Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
DOI: 10.29261/pakvetj/2019.100

RESEARCH ARTICLE

Effect of High Cassava Ration on Insulin Sensitivity and Clinical Signs of Laminitis in Dairy Heifers

R Pilachai[1,*], WH Hendriks[2,3], S Aiumlamai[4] and JTh Schonewille[3]

[1]Faculty of Technology, Udon Thani Rajabhat University, Thailand; [2]Wageningen University & Research, Wageningen, the Netherlands; [3]Faculty of Veterinary Medicine, Utrecht University, Utrecht, the Netherlands; [4]Faculty of Veterinary Medicine, Khon Kean University, Khon Kaen, Thailand
*Corresponding author: r.pilachai@udru.ac.th

ARTICLE HISTORY (19-220)

Received: May 25, 2019
Revised: August 01, 2019
Accepted: August 18, 2019
Published online: September 11, 2019

Key words:
Heifer
Insulin resistance
Insulin sensitivity
Laminitis
Rumen acidosis

ABSTRACT

The relationship between high intake of rapidly fermentable carbohydrates and the subsequent response on plasma glucose, insulin sensitivity and clinical signs of laminitis in dairy heifers was studied. Ten dairy heifers with a mean body weight of 275 kg (SD 13.8 kg), were parallelly subjected (n=5/treatment group) to either a total mixed ration (TMR) low (LC, 130 g DM/kg) or high (HC, 480 g DM/kg) in cassava (starch). Results showed that mean DM intake, rectal temperature, heart rate, respiration rate and rumen contractions were not affected by the dietary treatments. All heifers fed the HC-TMR had diarrhea while all the heifers fed the LC-TMR had a pasty, soft consistency of feces. Three of the 5 heifers fed the HC-TMR displayed clinical signs of acute laminitis compared to none in the LC-TMR group. Mean postprandial rumen pH values of heifers fed the HC-TMR, but not the LC-TMR, were lower than 6.0 at all the time points measured. The reduction in rumen pH was associated with 49% greater concentration of total VFA and a concomitant shift from acetic to propionic acid and increased rumen lactic acid concentrations. Moreover, the HC-TMR caused greater plasma insulin levels and insulin resistance. The three laminitis animals had 1.9 greater basal plasma insulin values and a 46% lower insulin sensitivity values, respectively compared to their non-laminitis counterparts fed the HC-TMR. Insulin sensitivity was shown to be associated with clinical signs of laminitis in heifers only when plasma insulin concentrations were greater than ~ 700 pmol/L.

To Cite This Article: Pilachai R, Hendriks WH, Aiumlamai S and Schonewille JTh, 2020. Effect of high cassava ration on insulin sensitivity and clinical signs of laminitis in dairy heifers. Pak Vet J, 40(1): 113-117. http://dx.doi.org/10.29261/pakvetj/2019.100

INTRODUCTION

Laminitis (*Pododermatitis diffusa aseptica*) is an important health problem in dairy cows and nutrition has been identified as a major contributing factor (Greenough *et al*., 2007). The precise etiology of laminitis still remains to be elucidated. High concentrations of endotoxins, histamine and lactic acid in rumen fluid and or plasma have been implicated to play an important role in the development of laminitis in dairy cattle (Nocek, 1997). The relevance of a number of these risk factors, however, has recently been questioned. Pilachai *et al*. (2012) and Pilachai (2013) observed cases of (sub) clinical laminitis that could not be explained by the observed concentrations of lipopolysaccharide (LPS), lactic acid or histamine in either rumen fluid or plasma. Moreover, systemic infusions of either LPS or histamine did not induce laminitis in horses (Boosman *et al*., 1991), thereby indicating that these components may be secondary metabolites associated with a hitherto unknown factor.

