INTRODUCTION

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The vertebrate skeletal muscle is a complex machinery. in which a host of chemical changes take place with the result that work is done or energy set free as heat. It is being realized more and more at present that the fuel for such energy in the muscle is not always carbohydrates, but where sustained activity is involved, energy is derived mostly from fat. How a muscle can oxidize fatty acids and obtain the necessary energy for the work it performs has been studied by several workers. But one important aspect of the metabolism of fat in the muscle that has never been sufficiently appreciated is, that all the fatty acid that is burnt in the muscle is not just poured into the muscle fibres when the metabolic wheels are set moving, but fatty acids are stored in the muscle fibres themselves in the form of esters, the most important of which being the glycerol ester. The muscle is thus always equipped with an adequate reserve of the fuel for energy. If so, the fat has to be broken down into the component fatty acids and glycerol before complete oxidation. The hydrolysis of fat is brought about by lipase, a fat splitting, enzyme. It follows then that the muscle should contain this enzyme and in large concentrations where more fat is utilized. This aspect of fat metabolism in muscle which was not studied previously has been done and constitutes the major part of the present thesis.

It has been shown by earlier workers that the flight muscles of birds and other flying animals utilize more fat

because of the high requirement of energy needed for flight. The powerful breast muscle of birds was therefore chosen for the present study.

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It was found that vertebrate skeletal muscle in general and the breast muscle of flying birds in particular contain a fat splitting enzyme. Comparing this enzyme with the pancreatic lipase it was shown that the fat splitting enzyme is a true lipase. The breast muscle of the bat was also shown to contain a similar fat splitting enzyme, and the view that the fat stored in the muscle could be directly utilized for energy purposes is thus supported.

Having thus demonstrated the occurrence of a lipase in muscle, some of the biochemical properties of the lipase were studied manometrically using the Warburg apparatus. This study was further extended to the cardiac muscle of vertebrates and a claim by an earlier author that the myocardial lipase is a lipoprotein lipase has been shown to be illfounded. A study of the lipase activity in the skeletal and cardiac muscles of a number of vertebrates showed that the concentration of the lipase in the heart muscle is related to the basal metabolism of the animal and the lipase concentration of the skeletal muscle is related to the extent of fat utilization in the muscle, which is dependent on its activity.

The lipase activity in the mitochondria of the pigeon breast muscle was also assessed and a study of the <u>in vitro</u> oxidation of fatty acids in the muscle with triglycerides as the starting material was made using the Warburg apparatus.

A histophysiological study of the lipase activity in the pigeon breast muscle showed that the lipase is confined to the narrow fibres (It was shown by other workers that the pigeon breast muscle contains two types of fibres, a broad white variety and a narrow red variety). This observation led to a similar study of other enzymes like alkaline phosphatase the dehydrogenases operating in the Krebs cycle and glycolysis and it was observed that all the enzymes are confined to the narrow fibres suggesting thereby that mostly the narrow fibres are concerned with energy metabolism. The co-existance of two such fundamentally distinct types of fibres in the same muscle thus present a conundrum to which a possible explanation is given. The histophysiological study of the enzymes was also extended to the cardiac muscle of the pigeon, the breast muscle and certain other muscles of the bat and a few other birds and the results obtained discussed.

In the course of the study a simple method was devised for the preparation of sections of tissues for histophysiological studies, a method which has a wide variety of other applications in the study of physiology and biochemistry. The method for the histophysiological study of dehydrogenases was modified and the drawbacks in some of the earlier methods pointed out. The triphenyltetrazolium chloride reduction method for the study of succinic dehydrogenase was extended to a number of other dehydrogenases.

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- On the occurrence of lipase in the skeletal muscles of vertebrates and its possible significance in sustained muscular activity. <u>J.Anim.Morph.Physiol.</u>, 3, 91 (1956).
- 2. Lipase activity in the vertebrate heart muscle and its relation to basal metabolism. <u>Ibid</u>.,4, 107 (1957).
- Histochemical demonstration of lipase activity in the <u>pectoralis major</u> muscle of the pigeon. <u>Nature</u>, 181, 783 (1958).
- 4. A quantitative and histochemical study of lipase activity in the <u>pectoralis major</u> muscle of the bat. <u>Naturwissenschaften</u>, 45, 93 (1958).
- 5. Histochemistry of muscle lipase. <u>J.Anim.Morph.Physiol.</u>,
 5, 43 (1958).
- Alkaline phosphatase ačtivity in the pigeon breast muscle.
 <u>Curr. Sci.</u>, 27, 172 (1958)
- 7. A histochemical study of dehydrogenase activity in the <u>pectoralis major</u> muscle of the pigeon and certain other vertebrate skeletal muscles. <u>Quart. J. Micr. Sci.</u> (In press).
- Alkaline phosphatase and succinic dehydrogenase activity in the breast muscle of bat, a histochemical study. J.Anim.Morph.Physiol.(In press).
- 9. Activation of pigeon pancreatic lipase by mercuric chloride. <u>Curr. Sci.</u>(In press).

The following abbreviations are used in the text. ATP, Adenosine triphosphate; ADP, Adenosine diphosphate; CP, Creatine phosphate; Col, Coenzyme 1; CoA, Coenzyme A; DPN, Diphosphopyridine nucleotide; Ph, Phosphate; Pi, inorganic phosphate