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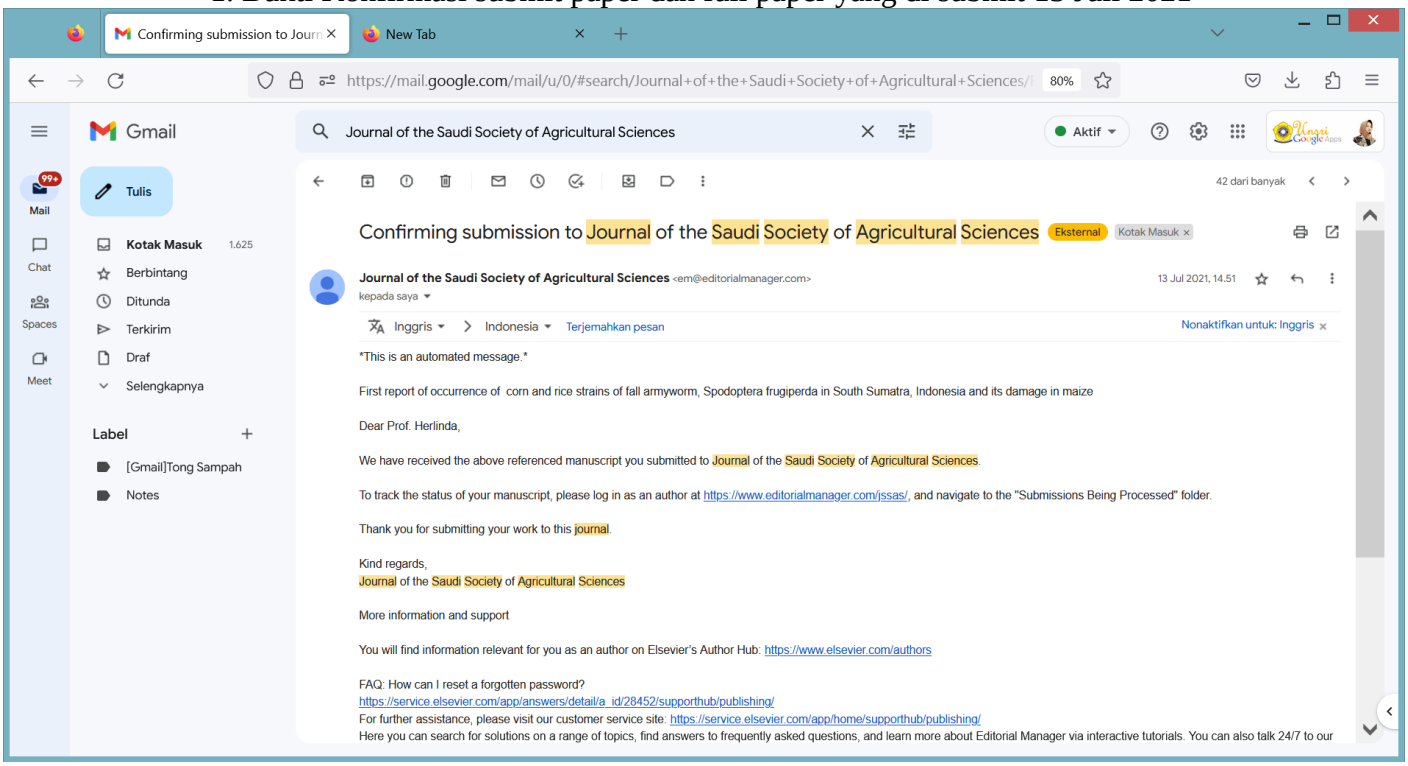

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First report of occurrence of corn and rice strains of fall armyworm, Spodoptera frugiperda in South Sumatra, Indonesia and its damage in maize

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Abstract:	Spodoptera frugiperda is a new invasive pest in Indonesia and its severity in maize ranges from 26.50 to 100%. However, information on the strains or genetic diversity of the S. frugiperda in Indonesia is still very limited. This research aimed to identify the genetic diversity of S. frugiperda from South Sumatra and determine its damage in maize. Surveys from January to June 2021 were carried out from the lowlands to highlands of South Sumatra. The observation of damage was carried out directly using a scouting system. The S. frugiperda larvae was identified based on morphological characters and molecular techniques using sequence analysis of Cytochrome c Oxidase subunit I (COI) gene. All larvae collected from South Sumatra showed identical morphological characteristics identified as S. frugiperda . The sequence analysis results showed that the 6 isolates of S. frugiperda shared 100% of sameness as the rice strain haplotype 1, S. frugiperda isolate from Lampung Province. The other 3 isolates of S. frugiperda shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan). All isolates have been deposited in the GenBank. This study confirmed the presence of rice and corn strains of Spodoptera frugiperda and this is the first report of the occurrence of both strains in South Sumatra. we also found that outbreaks of S. frugiperda have occured in the South Sumatra. The incidence and severity of S . frugiperda reached 100% and 65% respectively. Comprehensive further study should be performed to confirm the presence of both strains and their damage in all corn producing areas in Indonesia.
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Cover Letter

Palembang, 13 July 2021

Chief Editor
Journal of the Saudi Society of Agricultural Sciences

Dear Editor,
We wish to submit an article entitled, "First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize" for intended publication in Journal of the Saudi Society of Agricultural Sciences for your kind consideration.

We declare that this manuscript has not been published and is not under consideration for publication to any other journal or any other type of publication (including web hosting) either by me or any of my co-authors. All authors have been read and agreed to the Ethical Guidelines. I also certify that all the authors have approved the paper for release and are in agreement with its content.

This study highlights findings that first report of the occurrence of rice and corn strains of *Spodoptera frugiperda* in South Sumatra, Indonesia and we also found that outbreaks of *S. frugiperda* have occured in the South Sumatra.

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Thank you for your consideration of this manuscript.

Sincerely,

Prof. Dr. Siti Herlinda

First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

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Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

All data are available in the article and the materials used in this work are of high quality and grade.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Authors' contributions

SH performed research concept and design, writing the article, and final approval of article. RS prepared and performed molecular identification and data analysis and interpretation. MES and

FF performed collection and assembly of data. SS prepared and performed morphological identification and critical revision of the article. All the authors read and approved the manuscript.

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First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

ABSTRACT

Spodoptera frugiperda is a new invasive pest in Indonesia and its severity in maize ranges from 26.50 to 100%. However, information on the strains or genetic diversity of the *S. frugiperda* in Indonesia is still very limited. This research aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize. Surveys from January to June 2021 were carried out from the lowlands to highlands of South Sumatra. The observation of damage was carried out directly using a scouting system. The *S. frugiperda* larvae was identified based on morphological characters and molecular techniques using sequence analysis of Cytochrome c Oxidase subunit I (*COI*) gene. All larvae collected from South Sumatra showed identical morphological characteristics identified as *S. frugiperda*. The sequence analysis results showed that the 6 isolates of *S. frugiperda* shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province. The other 3 isolates of *S. frugiperda* shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan). All isolates have been deposited in the GenBank. This study confirmed the presence of rice and corn strains of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra. we also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. The incidence and severity of *S. frugiperda* reached 100% and 65% respectively. Comprehensive further study should be performed to confirm the presence of both strains and their damage in all corn producing areas in Indonesia.

Keywords: genetic diversity; incidence; outbreaks; scouting; severity

1. Introduction

Fall armyworm (FAW), *Spodoptera frugiperda* is a new invasive maize pest in Indonesia. This insect pest comes from the American continent (Nagoshi et al., 2017; Otim et al., 2018). In 2016, the FAW was reported to have come into Africa (Goergen et al., 2016). In 2017, FAW crossed over to Europe (Early et al., 2018). This pest began to move into Asia in 2018 (Mahat et al., 2021) and was first discovered in India (Ganiger et al., 2018) and came into Indonesia for the first time on March 26, 2019 in West Sumatra (Sartiami et al., 2020). Then, it began to spread to other provinces

and islands in Indonesia, such as South Sumatra (Hutasoit et al., 2020), West Java (Maharani et al., 2019), Lampung (Trisyono et al., 2019), Bengkulu (Ginting et al., 2020), Bali(Supartha et al., 2021).

In addition to spreading throughout the world, the FAW has caused maize yield losses of up to 18 million tons/year and losses of up to 13 million US$ in 12 African countries (Harrison et al., 2019). In Kenya, the loss due to this pest reaches 1 million ton/year (De Groote et al., 2020). Besides attacking the maize, this pest attacks paddy, sugarcane, cotton, and ornamental plants (IPPC, 2019). In Brazil it has been reported that about 76 plant families were destroyed by this pest (Montezano et al., 2018). In Indonesia, the FAW generally attacks maize with damage in Lampung ranging from 26.50% to 70% (Lestari et al., 2020), in Bali reaching 47.84% (Supartha et al., 2021), in East Nusa Tenggara ranging from 85% to 100% (Mukkun et al., 2021). This pest can also attack paddy leaves, for example, in Banten the FAW larvae was found to attack paddy (Sartiami et al., 2020); yet there is no information on damage by this pest to paddy in Indonesia.

There are two strains of *S. frugiperda* in the world, namely corn strain (C) and rice strain (R) (Unbehend et al., 2013; Va et al., 2014). The genetic diversity of *S. frugiperda* in Indonesia was first reported by Sartiami et al. (2020) stating that the strain of *S. frugiperda* found in Banten was only the rice strain. In Lampung, the strain of *S. frugiperda* found was only the corn strain (Lestari et al., 2020), while in West Sumatra both the corn and rice strains were found (Nelly et al., 2021). The corn and rice strains of *S. frugiperda* if they have spread in Indonesia can harm not only maize but also paddy and other important crops. However, information on the strains/genetic diversity of *S. frugiperda* in Indonesia is still very limited and until now the information on the strain of *S. frugiperda* originating from South Sumatra and its attack has not been reported. For this reason, information on the genetic diversity of the FAW in South Sumatra and its attacks is needed so that it can be used as a basis for controlling this pest and can complement information on *S. frugiperda* strains in Indonesia. This study aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize.

2. Materials and methods

2.1. Survey sites

Surveys to obtain specimens of *S. frugiperda* larvae were carried out from the lowlands to the highlands of South Sumatra, such as Palembang City (2°59′27.99″S 104°45′24.24″E), Pagar Alam City (3°52′43.8″S 103 °21′30″E), Lahat City (3.78639°S 103.54278°E), Ogan Ilir District

(3.43186°S 104.6727°E), Prabumulih City (3.4328°S 104.2356°E), Muara Enim District (4.2327°S 103.6141 °E), and Banyuasin District (2.8833°S 104.3831°E) (Fig. 1 and Table 1). The survey started from January to June 2021. The specimens obtained were then identified molecularly at the Laboratory of Agricultural Biotechnology (accredited according to the ISO 17025 standard), Department of Plant Protection, Faculty of Agriculture, Lampung University, Indonesia.

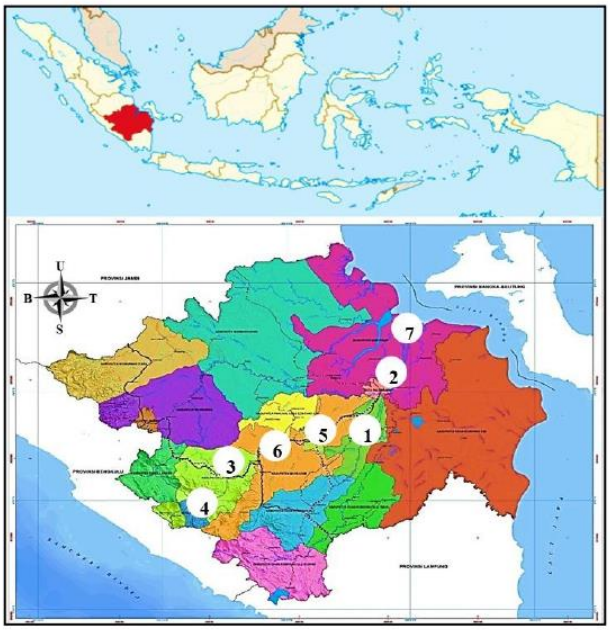

Fig. 1. Surveys locations in South Sumatra, Indonesia: Ogan Ilir District (1), Palembang City (2), Lahat District (3), Pagar Alam City (4), Prabumulih City (5), Muara Enim District (6), dan Banyuasin District (7).

2.2. Morphological identifications for Spodoptera frugiperda

S. frugiperda larvae were collected from maize fields in various districts/cities in South Sumatra, Indonesia. The larvae were brought to the Entomology Laboratory, Department of Plant Protection, Faculty of Agriculture, Universitas Sriwijaya, Indonesia to be reared individually in porous plastic cups (Ø 6.5 cm, height 4.6 cm). Maize leaves (2 cm x 5 cm) were placed into the cup to feed *S. frugiperda* and then the larvae were observed for further identification. Some samples of larvae were put into vials containing 70% alcohol for molecular identification at the Laboratory of Agricultural Biotechnology, Department of Plant Protection, Faculty of Agriculture, Universitas Lampung. Some other samples of larvae from the same population were kept until the adult completed one life cycle following the method of Gustianingtyas et al. (2021) to observe the morphology of the adult.

Table 1.

The origin of sample of *Spodoptera frugiperda* from South Sumatra, Indonesia.

Location (village, district/city)	Coordinate	Sample ID	Sample source	Species	GenBank Acc. No.
Alang-alang Lebar, Palembang City	2°59'27"S 104°45'24"E	AaFaw	Maize	*Spodoptera frugiperda*	MZ497020
Pagar Alam, Pagar Alam City	3°52′43.8″S 103°21′30″E	PaFaw	Maize	*Spodoptera frugiperda*	MZ497021
Tanjung Pering, Ogan Ilir District	104°38'29.058"E 3°12'47.1132"S	TpFaw	Maize	*Spodoptera frugiperda*	MZ497022
Prabumulih, Prabumulih City	3.4328°S 104.2356°E	FawPram	Maize	*Spodoptera frugiperda*	MZ497023
Lahat, Lahat City	3.78639°S 103.54278°E	LasFaw	Maize	*Spodoptera frugiperda*	MZ497024
Muara Enim, Muara Enim District	4.2327°S 103.6141°E	MeFaw	Maize	*Spodoptera frugiperda*	MZ497025
Purwasari, Banyuasin District	2°30'47.268"S 104°40'58.9296"E	PuFaw	Maize	*Spodoptera frugiperda*	MZ497026
Sukarami, Palembang City	2°54'35.3016"S 104°42'14.976"E	SFaw	Maize	*Spodoptera frugiperda*	MZ497027
Tanjung Seteko, Ogan Ilir District	3°13'08"S 104°41'01"E	TsFaw	Maize	*Spodoptera frugiperda*	MZ497028

2.3. Molecular identifications for Spodoptera frugiperda

2.3.1. DNA Extraction

DNA extraction was carried out based on the method of Lestari et al. (2020) with several modifications. *S. frugiperda* larvae that had been preserved in 70% alcohol solution were taken and dried on a tissue for 30 minutes. After that, the caterpillars were soaked in hot water (85 ºC) for 30 minutes until they got slightly whitish in color. Two abdominal segments were then cut and inserted into a 1.5 µl tube. A total of 5 µl Proteinase K was added and crushed until completely crushed. After being crushed, 300 L of TNES buffer was added (Tris HCl 1M(pH 7.5), NaCl 5M, EDTA 0.5 M,ddH2O, and 20% SDS), homogenized and incubated at 60 °C for 3 hours. After the incubation, 85 µL of 5 M NaCl was added and then shaken by hand for 15 seconds and centrifuged for 10 minutes at 14000 rpm. A total of 400 µL of supernatant was taken, put into a new tube and added Isopropanol as much as 60% of the taken volume of supernatant and put in a -40 °C freezer for 20 minutes. After that, it was centrifuged for 5 minutes at a speed of 14000 rpm. The supernatant was then discarded, added 500 µL of cold 70% alcohol and centrifuged for 5 minutes at 14,000 rpm. The

supernatant was then discarded and dried at room temperature for 24 hours (one night). After drying, 20 µL buffer TE (1st Base, Malaysia). Before being used, the DNA suspension was stored at -4 °C. The centrifugation process was carried out using Microspin12 (Biosan, Latvia).

