

MYCORRHIZAL FORMATION BY VARIOUS CARNIVOROUS PLANTS

YANA SANTIAGO • DOUGLAS W. DARNOWSKI • Indiana University Southeast • 4201 Grant Line Road
• New Albany, Indiana 47150 • USA • ddarnows@ius.edu

Keywords: physiology: mycorrhizae, roots.

Introduction

Carnivorous plants receive the greatest amount of attention for their modified leaves used for obtaining extra nutrients while they live in low-nutrient soils (Slack 2000). Another way in which plants obtain more nutrients when nutrients in the soil are limiting is the formation of mycorrhizae with fungi (Nemec 1982). The fungi provide a large, highly-absorptive surface area for the plants, able to scavenge rare nutrients efficiently, while the plants provide their fungal partners with carbohydrates. This mycorrhizal habit is found in many forest trees and also in most heaths and in some epiphytes and orchids (Marx 1982; Lambers & Colmer 2005). More and more plants are being discovered to be mycorrhizal (Lambers & Colmer 2005).

With this effect of enhancing the uptake of nutrients from such a relationship, carnivorous plants might be expected to form mycorrhizae, but there are very few reports of such symbioses among carnivores, or even among their fellow wetland plants in general, with one recent report in *Drosera* a rare exception (Fuchs & Haselwandter 2004). Also, Venugopal & Raseshowri (2007) have recently reported the formation of mycorrhizal associations by *Drosera peltata* which actually uses modified underground stems and leaves in place of roots for their connections with mycorrhizal fungi. This contradicts the common belief that there is no association of mycorrhizal fungi with roots of either hygrophyllic or xeromorphophic carnivorous plants (MacDougal 1899, reviewed by Juniper *et al.* 1989).

In our study, 23 species of greenhouse-grown carnivorous plants from various genera were examined to identify the extent of mycorrhizal colonization and the degree of mycorrhizal development within their root systems.

Materials and Methods

Plants that were used during this study were greenhouse grown in Corydon, Indiana, USA. They were propagated either from seed or from greenhouse-grown vegetative fragments. Plants were not inoculated with mycorrhizal spores. Thus, any mycorrhizae found would have come from the potting materials used, or from other environmental sources in the greenhouse.

Twenty three species were examined in total. A representative sample of different parts of the root (or stolons, for rootless species) from each species was obtained and preserved in 95% ethanol for a minimum of 24 hr. Individual root samples of 1-2 cm were placed on a slide, washed with deionized water and stained using a mixture of 80% (by volume) solution A, and 20% (by volume) solution B. Solution A was 0.3% (by mass) aniline blue in 90% ethanol. Solution B was Lactophenol Blue Solution (FLUKA, Fuchs, Switzerland; Marx 1982; Nemec 1982; Ruzin 1999).

Three 1-2 cm sections of root or stolons were selected randomly from samples of each spe-

Table 1: Mycorrhiza-formation scores (VAM=Vesicular Arbuscular Mycorrhizae)		
Taxon	Root system	Score (0 -10)
Nepenthaceae		
<i>Nepenthes sanguinea</i>	Weak/medium	4
Lentibulariaceae		
<i>Pinguicula laeueana</i>	Weak, and large	6
<i>Pinguicula moranensis</i>	Weak	5
<i>Pinguicula planifolia</i>	Weak, and large	5
<i>Utricularia cornuta</i>	None	0
<i>Utricularia inflata</i>	None	6
<i>Utricularia longifolia</i>	None	2
<i>Utricularia nephrophylla</i>	None	0
<i>Utricularia paulineae</i>	None	3
<i>Utricularia striata</i>	None	0
Sarraceniaceae		
<i>Sarracenia purpurea</i>	Weak/medium	2-3
<i>Sarracenia psittacina</i>	Weak/medium	4
<i>Sarracenia rubra</i> subsp. <i>gulfensis</i>	Weak/medium	6
<i>Darlingtonia californica</i>	Weak/medium	7
Droseraceae		
<i>Dionaea muscipula</i>	Weak, but fleshy	4
<i>Drosera adela</i>	Weak, and large	3
<i>Drosera androsaeca</i>	Weak, and large	4
<i>Drosera aliciae</i>	Weak, and large	7
<i>Drosera binata</i>	Weak, and large	6
<i>Drosera patens</i> × <i>occidentalis</i>	Weak, and large	3
<i>Drosera scorpioides</i>	Weak, and large	3
<i>Drosera omissa</i>	Weak, and large	5
<i>Drosera prolifera</i>	Weak	3, mostly VAM
<i>Drosera aliciae</i>	Weak, and large	0 (Control)
<i>Drosera patens</i> × <i>occidentalis</i>	Weak, and large	0 (Control)

cies and used for staining and analysis. Sections were stained for approximately 30 min, with the exceptions of *Drosera binata* and *Nepenthes sanguinea*, both of these plants having a thick, dark root structure. Their root sections were stained for less than 2 min to allow some detail to be seen after staining and then washed thoroughly, like other samples, with deionized water to remove the excess stain. A cross-section cut from the sample was used in the situation when entire root was too thick or too dark to be viewed under the microscope. After being washed with deionized water for approximately 5 min to remove excess stain the root samples were ex-

amed under a compound microscope. Fungal colonization was measured using the following numerical scale devised by Moberly & Darnowski (unpublished):

- 0: No staining or staining of bodies clearly identified as structures other than mycorrhizae.
- 1: 1-3 cells in the field of view with mycorrhizal infection.
- 2: 4-5 cells in the field of view with some mycorrhizal infection or a similar number or bodies or mass of bodies in a smaller number of cells in the field of view.
- 3: 6-8 cells in the field of view with some mycorrhizal infection or a similar number or bodies or mass of bodies in a smaller number of cells in the field of view.
- 4: 9 or more cells in the field of view with some mycorrhizal infection or a similar number or bodies or mass of bodies in a smaller number of cells in the field of view.
- 5-10: Multiple clusters of cells scoring as in 1-4 above, with the total score for all clusters in the field of view being 5-10.

