



## Effect of Carbon and Nitrogen Sources on Growth and Sclerotial Development of *Sclerotinia sclerotiorum*

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**ABSTRACT:** *Sclerotinia sclerotiorum* in rapeseed-mustard is a soil borne, necrotrophic pathogen. In this study the effect of Carbon and nitrogen sources on growth and sclerotial development of *Sclerotinia sclerotiorum*. Among the nitrogen sources, the maximum mycelium growth was recorded in ammonium sulphate @2% at 3 and 5 DAI. The maximum number and size of sclerotia were obtained in ammonium nitrate @2%. Among the carbon sources, the maximum mycelium growth was recorded in sucrose @2%. The maximum number of sclerotia per plate was recorded in sucrose @2%. The maximum size of sclerotia was recorded in D-glucose @2%. The present study helped in determining the carbon and nitrogen sources favourable for growth and sclerotial development of the pathogen.

**Keywords:** *Sclerotinia sclerotiorum*, Carbon, Nitrogen, Ammonium sulphate, Sucrose

### INTRODUCTION

Rapeseed-mustard group of crops is among the oldest cultivated plants in human civilization. The crops of this group are cultivated all over the world but their cultivation is mainly confined to India, China, Canada, Germany, France, Australia, USA, etc. It is a group of oilseed crops which assumes the significance in Indian national economy by occupying the second position next to groundnut and is considered as a 'cash crop'. Biologically, the rapeseed-mustard plants belongs to the family *Brassicaceae* (Cruciferae) and under the genus *Brassica* with large number of species and sub species cultivated in India. This group is mainly constituted by *Brassica juncea*, *B. napas*, *B. rapa*, *B. carinata* and *Erucasativa*. In India, rapeseed-mustard crops are cultivated on an area of 6.69 million ha with a production of 10.11 million tones and productivity 1511kg/ha (Anon, 2021). Rajasthan, Madhya Pradesh, Haryana, Uttar Pradesh, West Bangal, Gujarat, Jharkhand and Assam etc. are major mustard growing states of the country. In Madhya Pradesh rapeseed-mustard crops are cultivated in an area about 0.77 million hectares with the production of 1.31 million tones and productivity 1713kg/ha. (Anon, 2021). Chambal and Gwalior division of Madhya Pradesh are the major rapeseed-mustard growing area of state as these two divisions jointly contribute more than 70 and 80 per cent share in area and production of these crops in the state respectively. The pathogen of Sclerotinia stem rot [*Sclerotinia sclerotiorum* (Lib) de Bary] is a soil borne, necrotrophic pathogen with worldwide distribution known to infect over 500 species of plants (Sharma *et al.*, 2015). It was earlier considered as minor problem in India but now it has become a serious

problem over the years in some parts of the country. In Madhya Pradesh Sclerotinia stem rot has occupied a key position among the diseases and become now a threat for the cultivation of mustard in several pockets of Gwalior and Chambal division. Where it is locally as Polio because of the symptoms shows like polio and death after podding stage. The initial infection and symptoms of sclerotinia stem rot visible 40-45 days after sowing. This fungus can cause systemic and aerial infection by myceliogenic and carpogenic germination. Large numbers of sclerotia are formed in soil on organic matter, on roots, on and inside the pith of stem in rapeseed-mustard crop, and serve as source of primary inoculum for the next season. Symptoms appear on stem, and on decay leaves as elongated water soaked spots. Small white bodies appear on the stem which later on covered by the cottony mycelial growth of *S. sclerotiorum* and white to black colour as hardened sclerotia develop. The lesions finally girdle the stem from where it breaks and the plant dries. Sometimes infection is restricted to a smaller area of pith, resulting in slow stunting of the plant and premature ripening (Kolte, 1988). The objective of this study was to determined the ability of *S. sclerotiorum* to use a Carbon and nitrogen sources on growth and sclerotial development of *Sclerotinia sclerotiorum*.

### MATERIAL AND METHODS

Six nitrogen and six carbon sources were evaluated for the growth and sclerotial development of *Sclerotinia sclerotiorum*. Richards's agar medium was used with six nitrogen sources *viz.*, ammonium sulphate@2%, potassium nitrate@2%, ammonium nitrate@2%, sodium nitrate@2%, peptone@2%, urea@2% and six

carbon sources viz., Dextose@2%, D-glucose@2%, starch@2%, sucrose@2%, lactose@2% and mannitol@2%. The medium without nitrogen and carbon sources served as control. The pathogen was inoculated at the centre of the plate by placing a 5-day-old 5 mm culture disc. The plates were kept in an incubator at 20±2°C and three replications were maintained for each medium and the diameter of mycelia growth of the fungus was measured at 3 and 5 days after inoculation (DAI) and Number of sclerotia per plate and size per sclerotia (mm) were recorded at 15<sup>th</sup> days after inoculation.

