Effects of isolated soy protein and broken rice in corn-soy pre-starter diet on performance, intestinal microflora, and gut morphology in broiler chickens

Ali Nabizadeh,* Abolghasem Golian,†,[1] Ahmad Hassanabadi,† and Saeed Zerehdaran†

*Department of Animal Science, Ferdowsi University of Mashhad – International Campus, PO Box 9176764955, Mashhad, Iran; and †The Excellence Centre for Animal Science, Department of Animal Science, Ferdowsi University of Mashhad, PO Box 91775-1163, Mashhad, Iran

Primary Audience: Nutritionists, Researchers

SUMMARY

A trial was conducted to evaluate the effect of type of protein and energy sources in pre-starter diet on performance, intestinal microflora and gut morphology in broiler chickens. Pre-starter diets were a 4 × 4 factorial design with 4 levels of isolated soy protein (ISP; 0, 1.5, 3.0, and 4.5%) and 4 levels of broken rice (0, 6, 12, and 18%). All pre-starter diets were formulated to be isocaloric and isonitrogenous. A common commercial grower and finisher diet was fed to all birds from 11 to 24 and 25 to 42 d of age, respectively. Birds fed pre-starter diet contained 4.5% and 18% or 3% and 6% and/or 1.5% and 18% ISP and broken rice, respectively had higher ($P < 0.05$) body weight gain (BWG) at 42 d of age compared to those fed control or other diets. Chickens fed pre-starter diet contained 4.5% ISP and 18% broken rice had lower ($P < 0.05$) feed conversion ratio (FCR) as compared to those fed control (1.602 vs. 1.653) or other diets during 1 to 42 d of age. *Lactobacillus* counts in ileum was reduced ($P < 0.05$) by 14% when birds fed pre-starter diet contained 4.5% ISP and 18% broken rice compared to control ones at 10 d of age. *Bifidobacteria* counts in ileum and *Escherichia coli* counts in ceca were reduced ($P < 0.05$) when pre-starter diet contained 4.5% ISP compared to control ones at 10 d of age. The inclusion of broken rice in diet did not have a significant effect on *Escherichia coli* and *Bifidobacteria* counts in ileum or *Bifidobacteria*, *Escherichia coli*, and *Lactobacillus* counts in ceca of 10 d old broiler chickens. The villus height in duodenum, jejunum and ilium in birds fed diet containing 4.5% ISP and 18% broken rice were increased ($P < 0.05$) by 6% at 5 and 4% at 10 d of age. It is concluded that, the inclusion of ISP and broken rice in pre-starter diet may have some beneficial effects on gut morphology, and performance of broiler chickens.

Key words: pre-starter, broken rice, soy protein, broiler chicken performance

2018 J. Appl. Poult. Res. 27:133–144
http://dx.doi.org/10.3382/japr/pfx025

[1]Corresponding author: golian-a@um.ac.ir

DESCRIPTION OF PROBLEM

Digestive tract of newly hatched broiler chicks in terms of physiological and morphological is not fully developed [1]. Intestinal villus height rapidly increase during the wk 1 of life [2], pancreatic and biliary secretion may be insufficient in wk 1 after hatch especially for protein digestion [3]. Because of undeveloped digestive tract and insufficient digestive enzyme secretion, feed is less digested in the pre-starter period. On the other hand, maturity and development of some body systems such as thermoregulation and immune system are closely correlated with nutrient absorption from digestive tract. In addition, due to positive correlation between 7-d live weight and final weight of broiler chickens [4], economic efficiency can be improved through feeding of highly digestible ingredients [5, 6]. Non-starch polysaccharides (**NSPs**) are significant parts of soybean meal (**SBM**) and corn that are commonly used as the main dietary source of protein and energy for broiler chicks. Total NSPs and soluble NSPs are 192 and 27 mg/g in SBM [7] and 76.3 and 27 mg/g in corn grain [8], respectively. Although, NSPs content of corn is lower than soybean meal, since corn is the main source of energy and it approximately constitutes 50 to 70% of the diets [9], thus the total amount of NSPs provided by corn is noticeable. Soluble NSPs could prevent the enzymatic digestion due to an increase in digesta viscosity, and thus reduce protein and amino acid digestibility [10]. Furthermore, soluble NSPs can exhibit anti-nutritive activities, leading to changes in gut physiology, microflora, and health of birds [11].

Isolated soy protein (**ISP**) has significantly higher nutritional value than SBM and is characterized by a high protein (90%), low trypsin inhibitor content (<1 mg/kg), and zero or negligible oligosaccharides and antigenic factors [12]. During the protein isolation process the non-protein components, including soluble and insoluble NSPs, are removed [13].

Starch is an important energy source in poultry diet, and type of starch and its digestion rate are important factors affecting birds performance. A positive correlation between starch digestion rate and broiler chicken's performance was reported [20]. Rice starch granules are smaller (3 to 8 μm vs. 2 to 30 μm) than corn [14]. Small granules are generally digested more rapidly than larger granules [15]. In addition, rice starch has less amylose than corn starch (200 vs. 250 g/kg) [16] and interestingly increased amylose content is negatively related to the digestibility of starch [17–19].

This experiment was conducted to study the inclusion rate of ISP and broken rice in corn-soy pre-starter diet on performance, intestinal microflora, and gut morphology of broiler chickens.

MATERIALS AND METHODS

The experimental protocol was approved by the Animal Welfare Committee, at the Ferdowsi University of Mashhad (FUM), Iran.

Birds' Husbandry and Diets

A total of 1,440 mixed sex, day-old Ross 308 broiler chicks were obtained from a local commercial hatchery, weighed (average initial body weight was 45 ± 0.8 g), divided into groups of 15 and randomly assigned to one of the 96 floor pens. Each independent pen (120 × 100 × 70 cm, L × W × H), was equipped with 2 nipple drinkers and a hanging plastic feeder for manual feed distribution. The pen's concrete floors were bedded with clean wood shavings. House temperature and lighting programs were similar to those described previously [21]. All birds were vaccinated against viral diseases such as, Newcastle Disease (**ND**) and Infectious Bursal Disease (**IBD**), as scheduled by the veterinary official. The 16 pre-starter diets were carried out in a 4 × 4 factorial arrangement with 4 levels of isolated soy protein (0, 1.5, 3.0, and 4.5%) and 4 levels of broken rice (0, 6, 12, and 18%). Each pre-starter diet was randomly fed to 6 replicate groups of 15 chicks each from 1 to 10 d of age. Thereafter, all birds were fed with a mash form common commercial grower (10 to 24 d) and finisher (24 to 42 d) diets (Table 1) based on Ross broiler nutrient specifications (2014).

Performance Parameters

Each replicate bird was weighed after arrival at the farm and on d 10, 24, and 42, using a digital electronic balance with 1 g accuracy. Feed

Table 1. Ingredients and nutrient composition of the pre-starter diets[1] fed from 1 to 10 d of age.

