ment at 6 (± 1)° C for 7 days alternated with a high temperature treatment at 28 (± 1)° C for 5 days[1].

In this laboratory, a very simple, quick, convenient and reliable method was developed to induce sporulation in *Helminthosporium gramineum* using PDA (Potato dextrose agar) containing 2% hot water extract of rice straw. This has induced sporulation in all isolates of the fungus collected from around Kalyani. The fungus was isolated by seeding infected leaf bits on slants containing the said medium. Incubation was done at 25 (± 1)° C in diffused light. Growth of the fungus was visible 2 days after seeding and the culture sporulated profusely by the 5th day. Subculturing on the above medium produced abundant sporulating culture within 3 days. Repeated isolation and subculturing on the above medium yielded sporulating cultures invariably in all cases. The medium adjusted at pH 5, 6 and 7 and incubation at 20 (± 1)° C and 25 (± 1)° C supported luxuriant sporulating culture of the fungus due perhaps to the amino acid content of the Straus extract.

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## EFFECT OF ALBINO RAT GASTRIC JUICE ON THE VIRULENCE OF *MYTHIMNA* (*PSEUDALETIA*) *SEPARATA* NUCLEAR POLYHEDROSIS VIRUS

A SUBSTANTIAL concentration of nuclear polyhedrosis virus (NPV) of the cabbage looper *Trichoplusia ni* is reported to occur naturally on cabbage leaves[1, 2]. Many animals, including man, thus consume such entomopathogens as natural contaminants of food but without toxic effects[3]. The lack of toxicity of entomopathogenic viruses for vertebrates has been attributed to the inactivation of viruses by acidic gastric juice[4]. In the present work, therefore, the effect of albino rat gastric juice on the virulence of *Mythimna* (*Pseudaletia*) *separata* NPV is investigated as orally administered NPV had no toxic effects in albino rats[5].

Albino rats starved for 12 h were operated to ligate the opening of stomach into duodenum. Twelve hours later, rats' stomach was cut open to collect the gastric juice. The latter was then centrifuged at 2,000 rpm for 30 min to remove solid particles. The treatments were as mentioned below.

Suspensions of polyhedral inclusion bodies were mixed with the following in 1 : 7 ratio by volume : (A) Normal gastric juice (pH 2·0–2·5), (B) Buffer (pH 2·3) and (C) Neutralised gastric juice (pH 7·0). The mixtures, incubated at 37° C for 2 h, were then mixed with artificial diet and fed individually to 3rd instar *M.* (*P.*) *separata* larvae. Groups D and E, containing the larvae fed untreated virus and distilled water, served as treated and untreated controls respectively. There were 25 larvae at each treatment and the concentration of virus used was $10{\cdot}0 \times 10^6$ polyhedral inclusion bodies/larva. All the larvae maintained individually in plastic cups were provided daily with fresh diet. Larval mortalities by NPV which determined the virulence were recorded every day.

Results are presented in Table I.

TABLE I

*Effect of albino rat gastric juice on the virulence of NPV of* M. (P.) separata

| Group | Treatment | Larval mortality (%) |
|---|---|---|
| A | NPV + Normal gastric juice | 8 |
| B | NPV + Buffer | 20 |
| C | NPV + Neutralised gastric juice | 88 |
| D | Untreated virus | 92 |
| E | Distilled water | 4 |

*N.B.* : Groups D and E served as treated and untreated controls respectively.

The result for the group A indicated that the normal gastric juice of albino rats inactivated the virus as evidenced by the low larval mortality (8%) by NPV. Similarly human gastric juice also inactivated the NPV of *Heliothis zea*[4]. This inactivation is due to the high acidic nature of gastric juice as neutralised gastric juice has little effect on virulence of virus and larval mortality by NPV did not vary significantly from that recorded in group D. The view is supported by the finding that buffer of pH 2·3 (group B) also inactivated the virus. Further, that acidic pH does decrease the virulence of virus is supported by several workers[6, 7]. Thus, the inactivation of *M.* (*P.*) *separata* NPV by albino rat gastric juice will further add to the safety of the said NPV for albino rats.

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## GLUCOSE-6-PHOSPHATASE ACTIVITY IN THE EPIDIDYMIS OF *TAPHOZOUS LONGIMANUS* (MICROCHIROPTERA : MAMMALIA)

GLUCOSE-6-phosphatase (G-6-Pase, E.C. 3.1.3.9) has been reported in a variety of mammalian tissues where its apparent function is to allow glucose to be released into the circulation. This enzyme has been localised in the epididymis of only a few mammalian species[1,2].

Despite their world-wide distribution, peculiar aerial life and strange reproductive strategies, very little is known about the epididymal enzymology of Chiroptera[3,4]. The present report concerns the regional differences in the histochemical site and distribution of G-6-Pase in the caput, corpus and cauda epididymis of sexually mature males of insectivorous Microchiroptera—*Taphozous longimanus*.

Males of *T. longimanus* were netted at dusk while emigrating from their roost for nocturnal activities. They were maintained in batches of 3–4 in steel cages with wire screen for 24 h during which they had *ad lib* access to sugared water which they sipped avidly.

The animals were sacrificed by cervical dislocation and the epididymis along with testis were dissected out surgically under semi-sterile conditions. They were freed from blood and connective tissue, and washed briefly in cold mammalian ringer.