2.3.2. DNA amplification

DNA amplification was performed to amplify the Cytochrome Oxydase Subunit I (COI) region using LCO 1490 and HCO 2198 primers (Folmer et al., 1994). PCR was performed using a Sensoquest Thermal Cycler Machine (Germany) with a total volume of 25 µL consisting of 1 µL DNA, 12.5 µL master mix (2x MyTaq HS Red Mix, Bioline, USA), 1 µL of each primer LCO 1490 and HCO 2198 (Folmer et al., 1994) with a concentration of 10 M and 9.5 µL of sterile distilled water. The PCR was carried out in stages: 1 cycle initiation at 95°C for 5 minutes, followed by 30 cycles consisting of denaturation at 95°C for 1 minute, annealing at 54°C for 1 minute, extension at 72°C for 1 minute and followed by 1 elongation cycle at 72 °C for 5 minutes. The PCR results were then electrophoresed using a 0.5% agarose gel suspension that had been given 1 µL of ethidium bromide (ETBr; 10 mg/mL, per 20 mL agarose) at 55 volts for 70 minutes. The results were then visualized using a DigiDoc UV transilluminator (UVP, USA).

2.3.3. Sequencing and data analysis of sequencing results

The obtained PCR results were then sent to 1st Base Malaysia for the sequencing process. The obtained sequencing results were analyzed using the Bio Edit ver. 7.2.6 for windows (Hall, 1999). The results of the analysis were then submitted to the Basic Local Alignment Search Tool (BLAST) (https://blast.ncbi.nlm.nih.gov/Blast.cgi) to determine their possible identity. The phylogeny tree was created using the Mega 7 for Windows program (Kumar et al., 2016) using the Unweighted-pair Group Method with Arithmetic means (UPGMA). The reference strains used in this study were obtained from NCBI (https://www.ncbi.nlm.nih.gov/).

2.4. Observation of Spodoptera frugiperda in maize fields

2.4.1. Damage by Spodoptera frugiperda

The observations of *S. frugiperda* attacks were carried out from the lowlands to the highlands in 10 locations in South Sumatra, namely Sukarami, Palembang City; Pagar Alam City (Curup Jare

and Suka Rejo); Ogan Ilir District (Tanjung Seteko and Tanjung Pering); Prabumulih. City (Gunung Ibul), Lahat City (Nantigiri); Muara Enim District (Muara Harapan); Banyuasin District (Telang Sari and Mulyasari). From each location, the sample land was taken with an area of 1−5 ha per location and the age of the selected maize ranged from 3 to 6 weeks following Lestari et al. (2020). The survey was carried out from January to June 2021 and the observation of attacks was carried out directly using a scouting system. The scouting system was chosen because the survey area was large and located in many locations (Kuate et al., 2019) and the scouting protocol follows the guidelines of Prasanna et al. (2018). The field scouting was performed to calculate the percentage of infested plant or incidence of damage and to estimate the intensity of attack or severity caused by *S. frugiperda* larvae (Kuate et al., 2019). The maize fields were scouted using a "W" pattern approach and the total sample observed was 50 plants (10 consecutive plants at five different spots along the "W" transect) (Prasanna et al., 2018). Damage to the plants was distinguished by severity of pin holes, shot-holes, lesions, tattering and dead hearts. The percentage of severity or attack intensity was calculated using a rating scale for scoring of damage severity on whorl-stage plants (Kuate et al., 2019).

2.4.2. Data Analysis of Spodoptera frugiperda attack

The percentage of plants infested by FAW larvae termed as an incidence was measured by calculating total of infested plants divided by the total plants observed and multiplied by 100% while the percentage of severity was calculated by dividing the sum of score (excluding score 1) by the number of plants damage (Kuate et al., 2019). The rating scale of damage severity scored from 1 to 5 was used as follows: 1) no damage; 2) 1−10% leaf damage or < 5 mm diameter or only the leaf cuticle destruction; 3) 11−25% leaf damage with presence chewed areas > 5 mm, funnel leaves uninjured; 4) 26−50% leaf damage with presence chewed areas > 1 cm, the funnel less severe; and 5) > 50% leaf damage, plant stunting and funnel damaged severely (Kuate et al., 2019). Data of average incidence (% of infested plant) and severity (on a scale of 1 to 5) per village were tabulated.

3. Results and Discussion

3.1. Morphological characteristics of Spodoptera frugiperda

The larvae collected from 9 survey sites in South Sumatra showed identical morphological characteristics and were identified as *S. frugiperda*. Morphological characters of all larvae found

were characterized by the presence of four pinacula (black dots) on the eighth (second segment of the last segment) abdominal segment forming a square (Fig. 2A). The head of the larvae was dark and there was a single line forming a white inverted Y line on the head (Fig. 2B). The body of the larvae had pale yellow lines along the body dorsally, and yellow stripes subdorsal, thick bands (Fig. 2C). Based on the key to the morphological identification for larvae of *S. frugiperda* illustrated by Lestari et al. (2020), the larvae found in this study were identical to *S. frugiperda*. The morphology of the larvae of this study was also the same as that of *S. frugiperda* as illustrated by Deshmukh et al. (2021) and Sartiami et al. (2020).

The adult *S. frugiperda* moths produced by the larvae rearing from the same colony as the genetic studies showed the following morphological characteristics, an adult male moth had gray-brown forewings with triangular white spots at the tip and mottled-colored (brown, light brown, dark gray) on the upper part forewing, while his hindwing was grayish white with brown outer margin (Fig. 3A). An adult female moth forewing was less distinctly marked and uniform grayish brown, while her hindwing was grayish white similar to the color of the male hindwing (Fig. 3B). The forewing and hindwing coloration of both sexes of adult FAW moths was the same as that of the FAW moths illustrated by Huesing et al. (2018), Lestari et al. (2020), Sartiami et al. (2020). The male and female wingspans documented in this study were also within the range (30 to 40 mm) of the observation result by Huesing et al. (2018).

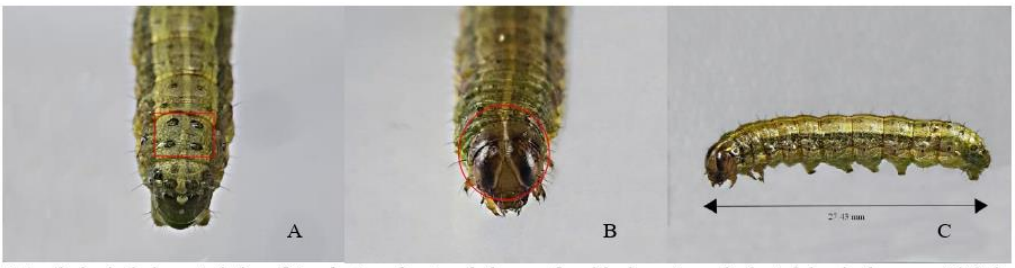

Fig. 2. Morphological characteristics of *Spodoptera frugiperda* larvae: four black spots on the last abdominal segment (A), inverted Y-shape on the head (B), longitudinal strips along the body (C)

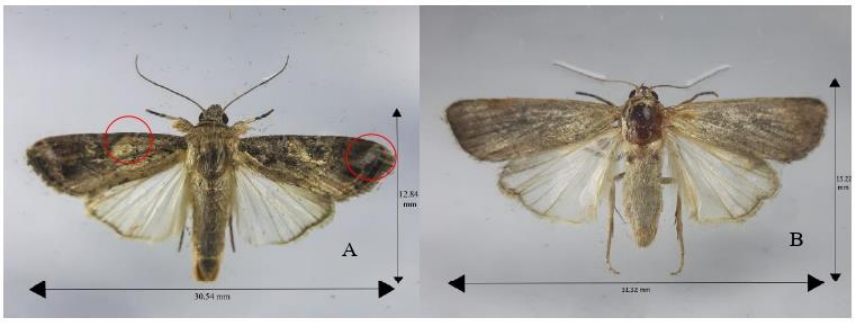

Fig. 3. Adult *Spodoptera frugiperda*: male (A) and female (B).

3.2. Molecular characteristics and genetic diversity of Spodoptera frugiperda

The sequence analysis results showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) were the same (100%). They also shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). They also shared 99.84% of similarity with rice strain haplotype 2 (Table 2).

The other 3 isolates from South Sumatra (AaFaw, PaFaw, TpFaw) were also identical (100% sameness) to each other. They shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan) and respective isolates of Group B from West Sumatra (Nelly et al., 2021). They shared 98.19% of similarity to the above mentioned 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), rice strain haplotype I and *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). The 3 isolates also shared 98.35% of similarity to the rice strain haplotype 2 (Table 2).

The nucleotide difference showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) and *S. frugiperda* from Lampung Province (Lestari et al., 2020) and respective isolates of Group A from West Sumatra (Nelly et al., 2021) were in the same pattern as the reference of rice strains. Meanwhile, the other 3 isolates (AaFaw, PaFaw, TpFaw) and the respective isolates of Group B from West Sumatra (Nelly et al., 2021) are in the same pattern as the reference of corn strains (Table 3).

The phylogenetic tree analysis revealed that the *S. frugiperda* isolates of South Sumatra were divided into 2 groups (I and II). The group I consisted of 6 isolates (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), and was placed within the groups of rice strain haplotype I (Acc. No. U72977.1) and haplotype 2 (Acc. No. U72978.1), the other reference rice strain voucher 93 Brits (Acc. No. MK493022), voucher 94 Brits (Acc. No. MK493021), isolate Yunnan (Acc. No. MK790611), VIE002 (Acc. No. MK913646), strain VIE003 (Acc. No. MK913647.1), strain VIE004 (Acc No. MK913648.1), Belagavi Voucher (Acc. No. MH753329. 1), and isolate Solok (Acc. No. MW876212.1) (Nelly et al. 2021) and Frug Adiluwih Unila (Acc. No. MZ501588) (Lestari et al. 2020). Group II consisted of 3 isolates (AaFaw, PaFaw, TpFaw), and was in the same group as the corn strain haplotype 1 (Acc. No. U72974.1), the other reference corn strain 1L-LepS6 (Acc. No. KX580614.1), Tanah Datar isolate (Acc. No. MW876210.1) (Nelly et al., 2021) and IS 1 (Acc. No. LC546855.1) (Fig. 4).

We confirmed the presence of rice and corn strain in South Sumatra as well as West Sumatra. This is the first report of the occurrence of the rice and corn strain of *S. frugiperda* from South Sumatra. This study also revealed that isolates of *S. frugiperda* in Lampung Province were in the group of a rice strain. Since the prompt spread of this pest, the rice strain is now may exist in the other areas in Sumatra Island, including Lampung province as well as the other corn producing areas in Indonesia. Comprehensive further study should be performed to confirm the presence of both rice and corn strains in all corn producing areas in Indonesia. Further study of strains or genetic diversity of fall armyworm in Indonesia will also provide valuable information on host plant preferences and the indigenous natural enemies for new association with *S. frugiperda*.

3.4. Maize damage caused by Spodoptera frugiperda

The surveys conducted at 10 locations in the lowlands and highlands in South Sumatra showed that all locations were invaded by *S. frugiperda.* The mean incidence of *S. frugiperda* in each location varied, the highest was found in Suka Rejo, Pagar Alam City (100%), while the lowest attack was in Sukarami, Palembang City (44.0%). The severity ranged from 11.5% to 65% (Table 4). It means that outbreaks of *S. frugiperda* have occured in the South Sumatra. In the lowlands and highlands of South Sumatra, the incidence and severity of *S. frugiperda* tended to be high in all locations. Therefore, the altitude of the location did not affect the severe or mild attack of this FAW. The observation during the surveys revealed that the egg mass was laid by the adult females on the leaf surface (Fig. 5A). The larvae found in the fields attacked the leaves and whorl (Fig. 5B−C). The attack symptoms by the larvae showed typical characteristics, namely holes used by the larvae on the leaves and leaves with transparent bite marks. On the stems or leaves there were brown larval frass similar to sawdust (Fig. 5D). Young leaves that were still curled up could also be attacked by the larvae as a result of the leaf rolls forming the holes. The larvae also perforated the maize stalks (Fig. 5E), flower (F), cobs (Fig. 5G−H), funnel damaged (I). The severe attack found in this study was in the vegetative phase, while in the generative phase the attack was low. However, in this survey the observation of attacks was limited to maize aged 3−6 weeks.

Table 2.
Similarity among *Spodoptera frugiperda* collected from South Sumatera, West Sumatera, Lampung, Indonesia and other countries including rice and corn strain isolates.

Isolate	Similarity (%)															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	100															
2	100	100														
3	100	100	100													
4	100	100	100	100												
5	100	100	100	100	100											
6	100	100	100	100	100	100										
7	98.35	98.35	98.35	98.35	98.35	98.35	100									
8	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100								
9	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100							
10	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100						
11	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100					
12	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100				
13	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100			
14	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100		
15	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	
16	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	100

1. Corn haplotype 1 (Maize, USA) (Acc. No. U72974), 2. AaFaw (Maize, South Sumatera) (Acc. No. MZ497020), 3. PaFaw (Maize, South Sumatera) (Acc. No. MZ497021), 4. TpFaw (Maize, South Sumatera) (Acc. No. MZ497022), 5. Isolate Tanah Datar (Maize, West Sumatera) (Acc. No. MW876210), 6. IS_1 (Sugarcane, Japan) (Acc. No. LC546855), 7. Rice (USA) haplotype 2 (Acc. No. U72978), 8. Rice (USA) haplotype 1 (Acc. No. U72977), 9. FawPram (Maize, South Sumatera) (Acc. No. MZ497023), 10. LasFaw (Maize, South Sumatera) (Acc. No. MZ4970224), 11. MeFaw (Maize, South Sumatera) (Acc. No. MZ497025), 12. PuFaw (Maize, South Sumatera) (Acc. No. MZ497026), 13. SFaw (Maize, South Sumatera) (Acc. No. MZ497027), 14. TsFaw (Maize, South Sumatera) (Acc. No. MZ497028), 15. Isolate Solok (Maize, West Sumatera) (Acc. No. MW876212), 16. Frugi_Adiluwih_UNILA (Maize, Lampung) (Acc. No. MZ501588). Length of the nucleotides: 607bp.

Table 3.

Nucleotides difference between *Spodoptera frugiperda* from South Sumatera and foreign countries.

