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# Continuous Monitoring of Oxygen Consumption in Sheep Head and Estimation of Energy Expenditure from Oxygen Consumption

Kouji HIGUCHI, Takehiro NISHIDA<sup>1)</sup>, Osamu ENISHI<sup>1)</sup>, Agung PURNOMOADI<sup>2)</sup>, Koichiro UEDA<sup>3)</sup>  
and Fuminori TERADA<sup>4)</sup>

Department of Animal Physiology and Nutrition

<sup>1)</sup> Department of Animal Feeding and Management

<sup>2)</sup> Diponegoro University

<sup>3)</sup> Hokkaido University

<sup>4)</sup> National Agricultural and Bio-oriented Research Organization

## Abstract

An experiment was conducted with two non-lactating and non-pregnant ewes to evaluate a new method of determining the energy expenditure of the head. The sheep were previously prepared with skin-covered loops and with ultrasonic blood flow probes around the carotid artery, and they were fed Italian ryegrass hay. After inserting the blood gas sensors into the carotid artery and jugular vein, the blood oxygen concentration, blood flow rate, and activity of the animals were continuously monitored. Oxygen consumption of the head was determined by multiplying the carotid blood flow rate and the difference in oxygen concentration between arterial and venous blood. The oxygen concentration of the carotid artery and jugular vein were relatively constant throughout a 3-day period; however, a rapid rise in the blood flow rate was observed. The rise in the blood flow rate was synchronized with the chewing activity of the sheep. Thus, the changes in the oxygen consumption of the head were associated with chewing. The calculated energy expenditure of the head was greater during eating and rumination than during resting. The rise in energy expenditure was greater during eating than that during rumination.

**Key words:** Oxygen consumption, Blood flow rate, Chewing activity, Head, Sheep

## Introduction

Open circuit respiratory procedures have been well established in indirect calorimetry<sup>2)</sup>, and such procedures have contributed to estimates made on the whole body energy requirement of farm animals. In recent years, in order to increase the productivity of farm animals, it has become necessary to optimize feeding management to reduce feeding costs and environmental waste. To optimize feeding management, the net nutrient requirement for the production and activity of farm animals must be estimated.

It has been reported that ruminants spend a large amount of time eating and ruminating, and that the energy required for chewing (eating and ruminating) accounts for a considerable proportion of the animals' total energy requirement<sup>15,19)</sup>. However, the changes in energy expenditure associated with the animals' activities can not be clearly distinguished from the changes in the whole body energy expenditure; such changes are difficult to determine due to the time lag response in the open circuit respiratory procedure. Thus, further advances in this procedure are required to determine the short-

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Present address: <sup>2)</sup> Semarang, 50275, Indonesia

<sup>3)</sup> Faculty of Agriculture, Hokkaido University, Kitaku, Sapporo-shi, Hokkaido 060-8589, Japan

