



First report of *Leucinodes africensis* and *Leucinodes laisalis* on *Solanum aethiopicum* and *Solanum melongena* in farmer's fields in southern Ghana

Research Paper

Cite this article: Fening KO, Okyere SO, Forchibe EE, Layodé BFR, Richmond TE, Agboyi LKBA, Afreh-Nuamah K, Wamonje FO (2024). First report of *Leucinodes africensis* and *Leucinodes laisalis* on *Solanum aethiopicum* and *Solanum melongena* in farmer's fields in southern Ghana. *Bulletin of Entomological Research* **114**, 359–373. <https://doi.org/10.1017/S0007485324000154>

Received: 19 June 2023
Revised: 14 November 2023
Accepted: 25 February 2024
First published online: 17 April 2024

Keywords:
DNA barcodes; *Leucinodes africensis*; *Leucinodes laisalis*; molecular taxonomy

Corresponding author:
Francis Onono Wamonje;
Email: francis.wamonje@niab.com

Ken Okwae Fening¹ , Stanley Osafo Okyere¹ , Ethelyn Echep Forchibe¹ , Babatoundé Ferdinand Rodolphe Layodé¹ , Tegbe Enyonam Richmond¹ , Lakpo Koku B. A. Agboyi² , Kwame Afreh-Nuamah¹ and Francis Onono Wamonje³

¹University of Ghana, Legon, Ghana; ²CABI, Accra, Ghana and ³Pest and Pathogen Ecology, National Institute of Agricultural Botany, East Malling, UK

Abstract

The eggplant fruit and shoot borer (EFSB) is a devastating pest of eggplants (*Solanum aethiopicum* L. and *Solanum melongena* L.) in Ghana, causing significant economic losses. Although initially thought to be the *Leucinodes orbonalis* Guenee species found in Asia, recent European and Mediterranean Plant Protection Organization reports suggest its absence in Africa. However, eight *Leucinodes* species have been recently described in Africa, including two new species, *Leucinodes africensis* sp. n. and *Leucinodes laisalis* Walker, which were intercepted in eggplant fruits exported from Ghana to the United Kingdom. Despite the reported absence of *L. orbonalis* in Africa, it remains on the pest list of Ghana as a species known to attack eggplants. To accurately determine the identity of the EFSB complex occurring on eggplant in Southern Ghana, molecular and morphological taxonomic tools were employed, and adult male populations were monitored in on-farm conditions. Our results revealed the presence of two EFSB species, *L. africensis* and *L. laisalis*, in the shoot and fruits of eggplants, with *L. africensis* being the dominant species and widely distributed in Southern Ghana. Notably, *L. africensis* males were attracted to the pheromone lure of *L. orbonalis* despite the two species being biologically distinct. This study provides crucial information on correctly identifying the EFSB species attacking eggplants in Southern Ghana and has significant implications for developing management interventions against these pests and their effects on international eggplant trade.

Introduction

Eggplant is a popular vegetable in Ghana due to its rich source of vitamins and minerals. The two primary cultivars, *Solanum aethiopicum* L. (African eggplant) and *Solanum melongena* L. (aubergine), are predominantly grown for local consumption and export, respectively (European Commission Health and Consumers Directorate-General, 2012; Fening and Billah, 2019a, 2019b; Fening *et al.*, 2020). While eggplants can be cultivated throughout the year, recent yield declines have had a negative impact on export value to Europe. In Ghana, achievable eggplant yields are estimated to reach 15,000 kg ha⁻¹; however, in 2016, the recorded average yield was only 50% of this attainable figure (Ministry of Food and Agriculture (MOFA), 2017). Moreover, the value of *S. melongena* fruit exports has experienced an annual decline of 11% from 2008 to 2013 (Food and Agriculture Organization – FAO, 2019). Several factors contribute to Ghana's low yields and diminished value of eggplant exports. These include high labour costs, arthropod pests, inadequate water management (Horna and Gruère, 2006; Horna *et al.*, 2007) and insufficient investment in efficient production technologies (Tsiboe *et al.*, 2019).

Among these factors, arthropod pests are a significant concern (Amengor *et al.*, 2017). Both *S. aethiopicum* and *S. melongena* are susceptible to various arthropod pests, with the eggplant fruit and shoot borer (EFSB), *Leucinodes orbonalis* Guenée (Lepidoptera: Crambidae), being the most destructive (EPPO, 2023). The larval stage of *L. orbonalis* is particularly destructive in its lifecycle. Larvae bore into and feed on the shoots and fruits of eggplants, leading to a reduction in fruit quality and quantity (EPPO, 2023). Infestations associated with *L. orbonalis* have resulted in significant yield losses, reaching up to 70% in eggplant fields in Ghana's Volta region (Amengor *et al.*, 2017).

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Leucinodes orbonalis is classified as an A1 quarantine pest. This classification is based on the regular intercepting of its larvae in eggplant fruits exported from African, Caribbean and Pacific (ACP) countries to Europe, where the European and Mediterranean Plant Protection Organization (EPPO) regions have declared it absent (EPPO, 2023). The presence of A1 quarantine pests can hinder international trade in eggplants, as exemplified by the European Union's (EU) ban on the export of eggplant fruits from Ghana. From October 2015 to December 2017, this ban was imposed due to the frequent interception of *L. orbonalis* larvae and other quarantine pests at border control points (BCPs) in EU Member States (Fening and Billah, 2019a, 2019b; Fening *et al.*, 2020).

Previously, *L. orbonalis* was known to be present in sub-Saharan Africa, as reported by Walker (1859), Frempong (1979) and CABI (2012). However, recent information regarding its distribution suggests that this pest is not as widespread in Africa (EPPO, 2023). Several studies conducted by Hayden *et al.* (2013), Gilligan and Passoa (2014) and Mally *et al.* (2015) focused on the identification of EFSBs intercepted from African consignments. The findings of Hayden *et al.* (2013) and Gilligan and Passoa (2014) indicated that the intercepted EFSB specimens from Africa consisted of three distinct species, distinct from the *L. orbonalis* found in Asia. Mally *et al.* (2015) further identified eight different species of EFSB intercepted from consignments in Africa, namely *Leucinodes africanensis* Mally, Korycinska, Agassiz, Hall, Hodgetts & Nuss, *Leucinodes laisalis* (Walker), *Leucinodes rimavallis* Mally *et al.*, *Leucinodes ethiopica* Mally *et al.*, *Leucinodes pseudorbonalis* Mally *et al.*, *Leucinodes kenyensis* Mally *et al.*, *Leucinodes ugandensis* Mally *et al.* and *Leucinodes malawiensis* Mally *et al.* However, among the intercepted eggplant fruits from Ghana, only *L. africanensis* and *L. laisalis* were found.

It is important to acknowledge that the presence of *L. africanensis* and *L. laisalis* in intercepted eggplant fruits from Ghana does not necessarily indicate that these are the sole *Leucinodes* species attacking eggplants in farmer's fields. The limited sampling of consignments intended for trade within the EU, which occurs at exit points such as airports, suggests that there may be other *Leucinodes* species causing damage to eggplants in the country that have yet to be identified (Everett, 2000; Surkov *et al.*, 2008; Saccaggi and Pieterse, 2013; Fening and Billah, 2019a, 2019b; Seidu, 2022).

Furthermore, a significant concern arises regarding the identification of the EFSB species attacking eggplants in Ghanaian farmer's fields. At the BCPs, consignments are usually sent into a containment facility upon arrival; and a visual inspection of consignments is carried out to detect signs or presence of EFSB species infestations by a trained phytosanitary officer. When the presence or signs attributed to EFSB infestations is detected, consignments are bagged, and sent to the laboratory for in-depth examination and taxonomic identification of EFSB species found in consignments (IPPC, 2020). Surprisingly, the pest list of eggplants in Ghana does not include *L. africanensis* and *L. laisalis*, which have been reported as the species intercepting eggplant consignments (Ministry of Food and Agriculture (MOFA), 2022). Instead, *L. orbonalis* is listed as the EFSB attacking eggplants on farmers' fields in Ghana, despite previous reports suggesting its absence in Africa (Mally *et al.*, 2015; EPPO, 2023). This raises the question of whether the EFSB species solely consists of *L. orbonalis*, as earlier studies suggested (Frempong, 1979; Owusu-Ansah *et al.*, 2001; Mochiah *et al.*, 2011; Ofori

et al., 2015; Ministry of Food and Agriculture (MOFA), 2022), or if *L. africanensis* and *L. laisalis*, as reported by Mally *et al.* (2015) and EPPO (2023), are also present.

Therefore, there is an urgent need to establish the precise identity of the EFSB species attacking eggplants in Ghana to make well-informed decisions. This study aimed to determine the species of EFSB attacking eggplants in eggplant hotspots in southern Ghana, study their phylogenetic relationships and monitor the population of adult males in on-farm conditions.

