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# Whole-body calorimetry in man and animals

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**Abstract**

This review assesses the role of calorimetry in studies of energy exchange in man and animals. The techniques of direct and indirect calorimetry for measuring energy expenditure of the whole body, as distinct from its constituent parts, are discussed. Attention is focused on the advantages and disadvantages of the various techniques and the experimental conditions under which measurements are made. In particular, the effects of thermal environment, nutrition, physical activity and the interactions between these variables are considered, because they can have a striking influence on the conclusions drawn from an investigation. Finally, consideration is given to current topics of controversy and problems of energy metabolism which remain to be investigated.

## INTRODUCTION

Whole-body calorimetry has been used extensively as a fundamental tool for investigating energy exchange in relation to problems of nutrition and metabolism, thermoregulation, exercise physiology and disease. The pioneering studies of Lavoisier, de LaPlace and Crawford [1–4] some 200 years ago paved the way for the modern science of calorimetry. At the turn of the century, considerable progress was made in nutrition by Atwater and Benedict [5] and Rubner [6] and later by Lusk [7]. The 1930s and 1940s were the start of an important period for advances in thermoregulation in man [8,9], while in the 1950s and 1960s significant progress was also made in our understanding of energy exchange in domestic animals [10,11]. The impetus for this work in animals was largely economic, with the aim being to ensure maximum efficiency of production.

It was not until the 1970s that the modern science of whole-body calorimetry in man became re-established. The impetus was now a need to determine man's requirement for energy in order to optimize health. Obesity was becoming an increasing health problem in affluent societies and there was an urgent need to determine whether there is a metabolic defect in individuals who become obese. However, it was first essential to obtain accurate information on the energy expenditure of normal-weight individu-

als before deciding whether the obese had a metabolic abnormality. Interest therefore lay in the extent to which there are fundamental differences in energy expenditure between individuals, and the degree to which the thermal environment and nutrition can influence energy metabolism. Additional aspects related to the effects of body composition, age, stress, drugs and disease, and the importance of physical activity.

The entire field of energy metabolism in animals and man has been reviewed recently by Blaxter [12]. The aim of this short review is to concentrate on some of those aspects which have helped in our understanding of the control of energy balance and to highlight some of the controversies and problems in this field.

## METHODOLOGY

A detailed account of the techniques of animal and human calorimetry has been published by McLean and Tobin [13]. This provides an extensive review of the historical background, instrumentation and problems associated with measurement of energy expenditure. Webb [14] has written a detailed review of direct calorimeters used for human subjects, while van Es [15] has described the experimental protocols in use for studies of energy expenditure in man over periods of 24 h. In this short review, it is only possible to provide an outline of the techniques used in calorimetry, and to draw attention to the particular advantages and disadvantages of the various techniques and the extent to which they can influence the conclusions drawn from an investigation. Discussion is limited to measurement of energy exchange of the whole body, as distinct from its constituent parts.

Both direct and indirect calorimetry can be used to estimate the energy expenditure of an individual. The former measures heat loss from the body while the latter estimates heat production. The relation between these two components of heat exchange can be described by the equation

$$\phi_p = \phi_l + \phi_{st}$$

where $\phi_p$ is the rate of heat production, $\phi_l$ is the rate of heat loss and $\phi_{st}$ is the rate of change of stored heat. Correction should also be made for any work done on the surroundings which does not appear as heat loss.

### *Direct calorimetry*

A wide variety of species has been investigated by direct calorimetry, including man and his domestic animals, rats, rabbits, lizards, snakes and tadpoles [16]. Direct calorimeters are usually either of the heat-sink or gradient layer type. The first type is sometimes termed an adiabatic calorimeter: it depends on absorbing the animal's heat loss and measuring it as a

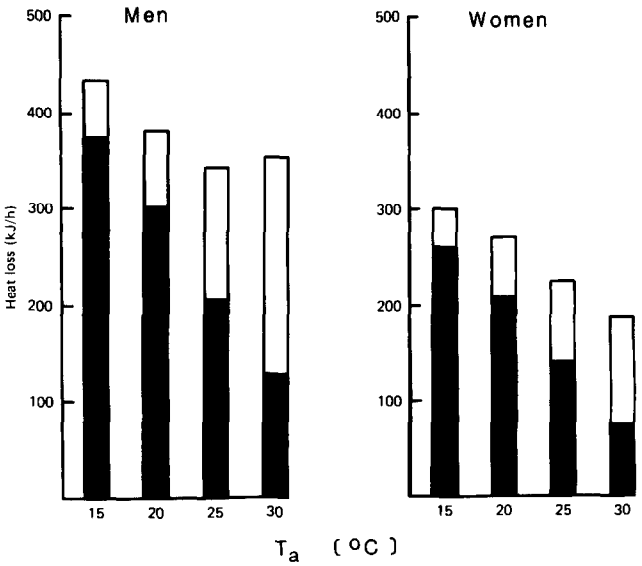


Fig. 1. Total heat loss divided into its sensible (■) and evaporative (□) components. Measurements were made for 2–3 h at a series of ambient temperatures ($T_a$) in a direct calorimeter. From Close et al. [36].

rise in temperature of the absorbing medium. Examples are the Atwater–Rosa–Benedict apparatus used for man at the turn of the century [17,18] and more recent calorimeters for the pig [19] and human subjects [20–22]. The second type is isothermal and uses a thermal gradient method: it depends on measuring the temperature difference produced across a thin layer of material lining the inside of the calorimeter. This was the type originally used by Rubner [23] and more recently for studies in human subjects [9,24] and cattle [25].

One advantage of the direct calorimeter is that it allows partition of total heat loss into its sensible and evaporative components; an example of such results is given in Fig. 1. The rate of heat loss is given by

$$\phi_l = \phi_{ls} + \phi_{le}$$

where $\phi_{ls}$ is the sensible or non-evaporative heat loss, and comprises heat exchanges by radiation, conduction and convection, and $\phi_{le}$ is the evaporative heat loss. The heat-sink calorimeter is probably less demanding in construction than the gradient layer type. The chamber for the subject consists of a well-insulated box which is best contained within a temperature-controlled room. If the temperatures inside and outside are the same, no heat should be lost through the walls. When a subject is introduced into the chamber, the heat lost from the body tends to make the air temperature rise. This rise is, however, prevented by a heat exchanger which removes exactly the amount of sensible heat lost from the body [20].

At its simplest, the heat exchanger consists of a radiator through which water passes at a constant rate. A control heater positioned in the circulatory system before the heat exchanger operates both to control the internal temperature of the calorimeter and to allow sensible heat to be taken up by the heat exchanger, by varying the heat input to the water [20]. Sensible heat loss can then be calculated from the temperature difference across the heat exchanger and the rate of flow of water.

A number of additional controls and variations can be introduced. The inner wall of the calorimeter can take the form of a metal sheet around which air is circulated. Any radiant heat picked up by the wall is then transferred rapidly by convection to the ventilating air stream [26]. The alternative solution, of a thin metal sheet backed with a thick layer of insulated material, allows the radiant heat to be reflected back into the calorimeter [20]. The use of proportional-with-integral controllers and a zero-gradient control across the walls [20,26], rather than control of outgoing air temperature [19], also improves the precision of the instrument. The other component of heat loss, the latent heat of vaporization, is estimated from the ventilation rate and difference in water content of ingoing and outgoing air streams. The accuracy for measurement of sensible heat loss can be better than 1%, while for evaporative heat loss it is in the order of 2%.

The gradient layer calorimeter depends on measuring the rate of heat flow through sensors on the inner wall of the calorimeter [24,25]. The sensors consist of a sheet of relatively poor conductor, such as epoxy resin, with thermocouples placed on either side. Heat flow through the sensors is then proportional to the temperature difference. In practice, a series of thermocouples, i.e. a thermopile, is used and the signal is integrated over the whole area of the calorimeter. Using a gradient layer, all the radiant component of heat loss and most of the convective component are detected. The small amount of convective heat loss which does not pass through the wall leaves the calorimeter in the exhaust air and some provision for its detection must be made. The latent heat of vaporization is also estimated from measurements made on the exhaust air. As the air is removed from the chamber, it passes over a plate which also contains a gradient layer. The air is cooled, causing water vapour to condense and thus liberate the latent heat. Details of construction vary from one calorimeter to another and details have been described previously [13,14].

