ORIGINAL ARTICLES

Free-living energy expenditure and behaviour in late infancy

J C K Wells, A Hinds, P S W Davies

Abstract

***Objectives*—The energy requirements of infants are determined by body size, growth rate, and physical activity. Little is known of the determinants of energy expended on activity. The relation between free-living energy expenditure and behaviour was investigated in infants aged 9 and 12 months.**

***Methods*—Total energy expenditure (TEE) was estimated by the doubly labelled water method and fat free mass was estimated from the ^{18}O dilution space. Behaviour was assessed by two 24 hour activity diaries.**

***Subjects*—Thirty four normal healthy infants.**

***Results*—TEE was negatively related to the time spent feeding and negatively related to the time spent upset. Body size, represented by fat free mass, accounted for only 19% of the variation in TEE, whereas the combination of fat free mass and two behavioural variables explained 46% of the variation in TEE.**

***Conclusions*—Behaviour contributed significantly to TEE. The energy requirements of individual subjects in this age group cannot be predicted with accuracy from body size alone.**

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Until recently, energy requirements of infants were based on records of energy intake in healthy populations. This approach led to a rapid revision of international recommendations[1–3] as both parental feeding habits and the body composition in the infant population changed over time. The publication of the World Health Organisation recommendations that energy requirements should be based on measurements of energy expenditure[3] coincided with the large scale application to human study populations of the doubly labelled water method for measuring energy expenditure non-invasively.[4 5] Since then a number of studies have reported the measurement of total energy expenditure (TEE) in young age groups.[6–9] TEE can be combined with an allowance for the energy cost of growth to calculate energy requirements.[5 10] Thus in all age groups energy requirements are increasingly being based on measurements of energy expenditure.[11]

Nevertheless, as with energy intake, the measurement of energy expenditure can only provide a description of, rather than prescription for, apparent optimum levels. The most variable aspect of infant energy expenditure is assumed to be physical activity, but there are few data available to support this assumption and the determinants of activity energy expenditure remain unknown. In the absence of such information infant energy requirements remain based on body size[11] or age, which acts as a proxy for body size,[3] in contrast with adults where the contribution of physical activity level is also included.[3] Basing the energy requirements of individual subjects on body size will only be successful if this factor is the main determinant of energy expenditure.

In a previous study we investigated infants aged 3 months, when mobility is poorly developed, and identified several factors which influenced infant activity energy expenditure.[12] In this study we report the investigation of free-living TEE and behaviour in later infancy. Energy expenditure in a sample of infants aged 9 and 12 months was estimated using the doubly labelled water method and infant behaviour was estimated using a parental activity diary. The aims of the study were to identify the aspects of infant behaviour that significantly influence TEE and to distinguish the contributions of behaviour and body size to TEE.

Subjects and methods

A sample of 38 full term infants, 19 aged 9 months and 19 aged 12 months, were recruited using the birth records of the Rosie Maternity Hospital, Cambridge. All infants had been healthy since birth and had no disorder that might adversely influence growth or development. Ethical permission for the study was granted by Cambridge Health Authority and the ethical committee of the Dunn Nutrition Unit, Cambridge.

Measurements were made in the homes of the infants over an eight day period. On day 1 nude body weight (Seca scales, CMS Weighing

Ltd, London), supine length (Rollametre, Raven Equipment Limited, Dunmow, Essex), and skinfold thicknesses (Holtain, CMS Weighing Ltd, London) at the triceps and subscapular sites were measured. Weight was measured again on day 8, with weight measurements being accurate to 20 g.

TEE was measured using the doubly labelled water technique.[6 13] In this technique two stable, non-toxic isotopes of water (^{2}H and ^{18}O) are administered. These isotopes equilibrate with the body water pool and act as tracers of water turnover (^{2}H) and the turnover of water and carbon dioxide combined (^{18}O). The difference between the disappearance rates of the two isotopes is equivalent to carbon dioxide production rate, which can be used to calculate energy expenditure using an assumed respiratory quotient[14] and Weir's equation.[15] Values for energy expenditure represent an average daily value for the eight day measurement period. The technique has been validated in infants against classical indirect calorimetry.[16–18]

