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THE EFFECTS OF DIFFERENT PROCESSING METHODS OF VELVET BEANS (*Mucuna pruriens*) ON L-DOPA CONTENT, PROXIMATE COMPOSITION AND BROILER CHICKEN PERFORMANCE

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SUMMARY

Recurrent droughts and soil degradation have had an adverse impact on food security and have increased malnutrition in Zambia. This has necessitated the promotion of conservation farming and diversification of food production. Velvet beans (*Mucuna pruriens*) is a crop that offers excellent soil conditioning properties and produces large quantities of beans of high protein and energy content. However, the beans contain anti-nutrients, particularly L-Dopa, that limit their use as a food or feed in humans and non-ruminant animals. Thus, this study was undertaken to investigate the efficacy of several preparation methods on the reduction of L-Dopa, on proximate composition, and on the performance of broiler chickens consuming the processed beans.

The processing methods resembled those used at household and community levels for preparing beans. Samples of 300 g of whole beans or grits of velvet beans were processed by soaking, boiling and both, with or without addition of sodium bicarbonate at 0.25% or 0.50%. The processed beans were then fed to 1000 Arbor Acre hybrid broiler chickens starting at day-old in a completely randomized block design. The birds were fed in triplicate in groups of 15 to assess their performance. The diets were designed so that 50% of the crude protein came from the velvet beans.

The processing results indicate that the lowest levels of L-Dopa (0.66-0.39 %, or an 88 % reduction) were achieved by soaking grits for 24 hours, then boiling for an hour and soaking again for 24 hours. The method of first boiling and then soaking for 24 hours had the next lowest L-Dopa level (68 % reduction). Soaking for 24 h alone had no effect on L-Dopa levels in whole beans or grits. Addition of sodium bi-carbonate at 0.25 and 0.50% showed variable influence on the L-Dopa levels. The feeding trial results confirmed these results and the processing methods that included boiling induced similar live weight gains, feed conversion ratios and carcass dressed percentages as a maize-soybean-based control diet.

Key words: *Mucuna*, velvet beans, sodium bicarbonate, L-Dopa, soaking, dehulling.

INTRODUCTION

Velvet beans in Zambia

Velvet bean (*Mucuna pruriens*) is a crop whose soil fertility improvement potential has been highly appreciated by Zambian farmers who have been growing it as a green manure and cover crop. Poor soil fertility has for years been one of the major constraints to increased crop production among small-scale farmers.

The removal of subsidies for farm inputs and closure of agricultural credit banks after 1991 have put purchases of inorganic fertilizers and other agricultural inputs out of reach of these farmers. In addition, the adverse effects caused by indiscriminate use of inorganic fertilizers have led to high reductions in yield or total crop failure. The use of several legumes, including velvet bean, has been found to be a viable alternative for improving soil fertility and main crop yield and thus, has been extensively promoted by the Ministry of Agriculture and Co-operatives (MACO) after 1991.

Velvet beans are indigenous to Zambia and are associated with uncultivated fertile soils. They were also commonly grown in Eastern Province to improve soil fertility prior to the introduction of the chemical fertilizer-based Lima Programme of the First and Second Republics of Zambia (Kaonga, 2002). Three cultivars have been developed for soil fertility improvement: Sam (white seed coat), green (green/beige), and NIRS 16 (black). Many other wild and itchy varieties grow in different parts of Zambia in virgin lands. Velvet bean consumption has been limited to famine periods and rough situations. They have been processed by elderly members of the community who know how to process the beans so that no intoxication follows when the beans are consumed. Additional uses for velvet beans include post-harvest grazing of fields by cattle.

The limited use of velvet bean as a food and feed in Zambia is due to the presence in the seed of anti-nutritional factors such as levodopa (L-Dopa), beherenic acids (Siddhuraju *et al*., 1996), lectins and enzyme inhibitors. The limited usage of velvet beans has provided a weak incentive for the farmers to expand the area of the crop despite the high yields of both herbage and seeds and its soil enriching characteristics.

