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Quantitative genetics and fitness effects of basal metabolism

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Abstract The physiological requirements of reproduction are predicted to generate a link between energy, physiology and life history traits. Simultaneously, low maintenance costs, measured by energy consumption, are expected to be advantageous. Here we investigated fitness relatedness of traits by estimating genetic correlations between, and inbreeding depression for, body mass, basal metabolic rate (BMR) and other life history characters in a wild rodent, *Myodes glareolus*. The narrow-sense heritability of absolute and mass corrected BMRs were high for females ($h^2 = 0.48$ and 0.42) but low and non-significant for males (0.32 and 0.09). A significant positive genetic correlation between BMR and litter size suggests that traits connected to female fecundity might favour higher metabolism (i.e. support increased intake hypothesis). However, the estimates of inbreeding depression indicate that, while higher values of body mass and female litter size could be positively associated with overall fitness, the association between BMR and overall fitness in bank voles would be negative (i.e. support compensation hypothesis). This result suggests that the advantages of larger litters and larger body mass might be evolutionary constrained by high costs of maintenance of those traits, as reflected by the level of basal metabolism.

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Introduction

It has been suggested that natural selection on basal metabolic rate (BMR) could proceed toward either high or low values (Nilsson 2002; Blackmer et al. 2005; Johnston et al. 2007; Burton et al. 2011). Positive directional selection is predicted by the "increased intake hypothesis", which assumes that BMR (or correlated "capacity" traits) reveals the general efficiency of energy acquisition (Blackmer et al. 2005; Boratyński and Koteja 2010). Alternatively, negative directional selection is predicted by the "compensation hypothesis", which considers BMR a measure of the maintenance costs, where a high level of BMR results in less energy left for other processes, such as reproduction (Jackson et al. 2001; Ketola and Kotiaho 2009). Along with these two contradictory hypotheses, fitness predictions for BMR are complicated by observations of sex-specific selection on energetics (Bouteiller-Reuter and Perrin 2005; Boratyński et al. 2010). Evidence of fitness dependency on metabolic rates in animals has only recently begun to accumulate (Hayes and O'Connor 1999; Jackson et al. 2001; Blackmer et al. 2005; Boratyński and Koteja 2009, 2010; Boratyński et al. 2010; Hayes 2010). Much of this work supports the hypothesis that natural selection could have shaped variation in these physiological traits. However, results on contemporary populations often show variable magnitude and signs of correlations between fitness components and energy metabolism, and it is still unresolved how selection could have produced a wasteful strategy such as a high level of BMR observed in homeothermic endotherms (Koteja 2000, 2004; Kemp 2006; see also: Nespolo et al. 2011 for a new concept). Likewise, controversy over whether high or low levels of basal, resting, maintenance metabolism, or BMR, are evolutionary beneficial, is still unresolved (Burton et al. 2011).

In this study, the quantitative genetic and inbreeding parameters for morphology, physiology and some life history traits were estimated to assess the genetic signal for fitness dependency on traits in the Eurasian rodent species, *Myodes* (or *Clethrionomys*) *glareolus*, the bank vole. The phenotypic traits investigated here have been correlated with individuals' performance and fitness, and their importance for survival and reproductive success has been experimentally tested in natural conditions (Mousseau and Roff 1987; Oksanen et al. 2001; Oksanen et al. 2007; Boratyński and Koteja 2010; Boratyński et al. 2010). Also, substantial genetic variation in metabolic rate and the genetic covariation with other (morphological and behavioural) traits have been successfully estimated (Ronning et al. 2007; Sadowska et al. 2009). However, the long term effects of relationship between BMR and fitness in endothermic animals, and its possible traces on the genetics of that trait were not assessed. Likewise, information on the genetic correlation between physiology and life history, and inbreeding effects on metabolic rate are very limited (see: Ketola and Kotiaho 2009, 2010 for studies on an ectotherm).

Investigations of the genetic background of traits by estimating the proportion of additive variation, genetic correlation with other (fitness) characters and sensitivity to inbreeding (as it is inversely linked to past selection; Charlesworth and Willis 2009) can help to reveal potential association between a trait and fitness. Traits strongly related to fitness are expected to share a substantial proportion of genetic variation with other fitness components, reflected by significant genetic correlation (Roff 1997; Koteja 2000; Ronning

et al. 2007; Sadowska et al. 2009). Inbreeding can lead to inbreeding depression, or a shift in the trait mean away from its fitness optimum. Inbreeding depression depends on many features (frequency of the recessive alleles, number of coding loci and amount of the genetic variation in the trait), however, the change in the mean of the fitness related trait, and the subsequent decrease in fitness, depend on harmful recessive alleles being expressed (in homozygotes) during the process of inbreeding (de Boer and van Arendonk 1992; Johansson et al. 1993). Therefore, the level of inbreeding depression cannot be linked directly to the strength of selection, however, it has been successfully used to estimate the direction of the trait-fitness association (e.g.: Ketola and Kotiaho 2010). Inbreeding and expressed recessive alleles are predicted to change the trait value in the opposite direction of past directional selection, which promoted the dominance genetic effects of the trait.

