

Introduction

- Legume crops are unique in their capacity to obtain substantial amounts of nitrogen by associating with rhizobia (1). Biological nitrogen fixation (BNF) by rhizobia offers an attractive alternative to chemical-nitrogen fertilization because it comes without fossil fuel costs or polluting byproducts. However, efficient BNF is difficult to attain in practice. The main challenge is that legumes encounter a diversity of rhizobial strains that vary in the degree of compatibility and benefits they provide for the host (2). To maximize fitness, legumes must exhibit efficient symbiosis traits, defined as characteristics that allow plants to invest in symbionts that provide benefits to the host and defend against ineffective or incompatible strains (3).
- Modern agricultural practices can degrade plant-microbial symbioses (4). Breeding practices select aboveground traits, while neglecting belowground plant features, and evolutionary tradeoffs between these traits can disrupt host control over microbiota (5). Moreover, the small effective population sizes of domesticated plants, the increased inbreeding, the loss of genetic diversity, and relaxed selection for traits that are not critical to agriculture (6), can each lead to the degradation of host mechanisms that regulate microbiota (4).
- We investigated how domestication has influenced symbiosis traits in cowpeas. Using eight wild cowpea genotypes and twelve early-cultivated landrace genotypes, we quantified mean trait values and genetic variance associated with clonal and mixed strain inoculation of *Bradyrhizobium diazoefficiens* as well as whole soil inoculation.

Objectives

- Compare genome-wide genetic diversity of wild and domesticated cowpeas.
- Investigate segregating variation in symbiosis traits and examine whether they became degraded during domestication
- Measure heritability of symbiosis traits and their potential to be selected upon in agronomic settings

