Effects of Isopropyl Alcohol Infusions on the Ruminating Behavior of Goats

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ABSTRACT : Metabolites, such as isopropyl alcohol (IPA) produced by rumen fermentation, were intravenously infused into a jugular vein of goats during feeding to explore the mechanism and roles of IPA in ruminating behavior (number of boli and ruminating time). Three female goats were confined in metabolism cages with a stanchion. The ruminating behavior measured by the number of ruminations, ruminating time, number of remastications, and remasticating time decreased ($p<0.05$) with intravenous IPA infusion. The IPA concentrations and VFA concentrations increased in the blood circulation. Our data suggest that sensitive receptors of rumination to IPA are more likely to be in an area such as the brain stem where they can respond to blood metabolite levels. *(Asian-Aust. J. Anim. Sci. 2001. Vol 14, No. 8 : 1085-1089)*

Key Words : IPA, Acetone, Rumination, Intravenous Infusion, Goats

INTRODUCTION

The role of rumination in ruminant animals is to reduce the particle size of fibrous material of feed and thus to allow the feed to pass from the rumen (Kennedy, 1985). The ruminating time is influenced by diet (Pearce, 1965). The masticating behavior influenced by room lighting is important to determine the ruminating behavior (Oshiro et al., 1996), and lighting is also important to determine the rumination circadian rhythm during 24 h (Oshiro, 1985; Oshiro and Katayama, 1987; Oshiro et al., 1996).

Ruminants adjust voluntary food intake in relation to the physiological demand for energy (Baile, 1971). The effect of intraruminal infusion of acetic, propionic and butyric acid on the masticating time of sheep was reported by Ulyatt (1965) and of cattle by Simkins et al. (1967). Intravenous volatile fatty acids (VFA) infusions have no effect on the food intake of sheep (Forbes, 1986). We showed previously that the number of ruminations in goats increases during two days of fasting (Oshiro et al., 1992), but the ruminating time (remasticating time per bolus) decreases from the second day after fasting starts (Oshiro et al., 1992). VFA production in the rumen decreases after fasting stars (Pothoven and Beitz, 1975). Feeding increases the VFA in the rumen and the blood (Pothoven and Beitz, 1975), and reduces the ruminating behavior immediately after feeding (Oshiro, 1985; Oshiro and Katayama, 1987). The ruminating behavior of goats (Oshiro et al., 1988), sheep (Murphy et al., 1983) and cattle (Suzuki et al., 1979) are influenced by diet. Diet influences rumen VFA and blood ketones and acetone in cows (Montgomery and Baumgardt, 1965). The ruminating behavior of goats increases, and the masticating behavior decreases with intravenous acetone infusion (Oshiro et al., 2000). But acetone is not the end product. Studies on cows showed that acetone is further metabolized (Luick et al., 1967). Increases in concentration of acetone and IPA in the blood, rumen liquor, milk and urine are associated with ketosis in cows that intensify symptoms from subclinical to severe (Thin and Robertson, 1953). IPA is derived from acetone that circulates form the blood stream into the rumen, where acetone is converted to IPA (Thin et al., 1959).

This study examined the relation between IPA and the ruminating behavior of goats fed under a continuous light environment to find out the effects of intravenous infusion of IPA on the ruminating behavior of goats.

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MATERIALS AND METHODS

Three goats (Saanen×Native Okinawan crossbred female goats, body weight: Means±S.D. 34.6±7.6 kg) were offered alfalfa hay cubes (2.3×2.3×1.9 cm) with auto feeder by free choice (Oshiro and Katayama, 1987) and water at 12:00 h every day. Each goat was fitted with a jugular vein cannula and was locked in a stanchion of a raised (1 m) metabolism cage in an experimental room where the temperature and the relative humidity were maintained at 21.3±1.1°C and 63.1±3.2%, respectively. The test room (10.4 m^2) was continuously lit by fluorescent electric bulbs (4 bulbs×40 W, 59.4±10.3 Lux). Goats had become accustomed for four weeks to the experimental room and handling. The goats were attached to chewing sensors to measure the jaw movements. A polyethylene catheter was inserted into a jugular vein toward the heart. Metabolites were slowly infused into each goat using a peristaltic pump (micro tube pump: MP-3, Tokyo Rikakikai CO. LTD.). Data were collected for 24 h from 12:00 h (noon) on day 1 until 12:00 h (noon) on day 2.

