

The fat formation occurred exclusively in viable cells. Those close to the site of oleic acid injection showed pyknotic degeneration but no sudanophilia. Although lipogenesis occurred equally in all the native cells of the cornea, none was found in the invading polymorphonuclear cells and only moderate amounts in the macrophages. Comparative studies on nonocular tissue are now under investigation.

DAVID G. COGAN  
TOICHIRO KUWABARA

*Howe Laboratory of Ophthalmology,  
Harvard Medical School,  
Massachusetts Eye and Ear Infirmary, Boston*

#### References

1. H. S. Simms and N. P. Stillman, *Arch. Pathol.* **23**, 316 (1937).
2. H. S. Simms and M. Sanders, *ibid.* **33**, 619 (1942).
3. H. S. Simms, M. S. Parshley, and R. B. Pitt, *J. Gerontol.* **2**, 205 (1947).
4. H. S. Simms, *ibid.* **6**, 159 (1951).
5. Work supported by funds from the American Heart Association and the Massachusetts Heart Association.

27 July 1954.

## Toxicity of Sarin in Bullfrogs

The Chemical Corps has a continuing and necessary interest in studies concerned with the mechanism of action of "nerve gases," compounds that have a powerful anticholinesterase activity. A major part of the fund of knowledge concerning the fundamental physiology of nervous activity has been obtained through studies on poikilotherms, especially the frog. Consequently, data concerning the effect of anticholinesterase drugs on cold-blooded animals are directly related to Chemical Corps research.

Sarin (isopropyl methyl phosphonofluoridate) is a powerful anticholinesterase drug (1). Details of its pharmacology and toxicology in the mouse, rat, guinea pig, rabbit, cat, dog, sheep, goat, and monkey have been published by British investigators (2). This communication summarizes the toxicity of this compound for the bullfrog, *Rana catesbeiana*.

Frogs, weighing 400 to 500 g each, were injected with the drug, dissolved in amphibian saline, into the dorsal lymph sac. Controls received similar injections of plain amphibian saline. The frogs were then put into glass aquariums kept at 22°C and observed until death or for 3 days after injection. Table 1 gives the

Table 1. Illustrating the toxicity of sarin to bullfrogs.

No. of frogs	Dose (mg/frog)	Results
6	0.060	No effect
6	.080	No effect
6	.100	No effect
5	.500	All alive and active 3 days later
17	1.000	1 dead
11	2.000	1 dead
6	4.000	4 dead
6	8.000	4 dead

results. If the logarithm of the dose is plotted as a function of percentage dead for that dose, the LD<sub>50</sub> is seen to be about 6 mg/kg body weight.

It has recently been reported that sarin is the most toxic of three known compounds of the nerve-gas type (3). The toxic dose for man is estimated to be 0.7 to 7.00 mg (3). For a 70-kg man, this value would amount to 10 to 100 µg/kg to kill. Rabbits given 40 µg/kg of sarin intravenously stop breathing in about 10 sec (2). The blood pressure of anesthetized cats, given 200 µg/kg of sarin intravenously, is rapidly depressed to about 30 mm Hg. At such pressure the heart continues to beat effectively for several minutes after respiration ceases (2).

It is evident that the bullfrog is resistant to relatively large amounts of sarin given by injection. The only signs of poisoning noticed were observed in frogs given doses exceeding 1 mg. Such animals were partially anesthetized. They showed no spontaneous movements and responded sluggishly to tactile stimuli. The righting reflexes were present. After 24 hr these signs disappeared, and the frogs seemed to be normal. No blood or tissue cholinesterase values were estimated.

The explanation of the great resistance to, and surprising recovery from, sarin poisoning in bullfrogs is not certain. In mammals two key effects of such poisoning are paralysis of external respiration and depression of circulation. In frogs such effects merely anesthetize the animals until detoxification takes place. In addition, the functional integrity of frog nerves persists even at remarkably low cholinesterase levels (4).

CHARLES G. WILBER

*Chemical Corps Medical Laboratories,  
Army Chemical Center, Maryland*

#### References

1. B. Holmstedt, *Acta Physiol. Scand.* **25**, suppl., 90 (1951).
2. C. A. DeCandolle *et al.*, *Brit. J. Pharmacol.* **8**, 466 (1953).
3. Anon., *Chem. Eng. News*, **32**, 2006 (1954.)
4. F. Crescitelli, G. B. Koelle, and A. Gilman, *J. Neurophysiol.* **9**, 241 (1946).

24 June 1954.

## Morphologic Variation and Mode of Growth of Devonian Trepomatous Bryozoa

Ramose fossil Bryozoa rarely are so situated in enclosing sediments that colonies can be assembled and studied in detail. The Hamilton group of Middle Devonian age in New York State contains colonies of at least three genera. Several have been restored and thin-sections made throughout their length.

The cortical, thick-walled portion of a colony (Fig. 1) contains diaphragms and other structures used in the classification of the order. The stage of development of the cortical region at any one level within the colony can be expected to control structural variations, at least of a quantitative nature. In the axial region of the colony the zoecia are thin-walled and diaphragms and other structures are rare.

