



## Research paper

### First report of Lygaeid bugs (Hemiptera: Lygaeidae) as a potential pollinator of little ironweed, *Vernonia cinerea* (L.) (Asteraceae)

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**Abstract:** Seed bugs, *Spilostethus hospes* and *Graptostethus verticalis* were collected from the flower of little ironweed, *Vernonia cinerea* (family Asteraceae) in and around Rabindra Sarovar Lake area, Kolkata. SEM study was conducted to examine the pollen grains on the various body parts. Both the species were reported as a potential pollinator of little ironweed for the first time.

**Keywords:** Pollination, Hemiptera, Lygaeidae, *Spilostethus hospes*, *Graptostethus verticalis*, *Vernonia cinerea*, SEM.

#### INTRODUCTION:

Hemiptera are recognized by having piercing and sucking type of mouthparts called rostrum and exhibits a wide spectrum of feeding habits, mostly phytophagous in nature (feed on roots, leaves, stems, seeds, fruits), some are predators, blood suckers,

fungivores, scavengers and ectoparasites. Lygaeid bugs are commonly known as seed bugs of ground bugs, recognised by having 4 segmented antennae and 4 segmented rostrum are important pests of agricultural crops.

As far as roll of bugs as insect pollinators or flower visitors are concerned, very scanty information is available and only a few studies are being reported on hemipterans or flower bugs as pollinators, despite the fact that they are frequently found in flowers, feeding on flower visitors such as thrips (Yasunaga, 1997). Augul (2016) studied insects pollinators and reported hemipteran abundance (about 5.23%), mainly lygaeids on flower of Asclepiadeae, Asteraceae, Fabaceae and Chenopodiaceae, *Nysius ericae* were collected from the flowers of Apiaceae and Asteraceae and *Deraeocoris* sp. on Asteraceae flowers play possible role in pollination of flowers. Cercopidae and

Scutelleridae were reported from the plants, *Cercopis vulnerata* and *Eurygaster* spp. respectively (Henneresse and Tyteca, 2016). Anderson (2003) studied relative importance of birds and insects as pollinators and observed Hemiptera as occasional pollen visitors. Young (1986) observed Hemiptera (Miridae) visiting inflorescences of *Dieffenbachia longispatha*. Mirid bugs among flower visitors reported from west Malaysia (Moog, 2002). The most abundant hemipteran flower visitors found on the male and female inflorescences were *Orius atratus* (Anthocoridae, Hemiptera), followed by *Decomioides schneirlai* (Miridae, Hemiptera) (Ishida *et al.*, 2009). In the recent past, Das *et al.* (2018) studied hemipteran dependency of beet flowers. Raj and Muttu (2014) reported two species of hemiptera, *Nysius* sp. and *Adolenda typicaic* as temperate fruit crops pollinators from different parts of Himachal Himalaya. Bhat and Kaveriappa (1995) reported Anthocorid bugs from flowering trees of *Myristica fatua* from Western Ghats. In order to understand carrying capacity and holding mechanism of pollen grains in lygaeid bugs, SEM studies have been conducted. *S. hospes* and *G. verticalis* (Lygaeidae) collected from the flower of little ironweed, *V. cinerea* (Asteraceae) were examined under Scanning Electron microscope and images were taken to various body parts (antennae, rostrum, legs, head and eyes) carrying pollen grains.

#### **MATERIAL AND METHOD:**

**Collection and field observations:** *S. hospes* and *G. verticalis* were collected by hand picking from the flower of *V. cinerea* (family Asteraceae) from the surrounding areas of Rabindra Sarovar Lake, Kolkata. The place was visited several times in

morning and evening hours and several examples were observed, of which five specimens of *S. hospes* and three specimens of *G. verticalis* were collected for SEM study. Images of Lygaeid bugs were taken in natural habitat while feeding and mating on little ironweed flowers by using Nikon D7000 camera.

