

experiments up to 15.4 cm./sec., which is equivalent to about twenty times the animal's length in a second.

Light-adapted *Mysidium* are positively phototactic and swim towards a source of light. Dark-adapted ones, however, always swim to the bottom of the tank when illuminated irrespective of the position of the light. After removal of the pair of statocysts situated in the uropods they always swim away from a light source, to the surface when illuminated from below and to the bottom when illuminated from above.

The behaviour of *Mysidium columbiae* is remarkable in several respects. Unlike most mysids it is a 'bright-light' animal living close to the water surface under the noonday tropical sun, while in its shoaling behaviour it shows a precision and co-ordination of locomotion comparable with that seen in fish. In some respects, however, the social responses of *Mysidium* are not so well developed as in shoaling species of fish. Two or more fish will form a shoal instantly but small numbers of mysids will not. It seems that critical density or number of animals is required before they will form shoals, which then build up rather slowly, taking from 30 sec. to 5 min. to become well developed. Also a proportion estimated at about 5 per cent do not join the shoals, but swim freely in other directions or remain motionless on the bottom. *Mysidium* is more suitable than fish in some ways for an experimental study of shoaling, since it shows no apparent capacity to modify its behaviour as a result of experience.

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### Evidence for a Sexual Phase in the Life-Cycle of an Amœba

THE amœba have generally been thought to be devoid of a sexual phase in their life-history<sup>1</sup>, though one earlier report<sup>2</sup>, and a recent one<sup>3</sup>, indicate a cycle involving fusion of flagellated gametes. This communication concerns evidence for sexuality in a diphasic amœba.

A flagellate ascribable to the genus *Heteramœba* and very near *H. globosa* Skuja<sup>4</sup> is a widespread, though by no means common, inhabitant of brackish supra-littoral rock pools. This organism, referred to here as *Heterochromonas clara* nom. prov., differs from *H. globosa* in being larger (30 $\mu$  diam.) and in not having contractile vacuoles, but, like *H. globosa*, it has no stigma. It was abundant in August 1960 in the polluted pools at the base of the cliff below the large gannetry on Ailsa Craig, where it was feeding voraciously on green flagellates. Two dozen bacteria-free clones were isolated from this source using a strain of *Brachiomonas submarina* for food. One clone was set aside for nutritional work and the remainder were maintained in composite culture for reference. The amœbæ first appeared in the latter culture. They were unexpected since *Heterochromonas* is not recorded as being the stage in the life-cycle of an amœba; moreover, an isolate of *H. clara* from a similar habitat in Finland had been kept for ten years without sign of amœbæ.

Several of the amœbæ were isolated and washed singly and used to start cultures. Without exception every one of these cultures quickly gave rise to flagellates. Twenty flagellated cells were similarly

isolated from one of the amœba isolates (145), the ensuing clones being designated 145/a-t. Amœbæ appeared in several of these within a few days, but others have given rise to no amœbæ after four months of maintenance. The latter, six in number, were then crossed with each other, that is, mixed in subculture two at a time in all combinations, for it seemed that the flagellate/amœba transformation might, in contrast to other diphasic amœbæ, be under genetic rather than environmental control. So indeed it proved, for with certain combinations amœbæ appeared within 24 hr. of the operation, while with others, and in the six parent clones, the populations remained uniformly flagellate (Table 1). Furthermore, the pattern of amœba-formation is consistent with the existence of two mating types among the clones, all of which, it will be recalled, are descended from the same amœba cell. This result is taken as indirect, though conclusive proof of a sexual phase in the life-cycle of this organism.

Table 1. PATTERN OF AMŒBA-FORMATION IN BI-CLONAL CULTURES

Clone	145d	145k	145r	145f	145i	145n
145d	—	—	—	A	A	A
145k	—	—	—	A	A	A
145r	—	—	—	A	A	A
145f	A	A	A	—	—	—
145i	A	A	A	—	—	—
145n	A	A	A	—	—	—

A, Formation of amœbæ within 24 hr.

The amœbæ are at first minute (7 × 30 $\mu$ ) and typically *limax*-like but later reach a size of 40 × 70 $\mu$ ; they have been observed to multiply by binary fission. Cysts (up to 25 × 30 $\mu$ , smooth-walled) are found only in amœba cultures; they germinate to produce single amœbæ. Possible isogametic fusions between minute flagellates can be observed in mixed cultures, but this is open to an alternative interpretation since *H. clara* is phagotrophic and able to devour cells of its own size or larger and is not above resorting to cannibalism.

The following hypothetical conclusions may be drawn from these preliminary observations: (1) the amœbæ are diploid; (2) flagellated cells may be diploid or haploid; (3) only diploid cells can encyst; (4) haploid cells are of two mating types; (5) multiplication can occur both in the diploid and haploid states and can continue indefinitely in the latter.

Multiplication of flagellated cells of diphasic amœbæ is not unknown; it has been reported, for example, for *Tetramitus rostratus*<sup>5</sup>, together with the suggestion of a sexual phase. It is not yet established that amœbæ of *H. clara* can multiply for an indefinite period without reversion to flagellates; nevertheless, considerable fluidity appears to exist in the balance between haploid and diploid phases of the life-cycle, the cycle being primitive on that count as compared with other sexual Sarcodina.

A cytological investigation of *H. clara* is being undertaken as an aid to a proper diagnosis of the organism and to a more detailed description of the life-history.

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<sup>1</sup> Jepps, M. W., *The Protozoa, Sarcodina* (Oliver and Boyd, London, 1956).

<sup>2</sup> Jones, P. N., *Arch. Protistenk.*, **43**, 322 (1928).

<sup>3</sup> Cigarda, L. N., *Inst. Lombardo (Rend. Sci.)*, **92**, 431 (1958).

<sup>4</sup> Skuja, H., *Symb. bot. Upsaliens.*, **9** (3), 1 (1948).

<sup>5</sup> Bunting, J., *J. Morph.*, **42**, 23 (1926).