



**Influence of Plant Interactions on Vesicular-Arbuscular Mycorrhizal Infections. I.
Host and Non-Host Plants Grown Together**

J. A. Ocampo; J. Martin; D. S. Hayman

New Phytologist, Vol. 84, No. 1. (Jan., 1980), pp. 27-35.

Stable URL:

<http://links.jstor.org/sici?sici=0028-646X%28198001%2984%3A1%3C27%3AIOPIOV%3E2.0.CO%3B2-C>

New Phytologist is currently published by New Phytologist Trust.

Your use of the JSTOR archive indicates your acceptance of JSTOR's Terms and Conditions of Use, available at <http://www.jstor.org/about/terms.html>. JSTOR's Terms and Conditions of Use provides, in part, that unless you have obtained prior permission, you may not download an entire issue of a journal or multiple copies of articles, and you may use content in the JSTOR archive only for your personal, non-commercial use.

Please contact the publisher regarding any further use of this work. Publisher contact information may be obtained at <http://www.jstor.org/journals/npt.html>.

Each copy of any part of a JSTOR transmission must contain the same copyright notice that appears on the screen or printed page of such transmission.

The JSTOR Archive is a trusted digital repository providing for long-term preservation and access to leading academic journals and scholarly literature from around the world. The Archive is supported by libraries, scholarly societies, publishers, and foundations. It is an initiative of JSTOR, a not-for-profit organization with a mission to help the scholarly community take advantage of advances in technology. For more information regarding JSTOR, please contact support@jstor.org.

INFLUENCE OF PLANT INTERACTIONS ON VESICULAR-ARBUSCULAR MYCORRHIZAL INFECTIONS. I. HOST AND NON-HOST PLANTS GROWN TOGETHER

BY J. A. OCAMPO*, J. MARTIN* AND D. S. HAYMAN

*Soil Microbiology Department, Rothamsted Experimental Station,
Harpenden, Herts*

(Accepted 6 April 1979)

SUMMARY

Mycorrhizal infections formed by different endophytes were examined in 10 crop species grown separately and in pairs in sterilized and unsterile soils. No infection was observed in cabbage, kale, rape or swede (in the supposedly non-mycorrhizal family Cruciferae) and only traces were seen in sugar beet (supposedly non-mycorrhizal Chenopodiaceae) when these plants were grown alone. However, slight (< 5%) infection (cortical mycelium and vesicles, but no arbuscules) developed in some when a mycorrhizal host plant was present and there were many clumps of endophyte mycelium on their root surfaces, usually attached to entry points which had often aborted. *Glomus fasciculatus* 'E3' was a more infective endophyte than *Gigaspora margarita*. Infection was usually well developed in the host plants barley, lettuce, maize, potato and onion. It was depressed only in a few pairs but no more by the presence of a 'non-host' plant than by a host plant. The results suggest that the barriers to mycorrhizal infection in 'non-hosts' are intrinsic and more probably related to characteristics of the root cortex or epidermis than to any infection-inhibiting factors that might be released in root exudates.

INTRODUCTION

Most plant species can develop vesicular-arbuscular (VA) mycorrhizal infections in their roots. Exceptions are members of plant families reported to form little or no mycorrhiza, e.g. Cruciferae and Chenopodiaceae, or which usually form other types of mycorrhiza, e.g. Pinaceae, Ericaceae and Orchidaceae (see Gerdemann, 1968). Within a family some species become more infected than others, e.g. Leguminosae, where lupins are much less infected than clovers (Morley and Mosse, 1976; Trinick, 1977) and Gramineae where rye is generally less infected than wheat and barley (Strzemska, 1955).

Most studies on the development of VA mycorrhizal infections have been with plants grown separately, even though in nature different plant species often grow together with their roots intermingled. Results from the few studies concerning two plant species grown together indicate various interactions: a non-mycorrhizal plant may inhibit infection in a plant that is normally strongly mycorrhizal (Hayman, Johnson and Ruddlesdin, 1975; Iqbal and Qureshi, 1976; Morley and Mosse, 1976), or a strongly mycorrhizal 'nurse' plant may increase infection in a weakly mycorrhizal plant (Trinick and Mosse, 1975), or one mycorrhizal species may increase infection in another similarly mycorrhizal species (Fitter, 1977). Plant-plant interactions can

* Present address: Sección de Microbiología, Estación Experimental del Zaidín, Granada, Spain.

affect VA fungi differently from other root-associated fungi and bacteria (Christie, Newman and Campbell, 1978).

