OPEN ACCESS
Advances in Animal and Veterinary Sciences
Research Article

Effect of Replace Soybean Meal with Fermented Soybean Meal on Growth Performance, Nutrient Digestibility, Serum Urea Nitrogen Concentration and Diarrhea Incidence of Sucking Calves

Jianying Zhang[1,2], Shad Mahfuz[2,3], Bao Nan[2], Zhi Jie Yuan[2], Gui Xin Qin[2*]

[1]College of Life Science, Linyi University, Linyi-276000, China; [2]College of Animal Science and Technology, Jilin Agricultural University, Changchun-130118, Jilin, China; [3]Department of Animal Nutrition, Sylhet Agricultural University, Sylhet-3100, Bangladesh.

Abstract | This experiment as a 2 (soybean or fermented soybean) x 2 (drinking 6L or 4L milk every day) multifactor experimental design was carried out at Jilin Agricultural University, China to study the effects of soybean meal (SBM) fermented with *Bacillus subtilis,* on the growth performance, nutrient digestibility, blood urea nitrogen and diarrhea incidence in sucking Holstein calves. A total of 20 newborn Holstein calves (BW: 45.7±5.5 kg) were randomly allotted into four groups for 45 days, every group has five calves. Four experimental groups were: 6SBM= drink 6L milk daily and feed soybean meal; 6FSBM = drink 6L milk daily and feed fermented soybean meal; 4SBM = drink 4L milk daily and feed soybean meal; 4FSBM=drink 4L milk and feed fermented soybean meal. Results indicated that fermented soybean meal (FSBM) could improve average daily weight gain (ADG), feed efficiency, crude protein (CP) digestibility and crude fat (EE) digestibility compare with non fermented soybean meal ($P<0.05$). Calves fed with FSBM had significant lower urea nitrogen concentration in serum than SBM diet. Diarrhea score was lower (($P<0.05$) in calves fed FSBM than SBM diet. In conclusion, soybean meal can be replaced by fermented soybean meal on improving growth performance, nutrient retention and for reducing diarrhea incidence in calves.

Keywords | Fermented soybean meal, Calves, Growth performance, Nutrient digestibility, Diarrhea score

Received | December 23, 2019; **Accepted** | March 03, 2020; **Published** | May 15, 2020
***Correspondence** | Gui Xin Qin, College of Animal Science and Technology, Jilin Agricultural University, Changchun-130118, Jilin, China; **Email:** qgx@jlau.edu.cn;
Citation | Zhang J, Mahfuz S, Nan B, Yuan ZJ, Qin GX (2020). Effect of replace soybean meal with fermented soybean meal on growth performance, nutrient digestibility, serum urea nitrogen concentration and diarrhea incidence of sucking calves. Adv. Anim. Vet. Sci. 8(6): 608-613.
DOI | http://dx.doi.org/10.17582/journal.aavs/2020/8.6.608.613
ISSN (Online) | 2307-8316; **ISSN (Print)** | 2309-3331

INTRODUCTION

Soybean meal (SBM) is the most commonly used protein source in animal diet because of its excellent protein with amino acid, excellent nutrient availability and favorable palatability (Jeong and Kim, 2015; Wang et al., 2014). SBM is the most commonly used supplemental plant proteins for dairy cows (Mjoun et al., 2010; Awawdeh et al., 2007). A variety of anti nutritional factors (ANF) have limited its application in the diets for young animals (Wang et al., 2014; Rojas and Stein, 2013; Kim et al., 2010a). Also ANF of SBM damages the animal intestinal wall, increase diarrhea and reduce the absorption rate of vitamins and proteins (Song et al., 2010).

A fermentation process could reduce the ANF in SBM which increased the nutrients digestibility in animals (Roh et al., 2015; Upadhaya and Kim, 2015). Furthermore, a variety of essential nutrients, such as vitamins, are also produced during the process (Kim et al., 2012). Previous studies have reported that fermented soybean meal (FSBM) improved growth performance and reduced diarrheal incidence in *Escherichia coli* challenged piglets (Hung et al., 2008; Feng et al., 2007). However, data on the effects of FSBM on growth performance and health parameter in sucking calves are not readily available.