The details of our methodology have been described previously.[19] Briefly, urine samples were collected daily for seven days after the administration by mouth of a dose of $^2H_2{}^{18}O$. Doses were intended to give 0.28 g ^{18}O and 0.10 g ^{2}H per kilogram body weight. Urine samples were obtained by leaving cotton wool balls in the nappies of the infants during the daytime, but not during the night. The parents were asked to check the nappy frequently for urination and the time of voiding was taken as the midpoint between the last two times of checking.[20] In the calculation of energy expenditure the fractionation factors were those derived by Lifson *et al* in their original experimental work[21] and the proportion of water subject to fractionation was taken to be 0.15.[22] The respiratory quotient was assumed to be 0.85 based on measurements of food quotients in approximately 200 infants.[14]

Isotope dilution spaces were calculated by the back extrapolation method.[23] This approach predicts isotopic enrichment immediately after dosing from the subsequent washout of each isotope and thus avoids any assumptions about the time at which isotopic equilibration with total body water is reached. Both ^{2}H and ^{18}O exchange with non-aqueous exchangeable hydrogen or oxygen, such that the isotope dilution space is slightly greater than the total body water itself. This difference has been estimated to be 1% for ^{18}O,[24] so the oxygen dilution space was divided by 1.01 to give a value for total body water. This value was then adjusted for the water content of lean tissue to give fat free mass, using age and sex specific hydration values from the reference child of Fomon *et al.*[25] These values were 79.3 and 79.0% (boys) and 79.0 and 78.8% (girls) at 9 and 12 months, respectively. The hydration of lean tissue is known to be higher in early life and to decrease subsequently towards the adult value.[25] Experimental work on 1 and 4 month old infants produced values similar to those given by Fomon *et al*, with low between-subject variability.[26] It is therefore unlikely that substantial errors are introduced to fat free mass by using these assumed hydration factors.

Table 1 Characteristics of the 34 infants at recruitment

Mean (SD) birth weight (g)	3560 (410)
Mean (SD) gestational age (weeks)	40.0 (1.4)
Mean (SD) maternal age (years)	31.8 (3.3)
No of boys/girls	17/17
No breast fed/formula fed	27/7
Parity	
1	12
2	13
3	5
4	4
Social class	
1	11
2	8
3	11
4	4
Delivery	
Normal	22
Forceps	7
Caesarean section	5

Behavioural activity was assessed using a parental activity diary based on a published version[27] which has previously been used to investigate energy expenditure in infants.[12] The diary distinguished five categories of behaviour, namely: sleep; awake and quiet; awake and active; upset; and feeding. Parents were asked to complete the diary over two 24 hour periods during the study week and to avoid unrepresentative days such as those with long car journeys or vaccinations. Infant activities were filled in on the chart as they occurred, with a resolution time of 15 minutes. Infants were classified as awake and active if they were crawling, walking, showing vigorous arm movements, or vocalising in an excited state. They were classified as awake and quiet if they showed minimal motor movement and were subdued. Parents were instructed to select whichever category of behaviour best described each 15 minute period of the 24 hour period.

TEE was regressed on fat free mass and weight to determine the proportion of TEE variation which could be attributed to body size. Multiple regression analyses of TEE were also undertaken, with the independent variables being in each case firstly categories of behaviour and secondly fat free mass, to adjust TEE for body size.

Results

Table 1 gives the characteristics of the infants at the time of recruitment. All the tables refer to the 34 infants in whom doubly labelled water measurements were successful. The four unsuccessful measurements were due to poor parental compliance in collecting daily urine samples.

Table 2 gives the results of anthropometric measurements and doubly labelled water measurements. The correlation between fat mass and the sum of two skinfold thicknesses was 0.44 ($p < 0.01$). Table 2 also gives the results of the activity diary. Table 3 gives the correlations between day 1 and day 2 records of the diary and table 4 gives the internal correlations of the activity diary.

Preliminary analysis indicated that neither behavioural measurements, nor body composition, nor TEE per kilogram body weight

Table 2 Age, anthropometry, energy expenditure, and behaviour; values are mean (SD)

Age (months)	10.7 (1.5)
Weight (kg)	9.84 (1.33)
Length (cm)	75.2 (3.6)
Triceps skinfold (mm)	9.2 (2.5)
Subscapular skinfold (mm)	6.4 (1.7)
N_o (l)	5700 (830)
N_d (l)	5900 (930)
k_o	0.238 (0.036)
k_d	0.192 (0.032)
Fat free mass (kg)	7.16 (1.10)
TEE (kJ/day)	3090 (600)
TEE (kJ/kg/day)	320 (60)
TEE error (%)	6.5 (3.9)
Sleeping (hours/day)	12.9 (1.1)
Awake and quiet (hours/day)	1.9 (1.2)
Awake and active (hours/day)	6.8 (1.8)
Upset (hours/day)	0.5 (0.4)
Feeding (hours/day)	1.9 (0.6)

N = dilution space; k = rate constant, for deuterium (d) and oxygen (o); TEE = total energy expenditure.

