

whereas in other concentrations larger aggregates (2–5 mm) appeared along with a few single cells. The two analogues, 2,4,5-T and 2,4,5-P were not effective in inducing a fine suspension of single cells compared to 2,4-D. Only the larger cell aggregates were observed without any suspension of single cells and small cell clumps.

The hormonal level in the medium was found to influence cell separation in cell suspensions. The 2,4-D analogues, 2,4,5-T and 2,4,5-P were found to be not efficient in inducing well dispersed fine cell suspension, whereas 2,4-D (2 mg/l) induced a fine cell suspension. These preliminary results suggest that the analogues may not be effective substitutes for 2,4-D in the growth and establishment of maize suspension cultures.

In cereals, 2,4-D has been used very frequently for callus induction⁴. In an earlier study, callus initiation and growth of most of the explants were favoured by 2 mg/l, of 2,4-D suggesting that 2,4-D is the choice of auxin required for callus induction in maize⁵. Analogues of 2,4-D have shown a wide range of response in cell and tissue cultures^{6,7}. Superiority of 2,4-D analogues over 2,4-D was also observed in maize embryo cultures suggesting that probably auxin sites have more affinity to analogues than 2,4-D⁸. In the present investigation, 2,4-D analogues were superior to 2,4-D in callus growth and maintenance suggesting differential response of analogues in cultured tissue. It is possible that different auxins have different metabolic breakdowns and hence greater auxin activity of the analogues.

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1. Vasil, I. K., *J. Plant Physiol.*, 1987, **128**, 193.
2. Green, C. E., In: *Basic biology of new developments in bio-technology*, Plenum Press, New York, 1983, p. 195.
3. Linsmaier, E. M. and Skoog, F., *Physiol. Plant.*, 1965, **18**, 100.
4. Yamada, Y., In: *Plant Cell Tissue and Organ Culture*, 1977, p. 144.
5. Rao, K. V., Suprasanna, P. and Reddy, G. M., In: *Gene structure and function in higher plants*, Oxford & IBH Publishers, New Delhi, 1986, p. 245.
6. Green, C. E. and Phillips, R. L., *Crop Sci.*, 1975, **15**, 473.

7. Sekiya, J., Yasuda, T. and Yamada, Y., *Plant Cell Physiol.*, 1977, **18**, 1155.
8. Sanchez de Jimenez, E., Albores, M. and Loyolla-Vargas, V. M., *Ann. Appl. Biol.*, 1981, **98**, 347.

EFFECT OF THYROID HORMONES ON THE ACTIVITIES OF HEPATIC ENZYMES IN THIOURACIL-TREATED TELEOST, *ANABAS TESTUDINEUS* (BLOCH)

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RECENTLY it has been reported that administration of physiological doses of thyroid hormones both T₄ and T₃ significantly influenced the activities of hepatic enzymes of *Anabas testudineus*¹. However, the role of these hormones in thiouracil (antithyroid drug) treated fish has not been investigated. Since hormonal effects on energy metabolism is mainly brought about by altering enzyme activities involved in metabolic pathways, an attempt was made to study the activities of hepatic enzymes such as lactate dehydrogenase (LDH), cytosolic and mitochondrial malate dehydrogenases (cyt. MDH, mit. MDH), glucose 6-phosphate dehydrogenase (G 6-PDH) and mitochondrial catalase of thiouracil-treated *A. testudineus* after the administration of thyroid hormones.

Five groups of fish (comprising six each) each, weighing 50 ± 5g BW were kept in aquaria with water at 28 ± 1°C. Fish were fed with fish feed every other day and were starved for two days prior to sacrifice. Each fish of the groups 2, 4 and 5 was injected i.p with 100 µg thiouracil and group 3 fish received 300 µg thiouracil (Sigma Chemical Co., USA) per fish over a total of 10 days. The fish of group 2 & 3 received hormone vehicle thereafter, while each specimen of group 4 received 5 µg of 1-T₃ and that of group 5 received 5 µg of 1-T₄ for total 5 days. The first group, which received vehicle alone served as the control. The fish were sacrificed after 24 h of last injection and the liver was excised. The chilled liver was weighed and homogenised in 0.25 M sucrose and centrifuged to isolate mitochondria and cytosol at 4°C in Beckman J2 21 centrifuge². The activities of LDH³, cyt. MDH mit. MDH⁴, G 6-PDH⁵ and catalase⁶ were assayed

Table 1 Activity levels of mitochondrial and cytosolic enzymes of *Anabas testudineus* after thiouracil and thyroid hormones administration (values are mean \pm S.E. for 6 fish)

