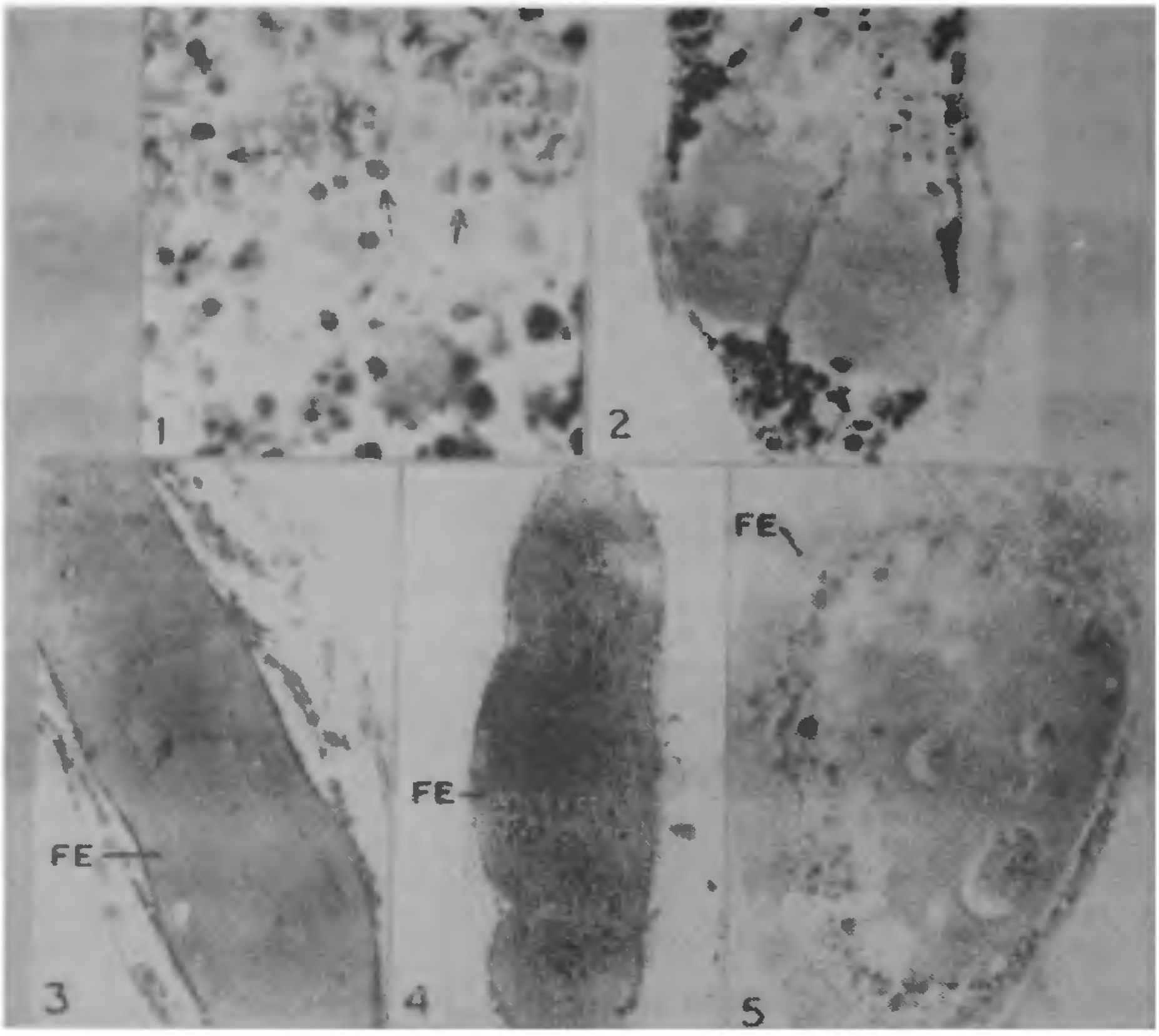

Figures 1–5. 1. A part of the germarium showing completely (arrows) and incompletely fused oogonia (broken arrows) × 675. **2.** Fusion between hypertrophied oogonia lacking follicular epithelium × 315. **3.** A compound follicle comprising a hypertrophied oogonium (arrow) and oocytes enclosed by a common follicular epithelium (FE) × 315. **4, 5.** Compound follicles with separate identities (figure 4) and without separate identities of the fused oocytes (figure 5) × 315. (All sections have been cut at 7.5 μ m and stained in Heidenhain's haematoxylin and eosin).

