Tissue and whole-body oxygen uptake in fed and fasted steers

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The effect of feeding *v.* fasting, on tissue blood flow, oxygen uptake and proportional contributions of the portal drained viscera (PDV), liver (Expts 1 and 2) and hindquarters (HQ; Expt 2) to whole-body O_2 uptake were studied in beef steers. The combined techniques of indirect calorimetry and net tissue flux, the latter being the product of arterio-venous concentration difference and blood flow, were used in the experiments. In response to fasting, whole-body O_2 consumption decreased as did O_2 uptake by all measured tissues except the liver (trend only in Expt 1). Blood flow to all measured tissues decreased during fasting and fractional uptake of O_2 decreased in PDV and increased in liver and HQ (Expt 2). Proportional contribution of specific tissues to whole-body O_2 uptake changed when animals were switched from the fed to the fasted state. The percentage consumed by PDV decreased from 25·4 to 19·9, by liver increased from 20·5 to 26·4 and by HQ was unchanged (9·6 and 10·5) in Expt 2. These significant responses in Expt 2 were observed as trends in Expt 1. The changes in proportional contribution of tissues to whole-animal O_2 uptake reflect the changing metabolic role of specific tissues to lack of food supply. These findings emphasize the central role of the liver in metabolism and indicate that fasting (catabolic) measurements may not reflect the previous fed (anabolic) physiological state.

Blood flow: Fasting: Oxygen uptake: Cattle

Oxygen consumption by a tissue is an index of aerobic energy use by the tissue. O_2 consumption by the portal-drained viscera (PDV) varied directly with level of intake in beef steers (Huntington *et al.* 1988) and lambs (Burrin *et al.* 1989) and decreased in dairy cows in response to fasting (Lomax & Baird, 1983). O_2 consumption by the liver was positively related to level of intake in lambs (Burrin *et al.* 1989) and decreased during fasting in dairy cows (Lomax & Baird, 1983). Similarly, whole-animal O_2 consumption or heat production is directly related to level of intake in cattle (Lobley *et al.* 1987) and lambs (McBride & Milligan, 1985*b*), reflecting the general decline in metabolic rate of body tissues as energy intake declines. Simultaneous determination of tissue and whole-body O_2 consumption allows calculation of the relative contribution of individual tissues to whole-body O_2 use.

Whole-animal O_2 consumption has been determined in fasted animals to assess the effect of treatment on basal energy requirements (e.g. Rumsey *et al.* 1980), thus using a catabolic state to make inference to an anabolic physiological state. Level of energy intake before fasting has been shown to affect fasting heat production (FHP) and parallel changes in weight of several visceral tissues, including the gastrointestinal tract and liver (for review, see Ferrell, 1988). The objectives of these studies were to measure proportional O_2 uptake by the PDV, liver and hindquarters (HQ) of steers and determine whether proportional uptake by individual body tissues varies between fed and fasted steers. In Expt 2 we included measurement of nitrogen flux across splanchnic tissues to quantify additional metabolic changes in response to fasting.

MATERIALS AND METHODS

Animals and diets

Six MARC III (1/4 Hereford, 1/4 Angus, 1/4 Pinzgauer, 1/4 Red Poll) steers were used in Expt 1. They were fed on a high-energy diet (Table 1) at 6 h intervals, beginning at 07.00 hours. The estimated metabolizable energy (ME) and crude protein (N × 6·25; CP) intakes (calculated from measured intake of feed) were 64·6 MJ/d and 955 g/d respectively during measurements in the fed state.

Six Simmental × Hereford steers were used in Expt 2. They were fed on a low-energy diet (Table 1) at 6 h intervals. The estimated ME and CP intakes were 94·4 MJ/d and 1107 g/d respectively during measurements in the fed state. In both experiments the CP concentration of the diet was adjusted so that protein intake would not limit gain at either level of energy intake (National Research Council 1984). All steers were housed in individual stalls throughout the experimental period. Water was available *ad lib.*

Surgery

Steers were fasted for 48 h and water removed 24 h before surgery to install chronic indwelling catheters in the abdominal aorta, hepatic-portal vein, an hepatic vein, branches of the cranial mesenteric vein (Expts 1 and 2), and the caudal vena cava (Expt 2 only). An ultrasonic flow probe (diameter 16 mm; Transonic Systems, Inc., Ithaca, NY) was placed around the abdominal aorta for measurement of blood flow to the HQ (Expt 2). Procedures for catheterization of the splanchnic tissues were similar to those described by Huntington *et al.* (1989). Placement of the ultrasonic flow probe was as described by Eisemann *et al.* (1988) except access was through an incision on the right side of the steer. Insertion of a catheter in the caudal vena cava was accomplished by direct access to the vessel through the visceral cavity.

The experiment began several months after surgery, following completion of another experimental protocol. For Expt 1 all six steers had patent catheters in the hepatic-portal vein and five had patent catheters in the hepatic vein. In the previous protocol two of the steers received clenbuterol (8 mg/animal per d) for a 3 week period ending 5 weeks before initiation of the present protocol. For Expt 2 all six steers had patent catheters in the hepatic-portal and hepatic veins, and four steers had patent catheters in the vena cava. In the previous protocol each steer received injections of bovine somatotropin for 10 d ending 9 weeks before initiation of the present protocol.

