Integrative animal behaviour and sociogenomics

Gene E. Robinson

Integrative animal behaviour is on the rise: some behavioural neuroscientists are increasingly asking 'why', while some behavioural ecologists are starting to ask 'how'. For example, the most recent edition of Krebs and Davies' authoritative *Behavioural Ecology: an Evolutionary Approach* contains, for the first time, a section on mechanistic analyses of behaviour within an ecological context. This nascent synthesis can be catalysed by molecular genetic analyses of behaviours that occur in a natural context. Recent findings can provide a foundation for increased integration in the study of social behaviour, and provide the basis for an agenda for research on 'sociogenomics'.

Gene Robinson is at the Dept of Entomology and Neuroscience Program, University of Illinois, Urbana, IL 61801, USA (generobi@uiuc.edu).

Both behavioural ecologists and neuroscientists are interested in genes. Behavioural ecologists study adaptations, which ultimately are the product of natural selection's actions on genes. Neuroscientists study neuroanatomical, neurochemical, neuroendocrinological and neurophysiological mechanisms of behaviour, which are built by proteins encoded by genes. One lesson from developmental biology is that focusing on genes provides a 'common language', which can spark a fusion of mechanistic and evolutionary analyses. For example, homeobox genes have provided important insights into both the mechanisms of development and the evolution of wings in insects[1].

Gene-based syntheses of mechanistic and evolutionary analyses can be conducted for several diverse types of behaviour, but social behaviour is attractive because it has already been studied at great depth in behavioural ecology. Readers of this journal scarcely need to be reminded of the rich theories that underlie much of the contemporary study of social behaviour. One of the outstanding achievements of behavioural ecology is the demonstration that the expression of complex patterns of social behaviour is not necessarily rigid, but can be strongly influenced by the environment. This has important implications for several different areas of behavioural biology, including behavioural evolution and animal cognition. For example, what environmental conditions, 'cognitive architectures'[2] and physiological and/or anatomical constraints lead to behavioural diversity within a particular lineage or population of animals? How does an animal acquire and process information from the environment that results in an adaptive

behavioural response, especially when the environment is richly textured with a variety of social interactions? Various types of social behaviour have been well studied mechanistically, including reproductive behaviour in rodents and fish, bird song learning, aggressive behaviour in birds and marine arthropods, and socially mediated changes in brain structure in bees[3]. These studies highlight the fact that many animals are especially attuned to their social environment and have been endowed by natural selection with mechanisms that allow for flexible responses to changing conditions.

Molecular genetic studies of social behaviour begin with one or both of the following questions. First, is allelic variation correlated with individual variation in the expression of a social behaviour? Results of quantitative genetic studies suggest that such correlations exist for many social behaviours[4], but specific genes have yet to be identified. Second, is gene expression in the brain sensitive to social context and, if so, to what extent do changes in gene expression control socially mediated neural and behavioural plasticity? As work in this area progresses, one possible insight will be that information acquired by the nervous system on social conditions can induce changes in genomic function that in turn adaptively modify the structure and functioning of the nervous system. Identifying genes that vary in structure, expression, or both, can lead to new ideas about the mechanisms governing the expression of a particular social behaviour, if such genes already have known functions in other systems. With the new science of genomics and its burgeoning efforts devoted to elucidating gene function in model organisms, more and more genes

are beginning to be understood in functional terms. Even so, some genes that have not yet been discovered in other contexts may be implicated in the regulation of social behaviour; if so, research can be conducted in 'model systems' (e.g. the fruit fly *Drosophila melanogaster*, the nematode *Caenorhabditis elegans* and the house mouse *Mus musculus*) where elucidation of gene function is more tractable. As discussed in the following two sections, gene identification can also contribute to evolutionary and ecological analyses of behaviour.