**Preservation:** The specimens were carefully killed in a closed container by putting cotton soaked in ethyl acetate to prevent discoloration and hardening of the specimens and to avoid direct contamination of ethyl acetate to pollen present on the bug. Specimens were transferred in ethyl alcohol (70%) after photography (by Leica microscope) and Scanning Electron Microscope study. After the detailed study was complete, the specimens were dried, set pinned, labeled and deposited in NZC.

**Identification:** External morphology of the specimen has been studied under light microscope (Leica M205A) and identification was made by comparing the voucher specimens including types held at National Zoological Collection, Hemiptera Section, Zoological Survey of India, Kolkata and with literatures. The weed plant was identified with the help of literature (Naidu, 2012).

**SEM Study:** Scanning Electron Micrographs were taken with SEM (EVO18) to study pollen on the collected lygaeid seed bug. Freshly killed specimens were fixed on carbon tape on an aluminium plate. After that gold palladium coating was made. During the process of gold plating, first the vacuum was created and then sputter coating was done and sample was kept for 40 min. After that the sample was placed in sample chamber in high vacuum at 20 KV. The different body parts of the specimen were focussed by electron beam

and images were taken in different magnifications (284 X- 1.18 KX).

#### **OBSERVATION AND DISCUSSION:**

##### **Lygaeidae as pollinator and flower visitor:**

Many lygaeid bugs are reported as flower visitors but very few studies have been made whether they act as pollinators or not. In the current study we have reported seed bug, *S. hospes* and *G. verticalis* as plant visitor and pollinator of little ironweed, *V. cinerea*. Many other orders of insects like Hymenoptera, Diptera, Coleoptera and Odonata have been also observed visiting little ironweed during the time period of study. It was observed that bugs visiting little ironweed for feeding, egg laying, mating etc. specimens collected from fresh flowers were found with numerous pollen grains trapped in the hairs on different parts of the body.

##### **Little ironweed, *Vernonia cinerea* (L.):**

The members of Asteraceae, together with the other families in the order Asterales, employ a system of pollination known as plunger, or secondary, pollination. *V. cinerea* is an erect, 15-75 cm long, slender, grooved and ribbed. The stems and leaves are covered with fine white hairs, leaves are variable in shape, broadly elliptic or lanceolate, margins toothed. The flowers are purple to pink, about 6 mm across, with

around 20 flowers and arranged in close terminal flower-heads. The fruits are small 1.3 mm long, 3-4angled and densely pubescent, one-seeded achenes fruits. (Akinnubi *et al.*, 2014). *V. cinerea* commonly grows in the proximity of habitation, thrives in any garden and agricultural soils and is very common in disturbed sites and degraded areas. It invades forest, woodland, grassland, cultivated land, riparian zones (banks of water courses), wetlands and coastal dunes. (Akinnubi *et al.*, 2014)

##### ***Spilostethus hospes* (Fabricius, 1794):**

*S. hospes* belong to the family Lygaeidae (seed bug) are a highly successful family of true bugs found worldwide. They are one of the three largest families within the Heteroptera, yet many aspects of their ecology and evolution remain obscure or unknown. Several lygaeid species are of economic importance due to their status as pests. *S. hospes* is reddish brown in colour with apex of head, inner margin of eyes, antennae, rostrum, two broad central discal band on pronotum, scutellum (excluding apex), central spot to corium, disk of sternum, abdomen and legs black, body hairy. *S. hospes* were observed dwelling both on fresh and dry flowers of little ironweed, but the pollen were observed only on those specimens collected from fresh flowers (Fig. 1-5).