We made three experiments: a non infusion experiment

without intravenous infusions, an experiment using Ringer's solution infused intravenously, and an experiment using Ringer's solution containing IPA infused intravenously. We used an infusion concentration of 0.0001527 mmoles/0.01 ml/kg body weight/min, which ranges in the blood within the subclinical level (Thin and Robertson, 1953). This IPA infusion amount was about one-seventh of an intravenous single infusion of sheep (Thin et al., 1959). All intravenous infusions started at 00:00 h (midnight) and continued until 12:00 h (noon) of the next day (day 2). The pH of the solution was adjusted to 7.0 using 0.1 N NaOH solution. The infused volumes in all infusion experiments were at a rate of 0.01 ml/kg body weight/min using the peristaltic pump. Blood and rumen were taken 12:00, 16:00, 20:00, 00:00, 01:00, 03:00, 06:00, 09:00 and 12:00 h after infusion period. The blood plasma was analyzed for VFA using a liquid chromatograph (model LC-10AS, Shimazu Corporation, Kyoto Japan), for IPA and acetone using a gas chromatograph (model GC-6A, Shimazu Corporation, Kyoto Japan), for non-esterified fatty acids (NEFA) using the Duncombe method (Duncombe, 1964), and for glucose using the glucose oxidase method (Trinder, 1969). The rumen juice was analyzed for VFA, IPA and acetone using an gas chromatograph (model GC-6A, Shimazu Corporation, Kyoto Japan).

The goat's jaw movement was recorded continuously for 24 h from 12:00 h of day 1 through to 12:00 h of day 2 using an auto-counter system (LB-8801, TECMO CO. LTD.) (Oshiro et al., 1987). The goats were allowed a period to adapt to the system prior to the experiment.

The results were reported as ruminations (number of boli; boli/12 h and ruminating time; min/12 h), mastcations (number of mastications; chews/12 h and masticating time; min/12 h) and resting time (min/12 h). The ruminations were further reported as number of remastications (chews/12 h), remasticating time (min/12 h), intermittent time (min/12 h), bolus formation time (sec/bolus), number of remastications per bolus (chews/bolus), remasticating time (sec/bolus) and intermittent time per bolus (sec/bolus) in each rumination; the averages of all these components in each 12 h of rumination and within each day were calculated.

Statistical analyses of all data were made between data collected before and after infusions using Tukey's method (Snedecor and Cochran, 1989).

RESULTS

Table 1 shows the food intake, ruminating behavior, masticating behavior and ruminating behavior per bolus with non infusion and with Ringer's solution and intravenous IPA infusion.

The ruminating behavior (number of boli, ruminating time, number of remastications and remasticating time) was constant at unvarying conditions during the non infusion and Ringer's solution infusion experiments.

The number of boli, ruminating time, number of remastications, and remasticating time were significantly different between the periods before and after infusion in intravenous IPA infusion experiments ($p<0.05$).

The masticating behavior (number of mastications and masticating time) was constant during the non infusion and Ringer's solution infusion experiments. Masticating behavior tended to decrease during intravenous infusion with IPA compared to before the infusions.

Food intake also tended to decrease during IPA infusion accompanied by decreased masticating behavior.

The bolus time, remasticating time per bolus, intermittent time per bolus and number of remastications per bolus were constant during the infusion periods in the three experiments.

Table 2 shows the concentrations of glucose, NEFA, VFA (acetic acid, propionic acid, butyric acid and isovaleric acid), acetone and IPA in the plasma in the three experiments.

The concentrations of glucose, NEFA, acetic acid, and acetone in the plasma did not change significantly in the three experiments, but these components tended to increase after intravenous IPA infusion. However, concentrations of propionic acid, butyric acid, isovaleric acid and IPA in the plasma increased significantly after the IPA infusion experiments ($p<0.05$).

Table 3 shows the concentrations of VFA, acetone and IPA in the rumen in the three experiments.

