

Effect of monensin on feed utilization and gastrointestinal fermentation in the hamster (*Mesocricetus auratus*)

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1. Three experiments were conducted to examine the effect of monensin on growth performance, feed utilization and volatile fatty acids (VFA) in the forestomach and caecum of hamsters.
2. In Expt 1, monensin was fed at levels of 0, 5, 10 and 20 mg/kg to the growing male and female hamsters given a commercial diet (major component: lucerne (*Medicago sativa*) meal). In Expt 2, monensin was fed at levels of 0, 5, 15, 45 and 135 mg/kg to the growing male hamsters given a semi-purified diet containing 10 g urea/kg (main components: maize starch, sucrose, casein and cellulose). In Expt 3, monensin was fed at levels of 0 and 40 mg/kg to the growing male hamsters given the commercial diet containing lucerne meal or a semi-purified diet.
3. In Expt 1, monensin improved feed conversion efficiency and growth performances in the young growing hamsters, but monensin did not affect the hamsters at a later growing stage.
4. In response to monensin the proportion of acetic acid increased and that of propionic acid decreased in the forestomach, whereas the proportion of acetic acid decreased and that of propionic acid increased in the caecum in Expt 2. The hamsters given 135 mg monensin/kg ate less, developed diarrhoea and died.
5. The apparent digestibility of crude protein (nitrogen \times 6.25) was improved by monensin but those of dry matter and neutral-detergent fibre (NDF) were decreased in hamsters given the semi-purified diet in Expt 3. Monensin did not appear to have a significant effect on the apparent digestibility of the diet containing lucerne meal.
6. The responses to monensin in hamsters are compared with those in ruminants.

Monensin, a biologically active compound produced by the actinomycetes *Streptomyces cinamonensis*, has been shown to increase feed conversion efficiency in ruminants on both high-roughage and high-concentrate diets (Gill *et al.* 1976; Potter *et al.* 1976; Raun *et al.* 1976; Utley *et al.* 1976). Monensin appears to be capable of altering rumen fermentation. Increased molar proportions of rumen propionic acid, caused by the addition of monensin to the diet, have been reported by many investigators (Dinius *et al.* 1976; Richardson *et al.* 1976; Boling *et al.* 1977; Perry *et al.* 1979). Van Maanen *et al.* (1978) indicated that increases in rumen propionate production and in glucose metabolism were caused by monensin. These influences have been implicated as the primary factors responsible for the increased feed conversion efficiency seen with monensin supplementation (Richardson *et al.* 1976).

The golden hamster possesses a distinctly compartmentalized stomach which consists of a forestomach and a glandular stomach. Great numbers of protozoa and bacteria are found in the forestomach (Kunstýr, 1974; Imai *et al.* 1976), and both the production and absorption of volatile fatty acids (VFA) have been demonstrated (Hoover *et al.* 1969).

The hamster also has a well-developed caecum which plays an important role in food utilization, especially in utilization of a high-fibre diet (Sakaguchi *et al.* 1981). The amount of VFA in the caecum was increased two to five times by roughage feeding compared with concentrate feeding (Manda, 1979). Furthermore, activities of enzymes of the hexose monophosphate pathway involved in lipid synthesis (glucose-6-phosphate dehydrogenase, EC 1.1.1.49; phosphogluconate dehydrogenase (decarboxylating), EC 1.1.1.44) in the

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Table 1. Comparison (g/kg) of the semi-purified diet

Expt no. . . .	2	3
Maize starch	420	500
Sucrose	100	100
Casein	130	160
Soya-bean oil	60	60
Cellulose	200	100
Vitamin mix*	60	60
Mineral mix†	20	20
Urea	10	—

* Vitamin mix (mg/kg): retinol 120, cholecalciferol 2, α -tocopherol 100, thiamin 790, pyridoxine 390, riboflavin 1590, calcium pantothenate 2520, nicotinamide 3440, choline 5000, pteroylmonoglutamic acid 50, biotin 10, hydroxycobalamin 2, ψ -inositol 11.76, ascorbic acid 5880, menadione 160.