Isolate	Accesion Number	Position of nucleotide difference										
		11	56	110	146	197	428	503	509	539	573	602
Corn haplotype 1 (USA)	U72974	G	G	T	T	C	T	T	C	C	T	T
AaFaw (Maize, South Sumatera)	MZ497020	.	.	.	.	.	.	.	.	.	.	.
PaFaw (Maize, South Sumatera)	MZ497021	.	.	.	.	.	.	.	.	.	.	.
TpFaw (Maize, South Sumatera)	MZ497022	.	.	.	.	.	.	.	.	.	.	.
Isolate Tanah Datar (Maize, West Sumatera)	MW876210	.	.	.	.	.	.	.	.	.	.	.
IS_1 (Sugarcane, Japan)	LC546855	.	.	.	.	.	.	.	.	.	.	.
Rice haplotype 2 (USA)	U72978	A	A	.	A	T	C	C	T	T	C	A
Rice haplotype 1 (USA)	U72977	A	A	C	A	T	C	C	T	T	C	A
FawPram (Maize, South Sumatera)	MZ497023	A	A	C	A	T	C	C	T	T	C	A
LasFaw (Maize, South Sumatera)	MZ4970224	A	A	C	A	T	C	C	T	T	C	A
MeFaw (Maize, South Sumatera)	MZ497025	A	A	C	A	T	C	C	T	T	C	A
PuFaw (Maize, South Sumatera)	MZ497026	A	A	C	A	T	C	C	T	T	C	A
SFaw (Maize, South Sumatera)	MZ497027	A	A	C	A	T	C	C	T	T	C	A
TsFaw (Maize, South Sumatera)	MZ497028	A	A	C	A	T	C	C	T	T	C	A
Isolate Solok (Maize, West Sumatera)	MW876212	A	A	C	A	T	C	C	T	T	C	A
Frugi_Adiluwih_UNILA (Maize, Lampung)	MZ501588	A	A	C	A	T	C	C	T	T	C	A

Length of the nucleotides: 607bp

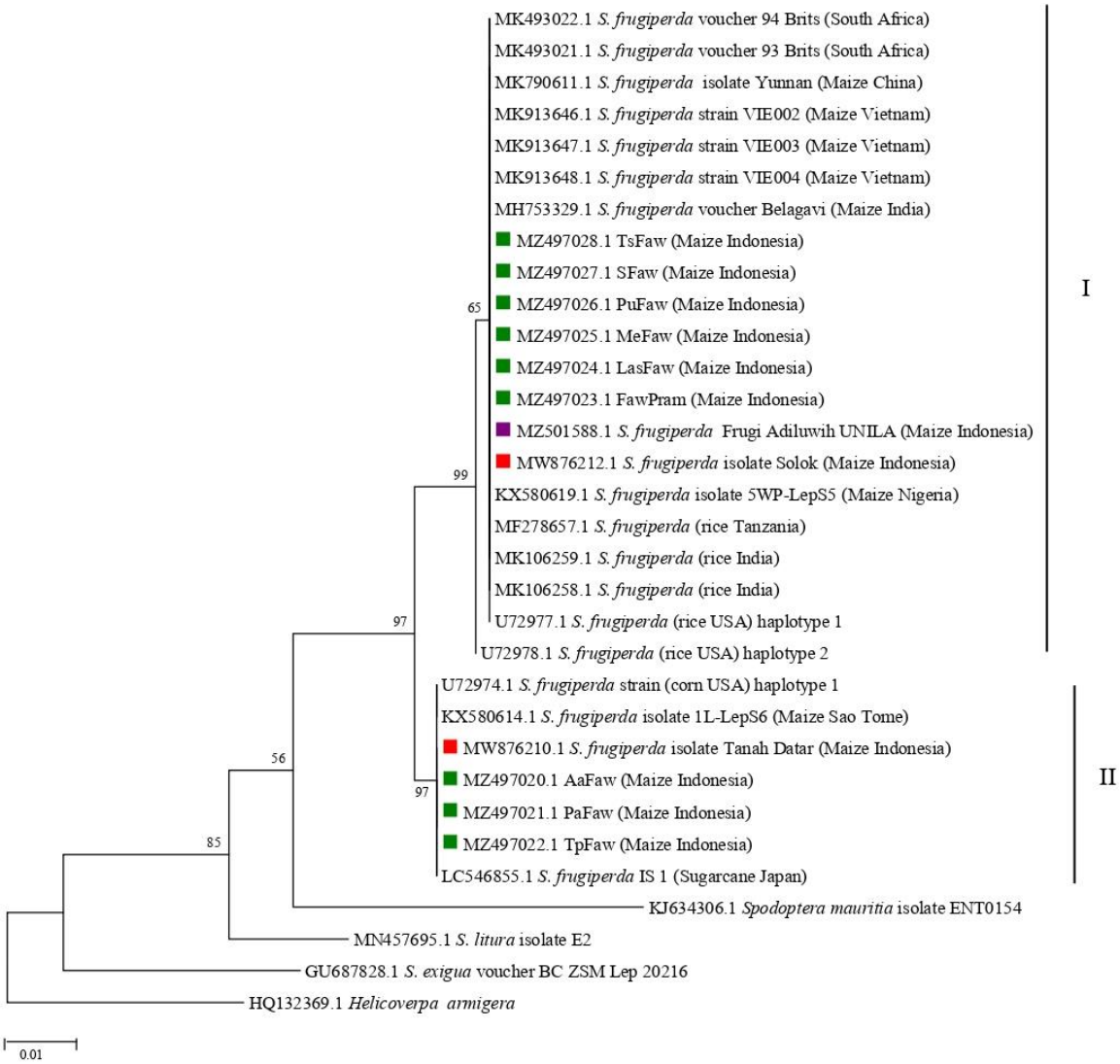

Fig. 4. Phylogenetic tree developed based on mitochondrial Cytochrome Oxydase I gene by maximum Likelihood method (Tamura-Nei model) using Mega7 for windows (Kumar et al 2016). The *Spodoptera frugiperda* isolates collected from South Sumatera were divided into 2 groups (I and II). The group I belong to the clade of "rice strain" meanwhile the group II was a member of the clade of "corn strain". *Helicoverpa armigera* (Acc. No. HQ132369.1) was used as outgroup. The *Spodoptera frugiperda* used in this study. The *Spodoptera frugiperda* from West Sumatera, Indonesia. The *Spodoptera frugiperda* from Lampung, Indonesia.

Table 4.

Damage by *Spodoptera frugiperda* in South Sumatra, Indonesia

Survey sites (village, district/city)	Survey date	Altitude (m)	Coordinate	Mean incidence (% of infested	Mean severity (on a scale of 1

				plant)	to 5)
Sukarame, Palembang	14-05-2021	32.0	2°55′07″S 104°16′02″E	44.0	11.5
Curup Jare, Pagar Alam City	30-05-2021	782.0	103°13'17.0904"E 4°0'58.7556"S	70.0	22.5
Suka Rejo, Pagar Alam City	30-05-2021	708.7	103°14′589″E 4°1′066″S	100.0	65.0
Tanjung Seteko, Ogan Ilir District	23-05-2021	23.3	3°13'08"S 104°41'01"E	82.0	38.7
Tanjung Pering, Ogan Ilir District	23-05-2021	35.0	104°38'29.058"E 3°12'47.1132"S	85.6	38.4
Gunung Ibul, Prabumulih City	17-06-2021	44.9	104°17'12.2856"E 3°24'52.3008"S	66.0	20.0
Nantigiri, Lahat City	31-05-2021	740.0	3°56'22"S 103°12'15"E	88.0	65.0
Muara Harapan, Muara Enim District	17-06-2021	99.8	3°38'01"S 103°49'24"E	84.0	27.5
Telang Sari, Banyuasin District	16-06-2021	24.5	104°38'40.2036"E 2°31'4.6596"S	50.0	22.5
Mulyasari, Banyuasin District	16-06-2021	27.0	104°39'27.2772"E 2°33'4.6764"S	45.3	17.5

The symptoms of *S. frugiperda* larvae attack in this study had the same characteristics as those of *S. frugiperda* found by Ginting et al. (2020) and Sartiami et al. (2020). The attack began with the larvae perforating the young leaves of the plant, and then perforating the young leaves that were still curled up, and at the worst the larvae cut the growing point of the maize (Ginting et al., 2020). Supartha et al. (2021) stated that the severity of attack by the larvae reached its peak when the maize was 4 weeks old, then the attack continued to decrease and at 8 weeks or more the attack was very low. During the survey, generally the severely affected maize was aged 3-6, the fruit bearing maize showed low attack.

This survey data showed that there was no consistent effect of the altitude of the survey sites with the severity of *S. frugiperda* attacks. However, Supartha et al. (2021) reported that in the highlands (> 500 m below sea level) there was no attack but in the lowlands this FAW attack was very high. This study has not been able to conclude the effects of corn and rice strain of *S. frugiperda* on the incidence and severity of the FAW. Therefore, further research on the effects of corn and rice strain of *S. frugiperda* on the incidence and severity of the FAW needs to be performed. In addition, the range of host plants of these two strains also needs to be studied comprehensively.

Fig. 5. Symptoms damage by *Spodoptera frugiperda* larvae in maize: egg mass on the leaf surface (A), larvae feeding on leaves (B), larvae feeding on leaf whorl (C), brown larval frass similar to sawdust (D), larvae feeding on corn stalks (E), larvae feeding on corn flower (F), larvae feeding on corn cobs (G), larvae feeding on corn cob tip (H), and funnel damaged (I).

4. Conclusion

This study have found and confirmed the presence of rice and corn strain of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra, Indonesia. The incidence and severity of *S. frugiperda* from the lowlands to the highlands is high with the incidence reaching 100% and the severity reaching 65%. we also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. Comprehensive further study should be performed to confirm the presence of both rice and corn strains in all corn producing areas in Indonesia.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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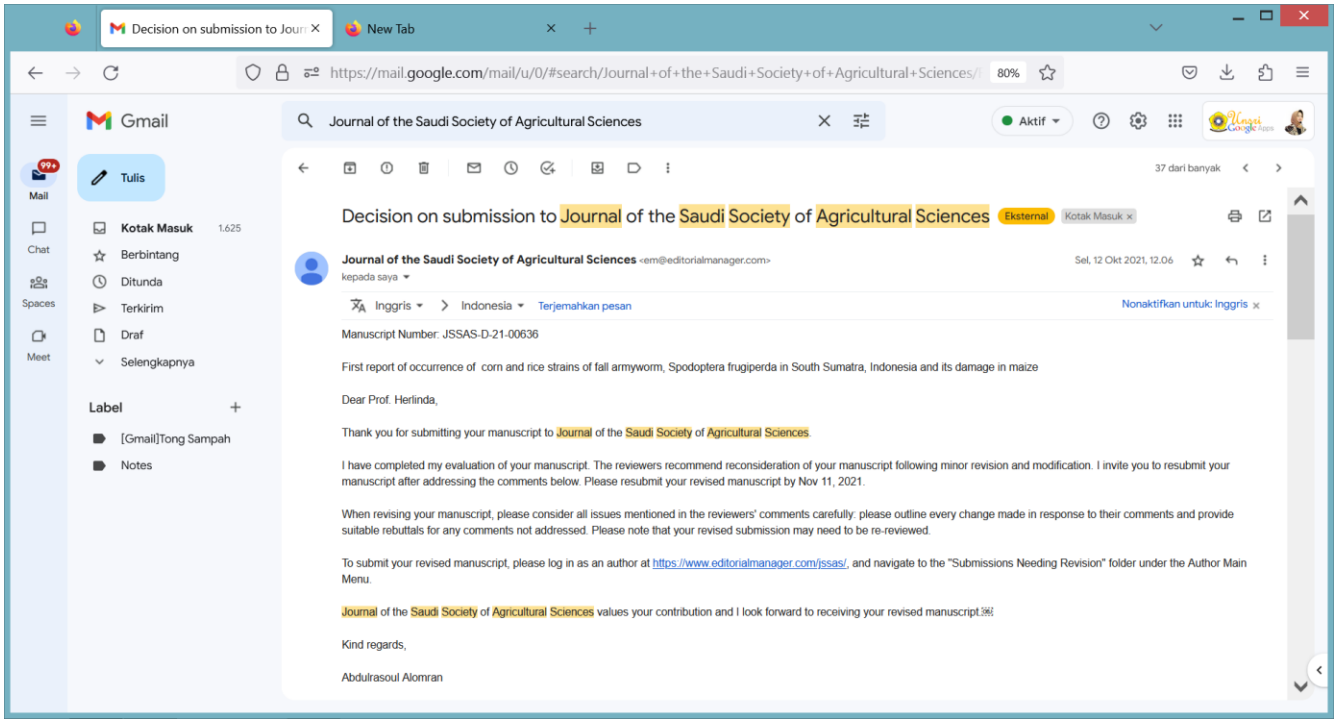
459

2. Bukti konfirmasi review pertama 12 Oktober 2021



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Sel, 12 Okt 2021, 12.06

Manuscript Number: JSSAS-D-21-00636

First report of occurrence of corn and rice strains of fall armyworm, Spodoptera frugiperda in South Sumatra, Indonesia and its damage in maize

Dear Prof. Herlinda,

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3. Bukti konfirmasi submit revisi dan hasil revisi pertama 16 Oktober 2021

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First report of occurrence of corn and rice strains of fall armyworm, Spodoptera frugiperda in South Sumatra, Indonesia and its damage in maize

--Manuscript Draft--

Manuscript Number:	JSSAS-D-21-00636R1
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Keywords:	Genetic diversity; incidence; outbreaks; scouting; severity
Corresponding Author:	Siti Herlinda, Dr. INDONESIA
First Author:	Siti Herlinda, Dr.
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	Radix Suharjo
	Melati Elbi Sinaga
	Fairuz Fawwazi
	Suwandi Suwandi
Manuscript Region of Origin:	Asia Pacific
Abstract:	Spodoptera frugiperda is a new invasive pest in Indonesia and its severity in maize ranges from 26.50 to 100%. However, information on the strains or genetic diversity of the S. frugiperda in Indonesia is still very limited. This research aimed to identify the genetic diversity of S. frugiperda from South Sumatra and determine its damage in maize. Surveys from January to June 2021 were carried out from the lowlands to highlands of South Sumatra. The field scouting was performed to calculate the incidence of damage and to estimate the severity caused by S. frugiperda larvae. The severity was assessed using a visual rating scale from 1 (no damage) to 5 (plant stunting and funnel damaged severely). The S. frugiperda larvae was identified based on morphological characters and molecular techniques using sequence analysis of Cytochrome c Oxidase subunit I (COI) gene. All larvae collected from South Sumatra showed identical morphological characteristics identified as S. frugiperda. The sequence analysis results showed that the 6 isolates of S. frugiperda shared 100% of sameness as the rice strain haplotype 1, S. frugiperda isolate from Lampung Province. The other 3 isolates of S. frugiperda shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan). All isolates have been deposited in the GenBank. This study confirmed the presence of rice and corn strains of Spodoptera frugiperda and this is the first report of the occurrence of both strains in South Sumatra. We also found that outbreaks of S. frugiperda have occured in the South Sumatra. The incidence and severity of S. frugiperda reached 100% and 65% respectively. Comprehensive further study should be performed to confirm the presence of both strains and their damage in all corn producing areas in Indonesia.
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	H. De Groote International Maize and Wheat Improvement Centre (CIMMYT), Nairobi, Kenya h.degroote@cgiar.org
	Rhett D. Harrison World Agroforestry Centre, 13 Elm Road, Woodlands, Lusaka, Zambia r.harrison@cgiar.org
Response to Reviewers:	Dear reviewer #1. Thank you very much for your valuable review and suggestions. Here are our detailed responses. All the revisions in the manuscript have been highlighted with yellow colour. Dear reviewer #2 . Thank you very much for no revision of our manuscript.