	Isolated soy protein[2] (ISP)%			
	0	1.5	3	4.5
Corn grain 8.5%	56.64	56.64	56.64	56.64
Broken rice[3]	–	–	–	–
Soybean meal 46%	27.81	25.00	22.16	19.34
ISP 90%	–	1.50	3.00	4.50
Soybean oil	0.18	0.36	0.55	0.74
Corn gluten meal	8.91	8.41	7.94	7.44
Wheat bran 15%	1.00	2.58	4.17	5.79
Casein 80%	1.00	1.00	1.00	1.00
Dicalcium phosphate	1.70	1.70	1.70	1.69
Calcium carbonate	1.39	1.40	1.41	1.42
Sodium chloride	3.90	3.90	3.90	3.80
Lysine-HCl	3.00	3.00	3.00	3.00
DL-methionine	0.18	0.21	0.22	0.23
Threonine	–	0.01	0.02	0.03
Vit. + min. premix[4]	0.50	0.50	0.50	0.50
Calculated nutrients (% *unless stated*)				
ME (kcal/kg)	3,000	3,000	3,000	3,000
CP (analyzed)	22.87	22.96	22.85	22.97
Lysine	1.29	1.29	1.29	1.29
Met + cys	0.95	0.95	0.95	0.95
Threonine	0.90	0.90	0.90	0.90
Calcium	0.96	0.96	0.96	0.96
Available P	0.48	0.48	0.48	0.48

[1]Similar commercial corn-soy grower and finisher diets fed to all birds from 10 to 24 and 24 to 42 d of age, respectively.

[2]It was provided from Shangdong Yuxin Bio-Tech Co., Qingdao, China.

[3]Four diets were formulated for each level of ISP by the addition of 0, 6.0, 12.0 and 18.0 per cent of broken rice and adjustment of all other ingredients with the exception of sodium chloride and vit-mineral premix to obtain similar nutrients in all diets.

[4]Each kg of diet contained: IU: vit. A 9,000, vit. D3 2,000, vit. E 18; mg: vit. K3 2, vit. B1 1.8, vit. B2 6.6, vit. B3 10, vit. B5 30, vit. B6 3, vit. B9 1, vit. B12 0.015, vit. H2 0.1, choline choloride 500; Mn 100, Fe 50, Zn 100, Cu 10, Mg 3.5, Se 0.2.

consumption was determined from the difference between supplied and residual feed in each replicate pen at the beginning and end of each period, respectively. The dead birds in each pen during each period were weighed and was used to calculate the corrected feed conversion ratio (**FCR**). The FCR was calculated by dividing total feed consumption by body weight gain (**BWG**) of birds for every pen during each period.

Samples Collection and Measurements

One chick from each replicate close to the average pen weight was selected to evaluate intestinal microflora and gut morphology at 5 and 10 d of age. Chicks were weighed, sacrificed, dissected, and the ileal and/or cecal (mixture of both cecum) contents were immediately collected, mixed, and placed in a microtube and stored at –70 °C until microbial assay. Since *Lactobacillus* and *Bifidobacteria* are the most important beneficial bacteria, and *E. coli* is the main pathogenic microorganism, that constitute a large part of the intestinal microflora in broiler chickens, these bacteria were studied in this experiment. Microbial populations were determined by serial dilution of ileal and/or cecal samples in anaerobic diluents before inoculation onto Petri dishes of sterile agar. *Lactobacilli* were grown on Rogosa SL agar, *Bifidobacteria* on BIM-25 agar, and E. coli on EMB agar. Plates for *Lactobacillus* and/or *Bifidobacteria* were incubated anaerobically for 48 h at 37 °C, but for *Escherichia coli* were incubated aerobically for 24 h at 37 °C. The number of colonies was then counted to determine the colony-forming units (**CFU**) per gram of fresh ileal and/or cecal contents, which were expressed in common logarithms.

Subsequently, the small intestine was divided into 3 segments: duodenum (from gizzard to pancreo-biliary ducts), jejunum (from pancreo-biliary ducts to Meckel's diverticulum), and ileum (from Meckel's diverticulum to the ileocecal junction). The empty weight and length of the fresh segments were recorded. About 1 cm in length from the midpoint of every segment was cut off, gently washed with physiological saline solution to remove any adherent of intestinal contents, and placed in 10% buffered formalin tube for fixation and further histological study. The buffered formalin solution was changed 2 times to refresh the tissue. Tissue samples dehydrated with increasing concentrations (70, 80, 95, and 100%) of ethanol, cleared with xylene, and placed into paraffin embedding wax. Tissue sections (2 μm) were cut by microtome [22], floated onto slides, and stained with hematoxylin (Gill no. 2, Sigma, St. Louis, MO) and eosin (Sigma). Villus height and crypt depth were measured by using a light microscope with a digital camera. Twelve images from 3 sections of each tissue sample were taken and 24 villus heights and crypt depths were measured by imaging software. Villus height was measured from the top of the villus to the top of the submucosa. Crypt depth was measured from the base upwards to the region of transition between the crypt and villus [23].

Statistical Analysis

Data were analyzed in a completely randomized design with factorial arrangement, using the GLM procedure of SAS [24]. Means were compared by Duncan's multiple-range test. Means difference was considered significant with a probability of equal or less than 5% ($P \leq 0.05$). Data were also subjected to orthogonal polynomial analysis for linear and quadratic trends, using SAS software.

RESULTS AND DISCUSSION

Growth Performance

The main effects of ISP and broken rice inclusion in pre-starter diet and their interaction on broiler chicken's performance are shown in Table 2. Chicks fed pre-starter diet contained 1.5% ISP had higher ($P < 0.0001$) BWG comparing to those fed control or diets containing 3 and 4.5% ISP at 10, 24, and/or 42 d of age. The BWG of chickens linearly increased with an increase of broken rice in pre-starter diet at 24 and/or 42 d of age. Birds fed pre-starter diet that contained 4.5% and 18%, 3% and 6%, and/or 1.5% and 18% ISP and broken rice, respectively had higher ($P < 0.05$) BWG compared with control and other treatments at 42 d of age (Table 3). The inclusion of ISP in pre-starter diet improved ($P < 0.05$) FCR during 1 to 10 and 1 to 42 d of age. The FCR was decreased ($P < 0.05$) with an increase of the broken rice in pre-starter diet. Inclusion of broken rice in the pre-starter diet improved ($P < 0.0001$) FCR during all periods. Chickens fed pre-starter diet contained 4.5% ISP and 18% broken rice had lower ($P < 0.05$) FCR as compared to those fed control (1.602 vs. 1.653) and/or other diets during 1 to 42 d of age.

