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Effect of feeding glycerol on rumen fluid and the parameters of the energetic profile in dairy cows

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Summary

The aim of the authors' study was to determine the effects of various concentrations of a glycerol-containing preparation on the dynamics of changes in individual parameters of rumen fluid and energetic profile in dairy cows. The presented results suggest a potential positive effect of glycerol as an energetic supplement to dairy cows. A number of studies have showed that the positive effect of glycerol is influenced by the administered doses, time of feeding and the form of administered glycerol. In this experiment a significant positive effect of glycerol was observed only in the third experimental group that was administered the highest dose of the investigated preparation and was reflected in pH ($P < 0.05$), acetic acid ($P < 0.05$), butyric acid ($P < 0.0$) and BHB ($P < 0.01$). The recorded moderate antilipolytic and glucoplastic effects of glycerol and the buffering capacity of the powder vehicle used (zeolite) on the health of dairy cows needs to be further verified in a long--term experiment with the highest dose of glycerol.

Keywords: feeding glycerol, dairy cows, transition period in dairy cows

Glycerol as a glucoplastic substance is used to prevent nutritional deficiencies in dairy cows predominantly during the transition period which encompasses the last 3 weeks prepartum and first 3 weeks postpartum (4). Dairy cows are at an increased risk of a variety of health disorders such as metabolic and infectious diseases (4). The prepartum depression of food intake and slow increase of appetite result in a negative energetic balance of dairy cows. During the transition period, the immune system is suppressed, rendering the cow susceptible to infectious diseases such as mastitis and endometritis (9). This period is characterized by decreased concentration of glucose in blood and mobilization of body fat reserves that are reduced to non-esterified fatty acid (NEFA) and glycerol. There are also changes in the level of acetic acid as a precursor for fat synthesis, propionic acid as a glucogenic precursor and in the acetate/propionate ratio in the rumen fluid of dairy cows (10). Excessive mobilisation of NEFA results in accumulation of triglycerides in the liver causing health disorders.

Glycerol as a glucoplastic substance is involved in the metabolism of glucose. This natural product can be found in all vegetable and animal fats. Its incorporation in the glycide metabolism is more expeditious than that of propylene glycol and other chemical glucoplastic substances (1).

Glycerol consumed in the feed is absorbed largely in the rumen and is metabolised to propionic acid by infusoria. Glycerol decreases the acetate to a propionate ratio and stimulates the water intake which will be of benefit to dairy cows in the transition period (11). Moreover, glycerol decreases the concentration of NEFA and increases blood glucose level (3). Feeding glycerol has been used to prevent ketosis, lipomobilisation syndrome and other disorders occurring during the transition period.

The aim of our study was to determine the effects of various concentrations of the glycerol-containing preparation Ketoglyc Powder on changes in some parameters in rumen fluid (volatile fatty acids – VFA, acidity – pH) and in blood serum (glucose, total cholesterol – TCH, total lipids – TL, triglycerides – TG, NEFA, and betahydroxybutyrate – BHB) and to determine optimum doses of glycerol. We expected a positive effect of glycerol as a glucoplastic substance on pH, decrease in acetate to propionate ratio, NEFA and BHB and increase in serum glucose.

Material and methods

Dairy cows of a lowland black spotted breed were used in the experiment. The experiment was carried out at the 2nd Internal Clinic of the University of Veterinary Medicine in Košice during 4 days. The first day of the experiment was considered a control (C) and each of the following days represented one experimental group (E1, E2, E3). Every group included 6 dairy cows. All procedures involving the use of animals were approved by the Ethics Committee for Handling of Animals at the University of Veterinary Medicine, Košice, the Slovak Republic.

Specified quantities of the glycerol-containing preparation (Ketoglyc Powder), dissolved in a constant volume of water, were administered orally in a single dose to animals in experimental groups through a nasopharyngeal applicator tube (tab. 1 and 2).