Reviewer #1: The authors have written a manuscript on 'First report of occurrence of corn and rice strains of fall armyworm, Spodoptera frugiperda in South Sumatra, Indonesia and its damage in maize'. The subject is topical and helps to understand the evolution of spodoptera frugiperda damage in Indonesia. The manuscript is well written. The methodology is adequate and follows the principles of Spodoptera sample collection. I recommand minor revisions.
Response to Reviewers #1: Thank you very much

Abstract
Lines 10 - 11: It must be clearly defined how the damage was assessed. What is the scouting system? Didn't you use the Davis scale method?
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Line 19: You must start the sentence with a capital letter.
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Introduction
It has been reported that the 2 strains of Spodoptera frugiperda have been found in the country. On which crops these 2 strains were identified? The crops on which these 2 strains were identified are of food importance in the country? What place do they occupy among the crops commonly consumed by populations?
Response to Reviewers #1: All the S. frugiperda were collected from maize. In Indonesia, maize is one of important crops which is widely benefitted as a raw material in the feed and food industries as well as staple food in some regions. It has been added in the text.

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Lines 70 - 71: The survey started from January to June 2021. Which season of the year is it? Rainy season or dry season? It is necessary to specify the characteristics of the vegetation according to the seasons. This can show the presence of host plants in the study area.
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Response to Reviewers #1: It has been deleted.

Data Analysis of Spodoptera frugiperda attack
Lines 164 - 172: What is presented here is not data analysis. You must present the statistical analysis of the data. Data of this study must be statistically analyzed to add value to the study.
Response to Reviewers #1: It has been revised. The revisions: Incidence and severity of S. frugiferda infestation was tested for normality using the Shapiro–Wilk test and for variance homogeneity by Levene's test. Square root transformation was performed to homogenous variance and to meet normality assumptions before being subjected to one-way analyses of variance. Means were compared using Tukey's honestly significant test (HSD) and back-transformed means were presented after analysis. R

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Dear reviewer #1. Thank you very much for your valuable review and suggestions. Here are our detailed responses. All the revisions in the manuscript have been highlighted with yellow colour.
Dear reviewer #2 . Thank you very much for no revision of our manuscript.

Reviewer #1: The authors have written a manuscript on 'First report of occurrence of corn and rice strains of fall armyworm, Spodoptera frugiperda in South Sumatra, Indonesia and its damage in maize'. The subject is topical and helps to understand the evolution of spodoptera frugiperda damage in Indonesia. The manuscript is well written. The methodology is adequate and follows the principles of Spodoptera sample collection. I recommand minor revisions.
Response to Reviewers #1: **Thank you very much**

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First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

Siti Herlinda[a,b*], Radix Suharjo[c], Melati Elbi Sinaga[a], Fairuz Fawwazi[a], Suwandi Suwandi[a,b]

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Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

All data are available in the article and the materials used in this work are of high quality and grade.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Authors' contributions

SH performed research concept and design, writing the article, and final approval of article. RS prepared and performed molecular identification and data analysis and interpretation. MES and FF performed collection and assembly of data. SS prepared and performed morphological

identification and critical revision of the article. All the authors read and approved the manuscript.

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First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

ABSTRACT

Spodoptera frugiperda is a new invasive pest in Indonesia and its severity in maize ranges from 26.50 to 100%. However, information on the strains or genetic diversity of the *S. frugiperda* in Indonesia is still very limited. This research aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize. Surveys from January to June 2021 were carried out from the lowlands to highlands of South Sumatra. The field scouting was performed to calculate the incidence of damage and to estimate the severity caused by *S. frugiperda* larvae. The severity was assessed using a visual rating scale from 1 (no damage) to 5 (plant stunting and funnel damaged severely). The *S. frugiperda* larvae was identified based on morphological characters and molecular techniques using sequence analysis of Cytochrome c Oxidase subunit I (*COI*) gene. All larvae collected from South Sumatra showed identical morphological characteristics identified as *S. frugiperda*. The sequence analysis results showed that the 6 isolates of *S. frugiperda* shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province. The other 3 isolates of *S. frugiperda* shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan). All isolates have been deposited in the GenBank. This study confirmed the presence of rice and corn strains of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra. We also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. The incidence and severity of *S. frugiperda* reached 100% and 65% respectively. Comprehensive further study should be performed to confirm the presence of both strains and their damage in all corn producing areas in Indonesia.

Keywords: genetic diversity; incidence; outbreaks; scouting; severity

1. Introduction

Fall armyworm (FAW), *Spodoptera frugiperda* is a new invasive maize pest in Indonesia. This insect pest comes from the American continent (Nagoshi et al., 2017; Otim et al., 2018). In 2016, the FAW was reported to have come into Africa (Goergen et al., 2016). In 2017, FAW crossed over to Europe (Early et al., 2018). This pest began to move into Asia in 2018 (Mahat et al., 2021) and was first discovered in India (Ganiger et al., 2018) and came into Indonesia for the first time on March 26, 2019 in West Sumatra (Sartiami et al., 2020). Then, it began to spread to other provinces

and islands in Indonesia, such as South Sumatra (Hutasoit et al., 2020), West Java (Maharani et al., 2019), Lampung (Trisyono et al., 2019), Bengkulu (Ginting et al., 2020), Bali(Supartha et al., 2021).

In addition to spreading throughout the world, the FAW has caused maize yield losses of up to 18 million tons/year and losses of up to 13 million US$ in 12 African countries (Harrison et al., 2019). In Kenya, the loss due to this pest reaches 1 million ton/year (De Groote et al., 2020). Besides attacking the maize, this pest attacks paddy, sugarcane, cotton, and ornamental plants (IPPC, 2019). In Brazil it has been reported that about 76 plant families were destroyed by this pest (Montezano et al., 2018). In Indonesia, the FAW generally attacks maize with damage in Lampung ranging from 26.50% to 70% (Lestari et al., 2020), in Bali reaching 47.84% (Supartha et al., 2021), in East Nusa Tenggara ranging from 85% to 100% (Mukkun et al., 2021). This pest can also attack paddy leaves, for example, in Banten the FAW larvae was found to attack paddy (Sartiami et al., 2020); yet there is no information on damage by this pest to paddy in Indonesia.

There are two strains of *S. frugiperda* in the world, namely corn strain (C) and rice strain (R) (Unbehend et al., 2013; Va et al., 2014). The genetic diversity of *S. frugiperda* in Indonesia was first reported by Sartiami et al. (2020) stating that the strain of *S. frugiperda* found in Banten was only the rice strain. In Lampung, the strain of *S. frugiperda* that was found from maize (Lestari et al., 2020) was also confirmed as the rice strain, while both the corn and rice strains were collected from maize production centers in West Sumatera (Nelly et al., 2021). In Indonesia, maize is one of important crops which is widely benefitted as a raw material in the feed and food industries as well as staple food in some regions (Nurina et al., 2021). The corn and rice strains of *S. frugiperda* if they have spread in Indonesia can harm not only maize but also paddy and other important crops. However, information on the strains/genetic diversity of *S. frugiperda* in Indonesia is still very limited and until now the information on the strain of *S. frugiperda* originating from South Sumatra and its attack has not been reported. For this reason, information on the genetic diversity of the FAW in South Sumatra and its attacks is needed so that it can be used as a basis for controlling this pest and can complement information on *S. frugiperda* strains in Indonesia. This study aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize.

2. Materials and methods

2.1. Survey sites

Surveys to obtain specimens of *S. frugiperda* larvae were carried out from the lowlands to the highlands of South Sumatra, such as Palembang City (2°59′27.99″S 104°45′24.24″E), Pagar Alam City (3°52′43.8″S 103 °21′30″E), Lahat City (3.78639°S 103.54278°E), Ogan Ilir District (3.43186°S 104.6727°E), Prabumulih City (3.4328°S 104.2356°E), Muara Enim District (4.2327°S 103.6141 °E), and Banyuasin District (2.8833°S 104.3831°E) (Fig. 1 and Table 1). The survey started from January to June 2021 covering rainy (January to February), transition (March to June) and early dry (June) seasons. The specimens obtained were then identified molecularly at the Laboratory of Agricultural Biotechnology (accredited according to the ISO 17025 standard), Department of Plant Protection, Faculty of Agriculture, Lampung University, Indonesia.

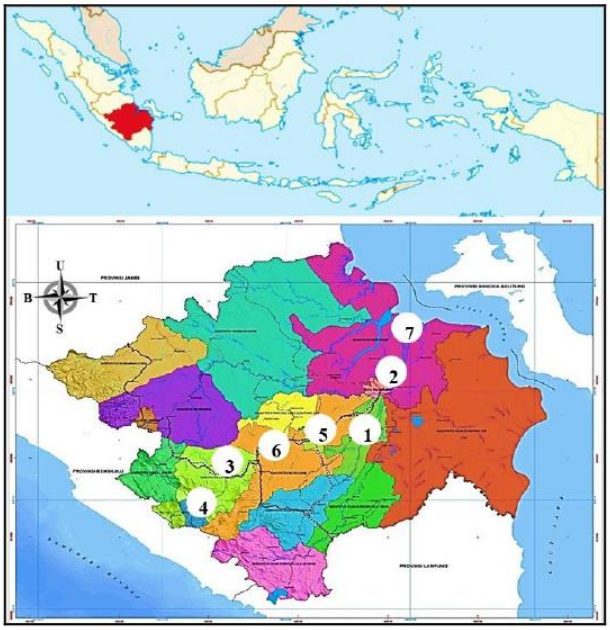

Fig. 1. Surveys locations in South Sumatra, Indonesia: Ogan Ilir District (1), Palembang City (2), Lahat District (3), Pagar Alam City (4), Prabumulih City (5), Muara Enim District (6), dan Banyuasin District (7).

2.2. Morphological identifications for Spodoptera frugiperda

S. frugiperda larvae were collected from maize fields in various districts/cities in South Sumatra, Indonesia. The larvae were brought to the Entomology Laboratory, Department of Plant Protection, Faculty of Agriculture, Universitas Sriwijaya, Indonesia to be reared individually in porous plastic cups (Ø 6.5 cm, height 4.6 cm). Maize leaves (2 cm x 5 cm) were placed into the cup to feed *S. frugiperda* and then the larvae were observed for further identification. Some samples of larvae were put into vials containing 70% alcohol for molecular identification at the Laboratory of

Agricultural Biotechnology, Department of Plant Protection, Faculty of Agriculture, Universitas Lampung. Some other samples of larvae from the same population were kept until the adult completed one life cycle following the method of Gustianingtyas et al. (2021) to observe the morphology of the adult.

Table 1.

The origin of sample of *Spodoptera frugiperda* from South Sumatra, Indonesia.

Location (village, district/city)	Coordinate	Sample ID	Sample source	Species	GenBank Acc. No.
Alang-alang Lebar, Palembang City	2°59'27"S 104°45'24"E	AaFaw	Maize	*Spodoptera frugiperda*	MZ497020
Pagar Alam, Pagar Alam City	3°52′43.8″S 103°21′30″E	PaFaw	Maize	*Spodoptera frugiperda*	MZ497021
Tanjung Pering, Ogan Ilir District	104°38'29.058"E 3°12'47.1132"S	TpFaw	Maize	*Spodoptera frugiperda*	MZ497022
Prabumulih, Prabumulih City	3.4328°S 104.2356°E	FawPram	Maize	*Spodoptera frugiperda*	MZ497023
Lahat, Lahat City	3.78639°S 103.54278°E	LasFaw	Maize	*Spodoptera frugiperda*	MZ497024
Muara Enim, Muara Enim District	4.2327°S 103.6141°E	MeFaw	Maize	*Spodoptera frugiperda*	MZ497025
Purwasari, Banyuasin District	2°30'47.268"S 104°40'58.9296"E	PuFaw	Maize	*Spodoptera frugiperda*	MZ497026
Sukarami, Palembang City	2°54'35.3016"S 104°42'14.976"E	SFaw	Maize	*Spodoptera frugiperda*	MZ497027
Tanjung Seteko, Ogan Ilir District	3°13'08"S 104°41'01"E	TsFaw	Maize	*Spodoptera frugiperda*	MZ497028

2.3. Molecular identifications for Spodoptera frugiperda

2.3.1. DNA Extraction

DNA extraction was carried out based on the method of Lestari et al. (2020) with several modifications. *S. frugiperda* larvae that had been preserved in 70% alcohol solution were taken and dried on a tissue for 30 minutes. After that, the caterpillars were soaked in hot water (85 ºC) for 30 minutes until they got slightly whitish in color. Two abdominal segments were then cut and inserted into a 1.5 µl tube. A total of 5 µl Proteinase K was added and crushed until completely crushed. After being crushed, 300 L of TNES buffer was added (Tris HCl 1M(pH 7.5), NaCl 5M, EDTA 0.5 M,ddH2O, and 20% SDS), homogenized and incubated at 60 °C for 3 hours. After the incubation, 85 µL of 5 M NaCl was added and then shaken by hand for 15 seconds and centrifuged for 10

minutes at 14000 rpm. A total of 400 µL of supernatant was taken, put into a new tube and added Isopropanol as much as 60% of the taken volume of supernatant and put in a -40 °C freezer for 20 minutes. After that, it was centrifuged for 5 minutes at a speed of 14000 rpm. The supernatant was then discarded, added 500 µL of cold 70% alcohol and centrifuged for 5 minutes at 14,000 rpm. The supernatant was then discarded and dried at room temperature for 24 hours (one night). After drying, 20 µL buffer TE (1st Base, Malaysia). Before being used, the DNA suspension was stored at -4 °C. The centrifugation process was carried out using Microspin12 (Biosan, Latvia).

2.3.2. DNA amplification

DNA amplification was performed to amplify the Cytochrome Oxydase Subunit I (COI) region using LCO 1490 and HCO 2198 primers (Folmer et al., 1994). PCR was performed using a Sensoquest Thermal Cycler Machine (Germany) with a total volume of 25 µL consisting of 1 µL DNA, 12.5 µL master mix (2x MyTaq HS Red Mix, Bioline, USA), 1 µL of each primer LCO 1490 and HCO 2198 (Folmer et al., 1994) with a concentration of 10 M and 9.5 µL of sterile distilled water. The PCR was carried out in stages: 1 cycle initiation at 95°C for 5 minutes, followed by 30 cycles consisting of denaturation at 95°C for 1 minute, annealing at 54°C for 1 minute, extension at 72°C for 1 minute and followed by 1 elongation cycle at 72 °C for 5 minutes. The PCR results were then electrophoresed using a 0.5% agarose gel suspension that had been given 1 µL of ethidium bromide (ETBr; 10 mg/mL, per 20 mL agarose) at 55 volts for 70 minutes. The results were then visualized using a DigiDoc UV transilluminator (UVP, USA).

2.3.3. Sequencing and data analysis of sequencing results

The obtained PCR results were then sent to 1st Base Malaysia for the sequencing process. The obtained sequencing results were analyzed using the Bio Edit ver. 7.2.6 for windows (Hall, 1999). The results of the analysis were then submitted to the Basic Local Alignment Search Tool (BLAST) (https://blast.ncbi.nlm.nih.gov/Blast.cgi) to determine their possible identity. The phylogeny tree was created using the Mega 7 for Windows program (Kumar et al., 2016) using the maximum Likelihood method (1000X bootstrap; Tamura-Nei model). The reference strains used in this study were obtained from NCBI (https://www.ncbi.nlm.nih.gov/).