The little information in the literature regarding the effect of ISP on performance of broiler chickens has conflicting results. Longo et al. [25] reported that starter diet containing 5.35% ISP did not affect ($P > 0.05$) BWG and FCR at 21 d of age, whereas Ebling et al. [26] found that diet containing 6% ISP decreased FI during 1 to 21 d of age, which negatively affected broiler growth performance ($P < 0.01$) at 7 and 21 d of age. In contrast with these findings, the study of Vander Eijk [27] showed that substitution of SBM with ISP significantly improved feed utilization. It has been reported that 5% processed soy protein improved BWG and feed efficiency of 7-day-old chicks, and improved ($P < 0.05$) FCR was reported for young turkeys fed a diet containing ISP [28]. The current study showed that BWG and FCR improved significantly when dietary SBM was replaced with ISP in pre-starter diet of broiler chickens. Soybean meal is known to contain anti-nutritional compounds, such as soy antigens and lectins [29] and trypsin inhibitors which have been shown to be detrimental to bird's performance [30]. Such anti-nutritional factors present in SBM, may affect small intestine characteristics and decrease digestive enzymes activities and consequently reduce broiler performance [31]. These factors not only reduce the nutritional value of SBM but reduce the digestion of nutrients from other ingredients. Processed soy products including soy protein concentrate (**SPC**) and ISP contain lower amount of oligosaccharides and antigenic substances. Therefore, these products have higher nutritive values than SBM [12]. Ebling et al. [26] showed that the coefficients of total tract retention of dry matter and GE of the pre-starter diets containing 6% ISP were higher ($P < 0.01$) than those fed control diet. Van Kempen et al. [32] reported that soybean oligosaccharides may decrease energy utilization. It is also shown that substitution of SBM with ISP significantly improved feed utilization as well as BWG of chickens [28]. The use of broken rice in pre-starter diet in our study improved FI, BWG, and FCR in broiler chickens, which are in agreement with the previous reports [26, 33]. Rice grain has less crude fiber and NSPs than corn (0.8 vs. 8.64%). Insoluble NSPs decrease the passage time of digesta through the gastrointestinal tract and consequently reduce nutrient digestibility. Negative

Table 2. Effect of pre-starter diet containing different levels of isolated soy protein (ISP) and broken rice (BR) on broiler chickens' performance.[1]

ISP (%) × BR (%)		Weight gain (g/chick)			Feed conversion ratio (g/g)		
		1–10 d	1–24 d	1–42 d	1–10 d	1–24 d	1–42 d
0	0	210^{i}	892^{h}	2,261^{i}	0.825^{a}	1.158^{a}	1.653^{a}
0	6	230$^{e–g}$	914^{g}	2,287^{g,h}	0.775$^{c–e}$	1.142^{b}	1.638^{b}
0	12	229^{f,g}	923^{f}	2,305^{f}	0.785^{c,d}	1.132^{b,c}	1.630^{c,d}
0	18	234$^{c–f}$	932^{c,d}	2,325^{d,e}	0.785^{c,d}	1.127^{c,d}	1.623^{d,e}
1.5	0	229^{g}	923^{f}	2,291^{g}	0.773^{d,e}	1.115$^{d–f}$	1.618^{e,f}
1.5	6	234$^{c–f}$	927$^{d–f}$	2,330^{c,d}	0.785^{c,d}	1.125$^{c–e}$	1.615^{f,g}
1.5	12	235$^{c–e}$	940^{b}	2,339^{b}	0.768^{d,e}	1.105^{f,g}	1.615^{f,g}
1.5	18	256^{a}	960^{a}	2,347^{a}	0.730^{g}	1.095^{g}	1.618^{e,f}
3.0	0	234$^{c–e}$	922^{f}	2,304^{f}	0.772^{d,e}	1.128^{c}	1.612^{f,g}
3.0	6	231$^{d–g}$	921^{f,g}	2,320^{a}	0.780$^{c–e}$	1.132^{b,c}	1.608^{g,h}
3.0	12	236^{c,d}	926$^{d–f}$	2,322^{e}	0.755^{e,f}	1.132^{b,c}	1.627^{c,d}
3.0	18	236^{c}	931$^{c–e}$	2,337^{b,c}	0.777$^{c–e}$	1.127^{c,d}	1.618^{e,f}
4.5	0	221^{h}	914^{g}	2,283^{h}	0.818^{a,b}	1.142^{b}	1.633^{b,c}
4.5	6	223^{h}	920^{f,g}	2,318^{e}	0.800^{b,c}	1.137^{b,c}	1.627^{c,d}
4.5	12	248^{b}	935^{b,c}	2,330^{c,d}	0.742^{f,g}	1.113^{e,f}	1.610^{g}
4.5	18	236^{c}	925^{e,f}	2,352^{a}	0.763$^{d–f}$	1.125$^{c–e}$	1.602^{h}
SEM[2]		1.65	2.28	2.39	0.01	0.004	0.03
Main Effect Means[3]							
ISP (%)							
0	–	271^{d}	915^{c}	2,294^{c}	0.79^{a}	1.14^{a}	1.64^{a}
1.5	–	284^{a}	938^{a}	2,327^{a}	0.76^{c}	1.11^{c}	1.62^{b}
3.0	–	279^{b}	925^{b}	2,321^{b}	0.77^{b,c}	1.13^{b}	1.62^{b}
4.5	–	277^{c}	924^{b}	2,321^{b}	0.78^{b}	1.13^{b}	1.62^{b}
	BR (%)						
–	0	268^{d}	913^{d}	2,285^{d}	0.80^{a}	1.14^{a}	1.63^{a}
–	6	274^{c}	920^{c}	2,314^{c}	0.79^{b}	1.13^{a}	1.62^{b}
–	12	282^{b}	931^{b}	2,324^{b}	0.76^{c}	1.12^{b}	1.62^{b}
–	18	286^{a}	937^{a}	2,340^{a}	0.76^{c}	1.12^{b}	1.61^{c}
SEM		0.83	1.14	1.19	0.004	0.002	0.001
P-value							
ISP		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
BR		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
ISP × BR		0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Orthogonal polynomials							
ISP							
Linear		0.0003	0.03	<0.0001	0.12	0.18	<0.0001
Quadratic		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
BR							
Linear		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Quadratic		0.22	0.50	<0.0001	0.10	0.01	0.40

[1] All birds fed a similar corn-soy commercial grower and finisher diets during 10 to 24 and 24 to 42 d of age, respectively.

[a–h] Means in the same column for every effect with uncommon superscript are significantly different at $P \leq 0.05$.

[2] Standard error mean.

[3] Means represent 15 birds per pen, 24 pens per diet.

effects of soluble NSP are associated with the viscous nature of these polysaccharides. Soluble NSPs increase gut viscosity [34], which decreases the rate of diffusion of substrates and digestive enzymes at the mucosal surface [35]. Soluble NSPs interact with the glycocalyx of the intestinal brush border and thicken the rate-limiting unstirred water layer of the mucosa, which reduce the efficiency of nutrient absorption through the intestinal wall [36]. It has been reported that FCR may be improved with an increase in dietary starch, because starch is the

Table 3. Effect of pre-starter diet containing different levels of isolated soy protein (ISP) and broken rice (BR) on microflora counts in ileal and cecal digesta of 10-day-old chicks (log10 cfu/g content).