Division of labour in the honeybee society

Social insects are exemplars of social behaviour because they must coordinate virtually all of their activities with other individuals for colony fitness. As in many insect societies, there is an age-related division of labour among adult worker honeybees, *Apis mellifera*[3]. Young bees primarily feed and care for larvae and the queen; middle-aged bees maintain the hive and store food; and the oldest bees forage for nectar and pollen and defend the hive. A bee typically begins to forage at about 21 days of age and then acts in this capacity for the remaining one to three weeks of her life. But bees can also accelerate, delay and even reverse their behavioural development in response to changes in colony age-demography. For example, in a colony deficient in foragers, some individuals develop precociously into foragers about two weeks early, when they are as young as seven days of age. The social environment influences plasticity in behavioural development in honeybees: older bees inhibit the behavioural development of younger bees[5–7]. Plasticity in behavioural development is essential to maintaining an appropriate division of labour in the face of constant changes in age demography, food availability, predation pressure and climatic conditions.

Do changes in gene expression in the bee brain underlie socially mediated plasticity in behavioural development? To begin to address this question, emphasis has been placed on understanding underlying sociophysiological mechanisms of behavioural development that can then be used to motivate molecular genetic research. These mechanisms involve the endocrine system, pheromones, structural and chemical changes in the brain and circadian rhythms[8]. Only endocrine and pheromone aspects will be discussed here.

Plasticity in behavioural development in honeybees is influenced by juvenile hormone[3]. The blood titre of juvenile hormone is low in bees that work in the hive and high in foragers, and treatment with

juvenile hormone or analogues induces precocious foraging. Removal of the corpora allata (the glands that produce this hormone) results in delayed foraging, whereas hormone analogue treatment eliminates this delay. Precocious foragers have a precociously high titre of juvenile hormone, over-age nurses have a low titre, and bees that revert from foraging to nursing show a titre drop. An inhibitory factor produced by older worker bees, unidentified at present, is suspected of playing a key role in endocrine-mediated behavioural development[7]. Queen mandibular pheromone has also been shown to be involved, suggesting that the control of honeybee behavioural development involves multiple social cues. Queen mandibular pheromone is already known to have a variety of effects on worker physiology and behaviour; recently, it has been shown to inhibit rates of juvenile hormone biosynthesis[9] and delay the onset of foraging[10].

Identification of compounds that regulate socially mediated changes in behavioural development in honeybees can be used as a starting point for molecular genetic studies of social behaviour. One potentially fertile line of investigation is to identify enzymes and other regulatory proteins involved in the production of juvenile hormone and pheromones[11], and their genes, and study how their expression varies during the regulation of colony division of labour. If colony age-demography is an adaptive trait of insect societies, as is widely assumed[12], it might be possible to use such genes to study the evolution of colony social organization. This endeavour could make good use of the fact that genetic variation exists for rate of behavioural development in honeybees. The ecological significance of this variation within populations and colonies of temperate European races of honeybees is not known. However, tropically derived 'Africanized' honeybees show faster rates of behavioural development than do European bees[3]; this type of analysis needs to be extended to other races and species of *Apis*. Comparative studies of honeybee behavioural development have great potential to integrate molecular genetics, social behaviour and ecology.

Page and colleagues[13,14] have developed another approach to the molecular genetics of honeybee division of labour. They studied differences in task specialization that occur during a particular phase of behavioural development, namely the tendency for some foragers to collect pollen while others collect nectar. These differences have been shown to have a genetic component[15]. Artificial selection, facilitated by the ability to inseminate a queen bee with the sperm of a single drone, resulted in two strains of bees: one more likely to collect pollen and the other more likely to collect nectar, even when co-fostered in a wild-type colony. Using genetic linkage methodology, these strains were used to identify two quantitative trait loci (QTL), *pln1* and *pln2*; variation in both of these chromosomal regions was significantly correlated with variation in the amount of stored pollen. Determining the frequency of high and low marker alleles at *pln1* and *pln2* in populations of bees in different environments might provide insight into how ecological forces shape the genetics of social foraging.

Studies of QTL might also lead to the identification of specific genes for food-collection preferences, as the honeybee linkage map becomes more saturated. Honeybees have a very high rate of recombination and a low level of repetitive DNA, which are helpful characteristics for map-based cloning[13]. Identification of such genes could lead to new insights into the mechanisms and evolution of division of labour, especially if homologous genes from other species have known functions.

