**Table 4.** Removal of silica from magnesite ore in the fermenter after treatment with the mutant  $U_{68}N_{66}E_{77}$ 

No. of days	Sinca content* in ore (%)		Total silica removal from ore (%)	
	Control	Treated with bacteria	Control	Treated
0	5.5	5.5	_	_
4	5.2	3.6	5.45	34.5
8	5,2	2.17	5.45	60 5

Control: No bacterial inoculum was added to the fermenter.

\* = average of three replicates.

hope for refractory industry, considering the rapid depletion of high grade magnesite in India.

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## Taxonomic status of rust on mulberry in India

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The morphology of the rust fungus on mulberry (Morus alba L.) reported by earlier workers is not in conformity with generic concepts of any of the genera to which the fungus has been assigned, viz. Caeoma, Aecidium, Uredo and Cerotelium. The fungus shows characteristic features of the form genus Peridiopsora Kamat & Sathe apud Sathe, based on peridial characters and urediniospores. Detailed histopathological studies revealed that the rust fungus occurring on mulberry in India is Peridiopsora. Hence the new combination Peridiopsora mori (Barclay) Prasad et al., comb. nov. has been suggested for the fungus.

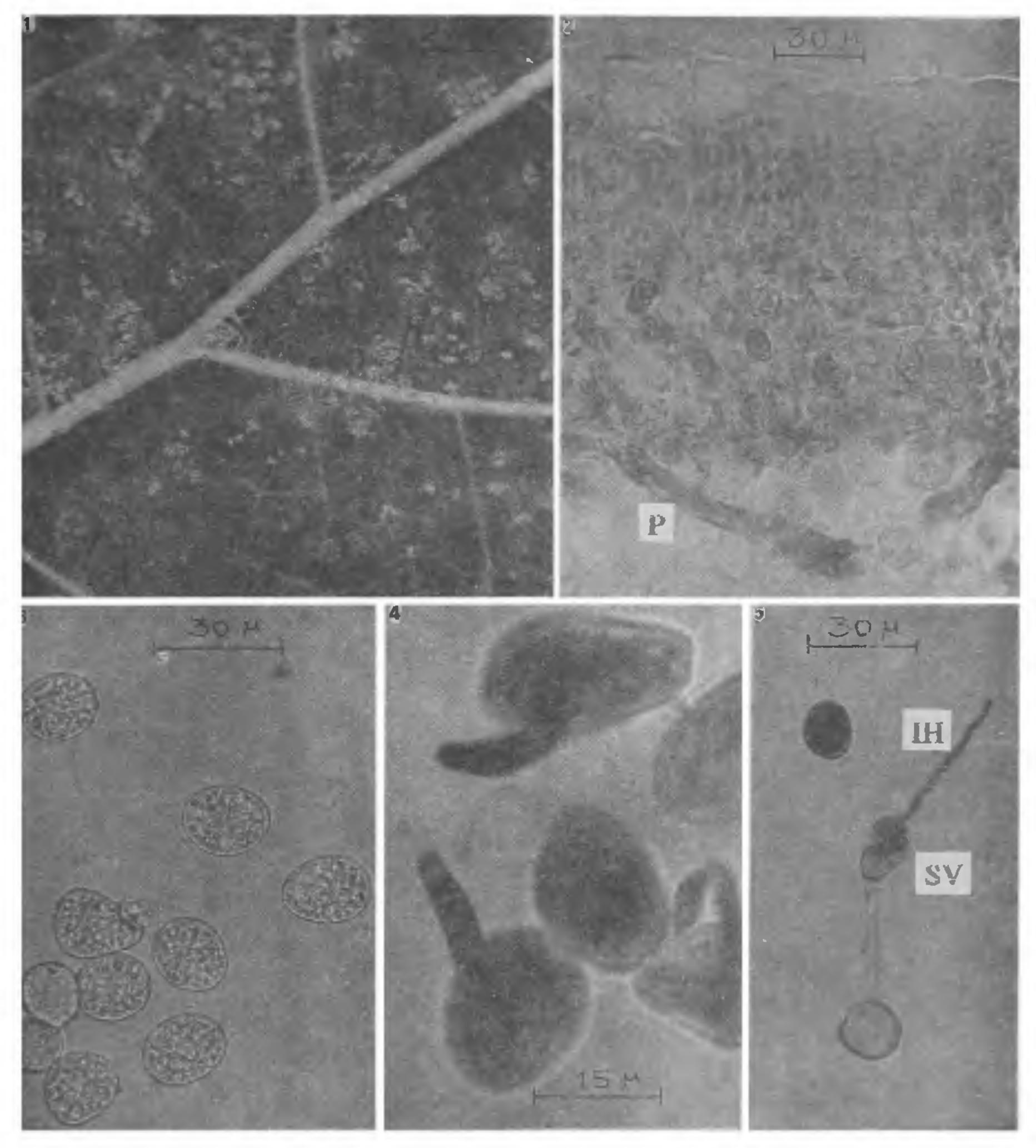
CAEOMA MORI Barclay<sup>1</sup>, Aecidium mori Barclay<sup>2</sup>, Uredo mori Barclay sensu Saccardo<sup>3</sup>, Aecidium mori (Barclay) Sydow et Butler<sup>4</sup>, and Cerotelium fici (Cast) Arthur<sup>5,6</sup> have been reported to be the causal organisms of rust disease in Morus alba L. and M. indica L.