By estimating quantitative genetic parameters and inbreeding effects we aimed to test whether the genetic variation in metabolic rate suggests fitness association, as reflected by significant genetic correlation with life history trait and phenotypic change due to inbreeding depression. Due to sexual dependency of selection on some traits (Bouteiller-Reuter and Perrin 2005; Boratyński et al. 2010) the parameters were identified for sexes separately and cross-sex correlations were also estimated (Ketola et al. 2012). It is predicted that if high levels of basal metabolism evolved due to selection, e.g. on capacity traits related to energy turnover (Koteja 2000; Kemp 2006), BMR will be genetically linked to life history trait that reflect high fitness. Therefore, we predicted to observe a significant positive genetic correlation between metabolic rate and the number of offspring (litter size); a trait related to reproductive success. Additionally, we predicted that the heritability and, more importantly, the additive genetic component, of BMR would be smaller than those of body mass. This prediction is based on the observation that additive variation will deplete faster in traits more closely related to fitness (Mousseau and Roff 1987; Houle 1992; DeRose and Roff 1999) and it is generally presumed that physiology is more functionally related to fitness than morphology. As previously described in different study systems, inbreeding depression is usually detected in traits also closely linked to fitness (Falconer and Mackay 1996; Roff 1997; DeRose and Roff 1999; Szulkin et al. 2007; Ala-Honkola et al. 2009; Ketola and Kotiaho 2009; Thiele et al. 2009; Michalczyk et al. 2010). This is because fitness traits are characterized by high proportion of dominance to additive genetic effects caused by segregation of recessive, deleterious alleles. Inbreeding reveals the recessive alleles and changes the trait means and decrease fitness (de Boer and van Arendonk 1992; Falconer and Mackay 1996; Roff 1997; Lynch and Walsh 1998; DeRose and Roff 1999). Therefore, even weak inbreeding can, in some circumstances, lead to severe phenotypic change and inbreeding depression. If inbreeding depression on BMR is detected, it is predicted that the decrease of BMR would support the positive fitness-BMR link and thus, the "increased intake hypothesis". An increase in BMR in response to inbreeding would support negative fitness effect of high maintenance costs and, thus, the "compensation hypothesis".

Materials and methods

Study organism

The bank vole (*Myodes glareolus*) is a Palearctic muroid rodent (Amori et al. 2008; Boratyński and Koteja 2010) frequently used in research of life history evolution and evolutionary physiology (Mappes and Ylonen 1997; Oksanen et al. 2001; Mappes et al.

2008). Studies have shown that morpho-physiological traits in bank voles, like body mass and size and basal metabolic rate, are highly variable on both the phenotypic and genetic level (Koivula et al. 2003; Labocha et al. 2004; Mappes and Koskela 2004; Sadowska et al. 2005). The substantial variation in size and metabolism was also described within and between natural populations, and the individual values remained repeatable even over relatively long term periods (intraclass correlation over an average of 56.5 days for body mass, head width and BMR: 0.67, 0.83 and 0.40, respectively; Boratyński and Koteja 2009). For the current study bank voles originated from a laboratory colony maintained at the University of Jyväskylä, Finland. Animals belonged up to 5th generation which ancestors were wild-trapped (150 females and 116 males) in central Finland (62°37′ N, 26°20′E). Total number of individuals in the pedigree used in the analysis was 1627. Standard breeding protocols developed in our laboratory were applied: voles were paired for 2 week, after that time males were separated from females, and females after parturition were separated from offspring after 2 weeks following parturition. After weaning sexes were kept in separate cages until the breeding procedure was applied in next generation (see details about the colony and breeding procedures in Boratyński et al. (2010) and Schroderus et al. (2010). The animals were housed in individual cages (43 × 26 × 15 cm) with bedding of wood shavings and hay, in standard conditions (a 16L:8D photoperiod, at 20 ± 2 °C, with food, Labfor 36, Lactamin AB, Stockholm, Sweden, and water available ad libitum). Measurements of litter size and mean offspring body mass after parturition were estimated from the first parturition. All experimental and housing procedures complied with the animal experimentation laws in Finland.

Basal metabolic rate

BMR measurements trials, including body mass measurements, were conducted once on not-reproducing adult individual belonging to the fourth and fifth generation. BMR was defined as the mean of the (third, fourth and fifth) lowest averages of oxygen consumption calculations based on the measurements conducted in respirometry systems (Boratyński et al. 2010; Boratyński and Koteja 2010). Measurements of oxygen consumption (ml h^{-1}) were conducted in an eight-channel open-flow respirometric system (Sable Systems, Henderson, NV) based on Fc-1B O_2 (Sable Systems) analyzer. Metabolic rate was calculated using O_2 values according to the formula: $VO_2 = \{V_i\ (FdO_2)/[1\text{-}FeO_2\ (1\text{-}RQ)]\}$, (eq. 1b in: (Koteja 1996)) where: VO_2—oxygen consumption rate, V_i—flow rate measured before chamber, FdO_2—difference of O_2 fractional concentrations in dry air flow before and after passing through the chamber, FeO_2—fractional concentration of O_2 in dry air flow after the chamber, assuming RQ = CO_2 eliminated/O_2 consumed of 0.75 for nearly starved animals. Prior to the measurements, seven animals were weighed and placed in chambers (180 ml volume; with the eighth channel remaining empty, as a reference) without access to water and food within the thermal neutral zone (30.0 ± 0.5 °C). The flow of dry air (dried with silica gel) of 260 ml/min was passing through the chambers. Oxygen consumption was recorded sequentially from 8 channels for a period of 7 h 30 min (from 09:00 to 16:30). A total of 29 oxygen consumption measures, sequential recordings for each chamber, were saved throughout one measurement trial.

Statistical analyses

The (co)variance components were estimated with the average information REML-animal model procedure using the ASReml 2.0 -program (Gilmour et al. 2002, 2006).