Methods

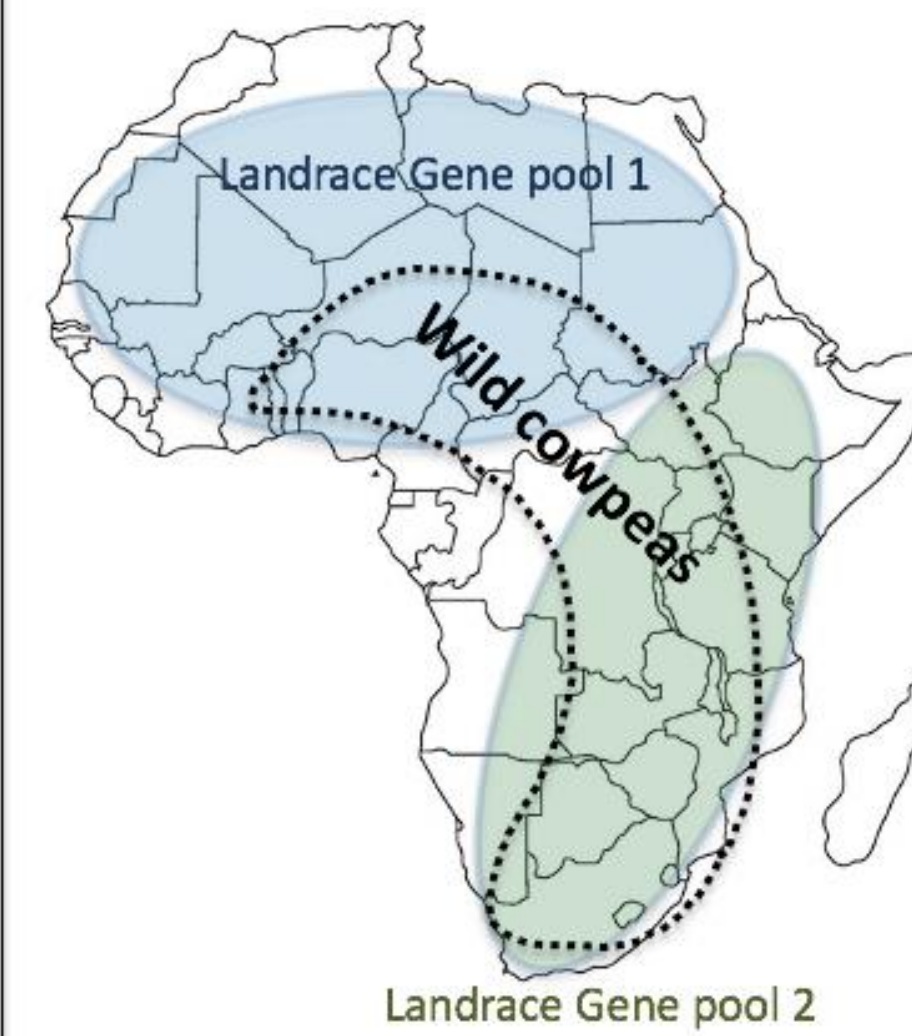
