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## Diversity of Arbuscular Mycorrhizal Fungi associated with common sugarcane varieties in Iran

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Sixteen species of AM fungi associated with four common sugarcane cultivating varieties of CP57-614, CP48-103, CP69-1062, NCO-310 in the Khuzestan province were identified. With this, four genera including *Glomus*, *Paraglomus*, *Pacispora* and *Kuklospora* were identified as *Glomus ambisporum*, *G. manihotis*, *G. eburneum*, *G. diaphanum*, *G. aggregatum*, *G. caledonium*, *G. coronatum*, *G. mosseae*, *G. geosporum*, *G. etunicatum*, *G. intraradices*, *G. lamellosum*, *G. microcarpum*, *Kuklospora colombiana*, *Pacispora scintillans* and *Paraglomus occultum*. This is the first report of *K. colombiana*, *G. eburneum*, *G. manihotis*, *G. diaphanum* and *P. occultum* from Iran. Moreover, *G. aggregatum*, *G. caledonium*, *G. coronatum*, *P. scintillans* are new recorded for sugarcane mycoflora in Iran. Results showed that the abundance and population diversity of these different species of AM fungi differ with variety they are associated with. The most diversity of association was found with CP48-103 and the least with NCO-310.

### Introduction

Sugarcane, a member of Gramineous plants, is among the plantations that has a long lasting relationship with the Khuzestan province soils. Although according what extracted from itinerary of ancient historians, the first purified white sugar was made in Khuzestan province in 600 AD, But it seems that development of industrial sugar production by raising different kinds of varieties dates back just to forty years ago where four common varieties of sugarcane i.e. CP57-614, CP48-103, CP69-1062, NCO-310 with distinct agronomic characteristics widely used in different sugarcane growing sites which now reaches to more than sixty thousand hectares all over the province. In spite of this long lasting history, few research works have been done on

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identification of AM Fungi associated with the plant, their diversities and beneficial efficacies and their interactions with other soil micro-organisms. Arbuscular Mycorrhizal Fungi (AMF) are a group of obligate plant symbionts belonging to Phylum Glomeromycota (Schussler *et al.*, 2001). Mycorrhizal fungi are relevant members of the rhizosphere mutualistic mycosymbiont populations that are known to carry out many critical ecosystem functions such as improvement of plant establishment, enhancement of plant nutrient uptake, plant protection against cultural and environmental stresses and improvement of soil structure (Smith and Read 1997). Mycorrhizal establishment is known to modify several aspects of plant physiology including mineral nutrient composition, hormonal balance, and carbon allocation patterns (Azcon-Aguilar and Bago 1994). Rhizosphere functioning is known to influence markedly plant fitness and soil quality because microbial developments in such environment can help the host plant to adapt to stress conditions concerning water and mineral deficits, and the presence of soil-borne plant pathogens (Lynch 1990; Bowen and Rovira 1999). Consequently, the mycorrhizal symbiotic status changes the chemical composition of root exudates, while the fungal mycelium serves as a carbon source to rhizosphere microbial communities, and further, introduces physical modifications in the environments surrounding the roots (Barea *et al.* 2002). On the other hand Nearly 5 to 23% of all photosynthetically fixed carbon is transferred to the rhizosphere through root exudates (Marschner 1995; Walker *et al.* 2003). The compounds secreted by plant roots serve important roles as chemical attractants and repellants in the rhizosphere (Estabrook and Yoder 1998; Bais *et al.* 2001). So Rhizosphere microbial communities can be regarded as a subset of the soil microbial community. The species composition of microbial communities in rhizosphere differs from that in the bulk soil (Foster 1986; Marilley and Aragno 1999). This is a clear indication that plants have a strong influence on the microbial populations on their roots. Indeed, in many cases the rhizosphere communities of different plant species growing in the same soil are distinct (Ibekwe and Kennedy 1998) and plants may even have very similar microbial community composition in different soils (Grayston *et al.* 1998; Miethling *et al.* 2000). Plant/host factors stimulate AMF hyphal growth *in vitro* and also during precolonization phase of AMF formation (Becard and Piche 1989). Specific compounds in the root exudates act as signal molecules capable of stimulation/induction of hyphal growth, branching, differentiation and host penetration (Becard and Piche 1989, 1990). This research was conducted to see if there are any preferences among different common cultivating sugarcane varieties to be colonized by distinct species of AM fungi or in other case whether root exudations of different

sugarcane varieties have attracting or repelling influences on their partnerships with AM fungi.