Experimental design

Expts 1 and 2 were conducted in identical fashion. The afternoon before measurements each steer was moved to a stall having an associated respiration box for measurement of whole-animal gas exchange. The box measured 0·9 m × 0·9 m × 1·8 m and was fitted with a waterer and feed box so that either fed or fasting measurements could be conducted. A 0·6 m^2 section of the front and each side of the box were covered with 5 mm Plexiglass for visibility. One side also held an access door. The remainder of the box was covered with 3 mm aluminium sheeting. The 0·4 m wide opening for the animal's head was fitted with a reinforced vinyl hood to provide a seal around the animal's neck using a pair of draw strings. There was sufficient amount of material in the hood to allow the animal to lay down. All steers had previous experience in the respiration box. The side door on the respiration box remained open until the start of measurements the following morning at 07.30 hours. From 07.30 to 13.30 hours simultaneous measurements were made of whole-

Table 1. *Composition of the diet for each experiment (g/kg dry matter)*

Ingredients	Expt 1*	Expt 2†
Smooth brome (*Bromus inermis*) chopped hay	—	300·0
Cracked maize	649·9	490·0
Ground lucerne (*Medicago sativa*) hay	122·5	78·6
Soya-bean meal	150·0	84·0
Molasses	50·0	29·9
Urea	9·6	—
$CaCO_3$	12·5	11·9
NaCl	4·9	4·9
Trace mineral premix‡	0·5	0·5
Vitamins A, D, E§	0·2	0·2

* Crude protein (nitrogen × 6·25) 183 g/kg, 12·38 MJ metabolizable energy/kg dry matter.
† Crude protein 132 g/kg, 11·25 MJ metabolizable energy/kg dry matter.
‡ Provided (mg/kg dry matter): iron 50, manganese 40, zinc 60, copper 8, iodine 1, cobalt 0·5.
§ Provided (μg/kg dry matter): vitamin A (retinol) 529, vitamin D 4·4, vitamin E 176.

animal gas exchange and tissue O_2 uptake. All tissue samples were taken while the steers were standing, to ensure uniformity in sample collection, and encompassed a 4 min interval. The steers were allowed to lay down in between samples but did not always do so. When they did lay down, at least 5 min were allowed in between getting an animal up and sampling. Changes in HQ blood flow in response to standing occurred over approximately a 30 s interval. Following measurements each steer was returned to its original stall. A second set of measurements was made 2 weeks later after steers were fasted for 3 d. The 6 h measurement period corresponded to 78–84 h post feeding.

Experimental procedures

Indicator dilution (*p*-aminohippurate, PAH) was used to measure blood flow in PDV and hepatic tissues. On each sample day a primed continuous infusion of PAH was initiated into a branch of the cranial mesenteric vein at least 45 min before the first set of simultaneous blood samples. The infusion rate of PAH varied with the size of the steers and feeding status and was 7200 and 3600 mg/h for fed and fasted steers respectively in Expt 1; and 8640 and 5400 mg/h for fed and fasted steers respectively in Expt 2.

There were three whole-body gas exchange measurements of 2 h duration during the 6 h sampling period. The first measurement began at 07.30 hours, 30 min after the 07.00 hours feeding (in the fed state). Air was circulated within the respiration box and exhausted through a dry test meter. A proportional sample of the exhaust air and a concurrently collected sample of intake air were held in Tedlar bags. The samples were taken to gas analysers for determination of oxygen, carbon dioxide and methane concentrations as outlined in Nienaber & Maddy (1985). The system recovery of O_2 and CO_2 exchange averaged 99 (SE 3.0) and 98 (SE 3·5)% respectively, as measured by combustion of alcohol within the box. Loss of energy as heat was calculated from O_2 consumed and CO_2 produced with correction for CH_4 production (Brouwer, 1965) and extrapolated to 24 h for comparison with literature values reported on that basis.

Within each whole-body measurement period, four sets of blood samples were taken simultaneously from the abdominal aorta, hepatic-portal vein, hepatic vein (Expts 1 and 2) and caudal vena cava (Expt 2 only) at 30-min intervals beginning at 07.45 hours. Two blood samples were taken. The first was withdrawn anaerobically into 3-ml heparinized syringes, capped with a rubber stopper and kept on ice until analysed for O_2 saturation and

haemoglobin (Hb) content using a Hemoximeter (OSM 2; Radiometer Corporation, Copenhagen, Denmark). These analyses were completed within 30 min of sampling. O_2 concentration was calculated by the following equation:

$$O_2\,(\text{mM}) = \frac{(\text{Hb}\,(\text{g/l}) \times 1{\cdot}34\ \text{ml}\,O_2/\text{g Hb} \times \%\,O_2\,\text{saturation}/100)}{22{\cdot}4\ \text{ml}\,O_2/\text{mmol}\,O_2}.$$

A second 5 ml blood sample was taken using a heparinized syringe. Blood from this sample was diluted 1:3 (v/v) with deionized distilled water and refrigerated until analysed for PAH. Blood flow was calculated by the following equation:

$$\text{blood flow (l/h)} = \frac{\text{PAH infusion rate (mg/h)}}{[\text{PAH}]_V - [\text{PAH}]_A},$$

where V is PAH concentration in the hepatic-portal vein (PDV flow) or hepatic vein (hepatic flow) and A is PAH concentration in the artery (background). Hepatic arterial flow is hepatic flow minus PDV flow. For Expt 2 only, ammonia-N, urea-N and α-amino-N were analysed also on the second blood sample. In addition the ultrasonic flow probe was used for measurement of blood flow to the HQ during the blood sampling interval. Blood flow was recorded every 10 s over the blood sampling interval and an average flow was then calculated. All analyses were completed on the day of sampling by automated procedures (Technicon Industrial Systems, Tarrytown, NY) as follows: PAH (Technicon Industrial Systems, 1972), ammonia-N by a hypochlorite method (Technicon Industrial Systems, 1974), urea-N by a diacetylmonoxime method (Technicon Industrial Systems, 1977) and α-amino-N by the method of Broderick & Kang (1980) modified to include sample dialysis. Under these conditions *N*-acetyl-PAH is not detected across the hepatic bed of cattle.

