

be the interspecific competition of space among biota along such a thermal gradient.

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### A LARGE-SIZED NEW SPECIES OF AMOEBA, *AMOEBA NAVINA* SP. NOV FROM TROPICAL RIVERS

This laboratory has been successful in cultivating a new species of large-sized amoeba, *Amoeba navina* sp. nov. collected from Mula-Mutha Rivers since 1967. The amoeba is visible to the naked eye. It ranges in length from 0.5 mm to 0.9 mm. However, as large as 1.2 mm sized forms have been noticed in petri dish cultures.

This new species, *Amoeba navina* sp. nov., has a distinct limiting membrane, hyaline ectoplasm and dark endoplasm. The latter contains numerous small spherical elongated vesicles. These vesicles appear dark in subdued illumination under the low power of the microscope. Besides these vesicles, the endoplasm also includes numerous food vacuoles, clear vesicles, single water expulsion vesicle and single spherical, oval or elongated polymorphic nucleus, lacking an endosome. Less than 1% population of *A. navina* sp. nov. shows binucleate condition, representing one of the stages of binary fission. Pseudopods are distinctly tubular, lobose and have clear tips. During normal locomotion, the amoeba shows one or two blunt pseudopods which determine the direction of its movement. The remaining pseudopods, when present, are either stationary or retracting. For quick unidirectional loco-

motion, the amoeba gives out only one pseudopod. Such a monopodal form of *A. navina* appears club-shaped; the blunt end being in the direction of locomotion. In unfavourable medium, these amoebae become thin, elongated and give out long 6 to 12 tubular pseudopods and the whole structure resembles a twig, or a branch of a tree.

*A. navina* feeds voraciously and favours Paramoecia as food. It also takes Rotifers, Chilomonas, Tetrahymena, Colpidia, Vorticella, eggs of worms, Arcella, as well as smaller amoebae of species other than its own.

*Amoeba navina* sp. nov. has been maintained in modified petri dish cultures during the last 3½ years. These amoebae were found to be comfortable between 16° C and 39° C carrying out all the vital functions of life, including feeding and fission. Curiously indeed they have survived the heat wave of May 1970, when amoeba medium temperature rose to 41° C! So also they have lived through the cold wave during the winter of 1970, when amoeba medium temperature dropped to 16° C.

