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A study on *Maruca vitrata* infestation of Yard-long beans (*Vigna unguiculata* subspecies *sesquipedalis*)

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Abstract

Globally, *Maruca vitrata* (Geyer) is a serious yield constraint on food legumes including Yard-long bean (*Vigna unguiculata* subspecies *sesquipedalis*). However, there is a dearth of information on its damage potential, distribution and population dynamics in Yard-long beans. In the present study, the level of *M. vitrata* larval infestation on flowers and pods of Yard-long beans in Sri Lanka was determined with respect to three consecutive cropping seasons, Yala, Off and Maha. Results indicated that larval infestation and abundance varied with developmental stage of flowers and pods, cropping season and their combined interactive effects. Flowers of Yard-long beans were more prone to *M. vitrata* larval attack compared to pods. Abundance and level of infestation of *M. vitrata* varied with plant parts, having a ranking of flower buds (highest) > open flowers > mature pods > immature pods (lowest). Peak infestation was observed six and eight weeks after planting on flowers and pods, respectively. Among the three cropping seasons, *M. vitrata* infestation was found to be higher during Maha and Off seasons compared to Yala. The findings of this study contribute to the identified knowledge gap regarding the field biology of an acknowledged important pest, *M. vitrata*, in a previously understudied crop in Sri Lanka.

1. Introduction

Food legumes provide low-fat protein in the human diet and hence are considered as "meat for the poor" (Heiser, 1990). They are also important as high quality livestock fodder and residual nitrogen suppliers in soil, fixing atmospheric nitrogen (Leikam et al., 2007). Amongst food legumes, Yard-long beans which originated in West Africa, is now extensively grown throughout Southeast Asia, Europe, Oceania and North America (Anonymous, 2014). It is often consumed as immature pods as a source of protein (24–27%) (Ano and Ubochi, 2008), vitamins, minerals and fibres (Messina, 1999; Singh, 2005).

Flower and pod-feeding Lepidopterans cause serious yield losses to edible legumes particularly in tropical and sub-tropical zones (Rouf and Sardar, 2011). *Maruca vitrata* (=*testulalis*) Geyer (Lepdoptera: Crambidae), a genetically complex species (Margam et al., 2011; Periasamy et al., 2015), is recognised as one of the most serious legume pests (Abate and Ampofo, 1996; Jackai, 1995; Shanower et al., 1999; Sharma, 1998) due to an extensive host range, high damage potential and cosmopolitan distribution (Margam et al., 2011; Sharma et al., 1999; Taylor, 1967). Larvae of *M. vitrata* feed on flowers, stems, peduncles and pods of food legumes, thus damage occurs at all developmental stages from seedling to podding stages (Singh and Taylor, 1978), however greatest damage occurs at flowering (Singh and Jackai, 1988). For example, typical yield losses on cowpea due to *M. vitrata* range from 20–88% (Singh et al., 1990). Thus, Yard-long bean growers face serious losses at pod harvest caused by *M. vitrata* infestation and consequently employ an array of agronomic management regimes such as application of conventional insecticides which cause adverse effects to the environment and human health, but fail to achieve satisfactory level of control (Srinivasan et al., 2012; Yule and Srinivasan, 2013). Globally, whilst much is known regarding *M. vitrata* infestation on cowpea and pigeon pea (Jackai, 1981; Karel, 1985), a knowledge gap exists with regard to *M. vitrata* infestation of Yard-long beans. Thus, there is a paucity of information regarding damage potential, distribution and population dynamics which are cornerstones for the implementation of successful management strategies. The objective of this study was to explore the epidemiology of *M. vitrata* on field grown Yard-long beans in Sri Lanka during three consecutive cropping seasons.

