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Escape from the chamber: alternative methods for large animal calorimetry

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Over the last few decades measurement of the metabolic rates of farm animals has been made using large, static respiration chambers (e.g., Kleiber, 1958; Flatt, van Soest, Sykes & Moore, 1958; Wainman & Blaxter, 1958), in marked contrast to the portable devices used in studies on human beings (Kofranyi & Michaelis, 1940; Wolff, 1958). The reasons for this divergence of apparatus lie mainly in the different questions which animal and human physiologists and nutritionists have been seeking to answer, and to a lesser degree in the constraints imposed by the behavioural reaction to apparatus by their respective subjects. To an extent the pendulum swings are now reversing and we find more human experimentation taking place in chambers (M. J. Dauncey, unpublished results; Garrow, Murgatroyd, Toft & Warwick, 1977; Jéquier, Pittet & Gygax, 1977), while the large animal physiologists are looking for techniques which will allow them to study the energy expenditure of animals living out-of-doors. One other group of workers concerned with measurement of energy exchange is the ecologists: their interest is comparatively recent and their discipline dictates that they adopt non-chamber techniques. Methods available to the ecologist have been reviewed by Gessaman (1973).

Non-chamber techniques with large animals

Any method of estimating energy expenditure in free-ranging animals must meet certain requirement before it can be adopted for general use. The method must work equally well independently of the nature of the physiological stimulus to alteration of metabolic rate, e.g. muscular activity, climatic stress or level of food intake. The accuracy of the technique should be such that energy expenditure can be estimated to within, say, ±10%: this value may not be good enough for some purposes, but anything worse is probably of little value. Energy expenditure should be measurable over periods of 1 h or less, rather than over the whole day. The method should involve minimal surgical interference, but where this is necessary, it is desirable that success rate and preparation survival times are high, and that after recovery the animal is not hampered in any way. The apparatus to be carried by the animal should be small, of light weight, robust and cheap: the first three factors are necessary if the animal is to behave normally and low cost is desirable as it is almost certain that damage to the apparatus will occur. The apparatus should be such that it can function for at least 24 h without attention from the experimenter. This probably implies that it contains either some type of recorder or a telemetry device. Radio telemetry does present some technical problems, but is probably more suitable for out-of-doors use than is infra-red telemetry (Weller, 1977). Magnetic tape recorders are technically more attractive than radio telemetry, but they are heavier, bulkier and more expensive than low-powered transmitters. The latter factor could be of real importance if the risk of damage to the apparatus is high.

Methods involving respiratory exchange

The most commonly practised method of measuring energy expenditure in large animals without the use of a respiration chamber is with a mask or hood, a method which is virtually identical with that used in human studies. This method was extensively used by Brody (1945) and his associates for many years with animals ranging from elephants to sheep, and more recently by Webster (1967) and Brockway & McEwan (1969*a*) with sheep, Hornicke, Ehrlein, Tolkmitt, Nagel, Epple, Decker, Kimmich & Kreuzer (1974) with horses and Ribeiro, Brockway & Webster (1977) with cattle. Hood and mask methods are totally unsuited for measurements on grazing animals and are essentially laboratory rather than field methods. A variation on this technique has been used on cattle by Flatt, Waldo, Sykes & Moore (1958) and on sheep by Young & Webster (1963). Their methods involve the use of the Kofranyi-Michaelis (1940) respirometer as in human studies but avoid the hood or mask by means of tracheal cannulation. In Flatt's method a simple cannula fitted with a two-way respiratory valve was used and both inspiration and expiration took place through the valve, completely by-passing the nasopharynx. It is questionable whether the Kofranyi-Michaelis respirometer could deal with the high pulmonary ventilation rates generated in an adult cattle beast during exercise or in a warm environment, but the method may be criticized more seriously (Young & Webster, 1963) in that the by-passing of the nasopharynx disturbed 'olfaction, communication and thermoregulation' (by respiratory evaporative cooling). The latter workers sought to avoid these difficulties by the use of a re-entrant tracheal cannula which allowed inspiration to take place through the nasopharynx and expiration through the respirometer. This method certainly allows normal olfaction, which is very important in relation to feeding behaviour, and probably allows normal communication, but it is difficult to envisage much improvement to the impaired respiratory thermoregulation as the sheep has a highly developed heat-exchange mechanism in the turbinate region, which heats and moistens inspired air and cools and extracts water from the expiratory airflow.