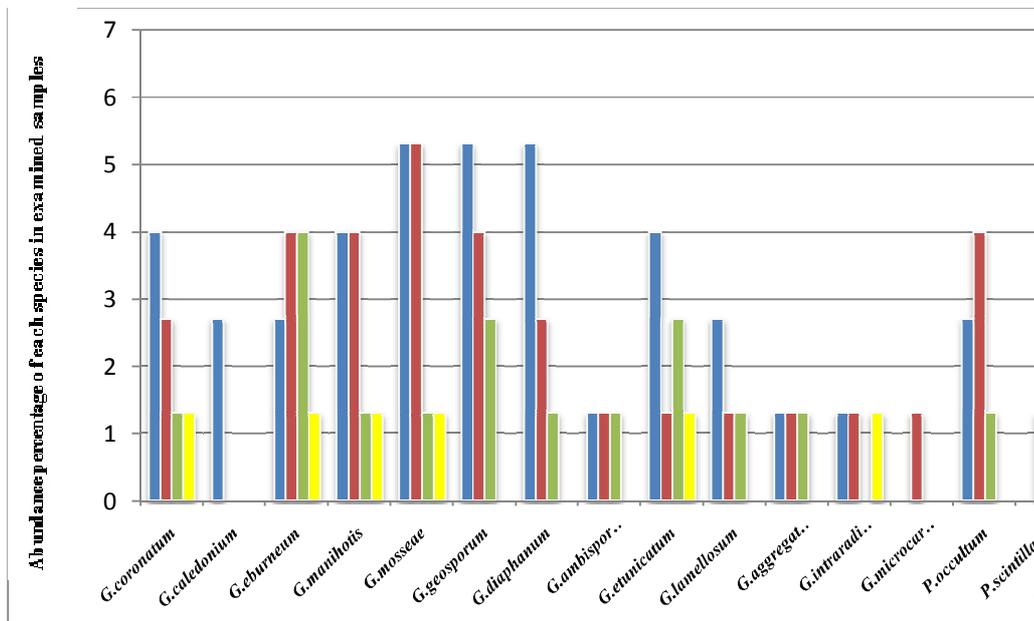
### Materials and Methods

Soil samples were collected from sixty fields of main sugarcane agro-industry companies including Haft-Tape, Karoon, Mian-Ab, Imam-Khomeini and Amir-kabir during November and December of 2004 during the end of growing season and beginning of harvest. Samples were collected from rhizosphere of four extensively grown sugarcane varieties, CP57-614, CP48-103, CP69-1062 and NCO-310 in the Khuzestan province that each variety shared the same number of samples (15 samples from each). Each sample was mixed 10 subsamples which were gathered accidentally from different parts of selected fields. In order to get the most diversities of AM Fungi, it was attempted to choose the oldest fields and the ones with sandy soil texture. Trap cultures 100 grams of soil and root of each sample mixed with sterilized sand, potted with corn seeds and kept for four random taken and half month in greenhouse. To confirm symbiotic association in sugarcane and corn, roots were boiled in 10% KOH, stained with 0.05% aniline blue solution in lacto phenol and were examined by optical microscope. Isolation of spores were carried out by centrifugation in 55% sucrose solution and then extracted spores were transferred to 1:1 mixture of PVLG (Koske and Tessier, 1983) and Melzer's reagent (Hall, 1984) and 400 microscopic slides (100 from each soil sample of each remarked variety) were prepared. Identification of species were carried out which based on morphological and morphometrical characteristics of spores, and subtending hyphae using stereomicroscope (Olympus ZSH10) and bright field microscope (Olympus BH-2). Following the complete examination of all microscopic slides the diversity and abundance of identified species with regard to mentioned varieties were studied.

### Results and discussion

In this research Sixteen species of AM fungi was found that belonging to four families including Glomaceae, Paraglomaceae, Diversisporaceae and Acaulosporaceae. Four genera including: *Glomus*, *Paraglomus*, *Pacispora* and *Kuklospora* were identified as *Glomus ambisporum*, *G. manihotis*, *G. eburneum*, *G. diaphanum*, *G. aggregatum*, *G. caledonium*, *G. coronatum*, *G. mosseae*, *G. geosporum*, *G. etunicatum*, *G. intraradices*, *G. lamellosum*, *G. microcarpum*, *Kuklospora colombiana*, *Pacispora scintillans* and *Paraglomus occultum*. This is the first report of *K. colombiana*, *G. eburneum* *G. manihotis*,

*G. diaphanum* and *P. occultum* from Iran. *G. aggregatum*, *G. caledonium*, *G. coronatum*, *P. scintillans* are new recorded for sugarcane mycoflora in Iran. *G. ambisporum* is new for Khuzestan sugarcane mycoflora whereas *G. mossae*, *G. geosporum*, *G. etunicatum*, *G. intraradices*, *G. lamellosum* that have been formerly reported by other Iranian investigators who reported with comparative descriptions. Also status of association of isolated species with common cultivating sugarcane varieties of CP57-614, CP48-103, CP69-1062 and NCO-310 was investigated. The results showed that the most diversity of association was found with CP48-103. Among identified species it was just *G. caledonium* that was not isolated from the mentioned variety. Comparing other varieties, was found *K. colombiana* or various other taxa of AM fungi which sampling from the rhizosphere of CP48-103 was the most diverse association with the identified AM fungi and with CP69-1062. *Kuklospora colombiana*, *Glomus microcarpum* and *Pacispora scintillans* were isolated from its soil samples. CP57-614 was associated with eleven species but *G. caledonium*, *G. intraradices*, *Glomus microcarpum*, *P. acintilans* and *K. colombiana*. The least number of association was NCO-310 where association of six species, *G. manihotis*, *G. mosseae*, *G. etunicatum*, *G. eburneum*, *G. coronatum*, *G. intraradices*. The results showed that apparently the abundance of AM fungi population differ with regard to colonized variety. The most and the least population frequencies of isolated species were found with CP69-1062 and NCO-310 respectively.



**Fig. 1.** Diversity and population frequencies of isolated species of AM fungi associated with common cultivating sugarcane varieties in Khuzestan

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