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Associations between basal metabolic rate and reproductive performance in C57BL/6J mice

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Summary

Basal metabolic rate (BMR) is highly variable, both between and within species. One hypothesis is that this variation may be linked to the capacity for sustained rate of energy expenditure, leading to associations between high BMR and performance during energy-demanding periods of life history, such as reproduction. However, despite the attractive nature of this hypothesis, previous studies have failed to show an association between BMR and fecundity. Our approach was to mate 304 C57BL/6J mice and allow them to wean pups before measuring BMR by indirect calorimetry. We did not find an association between BMR and litter mass, size or pup mass at birth or weaning that could not be accounted for by the body mass of the dam. There was also no relationship between BMR (or BMR corrected for body mass) and birth or weaning success, losses during weaning, or sex ratio. However, a significant relationship was found between BMR and gestational weight loss indicative of foetal resorption. This suggests that during pregnancy the available energy may be limited and partitioned away from the growing foetus and towards maintenance of the mother. In this context, a high BMR may actually be disadvantageous, conflicting with the idea that high BMR may bring reproductive benefits.

Key words: resource allocation, sustained metabolic rate, fecundity, mouse, BMR.

Introduction

Basal metabolic rate (BMR) represents the minimal metabolic rate of an individual when it is inactive, performing no thermoregulatory work and not digesting any food (Kleiber, 1961). Despite the expectation that it would reflect some minimal level of homeostatic cellular processes, BMR shows tremendous variability both between and within species (McNab, 2005; Rezende et al., 2004; Snodgrass et al., 2005), raising two key issues: why does the variation exist and what are the functional implications of the variability? Clearly, an individual with a high BMR would need to feed for longer to fuel its metabolism than one with a low BMR. Alternatively, if both animals fed for the same time, the one with the higher BMR would have less surplus energy available to devote to alternative activities such as reproduction (Gadgil and Bossert, 1970). Previous focus has therefore been on the benefits that individuals with high BMR might derive to offset the potential disadvantages of their high rates of metabolism.

During the late 1970s and early 1980s two similar hypotheses were published about the potential advantages that individuals having high BMR might have. Bennett and Ruben suggested that endotherms maintain high BMRs, as this enables them to achieve substantially higher maximal rates of energy expenditure than ectotherms (Bennett and Ruben, 1979). This has been called the ‘aerobic capacity model’ for the evolution of endothermy (Bennett and Ruben, 1979; Taigen, 1983; Bozinovic, 1992; Hayes and Garland, 1995). Generalising this idea to the level of the individual within an endothermic species, the aerobic capacity model suggests that individuals with high BMRs sustain these rates because they derive an advantage in situations where a maximal rate of energy metabolism is required. The aerobic capacity model links BMR to maximal rates of expenditure over short periods of time. The second hypothesis was that a higher BMR may be related to a higher sustained metabolic rate (SusMR); the maximal rate of metabolism that an individual could sustain over days and weeks (Drent and Daan, 1980). This may also provide a context for the evolution of endothermy (Farmer, 2000; Koteja, 2000). SusMR is presumed to be limited by an animal's morphology. A big gut, and associated organs (such as the liver) that can process energy faster, makes more energy available to support SusMR but also requires a greater amount of maintenance (BMR) (Drent and Daan, 1980; Hammond and Diamond, 1992; Hammond and Diamond, 1997; Peterson et al., 1990; Weiner, 1989; Weiner, 1992). Therefore, an individual with a higher BMR will have greater capacity for SusMR and, if available energy is unlimited, they may be able to sustain greater reproductive output.

A direct prediction of the SusMR hypothesis is that individuals with higher BMRs will have greater capacity for SusMR. One of the most energetically demanding periods in a small mammal's life history is late lactation (Thompson, 1992). Our work on the lactating MF1 mouse (Johnson and Speakman, 2001; Johnson et al., 2001a; Johnson et al., 2001b; Johnson et al., 2001c; Krol et al., 2003; Krol and Speakman, 2003a; Krol and Speakman, 2003b; Speakman et al., 2001; Speakman and Johnson, 2000; Speakman and Krol, 2005; Speakman and McQueenie, 1996) and studies of other lactating small rodents (Hammond and Diamond, 1992; Hammond and Diamond, 1994; Hammond et al., 1994; Hammond et al., 1996; Konarzewski and Diamond, 1995; Rogowitz, 1998; Rogowitz and McClure, 1995) have shown that BMR and SusMR are associated because, under different conditions (e.g. between non-breeding and lactating, or between different groups of lactating mice held at different ambient temperatures), they share a dependence on aspects of morphology that limit the uptake of energy or its utilisation. However, support for the existence of such a link at the individual level is sparse (Daan et al., 1989; Daan et al., 1990). In lactating mice, individual variations in BMR do not correlate with individual variation in organ morphology (Krol et al., 2003; Speakman and Johnson, 2000). Other studies have also failed to find associations between individual variation in morphology and BMR in birds and non-lactating mammals (Burness et al., 1998; Corp et al., 1997; Geluso and Hayes, 1999; Koteja, 1996).

