

# Effect of Age on Susceptibility of Groundnut Plants to *Sclerotium rolfsii* Sacc. Caused Stem Rot Disease

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## Abstract

Stem rot of groundnut caused by *Sclerotium rolfsii* (Sacc.) is a soil borne disease favoured in humid and warmer soil condition at all growth stages. Our objective was to determine how plants ages affect susceptibility of plants exposed to *Sclerotium rolfsii*. Groundnut seeds were grown in pots containing sterilized soil. Groundnut plants were inoculated 0, 15, 30, 45 and 60 days after sowing (DAS) by actively mycelium and sclerotia developed on sorghum grains placed near the seeds/plants. Stem rot developed in all inoculated plants but severity decreased with increasing plant age at inoculation. Highest disease severity (79.04%) was recorded in 45 DAS inoculated plants. Whereas plants inoculated 0 DAS may cause pre-emergence rotting and few plants emerged. Plants were inoculated at 15, 30 and 60 DAS developed stem rot symptoms. Our findings suggest that plants are more susceptible to infection at early development stages (0-45 DAS). However, susceptibility to stem infection was reduced after 45 DAS of inoculation. Moreover, young stage of maturity was more susceptible to *S. rolfsii*.

**Keywords:** Groundnut; *Sclerotium rolfsii*; Stem rot; Disease severity; Plant ages

## Introduction

Groundnut (*Arachis hypogea* L.) is commonly called peanut, goober pea goober, pindad jack nut, manila nut, pygmy nut, pignut and monkey nut [1]. It is also known as 'king of oil seeds' [2]. It has wide range of cultivation tropical and subtropical countries in the world. Groundnut is an important oilseed crop of India, grown extensively in various parts of the country in both Kharif and Rabi/Summer seasons. Groundnut plants suffer from several diseases caused by fungi, viruses, bacteria and nematodes resulting yield losses. *Sclerotium rolfsii* Sacc. is a soil born pathogen have wide host range (>500) including agricultural and horticultural crops [3,4]. Groundnut plants infected by *S. rolfsii* caused stem rot, root rot, sclerotial wilt, [5] and stem and pod rot [6]. Stem rot also known as white mold or southern blight, is a devastating soil borne disease in the India. Stem rot has been observed, where moisture and temperature conditions are sufficiently high to allow the growth and survival of *S. rolfsii*. Groundnut plants were infected by *S. rolfsii* at all growth stage including the germinating stage of the seed causing pre-emergence rot and young plant shown stem rot. The time taken for wilting varied from 8 to 15 days. The younger plants were found more susceptible as the infection was more and rapid [7].

The stem and pod rot caused by *S. rolfsii* Sacc. is major constraints and potential to reduces summer groundnut production in South Gujarat region. The objective of this study was to determine how plants ages affect susceptibility of plants exposed to *Sclerotium rolfsii*.

## Materials and Methods

The stem rot pathogen *Sclerotium rolfsii* was isolated from tissue-segmented method from groundnut plants with typical showing stem rot symptoms collected from collected the Regional Rice Research Station N.A.U., Vyara farm and farmers field of Tapi district during 2015-2016.

Infected stem tissues were surface sterilized with 0.1% HgCl<sub>2</sub> (1 g/lit) for 1 minute followed by three subsequent washing with sterilized distilled water in aseptic condition. The sterilized pieces were then transferred aseptically under laminar airflow on sterilized Petri plates containing 20 ml potato dextrose Agar (PDA) medium. The Petri plates were incubated in biological oxygen demand (BOD) at 27°C to

2°C temperature for optimum growth. The fungal hyphae developing from the infected tissues were sub-cultured aseptically on PDA media containing in Petri plates. Thus, pure culture was obtained by hyphal tip method and microscopically examined for identification and it was further purified by using single sclerotial body. The culture was maintained on PDA slants for further investigations.

