Prophenolase and the Role of Mehlis' Gland in Helminths

Enzyme phenolase is known to occur along with phenol and protein in vitelline cells of helminths having phenolically tanned eggs[1]. Yet the reasons as to why tanning does not occur before the shell globules coalesce to form the shell, and the nature of the association of the precursors of sclerotin while in vitelline cells, as well as the role of Mehlis' gland in this regard, have defied helminthologists[1-6], inspite of their realization that Mehlis' gland, by its location around the ootype, may have some role in the formation of the shell.

In an attempt at elucidating these problems, studies on the histochemistry of vitelline cells and Mehlis' gland of monogeneans *Pricea* and *Protomicrocotyle* were undertaken. As this study has yielded some interesting information bearing on the questions mentioned above, it is therefore being reported in this communication.

Materials and methods. Monogenetic trematodes *Pricea* and *Protomicrocotyle* obtained from the gills of marine fishes *Scomberomorus guttatus* and *Caranx sexfasciatus* respectively were employed in the study. Phenolase in these helminths was detected by following the method of SMYTH[7]. The experiments designed in the study were as follows: 1. Live specimens and specimens fixed and stored in 5% neutral formalin for 15 min were separately incubated in catechol. 2. Specimens stored in fixatives (5% neutral formalin and 70% alcohol) for 24 h were separately incubated in catechol. 3. Live specimens and specimens fixed and stored in 5% neutral formalin for 15 min were separately treated in 0.2% sodium oleate (prepared in phosphate buffer of pH 7) for 30 min and then separately incubated in catechol. 4. Live specimens and specimens fixed and stored in 5% neutral formalin for 15 min were injured by rendering them asunder and were separately incubated in catechol.

Results and discussion. Results obtained on specimens in experiment 1 reveal that vitelline cells in the vitellaria were negative whereas the shell precursors released from these cells and egg shell in the uterus after their passage through the ootype surrounded by Mehlis' gland were positive. The results suggest that the enzyme, while in vitelline cells in situ, remains unreactive to catechol but is enabled to react with it on coming under the influence of Mehlis' gland secretion.

The catechol negative reaction revealed by vitelline cells in situ in experiment 1 is at variance with the observations reported by previous workers[1-6]. To determine whether the catechol-positive reaction reported by previous workers was due to the influence of storage in fixative, experiment 2 was designed and the results obtained in the specimens revealed the vitelline cells in situ to be catechol-positive: thus agreeing with the observations reported by earlier workers and suggesting that storage in fixatives has influenced the enzyme in rendering it to be catechol-positive.

To determine whether the catechol-negative reaction revealed by vitelline cells in situ in experiment 1 was due to the enzyme existing as proenzyme, experiment 3 was designed employing sodium oleate treatment, as such a treatment is known to activate prophenolase of insects[8]. The results obtained on specimens in experiment 3 revealed vitelline cells in situ to be catechol-positive, suggesting thereby that not only the enzyme exists as prophenolase in them but is also activated by sodium oleate treatment.

To confirm the observation as regards the occurrence of phenolase as proenzyme in these helminths, experiment 4 was designed based on the evidence of injury causing an activation of cuticular prophenolase in insects[9]. Results obtained on specimens in this experiment revealed that vitelline cells at the regions of injury were catechol-positive whereas those in the intact regions were catechol-negative; this supports the observation made that phenolase in the vitelline cells in situ exists as prophenolase.

Conclusions. This study provides direct evidence not only of the existence of phenolase as proenzyme (to be reported for the first time in helminths) but also of the role of Mehlis' gland in activating the enzyme. By remaining a proenzyme it constitutes a factor responsible for preventing the precursors of sclerotin from tanning in situ.

The author has recently obtained evidence from a continuation of the study[10] for the presence of sulphated acid mucopolysaccharide in the vitelline cells in association with phenol which constitutes as an in-built inhibition system preventing the precursors from tanning in situ. Mehlis' gland secretion functions as a releasor of this inhibition, facilitating the process of tanning which will be published elsewhere[11].

Zusammenfassung. Die Phenoloxydase der Vittellinzellen von Trematoden wirkt als inaktive Prophenoloxydase und bedingt, dass die von Dotterstockzellen synthetisierten Vorläufer von Sklerotin in situ nicht tanniert werden. Das Sekret der Mehlis-Drüse beeinflusst die Fermentaktivierung.

K. RAMALINGAM

Department of Zoology, University of Madras, Madras 5 (India), 3 February 1970.

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The Isolation and Identification of 2-Amino-3-Hydroxyacetophenone from the Urine of Rats

In a survey of aromatic metabolites in the urine of rats following ingestion of tryptophan, it was noticed that the urine contained a small amount of substance with a greenish blue fluorescence. This compound has now been identified as 2-amino-3-hydroxyacetophenone. It was reported that 2-amino-3-hydroxyacetophenone-*O*-sulfate were excreted in a small amount in some normal human urines, and in appreciably larger amounts in certain pathological urines[1]. But there are no reports on the occurrence of 2-amino-3-hydroxyacetophenone in any