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Effect of heat-moisture treated cassava *(Manihot utilissima)* supplementation on lactational performance and rumen fermentation of early lactating crossbred cows[#]

C.S. Gopu Krishnan[1], K. Lalu[2*], K. Ally[1], K.S. Ajith [1], Lasna Sahib[1] and V. Babitha[3]

[1]Department of Animal Nutrition, [3]Department of Veterinary Physiology College of Veterinary and Animal Sciences, Mannuthy, [2]College of Dairy Science and Technology, Kaimanam, Thiruvananthapuram, Kerala Veterinary and Animal Sciences University, Pookode, Wayanad, Kerala

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Abstract

A feeding trial of 90 days was carried out in early lactating crossbred cows to evaluate the effect of heat-moisture treated (HMT) cassava (Manihot utilissima) supplementation on lactational performance and rumen fermentation of the animals. Twenty early lactating crossbred cows in the Cattle Breeding Farm, Thumburmuzhy were randomly assigned to four treatment groups which were T1 (control with 20 % crude protein, 68 % total digestible nutrients), T2 (T1 with 250 g/ day HMT0 cassava), T3 (T1 with 250 g/day HMT2 cassava) and T3 (T1 with 250 g/ day HMT3 cassava), with five animals in each group. Green fodder and paddy straw were used as roughages (ICAR, 2013). The result revealed that average daily dry matter intake (DMI), average body weights, fortnightly average daily milk yield of the animals among different treatments remain statistically similar (p>0.05) throughout the study. It was also observed that the rumen fermentation parameters such as rumen pH, total volatile fatty acids and rumen ammonia nitrogen were non-significant (p>0.05) among the dietary treatments. The study revealed that supplementing HMT cassava at the level of 250 g/day did not have any significant impact on the lactational performance of early lactating cows.

Keywords: *Heat-moisture treatment, cassava, lactation, rumen fermentation*

Proper nutrition is critical for the health and productivity of dairy cattle, particularly high-yielding animals with elevated energy needs. Intensive management often leads to feeding lactating dairy cows with diets rich in easily digestible carbohydrates to enhance milk production (Khiaosa-Ard and Zebeli, 2012). However, excessive intake of rapidly fermentable starches can disrupt the rumen ecosystem, leading to sub-acute ruminal acidosis (SARA) and in more severe cases, acute ruminal acidosis (ARA) when ruminal pH falls below 5.8 and 5, respectively (Nagaraja and Titgemeyer, 2007). These conditions might lead to various health issues, including impaired digestion, laminitis and liver abscesses, which can compromise cattle welfare and dairy profitability (Plaizier *et al*., 2008; Karapinar *et al*., 2010).

While optimising feeding strategies for ruminants, it is essential to maintain favourable rumen metabolism. Given the limited fibre intake in high-producing cattle, strategies that slow the degradation of starch-rich feeds, in the rumen, could mitigate the risk of SARA and ARA (Zebeli *et al.,* 2008). Enhanced digestion of starch in the small intestine may

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**Corresponding author: lalu@kvasu.ac.in, Ph. 6282088162*

also boost milk protein production by sparing amino acids, which otherwise will be used for gluconeogenesis in the liver and it is also considered to be providing greater energy efficiency compared to rumen digestion of starch (Owens *et al*., 1986). The potential of various chemical and physical treatments, such as heat-moisture treatment, to increase resistant starch (RS) content in these feeds remains an area of active research (Deckardt *et al*., 2013).

Heat-moisture treatment (HMT) and subsequent cooling may be an effective way to improve the RS content in starch rich feeds (Dupuis *et al*., 2014). Cassava (*Manihot utilissima*) is one of the easily available starch sources in tropics, which can be used as a feed source for ruminants. Even though there are studies showing good production and reproduction performance of lactating animals, supplemented with bypass fat as energy source (Das *et al.,* 2020), those regarding the use of bypass/resistant starch in the diet of ruminants in India are very less. Considering the benefits of the feeding diet which are degraded seldom or really slowly in the rumen, this work was undertaken to assess its effect on production performance and rumen fermentation parameters of early lactating crossbred cows.

