“Is parasitic manipulation still a worthwhile research topic?” The answer, according to Thomas et al. (2005), is a resounding YES. A major reason for their confidence is the complexity of the topic, requiring significant input from field biologists, evolutionary biologists, experimentalists, neurobiologists and biochemists. Combining the different approaches, techniques, and backgrounds of such disparate scientists is already producing vibrant, exciting research, and promises more in the future. Their review provides some cautionary notes, but is predominately a challenging blueprint for future work. Here, we comment briefly on some of the salient features of that review, and add a few suggestions on “where we could go” in our quest to understand manipulative parasites.

1. Adaptive versus non-adaptive changes

Thomas et al. (2005) start with a well-reasoned discussion of the debate on adaptive versus non-adaptive changes, focusing on the distinction in the literature between adaptations and “by-products” of infection, especially pathology. As they note, “if pathology is linked to transmission, then it is highly likely that natural selection has not been blind to that pathology.” To a field biologist (JH), this makes eminent sense.

It is perhaps regrettable that the authors did not extend that argument to the “cost” of host manipulation. The literature on sexual selection is rife with examples of strong selection for very expensive ornamentation and/or behaviours. Similarly, host manipulation that leads to increased transmission to a suitable host would presumably be selected regardless of the costs of manipulation or of losses to non-host predators (but see Mouritsen and Poulin, 2003). Of course other selective pressures might very well operate to reduce those costs or limit such losses, and their ecological consequences could be different (Lafferty et al., 2000).

2. Mechanisms

In this section, the authors use the narrow definition of “manipulation” (modulation of the neuroendocrine control systems, Holmes and Zohar, 1990), rather than the broad usage (parasite-induced altered behavior in general) of the rest of the review. They highlight the vast progress accomplished in the past few years using vertebrate host systems. However, invertebrates present unique advantages for neuroethological studies, i.e., simpler nervous systems and more stereotyped...
behaviours. The following comments emphasize the role invertebrates models could play in elucidating the neural basis of parasite induced altered behaviours.

2.1. Advantages of invertebrate systems

Invertebrates possess fewer neurons with often larger cell bodies and thicker axons, and, most importantly, identifiable neurons. These cells can be found reliably from one individual to the next in a given species, due to their specific morphological, electrophysiological, and biochemical properties. For example the biogenic amine serotonin has been implicated in a number of parasitic manipulations, and it is interesting to contrast the complexity of the serotonergic network in vertebrates and invertebrates. The diffuse serotonergic modulatory system of mammals, the dorsal raphe nuclei in the brain stem, consists of thousands of neurons with widespread projections. In contrast, fewer than 60 neurons labeling for serotonin can be identified in the brain of isopods (Thompson et al., 1994) and amphipods (Helluy and Thomas, 2003). In vertebrates, most altered behaviours already described involve complex behavioural patterns such as social behaviour (aggression, reproduction, and social rank, review in Klein, 2003) or feeding/predator avoidance (reviews in Holmes and Zohar, 1990; Moore, 2002). In invertebrates, manipulated behaviours consist often of altered reflex arcs, for instance changes in responses to mechanical stimuli leading to altered escape behaviours (reviews in Holmes and Zohar, 1990; Moore, 2002).

2.2. Contribution of invertebrate systems

Researchers in the field of parasitic manipulation have taken advantage of the fact that the neural circuitry of escape behaviours is known in exquisite details in some arthropods to investigate at which level of reflex arcs impairment occurs. Electrophysiological studies show that the paralyzing venom of the wasp Ampulex compressa, unlike that of other solitary wasps, does not interfere with signal transmission at the neuromuscular junction of cockroaches and that sensory pathways are not altered by the wasp’s venom. Instead, the venom of A. compressa acts centrally to modulate the response of specific interneurons and/or motoneurons to various sensory pathways (Fouad et al., 1996). Liberstat and Moore (2002) demonstrate longer latency and higher threshold for escape behaviours in cockroaches infected by Moniliformis moniliformis and conclude that the disruption in the escape circuitry is likely to take place centrally between thoracic interneurons.

