P-ISSN: 2304-3075; E-ISSN: 2305-4360

International Journal of Veterinary Science

www.ijvets.com; editor@ijvets.com

Research Article https://doi.org/10.47278/journal.ijvs/2024.194

Efficacy of Enzyme-Treated Soybean Meal on Broiler Performance, Nutrient Digestibility and Carcass Quality

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Article History: 24-479	Received: 26-Apr-24	Revised: 20-May-24	Accepted: 21-May-24	Online First: 15-Jun-24

ABSTRACT

The study evaluated the impact of enzyme-treated soybean meal with yeast (HP AviStart; HPA) on broiler performance, nutrient digestibility, and carcass quality. A total of 900 Ross 308 chickens of mixed sexes were randomly allocated to six treatments, each with six repetitions. The dietary treatments were corn-soybean diet with HPA 5% replacement soybean meal in four treatments: 1-10d for pre-starter; 1-21d for both pre-and starter in combination with methylene disalicylate (BMD) in pre-starter (1-10d) and 1-21d for both pre- and starter compared with the control (corn soybean diet) and the control with BMD in feeding. The overall study (1–42d) showed that the overall feed conversion ratio values were similar to those of BMD or HPA, but they were much better than the control group (P<0.05). They were also better for the percentage of survival rate, the productive index, and the drop in feed cost per gain, but these differences were not significant. However, supplemental HPA, including 5% replacement of SBM in the control diet, showed evidence of economic returns and showed a higher return on investment than the control group; it was higher when supplementing HPA for the whole period of the pre-starter and starter diets (1-21d). For nutrient digestibility in 21-year-old broilers, the results showed better significant digestibility of crude protein, ether extract, calcium, and phosphorus when broilers were fed with HPA or HPA plus BMD in the diets (P<0.05). There were no big differences in the carcass's quality or the organs' weight between the treatments. However, the white stripes on the breast meat got significantly less noticeable (P<0.05), and footpad lesion scores decreased in all treatments that included HPA in the feed. Supplementation with HPA showed a significant increase in fat deposition in the small intestine at 42d of age versus the control and antibiotic BMD groups (P<0.05). HPA supplementation (1-10d or 1-21d) promotes antibiotic growth with improved broiler performance and carcass quality and, thus, a higher return on investment. These data indicate that HPA provided both a high quality of protein and metabolizable energy value compared to a control diet.

Key words: Broiler, Enzyme treated soybean meal, Performance, Nutrients digestibility, Carcass quality

INTRODUCTION

Soybean meal (SBM) is one of the most consistent and highest-quality protein sources for animal nutrition, especially poultry and swine. To understand the nutritional values of SBM and full-fat soybean meal, they must be properly processed to destroy toxic factors or anti-nutritional factors (ANFs), e.g. trypsin inhibitors, hemagglutinin, saponin, phytic acid, oligosaccharides, lectin, antigen, etc. These all, decrease the nutrition value (Thomasen 1988; Swick 1999; Marsman et al. 1997; Mehri et al. 2010; Van der Eijk 2012, 2013; Wang et al. 2020). Chemical or enzymatic treatments are much more effective, but destroying protein antigenicity by heat treatment is impractical because it seriously impairs the quality of the nutritionally beneficial protein (Deng et al. 2023; Zhu et al. 2023; Tan et al. 2024).

Due to the varying quality of commercial SBM processing, SBM products have been developed via enzyme technology and co-processed with yeast and a non-fermentative, all-natural yeast culture component (Leeper et al. 2023; Uyisenga et al. 2023). Enzyme-treated soybean meal (HP AviStart; HPA) is one example with enhanced phosphorus-bioavailability for applications in animal nutrition. HPA, a highly digestible product that is very low in ANFs and is an excellent protein source for poultry pre-starters or starters. With feeding HPA replacement SBM at 0, 2.5, 5 and 10% in pre-starter broiler diets, it was reported that the addition of HPA to the starter diet significantly improved the live weight and feed conversion ratio of

Cite This Article as: Boonmee T, Jaikan W, Brokner C, Blanch A and Khajarern J, 2024. Efficacy of enzyme-treated soybean meal on broiler performance, nutrient digestibility, and carcass quality. International Journal of Veterinary Science x(x): xxxx. https://doi.org/10.47278/journal.ijvs/2024.194

broilers up to 10 or 35d and marginally improved the meat yield and carcass weight, especially breasts and thighs. According to van der Eijk (2013), these findings confirm that the inclusion of HPA at a level of 5% in broiler starter diets for 7d improved carcass yield and breast weight versus the control. Saengpookhiaw et al. (2018) also indicated that feeding HPA replacement SBM in the pre-starter (1-7d) period for the entire period of broiler diets (42d) in an open-house system showed beneficial effects on better or comparable performance regarding growth, feed efficiency, productive index and feed cost per gain with higher economic benefits and an increase in return on investment versus control SBM. HPA replacement SBM also showed no negative effect on organ weight or abdominal fat, but there were advantages in terms of footpad lesions, white striping, and stool appearance.