ISP (%) × BR (%)		Ileum			Cecum		
		E. coli	*L.*[1]	*B.*[2]	*E. coli*	*L.*	*B.*
0	0	5.0	7.4^{a}	$6.5^{a,b}$	5.1	8.4	7.8
0	6	4.9	$7.2^{a–c}$	6.6^{a}	5.1	8.4	7.8
0	12	4.9	$7.2^{a–c}$	$6.2^{a,b}$	5.0	8.3	7.6
0	18	4.8	$7.2^{a–c}$	$6.2^{a,b}$	4.9	8.3	7.5
1.5	0	4.8	$7.3^{a,b}$	6.6^{a}	4.6	8.4	7.8
1.5	6	4.8	$7.2^{a–c}$	$6.2^{a,b}$	4.8	8.4	7.7
1.5	12	4.9	$7.1^{a–c}$	$6.2^{a,b}$	4.7	8.3	7.6
1.5	18	4.9	$6.9^{a–c}$	$6.1^{a,b}$	4.8	8.8	7.5
3.0	0	4.8	$6.7^{c–e}$	$6.2^{a,b}$	4.8	8.2	7.6
3.0	6	4.8	$6.4^{d,e}$	$6.1^{a,b}$	4.6	8.2	7.5
3.0	12	4.8	$6.8^{c–e}$	$6.1^{a,b}$	4.7	8.0	7.7
3.0	18	4.9	$6.8^{b–e}$	$6.2^{a,b}$	4.6	8.1	7.6
4.5	0	4.9	$6.9^{b–d}$	$6.0^{a,b}$	4.7	8.2	7.5
4.5	6	4.9	$7.0^{a–c}$	$6.0^{a,b}$	4.6	8.0	7.4
4.5	12	4.8	$6.9^{b–d}$	$6.0^{a,b}$	4.7	8.1	7.5
4.5	18	4.8	6.3^{e}	5.9^{b}	4.6	8.1	7.5
SEM[3]		0.07	0.15	0.20	0.19	0.18	0.19
Main Effect Means[4]							
ISP (%)							
0	–	4.9	7.2^{a}	6.4^{a}	5.0^{a}	$8.3^{a,b}$	7.7
1.5	–	4.9	7.1^{a}	$6.3^{a,b}$	$4.8^{a,b}$	8.4^{a}	7.6
3.0	–	4.8	6.7^{b}	$6.2^{a,b}$	4.7^{b}	8.1^{b}	7.6
4.5	–	4.9	6.8^{b}	6.0^{b}	4.6^{b}	8.1^{b}	7.5
	BR (%)						
–	0	4.9	7.1^{a}	6.3	4.9	8.3	7.7
–	6	4.9	$7.0^{a,b}$	6.3	4.8	8.2	7.6
–	12	4.9	$7.0^{a,b}$	6.1	4.8	8.2	7.6
–	18	4.9	6.8^{b}	6.1	4.7	8.2	7.5
SEM		0.04	0.07	0.10	0.10	0.09	0.10
P-value							
ISP		0.47	<0.0001	0.05	0.03	0.05	0.52
BR		0.89	0.10	0.33	0.79	0.89	0.87
ISP × BR		0.48	0.14	0.80	1.00	1.00	0.97
Orthogonal polynomials							
ISP							
Linear		0.22	<0.0001	0.01	0.005	0.02	0.14
Quadratic		0.33	0.21	0.75	0.42	0.69	0.92
BR							
Linear		0.59	0.03	0.07	0.36	0.68	0.45
Quadratic		0.63	0.64	0.85	1.00	0.57	0.98

[1] *Lactobacillus*. [2] *Bifidobacteria*.

[a–e] Means in the same column for every effect with uncommon superscript are significantly different at $P \leq 0.05$.

[3] Standard error mean.

[4] Means represent 15 birds per pen, 24 pens per diet.

main source of dietary energy for poultry [37]. Rice starch has smaller granule size (5 to 15 mm of diameter) and lower amylose content (170 to 270 g/kg) than those in corn starch [38]. Also, total insoluble NSP content and encapsulation of starch are lower for rice than for corn, consequently the digestion process in the gastrointestinal tract is easier for rice [39]. The rate of starch digestion (**RSD**) is an important factor affecting bird's performance [40, 20]. Gutierrez del Alamo et al. [40] reported that broiler growth and FCR were improved quadratically with an increase in RSD. Similarly, Ball et al. [20] found a positive correlation between the RSD and performance parameters. Our results support the positive effect of rice inclusion in pre-starter diet on broiler chicken's performance at market age.

Intestinal Microflora

Effect of the pre-starter diet containing different levels of ISP and broken rice and their interaction on microflora counts in ileal and cecal digesta of broiler chickens at d 10 (log10 cfu/g content) are shown in Table 3. The inclusion of ISP in pre-starter diet did not have a significant effect ($P > 0.05$) on *Escherichia coli* counts in ileal content. Whereas the pre-starter diet containing 3% or 4.5% ISP decreased ($P < 0.05$) *Lactobacillus* counts and the inclusion 4.5% ISP decreased ($P < 0.05$) *Bifidobacteria* counts in ileal content compared to those fed control diet. The use of ISP in pre-starter diet decreased ($P <$ 0.05) *Escherichia coli* and *Lactobacillus* counts in cecal content. The *Bifidobacteria* counts in cecal content of chicks was not influenced ($P > 0.05$) by the use of different levels of ISP in the pre-starter diet at 10 d of age. The inclusion of broken rice in pre-starter diet did not have an effect ($P > 0.05$) on miroflora counts in ileal and cecal contents at 10 d of age, with the exception of 18% broken rice that decreased *Lactobacillus* counts in ileal content. The pre-starter diet contained different levels of ISP and broken rice did not have a significant effect on *Escherichia coli* counts in ileum and *Escherichia coli*, *Lactobacillus* and *Bifidobacteria* counts in ceca content at 10 d of age. The use of 4.5% ISP and 18% broken rice in the pre-starter diet decreased ($P < 0.05$) *Lactobacillus* counts in ileum compared with those fed control diet at 10 d of age.

Based on the results of the current study, the use of ISP and broken rice in pre-starter diet decrease intestinal microflora counts. It has been reported that high level of NSPs leads to increased digesta viscosity and a decline in nutrient digestibility [41]. In addition, there is a negative correlation between the amount of NSPs and the nutritive value of feedstuffs for poultry [33]. Because ISP contains trace amount of oligosaccharide, total NSPs of the diets have been declined by the use of ISP in diet. Therefore, with an increase in dietary level of ISP, digesta viscosity may decrease, nutrients digestibility increase and consequently cause a reduction in substrates for microflora in the ileum and ceca. The intestinal microflora counts were reduced, because of a limit in the amount of available substrates to microflora in ileum and ceca. Diet composition can affect gut microflora at least by 2 ways; first, changing the physico-chemical properties (i.e., viscosity or pH) of chyme and second, supplying the nutrients for the growth and proliferation of specific groups of microflora [42]. In our study, *Lactobacillus* counts in ileum were reduced by 14% when chicks fed a pre-starter diet contained 4.5% ISP and 18% broken rice compared to control at 10 d of age.

