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Arbuscular Mycorrhizal Fungi Associated with Wheat in Semi-arid Production System of North Gujarat

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Abstract: Arbuscular mycorrhizal fungi (AMF), a dynamic micro-symbiont form an intimate link between the soil environment and plant-root-systems and provide them better adaptability especially in stress agro-ecosystems. Different wheat based cropping systems were studied for AMF associations. A wide variability was observed for both spore density and root colonization by AMF in wheat based cropping systems. Thirty seven species representing five genera were isolated from rhizosphere soils of different wheat based crop-sequences including Mehsana, Sabarkantha and Banaskantha districts of North Gujarat. Among the AM species isolated from rhizosphere soils under different crop sequences, *Glomus* was found dominating which recorded 18 species followed by *Acaulospora* (10), *Sclerocystis* (4), *Gigaspora* (3) and *Scutellospora* (3). In general, *Glomus mossae*, *G. hoi*, *G. fasciculatum*, *Acaulospora sporocarpia* and *A. elegans* are the dominating AMF species frequently occurred. About 6-10 times higher sporulation was observed compared to no AMF application in wheat. Five most efficient AMF species namely *Glomus mossae, G. constrictum, G. fasciculatum, G. intraradice and G. restrictum* were identified having compassion with wheat.

Key words: AMF associations, sporulation, root colonization, wheat, north Gujarat

India harvested record wheat production of 109.24 million ton during 2020-2021 from an area of 31.58 million ha (https://agricoop.nic.in/). Besides major wheat growing states of India, Gujarat is also an important wheat producing state in the central zone of India which is well known for producing high quality wheat. The cultivation of wheat in Gujarat is highly variable depending upon the short window of the winter months. Typically it is around 14 weeks compared to over 25 weeks elsewhere in the wheat bowl of North India. Therefore, it is very impressive to note that per day productivity of wheat in Gujarat is comparable to the best global per day productivity of wheat Patel *et al*. (2018). Recently, a substantial increase was observed in acreage (1-25%), production (4-35%) and productivity (8-12%) since 2005 onward in wheat cultivation in Gujarat state (https//:agri.gujarat.gov.in). This increase in wheat production was attributable to high yielding varieties, improved technology, back up irrigation facilities, congenial administrative policies, and favorable weather conditions, especially prolonged winter. The wheat cultivation area is almost equally distributed in three major wheat-growing regions namely north Gujarat (31%), Saurashtra (37%) and middle Gujarat (30%). However, despite a better winter temperature window, the wheat productivity was comparatively less in north Gujarat situation due to poor soils and environmental stresses. The abiotic stresses like salinity and drought, coarse textured soils and poor organic matter and available nutrients are the common concern in these soils. Besides nitrogen, phosphorus and micronutrients are the important nutrients for wheat productivity in these soils.

The AMF are ubiquitous in nature and a dynamic micro-symbiont especially for nutrient-deficient, poor- quality and marginal land. These are soil-borne fungi that helps to cope with the abiotic and biotic stresses, particularly by alleviating water and mineral deficiencies and safeguarding the plants from pathogens (Panwar *et. al*., 2008: Smith and Read, 2008). Therefore, AMF are vital endosymbionts playing an effective role in plant productivity and the functioning of the ecosystem (Gianinazzi *et al*., 2010). It provides a nature-friendly alternative

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to chemical fertilizers. Thus, AM fungi may play a key role in sustainable wheat production, particularly in semi-arid regions of north Gujarat agro-ecosystems. The present study was planned to assess the diversity of AMF associated with wheat plants in sandy loam soils of north-Gujarat and pick-up the most efficient strains for the enhanced productivity of wheat sustainably.

Materials and Methods

The ecological conditions in north Gujarat are sparse having arid to semi-arid climate with limited water availability and poor soil conditions. An intensive field survey was conducted for Mehsana, Sabarkantha and Banaskantha districts of North Gujarat covering different wheat based cropping systems during 2007 to 2009.

Soil sampling

The survey was undertaken during rabi season (December to March) and roots and soil samples from wheat rhizosphere were collected and transported to laboratory using insulated carrier bags. These soil samples were stored at 4°C till further processing and the roots were processed immediately. The soil samples collected from specific location and cropping systems were homogenized before processing by sieving (2 mm mesh size) to remove stones, plant material and coarse roots. Two subsamples for each soil were used for characterization of soil properties and trap culture (pot) studies. The collected soil samples were analyzed for pH and electrical conductivity (EC) using soil to water suspension (1:2.5) by digital pH and EC meter, respectively. The soil properties were estimated using standard procedures described in Jackson (1967). The organic carbon using Walkley and Black (1934) was used for estimation of organic carbon, available P by Olsen *et al*. (1954) and available potassium using flame photometer. Microsoft Excel 2000 was used in the statistical processing of the data.

