Article

Performance and some blood biochemical in male Holstein calves fed by different levels of calcium salts of fatty acids supplementation

Ahad Bigdeli Khajehdizaji[1], **Rasoul Pirmohammadi**[2], **Akbar Taghizadeh**[3], **Ahad Golghasem Gharehbagh**[4], **Omid Hamidi**[1], **Zeinal Hemmati**[2]

[1]Department of Animal Sciences, Maragheh Branch, Islamic Azad University, Maragheh, Iran

[2]Department of Animal Sciences, Agriculture Faculty, Urmia University, Urmia, Iran

[3]Department of Animal Sciences, Agriculture Faculty, Tabriz University, Tabriz, Iran

[4]Agriculture Sciences Faculty, Payam e Noor University, Tehran, Iran

E-mail: bigdelikhajehdizaji1980@gmail.com

Received 16 December 2018; Accepted 1 January 2019; Published 1 March 2019

Abstract

This study was conducted to investigate the effect of different levels of calcium salts of fatty acids (CSFA) on performance in Holstein calves. A total of sixteen17 month of age male Holstein calves with average of (540±5 kg) weight were selected and divided into four groups in order to the experimental complete randomized block design. The calves were fed by control groups with no fat protected and 2, 4 and 6 percentages of fat protected oils until 60 days. Feed intake, Body weight gain and feed conversion ratio were measured. The blood samples were taken to investigate the serum content of plasma protein, glucose, calcium, phosphorus, cholesterol, triglyceride, LDL and HDL. Additionally, the fat and protein digestibility in each group were measured. Data from this study showed that the highest FI and BW and the better FCR was related to the calves fed by 2% fat protected supplement but there were no significant effects on calves performance between treatments. Feeding the fat protected supplementations lead to higher protein and fat digestibility none significantly. The higher use of calcium salts of fatty acids supplements lead to higher serum triglyceride, cholesterol, calcium and LDL significantly ($p \leq 0.05$). There were no significant differences between treatments about glucose, phosphorous and protein content in the calves feed by different levels of calcium salts of fatty acids supplements. In conclusion, we could demonstrated that using of different levels of calcium salts of fatty acids may have beneficial effects on performance and some blood biochemical in the male Holstein Calves.

Keywords blood biochemical; calcium salts of fatty acids; performance; Holstein calves.

Proceedings of the International Academy of Ecology and Environmental Sciences
ISSN 2220-8860
URL: http://www.iaees.org/publications/journals/piaees/online-version.asp
RSS: http://www.iaees.org/publications/journals/piaees/rss.xml
E-mail: piaees@iaees.org
Editor-in-Chief: WenJun Zhang
Publisher: International Academy of Ecology and Environmental Sciences

1 Introduction

Fat supplementation also increases energy density of the diet, but high dietary fat can lead to a reduction in fiber digestion in the rumen and a decline in milk fat percentage, depending on the amount and type of fat fed

