Research Article

Investigating arbuscular mycorrhizal fungal infection in medicinal plant roots in different localities of tehsil Shabqadar, district Charsadda, and Khyber Pakhtunkhwa Province, Pakistan

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Citation

Tabassum Yaseen, Farah Mabood¹, Rani Gul, Kushnood Ur Rehman and Naveed Akhtar. Investigating arbuscular mycorrhizal fungal infection in medicinal plant roots in different localities of tehsil Shabqadar, district Charsadda, and Khyber Pakhtunkhwa Province, Pakistan. Pure and Applied Biology. Vol. 9, Issue 1, pp427-435.

<http://dx.doi.org/10.19045/bspab.2020.90046>

Received: 22/07/2019

Revised: 19/09/2019

Accepted: 07/11/2019

Online First: 15/11/2019

Abstract

Arbuscular mycorrhiza represents plant-fungal association in which fungal partner gets carbohydrates and improves the nutrient uptake of host plants enhancing plant growth and productivity. The present work was done with the aim of generating baseline information on Arbuscular mycorrhizal fungi (AMF) in medicinal plants of different locations of Tehsil Shabqadar, District Charsadda, Khyber Pukhtunkhwa Pakistan. During the study, total of 40 medicinal plants were evaluated at different growth stages (i.e. vegetative, fruiting, and flowering) for investigating the AMF root colonization. During the study, high root colonization (95-100%) was shown by family Asteraceae, and Meliaceae, and the lowest in families of Libatacea, Solanacea, Lamaceae, Chenopodiaceae and Rhamnaceae respectively (10-20%). Among the localities, the highest host colonization by AMF was found in village Gonda and Nasratzai (100%) followed by Village Kangra and Pindi kaly, whereas no colonization/infection was found in Karimo. Similarly, the concentration of sand, silt, clay, organic matter, Calcium etc. determined at all locations studied were also different. As large number of medicinal plants can be found in different Locations of District Charsadda, extensive research work is required to create database of mycorrhizal species colonizing these medicinal plants and to determine their medicinal values for treatment of different infections.

Keywords: Arbuscular mycorrhizal fungi; Medicinal plants; Roots colonization; Shabqadar

Introduction

Mycorrhiza is symbiotic association between arbuscular mycorrhizal fungi (AMF) and plant roots [1]. This association is found

between the majority (80%) of terrestrial plant species [2]. It can benefit plants through improved nutrients uptake such as phosphorus (P), nitrogen, (N) and zinc (Zn)

[3-5]. The role of AMF on nutrient uptake (N, P, K) as well as effect of nutrient intensity on AMF crops has previously been studied [6, 7]. Colonization of AM fungi has negatively been affected by P and positively affected by N addition [8]. Colonization of roots by AM fungi improve growth and productivity of several field crops [9, 10], increasing uptake of nutrient elements [7]. Medicinal plants may be defined as those plants that are commonly used in treating and preventing specific ailments and diseases and that are generally considered to be harmful to humans [11].

In Pakistan, more than 60% of the population living in rural areas are not either aware of modern health care facilities or cannot afford it due to which they rely on old health care practices including use of extracts of medicinal plants [12]. Considering the high dependency on medicinal plants, it is suggested to analyze their metal composition, side effects and proximate analysis etc. [13] so that it can be safely administered to patients. These plants species are not only used for medicinal purposes but also been used as food source [13, 14]. To get an overview on AMF infection in the medicinal plants, the present study aims to generate baseline information on Arbuscular mycorrhizal fungi (AMF) in medicinal plants of different locations of Tehsil Shabqadar, District Charsadda, Khyber Pukhtunkhwa Pakistan.

Materials and methods

Forty samples of plant roots and soil were collected from different areas of District Charsadda during 2015-2016. These areas include Kangra, Nasratzai, Shahganga, Gonda, Yaghyband, Pindi kally, lalma, Shakoor, Bulb kally and Karimo. Plant samples studied belonged to different families, including Nyctaginaceae (1 specie), Poaceae (2 specie), Lamiaceae (4 species), Cannabaceae (2 species), Amaranthaceae (2 species), Meliaceae (2

species), Solanaceae (6 species), Asteraceae (5 species), Fabaceae (2 species), Chenopodiaceae (1 species), Verbenaceae (3 species), Apiaceae (1 species), Asclepiadaceae (2 species), Marsilaceae (1 specie), Zygophyllaceae (1 specie), Sapindaceae (1 specie), Rhaminaceae (1 specie), Cucurbitaceae (1 specie), Malvaceae (1 specie) and Labitaceae (1 specie).

The plants were carefully dug up along with their rhizosperic soil in triplicate and transported to laboratory in polythene bags. The plant roots were gently washed under tap water to remove soil particles (Table 1).

