# RESISTANT LEVEL OF SOYBEAN GERMPLASM AGAINST POD SUCKING BUGS (*Riptortus* spp.)

# Ketahanan Plasma Nutfah Kedelai Terhadap Kepik Pengisap Polong (Riptortus spp.)

Lina Herlina<sup>a\*</sup>, Bonjok Istiaji<sup>b</sup>, Dodin Koswanudin<sup>a</sup>, and Sutoro<sup>c</sup>

<sup>a</sup>Indonesian Center for Agricultural Biotechnology and Genetic Resources Research and Development
Jalan Tentara Pelajar No. 3A, Bogor 16111, West Java, Indonesia
phone: +62 251833797, Fax.: +62 251 833882

<sup>b</sup>Plant Protection Department, IPB University
Jalan Kamper - Kampus IPB Darmaga, Bogor 16680, West Java, Indonesia
<sup>c</sup>Binus University, Jalan. K H. Syahdan No. 9, Kemanggisan, Palmerah, Jakarta 11480, Indonesia

\* Correspondence author: tydars66@gmail.com

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# **ABSTRACT**

Increasing productivity of soybean has often been constrained by pod sucking bugs (Riptortus spp.) which caused a serious damage and yield losses up to 80%. Breeding for obtaining soybean variety resistant to pod suckers needs the availability of soybean germplasm resistant to the pest. The study aimed to obtain a candidate for soybean variety resistant to Riptortus spp. through the selection of 100 accessions of soybean. The study included the preparation of test plants and test insects, pest infestations, observations, and looking for a practical screening method for pod sucking pests. The experiment used a completely randomized design for two treatments (infested and non-infested Riptortus spp.). Cikuray variety and PI-092734 accession were used as a control. Results showed that there was a very low correlation among variables observed. Twelve soybean accessions showed a resistance to Riptortus spp., i.e. C7301-113AC-POP, Lokal Madiun-3549, Lokal Klungkung, ML.2974, Singgalang, Lokal Jepara, Lokal Jatim, Lokal Trenggalek, Lokal Tulungagung, Lokal Tabanan, Lokal Blitar, and Lokal Kuningan 10. These accessions were more resistant than the popular released variety such as Wilis, Grobogan, Detam 2, and Gepak Ijo. Small seed size was not a major determinant of soybean resistant to pod suckers. The addition of observational components, i.e. probing preference and oviposition, indicated that crop damage was indirectly influenced by the high frequency of probing and oviposition, although its relation to plant tolerant mechanisms still needs further investigation. Indeterminate plant types require further validation as to whether they contribute significantly to plant resistance against pod sucking insects.

[Keywords: oviposition, pod sucking insect, probing, *Riptortus*, soybean]

#### **ABSTRAK**

Peningkatan produktivitas kedelai sering terkendala oleh hama pengisap polong (Riptortus spp.) yang menyebabkan kerusakan parah dan kehilangan hasil hingga 80%. Upaya perakitan varietas kedelai tahan terhadap hama pengisap polong membutuhkan ketersediaan plasma nutfah kedelai yang tahan terhadap hama tersebut. Penelitian bertujuan untuk mendapatkan calon varietas kedelai tahan Riptortus spp. melalui seleksi terhadap 100 aksesi kedelai. Tahapan penelitian meliputi persiapan tanaman uji dan serangga uji, infestasi hama, observasi, dan mencari metode seleksi yang praktis untuk hama pengisap polong. Percobaan menggunakan rancangan acak lengkap untuk dua perlakuan (diinfestasi dan tanpa infestasi Riptortus spp.). Varietas Cikuray dan aksesi PI-092734 digunakan sebagai kontrol. Hasil penelitian menunjukkan bahwa terdapat korelasi yang sangat rendah antarvariabel yang diamati. Dua belas aksesi kedelai menunjukkan tingkat ketahanan terhadap hama pengisap polong, yaitu C7301-113AC-POP, Lokal Madiun-3549, Lokal Klungkung, ML.2974, Singgalang, Lokal Jepara, Lokal Jatim, Lokal Trenggalek, Lokal Tulungagung, Lokal Tabanan, Lokal Blitar, dan Lokal Kuningan 10. Aksesi ini lebih resisten dibandingkan dengan varietas populer yang telah dilepas seperti Wilis, Grobogan, Detam 2, dan Gepak Ijo. Ukuran biji kecil bukan penentu utama ketahanan kedelai terhadap pengisap polong. Penambahan komponen pengamatan yaitu preferensi probing dan oviposisi menunjukkan bahwa kerusakan tanaman secara tidak langsung dipengaruhi oleh tingginya frekuensi probing dan oviposisi, meskipun kaitannya dengan mekanisme toleransi tanaman masih perlu diteliti lebih lanjut. Tipe kedelai indeterminate memerlukan validasi lebih lanjut mengenai kontribusinya terhadap ketahanan tanaman terhadap serangga pengisap polong.

