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Research Article

Screening of Effective Media for the Growth of *Pyricularia grisea* under *In-vitro* Condition

Ajay Singh Kaurav, Pramod Kumar Fatehpuria*, R.K. Pandya and Rajni Singh Sasode

Department of Plant Pathology, College of Agriculture, Rajmata Vijayaraje Scindia Krishi Vishwavidyalaya, Gwalior, (M.P.) 474002, India

*Corresponding Author E-mail: pramodfatehpuria@gmail.com

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ABSTRACT

Pearl millet [Pennisetum glaucoma (L.) R. Br.], belongs to family Poaceae (section Paniceae), is the world's hardiest warm-season cereal crop with the annual rainfall of 150 mm to 1000 mm. Maximum mycelial growth was recorded in potato dextrose carrot agar medium (86.33 mm) followed by Pearl millet Potato dextrose agar (83.67 mm), potato dextrose agar (78.33 mm), Pearl millet carrot dextrose agar (71.67 mm), Pearl millet potato agar (68.33 mm), Carrot dextrose agar (60.67 mm), Pearl millet grain potato dextrose agar (58.33 mm), Oat meal agar (51.67 mm), Pearl millet dextrose agar (51.67 mm), Calcium carbonate agar (42.67 mm), Malt agar (41.00 mm), Water agar (37.33 mm), Pearl millet grain dextrose agar (35.33 mm), Pearl millet agar (30.00) and Yeast extract agar (28.33 mm) while minimum growth was recorded in Pearl millet grain agar (27.33 mm).

Keywords: *Pearl millet, Pyricularia grisea, medium.*

INTRODUCTION

Pearl millet [*Pennisetum glaucum* (L.) R. Br.], belongs to family Poaceae (section Paniceae), is the world's hardiest warm-season cereal crop with the annual rainfall of 150 mm to 1000 mm. It is cultivated in over 30 countries of Asia, Africa and America where dry land system is possible. India and Africa are together occupying approximately 90 per cent area of total pearl millet in the world (Yadav et al., 2012). In India, it is popularly known as bajra grown for grain and fodder purpose and cultivated over an area of 6.98 million ha with the production of 8.06 million tones and the productivity is 1154 kg/ha (Anon, 2016). Madhya Pradesh occupies 0.27 million ha with an annual production 0.59 million tones and productivity is 2203 kg/ha (Anon, 2016). The important pearl millet growing states in the country are Rajasthan, Gujarat, Maharashtra, Uttar Pradesh, Tamil Nadu, Karnataka and Madhya Pradesh. It is being grown in Madhya Pradesh as sole crop. Rajasthan ranks first in the area and annual production.

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The major area of pearl millet cultivation in the state is confined to its northern region compressing Morena, Bhind, Gwalior, Datia, Sheopur and Shivpuri, districts. These districts jointly contribute more than 75 per cent share of the state in its area and production. It's grain is chiefly served as food, because of higher protein (27% - 32%), higher concentration of essential amino acids, twice the extract (fat) and higher gross energy than maize (Ejeta et al., 1987, & Davis et al., 2003). The crude protein content of green pearl millet forage varies from 6 to 20 per cent. The fresh forage is fairly well digested by ruminants, with DMD being about 66-69 per cent. In pearl millet silage, crude protein content is low (from 4% to 10%) due to protein losses, and the rumen degradable fiber fraction is low (Guimaraes et al., 2010).

Several diseases caused by fungi, bacteria, viruses and nematodes have been recorded (Rachie & Majmudar, 1980). Out of them downy mildew, blast, ergot, smut and rust are important. Blast incited by *Pyricularia grisea* (Cooke) Sacc. has become wide spread and destructive disease of pearl millet particularly in the crop cultivated for fodder purpose. The disease was first observed during 1952 at Kanpur, Uttar Pradesh (Mehta et al., 1953). More than 50 species of grass family are infected by blast pathogen including rice, wheat, barley, oats, pearl millet, finger millet and foxtail millet (Tanweer et al., 2015). The pathogen is highly variable in nature but certain strains are specific in their host range. Thus, *M. grisea* strains from pearl millet do not infect rice and *vice versa* (Mehta et al., 1953). The fungus can infect at all growth stages from seedling to adult plant thereby reducing grain yield as well as forage at varied quantity, sometimes extreme negativities (Lukose et al., 2007). The disease appears as grayish, water-soaked foliar lesions that enlarge and become necrotic, resulting in extensive chlorosis and premature drying of young leaves (Wilson et al., 1989). It causes light to dark brown, boat shaped lesions; appear on the lower surface of the leaf. At severe infection, leaves become completely dry. This disease becomes more severe during humid weather conditions especially with dense plant stands. Leaf blast on pearl millet has been found to be negatively correlated with forage yield, dry matter yield and digestive dry matter thus affecting the productivity and quality of the crop (Wilson & Gates, 1993). In the present scenario there is need to evaluate a suitable media most suitable medium for the growth and sporulation of the pathogen.it is need to have the clear understanding of the biology of the pathogen and to study the bast patho system.