This experiment was aimed to study the effects of soybean meal (SBM) fermented with *Bacillus subtilis,* on per-

formance, nutrient digestibility and blood urea nitrogen concentration in sucking Holstein calves. We hypothesized that calves fed with FSBM diet can improve growth performance via improving nutrient retention during the sucking period.

MATERIALS AND METHODS

Preparation of Fermented Soybean Meal

The bacteria were obtained from the Jilin Agricultural University, as *Bacillus subtilis* (Patent No:201510205215.1). The solid-state fermentation culture medium (containing 2.64kg soybean, 0.37kg wheat bran, 0.09kg sucrose and 2.48L distilled water) was mixed with probiotics (0.31L *Bacillus subtilis*), incubated at 37˚C for 24h.

Animals and Feeding

A total of 20 newborn Holstein calves (BW: 45.7±5.5 kg) were randomly allotted to four groups, every group has five calves. Each calf was in a single cage and a mechanical ventilation system. The calf was fed milk using plastic bottles (2L capacity) and fed diet three times a day. Liger-EDTA (0.4g) was provided in 1L milk and Tio_2 (0.4g) was provided in 100g diet at 20-30d for nutrient digestibility test.

Date Recording and Sample Collection

The feed intake was recorded daily. Body weight was measured on every phase of evaluation and blood was collected in 5-mL coagulation-promoting tubes (BD, USA) from the jugular vein at initial day, 20 day and 45 day. Serum was collected from coagulation blood samples after centrifugation at 1,600 × g at 4˚C for 15 min and stored at -80˚C until subsequent assays were conducted. Feces sample was collected from 23d to 30d, 10% sulfuric acid were added in feces and then keep it into the -20˚C for further analysis.

Growth Performance and Feed Efficacy

Growth performance results as average daily gain (ADG) and feed efficiency were calculated at phase I (1-20d), phase II (21-45d), phase III (1-45d).

Chemical Analysis

All of feed and fecal sample were analyzed for dry matter (DM) by an oven at 105˚C for 5 h. Percentages of crude protein and crude fat were measured following the method AOAC (2004). Serum urea nitrogen (UREA) in whole blood was determined using fully automatic blood biochemical analyzer.

Diarrhea Scoring

Diarrhea scoring was done followed the method by Kim et al. (2012). In brief, fecal scoring composed of fecal fluidity, consistency, and odor was conducted daily at 0800 h as follows: for fecal fluidity, normal=1, soft = 2, runny = 3, and watery = 4; for fecal consistency, normal = 1, foamy = 2, mucouslike = 3, sticky = 4, and constipated = 5; for fecal odor, normal = 1, slightly offensive = 2, and highly offensive= 3.

Statistical Analyses

Experiments were analyzed as multifactor experimental design, and analysis of variance was conducted on all data using the general liner models (GLM) followed SPSS 17.0. A significant level of 0.05 was used.

RESULTS

Chemical Composition of Diets

Diets were formulated to meet or exceed the nutrient requirements by the NRC (2001) for calves. Nutrient and feed ingredients composition of the basal diet are shown in Table 1.

Body Weight Gain

As shown in Table 2. Average daily gain (ADG) of calves fed FSBM was significantly (P<0.01) improved in 6L milk at phase I (1-20d), and was increased (P <0.01) in 4L milk at phase II (20 to 45d). But the interaction between fed milk and diet was not significant (P=0.071). Compared to calves fed with SBM, FSBM significantly increased ADG in fed 6Lmilk and diet at phase III (1-45d), but the interaction between fed milk and diet was no significant.

Feed Efficiency

As shown in Table 3. Feed efficiency of calves fed FSBM were significantly (P<0.01) decreased in 6L milk during phase I (1-20d), but at Phase II (20 to 45d) feed efficiency was significant decreased in calves fed with 4L milk and diet (P<0.01; P<0.05). However, the interaction between fed milk and diet was no significant (P=0.186). At phase III (1-45d), FSBM significantly increased feed efficiency in calves fed with 4L milk and diet (P<0.05). But the interaction between fed milk and diet was no significant.