† Mineral mix: Harper (1959).

liver were decreased with the reduction of malic enzyme activity in hamsters given a roughage diet, and high uptake of [$1\text{-}^{14}\text{C}$]acetate and low uptake of [$U\text{-}^{14}\text{C}$]glucose were observed in the liver and epididymal adipose tissue of hamsters given a forage-containing diet compared with hamsters given concentrates (Murai & Manda, 1977). These reports may indicate that VFA produced in the caecum contributes significantly to the energy metabolism in the hamster given a forage-containing diet.

The following experiments were conducted to study the responses to monensin in growth, feed utilization and gastrointestinal fermentation in the hamster.

MATERIALS AND METHODS

Animals and feeding

Expt 1. Random-bred, male and female hamsters, average weight 52 g, were initially allocated to four treatment groups to receive 0, 5, 10 and 20 mg monensin/kg diet. Each group contained four male and four female hamsters.

The animals were individually housed in wire-bottom cages. The animal room was maintained at a temperature of $22 \pm 2^\circ$. A 14 h light–10 h dark cycle was automatically maintained. During the 42 d experimental period, body-weight and feed consumption were recorded at 6 and 2 d intervals respectively.

The hamsters were given a commercial diet (ZF; Oriental Yeast Co. Ltd, Tokyo) containing 0, 5, 10 and 20 mg monensin/kg *ad lib.* for 42 d. ZF was composed of lucerne (*Medicago sativa*) meal as a major component with supplements of wheat bran, soya-bean meal, beet pulp, wheat flour, skim-milk powder, fish meal, yeast, vitamins and minerals, and contained (g/kg dry matter): 198 crude protein (nitrogen $\times 6.25$), 35 crude fat, 149 crude fibre, 76 crude ash, 541 N-free extract and 389 neutral-detergent fibre (NDF).

Expt 2. Growing, male golden hamsters, average weight 66 g, were initially allocated to five treatment groups to receive 0, 5, 15, 45 and 135 mg monensin/kg diet. Each group contained five animals.

The animals were housed under the same conditions as in Expt 1. A semi-purified diet containing 10 g urea/kg, as shown in Table 1, was fed *ad lib.* to the hamsters for 32 d. Water was available *ad lib.* During the experimental period, body-weight and feed consumption were recorded at the same intervals as in Expt 1.

During the last day of the experimental period, all animals were fasted for 6 h and then

fed for 15 min. At 2 h after the beginning of the 15 min feeding, the animals were killed by diethyl ether inhalation. The forestomach and caecum were removed, chilled and stored at -20° .

Expt 3. Growing, male golden hamsters, average weight 45.2 g, were allocated to the following four groups, each containing six or seven animals: (1) semi-purified diet containing no monensin, (2) semi-purified diet containing 40 mg monensin/kg, (3) ZF (same as in Expt 1) containing no monensin, (4) ZF containing 40 mg monensin/kg. The diets were fed *ad lib.* to the animals for 14 d. Faeces were collected and food consumption was recorded daily during the last 6 d. The animals were housed individually in 150 mm \times 200 mm wire-bottom metabolism cages during the experimental period. The composition of the semi-purified diet is shown in Table 1.

Analytical methods

Fractions of VFA in the forestomach and caecum whole-contents were obtained by the method of Fenner & Elliot (1963) and analysed with a gas-liquid chromatograph (Yanaco G 180) equipped with a flame-ionization detector and a glass column (2.0 m) packed with Chromosorb 101 and operated at 180° with N_2 as a carrier gas.

The pooled faecal samples were oven dried at 60° and analysed in duplicate for N by the Kjeldahl procedure (Association of Official Analytical Chemists, 1975) and for NDF by the method proposed by Van Soest & Wine (1967). Dry matter in the samples was determined after drying for 2 h at 135° .

Statistics

The results for weight gain and feed conversion efficiency were analysed statistically as a 4×2 factorial design. The analysis included monensin treatment with four dose levels (0, 5, 10 and 20 mg/kg) and both sexes (male and female). All other results were tested for statistical difference by analysis of variance (Snedecor & Cochran, 1967) and Duncan's multiple-range test (Duncan, 1955).