Materials and methods

Study and sampling sites

In 2022, a survey was conducted to investigate the occurrence of EFSBs in major eggplant production regions of Ghana, specifically the Deciduous Forest and Coastal Savannah agroecological zones. The surveyed regions included Eastern, Greater Accra and Volta (Asenso-Okyere *et al.*, 2000; Ministry of Food and Agriculture (MOFA), 2018). The survey spanned from March to November, covering both the major and minor rainy seasons to capture the complete seasonal cycle of the pest.

A total of ten fields were selected for sampling, consisting of six exporter farms and four local eggplant fields across eight study areas: Adeiso, Asuboi, Azagonorkope, Begoro, Legon, Nsawam, Okorase, and Senchi (fig. 1). The aim was to determine the identity of the EFSB. Furthermore, four exporter farms were specifically chosen for monitoring the population of adult EFSB males in on-farm conditions. These farms were Eric and Trosky at Adeiso, Joekopan at Azagonorkope and Tacks at Senchi. The geographical coordinates of the sampling sites (farmer's fields) are provided in table 1.

Sampling of EFSB

During the survey, a systematic approach was followed to examine the presence of EFSBs. Seventy-five (75) eggplants were randomly selected using an 'X' pattern at each sampling site. The shoots and fruits of these eggplants underwent a thorough examination to identify signs of EFSB infestation. These signs included shoot drooping caused by larval tunnelling inside the shoots, the presence of EFSB larvae within the shoots, and emergence holes created by mature (5th) instar larvae exiting eggplant fruits to pupate in the soil.

Infested eggplant shoots were carefully separated from the plants and opened to extract the EFSB larvae found inside the tunnels. These larvae were preserved in vials containing 95% (v/v) ethanol, appropriately labelled and transported to the laboratory. Upon arrival, they were stored in a refrigerator at 4 °C for identification.

Furthermore, infested eggplant fruits were collected from the sampling sites and placed in containers for transportation to the laboratory. A rearing procedure, adapted from Padfwal and Scrivastava (2018), was employed to rear the EFSB larvae found within the eggplant fruits to the adult stage in a controlled laboratory environment.

EFSB rearing procedure

To facilitate the pupation process of EFSB larvae, the collected infested eggplant fruits were carefully placed in transparent plastic containers. The bottom of each container was covered with

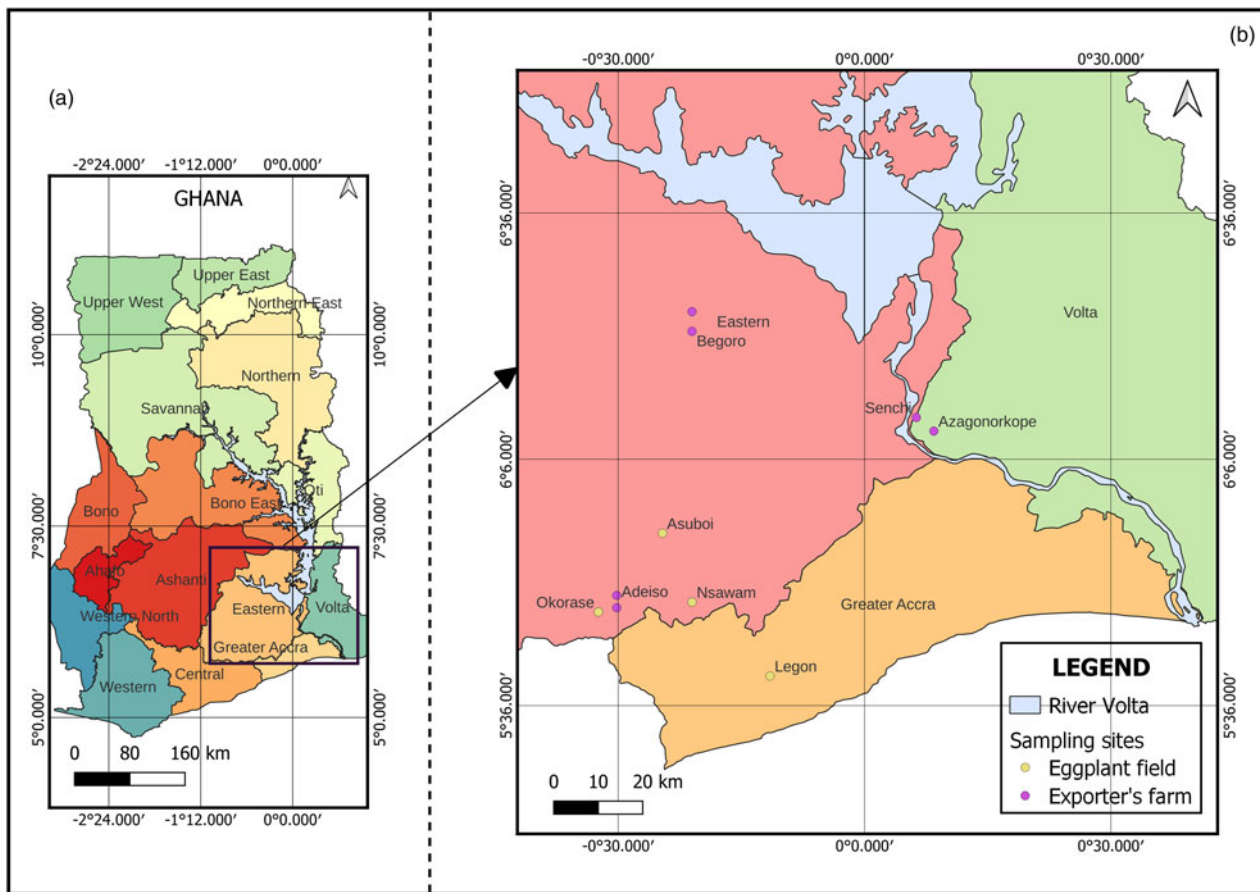


Figure 1. Map of Ghana showing the study areas and sampling sites. The map of Ghana (A) shows the regional political boundaries in different colours. The inserted purple box demarcates the geographic region where our study was conducted. The sampling sites (B) are shown in the blown-up image. The purple and yellow dots show the sites where fieldwork was conducted in the Eastern, Greater Accra and Volta regions.

muslin cloths, providing a suitable pupation site for the larvae. Another muslin cloth was used to cover the exposed area at the top of the plastic container. This setup ensured a controlled environment for pupation. Once the pupae emerged from the larvae in the rearing cages, they were transferred to glass tubes. These tubes

were lined with a muslin cloth at the bottom, providing a comfortable surface for adult emergence. The top of the glass tubes was covered with another muslin cloth. This arrangement allowed for the emergence of adult EFSB specimens while keeping them contained. To provide sustenance for the emerging adults, cotton

Table 1. Study areas and sampling sites of the EFSB (Lepidoptera: Crambidae)

Region	Agroecological zone	Study area	Sampling site	Type of eggplant	Variety	Latitude	Longitude
Eastern	Deciduous forest	Adeiso	*Eric farms	<i>S. aethiopicum</i>	Yogbe	5.81174	-0.50266
			*Trosky farms	<i>S. melongena</i>	Black beauty	5.81058	-0.50309
		Asuboi	Eggplant field	<i>S. aethiopicum</i>	Yogbe	5.95	-0.41
		Begoro	Exporter's farm	<i>S. aethiopicum</i>	Yogbe	6.36	-0.35
			Exporter's farm	<i>S. aethiopicum</i>	Yogbe	6.40	-0.35
		Nsawam	Eggplant field	<i>S. aethiopicum</i>	Yogbe	5.81	-0.35
		Okorase	Eggplant field	<i>S. aethiopicum</i>	Yogbe	5.79	-0.54
Greater Accra	Coastal Savannah	Legon	*Tacks farm	<i>S. melongena</i>	Black beauty	6.18494	0.10538
			Eggplant field	<i>S. aethiopicum</i>	Yogbe	5.65989	-0.19178
				<i>S. melongena</i>	Black beauty		
Volta	Deciduous forest	Azagonorkope	*Joekopan farms	<i>S. melongena</i>	Black beauty	6.15734	0.14091

*Exporters farms.

balls soaked in a 10% sugar solution were placed in the adult cages as a food source. Subsequently, the adult EFSB specimens were humanely euthanised by freezing to preserve them for identification.

Monitoring adult EFSB males' population in on-farm conditions

During the vegetative stage of eggplant cultivation in Eric, Joekopan, Tacks and Trosky farms, a delta trap was set up to capture adult male *L. orbonalis*. A delta trap was baited with sex pheromone lures specifically designed for *L. orbonalis* (P308-Lure manufactured by Chemtica Internacional SA). The active ingredients of the lure were E-11-hexadecenyl acetate and E-11-hexadecenol. The installation of the delta trap occurred at the farms mentioned above, and the trap was monitored every week from the vegetative stage until the maturity stage of the eggplants. Each week, the adult EFSB males captured in the trap were collected. These captured specimens were then identified and counted to determine the population dynamics of the pest.

Morphological identification of EFSB

The dead adults were identified morphologically on a Leica EZ4 D stereomicroscope using the identification keys published by Mally *et al.* (2015) by a curator, H. Davies, at the Insect Museum of the Department of Animal Biology and Conservation Science (DABCS), University of Ghana.