Intestinal Size and Morphology

The effect of pre-starter diet containing different levels of ISP and broken rice on relative weight (g/100 g body weight) and length (cm/100 g body weight) of intestinal segments of broiler chicks at 5 and 10 d of age are shown in Table 4. There was no interaction among treatments on relative weight and length of different segments of small intestine in 5- and 10-day-old chicks with the exception of ileum relative length at 10 d of age. The inclusion of 4.5% ISP in pre-starter diet increased relative weight of duodenum ($P = 0.058$) compared with those fed other diets and increased relative weight of ileum ($P = 0.023$) comparing with control fed birds at 5-day-old chicks. The use of ISP or broken rice in the pre-starter diet did not have an effect ($P > 0.05$) on relative weight of jejunum at 5- or 10-day-old chicks. The chick's relative weight of duodenum and jejunum were not affected when they were fed diet containing ISP at 10 d of age. However, the relative weight of duodenum in chick's fed pre-starter diet containing 4.5% ISP was significantly higher at 5 d of age than those fed control or other diets containing ISP. Chicks fed diet contained 4.5% ISP had higher ($P < 0.05$) relative weight of ileum than those fed control or other diets containing ISP at 10 d of age. The dietary ISP did not have an effect ($P > 0.05$) on relative length of intestinal segments of broiler chicks at 5 or 10 d of age. The use of broken rice in the pre-starter diet did not have an effect ($P > 0.05$) on the relative weight and relative length of duodenum, jejunum, and ileum at 5 or 10 d of age, with the exception of

Table 4. Effect of pre-starter diet containing different levels of isolated soy protein (ISP) and broken rice (BR) on intestinal segments of 5- and 10-day-old chicks.

	Relative weight (g/100 g body weight)						Relative length (cm/100 g body weight)					
Dietary treatments	Duodenum		Jejunum		Ileum		Duodenum		Jejunum		Ileum	
	5 d	10 d	5 d	10 d	5 d	10 d	5 d	10 d	5 d	10 d	5 d	10 d
ISP (%)												
0	1.6^{b}	1.4	2.6	2.9	2.0^{b}	1.8^{b}	13.4	7.3	27.5	16.9	25.1	17.0
1.5	1.6^{b}	1.3	2.5	2.7	2.1^{a,b}	1.8^{b}	13.4	7.4	26.3	17.0	24.7	17.1
3.0	1.6^{b}	1.3	2.5	2.8	2.1^{a,b}	1.8^{b}	13.3	7.1	25.7	16.9	25.1	16.6
4.5	1.7^{a}	1.3	2.6	2.8	2.2^{a}	2.0^{a}	13.6	7.1	26.4	16.9	25.7	17.4
BR (%)												
0	1.6	1.3	2.5	2.8	2.1	1.8	13.5^{a,b}	7.2	26.0	16.9	25.2	16.8
6	1.6	1.4	2.5	2.7	2.2	1.9	13.1^{b}	7.3	25.9	16.9	25.1	17.1
12	1.6	1.4	2.6	2.9	2.0	1.8	13.0^{b}	7.2	26.2	17.0	25.1	17.4
18	1.6	1.3	2.5	2.8	2.1	1.9	14.0^{a}	7.1	27.8	17.0	25.2	16.8
SEM[1]	0.04	0.02	0.06	0.06	0.06	0.04	0.22	0.11	0.73	0.09	0.33	0.24
P-value												
Sources of variation												
ISP	0.06	0.67	0.64	0.22	0.02	0.01	0.85	0.08	0.36	0.67	0.23	0.23
BR	0.98	0.15	0.65	0.42	0.48	0.26	0.01	0.80	0.22	0.75	1.00	0.28
ISP × BR	0.41	0.81	0.86	0.13	0.91	0.47	0.25	0.67	0.89	0.05	0.21	0.01
Orthogonal polynomials												
ISP												
Linear	0.03	0.55	0.90	0.63	0.002	0.002	0.66	0.03	0.21	0.97	0.16	0.54
Quadratic	0.14	0.87	0.23	0.22	1.00	0.07	0.50	0.81	0.20	0.33	0.13	0.22
BR												
Linear	0.89	0.88	0.46	0.99	0.42	0.94	0.16	0.46	0.08	0.32	0.91	0.94
Quadratic	0.97	0.02	0.43	0.52	0.95	0.62	0.003	0.51	0.26	0.73	0.90	0.08

^{a,b}Means in the same column for every effect with uncommon superscript are significantly different at $P \leq 0.05$. [1]Standard error mean.

relative length of duodenum at 5-day-old. Chicks fed the pre-starter diet containing 18% broken rice had higher ($P = 0.011$) relative length of duodenum than those fed diets contained 6% or 12% broken rice, but were similar to those fed control diet at 5 d of age.

Effect of ISP and broken rice in the pre-starter diet and their interaction on intestinal villus height and villus height to crypt depth ratio (**VCR**) of broiler chicks are shown in Tables 5 and 6, respectively. The villus height of duodenum increased ($P < 0.05$) when chicks fed pre-starter diet containing 3% or 4.5% ISP at 5 or 10 d of age. Feeding pre-starter diet containing different levels of ISP linearly increased ($P < 0.05$) villus height in jejunum and ileum of 5- or 10-day-old chicks. The VCR for duodenum was unaffected by dietary ISP at 5- or 10-day-old chicks. The use of ISP in the pre-starter diet increased ($P < 0.05$) VCR in jejunum of 5- or 10-day-old chicks and linearly increased ($P < 0.05$) VCR in ileum of 5-day-old chicks. A significant linear and quadratic effect of broken rice inclusion was observed on villus height in jejunum and ileum ($P < 0.05$) of 5- or 10-day-old chicks. The use of broken rice in the pre-starter diet increased ($P < 0.0001$) villus height in duodenum of 5-day-old chicks. In addition, villus height in jejunum and ileum were increased ($P < 0.0001$) when chicks fed pre-starter diet contained broken rice at 5- or 10-day-old birds. There were no differences ($P > 0.05$) in villus height of duodenum among chicks fed pre-starter diet contained different levels of broken rice at 10-day-old chicks. The VCR in duodenum was unaffected ($P > 0.05$) by the inclusion of broken rice of 5- or 10-day-old chicks. Dietary broken rice caused a significant increase ($P < 0.05$) in VCR of jejunum and ileum of chicks compared to those fed control diet at 5- or 10-day-old chicks. The villus height of duodenum and jejunum increased ($P < 0.05$)

Table 5. Effect of pre-starter diet containing different levels of isolated soy protein (ISP) and broken rice (BR) on intestinal villus height of 5- and 10-day-old chicks.