(Bines et al., 1978; Alyev, 1980). In order to counter affect these undesirable effects, dietary supplementation of fat as a salt of long chain fatty acids is a good alternative (Vovk et al., 2011).Saturated and unsaturated long chain fatty acids have less effect on rumen fermentation when supplemented as calcium salt than as free fatty acids (Drochner and Yildiz, 1999). Polyunsaturated fatty acids (PUFA), such as linoleic (LA) and linolenic (LNA) acids have been associated with enhanced reproduction of dairy and beef cows (Pantoja et al., 1994; Lopes et al., 2009). As demand for higher productivity increases, farmers of dairy cows are increasing faced with the need to supplement energy sources that could no longer be assuaged by the feeding of more dry matter (Grummer, 1991). Moreover, in the balance of economics, the cheapest energy source based one calorimetric contribution has to be derived from oils and fats. However the feeding of more oils and fats will invariably cause more harm than good because they break down in the rumen, into fatty acids (Staples and Thatcher, 2005). These same fatty acids will then be subjected to bio-hydrogenation processes that will inhibit microbial activities in the same rumen. If rumen activities are so crucial, then energy supplements via oils and fats will have to be modified in a way that it by-passes the rumen before it could be absorbed (Gassman et al., 2000). The Calcium Soap offers one such alternative. They offers beef cattle producers a method of increasing energy density in range supplements without harming forage utilization (Hightshoe et al., 1990). The calcium soap of long chain fatty acid is a type of rumen bypass fat made especially for high ruminants such as cows, goats and sheep (Doreau et al., 1993). The calcium bonding with fatty acids prevents the product from being reacted upon in the rumen stage thereby providing the crucial inertness. They stays inert in the rumen and then passes on through to the abomasums where the relative acidity condition there will naturally break down the calcium bonding and allow for optimal intestinal digestions (Ngidi et al., 1990). Formation of calcium soaps of fatty acids is another method of producing dry fats. Such products are easily handled and are more inert in the rumen than native fats. The unsaturated fatty acids in calcium soap products are only partially protected (Vovk et al., 2006). From rumen bio-hydrogenation and this protection is less complete with UFA than with the oleic acid. The experiences clearly showed that the calcium soaps consistently have a negative impact on dry matter intake (DMI). This has been stated by the National Research Council of Dairy Cattle, 2000) as well as by reviews of studies by other scientists. The NRC concluded that calcium soaps decreased DMI by 2.5% for each percentage unit added to the diet, while hydrogenated fatty acids or triglycerides did not affect DMI. The objective of this study was to measure the effect of calcium salt of fatty acid on performance and some blood biochemical of males Holstein calves.

2 Materials and Methods

2.1 Experimental procedures

The 17 month of age sixteen male Holstein calves with average of (540±5 kg) weight were selected from the dairy cattle herd of instructional dairy farm of Tabriz, Iran. They were fed under a conventional feeding system with concentrate mixture as per requirements (NRC, 2000). The calves were divided into four groups in order to the experimental complete randomized block design. The calves were fed by control groups with no fat protected and 2, 4 and 6 percentages of fat protected oils (Energizer gold) for 60 days respectively. The general analysis and chemical compositions of fatty acids that used in this study are shown in Table 1.

Ingredients of diets were analyzed according to AOAC (2000) methods. Dry matter (DM) with putting feed in 100 °C for 24 h., method 967.03, ash by incineration in 550°C for 8 h; method 942 and crud protein (CP) by Kjeldahl procedure. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed according to two stage procedures described by Stern et al. (1997) and Van Soest et al. (1994). The experimental total mixed ration (TMR) diets were fed in the morning and afternoon and the animal was also vaccinated against tuberculosis before the start of the project. The ingredient composition of different concentrate mixtures is given in Table 2.

Table 1 General analysis and chemical composition of fatty acids of the experimental rations.

The Chemical Composition	**(DM basis %)**
Metabolizable energy (M j /kg)	31
Ca %	8.5-9.5
P %	5.82
Fat content %	2.5-5
Moisture %	0.5
Fatty acid Contents	
Lauric acid (C12:0)	0.2
Myristic acid (C14:0)	1.2
Palmitic acid (C16:0)	47
Stearic acid (C18:0)	5
Oleic acid (C18:1)	38
Linoleic acid (C18:2)	8

Table 2 Ingredients and chemical composition of the experimental rations.

Ingredients (DM basis %)	Control	Protected fat (2%)	Protected fat (4%)	Protected fat (6%)
Alfalfa hay	15	15	15	15
Corn Silage	10	10	10	10
Barely	20	18	12	10
Wheat bran	4.6	6.5	20	24
Biscuit waste	15	15	15	15
Soybean meal	2	2	2	2
Maylo	30	28	19.7	15.1
Protected Fat	0	2	4	6
Nacl	0.5	1	0.5	1
Calcium bicarbonate	0.5	0.5	0.1	0.2
Urea	1	1	0.9	1
Mineral and vitamin per mix*	0.5	0.5	0.5	0.5
Chemical Composition				
Crude protein	14.72	14.58	14.45	14.61
Metabolizable energy(M cal/kg DM)	2.59	2.57	2.56	2.50
Net Energy maintenance (M cal/kg DM)	1.62	1.59	1.56	1.50
Net Energy growth(Mcal/kg DM)	1.05	1.02	1.02	1.00
Ca %	1.1	0.86	0.81	0.81
P %	0.51	0.43	0.40	0.40
Concentrate to forage ratio %	25.75	25.75	25.75	25.75