## RESULTS AND DISCUSSION

Among the six nitrogen sources, Ammonium sulphate@2% was significantly superior over the other tested nitrogen sources. The maximum growth was obtained in ammonium sulphate@2% (60.33 mm and 88.33 mm) followed by sodium nitrate@2% (53.33 mm and 81.33 mm), potassium nitrate@2% (42.33 mm and 72.67 mm) at 3 and 5 DAI. While minimum growth was obtained in peptone@2% (24.67 mm and 61.33 mm) at 3 and 5 DAI. The maximum number and size of sclerotia were obtained in ammonium nitrate @2% (11.33 and 4.00 mm) but it was statistically at par with ammonium sulphate @2% (9.33 and 3.67 mm), respectively. Among the six carbon sources, the maximum mycelium growth was obtained in sucrose@2% (63.33 mm and 82.67 mm) followed by starch@2% (57.00 mm and 75.00 mm), D-glucose@2% (49.00 mm and 72.00 mm) at 3 and 5 DAI. While

minimum mycelium growth mm was obtained in mannitol@2% (24.33 mm and 51.67), respectively. In respect mycelium growth Sucrose @2% was significantly superior over other tested carbon sources at 3 and 5 DAI. The maximum number of sclerotia was also formed in sucrose@2% (13.00/Plate) but it was statistically at par with starch @2%(12.33/Plate), D-glucose@2% (11.67/ Plate), Dextose@2% (10.00/Plate) and lactose @2% (9.33/Plate). However, it was superior over mannitol@2% (5.67/Plate). The maximum size of sclerotia was obtained in D-glucose@2% (4.33 mm) but it was statistically at par with sucrose @2% (4.00 mm). The present finding supported by Elgorban *et al.* (2014) reported that the mycelial growth of the *S. sclerotiorum* was increased when D-glucose and saccharose were added that produced. Whereas, L-alanine and L-arginin were the best nitrogen sources for the growth of pathogen. Krishnamoorthy *et al.* (2016) also reported that the effect of various carbon and nitrogen sources on the growth and sclerotial formation of this pathogen. Among the carbon sources, sucrose was found to promote rapid growth of the pathogen as well as sclerotial formation which was followed by starch and glucose. Among the nitrogen sources, ammonium sulphate promoted the rapid growth of the pathogen followed by potassium nitrate and sodium nitrate. Ayed *et al.* (2020) also found that the radial growth was optimum in ammonium chloride. *S. sclerotiorum* uses simple carbohydrates (glucose, mannitol, and sucrose) and converts them to oxalate (Marciano *et al.*, 1989; Maxwell and Lumsden 1970).

**Table 1: Influence of nitrogen sources on mycelial growth and sclerotial formation of *Sclerotinia sclerotiorum*.**

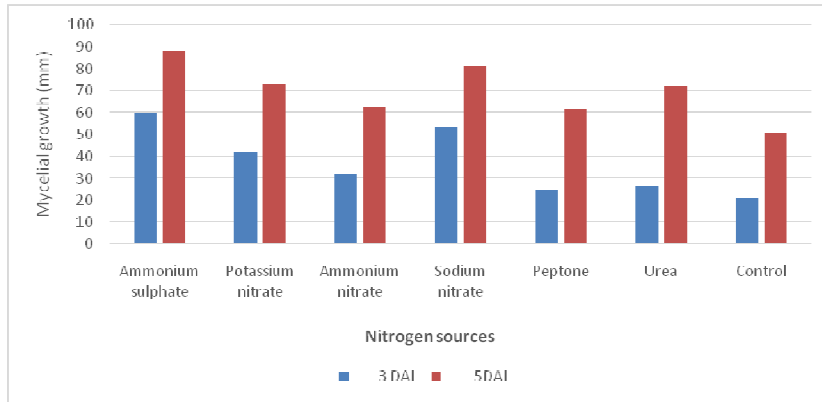
Nitrogen sources	Mycelium growth (mm)		Number of Sclerotia/Plate	Size of Sclerotia (mm)
	3 DAI	5DAI		
Ammonium sulphate @2%	60.33	88.33	9.33	3.67
Potassium nitrate @2%	42.33	72.67	5.33	3.00
Ammonium nitrate @2%	32.00	62.67	11.33	4.00
Sodium nitrate @2%	53.33	81.33	7.00	2.33
Peptone @2%	24.67	61.33	6.67	2.00
Urea @2%	26.33	72.33	3.67	3.33
Control	21.33	51.00	6.33	2.67
<b>Sem±</b>	<b>1.02</b>	<b>1.57</b>	<b>1.11</b>	<b>0.21</b>
<b>CD at 5%</b>	<b>3.13</b>	<b>4.81</b>	<b>3.40</b>	<b>0.65</b>

\*Data are the mean of three replication; \*DAI : Days after inoculation ; \*Sclerotial observation were carried out at 15<sup>th</sup> days after inoculation