It was hypothesized that HPA can improve performance and offer advantages in terms of edible meat, lower fat, the highest return on investment, and carcass quality. Therefore, we conducted this work to investigate the impact of HPA on broiler performance, carcass quality, and nutrient digestibility in an open-house system, both with and without antibiotic growth promoters (AGPs).

MATERIALS AND METHODS

The operations involving animals were conducted in accordance with the National Research Council of Thailand's Ethical Principles for the Use of Animals for Scientific Purposes (U1-07431-2561).

Animals and preparation

There were six treatments with a total of 900 mixed-sex commercial broiler breed (Ross 308) broilers distributed evenly among them. Each treatment included three male and three female replications, with 25 broilers per duplicate. The broilers were assigned to groups based on weight, ensuring each replication group had a similar total weight and weight range. The broilers were kept in pens with fresh rice hull litter on the floor. Each pen had two hanging feeders and one bell drinker. The test facility maintained an ambient temperature between 21.7 and 34.5°C during the trial. Continuous incandescent illumination was provided in each pen. The starting temperature of the room was set at around 30°C and gradually decreased by 2 to 3°C each week in order to find the surrounding temperature. The broilers received vaccinations for Marek's disease, Newcastle disease, and infectious bronchitis in the hatchery. They were also immunized against infectious bursa disease at 12d old. All treatments contained crumbled feed between 1 and 21d and pelleted feed between 22 and 42d. Both meals and drinks were given freely.

Diets

The control diets for pre-starter, starter, grower, and finisher stages were formulated with specific levels of metabolizable energy (ME) and crude protein (CP). The pre-starter diet contained 3,175kcal/kg ME and 23.21% CP, the starter diet contained 3,168kcal/kg ME and 22.84% CP, the grower diet contained 3,174kcal/kg ME and 20.23% CP, and the finisher diet contained 3,631 kcal/kg ME and 18.26% CP (Table 1). Salinomycin (50ppm) supplementation was added in the pre-starter, starter and grower diets to control coccidiosis. The six dietary treatments were: a standard corn soybean diet (negative control, NC, T1), NC with enzyme treated soy HPA 5.0% replacement SBM in pre-starter (1-10d) for T2 and both pre-starter and starter (1-21d) for T3, NC diet with antibiotic Bracitracin methylene disalicylate (BMD) 50g/ton feed in pre-starter, starter and grower diets (1-35d) for T4, T5 is T2 supplementation with BMD 50g/ton feed in pre-starter, starter and grower diets (1-35d) and T6 is T3 supplementation with BMD 50g/ton feed in pre-starter, starter and grower diets (1-35d). HPA composition is presented in Table 2.

Parameters studied

Growth performance and economic benefit

Pen body weight, survival rate (SR), feed efficiency or feed conversion ratio (FCR), feed cost per gain (FCG), economic benefit return, and return on investment (ROI) were recorded. Calculations were also made on the productivity index (PI). The FCR was adjusted correspondingly for the birds that passed away throughout each phase of the study.

Nutrients digestibility

At 16-21d of age, the broilers were taken from each treatment and placed in metabolic cages for fecal collection and digestibility studies. The broilers were fed with diets containing 0.2% chromic oxide as an indigestible marker for five days. The feces were collected during the last 3d of the digestion period. All the collection was kept in a freezer (-20°C) until the analyses were performed. Then, using AOAC (1990), those samples—dry matter (DM), crude protein (CP), crude fiber (CF), and ether extract (EE) were analyzed.

Characteristic of carcass and carcass quality

Following Kuttappan et al. (2012), broilers were randomly chosen with equal weight (three males and three females) from each pen at the end of the experiment (42d of age) and processed to determine carcass quality (dressing percentage of breast meat, thigh, leg, drumstick, wing, and cut up pert of yield as a percentage at 42d of age). The footpad and white stripe scores were evaluated following the methodology described by Kuttappan et al. (2012), as illustrated in Fig. 1 and 2. Furthermore, the heart, liver, pancreas, spleen, and gizzard weights were also recorded. According to Martrenchar et al. (2001), footpad lesion scores were also recorded. Fat deposition in the small intestine, with equal sample weight from each pen, was assessed at 21 and 42 days of age as illustrated in Fig. 3 and 4.

Statistical evaluation

Utilizing the general linear model process of statistical analysis system software, the data were analyzed using analysis of variance (SAS 1995). The Duncan's multiple range test was able to identify that there were differences between the means. The criteria for statistical significance was determined to be a level of P that was less than or equal to 0.05.

