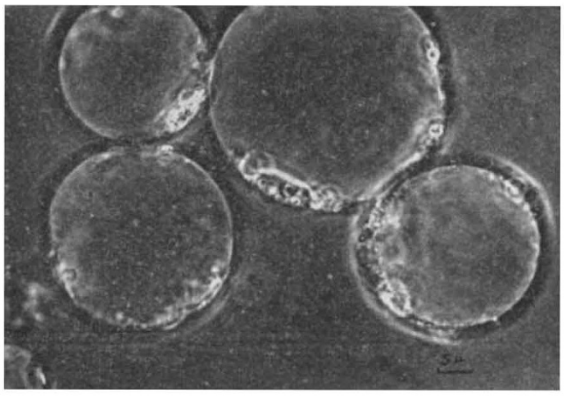

Fig. 3. Vacuoles (phase contrast)

spherical form, both in cellulase-treated root tips and in cellulase-treated single unvacuolated cells isolated from root tips by the action of ethylenediamine tetraacetic acid[9].

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ENTOMOLOGY

Bacteria as a Source of Food for Black-Fly Larvæ

Black-fly larvæ (Diptera : Simuliidae) are almost entirely confined to flowing water. They have long been known as 'filter-feeders', but the minimum particle-size acceptable as food by the several instars has not been previously investigated. Several authors have reported the occurrence of larvæ in sewage-contaminated waters ; but Petersen[1] was apparently the first to suggest that they might feed on suspended particles as small as bacteria.

This hypothesis is under investigation at Saskatoon in an attempt to determine some of the causes of destructive outbreaks of the black-fly, *Simulium arcticum* Mall. Larvæ of this species are sometimes excessively abundant in the north and south branches of the Saskatchewan River which originate in the Rocky Mountains of western Canada and discharge up to 140,000 c.f.s. into the confluence. During the spring and early summer, when these larvæ are most abundant, this river is generally in flood and the water very muddy with up to 1,000 p.p.m. of suspended solids. At the same time, macroplankton, the usual food of black-fly larvæ, are relatively scarce but bacteria are abundant. Species of bacteria present indicate an origin in sewage from the several cities situated on the river as well as from soil erosion.

In preliminary experiments at Saskatoon an attempt was made to rear first-instar larvæ of *S. vittatum* Zett. and *S. venustum* Say in sterile solutions of soluble nutrient in water flowing at 0·3–1·4 ft. per sec. In the absence of discrete particles these larvæ failed to develop beyond the second instar[2]. In similar, but non-sterile, solutions the larvæ developed to maturity, and adults were produced. Five species of bacteria, including three commonly found in soil and water, were isolated by Dr. G. Dempster, University of Saskatchewan, Saskatoon.

In further experiments 90 adults of these two species of black-flies were reared from 1,360 first-instar larvæ in distilled water containing only a washed suspension of *Bacillus subtilis* (Ehr.) Cohn, a normal component of soil. Fresh bacteria were added at 2- to 3-day intervals, keeping populations at about 160,000 viable bacteria per ml. of aquarium water. Microscopic examination showed that the bacteria were not clumped, and that most were present as single cells rather than in chains. About 65 per cent of the larvæ were lost in the first instar, possibly as a result of injuries received in handling. Rate of development was comparable to that in Nature : 21 days from hatching to the first pupa and a further 6 days to the first adult. These investigations are continuing and a detailed report will be published later.

Laboratory experiments have thus far been confined to larvæ of *S. vittatum* and *S. venustum* because of the difficulty in providing rapidly flowing water as in aquaria required by *S. arcticum*. However, the mouth-parts of these three species are very much alike, and thus similar results with the latter species may eventually be obtained.

The fact that all instars of larvæ of *S. vittatum* and *S. venustum* can obtain adequate nutriment from particles as small as *B. subtilis* is significant from two points of view : (1) Sewage bacteria entering a river may serve as food for black-fly larvæ. Thus the severity of black-fly outbreaks originating in rivers such as the Saskatchewan, Mississippi, Danube, Leine, Nile, etc., may be expected to increase as the amount of sewage entering them is increased. Reports[3,4] and personal observations indicate that livestock losses as a result of black-fly outbreaks along the Saskatchewan River have increased in the past 40 years. Enigk[5] reports that black-flies are appearing in larger numbers in Germany, and suggests that this is due to increased soiling of brooks and rivers. Losses adjacent to the Danube River appear to have been increasing during the past two hundred years[6]. (2) This suggests an unusual food-chain in the Saskatchewan River and perhaps other rivers which seasonally contain relatively little macroplankton but considerable bacteria and numerous filter-feeding black-fly larvæ. Fish, including *Moxostoma aureolum* (Le Sueur) and *Hiodon alosoides* Raf., containing black-fly larvæ were collected from the Saskatchewan River. Numerous larvæ-eating fish are known from the Danube[6].

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