

Original Research Article

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## Testing of pH Range for the Better Growth and Sporulation of *B. theobromae*

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### ABSTRACT

#### Keywords

Rose, Die-back, pH, Sporulation, Growth

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Rose is one of the important floricultural crops and is universally acclaimed as the 'Queen of flowers'. It is principle cut flower and growing more or less in all parts of north Gujarat. Therefore solving the problem associated with rose cultivation is necessary to increase the productivity. Die-back is a serious malady of rose causing considerable economic losses and a threat to rose cultivation in the area. The present investigations indicated that fungus could grow under wide range of pH from 4.5 to 9.0. However pH 6.5 and 6.0 proved to be optimum for the growth and sporulation of the fungus. More information on this problem for devising suitable control measures for preventing crop losses.

### Introduction

Rose (*Rosa* spp., Family: Rosaceae) is one of the nature's beautiful creations and is universally acclaimed as "Queen of flowers". No other flower is a better symbol of love, adoration, innocence and other virtues than the rose and not in our time only but so it has been for thousands of years (Fairbrother, 1965; Gaulf and Synge, 1971).

Rose has become the part and parcel of life, being connected with all phases of life right from birth to death. A large quantity of rose flowers is used for decorative purpose.

Besides it is used for making essence, rose water for flavouring sweets and other food articles as well as sprinkle for welcoming guests on festive occasions. Hips of some rose species are rich in vitamin C while its petals are used for preparing Gulkand and Pankhuri-two food articles of delicacy (Dhua, 1999).

Rose is affected by several fungal, bacterial, and viral diseases. The important fungal diseases are Die-back *Diplodia rosarum* (Srivastava, 1961), Powdery mildew *Spherotheca pannosa* var. *rosae* (Wallr.) Lev. (Pal, 1972), Rust *Phragmidium butleri* syd. (Chakravarti et al., 1969), Botrytis bud and

twig blight *Botrytis cinerea* (Pers.) Fries (Chohan and Kour, 1976), black leaf spot *Diplocarpon rosae* (Walf.) (Bardoloi and Ganguli, 1963) and leaf blight *Alternaria alternata* (Rao, 1964).

Among all the fungal diseases, die-back in one of the serious disease throughout the country caused by *Botryodiplodia theobromae* (Pat.) *Colletotrichum gloeosporioides* (Penz.), *Fusarium solani* (Mart.) Sacc. and *Diplodia rosarum* (Pal, 1972; Vir and Sharma 1985; Shukla and Choudhury, 1991; Dhua, 1991; Malik and Dadlani, 1984).

### Materials and Methods

For studying the effect of pH on the growth and sporulation of pathogen, Richard's medium was adjusted to pH 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5 and 9.0 in liquid state by using 0.1N HCl and 0.1N NaOH solutions with the help of Beckman's pH meter. The pH was adjusted before sterilizing and adding agar in case of solid media. Solid and liquid media in Petri plates and flasks were inoculated with 4 mm diameter dished of 15 days old pure culture of *B. theobromae* was incubated at room temperature ( $27^{\circ} \pm 2^{\circ}\text{C}$ ).

The observations for linear growth were recorded at every 24 hours interval in solid media while, dry mycelial weight, sporulation and pH were recorded after 15 days of inoculation both in solid and in liquid media. In solid media, dry mycelial weight was obtained by melting the inoculated petri dishes of all treatments after 15 days of inoculation. Then the mycelia mat from the melted media taken out with the help of forceps and put in between two previously weighted Whitman's filter paper No. 42. Then after in both the cases mycelial mats were washed with hot water to remove adhering media. Mycelia mat with filter paper were oven dried at  $60^{\circ}\text{C}$  till constant weight and cooled at room

temperature before weighing. The dry weight was measured on monopan balance.

### Results and Discussion

Richard's medium with and without agar was selected as a basal medium in the study. In solid media linear growth, spore count.

The results revealed that the fungus could grow in wide pH range from 4.5 to 9.0. Considering the linear growth, significantly maximum mycelial growth (90.00 mm) was obtained at pH 6.5 as compared to the rest. The next best was pH 6.0 (84.33) followed by pH 7.0 (82.00). With increase in pH above 7.0 and decrease below pH 5.5, linear mycelial growth was reduced.

Considering sporulation in solid media, maximum spores per optical field were observed in pH 6.5 which was significantly higher than the rest. The next best in order of merit were pH 6.0, 7.0, 7.5, and 5.5 in all solid media.

In liquid media, dry mycelial weight, spores per optical field and drift in pH were recorded. In liquid media maximum dry mycelial weight was yielded in pH 6.0 (1017.33 mg) followed by pH 6.5 (1003 mg). The next best in order of merit were pH 7.0, 5.5, 7.5 and 5.0. Considering sporulation in liquid media, maximum spores per optical field were observed in pH 6.5 (35) followed by pH 6.0 (26). The next best were pH 5.5, 5.0, 7.0 and 7.5, which were statistically at par with each other. In pH below 5.0 and in above 8.0 sporulation was not observed.

