ONION (*Allium cepa* 'Calibra')
Host response; Mycorrhizal colonization

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The effect of soil phosphorus levels on colonization of onion roots by arbuscular mycorrhizal fungi, 2016-2017.

Symbiotic arbuscular mycorrhizal fungi (AMF) colonize the roots of many plant species, helping mine soil for immobile nutrients, particularly phosphorus (P), enhancing plant growth, and limiting the severity of some plant diseases. Onions depend on AMF to compensate for the relatively sparse, unbranched root system. Colonization of onion roots by natural populations of AMF can be inhibited by practices such as fumigation, tillage, cover cropping with AMF incompatible crops (e.g., brassicas), and excessive fertilization, particularly high levels of soil P. A growth chamber trial was completed in 2016-2017 to assess the influence of soil P level on colonization of onion roots and growth of onion plants in soil amended with a commercial AMF product, MYKOS Gold Granular (Reforestation Technologies Inc., Gilroy, CA). The chamber was at 15 ± 1°C with a 12 h photoperiod/day to evaluate the AMF inoculant using a randomized complete block design with 5 replications of a 2 x 3 factorial treatment design: inoculated or not with AMF at each of three soil P levels (19, 40, and 81 mg/kg). Soil with low P (average Olsen P of 19 mg/kg) and nitrogen (average total N of 4.0 mg/kg) was collected from the Washington State University Extension Farm in Pasco, WA, steam-pasteurized twice (30 min at 65-70°C each time, with 24 h between pasteurizations), air-dried, and sieved to a particle size ≤2 mm. Triple superphosphate (0-45-0, Wilbur Ellis Co., Mount Vernon, WA) was blended at 0, 304.11, and 578.03 mg/kg into samples of the pasteurized soil for 10 min using a cement mixer to achieve the three levels of soil P. After 2 weeks, 30 g of MYKOS Gold granular were mixed with 2,970 g pasteurized soil for each of the three P levels using a twin shell blender (1% inoculant concentration by weight). A 500 g aliquot of the control soil (not amended with MYKOS Gold Granular) and inoculated soil for each level of P was added to each of five replicate D40 deepots (each 25 cm deep x 7 cm diameter, Stuewe & Sons, Inc., Tangent, OR) after sealing the drain holes at the lower end of each deepot with paper towels. Water (150 ml) was added to the soil in each deepot in 50 ml increments. Seeds (n = 10) of the onion cv. Calibra were placed on the soil surface and covered with 17 g (1 tablespoon) of dry soil not amended with MYKOS Gold Granular. The top end of each deepot was covered for 2-3 days with a plastic bag to reduce evaporation during emergence. Water was added to the soil in each deepot at 2- to 3-day intervals, as needed. At 4 weeks after planting, 25 ml of a suspension of 2/5th-strength nitrate-type Long Ashton fertilizer (without micronutrients or P) was added to the soil in each deepot. Starting 5 weeks after planting, the fertilizer was added to each deepot twice a week. Onion seedling height and total above-ground dry weight of all plants/deepot were measured 8 weeks (56 days) after seeding. The roots in each deepot were washed with running tap water, and then stained for AMF by boiling in 10% KOH followed by boiling in a 5% Sheaffer black ink and vinegar solution (Verheilig et al., 1998) to quantify the extent of root colonization by AMF microscopically (5x to 20x magnification) based on the presence or absence of AMF in each of 40 root sections, using a modified gridline intersection method (Giovannetti and Mosse, 1980). Foliar nutrient analyses were completed for the dried leaves by Soiltest Farm Consultants (Moses Lake, WA). Analyses of variance (ANOVA) and Fisher's protected least significant differences (LSD) at P <0.05 for means comparisons were computed with SAS (SAS Institute Inc., Cary, NC). The trial was repeated with soil P levels of 19, 42, and 70 mg/kg following addition of the same amounts of triple super phosphate noted above.