In equines, obesity and insulin resistance have been implicated to play a role in the etiology of laminitis (Treiber *et al*., 2005). Moreover, the role of glucose and insulin was experimentally confirmed by De Laat *et al*. (2010) who were able to induce laminitis by prolonged infusions of glucose with or without insulin in clinically healthy horses. It can be hypothesized that in bovines as well, insulin plays a role in the development of laminitis. In dairy cattle, laminitis typically occurs during the first week lactation (Tarlton *et al*., 2002) and insulin resistance also is often observed in early lactating cows (Kawashima *et al*., 2016). In bovines, however, the potential role of insulin in the development of laminitis has not yet been studied.

Early lactating cows are typically fed high concentrate rations to support milk production. High concentrate rations generally contain a high proportion of rapidly fermentable carbohydrates which not only entails a risk of rumen acidosis, but may also decrease insulin sensitivity (Holtenius *et al*., 2003). In earlier studies, Pilachai *et al*. (2012) successfully induced (sub) clinical laminitis with the use of high concentrate rations but in these studies, insulin sensitivity was not investigated.

The objective of the current study was to gain more insight into the relationship between high intakes of rapidly fermentable carbohydrates and the subsequent response in plasma glucose, insulin sensitivity and laminitis in dairy heifers. It was expected that (sub) clinical cases of laminitis in dairy cows are negatively associated with insulin sensitivity.

MATERIALS AND METHODS

Animals and housing: The current experiment was approved by the committee of Ethics of Animal Experimentation of Udon Thani Rajabhat University, Udon Thani, Thailand (ID. AREC. UDRU 02/2015 No. 0543.7/333). Ten, Holstein-Friesian (HF) × Brahman crossbred (HF >75%) heifers ranging from 15 to 20 mo. of age with a mean (±SD) body weight of 275±13.8 kg was individually housed under natural ventilation conditions. The concrete floor of the individual pens (3 × 3 m^2) was covered with a ~10 cm thick layer of sand bedding.

Experimental design and rations: The experiment involved 10 heifers which were parallelly subjected to two dietary treatments (*n*=5/treatment group) and consisted of a 7-day adaptation period followed by a 6-day measurement period. During the adaptation period, all 10 heifers were offered 2 kg of a total mixed ration (TMR) containing 130 g cassava chips (Table 1) with rice straw provided *ad libitum*. On the last day of the adaptation period, the heifers were randomly allocated to one of two following experimental rations: 1) a TMR similar to the one that was also fed during the adaptation period (low cassava TMR, LC-TMR) or 2) a TMR where the cassava chips and part of the rice straw were replaced (total 480 g /kg DM) by ground cassava chips (high cassava TMR, HC-TMR) (Table 1). All 10 animals received 12 kg dry matter (DM) of TMR which was offered daily in two equal portions at 08:00 and 17:00 h.

Intravenous glucose tolerance test: On the 6th day of the measurement period, all 10 animals were subjected to an intravenous glucose tolerance test (IVGTT) as described by Opsomer *et al.* (1999). Briefly, a blood sample was obtained, 15 min before 0.5 g/kg BW of glucose (1000 mL of a 50% sterile glucose solution) was manually infused over a 5 min period. Blood samples (~10 mL) were subsequently taken at 5, 10, 15, 30, 45, 60, 75, 90, 105 and 120 min after the infusion of the glucose solution. The sample was transferred into two tubes containing either K_2EDTA or NaF and both tubes were immediately centrifuged at 3,000 × *g* for 20 min and stored at -20ºC pending analysis.

Collection of samples and clinical observations: The TMRs were sampled daily and stored until chemical analysis. On day 5 of the measurement period, both blood and rumen fluid samples were taken ~15 min before the morning feeding and 2, 4 and 8 h after the morning feeding. Rumen fluid samples (~200 mL) were obtained by means of a stomach tube with a suction strainer and the pH was measured using a calibrated pH meter (Mettler-Toledo GmBh 8603, Schwerzenbach, Switzerland). The rumen fluid was centrifuged at 5,000 × *g* for 30 min with the supernatant collected stored at -20°C pending analysis.