[Kata kunci: serangga pengisap polong, probing, oviposisi, kedelai, Riptortus]

#### INTRODUCTION

Over the decades, soybean (*Glycine max* (L) Merr) has been established as an important food commodity in Indonesia. It is the most important legume crop produced and consumed globally as an animal feed, cooking oil, and a component in many processed foods (Luthria et al. 2018). Soybean has also been proposed as one of

strategic food commodity by the Ministry of Agriculture, Republic of Indonesia, in addition to eleven other strategic food types, including rice, corn, shallots, garlic, large chilies, bird's eye chilies, beef/buffalo, purebred chicken, eggs, sugar, and cooking oil (Yohana and Bambang 2021). The increase in soybean productivity is absolutely necessary in line with the increasing demand for this commodity. Moreover, soybeans are one of the legumes with high nutritional content and are also important for industry, including being the only supplier of the highest vegetable protein in the world (Bae et al. 2014).

In relation with the government policy, efforts to increase soybean productivity were carried out by preparing a planting area of 325,000 hectares, covering dry land, rainfed land, intercropping land with maize and sugar cane, and on oil palm plantations. The planting areas are scattered in various parts of Indonesia, including West Sulawesi, Central Sulawesi, South Sulawesi, Central Java, West Java, East Java, South Kalimantan, West Nusa Tenggara, Lampung, Jambi, and Banten (Yohana and Bambang 2021). Increased production also means the potential for increased pest attacks, because the availability of more food sources also means opportunities for soybean pests to multiply their population. In cereal, yield losses caused by insect pest pressure on crops are predicted to increase by 10-25% per degrees due to global surface warming (Deutsch et al. 2000; Straub et al. 2020).

For soybean, several species of Riptortus spp. have been known as important pests that cause serious damage and yield losses (Maharjan and Jung 2009; Lee et al. 2009; Li et al. 2021). Insects that have similar shape to stink bugs are known as pod suckers and considered economically important pests of soybean (Kang et al. 2003). These pod suckers take advantage of the nutrients found in the pods and water in the soybean plant. Insects have a stylet-sheath which produces saliva which contains digestive enzymes that cause significant damage in the form of reduced yield, seed quality, and germination rate of soybean seeds (Bae et al. 2014). Yield losses due to these pests can reach up to 80% (Marwoto et al. 2014). However, usually Riptortus spp. cannot develop or reproduce if the plant does not produce seeds, presumably because seeds supply nutrients where the highest concentration of nutrients are contained in them compared to other parts of soybean plants (Mainali et al. 2014). Soybean seeds have a relatively higher protein and lipid contents than other legumes (Luthria et al. 2018; Min et al. 2020; Huang et al. 2019).

Riptortus clavatus (Thunberg) (Hemiptera: Alydidae) is reported to be a very detrimental pest, causing 89-

91% seed injury (Jung et al. 2005) and difficult to control with insecticides (Maharjan and Jung 2009). This species has the ability to "avoid" insecticide application. Usually insects quickly move around and stay away from the planted area when spraying with insecticides, but can quickly return and infest the area around the crop. *R. clavatus* is reported to have the ability to fly long distances, reaching a distance of 1.6-5.1 km with a speed of up to 0.8 m per second (Maharjan and Jung 2009). Several control efforts have also been reported, including using pheromone traps (Huh et al. 2005), exploiting natural enemies (Son et al. 2008), and using trap plants (Youn and Jung 2008). However, it turns out that all these control efforts are not effective enough to control this pest in the field.