RESULTS

Expt 1. Growth and feed conversion efficiency

Weight gain and feed conversion efficiency of male and female hamsters during the experimental period are presented in Table 2. The experimental period (42 d) was divided into two periods: the first part (period 1) 24 d and the second part (period 2) 18 d. The weight gain and feed conversion efficiency of male hamsters tended to improve by addition of monensin during both periods. In the female hamsters, the weight gain and feed conversion efficiency were higher in the animals given the monensin diet than in the control animals in period 1, but the effects of monensin on the weight gain and food conversion efficiency of female hamsters were not marked in period 2. Values obtained by a 4×2 factorial analysis of variance (Table 3) showed that the effects of addition of monensin on the weight gain and feed conversion efficiency were significant ($P < 0.05$) in period 1, but not significant in period 2. The values obtained by analysis of variance showed that the difference of sex significantly affected ($P < 0.05$) the weight gain and feed conversion efficiency of hamsters in period 2 and, hence, overall. Average daily weight gain and feed conversion efficiency were significantly higher in period 1 than in period 2.

Expt 2. Growth performance and gastrointestinal VFA

Average weight gain, feed consumption and feed conversion efficiency of male hamsters during the experimental period are presented in Table 4. Weight gain and feed conversion

Table 2. *Expt 1. Average weight gain and feed conversion efficiency in hamsters given monensin*

(Mean values and standard deviations for four animals per group)

Dietary monensin (mg/kg)	Period 1 (24 d)*							
	Wt gain (g)				Feed conversion efficiency†			
	♂		♀		♂		♀	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0	24.7	1.3	26.1	3.7	9.2	0.5	9.9	2.8
5	33.3	5.4	32.8	7.4	12.9	2.0	11.4	2.3
10	25.6	2.6	30.4	5.3	10.4	0.4	11.5	1.0
20	28.8	7.1	32.4	1.5	11.2	1.9	12.0	1.0
Dietary monensin (mg/kg)	Period 2 (18 d)*							
	Wt gain (g)				Feed conversion efficiency†			
	♂		♀		♂		♀	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0	7.5	2.5	16.4	2.9	4.2	1.4	8.3	1.5
5	13.4	5.5	11.0	1.7	7.2	2.8	6.0	0.8
10	10.0	1.8	23.4	5.0	5.5	1.1	11.6	2.4
20	12.5	2.2	14.7	1.3	6.8	0.8	7.6	0.7

* For details, see p. 149.

$$\dagger \text{ Feed conversion efficiency} = \frac{\text{weight gain}}{\text{feed consumption}} \times 100.$$

Table 3. *Expt 1. Results of two-way factorial analysis of variance*

Experimental period...	1 (24 d)		2 (18 d)	
	Wt gain	Feed conversion efficiency	Wt gain	Feed conversion efficiency
Monensin	*	*	NS	NS
Sex	NS	NS	**	*
Interaction	NS	NS	NS	NS

NS, not significant.

* $P < 0.05$, ** $P < 0.01$.

efficiency of male hamsters given 5–45 mg monensin/kg tended to increase with dose level, but all animals that were given 135 mg monensin/kg had diarrhoea and died early in the experimental period: three animals on the 6th day, one animal on the 8th day, one animal on the 12th day after the start of feeding of monensin. Feed consumption during the initial 4 d of the experimental period was depressed significantly ($P < 0.05$) by the 135 mg monensin/kg diet.

Table 4. *Expt 2. Body-weight gain, feed consumption and feed conversion efficiency in growing male hamsters (32 d trial)*
(Five animals per group)

Dietary monensin (mg/kg)	Wt gain (g) (A)	Feed consumption (g)		Feed conversion efficiency $\left(\frac{A}{B} \times 100\right)$
		Initial (4 d)	Total (32 d) (B)	
0	28.7	39.6 ^a	288.1	10.0
5	30.3	40.4 ^a	289.3	10.4
15	32.8	41.2 ^a	285.5	11.5
45	34.7	42.8 ^a	293.2	11.8
135*	—	30.0 ^b	—	—
SE of mean	2.92	2.95	6.90	0.92

^{a, b} Mean values in the same vertical column with different superscript letters were significantly different ($P < 0.05$).

* All animals died within 12 d of the monensin feeding period.

