

*phiprion* while in *Mugil* the deep part of this layer shows very strong reaction, and the superficial part shows 4 thin bands with weak or moderate reaction. Optic layer: the inner part, corresponding to afferent retinal fibres, shows no reaction; the outer part, corresponding to the area where the marginal neurons of the optic layer are mainly arranged, exhibits very strong reaction. Marginal fibrous layer: no reaction.

In *Scomber* (Figure 4) a distribution pattern similar to that of *Mugil* is present but the intensity of the histochemical reaction appears reduced in all layers. In *Gobius* (Figure 5) AChE localization in optic tectum differs from that just described for the other species: in deep and central tectal layers AChE activity appears substantial, but the distribution pattern does not show marked variations among the different layers, while the marginal layer, which is lacking in histochemical reaction in all the species described above, exhibits very strong reaction.

Some experimental and ultrastructural works<sup>8-17</sup> have outlined the synaptic arrangement and the distribution pattern of fibre systems in teleost optic tectum. These results help to explain the laminar distribution of AChE and the role played by the enzyme. Previous observations<sup>2,3</sup> and the results of present work show a prevalent AChE distribution which suggests that most of the sensitive discharge and some important systems of stimuli propagation and modulation might be mediated by cholinergic mechanisms in teleost optic tectum. This con-

clusion is supported by prominent AChE activity at level of inner and outer gray layers, periventricular gray layer and marginal neuron layer, i.e. all layers which directly or indirectly receive the input of sensitive stimulation.

The above-mentioned AChE distribution pattern, however, is not the only one among the teleosts. In fact in the optic tectum of *Gobius* among seawater teleosts and *Ictalurus* among freshwater ones<sup>3</sup>, an alternative type of AChE distribution exists. This different AChE distribution might be indicative of specific differences in synaptic patterns among teleosts or differences in chemical mediators between analogous synaptic systems. It seems likely that further experimental and ultrastructural studies on the optic tectum of teleosts with dissimilar AChE localization will be necessary in order to solve the problems arising from histochemical analysis.

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## Copper Utilization During Embryogenesis of *Palaemon lamarrei*

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**Summary.** The pattern of copper utilization during the embryogenesis of the freshwater prawn, *Palaemon lamarrei* has been described. Throughout the incubation period, lasting for 14 to 16 days, the egg of *P. lamarrei* is permeable to salts and against a concentration gradient, it absorbs 11  $\mu\text{g}$  of salt from the freshwater medium. Out of this total salt uptake, as much as 0.143  $\mu\text{g}$  is due to the absorption of copper. Intake of copper, as a function of incubation time, exhibited a more or less similar trend to that of total salt intake and this can be attributed to the increased synthesis of haemocyanin.

Unlike in marine crustaceans<sup>1</sup>, the required salts for the normal development of the embryo in freshwater have to be either made available along with the yolk in the egg and/or have to be absorbed against a great concentration gradient. Among the trace elements, copper is an important element present in enzyme systems as well as respiratory pigments of crustaceans<sup>2</sup>. There are numerous publications dealing with the utilization of energy and

substance in several marine crustaceans<sup>3-6</sup>. VON HENTIG<sup>7</sup> has studied the yolk utilization in *Artemia salina*. Apart from this, there is no publication which deals with the utilization of yolk and especially copper during the embryogenesis of other freshwater crustaceans. The present paper reports on the pattern of utilization of copper during the different developmental stages of the freshwater prawn, *Palaemon lamarrei*.

**Material and methods.** Berried female specimens of *Palaemon lamarrei* (Edwards) were collected from the Bellandur fish farm, near Bangalore. The prawns caught in several collections throughout 1969-1972 were transferred into individual glass battery jars containing aerated freshwater. The egg mass was slowly released from the plumose hairs of the pleopods onto a glass slide and the

Changes in ash and copper content of developing eggs and zoea of *Palaemon lamarrei*

Develop- mental stage	Ash content		Copper content	
	$\mu\text{g}/\text{egg}$	% ash/egg	$\mu\text{g}/\text{egg}$	$\mu\text{g}/\text{g egg}$
I	15.30 $\pm$ 0.82	3.10 $\pm$ 0.12	0.173 $\pm$ 0.002	338.7 $\pm$ 4.10
II	19.20 $\pm$ 0.44	3.90 $\pm$ 0.12	0.171 $\pm$ 0.006	356.3 $\pm$ 12.70
III	19.80 $\pm$ 0.38	4.30 $\pm$ 0.33	0.182 $\pm$ 0.018	398.3 $\pm$ 39.40
IV	22.10 $\pm$ 0.92	4.90 $\pm$ 0.10	0.245 $\pm$ 0.014	544.8 $\pm$ 32.50
V	22.90 $\pm$ 0.78	5.20 $\pm$ 0.22	0.286 $\pm$ 0.033	623.9 $\pm$ 71.60
VI	26.30 $\pm$ 0.48	7.07 $\pm$ 0.32	0.316 $\pm$ 0.019	736.7 $\pm$ 43.80

<sup>1</sup> J. NEEDHAM, *Biochemistry and Morphogenesis* (The University Press, Cambridge 1950), p. 785.

<sup>2</sup> W. WIESER, Helgoländer wiss. Meeresunters. 15, 282 (1967).

<sup>3</sup> T. J. PANDIAN, Helgoländer wiss. Meeresunters. 16, 216 (1967).

<sup>4</sup> T. J. PANDIAN, Marine Biol. 5, 153 (1970).