Apart from heat-sink and gradient layer calorimeters, direct calorimeters can be of either the convection or the differential type. In the convection calorimeter, the mean temperature rise of the ventilating air stream is measured. It therefore requires extremely accurate temperature measurement and only in relatively recent years has it proved a reliable technique [27–29]. The differential calorimeter is well suited for use with small animals and operates with two identical chambers: the control chamber contains a heater whose measured input is varied to produce exactly the same temperature rise

as the test chamber containing the subject [30,31]. In general, however, the heat-sink and gradient layer types of calorimeter are most widely used. The size can vary considerably, from one in which there is just enough space for the subject to lie or sit, to the larger type with the dimensions of a bed-sitting room. This clearly affects the lag-time between heat being lost from the subject and being recorded. In practice, the response time for sensible heat can be short, provided the furniture and bedding are of low heat capacity.

Using either the heat-sink or gradient layer principle, it is possible to construct a suit calorimeter which is worn by the subject. Tubing can form the walls of a suit and act as a heat-sink [32], resistance thermometers as wire grids on either side of an insulated suit can operate on the gradient layer principle [33]. Alternatively, a series of heat-flow meters as described by Hatfield [34] and McGinnis and Ingram [35] can be used. The major advantage of these systems is that they are fully portable, although they are only appropriate for relatively hairless subjects such as man and pig. However, because they impede evaporative heat loss, they are unsuitable for a subject who is sweating. The suit must therefore be controlled such that there is no thermal sweating or, alternatively, the ambient temperature must be adjusted to the point below which sweating would occur, and this clearly limits the environments in which it can be used. Comparisons have been made between heat loss measured simultaneously from heat-flow meters attached around the waist and from a whole-body calorimeter in human subjects at several environmental temperatures between 15 and 30°C [36]. Heat loss determined by the meters was lower than that determined by the calorimeter and the difference increased at the higher temperatures. It was concluded that heat-flow meters could provide a useful estimate of total heat loss when the evaporative component is low. This could be of particular value in sedentary individuals when the heart-rate method may be inappropriate [37]. The advantage of an animal such as the pig is that there is no thermal sweating, making it especially suitable for use with heat-flow meters [38].

A disadvantage of the direct calorimeter is that it cannot provide reliable estimates of energy expenditure by the body over short periods of time. This is because of the thermal capacity of the body which allows changes in heat storage, resulting in a change in deep body temperature and an alteration in the body's heat balance which is not immediately reflected in heat loss from the body. Since deep body temperature rather than total body temperature is controlled, the core itself contracts and expands depending on the environmental and nutritional conditions, time of day and level of activity. Therefore, although reliable estimates of energy expenditure can be made under steady-state conditions, rapid changes in metabolic rate cannot be determined accurately. Thermal capacity and body heat content in man have been discussed by Minard [39]. Although a value of 3.47 kJ per kg body

weight per °C is commonly used in man, a value based on the fat content of the subject may be more appropriate.

*Indirect calorimetry*

Using indirect calorimetry, energy exchanges are estimated from the measurement of material exchanges. Heat production, or metabolic rate, is usually measured as the rate of oxygen consumption, sometimes in combination with measurements of carbon dioxide and methane production and urinary nitrogen excretion. Estimates are occasionally based on carbon dioxide alone or calculated from rates of turnover of carbon and nitrogen in the body. Other indirect methods are based on isotopic estimation of carbon dioxide production or on recording heart rate and correlating this with oxygen consumption.

Methods used for assessing respiratory gas exchange either contain the subject within a respiration chamber, or use a face-mask, hood or tube which is more closely associated with the lungs. Respiration chambers, sometimes incorrectly termed indirect calorimeters, can be of three main types: confinement, closed circuit [40] and open circuit [41]. In the confinement system, which was probably used first by Lavoisier, changes in gas concentration are estimated while the subject is contained within a sealed chamber. The obvious disadvantage of such a system is the fall in oxygen concentration with time. This was overcome by Blaxter et al. [42] who alternated a 50 min confinement period with 3 min in which fresh air was allowed into the chamber. The confinement system has been used for a wide variety of species including guinea-pigs, rabbits, dogs, pigs and cattle [42–45].

In the closed-circuit system, air is circulated within a closed circuit, carbon dioxide and water produced by the subject are absorbed, and the amount of oxygen needed to replace that used by the subject is measured. This system is not only well suited for use with small animals such as the mouse [46], rat [47] and chick [48], but was also used by Atwater and Benedict [18] for man, and later for sheep [49], cattle [50] and newborn babies [51]. In practice, the gas composition and water vapour in the chamber need to be estimated at the start and end of the run, because they are very unlikely to remain exactly the same as in fresh air. Moreover, even a very small change in temperature inside the chamber will cause gas either to expand into or be sucked out of the chamber. The system must also be absolutely leak-free, because even a very small leak may lead to significant error.

In the open-circuit system, the Fick principle is used to estimate metabolic rate: air flows through the chamber at a known rate and oxygen consumption is estimated from a knowledge of this flow rate and the concentrations of ingoing and outgoing oxygen and carbon dioxide. Such a

system was used by Atwater and Rosa [17] before their design was modified to a closed-circuit apparatus. However, the open-circuit system demands highly accurate measurement of gases at frequent intervals and it was only after the development of appropriate instrumentation that the technique gained in popularity. It has been used extensively in recent years for studies with human subjects [15,20,52,53], primates [54], pigs [55,56], cattle [57], poultry [58,59], rats [60,61] and mice [62].

The principles of indirect calorimetry are extremely straightforward. In practice, however, there are numerous places where errors in the estimation of heat production can arise. It is therefore essential that a wide range of calibrations and checks are incorporated into any system and these have been described in detail by McLean and Tobin [13].

Although relatively cheap and easy to construct, closed-circuit and confinement chambers lack precision over short periods. With the development of instrumentation that can monitor the gas contents of a stream of air continuously, the closed-circuit system is now used infrequently. The open-circuit system can provide information on minute-to-minute changes over a 24-h period, whereas the other chamber systems can rarely provide information over less than 30 min. Although a respiration chamber has an inherent lag-time because of the volume of air in the chamber, the flow-through method operated in open-circuit calorimetry allows rates of changes of gases to be determined. Thus, after appropriate calculation, instantaneous rates of gas exchange can be estimated [63–65]. The use of such a calculation becomes extremely important in studies where the immediate metabolic response to a particular stimulus such as food, temperature, exercise or a drug is being investigated, and its use is demonstrated in Fig. 2. Combined with the use of an "activity meter", it is then also possible to separate total heat production into components due to rest and activity (Fig. 3) [61,62,66].

In open-circuit systems, oxygen consumption cannot be estimated accurately from estimates of ventilation rate and oxygen concentration difference alone [64,67] and, to reduce the error of up to approximately $\pm 6\%$, measurements of carbon dioxide must also be made. Paradoxically, however, heat production as opposed to oxygen consumption can be estimated accurately, with an error of only $\pm 1\%$, if only ventilation rate and oxygen concentrations are measured. This is because the percentage error of assuming that oxygen consumption is equal to ventilation rate multiplied by the oxygen difference is virtually cancelled by the percentage change in the energetic value of oxygen consumed with variation in respiratory quotient.

It is possible to obtain very good agreement between 24-h estimates of heat loss and heat production when simultaneous measurements are made within a chamber. In a series of 36 subject runs using direct and indirect calorimetry as described by Dauncey et al. [20], a mean difference of only $1.0\% \pm 0.2$ (SEM) was obtained between the two measures of 24 h energy expenditure [68–70]. Such accuracy allows investigation of factors that may

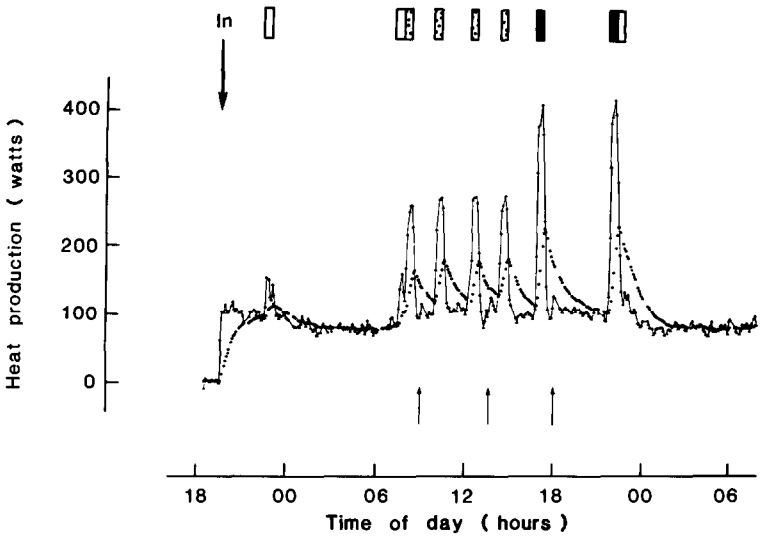


Fig. 2. Demonstration of the method for calculating rapid changes in gaseous exchange. A human subject lived in an open-circuit respiration chamber for 36 h and results are given with (———) and without (· · · · · ·) the correction for lag-time due to the volume of air in the chamber. Note the difference this calculation makes to the response time and peak value after a change in activity such as standing (□) or cycling at different work loads (▨, ■). Meals were provided at times indicated by arrows (↑); the subject was lying in bed between 23.00 and 07.30 h each night and, except where indicated, was usually sitting down. From Brown et al. [64].

influence energy expenditure by only a few percent and yet be of critical importance to energy balance.

