www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(2): 355-358

www.thepharmajournal.com
Received: 07-11-2022
Accepted: 14-12-2022

Dasharathe BG
M.Sc. Student, College of Agriculture, Dhule, MPKV, Rahuri, Maharashtra, India

Deore PB
Assistant Professor, College of Agriculture, Dhule, MPKV, Rahuri, Maharashtra, India

MN Agam
M.Sc. Student, College of Agriculture, Dhule, MPKV, Rahuri, Maharashtra, India

Corresponding Author:
Dasharathe BG
M.Sc. Student, College of Agriculture, Dhule, MPKV, Rahuri, Maharashtra, India

Variability of different isolates of stem rot (*Sclerotium rolfsii)* pathogen of groundnut from *Khandesh* region

Dasharathe BG, Deore PB and MN Agam

DOI: https://doi.org/10.22271/tpi.2023.v12.i2e.18440

Abstract
The experiment on "Effect of fungicides on *Sclerotium rolfsii* isolates of groundnut from *Khandesh* region" was conducted at Plant Pathology Section, College of Agriculture, Dhule. The results obtained are presented here under. Groundnut (*Arachis hypogea* L.) is a major edible oil seed crop of tropical and subtropical region of the world. It is the world's fourth important source of edible oil and third important source of vegetable protein also seeds, vines and dry fodder are excellent nutrient for cattle and *Rhizobium* bacterial root nodule provides nitrogen status of the soil. Isolates of *Sclerotium rolfsii* were isolated from stem rot infected groundnut samples collected from diverse geographic locations of *Khandesh* region of Maharashtra and pathogenicity of all the six isolates were proved.

Keywords: *Rhizobium, Sclerotium rolfsii, Arachis hypogea,* pathogenicity etc.

1. Introduction
Groundnut is believed to be the native of Brazil. The plant was introduced by Portuguese into Africa from where it was introduced into North America. It was introduced into India during the first half of the sixteenth century from one of the Pacific Islands of China, where it was introduced earlier from either Central America or South America (Jaisani, 2009) [8]. The major groundnut producing countries of the world are India, China, Nigeria, Senegal, *Sudan*, Burma and USA which occupied 18.9 million ha area with 17.8 million tons production of groundnut in the world, these countries account for 69% of the area and 70% of the production. The China is the highest groundnut producer in world followed by India. In India, groundnut is grown on 58.56 lakh hectares with production of about 82.64 lakh tons. The 58% of area and 82% of production are concentrated in Gujarat, Andhra Pradesh, Tamil Nadu and Karnataka. Andhra Pradesh, Tamil Nadu, Karnataka and Orissa have irrigated groundnut of about 6% of the total groundnut area in India. The Gujarat is the highest groundnut producer state in India and the highest productivity is recorded in Tamil Nadu (1.64 tonnes/ha). The total area in Maharashtra under groundnut is 2.38 lakh hectares with total production of 2.57 lakh tonnes and productivity is 1082 kg/ha. Summer Groundnut area in Maharashtra is 0.824 lakh ha with production 11.96 lakh tonnes and productivity 1451 kg/ha (Anonymous, 2011-12) [2]. Groundnut (*Arachis hypogea* L.) is a major edible oil seed crop of tropical and subtropical region of the world. It is the world's fourth important source of edible oil and third important source of vegetable protein also seeds, vines and dry fodder are excellent nutrient for cattle and *Rhizobium* bacterial root nodule provides nitrogen status of the soil. It is native of South America and belongs to annual legume group. It has an outstanding nutritive value with 40-50% oil, 25-30% protein, on equal weight basis, groundnut contains more protein than meat and about two half times more than eggs and 18% carbohydrates in addition to minerals *i.e.*, calcium, magnesium, iron and also vitamins like B_1, B_2, E and niacin. Groundnut cake is used as cattle and poultry feed. It is also good organic manure because of its high nitrogen content (7.0-8.0%) and other nutrients. Groundnut being a legume crop fixes a significant amount of nitrogen and improves the fertility status of the soil. It is high in calories, 5.6 calories per nut (calorific value of 567 per gram), its food value is: starch (11.55%), soluble sugar (4.5%) and moisture (6%). The low productivity in groundnut is attributed by many production constraints. Among these, biotic factors particularly diseases play a major role in limiting the yield of groundnut. This crop is known to be attacked by a number of fungal, bacterium and viral diseases.