Molecular identification of EFSB

The molecular identification process was performed at the CABI Plantwise Diagnostic and Advisory Service laboratory in the United Kingdom and the National Institute of Agricultural Botany (NIAB) laboratories in the UK. DNA was extracted from adult and larval specimens of the EFSB for samples sent to CABI using the microLYSIS[®]-PLUS extraction technique. For samples sent to NIAB, DNA was extracted using the Norgen Cells and Tissue DNA kit (Norgen, Thorold, 4Y6, Canada). The extracted DNA was then amplified by PCR using the universal mitochondrial cytochrome oxidase (COI) gene primers LCO1490 5'-GGTCAACAAATCATAAAGATATTGG-3' and HCO2198 5'-TAAACTTCAGGGTGACCAAAAAATCA-3' (Folmer *et al.*, 1994) to amplify a section of the COI gene. The quality of the PCR products was assessed using gel electrophoresis, followed by purification using a commercial kit. Samples were diluted to the required concentrations and submitted for semi-automated Sanger sequencing (Sanger *et al.*, 1977; Smith *et al.*, 1986) on the ABI 3130 Genetic Analyzer. The generated DNA sequences were compared with existing sequences in the Barcode of Life Data System (BOLD) and the National Center for Biotechnology Information (NCBI) to identify the *Leucinodes* species.

Phylogenetic analysis

The sequences of the EFSB from GenBank that were similar to the DNA sequences of the specimens used for molecular identification were downloaded in FASTA format and were used for phylogenetic analyses. Pairwise comparison was performed to establish similarity of COI sequences of EFSB obtained in this study. The COI sequences of EFSB identified in this study, some reference

sequences of *Leucinodes* species downloaded from GenBank viz. *L. orbonalis* from Bangladesh, India, Malaysia, Pakistan and Thailand; *L. africensis* from Bangladesh and Nigeria; *L. laisalis* from Kenya, Nigeria and South Africa; *L. kenyensis* and *L. rimavallis* from Kenya; *L. malawiensis* from Malawi; and *L. pseudorbonalis* from Uganda, and a reference sequence of the Mediterranean fruit fly, *Ceratitidis capitata* (included as an out-group for comparison) were aligned using MUSCLE algorithm (Edgar, 2004), and percentage similarity computed in SDT v 1.2 software (Muhire *et al.*, 2014). Following that, the sequences were aligned using MUSCLE (Edgar, 2004) in Molecular Evolutionary Genetics Analysis Version 11 (MEGA 11) (Tamura *et al.*, 2021) and used to construct a phylogenetic tree using the Neighbor Joining (NJ) tree algorithm (Saitou and Nei, 1987) with Tamura-3 parameter (Tamura, 1992). The statistical support for the nodes in the phylogenetic tree was assessed using 1000 bootstrap replicates. All the data used for the phylogenetic analysis can be found in the supplementary file (see Supplementary File 1).

Data analysis of the prevalence of the EFSB male population in eggplant fields

The prevalence of the adult EFSB males was estimated using the fruit fly prevalence estimation indices F/T/W where F = the total number of adult EFSB males captured, T = the number of inspected traps and W = the number of weeks traps exposed in the farmer's field (International Standards for Phytosanitary Measures (ISPM) 30, 2008; Billah and Fening, 2019; Fening and Billah, 2019a, 2019b).

Results

Identification of the EFSB

A total of 834 EFSB (Lepidoptera: Crambidae) were found in the shoot and fruits of the eggplants and pheromone traps mounted at the sampling sites. Following molecular and morphological taxonomic examination, the *L. africensis* and *L. laisalis* were identified as the EFSB infesting eggplants in southern Ghana (table 2). The BLAST search for similarity revealed that the generated DNA sequences of EFSB samples 1–83 selected for identification were >99% identical to the mitochondrial COI sequence of the *L. africensis* identified in eggplant fruits from Nigeria (GenBank Accession number: KM987391.1) (Mally *et al.*, 2015). However, the generated DNA sequences of the EFSB specimens 84 and 85 were both found to be 100% identical to the mitochondrial COI sequence of the *L. laisalis* identified in eggplant fruits from Nigeria (GenBank Accession number: KM987397.1) (Mally *et al.*, 2015). The DNA sequences generated from the specimens used for identification have been deposited in GenBank and assigned accession numbers (table 2). The complete list of all sequenced specimens and the GenBank accession numbers assigned is included in the supplementary information (see Supplementary table 1).

The pairwise comparison of the DNA sequences showed a clear species demarcation between the *L. orbonalis*, *L. africensis*, *L. laisalis*, *L. rimavallis*, *L. kenyensis*, *L. malawiensis* and *L. pseudorbonalis* (fig. 2). Three clusters of closely related sequences having >96% identity were identified. The first cluster comprised the mitochondrial COI sequences of the *L. orbonalis* identified in eggplant fruits from Bangladesh (accession number: LN624686.1),

Table 2. Summary of results of EFSB specimens subjected to molecular identification

Sample number	Sample ID	Location	Crop sampled from	GenBank accession number	BLAST hit	% Similarity
1	Adeiso_1_1	Adeiso	<i>S. aethiopicum</i>	OR058944	<i>L. africensis</i> : KM987391.1	99.68
2	Adeiso_1_2	Adeiso	<i>S. aethiopicum</i>	OR058946	<i>L. africensis</i> : KM987391.1	99.84
3	Adeiso_1_3	Adeiso	<i>S. aethiopicum</i>	OR058948	<i>L. africensis</i> : KM987391.1	99.68
4	Adeiso_1_4	Adeiso	<i>S. aethiopicum</i>	OR058950	<i>L. africensis</i> : KM987391.1	99.68
5	Adeiso_1_6	Adeiso	<i>S. aethiopicum</i>	OR058952	<i>L. africensis</i> : KM987391.1	99.84
6	Adeiso_1_7	Adeiso	<i>S. aethiopicum</i>	OR058942	<i>L. africensis</i> : KM987391.1	99.68
7	Adeiso_1_8	Adeiso	<i>S. aethiopicum</i>	OR058943	<i>L. africensis</i> : KM987391.1	99.68
8	Adeiso_1_9	Adeiso	<i>S. aethiopicum</i>	OR058954	<i>L. africensis</i> : KM987391.1	99.65
9	Adeiso_2_1	Adeiso	<i>S. melongena</i>	OR058941	<i>L. africensis</i> : KM987391.1	99.84
10	Adeiso_2_2	Adeiso	<i>S. melongena</i>	OR058945	<i>L. africensis</i> : KM987391.1	99.84
11	Adeiso_2_3	Adeiso	<i>S. melongena</i>	OR058947	<i>L. africensis</i> : KM987391.1	99.68
12	Adeiso_2_4	Adeiso	<i>S. melongena</i>	OR058949	<i>L. africensis</i> : KM987391.1	99.68
13	Adeiso_2_5	Adeiso	<i>S. melongena</i>	OR058951	<i>L. africensis</i> : KM987391.1	99.84
14	Adeiso_2_6	Adeiso	<i>S. melongena</i>	OR058953	<i>L. africensis</i> : KM987391.1	99.84
15	Adeiso_2_7	Adeiso	<i>S. melongena</i>	OR058955	<i>L. africensis</i> : KM987391.1	99.84
16	Asuboi_1	Asuboi	<i>S. aethiopicum</i>	OR062317	<i>L. africensis</i> : KM987391.1	99.84
17	Asuboi_2	Asuboi	<i>S. aethiopicum</i>	OR062316	<i>L. africensis</i> : KM987391.1	99.84
18	Asuboi_3	Asuboi	<i>S. aethiopicum</i>	OR062313	<i>L. africensis</i> : KM987391.1	99.84
19	Asuboi_4	Asuboi	<i>S. aethiopicum</i>	OR062312	<i>L. africensis</i> : KM987391.1	99.84
20	Asuboi_6	Asuboi	<i>S. aethiopicum</i>	OR062315	<i>L. africensis</i> : KM987391.1	99.84
21	Asuboi_8	Asuboi	<i>S. aethiopicum</i>	OR062314	<i>L. africensis</i> : KM987391.1	99.84
22	Azagonorkope_1	Azagonorkope	<i>S. aethiopicum</i>	OR062327	<i>L. africensis</i> : KM987391.1	99.84
23	Azagonorkope_2	Azagonorkope	<i>S. aethiopicum</i>	OR062328	<i>L. africensis</i> : KM987391.1	99.84
24	Azagonorkope_3	Azagonorkope	<i>S. aethiopicum</i>	OR062329	<i>L. africensis</i> : KM987391.1	99.84
25	Azagonorkope_4	Azagonorkope	<i>S. aethiopicum</i>	OR062330	<i>L. africensis</i> : KM987391.1	99.84
26	Azagonorkope_5	Azagonorkope	<i>S. aethiopicum</i>	OR062331	<i>L. africensis</i> : KM987391.1	99.84

(Continued)

Table 2. (Continued.)

