pounds are found in enzymatic hydrolysates of the 6 proteins; however, when citrulline is added to such hydrolysates, its "recovery" in terms of colorimetric units corresponds to several times the theoretical value. The reason for this anomalous result has yet to be determined.

4. It is probable that citrulline is either absent or present only in minimal quantities in these proteins.

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Comparative Study of Lipids in Whole Carcasses of Arctic and Non-Arctic Fish.* (17453)

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Information concerning lipid content and metabolism in cold-blooded animals is relatively meager.[1] Concerning these matters in Arctic poikilotherms, information is non-existent. A survey was, therefore, made of the lipid content of whole fish, *Pygosteus pungitius*,[2] the Arctic 9-spined stickleback. The results were evaluated in comparison with similar analyses made on the common guppy *(Girardinus guppyi)*.

Material and methods. Whole sticklebacks or guppies were carefully weighed and then dropped into a mixture consisting of 3 parts of pure ethanol and 1 part benzine. After 24 hours the fish were ground with sand and the lipids extracted according to Bloor's method.[1] Total fatty acids were estimated using the method of Bloor as modified by Snell.[3] Lipid phosphorus was estimated by a modified Youngburg technic;[4] from the latter results phospholipids were calculated as lecithin. Measurements of cholesterol were based on methods developed by Bloor.[5] Since all the procedures are colorimetric, a Klett-Summerson photometer was used to measure the various color intensities. Ten sticklebacks were analyzed. Two groups of 5 each guppies were tested: one group was fed ordinary commercial fish food; the other, chopped earthworms.

Results. The results are summarized in Table I, which shows that there is a mean value of 4.88% fatty acids, 0.32% cholesterol, and 1.87% phospholipids in the Arctic stickleback.

Table I also shows that the group of guppies fed on commercial fish food have the following mean lipid values: fatty acids, 8.87%; cholesterol, 0.56%; phospholipid, 2.31%. Guppies fed on chopped worms have lipid values practically identical with those fed commercial fish food.

Discussion. The results indicate that there is about the same amount of cholesterol in the guppies on either diet, but that there is less cholesterol in the Arctic fish than in the guppies. Total fatty acids are higher in the guppies, on either diet, than in the Arctic stickleback. The phospholipid content of all

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1 Bloor, W. R., *Biochemistry of the Fatty Acids*, New York, 1943.

2 Preble, E. A., A biological investigation of the Athabaska-Mackenzie Region, North American Fauna, No. 27, Washington, 1908.

3 Snell, F. D., and Snell, C. T., Colorimetric methods of analysis, New York, 1937.

4 Youngburg, G. E., and Youngburg, M. V., *J. Lab. Clin. Med.*, 1930, **16**, 158.

5 Bloor, W. R., *J. Biol. Chem.*, 1916, **24**, 227.

TABLE I.
Results of Lipid Analyses of the Arctic Stickleback and of 2 Groups of Guppies. All values expressed in percent fresh tissue.

Substance	Arctic stickleback	Guppies fed commercial fish food	Guppies fed earthworms
Fatty acid—mean	4.88	8.87	7.30
Range	2.46-7.06	8.14-9.94	6.22-9.33
Cholesterol—mean	0.32	0.56	0.55
Range	0.29-0.35	0.4-0.93	0.44-0.63
Lipid P—mean	0.08	0.09	0.08
Range	0.06-0.08	0.07-0.12	0.06-0.09
Phospholipid—mean	1.87	2.31	2.05
Range	1.57-2.08	1.83-2.60	1.52-2.31

TABLE II.
Table Showing the Various Lipid Ratios for the Arctic Stickleback and for the 2 Groups of Guppies. The ratios were calculated from the means given in previous table.

Group	Chol/fa	Chol/phosph	Chol/lipid P	Fa/lipid P
Stickleback	.07	.17	4.0	61.0
Guppy—worm fed	.08	.27	6.9	91.0
Fish food	.06	.24	6.2	99.5

3 groups is the same.

Calculations of the various lipid ratios were made for each group (Table II). Examination of these ratios indicates that, in relation to cholesterol, there is more phospholipid in the Arctic fish than in any of the guppies. It is maintained that the higher the ratio, cholesterol/lipid phosphorus, the greater the water content of the tissue in question.[6] If this generalization is universally true, the Arctic stickleback contains less water than does the guppy. It might be that this lower water content is correlated with the ability of the stickleback to withstand freezing.[7]

The fatty acid/lipid phosphorus ratios show that in the Arctic fish there is much less fatty acid, in proportion to phospholipid, than in the guppies. This may indicate that the turnover of fats is more rapid in the stickleback than in the guppies.