The absence of an association between variations in BMR and morphology is consistent with the large number of studies that have failed to establish the expected links between individual variations in BMR and markers of reproductive output (Derting and McClure, 1989; Earle and Lavigne, 1990; Hayes et al., 1992b; Johnson et al., 2001b; Stephenson and Racey, 1993a; Stephenson and Racey, 1993b). One possible reason for this failure is that previous studies have tended to measure BMR in pre-reproductive animals. Lactating mice show considerable morphological plasticity during lactation relative to the situation when they are virgins, including expansion of their alimentary tracts and liver sizes (Fell et al., 1963; Kennedy et al., 1958) with associated modulations of their resting metabolic rates (Speakman and McQueenie, 1996). Previously pregnant mice have a greater body mass, attributed to protein accumulation, than nonparous mice (Holinka, 1980). Therefore, we would argue that lactating mice never return to their original pre-reproductive form, and their post-reproduction morphology retains a shadow of the lactation experience. Perhaps after lactation, rather than before reproduction, is a more appropriate time to seek links between reproductive performance and BMR, if we are to deepen our understanding of the functional significance of its variation.

In the present study, we set out to determine whether animals with greater BMR can sustain higher levels of reproductive output or if reproduction is sacrificed for maintenance, by measuring the BMR in the post-reproductive period of a large cohort of female C57BL/6J mice (*N*=304). In addition to the usual ways to characterise reproductive output (litter mass and size at birth and weaning), we also set out to measure the association between BMR and the probability that dams will give birth to live pups and successfully wean pups, the offspring losses that occur during gestation and lactation, and the sex ratio of the offspring produced. We found no positive association between post-reproductive BMR and prior reproductive performance. Indeed, females with greater BMR had an increased frequency of gestational body mass losses suggestive of foetal resorption, suggesting that a high BMR was disadvantageous.

Materials and methods

Animals

Female C57BL/6J mice (*N*=304) were purchased in three batches from Harlan UK Ltd, Oxon, UK, at 6 weeks of age, and were housed in groups of six in a 12 h:12 h light:dark photoperiod at approximately 23°C, and in accordance with Home Office regulations. Sawdust and paper bedding were provided for nest building. Animals received an *ad libitum* supply of water and laboratory chow (Rat and Mouse Breeder Grower diet CRM; Special Diet Services, Essex, UK; 12.5 kJ g^{-1} metabolisable energy). At 7 weeks of age, two to three females were housed together with a proven breeder male C57BL/6J mouse, also from Harlan UK Ltd. Females were weighed every second day until birth to monitor pregnancy. After 16 days, males were removed and females were individually housed and monitored daily. Pups were weighed within 18 h of birth and at weaning 21 days later. The number of male and female pups per litter was recorded for 170 litters at weaning. Pups were removed at weaning and females were allowed to acclimatise for 10 days before measurements.

Measurements

Animals were placed on a 30-day measurement schedule where body mass was measured daily for two weeks, and the basal metabolic rate (BMR) of each animal was measured once using an open-circuit respirometry system as described by

Hayes et al. (Hayes et al., 1992a). Briefly, individual animals were placed in sealed Perspex™ chambers in an incubator (Sanyo Gallenkamp PLC, Leicestershire, UK) set to 30°C (within thermoneutral). Fresh air was dried through cylinders of self-indicating silica gel (VWR International Ltd, Dorset, UK) and pumped through the system (Charles Austin Pumps Ltd, Surrey, UK). Mass-flow controllers (MKS Instruments UK Ltd, Cheshire, UK) provided 500–700 ml air per min, which was monitored using Alexander Wright DM3A flow meters (GH Zeal Ltd, London, UK). Air leaving the animal chamber was dried using silica gel and 150 ml min^{-1} was passed through a gas analyser (Servomex Group Ltd, East Sussex, UK). Gas concentrations were measured continuously, and means were calculated every 30 s for 200 min. Values of oxygen consumption and carbon dioxide production were calculated with corrections for temperature and pressure from the 10 lowest consecutive measurements (equivalent to 5 min) using the appropriate equation (Hill, 1972). The BMR (Watts) for each animal was calculated from the respiratory quotient using the Weir equation, as shown by Speakman (Speakman, 2000).

Data analysis

Statistical analysis was carried out using MINITAB® Release 14.1 (MINITAB Inc., State College, PA, USA). The mean body mass (M_b) for the two-week period was calculated and used to correct BMR by plotting M_b *vs* BMR and storing the residuals. Animals were considered successful at birth if they gave birth to live pups and at weaning if they weaned any pups. Binary logistic regression was performed with birth (0 or 1) as the response and either BMR or residual BMR corrected for M_b as the model. The percentage of animals that were successful at birth or weaning was calculated from the number of successes out of the total observations for each centile of BMR, or residual BMR corrected for M_b. Animals that lost some or all pups during gestation were identified by observed deviations from normal body mass curves following pairing with a proven breeder male. Binary logistic regression was performed with pup loss (0 or 1) as the response and either BMR or residual BMR corrected for M_b as the model. The percentage of animals that lost pups was calculated for each centile of BMR or residual BMR corrected for differences in M_b. Least-squares linear regression was used to determine if there were any relationships between litter mass and size at birth and weaning and M_b, and either BMR or residual BMR corrected for M_b. The $\log_e$ ratio of female to male pups per litter was calculated and analysed by simple linear regression for a relationship with either BMR or the residual BMR corrected for M_b.

Results

Animals were, on average, 72 days old at parturition and BMR was measured, on average, 43 days later (Table 1). Of the animals that we measured for BMR (N=304), 87% had successfully given birth and 84% maintained litters to weaning age (Table 1). Our use of proven breeder males makes it unlikely that failure to produce offspring was a consequence of male infertility.