Table 1 Descriptive statistics of measured traits included in all analyses

	Females			Males		
Trait	N	Mean	SD	N	Mean	SD
Body mass (g)	161	20.5	3.69	184	25.1	4.37
BMR (ml O2/h)	161	39.8	5.83	184	44.9	6.77
BMR (residuals)	161	−0.03	4.04	184	0.02	5.21
Litter size[a]	876	4.20	1.30			
Offspring body mass[a]	876	1.93	0.22			

N number of individuals, *SD* standard deviation. Residual values were derived from linear regression models where BM was adjusted for variation in BMR, and BMR was adjusted for variation in BM

[a] Estimated for females only

REML-animal model has become the default method for quantitative genetic studies. It can handle complex pedigree structures and thus utilizes all available phenotypic data without need for breeding designs (Thompson et al. 2005; Thompson 2008). Linear regression on the inbreeding coefficient (estimated from pedigree) was fitted as a fixed effect for both body mass and BMR in all the models, including all phenotypic observations for each trait (Table 1). It allowed to estimate inbreeding depression/directional dominance and to correct additive genetic estimates from inflation caused by similarity due to directional dominance/inbreeding depression (de Boer and van Arendonk 1992; Ketola and Kotiaho 2009; Reid and Keller 2010). Statistical significance of inbreeding effect was assessed with Wald test implemented in the ASReml 3.0 (Gilmour et al. 2009). Conditional Wald statistics were compared against χ^2 distribution with one degree of freedom. Sex was included in the models as a fixed effect in all the analyses where the traits were treated as the same character across sexes. Age was included in all models as a covariate and models excluding age were also tested (Table S1). A set of linear models of increasing complexity hierarchically was fitted to determine the appropriate random effects structure for each trait:

$$y = Xb + e$$

$$y = Xb + Z_1 a + e$$

$$y = Xb + Z_1 a + Z_1 c + e$$

In which y is the vector of phenotypic observations; b is the vector of fixed effects; a, c and e are the vectors of direct additive genetic effects, common environmental effects and residuals, respectively. The common environmental effect fitted for each litter may include maternal, purely environmental and dominance genetic effects (Johansson et al. 1993; Wilson et al. 2010). Fixed and random effects are fitted to individual records by incidence matrices X, Z_1 and Z_2. The significance of the additive genetic and common environmental effect was assessed with a Log Likelihood ratio test. In the initial analysis, also models with genetic or permanent maternal effect in addition to common environmental effect were experimented. However, they were excluded from the final analysis since variation due to maternal effects proved to be negligible. Only half of the dams have more than one litter with phenotypic data, which means that litter specific common environmental effect most probably takes into account also possible maternal effects. Genetic correlations were estimated with bivariate models using the best model from the univariate analysis for each trait. Significance of the covariance between two traits was assessed by performing a Log Likelihood ratio test between a full model and model in which covariance in question

(additive genetic or common environmental) was constrained to zero. Female and male characters were treated as different traits when cross-sex correlations were estimated. Significance of the cross-sex correlation was tested by comparing Log Likelihood of a model used to estimate the correlation to a Log Likelihood of a model in which the correlation was constrained to 0.999. Due to high correlations and a functional link between morphological and physiological characters (bigger animals may have higher absolute metabolism, animals with faster energy processing may grow bigger; Sadowska et al. 2009), all the analyses of the basal metabolic rate were conducted for absolute values and on residual trait values (metabolic rate corrected for body mass, residual BMR). In these cases, estimated genetic parameters refer to the genetic background of the trait corrected for the (phenotypic) variation in the other trait. Estimates from bivariate models did not differ markedly from the univariate models therefore for simplicity results from bivariate models are included only in the supplementary material (Table S2). The quantitative genetic analyses described above were conducted on whole pedigree composed of 1627 individuals. The breeding values were extracted from ASReml output files.

Results

Heritability and inbreeding

Both of the characters measured, body mass and BMR, were highly variable (Table 1). Narrow-sense heritability (further referred as heritability) was high and significant for body mass ($h^2 = 0.58$; see Table 2 for standard errors). In more complicated models marginal common environmental effect was detected ($c^2 = 0.11$). The common environmental effect was also small for absolute and mass corrected BMR (Table 2), and the models including common environment effect were non-significant (Table 2). The heritability of absolute BMR was moderate ($h^2 = 0.34$), but when corrected for body mass it was very low (0.13). Heritabilities for litter size ($h^2 = 0.16$) and mean offspring body mass ($h^2 = 0.07$) were also low (Table 2).

Average inbreeding coefficients for individuals used in the BMR analysis was 0.024 in both males and females and for females with litter size data it was 0.017. Mean inbreeding coefficients for the females with metabolic measurements were 0.013 in the fourth and 0.041 in the fifth generation, whereas for males those values were 0.012 and 0.042, respectively. The inbreeding led to a decreased body mass ($b = -16.22$). This result was marginal but consistent over models (Table 2, see also Table S1). Inbreeding had little or no effect on the absolute values of BMR. However, inbreeding caused a significant increase in the residual (mass corrected) value of BMR ($b = 20.16$). The effect was consistent and significant among different models (Table 2, Table S1 and S2). For life-history traits, inbreeding had moderate, significant and negative effects only on litter size (Table 2 and Table S1).

The sex-specific heritability was comparable between sexes for body mass (Table 3). The common environmental effect was significant for body mass for males but not for females. The effect of inbreeding on body mass was stronger and more consistent among models for males compared to females. Sex-specific heritability for BMR differed markedly between sexes (Table 3). For females, absolute and residual BMR heritability were higher (0.48–0.42) than in males (0.32–0.09), for whom it approached zero for mass corrected BMR. The inbreeding was negligible for absolute but it was much stronger and increased BMR for mass corrected values (Table 3, Table S1).

