Proc. Indian natn. Sci. Acad. **B47** No. 6 pp. 823–827 (1981)

Studies on Physiology of Mammalian Cardiac Muscle: Differential Distribution of Certain Metabolites in the Myocardia of Ox, *Bos* sp.

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(Received on 27 July 1981; after revision 24 August 1981)

Biochemical assay of certain metabolites in different regions of the ventricular myocardia Ox, *Bos* sp., have been carried out. Definite biochemical gradients exist in the distribution pattern of some metabolites as well as enzymes in the different myocardial regions. The glycogen content of the outer region of both ventricles was higher than the inner region, whereas the lipids, myoglobin and SDH were higher in the inner region.

Key Words: **Myocardia, Gradients, Myoglobin, Glycogen, Lipids and SDH**

Introduction

Considerable attention has been focussed on the physiology of cardiac muscle of vertebrates. Despite the earlier concept of homogeneity in the distribution pattern of biochemical constituents in the cardiac muscle, recent investigations indicate a varied distribution pattern of certain metabolites in relation to their functional requirements (Bing 1953, Weber 1957, Arnold 1968, George 1969, Hamoir et al. 1971, Melnick et al. 1973, Morel & Pinset Harstorm 1975, Webber & Janicki 1977). The specific role of myoglobin in facilitating oxygen supply to the red skeletal muscle fibres of the breast muscle of pigeon has been well established (Wittenberg 1975). Studies on various chambers of the myocardia of some mammals have revealed a differential distribution pattern of myoglobin (Alexander 1975). Further specific regional variations in the distribution of glycogen and glycolytic enzymes have been observed in the ventricles of some mammals (Lilian 1963, 1964). It has been generally accepted that in prolonged muscular activity, lipid is the chief fuel (Fritz 1961, George & Berger 1966, Drummond 1967). Observations of Josephine and Harris (1971) on lipid levels of different chambers of the dog's heart have shown a differential pattern of distribution. The present study has been undertaken to evaluate the distribution pattern of certain metabolites in the various muscle layers of Ox, *Bos*. sp.

Materials and Methods

Myocardia of a large domesticated mammal

Ox, *Bos.* sp. were used for the present studies. Hearts were collected from selected, healthy, freshly slaughtered adult animals from the Corporation Slaughter House, Trivandrum and swiftly brought over to the laboratory in a refrigerated container. Cardiac muscle samples were taken from the outer, middle and inner regions of the mid left ventricle and from the inner and outer regions of the mid right ventricle. Tissue samples were also taken from the interventricular septum (figure 1). Myoglobin was assayed by the method of Tappan and Reynafarjee (1957). The succinic dehydrogenase activity of myocardial samples was quantitatively assayed by the method of Kun and Abood (1949) using triphenyl tetrazolium chloride (TTC) as an electron acceptor. Glycogen was estimated according to the procedure of Seifter et al. (1950) and the total lipids were assayed by the method of Bragdon (1951). The data were analysed statistically using student's t-test.

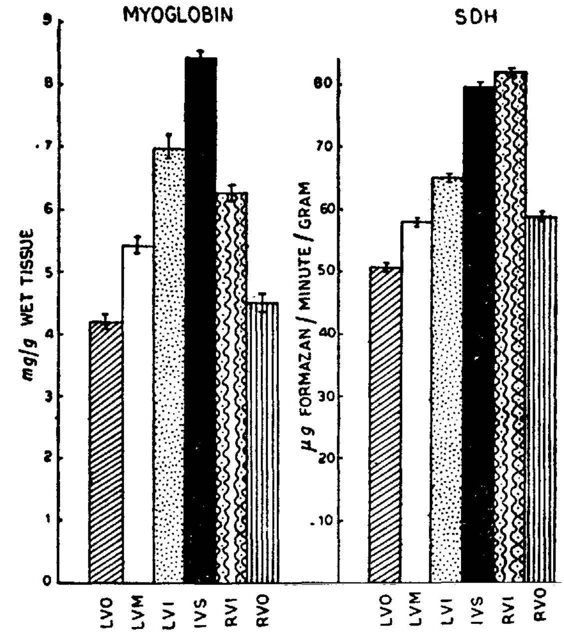

Figure 2 Myoglobin content and SDH activity of the Ox heart (*For abbreviations refer figure 1*)