Pest control through the use of resistant crops is one of the safest and most effective alternatives to develop. Unfortunately, breeding soybean for resistance against pod sucking pest in Indonesia has not maximally implemented. Until now, superior soybean varieties which also resistant against pod sucking bugs were still difficult to find. Although many superior soybean varieties have been produced, which reaching more than 50 varieties in Indonesia, it was only limited number of varieties which already known as resistant varieties, such as MLG 3032, IAC 80, and IAC 100 (Krisnawati et al. 2016).

To support the breeding program for resistance to pod suckers, it is absolutely necessary to provide soybean germplasm containing these resistant genes. Related to this, the research being conducted is still limited. Asadi (2009) reported several soybean varieties that potentially contain resistance genes against some pod sucking pests obtained from screening of 100 soybean accessions. Out of 100 accessions tested, only 17 accessions were reported to have resistance to pod sucking pests (Asadi 2009). This means that hard work is still needed to find new sources of resistance genes against pod sucking pests in Indonesia. This study aimed to obtain candidate for soybean genotype resistant to Riptortus spp. through the selection of 100 accessions of soybean collection. Those candidate would be useful for further research particularly to support breeding for soybean variety resistant to pod sucking pests. Plant resistance to Riptortus spp. in this study was determined by including additional variables derived from insect pest activity attributes, i.e. probing behavior and oviposition preference in scoring. This approach is also intended to find an available method for evaluation or screening plant responses to pest infestations which tested in large quantities of accessions, in a short time but was valid.

#### **MATERIALS AND METHODS**

#### **Plant Materials**

The research was conducted at the greenhouse of the Indonesia Center for Agricultural Biotechnology and Genetic Resources Research and Development (ICABIOGRAD), Bogor, from June until September 2015. The average temperature in the greenhouse was 24.6 °C in the morning and 36 °C in the afternoon, with the daily air humidity of 70-85% (data not shown). A total of 102 accessions of soybeans consisted of 100 accessions mainly from local varieties as tested materials and 2 accessions as check varieties were used in this study (Table 1)

### **Green House Assay**

The experiment was arranged in completely randomized design with two treatments. i.e. infested and non-infested with Riptortus spp., with three replications consisted of ten individual plants for each replication. Screening method was basicly adopted from IRRI (1996) with modification in insect infestation and scoring determination. Cikuray variety was used as a susceptible check, and PI-092734 was as a resistant check. The agronomic characters observed for all accessions were growth type (determinate, semi-determinate or indeterminate), 100 grain weight (g), seed size (small, medium and big), weight of total grains (bulk) harvested (g), and weight of sample grains harvested (g). Seed size was categorized differently from Susanto et al. (1994), because most accessions in this study were local landraces which had a smaller size. Here we classified seed type into three categories based on their 100 seed weight: small (< 7 g), medium (7 to < 9 g), and big (> 9 g).

Preparation was carried out by planting three seeds within each polybag contained 5 kg of soil (a mixture of filtered garden soil + compost with a ratio of 1: 1), while only one plant was maintained fertilizers consisted of 50 kg ha<sup>-1</sup> urea, 75 kg ha<sup>-1</sup> TSP, and 60 kg ha<sup>-1</sup> KCl were applied at planting. The plants were irrigated as needed to avoid water stress. Installation of stakes to prevent plant extension to other polybags due to relatively more elongated growth in the screenhouse (compared to the field) was done at 21 days after planting (DAP).

#### **Pest Infestation**

Preparation of the test insects was carried out by collecting the nymphs of *Riptortus* spp. as much as possible from the field, then kept in insect cages until

the first generation produced imago and is sufficient for testing. Insect maintenance was carried out by providing feed in the form of podded soybeans which is renewed every two days

Insect infestation was carried out simultaneously to the infested plants when the plants have formed 90% of pods (aged 55 DAP) by releasing 200 unsexed *Riptortus* spp. in the screenhouse. Two insect cages were placed in the greenhouse, then the gauze cover was slowly opened so that the insects get out of the cage. Insect food (soybean plants that have many pods) was removed from the cage. Insects were allowed to infest the plant for 21 days (until the plant enters harvest time at 90 DAP).

