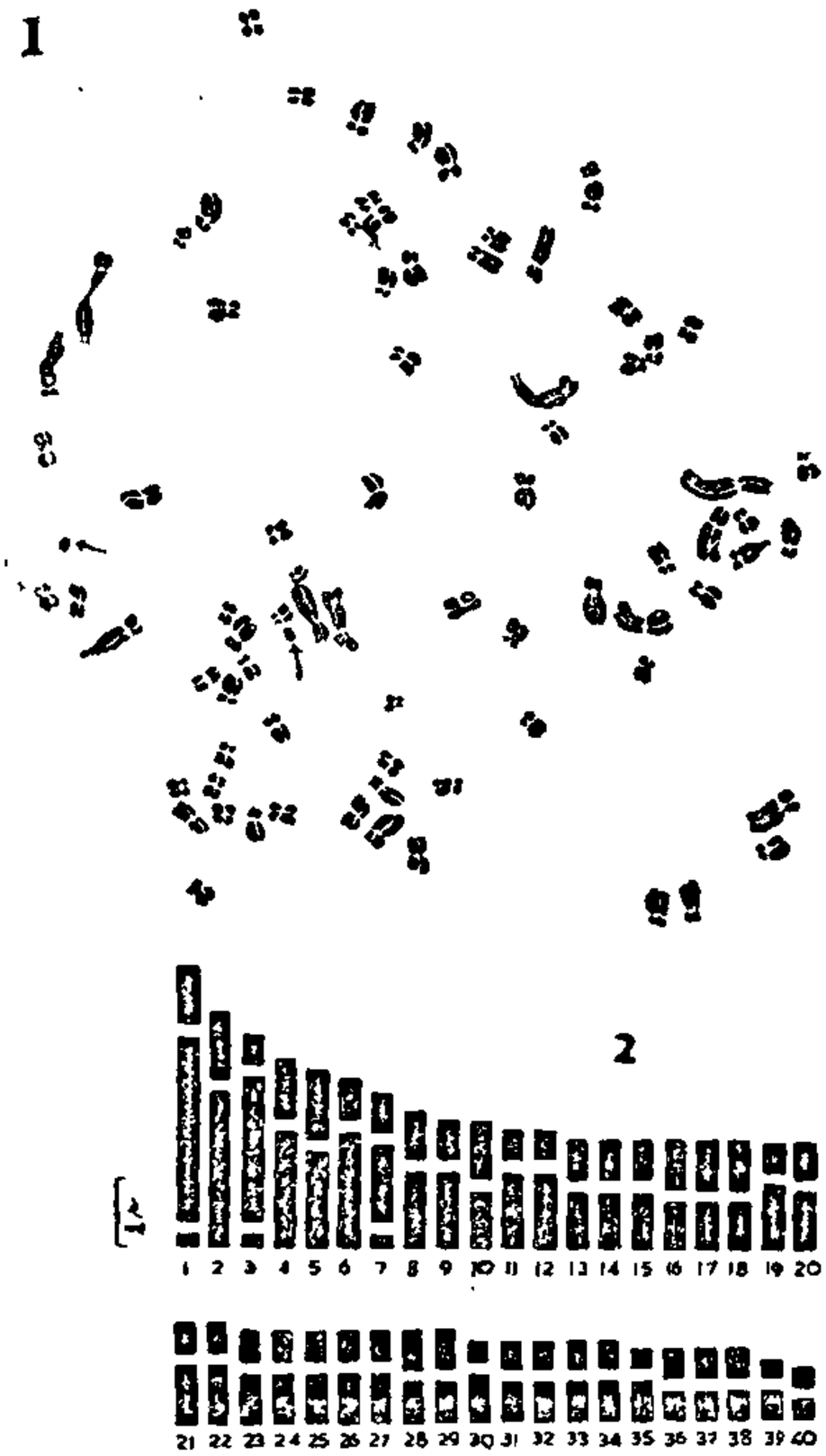
flora, are indicative that this wild species exists at a fairly high polyploid level (Octoploid level) under the 10-basic series and is presumably of allopolyploid origin. Further, the karyotypic data obtained by us for this wild species are in conformity with the data obtained by Sharma and Datta (1960) for the twenty-one species studied by them, and as such do not favour the inclusion of the genus *Dracaena* along with genera like *Agave, Yucca* and *Polyanthes*.



FIGS. 1–2. Fig. 1. Karyotype of *D. terniflora* ($2n = 80$) $\times$ 3,000 $\times$ ½. Fig. 2. Its idiogram.

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TRANSLOCATION OF TOBACCO MOSAIC VIRUS FROM TOBACCO CALLUS CULTURES THROUGH HYPHAE OF *PYTHIUM DEBARYANUM* HESSE

HOLLINGS[1] was the first to report the presence of virus particles in cultivated mushrooms affected with "die back disease". Todate at least five different viruses are known to infect, multiply in cultivated mushrooms and cause severe losses[2]. Later, virus particles have been demonstrated in many other fungi such as *Penicillium, Stemphylium, Laccaria, Alternaria* and *Plicaria*[3-7]. Several plant viral transmissions by fungi belonging to Phycomycetes have also been demonstrated where the virus is merely transmitted without any evidence of multiplication in these fungi[8,2]. Recently, Brants[9,10] has successfully obtained the infection of *Pythium* sp. with tobacco mosaic virus (TMV) by deliberately pouring sterile TMV suspension into culture medium in which the fungus was growing. She also got indications to show that the virus persists in mycelium and might mutiply in the hyphae.

