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ROLE OF THIAMINE HYDROCHLORIDE AS A HATCHING FACTOR IN LARVAL EMERGENCE FROM CYSTS OF *HETERODERA ORYZICOLA* (NEMATODA: HETERODERIDAE) ON RICE

THIAMINE hydrochloride (3 mM), undiluted rice root diffusate and flavionic acid (3 mM) stimulated the hatching of larvae from cysts of *Heterodera oryzicola* n.sp. While the chain length between the two terminal polarisable atoms of flavionic acid was the hatching factor, the effectiveness of thiamine hydrochloride with only one polarisable atom was through the enzyme system. Seed coat and roots of rice are rich in thiamine which along with the root diffusates stimulate hatching continuously in soils with monoculture of rice.

Root diffusates of plants^{1,2}, certain dyes³ and metallic ions like zinc^{4,5} stimulate the emergence of larvae of *Heterodera schachtii*. Organic compounds, viz., anhydrotetrone acid⁶, picrolonic acid⁷ against *Globodera rostochiensis*, flavionic acid^{8,9} against several *Heterodera* spp. and nematicides like nabam (sodium ethylene bis dithiocarbamate)¹⁰ against *H. schachtii* at concentrations of 0.6 to 3 mM proved effective as hatching agents. Some of these along with some constituents of vitamin B and undiluted diffusates from roots of rice c.v. CRM13-3241 (NSJ 200 × Padma) were tested against the cysts of *H. oryzicola* n.sp.¹¹ by the methods described earlier^{4,5,10}.

Hatching was significantly high in thiamine hydrochloride (vitamin B₁) followed by rice root diffusate and flavionic acid (Table I). Pyridoxine, folic acid and sodium metavanadate were moderately active. The other vitamin constituents, dyes and zinc salts were inactive. Larvae emerging from thiamine hydrochloride treatment were active and infective like those emerging from the rice root diffusate. Hatching agents stimulated the coiled larva inside the egg and

the emerged larva was active upto 72 h until penetration into rice roots suggesting that it has the energy reserves for motility but lacked stimulus for breaking the dormancy while still in egg.

TABLE I

Emergence of larvae from cysts of Heterodera oryzicola n.sp. soaked in test compounds at conc. 3 mM (mean cumulative hatch of 3 batches of 50 cysts each during 21 days)

Compound	Hatch rating*
Thiamine hydrochloride	123
Pyridoxine	62
Folic acid	40
Riboflavin	2
Calcium pantothenate	1
Nicotinic acid	1
Rice root diffusate (undiluted)	100
Flavionic acid	92
Picrolonic acid	1
Sodium metavanadate	33
Zinc chloride	5
Zinc sulphate	2
Auramine	2

* Based on Clarke and Shepherd⁶.

The role of thiamine hydrochloride as a co-enzyme with pyruvic acid dehydrogenase in the further metabolism of the energy rice pyruvic acid in animals has been established^{12,13}. The effectiveness of thiamine as a hatching factor in *H. oryzicola* n. sp suggests that the dormancy had been effected by blocking the intermediary metabolism. Unlike in the other cyst nematodes responding to flavionic acid⁸ where the chain length between the two terminal polarisable atoms was the factor influencing hatching, thiamine hydrochloride with only one polarisable atom was also found to be efficient as a hatching stimulant. Though the role of flavionic acid in influencing the emergence of larvae from cysts through the enzyme system has not yet been established, the effectiveness of thiamine is considered to be through the enzyme system.

Most enzymes of Glycolytic and TCA cycles in nematodes have been demonstrated¹⁴ and homogenates of the nematode *Ditylenchus trifurmis* were able to utilise labelled glucose and pyruvate and convert them into CO₂¹⁵. Seed coat and roots of rice are rich in thiamine and the present study confirms that the root diffusates and these may effect hatching of cysts of *H. oryzicola* n. sp through thiamine via the enzyme system. Due to the above reason, hatching was

continuous and larvae were prevalent round the year in soils with monoculture of rice.

