Effects of toasting, inclusion levels and different enzyme supplementations of faba beans on growth performance of broiler chickens

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Primary Audience: Nutritionists, Researchers, Live Production Managers

SUMMARY

Faba beans (*Vicia faba*) are an alternative protein source that likely can be used to a higher extent in broiler diets. White-flowered faba beans contain antinutritional substances (ANS) such as non-starch polysaccharides (NSP), trypsin inhibitors, and lectins, which might limit its inclusion level. Lectins and trypsin inhibitors are heat labile and previous studies have shown that steam-pelleting and enzyme treatment improves the nutritional value of faba beans. However, alternative to pelleting would facilitate for farmers to add faba beans on-farm. Currently, there are machines available for toasting faba beans on-farm, which might be used for broiler mash diets. The objective of this study was to investigate the effect of inclusion level (0, 10, 20 and 30%), toasting (140°C 5.5 min) and different enzymes (xylanase + phytase *vs.* xylanase, phytase, amylase, protease) of faba bean diets on growth performance and organ parameters in broilers. To test this, 2 experiments 34 and 35 days, using a total of 480 chickens were performed. Feed intake, body weight (BW) and feed conversion ratio were registered weekly, in addition, organ and carcass weights were registered at slaughter. The results showed that inclusion of 20% faba beans is possible in a pelleted diet with maintained broiler growth performance. When 20% was included in a mash diet, feed intake and BW decreased compared to chickens fed pelleted diets, irrespectively of pre-toasting of the beans. It can be concluded that toasting cannot replace pelleting. Supplementation of protease and amylase in addition to xylanase and phytase did not improve the nutritional value of faba beans.

Key words: faba beans, toasting, feed structure, protease, amylase

2017 J. Appl. Poult. Res. 26:467–475
http://dx.doi.org/10.3382/japr/pfx016

DESCRIPTION OF THE PROBLEM

About 70% of the protein feed used in the European Union (**EU**) for livestock production is imported. To address EU's protein deficit, the European Parliament adopted a resolution in 2011, stating that urgent action is needed to replace imported protein crops with European sources. Since then, a wide range of actions have been taken to increase the production of protein crops within EU [1]. Faba beans (*Vicia faba* minor.) is a crop with reasonable high crude protein (**CP**) content (~30%) [2] that can be grown in Sweden and throughout Europe. It likely has a

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potential to be used in a higher extent in broiler chicken production. As most plant based protein crops, faba beans contain antinutritional substances (**ANS**) that might limit the possible inclusion level in broiler diets. Tannins, vicine, convicine, lectins and trypsin inhibitors are ANS that are associated with faba beans [3]. Tannins are known to decrease both protein and energy digestibility [4], but have through plant breeding been reduced in white-flowered cultivars and is not an issue when these are used. Lectins and trypsin inhibitors (**TI**) may impair the protein utilization, and trypsin inhibitors might also cause an overactive and thereby enlarged pancreas [5]. Vicine and convicine may impair egg production and increase the liver weight in laying hens [6]. However, lectins and trypsin inhibitors are heat labile and the levels of these are substantially lower in faba beans than in soybean meal. Valdebous [7] reported that TI activity in faba beans was 7% of that of defatted soybean, and the corresponding value for lectins was 2% of that for defatted soybean. Moreover, the negative effect of vicine and convicine does not seem to be of major concern in growing animals [4], so if the feed is heat treated, lectins and trypsin inhibitors should not be of major concern in broiler diets. But still, the amount of white-flowered faba beans that can be included in a broiler diet does not seem to be unlimited; the current recommended inclusion level is about 20% in pelleted broiler diets [8, 9]. With higher inclusion levels, decreased broiler growth performance has been reported [8]. Moreover, if fed unprocessed in a mash feed, a linear decrease in body weight (**BW**) gain with increasing inclusion level (5-25%) has been observed [10], and the author suggested that this was due to some unknown heat-labile ANS being destroyed during the pelleting. Moreover, Lacassange et al. [11] found that pelleting improves both starch and protein digestibility and thereby the AME_n value of faba beans. However, few farmers have the ability to make investments in pelleting facilities, and use of mash feed can be a way for the farmer to facilitate addition of protein crops on-farm and increase the use of home-grown protein feed. Currently there are small-scale machines available on the market that can be used on-farm to toast faba beans before being milled and mixed in a diet. To the best of our knowledge, no study has been performed that investigates effects on the production performance of broiler chickens fed toasted faba beans included in mash feed, in comparison with pelleted faba bean diets.