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EFFECTS OF SCORPION (*HETEROMETRUS FULVIPES*) VENOM ON ITS CARDIAC ACTIVITY

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ALTHOUGH some information is available on the effects of scorpion venom on the physiology of several animals, studies on autoenvenomation are scarce. In the present study the *in vitro* and *in vivo* effects of scorpion venom on its own cardiac activity are presented.

Venom from the scorpion *Heterometrus fulvipes* (C. Koch) was collected by gently pressing the telson, preserved in refrigerator (−9° C), and used within 36 hr. Part of the collected venom was dialysed in a cellulose acetate dialysis tube against distilled water for 24 hr at 5° C. Another part of the crude venom was exposed in a glass tube to boiling water for 10 min to coagulate the protein, then centrifuged at 1000 g for 10 min and the supernatant stored in the refrigerator (−9° C).

In vitro heart rates of the scorpion were determined separately at 5 min intervals upto 1 hr after the administration of the three samples of venom mentioned above. LD_{50} was determined as given by Carpenter[1] and 1/3 of LD_{50} was taken as sublethal dose. For *in vivo* studies sublethal doses of the three venom samples were injected separately into the animal and the rate of heart-beat was determined at 1, 3, 6, 12, 24 and 48 hr following the administration of venom. The results were compared against the control rates from normal animals at these timings to waive off the differences due to diurnal rhythmicity.

Undiluted crude, dialysed and heat-denatured venom caused total cessation of the heart-beat *in vitro* and *in vivo*. With dilution of the venom to 50% (table 1) the heart-rates were found to decrease from control. Maximum decrease *in vivo* was observed at 12 hr after envenomation (table 2). Dialysed venom reduced the heart-rate maximally, followed by crude and heat-denatured venoms in that order.

TABLE 1

Rate of heart-beat in the scorpion (H. fulvipes) after in vitro auto-envenomation with crude, dialysed and heat-denatured venoms

Venom sample	Control heart-rates	Heart-rates 1 hr after envenomation
Crude	75.0 ± 2.4	21.2 ± 2.0 $P < 0.001$
Dialysed	75.0 ± 2.4	19.9 ± 3.2 $P < 0.001$
Heat-denatured	75.0 ± 2.4	33.8 ± 1.5 $P < 0.001$

Each value is the mean of observations from 6 scorpions ± standard deviation, followed by 't' test.

TABLE 2

Rate of heart-beat (beats/min) in the scorpion H. fulvipes with reference to time course following auto-envenomation by injection of crude, dialysed and heat-denatured venom

Venom sample	Control heart-rates	Heart-rates 12 hr after envenomation	Heart-rates 48 hr after envenomation
Crude	75.2 ± 2.0	59.0 ± 1.3 $P<0.001$	75.0 ± 2.2 NS
Dialysed	75.0 ± 2.0	54.9 ± 1.7 $P<0.001$	75.1 ± 2.0 NS
Heat-denatured	75.2 ±2.0	61.1 ± 2.0 $P<0.001$	75.0 ± 2.0 NS

Each value is the mean of observations from 6 scorpions ± standard deviation, followed by 't' test.

NS: Not significant.

The toxic fractions of the venom have been reported to be proteinaceous in nature[2,3]. Though the protein content of crude and dialysed venoms was similar, greater reduction in heart-rate with dialysed venom indicates that the putative substances, which might be acting as a 'check' on the toxicity in the crude venom have escaped out during dialysis. These substances could be amino acids and other ions of low molecular weight.

Minimal reduction of the heart-rate by heat-denatured venom in the present study demonstrates that during heat-denaturation most of the inhibitory principles (proteins) have been eliminated. However, persistence of some amount of toxicity in the heat-denatured venom suggests that all the toxic principles have not been eliminated on heat-denaturation, and that some are still present. These could be again proteins which are heat-resistant . The presence of heat-stable proteins in the scorpion venom is known[4]. Thus it may be concluded here that some of the toxic proteins in *H. fulvipes* venom are heat-resistant.

