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Effects of Feeding Processed Kidney Bean Meal *(Phaseolus Vulgaris)* by Replacing Soybean Meal on Egg Fertility and Qualities of Chicks of White Leghorn Hens

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Strictly as per the compliance and regulations of:

Effects of Feeding Processed Kidney Bean Meal (*Phaseolus Vulgaris*) by Replacing Soybean Meal on Egg Fertility and Qualities of Chicks of White Leghorn Hens

Taju Hussein [α], Mengistu Urge [σ], Getachew Animut [ρ] & Sisay Fikru [ω]

Abstract- The study was to evaluate feeding processed kidney bean meal (PKBM) instead of soybean meal (SBM) on fertility, hatchability, embryonic mortality and chick quality of white leghorn (WL) hens. Replacement of SBM with PKBM in the diet did not affect the fertility, hatchability and embryonic mortality. Chick length (15.63, 15.00, 15.33, 15.03 and 14.33 (SEM =±0.02)) and chick weight (34.13, 34.20, 33.13, 33.06 and 32.47(SEM=±1.39)) for T1, T2, T3, T4 and T5 respectively, were significantly ($P<0.05$) lower for T5 than the rest treatments. Visual assessment of chick quality is lower for treatments containing higher proportion of PKBM than treatments containing lower proportion of PKBM. Therefore, as it affects the quality of chicks at 100% replacement, it is only up to 75% replacement of SBM by PKBM (dosed at 195 g/kg concentrate diet) is possible without having significant negative effect on chick quality.

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I. Introduction

Chick quality is affected by pre-incubation storage conditions, time in the Hatcher, and size of egg. Tona et al., (2004) Found that increased incubation storage produced poor quality chick. Larger eggs tend to have significantly poor quality chicks as compared to other egg size Tona *et al.,* (2004).

The fertility of an egg is affected by several factors originating from the hen such as her ability to mate successfully, to store sperm, to ovulate an egg cell, and to produce a suitable environment for the formation and development of the embryo. The fertility also depends on the cock's ability to mate successfully, quantity and quality of semen deposited Brillard (2007). When fertility is low, it can affect other categories because of the lack of uniformity of embryo temperature inside the egg set (not as much heat provided by the developing embryos).

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Hatchability is a process that has several critical points that can be monitored and controlled to produce consistently healthy and mature hatchlings. These includes assessing hatching egg, fertility, egg storage and care, evaluation of hatch residue, poultry processing, sanitation, and poultry health and viability Hullet (2007).

The cost of poultry feed is very high and it accounts for 60-70% of the layer production cost Wilson and Beyer (2000). In recent years, the price of conventional or basic feed ingredients has tremendously increased. This has made poultry and live-stock production very expensive. In Ethiopia where soybean and its meal are in short supply and very expensive, the use of soybean meal as protein source of poultry ration is limited. Thus, an alternative protein source should be assessed and used. Sisay et al., (2015b) suggested that 100% replacement of SBM with PKBM is possible at 10% (100 g/kg) of soybean in layer ration. Therefore, the present research was initiated to evaluate the effect of feeding processed kidney bean meal (*Phaseolus vulgaris*) by replacing soybean meal at 19.5% (195g/kg) on egg fertility and qualities of chicks of white leghorn hens.

II. Materials and Methods

a) Management of Experimental Animals

Experimental house which were partitioned into 15 pens with wire-mesh and covered with grass litter material of 10 cm depth were used for experiments. Before the commencement of the actual experiment, the experimental pens, (2.5*2m), watering equipment, feeding troughs and laying nests were disinfected, sprayed against external parasites and thoroughly cleaned. Disinfectant was placed at the gate of the experimental house for workers and other visitors to step on before they enter into the house, which is in addition to the one placed at the main gate of poultry farm, for prevention of disease introduction.

