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ROOT SIZE, ROOT HAIRS AND MYCORRHIZAL INFECTION: A RE-EXAMINATION OF BAYLIS'S HYPOTHESIS WITH TROPICAL TREES

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Summary

A series of 89 Brazilian forest species, for which root characteristics and VA mycorrhizal condition were known, was used to test an aspect of Baylis's magnolioid root hypothesis. A non-parametric statistical test showed a significant association between magnolioid root characteristics and VA infection.

Introduction

Baylis (1975) suggested that primitive angiosperm roots, typified by those of the order Mangoliales, are especially dependent on vesicular–arbuscular mycorrhizal fungi for mineral uptake. He described the magnolioid root, which need not occur only in the Mangoliales, as being coarsely branched and having ultimate rootlets generally more than 0·5 mm in diameter. These roots generally lack root hairs, and many of these plant species respond to mycorrhizal infection even in relatively fertile soil.

In contrast, Baylis noted, graminoid roots, which may have branches less than 0·1 mm in diameter and frequently have a dense cover of long root hairs. Plant species having such roots generally respond to mycorrhizal infection only in the most phosphorus-deficient soil. Most species, however, are intermediate types, with the size of ultimate rootlets and the cover of root hairs between these extremes.

Baylis arranged 20 plant species, most having intermediate root types, in a series reflecting their dependence on mycorrhizal infection. The ordering showed a slight tendency toward decreasing root diameter and a strong tendency toward a continuous cover of long root hairs as mycotrophy decreased.

Baylis noted that tropical species were largely absent from his investigation of root types and mycotrophy. An examination of vesicular–arbuscular mycorrhizae in an Amazonian rain forest has afforded an opportunity partly to reconstruct Baylis's study. Data on response to infection and phosphorus fertilization are lacking, since all collections presented here are from mature forest trees. However, data on root diameter and frequency and length of root hairs suggest a slightly different way of arranging the species: from magnolioid to graminoid, with those having root hairs at the end of the series. A relative measure of mycorrhizal status in the field is then used to test the specific hypothesis that magnolioid roots are more likely than graminoid roots to support mycorrhizal infection.

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Materials and Methods

Collection of specimens

Most species were collected from a 1 ha plot of primary lowland rain forest on a very heavy oxisol near Manaus, Brazil. The site was surveyed by Prance *et al.* (1976), and all trees more than 15 cm in diameter were identified and labelled. A few species, including several herbs and shrubs in addition to trees, were collected on an extremely nutrient-poor spodosol (tropical podzol), also near Manaus. They were identified by A. Anderson of the Brazilian National Institute for Amazonian Research. In every case a root specimen was collected by digging outward from the base of an identified plant until its roots produced non-woody, ultimate-order rootlets. Usually about 50 cm^3 (loosely arranged) of these roots were returned to the laboratory for preparation.

Determination of mycorrhizal condition

Specimens of the roots were cleared and stained by the method of Phillips and Hayman (1970) and examined with a dissecting microscope. Sub-samples thought likely to be infected were mounted in glycerin on slides and examined under a compound microscope. The usual basis for judging infection was the presence of internal hyphae. Vesicles were not common and arbuscules were very rarely evident in field-collected native plants. Root specimens were occasionally so opaque that clearing with these techniques was unsatisfactory, and judgement had to be based on external hyphae of the characteristic endogonaceous type.

Plants rated 'non-mycorrhizal' were never observed to be infected. Those rated 'lightly' infected had occasional, isolated infection points; those 'moderately' infected had many parts infected; and those 'heavily' infected had most of the roots infested with hyphae or other VA structures. A more quantitative method was not sought, because of the large number of samples to be examined and because each sample represented only a small, non-random sample of each tree's root system; greater precision would be of little use without a measure of variability. The broad subjective categories were thought more likely to be consistent between replicates, a contention largely borne out for repeatedly sampled species.

Unless they were very rare, each non-mycorrhizal species was collected at least twice. The most common species were collected several times, usually from different trees, regardless of mycorrhizal condition.

Samples of all species, infected or not, were mounted on slides in glycerin for determination of root characteristics. In each case an example of the finest roots in the sample was mounted.

Measurements of roots and root hairs

The presence of root hairs on any part of the sample was recorded during examination with a dissecting microscope.

Root diameter was measured on the finest rootlets present in the sample, and the average of these recorded for the species. When, as in a number of cases, the ultimate-order rootlets were swollen, the swollen branches were measured, even though they were slightly larger than the penultimate-order rootlets. Because the slides had been prepared for determining mycorrhizal condition, the samples were somewhat flattened. A standard diameter correction factor of 0·7 was calculated from samples for which both compressed and non-compressed roots were available. Although the uniform

application of this factor to somewhat variably flattened specimens probably introduced a small error in the relative widths of the various species, it certainly did not substantially alter their positions in the sequence and is a better estimate of the original diameter than the uncorrected measurements.

If no root hairs were included on the permanent mounts, length of the root hairs on a few species was estimated from notes taken during initial examination.

Although other types of mycorrhizae (several ectomycorrhizal and orchidaceous and one ericaceous species) were examined, these were excluded from the analyses because they may not relate in the same way to root morphology.