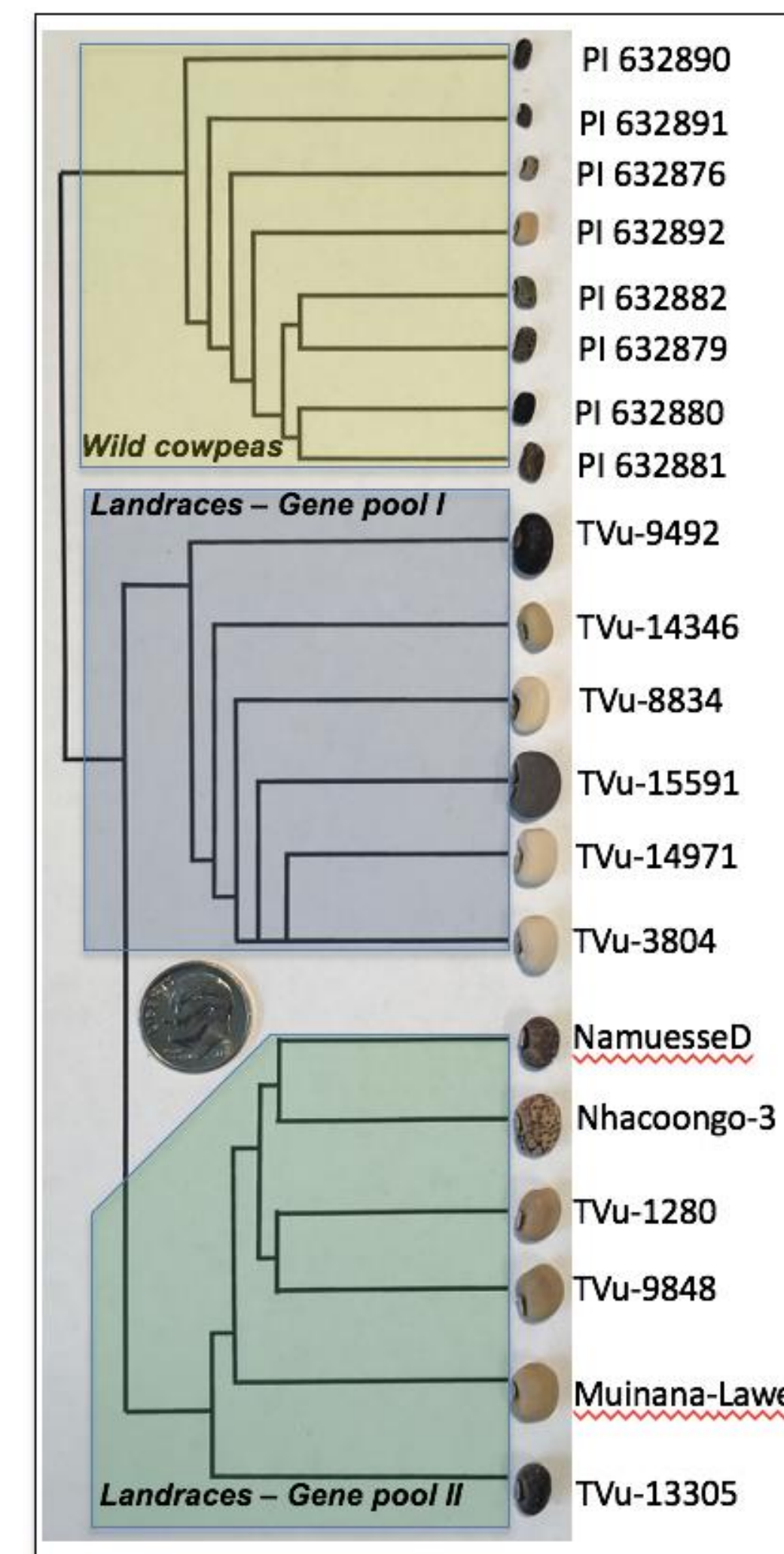