Broilers and piglets are the livestock species with the highest CP and amino acid requirements; it is therefore challenging to optimize a balanced diet without use of high-quality protein feedstuff. A way to overcome this challenge is to optimize the use of nitrogen in the feed by increasing the availability and reduce the excretions. Ospina-Rojas et al. [12] showed that in broiler diets, lowering the dietary CP level with 3% while the amino acid level was maintained decreased the nitrogen content in the litter and lowered the ammonia emissions without undermining production performance. Masey O'Neill et al. [13] showed that faba beans compared to soybean meal have a lower digestibility of several amino acids including methionine, cysteine, and threonine, which also means that there is a possibility for improvements in utilization of the amino acids in faba beans. Apart from CP, faba beans contain a considerable amount of starch and non-starch polysaccharides (**NSP**) [3]. About 26% of total NSP in faba beans are soluble NSP [14], which are well known to increase digesta viscosity and in turn decrease the nutrient availability and cause problems such as sticky droppings and wet litter. Supplementations of NSP-degrading enzymes (xylanases) have been shown to increase both energy and protein values of legumes in the same way as in wheat. In addition, enzyme mixtures containing xylanase, amylase, and protease have been shown to further improve the availability of the protein ratio in legumes [15]. The use of different enzyme mixtures can therefore be a strategy to further improve the nutritional value of faba beans.

The objective of the present study was to investigate the effect of inclusion level, toasting, and enzyme supplementation of faba bean diets on growth performance and organ parameters in grower broilers.

MATERIALS AND METHODS

Experimental Design and Diets

Experiment 1 A total of 240 unsexed day-old broiler chickens (Ross 308) were used in a

Table 1. Diet composition (%) and analyzed chemical composition (g/kg).

	Experiment 1						Experiment 2		
	C[1]	FB10[2]	FB20[3]	FB30[4]	FB20M[5]	FB20MT[6]	HP[7]	LP[8]	LP+E[9]
Ingredient									
Wheat	65.9	60.0	53.8	47.3	53.8	53.8	50.3	53.2	53.2
Soybean meal	24.5	20.3	16.2	12.1	16.2	16.2	18.1	15.7	15.7
Faba bean	0	10.0	20.0	30.0	20.0	20.0	20.0	20.0	20.0
Vegetable oil	4.0	4.6	4.7	5.3	4.7	4.7	5.3	5.0	5.0
Limestone	2.1	2.0	2.0	2.1	2.0	2.0	1.9	1.9	1.9
Monocalcium phosphate	1.6	1.6	1.5	1.4	1.5	1.5	1.4	1.5	1.5
L-Lysine- HCl	0.3	0.3	0.3	0.2	0.3	0.3	0.5	0.4	0.4
DL- Methionine	0.2	0.2	0.3	0.3	0.3	0.3	0.2	0.1	0.1
L-Threonine	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Premix[10]	0.8	0.8	0.8	0.8	0.8	0.8	0.7	0.7	0.7
Sodium chloride	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Sodium bicarbonate	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Xylanase + phytase	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	
Protease + xylanse + amylase	.	.	.	.	.	.	.	.	0.03
Phytase	.	.	.	.	.	.	.	.	0.01
Analyzed chemical composition									
Metabolizable energy kcal/kg (calculated)	2,938	2,938	2,938	2,938	2,938	2,938	2,914	2,914	2,914
DM	869	868	867	863	875	877	889	885	884
Ash	62	61	62	59	64	54	65	55	58
CP	193	192	189	200	204	198	199	181	187
CF	34	35	38	45	31	39	47	53	51
EE	48	44	38	47	59	49	63	74	71
Lysine	12.0	11.8	10.7	12.0	10.9	11.0	13.9	13.1	13.6
Methionine	6.5	6.7	6.6	6.5	6.10	5.80	5.5	4.7	5.0
Cysteine	6.3	8.5	7.6	6.3	7.30	8.20	6.8	6.8	6.9
Threonine	7.2	6.8	5.8	6.9	7.90	7.00	7.7	6.5	6.4

C[1] = Control; FB10[2] = Faba bean 10%; FB20[3] = Faba bean 20%; FB30[4] = Faba bean 30%; FB20M[5] = Faba bean 20%, mash; FB20MT[6] = Faba bean 20%, mash toasted; HP[7] = High protein; LP[8] = Low protein; LP+E[9] = Low protein + additional enzymes.

[10]The premix provided (per kg diet): retinyl acetate: 10,000 IU; cholecalciferol: 2,750 IU; dl-α-tocopherol acetate: 40 mg; menadione-nicotinic amide bi-sulphite: 3 mg; thiamin mono-nitrate: 2 mg; riboflavin: 6 mg; pyroxinhydrocloride: 3 mg; cynocobalamin: 0.02 mg; calcium pantothenate: 10 mg; folinic acid: 1.0 mg; nicotinic acid: 35 mg; d-biotin 0.5 mg; choline chloride: 180 mg; betaine anhydrous: 285 mg; Fe: 25 mg; Cu: 6 mg; Mn: 79 mg; Zn: 60 mg; I: 1.0 mg; Se: 0.50.

