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## Association of dark septate endophytes and arbuscular mycorrhizal fungi in potato under field conditions in the northeast region of India

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Despite numerous studies on arbuscular mycorrhizal fungi (AMF) in potato (*Solanum tuberosum* L.), nothing is known about colonization by dark septate endophytes (DSE). In a study of DSE and AMF associations in a potato field at Meghalaya, northeast India, monthly DSE and AMF colonization ranged from  $7.9 \pm 2.9$ – $13.2 \pm 1.65$  and  $6.98 \pm 3.13$ – $12.4 \pm 3.53\%$ , respectively. There were no significant differences in monthly DSE and AMF colonization. AMF hyphal colonization had a significant negative correlation with pH and available phosphorus ( $p < 0.05$ ). Total AMF colonization was negatively correlated with organic carbon ( $p < 0.05$ ). DSE colonization showed a significant negative correlation with root hair density ( $p < 0.05$ ). However, AMF colonization, although negatively correlated with root hairs, was not significant. Sixteen AMF morphotypes were identified from the genera *Acaulospora*, *Gigaspora*, *Glomus*, *Pacispora* and *Scutellospora*; a few remain unidentified. The results indicated that DSE and AMF colonization progressed synchronously with the dominance of *Glomus tortuosum*, *Pacispora boliviensis* and *Gigaspora margarita*.

**Keywords:** arbuscular mycorrhizal fungi; dark septate endophyte; *Solanum tuberosum*; morphotypes; root hair density

### 1. Introduction

Mycorrhizal symbiosis has been frequently thought to be identical with mutualistic symbiosis, where fungi benefit by acquiring carbohydrates from a host and the host, in turn, gains by improved growth and more efficient nutrient acquisition. Dark septate endophytes (DSE) are a miscellaneous group of root-inhabiting conidial sterile ascomycetous fungi that colonize living plant roots without causing noticeable harmful effects (Jumpponen 2001). Arbuscular mycorrhizal fungi (AMF) are involved in plant mineral nutrition, an association which is prevalent in agricultural plants (Smith and Read 1997). Both groups are essential for successful low-input agricultural practices. Hence, the formation and functioning of AMF symbiosis will play an important role in sustainable agriculture (Schreiner and Bethlenfalvay 1995).

*Solanum tuberosum* L. (Potato) is a staple food in northeast India and studies on AMF and DSE will assist sustainable agriculture in this region. Studies on AMF associations in potato have shown that phosphate fertilization suppressed AMF colonization (Black and Tinker 1977). Bhattarai and Mishra (1984) reported that AMF colonization increased with age in potato plants. Potato plants have a low root density (Pursglove and Sanders 1981) and high growth potential; thus, AMF symbiosis may be of particular significance in coping with P deficiency stress in natural ecosystems. This is also true for

commercial potato production, since significant yield increases due to AMF have been recorded (McArthur and Knowles 1991). A reduction in phosphate fertilizer use can also be achieved in potato by inoculating with AMF (Sieverding 1991). Morphological features, such as root hair development, were greater on non-mycorrhizal roots than on AMF roots (McArthur and Knowles 1993). Davies et al. (2005) reported an increase in potato yield with AMF inoculation; in addition, flavanoid increased the extraradical hyphae of AMF. A study of the AMF large-subunit (LSU) rRNA gene analyzing the fungal community suggested that potato roots were preferentially colonized by *Glomus intraradices* Schenck & Smith 1982 (Cesaro et al. 2008).

Despite these studies, nothing is known about DSE colonization. The present study investigated the association between DSE and AMF in potato.

### 2. Methods

#### 2.1. Site description

In September 2007, two potato plots, each  $100 \times 50 \text{ m}^2$  in area, were selected for root and soil sampling in Swer village (latitude  $25^\circ 25' \text{ N}$  and longitude  $91^\circ 47' \text{ E}$ ) in the East Khasi Hills District of Meghalaya in northeast India. The site is situated about 34 km south of Shillong on the road to Cherrapunjee. Altitude varies between 1910 and 1975

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m.a.s.l. The climate is monsoonal with distinct warm-wet and cold-dry seasons. The rainy season starts in May and continues till October.

Potato is grown in two seasons i.e. in summer it extends from February to June/July, while in autumn it extends from July/August to November/December. The local Khasi tribal farmers grow varieties of *Kufri Jyoti*, a high yielding potato cultivar which covers over 50% of the total potato growing area in the state.

