Hybrid vigour and maternal environment in mice. I. Body and brain growth

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Abstract:
Ovarian grafting and surrogate fostering were used to manipulate the pre- and postnatal maternal environments, respectively, in order that the inbred mouse strains BALB/c and C57BL/6J and their reciprocal F_1 hybrids experienced either an inbred or an F_1 hybrid environment pre- and/or postnatally. Results revealed sizeable heterotic as well as maternal environmental effects on birth, weaning and 100-day body weights as well as on brain weight at 100 days. The maternal environmental effect on brain weight was mediated by its effect on body weight, but there was heterosis for brain weights even when body weight was taken into account. At birth and weaning, inbreds appeared to benefit more from the hybrid maternal environment than did hybrids, but we found no evidence of increased variability of inbreds compared to hybrids (homeostasis) within experimental conditions.
Key words: Hybrid vigour; Maternal environment; Brain weight; Body weight; Ovarian grafting

Article:

Introduction

Inbreeding generally reduces growth, reproductive fitness and performance on a variety of behavioural tests, and its counterpart, hybrid vigour or overdominance, is often apparent when inbred strains are crossed (Falconer, 1981). Simple dominance at a single locus can yield overdominance for traits influenced by many segregating loci, but the increased phenotypic variability of highly inbred animals (Hyde, 1973; Lassalle, Medioni and Le Pape, 1979; Leamy, 1985; Wainwright, 1981) cannot be explained in this way. It has been hypothesized that inbreeding disrupts developmental homeostasis or "buffering" and thereby renders the developing organism more vulnerable to small variations in the environment (Hyde, 1973; Lerner, 1970).

The "buffering" metaphor actually hypothesizes genotype-environment interaction. It implies that variations in the early environment are similar for both inbred and hybrid animals, but that inbreds are more sensitive to these variations. This is not the only possible explanation, however. It is conceivable that the early maternal environment is more variable for inbred fetuses and neonates, so that instability of the mother's physiology or behaviour contributes to greater phenotypic differences among her offspring. Alternatively, the phenotypic variation among animals with the same genotype may not arise from the external environment; rather, it may reflect regulatory processes internal to the developing organism which give rise to non-genetic and non-environmental variation (Wahlsten, 1987) that constitutes epistatic interaction among several loci rather than genotype-environment interaction. These alternative explanations cannot be evaluated with purely genetic techniques. Instead, an experimental approach is required whereby inbred and hybrid animals are reared in distinctly different environments.

In laboratory mammals such as mice, the early environment is largely provided via the mother. Reciprocal hybrid crosses have demonstrated that the inbred maternal environment is unfavourable for growth of the body and brain and for several kinds of behaviour (McLaren, 1981; Wahlsten, 1983; Wahlsten & Wainwright, 1980). Unfortunately, reciprocal crosses alone cannot yield inbred or F_1 hybrid fetuses in a hybrid prenatal maternal environment and they cannot separate prenatal from postnatal effects.

One solution is ovary grafting (Jones & Krohn, 1960; Krohn, 1977), in which ovarian follicle cells of an inbred BALB female, for example, are placed into the ovarian capsule of an ovariectomized hybrid female, provided the hybrid female has the histocompatibility alleles of the inbred donor. If the hybrid host female is then mated to a BALB male, offspring will be genetically BALB in a hybrid mother. If, on the other hand, the hybrid female with BALB ovaries is mated with a C57BL/6 inbred male, the offspring will be F_1 hybrids in a hybrid mother. Prenatal versus postnatal contributions and their interactions with genotype can then be assessed by fostering at birth (Carlier & Nosten, 1987).

The present study combined ovary grafting and surrogate fostering to compare inbred and F_1 hybrid mice in inbred and F_1 pre- and postnatal maternal environments using the design shown in Table 1. This made it possible to ask whether inbred mice were indeed more sensitive to particular differences in the maternal environment. Several measures of growth, behaviour and brain anatomy were obtained at various ages.

The results of this project are presented in three parts concerning (I) measures of body and brain growth, (II) behaviour, and (III) hippocampal anatomy in relation to behaviour. Part I involves 337 mice with complete data at 100 days after birth, whereas Part II presents only the 300 mice with complete behavioural data and Part III presents only the 103 with adequate histological detail of the hippocampus. The rationale for measures and technical problems are presented in the pertinent part.