The birds were vaccinated against Newcastle disease according to the vaccination program of the farm. The birds were offered with experimental diets for 7 days as period of adaptation before actual data collection takes places. The feed was measured and given to the birds in groups twice a day at 8 am and 2 pm on *ad libitum* basis by dividing the daily offer into two equal portions. Feed refusals were collected every morning at 7:30 am before providing new feed, external contaminants were removed by visual inspection, weighed and recorded for each pen separately. Feed were offered into two metal tubular feeders per pen that was hanged approximately at a height of the backs of the birds. Water was provided in a plastic fountains placed on a flat stone at the center of the pen. The watering trough was cleaned every morning before feeding. Clean and fresh water was available to the birds' *ad libtum*. The experimental duration lasted for 12 weeks.

b) Treatments and Experimental Design

The ration of the experiment consists of PKB, which replaced SBM at five levels, namely 0, 25, 50, 75 and 100% PKB for T1, T2, T3, T4 and T5, respectively (Table 1). Initially weighed two hundred ten (210) birds consisting of 180 White leghorn layers and 30 cocks of mature white leghorn breeds of similar age and weight, which were obtained from the University's poultry farm, were randomly assigned to the five treatment rations. Thus, the design employed was a completely randomized design (CRD) with five treatments and three replications (pen) per treatment (Table 1).

c) Chemical Composition of Experimental Feeds

Representative samples of feed ingredients was taken and analyzed before formulating the actual dietary treatment rations. The results of the analysis were used to formulate the treatment rations (Table 2). The feed ingredient offer samples were analyzed for dray matter (DM), ether extracts (EE), crude fiber (CF) and ash following the procedure of Weende (proximate analysis method) of the AOAC (1990). Kjeildhal procedure was employed to determine the nitrogen (N) content of the feeds and crude protein (CP) was determined by multiplying the N value with 6.25. The total metabolizable energy (ME) contents were calculated indirectly by using the formula presented by Wiseman (1987). ME (Kcal/kg DM) = 3951 + 54.4 EE – 88.7 CF – 40.8 Ash.

d) Data Collection and Measurements

i. *Fertility and hatchability*

Hundred ninety-five, that is 39 and 13 eggs from each treatment and replication, respectively, of medium sized, non-defected and normal shaped eggs were collected in three consecutive days. The eggs were kept at room temperature until incubated within 4 days after the first day of collection. The eggs were lightly coded by marker before they were placed into the incubator. The incubation temperature and relative humidity during the 18 days of incubation was auto fixed at 37-38°C and 65-70%, while that of hatchery unit was adjusted to 38.5-39°C and 90% relative humidity.

The eggs kept in the tray with small end down and turned automatically by slanting the tray at 45°. The incubator is equipped with the turner that facilitates the turning operation at an interval of two hours. Fertility was determined by candling the incubated eggs on the 7th day of incubation. Candling was done at dark room with egg Candler powered by electricity. Eggs found to be infertile, which are characterized by clear appearance, egg with blood adhering to one sides of the eggs were drown from the incubator. Finally eggs found fertile, i.e. eggs having small dark spot, numerous blood vessels arising from those dark spot of yolk at day of candling, clearly visible thick and dark and well fill structure was further kept in the incubator for hatching North (1984). Eggs with living embryo were transferred to the hatching section of the incubator at the end of the 18th day. Hatched chicks were counted and chick quality was determined. Fertility and hatchability were determined by using the following formulae, respectively.

$$\%\text{Fertility} = \frac{\text{number of fertile eggs}}{\text{total egg set}} x100$$

$$\%\ \text{Hatchability on fertile eggs base} = \frac{\text{number of chick hatched}}{\text{fertile eggs}} x100$$

$$\%\ \text{Hatchability on total eggs base} = \frac{\text{number of chick hathced}}{\text{total eggs set}} x100$$

ii. *Embryonic mortality*

Embryonic mortality was determined by candling eggs at 7th, 14th and 18th days of incubation and at hatching. Eggs that had a structure encircled with blood ring, absence of blood vessels, adhering to the shell membrane and absence of clear demarcation between embryo and air cell was considered as dead embryo and removed from the incubator North (1984). The dead embryo further categorized in to early, mid, late dead and pip embryo, based on the classification criteria set by Butcher (2009). The embryonic mortality was computed by using the formulae indicated by Rashed (2004).

