"Biodiversity of fungal endophytes in rubber trees: Towards understanding their role as plant protection agents"

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Understanding the role of fungi in communities and ecosystems has been hampered by little sampling and characterization of fungal diversity. Fungal endophytes exemplify this situation. Endophytes reside in living plant tissues and form relationships with plants that range from pathogenic to mutualistic. Of the estimated 500,000 plant species in the world, only a few have been studied to determine their endophytic microflora. The proposed project will characterize fungal endophytic species in rubber trees (*Hevea brasiliensis*), to evaluate their potential role as plant protection agents. Some suggest that symbiotic fungal endophytes have coevolved with host plants to protect them from natural enemies. Rubber trees grown in plantations suffer from a variety of plant diseases, and endophytes may play a role in their protection. Because most fungal endophytes are horizontally transmitted, seeds from *H. brasiliensis* that are used in plantations may lack coevolved endophytes. Based on this premise, it is hypothesized that rubber trees in the wild host greater species richness and abundance of fungal endophytes than trees in plantations. If this hypothesis is supported, plantation trees may lack beneficial endophytes that could prevent some diseases and pests found on plantations. Preliminary data obtained for other tropical tree species support this hypothesis. The endophytic biota of rubber is unknown and few studies have sampled rubber trees in their native habitat, the Amazon region. The diversity and species composition of fungal endophytes in rubber trees in their native (i.e. Peruvian Amazon, specially Iquitos and Madre de Dios) and managed settings (i.e. plantations in Peru, Mexico and Cameroon) will be studied through intensive fieldwork; isolation and cultivation of endophytes from different parts of the tree; and identification of fungal species using morphological and molecular approaches.

During 2007 to 2009, under NSF and Amazon Conservation grants, 5 expeditions were conducted. Three sites were located in the southeast Peruvian Amazon (2 wild populations and one plantation), one located in the north east of the Peruvian Amazon (wild population) and one located in the southeast of Cameroon (1 plantation population). Collection methods were put to test under field conditions, resulting to be very successful for the proposed objectives. The data obtained so far has supported the projects hypotheses: populations distributed within the natural range of rubber tree hold a more diverse and abundant fungal endophyte community and this diversity has a distinctive distribution within the tree. In addition, we have found that the fungal endophyte composition differs between plantation populations as well, which support our hypothesis that crops that are cultivated far away from their center of origin lack of symbiotic endophytes. More importantly, we identified species that have potential to be good strains for the antagonistic experiments such as species from the genera: *Trichoderma, Fusarium, Colletotrichum*, within others. *Trichoderma* is of special importance since several studies have demonstrated its properties as a biocontrol agent (i.e. in the control of frost pod cacao disease). We have collected 100 strains of *Trichoderma* in the center of origin of rubber trees, several of which are taxonomically related to those used in biocontrol of cacao diseases.

The project aims to target the most significant diseases of rubber: the "South American leaf blight" (*Microcyclus ulei*), white root rot (*Rigidoporus lignosus*), Black thread (*Phytophtora palmivora*), leaf spot (*Colletotrichum gloesporioides)*, and leaf fall (*Corynespora cassiicola*). Effective biocontrol endophytes may be found in the native habitat of rubber trees, because these endophytes may have coevolved to protect the host plant from disease. Preliminary assays and experiments on the antifungal properties have started in collaboration with other scientists from USDA[1] and CIRAD[2]. Findings from this study will uncover several new endophytic species and will enhance infrastructure by creating a culture collection that can be used to study endophytic systematics and diversity that can be applied to other lines of study, including pharmaceuticals and biopesticides. More importantly, these findings will collaborate with national institutions/projects in the search for alternative renewable resources.

[1] United States of Department of Agriculture
[2] CIRAD is a French agricultural research centre working for international development.