Physiological and pathological studies on vesicular abrbscular mycorrhizal fungi grown under lab condition

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1.Three VAM isolates namely VAM-O, VAM-B and VAM-S were isolated from roots of field grown plants (onion and broad bean), aerial roots of Swiss cheese ornamental plants, respectively. Another isolate (VAM-M) was isolated from roots of maize seedlings emerged from surface sterilized seeds planted in sterilized soil. The in vitro growth of isolated VAM fungi (on the modified MS-agar medium) characterized by white-grayish dichotomously branched mycelium with aseptate, hyaline and vacuolated hyphae and varied in thickness. Oil-like DROPlets were observed repeatedly particularly in the widest hyphae.2.The isolated VAM fungi formed terminal sporangia (varied in diameter) contained rounded hyaline 1-celled sporangiospores in addition to apical, intercalary chlamydospores formed singly and/or in chains. Zygospore formation was noticed only in VAM-0 isolate.3.Inoculation with VAM fungi significantly improved most of the examined growth criteria of maize seedlings comparing with the control (non-inoculated). Degree of improvement was depending on source of VAM isolate, kind of inoculum used, inoculum level (i.e. 1-6g and 0.5-0.3mj/kg soil for BS-cultures and sporangiospore suspension, respectively) and the target growth parameter (shoot and root lengths, stem diameter, fresh and dry weights of shoots and roots/plant). Compared with the uninoculated control, VAM-0 shows retarding effects on most growth parameter tested when used at the highest inoculum level.4.Typical VA-mycorrhizal infection structures (arbuscules, vesicles and aseptate hyphae grow mainly between and rarelyacross cells in root cortex were clearly seen in mycorhizal (inoculated) and non-mycorhizal roots of maize seedlings. Roots of inoculated seedlings showed no any detectable changes in their external morphology when compared with those of control treatment.5. Among detected DNA bands, the band number 3 was found inall isolated VAM fungi either in cultures or inside the mycorrhizal roots. Specific DNA bands were detected in mycelia and inoculated roots such as bands number 5, 10, 11 and 12 in VAM-M isolate and bands number 11 & 12 in VAM-B isolate while, bands number 10 & 12 were specific to roots inoculated with VAM-0 isolate or Glomus spp. In all cases, these specific bands were missed from the non-mycorhizal roots (control). Similarity between the 4 VAM fungal isolates (in cultures) was ranged between 87.27-100.00%. As for mycorrhizal roots (inoculated with any VAM isolate including Gloms sp.) and non-mycorhizal roots, percentage similarity was ranged between 77.39-85.03%.6.The isolated VAM fungi could grow and sporulate on different nutritive media. Bushnell’s medium was the superior while, Czapek’s medium was the inferior.7.Growth and spore formation of isolated VAM fungi, on Bushnell’s medium, were sharply increased as temperature increased from 5°C to 28 or 31°C, depended on VAM isolate, then appreciably decreased by raising temperature up to 33°C. However, these parameters were significantly decreased as R.14.% increased from 14% to 100% comparing with the control (un-controlledhumidity conditions).8.The yeast extract, beef extract and aspragine were the best N-sources for growth and sporulation of the isolated VAM fungi. While, no growth or sporulation was detected on media containing urea or NaNO2.9.As for carbon sources, growth and sporulation of all isolates were completely stopped on citric acid. The linear growth of VAM-0 and growth dry weight of all VAM isolates, however, was stopped also on media containing inositol. In general, glucose

produces the highest growth dry weight followed by galactose, fructose, starch, maltose, arabinose and sucrose, respectively. whereas, sucrose was the best for spore formation followed by glucose, galactose, fructose, starch, maltose and arabinose, respectively.10.Growth and spore formation of VAM fungal isolates was significantly increased as pH increased from pH 5.0 to pH 7.0 then gradually decreased by elevating pH value up to 7.6, 8.0, 8.6 or 9.0, respectively. In general, the higher p1-1 values (pH 7.0-9.0) seemed more effective in this respect than the lower ones (pH 5.0-6.6).11.The linear growth of isolated VAM fungi was not affected significantly by different light waves tested. whereas, the hyaline light was the best for growth dry weight and spore* formation followed by the green, blue, yellow, red light waves and dark, respectively. In general, the hyaline light was the best for isolates VAM-0, VAM-M and VAM-S whereas green light was the best for VAM-B.12.Based on % survival, as accurate expression for the damping-off disease development, Rhizoctmia solani was the most virulent191 Summaryfollowed by Cephalo.sporium maydis and FusariumAll pathogenic