Body mass (M_b) of females that successfully gave birth was greater than unsuccessful females at the time when BMR was measured, but BMR was not different (Table 1). The variation in BMR for successful females was large (Table 1), and although the variation in M_b for the same sample was also great, only 6.9% of the variation in BMR could be accounted for by M_b (Fig. 1). Nevertheless, for successful females, the association was significant because of the large sample size (Fig. 1). However, because M_b accounted for so little of the

Table 1. *Characteristics of female C57BL/6J mice used in this study*

	Birth success	*N*	Mean ± s.e.m.	Range
Age at parturition (days)		265	72±0.2	68–85
Body mass at parturition (g)		265	24.1±0.10	19.8–30
Number of pups born		265	6.4±0.10	2–10
Birth litter mass (g)		265	12.4±0.14	4.4–18.2
Birth pup mass (g)		265	2.0±0.03	1.3–4.6
Number of pups weaned		256	6.1±0.12	1–10
Weaned litter mass (g)		256	54.0±0.76	4.9–83.9
Weaned pup mass		256	8.7±0.06	4.9–12.5
Age at BMR measurement (days)		304	115±0.5	100–135
Body mass at BMR measurement (g)	No	39	22.2±0.25	19.6–28.0
	Yes	265	24.2±0.08*	17.7–26.0
	Total	304	24.0±0.09	17.7–28.0
BMR (W)	No	39	0.185±0.0070	0.083–0.303
	Yes	265	0.194±0.0020^{NS}	0.102–0.299
	Total	304	0.193±0.0019	0.083–0.303

Pups were weighed individually at weaning, but at birth the pup mass was calculated from birth litter mass and the number of pups in each litter. Animals were considered successful at birth if they gave birth to live pups. Successful and unsuccessful animals were compared by unpaired *t*-test. *$P<0.001$, $t=-7.37$; $^{NS}P=0.237$, $t=-1.2$. BMR=basal metabolic rate.

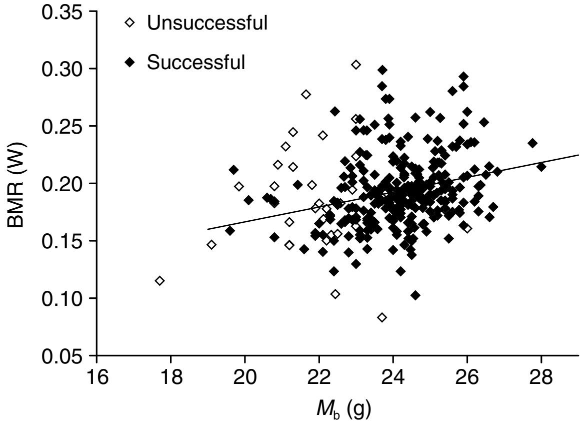

Fig. 1. Scatter plot of basal metabolic rate (BMR) and body mass (M_b) of female C57BL6/J mice during a 30-day measurement period starting 10 days after weaning. The trend-line (y=0.0054x+0.0639) is for females that successfully gave birth to pups (N=265, r^2=6.9%, P<0.001). There was no association between BMR and M_b for unsuccessful females (N=39; r^2=0.3%, P=0.748).

variance, there was a large residual variation in BMR once the effects of M_b had been accounted for. We have presented results concerning BMR as uncorrected BMR as well as residual BMR corrected for M_b effects. There was no association between BMR and M_b for animals that did not successfully give birth (Fig. 1).

Litter size for dams that gave birth to live litters varied at birth between two and 10 pups (Fig. 2). The mean litter size was 6.4 pups and the modal litter size was 7 (Fig. 2). Litter mass at birth was positively related to litter size (Fig. 3A) and ranged from 4.4 g for a litter of two pups to 18.2 g for a litter with seven pups. Although four animals gave birth to 10 live pups, the heaviest of these litters at birth was 16.6 g. Because the relationship between litter mass and litter size was not linear (Fig. 3A) the mean pup mass at birth was negatively related to litter size (Fig. 3B). Hence, pups in the smallest litters weighed on average 3.2 g, while those in litters of seven averaged 1.9 g and those in the largest litters weighed on average only 1.6 g. (Fig. 3B).

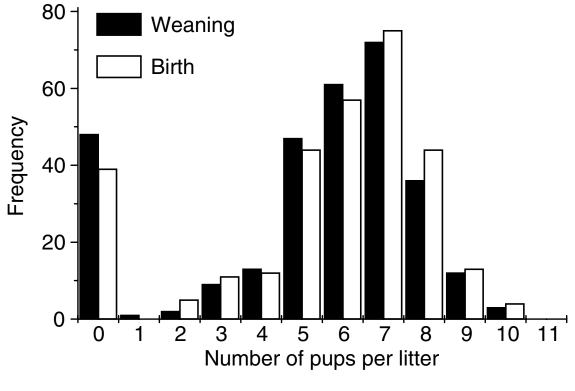

Fig. 2. Histogram showing the number of C57BL/6J mice that gave birth and weaned a given number of pups.