Assessment of roots colonization was done using the protocol of Phillips and Hayman, [15, 16]. The power of hydrogen (pH) of soil water suspension was determined using Equipments, power of hydrogen (pH)meter as described by McClean, [17] and electric conductivity of soil samples was measured by EC meter [18]. Texture class of the soil was determined using textural triangle [19]. Organic matter was determined using Walkly-Black method [20].

Results and discussion

This Study illustrates Arbuscular mycorrhizalfungi infection in roots of medicinal plants from different study sites of District Charsadda. AMF infection in roots of medicinal plant has widely been studied [21-27].

In our study, about 85% AMF infection was seen in plant *Mirabilis Jalapa*, (Family, Nyctaginaceae) (Table 1, Fig. 1-4) with Mean values of EH (0 ± 0), IH (16.7 ± 14.433) A (40 ± 0) and (20 ± 0). The present study is in line with study of Sharma and Jha [28] who has investigated *Mirabilis jalapa* root length with hyphae, root length with arbuscules, root length with vesicles.

In the present study, the highest root colonization was observed in family Asteraceae (100) which is in concordance to previous results [29].

In our study, the lowest root colonization was observed in families Labitaceae, Lamiaceae showing 10% and 15% colonization respectively. No colonization was observed in non-mycorrhizal plants *Chinopodium album* which is in agreement to results of Hildebrandt [30]. Although plants have been categorized as mycorrhizal and non-mycorrhizal plants but this classification is not very convincing as several non-mycorrhizal plants have been demonstrated to be colonized by AM fungi [31-33]. Similarly, some mycorrhizal plants also did not get colonized due to unknown reasons. Our results of getting no colonization even in case of mycorrhizal plants has also previously been reported by other researchers [30].

Our results are also in line with those of Karagiannides *et al.* [34] who also reported the occurrence of mycorrhiza in some members of the family Lamiaceae, Poor roots

colonization was noted from family Poaceae (20%) during the study which is in contrast to study conducted by Olfat and Jalil [35] where highest roots colonization (i.e. 67%) has been reported from Poaceae. The AMF infection observed from family Fabacea (40%) during our study is in concordance to those reported by Olfat and Jalil [35] where the infection percentage was 59%. Similarly the high infection reported from families Astereace and Lamiceae during our study is in contrast to these authors who reported nearly 40-50% infection in these families. The lowest infection reported by their studies are from families Apiaceae and Solanaceae which is in contrast to our study as we have reported high infection percentage from these families. Our results of getting low infection in family Chenopodiaceae does not support the finding of Alejandra *et al.* [36] who have reported moderate roots infection in their experiment.

Figure 1. AM fungi detected in different medicinal plants (*Mirabilis jalapa*, *Cynodondactylon*, *Oscimumbasilicum*, *Minthapiperata*, *Canabus sativa*, *MeliaAzedric* *Acharanthusapera* *Solinum Nigrum*, *Xnthium Stromarium*, of Shabqadar villages. EH (External hyphae), IH (Internal hyphae), A (Arbuscules), V (Vesicles)

Table 1. AMF root colonization of different medicinal plants of District Charsadda

S. No.	Plant species	Families	EH%	IH%	A%	V%	Root colonization%
1	Mirabilis Jalapa	Nyctaginaceae	0±0	16.7±14.433	40±0	20±0	85
2	Cynodon dactylon	Poaceae	10±0	5±0	0±0	5±0	20
3	Ocimum basilicum	Lamiaceae	0 ±0	30±17.32051	25±0	16.67±14.43376	95
4	Mintha piperata	Lamiaceae	0±0	31.67±2.8867	30±0	25±0	90
5	Cannabis sativa	Cannabaceae	0±0	0±0	0±0	3.33±5.773503	0
6	Melia azedarach	Meliaceae	35±0	16.667±5.773503	25±0	15±0	95
7	Achranthus aspera	Amaranthaceae	0± 0	16.667±5.773503	26.67±23.094	25±0	90
8	Solanum nigrum	Solanaceae	6.666±2.886751	23.3±2.886751	5±8.660254	10±0	70
9	Xanthium strumarium	Asteraceae	38.5±0	0±0	0±0	10±0	15
10	Conyza Canadensis	Asteraceae	13.3±2.886751	25±0	35±0	25±0	100
11	Acacia concinna	Fabaceae	6.6672.886751	10±8.660254	30±0	35±0	40
12	Chenopodiaceae ambrosioides	Chinopodiaceae	0± 0	15±0	0±0	0±0	20
13	Pertinum hysteroporus	Asteraceae	10±5	15 ± 0	0±0	15±0	60
14	Verbiscum Thapsus	Verbenaceae	15±10	0±0	15±0	25±0	45
15	Chenopodiaceae album	Chinopodiaceae	0± 0	0± 0	0± 0	0± 0	0
16	Verbinia officinalis	Verbenaceae	1±0	0±0	3.33±2.88675	20±0	50
17	Apium graveolens	Apiaceae	21.7±2.886751	18.334±2.88671	20±0	0±0	70
18	Datura stramonium,	Solanaceae	6.67±2.886751	30±0	0±0	15±0	10
19	Calotropis procera	Apocynaceae	18.3±7.637626	20±0	5±0	45±0	50
20	Marsilea quadrifolia	Marsilaceae	8.33±2.886751	10±8.660254	11.67±20.207	45±0	65
21	Peganum harmala	Nitrariaceae	18.33±2.886751	0±0	15±0	25±0	60
22	Calotropis procera	Apocynaceae	0±0	0±0	0±0	0±0	0
23	Dodonaea viscosa	Sapindaceae	23.3±2.886751	18.334±5.773503	30±0	0±0	65
24	Rhazya stricta	Apocynaceae	18.33±2.886751	15±0	31.67±5.77350	16.67±14.43376	70
25	Ziziphus mauritiana	Rhamnaceae	0±0	0±0	0±0	10±0	20
26	Verbascum thapsus	Scrophulariaceae	1.667±2.886751	0±0	5±0	5±8.660254	25
27	Acacia concinna	Fabaceae	0±0	10±0	40±0	45±0	45
28	Solanum surattense.	Solanaceae	0±0	0±0	10±0	45±0	55