## **Observation of Damage Incidence**

The observed parameters related to plant resistance against *Riptortus* spp. were as follows:

a. Percentage of damage pods. Pods with damaged symptoms of being deflated/empty or yellowing or dry were picked and counted, while the healthy pods were counted and left to remain on the plant. Observations were started at 5 days after insect infestation. The percentage of damage pods was determined following the equation of Bayu et al. (2017).

$$N(\%) = \frac{a}{b} \times 100\%$$

Note:

N = percentage of damage pods (%);

a = number of damage pods;

b = number of all pods (healthy pods + damage pods)

- b. Frequency of pod-sucking pest probing. Probing is the insect activity to visit the plants to fulfill their food needs. Probing or visiting activities of *Riptortus* spp. to the plant were differentiated based on the plant parts exposed, i.e. pods, leaves, stalks, or stems. Observations were made by counting the number of *Riptortus* spp. in the plant or stalk, along with the insect stadia. Observations were made in the morning before 10:00 am, because insect is usually active in the morning (before the temperature is warmer).
- c. Preference for oviposition. Eggs present in the test plant parts were counted and collected in a small plastic bottle. Observations were made every two days, starting from seven days after insect infestation to the test crop.

The plant resistant level against *Riptortus* spp. was determined based on the scoring of the percentage of pod damage (Table 2) which was developed by modifying

Table 1. List of 100 soybean accessions used in this study.

No. of accession	Name of accession	Collection site (province)	No. of accession	Name of accession	Collection site (province
2627	MLG2627	West Java	3736	Lokal Bojonegoro-3736	East Java
2812	C7301-113AC-POP	West Java	3737	Lokal Bojonegoro-3737	East Java
2984	MLG2984	West Java	3739	Nona	East Java
3186	Kepet Hitam	West Java	3740	Otok	East Java
3189	Kepet Godek	West Java	3743	Si Nyonya	East Java
3191	Kedele Godek	West Java	3749	MLG.2965	East Java
3207	Kedele Susu	West Java	3758	ML.2974	East Java
409	No.3409	West Java	3762	N0.3762	East Java
3460	Wilis	Centra Java	3763	MLG.2981	East Java
3466	Kedele Hitam	Central Java	3764	No.3764	East Java
473	Lokal Jatim	East Java	3770	MLG.2995	East Java
489	Hitam	East Java	3774	M.2996	East Java
498	Kretek Balap	East Java	3799	Lokal (ML 3027)	East Java
549	Lokal Madiun-3549	East Java	3780	MLG.3002	East Java
568	GM.920 SI	West Java	3793	MLG.3019	East Java
582	No.29 ex Bogor	West Java	3796	ML.3024	East Java
583	29 ex Mojosari	East Java	3797	MLG.3025	East Java
585	Lokal Jombang	East Java	3800	ML.3028	East Java
588	Lokal Nganjuk	East Java	3803	M.3031	East Java
595	Lokal Magetan	Central Java	3806	M.3289	East Java
596	No.3596	East Java	3898	LB-80	Bali
598	Lokal Madiun-3598	East Java	4116	Pangrango	West Java
600	Lokal Madiun-3600	East Java	4194	Lokal Ongko-2	West Nusa Tenggara
605	Lokal Trenggalek	East Java	4227	Lokal Bima Hijau	East Nusa Tenggara
607	Lokal Tulungagung	East Java	4229	Kedelai Langkat	North Sumatra
610	Lokal Kediri	East Java	4283	Singgalang	West Sumatera
612	Lokal Pasuruan-3612	East Java	4295	Lokal Bombongan III-4295	South Sulawesi
623	Lokal Sumenep	East Java	4296	Lokal Bombongan III-4296	South Sulawesi
625	Lokal Bangkalan	East Java	4308	Lokal Bombongan II	South Sulawesi
627	No.3627	Unknown	4372	Sindoro	Unknown
634	MLG.2759	East Java	4391	GM.374 SI	West Java
640	Lokal Pasuruan-3640	East Java East Java	4391	GM.374 SI	West Java
641	Lokal Pasuruan-3641	East Java East Java	4400	GM.4779 SI	West Java
649	Lokal Banyuwangi	East Java	4401	GM.4779 SI GM.4783 SI	West Java
	, ,	East Java			West Java West Java
650	Lokal Jember Kretek		4402 4403	GM.4596 SI GM.4839 SI	West Java West Java
652 654	No.3654	East Java	4403	GM.363 SI	West Java
	Lokal Lumajang	Unknown East Java			Unknown
6660	Lokal Pasuruan-3662		4413	Lokal Kuningan 10	
662		East Java	4414	Lokal Cikapak	Unknown
6665	Lokal Pasuruan-3665	East Java	4415	Rajabasa	Unknown
666	Lokal Pasuruan-3666	East Java	4418	Lokal Jepara	Central Java
684	Lokal Klungkung	Bali	4423	Ijen	Unknown
686	Lokal Tabanan	Bali	4426	No.4426	Unknown
692	Lokal Badung	Bali	4427	No.4427	Unknown
699	Lokal Karangasem-3699	Bali	4429	No.4429	Unknown
3700	Lokal Karangasem-3700	Bali	4430	Detam 2	Unknown
3701	Lokal Karangasem-3701	Bali	4432	Gepak Ijo	Unknown
708	Lokal Buleleng	Bali	4434	MLG.3017	East Java
724	Lokal Blitar	East Java	4441	Grobogon	Central Java
3732	Lokal Madiun-3732	East Java	B 27	Cikuray (susceptible check)	West Java
3735	Lokal Ngawi	East Java	4596	PI-092734 ( resistant check)	Introduction from other country

