freezing and thawing, so that eventually the proteins settle down in the form of a flocculum in the distal one-fourth or less of the total column. The junction is not very distinct, but the two layers can be maintained as such if the tubes are left undisturbed at room temperature or in refrigerator without freezing. The contents can be readily mixed by shaking, and the serum so reconstituted stays unchanged but responds like fresh serum if again allowed to freeze slowly as indicated above.

That the clear material at the top is completely devoid of proteins (and of antibodies in the case of immune sera), and that these are concentrated in the flocculum below, has been verified by (*a*) electrophoresis, (*b*) colour reactions, and (*c*) serological tests, such as the agglutinationlysis test for leptospirosis, the capillary agglutination tests for Q fever and for anaplasmosis, and the standard tube agglutination test for brucellosis. The antibody content of the protein flocculum is found to be higher than that of the original serum not subjected to this process of slow freezing and of the serum so treated but reconstituted by shaking. The antibody titres of the protein column, in general, correspond with the degree of sedimentation of the proteins.

Sedimentation of proteins in the form of a flocculum is readily observed in cattle serum owing to its colour, but it occurs equally well with serum of buffaloes, sheep, goats, horses, and pigs. It does not occur if the serum is frozen more repidly at — 10° C. or below. The critical temperature for separation to occur effectively in the test-tubes used would appear to be around — 5° C., a temperature commonly obtained in the freezing chamber of the ordinary refrigerator. Bacterial contamination seems to interfere with freezing and also with proper sedimentation.

The process is simple, inexpensive, and independent of the use of any chemicals. The repeated freezing and thawing involved does not appear to damage the proteins or, at any rate, lower the antibody titre. The method can be used with advantage in diverse diagnostic, therapeutic, and other biological techniques. Indications are that, with suitable modifications, perhaps it can also be developed for separation of the different protein fractions.

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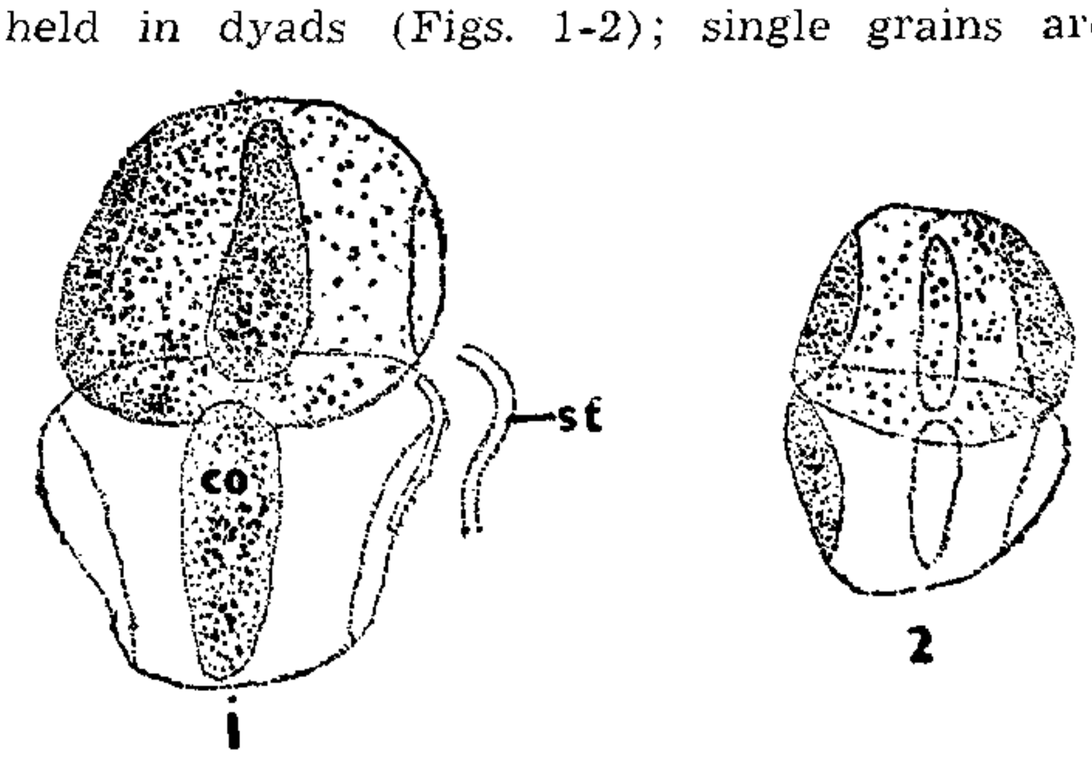
POLLEN MORPHOLOGY OF INDIAN PODOSTEMACEAE

