

# Effect of Coprophagy Prevention on Heat Production and Body Composition of Fasting Rats

By

Seizi SUKEMORI\*, Shuhei IKEDA\* and Yoshio KURIHARA\*

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**Summary** : This study aims to clarify the effect of coprophagy on heat production and body composition of fasting rats. Eight male Sprague-Dawley (SD) strain rats were used to determine heat production under coprophagy -prevented and -allowed conditions. Twelve male SD strain rats were used to determine whole body chemical composition under coprophagy -prevented and -allowed conditions. Though heat production 72 hrs from the starting point showed no significant difference, carbon dioxide generation in the coprophagy-prevented group was significantly higher than that in coprophagy-allowed group. Rats in the coprophagy-prevented group attained the fasting condition, which was indicated in the RQ value of 0.69, 12 hrs earlier in comparison with coprophagy-allowed group rats. Ether extracts in the coprophagy-prevented rat body were significantly lower than that in the coprophagy-allowed rat. These results suggest that coprophagy prevention accelerates body fat utilization.

**Key Words** : rats, coprophagy, heat production under fasting condition

## Introduction

In previous studies<sup>18)</sup>, we learned rats obtained vitamins from their own hydrous feces by eating them as coprophagous behavior. The prevention of coprophagy by physical apparatus reduced the growth of animals<sup>1,5,8,17)</sup>, i.e. deficiency of vitamins affected protein metabolism and/or energy metabolism. Caecectomy prevents the ingestion of hydrous feces by biological means, but caecectomized rats consumed a large amount of feed instead of ingested hydrous feces<sup>15)</sup>. Therefore, coprophagy prevention disturbs the nutritional phenomenon. Simplified recognition of the effect of coprophagy on heat production is possible under fasting conditions. The present experiment aims to compare heat production in coprophagy-allowed and coprophagy-prevented rats during fasting conditions. Furthermore, whole body chemical composition related to heat production was also compared.

## Materials and methods

### Animals and feed :

Twenty healthy male Sprague-Dawley strain rats 4-week-old were purchased from Japan Clea Co. Ltd., Tokyo. Eight with an initial body weight of  $163.7 \pm 1.9$  g were used to determine heat production and 12 with an initial body weight of  $170.3 \pm 3.2$  g were used to determine whole body chemical composition. They were divided into two groups, coprophagy-allowed and coprophagy-prevented groups. They were usually given commercial feed (CE-II, Japan Clea Co. Ltd., Tokyo) and water ad libitum. They were fed in individual dormitory-type cages ( $38 \times 25 \times 21$  cm) and were kept in a temperature controlled room at  $23 \pm 1^\circ\text{C}$  except during the period of heat production determination. Daylight period was from 6 : 00 to 18 : 00.

### Determination of heat production :

Heat production was determined by using a HALDANE type respiration apparatus<sup>12,14)</sup>. In this experiment we used 2 sets of HALDANE type respiration apparatus for

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\* Department of Animal Science, Faculty of Agriculture, Tokyo University of Agriculture

simultaneous determination. Rats were accommodated in a coprophagy-prevented tube cage<sup>13)</sup> and a coprophagy-allowed small cage fitting their body size without feed, respectively. The coprophagy-allowed small cage prevented wasteful feed investigation behavior that was induced by fasting. They could drink water freely. These cages were placed into the chamber of the HALDANE type respiration apparatus. Aeration was 3 l/min and a 72-hour test was conducted. Consumption of oxygen, generation of carbon dioxide and respiration quotient (RQ) were determined every 12 hours. Heat production was calculated from data according to BENEDICT<sup>2)</sup>.

#### Determination of chemical composition of fasting rat body :

After the appointed fasting period, rats were killed with diethyl ether. Their entire body was processed with a meat chopper to determine the chemical composition. Determination of chemical composition (moisture, crude protein and ether extracts as crude fat) and calories was conducted using the usual methods for meat<sup>3)</sup>. Glycogen was also determined by the following procedure : settled meat was dissolved by 40% KOH and 95% alcohol poured into it for the condensation of glycogen. After the filtration of glycogen, it was dissolved again by 25% HCl and determined using BERTLAND method<sup>4)</sup>.