Calculated using published values of feed ingredient NRC (2001) by UFFDAM software. *Supplies per kg of feed: 500000 IU Vitamin A, 100000 IU of Vitamin D_3, 100 IU Vitamin E, 180000 mg of Calcium, 90000 mg of Phosphorous, 20000 mg of Magnesium,300 mg of Cu, 3000 mg of Fe, 3000 mg of Zn, 100 mg of Cobalt and 100 mg of Iodine.

2.2 Sampling of data

Samples of food and stools of calves in each group were taken from in morning and afternoon for 5 day. Then the samples were mixed and the same mixture was prepared, grinded and assessed by the method described by (Van Keulen and Young, 1977). Blood samples were taken and collected from the coccygeal vein or artery into commercial blood collection after the morning supplement feeding (3 hours after feeding) for determination of serum concentrations of plasma protein, glucose, calcium, phosphorus, cholesterol, triglyceride, LDL and HDL. The blood tubes placed on ice immediately and centrifuged at 1500 × g for 30 min for serum collection. Harvested serum was stored frozen at –20 °C until laboratory analysis.

2.3 Statistical analysis

According to the treatments and replications a randomized complete block design was used in the current study and the statistical model was: Yij=μ+Ti+βj+εIj, where Y_{ij} is the individual observation, μ is the overall mean, T_i is the effect of treatment, T_i is the effect of blocks and e_{ij} is the remainder effect.

The data were analyzed in a completely randomized design using the GLM of (SAS, 2001). The means were compared with Duncan multiple range test and P value less than 0.05 was considered as significant.

3 Results

The result of using different levels of calcium salts of fatty acids on performance of calves are shown in tabe3. As the result showed that using of different levels of calcium salts of fatty acids had some goods on feed intake and body weight gain in experimental Holstein male calves. The highest feed intake and body weight gain were related to calves fed by 2% of calcium salts of fatty acids compared to the others. Also the lower FCR was seen were the 2% of calcium salts of fatty acids fed to the calves. In this case there were no significant differences about performance between treatments.

Table 3 The effect of using different levels of calcium salts of fatty acids on performance of calves.

Treatment	FI (Kg)	BW (Kg)	FCR
Control	871.6[a*]	1.190[a]	12.38[a]
2%	1111.5[a]	1.505[a]	11.78[a]
4%	936.7[a]	1.087[a]	13.91[a]
6%	940.7[a]	1.360[a]	13.57[a]
SEM*	97.87	0.147	1.08
p-value	0.21	0.24	0.55

abc: Means within columns with differing letters are different (p<0.05); *SEM=Standard Error Mean.

The protein and fat digestibility were not affected by the influence of the calcium salts of fatty acids dietary intake. Although their digestibility increased but there were no significant differences compared to the control group.

Table 4 The effect of using different levels of calcium salts of fatty acids on fat and protein digestibility percentage.

Treatment	Protein (%)	Fat (%)
Control	73.12[a*]	70.77[a]
2%	73.48[a]	90.83[a]
4%	76.34[a]	94.08[a]
6%	77.44[a]	92.11[a]
SEM**	1.58	3.40
p-value	0.21	0.001

abc: Means within columns with differing letters are different (p<0.05); **SEM=Standard Error Mean

Data showed that some blood biochemical such as glucose, phosphorus and protein didn't differed significantly. The serum blood triglyceride, cholesterol, calcium, HDL and LDL were increased when the calves fed by 2, 4 and 6 percentage of calcium salts of fatty acids respectively ($p \leq 0.05$).

