



# The Effects of Probiotic, Prebiotic and Synbiotic on Growth Performance and Immune Responses of Broiler Chicks Challenged with Heat Stress by Multiattribute Decision-Making Method

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Received: 30 January 2021, Revised: 09 February 2021, Accepted: 23 March 2021

## ABSTRACT

**Background:** Heat stress (HS) has negative effects on economic parameters of the poultry industry. Different strategies such as nutritional additives are used to alleviate negative effects of HS. The use of multiattribute decision-making (MADM) could help to select the best additive for alleviating the effects of the HS. Thus, the present study was conducted to investigate the effects of probiotic, prebiotic and synbiotic on growth performance and immune responses of broiler chicks affected by HS via MADM method.

**Methods:** Two-hundred and forty broiler chicks were randomly assigned into 4 treatments with 6 replications and 10 broiler chicks per replication. The birds were treated with probiotic, prebiotic, and synbiotic for 42 days. A group was considered as control and received only basal diet. Stress condition was induced from 21 to 42 days of age. Growth performance and humoral immunity were assessed, then calculated and analyzed by MADM method.

**Results:** The results showed that dietary supplementation of probiotic, synbiotic, prebiotic and control had coefficients of 0.762, 0.702, 0.581 and 0.00, respectively. Dietary supplementation of probiotic, prebiotic and synbiotic had better effects compared with control group.

**Conclusions:** Therefore, dietary supplementation of probiotic may have better efficiency compared with other additives based on the MADM. The use of probiotics can be suggested for improving growth and immunity under the HS condition in the poultry industry.

**Keywords:** Probiotic, Prebiotic, Symbiotic, Heat stress, Multiattribute decision-making

## 1. Introduction

Heat stress (HS) has negative effects on economic parameters in the poultry industry [1]. It is defined as a status in that animals cannot maintain their body temperature against thermal environment [2]. HS decreases growth

performance and defaults immune responses in broiler chicks [1, 3, 4]. HS shows its negative effects on immune responses by the decrease in level of total circulating antibodies [5]. It also has adverse effects on the intestinal mucosa and microbiota of broiler chickens [6].

Different strategies are used for alleviation negative effects of HS on economic parameters of broiler chicks such as dietary supplementation of additives.

Probiotics are live microbial feed supplements that have beneficial effects for their host by improving microbial balance in host [7-9]. Probiotics not only improve intestinal microbial balance, but they also promote the growth and activity of some bacteria in gastrointestinal system [10]. Studies have reported positive effects of probiotics on economic parameters in the poultry industry [11, 12].

Prebiotics increase population of health-promoting bacteria in the intestinal tract [10]. Prebiotics are non-digestible feed ingredients that could have beneficial effects for their hosts by improving the growth and activity of beneficial bacteria in intestinal system, such as glucose, fructose, galactose, and mannose [13]. Studies have reported that a combination of probiotic and prebiotics (synbiotics) have synergistic effects on improving immune responses and growth performance by the modulation in microbial system [14].

In sum, prebiotic, probiotic and synbiotic may have beneficial effect on growth performance and immunity of broiler chicks under HS condition. Comparative effects of different levels of dietary supplements in animal production could help decision makers [15]. Multiple Attribute Decision Making (MADM) is a method for selecting the best treatment, but it is not commonly used in the animal science. The use of the MADM may help to select the best the treatment. This study, for first time, evaluated the effects of probiotic, prebiotic and synbiotic on growth performance and immune responses of broiler chicks affected by the HS through MADM method.

## 2. Materials and methods

### 2.1. Materials

We prepared probiotic from IROST (Tehran, Iran), containing *B. subtilis*  $4 \times 10^8$  CFU/g in per mL. The prebiotic product contained hydrolyzed yeast and yeast cell wall. Synbiotic was a mixture of 50% probiotic and 50% prebiotic.

### 2.2. Housing

Two-hundred and forty commercial one-day old broiler chicks (Ross 308) of male sex were prepared. The current study was conducted in a farm in Sarpole-Zahab town (Kermanshah-Iran). All the efforts were conducted to minimize pain in the animals and the study was conducted based on Ethical Committee of Islamic Azad University, Qaemshahr Branch (No. 14062). The broiler chicks were reared in pens for a three phases period of starter, grower and finisher. The diets were formulated based on Ross 308 broiler management manual [16] and analyzed for proximate composition by AOAC methods [17]. A lighting program of 23 h light and 1 h darkness was used. The broiler chicks had unrestricted access to water and food. Thermal program was adjusted based on catalogue on 21 first days. Heat stress program was conducted from 21 days to 42 days, as reported by Akbari and Toriki [18].

### 2.3. Growth performance

Feed intake, body weight and feed conversion ratio (FCR) were calculated as growth performance parameters.

### 2.4. Immune responses

Immune responses were investigated by sheep red blood cell (SRBC) solution based on previous studies [19] and the samples were investigated for antibody titer, IgG and IgM.