Tab. 1. Chemical composition of Ketoglyc Powder, percentage abundance of components (weight %)

Components	Percentage abundance
Zeolite	38.8 w.%
Glycerol	34.0 w.%
SiO_2	16.7 w.%
Water	9.6 w.%
Sodium chloride	3-9 w.%
Choline chloride	4.5×10^{-3} w.%
DL-methionine	0.9×10^{-3} w.%
$ZnCl_2$	0.8×10^{-3} w.%
E vitamin (α-tocopherol 100%)	8.9×10^{-5} w.%
β-carotene	3.8×10^{-5} w.%
$CoCl_2 \times 6\ H_2O$	1.8×10^{-5} w.%
B_3 vitamin (niacin 100%)	1.6×10^{-5} w.%
KI	0.6×10^{-5} w.%
$Na_2SeO_3 \times 5\ H_2O$	0.4×10^{-5} w.%

Tab. 2. Scheme of groups, number of used animals, applied oral doses of preparation in g/animal/day, and the volume of glycerol in selected individual doses (g)

Group	The number of animals	Oral application of preparation (g/animal/day)	The volume of glycerol (g)
Control	6	–	–
Experiment 1	6	150	50.0
Experiment 2	6	200	66.6
Experiment 3	6	300	100.0

Samples of blood and rumen fluid were taken during the day at specified time intervals (rumen fluid after 0, 3, 4, 6 and 10 hours, blood after 0, 4, 8 and 10 hours) after administration of glycerol. The initial samples (hour 0) were collected before administration of the preparation.

Rumen fluid acidity (pH) was determined by a pH meter Orion 720 A, volatile fatty acids (VFA) by isotachophoresis employing an Isotachophoretic analyser ZKI 02 (Laberko, Spišská Nová Ves). The remaining parameters (glucose, total cholesterol, total lipids, triglycerides, NEFA and betahydroxybutyrate) were analysed by an Automatic Spectrophotometric Analyser (Alize, Lisabio) using Randox kits (UK).

The results obtained were evaluated statistically applying One-Way Anova and Tukey's Multiple Comparison Tests within the groups for respective days.

Results and discussion

The day's dynamics of selected parameters was determined in the control and experimental groups after application of glycerol.

On the basis of results from the control group a graphic model of changes in the rumen fluid was developed. An effect of glycerol feeding was noted in the third experimental group ($P < 0.05$, $P < 0.01$). The day's dynamics of all selected parameters in the first and second experimental groups was identical with that in the control group.

Tab. 3. Selected parameters of rumen fluid in the 3rd experimental group (x ± sd)

	0 hour	3rd hour	4th hour	6th hour	10th hour
pH	6.88	6.83	7.04^a	6.95	6.75^b
	0.063	0.110	0.137	0.190	0.123
VFA	73.89	71.51	68.74	65.28	72.03
	11.872	11.556	6.232	8.267	13.437
Acetic acid	75.73^a	69.26^a	76.08^b	75.96^a	79.83^b
	3.672	6.093	2.067	3.818	11.209
Propionic acid	12.46	13.48	11.09	10.93	13.29
	2.514	3.822	1.398	2.207	3.680
Lactic acid	1.42	1.11	1.32	1.28	1.18
	0.147	0.258	0.233	0.288	0.464
Butyric acid	11.77^A	17.23^B	12.81^A	12.94^A	8.86^B
	1.987	2.265	1.030	2.164	5.320
Ratio C2 : C3	6.30	5.51	6.96	7.23	6.76
	1.377	1.694	1.050	1.761	3.421

Explanations: means with different superscript letters differ significantly – a, b at $p < 0.05$; A, B at $p < 0.01$

In the third experimental group the following changes were recorded (tab. 3): pH showed a decrease after 3 and 10 hours and an increase after 4 hours from administration of glycerol ($P < 0.05$). VFA showed no significant decrease during the experiment, acetic acid (C2) decreased significantly after 3 hours and then increased gradually up to the 10th hour ($P < 0.05$), propionic acid (C3) showed a slight increase after 3 and 10 hours, butyric acid (C4) increased significantly after 3 hours ($P < 0.01$) and then showed a gradual decrease, while the control group showed an increase in these parameters throughout the experiment and a decrease in C2 : C3 ratio after 3 and 10 hours. The day's dynamics of lactic acid was the same in the control and experimental groups.

Similarly to the changes in the rumen fluid, the changes in parameters of energetic profile – glucose, NEFA, betahydroxybutyrate (BHB), total lipids (TL), triglycerides (TG), total cholesterol (TCH) – were practically the same in the control animals and in the cows from the first and second experimental groups.

In the third experimental group the following tendencies were observed (tab. 4): the glycaemic line showed a relatively uniform slow decrease compared to progressive hyperglycaemia in the control group; concentration of NEFA decreased in comparison with the continuous increase in the control group; the authors observed a contradictory dynamic in the concentration of TL and BHB in individual time intervals ($P < 0.01$); changes in TG and TCH showed an identical tendency in all observed groups.

