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Behavioral Genetics: Guanylyl Cyclase Dispatch Prompts Worms to Party

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When it comes to foraging, there are two types of worm in the world: those who enjoy a party, and those who prefer to dine alone. Two recent reports describe roles for guanylyl cyclase in the neuromolecular signaling systems that effect this natural behavioral dimorphism.

Understanding the intricacies of the relationships between genes, the environment and behavior is a major challenge for modern biology. The complex interplay between molecular and neuronal networks and the varying effects of environmental contexts complicate our investigations. Work towards this understanding is often impractical in extremely complex and incompletely characterized systems, such as mammals, and so we turn to model organisms, such as the nematode *Caenorhabditis elegans*. *C. elegans* has a small, well-defined nervous system which it uses to perform complex behaviors. One such behavior that has attracted much interest recently involves a natural foraging behavior dimorphism: upon exposure to a nutritive bacterial lawn, some worms will quickly aggregate and form tight foraging groups, while others continue to feed individually [1].

Since the discovery of this natural dimorphism in foraging behavior, researchers have located the gene responsible, *npr-1* [1], and, remarkably, the single amino acid difference that underlies the dimorphism. They have shown that the *npr-1* gene encodes a neuropeptide receptor and is expressed in a small number of cells [1], and identified the receptor's ligands [2]. Furthermore, using mutant and transgenic analyses, together with cellular ablation techniques, they have described several interacting neural circuits and signal transduction cascades that affect the group foraging phenotype [3,4]. Two new papers [5,6] have now extended our understanding of the signaling systems in which the naturally variable NPR-1 protein acts. Both studies have revealed a novel role for soluble guanylyl cyclase in group foraging behaviors.

In a paper published recently in *Current Biology* [5], Mario de Bono and colleagues describe how their search for modifiers of group foraging behavior led to the identification of two genes for soluble guanylyl cyclases. The authors demonstrated that these genes are expressed in very few cells, and more specifically, in three critical neurons that express the naturally variable *npr-1* gene and a heterodimeric cGMP-gated ion channel, TAX-2–TAX-4, also known to affect group foraging behavior [4]. Signaling by these three neurons influences group foraging, and Cheung *et al.* [5] discovered that guanylyl cyclase function is essential for their effect.

The anatomy of these neurons is of interest, as not only are they directly exposed to the body fluid [4] but at least one of them, URX, also has dendritic extensions to the tip of the worm's nose [6]. The functional implications of this neural anatomy are not yet clear, but it does fit tantalizingly well into a theory that this behavior involves the collection and integration of information from several sources in both the external and internal environments. It will be interesting to learn what internal cues in the body fluid, if any, are being monitored, and how the state of the animal — its nutrition, for example — affects group foraging behavior. It has also recently been shown that these neurons may use guanylyl cyclase to monitor external O_2 concentrations [6] (see below).

The anatomical colocalization with NPR-1 and the TAX-2–TAX-4 channels, together with the behavioral phenotypes caused by mutations of these two guanylyl cyclase genes, provides compelling evidence that these cyclases act in a signaling pathway that contributes to the natural variation in behavior. Cheung *et al.* [5] investigated the molecular roles of these cylases in the performance of foraging behaviors, and in so doing they uncovered a novel attribute of guanylyl cyclases in *C. elegans*. The sequences of all seven *C. elegans* guanylyl cyclases bear closer similarity to the β subunits of mammalian guanylyl cyclases than α subunits [7]. As mammalian soluble guanylyl cyclases are classically believed to function primarily as αβ heterodimers [8], it was not clear how the nematode guanylyl cyclases would work with only β-like subunits [9]. Cheung *et al.* [5] report evidence that these two worm guanylyl cyclases form a heterodimeric enzyme with an active site functionally similar to those in mammalian αβ heterodimers. This is intriguing, as it has been observed in other systems that the expression pattern of α-like and β-like guanylyl cyclase subunits may not completely overlap, hinting that other, non-traditional, couplings are possible [8].

Once they had demonstrated a role for the guanylyl cyclases in the *npr-1* cascade, Cheung *et al.* [5] next tested the genetic interaction of guanylyl cyclase with a mutation (*daf-7*) of the TGF-β signaling cascade, which affects group foraging behavior independently of the NPR-1 system [3]. The guanylyl cyclase mutations had little effect on the group foraging behavior of *daf-7* mutants, further demonstrating that all group foraging is not the same, and that even something as simple as *C. elegans* may require fairly complex neuronal interactions to behave appropriately in response to internal and/or external cues.

In the second paper [6], Cori Bargmann's and Michael Marletta's groups describe their investigations of another behavioral response — aerotaxis.

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Coincidentally, they report that the same guanylyl cyclase, in the same neurons identified by de Bono's group [5], also mediates an avoidance response to hyperoxia. Furthermore, they observed that group foraging behaviors are induced upon exposure to high O_2 concentrations and propose that this hyperoxia cue is directly sensed by guanylyl cyclase via a novel activation mechanism whereby guanylyl cyclase is stimulated by binding O_2.