Clearly there is a need to extend our knowledge of plant factors affecting VA infections. We have attempted this with 10 crop species, growing them in various pairs in different soils and in the presence of different endophytes.

MATERIALS AND METHODS

The five mycorrhizal host plants used were barley (*Hordeum vulgare* L. cv. Julia), lettuce (*Lactuca sativa* L. cv. Fortune), maize (*Zea mays* L. cv. Cargill Primeur 170), onion (*Allium cepa* L. cv. Bedfordshire Champion) and potato (*Solanum tuberosum* L. cv. King Edward). The five plant species tested that were believed to be non-mycorrhizal (subsequently referred to as 'non-hosts') were cabbage (*Brassica oleracea* L. cv. Ormskirk), kale (*Brassica oleracea* L. cv. Maris Kestrel), oilseed rape (*Brassica napus* L. cv. Victor) and swede (*Brassica campestris* L. cv. Best of All), all in the family Cruciferae, and sugar beet (*Beta vulgaris* L. var. *rapa* cv. Kleine) in the Chenopodiaceae. They were raised in seed trays and transplanted as small seedlings to 75 mm pots of soil. There were five host/non-host combinations, namely barley-rape, lettuce-cabbage, maize-sugar beet, onion-swede and potato-kale, and controls where single non-mycorrhizal species and single or paired mycorrhizal species were grown. There were three replicates per treatment. The pots were placed on sand trays and the soil kept moist by water from below.

Two soils were used. In experiments 1 and 3 a fallow sandy loam from Woburn stackyard was used (40 parts 10^{-6} Olsen P, pH 4.9) after sterilizing with γ -irradiation (1 Mrad) and mixing 50:50 with sterilized sand. In experiment 2 a clay loam (56 parts 10^{-6} Olsen P, pH 6.1) from Long Hoos field at Rothamsted was used unsterile and mixed 50:50 with sand. This soil was collected from the root region of maize plants growing in Long Hoos and contained 204 spores (mostly *Glomus macrocarpus* var. *geosporus* and some *Glomus mosseae* and 'white reticulate') per 100 g soil and fragments of maize roots in which 57% of their length had VA infection (Ocampo and Hayman, 1980).

All plants in the sterilized soils were inoculated by spreading about 5 g of soil inoculum mixed with 5 g of sand throughout each pot. Soil inocula of *Glomus fasciculatus* 'E3' for experiment 1 and *Gigaspora margarita* for experiment 3 were collected from stock plant cultures and contained spores, mycelium and infected root fragments. In experiment 2 the natural mixed field endophyte population was kept unchanged.

Plants were harvested after 10 weeks and roots examined in detail for VA infection. To avoid the possibility of confusing hosts with 'non-hosts', the roots from each pot were carefully washed to separate the two species and only roots clearly attached to the main root system were examined, although in most pairs the two species were anatomically quite distinct. The whole root system of each plant was cleared in KOH and stained with trypan blue in lactophenol (Phillips and Hayman, 1970), cut into 1 cm segments that were mixed and repeatedly subdivided to yield random samples of 20 root segments per plant that were mounted on slides and examined at $\times 160$ magnification. Root infection was recorded as a percentage of root length infected and as a percentage of the cortex infected, the latter obtained by multiplying the length of infection by the average width of cortex infected for each segment. Numbers

of entry points (appressoria) and vesicles were counted and the proportion of infected roots containing arbuscules was estimated. For 'non-host' plants the amounts of external mycelium attached to each root segment were also noted.