## 2.2. Sampling

Potato tubers were planted in August 2007 and sampling was conducted on the first week of subsequent months, i.e. September, October and November; after the emergence of stalks from the tuber. Three plants from each plot were sampled for root and 500 g soil at a depth of 0–25 cm, and combined into one sample. In addition, six samples were collected randomly, three from each plot, during the last month (November) and again combined into one sample for an AMF diversity study. The roots were fixed in FAA (Johansen 1940) and soil samples were air dried after analysis of pH and moisture content for further soil physico-chemical studies.

## 2.3. Root processing

The fixed roots were washed in tap water, cleared in 10% KOH at 90°C and stained with black Faber Castell stamp pad ink (Das and Kayang 2008). Fifty segments of ~1 cm long stained root samples were mounted on slides in lactoglycerol and examined for AMF and DSE structures under a light microscope (Olympus 41209). Estimations of AMF and DSE colonization were done by the magnified intersection method (McGonigle et al. 1990).

## 2.4. Root hair density measurement

Root hair density measurements were performed on root segments similar to those examined for quantification of fungal colonization. The root segments were counted three-dimensionally by adjusting the microscope's plane of focus (Ma et al. 2001).

## 2.5. AMF spore diversity

The spores were extracted by a modified wet sieving and decanting method (Muthukumar et al. 2006). The soil weighing 100 g was dispersed in 1 l of water and decanted through a series of 710- to 38- $\mu$ m sieves. The residues were filtered through gridded filter papers and all whole spores were counted using a light microscope at 40 $\times$  magnification. Sporocarps and spore clusters were considered as one unit. The isolated spores were picked up with a needle in polyvinyl alcohol/lactoglycerol under a microscope

(Koske and Tessier 1983) and also in mixed polyvinyl alcohol/lactoglycerol–Meltzer's reagent (1:1, v/v) for identification. Complete and broken spores were examined using a light microscope. Taxonomic identification of spores to species level was based on sporocarpic size, colour, ornamentation and wall characteristics by matching original descriptions (<http://www.invam.caf.wvu.edu>; Koske et al. 1986; Blaszkowski 1989; Almeida and Schenck 1990; Wu et al. 1995; Oehl and Sieverding 2004). Photography of the root segments colonized by fungi and spores of AMF was via a Leica EC 3 camera attached in a Leica dm 1000 microscope. Spore density, relative abundance (RA) and species richness (SR) were all recorded (Zhao and Zhao 2007).

## 2.6. Soil properties

Soil texture was analyzed using the sodium hexameta-phosphate method (Allen et al. 1974). For moisture content (%), 10 g subsamples of soil were oven-dried and the final weight determined. Measurement of pH was done using a microprocessor-based Pocket pH yester 2 (Eutech Instruments). Available phosphorus in the soil was determined following the molybdenum-blue method (Allen et al. 1974). Soil organic carbon was estimated using a colorimetric method (Anderson and Ingram 1993).

## 2.7. Data analysis

A one-sample *t*-test was conducted to analyse the monthly variation in the soil physico-chemical properties. All colonization variables were submitted to one-way ANOVA and Tukey's HSD test was used for comparison of means. Pearson correlation was done to determine relationships between the soil physico-chemical properties, fungal colonization and spore density. The data were analyzed with Statistica 9.0 software.

# 3. Results

## 3.1. Soil properties

The mean values of soil physico-chemical characteristics are presented in Table 1, which varies significantly in all

Table 1. Selected physico-chemical properties of soil from a potato field in Meghalaya, India.

Soil properties	Mean values
Sand (%)	82.36
Silt (%)	3.56
Clay (%)	14.08
Moisture content (% $\pm$ SE)	28.21 $\pm$ 1.76
pH ( $\pm$ SE)	5.36 $\pm$ 0.08
Organic carbon (% $\pm$ SE)	11.45 $\pm$ 0.61
Available phosphorus (mg/kg $\pm$ SE)	5.94 $\pm$ 0.94

three months ( $p < 0.05$ ). Moisture content of soil samples ranges between  $25.38(\pm 3.9)$  and  $31.17(\pm 2.35)\%$ . pH varies between  $5.17(\pm 0.06)$  and  $5.43(\pm 0.11)$ . The percentage of organic carbon and available phosphorus ranged from  $10.28(\pm 0.26)$ – $12.36(\pm 0.74)\%$  and  $4.7(\pm 0.54)$ – $7.8(\pm 0.92)$  mg/kg, respectively.