$$\%\ \text{of early mortality} = \frac{total\ numbr\ of\ early\ dead\ embryo}{total\ number\ of\ fertile\ eggs} x100$$

$$\%\ \text{of mid mortality} = \frac{total\ number\ of\ mid\ dead\ embryo}{total\ number\ of\ fertile\ eggs} x100$$

$$\%\ \text{of late mortality} = \frac{total\ number\ of\ late\ dead\ embryo}{total\ number\ of\ fertile\ eggs} x100$$

$$\%\ \text{of pip mortality} = \frac{total\ number\ of\ pip\ dead\ embryo}{total\ number\ of\ fertile\ eggs} x100$$

iii. *Chick quality*

Chicks' quality were measured using two different methods, which includes visual scoring and measuring of day old chick weight and length. Visual scoring of chicks was done by visual examination based on the quality standards described by North (1984). Accordingly, a chick that was not malformed, physically active, stands up well, and looks live has been taken as good quality chicks. The researcher and two technicians were under taken the visual quality assessment. Based on common decisions, the chicks were classified in to poor and normal chicken. After the chicks were classified in to two groups, namely quality (chicks with dried body, stand well, and active) and non-quality chicks (chicks with wet body, not firm, non-straight, and not having perpendicular leg to the ground), five chicken from normal chicken were taken randomly from each replicate and their weight and length were measured using sensitive balance and ruler for further quality assessments respectively. The chick's length was taken by stretching the chicks on the table and taking the length from the tip of the beak to the middle toe by a ruler. Percentage of quality chicks were calculated as

Quality chicks =

$$\frac{\text{total number of quality chicks}}{\text{total number of hatched chicks}}\, x100$$

e) *Statistical Analysis and Models*

The data collected during the study period were subjected to statistical analysis using SAS computer software version 9.1.3. SAS (2008). during data analysis, chick weight and length were analyzed following one way analysis of variance procedure. When the analysis of variance indicated the existence of significant difference between treatment means, list significant difference (LSD) method was used to locate the treatment means that were significantly different from the other Gomez and Gomez (1984).

The model used for statistical analysis was:

Yij = μ+Ti + eij

Where: Yij=Individual observation,

Ti=Treatment effect, μ=Overall mean, eij= Error term

General logistic regression analysis was employed for analysis of data recorded on fertility (fertile/infertile), hatchability (hatched/un-hatched), embryonic mortality (alive/dead), and visual scoring (normal/poor). The general logistic regression model used is given below:

Model: $1n\left\{\frac{\pi}{1-\pi}\right\} \beta 0+\beta 1^*$ (x)

Test H0: No treatment effect (*i.e.*, β1=0) vs.

HA: Significant treatment effect (β1≠ 0).

Where, π=probability,

β=slope, x=treatment

III. Results and Discussion

a) *Nutrient Composition of Ingredients and the Treatment*

The results of the chemical analysis of ingredients used and nutritional composition of the ration–for each treatment are given in Table 3 and 4 respectively. The DM contents of PKB obtained in the present study was slightly lower than reported by Marzo et al., (2002) (93.2%) and Audu and Aremu (2011) (96.8) but very close to Sisay et al., (2014) and Sisay et al. (2015a) (88) while CP contents is slightly higher than reported by Marzo et al., (2002), Audu and Aremu (2011), Emiola (2011), Sisay et al., (2014), Sisay et al., (2015b) and Emiola and Olghobo (2006) which were 20.9%, 23.6%, 24.7%, 25.8% and 26.8%, respectively. The EE content of kidney bean is the same with that reported by Emiola (2011) but Marzo et al., (2002) reported lower than the current results. The CF content of kidney bean used in the present study was comparable to that reported by Arija et al., (2006), Emiola (2011), Sai-ut et al., (2009), Audu and Aremu (2011), Sisay et al., (2014) and Sisay et al., (2015b) who reported 5.1, 5.0, 6.0, 4.7% and 4.5% respectively. The result of chemical composition of kidney bean used in the present experiment showed comparable ME contents to that noted by Ofongo and Aloghobo (2007) and Arija et al., (2006) who reported 3342.2 and 3365 kcal/kg, respectively.

