SYNERGISTIC EFFECT OF DIETARY LIVE YEAST AND OLIGOSACCHARIDE ON GROWTH PERFORMANCE OF BROILER CHICKEN

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This is to certify that the thesis entitled, "SYNERGISTIC EFFECT OF DIETARY LIVE YEAST AND OLIGOSACCHARIDE ON GROWTH PERFORMANCE OF BROILER CHICKEN" Submitted to the Department of Animal Nutrition, Genetics and Breeding, Faculty of Animal science and veterinary medicine, Sher-E-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (MS) in Animal Nutrition embodies the result of a piece of bonafide research work carried out by Sharmin Akhter, Registration No. 17-08229 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has been duly acknowledged by her.

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ABBREVIATION		FULL MEANING
ADO	=	Alginate-derived oligosaccharide
AGP	=	Antibiotic growth promoters
ANOVA	=	Analysis of variance
Avg.	=	Average
BCR	=	Benefit cost ratio
BWG	=	Body weight gain
Ca	=	Calcium
CFU	=	Colony forming unit
CM^2	=	Square centimeter
COS	=	Chito-oligosaccharide
DOC	=	Day old chick
E. coli	=	Escherichia coli
e.g.	=	For example
et al.	=	And others/associates
EU	=	European union
FAO	=	Food and agricultural organization
FC	=	Feed consumption
FCR	=	Feed conversion ratio
FI	=	Feed intake
FMO	=	Fructo monosaccharide
FOS	=	Fructo oligosaccharide
Ft	=	Feet
G	=	Gram
i.e.	=	That is
IB	=	Infectious bronchitis
IBD	=	Infectious bursal disease
K Cal	=	Kilo calorie
Kg	=	Kilogram
L	=	Litre
Μ	=	Metre
M.S	=	Master of science

LIST OF ACRONYMS AND ABBREVIATION

ABBREVIATION		FULL MEANING
ME	=	Metabolizable energy
Ml	=	Mililitre
Mm	=	Milimetre
MOS	=	Mannan oligosaccharide
ND	=	Newcastle disease
No.	=	Number
NS	=	Non-significance
Р	=	Phosphorus
PPB	=	Profit per bird
RH	=	Relative humidity
SAU	=	Sher-e-bangla agricultural university
SC	=	Saccharomyces cerevisiae
SCFOS	=	Short chain fructo oligosaccharide
SE	=	Statistical error
SPSS	=	Statistical package for social sciences
Viz.	=	Such as
Vs.	=	Versus
WHO	=	World health organization
Wks.	=	Weeks
YC	=	Yeast culture
YE	=	Yeast extract

ACRONYMS AND ABBREVIATION (CON'D)

SYMBOLS		FULL MEANING
*	=	5% level of significance
&	=	And
@	=	At the rate of
°C	=	Degree celcius
°F	=	Degree Fahrenheit
\$	=	Dollar
>	=	Greater than
<	=	Less than
/	=	Per
%	=	Percentage
±	=	Plus-minus
:	=	Ratio

LIST OF SYMBOLS

SYNERGISTIC EFFECT OF DIETARY LIVE YEAST AND OLIGOSACCHARIDE ON GROWTH PERFORMANCE OF BROILER CHICKEN

BY SHARMIN AKHTER

ABSTRACT

This study was conducted to evaluate the effect of feeding graded levels of live yeast (Saccharomyces cerevisiae) and oligosaccharide on broiler performance. One-day old of Cobb-500 broiler chicks (n=150) were randomly allocated into five treatments. Each dietary treatment consisted of 3 replicates having 10 broilers in each of the replication. The dietary treatment contained no live yeast and oligosaccharide considered as control (T_0) and the other four treatments were T_1 (1g yeast and 0.5g oligosaccharide/kg feed), T_2 (2g yeast and 0.5g oligosaccharide/kg feed), T_3 (1g yeast and 1g oligosaccharide/kg feed) and T_4 (2g yeast and 1g oligosaccharide/kg feed). During the experimental periods of 4 weeks, feed intake, body weight gain, feed conversion ratio (FCR), survivability, flock uniformity values were calculated. Growth performance parameters were significantly (P<0.05) affected by experimental diets. Birds fed 1g yeast and 1g oligosaccharide/kg feed gained superior body weights (1765.90±4.89g) compared to control $(1469.43\pm18.29g)$, and other dietary treatment. The mean body weight gains (g) at the 1st, 2nd, 3rd and 4th week of different treatment groups were significantly higher (P<0.05) than control group. The feed intake of T_3 (1g yeast and 1g oligosaccharide/kg feed) group was lower (2307.30±3.66g) compared to control (2421.07±25.99g). The groups fed diets containing 1g yeast and 1g oligosaccharide had lower FCR (1.33±0.00) compared to control (1.69±0.03). The inclusion of different dietary treatments had no significant (P>0.05) effects on survivability and flock uniformity. It is concluded that live yeast and oligosaccharide can be included in broiler diet at the rate of 1g yeast and 1g oligosaccharide/kg feed for better performance and higher economical return.

CHAPTER-1 INTRODUCTION

CHAPTER 1 INTRODUCTION

Modern intensive poultry production produces market ready broiler chicken with in four weeks of their age. Feed as a major input item of broiler rearing which occupies 75% of the production cost and has a vital role in broiler economics. The major objectives of poultry farming are to increase the profit margin by improving feed efficiency and exploiting maximum growth potential of the birds. Hence, it is imperative to give due attention to proper utilization of feed without adversely affecting the growth or production performance of broilers (Kokje, 1999). In recent years, there has been great attention to minimize or completely avoid usage of antibiotics in animal and poultry feeding, as well as an increasing consumer concern for poultry drug resides in meat and egg. Hence, non-antibiotic alternative live probiotic, prebiotic, symbiotics and phytobiotic are being used in poultry feed to improve growth and production performance.

Feed additives were used in poultry industry for different purposes for example to increase performance and decrease of mortality rate. These additives are including antibiotics, probiotics, coccidiostats etc. (Panda *et al.*, 2009). Among these feed supplements, probiotics have drawn much great attention. In recent years use of growth promoters, like yeast in poultry industry the world over of some can effective in decrease of feed intake, cost and increase of gain weight and amend of violations. *Saccharomyces cerevisiae* (SC), one of the most widely commercialized yeast and it was reported that feeding yeast to chicks improved weight gain and feed/gain ratio (Nilson *et al.*, 2004). Due to increasing population, there is an increasing demand for meat and eggs which led to commercialization of poultry production, with a large number of farms now operating across the country (Raha, 2007). One of the major challenges this industry faces is the spreading of diseases among the poultry population due to bacterial pathogens which results in serious economic losses (Huque *et al.*, 2011). As a result, the use of antimicrobial agents and growth promoters is substantially increasing in the poultry industry to prevent diseases and to promote faster growth (Islam *et al.*, 2016).

Prebiotics are a possible alternative to antibiotics in poultry diets. Prebiotic usually refers to oligosaccharides which are not digested by the animal enzymes, but can selectively stimulate certain intestinal bacteria species, which have potential beneficial effects on the host health. While probiotics are meant to bring beneficial microbes to the gut, oligosaccharides are supposed to selectively stimulate the beneficial microbes that already live there (Yang *et al.*, 2007). Prebiotic have two advantages relative to probiotics: a technological, because there are no problems with the thermal processing of the feed and the acidic conditions of the digestive system, and a safety, because there is no introduction of any foreign microbial species into the gut. However, similar to probiotics, results of the effects of prebiotics on broiler performance are contradictory.

FAO/WHO (2001) describes feed additive as 'live micro- organisms which, when administered in adequate amounts, confer a health benefit on the host. However, increasing concerns regarding over use of antibiotics and bacterial resistance has encouraged extensive investigation into alternatives for sub-therapeutic antibiotics in feeds. Probiotics act by competitive exclusion, lower gut pH, produce bacteriocins, of lysozyme and peroxides, and stimulate the immune system (Grashorn, 2010).

The presence of living yeast cells may also act as a reservoir for free oxygen, which could enhance growth of other anaerobes (Leeson and Summers, 2008). The action mechanism of live yeast for improving performance is not fully understood, but there are two probabilistic explanations. The first, action of yeast is most probably supporting the growth of lactic acid bacteria. The second, a competitive exclusion of pathogenic bacteria by yeast and its products especially the cell wall component (Onifade, 1998).

Mannan oligosaccharide (MOS) is one of the alternatives being explored to replace AGP. One of major mechanisms of actions for MOS is to act as a receptor analogue to block pathogens, which possess mannose-binding-lectin, from attaching to the gut wall (Spring *et al.*, 2000). Nowadays, there has been growing interest among researchers and the feed industry to prepare a probiotic feed supplement containing a combination of beneficial microbial strains to obtain a synergistic interaction for better growth performance, health status, and product quality of poultry.

Mannan oligosaccharides (MOS), which are derived from the cell wall of *Saccharomyces cerevisiae*, are an alternative to antibiotic growth promoters. It contains phosphorylated mannans, glucans and some protein intermixed. MOS have shown promising effects, such as decreasing pathogenic microflora of the gut, stimulating a strong immune response, and elevating the strength of the intestinal mucosa in studies with poultry (Spring, 1999a; Spring, 1999b; Spring *et al.*, 2000). By balancing the intestinal microflora and stimulating the immune response, MOS have been shown an increase at

the growth of broilers (Hooge, 2004). MOS supplementation to broiler diets improves growth performance in terms of body weight gain and feed conversion (Hooge, 2004;

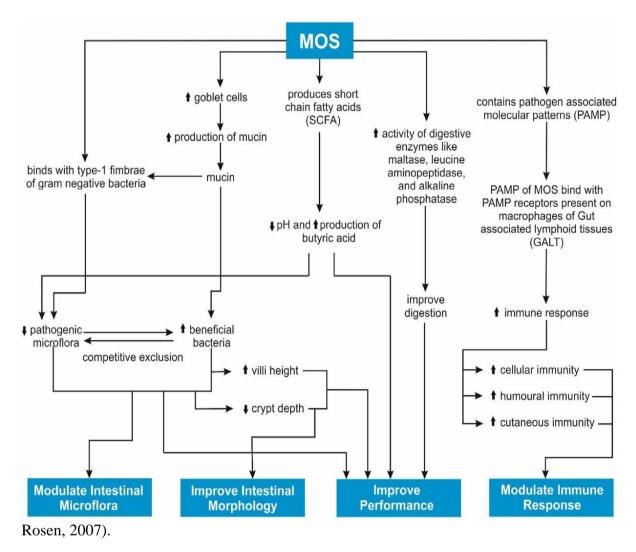


Figure 1: Mechanism of MOS

Source: Chacher (2017)

1.1 Objectives

With this background, the work was planned to explore the possibilities of live yeast (*Saccharomyces cerevisiae*) and oligosaccharide on performance of broiler chicken with the following specific objectives:

- To investigate the synergistic effects of yeast (*Saccharomyces cerevisiae*) and oligosaccharide on growth performance of broiler chicken
- To recommended the inclusion level of live yeast and oligosaccharide in broiler ration
- To determine the flock uniformity of broiler chicken under different treatment

CHAPTER-2

REVIEW OF LITERATURE

CHAPTER 2 REVIEW OF LITERATURE

Performing any type of survey or experiment review of literature is important which are linked to the proposed study for the convenient of research work. During the last decade, different studies have been attempted to find nutrition- based health approaches and natural feed additives to improve performance and immunity of poultry, and strongly recommended the use of probiotics, prebiotics, phytogenic additives or organic acids. Among these feed supplement oligosaccharide and live yeast individually have drawn much great attention. Nowadays, there has been growing interest among researchers and the feed industry to prepare a probiotic feed supplement containing a combination of live yeast and oligosaccharide to obtain a synergistic interaction for better growth performance, health status, and product quality of poultry. The literature reviewed here have been limited to these which are considered compatible and related to the objectives of the present study.

