

these two sub-families to family level, as *Gentianaceae sensu lato* and *Menyanthaceae*; as already practised by Engler<sup>3</sup> and Hutchinson<sup>8</sup>. Other chemical evidences listed by Gibbs<sup>4</sup> (hot-water test, cyanogenetic glycosides and some other data on saponins) and also the abundance of L-(+)-bornesitol in *Gentianoideae*<sup>13</sup> lend further support to this view.

In the *Gentianaceae sensu stricto*, flavone-*o*-glycosides are restricted to the tribe *Exacineae* (Type genus *Exacum* is screened here). The xanthenes and glycoflavones so abundant in the rest of the *Gentianaceae* are conspicuously absent in the *Exacineae*. L-(+)-bornesitol which is very common in the *Gentianaceae* also eludes this tribe<sup>13</sup>. The bilocular ovary of the tribe, in the otherwise unilocular *Gentianaceae*, is a characteristic, morphological feature.

The available morphological and chemical data, therefore, evoke a few pertinent queries.

- (1) Do all these formidable evidences suggest raising of the tribe *Exacineae* to a family or a sub-family level?
- (2) If the morphological and chemical differences of the same magnitude could warrant a family status for *Menyanthaceae*, would it not be in the fitness of things to segregate the tribe *Exacineae* and elevate it to the family category?

Answers to these queries are apparently in the affirmative. The tribe *Exacineae*, in our

opinion should be raised to family *Exacaceae*. However, chemical data on the remaining genera of *Exacineae* in particular and the *Gentianaceae* in general, are necessary to draw valid conclusions based on sound taxonomic judgement.

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## INFLUENCE OF SEX ON HEPATOPANCREATIC GLYCOGENOLYSIS OF SCORPION *HETEROMETRUS FULVIPES* (C. KOCH)

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### ABSTRACT

The levels of haemolymphatic glucose, hepatopancreatic glycogen were shown to differ between male and female scorpions. The hepatopancreatic phosphorylase 'a' and 'ab' activity levels were higher in males than in the females. The higher levels of phosphorylase activity in males have been correlated with higher glycogenolysis and haemolymphatic glucose. The sex-based differences in glycogenolysis have been discussed.

### INTRODUCTION

THE sex of the animals has been indicated to play a vital role in various physiological activities<sup>1-4</sup>. The differences in morphological features and the tissue somatic indices have been

clearly established between the two sexes of different animals<sup>1-5</sup>. There seem to be metabolic differences at the enzymatic levels between male and female scorpions<sup>6</sup>. In comparison to other groups of animals, there is less work on the scorpions in relation to sex. Hence an attempt has been made to study the sex-based differences in glycogenolysis to understand the probable cause

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for the difference in the activity levels of the two sexes of the scorpions.

#### MATERIAL AND METHODS

The commonly available South Indian scorpions, *Heterometrus fulvipes* (C. Koch) were collected around Tirupati and brought to the laboratory and were maintained in large moist vivaria. They were fed with cockroaches regularly *ad libitum*. The animals were starved for 24 h preceding the estimations. For all experimental purposes similar sized scorpions of the weight range of 5-6 g were used in the present investigation. All investigations were carried out at 20.00 h, since metabolites<sup>7</sup> and enzymes<sup>8,9</sup> vary in rhythmic manner in the 24 h of the day. The haemolymph from both the sexes was drawn separately with a hypodermic syringe through the arthroal membrane of the chelate leg. The hepatopancreas was isolated into scorpion Ringer<sup>10</sup> and kept for 5 min for recovery. The levels of haemolymphatic glucose and hepatopancreatic glycogen were assayed using the methods of Mendel, Kemp and Myers<sup>11</sup> and Kemp and Keto Van Heijningen<sup>12</sup> respectively. The activities of phosphorylase 'a' and 'ab' were estimated in the direction of glycogen synthesis<sup>13</sup>.

