Asian-Aust. J. Anim. Sci.
Vol. 19, No. 12 : 1671 - 1677
December 2006
www.ajas.info

Correlations between Heterozygosity at Microsatellite Loci, Mean d^2 and Body Weight in a Chinese Native Chicken

G. Q. Liu, X. P. Jiang*, J. Y. Wang and Z. Y. Wang

[1]College of Animal Science and Technology, Yangzhou University, Yangzhou, Jiangsu, 225009, P. R. China

ABSTRACT : A total of two hundred twenty eight half-sib chickens were scored for allele size at 20 microsatellite loci to estimate individual heterozygosity and mean d^2. The averages of microsatellite heterozygosity, allele per locus and mean d^2 were 0.39, 3.6 and 49, respectively. The body weight was measured biweekly from birth to twelve weeks of age. *Gompertz* function was assumed to simulate body weight and to estimate the growth model parameters. Due to sex effect on body weight, the regression of body weight on heterozygosity as well as on mean d^2 in males and females was analyzed separately in the present study. Positive correlations were found between microsatellite heterozygosity and body weight in males and females ($p<0.05$). Positive correlation also observed between individual heterozygosity and simulated maximum daily gain estimated from *Gompertz* function in female chickens ($p<0.05$). There were no significant correlations between mean d^2 and body weight. The results suggest that local effect hypothesis could explain the correlations between heterozygosity and fitness-related traits in the domesticated chicken population, rather than the general effect hypothesis does. (**Key Words :** Microsatellite Heterozygosity, Mean d^2, Body Weight, Chicken)

INTRODUCTION

Numerous efforts have attempted to correlate fitness-related traits, such as growth rate, with molecular marker heterozygosity in natural population. Most of the studies found positive correlations between growth traits and individual heterozygosity, and allozyme markers were widely used in these studies (Hansson and Westerberg, 2002). Based on these studies, one mechanism, the direct effect hypothesis, was founded on the multilocus heterozygosity-fitness correlations (HFCs) resulting from functional overdominance at the marker loci (Smouse, 1986; Houle, 1989; Pogson and Zouros, 1994; Pogson and Fevolden, 1998).

In the near decade, DNA markers were employed in many researches because DNA markers have abundant variability in most eukaryote genomes (Hughes and Queller, 1993; Jarne and Lagoda, 1996; Tsitrone et al., 2001; Wang et al., 2004; Tu et al., 2006). Microsatellite loci were introduced and positive correlations were also found in studies involving HFCs (Coltman et al., 1998; Jiang et al., 2003; Markert et al., 2004; Jiang et al., 2005; Pujolar et al., 2005; Zhang et al., 2005). The use of selectively neutral genetic markers stimulate associative overdominance hypothesis as the underlying mechanism, that is, the loci are mere markers, with other genes being responsible for the measured traits, the HFCs resulting from genetic associations between the neutral marker loci and the loci under selection (Zouros, 1993; David et al., 1995; David, 1998). The associative overdominance hypothesis involved two aspects: the local effect hypothesis, based on the linkage disequilibrium between the marker loci and the fitness-related traits loci in the local chromosomal vicinity, and the general effect hypothesis which based on the linkage disequilibrium throughout the genome resultant of inbreeding (David, 1998; Pogson and Fevolden, 1998; Bierne et al., 1998; Thelen and Allendorf, 2001; Hansson and Westerberg, 2002; Curik et al., 2003; Markert et al., 2004). A possible method to distinguish local effects and general effects is to use data from populations where detailed pedigrees can be constructed. Then it can be examined whether the inbreeding coefficient correlates with the marker heterozygosity, or variation in marker heterozygosity (but not in inbreeding coefficient) correlates with the trait. Coulson et al. (1998), assuming that microsatellites mutate mainly by stepwise changes in the number of repeat units, proposed d^2, the squared difference

* Corresponding Author: X. P. Jiang. College of Animal Science and Veterinary Medicine, Huazhong Agricultural University, Wuhan 430070, P. R. China. Tel: +86-027-6211-6868, Fax: +86-027-8739-4184, E-mail: xpjiang@mail.hzau.edu.cn
Received February 6, 2006; Accepted April 13, 2006

in repeat units between the two alleles of an individual, as a useful indicator of inbreeding. This makes it possible to distinguish local and general effects by using microsatellite loci.