Fig. 1. Cowpea genetic diversity: Wild cowpeas form a basal lineage that grow across sub-Saharan Africa, have small seeds, shattering pods, and flower under short days. Cultivated cowpeas include two sister groups of landraces originating in North & South Africa, and have large seeds non-shattering pods, and can flower in short or long days. Phylogenetic tree generated from data in Huynh et al., 2013.

Results

- Domesticated lineages experienced a modest reduction in heterozygosity (H_o) and gene diversity (H_s) relative to the wild lineage (Fig. 2).
- Domesticated plants formed more nodules and varied more between treatments (Fig. 3)
- Cultivated lineages invested a higher proportion of biomass into nodules compared to wild cowpeas.
- Host Growth response for each inoculation treatment varied among lineages (Fig. 3)
- Moderate levels of heritability were observed for number of nodules and host growth response.

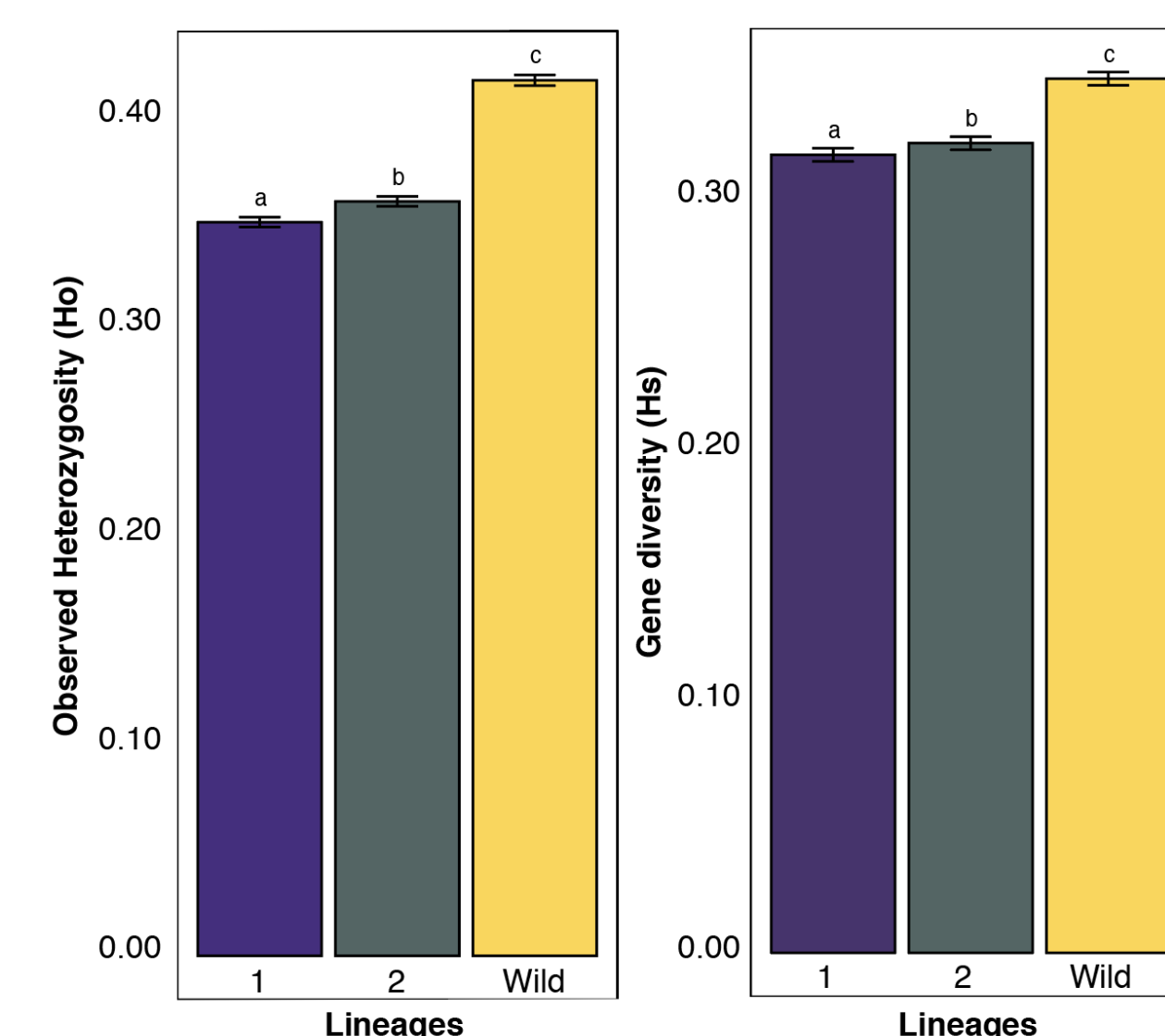


Figure 2. Genome-wide diversity of domesticated and wild cowpeas. Estimates of expected heterozygosity and gene diversity within each lineage are shown.

- We inoculated our twenty cowpea genotypes (Fig.1) with *Bradyrhizobium diazoefficiens* type strain USDA 110-ARS (Fix+), a nonfixing mutant derived from it USDA110-LI (Fix-), a coinoculation of equal proportions of both strains, and a microbial community made of soil rinsates from the field. We measured the total biomass, Host growth response (%), Investment into symbiosis, nodule number, mean nodule biomass and Ndfa(%) as well as the genome wide diversity and the additive genetic variation of the traits for all our cowpea genotypes.

