The Effect of Single or Combined Dietary Supplementation of Mannan Oligosaccharide and Probiotics on Performance and Slaughter Characteristics of Broilers

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Abstract: A feeding trial was conducted to investigate the effects of dietary supplementation of prebiotic (mannan oligosaccharide, MOS), Probiotic (Saccharomyces cerevisiae) and their combination (synbiotic) on growth performance, some carcass traits and meat quality in broilers. One hundred, one day-old broiler chicks (Avian 48), were randomly assigned to 4 dietary treatments (25 birds/treatment) for 6 weeks. Treatment groups were as follow: 1. Basal diet (control); 2. Basal diet plus Mannan-Oligosaccharide (MOS) (2 g of MOS/kg of the starter diets and 0.5 g/kg of the grower diets); 3. Basal diet plus probiotic (Saccharomyces cerevisiae) 3 g/kg diet and 4. Basal diet plus the combination of pre and probiotics (synbiotic). The final BW, weight gain, carcass yield percentage and immune organ weights were significantly (p<0.05) increased in probiotic and synbiotic supplemented broilers in comparison with the control and prebiotics groups. Meat from prebiotics supplemented broilers was more tender and juicy. Serum total cholesterol and triglycerides were significantly decreased in all dietary treatments compared with the control broilers. Supplementation of broiler diets with pre, pro and synbiotics significantly improved body weight, weight gain and feed conversion, some carcass traits and meat quality, compared with the un-supplemented control. The feed conversion rate was significantly lower in synbiotic supplemented group. The results of the present study demonstrated that synbiotic as growth promoters appeared to be superior compared to use pre or probiotic alone in optimizing digestion to convert feed into body mass more efficiently and improve broiler chickens growth performance.

Key words: Mannan oligosaccharides, prebiotic, probiotic, synbiotic, broilers, performance

INTRODUCTION

Increased bacterial resistance to antibiotics in humans has caused an increase in public and governmental interest in eliminating sub-therapeutic use of antibiotics in livestock. An alternative approach to sub-therapeutic antibiotics in livestock is the use of probiotic microorganisms, prebiotic substrates that enrich certain bacterial populations, or symbiotic combinations of prebiotics and probiotics. Prebiotics such as inulin, Fructo-oligosaccharides (FOS), Isomalt-oligosaccharides (IMO) and Mannan Oligosaccharides (MOS) have been defined by Gibson and Roberfroid (1995) as non-digestible food ingredients that selectively stimulate the growth and activity of one or a limited number of bacteria in the intestine that can improve the host health (Gibson and Roberfroid, 1995). These fermentable substrates are not hydrolyzed by the host digestive enzymes and pass intact in the lower digestive tract. Prebiotics have been reported to produce a beneficial effect upon the animal that receives them. This is due to the proliferation of certain beneficial bacteria such as Bifidobacterium sp. and Lactobacillus sp. or an increase in their metabolic activity (Gibson and Roberfroid, 1995). Inulin, FOS and IMO are reported to be substrates for certain species of beneficial bacteria (Chung and Day, 2004). MOS had the ability to block the colonization of the pathogens as E. coli and Salmonella sp., by blocking the ability of these bacteria to connect via their fimbriae, with the intestinal epithelial cells and therefore facilitating their excretion (Spring et al., 2000; Duncan et al., 2005). Also, the MOS have a positive effect on the immunity preventing the infections (Shashidahara and Dewegowda, 2003).

Gong et al. (2002) define probiotics as health-promoting bacteria inhabiting the gastrointestinal tract of humans and animals. Research is focused on identifying beneficial bacterial strains and substrates along with the conditions (Patterson and Burkholder, 2003). The major probiotic strains include Lactobacillus, Saccharomyces, Bacillus, Streptococcus and Aspergillus (Tannock, 2001). Positive effects of probiotics on animals can result either from a direct nutritional effect of the probiotic, or a health effect, with probiotics acting as bioregulators of intestinal microflora and reinforcing the host's natural defenses. There have been numerous studies in humans and animals on the ability of probiotics to change the types