fungi significantly reduced shoot and root lengths, stem diameter, number of leaves/plant, fresh and dry weights/plant, the total photosynthetic pigments (chlorophyll a & b, and carotenoids) in leaves compared with the control.13.All VAM isolates, particularly in the second sowing, significantly increased ()/0 survival in the pathogen-infested soil compared with the control. Inoculation with VAM-M was the superior in this respect followed by VAM-O, VAM-S and VAM-B, respectively.14.All VAM isolates significantly increased the shoot length compared with the control. Using VAM-B either in the pathogen-free soil or in soils infested with C. maydis, F. monioliforme or R. solani caused the highest increase in the shoot length compared with their particular controls particularly in the second sowing.15.All VAM isolates increased root length in both sowings compared with the controls. Using VAM-B in the first sowing and VAM-S in the second one against C. maydis and F. monioliforme and VAM-B against R. solani in both sowings caused the highest increase in the root length compared with the pathogen-infested controls.16.All VAM isolates increased stem diameter in both sowings compared with the controls. Using VAM-B against any of the pathogens tested (C. maydis, F. monioliforme and R. solani) caused the highest stein diameter in both sowings compared with pathogen-infested soils.192 Summary17.All VAM isolates tested significantly increased the number of leaves/plant compared with the control treatments. Using VAM-B against F. monioliforme or R. solani (in both sowings) and VAM-B and VAM-S in the first and second sowings, respectively against C. maydis produced the highest number of leaves/plant.18.All VAM isolates significantly increased both total fresh and dry weights/plant compared with the control treatments. Using VAM-B against F. monioliforme or R. solani particularly in the first sowing recorded the highest values of both determined criteria. VAM-M, however, recorded the lowest increase in both criteria in both sowings compared with the control treatments.19.All VAM isolates tested caused considerable increase in the photosynthetic pigments (chlorophyll and carotenoids) compared with the controls. In general, using VAM-0 against F. monioliforme and VAM-S against C. maydis (in both sowings) and VAM-B and VAM-S against R. solani in the first and second sowings, respectively produced the highest values of the total leaf pigments in comparison with their particular control.20.All pathogens and VAM isolates tested caused conspicuous increase in the pH value in the rhizosphere region compared with their controls. As for pathogens, F. monioliforme induced the highest increase in the rhizosphere-pli (in both sowings) followed by R. solani and C. maydis, respectively. Concerning VAM isolates, VAM-S was the most effective for increasing the rhizosphere-pH (in both sowings) whereas VAM-M was the least effective in this respect.21.Unlike rhizosphere-pH, all pathogens tested caused appreciable decrease in the soil-pH compared with the control. The highest decrease, however, was induced by I?. solani followed by F. moniolilin-me and C. maydis particularly in the first sowing. On the contrary, all VAM isolates increased the soil-pfl in comparison with the control. The highest increase, however, was induced by VAM-M in both sowings.22.Inoculation with different levels of VAM fungi significantly increased seed germination compared the unfertilized, P or N fertilized plots particularly in 2002 season. N fertilizer + VAM inoculation produced more significant increase in seed germination than VAM inoculation. N fertilizer + inoculation with: VAM-S at the low level (10Kg/fed.) or VAM-O, VAM-B and VAM-M at the high level (20Kg/fed.) recorded the highest increase in seed germination. In general, VAM-0 was superior for improving seed germination while, VAM-M was the inferior one during bothseasons.23 VAM

inoculation at the high level only in 2002 season and allinoculation levels (high, middle and low) in 2003 season significantly decreased wilt disease incidence compared with P fertilizer. In 2003 season, N fertilizer + inoculation with VAM-B at the middle (15kg/fed.) or high level (20 Kg/fed.) were the most effective for suppressing wilt disease compared with P or N fertilizers each alone. On the contrary, N fertilizer + inoculation with VAM-M (all levels) significantly increased the wilt24.Inoculation with VAM-M, in 2002 season, caused the highest decrease in the common smut disease incidence followed by VAM-0, VAM-S and VAM-B, respectively. Inoculation with VAM (different levels) alone was more effective in this respect than P fertilizer in 2002 season and N fertilizer alone or combined with VAM inoculum in both seasons.25.Inoculation with VAM (different levels) significantly decreased percentage of stunt disease incidence compared with P fertilizer in