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## Arterio-Venous Differences of Nitrogenous Compounds in Blood Circulating in the Hind Limb of a Goat

### I. Arterio-Venous Differences after an Overnight Fast

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#### Summary

1. The arterio-venous differences(A-V) of the nitrogenous compounds and the other compounds in the blood in the hind limb of the goat after an overnight fast were measured in order to investigate the role of 'tissue nutrient' in the skeletal muscle tissue.

2. The A-V differences in total, protein, non-protein and urea nitrogens, globulin and water were hardly observable in all fractions.

3. The A-V differences of albumin, total free amino-N, and lactic acid were  $-0.12$  mg/dl (V/A was 103.5 per cent),  $-0.17$  mg/dl (V/A was 103.2 per cent) and  $3.9$  mg/dl(V/A was 94.8 per cent) respectively. The higher values in venous blood than in arterial in albumin and total free amino-N suggested that they were released from muscle tissue into the venous blood. The paired difference analysis, however, showed that the above-mentioned differences were all non-significant.

4. The A-V difference of blood sugar was  $3.9$  mg/dl higher in arterial blood (V/A was 94.8 per cent). This significant high value ( $P < 0.005$ ) indicated that blood sugar might be used in the hind limb muscle during circulation.

The foodstuffs ingested by the animal are transformed in the digestive canal by the digestive process and are distributed to the tissues by the blood stream after the retransformation in the liver.

The particular nutrients which were supplied through the arterial blood to the particular tissues are utilized not only for maintenance but also for performance of their functions. In other words, each tissue requires its own 'tissue nutrients' in order to operate its own activities. UmezU, one of the present authors, defined the word 'tissue nutrient' as the substances which are mainly produced in the liver or other organs and which are transported by the blood stream to each tissue for its particular utilization. He mentioned that the investigation on tissue nutrient will throw light on the relationship between the resultant product of digestion and

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the exhibition of tissue function. The approach in this way to the significance of nutrient in the animal could become a new phase of nutritional physiology.

The amount of consumed substance by a tissue is often assumed from the concentration determination of that substance in the venous blood draining the tissue. For instance, the difference of blood sugar concentration in jugular vein between zero time and one hour later is often discussed as the reflection of the sugar utilization in the brain during one hour of circulation. However, it is true with the assumption that the arterial blood concentration of sugar is constant during one hour. In order to know the extent of utilization of the substance in the organ more accurately the arterio-venous difference determination may be helpful. In the present experiment, therefore, the arterio-venous difference is adopted as the most suitable technique to meet our purpose.

The skeletal muscle tissue of the hind limb of the goat was used as the experimental organ because it is active but is a simpler organ than a secretory gland for example.

Knowledge on the utilization and metabolism of carbohydrate or lipid in the tissues is rather well accumulated, but that of the nitrogenous compound is not necessarily adequate. It seems important not only from the aspect of nutritional physiology but also from the aspect of meat production to know the dynamic state of nitrogenous compound in the blood circulating muscle tissue which is the greatest protein depot in the animal body. In the present paper, the arterio-venous differences of nitrogenous compound in the blood circulating in the hind limb of the goat were measured under the conditions of an overnight fast.

### Materials and Methods

*Animals:* Fifteen adult female goats of Saanen breed, weighing 26–43 kg in body weight, were used in the present experiment. Each animal was fed with orchard grass hay and commercial formula feed.

*Treatments:* At least 3 weeks prior to the beginning of the experiments carotid-arterial loop was surgically made up. The polyethylen catheter was inserted into the saphenous vein and its far tip was located at the point where the iliac vein inosculated with the posterior vena cava. The exact location of the tip was not visually confirmed, however, the distance between inserted portion of saphenous vein on the shank and ilio-vena cava junction was assumed to be about 45 cm on the present experimental animals. The inserted catheter was maintained in the body through the experimental period. The catheter was filled with about 2 ml heparin saline solution (1000 units/ml) to prevent blockage inside the catheter from blood coagulation and was flushed with saline solution every second day (Fig. 1).

After an overnight fast the experiments were started at 10 a.m.. The arterial and venous blood samples were simultaneously withdrawn from the carotid-arterial loop and through the polyethylen catheter in the hind limb. The blood samples

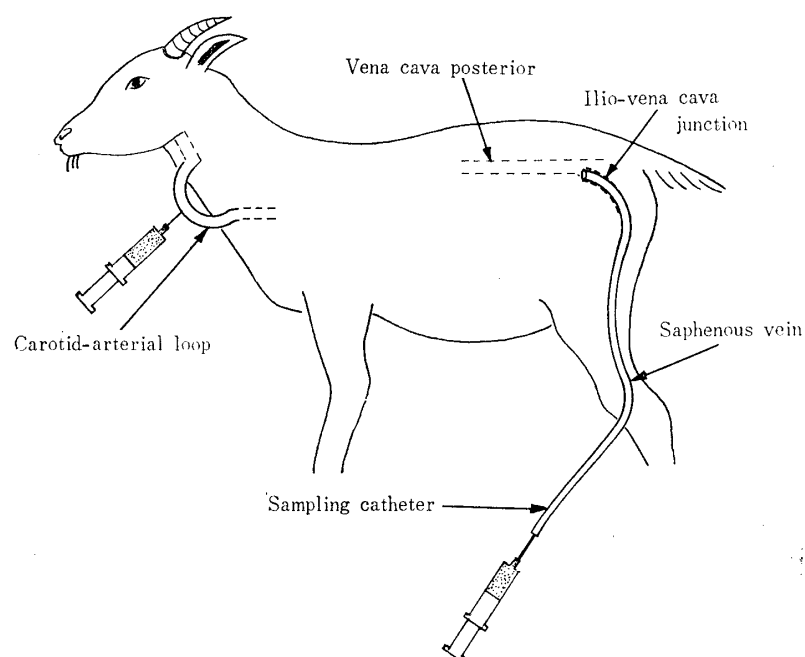


FIG. 1. Schematic diagram of arterial and venous blood sampling.

were immediately examined. In 13 of 15 animals the experiments were repeated two or three times on every third day to observe the quantitative changes of blood constituents.