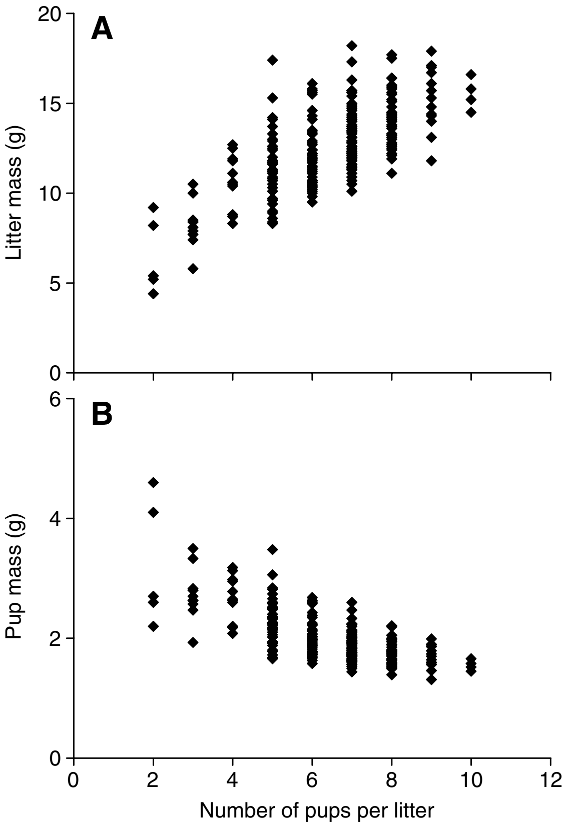

Fig. 3. Scatter plots showing the relationship between the number of pups born in a litter and (A) litter mass and (B) mean pup mass per litter, which was calculated from litter mass and pup number.

We ordered the mothers by either their absolute BMR or their residual BMR corrected for M_b and then divided these ordered data into 10 equal-sized groups. Within each group we calculated the proportion of animals that successfully gave birth to live litters (265 out of 304 mated) and those that went on to wean pups (256 out of 265 that gave birth; Fig. 4). There was no significant association between the probability of birth success and either BMR (Fig. 4A) or residual BMR corrected for M_b (Fig. 4B). From the 265 animals that gave birth to live litters, only nine failed to maintain them to weaning age. Four of the five (80%) litters with two pups did not reach weaning and four out of the 11 (36%) litters with three pups failed. One of the 57 litters with six pups (1.8%) also failed to reach weaning. Failure to maintain pups to weaning was not related to BMR (Fig. 4C) or residual BMR corrected for M_b (Fig. 4D). However, the females that lost their litters before weaning all gave birth to low numbers of pups. These two factors may have been related and may have resulted from some undiagnosed reproductive problem, so these animals were excluded from further analyses.

Many females lost offspring during the course of lactation but did successfully wean some pups. We used contingency tables to test if there was an association between losses and the number of pups per litter at birth. Excluding the animals that lost their entire litters, we found that the extent of the losses

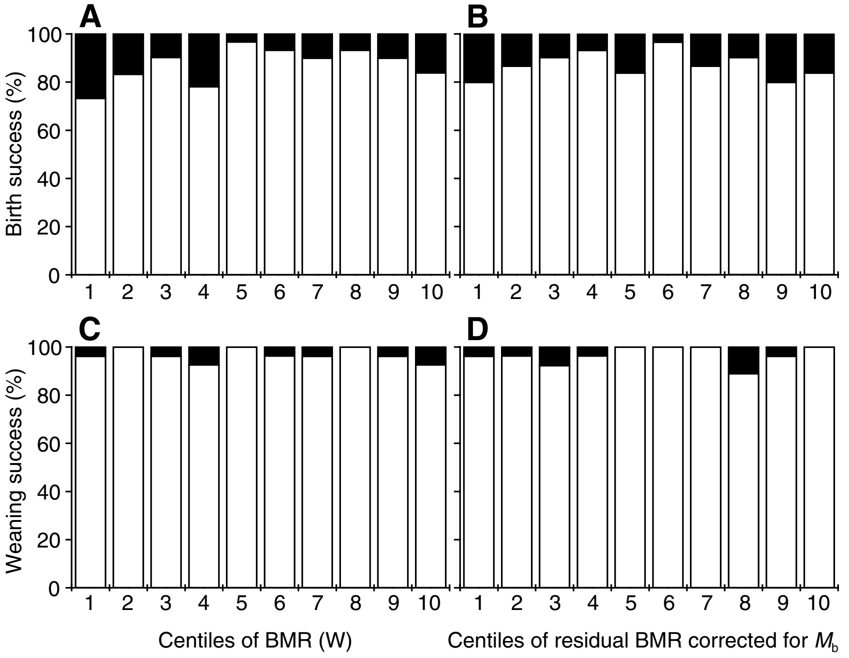

Fig. 4. The percentage of C57BL/6J mice that gave birth to live litters (N=304) and successfully weaned pups (N=265) in each centile of basal metabolic rate (BMR) values or residual BMR corrected for body mass (M_b). There was no significant association between the probability of birth success and either BMR (A; binary logistic regression, P=0.129) or BMR corrected for M_b (B; P=0.754), or weaning success and BMR (C; P=0.954) or BMR corrected for M_b (D; P=0.373).

during lactation was not related to large initial litter size (Pearson chi-squared value=5.66, d.f.=8, P=0.686; data not shown).