Table 5. *Expt 2. Volatile fatty acid (VFA) composition in the forestomach and caecum of young male hamsters*
(Five animals per group)

Dietary monensin (mg/kg)	Forestomach			
	Total VFA ($\mu\text{mol/pouch}$)	mmol/mol		
		Acetic	Propionic	Butyric
0	75	778 ^a	117	105 ^a
5	81	817 ^{a, b}	129	54 ^{a, b}
15	54	837 ^{a, b}	121	43 ^{a, b}
45	88	914 ^b	66	20 ^b
135*	—	—	—	—
SE of mean	16.2	26.1	24.6	11.1
Dietary monensin (mg/kg)	Caecum			
	Total VFA ($\mu\text{mol/pouch}$)	mmol/mol		
		Acetic	Propionic	Butyric
0	135	750 ^a	182 ^a	68 ^a
5	130	763 ^a	180 ^a	57 ^a
15	126	724 ^a	233 ^a	43 ^b
45	150	647 ^b	313 ^b	40 ^b
135*	—	—	—	—
SE of mean	12.8	19.3	17.9	5.8

^{a, b} Mean values in the same vertical column with different superscript letters were significantly different ($P < 0.05$).

* All hamsters died within 12 d of the monensin feeding period.

Table 6. *Expt 3. Apparent digestibilities (g/g) of dry matter, crude protein (nitrogen \times 6.25) and neutral detergent fibre (NDF) in young male hamsters*

(Values were obtained in a 6 d digestion trial)

Diet	Dietary monensin (mg/kg)	No. of hamsters	Dry matter	Crude protein	NDF
Semi-purified	0	7	0.91	0.79	0.79
	40	7	0.84**	0.87**	0.08**
SE of mean			0.006	0.012	0.026
ZF (high fibre)	0	7	0.55	0.50	0.40
	40	6	0.57	0.55	0.42
SE of mean			0.014	0.020	0.016

** Values were significantly different from the control value (no monensin) in the same vertical column ($P < 0.01$).

The effect of monensin on VFA of forestomach and caecum contents is shown in Table 5. Monensin increased the proportion of acetic acid and decreased that of butyric acid in the forestomach. Propionic acid in the forestomach was not affected by monensin. On the other hand, in the caecum, the proportions of acetic and butyric acids were decreased and that of propionic acid was increased by monensin feeding. The changes in VFA composition caused by monensin were approximately proportional to the dose level, although the effect of monensin on total VFA concentration was not apparent. The weights of the whole contents of the forestomach and caecum were (mean (SD)) 1.09 (0.51) g and 1.67 (0.55) g respectively for thirteen animals.

Expt 3. Apparent digestibility

Apparent digestibilities of dry matter, crude protein and NDF in the hamsters given 0 or 40 mg monensin/kg are shown in Table 6. In hamsters given the semi-purified diet, apparent digestibilities of dry matter and NDF were significantly ($P < 0.01$) decreased by monensin; however, the digestibility of crude protein was significantly ($P < 0.01$) increased by monensin. In the ZF-fed hamsters, digestibilities of dry matter, crude protein and NDF were improved by monensin but not significantly.

DISCUSSION

Stimulative effects of monensin on weight gain and feed conversion efficiency in the hamster were observed in the present experiment. Monensin has been shown to increase feed conversion efficiency in ruminants on various diets (Potter *et al.* 1976; Raun *et al.* 1976; Boling *et al.* 1977; Turner *et al.* 1977). However, monensin has a tendency to depress the rate of weight gain and feed conversion efficiency in urea-fed growing steers (Coombe *et al.* 1979; Hanson & Klopfenstein, 1979) but it had no negative effect on growth and feed conversion efficiency in urea-fed hamsters in this experiment.

The forestomach of the golden hamster and the rumen of the ruminant are similar in that they both play a significant role in the utilization of dietary urea for microbial protein synthesis (Sakaguchi *et al.* 1981). However the microbial flora in the hamster forestomach (Kunstýr, 1974; Imai *et al.* 1976) is different from that in the rumen (Hungate, 1966) and a lower pH value (3.7–6.0) has been observed in the hamster forestomach (Hoover *et al.*

1969; Kunstýr, 1974) than the pH value (about 6.7) in the rumen (Maynard *et al.* 1979). The effect of monensin on the proportions of VFA in the forestomach was different qualitatively from that in the caecum in Expt 2, although the response of caecal proportions of VFA to monensin was similar to that of rumen proportions of VFA (Potter *et al.* 1976; Raun *et al.* 1976; Boling *et al.* 1977). This may suggest a difference in the response of microbial protein synthesis to monensin in the rumen and in the hamster forestomach.

