

PHOSPHOMONOESTERASES IN DIFFERENT TISSUES OF THE DESERT LOCUST, SCHISTOCERCA REGGARIA (FORSKAL)

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Phosphomonoesterases were determined quantitatively in different tissues of desert locust according to the procedure of Naqvi *et al.* Acid phosphatase activity was found to be high in Malpighian tubules (proximal and distal end both), caecae, midgut and male accessory glands. It was moderate in hindgut, spermatheca, oviduct, salivary glands, ovariole, blood, heart, testes, foregut, brain and low in male ducts, ova, fat bodies, nerve cord and integument. Activity was negligible in muscles and absent in tracheae.

Alkaline phosphatase activity was recorded high in male accessory glands, caecae, midgut, and salivary glands. The activity was moderate in foregut, testes, ovariole and low in blood, male ducts, hindgut, fat bodies, oviduct, Malpighian tubules (distal and proximal both), and spermatheca. It was negligible in brain and absent in ova, integument, muscles, heart, nerve cord, and tracheae. The results were compared with the histochemical findings of Qureshi³ for the same insect and were correlated with different physiological functions.

Introduction

The work on biochemical characterization and histochemical localization of phosphomonoesterases in different tissues of animals and plants has been in progress since 1923,¹ but Nakamura² studied the phosphatase activity in insect tissue for the first time. Later on, histochemical and biochemical studies were done by several workers. All those workers who histochemically localized these enzymes, correlated the color intensity of the end product of the chemical reaction with the quantity of the enzyme present in a tissue. In the present investigation the biochemical studies of the phosphatases have been carried out to compare the histochemical findings of Qureshi,³ in different tissues of the same insect and to assign probable functions to these enzymes.

Material and Methods

Acid and alkaline phosphatase activity was determined in desert locust according to the methods^{4,5} modified by Naqvi *et al.*⁶ Colorimetric determination was done on Beckman DB 620 spectrophotometer.

Reagents.—Disodium *p*-nitrophenyl phosphate was used as substrate and *p*-nitrophenol as colorimetric standard. Solutions were prepared according to Sigma Technical Bulletin.⁷ Acid and alkaline buffers were prepared according to procedures of refs. 8 and 9.

Enzyme Source.—For enzyme source 28-day old adult insects were used for the estimation of the enzymes. These were chilled at -15°C in a cold chamber and were dissected in chilled water. Tissues after separation were kept in cold double

distilled demineralized water. After grinding the filtrate was collected in a microfilter tube under moderate suction pressure through a 2-mm thick glass fibre layer, placed in a Gooch crucible. Blood was sucked out of the body through a microsyringe. All the tissues were weighed prior to the grinding and a 0.3% homogenate was prepared in buffer of known pH.

Procedure.—Procedure and optimum factors including standard curves for the determination of acid and alkaline phosphatases were followed as described by Naqvi *et al.*⁶ Each experiment was repeated five times. Activity of the enzyme was measured in terms of micromoles of *p*-nitrophenol liberated per mg fresh tissue weight under standard assay conditions and was divided into four categories: (a) high activity—80 μmol or above. (b) moderate activity—40 μmol to 79 μmol (c) low activity—10 μmol to 39 μmol (d) negligible activity—5–9 $\mu\text{mol}/\text{mg}$.

Results and Discussion

Acid phosphatase activity as indicated by Fig. 1 was found to be 120 $\mu\text{mol}/\text{mg}$ in the distal end of the Malpighian tubules. It was 115 μmol in caecae, 110 in midgut, 90 μmol in male accessory glands and 90 $\mu\text{mol}/\text{mg}$ in proximal end of the Malpighian tubules. The activity was moderate i.e. 75 in salivary glands, 62 in ovariole, 60 in foregut and 48 $\mu\text{mol}/\text{mg}$ in brain. It was 38 in nerve cord and 18 $\mu\text{mol}/\text{mg}$ in integument. The activity was negligible i.e. 9 $\mu\text{mol}/\text{mg}$ in muscles and absent in tracheae.