2.1 Effect of live yeast on growth performance

Rodriguez *et al.* (2003) reported that yeast cell wall is containing chitin, mannan and glucan that have been known as immune stimulant. The presence of living yeast cells may also act as a reservoir for free oxygen, which could enhance growth of other anaerobes.

Reed and Nagodawithana (1999) stated that *Saccharomyces cerevisiae* (SC), one of the most widely commercialized types of yeast, rich in crude protein (40-45%) and its biological values were high and also rich in vitamin B complex, biotin, niacin, pantothenic acid and thiamin.

Santin *et al.* (2001) and Santin *et al.* (2003) showed the cell wall as SC improve the intestinal mucosa aspects and suggested that it might be the explanation for the improve in performance of broilers supplemented with cell wall of SC observed in the same study.

Nilson *et al.* (2004) reported broilers receiving yeast to replace part of the premix had better average weight gain and feed conversion ratio. Yeast products are important natural growth promoters. Several digestive enzymes are also excreted by the yeast that help the gastrointestinal tract to boost the nutrient digestibility, growth rate and feed conversion ratio.

Churchil and Mohan (2000) found better weight gain and feed conversion in broilers fed from 0.2 to 1% brewer's yeast. Also, 5-20% brewer yeast also improve growth performance of broiler chicken. Experiments showed that inactive form of SC cells very effective. Alive form of SC, such of probiotics have a low active, this possible by reason that inactive SC can lower defense from internal organs of the body.

Hyginus and Chukwu (2003) surveyed the dietary *Saccharomyces cerevisiae* and mannan oligosaccharide in reduced the deleterious effects of heat stress on White leghorn laying hens and the results proved that the mean of weight has increased in the groups having *Saccharomyces cerevisiae* and mannan oligosaccharide were added singly and combined at 0.05% per kg of feed, the mean of gain weight has been 58 g among groups.

Duk and Zhang (2004) reported that using of *Saccharomyces cerevisiae* supplement on the growth performance showed that the performance of the broilers which were fed from different levels of SC has increased in three-week old (P<0.05) and this increase can be witness in five-week-old as well.

Patterson and Burkholder (2003) reported that *Saccharomyces cerevisiae* is considered as one of the live microorganism's probiotic that, when administered through the digestive tract, have a positive impact on the hosts health through its direct nutritional effect.

Barnet *et al.* (2000) reported that using of *Saccharomyces cerevisiae* supplement on the growth performance showed that the performance of the broilers which were fed from different levels of SC has increased in three-week old (P<0.05) and this increase can be witness in five weeks old as well. But by increase the rate of SC in (P<0.05) feed intake in the groups fed by enriched SC has been low compared with gain weight. On the whole they point that there was no significant difference between treatment and control group used from SC in (P>0.05).

Barnet *et al.* (2000) reported that *Saccharomyces cerevisiae* are yeast organisms that unicellular gram-positive stain would be unable to adhesion to the intestinal wall, but it is capable of high consumption of oxygen and thus provides the presence of anaerobic conditions suitable for the growth and proliferation of *lactobacilli*.

Loddi *et al.* (2002) published that the yeast presents freely in the gastro intestinal tract cavity and it is called transient microorganisms that means it does not stick in the epithelial cells, and sorting of enzymes to the intestines which works to increase the

readiness of nutrients to feed as well as increase the percentage of digested protein in the gut of poultry.

Toma *et.al.* (2005) reported that *S. boulardii*, the probiotic being tested, is one supplement that may provide enteric benefits for broilers. It was originally isolated from the lychee plant from India and Southeast Asia. Locals had been using the fruit as a cure for diarrhea. Now one source of this yeast is in a product called Luvacell, produced by an animal health company called Lallem and One study showed that supplementing broiler feed with yeast decreased feed consumption and improved feed efficiency, while also increasing villi size as compared with an AGP.

Kelesidis (2012) reported that *Saccharomyces boulardii* may work in several different ways: helping to control intestinal homeostasis, by preventing pathogens from colonizing, by promoting beneficial enzyme production, by improving the gastrointestinal lining permeability, or by improving immune responses.

Stanley *et al.* (2004) reported that yeast could be used as an alternative for antibioticbased drugs in feed in broiler chicks or in recycled litter. It well documented that antibiotic have beneficial effect on animal growth performance and health. However, increasing concerns regarding over use of antibiotics has promoted extensive investigations into alternative to use the sub-therapeutics antibiotics in production yeast.

Zhang *et al.* (2005) observed greater villi height and superior ileal mucosa development at 21 d in chickens supplemented with a yeast cell wall product prepared from *Saccharomyces cerevisiae*. Histology results revealed significantly greater goblet cell densities and sizes for chicks receiving cell wall preparations than those of the control treatment chicks, while villi width and height measurements indicated no differences between treatments.

Rutz et al. (2006) reported that yeast extract is also derived from the cell content of live yeast and contain high levels of nucleotides, inositol and glutamic acid and have also resulted in beneficial effects on the feed conversion of broilers.

2.2 Synergistic effect of live yeast on growth

Stanley *et al.* (2004) showing that whole cells and several derivatives from yeast such as yeast culture (YC), yeast extracts (YE) and yeast fermentation products, that retain the

cell wall components, exert similar beneficial effects as MOS on the growth and feed conversion ratio of poultry.

Saleh *et al.* (2013) showed that dietary supplementation with *S. cerevisiae* and *A. awamori* improved the growth performance of broilers synergistically by increasing muscle protein metabolism. Yeast cell wall preparations could contribute to the gastrointestinal health and performance of broiler chickens by affecting mucus secreting goblet cells in a favourable manner.

Stanley *et al.* (2004) reported that improved growth performance have been noticed in broilers fed YC, YC residue, whole cells, cell wall components or a fermentation product from yeast. Yeast culture contains viable cells, cell wall components, metabolites, and the media on which the yeast cells were grown, the addition of a soluble fraction of YC showed an anti-inflammatory effect in conjunction with activation of natural killer cells and B lymphocytes.

Tortureo (1973) reported that in broiler nutrition, probiotic species belonging to *Saccharomyces, Lactobacillus, Streptococcus, Bacillus, Bifidobacterium, Enterococcus, Aspergillus* and *Candida*, and have a beneficial effect on broiler performance, modulation of intestinal microflora and pathogen inhibition, and promoting microbiological meat quality of broilers.

Stanley *et al.* (2004) published that under stress conditions, YC have a beneficial effect on broiler performance when birds were challenged with *Eimeria spp.* or aflatoxin. The response to antimicrobial agents was greater in a "dirty" environment.

Santin *et al.* (2001) and Zhang *et al.* (2005) reported greater villus height and improved performance in birds with supplementation of whole yeast or yeast cell wall. Cell wall components of YC (β -glucans and α -mannans) may provide a protective function to mucosa by preventing pathogens from binding to villi and allowing fewer anti-gens to be in contact with the villi.

Montes and Pugh (1933) reported that probiotics, such as *Saccharomyces cerevisiae*, are defined as non-digestible ingredients with mannan oligosaccharide and they have several modes of action: beneficial changes in gut flora with reductions in the population of pathogenic bacteria, lactate production with subsequent changes in intestinal pH, production of antibiotic-type substances, production of enzymes, competition for

adhesion receptors in the intestine, competition for nutrients, reduction of toxin release, and immuno-stimulation.

2.3 Effect of oligosaccharide on growth performance

Spring *et al.* (2000) reported that mannan oligosaccharides (MOS) are mannose-based carbohydrates found in the yeast cell wall and are capable of adsorbing enter pathogens. MOS supplementation had no discernable effect on body weight gain or composition during this 12-week study, challenging the potential use of MOS as a calorie restriction mimetic or body composition enhancer.

Kocher *et al.* (2004) reported that MOS have been shown to improve nutrient utilization through stimulation of specific microbial populations in the gastrointestinal tract. Mannan-oligosaccharides (MOS), is a type of probiotics originated from the yeast cell wall (*Saccharomyces cerevisiae*) has gained more prominent attention, mainly due to its ability to bind the threadlike fimbriae on pathogenic bacteria preventing them from attaching to the gut wall, thereby averting their stabilization and the resulting colonization and multiplication, up to the disease level, so it had been showed to be a most capable solution for antibiotic-free diets, as well as furnishing effective support for digestion and immunity in poultry.

Mc Donnell *et al.* (1989) reported that the addition of *Saccharomyces cerevisiae* (yeast) to the feed increases the production of calcitriol receptors. It plays a role as an antioxidant, helping with mineral retention, improving bone mineralization and subsequently the overall improvement the performance of poultry birds.

Spring *et al.* (2000) published that mannan oligosaccharides (MOS), which are derived from the cell wall of *Saccharomyces cerevisiae*, are an alternative to antibiotic growth promoters. MOS have shown promising effects, such as decreasing pathogenic microflora of the gut, stimulating a strong immune response, and elevating the strength of the intestinal mucosa in studies with poultry.

Hooge (2004) reported that MOS supplementation to broiler diets improves growth performance in terms of body weight gain and feed conversion. Dietary MOS improved the growth performance of birds given the wheat-based diet compared to that of birds given the corn-based diet during 7-21 days of age. The addition of MOS modulated the development of gut microflora. From day 7 to day 21, the numbers of mucosa-associated coliforms along the small intestine were decreased; whereas the numbers of mucosa-

associated lactobacilli were increased by MOS, regardless of the cereal type in the diets. Dietary MOS also reduced the counts of coliforms and *Clostridium perfringens* in the caeca of birds by 21 days of age.

Gibson and Roberfroid (1995) reported that the inhibition of pathogenic bacteria, such as *Salmonella* and *Campylobacter spp.* or putrefactive bacteria such as *Clostridium perfringens* can partially be explained by the fermentation products of these oligosaccharides in the intestine.

Bezkorovainy (2001) reported that MOS contributes in the described ways to increase of vitality in animals, reduction of losses and improvement in food utilizing, what causes optimal results in production and acceptable economic effects, so for a long time in a world they have been an integral part of majority of industrial mixture for poultry nutrition.

Liukkonen-Anttila (2001) published that, mode of action mannasase based in compatibility structure of mannose and lectins which are on pills as surface and fimbriae of bacteria. The addition of mannan-oligosaccharide results with complex mannans-bacteria and prevent adherence pathogens to intestinal wall although, bacteria have the other mechanisms of adherence for intestinal epithelia cells which are resistant on mannose inhibition, a great number of *E. coli* (66%) and *Salmonella* (53%) stains, have mannose sensitive adhesions.

Erener *et al.* (2009) stated that non-dissolved (indigestible) MOS pass to distal parts of digestive system. That is the way to prevent colonization of distal parts of digestive system with pathogens and their elimination. In unfavorable condition (change pH of intestinal content and lesion of intestinal mucosa) and due to passage of pathogens in front parts gastrointestinal tract, MOS acts on the same way.

Hatemink (1995) published that, selective activity of MOS are based on fact that beneficial bacteria in digestive system (*Bifidobacterium longum*, *Lactobacillus casei*, *L. acidophilus*, *L. delbrekii*) contain manase enzyme which prevent making complex. That ensures selectivity binding mannan oligosaccharide only for pathogens which normally do not have this enzyme. Previously described mode of binding MOS is not typical only for bacteria. Some toxin, viruses and eukaryotic cells have the ability to recognize determined sugar on surface the other cells too.

