

chromosome condensation in metaphase can be removed by vibration of metaphase cells in hypotonic solution prior to fixation.

From the present results, it is inferred that uncoiled chromosome arms show irregularity in their capacity to absorb acetic orcein along their length. The observation of dense regions at irregular distances from each other along the arms indicates that contraction of the spiralized chromatin in metaphase chromosomes conceals details along the chromatid thread. On the other hand, these dense regions may affect the banding patterns of contracted fixed chromosomes; on this line, differential condensation of the chro-

matid spirals has already been proposed as an explanation of banding patterns^{7,13}. In favor of this aspect, adequate evidence has accumulated showing that agents causing banding (e.g. SDS¹⁵; urea¹⁶; heat¹⁷; daunomycin¹⁸) may also cause despiralization under different conditions (SDS, urea⁷; heat¹⁴; daunomycin⁹).

In comparison with other methods for producing uncoiled chromosomes^{8,9,14}, vibration provides speed and elimination of chemical treatments. A combination of the uncoiling effect of vibration with biochemical techniques may be useful in the elucidation of the general chromosome architecture.



Fig. 3. A nonvibrated Chinese hamster metaphase cell.

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Changes in glutamine levels during starvation and aestivation in the Indian apple snail *Pila globosa* (Swainson)

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Summary. The glutamine levels in digestive gland, foot and body fluids of normal, starved and aestivated *Pila globosa* were estimated. Glutamine content was decreased during starvation and aestivation. The percent decrease was more during aestivation than starvation. The decrease may be due to low glutamine-synthetase activity or increased utilization for uric acid synthesis. This may have an adaptive value during aestivation and starvation.

The Indian apple snail, *P. globosa*, represents an interesting case where the animal is ammonotelic in its active life and uricogenic during aestivation². The urea and ammonia levels decrease in digestive gland, foot and body fluids, while uric acid levels increased several fold in the tissues and body fluid in aestivating *P. globosa*^{3,4}. Similar changes were observed in chicken during fasting⁵.

Though the synthesis of glutamine plays an important role in the detoxification of ammonia through the mediation of glutamate dehydrogenase and glutamine synthetase⁶, this aspect of metabolism has received scant attention in molluscs⁷. Earlier reports have shown a gradual decrease in glutamine synthetase activity during the course of aestivation⁸ and during starvation stress. Hence, it is necessary to estimate the levels of glutamine in digestive gland, foot and body fluids of active, starved and aestivated snails and to

see whether it has any bearing in starvation and aestivation metabolism.

Collection, maintenance of snails and mode of aestivation were described elsewhere³. A batch of active snails were kept in water without food for 1 month. Starved animals and snails aestivated for 1 year were used in the present investigation. Digestive gland and foot were isolated in cold and a 10% homogenate of each tissue was prepared in ice-cold distilled water. The homogenates were centrifuged at 3000 rpm for 10 min to remove the cell debris and 0.1 ml of the centrifuged homogenates and body fluid were subjected to glutamine assay, employing the method described by Wilcox⁹, and the liberated ammonia was estimated by Nesslerization.

The glutamine content showed a decrease in the tissue and body fluids of both starved and aestivated snails (table).

Glutamine levels in digestive gland, foot and body fluid of normal, starved and aestivated *Pila globosa*

	Digestive gland			Foot			Body fluid		
	Normal	Starved	Aestivated	Normal	Starved	Aestivated	Normal	Starved	Aestivated
Mean	7.33	5.4	4.26	6.68	6.23	2.64	1.09	1.09	0.33
SD	± 0.37	± 0.54	± 0.65	± 0.33	± 0.26	± 0.65	± 0.02	± 0.39	± 0.04
Difference (%)		-26.3	-41.8		-6.7	-60.4		+0.76	-69.4
p-value		< 0.001	< 0.001		NS	< 0.001		NS	< 0.001

Values expressed in $\mu\text{moles NH}_3/\text{g wet wt}$ and $\mu\text{moles NH}_3/100 \text{ ml body fluid}$. p-value indicates level of significance. NS: Nonsignificant. Values are mean \pm SD of 6 independent observations.

The decrease was relatively more in aestivated snails as compared with the starved ones. The changes during starvation were not statistically significant in the body fluid and foot tissue, while they were significant in the digestive gland.

Using $1,4\text{-}^{14}\text{C}$ succinate, ^{14}C succinate and ^{14}C bicarbonate¹⁰⁻¹² showed glutamine synthesis in some terrestrial snails, *Periwinkles* and *Otala lactea*, respectively. Glutamine was shown to contribute the N-3 and N-9 of purine and uric acid biosynthesis during aestivation^{13,14}. The decrement in the levels of glutamine during aestivation and

starvation in all the tissues studied, except in the body fluid of starved snails, as found in the present study, and increased levels of glutamate¹⁵ indicate the low level of glutamine synthetase activity as well as the enhanced synthesis of uric acid from glutamine. The increase in uric acid accumulation during aestivation and starvation is well in line with the present work^{3,5}. The glutamine, serving as a precursor in the synthesis of uric acid during the torpid state, may be of adaptive value for the snail in its shift from ammonogenesis to uricogenesis during starvation and aestivation.

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Substances fluorescentes dans les téguments à pigment blanc de *Processa robusta* (Crustacé, Natantia)

Fluorescent substances in the white pigment teguments of *Processa robusta* (Crustacean, Natantia)

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Summary. Riboflavin and isoxanthoptérin are identified as possible constituents of the white pigment chromatophores of the shrimp *Processa robusta*. These chromatophores contain 11.76 times more riboflavin than the surrounding tegument.

La crevette *Processa robusta* possède sur la face dorsale de son abdomen une tache blanche constituée de chromatophores contenant des granules de pigment blanc. Chez l'animal, ces granules, présentent au sein du chromatophore des migrations qui suivent un rythme nyctéméral: étalement le jour, concentration la nuit¹; ces migrations peuvent être induites par des stimulus hormonaux^{2,3} ou lumineux, in vivo⁴ comme in vitro⁵. La nature chimique du pigment blanc n'a pu être, jusqu'ici, clairement établie chez les espèces de crustacés où elle a été recherchée: acide urique chez *Eriocheir sinensis*⁶, ptérines chez diverses autres espèces⁷. Comme *P. robusta* est une espèce particulièrement favorable à l'étude de la physiologie du chromatophore, nous avons examiné son pigment blanc du point de vue de sa composition en substances photosensibles du type flavine et ptérine.

Matériel et méthodes. Les animaux (55 individus) proviennent de la région de Banyuls-sur-mer (juin 1977). On prélève sur le vivant la partie pigmentée en blanc (lot B) et, pour comparaison, une portion de surface équivalente (lot T) ne contenant qu'une très faible quantité de pigment blanc (environ 10% de celle contenue dans le lot B). On conserve des exuvies de *P. robusta* (E.P.R.) et celles de *P. edulis* (E.P.E.), espèce très voisine. On congèle le matériel jusqu'aux analyses qui sont faites en lumière réduite, suivant la méthode utilisée par l'un d'entre nous⁸: 3 broyages et macérations dans NH_4OH à 1% (1 h à 27 °C), filtration, acidification du filtrat à pH 3 par de l'acide acétique, adsorption sur colonne de Florisil (\varnothing 12 mm, h = 4 cm), lavage par de l'acide acétique à 1%, élution par de l'acétone à 30%, chromatographie de l'éluat sur du papier Whatman no 1 dans du n-propanol:ammoniaque à 1%