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Structure and Development of the Gametophytes of Philippine Cheilanthoid Ferns, III. *Cheilanthes concolor* (Langsdorff et Fischer) R. Tryon

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ABSTRACT

The gametophytes of Cheilanthes concolor *(Langsdorff et Fischer) R. Tryon from the Philippines (Quezon City) are no different in pattern of development and in morphology from those from India with one possible notable difference, which is, that the gametophytes under report (which were grown from spores produced from 32-sporate sporangia) are apogamous while those from India (sporangial type of source was not stated) are sexual.*

INTRODUCTION

This paper on the structure and development of the gametophyte of *Cheilanthes concolor* (Langsdorff et Fischer) R. Tryon (=*Doryopteris concolor* [Langsdorff et Fischer] Kuhn) is the third in a series dealing with the developmental morphology of the gametophytes of Philippine cheilanthoid ferns. As far as we have been able to determine, there is only one paper that was published earlier on the morphology of the gametophytes of *C. concolor*, namely, that of Nayar (1960).

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MATERIALS AND METHODS

The spores were obtained from freshly collected fertile fronds of plants growing on slightly shaded adobe wall in the Balara Parks and Filters, Diliman, Quezon City in August 1990 and August 1991.

As with *C. dilimanensis* (Zamora et al., 1993b), *C. javensis* (Zamora et al., 1992) and *C. tenuifolia* (Zamora et al., 1993a), the spores were separated from sporangia (previously scraped with scalpel from the fertile laminae) by sieving through a fine-mesh silkscreen cloth, subsequently sterilized by immersing them in 10–15 per cent calcium hypochlorite solution for 30 minutes through filter paper (Watman No. 25) filtration, placed in suspension by continuous shaking, then washed in sterile distilled water 8 times through continuous filter paper filtration; subsequently, 1 ml of the spore suspension was inoculated on the surface of the culture medium* in petri dishes with a sterile medicine dropper. Finally, the culture plates were taped (to prevent water loss), put in the culture chamber under continuous illumination (light intensity was about 500 foot candles as determined by a foot candle meter sourced from two GE 40-Watt, 120-Volt fluorescent bulbs) and maintained at 25–28° C up to seven months. As many plates as practicable were maintained for periodic microscopic monitoring. Cultures were observed under a compound microscope and a stereoscopic microscope every three to five days up to the appearance of the sporophytes.

Suitable specimens of spores, gametophytes and young sporophytes were photographed at specified intervals under a compound microscope.

Specimen vouchers of spore sources were deposited at the Herbarium of the Institute of Biology, College of Science, University of the Philippines, Diliman, Quezon City.

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* The culture medium was prepared following Zamora and Trasmonte (1987). Thus: 12.5 gm of Bacto Agar (DIFCO) was dissolved in 500ml distilled water over a water bath. To facilitate dissolution of agar, the mixture was stirred with Kimax Number 5 Magnetic Stirrer. To this were added and thoroughly mixed with agar the following nutrient sources (previously hydrated to 10ml distilled water): ammonium nitrate, 0.25gm; potassium phosphate, 0.10 gm; magnesium sulfate, 0.10 gm; calcium chloride, 0.05 gm; ferric chloride, 0.10 gm. Agar solution was allowed to boil for five minutes until the solution cleared up. The pH was adjusted to 5.5–5.8. The solution was sterilized by autoclaving a 15 psi for 15 minutes, poured into petri dishes, covered and then allowed to solidify. Petri dishes and glasswares used were sterilized by autoclaving at 15 psi for 15 minutes.

RESULTS AND OBSERVATIONS

The spores are globose as may be gleaned from Figure 1 with line-like triradiate marks. Sourced from 32-sporate sporangia, they are 30 x 35 microns in diameter. The germinate within 8–10 days from sowing. They following changes were observed from the first day through 180 days (6 months) of sowing:

Days 1 through 15.