Gibson and Roberfroid (1995) stated the examination of the MOS effects on production results in broilers showed that daily increase gain was 4-8%. At the same time, in the case of the same or lower consumption there was significantly better conversion for 5-8%. Statistical differences in broilers mortality between groups were not found but numerically it was lower in groups which were fed with MOS. Also, comparative examinations established that differences between broilers performance which were fed with mixtures containing antibiotics, that is, prebiotics was not significant, but the both additives are equally effective in promoting growth.

Awad *et al.* (2008) reported that the effects of MOS on poultry production can be expressed in reduction of diseases by inhibition of pathogenic bacterial colonization to gut lining by binding to them and thus preventing them of proliferating and producing toxins, reducing intestinal pathogen counts, improving the immune system and exhibit influence on morpho-functional characteristics of intestines.

Strickling *et al.* (2000) reported that the small intestine does not contain the digestive enzymes required to break down mannan oligosaccharide bonds, therefore they arrive at the large intestine intact after ingestion and passage through the small intestine.

Hooge *et. al.* (2003) reported that the addition of MOS to broiler chicken diets was reported to have positive effects on growth performance but the supplementing levels of MOS varied by trials and by feed phase in different studies, ranging from 0.5 g MOS/kg diet to 5 g MOS/kg diet.

Spring *et al.* (2000) reported that mannan oligosaccharide (MOS) is one of the alternatives being explored to replace AGP. One of major mechanisms of actions for MOS is to act as a receptor analogue to block pathogens, which possess mannose-binding-lectin, from attaching to the gut wall.

2.4 Synergistic effect of oligosaccharide on growth

Pelicano *et al.* (2005) reported that MOS and *Bacillus* combined may reduce the depth of the crypt, due to a lower enterocyte replacement rate, and by increasing the villus density in the duodenum.

Parks *et al.* (2001) reported that MOS is derived from mannans on yeast cell surfaces. The benefits of MOS are based on specific properties, including modification of the intestinal micro-flora, reduction in turnover rate of the intestinal mucosa, and modulation of the immune system in the intestinal lumen. These properties have the potential to enhance growth rate, feed efficiency, and livability in poultry species.

Nicholasville (1997) reported oligosaccharides are carbohydrates that yield 2 to 10 monosaccharides upon hydrolysis. The idea to use yeast MOS in poultry feeds evolved from the concept that certain sugars, particularly mannose, could be used to largely block the colonization of intestinal pathogens such as *Salmonella* species and *Escherichia coli*, which contain type 1 fimbriae with mannose-seeking lectins. When they bind to the MOS product, the pathogens are prevented from attaching to intestinal mannose, proliferating, and producing toxins. A second reason for developing the MOS product was because of the effectiveness of some strains of live yeast at binding and reducing intestinal pathogen counts.

Yan *et al.* (2011) reported that the supplementation of alginate-derived oligosaccharide (ADO) and chito-oligosaccharide (COS) was also found to be effective against *Salmonella* colonization.

Hidaka *et al.* (1986) reported that fructo oligosaccharides (FOS) that include inulin, oligofructose, and short-chain fructo oligosaccharide (SCFOS) can be fermented by bifido bacteria and lactobacilli. These 2 bacteria are generally classified as beneficial bacteria. These FOS also may help control or reduce the growth of harmful bacteria such as *Clostridium perfringens*, which is especially important to the poultry industry because this bacterium is a primary cause of necrotic enteritis that has been estimated to cost the worldwide poultry industry \$2 billion each year.

Newman (1994) reported that mannose is the main component of MOS and is unique because it is bound by the type 1 fimbriae used by many enteric bacteria to attach to host cells. Therefore, mannose can result in the movement of undesirable bacteria through the intestine without colonization.

Santin *et al.* (2001) published that probiotics are mainly represented by mannan oligosaccharides (MOS) and fructo oligosaccharides (FOS), present in the cell wall of yeasts, such as *Saccharomyces cerevisiae*. They exert their action by maintaining or reestablishing the conditions of eubiosis in the digestive tube, and thus, the normal microbial flora and the balance of the gastrointestinal tract.

Oyofo *et al.* (1989) reported that eutropic bacteria and mannan oligosaccharides are added, balance conditions become permanent, preventing the establishment of salmonella, *E. coli*, clostridium, among others, and increasing the number of beneficial lactic-acid producing bacteria, thus maintaining eubiosis.

CHAPTER-3

MATERIALS AND METHODS

CHAPTER 3 MATERIALS AND METHODS

3.1 Statement of the experiment

The study on effects of live yeast and oligosaccharide on growth performance in broilers was carried out in poultry farm, Sher-e-Bangla Agricultural University and in the laboratory of Animal Nutrition, Genetics and Breeding, Faculty of Animal Science and Veterinary Medicine, Sher-e-Bangla Agricultural University, Dhaka-1207 during the period from 21th February 2019 to 20th March 2019.

3.2 Collection of experimental broilers

A total of 150 day old chicks of "Cobb-500" strain having 43.2±0.3g average body weight were obtained from Kazi hatchery, Gazipur, Dhaka.

3.3 Experimental design

A total of 150, day old Cobb-500 strains were collected from Kazi Hatchery. At day 7, broiler chicks were randomly divided into 5 experimental group of 3 replicates each with 10 chicks per replicate. Birds were housed in 3ft x 2ft floor pens on fresh rice husk litter with a 24-h lighting plan. The height of litter was 3 cm. Before being used in the experiment, birds were adapted for 7 days in order to acclimatize in the environment. The experimental diets were prepared by supplementing the control diet with different levels of yeast *Saccharomyces cerevisiae* (Levucell- SB) at concentration of 1.0×10^{10} CFU/gm and oligosaccharide (Original XPC_{TM}). The control diet was formulated without supplementation of any yeast and oligosaccharide. The required amount of yeast and oligosaccharide weighed and initially mixed with small amount of feed and then mixed with bulk quantity of feed. The collected birds have neither developmental disorders, detectable genital diseases nor other diseases that may cause any problem in the experiment or affect the result of the experiment.

3.4 Experimental materials

The collected chicks were carried to the university poultry farm early in the morning. They were kept in electric brooder protocol. During brooding time only basal diet was given no yeast and oligosaccharide was used as treatment. Among 150 DOC, 120 chicks were selected from brooder and distributed randomly in four (4) treatments of yeast and oligosaccharide, remaining 30 chicks were distributed another treatment of control.

The chicks of each treatment group were divided into three (3) replications and in each replication, there were 10 birds. Each pen was provided with feeder and drinker. Feed and water were offered *adlibitum*. After 28 days of, data were collected for the following parameters: feed intake, live weight, body weight gain, feed conversion ratio, profit per bird and benefit-cost ratio.

3.5 Experimental treatments

- $T_{1\,=}\,1g$ yeast and 0.5g oligosaccharide/kg feed
- $T_2 = 2g$ yeast and 0.5g oligosaccharide/kg feed
- $T_{3=}$ 1g yeast and 1g oligosaccharide/kg feed
- $T_{4=}2g$ yeast and 1g oligosaccharide/kg feed

 T_{0} = Normal feeding & watering without any addition of yeast and oligosaccharide.

Treatment groups	No.	of replications	5	Total
	R ₁	R ₂	R ₃	_
T ₀	10	10	10	30
T_1	10	10	10	30
T_2	10	10	10	30
T ₃	10	10	10	30
T_4	10	10	10	30
Total	50	50	50	150

Table 1. Layout of the experiment

3.6 Preparation of broiler house

The broiler shed was an open sided natural house. Cross ventilation system was provided by using wire-net. It was a tin shed house with concrete floor. There was 1ft. side wall around the shed with no ceiling. The floor was above 1ft. from the ground and the top of the roof was above 15 ft. from the floor. Polythene sheet was hanged around the side wall to protect the chicks from cold, storm, dusts and heavy rainfall. The house was properly cleaned, rubbed with bleaching powder and washed the floor by using tap water and then disinfected by diluted iosan solution before starting the experiment. After proper drying of floor, equal size $(1 \text{ m} \times 1 \text{ m})$ wire net floor pens were made as per layout of the experiment. The height of the pens was 0.5m. Before placement of chicks the house was fumigated by formalin and potassium permanganate @ 500 ml formalin and 250 g potassium permanganate (i.e. 2:1) for 35 m^3 experimental area.

3.7 Experimental diets

Starter and grower commercial fresh broiler feed were purchased from the market. Starter diet was enriched with minimum:

Starter ration	Minimum percentage present (%)
Moisture	11
Protein	24
Fat	5.80
Calcium	1.30
Phosphorus	0.60
ME (kcal/kg)	3050
Grower ration	Minimum percentage present (%)
Moisture	11
Protein	23.50
Fat	5.80
Tat	5.00
Calcium	1.20

Feed were supplied 4 times daily by following Cobb 500 Manual and *ad libitum* drinking water 2 times daily.

3.7.1 Collection of live yeast and oligosaccharide

Levucell SB is the probiotic live yeast *Saccharomyces cerevisiae*. It is available in many other countries outside the EU. Levucell SB benefits from the patented titan technology of microencapsulation that protects the live yeast against extreme pelleting process (heat, pressure, humidity) and interaction with other chemical compounds used in fed manufacture. It was collected from Md. Shariful Islam, Director, SMC Poultry and Hatchery Ltd. Oligosaccharide (Original XPC_{TM}) is a natural nutritional health product used in all types of animal diets. Product of the USA. Original XPC_{TM} are available in the market of our country and it was collected from EON Group Company. Photographs of Live yeast and oligosaccharide were given in below:

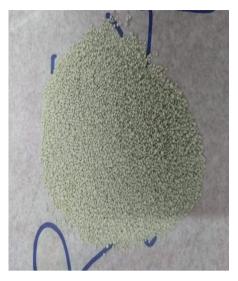


Plate 1. Live yeast



Plate 2. Oligosaccharide

Table 3. Nutritional composition of live yeast and oligosaccharide

Live yeast (Levucell SB)		
Saccharomyces cerevisiae contain 1.0×10^{10} CFU/g	gm (Microencapsulated	
formulation for premix & concentrated formulation for	or premix and pelleted	
feed)		
Oligosaccharide (Original XPC _{TM})		
Nutrient Component	Amount (%)	

Nutrient Component	Amount (%)
Crude Protein (min.)	15.0
Crude Fat (min.)	1.5
Crude Fiber (max.)	25.0
Ash (max.)	9.0
Moisture (max.)	11.0

Typical Analysis (as fed)		
	Amino Acids	Amount (%)
Arginir	ne	0.78
Cysteir	ie	0.45
Glycine	2	0.92
Histidi	ne	0.42
Isoleuc	ine	0.56

	1	Typical Analysis (as fed)
	Amino Acids	Amount (%)
Leucine		1.13

Lysine	0.81
Methionine	0.33
Phenylalanine	0.62
Proline	1.06
Threonine	0.63
Tyrosine	0.60
Tryptophan	0.23
Valine	0.81
Minerals	Amount (%)
Calcium (Ca)	0.53
Chloride (Cl)	0.42
Magnesium (Mg)	0.42
Phosphorus (P)	0.54
Potassium (K)	2.43
Sodium (Na)	0.05
Sodium (Na) Sulfur (S)	0.05 0.42
Sulfur (S)	0.42
Sulfur (S) Carbohydrates	0.42 Amount
Sulfur (S) Carbohydrates Starch	0.42 Amount 5.20

ADF = Acid Detergent Fiber; NDF = Neutral Detergent Fiber;

3.7.2 Preparation of feed with live yeast and oligosaccharide

Saccharomyces cerevisiae contains 1.0×10^{10} CFU/gm microencapsulated formulation for premix & concentrated formulation for premix and pelleted feed. Rate of use of live yeast was 1kg/metric ton feed. Recommendation dose of oligosaccharide for broiler starter is 100-125gm/100 kg feed and broiler grower 50gm/100 kg feed. At first 1g live yeast and 2g live yeast was measured separately and then the yeast was mixed with feed properly according to the inclusion level in different treatment. With the help of microbalance 0.5g oligosaccharide and 1g oligosaccharide was mixed properly with the commercial broiler feed. Mixing of live yeast and oligosaccharide was done carefully so that each of the ingredient of feed can close contact with the live yeast and oligosaccharide. After mixing the live yeast and oligosaccharide with commercial broiler feed, the recommendation level of feed allowed for 1^{st} , 2^{nd} , 3^{rd} and 4^{th} week was separated in bag for feeding.