Table 2 Components of variance (±SE) (sexes pooled) from quantitative genetic analyses, likelihood ratio test (LR) and significance (p) for univariate models, and coefficients (±SE) of explained variance and inbreeding effect on the trait

	Components of variance			Models tests		Coefficients								
Traits	A	C	E	LR	p	CV_A	h^2	c^2	b	F-con	p	Age	F-con	p
Body mass	8.25 ± 2.44		6.07 ± 1.42	49.3	<.0001	15.41	0.58 ± 0.12		−16.22 ± 8.25	3.86	0.049	0.007 ± 0.001	37.01	<.0001
	6.14 ± 2.49	1.56 ± 0.79	6.11 ± 1.42	6.64	0.01*	13.29	0.44 ± 0.15	0.113 ± 0.057	−13.34 ± 8.78	2.31	0.13	0.007 ± 0.001	30.98	<.0001
BMR	14.02 ± 5.75		26.85 ± 4.13	18.9	<.0001*	6.76	0.34 ± 0.12		7.71 ± 13.64	0.32	0.57	0.004 ± 0.002	4.92	0.027
	8.35 ± 5.48	4.74 ± 2.61	26.78 ± 3.82	4.63	0.03	5.21	0.21 ± 0.13	0.119 ± 0.064	6.47 ± 14.08	0.21	0.65	0.005 ± 0.002	5.74	0.017
Residual BMR	2.74 ± 1.70		18.65 ± 1.93	8.84	0.003*		0.13 ± 0.08		20.16 ± 9.03	4.99	0.025	−0.003 ± 0.001	4.91	0.027
	2.43 ± 1.83	0.72 ± 1.21	18.27 ± 1.95	0.35	0.55		0.11 ± 0.08	0.034 ± 0.057	19.41 ± 9.21	4.44	0.035	−0.003 ± 0.001	4.19	0.041
Litter size[a]	0.30 ± 0.10		1.58 ± 0.11	27.4	<.0001*	13.07	0.16 ± 0.05		−5.63 ± 1.95	8.31	0.004	−0.0012 ± 0.0003	15.96	<.0001
	0.29 ± 0.11	0.03 ± 0.09	1.56 ± 0.13	0.11	0.74	12.89	0.16 ± 0.06	0.017 ± 0.050	−5.60 ± 1.96	8.16	0.004	−0.0012 ± 0.0003	15.72	<.0001
Offspring body mass[a]	0.01 ± 0.01		0.05 ± 0.01	4.93	0.03*	2.98	0.07 ± 0.04		−0.02 ± 0.30	0.01	0.93	0.00002 ± 0.00005	0.13	0.72
	0.01 ± 0.01	0.01 ± 0.01	0.05 ± 0.01	0.76	0.38	2.73	0.06 ± 0.04	0.042 ± 0.049	−0.02 ± 0.30	0.001	0.99	0.00002 ± 0.00005	0.14	0.71

Components of variance: A—additive genetic, C—common environment, E—residual environment. Coefficients: h^2—heritability, proportion of additive genetic variance to phenotypic variance, c^2—proportion of common environment variance to phenotypic variance, CV_A—coefficients of additive genetic variation in % [(SD/mean)*100], b—fixed regression coefficient on inbreeding

[a] Estimated for females only

* Indicates the best model per character

Table 3 Sex-specific components of variance from quantitative genetic analyses, likelihood ratio test (LR) and significance (p) for univariate models and coefficients of explained variance and inbreeding effect on the trait

	Components of variance				Models tests		Coefficients								
Sex	Traits	A	C	E	LR	*p*	CV_A	h^2	c^2	b	F-con	p	Age	F-con	*p*
Females	Body mass	5.06 ± 2.28		5.00 ± 1.57	13.7	0.0002*	12.2	0.50 ± 0.18		−3.59 ± 8.72	0.17	0.68	0.009 ± 0.001	49.69	<.0001
		3.79 ± 2.31	1.37 ± 1.08	4.71 ± 1.49	2.11	0.15	10.60	0.38 ± 0.21	0.14 ± 0.11	−2.26 ± 9.08	0.06	0.81	0.009 ± 0.001	42.17	<.0001
	BMR	15.81 ± 7.41		17.27 ± 5.20	14.44	0.0001*	7.19	0.48 ± 0.18		14.09 ± 15.80	0.79	0.37	0.006 ± 0.002	6.36	0.012
		13.42 ± 7.66	3.01 ± 3.51	16.40 ± 5.09	0.82	0.36	6.62	0.41 ± 0.20	0.09 ± 0.11	14.59 ± 16.31	0.80	0.37	0.006 ± 0.003	5.81	0.016
	Residual BMR	6.43 ± 3.15		9.02 ± 2.34	16.0	0.0001*		0.42 ± 0.17		17.93 ± 10.77	2.71	0.10	−0.004 ± 0.002	7.28	0.007
		5.95 ± 3.31	1.07 ± 1.67	8.44 ± 2.38	0.42	0.52		0.38 ± 0.19	0.07 ± 0.11	17.54 ± 11.08	2.51	0.11	−0.004 ± 0.002	6.44	0.011
Males	Body mass	12.26 ± 4.61		5.67 ± 2.79	24.2	<.0001	22.43	0.68 ± 0.18		−22.40 ± 12.66	3.13	0.08	0.005 ± 0.002	9.46	0.002
		7.50 ± 4.38	3.77 ± 1.77	5.70 ± 2.49	8.04	0.0046*	14.50	0.44 ± 0.22	0.23 ± 0.10	−21.00 ± 13.14	2.55	0.11	0.006 ± 0.002	10.99	0.001
	BMR	15.12 ± 8.72		31.98 ± 6.95	5.96	0.015*	7.01	0.32 ± 0.17		2.70 ± 19.35	0.02	0.89	0.004 ± 0.003	2.32	0.13
		6.16 ± 8.84	9.08 ± 5.53	30.92 ± 6.31	3.53	0.060	4.47	0.13 ± 0.19	0.20 ± 0.12	−0.16 ± 16.68	0.01	1.0	0.006 ± 0.003	3.80	0.051
	Residual BMR	2.55 ± 3.11		24.40 ± 3.63	1.20	0.27		0.09 ± 0.11		20.79 ± 13.26	2.46	0.12	−0.002 ± 0.002	0.65	0.42
		1.42 ± 3.3.23	1.91 ± 2.80	23.60 ± 3.58	0.49	0.49		0.05 ± 0.12	0.07 ± 0.10	19.26 ± 13.50	2.04	0.15	−0.001 ± 0.002	0.37	0.54

