rock, than to the Nuggihalli Belt peridotite which is unaltered. Among the oxide ratios, the value of Al_2O_3/TiO_2, and the metal ratio of $Mg/Mg+Fe^{2+}$ are remarkably closer among the three peridotites.

The occurrence of peridotite among the crustal rocks has more than a casual significance in regional tectonics and petrogenesis since it is referred to as Upper Mantle rock. In the Khammam Belt, peridotite occurs amidst the high grade supracrustal rocks which were formed in partly ensialic basins. The peridotite of Khammam is derived from differentiation of gabbroic magma of tholeiitic parentage which itself is derived by partial fusion of the Upper Mantle.

DSP thanks CSIR for the Fellowship and VSVPR thanks A.P.S. Ground Water Department for encouragement.

4 July 1982; Revised 8 November 1982

1 Subbaraju, M., *Indian Min.*, 1976, **16**, 35.
2. Ramamohana Rao, T. and Shah, Jatin, Kumar., *Sci. Cult.*, 1981, **47**, 184.
3. Swaminath, J. and Ramakrishna, M., *Memoir G.S.I.*, 1981, 112.
4. Windley, B. F., *The Evolving Continents*, 1977.
5. Winkler, H. G. F., *Petrogenesis of metamorphic rocks.*, 1976.
6. Hussain, S. M., Naqvi, S. M. and Gnaneshwar Rao, T., *J. Geol. Soc. India.*, 1982, **23**, 19.
7. Naqvi, S. M., Viswanathan, S. and Viswanatha, M. N., *Archaean Geochem.*, 1976.
8. Divakara Rao, V., Satyanarayana, K., Naqvi, S. M. and Hussain, S. M., *Lithos*, 1975, **8**, 77.
9. Wyllie, P. J., *Ultramafic and related rocks.*, 1967.

HOST PARASITE RELATIONSHIPS IN *CHANNA PUNCTATUS* AND *EUCLINOSTOMUM HETEROSTOMUM-II*

A. K. GUPTA AND S. M. AGARWAL
Department of Bioscience, Ravishankar University, Raipur 492 010, India.

ENCYSTED metacercariae of *Euclinostomum heterostomum*, occurring in liver of *Channa punctatus*, obtain nourishment from host liver through cyst wall. Studies, thus far, on host parasite interactions, include only a preliminary report on electrophoretic analysis[1] and free and protein amino acids in liver of *C. punctatus* (both uninfected and infected), cyst wall and metacercariae of *E. heterostomum*[2]. The present work includes an account of glycogen, lactate, pyruvate levels and activity of non-specific phosphomonoesterases in liver (both uninfected and infected) of *C. punctatus*, cyst wall and metacercaria of *E. heterostomum*.

Cyst of *E. heterostomum* were recovered from liver of *C. punctatus*, washed thoroughly with distilled water dried on Whatman paper and teased. Cyst wall, metacercariae, uninfected and infected liver were washed in distilled water several times (in ice cold normal saline for enzymes). While, glycogen was estimated on dry weight basis (tissue dried at 80° C in an oven until constant weight) by the method of Seifter *et al*[3], fresh weights were taken for studying pyruvic acid, lactic acid and non-specific phosphomonoesterases. Known amounts of fresh tissue were homogenised in 10% Trichloro-acetic acid and centrifuged at 3000 rpm. Supernatant was used for estimation of pyruvic and lactic acids respectively by the methods of Friedeman and Haugan and Barker and Summerson as described by Oser[4]. Homogenates (10% w/v), prepared in ice cold normal saline, were centrifuged at 3000 rpm at 4° C for 15 min. Supernatant was used for enzyme assay.