### 3.2. Fungal colonization and root hair density

Root hairs and mycorrhizal structures were confirmed, viz. intraradical hyphae, vesicles, arbuscules, microsclerotia and septate hyphae (Figure 1). The monthly means for fungal colonization and AMF spore density are presented in Table 2. There was a minor increase in monthly fungal colonization. AMF and DSE colonization ranged from  $6.98(\pm 3.13)$ – $12.4(\pm 3.53)\%$  and  $7.9(\pm 2.9)$ – $13.2(\pm 1.65)\%$ , respectively. There were no significant differences in the monthly total AMF and DSE colonization ( $p < 0.05$ ). However, there was a significant difference in monthly AMF structural colonization i.e. vesicle colonization ( $p < 0.05$ ). Monthly fungal colonization is shown in Figure 2. Root hair density/cm of root segment ranged from  $42.12(\pm 6.54)$ – $58.86(\pm 6.67)$ .

Correlation values are given in Table 3, where AMF hyphal colonization was negatively correlated with pH and available phosphorus. There was an insignificant negative colonization between root hair density and AMF colonization. However, there was a significant negative correlation between root hair density and DSE. In addition, there was a significant negative correlation between organic carbon and AMF colonization. No significant relation was observed between spore density and AMF colonization.

### 3.3. AMF spore diversity

Sixteen morphotypes were identified from soil in the potato field (Figure 3). There were seven species from genus *Glomus*, three from *Pacispora*, four from *Acaulospora* and one each from *Gigaspora* and *Scutellospora*, and a few were unidentified. The dominant AMF species were *Glomus tortuosum* Schenck & Smith, followed by *Pacispora boliviana* Sieverd. & Oehl, and *Gigaspora margarita* Becker & Hall (Table 4). The relative abundance of AMF species in the potato field follows the trend *Glomus* > *Pacispora* > *Gigaspora* > *Acaulospora* > *Scutellospora* with 52.54, 23.25, 19.23, 4.55 and 0.43%, respectively. There were significant differences ( $p < 0.05$ ) in relative abundance between genera. The species richness of *Glomus*, *Acaulospora*, *Pacispora*, *Gigaspora* and *Scutellospora* was 7.3, 4.7, 2.7, 1.3 and 0.7, respectively. There were no significant differences ( $p < 0.05$ ) in species richness of the different genera.

## 4. Discussion

AMF colonization was found to increase slightly from 1-month to 2-month-old plants; Bhattarai and Mishra (1984) reported that AMF colonization increased with the age of the plant. There was also an increase in DSE colonization from the first to last month. The arbuscular peak was observed earlier than the vesicular peak, which supports the observation of Bhattarai and Mishra (1984). DSE colonization has also been observed to occur simultaneously with AMF, as reported previously (Jumpponen and Trappe 1998), and the present study suggest that DSE colonization progress in the same trend as AMF.

Potato plants have been characterized as relatively ineffective at acquiring soil phosphorus (Pursglove and Sanders 1981), but are capable of morphological and physiological adaptations during phosphorus deficiency that may substantially improve P acquisition (Cogliatti and Clarkson 1983). Morphological changes, such as the presence of root hairs, may help in phosphorus acquisition. In general, colonization is comparatively low, which may be due to the high availability of phosphorus coupled with the presence of root hairs, as decreased fungal colonization in the roots with high levels of phosphorus was reported by Galvez et al. (2001). Moreover, DSE possesses a penetrating potential through root hairs (O'Dell et al. 1993). However, the inverse correlation between DSE and root hair density was significantly high, which is in accordance with the study of Muthukumar and Udaiyan (2002). They reported an inverse relationship between root morphology and AMF colonization levels in Cycads, where the length of root colonized by AMF was proportional to the length and density of root hairs. Since mycorrhiza function as extensions of the root system, it is possible that species with fewer and short root hairs are more dependent on AMF than species with more abundant and longer root hairs.

Synchronized colonization by DSE and AMF could possibly provide a support system during periods when mycorrhizal fungi are adversely affected by environmental conditions, as reported previously (Jumpponen and Trappe 1998), and, in the present case, to combat host susceptibility to disease. The negative correlation between AMF colonization and soil organic carbon may be due to the input of organic manure, which agrees with the report of Piotrowskia et al. (2008) that AMF hyphal length, inoculum potential and colonization of roots were all suppressed at sites with greatest litter and soil organic matter accumulation. No significant relation was observed between spore density and AMF colonization, which is in accordance with the study of Moreira et al. (2006).