In the diets given to steers, 360–660 g/kg total N were replaced by urea-N (Hanson & Klopfenstein, 1979; Poos *et al.* 1979) but the diet given to the hamsters in this experiment contained only about 190 g urea-N/kg total N. This smaller contribution of urea-N to protein metabolism in the hamster might be another reason for the difference in the response to monensin between the ruminants and the hamsters on the urea-containing diet.

Acetic acid concentration was increased in the forestomach, while propionic acid concentration was increased in the caecum, by monensin addition to the diet. Van Maanen *et al.* (1978) indicated that monensin increased glucose metabolism and rumen propionate production. Chen & Wolin (1979) showed that monensin enhanced selection of microbial communities that produce relatively more propionic acid than other rumen VFA. The difference in the response of microbial fermentation to monensin between the forestomach and the caecum may suggest a difference of microbial flora between the two compartments. It is possible that VFA produced in the caecum contribute more to the hamster's energy supply than VFA produced in the forestomach (Sakaguchi *et al.* 1981). Therefore, the improvement of feed conversion efficiency caused by monensin feeding may have been partly associated with the alteration of proportions of VFA in the caecum.

Monensin decreased the digestibilities of dry matter and NDF in the hamsters on a semi-purified diet. Poos *et al.* (1979) reported that, in the lamb, the digestibilities of dry matter and acid-detergent fibre (ADF) were significantly reduced by monensin. These results are in agreement with those obtained from *in vitro* rumen incubation studies (Lemenager *et al.* 1978; Wallace *et al.* 1981). On the other hand, other workers reported significantly higher digestion coefficients for dry matter and fibrous compounds (crude fibre or NDF) for ruminants given monensin (Utley *et al.* 1977; Horton & Stockdale, 1979; Horton *et al.* 1980; Thompson & Riley, 1980; Wedegaertner & Johnson, 1983). However, some reports show little significant effect of monensin on the digestibilities of dry matter (Dinius *et al.* 1976; Utley *et al.* 1977; Muntifering *et al.* 1980; Thompson & Riley, 1980) and crude fibre or NDF (Dinius *et al.* 1976; Utley *et al.* 1977; Horton, 1980).

In hamsters on the high-fibre diet (ZF), it appears that monensin slightly increased the digestibilities of dry matter, crude protein and NDF. As the digestive system of hamsters changes to the herbivorous type by feeding roughage (Manda, 1979), the difference in the response of digestibilities of dry matter and NDF to monensin between the hamsters given a high-fibre diet and those given a semi-purified diet may be attributable to the difference in the adaptation of the digestive system to each diet.

The digestibility of crude protein was increased by monensin for hamsters on the semi-purified diet. This may be attributable to a decrease of proteolytic activity and microbial protein synthesis in the forestomach and caecum because *in vitro* studies showed that total and net microbial growth were considerably decreased by addition of monensin. Furthermore, during incubations with casein, monensin lowered protein degradation in line with a lowered ammonia production (Van Nevel & Demeyer, 1977). Even though an increase in the digestibility of crude protein does not always reflect an improvement of protein utilization, it is possible that protein utilization was improved by monensin in the hamsters as in the ruminant (Joyner *et al.* 1979; Perry *et al.* 1979, 1983).

All the hamsters given 135 mg monensin/kg drastically reduced their feed intake and died after several days. Raun *et al.* (1976) reported that feed intake and weight gain were also

reduced by giving 88 mg monensin/kg to cattle kept in feedlots. Although it is not clear from this experiment what the maximum permissible dose of monensin is for the hamster, it seems that it may be similar to that for the ruminant.

In conclusion, monensin promotes growth and improves feed conversion efficiency in the hamster. However, the effect is dependent on dietary conditions and the growth stage. Alteration of the gastrointestinal microbial fermentation pattern caused by monensin may be at least partly responsible for the stimulating effect on growth and feed conversion efficiency. In the hamster, the caecum is considered to be more important as a fermentation chamber than the forestomach.

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