RESULTS

Experiment 1

Table 1 shows that in irradiated Woburn soil all five mycorrhizal plants grown singly and inoculated with the E3 endophyte developed much VA infection (80 to 90% of root length infected). This was not significantly decreased by the presence of a 'non-host'. By contrast, infection was reduced in some instances where two mycorrhizal species were grown together, e.g. onion in which arbuscular (but not total) infection and numbers of entry points were higher in the presence of 'non-host' swedes but lower in the presence of the host plants barley and potato compared

Table 1. *VA infections in mycorrhizal host plants inoculated with E3 in sterilized soil*

Plant	Root infection		Entry points per cm root	Vesicles per cm root	Arbuscules*
	% length	% cortex			
Barley					
Alone	79	52	3.3	1.0	14
With rape	94	58	4.4	2.2	19
With onion	82	54	3.5	2.4	16
L.S.D. 5 %	27.6	6.6	1.70	4.19	6.5
Lettuce					
Alone	91	69	4.9	0.6	16
With cabbage	92	71	5.4	0.2	19
With onion	96	72	4.5	0.6	19
L.S.D. 5 %	21.2	19.4	3.17	1.05	14.1
Maize					
Alone	82	66	3.8	9.5	17
With sugar beet	95	87	8.4	2.7	33
With onion	62	51	4.0	1.1	12
L.S.D. 5 %	35.9	32.7	2.55	5.50	11.4
Potato					
Alone	79	59	5.3	2.4	21
With kale	48	39	2.7	1.3	16
With onion	68	44	4.7	2.0	16
L.S.D. 5 %	45.2	43.8	1.72	2.19	21.5
Onion					
Alone	82	63	5.4	2.9	19
With swede	90	82	8.5	2.4	31
With barley	83	58	3.3	1.2	5
With lettuce	84	58	3.5	10.6	17
With maize	76	62	6.3	5.7	10
With potato	88	62	3.2	0.7	1
L.S.D. 5 %	15.4	21.7	2.44	4.53	8.0

Each figure is the mean for 3 plants and 60 cm root.

* Estimated in categories: 10 = ~ 25% of the infection was arbuscular.

20 = ~ 50% of the infection was arbuscular.

30 = ~ 75% of the infection was arbuscular.

to onions growing alone. Both maize and onions appeared to develop more infection when grown alone or with a 'non-host' than when grown with each other, but this was not statistically significant. 'Non-hosts' increased numbers of entry points in host species in four (significantly in two) out of five pairs. Many more vesicles formed in onions when grown with lettuce than when grown alone or with other species, yet lettuce itself developed few vesicles.

Table 2. *VA* infections in 'non-host' plants inoculated with *E3* in sterilized soil

Plant	Internal mycelium*	External mycelium		Entry points per cm root	Internal vesicles per cm root	Arbuscules
		% root segments	% root length			
Cabbage						
Alone	0	2	< 1	0	0	0
With lettuce	0.7	52	12	0.6	0.2	0
Kale						
Alone	0	0	0	0	0	0
With potato	0.5	56	11	0.7	< 0.1	0
Rape						
Alone	0	7	1	0	0	0
With barley	0.1	47	9	0.4	< 0.1	0
Sugar beet						
Alone	0.2	18	4	0.1	0	0
With maize	2.0	60	23	1.2	8.9	0
L.S.D. 5%	—	36.2	26.1	0.38	—	—
Swede						
Alone	0	3	< 1	0	0	0
With onion	0.8	42	7	0.3	0.2	0
L.S.D. 5%	—	21.6	—	—	—	—

* 0.5 \approx 1% root length infected.

No infection developed in 'non-host' plants when grown alone except for traces of mycelium in the cortex of some sugar beet seedlings (Table 2). However, four of the five 'non-host' species developed slight (0.5 to 2.0%) infection and one had a trace when grown with a host plant (Table 2; Plate 1, Nos 1, 2 and 3; Plate 2, Nos 2 and 3). Furthermore, several clumps of external mycelium were observed around roots of all five 'non-host' species in the presence of a host species and some entry points were formed although these usually aborted (Plate 2, No. 4). Secondary spores (external vesicles) were formed on the external mycelium attached to three of these species (Plate 2, No. 1). No arbuscules were observed in any 'non-host' plant.

Experiment 2

Considerably less VA infection developed in barley, lettuce, potato and onion from the mixed endophyte population in unsterile field soil (Table 3) than when these same crops were inoculated with *E3* in sterilized soil (Table 1). Maize, in contrast, was equally highly infected in both soils, but whether this was due to any specificity in host susceptibility to endophytes in the naturally infected maize root fragments present in this soil was not ascertained.