Table 2. Scoring for percentage of soybean pods attacked by *Riptortus* spp., probing frequency, and oviposition activity.

Percentage of damage pods	Score (N1)	Criteria
0	1	Highly resistant (HR)
$0 \le x \le 5$	2	Resistant (R)
$5 < x \le 10$	3	Moderately resistant (MR)
$10 \le x \le 25$	4	Susceptible (S)
$X \ge 25$	5	Highly susceptible (HS)
Probing frequency	Score (N2)	
0	1	Highly resistant (HR)
$0 < x \le 1$	2	Resistant (R)
$1 \le x \le 2$	3	Moderately resistant (MR)
$2 \le x \le 3$	4	Susceptible (S)
$x \ge 3$	5	Highly susceptible (HS)
Oviposition activity	Score (N3)	
0	1	Highly resistant (HR)
$0 \le x \le 1$	2	Resistant (R)
$1 \le x \le 2$	3	Moderately resistant (MR)
$2 \le x \le 3$	4	Susceptible (S)
$x \ge 3$	5	Highly susceptible (HS)
Cumulative score (N cum)		
0		Highly resistant (HR)
$0 \le x \le 5$		Resistant (R)
$5 < x \le 7$		Moderately resistant (MR)
$7 < x \le 12$		Susceptible (S)
$x \ge 12$		Highly susceptible (HS)

the scoring by Chiang and Talekar (1980) whereis in this study the criteria applied were stringent in determining the resistant level.

For the determination of plant resistance to pests, the cumulative score (N cum) was used as one of the development of scoring method. The cumulative score is the sum of the respective scores from the percentage of damaged pods, probing frequency, and oviposition activity. The use of cumulative score aims to make a tighter selection in determining resistant accessions. We adopt this method from Herlina et al. (2021) who classify plant resistance to disease based on disease index scoring, by modifying it according to the resistance to pests.

#### **Data Analysis**

The data obtained were analyzed using one-way analysis of variance (ANOVA). Several yield component variables were analyzed based on the comparison of their mean values with the Tukey Pairwise Comparison method (95%)

confidence). The relationship between the observed variables were determined based on Pearson correlation analysis.

Resistant level of the accessions were determined based on cumulative score of percentage of infected pods, probing frequency, and oviposition activity. The resistance level was determined after all the values obtained were summed up, then grouped/sorted according to accessions with the largest to the smallest values. Ten accessions with the highest score were selected and considered to have a high level of resistance to *Riptortus* spp. based on the results of the grouping. All data analysis was performed using Minitab ver 19.

#### **RESULTS AND DISCUSSION**

#### **Yield Loss**

The results of mean analysis of yield components (total yield weight and total sample weight) based on Tukey's test to both infested and non-infested groups showed that the two groups were significantly different (Table 3 and 4).

The results showed that the soybean infested by *Riptortus* spp. had a lower yield compared to the non-infested plant. The decrease in the total yield weight reached 13.04 g (16.2%) and the decrease in the sample yield weight was 11.36 g (27.13%). This is suggested that *Riptortus* spp. infestation might significantly contribute to the decreasing soybean yields. Yield weight might be consider consisted of seed yield, while seed yield is one of the criteria for plant resistance to

Table 3. Mean analysis on total weight of soybean infested and not infested by *Riptortus* spp.

