They were divided into five groups each 20. First group was subjected Gum arabic in a concentration 5mg /200 ml, 2nd group was subjected Gum arabic in a concentration 15 g/200 ml, 3rd group administrated to Bifidobacterium animalis subsp. lactis (Bb12) (1x 10^7 colony forming unit (cfu)/g diet, 4th group administrated a combination
between *Bifidobacterium animalis* subsp. *lactis* (Bb12) 1x 10^5 cfu/g diet and Arabic gum in a concentration 5gm/, while 5th group was used as a control group subjected skinned milk only without Gum Arabic. All experimental groups administrated skinned milk with Gum arabic, (Bb12) and combination (Gum arabic 5 gm/200 ml) two times daily for 14 successive days before the challenge.

Challenge experiment:
Each mouse in all groups were orally challenged with 0.2 ml of the prepared *Salmonella typhimurium* suspension (1.5x10^9 CFU/ml) (Silva et al. 2004). The feces of the mice in each group were individually collected after the administration of the last dose of skinned milk with Gum arabic, (Bb12) and combination (Arabic gum 5 gm/200 ml) on the 2nd, 5th and 10th day post infection for detection of *Salmonella typhimurium*.

Strains:
*Salmonella typhimurium* ATCC 14028 kindly obtained from King Abdul Aziz Medical City (National Guard Hospital) in Jeddah, Saudi Arabia.

Arabic gum as prebiotic:
Arabic gum obtained from Khartoum, Sudan naturally grinded then mixed with milk powder (SM) for feeding mice along the experimental period. Gum arabic was prepared by dissolving the powdered form of Arabic gum in cold water for complete dissolving, making solutions in tow concentrations 5 gm, 15 gm in 200 ml water.

In vitro antagonistic activity of the prebiotic:
By a sterile swab, *Salmonella typhimurium* with a concentration of 1.5 x 10^8 CFU was spread over a nutrient agar plate and inoculated at 37°C for 24 h. about 10μl of each concentration of Gum arabic solution was inoculated in wells in the nutrient agar and incubated at 37°C for 24 hours. The inhibition zones was measured in mm. and recorded (Jakee et al. 2010).

Preparation of prebiotic milk:
Prebiotic milk was prepared from reconstituted skimmed milk SM (Silva et al,1999). 2 concentrations of Gum arabic were prepared and mixed with skimmed milk except of control. Skimmed milk prepared alone without Gum arabic.

*Salmonella typhimurium* fecal colony count:
Five grams of Pooled fresh feces were collected from each group separately; feces were diluted in sterile buffered saline (pH 7.2) viable *S. typhimurium* were determined (Jakee et al,2010).

Phagocytic assay:
Macrophage monolayers were obtained and prepared it were harvested from the peritoneal cavities of mice and were resuspended in Hanks’ balanced salt solution (HBSS) (Sigma Chemical Co., St. Louis, MO). The number of viable cells was determined by trypan blue dye exclusion and the coverslips were washed with HBSS. *Candida albican* particles were added to the monolayers in a 5:1 (particle: macrophage) ratio and the coverslips incubated at 37°C in humidified atmosphere. After 30 min, the coverslips were washed with HBSS, fixed in methanol, and stained with Giemsa. After drying, the coverslips were mounted on glass slides and examined microscopically. The percentage of cells with ingested particles was multiplied by the average number of particles per macrophage to calculate phagocytic index. At least 100 macrophages were counted per cover slip (Belline, et al., 2004).

Lysozyme assay:
The lysozyme concentration in the serum of mice was determined according to Schultz (1987) on the 2nd, 5th and 10th days after oral challenge with *S. typhimurium*.

Determination of (AST), (ALT), Uric acid and Creatinine:
At the end of the experimental period, blood samples were collected from tail vein in sterile test tubes centrifuged for serum separation at 1500 rpm for 15 min. to estimate AST, ALT according to methods described by Kapalan and Pesce (1996) uric acid and creatinine according to Tietz et al.(1995).

