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Influence of nutritional sources on biomass production of *Sporothrix schenckii* isolated from *Momordica charantia*

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Abstract
Sporothrix schencki infesting the Bitter gourd (*Momordica charantia* L.) was collected from the research area to study the production of dry mycelial biomass. For the present study, Czapek-Dox solid and liquid media was used. For the physiological studies, 10 days old culture of *Sporothrix schencki* was used. The fungal pathogen was isolated on the Czapek-Dox solid media and then transferred in the test tubes slants. The dry mycelial biomass production was recorded for 10 days and it was observed that the maximum dry biomass was recorded on the 9th day i.e. 189 mg. The media was later inoculated with ten different carbon and nitrogen sources. It was observed that the maximum dry biomass production was recorded on galactose and control i.e. $NaNo_3$ while the minimum dry mycelial biomass was recorded on mannitol and $CO(NO_3)_2$.

Keywords: Sporotrix schenckii, bitter gourd, momordica charantia l, czapek-dox media, carbon source, nitrogen source

Introduction

Sporotrix belongs to the genus of saprophytic dimorphic fungi, and is found to grow on plant material and in the soil. *Momordica charantia* L. belonging to the family Cucurbitaceae and is commonly known as Bitter gourd. Abascal and Yarmell, 2005 and Raj et al., 2010 mentioned that bitter gourd is grown in countries that include India, China and Nepal. It is grown in number of states in India including Maharashtra, Gujrat, Rajasthan, Punjab, West Bengal, Orissa, Assam, Uttar Pradesh, etc. Bitter gourd is grown throughout Nashik district of Maharashtra.

Cucurbits prefer warm and humid climatic conditions (Bajpai. S., 2018). Temperature ranging between 24ºC to 27ºC is preferred by bitter gourd. It thrives on soil that is sandy loam or loam; well drained and rich in organic matter and a pH ranging between 6.0 to 7.0.

During the favourable conditions, the cucurbits are generally attacked by the fungi causing fungal diseases that ultimately result into loss yield (Thammaihraj. S. A et al., 2021). Tangonam 1999 and Begum et al, 2012 mentioned in their studies that one of the most important diseases of bitter gourd in Philippines is *Cercospora* leaf spot caused by *Cercospora citrulline* Cooke.

Mathew and Alice, 2002, Pandey et al. 2002, Palada and Change, 2003, Rai et al., 2008 and Khan et al., 2014 mentioned that bitter gourd is suspectable to various fungal diseases viz. Fungal root rots, damping off of seedlings, anthracnose, *Cercospora* leaf spot, Powdery mildew, downy mildew, *Phytophthora* blight, *Fusarium* wilt, *Phoma* blight, collar rot, white rot, etc.

One of the important foliar diseases of bitter gourd is downy mildew that is caused due to the fungal pathogen *Pseudoperonospora cubensis* (Kumar. V et al., 2018).

Balachandran, 2011 and Abraham et al., 2015 in their studies mentioned that considerable loss is caused due to the fungal diseases *Fusarium* wilt (Rakhollya et al., 2003), powdery mildew (Watson and Napier, 2009), downy mildew (Hansen, 2000 and Rai and Yadav, 2005), leaf curl (Raj et al., 2010) and leaf spot.

Bitter gourd infested by seed borne fungi were mentioned by Furukawa T et al.,2007 they stated the diseases such as White rot of fruit caused by *Sclerotium rolfsii*, Fruit rot caused by *Rizoctonia solani*, Powdery mildew caused by *Oidium sps*., Leaf spot caused by *Cercospora sps*., *Fusarium sps*. and *Penicillium purpurogenum*.

In a study carried out by Khan F M et al.,2014, the seed borne fungi of Bitter gourd included *Aspergillus flavus*, *Alternaria alternata*, *Rhizopus stolonifera*, *Aspergillus niger*, *Fusarium solani* and *Myrothecium roridum*.

Sultana Net al.,2009 in their studies conducted in Taiwan stated that *Alternaria alternata f. sp. Cucurbitae* was found to cause seedling blight in grafted seedlings of Bitter gourd.

