



## Effects of feeding Isomaltooligosaccharides on the growth performance, carcass traits and immune response of broiler chickens

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### Abstract

The research was conducted to study the effect of feeding broiler chickens on diets containing isomaltooligosaccharides on the growth performance, carcass traits and immune response. 90-one day old broiler chicks were used according to completely randomized two treatment groups and one control, 30 birds each. Birds fed *ad-libitum* on basal starter and grower-finisher diets for 35 day. Diets of treatment's groups contained 0.5 g/Kg and 1 g/Kg of Isomaltooligosaccharides, while the control group fed on the basal diets without Isomaltooligosaccharides supplementation. Dietary supplementation of broiler chickens with Isomaltooligosaccharides improved body weight, feed conversion, carcass traits, two lymphoid organs weight and log antibody titer against avian flu vaccine. Most of the highest values were for birds fed low levels of Isomaltooligosaccharides. Feed intake decreases as Isomaltooligosaccharides level increases. Dietary supplementation with Isomaltooligosaccharides did not affect the lipids profile (triglycerides, total cholesterol, LDL and HDL), however the blood VLDL levels decreased with increased levels of Malondialdehyde and Glutathione reductase. Collectively, Dietary supplementation of broiler chickens with 0.5 g/Kg diet of Isomaltooligosaccharides improved growth performance, carcass traits and immune status.

**Key words:** Isomaltooligosaccharides; Broilers; growth performance; Carcass; and Immunity.

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## Introduction

Oligosaccharides are composed of between two and nine monosaccharides linked through glycosidic bonds. Nondigestible oligosaccharides are oligosaccharides that are not hydrolyzed by digestive enzymes in the gastrointestinal tract. Isomaltooligosaccharides (IMO) are produced from glucose by enzymatic transgalactosylation [Hayashi *et al.*, 1994; Vetere *et al.*, 2000]. The mixture contains isomaltose (O- $\alpha$ -D-glucopyranosyl-(1-6)-D-glucopyranose), panose, isomaltotriose, and several other branched oligosaccharides composed of four or five glucose residues. More importantly, IMO have been used as a prebiotic similar to other nondigestible oligosaccharides [Crittenden, 1999; Zhang, 2000; Chen *et al.*, 2001]. IMO was tested as prebiotic for poultry and stimulated the growth of *Bifidobacterium* and *Lactobacillus* and are not used by *Salmonella* or *Escherichia coli* [Chung and Day, 2004]. This wasn't the aim of this paper. However, this role has been a matter of debate because prebiotics are a non-digestible feed ingredient that may benefit the host by selectively stimulating the growth or activity of one or a limited number of bacteria in the colon, resulting in improved host health and performance [Gibson and Roberfroid, 1995]. Although, IMO can be cleaved at the brush border of small intestinal mucosal cells [Hunziker *et al.*, 1986; Najjar *et al.*, 1991; Gu" nther and Heymann, 1998; Hertel *et al.*, 2000], It enhance the growth performance of broilers [Chung and Day, 2004], potentially activate the immune system, thereby enhancing the resistance to diseases [Li *et al.*, 2009]. Because of a very few investigations of IMO have been undertaken on broilers to date, the aim of this work was to study the influence of dietary supplementation of broilers with IMO on the growth performance, carcass traits and immune response.

