



Research Article

INCIDENCE OF FUNGAL PATHOGENS ASSOCIATED WITH MUNGBEAN (*Vigna radiata*) IN ALLAHABAD REGION

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Received: April 06, 2017; Revised: April 26, 2017; Accepted: April 27, 2017; Published: May 18, 2017

Abstract- The incidence of fungal pathogens associated with Mungbean (*Vigna radiata*) in Allahabad region was carried out with the objectives to isolate and identify the fungal pathogens from the rhizosphere of Mungbean crop. As fungal diseases is one of the most important constraints to Mungbean production in India. 50 soil samples which are collected from the rhizosphere of infected Mungbean crop were found to have fungal incidence (78%). Among 39 fungal isolates, 6 fungal isolates were found to have incidence of which *Aspergillus terreus* (12.82%) have shown high incidence followed *Cladosporium sphaeroperumum*, *Curvularia lunata*, *Aspergillus awamori* and *Chaetomium globosum* (10.26%), *Alternaria alternate* and *Rhizoctonia solani* (7.69%), *Penicillium crateriforme*, *Helminthosporium australiensis*, *Mucor circinelloides*, *Rhizopus stolonifer*, *Trichoderma harzianum* (5.13%), *Aspergillus flavus* and *Fusarium oxysporium* (2.56%).

Keywords- Mungbean, Fungal pathogens, Incidence, Rhizosphere, isolates.

Citation: Sandhya N., et al., (2017) Incidence of Fungal Pathogens Associated with Mungbean (*Vigna radiata*) in Allahabad Region. International Journal of Agriculture Sciences, ISSN: 0975-3710 & E-ISSN: 0975-9107, Volume 9, Issue 23, pp.-4267-4271.

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Academic Editor / Reviewer: Mukesh Kumar, Bakshi Sankar

Introduction

India is the largest producer and consumer of pulses occupying 33 % of the world's area and 22 % of the production [28]. Pulses are a rich source of vegetative protein and are fairly drought tolerant due to their deep root system. These are also ideal for intercropping and many of them are short duration crops as well as for multiple cropping systems. The latest estimate indicates that the present production of pulses has reached 14.7 MT with the productivity of 637 kg/ha although the pulse requirement by the year 2030 (32 MT) that is double the present production [1]. Mungbean is a short duration, herbaceous, annual, self-pollinated legume pulse crop under family Fabaceae, through symbiotic nitrogen fixation this leguminous crops have the capacity to fix atmospheric nitrogen [19]. It is used as green fodder or green manure, fits well in various multiple and intercropping systems. It is grown in summer and Kharif season in northern India and in southern India [29]. Mungbean is an excellent source of protein (24.5 percent) with a high quality of lysine (460 mg/g N) and tryptophan (60mg/g N). It has also a remarkable quantity of ascorbic acid when sprouted and also contains riboflavin (0.21 mg/ 100 g) and minerals (3.84 g/100g) [13]. Cereals, on the other hand, are rich in both these amino acids but deficient in lysine which is available in pulses. Consumption of Mungbean along with cereals supplement each other's deficiency and protein rich food with a balanced amino acid profile may be obtained. It is an excellent source of proteins considered as a poor men's protein [18]. Areas for cereals and other pulse have decreased, that for Mungbean has doubled in the last two decades with an annual rate of 2.5 %. The total area and production under the green gram in India were about 3.44mha and 1.20mt and productivity were 351 kg ha⁻¹. Mungbean is the third most important pulse crop of India because nearly 8% of this area is occupied in terms of area cultivated and production next to gram and pigeon pea [23].

On a world scale, almost 90 % of Mungbean production is produced in Asia, with India. Mungbean biomass incorporation as green manure after grain harvest

