CHAPTER 10

A QUANTITATIVE STUDY OF SUCCINIC DEHYDROGENASE IN THE MUSCLES OF A FEW REPRESENTATIVE TYPES OF INSECTS.

It has been more or less well established that animals which indulge in long and sustained muscular activity have a higher metabolic rate in general and their muscles possess considerably high concentrations of the oxidative enzymes. It is also known that in the case of the flight muscles of some birds, bat and insects which indulge in sustained activity chiefly burn fat for energy (Weis-Fogh, 1952; Zebe, 1954; George and Jyoti, 1955a, 1955b, 1957; George and Naik, 1958a; George and Scaria, 1956, George, Susheela and Scaria, 1958a; George, Vallyathan and Scaria, 1958). These animals have got considerable amounts of fat stored in their muscles, and high concentrations of an emzyme (lipase) which can split up the neutral fats into fatty acids and glycerol. A recent study on the SDH concentration in the flight muscles of some representative types of birds (George and Talesara, 1961) have revealed that there do exist a relationship between the matabolic activity, the capacity to utilize fat for energy and the concentrations of various oxidative enzymes. In the previous chapters it has been observed that in the insect muscles whether flight muscles or leg muscles the concentration of SDH has a relation with the lipase activity of the same. In the present investigation, the SDH activity in the wing muscles of a number of insects belonging to different orders and indulging

in different modes of muscular activity, have been studied with a view to obtain more quantitative data.

Materials and Methods

The following insects were caught from their natural environments and used immediately.

Order	Name
Orthoptera	*Periplaneta americana*
"	*Mecopoda elongata*
Lepidoptera	*Danais chrysippus*
"	*Appias libythea*
"	*Papilio demoleus*
"	*Hypolimnas misippus*
"	*Hippotion rafflasi*
"	*Hippotion celerino*
"	*Dielephila nerii*
"	*Acherontia styx styx*
"	*Theretra nessus*
"	*Ophideres meterna*
"	*Nyctipao macrops*
Coleoptera	*Oryctes rhinoceros*
"	*Cybister confusus*
"	*Dineutes indicus*
"	*Anthia sexguttata*
Hymenoptera	*Xylocopa*
"	*Apis mellifera*
Diptera	*Musca domestica*

The method employed was the one, developed by Kun and Abood (1949) using TTC as the hydrogen aceptor. The procedure was exactly as described in the earlier chapters. The muscles of the above mentioned insects were removed carefully from their attachments and after determining the weight an homogenate of known percentage was prepared in ice cold distilled water. 1 ml of the homogenate was used to estimate the SDH activity in all the samples. SDH activity is expressed as µg formazan formed per mg. dry tissue per 15 minutes. Dry weight of the homogenate used was determined by drying the same volume of the homogenate in a pyrex weighing bottle in a hot air oven at 100°C.

Results and Discussion

The average values of SDH activities in the muscles of the various insects are presented in Table 1. It can be noticed that the insects belonging to the higher orders contain more SDH in their flight muscles than the leg muscles of the same as well as the flight muscles of the lower insects. A comparison of the oxygen uptake and SDH activity between teased leg and flight muscles has been made by Perez-Gonzales and Edwards (1954) for Preplaneta americana, Schistocerca infumata, Belostoma and Hydrophilus ater. The cockroach showed little difference between the muscles and in locust the SDH activity of the flight muscles was three times that of leg muscle. In Belostoma and Hydrophilus however, the flight muscles which are fibrillar were 15 to 20 times more active

than the leg muscles. Though the method employed by the above authors and the one adopted in the present study are different, the relative differences between the leg and flight muscles of the beetle flight muscles are more or less the same.

Table 1

Table 1. The value of SDH activity presented is the average of the number of experiments done in each case.