2. Materials and methods

The study area was located at 'Wilpita North', Matara district (6°5′ 60N″, 80°31′ 0E) in the southern province of Sri Lanka. Average annual rainfall in

this region is < 2500 mm (Punyawardana et al., 2003) with a mean diurnal air temperature depending upon season in the range 25–30 °C. The terrain is rolling, undulating and flat. Major soil groups in this region constitute red yellow "podsolic" soils with strongly mottled sub-soil and bog and half bog soils (Punyawardana et al., 2003). Rain fed rice cultivation is prominent in this region and has two main cropping seasons, April to August, known as Yala and November to February known as Maha. For approximately 15 years, common agronomic practice is to cultivate Yard-long beans on a commercial scale, integrated with rice during the Yala and Maha seasons. In addition, some Yard-long bean growers cultivate between August and November, i.e., the so-called Off season. Typical pre-plant fertiliser applications are a 45:135:65 kg ha^{-1} mixture of urea, triple super phosphate (TSP) and murate of potash (MOP). Two weeks later, urea (45 kg ha^{-1}) is applied as a top dressing. Weeding is done manually at weekly intervals.

Four, 75 m^2 fields of Yard-long beans (variety, "Mas mae") were selected at random in "Wilpita North" region. Plants were grown supported by wooden poles in two rows, 75 m long and spaced 0.9 m apart, with 0.3 m between plants. The level of *M. vitrata* infestation was determined based on the larval infestation at two different developmental stages of flowers (fully opened flowers and flower buds) and pods (immature and mature).

Plant parts were sampled during three consecutive cropping seasons Yala, Off and Maha. During Yala and Maha, flower and pod samples were collected from all four study fields. Samples were also taken during the Off season but only from three fields as the fourth was not in cultivation during this period. During each season, 10 samples were collected from each plant part at 6, 7, 8 and 9 weeks after planting (WAP).

In total, 1760 flowers and pods were sampled, 480 for fields 1, 2 and 3, and 320 for field 4. Sampled flowers and pods were kept separate in self-sealing plastic bags and transported to the laboratory in cool boxes. Subsequently, flowers and pods were carefully dissected under a stereo-microscope and any *M. vitrata* larval stages encountered were removed and placed in 70% ethanol. Thereafter, the total length of all collected larval stages was measured. Total larval abundance from individual flowers and pods was recorded. A plant was deemed infested if at least one larva was detected within a flower or pod. The percentage of infested flowers and pods per field with respect to season was calculated.

Larval infestation and abundance data were arcsin and square root ($\sqrt{x + 1}$) transformed, respectively, prior to statistical analysis for normalization of the experimental data. Level of infestation and abundance between flowers and pods as well as among the different developmental stages of flowers and pods in three consecutive cropping seasons were compared using factorial

ANOVA. Also, a one-way ANOVA was performed to determine whether *M. vitrata* infestation and abundance changed temporally across cropping seasons. All analyses were performed using SAS software (SAS Institute, 1999) at a probability level of 5%.

3. Results

3.1. Infestation and abundance of *M. vitrata* larvae

Of the 1760 flowers and pods sampled, mean percentage flower and pod infestation per field was 33.9 ($\pm$1.9) and 6.6 ($\pm$0.9) larvae, respectively. In addition, mean larval abundance on flowers and pods per field was 220.7 ($\pm$36.6) and 44.0 ($\pm$5.4), respectively. Larval infestation and abundance were significantly greater in flowers than pods in Off ($P < 0.0001$) and Maha ($P < 0.0001$) seasons (Fig. 1a,b). Overall infestation and abundance during Yala season were significantly lower compared with the other seasons (infestation, Off and Maha $P < 0.0001$; abundance, Maha $P < 0.0001$, Off $P = 0.0032$). Moreover, a significant interactive effect was detected between the plant unit and cropping season, for both larval infestation ($P < 0.0001$) and abundance ($P = 0.0016$). Greatest flower infestation (50.6 $\pm$ 3.6) and abundance (130.7 $\pm$ 20.6) were both recorded during the Off season (Fig. 1a,b). Pods exhibited no significant differences with respect to *M. vitrata* larval infestation and abundance across the three cropping seasons (Fig. 1a,b).