The data on growth were relevant for the project in two ways. First, they showed that the experimental manipulations of maternal environment did indeed affect the developing mice and, therefore, the failure to detect maternal effects on certain behaviours was not a trivial result of nearly identical treatments. Second, they assessed the generality of an earlier demonstration of a hybrid maternal environment effect on the BALB mouse brain (Bulman-Fleming and Wahlsten, 1988).

TABLE 1

Number of pups (N) and mean litter size (at weaning).

Genotype	Prenatal maternal environment	Postnatal maternal environment			
		Inbred		F_1 Hybrid	
		Litter size	N	Litter size	N
BALB/c	BALB/c	5.3	32	5.4	28
	F_1 Hybrid	3.6	14	4.1	16
C57BL/6J	C57BL/6J	5.7	29	5.1	40
	F_1 Hybrid	3.9	11	6.5	13
BALB × C57	BALB/c	4.9	18	4.5	26
	F_1 Hybrid	3.6	7	2.0	4
C57 × BALB	C57BL/6J	6.0	33	7.1	43
	F_1 Hybrid	7.0	7	8.8	22

Methods

Design

The design for this experiment was a 4 × 2 × 2 factorial wherein four genotypes experienced either an inbred or a hybrid prenatal maternal environment (ME) followed by either an inbred or hybrid postnatal ME (see Table 1). The four genotypes were BALB/cWah2, C57BL/6J, and the two reciprocal crosses resulting from BALB × C57 matings; BALB × C57 and C57 × BALB. In the case of crosses the female is designated first: i.e. C57 (female) × BALB (male).

All mice were derived from grafted ovaries and all were fostered to ungrafted lactating females within 48 h of birth. The inbred pre- and postnatal ME for BALB and BALB × C57 was always BALB and for C57 and C57 × BALB was always C57. The hybrid pre- and postnatal ME for all genotypes was either BALB × C57 or C57 × BALB.

Occasionally, small fragments of host ovarian tissue, which were inadvertently left behind during the ovariectomy procedure preceding the grafting of donor ovaries, produced contaminant offspring of host rather than donor origin. Pigmented BALB mice, homozygous for the wild allele at the locus for albinism, were used to produce the F_1 females which were grafted. In this way, unequivocal identification of the origin of albino

offspring from F_1 mothers grafted with albino ovaries was ensured (Bulman-Fleming and Wahlsten, 1988). Unfortunately, unequivocal identification of offspring genotype using a coat color marker was not possible for three of the remaining seven combinations of genotype and prenatal ME (C57 in hybrid ME, BALB × C57 in hybrid ME, and C57 × BALB in hybrid ME). None of the members of litters containing one or more contaminant mice (those whose coat color identified them as being of host ovary origin) was used in the study.

Animals

The inbred strains BALB/cWah2 and C57BL/6J were maintained using full-sibling mating pairs in D.W.'s laboratory at the University of Waterloo. The F_1 hybrid females used for ovarian grafting were offspring of reciprocal crosses between C57BL/6J mice and pigmented BALB mice homozygous for the wild allele at the albinism locus. The origins of the BALB/cWah2, pigmented BALB, and C57BL/6J strains have been outlined in earlier reports (Bulman-Fleming and Wahlsten, 1988; Wahlsten & Bulman-Fleming, 1987). The ungrafted F_1 hybrid females used as surrogate mothers were derived from crosses between C57BL/6J mice and either pigmented or albino BALB mice. All animals were maintained under standard laboratory conditions which have been described previously (Wahlsten, 1982).

Ovary grafting and surrogate fostering

The grafting procedure was essentially that used by Jones and Krohn (1960) with only minor adaptations (Bulman-Fleming, 1988). All animals were between 6 and 10 weeks of age at the time of the operations, and were allowed to recover for at least two weeks prior to being placed with a male. Inbred or hybrid animals experiencing inbred prenatal environments were produced by autografts (donor and host of the same genotype) of BALB and C57 ovarian tissue into BALB and C57 hosts, which were then mated to males of the same or to those of the different inbred strain, respectively. To produce inbreds or hybrids in an F_1 prenatal ME, inbred ovaries were grafted into F_1 host dams which were subsequently mated, as above, to males of either the same or the other inbred strain.