Total weight	N	St Dev	Mean	Grouping
Non-infested	102	22.34	80.49	A
Infested	102	19.96	67.45	В

Means that do not share a letter are significantly different based on Tukey comparison by 95% confidence level.

Table 4. Mean analysis on sample weight of soybean.

Sample weight	N	St Dev	Mean	Grouping
Harvested I	102	10.76	41.87	A
Harvested N	102	11.96	30.51	В

Means that do not share a letter are significantly different based on Tukey comparison by 95% confidence level. .

pests according to Da Fonseca Santos et al. (2018). These results are consistent with those reported by Maharjan and Jung (2009), Marwoto et al. (2014), Li et al. (2021) that *Riptortus* spp. causes yield loss which is worth considering for resistance.

According to Marwoto (2006), the sensitive phase of soybean against *Riptortus linearis* attack is during the pod initiation and pod filling phase, known with code as R5-R6. The presence of *Riptortus* infestation during this phase resulted in seed damage up to 15-20% and yield loss of 80%. Thus results of this study confirm that until now this statement is remain relevant.

In a normal situation, the yield is a complex character, which depends on the various characters associated with plant development and yield components. Seed yield is strongly influenced by various reproductive development processes as well as secondary characters that have a direct or indirect effect on yield formation (Sellamuthu et al. 2011). In addition, Sui et al. (2013) stated that increasing the yield potential is relatively more difficult if you only manipulate certain characters. From all of these statements it is clear that significant disruptions to the development of seeds contained in the

pods will have a very detrimental and fatal impact on yield. The damage caused by pod sucking pests also included.

## **Pod Sucking Bug Evaluation**

The average *Riptortus* spp. encountered by probing was 1.2 insects per accession during the observation period (Table 5), with the maximum number of probing was 4 insects per accession. The highest probing site preference was in leaves (Table 5) (Figure 2). This result is reasonable, because of all the sites evaluated, leaves were the most abundantly available and easily occupied by the insects and also support the probing activity of insects. As with oviposition, the probing site most entered by *Riptortus* spp. was leaves. The preference of insects to leaves than other plant parts indicates that leaves may have nutrients that are indispensable for insect pests.

Pods are the second most common choice for probing, after leaves, because they are also the second dominant in quantity (Table 5). Not all *Riptortus* stages require pods for their survival. Imago exploits more

Table 5. Probing and oviposition preference site and stadia of Riptortus sp. on soybean.

	Pro	obing	Ovipo	osition
Site preference	Frequency	%	Frequency	%
Stake	46	21.8	15	23.44
Stalk	32	15.17	3	4.69
Pod	56	26.54	11	17.19
Leaf	68	32.23	35	54.69
Other*	9	4.27	0	0
Stadia	Frequency	%		
Imago	95	92.23		
Nymph	8	7.77		

<sup>\*</sup>Gauze or bucket or soil









Fig. 2. Probing site of Riptortus spp. mostly on leaves and pods of soybean.

pods than nymphs. *Riptortus* of first instar nymphs do not even suck soybean pods (Rahman et al. 2017; Talekar et al. 1995) and their nutritional needs are mostly met from the leaves. Pod sucking activity usually begins when the insect has entered the second instar nymph stage (Rahman et al. 2017).

The term of probing in a broad definition is any behavior in which the mouth is in contact with food (Backus 2000). The duration of probing can be used as an indication of the preference of insects for plant parts. The longer the insect probes there, the part of the plant is the main site that the insect prefers. By implication, there is a high probability that these sites are also the most active in their defense in response to insect intimidation. This is a plant response as a form of plant resistance to herbivorous insects that interfere with it. Many studies have reported on this, where plants develop morphological resistance due to herbivorous insect infestations, namely in the form of the growth of mechanical protection on the plant surface in the form of thicker hair, trichomes, spines, and leaves, all of which are targeted to inhibit insect development (Dalin et al. 2008; Liu et al. 2010; Peschiutta et al. 2018). Likewise, it is suspected that soybean plants develop the same morphological resistance.

Apart from the morphological resistance that plants develop, another unique response of soybean to pest infestation is increasing their vegetative phase by keeping their leaves green or known as stay-green syndrome (Li et al. 2019). However, the interesting point is this accession actually showed quite dense leaf growth and longer green

color (both on the stalk and leaves) than the other accessions. It is suggested that the stay-green syndrome might play important role in tolerance mechanism of plant against insect infestation. Kuswantoro et al. (2020) also reported the presence of stay-green syndrome symptoms in soybean stalks in an evaluation of resistance to *Nezara viridula* (soybean pod sucker).