3.2. Effect of developmental stages of flowers and pods on infestation and abundance of *M. vitrata* larvae

Larval infestation ($P < 0.0001$) and abundance ($P < 0.0001$) were significantly different amongst the four sampled plant components with a ranking of flower
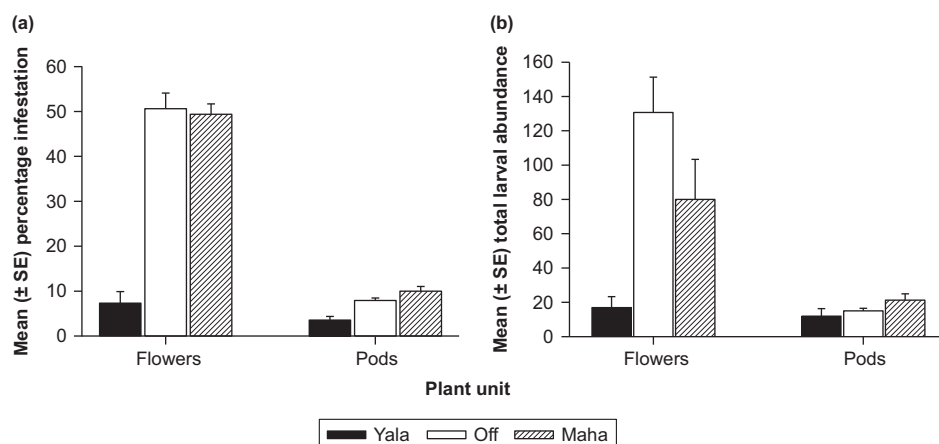

Fig. 1. Mean ($\pm$SE) percentage larval (a) infestation and (b) abundance on flowers and pods of Yard-long beans with respect to three consecutive cropping seasons (Non transformed data are presented).
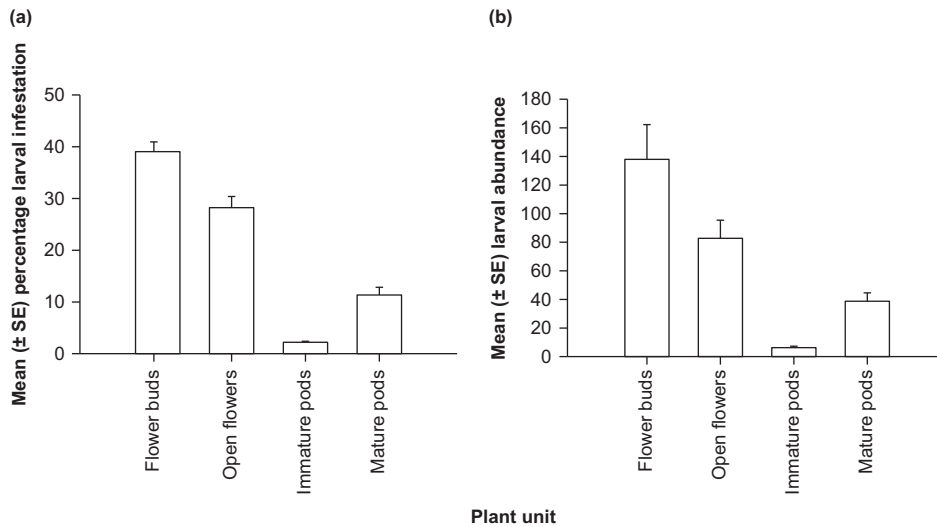
Fig. 2. Mean (±SE) percentage larval (a) infestation and (b) abundance on four different developmental stages of plant units irrespective of cropping season (Non transformed data are presented).