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and numbers of gut microflora (Endo et al., 1999). Exactly how dietary microbial products function in the digestive system is not known, but some suggested mechanisms are that they: 1) provide nutrients, 2) aid in digesting and 3) inhibit harmful bacteria (Owings et al., 1990). Moreover, *Saccharomyces cerevisiae* could act as bioregulator of the intestinal micro flora and reinforcing the host natural defenses, through the sanitary effect by increasing the colonization resistance and stimulation of the immune response (Line et al., 1998). Effects of yeast products on production and their mode of action in monogastrics have been reported in poultry (Stanley et al., 2004, Zhang et al., 2005). However, mode of action of yeast products in these animals is less clear. Some studies have confirmed the effects of Yeast Culture (YC) in increasing concentrations of commensal microbes or suppressing pathogenic bacteria (Stanley et al., 2004). Gao et al. (2008) hypothesized that there may be other mechanisms responsible for effects of YC in monogastrics other than modulation of microbial ecology. Mannan oligosaccharides and 1,3/1,6 β-glucan are components of the yeast cell wall that modulate immunity (Shashidhara and Dewewgoda, 2003), promote growth of intestinal microflora (Spring et al., 2000) and increase growth (Parks et al., 2001). YC contains viable cells, cell wall components, metabolites and the media on which the yeast cells were grown (Miles and Bootwalla, 1991).

The combination of a pre- and probiotic in 1 product has been shown to confer benefits beyond those of either on its own (Gallaher and Khil, 1999). Symbiotics is defined as a mixture of probiotics and prebiotics that beneficially affects the host by activating the metabolism of one or a limited number of health promoting bacteria and/or by selectively stimulating their growth improving the host’s welfare (Gibson and Roberfroid, 1995). Zhang et al. (2006) found that some probiotics or symbiotics were effective in increasing the body weight of chickens. In addition, Mohni et al. (2007) found that the symbiotic product (Biomix® PoultryStar) had a comparable potential to improve broiler performance as Avilamycin (an antibiotic growth promoter). It seems that synergistic effects of prebiotics and probiotics can be useful in stimulating beneficial bacteria and improving the health of the gut. However, there is scarce information available to date on symbiotics and its possible mechanisms in broiler chickens. Symbiotics include both beneficial microorganisms and substrates, which may have synergetic effects on the intestinal tract of animals. Thus it may offer a considerable promise for growth promotion in poultry production and a new alternative to antibiotic growth promoters (Patterson and Burkholder, 2003; Awad et al., 2008, 2009).

Therefore, the objective of this study was to evaluate effects of dietary supplementation of prebiotics (Bio-MOS), probiotic (*Saccharomyces cerevisiae*) and their combination on growth performance, carcass traits and meat quality in broilers.

**MATERIALS AND METHODS**

**Birds and housing:** The experiment was conducted in the experimental poultry house of the Department of Nutrition and Clinical Nutrition, Fac. of Vet. Med., Assiut University, Assiut, Egypt. A total number of 100 One-day chicks (Avian 48) were randomly divided into 4 groups (25 birds/group) and housed in pens of identical size (2 m²) in a deep litter system with a wood shaving floor and equipped with feeders and drinkers. Each group had 3 replicates (8 birds/pen) one pen contain 9 birds in each group. The birds had free access to water and feed. The climatic conditions and lighting program followed the commercial recommendations. Environmental temperature in the first week of life was 32°C and decreased to 25°C until the end of the experiment. During the first week, 22 h of light was provided with a reduction to 20 h afterward.

