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A possible role for imprinted genes in inbreeding avoidance and dispersal from the natal area in mice

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The expression of a subset of mammalian genes is subject to parent of origin effects (POE), most of which can be explained by genomic imprinting. Analysis of mutant animals has demonstrated that a number of imprinted genes influence brain development and behaviour. Here we provide evidence for POE on olfactory related behaviour and sensitivity to maternal odour cues. This was investigated by examining the odour preference behaviour of reciprocal cross F_1 mice made by embryo transfer to genetically unrelated foster parents. We determined that both adult males and females show an avoidance of female urinary odours of their genetic maternal but not paternal origin. This was found not to be due to any previous exposure to these odours or due to self-learning, but may be related to direct effects on the olfactory system, as reciprocal F_1 males show differential sensitivity to female odour cues.

Currently the most robust theory to explain the evolution of imprinting is the conflict hypothesis that focuses on maternal resource allocation to the developing foetus. Kinship considerations are also likely to be important in the selection of imprinted genes and we discuss our findings within this context, suggesting that imprinted genes act directly on the olfactory system to promote post-weaning dispersal from the natal area.

Keywords: parent of origin effects; genomic imprinting; olfaction; inbreeding avoidance; mice

1. INTRODUCTION

There is a growing body of evidence demonstrating that imprinted genes, those genes that are subject to parent of origin specific expression, influence brain and behavioural phenotypes (Keverne 1997; Isles & Wilkinson 2000). In studies of mice, imprinted genes have been implicated in a range of behaviours, including maternal behaviour (Lefebvre *et al.* 1998; Li *et al.* 1999), long-term memory (Brambilla *et al.* 1997), and suckling (Cattanach & Beechey 1990).

Our own studies following the fate of parthenogenetic and androgenetic cells in chimeras demonstrated that these cells have distinct developmental fates within the brain, a finding that indicates that paternally and maternally expressed imprinted genes show distinct expression within the brain and may have contrasting functions (Allen *et al.* 1995; Keverne *et al.* 1996). However, both uniparental cell types contributed to the main olfactory and vomeronasal systems, and in particular to the receptor neurones themselves (Allen *et al.* 1995). This is noteworthy, given research demonstrating that olfactory receptor genes are only expressed from one allele (Chess *et al.* 1994). In addition to these neuronal findings, adult normal–parthenogenetic chimeric mice also demonstrated a change in aggression, as measured by latency to attack (Allen *et al.* 1995). This aggressiveness was positively correlated with the contribution of parthenogenetic cells to the chimera. Aggressive behaviour in mice is mediated in part by olfactory cues (Mugford & Nowell 1970), and this, coupled with the fact that uniparental cells contribute directly to the olfactory receptor neurones, suggests that imprinted genes may affect olfactory system functioning.

Olfaction is the main sensory modality in mice, and social behaviour is predominantly controlled by odour cues (Hurst 1990*a*,*b*,*c*). Many of these behaviours rely upon kin recognition mechanisms, which are provided by information in odour cues, often carried by the urine.

We have shown recently that parent of origin effects (POE) influence olfactory related behaviour (Isles *et al.* 2001). Here, we provide further evidence to support this, and also examine the level at which imprinted genes may be exerting this influence. We also go on to discuss the possible functional role that imprinting has to play in the olfactory system, focusing on natal dispersal and inbreeding avoidance.

2. MATERIAL AND METHODS

(a) *Subjects*

The inbred strains used to generate reciprocal F_1 mice were C57Bl/6 (B6) and CBA/Ca (CBA), and C57Bl/6 and DBA. Reciprocal F_1 mice were generated by pairing males with superovulated females, zygotes were then recovered and transferred to pseudopregnant females of the outbred strain CD1. Offspring

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from the two crosses were kept separate at all times. The F_1 mice were weaned at three weeks post-partum and housed in same-sex groups. After puberty and prior to testing, mice were housed singly under reversed-cycle lighting (12 D : 12 L). Animals were 6 to 10 weeks old when tested. Bilateral castration was carried out on some males via a single midline incision using Ketamine (130 mg kg^{-1}) and Xylazine (13 mg kg^{-1}) anaesthesia. Following gonadectomy, mice continued to be housed in same-sex groups for two months prior to behavioural testing. Females were always tested in the preference test when in proestrus/oestrus.