Statistical analysis
Data were represented as mean ± standard error (S.E). The data obtained in this study were analyzed statistically using a t-Student test. The differences were considered significant at * p < 0.05 and high significant**p < 0.01 as compared to the control group. The data were analyzed according to Sendecor (1964).

Results:
Antagonistic activity of Gum Arabic and Bb12 against *Salmonella typhimurium* in vitro:
Present study revealed that the antagonistic activity of Gum Arabic (prebiotic) and Bb12 (probiotic) against *Salmonella typhimurium* in vitro was recorded as the 1st group (5gGA) recorded 0.31mm inhibition zone while 2nd group recorded 0.42 mm and 3rd group recorded 9.65 mm and 4th group recorded 9.77 mm in contrast the control group revealed 0 mm inhibition zone table 1.

<table>
<thead>
<tr>
<th>group</th>
<th>Inhibition zone</th>
</tr>
</thead>
<tbody>
<tr>
<td>gp1 (5gGA)</td>
<td>0.31 mm</td>
</tr>
<tr>
<td>gp2 (10gGA)</td>
<td>0.42 mm</td>
</tr>
<tr>
<td>gp3 (Bb12)</td>
<td>9.65 mm</td>
</tr>
<tr>
<td>gp4 (5 g GA+ Bb12)</td>
<td>9.77 mm</td>
</tr>
<tr>
<td>gp5 (Saline control)</td>
<td>0 mm</td>
</tr>
</tbody>
</table>
**Salmonella typhimurium** colony count from fecal samples of mice:

From the obtained results it was noticed that in 2nd day after challenge the lowest bacterial colony count was recorded in the 4th group (SM + 5 g GA+ Bb12) \(9 \times 10^4\) followed by 3rd group (Bb12) \(12 \times 10^4\) followed by 2nd group (SM+10g GA) \(17 \times 10^4\) while the highest bacterial colony count was recorded in the 1st group (SM+5g GA) \(23 \times 10^4\) in comparison with the control group. With the time after 5 days and 10 days, bacterial colony count decreased and *Salmonella typhimurium* bacteria completely diminished after 10 days in 4th group (SM +5 g GA+ Bb12) and 2nd group (SM+10 gGA) table 2.

<table>
<thead>
<tr>
<th>Table 2: Results of <em>Salmonella typhimurium</em> colony count from fecal samples of challenged mice in the experimental groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>group</td>
</tr>
<tr>
<td>gp1 (SM+5gGA)</td>
</tr>
<tr>
<td>gp2 (SM+10gGA)</td>
</tr>
<tr>
<td>gp3 (SM+Bb12)</td>
</tr>
<tr>
<td>gp4 (SM+5 g GA+ Bb12)</td>
</tr>
<tr>
<td>gp5 (SM control)</td>
</tr>
</tbody>
</table>

*significance; **high significance

**Daily weight gain of the examined mice:**

In all examined groups there is no significant increase of daily weight gain 2.5 day post administration while there is significant increase after 10 and 20 day particularly in 4th group, 3rd group and 2nd group table 3.

<table>
<thead>
<tr>
<th>Table 3: The daily weight gain of the examined mice groups in the experimental groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>group</td>
</tr>
<tr>
<td>gp1 (SM+5gGA)</td>
</tr>
<tr>
<td>gp2 (SM+10gGA)</td>
</tr>
<tr>
<td>gp3 (SM+Bb12)</td>
</tr>
<tr>
<td>gp4 (SM+5 g GA+ Bb12)</td>
</tr>
<tr>
<td>gp5 (SM control)</td>
</tr>
</tbody>
</table>

mean ± S.E; *significance; **high significance

**Results of estimation of phagocytic activity:**

The results displayed that phagocytic activity day 2 after oral challenge was significant recorded for group 3 and 4 only in comparison with the control group, while at day 10 after challenge was significant only for 2 groups 2nd (77.34±6.13) and 3rd (78.67±3.24) groups while 4th group was high significance (85.67±6.78) in comparison with the control group furthermore, Day 15 after oral challenge significance was for 3rd (76.56±5.23) group and high significance for 4th (79.25±6.21) group table 3.