Many studies have estimated respiratory gas exchange without the subject being confined within a room. These tend to allow greater freedom of movement than the chamber methods, but at the expense of reducing the

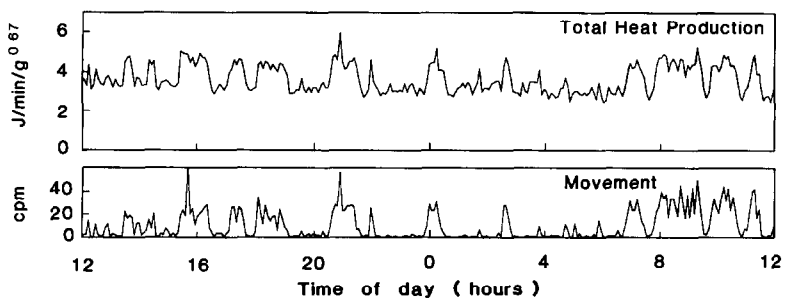


Fig. 3. Total heat production corrected for lag-time as demonstrated in Fig. 2, and physical activity estimated as discrete movements in counts per min (cpm) using a microwave Doppler system, in a Wistar rat living at 28°C in an open-circuit respiration chamber. Note coincidence of peaks in heat production and movement, which allows assessment of resting or "underlying" thermogenesis as described in Brown et al. [61].

level of accuracy. However, complete freedom of movement is not necessarily implicit in all such methods and some are more ideally suited to measurements of resting metabolic rate. Spirometers, such as those of Krogh [71,72] and Benedict and Roth [73–75] operate on the closed-circuit principle, and the measurement time of 10–20 min can be extended by addition of known amounts of oxygen to the spirometer. Total collection systems allow for collection of all expired air and its subsequent analysis, and two such systems include the Tissot gasometer [76] and the Douglas bag [77]. Inherent in all these systems are several sources of error including those associated with leaks, temperature changes, incomplete absorption of carbon dioxide, respiratory valves with a high resistance and diffusion of gases through the container. They have, however, been used in a wide variety of studies in man [71,74,78,79] and farm animals [49,80]. These methods have also proved especially useful for practical classes with students.

One of the most popular indirect open-circuit systems in use today with resting subjects is the ventilated hood flow-through method [81]. This is often used in clinical studies and, as with all open-circuit calorimetry, it has benefited considerably from the development of appropriate technology for continuous accurate measurements of gas concentrations. Nevertheless, with all these methods, although the subject is not confined within a chamber, freedom of movement is considerably limited.

A particularly valuable open-circuit system which is also portable is the Kofranyi–Michaelis or Max Planck Respirometer [82,83]. Despite the possibility of major errors associated with contaminated samples of expired air and faulty estimates of flow [84], the method has been used extensively in studies of man carrying out a wide range of activities [85–87]. It has proved invaluable in activities where the Douglas bag is either too bulky or of limited capacity. A number of other highly portable open-circuit systems have since been developed and used with varying degrees of success [13]. These include the integrating motor pneumotachograph or "Imp" [88], the "Miser" (a miniature, indicating, sampling, electronic respirometer) [89], and the "Oxylog" [90]. The aim has always been to produce an accurate, light-weight system which can be used over long periods of time. Problems have tended to involve leaks or electronic components used in the system. A simple, fast-responding system is the Metabolic Rate Monitor (MRM) [91] but, despite its advantages, it has been very little used.

A problem inherent in all the non-chamber techniques is that total gaseous exchange does not take place exclusively through the lungs [12]. In man, oxygen uptake by the skin is in the order of 2% of total consumption and carbon dioxide loss can be close to 3% [92]. In bats, whose wings allow a large area for diffusion, the loss of carbon dioxide can be as high as 6%. The loss of carbon dioxide through the skin is even greater in lizards and snakes and can be up to 10% and 35%, respectively [93].

Because of the restrictions imposed by a hood, mask or mouth-piece,

attempts have been made to estimate heat production by less intrusive techniques. Two isotopic techniques for estimating carbon dioxide production are the doubly labelled water method [94] and the carbon dioxide entry rate method [95]. The doubly-labelled water method is expensive and of limited accuracy: the accuracy for measuring carbon dioxide output is within about ±2% whereas, because of the error in estimating respiratory quotient, the overall uncertainty in estimating heat production is at least 6% [13]. Nevertheless, it is well suited to relatively long-term measurements, in the order of 5–14 days in man, in whom it has been used extensively [96–99]. It has also proved an ideal technique for the estimation of "field" metabolic rate in free-living animals and birds [100]. The carbon dioxide entry rate method is ideal for use with freely grazing animals such as sheep [101]. However, it overestimates carbon dioxide production by about 2–4% [102], because of the slow equilibration of carbon dioxide with carbonate in bone. As with the doubly labelled water method, there are also limitations associated with making assumptions about respiratory quotient in order to predict heat production.

There tends to be a close relation between heart rate and oxygen consumption during graded levels of exercise and with the development of relatively cheap, portable heart-rate monitors, heart-rate has been used as a predictor of 24 h energy expenditure [103,104]. However, extreme caution should be used with this method because very large errors can occur in the predicted values of energy expenditure [37]. This is because the relation between heart rate and oxygen consumption can be influenced by posture and in many sedentary individuals the standard calibration curve is inappropriate. Ideally, each subject should be calibrated within a whole-body calorimeter. However, this is not always possible and a compromise is probably to use heart rate as a predictor of energy expenditure only when it is above a certain minimum value. Combined with appropriate calibration curves, the heart-rate method can then provide reasonably accurate estimates of total energy expenditure for groups of subjects, and the precision of individual estimates can be better than ±10% [105]. Although these various field methods can be invaluable for use in free-living subjects, the level of control of the experimental variables is limited and may lead to considerable errors in interpreting the results.

## BASAL OR STANDARD METABOLISM

In order to allow comparisons of metabolic rate either within or between individuals, accurate measurements as described in the previous section need to be made under very carefully controlled conditions of nutrient intake, thermal environment and physical activity. Attempts at standardization have led to a large number of terms being used to describe this rate of metabolism, including basal, standard, fasting and resting metabolic rate

[12,106,107]. The important point is that, for valid comparisons to be made, the conditions should be as similar as possible.

The basal or standard metabolism makes a significant contribution to total 24 h energy expenditure. In women, ratios of 24 h energy expenditure/"basal" metabolic rate between 1.3 and 1.4 have been recorded [70,108]. This is considerably less than the suggested minimum value for maintenance of 1.5 for women [109], indicating the low level of activity engaged in by many individuals. Even in the wild the standard metabolic rate can still account for some 50% of the "field" metabolic rate of larger animals [110].

Physiological and biochemical mechanisms accounting for the standard metabolic rate are outside the scope of this review and have been discussed in detail elsewhere [12,111,112]. The aim of this section is to discuss those factors which influence resting metabolism and the standard conditions under which it is best measured.

*Food intake*

In adult man, the resting metabolic rate may be elevated even when measured at least 14 h after the last meal [68]. Similarly, in the young pig, this rate of metabolism can remain elevated for at least 20 h after feeding [113]. The extent to which this elevation is due to post-absorptive, as distinct from absorptive, processes is not known. Nevertheless, the post-absorptive

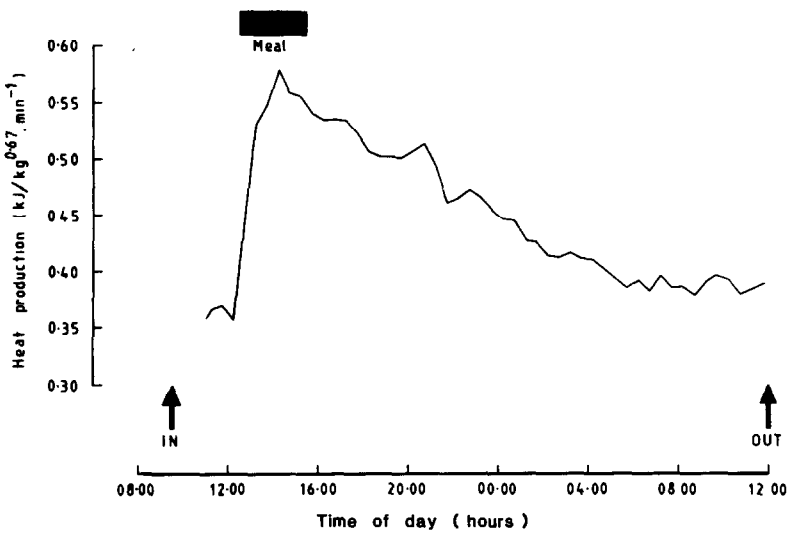


Fig. 4. Energy expenditure of a young pig living at 25°C in an open-circuit respiration chamber. Estimate of standard or "basal" metabolic rate would start at approximately 06.00 h, 14 h after the end of the meal, when the thermogenic effects of the food and overt physical activity were minimal. From Ingram and Dauncey [55].

state is not thought to be reached until about 3–5 days in ruminants, 2 days in chickens and 10–20 h in small omnivorous animals [114].