The noxious effects of L-Dopa (e.g., nausea and vomiting) appear to be the most notorious of the anti-nutritional factors. Traditionally, the L-Dopa is extracted from the beans by leaching and boiling methods, processes that have proven to be rather tedious and laborious, making the high protein bean unpopular as a food except in times of scarcity or famine. Soaking velvet beans in alkaline solutions of 4% calcium hydroxide ($Ca(OH)_2$) has been found to reduce L-Dopa content to extremely low levels in Mexico (Diallo *et al*., 2002). However, the black color of seed processed in this manner makes the beans visually objectionable for human consumption. In Zambia, cooking in sodium bicarbonate (baking soda, NaHCO3), a cheap, harmless, widely accepted and readily available chemical, is common. Wood pottash is also used in most rural areas. Baking soda or pottash are used, among other reasons, to reduce cooking time, to eliminate poison in mushrooms, and to improve the taste and digestibility of vegetables. If a simple and effective method to reduce or eliminate L-Dopa can be developed, the velvet beans can be increasingly used for soil fertility improvement, weed control and high protein food for humans and livestock.

Processing velvet beans

Heat treatment is the most common method of preparing velvet beans for consumption by humans and other monogastric animals because most of the deleterious compounds in velvet beans are heat labile. Heat treatment has been achieved by various means, e.g., by roasting at 120º C for 30 min, by grilling for an hour after pre-soaking the seeds for 24 h and thereafter dehulling, and by autoclaving for 30 min (Siddhuraju *et al*., 1996; Dossa *et al*., 1998; Del Carmen *et al*., 1999).
Siddhuraju *et al*. (1996) found dry heat treatment to be effective in reducing L-Dopa in velvet beans. They attributed the reduction in L-Dopa to its racemization under roasting. Studies by Hayase *et al*. (1975) revealed that amino acid residues in proteins and in synthetic peptides can racemize under roasting conditions. Diallo *et al*. (2002) showed that adding $Ca(OH)_2$ to broken velvet beans resulted in extremely low L-Dopa content (0.01%). However, the resulting mixture was black, probably due to conversion of L-Dopa to melanin.

Heat treatment through roasting or boiling has been found to be most effective in reducing other anti-nutrients in velvet beans. According to Dossa *et al*. (1998), nutritional quality is maintained better through grilling than cooking. This was contradictory to the findings of an earlier study where boiling was reported to be better than roasting velvet beans (Laurena *et al*., 1991). Heat treatment by thorough roasting and cooking can successfully reduce HCN levels by as much as 68%. Dry heat treatment reduced the content of phytic acid by 36% and autoclaving reduced phytate content by 47% in velvet bean (Siddhuraju *et al*., 1996). Beherenic acid is more stable to heat and soaking treatment. Autoclaving at 130º C for one hour and soaking the seed for 20 h only removed 15 and 1.5%, respectively, of the acid in winged bean. Trypsin inhibitors (TI) are known to be heat labile as heat disintegrates their structure. Significant reductions in velvet bean TI, 93% by roasting and 96% by autoclaving, were reported by Siddhuraju *et al*. (1996). Pre-soaking prior to cooking may improve the extraction efficiency of protease and alpha-amylase inhibitors (Moneam, 1990).

In general, a significant reduction of hemagglutinating activity due to lectins was reported among all blood groups (ABO) when the seeds were subjected to both dry heat treatment and autoclaving (Siddhuraju *et al*., 1996). Roasting can reduce the negative effects of protease and alpha-amylase inhibitors on digestion by 96% and cooking velvet beans removes the negative effects of these anti-nutritional factors completely (Siddhuraju *et al*., 1996).

Thus, the general aim of this study was to define appropriate methods for reducing L-Dopa that could be used at village level for both human and animal feeding of velvet beans. Specifically, the study investigated the efficacy of several velvet bean preparation methods that included soaking, boiling or both, and treatment with or without sodium bicarbonate in the reduction of L-Dopa and on the performance of broiler chickens fed the processed beans.

MATERIALS AND METHODS

Processing of samples was conducted at the Field station of the School of Agricultural Sciences, University of Zambia. Local speckled velvet beans were procured from small-scale farmers practicing conservation farming and using them for improving soil fertility.

Three hundred gram samples of whole velvet beans and grits were subjected to varying combinations of treatment processes that included soaking, heat treatment by boiling and addition of sodium bicarbonate to the first soaking water as presented in Table 1.

Soaking and soda treatment

All the treatments began with soaking of the samples in 300 mL of water (weight of water equivalent to the weight of sample) after soda was added to the respective treatments, in zip-lock plastic bags used as containers. This first soaking lasted approximately 12 h or until the beans absorbed the water in the plastic bags. Subsequent soakings lasted for 24 h and used water at five (1.5 L) or ten (3.0 L) times the dry weight of the samples.

