The Impact of Inoculation on the Tripartite Association Between Pulse Crops, Rhizobia and Arbuscular Mycorrhizal Fungi

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INTRODUCTION

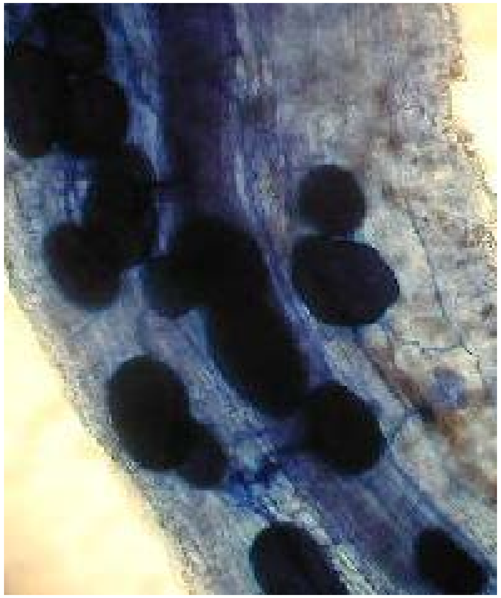

Fig. 1. A stained AMF colonized root *(http://aggie-horticulture.tamu.edu)*

Fig. 2. Nodulated roots of pea*(Dr. J. Burgess/Science Photo Library)*

- Arbuscular mycorrhizal fungi (AMF): obligate biotrophs, improved P uptake.
- Rhizobia: symbiotic N-fixing bacteria, form root nodules in legumes

- Rhizobia are known to improve the bioavailability of N, and this is may be enhanced in mycorrhized roots (Barea et al., 2002), and

- Jia et al. (2004) concluded that plant productivity was enhanced by the the rhizobia/AMF/*Vicia* tripartitite relationship and some effects were either synergistic or additive, depending on the growth parameter assessed.

- Legume mutant plants impaired in nodulation are also unable to associate with Glomeromycota (Lima et al., 2009)

- Lipochitooligosaccharides(LCOs) activate a signaling pathway called the common symbiotic pathway (CSP), which controls both the rhizobium-legume (RL) and the arbuscular mycorrhizal (AM) symbioses and they are conserved (Kouchi et al., 2010)

Rhizobium–legume symbiosis shares an exocytotic pathway required for arbuscule formation

Sergey Ivanov[a,1], Elena E. Fedorova[a], Erik Limpens[a], Stephane De Mita[a,2], Andrea Genre[b], Paola Bonfante[b], and Ton Bisseling[a,c,3]

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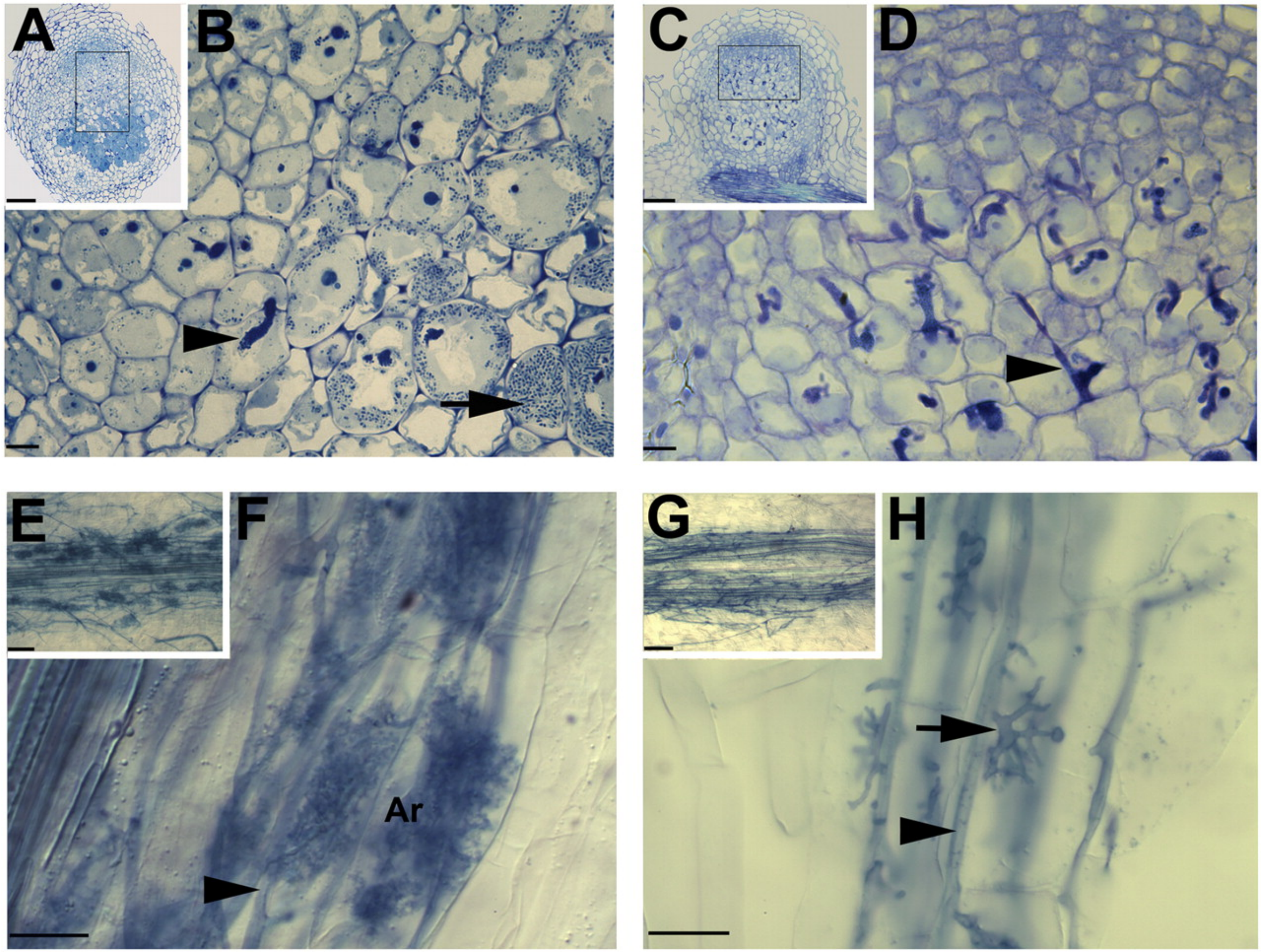
- Both AMF and rhizobial associations involve the formation of specialized plant-based membranes, which surround the microbes and forms a "symbiotic interface" where the host plant and the microbes exchange nutrients in a controlled manner.
- Ivanov et al. (2012) suggests that the signaling pathway for the formation of the membrane is similar between rhizobia-legume and AMF-legume, and that the former "co-opted" the pathway from the more ancient arbuscular mycorrhizal symbiosis to form a symbiotic interface.