Nutrient Digestibility Analysis

The effects of dietary treatment on nutrient digestibility were shown in Table 4. Compared with SBM, FSBM could significantly increase the digestibility of CP in calves fed with 6 L milk and diet (P<0.05; P<0.01).The digestibility of EE was improved in calves fed with 6 L milk and diet (P<0.05; P< 0.01) and the interaction between milk and diet was significant (P< 0.05).

Serum urea-nitrogen Concentration

The effects of the different feeding conditions on urea-nitrogen concentration in serum of calves were shown in Table 5. At 20d, urea concentration in serum was decreased in calves fed FSBM compared with SBM (diet effect, P<0.05)

Table 1: Nutrition and ingredients composition (%) of the soybean meal (SBM) and fermented soybean meal (FSBM) in experimental diets

Items	SBM	FSBM
Ingredient,%		
Corn	49	50
SBM (42% crude protein)	26	-
FSBM (44% crude protein)	-	25
Cottonseed meal	5	5
Distiller's dried grains with solubles	11.8	11.8
Wheat bran	5	5
Calcium hydrogen phosphate	0.5	0.5
Sodium chloride	0.4	0.4
Mountain flour	0.8	0.8
Sodium bicarbonate	1	1
Premix[1](vitamins and trace minerals)	0.5	0.5
Analyzed chemical composition ,%		
Metabolic energy, kcal/g	13.46	13.79
Crude protein	21.45	21.14
Calcium	0.55	0.47
Phosphorus	0.57	0.41
Sodium	0.38	0.37
Chlorine	0.32	0.31

[1]Premix supplied the following nutrients per kilogram of mixed feed: vitamin A, 1200KIU; vitamin D_3, 400KIU; vitamin E,12000IU; $CuSO_4 \cdot 5H2O$, 4000 mg; $ZnSO_4$, 10000 mg; $MnSO_4$, 8000mg; CoCL, 60mg;KI, 160mg; Na_2SeO_3,140mg and $FeSO_4$,2000mg.

Table 2: Effect of experimental diets on average daily weight (g) gain in calves

	Treatment[2]		Significance(P-value)[3]		
Items[1]	SBM	FSBM	M	B	M×B
Phase I (1-20d)					
6L milk	290±80	360±80	**		
4L milk	100±40	110±30			
Phase II (21-45d)					
6L milk	600±140	650±30			
4L milk	500±110	710±30		**	0.071
Phase III (1-45d)					
6L milk	430±40	475±40	**	**	
4L milk	320±70	380±50			

[1]6L milk=feeding 6Lmilk each calf daily; 4L milk=feeding 4Lmilk each calf daily.
[2]SBM= soybean meal; FSBM=fermented soybean meal.
[3]M=milk effect; B=diet effect; M×B= the interaction between milk and diet effect. **P<0.01.

Table 3: Effect of feed efficiency in calves

Items[1]	Treatment[2]		Significance(P-value)[3]		
	SBM	FSBM	M	B	M×B
Phase I (1-20d)					
6L milk	2.64±0.86	2.11±0.65	**		
4L milk	5.83±1.55	5.39±0.96			
Phase II (21-45d)					

6L milk	2.06±0.34	1.87±0.14			
4L milk	1.91±0.20	1.62±0.02	**	*	0.186
Phase III (1-45d)					
6L milk	2.32±0.29	2.42±0.31			
4L milk	1.92±0.23	2.00±0.58		*	

[1]6L milk=feeding 6Lmilk every calf daily; 4L milk=feeding 4Lmilk every calf daily.
[2]SBM= soybean meal; FSBM=fermented soybean meal.
[3]M=milk effect; B=diet effect; M×B= the interaction between milk and diet effect.
**P<0.01; *P<0.05.