Heterozygosity-fitness correlations are not only of theoretical interest, they are also valuable in animal breeding, since highly heterozygosity should be carried out with breeding method if positive HFCs do exist in populations. Chicken is one of the important fowls and its body weight has significant economical importance. Here we detected relationships between body weight and estimates of genetic variation in Yangzhou Chicken by using half-sib data to exclude variation in the inbreeding coefficient. The present experiment was set up (1) to serve as a basis for future studies on the relationship among genetic variability (microsatellite heterozygosity and mean square allelic distance, d^2) and body weight, (2) to test optimum individual heterozygosity level for maximum body weight, and (3) to probe the mechanism underlying the association of body weight and individual heterozygosity.

MATERIAL AND METHODS

Experimental animals

Yangzhou chicken is a native breed famous for its high meat quality and it mainly distributed in the Yangtze Delta in China. The breed was qualified a dual-purpose breed of "Chinese poultry breed" by authoritative Committee of "The Breeds of Domestic Animal and Poultry in China" in 1984. In present study, 250 unrelated hens were randomly inseminated with semen from 50 cocks without sibling mating, and their 280 progeny were reared for experiment. The data of 228 half siblings (101 males and 127 females) were employed in this study. The experimental chickens were reared on a deep-litter floor pen (5 m×8 m). They were free access to water and were fed *ad libitum* starter diet (12.28 MJ/kg and 18% CP) until 6 week of age and grower diet (12.18 MJ/kg and 16% CP) from the 7th week until slaughtered. The lighting regime started with 23 h/d to 14 days post hatch after which lighting was reduced to 12 h/d. At 7 week of age, the chicks were exposed to natural day light. Body weight of each chicken was measured biweekly using electronic scale until 12 weeks of age.

Microsatellite genotyping

The PCR for microsatellite genotyping comprised a total reaction volume of 25 μl:1.0 μl of template DNA (100 ng), 0.5 μl of Taq polymerase (2 U/μl), 0.5-1.0 μl dNTPs (10 mM), 0.5 μl of each primer (100 ng/μl), 18-18.5 μl ddH_2O and 2.5 μl 10×reaction buffer (with 20 mM $MgCl_2$) provided by the enzyme supplier. An Eppendorf Thermal Cycler was programmed for an initial incubation at 94°C for 3 min; 35 cycles each with denaturing at 94°C for 50 s, annealing at 56-60°C for 45 s and extension at 72°C for 50 s; and a final cycle at 72°C for 5 min. The PCR products were electrophoresed with polyacrylamide gel and silver stained with standard methods. Fragment sizes were determined by DNA sequencer (Shanghai Sangon Corporation).

Data analysis

The growth data of each individual were simulated with the following *Gompertz* growth function,

$$W = k*\exp(-A*\exp(-B*d)),$$

where W is the expected body weight of individual at age d-days; K is the maximum individual body weight estimated by *Gompertz* function; A is a growth constant; B is a rate of maturing. According to this model, age at the point of inflection is Ln (A/B), and the maximum day-gain for body weight is $B*K$/e.

Data were analyzed using the nonlinear regression model procedure (PROC NLIN) of Statistical Analysis System software package (SAS 1996). The R^2-value of the linear regression between observed and predicted body weight was used to indicate the goodness of fit.

A x^2-test for goodness of fit was applied to determine whether genotype frequencies at each locus were in Hardy-Weinberg equilibrium.

Individual heterozygosity was the proportion of loci that are heterozygous among the twenty microsatellite loci for each chicken. Mean d^2, the squared difference in repeat units in microsatellite loci between two alleles in an individual was calculated with the following function (Coulson et al., 1998).

$$\text{mean } d^2 = \sum_{i=1}^{n} \frac{(i_a - i_b)^2}{n}$$

where i_a and i_b are the lengths in repeat units of alleles a and b at locus i, and n is the total number of loci at which an individual was scored.