Sample number	Sample ID	Location	Crop sampled from	GenBank accession number	BLAST hit	% Similarity
27	Azagonorkope_6	Azagonorkope	<i>S. aethiopicum</i>	OR062324	<i>L. africensis</i> : KM987391.1	99.84
28	Azagonorkope_7	Azagonorkope	<i>S. aethiopicum</i>	OR062325	<i>L. africensis</i> : KM987391.1	99.84
29	Azagonorkope_8	Azagonorkope	<i>S. aethiopicum</i>	OR062326	<i>L. africensis</i> : KM987391.1	99.84
30	Begoro_1_1	Begoro	<i>S. aethiopicum</i>	OR060667	<i>L. africensis</i> : KM987391.1	99.84
31	Begoro_1_2	Begoro	<i>S. aethiopicum</i>	OR060669	<i>L. africensis</i> : KM987391.1	99.68
32	Begoro_1_3	Begoro	<i>S. aethiopicum</i>	OR062345	<i>L. africensis</i> : KM987391.1	99.84
33	Begoro_1_4	Begoro	<i>S. aethiopicum</i>	OR060674	<i>L. africensis</i> : KM987391.1	99.84
34	Begoro_1_5	Begoro	<i>S. aethiopicum</i>	OR060677	<i>L. africensis</i> : KM987391.1	99.84
35	Begoro_1_6	Begoro	<i>S. aethiopicum</i>	OR060679	<i>L. africensis</i> : KM987391.1	99.84
36	Begoro_1_7	Begoro	<i>S. aethiopicum</i>	OR060682	<i>L. africensis</i> : KM987391.1	99.84
37	Begoro_1_8	Begoro	<i>S. aethiopicum</i>	OR060684	<i>L. africensis</i> : KM987391.1	99.84
38	Begoro_2_1	Begoro	<i>S. aethiopicum</i>	OR060668	<i>L. africensis</i> : KM987391.1	99.84
39	Begoro_2_2	Begoro	<i>S. aethiopicum</i>	OR060670	<i>L. africensis</i> : KM987391.1	99.84
40	Begoro_2_3	Begoro	<i>S. aethiopicum</i>	OR060672	<i>L. africensis</i> : KM987391.1	99.84
41	Begoro_2_4	Begoro	<i>S. aethiopicum</i>	OR060675	<i>L. africensis</i> : KM987391.1	99.84
42	Begoro_2_5	Begoro	<i>S. aethiopicum</i>	OR060688	<i>L. africensis</i> : KM987391.1	99.68
43	Begoro_2_6	Begoro	<i>S. aethiopicum</i>	OR060680	<i>L. africensis</i> : KM987391.1	99.84
44	Begoro_2_7	Begoro	<i>S. aethiopicum</i>	OR060687	<i>L. africensis</i> : KM987391.1	99.84
45	Begoro_2_8	Begoro	<i>S. aethiopicum</i>	OR060685	<i>L. africensis</i> : KM987391.1	99.84
46	Begoro_L_LP_1A	Begoro	<i>S. aethiopicum</i>	OR060686	<i>L. africensis</i> : KM987391.1	99.84
47	Begoro_L_LP_1B	Begoro	<i>S. aethiopicum</i>	OR060689	<i>L. africensis</i> : KM987391.1	100
48	Begoro_L_DP_1A	Begoro	<i>S. aethiopicum</i>	OR060671	<i>L. africensis</i> : KM987391.1	99.84
49	Begoro_L_DP_1B	Begoro	<i>S. aethiopicum</i>	OR060673	<i>L. africensis</i> : KM987391.1	99.84
50	Begoro_L_DP_2A	Begoro	<i>S. aethiopicum</i>	OR060676	<i>L. africensis</i> : KM987391.1	99.84
51	Begoro_L_DP_2B	Begoro	<i>S. aethiopicum</i>	OR060678	<i>L. africensis</i> : KM987391.1	99.84
52	Begoro_L_DP_2C	Begoro	<i>S. aethiopicum</i>	OR060681	<i>L. africensis</i> : KM987391.1	99.84

(Continued)

Table 2. (Continued.)

Sample number	Sample ID	Location	Crop sampled from	GenBank accession number	BLAST hit	% Similarity
53	Begoro_L_W_2	Begoro	<i>S. aethiopicum</i>	OR060683	<i>L. africensis</i> : KM987391.1	99.84
54	Legon_1_1	Legon	<i>S. melongena</i>	OR062342	<i>L. africensis</i> : KM987391.1	99.84
55	Legon_1_2	Legon	<i>S. melongena</i>	OR062344	<i>L. africensis</i> : KM987391.1	99.84
56	Legon_1_3	Legon	<i>S. melongena</i>	OR062333	<i>L. africensis</i> : KM987391.1	99.84
57	Legon_1_4	Legon	<i>S. melongena</i>	OR062335	<i>L. africensis</i> : KM987391.1	99.84
58	Legon_1_5	Legon	<i>S. melongena</i>	OR062337	<i>L. africensis</i> : KM987391.1	99.84
59	Legon_1_6	Legon	<i>S. melongena</i>	OR062338	<i>L. africensis</i> : KM987391.1	99.84
60	Legon_1_7	Legon	<i>S. melongena</i>	OR062346	<i>L. africensis</i> : KM987391.1	99.84
61	Legon_1_8	Legon	<i>S. melongena</i>	OR062347	<i>L. africensis</i> : KM987391.1	99.84
62	Legon_2_1	Legon	<i>S. aethiopicum</i>	OR062343	<i>L. africensis</i> : KM987391.1	99.84
63	Legon_2_2	Legon	<i>S. aethiopicum</i>	OR062332	<i>L. africensis</i> : KM987391.1	99.84
64	Legon_2_3	Legon	<i>S. aethiopicum</i>	OR062334	<i>L. africensis</i> : KM987391.1	99.84
65	Legon_2_4	Legon	<i>S. aethiopicum</i>	OR062336	<i>L. africensis</i> : KM987391.1	99.84
66	Legon_2_6	Legon	<i>S. aethiopicum</i>	OR062339	<i>L. africensis</i> : KM987391.1	99.84
67	Legon_2_7	Legon	<i>S. aethiopicum</i>	OR062340	<i>L. africensis</i> : KM987391.1	99.84
68	Legon_2_8	Legon	<i>S. aethiopicum</i>	OR062341	<i>L. africensis</i> : KM987391.1	99.84
69	Nsawam_1	Nsawam	<i>S. aethiopicum</i>	OR062318	<i>L. africensis</i> : KM987391.1	99.84
70	Nsawam_2	Nsawam	<i>S. aethiopicum</i>	OR062319	<i>L. africensis</i> : KM987391.1	99.84
71	Nsawam_3	Nsawam	<i>S. aethiopicum</i>	OR062320	<i>L. africensis</i> : KM987391.1	99.84
72	Nsawam_4	Nsawam	<i>S. aethiopicum</i>	OR062321	<i>L. africensis</i> : KM987391.1	99.84
73	Nsawam_5	Nsawam	<i>S. aethiopicum</i>	OR062322	<i>L. africensis</i> : KM987391.1	99.84
74	Nsawam_8	Nsawam	<i>S. aethiopicum</i>	OR062323	<i>L. africensis</i> : KM987391.1	99.84
75	Senchi_1	Senchi	<i>S. aethiopicum</i>	OR064752	<i>L. africensis</i> : KM987391.1	99.84
76	Senchi_2	Senchi	<i>S. aethiopicum</i>	OR064753	<i>L. africensis</i> : KM987391.1	99.84
77	Senchi_3	Senchi	<i>S. aethiopicum</i>	OR064750	<i>L. africensis</i> : KM987391.1	99.84
79	Senchi_4	Senchi	<i>S. aethiopicum</i>	OR064749	<i>L. africensis</i> : KM987391.1	99.84

(Continued)

Table 2. (Continued.)

Sample number	Sample ID	Location	Crop sampled from	GenBank accession number	BLAST hit	% Similarity
79	Senchi_5	Senchi	<i>S. aethiopicum</i>	OR064748	<i>L. africensis</i> : KM987391.1	99.68
80	Senchi_6	Senchi	<i>S. aethiopicum</i>	OR064747	<i>L. africensis</i> : KM987391.1	99.84
81	Senchi_7	Senchi	<i>S. aethiopicum</i>	OR064746	<i>L. africensis</i> : KM987391.1	99.84
82	Senchi_8	Senchi	<i>S. aethiopicum</i>	OR064751	<i>L. africensis</i> : KM987391.1	99.84
83	Senchi_9	Senchi	<i>S. aethiopicum</i>	OR064754	<i>L. africensis</i> : KM987391.1	99.84
84	Adeiso_1_10	Adeiso	<i>S. aethiopicum</i>	OR058652	<i>L. laisalis</i> : KM987397.1	100
85	Legon_2_9	Legon	<i>S. aethiopicum</i>	OR058653	<i>L. laisalis</i> : KM987397.1	100

Thailand (accession number: LN624707.1), India (accession number: LN624690.1), Pakistan (accession number: LN624679.1) and Malaysia (accession number: LN624689.1). Likewise, the second cluster comprised the mitochondrial *COI* sequences of the *L. africensis* identified in this study, and those in eggplant fruits from Bangladesh (accession number: OL693251.1), Nigeria (KM987391.1). The third cluster comprised the sequences of the *L. laisalis* identified in eggplant fruits at Adeiso (Adeiso_1_10) and Legon (Legon_2_9) in southern Ghana, and those from Nigeria (accession number: KM987397.1), Kenya (accession number: KM987403.1) and South Africa (accession number: KM987697.1).