MtVAMP721d and MtVAMP721e are required for symbiosome and arbuscule formation.



Ivanov S et al. PNAS 2012;109:8316-8321

- Legumes spend approximately 4-16% of their photosynthetic activity on each of the rhizobial and AMF symbioses (Kaschuk et al. 2010).

- AMF associations vary from mutualistic to parasitic depending on AMF species, host crop and environmental conditions (Jones and Smith 2004; Schwartz et al. 2006).

- Positive effects are only achieved if the benefits outweigh the costs of maintaining association (Kaschuk et al. 2010).

- Recent biotech advances have led to the introduction of AMF inoculants as ‘biofertilizers’.

- Pulse crops have typically high P requirement, alternative P management strategy.

- Additionally the possibility exists for the AMF inoculant to influence the rhizobial association because AMF and rhizobia share similar signaling pathways.

OBJECTIVES

- To investigate the impact of AMF and rhizobial inoculation on the efficacy of the tripartite symbioses under field conditions.

- To assess the impact of the tripartite symbioses on the success of each participant in the symbiosis (i.e., AMF colonization, rhizobial nodulation and pulse crop growth parameters).

MATERIALS AND METHODS

- Field trials with lentil (*Lens culinaris* cv. Clearfield Impress) and field pea (*Pisum sativum* L. cv. CDC Meadow) were conducted in 2012 at Kelvington and Stewart Valley, SK.
- Sites were chosen on the basis of low levels of available P (4 kg ha^{-1} and 10 kg ha^{-1} to 30 cm, respectively).
- Completely randomized block design with four replicates.

TREATMENT DESIGN

Phosphorous Applied	Rate of MYKE® PRO GR	Rate of Nodulator®	Rate of JumpStart®
-	-	+	-
-	7.5 kg ha^{-1}	+	-
-	15 kg ha^{-1}	+	-
-	-	+	+
16.8 kg ha^{-1}	-	+	-
16.8 kg ha^{-1}	7.5 kg ha^{-1}	+	-
16.8 kg ha^{-1}	15 kg ha^{-1}	+	-
16.8 kg ha^{-1}	-	+	+

Figure 3(a). Field pea at Kelvington 2 wks after emergence

Figure 3(b). Field pea at Kelvington at mid season.

- Sampling : Midseason (early pod fill) and final harvest.
- Shoot biomass, nodulation and AMF colonization, seed yield and nutrient uptake.
- Biological nitrogen fixation is to be determined using the ^{15}N natural abundance method.
- Results from the 2012 field season (field pea and lentils) from Kelvington and Stewart Valley are presented.

Fig 4(a). Mid-season biomass sampling at Kelvington.

Fig 4(b). Mid-season root sampling at Kelvington.