Table 1 shows results of the main effects and means comparisons for each trial. Table 2 shows results of variables in each trial for which the ANOVA interaction term was significant. AMF colonization of onion roots averaged 30.3 ± 4.5 and 19.7 ± 4.6% for plants grown in soil inoculated with MYKOS Gold Granular in Trials 1 and 2, respectively (averaged across soil P levels) vs. 0% colonization for plants in control soil in both trials (Table 1). In each trial, the amount of root colonization decreased with increasing soil P level, from >40% in soil with 18-19 mk P/kg soil to <20% in soil with 70-81 mg P/kg soil (Table 1). Despite root colonization by AMF, onion plants growing in soil inoculated with MYKOS Gold Granular in Trial 1 had significantly less shoot dry weight and foliar K, Fe, and Mn; but greater foliar total N, S, and Na (Table 1). There were no significant differences in shoot height and foliar P, B, Ca, Cu, Mg, P, and Zn between the control soil and soil amended with MYKOS Gold Granular. As expected, foliar P levels increased with increasing soil P levels (0.316, 0.479, and 0.612% in soil with 19, 40, and 81 mg P/kg soil, respectively) (Table 1). Plants grown in soil with 40 mg P/kg had the greatest shoot height and dry weight, but the least foliar Fe and Mn compared to plants growing in soil with 19 and 81 mg P/kg. Soil P levels did not affect foliar B, Ca, Cu, K, Mg, N, Na, S, or Zn significantly. There were significant interactions between the effects of MYKOS Gold Granular and soil P levels on shoot dry weight as well as foliar Cu and K (Table 2). Shoot dry weight was greater in plants grown at 40 mg P/kg soil then in soil with 19 or 81 mg P/kg, regardless of whether the soil was amended or not with MYKOS Gold Granular. Foliar Cu and K were most concentrated in plants grown in soil with the least P (19 mg/kg) and amended with MYKOS Gold Granular compared to plants in other soil treatment combinations. In Trial 2, there was no significant differences in shoot height, shoot dry weight and levels of any foliar nutrients of plants growing in non-inoculated soil or soil amended with MYKOS Gold Granular (Table 1). In fact, shoot height and dry weight were greatest for plants in soil with the least P (18 mg/kg soil), with no differences in shoot height or dry weight of plants in soil with 42 and 70 mg P/kg soil. As expected, foliar P concentration increased significantly (almost doubled) with increasing soil P level from 18 to 42 mg/kg soil, but there was no significant difference in foliar P levels of plants in soil with 42 vs. 70 mg P/kg. Total Foliar N was greatest in plants grown in soil not amended with AMF that had an intermediate P level (42 mg/kg soil) compared to plants in the other five soil treatment combinations, reflecting a significant interaction effect (Table 2). Overall, the results demonstrate the potential negative effect of higher soil P levels on colonization of onion roots by AMF, and the need to assess the potential impacts of soil fertility levels on the value of using AMF inoculants in onion production. Further studies are needed to examine interactions between soil nutrient levels, AMF colonization of onion roots, and onion growth and nutrient status.

Table 1.