Boiling

Heat treatment of the samples used volumes of water equivalent to five times the sample dry weight. Enamel pots were used for boiling the beans on charcoal burners with wooden spoons used to stir the contents. The samples were added to the pots once boiling started. One to three minutes later the beans would boil and they were left to boil for 60 min.

Drying

The samples were dried on metal trays in a draft air oven at 50º C. It required 17 to 18 h for the samples to be dry enough for grinding without clogging the grinder. Some attempts were made to dry the beans on mesh in the sun, when the draft oven developed an electrical fault, but these samples were difficult to grind.

Grit making

Grits were prepared by soaking velvet beans in warm water for a few minutes until the seed coat could be easily removed, and then dried in a draft air oven at 50º C for 12 h. Grits were made initially by pounding velvet beans in a mortar with a pestle. However, this process proved to be too time consuming and the particle size of the samples was not uniform. Thus, a Wiley Mill was used with 0.5 cm sieve to produce 4 mm grits.

Proximate chemical composition analysis of velvet beans

Chemical analysis followed AOAC methods (1998). Moisture content was determined by drying 2 g samples in a Memment oven (Model 500, Memmert Company, Schwabach, Germany) at 110 ºC for 2 hours. For crude protein analysis (according to AOAC, 1998), samples weighing approximately 2 g were digested in a Foss Tecator Digestion System (Foss Tecator Company, Hoganas, Sweden) at 420 ºC for 1 h and distilled in a Markham semi-micro Kjeldahl apparatus. Ether extract analysis was determined using 5 g samples placed in extraction thimbles in soxhlet flask. Determination of ash and mineral extraction was done by combusting approximately 2 g in a Nabertherm muffle furnace (Nabertherm Company, West Germany) to ash at 550 ^{0}C. The ash was used to determine Ca^{2+} and P content of the sample.

The minerals in the ash were first extracted by boiling ash in 10 mL of 2 N hydrochloric acid. The solution was then filtered out into a 100 mL flask and made up to the 100 mL mark by washing the residue with hot distilled water. Calcium was then precipitated with ammonium oxalate and titrated to a faint pink with N/10 potassium permanganate. Phosphorous-diluted solutions were read by a colorimeter at 660 nm wavelength. A standard curve was used to determine the concentration of the sample solutions. Determination of neutral detergent fibre was done according to AOAC (1998).

L-Dopa analysis

The raw and processed samples were ground in a Wiley Mill with 0.8 mm sieve. Samples of 50 g were sealed in zip-lock bags and shipped to Judson College in Elgin, Illinois, after phytosanitory certification. The L-Dopa content was analysed by Professor Myhrman's Laboratory at Judson College.

Feeding trial

The raw and processed speckled beans were fed to 1000 Cobbs hybrid broiler chickens starting at day old in a completely randomized block design. The birds were fed in triplicate in groups of 15 to assess growth performance on the processed beans. The diets were designed so that 50% of the crude protein was provided by the velvet beans, 36 % by soybeans, and the remaining 14% by maize. The birds were weighed twice a week, the feed intake was recorded daily, and the dressed weights were determined at end of the feeding trial (41days). Data collected were subjected to statistical analysis of variance (ANOVA) using the technique of Steele and Torrie (1980) and differences between paired means were tested with Duncan multiple range test.

RESULTS AND DISCUSSION

Observations during processing

Boiling the beans for 40 min was not adequate to make the beans soft, thus boiling was continued for 60 min. During boiling, the grits tended to froth. The frothing was more pronounced with and at higher levels of sodium bicarbonate. With sodium bicarbonate, the beans changed to an unappealing brown colour that may be objectionable for human consumption.The beans were not adequately dry for grinding even after 3 d of sun drying. Odors on the second day of drying indicated the occurrence of some fermentation in the beans. All the beans completed their drying in the draft oven. Overall, the process of boiling and drying was labour- and energy-intensive and time consuming (it took 3 wk to process 50 samples).

Grit making by means of pounding with a mortar and pestle was extremely difficult. Only the bean hulls managed to peel off the hard dry beans while partially dry beans tended to get mushy. Successful grit making was achieved only by using an electric grinder with a 0.5 cm sieve to produce 4 mm grits. Smaller grits tended to mash when boiled and lost a lot of soluble particles on decanting.