**Dietary treatments:** Two basal diets, starter and grower-finisher were formulated. The starter diet was fed for the first 3 weeks, while the grower finisher one was fed for the last 3 weeks of the experimental period. The physical and the calculated chemical composition of the basal diets are illustrated in Table 1. The dietary treatments were: 1 Basal diet (control); 2. Basal diet + 0.2% prebiotic (2 g prebiotic/kg) in the starter phase and 0.05% (0.5 g prebiotic/kg) in the grower phase; 3. Basal diet plus 0.3% (3 g prebiotic/kg) 4. Basal Diet + 0.2% prebiotics and 0.3% probiotics in the starter phase or 0.05% Bio-mos and 0.3 Probiotic in the grower phase. The chicks were fed with the starter diets from d 1 to d 21 of age and grower feed from d 22 to d 42 (Table 1) in mash form. Diets were formulated according to NRC (1994) recommendations. A commercial prebiotic source Bio-Mos® (Alltech Inc., Nicholasville, KY, USA) and commercial probiotics source (Yeast culture probiotics (Bro-bio-fair, Vitality Co., Egypt), which contained *Saccharomyces Servisia* (labelled as each kg contains: 100 gram *Saccharomyces servisia* (10¹⁰ cell/gram) and 900 gram carrier (90% soy meal, 5% malt, 5% Fenu Greek). The birds were fasted for 10-12 h prior to determination of the final body weight at slaughter.

**Growth performance traits:** All birds were weighed individually after their arrival from the hatchery to the experimental farm (initial weight) and every week tell the end of the experiment the BW was recorded. Weight gain for each dietary treatment was calculated. Feed consumption was recorded in the course of the whole experiment for each treatment and the feed conversion
Table 1: Inclusion rates for mannur oligosaccharide (Bio-MOS) and probiotic products in broiler chicken diets by treatment

<table>
<thead>
<tr>
<th>Item</th>
<th>Additives</th>
<th>Starter (d 0 to 21)</th>
<th>Grower and finisher (d 21 to 42)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1 (control)</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>T2</td>
<td>Prebiotic (Bio-Mos)</td>
<td>2</td>
<td>0.5</td>
</tr>
<tr>
<td>T3</td>
<td>Probiotic</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>T4</td>
<td>Bio-Mos + probiotic (synbiotic)</td>
<td>2 + 3</td>
<td>0.5 + 3</td>
</tr>
</tbody>
</table>

Table 2: Composition and nutrient content of starter (d 1 to 21) and grower (d 22 to 42) basal diets for broiler chicks (% as fed-basis)

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Starter</th>
<th>Grower</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yellow corn</td>
<td>55.45</td>
<td>64.00</td>
</tr>
<tr>
<td>Soybean meal</td>
<td>32.70</td>
<td>25.00</td>
</tr>
<tr>
<td>Fish meal (60%)</td>
<td>4.00</td>
<td>4.00</td>
</tr>
<tr>
<td>Sunflower oil</td>
<td>4.50</td>
<td>3.70</td>
</tr>
<tr>
<td>Dicalcium phosphate</td>
<td>1.50</td>
<td>1.50</td>
</tr>
<tr>
<td>Limestone</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>Salt (NaCl)</td>
<td>0.40</td>
<td>0.36</td>
</tr>
<tr>
<td>Lysine</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>DL-Methionine</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Premix¹</td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td>Antioxidant</td>
<td>0.04</td>
<td>0.04</td>
</tr>
<tr>
<td>Total</td>
<td>100.00</td>
<td>100.00</td>
</tr>
</tbody>
</table>

Calculated analysis

| ME (kcal/kg) | 3202.91 | 3224.18 |
| CP (%)       | 23.03   | 20.03   |
| Met and Cys (%) | 0.93 | 0.87 |
| Lysine (%)   | 1.39    | 1.19    |
| Ca (%)       | 1.01    | 1.00    |
| Av. P (%)    | 0.49    | 0.49    |
| Na (%)       | 0.19    | 0.18    |
| CF (%)       | 2.97    | 2.92    |

¹Mineral and vitamin premix Heromix broilers (Heropharma Co., Egypt).

Each 2.5 kg contain: 12,00000 IU Vit. A, 2,000000 Vit D3, 10 g vit. E, 2 g Vit K3, 1 g Vit. B1, 5 g Vit B2, 1.5 g Vit B6, 10 mg Vit B12, 30 g niacin acid, 10 g panthanic acid, 1 g folic acid, 50 g biotin, 250 g choline chloride 50%, 30 g iron, 10 g copper, 50 g zinc, 60 g manganese, 1 g iodine, 0.1 g selenium, 0.1 g cobalt and carrier CaCO₃ to 2.5 kg

Rates were calculated subsequently. Mortality was recorded as it occurred and feed per gain values were corrected for mortality (regarding feed intake).