All data were checked for normality and the variables approximated a normal distribution, so the data were not standardized before parametric statistical analysis. Correlations between individual heterozygosity, mean d^2, and body weight were tested with regression (REG) procedure of SAS.

RESULTS

Phenotypic means

Means and standard deviations for body weight were listed in Table 1. The body weight by sex was normally

Table 1. Sample size, means and standard deviations for body weight and its simulated parameters

Traits	Male		Female	
	Sample size	Mean	Sample size	Mean
Birth weight (g)	101	34.62±3.20	127	34.65±3.33
Weight at 2 week (g)	101	136.56±16.69^{A}	126	128.58±14.15^{B}
Weight at 4 week (g)	97	308.97±40.71^{A}	124	272.55±31.97^{B}
Weight at 6 week (g)	97	506.47±69.34^{A}	119	427.99±57.49^{B}
Weight at 8 week (g)	101	791.69±110.70^{A}	125	656.03±83.27^{B}
Weight at 10 week (g)	99	1,100.52±138.25^{A}	118	888.29±112.74^{B}
Weight at 12 week (g)	101	1,295.34±193.49^{A}	127	1,024.18±131.22^{B}
Maximum weight (g)	100	2,309.13±631.04^{A}	127	1,723.47±454.18^{B}
Inflection age (day)	100	74.06±3.00	127	72.30±2.43
Maximum day-gain (g)	100	20.69±3.10^{A}	127	15.40±2.26^{B}

In the same age (week) means with different letter indicates significant difference ($p<0.01$).

Table 2. Chromosome location, allele size, number of allele, observed heterozygosity (H_o), expected heterozygosity (H_e), difference between heterozygote and homozygote (*D*) and polymorphism information content (*PIC*) for microsatellite loci

Locus	Chromosome location	Allele size	Number of alleles	*Ho*	*He*	*D*	*PIC*
ADL155	3	108-116	2	0.14	0.16	-0.13	0.15
ADL183	1	291-340	6	0.74	0.75	-0.01	0.71
ADL185	2	148-166	5	0.58	0.58	-0.00	0.53
ADL201	Z	277-309	2	0.81	0.50	0.62	0.37
ADL217	2	158-172	5	0.34	0.34	0.00	0.31
ADL273	Z	150-168	2	0.16	0.27	-0.41	0.24
ADL292	5	120-156	4	0.91	0.66	0.38	0.60
LEI66	-	303-319	3	0.38	0.65	-0.42	0.58
LEI94	4	195-223	3	0.14	0.16	-0.13	0.15
LEI166	3	252-264	3	0.33	0.43	-0.23	0.38
MCW58	1	158-186	4	0.24	0.68	-0.65	0.62
MCW85	4	274-284	3	0.28	0.61	-0.54	0.54
MCW120*	7	270-284	5	0.25	0.63	-0.60	0.57
MCW154	Z	171-192	5	0.26	0.25	0.04	0.24
MCW170	4	263-290	4	0.58	0.72	-0.19	0.67
MCW180	4	70-84	5	0.61	0.72	-0.15	0.67
MCW258	Z	141-162	3	0.29	0.62	-0.53	0.55
MCW264	2	227-241	2	0.21	0.19	0.11	0.17
MCW294	Z	305-317	2	0.02	0.02	0.00	0.02
MCW330	17	260-290	3	0.59	0.60	-0.02	0.52

* The locus is out of Hardy-Weinberg equilibrium; $D = (H_o-H_e)/H_e$.

distributed, and the body weight of cocks was significantly higher than that of hens ($p<0.01$). Range of the body weight and the simulated maximum body weight varied widely in males and females, which suggested that the population might contain sufficient genetic variability and the data could be used to analysis correlations between heterozygosity and body weight. The R^2-value for *Gompertz* function was about 0.97 and the simulated body weight curve was similar to the actual curve, which implied that body weight of this population could be simulated with *Gompertz* function.