The neighbour-joining tree grouped all *Leucinodes* taxa into two major clades (I and II) with the exception of *L. malawiensis* (fig. 3). In the first clade, all *L. orbonalis* specimens clustered into one monophyletic clade (subclade A) consisting of two distinct groups. One group comprised a single Malaysian specimen (GenBank accession number: LN624689.1) and the second group formed a polytomy comprising specimens found in eggplant fruits in Bangladesh (GenBank accession number: LN624686.1), Pakistan (GenBank accession number: LN624679.1), India (GenBank accession number: LN624690.1) and Thailand (GenBank accession number: LN624707.1).

Likewise, *L. pseudorbonalis*, *L. rimavallis* and *L. africensis* specimens clustered into another monophyletic clade (subclade B). Within this monophyletic clade, three distinct groups were found. One group comprised *L. pseudorbonalis* specimen identified in fruits from Uganda (GenBank accession number: LN624707.1). The second group comprised *L. kenyensis* (GenBank accession number: KM987390.1) and *L. rimavallis* specimens (accession number: LN624678.1) identified in eggplant fruits from Kenya. Similarly, the third group comprised *L. africensis* specimens found in eggplant fruits in this study and *L. africensis* identified in eggplant fruits from Nigeria (GenBank accession number: KM987697.1) and Bangladesh (GenBank accession number: OL693251.1) respectively.

Notwithstanding, it is interesting to note that *L. africensis* specimens clustered into two distinct sub-groups. The first group formed a polytomy comprising all *L. africensis* specimens identified in this study and the reference specimen imported with fruits from Nigeria to Europe (GenBank accession number:

KM987697.1); and the second comprised a single specimen found in eggplant fruits in Bangladesh (GenBank accession number: OL693251.1).

Two groups were identified in the second major clade (clade II) containing all *L. laisalis* specimens. One group comprised all *L. laisalis* specimens identified in this study (GenBank accession numbers: OR058652.1 and OR058653.1), and reference specimens identified in fruits from Kenya (GenBank accession number: KM987403.1) and Nigeria (GenBank accession number: KM987397.1) (subclade C). However, the second group comprised a single specimen from South Africa (GenBank accession number: KM987697.1).

The morphological examination of the *L. africensis* and *L. laisalis* revealed similar and marked distinguishing features between the two species (fig. 4). Both species were found to possess white-coloured first abdominal segments. However, the remaining abdominal segments of the *L. africensis* were dark brown, compared to that of *L. laisalis*, which was light brown. Likewise, the ground colour of the forewings of the *L. africensis* was white with brown coloured half-moon-shaped patches and black patches at the wing tips, whilst that of the *L. laisalis* was light brown with brown coloured half-moon-shaped patches and dark brown patches at the wing tips (fig. 4).

Distribution and abundance of the EFSB species in southern Ghana

Leucinodes africensis was found in the shoot and fruits of eggplants on farmer's fields in all the study areas. However, *L. laisalis* was found in the shoot and fruits of eggplants on farmer's fields in only five study areas; Adeiso, Nsawam, Okorase and Senchi; and Legon in the Deciduous Forest and Coastal Savannah agroecological zones, respectively (fig. 5). Overall, the abundance of *L. africensis* was found to be higher than that of *L. laisalis* (table 3). Similarly, the percentage abundance of *L. africensis* was also higher than that of *L. laisalis* in all the study areas where both species occurred. The percentage abundance of the *L. africensis* identified in the shoot and fruits of eggplants on farmer's fields in southern Ghana was >90% in all the study areas, whereas that of *L. laisalis* was <10%.

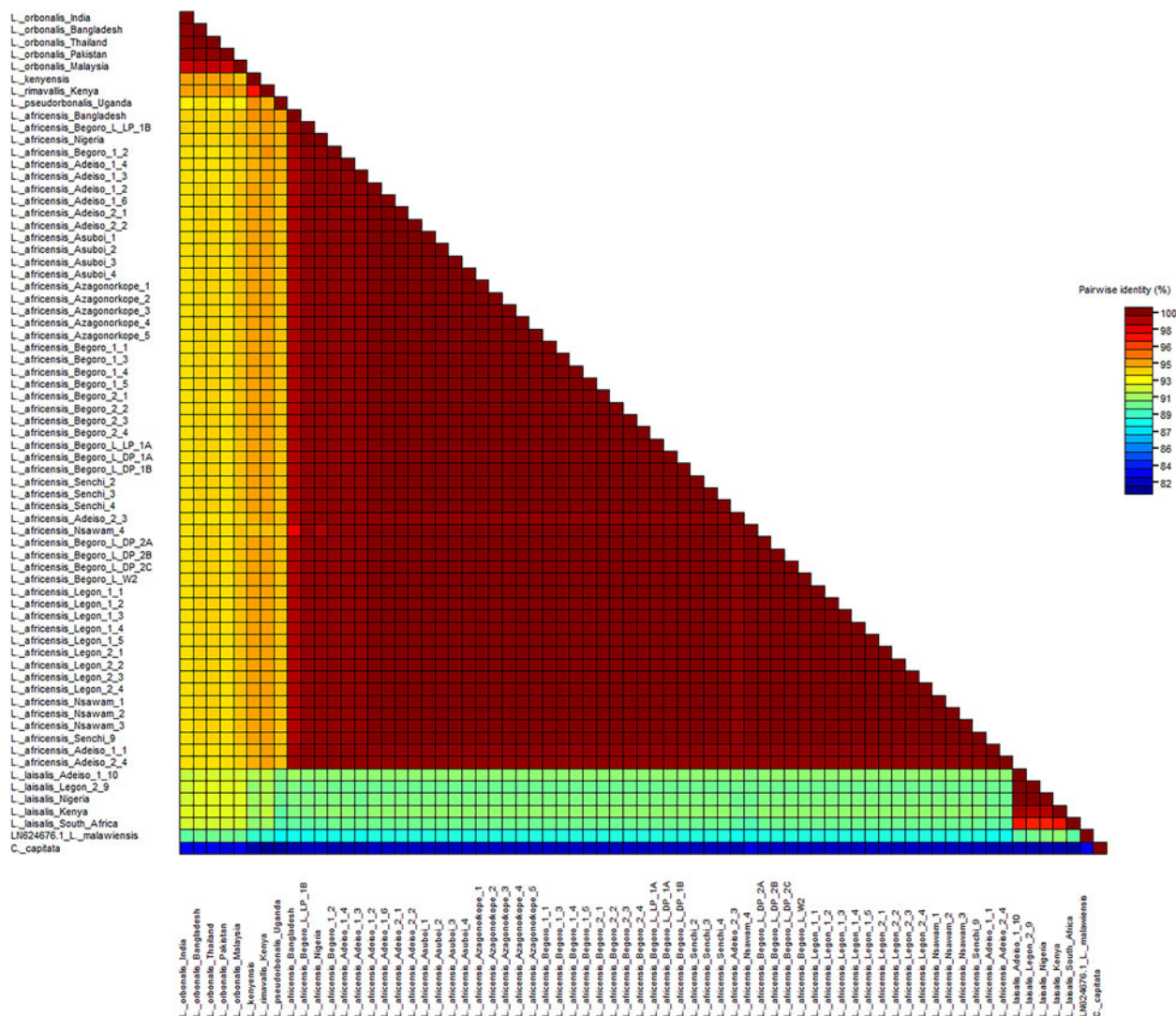


Figure 2. Pairwise sequence similarity of different *Leucinodes* species. Pairwise sequence comparison of *L. africensis* and *L. laisalis* identified in this study and reference sequences downloaded from GenBank: *L. orbonalis*, *L. africensis*, *L. laisalis*, *L. rimavallis*, *L. kenyensis*, *L. malawiensis* and *L. pseudorbonalis* was performed by aligning the sequences using MUSCLE algorithm and computing percentage similarity in SDT v 1.2 software. The intense crimson colour (as shown in the scale) indicated close similarity in the sequences and was more pronounced within species. Notably, there was a clear species demarcation between sequences from *L. africensis* and *L. laisalis* which had been identified in our study.

Monitoring of EFSB males in on-farm conditions

The *L. africensis* was the only EFSB identified in the pheromone traps mounted at Eric, Trosky, Tacks and Joekopan farms following a molecular and morphological taxonomic examination of the specimens.