34-d growth experiment. The chickens were randomly distributed into 30 pens (1.50 × 0.75 m) with 8 chickens/pen and 5 replicates per treatment. The experiment was organized as a randomized block design with 6 dietary treatments to test the effect of inclusion level and toasting. The experimental diets, Table 1, were formulated to meet nutritional requirements of chickens according to NRC [16]. However, no phase feeding was used, and the nutrients were kept constant throughout the study, leading to a lower nutrient content than recommended during the starter phase and a higher during the finisher phase. All diets were supplemented with 2,300 U xylanase [17]and 500 FTU of phytase [18] per kg diet. The control diet (**C**) was based on wheat and soybean meal and in the experimental diets, part of the wheat and soybean meal was substituted with 10 (**FB10**), 20 (**FB20**) or 30% (**FB30**) faba bean of the white-flowered cultivar Tattoo (Table 1). The CP and major amino acids in the faba beans were analyzed and were (g/kg dry matter [**DM**]) CP: 312; methionine: 3.86; cysteine: 0.20; lysine: 19.10 and threonine: 10.56. For apparent metabolizable energy (AME_n), the value 2,761 kcal/kg DM from European table of energy value for poultry feedstuffs [19] was used.

The diets were steam-pelleted (75°C) at a commercial feed mill. Moreover, to test the effect of toasting, 2 mash diets with 20% faba beans were prepared. In one of the diets the faba

beans were untreated (**FB20M**), whereas in the other (**FB20MT**) the beans were toasted (140°C, 5.5 min). In both mash diets, the wheat was kept as whole wheat, whereas the beans and other ingredients where milled through a 3-mm sieve before being mixed in the diets.

Experiment 2 A total of 240 unsexed day-old broiler chickens (Ross 308) were used in a 35-d growth experiment. The chickens were randomly distributed into 30 pens (1.50 × 0.75 m) with 8 chickens per pen and 10 replicates per treatment. The experiment was organized as a randomized block design with 3 dietary treatments to test the effect of different enzymes. All diets contained 20% faba beans. A high protein (**HP**) diet was formulated to meet the nutritional requirements of the chickens according to NRC [16] (Table 1). However, no phase feeding was used, and the nutrients were kept constant throughout the study, leading to a lower nutrient content than recommended during the starter phase and a higher during the finisher phase. The HP diet was supplemented with xylanase (2,300 IU) and phytase (500 FTU) [17, 18]. A low protein (**LP**) diet with a 9% reduction in CP, 6% reduction in lysine, and 15% reduction in methionine and threonine compared to HP was formulated (Table 1). The LP diet contained the same enzymes as the HP diet [17, 18]. The third diet, **LP+E**, was formulated to be nutritionally identical to the LP diet, but in addition to the phytase (500 FTU) [18], an enzyme mixture was used [20] to supply the same amount of xylanase (2,300 IU) as in HP and LP, and in addition, 4,000 IU protease and 400 IU amylase were added. Recovery analyses of enzymes showed expected values, verifying correct enzyme addition. The diets were steam-pelleted (75°C) at a commercial feed mill, the pellets were crushed before fed to the chickens, which was done to distinguish between the effect of physical treatment and the effect of feed structure for results in Experiment 1.

Housing

In both experiments, the chickens were housed in pens raised from the floor, equipped with solid floor covered with fresh wood shavings. Chickens had free access to water and feed throughout the experiment. Room temperature was gradually decreased from 33°C on d 0 to 23°C on d 24 and kept at 23°C until the chickens were euthanized at the end of the trial. Chickens had continuous artificial light for the first 2 d; thereafter, the dark period was gradually increased to 6 h from d 8 to the age of slaughter. The experiments were carried out at The Swedish Livestock Research Center and were approved by the ethical committee of the Uppsala region, approval number C 334/12.