Genetic diversity

Genetic parameters of the loci were depicted in Table 2. The twenty microsatellite loci were characterized by considerable allelic variation in number of alleles and observed heterozygosity across all chickens. Observed heterozygosity of microsatellite loci in the population ranged from 0.02 to 0.91. The average heterozygosity and polymorphism information content (*PIC)* estimated was 0.39 and 0.43, respectively. Mean d^2 in the population ranged from 5 to 227, and the averaged mean d^2 was 49. Heterozygosity and mean d^2 varied among individuals, which indicated a wide range of genetic variation within the chicken population, an important consideration when testing for HFCs. One of the twenty microsatellite loci was significantly out of Hardy-Weinberg proportions. The departure from Hardy-Weinberg equilibrium was due to an excess of homozygotes.

Correlations between heterozygosity and body weight

Due to sex effect on body weight, the regression of body weight on heterozygosity as well as on mean d^2 in males and females was analyzed separately. The univariate

Table 3. Univariate regression of body weight on individual heterozygosity

Trait	Female				Male			
	Interception	Slope	r^2	P	Interception	Slope	r^2	P
Birth weight (g)	36.16	-4.24	0.014	0.184	35.94	-3.21	0.011	0.297
Weight at 2 week (g)	123.75	11.71	0.005	0.529	140.90	-9.37	0.003	0.602
Weight at 4 week (g)	270.65	1.21	0.019	0.127	312.67	-7.98	0.000	0.857
Weight at 6 week (g)	420.57	12.31	0.024	0.091	498.42	21.29	0.021	0.101
Weight at 8 week (g)	631.05	51.35	0.031	0.042	770.95	44.81	0.025	0.086
Weight at 10 week (g)	829.62	119.14	0.040	0.035	1,022.56	167.43	0.041	0.032
Weight at 12 week (g)	949.58	190.92	0.062	0.017	1,179.48	250.04	0.038	0.036
Maximum weight (g)	1,539.77	508.18	0.012	0.273	1,853.73	957.75	0.018	0.191
Inflection age (day)	71.89	0.62	0.002	0.696	71.70	2.47	0.002	0.678
Maximum day-gain (g)	13.59	4.77	0.047	0.021	18.74	4.23	0.023	0.095

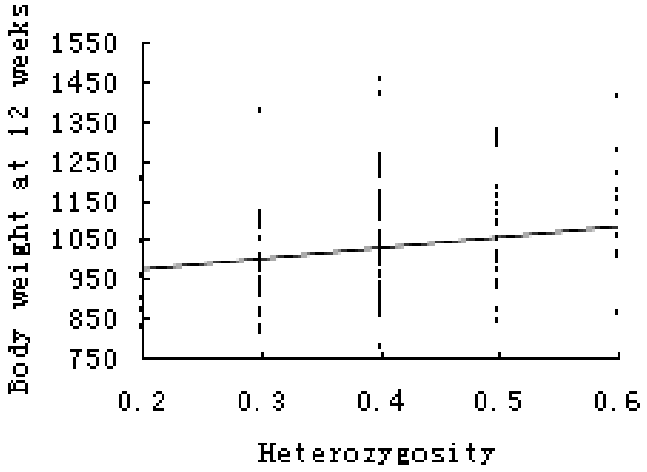

Figure 1. Univariate linear regression of female body weight at 12-week against microsatellite heterozygosity.

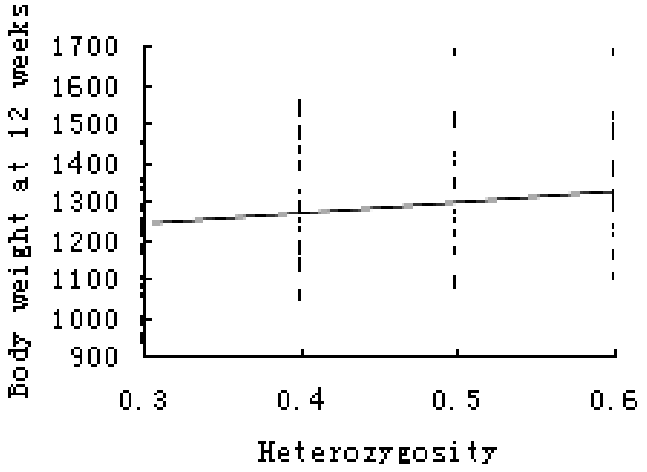

Figure 2. Univariate linear regression of male body weight at 12-week against microsatellite heterozygosity.

