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Ant-microbe metabolic integration supporting the nutrition of agricultural pest leaf-cutters

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Leaf-cutter ants (Hymenoptera: Attini) are important neotropical agricultural pests. This pest status is related to the high population size and geographic density of ants nests. In this work, we propose that high biomass production by leaf-cutters depends on the association with two classes of key microbes. The fist one is the mutualistic fungus *Leucoagaricus gongylophorus* that we show able to degrade plant polysaccharides and to accumulate in the extracellular space simple sugars which are taken up by the ants. Hemicellulose and starch are the major plant polysaccharides supporting mutualistic nutrition. A metabolic pathway is described involving starch degradation by fungal enzymes to generate maltose and maltose degradation, likely by an ant maltase, to generate glucose, which is a major food source for the mutualists. This mutualistic pathway may have evolved in the ant gut by selection of enzymes which are not sensitive to proteolytic inactivation or catabolic repression. We also show that ant nutrition on plant matter does not occur in the absence of fungal carbohydrases, which are thus essential for ants to access carbons from plants. A second class of mutualistic microbes seem involved on leaf-cutter ant nutrition on nitrogen. Nitrogen from plant leaves is present in very low amounts which are not enough to support the generation of billions of individuals living in a leaf-cutter ant colony. Using metagenomic analysis, we show that the guts of leaf-cutters harbor a narrow variety of microbial species closely related to nitrogen-fixing bacteria. These microbes are proposed as mutualists mediating ant feeding on N2. Therefore, metabolic integration is assumed as a central force modulating the evolution of the association between leaf-cutters and microbes. This association has likely enabled the ants to assimilate great amounts of nutrients necessary to reach high population size and to become major agricultural pests.