A more general criticism of methods involving tracheal cannulation lies in the fact that they involve surgery, which, while admittedly not of the most severe kind, does imply some risk and expense, and more seriously, general experience with preparations involving such cannulation indicates variable and often quite short survival (Blaxter & Joyce, 1963). Quite apart from any ethical considerations, and these cannot be too lightly dismissed, the expense of losing an adult cattle beast, after perhaps only 2 or 3 weeks of useful experimental life, is considerable.

Isotope dilution methods

If methods involving measurement of respiratory exchange to estimate energy expenditure are unsuitable the experimenter is obliged to look for other physiological correlates of energy expenditure. Two isotope dilution techniques have been proposed for this purpose: the double-labelled water method (Lifson, Gordon, Visscher & Nier, 1949) and the carbon dioxide entry rate technique (Young, Leng, White, McClymont & Corbett, 1969). Both of these methods measure carbon dioxide production, and so introduce an uncertainty, albeit a small one, due to an unknown respiratory quotient (RQ), into the assumption of the energy equivalent of unit oxygen consumption. (The energy equivalent of each litre of oxygen consumed ranges from 19·6 kJ at RQ 0·70 to 21·1 kJ at RQ 1·00 (Lusk, 1928), i.e. a total range of about 7·5%.) With large animals the double labelled water technique is expensive in terms of both reagent and analytical costs, and it is only suitable for measurements over periods in excess of 24 h (Lifson, Little, Levitt & Henderson, 1975). The carbon dioxide entry rate technique seems more suited to work with large animals and has been used with both cattle and sheep (Young, 1970; Corbett, Farrell, Leng, McClymont & Young, 1971; Whitelaw, Brockway & Reid, 1972; Whitelaw, 1974; Engels, Inskip & Corbett, 1976). The method suffers some disadvantages, particularly if short-term changes in metabolic rate are of interest. Measurements can only be made over periods of 2 to 3 h, and if the animal is to remain undisturbed by regular attention from the experimenter, some system of automatic collection of sequential samples would be required, which would add to the bulk and vulnerability of the equipment package to be carried by the animal. Further increase in the accuracy of the technique could be obtained if simultaneous estimates of RQ could be made, but again this would entail increased bulk of apparatus: this may be practicable for cattle but would not be so for sheep. In its most developed form (Engels *et al.* 1976) the technique with sheep involves the continuous infusion of $NaH^{14}CO_3$ in saline solution into the jugular vein and continuous collection of parotid saliva. Infusion and collect rates are 5 ml/h. The technique is undoubtedly both useable and useful, being able to predict energy expenditure with an error of better than ±10%, over periods of 2–3 h. It is relatively inexpensive, and requires little surgical interference and no individual 'calibration' of the animals.

Heart rate as a measure of energy expenditure

The use of heart rate to predict energy expenditure is superficially very attractive, but in ruminant animals, at least, the relationship between the parameters is less than satisfactory. Oxygen consumption and heart rate are related in the following manner:

$$\dot{V}_{O_2} = (\mathrm{HR} \times \mathrm{SV}) \times (C_{A_{O_2}} - C_{V_{O_2}})$$

where $\dot{V}_{O_2}$ is oxygen consumption, HR is heart rate, SV is stroke volume of the heart, C_{AO_2} is arterial blood oxygen content and C_{VO_2} is venous blood oxygen content. The product HR×SV is the cardiac output. If $\dot{V}_{O_2}$ is to be predicted from HR it is necessary that the remaining terms on the right-hand side of the above equation vary in a systematic way, or that they remain constant.

Webster (1967) and Brockway & McEwan (1969*b*) have reported on the relationship between oxygen consumption and heart rate in sheep: individual 'calibration' of the animals is essential and while a possibly acceptable relationship can be discerned in some animals, in others no detectable relationship appears to exist. Brockway & McEwan (1969*b*) suggested that the relationship appeared to be disrupted by highly variable cardiac stroke volumes, probably in response to emotional factors.

