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HAEMAGGLUTINATION BY CERTAIN BACULOVIRUSES

K. NARAYANAN
Division of Entomology and Nematology,
Indian Institute of Horticultural Research,
Hessaraghatta Lake Post, Bangalore 560089, India.

AMONG the various entomopathogens, baculoviruses, comprising of nuclear polyhedrosis virus (NPV) and granulosis virus (GV) seems to have a great potential for the control of insect pests. Nearly 1271 host-virus records have been reported[1]. However, before considering the widespread field application of such viruses, a detailed knowledge of their characteristics appears essential. Haemagglutination is a property of a number of animal viruses[2]. Since the discovery by Hirst[2], that influenza virus could cause haemagglutination, this reaction has provided an effective tool for the study of certain basic properties like invasive mechanism of animal viruses. But in the case of insect viruses, haemagglutination was generally unsuccessful[3], until Miyajima and Kawase[4] showed that cytoplasmic polyhedrosis virus (CPV) and NPV of silk worm, *Bombyx mori* agglutinated chicken, sheep and mouse erythrocytes and by Shapiro and Ignoffo[5] with NPV of *Heliothis zea* on chick red blood cells (CRBC). In several cases, the adsorption of the viruses to cells has been mediated through the same receptors that are involved with the agglutination of vertebrate erythrocytes[6]. Hence, an attempt has been made to find out the haemagglutinating property of certain baculoviruses and the results of the study have been presented here.

The NPVs infected larvae of *Spodoptera litura*, *Heliothis armigera* and *Corcyra cephalonica* and GV of *Pericallia ricini*, were triturated in a solution containing 0.14 M sodium chloride and 0.115 M sodium citrate and 0.001 M 1-phenyl-2-thiourea. The triturate was spinned through several layers of muslin cloth. Both polyhedral inclusion bodies (PIB) of NPVs and capsules of GV obtained from those larvae were purified by repeated differential centrifugation and saline water washes and stored at 5°C and used as and when needed. For haemagglutination test, pooled fresh blood from several chicks were drawn into Alsever's solution, centrifuged, and the pellets of RBCs were washed thrice with normal saline. Washed cells were suspended in saline to obtain a 0.5% suspension. Two-fold serial dilutions of virus suspensions were made from a stock containing 1.1×10^6 PIB/ml in saline in perspex plate and to each well equal quantity of erythrocytes suspension was added and incubated at room temperature. The highest dilution of inclusion body suspensions producing 100% haemagglutination was considered as one HA unit. In the case of doubtful results, the perspex plates were incubated at 5°C overnight and observed for HA on the next day. Both normal saline solution and erythrocyte suspension mixture and extract of healthy insect tissue were kept as controls.

It is evident from the result (table 1) that the NPVs of *S. litura* and *H. armigera* and GV of *P. ricini* had the property of agglutinating chick erythrocytes whereas NPV of *C. cephalonica* was negative. The above results with NPV of *S. litura* confirmed the earlier report of Wani *et al*[7]. But the titre obtained in this case is of 1 : 80 which is comparatively low. This may be due to

Table 1 *Haemagglutination response of chick erythrocytes by certain baculoviruses*

Sample	Response	Titre
NPV of *S. litura*	+ ve	1 : 80
NPV of *H. armigera*	+ ve	1 : 160
NPV of *C. cephalonica*	− ve	—
GV of *P. ricini*	+ ve	1 : 80

the long storage of virus or low titre of antigen itself *i.e.* polyhedra. Similar decrease in haemagglutination of the CPV of *B. mori* has been reported when the virus is kept at 4°C for more than 2 days[4]. Specific components like PIB protein, virion or PIB have been shown to contain haemagglutinins and they are capable of agglutinating mammalian or avian erythrocytes[4,5]. The negative response of NPV of *C. cephalonica* may be due to the fairly large number of spindle shaped bodies found along with PIBs of NPV of *C. cephalonica*[8].

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HISTOMORPHOLOGY OF CEREBRAL NEUROSECRETORY SYSTEM IN THE CARIDIAN PRAWN, *CARIDINA RAJADHARI* (BOUVIER)

B. VICTOR* and R. SAROJINI
Department of Zoology, Marathwada University, Aurangabad 431 004, India.
** Department of Zoology, St. Xavier's College, Palayamkottai 627 002, India.*

THE study of hormonal regulation in decapod crustacea has shown the important role played by the neurosecretory system. Histological investigations of neurosecretory cells furnish more information on their nature and secretory activity. Cerebral ganglion (brain) is an important neurosecretory centre in several decapods[1–3]. In many animals, the neurosecretory cells are often clumped into groups, which are very conspicuous features of central nervous system[4]. In the present study the various kinds of neurosecretory cells (NSCs) in the cerebral ganglion of *Caridina rajadhari* were histologically studied and their distribution was demonstrated.

Mature animals of *C. rajadhari* (Crustacea, Decapoda, Atyidae) (2.5 cm long) were collected from Kham river, near Aurangabad, Maharastra. Immediately after collection, the cerebral ganglion of non-ovigerous, intermoult (stage-c) animals with matured ovary was carefully dissected out in Van Harreveld's solution[5] and fixed in aqueous Bouin's for 24 hr. The tissues were dehydrated in ethanol, cleared in xylene and embedded in paraffin wax (m.p. 58–60°C). Serial sections, 5–6 μm thick were cut and mounted on glass slides. The staining method used to locate neurosecretory cells and structures was Ewen's (1962) modification of Gomori's paraldehyde fuchsin (PF) technique with Halmi's (1952) counterstain[6]. The size, shape, differential stainability and cell inclusions were used as the main criteria in identifying the neurosecretory cell types[7].

The cerebral neurosecretory cells of *C. rajadhari* in their morphology and general pattern of distribution were similar to those found in other natantians *Caridina weberi*[1], *Penaeus japonicus*[2] and *Metapeneus monoceros*[3]. In *C. rajadhari*, greater number of NSCs occured in the protocerebrum (figure 1). Two paired groups of NSCs were found on each side of the deutocerebrum (figure 2). The tritocerebrum had small group of NSCs, especially noticeable on the base of the cerebral ganglion (figure 3). The size and staining reactions of the NSCs allowed the differentiation of 3 types called, A-, B- and C-neurosecretory cells (figures 4–5). Three types of NSCs were also observed in the brain of other decapods *Paratelphusa hydrodromous*[8], *Scylla serrata*[9], and *Paragrapsus gaimardii*[10].

A-type cells of *C. rajadhari* were large monopolar PF-positive cells and measured about 21–22 μm in diameter. They were fewer in number and more secretory in activity. They had a coarsely granular fuchsinophilic cytoplasm with perinuclearly arranged patches of large purple granules. The nuclei were oval in shape and the nucleoli had a noticeable affinity for the orange-G component of Halmi's counterstain. Only one nucleolus occurred per nucleus (figure 4). The B-type cells were more numerous than A-cells with 17–18 μm in diameter. The cytoplasm was granular and the perikarya contained secretory granules (figure 5). The most abundant type of cell was the