

## Effect of Temperature and Hydrogen ion Concentration on three pathogenic Fungi

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### Introduction

Most important environmental factors governing the growth and sporulation of fungi are temperature and hydrogen-ion concentration. A little variation in these factors may induce marked differences in their morphological characters, growth and sporulation. It is an established fact that for each fungus there is a minimum, optimum and maximum temperature for growth and sporulation. Wolf and Wolf (1947) stated that the growth of most of the fungi stops at 0° C, and only a few fungi are active beyond 40° C, whereas the optimum temperature lies somewhere between the two. Singh and Khanna (1966) working on *Alternaria tenuis* found best growth and sporulation of the fungus at 24—25° C. Tandon and Varma (1962) reported 25—27° C to be the optimum temperature for the growth and sporulation of *Colletotrichum gloeosporioides* and *Chaetostylum* showed best growth at 20° C. This shows that the fungi are highly sensitive to the temperature of the medium.

Fungi differ considerably in their tolerance to different pH values. The growth of fungi may be completely inhibited in media, which are either too acidic or too alkaline. Most of the fungi, however, tend to grow better on the acidic side. Cochran (1958) states that many fungi, with few exceptions, grow best on media with an initial pH of 5.0 to 6.5. Tandon and Chaturvedi (1963) working on *Alternaria tenuis*, found the optimum growth at pH 5.5. Grewal (1954) observed that *Colletotrichum papayae* and *Gloeosporium musarum* grew fairly well even at pH 2. Mathur et al. (1950) reported a bimodal type of curve showing the two peaks at pH 3.4 and 5.4 for *C. lindemuthianum*. Sattar and Malik (1939) reported that *C. gloeosporioides* showed best growth at pH 7.5. This variation in environmental conditions initiated the author to study the effect of various temperature and pH on the growth and sporulation of three different pathogenic fungi isolated by him.

### Materials and Methods

Three leaf spot fungi, viz; *Alternaria tenuis* from *Ixora sp.*, *Alternaria solani* from *Barleria sp.*, and *Colletotrichum gloeosporioides* from

*Gardenia sp.*, were purified by hyphal tip methods and grown on Asthana and Hawker's Medium A.\*). Extra pure chemicals (B. D. H. or E. Merck) were used. For cultural work 25 ml. of the liquid medium was poured in 150 ml. Erlenmeyer Pyrex flasks; five replicates were used in each treatment. After autoclaving (at 15 lbs. pressure for 15 minutes) the flasks were kept at various temperatures at which the growth was to be observed for at least twenty four hours before inoculation to remove the lag effect. The various temperatures tried were 0, 15, 15, 20, 25, 30, 35, and 40° C. Inoculated flasks were incubated for fifteen days at respective temperatures. For pH experiment the fungi were grown at 21 different pH values viz; 2.0, 2.5, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.5, 9.0, 9.5, 10.0, 10.5, 11.0, 11.5, and 12.0. The pH was adjusted with the help of Beckman's pH meter so that it gives the desired pH after autoclaving. The cultural procedures were the same as above. These flasks were incubated for 15 days at 25° C ( $\pm 1$ ). After the incubation period, the sporulation and dry weight were recorded. Degree of sporulation was classified by microscopic examination, using eye piece 15 X and objective 40 X, on the following basis: 1—10 spores per field of the microscope, poor; 11—30, fair; 31—70; good, 71 and above very good. For obtaining dry weight the contents of the flasks were filtered through Whatman's filter paper No. 42. The mycelial mat was subsequently dried at 70° C in an electric oven for two days and then weighed on an electrical balance.

Table I

Showing growth (in mg), and sporulation of *Alternaria tenuis*, *Alternaria solani* and *Colletotrichum gloeosporioides* at different temperatures

No.	Temp. in Centi-grade.	<i>Alternaria tenuis</i>		<i>Alternaria solani</i>		<i>Colletotrichum gloeosporioides</i>	
		Dry wt. in mg.	sporulation.	Dry wt. in mg.	sporulation.	Dry wt. in mg.	sporulation.
1.	0° C	—	—	—	—	—	—
2.	10° C	8.3	—	10.1	—	4.1	—
3.	15° C	19.5	—	22.7	+	16.6	—
4.	20° C	49.2	+++	56.2	++++	39.7	+++
5.	25° C	59.2	++++	69.3	++++	47.9	++++
6.	30° C	28.6	+	42.2	+	16.8	—
7.	35° C	20.4	—	28.7	—	8.9	—
8.	40° C	—	—	—	—	—	—
Average Mean:-		30.9		41.5		22.3	

++++ = Very Good; +++ = Good; ++ = Fair; + = Poor; — = Nil.