<sup>4)</sup> Kannondai 3-1-1, Tsukuba-shi, Ibaraki 305-8514, Japan

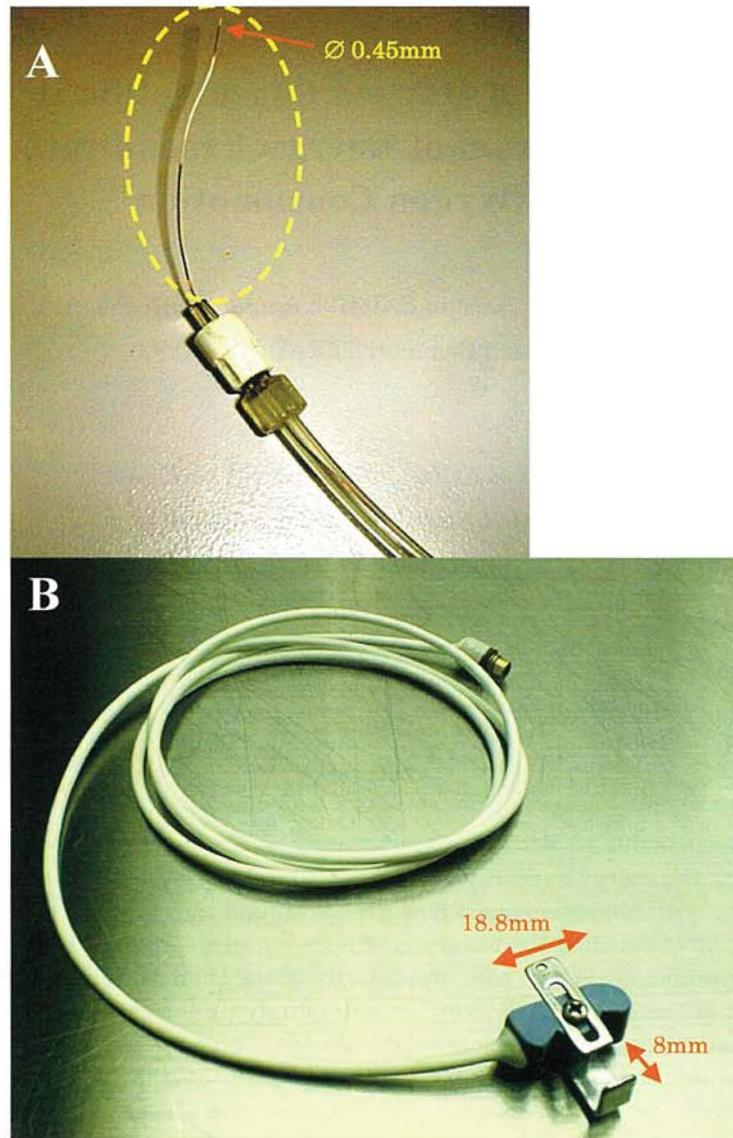


Fig. 1. A blood gas sensor (A) and a transit-time ultrasound blood flow probe (B). The yellow circle indicates the tip of the sensor.

term changes in energy metabolism in local tissues or in specific organs.

Energy expenditure in local tissues or in organs can be measured as oxygen consumption, as calculated from the blood flow and (multiplied by) the difference in the oxygen content between arterial and venous blood; such calculations are based on the application of the Fick principle. An ultrasound blood flow probe was developed for chronic measurement of the blood flow rate in the blood vessels, and an intra-vascular blood gas sensor for continuous monitoring of blood gas concentrations, such as those of oxygen and carbon dioxide, has been developed for the management of patients in intensive care units.

The objective of the present study was to develop a new method for continuously monitoring the oxygen consumption of the organs in order to esti-

mate their energy expenditure. The oxygen consumption of the sheep head was continuously monitored using an ultrasound blood flow meter and intra-vascular blood gas sensors. The energy expenditure of the entire body was also measured using indirect calorimetry in a respiration chamber.

#### Materials and Methods

Two non-lactating and non-pregnant ewes (one Corriedale, 28 kg; and one Suffolk, 31 kg) were kept in metabolic cages. Each of the two sheep had been surgically prepared with skin-covered loops established around the left carotid artery, and with chronic implantation of an ultrasonic flow probe (diameter, 8 mm; Transonic Systems Inc., Ithaca, New York, USA; Fig. 1) placed around the right carotid artery. Anaesthesia was induced with intravenous administration of 30 mg of ketamine hydrochloride and

0.6  $\mu$ g xylazine. Surgical anaesthesia was maintained with halothane and oxygen in a closed circuit during the surgical operation. The sheep were fed Italian ryegrass hay (884 g dry matter (DM)/kg, 87 g crude protein/kg DM and 18.0 MJ gross energy/kg DM) at a near-maintenance requirement level<sup>(10)</sup> of total digestible nutrients once daily at 10:00 h, and the sheep also had free access to water. After a seven-day preliminary period, whole feces and urine were collected for 6 days (experimental period). On the first days of the experimental period, intravascular blood gas sensors (Paratrend 7, Biomedical Sensors Ltd., High Wycombe, UK; Fig. 1) were calibrated and inserted into the sheep to a length of 10 cm in order to measure the blood oxygen concentration via the carotid cannulae and the jugular cannulae on the left side. The blood gas sensors were secured by an adhesive to the skin adjacent to the skin-covered loops of the carotid artery and jugular vein, and the sensors were connected to Paratrend 7 monitors. All cannulae were flushed continuously with a solution of normal saline containing heparin (2 units/ml) from a pressurized flush bag maintained at 300 mmHg through a side-port of the sensor. The ultrasound flow probe was connected with a Medical Flowmeter (T106, Transonic Systems Inc., Ithaca, New York, USA). The blood oxygen concentration and the blood flow rate were continuously measured throughout the first 3 days of the experimental period. Data were collected from the monitors at 10-sec intervals for the blood gasses, and at 1-min intervals for the blood flow, respectively. The data were transferred into personal computers via RS-232C serial ports and were then averaged every minute, and were then recorded as an ASCII file. The oxygen consumption was calculated using the following equation:  $OC = (O_{concA} - O_{concV}) \times BF$ , where  $OC$  = the oxygen consumption (ml/min),  $O_{concA}$  = the oxygen concentration of the carotid artery (ml/100 ml),  $O_{concV}$  = the oxygen concentration of the jugular vein (ml/100 ml), and  $BF$  = the blood flow rate of the carotid artery (ml/min). The oxygen consumption of the head was calculated as being equal to twice the  $OC$ , because there are two pairs of carotid arteries and jugular veins in the neck. The oxygen concentration was calculated using the following equation:  $O_{conc} = pO_2 / 760 \times 100 \times 0.024 + 1.34 \times Hb \times O_2 \text{ sat} / 100$ , where  $O_{conc}$  = the oxygen concentration (ml/100 ml),  $pO_2$  = the partial pressure of the oxygen,  $Hb$  = the hemoglobin concentration (g/100 ml), and  $O_2 \text{ sat}$  = the oxygen saturation (%). The hemoglobin concentration of the blood was determined by a fully automated blood gas analyzer (CHIRON 840, Bayer Medical Ltd., Tokyo, Japan) at the beginning of the experimental period. Chewing behavior was monitored using a video camera, and was recorded manually

