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In Vitro Impact of Temperature on the Radial Growth of Pathogenic Fungi

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ABSTRACT *Environmental factors play an important role in the development of post harvest diseases associated with fruits. Fruits rotting fungi grow best at optimum temperature and related moisture content. The extremely high and very low temperature decreases the growth of fungi. To determine the growth rate of fungi, fungal isolates were grown in in vitro on agar. Among the various isolates, studies were conducted on the growth of most dominant five isolates Apergillus niger, Penicillium, Fusarium oxysporum, A.fumigatus and A. flavus fungi by incubating at different temperature 10,15,20,25,30,35 and 40˚C for six days. This study indicated that isolated fungi were able to grown in large temperature range. However, a temperature below 10˚C and above 35˚C presented inhibitory effects on mycelial growth. Results revealed that optimum temperature for maximum growth, range from 25˚C to 30˚C.The findings suggest that storage of fruits at low temperature reduces spoilage of fruits. Controlling the storage conditions can play a significant role in reducing the growth of fruit rotting fungi.*

INTRODUCTION

Temperature is one of the environmental factor which plays an important role in the establishment of fungal diseases. Fruits are more sensitive to elevated temperature in their later stages of maturation. The post harvest diseases such as Penicillium, Aspergillus, Goetrichum, Alternaria, Penicillium, Curvularia and Rhizopus sp., are important fungi which infect market storage of fruits (Rathod, 2010, Bhale, 2011 and Singh, 2012). The productivity of fruits is affected by various post harvest fungal diseases which may reduce fruit quality and cause severe losses, because they lead to unmarketable fruits. The optimum growth temperatures for the majority of fungi studied was found to fall between 25˚C to 30˚C and above 40˚C the growth was poor and in some cases mortality may occur (Sharma and Razak, 2003).The temperature is an important physical factor affecting the growth and metabolic activity of pathogenic fungi. The better preservation of the fruits at low temperature may help checking of fungus growth. The prevailing climatic condition of Barak Valley is quite conducive for infection of the fruits by various microorganisms mostly fungi.

In the present study an attempt has been made to find out the dominant fungi associated with fruit rots and the effect of temperatures on the growth of pathogenic fungi.

MATERIAL AND METHODS

Isolation of pathogens:

Fungal isolates were obtained from the decayed fruits collected from the market in a pre sterilized polyethylene bag and brought to the laboratory. Isolation from fruits was made by cutting a small section of the infected portion which was sterilized with 1% NaOCl solution for 2mins and subsequently washed in sterilized distilled water. Using a sterile scalpel, and forceps infected part of the fruit was peeled from the marginal area between decayed and healthy tissues was cut into 2mm² then inoculated on sterile potato dextrose agar (PDA) in 9cm petri dishes and incubated in an inverted position at 28±2ºC for 48-72 hours for growth of pathogens. Isolated fungi were purified with single spore technique and then kept in a refrigerator on PDA slants.

Identification:

The morphological identification of the fungal strains was based on the morphology of the fungal culture colony, size, colour and characteristics of the spores or hyphae and reproductive structure were examined critically with reference to mycological texts (Gilman,1957 and Barnett and Hunter,1972).Moisture content was determined both healthy and infected by inoculation of respective fungi.

Effect of different culture media:

The isolated fungi were cultured on three different media i.e., PDA, Fungal agar and Czapeck`s dox agar. Petriplates with different media were inoculated with the tests fungi and incubated in darkness at 28±2˚C, their linear growth (cm) was determined.

Effect of temperature:

The effect of temperature on the rate of disease development was determined by inoculating 5mm disc of mycelia plug cut with a sterile cork borer from the margin of a 6 days old colony of five dominant isolates. Inoculated plates were incubated at different temperatures (10,15,20,25,30,35 and 40˚C) for six days. After the incubation period, the linear growth (cm) was determined.

Experimental Design

All the experiments were carried-out in completely randomized blocked design with three replications.