### 2.5. Microbiological isolation and enumeration

To isolate lactic acid bacteria, the ileal samples were collected from 2 broiler chicks per replication and investigated as reported by Jazi et al. [20].

### 2.6. The MADM method

The MADM method was calculated as reported by Meimandipour et al. [15]. Summary, decision matrix based on fuzzy set membership values for the production criteria was calculated as follows;

$$n_{ij} = \frac{a_{ij}}{\sqrt{\sum_{i=1}^m a_{ij}^2}}$$

Normalized weights for selecting the treatments were calculated and the probability distribution (Pij) was calculated as follows;

$$P_{ij} = \frac{a_{ij}}{\sum_{i=1}^m a_{ij}}$$

The entropy value (Ei) was calculated, as follows;

$$E_j = -k \sum_{i=1}^m [p_{ij} \ln p_{ij}]$$

The amount of uncertainty (di) was calculated, as follows:

$$d_i = 1 - E_j$$

The weights were calculated, as follows;

$$W_j = \frac{d_j}{\sum_{j=1}^n d_j}$$

Setting the decision set (D) was calculated by multiplying the probability distribution (Pij) matrix with weighting matrix based on multiple matrices. In TOPSIS, all criteria are based on weight of significance and weighting is based on the entropy method.

### 3. Results

The results for mean of parameters are shown in Table 1. FCR was considered as negative index, but other parameters were considered as positive parameter. As the results show, synbiotic had highest mean for feed intake, body weight and lactobacillus, while probiotic had the highest mean for immune responses.

**Table 1.** The results for mean of parameters

Treatments	Feed intake (g)	Body weight (g)	FCR	IgG (log <sub>2</sub> )	IgM (log <sub>2</sub> )	Lactobacillus (CFU/g)
Control	3610.21	1930.21	1.88	2.12	1.89	7.12
Probiotic	3921.15	2150.12	1.82	2.39	2.22	7.51
Prebiotic	3858.41	2130.51	1.81	2.32	2.05	7.43
Synbiotic	3978.12	2210.21	1.79	2.26	2.05	8.02
Positive/negative	+	+	-	+	+	+

Decision matrix based on fuzzy set membership values for the production criteria are shown in Table 2. In this part, the data, feed intake for instance, were grouped:

$$[3610.21+3921.15+3858.41+3978.12]$$

The data were then calculated as follows;

A=

$$[(3610.21)^2+(3921.15)^2+(3858.41)^2+(3978.12)^2]$$

The calculation was continued for radical, as follows;

$$\sqrt{A}$$

The data were divided on  $\sqrt{A}$  as follows;

$$3610.21/\sqrt{A}$$

The data were calculated for all the parameters and values.

**Table 2.** Decision matrix based on fuzzy set membership values for the production criteria

Treatments	Feed intake (g)	Body weight (g)	FCR	IgG (log <sub>2</sub> )	IgM (log <sub>2</sub> )	Lactobacillus (CFU/g)
Control	0.469525	0.457854	0.515068	0.465934	0.459854	0.47309
Probiotic	0.509964	0.510017	0.49863	0.525275	0.540146	0.499003
Prebiotic	0.501805	0.505366	0.49589	0.50989	0.498783	0.493688
Synbiotic	0.517373	0.524271	0.490411	0.496703	0.498783	0.53289

The data were corrected based on weights. The weights were reported as 0.15 for FCR and 0.1 for other treatments, as follows; Feed intake in control group: 0.469525\*0.1=0.0469525 FCR in prebiotic group= 0.49589\*0.15=0.74384

**Table 3.** Correction of the data based on fuzzy set membership values for the production criteria

Treatments	Feed intake (g)	Body weight (g)	FCR	IgG (log <sub>2</sub> )	IgM (log <sub>2</sub> )	Lactobacillus (CFU/g)
Control	0.0469525	0.0457854	0.07726	0.0465934	0.0459854	0.047309
Probiotic	0.0509964	0.0510017	0.074795	0.0525275	0.0540146	0.0499003
Prebiotic	0.0501805	0.0505366	0.074384	0.050989	0.0498783	0.0493688
Synbiotic	0.0517373	0.0524271	0.073562	0.0496703	0.0498783	0.053289

Table 4 shows selection of negative and positive values for the production. In animal science, more consumption of feed, higher body weight, lower FCR, higher antibody titer immune responses and lactobacillus are desirable. We selected lowest value for FCR (0.073562) as positive ideal and highest values for other groups, 0.0769525 (feed intake), 0.0457854 (body weight), etc. Highest value for FCR (0.07726) and lowest values for other parameters in per column were considered as negative ideal.