29	<i>Momordica Cochinchinensis</i>	Cucurbitaceae	8.33±2.886751	30±7.071068	25±0	20±0	85
30	<i>Datura stramonium,</i>	<i>Solanaceae</i>	31.67±2.886751	15±7.071068	40±0	20±0	85
31	<i>Canabis sativa</i>	Cannabaceae	16.675.773503	20±0	10±0	0±0	40
32	<i>Lycopersicon esculentum</i>	Solanaceae	0±0	7.5±3.535534	5±0	1.667±2.886751	25
33	<i>Ocimum basilicum</i>	Lamiaceae	10±0	10±0	5±0	0±0	15
34	<i>Mentha spicata</i>	Lamiaceae	3.33±5.773503	0±0	0±0	10±0	20
35	<i>Calendula officinalis</i>	Asteraceae	6.667±2.886751	0±0	45±0	5±2.886751	55
36	<i>Hibiscus esculentum</i>	Malvaceae	10±17.32051	0±0	30±25.98076	30±0	75
37	<i>Mentha vulgaris</i>	Lamiaceae or Labiatae	0±0	12.5±3.535534	5±0	24.667±21.36196	10
38	<i>Melia azedarach</i>	Meliaceae	13.34±2.88675	7.5±10.6066	30±0	0±0	45
39	<i>Capsicum annuum</i>	Solanaceae	1.667±2.88675	0±0	0±0	35±0	40
40	<i>Carthamus oxyacantha</i>	Asteraceae	0±0	0±0	0±0	0±0	0

Key: EH (External hyphae), IH (Internal hyphae), A (Arbuscules), V (Vesicles)

Figure 2. AM fungi detected in Different medicinal plants (*Accaciacoincina*, *Chenopodiaceae ambrosioids*, *Pertinum hysteroporus*, *Verbescum Thapsus*, *Chenopodiaceae album*, *Verbena officinale*, EH (External hyphae), IH (Internal hyphae), A (Arbuscules), V (Vesicles) *apigravelones*, *Dhatora stramonium*, *callotropus procera*, *marseliaquadrifolia*)

Figure 3. AM fungi detected in Different medicinal plants (*Piganumharmalla*, *Calotropas procera*, *Dodonea viscosa*, *Raziastratica*, *Zizipus muratiana*, *Verbescum Thapsus*, *Alhiji marjoram*, *Solanum soratense*, *Memordicachenchinensis*, *Dhatora stramonium*). EH (External hyphae), IH (Internal hyphae), A (Arbuscules), V (Vesicles)

Figure 4. AM fungi in Different medicinal plants (*Canabus sativa*, *Lycopersicum esculentum*, *Oscimum basilicum*, *Menthaspicata*, *Calendula officinale*) (*Hibiscus esculentum*, *Mentha sylestris*, *Melia azedaric*, *Carthamus oxicantha*). EH (External hyphae), IH (Internal hyphae), A (Arbuscules), V (Vesicles)

Conclusion

This study confirms that AMF effectively improved the growth of medicinal plant. The growth promotion of medicinal plants depends on the soil mycorrhizal infectivity and functional abilities of soil microorganisms rely on inoculation of AM fungi. Inoculation of field crops with AM fungi can prove an effective practice in improving the production of medicinal plants and also improve the soil bio-functioning. It is also worth thinking to have detailed knowledge of AMF ecology, better fungal strain, adaptability to environmental conditions for getting better yields and sustainability.

Authors' contributions

Conceived and designed the experiments: T Yaseen & F Mabood, Performed the experiments: F Mabood, Analyzed the data: R Gul, KU Rehman & N Akhtar, Contributed materials/ analysis/ tools: R Gul, KU Rehman & N Akhtar, Wrote the paper: T Yaseen & F Mabood.

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