Table 5 The effect of using different levels of calcium salts of fatty acids on some blood biochemical.

Treatment	Glucose (Mg.dl)	Triglyceride (Mg.dl)	Cholesterol (Mg.dl)	Calcium (Mg.dl)	Phosphorus (Mg.dl)	Protein (g.dl)	HDL (Mg.dl)	LDL (Mg.dl)
Control	70	17[b]	111[b*]	9.35[b]	5.87	8.77	31[b]	61.1[b]
2%	68	18.25[ab]	174.75[ab]	10.45[a]	5.75	8.95	47.5[a]	99.8[ab]
4%	63.25	24.25[ab]	182.50[ab]	9.40[b]	5.92	8.35	42.2[ab]	114.3[ab]
6%	78.33	27.00[a]	232.33[a]	10.76[a]	6.64	8.80	51.3[a]	149.9[a]
SEM*	5.20	2.8	21.77	0.296	0.412	0.361	4.40	17.2
p-value	0.67	0.09	0.06	0.01	0.91	0.67	0.08	0.08

abc: Means within columns with differing letters are different (p<0.05); *SEM=Standard Error Mean.

4 Discussion

There is some evidences that feeding fatty acids improved the energy status of dairy cows (Staples et al., 1998), an enhanced performance may occur apart from an improved energy status of the animals (Sklan et al., 1991). The effects of fat supplementation on insulin concentrations are varied. Reis et al., 2012 showed that insulin increase, and Relling and Reynolds (2007) mentioned that decrease in concentrations of insulin after fat supplementation. Insulin concentrations reflect energy intake, and therefore, differences in insulin concentrations can be due to differences in energy status (Lucy et al., 1991). Lopes et al. (2009) did not observe differences in serum concentrations of insulin in beef cows supplemented with up to 0.20 kg per day rumen-inert PUFA. They suggested that rumen-inert dry fats are used to avoid interference with rumen function that in turn could decrease nutrients supplied by the rest of the ration ingredients.

It is known that application of fat supplements in a diet of cattle increases their caloric content and makes positive impact on milk productivity, feed payment as well as growth and development of animals (Vovk et al., 2006). An increased level of vegetable fats in a diet of ruminants makes sparing action on activity of microbial flora of fore stomach, making negative influence on fermentation processes in a rumen and absorption of nutrients in general (Pantoja et al., 1994). That is why lately protection of fat supplements in a diet is used before feeding in order to decrease negative effect of fat supplements on metabolic activity of microbial flora of fore stomach as well as to lower the level of hydrogenation of polyene fatty acids in four stomach of the given kind (Wu et al., 1991). Application of protected fatty acids on a base of oil in a diet of cattle makes a positive impact on their productivity and increases level of polyene fatty acids in a content of lipids of milk, organs and tissues, improving their quality (Enjalbert et al., 1997).

Calcium salts of long-chain fatty acids are a high-energy ingredient that is highly digestible in the small intestine, does not interfere with rumen function, and provides an optimum fatty acid profile (Gassman et al., 2000). Feeding of cattle with CSFA, made on the base of vegetable oils, increases share of unsaturated fatty acids in the content of tissues in a body (Sklan et al., 1991).

Feeding Ca salts of PUFA to dairy cows increased milk production, did not alter feed intake, and reduced pregnancy losses per service. Further, the total daily amount of Ca salts of PUFA should be fed during the first feeding of the day to optimize its benefits on pregnancy maintenance of dairy cows (Vieira et al., 2010).

It is also more likely that protozoa number may not have been affected due to Ca-FA rather than the consumption of diet as a whole as the source off at is protected from ruminal degradation (Bhatt et al., 2009).

In this study the digestibility of dry matter and crude protein were not significantly affected by the addition of CSFA. But there was a trend for increased digestibility of fat and protein with CSFA supplementation. Calcium salts of fatty acids have increased the apparent digestibility of total lipid in other studies when compared with digestibility of control rations (Hightshoe et al., 1990). The improved digestibility of total lipid suggests that added fat is perhaps more digestible than the lipid fraction of an un-supplemented diet.

