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Introduction

When I was a high school student in the early 1930's, the brawniest tackle on our football team -- after an energetic afternoon of swimming and diving in our suburban city pool one, hot, summer day -- was carted off by ambulance to the hospital that evening with a splitting headache and a rapidly rising fever. He died about three days later. The official verdict -- "atypical meningitis" -- atypical, because no bacteria of the *meningococcus* sort could be found in his brain on autopsy.

Our family shook in trepidation, for my sister -- an athletic girl -- had been doing tandem stunt-diving with that youth the day he became ill; but nothing more happened. Neither she nor anyone else who had swum in that pool on that day, nor anyone else in town contracted meningitis.

What killed our muscular football player? No one then knew; but now I suspect, knowing of biological and medical researches on it for the past 20 years, that he was killed by a small amoeba, which invaded his brain and "blew his mind".

Until the mid-1950's it was assumed by biologists and physicians that the only amoeba that endangered human comfort is the one that infects the digestive tract, causing amoebic dysentery in the susceptible and unwary tourist exposed to its more virulent varieties. Although that one, known to biologists as *Entamoeba histolytica*, sometimes escapes to the other parts of the body to cause sores and disturbances, it rarely causes death of its host, being a generally "well-behaved" parasite.

Acanthamoeba

During the search for viruses in the 1950's, especially "polio" viruses, other amoebas of lethal portent were discovered, masquerading as viruses until their "masks" were torn away by Dr. C. G. Culbertson of the Indiana Medical School and his staff, in cooperation with the Indianapolis laboratories of the Eli Lilly Company. Culbertson and his co-workers (5) found that an amoeba that grew in kidney-tissue cultures (as viruses also do) could cause a fatal, virus-like meningitis in laboratory mice and monkeys!

The most potent route of invasion by the amoeba, Culbertson found, is the mucus lining of the nose, by way of which the amoebas dissolve and gobble their way up the olfactory nerves to the frontal lobes of the brain. There, they rapidly produce (in a day or two) a meningitis that is quickly fatal, by killing and devouring brain tissue.

The amoeba was sent to Dr. E. C. Bovee, an amoeba-specialist then at the University of Florida, who identified it as of the genus *Acanthamoeba*, closely related to the common "soil" amoeba, *Acanthamoeba castellanii* (named after the British-Italian microbiologist, Aldo Castellani, who first discovered it).

What had caused a small, innocuous, bacteria-eating amoeba to turn into a virulent, lethal, tissue-destroying monster? Nobody knew.

In rapid succession, however, researches of the 1960's showed many things.

Dr. Bovee and his staff showed that the virulent *Acanthamoeba* could enter the body —— of rats and mice, at least —— anywhere a wound might occur, especially if bacteria also invaded the wound. Those mice and rats then usually suffered kidney, liver, spleen and lymphatic node invasion, sometimes with death resulting. Other strains of *Acanthamoeba* he had collected were less virulent (12).

Another puzzle was solved when Dr. R. S. Chang and his group at the Harvard School of Public Health (4) found that their "lipo-virus" was *Acanthamoeba* in masquerade; and the "Ryan virus" was found by Drs. J. Armstrong and M. Pereira (1) of England also to be a masquerading *Acanthamoeba*.

Now that the mask was off, Dr. E. C. Nelson of the Medical College in Richmond, Virginia, solved the problem of the haunting spector of "atypical meningitis," an epidemic of which had occurred among adolescent swimmers in a warm, outdoor pool in Virginia in 1951-52, killing 15. Dr. Nelson's old tissue-slides from the brains of the victims were taken from the files and reexamined after 15 years and clearly showed the amoebas nestled healthily among the diseased brain tissue (7).

Other reports came in —— a fatal case in a swimmer from Virginia in 1967, and three more in 1969; four cases from Florida, 1962-1967; one from California, 1972 —— in pools warmed to body temperature and above, across the U.S.A.

Naegleria gruberi

Also, there came disturbing accounts from abroad —— meningitis caused by **another** "soil" amoeba, this time an amoeboflagellate (an amoeba which can grow flagella and swim), known as *Naegleria gruberi* (named after two German microbiologists, Nagler and Gruber, by their student, Schardinger). Seventeen cases were reported from Czechoslovakia, seven from Australia, four from New Zealand and five from Great Britain (3, 2) and one from Virginia in the U.S.A.

Again, the conditions of infection were similar, although the amoebas were different —— young, energetic swimmers got the disease after swimming in over-warmed open-air pools!

New Species

Can **any** amoeba produce such a virulent strain? Probably not, says Dr. J. L. Griffin (6; and unpublished data) of the Armed Forces Institute of Pathology in Washington, D.C. He has collected many strains of amoebas and grown them in conditions similar to those in which the virulent strains grow —— warm, *chlorinated* water with bacteria for food —— and he finds that few amoebas grow in such situations. Among those that do, only a very few strains (to date only of *Acanthamoeba* and *Naegleria*) are infective. Some claims are made for a third genus, *Hartmannella*, especially by Dr. B. N. Singh of the Lucknow Drug Institute in India, who thinks *Acanthamoeba* and *Hartmannella* are the same (most others disagree).

Can a non-virulent strain shift to virulent form? Probably not without distinct genetic mutation. Many physiological, biochemical and immunological tests, especially by Drs. Visvesvera and Balamuth (11) of the University of California at Berkeley, show that the Lilly-Culbertson strain of *Acanthamoeba* is distinctly different from *Acanthamoeba castellanii*. Similar tests show that the infective strain of *Naegleria* is significantly different from *Naegleria gruberi*.