Alkaline phosphatase activity was found to be 157 $\mu\text{mol}/\text{mg}$ in male accessory glands as indicated by Fig. 2. It was 95 in caecae, 90

in midgut and 87 $\mu\text{mol}/\text{mg}$ in salivary glands. The activity was 48 in foregut, 48 in testes and 43 $\mu\text{mol}/\text{mg}$ in ovariole. It was 35 in blood, 32 in male ducts, 22 in hindgut, 17 in fat bodies, oviduct and distal Malpighian tubules, 15 in proximal Malpighian tubules and 13 $\mu\text{mol}/\text{mg}$ in spermatheca. The activity was 9 $\mu\text{mol}/\text{mg}$ in brain and almost absent in ova, integument, muscles, heart, nerve cord and tracheae.

High activity of acid phosphatase in Malpighian tubules has been reported by other workers. Among them Przelecka and Wroniszewska^{14,15} has pointed out that the activity is higher in the distal end than the proximal end of the Malpighian tubules. During excretion maximum absorption takes place through the proximal end which mostly plays a role of pouring excretory fluid in the intestine.¹⁷ Thus the acid phosphatase probably seems to be

helping in absorption and transportation of materials.

The high acid phosphatase activity in caecae and midgut may be perhaps due to absorption and assimilation processes in the mesenteron. This coincides with the reports of previous workers.^{3,5,12,18,19} Acid phosphatase activity was high in male accessory glands also, but it was less than alkaline phosphatase. Higher activity in these glands may be correlated with transportation of the secretion of glands.

Moderate acid phosphatase activity in the salivary glands may be related with transportation of the saliva. In foregut also the acid phosphatase activity is moderate and almost equal to the alkaline phosphatase. This may be due to lesser absorption and secretion phenomena in this region. There is a lesser decrease in the acid phosphatase