For tissue O_2 and metabolite measurements, a mean value was calculated for each four samples corresponding to a whole-body measurement interval (2 h). The resulting three means were then averaged to obtain an overall mean estimate for each steer and sample day. Net tissue uptake of O_2 and net tissue uptake or release of other metabolites was calculated as the product of arterio-venous (A-V) concentration difference and blood flow. Fractional uptake (uptake relative to tissue supply) of O_2 for the PDV and HQ was calculated as (A-V)/A where V is the O_2 concentration in the portal vein or vena cava respectively. Fractional uptake of O_2 by the liver was calculated as:

$$\frac{\text{hepatic } O_2 \text{ uptake}}{((\text{HA flow} \times [O_2]_{HA}) + (\text{PDV flow} \times [O_2]_P))},$$

where HA is hepatic artery and P is portal vein.

Statistical analysis

Statistical analysis of all variables was performed using a Student's paired *t* test to compare values obtained during the fed and fasted state (Snedecor & Cochran, 1967).

RESULTS

Whole-body measures

Whole-animal responses were similar in Expts 1 and 2 (Table 2). Uptake of O_2 and production of CO_2 and CH_4 were lower ($P < 0{\cdot}001$) in fasted than in fed steers. The

Table 2. *Whole-body measurements in fed and fasted steers**

Variable	Fed	Fasted	SED	Statistical significance of difference: $P <$
Expt 1				
BW (kg)	339	333	2	0·05
Gas exchanges (l/2 h):				
Oxygen	197·2	131·0	7·2	0·001
Carbon dioxide	198·8	96·1	7·0	0·001
Methane	8·2	1·0	0·5	0·001
RQ	1·01	0·73	0·008	0·001
Heat production†:				
kJ/d per kg BW	147·3	93·7	4·2	0·001
kJ/d per kg $BW^{0·75}$	632·6	400·8	19·7	0·001
Expt 2				
BW (kg)	426	416	2	0·01
Heart rate (beats/min)	81	51	1	0·001
Gas exchanges (l/2 h):				
Oxygen	242·8	153·8	2·3	0·001
Carbon dioxide	248·8	110·8	2·6	0·001
Methane	18·2	0·9	0·6	0·001
RQ	1·02	0·72	0·005	0·001
Heat production†:				
kJ/d per kg BW	144·8	87·9	1·2	0·001
kJ/d per kg $BW^{0·75}$	658·1	396·6	5·4	0·001

SED, standard error of the difference between means; BW, body-weight; RQ, respiratory quotient.

* For details of procedures, see p. 401.

† Calculated from O_2 consumed and CO_2 produced with correction for CH_4 production (Brouwer, 1965). Extrapolated to 24 h.

respiratory quotient (RQ) decreased ($P < 0·001$) as well, indicating a change in oxidative substrate. CH_4 production and the RQ together suggested attainment of the fasting state. Loss of energy as heat, expressed relative to body weight (BW) or $BW^{0·75}$ decreased ($P < 0·001$) with fasting.

For Expt 1, O_2 uptake did not differ across whole-body periods and was 195·6, 195·8 and 199·5 litres/h during fed measurements compared with 131·8, 128·4 and 132·6 litres/h during fasting (SEM 3·0). For Expt 2, O_2 uptake did vary across whole-body periods and was 254·4, 241·0 and 233·1 litres/h during fed measurements compared with 158·7, 153·1 and 149·6 litres/h during fasting (SEM 2·9).

Tissue blood flow and O_2

In Expt 1, arterial O_2 concentration and arterio-portal concentration differences were not changed with fasting (Table 3). Both arterio-hepatic ($P < 0·03$) and portal-hepatic ($P < 0·06$) O_2 differences increased with fasting, while blood flow in both the PDV ($P < 0·004$) and liver ($P < 0·02$) decreased. The percentage of liver blood flow derived from the PDV was unchanged with treatment, although it tended to be lower during fasting (85 *v.* 79% of total flow).

In Expt 2, arterial O_2 concentration ($P < 0·001$) and arterio-hepatic ($P < 0·07$), portal-hepatic ($P < 0·02$) and arterio-venous ($P < 0·02$) concentration differences increased with fasting. O_2 concentration difference across the PDV was unchanged. Blood flow decreased ($P < 0·001$) in all tissue beds measured as did the contribution of PDV blood flow to liver blood flow (91 *v.* 79%, $P < 0·01$).

Table 3. *Oxygen concentrations (mM), arterio-venous concentration differences (mM), and blood flow (l/h) in fed and fasted steers**

Variable	Fed	Fasted	SED	Statistical significance of difference: $P <$
Expt 1				
Arterial O_2	6·59	6·80	0·16	NS
O_2 concentration difference:				
A-P	1·45	1·55	0·07	NS
A-H	2·59	2·79	0·04	0·03
P-H	1·09	1·28	0·05	0·06
Blood flow				
PDV	611	336	38	0·004
Liver	683	454	44	0·02
PDV/liver	0·85	0·79	0·07	NS
Expt 2				
Arterial O_2	6·74	7·26	0·06	0·001
O_2 concentration difference:				
A-P	1·80	1·76	0·03	NS
A-H	2·96	3·20	0·07	0·07
P-H	1·16	1·44	0·06	0·02
A-V	2·72	3·45	0·12	0·02
Blood flow				
PDV	768	394	5	0·001
Liver	849	508	20	0·001
PDV/liver	0·91	0·79	0·02	0·01
HQ	210	127	7	0·001

SED, standard error of the difference between means; A, artery; P, portal vein; H, hepatic vein; V, caudal vena cava; PDV, portal-drained viscera; HQ, hindquarters; NS, not significant.