every minute. Data regarding the blood oxygen concentration and the blood flow rate were classified and averaged every minute according to the chewing activity of the animal during resting, eating, and ruminating. During the last 3 days of the experimental period, the sheep were tested to determine the heat production by their entire bodies using an open-circuit respiration apparatus<sup>(9)</sup>. Heat production was calculated from the gaseous exchange and urinary excretion of nitrogen using the equation of Brouwer<sup>(31)</sup>:  $\text{Heat (kJ/day)} = O_2 \times 16.18 + CO_2 \times 5.02 - CH_4 \times 2.17 - N \times 5.99$ , where  $O_2$  = the oxygen consumption (L/day);  $CO_2$  and  $CH_4$  = the emission of carbon dioxide and methane (L/day), respectively; and  $N$  = the urinary nitrogen excretion (g/day). Heat production of the head was calculated using the following equation:  $\text{Heat (kJ/min)} = EW / OW \times OC$ , where  $EW$  = the whole body heat production (kJ/day),  $OW$  = the whole body oxygen consumption (L/day), and  $OC$  = the oxygen consumption (L/day). The methods used for the energy and chemical analyses of the feed, feces, and urine were the same as those described in our previous report<sup>(7)</sup>.

## Results

The total measuring times for the blood oxygen concentration during the first 3 days of the experimental period are shown in Table 1. The monitoring of the blood oxygen concentration was successful for more than 60% of the total experimental period (60 min  $\times$  24 h  $\times$  3 days = 4320 min). The blood gas sensors sometimes failed at monitoring, because the tip of the sensor was bent in the blood vessel due to the movement of the sheep. However, we were able to continuously monitor the blood oxygen concentration, using these sensors, for at least 24 hours in both sheep.

The oxygen concentrations in the carotid artery and jugular vein, the blood flow rate of the carotid artery, and the calculated oxygen consumption, and energy expenditure of the sheep's heads are shown in Table 2, and these data are categorized and averaged according to the activity of the animal, such as resting, eating, and ruminating, throughout the 3-day period. The oxygen concentration of the carotid artery and jugular vein were almost constant in sheep 1, and were about 8.0 and 5.2 ml/100 ml, respectively. The oxygen concentration in the carotid artery and jugular vein in sheep 2 were about 8.0 and 5.6 ml/100 ml, respectively, but these values were lower during eating.

The blood flow rates of the carotid artery during resting were 146 and 194 ml/min in sheep 1 and 2, respectively. In both sheep, the blood flow rates during eating and ruminating were 2.3–2.6 and 1.6–1.7 times higher than during resting, respectively. A

typical example of the changes in the blood flow rate of the carotid artery is shown in Fig. 2. The values were smoothed by calculating the 10-point moving average, ranging from about 200 to 800 ml/min. Rapid rises in the blood flow rate associated with eating and ruminating were clearly observed among the changes in the blood flow rate.

The changes in oxygen consumption and the energy expenditure of the head were accompanied with changes in the blood flow rate, because the difference in the blood oxygen concentration between the carotid artery and jugular vein were almost constant. The energy expenditure of the head during eating and ruminating was about 2.5 and 1.6 times higher than during resting, respectively (Table 2).