regression analyses of body weight and microsatellite heterozygosity was listed in Table 3. There were no significant correlations between individual heterozygosity and female body weight at birth, two, four and six weeks ($p>0.05$). Yet microsatellite heterozygosity was positively correlated with their body weight at eight, ten and twelve weeks ($p<0.05$, Figure 1). The r-squared-values of the significant positive slopes of the linear regression ranged from 0.031 to 0.062, which suggested that microsatellite heterozygosity could explain 3.1-6.2% phenotypic variance of female body weight. Microsatellite heterozygosity was positively correlated with male body weight at ten and twelve weeks ($p<0.05$, Figure 2). The r-squared-values of the significant positive slopes of the linear regression were 0.041 and 0.038, respectively, which suggested that microsatellite heterozygosity could explain 3.8-4.1% phenotypic variance of male body weight.

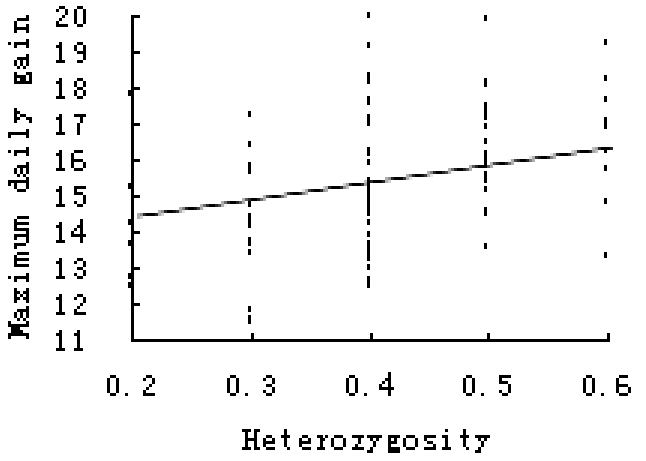

Figure 3. Univariate linear regression of simulated maximum daily gain against microsatellite heterozygosity in females.

Individual heterozygosity positively correlated with simulated maximum daily gain estimated from *Gompertz* function in females ($p<0.05$, Figure 3), but there were no significant correlations between heterozygosity and other simulated parameters ($p>0.05$). The r-squared-value of the significant positive slope of the linear regression was 0.047 ($p<0.05$), which suggested that microsatellite heterozygosity could explain 4.7% variance of the female chicken maximum daily gain. There were no significant correlations between heterozygosity and simulated parameters from *Gompertz* function in males ($p>0.05$).

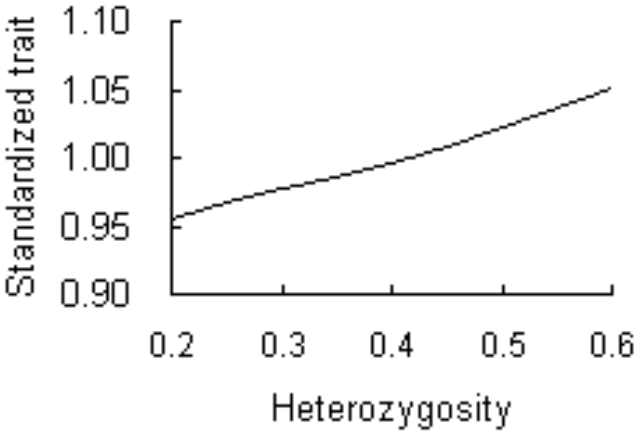

Figure 4. Changes of female body weight with microsatellite heterozygosity.