Materials and methods

Cassava (*Manihot utilissima*) was procured in dried form, locally and ground to flour in School of Animal Nutrition and Feed Technology (SANFT) Feed mill, Mannuthy. The cassava flour was subjected to heat-moisture treatment (HMT) cycle of 121°C with moisture level of 1:3.5 w/v. Three samples of cassava such as HMT I, HMT II and HMT III were prepared with one, two and three cycles of HMT respectively, and each sample were cooled at -20°C in deep freezer for six hours (modified from Putra *et al*., 2023). The resistant starch levels in the cassava samples were quantified (modified from Sopade and Gidely, 2009) and the samples having highest resistant starch levels were selected for the *in vivo* study.

Twenty early lactating crossbred cows were selected from the Cattle Breeding Farm, Thumburmuzhy, Kerala Veterinary and Animal Sciences University and distributed randomly into four treatment groups (T1, T2, T3 and T4) of five animals each. Throughout the 90-day trial period, the animals were fed a concentrate mixture (CP- 20 % and TDN- 68%), green fodder and paddy straw (ICAR, 2013). Additionally, 250 g of HMT0, 250 g of HMT II and 250 g HMT III were supplemented to T2, T3 and T4, respectively, per day. Ingredient composition of the four experimental rations fed to the animals are given in Table 1. All the animals were maintained under uniform managemental conditions in the farm.

The data on daily dry matter intake of the animals were recorded throughout the feeding trial. Body weight of the experimental animals was recorded, every fortnight. Rumen liquor samples were collected at the end of the trial and analysed for rumen pH, total volatile fatty acids (Barnett and Reid, 1957) and rumen ammonia nitrogen (Beecher and Whitten, 1970). The proximate composition of feed and fodder was analysed (AOAC, 2016). Analyses of variance was used for statistical analysis (Snedecor and Cochran, 1994) using statistical packages for social services (SPSS), version 24.

Table 1. Ingredient composition of the experimental concentrate mixture* fed to experimental animals

Ingredient	Percentage composition			
	T1	T2	T3	T4
Corn gluten fibre	25.0	25.0	25.0	25.0
Coconut cake	10.0	10.0	10.0	10.0
De-oiled rice bran	16.0	16.0	16.0	16.0
Black gram husk	8.0	8.0	8.0	8.0
Maize	16.0	16.0	16.0	16.0
Rice polish	8.0	8.0	8.0	8.0
Tapioca starch waste	1.0	1.0	1.0	1.0
Alfalfa	13.0	13.0	13.0	13.0
Calcite	1.5	1.5	1.5	1.5
Mineral mixture	0.5	0.5	0.5	0.5
Salt	1.0	1.0	1.0	1.0
Total	**100**	**100**	**100**	**100**
HMT 0 cassava		250.0 g		
HMT II cassava			250.0 g	
HMT III cassava				250.0 g

*To every 100 kg of the concentrate feed,100 g of Toxfia was added

Results and discussion

Chemical composition of feed

The per cent composition of the concentrate mixture, green grass and paddy straw fed to the experimental animals in four experimental rations are given in Table 2 and chemical composition of the cassava samples supplemented are given in Table 3.

Body weight

The average body weight of experimental lactating cows maintained on four treatments viz. T1, T2, T3 and T4 at fortnightly intervals are given in Table 4. The average initial and final body weights (kg) of the experimental animals were 385.40±21.95, 424.20±34.78, 447.20±15.75, 418.00±21.53 and 387.20±21.40, 428.00±33.68, 452.80±17.88, 422.60±19.17 respectively. The statistical analysis showed that the average bodyweight of the experimental animals was similar among the four treatment groups throughout the feeding trial. Similar results were reported by Ally *et al.* (2007). All the experimental groups showed a decline in body weight

Table 2. Chemical composition[1] of the concentrate feed, green fodder and paddy straw fed to the experimental animals

Parameters	Concentrate	Green fodder	Paddy straw
Dry matter	90.54±0.26	17.52±0.14	90.83±0.33
Crude protein	20.91±0.24	10.55±0.15	2.98±0.02
Ether extract	4.37±0.10	2.04±0.03	1.14±0.02
Crude fibre	8.82±0.08	30.90±0.12	30.81±0.58
Total ash	10.55±0.12	10.10±0.09	14.48±0.10
Nitrogen free extract (NFE)	53.89±0.32	46.41±0.18	50.60±0.61
Calcium	0.86±0.01	0.55±0.01	0.28±0.00
Phosphorus	0.53±0.01	0.24±0.01	0.09±0.00
Neutral detergent fibre (NDF)	32.74±0.15	61.84±0.09	68.83±0.25
Acid detergent fibre (ADF)	14.51±0.11	40.89±0.10	47.22±0.22