Bharadwaj et al. (2007) showed that *G. intraradices* and *G. mosseae* (Nicol. & Gerd.) (Gerd. & Trappe) are



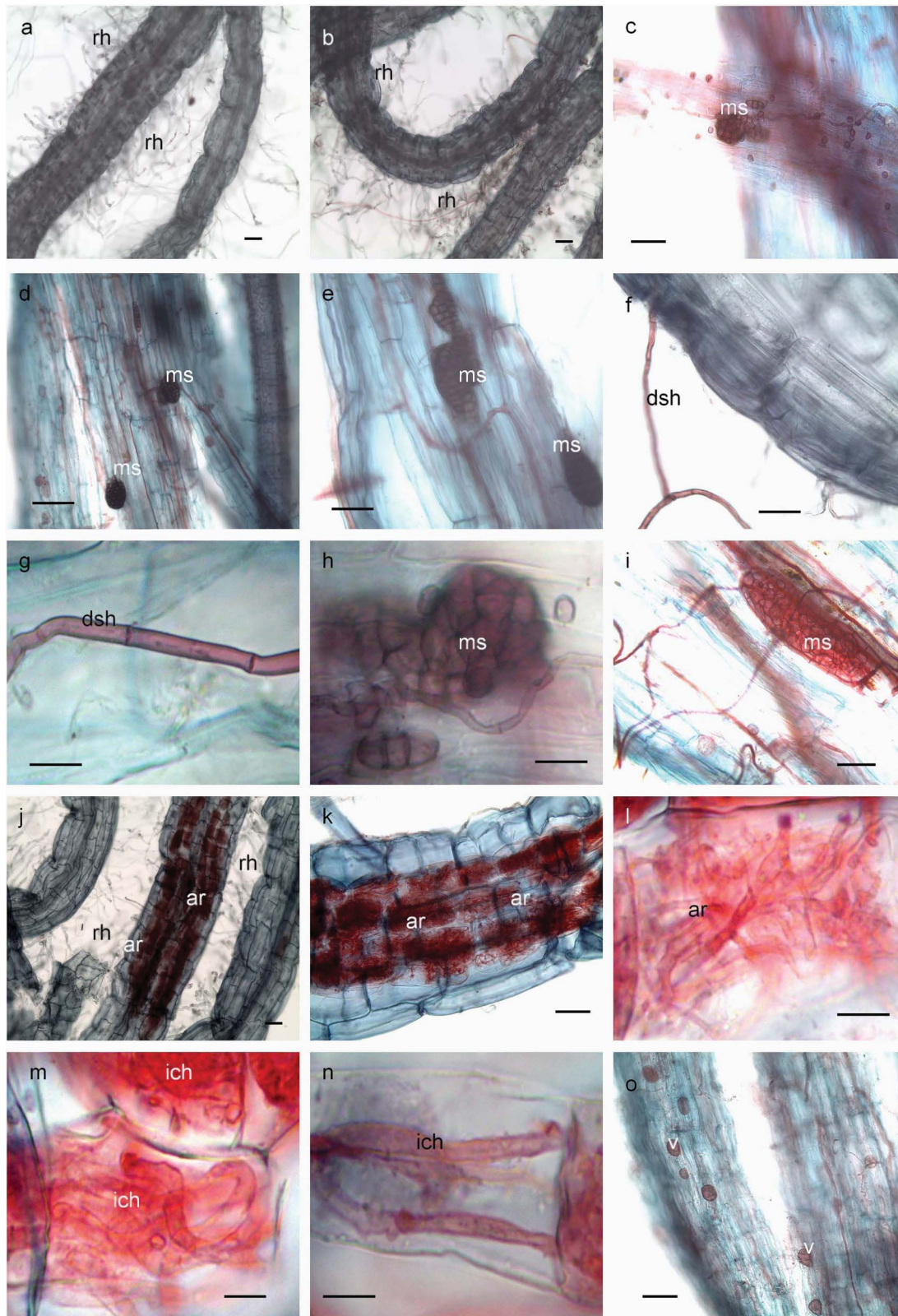


Figure 1. Dark septate endophyte, arbuscular mycorrhizal colonization and root structure of potato. (a & b) Root segments with root hairs. Bar = 1.2 mm & 1.2 mm, respectively; (c-i) portions of roots showing DSE colonization. Bar = 300µm, 100µm, 100µm, 100µm, 50µm, 50µm, & 150µm, respectively; (j-o) roots showing AMF structures. Bar = 1.2mm, 300µm, 50µm, 75µm, 50 µm & 100µm, whereas; rh-root hairs, ms-microsclerotia, dsh-dark septate hyphae, ar-arbuscules, ich-intracellular hyphae and v-vesicles.

Table 2. AMF and DSE colonization of potato and monthly AMF spore density.

Months	DSE	Arbuscules	Vesicles	Hyphae	Total AMF	Spore density/100 g soil
September	7.9 ± 2.9a	6.16 ± 2.85a	0.0b	0.82 ± 0.34a	6.98 ± 3.13a	502.77 ± 19.74c
October	11.1 ± 1.45a	6.40 ± 2.29a	0.04 ± 0.04ab	2.36 ± 0.73a	8.8 ± 2.94a	366.0 ± 23.71c
November	13.2 ± 1.65a	9.08 ± 2.85a	0.52 ± 0.26a	2.8 ± 0.66a	12.4 ± 3.53a	405.0 ± 17.21c

Notes: ± indicate standard error.