Data commonly considered to be of value in dis-

crimination of species include the number of diaphragms, breadth of the cortical region, thickness of the zooecial walls, and the angle at which the zooecia reach the surface. These measurements are found to vary so widely within one colony that as many as three "species" can be designated from one end to the other.

The axial ratio (ratio of the diameter of the axial region to the diameter of the branch) was considered constant for any one species by G. W. Lee [*Mem. Geol. Survey G. Brit.* 1 (3), 135-195 (1912)] and subsequent workers. According to Lee, the proximal ends of the cortex were absorbed as the distal ends were extended. The rate of absorption was assumed to be balanced with the rate of growth, so that the axial ratio remained constant. The axial ratios in different parts of single colonies of the Hamilton group, however, show more variation than Lee allowed within species. On this basis also, as many as three "species" can be differentiated within a colony.

The present investigations resulted in an alternate hypothesis of the mode of growth of ramose colonies that is more consistent with axial ratio measurements and other observations. This new interpretation assumes resorption of the growing tips at the distal ends of the thick-walled regions. The cycle presumably began with growth of the zooecia in the thin-walled region so as to extend the growing tips of the colony. A thick-walled zone was then formed around the tips. Next, resorption from the outer ends of the zooecia back toward the base of the thick-walled region removed distal portions of walls and associated structures such as diaphragms. The amount of resorption varied greatly among species, but usually some traces of the thick-walled region remained. Another cycle began with the extension of the thin-walled region. This growth, in the distal ends of the colony, lengthened the branches.

Proximal to the growing tips of the colony, where

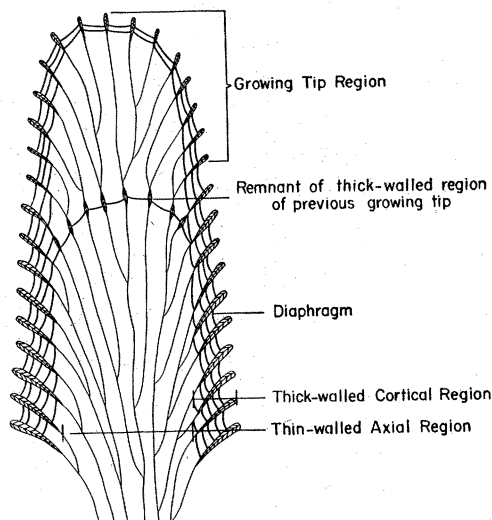


Fig. 1. Idealized diagram of a ramose bryozoan in axial view.

the zooecia established their thick-walled regions in permanent positions opening along the sides of the branches, there are no signs of resorption. Zooecial growth was restricted to the cortex. Thickness of walls, number of diaphragms, and other intramural structures increase progressively toward the base of the colony. The thin-walled, axial region was not affected. Thus, changes in the axial ratio result from a slow proximal increase in the breadth of the cortex, with resulting increase in the diameter of the colony and no accompanying changes in the axial region.

A study of complete colonies indicates that a wider range of specific variation must be allowed in the structures that are controlled largely by the stage of development of the colonies.

RICHARD S. BOARDMAN

*U.S. Geological Survey  
Washington 25, D.C.*

19 July 1954.

## Chromosomes of *Hypomyces solani* f. *cucurbitae*

The cytology of *Hypomyces solani* f. *cucurbitae* was first investigated by Hirsch (1, 2). On the basis of her investigations she concluded that the haploid chromosome number is four in the hermaphroditic strain, three in the male, three in the female, and two in the neuter. When the hermaphrodite mutates to male, it loses one of its sex chromosomes; when it mutates to female it loses the other. Hirsch claimed that hermaphrodites and neuters, which are produced among the offspring of the cross  $\text{♀} \times \text{♂}$  (3), arise as a result of occasional nondisjunction of sex chromosomes.

However, my genetic investigations (4) were not in accord with Hirsch's interpretations, and hence the cytology of *Hypomyces solani* f. *cucurbitae* required reinvestigation. It is the purpose of this paper to describe briefly the results of such reinvestigation (5).

By employing the aceto-orcein smear technique, which was used successfully by McClintock (6) on *Neurospora crassa*, it was possible to study the nuclear behavior in the ascus and to determine the number of chromosomes in each of the four strains. Priority was given to the cross  $\text{♀} \times \text{♂}$ , since the four sex strains can be obtained from such a mating, and hence the chromosome number in each strain would be determined. Careful analysis of diakinesis, prometaphase I, and metaphase I figures has revealed that the number of bivalents at these stages was consistently four. At early anaphase I and mid-anaphase I, eight dyads could be counted. During the following divisions each nucleus was observed to contain four chromosomes. Nondisjunction of what are called the two sex chromosomes, as reported by Hirsch to occur frequently during the first anaphase, has never been seen. Since, from the cross  $\text{♀} \times \text{♂}$ , the four sex strains are produced, and since all nuclei in the ascus at the same stage of division have the same number of chromosomes, it seems justifiable to conclude that the haploid chromosome number in hermaphrodite, female, male,