Variation in mating behaviour in vertebrate societies

Variation in mating behaviour has been well documented at three levels: differences between closely related species, alternative mating strategies within populations of the same species and plasticity in the mating behaviour of individual animals[16]. Recent studies of voles and cichlid fish have revealed provocative differences in gene expression associated with all three types of variation.

Most species of voles, like most mammals, are not monogamous. The prairie vole (*Microtus ochrogaster*) is a striking exception; there is both long-term pair bonding and high levels of biparental care. Closely related species, such as the montane vole (*M. montanus*), are polygamous. This difference in naturally occurring social behaviour is reflected in laboratory assays of conspecific tolerance; prairie vole females, for example, are much more tolerant of conspecific males than are montane vole females. Laboratory studies have demonstrated a strong role for the neuropeptide hormone oxytocin in the expression of vole affiliative behaviour. This led Insel and Shapiro[17] to explore whether variation in the oxytocin system might be associated with variation in vole mating behaviour. They found a dramatically different pattern of distribution of the oxytocin receptor in the brains of prairie voles and montane voles. In the prairie vole, these receptors

are found in the prelimbic cortex and nucleus accumbens, which are part of the 'reward circuit'; in the montane vole, they are located elsewhere. Because mating causes the release of oxytocin and also facilitates pair bond formation, variation in oxytocin receptor distribution in brain circuits might provide a proximal explanation for variation in mating behaviour in voles. Did the presence of oxytocin receptors in the reward circuit facilitate the evolution of monogamy in this lineage, or did receptor distribution evolve in concert with other aspects of neural organization that are associated with monogamy? Molecular genetic analyses of behaviour conducted within a phylogenetic framework could be used to address these issues. Insel and Shapiro[17] also demonstrated an increase in the number of oxytocin receptors in the brains of montane voles just after parturition, which coincides with an increase in maternal care. Studies using vole oxytocin-receptor genes to make transgenic mice are in progress to determine whether an increase in the expression of the oxytocin-receptor gene in the brain does indeed influence the expression of monogamy.

Fish have been used in elegant studies of alternative mating strategies that integrate evolutionary theory with behavioural, endocrine and neurobiological analyses[18]. As in many fish, the African teleost *Haplochromis burtoni* has two forms of males. Dominant males, which are aggressively territorial, brightly coloured, and have high levels of circulating testosterone, enjoy extremely high levels of reproductive success, whereas subordinate males, lacking all these attributes, do not. Francis *et al.*[19] showed that the brains of dominant males have larger cells containing the neuropeptide gonadotropin-releasing-hormone (GNRH) than do subordinate males. GNRH plays a pivotal role in coordinating physiological and behavioural aspects of reproduction in fish and other vertebrates, and the difference in GNRH neuron size is most marked in the hypothalamus, a brain region known to mediate reproductive behaviour. GNRH neurons are directly sensitive to changes in social context; subordinates that become dominant show an increase in neuron size, whereas dominants that become subordinate show a decrease. It is thought that larger cells reflect increased expression of the GNRH gene, which results in more release of this neurohormone (R.D. Fernald, pers. commun.).

These results raise several questions that involve both mechanistic and evolutionary issues, including the following. What are the ecological determinants of

variation in genes that influence behaviour, between individuals, populations and species? How did variation in mating behaviour evolve at the molecular level? For example, spatial differences in gene expression within the brain can be caused by variation in the promoter region of a gene[20]. The evolution of diversity in mating behaviour in voles might thus be related to variation in the oxytocin receptor promoter sequence (T.R. Insel, pers. commun.). This suggests that insights into the evolution of social behaviour could come from studies of gene regulation, rather than from the discovery of novel genes; this has been demonstrated repeatedly in evolutionary studies of animal development[1]. Molecular genetic analyses could also provide a firm basis for analyses of phylogenetic constraints on the evolution of behaviour. In addition, genes that have been shown to vary in expression in response to changing social conditions can be used as probes for pathways of information acquisition and processing that result in adaptive changes in individual neural and behavioural function. Research addressing these questions can begin with genes that encode the oxytocin receptor or GNRH, or analogous genes in species that can be studied more easily in the field. Given the strong conservation of function for genes discovered by developmental biologists[1], it is likely that genes can be used by behavioural ecologists to help discern the evolution of behavioural diversity.