<table>
<thead>
<tr>
<th>Table 3: Results of detection of phagocytic activity of Gum Arabic in mice.</th>
</tr>
</thead>
<tbody>
<tr>
<td>groups</td>
</tr>
<tr>
<td>gp1 (SM+5gGA)</td>
</tr>
<tr>
<td>gp2 (SM+10gGA)</td>
</tr>
<tr>
<td>gp3 (SM+Bb12)</td>
</tr>
<tr>
<td>gp4 (SM+5 g GA+ Bb12)</td>
</tr>
<tr>
<td>gp5 (SM control)</td>
</tr>
</tbody>
</table>

mean ± S.E; *significance; **high significance

**Lysozyme concentrations:**

The results displayed that lysozymal concentrations in the serum, day 2 after oral challenge was significant recorded for group 3 and 4 only in comparison with the control group, while at day 10 after challenge was significant only for 2 groups 2nd (77.34±6.13) and 3rd (78.67±3.24) groups while 4th group was high significance (85.67±6.78) in comparison with the control group furthermore, Day 15 after oral challenge significance was for 3rd (76.56±5.23) group and high significance for 4th (79.25±6.21) group table 3.
Table 4: Results of lysozymal concentrations in the serum of mice after oral challenge.

<table>
<thead>
<tr>
<th>groups</th>
<th>Day 2 after oral challenge</th>
<th>Day 10 after oral challenge</th>
<th>Day 15 after oral challenge</th>
</tr>
</thead>
<tbody>
<tr>
<td>gp1 (SM+5gGA)</td>
<td>298.45±34.40</td>
<td>276.15±35.12</td>
<td>272.67±56.23</td>
</tr>
<tr>
<td>gp2 (SM+10gGA)</td>
<td>313.43±56.34*</td>
<td>312.34±54.45</td>
<td>309.46±63.56</td>
</tr>
<tr>
<td>gp3 (SM+Bb12)</td>
<td>356.24±34.12***</td>
<td>335.35±68.67*</td>
<td>315.34±27.23*</td>
</tr>
<tr>
<td>gp4 (SM+5 g GA+ Bb12)</td>
<td>345.31±45.34**</td>
<td>365.23±34.98**</td>
<td>333.23±67.45**</td>
</tr>
<tr>
<td>gp5 (SM control)</td>
<td>304.24±45.34</td>
<td>307.33±43.32</td>
<td>287.13±67.24</td>
</tr>
</tbody>
</table>

mean ± S.E; *significance; **high significance

Concentrations of AST, ALT, uric acid, and creatinine:

Present study revealed that AST significantly decreased in 1st group (SM+5gGA) (40.45 µg/l) but 2nd group (SM+10gGA) and 3rd group (SM+Bb12) were higher (42.5 µg/l) and (41.15 µg/l) respectively than 1st and 4th group (SM + 5 g GA + Bb12) (39.33 µg/l) while ALT significantly decreased in 2nd group (SM+10gGA) (29.77µg/l) furthermore, 3rd group (SM+Bb12) and 4th group (SM+ 5 g GA+ Bb12) recorded high significance in reduction of ALT (27.56 µg/l) and (25.23 µg/l) respectively in comparison with the control group (35.34 µg/l). uric acid was not significant decreased in 1st group (SM+5gGA) (3.64 mg/dl) in comparison with the control group (3.94 mg/dl) while 2nd (SM+10gGA) was high significant and 3rd (SM+Bb12) group significantly decreased (2.16 mg/dl) and (2.45 mg/dl) respectively on the other hand 4th group (SM+5 g GA+ Bb12) was not significant (2.86 mg/dl) decreased in comparison with the control group (3.94 mg/dl). concerning, creatinine 1st (SM+5gGA) did not significantly decreased (0.64 mg/dl) in comparison with the control group (0.76 mg/dl) in contrast 3rd (SM+Bb12) and 4th group (SM+5 g GA+ Bb12) were significantly (0.46 mg/dl) and (0.61 mg/dl) decreased while 2nd (SM+10gGA) group was high significant (0.38 mg/dl) decreased in comparison with the control group (0.76 mg/dl) table 5.