The amount of time the sheep spent on each behavior, and their oxygen consumption and energy expenditure while exercising that behavior, are shown on a per-day basis in Table 3. The time spent eating was 166 and 136 min in sheep 1 and 2, respec-

tively. Both sheep ruminated for about 13 hours (54% of the day). The oxygen consumption and energy expenditure per day were calculated by multiplying the oxygen consumption (and energy expenditure) per minute in Table 2 by the time spent on each behavior. Both sheep consumed about 75 kJ/day (20% of the total energy consumption in the head) during eating, and about 241 kJ/day (60%) during rumination. The total energy consumption of the head was about 419 kJ/day in both sheep.

Table 4 shows the energy intake and expenditure of the two sheep. The gross energy intakes (MJ/day) were 13.1 and 14.2, and the levels of heat production (MJ/day) were 5.1 and 6.0 for sheep 1 and 2, respectively. The energy balance of sheep 2 was slightly negative. The total energy expenditure of the head, averaged for both sheep, was estimated to be 0.4 MJ (Table 4), indicating that this compartment accounted for about 8 and 6% of the entire body energy expenditure in sheep 1 and 2, respectively.

### Discussion

In this study, the oxygen consumption of the sheep head was calculated by continuously monitoring the blood gas concentration and blood flow rate.

The blood flow was monitored by an ultrasound flow probe. Drost<sup>5)</sup> introduced a blood flow measuring system based on ultrasonic transit time principles. The cuff-type blood flow probe implanted around the vessel requires only acoustic contact with the vessel; thus, a loose fit is acceptable. The ultrasound flow probe has advantages over the other commonly used methods, such as thermodilution, dye-dilution, and the electromagnetic flow meter, in

Table 1. Total measuring times of the blood oxygen concentration of the carotid artery and jugular vein throughout the 3-day period.

Sheep	Items	Time (min)
1	Carotid artery	3771 (87)
	Jugular vein	2456 (57)
2	Carotid artery	2809 (65)
	Jugular vein	3191 (74)

The number in parentheses indicates % of total time (4320 min).

Sheep no. 1, Corriedale; sheep no. 2, Suffolk.

Table 2. Oxygen concentrations in the carotid artery and jugular vein, blood flow rate of the carotid artery, and calculated oxygen consumption and energy expenditure of the sheep's heads during resting, eating and ruminating throughout the 3-day period.

Items	Sheep	Resting	Eating	Ruminating
Oxygen concentration in carotid artery (ml/100 ml)	1	8.0±0.1	8.0±0.1	8.0±0.0
	2	8.0±0.1	8.0±0.0	8.0±0.1
Oxygen concentration in jugular vein (ml/100 ml)	1	5.2±0.3	5.2±0.0	5.1±0.5
	2	5.8±0.7	5.2±0.6	5.7±0.5
Blood flow of the carotid artery (ml/min)	1	146±64	381±95	240±74
	2	194±98	438±173	334±102
Oxygen consumption (ml/min)	1	9.1±3.8	19.7±4.8	14.6±3.9
	2	9.7±6.3	27.2±11.3	14.6±5.8
Energy expenditure (J/min)	1	195±81	420±102	310±83
	2	208±134	582±243	313±123
Energy expenditure (J/kg <sup>0.75</sup> /min)	1	16.0±6.7	34.5±8.4	25.5±6.9
	2	15.8±10.2	44.3±18.5	23.8±9.4

Values are means±S.D.

Sheep no. 1, Corriedale; sheep no. 2, Suffolk.

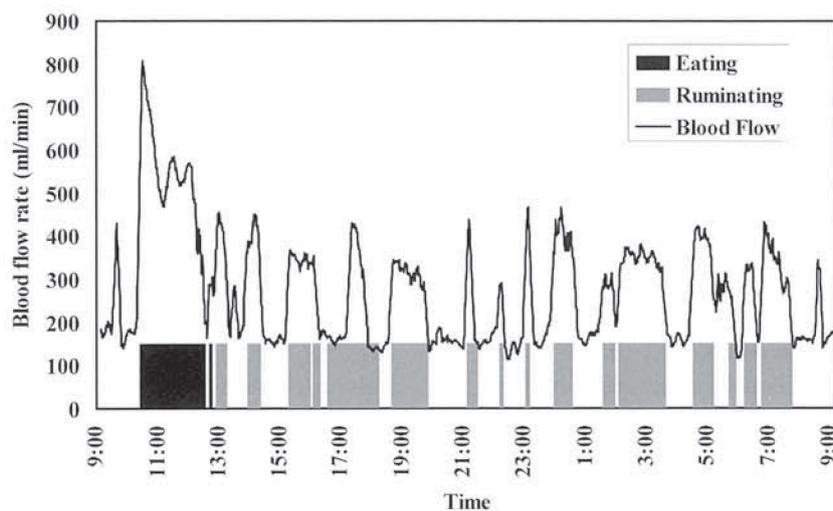


Fig. 2. Changes in the blood flow rate of the carotid artery associated with eating and ruminating in sheep 1. (■) and (■) indicate the time the sheep spent eating and ruminating, respectively.