considerably improved the productivity of rice in rice-wheat system. Chopping Mungbean-straw and its incorporation gave highest available N (206–304 kg/ha) as compared to without chopping (170–195 kg available N). Mungbean average yield is very low (763.50kg ha⁻¹) as compared to its potential yield of 2-4 ton ha⁻¹. Rajasthan, Maharashtra, Andhra Pradesh, Karnataka, Orissa, and Bihar are the major Mungbean producing states [21]. Major constraints for poor productivity could be described to the lack of scientific efforts for bringing desired genetic improvement in this crop. Mungbean is vulnerable to about 26 diseases in the world [6]. The loss due to diseases to pulse crops has been estimated up to 44 %, depending upon the crop variety [5]. *Aspergillus flavus*, *Fusarium oxysporium*, *Penicillium crateriforme*, *Trichoderma harzianum*, *Alternaria alternate*, *Rhizoctonia solani*, *Cladosporium sphaerospermum*, *Curvularia lunata*, and *Aspergillus terreus* are most common fungi. Among the biotic factors, *Rhizoctonia solani* is a seed and soil borne disease [9] in pulse crops besides other agricultural and horticultural crops. The pathogen causes considerable yield loss in Mungbean [8], that commonly occurs in the tropics, sub-tropics and other warm temperate regions of the world causing root rot, stem rot, wilt and foot rot in almost all crops [4, 7, 10]. Allahabad is situated in Southern-Eastern part of the State Uttar Pradesh. It lies between the parallels of 24° 47' north latitude and 81° 19' east longitudes. The Ganga par and Jamuna par and the City comprise of Allahabad district. In this area, Mungbean is in 3532 area (ha), production is 2599(MT) and Productivity is 7.36 (q/ha). Pulses have occupied a focal attention in recent years due to increasing awareness and concern for sustainable production, food, and nutritional security. Pulses crops like Mungbean are now being introduced under intensive cropping systems to diversify the production systems and to bring sustainability in the cropping system in Mungbean crop losses due to disease is estimated to be 20-45% [3]. Among, fungal incidence causes more than 35-55% yield loss on Mungbean plant. More emphasis should be given to disease management of the crops in spite of the importance it renders to the masses.

Materials and Methods

Collection of soil sample

Total Fifty soil samples were collected from the rhizosphere of Mungbean crop, from the growing areas of Ganga par and Jamuna par in Allahabad region. Soil samples (50g each) were collected from four corners and the center of the field. The samples were collected in polyethylene bags with the help of auger and samples were sun dried. Before the collection of samples, auger was cleaned with sterilized water.

Isolation of fungal pathogens

Isolation of fungal pathogens was performed on potato dextrose Agar medium by serial dilution technique (at 10^{-3} , 10^{-4} & 10^{-5} dilutions). Plates were incubated at 25 ± 2 °C for four to five days. The pure culture of these fungi was prepared and maintained on PDA slants for further study.

Identification

Cultural and morphological characteristics- The cultural characteristics of different isolates of monoonidial origin were studied on potato dextrose Agar Media. The observations were made on basis of cultural and morphological characteristics. Culture character included colony color, texture, hyphae and morphological character included types of spores, size, shape, septation and arrangement of the spores. The measurement was done by using calibrated ocular micrometer [12].

Results

50 soil samples which are collected from the rhizosphere of infected Mungbean crop were found to have (78%) fungal incidence [Table-1] in the selective isolation process on selective media Potato dextrose Agar medium. Among 39 isolates, 6 isolates of *Aspergillus* sp.(25.6%) found to have high incidence on the Mungbean crop when compared to remaining isolates .the difference was found to be statistically significant ($P < 0.05$) [Table-1]

In the findings of [2] *Rhizoctonia solani* and *Fusarium* sp is observed which cause Root rot and wilt. In comparison to the incidence of 12 fungal isolates in the present study on Mungbean (*Vigna radiata*), 14 fungus isolates associated with Mungbean was reported in the study of [24].

In the present study higher incidence of *Aspergillus* sp was recorded which was in agreement with the studies of [26] [22].

As in the present study, 78% fungal incidences were observed which causes high yield losses in the Mungbean crop. To manage this fungal disease, farmers used commercial fungicides and indigenous method and a combination of both. Recommendation by the agriculture scientist was in line as reported in the study of [27].

In the present study, there is no incidence of *Macrophomina phaseolina* which was contrast to the studies of sheikh and [11,15] that *Macrophomina phaseolina* persisted in the soil in the form of black Sclerotia grows well at optimum temperature of 35°C and disease development was best at 30-35 °C.