The choice of urine odours in the preference tests was between that of a genetic parental strain and that of a genetically unrelated strain (Balb/C). This provided a conservative comparison, unbiased by parental factors that would have been present in mother versus paternal sister, or father versus maternal brother choices. The particular urinary odours presented had not been encountered prior to these tests and were equally unfamiliar. To ensure these factors remained constant throughout all tests, different cohorts of embryo-transferred mice were used in each part of the study.

(b) *Behaviour*

Odour preference tests were performed in a Perspex testing arena divided into three equally sized chambers. Animals were habituated to the conditions and apparatus for 5 min. All tests involved application of 10 µl of urine to a disc of filter paper. The papers were placed in petri dishes at each end of the testing arena, and a wire mesh was placed over each dish so that subjects could smell volatile odours from the urines, but not gain direct access, thus preventing vomeronasal sensory input. The time an animal spent in either choice chamber was then measured over a total 5 min test period. Time readings for the investigation time were taken 2 min into the test and at the end (5 min), although all data presented here are for the 2 min timepoint. Individuals were tested on three separate days for a particular preference. Mean scores were calculated for each subject, and these were used to compute group means. The oestrus cycle of female subjects was monitored by vaginal smear; all females were tested when in vaginal late proestrus/oestrus. Males were tested with urine that was collected and pooled from 4 to 5 females in late proestrus/oestrus (confirmed by a vaginal smear). Females were tested with urine from females maintained in dioestrus by a subcutaneous progesterone implant to mimic pregnancy. Male urinary samples were also pooled according to strain.

Habituation–dishabituation tests (Gregg & Thiessen 1981; Sundberg *et al.* 1982) were performed in the animals' home cages. Animals were exposed to odours sequentially for a 2 min period, with a 1 min non-exposure period intervening between each odour presentation. Filter paper soaked with 10 µl of a given urinary odour was secured to a plastic weighing boat and placed inverted on the lid of the cage. Subjects were unable to make physical contact with the filter paper using either their snout or paws. The time the mice spent rearing beneath the weighing boat with their noses through the bars of the cage was taken as investigation time. Initially, mice were exposed three times to water to allow them to habituate to the procedure. One odour was then presented three successive times and dependent on the requirements of the test was followed by three presentations of the second odour. If two urinary odours were used in the test, the order of presentation of these two odours was randomized to account for any order effects.

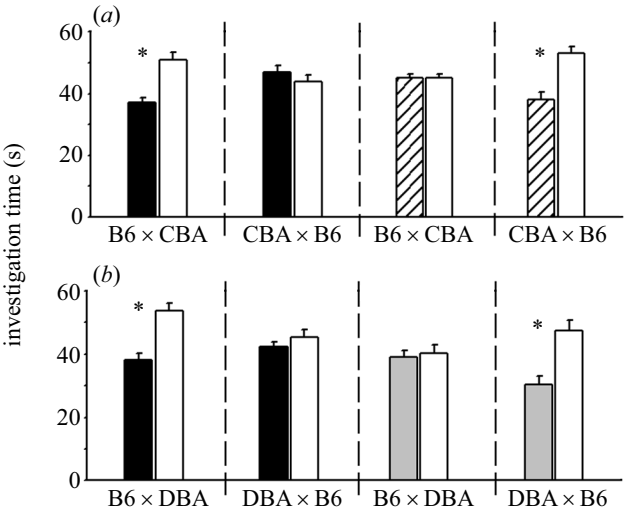

Figure 1. Preferences of reciprocal cross F_1 mice for female urine. Our previous study with B6 × CBA reciprocal F_1 mice had demonstrated that males and females had the same preference with regards to female paternal strain and maternal strain odour cues (Isles *et al.* 2001) and consequently the data presented here are pooled male and female data. (*a*) Preference of F_1(B6 × CBA) and F_1(CBA × B6) mice between B6 and Balb/C urine, and CBA and BALB urine. (*b*) Preference of F_1(B6 × DBA) and F_1(DBA × B6) mice between B6 and Balb/C urine, and DBA and BALB urine. (Black bars, B6 urine; white bars, Balb/C urine; hatched bars, CBA urine; grey bars, DBA urine.) Values shown are means ± s.e. Asterisks indicate $p > 0.05$.

(c) *Statistics*

Preference test measures were analysed using Wilcoxon signed-rank tests. Habituation–dishabituation test measures were analysed using paired, one-tailed *t*-tests. All statistical analysis was carried out either using preprogrammed tests within, or by programming equations into Microsoft Excel and then using statistical tables to generate *p* values.