2.4. Observation of Spodoptera frugiperda in maize fields

2.4.1. Damage by Spodoptera frugiperda

The observations of *S. frugiperda* attacks were carried out from the lowlands to the highlands in 10 locations in South Sumatra, namely Sukarami, Palembang City; Pagar Alam City (Curup Jare and Suka Rejo); Ogan Ilir District (Tanjung Seteko and Tanjung Pering); Prabumulih. City (Gunung Ibul), Lahat City (Nantigiri); Muara Enim District (Muara Harapan); Banyuasin District (Telang Sari and Mulyasari). From each location, the sample land was taken with an area of 1−5 ha per location and the age of the selected maize ranged from 3 to 6 weeks following Lestari et al. (2020). The observation of attacks was carried out directly using a scouting system. The scouting system was chosen because the survey area was large and located in many locations (Kuate et al., 2019) and the scouting protocol follows the guidelines of Prasanna et al. (2018). The field scouting was performed to calculate the percentage of infested plant or incidence of damage and to estimate the intensity of attack or severity caused by *S. frugiperda* larvae (Kuate et al., 2019). The maize fields were scouted using a "W" pattern approach and the total sample observed was 50 plants (10 consecutive plants at five different spots along the "W" transect) (Prasanna et al., 2018). Damage to the plants was distinguished by severity of pin holes, shot-holes, lesions, tattering and dead hearts. The percentage of severity or attack intensity was calculated using a rating scale for scoring of damage severity on whorl-stage plants (Kuate et al., 2019).

The percentage of plants infested by FAW larvae termed as an incidence was measured by calculating total of infested plants divided by the total plants observed and multiplied by 100% while the percentage of severity was calculated by dividing the sum of score (excluding score 1) by the number of plants damage (Kuate et al., 2019). The visual rating scale of damage severity scored from 1 to 5 was used as follows: 1) no damage; 2) 1−10% leaf damage or < 5 mm diameter or only the leaf cuticle destruction; 3) 11−25% leaf damage with presence chewed areas > 5 mm, funnel leaves uninjured; 4) 26−50% leaf damage with presence chewed areas > 1 cm, the funnel less severe; and 5) > 50% leaf damage, plant stunting and funnel damaged severely (Kuate et al., 2019).

2.4.2. Data Analysis of Spodoptera frugiperda attack

Incidence and severity of *S. frugiferda* infestation was tested for normality using the Shapiro–Wilk test and for variance homogeneity by Levene's test. Square root transformation was performed to homogenous variance and to meet normality assumptions before being subjected to one-way analyses of variance. Means were compared using Tukey's honestly significant test (HSD) and

back-transformed means were presented after analysis. R studio Version 1.4.1106 (RStudio PBC, Boston, MA, USA) was used for analyses of infestation data.

3. Results and Discussion

3.1. Morphological characteristics of Spodoptera frugiperda

The larvae collected from 9 survey sites in South Sumatra showed identical morphological characteristics and were identified as *S. frugiperda*. Morphological characters of all larvae found were characterized by the presence of four pinacula (black dots) on the eighth (second segment of the last segment) abdominal segment forming a square (Fig. 2A). The head of the larvae was dark and there was a single line forming a white inverted Y line on the head (Fig. 2B). The body of the larvae had pale yellow lines along the body dorsally, and yellow stripes subdorsal, thick bands (Fig. 2C). Based on the key to the morphological identification for larvae of *S. frugiperda* illustrated by Lestari et al. (2020), the larvae found in this study were identical to *S. frugiperda*. The morphology of the larvae of this study was also the same as that of *S. frugiperda* as illustrated by Deshmukh et al. (2021) and Sartiami et al. (2020).

The adult *S. frugiperda* moths produced by the larvae rearing from the same colony as the genetic studies showed the following morphological characteristics, an adult male moth had gray-brown forewings with triangular white spots at the tip and mottled-colored (brown, light brown, dark gray) on the upper part forewing, while his hindwing was grayish white with brown outer margin (Fig. 3A). An adult female moth forewing was less distinctly marked and uniform grayish brown, while her hindwing was grayish white similar to the color of the male hindwing (Fig. 3B). The forewing and hindwing coloration of both sexes of adult FAW moths was the same as that of the FAW moths illustrated by Huesing et al. (2018), Lestari et al. (2020), Sartiami et al. (2020). The male and female wingspans documented in this study were also within the range (30 to 40 mm) of the observation result by Huesing et al. (2018).

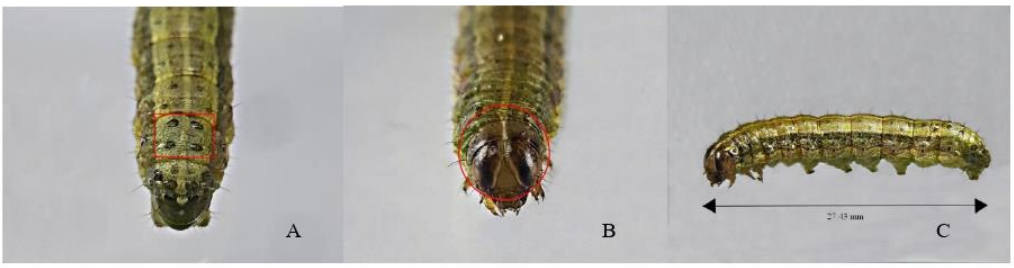

Fig. 2. Morphological characteristics of *Spodoptera frugiperda* larvae: four black spots on the last abdominal segment (A), inverted Y-shape on the head (B), longitudinal strips along the body (C)

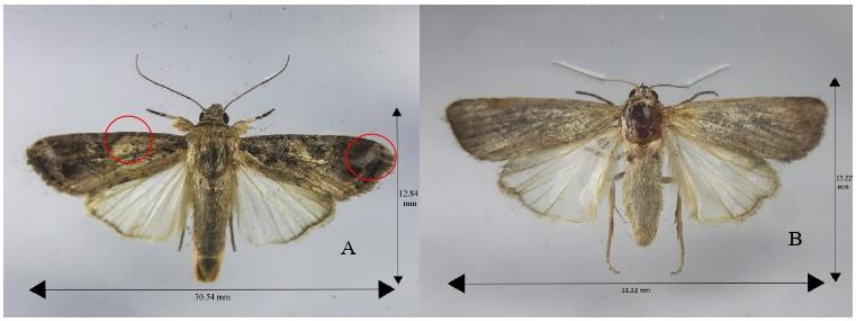

Fig. 3. Adult *Spodoptera frugiperda*: male (A) and female (B).

3.2. Molecular characteristics and genetic diversity of Spodoptera frugiperda

The sequence analysis results showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) were the same (100%). They also shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). They also shared 99.84% of similarity with rice strain haplotype 2 (Table 2).

The other 3 isolates from South Sumatra (AaFaw, PaFaw, TpFaw) were also identical (100% sameness) to each other. They shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan) and respective isolates of Group B from West Sumatra (Nelly et al., 2021). They shared 98.19% of similarity to the above mentioned 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), rice strain haplotype I and *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). The 3 isolates also shared 98.35% of similarity to the rice strain haplotype 2 (Table 2).

The nucleotide difference showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) and *S. frugiperda* from Lampung Province (Lestari et al., 2020) and respective isolates of Group A from West Sumatra (Nelly et al., 2021) were in the same pattern as the reference of rice strains. Meanwhile, the other 3 isolates (AaFaw, PaFaw, TpFaw) and the respective isolates of Group B from West Sumatra (Nelly et al., 2021) are in the same pattern as the reference of corn strains (Table 3).

The phylogenetic tree analysis revealed that the *S. frugiperda* isolates of South Sumatra were divided into 2 groups (I and II). The group I consisted of 6 isolates (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), and was placed within the groups of rice strain haplotype I (Acc. No.

239 U72977.1) and haplotype 2 (Acc. No. U72978.1), the other reference rice strain voucher 93 Brits
240 (Acc. No. MK493022), voucher 94 Brits (Acc. No. MK493021), isolate Yunnan (Acc. No.
241 MK790611), VIE002 (Acc. No. MK913646), strain VIE003 (Acc. No. MK913647.1), strain
242 VIE004 (Acc No. MK913648.1), Belagavi Voucher (Acc. No. MH753329. 1), and isolate Solok
243 (Acc. No. MW876212.1) (Nelly et al. 2021) and Frug Adiluwih Unila (Acc. No. MZ501588)
244 (Lestari et al. 2020). Group II consisted of 3 isolates (AaFaw, PaFaw, TpFaw), and was in the same
245 group as the corn strain haplotype 1 (Acc. No. U72974.1), the other reference corn strain 1L-LepS6
246 (Acc. No. KX580614.1), Tanah Datar isolate (Acc. No. MW876210.1) (Nelly et al., 2021) and IS 1
247 (Acc. No. LC546855.1) (Fig. 4).
248 We confirmed the presence of rice and corn strain in South Sumatra as well as West Sumatra.
249 This is the first report of the occurrence of the rice and corn strain of *S. frugiperda* from South
250 Sumatra. This study also revealed that isolates of *S. frugiperda* in Lampung Province were in the
251 group of a rice strain. Since the prompt spread of this pest, the rice strain is now may exist in the
252 other areas in Sumatra Island, including Lampung province as well as the other corn producing
253 areas in Indonesia. Comprehensive further study should be performed to confirm the presence of
254 both rice and corn strains in all corn producing areas in Indonesia. Further study of strains or
255 genetic diversity of fall armyworm in Indonesia will also provide valuable information on host plant
256 preferences and the indigenous natural enemies for new association with *S. frugiperda*.
257
258 ***3.4. Maize damage caused by Spodoptera frugiperda***
259
260 The surveys conducted at 10 locations in the lowlands and highlands in South Sumatra showed
261 that all locations were invaded by *S. frugiperda.* The mean incidence of *S. frugiperda* in each
262 location were significantly different (P = 0.00174) (Table 4). The highest incidence was found in
263 Suka Rejo, Pagar Alam City (100%), while the lowest was in Sukarami, Palembang City (44.0%).
264 The severity found on all locations ranged from 11.5% to 65% and were also significantly different
265 (P < 0.0001). In the lowlands and highlands of South Sumatra, the incidence and severity of *S.*
266 *frugiperda* tended to be high in all locations. It means that outbreaks of *S. frugiperda* have occured
267 in the South Sumatra. Therefore, the altitude of the location did not affect the severe or mild attack
268 of this FAW. The observation during the surveys revealed that the egg mass was laid by the adult
269 females on the leaf surface (Fig. 5A). The larvae found in the fields attacked the leaves and whorl
270 (Fig. 5B−C). The attack symptoms by the larvae showed typical characteristics, namely holes used
271 by the larvae on the leaves and leaves with transparent bite marks. On the stems or leaves there
272 were brown larval frass similar to sawdust (Fig. 5D). Young leaves that were still curled up could

273 also be attacked by the larvae as a result of the leaf rolls forming the holes. The larvae also
274 perforated the maize stalks (Fig. 5E), flower (F), cobs (Fig. 5G–H), funnel damaged (I). The severe
275 attack found in this study was in the vegetative phase, while in the generative phase the attack was
276 low. However, in this survey the observation of attacks was limited to maize aged 3–6 weeks.

Table 2.

Similarity among *Spodoptera frugiperda* collected from South Sumatera, West Sumatera, Lampung, Indonesia and other countries including rice and corn strain isolates.

Isolate	Similarity (%)															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	100															
2	100	100														
3	100	100	100													
4	100	100	100	100												
5	100	100	100	100	100											
6	100	100	100	100	100	100										
7	98.35	98.35	98.35	98.35	98.35	98.35	100									
8	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100								
9	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100							
10	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100						
11	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100					
12	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100				
13	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100			
14	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100		
15	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	
16	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	100

1. Corn haplotype 1 (Maize, USA) (Acc. No. U72974), 2. AaFaw (Maize, South Sumatera) (Acc. No. MZ497020), 3. PaFaw (Maize, South Sumatera) (Acc. No. MZ497021), 4. TpFaw (Maize, South Sumatera) (Acc. No. MZ497022), 5. Isolate Tanah Datar (Maize, West Sumatera) (Acc. No. MW876210), 6. IS_1 (Sugarcane, Japan) (Acc. No. LC546855), 7. Rice (USA) haplotype 2 (Acc. No. U72978), 8. Rice (USA) haplotype 1 (Acc. No. U72977), 9. FawPram (Maize, South Sumatera) (Acc. No. MZ497023), 10. LasFaw (Maize, South Sumatera) (Acc. No. MZ4970224), 11. MeFaw (Maize, South Sumatera) (Acc. No. MZ497025), 12. PuFaw (Maize, South Sumatera) (Acc. No. MZ497026), 13. SFaw (Maize, South Sumatera) (Acc. No. MZ497027), 14. TsFaw (Maize, South Sumatera) (Acc. No. MZ497028), 15. Isolate Solok (Maize, West Sumatera) (Acc. No. MW876212), 16. Frugi_Adiluwih_UNILA (Maize, Lampung) (Acc. No. MZ501588). Length of the nucleotides: 607bp.

Table 3.

Nucleotides difference between *Spodoptera frugiperda* from South Sumatera and foreign countries.

Isolate	Accesion Number	Position of nucleotide difference										
		11	56	110	146	197	428	503	509	539	573	602
Corn haplotype 1 (USA)	U72974	G	G	T	T	C	T	T	C	C	T	T
AaFaw (Maize, South Sumatera)	MZ497020	.	.	.	.	.	.	.	.	.	.	.
PaFaw (Maize, South Sumatera)	MZ497021	.	.	.	.	.	.	.	.	.	.	.
TpFaw (Maize, South Sumatera)	MZ497022	.	.	.	.	.	.	.	.	.	.	.
Isolate Tanah Datar (Maize, West Sumatera)	MW876210	.	.	.	.	.	.	.	.	.	.	.
IS_1 (Sugarcane, Japan)	LC546855	.	.	.	.	.	.	.	.	.	.	.
Rice haplotype 2 (USA)	U72978	A	A	.	A	T	C	C	T	T	C	A
Rice haplotype 1 (USA)	U72977	A	A	C	A	T	C	C	T	T	C	A
FawPram (Maize, South Sumatera)	MZ497023	A	A	C	A	T	C	C	T	T	C	A
LasFaw (Maize, South Sumatera)	MZ4970224	A	A	C	A	T	C	C	T	T	C	A
MeFaw (Maize, South Sumatera)	MZ497025	A	A	C	A	T	C	C	T	T	C	A
PuFaw (Maize, South Sumatera)	MZ497026	A	A	C	A	T	C	C	T	T	C	A
SFaw (Maize, South Sumatera)	MZ497027	A	A	C	A	T	C	C	T	T	C	A
TsFaw (Maize, South Sumatera)	MZ497028	A	A	C	A	T	C	C	T	T	C	A
Isolate Solok (Maize, West Sumatera)	MW876212	A	A	C	A	T	C	C	T	T	C	A
Frugi_Adiluwih_UNILA (Maize, Lampung)	MZ501588	A	A	C	A	T	C	C	T	T	C	A