All heifers were clinically monitored each day, including auscultation of heart rate and rumen contractions, rectal temperature and respiration frequency. The consistency of freely voided feces was inspected visually and graded during the day time (Hughes, 2001). Furthermore, all heifers were clinically examined daily for signs related to sub/acute laminitis as modified by Greenough *et al*. (2007). Briefly, the coronary band was observed and evaluated for a swelling and pink in color. Weight shifting was observed as a lateral transfer of weight by the animal from one leg to another in a monotonous manner.

Chemical analysis: The contents of DM, ash, ether extract and crude protein of the feedstuffs were determined to the procedure of the AOAC (1990). The contents of neutral- and acid detergent fiber were determined according to the method described by Van Soest *et al*. (1991).

Rumen fluid concentrations of volatile fatty acids (VFA) and lactate were determined by HPLC (Shimadzu Scientific Instruments Inc., Colombia, MD, USA) using an UV-Detector (210 nm) and a flow rate of 0.5 mL/min (Samuel *et al*., 1997). Insulin was measured in EDTA-plasma using a radioimmunoassay kit (Cisbio Bioassays, Codolet, France) and glucose was determined in NaF-plasma according to the hexokinase method (BS-400, Mindray Co., Ltd., Shenzhen, China).

Calculations and statistical analysis: The time patterns of the plasma glucose and insulin concentrations after the glucose challenge were used to calculate various indices of glucose tolerance (Opsomer *et al*., 1999). Briefly, the glucose clearance rate (*k*, %/min) was calculated on the basis of the glucose concentrations measured at 15 and 45 min (T15 and T45, respectively) after the start of the glucose infusion, i.e. 100 × [(log glucose T15 − log glucose T45)/(T45 − T15)]. The half-life ($T_{1/2}$; the time required for the glucose concentration to fall by one-half) was calculated as: 100 × ln 2/*k*. The areas under the curve (AUC) of both glucose and insulin were calculated using a trapezoid model. Insulin sensitivity was calculated using the homeostasis model assessment (HOMA) calculator (Diabetes Trials Unit, Oxford, UK). Insulin resistance was calculated as 1/ (insulin sensitivity/100).

Dry matter intake and selected indices of rumen fermentation were subjected to repeated measures analyses of variance (ANOVA) with dietary treatment as factor. Fisher's least significant difference test was subsequently used to compare the mean differences between the two treatments. Effects of dietary treatments on the consistency of feces and signs of subacute laminitis were determined using Fisher's exact test. Throughout, data are presented as a mean ± standard error. Statistical significance was preset at $P<0.05$.

RESULTS

Feed intake and clinical observations: Throughout the measurement period, the mean DM intake (DMI) was not significantly affected by the dietary treatments (Table 2). Mean rectal temperature, heart rate, respiration rate and rumen contractions were similar (P≥0.476) between treatments. From the 3rd day of the measurement period, all heifers fed the HC-TMR had a feces score of 4 while all the heifers fed the LC-TMR had a feces consistency of 3 (P=0.008). No clinical signs of laminitis were observed in the heifers fed the LC-TMR, while 3 out of the 5 heifers fed the HC-TMR displayed marked weight shifting (score 2) and swelling over the coronary band (score 1). The first marked weight shifting and swelling over the coronary band were observed on d 3 within 4 to 8 h after feeding. The difference in laminitis scores between the two dietary treatments, however, did not reach statistical significance (Table 2).