The population of *L. africensis* males in eggplant fields at the exporter’s farms followed an irregular pattern from the vegetative to the maturity stage of the eggplants (fig. 6). Except for Joekopan farms, the number of *L. africensis* males remained stable at counts of zero from the 3rd to at least the 5th week after transplanting of the eggplants. Likewise, the number of *L. africensis* males peaked in the 4th, 9th, 11th and 14th weeks after transplanting the eggplants at Joekopan, Trosky, Tacks and Eric farms, respectively. Generally, the relative density of *L. africensis* males in eggplant fields at all the exporter’s farms was low (<2.00) (table 4). The highest relative density of *L. africensis* males was recorded at

Trosky farms, followed by Eric farms, Joekopan farms and Tacks farms.

Discussion

This study aimed to ascertain the identity of EFSB species (Lepidoptera: Crambidae) attacking eggplants on farmer’s fields in southern Ghana and monitoring the adult male population in on-farm conditions. Earlier studies by Frempong (1979), Owusu-Ansah *et al.* (2001), Mochiah *et al.* (2011), Ofori *et al.* (2015) and Ministry of Food and Agriculture (MOFA) (2022) noted the presence of the *L. orbonalis* on farmer’s fields in Ghana. However, this study did not detect any evidence of the *L. orbonalis* in the shoot and fruits of *S. aethiopicum* and *S. melongena* found on farmers’ fields in southern Ghana. Instead, *L. africensis* and *L. laisalis* were the only EFSB species identified

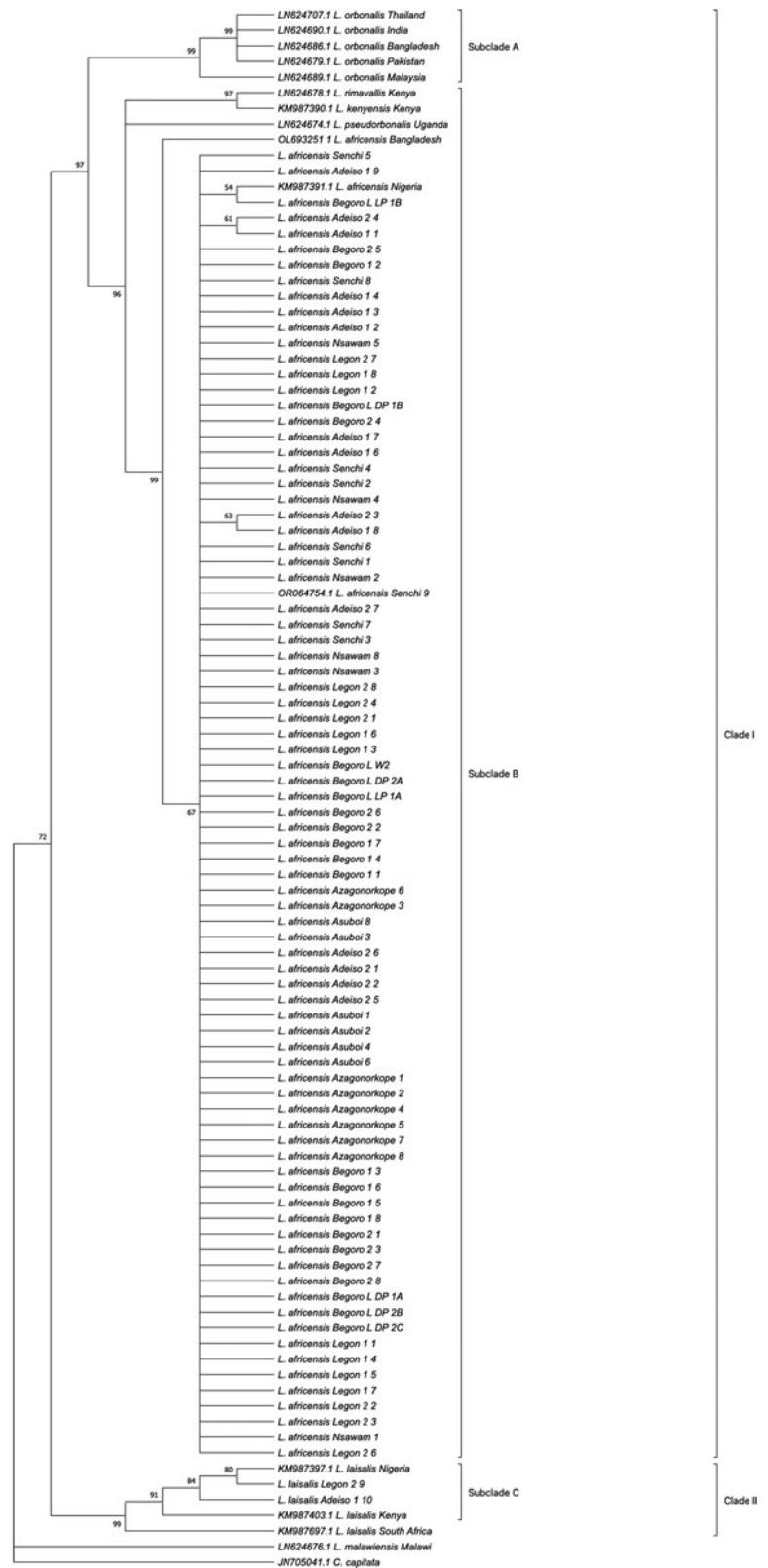


Figure 3. Phylogenetic analysis of *Leucinodes* species. Phylogenetic analyses to show the evolutionary relationship between *L. orbonalis*, *L. africensis*, *L. laisalis*, *L. malawiensis*, *L. pseudorbonalis*, *L. rimavallis* and *L. kenyensis* were constructed using the Neighbor-Joining method based on the Tamura-3 parameter in MEGA 11. There were two major clades (clade I and clade II). Notably, *L. africensis* and *L. laisalis*, which were of significant interest to this study, clustered in separate clades. Generally, there was a clear demarcation between the species whose representative sequences were used in the phylogenetic analyses. The mitochondrial *COI* sequence of the *C. capitata* was included as an outgroup.

on farmer's fields in southern Ghana, corroborating the findings of Mally *et al.* (2015), who identified the *L. africensis* and *L. laisalis* in intercepted eggplant fruits from Ghana.

Previous studies by Mally *et al.* (2015) and EPP0 (2023) have highlighted that *L. africensis* infests both *S. aethiopicum* and *S.*

melongena. Similarly, Boateng *et al.* (2005) documented the presence of *Sceliodes laisalis* (syn. *L. laisalis*) in *S. melongena* fruits in Ghana. However, there is limited literature on the occurrence of *L. laisalis* in *S. aethiopicum* fruits. Nevertheless, Mantey (2021) reported the presence of *L. laisalis* in *S. aethiopicum* fruits in

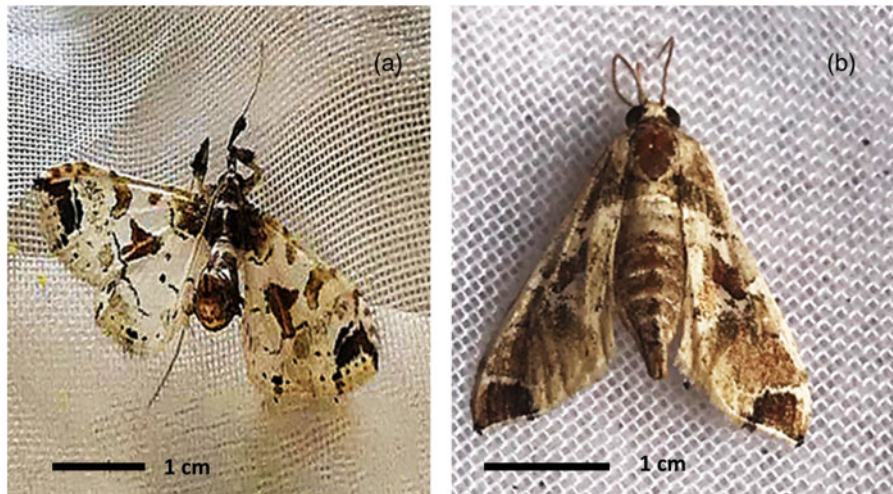


Figure 4. Eggplant fruit and shoot borer identified from fruits of eggplants. The adult insects were obtained by following the incubation of larvae-infested eggplant until the larvae pupated, and subsequently, adult forms emerged. The left panel (a) shows female *Leucinodes africensis*, while the right panel (b) shows female *Leucinodes laisalis*. Both species can readily be identified by the brown half-moon-shaped patches on their forewings and a white-coloured first abdominal segment. However, the *L. africensis* possess forewings with a white ground colour and its remaining abdominal segments being dark brown coloured, distinguishing it from the *L. laisalis*, which possesses forewings with a light brown ground colour, and remaining abdominal segments that are also light brown.

eggplant fields at Legon in southern Ghana in an unpublished thesis. The findings of this study support the report by Mantey (2021) and provide formal confirmation of the presence of *L. laisalis* in *S. aethiopicum* fruits in eggplant hotspots in southern Ghana.