Ideally, the subject should not only be in the post-absorptive state but should also have reached a point where the decline in metabolic rate since the last meal has reached a plateau. In practice, this may involve a prolonged period of fasting and there is even the possibility that metabolic rate would continue to decline until death. Conditions such as these clearly yield information about the truly minimal metabolism but they are obviously obtained under conditions which rarely occur. Moreover, in some cases it might be quite impractical to use long periods of starvation. The temptation might, therefore, be to standardize the conditions by measuring metabolic rate for example at 24 h after the last meal. However, this could be quite inappropriate for comparisons between different species such as the mouse and elephant.

For practical purposes, a standard set of conditions is required under which metabolic rate can be measured and then compared between, for example, different species, ages or treatments. The compromise solution should involve measuring metabolic rate at least after the initial elevation associated with feeding has subsided and metabolism is declining only slightly (Fig. 4).

*Critical temperature*

Standard metabolic rate should be measured in a thermally neutral environment, with the ambient temperature being between the lower and upper critical temperatures [115]. For all homeothermic animals, the lower critical temperature ($T_c$) is the environmental temperature below which metabolic rate must increase in order to prevent a fall in deep body temperature. As environmental temperature increases, there is eventually a point at which body temperature increases and this will result in a rise in metabolic rate because of the Arrhenius–van't Hoff effect [116]. This environmental temperature is known as the upper critical temperature.

For some species, the difference between lower and upper critical temperatures may be only one or two degrees Celsius. Different species can have widely differing values: in the adult rat the lower critical temperature is about 28°C, while in the Arctic fox in its winter coat, the value is approximately −40°C [115]. Furthermore, values may also be quite different for animals of the same species but of different age. Lower critical temperatures of 34 and 25°C have been reported for newborn and 3-month-old pigs, respectively [11], while a value of 33°C in the human newborn infant compares with 28°C in adult man [115]. In addition, the level of insulation will affect the zone of thermal neutrality. The lower critical temperature falls to about 15°C when adult man is clothed, and values in sheep are strikingly affected by whether or not they have been shorn. Moreover, in animals with

a fur coat, the depth of fur tends to vary from one season to another and so alter the critical temperature. Whether the animals are kept in groups or singly will affect critical temperature [11], while the important influence of nutrition can be deduced from the earlier discussion on food intake and will be considered in more detail in a later section. Other environmental conditions also need to be kept constant and these will be considered in the next main section.

*Body size and deep body temperature*

It has long been recognized that large animals have a higher metabolic rate than small ones, although they produce less heat per unit body weight. At the turn of the century, Rubner [117] and Voit [118] showed that the fasting metabolism, for eight different species from an 18-g mouse to a 441-kg horse, is closely related to surface area, with a mean value of 48 W $m^{-2}$. This would be expected, on theoretical grounds, as the rate of heat loss of an individual is proportional to surface area [119]. In practice, the relation between body weight and metabolism is more closely described by the three-quarters power, when comparisons are made between species [120–123]. However, detailed inspection of a large number of results shows that, although metabolism increases with $(\text{body weight})^{0.75}$ within subgroups of both homeotherms and poikilotherms, there is considerable variation from one subgroup to another [12,124,125]. For example, placental animals have a higher metabolism than marsupials, whereas bats have a considerably lower fasting metabolism than small insectivores such as the shrew. In general, it appears that the power value for different groups of animals ranges from less than 0.6 to more than 0.9 [126], and care should therefore be taken in interpreting results where 0.75 is used [127].

Another factor which affects the difference between species is their shape. The more closely the animal approximates a sphere, the lower the surface area in relation to unit body mass. Thus, although metabolic rate is proportional to a power function of body weight, different shape factors must be used. These factors represent an average for the species, and body composition within a species is also of importance. Subjects with a high body fat content will probably approximate a sphere more closely than those with little fat.

Metabolic rates of the various orders or classes are, in general, proportional to deep body temperature, with birds having higher body temperatures and metabolic rates than mammals, and placental mammals having higher values than marsupials [12]. Such a relation would be predicted from the Arrhenius–van't Hoff effect. Therefore, of particular relevance to any comparisons of metabolic rate is the deep body temperature which is being maintained. If the rise in metabolic rate for a 10°C increase in ambient temperature ($Q_{10}$) is known, a correction can be made to a standard body

temperature. Alternatively, a $Q_{10}$ of about 2 can be assumed. The importance of this correction clearly depends on the temperature difference between individuals. This is usually small within a given species and is often ignored, although under some conditions and between species it can be significant [128].

*Temporal variations*

Variations in deep body temperature, metabolic rate, thermal conductance and heat loss occur cyclically over 24 h in most species [115,129,130]. These appear to be innate and occur independently of nutritional status and physical activity. Nevertheless, these two variables themselves have a profound influence on metabolic rate, as will be discussed later. In resting man, circadian variations of heat loss are responsible for about 75% of the range of oscillation in core temperature, while the variation in heat production contributes only about 25% [129].

In human subjects measured from day-to-day or week-to-week under carefully controlled conditions, the variation in energy expenditure is small. Coefficients of variation in the order of 2% have been reported [131–134]. Changes do, however, occur in relation to the stage of the menstrual cycle: metabolic rate during sleep is lowest in the late follicular phase and highest in the late luteal phase [135]. There are many conflicting reports on the seasonal variation of basal metabolism in man [136,137]. The large variations which have been reported in some studies are possibly related to differences in diet.

From the previous discussion, it is clear that the evaluation of standard metabolic rate is subject to a variety of errors and the degree of control which needs to be exercised is dependent on the precision of the comparisons to be made.

## THERMAL ENVIRONMENT

In many studies, environmental temperature is represented as air temperature measured with a dry bulb thermometer. No indication is given that radiant temperature, air movement or humidity might be involved. However, if the critical temperature ($T_c$) is determined at constant air temperature, it is found that when the radiant temperature is high, $T_c$ is reduced. Moreover, $T_c$ is higher in windy conditions than in still air. In any calorimetric study, it is therefore desirable to have the radiant temperature equal to the air temperature, to have the air movement as low as possible and to control humidity. However, even when climatic conditions have been fixed, other environmental factors can still influence the critical temperature. The use of bedding or clothing will lower $T_c$ because of an increase in external insulation, huddling between animals will also reduce $T_c$ and a change in

posture will alter the surface area of the body available for radiant, convective and conductive heat transfer. Additional factors which can alter the effect of a given environment include previous thermal history, and nutritional and endocrine status [138].

The three modes of sensible heat transfer depend primarily on the difference between the animal's surface temperature and the corresponding environmental temperature. For radiative, convective and conductive heat transfer, these temperatures are radiant, air and floor, respectively. Radiative heat exchanges can be considered in relation to long-wave and short-wave radiation. Long-wave radiation is emitted by surfaces which are at a range of temperatures extending downwards from several hundred degrees Celsius, whereas solar radiation occurs at short wavelengths including the visible spectrum and the ultraviolet. A natural division occurs between these two radiant fluxes in the wavelength region between 2 and 3 $\mu$m [139]. An important point is that long-wave radiation is transmitted by very few substances which are transparent to the shorter wavelengths of the visible spectrum, as for example glass and Perspex (polymethylmethacrylate) [11].

For determining metabolic rate under standard conditions, it is simplest to ensure that the air and radiant temperatures are the same. Two methods can be used. First, by using a heavily insulated wall covered by a thin metal sheet [20]. The metal rapidly reaches air temperature and hence the conditions are standardized. Second, by using a false inner wall in the form of a sheet of material hanging approximately 5 cm from the structural wall [26]. The air then circulates around the inner wall, bringing it to the same temperature. In either case, the nature of the surface of the wall should be such that it will not reflect long-wave radiation. In practice this means that shiny metal surfaces should be avoided. If such a surface is used it will simply reflect back the surface temperature of the subject. With small animals, similar conditions can be achieved by keeping the animal in a thin-walled box and varying the temperature of the outer room [62]. However, the use of a small heater or cooler within the animal enclosure alone would result in control of air temperature but not the wall temperature and hence not the radiant temperature to which the animal is exposed.