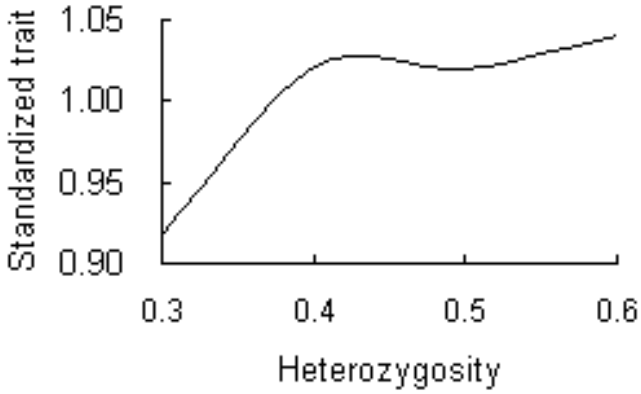

Figure 5. Changes of male body weight with microsatellite heterozygosity.

There were no significant correlations between mean d^2 and body weight in males and females ($p>0.05$).

Changes of body weight with microsatellite heterozygosity

Body weight increased when individual heterozygosity increased and the changing trend was similar from 2 to 12 weeks in females (Figure 4). Males' body weight increased when individual heterozygosity increased, with a little lower when the heterozygosity was 0.5, and the changing trend was similar from 6 to 12 weeks (Figure 5). There was no difference of simulated maximum body weight at each heterozygosity level in males and females, but the simulated maximum daily gain increased with microsatellite heterozygosity in females (Figure 6).

DISCUSSION

According to our knowledge this work was the first one on heterozygosity-fitness correlations (HFCs) in domesticated chicken population. Positive association between microsatellite heterozygosity and body weight in males and females was observed and more heterozygous individuals presented a higher body weight, which suggested the existence of HFCs in domesticated population. Body weight increased with microsatellite heterozygosity, which implied body weight could be improved by increasing heterozygosity. Estimated maximum daily gain of males and females increased when microsatellite heterozygosity increased, however, the correlation between them was significant only for females ($p<0.05$), which implied that improving the growth rate of body weight might be possible by maintaining high level of heterozygosity. All the results indicated that heterozygosity should be maintained at high level with breeding method in standard chicken breed.

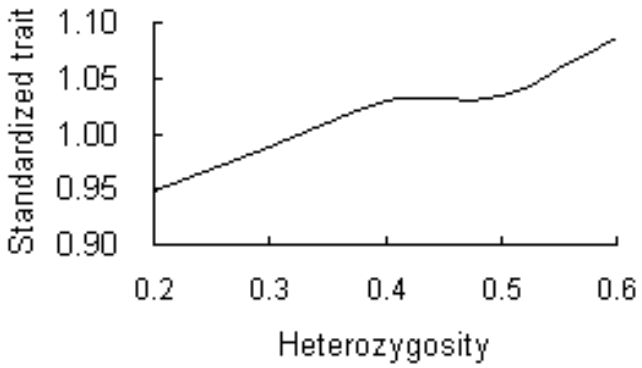

Figure 6. Changes of simulated maximum daily gain with microsatellite heterozygosity in females.

The mechanism for generating HFCs is not yet well understood. The observations of HFCs with neutral genetic markers prove that at least some correlations are due to the genetic association between the neutral markers and fitness-related genes. In this work, we used selectively neutral microsatellite loci to detect heterozygosity because they are highly mutable DNA markers that are particularly suitable for the detection of HFCs evolve in relation to short-term processes (Hughes and Queller, 1993; Jarne et al., 1996; Tsitrone et al., 2001), and significant HFCs had been found. Accordingly, the genomic pattern responsible for generating HFCs in this population should be local and/or general effects hypothesis.