Material and Methodology

Sporothrix schencki infecting the bitter gourd was collected from the research area and brought to the laboratory for the isolation procedure. The plant material that was infected was surface sterilized using 0.5% Sodium hypocholride to avoid contamination. The petri plates, glassware was sterilized in an autoclave at 15 psi pressure for 15 minutes. For the isolation of the fungal pathogen Czapek- Dox solid and liquid media was prepared and sterilized in the autoclave at 25 psi pressure for 15 minutes.

Under aseptic conditions, the pathogen was isolated on the Czapek-Dox solid media. The petri plates were neatly labelled and incubated at room temperature until good growth of the pathogen was observed. Later the pathogen was transferred on the Czapek-Dox slants in the test tubes.

The pathogen was isolated on the Czapek-Dox solid media to study the dry mycelial biomass production for 10 days. In order to study the effect of these carbon and nitrogen sources on the growth of the pathogen, the Czapek -Dox liquid media was incubated with 10 different carbon and nitrogen sources. After the incubation period of 8 to 10

days, the pathogen harvested using Whatman filter paper and then dried in the oven and the dry mycelial biomass was recorded on the different carbon and nitrogen sources.

Result and Discussion

From Janori region of Nashik district, Bitter gourd was collected and brought to the laboratory to study the dry mycelial biomass production of the pathogen *Sporotrix schenckii*.

The pathogen was isolated on the Czapek-Dox solid media to study the dry mycelial biomass production. During the incubation period of 10 days, it was observed that maximum dry mycelial biomass production was recorded on the 9th day i.e. 189 mg.

For studying the effect of different carbon sources on the dry mycelial biomass production the Czapek-Dox liquid media was inoculated with 10 different carbon sources. It was observed that the maximum dry mycelial biomass production was recorded on Galactose i.e. 306 mg followed by starch, glucose and dextrose. While minimum dry mycelial biomass production was recorded on Mannitol i.e. 228 mg followed by lactose, maltose and CMC.

Similarly, the Czapek-Dox liquid was also inoculated with 10 different nitrogen sources. The maximum dry mycelial biomass production was observed on Control i.e. 302 mg followed by Ca $(NO_3)_2$, $(NH_4)_2SO_4$ and NH_4NO_3. The minimum dry mycelial biomass production was recorded on $CO(NO_3)_2$ i.e. 225 mg followed by Ba $(NO_3)_2$, $CO(NH_2)_2$ and KNO_3.

Conclusion

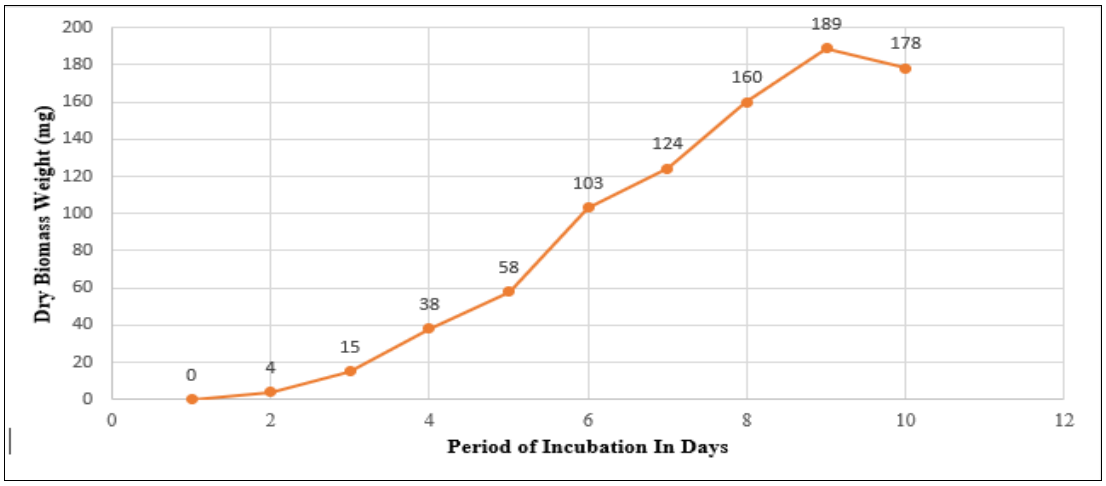
Growth of *Sporothrix schencki* on Bitter gourd was studied by inoculating the fungal pathogen on Czapek-Dox liquid media along with ten different carbon and nitrogen sources. There was a gradual increase in the dry mycelial biomass production, maximum dry mycelial biomass was recorded on the 9th day i.e. 189 mg and on the 10th day there was decrease in the dry mycelial biomass production.