3. RESULTS

(a) *Parent of origin effects on preferences for odours*

(i) *F_1 mice have reduced preference for female maternal-strain odours*

Male and female reciprocal cross F_1 mice (CBA × B6 and vice versa—maternal strain given first) were tested for their preference between female urinary odours of either their genetic maternal or paternal strain, and an unrelated standard (Balb/C). A preference for the standard urinary odour over odours from their maternal strain was found (Wilcoxon signed-rank test: F_1(B6 × CBA), $n = 20$, $p < 0.01$; F_1(CBA × B6), $n = 20$, $p < 0.01$; figure 1*a*). This avoidance of maternal odour occurred regardless of whether the genetic maternal strain was CBA or B6. A separate group of F_1 mice showed no preference when given the choice between a standard and their paternal strain female urinary odours (Wilcoxon signed-rank test: F_1(B6 × CBA), $n = 22$, $p \geqslant 0.1$; F_1(CBA × B6), $n = 21$, $p \geqslant 0.1$). This pattern of preference was also seen in a repeat experiment using a separate reciprocal cross (DBA × B6 and vice versa; figure 1*b*). Male and female

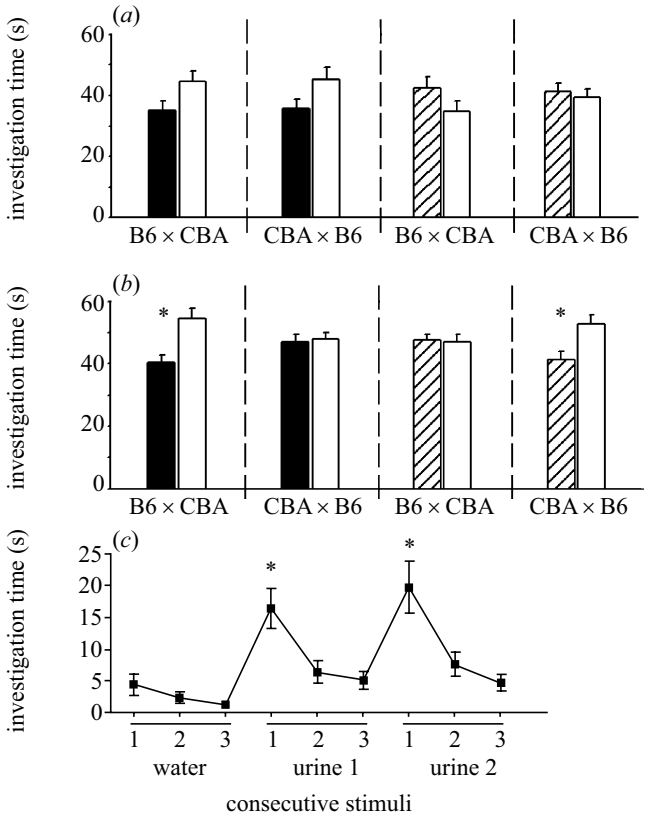

Figure 2. (*a*) Preference of F_1(B6 × CBA) and F_1(CBA × B6) females between B6 and Balb/C male urine, and CBA and Balb/C male urine. (*b*) Preference of F_1(B6 × CBA) and F_1(CBA × B6) females between B6 and Balb/C dioestrous female urine, and CBA and Balb/C dioestrous female urine. (Black bars, B6 urine; white bars, Balb/C urine; hatched bars, CBA urine.) (*c*) Habituation–dishabituation tests showing that reciprocal cross F_1 (B6 × CBA, and vice versa) females can discriminate between B6 and CBA male urine. Urine 1 indicates the first odour presented (either B6 or CBA urine), urine 2 indicates the second (either CBA or B6 respectively). Values shown are means ± s.e. Asterisks indicate $p > 0.05$.