Nutrient composition

The nutrient composition results are presented in Table 1. Boiling before soaking increased crude protein content but reduced the total energy content due to leaching of nutrients into the discarded water. Soaking and boiling grits increased both energy and protein content. The acid detergent fiber and ash were reduced in grits compared to whole beans. For samples processed under water the longest, the process of fermentation resulted in increased nutrients, particularly crude protein. Boiling and soaking in sequence reduced nitrogen free extract and total metabolizable energy. This would indicate higher loss of soluble nutrients when boiled grits were soaked and decanted.

L-Dopa content

The L-Dopa content of the raw local velvet bean varieties was 3.75, 3.90, and 4.36% for the white, speckled, and black beans, respectively. The L-Dopa content of the pre-soaked speckled beans used for the feeds was 4.02%. Soaking grits in 1.5 L, boiling in 1.5 L and then soaking in 1.5 L water for 24 h, all in the presence of sodium bicarbonate (0.25%), extracted the most L-Dopa (90%; from 4.02 to 0.39 %) (Table 2).

Table 1. Proximate composition of processed and unprocessed velvet beans.

Type	Processing method	Moist	CP	ADF	EE	Ash	NFE	ME
		- - - - - - - -	- - - - -	- - - %	- - - - -	- - - - -	- - - - -	Mcal kg^{-1}
Local speckled	None	8.87	24.92	n.d.	3.44	3.30	n.d.	n.d.
Local white	None	9.62	24.71	n.d.	3.88	4.59	n.d.	n.d.
Local green	None	8.48	23.50	n.d.	6.81	4.14	n.d.	n.d.
Local black	None	8.68	22.51	n.d.	5.40	3.30	n.d.	n.d.
Nicaraguan speckled	None	8.29	24.00	n.d.	3.72	3.06	n.d.	n.d.
Speckled whole	Soaked (10x)	6.24	24.48	7.48	5.45	3.01	53.34	3.47
	Boiled	7.56	24.46	11.85	6.03	2.10	48.09	3.13
	Soaked (10x), boiled	8.20	24.48	8.55	6.78	2.04	49.95	3.25
	Boiled, soaked (10x)	8.36	25.34	7.69	6.67	1.55	50.39	3.28
	Soaked (5x), boiled, soaked (5x)	7.81	24.46	7.37	6.48	1.84	52.04	3.39
Speckled grits	Soaked (5x)	5.49	24.01	3.03	6.62	2.22	58.63	3.58
	Soaked (10x), boiled	6.15	25.37	4.62	6.84	1.54	55.48	3.47
	Boiled, soaked (10x)	5.99	27.28	6.42	6.79	1.76	51.76	3.23
	Soaked (5x), boiled, soaked (5x)	6.54	26.00	3.11	6.90	0.83	56.65	3.61

Note: All samples were first soaked in water weight that equaled sample weight and lasted approximately 12 h (until all water was absorbed by beans).
Subsequent soakings lasted 24 h and were done using water at five (5x) or ten (10x) times the dry weight of the samples.
n.d.=not determined.

Table 2. L-Dopa content of processed velvet beans, whole and grits.

$NaHCO_3$	Processing method	L-Dopa (%)	SD
Whole beans		4.17	
None	Soaked (12 h; 5x)	4.02	0.333
None	Boiled	2.17	0.010
None	Soaked (24 h; 10x)	3.81	0.178
None	Soaked (10x), boiled	2.15	0.047
None	Boiled, soaked (10x)	1.23	0.098
None	Soaked (5x), boiled, soaked (5x)	2.06	0.036
0.25	Soaked (12 h; 5x)	4.16	0.074
0.25	Boiled	2.07	0.010
0.25	Soaked (24 h; 10x)	3.85	0.107
0.25	Soaked (10x), boiled	1.98	0.108
0.25	Boiled, soaked (10x)	1.37	0.414
0.25	Soaked (5x), boiled, soaked (5x)	1.31	0.720
0.50	Soaked (12 h; 5x)	4.08	0.040
0.50	Boiled	2.39	0.066
0.50	Soaked (24 h; 10x)	3.85	0.205
0.50	Soaked (10x), boiled	1.35	0.186
0.50	Boiled, soaked (10x)	n.d.	n.d.
0.50	Soaked (5x), boiled, soaked (5x)	1.52	0.032
Grits			
None	Boiled	1.72	0.103
None	Soaked (24 h; 10x)	1.86	0.023
None	Soaked (10x), boiled	2.02	0.013
None	Boiled, soaked (10x)	1.94	0.191
None	Soaked (5x), boiled, soaked (5x)	0.66	0.161
0.25	Boiled	2.53	0.174
0.25	Soaked (24 h; 10x)	3.45	0.110
0.25	Soaked (10x), boiled	1.10	0.015
0.25	Boiled, soaked (10x)	1.14	0.050
0.25	Soaked (5x), boiled, soaked (5x)	0.39	0.039
0.5	Boiled	2.15	0.103
0.5	Soaked (24 h; 10x)	3.38	0.147
0.5	Soaked (10x), boiled	1.55	0.008
0.5	Boiled, soaked (10x)	0.93	0.073
0.5	Soaked (5x), boiled, soaked (5x)	0.47	0.055