Table 3 Consistency and agreement between day 1 and day 2 records from the activity diary

Variable	*r Value*	*p Value*	*SD of difference (hours)*
Sleeping	0.19	0.279	1.75
Awake and quiet	0.56	< 0.001	1.28
Awake and active	0.62	< 0.001	1.68
Upset	0.61	< 0.001	0.55
Feeding	0.80	< 0.001	0.43

r = correlation between day 1 and day 2 values; SD = standard deviation of the difference between day 1 and day 2 values.

Table 4 Internal correlation coefficients for the activity diary

	Awake and quiet	*Awake and active*	*Upset*	*Feeding*
Sleeping	−0.08	−0.47**	0.06	−0.27
Awake and quiet		−0.72**	0.16	0.12
Awake and active			−0.45*	−0.29
Upset				0.03

* p < 0.01; ** p < 0.005.
Correlations between all possible combinations of pairs of behaviour categories indicate whether one category is significantly associated with another category, and whether this association is positive or negative.

differed significantly between the 9 and 12 month old infants. The results of the two age groups were therefore combined and table 5 gives the results of the regression analyses. The time spent feeding and the time spent upset were both inversely related to the TEE. On its own fat free mass explained 19.7% of the variation in TEE and body weight explained 18.7%, whereas the combination (sum of partial r^2 values) of fat free mass and the time spent upset and feeding explained 46.1% of the variation in TEE.

Table 5 Regression of total energy expenditure (TEE) on fat free mass and behaviour for the 34 infants

	Partial R^2 value	*Regression coefficient*	*p Value*
Fat free mass	19.7	241	0.009
Fat free mass	21.1	251	0.007
Sleeping	3.3	99	0.259
Fat free mass	19.6	241	0.010
Awake and quiet	0.1	−16	0.844
Fat free mass	17.8	231	0.012
Awake and active	2.8	57	0.296
Fat free mass	27.0	287	0.001
Upset	27.5	−792	0.001
Fat free mass	12.6	197	0.019
Feeding	16.3	−398	0.008
Fat free mass	19.2	249	0.001
Upset	19.0	−681	0.001
Feeding	7.9	−286	0.029

TEE (kJ/day) regressed on fat free mass, or on combinations of fat free mass and behaviour.

Discussion

In adults it is generally accepted that energy requirements are determined firstly by basal metabolism, which incorporates the effects of body size and body composition, and secondly by physical activity level. Such an approach is utilised by current international energy requirement recommendations[3] and is practicable because the energetic costs of a wide range of adult activities have been studied. In infants and children energy is also required for growth, but by late infancy the proportion of energy intake utilised in growth is low compared with that required for physical activity. Little is known about the relation between energy expenditure and physical activity in infants, however, due to the practical difficulties of measuring free-living energy expenditure in this age group which have only recently been surmounted.

Physical activity in infants and children can be conceptualised in various ways—for example, in terms of energy expenditure, behaviour, mood, or movement. For energy metabolism studies, focusing on energy balance or energy requirements, the quantity of significance is the total daily activity energy expenditure, whatever its cause. There are likely to be variations between subjects in the extent to which the different aspects of physical activity contribute to activity energy expenditure, however. Furthermore, different aspects of physical activity may be important at particular developmental time points.

In a previous study of 12 week old infants we reported that the main aspect of behaviour influencing energy expenditure was the time spent sleeping, which was inversely related to the activity energy expenditure.[12] Other important factors were irritability, positively related to energy expenditure, and the number of siblings, negatively related to activity energy expenditure, as has been reported previously.[28 29] In the present study we sought to explore similar relations in older infants and to distinguish the relative contributions of body size and behaviour to TEE. Although TEE includes basal metabolism and growth costs, it is a more precise measurement than activity energy expenditure, which includes measurement errors from several different methodologies.[30] Thus providing an adjustment is made for body size, TEE is a good index of energy expenditure on physical activity. Furthermore, in late infancy growth costs are relatively low,[25] such that most TEE, after adjustment for body size, can be attributed to activity energy expenditure.

A limitation of our methodology was that although energy expenditure was averaged over a seven day period, behaviour was recorded for only two days within this longer period. This approach was used because of the importance of obtaining 24 hour records of free-living infant behaviour, which only the parents could achieve.