Length of the nucleotides: 607bp

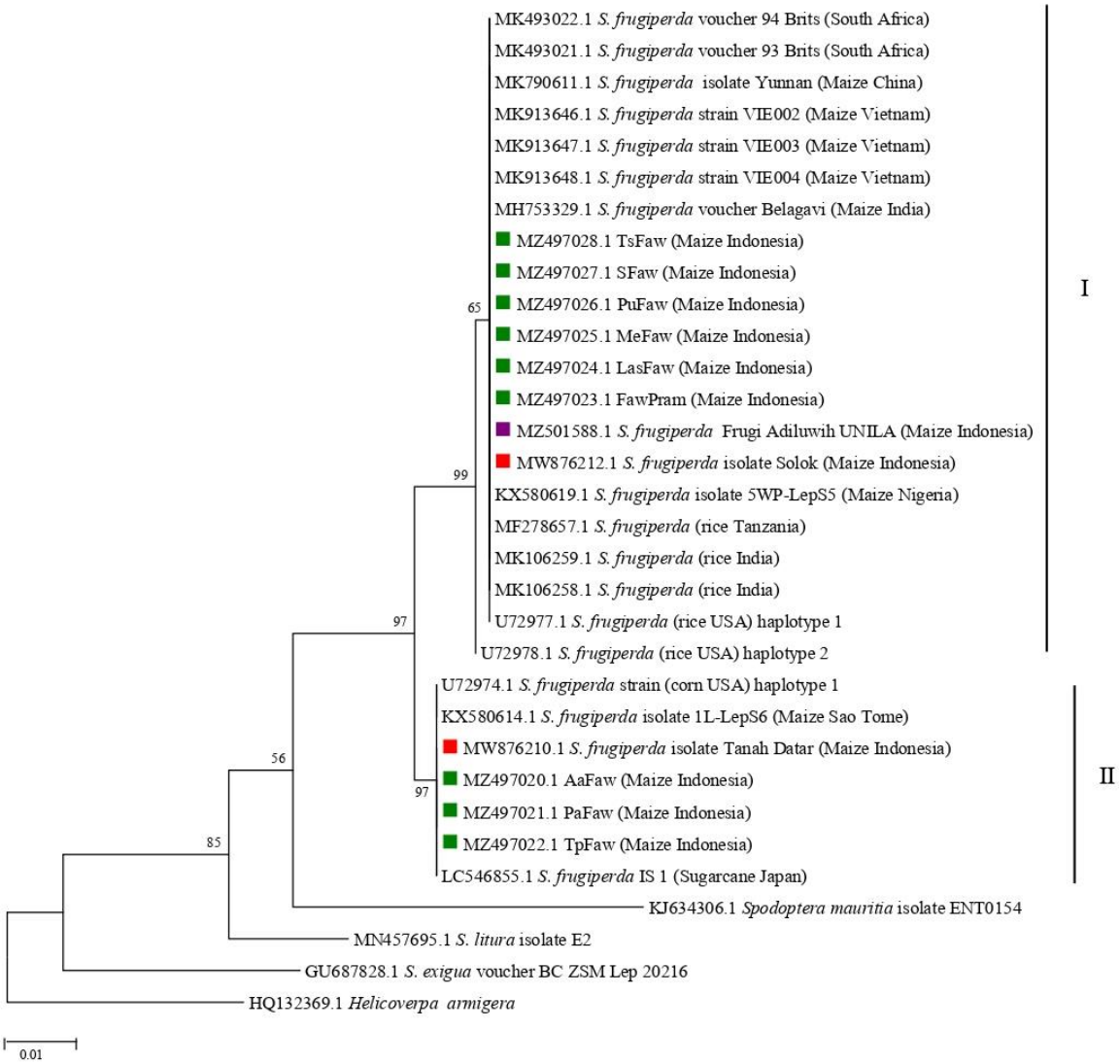

Fig. 4. Phylogenetic tree developed based on mitochondrial Cytochrome Oxydase I gene by maximum Likelihood method (1000X bootstrap; Tamura-Nei model) using Mega7 for windows (Kumar et al 2016). The *Spodoptera frugiperda* isolates collected from South Sumatera were divided into 2 groups (I and II). The group I belong to the clade of "rice strain" meanwhile the group II was a member of the clade of "corn strain". *Helicoverpa armigera* (Acc. No. HQ132369.1) was used as outgroup. The *Spodoptera frugiperda* used in this study. The *Spodoptera frugiperda* from West Sumatera, Indonesia. The *Spodoptera frugiperda* from Lampung, Indonesia.

Table 4.

Damage by *Spodoptera frugiperda* in South Sumatra, Indonesia

Survey sites (village, district/city)	Survey date	Altitude	Coordinate	Mean incidence[a]	Mean severity[b]

		(m)		(% of infested plant)	(on a scale of 1 to 5)
Sukarame, Palembang	14-05-2021	32.0	2°55′07″S 104°16′02″E	44.0c	11.5d
Curup Jare, Pagar Alam City	30-05-2021	782.0	103°13'17.0904"E 4°0'58.7556"S	70.0abc	22.5bcd
Suka Rejo, Pagar Alam City	30-05-2021	708.7	103°14′589″E 4°1′066″S	100.0a	65.0a
Tanjung Seteko, Ogan Ilir District	23-05-2021	23.3	3°13'08"S 104°41'01"E	82.0abc	38.7b
Tanjung Pering, Ogan Ilir District	23-05-2021	35.0	104°38'29.058"E 3°12'47.1132"S	85.6abc	38.4b
Gunung Ibul, Prabumulih City	17-06-2021	44.9	104°17'12.2856"E 3°24'52.3008"S	66.0abc	20.0bcd
Nantigiri, Lahat City	31-05-2021	740.0	3°56'22"S 103°12'15"E	88.0ab	65.0a
Muara Harapan, Muara Enim District	17-06-2021	99.8	3°38'01"S 103°49'24"E	84.0abc	27.5bc
Telang Sari, Banyuasin District	16-06-2021	24.5	104°38'40.2036"E 2°31'4.6596"S	50.0bc	22.5bcd
Mulyasari, Banyuasin District	16-06-2021	27.0	104°39'27.2772"E 2°33'4.6764"S	45.3bc	17.5cd

Note: Means of [a]incidence and [b]severity of damage plants by different survey sites labelled by same letter in one column for each mean are not significantly different from each other at (alpha 0.05) based on the Tukey's honestly significant test (HSD) and back-transformed means

The symptoms of *S. frugiperda* larvae attack in this study had the same characteristics as those of *S. frugiperda* found by Ginting et al. (2020) and Sartiami et al. (2020). The attack began with the larvae perforating the young leaves of the plant, and then perforating the young leaves that were still curled up, and at the worst the larvae cut the growing point of the maize (Ginting et al., 2020). Supartha et al. (2021) stated that the severity of attack by the larvae reached its peak when the maize was 4 weeks old, then the attack continued to decrease and at 8 weeks or more the attack was very low. The larval population dynamics follows the same pattern during the season, the larval population peaks three times, in the 14 and 21 days, in the 42 and 49 days and in the 77 days after planting (Dassou et al., 2021). During the survey, generally the severely affected maize was aged 3-6, the fruit bearing maize showed low attack.

This survey data showed that there was no consistent effect of the altitude of the survey sites with the severity of *S. frugiperda* attacks. However, Supartha et al. (2021) reported that in the highlands (> 500 m below sea level) there was no attack but in the lowlands this FAW attack was very high. This study has not been able to conclude the effects of corn and rice strain of *S.*

frugiperda on the incidence and severity of the FAW. Therefore, further research on the effects of corn and rice strain of *S. frugiperda* on the incidence and severity of the FAW needs to be performed. In addition, the range of host plants of these two strains also needs to be studied comprehensively.

Fig. 5. Symptoms damage by *Spodoptera frugiperda* larvae in maize: egg mass on the leaf surface (A), larvae feeding on leaves (B), larvae feeding on leaf whorl (C), brown larval frass similar to sawdust (D), larvae feeding on corn stalks (E), larvae feeding on corn flower (F), larvae feeding on corn cobs (G), larvae feeding on corn cob tip (H), and funnel damaged (I).

4. Conclusion

This study have found and confirmed the presence of rice and corn strain of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra, Indonesia. The incidence and severity of *S. frugiperda* from the lowlands to the highlands is high with the incidence reaching 100% and the severity reaching 65%. we also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. Comprehensive further study should be performed to confirm the presence of both rice and corn strains in all corn producing areas in Indonesia.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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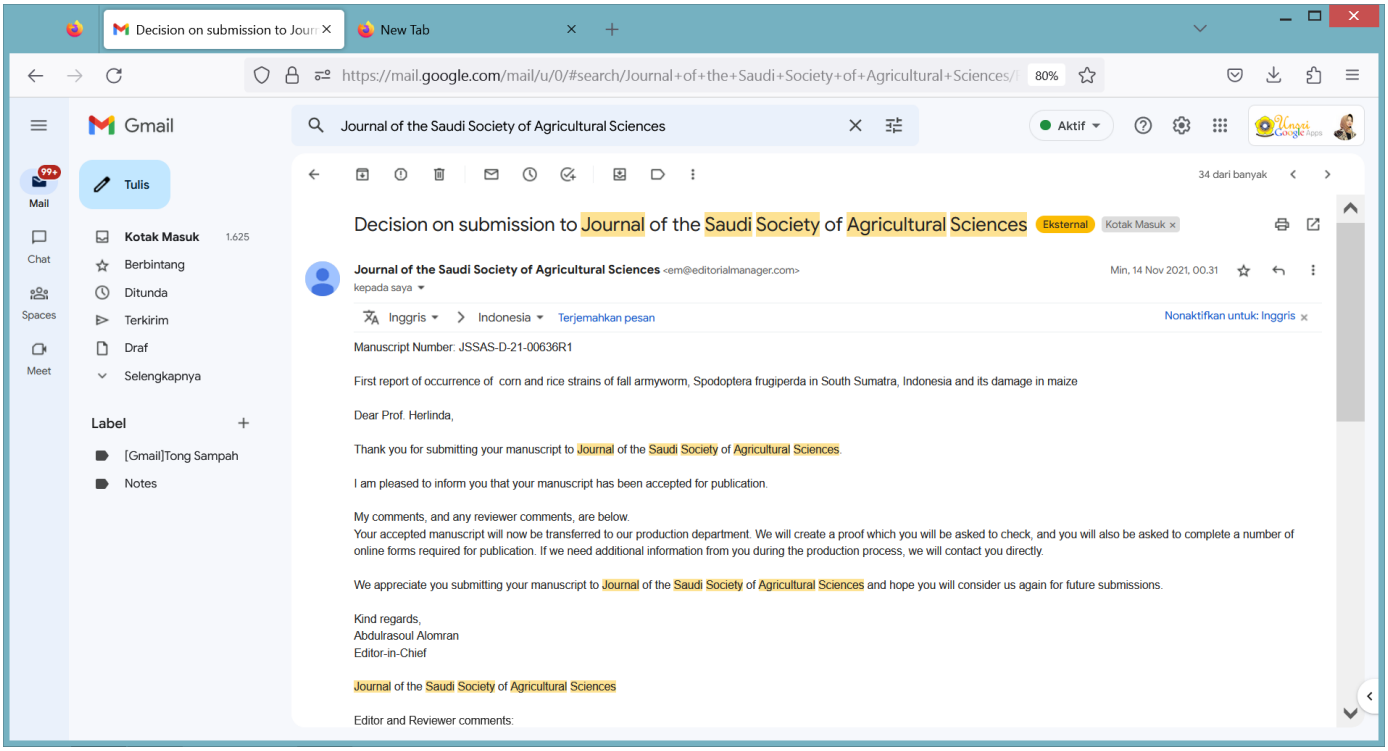

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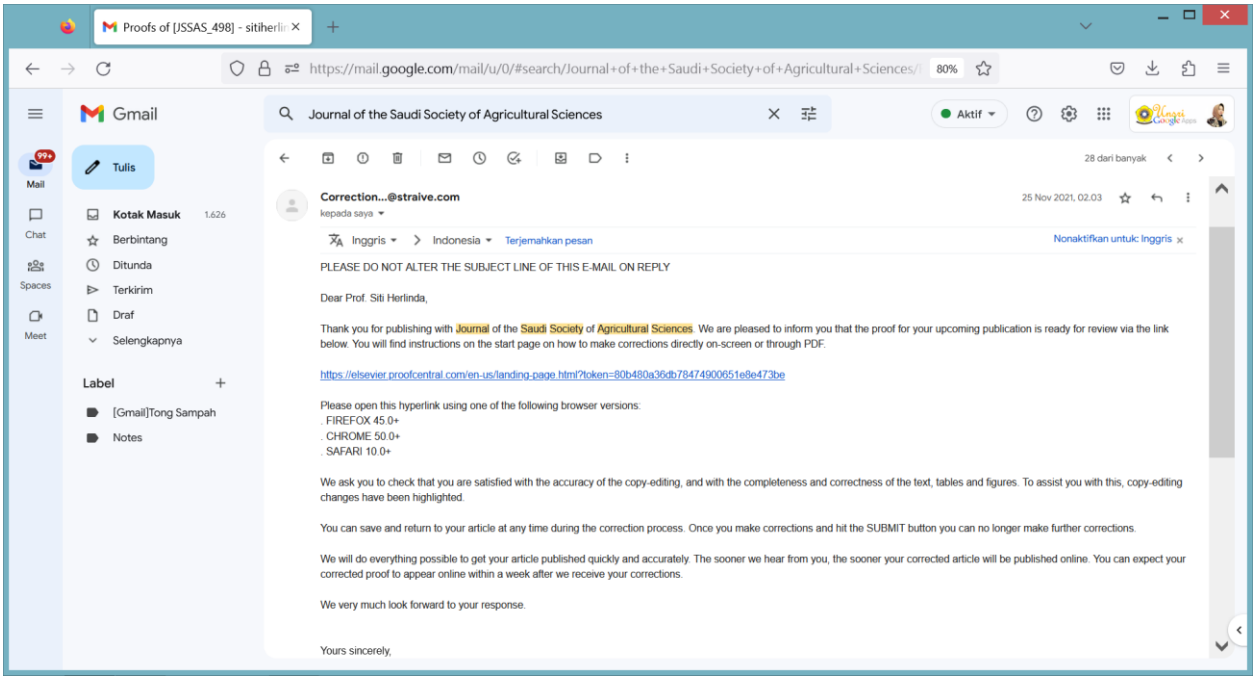

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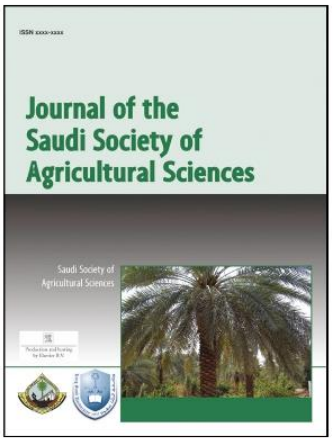

First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

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First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

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Declarations

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Consent for publication

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All data are available in the article and the materials used in this work are of high quality and grade.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Authors' contributions

SH performed research concept and design, writing the article, and final approval of article. RS prepared and performed molecular identification and data analysis and interpretation. MES and FF performed collection and assembly of data. SS prepared and performed morphological identification and critical revision of the article. All the authors read and approved the manuscript.

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First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize

ABSTRACT

Spodoptera frugiperda is a new invasive pest in Indonesia and its severity in maize ranges from 26.50 to 100%. However, information on the strains or genetic diversity of the *S. frugiperda* in Indonesia is still very limited. This research aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize. Surveys from January to June 2021 were carried out from the lowlands to highlands of South Sumatra. The field scouting was performed to calculate the incidence of damage and to estimate the severity caused by *S. frugiperda* larvae. The severity was assessed using a visual rating scale from 1 (no damage) to 5 (plant stunting and funnel damaged severely). The *S. frugiperda* larvae was identified based on morphological characters and molecular techniques using sequence analysis of Cytochrome c Oxidase subunit I (*COI*) gene. All larvae collected from South Sumatra showed identical morphological characteristics identified as *S. frugiperda*. The sequence analysis results showed that the 6 isolates of *S. frugiperda* shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province. The other 3 isolates of *S. frugiperda* shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan). All isolates have been deposited in the GenBank. This study confirmed the presence of rice and corn strains of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra. We also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. The incidence and severity of *S. frugiperda* reached 100% and 65% respectively. Comprehensive further study should be performed to confirm the presence of both strains and their damage in all corn producing areas in Indonesia.