(1) Initially, the spore content enlarges causing a crack at the triradiate mark, the spore coat subsequently splitting into three valves (arrows in Figure 2), accompanied by the emergence of a rhizoid at the posterior end (Figure 2) and of the germ papilla at the anterior end (Figure 3). As may be gleaned from Figures 2 and 3, the rhizoid is slightly dilated at the base (Figure 2) and the germ papilla is densely chlorophyllous (Figures 2 and 3).

(2) Subsequently, the germ papilla divides (the new walls oriented essentially perpendicularly vis-à-vis to the axis) giving rise to two derivative cells (Figure 4).

(3) Then the two derivative cells themselves divide 1 to 2 times in succession giving rise to 4–6-celled germ filament respectively whose component cells are broader than long. The foregoing changes occur within 8–10 days from sowing.

(4) Next, the cells of the germ filament, except the basal one, divide longitudinally, i.e. the new walls perpendicular to the long axes of the component cells, as may be gleaned from Figure 5.

(5) Following the close series of divisions and concomitant enlargement of the component cells vis-à-vis the so-called *Adiantum* type of prothallial development (*sensu* Nayar and Kaur, 1969, 1971), the germ filament is transformed into a spatulate structure by the activity of an apical cell (Figures 6, 7) 15 days from sowing.

Days 16 through 50

(6) As a result of the rather active growth of the anterior end of the rapidly developing gametophyte, the apex gradually becomes notched (Figures 8, 9) with the obconical apical cell occupying the bottom of the shallow notch (Figure 9).

(7) Subsequently, the gametophyte becomes prominently cordate and the apical cell becomes replaced by a multicellular meristem that is composed of several narrow columnar cells.

(8) At 50 days from sowing, the gametophyte is cordate, naked, broader than long with a thin central midrib bearing semi-circular wings (that are slightly overlapping as may be seen in Figure 10) on either side of the notch.

Days 51 through 110

(9) The gametophytes produce antheridia among the rhizoids within 75–95 days from sowing and archegonia immediately below the apical notch much later, i.e. within 95–110 days from sowing. Figure 11 is a highly magnified view of an antheridium containing sperms and Figure 12 is a highly magnified view of an archegonium containing an egg.

Days 111 through 180

(10) The gametophytes produce sporophyte directly from cells just below the apical notch, i.e. part that normally producse archegonia (Figure 13). Figure 14 is a photograph of a 6-month old culture containing several apogamous sporophytes still associated with their respective gametophytes.

DISCUSSION

Our findings on the structure and development of the gametophytes of *Cheilanthes concolor* from the Philippines (Quezon City) are essentially the same as those of Nayar (1960) from India with one notable difference: our material is apogamous while that of Nayar (1960) is sexual. The 32-sporate character of the sporangia of our material correlates well with its apogamous nature (inferred earlier by Zamora, 1978). The type of sporangium of Nayar's material is not known, but its sexual nature indicates that its sporangia may have been of the 64-sporate type. This is entirely possible for, as Anthony (1984) has shown, populations of *C. concolor* that produce 64-sporate type of sporangium do exist (in South Africa). It is thus possible here, based on the foregoing facts, to recognize two forms of *C. concolor* as follows:

(1) a form that produces 32-sporates type of sporangium and exhibits obligate type of apogamy (Philippines, this paper)* and
(2) a form that produces 64-sporate type of sporangium and exhibits sexual type of reproduction (India, Africa) as per the reports of Nayar (1960) and Anthony (1984).

Earlier, Tryon (1968) reported the existence of apogamous as well as sexual races in the same species of cheilanthoid fern, i.e. *Pellaea glabella.*

C. concolor is one of two species of *Cheilanthes* that is amphiatlantic in geographical distribution occurring in Africa, Asia, Malaysia, northern Australia, Pacific Islands, Central and South America (Tryon, 1942; Tryon and Tryon, 1973). It would be interesting to find out at some future time if the foregoing observations are biogeographically related.

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* It may be stated here as was done for *Cheilanthes tenuifolia* (Zamora et al., 1993a) that the mere production of 32 spores per sporangium has been reported for *Cheilanthes catenensis* and *Ceratopteris thalictroides* (see Zamora et al., 1993a). It is thus still necessary to germinate spores, count chromosomes and raise gametophytes to prove beyond doubt the true pattern of reproduction (whether sexual or apogamous) of a species in question.