Table-1 incidence of fungal isolates from rhizosphere soil of Mungbean crop

Soil Samples	Total incidence	Incidence (%)											
		<i>Aspergillus</i> sp.	<i>Penicillium</i> sp.	<i>Alternaria</i> sp.	<i>Cladosporium</i> sp.	<i>Curvularia</i> sp.	<i>Helminthosporium</i> sp.	<i>Mucor</i> sp.	<i>Rhizopus</i> sp.	<i>Trichoderma</i> sp.	<i>Chaetomium</i> sp.	<i>Rhizoctonia</i> sp.	<i>Fusarium</i> sp.
50	39(78%)	10(25.6)	2(5.12)	3(7.69)	4(10.25)	04(10.25)	2(5.12)	2(5.12)	2(5.12)	2(5.12)	4(10.25)	3(7.69)	1(2.56)

Values in parenthesis indicate percentage incidence: $X^2_{cal} = 18.5144 < X^2_{tab} (5\%) = 19.675$, *NS=non-significant

*Accepted – no significance difference between observed and expected values. Acceptance of null Hypothesis.

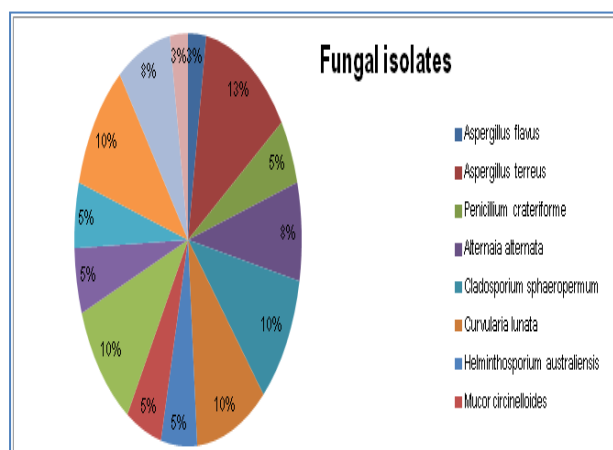


Fig-1 Incidence of different fungal species isolated from soil samples of Allahabad region

Identification of fungal isolates

Based on cultural characteristics and morphological characteristics the following isolates were isolated.

Summary and conclusion

Identification of fungal isolates on the basis of cultural and morphological characteristics revealed presence of *Aspergillus flavus* and *Fusarium oxysporum* (2.56%), *Penicillium crateriforme* and *Trichoderma harzianum* (5.12%), *Alternaria alternata* and *Rhizoctonia solani* (7.69%), *Cladosporium sphaerospermum* (10.25%), *Chaetomium globosum*, *Curvularia lunata*, *Aspergillus awamori* (10.25%) and *Aspergillus terreus* (12.82%) [Fig-1]

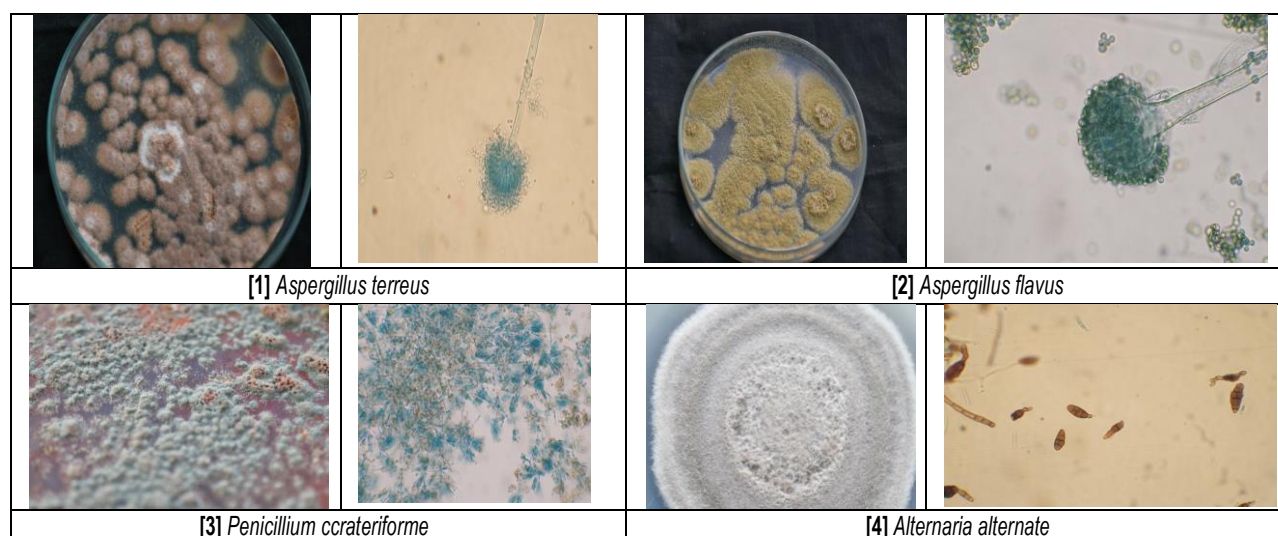
In the present study, rhizosphere soils are rich in microbial population, which leads to increases microbial activity. The increased number can be attributed to the enriched effect of root exudates, sloughed off cells leading to increased microbial activity that is characteristic to rhizosphere zone .the highly dynamic nature of the rhizosphere is reported by Katznelson, 1965. It is well known that root exudates have direct influence on the microbial population in the rhizosphere as reported by [20].