Organ weights and carcass yield percentages: At the end of the experiment, after weighing, 4 birds per treatment were randomly selected and weighed live, slaughtered by neck cut and allowed to bleed. Afterward, the birds were scalced, de-feathered and carcasses were eviscerated. The gizzard, heart, liver, were excised and weighed. The weight of carcass refers to the weight of the eviscerated carcass plus giblet (liver, heart, skinned empty gizzard). Carcass yield (dressing percentage) was obtained by expressing the dressed carcass weight (with giblet) as percentage of live body weight.

Sensory analysis: Three consumer-based sensory panel were conducted to evaluate the acceptability of live body weight.

Serum biochemistry: Four broilers were randomly selected from each treatment group for blood sample collections from the brachial vein during at day 21 and from the bronchial vein during slaughter at day 42. The collected blood samples were centrifuged at 2000 rpm for 10 min and the sera were collected, stored at -20°C until further analysis. Serum samples were assayed for estimation of total protein, triglycerides, cholesterol, calcium, phosphorus, ALT and AST enzymes by spectrophotometer using commercial test kits (Spectrum, Cairo, Egypt).

Statistics: The data were subjected to statistical analysis with one way ANOVA of (SPSS for Windows Version 13; SPSS GmbH, Munich, Germany) to determine if variables differed between groups. The Kolmogorov-Smirnov test was used to test the normal distribution of the data before statistical analysis was performed. The BW, weight gain, feed intake, feed conversion and organ weights, were compared between groups by one way ANOVA and subsequent Duncan’s multiple range test. Probability values of less than 0.05 (p<0.05) were considered significant. Results are expressed as means ± pooled SEM.

RESULTS

The initial BW of chicks did not differ (p>0.05) between the dietary treatments (Table 3). Birds supplemented with prebiotic (BIO-MOS), probiotic and those supplemented with symbiotic had a greater (p<0.05) BW compared with controls at d 21 of age. Moreover, MOS supplemented birds had a greater BW (706.00) (p<0.01) than birds supplemented with the symbiotic (632.32) at d 21. However, at the end of the experiment (d 42), birds
supplemented with symbiotic exhibited a greatest (p<0.05) BW (2399.19) compared to probiotic group (2247.14) and control group (2099.72), however the prebiotic supplemented group had an intermediate (2127.38) values. The cumulative feed intake from d 1 to 21, from d 22 to d 42 and during the whole experiment was increased (p<0.01) for birds supplemented with MOS compared with the other dietary treatments and the control groups.

**Mortality rate:** The mortality rate was numerically lower (Table 3) for all the dietary treated groups in comparison with the control group at day 21 and at the last 3 weeks from the experiment (d 22 to 42 day). Moreover, at the end of the experiment the mortality rate was lower numerically not statistically in prebiotic and symbiotic supplemented groups.

**Carcass traits and absolute organ weights:** The data in Table 4 showed that there is a significant increase (p<0.05) in the carcass weight and dressing % in symbiotic supplemented broilers compared with either pre or probiotic alone supplemented group. There is also a significant increase in the absolute weight of the immune organs (bursa and thymus) and numerical increase in the spleen in symbiotic supplemented broilers. However, the liver also tended to be higher (p<0.1) in symbiotic supplemented broilers.
Sensory characteristics: Sensory analysis data (Table 5) declared the presence of significant difference (p<0.05) between control samples and those from chicken fed Bio-MOS concerning tenderness and juiciness. Where samples from chicken fed Bio-mos were rated higher. Like wise, there were a significant difference in the tenderness and juiciness between samples from chicken fed Bio-Mos and those from the other two treatments, where Bio-Mos fed chicken had more tender and juicy meat. However, there were no significant difference between the control sample and those from chicken fed probiotic or symbiotic concerning tenderness and juiciness. As well, no significant difference was found between control samples and all the other treatments or between the three different treatments concerning the flavor of the chicken meat samples.

