Original Research Article

Pest status, Damage and Biology of an Invasive Pink Pineapple Mealybug, *Dysmicoccusbrevipes* (Cockrell) (Hemiptera: Pseudococcidae) on Sweet flag (*Acorus calamus* Linn.): First Report in India

ABSTRACT

Acorus calamus Linn is a medicinal plant that also has pesticidal properties, primarily against storage pests of agricultural commodities. The creeping rhizomes are the economic part of the plant and are in high demand in Ayurvedic medicine preparation.So far, the pest attacks have not posed a major issue in terms of the economic yield of the plant. In 2022, the presence of an invasive pest, *Dysmicoccusbrevipes*Cockerell, was recorded in the rhizosphere of sweet flag plants maintained at the Pepper Research Station campus, Panniyur, Kannur, Kerala, India. The polyphagous pest has expanded its host range on *A. calamus*. The pest attack was recorded from February 2022 onwards, which was the world's first report. Its biology was evaluated in the laboratory, and it was found that the pest is a parthenogenetic race with an average lifecycle of 59±2.26 days and a fecundity range of 30-74 offspring per adult. Future research should focus on bio-intensive sustainable management packages to protect the IUCN-red list categorized sweet flag from extinction.

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*Keywords: Acoruscalamus,*Below ground pest*Dysmicoccusbrevipes*, Fecundity, Medicinal plant, Pink pineapple mealybug

1. INTRODUCTION

India is home to a wide variety of medicinal plants. Hosting about 7,000 different plants, India is one of the seventeen mega-biodiversity countries in the world recognized by the World Conservation Monitoring Centre (WCMC) of the United Nations Environment Programme.

The sweet flag, *Acorus calamus* Linn. (Acoraceae), an important medicinal herb with a conservation status of least concern on the IUCN red list, is located in the wetland habitats of the Himalayas, Manipur, Naga Hills, and South India. It is a monocotyledonous, semi-aquatic perennial plant with creeping rhizomes (Balakumbahan*et al.,* 2010; Gowthami *et al*., 2021)[1,2]. Apart from that, it is also used as a botanical insecticide against field and storage pests in agriculture. The use of sweetflag in stored cereals and pulses can prevent coleopteran pests' (*Sitophilus granarius* (L.), *S. oryzae* (L.), and *Callosobruchuschinesis* (L.)) buildup by reducing fecundity. (Schmidt *et al., 1991*). Asarones, the phytochemicals present in the essential oil, are the main reason for the insecticidal, growth-inhibiting, and antifeedant potential of sweetflag against insect pests(Balakumbahan*et al.,* 2010; Wang et al., 2022)[1,3]. Direct exposure of the essential oils to the storage pest,sawtoothed grain beetlecauses morphological alterations on insects' cuticles. It also causes the weakest feeding deterrence,which leads to phagostimulation and is followed by post-ingestive lethality (Wijerathna et al., 2023)[4]. The rich insecticidal properties of sweetflag may be the reason for the lower pest attack at the field level, but the presence of rootmealy was mentioned by Balakumbahan*et al. (*2010)[1]. Even though studies for morphological identification and genus-level confirmations have not been reported till now, Thus, the objective of the study was to report the attack, symptoms, and biology of the pink pineapple mealybug (PPMB) on *Acorus calamus*.

Dysmicoccusbrevipes (Cockrell), pink pineapple mealybug, is a polyphagous pest that commonly clusters belowground and above portions of the plant. *D. brevipes* (Cockrell) (Hemiptera: Pseudococcidae) is native to the American tropics (Rohrbach *et al.*, 1988)[7] and distributed to countries in Africa, Asia, Europe, and Oceania (CABI, 2023)[8]. The mealybug colonies were mainly seen on the roots and lower stem (Beardsley, 1993)[9]. According to CABI 2023[8], both nymphs and adults were fed 86 host plants belonging to 35 families and 75 genera. The wide variety of host plants includes perennial grasses, ornamentals, spices, tuber crops, fruit and vegetable crops, and forest trees. The major crops affected by *D. brevipes* (Cockrell) are pineapple, cashew nut, apple, mango, sapota, banana, avocado, guava, grapevine, rambutan, groundnut, areca nut, red gram, citrus, coconut, Arabica coffee, colocasia, taro, cucumber, pumpkin, soybean, potato, African oil palm, date palm, betel vine, black pepper, sugarcane, cotton, sweet potato, clove, cocoa, maize, and ginger (Rajagopal *et al.,* 1982[10]; CABI, 2023 [8]). *D. brevipes* (Cockrell).