Note: All samples were first soaked in water weight that equaled sample weight and lasted approximately 12 h (until all water was absorbed by beans). Subsequent soakings of whole beans and all soakings of grits lasted 24 h and were done using water at five (5x) or ten (10x) times the dry weight of the samples.

Similar treatment of whole beans reduced L-Dopa by 67% (to 1.31%). Soaking grits for 24 h in 3.0 L water (with no sodium bicarbonate) reduced L-Dopa by 54% (from 4.02 to 1.86%) but soaking whole beans in the same water volume reduced it only by 5% (to 3.81%). In the absence of sodium bicarbonate, boiling whole beans and grits without soaking in water reduced L-Dopa only by 48.5% (from 4.02 to 2.07%) and 57% (to 1.72%), respectively. These results indicate that soaking grits before and after boiling (5 times the weight of sample) extracted more L-Dopa than the same treatment on whole beans or with one larger water volume (10 times the weight of dry sample).

The sodium bicarbonate treatment did not result in a marked additional decline of L-Dopa level. Overall, the variance between samples within the same treatment was very high. Notable was the increase in dark coloration of the beans with increased sodium bicarbonate levels.

Broiler chicken performance

The results of the broiler chicken study are presented in Table 3 and Figure 1 and 2. Broiler chickens fed from day-old with diets containing boiled velvet beans treated with or without sodium bicarbonate had significantly ($P<0.05$) higher live weights than those fed soaked raw velvet beans (typically values were

1500-1600 g vs. 900-1050g). Most of the chicks on raw beans died within the first 2 weeks; however, a few survived and grew sub-optimally up to the end of the feeding trial. In general, all the birds on velvet beans showed lower growth rates in the first 2 weeks. Those on boiled velvet beans with or without sodium bicarbonate treatment caught up with the birds on the control diet by the fourth week (Figure 1). Feed conversion ratio (FCR) was significantly better for broilers on the boiled than on the raw beans even if they were soaked for a long time (2.01-2.16 and 2.47-3.45, respectively). The two levels of sodium bicarbonate in combination with boiling did not show any adverse effect on broiler performance and their results were similar to the maize-soybean control diet.

The birds on velvet bean diet, particularly those on diets treated with sodium bicarbonate, had dark-coloured shanks and pale carcasses. In addition, birds on raw or unboiled velvet beans tended to be hyperactive (i.e., jumping out of baskets at weighing time and moving a lot in their pens).

Although combinations of boiling, sodium bicarbonate, and additional soaking at times reduced L-Dopa far more than 50%, the results suggest that boiling alone was the principal method for eliminating L-Dopa and that 50% reduction in L-Dopa is adequate for improving broiler diets. This is an important result in that the additional work of soaking for 24 or 48 h may not be worthwhile.

Table 3. Performance of broiler chickens fed processed and sodium bicarbonate-treated velvet beans.