both 2002 and 2003 seasons. Also, N fertilizer + inoculation with VAM (different levels) was more effective in this respect than N fertilizer alone.26.Inoculation with VAM significantly increased plant height more than application of P fertilizer. Plant height was proportionally increased as VAM Inoculation level increased. On the contrary, N fertilizer 1- VAM Inoculation significantly increased plant height in 2002 season and decreased it in 2003 season compared with application of N fertilizer alone.27.VAM-0 was the most effective while VAM-M was the least effective one for increasing total fresh and dry weights/plant. Inoculation with VAM alone or combined with N fertilization significantly increased both parameters compared with P and N fertilizers, respectively. The increase in both parameters was proportionally parallel to the increase in inoculation level particularly in 2003 season. In both seasons, N fertilizer I VAM inoculation particularly at the middle or the high levels was significantly better than N fertilizer alone. Using N fertilizer -±-inoculation with VAM-0 (at high level) or VAM-S (at low level)produced the highest increase in both parameters without significant differences in between.28.In 2002 season, VAM-0 and VAM-M were significantly better for increasing the fresh weight of roots (FWR)/plant than VAM-B and VAM-S. Inoculation with VAM particularly at the middle level in 2002 and high level in 2003 seasons significantly increased the FWR/plant compared with P fertilizer. Moreover, N fertilizer + VAM inoculation at any level was significantly better in this respect than N fertilizer alone without significant variations between the 3 used VAM levels particularly in 2002 season.29.In 2003 season, VAM-0 and VAM-S significantly increased dry weight of roots (DWR)/plant more than VAM-B and VAM-M. Inoculation with VAM (at the middle and high levels) was more effective in this respect compared with P fertilizer. The DWR/plant was not affected significantly by using N fertilization + VAM inoculation compared with N fertilizer alone.30.The isolate VAM-0 recorded the highest significant increases in ear weight in 2003 season followed by VAM-S and VAM-I3 and VAM-M, respectively. The ear weight was significantly increased by applying VAM inoculation alone (compared with P fertilizer) or combined with N fertilization (compared with N fertilizer alone). This trend was holds true in both seasons. The ear weight was significantly increased as VAM inoculation level increased.31.The isolate VAM-S recorded the highest significant increases in the grain yield/plant in 2003 season followed by VAM-0 and VAM-B and VAM-M, respectively. The grain yield/plant was significantly increased by using VAM inoculation alone (comparedwith P fertilizer) or combined with N fertilization (compared with N fertilizer alone). This trend was holds true in both seasons. The grain yield/plant was proportionally and significantly increased, in general, as VAM inoculation level increased.32.In 2002 season, VAM-B, VAM-S and VAM-0 significantly increased the highest weight of 100-kernels more than VAM-M. While, VAM-B was the most effective in 2003 season followed by VAM-S, VAM-0 and VAM-M, respectively. Applying VAM inoculation at any level alone or combined with N fertilizer significantly increased weight of 100-kernels compared with application of P and N fertilizers, respectively. This trend was true in both 2002 and 2003 seasons.33.VAM-B recorded the highest significant increases in the crude protein (CP) in the maize kernels in 2002 season followed by VAM-M, VAM-0 and VAM-S isolates, respectively. In 2003 season, VAM-B was the best followed by VAM-O, VAM-S and VAM-M, respectively. The CP was significantly increased, in general, by increasing VAM inoculation level compared with P application. Also, CP significantly increased by using N fertilizer + VAM inoculation at any level compared with N fertilizer alone in both seasons. Using N fertilization + VAM-0 (high level), VAM-B (middle level) or VAM-M (low and the middle levels) in 2002 season and N fertilizer + VAM-S and VAM-0 (high level) in 2003 season produced

the highest CP content.34.VAM inoculation at different levels alone or combined with N fertilizer significantly increased phosphorous (P) content in maize kernels compared with P and N fertilizers, respectively. This trendwas true in both 2002 and 2003 seasons. The highest P content in the maize kernels was produced by N fertilizer + VAM-B (middle level) in 2002 season and VAM-0 (high level) in 2003 season.35. VAM inoculation significantly increased potassium (K) contentin the corn kernels compared with P fertilization. The P content increased proportionally as VAM inoculation level increased. However. N fertilizer + VAM inoculation (all levels) had no significant effect on K content in the corn kernels compared with N fertilization alone.