Non-specific phosphomonoesterase activity was determined colorimetrically at 660 nm with appropriate blank of buffered sodium β-Glycerophosphate substrate, as suggested by Hawk *et al*[5], and the liberated inorganic phosphorus was estimated by the method of Fiske and Subbarao[6]. Protein concentration in homogenate was estimated by Lowry's[7] method using bovine serum albumin as standard. Enzyme reaction was stopped with 30% Trichloroacetic acid after 60 min and phosphorus was estimated in protein free supernatant at room temparature. Specific enzyme activity was expressed in terms of μg inorganic phosphorus/mg protein/hr. Effect of pH on enzyme activity was studied using buffered (sodium diethyl barbitone) sodium β-Glycerophosphate as

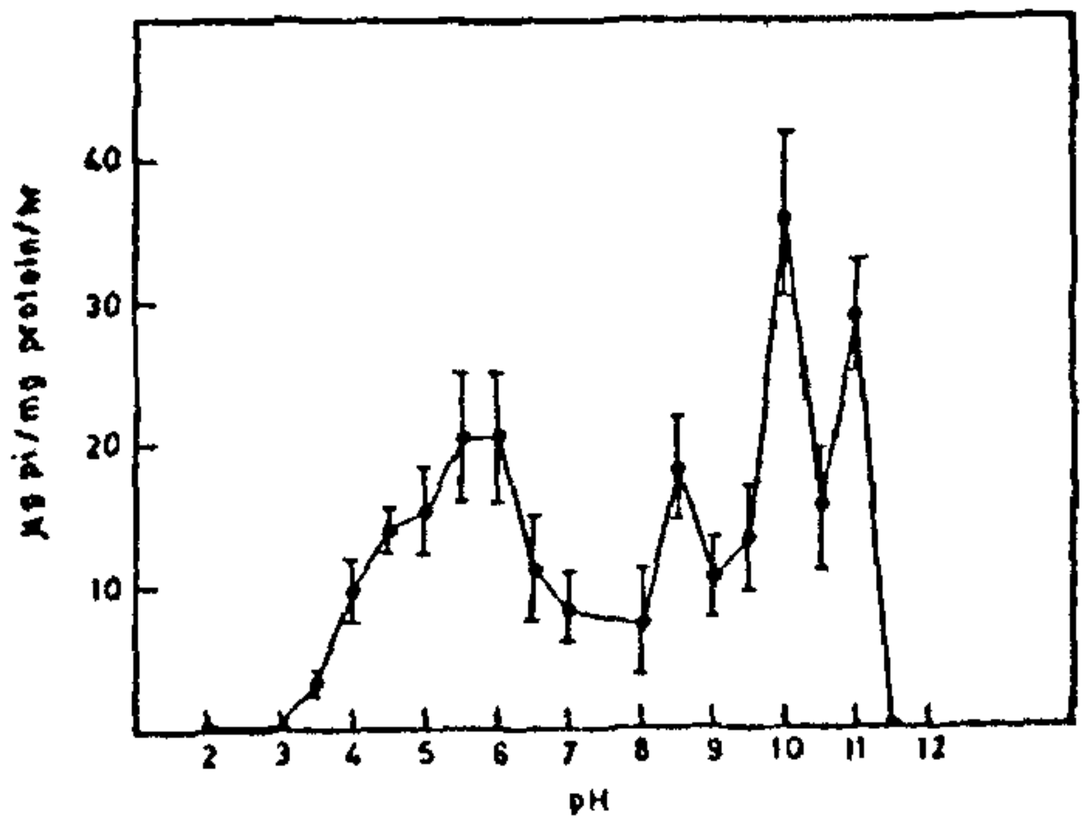

Figure 1. Effect of pH on acid and alkaline phosphatase activity of *E. heterostomum* (Metacercariae).

TABLE I

Glycogen, Pyruvate, Lactate and Phosphomonoesterases in metacercaria and cyst wall of E. heterostomum and uninfected and infected liver of C. punctatus.