Table 1: Composition and calculated analysis of broiler diets

Ingredients	Price *)Baht/kg(	Pre-starter (0-10d)				Pre-starter+ Starter (11-21d)		Starter (11-21d)		Grower (22-35d)		Finisher (36-42d)
		[1]T1)Control(	T2, T3 [2])HPA5(%	T4)[3]BMD(	T5, T6 [4])CMD(	T3)[2]HPA5(%	T6)[4]CMD(	T1, T2)Control(	T4, T5)[3]BMD(	T1, T2, T3)Control(	T4, T5, T6 [3])BMD(	T1-T6)Control(
Corn 8 %CP	11.50	47.69	49.19	47.66	49.96	49.19	49.96	48.89	48.86	56.09	56.06	60.34
Soybean meal 46 %CP	18.00	27.50	20.90	27.50	20.80	20.90	20.80	26.50	26.50	18.00	18.00	13.20
[2]HPA 55.5% CP	42.00	0.00	5.00	0.00	5.00	5.00	5.00	0.00	0.00	0.00	0.00	0.00
Full fat soybean 36 %CP	21.50	18.00	18.00	18.00	18.00	18.00	18.00	18.00	18.00	20.00	20.00	20.00
Rice bran crude-oil	42.00	1.50	1.30	1.50	1.00	1.30	1.00	1.30	1.30	0.60	0.60	1.30
L-Lysine HCl	55.00	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.21	0.21	0.15
DL-Methionine	108.00	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.22	0.22	0.17
MDCaP 21	28.50	1.80	1.70	1.80	1.70	1.70	1.70	1.80	1.80	1.81	1.81	1.84
Limestone	3.50	1.60	2.00	1.60	1.60	2.00	1.60	1.60	1.60	1.63	1.63	1.61
Salt	5.50	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Choline Chloride 60%	36.50	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.08	0.08	0.08
[5]Premix	250.00	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Pellet binder	77.00	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Salinomycin 12%	264.00	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.00
Antimold	250.00	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Toxin binder	250.00	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Antioxidant	450.00	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
[3]BMD 15%	275.00	0.00	0.00	0.03	0.03	0.000	0.03	0.00	0.03	0.00	0.03	0.00
Total		100	100	100	100	100	100	100	100	100	100	100
*Cost (Baht/kg)		17.89	18.87	17.97	18.89	18.87	18.89	17.76	17.85	17.14	17.22	16.84
Calculated analysis												
Energy, kcal ME/kg		3,175	3,175	3,175	3,175	3,175	3,175	3,168	3,168	3,174	3,174	3,631
Crude protein, %		23.21	23.21	23.21	23.21	23.21	23.21	22.84	22.84	20.23	20.23	18.26

*All costs were shown in Thailand bath; 1US$=33.82 bath. [1]T1=Control diet) corn-soybean diet=(NC, T2=NC+HP AviStart 5.0% replacement SBM on pre-starter)d1-10(, T3=NC+HP AviStart 5.0 %replacement SBM on pre-starter)d1-21(. T4=NC+Bracitracin methylene disalicylate) BMD (50g/ton feed)d1-35(, T5=NC+HP AviStart 5.0 % replacement SBM)d1-10+(BMD 50g/ton feed)d1-35(, T6=NC+HP AviStart 5.0 %replacement SBM)d1-21+(BMD 50g/ton feed)d1-35). [2]HPA=HP AviStart 5% replacement soybean meal; [3]BMD=Bracitracin methylene disalicylate) BMD (50g/ton feed. [4]CMD=HP AviStart 5.0 %replacement SBM combine with BMD 50g/ton feed. [5]Premixed mix supplied/kg of diet: Vitamin A: 1,200,000IU, Vitamin D3: 240,00IU, Vitamin E: 2,000mg, Vitamin K: 250mg, Vitamin B1:200mg, Vitamin B2:500 mg, Vitamin B6: 200mg, Vitamin B12: 0.2mg, Biotin: 40mg, Folic acid: 100mg, Niacin: 5,000mg, Pantotenic: 1,500mg, Cu: 900mg, Mn: 6,000mg, Zn: 4,500mg, Fe: 4,000mg, Iodine: 75mg, Se: 30mg. Monodicalciumphosphate21: MDCaP 21.

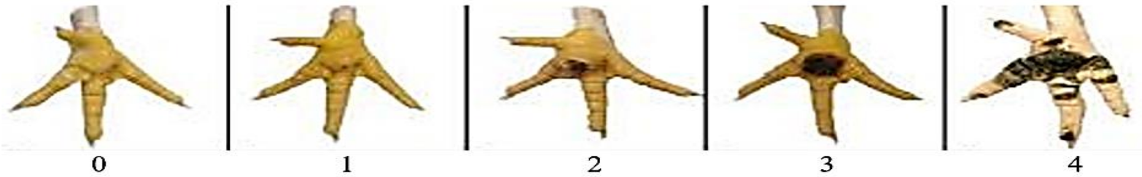

Fig. 1: Evaluation of food pad scores; 0=no lesions; 1=lesions on <25% of pads; 2=lesions on 25%-50% of pads; 3=lesions on 50%-75% of pads; 4=lesions on >75% of pads (Kuttappan et al. 2012).

Table 2: Nutritional component profile of HP AviStart

Item	*HP Avistart (HPA)	*Soybean meal (SBM)
Energy, kcal ME/kg	2,287	3,180
Crude protein, %	55.50	46.20
Phosphorus, %	0.80	0.65
Calcium, %	0.25	0.32
Anti-nutritional factors		
Trysin inhibitor (mg/g)	1.30	5.80
Beta-conglycinine (ppm)	2.00	16.00
Oligosaccharides (%)	1.00	-
- Stachyose	0.30	3.70
- Raffinose	0.40	1.70
Lectins	<1	-
Phytic acid	0.20	0.60

*Hamlet Protein A/S Denmark Company determined the nutritional composition of HP Avistart (HPA) and soybean meal (SBM).