Keywords: genetic diversity; incidence; outbreaks; scouting; severity

1. Introduction

Fall armyworm (FAW), *Spodoptera frugiperda* is a new invasive maize pest in Indonesia. This insect pest comes from the American continent (Nagoshi et al., 2017; Otim et al., 2018). In 2016, the FAW was reported to have come into Africa (Goergen et al., 2016). In 2017, FAW crossed over to Europe (Early et al., 2018). This pest began to move into Asia in 2018 (Mahat et al., 2021) and was first discovered in India (Ganiger et al., 2018) and came into Indonesia for the first time on March 26, 2019 in West Sumatra (Sartiami et al., 2020). Then, it began to spread to other provinces and islands in Indonesia, such as South Sumatra (Hutasoit et al., 2020), West Java (Maharani et al., 2019), Lampung (Trisyono et al., 2019), Bengkulu (Ginting et al., 2020), Bali(Supartha et al., 2021).

In addition to spreading throughout the world, the FAW has caused maize yield losses of up to 18 million tons/year and losses of up to 13 million US$ in 12 African countries (Harrison et al., 2019). In Kenya, the loss due to this pest reaches 1 million ton/year (De Groote et al., 2020). Besides attacking the maize, this pest attacks paddy, sugarcane, cotton, and ornamental plants (IPPC, 2019). In Brazil it has been reported that about 76 plant families were destroyed by this pest (Montezano et al., 2018). In Indonesia, the FAW generally attacks maize with damage in Lampung ranging from 26.50% to 70% (Lestari et al., 2020), in Bali reaching 47.84% (Supartha et al., 2021), in East Nusa Tenggara ranging from 85% to 100% (Mukkun et al., 2021). This pest can also attack paddy leaves, for example, in Banten the FAW larvae was found to attack paddy (Sartiami et al., 2020); yet there is no information on damage by this pest to paddy in Indonesia.

There are two strains of *S. frugiperda* in the world, namely corn strain (C) and rice strain (R) (Unbehend et al., 2013; Va et al., 2014). The genetic diversity of *S. frugiperda* in Indonesia was first reported by Sartiami et al. (2020) stating that the strain of *S. frugiperda* found in Banten was only the rice strain. In Lampung, the strain of *S. frugiperda* that was found from maize (Lestari et al., 2020) was also confirmed as the rice strain, while both the corn and rice strains were collected from maize production centers in West Sumatera (Nelly et al., 2021). In Indonesia, maize is one of important crops which is widely benefitted as a raw material in the feed and food

industries as well as staple food in some regions (Nurina et al., 2021). The corn and rice strains of *S. frugiperda* if they have spread in Indonesia can harm not only maize but also paddy and other important crops. However, information on the strains/genetic diversity of *S. frugiperda* in Indonesia is still very limited and until now the information on the strain of *S. frugiperda* originating from South Sumatra and its attack has not been reported. For this reason, information on the genetic diversity of the FAW in South Sumatra and its attacks is needed so that it can be used as a basis for controlling this pest and can complement information on *S. frugiperda* strains in Indonesia. This study aimed to identify the genetic diversity of *S. frugiperda* from South Sumatra and determine its damage in maize.

2. Materials and methods

2.1. Survey sites

Surveys to obtain specimens of *S. frugiperda* larvae were carried out from the lowlands to the highlands of South Sumatra, such as Palembang City (2°59′27.99″S 104°45′24.24″E), Pagar Alam City (3°52′43.8″S 103 °21′30″E), Lahat City (3.78639°S 103.54278°E), Ogan Ilir District (3.43186°S 104.6727°E), Prabumulih City (3.4328°S 104.2356°E), Muara Enim District (4.2327°S 103.6141 °E), and Banyuasin District (2.8833°S 104.3831°E) (Fig. 1 and Table 1). The survey started from January to June 2021 covering rainy (January to February), transition (March to June) and early dry (June) seasons. The specimens obtained were then identified molecularly at the Laboratory of Agricultural Biotechnology (accredited according to the ISO 17025 standard), Department of Plant Protection, Faculty of Agriculture, Lampung University, Indonesia.

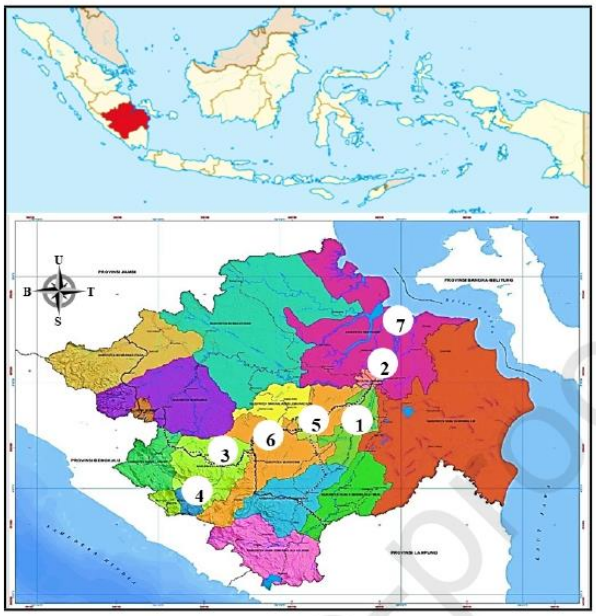

Fig. 1. Surveys locations in South Sumatra, Indonesia: Ogan Ilir District (1), Palembang City (2), Lahat District (3), Pagar Alam City (4), Prabumulih City (5), Muara Enim District (6), dan Banyuasin District (7).

2.2. Morphological identifications for Spodoptera frugiperda

S. frugiperda larvae were collected from maize fields in various districts/cities in South Sumatra, Indonesia. The larvae were brought to the Entomology Laboratory, Department of Plant Protection, Faculty of Agriculture, Universitas Sriwijaya, Indonesia to be reared individually in porous plastic cups (Ø 6.5 cm, height 4.6 cm). Maize leaves (2 cm x 5 cm) were placed into the cup to feed *S. frugiperda* and then the larvae were observed for further identification. Some samples of larvae were put into vials containing 70% alcohol for molecular identification at the Laboratory of Agricultural Biotechnology, Department of Plant Protection, Faculty of Agriculture, Universitas Lampung. Some other samples of larvae from the same population were kept until the adult completed one life cycle following the method of Gustianingtyas et al. (2021) to observe the morphology of the adult.

Table 1.

The origin of sample of *Spodoptera frugiperda* from South Sumatra, Indonesia.

Location (village, district/city)	Coordinate	Sample ID	Sample source	Species	GenBank Acc. No.
Alang-alang Lebar, Palembang City	2°59'27"S 104°45'24"E	AaFaw	Maize	*Spodoptera frugiperda*	MZ497020
Pagar Alam, Pagar Alam City	3°52′43.8″S 103°21′30″E	PaFaw	Maize	*Spodoptera frugiperda*	MZ497021
Tanjung Pering, Ogan Ilir District	104°38'29.058"E 3°12'47.1132"S	TpFaw	Maize	*Spodoptera frugiperda*	MZ497022
Prabumulih, Prabumulih City	3.4328°S 104.2356°E	FawPram	Maize	*Spodoptera frugiperda*	MZ497023
Lahat, Lahat City	3.78639°S 103.54278°E	LasFaw	Maize	*Spodoptera frugiperda*	MZ497024
Muara Enim, Muara Enim District	4.2327°S 103.6141°E	MeFaw	Maize	*Spodoptera frugiperda*	MZ497025
Purwasari, Banyuasin District	2°30'47.268"S 104°40'58.9296"E	PuFaw	Maize	*Spodoptera frugiperda*	MZ497026
Sukarami, Palembang City	2°54'35.3016"S 104°42'14.976"E	SFaw	Maize	*Spodoptera frugiperda*	MZ497027
Tanjung Seteko, Ogan Ilir District	3°13'08"S 104°41'01"E	TsFaw	Maize	*Spodoptera frugiperda*	MZ497028

2.3. Molecular identifications for Spodoptera frugiperda

2.3.1. DNA Extraction

DNA extraction was carried out based on the method of Lestari et al. (2020) with several modifications. *S. frugiperda* larvae that had been preserved in 70% alcohol solution were taken and dried on a tissue for 30 minutes. After that, the caterpillars were soaked in hot water (85 ºC) for 30 minutes until they got slightly whitish in color. Two abdominal segments were then cut and inserted into a 1.5 µl tube. A total of 5 µl Proteinase K was added and crushed until completely crushed. After being crushed, 300 L of TNES buffer was added (Tris HCl 1M(pH 7.5), NaCl 5M, EDTA 0.5 M,ddH2O, and 20% SDS), homogenized and incubated at 60 °C for 3 hours. After the incubation, 85 µL of 5 M NaCl was added and then shaken by hand for 15 seconds and centrifuged for 10 minutes at 14000 rpm. A total of 400 µL of supernatant was taken, put into a new tube and added Isopropanol as much as 60% of the taken volume of supernatant and put in a -40 °C freezer for 20 minutes. After that, it was centrifuged for 5

minutes at a speed of 14000 rpm. The supernatant was then discarded, added 500 µL of cold 70% alcohol and centrifuged for 5 minutes at 14,000 rpm. The supernatant was then discarded and dried at room temperature for 24 hours (one night). After drying, 20 µL buffer TE (1st Base, Malaysia). Before being used, the DNA suspension was stored at -4 °C. The centrifugation process was carried out using Microspin12 (Biosan, Latvia).

2.3.2. DNA amplification

DNA amplification was performed to amplify the Cytochrome Oxydase Subunit I (COI) region using LCO 1490 and HCO 2198 primers (Folmer et al., 1994). PCR was performed using a Sensoquest Thermal Cycler Machine (Germany) with a total volume of 25 µL consisting of 1 µL DNA, 12.5 µL master mix (2x MyTaq HS Red Mix, Bioline, USA), 1 µL of each primer LCO 1490 and HCO 2198 (Folmer et al., 1994) with a concentration of 10 M and 9.5 µL of sterile distilled water. The PCR was carried out in stages: 1 cycle initiation at 95°C for 5 minutes, followed by 30 cycles consisting of denaturation at 95°C for 1 minute, annealing at 54°C for 1 minute, extension at 72°C for 1 minute and followed by 1 elongation cycle at 72 °C for 5 minutes. The PCR results were then electrophoresed using a 0.5% agarose gel suspension that had been given 1 µL of ethidium bromide (ETBr; 10 mg/mL, per 20 mL agarose) at 55 volts for 70 minutes. The results were then visualized using a DigiDoc UV transilluminator (UVP, USA).

2.3.3. Sequencing and data analysis of sequencing results

The obtained PCR results were then sent to 1st Base Malaysia for the sequencing process. The obtained sequencing results were analyzed using the Bio Edit ver. 7.2.6 for windows (Hall, 1999). The results of the analysis were then submitted to the Basic Local Alignment Search Tool (BLAST) (https://blast.ncbi.nlm.nih.gov/Blast.cgi) to determine their possible identity. The phylogeny tree was created using the Mega 7 for Windows program (Kumar et al., 2016) using the maximum Likelihood method (1000X bootstrap; Tamura-Nei model). The reference strains used in this study were obtained from NCBI (https://www.ncbi.nlm.nih.gov/).

2.4. Observation of Spodoptera frugiperda in maize fields

2.4.1. Damage by Spodoptera frugiperda

The observations of *S. frugiperda* attacks were carried out from the lowlands to the highlands in 10 locations in South Sumatra, namely Sukarami, Palembang City; Pagar Alam City (Curup Jare and Suka Rejo); Ogan Ilir District (Tanjung Seteko and Tanjung Pering); Prabumulih. City (Gunung Ibul), Lahat City (Nantigiri); Muara Enim District (Muara Harapan); Banyuasin District (Telang Sari and Mulyasari). From each location, the sample land was taken with an area of 1−5 ha per location and the age of the selected maize ranged from 3 to 6 weeks following Lestari et al. (2020). The observation of attacks was carried out directly using a scouting system. The scouting system was chosen because the survey area was large and located in many locations (Kuate et al., 2019) and the scouting protocol follows the guidelines of Prasanna et al. (2018). The field scouting was performed to calculate the percentage of infested plant or incidence of damage and to estimate the intensity of attack or severity caused by *S. frugiperda* larvae (Kuate et al., 2019). The maize fields were scouted using a “W” pattern approach and the total sample observed was 50 plants (10 consecutive plants at five different spots along the “W” transect) (Prasanna et al., 2018). Damage to the plants was distinguished by severity of pin holes, shot-holes, lesions, tattering and dead hearts. The percentage of severity or attack intensity was calculated using a rating scale for scoring of damage severity on whorl-stage plants (Kuate et al., 2019).

The percentage of plants infested by FAW larvae termed as an incidence was measured by calculating total of infested plants divided by the total plants observed and multiplied by 100% while the percentage of severity was calculated by dividing the sum of score (excluding score 1) by the number of plants damage (Kuate et al., 2019). The visual rating scale of damage severity scored from 1 to 5 was used as follows: 1) no damage; 2) 1−10% leaf damage or < 5 mm diameter or only the leaf cuticle destruction; 3) 11−25% leaf damage with presence chewed areas > 5 mm, funnel leaves uninjured; 4) 26−50% leaf damage with presence chewed areas > 1 cm, the funnel less severe; and 5) > 50% leaf damage, plant stunting and funnel damaged severely (Kuate et al., 2019).

2.4.2. Data Analysis of Spodoptera frugiperda attack

Incidence and severity of *S. frugiferda* infestation was tested for normality using the Shapiro–Wilk test and for variance homogeneity by Levene's test. Square root transformation was performed to homogenous variance and to meet normality assumptions before being subjected to one-way analyses of variance. Means were compared using Tukey's honestly significant test (HSD) and back-transformed means were presented after analysis. R studio Version 1.4.1106 (RStudio PBC, Boston, MA, USA) was used for analyses of infestation data.

3. Results and Discussion

3.1. Morphological characteristics of Spodoptera frugiperda

The larvae collected from 9 survey sites in South Sumatra showed identical morphological characteristics and were identified as *S. frugiperda*. Morphological characters of all larvae found were characterized by the presence of four pinacula (black dots) on the eighth (second segment of the last segment) abdominal segment forming a square (Fig. 2A). The head of the larvae was dark and there was a single line forming a white inverted Y line on the head (Fig. 2B). The body of the larvae had pale yellow lines along the body dorsally, and yellow stripes subdorsal, thick bands (Fig. 2C). Based on the key to the morphological identification for larvae of *S. frugiperda* illustrated by Lestari et al. (2020), the larvae found in this study were identical to *S. frugiperda*.

The morphology of the larvae of this study was also the same as that of *S. frugiperda* as illustrated by Deshmukh et al. (2021) and Sartiami et al. (2020).

The adult *S. frugiperda* moths produced by the larvae rearing from the same colony as the genetic studies showed the following morphological characteristics, an adult male moth had gray-brown forewings with triangular white spots at the tip and mottled-colored (brown, light brown, dark gray) on the upper part forewing, while his hindwing was grayish white with brown outer margin (Fig. 3A). An adult female moth forewing was less distinctly marked and uniform grayish brown, while her hindwing was grayish white similar to the color of the male hindwing (Fig. 3B). The forewing and hindwing coloration of both sexes of adult FAW moths was the same as that of the FAW moths illustrated by Huesing et al. (2018), Lestari et al. (2020), Sartiami et al. (2020). The male and female wingspans documented in this study were also within the range (30 to 40 mm) of the observation result by Huesing et al. (2018).