As soon as possible after birth, pups were weighed to the nearest mg and then were fostered to ungrafted lactating mothers who had themselves given birth within 48 h of the birth of the "grafted" litter. The foster mother's own pups were euthanized. One C57 dam bit several of her foster pups within seconds after they had been placed into the cage. The unbitten pups were removed immediately and were placed with a different mother, and the C57 dam and injured pups were sacrificed. With the exception of this one incident, all foster mothers readily accepted their new litters.

Except for cage cleaning, litters were left undisturbed until weaning, which occurred when pups were 21 days of age. At this time, pups were weighed to the nearest 0.1 g and thereafter housed with siblings of the same sex.

Histology and fibre tract measurement

At between 98 and 102 days of age, each animal was weighed, anaesthetized with an overdose of sodium pentobarbital, and perfused intracardially with 0.9% saline followed either by buffered 10% formalin or by 0.1% sodium sulfide and 3% glutaraldehyde in 0.15 M Sorensen phosphate buffer. The latter perfusion was in preparation for the Timm's sulfide silver stain for hippocampal mossy fibres (Danscher, 1981) and was performed on two animals from each litter (one male and one female where possible).

In the case of the Timm's perfusions, the brains were trimmed to remove olfactory bulbs, paraflocculi, optic nerve, and spinal cord, and were weighed to the nearest mg immediately after having been removed from the skull. These brains were then stored in the glutaraldehyde fixative for 1-2 h prior to being placed in vials containing a solution of 30% sucrose. The brains fixed with formalin were stored for one week in jars containing an excess of formalin before being trimmed and weighed as above.

All brains were bisected with a razor blade at the interhemispheric fissure; then 33Um frozen sections were cut from the left hemisphere in the sagittal plane and were stained with Sudan Black B for myelin. Midline areas of corpus callosum, hippocampal commissure and anterior commissure were determined by digitized

morphometry (R & M Biometrics, Nashville, TN) from drawings made using a Leitz tracing device. At the time of perfusion, the brains were coded so that the experimenters were blind to the genotype and experimental condition of the animals when trimming and weighing brains as well as for subsequent measurement of fibre tract areas. The brains which were fixed for Timm's staining were included in the brain weight analyses but not in the analyses of midline fibre tract areas, because the very poor quality of the frozen sections with Sudan Black B staining precluded accurate measurements in most cases.

Data analysis

The data were analyzed using the MGLH (multivariate general linear hypothesis) module of SYSTAT (Wilkinson, 1986). The unit of analysis was the individual pup score, because an issue of primary interest was the correlation of individual fibre tract areas with brain weights. Multiple regression using effect coding was the method of analysis in this study. The following terms were used: STR (strain: BALB + 1, C57 — 1); HET (heterosis: hybrids + 1, inbreds — 1); RECIP (reciprocals: BALB × C57 + 1, C57 × BALB — 1); PRE (prenatal ME: hybrid + 1, inbred — 1); POST (postnatal ME: hybrid + 1, inbred — 1); SEX (sex: males + 1, females — 1).

Results

In total, 30 mice (15 BALB and 15 hybrid) were grafted with BALB ovaries and 29(14 C57 and 15 hybrid) with C57 ovaries for this experiment. Eight grafted females and 12 litters were destroyed because of the above-mentioned problems with contaminant host ovarian tissue which produced viable ova. Notwithstanding this problem, the sixteen combinations of genotype, pre- and postnatal ME resulted in 361 and 337 mice at birth and at 100 days of age, respectively (see Table 1).