buds (highest) > open flowers > mature pods > immature pods (lowest) (Fig. 2a,b), irrespective of the cropping season. A seasonal effect was also noted for infestation ($P < 0.0001$) and abundance ($P < 0.0001$). Moreover, a significant interactive effect was detected for different flower and pod developmental stages and cropping season with respect to both infestation ($P < 0.0001$) and larval abundance ($P < 0.0001$). A significantly greater larval infestation in flower buds ($P = 0.006$) was recorded compared to open flowers but only in Maha season. However, the larval abundance in flower
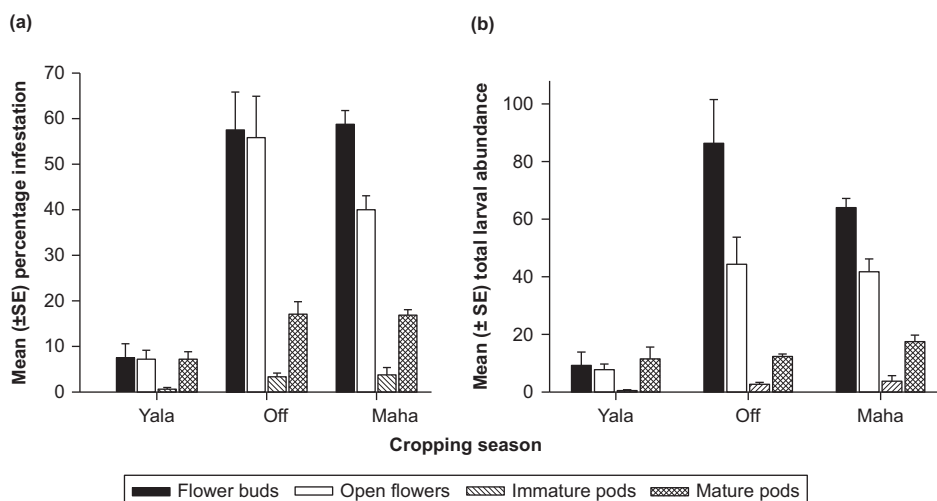

Fig. 3. Mean (±SE) percentage larval (a) infestation and (b) abundance on flowers and pods of Yard-long beans with respect to four different plant units in three cropping seasons (Non transformed data are presented).
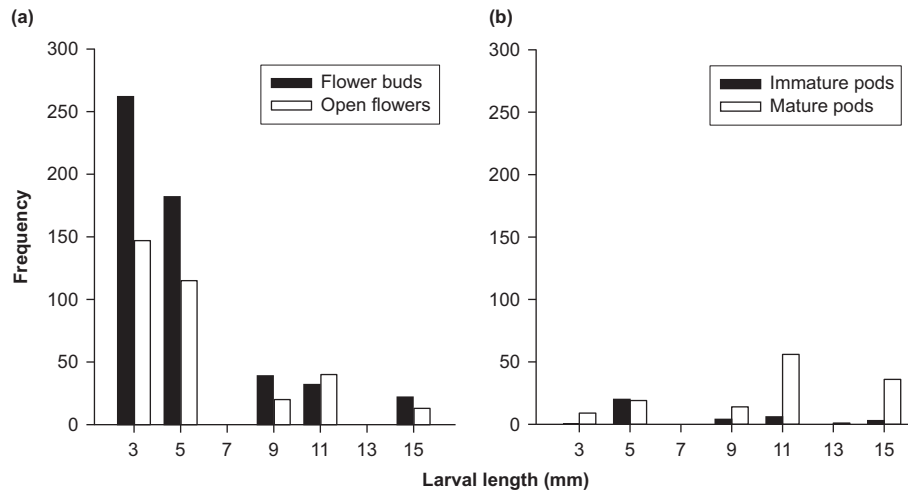
Fig. 4. Length frequency distribution of *M. vitrata* larvae found in (a) flower buds and open flowers and (b) immature and mature pods of Yard-long beans.

buds was found to be higher in both Maha ($P = 0.018$) and Off ($P < 0.001$) seasons (Fig. 3a,b). Infestation level and larval abundance were significantly greater in mature compared with immature pods during all seasons (Maha, $P = 0.0011$; Off, $P = 0.0209$; Yala, $P = 0.0015$) (Fig. 3a,b).