Indices of rumen fermentation and plasma concentrations of glucose and insulin: On the 5th day of the measurement period, mean rumen pH of the heifers fed the HC-TMR was found to 9.6% (P<0.001) lower compared to the heifers fed the LC-TMR (Table 3). Moreover, postprandial rumen pH of the heifers provided the HC-TMR was found to be <6.0 at each time point measured (i.e. mean postprandial rumen pH values were 5.99, 5.78 and 5.75 at 2, 4 and 8 h, respectively). The concentration of total VFA was 49% greater (P<0.001) with the feeding of the HC-TMR and this was associated with a shift in VFA from acetate to propionate; the proportions of acetate decreased by 14.7% units (P=0.001) while the proportion of propionate increased by 13.9% units (P=0.001). The proportion of butyrate was not affected (P=0.563) by the dietary treatment. The rumen lactate concentration was found to be 1.5 times greater (P=0.010) when the HC-TMR was provided. Mean pre- and postprandial plasma glucose concentrations were greater (P≤0.033) when the HC-TMR was provided (Table 3) and plasma insulin concentrations were likewise affected (P≤0.024).

Intravenous glucose tolerance test: Basal plasma glucose concentrations (i.e. the glucose concentrations measured 15 min prior to the start of the glucose infusions) were 23% greater (P=0.008) when the heifers were provided the HC-TMR (Table 4). The difference between the peak- and basal glucose values was similar between the two dietary treatments (P=0.614). The clearance rate of glucose was greater (P=0.026) when the heifers were offered the HC-TMR. The time required for the glucose concentration to fall by one-half ($T_{1/2}$) and the time that glucose fell to basal value were shorter (P≤0.041) after the feeding of the HC-TMR. Likewise, the area under the curve (AUC) of glucose for the two indicated time intervals (Table 4) was lower (P≤0.036) in heifers offered the HC-TMR.

Basal plasma insulin concentrations (Table 4) were greater (P=0.041) when the HC-TMR was fed. However, the difference between the rations did neither affect (P≥0.116) the insulin increment (i.e. peak minus basal values) nor the AUC, irrespective of the time frame used. Furthermore, the feeding of the HC-TMR increased 1.8 times the insulin resistance value (P=0.041) with a concomitant decrease of almost 40% (P=0.020) in insulin sensitivity.