## Materials and Methods:

The experiment was carried out under the protocol approved by the Faculty of Veterinary Medicine, Department of Nutrition and Clinical Nutrition, Sadat City University Egypt. A total of 90 one day old chicken "cobb<sub>500</sub>" were randomly allotted to 3 floor pens 30 birds each. The pens were assigned to control and two treatment's groups. The average body weights of the birds in groups were similar. Birds were raised to 35 days and fed corn-soya bean based diet (1-15 day of age) and grower-finisher diet (15-35 day of age) which formulated to guarantee the strain's requirements or exceed recommended nutrient requirement [NRC, 1994]. One basal diet was mixed for each dietary period and the additives were blended in according to treatment distribution. The basal diets composition for all experimental phases is listed in Table 1. Basal diets (starter and grower-Finisher) were used without IMO supplementation for the control group. Supplemental IMO 93% was substituted for corn in the basal diets (starter and grower-Finisher) at inclusion rate of 0.5 g/kg and 1 g/kg for the two treatment's groups (T1 and T2), respectively. The IMO product contained the following sugars: glucose (0.27%), maltose (7.4%), isomaltose (24.5%), panose (7.39%), and isomaltotriose (16.07%), and the remainder consisted of maltotriose and other oligosaccharides. Diets were antibiotic free and were provided in mash form.

## Management and housing

Top-dressed litter with 2 inch of fresh pine wood shavings was used as bedding. The temperature was set at 30 to 33°C during the first week and was reduced by 2 °C per week until 23 °C was reached. Relative humidity was about 60 to 80%. The lighting program was 23L:1D. The chicks were vaccinated against Avial Flu disease. Access to feed and water was provided on an *ad libitum* basis.

## Birds' performance measurements

Body weight and feed intake were monitored on a pen basis weekly and calculated individually, while weight gain, relative growth rate [Brody, 1945] and feed conversion ratio [Singh and Panda, 1992] values were calculated at the end of starting (0-14day of age ) and growing-finishing (14-35 day of age) periods. Mortality was also recorded on a daily basis in each pen. Chickens were killed by cervical dislocation at the end of the trial. Six birds per treatment group were randomly selected for determining carcass traits. They were de-feathered, eviscerated and dressed. Liver, gizzard, proventriculus, heart, thigh, breast, spleen and bursa of fabricius were collected, weighed and calculated as percentage of live weight.

## Sampling and methods of analysis

Representative samples of mixed rations were taken for chemical analysis [A.O.A.C 2000]. At the end of 5<sup>th</sup> week of age, blood samples were obtained from wing veins and directly liquated into 2-mL sterile vials, and allowed for one hour at room temperature (23C<sup>o</sup>) for complete coagulation and for 3h in refrigerator (4C<sup>o</sup>) for complete separation of serum before centrifugation at 1500 rpm, 6C<sup>o</sup> for 20 min. The serum samples were used for determination of triglycerides [Fassati and Principe 1982], total cholesterol [Barnes *et al.*, 1972], high density lipoprotein HDL [Najjar *et al.*, 1991], low density lipoprotein LDL [Lee and Nieman, 1996], very low density lipoprotein VLDL [Lee and Nieman, 1996] and malondialdehyde [Wong *et al* 1987], glutathione reductase [Anderson, 1996] using Spectrophotometer and commercially available kits (Biosystem S.A, Costa Brava, 30, Barcelona, Spain) according to manufacturer's instructions.

## Results and Discussion

Many investigations proved the role of IMO as prebiotic for human [Chen *et al.*, 2001, Fu *et al.*, 1999], animals [Crittenden, 1999, Kaneko *et al.*, 1995], and poultry [Zhang, 2000 and Zhang *et al.* 2003]. However, very few investigations were conducted on the role of IMO in the poultry as growth promoter or immune stimulant. In the present study, birds fed on diets supplemented with IMO had numerically the heavier body weight than the birds of control group. The heaviest significant body weights were for the birds fed on diet supplemented with 0.5g/Kg diet IMO compared with the control group but no difference when compared with that fed on diets contained 1g IMO/Kg of diet (Table 2). This effect was observed all over the experimental periods as reflex for improved relative growth rate for the treatment's groups. This