The body length of *M. vitrata* larvae collected from the flower buds (4.69 ± 0.13 mm) and the open flowers (5.02 ± 0.17 mm) were significantly shorter compared with those from immature (7.05 ± 0.64 mm) and mature pods (9.92 ± 0.32 mm); flower buds vs immature pods $P < 0.0001$; flower buds vs mature pods $P < 0.0001$; open flowers vs immature pods $P = 0.0002$; open flowers vs
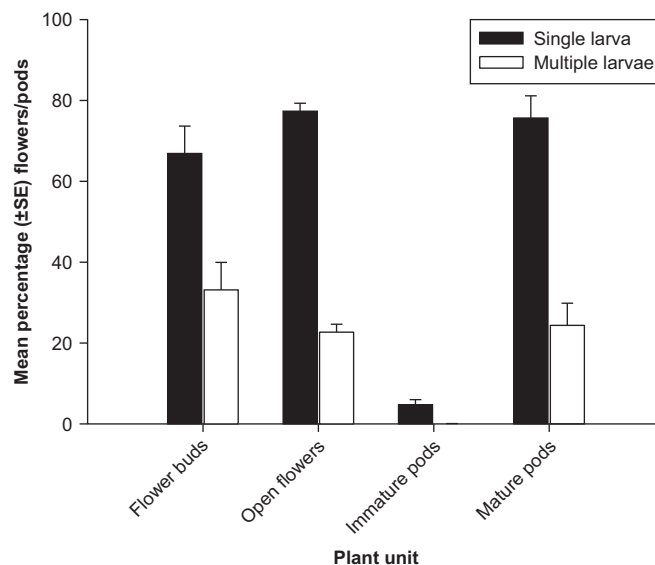

Fig. 5. Mean (±SE) percentage flower buds, open flowers, immature and mature pods of Yard-long beans containing single and multiple larvae of *M. vitrata* (Non transformed data are presented).

mature pods $P < 0.0001$. The most prevalent larvae on flower buds and open flowers were in the range of 2–6 mm. In contrast, the prevalent larvae on pods were in the size ranges 11–12 and 15–16 mm (Fig. 4a,b). A significantly higher ($P < 0.0001$) component of the Yard-long bean crop at all developmental stages was infected by a single rather than multiple larvae (Fig. 5).

3.3. Infestation and abundance of *M. vitrata* larvae over time

Whilst a trend of higher larval infestation and abundance on flowers was recorded at six WAP (Fig. 6a–f), only during Maha season was this significant
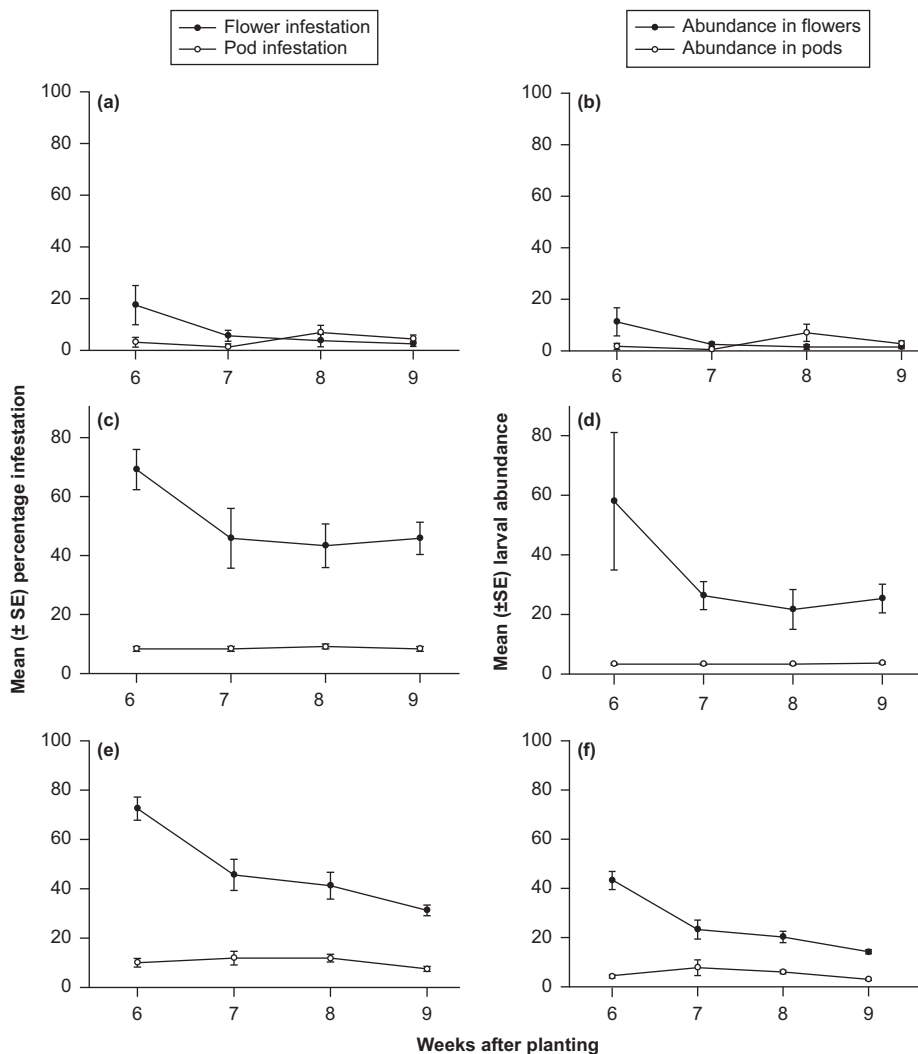