Tukey's HSD test showing different letters differed significantly ( $p < 0.05$ ).

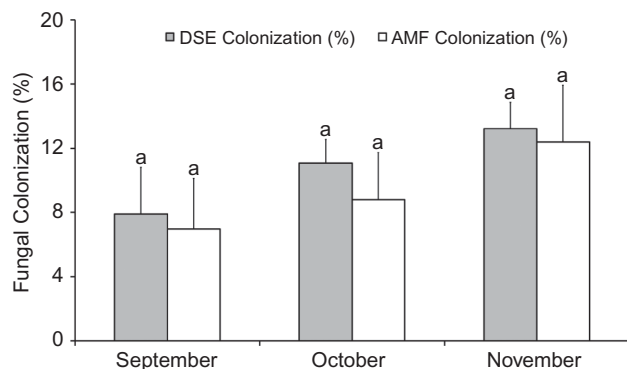


Figure 2. Arbuscular mycorrhizal and dark septate endophyte colonization in the roots of potato collected from three months. Tukey HSD test showing same alphabet with no significant differences ( $p < 0.05$ ).

the most common fungi in potato roots and soil. Cesaro et al. (2008) confirmed that *Glomus* species were identified, regardless of some pedological dissimilarity. In addition, they reported that *G. intraradices* was the preferred AMF species colonizing potato root. Our findings suggest that *Glomus* spp. were abundant, particularly *G. tortuosum*. This is the first report of *Pacispora* spp. in these soils and also to our knowledge from soils of India (Das and Kayang 2010). *P. boliviana* may favour this sub-tropical region, as ubiquitously

reported (Oehl and Sieverding 2004). However, *G. microaggregatum* Koske, Gemma & P.D. Olexia 1986 was found inside a nematode cyst, whereas the same species was reported to occur inside the dead spores of other AMF species (Koske et al. 1986). *G. aggregatum* Schenck & Smith 1982 was also recovered inside an unknown spore-like structure.

## 5. Conclusion

In the present investigation, a number of species isolated from the potato field remain unidentified; therefore, appropriate trap culture techniques will be the focus of further studies. For instance, species such as *G. tortuosum* and *P. boliviana*, were found in high abundance. A comprehensive study is now required, including the highly abundant AMF species, to examine the growth and nutrition of potato plants. Furthermore, it is unclear what factors are responsible for the suppression of mycorrhizal fungal colonization, although they are likely related to the introduction of inorganic fertilization as normal agriculture practice in this region. However, AMF species were found to be abundant; therefore, a proper awareness program is necessary for local farmers to access the benefits of these biological fertilizers. This work provides a platform to analyse the role of AMF and DSE in plant growth and nutrition in several other crop plants in northeast India.

Table 3. Correlation coefficients between fungal colonization, root hair density and soil physico-chemical properties.

Variables	DSE	Ves	Ar	Hy	AMF	MC	pH	C	P	RH
Dark septate endophyte (DSE)	1.0	0.84	0.84	0.98	0.95	0.42	-0.97	-0.94	-0.97	-0.99*
Vesicles (Ves)		1.0	1.0**	0.72	0.96	0.84	-0.69	-0.97	-0.71	-0.87
Arbuscules (Ar)			1.0	0.72	0.96	0.84	-0.7	-0.97	-0.71	-0.87
Hyphae (Hy)				1.0	0.87	0.24	-0.99*	-0.86	-0.99*	-0.96
Total arbuscular mycorrhizal fungi (AMF)					1.0	0.68	-0.86	-0.99*	-0.86	-0.97
Moisture content (%) (MC)						1.0	-0.21	-0.7	-0.22	-0.48
pH							1.0	0.84	0.99**	0.95
Organic carbon (%) (C)								1.0	0.85	0.96
Available phosphorus (mg/kg) (P)									1.0	0.96
Root hair density (RH)										1.0

Note \*Significant at  $p < 0.05$ . \*\*Significant at  $p < 0.01$ .



