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J. Mycopathol. Res. 60(4) : *625-627, 2022;*
ISSN 0971-3719

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J. Mycopathol. Res. **60(4)** : 625-627, 2022; (ISSN 0971-3719)
doi:10.57023/JMycR.60.4.2022.625

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Received : 24.05.2022 Accepted : 14.10.2022 Published : 26.12.2022

Curvularia lunata, Fusarium semitectum and *Alternaria alternata* have been detected as grain molds from the ear heads of *Sorghum* cultivar SPV-881. The moldy grains were collected from fields and pure cultures of above three grain molds have been raised. All the three fungi were subjected for the elaboration of cellulases and pectinases as they are involved in the grain deterioration. *Curvularia lunata* and *Fusarium semitectum* elaborated both cellulase and pectinase while *Alternaria alternata* showed the presence of only cellulase. The data clearly indicates the role of such enzymes in the deterioration of Sorghum cultivar SPV-881.

Key words :Cellulase, fungi, grain mold, , pectinase,sorghum

INTRODUCTION

In recent times, grain molds have been worked out by some researchers in India and other countries (Ackerman *et al.* 2021;Fatima *et al.* 2021). Marked reduction in size and weight of the sorghum grain along with germination loss due to grain molds have also been previously reported. It has been observed that during grain mold infection, several enzymes elaborated by grain mold fungi play an important role. Suresh Nath and Naveen Kango (2022) have elaborated recent developments and role of various fungal enzymes in the field of Biotechnology and Industry. Therefore, it has been thought worthwhile to evaluate the elaboration of cellulases and pectinases with reference to some grain mold fungi.

MATERIALS AND METHODS

Curvularia lunata, Fusarium semitectum and *Alternaria alternata*, the grain mold fungi associated with the Cultivar SPV-881 were subjected for the elaboration of cellulases andpectinases.

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The fungi were inoculated in 25ml Lilly and Barnett sterilized medium in 100ml conical flasks (triplicates) and were incubated at 27^0 – 29^0C, at the end of 4^{th}, 8^{th} and 12 days of incubation period. The set of flasks were harvested for its mycelium on previously dried and weighed Whatman filter paper No. 42. They were weighed to a constant weight in an analytical balance after cooling to room temperature in a desiccator.Since the difference in dry weight of the mycelium (in mg) among the replicates was statistically insignificant, average of three replicates was taken as criterion for determining the mycelial growth rate. The filtrates were passed through Seitz filter and thus obtained was used as enzyme source.

Cellulases

a) Endoglucanase – cx (EC 3.2.1.4) activity was assayed viscomertically as suggested by Reese *et al.* (1950). The activity of Endoglucanase – cx was expressed in relative viscomertic units (RVU).
RVU = 1000 / tv50,
where
tv50= time required in minutes to reduce the viscosity of CMC to 50% of the initial velocity
b) Cellobiohydrolase – C1 (EC 3.2.1.9) activity was determined by DNS method as suggested by Berber *et al.* (2021) and Miller (1959).

The reaction mixture consisting of 3.5 ml of 1% cellulose solution, 0.5 ml enzyme and 1 ml citrate buffer (pH 5.5) was incubated at 30 ± 1^0 C for 6 hours. One ml of distilled water in place of enzyme extract served as blank and the enzyme activity was expressed as the increase in mg of reducing groups (as glucose/ml) liberated in 6 hours.

2. Pectinase

a) Pectin methyl esterase (PME) : PME activity was determined according to the method of Kertesz (1937).

The reaction mixture consists of 10 ml of enzyme and 5 ml of distilled water and 25 ml of 1% pectin solution was taken in a 250 ml Erlenmeyer conical flask. 1 or 2 drops of Methyl red was added to it. The pH was adjusted to 6.2 with the addition of 0.1 NNaoH solution to the reaction mixture solution to prevent it from turning pink at every 10 minutes. The amount of methoxyl groups (in mg) which got split off during the reaction time by 1 ml of the enzyme was calculated according to the formula.

$$\text{No. of methoxyl groups split off by 1 ml of enzyme} = \frac{\text{Total ml of 0.1 N NaoH}}{\text{Total volume of enzyme used}} \times 3.1$$

RESULTS

The grain mold fungi, associated with the Cultivar SPV-881 namely *Curvularia lunata, Fusarium semitectum* and *Alternaria alternata* were subjected for the elaboration of cellulases and pectinases following standard methods. The *Sorghum* grain damage was also more evident because of the grain mold fungi. *Curvularia lunata* and *Fusarium semitectum* have shown the presence of both cellulases and pectinases while *Alternaria alternata* has elaborated only cellulases (Table1).

Table.1 : Cellulases andpectinases elaborated by Grain mold Fungi

Grain mold Fungi	Endoglucanase	Cellobiohydrolase	Pectin methyl esterase
Curvularia lunata	3.7	2.0	20.9
Fusarium semitectum	4.6	1.6	30.0
Alternaria alternata	1.8	0.8	0.0

Units are expressed as per methodology.

1. Endoglucanase expressed as RVU units.
2. Cellobiohydrolase expressed as glucose / ml per 6 hours.
3. PME calculated as mg of methoxy groups split by 1 ml. of enzyme every 10 mins..

From the Table1,it is clear that the enzymes elaborated by grain molds played an important role in the biodegradation of sorghum grains.

DISCUSSION

Enzymes are group of proteinoid organic compounds elaborated by fungi and every enzyme has its own application. Enzymes are derived from different sources including from fungi by means of various catalytic activities and genetic manipulation (Yadav, 2021). Gendi*et al.* (2022) and Swami (2020) have dealt with the structure, classification and role of enzymes in Mankind's challenges. Fungal enzymes are known to play significant role in the biodeterioration of grains. In recent times, cellulase enzyme research gained importance in the production of ethanol. Many microbes and fungi have evolved the necessary enzymatic machinery for the biodeterioration of seed materials and also for converting cellulose to a product of energy source (Christina *et al*. 2015). Therefore, data presented in this communication gains importance.

ACKNOWLEDGEMENT

The Author, Aravinda, T. is thankful to the Commissioner, Collegiate Education, Telangana State for consistent support and encouragement.

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