In general, as different report showed the processing (treatments) could cause drop or increase in nutrients compared to the raw seed. The treatment method (boiling) employed was suggested increased total carbohydrates and decrease the CP, EE, CF and ash contents Marzo et al., (2002), Audu and Aremu (2011). Similarly, Akaerue and Onwuka, (2010) and Mubarak (2005) reported reduced CP, EE, CF and ash for Mung bean (*Vigna radiata*). The low content of CF of PKB and the further reduction as a result of boiling might favored the feed intake of the layers by decreasing the problem of feed digestibility.

SBM and NSC are rich in CP content that make these ration to be ideal source of protein supplement for poultry. The NSC used to formulate experimental ration composed 26% CP and 2006 kcal ME. The CP values are comparable with that of Shewangzaw et al., (2011) and Meseret et al., (2011) but low in ME. Previously under taken studies indicated that the CP content of SBM to be in the range of 41 to 50% Waldroup (2002), Ekeren et al., (2006). However, similar to the value obtained in this study, a 38% CP content of SBM were reported in Ethiopia Meseret et al., (2011).

When SBM and PKB are compared, SBM contain by far higher CP, EE and ME. On the other hand, PKB has lower CF and ash than SBM. As a result, the CP of the formulated ration ranged between 16% (diet 5) and 18% (diet 1). Crude fiber increased, EE decreased because of total replacement of SBM by PKB. The energy content of SBM is higher than that of PKB. For this reason, the energy content decreased with increasing level of processed kidney bean. Nevertheless, energy and protein content of all rations ranged within the recommended level for layers. Lesson and summer (2001) recommended 16-18% of CP and 2500-3300 kcal/kg ME, respectively for white leghorn layers. Furthermore, the energy of compound ration is the same with that used by Zebiba (2012) and Senayt et al., (2011) in their experiment for the same bird in the same farm.

b) Fertility and Hatchability of Eggs

Mean values of fertility and hatchability for the treatments are presented in Table 5. The logistic regression results for fertility and hatchability of eggs showed no significant difference among treatments. However, there was a numerical decrease in fertility and hatchability percentage for T5 as compared toT1 which is similar with Sisay et al., (2015b) finding. The numerical decrease in fertility and hatchability from T1 to T5 could be the result of increased, level of kidney bean in the ration that caused reduction of level of protein, calcium and phosphorus found in the rations. The diets of breeder poultry should be adequate in both quantity and quality to meet the recommended levels of feed standard Brillard (2007). The level of dietary protein significantly affected egg fertility and hatchability Gareil et al., (2006). Poor hatching happen when nutritionally deficient feed is used for layers Hocking et al., (2002). The authors have shown that low calcium levels tended to decrease percent of fertility and hatchability. On the contrary, El-Ghamry et al., (2010) reported improved fertility (83% and 77%) and hatchability (78% and 71%) for the group fed 2.4% and 2.6% calcium, respectively.

c) Embryonic Mortality

The mean values of embryonic mortality at different stages of development are presented in Table 5. The logistic regression result showed no significant differences among treatments for early, mid, late and pip embryonic mortality. Nevertheless, the mortality tends to numerically increase with increased level of PKB compared to control diets that do not contain PKB. This might occurred because of the decreased crude protein level with increased level of PKB. Embryonic mortality of eggs of breeder hens' fed low protein is reported to be higher than hens fed high protein diets. Low-protein rearing rations were associated with higher rates of food intake, higher mortalities and lower rates of egg production than the conventional protein ration Hocking et al., (2002).

d) Chick Quality

i. Visual Scoring (Observation)

The visual scoring of chicks is presented in Table 6. Wald chi-square statistics indicated that visual scoring of chicks was insignificant (0.64) at (α = 0.05) among the treatments, but Sisay et al., (2015b) indicated that visual scoring of chicken was not significant (*pr>chisq* 0.641 α=0.05). The visual assessment showed that the quality of chicken is better in the order of T2>T1>T3>T4, while the chick quality of T5 is inferior or poor on visual scoring. The chicken in T5 were not well standing, they are dehydrated and seems inactive at their day old age. This assessment was supported by length and weight of chicks that indicated significant differences among treatments. Utilization of visual score parameters such as naval quality, firmness of leg, size of beak, eye and vital chick are recommended ways of determining highest quality chicks Petek et al., (2010).