The presence of *L. africensis* and *L. laisalis* in the shoots and fruits of both *S. aethiopicum* and *S. melongena* in southern

Ghana has significant implications for the bilateral trade of eggplants between Ghana and European Union (EU) Member States. These implications may also extend to the international trade of other Solanaceae plants, such as *Capsicum annum* and *Solanum lycopersicum*, between Ghana and EU Member States, as these crops have been reported as host plants for these pest species (Mally *et al.*, 2015; EPPO, 2023). The implications include but

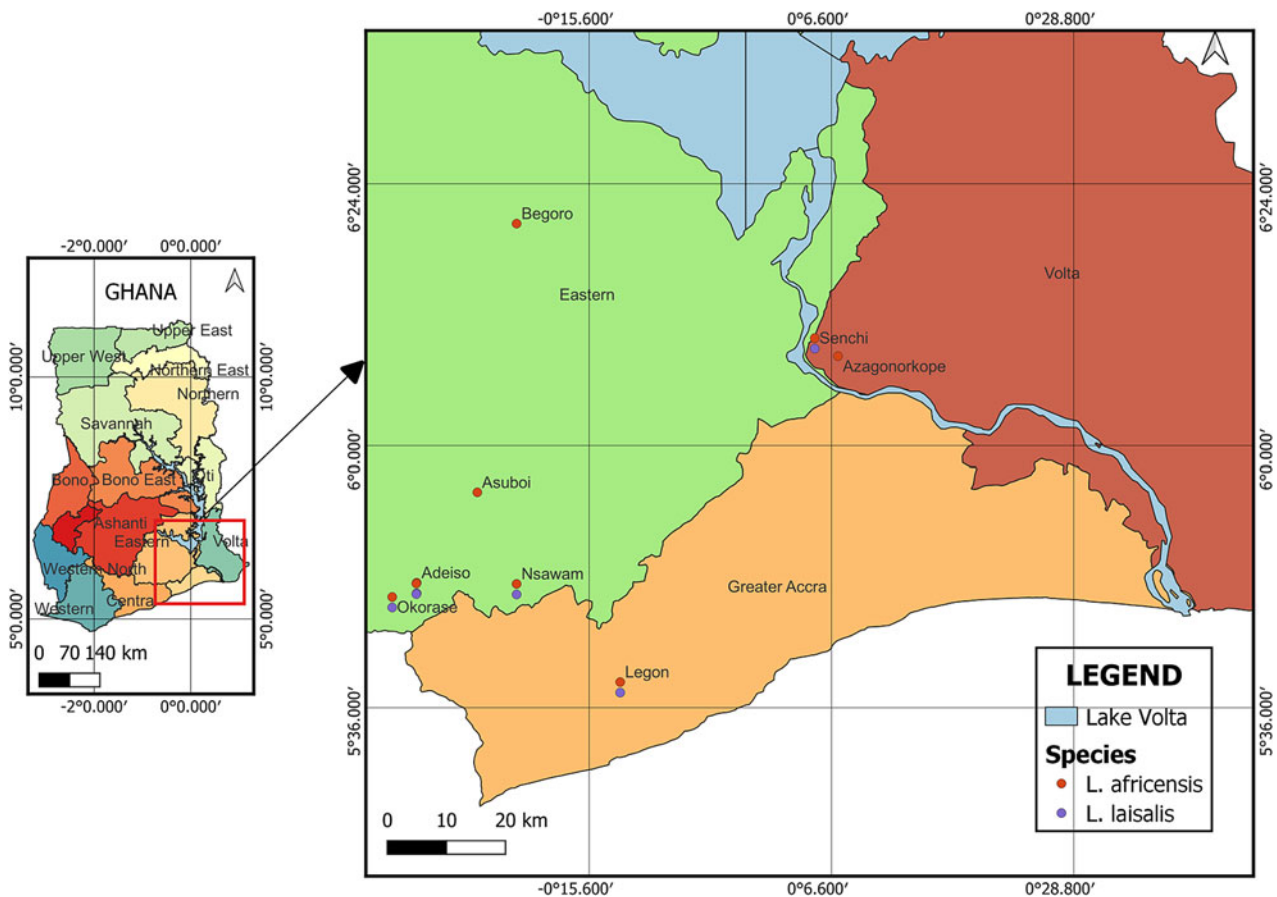


Figure 5. Distribution of the *L. africensis* and *L. laisalis* in southern Ghana. This study revealed that both *L. africensis* (red dots) and *L. laisalis* (purple dots) were present in southern Ghana. We did not detect any *L. laisalis* in Begoro, Asuboi and Azagonorkope. The complete map of Ghana on the left shows the regional political boundaries in different colours, and the inserted red box demarcates the geographic region where our study was conducted.

Table 3. Percentage (%) abundance of the *L. africanis* and *L. laisalis* in the shoot and fruits of eggplants at different locations in southern Ghana

Study area	Sampling site	EFSB sampled from eggplants	Abundance	% Abundance for each study area
Adeiso	*Eric + Trosky Farms	<i>L. africanis</i>	171	92.4
		<i>L. laisalis</i>	14	7.6
Asuboi	Eggplant field	<i>L. africanis</i>	91	100.0
Azagonorkope	*Joekopan Farms	<i>L. africanis</i>	57	100.0
Begoro	Exporter's farms	<i>L. africanis</i>	44	100.0
Nsawam	Eggplant field	<i>L. africanis</i>	68	93.2
		<i>L. laisalis</i>	5	6.8
Okorase	Eggplant field	<i>L. africanis</i>	69	93.2
		<i>L. laisalis</i>	5	6.8
Senchi	*Tacks Farms	<i>L. africanis</i>	121	99.2
		<i>L. laisalis</i>	1	0.8
Legon	UG Farm	<i>L. africanis</i>	103	90.4
		<i>L. laisalis</i>	11	9.6

*Exporter's farms.

are not limited to interception of produce and, in the absence of robust phytosanitary measures, could negatively impact the export of these Solanaceae crops from Ghana to the EU Member States, a situation that has occurred before in Ghana. The first local ban on the export of eggplants and other crops within the Solanaceae family was issued by the Plant Protection and Regulatory Service Directorate (PPRSD) of the Ministry of Food and Agriculture (MOFA) in September 2011 due to high interceptions of the *L. orbonalis* (now referring to individuals within the *Leucinodes* genus native to Africa (EFSA *et al.*, 2021)) and *Thrips* spp. (European Commission Health and Consumers Directorate-General, 2012); while the EU ban, also due to high interceptions of harmful organisms, including possibly the misidentified *L. orbonalis*, was issued by the EU in October 2015 and extended to December 2017 (Fening and Billah, 2019a, 2019b).

It is interesting to note that in the neighbour-joining tree, *L. orbonalis*, *L. pseudorbonalis*, *L. kenyensis*, *L. rimavallis* and *L.*

africanis clustered together into one clade, while *L. laisalis* clustered into another clade. This suggests that *L. orbonalis*, *L. pseudorbonalis*, *L. kenyensis*, *L. rimavallis* and *L. africanis* are more closely related to each other than to *L. laisalis* as far as mitochondrial COI gene is concerned. The present finding is broadly consistent with the findings of Mally *et al.* (2015), who also demonstrated that *L. orbonalis*, *L. africanis*, *L. rimavallis*, *L. pseudorbonalis* and *L. kenyensis* clustered together in one clade, while the *L. laisalis* and *L. malawiensis* clustered together in another clade.

The morphological examination of the *L. africanis* and *L. laisalis* showed that both species possessed brown-coloured half-moon-shaped patches in theirs and a white-coloured first abdominal segment. These features have been reported to be characteristic of species found in the *Leucinodes* genus (Mally *et al.*, 2015). Additionally, differences were observed in the ground colour of the forewings and remaining abdominal segments of the *L. africanis* and *L. laisalis*; this is in concurrence with the reports

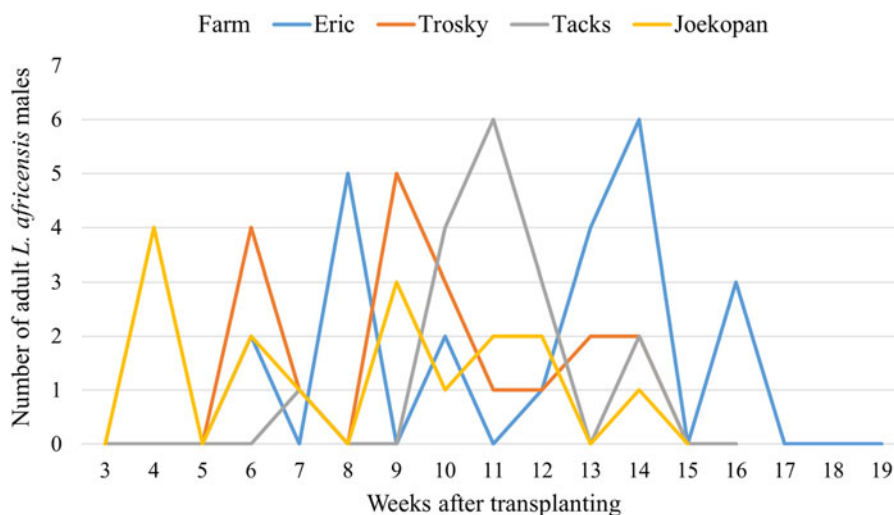


Figure 6. Weekly trap catches of adult *L. africanis* males at Eric, Trosky, Tacks and Joekopan farms. This study revealed that adult *L. africanis* males were attracted to the sex pheromone lure of the *L. orbonalis* as this EFSB was the only species found in the delta pheromone traps mounted on farmer's fields. The population of adult *L. africanis* males followed an irregular pattern from the vegetative to the maturity stage of the eggplants in the farmers' fields, peaking in the 4th, 9th, 11th and 14th weeks after transplanting of the eggplants at Joekopan, Trosky, Tacks and Eric Farms, respectively.