ISP × BR		Villus height (μm)					
(%)	(%)	Duodenum		Jejunum		Ileum	
		5 d	10 d	5 d	10 d	5 d	10 d
0	0	989^{g}	1,175^{f,g}	621^{h}	817^{f}	432^{i}	525^{f,g}
0	6	990^{f,g}	1,184^{e,f}	631^{f,g}	823^{f}	436^{h,i}	528^{e-g}
0	12	997^{d,e}	1,192^{c-e}	629^{f,g}	818^{f}	439^{g,h}	531^{d-f}
0	18	1,000^{b-d}	1,168^{g}	631^{f,g}	823^{f}	443^{g}	536^{b-d}
1.5	0	983^{h}	1,181^{f}	626^{g,h}	820^{f}	446^{g}	526^{e-g}
1.5	6	997^{d,e}	1,197^{b-d}	645^{b,c}	845^{c}	455^{e,f}	541^{a,b}
1.5	12	998^{d,e}	1,176^{f,g}	645^{b,c}	837^{d,e}	467^{b,c}	525^{g}
1.5	18	1,004^{b}	1,169^{g}	642^{c,d}	857^{b}	466^{c,d}	535^{c,d}
3.0	0	985^{g,h}	1,200^{a-c}	639^{d,e}	818^{f}	443^{g}	528^{e-g}
3.0	6	1011^{a}	1,180^{f}	647^{b,c}	845^{c}	453^{f}	528^{e-g}
3.0	12	999^{c-e}	1,183^{e,f}	634^{e,f}	845^{c}	465^{c,d}	543^{a}
3.0	18	1,003^{b,c}	1,203^{a,b}	661^{a}	833^{e}	462^{c-e}	532^{c-e}
4.5	0	994^{e,f}	1,191^{d,e}	649^{b}	834^{e}	461^{c-e}	530^{d-g}
4.5	6	1,011^{a}	1,197^{b-d}	659^{a}	841^{c,d}	460^{d,e}	537^{b,c}
4.5	12	1,012^{a}	1,196^{b-d}	660^{a}	876^{a}	476^{a}	534^{c,d}
4.5	18	1,011^{a}	1,208^{a}	660^{a}	875^{a}	473^{a}	536^{b-d}
SEM[1]		1.62	2.82	2.06	2.02	2.13	1.81
Main Effect Means[2]							
ISP (%)							
0	–	994^{c}	1,180^{c}	628^{d}	820^{d}	437^{c}	530^{b}
1.5	–	995^{c}	1,181^{c}	639^{c}	839^{b}	458^{b}	532^{a,b}
3.0	–	999^{b}	1,191^{b}	645^{b}	835^{c}	456^{b}	532^{a,b}
4.5	–	1,007^{a}	1,198^{a}	657^{a}	856^{a}	467^{a}	534^{a}
	BR (%)						
–	0	988^{c}	1,187	634^{c}	822^{d}	445^{c}	527^{b}
–	6	1,002^{b}	1,189	645^{a}	838^{c}	451^{b}	534^{a}
–	12	1,001^{b}	1,187	642^{b}	843^{b}	461^{a}	533^{a}
–	18	1,004^{a}	1,187	648^{a}	847^{a}	461^{a}	534^{a}
SEM		0.81	1.41	1.03	1.01	1.07	0.90
P-value							
ISP		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.02
BR		<0.0001	0.52	<0.0001	<0.0001	<0.0001	<0.0001
ISP × BR		<0.0001	<0.0001	<0.0001	<0.0001	0.03	<0.0001
Orthogonal polynomials							
ISP							
Linear		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.002
Quadratic		0.001	0.07	0.72	0.39	<0.0001	0.84
BR							
Linear		<0.0001	0.76	<0.0001	<0.0001	<0.0001	<0.0001
Quadratic		<0.0001	0.49	0.01	<0.0001	0.004	0.01

$^{a-h}$Means in the same column for every effect with uncommon superscript are significantly different at $P \leq 0.05$.
[1]Standard error mean. [2]Means represent 15 birds per pen, 24 pens per diet.

when chicks fed pre-starter diet containing 4.5% ISP and 6%, 12% or 18% broken rice at 5 d of age. Chicks fed pre-starter diets contained 4.5% ISP and 12% or 18% broken rice had higher ($P < 0.05$) villus height in ileum at 5 d of age. The use of 4.5% ISP and 12% or 18% broken rice in the pre-starter diet increased ($P < 0.05$) villus height in jejunum and ileum of 10-day-old chicks.

The use of 4.5% ISP in the pre-starter diet increased relative weight of duodenum and ileum, but there were no significant differences in the

Table 6. Effect of pre-starter diet containing different levels of isolated soy protein (ISP) and broken rice (BR) on intestinal villus height to crypt depth ratio (VCR) of 5- and 10-day-old chicks.

ISP × BR		VCR					
(%)	(%)	Duodenum		Jejunum		Ileum	
		5 d	10 d	5 d	0 d	5 d	10 d
0	0	7.85	9.04^{a-e}	5.32	6.04^{e}	3.83	4.64
0	6	7.78	9.13^{a-c}	5.40	6.25^{b-e}	3.76	4.67
0	12	7.93	8.74^{c-f}	5.42	6.12^{c-e}	3.93	4.70
0	18	7.83	8.70^{d-f}	5.45	6.15^{c-e}	3.85	4.68
1.5	0	7.71	9.09^{a-d}	5.36	6.05^{e}	3.94	4.38
1.5	6	7.96	8.91^{a-f}	5.52	6.32^{a-d}	3.96	4.63
1.5	12	7.88	8.68^{e-g}	5.49	6.31^{a-d}	4.08	4.72
1.5	18	8.01	$8.58^{f,g}$	5.49	6.34^{a-c}	4.10	4.86
3.0	0	7.99	$8.63^{f,g}$	5.47	6.27^{a-e}	4.04	4.48
3.0	6	7.84	8.76^{b-f}	5.54	6.36^{a-c}	4.05	4.41
3.0	12	7.70	8.93^{a-f}	5.32	6.12^{c-e}	4.26	4.65
3.0	18	7.61	9.25^{a}	5.63	6.36^{a-c}	4.19	4.86
4.5	0	7.88	8.31^{g}	5.56	6.28^{a-e}	4.24	4.46
4.5	6	7.92	8.89^{a-f}	5.61	$6.07^{d,e}$	4.24	4.70
4.5	12	7.89	$9.15^{a,b}$	5.58	$6.46^{a,b}$	4.40	4.71
4.5	18	7.98	9.27^{a}	5.59	6.52^{a}	4.35	4.81
SEM[1]		0.12	0.12	0.05	0.08	0.06	0.08
Main Effect Means[2]							
ISP (%)							
0	–	7.85	8.91	5.40^{c}	6.14^{b}	3.84^{d}	4.67
1.5	–	7.89	8.82	$5.47^{b,c}$	6.26^{a}	4.02^{c}	4.65
3.0	–	7.79	8.90	5.49^{b}	6.28^{a}	4.14^{b}	4.60
4.5	–	7.92	8.91	5.58^{a}	6.33^{a}	4.31^{a}	4.67
	BR (%)						
–	0	7.86	8.77	5.43^{c}	6.16^{b}	4.01^{b}	4.49^{c}
–	6	7.88	8.92	$5.52^{a,b}$	$6.25^{a,b}$	4.00^{b}	4.60^{b}
–	12	7.85	8.88	$5.45^{b,c}$	$6.25^{a,b}$	4.17^{a}	$4.69^{a,b}$
–	18	7.86	8.95	5.54^{a}	6.34^{a}	4.12^{a}	4.80^{a}
SEM		0.06	0.06	0.03	0.04	0.03	0.04
P-value							
ISP		0.42	0.69	<0.0001	0.01	<0.0001	0.53
BR		0.99	0.18	0.01	0.02	0.0003	<0.0001
ISP × BR		0.29	<0.0001	0.06	0.003	0.98	0.11
Orthogonal polynomials							
ISP							
Linear		0.65	0.77	<0.0001	0.001	<0.0001	0.77
Quadratic		0.45	0.42	0.64	0.47	0.94	0.22
BR							
Linear		0.90	0.08	0.02	0.003	0.001	<0.0001
Quadratic		0.95	0.52	0.89	0.97	0.54	0.97