PLATE: 1



Fig. 1



Fig. 2



Fig. 3



Fig. 4



Fig. 5

Fig. 1-5. The seedbug, *Spilostethus hospes* (Fabricius, 1794) visit flowers for, feeding, egg laying and mating

PLATE: 2

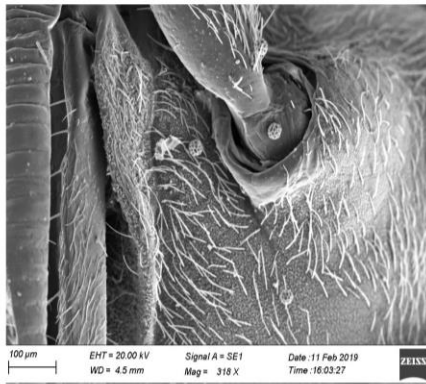


Fig. 6. Pollen grains on base of antennae

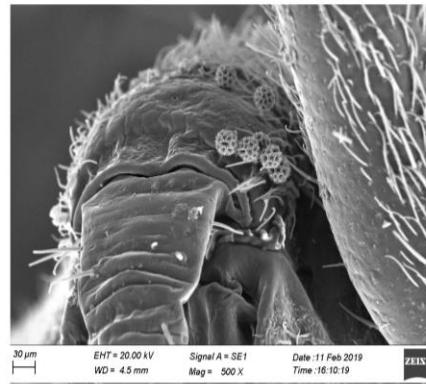


Fig. 7. Pollen grains on base of rostrum

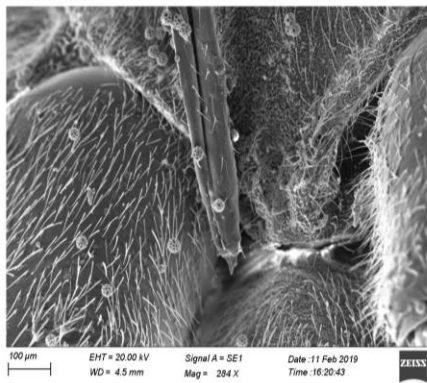


Fig. 8. Pollen grains on apex of rostrum

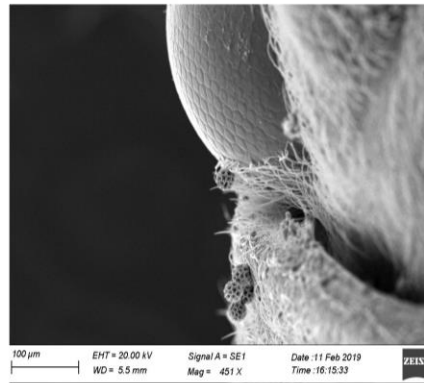


Fig. 9. Pollen grains on base of eye

Fig. 6-9. Scanning Electron Microscope studies of different body parts of *Spilostethus hospes* (Fabricius, 1794) showing pollen grains