There was a positive correlation between litter mass at birth and M_b of females when BMR was measured (Fig. 5A), but there was no relationship between litter mass and BMR (Fig. 5B) or BMR with the effects of M_b removed (Fig. 5C). However, litter size at birth was significantly related to both female M_b (Fig. 6A) and BMR (Fig. 6B). Females that were heavier, or had greater BMR, on average gave birth to larger litters. However, the explained variance in litter size by both M_b and BMR was small (15.3% and 2.1%, respectively) and the gradients of the least-squares fitted regression lines were very shallow. Hence, on average, a female mouse with a BMR of 0.15 W gave birth to a litter of 6.1 pups and a female with a BMR of 0.25 W (67% greater) gave birth to a litter of 6.8 pups (11% greater). Moreover, this effect of BMR on litter size was completely dependent on the shared variation in both traits due to body mass, as there was no significant association between litter size and residual BMR with the effects of mass removed (Fig. 6C).

At weaning, litter mass and litter size both correlated with M_b (Fig. 7A and Fig. 8A, respectively) and BMR (Fig. 7B and Fig. 8B, respectively) and the relationships were again positive. Heavier females and those with greater BMR weaned heavier and larger litters. However, as with the relationships for litter size at birth (Fig. 6), the gradients of the least-squares fitted regression lines were very shallow. Increases in BMR from 0.15 to 0.25 W (67% greater) were on average associated with increased masses at weaning of 6.4 g (12% of mean litter mass at weaning) and 0.8 extra pups (13% of litter size at weaning). As with the relationships at birth, the significance of the BMR effect was entirely dependent on the shared variation due to M_b as there was no relationship when residual BMR taking mass into account was used (litter mass in Fig. 7C and litter size in Fig. 8C).

The monitoring of body mass every second day enabled us to identify 13 animals that were pregnant but were not successful at birth. Two of these animals gave birth, but only dead pups were found, and the remaining 11 animals experienced M_b gain indicative of pregnancy but later returned to normal M_b, suggesting that foetuses were absorbed or miscarried. We also identified that 25 of the animals that successfully gave birth to live litters had M_b loss or plateau during pregnancy, which indicated that they had lost some pups during gestation. We were unable to quantify what the litter sizes might have been for these animals if they had kept all pups conceived. We ordered these 278 (265 successful and 13 pregnant but unsuccessful) animals by either their absolute BMR or residual BMR corrected for BM and then divided these ordered data into 10 equal-sized groups. Within each group, we calculated the proportion of animals that appeared to have absorbed or miscarried foetuses (Fig. 9). We found a significant relationship between the loss of pups during gestation and BMR (Fig. 9A). This indicates that animals with high BMR have an increased probability of losing pups either through miscarriage or the absorption of foetuses. This relationship was independent of the variation found in M_b because the relationship with residual BMR corrected for differences in M_b was still significant (Fig. 9B).

The sex of the pups from 170 litters was recorded at weaning. Two litters contained only male pups and four litters contained only female pups. There was no relationship between the number of female pups weaned and M_b (N=168, r^2=0.9%, P=0.228), but the number of male pups weaned per litter was related to the M_b of the dam at the time when BMR was

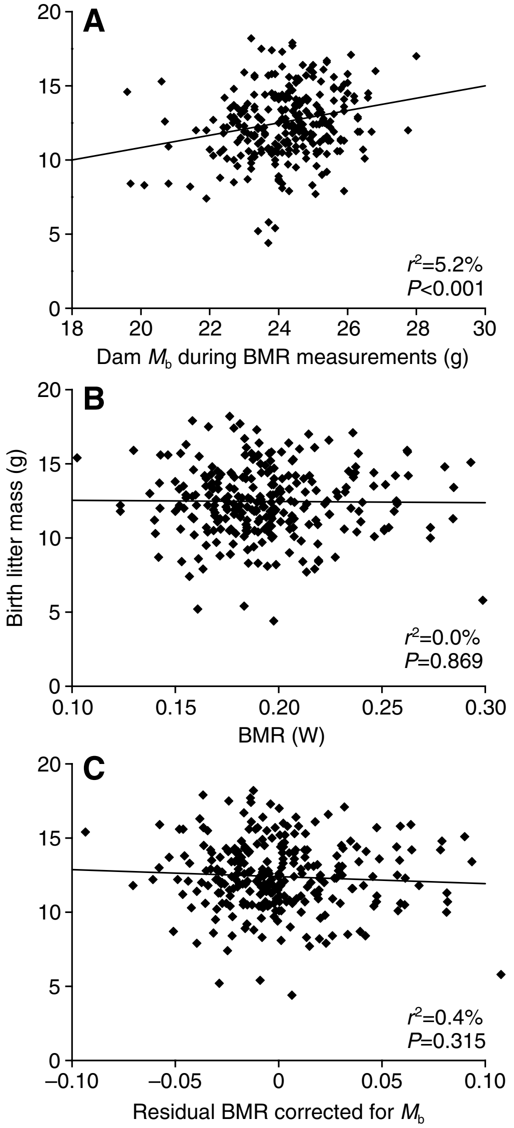

Fig. 5. Scatter plots of litter mass at birth against (A) body mass (M_b) ($y=0.414x+2.41$), (B) basal metabolic rate (BMR) ($y=-0.75x+12.6$) and (C) BMR corrected for M_b ($y=-4.75x+12.4$) for C57BL/6J mice ($N=265$).