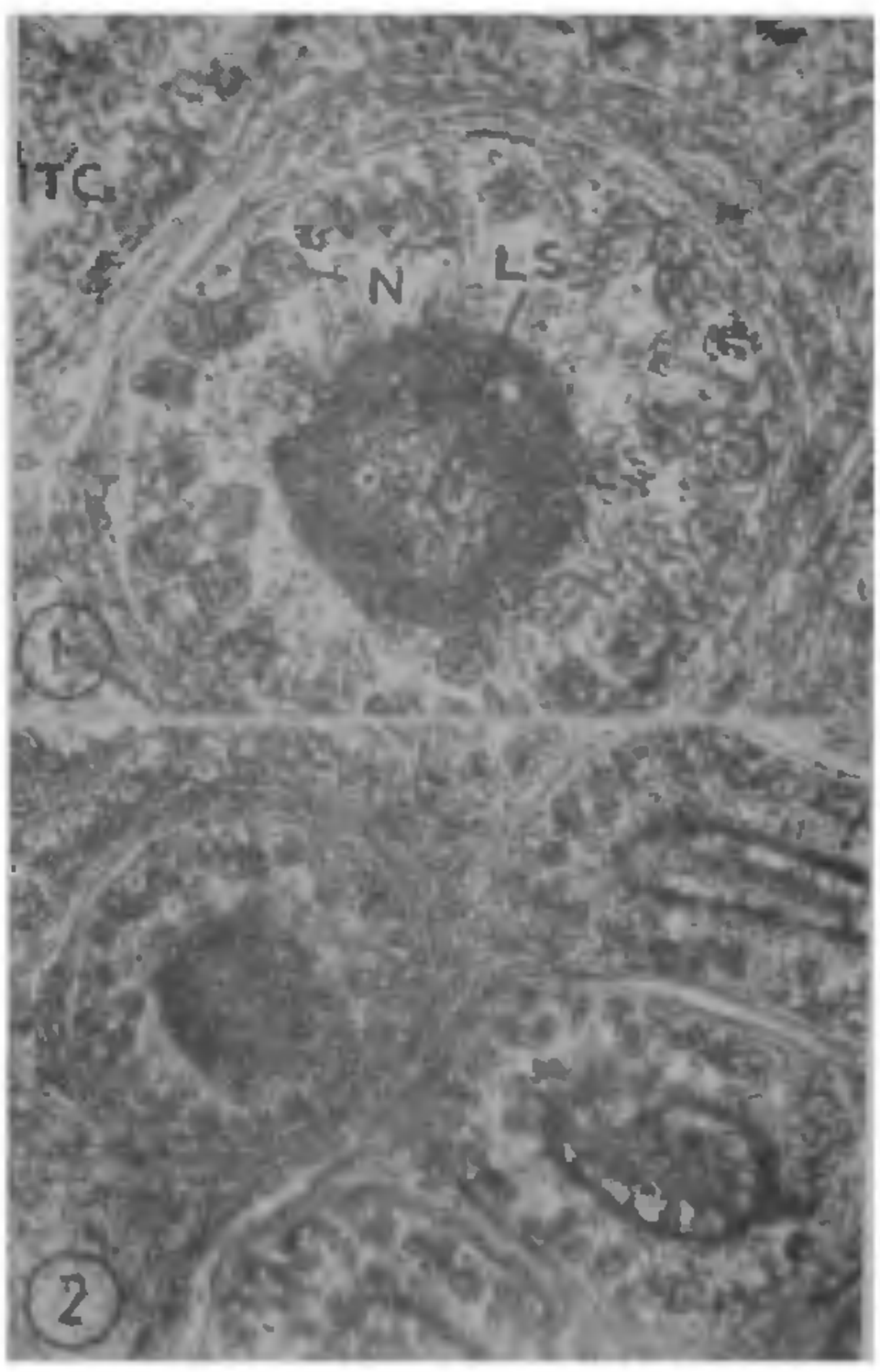
Fresh frozen sections of caput, corpus and cauda epididymis were cut at 10 $\mu$m. The sections from each epididymal segment were identically incubated in the substrate medium (pH 6·0) at 37° C for 40 min according to Wachstein and Meisel's method[5] and fixed in neutral formalin (10%). Brownish black deposit of lead sulphide was taken as the index for ascertaining the site and distribution of G-6-Pase activity in the epididymal segments. Appropriate controls were run simultaneously.

Enzyme activity was visually scored as intense (++++), high (+++), moderate (++), slight (+) and negative (0). An integrated table was made to display the regional histochemical differences in G-6-Pase activity in various epididymal segments.

G-6-Pase activity was observed to show regiona-differences in the caput, corpus and cauda epididymis (Table I, Figs. 1–6). The order of enzyme intensity was corpus > cauda $\geq$ caput. High enzyme reaction was discerned in the intertubular connective tissue; luminal spermatozoa were G-6-Pase positive (Figs. 2, 3, 5 and 6).

Maximal enzyme activity was exhibited at the base of the epididymal epithelium in all the regions. The apex of the cells showed moderate G-6-Pase reaction while the cytoplasm indicated slight enzyme activity. Nuclei of the cells exhibited positive reaction (Figs. 1, 3 and 5).

FIGS. 1–6. Glucose-6-phosphatase (G-6-Pase) activity in the caput, corpus and cauda epididymis of *Taphozous longimanus*.




FIGS. 1–2. Caput epjdidymis showing varying profiles of G-6-Pase reaction in the tubule epithelium, inter-tubular connective tissue and lumen. Note the enzyme activity in the luminal spermatozoa (400 ×).