in the brain the hormone is made: in the nerves of the dorsomedial hypothalamus. The axons of these nerves project to numerous areas of the brain where nerve cells produce GnRH, and staining showed that these axons contacted GnRH-producing cells, suggesting direct effects. They showed that as in birds, GnIH also rapidly inhibits production of luteinizing hormone by the pituitary. Along with the fact that the cells containing GnIH also have receptors for oestrogen-like compounds such as oestradiol, the combined evidence suggests that GnIH is a direct inhibitor of GnRH. Because the brain cells producing and secreting GnIH send their axons into many areas of the brain, GnIH may have other effects on the brain, too. Though we do not know in mammals where the receptors are for GnIH, it looks like the hormone produces multiple effects in the brain. In birds, the hormone not only affects reproductive hormones, but also sexual behaviour in females, such as readiness to copulate. Interestingly, the neurons producing GnIH are in an area of the brain, the dorsomedial nucleus of the hypothalamus, that coordinates and integrates information from external stimuli, the senses and from motivational and emotional inputs.

Kriegsfeld and Bentley along with Takayoshi Ubuka, Kazuhiko Inoue and Kazuyoshi Ukena plan to continue their investigation of the role of GnIH in birds and mammals.

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Fungal farming: A story of four partner evolution

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Agriculture or farming can be loosely defined as the science and art of growing one's own food. It is a science that humans have evolved some 10,000 years ago; however, a group of ants could be the first ones to invent this practice ~50 million years ago. Over time, they have perfected the technique of propagating fungi that serve as food in specialized fungal farms and keeping this farm free from any disease. There is an interesting quadripartite association between the ant, fungus, fungal parasite and antibiotic-producing bacterium, which we are beginning to understand only recently[1].

Attine ants are found mostly in North America and comprise 200 described species in 12 genera that are obligatorily dependent on fungiculture for food. Although attines derived from a single ancestor extant species, cultivate multiple and phylogenetically distant lineages of fungi, most of these fungi belong to the family Lepiotaceae (Agaricales: Basidiomycota). Fungal cultivars, serving as the primary food source for the ants, are carefully manured by the ants with plant substrate, insect frass or seeds. Foundress queens propagate the fungus clonally, by carrying inocula in their mouths during their nuptial flight to establish new colonies and farms. Although at least some 'lower' attines (phylogenetically basal lineages) propagate cultivars that were recently domesticated from free-living populations of Lepiotaceae, the 'higher' attines (a derived monophyletic clade that includes the leaf-cutting ants) are thought to propagate ancient clones, likely several million years old. Among the attines, the two genera of leaf-cutting ants, namely *Acromyrmex* and *Atta* exhibit the most complex fungicultural systems. Colonies of *Atta* spp. can survive for extended periods, often living for 8–10 years or more after initial nest-founding. Foundress queens of these species establish new colonies by digging chambers in the soil, expelling the fungal pellet that they bring from their natal nest, and initiating the cultivation of their own gardens, started using faecal material provided by the queen. Nest-founding occurs within a claustral chamber that remains closed until the first brood is reared, at which point the new workers begin foraging for leaf material outside the initial chamber. Fungus-growing ants are thought to be capable of cultivating their fungus in axenic (single species) 'monocultures', despite continuous exposure to the competitively advantaged microbes already present in the vegetative material that is added to the garden. It also appears that the fungi cultivated by these higher attines are ancient clones that have been evolving strictly within this mutualism for millions of years. On the other hand, in the lower attines there is vertical transmission of the fungus from parent to offspring colony, with occasional horizontal (lateral) transfers of cultivars between ant colonies and perhaps domestication of novel cultivars.

It was initially thought that these gardens are maintained as monocultures despite continuous exposure to the competitively advantaged microbes already present in the vegetative material that is added to the garden. Currie *et al.*[2] conducted the first extensive study of nonmutualistic filamentous fungi associated with fungus-growing ant gardens and found that fungi in the hyphomycete genus *Escovopsis* (anamorph allied with Hypocreales: Ascomycota) were the most common nonmutualistic ones present. *Escovopsis* can have a dramatic impact on the health and survivorship of fungus-growing ant gardens. As previously noted, *Escovopsis* can rapidly overgrow gardens in the absence of ants. More importantly, under some conditions, even in the presence of ants, it can completely devastate colonies by rapidly overgrowing the whole fungus garden.

To help defend their cultivars from the garden parasite, attine ants have a mutualistic association with filamentous bacteria that produce antibiotics, with potent antagonistic properties against *Escovopsis*. These bacteria of the genus *Pseudonocardia*, were found to be associated with all 22 species of fungus-growing ants examined, representing eight genera and the phylogenetic diversity of the Attini[3]. They also found it to be associated with all colonies that were examined closely. Bioassays indicate that this actinomycete apparently does not produce secondary metabolites with any general antifungal properties. However, they produce specialized antibiotics that have potent inhibitory properties against the pathogen *Escovopsis*. This clearly indicates that the actinomycete is a highly evolved mutualist within this ancient association, utilized by fungus-growing ants to suppress *Escovopsis*. This bacterium can be regarded as a specific 'biocontrol agent' against parasitic attack on the fungal garden maintained by ants.

In their latest publication, Currie *et al.*[4] provide evidence that over the million years of evolution, ant species have developed specialized body structures to host and nurture this agent. They thoroughly examined the anatomy and morphology of ant species in the genus *Cyphomyrmex* because of the conspicuous white bloom of bacterium present on the propleural plates. The species were found to have specialized cavities on these plates in which bacteria grow. Besides, there are exocrine glands located on the inner surface of the cuticle, just below the foveae. The gland consists of bicellular units, each formed by a gland cell and duct cell. The duct cells cross the cuticle and open within the foveae where the bacteria are cultured. In addition to foveae occurring on the propleural plates, *C. longiscapus*, *C. muelleri* and *C. costatus* ants also have bacteria-filled foveae covering most of the surface of worker exoskeletons, including the head, thorax, abdomen and legs. These crypts have small openings to the external surface of the ant, with minute microtrichia (hairlike cuticular projections) that appear to shield the opening of the crypt. At the bottom of each fovea is a porous tubercle (integumental protrusion), connected via a duct cell to the corresponding gland cell directly beneath the crypt.

Ant genera closely related to attine ants, *Wasmannia* and *Blepharidatta* do not have filamentous bacteria, fovea or tubercles. In the most phylogenetically basal attine ants (paleoattines), such as the genus *Apterostigma*, the filamentous bacterium occurs on the mesopleura (under the forelegs), where it grows directly on the cuticle over the pores of duct cells connected to the corresponding gland cells. In most species of lower attine ants, mutualistic bacteria occur on the propleural plates, e.g. *Cyphomyrmex costatus*, in which the bacterium grows on tubercles within the foveae. Similarly, bacteria are also concentrated on the propleural plates in the Bhigher attine genus *Trachymyrmex* and the leaf-cutter genus *Acromyrmex*, although in these two genera the bacteria grow on gland cell-associated tubercles directly on the exoskeleton rather than in the foveae.

Presence of these specialized morphological structures, coupled with molecular phylogenetic analysis of fungi, parasite and ants indicate that this quadripartite relationship has evolved over millions of years. The most interesting question is how the antibiotics have remained effective without rampant evolution of resistance in the parasite. If we understand the mechanisms responsible for this, it will definitely help us in solving the issue of development of multidrug-resistant pathogens, where the resistance develops in a matter of just a couple of years.

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