Homogenate (5% W/V) was prepared in aqueous medium containing 0.037 M ethylene diamine tetraacetic acid (EDTA), pH 6.5 and 0.1 M sodium fluoride, pH 6.5 as recommended by Guillory and Mommaerts<sup>14</sup>. The homogenate was centrifuged for 15 min at 2,500 rpm and the supernatant was diluted four times with cysteine (0.03 M),  $\beta$ -glycerophosphate (0.015 M) buffer, pH 6.5. The diluted enzyme (0.4 ml) was added to 0.2 ml of 2% glycogen and incubated for 20 min at 35° C. The reaction was started by the addition of 0.2 ml of 0.016 M glucose-1-phosphate (G-I-P) to one tube (phosphorylase 'a'), 0.2 ml of G-I-P and 0.004 M adenosine-5-monophosphate to the other (phosphorylase 'ab'). After incubation for 15 min for phosphorylase 'ab' (Total) and 30 min for phosphorylase 'a' (active) activities, the reaction was stopped by the addition of 10% sulphuric acid. The inorganic phosphate (Pi) liberated was estimated by the method of Taussaky and Shorr<sup>15</sup> and protein by the method of Lowry *et al.*<sup>16</sup>.

#### RESULTS AND DISCUSSION

The results presented in Table I indicate the sex-based differences in the levels of metabolites and enzyme activities. The level of

glucose in haemolymph of males was found to be at a higher level than that of the females, indicating the possible induction of activity difference in the two sexes. The observation is in consonance with the earlier findings indicating high rate of oxygen consumption in males in comparison with the females<sup>2,3</sup>. Since the glucose of haemolymph is known to be derived from the hepatopancreatic glycogen, the levels of glycogen in hepatopancreas of males and females were analysed.

TABLE I

The levels of haemolymph glucose (mg glucose/100 ml of blood), glycogen (mg glycogen/g wet weight), phosphorylase 'a' and 'ab' ( $\mu$  moles of pi/mg protein/hr) in hepatopancreas of scorpion, *Heterometrus fulvipes*.

All values are mean  $\pm$  S.D. of 10 observations.

+ or - indicates increase or decrease respectively.

Component	Sex of the animal		% Change male over female
	Female	Male	
Haemolymph glucose	19.92 $\pm 1.08$	25.02 $\pm 1.51$	+25.60 P < 0.001
Glycogen	14.24 $\pm 1.45$	11.00 $\pm 1.01$	-22.76 P < 0.001
Phosphorylase 'a'	8.00 $\pm 0.33$	11.03 $\pm 1.67$	+37.87 P < 0.001
Phosphorylase 'ab'	27.33 $\pm 1.21$	33.26 $\pm 1.78$	+21.73 P < 0.001
% of 'a' from 'ab'	29.62	33.42	+12.83

It was found that males had 22.8% less glycogen in their hepatopancreas than the females, indicating the possible depletion of glycogen reserve in male to maintain high glucose level in haemolymph. Hence it can be suggested that the reported higher metabolic rate in males induces higher glycogenolytic activity, which is regulated by phosphorylase. Hence phosphorylase 'a' and 'ab' activity levels are determined in the hepatopancreas of male and female scorpions. The active phosphorylase ('a') and total phosphorylase ('ab') are found to be higher in males over the females. The level of the active form among the total phosphorylase (a/ab%) was also greater in males than in the females. These observations indicate the occurrence of higher rate of phosphorylase activity in male hepatopancreas which in turn might be responsible for higher glycogenolytic activity resulting in the depletion of glycogen. Since the

hepatopancreatic glycogen was the source for the glucose of haemolymph, higher rate of glycogenolysis might have been responsible for the higher glucose level of haemolymph in males over the females. The present investigation clearly suggested the occurrence of variations in the rate of glycogenolysis of the hepatopancreas of male and female scorpions, while the female is tending towards decreased glycogenolysis for the preservation of glycogen, the male is tending towards higher glycogenolysis. This differential rate of glycogenolysis might be responsible for the differences in haemolymphatic glucose and metabolic rate between male and female scorpions. Hence the hepatopancreatic glycogen degradation seemed to be under the influence of the sex of the animal.

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