The general effect hypothesis suggested HFCs because of the homozygosity at genome-wide distributed loci. According to this view, individual heterozygosity should be changed with inbreeding coefficient and there should be no heterozygosity-fitness correlation in population having no variation in inbreeding coefficient (David, 1998; Hansson and Westerberg, 2002). All chickens were half-sibs in this study, so we could suppose there was no difference in inbreeding coefficient among individuals. Different individual heterozygosity existed among this population and body weight correlated with heterozygosity in males and females, which implied that heterozygosity didn't

correlate with inbreeding coefficient (as found by Leary et al., 1987; Curik et al., 2003; Balloux et al., 2004; Markert et al., 2004; Slate et al., 2004). The results, on the surface therefore, could rule out the general effect hypothesis in explaining the HFCs in this chicken population.

Coltman and Slate (2003) reported that if HFCs exist, they will be better detected by individual heterozygosity than by d^2 values because heterozygosity usually provides higher correlations than that of d^2 under most biologically plausible conditions. In our study, no correlation was found between d^2 values and body weight in males and females. Rowe and Beebee (2001), in *Bufo calamita and Rana temporaria*, Shikano and Taniguchi (2002), in *Poecilia reticulata*, and Borrell et al. (2004), in *Atlantic salmon*, also found no association between fitness traits and d^2 measurements. Pujolar et al. (2005) even found negative relation between mean d^2 and growth rate. Mean d^2 is a useful indicator of inbreeding and related to more distant events in measuring the consequences of recent breeding patterns. In this way, positive correlations between some fitness components and mean d^2 should be found in species with high inbreeding coefficients (Coltman et al., 1998; Coulson et al., 1998), but not between these traits and microsatellite heterozygosity. No relationship between body weight and mean d^2 in this population indicated that inbreeding was not the cause of HFCs.

In this work, the general effect hypothesis could not explain the HFCs in the chicken population while the local effect hypothesis has been responsible for generating HFCs.

ACKNOWLEDGEMENT

This research was supported by National Nature Science Foundation of China (NSFC, 30300253) and Yangzhou University Foundation (NK0313102). The kind cooperation of workers in sampling is greatly acknowledged.

REFERENCE

Balloux, F., W. Amos and T. Coulson. 2004. Does heterozygosity estimate inbreeding in real populations? Mol. Ecol. 13:3021-3031.

Bierne, N., S. Launey, Y. Naciri-Graven and F. Bonhomme. 1998. Early effect of inbreeding as revealed by microsatellite analyses on *Ostrea edulis larvae*. Genet. 148:1893-1906.

Borrell, Y. J., H. Pineda, I. McCarthy, E. Vazquez, J. A. Sanchez and G. B. Lizana. 2004. Correlations between fitness and heterozygosity at allozyme and microsatellite loci in the Atlantic salmon, *Salmo salar L*. Heredity 92:585-593.

Coltman, D. W., W. D. Bowen and J. M. Wright. 1998. Birth weight and neonatal survival of harbour seal pups are positively correlated with genetic variation measured by microsatellites. Proc. R. Soc. Lond. B 265:803-809.

Coltman, D. W. and J. Slate. 2003. Microsatellite measures of inbreeding: A meta-analysis. Evolution 57:971-983.

Coulson, T. N., J. M. Pemberton, S. D. Albon, M. Beaumont, T. C. Marshall, J. Slate, F. E. Guinness and T. H. Clutton-Brock. 1998. Microsatellites reveal heterosis in red deer. Proc. R. Soc. Lond. B 265:489-495.

Curik, I., P. Zechner, J. Solkner, R. Achmann, I. Bodo, P. Dovc, T. Kavar, E. Marti and G. Brem. 2003. Inbreeding, microsatellite heterozygosity, and morphological traits in *Lipizzan* horses. J. Hered. 94:125-132.

David, P. 1998. Heterozygosity-fitness correlations: new perspectives on old problems. Heredity 80:531-537.

David, P., B. Deley, P. Berthou and P.Jarne. 1995. Alternative models for allozyme-associated heterosis in the marine bivalve *Spisula ovalis*. Genet. 139:1719-1726.

Hansson, B. and L. Westerberg. 2002. On the correlation between heterozygosity and fitness in natural populations. Mol. Ecol. 11:2467-2474.

Houle, D. 1989. Allozyme-associated heterosis in Drosophila melanogaster. Genet. 123:789-801.