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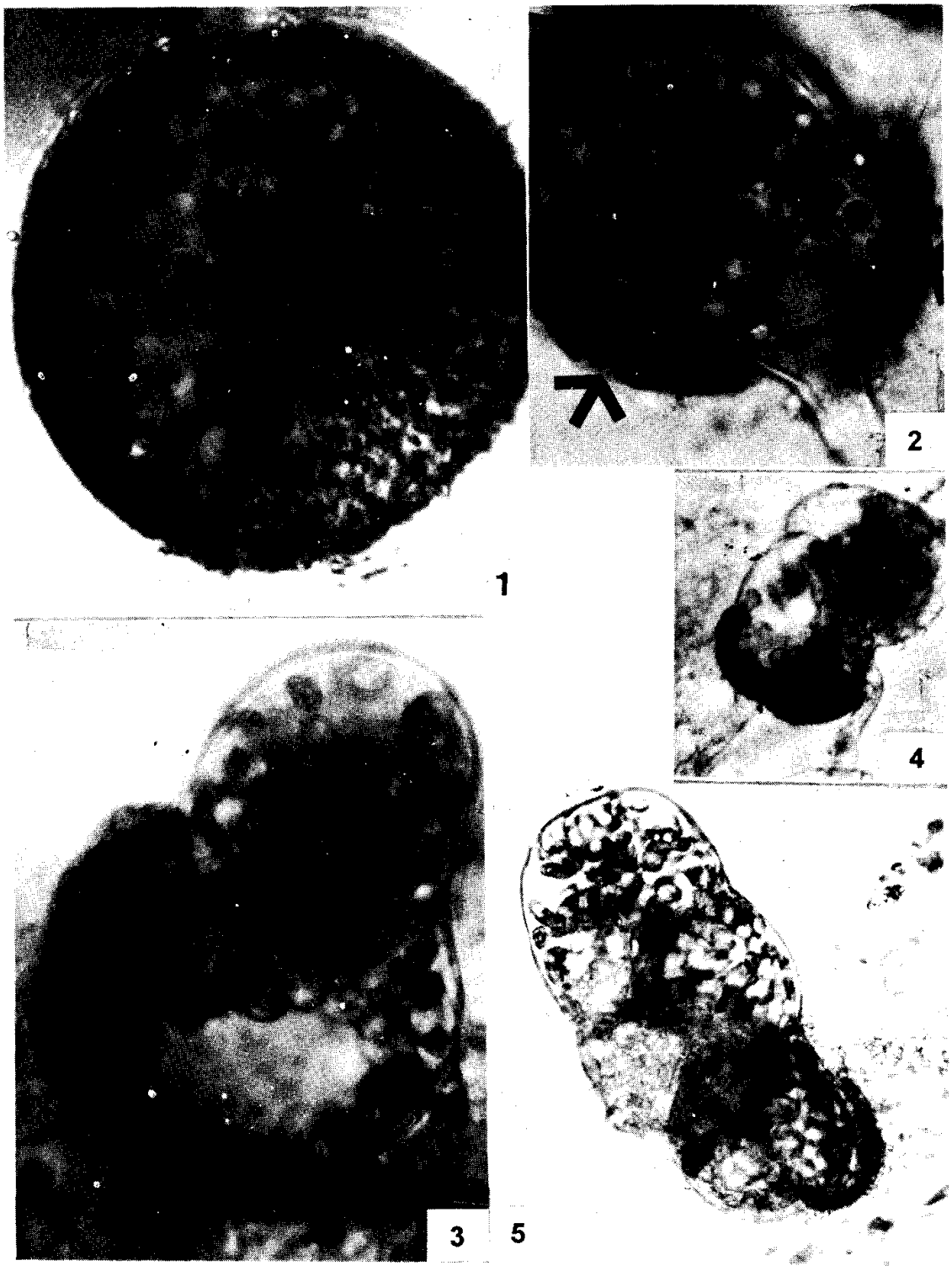
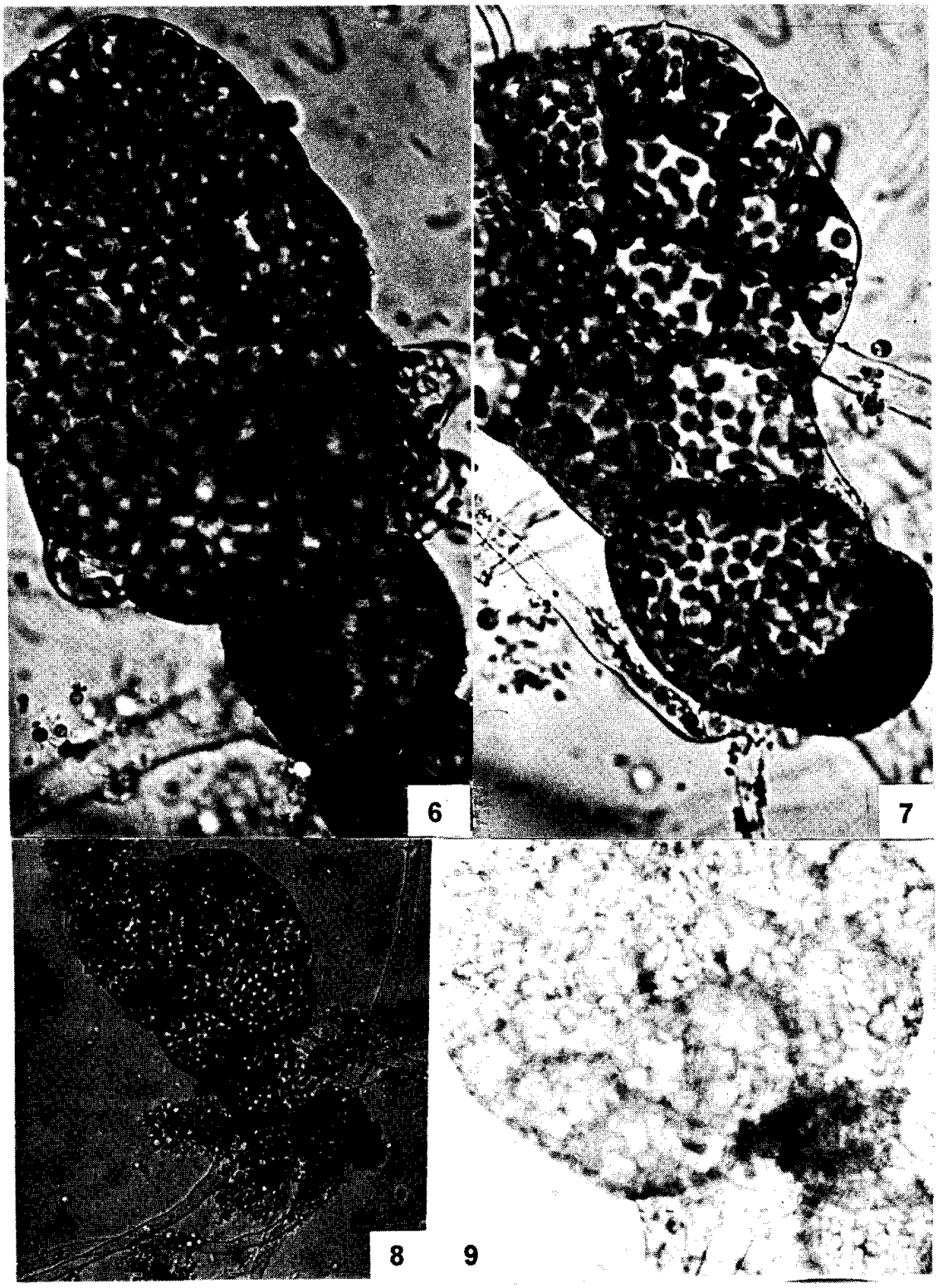