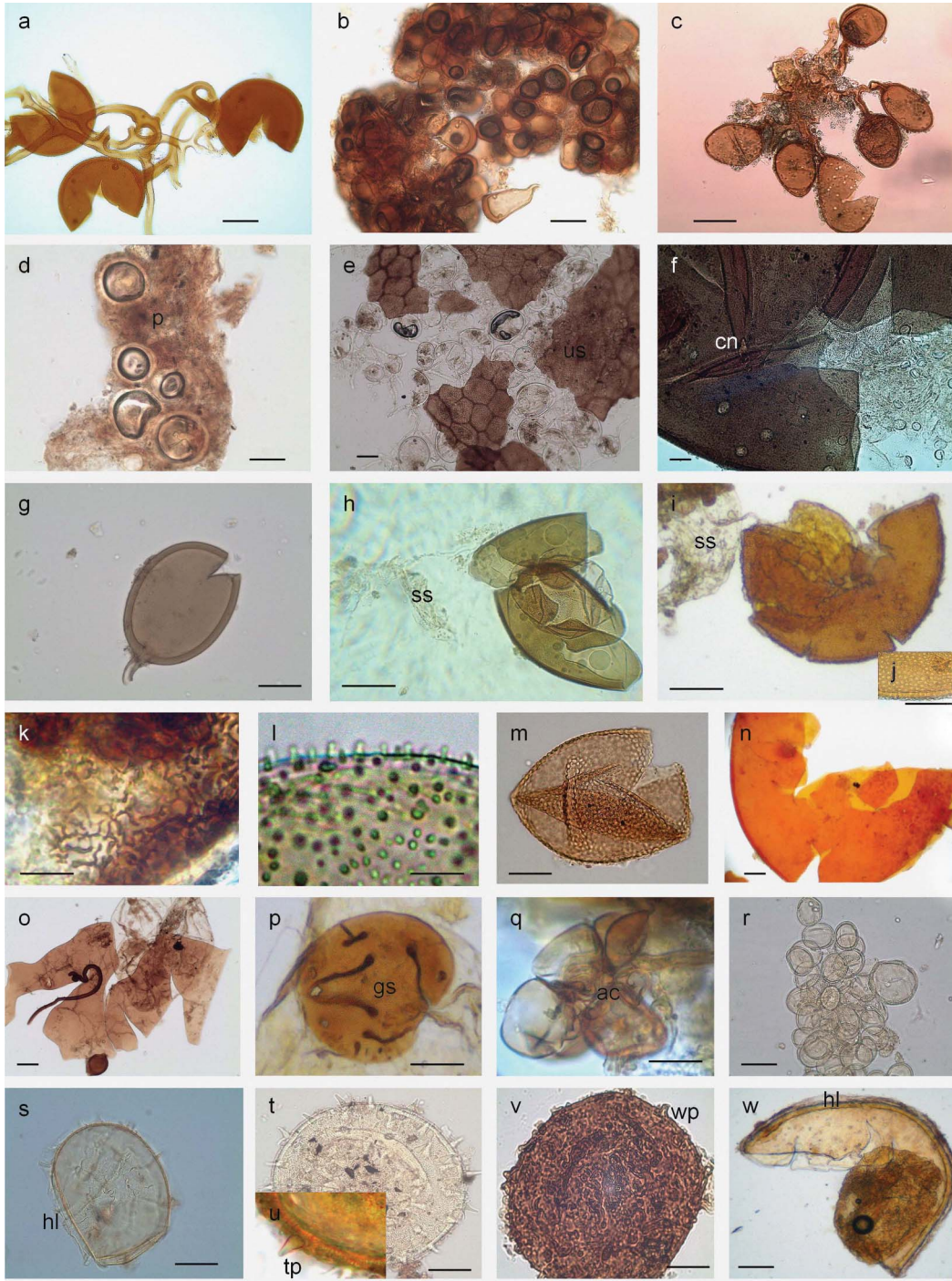


Figure 3. Arbuscular mycorrhizal fungi isolated from potato field. (a) *Glomus macrocarpum*. Bar = 200 $\mu$ m; (b) *G. clavisporum*. Bar = 150 $\mu$ m; (c) *G. fuegianum*. Bar = 200 $\mu$ m; (d) peridium [p] surrounding the spores of *G. tortuosum*. Bar = 200 $\mu$ m; (e) spores of *G. aggregatum* inside unidentified spore [us]. Bar = 500 $\mu$ m; (f) spores of *G. microaggregatum* inside cyst nematode [cn]. Bar = 500 $\mu$ m; (g) *Glomus* sp 1. Bar = 100 $\mu$ m; (h) *Acaulospora tuberculata* with sporiferous saccule [ss]. Bar = 50 $\mu$ m; (i) *A. cavernata* with sporiferous saccule. Bar = 50 $\mu$ m; (j) pitted ornamentation on the wall of *A. cavernata*. Bar = 10 $\mu$ m; (k) ridges on the surface of *A. rehmi*. Bar = 10 $\mu$ m; (l) wall of *Pacispora chimonobambusae* with clavate projections. Bar = 10 $\mu$ m; (m) *P. boliviana* with ornamented pits. Bar = 100 $\mu$ m; (n) *Gigaspora margarita*. Bar = 400 $\mu$ m; (o) *Scutellospora* sp 2. Bar = 300 $\mu$ m; (p) germination shield [gs] of *Scutellospora* sp 2. Bar = 20 $\mu$ m; (q) auxiliary cells [ac] of *Scutellospora* sp 2. Bar = 10 $\mu$ m; (r) *Pacispora* like spore with ornamentation on the wall. Bar = 200 $\mu$ m; (s) hyaline layer [hl] on the wall of unidentified species. Bar = 100 $\mu$ m; (t) unidentified species. Bar = 100 $\mu$ m; (u) thorn like projection [tp] on the wall of unidentified species. Bar = 25 $\mu$ m; (v) wart like projection [wp] on the wall of unidentified species. Bar = 100 $\mu$ m & (w) hyaline layer on the wall of unidentified species. Bar = 200 $\mu$ m.