Fig. 6. Mean (±SE) percentage infestation larval abundance with respect to flowers and pods of Yard-long beans in three consecutive seasons; (a) flower and pod infestation in Yala; (b) larval abundance in flowers and pods in Yala; (c) flower and pod infestation in Off; (d) larval abundance in flowers and pods in Off; (e) flower and pod infestation in Maha; (f) larval abundance in flowers and pods in Maha.

($P < 0.0001$) (Fig. 6e,f). In contrast, *M. vitrata* larval abundance and infestation in pods did not vary during the same period (Fig. 6a–f).

4. Discussion

Maruca vitrata is a serious pest of food legumes in several countries in Asia, for example, India (Sharma, 1998), Thailand (Buranapanichpan and Napompeth, 1982), Bangladesh (Das and Islam, 1985) and Pakistan (Ahmed et al., 1987). Although there are several reports of *M. vitrata* infestation on Pigeon pea (Dharmasena et al., 1992; Fellows et al., 1977; Sharma et al., 1999), data is lacking for Yard-long beans. Here, we address this knowledge gap and report data on *M. vitrata* from field grown Yard-long beans in Sri Lanka.

We have identified season as an influencing factor for *M. vitrata* larval incidence and abundance of Yard-long bean flowers and pods. Larval infestation and abundance was significantly reduced during the period April to August, known locally as Yala. Previously, a significant relationship was documented between *M. vitrata* incidence and total rainfall, including number of rainy days, between crop emergence to flowering i.e., wetter conditions led to an increase in *M. vitrata* infestation (Liao and Lin, 2000). Our study area is located in the southern region of Sri Lanka, with an average rainfall of 84.4 mm during Yala, compared to that for both Maha (355.9 mm) and Off seasons (335.7 mm). Thus, our findings are consistent with the previous reports of Fellows et al. (1977) and Saxena et al. (1992) who recorded a high density of *M. vitrata* larvae during Maha and Off seasons on Pigeon pea in Sri Lanka. Furthermore, our data indicates that flowers of Yard-long beans are more vulnerable to damage by *M. vitrata* larvae than pods with overall flower infestation and larval abundance approximately five fold greater than pods. This concurs with previous studies on cowpea, *Vigna unguiculata* (L.) Yalp in Tanzania and Nigeria (Jackai, 1981; Karel, 1985).