Variable measured (unit of measurement)	AMF treatment[z]			Soil P level (mg/kg)			
	Trial 1						
	Control	MYKOS	*P* value[y]	19 mg/kg	40 mg/kg	81 mg/kg	*P* value[y]
AMF root colonization (%)	0	30.3	-	48.0 a	22.3 b	17.5 b	0.0021
Shoot height (cm)	16.71	16.50	0.9801	16.27 b	17.94 a	16.02 b	0.0624
Shoot dry weight (g)	0.636 a	0.555 b	0.0123	0.561 b	0.696 a	0.565 b	0.0037
Foliar P (%)	0.468	0.468	0.9227	0.316 c	0.479 b	0.612 a	0.0001
Foliar K (%)	6.261 a	5.904 b	0.0388	6.36	6.03	5.74	0.0966
Total foliar N (%)	3.05 b	3.61 a	0.0157 Rank[x]	3.72	3.04	3.12	0.1200 Rank
Foliar Ca (%)	1.21	1.16	0.1511 Rank	1.20	1.20	1.16	0.1507 Rank
Foliar Mg (%)	0.302	0.299	1.0000 Rank	0.292 a	0.300 a	0.310 a	0.0637 Rank
Foliar S (%)	0.666 b	0.751 a	0.0097	0.701 a	0.697 a	0.720 a	0.0575
Foliar Zn (mg/kg)	24.00	26.23	0.1551 Rank	27.70	23.43	23.6	0.1669 Rank
Foliar Fe (mg/kg)	6.08 a	5.86 b	0.0123 Log[x]	6.13 a	5.69 b	6.02 a	0.0060
Foliar Mn (mg/kg)	180.29 a	118.62 b	0.0001	175.30 a	89.57 b	168.60 a	0.0001
Foliar Cu (mg/kg)	4.36	4.36	0.9821	4.66	4.24	4.15	0.5490
Foliar B (mg/kg)	25.86	25.31	0.6769 Rank	25.00	24.57	26.90	0.3168 Rank
Foliar Na (%)	0.032 b	0.040 a	0.0234	0.037	0.029	0.040	0.1166
	Trial 2						
	Control	MYKOS	*P* value[y]	18 mg/kg	42 mg/kg	70 mg/kg	*P* value[y]
AMF root colonization (%)	0	19.7	-	42.9 a	27.3 a	5.1 b	0.0239
Shoot height (cm)	17.20	15.07	0.5192 Rank	18.81 a	16.74 b	12.55 b	0.0083
Shoot dry weight (g)	0.70	0.86	0.4946 Rank	1.23 a	0.61 b	0.54 b	0.0178 Rank
Foliar P (%)	0.41	0.40	0.9520	0.24 b	0.43 a	0.53 a	0.0001
Foliar K (%)	4.39	4.84	0.1335	4.61	4.57	4.71	0.9463
Total foliar N (%)	2.51	2.38	0.4046	2.38 ab	2.67 a	2.25 b	0.0273
Foliar Ca (%)	0.91	1.00	0.2119	0.94	0.91	1.04	0.2594
Foliar Mg (%)	0.23	0.25	0.2792	0.23	0.23	0.26	0.2895
Foliar S (%)	0.46	0.51	0.1215	0.47	0.52	0.46	0.2698
Foliar Zn (mg/kg)	21.62	23.67	0.8460 Rank	20.00	23.40	24.67	0.3151 Rank
Foliar Fe (mg/kg)	635.80	506.20	0.8403 Rank	375.20	717.30	589.80	0.0886 Rank
Foliar Mn (mg/kg)	135.15	162.53	0.1029	141.56	128.50	181.78	0.0694 Sqrt[x]
Foliar Cu (mg/kg)	4.22	4.11	0.6909 Rank	3.87	4.45	4.13	0.6708 Rank
Foliar B (mg/kg)	17.75	18.73	0.5375 Rank	17.20	20.17	17.26	0.8078 Rank
Foliar Na (%)	0.039	0.048	0.1706 Rank	0.037	0.046	0.049	0.3585 Rank

[z] MYKOS Gold Granular was added to pasteurized soil at 1% by weight (30 g product + 2,970 g soil) = 0.8 spores/g soil.

[y] Within each row, numbers followed by the same letter are not significantly different based on Fisher's protected LSD.

[x] Original means are shown for all variables. Rank = means separation is based on rank-transformed data because of heterogeneous variances among treatments. Log and Sqrt = means separation was based on log and square root transformations of the data, respectively, to meet assumptions for parametric data analysis.

Table 2.

Variable (unit of measurement)	No AMF (control)			AMF-inoculated soil			*P* value[*]
	Trial 1						
	19 mg/kg	40 mg/kg	81 mg/kg	19 mg/kg	40 mg/kg	81 mg/kg	
Shoot dry weight (g)	0.640 ab	0.780 a	0.576 bc	0.482 c	0.680 a	0.554 bc	0.0016
Foliar K (%)	5.89 b	5.82 b	5.99 b	6.83 a	5.64 b	6.06 b	0.0254
Foliar Cu (mg/kg)	4.36 ab	4.35 ab	4.38 ab	4.96 a	4.10 b	3.92 b	0.0434
	Trial 2						
	18 mg/kg	42 mg/kg	70 mg/kg	18 mg/kg	42 mg/kg	70 mg/kg	
Total foliar N (%)	2.16 b	3.07 a	2.16 b	2.55 b	2.27 b	2.32 b	0.0031

[*] Within each row, numbers followed by the same letter are not significantly different based on Fisher's protected LSD.