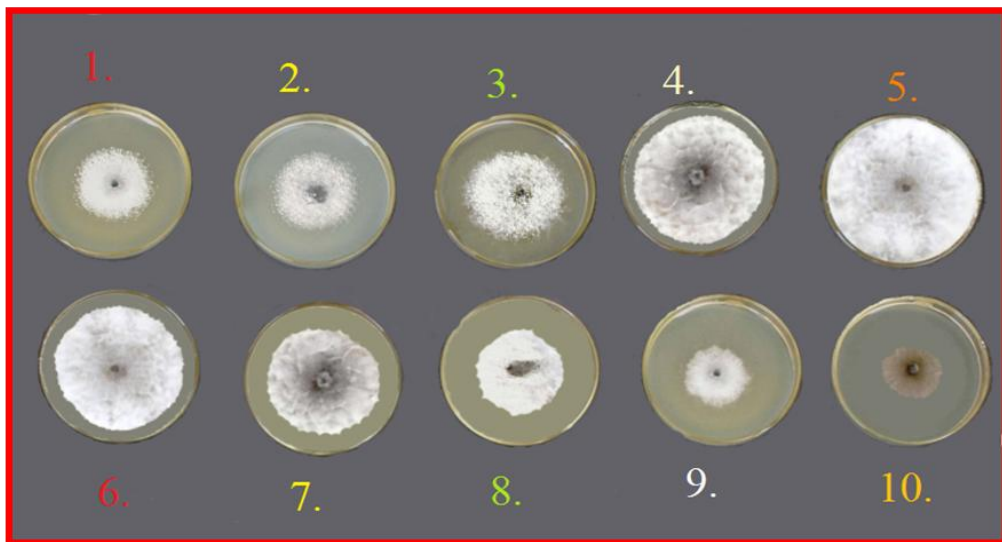
From the above results (Table 1 and 2), it is observed that neutral to slightly acidic media were proved to be more favourable as compared to alkaline. For the growth and sporulation of *B. theobromae* optimum pH 6.0 to 6.5 in both solid and liquid media.

The effect of pH on the growth and sporulation of *B. theobromae* indicated that the fungus could grow at a wide range of pH from 4.5 to 9.0 but the optimum pH laid 6.0 to 6.5 in solid and liquid media. The earlier

workers Bhatnagar (1970), Sabalpara (1983), Patel (1971), Patel (1989) and Dambhla (2001) proved that *B. theobromae* grow well in neutral to acidic medium. Our results are in line with the findings of earlier workers.

**Table.1** Growth and Sporulation of *B.theobromae* at different hydrogen-ion concentration in solid media

Sr. No.	Initial pH	Linear growth (mm)	No. of spores/ optical field
1.	4.5	36.00	0
2.	5.0	46.33	4
3.	5.5	64.67	7
4.	6.0	84.33	17
5.	6.5	90.00	43
6.	7.0	82.00	10
7.	7.5	64.67	8
8.	8.0	42.67	3
9.	8.5	24.33	0
10.	9.0	19.00	0
S. Em. ±		1.12	0.66
C. D. AT 5%		3.32	1.98
C. V. (%)		3.5	12.40



Sr. No.	Initial pH	Sr. No.	Initial pH
1.	4.5	6.	7.0
2.	5.0	7.	7.5
3.	5.5	8.	8.0
4.	6.0	9.	8.5
5.	6.5	10.	9.0

**Table.2** Dry mycelial weight and sporulation of *B. theobromae* at different hydrogen-ion concentration in liquid media

Sr. No.	Initial pH	Dry mycelial weight (mg)	No. of spores/ optical field	Final pH
1.	4.5	455.33	0	7.5
2.	5.0	690.00	11	7.5
3.	5.5	782.67	13	8.5
4.	6.0	1017.33	26	8.0
5.	6.5	1003.00	35	8.0
6.	7.0	838.67	11	8.5
7.	7.5	744.67	11	8.0
8	8.0	628.33	3	8.0
9	8.5	435.00	0	8.5
10.	9.0	332.33	0	8.5
	<b>S. Em. ±</b>	<b>6.98</b>	<b>0.49</b>	
	<b>C. D. AT</b>	<b>20.72</b>	<b>1.48</b>	
	<b>5%</b>	<b>1.74</b>	<b>7.73</b>	
	<b>C. V. (%)</b>			

The studies indicated that fungus could grow under wide range of pH from 4.5 to 9.0. However pH 6.5 and 6.0 proved to be optimum for the growth and sporulation of the fungus.

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