Many animals form groups in response to stressful conditions [10], and indeed, *C. elegans* aggregates on exposure to aversive food stimuli or stimulation of nociceptive neurons [3]. *C. elegans* was found to show a preference for O_2 concentrations of 2–12%, avoiding higher concentrations [6], thus implying that high O_2 concentrations present an aversive cue. It was hypothesized that the worms may invoke group foraging behaviors when exposed to aversive O_2 levels, and that is exactly what was observed.

Gray *et al.* [6] suggest that aversion to high O_2 concentrations may explain one of the behaviors commonly associated with group foraging — 'bordering'. Bordering is the tendency for animals to accumulate at the thick outer edges of a bacterial lawn, and is often associated with group foraging [1]. The edges of the lawn represent regions of vigorous bacterial growth and thus O_2 levels are locally depleted. In a 21% O_2 chamber, the authors measured the local O_2 concentration at the center of the lawn to be 17.1%, while it averaged only 12.8% at the edge. This difference may present enough of a gradient to induce animals averse to high O_2 concentrations to accumulate at the lawn border.

Gray *et al.* [6] note, however, that O_2 aversion, alone, is not likely to explain all aspects of group foraging behavior as, for example, small clusters of animals are unlikely to create strong enough O_2 gradients to attract other individuals. In the future, it may be informative to study the O_2 avoidance behaviors of the naturally occurring group and solitary foraging variants to investigate further the link between group foraging and aerotaxis. This work raises several questions of ecological significance, such as what O_2 concentrations are common in *C. elegans* habitats and what are the proportions of group *versus* solitary feeders in different habitats, with respect to O_2 levels? Additionally, what advantages does forming foraging groups confer?

These two new studies [5,6] detail a novel role for guanylyl cyclase activity in group foraging and bordering behaviors. The induction of this cyclase activity may involve some internally derived signal, and/or environmental cues such as O_2. Thus, another important thread of this signaling web has been identified, yet we are likely only beginning to sense the overall fabric of signals effecting these naturally variable behaviors. Many exciting questions remain to be investigated, in the sensory systems (where most of the studies to date have been focused) as well as in the central processing and final performance of the behaviors. What are the connections between the various neuronally mediated signals, and what are the other molecular components of these systems? What other internal and external cues can induce or suppress group foraging, and in what contexts? How are sensory responses to these cues combined and integrated into appropriate actions from the motor systems? Finally, what is the evolutionary significance of this behavioral variation, and what amount of conservation exists in other species [11]? Given the extraordinary rate of progress to date, we can look forward to learning the answers to many of these questions in the not too distant future.

References

1. de Bono, M. and Bargmann, C.I. (1998). Social feeding in *Caenorhabditis elegans* is induced by neurons that detect aversive stimuli. Cell *94*, 679-689.
2. Rogers, C., Reale, V., Kim, K., Chatwin, H., Li, C., Evans, P., and de Bono, M. (2003). Inhibition of *Caenorhabditis elegans* social feeding by FMRFamide-related peptide activation of NPR-1. Nat. Neurosci. *6*, 1178-1185.
3. de Bono, M., Tobin, D.M., Davis, M.W., Avery, L., and Bargmann, C.I. (2002). Social feeding in *Ceanorhabditi elegans* is induced by neurons that detect aversive stimuli. Nature *419*, 899-903.
4. Coates, J.C. and de Bono, M. (2002). Antagonistic pathways in neurons exposed to body fluid regulate social feeding in *Ceanorhabditi elegans*. Nature *419*, 925-929.
5. Cheung, B.H., Arellano-Carbajal, F., Rybicki, I., and De Bono, M. (2004). Soluble guanylate cyclases act in neurons exposed to the body fluid to promote *C. elegans* aggregation behavior. Curr. Biol. *14*, 1105-1111.
6. Gray, J.M., Karow, D.S., Lu, H., Chang, A.J., Chang, J.S., Ellis, R.E., Marletta, M.A., and Bargmann, C.I. (2004). Oxygen sensation and social feeding mediated by a *C. elegans* guanylate cyclase homologue. Nature *430*, 317-322.
7. Morton, D.B., Hudson, M.L., Waters, E., and O'Shea, M. (1999). Soluble guanylyl cyclases in *Caenorhabditis elegans*: NO is not the answer. Curr. Biol. *9*, R546-R547.
8. Lucas, K.A., Pitari, G.M., Kazerounian, S., Ruiz-Stewart, I., Park, J., Schulz, S., Chepenik, K.P., and Waldman, S.A. (2000). Guanylyl cyclases and signaling by cyclic GMP. Pharmacol. Rev. *52*, 375-413.
9. Wedel, B.J. and Garbers, D.J. (2001). The guanylyl cyclase family at Y2K. Annu. Rev. Physiol. *63*, 215-233.
10. Wilson, E.O. (1975). Sociobiology: The new synthesis. (Cambridge, Mass.:Harvard University Press.)
11. Sokolowski, M.B. (2002). Neurobiology: Social eating for stress. Nature *419*, 893-894.