Experimental Procedures and Analyses

In Experiment 1 and 2 feed intake, BW and FCR were registered per pen on a weekly basis. FCR was corrected for mortality. At d 34 (Experiment 1) and d 35 (Experiment 2), individual BW and sex (visually determined based on exterior appearance) were registered and CV of BW within pen was calculated as CV % = (Standard deviation/mean BW) × 100. Sex ratio was calculated as: Number of rooster/total number of bird in pen. Occurrence of sticky droppings (scale 0–1; where 0 is no occurrence and 1 is occurrence) was registered on pen basis at d 7 and 14. Excreta samples were collected for DM analysis at d 12, 22, and 33 in both experiments by covering the litter area with plastic foil and collect the excreta during 2 h. At d 35 in Experiment 1, one hen and one rooster representative for the pen (visually determined based on exterior appearance) were selected and killed by an intravenous injection of sodium pentobarbital through the wing vein. The BW and weight of liver and pancreas, respectively, were recorded. Moreover, the foot pad lesion was scored according to Ekstrand et al. [21]. At d 36 in Experiment 2, one hen and one rooster representative for the pen were selected and brought to the research slaughter facility. The individual BW was recorded before the chickens were killed. Chickens were individually stunned by electricity, killed by bleeding, and then scaled in hot water and de-feathered by a machine. The liver weight and pancreas weight were recorded before all organs, head, feet, and neck were removed and hot carcass weight recorded. The breast meat and drumstick were cut out according to Hudspeth et al. [22] and their weights were recorded. In addition, foot pad lesions were scored in accordance with Ekstrand et al. [21].

Feed and excreta samples were analyzed for DM by drying at 103°C for 16 h and ash of feed samples after ignition at 600°C for 3 h [23]. The CP (N × 6.25) in the feed was determined by the Kjeldahl method [24]. The EE of feed was determined according to Official Journal of European Communities [25]. Amino acids were analyzed with the Waters AccQ Tag^{TM}method as described by Langeland et al. [26].

Statistical Analyses

Statistical analyses were performed with the GLM procedure in SAS [27] to determine treatment effects by one-way analysis of variance (**ANOVA**). The model included diet as a fixed factor, the effect of sex ratio was tested in the models, but without significant effect and was therefore excluded. Pen served as experimental unit for performance data, sticky droppings, and DM of excreta. Since DM of excreta was determined 3 times (d 11, 22, and 33), the diet effect was analyzed with Proc Mixed and a repeated statement with unstructured covariance, pen served as random factor. For relative carcass traits and organ weights, the individual bird was considered as the experimental unit and dietary treatment and sex were considered as fixed factors. The sticky droppings data was arcsin-root transformed before analysis [28]. All differences were considered significant at $P \leq 0.05$.

RESULTS AND DISCUSSION

Effect of Inclusion Level and Toasting

Experiment 1 showed that inclusion of 20% faba beans in a pelleted diet was possible without affecting production performance negatively compared to the control diet. With 30% inclusion, feed intake and BW were impaired, whereas the FCR was improved compared to C and FB20, showing that the lower feed intake was responsible for the decreased BW. Moreover, a very low feed intake and consequently low BW were observed when the faba beans were included in mash diet, independently of pre-toasting of the beans (Table 2). The chickens fed the mash diets also had a higher relative pancreas weight than chickens fed pelleted diets, independently of pre-toasting. No effect of dietary treatment on liver weight was observed (Table 3). The CV in BW ranged between 13.1 to 16.4% for all treatments except for chickens fed FB20M that had a CV of 22.3%, which was about 7% higher than FB20MT and 8% higher than the control. The results show that inclusion of 20% white-flowered faba beans in a diet is possible without interfering with broilers production performance on the condition that the diet is pelleted and not fed as mash. Maintained growth performance with 20% inclusion is in agreement with Nalle et al. [9]. Similarly, Farell et al. [8] showed a curve linear response in chicken performance with increasing levels of faba beans, with the best performance at an inclusion of 18% faba beans and a decreased performance with 24 and 36% faba beans. In the present study and in the study of Farrell et al. [8], diets were optimized on total CP and amino acid content. A lower digestibility of several amino acids including methionine, cysteine and threonine in faba beans compared to soybean meal was proved by Masey O'Neill et al. [13]. Therefore, a lower level of available amino acids might explain the decreased growth rate with 30% faba bean inclusion. The lower growth performance observed on the mash diets is in agreement with Gous [10]. In the study by Gous [10], it was hypothesized that the impaired performance in broilers fed mash diets was due to an unknown heat labile ANS, that got destroyed in the pelleting process. The higher relative pancreas weight in the present study on mash diets indicates a higher activity of pancreatic enzymes. However, in the present study no differences in growth performance or pancreas weight between the toasted and non-toasted faba bean groups were observed. This suggests that the difference was not caused by a heat labile ANS, instead it seems like the pelleting process per se has a beneficial effect. It is known that steam-pelleting increases both starch and protein digestibility in faba beans [11]. One explanation for this is that during steam-pelleting, starch is gelatinized. For this to happen, a sufficient amount of water has to be present during the feed processing [29]. The toasting was performed without addition of water which might explain why toasting was not able to improve the nutritional value in the same way as pelleting. However, increased within group variation

Table 2. Weekly broiler chicken production performance, accumulated BW, feed intake (FI) and calculated FCR in Experiment 1 and 2. Least square means ± pooled SEM.