Grummer (1991) hypothesized that concentration of glucose was not affected by CSFA and Plasma triglyceride and free fatty acids concentration increased as a result of CSFA supplementation. He also mentioned that the effect of feeding rumen protected fat on plasma glucose concentration is variable.

Bhatt et al (2018) showed that the Ca-FA supplementation improved dressing percentage both at 2% and 4% CA-FA inclusions levels with no change in fat yield and composition. It is concluded that supplementation of Ca-FA prepared from industrial-grade rice bran oil in growing lamb rations improved gain, organic matter intake and digestibility during the post-weaning period and a linear response to the level of supplementation recommends Ca-FA at 40 g/kg for improved lamb performance.

Research showed unsaturated soaps were generally more digestible (Enjalbert et al., 1997) and tended to dissociate at a higher pH compared with those that were more saturated.

Pantoja et al. (1994) and Wu et al. (1991) showed that the unique properties of unsaturated Fatty acids allowed for DM intake that was similar to tallow supplemented diets but did not negatively influence digestion and metabolism as would be expected with diets containing unprotected fats. Studies generally agreed that digestibility of major nutrients such as organic matter, starch, nitrogen, and energy were similar while NDF and ADF digestion was improved when unsaturated fatty acids was supplemented compared with tallow and vegetable fat supplemented at the same level (Doreau et al., 1993; Zinn, 1996; Ngidi et al., 1990).

5 Conclusion

In conclusion, application of supplements of calcium salts of fatty acids, in a diet of cattle makes positive impact on intensity of growth of calves. It also increases a share of unsaturated fatty acids in the content lipids of tissues of calves. Supplementation of different levels of calcium salts of fatty acids had beneficial effects on performance and fat and protein digestibility in male Holstein calves. Additionally, the triglyceride, cholesterol, calcium, HDL and LDL serum contents were increased when the calves fed calcium salts of fatty acids. Also Further tests are needed to explore and more detail explanation.

Acknowledgment

The authors would like to express special thanks to the vice chancellor for research on Department of Animal Science, Tabriz, Iran to support for the research.

References

AOAC.2000. Official Methods of Analysis, Association of Official Analytical Chemists. AOAC press, Gaithersburg, USA

Alyev A. 1980. Lipid Metabolism and Productivity Of Ruminants. 381.

Bhatt RS, Sahoo A, Shinde AK, Karim SA. 2018. Effect of calcium salt of fatty acids supplementationon performance of Malpura lambs. Animal Production Science, 55(9): 1123-1130

Bhatt RS, Tripathi MK, Verma DL, Karim SA 2009. Effect of different feeding regimes on pre-weaning growth, rumen fermentation and its effect on post weaning performance of lambs. Journal of Animal Physiology and Animal Nutrition, 93: 568-576

Bines JA, Brumby PE, Stony JE, Fulford RJ, Braithwaite GD. 1978. The effect of protected lipids on nutrient intakes, blood and rumen metabolites and milk secretion in dairy cows during early lactation. Journal of Agricultural Science, 91: 135

Doreau, M, Ferlay A, Elmeddah Y. 1993. Organic matter and nitrogen digestion by dairy cows led calcium salts of rapeseed oil fatly acids or rapeseed oil. Journal of Animal Science, 71: 499-504

Drochner W, Yildiz G. 1999. Rumen fermentation and digestibility of nutrients studied by the addition of Ca soaps of palm oil fatty acids and their analogous fatty acids in the sheep model. Berl. Munch. Tierarztl.Wochenschr., 112(12): 472-478

Enjalbert F, Nicol MC, Bayourthe C, Vernay M, Moncoulon R. 1997. Effects of dietary calcium soaps on digestion, milk composition, and physical properties of butter. Journal of Dairy Research, 64: 181-195

Gassman K, Beitz D, Parrish F. 2000. Effects of feeding calcium salts of conjugated linoleic acid to finishing steers. Journal of Animal Science, 78: 275-276