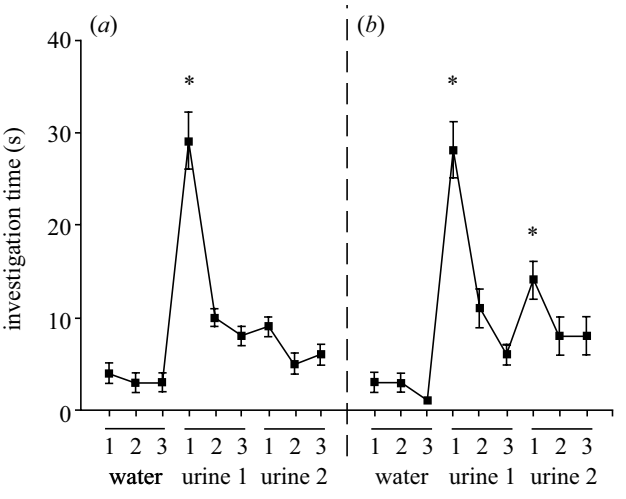

Figure 3. (*a*) Habituation–dishabituation tests of Balb/C females with male urinary odours from reciprocal crosses between CBA and B6. Urine 1 indicates the first odour presented (either F_1(CBA × B6) or F_1(B6 × CBA) urine), urine 2 indicates the second (either F_1(B6 × CBA) or F_1(CBA × B6) respectively). (*b*) Habituation–dishabituation using the same group of mice, this time tested with CBA and B6 urine. Urine 1 indicates the first odour presented (either B6 or CBA urine), urine 2 indicates the second (either CBA or B6 respectively). Values shown are means ± s.e. Asterisks indicate $p > 0.05$.

mice of these crosses also preferred the urinary odours of the unrelated standard to odour cues from their genetic maternal strain (Wilcoxon signed-rank test: F_1(B6 × DBA), $n = 22$, $p < 0.01$; F_1(DBA × B6), $n = 10$, $p < 0.01$), but not their paternal strain (Wilcoxon signed-rank test: F_1(B6 × DBA), $n = 22$, $p > 0.1$; F_1(DBA × B6), $n = 10$, $p > 0.1$).

(ii) *F_1 female mice show no preference between male parental strain and unrelated urinary odours*

A distinct group of reciprocal cross F_1 females (B6 × CBA and vice versa) were tested for their preference between male urinary odours of their genetic parental strains. None of these F_1 females showed any pattern of preference between either male paternal strain and standard urinary odour (figure 2*a*) (Wilcoxon signed-rank test: F_1(B6 × CBA), $n = 8$, $p \geqslant 0.1$; F_1(CBA × B6), $n = 8$, $p \geqslant 0.1$) or male maternal strain and a standard urinary odour (Wilcoxon signed-rank test: F_1(B6 × DBA), $n = 10$, $p \geqslant 0.05$; F_1(CBA × B6), $n = 7$, $p \geqslant 0.1$). However, females of the same F_1 types do show the previously described preference for unrelated oestrus female urine over maternal strain female urine (figure 2*b*; Wilcoxon signed-rank test: F_1(B6 × CBA), $n = 11$, $p < 0.01$; F_1(CBA × B6), $n = 10$, $p < 0.05$), and not for unrelated oestrus female urine over paternal strain female urine (Wilcoxon signed-rank test: F_1(B6 × CBA), $n = 11$, $p > 0.1$; F_1(CBA × B6), $n = 10$, $p > 0.1$). Although no preference was shown for male urinary odours of the two parental strains, the F_1 females were able to distinguish these odours in an habituation–dishabituation test (figure 2*c*; paired, one-tailed *t*-test: $n = 10$, $p < 0.001$).

(b) *No parent of origin effects on urinary odours themselves*

The avoidance of maternal urine odour that the reciprocal cross F_1 male and female mice demonstrate could have resulted from one of two mechanisms. First, the odour preference may be due to genes that are subject to parent of origin effects, acting directly on the olfactory system, either at the level of perception or information processing. Alternatively, the preference may be a learnt response, although this cannot be due to prior exposure to either set of parents as the F_1 mice were transferred as embryos to a foster mother of unrelated strain. Nevertheless, the animals may learn their own urinary odours, and use these as a template against which to base odour preference decisions (Mateo & Johnston 2000). In this context, the POE would affect the production of urinary odour.

In order to test this latter possibility we examined the ability of Balb/C females to discriminate between the urinary odours of reciprocal cross F_1 males (B6 × CBA and vice versa) in a habituation–dishabituation paradigm. The mice were unable to distinguish between the two reciprocal cross F_1 odours (figure 3*a*; paired, one-tailed *t*-test: $n = 7$, $p = 0.35$) regardless of order of presentation. However, in a separate test the same mice could readily