More recently we have made further observations of the relationship in two steers exercised on a treadmill. Energy expenditure was measured by indirect calorimetry using an open-circuit ventilated-hood system for 30 min when the animal was standing quietly and then for a further 30 min when walking at speeds between 40 and 80 m/min. Average heart rates for the standing and walking periods were calculated from measurements made at 5-min intervals through the experiment. Twelve experiments were performed on one animal and eleven on the other. The results for the two animals were similar, although not statistically combinable, that is, distinct relationships existed for each animal. Due to large

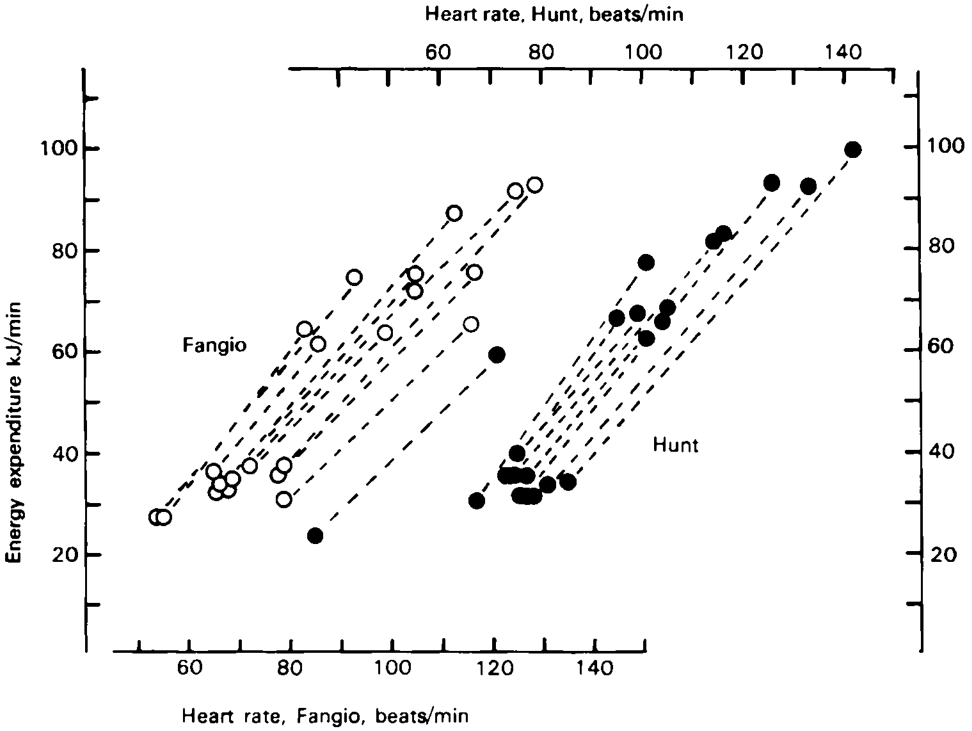

Fig. 1. The relationships between heart rate and energy expenditure in two steers, Fangio and Hunt. Each dotted line connects the standing and walking values of the parameters, measured over 30 min, on different days.

within-animal between-day variation it was not possible to derive meaningful, predictive relationships. The variation was mainly due to differences in the intercepts, with much smaller differences in the slopes (Fig. 1).

The intercept terms of the lines shown in Fig. 1 varied in a random manner with respect to time. For each animal energy expenditure when walking could be estimated from heart rate if the values of these parameters when standing are known on any given day. The prediction equations were $\Delta E = 1{\cdot}092\ \Delta R$ for steer Fangio and $\Delta E = 1{\cdot}190\ \Delta R$ for Hunt, where ΔE is the difference between walking and standing energy expenditure (kJ/min) and ΔR is the difference between walking and standing heart rates (beats/min). The relationships are shown in Fig. 2.

The regression coefficients differed significantly ($P<0{\cdot}05$), but both equations predict the additional energy expenditure when walking to within ±8%. At first sight this approach seems to have but limited application, but perhaps it could be combined with the carbon dioxide entry rate technique, to estimate the energy cost of short-term activities over and above a longer term estimate of resting energy expenditure provided by the entry rate measurement.