In many studies, it is useful to be able to see inside the calorimeter, and glass or Perspex is often used as a window. However, it then needs to be appreciated that the window can transmit short-wave radiation and any sun falling on the window will impose an extra heat load. Such a problem was frequently encountered in incubators for pre-term infants before the significance of short-wave radiation and transmittance were fully appreciated [140].

Convective heat loss will increase as air movement increases and this should therefore be kept to a minimum. Still-air conditions are impossible to achieve simply because the subject's body will set up convection currents. In addition, some air movement is necessary in a calorimeter for ventilation

and the control of air temperature. Nevertheless, it is usually possible to keep air movement below the level of conscious perception or to standardize the position of the subject within the chamber.

Conductive heat loss will depend on the contact between the subject and the surrounding surfaces. For human subjects this can be controlled, whereas for animals it is more of a problem. The nature of the surfaces is particularly important because contact with a good conductor will transfer heat away from the body very much more rapidly than from a poor conductor. For example, heat loss will be considerably greater when sitting on a stone slab than on a feather cushion, even when both are at the same temperature.

The above conditions are relatively easy to achieve within the laboratory. However, in free-living subjects, the environmental conditions can vary considerably and strict comparisons between subjects or treatments are no longer possible. For example, if two individuals walk the same distance and carry the same load at an air temperature of 20 °C, one when the sun is shining and the other on a cloudy, windy day, they will be subjected to very different conditions of heat exchange. These could negate any effects which the investigator is attempting to examine.

Metabolic rate increases on exposure to cold, in man as in other animals [36,138,141–143]. Summit metabolism is the maximal rate of metabolism which can be maintained for 1 h in response to cold, without a decline in deep body temperature. This is in the order of 25 W $kg^{-0.75}$, which is approximately seven times the fasting metabolic rate and two to three times the "field" metabolic rate [12].

Many studies on cold exposure have concentrated on relatively low temperatures which cause at least a doubling of metabolic rate [144–146]. Nevertheless, even an environmental temperature which is only slightly below thermal neutrality can have a significant influence on energy expenditure. Women living under controlled conditions of food intake, activity, clothing and bedding were found to have an average 7% increase in 24 h energy expenditure when living at 22 compared with 28°C [147]. Before this investigation, it had been suggested that, given the chance, people do not expose themselves to cold severe enough to induce a metabolic response [148]. However, the upper temperature of 28°C was comfortably warm while the lower temperature of 22°C was judged to be typical of the level at which people often live without any obvious sign of shivering. It was calculated that these findings could have important implications for the maintenance of body weight. A recent study has shown that, even when the subjects are provided with "normal" clothing and bedding, there is a 7% increase in 24 h heat production in women at 20 °C compared with those at 28°C [149].

Recent whole-body calorimetry studies in the rat have shown that 24 h energy expenditure is approximately 25% greater in animals living at 21 °C compared with those at 28°C [61]. This has important implications for those studies in which animals were living at "normal" room temperatures of

between 20 and 25°C, which are below thermal neutrality, especially if they are kept singly without bedding.

Ambient temperature also has a significant influence on the partition of heat loss into its sensible and evaporative components (Fig. 1). In human subjects at a series of temperatures between 15 and 25°C, the rate at which sensible heat loss decreased for an increase in temperature was 8.2 kJ $m^{-2}$ $h^{-1}$ $°C^{-1}$, while evaporative heat loss increased by 3.4 kJ $m^{-2}$ $h^{-1}$ $°C^{-1}$ [36]. Similarly, in women at 28 compared with 22°C, there was a 22% decrease in sensible heat loss and 64% increase in evaporative heat loss [147]. The channels of heat loss can also be influenced by food intake [68,69], indicating that the effects of temperature and nutrition can never be entirely separated. In human subjects at 26°C, a high protein compared with an isoenergetic high glucose diet induced increases in sensible and evaporative heat losses of 7 and 39%, respectively [69].

Mechanisms by which energy expenditure increases in the cold must be via shivering or non-shivering thermogenesis and these have been discussed in detail elsewhere [12,61,70,115,138,147,150]. Shivering plays an important role in many species on immediate exposure to cold but, after a period of about 2 weeks, although shivering remains important in large animals, non-shivering thermogenesis in brown adipose tissue is the dominant factor in small rodents [144,151–153]. The extent to which cold-induced non-shivering thermogenesis occurs in human subjects and other large animals remains to be established.

Mathematical modelling has been used recently to assess the components of 24-h thermogenesis in rats which had been living under controlled conditions of energy intake for 2 weeks in the mild cold [61]. In animals at thermal neutrality (28°C), total heat production was accounted for by approximately 82% underlying or "basal" thermogenesis and 18% movement-induced thermogenesis. The underlying component increased by about 20% in the mild cold (21°C) and this was probably due mainly to non-shivering heat production in brown adipose tissue. There was, however, no difference in movement-induced thermogenesis over 24 h, even though the animals moved less at the lower temperature. The possibility is that this reduction in energetic efficiency of movement at 21°C was related to the increased heat loss associated with a change in posture. During periods of prolonged inactivity, an additional form of heat production, termed supplementary thermogenesis, was observed at 21°C. Although this could theoretically be due to shivering or brown adipose tissue thermogenesis, the possibility is that it was related to increased muscle tone. Animals were seen to adopt a cold-defensive posture during periods when this supplementary heat production occurred, similar to the posture adopted by other species living at a low temperature [154]. Supplementary thermogenesis accounted for some 6% of 24 h energy expenditure at 21°C, while the peak value contributed 20% to total heat production.

The metabolic response to a high environmental temperature has been discussed in detail elsewhere [12,115]. Thermal balance can be maintained as long as heat can be lost by evaporation and there is a clear difference between species in their ability to lose heat by this channel. Behavioural strategies become particularly important in those species which do not have a high capacity for evaporative heat loss [154]. If there is an imbalance between heat production and heat loss, body temperature will rise and this itself will cause an increase in metabolic rate because of the Arrhenius–van't Hoff effect. It has, however, been observed that, particularly in small birds, hyperthermia may occur without an increase in metabolic rate [155]. The advantage of such a response is that it will slightly increase the temperature gradient for sensible heat loss.

The effects of thermal environment on energy expenditure interact significantly with those of nutrition. Thus, critical temperature decreases as food intake is increased [146,156,157]. The effects of nutrition are considered in more detail in the following section.

## NUTRITION

Throughout this century, whole-body calorimetry has been used to investigate the influences of nutrition on energy balance in a wide variety of species [7,11,12]. Studies have been carried out on over- and under-nutrition and also in relation to nutritional effects on substrate utilization. In recent years, particular interest has centred on the extent to which the quantity and composition of the diet affect energy expenditure and whether the metabolic response is entirely obligatory or may also be regulatory/facultative/adaptive in nature [158,159].

There is a normal energy cost of processing food and this will increase as energy intake increases, and vice versa. A point which has received much attention is whether all the energy expended as a result of food intake is related to necessary processes or whether there is an additional and quite separate set of processes, the intensity of which increases as energy intake increases. A major difficulty is that there is no direct way of measuring the relative contributions made by obligatory and regulatory components to the total thermogenic effect of food. Attempts at demonstrating the presence of a regulatory component have, therefore, aimed to show a change in metabolic rate which cannot be accounted for by changes in obligatory processes or body weight. Pharmacological means have also been used in an attempt to enhance or block the regulatory component.

An additional problem which has hampered the interpretation of some nutritional studies is that the level of energy intake has a striking effect on critical temperature. Therefore, if metabolic rate is to be measured at thermal neutrality, the ambient temperature would have to be lower in

overfed than underfed subjects. Differences also occur in the channels of heat loss, although these will affect only the partition of total heat loss.

*Overnutrition*

Earlier this century, Neumann [160] and Gulick [161] concluded from studies on themselves that there is an energy-dissipating mechanism, termed luxuskonsumption, which allowed them to maintain a reasonably constant body weight despite changes in energy intake. The overfeeding studies of Miller and coworkers [162,163] lent support to this idea but with the advent of accurate methods for measuring energy expenditure over long periods of time, the evidence now appears less convincing [158,164,165].