As in experiment 1, VA infection in a host plant was not significantly decreased by the presence of a non-host plant in all five pairs tested. Indeed in barley it was highest when rape roots were present. When two mycorrhizal host plants were grown together the amount of VA infection appeared unaffected, except for a large decrease in onion roots grown with lettuce, although there was no converse effect of onion on lettuce. Compared to the sterilized soil amended with E3, there was less arbuscular infection in all crops except maize grown in the unsterile field soil and fewer entry points were formed. Few distinct arbuscules were seen in onion.

Table 3. VA infections in mycorrhizal host plants in unsterile soil

Plant	Root infection		Entry points per cm root	Vesicles per cm root	Arbuscules
	% length	% cortex			
Barley					
Alone	25	15	1.1	0.9	3
With rape	61	33	1.6	1.0	11
With onion	37	21	1.0	0.7	6
L.S.D. 5 %	23.7	12.4	0.75	2.87	4.3
Lettuce					
Alone	51	25	2.0	1.4	6
With cabbage	41	20	1.8	1.1	1
With onion	52	32	2.0	2.4	1
L.S.D. 5 %	23.7	12.1	1.04	4.19	3.7
Maize					
Alone	79	59	4.0	5.3	14
With sugar beet	88	66	4.4	2.8	17
With onion	77	55	5.4	2.5	16
L.S.D. 5 %	5.5	11.6	2.02	6.16	8.9
Potato					
Alone	34	23	1.4	0.2	9
With kale	33	21	1.2	0.3	10
With onion	24	17	0.9	0.1	8
L.S.D. 5 %	22.0	17.7	0.41	0.60	8.1
Onion					
Alone	59	37	4.1	3.6	1
With swede	62	39	2.1	6.1	0
With barley	38	20	1.0	1.5	0
With lettuce	15	5	0.5	1.1	1
With maize	50	30	2.0	5.4	2
With potato	81	46	1.9	0.8	0
L.S.D. 5 %	40.9	34.6	2.10	6.51	2.7

Legend as for Table 1.

The pattern for the five 'non-host' plants (Table 4) was similar to experiment 1, namely VA infection and external mycelium were absent when they were grown alone but there was considerable external mycelium on their roots when a host plant was present. Kale, rape and sugar beet had traces of infection (< 1% root length) when grown with potato, barley and maize, respectively. There were a few aborted entry points in all cases but no secondary spores or arbuscules were seen.

Experiment 3

Gigaspora margarita (Table 5) proved to be a less infective endophyte than E3 (Table 1) on lettuce and maize in the same sterilized soil, although it was not significantly less infective than E3 on onions. This suggests some host-endophyte specificity in terms of the relative susceptibility of different plant species to different mycorrhizal fungi. Maize significantly reduced infection in onion and lettuce. *G. margarita* also formed fewer entry points and arbuscules than E3. No vesicles were observed in the root cortex of any *G. margarita*-inoculated host plants.

Table 4. *VA infections in non-host plants in unsterile soil*

Plant	Internal mycelium*	External mycelium		Entry points per cm root	Internal vesicles per cm root	Arbuscules
		% root segments	% root length			
Cabbage						
Alone	0	0	0	0	0	0
With lettuce	0	13	2	0.1	0	0
Kale						
Alone	0	0	0	0	0	0
With potato	0.1	28	10	0.1	0	0
Rape						
Alone	0	0	0	0	0	0
With barley	0.1	13	2	0.1	0	0
Sugar beet						
Alone	0	0	0	0	0	0
With maize	0.1	25	4	0.1	0	0
Swede						
Alone	0	0	0	0	0	0
With onion	0	18	3	0.1	0	0

* 0.5 \approx 1 % root length infected.

'Non-host' plants were also less infected by *G. margarita* than E3, with no internal mycelium observed in either species tested (Table 6). No external mycelium of *G. margarita* was observed around cabbage or swede roots grown without a host plant whereas there was a little on those inoculated with E3. There was much external mycelium of *G. margarita* on 'non-host' roots in the presence of a host plant (Plate 1, No. 4) but again considerably less than on similar plant pairs with E3. No *G. margarita* entry points, aborted or not, were observed on cabbage roots with or without lettuce but a few occurred on swedes grown with onions.