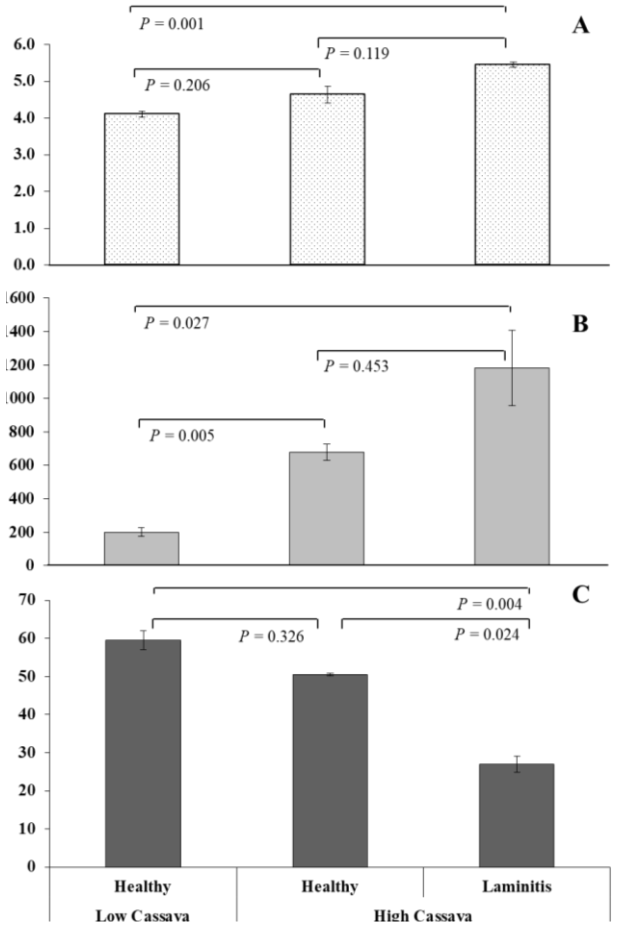

Fig. 1: Mean post feeding plasma glucose- (mmol/L, panel A) and insulin concentrations (pmol/L, panel B) at day 5 of the measurement period and the insulin sensitivity index (%, panel C) at d 6 of the measurement period (Glucose tolerance test) in heifers fed either a low or a high cassava containing TMR. Laminitis = heifers showing clinical signs of laminitis (*n* = 3), Healthy = heifers without clinical signs of laminitis (low cassava, *n* = 5; high cassava, *n* = 2). Error bar = SE. The indicated P values are calculated by means of an unpaired, two-sided *t*-test.

Table 1: Ingredient and analyzed composition of the experimental total mixed rations (TMR)

	Cassava containing TMR	
Composition	Low	High
Ingredient (g/kg of DM)		
Rice straw	500	150
Cassava chip	130	-
Ground cassava chip	-	480
Rice bran	100	100
Soybean meal	250	250
Premix[a]	10	10
Urea	5	5
Sulphur	5	5
Analyzed (g/kg of DM)		
DM (g/kg)	904	914
Ash	91	74
CP	152	161
NDF	428	242
ADF	291	166
Ether extract	2	0.2
NSC	328	523

NSC = non-structural carbohydrates = (1000 - ash - CP - NDF – Ether extract); [a] Mineral premix consisted of (g per kg): 10 Mg; 6 Zn; 8 Mn; 10 Fe; 1.6 Cu; (mg per kg) 100 I; 20 Co; 60 Se; 2,000,000 IU vitamin A; 400,000 IU vitamin D; 3000 IU vitamin E (TS Dairy mix®, Thailand).

Table 2: Mean dry matter intake (DMI) during the first 4 days of the measurement period, selected clinical parameters and laminitis score in heifers fed either a low or a high cassava total mixed ration (TMR)

Item	Cassava containing TMR		SEM	P-value
	Low	High		
DMI, kg/day	9.0	9.5	0.35	0.300
Rectal temperature, °C	38.7	38.6	0.19	0.998
Heart rate, beats/min	94.9	99.9	6.87	0.476
Respiration rate, times/min	52.8	55.4	8.25	0.756
Rumen motility, ontractions/5 min	7.1	7.4	0.46	0.575
Feces score	3	4	NA	0.008
Laminitis				
%	0	60	NA	0.167
No. of cases/No of animals	0/5	3/5		

NA=Not Applicable, Fisher's exact test.

Table 3: Mean values on selected indices of rumen fermentation and plasma concentrations of glucose and insulin in heifers fed either a low or a high cassava total mixed ration (TMR) at d 5 of the measurement period

Item	Cassava containing TMR		SEM	P-value
	Low	High		
Rumen[a]				
pH	6.57	5.94	0.033	<0.001
Total VFA (mM)	88.1	131.4	3.45	<0.001
Individual VFA (mol/100 mol)				
Acetic acid	75.8	61.1	2.06	0.001
Propionic acid	16.0	29.9	1.77	0.001
Butyric acid	8.2	9.1	1.02	0.563
Lactic acid (mM)[b]	2.3	3.8	0.30	0.010
Plasma				
Glucose (mmol/L)				
Pre-feeding	4.1	4.9	0.21	0.033
Post-feeding[c]	4.1	5.1	0.22	0.010
Insulin (pmol/L)				
Pre-feeding	153	795	70.0	<0.001
Post-feeding	199	982	200.0	0.024

[a] Indices of rumen fermentation represent the means of the pre-feeding values and the three values measured at 2, 4 and 8 h after feeding; [b] Total lactic acid (sum of D and L isomers); [c] Means of three values measured at 2, 4 and 8 h after feeding.