Consequently, the lethal *Acanthamoeba* has been named *Acanthamoeba culbertsoni* by Dr. Singh (9) in honor of Dr. Culbertson and the lethal *Naegleria* has been termed *Naegleria fowleri* in honor of Dr. M. Fowler of Australia, who first isolated it in culture. Another strain pathogenic to mice, resembling *N. fowleri*, has been called *Naegleria aerobia* by Singh (9, 10).

Safety Precautions

Does this mean we'll have to give up the "old swimming hole"?

Dr. Griffin doesn't think so. A relatively-cool, country pool, pond or lake, he says, is ideally safe. It's only when a *warm-water-and-chlorine-resistant* strain is present that "blooms" of lethal amoebas occur. Those strains don't compete well with other bacteria-eating amoebas at lower temperatures, and "bloom" only when other amoebas are killed by higher temperatures and chlorine.

How can you avoid amoeba-caused meningitis? It's easiest to do so if you don't go swimming. All (but perhaps one) of the known cases of the disease have occurred in swimmers, always among those who swam in warm, chlorinated pools on a hot summer's day! If you *must* swim then, Dr. Griffin suggests you don't dive; don't get water into your nose and blow it out often; or better yet, wear a nose-clip to keep water out of your nose when you swim and especially if you dive. It's only when the mucus which covers the nasal passage is broken or absent that these amoebas can invade.

Is there a cure for this amoeba-caused meningitis? Not yet! A few drugs have been found capable of killing the amoeba, but those same drugs are also dangerous to humans. Amphotericin-B, an antifungal drug, shows some promise, but has severe side-effects to humans (9).

That's enough to "blow your mind," too, if you're the worrisome sort!

Culturing

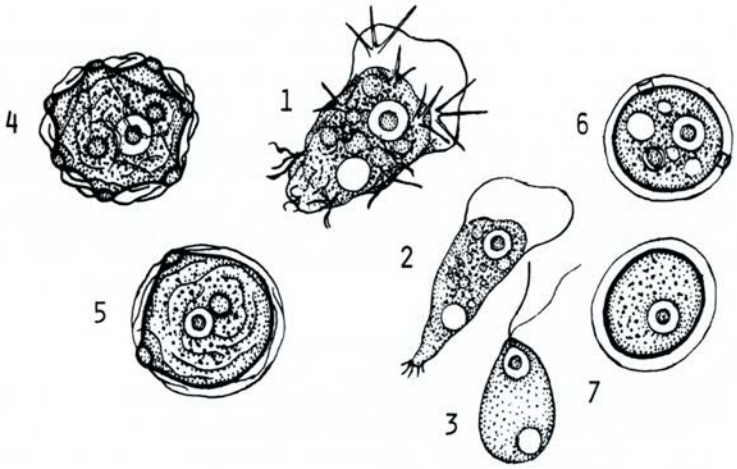
Is there danger in culturing these amoebas in the laboratory? Practically none. No one who has raised them in the laboratory has ever caught the disease.

They can be easily grown on a moist nutrient agar (2.0%) with bacteria or yeasts streaked on the plate as food. When the food is gone, the amoebas enclose themselves in resistant capsules, called *cysts*. These cysts stick to the agar, and can be transferred from one agar plate to another with a wire loop of the type used for making transfers of bacterial cultures. The amoebas can also be grown clonally (axenically, as a "pure" culture) in a peptone, yeast-extract solution in flasks, on a bacteriological shaker. Their cysts are so tough they can be cleaned of bacteria with metal salts or antibiotics before being transferred to the peptone. Even the pathogenic strains offer no danger if handled by standard laboratory methods used in growing bacteria.

Conclusion

Apparently there is little danger of infection from *Acanthamoeba* or *Naegleria-except to swimmers!* Their cysts blow about with other dust-sized particles in the air, along with bacteria, viruses, pollen grains and other

Figures and Legends



1. Active *Acanthamoeba*; all species look much alike in the active state.

2. Active *Naegleria*; all species look much alike in the active state.

3. Flagellated stage of *Naegleria*; all species are similar.

4. Encysted *Acanthamoeba castellanii*; endocyst is polygonal with many plugged pores; ectocyst is much wrinkled; non-pathogenic.

5. Encysted *Acanthamoeba culbertsoni*; endocyst is rounded or slightly polygonal, with a few plugged pores; ectocyst round, slightly wrinkled; pathogenic.

6. Encysted *Naegleria gruberi*; endocyst is round, with several pores; ectocyst is thin, smooth; non-pathogenic.

7. Encysted *Naegleria fowleri*; endocyst is more-or-less ovoid, thick; ectocyst is gelatinous; pathogenic. Encysted *Naegleria aerobia* is nearly the same; also pathogenic (at least to mice).

All figures modified after Singh and Das (10) and Page (8).

dust-components. No one gets the disease, thus, because the mucus sheet of the nasal, bronchial and digestive passages prevents their entry into the tissues.

Swimmers get the disease because they blow the mucus out of their noses along with water, especially after diving, leaving the nasal, mucosal cells bare and open to invasion. Either active amoebas, or those which actively crawl from cysts which have been inhaled, can start an infection — most capably if bacteria are also present to assist pentration (as they always are).

How many other amoebas there may be which can cause such a meningitis is not known. Probably few. Most are not able to tolerate water temperatures as high as human body-temperature. Only a few, tough strains are implicated and then only under special conditions produced in swimming pools and swimmers. The danger is slight, but it is there!

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