Structure and development of the gametophyte of Cheilanthes concolor *(Langsd et Fisch) Tryon*



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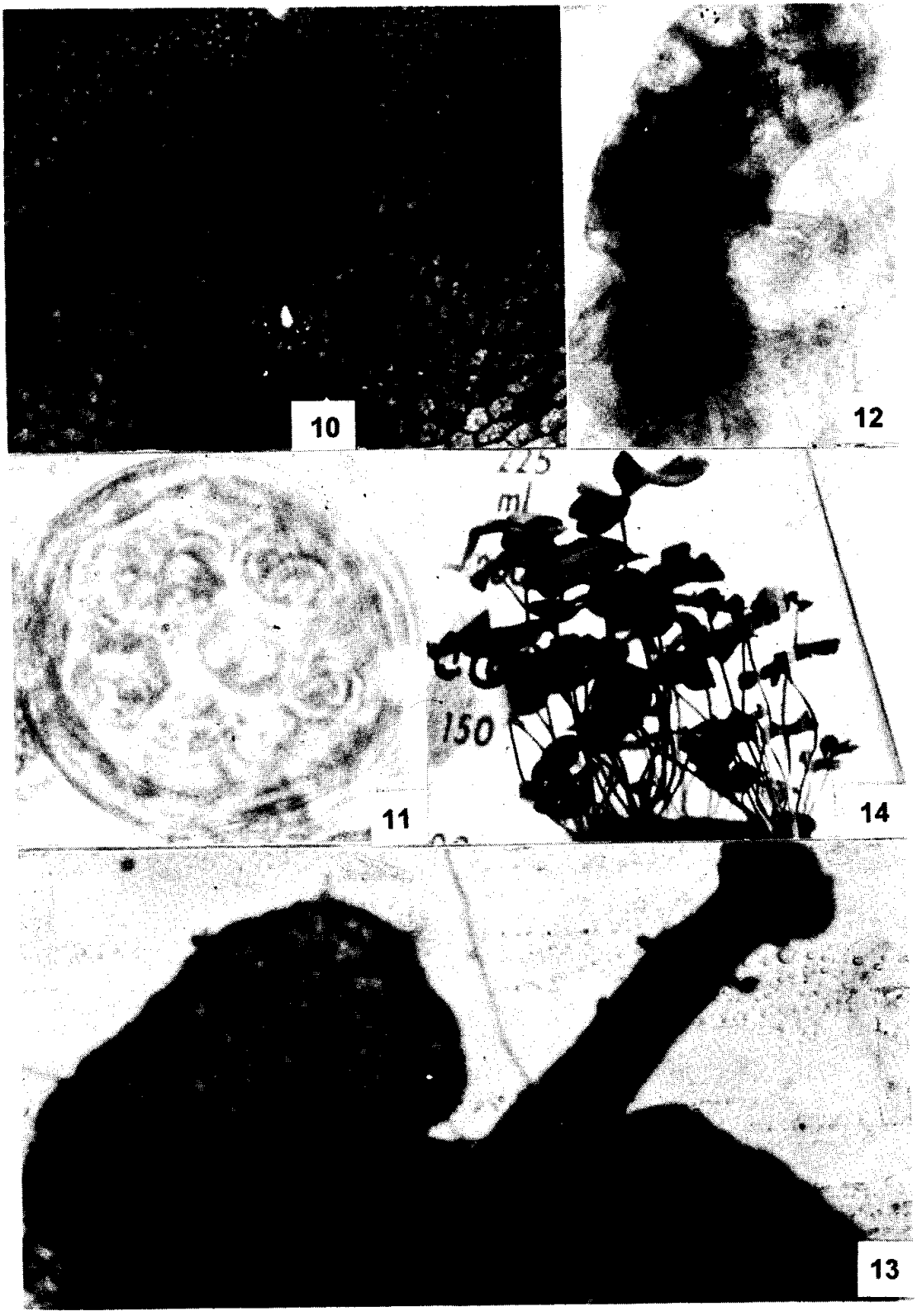

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