$NaHCO_3$	Processing method	Live body wt. (g)	Feed intake (g)	Feed conversion ratio	Dressed weight (g)	L-Dopa (%)	SD
None	Soaked (5x)	877.5a	2711a	3.09bc	57.91a	4.02	0.333
None	Boiled	1548.7b	3149ab	2.05a	76.11	2.17	0.01
None	Soaked (24 h; 10x)	931.7a	2939ab	3.18bc	61.84a	3.81	0.178
None	Soaked (10x), boiled	1509.5b	3241b	2.15a	67.03bc	2.15	0.047
None	Boiled, soaked (10x)	1466.2b	3212b	2.19a	65.85b	1.23	0.098
None	Soaked (5x), boiled, soaked (5x)	1479.5b	3013ab	2.03a	65.85b	2.06	0.036
0.25%	Soaked (5x)	1057.7a	2610a	2.47ab	63.54ab	4.16	0.074
0.25%	Boiled	1618.8b	3247b	2.01a	67.35bc	2.07	0.01
0.25%	Soaked (24 h; 10x)	1015.8a	2759a	2.72ab	63.54ab	3.85	0.107
0.25%	Soaked (10x), boiled	1416.4b	3208b	2.63ab	67.35bc	1.98	0.108
0.25%	Boiled, soaked (10x)	1648.2b	3298b	2.01a	63.29ab	1.37	0.414
0.25%	Soaked (5x), boiled, soaked (5x)	1486.8b	3111b	2.09a	64.14b	1.31	0.72
0.50%	Soaked (5x)	882.2a	3029ab	3.45c	59.42a	4.08	0.040
0.50%	Boiled	1521.8b	3291b	2.16a	67.08bc	2.39	0.066
0.50%	Soaked (24 h; 10x)	968a	2687a	2.78ab	61.92a	3.85	0.205
0.50%	Soaked (10x), boiled	1507.3b	3259b	2.23a	66.98bc	1.35	0.186
0.50%	Boiled, soaked (10x)	1553.9b	3513bc	2.26a	67.12bc	n.d.	n.d.
0.50%	Soaked (5x), boiled, soaked (5x)	1613b	3256b	2.02a	67.14bc	1.52	0.032
None	Control (maize-soybean)	1767.3b	3196.31b	1.81a	1.81a		
	SD	320.8	303.4	0.49	3.7		
	P-value	<0.05	<0.01	<0.01	<0.075		

Note: All samples were first soaked in water weight that equaled sample weight and lasted approximately 12 h (until all water was absorbed by beans). Subsequent soakings lasted 24 h and were done using water at five (5x) or ten (10x) times the dry weight of the samples.
n.d.=not determined.

CONCLUSION

These results indicate that velvet beans, if boiled, can be safely fed to broiler chickens at levels up to 50 % of both the protein and energy and that local speckled Zambian velvet beans are high in crude protein (24-27 %) and in metabolizable energy (3.2-3.5 Mcal kg^{-1}). Dehulling and boiling combined with soaking, with or without the addition of sodium bicarbonate, could be used as a starting point in processing for human consumption. However, these methods are labor intensive and more work is needed to confirm the processing efficiency of velvet beans for human consumption.

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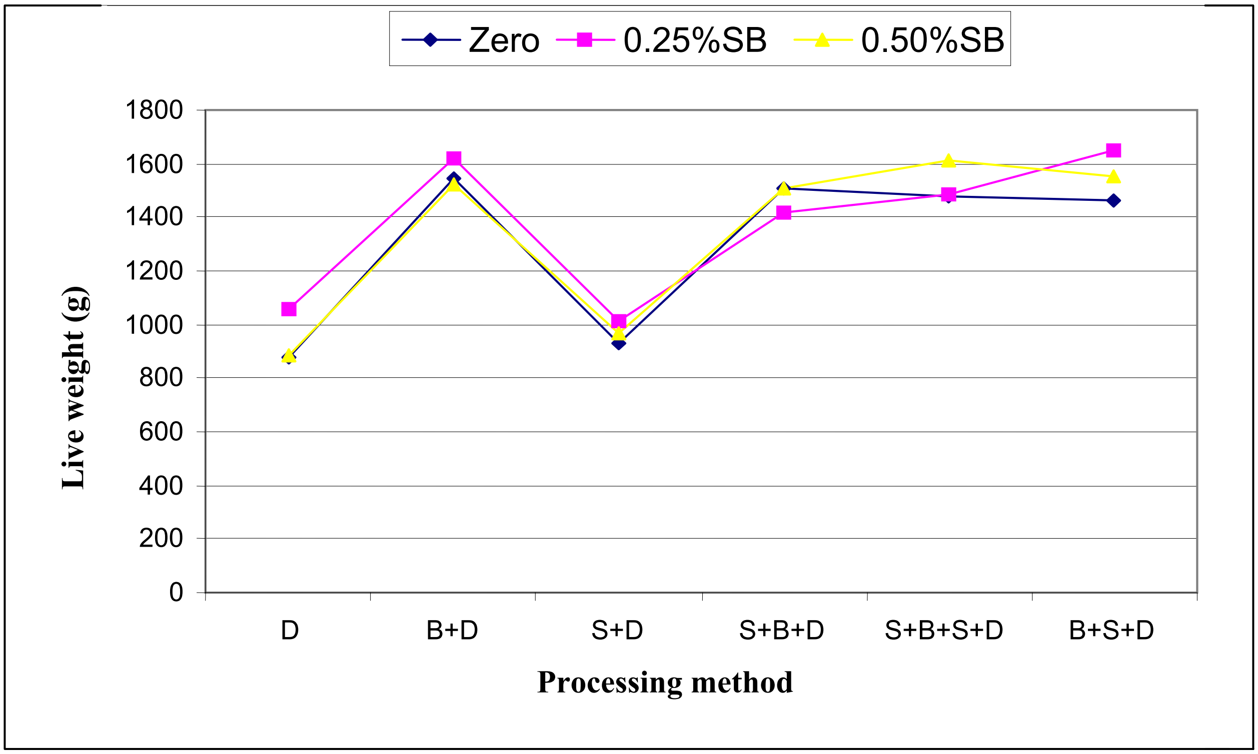

