

fungus which has recently been described by the authors<sup>7</sup> as a new species of *Teichospora* (*T. indica*). It was also observed that late in December or early January, the perithecia of *Teichospora indica* were also produced on the same lesions which were developed earlier on account of the infection by *P. cycadina*. The close and constant association of these two organisms suggested the possibilities of some relationship. The present investigation was started to find out the possible relationship between *P. cycadina* and *T. indica*.

Infected leaves of *Cycas revoluta* which contained both pycnidia of *P. cycadina* as well as

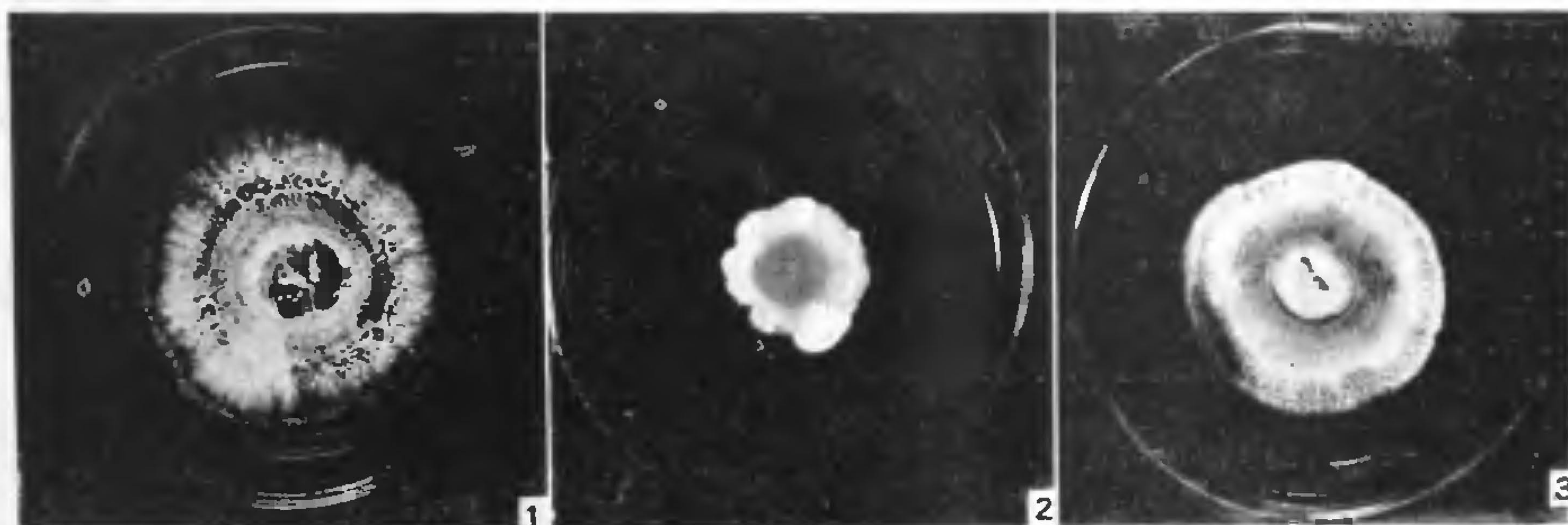


FIG. 1. Sp-A. FIG. 2. Sp-B. FIG. 3. Sp-C.

#### A NOTE ON THE PERFECT STAGE OF *PHYLLOSTICTA CYCADINA* (PASS)

DURING 1956-59 the authors noted that the infected leaves of *Cycas revoluta* bore the pycnidia of *Phyllosticta cycadina* in close association with the perithecia of an ascomycetous