Grummer R. 1991. Effect of feed on the composition of milk fat. Journal of Dairy Science, 74(9): 3244-3257

Grummer RR. 1988. Influence of prilled fat and calcium salts of palm oil fatty acids on nominal fermentation and nutrient digestibility. Journal of Dairy Science, 71: 117

Hightshoe RB, Cochran RC, Corah LR, Harmon DL, Vanzant ES. 1990. Influence of level and source of rumen- escape lipid in a supplement on forage intake and digestibility. Journal of Animal Science, 68 (Suppl. 1): 571

Lopes CN, Scarpa AB, Cappellozza BI, Cooke RF, Vasconcelos JLM. 2009. Effects of rumen-protected polyunsaturated fatty acid supplementation on reproductive performance of *Bos indicus* beef cows. Journal of Animal Science, 87: 3935-3343

Lucy MC, de la Sota RL, Staples CR, Thatcher WW. 1993. Ovarian follicular populations in lactating dairy cows treated with recombinant bovine somatotropine (Sometribove) or saline and fed diets differing in fat content and energy. Journal of Dairy Science, 76: 1014-1027

Ngidi ME Welch SC, Fluharty FL, Palmquist DL. 1990. Effects of calcium soaps of long-chain fatly acids on feed lot performance, carcass, characteristics, and ruminal metabolism of steers. Journal of Animal Science, 68: 555-565

Official Methods of Analysis. 1990. Kjeldahl protein (crude) determination in animal feed, copper catalyst Kjeldahl method. (984.13). Association of Official Analytical Chemists (15th Edition)

Pantoja J, Firkins JL, Eastridge ML, Hull BL. 1994. Effects of fat saturation and source of fiber on site of nutrient digestion and milk production by lactating cows. Journal of Dairy Science, 77: 341-356

Reis M, Cooke R, Ranches J. 2012. Effects of calcium salts of polyunsaturated fatty acids on productive and reproductive parameters of lactating Holstein cows. Journal of Dairy Science, 95(12): 7039-7050

SAS Institute, SAS/STAT User's Guide for Personal Computer. 2001. Release 6.12 SAS Institute Inc., Cary, NC, USA

Sklan D, Moallem U, Folman Y. 1991. Effect of feeding calcium soaps of fatty acids on production and reproductive responses in high producing lactating cows. Journal of Dairy Science, 74: 510-517

Staples C, Thatcher W. 2005. Effects of fatty acids on reproduction of dairy cows. In: Recent Advances in Animal Nutrition. 229-256

Stern MD, Bach A, Calsamiglia S. 1997. Alternative techniques for measuring nutrient digestion in ruminants. Journal of Animal Science, 75: 2256-2276

Van Keulen J, Young BA. 1977. Evaluation of acid-insoluble ash as natural marker in ruminant digestibility. Journal of Animal Science, 44: 262-266

Van Soest PJ. 1994. Nutritional Ecology of The Ruminant (2^{th} ed). Cornell University Press. Ithaca, NY, USA

Vieira FVR, Lopes CN, Cappellozza BI, Scarpa AB, Cooke RF, Vasconcelos JLM. 2010. Effects of intravenous glucose infusion and nutritional balance on serum concentrations of NEFA, glucose, insulin, and progesterone in non-lactating dairy cows. Journal of Dairy Science, 93: 3047-3055

Vovk S, Pavkovych S, Martin M. 2006. Protected fats and fatty acids in the diets of cattle fodder. Bulletin of Agricultural Science, Special Issue: 83-86

Vovk S, Snitynskyy V, Pavkovych S, Kruzhel B. 2011. Fat Additives in Animal Feed and Poultry: monograph 208

Wu Z, Ohajuruka OA, Palmquist DL. 1991. Ruminal synthesis, biohydrogenation, and digestibility of fatly acids by dairy cows. Journal of Dairy Science, 74: 305-3034

Zinn RA, Shen Y. 1996. Interaction of dietary calcium and supplemental fat oil digestive function and growth performance in feedlot steers. Journal of Animal Science, 74: 303-309