When the Czapek-Dox, liquid media was inoculated with carbon source it was observed that maximum dry mycelial biomass production was recorded on galactose and minimum dry mycelial biomass was recorded on mannitol. When the Czapek-Dox, liquid media was inoculated with 10 different nitrogen sources, the maximum dry mycelial biomass was recorded on control and minimum dry mycelial biomass production was recorded on $CO(NO_3)_2$.

Observation Tables

Table 1: Growth of *Sporothrix schencki* grown on Czapek -Dox liquid media at different Incubation Periods

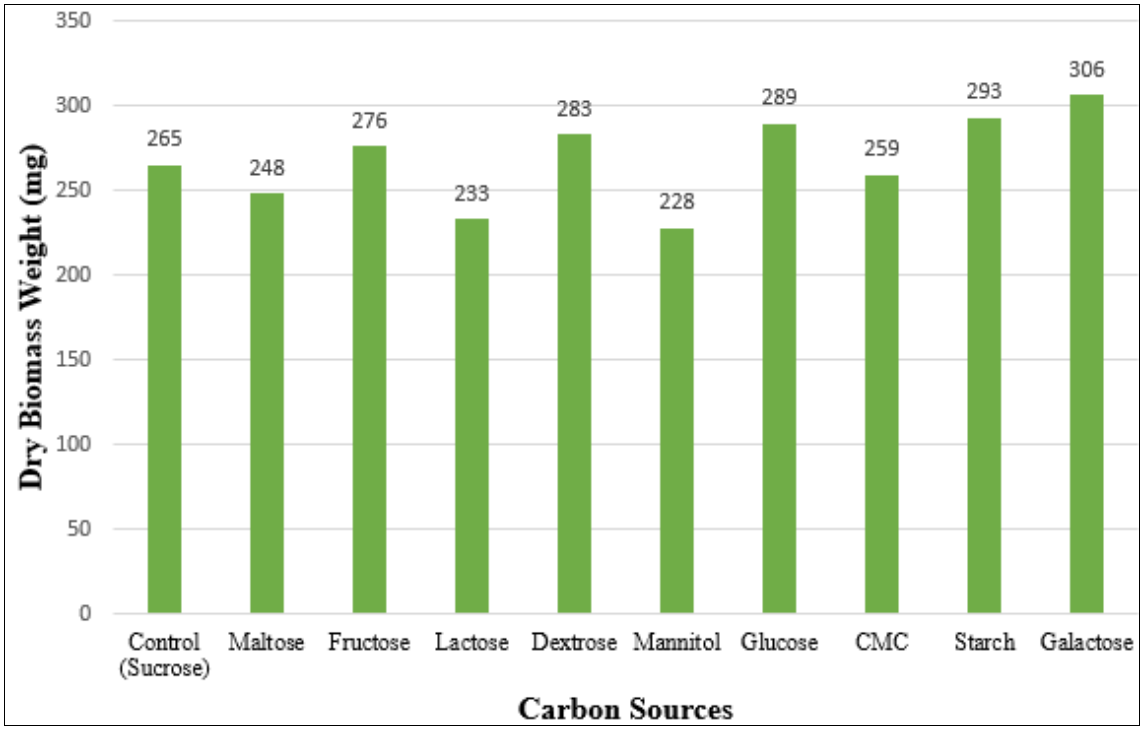
Period of Incubation Days	Dry Biomass Weight of *Sporothrix schencki* (mg)
1	0 mg
2	4 mg
3	15 mg
4	38 mg
5	58 mg
6	103 mg
7	124 mg
8	160 mg
9	189 mg
10	178 mg