* For details of procedures, see pp. 401–402.

In Expt 1, O_2 uptake decreased in PDV ($P < 0·004$) and total splanchnic tissues ($P < 0·05$) in response to fasting (Table 4). There was an 18% decrease in liver O_2 uptake with fasting, although this was not significant. Fractional uptake of O_2 was unchanged in the tissues measured, although it tended to be higher in the liver. In Expt 2, O_2 uptake decreased ($P < 0·001$ except liver, $P < 0·06$) in all tissues with fasting. Fractional uptake decreased in PDV ($P < 0·01$) and increased ($P < 0·05$) in liver and HQ tissues. The increase in uptake by the liver in Expt 2 was similar to the trend in Expt 1.

Values for tissues and the whole animal were combined to calculate proportional contribution of tissues to whole-animal O_2 uptake (Table 5). In Expt 2 the proportional contribution of PDV tissues decreased ($P < 0·001$) and of liver increased ($P < 0·02$) with respect to whole-animal O_2 consumption when steers were fasted. Similar trends were apparent in Expt 1.

Net flux of nitrogenous compounds

Fasting resulted in increased arterial concentration of urea-N ($P < 0·05$) and α-amino-N ($P < 0·01$) but not ammonia-N (Table 6). Net flux of these compounds across PDV tissue changed such that release of ammonia-N and α-amino-N by the PDV decreased ($P < 0·001$) and transfer of urea-N to the PDV decreased ($P < 0·07$) with fasting. Net flux across the liver was unchanged by fasting except for uptake of ammonia which decreased ($P < 0·003$).

Table 4. *Oxygen uptake (mmol/h) and fractional uptake in tissues of fed and fasted steers**

Variable	Fed	Fasted	SED	Statistical significance of difference: $P <$
Expt 1				
O_2 uptake†				
PDV	876	520	48	0·004
Liver	900	742	141	NS
Splanchnic	1765	1269	130	0·05
Fractional uptake:				
PDV‡	21·9	22·8	0·8	NS
Liver§	24·7	29·2	2·6	NS
Splanchnic‡	39·2	42·0	1·1	NS
Expt 2				
O_2 uptake†				
PDV	1377	684	21	0·001
Liver	1114	911	57	0·06
Splanchnic	2486	1594	70	0·001
HQ	518	357	7	0·001
Fractional uptake:				
PDV‡	26·7	24·2	0·4	0·01
Liver§	25·9	30·7	1·1	0·03
Splanchnic‡	43·9	44·0	1·0	NS
HQ‡	41·0	46·0	1·0	0·04

PDV, portal-drained viscera; HQ, hindquarters; NS, not significant.

* For details of procedures, see pp. 401–402.

† Uptake (mmol/h) = arterio-venous concentration difference (mM) × blood flow (l/h).

‡ Fractional uptake = $[O_2]_{A-X}/[O_2]_A \times 100$ where A is arterial O_2 concentration, X is portal vein O_2 concentration for PDV, hepatic vein O_2 concentration for splanchnic, and vena cava O_2 concentration for HQ.

§ Fractional uptake = liver O_2 uptake/((HA flow × $[O_2]_{HA}$) + (PDV flow × $[O_2]_P$)) × 100 where HA is hepatic artery and P is portal vein.

Table 5. *Tissue oxygen uptake as a percentage of whole-body (WB) oxygen uptake expressed on an absolute and incremental basis in fed and fasted steers*†

Variable	Absolute*		SED	Statistical significance of difference: $P <$	Incremental*	
	Fed	Fasted			Mean	SEM
Expt 1						
PDV	20·0	17·8	1·2	NS	27·2	6·7
Liver	20·7	25·2	4·1	NS	8·1	17·5
Splanchnic	40·8	43·3	3·1	NS	36·2	13·2
Expt 2						
PDV	25·4	19·9	0·3	0·001	34·4	1·4
Liver	20·5	26·4	1·3	0·02	9·5	3·7
Splanchnic	45·8	46·4	1·4	NS	43·7	3·6
HQ	9·6	10·5	0·4	NS	8·0	0·3

SED, standard error of the difference between means; SEM, standard error of the mean; PDV, portal-drained viscera; HQ, hindquarters; NS, not significant.

* Absolute basis = (tissue/WB) × 100; Incremental = $((\text{tissue}_{fed} - \text{tissue}_{fasted})/(\text{WB}_{fed} - \text{WB}_{fasted})) \times 100$.

† For details of procedures, see pp. 401–402.

Table 6. *Expt2. Arterial concentration (mM) and net flux (mmol/h) of nitrogenous variables in fed and fasted steers**

Variable	Fed	Fasted	SED	Statistical significance of difference: $P <$
Concentration				
Ammonia	0·374	0·409	0·016	NS
Urea-nitrogen	5·83	7·19	0·36	0·05
α-Amino-N	3·28	3·50	0·04	0·01
Net flux†				
PDV				
Ammonia	−185	−78	11	0·001
Urea-N	123	39	25	0·07
α-Amino-N	−119	4	6	0·001
Liver				
Ammonia	190	100	12	0·003
Urea-N	−238	−183	23	NS
α-Amino-N	61	87	11	NS

SED, standard error of the difference between means; PDV, portal-drained viscera; NS, not significant.