DISCUSSION

These experiments show clearly that so-called 'non-host' plants can become slightly mycorrhizal in the presence of a host plant, *Beta vulgaris* var. *rapa* more so than various *Brassica* crops. Nevertheless, infection was usually limited to well under 5% of the root length and arbuscules were never seen. Typical VA infections, for which the presence of both vesicles and arbuscules in the cortex is a diagnostic feature, were absent from all 'non-host' roots. Denser infections have been reported in another crucifer (*Atriplex canescens*; Williams, Wollum and Aldon, 1974) but again

arbuscules were not observed. Also, Kruckelmann (1975) found some mycorrhizal infection in some Cruciferae and Chenopodiaceae species that he inoculated. More recently Hirrel, Mehravaran and Gerdemann (1978) reported some vesicular infection in four species of Chenopodiaceae and two species of Cruciferae when grown with mycorrhizal onion or citrus. The lack of infection of 'non-hosts' by *G. margarita*

Table 5. *VA infections in mycorrhizal host plants inoculated with Gigaspora margarita in sterilized soil*

Plant	Root infection		Entry points per cm root	Vesicles per cm root	Arbuscules
	% length	% cortex			
Lettuce					
Alone	26	15	1.1	0	7
With cabbage	33	24	0.9	0	10
With onion	22	13	0.9	0	6
With maize	6	2	0.4	0	<1
L.S.D. 5%	14.1	11.9	0.38	—	5.5
Maize					
Alone	39	26	1.7	0	10
With onion	21	12	1.3	0	6
With lettuce	24	14	1.2	0	5
L.S.D. 5%	11.3	10.2	0.54	—	4.3
Onion					
Alone	73	52	1.6	0	11
With swede	66	38	0.7	0	5
With lettuce	9	3	0.3	0	1
With maize	5	1	0.3	0	1
L.S.D. 5%	22.7	17.5	0.21	—	8.1

Legend as for Table 1.

Table 6. *VA infections in 'non-host' plants inoculated with Gigaspora margarita in sterilized soil*

Plant	Internal mycelium*	External mycelium		Entry points per cm root	Internal vesicles per cm root	Arbuscules
		% root segments	% root length			
Cabbage						
Alone	0	0	0	0	0	0
With lettuce	0	17	2	0	0	0
Swede						
Alone	0	0	0	0	0	0
With onion	0	35	5	0.1	0	0

* 0.5 \approx 1% root length infected.

may be related to the fact that it is primarily an arbuscular endophyte and appears not to form vesicles in typical host plants.

The appreciable development of mycorrhizal hyphae around roots of 'non-host' plants and the numerous attempts at penetration as shown by the many aborted entry points suggests that the barriers to infection are intrinsic and that outside the