	Experiment 1								Experiment 2				
Item	C[1]	FB10[2]	FB20[3]	FB30[4]	FB20M[5]	FB20MT[6]	Pooled SEM	*P*-value	HP[7]	LP[8]	LP+E[9]	Pooled SEM	*P*-value
BW (g)													
d 7	152^{b}	148^{b,c}	174^{a}	148^{b,c}	135^{c,d}	136^{c,d}	4.91	<0.0001	143	134	134	3.26	0.108
d 14	439^{b}	417^{b}	518^{a}	388^{b}	296^{c}	300^{c}	19.73	<0.0001	337	304	317	11.17	0.160
d 21	898^{b}	847^{b}	1019^{a}	794^{b}	591^{c}	592^{c}	37.49	<0.0001	752	646	648	35.50	0.085
d 28	1,511^{b}	1,423^{b,c}	1,681^{a}	1,372^{c}	1,018^{d}	1,037^{d}	48.89	<0.0001	1,257	1,154	1,185	34.57	0.135
d 34 (35)[10]	2,098^{a}	2,003^{a,b}	2,167^{a}	1,900^{b}	1,478^{c}	1,544^{c}	66.91	<0.0001	1,959^{A}	1,770^{B}	1,787^{B}	42.35	0.012
FI (g)													
d 7	198^{b,c}	202^{b}	223^{a}	203^{b}	194^{b,c}	202^{b}	4.87	0.001	200	188	194	6.63	0.435
d 14	592^{b}	565^{b,c}	677^{a}	518^{c}	453^{d}	468^{d}	17.27	<0.0001	474	477	493	10.28	0.329
d 21	1,290^{b}	1,211^{b,c}	1,419^{a}	1,124^{c}	949^{d}	954^{d}	37.18	<0.0001	1,025	982	1,034	25.24	0.299
d 28	2,306^{b}	2,141^{b,c}	2,565^{a}	2,020^{c}	1,667^{d}	1,679^{d}	65.12	<0.0001	1,959	1,813	1,837	51.41	0.134
d 34 (35)[10]	3,641^{a}	3,214^{b}	3,726^{a}	2,951^{b}	2,524^{c}	2,562^{c}	115.68	<0.0001	3,170^{A}	2,940^{B}	2,950^{B}	66.18	0.045
FCR													
d 7	1.31	1.38	1.28	1.38	1.44	1.49	0.058	0.0890	1.42	1.42	1.46	0.055	0.844
d 14	1.35^{a}	1.36^{a}	1.31^{a}	1.34^{a}	1.57^{b}	1.57^{b}	0.071	<0.0001	1.42^{B}	1.57^{A}	1.56^{A}	0.036	0.015
d 21	1.44^{a}	1.43^{a}	1.39^{a}	1.42^{a}	1.63^{b}	1.62^{b}	0.063	<0.0001	1.40^{B}	1.54^{A,B}	1.59^{A}	0.050	0.038
d 28	1.52^{a,b}	1.51^{a,b}	1.53$^{a–c}$	1.47^{a}	1.64^{c}	1.62^{b,c}	0.045	<0.0001	1.57	1.57	1.55	0.045	0.945
d 34 (35)[10]	1.73^{b,c}	1.61^{a,b}	1.73^{b,c}	1.55^{a}	1.71^{b,c}	1.66^{a,b}	0.050	0.0089	1.62	1.66	1.65	0.021	0.368

C[1] = Control; FB10[2] = Faba bean 10%; FB20[3] = Faba bean 20%; FB30[4] = Faba bean 30%; FB20M[5] = Faba bean 20%, mash; FB20MT[6] = Faba bean 20%, mash toasted; HP[7] = High protein; LP[8] = Low protein; LP+E[9] = Low protein + additional enzymes. [10] = d 34 Experiment 1, d 35 Experiment 2.

$^{a–d}$Least square means within the same row (Experiment 1) with different superscripts were significantly different ($P < 0.05$).

^{AB}Least square means within the same row (Experiment 2) with different superscripts were significantly different ($P < 0.05$).

Table 3. Relative organ weights, sticky droppings DM – excreta and foot pad score, and carcass characteristics. Least square means ± pooled SEM.