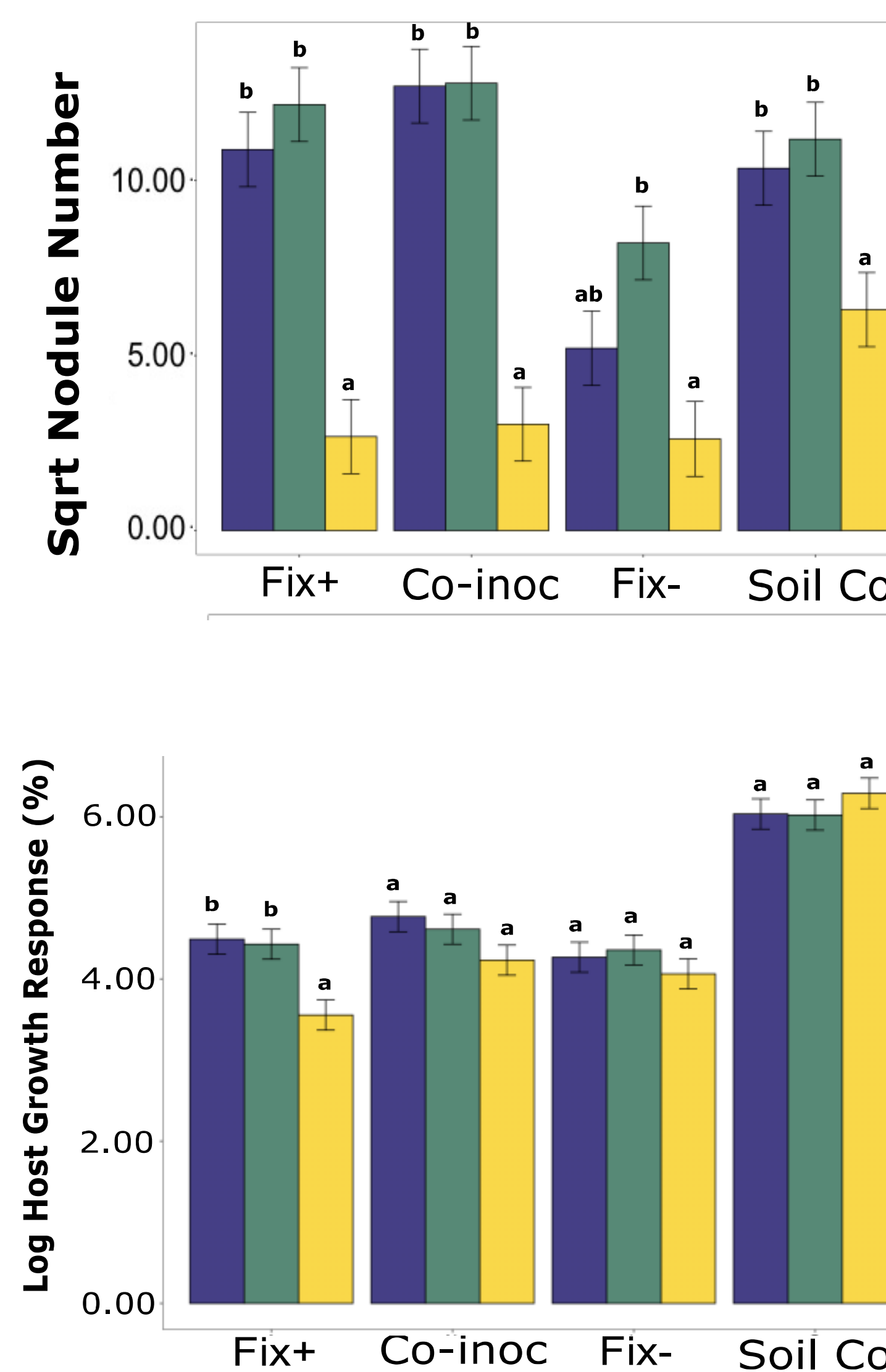


Figure 3. Sqrt nodule counts (least-squared means) and Log Host Growth Response (%) of cowpea lines under the four inoculation treatments. The blue bars represent plants that belong to landrace gene pool 1, green bars represent landrace gene pool 2 and yellow bars the wild gene pool. Error bars show the standard error of the means for each group.

Discussion

- We found little evidence for degradation of symbiosis associated with domestication. There was a modest decline in genetic diversity during the early stages of cowpea domestication (Fig. 2). Domesticated cowpeas experienced a less severe bottleneck than other crops, additionally, the low nutrient conditions in Africa where these landraces were developed likely favored plant genotypes that benefit from symbiosis.
- Reduction in genome-wide genetic variation did not indicate loss of additive genetic variance of all symbiosis traits. For number of nodules produced, we found that domesticated landraces had higher genetic variance compared to the wild lineage.
- Among the cowpeas studied here, Genepool-2 contains the best potential for further breeding given its high heritability for number of nodules and host growth response. Further screening for these traits could allow growers to select accessions that can improve plant growth in the presence of compatible rhizobia.
- Our work was focused on examining the early steps of domestication, and thus the conclusions that we can draw might not apply to modern cowpea cultivars. Moreover, conditions of propagation exposure might be important in the disruption of symbiosis traits. Thus, it could be that degradation of symbiosis traits occurs more commonly with intense artificial selection during the latter stages of domestication.

Implications

- Breeders have largely neglected symbiosis traits, but artificial selection for improved plant responses to microbiota could increase plant performance and sustainability.
- Our results highlight potential breeding strategies to include symbiosis traits as tools to potentially use them to sustainably increase crop productivity and reduce the impact of nitrogen fertilizers.

Literature Cited

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