#### Statistical treatment :

Data were treated by one-way analysis of variance and multiple range test by Tukey at significance level

of 0.05.

## Results

#### Heat production of fasting rats :

Oxygen consumption, carbon dioxide generation, RQ and heat production after 48 hrs from the start of fasting are shown in Table 1. There was no significant difference. The results obtained after 60 hrs from the start of fasting are shown in Fig. 1. Carbon dioxide generation in the coprophagy-prevented group showed significantly ( $P < 0.05$ ) lower values than that of the coprophagy-allowed group. RQ value in coprophagy-prevented group was also significantly lower than that of coprophagy-allowed group. There was no significant difference in heat production between coprophagy-prevented and -allowed groups, and it showed the similar tendency.

#### Chemical composition of fasting rats :

Chronological change in the body weight is shown in Fig. 2. Although coprophagy prevention slightly decreased the body weight, there was no significant difference between both groups. Chemical composition of fasting rats is shown in Table 2. There was no significant difference in the moisture, crude protein, calories and glycogen between both groups. Ether extracts in coprophagy-prevented group was significantly ( $P < 0.05$ ) lower than that in coprophagy-allowed group 48 hours from the start point.

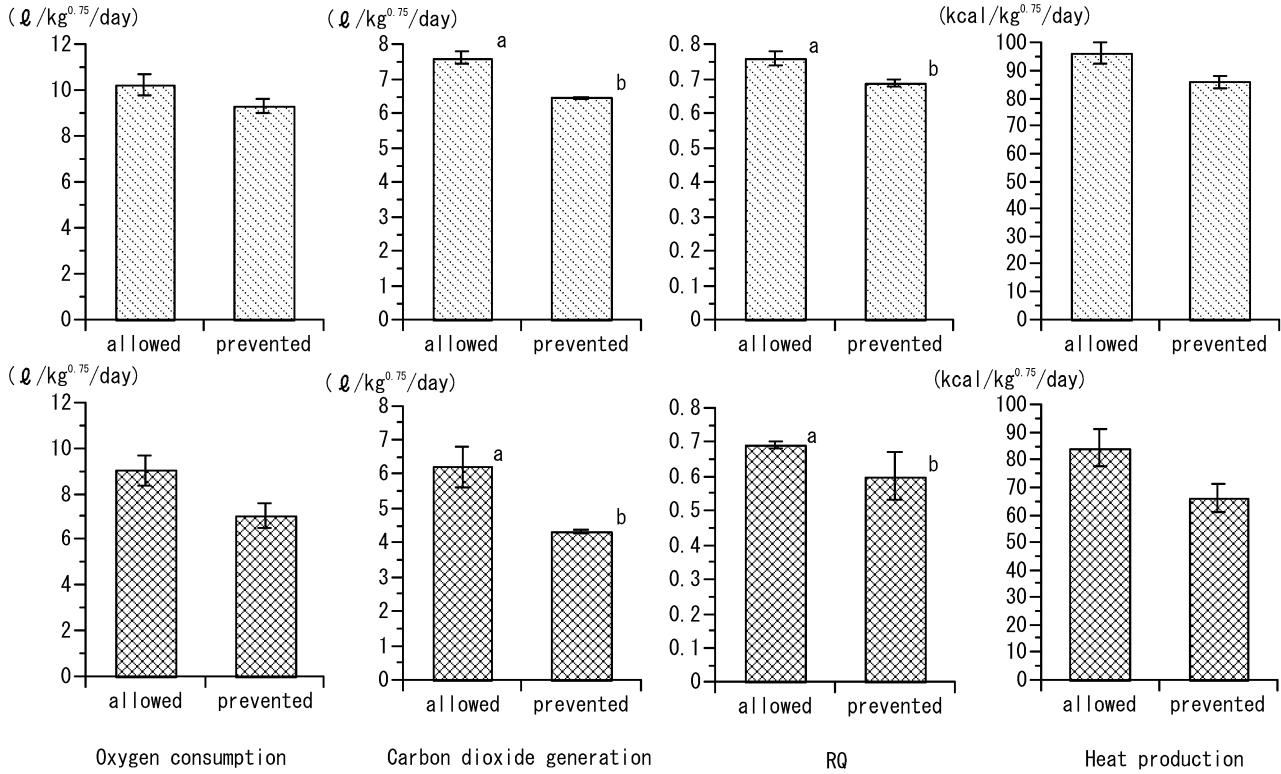
## Discussion

Basal metabolism of energy, i.e. heat production of