Fig. 2: Evaluation of white stripe score. White stripe scores were 0=normal, 1=moderate, 2=severe, and 3=extreme. Normal – No distinct white lines. Moderate – Small white lines, generally <1 mm thick, but visible on the fillet surface. Severe – Large white lines (1-2mm thick) very visible on the fillet surface. Extreme – Thick white bands (>2mm thickness) covering almost entire surface of fillet (Kuttappan et al. 2012).

RESULTS

Growth performance

Data on the performance of the four feeding phases and overall periods (1-42d of age) are presented in Table 3. During the pre-starter period (1-10d of age), there were no significant differences (P>0.05) in BW, BWG, FI, SR, and FCG among the different treatments. However, when feeding with a diet containing 5% replacement of HPA with SBM, the FCR showed a higher level of significance (P<0.05) compared to diets without BMD. Additionally, the diet with HPA 5% replacement showed a higher PI, although this difference was not statistically significant (P>0.05) compared to the control group fed with SBM. The results indicated that the HPA protein source was a better protein source for easier digestion in terms of amino acid content for retention in the body of the early life of young broilers than the control SBM group when FCR and PI were evaluated in the pre-starter period (1-10d of age).

During the initial period (11-21d of age), there were no statistically significant differences (P>0.05) in BW, BWG, SR and PI across the groups. However, there was an observed increasing trend in PI for broilers fed diets containing HPA, BMD, or a combination of HPA and BMD. The results of FCR and FCG showed that the birds fed HPA, BMD, and HPA plus BMD exhibited significant

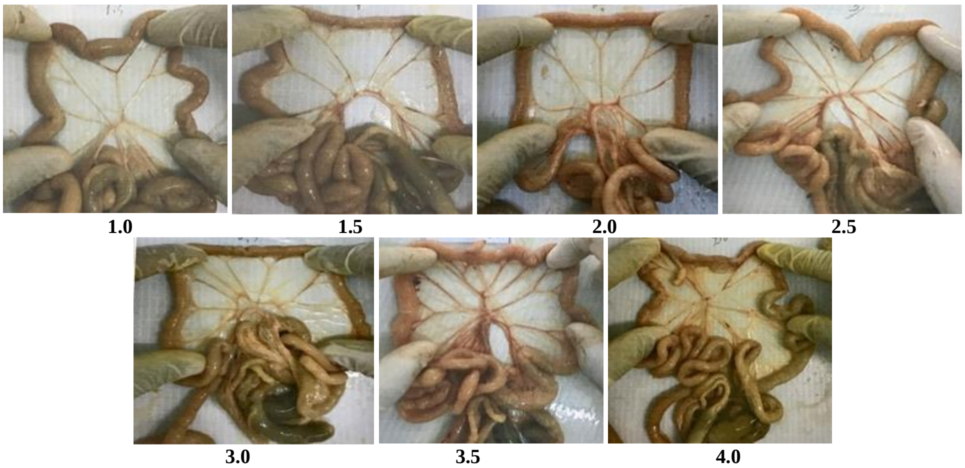

Fig. 3: Evaluation of fat deposition in small intestine score of broilers at 21d.

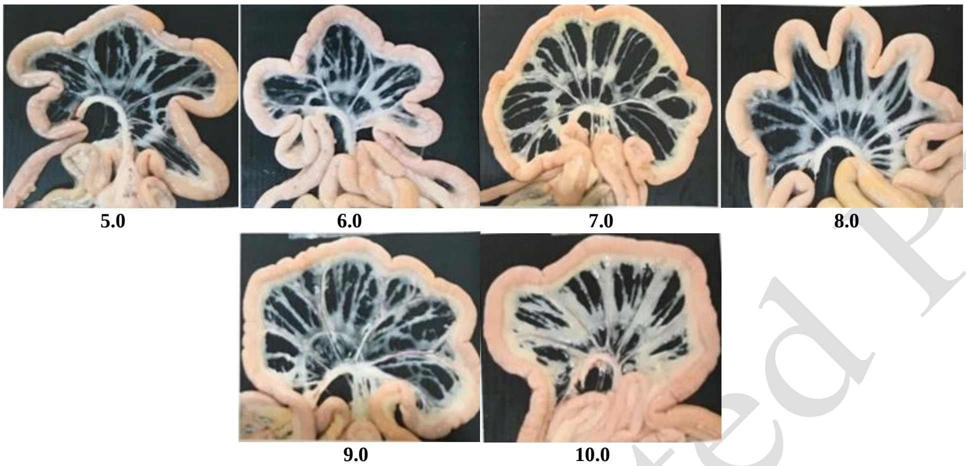

Fig. 4: Evaluation of fat deposition in small intestine score of broilers at 42d.

(P<0.05) improvement of FCR and decreasing FCG more than the control birds.