Shorter, more detailed, measurement periods would not necessarily be indicative of the total daily energy expenditure, particularly if an observer from outside the family was involved. Given that the parents were also assisting with the doubly labelled water method by collecting a daily urine sample, we limited the diary to two 24 hour periods. Correlations between day 1 and day 2 diary records indicate consistency in most categories of behaviour, but not for time spent sleeping. More importantly, day to day variability was lowest in the two aspects of behaviour which successfully predicted energy expenditure. Longer periods of observation would be expected to show stronger associations between behaviour and TEE, but we nevertheless were able to show important effects.

We found no significant relation between energy expenditure and the time spent awake and active, or awake and quiet in the 9 and 12 month old infants. This may be due in part to our methodology: the diaries distinguished quiet and active periods, whereas a continuum between extremes of activity might have been a more suitable model. It is possible that the parents found applying the dichotomy difficult, but correlations between the records of the two 24 hour periods were highly significant for both aspects of behaviour, indicating that the infants showed consistency in their behaviour, or at least the parents in their reporting. It is therefore more likely that the distinction between being 'quiet' and 'active' is too simplistic to relate successfully to the total daily energy expenditure. Also, the three categories of sleeping, awake and quiet, and awake and active all showed high day to day variability even if the parents ranked the infants consistently. Thus for these categories of behaviour it is probable that two day records were insufficient to overcome day to day variations within subjects and could not give representative daily values.

The remaining two categories of behaviour showed lower day to day variability and high consistency in parental rating, however. The time spent feeding was negatively related to the TEE. This may at first appear to be paradoxical, given the close relation between energy intake and energy expenditure in this age group. We speculate, however, that active infants may feed quickly and inactive infants more slowly. The negative relation between the time spent feeding and the time spent awake and active, although of moderate magnitude, is consistent with this hypothesis.

The time spent upset was highly significantly associated with reduced TEE and was the strongest behavioural predictor of TEE. Again, this result initially appears to confound expectations, as greater distress in younger infants was associated with increased energy expenditure.[12] We suggest that there may be two possible explanations for this relation. Firstly, the relation could actually represent a link between energy intake and distress, indicating that upset infants might be difficult to feed and therefore have both a lower energy intake and less energy to expend on physical activity. Internal correlations of the activity diary suggest this was not the case, however, as the time spent upset showed no association with the time spent feeding (r = 0.03; NS). Furthermore, there was no relation between the time spent upset and the percentage of fat (r = −0.09; NS) or skinfold thickness (r = 0.10; NS), which might be expected if energy intake was significantly reduced.

An alternative possibility is that infants spending more time upset are less active. Internal correlations of the activity diary show that infants spending more time being upset spent significantly less time awake and active (r = −0.45; p = 0.005), whereas the time spent upset did not correlate with the time spent being quiet or the time spent sleeping. Thus being upset appears to reduce the time available for more active behaviour.

Our data indicate that the size of the metabolically active tissue, represented here by fat free mass, explains less than 20% of the variation in TEE. This is negligibly better than predicting TEE from body weight, as is currently the practice when using contemporary national guidelines[11] to estimate the energy requirements of subjects in younger age groups. It should be noted that the fat free mass includes both organ tissues, with a high metabolic rate, and muscle tissues, with a low metabolic rate. Intersubject variation in the ratio of organs to muscles is not taken into account if fat free mass or weight is used as the index of body size. It is unlikely, however, that this explains the poor ability of fat free mass to predict TEE in this age group. A more plausible explanation is that infants differ markedly from adults in the determinants of energy expenditure. In adults, both basal metabolism and many activities which involve supporting body weight are strongly related to size. In infants with poorly developed mobility various aspects of energy expenditure (growth rate, non-weight bearing movement, temperament) are minimally related to size. Hence, paradoxically, energy requirements are based on body size in the population where the relation between body size and TEE is weakest.

The combination of two aspects of behaviour was found to contribute significantly to TEE, explaining, in combination with the fat free mass, almost half the variation. This shows the importance of behaviour for energy requirements in this age group and reinforces the argument that the requirements of individual subjects cannot be predicted with accuracy on the basis of body size alone. In the absence of actual measurements, energy requirements for individual infants and children can only be estimated, but our study has shown that such estimations could be improved if behaviour is taken into account. The relation between behaviour and energy expenditure merits more attention in both healthy and unhealthy infants.

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