Table 4: Nutrient digestibility in calves

Items[1]	Treatment[2]		Significance(P-value)[3]		
	SBM	FSBM	M	B	M×B
CP (%)					
6L milk	67.24±2.82	80.02±4.90	*	**	0.067
4L milk	67.56±2.49	72.09±4.39			
EE (%)					
6L milk	81.73±3.88	91.01±1.71	*	**	*
4L milk	82.33±1.77	83.07±3.01			

[1]Cp=crude protein; EE=crude fat. 6L milk=feeding 6Lmilk every calf daily; 4L milk=feeding 4Lmilk every calf daily.
[2]SBM= soybean meal; FSBM=fermented soybean meal.
[3]M=milk effect; B=diet effect; M×B= the interaction between milk and diet effect.
**P<0.01; *P<0.05.

Table 5: Effect fermented soybean meal on urea-nitrogen concentration

Items[1]	Treatment[2]		Significance(P-value)[3]		
	SBM	FSBM	M	B	M×B
20d					
6L milk	3.53±0.59	2.90±0.64	0.092	*	
4L milk	4.17±0.51	3.15±0.43			
45d					
6L milk	3.65±0.64	3.10±0.56	*	*	
4L milk	4.29±0.45	3.87±0.42			

[1] 6L milk=feeding 6Lmilk every calf daily; 4L milk=feeding 4Lmilk every calf daily.
[2]SBM= soybean meal; FSBM=fermented soybean meal.
[3]M=milk effect; B=diet effect; M×B= the interaction between milk and diet effect.
**P<0.01; *P<0.05.

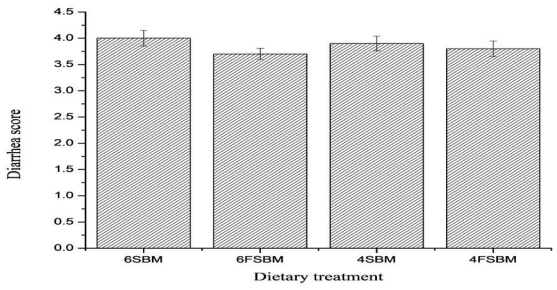

Figure 1: Effect of different treatment on Diarrhea incidence of calves. 6SBM=6L milk and soybean meal; 6FSBM=6L milk and fermented soybean meal; 4SBM=4L milk and soybean meal; 4FSBM=4L milk and fermented soybean meal.

Similarly, at day 45, urea-nitrogen concentration was decreased in calves fed with FSBM compared with SBM (milk and diet effect, P<0.05).

Diarrhea Score

As shown in Figure 1, the diarrhea rate in calves fed 6L milk fermented soybean meal (6FSBM) was the most lowest (P<0.05) and the diarrhea rate in calves fed 6L milk with soybean meal (6SBM) was the most highest (P<0.05).

DISCUSSIONS

In this study, soybean meal was replaced 25% by fermented soybean meal in calves diet, could significantly improve

growth performance, feed efficiency, crude protein, crude fat digestibility, and could decrease serum urea concentration as well as diarrhea score on drinking 6Lmilk and 4L milk daily. Milk and diet was the main factor could improve ADG, feed efficiency and nutrient digestibility in calves. This study ensured the higher average daily body weight gain in calves fed with fermented soybean meal (FSBM). The present study hypothesized that the growth-promoting effects of calves may be due the improvement of nutrition retention especially crude protein (CP) and ether extract (EE) in calves fed with FSBM. A similar growth promoting effects of fermented soybean meal in calves were noted by Kim et al. (2012) who observed the higher body weight gain and higher dry matter intakes, when compared with those fed non fermented soybean meal (SBM) in calves at 4 wk of age. In addition, growth-promoting effect was also found by Wang et al. (2014) who reported that pigs fed soybean meal fermented with *Streptococcus thermophilus* and *Saccharomyces cerevisiae* had a greater average daily body weight gain (ADG) and improved gain/feed ratio than the control diets. Furthermore, Kim et al. (2014) showed soybean fermented with *Aspergillus oryzae* increased growth performance of nursery pigs. Improved growth performance in broilers fed with fermented soybean meal was also reported by Chun et al. (2014).