For the grower period (22-35d of age), no significant differences (P>0.05) were detected on BW, BWG, FI, SR and PI among the treatment groups, but there was an increasing trend in PI with HPA, BMD and HPA plus BMD. However, the results of FCR and FCG showed that birds fed with HPA or BMD or HPA plus BMD showed a significantly (P<0.05) improved FCR and decreased FCG. Feeding with HPA replacement SBM 21d (T3) and T3 plus BMD (T6) showed a significant (P<0.05) improvement in FCR and a significant decrease in FCG. The results of this study further indicated that replacement of 5% HPA from SBM in the pre-starter (1-10d) showed that all performance was comparable with no significant (P>0.05) differences with BMD (T4), but there was significantly (P<0.05) improved FCR and reduction of FCG with 5% HPA replacement SBM in both the pre-starter and starter periods for 1-21d (T3) compared to the BMD as well as the other supplementation with BMD and the control diet. These findings suggest that a well-formulated diet rich in high-quality protein sources (HPA) best serves the nutritional needs of the birds while also fostering healthy digestive tracts, which are essential in any antibiotic-free (ABF) program.

During the finisher phase (36-42d of age), the findings of this study showed a similar pattern to the grower period. However, there were no statistically significant changes (P>0.05) seen in the productive performance measures, including BW, BWG, FI, FCR, SR, PI and FCG. During this period, it was observed that feeding with HPA 5% (T2) resulted in the best performance in terms of FCR, PI, and FCG compared to other treatments. Although there were no statistically significant differences (P>0.05) in the productive performance indicators, there was an improvement when the control diet was supplemented with HPA or BMD, or a combination of HPA and BMD.

During the whole research period (1-42d of age), the results indicated that supplementing with HPA, BMD, or a combination of both increased the overall productivity. Feeding HPA 5% during the pre-starter and starter periods 1-21d (T3) showed the highest productive performance with a significant (P<0.05) improvement in the value of FCR compared to the control group; it, also showed the lowest FCG, but this was not significantly different (P>0.05). Birds fed HPA 5% in the diet 1-21d (T3) demonstrated superior results, while there was no statistically significant difference (P>0.05) in terms of BWG, FCR, SR and PI compared to the BMD (T4). The results indicated that supplementation of 5% HPA replacement SBM in the study period of 21d showed no need to supplement AGPs for the overall period of feeding and showed better results but did not differ significantly (P>0.05) on BWG, FCR, SR and PI.

Economic benefit

This study demonstrated that feeding HPA 5% replacement SBM in the control diet (T3) during the early 1-21d of age showed evidence of economic benefit return (EBR; Table 4). The 21d feeding with HPA 5% replacement SBM in the control diet for 21d (T3) showed the highest return on investment at 6.54baht/bird or 2.60baht/kg over the control

Table 3: Effects of HP AviStart in the diets on growth performance for four phases of feeding diets and the overall period from 1to 42d of age

	Treatment	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
1-10 days	Initial weight (g)	41.40	41.56	41.40	41.253	41.18	41.12	0.16	0.60
	BW)g(	235.63	232.98	231.93	240.33	233.73	234.67	7.60	0.49
	BWG)g(	194.23	191.42	190.53	199.08	192.55	193.55	7.61	0.47
	FI)g(	253.34	238.30	237.50	251.87	241.51	241.35	10.78	0.06
	FCR	1.31a	1.25b	1.25b	1.26ab	1.26b	1.25b	0.04	0.05
	%SR	99.33	100.00	100.00	100.00	99.33	100.00	0.94	0.56
	PI	148.12	153.77	152.89	157.59	152.65	155.25	8.27	0.51
	FCG) Baht/kg BW(	23.37	23.52	23.55	22.72	23.87	23.56	0.84	0.28
11-21 days	Initial weight (g)	235.63	232.98	231.93	240.33	233.73	234.67	7.60	0.49
	BW)g(	790.00	780.33	805.00	798.00	789.00	783.33	72.28	0.99
	BWG)g(	554.37	547.36	573.07	557.67	555.27	548.67	69.25	0.99
	FI)g(	886.55	856.75	783.01	777.11	876.60	789.15	105.96	0.17
	FCR	1.60a	1.57a	1.37b	140.b	1.59a	1.45ab	0.13	0.00
	%SR	99.28	99.28	100.00	100.00	100.00	99.33	1.22	0.70
	PI	344.34	349.21	420.10	400.24	363.81	380.97	69.35	0.28
	FCG) Baht/kg BW(	28.86a	28.25ab	25.82ab	25.06b	28.54a	27.51ab	2.51	0.03
22-35 days	Initial weight (g)	790.00	780.33	805.00	798.00	789.00	783.33	72.28	0.99
	BW)g(	2022.70	1970.70	2048.00	2046.00	2029.30	1991.30	180.62	0.97
	BWG)g(	1232.67	1190.33	1243.00	1248.00	1240.33	1208.00	132.42	0.97
	FI)g(	2130.20	2011.70	2074.30	2142.70	2087.60	2004.10	231.36	0.86
	FCR	1.73a	1.69ab	1.67b	1.72a	1.68ab	1.66b	0.04	0.02
	%SR	94.93	96.38	95.65	94.20	94.93	94.93	3.61	0.93
	PI	485.33	485.51	510.02	488.02	499.41	495.90	61.70	0.98
	FCG) Baht/kg BW(	29.62a	28.95ab	28.59b	29.59a	28.98ab	28.59b	0.64	0.02
36-42 days	Initial weight (g)	2022.70	1964.00	2034.70	2046.00	2029.30	1994.50	180.07	0.97
	BW)g(	2508.00	2472.00	2556.00	2522.70	2538.70	2474.50	219.68	0.98
	BWG)g(	485.33	508.00	521.33	476.67	509.33	480.00	74.05	0.86
	FI)g(	945.03	931.93	998.47	915.50	986.17	904.70	90.42	0.40
	FCR	1.96	1.89	1.92	1.94	1.95	1.88	0.18	0.96
	%SR	98.55	99.28	100.00	100.00	100.00	99.28	1.78	0.65
	PI	353.65	402.78	389.53	359.60	377.14	356.28	94.45	0.94
	FCG) Baht/kg BW(	33.65	32.01	32.29	32.68	32.77	31.99	3.37	0.96
1-42 days	Initial weight (g)	41.40	41.56	41.40	41.25	41.18	41.12	0.47	0.60
	BW)g(	2474.70	2455.30	2556.00	2522.70	2538.70	2462.00	226.75	0.95
	BWG)g(	2433.30	2413.80	2514.60	2481.40	2497.50	2433.40	223.67	0.96
	FI)g(	4215.10	4072.00	4093.30	4087.20	4303.70	3956.60	348.75	0.61
	FCR	1.74a	1.69abc	1.63c	1.65bc	1.73ab	1.64c	0.07	0.03
	%SR	92.67	96.00	96.00	94.67	94.67	94.00	3.50	0.57
	PI	312.00	328.37	353.93	339.31	326.57	332.72	43.29	0.68
	FCG) Baht/kg BW(	32.79	31.18	30.45	30.44	32.33	31.36	1.98	0.24