Table 3. Amount of time the sheep spent resting, eating, and ruminating; and oxygen consumption and energy expenditure of the sheep's heads during resting, eating, and ruminating.

Items	Sheep	Resting	Eating	Ruminating	Total
Times sheep spent (min/day)	1	510	166	765	1440
	2	524	136	780	1440
Oxygen consumption (L/day)	1	4.7	3.3	11.1	19.1
	2	5.1	3.7	11.4	20.2
Energy expenditure (kJ/day)	1	99	70	237	406
	2	109	79	244	432

Sheep no. 1, Corriedale; sheep no. 2, Suffolk.

that it is non-invasive and provides continuous measurements for experiments in which continuous monitoring is required<sup>6)</sup>. It is believed that the transit-time ultrasonic blood flow sensor provides an accurate and highly reliable system for measuring blood flow on a chronic basis. Bednarik and May<sup>1)</sup> have reported that transit-time flow probes reliably and accurately measured the regional blood flow over many months in adult sheep, although the probes could not be used to measure cardiac output.

There are four representative arteries, two carotid arteries and two vertebral arteries, in the neck. The maxillary artery branches from the carotid artery and vertebral artery, providing the cerebral blood flow in ruminants<sup>17)</sup>. It is thought that the contribution to the blood supply from the vertebral artery is low in cattle and sheep<sup>17)</sup>; however, there is scant data describing the blood flow rate of the vertebral artery in sheep. Since we ignored the blood supply from the vertebral artery, the total blood supply to the head might be a low estimate in this study. There have also been few reports describing

Table 4. Energy intake and energy expenditure of the sheep's heads. (MJ/day)

Items	Sheep no.	
	1	2
Gross energy intake	13.1	14.2
Metabolizable energy intake	5.4	5.9
Heat production		
whole body	5.1	6.0
head	0.4 (8)*	0.4 (7)

\*The number in parentheses indicates %HP of the whole body.

Sheep no. 1, Corriedale; sheep no. 2, Suffolk.

the blood flow rate of the carotid artery in conscious adult sheep (discussed below). However, this report clearly shows that the blood flow of the carotid artery increased dramatically during eating and ruminating. It is possible that the increase in blood flow was a result of the increase in the muscle activity required for mastication, salivation, and brain activity. Takahashi and Eda<sup>16)</sup> reported that the post-

prandial increment of cerebral oxygen consumption was only 8%, using *in situ* near-infrared ray spectroscopy in sheep. On the other hand, it has been reported that the saliva flow during eating is 1.5~4 times greater than that during resting<sup>13)</sup>. Based on these studies, it is likely that the increases in muscle activity and salivation mainly contributed to the rise in blood flow.

Blood gas concentrations in the carotid artery and jugular vein were monitored by intravascular blood gas sensors. Continuous monitoring of the arterial blood gas concentration is essential for the optimum management of mechanically ventilated patients in intensive care units. Intermittent blood gas measurement using a blood gas analyzer is the standard method of monitoring the blood gas concentration, and the only routinely available continuous monitor of oxygen saturation is the pulse oximeter. However, the oximeter cannot measure pH and pO<sub>2</sub>. A continuous intra-arterial blood gas monitoring system has been developed by Biomedical Sensors (Paratrend 7, Biomedical Sensors Ltd., High Wycombe, UK). It has been reported that the results obtained from monitoring arterial blood gases using this system showed an acceptable level of clinical accuracy in humans and porcine models<sup>4,18)</sup>. However, the impossibility of measuring the blood hemoglobin concentration is likely to be an important issue with regard to the continuous monitoring of the blood oxygen concentration; in this context, it is of note that the results of our calculations for the oxygen concentration in the carotid artery (Table 2) were lower than the results reported in another study<sup>11)</sup>, and in addition, the co-efficient value of the blood hemoglobin concentration from our laboratory data (not published) revealed approximately 20% day-to-day variations. Thus, there might have been a maximum of 20% variation among the calculated oxygen consumption results of the present study.

