VARIATIONS IN THE POPULATION OF ARBUSCULAR MYCORRHIZAL FUNGI IN AN AGRICULTURAL SOIL IRRIGATED WITH TEQUILA VINASSES

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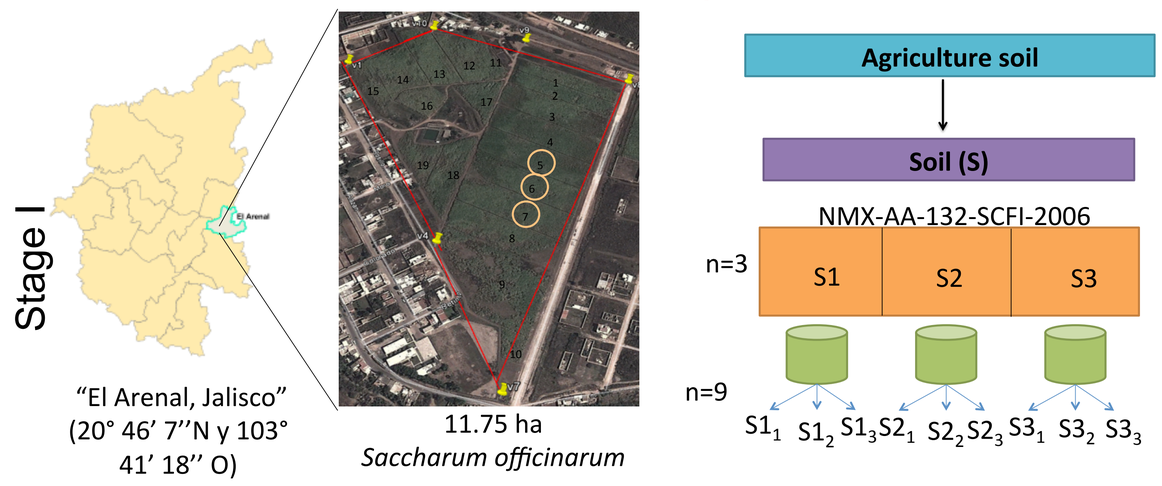
1. Introduction. Tequila production generates a waste called vinasse. It's estimated that 1763 million liters of tequila vinasses were generated in 2014. Because of the high concentration of organic matter, it is considered that the application of vinasses can be beneficial to soil, which is a common practice in many tequila factories. However, there is not information about the effects on populations of beneficial organisms as arbuscular mycorrhizal fungi (AMF). There isn't any registers that indicate if there are AMF in soils irrigated with tequila vinasses. This fungi (AMF) are symbiotic organisms which establish mutual symbiosis with most higher plants, providing a link between the soil and plant roots (1). The objective of this study was to evaluate the effect of the addition of different concentrations of tequila vinasses in AMF spores.

2. Methods.

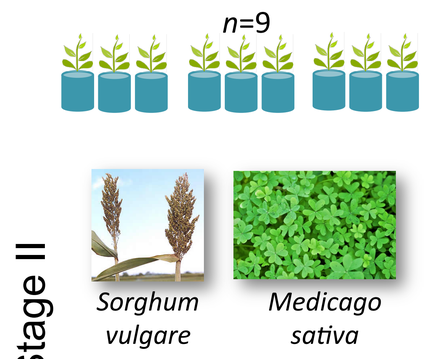
Characterization of soil and tequila vinasse



Soil: pH, water holding capacity (WHC), electrolytic conductivity (EC), organic and inorganic carbon, total nitrogen (TN), available phosphorus (AP) (2) (3).

Vinasses: pH, EC, volatile suspended solids, setteable solids, AP, TN, COD, BOD, heavy metals (4).