The concentrations of VFA, acetone, and IPA in the rumen were the same levels for both before and after infusion in the three experiments, but the VFA concentration in the rumen tended to increase after infusion in the IPA infusion experiment.

DISCUSSION

Feeding increases mastication and decreases rumination (Oshiro, 1985; Oshiro and Katayama, 1987). The ruminating behavior during and after feeding in continuous light either stays the same or decreases (Oshiro et al., 1988). Therefore, we postulate that a decrease in the ruminating behavior after feeding (Oshiro, 1985) is related to the increase in acetic acid in the rumen and plasma after feeding (Pothoven and Beitz, 1975). But fatty acids do not pass through the blood brain barrier to the ruminating brain center (Voet and Voet, 1995).

In this study, an IPA infusion into a jugular vein decreased the ruminating behavior, suggesting that the ruminating behavior decreases during and after IPA infusion because of the increase in blood IPA during and after IPA infusion.

Table 1. Food intake, ruminating behavior, masticating behavior and ruminating behavior per bolus in three experiments

Items		Controls		Treatment
		Non-infusion	Ringer-infusion	IPA-infusion
Food intake (g/12 hours)	P-I	680±147	610±220	663±310
	D-I	677±212	583±222	503±297
Number of boli	P-I	215±37	200±56	216±21
(boli/12 hours)	D-I	213±14	202±38	173±9*
Ruminating time	P-I	192±15	189±42	191±36
(min/12 hours)	D-I	197±25	218±32	141±10*
Number of remastications	P-I	13584±878	12863±3426	16641±4742
(chews/ 12 hours)	D-I	13163±256	15221±4035	11501±2506*
Remasticating time	P-I	164±9	165±36	168±34
(min/12 hours)	D-I	165±22	192±27	122±10*
Number of mastications	P-I	3601±578	4450±420	5078±1813
(chews/12 hours)	D-I	3787±982	4503±179	3942±1956
Masticating time	P-I	35±5	37±5	38±6
(min/12 hours)	D-I	36±8	37±1	27±21
Intermittent time	P-I	16±6	24±9	22±6
(min/12 hours)	D-I	19±7	26±7	19±3
Resting time	P-I	493±18	500±45	492±35
(min/12 hours)	D-I	487±73	448±62	553±16*
Bolus time (sec/bolus)	P-I	53.6±6.2	56.8±4.0	52.7±8.0
	D-I	55.5±63	58.4±5.5	48.9±4.1
Remasticating time per	P-I	45.8±67	49.4±4.8	46.6±8.4
bolus (sec/bolus)	D-I	46.5±6.3	51.4±6.0	42.3±3.4
Intermittent time per bolus	P-I	7.8±0.6	7.3±0.8	6.2±1.2
(sec/bolus)	D-I	9.0±0.6	7.1±0.7	6.5±1.3
Number of remastications	P-I	63.2±6.6	64.4±4.2	77.0±21.0
per bolus (chews/bolus)	D-I	61.8±6.3	68.0±7.8	66.0±11.4

P-I: pre-infusion, D-I: during-infusion.

* Means±S.D. in the same row having the superscripts are significantly different ($p<0.05$).

The results of this study may indicate an effect of IPA infusion on masticating behavior, because masticating behavior decreased by the IPA infusion into the jugular vein of goats during spontaneous meals, suggesting that ruminants may adjust their ruminating and masticating behavior according to the IPA concentration in the blood. Montgomery and Baumgardt (1965) supported the hypothesis that ruminants adjust voluntary food intake in relation to the physiological demand for energy if the rumen fill or the rumen load do not limit their consumption. The results of this study showed strong evidence that IPA concentrations in the jugular vein decrease ruminating and masticating behavior.

Plasma glucose and VFA concentrations increased after IPA infusion, suggesting that the IPA may be converted to these compounds. Other studies support this hypothesis. IPA is metabolized to acetone in rats (Lahman et al., 1980) and cows (Thin et al., 1959), and, finally, acetone is metabolized to glucose in cows (Black et al., 1972), and to VFA in goats (Oshiro et al., 2000). However, we consider that the increases in plasma glucose and VFA do not affect the decrease in masticating behavior in this experiment, because other studies have indicated that glucose (Robert et al., 1959) and VFA (Forbes, 1986) infused intravenously do not reduce food intake in ruminants. Also fatty acids can not pass through the blood brain barrier (Voet and Voet, 1995).