Table 4. Relative abundance of AMF species in the potato field.

Species	Relative abundance (%)
Acaulosporaceae	
<i>Acaulospora cavernata</i> Blaszk.	1.67
<i>A. rehmi</i> Sieverd. & Toro	0.22
<i>A. tuberculata</i> Janos & Trappe	1.35
<i>Acaulospora</i> sp. 2	1.32
Gigasporaceae	
<i>Gigaspora margarita</i> Becker & Hall	19.23
<i>Scutellospora</i> sp. 2	0.43
Glomeraceae	
<i>Glomus aggregatum</i> Schenck & Smith	0.39
<i>G. clavisporum</i> (Trappe) Almeida & Schenck	0.22
<i>G. feugianum</i> (Speg.) Trappe & Gerd.	0.22
<i>G. microaggregatum</i> Koske, Gemma & P.D. Olexia	1.25
<i>G. macrocarpum</i> Tul. & Tul.	0.57
<i>G. tortuosum</i> Schenck & Smith	49.7
<i>Glomus</i> sp. 1	0.22
Pacisporaceae	
<i>Pacispora boliviana</i> Sieverd. & Oehl	22.43
<i>P. chimonobambusae</i> (C.G. Wu & Y.S. Liu) Sieverd. & Oehl.	0.64
<i>Pacispora</i> sp. 1	0.18
Total AMF 16	99.9