It had been postulated that the luxuskonsumption mechanism occurred in some individuals during overfeeding such that weight gain was either zero or much less than would be predicted, and only operated after a period of several days of overfeeding or after a critical amount of additional energy had been consumed [148,166]. However, Dauncey [68] used 24-h direct and indirect calorimetry to show that the increase in metabolic rate after only 1 day of overfeeding was of the same order as that estimated to occur after several weeks of overfeeding. An important finding was that only 14% of the additional 24-h intake of 5400 kJ was lost as heat and the rest must have been stored in the body.

The average increase in 24 h energy expenditure after overfeeding for 1 day was approximately 10% [68]. Figure 5 shows that not only did metabolic rate increase during the day time but it remained elevated at night and for at least 14 h after the last meal. There was, however, a wide range in response between individuals, with one subject showing no change in metabolic rate while in another it increased by 21%. These results are similar to increases of the order of 5–15% in a wide range of overfeeding studies [164], with the increase being related to the extent of overfeeding.

In another study where measurements were also made continuously during a 24-h period, five men were persuaded to eat an additional 8010 kJ $day^{-1}$ for 9 days [167]. On average, 24 h energy expenditure increased by 21%, although in four of the men the rise was close to 14% while in the fifth man the value was 47%. This very marked increase was probably related to increased physical activity in the one subject, a topic which will be considered in more detail in the next section. On average, only a quarter of the extra energy was lost as heat, with the remaining 75% being stored in the body. It was estimated that one-third of the rise in energy expenditure could be explained by a rise in "basal" metabolic rate while the rest was due to the thermic effect of food and the increased cost of physical activity related to weight gain.

It appears that the thermogenic response is dependent on whether the overfeeding is of a mixed diet or of a diet high in fat or carbohydrate [158].

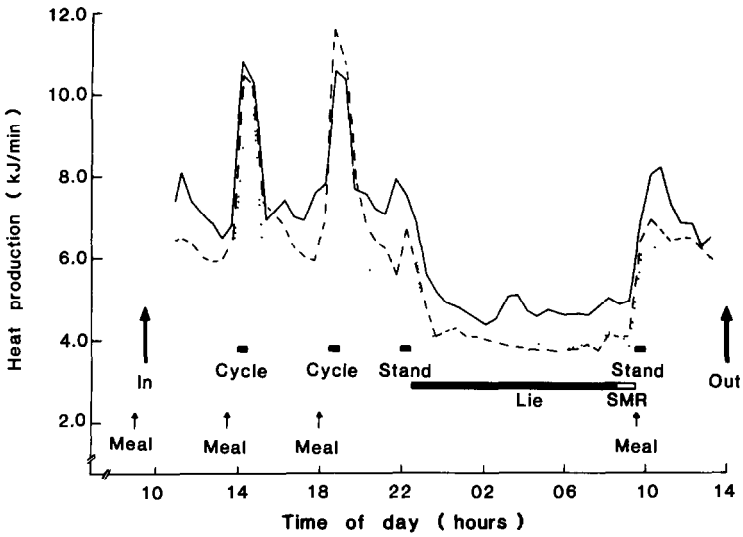


Fig. 5. Influence of over- or under-feeding for only 1 day on the metabolic rate of an adult man during three sessions of 28 h in an open-circuit respiration chamber at 26°C. Energy intakes were high (———), medium (- - - - - -) or low (· · · · · ·) and percentage contributions from protein, fat and carbohydrate were identical. Cycling was at the low workload of 5 N and 50 rpm and standard metabolic rate (SMR) was measured with the subject lying quietly awake. From Dauncey [68].

In general, more of the excess intake is expended as heat when the extra energy is consumed as carbohydrate rather than fat. One week of overfeeding a high carbohydrate diet, mainly as sucrose, resulted in a marked stimulation of energy expenditure, amounting to 33% of the excess intake by the seventh day of overfeeding [168]. This was attributed in part to stimulation of lipogenesis and sympathetic activity, and studies on the effects of $\alpha$- and $\beta$-adrenergic blockade have suggested that the $\beta$-receptor may play an important role in glucose-induced thermogenesis in man [169].

In summary, it therefore appears that no overfeeding studies in man which involve measurement of energy expenditure over prolonged periods, under carefully controlled experimental conditions, support the hypothesis of a luxuskonsumption mechanism in which all or most of the extra intake is lost as heat. Many of the original conclusions were based on studies in which 24 h energy expenditure was not measured accurately and in which environmental temperature and physical activity were poorly controlled. In addition, conclusions were sometimes based on inappropriate interpretation of experimental results. Forbes [170] has shown that the original studies of Neumann [160] and Gulick [161] do not provide evidence of luxuskonsumption. Also, studies in which subjects were overfed for 15 days [171] or 6 weeks [172] were quoted by others as providing evidence for luxuskonsumption, even though the increases in metabolic rate were not great enough to have been this postulated mechanism.

Although a luxuskonsumption mechanism clearly does not appear to operate in man, there is the possibility that such a mechanism can be induced in rodents living at temperatures below thermal neutrality. In 1979, Rothwell and Stock [173] reported that in rats fed a highly palatable and varied "cafeteria" diet, there was an increase in ad libitum intake of 80% without the occurrence of obesity. Comparative slaughter experiments were used to estimate that heat production had increased substantially and it was suggested that this "diet-induced thermogenesis" was a mechanism which tends to maintain zero energy balance during overfeeding.

Numerous investigations have been carried out since this 1979 report [173] to determine whether overfeeding of rodents causes a regulatory/facultative/adaptive "diet-induced thermogenesis" in addition to an obligatory component [174]. For example, indirect calorimetry and comparative slaughter techniques were used to show that energy expenditure is 40% greater in mice fed a high-fat compared with a stock diet, and that nadolol suppresses a large part of this increase [175]. However, because of the difficulty in determining the relative proportions of obligatory and possible regulatory heat production, there has been much controversy regarding the extent to which the total thermogenic response comprises obligatory and regulatory processes [176,177]. A problem has been the comparison between investigations carried out under different conditions, because, for example, the elevated heat production during overfeeding is inhibited at thermally neutral temperatures [178,179].

It has been suggested that this regulatory "diet-induced thermogenesis" in rodents operates via the same mechanism as cold-induced non-shivering thermogenesis, i.e. through sympathetic stimulation of a proton conductance pathway in the inner mitochondrial membrane of brown adipose tissue. The presence of a 32000 $M_r$ uncoupling protein can be used to test for the presence of brown adipose tissue and its thermogenic capacity [180], and the binding of the purine nucleotide GDP is used to estimate its activity [153]. "Cafeteria" feeding has been found to cause changes in brown adipose tissue in rodents, although these are considerably smaller than those induced by a low temperature. The 100-fold increase in the amount of interscapular uncoupling protein in rats at 4°C compared with 29°C is at least an order of magnitude greater than the changes induced by either over- or underfeeding [180].

Even the role of brown adipose tissue in "diet-induced thermogenesis" has recently been questioned. From measurements of the oxygen consumption of the whole body and brown adipose tissue, Ma and Foster [181] concluded that it does not play a major role in the "diet-induced thermogenesis" of "cafeteria"-fed rats. Instead, they suggested a major role for the liver. In addition, other factors such as substrate cycles have been suggested to be of importance in the thermogenic response to food [182].

Because "diet-induced thermogenesis" in rodents is most readily demon-

strated at temperatures below thermal neutrality, it appears that overfeeding simply increases the magnitude of the thermoregulatory response to cold. Thus, it would provide a system for limiting weight gain under circumstances where thermoregulatory thermogenesis already operates [174,183]. However, under natural conditions such a mechanism may seldom be evoked because overfeeding would rarely occur in wild rodents. The size of a rat population is usually limited by food supply and an increase in this supply would rapidly lead to an expansion of the population rather than to prolonged overfeeding.

In man and some domestic animals such as the pig, the evidence for the operation of a regulatory system depending on brown adipose tissue is not strong. If it does occur, the magnitude is considerably less than in rodents. Resting metabolic rate of those on a high energy intake remains elevated for at least 14 h after the last meal in adult man [68,167] and the young pig [113]. Whether any of this increase is regulatory is not certain [158,184]. Nevertheless, the 14% reduction in resting metabolic rate of the young pig by the $\beta$-blocker propranolol is of the same order as the reduction in the elevated metabolic rate due to mild cold exposure [113]. Whether this is a direct action on brown adipose tissue [185], or an indirect effect on thyroid hormone metabolism, muscle tone or some other mechanism remains to be established.

In conclusion, this section clearly highlights the need for more experimental work, including measurements of 24 h energy expenditure under carefully controlled conditions, not only of temperature and diet, but also of physical activity. Calorimetry undoubtedly has a further part to play in elucidating problems related to obligatory and regulatory heat production during overfeeding.