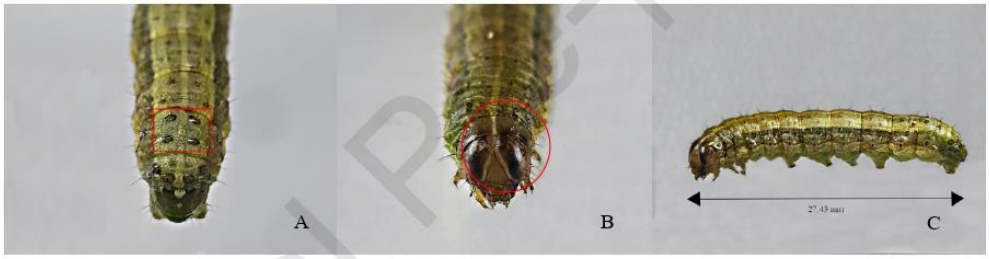

Fig. 2. Morphological characteristics of *Spodoptera frugiperda* larvae: four black spots on the last abdominal segment (A), inverted Y-shape on the head (B), longitudinal strips along the body (C)

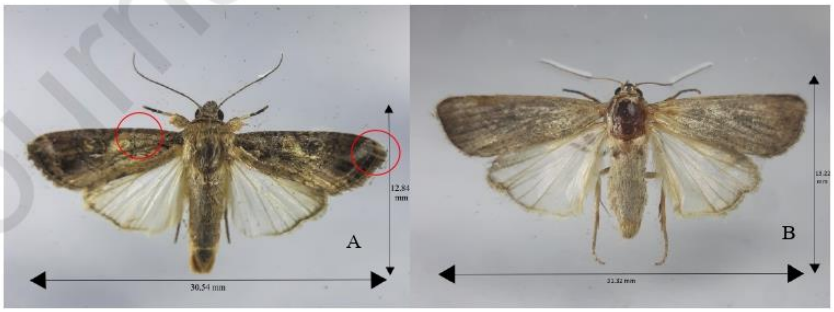

Fig. 3. Adult *Spodoptera frugiperda*: male (A) and female (B).

3.2. Molecular characteristics and genetic diversity of Spodoptera frugiperda

The sequence analysis results showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) were the same (100%). They also shared 100% of sameness as the rice strain haplotype 1, *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). They also shared 99.84% of similarity with rice strain haplotype 2 (Table 2).

The other 3 isolates from South Sumatra (AaFaw, PaFaw, TpFaw) were also identical (100% sameness) to each other. They shared 100% sameness as the corn strain haplotype I and IS 1 (obtained from sugarcane in Japan) and respective isolates of Group B from West Sumatra (Nelly et al., 2021). They shared 98.19% of similarity to the above mentioned 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), rice strain haplotype I and *S. frugiperda* isolate from Lampung Province (Lestari et al., 2020) and respective isolate of Group A from West Sumatra (Nelly et al., 2021). The 3 isolates also shared 98.35% of similarity to the rice strain haplotype 2 (Table 2).

The nucleotide difference showed that the 6 isolates of *S. frugiperda* (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram) and *S. frugiperda* from Lampung Province (Lestari et al., 2020) and respective isolates of Group A from West Sumatra (Nelly et al., 2021) were in the same pattern as the reference of rice strains. Meanwhile, the other 3 isolates (AaFaw, PaFaw, TpFaw) and the respective isolates of Group B from West Sumatra (Nelly et al., 2021) are in the same pattern as the reference of corn strains (Table 3).

The phylogenetic tree analysis revealed that the *S. frugiperda* isolates of South Sumatra were divided into 2 groups (I and II). The group I consisted of 6 isolates (TsFaw, SFaw, PuFaw, MeFaw, LasFaw, FawPram), and was placed within the groups of rice strain haplotype I (Acc. No. U72977.1) and haplotype 2 (Acc. No. U72978.1), the other reference rice strain voucher 93 Brits (Acc. No. MK493022), voucher 94 Brits (Acc. No. MK493021), isolate Yunnan (Acc. No. MK790611), VIE002 (Acc. No. MK913646), strain VIE003 (Acc. No. MK913647.1), strain VIE004 (Acc No. MK913648.1), Belagavi Voucher (Acc. No. MH753329. 1), and isolate Solok (Acc. No. MW876212.1) (Nelly et al. 2021) and Frug Adiluwih Unila (Acc. No. MZ501588) (Lestari et al. 2020). Group II consisted of 3 isolates (AaFaw, PaFaw, TpFaw), and was in the same group as the corn strain haplotype 1 (Acc. No. U72974.1), the other reference corn strain

1L-LepS6 (Acc. No. KX580614.1), Tanah Datar isolate (Acc. No. MW876210.1) (Nelly et al., 2021) and IS 1 (Acc. No. LC546855.1) (Fig. 4).

We confirmed the presence of rice and corn strain in South Sumatra as well as West Sumatra. This is the first report of the occurrence of the rice and corn strain of *S. frugiperda* from South Sumatra. This study also revealed that isolates of *S. frugiperda* in Lampung Province were in the group of a rice strain. Since the prompt spread of this pest, the rice strain is now may exist in the other areas in Sumatra Island, including Lampung province as well as the other corn producing areas in Indonesia. Comprehensive further study should be performed to confirm the presence of both rice and corn strains in all corn producing areas in Indonesia. Further study of strains or genetic diversity of fall armyworm in Indonesia will also provide valuable information on host plant preferences and the indigenous natural enemies for new association with *S. frugiperda*.

3.4. Maize damage caused by Spodoptera frugiperda

The surveys conducted at 10 locations in the lowlands and highlands in South Sumatra showed that all locations were invaded by *S. frugiperda*. The mean incidence of *S. frugiperda* in each location were significantly different ($P = 0.00174$) (Table 4). The highest incidence was found in Suka Rejo, Pagar Alam City (100%), while the lowest was in Sukarami, Palembang City (44.0%). The severity found on all locations ranged from 11.5% to 65% and were also significantly different ($P < 0.0001$). In the lowlands and highlands of South Sumatra, the incidence and severity of *S. frugiperda* tended to be high in all locations. It means that outbreaks of *S. frugiperda* have occured in the South Sumatra. Therefore, the altitude of the location did not affect the severe or mild attack of this FAW. The observation during the surveys revealed that the egg mass was laid by the adult females on the leaf surface (Fig. 5A). The larvae found in the fields attacked the leaves and whorl (Fig. 5B−C). The attack symptoms by the larvae showed typical characteristics, namely holes used by the larvae on the leaves and leaves with transparent bite marks. On the stems or leaves there were brown larval frass similar to sawdust (Fig. 5D). Young leaves that were still curled up could also be attacked by the larvae as a result of the leaf rolls forming the holes. The larvae also perforated the maize stalks (Fig. 5E), flower (F), cobs (Fig. 5G−H), funnel damaged (I). The severe attack found in this study was in the vegetative

phase, while in the generative phase the attack was low. However, in this survey the observation of attacks was limited to maize aged 3−6 weeks.

Table 2.
Similarity among *Spodoptera frugiperda* collected from South Sumatera, West Sumatera, Lampung, Indonesia and other countries including rice and corn strain isolates.

Isolate	Similarity (%)															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	100															
2	100	100														
3	100	100	100													
4	100	100	100	100												
5	100	100	100	100	100											
6	100	100	100	100	100	100										
7	98.35	98.35	98.35	98.35	98.35	98.35	100									
8	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100								
9	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100							
10	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100						
11	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100					
12	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100				
13	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100			
14	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100		
15	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	
16	98.19	98.19	98.19	98.19	98.19	98.19	99.83	100	100	100	100	100	100	100	100	100

1. Corn haplotype 1 (Maize, USA) (Acc. No. U72974), 2. AaFaw (Maize, South Sumatera) (Acc. No. MZ497020), 3. PaFaw (Maize, South Sumatera) (Acc. No. MZ497021), 4. TpFaw (Maize, South Sumatera) (Acc. No. MZ497022), 5. Isolate Tanah Datar (Maize, West Sumatera) (Acc. No. MW876210), 6. IS_1 (Sugarcane, Japan) (Acc. No. LC546855), 7. Rice (USA) haplotype 2 (Acc. No. U72978), 8. Rice (USA) haplotype 1 (Acc. No. U72977), 9. FawPram (Maize, South Sumatera) (Acc. No. MZ497023), 10. LasFaw (Maize, South Sumatera) (Acc. No. MZ4970224), 11. MeFaw (Maize, South Sumatera) (Acc. No. MZ497025), 12. PuFaw (Maize, South Sumatera) (Acc. No. MZ497026), 13. SFaw (Maize, South Sumatera) (Acc. No. MZ497027), 14. TsFaw (Maize, South Sumatera) (Acc. No. MZ497028), 15. Isolate Solok (Maize, West Sumatera) (Acc. No. MW876212), 16. Frugi_Adiluwih_UNILA (Maize, Lampung) (Acc. No. MZ501588). Length of the nucleotides: 607bp.

Table 3.

Nucleotides difference between *Spodoptera frugiperda* from South Sumatera and foreign countries.

Isolate	Accesion Number	Position of nucleotide difference										
		11	56	110	146	197	428	503	509	539	573	602
Corn haplotype 1 (USA)	U72974	G	G	T	T	C	T	T	C	C	T	T
AaFaw (Maize, South Sumatera)	MZ497020	.	.	.	.	.	.	.	.	.	.	.
PaFaw (Maize, South Sumatera)	MZ497021	.	.	.	.	.	.	.	.	.	.	.
TpFaw (Maize, South Sumatera)	MZ497022	.	.	.	.	.	.	.	.	.	.	.
Isolate Tanah Datar (Maize, West Sumatera)	MW876210	.	.	.	.	.	.	.	.	.	.	.
IS_1 (Sugarcane, Japan)	LC546855	.	.	.	.	.	.	.	.	.	.	.
Rice haplotype 2 (USA)	U72978	A	A	.	A	T	C	C	T	T	C	A
Rice haplotype 1 (USA)	U72977	A	A	C	A	T	C	C	T	T	C	A
FawPram (Maize, South Sumatera)	MZ497023	A	A	C	A	T	C	C	T	T	C	A
LasFaw (Maize, South Sumatera)	MZ4970224	A	A	C	A	T	C	C	T	T	C	A
MeFaw (Maize, South Sumatera)	MZ497025	A	A	C	A	T	C	C	T	T	C	A
PuFaw (Maize, South Sumatera)	MZ497026	A	A	C	A	T	C	C	T	T	C	A
SFaw (Maize, South Sumatera)	MZ497027	A	A	C	A	T	C	C	T	T	C	A
TsFaw (Maize, South Sumatera)	MZ497028	A	A	C	A	T	C	C	T	T	C	A
Isolate Solok (Maize, West Sumatera)	MW876212	A	A	C	A	T	C	C	T	T	C	A
Frugi_Adiluwih_UNILA (Maize, Lampung)	MZ501588	A	A	C	A	T	C	C	T	T	C	A

Length of the nucleotides: 607bp

Fig. 4. Phylogenetic tree developed based on mitochondrial Cytochrome Oxydase I gene by maximum Likelihood method (1000X bootstrap; Tamura-Nei model) using Mega7 for windows (Kumar et al 2016). The *Spodoptera frugiperda* isolates collected from South Sumatera were divided into 2 groups (I and II). The group I belong to the clade of "rice strain" meanwhile the group II was a member of the clade of "corn strain". *Helicoverpa armigera* (Acc. No. HQ132369.1) was used as outgroup. The *Spodoptera frugiperda* used in this study. The *Spodoptera frugiperda* from West Sumatera, Indonesia. The *Spodoptera frugiperda* from Lampung, Indonesia.

Table 4.

Damage by *Spodoptera frugiperda* in South Sumatra, Indonesia

Survey sites (village, district/city)	Survey date	Altitude (m)	Coordinate	Mean incidence[a] (% of infested plant)	Mean severity[b] (on a scale of 1 to 5)
Sukarame, Palembang	14-05-2021	32.0	2°55′07″S 104°16′02″E	44.0c	11.5d
Curup Jare, Pagar Alam City	30-05-2021	782.0	103°13'17.0904"E 4°0'58.7556"S	70.0abc	22.5bcd
Suka Rejo, Pagar Alam City	30-05-2021	708.7	103°14′589″E 4°1′066″S	100.0a	65.0a
Tanjung Seteko, Ogan Ilir District	23-05-2021	23.3	3°13'08"S 104°41'01"E	82.0abc	38.7b
Tanjung Pering, Ogan Ilir District	23-05-2021	35.0	104°38'29.058"E 3°12'47.1132"S	85.6abc	38.4b
Gunung Ibul, Prabumulih City	17-06-2021	44.9	104°17'12.2856"E 3°24'52.3008"S	66.0abc	20.0bcd
Nantigiri, Lahat City	31-05-2021	740.0	3°56'22"S 103°12'15"E	88.0ab	65.0a
Muara Harapan, Muara Enim District	17-06-2021	99.8	3°38'01"S 103°49'24"E	84.0abc	27.5bc
Telang Sari, Banyuasin District	16-06-2021	24.5	104°38'40.2036"E 2°31'4.6596"S	50.0bc	22.5bcd
Mulyasari, Banyuasin District	16-06-2021	27.0	104°39'27.2772"E 2°33'4.6764"S	45.3bc	17.5cd

Note: Means of [a]incidence and [b]severity of damage plants by different survey sites labelled by same letter in one column for each mean are not significantly different from each other at (alpha 0.05) based on the Tukey's honestly significant test (HSD) and back-transformed means

The symptoms of *S. frugiperda* larvae attack in this study had the same characteristics as those of *S. frugiperda* found by Ginting et al. (2020) and Sartiami et al. (2020). The attack began with the larvae perforating the young leaves of the plant, and then perforating the young leaves that were still curled up, and at the worst the larvae cut the growing point of the maize (Ginting et al., 2020). Supartha et al. (2021) stated that the severity of attack by the larvae reached its peak when the maize was 4 weeks old, then the attack continued to decrease and at 8 weeks or more the attack was very low. The larval population dynamics follows the same pattern during the season,

the larval population peaks three times, in the 14 and 21 days, in the 42 and 49 days and in the 77 days after planting (Dassou et al., 2021). During the survey, generally the severely affected maize was aged 3-6, the fruit bearing maize showed low attack.

This survey data showed that there was no consistent effect of the altitude of the survey sites with the severity of *S. frugiperda* attacks. However, Supartha et al. (2021) reported that in the highlands (> 500 m below sea level) there was no attack but in the lowlands this FAW attack was very high. This study has not been able to conclude the effects of corn and rice strain of *S. frugiperda* on the incidence and severity of the FAW. Therefore, further research on the effects of corn and rice strain of *S. frugiperda* on the incidence and severity of the FAW needs to be performed. In addition, the range of host plants of these two strains also needs to be studied comprehensively.

Fig. 5. Symptoms damage by *Spodoptera frugiperda* larvae in maize: egg mass on the leaf surface (A), larvae feeding on leaves (B), larvae feeding on leaf whorl (C), brown larval frass similar to sawdust (D), larvae feeding on corn stalks (E), larvae feeding on corn flower (F), larvae feeding on corn cobs (G), larvae feeding on corn cob tip (H), and funnel damaged (I).

4. Conclusion

This study have found and confirmed the presence of rice and corn strain of *Spodoptera frugiperda* and this is the first report of the occurrence of both strains in South Sumatra, Indonesia. The incidence and severity of *S. frugiperda* from the lowlands to the highlands is high with the incidence reaching 100% and the severity reaching 65%. we also found that outbreaks of *S. frugiperda* have occured in the South Sumatra. Comprehensive further study should be performed to confirm the presence of both rice and corn strains in all corn producing areas in Indonesia.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Palembang, 13 July 2021
Chief Editor
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Dear Editor,
We wish to submit an article entitled, "First report of occurrence of corn and rice strains of fall armyworm, *Spodoptera frugiperda* in South Sumatra, Indonesia and its damage in maize" for intended publication in Journal of the Saudi Society of Agricultural Sciences for your kind consideration.
We declare that we have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper
On behalf of the authors,
Prof. Dr. Siti Herlinda