* For details of procedures, see pp. 401–402.

† Net flux (mmol/h) = arterio-venous concentration difference (mM) × blood flow (l/h). Positive number indicates net uptake, a negative number indicates net release.

DISCUSSION

The values for whole-animal gas exchange or FHP are similar in magnitude to those previously reported for fasted cattle by Rumsey *et al.* (1980), and on the low end of the range reported by Webster *et al.* (1974). They are, however, higher than equations derived by the Agricultural Research Council (1980) which estimate FHP as a function of $BW^{0·67}$ and would equate to 26 MJ/d for Expt 1 and 30·1 MJ/d for Expt 2. The values for heat production in fed and fasted steers in the present study compare well with those of Lobley *et al.* (1987). On average they observed heat production of 65·9, 52·4 and 39·6 MJ/d for steers fed at 1·6 M, 1·0 M or fasted respectively (where M is the ME calculated for zero energy retention). Their steers were slightly heavier than those of Expt 2. The level of energy intake before fasting has been shown to affect FHP (for review, see Ferrell, 1988); however, the steers of Lobley *et al.* (1987) were fed on a maintenance ration for 3 weeks before fasting.

In concert with observed changes in heart rate (Table 1 and Rumsey *et al.* 1980) and trends in cardiac output (Huntington *et al.* 1990) in fed *v.* fasted steers, blood flow to all tissue beds declined with fasting. The decrease was greatest for the PDV. There was also a shift in the proportion of liver blood derived from PDV tissues and the hepatic artery (significant in Expt 2, trend in Expt 1) such that the hepatic arterial contribution increased with fasting. Lomax & Baird (1983) observed a greater decline in portal than hepatic blood flow in non-lactating dairy cows in response to fasting. Hepatic arterial flow was unchanged by fasting in the study of Lomax & Baird (1983). Along with the decrease in blood flow, arterio-venous concentration difference of O_2 increased across all tissue beds except the PDV. The increase in O_2 concentration difference across tissues other than PDV reflects the greater decrease in blood flow than O_2 uptake across these tissues with fasting. In general, Burrin *et al.* (1989) observed changes in blood flow but not arterio-venous concentration difference in lambs fed at maintenance or *ad lib.*

Regression of O_2 uptake *v.* O_2 supply across both experiments (Fig. 1) showed a strong positive relationship ($P < 0·01$) between these variables in all three vascular beds for fed and fasted steers. These observations are similar to those of Edelstone & Holzman (1981)

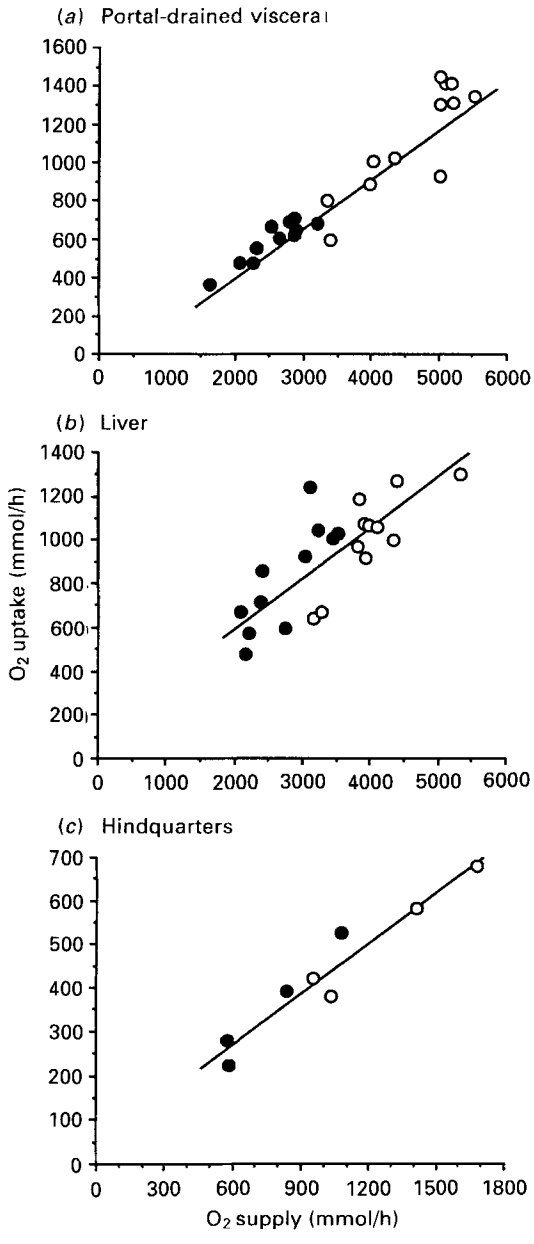

Fig. 1. Relation between oxygen uptake and O_2 supply in (*a*) portal-drained viscera $y = 0{\cdot}271x - 105{\cdot}5$, R^2 0·90, SE 0·019, residual standard deviation (RSD) 110·00, (*b*) liver $y = 0{\cdot}225x + 167{\cdot}5$, R^2 0·59, SE 0·040, RSD 154·7; and (*c*) hindquarters $y = 0{\cdot}387x + 44{\cdot}5$, R^2 0·92, SE 0·043, RSD 43·3 tissues in fed (○) and fasted (●) steers. Values were pooled across Expts 1 and 2. For details of procedures, see pp. 401–402.