Means within row with no common alphabets differ significantly (P≤0.05). Productive index)PI)=(BWG)kg (x Survival rate ((%)x 100)/Age x FCR. Feed cost per gain)FCG1=((FCR x Feed cost x 100)/Survival rate (%).

group (T1), the second is BMD treatment (T4) with 5.81baht/bird or 2.34baht/kg, the third is HPA 5% replacement SBM for 10d (T2) with 3.45 baht/bird or 1.43baht/kg, the fourth is HPA 5% replacement SBM for 21d (T6) with 3.18 baht/bird or 1.31baht/kg, and the fifth is HPA 5% replacement SBM for 10d (T5) with 1.5 baht/bird or 0.61 baht/kg compared with the control (T1) group.

Nutrients digestibility

Table 5 shows how feed affects 21d old broilers' nutritional digestibility. The study found no significant changes (P>0.05) in digestibility of DM and CF, however broilers fed HPA or HPA with BMD had considerably (P<0.05) improved digestibility of CP, EE, Ca, and P compared to the control group.

Carcass characteristics and carcass quality

Table 6 and 7 display the impact of dietary supplements on the physical parameters of the carcass and the weight of organs in broilers that are 42d old. There were no statistically significant changes (P>0.05) found in terms of carcass quality. The white striping of breast meat and footpad lesion scores (Table 5) showed strong evidence of benefit return. The study found that feeding with HPA 5% with or without BMD resulted in a highly significant decrease in white striping (P<0.05).

Table 4: Effects of HP AviStart in the diets on economic benefits return for overall 42d of age

Treatments	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
FCG2[1]) Baht/bird(	79.33	75.01	76.46	75.70	80.70	75.60	6.45	0.57
SBR[2]) Baht/bird(	109.50	108.62	113.16	111.67	112.39	108.94	10.20	0.95
NPR1[3])Baht/bird(	30.16	33.61	36.70	35.97	31.69	33.34	6.74	0.55
NPR2[4]) Baht/kg(	12.21	13.82	14.55	14.56	12.67	13.64	1.98	0.24
ROI1[5]) Baht/bird(	-	3.45	6.54	5.81	1.53	3.18	-	-
ROI2[6]) Baht/kg(	-	1.43	2.60	2.34	0.61	1.31	-	-

All costs were shown in Thailand bath; 1 USD=33.82 bath. Means within row with no common superscript differ significantly (P≤0.05). [1]Feed cost per gain)FCG2=(FCG1 x BWG)kg(. [2]Salable bird return)SBR=(Price of live chicken)36 Baht (x BW)g.([3]Net profits return per bird)NPR1=(SBR - FCG2. [4]NPR2=NPR1/BWG)kg.([5]Return on investment by comparing with and the control group)ROI1=(NPR1 (group test) –NPR1) control.([6]ROI2=NPR2 (group test) –NPR2) control.(*Selling price 45 baht/kg.

