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Record of *Etiella zinckenella* (Treitschke) affecting soybeans in Itapúa, Paraguay

Registro de *Etiella zinckenella* (Treitschke) afectando cultivo de soja en Itapúa, Paraguay

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ABSTRACT: During the 2021/2022 soybean production season in Itapúa, Paraguay, the presence of *Etiella zinckenella* (Treitschke) caterpillars affecting soybean pods in the country was documented. The observations were made amidst a widespread drought that adversely affected soybean production. Caterpillars were collected from pods that exhibited signs of pest entry. Morphological characterization of the collected specimens



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matched the descriptions provided in taxonomic keys for *E. zinckenella* larvae. Molecular identification through DNA extraction and sequencing confirmed the identity of the pest as *E. zinckenella*, exhibiting a 96% similarity with known accessions of the species in the GenBank database. As a challenging pest to control, efforts should be directed towards evaluating the economic and damage thresholds caused by *E. zinckenella*, assessing resistance in soybean varieties used in Paraguay, and investigating the efficacy of biological and chemical insecticides. These findings can contribute to the development of integrated management strategies for effectively mitigating the impact of this pest on soybean production in Paraguay.

Keywords: entomology; *Glycine max*, harvesting, pests.

RESUMEN: Durante la temporada de producción de soja 2021/2022 en la región de Itapúa, Paraguay, se documentó la presencia de orugas *Etiella zinckenella* (Treitschke) afectando las vainas de soja en el país. Las observaciones se realizaron en medio de una sequía generalizada que afectó negativamente a la producción de soja. Las orugas se recolectaron de las vainas que mostraban signos de entrada de plagas. La caracterización morfológica de los especímenes recolectados coincidió con las descripciones proporcionadas en claves taxonómicas para las larvas de *E. zinckenella*. La identificación molecular a través de la extracción y secuenciación de ADN confirmó la identidad de la plaga como *E. zinckenella*, exhibiendo un 96% de similitud con las accesiones conocidas de la especie en la base de datos *GenBank*. Como plaga difícil de controlar, los siguientes esfuerzos deben dirigirse a evaluar los umbrales económicos y de daño causados por *E. zinckenella*, evaluar la resistencia en las variedades de soja utilizadas en Paraguay e investigar la eficacia de los insecticidas biológicos y químicos. Estos hallazgos pueden contribuir al desarrollo de estrategias de manejo integrado para mitigar de manera efectiva el impacto de esta plaga en la producción de soja en Paraguay.

Palabras clave: cosecha, entomología, *Glycine max*, plaga.

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INTRODUCTION

The pod borer *Etiella zinckenella* (Treitschke) (Lepidoptera: Pyralidae) is considered a cosmopolitan pest. This species feeds mainly on fabaceae seeds, including peas (*Pisum sativum*), common bean (*Phaseolus vulgaris*) and soybean (*Glycine max*)⁽¹⁾.

The population of *E. zinckenella* increases in dry and warm seasons, which also allows a higher incidence in crops⁽²⁾. Furthermore, this pest can survive during the winter on host crops such as *Crotalaria* sp.⁽³⁾.

In addition, *E. zinckenella* is considered a difficult-to-control pest when soybeans are in the pod stage, where insecticide applications are made without considering their effectiveness or economic thresholds ⁽⁴⁾. When the attacks of *E. zinckenella* to the soybean crop are severe, the losses can reach to 80%, when control tactics of this pest are not carried out ⁽⁵⁾.

In Paraguay, this pest was recorded in Villarrica in 1956⁽⁶⁾ and later a mention was made of its presence in Paraguay⁽⁷⁾, so the aim of this work was to document the record of this insect attacking soybean crops in Itapúa department, Paraguay.

During the 2021/2022 soybean production season, the presence of *E. zinckenella* caterpillars was observed in Itapúa (Paraguay) fields (Table 1). This season was characterized by the occurrence of a generalized drought in the country, which affected soybean production⁽⁸⁾. Caterpillars were collected from individual pods of soybean plants previously identified with pest entry holes (Figure 1). Subsequently, they were preserved in 70% ethanol in the Multiple Uses Laboratory of the Technological Development and Innovation Center (CEDIT) in Natalio, Itapúa (Paraguay), where morphological characterization was carried out using taxonomic keys of immature stages of the Pyralidae family⁽⁹⁾ and keys to immature stages of lepidoptera⁽¹⁰⁾.

Table 1. Location and date of observation of *Etiella zinckenella* in soybean fields in

Location	Geographical coordinates		Observation date
	S	W	
Bella Vista	27°02'07.3"	55°31'38.5"	10/12/2021
Capitán Meza	26°52'11.2"	55°18'53.9"	10/12/2021
Edelira	26°43'50.4"	55°17'14.1"	15/12/2021
Natalio	26°40'33.7"	55°07'53.9"	20/01/2022
Pirapó	26°46'38.3"	55°29'35.5"	10/12/2021
San Rafael del Paraná	26°32'04.4"	54°56'40.4"	11/02/2022
Tomás Romero Pereira	26°34'49.5"	55°14'09.0"	11/02/2022

Itapúa, Paraguay.