PLATE: 3

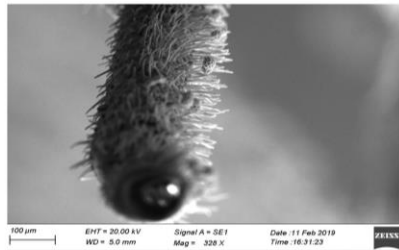


Fig. 10. Pollen grains on apex of antennae

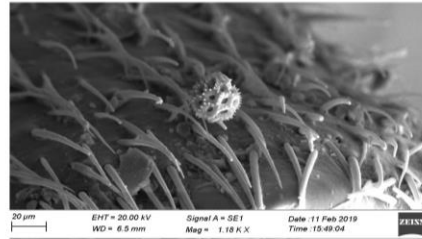


Fig. 11. Pollen grains on sternum

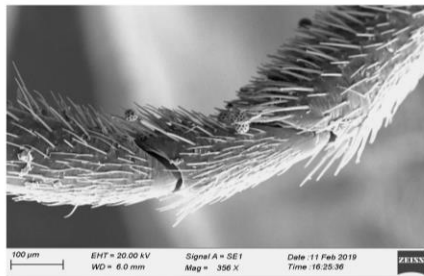


Fig. 12. Pollen grains on tarsi

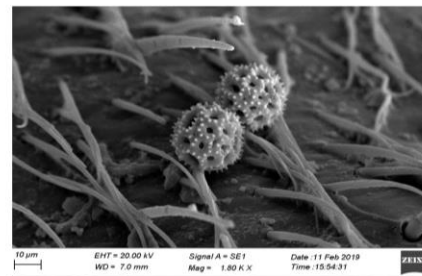


Fig. 13. Pollen grains on head

Fig. 10-13. Scanning Electron Microscope studies of different body parts of *Spilostethus hospes* (Fabricius, 1794) showing pollen grains

***Graptostethus verticalis* (Dallas, 1852):**

*G. verticalis* is less diversified and confined to eastern part of India, can be recognised by having body brick red with dark brown spots and patches between the eyes, two small transverse and two large spots on pronotum, a transverse spots on the base of scutellum, an oval dull grey spot partly on clavus and partly on corium, a dark brown streak on clavus superimposing oval spots, a large dark brown spots on corium towards outer

margins and apical part of corium whitish pink. Membrane dark brown to black with the apical margin white; Legs and antennae brownish black, rostrum black, extending to the posterior coxae. *G. verticalis* was observed dwelling mainly on fresh but few specimens were also found on dry flowers of little ironweed (Fig. 14-16). Relatively more pollen was observed on *G. verticalis* as their body was provided with longer hairs.

PLATE: 4



Fig. 14

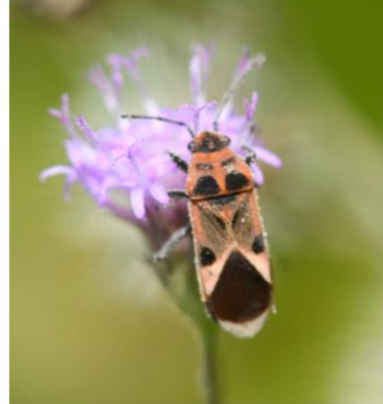


Fig. 15



Fig. 16

Fig. 14-16. The seedbug, *Graptostethus verticalis* (Dallas, 1852) visit flowers for, feeding