Sociogenomics

I propose to call integrative studies of the molecular genetics of social behaviour 'sociogenomics'. This should involve species in which naturally occurring social behaviour can be studied. Such species are then used for studies that: (1) identify genes that influence social behaviour; (2) determine the influence of these genes on underlying neural and endocrine mechanisms; (3) explore the effects of the environment, particularly the social environment, on gene action; and (4) use these genes to study the evolution of behavioural diversity. Sociogenomics should use molecular genetic studies to strive for a dynamic interplay between mechanistic and evolutionary analyses. I am not advocating the creation of a totally new field; studies reviewed in previous sections[13,14,17,19], and others not cited for the sake of brevity, reflect pioneering efforts initiated some time ago (see also Ref. 21). What I am advocating is implicit in the name sociogenomics; that many genes must be studied to understand a particular behaviour, aided by the revolutionary advances that

are emerging from the Human Genome Project. For example, there are new techniques that allow large numbers of genes to be screened for variation in sequence and expression[22]. The application of these techniques requires at least partial sequencing of a species' genome, which, although now technically straightforward, raises the issue of how to fund new initiatives in sociogenomics.

The challenge of studying the molecular genetics of social behaviour in ecologically relevant contexts is daunting. Perhaps the biggest difficulty is that sociogenomics requires studying the behaviour of a much wider variety of animals than are currently being analysed at the molecular level. At present, it is possible to engineer genetically only a few animal species (the fruit fly, nematode and house mouse), so even if a specific gene were implicated in a behaviour in another species, the techniques of gene addition, deletion and substitution are not available and one cannot yet go beyond establishing a correlation. A truly rigorous molecular biology of social behaviour requires the ability to turn on or off specific genes in specific brain regions at specific points in an animal's life, in a similar manner to that currently employed in studies of learning and memory[20] and chronobiology[23]. Fortunately, techniques are being developed, such as antisense therapy, in the hope of manipulating the human genome. It should be possible to use these techniques to begin testing hypotheses of gene function in animal species that are favourable for studies of social behaviour but lack advanced genetic resources[24].

Despite these limitations, the time is right to develop a molecular biology of social behaviour. Thanks to the Human Genome Project, commercial development of molecular genetics has been extensive and rapid, and many techniques for the study of specific genes are now more accessible to nonspecialists than ever before. In addition, many of these techniques are similar to those used by behavioural ecologists who use molecular markers to study patterns of relatedness among individuals[25]. Social behaviour, like many other forms of behaviour, provides rich material for analyses that integrate neuroscience and evolutionary biology. But social behaviour is exceptionally well suited to demonstrate, in molecular terms, the fundamental principle that the phenotype is a product of the genotype and the environment. It is extremely important to develop and communicate to the lay public such demonstrations, to address the concerns raised by the spectre of 'biological determinism'. Behavioural ecology is the discipline that

has shown that the expression of complex patterns of social behaviour depends heavily on the environment. It is essential that this perspective help guide the development of modern behavioural genetics, both for the sake of the science and its application to societal concerns. For these reasons, behavioural ecologists have an important role to play in the development of sociogenomics.