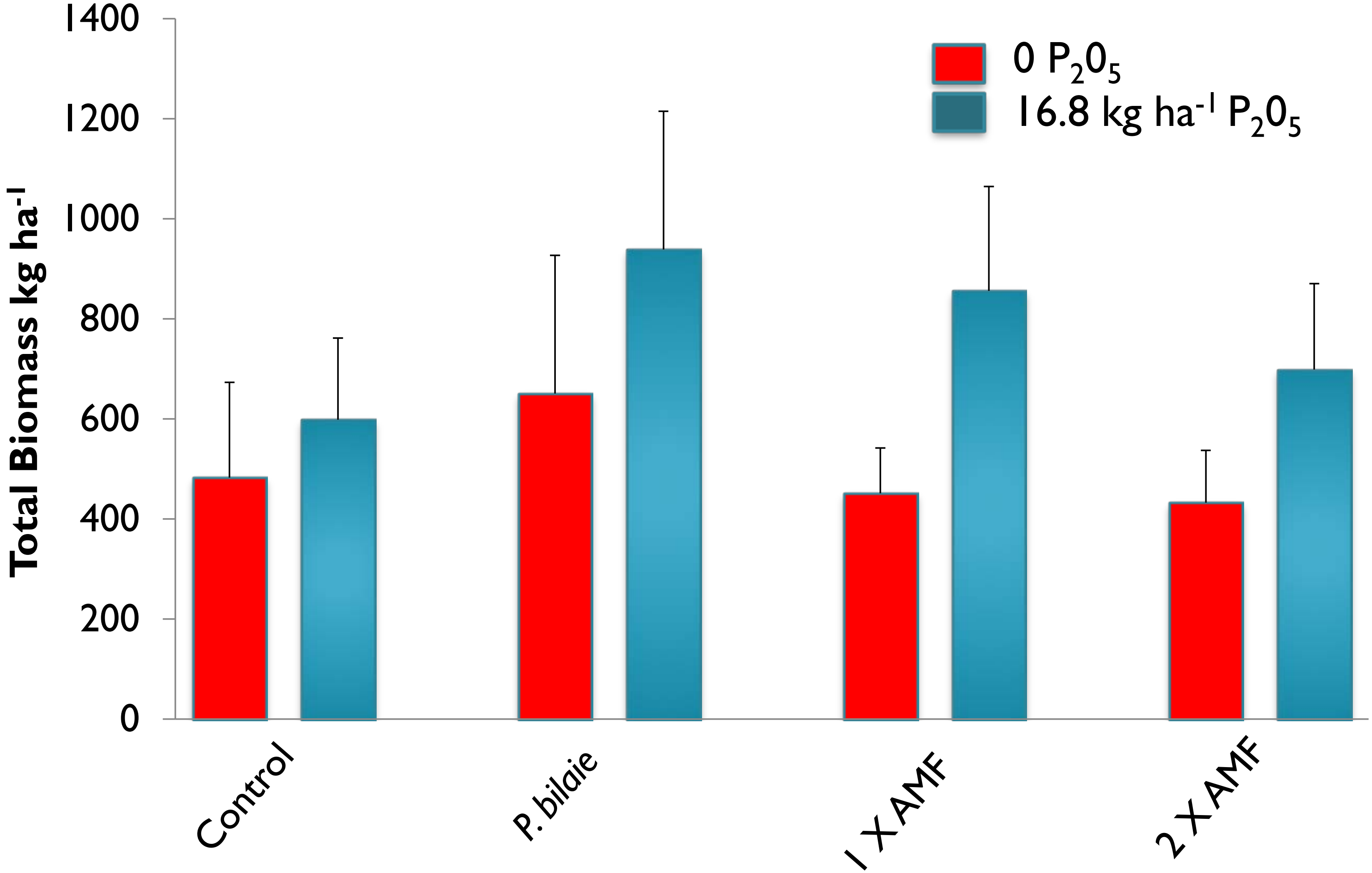


Figure 5. Impact of inoculation on midseason biomass of field pea at Kelvington.

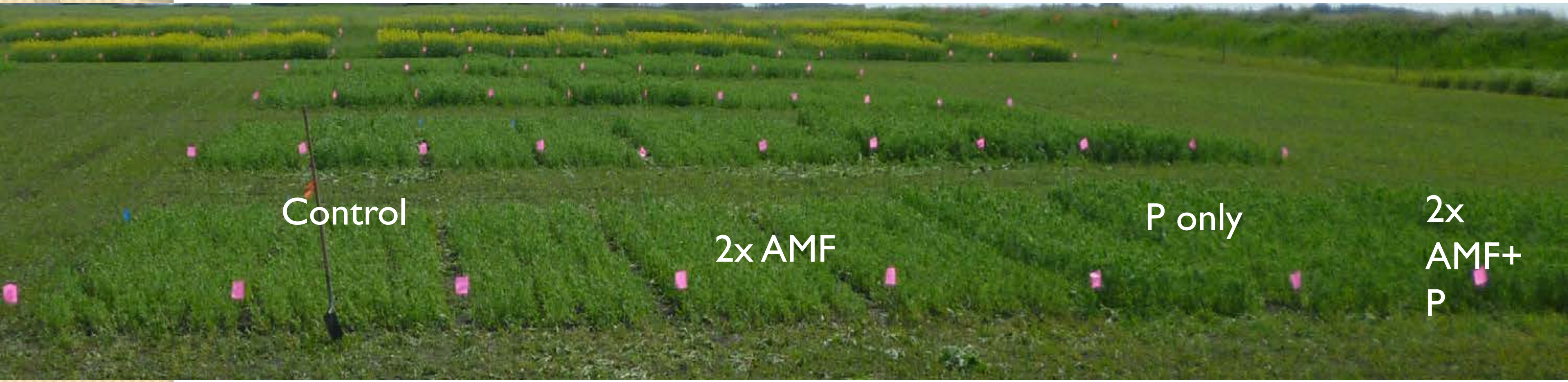


Fig 6. Pea biomass response to fertilizer P and inoculation at midseason at Kelvington