Table 5: Results of AST, ALT, uric acid, and creatinine in the serum of mice 10 days post oral challenge

<table>
<thead>
<tr>
<th>groups</th>
<th>AST (µg/l)</th>
<th>ALT (µg/l)</th>
<th>Uric acid (mg/dl)</th>
<th>Creatinine (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>gp1 (SM+5gGA)</td>
<td>40.45±6.45*</td>
<td>31.34±4.34*</td>
<td>3.64±0.50</td>
<td>0.64±0.05</td>
</tr>
<tr>
<td>gp2 (SM+10gGA)</td>
<td>42.50±7.12*</td>
<td>29.77±5.23*</td>
<td>2.16±3.43**</td>
<td>0.38±0.04**</td>
</tr>
<tr>
<td>gp3 (SM+Bb12)</td>
<td>41.15±5.65*</td>
<td>27.56±7.23**</td>
<td>2.45±2.23*</td>
<td>0.46±0.06*</td>
</tr>
<tr>
<td>gp4 (SM+5 g GA+Bb12)</td>
<td>39.33±5.23**</td>
<td>25.23±5.45**</td>
<td>2.86±3.12</td>
<td>0.61±0.09*</td>
</tr>
<tr>
<td>gp5 (SM control)</td>
<td>49.53±6.45</td>
<td>35.34±4.45</td>
<td>3.94±4.11</td>
<td>0.76±0.13</td>
</tr>
</tbody>
</table>

mean ± S.E; *significance; **high significance

Discussion:

The symbiotic relationship between the host and intestinal microbiota has been extensively studied, in part because of its implications in intestinal health. To promote the development of beneficial microbiota in the intestine. Prebiotics and probiotics can restore the balance of bacteria in the digestive tract. Probiotics are beneficial bacteria that can be found in various foods, or in the form of dietary supplements. When you consume probiotics, you add these healthy bacteria to your intestinal tract (Gourbeyre et al., 2001). Common strains include Lactobacillus and Bifidobacterium families of bacteria. Prebiotics are non-digestible foods that make their way through our digestive system and help good bacteria grow and flourish. Prebiotics help feed and keep beneficial bacteria healthy. on its impact towards human health since the history of mankind. This exclusive Natural Prebiotic is exudates from Acacia senegal and Acacia seyal solely obtained from Sudan. Sudanese used it traditionally as food to maintain health and longevity (Roller et al., 2003; Calame, 2008; Kamada et al., 2013).

The finest quality of Gum arabic maintains more than 85% soluble dietary fiber (oligosaccharide) and high combination of minerals (calcium, potassium, magnesium and iron) from the resin (part of plant used) based on size reduction method (invented and patented) with no introduction to high temperature and no chemical extraction (Williams and Phillip, 2000; Calame et al., 2008).

Present study aimed to investigate the effect of gum Arabic as natural prebiotics in comparison with Bifidobacterium animalis subsp. lactis (Bb12) as probiotic when administrated as food additive for experimentally infected albino mice with salmonella typhimurium.

Concerning the Antagonistic activity of Gum Arabic and Bb12 against Salmonella typhimurium in vitro; present study revealed that the antagonistic activity of Gum Arabic (prebiotic) and Bb12 (probiotic) against Salmonella typhimurium in vitro was recorded as the 1st group (5gGA) recorded 0.31 mm inhibition zone while 2nd group recorded 0.42 mm and 3rd group recorded 9.65 mm and 4th group recorded 9.77 mm in...
contrast the control group revealed 0 mm inhibition zone it was shown that gum Arabic in 2 doses recorded very small inhibition zone while probiotic bacteria recorded 9.65mm and in association with gum Arabic recorded 9.77mm inhibition zone this maybe due to that Bb12 bacteria secrete lactic acid inhibit the growth of Salmonella typhimurium the results confirmed by Vandenbergh (1993); Jakee et al. (2010).