However, the oxygen concentrations in the carotid artery and jugular vein were almost constant. The variations found in the jugular vein were caused by the activity of the head, associated with jaw movement. It is known that oxygen and carbon dioxide are classic metabolic regulators of tissue blood flow<sup>14)</sup>. The rise in both oxygen absorption and carbon dioxide release associated with jaw movement caused an increase in blood supply as the heart beat increased.

The calculated energy cost of eating and ruminating in the sheep's heads were 39 and 25 J/min/kg<sup>0.75</sup>, and the increases in energy expenditure compared with those of the resting condition amounted to approximately 23 and 9 J/min/kg<sup>0.75</sup> during eating and ruminating, respectively. There have been few reports focusing on changes in the oxygen consump-

tion of the head of conscious adult sheep. However, the data from this study were comparable to the data that are presently available; for example, it has been reported that the increase in the whole body energy expenditure of sheep during eating and ruminating was about 70<sup>13)</sup> and 50<sup>8)</sup> J/min/kg<sup>0.75</sup>, respectively. These figures also indicate that the data regarding the blood flow rate in this study were within a reasonable range. The differences between the energy expenditure of the head and the whole body during eating and ruminating might primarily be due to the activity of the digestive tract. The contribution of the activity of the digestive tract may be lower during rumination because the reticulo-rumen contraction intervals are longer during rumination than during eating<sup>14)</sup>. Thus, it is thought that the heat increment is mainly affected by an increase in the activity of the digestive tract and of other organs associated with digestion; however, the amount of energy consumed by mastication was not negligible. It is well known that the uptake of certain metabolites in target organs is determined by the blood supply. Oshibe *et al.*<sup>12)</sup> indicated that the rise in blood flow is a consequence of increased organ activity in the mammary glands of lactating ewes. The rise in the blood flow rate of the carotid artery associated with chewing is likely to be a consequence of increased oxygen consumption in related organs.

In conclusion, the oxygen consumption of the head was measured in this study by continuously monitoring the blood flow and the blood oxygen concentration. The oxygen concentrations of the carotid artery and jugular vein were relatively constant, but the blood flow rate of the carotid artery was dramatically increased during eating and ruminating. The amount of energy consumed in the head was of note, and was mainly determined by changes in the blood flow rate associated with jaw movement. The mechanism regulating blood flow to the head will be examined in a future study, and measurement of the blood flow rate of the vertebral artery as well as of the blood hemoglobin concentration will also be performed.

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## ヒツジ頭部における酸素消費量の連続測定ならびにエネルギー消費量の推定

樋口浩二・西田武弘<sup>1)</sup>・永西 修<sup>1)</sup>・Agung PURNOMOADI<sup>2)</sup>・上田宏一郎<sup>3)</sup>・寺田文典<sup>4)</sup>

家畜生理栄養部

<sup>1)</sup> 家畜生産管理部

<sup>2)</sup> ディポネゴロ大学

<sup>3)</sup> 北海道大学

<sup>4)</sup> 農業技術研究機構本部

### 摘 要

動静脈差法による家畜臓器のエネルギー消費量測定系を開発する目的で、2頭のヒツジの頸動脈と頸静脈の酸素濃度差ならびに頸動脈血流量を連続的に測定した。ヒツジにはあらかじめ左側に頸動脈ループの作成、右側頸動脈に超音波血流計センサーの装着をおこない、日本飼養標準に基づいてイタリアンライグラス乾草をTDN維持要求量を満たすように給餌し、7日間の予備試験の後、6日間の本試験をおこなった。本試験前半3日間は、頸動脈ループと頸静脈に血液ガスセンサーを挿入して酸素濃度を10秒間隔で測定し、血流量は1分間の平均値を記録した。同時にビデオカメラでヒツジの採食・反すう行動を観察した。後半3日間は開放式呼吸試験装置で全身の酸素消費量を測定した。その結果、頸動脈ならびに頸静脈酸素濃度は、1日を通じてほぼ一定の値を示した。血中酸素濃度には動物の行動の影響は見られず、見かけの酸素濃度差に変化はなかった。しかし頸動脈血流量は採食・反すう行動に伴って大きく変動し、採食時では安静時に比べて約2.5倍に増加した。単位時間あたりの頭部のエネルギー消費量は、安静時に比べて採食・反すう時に増加し、その増加は反すう時よりも採食時のほうが大きかった。

キーワード：酸素消費量，血流量，咀嚼行動，頭部，ヒツジ