[1]Mean of six values with SE

Table 3 Chemical composition[1] of the cassava samples fed to the experimental animals

Parameters	Cassava samples		
	HMT 0	HMT II	HMT III
Dry matter	94.04±0.09	88.80±0.22	87.85±0.10
Crude protein	2.39±0.02	2.73±0.01	2.91±0.01
Ether extract	0.96±0.02	1.35±0.03	1.73±0.06
Crude fibre	4.40±0.06	5.25±0.05	5.83±0.07
Total ash	1.68±0.02	1.72±0.03	1.73±0.03
Nitrogen free extract	90.57±0.21	88.94±0.23	87.8±0.32
Calcium	0.46±0.03	0.44±0.01	0.40±0.00
Phosphorus	0.36±0.02	0.32±0.03	0.31±0.01
Neutral detergent fibre	5.61±0.08	8.90±0.08	9.83±0.06
Acid detergent fibre	2.73±0.05	2.95±0.13	3.33±0.10

[1]Mean of six values with SE

Table 4. Fortnightly average body weight[1] of the experimental animals maintained on the four experimental rations

Fortnight	Dietary treatments				p value
	T1	T2	T3	T4	
0	385.40±21.95	424.20±34.78	447.20±15.75	418.00±21.53	0.385^{ns}
1	372.00±22.00	414.80±33.12	441.60±18.66	404.60±20.62	0.277^{ns}
2	366.60±21.67	412.00±34.27	436.00±19.08	403.00±19.66	0.283^{ns}
3	371.60±21.04	417.60±34.46	441.40±19.79	417.20±18.84	0.270^{ns}
4	378.00±21.40	422.60±33.37	447.20±18.70	419.00±18.72	0.266^{ns}
5	381.80±20.71	423.80±33.86	451.00±18.23	420.20±19.27	0.273^{ns}
6	387.20±21.40	428.00±33.68	452.80±17.88	422.60±19.17	0.314^{ns}

[1]Mean of five values with SE; ns- non significant (p>0.05)

for the first two fortnights and then body weight gain was experienced throughout the feeding trial. Roche *et al.* (2015) reported that high yielding dairy cows exhibit a loss in body weight for the first month post-partum.

Daily dry matter intake

Fortnightly average daily dry matter intake of experimental animals fed on four dietary treatments are displayed Table 5. The average DMI in each group were 13.98±0.39, 14.38±0.34, 14.58±0.18 and 14.58±0.26 kg respectively for T1, T2, T3 and T4, respectively. Iqbal *et al*. (2009) evaluated the effect of barley grain steeped in 0.5 per cent lactic acid, as a rumen bypass starch source and found that there was no significant difference in the dry matter intake of animals fed barley grains which were steeped for 48 hours either in 0.5 per cent lactic acid (treatment group) or in an equal quantity of tap water (control). Similarly, Delahoy *et al.* (2003) noted that there was no difference in total dry matter intake (DMI) between cows that were fed cracked corn and those that received steam-flaked corn, as a supplement. Carmo *et al.* (2015) reported a linear increase in the DMI when fed with a diet

Table 5. Fortnightly average daily dry matter intake[1] of the experimental animals maintained on the four experimental rations, kg

Fortnight	Dietary treatments				p value
	T1	T2	T3	T4	
1	13.44±0.48	13.87±0.34	14.00±0.30	14.10±0.33	0.609 ns
2	14.05±0.39	14.28±0.40	14.65±0.18	14.48±0.26	0.586ns
3	14.09±0.35	14.32±0.37	14.67±0.15	14.67±0.15	0.392ns
4	14.17±0.34	14.49±0.38	14.76±0.17	14.73±0.35	0.555 ns
5	14.11±0.36	14.64±0.36	14.69±0.20	14.75±0.33	0.480 ns
6	14.01±0.42	14.70±0.30	14.71±0.10	14.78±0.32	0.292 ns
Mean±SE	13.98±0.39	14.38±0.34	14.58±0.18	14.58±0.26	0.466ns

[1]Mean of five values with SE; ns- non significant ($p>0.05$)

Table 6. Fortnightly average milk production[1] of the experimental animals maintained on the four experimental rations, kg