*Determination of blood constituents:* Following methods were used for the determination of each blood constituent (Table 1).

TABLE 1. *Determination Methods of Blood Constituents*

Blood constituent	Determination method	Blood composition	Reference
Total nitrogen Protein nitrogen Non-protein nitrogen	Micro-Kjeldahl	Plasma	(1)
Protein	Refractometer	Plasma	
Albumin Globulin	Howe and Micro-Kjeldahl	Plasma	(2)
Urea nitrogen	Conway	Plasma	(3)
Total free amino nitrogen	Colorimetry ( $\beta$ -naphtho-quinone)	Plasma	(4)
Water	Gravimetry	Whole blood	
Sugar	Hagedorn-Jensen	Whole blood	(5)
Lactic acid	Barker-Summerson	Whole blood	(6)

### Results and Discussion

The obtained results on the A-V differences of water content and sugar and lactic acid concentrations were shown in Table 2.

The A-V difference of water content was not observed. Therefore, the effect of water content change in the blood on the A-V differences of several substances under investigation may be eliminated from the discussion.

TABLE 2. *A-V Differences of Water Content, Sugar and Lactic Acid Concentration in Blood circulating in the Goat Hind Limb at an Overnight Fast*

Blood constituent	No. tests	No. subjects	Arterial blood (A)	Venous blood (V)	(A)-(V)	V/A (%)
Water (%)	22	11	83.22±1.23	83.25±1.23	-0.03	100.0
Sugar (mg/dl)	27	13	72.8±12.9	69.0±13.8	3.9*	94.8
Lactic acid (mg/dl)	13	7	12.51±6.51	13.14±5.99	-0.63	105.0

\*  $0.001 < P < 0.005$

The blood sugar level was 3.9 mg/dl lower in venous blood than in arterial blood. This figure is highly significant ( $P < 0.005$ ). This fact is undoubtedly not a new finding but has not been adopted to all experimented results. In 2 cases out of 27, higher blood sugar value was obtained in venous blood.

It is reasonable to assume that the higher level of lactic acid in venous than in arterial blood was the reflection of blood sugar utilization in the muscle tissues of the hind limb during circulation. However, the A-V difference of lactic acid is not statistically significant.

It should be noted that the nitrogen concentration difference between arterial and venous blood was not significant (Table 3). It is suggested that the dissimilation and assimilation of nitrogenous substances in skeletal muscle tissues proceeds, as a whole, at the same rate as shown in the present experiment. As to non-protein nitrogen, no significant difference was measured between the arterial and venous blood concentration. However, the arterial concentration of non-protein nitrogen was higher in arterial blood than in venous blood in 5 subjects out of 7.

Total protein concentration was determined by two methods. One is by the use of a hand protein refractometer (Hitachi) and the other was by multiplying the protein nitrogen concentration obtained by Kjeldahl method by 6.25. The data obtained by the former method was expressed as total protein I and the latter was as total protein II in Table 3. In the comparison of the two methods, the data resulting from the hand refractometer was always lower than the Kjeldahl method, though the difference between the two was not large.

The albumin concentrations were higher in venous blood than in the arterial, though they were not statistically significant. This fact may suggest that albumin was released from the muscle tissue or tissue fluid to the venous blood. On the

TABLE 3. A-V Differences of Nitrogen Compounds in Blood circulating in the Goat Hind Limb at an Overnight Fast

Blood constituent	No. tests	No. subjects	Arterial blood (A)	Venous blood (V)	(A)-(V)	V/A (%)
Total nitrogen (mg/dl)	15	7	1196.7±63.6	1198.9±58.5	-2.2	100.2
Protein nitrogen (mg/dl)	14	7	1164.1±61.6	1167.9±55.1	-3.8	100.3
Non-protein nitrogen (mg/dl)	14	7	37.9± 5.9	36.4± 5.9	1.5	96.0
Total protein (%) I	17	9	7.06± 0.57	7.01± 0.64	0.05	99.3
Total protein (g/dl) II	19	11	7.13± 0.51	7.15± 0.48	-0.02	100.3
Albumin (g/dl)	18	10	3.05± 0.59	3.17± 0.75	-0.12	103.5
Globulin (g/dl)	18	10	4.05± 0.49	4.03± 0.59	0.02	99.5
Urea nitrogen (mg/dl)	26	15	21.4± 3.6	20.9± 2.7	0.5	97.6
Total free amino nitrogen (mg/dl)	28	15	5.32± 0.87	5.49±0.98	-0.17	103.2

contrary, no globulin difference was observed between the venous and arterial blood. These results are open for further investigation.

The A-V difference of urea showed a tendency to decrease in the venous blood. Shiga *et al.*\* also found that in a horse the urea concentration was lower in venous blood than in the arterial.

On the free amino acids difference, however, the venous concentration in 20 trials out of 28 was higher than in the arterial ( $P < 0.05$ ). London *et al.*(7) reported that there was a release of amino acid from the human forearm to the venous blood upon resting after an overnight fasting condition. These results seemed to indicate that an overnight fast often caused amino acids release to the venous blood.

On the whole, the A-V differences of nitrogenous compounds in blood circulating in the hind limb of the goat was non-existent or hardly observable. The quantitative changes of those blood constituents from day to day also was hardly observable.

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