Regarding Salmonella typhimurium colony count from fecal samples of mice, it was noticed that in 2nd day after challenge the lowest bacterial colony count was recorded in the 4th group (SM + 5 g GA + Bb12) 9x10³ followed by 3rd group (Bb12) 12x10³ followed by 2nd group (SM+10g GA) 17x10³ while the highest bacterial colony count was recorded in the 1st group (SM+5g GA) 23x10³ in comparison with the control group. With the time after 5 days and 10 days, bacterial colony count decreased and Salmonella typhimurium bacteria completely diminished after 10 days in 4th group (SM +5 g GA + Bb12) and 2nd group (SM+10 g GA) it was clear that with time (after 10 day) the activity of probiotic Bb12 due to the activation of natural prebiotic gum arabic with the large dose 10 g the results confirmed by Silva et al. (2004) who observed that there was improved survival of mice pretreated with Bifidobacterium longum during challenge with salmonella sp. Furthermore Kamada et al. (2013) who reported that the interaction of the microbiota with pathogens and the host might provide new insights into the pathogenesis of disease, as well as novel avenues for preventing and treating intestinal and systemic disorders. Various carbohydrate sources including gum Arabic are known to support the growth and activity of Bb12 inhibiting growth of salmonella typhimurium reducing its colony count in feces.

Regarding daily weight gain of the examined mice, present study revealed that, in all examined groups there is no significant increase of daily weight gain 2.5 day post administration while there is significant increase after 10 and 20 day particularly in 4th group, 3rd group and 2nd group the results indicated that there is great role of combination between probiotic as Bb12 and prebiotic as Gum Arabic in association of time for significance increase of daily weight gain in experimental animals, the results nearly agree with Isk et al. (2004) and Osman et al. (2010).

Regarding the immunomodulatory effect of Probiotics (PRO) modulate systemic immunity in animals and humans. In contrast, the effects of prebiotics (PRE) on systemic and intestinal immunity have not been investigated. Whether the combined application of PRO and PRE [synbiotics (SYN)] has synergistic or additive effects. The phagocytic activity in present study revealed that, day 2 after oral challenge was significant record for group 3 and 4 only in comparison with the control group, while at day 10 after challenge was significant only for 2 groups 2nd and 3rd groups while 4th group was high significance in comparison with the control group furthermore, Day 15 after oral challenge significance was for 3rd group and high significance for 4th group the results nearly agree with. Sheil et al. (2006) and Calame et al. (2008) who reported that gum Arabic improve all physiological functions for human. The colonic microbiota affects mucosal and systemic immunity in the host (Famularo et al., 1997). Intestinal epithelial cells, blood leucocytes, B and T lymphocytes, and accessory cells of the immune system are all implicated (Schiffir et al., 1997). Bacterial products with immunomodulatory properties include endotoxic lipopolysaccharide, peptidoglycans, and lipoteichoic acids (Standiford et al., 1994) Lipoteichoic acids of Gram positive bacteria such as bifidobacteria possess high binding affinity for epithelial cell membranes and can also serve as carriers for other antigens, binding them to target tissues, where they provoke an immune reaction (Op et al., 1984) while lactobacilli which adhere to human intestinal epithelial cells are capable of activating macrophages (Kleeman and Klaenhammet, 1982; Perdigon et al., 1986) There are as yet no experimental data to support the immunostimulatory properties of non digestible oligosaccharides in humans. However, probiotic organisms interact with the immune system at many levels, including cytokine production, mononuclear cell proliferation, macrophage phagocytosis and killing, modulation of autoimmunity, and immunity to bacterial and protozoan pathogens (Famularo et al., 1997; Schiffir et al., 1997; Matsumar et al., 1992).