The literature reveals that the yield losses caused by major fungal diseases like stem root, root rot, collar rot and pod rot, singly or in combination as high as 15-70% during both *kharif* and *rabi-summer* seasons (Ghewande *et al*., 1983, Subrahmanyam *et al*., 1984) [7, 12].

2. Materials and Methods

2.1 Laboratory instruments

Different laboratory instruments used during the course of investigator were autoclave, laminar air flow, BOD incubator, binocular research microscope, refrigerator, pH meter, electronic weighting balance, gas burner, spirit lamp, inoculating needles, forceps, blades, marker pen etc.

2.2 Glassware's

Different Borosil makes glassware *viz.,* Petri plates, test tube, glass slides, conical flasks and beakers of various capacities, glass rods, bowls, volumetric flasks, measuring cylinders of different capacities, pipettes, *etc.* were used.

2.3 Glassware cleaning

During the experimental studies, glassware were kept for a day in the cleaning solution containing 60 g potassium dichromate ($K_2Cr_2O_7$), 60 ml of concentrated sulphuric acid (H_2SO_4) dissolved in one lit of water. Then they were cleaned by washing with detergent soap solution followed by rinsing several times in tap water and finally with distilled water.

2.4 Sterilization

The glassware was sterilized in an autoclave at 1.1 kg/cm^2 pressure for 20 minutes and then they were kept in hot air oven at 60 °C for one hour. The media were sterilized at 1.1 kg/cm^2 pressure for 20 minutes. Soil used for pot culture experiments was sterilized in an autoclave at 1.1 kg/cm^2 for 2 h for two consecutive days.

2.5 Chemicals

The laboratory grade standard and pure chemicals were used for media preparation.

2.6 Collection of diseased samples and isolation of *Sclerotium rolfsii*: The groundnut plants showing the typical symptoms of stem rot (*Sclerotium rolfsii*) were collected from diverse geographic locations of *Khandesh* region and code numbers were allotted to each collected disease sample for further study (Table 1). The details are as follows.

Table 1: Locations of collection and codes of *Sclerotium rolfsii* isolates from *Khandesh* region

No.	Location/Place	Tahasil	District	Code No.
1.	Dhule (Agronomy Farm, College of Agriculture, Dhule)	Dhule	Dhule	Sr1
2.	Dhule (KVK, Dhule)	Dhule	Dhule	Sr2
3.	Chahardi	Chopada	Jalgaon	Sr3
4.	Gambhi (Farmer's field)	Bhusawal	Jalgaon	Sr4
5.	Belvia (Farmer's field)	Bhusawal	Jalgaon	Sr5
6.	Sanasgaon (Farmer's field)	Bhusawal	Jalgaon	Sr6

The groundnut plants infected with stem rot (*Sclerotium rolfsii)* was used for isolation. Isolations were made from the fresh diseased groundnut plant samples collected from various locations. The isolation of fungus was done from infected groundnut portion by following standard tissue isolation procedure.

The infected samples were cut into small bits and washed in running water. Then, bits were surface sterilized with 0.1% mercuric chloride ($Hgcl_2$) solution for one minute and washed thoroughly with sterile distilled water for three times to remove the traces of mercuric chloride. Then the surface sterilized sample bits were dried by placing gently in between the two sterile blotter paper and aseptically transferred to petriplates containing the sterilized PDA medium. The plates were incubated at 28±2 °C in BOD incubator for three days. On fourth day, the fungal growth arose through the infected tissue was picked up by inoculation loop and transferred aseptically to the PDA slants. The pure cultures of the fungus isolates were obtained by further growing the cultures and following hyphal tip culture method under aseptic conditions.

2.7 Pathogenicity

Sterilized soil was taken in plastic pots of size 11 x 8 cm. The sick soil was prepared by mixing *Sclerotium rolfsii* isolate cultures thoroughly with sterilized soil. Then, healthy surface sterilized two groundnut seeds (SB XI variety) were planted in each pots filled with sick soil and replicated thrice. The groundnut seeds planted in pots without *Sclerotium rolfsii* inoculum served as control. Throughout the growing period, the soil moisture was maintained at 25 per cent moisture holding capacity of soil by adding sterilized water. After 25 days of germination the plants showing the typical stem rot symptoms were observed. Reisolation of each isolate was made from such affected portion of the plant tissue and compared with that of original culture.