Results

The inner layer of the left ventricle has a higher myoglobin content than either the middle or the outer layers ($p < 0.001$) (figure 2). Similarly, the inner layer of the right ventricle has more myoglobin than the outer layer ($p < 0.001$). The highest level of myoglobin, however, is found in the interventricular septum (figure 2). SDH activity is also higher in the inner myocardial regions than the outer zones of both left and right ventricles ($p < 0.001$) (figure 2). The inner layer of the right ventricle has more SDH activity than the inner region of the left ventricle ($p < 0.05$).

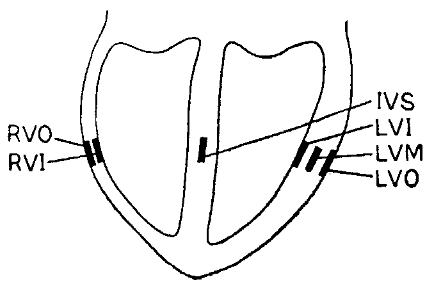

Figure 1 Schematic representation of the regions of the Ox heart under study, **RVO**, right ventricle outer region; **RVI**, right ventricle inner region; **IVS**, interventri cular septum; **LVO**, left ventricle outer region; **LVM**, left ventricle middle region; **LVI**, left ventricle inner region

A higher concentration of glycogen is discernible in the outer layer than in the middle and inner layers of the left ventricle ($p < 0.001$) (figure 3). The pattern is similar in the right ventricle in as much that the outer layer has more glycogen than the inner layer ($p < 0.005$) (figure 3). Further the outer region of the left ventricle shows a higher level of glycogen than the outer region of the right ventricle ($p < 0.001$). Total lipids also exhibit region-wise differences in distribution (figure 3). The inner layers show more lipids than the outer layers. Regarding myoglobin, the interventricular septum has the highest amount of lipid (figure 3).

Discussion

The data suggest that distinct variations exist in the distribution pattern of myoglobin, fuel reserves such as glycogen and lipid and succinic dehydrogenase activity in the various regions of the ventricles. The inner regions of the ventricular myocardia possess a greater amount of myoglobin than the outer layers. Myoglobin content of the muscle generally increased with an enhanced percentage of red fibres (Beecher et al. 1965). Whether it is the vertebrate heart which undergoes rhythmic contractions or the flight muscles of birds, myoglobin would supply the muscles with oxygen during contraction whereas the blood supply is assumed to be much reduced. Further, histochemical studies of the myocardium and its vasculature in rats have shown that the arterioles which supply the inner regions of the myocardia have a higher capacity for aerobic metabolism, than do the arteries, while the more abundant arteries seen on the outer region of the myocardia are more adapted for anaerobic metabolism (Cannon et al. 1980). Myoglobin is found in higher concentration in the red muscles of fishes which are concerned with slow and repetitive activity (Love 1970). Accordingly the higher myoglobin values of the inner region of the Ox myocardia may possibly be a physiological adaptation to facilitate an adequate oxygen supply in accordance with its greater intramyocardial pressure concomitant to hemodynamic needs and relatively lower vascularisation.

The present study indicates variations in glycogen levels in the different regions of the Ox ventricular myocardia. The left ventricle has a higher quantum of glycogen than the right ventricle possibly because the former has to exert a relatively greater haemodynamic force in order to pump the blood into arterial circulation. Further, the outer region of the ventricles shows a greater amount of glycogen than the inner region. This may be due to the fact that fibres of the outer region are relatively more glycolytic than those of the inner region (Eapen & Alexander, Unpublished data).