See Table 2 for details

* Indicates the best model per character

Table 4 Additive genetic (r_A) and phenotypic Pearson's product-moment (r_P) correlations (±SE) between traits

Correlations	Additive genetic			Phenotypic		
Pair of traits	r_A	se	*p*	r_P	se	*p*
Body mass-BMR	0.87	0.10	0.009	0.73	0.03	<.0001
Females: body mass-BMR	0.78	0.13	0.005	0.72	0.04	<.0001
Males: body mass-BMR	0.95	0.09	0.001	0.65	0.04	<.0001
Body mass-Litter size[a]	0.19	0.45	0.66	0.39	0.03	<.0001
Body mass-offspring mass[a]	0.64	0.59	0.29	0.17	0.03	<.0001
BMR-litter size[a]	0.92	0.42	0.064	0.23	0.14	0.13
BMR-offspring body mass[a]	0.34	0.69	0.57	−0.36	0.13	0.017
Residual BMR-litter size[a]	1.11	0.40	0.022	0.12	0.15	0.43
Residual BMR- offspring mass[a]	−0.32	0.72	0.65	−0.42	0.13	0.005
Litter size-offspring body mass[a]	0.27	0.44	0.47	−0.50	0.03	<.0001

[a] Estimations based only on females

Genetic and phenotypic correlations

Bivariate genetic analyses (Table 4) revealed a high additive genetic correlation between body mass and BMR, both when including all data ($r_A = 0.87$) and in sex-specific analyses (females: $r_A = 0.78$, males: $r_A = 0.95$; Fig. 1). In general, the correlations were consistent with those observed at the phenotypic level (Pearson's product-moment correlations) which were lower ($r_P = 0.73$; Table 4; Fig. 1a). Moreover, for the life-history traits (calculated only for females), absolute BMR was nearly significant and residual BMR was significantly genetically correlated with litter size (Table 4; Fig. 2). This genetic correlation converged outside parameter space ($r_A > 1$) which might be due to sampling variance in a modestly sized data set. Other genetic correlations with litter size and offspring body mass were all non-significant, despite significance of some phenotypic correlations (Table 4), which was probably due to a low heritability of life-history traits (Table 2).

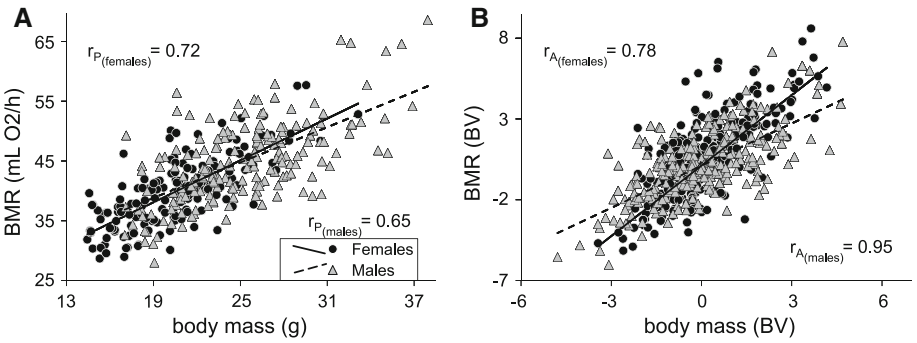

Fig. 1 Sex-specific correlations between body mass and basal metabolic rate (BMR) on phenotypic (**a** absolute trait values) and genetic (**b** breeding values—BV) levels. r_P—Pearson's product-moment, r_A—additive genetic correlations

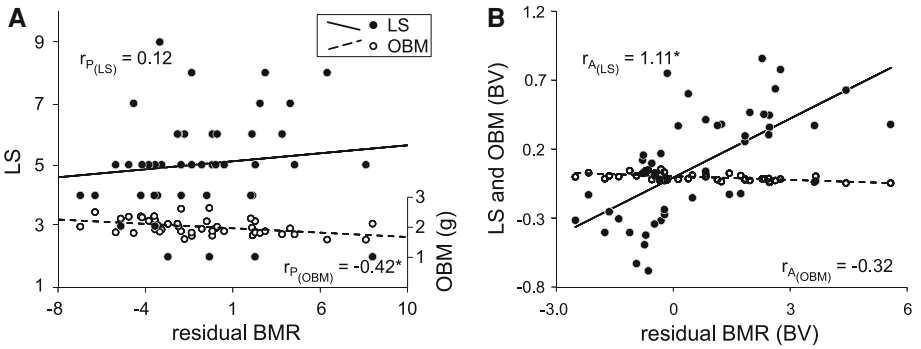

Fig. 2 Correlations between life history traits and residual basal metabolic rate (BMR corrected for body mass) on phenotypic (**a**, absolute trait values) and genetic (**b**, breeding values—BV) levels. *LS* litter size, *OBM* offspring body mass, r_P—Pearson's product-moment, r_A—additive genetic, correlations. Data presented for females only

Cross-sex genetic correlations

The cross-sex additive genetic correlation for body mass was high and significantly different from 0 ($r_A = 0.95$, se = 0.15, $p_0 = 0.001$). In contrast the cross-sex correlation for BMR was low and non-significant ($r_A = 0.60$, se = 0.28, $p_0 = 0.084$). Cross-sex correlation for residual BMR was even lower ($r_A = 0.41$, se = 0.65, $p_0 = 0.507$; Fig. 3). Due to the large standard errors none of the estimated correlations significantly differed from 0.999. However, cross-sex correlation for absolute BMR values was the closest to meet standard significance level of 0.05 when tested against 0.999 ($p_1 = 0.060$; for body mass and residual BMR $p_1 > 0.74$).