Our results showed an increase in nutrient utilization particularly protein and energy in calves fed with fermented soybean was also related with improve feed efficiency in FSBM groups. Feng et al. (2007a) reported that fermentation may reduce the anti nutritional factors like raffinose and stachyose in soybean meal which lead improve nutrient retention in calves. We also hypothesized that fermentation may increase the availability of nutrients in experimental fed group. It is certain that some of the anti-nutritional factors were eliminated through microbial fermentation, and nutrients were supposed to be more digested. Fermentation of soybean meal using *Bacillus* spp. has increased digestibility of soy proteins as well (Kiers et al., 2003).

Serum urea nitrogen is a microbial product that is known to have a negative health status in birds and animals (Mahfuz et al., 2020). In this report, calves fed with FSBM showed lower serum urea concentration than unfermented one which ensured the sound health status in experimental calves which was similar with previous report (Feng et al., 2007b).

In our study, the diarrhea score of newborn calves were reduced at the whole period, which was similar with Kim et al. (2010b) who reported that the incidence and severity of diarrhea of calves were significantly lower in the FSBM group compared to those in the SBM group at 21days and 42 days of age. In addition, Kim et al. (2012) noted that calves fed the FSBM had significantly lower fecal scores than those fed the SBM during both pre and post weaning periods.

CONCLUSION

Calves fed with fermented soybean meal (FSBM) diet could improve growth performance via improved nutrient retention, decrease serum urea concentration and lowered the incidence of diarrhea. Further experiment can be conducted to evaluate the health status of calves fed with FSBM in commercial level.

ACKNOWLEDGEMENTS

The work was supported by National Key R&D Program of China (No. 2018YFD0500600).

CONFLICT OF INTEREST

The authors claim that they have no competing interests.

AUTHORS CONTRIBUTION

All authors contributed equally in the planning of the study, drafting the manuscript. All of them approved the final version of the article.

REFERENCES

- AOAC (2004). Official Methods for Analysis, 13th Ed. Association of Official Analytical Chemists.Washington, DC.
- Awawdeh MS, Titgemeyer EC, Drouillard JS, Beyer RS, Shirley JE (2007). Ruminal degradability and lysine bioavailability of soybean meals and effects on performance of dairy cows. J. Dairy Sci. 90:4740-4753. https://doi.org/10.3168/jds.2007-0210
- Chun-Yi L, Lu JJ, Wu CP, Lien TF (2014). Effects of probiotics and bremelain fermented soybean meal replacing fish meal on growth performance, nutrient retention and carcass traits of broilers. Livestock Sci.163:94-101. https://doi.org/10.1016/j.livsci.2014.02.005
- Feng, J, Liu X, Xu ZR, Lu YP, Liu YY (2007). Effect of fermented soybean meal on intestinal morphology and digestive enzyme activities in weaned piglets. Dig. Dis. Sci. 52:1845-1850. https://doi.org/10.1007/s10620-006-9705-0
- Feng J., Liu X, Xu ZR., Liu YY., Lu YP (2007b). Effects of *Aspergillus oryzae* 3.042 fermented soybean meal on growth performance and plasma biochemical parameters in broilers. Anim. Feed Sci. Technol.134:235-242. https://doi.org/10.1016/j.anifeedsci.2006.08.018
- Hung ATY, Su TB, Liao CW, Lu JJ (2008). Effect of probiotic combination fermented soybean meal on growth

performance, lipid metabolism and immunological response of growing-finishing pigs. Asian J. Anim. Vet. Adv. 3:431-436. https://doi.org/10.3923/ajava.2008.431.436