for PDV of newborn lambs but differ in that they observed no relation between O_2 uptake by the liver and either blood flow or O_2 supply. In addition, Edelstone & Holzman (1981) observed no relationship between O_2 supply and extraction in the gut of newborn lambs that were fed or fasted but a negative relationship between supply and extraction in the

liver. Across both experiments O_2 extraction by the PDV, liver or HQ did not vary with blood flow or O_2 supply ($P > 0{\cdot}10$, values not shown). In general, across the tissues studied, extraction was higher in the HQ (38·7%) than the PDV (27·1%) or liver (22·5%), suggesting an inverse relationship between extraction and supply (or flow). O_2 extraction by the HQ decreased in steers, concomitant with increased blood flow and increased O_2 uptake in response to clenbuterol (Eisemann *et al.* 1988). The observation that changes in the PDV differ from those of the other tissues suggests interrelationships among supply, extraction and metabolic need.

Comparison of whole-body O_2 consumption in fed *v.* fasted cattle in these two experiments indicates an increase of 1·5- to 1·6-fold in the fed state. The tissue bed showing the largest change was the PDV, which increased from 1·7- to 2-fold (Expts 1 and 2, respectively), whereas the smallest change was in liver O_2 use which increased 1·2-fold. Changes in HQ tissues reflected the average of the whole body. The increase in PDV accounted for 27–34% and in splanchnic tissues accounted for 36–44% (Expts 1 and 2, respectively; Table 5) of the increase for the whole body. The variation in tissue change relative to the whole body is reflected in the proportional calculations (Table 5) where trends were evident in Expt 1 and significant changes were observed in Expt 2. The major difference between the two experiments is in the PDV which may relate to the higher level of feeding in Expt 2 or the differences between diets.

Lomax & Baird (1983) observed a decline in O_2 uptake of both PDV and hepatic tissues in fasted dairy cattle that was of similar magnitude in the two tissues. A positive relation between ME intake and O_2 uptake by the PDV (Webster *et al.* 1975; Burrin *et al.* 1989) and liver (Burrin *et al.* 1989) in lambs and PDV of steers (Huntington *et al.* 1988) have been reported. McBride & Milligan (1985*b*) observed no change in total O_2 uptake of duodenal mucosa per unit dry weight (measured in vitro) in response to level of feeding, including a 48 h fast, in sheep. They (McBride & Milligan, 1985*a*) also observed no change in O_2 uptake by hepatocytes per unit dry weight from sheep fed either at maintenance or fasted for 5 d. Similarly, Burrin *et al.* (1988), comparing fed and starved rats, showed no change in O_2 consumption of hepatocytes per unit weight. As reviewed by Ferrell (1988), considerable information exists to demonstrate a positive relationship between level of nutrition and visceral organ mass. These findings imply that changes in O_2 consumption with level of feeding relate more to changes in tissue mass than metabolic activity. These approaches in vitro, however, may not reflect the situation in vivo.

Energy-requiring processes in body tissues include macromolecular turnover, transport and substrate cycling (for review, see Summers *et al.* 1988). Webster (1980) discussed some of the processes occurring in the gut that contribute to O_2 use by these tissues. He calculated that protein synthesis could account for 50% of the aerobic O_2 use by gut tissues. McNurlan *et al.* (1979) showed both a decrease in fractional rate and absolute rate of protein synthesis in the jejunal mucosa and liver of starved rats. In cattle (Lobley *et al.* 1987), whole-body protein synthesis declined with decreasing level of intake including fasting; however, heat production attributed to protein synthesis relative to total heat production did not change with level of intake. Protein oxidation declined with level of intake above and including maintenance and increased with fasting. Thus, amino acid oxidation is one process that increases with fasting. The whole-body measurements of Lobley *et al.* (1987) may hide important differential effects in specific body tissues that explain the proportional changes seen in response to fasting in Expt 2 and the trends in Expt 1.

O_2 consumption measures in hepatocytes (McBride & Milligan, 1985*a*) and duodenal mucosa (McBride & Milligan, 1985*b*) showed that ouabain-sensitive O_2 consumption was a greater proportion of total O_2 consumption in fed than fasted sheep, demonstrating a

shift in energy expenditure with fasting. These findings give no indication of other metabolic processes, independent of Na^+, K^+-ATPase, that show a proportional increase with fasting to account for the similarity in total O_2 consumption per unit weight.

The information on N flux across PDV and liver tissues gives some rationale for the observations on O_2 use across these tissues. Net flux of α-amino-N across the PDV was not different from zero in fasted steers and yet uptake of α-amino-N by the liver was unchanged, indicating a change in origin of α-amino-N from the gut to the periphery. Arterial supply of α-amino-N to the liver increased from 266 to 399 mmol/h during fasting. Heitmann & Bergman (1980) in sheep and Lomax & Baird (1983) in dairy cattle showed little change in amino acid uptake by hepatic tissues in fasted animals despite the cessation of absorption from the PDV. Amino acids may be used for synthesis of liver and plasma proteins, they may be completely oxidized or the carbon skeletons used for gluconeogenesis. On a proportional basis, N from uptake of α-amino-N by the liver could contribute 0·26 to urea-N output during the fed state, increasing to 0·48 during fasting, while proportional contribution of NH_3-N uptake to urea-N output decreased from 0·80 in the fed state to 0·55 during fasting. In part this reflects a decrease in NH_3-N uptake without a similar decrease in urea-N output during fasting as well as a trend for higher uptake of α-amino-N during fasting. These trends are consistent with increased amino acid oxidation. Similarly transfer of urea to the PDV decreased from 0·52 of hepatic output in the fed state to 0·21 during fasting. On a net N basis for each mmol urea-N transferred to the gut, 2·0 mmol NH_3-N were absorbed during fasting and only 1·5 during the fed state, suggesting greater microbial use of the recycled N in the fed state. Findings of Bergman & Wolff (1971) showed that net absorption of propionate by the PDV ceases in sheep fasted for 3 d thereby increasing the quantitative significance of amino acids for gluconeogenesis, although glucose output by the liver declines with fasting as well (Bergman *et al.* 1974). Similar observations were reported in cows (Lomax & Baird, 1983). Therefore, while digestive and absorptive functions decline in PDV, diverse metabolic functions continue in the liver.