PLATE: 5

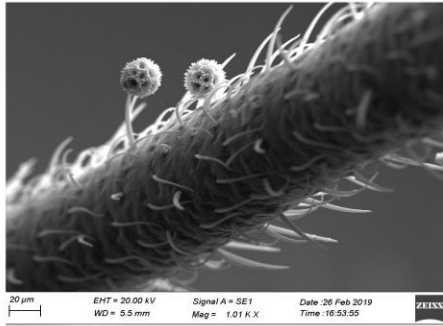


Fig. 17. Pollen grains on 2nd antennal segment

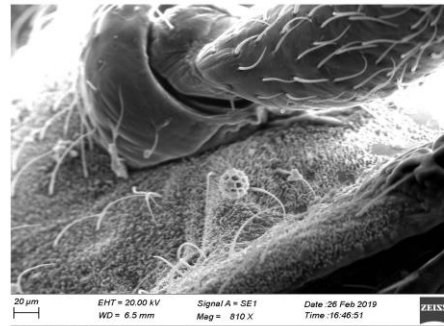


Fig. 18. Pollen grains on base of antennae

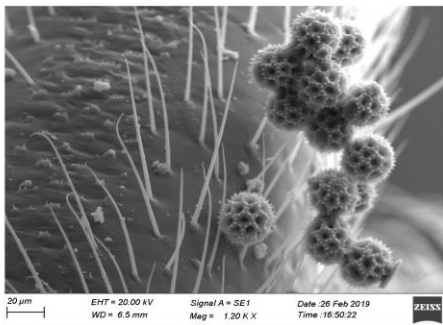


Fig. 19. Pollen grains on fore femur

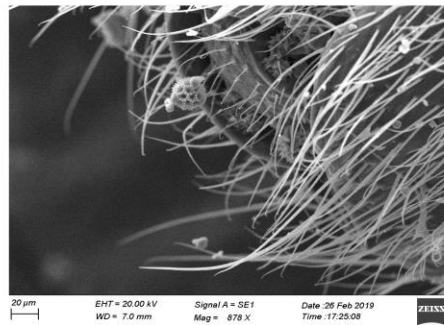


Fig. 20. Pollen grains on external genital segment

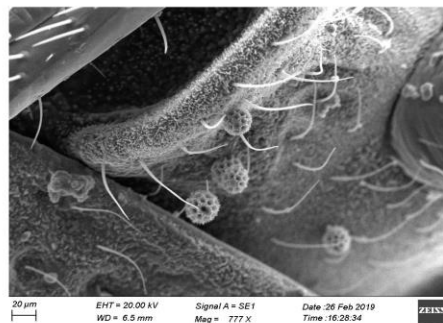


Fig. 21. Pollen grains on ventral side of head

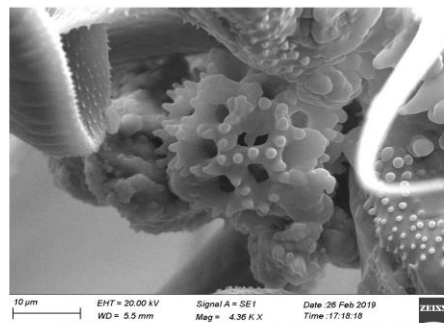


Fig. 22. Pollen grains on hind tarsi

Fig. 17-22. Scanning Electron Microscope studies of different body parts of *Graptostethus verticalis* (Dallas, 1852) showing pollen grains

PLATE: 6

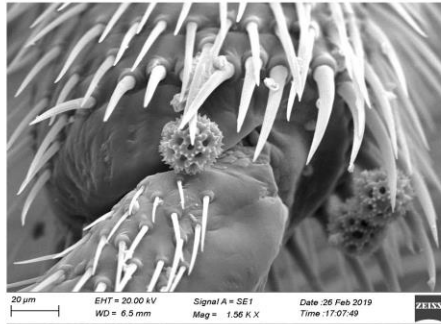


Fig. 23. Pollen grains on hind tarsi

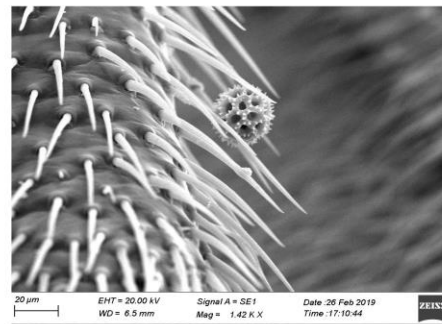


Fig. 24. Pollen grains on hind tibia



Fig. 25. Pollen grains on abdomen (lateral)

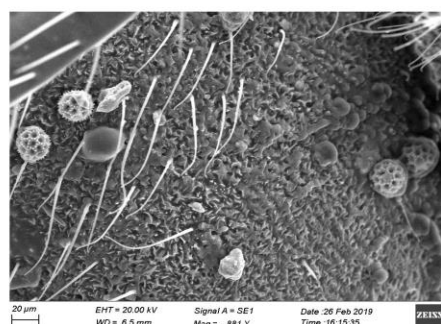


Fig. 26. Pollen grains on mesosternum

Fig. 23-26. Scanning Electron Microscope studies of different body parts of *Graptostethus verticalis* (Dallas, 1852) showing pollen grains