The lysozyme concentration, present study displayed that lysozymal concentrations in the serum, day 2 after oral challenge was significant recorded for group 3 and 4 only in comparison with the control group, while at day 10 after challenge was significant only for 2 groups 2nd and 3rd groups while 4th group was high significance in comparison with the control group furthermore, Day 15 after oral challenge significance was for 3rd group and high significance for 4th group the results nearly agree with Roller et al. (2003) and Gourbrey et al. (2001) who reported that Functions of immune cells isolated from peripheral blood mononuclear cells (PBMC), spleen, mesenterial lymph nodes and Peyer’s patches were investigated. The SYN supplement increased secretory immunoglobulin A (sIgA) production in the ileum compared with control, and decreased the oxidative burst activity of blood neutrophils compared with rats fed PRO. The PRE supplement enhanced the production of interleukin-10 in PP as well as the production of sIgA in the cecum, compared with controls. The PRO
supplement modestly affected immune functions, whereas systemic immunomodulatory effects were observed in rats fed SYN.

Concerning the concentrations of AST, ALT, uric acid, and creatinine, Present study revealed that AST significantly decreased in 1st group (SM+5gGA) but 2nd group (SM+10gGA) and 3rd group (SM+Bb12) were higher than 1st and 4th group (SM + 5 g GA + Bb12) while ALT significantly decreased in 2nd group (SM+10gGA) furthermore, 3rd group (SM+Bb12) and 4th group (SM+5 g GA+Bb12) recorded high significance in reduction of ALT in comparison with the control group. the results disagree with Antuncovic et al.(2005) who reported that the probiotic pioneer PDFM significantly reduced serum glucose and urea levels and activates AST and ALT in lambs and agree with El-Jakee et al.(2010) who recorded that AST and ALT were reduced in mice supplemented probiotics with skimmed milk.

Uric acid was not significant decreased in 1st group (SM+5gGA) in comparison with the control group while 2nd (SM+10gGA) was high significant and 3rd (SM+Bb12) group significantly decreased on the other hand 4th group (SM+5 g GA+Bb12) was not significant decreased in comparison with the control group. concerning, creatinine 1st (SM+5gGA) did not significantly decreased in comparison with the control group in contrast 3rd (SM+Bb12) and 4th group (SM+5 g GA+Bb12) were significantly decreased while 2nd (SM+10gGA) group was high significant (0.38 mg/ml) decreased. the results nearly agree with Aled et al.(2011) who reported that in vivo and in vitro with gum arabic, which show its compatibility in the diet of patients suffering with diabetes mellitus and reduction in systolic blood pressure, which may translate into improved cardiovascular outcome and a reduction in the progression of renal diseases. Nasir (2007) reported that GA treatment was associated with an increased 24 h-creatinine clearance in healthy mice. The exact mechanism for this remains unclear, since it represents a remote effect of GA on the kidney, which requires one or more humoral factors. It is well known that GA is fermented by intestinal bacteria leading to formation of various degradation products, such as short chain fatty acids (Bliss et al., 1996). In a recent study, serum butyrate concentrations were increased following treatment with GA in healthy subjects (Matsumoto et al., 2006) and this may have a role in the claimed salutary effect on creatinine clearance. Recently, a report from Sudan assessed the effect of GA on the concentration of certain metabolites in the sera of patients with CRF on a low-protein diet. GA was given at an oral dose of 50 g/day for 3 months, with or without supplementing the diet with ferrous sulfate (200 mg/day) and folic acid (5 mg/day). Serum creatinine, urea, phosphate and uric acid concentrations were reported to be significantly reduced by GA, (Ali et al.,2008).

From present study it was concluded that there are symbiosis between prebiotics and probiotics, the results indicated that gum Arabic is good for health as natural prebiotic especially in the larger dose, *Bifidobacterium lactis* Bb12 also good for health and immune status while the combination between gum Arabic as prebiotic and *Bifidobacterium* as probiotic (5 g GA+ Bb12) was better whether the combined application of PRO and PRE (symbiotics) has synergistic or additive effects significantly, they give very good results than each one alone and improve the health conditions, physiological and immune response; cellular and humeral for challenged mice with *salmonella typhimurium*.

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