Identification of the pathogen causing stem rot of groundnut was carried out by studying the cultural and morphological characters were recorded right from initiation of mycelial growth till the period of 15 days. The morphological characters *viz.*, mycelia growth and sclerotial formation, its size, shape and colour were studied under low power magnification (10X) from 10 days old culture of *S. rolfsii* and were compared with identification key described in "Illustrated Genera of Imperfect Fungi" [8]. The pathogenicity test of the pathogen was also carried out in pots by stem inoculation technique as described [9].

## Preparation of inoculums

The pathogen *Sclerotium rolfsii* was multiplied on sorghum grains (200 g) soaked overnight in water for pot experiment. About 100 g of soaked sorghum grains were taken in 500 ml capacity saline bottles tightly plugged. The bottles were then sterilized for 20 min at 121°C. After sterilization the sorghum seeds in saline bottles were inoculated with 5 mm mycelial disc from 7-day-old pure culture of *S. rolfsii* at each bottle and bottles were incubated for a 15 days at 27°C ± 2°C for proper mycelial growth.

Experiment was conducted at Regional Rice Research Station, N.A.U., Vyara during the year 2015-2016 under pot condition. Five

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stages i.e. 0, 15, 30, 45 and 60 DAS of the groundnut plants were taken for their susceptible reaction against stem rot causal pathogen *S. rolfsii*. These stages of plants were maintained in the eighteen plastic pots of 15 × 30 cm diameter replicated in three times and filled with sterilized soil. In each pot 10 seeds of groundnut (cv. GJG-9) was shown and fertilizer dose applied as per recommended. After raising all the respective stages, the sorghum grain inoculums were added at near the stem up to 4-5 grain on each plants of groundnut. Inoculated pots were kept in open place for observation and the pots were irrigated as when required. Stem rot disease severity was made at 15, 30, 45, 60 and 75 days after inoculation at respective stages, number of plants showed typical symptoms i.e. stem rot, lesion of stem, weathering of leaf and dead plants due to *S. rolfsii* was observed and per cent disease incidence was calculated using formula [10] (Table 1).

$$\text{Disease incidence (\%)} = \frac{\text{No of infected plant}}{\text{Total no. of observed plants}} \times 100$$

Disease rating	Description
1	Healthy
2	Lesions on stem only
3	Up to 25% of the plant symptomatic (wilt, dead or dying)
4	26% to 50% of the plant symptomatic
5	>50% of the plant symptomatic

Disease severity (Ds) was calculated as [12]

Table 1: Symptoms on groundnut plants were observed as per 1-5 rating scale [11].

Sr. No	Treatments (Days)	Disease incidence	PDI*
1	0	100	25.71 (5.10) <sup>a</sup>
2	15	100	69.36 (8.35)
3	30	100	74.45 (8.64)
4	45	100	79.04 (8.89)
5	60	100	49.68 (7.06)
6	Control	0.00	0.00 (0.70)
S. Em. ±			0.29
C.D. at 5%			0.96
C.V. %			8.35

\*=Average of three replications. a=Figures in parentheses are the corresponding square root transformed values + 0.5 added

Table 2: Effect of age of groundnut plants on stem rot disease development in pot conditions.

$$\text{Disease severity} = \frac{\sum ab}{AK} \times 100$$

Where, a=No. of disease plants having the same degree of infection, b=Degree of infection, A=Total no. of examine plant, K=Highest degree of infection

## Result and Discussion

Stem rot fungal pathogen showed white fluffy mycelium growth appearance on PDA medium. Microscopic view of mycelium was hyaline, branched, compact with septet and clamp connection. Initially sclerotia formation was observed 4 days after incubation and continued till 7 day old, numerous round to oval, globose or irregular mustard seed like sclerotia were produced. Initially, white colored sclerotia were formed then their color changed from white to off-white, light brown and dark brown as they attained maturity within 10-12 days. However, dark brown and black coloured sclerotia survived for longer times. The change color of sclerotia might also be due to utilization/exhaustion of nutrients. Also, found that sclerotia of some pathogen showed shiny appearance due to presence of gummy material on surface. All the above morphological characteristics of fungus was identified as *Sclerotium rolfsii* Sacc. and further confirmed with identification key described in "Illustrated Genera of Imperfect Fungi" [8,11-17]. Proved pathogenicity on 15 days olds groundnut plants (cv. GJG-9) under pot conditions. 4 days after of inoculation, the first symptoms were observed as water soaked brown to dark brown spots at basal portion of plants. The leaves of infected plants gradually yellowing and dry up. The professed white cottony growth of the fungus was also observed near collar region of the plant. Mycelium on stem/soil produced naked mustard seed like white sclerotia, later become dark brown. The collar region was weakened by the pathogen which resulted in to withering and death of the plant. Re-isolation of pathogen was done from inoculate infected plants and proved pathogenic nature of fungus. Un-inoculated seedlings did not develop any symptoms