Body weight at birth

A multiple regression analysis of birth weight (including terms for strain, heterosis, reciprocals, prenatal ME, litter size, and First-order interactions between the genotype contrasts and both prenatal maternal environment and litter size) yielded a squared multiple R of 0.583. Heterosis, maternal environment, and litter size were all highly significant (all $p < 0.01$, two-tailed). The directions of the effects were as predicted; hybrid pups weighed more than inbred pups, pups with hybrid mothers weighed more than those with inbred mothers, and pups from smaller litters were larger than pups from larger litters. There was a suggestion of an interaction between heterosis and maternal environment ($p = 0.09$, one-tailed), such that inbred pups benefitted more from an F_1 hybrid prenatal ME than did hybrid pups. The difference in average birth weight between inbred pups whose mother was hybrid and inbred pups with an inbred mother was about 0.1 g, and the difference for hybrid pups was 0.06 g. No other interaction terms were significant. When the insignificant terms were dropped, the squared multiple R fell to 0.562. The results of this analysis are summarized in Table 2.

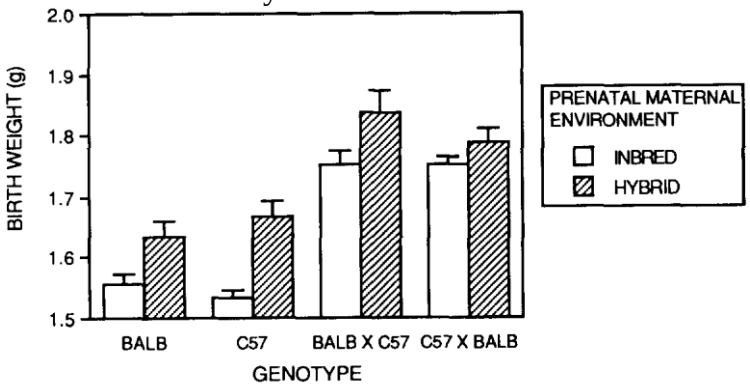

Fig. 1. Body weight at birth. Bars indicate 1 standard error of the mean.

The analysis of birth weight data did not include runts (body weight less than 1.2 g, n = 8) nor two small litters of F_1 animals (n = 6) which received very poor care at birth and were judged to be outliers.

For presentation in Figure 1, body weights at birth were adjusted to a common litter size of 5, using a common slope of —0.05. This factor was derived from results of a linear regression of birth weight on litter size in the

present sample, as well as from the results of a previous experiment which employed a very large sample size (Bulman-Fleming & Wahlsten, 1988). As can be seen from Figure 1, the inbreds were no more variable than the hybrids, irrespective of the maternal environment.

TABLE 2

Results of multiple regressions on body weights at 3 ages and brain weight at 100 days of age [a].

	Variables	df [b]	Coefficient	Multiple R^2	SEE	Overall Regression
Body Weight at Birth	LSB	351	−0.054 **	0.562	0.131	***
	PRE		0.042 **			
	HET		0.101 **			
Body Weight at Weaning	LSW	334	−0.829 ***	0.809	1.103	***
	POST		1.228 ***			
	SEX		0.214 ***			
	STR		0.439 ***			
	HET		0.543 ***			
	RECIP		0.499 ***			
	STR × POST		−0.467 ***			
	HET × POST		−0.174 **			
	RECIP × POST		0.372 ***			
Body Weight at 100 Days	LSW	329	−0.530 ***	0.769	1.738	***
	POST		0.691 ***			
	SEX		2.508 ***			
	STR		0.687 ***			
	HET		1.685 ***			
	RECIP		0.512 **			
	STR × SEX		0.439 **			
Brain Weight at 100 Days for Males	FIX	142	25.57 ***	0.858	12.22	***
	STR		5.75 ***			
	HET		5.33 ***			
	RECIP		3.28			
	POST		1.87			
	BOD		3.97 ***			
Brain Weight at 100 Days for Females	FIX	181	27.11 ***	0.788	15.97	***
	STR		−0.39			
	HET		4.22 **			
	RECIP		4.83 *			
	POST		1.78			
	BOD		6.94 ***			

[a] Abbreviations: PRE and POST—prenatal and postnatal maternal environment, respectively: hybrid = +1, inbred = −1; LSB and LSW—litter size at birth and at weaning, respectively; BOD—body weight; FIX—fixative: Formalin = +1, Timm's = −1; STR—strain: BALB/c = +1, C57 = −1; HET—heterosis: hybrid = +1, inbred = −1; RECIP—reciprocals: BALB × C57 = +1, C57 × BALB = −1; SEE—standard error of estimate; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ two-tailed.

