Factors Affecting Conidial Germination of *Alternaria alternata*(Fries) Keissler Causing Fruit Rot of Apple

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S.K. 탁 · O.P. 베르마 · V.N. 파닥 : 사과과일썩음증상을 일으키는 *Alternaria alternata* 의 포자발아요인

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ABSTRACT Effect of some physical and chemical factors on germination of conidia of *Alternaria alternata* (Fries) Keissler causing fruit rot of apple was investigated. The germination was maximum at 30°C, 100 per cent RH and at 5.5 pH Syllit, amongst the 11 fungicides and Planofix, amongst the 5 growth regulators caused maximum inhibition of conidial germination.

KEY WORDS *Alternaria alternata*, conidial germination, apple, fruit rot.

INTRODUCTION

Alternaria alternata (Fries) Keissler has been reported as an important pathogen of apple causing fruit rot in storage and markets from various parts of the world viz., U.S.A. (Rose, 1924), U.K.(Kidd and Beaumont, 1924), New Zealand(Brien, 1934), Rhodesia(Hopkins and Bacon, 1938), India(Agrawala and Sharma, 1968) and Canada(Coulombe, 1976). Since the knowledge of factors affecting conidial germination may help in devising the effective control measures, the present investigation was undertaken to know the germination of conidia of the pathogen under the influence of various physical and chemical factors.

MATERIALS AND METHODS

Except mentioned otherwise, following methodology was followed for all the experiments on conidial germination.

Spores from 10-day-old culture of the fungus grown in Petri dishes containing PDA were tapped on glass slides. The spore concentration on slides was maintained as 20-25 conidia per low power (100X) field of microscope. The spores on glides were given the respective treatments and the slides were placed in humid chambers consisting of Petri dishes containing double layers of moist filter paper. Each Petri dish contained two slides. The humid chambers containing slides were incubated at 30°C for 12 hours. After this, the slides were taken out, a drop of lactophenol was added to spores and examined under the microscope at 100 X to record germination percentage. For each treatment four slides were examined and each slide was observed at four microscopic fields selected randomly. To record speed of lgermination, length of five randomly selected germ tubes from each of the four microscopic fields were measured at 400X. Other specific details of the individual experiments were as under:

(i) **Effect of temperature:** A drop of sterilized water was added to the dry spores on slides and incubated at 10, 20, 30, 35 and 40°C for 12 hours after keeping in humid chambers.

(ii) **Effect of H-ion concentration:** Seven pH levels viz., 3.0, 3.5, 4.0, 5.0, 5.5 an 6.0 were adjusted in sterilized water with the help of pH meter using N/10 HCl A drop of

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respective pH solution was mixed with dry spores on slides and incubated at 30°C for 12 hours after keeping in humid chambers.

(iii) **Effect of relative humidity:** Effect of 50, 60, 70, 80, 90 and 100 per cent relative humidities was investigated. Different levels of RH were produced by mixing concentrated sulphuric acid and distilled water (Buxton and Mellanby, 1934). Ten ml of the solution was poured in the bottom of a Petri dish. Two slides containing dry conidia were kept in each Petri dish was then sealed with the help of adhesive tape and incubated at 30 °C for 12 hours.

(iv) **Effect of sugar concentration:** Four concentrations(5, 10, 15 and 20 per cent) of sucrose prepared in sterilized distilled water were used in the test. A drop of respective sugar solution were mixed with dry spores on the slides, kept in humid chambers and incubated at 30°C for 12 hours. In control treatment a drop of sterilized distilled water was added instead of sugar solution.

(v) **Effect of growth regulators:** Effect of five growth regulators namely maleic hy drazide, indole acetic acid, gibberellic acid, planofix(α naphthyl acetic acid) and Microcil (choline dichloride) was tested at 100ppm. A drop of respective growth regulator solution was added to the dry spores on glass slide and kept in humid chamber for incubation at 30°C for 12 hours. Proper control was maintained using sterilized distilled water.

(vi) **Effect of fungicides:** Effect of eleven fungicides each at three concentrations (50, 100 and 150ppm) was tested. A drop of respective fungicide solution prepared in sterilized distilled water was mixed with dry spores on slide and incubated at 30°C for 12 hours after keeping in humid chambers. Drop of sterilized distilled water served as control. Fungicides used in the test were: Aliette (aluminium tris ethylphosphonate), Aureofungin-Sol. (A haptane antibiotic), Bavistin WP(carbendazim) Bayleton(triadimefon), Baytan (triadimenol), Benlate(benomyl), Dexon (fenaminosulf), Dit hane M-45(mancozeb), Rovral (iprodione), Syllit(dodine) and Vitavax(carboxin).

RESULTS

(i) **Effect of temperature:** Maximum germination was recorded at 30°C which was significantly more than at all other temperatures(Table 1). Lowest germination was recorded at 40°C. Although there was significant difference in conidial germination at 25 and 30°C but length of germ tubes at these two temperatures did not differ much.