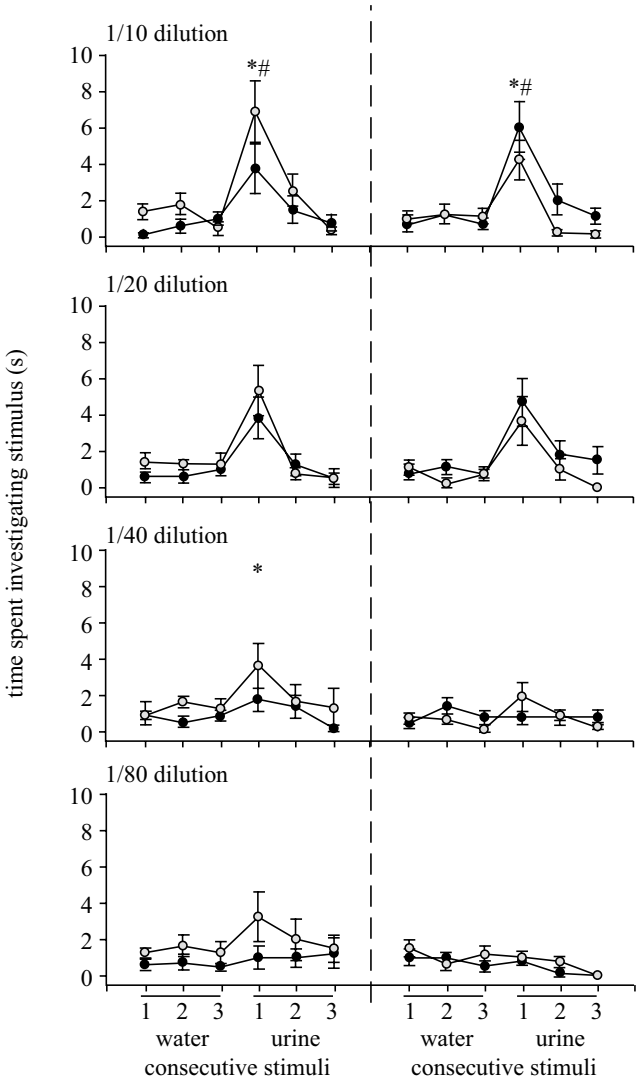

Figure 4. Habituation–dishabituation tests of reciprocal F_1 males with increasing dilutions of B6 oestrus female urine (left block), or B6 male urine (right block). An asterisk indicates a significant ($p < 0.05$) increase in investigation for F_1(B6 × CBA) males, and a hash indicates a significant increase in investigation for F_1(CBA × B6) males. Values shown are means ± s.e.

discriminate samples of the individual genetic parental strains (B6 and CBA) (figure 3*b*; paired, one-tailed *t*-test: $n = 7$, $p < 0.001$).

(c) *F_1 mice have a lower threshold of maternal odour cue detection*

Reciprocal cross F_1 males (B6 × CBA and vice versa, gonadectomized to control for hormonal influences) were tested for their sensitivity to various dilutions of both male and oestrus female B6 urine in an habituation–dishabituation paradigm. The males of both F_1 types were equally sensitive to B6 male urinary odours, neither being able to distinguish a 1/40 dilution of the urine from water (figure 4). However, when tested with dilutions of B6 oestrus female urine, F_1 males whose maternal genotype was B6 were much more sensitive than males of the reciprocal F_1 cross whose maternal genotype was CBA. The former (B6 × CBA) could discriminate a 1/40 dilution of the urine from water (Wilcoxon signed-rank test, one-tailed: $n = 10$, $p < 0.05$), whereas the latter (CBA × B6) could not.

4. DISCUSSION

We have shown through the use of reciprocal cross F_1 mice the existence of POE that influence odour preference of mice. Two independent reciprocal F_1 crosses show the same behavioural pattern, implying that this phenomenon is real, and is not simply a genetic anomaly particular to one F_1 type.

Specifically, male and female reciprocal F_1 mice avoid maternal odour cues, despite having had no prior exposure pre- or postnatally to odours of the maternal genotype. Furthermore, as the urinary odours produced by reciprocal F_1 males themselves cannot be discriminated by females, we suggest that these effects are not due to a self-learning mechanism, but the result of genetic influences on the olfactory system itself, at the level of detection. This is supported by the fact that reciprocal F_1 males have differential detection levels of female urine, in that F_1 males have a lower detection threshold of their maternal strain female urine. As the males were tested two months after castration, it is probable that these data reflect a non-sexual aspect of sensory function.

The use of offspring from reciprocal crosses between inbred strains of mice provides a methodology to examine parent of origin effects that relies on underlying gene polymorphisms and not upon a genetic manipulation. Reciprocal cross strategies have been employed to demonstrate parental effects on a number of phenotypes at the behavioural (McGill & Manning 1976; Banko *et al.* 1997), cellular (Bander *et al.* 1989; McDonald & Jackson 1994), and molecular level (Vogel & Klose 1992). In addition, we also observed POE differences in weight between our reciprocal cross F_1 mice (A. R. Isles, M. J. Baum, D. Ma, A. Szeto, E. B. Keverne and N. D. Allen, unpublished data). The POE influencing olfactory related behaviour reported here occurs in both males and females (Isles *et al.* 2001) and therefore is unlikely to be due to sex-linked genes. Consequently the most parsimonious explanation is that this effect is caused through genomic imprinting, although cytoplasmic inheritance, such as via mitochondria, cannot be ruled out.