Hughes, C. R. and D. C. Queller. 1993. Detection of highly polymorphic microsatellite loci in a species with little allozyme polymorphism. Mol. Ecol. 2:131-137.

Jarne, P. and P. J. L. Lagoda. 1996. Microsatellites, from molecules to populations and back. Trends Ecol. Evol. 11:424-429.

Jiang, X. P., G. Q. Liu and Y. Z. Xiong. 2005. Investigation of Gene and Microsatellite Heterozygosities Correlated to Growth Rate in the Chinese Meishan Pig. Asian-Aust. J. Anim. Sci. 18:927-932.

Jiang, X. P., Y. Z. Xiong, G. Q. Liu, C. Y. Deng and Y. C. Qu. 2003. Effects of individual gene heterozygosity on growth traits in swine. Acta Genetica Sinica 30:431-436.

Leary, R. F. 1987. Differences in inbreeding coefficients do not explain the associations between heterozygosity at allozyme loci and developmental stability in rainbow trout. Evolution 41:1413-415.

Markert, J. A., P. R. Grant, B. R. Grant, L. F. Keller, J. L. Coombs and K. Petren. 2004. Neutral locus heterozygosity, inbreeding, and survival in Darwin's ground finches (*Geospiza fortis and G. scandens*). Heredity 92:306-315.

Pogson, G. H. and S. E. Fevolden. 1998. DNA heterozygosity and growth rate in the Atlantic cod *Gadus morhua*. Evolution 52:915-920.

Pogson, G. H. and E. Zouros. 1994. Allozyme and RFLP heterozygosities as correlates of growth rate in the Scallop *Placopecten magellanicus*: a test of the associative overdominance hypothesis. Genet. 137:221-231.

Pujolar, J. M., G. E. Maes, C. Vancoillie and F. A. M. Volckaert. 2005. Growth rate correlates to individual heterozygosity in the European eel, *Anguilla Anguilla L*. Evolution 59:189-199.

Rowe, G. and T. J. C. Beebee. 2001. Fitness and microsatellite diversity estimates were not correlated in two outbreed anuran populations. Heredity 87:558-565.

Shikano, T. and N. Taniguchi. 2002. Relationships between genetic variation measured by microsatellite DNA markers and a fitness-related trait in the guppy (*Poecilia reticulata*). Aquac. 209:77-90.

Slate, J., P. David, K. G. Dodds, B. A. Veenvliet, B. C. Glass, T. E. Broad and J. C. McEwan. 2004. Understanding the

relationship between the inbreeding coefficient and multilocus heterozygosity: theoretical expectations and empirical data. Heredity 93:255-265.

Smouse, P. E. 1986. The fitness consequences of multiple-locus heterozygosity under the multiplicative overdominance and inbreeding models. Evolution 40:946-957.

Thelen, G. C. and F. W. Allendorf. 2001. Heterozygosity-fitness correlations in rainbow trout: effects of allozyme loci or associative overdominance? Evolution 55:1180-1187.

Tu, Y. J., K. W. Chen, S. J. Zhang, Q. P. Tang, Y. S. Gao and N. Yang. 2006. Genetic diversity of 14 indigenous grey goose breeds in China based on microsatellite markers. Asian-Aust. J. Anim. Sci. 19:1-8.

Tsitrone, A., F. Rousset and P. David. 2001. Heterosis, marker mutational processes and population inbreeding history. Genet. 159:1845-1859.

Wang, X., H. H. Cao, S. M. Geng and H. B. Li. 2004. Genetic diversity of 10 indigenous pig breeds in China by using microsatellite markers. Asian-Aust. J. Anim. Sci. 17:1219-1226.

Zhang, J. H., Y. Z. Xiong and C. Y. Deng. 2005. Correlations of genic heterozygosity and variances with heterosis in a pig population revealed by microsatellite DNA marker. Asian-Aust. J. Anim. Sci. 18:620-625.

Zouros, E. 1993. Associative overdominance: evaluating the effects of inbreeding and linkage disequilibrium. Genetica 89:35-46.