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Phosphate in Lobster Nerve.*

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The nerves of the large claws and walking legs of the lobster (*Homarus americanus*) with or without the abdominal ganglion chain were carefully dissected into iced mortars. They were ground and analyzed, either at once or after standing for $\frac{1}{2}$ to 3 hours in closed vessels in moist oxygen or nitrogen at 23-25° C. In a few cases companion analyses were run on nerves resting or stimulated in oxygen, stimulation being continued with fairly strong induction shocks at 100 per second for half an hour. The method of analysis was essentially that described by Fiske and Subbarow¹, involving extraction with 10% iced trichloroacetic acid, part of the filtrate being used to determine total acid-soluble phosphorus after ashing with sulphuric and nitric acids, the larger portion being neutralized and treated with calcium chloride-calcium hydroxide precipitant. After centrifuging and washing, the inorganic phosphate remained in the precipitate. Phosphagen was determined in the clear solution after reacidification with hydrochloric acid and standing at room temperature for 2 days, according to Meyerhof². Phosphate was finally estimated in each fraction by the Fiske-Subbarow method, using a total volume of 15 cc. and comparing with a phosphate standard of 8.0 mg. % strength.

The rate of hydrolysis of the phosphagen compound, as tested with acid for different time intervals, clearly showed it to be of the arginine phosphate rather than the creatin phosphate type, which is in conformity with the difference in muscle phosphagens in vertebrates and crustaceans. The usual controls of the adequacy of separation and determination demonstrated the validity of our procedure. Especially, the sum of the inorganic and phosphagen fractions separately determined was equal to the acid-labile phosphorus (Gerard and Wallen³) determined directly.

The total soluble phosphate of the nerves, at once or after a period in oxygen, averaged 41 mg. % in 7 experiments, the variation being

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¹ Fiske, C. H., and Subbarow, Y., *J. Biol. Chem.*, 1929, **81**, 629.

² Meyerhof, O., *Die chemischen Vorgänge im Muskel*. Springer, Berlin, 1930.

³ Gerard, R. W., and Wallen, J., *Am. J. Physiol.*, 1929, **89**, 108.

31 to 46. After an average stay in nitrogen of 2 hours, the value for 20 experiments was 43.4, extremes 30 to 64. The phosphagen content of those analyzed at once averaged 5.1 mg. % and showed little change on standing in oxygen. After 2 hours' anoxia, the value had fallen to 1.6. A definite decrease in phosphagen in the asphyxiated compared with the control nerve was apparent in each experiment. The phosphate present in the soluble fraction exclusive of the inorganic and phosphagen phosphate was 12 mg. %. This fraction was decreased to 8 mg. % during asphyxia. The inorganic phosphate increased in asphyxia by an amount corresponding to the decrease in the other two. The few experiments on stimulation did not lead to a definite change in the phosphate fractions, but neither the number of experiments nor the conditions of stimulation justify a conclusion on this point. Chopping and warming the nerves led to a complete breakdown of phosphagen with but little decrease in the other combined phosphate fraction. These results closely parallel previous findings on the frog sciatic³.

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Studies on Calcium. VI. Effective Dose of Calcium Gluconate Subcutaneously Administered in Completely Parathyroidectomized Dogs.*

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Lieberman and Szurek¹ have previously shown that parathyroid tetany can be prevented (dogs) by injecting subcutaneously large quantities of calcium gluconate (in 10% concentration). Whereas this salt of calcium is the only one so far described which can be injected subcutaneously without causing necrosis, no attempt was made to determine how *little* of a 10% solution sufficed to prevent the appearance of tetany following a single daily subcutaneous injection of this salt on the basis of cc/kg body weight in the thyroparathyroidectomized animal.

Method. Complete data were obtained from 5 dogs. The average weight of the dogs was 10 kg. (Extremes: 7.5 kg.-13.5 kg.). For the the first 2 weeks after operation the animals were put on milk and meat diet. Thereafter, on dog biscuit diet.

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¹ Lieberman and Szurek, *J. Pharm. and Exp. Therap.*, 1931, **41**, 333.