Order	Name	µg. Formazan/mg. dry tissue/ 15 minutes.	Expts.
Orthoptera	Periplaneta americana		
	Male	8.35	6
	Female	1.92	6
"	Mecopoda elongata	1.8	3
Lepidoptera	Danais chrysippus	11.2	5
"	Appias libythea	10.85	3
"	Papilio demoleus	12.78	3
"	Hypolimnas misippus	8.1	4
"	Hippotion rafflasi	12.0	5
"	Hippotion celerino	11.3	5
"	Dielephila nerii	9.7	6
"	Acherontia styx styx	8.7	4
"	Theretra nessus	13.1	4
"	Ophideres meterna	8.66	3
"	Nyctipao macrops	8.57	5
Coleoptera	Oryctes rhinocerus		
	Flight muscle	10.12	6
	Leg muscle	2.63	6
"	Cybister confusus		
	Flight muscle	6.25	5
	Leg muscle	0.14	5

Order	Name	µg Formazan/mg. dry tissue/ 15 minutes.	Expts.
Coleoptera	Dineutes indicus		
	Flight muscle	5.3	4
	Leg muscle	0.11	4
"	Anthia sexguttata	0.5	3
Hymenoptera	Xylocopa	10.7	3
"	Apis mellifera	12.8	8
Diptera	Musca domestica	10.4	5

It is evident from Table 1 that the higher orders of insects possess more of SDH in their flight muscles. The lepidopteran and hymenopteran insects possess the highest SDH activity. The difference obtained between the male and female cockroaches for SDH activity has been observed by Allen and Richards (1954) by the difference in oxygen uptake in the presence of added succinate. Similar metabolic differences between the male and female cockroaches have been observed by Barron and Tahmisian (1948) and Chapter 6. This difference could be related to the amount of mitochondria present in their flight muscles, the male possessing nearly three times that in the female (Kwatwaza, 1960). The grass hopper examined is not a good flier and the muscle was almost white in colour. The lepidopteran insects which have studied in

Some detail for the lipase activity and fibre diameter of the flight muscles also show some differences among different species. Most of these lepidopterans were migratory forms and their flight muscles were red in colour. It was observed in chapters 1 and 2 that there is a relationship between the lipase activity and fibre diameter of the flight muscles in insects. Though the relationship with regard to SDH activity and the fibre diameter is not as conspicuous as that with lipase, still a significant difference is nevertheless evident. This can be explained when it is realized that the presence of SDH is required in considerable amounts in muscles whether they burn aerobically fat or carbohydrates for energy. Further, fatty acid oxidation being an aerobic process the need for sufficient concentrations of oxidative enzymes is more as has been observed in chapters 4 and 8, Further support is available from the values obtained for the leg and flight muscles of the beetles studied as well as the muscles of the wingless form. This suggests that in muscles which do not indulge in prolonged activity, there is no necessity of having high concentrations of oxidative enzymes.

In the case of hymenopteran and dipteran insects it is generally believed that carbohydrate is the chief fuel, but in the present study considerable amount of SDH have been noted in their flight muscles. The giant size of the fibres and their high frequency of isometric contractions are obviously supported by the energy the aerobic metabolism of the substrates in the muscles. The efficient system of

intracellular tracheoles further promote the oxidative metabolism in these muscles. Therefore the high concentration of SDH in these muscles could be correlated with the high requirement of energy by these flight muscles. This is also supported by the fact that these muscles contain large sarcosomes (mitochondria) and nearly half of the cross sectional area of the fibre is studded with mitochondria (Chapter, 3).

From these observations it can be assumed that the concentrations of SDH in the insect muscle would be related to the high aerobic metabolism supporting the high degree of muscular activity. The high concentrations of cytochromes and the oxidative enzymes usually present in muscles which indulge in sustained muscular activity as observed by Klein,(1929); Watanabe and Williams (1951), Sacktor and his collaborators (1953a, 1953b; 1955; 1956; 1958) and the data obtained in the present dtudy, also support the above contention. The presence of lactic and glycerophosphate dehydrogenases in the insect muscle as studied by Zebe and McShan (1957) also support this view. Zebe and McShan (1957) observed that the negligible amount of LDH and the high concentrations of glycerophosphate dehydrogenase in the flight muscles of various insects utilizing fat and carbohydrates for energy did not show any relation with the fuel consumed and they suggested that insect flight muscles are adapted for aerobic metabolism.

The SDH activity of insect flight muscle is higher than that of any vertebrate skeletal muscle hitherto known. A recent investigation by George and Talesara (1961) on the SDH content of a few representative types of birds exhibiting different modes of activity shows that birds indulging in sustained muscular activity such as migratory flights, possess more of this enzyme than the poor fliers. The values obtained for the insect muscle indulging in sustained activity are twice more than the highest values obtained for bird muscles. The above authors also showed a relationship between the fibre diameter, body weight and the SDH activity. In birds such a relationship is comparatively more obvious due to the limited variations in the activity and the structure of the muscle. Insects on the other hand are a much larger group with greater diversities.