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ENZYME PRODUCTION BY BACTERIA ISOLATED FROM FRUITS COLLECTED FROM FRUIT MARKETS

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The bacteria which are responsible for post harvest losses of fruits produce various enzymes such as cellulase, chitinase, amylase, lipase, protease, pectinase, which utilize the nutrients present in these fruits as macromolecules. These bacteria have been isolated from apple, grapes, guava and sapota fruits. The isolates were purified and were identified to generic levels on the basis of morphological, cultural, physiological and biochemical characteristics. Out of the nine forms isolated two belonged to the genus *Erwinia*, two *Pseudomonas*, two *Xanthomonas*, one *Bacillus*, one *Micrococcus* and one *Staphylococcus*.

Keywords : Bacterial genera; Extra cellular enzymes; Market fruits; Polysaccharides.

Fruits are one of the most vital source of nutrients which are necessary for the existence of life. Nutritional value of fruits chiefly depends on their quality and concentration of sugars, vitamins and other essential substances. Microbes cause damage and loss to these fruits. Bacteria is one amongst them. They are able to decompose various polysaccharides by the secretion of enzymes, which results in depletion of nutrients and rotting of fruits. In the present paper various bacterial isolates that decomposed polysaccharides of fruits namely guava, apple, grapes and sapota are reported.

Selected fruits showing rotting were collected in sterile polythene bags from fruit markets of Ajmer and Kishangarh. They were brought to the laboratory for the isolation of various bacterial forms. After isolation these bacteria were further purified and were identified using Bergey's[1] and Holt *et al.*[2]. determinative tests. Later these bacterial isolates were tested using various media (pH-6.8) as given below to know their ability to decompose various polysaccharides. All the tests were performed on prepoured plates by spread plate method.

Cellulolytic activity : To detect cellulolytic activity the medium described by McIntyre and Hankin[3] was used. After a period of 6 to 8 days incubation the plates were flooded with 1% aquous solution of hexadecyl trimethyl ammonium bromide. This reagent precipitates intact cellulase and thus a clear zone around the colony in an otherwise opaque medium indicated degradation of cellulose.

Chitinolytic activity : Chitinolytic activity was determined with the medium described by Hankin and Anagnostakis[4]. After 5 to 8 days of incubation clear zones were seen in the opaque agar around colonies able to degrade chitin.

Amylolytic activity : The ability to degrade starch was used as the criterion for the determination of ability to produce amylolytic enzymes. The medium used contained N.A. with 0.2% soluble starch. After 3 to 5 days of incubation the plates were flooded with an iodine solution. A yellow zone around colony in an otherwise blue medium indicated amylolytic activity.

Proteolytic activity : The ability to degrade gelatin (as the protein substrate) was used as the criterion for the detection of proteolytic enzymes. The medium used contained N.A. with 0.4% gelatin. After 2 to 3 days of incubation the plates were flooded with saturated solution of ammonium sulphate. A clear zone around the colony in an otherwise opaque medium indicated proteolytic activity.

Lipolytic activity : The medium described by Hankin *et al.*[5] was used to detect lipolytic activity. The plates were incubated for 3 to 5 days. The production of lipolytic enzymes by the bacterial isolates was seen in the formation of crystals or as a clear zone.

Enzymatic activity by the phytopathogenic bacteria which is used as routine biochemical character has also been employed by investigators[6-8] in the past to group them into various genera. Verma[9] characterised bacterial isolates belonging to genus *Pseudomonas* isolated from rotted vegetable specimens and stated that this character help in characterisation of bacteria. Similar observation were made in the present study on the bacteria isolated from rotted fruit specimens collected from the local markets. The ability of the bacterial isolates obtained from fruits to produce enzymes on solid media is shown in Table 1. The word enzyme production is here intended to mean both synthesis of the enzyme by the bacteria and the ability of the enzyme to degrade the substrate in the medium after its production. From the data in the table it is clear that some bacteria *Bacillus* sp. and *Micrococcus* sp. produced cellulase, amylase, protease and lipase. While the others *Erwinia* sp. 2. *Xanthomonas* sp. 1 and 2, *Pseudomonas* sp. 1 and 2 and *Staphylococcus* sp. produced amylase, protease and lipase only.

Erwinia sp. 1 produced only amylase and protease. None of the isolates of the present study could

Table 1. Bacterial isolates obtained from post harvest fruits producing extracellular hydrolytic enzymes on solid media

Bacterial isolates	Cellulolytic	Chitinolytic	Amylolytic	Proteolytic	Lipolytic
Erwinia sp.1	-	-	+	+	-
Erwinia sp. 2	-	-	+	+	+
Bacillus sp.	+	-	+	+	+
Micrococcus sp.	+	-	+	+	+
Pseudomonas sp.1	-	-	+	+	+
Pseudomonas sp.2	-	-	+	+	+
Xanthomonas sp.1	-	-	+	+	+
Xanthomonas sp.2	-	-	+	+	+
Staphylococcus sp.	-	-	+	+	+

+ : Indicates production and utilization of substrate
- : Indicates non-production and non utilization

use chitin even after prolonged incubation periods. This observation is in full agreement with that of Liao and Well[10] who reported that neither of the yellow pigmented *Xanthomonas, Pseudomonas* and *Erwinia* produced enzyme chitinase.

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