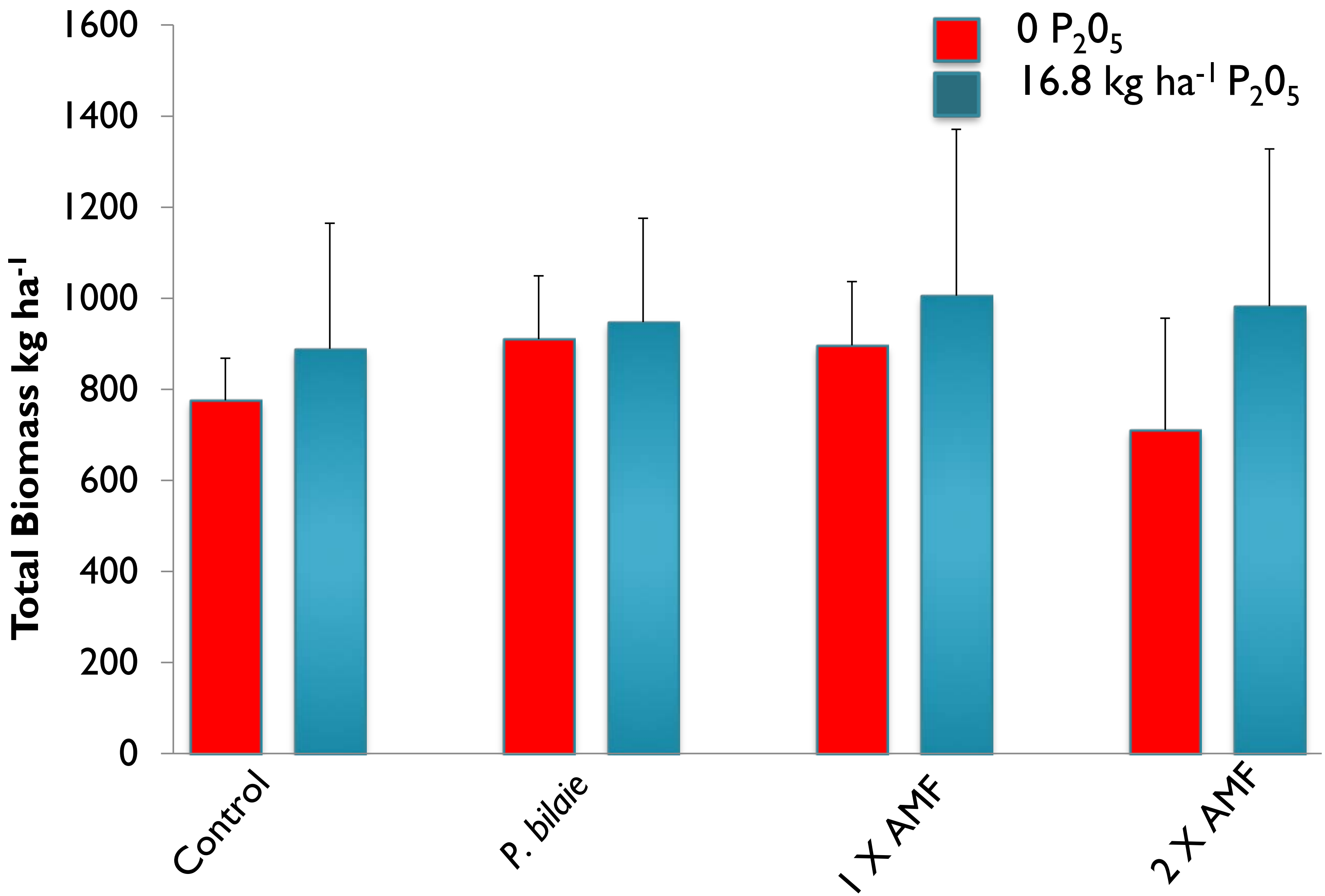


Figure 7. Impact of inoculation on midseason biomass of lentils at Stewart Valley.

