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Cholesterol in Blood of Horseshoe Crab and Woolly Bear Caterpillar.

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It is generally considered that the blood of all animals contains both free cholesterol and cholesterol esters, the former existing in corpuscles and plasma and the latter occurring only in the plasma or serum. Since this conception is founded upon the results of determinations carried out on the blood of higher animals it seemed of interest to determine whether lower and more primitive forms would also contain these constituents in their blood corpuscles and serum.

The horsehoe crab *(Limulus polyphemus)* and the woolly bear caterpillar *(Isia isabella)* were chosen as suitable experimental animals both as to position in the animal scale and as to the ease with which sufficient blood could be obtained. The work on the horseshoe crab was carried out on 2 adult animals, and that on the caterpillars on pooled blood samples from 40 animals. Bloor's[1] method was used in determining total cholesterol and Bloor and Knudson's[2] in determining cholesterol ester, both modified as to amount of material tested and amount of extractive used in anticipation of very low values. The results obtained are given in Tables I and II.

Summary. The blood serum of *Limulus polyphemus* was found to be entirely free of cholesterol or cholesterol esters and thus differs

TABLE I. *Horsehoe Crab (Limulus polyphemus).*

Animal No.	Total cholesterol in blood cells.	Cholesterol ester in blood cells.	Total cholesterol in blood serum.	Cholesterol ester in blood serum.
	mg. %	mg. %	mg. %	mg. %
1	67.2	0	0	0
2	75.6	0	0	0

TABLE II. *Woolly Bear Caterpillar (Isia isabella).*

Total cholesterol in blood cells.	Cholesterol ester in blood cells.	Total cholesterol in blood serum.	Cholesterol ester in blood serum.
mg. %	mg. %	mg. %	mg. %
240.0	0	27.7	0

[1] Bloor, W. R., *J. Biol. Chem.*, 1916, xxiv, 227.

[2] Bloor, W. R., and Knudson, A., *J. Biol. Chem.*, 1916, xxvii, 107.

from the blood sera of higher animals. The blood cells, however, contained free cholesterol and were similar to blood cells of higher animals in that they contained no cholesterol esters. Both blood cells and serum of the woolly bear caterpillar contained free cholesterol but no cholesterol ester was present.

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Comparison of the Resistance of Streptococci and of Poliomyelitic Virus in Glycerol.

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The virus of poliomyelitis, present in monkey brain tissue has been shown to be virulent after being kept 8 years in 50% glycerol at 4° C.[1] Micrococci have been obtained in culture from the "glycerolated nervous system of cases of poliomyelitis after a period of 15 months."[2]

Streptococci and other microorganisms were isolated from the brains of monkeys recently infected with the virus of poliomyelitis and from those of normal monkeys.[3]

Rabbits were inoculated intracerebrally with certain of the streptococci.[3] The brains of these rabbits were removed under aseptic precautions and were stored at 4° C. in 50% glycerol. At frequent intervals cultures were made by placing a small fragment of washed glycerolated brain material in dextrose broth. Seven brain specimens were thus tested. One brain inoculated with streptococci yielded a pure culture of the organisms after 82 days, but not after 116 days; one after 116 but not after 146 days, and three after 117 but not after 204 days. In 2 of the brains tested streptococci were still viable after 303, but not after 333 days.

It is evident that while streptococci can survive in brain tissue in 50% glycerol for periods up to nearly a year, there is wide disparity between the resistance of the virus of poliomyelitis and the resistance of certain strains of streptococci in glycerol.

[1] Rhoads, C. P., *J. Exp. Med.*, in press.

[2] Mathers, G., and Weaver, G. H., *J. Infect. Dis.*, 1918, xxii, 559.

[3] Long, P. H., Olitsky, P. K., and Stewart, F. W., *J. Exp. Med.*, 1928, xlviii, 431.