Figure 1. Damage caused by *Etiella zinckenella*. (a) perforated pods; (b, c) soybeans with damage and caterpillar feces; (d) *Etiella zinckenella* caterpillar feeding.

The DNA extraction and molecular identification were carried out in the Molecular Biology Laboratory of the Faculty of Science and Technology at the National University of Itapúa, Encarnacion, Itapúa, Paraguay. The larvae morphology detected in soybean pods and seeds concord with that described by other researchers^(9, 10, 11, 12) for *E. zinckenella* larvae, presenting the eighth-grade blowhole, abdominal segment at the same level as the previous ones, three ventral setae can be seen from the third to the sixth abdominal segment, a bright ring around the SD1 bristle on the

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eighth abdominal segment, with three setae on the lateral group of the ninth abdominal segment and thoracic plate with a characteristic pattern for the species (Figure 2).

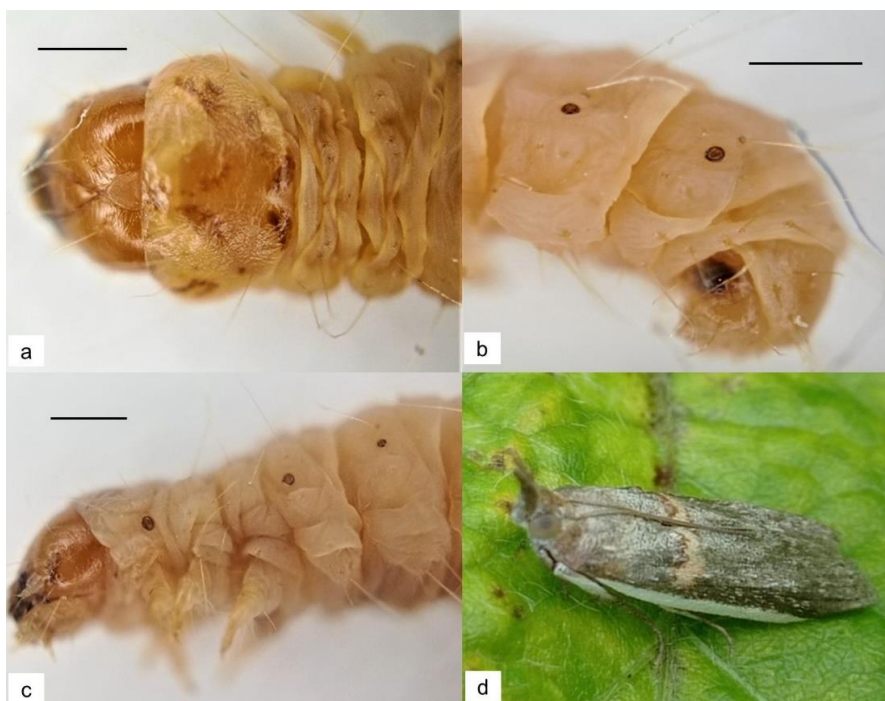


Figure 2. Morphological characteristics of *Etiella zinckenella* observed in Paraguayan soybean fields. (a) Chest plate; (b) Lateral detail of the last abdominal segments; (c) Lateral view of the head and thorax; (c) Adult captured in soybean culture. Bars = 1mm.

Genomic DNA was extracted from the thorax and head of one adult, with The Wizard[®] Genomic DNA Purification Kit (Promega). To verify the

identity, the barcoding region of the mitochondrial gene cytochrome oxidase 1 (CO1) was amplified with the following primers: LepF1: ATTCAACCAATCATAAAGATATTGG and LepR1: TAAACTTCTGGATGTCCAAAAAATCA⁽¹³⁾. The resultant amplicons were sequenced at Macrogen (Seoul, Republic of Korea), and the consensus sequence was compared with the GenBank database with the Blastn tool. Obtaining a consensus sequence of 599 bp, which presented a high percentage of identity (96%) with accesses of the species *E. zinckenella*.

This is the first record of *E. zinckenella* affecting pods in soybeans fields in Itapúa, Paraguay. Future investigations could focus on evaluating the loss caused by this pest to determine economic and damage thresholds, as well as determining resistance in the varieties used in Paraguay and evaluating the effectiveness of biological and chemical insecticides with the aim of making proposals for their management integrated from this plague.

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AUTHOR CONTRIBUTIONS

JAST conceived the research and designed the sampling, collected the field data and carried out the morphometric studies; LNTS developed the laboratory methodology and analyzed the molecular data; CNBC carried out the investigation, specifically, processing the samples in the laboratory; JYMF carried out the research, specifically processing the samples in the laboratory, also analyzed molecular data; GAEM supervised and led the planning and execution of the research, also prepared the published work, specifically writing the initial draft (including a substantial translation). All authors reviewed and approved the final version of the manuscript.

DECLARATION OF CONFLICTS OF INTEREST

The authors declare that they have no conflict of interest with respect to this research article.

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