Conclusion

78% of fungal incidence on Mungbean was recorded during this study. Soil properties can modify the influence of fungi on plants and management practices that modify soil properties could be used to maximize the beneficial effects of inoculants. By Application of organic manures to the soil and adaption of crop rotation resident antagonistic organisms are activated which results in reduction of disease incidence and also have beneficial effect on plant growth. The fungi, *Aspergillus* sp, *Penicillium* sp, *Mucor* sp , *Rhizopus* sp and *Trichoderma* sp isolated from rhizosphere doesn't cause more yield loss to the Mungbean crop, but were found to have inhibitory effect on the pathogenic organisms like *Fusarium* spect[13].The fungi, *Cladosporium* sp, *Curvularia* sp and *Chaetomium* sp cause leaf spot, *Alternaria* sp causes a black spot, *Helminthosporium* sp causes leaf crown and root diseases, *Rhizoctonia* sp causes root rot and *Fusarium* sp causes wilt diseases which are important in yield loss and low productivity. Fungal incidence causes more than 35-55% yield loss on Mungbean plant. More emphasis is needed on disease management of this crop as this field lacks attention in spite of the importance it renders to the masses. To combat such problems, there is a need of proper fungal diagnosis program. To identify fungi, Conventional methods have often relied on identification of disease symptoms, isolation and culturing of environmental organisms, and laboratory identification by morphology tests.