Table 1. Germination of conidia of *A. alternata* at dffierent temperatures after 12 hours incubation.

Temperature (°C)	Average germination (%)	Average length of germ tube (μm)
10	21(27.25)	22.50
15	35(36.25)	43.75
20	63(52.55)	53.75
25	96(82.04)	102.00
30	100(90.00)	103.75
35	66(54.34)	62.50
40	14(21.92)	11.25
LSD(5%)	(5.95)	

Figures in parentheses are angular values.

Table 2. Germination of conidia of *A. alternata* at different H-ion concentrations after incubation of 12 hours at 30±1°C.

pH	Average germination (%)	Average length of germ tube (μm)
3.0	12(20.27)	12.50
3.5	19(25.81)	23.75
4.0	20(26.56)	25.00
4.5	31(33.82)	45.00
5.0	67(55.13)	75.00
5.5	94(77.94)	92.50
6.0	60(53.78)	45.00
LSD(5%)	(12.14)	

Figures in parentheses are angular values.

(ii) **Effect of H-ion concentration:** Conidia of the fungus germinated at all the pH levels(3.0 to 6.0) tested(Table 2). The most favourable was a pH of 5.5 supporting maximum germination and length of germ tube.

(iii) **Effect of relative humidity:** Increasing levels of humidity caused significant increase in conidial germination except that the difference between 50 and 60 per cent relative humidities was non-significant(Table 3). Both, per cent germination and length of germ tube were maximum at 100 per cent relative humidity.

Table 3. Germination of conidia of *A. alternata* at different relative humidity after incubation of 12 hours at 30±1°C.

R.H.(%)	Average germination (%)	Average length of germ tube (μm)
50	6(13.985)	10
60	16(18.87)	15
70	24(29.33)	35
80	57(49.03)	50
90	72(58.08)	62.5
100	94(77.94)	90
LSD(5%)	(9.33)	

Figures in parentheses are angular values.

(iv) **Effect of sugar concentration:** Although presence of sugar significantly increased the conidial germination but differences in the concentration of sugar did not produce significant effect on conidial germination (Table 4). Presence of sugar also increased length of germ tube.

Table 4. Effect of different concentrations of sugar on germination of conidia of *A. alternata* after incubation of 12 hours at 30±1°C

Sugar concentration (%)	Average germination (%)	Average length of germ tube (μm)
5	99(87.11)	225
10	100(90.00)	237.5
15	100(90.00)	250
20	100(90.00)	218.75
Control	94,(77.94)	92.5
LSD(5%)	(7.09)	

Figures in parentheses are angular values.

(v) **Effect of growth regulators:** Except I.A.A., all the growth regulators caused significant reduction in germination of conidia (Table 5). I.A.A. resulted in significant increase in conidial germination. Planofix caused complete inhibition of germination. It was followed bymicrocil and maleic hydrazide. The similar trend was observed for length of germ tube.

Table 5. Effect of growth-regulators on germination of conidia of *A. alternata* at 30±1°C after incubation of 12 hours.

Growth regulator	Average germination (%)	Average length of germ tube (μm)
Gibberellic acid	78(62.09)	187.5
Indol Acetic Acid	100(90.00)	290.0
Maleic Hydrazide	38(37.99)	93.75
Microcil	14(21.92)	10.00
Planofix	0(0.0)	0.0
Control	94(77.94)	92.5
LSD(5%)	(6.32)	

Figures in parentheses are angular values.

(vi) **Effect of fungicides:** All the fungicides tested caused significant reduction in conidial germination. Although Syllit was the most effective fungicide in reducing conidial germination, its inhibitory effect was statistically at par to that of Dexon and Dithane M45. Fungicides x concentration interactions were significant indicating the differential behaviour of fungicides at different concentr ations. The differences were more pronounced between lowest(50ppm) and the highest(150 ppm) concentrations of a fungicideas compared to next lower or higher concentration. (Table 6)

DISCUSSION

Although optimum temperature for conidial germination was 30°C but ability of conidia to germinate on wide range of temperature

Table 6. Effect of fungicides on germination of conidia of *A. altrenata* incubated for 12 hours at 30±1°C.

Fungicide	Average germination(%)			Mean
	50ppm	100ppm	150ppm	
Aliette	22(27.94)	20(26.50)	20(26.50)	(26.98)
Fureofungin Sol	80(67.94)	76(57.42)	68(55.59)	(60.31)
Bavistin	94(77.94)	94(76.01)	86(68.29)	(74.08)
Bayleton	50(44.97)	49(44.42)	34(40.95)	(43.44)
Baytan	94(76.01)	73(58.77)	69(56.19)	(63.65)
Benlate	61(51.36)	59(59.19)	56(48.46)	(50.00)
Dexon	9(17.39)	8(16.43)	4(11.54)	(15.12)
Dithane M-45	10(18.35)	8(16.16)	7(15.20)	(16.57)
Rovral	58(49.61)	50(45.00)	46(42.69)	(45.76)
Syllit	9(17.39)	8(16.43)	3(8.65)	(14.15)
Vitavax 75 WP	14(21.27)	11(19.17)	9(17.12)	(19.19)
Control (sterilized water)	—	—	—	(77.94)
Mean	(49.58)	(41.45)	(37.36)	
LSD(5%) for fungicides		(3.25)		
for concentration		(1.69)		
for fungicides X concentration		(5.63)		