Only a handful of other invertebrate systems have been explored in a systematic way, each one yielding essential information (review in Adamo, 2002). In one of them the relationship between the parasitic wasp Cotesia congregata and Manduca sexta has been investigated demonstrating that feeding is suppressed in the insect as octopamine concentrations increase and disrupt neural activity in the frontal ganglion of the brain. The study of schistosome/snail associations reveals that parasites interfere with their host physiology at multiple levels to suppress egg-laying (review in de Jong-Brink et al., 2001). A complex picture emerges in which larval schistosomes secrete a number of neuropeptide-like compounds, alter gene expression in various host tissues, induce changes in the production of a panoply of peptides by the host, and cause the reduction of brain parts associated with underdeveloped reproductive target organs in the mollusk.

Mechanisms recently unraveled in manipulated hosts, mostly in vertebrates, implicate changes in the metabolism of molecules that have an impact on both neural and immune systems such as cytokines and opioids. These compounds are also part of the molecular arsenal of invertebrates’ innate immunity (Ottaviani and Franceschi, 1998; Cooper, 2003). Complementary methods such as molecular biology, proteomics, immunocytochemistry, electrophysiology, biochemistry, and pharmacology deserve to be applied to a greater number of invertebrate systems in order to resolve the cascades of molecular, biochemical, and neural events that start at the host parasite interface and result in the altered motor output of the host.

2.3. Brain worms

Many vertebrates displaying manipulated behaviours harbour intra or extra cellular parasites in their central nervous system. By contrast, in invertebrates, manipulative parasites are generally located in non-neural tissues or in the hemocoel. Therefore, brain worms found in arthropods represent a potentially useful tool for mechanistic studies. They enable us to visualize the interface between a single parasite and an entire brain yielding data on localization that complement
biochemical results. The brain worm Microphallus papillorobustus alters responses to mechanical and photic stimuli in its intermediate host Gammarus insensibilis. This trematode also induces alterations in the architecture of specific neurons as revealed by immunocytochemical methods (Helluy and Thomas, 2003). The giant serotonergic neurons extend projections throughout the central nervous system, and these giant neurons are stunted in infected gammarids. Thus, it is possible that parasites may have an impact on regions of the brain where they are not present by modifying the architecture of neurons, and the topography of their far-reaching projections.

A possible mechanism of action involves nitric oxide (NO), yet another molecule with neural and immune properties. NO which is known to be a major effector molecule of macrophage toxicity against many parasites including helminths (Oswald et al., 1994), is produced by microglia in the invertebrate nervous system (Sonetti et al., 1997), and may also play a general role in the development of discrete neural networks (Scholz et al., 1998). Therefore, NO released by immunocompetent cells at the interface between host and parasite tissues could alter the distribution of terminals, disrupting neural connectivity. Parasites encysted in the brain could also be coated with compounds such as cell-adhesion molecules (e.g. N-cam, cadherins and integrins) or growth factors that attract or redirect neuronal terminals.

Sonetti et al. (1997) conclude an article on the properties of microglia across taxa by stating that invertebrates “may serve as an excellent and economical model for the study of the relations between the nervous, neuroendocrine and immune systems”. The presence of a manipulative parasite in an invertebrate brain could add another dimension to the model and allow the study of neuroinflammation. On a less optimistic note regarding the potential contribution of immune responses and the complex interactions between immunity and the nervous system, the authors speculate that most parasites effect behavioural changes indirectly, through the immune system, as a form of “sickness behaviour”. This is an interesting suggestion that should be followed up, and extended to include the complex interactions between immunity and the endocrine system (Morales-Montor et al., 2004). A major manipulation by parasitoids involves host maturation and moulting, so it is not surprising that they influence host hormone levels directly, either by secretion of active hormones, by inactivating those secreted by the host, or by influencing host secretion of hormones (see review by Beckage and Gelman, 2004). Feminizing parasites, such as sacculinid barnacles (Bishop and Cannon, 1979), nematodes (Vance, 1996) or microsporidia (Rogers-Gray et al., 2004), may have similar direct actions on the host hormonal systems (although the specific mode of action of these parasites is unknown). As pointed out by de Jong-Brink et al. (2001) parasites interfere at multiple levels with their host’s metabolism and direct actions may complement those mediated by the immune system in many cases.