The most robust explanation for the evolution of genomic imprinting is the conflict hypothesis (Hurst 1997), although this theory cannot explain all incidences of imprinting (Hurst & McVean 1998). The conflict hypothesis focuses on maternal resource allocation to the developing foetus (Moore & Haig 1991). This theory predicts that paternal genes within the foetus will maximize the contribution of resources from the mother, whereas maternal genes will be expected to reduce the contribution. Many of the known imprinted genes concur with the predictions (Dechiara *et al.* 1990; Lau *et al.* 1994; Leighton *et al.* 1995; Lefebvre *et al.* 1998; Li *et al.* 1999). Although most attention has focused on the developing embryo, other evidence, such as the concentration of androgenetic cells in the hypothalamus (Keverne *et al.* 1996), indicates that manipulation of metabolism and resource allocation may also occur postnatally. In line with this, it has been suggested that imprinting may also influence kin recognition abilities and their subsequent

behavioural outcomes (Hurst 1997). The data presented here provide evidence that this is indeed the case.

Odour preference was used in this research as it provides an experimental paradigm that is relevant to behaviours that are dependent upon olfactory discrimination. Such behaviours include kin recognition, inbreeding avoidance, mate preference and juvenile dispersal from the natal area (Moore & Ali 1984; Barnard & Fitzsimons 1988; Waldman *et al.* 1988), all of which could be influenced by parental conflict (Burt & Trivers 1998). The data imply there is a genetic mechanism that influences postnatal behaviours of mice in response to cues provided by their genetic mothers, in that the F_1 mice are more sensitive to, and avoid maternal odour cues. These POE may promote dispersal of post-weaning young from the natal area. The evolution of dispersal is a complex issue, which is intricately associated with a number of variables including resource availability (food and habitat) and population density, and contributes to inbreeding avoidance (Moore & Ali 1984; Pusey 1987; Clutton-Brock 1989; Gandon 1999). In this context a genetic mechanism that uses imprinting to 'avoid mother' would be in the maternal interest to safeguard resources in the natal area.

Here we have shown a POE which, for both sons and daughters, would facilitate dispersal from the natal area and would further facilitate inbreeding avoidance in sons. That a similar effect is not found for genomic imprinting in daughters avoiding paternal urine can be explained at a number of different levels. At the mechanistic level, the receptors that respond to the two urine types (male or female) are likely to be different. Moreover, inbreeding avoidance for females may not be an issue if males are dispersing, nor may it be the optimal strategy for females.

A recent study has shown that inbreeding in mice under semi-natural conditions results in reduced reproductive success, especially in competitive situations (Meagher *et al.* 2000). However, there was disparity between the sexes with regard to the extent of the fitness decline, with an 81% reduction for males, but only 22% for females. Previous laboratory studies have also shown that male and female mice have a difference in their optimal mate preference, in that males prefer to outbreed, whereas females prefer to inbreed (Barnard & Fitzsimons 1988). In addition, these mate preferences produce differences in litter size, with the largest litter produced by the males optimal pairing choice (coefficient of relatedness equals 0.125; Barnard & Fitzsimons (1989)). Although a large litter size would be of benefit to a male, it may not be the optimal choice for a female, whose total lifetime reproductive output may be reduced. Consequently, in this context, a genetic mechanism that uses genomic imprinting to 'avoid mother' would also be in the paternal interest to ensure optimal outbreeding.

For such a genetic system to operate in outbred populations it may be necessary to postulate that the genes determining odour and those subject to POE that act on the perception and processing of odours would be in linkage disequilibrium. This idea has been suggested previously (Yamazaki *et al.* 1976), and linkage between olfactory receptor genes and major histocompatibility complex (MHC) loci has been demonstrated in both mice (Amadou *et al.* 1999) and humans (Fan *et al.* 1995; Ehlers *et al.* 2000). However, the interaction between olfactory receptors and linked MHC genes or molecules associated with them has not been shown in mammals, although there is a precedent in fungi (O'Shea *et al.* 1998).