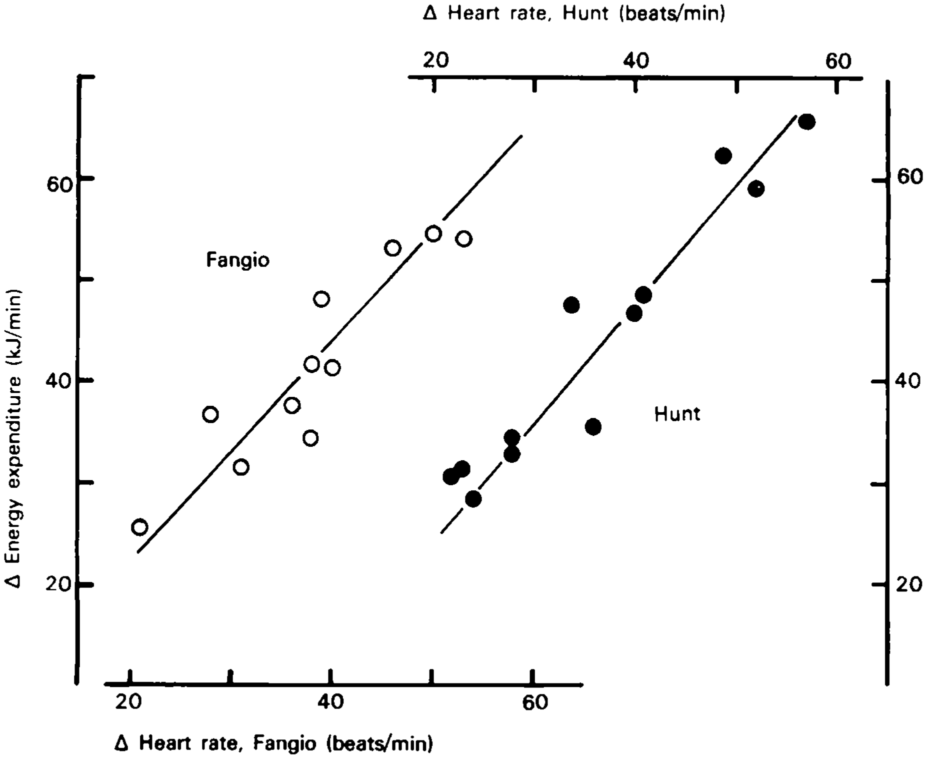

Fig. 2. The relationships between the differences in heart rate and energy expenditure while standing and walking, in two steers, Fangio and Hunt. Each point represents the (walking-standing) value of the parameters on different days.

Other correlates of energy expenditure

One approach to the problem is to estimate sensible heat loss from the animal. The major limitation of this method is that it does not estimate evaporative heat loss, but this may be of no major significance in cool environments. This topic is discussed by Mount (1977) in this meeting and is only mentioned at this point for the sake of completeness.

We have currently been looking at the possibility of estimating energy expenditure in exercising cattle from the partial pressure of oxygen in arterial and venous blood ($P_{A_{O_2}}$, $P_{V_{O_2}}$), heart rate and rectal and skin temperatures. This work is at a very early stage and comments on it must be of a highly tentative nature. A series of eight experiments on one animal over 5 d suggest the following: $P_{A_{O_2}}$, $P_{V_{O_2}}$ or $\Delta P_{A\text{-}V_{O_2}}$, or heart rate, by themselves are not useable predictors; but a combination of $\Delta P_{A\text{-}V_{O_2}}$ and heart rate, or of $k - P_{V_{O_2}}$ and heart rate (where k is a constant, assumed, value for $P_{A_{O_2}}$) may provide the basis for an acceptable prediction of energy expenditure. The introduction of body temperature into the regression does not, at this stage, appear to offer improved accuracy of prediction of energy expenditure.

Further work is in progress and if these tentative views can be substantiated a quite attractive technique can be envisaged where a combined P_{O_2} electrode (Soutter, Conway & Parker, 1975) and ECG electrode is introduced into the vena cava, by way of the jugular vein, with the animal carrying battery-powered preamplifiers and a two-channel radio transmitter. It must be emphasized that the development of such a technique requires much further work; we remain without a really satisfactory solution to the basic problem.

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