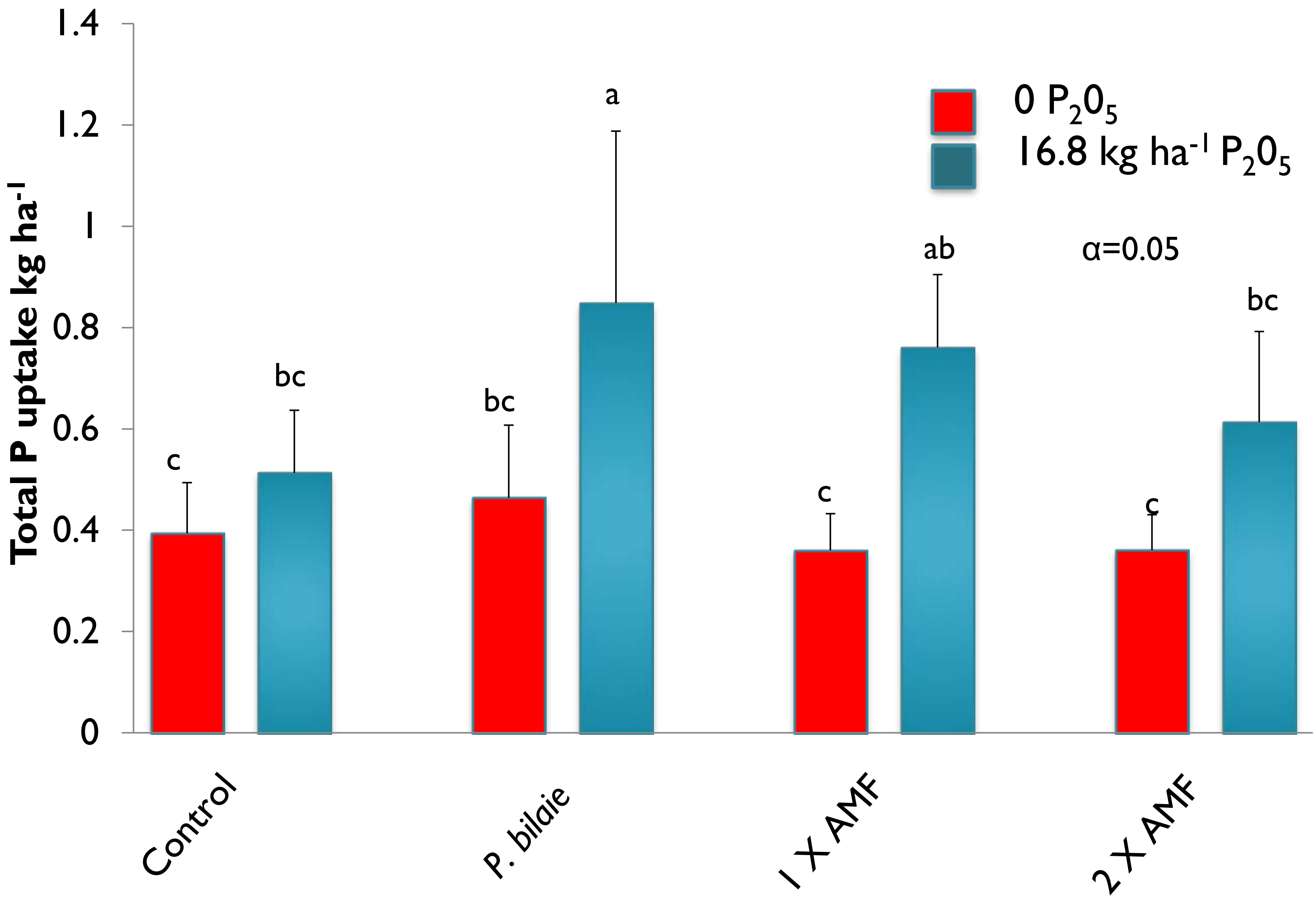


Figure 8. Impact of inoculation on midseason P uptake of field pea at Kelvington.

