CHAPTER 5

LIPASE ACTIVITY IN THE ADIPOSE TISSUE OF VERTEBRATES

Lipase was detected in the adipose tissue by earlier workers (Quagliariello and Scoz,1938; Gomori,1946). It has already been mentioned that some authors have expressed doubt as to the occurrence of a 'true lipase' in the adipose tissue and that the present studies indicate the presence of a 'true lipase' in this tissue. Here the results of a quantitative study of the lipase activity in the adipose tissue of the frog (Rana tigrina), lizard (Calotes versicolor), fowl (Gallus domesticus), rosy pastor (Pastor roseus), pigeon (Columba livia) and bat (Hipposideros speoris), are reported.

Material and Methods

In all the cases, visceral adipose tissue was used except of the bat in which the subcutaneous adipose tissue was taken. The animals were killed by decapitation and the tissues taken after most of the blood was drained off. The adipose tissue covering the coils of the intestine in the fowl, rosy pastor and pigeon, the same tissue found in association with the reproductive organs in the frog and lizard and the subcutaneous, interscapular brown adipose tissue and the yellow adipose tissue from the lower lateral sides of the abdomen of the bat, were carefully removed, separately minced and defatted in two changes of ethyl ether at room temperature for 2 hours. The defatted tissue was dried in a vacuum desiccator at room temperature. An extract of each was made in cold distilled water by grinding with clean sand by means of a glass rod in a test tube for 1 hour in

cold (4°C), centrifuged at approximately 2500 r.p.m for 5 minutes and the resulting supernatant was used as the enzyme material.

The method of assay was a manometric method adopted from Martin and Peers (1953) using the Warburg apparatus, with a bicarbonate carbon dioxide buffer system of pH 7.4 at 37°C, tributyrin being used as the substrate. The reaction flask contained 1.5 ml of 0.025 M bicarbonate solution, and 1 ml enzyme solution in the main chamber and 0.5 ml 4% (v/v) tributyrin in 0.0148 M bicarbonate solution (emulsified by shaking with a drop of "Tween 80") in the side arm thus making up a total fluid volume of 3 ml. The procedure was the same as the one adopted for the study of lipase activity in the fat body of the locust (Chapter 4). The enzyme activity was calculated on the basis of the protein concentration of the enzyme solution used and is expressed as the number of µl CO_2 produced/ mg protein/ hour. Protein was estimated according to the micro-Kjeldahl steam distillation method for total proteins (Hawk et al, 1954).

Results and Discussion

Table 1 presents the lipase value in the adipose tissue of the six vertebrates studied. The study of lipase activity in the rosy pastor and fowl adipose tissue, gave contrasting results, the former showing very high lipase activity than the latter. This is in conformity with the results of the histochemical study on the adipose tissue of these two birds (Chapter 2, II).

Table 1

Animal	Lipase activity: µl CO_2/ mg protein/ hour
Frog	16.43
Lizard	275.85
Fowl	21.02
Rosy pastor	254.40
Pigeon	246.40
Bat	
(yellow)	37.85
(brown)	30.45

A possible explanation for the higher lipase concentration in the rosy pastor adipose tissue, that it is an indication of higher metabolic activity especially involving lipid metabolism was also suggested. This is noteworthy due to the fact that the rosy pastor is a migrant and the birds used in the present study were procured few days before they started on their migration to Europe. For the synthesis of the large amounts of fat (mainly glycerides) accumulated in the adipose tissue during premigratory period, considerable amount of lipase should be deemed essential. In the fowl also at times adipose tissue is found to contain large quantities of fat, but the lipase activity when calculated on the basis of the protein concentration of the enzyme solution, remains low. This may be because, all the fat that is found in the adipose tissue of the fowl may not be synthesized there but transported from other sites of synthesis and deposited in the adipose tissue. This view is apparently supported by the findings

of Bumgardner (1957) who investigated the origin and mobilization of the fatty acids of the adipose tissue of avian embryos using tracer techniques. According to this author a large majority of the synthesized fat found in the adipose tissue of the chick embryo has its origin in other tissues. But this however, does not disprove the fact that the synthesis of fat takes place in the adipose tissue itself. If so, the ability to synthesize fat (glycerides) by the adipose tissue may be different in different animals and the concentration of lipase may be regarded as an index of the varying capacities for fat synthesis. The amount of lipids present in the adipose tissue of the frog and the lizard is reported to be almost the same (Shah,1952). But the lipase activity in this lizard adipose tissue is comparatively very high. The condition in these two animals parallels that of the fowl and rosy pastor respectively. I have also observed that there is a seasonal variation in the lipase concentration of the frog adipose tissue. The value of 16.43 µl CO_2/ mg protein/ hour (Table 1) obtained during midsummer (May) rose to 21.60 µl CO_2/ mg protein/ hour during the rains (July). These observations lead to show that irrespective of the fact that an animal is aquatic or terrestrial the lipase activity in the adipose tissue is an index of the extent of fat synthesis or breakdown in it. It also shows that lipase activity is less in the adipose tissue of those animals in which the fat is gradually built up and gradually used up, but on the other hand it is very high in those in which fat is built up rapidly for a large scale utilization in a short period as in the case of the migratory

birds before and during migratory flights. Even though pigeon is not a migrant, it is known to be a good flier indulging many a time in sustained flight, and the energy for this is reported to be obtained mainly from the utilization of fat. The high figure in the lizard which may be true for other reptiles as well, is perhaps due to the fact that they are constantly threatened with starvation during some seasons and also they lay numerous, large, yolked eggs. Moreover for a terrestrial reptile more fat stored means more water conserved and more fat utilized means more metabolic water made available to meet the acute conditions of draught. Further it should be stated that the fowl adipose tissue was reported to be containing only lipoprotein lipase and no lipase (Korn and Quigley,1957). But now it is possible to say that not only a 'true lipase' is present but also that it occurs in appreciable quantities in the adipose tissue of the fowl.

From the results obtained it is evident that the yellow adipose tissue of the bat has a higher lipase activity than the brown, but at the same time the difference in concentration of the enzyme in the two types of adipose tissue is not sufficient to show a pronounced difference in the histochemical preparations. The amount of neutral fat is shown to be more in the yellow adipose tissue than in its counterpart (Fawcett,1952) and the fat in the yellow adipose tissue is reported to be mobilized in a greater amount and faster than in the brown adipose tissue (Remillard,1958). These could be accounted for, because of the presence of a higher concentration of lipase in the yellow adipose tissue.