ii. Chick Length

The length of chicks hatched from eggs of hens fed diet containing 0, 25, 50 and 75% of PKB are not significantly different which is similar with Sisay et al., (2015b) finding that there was no significant difference (*p*>0.05) among treatments in chick weight and chick length. However, the chicken hatched for eggs harvested from layers fed 100% PKB (T5) had significantly lower length as compared to that from the layers fed SBM at different levels. This occurred either because of the egg size that accommodate larger embryo of chicken compared to the small eggs that has less environment to hold large chicken or the nutrition of the layers that promote better growth of the chicks. Mukhtar et al., (2013) Found similar result and stated that hatching length is an easy and repeatable quality evaluation parameter for newly hatched chicks. This important trait has a positive correlation with the size of the egg and the chick's weight. It is an important economic trait to predict chick development because it is positively related to yolk-free body mass at hatch and potential of chicks for optimum future performance.

Furthermore, Petek et al., (2010) pointed out that each extra cm of hatchling length at day of hatch meant an increase of 18 g BW at seven days of age. Chicks with longer hatchling length have better-feed efficiency and survival rates as compared to smaller chicks. Petek et al., (2009) Classified length intervals in to short, middle and long for a day old chicks. Accordingly, layer chick with a length of < 17.8, 17.8-18.2 and > 18.2cm, respectively are grouped as short, medium and long chicks. As to this classification, chick lengths in the present experiment for all treatments fall within a short category. On other hand, Petek et al., (2008) reported that the body weight and chick length uniformity in long group in all poultry to be better than the shorter group

iii. Chick Weight

The chick weight of layers were higher (P<0.05) in ration that do not contain SBM as compared to the ration containing 100%PKB (Table 6). The variation in chick weight may be due to the weight of eggs, which is slightly decreasing across treatment as well as the amino acid content of the rations, which is higher in control diet while decreasing across treatment because of decreased SBM. The present finding is in agreement with many previous of findings. For instance the chicks injected with amino acids invariably had higher plasma protein and lower plasma glucose on the day of hatch, because methionine and threonine are critical for the growth of chicken embryo Subrat et al., (2012). Egg weight has a direct impact on the weight of chick and there is a positive correlation between egg and chick weight Petek et al., (2010). Chicken hatched from large eggs are heavier than those hatched from comparatively smaller eggs Al-Murrani (1978). A heavy chick indicates a good development. However, this is sometimes not true and evaluating chick quality by measuring only body weight can be misleading Molenaar (2009).

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Table 1: Experiment lay out

Treatments	Replications	Layers per replication	Cocks per replication
T1(100%SBM:0%PKB)	3	12	2
T2 (75% SBM:25%PKB)	3	12	2
T3 (50% SBM:50%PKB)	3	12	2
T4 (25% SBM:75%PKB)	3	12	2
T5 (0% SBM:100% PKB)	3	12	2

SBM=Soybean meal; PKB=Processed kidney bean; 100% PKB represents replacement of maximum SBM (260g/kg) as recommended by earlier study Senayt et al. (2011)[6]

Table 2 : Proportion of feed ingredient used in formulating experimental rations

	Treatments				
Feed (%)	T1	T2	T3	T4	T5
CG	56	53	52	48	37
WS	7	7	7	7	16
SBM	26	19.5	13	6.5	0
PKB	0	6.5	13	19.5	26
NSC	4	7	8	12	14
LS	5.5	5.5	5.5	5.5	5.5
Salt	0.5	0.5	0.5	0.5	0.5
VPM	1	1	1	1	1
Total	100	100	100	100	100

CG= corn grain; WS= wheat short; SBM= soybean meal; PKB= processed kidney bean; NSC= noug seed cake; LS= limestone; VPM= vitamin pre mix; T1 100%SBM: 0%PKB; T2= 75%SBM: 25%PKB; T3= 50%SBM: 50%PKB; T4=25%SBM: 75%PKB; T5= 0%SBM: 100%PKB