The trend toward a decrease in hepatic urea output was similar to the decline in O_2 use by the liver during fasting, resulting in a similar proportional contribution of urea production to energy use by the liver. Using values of 4 ATP per mol urea synthesized and 6 ATP per mol O_2 consumed, approximately 7·1 and 6·7 % of the O_2 used by the liver could be attributed to urea production for fed and fasted steers respectively. This implies that urea synthesis does not make a major contribution to liver energy expenditure. These estimates do not account for a decrease in ATP produced per mol O_2 consumed in general or in response to fasting. Using the previous assumptions, Huntington & Reynolds (1987) suggested a value of approximately 8·5 % (the original value reported was for urea-N, assuming 4·8 ATP per mol urea and 5 ATP per mol O_2) in lactating cows, and Summers *et al.* (1988) calculated 8–12 % based on findings of Rabkin & Blum (1985) from rat hepatocytes.

The proportional contribution of tissue O_2 uptake to whole-body O_2 uptake was similar in the two experiments except for slightly higher values in Expt 2 for the PDV (20 *v.* 25 %). The diet in Expt 2 was higher in forage than that of Expt 1 and fed at a slightly higher level. Either of these could alter proportional contribution of the PDV to whole-animal energy use. The value for contribution of the PDV was similar to the value of 18 % reported for dairy cows (Huntington & Tyrrell, 1985) and 20 % for lambs (Webster, 1980). Huntington *et al.* (1988) reported values from 23 to 29 % in beef steers. Values for percentage contribution of liver, 20–26 %, were slightly lower than values of 24–31 % in beef steers reported by Huntington *et al.* (1990) and higher than those of Thompson & Bell (1976) who reported 16–18 % in lambs. It is noteworthy that the total contribution of splanchnic tissues to whole-body O_2 use remained constant with fasting despite the change in the individual

components. The high contribution of these tissues to O_2 use in the body relative to their contribution to BW was previously discussed (for review, see Huntington & Reynolds, 1987). In contrast, tissues of the HQ contributed only 10% to O_2 use. In steers of 350 kg live weight, HQ tissues represented approximately 28% of empty BW (J. H. Eisemann, G. B. Huntington and D. R. Catherman, unpublished results). Therefore, while the PDV overcontributes and the HQ undercontributes, each by a factor of 2–3, the liver makes the most disproportionate contribution to O_2 use.

In conclusion, the findings presented here quantify blood flow and O_2 use in body tissues of fed and fasted cattle. Blood flow to and O_2 uptake by all tissues measured decreased as did O_2 uptake by the whole body during fasting. Proportional contribution of O_2 use by major tissues varied between fed and fasted states (Expt 2) such that the contribution of PDV tissues decreased and of liver tissue increased during fasting. Therefore, food supply altered the partition of energy use as well as the total. Because proportional contribution of tissues may change with fasting, relationships observed in the fasting state may not reflect those present in the normal anabolic situation. Similarly, proportional relationships may vary with level of anabolism, and, thus, should not be extrapolated beyond conditions under which they are measured.

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REFERENCES

Agricultural Research Council (1980). *The Nutrient Requirements of Ruminant Livestock*. Slough: Commonwealth Agricultural Bureaux.

Bergman, E. N., Brockman, R. P. & Kaufman, C. F. (1974). Glucose metabolism in ruminants: comparison of whole-body turnover with production by gut, liver and kidneys. *Federation Proceedings* **33**, 1849–1854.

Bergman, E. N. & Wolff, J. E. (1971). Metabolism of volatile fatty acids by liver and portal-drained viscera in sheep. *American Journal of Physiology* **221**, 586–592.

Broderick, G. A. & Kang, J. H. (1980). Automated simultaneous determination of ammonia and total amino acids in ruminal fluid and in vitro media. *Journal of Dairy Science* **63**, 64–75.

Brouwer, E. (1965). Report of subcommittee on constants and factors in energy metabolism. In *Energy Metabolism. Proceedings of the 3rd Symposium on Energy Metabolism, European Association of Animal Production Publication* no. 11, pp. 441–443 [K. L. Blaxter, editor]. New York: Academic Press.

Burrin, D. G., Britton, R. A. & Ferrell, C. L. (1988). Size and metabolic activity of visceral organs in fed and fasted rats. *Journal of Nutrition* **118**, 1547–1552.

Burrin, D. G., Ferrell, C. L., Eisemann, J. H., Britton, R. A. & Nienaber, J. A. (1989). Effect of plane of nutrition on splanchnic blood flow and oxygen consumption in sheep. *British Journal of Nutrition* **62**, 23–34.

Edelstone, D. I. & Holzman, I. R. (1981). Oxygen consumption by the gastrointestinal tract and liver in conscious newborn lambs. *American Journal of Physiology* **240**, G297–G304.