The present study was undertaken to find out whether or not *Pythium debaryanum* could infect the tobacco callus containing TMV, pick up the virus and translocate the virus beyond the source.

Pythium debaryanum Hesse culture was obtained from type culture collection of this Division and maintained on Czapek's agar medium by regular weekly subcultures. The callus cultures were isolated from healthy and TMV-infected tobacco plants and maintained on Murashige and Skoog's medium. Actively growing calli of 15 days old were employed for the present study.

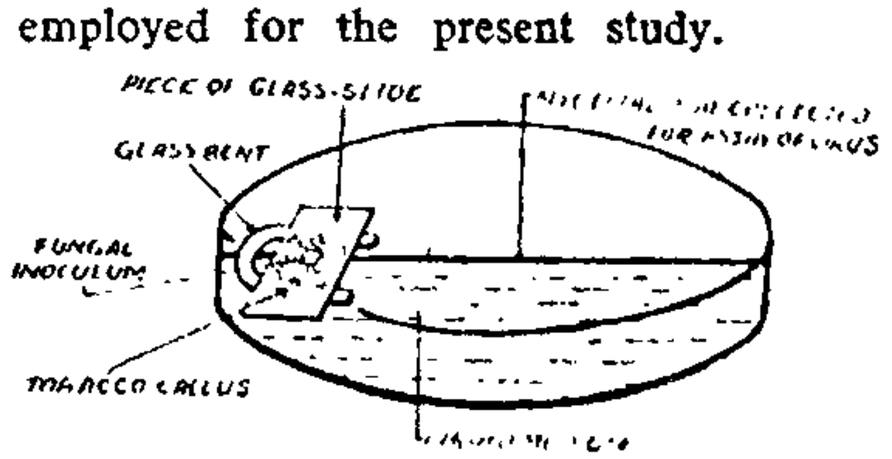

FIG. 1. Diagram of the petriplate used for the study of TMV translocation from tobacco callus cultures through hyphae of *Pythium debaryanum*.

In a petriplate a glass bent and a piece of glass slide were placed to raise the height and make a platform. Healthy or TMV containing tobacco callus was placed on top of slide as shown in the figure. About 15 ml of Czapek's liquid medium

TABLE I

Number of local lesions on C. amaranticolor *leaves after inoculation with macerated mycelium of* P. debaryanum *grown on tobacco callus containing TMV*

No of trial	No. of mycelial mats employed	No. of mycelial mats showing virus	*C. amaranticolor*		Total number of lesions produced
			No. of leaves inoculated	No. of leaves showing lesions	
1	6	6	36	18	30
2	6	5	36	7	20
3	6	5	36	9	32
4	6	3	36	6	21

was poured in the petriplate taking care that the level of the medium stays well below the platform and does not touch the calli. The calli were inoculated with *P. debaryanum* by cutting a small square of agar along with the fungus from culture tube and placing it on top of the callus. After making good establishment with callus the fungus reached below, on to the liquid medium and started to grow further. After 5 days, the fungus on the medium was separated by a pair of scissors at least 1 cm beyond callus without disturbing it. The mycelial mat thus collected was thoroughly washed in several changes of distilled water and macerated in few drops of phosphate buffer solution (pH 7·0) and bioassayed on local lesion host *Chenopodium amaranticolor* leaves for the presence of the virus. Mycelial mat from one petriplate served as one unit. The lesions were counted 5 days after inoculation.

Mycelial extracts of pure culture of *P. debaryanum* did not show the presence of the virus indicating that the fungal culture under study was virus free. Further, the fungus that grew on healthy tobacco callus also did not contain the virus.

In the preliminary tests conducted the virus was detected in the mycelium that grew on calli containing virus. Countable lesions were produced on *C. amaranticolor*, numbering 0 to 6 per leaf. Experiment was therefore, repeated four times to confirm the translocation of the virus through hyphae. Out of 6 mycelial mats employed in each experiment, 3 to 6 indicated the presence of the virus. Although a total of 36 leaves of *C. amaranticolor* leaves were inoculated in each experiment, 6 to 18 leaves have produced local lesions (Table I). However, in all the four experiments the translocation of the virus was demonstrated in the mycelium collected away from the callus.

The results have clearly shown that the fungus infects the tobacco callus containing TMV, picks up the virus and translocates to distances. The microtome sections of the callus revealed that the fungus was intercellular and intracellular. Since the contact between the callus on top of the glass slide platform and mycelium on liquid medium is only through hyphae, it is strongly felt that the virus is being translocated only through hyphae. However, electron microscopic studies are also important to show the virus particles within hyphae. We have no idea how the virus particles inside the callus cells, infect the hyphae and enter. But repeated bioassay of the mycelium collected at a distance from the callus does prove that the virus in the callus is being picked up and translocated. It is not clear whether the virus is merely translocated or it also multiplies in the mycelium. Brants (1971) obtained evidence to show that the virus persists and multiplies in hyphae when *Pythium* sp. was deliberately inoculated with tobacco mosaic virus.

The present investigation clearly demonstrated the translocation of the virus through fungal hyphae. In some of the plant diseases of complex etiology where fungi and viruses are associated such as coconut root-wilt, citrus die-back and mango malformation, the possibility of a similar phenomenon cannot be ruled out.

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