	Experiment 1							Experiment 2				
	C[1]	FB10[2]	FB20[3]	FB30[4]	FB20M[5]	FB20MT[6]	Pooled SEM	*P*-value	HP[7]	LP[8]	LP+E[9]	Pooled SEM	*P*-value
Relative pancreas weight (g/kg BW)	2.06[c]	2.32[b,c]	2.26[b,c]	2.16[c]	2.68[a]	2.51[a,b]	0.096	<0.0001	1.7	1.6	1.8	0.08	0.264
Relative liver weight (g/kg BW)	30.48	28.84	27.92	28.27	30.78	29.41	1.124	0.4809	31.3	16.7	29.2	5.85	0.199
Sticky droppings % affected d 7	11.0	16.0	12.8	10.7	19.0	4.0	4.87	0.3385	18.3	19.4	19.4	0.39	0.098
Sticky droppings % affected d 14	13.0	1.7	13.5	10.67	11.0	6.3	4.39	0.2177	15.5	12.5	12.5	2.50	0.528
DM excreta %	22.9[c]	22.9[c]	24.5[c]	24.11[c]	29.2[a]	30.0[a]	0.605	<0.0001	22.3[B]	23.2[A]	23.1[A]	0.31	0.006
Carcass characteristics													
Carcass weight (CW) % (CW/BW)	.	.	.	.	.	.	.	.	73.0[A]	71.7[B]	70.8[B]	0.41	0.002
Relative breast meat weight % (breast meat/CW)	.	.	.	.	.	.	.	.	25.4	23.7	23.6	0.58	0.054
Relative drumstick weight% (drumstick/CW)	.	.	.	.	.	.	.	.	14.9	14.6	15.0	0.20	0.431

C[1] = Control; FB10[2] = Faba bean 10%; FB20[3] = Faba bean 20%; FB30[4] = Faba bean 30%; FB20M[5] = Faba bean 20%, mash; FB20MT[6] = Faba bean 20%, mash toasted; HP[7] = High protein; LP[8] = Low protein; LP+E[9] = Low protein + additional enzymes.

[a–d]Least square means within the same row (Experiment 1) with different superscripts were significantly different ($P < 0.05$).

[AB]Least square means within the same row (Experiment 2) with different superscripts were significantly different ($P < 0.05$).

in body weight is a sign of specific nutrient deficiency [30]. The higher CV in the non-toasted group indicate that the toasting might improve the nutrient availability in some extent, although the low feed intake limits the use of faba beans in mash diets.

Effect of Enzymes

Experiment 2 showed no differences between LP and LP+E for any parameter, but chickens fed HP had both higher BW and feed intake at d 35 than chickens fed the LP diets. Moreover, the carcass weight was higher and relative breast meat weight tended to be higher in chickens fed HP diets, whereas the excreta DM was lower in chickens fed HP compared to LP diets. The decreased performance in chickens fed the LP diets confirms that they were fed below their nutrient requirement and that supplementation of protease and amylase in addition to xylanase and phytase did not improve the nutritional value of faba beans.

The lack of effect of additional enzyme supplementation is in agreement with Kalmendal and Tauson [31], who did not find any additional effect of the combination of xylanase and protease compared to when the enzymes were fed as monocomponent enzymes in wheat-soybean meal based diets. Masey O'Neill et al. [13] showed that the standardized ileal digestibility (**SID**) of methionine in Tattoo is about 60%, which means that about 40% is unused, leading to a large potential for improvements. To get an effect of an enzyme, there has to be specificity between the substrate and the enzyme [32]. Ghazi et al. [33] showed different animal response using protease from different microorganisms in soybean meal diets. To the best of our knowledge, there are no published studies that test the specificity between faba beans and different proteases; such a study would be very valuable in the future.

Effect of Feed Structure

To distinguish between the effect of pelleting and the effect of feed structure, the pelleted diets were fed intact in Experiment 1 and crushed in Experiment 2. FB20, FB20M, FB20MT in Experiment 1 and HP in Experiment 2 did all

have very similar ingredient and dietary composition. A comparison in feed intake at d 34 between FB20 (pelleted feed) HP (crushed pellet) and FBMT (mash feed) shows a linear decrease ($r^2 = 0.55$) from pelleted to mash feed. The feed intake was about 15% lower in chickens fed crushed pelleted feed and about 30% lower in chickens fed mash feed compared to the pelleted feed. The feed structure did also have an effect on excreta DM with higher values in chickens fed mash diets compared to pelleted diets, however no remarks on foot fad score were observed in any treatment. Moreover, no differences in occurrence of sticky droppings were observed in either Experiment 1 or 2 (Table 3). This indicates that the major effect of feed structure is on feed intake.