Fortnight	Dietary treatments				p value
	T1	T2	T3	T4	
1	14.57±1.54	15.66±0.67	16.55±0.82	17.08±1.14	0.418 ns
2	15.08±1.30	16.36±0.78	16.73±0.78	17.48±0.81	0.366 ns
3	15.08±1.22	16.41±0.68	16.67±0.54	17.38±0.70	0.364 ns
4	14.43±1.21	15.92±0.91	16.01±0.53	16.73±0.79	0.403 ns
5	14.02±1.24	15.34±1.23	15.64±0.49	16.55±0.67	0.356 ns
6	13.52±1.00	14.98±1.28	15.43±0.41	16.02±0.94	0.258 ns
Mean±SE	14.32±1.23	15.55±0.85	16.02±0.55	16.72±0.80	0.313ns

[1]Mean of five values with SE; ns- non significant ($p>0.05$)

containing different levels of starch at 15, 20, 25 or 30 per cent ($p<0.05$), contrary to the current study.

Milk production

The fortnightly average daily milk yield of the experimental animals in four treatment groups showed mean values of 14.32±1.23, 15.55±0.85, 16.02±0.55 and 16.72±0.80 kg for T1, T2, T3 and T4, respectively. The data is represented in Table 6 and the average daily milk yield was found to be statistically similar ($p>0.05$) among treatment groups. In agreement to this study, Iqbal *et al.* (2009) found that there was no significant difference in the milk production between groups fed barley grains which were steeped for 48 hours either in 0.5 per cent lactic acid (treatment group) or in an equal quantity of tap water (control). Similarly, Delahoy *et al.* (2003) noted that there was no significant difference in milk yield between cows that were fed cracked corn supplement compared to those that received steam-flaked corn supplement. Carmo *et al.* (2015) found that feeding lactating cows with diets containing starch at different levels of 15, 20, 25 or 30 per cent increased the milk yield linearly up to 25 per cent starch level and decreased at 30 per cent level.

Rumen fermentation parameters

The results of rumen fermentation parameters such as, rumen pH, total volatile fatty acids and rumen ammonia nitrogen are listed in Table 7.

Rumen pH

The average rumen pH values obtained for animals maintained on four experimental rations were 7.10±0.05, 6.94±0.05, 7.06±0.05 and 7.08±0.06 respectively for T1, T2, T3 and T4. The results were similar among all the treatment groups ($p>0.05$). Dominic (2013) also reported comparable values for rumen pH in his study of the effect of post-partum energy supplementation on rumen fermentation patterns of early lactating cows. Corresponding to this study, Reis and Combs (2000) found that the ruminal pH of animals supplemented with a concentrate mixture containing 90 per cent dry shell corn with CP per cent at two different levels of five or ten kg/day, was similar to that of the control group. In contrast to the findings of the current study, Khampa and Wanapat (2006) reported that two per cent level of supplementation of cassava chips lowered ruminal pH in lactating animals compared to one per cent level of supplementation. Iqbal *et al.* (2012) reported an increase in rumen pH of animals fed TMR ration containing barley grains steeped in lactic acid (10 g/L) for 48 hours and then heated at 55^{0}C compared to those fed with TMR having barley grains steeped in equal amount of tap water.

Table 7. Rumen fermentation parameters[1] of the experimental animals maintained on the four experimental rations

Parameter	Dietary treatments				p- value
	T1	T2	T3	T4	
Rumen pH	7.10±0.05	6.94±0.05	7.06±0.05	7.08±0.06	0.726 [ns]
TVFA (mM/L)	79.90±1.00	84.08±3.41	82.03±1.67	80.20±1.11	0.464 [ns]
Rumen ammonia nitrogen (mg/dL)	14.40±0.69	16.04±0.94	14.85±0.71	14.87±0.69	0.491 [ns]

[1]Mean of five values with SE; ns- non significant (p>0.05)

Total volatile fatty acids

The mean total volatile fatty acids values obtained for experimental animals maintained on four dietary treatments were 79.90±1.00, 84.08±3.41, 82.03±1.67 and 80.20±1.11 mM/L for T1, T2, T3 and T4 respectively. There was no significant among the four treatment groups fed with four different treatment rations (p>0.05) and the data obtained were in the normal range. Similarly, Reis *et al.* (2001) reported that there was no significant difference in TVFA production between animals supplemented with finely ground dry shelled corn or coarsely ground high moisture corn compared to control group. Gulmez and Turkemen (2007) found that there was no significant difference in TVFA production among groups fed with diets containing maize and wheat in different proportions. Unlike the findings of this study Khampa and Wanapat (2006) reported that two per cent level of supplementation of cassava chips and corn meal, increased TVFA in animals compared to one per cent level of supplementation.