Table 4: Selected indices of the intravenous glucose tolerance test of heifers fed either a low or a high cassava containing total mixed ration (TMR) at day 6 of the measurement period

Item	Cassava containing TMR		SEM	P-value
	Low	High		
Glucose				
Basal value[a] (mmol/L)	4.0	4.8	0.16	0.008
Peak minus basal (mmol/L)	16.4	18.3	2.53	0.614
Clearance rate (/min)	0.44	0.59	0.039	0.026
$T_{1/2}$ (min)[b]	165	118	12.8	0.034
T_{basal} (min)[c]	170	112	16.8	0.041
AUC (mmol/L×min)[d]				
between 5 and 60 min	762	649	31.9	0.036
between 5 and 120 min	1253	1043	58.3	0.034
Insulin				
Basal value (pmol/L)	100	176	22.2	0.041
Peak minus basal (pmol/L)	452	652	112.5	0.244
AUC (pmol/L×min)				
between 5 and 60 min	19790	34060	5723.0	0.116
between 5 and 120 min	38964	62185	9874.3	0.135
Insulin resistance	1.7	3.1	0.40	0.041
Insulin sensitivity index[e]	59.6	36.4	5.62	0.020

[a]Values measured 15 min before the start of the glucose infusion; [b]Time required for glucose concentration to fall by one-half; [c]Time that glucose falls to its basal value; [d]Area under the curve; [e]Calculated with the use of the HOMA2 calculator (Diabetes Trials Unit, Oxford, UK).

DISCUSSION

To the best of the authors' knowledge, the current study is the first in heifers showing that clinical signs of bovine laminitis are associated with insulin resistance. Three out of the five heifers fed the HC-TMR developed clinical signs of laminitis and these animals had almost 18% greater plasma glucose values compared to their two non-laminitis counterparts fed the same ration (Panel A, Fig. 1). The greater plasma glucose values were associated with 1.7 times greater plasma insulin values (Panel B, Fig. 1). Thus, despite the greater plasma insulin levels in the animals that suffered from laminitis, they were not able to maintain their plasma glucose level around 4 mmol/L. These observations can be interpreted that those animals were insulin resistant (Kahn, 1978). This interpretation is in line with a 46% lower insulin sensitivity value (Panel C, Fig. 1) in the animals that suffered from laminitis, thereby confirming the notion that insulin sensitivity may play a role in the a etiology of bovine laminitis. Two heifers that were fed the HC-TMR did not develop clinical signs of laminitis although their plasma insulin levels were 3.4 times greater than those of the animals fed the LC-TMR. This observation cannot be explained by a difference in DMI but it can indicate that plasma insulin levels have to exceed a certain threshold for laminitis to occur. In the present study, clinical signs of laminitis only occurred in case mean post feeding plasma insulin concentrations were greater than ~700 pmol/L. This observation is in line with Walsh *et al*. (2009) who reported that the occurrence of laminitis in horses also is associated with plasma insulin concentrations of 695 pmol/L or greater. Needless to say that caution is warranted to generalize the outcome of the current study and future studies are required for confirmation.

In the current study, the mean postprandial plasma insulin concentration in heifers fed the LC-TMR, was found to be 199 pmol/L. Insulin concentrations between 14-226 pmol/L are considered to be within the physiological range as observed in cattle (Radostits *et al*., 2000) and are in line with values observed in healthy, early lactating cows fed a low starch diet; i.e. 135 pmol/L (van Knegsel *et al*., 2007) and 118 pmol/L (Kerestes *et al*., 2009).