Blood metabolites: The data at Table 6 showed that at day 21 serum total protein increased (p<0.01) in MOS supplemented group in comparison with the control and probiotic supplemented groups. Moreover, serum calcium and phosphorus level was numerically higher in all dietary supplemented groups in comparison with the control one.

At day 42 there was a significant decrease in serum total cholesterol and triglycerides in all dietary supplemented groups in comparison with the control one. There is a numerical decrease in serum AST and ALT in all dietary supplemented groups in comparison with the control one.

DISCUSSION
The alternatives to in feed antibiotics possibly can improve the health and performance of birds by increasing the growth of beneficial microbes or by reduction and removal of potential pathogens. Probiotics, prebiotics and sybiontics are either beneficial microorganisms or substances that facilitate the growth of these microorganisms, which can be suitably used by the food manufacturers and hold considerable promise for the health care industry.

At day 21 BW, weight gain and feed conversion efficiency was significantly improved in all dietary treatments in comparison with the control group. Prebiotic (MOS) supplemented broilers are superior to probiotic and symbiotic birds at day 21 in BW and weight gain and feed intake. These results were in agreement with several studies, which have reported that probiotics can improve performance in broilers (Olsen, 1995; Parks et al., 2001; Ferket et al., 2002; Ferket, 2004; Hooge, 2004; Benites et al., 2008). Three major modes of action by which broiler performance is improved by MOS are proposed: 1) control of pathogenic or potential pathogenic bacteria which possess type-1 fimbriae (mannosensitivite lectin), 2) immune modulation and 3) modulation of intestinal morphology and expression of mucin and brush border enzymes (Ferket, 2004). A unique character of MOS in immune modulation is that it enhances the protective antibody response to enhance disease resistance while at the same time suppress the acute phase (fever) response (Ferket et al., 2002). Mannanoligosaccharides, specifically Bio-MOS, were shown to alter mucosal architecture and longer villi were noticed in birds fed MOS-supplemented diets (Loddi et al., 2002; Yang et al., 2007). Further, Bio-MOS consistently reduced the crypt depth of the mucosa of the small intestine where its growth-promoting effects were observed.

At day 42 body weights, the cumulative weight gains and feed conversion rate were significantly (p<0.05) increased by the dietary inclusion of the probiotics and symbiotic as compared to the control fed broilers. Furthermore, the mortality % was numerically decreased by dietary supplementation of prebiotic and symbiotic. These results are fully consistent with the results of Kim
et al., 1988; Awad et al., 2008; Bozkurt et al., 2008; El-Banna et al., 2010; Falaki et al., 2011. Improvement in growth performance and feed efficiency in broilers chickens fed probiotic and symbiotic may be attributed to the total effect of probiotic action including the maintenance of beneficial microbial population (Fuller, 1989), improving feed intake and digestion (Nahanshon et al., 1992) and altering bacterial metabolism. The results of our study are supported by other studies (Kim et al., 1988; Jin et al., 1998; Jamroz and Kamel, 2002; Mitsch et al., 2004; Li et al., 2007; Mountzouris et al., 2007; Awad et al., 2008, 2009). These studies suggested that probiotics, prebiotics, and symbiotics could enhance the performance of broilers by (1) improving intestinal morphology and microbial balance associated with suppressing intestinal pathogens, by competitive exclusion and antagonism, such as Salmonella, Campylobacter and E. coli (Fuller, 1989; Jin et al., 1998; Kabir et al., 2005; Line et al., 1998), (2) increasing digestive enzyme activity and decreasing bacterial enzyme activity and ammonia production (Chiang and Hsieh, 1995; Jin et al., 2000), (3) also by an increase in feed intake and the uptake of nutrients (fatty acids and glucose), (4) fixation of nitrogen and reduction of excretion of fat in feces and microbial urea (Willis et al., 2007). (5) Stimulating the immune system and reducing mortality (Huang et al., 2004, Kabir, 2009). Therefore, beneficial effects of these supplements in the gastrointestinal tract could result in an improvement of overall health and performance of animals. However, Karaoğlu and Durdag (2005) used Saccharomyces cerevisiae as a dietary probiotic to assess performance and found no overall weight gain difference. Variations among reports of researches could be related to environmental conditions that exist in various experiments.