3.8 Management procedures

Body weight and feed intake were recorded every week and survivability was recorded for each replication up to 28 days of age. The following management procedures were followed during the whole experimental period.

3.8.1 Litter management

Bedding material that was high absorbing was used as litter on floor. Clean, fresh and sun-dried rice husk was used as shallow litter to absorb moisture from fecal discharge of broiler chicken. The shallow litter was 5 cm (2 inch) in depth. About 200g calcium powder was mixed with rice husk in every pen as disinfectant. At the end of each week the litter was harrowed to prevent accumulation of toxic gases and to reduce moisture and parasitic infestation. At 3rd and 4th week of rearing period, dropping was cleaned from the surface level by removing a thin layer of litter and same amount new litter was placed in each pen.

3.8.2 Care of day-old chicks

Just after arrival of day-old chicks to the poultry house the initial weight of the chicks were recorded by a digital electronic balance and distributed them under the hover for brooding. The chicks were supplied glucose water with vitamin C to drink for the first 6 hours to overcome dehydration and transport stress. Subsequently small feed particles were supplied on the newspapers to start feeding for the first 24 hours.

3.8.3 Brooding of baby chicks

Electric lamp brooder was used to brood the chicks. Partitioning was done due to different experimental treatment. Each brooder had one hover and a round chick guard to protect chicks and four partitioning chambers. The brooding was adjusted on the behavior and comfortable of the chicks. Thereafter, healthy baby chicks were randomly distributed to the pen according to the design of the experiment. The recommended brooding temperature was 35-23°C from 1st to 4th weeks of age. Due to low environmental temperature at first week 200-watt electric bulb was hanged in every pen to maintain chicks body temperature. Moreover, at that time the wall polythene sheet spread over the net-wire to protect the chicks from cold and wind.

3.8.4 Room temperature and relative humidity

Daily room temperature (°C) and humidity were recorded with a thermometer and a wet and dry bulb thermometer respectively. Averages of room temperature and percent relative humidity for the experimental period were recorded and is presented in Table 4.

Week	Date	Temperature (°C)		Relative Humidity (%)	
		Avg. Maximum	Avg. Minimum	Avg. Maximum	Avg. Minimum
1^{st}	21.02.19 -27.02.19	31.72	25.18	61.20	45.00
2^{nd}	28.02.19 -06.03.19	28.34	17.79	78.00	54.57
3 rd	07.03.19-13.03.19	31.64	21.41	77.71	46.14
4^{th}	14.03.19-20.03.19	34.66	24.76	71.43	42.71

 Table 4. Temperature and relative humidity

3.8.5 Feeding and drinking

Crumble feed was used as starter (0–2 weeks), pellet feed was used as grower and finisher at 3-4 weeks and 4 weeks to last day of sell. *Ad libitum* feeding was allowed for rapid growth of broiler chicks up to the end of the 4th weeks. Live yeast and oligosaccharide were supplied by mixing with the feed at recommended dose of producer. Fresh clean drinking water was supplied *ad libitum*. Feed were supplied 3 times: morning, noon and night. Water was supplied two time daily: morning and evening. Left over feed and water were recorded to calculate actual intake of feed and water. Digital electric balance and measuring cylinder were used to take record of feed and water. Daily water consumption (ml) and weekly feed consumption (gm/bird) were calculated to find out weekly and total consumption of feed and water. Manual plastic feeder and drinker were used. All feeders and drinkers were washed and sun-dried before starting the trial. One plastic made round and one drinker were kept in the experimental pen. Feeder and drinker size were changed according to the age of the bird. Feeders were washed at the end of the weeks and drinkers once daily.

3.8.6 Lighting

At night there was provision of light in the broiler house to stimulate feed intake and rapid body growth. At night 4 energy lights were provided to ensure 24 hours' light for the first 2 weeks. Thereafter, 23 hours light and one-hour dark were scheduled up to marketable age. At night one-hour dark was provided in two times by half an hour.

3.8.7 Ventilation

The broiler shed was south facing and open sided. Due to wire net cross ventilation it was easy to remove polluted gases from the farm. Besides, on the basis of necessity ventilation was regulated by folding polythene screen.

3.8.8 Biosecurity measures

Biosecurity is the product of all actions undertaken by an entity to prevent introduction of disease agents into a specific area. To keep disease away from the broiler, farm the following vaccination, medication and sanitation program was undertaken.

3.8.9 Vaccination

The vaccines were collected from medicine shop (Ceva Company) and applied to the experimental birds according to the vaccination schedule. At day old a combined vaccine was used to the entire flock against Infectious Bronchitis (IB) and Newcastle Disease (ND). The flock was vaccinated against gumboro disease at 9th and booster dose at 17th days of age. The vaccination schedule is shown in Table 5.

 Table 5. Vaccination schedule

Age (day)	Name of disease	Name of vaccine	Route of vaccination
0	Infectious Bronchitis +	CEVAC BI L	One drop in eye
	Newcastle Disease (IB+ND)		
09	Gumboro (IBD)	CEVAC IBDL	Drinking water
17	Gumboro (IBD)	CEVAC IBDL	Drinking water

3.8.10 Medication

Broiler medication program is an important to keep disease free flock in commercial broiler farming. This process includes receiving day-old chick (DOC) and medication program in different days of bird's age. Vitamin-B complex, Vitamin-A, D₃, E were used against deficiency diseases. Electrolyte and vitamin-C also used to save the birds from heat stress. The medication program is presented in Table 6.

Medicine	Composition	Dose	Period
Ultravit B+C	Vitamin B-complex + vit C	1g/1L water	3-5 days (all groups)
Renavit AD ₃ E	Vitamin A, D & E	1 ml/5L water	3 -5 days (all groups)
Electromin Powder	Electrolytes	1g/2L water	4-5 days (all groups)
Revit-C	Vitamin-C Premix	1g/5L water	4-5 days (all groups)
Calplex	Ca, P and Vit-D	10 ml/100 bird	3-5 days (all groups)
COCCI-OFF (water soluble powder)	Anticoccidial	1g/ L water	5-7 days (all groups)

Table 6. Medication program

3.8.11 Sanitation

The single most important factor in keeping poultry healthy is maintaining good hygiene. Healthy parents and hygienic hatchery conditions contribute greatly to disease free chicks. Good hygiene standards reduce disease challenge. Farm sanitation does not just mean the choice of the right disinfectant. The key to farm sanitation is effective cleaning. Throughout the experimental period proper hygienic measures were maintained. Cleaning and washing of broiler shed and its premises were under a routine sanitation work. Flies and insects were controlled by spraying phenol and lysol to the surroundings of broiler shed. There was a provision of foot bath at the entry gate of the broiler shed to prevent any probable contamination of disease. Farm dress, shoes and hand gloves were used during the experimental period.

3.9 Recorded parameters

Weekly lives weight, weekly feed consumption and death of chicks to calculate mortality percent. FCR was calculated from final live weight and total feed consumption per bird in each replication.

3.10 Data collection

The experiment was carried out by collecting data from the five treatment. Feed intake (FI), body weight gain (BWG), feed conversion ratio (FCR), profit per bird, mortality

percentage, uniformity, benefit cost ratio of different experimental birds were calculated. Detail of each data collection procedure are given below:

3.10.1 Live weight

The initial day-old live weight and weekly live weight of each replication was kept to get final live weight record per bird.

3.10.2 Feed consumption

Daily feed consumption record of each replication was kept to get weekly and total feed consumption record per bird.

3.10.3 Mortality of chicks

Daily death record for each replication was counted up to 28 days of age to calculate mortality.

3.10.4 Flock uniformity

Uniformity is a measure of the variability of bird size in a flock. To determine the average weight and uniformity of flock, divided the house into three sections.150 birds were weighed individually to determine flock. It is important to weigh all birds within the catch pen, excluding culls. Of the 100 birds sampled, counted the number of birds 10% either side of the average body weight.

3.11 Calculations

3.11.1 Live weight gain (LWG)

The average body weight gain of each replication was calculated by deducting initial body weight from the final body weight of the birds.

Body weight gain = Final weight – Initial weight

3.11.2 Feed intake (FI)

Daily feed consumption record of each replication was kept to get weekly and total feed consumption record per bird. Total feed intake in a replication was divided by number of live birds in each replication to get average feed intake per bird.

Average FI (g/bird) = $\frac{\text{Feed intake in replication (g)}}{\text{No. of live birds in a replication}}$

3.11.3 Feed conversion ratio (FCR)

Total feed consumption per bird was divided by weight gain per bird

 $FCR = \frac{Feed intake (g)}{weight gain (g)}$

3.11.4 Flock uniformity

Uniformity can be determined using the following equation-

Uniformity = $\frac{\text{Standard deviation}}{\text{Average body weight}} \times 100$

3.12 Economic analysis

3.12.1 Cost record

The production cost was calculated involved in chicks, feed, vaccine and medication. Feed cost was calculated by the average amount of feed consumed in each treatment on phase basis. Litter cost was calculated with the required amount of rice husk bags multiplying rice divided by number of birds in each replication. Cost of live yeast and oligosaccharide was calculated with the required amount multiplying price divided by number of replication birds in each treatment groups. All expenses and income were calculated on the basis of market price at the time of experimental period.

3.12.2 Benefit cost ratio (BCR)

The capital expenditure, recurring expenditure and depreciation cost were considered to calculate total expenditure. The major expenditure included cost of chick, feed, litter, medicine, vaccine and labor and electricity charges. The common expenditure per bird was found out from the total expenditure of one batch. The consumption of feed was not same in different replications, so feed expenditure was calculated for every individual replication. Similarly, due to difference of live weight gain, the sale value of bird was calculated for every individual replication. The sale value of poultry manure and feed bags were also considered to compute income. Number of live birds in each replication considered here to calculate average value. Finally, treatment wise production cost and income was calculated. Net profit was found out by deducing the total expenditure from the total income according to replication under each treatment.

 $BCR = \frac{Total income}{Total cost of production}$

3.12.3 Profit per bird (PPB)

The major expenditure included cost of chick, feed, litter, medicine, vaccine, labor and electricity bill. The common expenditure per bird was found out from the total

expenditure of one batch. The consumption of feed was not same in different replications, so feed expenditure was calculated for every individual replication. The sale value of poultry manure and feed bags were also considered to compute income. Number of live birds in each replication considered here to calculate average value. Finally, treatment wise production cost and income was calculated. Profit per bird was found out by deducting the total expenditure from the total income according to replication under each treatment.