\*) Glucose, 5 g; KNO<sub>3</sub>, 3.5 g; KH<sub>2</sub>PO<sub>4</sub>, 1.75 g; MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.75 g; distilled water, 1 l.

## Results and Discussion

It is evident from Table I that all the three species showed no growth below 10° C. The optimum temperature for the growth of all the species under present investigation was found to be 25° C. The growth of these three fungi, however, ceased at 40° C. Their sporulation was best at 20—25° C. Similarly Green (1927) working on *Zygorhynchus moelleri* reported that the growth of the fungus increased with rise of temperature up to 26° C but above 26° it decreased and later ceased at 32° C.

Hydrogen-ion concentration of the medium can affect the growth of the fungus in two ways. Externally, it can control the degree of dissociation of the inorganic ions in the culture solution. Since dissociation plays a part in the movement of ions in the fungus, degree of dissociation will affect fungus growth. Internally it can cause changes in pH in the mycelium. It is clear from Table II that all the three fungi grew well within a pH range of 4.5—8.5. None could grow at pH 2.

Table II

Showing the effect of different pH on the growth and sporulation of *Alternaria tenuis*, *Alternaria solani* and *Colletotrichum gloeosporioides*

No.	pH	<i>Alternaria tenuis</i>		<i>Alternaria solani</i>		<i>Colletotrichum gloeosporioides</i>	
		Dry wt. in mg.	sporulation.	Dry wt. in mg.	sporulation.	Dry wt. in mg.	sporulation.
1.	2	—	—	—	—	—	—
2.	2.5	7.2	—	8.9	—	4.1	—
3.	3	13.7	—	15.6	—	9.3	—
4.	3.5	19.2	+	21.4	+	17.2	+
5.	4	25.3	+	30.0	+++	24.4	+
6.	4.5	36.0	+++	38.6	+++	29.3	+++
7.	5	44.5	+++	50.2	+++	35.7	+++
8.	5.5	49.9	+++	55.1	+++	44.0	++++
9.	6	56.5	++++	62.5	++++	51.1	++++
10.	6.5	59.8	++++	69.7	++++	50.1	++++
11.	7	58.2	++++	67.8	++++	49.1	++++
12.	7.5	49.4	+++	57.0	++++	43.0	+++
13.	8	38.6	+++	49.1	+++	38.0	++
14.	8.5	32.0	+++	39.9	+++	28.7	+
15.	9	25.1	++	33.6	+++	22.3	+
16.	9.5	18.6	++	26.5	++	19.5	+
17.	10	17.0	++	23.0	++	16.5	+
18.	10.5	14.9	—	19.2	—	13.6	+
19.	11	12.1	—	16.9	—	10.2	—
20.	11.5	8.9	—	11.3	—	6.9	—
21.	12	6.9	—	11.2	—	5.2	—
Average Mean		29.69		35.38		25.91	

++++ = Very Good; +++ = Good; ++ = Fair; + = Poor; — = Nil.

Maximum growth of *A. tenuis* and *A. solani* was recorded at pH 6.5 while *C. gloeosporioides* showed best growth at pH 6. Generally alkaline media were not favourable for growth and sporulation of these three pathogenic fungi. Similarly Johnson (1923), Brancato and Golding (1953), Agnihotri (1964) and Sarbhoy (1965) also found that the growth of fungi investigated by them was more on the acidic media than on the alkaline.

In the present case a single optimum peak of the pH was recorded for all the three pathogens, which agrees with the behaviour of fungi studied by Mehrotra (1964) and Sarbhoy (1965). On the other hand Webb (1924), Saksena (1936), Mathur et al. (1950), Mehrotra and Mehrotra (1962) and Tandon & Varma (1962) found a bimodal type of curve in the fungi investigated by them.

In the present study there was always a correlation between growth and sporulation of the three fungi.

#### Summary

The effect of two environmental factors, temperature and hydrogen-ion concentration on the growth and sporulation of *A. tenuis*, *A. solani*, and *C. gloeosporioides* was studied. It was found that 25° C was the optimum temperature for growth and sporulation of the three fungi. It was also found that pH of the medium had a marked effect on the growth and sporulation of the fungi. The best growth in case of *A. tenuis* and *A. solani* was observed at pH 6.5 while *C. gloeosporioides* has best growth at pH 6.

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