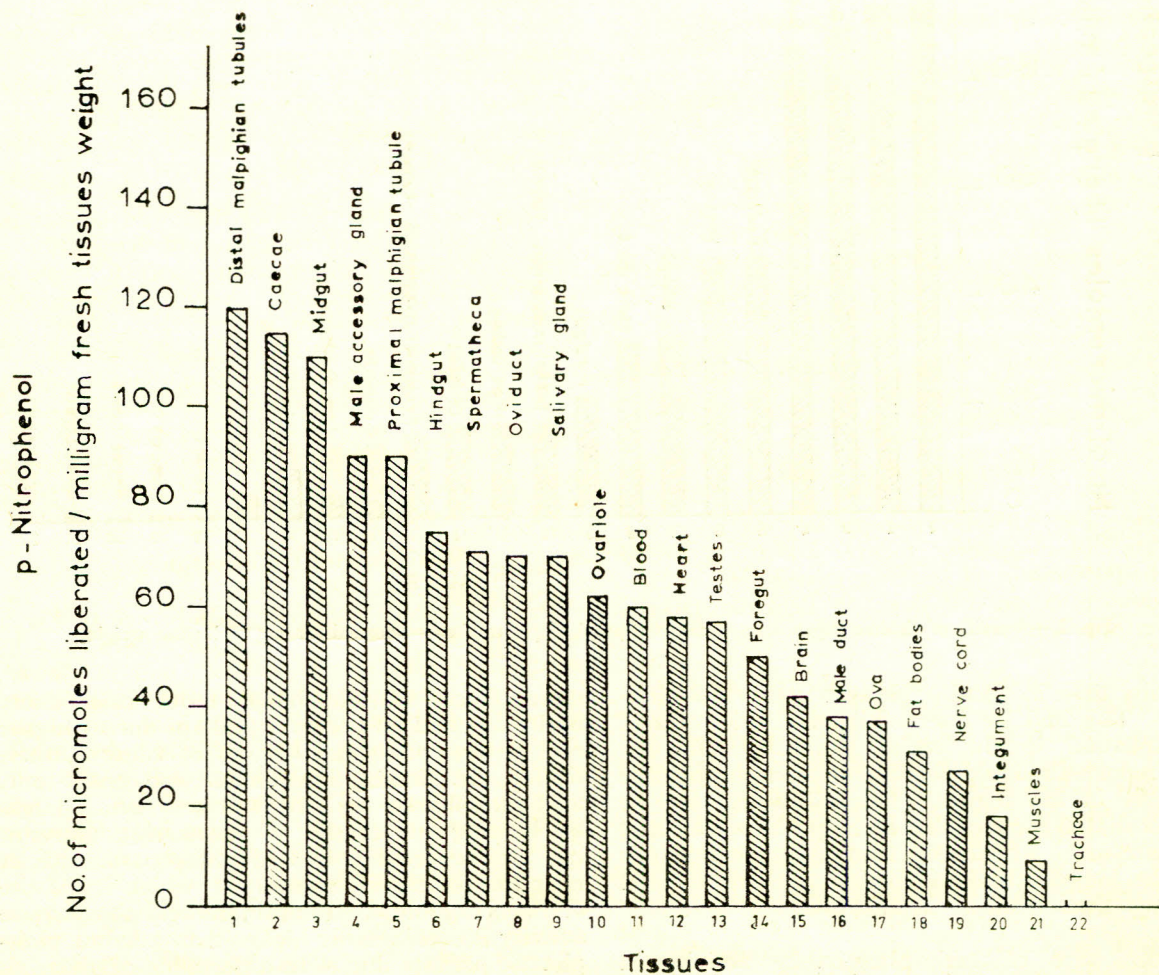


Fig. 1.—Activity of acid phosphatase in terms of μmol of *p*-nitrophenol liberated per mg fresh tissue weight.