It is well known that feed intake is highly related to feed structure and that birds have a higher feed intake on pelleted compared to mash feed. However, the decrease in feed intake on mash diets was higher than expected. Between 8 to 25% higher intake on pelleted compared to mash diets have been reported in previous studies [34, 35]. The lower feed intake on mash compared to the crushed pellet observed in the present study indicate that the pelleting process per se has an important effect also on the palatability of faba beans. The feed intake can apart from affecting BW also influence the excreta moisture content and an increased excreta moisture content has previously been linked to increased feed intake [36]. The observed lower excreta DM in chickens fed pelleted compared to mash diets is likely due to a higher feed intake, but since no effects were observed on the foot pad; this is judged to be of minor importance for animal welfare.

CONCLUSIONS AND APPLICATIONS

1. Inclusion of 20% white-flowered faba beans in a pelleted diet is possible without interfering with broilers production performance.
2. When 20% faba bean was included in a mash feed, adverse effect on feed intake was observed, irrespectively of pre-toasting, and it can be concluded that toasting cannot replace pelleting.
3. Supplementation of protease and amylase in addition to xylanase and phytase did not improve the nutritional value of faba beans. Studies that investigate the specificity between enzyme and substrate are needed.

REFERENCES AND NOTES

1. Schreuder, R., and C. de Visser. 2014. Report EIP-AGRI Focus group protein crops. Accessed March 2017. https://ec.europa.eu/eip/agriculture/en/content/eip-agri-focus-group-protein-crops-final-report.

2. Swedish University of Agricultural Sciences. 2011. Fodertabell för gris. Accessed March 2017. http://www.slu.se/institutioner/husdjurens-utfodring-vard/Verktyg/fodertabell-for-gris/.

3. Jezierny, D., R. Mosenthin, and E. Bauer. 2010. The use of grain legumes as a protein source in pig nutrition: A review. Anim. Feed Sci. Technol. 157:111–128.

4. Grosjean, F., P. Cerneau, A. Bourdillon, D. Bastianelli, C. Peyronnet, and G. Duc. 2001. Feeding value, for pig, of near isogenic faba beans containing or not tannins and with high or low levels of vicine or convicine./Valeur alimentaire, pour le porc, de féveroles presque isogéniques contenant ou non des tanins et à forte ou faible teneur en vicine et convicine. J. Rech Porcine en France, 33:205–210.

5. Rubio, L. A., A. Brenes, and M. Castaño. 1990. The utilization of raw and autoclaved faba beans (*Vicia faba L., var. minor*) and faba bean fractions in diets for growing broiler chickens. Br. J. Nutr. 63:419–430.

6. Muduuli, D., R. Marquardt, and W. Guenter. 1982. Effect of dietary vicine and vitamin E supplementation on the productive performance of growing and laying chickens. Br. J. Nutr. 47:53.

7. Valdebouze, P., E. Bergeron, T. Gaborit, and J. Delort-Laval. 1980. Content and distribution of trypsin inhibitors and haemagglutinins in some legume seeds. Can. J. Plant Sci. 60:695–701.

8. Farrell, D. J., R. A. Perez-Maldonado, and P. F. Mannion. 1999. Optimum inclusion of field peas, faba beans, chick peas and sweet lupins in poultry diets. II. Broiler experiments. Br. Poult. Sci. 40:674–680.

9. Nalle, C. L., V. Ravindran, and G. Ravindran. 2010. Nutritional value of faba beans (*Vicia faba L.*) for broilers: Apparent metabolisable energy, ileal amino acid digestibility and production performance. Anim. Feed Sci. Technol. 156:104–111.

10. Gous, R. M. 2011. Evaluation of faba bean (*Vicia faba* cv. Fiord) as a protein source for broilers. S. Afr. J. Anim. Sci. 41:71–78.

11. Lacassange, L., M. Francesch, B. Carré, and J. P. Melcion. 1988. Utilization of tannin-containing and tannin-free faba beans (*Vicia faba)* by young chicks: Effects of pelleting feeds on energy, protein and starch digestibility. Anim. Feed Sci. Technol. 20:59–68.

12. Ospina-Rojas, I. C., A. E. Murakami, C. Eyng, R. V. Nunes, C. R. A. Duarte, and M. D. Vargas. 2012. Commercially available amino acid supplementation of low-protein diets for broiler chickens with different ratios of digestible glycine+serine:lysine. Poult. Sci. 91:3148–3155.

13. Masey O'Neill, H. V., M. Rademacher, I. Mueller-Harvey, E. Stringano, S. Kightley, and J. Wiseman. 2012.

Standardised ileal digestibility of crude protein and amino acids of UK-grown peas and faba beans by broilers. Anim. Feed Sci. Technol. 175:158–167.