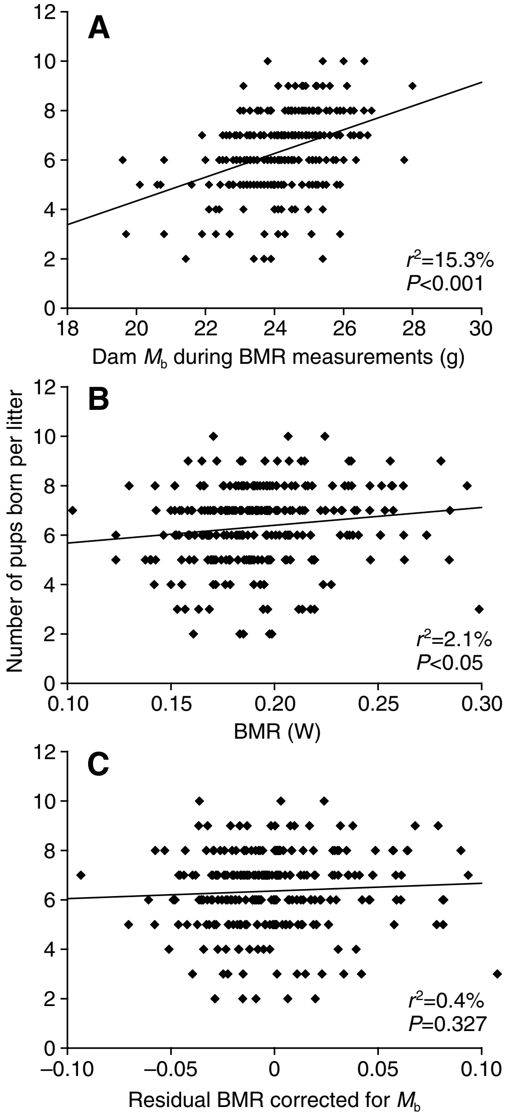

Fig. 6. Scatter plots of litter size at birth against (A) body mass (M_b) ($y=0.48x-5.26$), (B) basal metabolic rate (BMR) ($y=7.2x+4.96$) and (C) BMR corrected for M_b ($y=3.11x+6.36$) for C57BL/6J mice ($N=265$).

measured ($N=166$, $r^2=12.1\%$, $P<0.001$). As pups were not sexed at birth, this could have occurred for two reasons: either dams with high M_b may have given birth to a greater number of male pups or dams with a low M_b may have reduced the number of male pups in litters during lactation. There was no relationship between the number of females or the number of males weaned and BMR ($r^2=1.4\%$, $P=0.122$ and $r^2=0.6\%$, $P=0.322$, respectively; data not shown) or residual BMR corrected for M_b ($r^2=1.0\%$, $P=0.202$ and $r^2=0.0\%$, $P=0.854$, respectively; data not shown). For dams with male and female pups, the ratio of female to male pups was also unrelated to M_b (Fig. 10A) and both BMR (Fig. 10B) and residual BMR corrected for M_b (Fig. 10C).

Discussion

The variation we observed in BMR in this strain of mice was consistent with our previous measures of BMR in mice (reviewed in Speakman and Krol, 2005). Since we have previously determined the repeatability of this trait in our measurement protocols at around 8% (Krol et al., 2003), only a minor proportion of the total variation we observed can be attributed to random error. This variability in domesticated mice is typically larger, and the corresponding relationships between BMR and body mass poorer, than is observed in other small wild rodents (Koteja, 1995; Sadowska et al., 2005). This probably reflects the high variability in the contribution of metabolically inert body components such as adipose tissue to the variance in body mass of domestic mice.

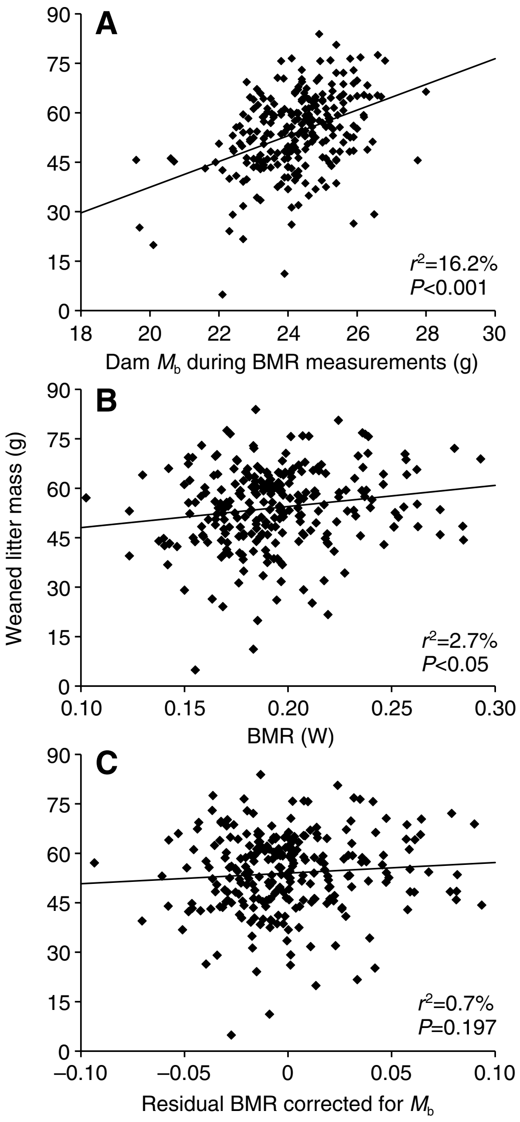

Fig. 7. Scatter plots of litter mass at weaning against (A) body mass (M_b) (y=3.9x–40.6), (B) basal metabolic rate (BMR) (y=63.8x+41.7) and (C) BMR corrected for M_b (y=32.4x+54.0) for C57BL/6J mice (N=256).