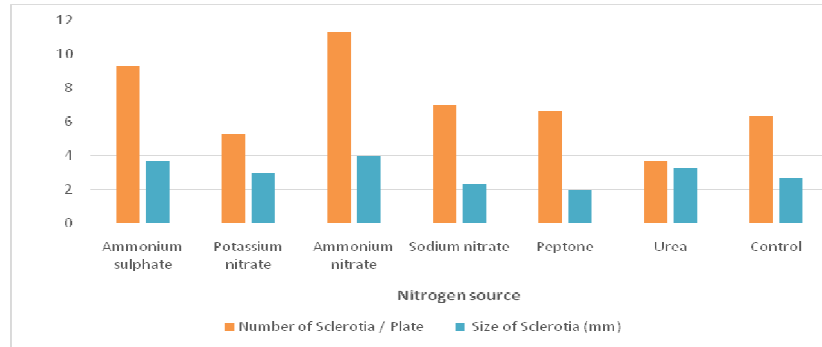
**Table 2: Effect of Carbon sources on mycelial growth and sclerotial development of *Sclerotinia sclerotiorum*.**

Carbon sources	Mycelium growth (mm)		Number of Sclerotia / Plate	Size of Sclerotia (mm)
	3 DAI	5DAI		
Dextose @2%	41.33	71.33	10.00	3.67
D-glucose @2%	49.00	72.00	11.67	4.33
Starch @2%	57.00	75.00	12.33	3.67
Sucrose @2%	63.33	82.67	13.00	4.00
Lactose @2%	36.33	64.67	9.33	3.00
Mannitol@2%	24.33	51.67	5.67	3.33
Control	22.67	45.33	7.33	3.33
<b>Sem±</b>	<b>1.49</b>	<b>1.37</b>	<b>1.50</b>	<b>0.19</b>
<b>CD at 5%</b>	<b>4.56</b>	<b>4.20</b>	<b>4.61</b>	<b>0.61</b>

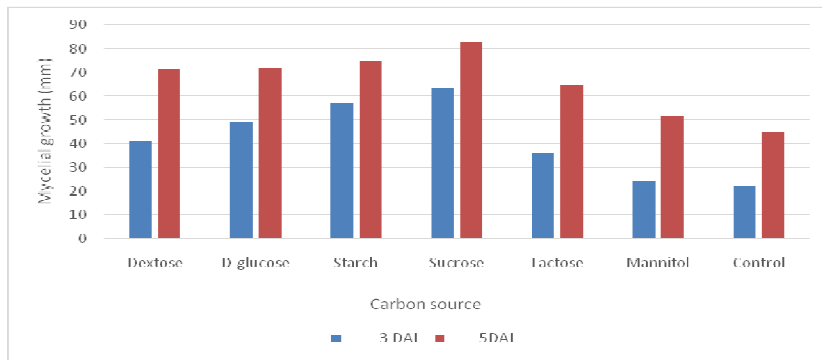
\*Data are the mean of three replication; \*DAI: Days after inoculation; \*Sclerotial observation were carried out at 15<sup>th</sup> days after inoculation



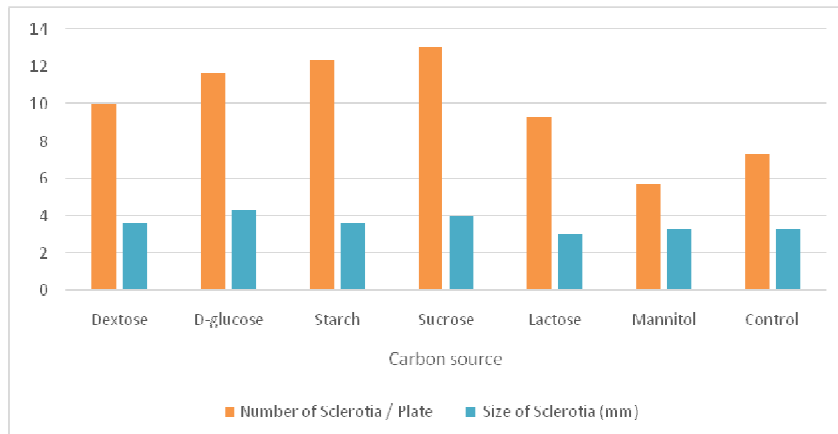
**Fig. 1.** Influence of nitrogen sources on mycelial growth of *Sclerotinia sclerotiorum*.



**Fig. 2.** Influence of nitrogen sources on sclerotial formation of *Sclerotinia sclerotiorum*.



**Fig. 3.** Effect of Carbon sources on Mycelial growth of *Sclerotinia sclerotiorum*.



**Fig. 4.** Effect of Carbon sources on sclerotial development of *Sclerotinia sclerotiorum*.

## CONCLUSIONS

Among the nitrogen sources, the maximum mycelium growth was recorded in ammonium sulphate@2% at 3 and 5 DAI. The maximum number and size of sclerotia were obtained in ammonium nitrate @2%. Among the carbon sources, the maximum mycelium growth was recorded in sucrose@2%. The maximum number of sclerotia per plate was recorded in sucrose@2%. The maximum size of sclerotia was recorded in D-glucose@2%. Thus, from the present study the nutritional requirement of *Sclerotinia sclerotiorum* affecting mustard crop was known.

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**Conflict of Interest.** None.

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