METHODS AND MATERIALS

A total of sixteen culture media were evaluated against *p. grisea* replicated thrice and CRD. The Culture Mediums were prepared by the standardized method and autoclaved at 121.6 °C, 15 psi pressure for twenty minutes. Uniform quantities (20 ml) of each medium were poured in 90 mm Petri plates. Each Petri plate was inoculated separately with uniform mycelium culture bits (5 mm) cut with the help of cork borer from young (5 days) vigorously growing culture were placed on the middle of the each pre poured medium and incubated at 25±1oC (Dela Paz et al., 2006). Each treatment was replicated three times. The diameter of the growth of the fungus was measured after inoculation 3, 5 and 7 days on radial growth of mycelium.

RESULTS

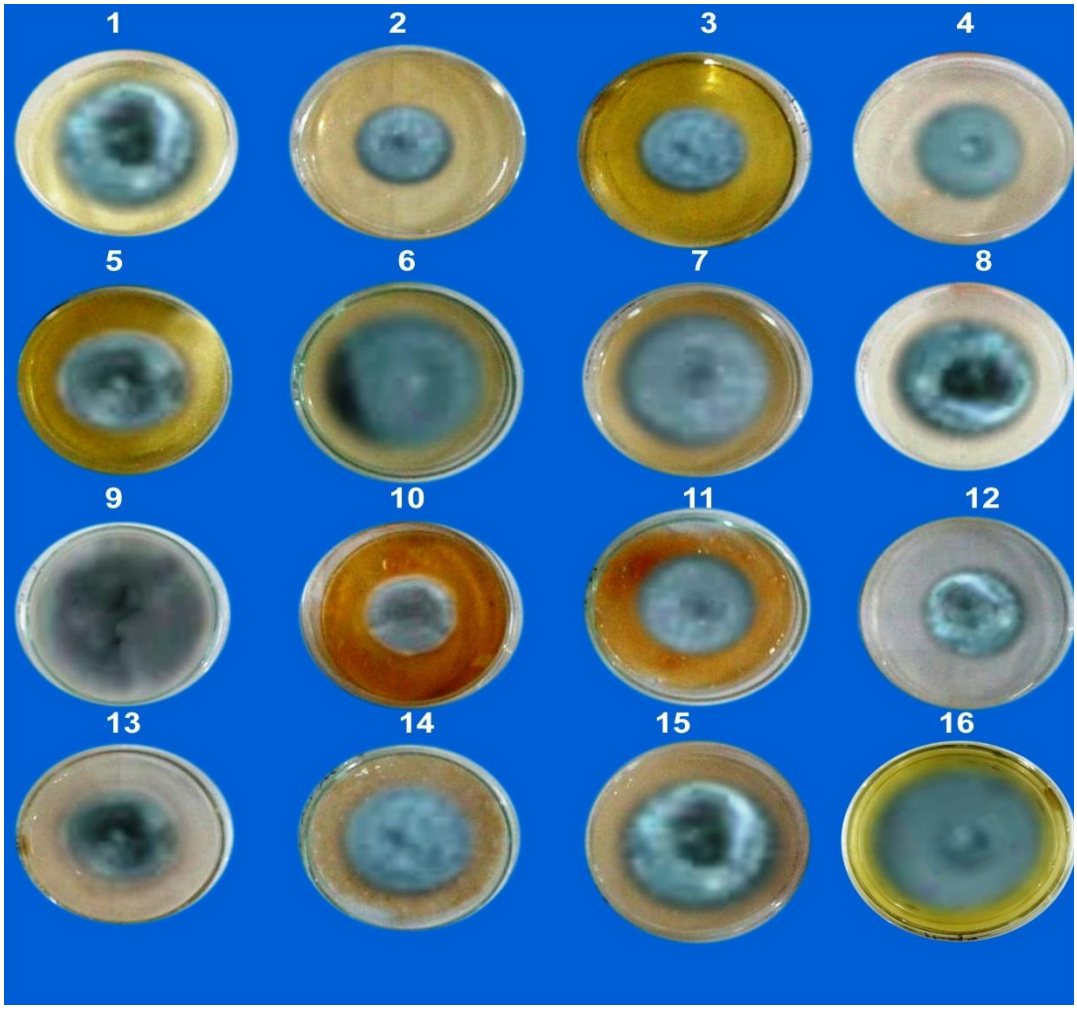
A total of sixteen culture media were evaluated for the growth of *pyricularia grisea* at three, five and seven days after inoculation and the data summarized in table-1 reveals that the maximum mycelial growth was recorded in potato dextrose carrot agar medium (86.33 mm) followed by Pearl millet Potato dextrose agar (83.67 mm), potato dextrose agar (78.33 mm), Pearl millet carrot dextrose agar (71.67 mm), Pearl millet potato agar (68.33 mm), Carrot dextrose agar (60.67 mm), Pearl millet grain potato dextrose agar (58.33 mm), Oat meal agar (51.67 mm), Pearl millet dextrose agar (51.67 mm), Calcium carbonate agar