Rumen ammonia nitrogen

The average values of rumen ammonia nitrogen obtained for experimental lactating cows maintained on four experimental rations, T1, T2, T3 and T4 weref14.40±0.69, 16.04±0.94, 14.85±0.71 and 14.87±0.69 mg/dL respectively. There was no significant among the four treatment groups fed with four different treatment rations (p>0.05). In a manner consistent with the current study, Rearte *et al.* (2001) reported that there was no significant difference in the rumen ammonia level in cows supplemented with high moisture corn when compared to the control. Conversely, Mc Cormick *et al.* (2001) reported that supplementation of sucrose in a corn-based ration at the level of five per cent lowered the rumen ammonia nitrogen concentration in dairy cows when compared to that of lactose supplementation.

Conclusion

On critical understanding of the data of milk production, it can be concluded that supplementation of HMT cassava at 250 g/day did not have any significant impact on the milk production of the lactating animals. So further studies with a higher level of supplementation of HMT cassava is needed before finalising its effect in the animal feed.

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Conflicts of interest

The authors declare that they have no conflict of interest.

References

Ally, K., Mercy, A.D. and Viswanathan, T.V. 2007. Effect of dietary crude protein levels on the performance of lactating cows. *Indian J. Anim. Nutr.* **24**: 84-87.

AOAC [Association of Official Analytical Chemists]. 2016. Official Methods of Analysis. (20th Ed.). Association of official analytical chemists, Washington DC, 684p.

Barnett, A.J.G. and Reid, R.L. 1957. Studies on the production of volatile fatty acids from grass by rumen liquor in an artificial rumen. 1. The volatile fatty acid production from fresh grass. *J. Agri. Sci. Camb*. **48**: 315-321.

Beecher, G.R. and Whitten, B.K. 1970. Ammonia determination: reagent modification and interfering compounds. *Anal. Biochem*. **36**: 243-246.

Carmo, C.A., Batistel, F., De Souza, J., Martinez, J.C., Correa, P., Pedroso, A.M. and Santos, F.A.P. 2015. Starch levels on performance, milk composition and energy balance of lactating dairy cows. *Trop. Anim. Health Prod.* **47**: 179-184.

Das, A.S., Joseph, M., Kurien, M.O., Simon, S., Usha, A.P., Shyama, K., Aslam, M.K.M. and Niyas, E. 2020. Postpartum reproductive efficiency in cows supplemented with rumen bypass fat. *J. Vet. Anim. Sci.* **51**: 8-13.

Deckardt, K., Khol-Parisini, A. and Zebeli, Q. 2013. Peculiarities of Enhancing Resistant Starch in Ruminants Using Chemical Methods: Opportunities and Challenges. *Nutrients*. **5**: 1970-1988.

Delahoy, J.E., Muller, L.D., Bargo, F., Cassidy, T.W. and Holden, L.A. 2003. Supplemental Carbohydrate Sources for Lactating Dairy Cows on Pasture. *J. Dairy Sci.* **86**: 906-915.

Dominic, G. 2013. Effect of high energy diet on early lactation in cross-bred cows. *M.V.Sc. thesis.* Kerala Veterinary and Animal Sciences University, Pookode, 95p.

Dupuis, J.H., Liu, Q. and Yada, R.Y. 2014. Methodologies for increasing the resistant starch content of food starches: A review. *Compr. Rev. Food Sci. Food Saf.* **13**: 1219-1234.

Gulmez, B.H. and Turkmen, I.I. 2007. Effect of starch sources with different degradation rates on ruminal fermentation of lactating dairy cows. *Rev. Med. Vet.* **158**: 92-99.

ICAR [Indian Council of Agriculture and Research]. 2013. *Nutrient Requirements of Animals – Cattle and Buffalo* (3rd Ed.). Indian Council of Agricultural Research, New Delhi, 24p.