The review of literature reveals that this rust is known to the Indian workers in the sield either as

Aecidium mori Barclay or as Cerotelium fici (Cast) Arthur<sup>5,6</sup>. It is noteworthy in this connection that Barclay himself had described this rust under Caeoma mori and as such the transfer effected as Aecidium mori (Barclay) Syd. & Butler is a valid transfer. As far as its placement under Cerotelium fici, it was made out of the confusion arising due to the fact that Barclay while describing the rust under Aecidium mori also included the rust on Ficus because the uredinial sori of Uredo fici Cast. are in fact peridiate and ostiolate<sup>8</sup>. Subsequently, however, the two rusts namely Aecidium mori and Cerotelium fici were separately treated by Sydow & Butler<sup>4</sup>. Cerotelium fici, therefore, is a misnomer for the rust on mulberry in India. The telial stage for the rust on mulberry has not been reported so far, and the rust is believed to be prevailing in its anamorphic state only.

During the course of studies on rust fungus on mulberry in Karnataka, it was noticed that the rust fungus prevailing in the state is not in conformity with generic concepts of either Caeoma, Aecidium or Uredo and therefore, it was decided to undertake the detailed studies of the same.

Leaves infected with the rust fungus were collected from the mulberry farm of the Institute (KSSDI). The rust was found to manifest itself on the lower (adaxial) surface of leaves in the form of yellowish to reddish brown blisters grouped together (Figure 1).



Figures 1-5. I, Leaf rust pustules on adaxial side; 2, Urediniosorus with peridium (P) and subsessile urediniospores, 3, Urediniospore with echinulate to verrucose surface; 4, Germinating urediniospores, 5, Urediniospore with substomatal vesicle (SV) and infectious hypha (IH).

The blister-like pustules measured 0.4 to 0.5 mm in diameter. Each pustule was provided with a central ostiole, through which creamish masses of spores were seen coming out. On microscopic examination the sectional view of the pustule revealed that it was a uredinial sorus bearing sessile or subsessile urediniospores (Figure 2). The uredinium was ostiolate and was provided with the peripheral thin cellular peridium,

each cell measuring  $18(21) \times 5(7) \, \mu \text{m}$ . The urediniospores were subglobose, oval to pyriform in shape and admeasuring  $15(24)-18(30) \times 12-15(18) \, \mu \text{m}$  and the surface was echinulate to verrucose (Figure 3). The urediniospores were provided with four equatorial germ pores. On germination they produced germ tube of 6-8  $\mu \text{m}$  length (Figure 4).

The germ tube prior to entering the leaves produced

characteristic appressorium from which infection hypha emerged (Figure 5).