effect agreed with the finding of Zhang *et al.* [2002] who showed enhanced body weights for birds fed on diets contained 0.1%, 0.2%, and 0.4% of IMO compared with control group and with those of Saminathan *et al.* [2014] who found an improved growth performance of broiler chickens fed on diets contained IMO. On the other hand, these results weren't agreed with the work of Dagcutan Broce [2013] who found no effect for feeding ration containing IMO on the growth performance of broilers. The overall Feed conversion ratio of the treatment's groups were improved compared to the control group. Birds fed on diets supplemented with 0.5 g IMO/Kg diet have had the best significant feed conversion values. This improvement was observed at the ends of starting and finishing periods. Moreover, the feed intakes values were numerically decreased as the IMO increased (Table 2). This effect was agreed with the finding of Thitaram *et al.* [2005] but not agreed with that of Yang *et al.* [2008] and Zhang *et al.* [2003] who found no effects for dietary supplementation of IMO on the feed conversion of broilers.

This effect was demonstrated as a result of more aspects, the first was due to the stimulated glycolytic bacteria in the crop, because the level of amylase in chicken saliva is very low [Jerrett and Goodge, 1973], and only a trace of maltose or glucose is produced in the crop [Pritchard, 1972]. Secondly, production of more sugars from the cleavage of IMO at brush border of the intestinal mucosal cells [Hunziker *et al.*, 1986; Najjar *et al.*, 1991; Günther and Heymann, 1998; Hertel *et al.*, 2000]. Both aspects might enhanced the feed utilization of the treatment's groups. On the other hand, *Lactobacillus spp.* is the most dominant bacterial species in the crop [Fuller, 1977; Mead, 1997; He *et al.*, 2000]. It can inhibit growth of pathogens [Fu *et al.*, 1999] by maintaining normal microbial balance in the crop [Fuller, 1973], which is important for broilers because the crop is the gateway for exotic bacteria to enter the intestines of birds. Also, *Lactobacillus spp.* is the only organism normally present in the duodenum and small intestine at levels significantly above 10<sup>3</sup> per gram in chickens [Barnes *et al.*, 1972]. Most of the organic acids produced from fermentation of *Lactobacillus spp.* (such as *L. fermentum*, *L. acidophilus*, and *L. plantarum*) are lactic and acetic acids [Mitsuhashi *et al.*, 1983] in addition to other organic acids, which initiate the prebiotic effect of IMO. Collectively, these suspected modes of action were included as causes for an improved growth performance. This increased growth performance proved the positive impacts for dietary IMO supplementation for broiler. These effects were observed in the trial of Zhang *et al.* [2003] during the first three weeks of age but weren't found after the 3<sup>rd</sup> week of age due to the occurrence of infectious outbreak. Our results were in agreement with the finding of Thitaram *et al.* [2005]. However, Sutawee [1997] found no effects for dietary IMO supplementation of broilers diets on body weight and gain, which was attributed it to low dose of IMO supplementation.

The effects of dietary treatments on carcass characteristics and some internal organs of 35 days old broilers are shown in Table 3. Dressing percentages values increased with dietary supplementation of broilers diets with IMO compared to the control group. The difference was significant in the group fed high level of IMO (T2). Percentages of relative weight of breast, liver, gizzard, and heart increased significantly in treatment's groups compared with the control group, where the largest value was for the group fed high level of IMO (T2). The difference was not significant for heart of group fed low level of IMO (T1). These improvement's effects of IMO on carcass yield and parts might be due to its stimulant effects on growth performance. There are very few data describing the precise effects of IMO on carcass traits of broilers, although our results were in agreement with those of Ghiyasi *et al.* [2007] who found improved carcass and breast yields of broilers fed on low protein diets contained IMO as fermentation product of *Aspergillus Orizae*.