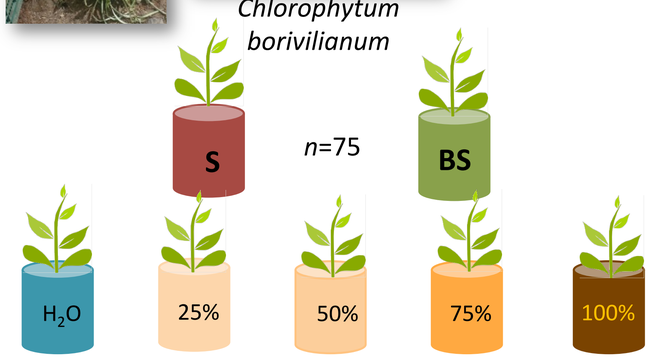
AMF propagation



Isolation, quantification and identification of AMF spores.

Evaluation of different concentrations of tequila vinasses

Addition of **5 g** of propagated soil as **initial inocolum** with a density of **270 spores** of AMF.



Each month for 4 months

Isolation, quantification and identification of AMF spores.	pH, EC, available phosphorus, NO_3^-, NO_2^-, NH_4, and ions (Na, K, Ca, Mg).

The AMF spores were isolated from S using wet sieving method, decanting and centrifugation in water and 50% saccharose (w/v), according to Brundrett et al. (5). The spores were analyzed under a stereomicroscope and mounted in preparations of PVLG and PVLG+Melzer (1:1 v/v) for their identification in an optical microscope followed the descriptions of the International Culture Collection of Vesicular Arbuscular Mycorrhizal Fungi (INVAM) (6).

3. Results and discussion. The sandy loam soil (S) was classified as Phaeozem (7) with neutral pH (6.5 ± 0.2) and high organic matter (93.7 ± 1.5 g kg^{-1}). The S was saline with high CEC (27.3 ± 5.9 Cmol kg^{-1}). The inorganic N was low, but the total N content was high (971 ± 147 mg kg^{-1}). The AP (0.1 ± 0.03 mg kg^{-1}) in the soil was low compared to standard regulations (8). The amount of heavy metals was low and did not exceed international standards. The tequila vinasse had an acid pH (3.5 ± 0.1), a high COD (57 800 ± 1 900 mg L^{-1}) and BOD (25 400 ± 1 700 mg L^{-1}) content, high settleable solids (120 ± 5.0 mg kg^{-1}), and total volatile solids (37.3 ± 3.0 mg kg^{-1}) and high total nitrogen 4 200 ± 100 mg L^{-1}).

The concentrations of heavy metals (< 1 ± 0.0 mg L^{-1} of As, Cu, Cr, Hg, Pb and Ni, < 0.2 ± 0.0 mg L^{-1} of Cd, < 2.3 ± 0.0 mg L^{-1} of Zn and 24 ± 0.9 mg L^{-1} of Fe) were below maximum limits as stipulated by USEPA regulations (9).

After the propagation stage, the spore abundance mean of AMF were 236 ± 79. Nine species of AMF were morphological identified. The most abundant species were *Acaulospora mellea*, *Funneliformis mossea* and *A. scrobiculata (*Figure 1).

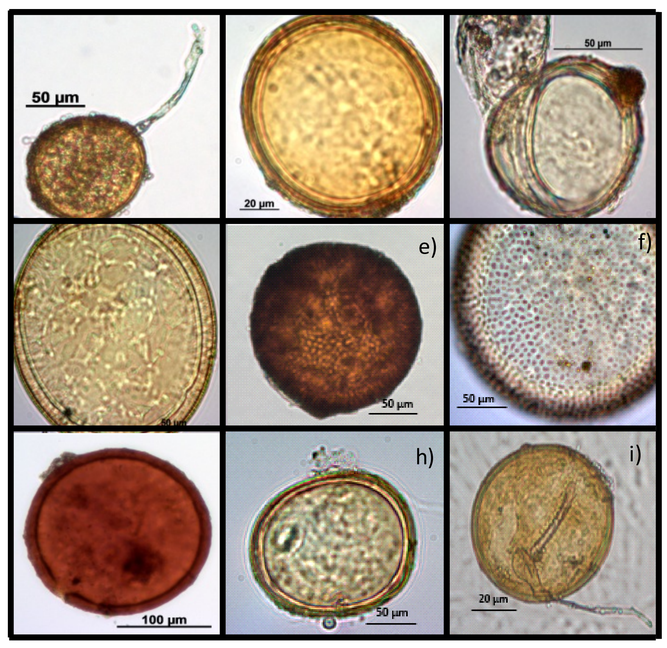


Fig 1. AMF propagated. a) *Funneliformis mosseae*, b) *Acaulospora mellea*, c) *Paraglomus occultum*, d*) A. spinosa*, e) *A. foveata*, f) *A. scrobiculata*, g) *F. geosporum*, h) *Gigaspora margarita* and i) *A. delicata*.