Graph 1: Growth SPOROTHRIX SCHENCKII on Czapek- Dox liquid media at different Incubation period

Table 2: Growth of *Sporothrix schencki* grown on Czapek-Dox Liquid media Containing different carbon sources after 10 days of incubation

Ten Carbon Sources	Dry Biomass Weight of Sporothrix schencki (mg)
Control (Sucrose)	265 mg
Maltose	248 mg
Fructose	276 mg
Lactose	233 mg
Dextrose	283 mg
Mannitol	228 mg
Glucose	289 mg
CMC	259 mg
Starch	293 mg
Galactose	306 mg

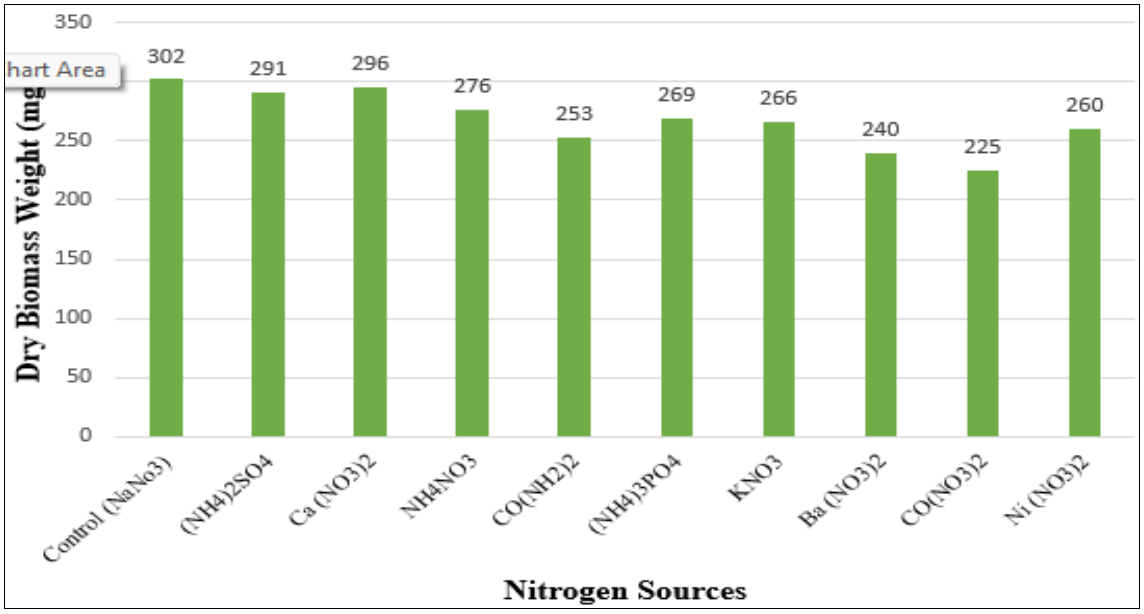


Graph 2: Growth of SPOROTHRIX SCHENCKII on Czapek- Dox liquid media containing different Carbon sources after 10 days of incubation

Table 3: Growth of *Sporothrix schencki* grown on Czapek-Dox liquid media

Ten Carbon Sources	Dry Biomass Weight of *Sporothrix schencki* (mg)
Control ($NaNo_3$)	302 mg
$(NH_4)_2SO_4$	291 mg
Ca $(NO_3)_2$	296 mg
NH_4NO_3	276 mg
$CO(NH_2)_2$	253 mg
$(NH_4)_3PO_4$	269 mg
KNO_3	266 mg
Ba $(NO_3)_2$	240 mg
$CO(NO_3)_2$	225 mg
Ni $(NO_3)_2$	260 mg

Containing different nitrogen sources after 10 days of incubation



Graph 3: Growth of SPOROTHRIX SCHENCKII on Czapek- Dox liquid media containing different Nitrogen sources after 10 days of incubation

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