Iqbal, S., Terrill, S.J., Zebeli, Q., Mazzolari, A., Dunn, S.M., Yang, W.Z. and Ametaj, B.N. 2012. Treating barley grain with lactic acid and heat prevented sub-acute ruminal acidosis and increased milk fat content in dairy cows. *Anim. Feed Sci. Technol.* **172**: 141-149.

Iqbal, S., Zebeli, Q., Mazzolari, A., Bertoni, G., Dunn, S.M., Yang, W.Z. and Ametaj, B.N. 2009. Feeding barley grain steeped in lactic acid modulates rumen fermentation patterns and increases milk fat content in dairy cows. *J. Dairy Sci.* **92**: 6023-6032.

Karapinar, T., Dabak, M. and Kizil, O. 2010. Thiamine status of feedlot cattle fed a high-concentrate diet. *The Can. Vet. J.* **51**: 12-51.

Khampa, S. and Wanapat, M. 2006. Influences of energy sources and levels supplementation on ruminal fermentation and microbial protein synthesis in dairy steers. *Pakist. J. Nutr.* **5**: 294-300.

Khiaosa-Ard, R. and Zebeli, Q. 2012. Dietary modulation of rumen metabolism: a key factor to enhancing ruminant production. *Albanian J. Agric. Sci.* **11**: 131-140.

Mc Cormick, M. E., Redfearn, D. D., Ward, J. D. and Blouin, D. C. 2001. Effect of protein source and carbohydrate addition on rumen fermentation and lactation performance of Holstein cows. *J. Dairy Sci*. **84**: 1686-1697.

Nagaraja, T.G. and Titgemeyer, E.C. 2007. Ruminal Acidosis in Beef Cattle: The Current Microbiological and Nutritional Outlook. *J. Dairy Sci.* **90**: E17-E38.

Owens, F.N., Zinn, R.A. and Kim, Y.K. 1986. Limits to starch digestion in the ruminant small intestine. *J. Anim. Sci.* **63**: 1634-1648.

Plaizier, J.C., Krause, D.O., Gozho, G.N. and McBride, B.W. 2008. Subacute ruminal acidosis in dairy cows: The physiological causes, incidence and consequences. *Vet. J.* **176**: 21-31.

Putra, L.O., Suharti, S., Sarwono, K.A., Sutikno, S., Fitri, A., Astuti, W.D., Rohmatussolihat, R., Widyastuti, Y., Ridwan, R., Fidriyanto, R. and Wiryawanv, K.G. 2023. The effects of heat-moisture treatment on resistant starch levels in cassava and on fermentation, methanogenesis, and microbial populations in ruminants. *Vet. World*. **16**: 811–819.

Rearte, D.H., Alvarez, H. and Santini, F. 2001. High moisture corn supplementation of different dry matter content to cows grazing temperature pasture. *J. Dairy Sci*. **29:** 424-441.

Reis, R.B. and Combs, D.K. 2000. Effects of increasing levels of grain supplementation on rumen environment and lactation performance of dairy cows grazing grass- legume pasture. *J. Dairy Sci*. **83**: 2888-2898.

Reis, R.B., San Emeterio, F., Combs, D.K., Satter, L.D. and Costa, H.N. 2001. Effects of corn particle size and source on performance of lactating cows fed direct-cut grass-legume forage. *J. Dairy Sci*. **84**: 429–441.

Roche, J.R., Meier, S., Heiser, A., Mitchell, M.D., Walker, C.G., Crookenden, M.A., Riboni, M.V., Loor, J.J. and Kay, J.K. 2015. Effects of precalving body condition score and prepartum feeding level on production, reproduction, and health parameters in pasture-based transition dairy cows. *J. Dairy Sci*. **98**: 7164–7182.

Snedecor, G.W. and Cochran, W.G. 1994. *Statistical Methods.* (8th Ed.). The Iowa State University Press, Ames, USA, 503p.

Sopade, P.A. and Gidley, M.J. 2009. A rapid *in-vitro* digestibility assay based on glucometry for investigating kinetics of starch disnedgestion. *Starch.* **61**: 245-255.

Zebeli, Q., Dijkstra, J., Tafaj, M., Steingass, H., Ametaj, B.N. and Drochner, W. 2008. Modelling the adequacy of dietary fibre in dairy cows based on the responses of ruminal pH and milk fat production to composition of the diet. *J. Dairy Sci.* **91**: 2046-2066. ■