

On the phospholipids of adult *Ancylostoma caninum*

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Summary. The phospholipid composition of *Ancylostoma caninum* has been examined. Phospholipids amounted to 0.9% of the wet weight of the parasite. Ethanolamine and choline containing lipids comprised about 76% of the total phospholipids. Lysolecithin, sphingomyelin, phosphatidyl inositol and phosphatidyl serine were present in minor amounts. Cerebrosides and sulfatides were also identified.

Ancylostoma caninum, the dog hookworm, is a parasite belonging to the class of nematodes. Adult stage of *A. caninum* inhabits the intestine of mammals and causes severe anemia³. At present our knowledge of the composition and origin of the phospholipids of parasites in general and those of *Ancylostoma* is very scarce⁴. As part of a comprehensive chemical study of the parasite, the lipids of the adult *A. caninum* have been investigated. This paper pertains to the phospholipid composition of this parasite.

Materials and methods. *A. caninum* were removed from the intestinal tract of the dogs immediately following the sacrifice and were placed in physiological saline. After repeated washing, they were stored at -20 °C. The worms were blotted on filter paper and weighed. Extraction, purification and isolation of phospholipids were as de-

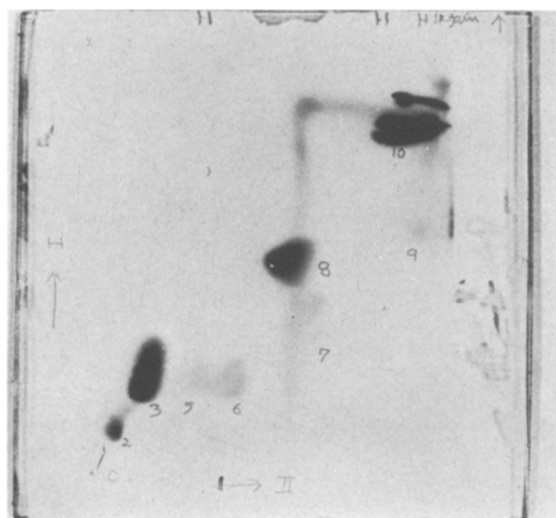
scribed previously^{5,6}. The separation, identification and characterization of phospholipids were as described elsewhere^{7,8}. Cerebrosides and sulfatides were separated on silica gel G plates in chloroform-methanol-conc. ammonia, 80:20:0.4. Plates were developed for several hours to achieve better resolution of these lipids. Sugar containing lipids were identified with α -naphthol sulphuric acid spray. **Results and discussion.** The total chloroform-methanol soluble fraction of *A. caninum* amounted to 2.7% and phospholipids to 0.9% of the wet weight of the parasite. The lipids gave components on thin layer chromatoplates with iodine vapors (figure). These phospholipids were identified as lysolecithin, sphingomyelin, lecithin, phosphatidyl inositol, phosphatidyl serine and phosphatidyl ethanolamine. The presence of these phospholipids was further confirmed by cochromatography with authentic standards. It is evident from the table that choline and ethanolamine containing lipids comprise about 76% of the total phospholipids of the parasite. On chromatography of these lipids in chloroform-methanol-ammonia, 80:20:0.4, 2 sugar positive components were identified with α -naphthol sulphuric acid spray. These components were identified as cerebrosides and sulfatides with authentic markers. The presence of these lipids was also confirmed by cochromatography with the respective glycolipids.

The chemical nature of phospholipids of helminths in general appears to be similar to that of mammalian tissues. The most abundant phospholipids found in parasites are phosphatidyl choline, phosphatidyl ethanolamine and phosphatidyl serine. The phosphoinositides and sphingomyelins are, often but not invariably, encountered as cardiolipin and plasmalogens⁴. The results obtained in the present study on the lipid content and phospholipid composition of *A. caninum* are similar to cestodes⁴. Ethanolamine and choline containing lipids comprise about 70% of the total phospholipids in *Ascaris lumbricoides*⁹, identical with the results of the present study. To the best of our knowledge, this is the 1st report on the presence of cerebrosides and sulfatides in nematodes, whereas these lipids have been already identified in trematodes i.e. *Fasciola hepatica*¹⁰. The functions of these lipids in metabolism of *Ancylostoma* is unknown and deserves investigation.

Phospholipids of *Ancylostoma caninum*

Phospholipid	Total phosphatides (%)*
Lysolecithin	5.0
Sphingomyelin	4.0
Phosphatidyl choline	39.1
Phosphatidyl inositol	7.5
Phosphatidyl serine	6.8
Phosphatidyl ethanolamine	37.5

* Each value is the mean of 3 different determinations.



2 dimensional thin. layer chromatogram of lipids of *Ancylostoma caninum*. Chloroform-methanol-water, 65:25:4 was used in the 1st direction and chloroform-methanol-acetic acid-water, 50:35:8:1 in the 2nd direction. Components were identified with iodine vapors. Identity of components: 1. Lysolecithin, 2. sphingomyelin, 3. phosphatidyl choline, 5. phosphatidyl inositol, 6. phosphatidyl serine, 7. sulfatide, 8. phosphatidyl ethanolamine, 9. cerebroside, 10. neutral lipids.

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