The current observations with respect to the occurrence of laminitis (3 of 5 heifers in the HC-TMR group) are consistent with previous work of Pilachai *et al*. (2012) and Pilachai (2013). In these studies, 3 out of the 4 (Pilachai *et al*., 2012) and 4 out of the 6 (Pilachai, 2013) animals showed clear signs of subacute laminitis when high cassava rations were fed. In these experiments, the cassava rich treatments caused increased rumen VFA concentrations with a concomitant shift from acetic to propionic acid and increased rumen lactic acid concentrations (Pilachai *et al*., 2012; Pilachai, 2013). In the current study, rumen fermentation was likewise affected by the feeding of the HC-TMR. The afore-mentioned results on rumen fermentation are in line with the generally accepted thesis that a high intake of readily fermentable carbohydrates, such as ground cassava chips, increase the production of VFA and lactic acid which in turn reduce the pH of rumen fluid (Plaizier *et al*., 2008). Interestingly, Nocek (1997) implicated the condition of rumen acidosis in the etiology of bovine laminitis, but a clear mode of action was not postulated. In the current study, insulin resistance occurred in association with a high intake of readily fermentable carbohydrates (starch), thereby,

implicating that fermentation products such as VFA and/or lactic acid may play a role in the development of insulin resistance. A clear relationship between rumen acidosis and insulin resistance has not been identified yet, but it is known that propionic acid can stimulate the pancreatic secretion of insulin in ruminants (Gong *et al*., 2002). Furthermore, Oh *et al*. (2015) demonstrated that in steers, serum insulin responded to intra-ruminal infusions of propionate. Thus, it can be speculated that the production of propionic acid provides a key to potentially explain hyperinsulinaemia and subsequent insulin resistance. On the other hand, it cannot be excluded that a portion of the cassava starch escaped from being fermented in the rumen, thereby, rendering the glucose available for absorption in the small intestine (Li *et al*., 2012). This mode of action would, at least partly, also contribute to the observed increase in plasma glucose when the high cassava ration was fed.

Clearly, current data do not provide clues as to the underlying mechanism by which hyperinsulinaemia causes laminitis. De Laat *et al*. (2012), however, suggested that the formation of advanced glycation endproducts (AGEs), at least in equine, may play a role. The mechanism by which AGEs may play a role in the development of laminitis is not yet fully elucidated, but it is known that AGEs can bind to its receptor, thereby, releasing pro-inflammatory cytokines and overexpression of some proteins in the extra-cellular matrix such as collagen and laminin (Simm *et al*., 2004).

Conclusions: The feeding of a high cassava ration caused greater plasma insulin levels and lower insulin sensitivity. Taken the current observations on plasma glucose and insulin into account, it can be hypothesized that the feeding of the HC-TMR ultimately caused formation of AGEs in the connective tissues of the hoofs resulting in clinical signs of laminitis.

Acknowledgements: The study was financially supported by The Thailand Research Fund and Udon Thani Rajabhat University, Thailand.

Authors contribution: RP and SA operated the experiment and analyzed samples and the data. JTS and WHH interpreted the data, reviewed and edited the manuscript.

REFERENCES

AOAC, 1990. Official methods of analysis, 15th ed. Association of official analytical chemists, Arlington, Virginia.

Boosman R, Nemeth F and Gruys E, 1991. Bovine laminitis: clinical aspects, pathology and pathogenesis with reference to acute equine laminitis. Vet Q 13:163-71.

De Laat MA, McGowan CM, Sillence MN, *et al*., 2010. Equine laminitis: induced by 48 h hyperinsulinaemia in Standardbred horses. Equine Vet J 42:129-35.

De Laat MA, Kyaw-Tanner MT, Sillence MN, *et al*., 2012. Advanced glycation end products in horses with insulin-induced laminitis. Vet Immunol Immunopathol 145:395-401.

Gong JG, Lee WJ, Garnsworthy PC, *et al*., 2002. Effect of dietary induced increases in circulating insulin concentrations during the early postpartum period on reproductive function in dairy cows. Reproduction 123:419-27.

Greenough PR, Bergsten C, Brizzi A, *et al*., 2007. Bovine laminitis and lameness: a handson approach. 1st ed. Saunders Elsevier, Philadelphia, PA.

Holtenius K, Agenäs S, Delavaud C, *et al*., 2003. Effects of feeding intensity during the dry period. 2. Metabolic and hormonal responses. J Dairy Sci 86:883-91.