Table 3 : Ingredient used in the study and its nutrients compositions

	Nutrient composition (% for DM and % DM for others)					
Feed type	DM	CP	EE	CF	Ash	ME kcal/kg
CG	89.5	8.7	4.3	8.0	6.21	3230.5
WS	90.3	12	3.3	6.2	6.8	3303.1
SBM	90.2	38	7.0	9	7.8	3215
PKB	87.5	28	0.9	6	7.0	3182.2
NSC	91.5	26	6.0	21.0	10	2006.0

CG=Corn grain; WS=Wheat short; SBM=Soybean meal; PKB=processed Kidney bean; NSC=Noug seed cake; DM=dry matter; CP=Crude protein; EE=ether extract; CF=crude fiber; ME=methabolizable energy

Table 4 : Nutritional composition of treatment diets containing different levels of processed kidney bean as a replacement for soybean meal

	Nutrient composition (% for DM and % DM for others)							
Treatments	DM	CP	EE	CF	Ash	Ca	P	ME kcal/kg
T1	91.85	18	5.64	6.26	9.96	3.4	0.39	3296.20
T2	91.56	17.8	5.63	6.36	9.97	3.26	0.38	3286.40
T3	91.17	17.6	5.58	6.52	9.98	3.28	0.38	3269.00
T4	90.21	16.3	5.40	6.56	9.98	3.01	0.36	3255.70
T5	89.86	16.0	4.90	6.86	10.20	2.79	0.32	3192.90

DM= dry matter; CP= crude protein; EE = ether extract; CF= crude fiber; SBM= soybean meal; PKB processed kidney bean; T1 100%SBM: 0%PKB; T2= 75%SBM: 25%PKB; T3= 50%SBM: 50%PKB; T4=25%SBM: 75%PKB; T5= 0%SBM: 100%PKB

Table 5 : Fertility and hatchability of eggs and embryonic mortality in layers fed different levels of processed kidney bean as a replacementz to soybean meal

	Treatments						
Parameter	T1	T2	T3	T4	T5	SEM	SL
Fertility (%)	94.87	94.87	94.87	94.87	92.3	2.29	NS
HTES (%)	71.79	71.79	71.79	64.10	61.54	4.59	NS
HFES (%)	75.43	75.64	75.43	69.44	66.67	3.50	NS
EEM (%)	5.56	5.56	5.56	8.33	8.33	2.14	NS
MEM (%)	5.56	8.12	8.12	11.11	8.33	1.76	NS
LEM (%)	5.56	5.56	5.56	8.30	13.89	2.48	NS
PIEM (%)	10.89	8.12	10.89	11.11	11.11	3.07	NS

SBM= soybean meal; PKB= processed kidney bean;T1=100% SBM:0% PKB; T2=75%SBM:25%PKB; T3=50%SBM:50%PKB; T4=25%SBM:75%PKB; T5=0%SBM:100PKB; HTES= hatchability on total egg set; HFES= hatchability on fertile egg set; EEM =early embryonic mortal; MEM= mid embryonic mortality; LEM = late embryonic mortality; PIEM =pip embryonic mortality

Table 6 : Chick quality parameters of white leghorn layers fed different levels of processed kidney bean as a replacement to soybean meal

	Treatment						
Parameters	T1	T2	T3	T4	T5	SEM	SL
Visual (%)	84.93	85.09	82.36	81.02	70.83	3.89	NS
Length (cm)	15.63^{a}	15.00^{a}	15.33^{a}	15.03^{a}	14.33^{b}	0.20	*
Weight (g)	34.13^{a}	34.20^{a}	33.13^{a}	33.06^{a}	32.47^{b}	0.28	*

$^{a\text{-}b}$ *means in the row without common superscript are significant; NS = Non- significant, SL= significance level; SEM= standard error of mean ; cm = cent meter, g = gram; % = percent; PKB = processed kidney bean meal; SBM= soybean meal; T1=100% SBM:0% PKB; T2=75%SBM: 25%PKB; T3=50%SBM: 50%PKB; T4=25%SBM: 75%PKB; T5=0%SBM: 100PKB;*