3. Results and Discussion

3.1 Collection and isolation of *Sclerotium rolfsii* isolates of groundnut

The groundnut plants showing the typical symptoms of stem rot (*Sclerotium rolfsii*) were collected from diverse geographic locations of Khandesh region of Maharashtra. The isolation of *Sclerotium rolfsii* was done from the infected samples (Plate 3). The Sclerotium rolfsii pathogen was identified based on following morphological and growth characters.

The fungus produced white, dense radiating mycelial growth on PDA. In early stages of growth, the mycelium was silky white, it gradually lost its luster and became dull in appearance. The sclerotia initiation was started from 6th day onwards. At initial stages, the sclerotial bodies were white but later they turned into buff brown colour and then to chocolate brown at maturity. Matured sclerotia were spherical to ellipsoidal. The fungus isolates isolated from the affected groundnut samples were compared with the original description and were found resembling with Sclerotium rolfsii in morphological characters. These isolates constituted the experimental results of present investigation.

3.2 Symptomatology

The *Sclerotium rolfsii* infected plants showed characteristics yellowing of leaves followed by loss of vigor. Infected plants were easily pulled out from the soil. The whitish mycelial growth was cropping around the collar region, stems as well as the roots. On stem, white to creamy coloured, round to spherical sclerotial bodies were seen (Plate 1). Heavily infected stems revealed the characteristics soft rot or decaying of tissue.

3.3 Pathogenicity: The Pathogenicity of isolates obtained from different location stem rot samples were proved by sick soil technique. The observations were recorded regularly after germination. The hundred percent seed germination and good growth of groundnut seedlings were observed in un-inoculated control treatment. While, delayed seed germination and seed rot was observed in treatments treated with individual isolates of *Sclerotium rolfsii*. The infected plants showed poor growth and rotting at collar region which were easily pulled out from the soil. The white radiating mycelium was creeping over the crown and also at the root region. In the later stages of infection, light to deep brown, spherical or round sclerotial bodies were formed in abundant adhering around the collar region. Later, the seedlings were seen exhibiting rotting appearance which finally dried (Plate 2). All the isolates were reisolated and compared with the original cultures. All the *Sclerotium rolfsii* isolates satisfied the Koch's postulates.

3.4 Collection and isolation of Sclerotium rolfsii isolates of groundnut: Isolates of *Sclerotium rolfsii* were isolated from stem rot infected groundnut samples collected from diverse geographic locations of *Khandesh* region of Maharashtra. The isolates of the fungus were confirmed as *Sclerotium rolfsii* by comparing with the characters described by Mundkur (1934) [9].