References
1 Raff, R.A. (1996) *The Shape of Life: Genes, Development, and the Evolution of Animal Form*, University of Chicago Press
2 Real, L.A., ed. (1994) *Behavioral Mechanisms in Evolutionary Ecology*, University of Chicago Press
3 Robinson, G.E., Fahrbach, S.E. and Winston, M.L. (1997) **Insect societies and the molecular biology of social behavior,** *BioEssays* 19, 1099–1108
4 Boake, C.R.B., ed. (1994) *Quantitative Genetic Studies of Behavioral Evolution*, University of Chicago Press
5 Huang, Z-Y. and Robinson, G.E. (1992) **Honeybee colony integration: worker–worker interactions mediate hormonally regulated plasticity in the division of labor,** *Proc. Natl. Acad. Sci. U. S. A.* 89, 11726–11729
6 Huang, Z-Y. and Robinson, G.E. (1996) **Regulation of honey bee division of labor by colony age demography,** *Behav. Ecol. Sociobiol.* 39, 147–158
7 Huang, Z-H., Plettner, E. and Robinson, G.E. (1998) **Effects of social environment and worker mandibular glands on endocrine-mediated behavioral development in honey bees,** *J. Comp. Physiol. A.* 183, 143–152
8 Robinson, G.E. (1998) **From society to genes with the honey bee,** *Am. Sci.* 86, 456–462
9 Kaatz, H-H., Hildebrandt, H. and Engels, W. (1992) **Primer effect of queen pheromone on juvenile hormone biosynthesis in adult worker honey bees,** *J. Comp. Physiol. B* 162, 588–592
10 Pankiw, T. *et al.* (1998) **Effects of queen mandibular pheromone on behavioural ontogeny and juvenile hormone titres in honey bees,** *J. Insect Physiol.* 44, 685–692
11 Plettner, E. *et al.* (1996) **Caste-selective pheromone biosynthesis in honeybees,** *Science* 271, 1851–1853
12 Oster, G.F. and Wilson, E.O. (1978) *Caste and Ecology in the Social Insects*, Harvard Press

13 Hunt, G.J. *et al.* (1995) **Major quantitative trait loci affecting honey bee foraging behavior,** *Genetics* 141, 1537–1545

14 Page, R.E., Jr *et al.* (1995) **Genetic determinants of honey bee foraging behaviour,** *Anim. Behav.* 50, 1617–1625

15 Calderone, N.W. and Page, R.E., Jr (1988) **Genotypic variability in age polyethism and task specialization in the honey bee,** *Apis mellifera* **(Hymenoptera: Apidae),** *Behav. Ecol. Sociobiol.* 22, 17–25

16 Alcock, J. (1998) *Animal Behavior* (6th edn), Sinauer

17 Insel, T.R. and Shapiro, L.E. (1992) **Oxytocin receptor distribution reflects social organization in monogamous and polygamous voles,** *Proc. Natl. Acad. Sci. U. S. A.* 89, 5981–5985

18 Bass, A.H. (1996) **Shaping brain sexuality,** *Am. Sci.* 84, 352–363

19 Francis, R.C., Soma, K. and Fernald, F.D. (1993) **Social regulation of the brain-pituitary-gonadal axis,** *Proc. Natl. Acad. Sci. U. S. A.* 90, 7794–7798

20 Mayford, M. *et al.* (1996) **Control of memory formation through regulated expression of a CaMKII transgene,** *Science* 274, 1678–1683

21 Crozier, R.H. (1987) **Towards a sociogenetics of social insects,** in *Chemistry and Biology of Social Insects* (Proceedings of the 10th International Congress of the International Union for the Study of Social Insects) (Eder, J. and Rembold, H., eds), pp. 325–328, Verlag J. Peperny, Munich, Germany

22 Chee, M. *et al.* (1996) **Accessing genetic information with high-density DNA arrays,** *Science* 274, 610–614

23 Brandes, C. *et al.* (1996) **Novel features of** *Drosophila* **period transcription revealed by real-time luciferase reporting,** *Neuron* 16, 687–692

24 Ogawa, S. and Pfaff, D.W. (1998) **Current status of antisense DNA methods in behavioral studies,** *Chem. Senses* 23, 249–255

25 Hughes, C. (1998) **Integrating molecular techniques with field methods in studies of social behavior: a revolution results,** *Ecology* 79, 383–399

Distinguishing fact from value?