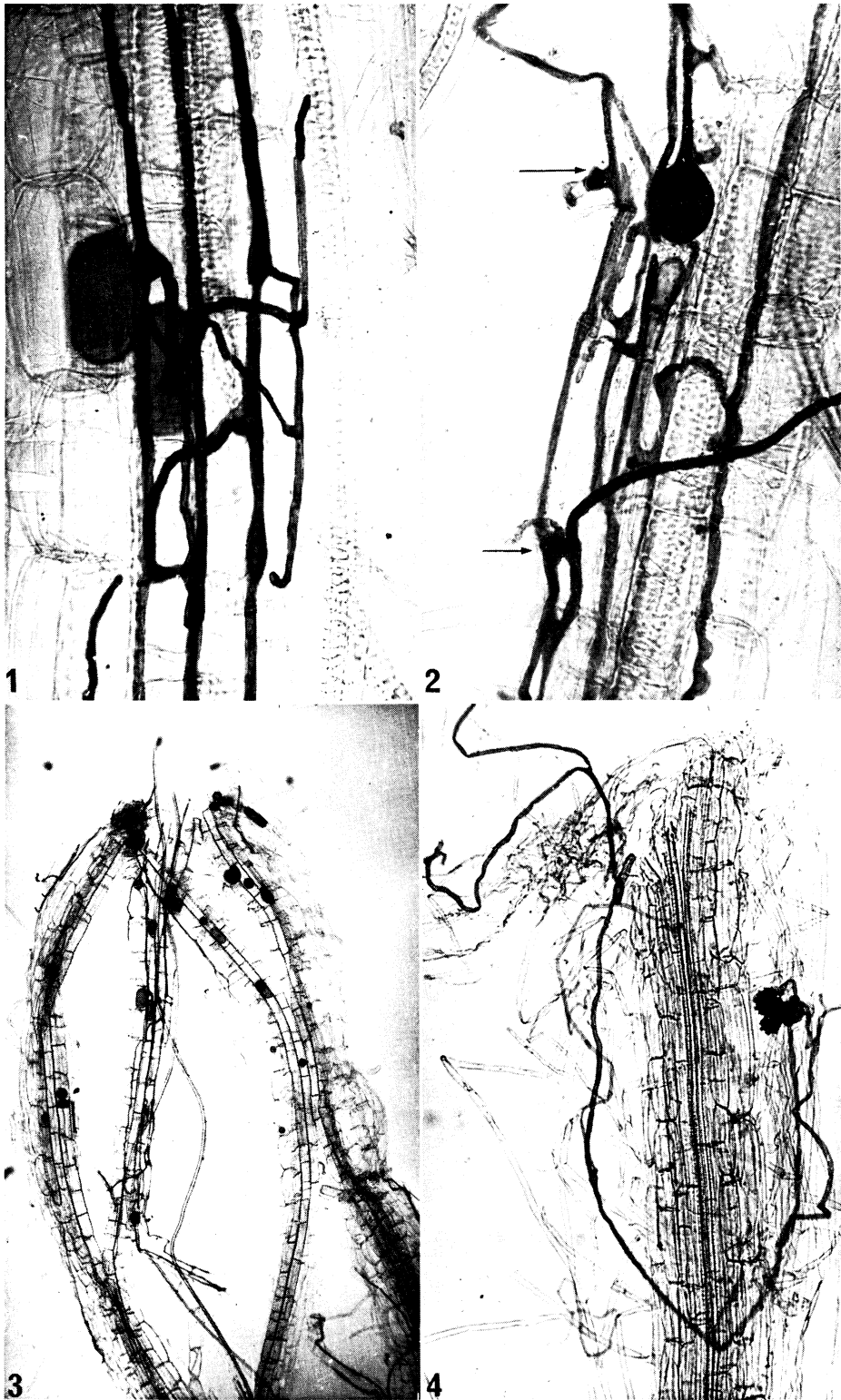
roots physical factors such as root hair density or structure of the epidermis, perhaps, may be more inhibitory than chemical factors such as root exudates. This contrasts with some earlier studies with host/'non-host' pairs where it was speculated that root exudates (Hayman *et al.*, 1975; Iqbal and Qureshi, 1976) or seed-coat components (Morley and Mosse, 1976) might depress VA infection in normal host plants. The present study suggests that the mycorrhizal fungus can derive some benefit from the roots of 'non-host' plants and make some growth around them, especially if host plant roots are present. Consequently the presence of a so-called 'non-host' plant may favour the development of VA mycorrhizal fungi in soil more than the absence of either 'non-hosts' or hosts. This agrees with Bevege and Bowen (1975) who observed some external colonization of roots of *Arabidopsis* (Cruciferae) and *Pinus radiata* by certain VA endophytes.