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OCCURENCE OF TWO POLYSTYRENES IN PROSOPIS JULIFLORA SEEDS

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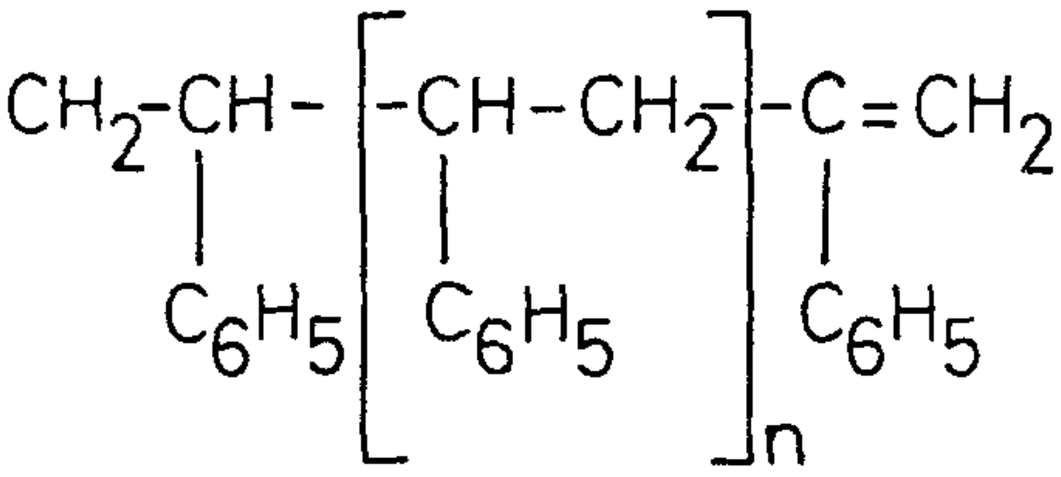
POLYSTYRENES are ragarded as a homologous series of polymers, the general constitution whereof, despite several varying suggestions, has generally been represented by the structural formula:



where *n* denotes the number of styrene units.

In view of the importance of styrene polymers it was considered pertinent to study the compounds isolated from the seeds of *Prosopis juliflora*. The following describes the isolation and properties of the two polystyrenes.

The defatted (with petroleum ether) dry seed powder (2 kg) was exhaustively extracted with diethyl ether (24 hr) in a Soxhlet extractor over a water bath. The ethereal extract on concentration gave a greenish viscous liquid. This was re-dissolved in a small quantity of diethyl ether (25 ml) and ethanol was added dropwise with constant stirring when a greenish white precipitate was obtained which was filtered by suction.

The crude compound was dissolved in benzene (20 ml), filtered and made turbid by dropwise addition of ethanol and allowed to stand overnight at ambient temperature when a light green compound got deposited. Repeated crystallization and decolourization with animal charcoal and final crystallization with ethyl acetate afforded a white amorphous powder melting at 155° C. Further crystallization did not improve the melting point. The purity of this compound 'A' was tested by thin layer chromatography when a single spot appeared on spraying the plate with sulphuric acid (2N) and subsequent heating for 30 min. in a hot air oven at 120° C (Yield, 753 mg).

After the isolation of the diethyl ether-soluble compound 'A', the residue was further exhaustively extracted with benzene. The extract on similar treatment as for compound 'A' afforded a dark brown viscous liquid. This was purified by repeated crystallization and boiling with animal charcoal in benzene and finally chromatographed over deactivated alumina column and eluted with benezene-ethanol mixture (7:3) when on evaporation of the solvent a greyish white amorphous powder, melting at 209-10° C was obtained. This was re-dissolved in benzene and made turbid with dropwise addition of ethanol and left overnight when an almost white amorphous substance melting at 209–10° was obtained. The purity of the substance was established by TLC, (Yield, 967 mg).

Both the compounds were found to be aromatic in nature. Both gave negative tests for nitrogen, halogens, phosphorus and sulphur and left no ash when ignited in a platinum boat. Both were soluble in benzene, chloroform, carbontetrachloride and ethyl acetate and insoluble in water, ethanol, methanol, petroleum ether, aqueous caustic alkali and mineral acids. The only difference with respect to solubility between the two compounds was that A was soluble in di-ethyl ether and the compound B was insoluble.

Neither of the compounds decolourised bromine water or bromine in carbontetrachloride or acidified potassium permanganate solution. They did not effervesce with sodium bicarbonate solution. They did not give colour reaction with ferric chloride nor reduce Fehling's solution. They did not react with 2:4 -dinitrophynylhydrazene nor give any colour with sulphuric acid.

Elementary analysis and molecular weight determination by Rast's method evinced the following:

Compound 'A' yield 37.6 mg/100 g seed powder

Found	Calculated for $C_{440}H_{440}OR(C_8H_8)_{55}$
C = 91.9%	C = 92.3%
H = 7.7%	H = 9.7%
Mol. wt. = 5725	Mol. wt. = 5720

Compound 'B' yield 48.3 mg/100 g seed powder

Found	Calculated for $C_{968}H_{969}OR(C_8H_8)_{121}$
C = 92.6%	C = 92.3%
H = 7.7%	H = 7.7%
Mol. wt. = 12580	Mol. wt. = 12584

The results showed that compounds are polymers of the aromatic hydrocarbon styrene. Finally the identity of the compounds was confirmed by comparing their infra-red spectra with that of an authentic specimen of polystyrene; they were found to be identical.

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