

RESEARCH COMMENTARY

Genetically engineered monogamy in voles lends credence to the *modus operandi* of behavioural ecology

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Behavioural ecologists investigate the evolutionary forces that select for one behavioural pattern over another (Krebs and Davies 1991, 1993.) Why do lions hunt in prides while the tiger stalks its prey alone? Why are honey bee workers so industrious while the drones are so lazy? Why do koels lay their eggs in the nests of crows while the latter go through the trouble of building nests and caring for chicks, their own as well those of the koel? Why do Siberian cranes fly some 6400 kilometres from their breeding grounds in Siberia to over winter in Bharatpur in Rajasthan, India, only to return to Siberia in summer? Why are males in many species of birds monogamous, pairing for life and providing paternal care to the chicks, while the males of many mammals are polygynous, mating with many females and contributing little more than a sperm-full of genes to their offspring?

Behavioural ecologists estimate, and wherever possible calculate, the relative costs and benefits of alternative behavioural strategies under the given ecological circumstances and attempt to predict the winning strategy. What are the relative advantages of cooperative hunting versus stalking in stealth, for the lion that lives in open savannas and the tiger that lives in dense jungle? What are the inclusive fitness benefits to the worker bee who rears sisters related to her by 0.75 instead of daughters related to her by 0.5, as compared to similar benefits for drones, of rearing sisters related to them by 0.5 instead of daughters related to them by 1.0? What are the benefits to the koel of saving the cost of nest building and brood care and to the crow of attempting to selectively destroy the koels' eggs without destroying her own? What are the costs for the Siberian crane of attempting to survive, let alone breed, in the severe winters of Siberia compared to the cost of risky and energy-consuming flights to Bharatpur and back? What are the costs to males of denying paternal care to their offspring when the mothers are up to the task by themselves and what are the costs to the females of abandoning their offspring?

Difficult as it is to do, these costs and benefits are often

measurable. The more risky gambit of the behavioural ecologists appears to be the assumption that alternative behavioural strategies are readily available or easily created by mutation so that natural selection can choose from among them (Grafen 1991). The source of uncertainty is our profound ignorance of the proximate physiological, genetic or epigenetic, neurobiological and developmental mechanisms that orchestrate these behaviours. This ignorance has to do, in part but only in part, with the formidable difficulty of unraveling the proximate mechanisms of complex behaviour patterns. The remaining part has undoubtedly to be attributed to the behavioural ecologists' obsession with ultimate factors and habitual neglect of proximate causation (Gadagkar 1997).

But of course there are exceptions, and a particularly spectacular exception concerns our present understanding of the proximate factors involved in the presence and absence of affiliation, pair-bonding and paternal behaviour in voles. Voles are small mouse like rodents that can sometimes be serious pests. Of particular interest for this tale are two North American species, the prairie vole, *Microtus ochrogaster*, and the meadow vole, *Microtus pennsylvanicus*. These two species have contrasting mating systems. The prairie vole is very social, forms lasting pair bonds, males prefer the company of their mates and are aggressive to other individuals, and both parents provide extended parental care to offspring. In contrast, the meadow vole is rather asocial, mating is polygynous or promiscuous, mated pairs do not pair-bond and males do not provide parental care to offspring. There are also two other similarly contrasting species, the monogamous pine vole (*Microtus pinetorum*) and the promiscuous montane vole (*Microtus montanus*) which have been substituted for prairie voles and meadow voles, respectively, in some of the studies. Despite such contrasting social behaviour, prairie/pine voles and meadow/montane voles share more than 99% genome homology and look very similar. The obvious advantage of these vole species as model systems to investigate the evolution of social behaviour in general, and mat-

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ing systems in particular, have long been recognized. Fortunately these contrasting behavioural patterns are also seen in the laboratory, so that simple behavioural assays involving preference for mates versus strangers, affiliative behaviour and time spent huddling with mated partners, have been developed and used to quantitatively assess behaviour patterns under experimental conditions.