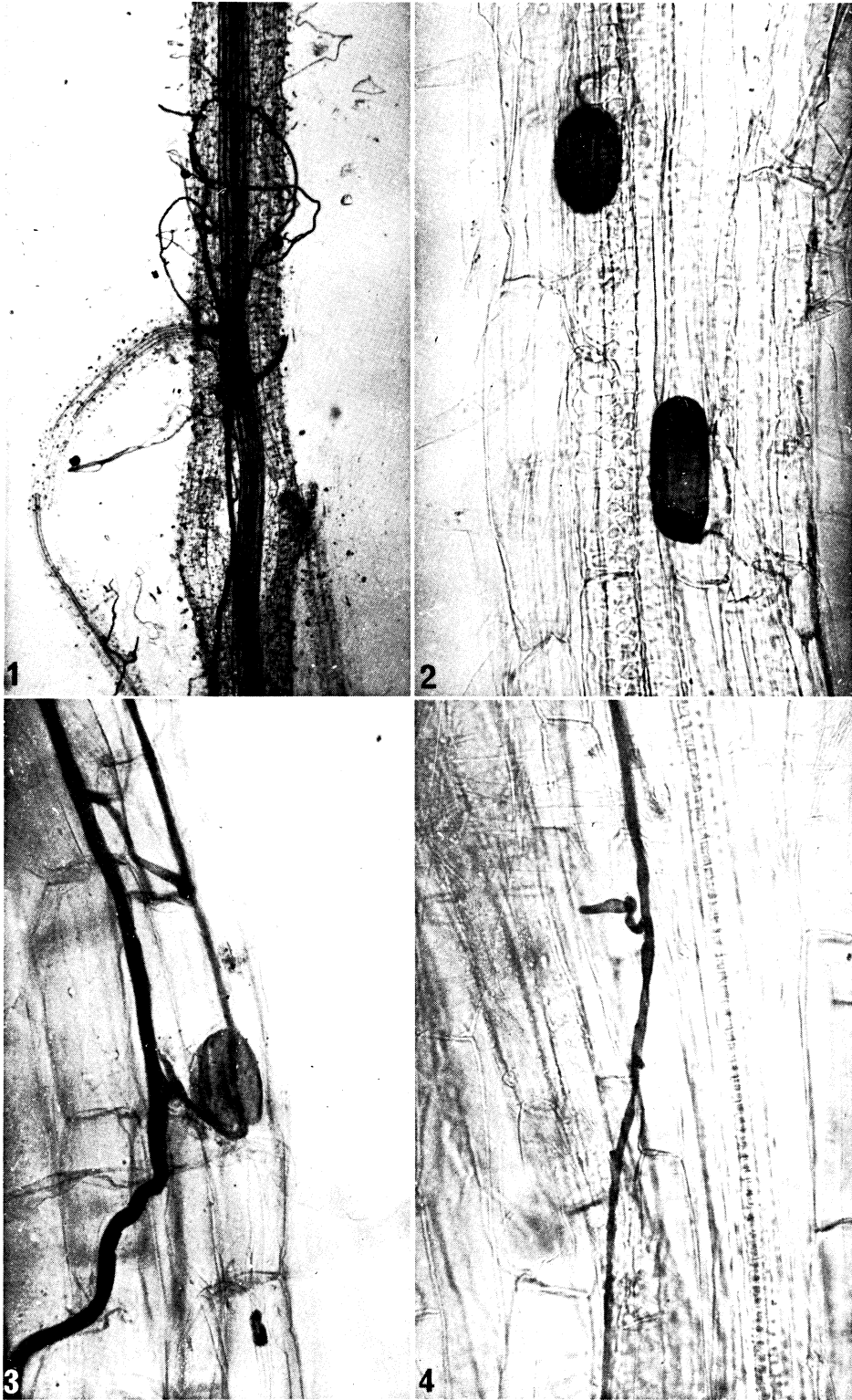
Not only was the possible, albeit limited, beneficial effect of a 'non-host' root on a VA endophyte unexpected in our study, but the occasional negative effect of mixing two host species was also surprising. This might be attributed to changes in root density or preferential colonization of one of the two host plants. There was no relationship between the amount of infection and plant size.

Finally, these results show that more attention should be paid to plant interactions affecting mycorrhizal development since plants do not normally grow in communities of one species. Interspecific mycorrhizal effects may be one of the factors contributing to allelopathic interactions and competition between plants in mixed communities.