We conclude that the immediate reduction of the mastication behavior and ruminating behavior after IPA infusion was not by the increase in blood glucose and VFA concentrations during IPA infusion, but by the increasing blood IPA concentrations themselves after IPA infusions.

Our data suggest that rumination receptors sensitive to blood IPA are more likely to be in such as the brain stem (Anderson, 1951), where they can respond to blood levels.

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Table 2. Concentration of plasma compounds in three experiments

Items		Controls		Treatment
		Non-infusion	Ringer-infusion	IPA-infusion
glucose (mg/dl)	P-I	62.2 ± 3.1	72.6 ± 1.9	70.2 ± 8.0
	D-I	67.3 ± 4.6	73.2 ± 4.1	89.8 ± 17.9
NEFA (mEq/l)	P-I	0.20 ± 0.003	0.22 ± 0.02	0.21 ± 0.07
	D-I	0.22 ± 0.01	0.23 ± 0.03	0.25 ± 0.04
Acetic acid (m mol/l)	P-I	0.949 ± 0.199	1.064 ± 0.126	0.873 ±0.522
	D-I	0.945 ± 0.340	1.095 ± 0.263	1.311 ±0.502
Propionic acid (m mol/l)	P-I	0.183 ± 0.062	0.072 ± 0.021	0.183 ±0.035
	D-I	0.113 ± 0.046	0.111 ± 0.041	0.449 ±0.070
Butyric acid (m mol/l)	P-I	0.159 ± 0.045	0.113 ±0.036	0.120 ±0.010
	D-I	0.165 ± 0.055	0.147 ±0.040	0.357 ±0.013*
Iso-Valeric acid (m mol/l)	P-I	0.152 ± 0.061	0.111 ±0.046	0.176 ±0.043
	D-I	0.185 ± 0.065	0.131 ±0.024	0.273 ±0.052*
Acetone (m mol/l)	P-I	0.601 ± 0.147	0.890 ±0.329	0.701 ±0.032
	D-I	0.412 ± 0.087	0.630 ±0.283	1.174 ±0.540
IPA (m mol/l)	P-I	0.027 ± 0.008	0.022 ±0.006	0.019 ±0.007
	D-I	0.028 ± 0.015	0.021 ±0.009	0.110 ±0.040*

P-I: pre-infusion, D-I: during-infusion.

* Means±S.D. in the same row having the superscripts are significantly different ($p<0.05$).

Table 3. Concentration of rumen compounds in three experiments

Items		Controls		Treatment
		Non-infusion	Ringer-infusion	IPA-infusion
Acetic acid (m mol/l)	P-I	71.088 ± 14.193	60.550 ± 3.333	43.800 ± 20.715
	D-I	71.226 ± 8.668	56.870 ± 2.871	62.490 ± 6.403
Propionic acid (m mol/l)	P-I	31.088 ± 6.059	18.651 ± 2.000	14.841 ± 4.513
	D-I	30.041 ± 4.805	17.253 ± 1.953	21.943 ± 6.455
Butyric acid (m mol/l)	P-I	7.738 ± 1.603	5.378 ± 0.549	5.514 ± 3.952
	D-I	7.827 ± 1.680	4.879 ± 0.667	8.357 ± 0.698
Iso-Valeric acid (m mol/l)	P-I	0.489 ± 0.254	0.380 ± 0.225	0.339 ± 0.109
	D-I	0.556 ± 0.305	0.475 ± 0.227	0.689 ± 0.492
Acetone (m mol/l)	P-I	0.247 ± 0.154	0.146 ± 0.109	0.462 ± 0.090
	D-I	0.340 ± 0.119	0.220 ± 0.124	0.543 ± 0.156
IPA (m mol/l)	P-I	0.059 ± 0.028	0.062 ± 0.005	0.039 ± 0.010
	D-I	0.083 ± 0.043	0.064 ± 0.022	0.046 ± 0.026

P-I: pre-infusion, D-I: during-infusion.

* Means±S.D. in the same row having the superscripts are significantly different ($p<0.05$).

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