The mortality of each treatment was very low, and at most only one bird per control group died. Xia *et al.* [2001] reported that dietary IMO improved survivability of chicks. The indices of two immune organs are shown in Table 3. Differences in bursa and spleen weights due to IMO supplementation were significant compared to the control group except for bursa in groups fed low level of IMO (T1), which is numerically increased. The largest values were for the groups fed on diet contained high level of IMO (T2). There is increasing evidence that IMOs have immunomodulatory effects on systemic immune response including gut-associated lymphoid tissues system [Sung *et al.*, 2004; Buchholz and Seibel, 2003]. Data in table (4) revealed a significant increased serum Log antibodies titer against avian flu by supplementation of broiler chicks with IMO at 21 day, 28 day, and 35 day of age compared with the control group. Difference between treatment's groups and control one were significant except in the group fed low level of IMO only at 28 day of age. The increased weights of immune organs and serum antibody titer against avian flu along the experimental periods of broilers with IMO supplementation proved a stimulant effect of IMO on the immune system. Our results were agreed with those of Zhang *et al.* [2003] who found increased thymus index and weights of immune organs with dietary supplementation of broilers with IMO. Studies in mice gave strong evidence that IMO might stimulate the immune system [Watanabe *et al.*, 2002].

As shown in table 5, although the serum levels of total protein and albumen were not affected by dietary supplementation with IMO, the serum globulin levels increased in all groups fed on diets supplemented with IMO compared with the control group. The difference was significant for group received low levels and insignificant for that received high levels of IMO. The increased levels of blood globulins confirmed the stimulant effects for dietary supplementation of broilers with IMO on the immunoglobulin's content of blood. Mizubuchi *et al.* [2005] observed greater levels of IgA in feces in mice fed for 4 weeks on a diet supplemented with 20% IMO. Unfortunately, our results data did not included the profile of white blood cells, which needs further investigation to assesses the finding of Li *et al.* [2000] who stated an enhanced lymphocytic transformation rate, macrophage activity, and erythrocyte rosette forming cell by inclusion of IMO in the diets of piglets.

Some of serum lipids profile (total triglycerides, cholesterol, LDL, HDL) did not significantly affected by dietary supplementation of broilers with IMO, however the serum levels of VLDL decreased and increased serum Malondialdehyde and Glutathione reductase. The decreased serum VLDL. This dietary supplementation with IMO increased serum Malondialdehyde and Glutathione reductase, which might be proved activation of lipid metabolism. This effect was not observed in the other lipids profile. Although, the increased serum Malondialdehyde and Glutathione reductase are considered as indicators for increased peroxidation, our results for lipids profile were not totally in



agreement with the findings of Wang *et al.* [2001] and Mizubuchi *et al.* [2005] who found an improving lipid metabolism as well as the functions of the liver and the kidneys.

## Conclusion

It was concluded that dietary supplementation of broiler chickens with 0.5 g/Kg of Isomaltooligosaccharides improved the growth performance, carcass traits and immune status but increasing dietary level of IMO to be 1 g/Kg decreased the total feed intake.

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**Table 1. Ingredients and nutrients composition (% DM) of broiler basal diets**

<b>Ingredients</b>	<b>Starter</b>	<b>Grower-Finisher</b>
Yellow corn %	56.55	58.6
Soybean meal (44%)	32.0	30.6
Corn gluten (60%)	6.0	3.6
Vegetable oil	1.0	3.0
Common salt	0.3	0.3
IMO <sup>2</sup>	0.05	0.1
Dicalcium phosphate <sup>1</sup>	1.7	1.7
Vit. <sup>3</sup> & min. mixture <sup>4</sup>	0.3	0.3
Limestone	1.6	1.44
DL-Methionine <sup>5</sup>	0.13	0.12
L- Lysine-HCL <sup>6</sup>	0.17	0.14
Sodium bicarbonate	0.20	0.10
<b>Chemical composition</b>		
Crude protein	21.80	19.90
ME Kcal/Kg	2900	3015
C/P ratio	133	151
Calcium	1.10	1.0
Available Phosphorous	0.45	0.42
Methionine	0.47	0.45
Lysine	1.2	1.0

<sup>1</sup> IMO: Yikangsu is a patent fermentation product contained 93% isomaltooligosaccharides, Guangdong VTR Bio-Tech Co., Ltd, China, Delta Vet. Center, sole agent, Egypt).