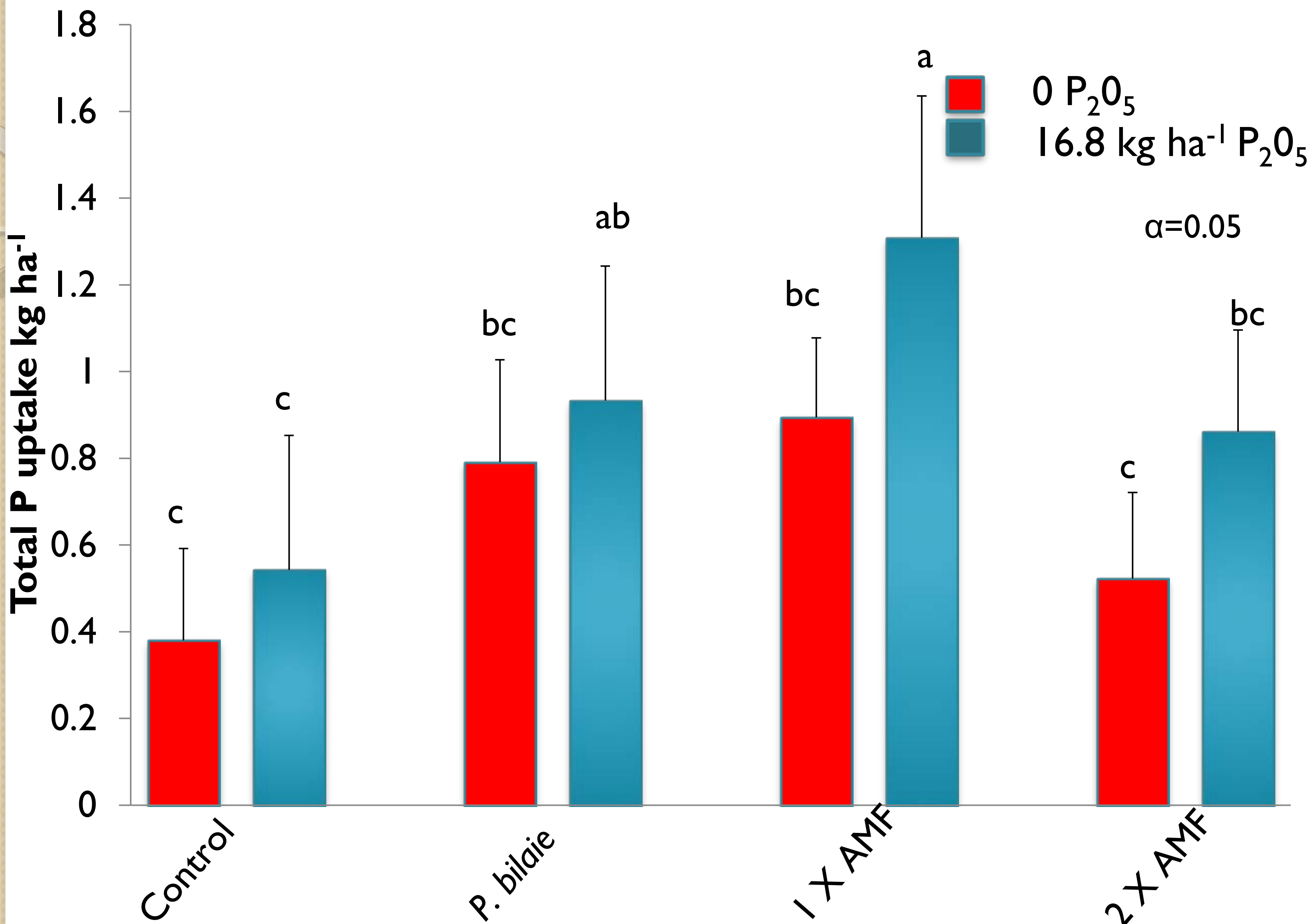


Figure 9. Impact of inoculation on midseason P uptake of Lentils at Stewart Valley.