**Table 1** Results of respiration test after 48 hrs from the start of fasting

Experimental group		Oxygen	Carbon dioxide	RQ	Heat production
		consumption ( $\ell/\text{kg}^{0.75}/\text{day}$ )	generation ( $\ell/\text{kg}^{0.75}/\text{day}$ )		( $\text{kcal}/\text{kg}^{0.75}/\text{day}$ )
Coprophagy-allowed	12hrs	12.6±0.82	10.2±0.4	0.82±0.08	121.2±5.22
	24hrs	14.0±0.94	9.83±0.83	0.71±0.01	131.5±8.89
	36hrs	11.6±0.11	8.81±0.08	0.76±0.01	110.6±0.64
	48hrs	11.3±0.94	8.50±0.40	0.76±0.02	107.3±8.24
Coprophagy-prevented	12hrs	14.5±0.49	11.3±0.01	0.78±0.04	138.5±3.46
	24hrs	14.4±0.77	10.5±0.51	0.73±0.01	138.1±4.70
	36hrs	12.4±0.71	9.89±0.50	0.80±0.01	118.6±6.62
	48hrs	10.1±0.51	7.85±0.19	0.78±0.02	96.2±4.40

Mean±S.D.; n=4.

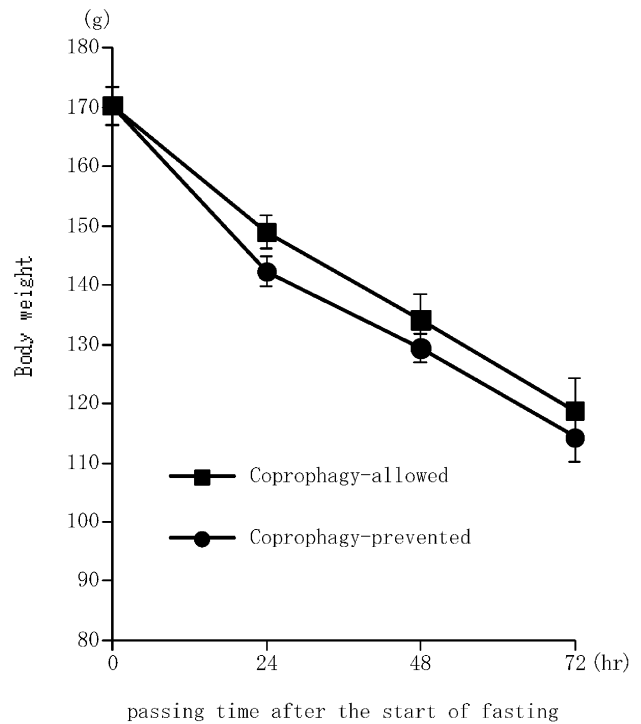


**Fig. 1** Results of respiration test

The upper row shows the results of respiration test after 60 hrs from the start of fasting and the lower row shows the results after 72 hrs from the start point. Significant ( $P < 0.05$ ) difference was recognized between different superscript letters.  $n = 4$ .

fasting animals, relates to the metabolic body size ( $\text{kg}^{0.75}$ ), and the rats having small body weight require large energy in comparison with other animals having large body weight. KLEIBER *et al.*<sup>11)</sup> reported that heat production of fasting rats decreased in accordance with body weight gain and this phenomenon was also observed in our previous report with piglets<sup>9)</sup>. Further, coprophagy frequency in rats also decreased in accordance with body weight gain<sup>6,7)</sup>.