<sup>2</sup> Dicalcium phosphate, 18% granular phosphate and 23 % calcium.

<sup>3</sup> Each 1.5 kg contain: vitamin A 12000000 IU, vitamin D3 3000000 IU, vitamin E 40000 mg, vitamin K3 3000 mg, vitamin B1 2000 mg, vitamin B2 6000 mg, vitamin B6 5000 mg, vitamin B12 20 mg, niacin 45000 mg, biotin 75 mg, folic acid 2000 mg, pantothenic acid 12000 mg.

<sup>4</sup> Each 1.5 Kg contain: manganese 100000 mg, zinc 600000 mg, iron 30000 mg, copper 10000 mg, iodine /1000 mg, selenium 200 mg and cobalt 100 mg.

<sup>5</sup> DL-Methionine, Met AMINO® (DL-2-amino-4-(methyl-thio)-butane acid, DL-methionine, α-amino-γ-methyl-oily acid) by Feed Grade 99% (EU).

<sup>6</sup> L-Lysine HCL 99% (Feed Grade) L-Lysine: 78.0% Min (Indonesia).



**Table 2. Effect of IMO supplementation on the growth performance of broilers**

	Control	T1	T2
<b><u>At the end Of 14 day of age:</u></b>			
Body weight (g)	618.57 ± 23.11 <sup>b</sup>	712.63 ± 18.18 <sup>a</sup>	673.68 ± 12.5 <sup>a</sup>
Body weight gain (g)	573.57 ± 23.11 <sup>b</sup>	667.63 ± 18.18 <sup>a</sup>	628.66 ± 12.5 <sup>a</sup>
Relative growth rate (%) <sup>*</sup>	172.45 ± 0.01 <sup>b</sup>	176.0 ± 0.01 <sup>a</sup>	174.8 ± 0.01 <sup>a</sup>
Feed conversion ratio	1.99 ± 0.08 <sup>a</sup>	1.79 ± 0.05 <sup>b</sup>	1.8 ± 0.03 <sup>b</sup>
Feed intake (g/bird)	1121.9	1176.8	1126.8
<b><u>From 14 to 35 day of age:</u></b>			
Body weight gain (g)	911.07 ± 42.23 <sup>b</sup>	1052.6 ± 44.7 <sup>a</sup>	936.1 ± 34.6 <sup>ab</sup>
Relative growth rate (%) <sup>*</sup>	84.76 ± 0.03 <sup>a</sup>	84.68 ± 0.03 <sup>a</sup>	81.75 ± 0.02 <sup>a</sup>
Feed conversion ratio	2.07 ± 0.098 <sup>a</sup>	1.80 ± 0.09 <sup>b</sup>	1.92 ± 0.06 <sup>ab</sup>
Feed intake (g/bird)	1835.8	1826.8	1760.8
<b><u>Total RGR and CR:</u></b>			
Total body weight (g)	1529.6 ± 48.5 <sup>b</sup>	1765.3 ± 41.4 <sup>a</sup>	1609.7 ± 31.3 <sup>b</sup>
Total body gain (g)	1484 ± 48.5 <sup>b</sup>	1720.3 ± 41.4 <sup>a</sup>	1564.7 ± 31.3 <sup>b</sup>
Relative growth rate (%)	161.11 ± 0.01 <sup>b</sup>	165.73 ± 0.01 <sup>a</sup>	164.63 ± 0.01 <sup>a</sup>
Feed conversion ratio	2.08 ± 0.07 <sup>a</sup>	1.77 ± 0.04 <sup>b</sup>	1.86 ± 0.03 <sup>b</sup>
Total Feed intake (g/bird)	3047.2	3002.8	2886.8
Mortality %	0.0	0.0	0.0

<sup>abcd</sup> Values in the same row with a different superscript differ significantly at P < 0.05