Table 5: Effects of HP AviStart in broiler diets on nutrient digestibility at 21d of age

Digestibility (%)	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
DM	81.09	81.19	81.38	81.19	81.31	81.46	0.38	0.59
CP	85.43^{d}	87.40^{bc}	89.30^{a}	85.92^{cd}	87.19^{bc}	88.64^{ab}	1.21	0.0001
EE	89.03^{b}	89.97^{a}	89.95^{a}	89.32^{ab}	89.61^{ab}	89.59^{ab}	0.55	0.04
CF	60.26	60.29	60.45	60.42	60.43	60.88	0.38	0.09
Ca	56.10^{bc}	57.78^{b}	60.49^{a}	55.96^{c}	57.06^{bc}	60.04^{a}	1.42	0.0001
P	51.29^{c}	53.20^{b}	56.80^{a}	52.58^{b}	52.44^{b}	56.99^{a}	0.65	0.0001

Means within row with no common superscript differ significant ($P \leq 0.05$).

Table 6: Effects of HP AviStart in the diets on carcass quality at 42d of age

Treatment	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
Dressing percentage[1]	72.91	72.27	72.46	72.94	73.28	72.30	1.47	0.79
Breast%) BW(	21.76	20.79	21.80	21.88	21.35	22.08	1.23	0.51
Drumstick +Thigh %)BW(	22.73	22.31	22.64	22.92	22.84	22.14	1.24	0.87
Wing %)BW(	7.75	8.30	7.83	7.67	8.09	7.84	0.68	0.61
Edible Meat%) [2]BW(	52.23	51.40	52.27	52.46	52.27	52.07	1.29	0.77
Abdominal fat %)BW(	1.65	1.78	1.58	1.66	1.81	1.57	0.33	0.75

Means within row with no common superscript differ significantly ($P \leq 0.05$); [1]Dressing percentage=Dressed bird or New York Cut – head and neck – all visceral organs – shanks and feet[2] ;Edible meat=Breast +Drumstick +Thigh+Wing.

Table 7: Effects of HP AviStart in the diets on organ weight, footpad lesion score and white stripe score at 42d of age

Treatment	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
Heart (%BW)	0.46	0.45	0.47	0.47	0.50	0.46	0.05	0.59
Liver (%BW)	2.05	2.02	2.04	2.04	2.11	2.106	0.25	0.98
Pancreas (%BW)	0.19	0.20	0.20	0.21	0.22	0.223	0.03	0.53
Spleen (%BW)	0.17	0.14	0.18	0.20	0.19	0.192	0.05	0.18
Gizzard (%BW)	1.43	1.34	1.45	1.49	1.31	1.357	0.13	0.13
Total visceral organs	4.29	4.14	4.34	4.41	4.33	4.340	0.26	0.59
Footpad lesion score[1]	1.42	1.11	1.15	1.67	1.21	0.72	0.54	0.12
White stripe score[2]	2.01^{a}	1.79^{c}	1.75^{c}	1.97^{ab}	1.90^{abc}	1.82^{bc}	0.13	0.01

Means within row with no common superscript differ significantly ($P \leq 0.05$); [1]Footpad lesion score, using a scoring system: 0-0.75=no lesions or black spots on footpads, 1.0-1.75=a few lesions or black spots on footpads (<25%), 2.0-2.75=a moderate number of lesions or black spots on footpads (25-50%) and 3.0-3.75=acute lesions or black spots on footpads (>50%) (Martrenchar et al. 2001).

Table 8: Effects of HP AviStart in the diets on fat deposition in small intestine appearance score at 21d and 42d of age

Treatment	T1 (Control)	T2 (HPA 10d)	T3 (HPA 21d)	T4 (BMD)	T5 (CMD 10d)	T6 (CMD 21d)	SEM	P-value
Fat deposition in small intestine score:								
21 days	2.180^{c}	2.603^{abc}	2.772^{abc}	2.328^{bc}	2.943^{ab}	3.120^{a}	0.495	0.0205
42 days	5.975^{b}	8.392^{a}	8.625^{a}	6.217^{b}	8.400^{a}	8.792^{a}	0.605	0.0001

Means within row with no common superscript differ significantly ($P \leq 0.05$).

Additionally, there was an increase in footpad lesion scores in the control group, although this increase was not statistically significant (P>0.05).

Table 8 displays the impact of dietary interventions on the accumulation of fat in the small intestine at 21 and 42d of age. With regard to feeding with HPA 5% replacement SBM during 1-21d of age in combination with BMD (T6), the appearance score at 21d of age showed significantly (P<0.05) increased fat deposition in the small intestine score compared to the control and BMD supplemented diets. The appearance score at 42d of age showed significantly (P<0.05) increased fat deposition in the small intestine score when supplemented with HPA 5% or in combination with BMD compared to the control and BMD-supplemented diets.