Acorus calamus L. and *A. gramineus*are members of the Acoraceae family. Halbert (2003)[11] has reported *D. brevipes* attack on *A. gramineus*. According to Yajun Bai *et al.,* 2020[12], the active chemical components present in *A. calamus* L. and *A. gramineus have a* similar chemical nature. The major constituents are α-asarone (1a) and β-asarone, which have antifeedent and insect repellent activities. These plants also contain a category of alkaloids, amides, diterpenes, flavonoids, lignans, phenylpropanes, sesquinlignans, and sesquiterpenoids. However, as of now, *D. brevipes*has not been reported to occur on *A. calamus* while attacking *A. gramineus*as a pest.

2. MATERIAL AND METHODS

2.1 Sample collection

Female mealybugs were collected from the below-ground portions of the sweet flag plants. Ten adult female mealybugs were collected for artificial rearing, and 20 samples were preserved in 95% alcohol for morphological identification. The morphological characters were examined with stereo zoom binocular microscope.

2.2. Artificial rearing and laboratory studies on biology of *D. brevipes*

The substrate for laboratory rearing of mealybugs was selected as mature pumpkin fruits, which underwent a thorough cleaning, carbendazim treatment (0.1%), and air drying. The treated pumpkins' physical wounds were sealed with wax, and twines were tied along the ridges to help the colony establishment. Mealybug instars were carefully placed on the pumpkin's stalk portion and covered with a basin for seven days. The basin was removed after one week, and colony growth was observed visually for further studies(Fig. 1) (Najitha Ummer, 2016)[5].

The first instars of *D. brevipes* were individually released on small sweetflag rhizome bits in 20 replications. Moultedexuvia of each instars were taken as developmental stage confirmation. The pre-larvipositon period of the newly moulted adult was recorded as the period in between the last moulting and the point of beginning larvipositon. During larviposition, the period of larviposition as well as the adult's fecundity were recorded. Fecundity was recorded by counting daily hatching instars, and counted instars were removed for accuracy in counting. The post-larviposition period is recorded as a duration that lies between the end of the larviposition and the death of the adult (Manjushree, 2016)[6].

2.3.Pest monitoring and symptoms
The pest attack of *D. brevipes*, visual observations on the attacked plant and its rhizosphere were recorded.

3. RESULTS AND DISCUSSION

3.1 *Dysmicoccusbrevipes*- Distribution and first report on Sweetflag

The pest attack of *D. brevipes* (Cockrell) was first recorded from *Acorus calamus o*n February 10th, 2022, from the sweet flag plants maintained in pots at Pepper Research Station, Panniyur (Lattitude 12° 4' 51.6648" N, Longitude 75° 23' 55.464" E) (Figs. 2, 3, 4, and 5). This is the first report in the world. The mealybug samples were identified as *Dysmicoccusbrevipes* (Cockrell). Species confirmation was done by Dr. Sunil Joshi, Principal Scientist, ICAR-NBAIR, Bangalore, India.

As a raw material for various ayurvedic medicine preparations, its IUCN conservation status, the pest problem recorded on sweetflag should be taken as a serious issue. Because the pest is expanding its ecosystem by identifying different types of crops, Apart from pineapple, pink pineapple mealybug has already invaded and succeeded their generations on fruit bunches of oil palm (Dhileepan, 1992)[13], immature nuts of coconut (Radhakrishnan, 2003)[14], pineapple (Joy *et al.,* 2021.)[15], and the roots and basal stem region of pepper (Devasahayam *et al.,* 2009)[16]. According to Egelie and Gillet-Kaufman (2015)[17], the pest can complete its life cycle on a single or more than one host plant. Hence, host expansion of the pest and its population buildup will be a threat in the future.

3.2. Biology

According to Beardsley (1959)[18], parthenogenetic and bisexual races are present in *D. brevipes,* where parthenogenetic races are common and bisexual races are restricted in Brazil, the Dominican Republic, Martinique, Malaysia, Madagascar, and Ivory Coast. During the laboratory studies on the biology of *D. brevipes* collected from sweetflag, it was found that the females are reproduced parthenogenically and male insects are not recorded. Similar observations were reported from Kerala, India, by Manjusree (2016)[6] on the laboratory rearing of *D. brevipes on* pineapple leaf bits*.*

The main characteristics of the adult female is pinkish in colour with humped oval body covered with a wax coating. Seventeen pairs of lateral wax filaments were seen on the marginal sides and were usually slightly curved in appearance (Mau *et al.,*2007)[19]. According to Egelie and Gillet-Kaufman (2015) [17] and Joy *et al.* (2021)[15], lateral filaments in the anterior region are smaller, and in the posterior region, the length may vary from one-third to one-half or as long as the body. The life cycle of *D. brevipes* has three nymphal instars and an adult stage (Table 1). The average duration taken for the nymphal

