

Original Research Article

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Evaluation of Culture Medium for the Growth of *Bipolaris sorokiniana* the Causal Agent of Spot Blotch of Wheat

K. Sinijadas*, Aparajita Dhar, Pulak Bhaumik and A.K. Chowdhury

Department of plant pathology, UBKV, Pundibari, West Bengal, India

*Corresponding author

ABSTRACT

Keywords

Evaluation, Culture medium, Colony morphology, *Bipolaris sorokiniana*

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Experiments were conducted to evaluate the effect of some culture media on the vegetative growth of *Bipolaris sorokiniana*. Five (5) treatment combinations each with three replications were employed. Potato Dextrose Agar (PDA) showed highest radial growth (6.7cm) after 9th day of observation followed by V8 juice agar (6.3cm). Oat Meal Agar showed lowest radial growth (4.6cm) for UBS-1 strain of *B. sorokiniana*. PDA, Czepek's-Doc Agar and Oat Meal Agar were showed blackish grey colony with irregular margins whereas V8 juice Agar and Leaf Decoction + PDA were showed whitish grey colony with regular margins.

Introduction

Wheat is the second most important food crop being next to rice in India, which contributes nearly one-third of the total food grain production. About one-tenth of the global production is contributed from India.

Wheat is susceptible to many diseases, Spot Blotch of wheat, one of the fungal foliar disease, which is the most prevalent disease in eastern parts of India. Spot blotch of wheat caused by the fungi *Bipolaris sorokiniana* (Sacc.) Shoem., in anamorph state (Saari, 1997). A key for distinguishing species of *Bipolaris* was described by Subhranian (1971). *B. sorokiniana* is characterized by thick-walled, elliptical conidia (60-120 µm

×12-20µm) with 5-9 cells. In axenic culture, the mycelium is composed of hyphae interwoven as a loose cottony mass and appears as white or light to dark grey depending on the isolates (Kumar *et al.*, 2002). These fungi are differentiated from the *Bipolaris* genus on the basis of morphological features of conidiophores and conidia.

On the leaf, lesions are due to anamorph *Bipolaris sorokiniana*, characterized by long multicellular spores, whereas the ascospores of *Cochliobolus sativus* are formed in pseudothecia developed on the wheat residue. The growth of microorganisms in an artificial medium is influenced by several physical and chemical factors. A nutrient material prepared for the growth of microorganisms in a

laboratory is called culture medium and the nutrient composition of a culture medium contribute a major role in microbial growth (Toratoro *et al.*, 1995) A critical and comprehensive knowledge of nutritional patterns and factor influencing the growth of fungi is prerequisite for any study leading to the understanding of host-pathogen relationship. Hence tested the different culture medium for the mycelia growth as well as colony characteristics of the *B.sorokiniana*

Materials and Methods

The different culture mediums (solid) were evaluated for obtaining maximum mycelial growth of *Bipolaris sorokiniana*. The experiment was conducted in complete randomized design with replicated 3 times. 5 solid medias viz., Potato Dextrose Agar, Oat meal Agar, Czapek's- Dox Agar, V8 juice Agar and Leaf decoction(wheat) + PDA. The culture mediums were prepared by the standardized method and autoclaved at 121.6 ° C, 15 psi pressure for 20 minutes.

Equal quantities of (20ml) of each medium were poured in 9 cm petri plate was inoculated separately with uniform 5mm mycelia culture bits cut with the help of cork borer from 7 days old culture of highly virulent strain of *Bipolaris sorokiniana* (UBS-1) were placed on the middle of petri plates filled with media. Plates were incubated at 27 ±1° C in BOD incubator and taken the observation. The diameter of the fungus growth was measured after inoculation of 5, 7, and 9 days on radial growth of mycelium.

Statistical Analysis

The experiments were done under controlled laboratory condition, and the data were analysed following completely randomized design (CRD) by using SPSS statistical software.

Results and Discussion

Radial growth of the pathogen

To find out the most effective culture media for the growth of *Bipolaris sorokiniana*, a total five culture media (solid) were evaluated again *B. sorokiniana* under in vitro condition and the data summarized in table 1, figure 1 reveals that Potato Dextrose Agar media was significantly superior over other tested media and followed by closely followed by V8 Juice Agar. The Oat Meal agar supported least growth. The fungus in PDA show fluffy blackish colony with higher number of Sporulation. The Wheat Leaf decoction media shows intermediate growth and sporulation. *Bipolaris* fungus showed better sporulation on PDA media supplemented with graminaceous plant extracts (Nasreen *et al.*, 2017)

These results corroborate the findings of former literatures such as Chattopadhyay and Dasgupta (1965) Found that production of conidia by *B. oryzae* was favoured by media either rich in or made exclusively of plant parts. Coleman *et al.*, (1990) showed that methionine and ethylene both reduced mycelium growth of *B. sorokiniana* when grown on Czapek's medium or on media containing grass leaf infusion.

The maximum mycelia growth on nine days after inoculation was recorded in PDA (6.7cm) followed by V8 juice agar (6.3cm), wheat leaf decoction + PDA (5.85cm) and Czapek's-Doc Agar (5.6cm) while minimum growth was recorded in Oat meal Agar (4.6cm). In case of statistical analysis the data shows highly significance with each other

Colony morphology of the Culture

Among 5 media tested were evaluated against *B. sorokiniana* under in vitro condition were data summarized under table 2.

Fig.1 Growth of CRP-473 on different culture media a) Czepek's-Doc Agar b) PDA c) Oat Meal Agar d) Leaf Decoction +PDA e) V8 juice Agar



Table.1 Radial growth of *B. sorokiniana* (UBS-1) in different growth medial at Different time interval

Sl. No	Solid culture medium	Radial growth of mycelia (cm)		
		5 DAS	7 DAS	9 DAS
1	PDA	4.65	5.45	6.7
2	V8 juice agar	4.15	5.45	6.3
3	Leaf decoction +PDA	4.45	5.2	5.85
4	Czepek's-Doc Agar	4.55	5.1	5.6
5	Oat meal Agar	3.75	4.35	4.6
SEm		0.0592	0.1176	0.2839
CD at 5%		0.1864	0.3706	0.8947

Table.2 Colony character of *B. sorokiniana* on different culture growth media

Sl. No	Solid culture media	Colony character	
		Colony colour	Margin of colony
1	PDA	slight fluffy blackish colony with whitish growth	irregular
2	V8 juice agar	fluffy whitish grey colony	regular
3	Leaf decoction +PDA	fluffy whitish colony	regular
4	Czapek's-Doc Agar	slight fluffy greyish black colony	irregular
5	Oat meal Agar	no fluffy growth greyish black colony	irregular

The PDA showed maximum radial growth with fluffy blackish colony with irregular margin. V8 juice agar was shown fluffy whitish grey colony with regular margin. Wheat leaf decoction PDA also showed fluffy whitish colony with regular margin but sporulation observed only at the margin of colony. Both Czapek's Doc and Oat meal Agar were showed fluffy greyish colony with irregular margin, but they differ only in the case of nature of colony. Oat meal agar showed no fluffy growth whereas Czapek's Doc showed slight fluffy growth. Colony morphology varied in five different culture media due to the difference in composition of medium as well as the response of pathogen towards that media.

In the present investigation, maximum radial growth of *Bipolaris sorokiniana* was observed in PDA medium and followed by V8 juice Agar and least shows in oat meal agar. In case of colony morphology PDA shows blackish colony with irregular margin and Leaf Decoction +PDA shows whitish colony with regular margin. That means colony morphology and radial growth of pathogen will be differ in different growth media due to their response towards medium contain components.

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