Discussion

In this study we analysed the quantitative genetic parameters of physiological, life history and morphological traits in the bank vole, *Myodes glareolus*. Our main aims were (1) to assess the genetic basis of traits (body mass, BMR, residual BMR, litter size, offspring body mass) and (2) to test sensitivity of the traits to inbreeding, for predicting how morpho-physiological characters are related to life history and fitness.

The heritability estimates are in agreement with previous quantitative genetic studies on the bank vole (Mappes and Koskela 2004; Sadowska et al. 2005; Sadowska et al. 2009) and showed substantial heritable variation in basal metabolism (at least for females) and body mass. Our estimates of heritability and coefficients of additive genetic variation are also in agreement with quantitative genetics theory (Mousseau and Roff 1987; Falconer and Mackay 1996; Roff 1997), which predicts a lower proportion of additive variation for traits more closely related to fitness (i.e.: physiological versus morphological; Table 2). In males, heritability estimate and coefficients of additive genetic variation for body mass were higher than for BMR. However, in females the heritability of body mass was only slightly higher than that of absolute and mass corrected BMR values (Table 3). Despite sex differences in heritability of BMR, coefficients of additive genetic variation for this trait were very similar between females and males and much smaller than for body mass.

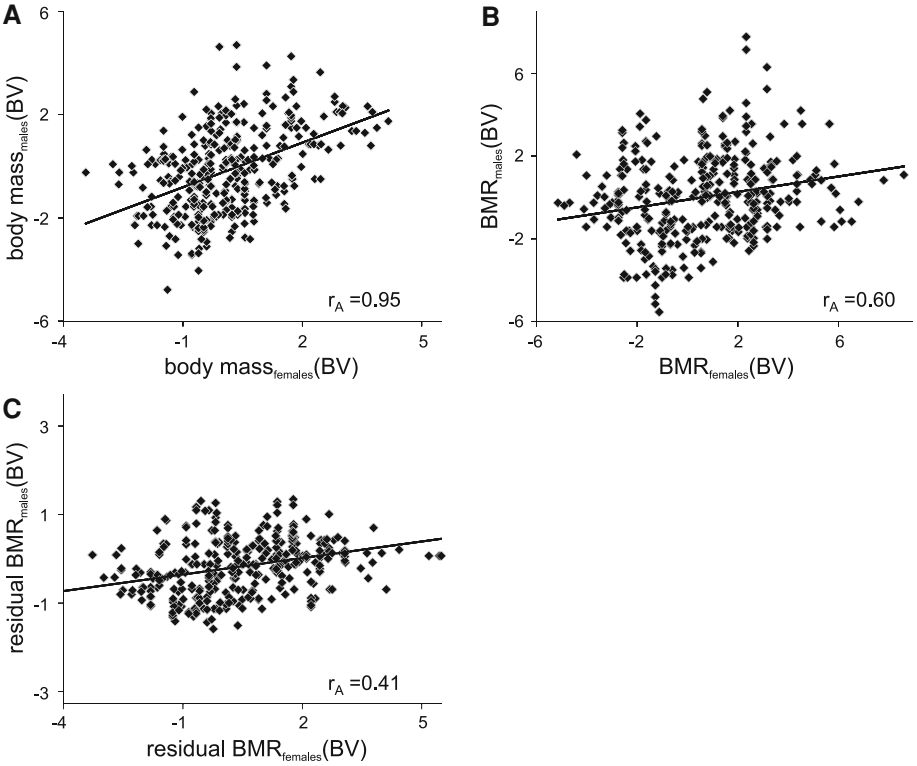

Fig. 3 Cross-sex correlations between female and male breeding values (BV) of body mass (A) and basal metabolic rate (BMR). r_A—additive genetic correlations

Traces of fitness effects on the genetic background of traits were indirectly estimated with inbreeding depression, which is predicted to modify the trait values in the opposite direction of past selection (e.g.: Roff and Emerson 2006). Here we detected a marginal effect of inbreeding depression on body mass—inbred individuals were characterized by decreased body mass (Table 2, Table S1) and the effect was mainly caused by inbreeding depression in males (Table 3). Since inbreeding decreased body mass, it suggests that higher body mass is positively associated with fitness. This result is consistent with our previous experimental data, which showed benefits of higher body mass both for survival and reproductive success in bank voles (Oksanen et al. 1999; Oksanen et al. 2001; Mappes and Koskela 2004; Boratyński and Koteja 2010).

Interestingly, we found a significant effect of inbreeding on metabolic rate corrected for body mass, but not on absolute BMR values (Table 2, Table S1). Inbred individuals were characterized by increased residual values of BMR and this pattern was consistent among different models (Table 2, see also: Table S1 and S2). Sex-specific analyses were non-significant, but showed a similar trend for both females and males (Table 3, see: Table S1 for significant effect in females). These findings indicated that inbreeding increased energy use for maintenance, reflected here by BMR, which may be considered disadvantageous for individuals. Assuming a negative link between fitness and inbreeding, the estimated inbreeding depression on BMR supports the compensation model for the evolution of maintenance metabolism, which predicts a negative link between fitness and BMR.

Two previous studies on endotherms also suggested that selection can promote low maintenance costs. In storm petrels (*Oceanodroma leucorhoa*) BMR was negatively associated with chick growth rate whereas in North American red squirrels (*Tamiasciurus hudsonicus*) juveniles were more likely to survive winter if they had low levels of resting metabolism (Blackmer et al. 2005; Larivée et al. 2010). On the contrary, other correlative studies indicated that selection on BMR might be positive [over-winter survival on mass-independent BMR: (Jackson et al. 2001)], differential between sexes [over-winter survival on BMR: (Boratyński et al. 2010)] and may vary between seasons and years [survival, reproductive and sexual selection on BMR: (Boratyński and Koteja 2009; Boratyński and Koteja 2010)].As these earlier studies measured rather short-time fitness effects, they cannot reveal the long term consequences of variation in a trait (see: Nespolo et al. 2011). By applying here an indirect inbreeding method we aimed to identify the direction of a long term fitness association with BMR.