PPB = Total income per bird – Total expenditure per bird

3.13 Statistical analysis

The data was subjected to statistical analysis by applying one-way ANOVA using statistical package for social sciences (SPSS) version 16. Differences between means were tested using Duncan's multiple comparison test and significance was set at P<0.05.

Some photograph of chick management and experimental procedure are preented in plate 3-16 below:



Plate 3. Washing of floor with detergent





Plate 4. Cleaning of drinker





Plate 5. Preparation of chick brooder guard

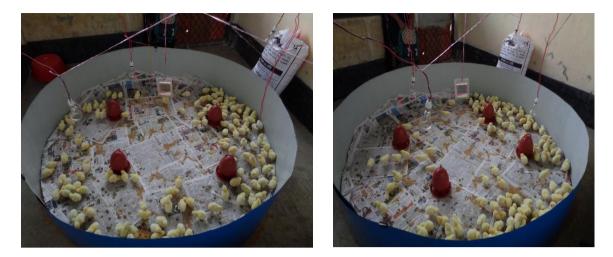


Plate 6. Arrival of day old chick (DOC)



Plate 7. Preparation of experimental room



Plate 8. Distribution of chick under different treatment

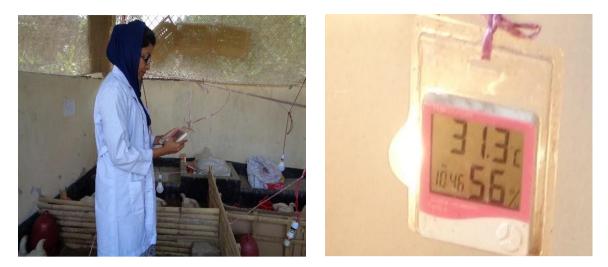


Plate 9. Measuring of temperature and humidity



Plate 10. During drinking of chicks



Plate 11. During feeding of chicks



Plate 12. Vaccination vial of IBD



Plate 13. Vaccination vial of ND and IB





Plate 14. Vaccination of chick



Plate 15. Medication (calplex)



Plate 16. Renasol AD₃E vitamin

CHAPTER-4

RESULTS AND DISCUSSION

CHAPTER 4 RESULTS AND DISCUSSION

4.1 Production performances of broiler chicken

Supplementation of live yeast and oligosaccharide to broiler diets improves growth performance in terms of feed consumption, body weight gain and feed conversion ratio (FCR). The chicks were randomly divided into five experimental treatment groups. The five groups were T_1 (1g yeast and 0.5g oligosaccharide/kg feed), T_2 (2g yeast and 0.5g oligosaccharide/kg feed), T_3 (1g yeast and 1g oligosaccharide/kg feed) and T_4 (2g yeast and 1g oligosaccharide/kg feed) and T_4 (2g yeast and 1g oligosaccharide/kg feed) and T_0 (control). The performance traits *viz*. final live weight, body weight gain, feed consumption, FCR, survivability, flock uniformity, were discussed here.

4.1.1 Final live weight

The relative final live weight (g) of broiler chickens in the different groups T_0 , T_1 , T_2 , T_3 and T_4 presented in (Table 7 & Figure 2) were 1469.43±18.29, 1759.53±16.86, 1596.23±30.06, 1765.90±4.89 and 1601.87±29.83 respectively. The highest result was found in T_3 (1765.90g±4.89) and lowest result was in T_0 (1469.43g±18.29) control and that was statistically significant (P<0.05). Results also demonstrated that the body weights also varied among the treatment groups having statistical significance (P<0.05) and all the treated groups had higher live weight than control group. The higher body weight in T_1 and T_3 group might be due to the synergistic effects of live yeast (*Saccharomyces cerevisiae*) and oligosaccharide.

These results are in agreement with those of previous researchers Mohamed *et al.* (2015) reported that live weight at different levels of yeast was not different (P>0.05) from either negative or positive controls. Nevertheless, overall body weight gains increased (P<0.05) in birds fed on 1% dietary yeast compared with the positive control during the entire period (0 to 6 weeks of age). Yang *et al.* (2007) found that supplementation of MOS to the basal diet improved the growth performance of birds compared to the negative control in the first three weeks but not in the last three weeks.

4.1.2 Weekly body weight gains (WBWG)

The mean body weight gains (g) at the 1st, 2nd, 3rd and 4th week of different treatment groups were significantly higher (P<0.05) than control. The mean body weight gains (g) of broiler chicks at 4th week in different groups were T_0 (542.60±11.10), T_1

(648.57±5.72), T₂ (592.40±18.25), T₃ (651.23±7.65) and T₄ (575.83±27.9) respectively. At the 4th week the highest result was found in T₃ (651.23g±7.65) and lowest result was in T₀ (542.60g±11.10) control group and that was statistically significant (P<0.05). The data of weekly body weight gains of broiler chicks presented in (Table 8 & Figure 3).

These results are in agreement with those of previous researchers Gao *et al.* (2008) reported that during the starter period (0-3 wk.), dietary treatments did not affect (P>0.05) broiler performance parameters, during the finisher (4-6 wk.) and overall (4-6 wk.) periods, supplemental yeast, significantly (P<0.05) affected growth performance. Yang *et al.* (2007) reported that 6% increase (P<0.05) in BWG were observed with birds in the high MOS group (2g/kg) compared to the negative control in the first three weeks.

4.1.3 Total feed consumption (FC)

Total feed consumption of different treated groups and control group have been presented in Table 7. T_0 (control) consumed higher amount of feed (2421.07g±25.99) and T_3 consumed lower amount of feed (2307.30g±3.66), whereas T_1 , T_2 , and T_4 consumed 2398.27g±3.72, 2380.17g±11.91 and 2377.77g±9.29 feed respectively. Result in total feed consumption demonstrated that treatment groups showed significant (P<0.05).

These results are in agreement with those of previous researchers Shareef and Al-Dabbagh (2009) reported that dietary treatment of 1.5% and 2% live yeast had significantly higher feed intake than others (P<0.05). Yang *et al.* (2007) reported that non-significant effect of MOS and/or AGP addition to diets on feed intake of young broilers. Spring (1999) reported that feed intake per bird from day-old to day 40 of age showed a significant (P<0.05) increase in the MOS treated group compared to control.

4.1.4 Weekly feed consumption (WFC)

The mean of weekly feed consumption of broiler chicks in different groups T_0 , T_1 , T_2 , T_3 , T_4 , showed in Table 9 & Figure 4. The result presented that feed consumption of the 1st weeks (starter phase) was significantly (P<0.05) higher in live yeast and oligosaccharide treated groups than control group. In 2nd week, the highest feed consumption was in T_1 group 458.33g±1.20 and the result showed that there was significant difference (P<0.05) among the treated group. In 3rd week, the highest feed consumption was in T_1 group (685.87g±2.26) and the lowest feed consumption was in T_3 group (622.03g±1.73) and the result showed that there was significant difference (P<0.05) among the treated group. In 4th week, the highest feed consumption was in T_0 group (1097.93g±1.39) and the lowest

feed consumption was in T_1 group (1016.67g±4.18) and the result showed that there was significant difference (P<0.05) among the treated group.

These results are in agreement with those of previous researchers Gao *et al.* (2008) reported that at the 4th-6th weeks 3% dietary yeast significantly (P<0.05) increased feed intake when compared to the positive control. Yang *et al.* (2007) reported that feed intake of birds numerically increased on the high MOS treatment (2g/kg) compared to the negative control in the first three weeks.

4.1.5 Feed conversion ratio (FCR)

The result of feed conversion ratio (FCR) of broilers under different treatment groups have been shown in Table 7. The lowest feed conversion ratio (FCR) 1.39 ± 0.02 and 1.33 ± 0.00 were significantly (P<0.05) found in T₁ and T₃ group supplemented 1g yeast with 0.5g oligosaccharide and 1g yeast with 1g oligosaccharide respectively than control birds (1.69 ± 0.03). However, feed conversion ratio (FCR) was significantly (P<0.05) higher in T₂ (1.53 ± 0.02) and T₄ (1.52 ± 0.03) groups compared to control.

These results are in agreement with those of previous researchers Mohamed *et al.* (2015) reported that a poorer feed conversion ratio was observed in birds fed 3% yeast and positive control compared with negative control. In contrast with this study, Onifade *et al.* (1999) reported the use of yeast cell in broiler diets improved feed conversion ratio. Shareef and Al-Dabbagh (2009) reported that dietary treatment of 1.5% and 2% live yeast had significantly higher in feed conversion ratio than others (P<0.05). Kamran (2013) observed improved feed conversion ratio on adding MOS. The enhancement in FCR might be due to an increased weight gain of the birds in the same group.

4.1.6 Weekly feed conversion ratio (Weekly FCR)

The mean weekly FCR of broiler chicks in different groups were presented in Table 10 and Figure 5. The FCR of 1^{st} weeks was significant (P<0.05) than control but there was no significant difference among the treated group. In 2^{nd} and 3^{rd} weeks FCR was significant (P<0.05) among the treated group compare to control. In 4^{th} week FCR was significantly lower (P<0.05) in T₁ and T₃ group than T₂, T₄ and control group.

These results are in agreement with those of previous researchers Shareef and Al-Dabbagh (2009) reported that dietary treatment of 1.5% and 2% live yeast at 3^{rd} weeks had significantly higher in feed conversion ratio than others (P<0.05). Yang *et al.* (2007)

reported that 2% decrease (P>0.05) in FCR were observed with birds in the high MOS group (2g/kg) compared to the negative control in the first three weeks.

4.1.7 Survivability

Survivability rate of broiler chickens treated with live yeast and oligosaccharide presented in Table 7. The result denoted that the survivability rate of the broilers in the treatment groups were higher than control group but did not differ significantly (P>0.05) with control group.

These results are in agreement with those of previous researchers Shareef and Al-Dabbagh (2009) reported that *Saccharomyces cerevisiae* is considered as one of the live microorganism's probiotic that, when administered at 1%, 1.5% and 2% through the digestive tract mortalities were not observed in all groups of experiment. Firon *et al.* (1983) reported that mannan-oligosaccharides are thought to block the attachment of pathogenic bacteria to the animal's intestine. It is also thought to stimulate the animal's immune system, thereby further increasing the rate of survivability.

4.1.8 Flock uniformity

Flock uniformity of broiler chicken were presented in table 11. The higher flock uniformity (73.33 \pm 3.33 %) was found in T₁ and T₄ group. The lower flock uniformity (63.33 \pm 3.33 %) was found in T₂ group. Flock uniformity of different treatment groups were statistically insignificant (P>0.05).

These results are in agreement with those of previous researcher of Susan, (2009) reported that on day 18, when the trial began with *saccharomyces cerevisiae*, there was no significant difference (P>0.05) in the weights and the variance was very small, indicating that all the treatments started with similar bird weights. On days 28 and 36, the weight difference was also insignificant (P>0.05). The difference in weight gain from both 18-28 days and 28-36 days was also insignificant (P>0.05), with p-values of 0.8913 and 0.1200. Yang *et al.* (2007) reported that diet treated MOS group birds were in a very health condition and flock uniformity was high around 90%.