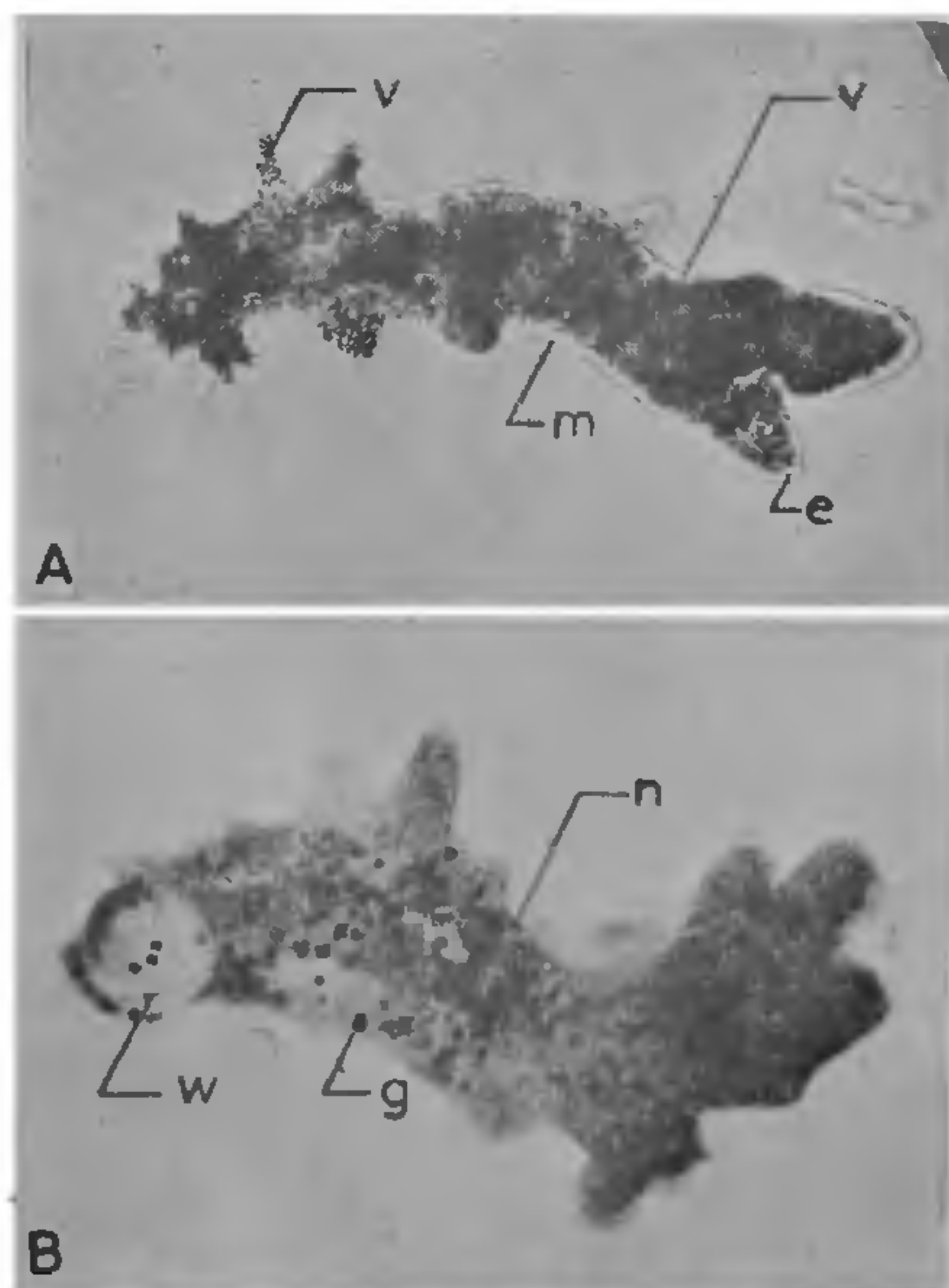


FIG. 1. A. *Amoeba navina* sp. nov., a living specimen during normal mode of locomotion, showing clear ectoplasm (e), outer membrane (m), and the dark endoplasm containing numerous small vesicles (v). Subdued light photomicrograph, × 130. B. *A. navina* sp. nov.—Hematoxylin and eosin stained preparation showing nucleus (n), granules (g), and water expulsion vesicle (w), × 130.

This amoeba has certain similarities with *A. nitida* (Penard<sup>1</sup>), *A. proteus* and *Chaos carolinensis*.<sup>2</sup> Like other Amoebidae, e.g., *Amoeba proteus*, *A. nitida*, it possesses clearly defined ectoplasm, granular endoplasm and tubular pseudopods with clear caps for locomotion. *Amoeba navina* sp. nov. happens to be the largest mononucleate form among Amoebidae known so far. Superficially it looks like the multinucleate form, *Chaos carolinensis* in respect of size, its feeding habits, general structure and formation of pseudopods, but differs from the latter in respect of possessing single nucleus, its ability to withstand tropical temperature variations and the lack of endoplasmic crystals. On the other hand, the largest mononucleate forms known so far are *Amoeba nitida* (Penard<sup>1</sup>) and *A. proteus*; both found in freshwaters. These are smaller than *A. navina* sp. nov. *A. navina* always gives out either one or two functional pseudopods at a time, while *A. nitida* shows three principal pseudopods. In monopodal forms, *A. navina* sp. nov. has a blunt end in the direction of its locomotion, while *A. nitida* has pointed tubular end in the direction of monopodal locomotion. *A. nitida* possesses narrow truncated bipyramidal crystals in independent vesicles. Such crystals are lacking in *A. navina* sp. nov.

The branched structure of *A. navina* giving out several slender, tubular pseudopods is the unique characteristic. Its fairly large, mononucleate structure showing lack of crystals in its endoplasm, and its ability to withstand tropical temperature variations between 16° C and 41° C, are the principal characters which have led to the classification of this animalcule into a new species, *Amoeba navina* sp. nov.

This one-celled large animalcule is also easy to cultivate and maintain in tropical laboratories. This makes it an ideal material for routine class work and cell biological research.

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### CELLULOSE POWDER AS LEGUME INOCULANT BASE

PEAT is the preferred carrier material in the manufacture of legume rhizobia inoculants. Superiority of peat over other carrier materials is due to (i) high protection it offers to the culture against high storage temperature, (ii) low moisture loss during storage, and (iii) the presence of certain essential nutrients which not only help survival of the organism but also support further multiplication during storage.

High-grade peat is not available in India but only small deposits of low-grade peat occur in the Nilgiri Hill ranges<sup>1</sup>. Initial studies carried out by Iswaran *et al.*<sup>2</sup> indicate that Indian peat could be satisfactory as inoculant-base.

In our preliminary comparative studies on suitability of soil, of coconut-shell powder and cellulose powder as substitutes for peat, cellulose powder appeared to be promising both from the point of view of the survival of rhizobia and the ease with which the seed could be treated. As an extension to these studies, survival of rhizobia in the inoculant during storage and on the treated seed, and the adequacy or otherwise of the surviving number to induce nodulation in legume roots were examined.

Soyabean-specific *Rhizobium* strain S-12 from the collection of the Department of Microbiology, University of Agricultural Sciences, Bangalore, was used in these studies. Four-day-old yeast-extract mannitol agar grown bacterial cells were suspended in sterile water containing 0.1% carboxyl methyl cellulose and the suspension was mixed with sterile cellulose powder of 200 mesh fineness. The inoculant powder thus prepared was divided into 12 lots and sealed in polythene bags. One set of six bags was stored at refrigeration temperature of 10° C and the other lot of six was stored at room temperature (28°-30° C). Inoculant samples were withdrawn at periodical intervals of six days over a period of 24 days and the surviving number of rhizobia was determined by dilution-plate-count method using yeast-extract-mannitol-congo-red agar. A portion of the sample withdrawn was used to treat the soyabean seed (variety Hill). The seed was moistened with water, rolled in the inoculant powder and was dried in shade for about 15 minutes. The number of rhizobia per treated seed was determined by washing five treated seeds in an aliquot of sterile water and plating appropriate dilutions of the latter in yeast-

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