Fig. 1 Artificial rearing of *D. brevipes* on pineapple for mass multiplication

period, adult phase, and total life cycle is 22.9±0.78 days, 36.65±1.59 days, and 59±2.21 days, respectively. There is a slight similarity in the duration of each life stage of *D. brevipes*reared on pineapple leaf bits from Kerala, India. The nymphal period, adult phase, and total life cycle were recorded as 40.2±1.10, 23.2±0.78, and 63.4±1.50 days, respectively (Manjushree, 2016). Although the fecundity of *D. brevipes* on sweet flagvaried to 30–74 offsprings/adult, which was found to be a lower rate compared to the studies on the biology of *D. brevipes* on pineapple leaf bits, where the fecundity of females was 80–143 instars/adult. Pre-larviposition, larviposition, and post-larviposition periods of *D. brevipes* on sweet flag rhizomes were recorded as 8.05±0.82, 4.9±0.78, and 9.95±0.95. The studies on *D. brevipes* on pineapple leaf bits by Manjushree(2016)[6] recorded a similar trend in the pre-larviposition, larviposition, and post-larviposition periods. The pre- and post-larviposition periods are 8.7±0.78 days, 4.5±0.5 days, and 9.8±1.16 days, respectively. While comparing the biology of *D. brevipes* on pineapple leaf bits and sweet flag, it was found that the duration of life stages is almost similar, whereas the fecundity of *D. brevipes* on sweet flag is lower, which may be due to the presence of phytochemicals in the plant. According to Wang *et al*. (2022)[3], the exposure of β-asarone to soft bodied hemipterans like *Bemisiatabaci* can cause significant reductions in oviposition duration, fecundity, and hatchability of the pest. Both *B. tabaci* and *D. brevipes* are soft bodied sucking pests under the order Hemiptera; hence, β-asarone may be the reason for reducing fecundity in *D. brevipes.*

Fig 2 &3 : of *D. brevipes*attack on sweet flag

Table 1. Biology of *D.brevipes* on sweetflag

Life Stage	Range (days)	Mean (Duration ±S.D)
1st Instar	8-10	8.85± 0.81
2nd Instar	11-14	12.4 ±1.046
3rd Instar	15-17	15.7 ±0.73
Total nymphal period	22-24	22.9±0.78
Adult pre-larviposition	7-9	8.05±0.82
Larviposition	3-6	4.9±0.78
Post larviposition	8-12	9.95±0.99
Total adult fecundity	30- 74	44.85±11.71
Total days	56-64	59±2.26

**Meandays±Standard deviation (SD)*

3.2. Dysmicoccusbrevipes- pest attack and symptoms on sweetflag

The initial stages of a pest attack are difficult to diagnose. The visual symptoms, like ant colonies on the basal region and, based on the pest attack on the rhizosphere, yellowing of the plant, can be seen in later stages.The most visible symptoms are active ant colonies with loosened soil in the basal region. The major food source of the associated ant colonies is the mealybug-excreted honey dews, rich in free amino acids and sugars. When the plant base is disturbed, the associated ant species will transport the nymphs by holding them in their mouths, which may aid in their protection and dispersal of the pest throughout the field. These ants will place the nymphs on nearby host plants and spread their attack to neighbouring plants. In the below-ground portions, rotting of basal portions by insect feeding injuries may invite associated secondary infections by soil-borne pathogens. Yellowing of lower leaves and arrested growth were observed in severely affected plants. Even so, the intensity of damage was less compared to their major hosts. In severe conditions, their incidence may be seen on the leaf axils of the sweet flag.

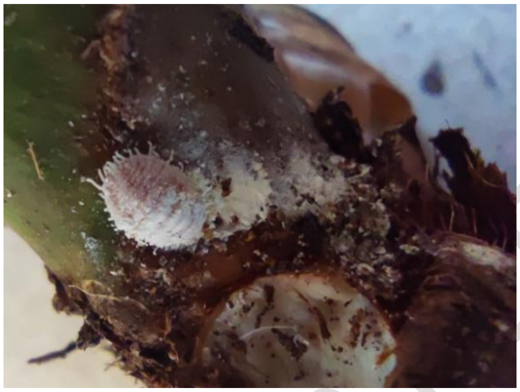

Fig 4. **Fig 5.**

Fig 4. Attack of *D. brevipes* on the leaf axils of sweetflag

Fig 5. *D. brevipes*colonies on the rhizosphere of Sweetflag

4. CONCLUSION

Sweetflag is an ayurvedic medicine and also a folk medicine traditionally used as neonatal medicine. Hence, we should emphasise pest management by preventing the entry of pests from pest-infected areas by focusing on localised quarantine and biointensive management measures at the field level. There should be an effective management measure to conserve the IUCN red list-categorised (least concern) medicinal herb by maintaining the pest population below the economic threshold level. By placing more emphasis on quarantine aspects of this pest, we can reduce its incidence and new host identification in the future. Hence, a comprehensive study of these aspects, as well as sustainable pest management practices, is required.

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