<sup>\*</sup> Relative growth rate =  $(W_2 - W_1) / (W_1 + W_2) / 2 * 100$  <sup>4)</sup>

**Table 3. Effect of IMO supplementation on carcass traits of broilers**

	Control	T1	T2
Dressing %	72.0 ± 0.003 <sup>b</sup>	73.2 ± 0.00 <sup>ab</sup>	75.6 ± 0.013 <sup>a</sup>
Breast%	12.7 ± 0.00 <sup>c</sup>	15.1 ± 0.02 <sup>b</sup>	17.9 ± 0.00 <sup>a</sup>
Liver %	1.83 ± 0.00 <sup>c</sup>	1.99 ± 0.01 <sup>b</sup>	2.19 ± 0.01 <sup>a</sup>
Gizzard %	2.11 ± 0.01 <sup>c</sup>	2.45 ± 0.00 <sup>b</sup>	2.78 ± 0.00 <sup>a</sup>
Heart %	0.47 ± 0.00 <sup>b</sup>	0.49 ± 0.00 <sup>b</sup>	0.55 ± 0.01 <sup>a</sup>
Bursa of fabricios %	0.16 ± 0.00 <sup>b</sup>	0.17 ± 0.00 <sup>ab</sup>	0.18 ± 0.00 <sup>a</sup>
Spleen %	0.06 ± 0.00 <sup>b</sup>	0.08 ± 0.00 <sup>a</sup>	0.08 ± 0.00 <sup>a</sup>

<sup>abcd</sup> Values in the same row with a different superscript differ significantly at P < 0.05

**Table 4. Effect of IMO supplementation on the antibodies titer against avian flu and Newcastle**

	Control	T1	T2
21 day	2.0 ± 0.4 <sup>b</sup>	4.5 ± 0.5 <sup>a</sup>	4.0 ± 0.41 <sup>a</sup>
28 day	4.25 ± 0.25 <sup>b</sup>	5.0 ± 0.58 <sup>b</sup>	6.6 ± 0.24 <sup>a</sup>
35 day	5.67 ± 0.33 <sup>b</sup>	9.0 ± 0.00 <sup>a</sup>	9.0 ± 0.00 <sup>a</sup>

<sup>abcd</sup> Values in the same row with a different superscript differ significantly at P < 0.05

**Table 5. Effect of IMO supplementation on the blood parameters of broilers**

Items mg/dl	Control	T1	T2	P
Total protein	3.51 ± 0.11	3.4 ± 0.11	3.23 ± 0.07	NS
Albumen	1.42 ± 0.11	1.62 ± 0.08	1.54 ± 0.07	NS
Globulin	205.5 ± 2.62 <sup>b</sup>	228.03 ± 5.22 <sup>a</sup>	223.5 ± 3.23 <sup>a</sup>	
Triglycerides	80.6 ± 3.37	70.0 ± 2.36	61.9 ± 8.3	NS
Cholesterol	90.37 ± 3.6	82.7 ± 3.01	83.4 ± 3.05	NS
HDL	170.8 ± 46.3	160.5 ± 37.4	169.6 ± 16.0	NS
LDL	92.03 ± 10.0	84.7 ± 3.50	77.15 ± 6.40	NS
VLDL	37 ± 0.6 <sup>a</sup>	33.4 ± 1.0 <sup>b</sup>	32.9 ± 1.1 <sup>b</sup>	
Malondialdehyde	11.74 ± 1.57 <sup>b</sup>	23.12 ± 4.92 <sup>a</sup>	18.78 ± 2.56 <sup>a</sup>	NS
Glutathione reductase	0.90 ± 0.17 <sup>b</sup>	1.93 ± 0.12 <sup>a</sup>	1.46 ± 0.21 <sup>a</sup>	

<sup>abcd</sup>Values in the same row with a different superscript differ significantly at P < 0.05

NS: Nonsignificant difference (P>0.05)

## Author' biography



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