*Underfeeding*

When energy intake is restricted, the resting metabolic rate tends to decline. This decline is greater than would be predicted from tissue loss alone and underfeeding causes not only a loss of metabolically active tissue but also in the metabolic activity per unit weight of tissue. In the short-term, the predominant factor is probably the decrease in metabolic activity [186] while in the long-term, the loss of tissue becomes important [187,188].

Studies with whole-body calorimetry have shown that reducing the usual energy intake for only 1 day causes a significant reduction in 24 h energy expenditure [68]. However, by contrast with overfeeding, the effects of underfeeding are most striking during the day time, while at night time there is no significant effect on metabolic rate (Fig. 5). Because there is no effect on resting metabolic rate measured 14 h after the last meal, the effects of a reduced energy intake on the post-absorptive metabolic rate must take several days to become established. The extent to which there may be a

suppression of the sympathetic nervous system during underfeeding analogous to an activation during overfeeding has been the subject of some investigation. There is, however, a discrepancy between different investigations, with some demonstrating a decline in activity of the sympathetic nervous system [189,190] though others find no such suppression [191].

Individual variation in the metabolic response to underfeeding is wide, with some subjects showing little change in metabolic rate while others show a substantial decrease [68]. Although such a decrease would be advantageous in times of chronic food shortage, it could also be a problem in obese individuals on a weight-reducing diet (the fall in metabolic rate would result in a reduced requirement for food). Interestingly, there is a marked reduction in "basal" metabolic rate during the first 6 months of pregnancy in both Gambian and British women [192]. This adaptation is not related to a low food intake but probably acts as a mechanism for conserving energy supplies which can then be used during lactation.

There is currently much interest in the extent to which individuals can adapt to long-term food shortage [193–195]. Minghelli et al. [196] recently found that in Gambian men a low "basal" and sleeping energy expenditure, reduced thermic response to food and high work efficiency allowed them to cope with a marginal food intake during the "hungry" season. However, even this aspect of thermogenic control is not clear-cut: Garby et al. [197] used direct calorimetry to show that 12 weeks of underfeeding in "normal"-weight subjects did not result in a significant change in 24 h energy expenditure.

Wild animals are seasonally subjected to food scarcity and in the American badger (*Taxidea taxus*) this is combined with severe cold winters. Starvation rapidly becomes a problem at these low temperatures because thermoregulatory demand is high. Whole-body indirect calorimetry was therefore used by Harlow [198] to investigate possible metabolic adaptations of adult badgers during a 30-day fast. After this time, a reduction of 26% in total metabolism was observed, and this was accounted for by reductions in the energy requirements for maintenance and physical activity of 44% and 37%, respectively. About 15% of the reduction in maintenance metabolism was attributed to a decline in deep body temperature; a finding similar to that observed in young pigs living in the cold on a low energy intake [146]. These adaptive responses all allow the badger to conserve energy, thereby increasing the chances for survival during winter food shortage.

Another species which experiences food shortage is the Amazonian manatee (*Trichechus inunguis*). This herbivorous marine mammal normally eats a diet low in metabolizable energy and, in addition, may undergo periods of fasting during the dry season. The low "basal" metabolism of the manatee [199,200] is probably one adaptation to a limited energy supply. Gallivan and Best [200] used respiratory masks to show that after a 14-day fast there was a 22% reduction in metabolic rate but no change in weight-

specific metabolic rate. Whether a decrease in the latter parameter occurs during a longer fast remains to be investigated. As with the badger, there was a reduction in physical activity and this is considered in the next main section.

*Nutrient composition*

Since the time of Rubner [6] and Lusk [7] it has been recognized that the composition of the food eaten can affect metabolic rate. It was demonstrated, for example, that a high protein intake elevated the metabolic rate after feeding in the dog. More recent studies have shown that in the young pig fed a high energy intake, the resting metabolic rate remains elevated for 12–20 h when the diet is high in protein whereas a high glucose diet has no such effect [113].

In the early 1970s it was suggested that man did not respond to diets of different nutrient composition in a way similar to that of other animals [201]. At that time there were indeed no clear-cut investigations on feeding isoenergetic meals of different protein and carbohydrate content to human subjects. However, Pittet et al. [202] then reported that amino acids had a greater effect than glucose on the resting metabolic rate during the 150 min following a test meal.

The maximum probable effect of nutrient composition on 24 h energy expenditure was later investigated by Dauncey and Bingham [69]. Subjects consumed 10 330 kJ per 24 h with either a high protein (37%) or high glucose (40%) content during two separate sessions in a whole-body calorimeter. The 12% increase in 24 h energy expenditure on the high protein intake was highly significant. All subjects exhibited this response and an example of the results is given in Fig. 6. This shows that there was a difference in metabolic rate during the day time and first part of the night. However, by 14 h after the last meal there was no significant effect on resting metabolic rate. The finding of no effect of the protein intake of the preceding day on "basal" metabolic rate has since been confirmed by Soares et al. [203]. Mechanisms accounting for the elevated energy expenditure on a high protein compared with a high glucose diet have been discussed elsewhere [69].

There appears to be some disagreement about the extent to which a high fat compared with a high carbohydrate diet may affect metabolic rate. In a recent study using the Douglas bag technique, no relation was found between meal composition and the thermic effect of feeding [204] and this result was discussed in relation to those of other studies. Undoubtedly some discrepancies arise because of differences in experimental conditions, measurement techniques, state of energy balance of the individual, and type of carbohydrate supplied.

There do, however, appear to be clearly documented differences in the thermogenic responses to different carbohydrates. The resting metabolic rate

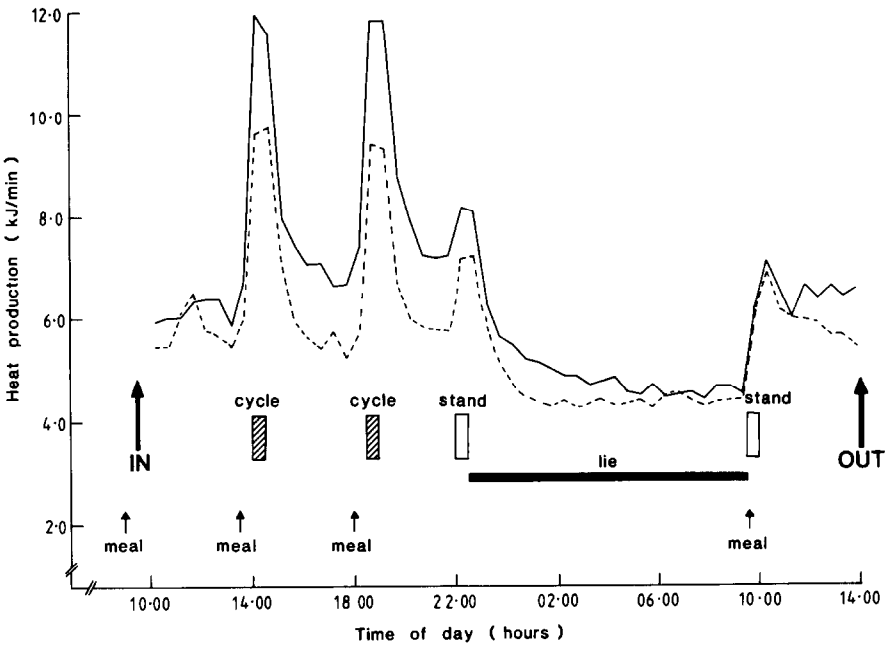


Fig. 6. Influence of nutrient composition on the metabolic rate of an adult man during two sessions of 28 h in an open-circuit respiration chamber at 26°C. Total energy intakes were identical and diets had either a high protein (———) or high glucose (- - - - -) content. Cycling was at the low workload of 5 N and 50 rpm. From Dauncey and Bingham [69].

of the young pig 12–21 h after feeding is greater after a meal high in sucrose or glucose polymer with five hexose units, compared with a high glucose intake. However, it is considerably less than after a meal of complex carbohydrate [55]. Moreover, in man, when fructose is the only carbohydrate source of a mixed meal, the thermic effect of the meal is greater than when glucose is the sole source of carbohydrate [205].

## PHYSICAL ACTIVITY

The muscular movement associated with physical activity can have a significant impact on 24 h energy expenditure [70]. Activities to be considered range from incidental activity due to muscle tone and "fidgeting", to sitting and standing, walking and running, and major activities such as exercise in man and flying in birds. The energetics of muscular work has been discussed in detail by Blaxter [12] and aspects of recent research on the influence of muscular activity and exercise on energy expenditure have also been reviewed [206–208].