- Jeong JS, Kim IH (2015). Comparative efficacy of up to 50% partial fish meal replacement with fermented soybean meal or enzymatically prepared soybean meal on growth performance, nutrient digestibility and fecal microflora in weaned pigs. Anim. Sci. J. 86:624-633. https://doi.org/10.1111/asj.12335
- Kiers JL, Meijer JC, Nout MJR, Rombouts FM, Nabuurs MJA, Meulen J (2003). Effect of fermented soya beans on diarrheoa and feed efficiency in weaned piglets. J. Appl. Micro. 95:545-555. https://doi.org/10.1046/j.1365-2672.2003.02011.x
- Kim SW, Heugten E, Ji F, Lee CH, Mateo RD (2010a). Fermented soybean meal as a vegetable protein source for nursery pigs: I. Effects on growth performance of nursery pigs. J. Anim. Sci. 88:214-224. https://doi.org/10.2527/jas.2009-1993
- Kim MH, Yun CH, Kim HS, Kim JH, Kang SJ, Lee CH, Ko JY, Ha JK (2010b). Effects of fermented soybean meal on growth performance, diarrheal incidence and immune-response of neonatal calves. Anim. Sci. J. 81, 475-481. https://doi.org/10.1111/j.1740-0929.2010.00760.x
- Kim MH, Yun CH, Lee CH, Ha JK (2012). The effects of fermented soybean meal on immunophysiological and stress-related parameters in Holstein calves after weaning. J. Dairy Sci. 95:5203-5212. https://doi.org/10.3168/jds.2012-5317
- Kim SW, Heugten E, Ji F, Lee CH, Mateo RD (2014). Fermented soybean meal as a vegetable protein source for nursery pigs: effects on growth performance of nursery pigs. J. Anim. Sci. 88:214-224. https://doi.org/10.2527/jas.2009-1993
- Mahfuz S, He T, Liu S, Wu D, Long S, Piao X (2019). Dietary inclusion of Mushroom (*Flammulina velutipes*) stem waste on growth performance, antibody response, immune status, and serum cholesterol in broiler chickens. Animals 9: 692. https://doi.org/10.3390/ani9090692
- Mjoun K, Kalscheur KF, Hippen AR, Schingoethe DJ (2010). Ruminal degradability and intestinal digestibility of protein and amino acids in soybean and corn distillers grains products. J. Dairy Sci. 93:4144-4154. https://doi.org/10.3168/jds.2009-2883
- NRC(2001). Nutrient requirements of dairy cattle, 7th Rev. Ed. Natl. Acad. Press, Washington, DC, USA.
- Roh SG, Jeffery A, Carrol L, Kim SW (2015). Effects of fermented soybean meal on innate immunity-related gene expressions in nursery pigs acutely challenged with lipopolysaccharides. Anim. Sci. J. 86:508-516. https://doi.org/10.1111/asj.12319
- Rojas OJ, Stein HH (2013). Concentration of digestible, metabolizable, and net energy and digestibility of energy and nutrients in fermented soybean meal, conventional soybean meal, and fish meal fed to weanling pigs. J. Anim. Sci. 91:4397-4405. https://doi.org/10.2527/jas.2013-6409
- Song YS, Pérez VG, Pettigrew JE, Martinez-Villaluenga C, Mejia EG (2010). Fermentation of soybean meal and its inclusion in diets for newly weaned pigs reduced diarrhea and measures of immuno reactivity in the plasma. Anim. Feed Sci. Technol.159: 41-49. https://doi.org/10.1016/j.anifeedsci.2010.04.011
- SPSS (2006). Statistical software package for the social sciences SPSS Inc., IL. 20.0.
- Upandhaya SD, Kim IH (2015). Ileal digestibility of nutrients and amino acids in unfermented, fermented soybean meal and canola meal for weaning pigs. Anim. Sci. J. 86:408-414 https://doi.org/10.1111/asj.12305.
- Wang Y, Liu XT, Wang HL, Li DF, Piao XS, Lu WQ (2014). Optimization of processing conditions for solid-state fermented soybean meal and its effects on growth performance and nutrient digestibility of weanling pigs. Lives. Sci. 170: 91-99. https://doi.org/10.1016/j.livsci.2014.07.020