THE Podostemaceæ is an aquatic family, and its systematic and phylogenetic position have been variously interpreted. Bentham and Hooker placed this family in the order Multiovulatæ aquaticæ under the Monochlamydeæ, while other taxonomists placed it under Rosales (Engler and Prantl), Caryophyllales (Bessey), Ranales (Hallier), or Podostemales (Hutchinson; *see* Lawrence[1]).

Pollen morphology of the plants belonging to the Podostemaceæ is little known,[2] possibly due to the difficulties in procuring the polliniferous material. Mention of pollen morphology is also available in embryological literature.[3-6]

The present study includes the following species: *Dicræa stilosa* Benth. Hook. f., *Farmaria metzeroides* Willis, *Podostemon subulatus* Gardn., and *Zeylanidium lichenoides* Engl. Pollen material preserved in FAA has been obtained from Prof. M. A. Razi, Botany Department, Central College, Bangalore (India). Pollen preparations have been made by the acetolysis method.

In all the species studied, pollen grains are held in dyads (Figs. 1-2); single grains are



FIGS. 1–2. Fig. 1. Pollen dyads of *Dicræa stilosa*. Fig. 2. *Zeylanidium lichenoides*. *co*, colpus; *st*, strata.

3-*zonocolpate* (*colpi indistinctly demarcated*; appearing as inaperturate). Exine is very thin (thickness below 1 μ), surface pattern granulose in the different species. The average size measurements of the dyads (longest diameter × shortest diameter) in the different species are as follows: *Dicræa stilosa* (39 × 21 μ); *Farmaria metzeroides* (43 × 29 μ); *Podostemon subulatus* (39 × 25 μ); *Zeylanidium lichenoides* (36 × 21 μ).

Pollen being held in dyads, the Podostemaceæ is unique among the members of the Monochlamydeæ. Erdtman[2] suggested that pollen grains are 3-colporoidate in Weddllinoideæ, 1-porate in Tristicholdeæ (*sensu* Engler), and

3-colpate as well as inaperturate in Podostemoideæ.

The phylogenetic position of the Podostemaceæ has been variously interpreted.[1] The family has been considered to be close to the Saxifragaceæ (Warming; Engler); to be much reduced apetalous types of Saxifragales; or to have affinities with Rosales. The indistinct nature of the colpus in the grains of members of Podostemaceæ might be considered to indicate reduction of colpi from similar forms in the Saxifragales. The Hydrostachyaceæ, allied with Podostemaceæ in the order Podostemales by Hutchinson, have sporomorphs united in tetrads (at maturity) and also inaperturate, as in some of the Podostemaceæ.

I am grateful to Prof. K. N. Kaul for encouragement, and to Dr. M. A. Razi for supplying the pollen material.

National Botanic Gardens, P. K. K. NAIR.
Lucknow, *December* 24, 1964.