[b] Degrees of freedom for the residual.

Body weight at weaning

For body weight at weaning, a multiple regression similar to that described for birth weight was performed, with the addition of terms for sex and postnatal ME (all $p < 0.01$, two-tailed). The directions of the effects were the same as for birth weights. All genotype contrasts were significant, as were terms for litter size, sex, postnatal ME, and the three first order interactions involving genotype contrasts and postnatal ME. For the strain variable, which was not significant at birth, the direction of the effect was such that BALB were heavier than C57. At this stage, there was no effect of the prenatal ME on body weight. The squared multiple R for this analysis was 0.810. When prenatal ME was eliminated from the equation, the squared multiple R was 0.809 (see Table 2). The interaction between postnatal ME and heterosis was consistent with the trend observed for birth weights, and at this stage the effect was significant. In other words, body weight differences between pups experiencing an inbred and those experiencing a hybrid maternal environment were greater for inbreds than for hybrids. These results could be interpreted as evidence of developmental homeostasis (Lerner, 1954). With respect to the strain by postnatal ME and reciprocals by postnatal ME interactions, both C57 and C57 × BALB mice in the inbred postnatal ME condition experienced a C57 maternal environment, whereas BALB and BALB × C57 mice were reared by BALB dams (see Methods). The regression results indicated that there was a greater difference in weaning weights between C57 mice in the inbred and the hybrid postnatal ME conditions than there was between BALB in these two conditions. Similarly, the difference between C × B weights in the two

ME conditions was greater than the difference between B × C weights in the inbred vs. the hybrid ME condition. Wahlsten (1983) has previously reported brain and body weight differences between reciprocal F_1 hybrids of BALB and C57. He demonstrated that the effects observed were a result of the superiority of the BALB pre- and/or postnatal ME. It would seem, from the present results, that the postnatal ME is the more critical, at least with regard to body weight at weaning.

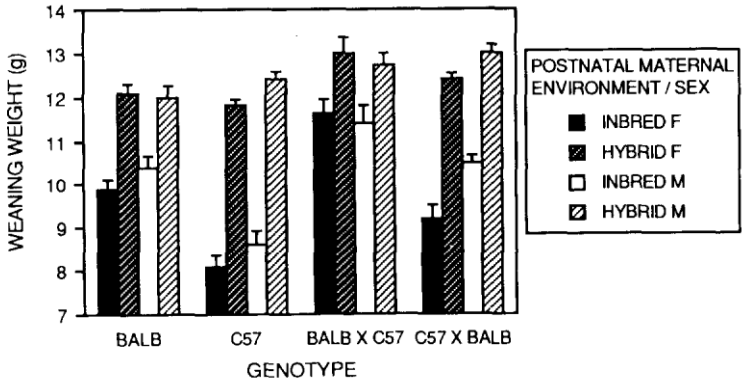

Fig. 2. Body weight at weaning. Bars indicate 1 standard error of the mean.

Weaning weights, adjusted to a common litter size of 5 using a common slope of —0.83, are presented in Figure 2. Here again, no less variability is evident for the F_1 hybrid pups than for the inbred strains.

Body weight at 100 days

Preliminary analyses of 100-day brain weights and body weights confirmed that the effect of the prenatal ME was no longer evident, so the 100 day data were pooled across prenatal ME for subsequent statistical tests.

Results of the multiple regression of 100-day body weight can be found in Table 2. The effects of sex, litter size, postnatal ME, strain, heterosis, and reciprocals were all highly significant (reciprocals $p < 0.01$; all others $p < 0.001$, two-tailed). The directions of the effects were consistent with those observed for weaning weights. There was also a significant interaction between strain and sex, in which, for females only, BALB animals were heavier that C57s. 100-day body weights were adjusted to a common litter size of 5 using a slope of —0.54 for both sexes. Inspection of these adjusted weights (not shown) revealed that, in contrast to the effect seen at birth and at weaning, in which body weight differences between inbred and hybrid ME conditions for inbred animals were greater than the differences for hybrid animals, by 100 days of age the differences were equivalent. Thus, the question of differential sensitivity to the maternal environment is not a simple one, but must be evaluated as a function of each stage in the organism's development.