Analysis of data

The species were arranged in order of decreasing root diameter, from magnolioid to graminoid-like with hairless roots, followed by species having root hairs, ordered first by frequency (rare, inconstant or constant), then, within frequency, by length of root hair. Each species was assigned a rank (graminoid-like roots receiving the lowest ranks), the ranks were assigned to categories of mycorrhizal infection, and Kruskal–Wallis one-way analysis of variance by ranks (Siegel, 1956) was applied to determine whether the ranks were distributed equitably among the mycorrhizal categories.

Taxonomy was based on Cronquist (1968) for flowering plants and on Lawrence (1951) for ferns.

Results

Magnolioid roots

The six members of the Magnoliales examined (*Aniba duckei*, *Licaria aurea* L. sp. and *Nectandra rubra* of the Lauraceae, *Unonopsis stipitata* of the Annonaceae, and *Siparuna* sp. of the Monimiaceae) had root diameters between 0·165 and 0·853 mm, with a mean of 0·429 mm (Table 1). This is somewhat smaller than Baylis's range, 0·35–1·5 mm, and the difference may be (1) because of the population of small root sizes in the tropical Magnoliales, or (2) it may be related to the correction factor used in this study (see Methods), or (3) it may be sampling error. In any case, these results may be taken to agree in a general way with Baylis's. Of the 24 orders represented in this study, five had larger mean root diameters than the Magnoliales. It should be noted that all are poorly represented and one is not an order of flowering plants. None among the Magnoliales has root hairs, and all were either heavily or moderately infected with VA mycorrhizal fungi.

From the range of values shown by the Magnoliales in this study, it appears that a diameter of 0·3 mm is a reasonable, though somewhat arbitrary, dividing line between magnolioid and intermediate root types in this series. Root diameters of 29 species lacking root hairs were larger than 0·3 mm, and that of only one member of the Magnoliales (*Licaria* sp.) was smaller.

Intermediate and graminoid roots

None of the species studied was truely graminoid in the sense of having a constant dense cover of 1 to 2 mm long root hairs. Only 15 species had root hairs at all, of which four were 'rare', five 'inconstant' and six 'constant'. They ranged in length from 0·044 to 0·400 mm; one species was undetermined, because both suitable notes and material to measure were lacking.

Forty-five species without root hairs fell below the arbitrary 0·3 mm size limit for magnolioid roots. Having roots as fine as 0·052 mm, they could be considered of 'intermediate' root type, as would those having root hairs by the original classification.

Table 1. *Root characteristics and mycorrhizal condition of 89 tropical American plants of 23 orders*

Order	No. of species examined	No. of species >0·3 mm diam. ('magnolioid')	No. of species <0·3 mm w/o root hairs ('intermediate')	No. of species with root hairs ('graminoid-like')	Mycorrhizal category*			
					Non.	Light	Mod.	Heavy
Arecales	2	2	0	0	1	0	1	0
Asterales	1	1	0	0	0	0	0	1
Celastrales	3	1	1	1	2	1	0	0
Cyperales	3	0	2	1	1	0	2	0
Ebenales	7	2	2	3	3	1	3	0
Eriocaulales	1	0	1	0	1	0	0	0
Eufilicales	2	2	0	0	0	0	1	1
Gentianales	1	1	0	0	0	1	0	0
Lecithidales	9	3	6	0	3	6	0	0
Linales	2	0	2	0	0	1	1	0
Magnoliales	6	5	1	0	0	0	3	3
Malvales	5	1	3	1	0	1	4	0
Myrtales	5	1	4	0	2	1	0	2
Polemoniales	1	1	0	0	0	1	0	0
Polygalales	4	0	4	0	0	0	4	0
Rhamnales	1	0	1	0	1	0	0	0
Rosales	13	6	7	0	3	1	4	5
Rubiales	2	0	1	1	0	1	1	0
Santalales	2	1	0	1	1	0	1	0
Sapindales	7	0	5	2	2	1	1	3
Theales	3	0	1	2	1	0	1	1
Urticales	7	2	3	2	3	1	1	2
Violales	2	0	1	1	1	0	0	1

* Non. = non-mycorrhizal; light = lightly infected; mod. = moderately infected; heavy = heavily infected.

Association of VA infection and root type

The H statistic generated by the Kruskal–Wallis one-way analysis of variance by ranks was 12·52. With 3 degrees of freedom, this value is statistically significant ($P < 0{\cdot}01$). The 'heavily mycorrhizal' category had a preponderance of high ranks and the 'non-mycorrhizal' category of low ranks, which clearly shows where the differences lie: magnolioid roots tend to be heavily infected, while graminoid-like intermediate roots tend to be uninfected.

Discussion

The present investigation, although organized somewhat differently from Baylis's, supports certain contentions of that study. Examination of six tropical American representatives of the Magnoliales largely confirms his description of the generalized magnolioid root system. That these root systems were found to fall into the 'heavily

mycorrhizal' category with disproportionate frequency, while the most graminoid-like root systems fell disproportionately into the 'non-mycorrhizal' category, supports the specific hypothesis that magnolioid roots are more likely to be infected.

Magnolioid root systems were found to be very common in the tropical forest, in contrast to truly graminoid roots, which were apparently absent. Relatively fine roots or those bearing some development of root hairs were considerably less common than magnolioid roots but did occur. A good many of the intermediate types, as well as many graminoid-like and a few magnolioid roots, were completely non-mycorrhizal or only lightly infected. The significance of this will be discussed in a subsequent publication.

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