**Table 4.** Selection of negative and positive values for the production criteria

Treatments	Feed intake (g)	Body weight (g)	FCR	IgG (log <sub>2</sub> )	IgM (log <sub>2</sub> )	Lactobacillus (CFU/g)
-	0.0469525	0.0457854	0.07726	0.0465934	0.0459854	0.047309
+	0.0517373	0.0524271	0.073562	0.0525275	0.0540146	0.053289

The amounts of uncertainty for the production criteria for positive and negative ideal were calculated (Table 5), as follows; Negative ideal in prebiotic group; Negative ideal:  $A = \sqrt{(0.0501805 - 0.0469525)^2 + (0.0505366 - 0.0457854)^2 + (0.074384 - 0.07726)^2 + (0.050989 - 0.0465934)^2 + (0.0498783 - 0.0459854)^2 + (0.0493688 - 0.047309)^2}$  The calculation was continued for radical, as follows;

**Table 5.** The amount of uncertainty for the production criteria for positive and negative ideal

Treatments	-	+
Control	0	0.0147
Probiotic	0.0125	.0039
Prebiotic	0.0089	0.0064
Synbiotic	0.0118	0.005

Normalized weights for selecting the best the treatment are shown in Table 7. The data were calculated as follows;

Synbiotic:  $\text{Negative ideal} / (\text{Positive ideal} + \text{negative ideal})$   
 $0.0118 / (0.0118 + 0.005) = 0.702380952$ .

The results showed that probiotic, synbiotic, prebiotic and control had coefficients of 0.762, 0.702, 0.581 and 0.00, respectively. Thus, probiotic, synbiotic and prebiotic had first to third orders.

#### 4. Discussion

The present study investigated the effects of probiotic, prebiotic and synbiotic on growth performance and immune responses of broiler chicks affected by HS by the MADM method. The results showed that dietary supplementation of synbiotic had higher values for means for some parameters, but dietary supplementation of probiotic had higher efficiency compared with synbiotic group. In the MADM analysis, all the parameters are to be investigated and decided based on several factors. The results show that adding probiotic into diet has highest efficiency for decreasing negative effects of the HS on growth performance and immune responses. The results show that dietary inclusion of probiotic could have highest coefficient (0.762) and it had a difference with synbiotic (0.702).

HS has negative effects on immune responses and growth performance by decreasing feed intake and insufficient absorption nutrients for production of antibody and also lower population for

lactic acid bacteria [11]. The results showed that the broiler chicks in control group consumed lesser feed and had lower growth and immunity. They also had lower population for lactic acid bacteria. Dietary supplementation of probiotic had the highest coefficient and desirable for growth, immunity and lactic acid bacteria compared with control group. The results of the effects of probiotic and synbiotics on growth performance are similar to those reported by previous studies [11, 12, 20-24]. The positive effects of probiotics on growth performance of broiler chicks might be attributed to its positive effects for improving immunity performance (23). Under stress condition, animals consume more food for improving immunity. Seemingly, supplementation of probiotic and synbiotic improve immunity and the improvement of immunity decrease food consumption for immune responses. Other reason for improvement of growth performance by probiotic and synbiotic might be attributed to their effects on intestinal microbiota. Probiotics and synbiotics increase beneficial bacteria population, as shown for lactic acid bacteria. Beneficial bacteria increase nutrient retention and digestive enzymes required for improving growth performance [25].

Similar results for the effects of probiotics and synbiotics on immunity responses have been previously reported [20, 22, 26, 27]. The efficiency of probiotics and synbiotics for improving immune responses is related to their

effects for improving intestinal epithelial cells [28] and preventing pathogen colonization under stress condition [29].

The broiler chicks in synbiotic had positive ideal values for some parameters, but it had lower values for immune responses compared to probiotic group. The results show that probiotics are more efficient under HS condition, but dietary supplementation of prebiotic decreased its efficiency for immune response compared with probiotic group. Studies have reported the efficiency of synbiotics for improving immune responses and growth performance by the modulation in microbial system [14], as observed in the current study.

## 5. Conclusions

The MADM analysis is a suitable tool to investigate data on several parameters. In the current study, the MADM investigated different parameters and the results showed that probiotic is more efficient under HS condition for improving growth performance and immune responses. Dietary supplementation of probiotics is suggested for improving growth and immunity under HS condition. We also recommend experts to apply MADM for selecting the best additives in the poultry industry.

## Conflict of interest

None.

## Consent for publications

Both authors read and approved manuscript.

## Availability of data and material

All the data are given.

## Authors' contributions

Both authors designed the study and first author conducted and analyzed the data.

## Funding

All the costs were supported by first author.

## Ethics approval and consent to participate

All the efforts were conducted to minimize pain in the animals and the study was conducted based on Ethical Committee of Islamic Azad University, Qaemshahr Branch (No.14062).

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**How to cite this article:** Ebrahim Babakhani, Reza Rostamian. The Effects of Probiotic, Prebiotic and Synbiotic on Growth Performance and Immune Responses of Broiler Chicks Challenged with Heat Stress By Multiattribute Decision-Making Method. *International Journal of Advanced Biological and Biomedical Research*, 2021, 9(2), 181-189. Link: <http://www.ijabbr.com/article/243168.html>