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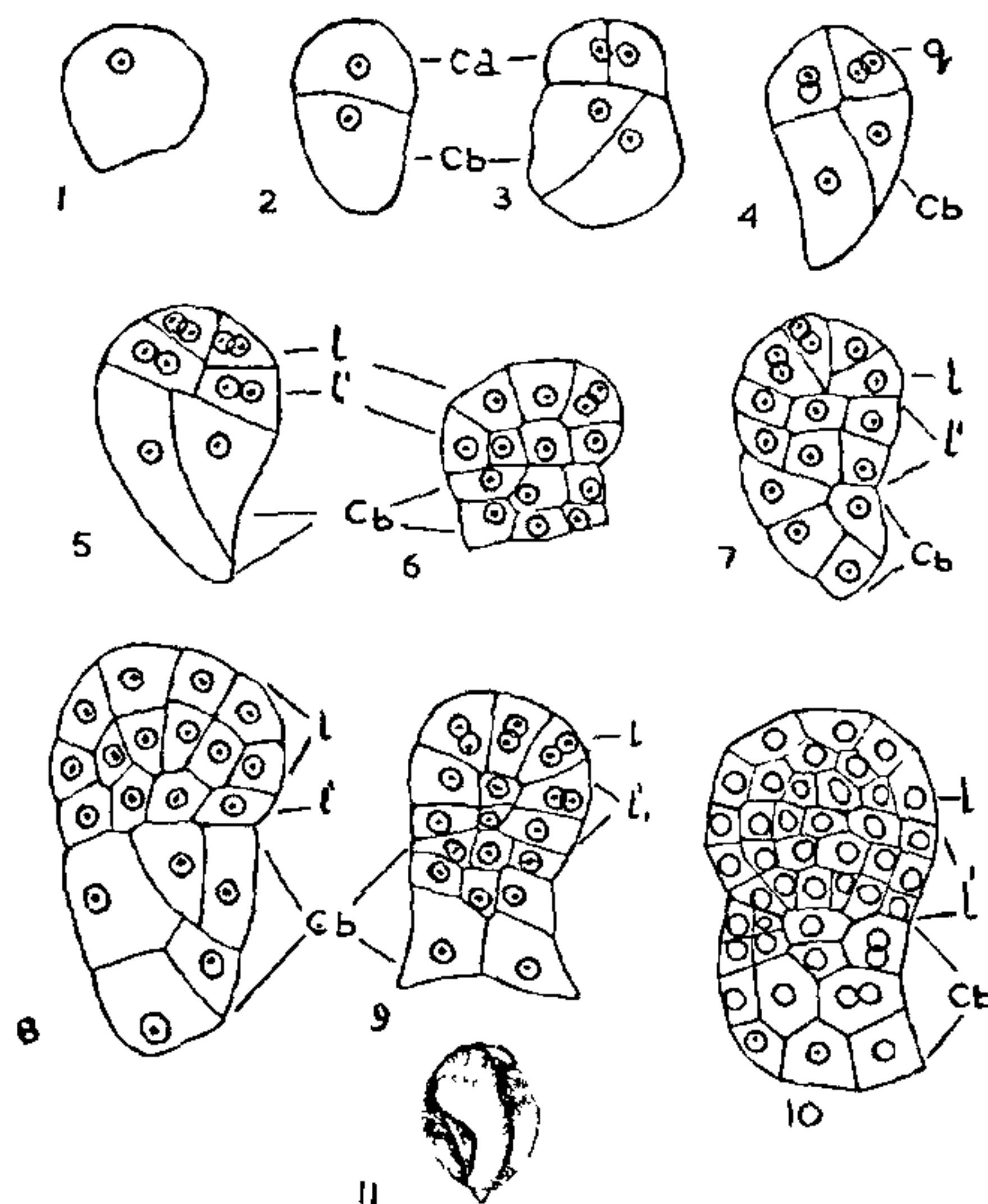
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EMBRYOGENY IN *GYROCARPUS AMERICANUS* JACQ.

THE embryo development in *Gyrocarpus* remains uninvestigated; hence a study was undertaken and reported here.

The fertilised egg enlarges and divides by a transverse wall giving rise to two superposed cells *ca* and *cb* (Figs. 1, 2). The cell *ca* divides vertically and *cb* by an oblique wall resulting in a tetrad of the A₁ category of Soueges (*vide* Crété¹) and the two juxtaposed cells of *ca* undergo another vertical division forming the quadrant *q* (Fig. 4). The cells of the tier *q* divide transversely forming the octant disposed in two tiers *l* and *l'* (Fig. 5). In some preparations the division in *cb* preceded that in *ca* and in others it is slightly belated. Further divisions in the tier *l*, although irregular, give rise to the stem apex and the two cotyledons, while those of the *l'* give rise to the hypocotyledonary part and hypophysis (Figs. 6-10). The cells of the tier *cb* divide rather irregularly forming a massive suspensor whose cells are highly vacuolated. The development of the embryo is referable to the

First Period, Series A, Megarchetype VI. In the mature embryo, the large cotyledons are convolute and enclose the plumule (Fig. 11), which shows the first leaf primordia.



FIGS. 1-11. *Gyrocarpus americanus* Jacq. Figs. 1-10 Stages in the development of embryo $\times 160$; Fig. 11, mature embryo showing convolute cotyledons \times nat. size.

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TWIN-POLLINIA IN *CEROPEGIA* LINN. (ASCLEPIADACEAE)

THE nature and importance of twin-pollinia of Asclepiadaceae was not mentioned in the works of earlier authors like Erdtman^{1,2} and Galil and Zoroni³. It was El-Gazzar and Hamza⁴ who utilized the palynological data for the classification of members of Asclepiadaceae. The present study was undertaken to evaluate the twin-pollinia data, in differentiating two species and two varieties of *Ceropegia* which are considered separate by some authors and merged by others.