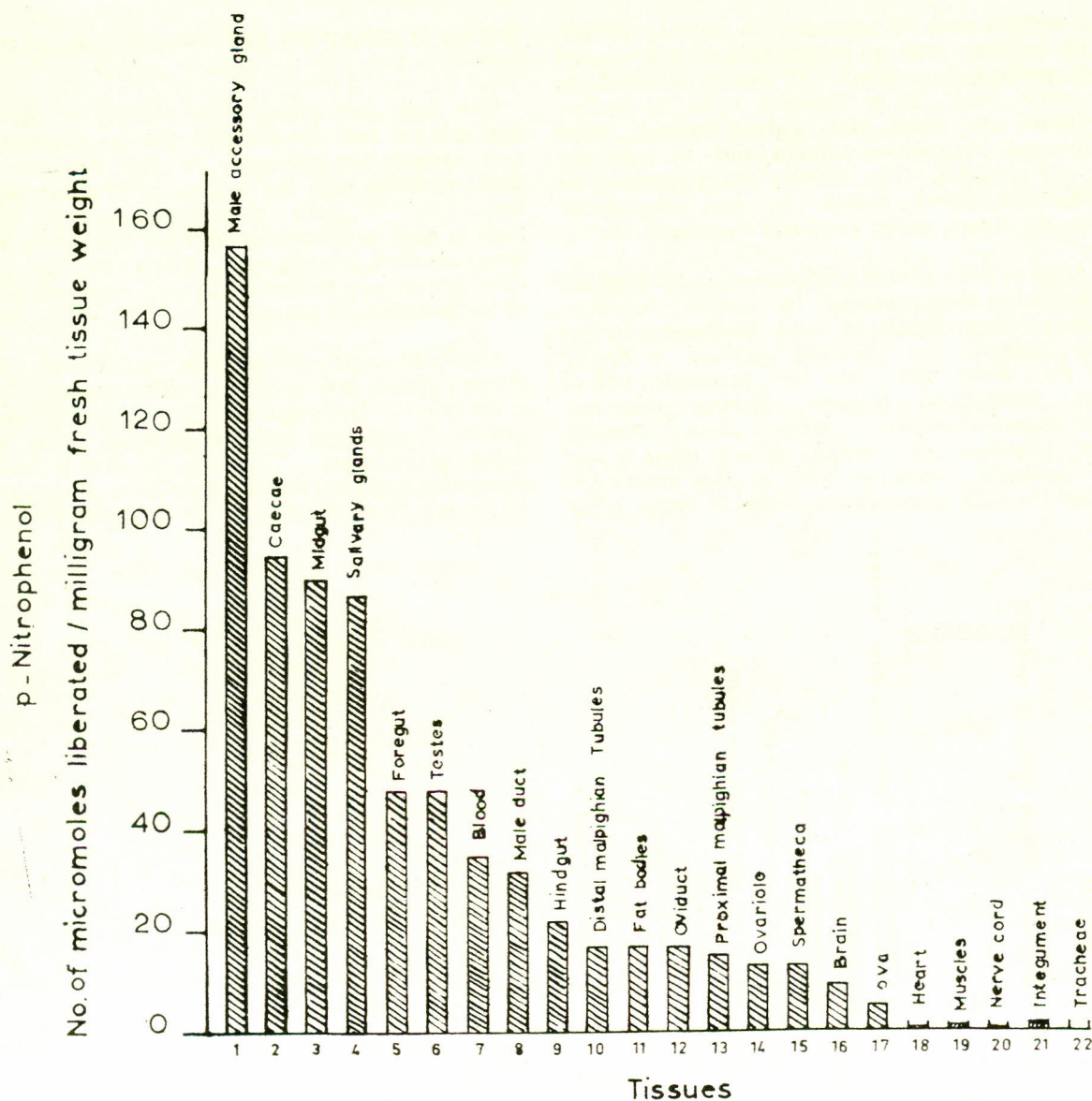


Fig. 2.—Activity of alkaline phosphatase in terms of μmol of *p*-nitrophenol liberated per mg fresh tissue weight.

activity than in alkaline phosphatase in the hindgut. This may be due to higher activity of acid phosphatase in the rectal glands.¹³ Qureshi³ has reported marked decrease of acid phosphatase activity in this region. This difference may be either due to the fact that he might have localized the enzyme in a region not containing rectal glands or the longer starvation period (48 hr) he used during histochemical localization.

Dominance of acid phosphatase activity in

spermatheca, ovariole, blood, testes, male ducts, brain, ova and fat bodies is perhaps due to higher rate of absorption and transfer of solute in these tissues. In nerves, integument and heart only acid phosphatase activity is present which might be helping in transfer of materials. Qureshi³ has also reported only acid phosphatase activity near the pericardial membrane, which is the site of transfer of solute. In brain the acid phosphatase activity is considerably higher than alkaline phosphatase perhaps due to transportation of materials

through cerebral membrane Qureshi³ has also reported presence of acid phosphatase only in the peripheral region of brain.

In the egg at earlier stages of development, acid phosphatase is prominently higher than alkaline phosphatase⁶ which is perhaps due to higher rate of absorption and transportation of materials. Moreover, Qureshi³ has reported a dense distribution of acid phosphatase near the chorion of the egg. These findings and the present data indicate that acid phosphatase is more related with absorption, exchange of materials and transfer of solute.

Dominance of alkaline phosphatase activity in the male accessory glands and salivary glands indicate that alkaline phosphatase is related with the secretion phenomenon. The high activity of alkaline phosphatase in the midgut and caecae may be due to the digestive enzyme secreting glands of these parts. This is in accordance with the findings of others.^{3,6,13,19-25} Comparatively high alkaline phosphatase activity in testes and ovarioles, than other tissues may be correlated with the development as has been pointed out by Banerjee.²⁶ High alkaline phosphatase activity in the salivary glands may be compared with the previous findings.^{6,13,19,27-30}

By comparing the histochemical results of Qureshi³ and the present biochemical findings it was found that both the findings coincide except for slight variation in hindgut and salivary glands. However, the general conclusion on the basis of the pattern of distribution of these enzymes is similar. Thus it seems quite probable that an important function of acid phosphatase is to help in transport of materials and absorption while alkaline phosphatase is related more with secretion, tissue growth and development.

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