Spore extraction

The AMF endophytes associated with wheat were isolated using wet sieving and decanting technique of Gerdemann and Nicolson (1963). The spore density (spore/100 g soil) was estimated using the method of Gaur and Adholeya (1994). For further studies, the isolated spores were also mounted using PVLG (polyvinyl lactoglycerol) and Meltzer's reagent (1:1 v/v). The taxonomic studies of AMF strains was done with the help of standard manuals (Schenck and Perez, 1990) and as described by international collection of vesicular and arbuscular mycorrhizal fungi (http://invam.caf.wvu.edu).

Trap culture studies in pots

The trap culture experiment was carried out in earthen pots (15 kg capacity) for *in vitro* induction of AMF spores using pearl millet roots. For this soils were autoclaved alternatively for three consecutive cycles and filled in earthen pots by mixing homogenously (1:1, v/v), the autoclaved soil plus 500 g soil collected from field for isolation of AMF species. The seeds of pearl millet were surface sterilized using 0.1% w/w mercuric chloride solution for 2 min and then washed with distilled water. About 8-10 of these seeds were sown as host crop for AMF sporulation and the pots were watered at field capacity using weight loss method.

Studies for AMF spore colonization using different wheat cultivars

The efficient AMF strains were identified using different wheat cultivars namely GW 322, GW 173, GW 273 and GW 496 for sporulation and root colonization using pot studies. To determine percent root colonization, root samples were washed in tap water and processed to investigate different infection patterns (hyphal, arbuscular, vesicular and total root colonization). The roots were stained and assessment of colonization was done for each sample using a microscope slide method (Phillips and Hayman, 1970). The presence or absence of AM colonization was evaluated in 100 randomly selected root segments under the dissection microscope. Total AMF colonization was expressed as the ratio of infected root sections to the total number of root sections examined (Biermann and Lindermann, 1981).

Results and Discussion

Soil properties and occurrence of AMF species in wheat based cropping system

The survey included Sabarkantha, Mehsana and Banaskantha districts of north Gujarat. The soils are generally coarse in texture representing sandy loam in part of Mehsana and Sabarkantha and loamy sand to sandy in part of Mehsana and Banaskantha districts. The soils were alkaline in pH (7.20 to 8.18) and mostly low to medium in organic carbon (0.22 to 0.49%). The available phosphorus and potassium were found medium to high in soils of these surveyed districts. The cotton-wheat-pearl millet, mung bean-wheat-pearl millet, cotton-wheat-mung bean, cotton-wheat, groundnut-wheat were the major cropping system adopted in the surveyed area. Total thirty eight species representing five AMF genera namely *Glomus, Acaulospora, Gigaspora, Sclerocystis* and *Scutellospora* were identified in the surveyed area associated with wheat plants (Table 1). The highest eighteen species of AMF were observed for *Glomus* followed by *Acaulospora* (10), *Sclerocystis* (4), *Gigaspora* (3) and *Scutellospora* (3). A very poor spore density (0.3 to 1.4 spores per gram soil) was recovered in the surveyed area. However, the level of root infection was observed appreciably high (22 to 84%) in the wheat plants collected from the surveyed area. This poor density of AMF sporulation might be due to the use of high inputs particularly in the preceding cotton crop, especially nitrogen and pesticides, which might be affecting the sporulation of AMF propagules in these soils. The highest root colonization was observed in the Sabarkantha (42 to 84%), followed by Mehsana (30-78%) and Banaskantha (22-87%) districts. The frequent occurrence of AMF species belonging to genus *Glomus* were reported widely in arid environment (Tarafdar and Praveen-Kumar, 1996; Panwar and Tarafdar, 2006a, 2006b). Generally the low spore density for these AMF species reported in these sparse environmental conditions might be due to multiple abiotic stresses, high temperature and poor availability of the hosts (Sun *et al*., 2018). Generally low spore density was reported in arid production system (Requena *et al*., 1996).