Table-2 Cultural and Morphological characteristics of isolated cultures

Cultural characteristics			Morphological characteristics					Organism
			Spores					
Color	Texture	Hyphae	Type	Size	Shape	Septation	Arrangement	
[1] Brownish black powdery	smooth	Aerial hyphae	Ascospores	Conidiophore varying 112.5-204.9µ×5-14.3µ. Vesicle upto 9-17µ×7-16.5µ. Conidial head 13-27µ×20-67.5µ	Vesicle globose	Septate or non septate	spores in Clustering	<i>Aspergillus terreus</i>
[2] Yellow basal with light green colour	Powdery smooth	Aerial hyphae	Ascospores	Conidiophore varying 990-1170×6-20µ. Vesicle upto 28-35×14-30µ. Conidial head 45-58µ×46-58µ	Vesicle globose and flattened at the apical part	Septate or non septate	spores in Clustering	<i>Aspergillus flavus</i>
[3] Dark green with orange basal	Velvety	decumbent	Ascospores	Conidiophores ranging about 69-124×3-13µ. Phialides upto 9-14×2-4µ.	Conidia shape partly globose and smooth	aseptate	Chain of single-celled conidia	<i>Penicillium ccrateriforme</i>
[4] Light olive green to brown	Smooth or verrucose	Branched hyphae	chlamydospores	Hyphae diameter: 5.25-8.6µ. Conidia about 25.5-46.75 µ×15-17.75µ.	Club- shaped spores single or form chains	Alternate septate	Conidia borne in long chain	<i>Alternaria alternata</i>
[5] Greenish black, lower brown colour.	Turf matted	Erect branched	chlamydospores	Conidiophore upto 100-150µ×3-15µ	Globose	septate	Chain of spores	<i>Cladosporium herbarium</i>
[6] Dark olive-gray greenish	Sub floccose ,woolly colonies	Dematiaceous hyphae	Ascospores	Conidiophores are thread like. Conidia 25-30µ×15µ. 5-5.25µ thickness at base. Thickness at apex 6-8.75µ. 3-5 septa	Conidia round at apex, often attenuated at the base	Septate	Branched conidia	<i>Curvularia lunata</i>
[7] White to dark purple	Felty woolly	Aerial mycelium	chlamydospores	Conidia 21.6-28µ×3.75-6µ	Spindle or sickle shape	Septate	Conidia scattered	<i>Fusarium oxysporum</i>
[8] Brown to black	Velvety	Aerial mycelium	conidia	Conidia about 32.5-37.45µ×6-7.5µ. conidia thickness 5-5.5µ	Conidia terminal or lateral on the geniculation, elongated, cylindrical.	Septate	Conidia terminal or lateral on the geniculation	<i>Helminthosporium australiensis</i>
[9] Short truf, deep brown –black	velvety	More or less branched in sympodia with branches alternating right and left	Zygospores, chlamydospores	Sporangiophores ranging 117-203µ×3.75-6.25µ. Columellae 40-85µ×45-85µ. spore 4-5µ×3µ. Zygospore :66-86µ	Columellae oval or pear- shape	Septate	Spores globose or elliptical	<i>Mucor circinelloides</i>
[10] White at young turns ochre- yellow.	cottony	More or less branched in sympodia branches alternating right and left	zygospores round or oval or angular	Sporangiophores about 561-716µ×18-24µ. Columellae up to 70-90µ×50-176µ. spore: 3-6µ×7.5-8µ. internodal length 3-6µ	Columellae oval or pear- shape	septate	Spores globose or elliptical	<i>Rhizopus stolonifer</i>
[11] Grayish green lower black	Horny fleshy	Sclerotial hyphae is short celled branching right angle	Basidia with 6-8 sterigmata. Barrel shape	Hyphae is 4.5- 14 µ in diameter, basidia 12-18µ×8-11µ	Spores are ellipsoid or oblong	septate	Spores scattered or clusters	<i>Rhizoctonia solani</i>
[12] Green or white – grey	Woolly smooth	Sporangia with smooth thin walled.	Zoospore Ellipsoid, 3.5×4.8µ	Sporangia 140µ×60µ. resting spore 13µ×17µ, gametophytes 106µ	Spores are spherical	septate	Spores scattered or clusters	<i>Sphaerocladia sps</i>
[13] Green lower side yellow	floccose	Vegetative aerial hyphae hyaline. di or trichotomously branched	conidia	Conidiophore 52-120µ×7.5-11.5µ. conidial head 26.5-28µ, spore 3-6µ	Spores are spherical or oval	septate	Spores scattered or clusters	<i>Trichoderma harzianum</i>