14. Bach Knudsen, K. E. 1997. Carbohydrate and lignin contents of plant materials used in animal feeding. Anim. Feed Sci. Technol. 67:319–338.

15. Wiryawan, K. G., J. G. Dingle, and D. Creswell. 1997. Enzyme supplementation improves protein quality of grain legumes for poultry production. Page 231 In: Recent Advances in Animal Nutrition in Australia. Corbett, J. L., M. Choct, J. B. Rowe, and J. V. Nolan, eds. University of New England, Armidale.

16. NRC. 1994. Nutrient Requirement of Poultry. 9th rev. ed. Natl. Acad. Press, Washington, DC.

17. Danisco Xylanase 8000 G, Danisco Animal Nutrition, Marlborough, Wiltshire, UK.

18. Phyzyme XP 5000 TPT, Danisco Animal Nutrition, Marlborough, Wiltshire, UK.

19. Subcommitee Energy of the Working group nr. 2 Nutrition of the European Federation of Branches of the World's Poultry Sciences Association. 1989. European table of energy values for poultry feedstuffs. 3rd rev. ed. Grafisch bedrijf Ponsen and Looijen bv, Wageningen.

20. Avizyme 1505 X, Danisco Animal Nutrition, Marlborough, Wiltshire, UK.

21. Ekstrand, C., T. Carpenter, N. Andersson, and B. Algers. 1998. Prevalence and control of foot-pad dermatitis in broilers in Sweden. Br. Poult. Sci. 39:318–324.

22. Hudspeth, J. P., C. E. Lyon, B. G. Lyon, and A. J. Mercuri. 1973. Weights of broiler paers as related to carcass weights and type of cut. J. Food Sci. 38:145–150.

23. Jennische, P., and K. Larsson. 1990. Traditonella svenska analysmetoder för foder och växtmaterial, In: Statens Lantbrukskemiska Laboratorium, Uppsala, Sweden.

24. Nordic committee on feed analysis. 2003. Determination in feeds and faeces according to Kjeldahl. Nitrogen, NMKL, Olso, Norway.

25. Official Journal of European Communities. 1994. Determination of crude oils and fat. Method B.

26. Langeland, M., A. Vidakovic, J. Vielma, J. E. Lindberg, A. Kiessling, and T. Lundh. 2016. Digestibility of microbial and mussel meal för artic charr (*Salvelinus alpinus*) and Eurasian perch (*Perca fluviatilis*). Aquac. Nutr. 22:485–495.

27. SAS User's guide. 2013. Version 9.4 ed. SAS Inst. Inc., Cary, NC.

28. Snedecoer, G. W., and W. G. Cochran. 1989. Statistical methods. 8th ed. Iowa State University Press, Ames, Iowa.

29. Svihus, B., A. K. Uhlen, and O. M. Harstad. 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.

30. Summers, J., H. Shen, and S. Leeson. 1982. The value of canola seed in poultry diets. Can. J. of Anim. Sci. 62:861–868.

31. Kalmendal, R., and R. Tauson. 2012. Effects of a xylanase and protease, individually or in combination, and an ionophore coccidiostat on performance, nutrient utilization, and intestinal morphology in broiler chickens fed a wheat-soybean meal-based diet. Poult. Sci. 91:1387–1393.

32. Angel, R., and J. Sorbara. 2014. Why is it important to understand substrates if we are to optimize exogenous efficacy? Poult. Sci. 93:2375–2379.

33. Ghazi, S., J. A. Rooke, H. Galbraith, and M. R. Bedford. 2002. The potential for the improvement of the nutritive value of soya-bean meal by different proteases in broiler chicks and broiler cockerels. Br. Poult. Sci. 43:70–77.

34. Choi, J. H., B. S. So, K. S. Ryu, and S. L. Kang. 1986. Effects of pelleted or crumbled diets on the performance and the development of the digestive organs of broilers. Poult. Sci. 65:594–597.

35. Jimenez-Moreno, E., A. de Coca-Sinova, J. M. Gonzalez-Alvarado, and G. G. Mateos. 2016. Inclusion of insoluble fiber sources in mash or pellet diets for young broilers. 1. Effects on growth performance and water intake. Poult. Sci. 95:41–52.

36. van der Hoeven-Hangoor, E., C. J. Rademaker, N. D. Paton, M. W. A. Verstegen, and W. H. Hendriks. 2014. Evaluation of free water and water activity measurements as functional alternatives to total moisture content in broiler excreta and litter samples. Poult. Sci. 93:1782–1792.

Acknowledgments

The authors thank "Stiftelsen Svenska Kycklinguppfödare" for financial support.