Identification of efficient AMF species

For identification of the efficient AMF strains, the collected species were screened in the pot culture using four different wheat cultivars namely GW 322, GW 173, GW 273 and GW 496. These four wheat cultivars were the most adopted and covering more than 95% of the total wheat acreage in the north Gujarat. All the AMF species varied greatly for sporulation and root colonization of the four wheat cultivars (data not shown). The AMF species showing fairly good sporulation (>200 spore/100 g soil) and root colonization (>30%) were considered as most efficient and thus identified four species namely *Glomus mossae, G. constrictum, G. fasciculatum and G. intraradices* in pot culture studies (Table 2). Generally *Glomuss mossae* showed the highest degree of average sporulation (182 to 244 spore/50 g soil) as well as root colonization (62 to 72%) followed by *G. constrictum* (129 to 212 spores/50 g soil; 62 to 71%) or *G. fasciculatum* (135 to 168 spores/50 g soil: 54 to 69%) and least by *G. intraradices* (111 to 145 spores/50 g soil; 49 to 67%), respectively (Table 2). All the wheat cultivars found as suitable hosts for the AMF species tested varied for their specificity in spore population and root colonization. For example sporulation of *G. mossae* was highest in all the wheat cultivars but the root colonization varied with cultivars. The highest root colonization was observed for *G. mossae* in GW 173 and GW 496, for *G. fasciculatum* in GW 322 and *G. constrictum* in GW 273. These variations in sporulation and root

Table 1. Distribution of AMF species recovered from wheat based cropping systems in semi-arid production system of north Gujarat

Area Surveyed (Name of district)	% Root colonization	Spore density*	Total No. of AMF species	AMF genus recovered from all the locations
Mehsana	30 - 78	0.5 - 1.2	25	*Acaulospora, Glomus, Gigaspora, Sclerocystis, Scutellospora*
Sabarkantha	42 - 84	0.3 - 0.9	16	
Banaskantha	22 - 87	0.5 - 1.4	37	

Dominant AM species:
Acaulospora elegans, A. sporocarpia, A. thomii, A. gerdemannii, A. denticulate, A. rehmii, A. nicolsonii, A. spinosa, A. lacunose, A. bireticulata, Glomus mossae, G. aggregatum, G. ambisporum, G. constrictum, G. convolutum, G. fasciculatum, G. geosporum, G. intraradices, G. rubiforme, G. sinuosum, G. heterosporum, G. clarodium, G. hoi, G. radiatum, G. etunicatum, G. macrocarpum, G. microcarpum, G. margarita, Gigaspora albida, Gig. margarita, Gig. candida, Sclerocystis cocogena, S. microcarpus, S. pachycaulis, Scutellospora heterogama, S. minuta, S. pellucida.

*No. of spores per gram soil

Table 2. Response of efficient AMF species for sporulation and root colonization using different cultivars of wheat growing in north Gujarat

AMF species	GW 322	GW 173	GW 496	GW 273
Spore population (Per 50 g soil)				
Glomus mossae	206±98	244±104	182±72	189±58
G. constrictum	129±54	212±84	154±36	134±38
G. fasciculatum	144±32	156±28	135±36	168±46
G. intraradices	111±27	118±51	145±41	114±41
Root colonization (%)				
Glomus mossae	48-89(62)	41-93(72)	46-79(68)	51-73(62)
G. constrictum	58-69(65)	35-92(69)	43-76(62)	40-81(71)
G. fasciculatum	55-72(67)	49-78(63)	44-62(54)	48-86(68)
G. intraradices	36-75(49)	32-74(59)	42-72(67)	39-69(62)

± represent the standard deviation; the parenthesis represents the mean values for root colonization.

colonization attributed to the soil properties as well as the host specificity. The spore density of AMF had a strong positive correlation with soil pH and organic carbon content and a negative correlation with Olsen's P content of the soil (Panwar and Tarafdar 2006a, 2006b).

Conclusion

A wide variability was observed for both spore density as well as root colonization by AMF in wheat based cropping systems in north Gujarat. *Glomus* spp. was found most dominating AMF associated with wheat-based cropping system and widely recovered from north Gujarat semi-arid agro-ecosystem soils. The efficient AMF species having high spore density (>1-2 spores g^{-1} soil) were recovered in the order of *G. mossae* > *G. constrictum* > *G. fasciculatum, G. intraradice* > *G. restrictum*. These species of AMF may have potential for enhanced yield and nutrient acquisition especially for immobile soil nutrients in wheat crop.

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