A great deal is now known about the neuroendocrine modulation of social and sexual behaviour of vertebrates in general and mammals in particular. In mammals, two neuropeptides, oxytocin and vasopressin, secreted by the hypothalamus, are being increasingly implicated in a variety of socio-sexual behaviours. Oxytocin and vasopressin are peptides with nine amino acids each and differ from each other only in two amino acids. Oxytocin is associated with female behaviours while vasopressin is associated with those of the male. The monogamous prairie vole and the polygynous meadow/montane voles have strikingly different spatial patterns of expression of oxytocin and vasopressin receptors in their brains. Not surprisingly, several investigators including Thomas R. Insel, Larry J. Young, Zuoxin Wang and their colleagues in Emory University in Georgia (USA) have pursued the bold hypothesis of a causal relationship between the spatial patterns of action of oxytocin and vasopressin and socio-sexual behaviour. The greatest interest is in the differences between the monogamous, pair-bonding, paternal prairie/pine vole males and the non-monogamous, non pair-bonding, non-paternal meadow/montane vole males, and most attention has been focused on a class of vasopressin receptors called V1a (Young *et al.* 1998). V1a receptors are present in higher amounts in the ventral forebrain in males of the monogamous species as compared to the promiscuous species (Insel *et al.* 1994) and this has been shown to be due to differences in gene expression (Young *et al.* 1997). This difference has been traced also to differences in gene sequences; the coding region of the gene for V1a receptor is highly (99%) conserved in monogamous and promiscuous species. However, in the monogamous prairie vole but not in the promiscuous meadow vole, there is 428 bp, unstable, microsatellite DNA element in the 5' flanking, promoter region of the receptor gene (Hammock and Young 2002). Nevertheless all this evidence remains merely correlational and therefore not conclusive.

Several lines of direct experimental evidence have also been brought to bear on this question. Intracerebroventricular injection of an antagonist of the V1a receptor into male prairie voles abolished their aggression towards strangers and their preference for their mates, clearly showing the role of vasopressin and its V1a receptor in bringing about this species-specific behaviour (Winslow *et al.* 1993). More convincing is the experimental result that transgenic mice that have received the prairie vole receptor gene acquire a neuroanatomical pattern of receptor binding reminiscent of the prairie vole itself. Moreover, the

transgenic mice which are otherwise hardly monogamous, now affiliate significantly more with their mated partners (Young *et al.* 1999). More recently, a novel adeno-associated viral vector has been employed to deliver the V1a receptor gene to male mice, male prairie voles as well as to males of the promiscuous meadow voles. The mice so treated become more social as compared to controls (Landgraf *et al.* 2003) and the male prairie voles become more affiliative and show stronger preference for their mated partners (Pitkow *et al.* 2001). The clinching evidence however comes from the normally promiscuous male meadow voles receiving the V1a receptor gene from the monogamous prairie voles, who now show significantly higher partner preference, so reminiscent of the prairie vole males (Lim *et al.* 2004).

These results, especially the last one which shows that the normally promiscuous meadow voles can be made to show greater partner preference, like the prairie voles from whom they have received a single gene, are undoubtedly of great significance. Not only do these results unravel the genetic, physiological and neurological pathway for the causation of as complex a behaviour as monogamy but they also provide evidence that a single gene, albeit in the presence of an appropriate genetic and neural background, can profoundly alter social behaviour. Quite predictably, the popular press has gone overboard in reporting these findings. There has been much discussion about the possibility of using transgenic technology to make men more monogamous and more interested in their wives and children! One newspaper even referred to the behaviour of a past President of the United States of America. Only time will tell whether these mechanisms, let alone these technological breakthroughs, will apply to the human species. But the more significant import of these results is the credence they lend to the *modus operandi* of behavioural ecology which should lead to more rapid progress in our understanding of the evolution of other more complex social behaviours in different animal taxa. In other words, these results vindicate the behavioural ecologists' gambit that assumed the availability of alternative behavioural phenotypes for the action of natural selection.