Eisemann, J. H., Huntington, G. B. & Ferrell, C. L. (1988). Effects of dietary clenbuterol on metabolism of the hindquarters in steers. *Journal of Animal Science* **66**, 342–353.

Ferrell, C. L. (1988). Contribution of visceral organs to animal energy expenditures. Current concepts of animal growth. IV. *Journal of Animal Science* **66**, Suppl. 3, 23–34.

Heitmann, R. N. & Bergman, E. N. (1980). Integration of amino acid metabolism in sheep: effects of fasting and acidosis. *American Journal of Physiology* **239**, E248–E254.

Huntington, G. B., Eisemann, J. H. & Whitt, J. M. (1990). Portal blood flow in beef steers – comparison of techniques and relation to hepatic blood flow, cardiac output and oxygen uptake. *Journal of Animal Science* **68**, 1666–1673.

Huntington, G. B. & Reynolds, C. K. (1987). Oxygen consumption and metabolite flux of bovine portal-drained viscera. *Journal of Nutrition* **117**, 1167–1173.

Huntington, G. B., Reynolds, C. K. & Stroud, B. H. (1989). Techniques for measuring blood flow in splanchnic tissues of cattle. *Journal of Dairy Science* **72**, 1583–1595.

Huntington, G. B. & Tyrrell, H. F. (1985). Oxygen consumption by portal-drained viscera of cattle: comparison of analytical methods and relationship to whole body oxygen consumption. *Journal of Dairy Science* **68**, 2727–2731.

Huntington, G. B., Varga, G. A, Glenn, B. P. & Waldo, D. R. (1988). Net absorption and oxygen consumption by Holstein steers fed alfalfa or orchardgrass silage at two equalized intakes. *Journal of Animal Science* **66**, 1292–1302.

Lobley, G. E., Connell, A. & Buchan, V. (1987). Effect of food intake on protein and energy metabolism in finishing beef steers. *British Journal of Nutrition* **57**, 457–465.

Lomax, M. A. & Baird, G. D. (1983). Blood flow and nutrient exchange across the liver and gut of the dairy cow. Effects of lactation and fasting. *British Journal of Nutrition* **49**, 481–496.

McBride, B. W. & Milligan, L. P. (1985*a*). Magnitude of ouabain-sensitive respiration in the liver of growing, lactating and starved sheep. *British Journal of Nutrition* **54**, 293–303.

McBride, B. W. & Milligan, L. P. (1985*b*). Influence of feed intake and starvation on the magnitude of Na^+,K^+-ATPase (*EC* 3.6.1.3)-dependent respiration in duodenal mucosa of sheep. *British Journal of Nutrition* **53**, 605–614.

McNurlan, M. A., Tomkins, A. M. & Garlick, P. J. (1979). The effect of starvation on the rate of protein synthesis in rat liver and small intestine. *Biochemical Journal* **178**, 373–379.

National Research Council (1984). *Nutrient Requirements of Beef Cattle*, 6th rev. ed. Washington, DC: National Academy Press.

Nienaber, J. A. & Maddy, A. L. (1985). Temperature controlled multiple chamber indirect calorimeter – design and operation. *American Society of Agricultural Engineers* **28**, 555–560.

Rabkin, M. & Blum, J. J. (1985). Quantitative analysis of intermediary metabolism in hepatocytes incubated in the presence and absence of glucagon with a substrate mixture containing glucose, ribose, fructose, alanine and acetate. *Biochemical Journal* **255**, 761–786.

Rumsey, T. S., Tyrrell, H. F. & Moe, P. W. (1980). Effect of diethylstilbestrol and Synovex-S on fasting metabolism measurements of beef steers. *Journal of Animal Science* **50**, 160–166.

Snedecor, G. W. & Cochran, W. G. (1967). *Statistical Methods*. Ames, IA: Iowa State University Press.

Summers, M., McBride, B. W. & Milligan, L. P. (1988). Components of basal energy expenditure. In *Aspects of Digestive Physiology in Ruminants*, pp. 257–285 [A. Dobson and M. J. Dobson, editors]. Ithaca, NY: Comstock Publishing Associates.

Technicon Industrial Systems (1972). Para-*amino-hippuric acid, Technicon Industrial Method no. 216-72T*. Tarrytown, NY: Technicon Industrial Systems.

Technicon Industrial Systems (1974). *Ammonia, Technicon Industrial Method no. 337-74T*. Tarrytown, NY: Technicon Industrial Systems.

Technicon Industrial Systems (1977). *Urea nitrogen, Technicon Industrial Method no. 339-01*. Tarrytown, NY: Technicon Industrial Systems.

Thompson, G. E. & Bell, A. W. (1976). The energy metabolism of the liver measured in vivo during cold exposure of sheep. In *Energy Metabolism of Farm Animals. European Association of Animal Production Publication* no. 19, pp. 37–40 [M. Vermorel, editor]. Clermont-Ferrand, France: G. de Bussac.

Webster, A. J. F. (1980). Energy cost of digestion and metabolism in the gut. In *Digestive Physiology and Metabolism in Ruminants*, pp. 469–484 [Y. Ruckebusch and P. Thivend, editors]. Westport, CT: AVI Publishing Company, Inc.

Webster, A. J. F., Brockway, J. M. & Smith, J. S. (1974). Prediction of the energy requirements for growth in beef cattle. 1. The irrelevance of fasting metabolism. *Animal Production* **19**, 127–139.

Webster, A. J. F., Osuji, P. O., White, F. & Ingram, J. F. (1975). The influence of food intake on portal blood flow and heat production in the digestive tract of sheep. *British Journal of Nutrition* **34**, 125–139.

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