The oral administration of preparations intended for treatment and prevention of ketosis had a positive effect on the status of rumen, particularly on pH, VFA and relative ratio of rumen acids (8). Our results of rumen fluid analyses after administration of Ketoglyc Powder showed a significant effect on rumen fermentation, particularly in the third experimental group. The changes recorded in the

Tab. 4. Selected parameters of blood serum in the 3rd experimental group (x ± sd)

	0 hour	4th hour	8th hour	10th hour
Glucose (mmol/l)	4.55	4.39	4.43	4.35
	0.212	0.126	0.316	0.156
TCH (mmol/l)	2.86	2.85	2.91	2.88
	0.593	0.427	0.433	0.503
TL (g/l)	3.38	3.25	3.58	3.39
	0.196	0.525	1.128	1.146
TG (mmol/l)	0.29	0.29	0.27	0.26
	0.066	0.049	0.067	0.069
NEFA (mmol/l)	0.14	0.14	0.15	0.12
	0.036	0.034	0.043	0.016
BHB (mmol/l)	0.50[a]	0.45[a]	0.44[a]	0.49[b]
	0.122	0.059	0.055	0.093

Explanations: means with different superscript letters differ significantly – a, b at $p < 0.05$

first and second experimental groups were similar to those in the control group.

The rumen acidity (pH) in the third experimental group (300 g of Ketoglyc Powder/d) decreased after 3 hours and then increased rapidly by the 4th hour and subsequently showed a gradual decrease up to the 10th hour after administration of the preparation. In the control and two remaining experimental groups the pH values continued to increase throughout the experiment. The significant difference in the third experimental group could be induced by energetic supplementation of glycerol (decrease) and, on the other hand, by buffering capacity of the powder vehicle (increase). The third experimental group differed also in the level of VFA in comparison with the control group. Concentration of acetic acid in this group decreased within 3 hours and that of propionic acid increased within 10 hours after application of glycerol. The tendencies mentioned also induced changes in the acetate to propionate ratio. The level of butyric acid in this group increased after 3 hours while the level of lactic acid recorded at the same time was the lowest in comparison with the remaining groups.

A positive effect on rumen fluid acidity (pH), decrease in concentration of acetic acid, increase in the level of propionic acid and decrease in the acetic to propionic acid ratio (10) was reported after the application of glycerol in a liquid form (21 days a. p. and 100 days p. p.). A similar tendency of changes in the rumen status was also described by other authors (3) but there were scientists who did not observe such an effect of glycerol (4). The difference in the results can be related to both different doses of administered glycerol and different lengths of administration (4, 7). Glycerol administered per os is absorbed in the rumen, metabolised eventually to propionic acid by infusoria and utilised for gluconeogenesis (12).

The changes in the rumen status recorded after a single administration to cows in experimental groups indicated that dosage, time of application and the time of the adaptation can influence changes in the selected parameters. Significant changes were observed particularly in the experimental group administered the highest dose of glycerol.

The antilipolytic effect of glycerol as a glucoplastic substance was manifested by changes in concentrations of the serum NEFA and ketone (acetoacetate, betahydroxybutyrate, acetone), while changes in several other parameters of the energetic status were insignificant (2, 3). A similar tendency was observed in the authors' experiment based on the administration of single doses of Ketoglyc Powder. The changes in the energetic profile resembled those determined in the rumen. Significant changes were observed particularly in the third experimental group.

The glycaemic line showed a relatively uniform gradual decrease in comparison with progressive hyperglycaemia in the control group. The levels of NEFA in the third experimental group decreased while in the remaining groups they showed an increase indicative of higher lipomobilisation. The highest doses of glycerol caused a significant decrease in the concentration of BHB. No significant changes were observed in the other energetic profile parameters (TCH, TL, TG).

A number of authors reported that supplementation of glycerol to dairy cows induced similar effects on the rumen status, energetic profile, milk yield and the quality of milk (3).

Interesting results obtained by feeding additives to dairy cows were described by Hutjens (6). The glucoplastic substances increased milk yield, quality of produced milk, dry matter intake, microbial synthesis of proteins and VFA in the rumen, digestibility in the digestive tract, stabilized the rumen status and rumen fluid acidity (pH) in rumen fluid, decreased weight loss, reduced the negative effect of stress factors, promoted health and decreased nutritional deficiencies in dairy cows (ketosis, syndrome of fatty liver, acidosis, lipomobilisation syndrome).

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