$^{a-g}$Means in the same column for every effect with uncommon superscript are significantly different at $P \leq 0.05$.

[1]Standard error mean. [2]Means represent 15 birds per pen, 24 pens per diet.

relative length of intestinal segments in chicks fed any of the ISP containing diets, compared to those fed control diet. Jankowski et al. [28] showed that the use of 19.5% ISP that contained only trace amounts of oligosaccharides in diet significantly decreased ileum weight of young turkeys. It is postulated that a total withdrawal of oligosaccharides from a diet may result in undesirable hypotrophy of small intestinal tissue. In our study, there were no significant differences in the relative weight and relative length of small intestine segments among chicks fed a pre-starter diet containing different levels of broken rice and those fed a control diet. Similarly, Kita and Okuten [33] reported that replacement of corn with rice did not have a significant effect on relative weight of jejunum and ileum in young chicks. The results of the current study revealed that the inclusion of ISP in pre-starter diet, increased villus height in duodenum, jejunum, and ileum in 10-day-old chicks. It also caused a linear increase in VCR in jejunum and ileum in 10-day-old chicks. Higher villus height and lower crypt depth in the small intestine are important indicators of gut development and health, and as such influence nutrient digestion and absorption [43]. Performance of broiler chickens in our study support more effective absorption of nutrients by villi in birds fed pre-starter diet containing different levels of ISP (1.5 to 4.5%) and broken rice (6 to 18%).

CONCLUSIONS AND APPLICATIONS

1. Pre-starter diet containing 4.5% ISP and 18% broken rice improved BWG by 4% and FCR by –3% in broiler chickens during 1 to 42 d of age.
2. The use of ISP and broken rice in pre-starter diet significantly increased ($P < 0.05$) the average villus height of duodenum, jejunum, and ileum by 6% at d 5 and 4% at d 10 of age.
3. The *Lactobacillus* counts in ileum was reduced by 14% when chicks fed a pre-starter diet contained 4.5% ISP and 18% broken rice compared with those fed control diet in 10-day-old chicks. Since, dietary ISP (4.5%) had a negative effect on beneficial bacteria, further research is needed to study the effect of pre- and probiotics in pre-starter diet containing ISP on gut microbiota.

REFERENCES AND NOTES

1. Jin, S. H., A. Corless, and J. L. Sell. 1998. Digestive system development in post hatch poultry. World's Poult. Sci. J. 54:335–345.

2. Wijtten, P. J. A., D. J. Langhout, and M. W. A. Verstegen. 2012. Small intestine development in chicks after hatch and in pigs around the time of weaning and its relation with nutrition: A review. Acta Agric. Scandi. A. Anim. Sci. 62:1–12.

3. Noy, Y., and D. Sklan. 1997. Post hatch development in poultry. J. Appl. Poult. Res. 6:344–354.

4. Willemsen, H., N. Everaert, A. Witters, L. De Smit, M. Debonne, F. Verschuere, P. Garain, D. Berckmans, E. Decuypere, and V. Bruggeman. 2008. Critical assessment of chick quality measurements as an indicator of post hatch performance. Poult. Sci. 87:2358–2366.

5. Sklan, D., and Y. Noy. 2003. Crude protein and essential amino acid requirements in chicks during the first week post hatch. Br. Poult. Sci. 44:266–274.

6. Kidd, M. T., A. Corzo, D. Hoehler, E. R. Miller, and W. A. Dozier, III. 2005. Broiler responsiveness (Ross × 708) to diets varying in amino acid density. Poult. Sci. 84:1389–1396.

7. Irish, G. G., and D. Balnave. 1993. Non-starch polysaccharides and broiler performance on diets containing soya bean meal as the sole protein concentrate. Aust. J. Agric. Res. 44:1483–1499.

8. Meng, X., and B. A. Slominki.2005. Nutritive values of corn, soybean meal, canola meal, and peas for broiler chickens as affected by a multicarbohydrase preparation of cell wall degrading enzymes. Poult. Sci. 84:1242–1251.

9. Salami, R. I., and A. A. Odunsi. 2003. Evaluation of processed cassava peel meals as substitutes for maize in the diets of layers. Int. J. Poult. Sci. 2:112–116.

10. Siddhuraju, P., and K. Becker. 2005. Nutritional and anti-nutritional composition, in vitro amino acid availability, starch digestibility and predicted glycemic index of differentially processed gela mucuna beans (Mucuna pruriens var. utilis): an under- utilized legume. Food Chem. 91:275–286.

11. Choct, M. 2015. Feed non-starch polysaccharides for mono gastric animals: classification and function. Anim. Prod. Sci. 55:1360–1366.

12. Peisker, M. 2001. Manufacturing of soy protein concentrate for animal nutrition. Cahiers Options Médit. 54:103–107.

13. Grieshop, C. M., and J. R. G. C. Fahey. 2000. The role of soy in companion animal nutrition. Pages 171–181 in Soy in Animal Nutrition. Drackley, J. K., ed. Federation of Animal Science Societies, Champaign, IL.

14. Gonzalez-Alvardo, J. M., E. Jimenz-Moreno, R. Lazaro, and G. G. Mateos. 2007. Effects of type of cereal, heat processing of the cereal, and inclusion of fiber in the diet on productive performance and digestive traits of broilers. Poult. Sci. 86:1705–1715.