Treatment	Final Live weight (g/bird)	Average BWG (g/bird)	Total FC (g/bird)	Final FCR	Survivability (%)
T ₀	1469.43 ^c ±18.29	1427.40 ^c ±18.26	2421.07 ^a ±25.99	$1.69^{a} \pm 0.03$	98.33±1.67
T_1	1759.53 ^a ±16.86	$1717.53^{a} \pm 16.85$	2398.27 ^a ±3.72	$1.39^{c} \pm 0.02$	100.00 ± 0.00
T_2	1596.23 ^b ±30.06	1554.23 ^b ±30.06	2380.17 ^a ±11.91	$1.53^{b} \pm 0.02$	100.00 ± 0.00
T ₃	$1765.90^{a} \pm 4.89$	1723.90 ^a ±4.89	$2307.30^{b} \pm 3.66$	$1.33^{c} \pm 0.00$	100.00 ± 0.00
T_4	1601.87 ^b ±29.83	1559.87 ^b ±29.83	$2377.77^{a} \pm 9.29$	$1.52^{b} \pm 0.03$	100.00 ± 0.00
Mean ± SE	1638.59±31.04	1596.57±31.05	2376.91±11.42	1.49±0.03	99.67±0.33

Table 7. Effects of live yeast and oligosaccharide on production performances of broiler chicken

Here, $T_0 =$ (Control), $T_1 =$ (1g yeast and 0.5g oligosaccharide), $T_2 =$ (2g yeast and 0.5g oligosaccharide), $T_3 =$ (1g yeast and 1g oligosaccharide), $T_{4=}$ (2g yeast and 1g oligosaccharide). Values are Mean \pm SE (n=15), one-way ANOVA (SPSS, Duncan method).

- \checkmark Mean with different superscripts are significantly different (P<0.05)
- \checkmark Mean within same superscripts don't differ (P>0.05) significantly
- ✓ SE= Standard Error

Treatment	1 st Week BWG	2 nd Week BWG	3 rd Week BWG	4 th Week BWG
T ₀	165.53 ^b ±0.60	326.90 ^b ±6.45	392.40 ^c ±13.81	542.60 ^b ±11.10
T_1	$201.73^{a} \pm 4.76$	$388.00^{a} \pm 1.34$	$479.23^{a}\pm8.78$	$648.57^{a}\pm5.72$
T_2	$199.50^{a}\pm0.29$	$330.17^{b} \pm 27.00$	$432.17^{b} \pm 12.32$	$592.40^{b} \pm 18.25$
T ₃	$200.43^{a} \pm 1.55$	$387.00^{a} \pm 1.40$	$485.23^{a}\pm3.02$	$651.23^{a} \pm 7.65$
T_4	$204.20^{a}\pm1.46$	$339.70^{ab} \pm 18.11$	$440.13^{b} \pm 3.00$	$575.83^{b} \pm 27.9$
Mean ± SE	194.28±3.97	354.35 ± 9.22	445.83±9.72	602.13±12.85

 Table 8. Effects of live yeast and oligosaccharide on body weight gain (BWG)
 (g/bird) of broiler chickens in different weeks

Here, $T_0 = (Control)$, $T_1 = (1g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_2 = (2g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_3 = (1g \text{ yeast and } 1g \text{ oligosaccharide})$, $T_4 = (2g \text{ yeast and } 1g \text{ oligosaccharide})$. Values are Mean \pm SE (n=15), one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ SE= Standard Error

Treatment	1 st Week FC	2 nd Week FC	3 rd Week FC	4 th Week FC
T ₀	$224.10^{b} \pm 6.46$	$448.30^{ab} \pm 4.20$	650.73 ^{bc} ±18.76	1097.93 ^a ±1.39
T_1	$237.40^{a} \pm 0.30$	$458.33^{a} \pm 1.20$	$685.87^{a} \pm 2.26$	$1016.67^{d} \pm 4.18$
T_2	$237.10^{a} \pm 1.51$	437.33 ^{bc} ±9.13	$668.93^{ab} \pm 3.88$	$1036.80^{b} \pm 0.89$
T ₃	236.70 ^a ±0.35	$428.53^{c}\pm0.58$	$622.03^{c} \pm 1.73$	$1020.03^{d} \pm 2.03$
T_4	$237.40^{a} \pm 1.11$	$442.00^{bc} \pm 2.10$	$670.90^{ab} \pm 7.42$	$1027.46^{c} \pm 1.16$
Mean ± SE	234.54±1.80	442.90±3.21	659.69±6.82	1039.78±8.03

Table 9. Effects of live yeast and oligosaccharide on feed consumption (g/bird) of broiler chickens in different weeks

Here, $T_0 = (Control)$, $T_1 = (1g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_2 = (2g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_3 = (1g \text{ yeast and } 1g \text{ oligosaccharide})$, $T_4 = (2g \text{ yeast and } 1g \text{ oligosaccharide})$. Values are Mean \pm SE (n=15), one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ SE= Standard Error

Treatment	1 st Week FCR	2 nd Week FCR	3 rd Week FCR	4 th Week FCR
T ₀	$1.35^{a} \pm 0.04$	$1.37^{a} \pm 0.01$	$1.66^{a} \pm 0.08$	2.03 ^a ±0.04
T_1	$1.18^{b} \pm 0.03$	$1.18b^{c} \pm 0.00$	$1.43^{b} \pm 0.02$	$1.57^{c} \pm 0.02$
T_2	$1.19^{b} \pm 0.01$	$1.34^{ab}\pm0.08$	$1.55^{ab} \pm 0.05$	$1.75^{b} \pm 0.05$
T ₃	$1.18^{b} \pm 0.01$	$1.11^{c}\pm0.01$	$1.28^{c}\pm0.01$	$1.57^{c}\pm0.02$
T_4	$1.16^{b} \pm 0.01$	$1.31^{ab} \pm 0.07$	$1.52^{ab} \pm 0.03$	$1.79^{b} \pm 0.09$
Mean ± SE	1.21 ± 0.02	1.26±0.03	1.49 ± 0.04	1.74 ± 0.05

Table 10. Effects of live yeast and oligosaccharide on feed conversion ratio (FCR) of broiler chickens in different weeks

Here, $T_0 = (Control)$, $T_1 = (1g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_2 = (2g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_3 = (1g \text{ yeast and } 1g \text{ oligosaccharide})$, $T_4 = (2g \text{ yeast and } 1g \text{ oligosaccharide})$. Values are Mean \pm SE (n=15), one-way ANOVA (SPSS, Duncan method).

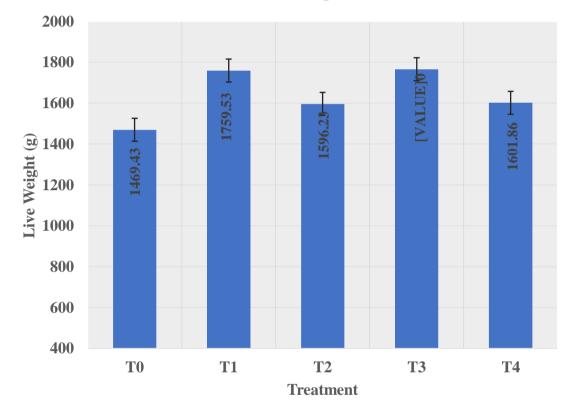
- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ SE= Standard Error

Treatment	Uniformity	Level of significance
T ₀	70.00±10.58	NS
T_1	73.33±3.33	NS
\mathbf{T}_2	63.33±3.33	NS
T ₃	$70.00{\pm}10.0$	NS
T_4	73.33±3.33	NS
Mean ± SE	70.00±2.82	NS

Table 11. Flock uniformity of broiler chickens under different treatment

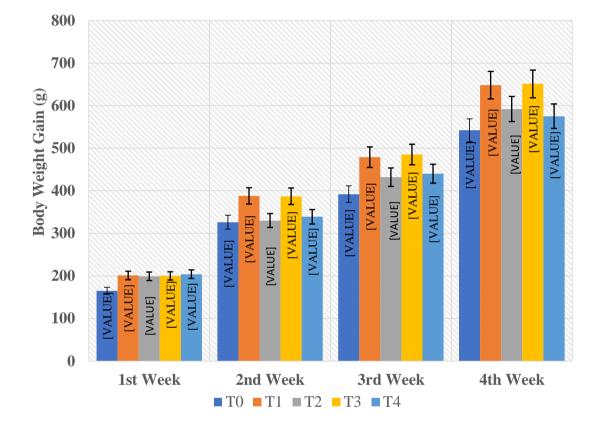
Here, $T_0 = (Control)$, $T_1 = (1g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_2 = (2g \text{ yeast and } 0.5g \text{ oligosaccharide})$, $T_3 = (1g \text{ yeast and } 1g \text{ oligosaccharide})$, $T_4 = (2g \text{ yeast and } 1g \text{ oligosaccharide})$. Values are Mean \pm SE (n=15), one-way ANOVA (SPSS, Duncan method). NS = Non-significance

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ SE= Standard Error



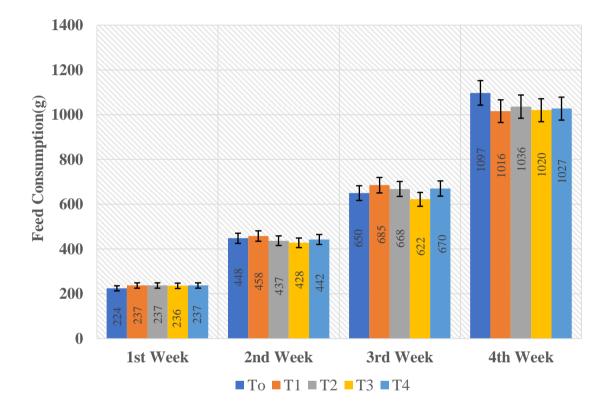
Average Live Weight Under Different Treatment Group

Figure 2. Effects of live yeast and oligosaccharide on live weight of broiler chickens



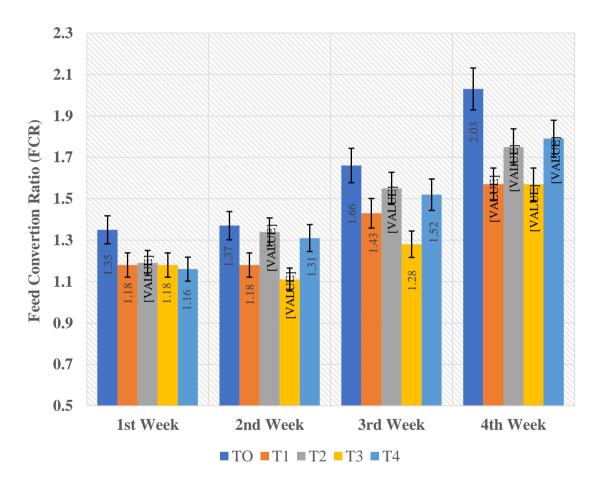
BWG of Broiler Chickens at Different Weeks

Figure 3. Effects of live yeast and oligosaccharide on body weight gain of broiler chickens in different weeks



Weekly Feed Consumption of Broiler Chickens

Figure 4. Effects of live yeast and oligosaccharide on feed consumption of broiler chickens in different weeks



FCR of Broiler Chickens at Different Weeks

Figure 5. Effects of live yeast and oligosaccharide on FCR of broiler chickens at different weeks

CHAPTER-5

SUMMARY, CONCLUSION & RECOMMENDATION

CHAPTER 5

SUMMARY, CONCLUSION & RECOMMENDATION

The present experiment was conducted at the Sher-e-Bangla Agricultural University (SAU), Dhaka Poultry Farm for a period of 28 days using live yeast (*Saccharomyces cerevisiae*) and oligosaccharide. The experiment was pe rformed by applying different concentration of live yeast and oligosaccharide mixing with commercial broiler feed. The specific objectives of this experiment were under taken to determine the synergistic effect of live yeast and oligosaccharide in broiler performance, to investigate the synergistic effects, to recommend the inclusion level, to determine the flock uniformity. A total of 150 day-old Cobb-500 broiler chicks were purchased from Kazi hatchery, Gazipur, Dhaka. The experimental broilers were distributed randomly to 4 treatments and a control group with three replications having 10 broilers per replication of experimented group. The experiment was conducted for 4 weeks and the treatment of various groups consisted of different inclusion level such as group T_1 was 1g yeast with 0.5g oligosaccharide, group T_2 was 2g yeast with 0.5g oligosaccharide, group T_0 was no yeast and oligosaccharide i.e. control group.