There is an interesting parallelism in the lipid content of the whole carcasses of guppies fed on commercial food and liver lipids in various fish. For example, Weill[8] found the liver cholesterol in various fish to be as follows (percent wet weight): *Truta fluvialitis*, 0.68; *Gadus merlangus*, 0.54; *Solea vulgaris*, 0.47. These values are quite close to that of 0.56% cholesterol in the guppies fed commercial food (Table I).

Mottram[9] reported that in the liver of the plaice fatty acids amount to 8.86% of the fresh tissue. This value compares closely with that of 8.87% fatty acids in whole guppies (Table I).

Similarly, Javillier and co-workers[10] found that in the liver of the carp lipid phosphorus amounts to 0.099% fresh tissue, a value similar to 0.09% for lipid phosphorus in the whole guppies. The Arctic stickleback also contains a similar amount of lipid phosphorus 0.08% (Table I). although its other lipids do not resemble so closely those of the guppy.

The present results indicate that in the stickleback phospholipids are high in proportion to other lipids. Such a condition suggests that in the single Arctic fish studied lipid metabolism is proceeding at a higher level than in the guppies, for phospholipid level is

[6] Mayer, A., and Schaeffer, G., *J. physiol. path. gen.*, 1913, **15**, 771.

[7] Pouchet, F. A., *J. Anat. Physiol.*, 1866, **3**, 1.

[8] Weill, J., *C. R. Acad. Sci.*, 1914, **158**, 642.

[9] Mottram, V. H., *J. Physiol.*, 1912, **45**, 363.

[10] Javillier, M., Cremieu, A., and Hinglais, A., *Bull. Soc. chim. biol.*, 1928, **10**, 327.

a good indication of overall level of fat metabolism.[1] One might expect just such a condition as an adaptation on the part of the stickleback to Arctic living: the high caloric values of the fats make them ideal sources of energy in an environment such as the Arctic.

Other adaptations of a metabolic nature are known to exist in Arctic fish: polar cod tissue shows Q_{02}-temperature curves peculiarly adapted to functioning at low temperatures.[11]

Summary. 1. Whole Arctic sticklebacks and ordinary guppies were analyzed for fatty acids, cholesterol and lipid phosphorus.

2. It was found that diet does not have an appreciable effect on the lipids in the guppies.

3. Cholesterol and fatty acid are higher in the guppies than in the Arctic fish. On the other hand phospholipid is essentially the same in all the fish examined.

4. There is more phospholipid in relation to cholesterol in the Arctic sticklebacks than in the guppies, a fact which may be interpreted as indicating a higher level of fat turnover in the former than in the latter.

5. The ratio of cholesterol/lipid phosphorus is lower in the stickleback than in the guppy, indicating a lower water content in the tissues of the former. This may be correlated with the reported ability of the stickleback to resist freezing injuries.

[11] Field, J., and Peiss, C. N., *Fed. Proc.*, 1949, **8**, 44.

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Electroencephalographic Changes Associated with Painful and Non-Painful Peripheral Stimulation. (17454)

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This work was undertaken to ascertain whether the perception of pain is associated with a specific change in the electroencephalogram (E.E.G.), which might be used as an objective criterion of pain.

Methods. A Grass Electoencephalograph was used with 6 or 8 electrodes in 17 experiments on as many subjects. In addition to the usual 8 electrodes (2 frontals, 2 parietals, 2 temporals and 2 occipitals) the Grinker needle electrode[1] in the sphenoid bone was employed in 2 experiments, and a saline saturated cotton electrode pressed against the posterior pharyngeal wall in one. For the purpose of exploring any changes from the region of the base of the skull or hypothalamus near the foramen ovale in still another experiment an insulated needle electrode was inserted (under infiltration anesthesia) between the mandible and zygoma to a depth of 5.5 cm. Monopolar registration was employed with the lobe of the ear as indifferent electrode. Pain was produced in several ways in each experiment. (a) A Von Frey hair stimulator was applied to a previously determined pain point on the right forearm with a pressure of 8 g. (b) The web of the skin between the thumb and index finger of the right hand was pressed between a forceps. (c) As a control in two experiments heat, cold and touch were applied to the right forearm without causing pain.

Results. In all experiments a definite change in the E.E.G. occurred. It consisted of a decrease in amplitude, which was in the nature of a loss of electrical entropy and must be separated from the attention wave, which frequently appeared as an additional factor. The change was not specific for a pain stimulus or sensation since a similar response was obtained with heat, cold and touch. The decrease was bilateral and was not confined to any large cortical area, though it was more pronounced in the parietal, occipital and temporal region in a decreasing order of mag-

[1] Grinker, R. R., and Serota, H. M., *J. Neurophysiol.*, 1938, **1**, 573.