Sharma et al. (1999) indicated that first instar larvae of *M. vitrata* had a strong feeding preference for flowers rather than pods. Our larval length data also suggest that the majority of the larvae found in flower buds were early stage (predominantly 2–5 mm), i.e., most probably the first instar larvae. In addition, Jackai (1980) and Sharma et al. (1999) reported that flower buds were the preferable oviposition site for *M. vitrata*. Hence, after emergence from eggs, larvae directly feed on buds leading to higher infestation. Furthermore, Smith (1979) indicated that the level of pod infestation reflected the intensity of the overall *M. vitrata* larval migration/secondary infestation. Migration intensity of *M. vitrata* larvae is a function of age and the density of larvae on the plant at a given time (Jackai, 1981). Hence, high larval densities in flowers, i.e. crowding of larvae, can result in larvae moving from flowers to pods. Our data show that

the majority of the flowers and pods had single larvae infestations of *M. vitrata* suggesting that crowding was not an issue. However, a small percentage of flowers had multiple larvae, including a range of age classes, implying either females deposited more than one egg or several females oviposit at the same location (Taylor, 1967, 1978). Moreover, our data is consistent with Traore et al. (2013) who reported that *M. vitrata* infest flowers and pods of cowpea irrespective of insect age.

Of the two developmental stages of flowers sampled, greater larval infestation and abundance were recorded on flower buds compared to open flowers. This contrasts with Atachi et al. (2002) who detected larval infestation of *M. vitrata* only on open flowers of *Lonchocarpus sericeus* Kunth ex DC (Fabaceae) but not on flower buds. However, Taylor (1978) reported open flowers were the oviposition sites for *M. vitrata* on cowpea rather than flower buds. These apparent contradictory data may be associated with raceme structure and the period between flower bud formation and flower opening (Atachi et al., 2002). In this study, mature pods exhibited greater larval infestation and abundance compared to immature pods similar to Jackai (1980) who reported minimal *M. vitrata* larval infestation on young cowpea pods. Also, Jackai (1981) stated that older pods of cowpea contained later stage larvae of *M. vitrata* similar to our findings. Larvae of *M. vitrata* feed on the young flower parts enclosed within the sepals leading to flower shedding which reduces flowering and subsequent pod setting (Sharma et al., 1999). Dharmasena et al. (1992) recorded 84% pod damage caused by *M. vitrata* larvae in Pigeon pea in Sri Lanka, significantly greater than the pod damage (6.6%) detected on Yard-long beans in this study and an estimated 25% damage on Yard-long beans in Indonesia (Hammig et al., 2008). However, our results were more similar to studies on cowpea, with pod damage of 17–53% in Taiwan (Liao and Lin, 2000) and Bangladesh (Zahid et al., 2008) for lablab bean (18%) and mung bean (20–30%).

The incidence of *M. vitrata* larvae as well as their abundance on flowers and pods of Yard-long beans varied over time within a cropping season with a detectable peak infestation at 6 WAP. This is likely to be related to initiation of flowering of Yard-long beans typically at 5 WAP. It has been reported that the egg stage of *M. vitrata* last five days (Sharma et al., 1999). The significant increase in infestation during Maha season detected 6 WAP was most probably associated with the first larval emergence from eggs. The gradual decrease of larval incidence on flowers after 6 WAP is likely due to the decline in numbers of non-infested flowers and fewer eggs remaining to hatch. Similarly, Atachi and Ahohuendo (1989) also recorded maximum larval density 6 WAP on cowpea in Benin.

In conclusion, the findings of this study indicated that *M. vitrata* larvae caused substantial infestation on Yard-long beans and the higher feeding preference of

M. vitrata larvae to flower buds could be beneficial for the larvae of *M. vitrata* to protect from natural enemies and insecticide treatments. Flower bud would be the most appropriate sampling unit in population dynamics studies of *M. vitrata*. It is recommended to apply control measures before 6 WAP against this pest. Unequivocally, this new data will help to inform growers to develop appropriate control measures for *M. vitrata* in Yard-long beans.

Declarations

Author contribution statement

Ramani C. Jayasinghe: Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

W.T.S. Dammini Premachandra: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Roy Neilson: Conceived and designed the experiments; Wrote the paper.

Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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