The synergistic effects of live yeast and oligosaccharide were measured. The parameters such as the final live weight, weekly body weight gain, total feed consumption, weekly feed consumption, feed conversion ratio (FCR), weekly feed conversion ratio, survivability, and flock uniformity. A statistically significant difference (P<0.05) was noted on body weight, feed consumption, BWG and FCR value of the birds treated with live yeast and oligosaccharide. The final live weight of T_1 , T_2 , T_3 and T_4 group treated with live yeast and oligosaccharide was significantly higher (P<0.05) than control group (T_0) . The mean body weight gains (g) at the 1st, 2nd, 3rd and 4th week of different treatment groups were significantly higher (P<0.05) than control group. The T₃ group consumed lower amount of feed compare to control. The feed consumption of T₃, T₄, T₁, T₂, and T₀ are ascending in number. At 1st week the feed consumption was significantly higher (P<0.05) in live yeast and oligosaccharide treated group than control and at the 4^{th} week the feed consumption was significantly higher (P<0.05) in T_0 group than live yeast and oligosaccharide treated group. The FCR was better in all live yeast and oligosaccharide treated groups compared to the control group but significant (P<0.05) difference with the T_3 and T_0 groups. The FCR of 1^{st} , 2^{nd} and 3^{rd} weeks was significant (P<0.05) than control

and in 2^{nd} and 3^{rd} weeks FCR was significant (P<0.05) among the treated group. In 4^{th} week FCR was significantly lower (P<0.05) in T₁ and T₃ group than T₂, T₄ and control group. The survivability rate of the broiler chickens in the treatment groups were higher than control group but statistically insignificant (P>0.05). The higher flock uniformity was found in T₁ and T₄ group but the flock uniformity was statistically insignificant (P>0.05).

Analyzing the above research findings, this study recommended that the use of live yeast and oligosaccharide was best for better production performance. One gram yeast and One gram oligosaccharide powder was more effective than others treatment. So, live yeast and oligosaccharide can be used as an alternative to AGP in broiler ration. The study therefore recommends for hematological parameters on birds immunity and conducting field trial on commercial poultry farm to fix up inclusion level of live yeast and oligosaccharide. Hence, live yeast and oligosaccharide could be decidedly used in broiler rearing for better performance and higher economical return.

REFERENCES

- Awad, W.A., Ghareeb, K., Nitsch, S., Pasteiner, S., Abdel-Raheem, S. and Bohm, J. (2008). Effects of purified lignin and mannan oligosaccharides on intestinal integrity and microbial population in ceca and litter of broiler chickens. J. of Poult. Sci. 86: 1070-1078.
- Barnet, J.A., Payne, R.W., and Yarrow, D. (2000). Yeasts: Characteristics and Identification. 3rd ed. Cambridge: Cambridge University Press.
- Bezkorovainy, A. (2001). Prebioitcs: Determinates of survival and growth in the gut. *American J. of nutr.* **65**(2): 399-404.
- Chacher, M.F.A (2017). Use of mannan oligosaccharide in broiler diets: an overview of underlying mechanism. *J. of World Poult. Sci.* **73**: 97-114.
- Churchill, R. and Mohan, B. (2000). Effect of supplementation of broiler rations with live yeast culture. 29(1-2): 23 27.
- Duk, L.B. and Zhang, Ai-Wu., (2004). Effects of saccharomyces cerevisiae supplementation on growth performance and meat quality in broiler chickens. XXII Worlds Poultry Congress, Istanbul, Turkey.
- Erener, G., Ocak, N., Ozturk, E., Cankaya, S. and Ozkanca, R. (2009). The effects of olive leaf extract on performance, some blood parameters and caeca micro flora of broilers. J. Appl. Bacteriol. 77(2): 412-420.
- FAO/WHO, (2001). Report of a joint FAO/WHO expert consultation on evaluation of health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria. Cordoba, Argentina (October 1-4, 2001). Available at: http://www.fao.org/3/a-a0512e.pdf.
- Firon, N. and Ofek, S.N. (1983). Carbohydrate specifity of the surface lectins of *Esherichia coli*, *Klebsiella pneuomoniae* and *Salmonella typhimurium*. *Carbohydr. Res.* **120**: 235-249.
- Gao, J., Zhang, H.J., Yu, S.H., Wu, S.G., Yoon, I., Quigley, J., Gao, Y.P. and Qi, G.H. (2008). Effects of yeast culture in broiler diets on performance and immunomodulatory functions. *Poult. Sci.* 87: 1377–1384.
- Gibson, G.R. and Roberfroid, M.B. (1995). Dietary modulation of the colonic microbiota. *Food microbial*. **11**: 491-498.
- Grashorn, M.A. (2010). Use of phytobiotics in broiler nutrition an alternative to infeed antibiotics. *J. Anim. Feed Sci.* **19**: 338-347.

- Hatemink, R. (1995). Non-digestible oligosaccharides: Healthy food for colon. Proceedings Symposium Wageningen. pp: 177-178.
- Hidaka, H., Eida, T., Takizawa, T., Tokunaga, T. and Tashiro, Y. (1986). Effects of fructooligosaccharides on intestinal flora and human health. *Bifidobacteria Microflora* 5: 37–50.
- Hooge, D. (2004). Meta-analysis of broiler chicken pen trials evaluating dietary mannan oligosaccharide. *Int. J. Poult. Sci.* **3**: 163-174.
- Hooge, D.M., Sims, M.D., Sefton, A.E., Connolly, A. and Spring, P. (2003). Effect of Dietary Mannan oligosaccharide, with or without bacitracin or virginiamycin, on live performance of broiler chickens at relatively high stocking density on new litter. J. Appl. Poult. Res. 12: 461-467.
- Huque, M.F., Alosh, M. and Bhore, R. (2011). Addressing multiplicity issues of a composite endpoint and its components in clinical trials. J. of Biopharm. Stats. 21: 610–634.
- Hyginus, I., and Chukwu. (2003). Dietary Saccharomyces cervisiae and mannan oligosaccharide reduced the deleterious effects of heat stress on white leghorn laying hens. Cooperative Agricultural Research Center, Prairie View A & M University, Prairie View, Texas 77446.
- Islam, K.B.M.S., Shiraj-Um-Mahmuda, S. and Hazzaz-Bin-Kabir, M. (2016). Antibiotic usage patterns in selected broilers farms of Bangladesh and their public health implications. *J. of Public Health in Developing Countries* **2**(3): 276-284.
- Kamran, Z. (2013). Performance of broiler chickens fed mannan oligosaccharides as alternatives to antibiotics from one to twenty-two days of age. J. of Ani. and Plant Sci. 23: 1482-1485.
- Kelesedis (2012). Efficacy and safety of the probiotic Saccharomyces boulardii for the prevention and therapy of gastrointestinal disorders. Therapeutic Advanced Gastroenterology. 5(2): 111–125.
- Kocher, A., Canolly, A., Zawadzki, J. and Gallet, D. (2004). The challenge of finding alternatives to antibiotic growth promoters. International Society for Animal Hygiene-Saint Malo pp. 227-229.
- Kokje, R.P. (1999). Effect of feeding probiotics on growth performance in commercial broilers. M.V.Sc. Thesis Submitted to Gujarat Agricultural University, Anand.
- Leeson, S. and Summers, J.D. (2008). Use of probiotic in broiler. Commercial Poultry Nutrition, 3rd Edition. Nottingham University Press, England.

- Liukkonen-Anttila, J. (2001). Nutritional and genetic adaptation of galliforms birds: Implications for hand rearing's and resticking. Acanic Dissertation, Faculty of Science, University of Oulu, Oulu Yilopisto, Finland.
- Loddi, M.M.L., Nakaghi, S.O., Edens, F.M., Hannas, M.I., Moraes, V.M.B. and Arikri, J. (2002). Mannan oligosaccharide and organic acid on intestinal morphology integrity of broiler evaluated by Scanning electron microscopy in proc. 11th European. Poult. Sci. Comf. Brenen. Germany Sept. 6: 109-121.
- Mcdonnell, D.P., Pike, J.W., Drutz, D.J., Butt, T.R., and O' Malle, B.W. (1989). Reconstitution of the vitamin d-responsive osteocalcin transcription unit in *saccharomyces cerevisiae*, *Mol. Cell. Biol.* **9**: 3517–3523.
- Mohamed, E.A., Talha, E.A., Mojahid, A., Abdlhag, and Dafaalla, E.M. (2015). Effect of dietary yeast (*Saccharomyces cerevisiae*) supplementation on Performance, carcass characteristics and some metabolic responses of broilers. *J. Ani. and Vet. Sci.* 3:(5-1): 5-10
- Montes, A.J. and Pugh, D.G. (1993). The use of probiotics in food-Animal practice. *Vet. Med.* **88**: 282-289.
- Newman, K. (1994). Mannan-oligosaccharides: Natural polymers with significant impact on the gastrointestinal microflora and the immune system. *Biotechnology in the Feed Industry*. PP. 167–174.
- Nicholasville, K.Y. (1997). Bio-Mos product specifications. Bio-Mos Poultry Dossier. Alltech Inc.
- Nilson, A., Peralta. J.M.F. and Miazzo, R.D. (2004). Use of brewer's yeast (S. cerevisiae) to replace part of the vitamin mineral premix in finisher broiler diets. XXII Worlds Poultry Congress, Istanbul, Turkey.
- Onifade, A.A. (1998). Proposing fortification of foods with yeast for optimal nutrition value and salubrious effects. *Nutr. and Food Sci.*. **4**: 223–226.
- Onifade, A.A., Odunsi, A.A., Olorede, B.R. and Muma, E. (1999). Comparison of the supplemental effects of *Saccharomyces cerevisiae* and antibiotics in low-protein and high-fiber diets fed to broiler chicken. *Arch. Anim. Nutr.* 52: 29–39.
- Oyofo, B.A., Deloach, J.R., Corrier, D.E., Norman, J.O., Ziprin, R.L. and Mollenhauer, H.H (1989). Prevention of Salmonella typhimurium colonization of broilers with D-mannose. *Poult. Sci.* **68**: 1357-1360.
- Panda, A.K., Reddy, M.R., Rama, R.S.V., Raju., M.V.L.N. and Paraharaj, N.K. (2009). Growth, carcass characteristics, immune componence and response to *Escherichia*

coli of broiler fed diets with various level of probiotic. *Archive fur Geflugelkunde*. **64**: 152 – 156.