perithecia of *T. indica* were collected in the months of December and January. Small pieces of the leaflets of the host were cut from the junction of healthy and diseased portions which were sterilized and placed in agar slants of potato dextrose. About 100 isolated ascospores of *T. indica* and pycnosporos of *P. cycadina* were cultured separately. The fungal colonies obtained from the above three sources were examined microscopically at different intervals and they were used for subsequent investigations. Artificial inoculations were made on the leaflets of *Cycas revoluta*. All the three types of cultures were used for artificial inoculations. The inoculated leaves were covered with polyethylene bags. Some bulbils of *Cycas revoluta* were placed in nutrient solution and kept inside sterilized glass chambers. The leaflets arising from the bulbils were also artificially inoculated. Besides the above three cultures the germinated ascospores of *T. indica* and germinated pycnosporos of *P. cycadina* were also used for testing the pathogenicity. After three months of artificial inoculation the percentage of infected leaflets was counted and some of the leaflets were microscopically examined for the presence



of pycnidia or perithecia. A preliminary study had revealed that a medium containing dead leaves of *Cycas revoluta* developed the fruiting bodies very readily. The following culture medium was, therefore, prepared. Dead leaves of *Cycas revoluta* 20.0 gm., maltose 10.0 gm., asparagine 2.5 gm.,  $\text{KH}_2\text{PO}_4$  1.75 gm.,  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  0.75 gm., distilled water 1 litre 2.0% agar was added to solidify the medium. Ascospore germination in 0.2% maltose solution was also studied.

Isolation experiments showed that infected leaves of *Cycas revoluta*, as well as the pycnosporos, always yielded *Phyllosticta cycadina* in culture. Cultures started from the ascospores developed pycnidia of *Phyllosticta* in about 15-20 days. They also developed perithecia of *T. indica* after 2 months.

The results of artificial inoculations have been summarized in Table I.

TABLE I

Showing the percentage of infection of *Cycas revoluta* leaflets under different conditions

Sources of inoculum	Percentage of infection
1 Culture obtained from infected leaves	85
2 do. pycnosporos ..	90
3 do. ascospores ..	55
4 Spraying germinated pycnosporos ..	70
5 do. ascospores ..	No infection

Table I clearly shows that ascospores were non-pathogenic, whereas pycnosporos caused infection to about 70% leaflets. Culture obtained from ascospores was also comparatively less effective than the cultures derived from pycnosporos or infected leaves of *Cycas revoluta*.

Experiments on spore germination showed that some of the ascospores started producing the germtube within 6 hours. Over 90% spores germinated between 12 and 13 hours. 100% germination was not attained even after 20 hours. It was observed that practically all the cells of the ascospores were capable of producing the germtube (vide Fig. 1). Some of the mature ascospores germinated even within the ascus. The germtube of such ascospores emerged by piercing the wall of the ascus.

Species of *Phyllosticta* have been reported to have their relationship with several ascomycetous fungi, viz., *Mycosphærella*,<sup>9</sup> *Venturia*,<sup>8</sup> *Guignardia*,<sup>3</sup> *Leptosphaeria*<sup>2</sup> and *Pleosphaerulina*,<sup>1</sup> etc. So far no species of *Phyllosticta* has been found to

be associated with *Teichospora*. Fischer<sup>4</sup> found *Teichospora salicina* in close association with a species of *Chaetophoma* in Australia. Gäumann