The various characters revealed by the uredinial sorus of the rust under the present consideration is in conformity with the form-genus *Peridiopsora* Kamat & Sathe apud Sathe<sup>9,10</sup>.

In Uredinales, by and large genera are identified primarily on the features of telial state. Many rusts where telial state has not been discovered, assigning them to any particular genus is rather difficult. Since rust fungi exhibit host specialization this attribute has been exploited by mycologists for making a detailed study of aecia and uredinia and assigning them to different form genera.

The entire structure of the uredinial sorus is also in conformity with the structure of Caeoma mori Barclay. It is, therefore, essential to transfer the species to the form genus Peridiopsora and this is effected here as Peridiopsora mori (Barclay) Prasad K. V. et al. comb. nov. ( $\equiv$  Caeoma mori Barclay in J. Asiatic Soc. Bengal,

1890, 59, 97 Figs.).

In conclusion, the rust fungus infecting Morus alba L. in India is Peridiopsora mori (Barclay) Prasad et al. and the other names become synonyms of the same.

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## Isolation of a chlorate-resistant line from protoplast cultures of *Hyoscyamus* muticus L.

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A chlorate-resistant line resistant to 40 mM potassium chlorate (KClO<sub>3</sub>) has been isolated from suspension culture protoplast derived colonies (30–35 celled stage) of *Hyoscyamus muticus*. In comparison to the wild type, the selected line ClO<sup>1</sup>1 registered nearly 3-fold decrease in the nitrate reductase enzyme activity (157.25 units/g fresh wt.). This characterization of ClO<sup>1</sup>1 was reflected in their slower growth on medium having NO<sub>3</sub> as sole nitrogen source.

Isolation of conditional mutants such as nitrate reductase deficient, amino acid and vitamin requiring mutants in vitro and their use have led to the progress of developing a majority of somatic hybrids to-date<sup>1,2</sup>. The importance of such mutations is further enhanced in developing universal hybridizers<sup>3,4</sup>. Although several technical advancements such as fluorescent activated cell sorting (FACS) and microculture techniques have emerged for the efficient screening of somatic hybrids<sup>5,6</sup>, the importance of inducing selectable markers to desirable genetic background still prevails. It has been studied earlier that the screening for chlorate resistance can indirectly result in recovery of nitrate reductase (NR) deficient mutants<sup>7</sup>. In this communication we report the isolation of a chlorate-resistant line from

protoplast cultures of *Hyoscyamus muticus*. The importance of this mutant with respect to somatic hybridzation will be discussed.

Protoplasts from suspension culture cells were isolated according to the method reported earlier8. Dose response studies were conducted to determine the concentration of potassium chlorate (KClO<sub>3</sub>) which completely inhibited the growth of cells. Protoplastderived colonies (30-35 celled stage) were plated on the medium incorporated with 10, 20, 30 and 40 mM concentrations of KClO<sub>3</sub>. 20 mM KClO<sub>3</sub> totally inhibited growth. Selection pressure was applied at a supralethal dose (40 mM). Resistant colonies were selected after 8-10 weeks of growth in the stress medium. Chlorate-resistant callus was subjected to characterization by testing its capacity to utilize nitrate as the sole nitrogen source in relation to ammonium and other reduced forms of nitrogen such as casein hydrolysate. Regeneration of plants from a chlorateresistant line was obtained.

Assay for nitrate reductase activity was measured by the *in vitro* assay method<sup>9</sup>. Tissue samples included callus and leaves of wild type and chlorate-resistant lines and leaves of *in vitro* grown double mutant plants of *Nicotiana tabacum*<sup>3</sup>. Double mutant of *N. tabacum*, obtained through the courtesy of Dr D. Pental, Tata Energy Research Institute, New Delhi, India was used as standard check.

Chlorate-resistant lines were selected by incorporating 40 mM of KClO<sub>3</sub> as supralethal dose. The screening for chlorate resistance was carried out by plating  $1 \times 10^3 - 5 \times 10^3$  protoplast-derived colonies (Table 1). After eight weeks of incubation on the stress medium, a total of six colonies survived in the first passage. Out of