PLATE: 7

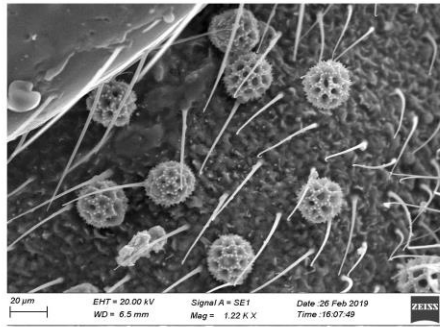


Fig. 27. Pollen grains on metasternum

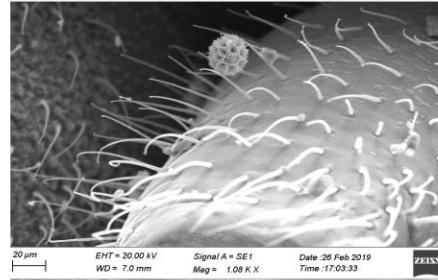


Fig. 28. Pollen grains on middle coxa

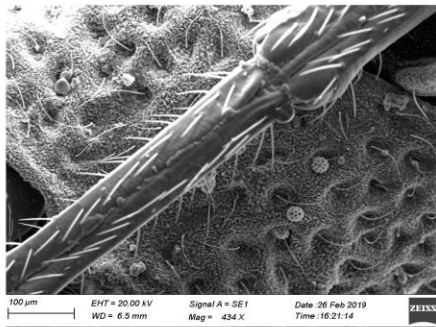


Fig. 29. Pollen grains on prosternum

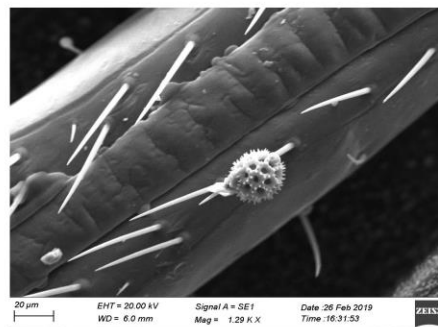


Fig. 30. Pollen grains on rostrum

Fig. 27-30. Scanning Electron Microscope studies of different body parts of *Graptostethus verticalis* (Dallas, 1852) showing pollen grains

### SEM studies:

Scanning Electron Microscope study has shown the presence of pollen both in clusters and in scattered condition in different parts of body mainly on the head, antennae, base of eyes, along with rostrum, sternum, tarsi in both the species of lygaeid bugs. Pollen grains were observed in clusters near base of eyes, apex and base of rostrum, and in scattered conditions to the apex and base of antennae, on head, sternum and tarsi in *S. hospes*. About 60-80 pollen grains were observed per specimen mainly to antennae, basal and apical parts of rostrum, sternum, near compound eyes, head and tarsi (Fig. 6-13). Pollen were found adhere more to the body parts with longer, curved and stout setae, but pollens were also observed on the body parts with smaller setae or even on smooth surface. In *G. verticalis*, more than 120 pollens were observed on various parts of body like, head (ventral side), antennae, base of eyes, few on mesosternum and abdomen, and on legs (mainly femur, tibia, tarsus) (Fig. 17 -30). Pollens observed were polyantoporate, prolatespheroidal, spines having sharp pointed ends with dense pores. Its diameter ranges from 17.14 - 25.72  $\mu$ m.

During the month of February, seed bug was dominated on the plants as compared to the other insects, is an indicative of importance of seed bug as potential pollen vector in little ironweed ecosystem. Mechanism of adherence of pollen grains in hemiptera and in particular lygaeid bugs has not been studied. Parts of the body of bugs (rostrum, legs, antennae, lower parts of body, margins of pronotum and corium) which come in direct contact to male part of the flower receive maximum number of pollen rather on the dorsal side, like pronotum, corium, scutellum. Presence of nectar on rostrum,

legs and other parts of body may facilitate the pollen adherence, even in the less or in the absence of setae, though it needs further detailed study to understand the mechanism of pollen transfer to the bugs and its sustenance on the body. As the insect visit very frequently to other flowers of the same plant and others, transfer of pollen grains are obvious. Synchronization of life cycles of these lygaeid bugs with the flowering of little ironweed provide ample opportunity of pollination.

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