Figure 1. Live weight of broiler chickens fed for 41 d a diet containing *Mucuna* beans processed by different combinations of boiling (B), soaking (S), and drying (D), without sodium bicarbonate (“zero”), or with 0.25% or 0.50% of sodium bicarbonate (SB).

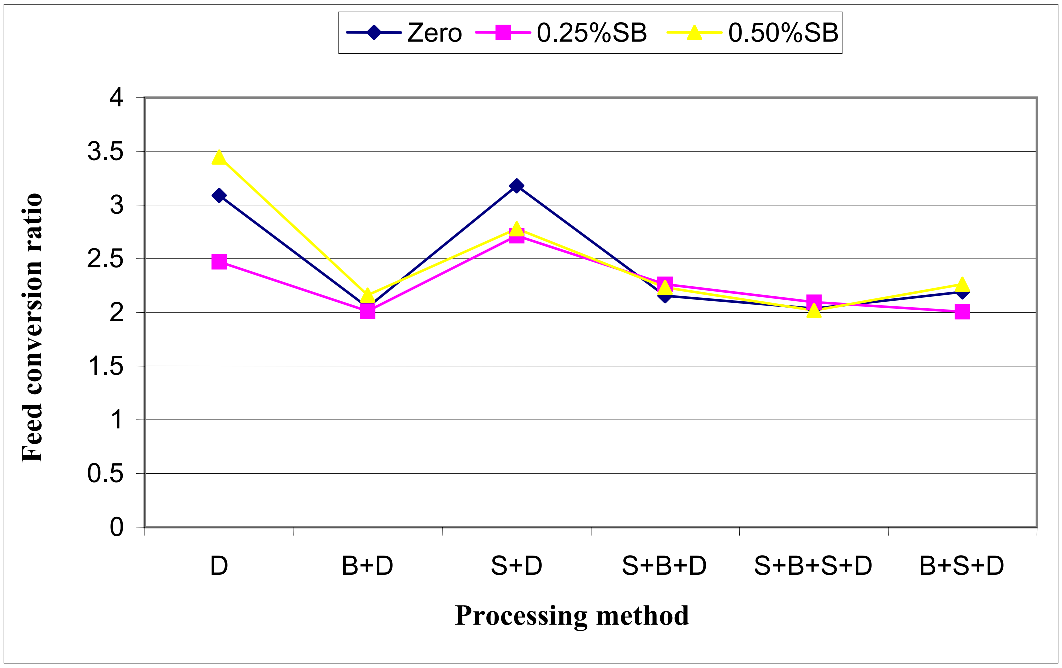

Figure 2. Feed Conversion Ratio of broiler chickens fed for 41 d a diet containing *Mucuna* beans processed by different combinations of boiling (B), soaking (S), and drying (D), without sodium bicarbonate ("zero"), with 0.25% or with 0.50% of sodium bicarbonate (SB).

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