Treatment	LDH (μ mol NADH/ min, mg protein)	Catalase (μ mol H ₂ O ₂ /min/mg protein)	(nmol NADH oxidized/min/mg protein)		
			Cyt. MDH	Mit. MDH	G 6-PDH
Control	2.61 \pm 0.39	136.17 \pm 43.42	866.05 \pm 30.77	184.00 \pm 42.16	58.13 \pm 12.80
100 μ g thiouracil	2.62 \pm 0.22	103.17 \pm 15.16	780.30 \pm 84.23	225.32 \pm 18.77	62.92 \pm 10.15
300 μ g thiouracil	2.53 \pm 0.27	121.73 \pm 24.75	547.21 \pm 97.98**	117.57 \pm 9.05**	51.51 \pm 5.11
100 μ g thiouracil + 5 μ g T ₃	2.19 \pm 0.35	120.59 \pm 15.21	331.86 \pm 44.82***	89.45 \pm 16.43***	34.87 \pm 4.73*
100 μ g thiouracil + 5 μ g T ₄	2.51 \pm 0.29	112.19 \pm 19.28	460.24 \pm 64.07***	102.76 \pm 12.35***	39.19 \pm 4.95*

* $P < 0.01$; ** $P < 0.005$; *** $P < 0.001$.

spectrophotometrically using B&L Spectronic 2000 spectrophotometer at 25°C. Protein was estimated⁷ using BSA as standard. Student's *t* test was used for statistical analysis⁸.

Administration of 100 μ g thiouracil while, produced no change in LDH, cyt. MDH mit. MDH, G 6-PDH and catalase activities, 300 μ g thiouracil produced significant decrease in the activities of cyt. MDH and mit. MDH. However, 300 μ g thiouracil injection had little effect on LDH, G 6-PDH and catalase activities. Administration of T₃ and T₄ to thiouracil-treated fish while, significantly inhibited the activities of cyt. MDH mit. MDH and G 6-PDH, produced no effect on LDH and catalase activities (table 1).

The unchanged activities of LDH and catalase after thiouracil and thyroid hormones administration seem to support the earlier report about the least response of these enzymes to thyroid hormones¹. Lee and Lardy⁹ reported that LDH activity was not altered by thyroid hormone administration or thyroidectomy in rat.

The decreased activity of G 6-PDH, cyt. MDH and mit. MDH after thyroid hormone injection to thiouracil-treated fish, clearly demonstrate the inhibition these enzymes by thyroid hormones. It has been shown in rat that thyroid hormones had little influence on MDH in either cytoplasmic or mitochondrial fractions⁹. The decreased activity of G 6-PDH implies reduced emphasis of HMP shunt and reduced synthesis of fatty acids in the liver of *A. testudineus*. It may be suggested that the decreased MDH and G 6-PDH activities after thyroid hormone injection to thiouracil-treated fish, may be due to the decreased availability of NAD and NADP in the liver as reported earlier¹.

The present study clearly establishes the fact that exogenous administration of thyroid hormones in *A. testudineus* inhibit the anabolic and stimulate or produce no effect on the catabolic enzymes,

regardless of thyroid dysfunction by antithyroid drug administration.

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1. Peter, M. C. S. and Oommen, O. V., *J. Reprod. Biol. Comp. Endocrinol.*, 1987, 7, 53.
2. Johnson, D. and Lardy, H., *Methods Enzymol.*, 1967, 10, 694.
3. Bergmeyer, H. U., *Methods Enzymol.*, 1963, 886.
4. Barrie-Kitto, G., *Methods Enzymol.*, 1969, 13, 106.
5. Glock, G. E. and Mc Lean, P., *J. Biol. Chem.*, 1953, 55, 400.
6. Bergmeyer, H. U., *Methods Enzymol.*, 1974, 574.
7. Gornal, A. G., Bardwill, C. S. and David, M. M., *J. Biol. Chem.*, 1949, 177, 751.
8. Snedecor, G. W. and Cochran, W. G., In: *Statistical methods*, Oxford and IBH Publishing Company, 1967.
9. Lee, Y. L. and Lardy, H. A., *J. Biol. Chem.*, 1965, 340, 1427.

LIPASE ACTIVITY IN AESTIVATED SNAIL, *PILA GLOBOSA*: NEUROENDOCRINE INVOLVEMENT

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AESTIVATION is an enigmatic phenomenon and several aspects of hibernatory and aestivatory torpor in gastropods have been touched briefly by the