# INTRODUCTION

In the animal kingdom, the ability to move from place to place varies with the habitat. In higher vertebrates which live on land or air, the organs of locomotion are increasingly efficient and specialized, as exemplified by the limbs in mammals and wings in birds. In fishes which are lower vertebrates, the motive force required for progression through the water is supplied by the trunk muscles and not the fins. Most fishes have retained the primitive arrangement of the body muscles, the myotomes which form a series of blocks or segments arranged one behind the other and separated by thin sheets of connective tissue or myocommata. The stream-lined shape of the body is well adapted to attain an efficient movement with great economy of energy for progression in a liquid medium. This is well illustrated in migratory fishes such as the salmon which travel hundreds of miles to reach their spawning grounds without feeding on the way. Drummond and Black (1960) have reviewed the studies on the changes in carbohydrate, protein and fat contents of the body in fishes during migration. They have shown that the glycogen content of fish muscle is very little and this could get depleted easily, whereas the fat store could serve as a potentially large source of energy. Black (1958) in his discussion on energy stores and metabolism gave a penetrating account of the problem in relation to biochemical exchange. He has postulated that energy must be provided through the metabolism of metabolite reserves, chiefly fat and protein,

to provide for the development and maturation of the gonads, as well as for other essential metabolic needs, including that of muscular energy. Bilinski (1963) found that the oxidation of some fatty acids takes place at a very much higher rate in red than in white muscles of the trout, and hence the greater utilization of fat for energy by the red muscle during prolonged muscular activity.

Muscles of different animals are known to differ in their fibre composition, colour and biochemical properties, and even within the same animal, characteristic differences are seen between the muscles from two different regions. Histophysiological studies on the avian pectoralis muscle have been one of the main areas of research in our laboratories. These studies have shown the existence of two types of muscle fibres in the pectoralis muscle of pigeon and a number of other birds. The narrow red tonic type of muscle fibres with large numbers of mitochondria, high concentrations of fat and oxidative enzymes are well equipped for an aerobic metabolism using fat as the chief fuel whereas the broad white tetanic type of fibres with very few mitochondria, low levels of fat and oxidative enzymes but with high concentrations of glycogen and glycolytic enzymes are adapted for an anaerobic metabolism using mainly glycogen (George and Naik, 1959).

On the basis of some exploratory observations reported by George (1962) in the red and white muscles of the mackerel,

which is a migratory fish, it was thought profitable to study the fish muscle along the same lines as were followed in the case of the pigeon breast muscle, with a view to obtain a better understanding of the structure and function of the red and white muscles in fishes. The trunk muscle situated along the lateral line is generally seen to comprise of dorsal red and ventral white muscle blocks, the proportions of which vary from species to species. The red colour of the muscle is due to the presence of the protein pigment myoglobin, and this muscle is also richer than the white muscle in mitochondria. These two types of muscles were studied in a few representative types of fishes. Variations in the cellular organization of the red and white muscle in regard to the nature of the fibres, the mitochondrial distribution and the concentration of fat in a number of marine and fresh water fishes were also studied.

A detailed histochemical study of the distribution pattern of myoglobin, the metabolites (glycogen and fat) and some of the glycolytic and fat metabolizing enzymes has been made in the fresh water carp, Cirrhina mrigala, with a view to obtain a better picture of the metabolic activities of the red and white muscles. The high levels of the metabolites as well as of the enzymes, oxidative and glycolytic, in the red fibres, suggest that these fibres are better adapted for an oxidative as well as glycolytic metabolism than the white fibres.

Quantitative estimations of fat, glycogen, lipase and succinic dehydrogenase have been made in the red and white muscles along the anterior, middle and posterior regions of the body of fish with a view to determine whether any significant regional differences as regards the metabolite load and enzyme concentrations were evident. Studies on the capacity for *in vitro* fatty acid (butyrate) oxidation by whole homogenates of the red and white muscles separately was assessed in three fishes of the family Cyprinidae viz. *Labeo rohita*, *Labeo fimbriatus* and *Cirrhina mrigala*, to obtain data for a better understanding of the mechanisms involved in fat utilization by both the muscles.

Publications of earlier investigators though dealt with various facets of the fish muscle, no comprehensive assessment at the subcellular levels has heretofore been presented. It was therefore thought desirable to assess the levels of lipase and succinic dehydrogenase in the cell particulate fractions of the red muscle of *Labeo fimbriatus* and *Cirrhina mrigala*. The submicroscopic analysis reported here has revealed the possibility of a high level of extramitochondrial oxidative metabolism.

A new method for the histochemical demonstration of lipase activity in muscle was attempted in order to obtain precise intracellular localization of the enzyme activity. Tween 85 was used as the substrate and alizarin red S as the agent to stain the calcium soap formed. The enzyme activity

was found to be localized in the mitochondria.

The localization of sarcotubular ATPase acting at a pH optimum of 7.2 in the presence of a sulphydryl compound has been reported by others. The occurrence in skeletal muscle of an acid ATPase which is adapted to function in the acid pH range was thought possible. Such an ATPase with pH optimum of 2.5 was demonstrated in muscle. In frog muscle, this enzyme had to be demonstrated after electrically stimulating the leg muscles for varying lengths of time, since acid ATPase activity was found to be very low in resting frog muscle. Mitochondrial ATPase was also studied.

This thesis has been prepared with a view to publish it in parts, in the form of separate papers. Each chapter therefore has been made as comprehensive as possible. This has inevitably resulted in some repetitions which could not be avoided.

The following papers have already been published.

1. Cellular organization and fat utilization in fish muscle. J. Anim. Morphol. Physiol. (C. J. George Felicitation Number) 11 : 124-132, 1964.
2. Histochemical demonstration of muscle lipase. J. Histochem. Cytochem. 12 : 768-771, 1964.