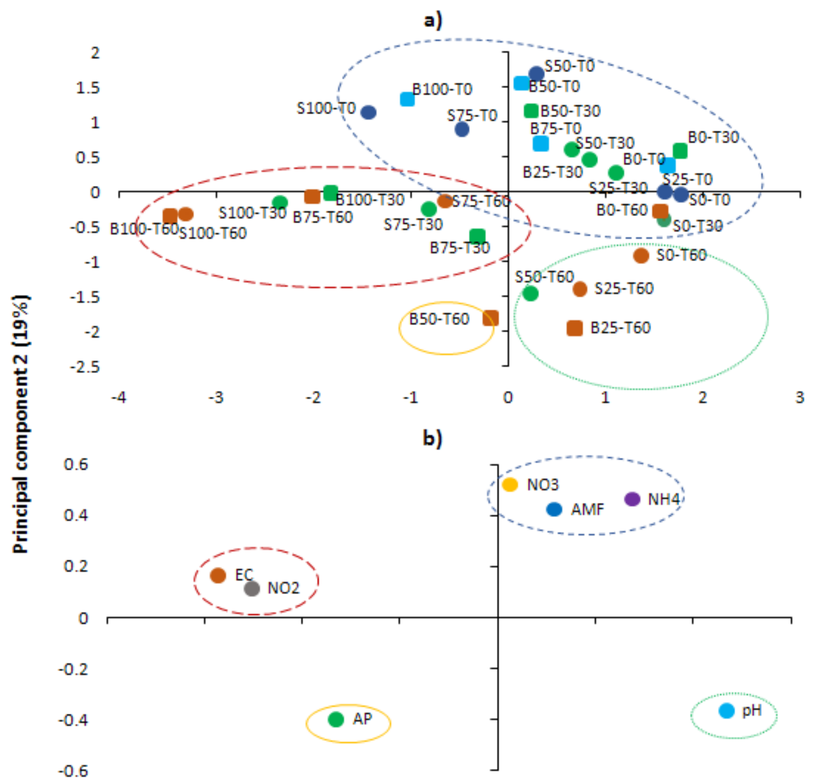


Fig 2. Principal component analysis (PCA) for **a)** scores and **b)** loadings

The principal component 1 (PC1) and component 2 (PC2) explain 54% of variability of data (Figure 2). Component PC1 in positive side separated to pH and negative side to EC, NO_2^-, while component PC2 separated to variables of NO_3^- AMF and NH_4^+ in positive side and negative side to AP. Also, PC1 group the higher concentrations of vinasses (75% and 100%) and the longer time (T30 and T60) with the highest values of EC and NO_2^- in the negative quadrant. The lowest pH values were found in 0, 25 and 50% of vinasse with longer time (T60) in the positive quadrant of PC1. The PC2 correlated the AP variable in the negative quadrant with intermediate concentrations (50%) with longer time (T60). While, high values of NO_3^- AMF and NH_4^+ were correlated in the positive quadrant with the lowest concentrations of vinasses (0, 25 and 50%) in time T0 and T30. This indicated that with higher amount of vinasses decreases the amount of spore and EC increases.

4. Conclusions. Nine species of AMF spores were propagated in a trap crop: *Funneliformis moseeae, Acaulospora mellea, Paraglomus occultum, A. scrobiculata, F. geosporum, A. foveata, A. spinosa, A. delicata and Gigaspora margarita.* High concentrations of tequila vinasses decreases the amount of AMF spores and increases the EC of soil.

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6. Reference.

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