REFERENCES

- BEVEGE, D. I. & BOWEN, G. D. (1975). *Endogone* strain and host plant differences in development of vesicular-arbuscular mycorrhizas. In *Endomycorrhizas* (Ed. by F. E. Sanders, Barbara Mosse & P. B. Tinker), pp. 77-86. Academic Press, New York and London.
- CHRISTIE, P., NEWMAN, E. I. & CAMPBELL, R. (1978). The influence of neighbouring grassland plants on each others' endomycorrhizas and root-surface microorganisms. *Soil Biology and Biochemistry*, **10**, 521-527.
- FITTER, A. H. (1977). Influence of mycorrhizal infection on competition for phosphorus and potassium by two grasses. *New Phytologist*, **79**, 119-125.
- GERDEMANN, J. W. (1968). Vesicular-arbuscular mycorrhiza and plant growth. *Annual Review of Phytopathology*, **6**, 397-418.
- HAYMAN, D. S., JOHNSON, A. M. & RUDDLES DIN, I. (1975). The influence of phosphate and crop species on *Endogone* spores and vesicular-arbuscular mycorrhiza under field conditions. *Plant and Soil*, **43**, 489-495.
- HIRREL, M. C., MEHRAVARAN, H. & GERDEMANN, J. W. (1978). Vesicular-arbuscular mycorrhizae in the Chenopodiaceae and Cruciferae: do they occur? *Canadian Journal of Botany*, **56**, 2813-2817.
- IQBAL, S. H. & QURESHI, K. S. (1976). The influence of mixed sowing (cereals and crucifers) and crop rotation on the development of mycorrhiza and subsequent growth of crops under field conditions. *Biologia (Pakistan)*, **22**, 287-298.
- KRUCKELMANN, H. W. (1975). Effects of fertilizers, soils, soil tillage, and plant species on the frequency of *Endogone* chlamydo spores and mycorrhizal infection in arable soils. In *Endomycorrhizas* (Ed. by F. E. Sanders, Barbara Mosse & P. B. Tinker), pp. 511-525. Academic Press, New York and London.
- MORLEY, C. D. & MOSSE, B. (1976). Abnormal vesicular-arbuscular mycorrhizal infections in white clover induced by lupin. *Transactions of the British Mycological Society*, **67**, 510-513.
- OCAMPO, J. A. & HAYMAN, D. S. (1980). Effects of pesticides on mycorrhiza in field-grown barley, maize and potatoes. *Transactions of the British Mycological Society* (in press).
- PHILLIPS, J. M. & HAYMAN, D. S. (1970). Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions of the British Mycological Society*, **55**, 158-161.
- STRZEMSKA, J. (1955). Investigations on mycorrhiza in cereals (in Polish). *Acta Microbiologica Polonica*, **4**, 191-204.





J. A. OCAMPO, J. MARTIN & D. S. HAYMAN

- TRINICK, M. J. (1977). Vesicular-arbuscular infection and soil phosphorus utilization in *Lupinus* spp. *New Phytologist*, **78**, 297-304.
- TRINICK, M. J. & MOSSE, B. (1975). *Rothamsted Report for 1974*, part 1, p. 253.
- WILLIAMS, S. E., WOLLUM, A. G. & ALDON, E. F. (1974). Growth of *Atriplex canescens* (Pursh) Nutt. improved by formation of vesicular-arbuscular mycorrhizae. *Soil Science Society of America Proceedings*, **38**, 962-965.

EXPLANATION OF PLATES

PLATE 1

Roots of cabbage grown with lettuce and mycorrhizal inoculum of either *Glomus fasciculatus* 'E3' (Nos 1 to 3) or *Gigaspora margarita* (No. 4).

No. 1. Vesicles and hyphae inside root. $\times 400$.

No. 2. Entry points (arrowed), internal hyphae and external spore/vesicle. $\times 400$.

No. 3. Vesicular infection. $\times 62$.

No. 4. External mycelium and secondary spores. $\times 160$.

PLATE 2

Roots of sugar beet, swede, kale and oilseed rape grown with maize, onion, potato and barley, respectively, and with inoculum of *Glomus fasciculatus* 'E3'.

No. 1. Sugar-beet root with external mycelium and spores. $\times 62$.

No. 2. Two vesicles inside swede root. $\times 400$.

No. 3. Kale root with external hypha (arrowed) and internal hyphae and vesicle. $\times 400$.

No. 4. External hypha and aborted entry point (arrowed) on rape root. $\times 400$.