Hughes J, 2001. A system for assessing cow cleanliness. In Practice 23:517-24.

Kahn CR, 1978. Insulin resistance, insulin insensitivity, and insulin unresponsiveness: a necessary distinction. Metabolism 27:1893-902.

Kawashima C, Munakata M, Shimizu T, *et al*., 2016. Relationship between the degree of insulin resistance during late gestation and postpartum performance in dairy cows and factors that affect growth and metabolic status of their calves. J Vet Med Sci 78:739-45.

Kerestes MV, Faigl V, Kulcár M, *et al*., 2009. Periparturient insulin secretion and whole-body insulin responsiveness in dairy cows showing various forms of ketone pattern with or without puerperal metritis. Domest Anim Endocrinol 37:250-61.

Li S, Khafipour E, Krause DO, *et al*., 2012. Effects of subacute ruminal acidosis challenges on fermentation and endotoxins in the rumen and hindgut of dairy cows. J Dairy Sci 95:294-303.

Nocek JE, 1997. Bovine acidosis: implications on laminitis. J Dairy Sci 80:1005-28.

Oh YK, Eun JS, Lee SC, *et al*., 2015. Responses of blood glucose, insulin, glucagon and fatty acids to intraruminal infusion of propionate in Hanwoo. Asian-Aust J Anim Sci 28:200-6.

Opsomer G, Wensing T, Laevens H, *et al*., 1999. Insulin resistance: the link between metabolic disorders and cystic ovarian disease in high yielding dairy cows. Anim Reprod Sci 56:211-22.

Pilachai R, 2013. Feeding practices and potential risk factors for laminitis in dairy cows in Thailand. PhD Thesis, Faculty of Veterinary Medicine, Utrecht University, Utrecht, the Netherlands.

Pilachai R, Schonewille JT, Thamrongyoswittayakul C, *et al*., 2012. Starch source in high concentrate rations does not affect rumen pH, histamine and lipopolysaccharide concentrations in dairy cows. Livest Sci 150:135-42.

Plaizier JC, Krause DO, Gozho GN, *et al*., 2008. Subacute ruminal acidosis in dairy cows: the physiological causes, incidence and consequences. Vet J 176:21-31.

Radostits OM, Blood DC, Gay CC, *et al*., 2000. Veterinary medicine: A textbook of the diseases of cattle, sheep, pigs, goats and horses. 9th ed. WB Saunders Company Ltd. London.

Samuel M, Sagathewan S, Thomas J, *et al*., 1997. An HPLC method for estimation of volatile fatty acids of rumen fluid. Indian J Anim Sci 67:805-7.

Simm A, Casselmann C, Schubert A, *et al*., 2004. Age associated changes of AGE-receptor expression: RAGE upregulation is associated with human heart dysfunction. Exp Gerontol 39:407-13.

Tarlton JF, Holah DE, Evans KM, *et al*., 2002. Biomechanical and histopathological changes in the support structures of bovine hooves around the time of first calving. Vet J 163:196-204.

Treiber KH, Hess TM, Kronfeld DS, *et al*., 2005. Insulin resistance and compensation in laminitis-predisposed ponies characterized by the Minimal Model. Pferdeheilkunde 21:91-2.

Van Knegsel ATM, Van den Brand H, Dijkstra J, *et al*., 2007. Effect of glucogenic vs. lipogenic diets on energy balance, blood metabolites, and reproduction in primiparous and multiparous dairy cows in early lactation. J Dairy Sci 90:3397-409.

Van Soest PJ, Robertson JB and Lewis BA, 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. J Dairy Sci 74:3583-97.

Walsh DM, McGowan CM, McGowan TW, *et al*., 2009. Correlation of plasma insulin concentration and laminitis score in a field study of equine Cushing's disease and equine metabolic syndrome. J Equine Vet Sci 29:87-94.