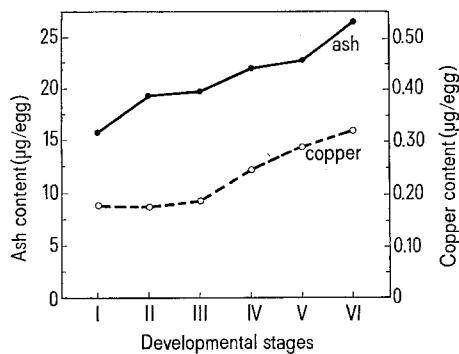
<sup>5</sup> T. J. PANDIAN, Marine Biol., 7, 249 (1970).

<sup>6</sup> T. J. PANDIAN and K. H. SCHUMANN, Helgoländer wiss. Meeresunters. 16, 225 (1967).

<sup>7</sup> R. VON HENTIG, rer. nat. Thesis, University of Hamburg (1970).

connections between the eggs removed. For ash and copper analyses, the following 6 stages of development were chosen: Stage I: Soon after spawning; eggs deep green in color, oval in shape (age: 0 to 24 h). Stage II: Transparent blastoderm visible at one end of the egg (age: ca. 48 h). Stage III: Streaks of pigmented eyes noticed. The egg slightly milky white. Occasional pulsations of the heart noticed (age: 6 to 7 days). Stage IV: Eyes and appendages fully developed (age: 10 to 12 days). Stage V: Jerky movements of the embryo noticed occasionally (age: 14 to 16 days, just prior to hatching). Stage VI: Freshly hatched zoeae (age: 16 to 17 days).

Ash content of the different stages of eggs was determined by incineration of the sample (50 mg) at 560°C for 5 h<sup>8</sup>. Copper contents of developing eggs and zoeae were estimated colorimetrically following the procedure detailed by KOLMER et al.<sup>9</sup>.



Intake of ash and copper in the eggs of *Palaemon lamarrei* as a function of incubation time.

**Results and discussion.** Marine crustacean eggs are known to be initially impermeable to water. However, PANDIAN<sup>4,5</sup> reported that they were permeable to salts throughout their development. In the eggs of *Palaemon lamarrei* also, salts were absorbed from the beginning of the incubation till hatching (Table). Ash (= salt) content increased during the developmental stages both as a percentage and quantitatively, indicating that the developing egg actively absorbed salts from the surrounding freshwater medium over a concentration gradient. It was therefore interesting to know whether this increase was due to the absorption of copper, a very important element necessary for the formation of haemocyanin. Copper content, which was about 175.3 µg/egg or 364.4 µg/g dry egg in stage I to III (age: 1 to 7 days) exhibited a marked increase in stage VI (freshly hatched zoeae). Intake of copper in the eggs of *P. lamarrei* as a function of incubation time also exhibited a more or less similar trend to that of total salt intake (Figure). In a closely related species, *Palaemon malcomsonii*, RAJYALAKSHMI<sup>10</sup> observed that the development of heart took place in 7- to 11-day-old embryos. In *P. lamarrei* pulsation of the heart was observed from 7 to 9 days of age. Hence, a marked increase in the copper content from 0.182 µg/egg in stage III to 0.245 µg/egg in stage IV and further to 0.316 µg/zoea can be attributed to an increased synthesis of haemocyanin. On the whole, it was observed that during the entire embryonic development, lasting for 14 to 16 days, out of a total salt intake of 11 µg as much as 0.143 µg was due to the absorption of copper.

<sup>8</sup> R. T. PAINE, *Ecology* 45, 384 (1964).

<sup>9</sup> J. A. KOLMER, E. H. SPAULDING and H. W. ROBINSON, *Approved Laboratory Techniques* (Scientific Book Agency, Calcutta, India 1969), p. 1022.

<sup>10</sup> T. RAJYALAKSHMI, *Proc. natn. Inst. Sci. India*, 26B, 395 (1960).

## Lipid and Phospholipid Content and Fatty Acid Composition of the Chick Lung During Embryonic Development

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**Summary.** The evolution of total phospholipid, phosphatidylcholine and phosphatidylethanolamine content of chick lung during embryonic development is in good agreement with morphological data. Saturated fatty acids are predominant. A sex-linked difference is observed in the evolution of phosphatidylcholine.

Although the existence of a pulmonary surfactant in birds was first denied<sup>1</sup>, it is now well established that the epithelium of the functional lung is lined by a surface active phospholipoproteic material, whose main features – high lecithin content, high percentage of saturated fatty acids – are similar to those of the mammalian surfactant<sup>2</sup>. Despite the particular structure of the respiratory parenchyma, which is a continuous network of air capillaries, the role of the surfactant in birds is comparable to that in mammals: it prevents transsudation of plasma fluids across the blood-air barrier, and it helps to keep the air capillaries open, to ensure sufficient ventilation<sup>3</sup>. Its physiological role is thus considerable, and its appearance in the course of foetal development is a very important step in functional differentiation.

In mammals, there is much information on normal evolution of lung lipids during foetal life, and recent results indicate that maturation of the pulmonary tissue, both morphological and biochemical, depends on the pituitary-adrenal axis<sup>4-12</sup>. In birds, the maturation of lung tissue has so far been studied only from the morphological point of view; as in mammals, the granular pneumocytes, which are responsible for surfactant synthesis, begin to differentiate at a late stage<sup>13</sup>, and as in mammals this differentiation depends on humoral factors, secreted by the hypophysis or controlled by it<sup>14-15</sup>. From the biochemical standpoint, however, there are, so far as we know, no available data, either about normal development, or about the endocrine control of lung maturation.