We suggest that the POE on preference behaviour we present here may be the result of a complex interaction between the conflict of different parental genomes in terms of resource allocation and inbreeding avoidance. In one case (resource allocation), it is in the maternal interest that offspring avoid maternal odour cues in order to promote dispersal from the natal area. In the other (inbreeding avoidance), it is in the paternal interest to avoid maternal odour cues, thus promoting outbreeding.

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REFERENCES

Allen, N. D., Logan, K., Lally, G., Drage, D. J., Norris, M. L. & Keverne, E. B. 1995 Distribution of parthenogenetic cells in the mouse-brain and their influence on brain-development and behavior. *Proc. Natl Acad. Sci. USA* **92**, 10 782–10 786.

Amadou, C., Kumanovics, A., Jones, E. P., Lambracht-Washington, D., Yoshino, M. & Lindahl, K. F. 1999 The mouse major histocompatibility complex: some assembly required. *Immunol. Rev.* **167**, 211–221.

Bander, S., Watson, S. & Shire, J. 1989 Paternal inheritance of egg traits in mice: a case of genomic imprinting. *Genet. Res.* **54**, 213–219.

Banko, M. L., Allen, K. M., Dolina, S., Neumann, P. E. & Seyfried, T. N. 1997 Genomic imprinting and audiogenic seizures in mice. *Behav. Genet.* **27**, 465–475.

Barnard, C. J. & Fitzsimons, J. 1988 Kin recognition and mate choice in mice: the effects of kinship, familiarity and social interference on intersexual interaction. *Anim. Behav.* **36**, 1078–1090.

Barnard, C. J. & Fitzsimons, J. 1989 Kin recognition and mate choice in mice: fitness consequences of mating with kin. *Anim. Behav.* **38**, 35–40.

Brambilla, R. (and 14 others) 1997 A role for the Ras signalling pathway in synaptic transmission and long-term memory. *Nature* **390**, 281–286.

Burt, A. & Trivers, R. 1998 Genetic conflicts in genomic imprinting. *Proc. R. Soc. Lond.* B **265**, 2393–2397.

Cattanach, B. M. & Beechey, C. V. 1990 Autosomal and X-chromosome imprinting. *Dev.* **5**(Suppl.), 63–72.

Chess, A., Simon, I., Cedar, H. & Axel, R. 1994 Allelic inactivation regulates olfactory receptor gene-expression. *Cell* **78**, 823–834.

Clutton-Brock, T. H. 1989 Female transfer and inbreeding avoidance in social animals. *Nature* **337**, 70–72.

Dechiara, T. M., Efstratiadis, A. & Robertson, E. J. 1990 A growth-deficiency phenotype in heterozygous mice carry an insulin-like growth factor-II gene disrupted by targeting. *Nature* **345**, 78–80.

Ehlers, A., Beck, S., Forbes, S. A., Trowsdale, J., Volz, A., Younger, R. & Ziegler, A. 2000 MHC-Linked olfactory receptor loci exhibit polymorphism and contribute to extended HLA/OR-haplotypes. *Genome Res.* **10**, 1968–1978.

Fan, W., Liu, Y. C., Parimoo, S. & Weissman, S. M. 1995 Olfactory receptor-like genes are located in the human major histocompatibility complex. *Genomics* **27**, 119–123.

Gandon, S. 1999 Kin competition, the cost of inbreeding and the evolution of dispersal. *J. Theor. Biol.* **200**, 345–364.

Gregg, B. & Thiessen, D. D. 1981 A simple method of olfactory discrimination of urines for the Mongolian gerbil, *Meriones unguiculatus*. *Physiol. Behav.* **26**, 1133–1136.

Hurst, J. L. 1990*a* Urine marking in populations of wild house mice *Mus domesticus* Rutty. 1. Communication between males. *Anim. Behav.* **40**, 209–222.

Hurst, J. L. 1990*b* Urine marking in populations of wild house mice *Mus domesticus* Rutty. 2. Communication between females. *Anim. Behav.* **40**, 223–232.

Hurst, J. L. 1990*c* Urine marking in populations of wild house mice *Mus domesticus* Rutty. 3. Communication between the sexes. *Anim. Behav.* **40**, 233–243.

Hurst, L. 1997 Evolutionary theories of genomic imprinting. In *Genomic imprinting*, vol. 18 (ed. M. Surani & W. Reik), pp. 211–237. Oxford University Press.

Hurst, L. D. & McVean, G. T. 1998 Do we understand the evolution of genomic imprinting? *Curr. Opin. Genet. Dev.* **8**, 701–708.