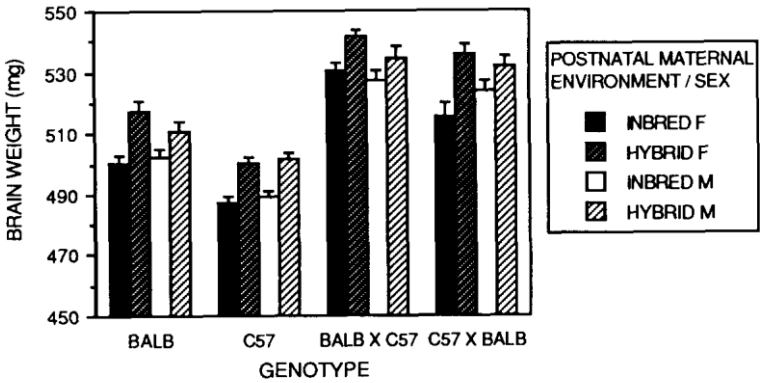

Fig. 3. Brain weight at 100 days. Bars indicate 1 standard error of the mean.

Brain weight at 100 days

As a result of large differences between weights of brains from animals perfused with formalin and those of animals perfused by the Timm's method, the multiple regression analysis included a term for the type of fixation. Coefficients obtained from regressions for each sex of brain weight on fixation, litter size at weaning, postnatal maternal environment and the three genotype contrasts were used to generate brain weights which

were adjusted to a common litter size of 5 and which corrected for effects of fixation. These adjusted brain weights are graphed in Figure 3.

In order to determine whether the postnatal maternal environment exerted any effect on brain weight over and above its effect mediated by body weight, regressions were performed again, but this time body weight was substituted for litter size. (Including both these independent variables would have resulted in problems with multicolinearity—see Pedhazur, 1982). For both males and females, when body weight was accounted for, there was no longer a significant effect of the postnatal maternal environment, nor was there a strain effect for females. In other words, there was no effect of the postnatal ME on brain weight over and above its effect on overall growth. The reciprocal effect, though still significant for females and almost significant for males, was weak, but heterosis was still significant ($p < 0.01$, two-tailed) for both females and males, even when body weight was taken into account. These results confirm and extend previous observations in our laboratory in which BALB mice were reared in either a BALB or an F_1 hybrid ME pre- and postnatally (Bulman-Fleming and Wahlsten, 1988).

TABLE 3

Means (±SD) of fibre tract areas (mm^2) [a].

	Genotype:	BALB/c		C57		F_1 [b]	
	Postnatal ME:	I	F_1	I	F_1	I	F_1
	N:	22	22	22	33	40	63
CC	MEAN	0.825	0.928	1.074	1.120	1.154	1.170
	SD	0.287	0.290	0.071	0.067	0.067	0.056
HC	MEAN	0.277	0.287	0.269	0.286	0.288	0.291
	SD	0.024	0.020	0.022	0.022	0.027	0.026
AC	MEAN	0.150	0.147	0.142	0.156	0.165	0.165
	SD	0.013	±0.011	0.009	0.012	0.014	0.014

[a] Abbreviations: Postnatal ME—postnatal maternal environment; I—inbred; F_1—F_1 hybrid; CC—corpus callosum; HC—hippocampal commissure; AC—anterior commissure.
[b] Results are pooled across reciprocal F_1 hybrids.

Fibre tract areas

Because of the poor tissue quality of the stained frozen sections from brains perfused by the Timm's method, analysis of commissure areas of only the 202 mice perfused with formalin are presented. Preliminary tests indicated neither effects of sex nor differences between the two reciprocal F_1 hybrids, so for the following analyses, males and females within each genotype were pooled, as were the F_1 hybrids. Table 3 presents mean values for the midline areas of the 3 fibre tracts.