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## References

- Allen SE, Grimshaw HM, Parkinson JA, Quaramby C. 1974. Chemical analysis of ecological materials. Oxford: Blackwell.
- Almeida RT, Schenck NC. 1990. A revision of the genus *Sclerocystis* (Glomeraceae, Glomales). *Mycologia* 82: 703–714.
- Anderson JM, Ingram JSI. 1993. Tropical soil biology and fertility: A handbook of methods. Wallingford (UK): CAB International.
- Bharadwaj DP, Lundquist P, Alströma S. 2007. Impact of plant species grown as monocultures on sporulation and root colonization by native arbuscular mycorrhizal fungi in potato. *Appl Soil Ecol.* 35: 213–225.
- Bhattarai ID, Mishra RR. 1984. Study on the vesicular-arbuscular mycorrhiza of three cultivars of potato (*Solanum tuberosum* L.). *Plant Soil* 79: 299–303.
- Black RLB, Tinker PB. 1977. Interaction between effects of vesicular-arbuscular mycorrhiza and fertiliser phosphorus on yields of potatoes in the field. *Nature* 267: 510–511.
- Blaszkowski J. 1989. *Acaulospora cavernata* (Endogonaceae): a new species from Poland with pitted spores. *Crypt Bot.* 1: 204–207.
- Cesaro P, van Tuinen D, Copetta A, Chatagnier O, Berta G, Gianinazzi S, Lingua G. 2008. Preferential colonization of *Solanum tuberosum* L. roots by the fungus *Glomus intraradices* in arable soil of a potato farming area. *Appl Environ Microbiol.* 74: 5776–5783.
- Cogliatti DH, Clarkson DT. 1983. Physiological changes in, and phosphate uptake by potato plants during development of, and recovery from phosphate deficiency. *Physiol Plant.* 58: 287–294.
- Das P, Kayang H. 2008. Stamp pad ink, an effective stain for observing arbuscular mycorrhizal structure in roots. *Wor J Agric Sci.* 4: 58–60.
- Das P, Kayang H. 2010. Mycorrhizal colonization and distribution of arbuscular mycorrhizal fungi associated with *Michelia champaca* L. under plantation system in northeast India. *J For Res.* 21:137–142.
- Davies Jr. FT, Calderón CM, Huaman Z. 2005. Influence of arbuscular mycorrhizae indigenous to Peru and a flavonoid on growth, yield, and leaf elemental concentration of ‘Yungay’ potatoes. *Hort Sci.* 40: 381–385.
- Galvez L, Douds DD, Drinkwater LE, Wagoner P. 2001. Effect of tillage and farming system upon VAM fungus populations and mycorrhizas and nutrient uptake of maize. *Plant Soil* 228: 299–308.
- Johansen DA. 1940. Plant microtechnique. New Delhi: Tata McGraw-Hill.
- Jumpponen A, Trappe JM. 1998. Dark septate endophytes: a review of facultative biotrophic root-colonizing fungi. *New Phytol.* 140: 295–310.
- Jumpponen A. 2001. Dark septate endophytes-are they mycorrhizal? *Mycorrhiza* 11: 207–211.
- Koske RE, Tessier B. 1983. A convenient, permanent slide mounting medium. *Mycol Soc Am News* 34: 59.
- Koske RE, Gemma JN, Olexia PD. 1986. *Glomus microaggregatum*, a new species in the Endogonaceae. *Mycotaxon* 26: 125–132.
- Ma Z, Bielenberg DG, Brown KM, Lynch JP. 2001. Regulation of root hair density by phosphorus availability in *Arabidopsis thaliana*. *Plant Cell Environ.* 24: 459–467.
- McArthur DAJ, Knowles NR. 1991. Response of ‘Russet Burbank’ potato to V.A. mycorrhizal fungi. *Proceeding of the Prairie Potato Council.*
- McArthur DAJ, Knowles NR. 1993. Influence of vesicular-arbuscular mycorrhizal fungi on the response of potato to phosphorus deficiency. *Plant Physiol.* 101: 147–160.
- McGonigle TP, Miller MH, Evans DG, Fairchild GL, Swan JA. 1990. A new method which gives an objective measure of colonization of roots by vesicular-arbuscular mycorrhizal fungi. *New Phytol.* 115: 495–501.
- Moreira M, Baretta D, Tsai SM, Cardoso EJBN. 2006. Spore density and root colonization by arbuscular mycorrhizal fungi in preserved or disturbed *Araucaria angustifolia* (Bert.) O. Ktze. ecosystems. *Sci Agric.* 63: 380–385.
- Muthukumar T, Udaiyan K. 2002. Arbuscular mycorrhizas in cycads of southern India. *Mycorrhiza* 12: 213–217.
- Muthukumar T, Senthilkumar M, Rajangam M, Udaiyan K. 2006. Arbuscular mycorrhizal morphology and dark septate fungal associations in medicinal and aromatic plants of Western Ghats, Southern India. *Mycorrhiza* 17: 11–24.
- O’Dell TE, Massicotte HB, Trappe JM. 1993. Root colonization of *Lupinus latifolius* Agardh. and *Pinus contorta* Dougl. by *Phialocephala fortinii* Wang & Wilcox. *New Phytol.* 124: 93–100.
- Oehl F, Sieverding E. 2004. *Pacispora*, a new vesicular-arbuscular mycorrhizal fungal genus in the Glomeromycetes. *J Appl Bot.* 78: 72–82.
- Piotrowskia JS, Morford SL, Rillig MC. 2008. Inhibition of colonization by a native arbuscular mycorrhizal fungal community via *Populus trichocarpa* litter, litter extract, and soluble phenolic compounds. *Soil Biol Biochem.* 40: 709–717.



- Pursglove JD, Sanders FE. 1981. The growth and phosphorus economy of the early potato (*Solanum tuberosum*). *Commun Soil Sci Plant Anal.* 12: 1105–1121.
- Schreiner RP, Bethlenfalvay GJ. 1995. Mycorrhizal interactions in sustainable agriculture. *Critic Rev Biotechnol.* 15: 271–285.
- Sieverding E. 1991. Vesicular–arbuscular mycorrhizal management in tropical agrosystems. Eschborn, Germany: Deutsche Gesellschaft für Technische Zusammenarbeit.
- Smith SE, Read DJ. 1997. Mycorrhizal symbiosis. Academic Press: London.
- Wu CG, Liu YS, Hwuang YL, Wang YP, Chao CC. 1995. Glomales of Taiwan: V. *Glomus chimonobambusae* and *Entrophospora kentinensis*, spp. nov. *Mycotaxon* 53: 283–294.
- Zhao D, Zhao Z. 2007. Biodiversity of arbuscular mycorrhizal fungi in the hot-dry valley of the Jinsha River, southwest China. *Appl Soil Ecol.* 37: 118–128.