(42.67 mm), Malt agar (41.00 mm), Water agar (37.33 mm), Pearl millet grain dextrose agar (35.33 mm), Pearl millet agar (30.00) and Yeast extract agar (28.33 mm) while minimum growth was recorded in Pearl millet grain agar (27.33 mm). The growth of *Pyricularia grisea* was significantly higher in potato dextrose carrot agar medium than the other tested medium except pearl millet potato dextrose agar medium where it was statistically at par. Potato dextrose carrot agar medium also showed maximum growth of the fungus at three and five days after inoculation (figure-2, plate 1).

Table 1: *In-vitro* evaluation of different media for the growth of *Pyricularia grisea*

S.No	Culture media	Mycelial growth (mm)*		
		3 DAI	5 DAI	7 DAI
1	Pearl millet potato agar	16.67	35.67	68.33
2	Pearl millet grain agar	9.33	17.33	27.33
3	Pearl millet agar	11.67	16.33	30.00
4	Pearl millet grain dextrose agar	12.00	18.33	35.33
5	Pearl millet dextrose agar	11.67	29.33	51.67
6	Pearl millet potato dextrose agar	19.00	44.33	83.67
7	Pearl millet carrot dextrose agar	17.33	39.33	71.67
8	Pearl millet grain potato dextrose agar	13.00	29.33	58.33
9	Potato dextrose carrot agar	21.67	46.33	86.33
10	Yeast extract agar	13.67	19.33	28.33
11	Malt agar	15.00	23.00	41.00
12	Water agar	14.67	24.33	37.33
13	Calciumcarbonate agar	16.67	26.00	42.67
14	Oat meal agar	16.00	28.33	51.67
15	Carrot dextrose agar	17.33	32.67	60.67
16	Potato dextrose agar (Control)	18.33	39.33	78.33
	SEm±	**1.17**	**1.426**	**1.470**
	CD at 5%	**3.385**	**4.128**	**4.253**

***Mean of three replications**



DISCUSSION

Out of sixteen culture media the maximum mycelial growth was recorded in potato dextrose carrot agar medium followed by Pearl millet Potato dextrose agar, potato dextrose agar, while minimum growth was recorded in Pearl millet grain agar. Similar finding were laid out by Hajano et al. (2013) reported that abiotic factors including culture media, photo-periods and temperature greatly influenced mycelial growth and sporulation of rice blast fungus *Magnapothe oryzae*. Under laboratory conditions maximum colony growth of *M. oryzae* was recorded on potato dextrose agar (PDA) followed by potato carrot agar (PCA), whereas oat meal agar (OMA) produced minimum growth. For sporulation, PDA appeared as most favorable medium, the number of conidia formed on it were even more than those produced in all remaining five media. Hossain (2000) also observed that potato dextrose agar supported maximum radial growth, next was host extract + 2 per cent sucrose agar medium followed by oat meal agar. Awoderu et al. (1991) investigated that the linear growth of *P. oryzae* was greatest on potato dextrose agar, while conidial production was greatest on one per cent soluble starch yeast extract agar. Tripathi (2006) also reported *P. grisea* was cultured on seven media Asthana and Hawkers, Czapek-Dox agar, PDA, maize meal agar, rice polished agar, carrot agar and kodo meal agar, and exposed to 3 light conditions, i.e. 12 h light and 12 h dark, 24 h continuous light, and 24 h continuous darkness. pH levels of 4-9 were maintained on the PDA medium to study the influence of pH on the growth of the fungus. Different carbon sources, i.e. glucose, fructose, maltose, sucrose, lactose and mannitol (20 g/l), and nitrogen sources, i.e. sodium nitrate, barium nitrate, lead nitrate and ammonium nitrate (2 ml/l), were added to the PDA medium. PDA alone served as the control. *P. grisea* grew well on PDA and carrot agar media. It required an optimum temperature of 25+or-2 degrees C and pH of 7 for growth and sporulation. The maximum mycelial growth and sporulation were recorded at 12 h of light and 12 h of darkness. Among the carbon sources, maltose supported the best growth (31.60 mm).

CONCLUSION

Potato dextrose carrot agar medium and pearl millet potato dextrose agar medium were found most suitable for the growth of *pyricularia grisea*.

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