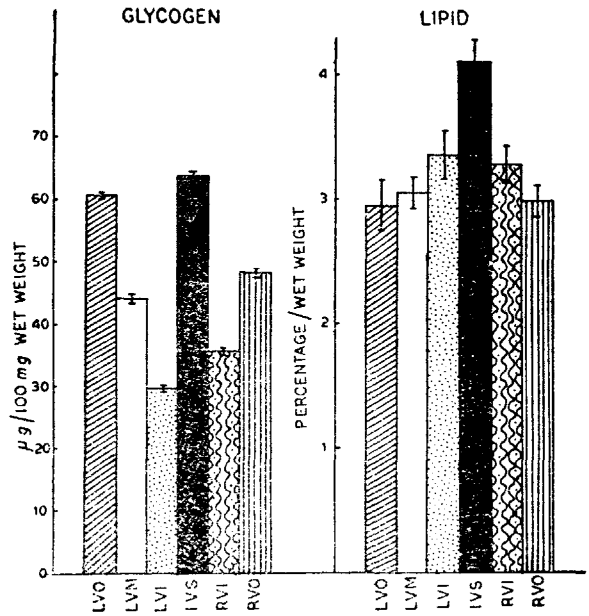

Figure 3 Glycogen and total lipid content of the Ox heart (*For abbreviations refer to figure 1*)

The levels of total lipids of the inner region of the right and left ventricles are higher than those of the outer region. Since the myocardia contract rhythmically deploying considerable haemodynamic force, they have the highest capacity for aerobic metabolism among mammalian muscles and concomitantly utilize lipids preferentially for their major energy requirements. The cardiac and skeletal muscles with higher rates of oxidative metabolism derive their energy for contraction from the aerobic breakdown of a variety of substrates especially fatty acids (Romanul 1965). The cardiac muscles, having to contract continuously cannot possibly derive their entire energy requirements for steady contraction from the breakdown of stored tissue glycogen having varied levels of metabolism, properties of contractile proteins and by the differential action of a number of enzymes (Rubel 1968). When oxygen is available, mammalian hearts are found to utilize fatty acids as the major fuel reserves and in the anoxic or hypoxic hearts, energy liberation through glycolysis may increase by 10–20 fold (Neely & Morgan 1974). Generally higher myoglobin content is associated with higher succinic dehydrogenase activity (Lawrie 1952). Accordingly, the variation in the lipid content of the fibres of the outer and inner region of the myocardia can be correlated to their specific fuel preferences.

The differential gradients in the levels of metabolites indicate the level of physiological adaptation of each region of the heart. Relatively higher levels of lipids, succinic dehydrogenase and myoglobin were observed in the inner regions than in the outer regions of the heart of Ox, *Bos*. sp. This suggests that the inner region of the myocardia depends on lipids and hence they have to depend on aerobic degradation of fatty acids (Bing et al. 1953).

Biochemical assay of different regions of the ventricular myocardia have shown that the inner regions of the left and right ventricles possess a relatively higher SDH activity than the outer layers. The myocardial fibres derive their energy predominantly from oxidative lipid metabolism and accordingly correspond to aerobic type of muscle. The SDH activity in different fibres can be correlated with their oxidative metabolic capacity. Further, the higher levels of SDH activity would possibly indicate the mitochondrial density and concomitant higher oxidative capacity. The high SDH levels and the resulting oxidative capacity are generally characteristic features of red muscle fibres effecting a higher functional efficiency which is absolutely essential for a constantly pumping muscular organ like the heart. Biochemical and ultrastructural studies on the inner and outer myocardial layers of the ventricles from tuna fish revealed a higher respiratory enzyme activity and the mitochondria of the inner layer were found to have a higher cristae density than those from the outer layer (Basile et al. 1976).

Different regions of the myocardia exhibit disparate contractile performance which is facilitated by mass of muscles as the main fuel source of energy. Conversely, the outer region exhibited a higher glycogen content than the inner region and the fibres of the outer region are relatively more glycolytic than the fibres of the inner region.

Acknowledgements

We are grateful to Dr K K Nair, Veterinary Surgeon, Corporation Slaughter House, Trivandrum for the assistance in collecting Ox heart samples. Our thanks are due to Mr T I Jacob for the technical assistance. One of us (GE) gratefully acknowledges the award of a Junior Research Fellowship by the Council of Scientific and Industrial Research, New Delhi.

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