To find out the susceptible stage of the groundnut to stem rot disease development, an experiment was conducted in pot conditions (Figure 1). The results are presented in the Table 2 depicted in Plate-I. The results revealed that there was no difference in disease severity percentage among the different stage of plant. Fourty five day old plant had maximum 79.04% disease severity was recorded followed by



Figure 1: Effect of age of groundnut plants on stem rot disease development in pot conditions.

30 and 15 days old plants with 74.45% and 69.36% disease severity, respectively. Least disease severity was recorded in 0 days old plants with 25.71% whereas; 60 days old plant had 49.68% disease severity. However, few plants were emerged from inoculums incorporated with seeds at the time of sowing. It may due to low germination or plant emergence may due to production of organic acid by *S. rolfsii*, which are toxic to living cell. Therefore, this result was used to identified most susceptible stages to evaluation of genotypes under artificial conditions.

The present findings revealed that groundnut plants were infected by *S. rolfsii* at all stage of plant growth from seed germinating to harvesting. Germinating stage of the seed causing pre-emergence rot and the susceptibility of groundnut plants against *S. rolfsii* was decreased with the increase in the age of groundnut plants. Our finding described the young stage of maturity was more susceptible against *S. rolfsii*, groundnut plants were infected by *S. rolfsii* at all growth stage of the plant including the germinating stage of the seed causing pre-emergence rot [7]. The younger plants were found more susceptible as the infection was more and rapid. The time taken for wilting varied from 8 to 15 days. Disease severity was decreased as the age of plant increased. Maximum plant mortality due to *S. rolfsii* was recorded in 15 days old groundnut seedling followed by 30 days old plants [18]. They also found that least mortality was recorded in 105 days old plants and susceptibility of groundnut seedling to *S. rolfsii* decrease with the increase in the age of groundnut plants. 10 days old plants were more susceptible to collar rot infection (80.00%) followed by 15 days old groundnut plants (75.00%). Plant mortality was increased with the increase in age of plant from 5 days (30.00%) to 10 days (80.00%), but it decreased thereafter i.e. at 15 days (75.00%), 20 days (65.00%) and 25 days (37.50%) plants were mortile [19]. Moreover, susceptibility or resistance of plants to stem rot disease is often influenced by their age. The per cent plant killing increased with increased in age up to (5 to 10 days) but it was decreased beyond 15 days, also in chick pea plant [20] and peppermint [21]. Hence, further studies are in progress to manage this disease at early stage of the crop growth.

## Conclusion

Initially *S. rolfsii* appeared as white fluffy mycelium growth on PDA as well as around the basal portion of stem than it produced light brown and dark brown round to oval, globose or irregular mustard seed like sclerotia were produced. Groundnut plants were infected by *S. rolfsii* at all growth stages of plant from seed germinating to maturity. But the younger plants were found more susceptible to infection by *S. rolfsii* caused highest plant mortality results to reduced pod yield. Maximum (79.04%) disease severity was recorded in 45 days old groundnut plants whereas, 30 and 15 days old plants had 74.45% and 69.36% disease severity, respectively. Few plants emerged from inoculums was incorporated with seeds at the time of sowing. Low germination or plant emergence may due to production of organic acid by *S. rolfsii* which are toxic to living cell. Germinating stage of the seed causing pre-emergence rot and the susceptibility of groundnut plants against *S. rolfsii* was decreased with the increase in the age of groundnut plants.

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