In the results of the respiration test offering heat production, significant differences were recognized in the values of carbon dioxide generation and RQ although not in heat production. RQ is about 0.7 under the fasting condition. Therefore, both the groups reached the fasting condition animals' 72 hrs. The coprophagy-prevented group showed fasting condition attained 12 hrs earlier than coprophagy-allowed group. This tendency suggested that the coprophagy prevention accelerates a decrease in the speed of heat production. The difference of RQ may be induced by the carbon dioxide generation. The difference in carbon dioxide generation reflected the consumption of ether extracts, which is an important material in energy production after glycogen consumption during fasting conditions<sup>10)</sup>. The results of statistical treatment heat



**Fig. 2** Chronological change of body weight after the start of fasting  
There was no significant difference in each time point.

**Table 2** Chemical composition of fasting rats' body

Experimental group	Moisture (g)	Crude protein (g)	Ether extracts (g)	Calories (kcal)	Glycogen (g)
Start point	125.3±2.4	33.3±0.22	7.96±0.93	249.6±5.5	3.60±0.07
	100%	100	100	100	100
After fasting					
Coprophagy -allowed	83.9±6.1	27.7±1.0	2.14±0.37 <sup>a</sup>	162.9±7.9	2.77±0.20
	67.2	83.4	28.8	65.4	76.9
Coprophagy-prevented	82.5±3.0	27.0±1.1	1.73±0.04 <sup>b</sup>	154.5±5.8	2.65±0.25
	65.9	81.3	22.6	60.5	73.6

Mean±S.D.; n=4. Significant (P<0.05) difference was recognized between different superscript letters in same vertical column.

production showed no significant difference, but the coprophagy-prevented group showed lower values than those in coprophagy-allowed group. Oxygen consumption also showed the same tendency. In our previous report<sup>18)</sup>, we noted that hydrous feces, i.e. ingested feces by coprophagous behavior includes large amount of vitamin B<sub>12</sub> and folic acid. These vitamins play a role in protein metabolism and DNA synthesis<sup>16)</sup>. The prevention of protein metabolism and/or DNA synthesis might induce a failure in nutritional metabolism affecting fat metabolism. Furthermore, a direct factor relating to fat metabolism, whether the acceleration of fat utilization or the prevention of fat synthesis, may be hydrous feces, although we have not yet recognized it. This hypothesis remains to be clarified in a future work.

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# ラットの食糞行動の阻止が基礎代謝量および体成分に及ぼす影響

祐森誠司\*・池田周平\*・栗原良雄\*

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要約：本試験はラットの食糞行動の阻止が基礎代謝量および体成分に及ぼす影響を明らかにすることを目的として行った。基礎代謝量の測定には8匹のSD系ラットを用い、平均体重が等しくなるように食糞行動を阻止した4匹と食糞行動を許容した4匹に配分した。飼料の制限を課して体成分を測定した試験には12匹のSD系ラットを用い、平均体重が等しくなるように試験開始時の補正用に4匹、食糞行動を阻止した4匹、食糞行動を許容した4匹に配分した。飼料の制限から72時間後の熱発生量には有意な差は認められなかったが、食糞行動阻止区の二酸化炭素発生量は食糞行動許容区の値よりも有意に高かった。食糞行動阻止区のラットは0.69というRQ（呼吸商）の値から判断すると食糞行動許容区よりも12時間早く飢餓状態に到達していた。食糞行動阻止区ラットの体脂肪は食糞行動許容区の値よりも有意に低かった。これらの結果は、食糞行動の阻止が体脂肪の利用を促進することを示唆している。

キーワード：ラット, 食糞行動, 基礎代謝量