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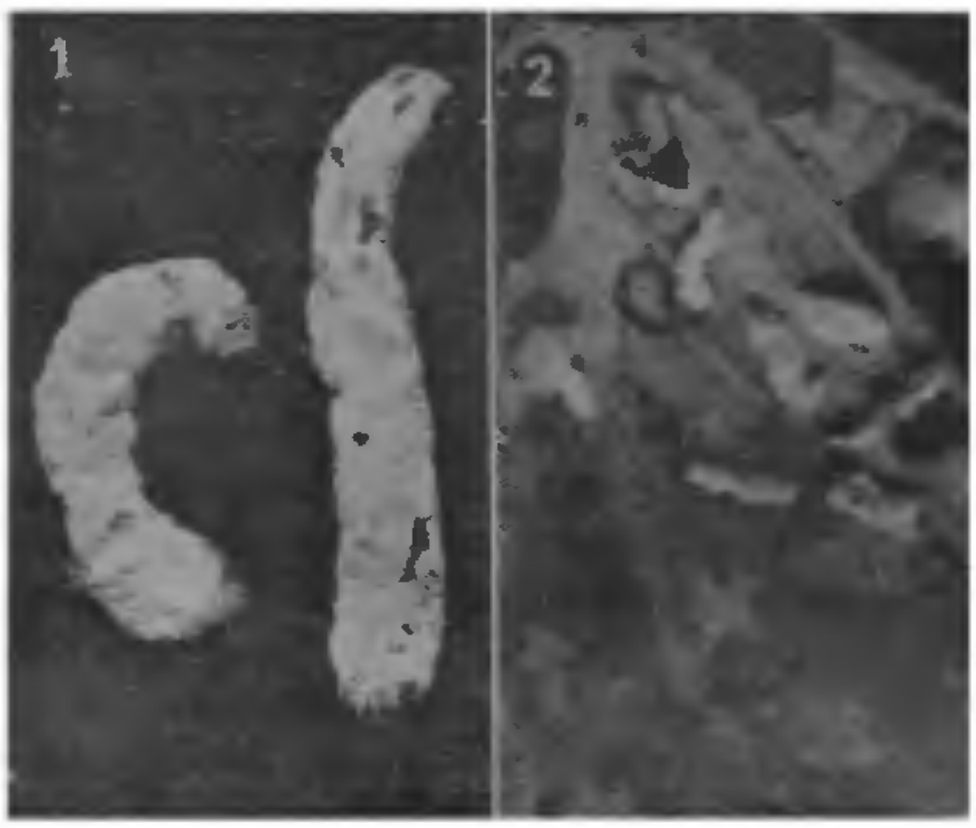
A PRELIMINARY STUDY OF WHITE MUSCARDINE FUNGUS ON CABBAGE SEMILOOPER FROM MYSORE

Beauveria bassiana (Bals.) Vuill., commonly referred to as white muscardine fungus-causing disease of silkworm, *Bombyx mori* Linn., is also a well-known entomogenous fungus of some importance in the control of many noxious insects. Preliminary laboratory studies were undertaken with *B. bassiana* attacking silkworm with a view to find out possibilities of utilising the same against the cabbage semilooper, *Plusia* sp. This is a common leaf-eating semilooper pest on a variety of economic crops like Cabbage, Tomato, Cotton Beans, Lettuce and Alfalfa.

The genus *Beauveria* has been established by Vuillemin in honour of Beauverie who studied the characteristics of the white muscardine fungus, *Botrytis bassiana* Bals., and the related species *B. effusa* Beauv., found on silkworm. He further recommended for the erection of a new genus *Beauveria* to include both these species.[6] Das Gupta[2] was one of the earliest workers to report *B. bassiana* under the name *Botrytis bassiana* from India as the causal organism of the muscardine disease of silkworm. MacLeod,[4] while making a critical examination of the genus *Beauveria*, is of the opinion that there are only two valid species, *viz.*, *B. bassiana* and *B. tenella* (Delacr.) Siem. Several corkers have reported 'fungus disease' other than *B. bassiana* on *Plusia* spp., occurring on various crops.[1,3,5] From review of literature there appears to be no record of any entomogenous fungus on *Plusia* sp. from India.

In the course of the present studies, fungus-affected silkworms which were collected (Fig. 1)



FIGS. 1-2. Fig. 1. Silkworm larvæ infected with *Beauveria bassiana* (Bals.) Vuill., with typical white muscardine growth of the fungus. Fig. 2. *Plusia* sp. caterpillar on damaged cabbage leaves parasitised by *B. bassiana*.

on examination revealed the presence of *Beauveria bassiana*. Dilute chlorine water (1 : 5) was used for surface sterilization to isolate the fungus. After repeated wash in sterile distilled water, the caterpillars were directly placed on the poured plates of Sabouraud maltose media. Few isolations were also made from the internal mycelium of the silkworm aseptically by squeezing with spatula to release the contents of the insects (including internal mycelium) to the petri dish containing a drop of sterilised distilled water. They were further streaked on the poured plates of the same media. Successful growth of the fungus with the characteristic cylindrical-shaped coremia was observed. The conidia observed on artificial culture measured 1·5–2·25 μ. Pathogenecity of the fungus on silkworm was also established in the laboratory.

Caterpillars of *Plusia* sp. were collected and bred into moths in the laboratory. Further generations were reared in the laboratory on cabbage leaves. Rearing was done in 6" diameter petri dishes. Inoculation was done by allowing