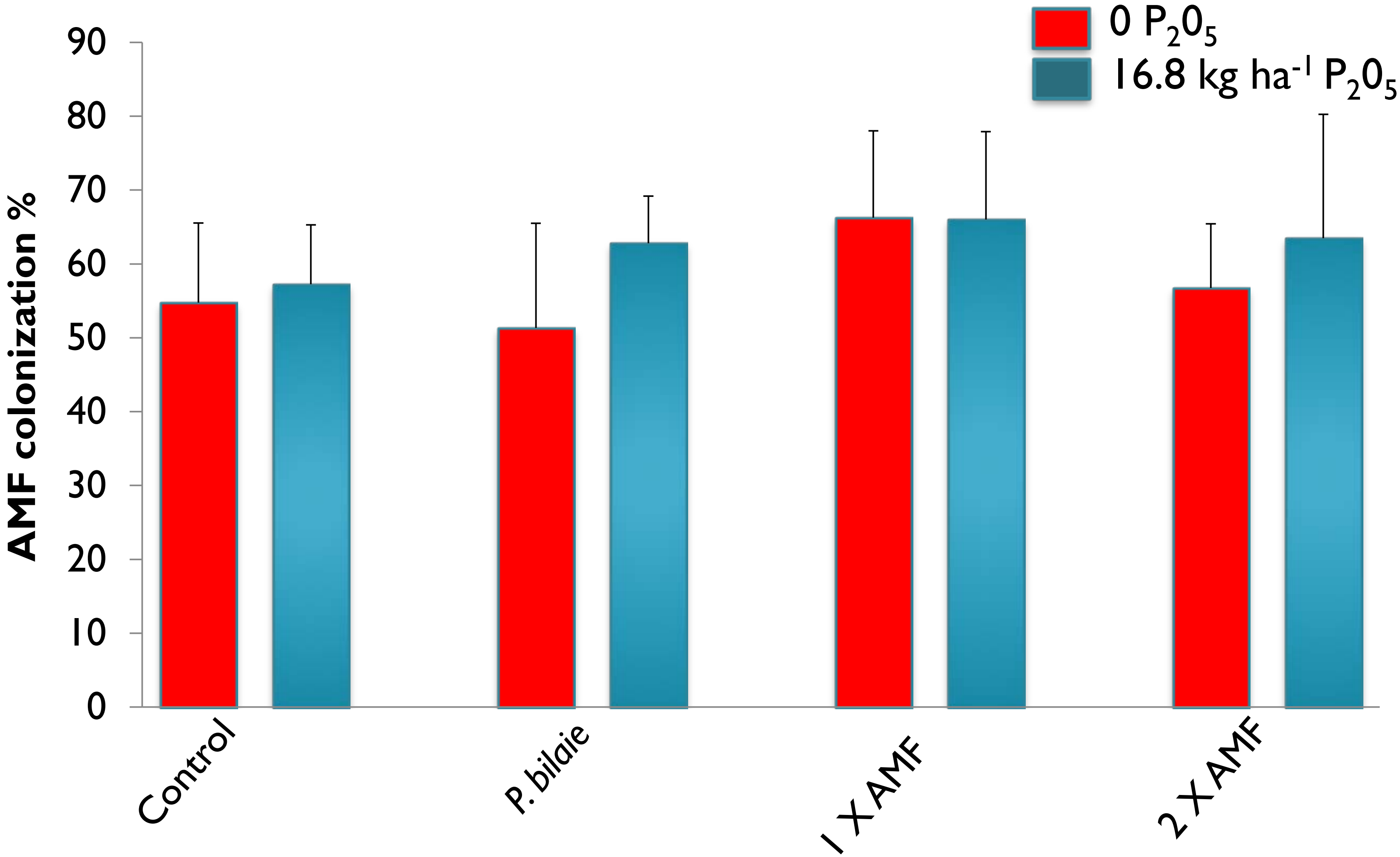


Figure 10. Impact of inoculation on midseason AMF colonization on field pea at Kelvington.

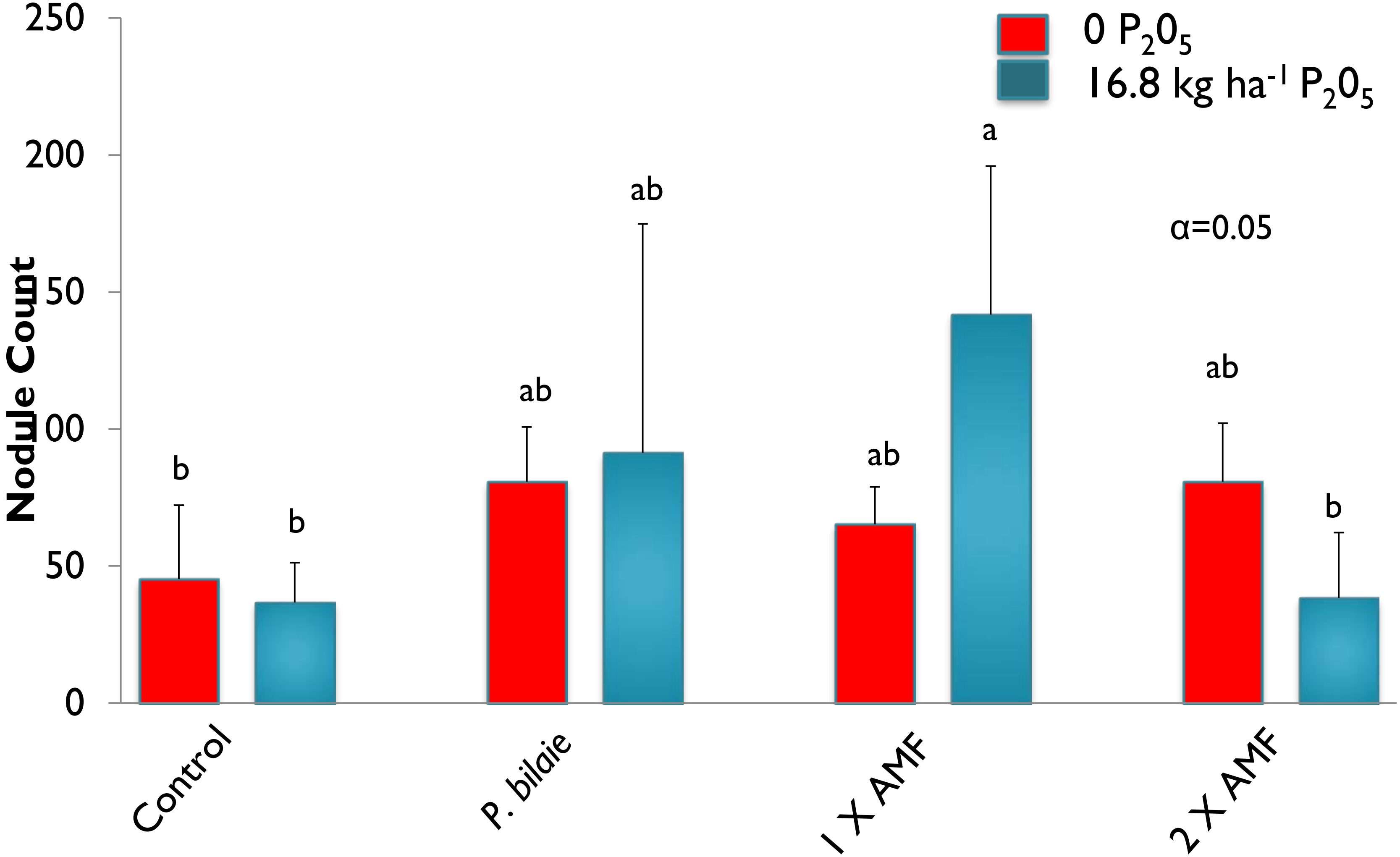


Figure 11. Impact of inoculation on midseason nodulation on field pea at Kelvington.