In poultry industry, one of the major concerns is to obtain a higher percentage yield of saleable products and consequently, to increase the edible portions. The significant increase in the carcass weight and dressing % in symbiotic supplemented broilers was totally coincided with the observations of (Awad et al., 2009) who reported that carcass yield percentage was significantly increased in the symbiotic-fed broilers compared with the control and probiotic-fed broilers. In consistent with that prediction, there are significant increases in the absolute weight of breast, proventriculus and pancreas in symbiotic supplemented group. Kabir et al. (2004) reported the occurrence of a significantly (p<0.01) higher carcass yield in broiler chicks fed with the probiotics on the 2nd, 4th and 8th week of age both in vaccinated and non-vaccinated birds. Although Mahajan et al. (1999) recorded in their study that mean values of givlets, hot dress weight, cold dress weight and dressing percentage were significantly (p<0.05) higher for probiotic (Lacto-Sacc) fed broilers. Dietary treatments had no significant effect on abdominal fat pad accumulation in the present study. These results are in harmony with results of (Waldroup et al., 2003; Aliciçek et al., 2004; Bozkurt et al., 2005). The significant increases in the absolute weight of the immune organs (thymus and bursa) were in harmony with the results of previous studies (Gao et al., 2001; Wang et al., 2003; Huang et al., 2007). Measurement of immune organ weight is a common method for evaluation of immune status in chickens (Heckert et al., 2002). Such related organs include bursa of Fabricius, liver and spleen. Good development of these organs is crucial for optimal Ig synthesis (Glick, 1977).

The improvement of sensory characteristics of meat samples from broilers supplemented with prebiotics was observed. Kabir et al. (2005) evaluated the effects of probiotics on the sensory characteristics and microbiological quality of dressed broiler meat and reported that supplementation of probiotics in broiler ration improved the meat quality both at pre-freezing and post-freezing storage. Likewise, Zhang et al. (2005) conducted an experiment with 240, day-old, male broilers to investigate the effects of Saccharomyces cerevisiae (SC) cell components on the meat quality and they reported that meat tenderness could be improved by the whole yeast or Saccharomyces cerevisiae extract. On the other hand, Loddi et al. (2000) reported that neither probiotic nor antibiotic affected sensory characteristics (intensity of aroma, strange aroma, flavour, strange flavour, tenderness, juiciness, acceptability, characteristic colour and overall aspects) of breast and leg meats.

The effects of probiotics and prebiotics on serum cholesterol and triglycerides concentrations are consistent among previous studies. These studies have shown that probiotics and prebiotics exhibited lipid-lowering properties which might be related to the changes in the intestinal bacterial flora composition, which ferments prebiotics to produce short-chain fatty acids in the gut and then causes a decrease in the systemic levels of blood lipids and cholesterol. Furthermore, some probiotic bacteria may interfere with cholesterol absorption in the gut by de-conjugating bile salts or by directly assimilating cholesterol (Mohan et al., 1996; Jin et al., 1998; Zhao and Yang, 2005; Lian and Shah, 2006; Li et al., 2007).

In conclusion, The BW, weight gain, carcass yield percentage and feed conversion rate were significantly (p<0.05) increased by the dietary inclusion of the symbiotic and probiotic compared with the control. The results of the present study revealed that supplementation of broiler diets with pre, pro and symbiotics increased body weight gain and feed conversion, some carcass traits and meat quality.
compared with the un-supplemented control. The combination of pre and probiotic (synbiotic) induced additive benefit in growth performance, feed conversion and carcass traits than that of individual use of these additives. Under present experimental condition, synbiotic as a growth promoter appeared to be superior compared to use pre or probiotic alone in optimizing digestion to convert feed into body mass more efficiently and improve broiler chickens growth performance.

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