

Cultural characteristics of *Cercospora beticola* sacc. causing leaf spot of palak

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ARTICLE INFO

Received : 22.07.2014
Revised : 01.09.2014
Accepted : 13.09.2014

KEY WORDS :

Cercospora beticola, Cultural characters, Palak

ABSTRACT

Palak (*Beta vulgaris* var. *bengalensis* Hort.) is one of the most popular leafy vegetables widely grown in India. This crop is severely affected by leaf spot disease caused by *Cercospora beticola* leading to brown coloured spots on the leaves which hinders the market quality of the leaves. All media are not equally good for all fungi, nor there is a universal substrate or artificial medium, upon which all fungi can grow. So, different media including both synthetic and non-synthetic were tried for *C. beticola* in the present experiment. Cultural studies of *C. beticola* grown on different solid media showed that Potato dextrose agar and Oat meal agar were good for growth. Among the nine liquid media evaluated, maximum dry mycelial weight of the fungus was obtained in Soypeptone broth which was significantly superior to all other media. Temperature requirement of the fungus was found 25°C where good growth was observed.

How to view point the article : Poornima and Hegde, Yashoda R. (2014). Cultural characteristics of *Cercospora beticola* sacc. causing leaf spot of palak. *J. Plant Protec.*, 7(2) : 441-443.

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INTRODUCTION

Palak (*Beta vulgaris* var. *bengalensis* Hort.) is one of the most popular leafy vegetables grown in India. It is widely grown in West Bengal, Uttar Pradesh, Bihar, Gujarat and Rajasthan. However, this crop is now gaining popularity in southern states like Karnataka. Since leaves are the edible parts in palak, the foliar diseases play an important role. Leaf spot disease caused by *Cercospora beticola* Sacc. is one of the major diseases of palak. Symptoms of the leaf spot disease are characterized by production of small brown spots with circular or irregular margin. Closely situated spots. Leaf spot is one of the limiting factors in the commercial cultivation of palak (Dange and Patel, 1968). However, there is paucity of information on severity of

disease in Karnataka. Therefore, in the present study an attempt was made to conduct survey for diseases of palak in order to locate hotspots for disease. It is difficult to obtain sporulation by many species of *Cercospora* on artificial media and this is true for *Cercospora beticola* also. Nagel (1934) observed difficulty in obtaining conidial production in pure culture of *C. beticola*. *C. zae* maydis and *C. asparagi* sporulated sparsely or not at all in culture unless specific requirements were met (Beckman and Payne, 1983; Cooperman and Jenkins, 1986). Mallappa (2007) reported that *C. nicotianae* grew well on most of the media but no conidia were produced in any of the media. Hence, an attempt was made to identify simple method for induction of sporulation and to study the morphological characters of *C. beticola*.

Palak belonging to family Chenopodiaceae is an important leafy vegetable grown in India. Since leaves are the edible parts in palak, so the foliar diseases play an important role. The leaves of palak are rich in vit. A, vit. C and iron. Palak being the store house of useful minerals and vitamins at the cheapest price are now-a-days considered as the corner stores of health care system due to presence of many helpful phytochemicals or phytochemicals in scavenging the dead free radicals generated as metabolic by products in alleviating many serious diseases (Kaur and Main, 2001). They also contain high quantity of ascorbic acid, being water soluble enters tissues easily and acts to neutralize the toxic free radicals generated by metabolic process, and it is a good source of carotenoids which acts as effective antioxidant in protecting cell damage of eye, and thus avoiding blindness of aged and elderly people.

This crop is severely affected by leaf spot disease caused by *C. beticola* leading to brown coloured spots on the leaves which hinders the market quality of the leaves. Every living being requires food for its growth and reproduction and fungi are not exception to it. Fungi secure food from the substrate upon which they live in. In order to culture the fungus in the laboratory, it is necessary to furnish the essential elements and compounds in the medium, for their growth and other life processes. All media are not equally good for all fungi, nor there is a universal substrate or artificial medium, upon which all fungi can grow. So, different media including both synthetic and non-synthetic were tried for *C. beticola* in the present experiment. The amount of vegetative growth was estimated by measuring diameter of the colony on solid media and by weighing the dry mycelial weight in liquid media.