Evidence from direct and indirect calorimetry suggests that differences in incidental muscular activity could readily alter 24 h energy expenditure by as much as 20%. It is therefore likely that the individual variability in

metabolic rate is due not only to differences in resting metabolism and the thermic responses to temperature and nutrition but also to differences in minor activity. There is a significant reduction in spontaneous activity during undernutrition in man [209] and wild animals [198,200]. Results for overfeeding studies are less clear-cut, but overnutrition may result in an increase in spontaneous activity and, after a prolonged period, the increase in body weight will certainly cause an increase in the energetic cost of physical activity. The possibility is that differences in the duration or cost of spontaneous activity could help to explain some of the discrepancies between overfeeding studies [70,167,171,210,211]. Even when major differences in activity are not observed, small differences in muscle tone and minor movement could also partially explain individual differences in response to overfeeding.

Environmental temperature also has a significant impact on spontaneous activity. At low temperatures, animals are often seen to adopt a cold-defensive posture; a response which will conserve energy by reducing the surface area available for heat loss [154]. In mice, for example, it has been found that less time is spent in spontaneous activity at 8° C compared with 33°C [212]. Even mild cold exposure can cause a significant reduction in movement. A recent investigation of rats living at 21°C (mild cold) or 28°C (thermal neutrality), on a controlled energy intake, showed that there was a significant reduction in movement at the lower temperature [61]. It was, however, energetically less efficient and there was no difference in 24-h movement-induced thermogenesis, which accounted for 18 and 15% of total heat production at 28 and 21°C, respectively. In addition, a supplementary form of thermogenesis was observed in animals at 21°C which occurred during periods of prolonged inactivity. Although this may have been due to shivering or brown adipose tissue thermogenesis, an alternative possibility is that it was due to an increase in muscle tone caused by maintenance of a cold-defensive posture. Differences in muscle tension may also help to explain part of the increase in metabolic rate in man during cold exposure [70,147,213].

Differences in spontaneous activity may be a significant factor in the development and maintenance of obesity. Studies of particular relevance have been carried out both in the genetically obese (*ob/ob*) mouse and in human subjects. Whole-body calorimetry has been used to show that young "pre-obese" *ob/ob* mice weighing only 16 g are significantly less active than their lean littermates of the same body weight [62]. Figure 7 shows that for 24-h energy expenditure to increase in the young *ob/ob* to the same value as that of the lean animals, the component due to rest would have to increase by about one-third and the activity component would have to double [70]. This finding is in accord with a study in man in which normal weight children with obese parents had a reduction not only in total energy expenditure and resting metabolism, but possibly also in physical activity [214].

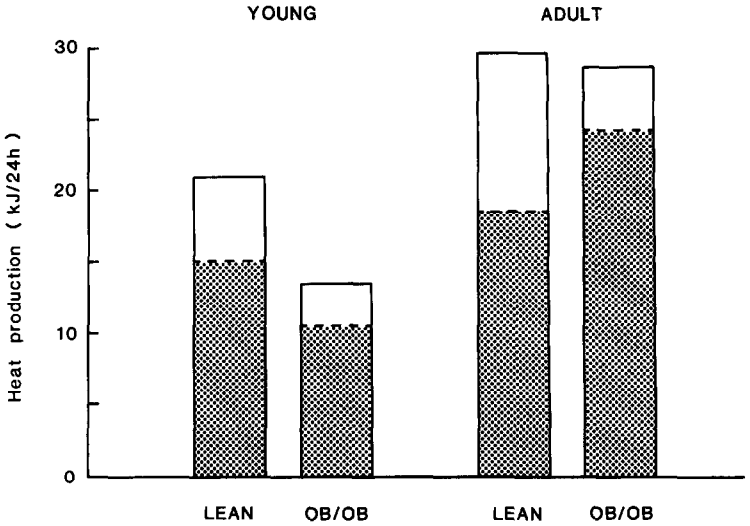


Fig. 7. Twenty-four hour energy expenditure divided into the contributions made by rest (▨) and activity (□) in six pairs of lean and *ob/ob* littermate mice at 28°C. Measurements were made in young animals before there was any difference in body weight and again when they were adult, after differences in body weight had developed. From Dauncey and Brown [62] and Dauncey [70].

After the development of obesity, the adult *ob/ob* mouse is considerably less active than its lean littermate (Fig. 7) [62,215]. In lean and obese mice weighing on average 28 and 54 g, respectively, activity-induced thermogenesis accounted for 11.2 and 4.6 kJ per 24 h in the two groups [62]. It has not been possible to carry out investigations on human obesity under such carefully controlled conditions. Nevertheless, studies using whole-body calorimetry and the doubly labelled water method have allowed the partition of free-living energy expenditure of lean (58 kg) and obese (88 kg) women into two components: "basal" metabolism and the thermic effects of food plus activity [216]. Both these components were higher in the obese women but were identical to the lean when corrected for differences in total body mass and fat-free mass. The findings on resting metabolism agreed with those for the *ob/ob* mouse [62] and with earlier studies on human obesity [217]. The results in relation to activity were, however, different from those in the obese mouse [62]. Apart from the obvious species difference, this could be because of differences in degree of obesity and genetics. As pointed out by the authors [216], their results should not be extrapolated to grossly obese people or those who become obese in childhood. A further possibility is that there is a marked difference in the relative activity levels of the two types of lean subject. Thus, in affluent societies there tends to be a very low level of activity in both lean and obese women and this trend towards exceptionally low levels of activity in many people is potentially a serious problem in relation to the development of obesity [70,108,218].

In man there is only a slight increase in metabolic rate when the posture changes from lying to sitting, whereas there is a much larger increase from sitting to standing [70]. Individual variation in the energy cost of standing is particularly wide because of the different types of posture which can be maintained [12]. In quadrupeds of the same body mass as man, there tends to be a smaller cost of standing, and in the horse, Winchester [219] found no increase in heat production on standing.

Over the last 20 years, significant progress has been made in our understanding of the energetics of walking and running [12,220]. Studies have been carried out in a wide variety of species including hedgehog, lion, quail, and ostrich [211,221,222]. Of particular significance have been the recently reported findings on the energy cost of movement which suggest that an increased economy is achieved by a reduction in the energetic cost of generating force, for example by straight-legged walking and tendon elasticity, rather than by improving the efficiency of work [223,224]. Kram and Taylor [224] have shown that there is a simple inverse relation between the energy cost of running and the period during which the foot applies force to the ground. These results therefore support the hypothesis that it is mainly the cost of supporting the body and the time course of generating force that determine the cost of running [225].

Investigations on the energy cost of walking in man have indicated that, within certain limits, extra loads can be carried without any measurable increase in energy expenditure [226,227]. Mechanisms accounting for this paradox have yet to be elucidated. However, since "head-carry" appears to be a particularly efficient way of transporting loads, and since the load can be either external or internal as body fat, the mechanism may depend on the load being evenly distributed about the vertical body axis.

An important consideration in relation to activity is that the heat it produces can substitute for thermoregulatory thermogenesis. This can occur during mild and severe cold exposure and has been observed in many species including mice, rats, and man [70,147,212,228,229]. Of particular importance, therefore, is the fact that the apparent cost of activity or exercise will be less in the cold than in the warm.

## FUTURE DIRECTIONS

Although whole-body calorimetry has been used since the turn of the century for numerous investigations in man and other animals, it continues to play an essential role in increasing our understanding of energy metabolism in health and disease.

The previous sections have highlighted a number of outstanding problems. There is still considerable uncertainty about the energy requirements of individuals when living under different conditions of food availability and climatic environment. In addition, much remains to be discovered about the

extent to which individuals may adapt to over- and undernutrition at different ambient temperatures, by changes in resting metabolic rate and physical activity. Studies on these aspects would have vital implications for the survival and optimal health of human and animal species. They would also be highly relevant to the efficient utilization of energy supplies in our domestic animals.

One area of current interest is the use of indirect calorimetry for determining substrate utilization and nutrient balance [230–232]. Measurements of oxygen consumption, carbon dioxide production and urinary nitrogen excretion allow calculation of the fuel mixture oxidized. With a knowledge of the nutrients ingested, the nutrient balance can then be calculated. Despite the limitations of this technique, invaluable studies have already been carried out in lean, obese and diabetic people [230]. Recent investigations have also examined the use of this technique as a non-invasive method for determining muscle glycogen stores [233]. This could be particularly useful for assessing the effectiveness of training and diet in endurance athletes.

In recent years there has been an increasing use of whole-body calorimetry in pre-clinical and clinical investigations. Studies have been carried out with pre-term [234] and small-for-gestational-age [235] individuals, elderly mental patients [236], and individuals with cancer [237], liver disease [238,239], cystic fibrosis [240,241] or growth defects [242]. A clear understanding of the changes in energy metabolism induced by these disorders can only come about with the use of appropriate methodology, controls, and conditions of environment, nutrition and activity as outlined in this review.

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