15. Franco, C. M. L., S. J. R. Preto, and C. F. Ciacco. 1992. Factors that affect the enzymatic degradation of natural starch granules effect of the size of the granules. Starch. 44:422–426.

16. Eliasson, A. C., and M. Gumundsson. 1996. Starch: physicochemical and functional aspects. Pages 431–503 in Carbohydrates in Food. Eliasson, A. C.. Marcel Dekker, Inc., New York, NY.

17. Weurding, R. E., A. Veldman, W. A. G. Veen, P. J. Van der Avar, and M. W. A. Verstegen. 2001. Starch digestion rate in the small intestine of broiler chickens differs among feedstuffs. J. Nutr. 131:2329–2335.

18. Li, J. H., T. Vasanthan, R. Hoover, and B. G. Rossnagel. 2004. Starch from hull-less barley: V. In vitro susceptibility of waxy, normal, and high amylose starches towards hydrolysis by alpha-amylases and amyloglucosidase. Food Chem. 84:621–632.

19. Riley, C. K., A. O. Wheatley, I. Hassan, M. H. Ahmad, E. Y. S. A. Morroson, and H. N. Asemota. 2004.

In vitro digestibility of raw starches extracted from five yam (Dioscorea spp.) species grown in Jamaica. Starch. 56: 69–73.

20. Ball, M. E. E., B. Owens, and K. J. Mccraken. 2013. Chemical and physical predictors of the nutritive value of wheat in broiler diets. Asian-Australas. J. Anim. Sci. 26:97–107.

21. House temperature was initially set at 32–34°C by the thermostatically controlled heating system on d 1 and decreased by 0.5°C per day to reach a temperature of 21–23°C on d 22 and kept constant thereafter. All birds exposed to 23L:1D throughout the experiment.

22. Pouyan – MK 1120- Pouyan Co, Mashhad, Iran.

23. Bilini, A., C. F. Quintana, A. Menconi, G. Kallapura, J. Latorre, C. Pixley, S. Layton, M. Dalmagro, X. Hernadez-Velasco, A. Wolfenden, B. M. Hargis, and G. Tellez. 2013. Evaluation of effects of EarlyBird associated with FloraMax-B11 on Salmonella Enteritidis, intestinal morphology, and performance of broiler chickens. Poult. Sci. 92:337–346.

24. SAS User's Guide. 2009. Version 9.2 ed. SAS Institute Inc., Cary, NC, USA.

25. Longo, F. A., J. F. M. Menten, A. A. Pedroso, A. N. Figueiredo, A. M. C. Racanicci, J. B. Gaiotto, and J. O. B. Sorbara. 2005. Different sources of protein in the diet of newly hatched chicks. Rev. Bras. Zootec. 34:112–122.

26. Ebling, P. D., A. M. Kessler, A. P. Villanueva, G. C. Pontalti, G. Farina, and A. M. L. Ribeiro. 2015. Rice and soy protein isolate in pre-starter diets for broilers. Poult. Sci. 94:2744–2752.

27. Van der Eijk, C. 2015. Reducing anti-nutritional factors in pre-starter feeds. Asian Feed Mag. 6–9.

28. Jankowski, J., J. Juskiewicz, K. Gulewicz, A. Lecewicz, B. A. Slominski, and Z. Zdunczyk. 2009. The effect of diets containing soybean meal, soybean protein concentrate, and soybean protein isolate of different oligosaccharide content on growth performance and gut function of young turkeys. Poult. Sci. 88:2132–2140.

29. Li, Z., D. Li., S. Qiao, X. Zhu, and C. Huang. 2003. Anti-nutritional effects of a moderate dose of soybean agglutinin in the rat. Arch. Anim. Nutr. 57:267–277.

30. Friedman, M., D. L. Brandon, A. H. Bates, and T. Hymowtiz. 1991. Comparison of a commercial soybean cultivar and an isoline lacking the Kunitz trypsin inhibitor: composition, nutritional value, and effects of heating. J. Agric. Food Chem. 39:327–335.

31. Saki, A. A., M. Abbasinezhad, S. H. Ghazi, M. M. Tabatabi, A. Ahmadi, and K. H. Zaboli. 2012. Intestinal characteristics, alkaline phosphatase and broilers performance in response to extracted and mechanical soybean meal replaced by fish meal. J. Agric. Sci. Tech. 14:105–114.

32. Van Kempen, T. A. T. G., E. Vanheugten, A. J. Moeser, N. S. Muley, and V. J. S. Sewalt. 2006. Selecting soybean meal characteristics preferred for swine nutrition. J. Anim. Sci. 84:1387–1395.

33. Kita, K., and A. Okuten. 2013. Influence of dietary rice grain replaced from corn on serum concentrations of amino acids in young chickens. J. Poult. Sci. 50:114–119.

34. Choct, M., and G. Annison. 1990. Anti-nutritive activity of wheat pentosans in broiler diets. Br. Poult. Sci. 31:811–821.

35. Ikegami, S., F. Tsuchihashi, H. Harada, N. Tsuchihashi, E. Nishide, and S. Innami. 1990. Effect of viscous indigestible polysaccharides on pancreatic-biliary secretion and digestive organs in rats. J. Nutr. 120:353–360.

36. Johnson, I. T., and J. M. Gee. 1981. Effect of gel-forming gums on the intestinal unstirred layer and sugar transport in vitro. Gut. 22:398–403.

37. Mc Nab, J. M. 1996. Factors affecting the energy value of wheat for poultry. World. Poult. Sci. J. 52:69–73.

38. Tester, R. F., J. Karkalas, and X. Qi. 2004. Starch-composition, fine structure and architecture. J. Cereal Sci. 39:151–165.

39. Svihus, B., A. K. Uhien, and O. M. Herstad. 2005. Effect of starch granule structure, associated components and processing on nutritive value of cereal starch: a review. Anim. Feed Sci. Technol. 122:303–320.

40. Gutierrez del alamo, A., M. W. A. Verstegen, L. A. Denhartog, P. Perez de ayala, and M. J. Villamide. 2008. Effect of wheat cultivar and enzyme addition to broiler chicken diets on nutrient digestibility, performance, and apparent metabolizable energy content. Poult. Sci. 87: 759–767.

41. Pan, D., and Z. Yu. 2014. Intestinal microbiome of poultry and its interaction with host and diet. Gut Microbes. 5:108–119.

42. Shakouri, M. D., P. A. Iji, L. L. Mikkelsen, and A. J. Cowieson. 2009. Intestinal function and gut microflora of broiler chickens as influenced by cereal grains and microbial enzyme supplementation. J. Anim. Physiol. Anim. Nutr. 93:647–658.

43. Wang, J. X., and K. M. Peng. 2008. Developmental morphology of the small intestine of African ostrich chicks. Poult. Sci. 87:2629–2635.

Acknowledgments

The authors would like to appreciate the Vice President in Research and Technology, at the Ferdowsi University of Mashhad for their financial support of this study (project # 37844).