A strong positive correlation between bank vole body mass and BMR was detected at both the phenotypic and the genetic level (Table 4; Fig. 1), however, the correlation between body mass and life-history traits (litter size and offspring mass) were only significant at the phenotypic level. The negative phenotypic correlation between offspring body mass and residual BMR was opposed by a strong positive genetic correlation between residual BMR and litter size (Fig. 2). This effect is compatible with the observed life-history trade-off between litter size and offspring quality (Roff 2002; Mappes and Koskela 2004; Oksanen et al. 2007). It could signify that females with a higher metabolic rate (BMR independent of body mass) also possess genes allowing them to produce larger litters. This result supports the prediction of parental care models for the evolution of endothermy, suggesting that females with a high level of energy flux (reflected by elevated BMR) are better prepared for costly parental provisioning during pregnancy and lactation, which are crucial behaviours for offspring fitness (Koteja 2000; Farmer 2003). As a result of correlated evolution, both metabolic rate and the nature of parental behaviour could have been increased by selection promoting increased energy flux. An additive genetic correlation between BMR and litter size is also in agreement with the recent empirical data showing a phenotypic correlation between reproductive success and BMR in a wild population of our study species (Boratyński and Koteja 2010).

Conclusions

Our results based on inbreeding method support the hypothesis of fitness advantage of individuals with lower maintenance costs, as BMR increased along with inbreeding effects. This is consistent with the allocation principle or the "compensation hypothesis" according to which BMR is considered a surrogate for maintenance costs. However, our results based on genetic correlations reveal a positive relationship between BMR and litter size. This suggests that females with high BMR are better prepared for parental provisioning due to higher energy flux that allows higher fecundity. These opposing results, based on overall fitness and female related fitness, might partly explain the low level of additive genetic co-variation of BMR shared between sexes (Fig. 3). Low cross-sex correlation for BMR will release energy metabolism, allowing it to approach its sex specific optima (Ketola et al. 2012). Whether this phenomenon is universal and whether it is due, for example, to sex-linked inheritance or sex-linked gene expression is for future studies to determine.

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References

Amori G, Hutterer R, Kryštufek B, Yigit N, Mitsain G, Muñoz LJP Henttonen H, Vohralík V, Zagorodnyuk I, Juškaitis R, Meinig H, Bertolino S (2008) *Myodes glareolus*. In: IUCN (2010) IUCN red list of threatened species. Version 2010.2. www.iucnredlist.org

Blackmer AL, Mauck RA, Ackerman JT, Huntington CE, Nevitt GA, Williams JB (2005) Exploring individual quality: basal metabolic rate and reproductive performance in storm-petrels. Behav Ecol 16:906–913

Boratyński Z, Koteja P (2009) The association between body mass, metabolic rates and survival of bank voles. Funct Ecol 23:330–339

Boratyński Z, Koteja P (2010) Sexual and natural selection on body mass and metabolic rates in free-living bank voles. Funct Ecol 24:1252–1261

Boratyński Z, Koskela E, Mappes M, Oksanen TA (2010) Sex-specific selection on energy metabolism: selection coefficients for winter survival. J Evol Biol 23:1969–1978

Bouteiller-Reuter C, Perrin N (2005) Sex-specific selective pressures on body mass in the greater white-toothed shrew, *Crocidura russula*. J Evol Biol 18:290–300

Burton T, Killen SS, Armstrong JD, Metcalfe NB (2011) What causes intraspecific variation in resting metabolic rate and what are its ecological consequences? Proc R Soc B 278:3465–3473

Charlesworth D, Willis JH (2009) The genetics of inbreeding depression. Nature Rev Genet 10:783–796

de Boer IJM, van Arendonk JAM (1992) Predicting of additive and dominance effects in selected or unselected populations with inbreeding. Theor Appl Genet 84:451–459

DeRose MA, Roff DA (1999) A comparison of inbreeding depression in life-history and morphological traits in animals. Evolution 53:1288–1292

Falconer DS, Mackay TFC (1996) Introduction to quantitative genetics. Longman, Essex

Farmer CG (2003) Reproduction: the adaptive significance of endothermy. Am Nat 162:826–840

Gilmour AR, Gogel BJ, Cullis BR, Welham SJ, Thompson R (2002) ASReml User Guide Release 1.0. VSN International Ltd, Hemel Hempstead, UK

Gilmour AR, Cullis BR, Harding SA, Thompson R (2006) ASReml Update: What's new in release 2.00. VSN International Ltd, Hemel Hempstead, UK

Gilmour A, Gogel B, Cullis B, Thompson R (2009) ASReml User Guide Release 3.0

Hayes JP (2010) Metabolic rates, genetic constraints, and the evolution of endothermy. J Evol Biol 23:1868–1877

Hayes JP, O'Connor CS (1999) Natural selection on thermogenic capacity of high-altitude deer mice. Evolution 53:1280–1287

Honkola O, Uddstrom A, Pauli Diaz B, Llindstrom K (2009) Strong inbreeding depression in male mating behaviour in a poeciliid fish. J Evol Biol 22:1396–1406

Houle D (1992) Comparing evolvability and variability of quantitative traits. Genetics 130:195–204

Jackson DM, Trayhurn P, Speakman JR (2001) Associations between energetics and over-winter survival in the short-tailed field vole *Microtus agrestis*. J Anim Ecol 70:633–640

Johansson K, Kennedy BW, Quinton M (1993) Prediction of breeding values and dominance effects from mixed models with approximations of the dominance relationship matrix. Livest Prod Sci 34:213–223