Plate 1: Symptoms of stem rot (*Sclerotium rolfsii)* of groundnut

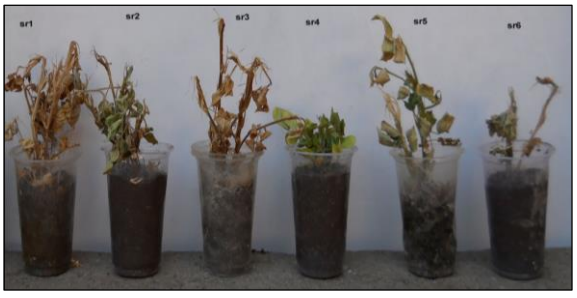

Plate 2: Pathogenicity test of *Sclerotium rolfsii* isolates of Groundnut

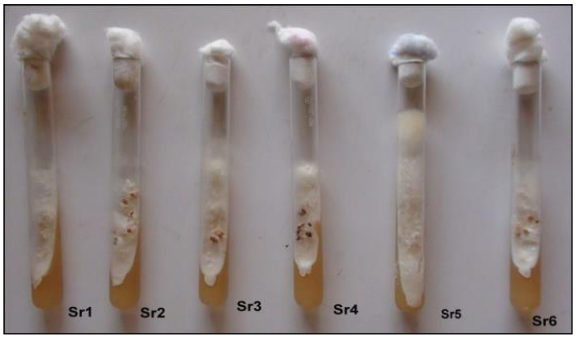

Plate 3: Isolates of *Sclerotium rolfsii*

3.5 Pathogenicity: The pathogenicity test of six isolates were carried out by sick soil technique. The diseased symptoms were characterized by seed rot, pre and post emergence death of seedlings. The young plants showed yellowing of leaves followed by premature death. On older plants brown lesions were noticed. Subsequently, such lesions exhibited profuse white mycelial growth. Later, there was production of sclerotial bodies on infested portion and such lesions on maturity turned brown. As the disease advanced, wilting of plant took place. Similar symptoms were observed by Beattie (1954) [5], Wheeler (1969) [13], Siddaramaiah *et al*., (1979) [11], Baruah *et al*., (1980) [4], Nyvall (1989) [10] and Ansari (2005) [3]. Hence, studies revealed that all the isolates of *Sclerotium rolfsii* were pathogenic to groundnut. All the isolates were reisolated and compared with the original cultures. All six *Sclerotium rolfsii* isolates satisfied the Koch's postulates.

4. Summary and Conclusions

Among the diseases of groundnut, particularly stem rot caused by *Sclerotium rolfsii* has become serious in recent years causing economic yield loss up to 80 per cent in severely infected fields. The less work has been done on the different isolates of *Sclerotium rolfsii* to different fungicides by poison food technique. Isolates of *Sclerotium rolfsii* were isolated from stem rot infected groundnut samples collected from diverse geographic locations of *Khandesh* region of Maharashtra and pathogenicity of all the six isolates were proved.

5. Acknowledgement

The authors are thankful to the Department of Plant Pathology, College of Agriculture, Dhule, MPKV, Rahuri, for providing the required experimental instruments for smooth conduct of the research work and providing the necessary facilities for the studies. The support and encouragement of the guide and research committee members, during the course of present investigation is gratefully acknowledged.

6. Conflict of Interest: None.

7. References

1. Anonymous. Distribution, area and production; c2010. http://www.krishi world.com.
2. Anonymous. Groundnut area, productivity and production, USDA Foreign Agricultural Service; c2011. Website: www.fas.usda.gov/wap/current/
3. Ansari MM. Growth survival, perpetuation and pathogenic variability of *Sclerotium rolfsii*, a polyphagous pathogen: A review. Journal of Oilseeds Research. 2005;22(2):240-244.
4. Baruah HK, Baruah P, Baruah A. Textbook of Plant Pathology, Oxford and IBH, Publishing Co. New Delhi; c1980. p. 498.
5. Beattie JH. Growing peanuts. United States Department Agriculture. Farmers Bullettin; c1954. p. 552.
6. Bindu M, Bhattiprolu SL. Integrated disease management of dry root rot of chilli incited by *Sclerotium rolfsii* (Sacc.) International Journal of Plant, Animal and Environmental Sciences. 2011;1(2):31-37.
7. Ghewande MP, Pandey RN, Shukla AK, Misrad DP. A new southern blight disease of Groundnut caused by *Sclerotium rolfsii* Sacc. Current Science. 1983;16:845-847.

8. Jaisani H. Geographical origin of groundnut; c2009. http://www.krishiworld.com/html/com_crops1.html.
9. Mundkur BB. Perfect stage of *Sclerotium rolfsii* Sacc. In pure culture. The Indian Journal of Agricultural Sciences. 1934;4:779-782.
10. Nyvall RF. Field crop Diseases Handbook, Second Edition, Published by Van Nartrand Renhold, New York, U.S.A.; c1989. p. 817.
11. Siddaramaiah AL, Krishnaprasad KS, Shivaram BN. Laboratory evaluation of fungicides against *Sclerotium rolfsii* Sacc. Causing foot rot of groundnut. Pesticides. 1979;13(11):31-32.
12. Subrahmanyam P, Williams JH, McDonald D, Gibbones RW. Studies on Sclerotial root rot disease of Groundnut caused by *Sclerotium rolfsii* Sacc. Madras Agricultural Journal. 1964;51:367-378.
13. Wheeler BEJ. An introduction to plant diseases. Published by the English language Book Society and John Wiley and Sons Limited., London; c1969. p. 374.