

## HOST PARASITE RELATIONSHIPS IN *CHANNA PUNCTATUS* AND *EUCLINOSTOMUM HETEROSTOMUM*. III. TRANSAMINASES AND TOTAL PROTEINS AND FREE AMINO ACIDS

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FREE and protein amino acids have been studied earlier<sup>1</sup> and glycogen, pyruvate and lactate levels and activities of non-specific phosphomonoesterases<sup>2</sup> in liver tissue of *Channa punctatus*, both uninfected and infected, cyst wall and metacercaria of *Euclinostomum heterostomum*. To explore whether nutrients are obtained by worms through cyst wall from host liver and, if so, to candidly assess the parasitic effects on host liver, total free amino acids, total proteins and activities of glutamate-oxaloacetate transaminase (GOT, E.C. No. 2.6.1.1) and glutamate-pyruvate transaminase (GPT, E.C. No. 2.6.1.2) were determined. The present communication presents the result of the study.

Cyst from liver of naturally infected *C. punctatus* were recovered and were washed thoroughly in distilled water and blotted dry. Cyst wall was separated from worms with a pair of needles. Cyst walls thus separated, metacercariae, liver tissue from infected and uninfected *C. punctatus* were washed in double-

distilled water (in normal ice cold saline for enzymes) several times and dried on Whatman paper no. 1.

Total free amino acids were extracted in 10% TCA and estimated (dry wt) colorimetrically by the method of Moore and Stein<sup>3</sup>, as described by Plummer<sup>4</sup>. Total protein (dry wt) was estimated following the method of Lowry *et al*<sup>5</sup>, using bovine serum albumin as standard. Fresh worm homogenate, prepared in ice-cold 0.25 M sucrose, was centrifuged at 3000 rpm at 4°C for 15 min. Supernatants were used for enzyme assay colorimetrically at 545 nm by the method of Reitman and Frankel<sup>6</sup>, as described by Bergmeyer<sup>7</sup>. The protein content in enzyme extracts was estimated by the method of Lowry *et al*<sup>5</sup>. Enzyme activity is expressed in terms of  $\mu\text{g}$  pyruvate formed/mg protein/hr.

A significant decrease in the total free amino acids, a slight increase in the total protein levels, significantly higher GOT and lower GPT activities were observed in infected as compared with the control host liver tissue (table 1). Further, significantly higher levels of protein, GOT and GPT activities were found in cyst wall as compared to metacercaria. Total free amino acids in the cyst wall, however, was much less than in metacercaria (table 1).

Encysted metacercaria, obtaining nourishment from host tissue, through cyst wall, provides an interesting model for study of host parasite relationships. Higher levels of total protein and GOT activity, as observed by the present authors, suggest augmented

**Table 1** Total free amino acids, proteins and transaminases in host liver, cyst wall and metacercaria of *E. heterostomum*. (Mean  $\pm$  standard deviation, figures in parentheses represent the number of samples studied).

	Metacercaria A	Cyst wall B	Uninfected liver C	Infected liver D
Total free amino acids (mg/100 mg)	1.89 $\pm$ 0.48 (8)	1.28 $\pm$ 0.70 (8)	1.10 $\pm$ 0.73 (11)	0.56 $\pm$ 0.18 (8)
Total protein (mg/100 mg)	18.07 $\pm$ 1.45 (13)	38.23 $\pm$ 4.27 (12)	29.99 $\pm$ 5.13 (11)	31.14 $\pm$ 4.09 (11)
GOT ( $\mu\text{g}$ pyruvate/mg protein/hr.)	16.59 $\pm$ 3.06 (7)	72.30 $\pm$ 18.87 (5)	28.63 $\pm$ 3.61 (10)	35.00 $\pm$ 0.75 (5)
GPT ( $\mu\text{g}$ pyruvate/mg protein/hr.)	6.31 $\pm$ 2.35 (8)	20.90 $\pm$ 5.58 (6)	56.97 $\pm$ 9.93 (10)	46.39 $\pm$ 1.24 (5)
<i>Comparison:</i>				
Total free amino acids		—	C:D*	
Total protein		A:B***	—	
GOT		A:B***	C:D***	
GPT		A:B***	C:D**	

\*\*\*P < 0.001    \*\*P < 0.01    \*P < 0.05

protein synthesis and consequent depletion in levels of total free amino acids in infected as compared to healthy liver tissue. Depleted levels of pyruvate and lactate, despite considerable glycogenolysis as observed by Gupta and Agarwal<sup>2</sup> in infected liver, corroborate this.

Total protein levels in the cyst wall, more than twice that of metacercaria, suggest the cyst material to be highly proteinaceous. Activity of GPT, although higher than in metacercaria, is not as prominent in the cyst wall. The most remarkable feature of the cyst wall, however, is the relatively very high GOT activity (more than 4 times that of metacercaria and thrice that of host tissue). This is clearly related to extensive transamination of substrates of energy metabolism into amino acids and, via the aspartate family of amino acids, into pyrimidines, and eventually their incorporation into proteins and nucleic acids in the metacercaria growing within the cyst wall. Relatively high alkaline phosphatase activity in metacercaria<sup>2</sup> suggests active uptake of these by the growing metacercaria.

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## A NOTE ON THE PHYTOCHEMICAL CONSTITUENTS OF SOME BIGNONIACEAE

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THE chemotaxonomy of Bignoniaceae is little known<sup>1</sup>. The present work on the chemotaxonomy of 14 species spread over 10 genera is taken up to fill the gap.

The following taxa have been collected during different periods and in different localities as mentioned in the parenthesis. *Anemopaegma scandens* Mello ex K. Schum (December, 1983, Public Garden, Hyderabad, India), *Bignonia gracilis* Lodd. (November, 1983, Public Garden, Hyderabad), *B. magnifica* Bull (November, 1983, Begumpet, Hyderabad), *B. purpurea* Lodd. ex Sweet. (December, 1983, S. R. Nagar, Hyderabad), *Crescentia cujete* L. (August, 1983, Kakinada, India), *Jacaranda mimosaefolia* D. Don. (November, 1983, Public Garden, Hyderabad), *Kigelia pinnata* (N. A. Jacq.) DC., (December 1983, Public Garden, Hyderabad), *Millingtonia hortensis* L.f. (November, 1983, Begumpet, Hyderabad), *Parmentiera cereifera* Seem. (December, 1983, Osmania University Campus, Hyderabad) *Pyrostegia venusta* (Ker-Gawl) Miers. (October, 1983, Public Garden, Hyderabad), *Spathodea companulata* Beauv. (October, 1983, Nizam College, Hyderabad), *Tecoma capensis* Lindl. (November, 1983, Begumpet, Hyderabad), *T. smithii* × Hort. ex Garten. (October, 1983, Public Garden, Hyderabad) and *T. stans* (L.) H. B. & K. (November 1983, Nizam College, Hyderabad). Standard tests with the fresh leaves and stems as well as 80% ethanolic extracts of the above taxa have been carried out to screen the presence of various phytochemical constituents.

Uniformly negative results are obtained for alkaloids, anthraquinones, cyanogenic glycosides (HCN test), Juglone (Juglone test A) and lignans and uniformly positive results for free phenols and syringyl radicals (Maule test). Notwithstanding the similarities in the above chemical characters, the taxa in the present study seem to be distinct in the possession of some chemical constituents, with restricted distribution. Thus, aucubin compounds (Ehrlich test) are found to be present in *Anemopaegma*, *B. gracilis*, *B. magnifica*, *Kigelia*, *Pyrostegia*, *Spathodea*, *T. capensis*,