The case of the voles is worth dwelling upon a little bit more. Behavioural ecologists have argued that monogamy would evolve if the cost of indulging in paternal behaviour is more than compensated by the benefit in improved offspring survival. The difficult question was whether the mechanisms that produce monogamy and polygamy permit mutating from one to the other without creating a lethal monster. Now we can conclude that this may not be such a serious problem after all. Indeed, monogamy in the prairie voles and promiscuousness in the meadow vole appear to be quite plastic and flexible even within each species. Not all prairie vole males are equally monogamous: those captured from a resource-abundant habitat in Illinois fit the description of monogamy better than those captured from a

more arid habitat in Kansas, in the USA. Crossing the two populations has shown that hybrid offspring males behaved like their fathers, especially if the father was present up until the birth of the offspring (Roberts *et al.* 1998). Similarly, not all meadow vole males are equally promiscuous. When forced to cohabit with a single female for periods longer than they would ever do in nature, meadow voles also developed significant preferences for their partners and aggression towards strangers (Parker *et al.* 2001).

The study of vole mating systems provides the most convincing proof yet of the mutually beneficial impact of simultaneously asking proximate and ultimate questions in behavioural ecology, if ever a proof was needed.

References

- Gadagkar R. 1997 *Survival strategies: cooperation and conflict in animal societies*. Harvard University Press, Cambridge, and Universities Press, Hyderabad.
- Grafen A. 1991 Modelling in behavioural ecology. In *Behavioural ecology: an evolutionary approach* (ed. J. R. Krebs and N. B. Davies), pp. 5–31. Blackwell Scientific, Oxford.
- Hammock E. A. D. and Young L. J. 2002 Variation in the vasopressin V1a receptor promoter and expression: implications for inter- and intraspecific variation in social behaviour. *Eur J. Neurosci.* **16**, 399–402.
- Insel T. R., Wang Z. X. and Ferris C. F. 1994 Patterns of brain vasopressin receptor distribution associated with social organization in microtine rodents. *J. Neurosci.* **14**, 5381–5392.
- Krebs J. R. and Davies N. B. (eds.) 1991 *Behavioural ecology: an evolutionary approach*. Blackwell Scientific, Oxford.
- Krebs J. R. and Davies N. B. (ed.) 1993 *An introduction to behavioural ecology*. Blackwell Scientific, Oxford.
- Landgraf R., Frank E., Aldag J. M., Neumann I. D., Sharer C. A., Ren X. et al. 2003 Viral vector-mediated gene transfer of the vole V1a vasopressin receptor in the rat septum: improved social discrimination and active social behaviour. *Eur. J. Neurosci.* **18**, 403–411.
- Lim M. M., Wang Z., Olazábal D. E., Ren X., Terwilliger E. F. and Young L. J. 2004 Enhanced partner preference in a promiscuous species by manipulating the expression of a single gene. *Nature* **429**, 754–757.
- Parker K. J., Phillips K. M. and Lee T. M. 2001 Development of selective partner preferences in captive male and female meadow voles, *Microtus pennsylvanicus*. *Anim. Behav.* **61**, 1217–1226.
- Pitkow L. J., Sharer C. A., Ren X., Insel T. R., Terwilliger E. F. and Young L. J. 2001 Facilitation of affiliation and pair-bond formation by vasopressin receptor gene transfer into the ventral forebrain of a monogamous vole. *J. Neurosci.* **21**, 7392–7396.
- Roberts R. L., Williams J. R., Wang A. K. and Carter C. S. 1998 Cooperative breeding and monogamy in prairie voles: influence of the sire and geographical variation. *Anim. Behav.* **55**, 1131–1140.
- Winslow J. T., Hastings N., Carter C. S., Harbaugh C. R. and Insel T. R. 1993 A role for central vasopressin in pair bonding in monogamous prairie voles. *Nature* **365**, 545–548.
- Young L. J., Nilsen R., Waymire K. G., MacGregor G. R. and Insel T. R. 1999 Increased affiliative response to vasopressin in mice expressing the V1a receptor from a monogamous vole. *Nature* **400**, 766–768.
- Young L. J., Wang Z. and Insel T. R. 1998 Neuroendocrine basis of monogamy. *Trends Neurosci* **21**, 71–75.
- Young L. J., Winslow J. T., Nilsen R. and Insel T. R. 1997 Species Differences in V1a receptor gene expression in monogamous and nonmonogamous voles: behavioral consequences. *Behav. Neurosci.* **111**, 599–605.