A Primer for Environmental Literacy
by F.B. Golley

Yale University Press, 1998.
£26.00 hbk, £12.50 pbk (xiv + 254 pages)
0 300 07315 1 / 0 300 07049 7

A primer on environmental literacy would surely go well beyond scientific content. Thus, we were pleased to see Frank Golley state in *A Primer for Environmental Literacy* that: 'science is limited by its point of view and its method. It seems odd to claim that environmental science is narrow, when it studies hierarchies that range from the planet to individual organisms. But the environmental sciences often do not include people in their studies, and almost never economics and politics. If they exclude these, how can they be practical?'

We wholeheartedly agree. Unfortunately, in the 254 pages of his book, the index contains only four references to economics, which are essentially brushes on the surface of a significant topic whose paradigms and methods should have been included in the discussion of environmental literacy. Politics does not even merit one entry in the index.

In essence, Golley spends the vast bulk of his pages, in the name of 'environmental literacy', discussing ecosystems across a hierarchy of scales that he rightly says are needed. He also argues that we need a sense of connectedness: 'Ecology is the study of connections', but his first prescription is that we acknowledge 'the connections we make with nature and the other. When students ask me how to do this, my answer is prayer.' This spirituality is telegraphed on page one when Golley rightly calls for a systems approach to environmental literacy, but then adds that there is a limitation to this approach: 'the main criticism is that it is mechanical and treats nature as a machine. This is a serious objection – nature is above all else not a machine! But in science I know of no better way to study and discuss wholes than in systems language.' However, he defines 'our task here is to understand how ecological systems operate' – which sounds pretty mechanistic.

He goes on to say that 'philosophy and religion offer alternative approaches. But in science we face a poverty of tools.' For us, the mechanistic part of environmental science is to discover the interactions of physical, biological and social subsystems of the planet. Just because we have feelings and beliefs does not remove the obligation to study nature and our institutions as a coupled set of mechanisms. But a strictly mechanistic paradigm of the environmental system can do little to help us decide what is the 'right thing to do' when faced with trade-offs among preserving natural habitats, improving material standards of living or discounting future damages in favor of near-term benefits. Our values and feelings towards nature are indeed 'spiritual'. However, they are more about personal and political values than the science of ecology. Economics at its best is about making trade-offs based on the different valuation of alternative actions. The 'value' of nature is clearly a 'spiritual' component of that valuation decision.

Unfortunately, Golley's book does virtually nothing to teach people how economics works (or does not) and how its conventional and recently more radical (i.e. ecological economics) paradigms impact on the environment. For a primer used in a core graduate course in environmental ethics, it is serious that there is little mention of environmental trade-offs and very little of management practices or the politics that drives them. Nor is the role of the mass media addressed, despite the fact that journalistic 'balance' often pits crackpot science or creationists as credible antagonists to large scientific assessments. Environmental literacy requires knowledge of the broad context in which environmentally relevant actions are pursued.

We differ in our definition of the scope of environmental literacy. Golley describes it as developing a 'sense of the spirit of the place'. He does not explain in depth the political, economic and social context that create and define or solve environmental problems. Golley does allow the humanities to enter into environmental studies, although more in the sense of fine arts than politics, but he states that the focus of his book is on 'science, and we can do little more than tip our hat to these other topics'. This was disappointing in view of his earlier 'practicality' statement.

We agree that environmental literacy should start with a whole systems approach, but it should also include the skills necessary to make educated decisions about environmental problems, by being able to integrate information from different disciplines and – most important – to discern credible sources from that of often elliptical advocacy groups. An environmentally literate person is not required to be – indeed, is unlikely to be – an expert in technical details but is expected to have a knowledge base of how science works in order to evaluate the assessments that surround most environmental policy debates. An understanding of media, economics and politics is necessary for a practical application of environmental science to solving environmental problems.

We taught a freshman seminar in environmental literacy at Stanford[1]. The main points of our curriculum were to teach students how to differentiate between issues of fact from those of value, levels of uncertainty and different sources of information, and to ask the three following questions of any environmental policy debate. (1) What can happen? (2) What are the odds? (3) How do you know? Students used role playing to see perspectives of environmentalists,