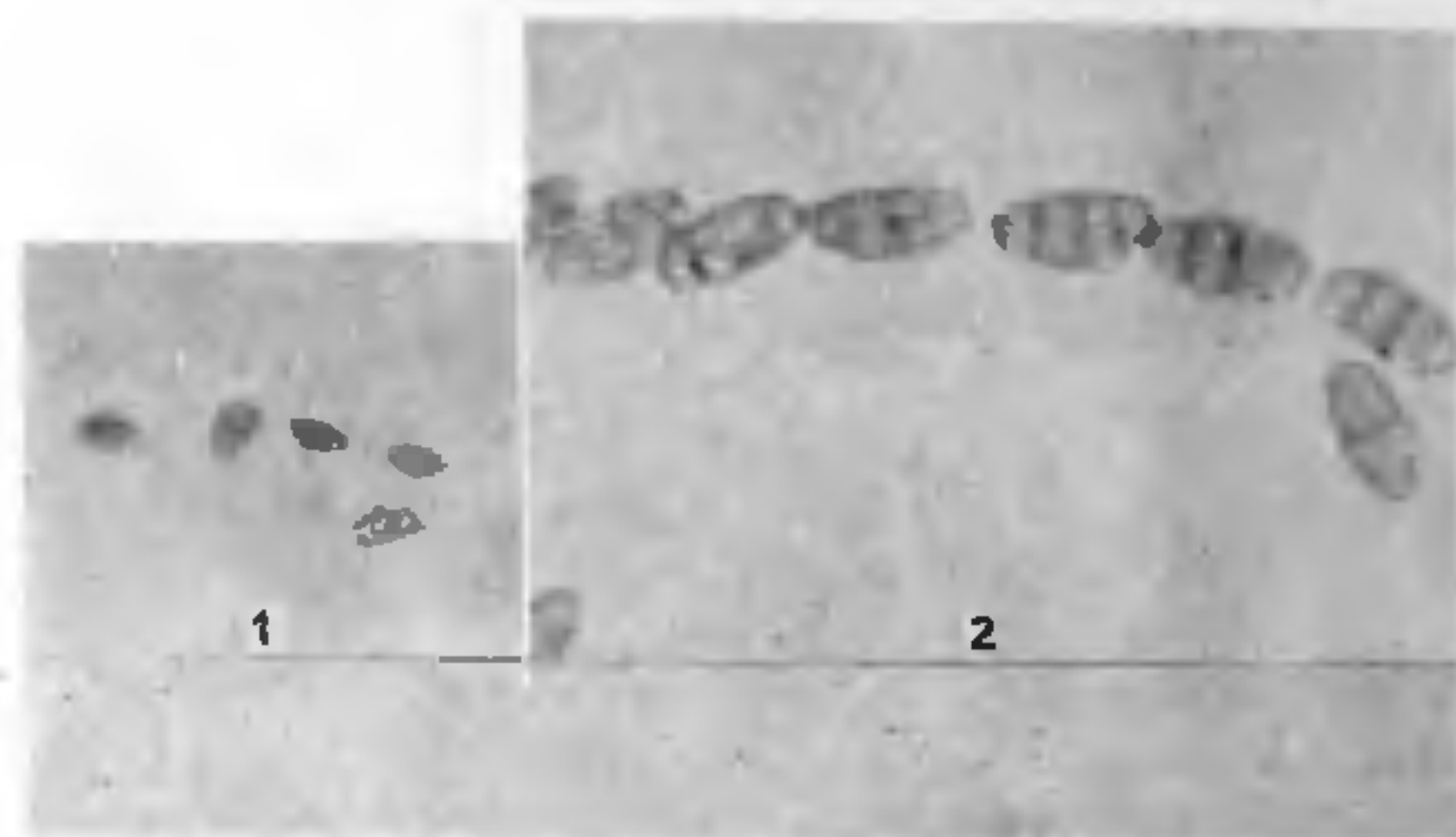


FIG. 1. Showing the formation of germ tubes from different cells of the ascospores,  $\times 347$ .

FIG. 2. Showing the germinating ascospores within the ascus,  $\times 740$ .

and Dodge<sup>5</sup> reported that all possible imperfect forms have been ascribed to the genus *Teichospora* without their appropriateness being culturally determined. In the present investigations it was found that the isolated ascospores of *T. indica* invariably yielded the culture of *Phyllosticta cycadina* on a medium which was prepared with dead leaves of *Cycas revoluta*. It also established that the leaflets of *Cycas revoluta* sprayed with the spores of *P. cycadina* or inoculated with its culture frequently developed the perithecia of *T. indica* when placed under perfectly sterilized conditions. Morphological characters of *P. cycadina* and *T. indica* showed resemblance. The thickness of the hyphae and the distance in their septation was almost similar. It is, therefore, concluded that *Teichospora indica* is the ascigerous stage of *Phyllosticta cycadina*.

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