		Metacercariae	Cyst wall	Uninfected Liver	Infected Liver
I	*Glycogen* (% on dry weight basis)				
	Range	17.87–27.03	15.90–18.02	4.24–13.30	0.80–3.88
	Mean	22.36	17.02	8.99	1.84
	S. D.	2.07	0.82	2.65	1.05
II	*Pyruvic Acid* (μg/100mg tissue)				
	Range	13.57–24.00	13.57–23.04	8.96–16.25	5.10–11.10
	Mean	17.10	19.77	11.64	6.56
	S. D.	4.39	3.88	2.74	2.25
III	*Lactic Acid* (μg/100mg tissue)				
	Range	13.41–27.27	6.25–12.50	41.90–154.16	14.28–40.00
	Mean	19.02	8.62	84.98	22.89
	S. D.	4.40	1.49	41.83	7.81
IV	*Phosphomonoesterases* (μg pi/mg protein/hr.)				
i	*Acid Phosphatase* (pH 5.0)				
	Range	2.90–9.68	139.79–166.67	70.84–94.08,	85.99–112.68
	Mean	8.38	156.36	85.30	102.01
	S. D.	2.11	11.58	6.44	8.29
ii	*Alkaline phosphatase* (pH 10.0)				
	Range	9.68–15.48	5.38–10.75	0.58–1.16	1.18–2.90
	Mean	11.83	8.95	0.90	1.74
	S. D.	2.15	2.77	0.30	0.57

S. D. = Standard Deviation.

substrate. Different pH levels, from 2.0 to 12.0, were adjusted using dilute acid and alkali. Relative activities of acid and alkaline phosphatases were carried out at pH 5.0 and 10.0 respectively.

Data obtained as percentage glycogen, pyruvate and lactate in μg/100mg tissue and non-specific phosphomonoesterases in μg pi/mg protein/hr. in metacercariae, cyst wall, infected and uninfected liver are included in table 1. Percentage glycogen is found relatively high in metacercariae, and very low in infected liver. Acid phosphatase activity is found to be very high (as compared from alkaline phosphatase) in the cyst wall; it is higher in infected liver than in uninfected liver and very low in metacercariae. Alkaline phosphatase activity, however, is higher than acid phosphatase activity in the latter. Effect of pH on enzyme activity of metacercariae is shown in figure 1. Maximum activity of acid phosphatase is observed at pH 5.5 to 6.0, whereas alkaline phosphatase is found to have three peaks of activities, respectively at pH 8.5, 10.0 and 11.0.

Depletion in glycogen, pyruvate and lactate levels and higher phosphomonoesterase (both acid and alkaline phosphatase) activity in *C. punctatus*, infected with cysts of *E. heterostomum*, obviously is the result of stress, since the worms obtain nourishment through cyst wall from the host liver. The reserve food in both cyst wall and metacercaria is glycogen. Higher levels of pyruvate in both (as compared from host liver) suggest anaerobic metabolism. Acid phosphatase activity is highest in cyst wall suggesting predominantly degradative metabolism. Higher alkaline phosphatase activity as compared with the acid phosphatase activity in metacercaria is further suggestive of a much greater need of active uptake by the worms. This is the first report of higher alkaline phosphatase activity in a trematode worm. Few other workers[8-17] have also reported higher acid phosphatase activity in the adult trematodes. Occurrence of three peaks of alkaline phosphatase activity (figure 1), respectively at pH 8.5, 10.0 and 11.0, is suggestive that more than one alkaline phosphatase enzyme is involved in the energy metabolism of metacercariae of *E. heterostomum*[18].

Grateful thanks are extended to UGC for sanctioning

a scheme on "Survey and Pathophysiology of fishes parasitized by helminths" to SMA and junior research fellowship to AKG.