RESULTS AND DISCUSSION

A total of 14 fungal isolates were isolated from the local fruits were identified and classified. Aspergillus, Penicillium, Fusarium, Alternaria, Trichoderma, Chalaropsis, Curvularia, Geotrichum and sterile mycelia were the dominant fungal genera. Apergillus niger, Penicillium sp., Fusarium oxysporum, A.flavus, and A.fumigatus were found to be the pathogenic associated with fruits rot.

Effect of different culture media:

Table 1. Shows the effect of growth in different medium. The FA medium was most suitable growth medium for A.flavus (7.23±0.12) and A.niger (7.00±0.15) with a significance difference at ($p<0.05$); whereas CDA medium was suitable medium for Fusarium (7.50±0.62) and A.fumigatus (7.35±0.12) for PDA Penicillium sp.(4.25±0.18).

Effect of temperature:

Temperature is most important physical environmental nor there can a universal substrates or artificial medium on factor for regulating the growth and reproduction of which

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all fungi grow well. It was apparent from the results (Table 2.) that the temperature exerted a significant effect on the growth of fungi during fruits storage. Maximum growth of Aspergillus flavus (5.45±0.12), A.fumigatus (5.42±0.1) and Penicillium sp.(4.20±0.1) were found at 30˚C and gradually decreased with further either increase or decrease in temperature. A.fumigatus and A.niger both can grow wide range of temperature i,e. 10 to 40˚C. From the study, it is clear that optimum temperature for maximum growth of fungi, ranges from 25˚C to 35˚C. The pathogens did not show any growth above 45˚C and below 10˚C. Ababutain (2013) reported similar observation of A.niger can grow wide range of temperature i,e. 10 to 40˚C.Gadgile et al (2010) have also reported that A.flavus was unable to cause disease in mango fruits at 10˚C. Rawal and Manjunatha (2002) have similar observation and recommended low temperature for storage of grapes fruits to minimize fruit spoilage. At 20, 15 and 10˚C the mycelial growth was correspondingly reduced. The temperatures 25 and 30°C favored colony diameter growth for respectively so they considered as the optimum growth temperature.

Temperature is important component of the environment which affects the life of fruits, especially the rate of respiration in micro-organisms. High temperature and long storage times favoured the development of fruit rots while low temperatures and fast ripening times inhibited the growth of fungi. The low temperature reduced respiration of the decaying micro organisms and other enzymatic reactions, which caused deterioration of stored fruits. Present investigation revealed that the storage of fruits below 10˚C did not favor the growth of any fungi.

CONCLUSION

From the above result, it can be concluded high temperature and low temperature reduce the growth of fungal pathogens in fruits but at high temperature may reduce the quality and texture of fruits. Therefore, it is highly recommended that fruits should be stored at low temperature to avoid post harvest infections this may contribute significantly in controlling the heavy losses.

Table 1: Effect of growth media on fungal isolates

Fungi	Colony diameter (cm)* ± SD				
	F.oxysporum	A.flavus	A.niger	Penicillium sp.	A.fumigatus
PDA	2.9±0.05	4.28±0.18	4.28±0.62	4.25±0.18	4.68±0.13
FA	5.08±0.13	7.23±0.12	7.00±0.15	4.02±0.02	7.2±0.21
CDA	7.50±0.62	5.06±0.02	6.25±0.12	4.00±0.15	7.35±0.12
*Each value is an average of 3 replicates; ± Standard deviation					

Table 2: Effect of temperature on the different fungal isolates (6 days after incubation)

Temperature (in˚C)	Colony diameter (cm)* ± SD				
	F.oxysporum	A.flavus	A.niger	Penicillium sp.	A.fumigatus
10	0	0	0	0	0
15	1.51±0.06	0.86±0.02	1.63±0.08	1.41±0.08	0.87±0.02
20	2.21±0.12	2.86±0.02	4.97±0.09	1.95±0.04	2.06±0.02
25	2.86±0.02	4.05±0.17	4.50±0.04	3.41±0.08	4.65±0.17
30	2.33±0.06	5.45±0.12	4.28±0.62	4.20±0.10	5.42±0.10
35	0.93±0.09	3.65±0.08	4.11±0.44	3.21.±0.02	5.08±0.04
40	0	0	1.90±0.04	0	3.63±0.04
* Each value is an average of 3 replicates; ± Standard deviation					

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