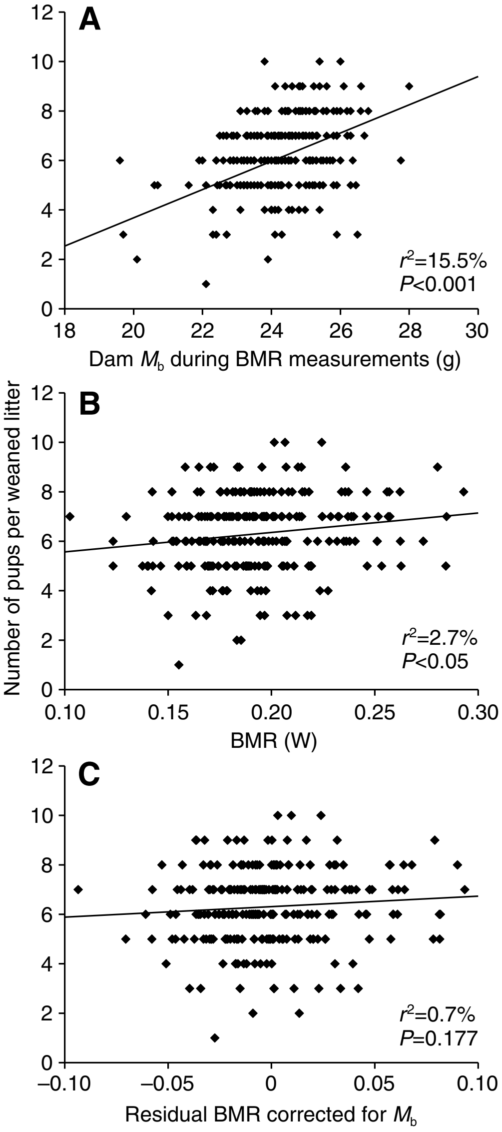

Fig. 8. Scatter plots of litter size at weaning against (A) body mass (M_b) (y=0.571x–7.74), (B) basal metabolic rate (BMR) (y=7.84x+4.79) and (C) BMR corrected for M_b (y=4.24x+6.31) for C57BL/6J mice (N=256).

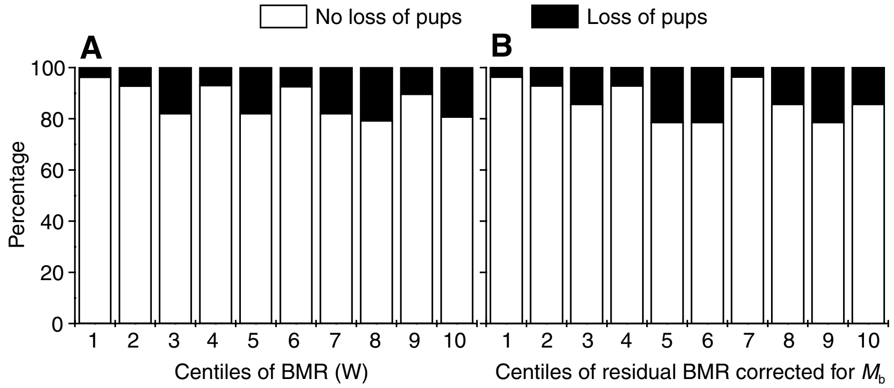

Fig. 9. The percentage of C57BL/6J mice (N=278) that were identified, by changes in body mass (M_b) following pairing with a proven breeder male, to have lost some or all pups by absorption or miscarriage in each centile of basal metabolic rate (BMR) or residual BMR corrected for M_b. Pup loss was associated with BMR (A; binary logistic regression, P<0.05) and BMR corrected for M_b (B; P=0.05).

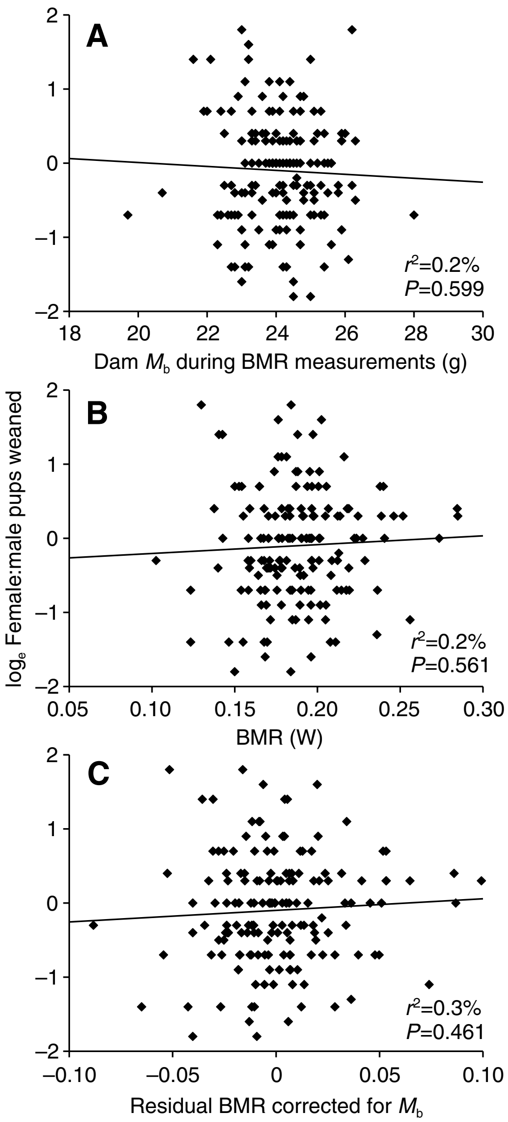

Fig. 10. The ratio of female to male pups at weaning against (A) body mass (M_b) (y=–0.0319x+0.668), (B) basal metabolic rate (BMR) (y=1.49x–0.412) and (C) BMR corrected for M_b (y=1.56x–0.099) for C57BL/6J mice (N=164).