DISCUSSION

In the current study, it was speculated that the efficacy of the HPA with low ANFs contained lower amounts of oligosaccharide and antigen substances (Table 2). That indicated a well-formulated diet with high-quality protein sources. The HPA is the most suitable option for fulfilling the birds' nutritional needs while also supporting a healthy digestive tract, which is crucial under antibiotic-free (ABF) conditions. The HPA is one of the SBM enzyme fermentation technologies plus inactivated yeast. The protein in fermented feed enhances nutritional value and health in an animal, reducing the use of antibiotics, acidifiers, and toxin binder (Hidalgo and Brunsgaard 2019; Somer 2019). The findings of this study suggest that supplementing with HPA 5% replacement SBM over a period of 21d did not require additional supplementation with AGPs during the whole feeding time. Furthermore, the supplementation demonstrated slight improvements, albeit not statistically significant (P>0.05), in terms of BWG, FCR, SR and PI. The results from this study showed that efficient broiler production without AGPs is possible with a well-formulated diet of high-quality plant protein that best meets the nutritional requirements of the birds, while promoting a healthy intestinal tract is imperative in any antibiotic-free environment, and more is also needed to ensure maximum digestibility of the feed. According to Zhang et al. ((2021, who reported that broiler body weight and feed intake were increased when supplemented with soy protein concentrate at 4and %8in broiler feed during the starter phase (10-1d). Similarly, incorporation of 7.7 10.0 and 12.5% SPC in prestarter (0-10d) diets increased growth performance (Kiarie et al. 2021).

The digestibility of nutrients was considered an indicator of nutrient absorption in the animals and substantiated their productive performance. In this study, the results showed better significant digestibility of CP, EE, Ca and P when broilers fed with HPA or HPA plus BMD in the diets compared no-supplemented group. In their study, Wang et al. (2011) found that the inclusion of enzymolytic soybean meal (ESBM) at a rate of 100g/kg in the diets of developing broilers resulted in the greatest improvement in the digestion of nutrients such as dry matter, crude protein, energy, calcium, and phosphorus. Broilers' apparent nitrogen digestion and truly metabolizable energy were found to be higher when fed soybean meal that had been treated with protease (Ghazi et al. 2002, 2003). Similarly, the results of enzyme-treated soybean meal supplemented in piglet diets showed better apparent ileal digestibility of total nitrogen Histidine and Tryptophan (Li et al. 2021).

These are several facts that determine the recondition of visual quality by consumers. The amount of fat meat texture and skin color can also be one of those factors. Footpad lesions (pododermatitis or footpad dermatitis, FPD) are an important aspect of the broiler business, both from an economic and a welfare point of view. It affects growth performance, walking ability, and carcass revenues. Chicken paws have recently become the third most significant commercial component of chickens, behind the breast and wings. The chicken sector is concerned about the lesions induced by FPD because to their impact on animal welfare, food safety, and product degradation issue (Shepherd and Fairchild 2010). These data suggest that sticky indigestible carbohydrates derived from plant sources, particularly soybean meal (SBM), may have a corrosive effect and contribute to FPD (Hess et al. 2004; Blanch 2020). The study findings indicate that HPA is a high-quality protein source with little non-starch polysaccharide (NSP) content, resulting in a reduction in FPD lesions (Table 7). According to Amer (2020), the incorporation of enzymes in chicken feed that are capable of breaking down non-starch polysaccharides has the ability to reduce the quantity of moisture present in the litter as well as the frequency and severity of FPD. There was a significant increase ($P<0.05$) in fat deposition in the small intestinal score when supplementing with either 5% HPA or BMD in conjunction with the control and BMD groups. The results of this study found that HPA is one of the SBM enzyme fermentation technologies plus inactivated yeast, showed high quality protein with low ANF activities, contained a low amount of oligosaccharide and NSP with a higher ME value, and showed higher digestibility of nutrients in the diet than SBM. The findings of the current investigation indicate that a well-designed diet consisting of superior components, particularly protein and cereal, is crucial in ensuring optimal nutrition for birds and maintaining a healthy digestive system, especially in the absence of antibiotics (ABF). Simultaneously, consuming an excessive amount of energy might lead to an increase in the accumulation of fat (Ahiwe et al. 2019).

Conclusion

The results of this study clearly indicate that feeding HPA replacement SBM during the pre-starter period (1-10d) or both the pre-starter and starter periods (1-21d) resulted in improved performance throughout the entire study period (1-42d). This improvement was observed in terms of higher BWG, PI, and ROI. Additionally, there were beneficial enhancements in carcass quality, reduced white striping of breast meat, and improved footpad lesion scores. Moreover, increasing nutrient digestibility of crude protein (CP), ether extract (EE), calcium (Ca), and phosphorus (P) showed better results at 21d when compared with the control without supplementation and the AGP BMD for the whole period of this study.

Acknowledgement

The authors would like to express their appreciation to Hamlet Protein A/S Denmark for providing the HP AviStart and funds that were used for conducting this experiment. Furthermore, we would like to thank Khon Kaen University and the Department of Animal Science, Non-Ruminant Nutrition team for their facility support both in the course and Non-Ruminant Nutrition laboratory.

Conflict of interest

The authors declare that this study received funding from Hamlet Protein A/S Denmark. The funder was not involved in the study design, collection, analysis, interpretation of data, the writing of this article or the decision to submit it for publication.

Author contributions

Conceptualization, Tanaphoom Boonmee, Winai Jaikan, Christine Brokner, Alfred Blanch, Jowaman Khajarern; methodology, Tanaphoom Boonmee, Winai Jaikan; writing-original draft preparation, Tanaphoom Boonmee, Winai Jaikan; writing-review and editing, Tanaphoom Boonmee, Winai Jaikan, Christine Brokner, Alfred Blanch, Jowaman Khajarern. All authors have read and agreed to the published version of the manuscript.

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