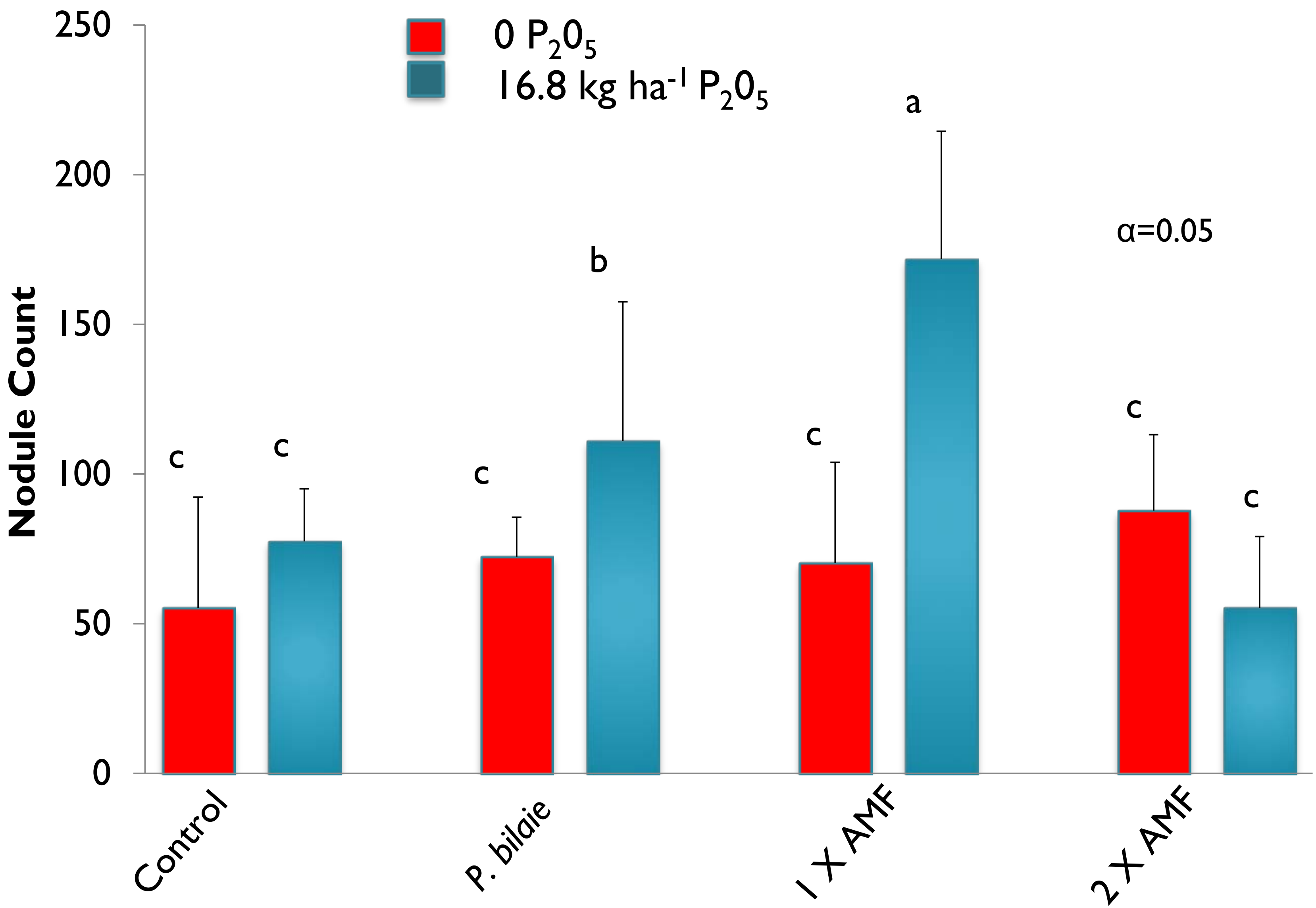


Figure 11. Impact of inoculation on midseason nodulation on Lentils at Stewart Valley.

DISCUSSION

- P fertilizer enhanced mid season biomass at Kelvington and Stewart Valley.
- Magnitude of the biomass response to P was enhanced in the presence of both AMF and *P. bilaiae*.
- No additional benefit of doubling the AMF application rate. (New recommended rate is 5 kg ha^{-1})
- Similar results were obtained for P uptake.

- AMF colonization (%) was unaffected by any treatment, including AMF inoculants. In 2013 field trials, % root length colonized will be measured to take into account potential "dilution" in case of larger root biomass.

- Nodulation was highly variable; however, trends suggest that nodulation was unaffected by P fertilizer alone, but was enhanced when applied with AMF.

SUMMARY

- Preliminary data suggest AMF inoculation may influence nodulation of both field pea and lentils.

- Doubling the application rate of AMF did not affect responses compared to the recommended rate.

- Biomass responses to AMF inoculation may reflect improved uptake of fertilizer P sources, in particular.

NEXT STEPS

- Sample processing is ongoing.
- Experiments will be repeated in 2013.
- Growth chamber experiments and *in vitro* experiments will be carried out to further study the mechanism controlling the interactions.

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THANK YOU !!