In this study, we set out to characterise the relationship between fecundity and BMR measured after weaning in C57BL/6J mice. Associations between individual variations in BMR and reproductive performance may not have emerged in previous studies because the traits used to reflect reproductive performance (litter mass and size at birth and weaning) appear to be inappropriate performance indicators. In addition to these measurements, we made some additional measures in the present study that might better reflect reproductive capacity. These additional traits were: the probability that the female would successfully give birth; the probability the female would successful wean offspring; the extent of losses that occurred during gestation and lactation; and the sex ratio of the offspring. Previous studies have not reported the variation in these traits nor how they were correlated with BMR. We found that weight loss in the pregnant dam, which we considered to be indicative of foetal resorption, was significantly associated with individual variability in BMR and in the residual variation in BMR with the effects of body mass removed.

Most mammals have evolved to invest nutrients into reproduction across gestation and lactation (matrotrophy). Energy can be diverted away from the growing foetus and towards maintenance at any stage, and the dam can abort or resorb the invested energy if conditions of food availability change (Trexler and DeAngelis, 2003). The inbred strain of mouse used in the current study has been shown to have an increased frequency of resorptions with aging (Holinka et al., 1979). This is consistent with animals allocating energy into somal protection rather than reproduction later in life (Yearsley et al., 2005). The re-evaluation of resources by the dam during gestation may be the reason why we observed a greater number of gestational body mass anomalies consistent with abortion or resorption with increasing BMR. This information supports an inverse relationship between reproductive output and BMR and challenges the theory that life-history demands are driving total energy requirements and thus BMR. However, a more direct method of measuring resorption is required to support the observations in the current study.

By measuring BMR in the period following rather than preceding reproduction we hoped to reveal associations between BMR and reproductive performance that have eluded previous attempts to establish such associations (Derting and McClure, 1989; Earle and Lavigne, 1990; Hayes et al., 1992b; Johnson et al., 2001b; Krol et al., 2003; Stephenson and Racey, 1993a). Despite this different protocol, the relationships between BMR and the parameters of reproductive output commonly used (birth litter size, weaning litter size and weaning litter mass) were all exceedingly weak and involved BMR explaining considerably less than 20% of the observed variance in any particular trait. Consequently, enormous differences in BMR (increased by 66%) were associated with very modest average differences in these measures of performance (increased by 11–13%). It might be argued that an average difference in reproductive output of 10% would be highly significant in evolutionary terms, supporting the idea that high BMR may be selected for because of the advantages in the enabled sustained metabolic rate and consequent reproductive performance enhancement. However, contrary to this argument, the associations that we observed between BMR and litter size, and BMR and weaning litter mass, only occurred because these traits were also related to M_b, and M_b and BMR were also weakly correlated. In all cases excepting the indirect measure of resorption, when we statistically removed the effect of M_b on BMR, the significant associations to the reproductive performance measures disappeared. Similar patterns of association between M_b, BMR and reproductive performance were observed in laboratory mice by Hayes and coworkers (Hayes et al., 1992b) utilising the BMR measured prior to rather than after reproduction. This indicates that measuring BMR after reproduction made no difference to the nature of the associations between these traits. Since we only measured

BMR after reproduction, it remains theoretically possible that there was an association between BMR prior to reproduction and reproductive performance, but after their large performance they downregulated their basal metabolism to compensate. Additional work measuring BMR at multiple points throughout the reproductive process might be necessary to ultimately test the hypothesis. Nevertheless, our data provide substantial support to the body of previous work that has suggested that individual variations in BMR in non-reproductive individuals do not provide an enabling mechanism for greater sustained metabolic rates leading to enhanced litter sizes or litter masses at either birth or weaning (Derting and McClure, 1989; Hayes et al., 1992b; Johnson et al., 2001b; Krol et al., 2003; Speakman and Krol, 2005).

Although we predicted that variations in BMR would be positively associated with parameters of reproduction, the only significant relationship we found was negative. Why high BMR should be associated with a greater probability of resorption is unclear. It is possible that resources are limited in pregnancy, and hence the female with a higher BMR has fewer resources to divert towards support of the developing foetuses. Yet, why resources should be limited in pregnancy, which exerts substantially less demand than lactation, is unclear. Moreover, this interpretation assumes that the single measure we made of BMR following reproduction was reflective of the BMR throughout pregnancy, which we did not establish. An alternative view is that for some reason individuals that sustained resorptions during reproduction ended up with an elevated BMR. Our protocol, which involved only a single measure of BMR, could not distinguish these effects. Whatever turns out to be the explanation of this effect, we found no support for a positive association between BMR and aspects of reproduction in this strain of mice.

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