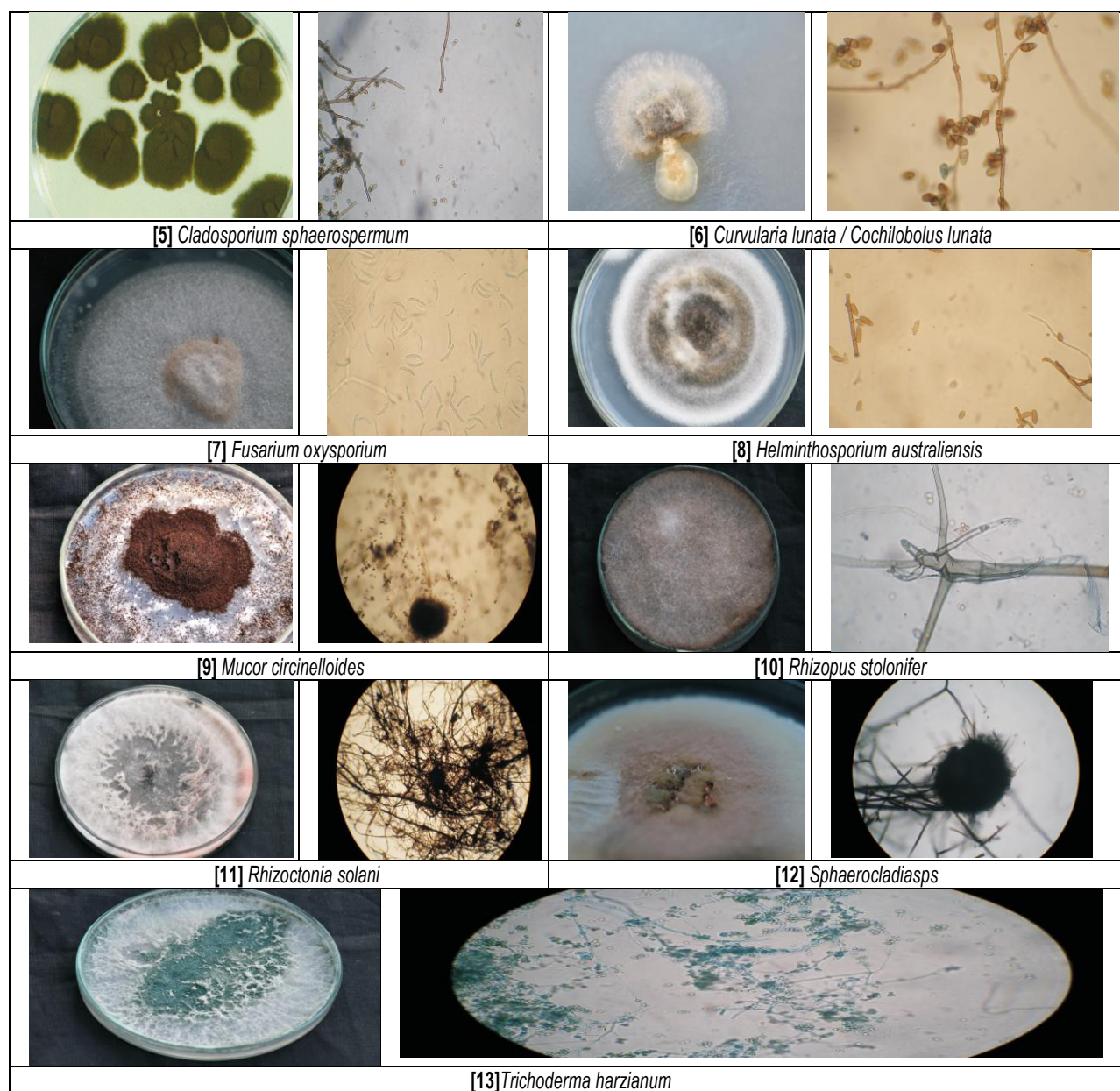


Fig-2 Cultures grown on PDA and observed under microscope at 40X.

Acknowledgement

I would like to express my sincere gratitude to all of them. First of all, I am extremely grateful to my research guide, Dr. Rubina Lawrence, Head, Department of Microbiology and Fermentation Technology, SHIATS, Allahabad. For her valuable guidance, scholarly inputs and consistent encouragement I received throughout the research work. This feat was possible only because of the unconditional support provided by Madam. I wish to express my sincere gratitude to my co-advisor Dr. Ebenezer Jeyakumar is a person with an amicable and positive disposition, Sir has always made himself available to clarify my doubts despite his busy schedules and to the encouragement throughout the time of research work. Thank you, Madam and Sir, for all your help and support. I was very much privileged to learn from my dear friend, Vijaya Kumar S who consistently extended his research expertise throughout my work and I owe a lot to him for this achievement. He has been very kind and patient and always willing to lend his service whenever I approached him and I acknowledge and appreciate him for all his efforts. Thank you, friend for your concern and good wishes. Overall thanks to God who, by His grace, has seen me through to the successful completion of this programme.

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.

List of Abbreviations

cal	calculated
cm	centimeter
et al	And others
ect	Many more
fig	figure
g	grams
ha	hectare
i.e.	That is
Kg/ha	Kilograms per hectare
μ	micron
μm	micrometer
μg	microgram
mg	milligram
mm	millimeter
MT	Million tons
q/ha	Quintal per hector
sp	species
S	Significance
tab	Tabulated
ton/ha	Tons per hectare

Author Contributions: All author equally contributed

Conflict of Interest: None declared

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