In addition, whenever the host is “induced” to produce substances that influence its behaviour, the question arises as to whether that production is controlled by the parasite or by the host. Hurd (2001) points out that early developmental metacestodes of Hymenolepis diminuta in Tenebrio molitor produce a manipulation factor that directly inhibits vitellogenin synthesis in the beetle fat body; female (but not male) T. molitor also produce a circulating antigonadotrophin that reduces ovarian protein content. Hurd argues that the latter “strongly suggests that the female host is regulating reproduction in response to infection”, and that this has benefits to the host as well as to the parasite.

3. Manipulated hosts within ecosystems

This is another section that resonates strongly with a field biologist. Laboratory experiments are neces-
P. paradoxus may be an example of another role for manipulative parasites, facilitating “host capture”. Members of the genus Polymorphus are almost exclusively parasites of birds; to our knowledge, P. paradoxus is the only species to have substantial populations in mammals. Manipulative parasites that elicit a change in habitat of the host should encounter a different set of potential definitive hosts; one would expect strong selection for characters that would allow the parasite to develop in a host that is regularly encountered. P. paradoxus is the only one of our local species that will excyst at mammalian body temperatures (Holmes, unpublished results). Perhaps this is an example of a trait allowing host capture. Given their strong central foraging behaviour, these mammals might efficiently recycle any parasites developing such a trait.

4. How complex are “parasitically modified organisms”?*

We fully agree that parasitically modified organisms are more complex than indicated thus far in the literature. For example, our observations in the field and in the laboratory indicate that Gammarus lacustris infected with P. paradoxus differ from uninfected gammarids in many respects. They are photophilic, found most frequently in shallower water, are more sensitive to disturbance, and have altered escape behaviour (Bethel and Holmes, 1973). Infected males are less competitive for mates, less likely to pair when isolated with a female, and both sexes overwinter in shallower water (Zohar and Holmes, 1998). Infected females have reduced overwinter survival, and have greatly reduced pairing success in the field or in laboratory tests (Zohar, 1993). Helluy (1988) has shown that there are at least two mechanisms involved. Injected serotonin elicits not only the clinging response (Helluy and Holmes, 1990), but also other aspects of the altered escape behaviour (escape towards the source of light and “skimming” along the surface). A migration of the accessory screening pigment in the eyes of infected gammarids, producing a more “light-adapted” eye, and probably associated with the habitat shift, was not elicited by serotonin. The infected individuals resembled juveniles in their shallower habitats, but there was no evidence for castration of either males or females (Zohar, 1993). We...
suspect that most parasitically modified organisms will show similar, or even greater, complexities.

5. Causes of interspecific variation in manipulative processes

Helluy (Helluy, 1988; see also Helluy and Holmes, 1990 and Zohar, 1993) studied individual variation in the extent of manipulation by *P. paradoxus* in *G. lacustris*, and in the photic responses of uninfected *G. lacustris*. Helluy noted four patterns. First, uninfected amphipods showed a season pattern in degree of photophilia, with maximum numbers of individuals active in the lighted zone during late summer. Second, in both infected and uninfected amphipods the migration of the white pigment in the eye covered only a small fraction of the potential range; that fraction had a similar amplitude, but a different set point, in different individuals. (Unfortunately, the photophilia and pigment migration were investigated in different individuals, so possible connections between these two patterns could not be discerned.) Third, infected animals did not invariably show altered behaviour; instead there was a predictable decrease over time in the number responding. And, finally, there was a high interindividual variability in strength of the elicited behaviours, with considerable consistency within individuals. At least part of the variability among individual infected gammarids was probably due to differences in the host; it is unclear whether or not there was also variability amongst the parasites in their ability to manipulate. It is probable that at least some of these patterns apply to other systems.

6. Concluding remarks

Parasitically-manipulated hosts sometimes do appear to be novel organisms (Curtis, 1990, Brodeur and Boivin, 2004), differing from their uninfected counterparts in many ways. However, it seems likely that the novel behaviour or physiological patterns are either normal patterns elicited at unusual times (Brodeur and Boivin, 2004) or under unusual circumstances, or are elaborations on normal patterns. It also seems likely that the parasites are eliciting those patterns through rather simple mechanisms. We totally agree with Thomas et al. (2005) that searching for those mechanisms will require interdisciplinary approaches, and is likely to produce information of wide interest.

References