Johnston SL, Souter DM, Erwin SS, Tolkamp BJ, Yearsley JM, Gordon IJ, Illius AW, Kyriazakis I, Speakman JR (2007) Associations between basal metabolic rate and reproductive performance in C57BL/6 J mice. J Exp Biol 210:65–74

Kemp TS (2006) The origin of mammalian endothermy: a paradigm for the evolution of complex biological structure. Zool J Linn Soc 147:473–488

Ketola T, Kotiaho JS (2009) Inbreeding, energy use and condition. J Evol Biol 22:770–781

Ketola T, Kotiaho J (2010) Inbreeding, energy use and sexual signaling. Evol Ecol 24:761–772

Ketola T, Kristensen TN, Kellermann VM, Loeschcke V (2012) Can evolution of sexual dimorphism be triggered by developmental temperatures? J Evol Biol 25:847–855

Koivula M, Koskela E, Mappes T, Oksanen TA (2003) Cost of reproduction in the wild: manipulation of reproductive effort in the bank vole. Ecology 84:398–405

Koteja P (1996) Measuring energy metabolism with open-flow respirometric systems: which design to choose? Funct Ecol 10:675–677
Koteja P (2000) Energy assimilation, parental care and the evolution of endothermy. Proc R Soc B 267:479–484
Koteja P (2004) The evolution of concepts on the evolution of endothermy in birds and mammals. Physiol Biochem Zool 77:1043–1050
Labocha MK, Sadowska ET, Baliga K, Semer AK, Koteja P (2004) Individual variation and repeatability of basal metabolism in the bank vole, *Clethrionomys glareolus*. Proc R Soc B 271:367–372
Larivée ML, Boutin S, Speakman JR, McAdam AG, Humphries MM (2010) Associations between over-winter survival and resting metabolic rate in juvenile North American red squirrels. Funct Ecol 24:597–607
Lynch M, Walsh B (1998) Genetics and analysis of quantitative traits. Sinauer, Sunderland
Mappes T, Koskela E (2004) Genetic basis of the trade-off between offspring number and quality in the bank vole. Evolution 58:645–650
Mappes T, Ylonen H (1997) Reproductive effort of female bank voles in a risky environment. Evol Ecol 11:591–598
Mappes T, Grapputo A, Hakkarainen H, Huhta E, Koskela E, Saunanen R, Suorsa P (2008) Island selection on mammalian life-histories: genetic differentiation in offspring size. BMC Evol Biol 8:296
Michalczyk Ł, Martin OY, Millard AL, Emerson BC, Gage MJG (2010) Inbreeding depresses sperm competitiveness, but not fertilization or mating success in male *Tribolium castaneum*. Proc R Soc B 277:3483–3491
Mousseau TA, Roff DA (1987) Natural selection and the heritability of fitness components. Heredity 59:181–197
Nespolo RF, Bacigalupe LD, Figueroa CC, Koteja P, Opazo JC (2011) Using new tools to solve an old problem: the evolution of endothermy in vertebrates. Tree 26:414–423
Nilsson JA (2002) Metabolic consequences of hard work. Proc R Soc B 269:1735–1739
Oksanen TA, Alatalo RV, Horne TJ, Koskela E, Mappes J (1999) Maternal effort and male quality in the bank vole, Clethrionomys glareolus. Proc R Soc B 266:1495–1499
Oksanen TA, Jonsson P, Koskela E, Mappes T (2001) Optimal allocation of reproductive effort: manipulation of offspring number and size in the bank vole. Proc R Soc B 268:1–6
Oksanen TA, Koivula M, Koskela E, Mappes T, Hughes K (2007) The cost of reproduction induced by body size at birth and breeding density. Evolution 61:2822–2831
Reid JM, Keller LF (2010) Correlated inbreeding among relatives: occurrence, magnitude, and implications. Evolution 64:973–985
Roff DA (1997) Evolutionary quantitative genetics. Chapman and Hall, New York
Roff DR (2002) Life history evolution. Sinauer Associates, Inc
Roff DA, Emerson K (2006) Epistasis and dominance: evidence for differential effects in life-history versus morphological traits. Evolution 60:1981–1990
Ronning B, Jensen H, Moe B, Bech C (2007) Basal metabolic rate: heritability and genetic correlations with morphological traits in the zebra finch. J Evol Biol 20:1815–1822
Sadowska ET, Labocha MK, Baliga K, Stanisz A, Wroblewska AK, Jagusiak W, Koteja P (2005) Genetic correlations between basal and maximum metabolic rates in a wild rodent: consequences for evolution of endothermy. Evolution 59:672–681
Sadowska ET, Baliga-Klimczyk K, Labocha MK, Koteja P (2009) Genetic correlations in a wild rodent: grass-eaters and fast-growers evolve high basal metabolic rates. Evolution 63:1530–1539
Schroderus E, Jokinen I, Koivula M, Koskela E, Mappes T, Mills SC, Oksanen AT, Poikonen T (2010) Intra- and intersexual trade-offs between testosterone and immune system: implications for sexual and sexually antagonistic selection. Am Nat 176:E90–E97
Szulkin M, Garant D, McCleery RH, Sheldon BC (2007) Inbreeding depression along a life-history continuum in the great tit. J Evol Biol 20:1531–1543
Thiele J, Hansen T, Siegismund HR, Hauser TP (2009) Genetic variation of inbreeding depression among floral and fitness traits in *Silene nutans*. Heredity 104:52–60
Thompson R (2008) Estimation of quantitative genetic parameters. Proc R Soc B 275:679–686
Thompson R, Brotherstone S, White IMS (2005) Estimation of quantitative genetic parameters. Phil Trans R Soc B 360:1469–1477
Wilson AJ, Réale D, Clements MN, Morrissey MM, Postma E, Walling CA, Kruuk LEB, Nussey DH (2010) An ecologist's guide to the animal model. J Anim Ecol 79:13–26