14 December 1982; Revised 3 August 1983.

1. Bhagavathiammai, A. and Ramalingam, K., *Third Natl. Congr. Parasit.*, Hissar, 1980.
2. Gupta, A. K. and Agarwal, S. M., *Curr. Sci.*, 1983, **52**, 474.
3. Seifter, S., Dayton, S., Novic, B. and Muntwyler, E., *Arch. Biochem.*, 1950, **25**, 191.
4. Oser, B. L., *Hawk's Physiol. Chem.*, 14th ed., Tata McGraw Co., New Delhi, 1965.
5. Hawk, P. W., Oser, B. L. and Summerson, W. H., *Practical Physiol. Chem.*, McGraw Hill Book Company, New York, 1954, p. 1118.
6. Fiske, C. H. and Subbarao, Y., *J. Biol. Chem.*, 1925, **66**, 375.
7. Lowry, O. H., Roserbrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265.
8. Pennoit-De Cooman, E. and van Grembergen, G., *Plathelminthen.*, 1942, **4**, 7.
9. Nimmo-Smith, R. H. and Standon, O. D., *Exp. Parasitol.*, 1963, **13**, 305.
10. Goil, M. M., *Parasitol.*, 1966, **56**,105.
11. Halton, D. W., *J. Parasitol.*, 1967, **53**, 46.
12. Probert, A. J. and Lwin, T., *Exp. Parasitol.*, 1974, **35**, 253.
13. Srivastava, M. and Gupta, S. P., *Zool. Anz.*, 1975, **195**, 35.
14. Tandon, R. S. and Mishra, K. C., *Indian J. Parasitol.*, 1978, **2**, 145.
15. Sathyanarayana, M. C. and Anantharaman, S., *Proc. Indian Acad. Sci.*, 1979, **B88**, 115.
16. Hora, C. and Sharma, P. N., *Proc. Indian Acad. Parasitol.*, 1980, **1**, 125.
17. Gupta, A. K., Gaur, A. S., Agarwal, S. M., *Jpn. J. Parasit.*, (in press).
18. Nizami, W. A., Siddiqui, A. H., Yusufi, N. K., *J. Helminth.*, 1975, **49**, 281.

IMPROVING SYMBIOTIC NITROGEN FIXATION AND PRODUCTIVITY IN BLACK GRAM (*VIGNA MUNGO* L.)

P. C. PARICHA AND M. KAR
Department of Plant Physiology,
Orissa University of Agriculture and Technology,
Bhubaneshwar 751003, India.

RESPONSES to nitrogen fertilization in tropical and other legumes have been confirmed in several studies[1-3]. With the escalation in the cost of fertilizers such a finding has very little relevance to practical farming. The alternative approach is to increase the efficiency of nitrogen fixation and assimilation in legumes. It is well known that iron and molybdenum are constituents of the enzyme nitrogenase[4] whereas the availability of iron is plentiful, molybdenum is limited to acid soils[5]. Liming and molybdenum fertilization in such soils are practiced to ameliorate the soil conditions.

The present study was undertaken as a field experiment in 1980-1981 using acid laterite soils of pH 5.4 to know the response of Mo seed treatment and to relate the benefits in terms of applied nitrogen to the soils. The plots were 16 m^2 replicated four times in an experimental design of randomized block.

The soil was sterilized with formaldehyde (40% w/v) in the proportion of 1:50 parts of irrigation water and seeds of blackgram were inoculated with appropriate *Rhizobium* culture. Molybdenum was applied at the rate of 3 g/kg of seed. Nitrogen was added to the soil in the form of urea at rates of 0, 15, 30, 45 kg/ha with *P* and *K* uniformly applied at rates of 80 and 50 kg/ha respectively.

Total nitrogen was estimated following the standard Kjeldahl method[6] and total chlorophyll according to the method of Arnon[7]. Control plants were less vigorous and exhibited premature yellowing of the lower leaves, typical as those of nitrogen deficiency. The plants attained lush green colour due to *N* fertilization or Mo seed treatment which is evident from the chlorophyll contents of the leaves (table 1).

The nodulation decreased considerably in the uninoculated series but could not be completely eliminated as is evident from sparse nodulation in the control plants. Evidences in literature indicate both promotion[8] and inhibition[9] of nodulation due to starter dose of *N* fertilization. In the present study, low levels of nitrogen fertilization consistently resulted in an increase in the nodule number, possibly, because of low *N* status (0.06%) of the soils. The nodule number increased significantly due to Mo seed treatment. Similar increases in nodulation due to Mo were also reported in pea[10].

Apart from nodule number, the dry weight of the nodules was higher in the inoculated series (62.0 mg) as compared to the uninoculated plants (13.2 mg) Mo seed treatment resulted in further increases in dry weight of the nodules[11]. The inhibitory effects of applied *N* were observed at or above 30 kg/ha on both nodule number and dry weight of nodules.

The inoculated plants contained higher concentration of *N* as compared to the uninoculated plants. Nitrogen concentrations increased progressively as a response to added nitrogen. Stimulation in *N* concentration due to Mo alone was equivalent to the highest