MATERIAL AND METHODS

Palak leaves infected with *Cercospora* were collected from infected field and used for isolation of the fungus *in vitro*. The isolation of the fungus was made by following standard tissue isolation technique. Identification of the fungus was made based on the morphological characters of the isolated fungus. Selection of basal medium for growth and sporulation of the fungus was done by using Potato dextrose agar, Malt extract agar, Oat meal agar, Corn meal agar, Host leaf extract agar, Sabourauds agar, Richards agar, V₈ juice agar, Browns agar, Soyseptone agar, Carrot dextrose agar and Potato carrot agar media. All the media were sterilized at 1.1 kg/cm² pressure for 15 min. To carryout the study, 20 ml of each of the medium was poured in 90 mm Petriplates. Such Petriplates were inoculated with 5 mm mycelial disc cut from periphery of actively growing culture and incubated at 25°C. Each treatment was replicated thrice. Observations were taken when the fungus covered complete Petriplate in any one of the media. The

colony diameter was recorded. The fungus colony colour, margin were also recorded. The data on radial growth were analyzed statistically. Thirty ml of the medium was added to each of 100 ml flasks. All the flasks were sterilized at 1.1 kg/cm² pressure for 15 min. Inoculum disc of 5 mm size was inoculated to all flasks and incubated at 27±1°C for 16 days. Each treatment was replicated thrice. Culture was filtered through Whatman no.42 filter paper disc of 12.5 diameter, which were dried to a constant weight at 60°C, cooled in a dessicator and weighted immediately on an analytical electrical balance. The composition and preparation of different liquid media used, were the same as that of solid media except that agar was not added. The data were analysed statistically. The best medium was found out and used for further studies. The different temperatures maintained for the growth of *C. beticola* were 15, 20, 25, 30, 35 and 40°C. Mycelial discs measuring 5 mm were taken from culture plates and inoculated into Potato dextrose broth. For each treatment, three replications were maintained.

RESULTS AND DISCUSSION

Among the twelve solid media evaluated, maximum radial growth of *C. beticola* was observed on Potato dextrose agar (89.66 mm) and Oat meal agar (81.67 mm). These two were on par with each other and significantly superior over all other media tested. These were followed by Soyseptone agar (80.00 mm), Malt extract agar (79.67), Carrot dextrose agar (78.00 mm). Minimum radial growth was observed in V₈ – juice agar (33.33 mm) (Table 1). Sporulation was not observed in any of the media tested. The results are in conformity with the findings of Verma and Angihotri (1972)

Table 1 : Growth of *Cercospora beticola* on solid media

Media	Mean colony diameter (mm)
Brown's agar	55.66
Carrot dextrose agar	78.00
Corn meal agar	62.33
Host extract agar	76.00
Malt extract agar	79.67
Oat meal agar	81.67
Potato carrot agar	66.66
Potato dextrose agar	89.66
Richardss agar	66.66
Sabouraud agar	71.00
Soyseptone agar	80.00
V8 – juice agar	33.33
S.E. ±	3.13
C.D. (P=0.01)	9.15
C.V. (%)	7.75

Table 2 : Effect of liquid media on mycelial growth of *Cercospora beticola*

Media/Broth	Dry mycelial weight (mg)
Browns broth	46.66
Host extract broth	170.0
Malt extract broth	276.6
Oat meal broth	393.3
Potato dextrose broth	343.3
Richards broth	213.3
Sabourauds broth	340.0
Soypeptone broth	526.6
V8 – juice broth	280.00
S.E. ±	9.44
C.D. (P=0.01)	28.04
C.V. (%)	6.29

Table 3 : Effect of temperature on mycelial growth of *Cercospora beticola*

Temperature (°C)	Dry mycelial weight (mg)
15	50.0
20	207.50
25	312.50
30	265.00
35	212.50
40	35.00
S.E. ±	4.75
C.D. (P=0.01)	19.31
C.V. (%)	5.25

and Dange and Patel (1968). Among the nine liquid media evaluated, maximum dry mycelial weight of fungus was obtained in Soypeptone broth (526.6 mg) which was significantly superior to all other media. This was followed by Oat meal broth (393.3 mg) and Potato dextrose broth (343.3 mg). Least mycelial weight was obtained in Browns broth (46.66 mg) and Host extract broth (170.00 mg) (Table 2). Hence, Cochrane (1958) has opined the determination of dry mycelial weight as the best method for precise work. The ability of the fungus to grow in Soypeptone's medium indicated the requirement of certain nutrients and vitamins

which may be present in the medium.

Temperature plays an important role among the external factors which influence the growth and reproduction of fungi. All the fungi have minimum temperature, below which they cannot grow and above which they are inactivated or killed. Each fungus has its temperature range for the growth and sporulation. *C. beticola* under study grew best at temperature of 25°C (312.50 mg), whereas optimum temperature range was 25-30°C (Table 3). Similarly Dange and Patel (1968) reported 25-30°C as the optimum temperature range for *C. beticola*. pH of the medium has profound effect on the rate and the amount of growth and many other life processes (Lilly and Barnett, 1951).

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