Separate multiple regressions were performed for each of the three genotypes (BALB, C57 and pooled F_1 hybrids) because of extreme differences in the variances of corpus callosum area at midplane (Wahlsten, 1989). (In the BALB inbred strain the midline area of the corpus callosum of about 20-25% of the animals is very much reduced in size and in some cases is actually absent—note standard deviations in Table 3.) For BALB, the results of the regressions (not shown) revealed that neither the maternal environment nor brain weight was important in the determination of adult corpus callosum area, but brain weight was correlated with the midline area of the anterior commissure and, to a lesser extent, with that of the hippocampal commissure. On the other hand, for C57 the hybrid postnatal maternal environment was favorable to midline commissure area growth independent of its effect on brain weight. All three midline fibre tract areas were correlated with brain weight for the hybrids, and maternal environment was not an important factor. Multiple regression analysis revealed that the larger mean values of midline commissure areas for the hybrids as compared to inbreds was due to their larger brains.

Discussion

Results from this study clearly demonstrate the existence of sizeable heterotic and maternal environmental effects on birth and weaning weight as well as on 100-day body and brain weight. The brain weight effect was mediated through an effect on whole body growth. Heterosis is a well-known phenomenon and has been

demonstrated for behavioural (Bruell, 1964; Lassalle, Medioni and Le Pape, 1979; Wainwright, 1980, 1981; see also review by Hyde, 1973) as well as morphometric (Hahn and Haber, 1978; Henderson, 1973; Leamy, 1982, 1985; Wahlsten, 1983; Wainwright, 1980) characteristics. The existence of maternal heterosis is also well established, though in studies employing non-inbred strains the magnitude of these effects is difficult to assess because of the presence of genetic variability both of mothers and of offspring (Bandy and Eisen, 1984; Leamy, 1988; Riska, Rutledge and Atchley, 1985). Our design, employing strains with virtually no genetic variability (inbred strains and their F_1 hybrid offspring) afforded advantages in this respect, but because of the complexity and scope of the experiment sample sizes were quite dissimilar.

There is no general agreement in the literature regarding the relative importance of prenatal vs. postnatal maternal environmental factors. The phenomenon is without doubt complex: effects vary depending on the age of the pups and the characteristic of interest. Also, the genotype of the pup can markedly influence the maternal performance both pre- and postnatally (Brumby, 1960). In this study, we found no significant effect of uterine (prenatal) environment of the mother on weaning or on 100-day body or brain weight. The data of Leamy (1988), on the other hand, would argue for a significant prenatal influence on brain, if not on body size. This inconsistency could be explained by the fact that Leamy used random-bred mice, as well as different methods of determining brain weights and different techniques for analysis. In this regard, it is instructive to compare the present results with those from a previous experiment conducted in our laboratory which employed BALB mice either raised pre- and postnatally by BALB or by F_1 hybrid mothers derived from reciprocal crosses between BALB and C57 matings (Bulman-Fleming and Wahlsten, 1988). In the prior experiment, brain weights of BALB animals experiencing a hybrid pre- and postnatal maternal environment were 5 to 6 mg heavier than those of animals that had inbred mothers. The comparable differences in this study, for which results were pooled over the two prenatal conditions, were 13, 11,9, and 14 mg for BALB, C57, BALB × C57 and C57 × BALB, respectively. If the prenatal component were important, we would have expected *smaller* differences in the present study. The inbred vs. hybrid differences for body weight showed a similar pattern between the two experiments: namely, those in the present study were almost twice the magnitude of those in the earlier experiment (1 to 2 g vs. 0.5 to 1 g, respectively). In the present experiment, of course, all mice were derived from grafted ovaries and were fostered at birth, manipulations which in themselves could conceivably account for these differences. However, results from other similar experiments in which there were controls for grafting (Brumby, 1960; Bulman-Fleming and Wahlsten, 1988) and for fostering (Brumby, 1960) suggest that this is unlikely.

Our results do not show decreased within-strain variability with increased heterozygosity (developmental homeostasis—Lerner, 1954) as reported by some other investigators (Hyde, 1973; Leamy, 1985; Soulé, 1979; Wainwright, 1981). This phenomenon is extremely complex and would appear to be a function of developmental age (Leamy, 1985) as well as of the particular trait of interest (Hyde, 1973).

In contrast to the apparent lack of increased within-group variability in the inbreds, the present results do indicate that inbred pups (at least at 21 days of age, and possibly also at birth) are more sensitive to the rather large maternal environmental differences provided by inbred vs. hybrid mothers.

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