Isles, A. R. & Wilkinson, L. S. 2000 Imprinted genes, cognition and behaviour. *Trends Cogn. Sci.* **4**, 309–318.

Isles, A. R., Baum, M. J., Ma, D., Keverne, E. B. & Allen, N. D. 2001 Urinary odour preferences in mice. *Nature* **409**, 783–784.

Keverne, E. B. 1997 Genomic imprinting in the brain. *Curr. Opin. Neurobiol.* **7**, 463–468.

Keverne, E. B., Fundele, R., Narasimha, M., Barton, S. C. & Surani, M. A. 1996 Genomic imprinting and the differential roles of parental genomes in brain development. *Dev. Brain Res.* **92**, 91–100.

Lau, M. M., Stewart, C. E., Liu, Z., Bhatt, H., Rotwein, P. & Stewart, C. L. 1994 Loss of the imprinted IGF2/cation-independent mannose 6-phosphate receptor results in fetal overgrowth and perinatal lethality. *Genes Dev.* **8**, 2953–2963.

Lefebvre, L., Viville, S., Barton, S. C., Ishino, F., Keverne, E. B. & Surani, M. A. 1998 Abnormal maternal behaviour and growth retardation associated with loss of the imprinted gene *Mest*. *Nature Genet.* **20**, 163–169.

Leighton, P. A., Ingram, R. S., Eggenschwiler, J., Efstratiadis, A. & Tilghman, S. M. 1995 Disruption of imprinting caused by deletion of the H19 gene region in mice. *Nature* **375**, 34–39.

Li, L. L., Keverne, E. B., Aparicio, S. A., Ishino, F., Barton, S. C. & Surani, M. A. 1999 Regulation of maternal behavior and offspring growth by paternally expressed *Peg3*. *Science* **284**, 330–333.

McDonald, T. & Jackson, C. 1994 Mode of inheritance of the higher degree of megakaryocyte polyploidization in C3H mice. I. Evidence for a role of genomic imprinting in megakaryocyte polyploidy determination. *Blood* **83**, 1493–1498.

McGill, T. & Manning, A. 1976 Genotype and retention of the ejaculatory reflex in castrated male mice. *Anim. Behav.* **24**, 507–518.

Mateo, J. M. & Johnston, R. E. 2000 Kin recognition and the 'armpit effect': evidence of self-referent phenotype matching. *Proc. R. Soc. Lond.* B **267**, 695–700.

Meagher, S., Penn, D. J. & Potts, W. K. 2000 Male–male competition magnifies inbreeding depression in wild house mice. *Proc. Natl Acad. Sci. USA* **97**, 3324–3329.

Moore, J. & Ali, R. 1984 Are dispersal and inbreeding avoidance related? *Anim. Behav.* **32**, 94–112.

Moore, T. & Haig, D. 1991 Genomic imprinting in mammalian development: a parental tug-of-war. *Trends Genet.* **7**, 45–49.

Mugford, R. A. & Nowell, N. W. 1970 Pheromones and their effect on aggression in mice. *Nature* **226**, 967–968.

O'Shea, S. F., Chaure, P. T., Halsall, J. R., Olesnicky, N. S., Leibbrandt, A., Connerton, I. F. & Casselton, L. A. 1998 A large pheromone and receptor gene complex determines multiple B mating type specificities in *Coprinus cinereus*. *Genetics* **148**, 1081–1090.

Pusey, A. E. 1987 Sex-biased dispersal and inbreeding avoidance in birds and mammals. *Trends Ecol. Evol.* **2**, 295–299.

Sundberg, H., Doving, K., Novikov, S. & Ursin, H. 1982 A method for studying responses and habituation to odors in rats. *Behav. Neural Biol.* **34**, 113–119.

Vogel, T. & Klose, J. 1992 Two-dimensional electrophoretic protein patterns of reciprocal hybrids of the mouse strains DBA and C57BL. *Biochem. Genet.* **30**, 649–662.

Waldman, B., Frumhoff, P. C. & Sherman, P. W. 1988 Problems of kin recognition. *Trends Ecol. Evol.* **3**, 8–13.

Yamazaki, K., Boyse, E. A., Mike, V., Thaler, H. T., Mathieson, B. J., Abbott, J., Boyse, J., Zayas, Z. A. & Thomas, L. 1976 Control of mating preferences in mice by genes in the major histocompatibility complex. *J. Exp. Med.* **144**, 1324–1335.