- Parks, C.W., Grimes, J.L., Ferket, P.R. and Fairchild, A.S. (2001). The effects of mannan oligosaccharide, bambermycin and virginiamycin on performance of large white male turkeys' toms. *Poult. Sci.* 80: 718-723.
- Patterson, J.A. and Burkholder, K.M. (2003). Application of prebiotics and probiotics in poultry production. *Poult. Sci.* **82**: 627-631.
- Pelicano, E.R.L., Souza, P.A., Souza, H.B.A., Figueiredo, D.F., Boiago, M.M., Carvalho, S.R. and Bordon, V.F. (2005) Intestinal mucosa development in broiler chickens fed natural growth promoters. *Rev Bras Cienc Avic*. 7: 221–1229.
- Raha, S.K. (2007). Poultry Industry in Bangladesh: Is it growing? Bangladesh J. of Agril. Econ. 2: 93-101.
- Reed, G. and Nagodawithana, T.W. (1999). Yeast technology (2nd Edn.), Van Nostrand Reinhold New York.
- Rodriguez, A., Cuesta, A., Ortuno, J., Esteban, M.A. and Meseguer, J. (2003). Immunostimulant properties of a cell wall-modified whole *Saccharomyces cerevisiae* strain administered by diet to sea bream (*Sparus aurata L.*). *Veterinary Immunology and Immunopathology*. **96**: 183-192.
- Rosen, G.D. (2007). Holo-analysis of the efficacy of Bio-Mos in broiler nutrition. *Br. Poult. Sci.* **48**: 21-26.
- Rutz, F., Anciuti, M.A., Rech, J.L., Goncalves, F.M., Delgado, A.D. Rosa, E.R. Zauk, N., Ribeiro, C.L.G., Silva, R.R. and Dallmann, P.R. (2006). Desempenhoe caracteristica de carcaca de frangos de corte recebendo extrato de levedura na dieta. Cienc. Anim. Bras. 7: 349-355.
- Saleh, A.A., Hayashi, K. and Ohtsuka, A. (2013). Synergistic effect of feeding *Aspergillus Awamori* and *Saccharomyces cerevisiae* on growth performance in broiler chickens; promotion of protein metabolism and modification of fatty acid profile in the muscle. *J Poult Sci.* 50: 242–250.
- Santin, E., Maiorka, A., Macari, M., Grecco, M., Okada, T.M. and Myasaka, A.M. (2001). Performance and intestinal mucosa development of broiler chickens fed diets containing *Saccharomyces cerevisiae* cell wall. *J. Appl. Poult.* Res. 10: 236– 24.

- Santin, E., Paulillo, A.P., Mariorka, A., Fischer, A.V., Silva and Alessi, A.C. (2003). Evaluation of efficacy of *Saccharomyces cerevisiae* cell wall to ameliorate the toxic effects of aflatoxin in broilers. *Int. J. of Poult. Sci.* 2(5): 341 – 344.
- Shareef, A.M. and Al-Dabbagh, A.S.A. (2009). Effect of probiotic (*Saccharomyces cerevisiae*) on performance of broiler chicks. *Iraqi J. of Vet. Sci.* 23(1): 23-29.
- Spais, A.B., Giannenas, I. and Christaki, A.E. (2003): Effect of the feed supplement Bio-Mos, a mannan-oligosaccharide, on the performance of broiler chickens. J. of Hellenic vet. made. sci. 54(2): 2585-3724
- Spring, P. (1999). Modes of action of dietary mannan oligosaccharide as a growth enhancer. *Zootech. Int.* 22: 34.
- Spring, P. (1999b). Mannan oligosaccharides as an alternative to antibiotic use in Europe. *Zootech. Int.* **22**: 38-41.
- Spring, P. and Privulescu, M. (1998). Mannan oligosaccharide: its logical role as a natural feed additive for piglets. Nottingham University Press, Nottingham, UK.
- Spring, P., Wenk, C., Dawson, K.A. and Newman, K.E. (2000). The effects of dietary mannan oligosaccharides on cecal parameters and the concentrations of enteric bacteria in the ceca of Salmonella-challenged broiler chicks. *Poult. Sci.* 79: 205-211.
- Stanley, U.G., OJO, R., Woldesenbat, S. and Hutchinson, D.H. (1993). The use of Saccharomyces cerevisiae to suppress the effects of aflatoxicosis in broiler chicks. *Poult. Sci.* 72: 1867-1872.
- Stanley, V.G., Gray, C., Daley, M., Krueger, W.F. and Sefton, A.E. (2004). An alternative to antibiotic-based drugs in feed for enhancing performance of broilers grown on Eimeria spp.-infected litter. *Poult Sci.* 83: 39–44.
- Strickling, J.A., Harmon, D.L., Dawson, K.A., Gross, K.L. (2000). Evaluation of oligosaccharide addition to dog diets: influences on nutrient digestion and microbial populations. An. Feed Sci. and Tech. 86: 205-219.
- Susan, W. (2009a). The role of probiotics in the poultry industry. *Int. J. of Mol. Sci.* **10**(8): 3531–3546.
- Susan, W. (2009b). Cell interactions and regulation of cell type in the yeast *Saccharomyces cerevisiae*. *Annu. Rev. Microbiol.* **37**: 623-660.

- Toma, Raipulus and Kalnina (2005). Effect of Probiotic Yeast on Genotoxicity: Does Probiotic Yeast Act as Antigenotoxin? *Food Techn. and Biotech.* 43 (3). Pgs. 301–305.
- Tortuero, F. (1973). Influence of the implantation of *Lactobacillus acidophilus* in chicks on the growth, feed conversion, malabsorption of fats syndrome and intestinal flora. *Poult. Sci.* **52**: 197–203.
- Yan, G.L., Yan, Y.M., Guo, J.M., Yuan, D. Liu., Zhang, B.K. (2011). Sodium alginate oligosaccharides from brown algae inhibit *Salmonella enteritidis* colonization in broiler chickens *Poult. Sci.* **90** (7): 1441-1448.
- Yang, Y., Iji, P.A. and Choct, M. (2007). Effects of different dietary levels of mannan oligosaccharide on growth performance and gut development of broiler chickens. *J. of appli. poult. sci.* 16: 280-288.
- Zhang, A.W., Lee, B.D., Lee, S.K., Lee, K.W., An, G.H., Song, K.B. and Lee, C.H. (2005). Effects of yeast (*Saccharomyces cerevisiae*) cell components on growth performance, meat quality, and ileal mucosa development of broiler chicks. *Poult. Sci.* 84: 1015-1021.

APPENDICES

Treatment	Replication	1 st Week FC	2 nd Week FC	3 rd Week FC	4 th Week FC	Total FC
T ₀	R ₁	218.10	442.60	618.60	1095.40	2374.70
	\mathbf{R}_2	237.00	445.80	683.60	1098.20	2464.60
	R ₃	217.20	456.50	650.00	1100.20	2423.90
T_1	R ₁	237.00	460.00	682.20	1025.00	2404.20
	\mathbf{R}_2	238.00	456.00	685.40	1012.00	2391.40
	R ₃	237.20	459.00	690.00	1013.00	2399.20
T_2	R ₁	234.30	420.00	670.70	1035.20	2360.20
	\mathbf{R}_2	239.50	441.00	661.50	1036.90	2378.90
	R ₃	237.50	451.00	674.60	1038.30	2401.40
T ₃	R ₁	237.10	429.50	619.00	1019.20	2304.80
	\mathbf{R}_2	236.00	427.50	622.10	1017.00	2302.60
	R ₃	237.00	428.60	625.00	1023.90	2314.50
T_4	R ₁	239.60	446.20	683.80	1025.20	2394.80
	\mathbf{R}_2	236.50	440.00	658.10	1028.20	2362.80
	R ₃	236.10	439.80	670.80	1029.00	2375.70

Appendix 1. Feed consumption (g/bird) of 1st, 2nd, 3rd and 4th week under different treatment groups

Treatment	Replication	1 st Week	2 nd Week	3 rd Week	4 th Week	Total BWG
To	R ₁	166.6	316.5	413.3	564.5	1460.8
	\mathbf{R}_2	165.5	325.5	397.6	534.9	1423.5
	R ₃	164.5	338.7	366.3	528.4	1397.9
T_1	\mathbf{R}_1	193.5	390.5	461.7	640.7	1686.4
	\mathbf{R}_2	201.7	385.9	489.0	645.3	1721.9
	R ₃	210.0	387.6	487.0	659.7	1744.3
T_2	\mathbf{R}_1	199.0	299.5	451.0	556.6	1506.1
	\mathbf{R}_2	199.5	307.0	436.5	604.1	1547.1
	R ₃	200.0	384.0	409.0	616.5	1609.5
T ₃	R ₁	200.0	386.6	489.5	639.7	1715.8
	\mathbf{R}_2	198.0	389.6	479.4	665.7	1732.7
	R ₃	203.3	384.8	486.8	648.3	1723.2
T_4	\mathbf{R}_1	207.0	343.5	435.0	520.5	1506.0
	\mathbf{R}_2	202.1	306.6	445.4	610.5	1564.6
	R ₃	203.5	369.0	440.0	596.5	1609.0

Appendix 2. Body weight gain (BWG) (g/bird) of 1st, 2nd, 3rd and 4th week under different treatments

Treatment	Replication	1 st Week	2 nd Week	3 rd Week	4 th Week
T ₀	R ₁	1.31	1.40	1.50	1.94
	R ₂	1.43	1.37	1.72	2.05
	R ₃	1.32	1.35	1.77	2.08
T ₁	R ₁	1.22	1.18	1.48	1.60
	\mathbf{R}_2	1.18	1.18	1.40	1.57
	R ₃	1.13	1.18	1.42	1.54
T ₂	R ₁	1.18	1.40	1.49	1.86
	R ₂	1.20	1.44	1.52	1.72
	R ₃	1.19	1.17	1.65	1.68
T ₃	\mathbf{R}_1	1.19	1.11	1.26	1.59
	\mathbf{R}_2	1.19	1.10	1.30	1.53
	R ₃	1.17	1.11	1.28	1.58
T_4	R ₁	1.16	1.30	1.57	1.97
	R ₂	1.17	1.44	1.48	1.68
	R ₃	1.16	1.19	1.52	1.73

Appendix 3. Feed conversion ratio (FCR) of birds under different treatments

Treatment	Replication	Final Live weight (g/bird)	Total FC (g/bird)	Total BWG (g/bird)	Final FCR	Survivability (%)
T ₀	R ₁	1502.90	2374.70	1460.8	1.63	100
	\mathbf{R}_2	1465.50	2464.60	1423.5	1.73	100
	R ₃	1439.90	2423.90	1397.9	1.73	95
T_1	\mathbf{R}_{1}	1728.40	2404.20	1686.4	1.43	100
	\mathbf{R}_2	1763.90	2391.40	1721.9	1.38	100
	R ₃	1786.30	2399.20	1744.3	1.38	100
T_2	\mathbf{R}_{1}	1548.10	2360.20	1506.1	1.57	100
	\mathbf{R}_2	1589.10	2378.90	1547.1	1.54	100
	R ₃	1651.50	2401.40	1609.5	1.49	100
T ₃	\mathbf{R}_1	1757.80	2304.80	1715.8	1.34	100
	\mathbf{R}_2	1774.70	2302.60	1732.7	1.33	100
	\mathbf{R}_3	1765.20	2314.50	1723.2	1.34	100
T_4	\mathbf{R}_{1}	1548.00	2394.80	1506.0	1.59	100
	\mathbf{R}_2	1606.60	2362.80	1564.6	1.51	100
	R ₃	1651.00	2375.70	1609.0	1.48	100

Appendix 4. Production performance of broiler chicken under different treatments

Treatment	Replication	Uniformity (%)	Average Uniformity (%)
T ₁	R ₁	70	
	\mathbf{R}_2	80	73
	R ₃	70	
T_2	\mathbf{R}_{1}	60	
	\mathbf{R}_2	60	66
	\mathbf{R}_3	70	
T ₃	R ₁	50	
	\mathbf{R}_2	80	70
	R ₃	80	
T_4	R ₁	70	
	\mathbf{R}_2	70	73
	R ₃	80	
T ₀	R ₁	90	
	\mathbf{R}_2	66	81
	R ₃	54	

Appendix 5. Flock uniformity of broiler chickens under different treatment