Figures in parentheses are angular values.

may be one of the factors for availability of the disease in markets during different periods of the year. Inability of good germination of the fungal conidia at lower pH and stimulation of conidial germination and length of germ tube by addition of sugar may be a poss ible reason for more susceptibility of the apple varieties having more sweetness. Similar results have been reported by Pearson and Hall(1975) for tomato isolate of *A. alternata* in which conidial germination was more in the presence of sugars. Like most plant pathogenic fungi, conidia of *A. alternata* also germinated better with higher humidities. Conidial germination of a tomato isolate of the fungus was also rapid in the presence of free moisture (Pearson and Hall, 1975). This suggests for regulation of humidity in storage houses and shops to the minimum possible level to check the infection or development of the disease.

Syllit was the most effective fungicide for inhibition of conidial germination of the pathogen. Unlike these results, Rovral has been proved to be most effective in checking conidial germination of *A. alternata* isolated from radish(Singh and Suhug, 1981). Such differences may be due to the differences in the isolates. Growth regulators are also known to possess antifungal properties(Davis and Dimond, 1953 Sinha and Wood, 1967; Martin *et al.*, 1976; Rahman, 1977). In the present studies Planofix and Micocil were most effective in checking the conidial germination of *A. alternata.* Planofix has also been reported effective in controlling fruit rot of papaya caused by *A. alternata*(Pathak *et al.*, 1981). I.A.A. Stimulated the germination. stimulat ory effect of growth regulators to lead the increased severity of various diseases has also been shown earlier(Fawcett and Spencer, 1970).

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REFERENCES

1. Agarwal, R.K. and Sharma, V.C. 1968.

Storage rot disease of apple. *Indian Phy topath.* 21 : 294—298.

2. Brien, R.M. 1934. The fungi causing rots of stored apples in New Zealand. *New Zealand J. Agric.* 48 : 143—149.
3. Buxton, P.A. and Mellanby, K. 1934. The measurement and control of humidity. *Bull. Ent. Res.* 25; 171175.
4. Coulombe, L.J. 1976. Storage rot caused by *Acremonium kiliense* Grutz and *Alternaria alternata*(Er.) Keissler in apples. *Mc.Intosh. Phytoprotection* 57 : 33—35.
5. Davis, D. and Dimond, A.E. 1953. Inducingdisease resistance with plant growth regulators. *Phytopathology* 43 : 137~140.
6. Fawcett, C.H. and Spencer, D.M. 1970. Plant chemotherapy with natural products. *Ann. Rev. Phytopath.* 8 : 403~414.
7. Hopkins, J.C.F. and Bancon, A.L. 1938. Common diseases of apples and their control in Southern Rhodesia. *Rhod. agric. J.* 35 : 452~466.
8. Kidd, M.N. and Beaumont, A. 1924. Apple rot fungi in storage. *Trans. Br t. Mycol. Soc.* 10 : 98~118.
9. Martin, D., Lewis, T.L., Cerny, J. and Rathowsky, D.A. 1976. The effect of tree sprays of calcium, boron, zinc and naphthaline acetic acid, alone and in all combination on the incidence of storage disorders of Marton apples. *Australian J. Agri. Res.* 27 : 391~398.
10. Pathak, V.N., Gupta, A.K. and Mazumdar, V.L. 1981. Management of postharvest fungal rots of papaya fruits with chemical and heat treatments. *Proce. 3rd Int. Symp. Pl. Patho.* New Delhi, Dec. 14~18, 1981 : 146(abst.).
11. Pearson, R.C. and Hall, D.H. 1975. Factors affecting the occurrence and severity of blackmold of ripe tomato fruitcaused by *Alternaria alternata. Phytopathology* 65 : 1352-1359.
12. Rahman, A.M. 1977. Interactions between ethephon and fungicides in tomato disease control. *Pl. Dis. Rept.* 61 : 106~110.
13. Rose, D.H. 1924. Diseases of apples on the market. *U.S. Dept. of Agric.Bull.* 1253 : 24.
14. Sinha, A.K. and Wood, R.K.S. 1967. The effect of growth substances on Verticillium wilt of tomato plants. *Ann. Appl. Biol.* 59 : 117~128.
15. Singh, R. and Suhug, L.S. 1981. *Alternaria alternata,* a sesious menace to radish seed multiplication programme in Haryana. *Proce. 3rd Int. Symp. Pl. Patho.* New Delhi Dec. 14~18, 1981 : 175~176(abst.).