In using the above ranking scheme, more elaborately detailed or larger mycorrhizal clusters in a given cell raised the score by 1. As negative controls, we used roots from *Drosera aliciae* and *Drosera patens* × *occidentalis* (i.e., *Drosera* sp. “Lake Badgerup”) that were grown in aseptic culture media at the lab.

Results

Table 1 illustrates the results of this survey on the presence of mycorrhizal colonization. All but three of the 23 species examined, the exceptions being *Utricularia striata*, *Utricularia cornuta*, and *Utricularia nephrophylla* (all bladderworts and the first and third aquatic plants), formed mycorrhizae when grown in the greenhouse. Wide variation in the shapes and abundance of mycorrhizae was observed in different species. *Drosera aliciae*, *Drosera scorpioides*, *Darlingtonia californica*, and *Sarracenia rubra* subsp. *gulfensis* formed a very large number of fungal colonizations, while other species, such as *Sarracenia purpurea*, *Utricularia longifolia*, and *Utricularia paulineae* formed very few associations.

Discussion

Since the observations of Nitschke (1860) and Burbidge (1897), there has been a general belief that carnivorous plants only have weakly developed roots, e.g., most *Drosera* species, *Dionaea*, *Pinguicula*, *Cephalotus*, *Sarracenia*, and most *Nepenthes*, or no roots at all, e.g., *Utricularia*, *Genlisea*, and *Aldrovanda*. The absence of roots does not necessarily mean, however, that the functions of roots are not needed; in some plants the stem and leaves have replaced their functions (Lambers & Colmer 2005). Therefore, mycorrhizal associations might occur, even in carnivorous plants where true roots have been replaced by modified shoot parts. In particular, this applies to species of *Utricularia*, and as can be seen in Table 1, even some aquatic *Utricularia* showed signs of fungal associations. Given that, it is not surprising that terrestrial and epiphytic species that grow in moist soil and in decomposing organic matter from other plants (Taylor 1989) also often showed signs of mycorrhizal infection.

On the basis of a recent ecophysiological study (Adamec 2005) carnivorous plant roots appear to be physiologically very active per unit biomass, in spite of their relatively weak proportion of total plant biomass. Since the prevailing amount of mineral nutrients in carnivorous plants gained by roots, the activity of which is greatly stimulated by foliar nutrient absorption from prey, the role of roots is crucial also for carnivory (Adamec 2005), making the general presence of mycorrhizae in cultivated carnivorous plants sensible. Further study of wild populations is needed to confirm the presence of mycorrhizae in natural growth conditions.

Acknowledgements: The authors thank the Indiana University Southeast Office of the Dean for Research for Large Grants and Indiana University for an Intercampus Grant and a Research Support Fund grant.

References

- Adamec, L. 2005. Ecophysiological characterization of carnivorous plant roots: oxygen fluxes, respiration, and water exudation. *Biol. Plant* 49: 247-255.
- Burbidge, F.W. 1897. Note on *Nepenthes*. *J. R. Hort. Soc.* 21: 256-262.
- Fuchs, B., and Haselwandter, K. 2004. Red list plants: colonization by arbuscular mycorrhizal fungi and dark septate endophytes. *Mycorrhiza* 14: 277-281.
- Juniper, B.R., Robins, R.J., and Joel, D.M. 1989. *The Carnivorous Plants*. Academic Press, London.
- Lambers, H., and Colmer, T.D. 2005. *Root Physiology: From gene to function*. Springer, London. p127-140.
- MacDougal, D.T. 1899. Symbiotic saprophytism. *Ann. Bot. London* 13: 1-46.
- Marx, D.H. 1982. Mycorrhizae in interactions with other microorganisms, ectomycorrhizae. In: Schenck, N.C. [ed.], *Methods and Principles of Mycorrhizal Research*, 225-228. APS Press, St. Paul, Minnesota.
- Nemec, S. 1982. Morphology and histology of vesicular-arbuscular mycorrhizae, histology and histochemistry. In: Schenck, N.C. [ed.], *Methods and Principles of Mycorrhizal Research*, 23-28. APS Press, St. Paul, Minnesota.
- Nitschke, T. 1860. Wachstumsverhältnisse des rundblattigen Sonnenthaues. *Bot. Zeit.* 18: 57-69.
- Ruzin, S.E. 1999. *Plant Microtechnique and Microscopy*. Oxford University Press, New York, NY.
- Slack, A. 2000. *Carnivorous Plants*. Marston House, Yeovil, UK.
- Taylor, P. 1989. *The genus Utricularia: a taxonomic monograph*. Royal Botanic Gardens, Kew, London. 724 pp.
- Venugopal, N., and Raseshowri, D.K. 2007. An interesting observation on the mycorrhizal symbiosis in the insectivorous plant, *Drosera peltata* Sm., in Meghalaya, North-East India. *Carniv. Pl. Newslett.* 36: 9-13.

Carnivorous Plant Nursery

*Promoting environmental stewardship through
engaging educational materials and low-cost, high-quality carnivorous plants.*



- Carnivorous Plants
- Bog Orchids, Bog Plants
- Live Wingless Fruitflies
- Activity Book for Carnivorous Plants
- Bugs in the Bog Board Game
- Free Monthly Drawing
- Online Shopping and Mail Order

www.carnivorousplantnursery.com

301-519-7505 | info@carnivorousplantnursery.com | 16128 Deer Lake Road, Derwood, MD 20855