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A Study of Betel Leaf Microflora

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Abstract : The Betal vine *(Piper betel)* is a perennial dioecius creeper belonging to the family Piperaceae. The leaves of the plant have been traditionally used for chewing. Over thirty five varieties of betal leaves are cultivated in out country. They are grown in special Pan orchids known as 'Barejas' and have a complex system of cultivation, cropping and development. The study focuses on a comprehensive microbiological investigations of pathogen causing leaf diseases. It deals with pathogen isolation, pathogen classification, pathogen biochemical analysis and properties of pathogen culture. The study also compares the disease incidents on different betel leaf varieties and the losses caused to farmers and consumers. The present high rate of oral cancers in Bhopal underline the importance of the study.

Key words : Microflora, Betal Leaf.

Introduction :

Pan is the local vernacular Hindi name used for leaves of Betel Vine Plant *(Piper betel)*. It is a perennial creeper belonging to family Piperaceae. On account of its immense medicinal, social, religious and export value betel vine is a crash crop of economic importance and is extensively grown on large scale in different parts of India in general and in particular MP. Madhya Pradesh is the leading State in betal vine production and in export. Approximately 15 different varieties are cultivated in MP. The pan leaves are affected by common agents which introduce microbial pathogens onto leaf surface, they are agency of wind, its direction and presence of microbial load, agency of water and its properties, soil, its properties and during post harvest stages, factors like packaging material, moisture content and finally water in which the leaves are submerged before they are converted into betel quid. The leaves, which are subjected to these factors, develop leaf diseases like 'Leaf Spot' and 'Leaf Blight' resulting in economic loss to distributors, shop keepers and people involved in this trade CSIR (1984).

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Materials and Methods :

Diseased leaves were collected from different locations in the city and studied in detail for their disease symptomatology and grouped as leaf spot and leaf blight infections (Bhale *et al*., 1985).

Young developing lesions from leaves were selected for isolation and a part of the infected tissue was cut using sterilized blade and the bacterial fluid was collected. The isolation was made by streaking under aseptic conditions on PSPA medium (potato sucrose peptone medium). The bacterial culture was investigated for its cellular morphology. Common staining methods were used to determine its properties (Dye, 1962). Biochemical studies were concluded on the bacterial isolate and using 48 hour old growth culture of bacterial isolate the gelatin columns were stabbed by a straight inoculating needle. The tube was incubated at 20^{o} C for one month. The isolate was also tested for its H_2S liberation quality. Peptone water medium was inoculated with 48 hours bacterial isolate culture and a lead acetate strip was placed just over the broth surface. The flask was incubated at 20° C for 14 days (Choi *et al*., 1982).

Results and Discussion :

Collected leaf samples from different locations in the city were grouped according to the leaf type and disease symptoms (Table 1).

Table 1 : Type of leaf and disease symptoms in pan.

S. No.	Type of Leaf	Disease Symptoms
1.	Madrasi patta/pan	Leaf Spot
2.	Bangla patta/pan	Leaf Blight
3.	Meetha patta/pan	Leaf Spot
4.	Kapoori patta/pan	Leaf Spot
5.	Banarsi pata/pan	Leaf Blight

Colony size (mm) of bacterial isolate on PSPA medium under 25° C temperature for a maximum of 240 hours.

Table 2 : Colony size of bacterial isolate on PSPA medium.

Colony Size of isolate in (mm)	48 hrs	96 hrs	144 hrs	192 hrs	240 hrs
Isolate I	1.1	2.0	2.8	3.5	4.0

Colony size increased with reference to time period and the size increased till 240 hours, when it had a final diameter of 4.0 mm, colony morphology was studied and the colony shape was round, its margin was entire, the colony elevation was raised and colony colour was yellow (Jindal and Patel, 1984).

The detail microscopic study of the colony showed that the cells were of rod shaped bacilli gram negative and non-spore formers. They also showed presence of capsule (Raguramulu and Madhaven, 1983). The biochemical study showed that the bacterial isolate culture had gelatin liquification property and could release H_2S resulting in appearance of black colour on lead acetate strips.

Based on disease symptoms, cellular morphology biochemical properties and colony characters the causative disease organism was identified as *Xanthomonas compestris* pv. Betticola bacteria causing leaf damage (Mathew *et al.,* (1978).

Madhya Pradesh is one of the leading states of betel-vine cultivation, has large areas under betel vine cultivation. Early recognition of disease symptoms and their control can reduce the economic loss caused to farmers and cultivers. So, a better betel leaf crop can be obtained.

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