Table 4. Comparison of relative density of adult *L. africensis* males based on weekly trap catches

Exporter's farm	Relative density (F/T/W) *
Eric farms	1.35
Trosky farms	1.46
Tacks farms	1.14
Joekopan farms	1.23

*F = the total number of *L. africensis* males captured, T = the number of pheromone traps inspected and W = the number of weeks pheromone traps were exposed in the eggplant fields.

made by Mally *et al.* (2015). Mally *et al.* (2015) reported that the ground colour of the forewings and remaining abdominal segments of the *L. africensis* was white and ranged from brown to grey, respectively, whilst that of the *L. laisalis* had colours ranging from orange-brown to greyish-white, and brown, respectively. Notwithstanding, Mally *et al.* recommended the use of male genitalia as another diagnostic feature to accurately distinguish between *L. africensis* and *L. laisalis*. The male genitalia of adult *L. africensis* has a long ventrad fibula; an elongated, strong-hooked or straight shaped, sometimes branching distal sacculus process that is projected towards the valva apex; an apically thin juxta; and a posterior phallus with an oval saw blade-shaped sclerotisation. However, the male genitalia of adult *L. laisalis* has a large and oval sacculus; a ventrad fibula that is broad and strongly sclerotised; well elongated saccus; and a phallus that has a keeled coecum and slim, fingerlike and strongly sclerotised apoderme.

Considering the technical knowledge involved in the use of male genitalia to distinguish between moth species in general, the differences found in the ground colour of the forewings and abdominal segments between adult *L. africensis* and *L. laisalis* could be helpful to farmers in their identification during pest monitoring activities on their farms, which can inform decision-making on the management of the infestations of the *L. africensis* and *L. laisalis*. Hence, the extension staff of the PPRSD of MOFA is encouraged to educate farmers on these diagnostic features during focus discussion sessions with farmer groups.

Leucinodes africensis was detected in all the study areas in southern Ghana, and it was found to coexist with *L. laisalis* in some of these areas. This indicates that the study areas provide favourable conditions for the establishment of these *Leucinodes* species. Moreover, the presence of both *L. africensis* and *L. laisalis* suggests their adaptability to the various environmental conditions in the study areas. The climatic conditions in these areas are predominantly hot and humid throughout the year, with an average annual temperature of 26.1 °C for locations near the coast (Ministry of Food and Agriculture (MOFA), 2018). This observation suggests that *L. africensis* and *L. laisalis* are moths that thrive in warm and humid environments.

Leucinodes africensis was found to have a wider distribution and greater dominance in southern Ghana compared to *L. laisalis*. This aligns with reports by Mally *et al.* (2015) highlighting the widespread presence of *L. africensis* in Africa among other *Leucinodes* species native to the continent. Additionally, Pace *et al.* (2022) reported frequent interceptions of *L. africensis* in exported eggplant fruits from Ghana. Interestingly, despite using an *L. orbonalis* sex pheromone lure, there was no evidence of adult *L. orbonalis* males in the pheromone traps installed on exporter's farms. Instead, only adult *L. africensis* males were

observed through molecular and morphological taxonomic examination. This provides further evidence that *L. orbonalis* may not be present in Ghana as previously described and suggests the possibility of interspecific pheromone attraction among *Leucinodes* species. While limited information is available on interspecific pheromone attraction among *Leucinodes* species, it is plausible that there are similarities in the components of sex pheromones released by adult females of *L. africensis* and *L. orbonalis*, considering their close relationship. However, it is essential to exercise caution in interpreting this finding, as factors other than those suggested may influence the behavioural responses of *L. africensis* to the *L. orbonalis* sex pheromone lure. Nevertheless, the attraction of *L. africensis* to the sex pheromone lures of *L. orbonalis* can be utilised as a population suppression tool for managing *L. africensis* in eggplant production in Ghana.

There was variation in the number of adult *L. africensis* males among the exporter's farms and the peak period of the adult *L. africensis* males. This is attributed to the variation in the growth stage of the eggplant and climatic conditions among the farms (McNeil, 1991; Rhino *et al.*, 2010). The EFSB has been reported by many studies to be present on eggplant fields in all the growth stages of eggplant, with their numbers varying throughout the lifecycle of eggplants. For instance, Ofori *et al.* (2015) reported the presence of EFSB previously reported as *L. orbonalis* in all the growth stages of eggplants in Ghana. Similarly, Taiwo *et al.* (2020) demonstrated the variation in numbers of EFSB previously reported as *L. orbonalis* at different weeks after transplanting of eggplants and in each growth stage. This has been attributed to the production of secondary metabolites in leaves, shoot, flowers and fruits of eggplants, whose levels vary throughout the lifecycle of eggplants and serve as kairomones for adult EFSB. For instance, Nusra *et al.* (2021) demonstrated that the production of secondary metabolites such as benzyl alcohol, 2,2'-(ethane-1,2-diylbis(oxy)) bis(ethane-2-1-diyl) dibenzoate and 3,7-dimethylocta-1,6-dien-3-ol as major constituents of leaves, flowers, fruits and shoots of eggplants, respectively, serves as kairomones for *L. orbonalis*. During the survey of farmer's fields in this study, a variation in planting dates of eggplants and cultivation period among farmer fields was observed, resulting in variation in onset and duration of growth stages of eggplants among farmer fields. This may explain the difference in the number and peak period of adult *L. africensis* males among the exporter's farms.

Similarly, climatic conditions such as temperature and rainfall have an effect on the development and survival of moths. Among the climatic conditions that affect the development and survival of moths, temperature and rainfall have a significant relationship with moth abundance in the tropics (Kato *et al.*, 1995; Intachat *et al.*, 2001; Brehm *et al.*, 2007), as revealed by Choi (2008). In this study, the population of adult male moths was monitored in farmer fields in the eastern and volta (found in south-eastern coastal area) regions of southern Ghana. Both regions experience predominantly warm and humid conditions; however, the eastern belt is comparatively warmer (Ministry of Food and Agriculture (MOFA), 2018). Likewise, both regions experience bimodal rainfall every year (Ministry of Food and Agriculture (MOFA), 2018). However, there is variability in rainfall amounts; with the eastern belt experiencing more rainfall than the south-eastern coastal areas (Braimah *et al.*, 2022). The variation in these conditions (especially temperature, humidity and rainfall) among the farms could have influenced the variation in the numbers and peak periods of adult *L. africensis* males.

The relative density of the adult *L. africensis* males recorded in all the exporter's farms was low, and this is attributed to the effectiveness of the management practices recommended by the PPRSD of MOFA in its roadmap to manage populations of the *L. orbonalis* (now referring to *Leucinodes* spp. native to Africa) (European Commission Health and Consumers Directorate-General, 2012) as the farmers adhered to this management protocol. The management practices recommended by the PPRSD of MOFA in its roadmap to manage populations of the misidentified *L. orbonalis* included on-farm sanitation, that is, proper disposal of rotten eggplant fruits, use of pheromone traps and application of selective insecticides (Fening et al., 2017). The extension staff of the PPRSD of MOFA is therefore encouraged to regularly visit exporters' farms to ensure that farmers adhere to these management practices to increase yield and revenues obtained from exports of eggplants.

In conclusion, this paper presents evidence that challenges the previous description of *L. orbonalis* presence in Ghana. Through identification efforts, it was determined that *L. africensis* and *L. laisalis* are the only species attacking eggplants on farmer's fields in southern Ghana. One notable finding is that *L. africensis* males were attracted to the sex pheromone lures of *L. orbonalis*, despite the species being distinct. This suggests the potential use of *L. orbonalis* sex pheromone lures as a tool to suppress *L. africensis* populations in eggplant fields. Further investigation and experimentation in this area are strongly recommended. The management protocol implemented by the PPRSD of MOFA in Ghana, aimed at managing *L. orbonalis* populations (now referred to as African *Leucinodes* spp.), was found to be effective, resulting in low numbers of *L. africensis* on exporter's farms. Eggplant farmers are therefore encouraged to adhere to this management protocol.

Supplementary material. The supplementary material for this article can be found at <https://doi.org/10.1017/S0007485324000154>.

Acknowledgements. The authors are very grateful for the financial support from GCRF AgriFood Africa Innovation Awards Round 3 grant awarded to Dr Francis Wamonje and Professor Ken Fening. We are also grateful to farmers from the Ghana Association of Vegetable Exporters (GAVEX) who offered their farms for the study.

Competing interests. None.

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