The oviposition activity by female imago reached 0.65 eggs per accession during the observation period with a maximum number of 6 eggs per accession (Table 5). Preferred site for oviposition was leaves (54.69%). This is in accordance with the results reported by Talekar et al. (1995) that in general imago prefers to lay eggs on leaves, especially leaves that appear on the fourth to sixth nodes (from the apex) on the stem.

The number of eggs laid was the same (equal) between the top surface and the bottom surface of the leaf (Talekar et al. 1995). But according to Marwoto (2006), *R. linearis* were laid eggs in groups on the bottom surface of leaf and in pods with a number of 3-5 grains. Egg was round shape with the center slightly concave, as shown in Figure 3. Freshly laid eggs are grayish blue, then changed to dreary brown, 1.20 mm diameter. Egg stage ranges from 6 to 7 days. The main consideration underlying the oviposition preference by the imago is the guarantee of the survival of the offspring. Leaves are preferred over other plant parts, possibly because they are the most readily available part and have a structure that is easily accessible to insects.



Fig. 3. Oviposition site preference of Riptortus spp. on soybean

In general, the small mean number of probing and oviposition per accession in Table 6 were probably due to the limited number of insects (Riptortus spp.) used in the study which was only 200 insects totally, comparing with observations in the Riptortus spp. endemic areas where each plant could be visited by 3-5 insects (the author's experience during field observations when collecting insects for testing materials for greenhouse assay). Therefore, based on the result of this study, we suggested that for future evaluation on the soybean resistance to *Riptortus* spp. in the screenhouse, at least the researcher will require larger number of test insects (minimum 500 individuals of insects for assessing 100 accessions of soybean). Li et al. (2021) stated that damage intensity on soybeans increased with increased pest density, which inferred that pest density highly contributed to the resistance measurement accurately.

The resistance level of 100 soybean accessions in this study was determined based on some parameters included probing activity, oviposition, and damaged pods. We determined resistance by including several attributes of pest behavior in this case probing activity and oviposition - because insect pest behavior is closely related to insect infestation in plants. The results showed that there were high variations on the number of damaged pods, the frequency of insect preference for probing, and the number of eggs at oviposition (Table 6). Cikuray as a check variety showed susceptible reaction to *Riptortus* spp. infestation, with the percentage of damaged pods reaching 31%, an average probing frequency of 0.5, and resistance score 8 (Appendix 2). This result is interesting because Cikuray was previously known to

have moderate tolerance to pod sucking pest, *Nezara viridula* (Kuswantoro et al. 2020). The PI-092734 variety showed a resistant reaction to pod sucking pests, with a percentage of damaged pods 0.25%.

Damage pod (Figure 5) is one of the variables that determines the level of soybean resistance to pod sucking insects as reported by Kuswantoro et al. (2020) and Da Fonseca Santos et al. (2018), where the lower the damage pods ratio, the more resistant the varieties to pod sucking bugs. In this study, several soybean accessions from East Java showed a low percentage of damage pods, including Lokal Madiun, Lokal Tulungagung, Lokal Pasuruan, and Lokal Blitar (Figure 4).

Variation in the percentage of damaged pods on 100 accessions tested was quite high and the highest number was reached by MLG-3017 (Figure 4). Damage pods ranged from 0 to 74.55%, with the average number of 17.64% (Appendix 2). This value is lower compared to those reported by Asadi (2009) of which the average damage reached 23-33%. The difference is probably due to differences in the measured damage components. Asadi (2009) measured the cumulative value of damage caused by several types of pod sucking pests (*Nezara* sp., *Piezodorus* sp., and *Riptortus* sp.), whereas in this study the damage was only caused by *Riptortus* spp.

Evaluation for pod-sucking bug resistance of 100 soybean accessions based on scoring value for damaged pods showed that 12 accessions were highly resistant, 12 accessions were resistant, and 13 accessions were moderately resistant (Figure 6). There were 39 susceptible accessions and 24 highly susceptible accessions.

Table 6. Descriptive statistics of	probing activity,	oviposition, and	percentage of	damaged pods.

Items	Probing activity (insect/accession)	Oviposition (egg/accession plant)	Damage pods/plant (%)
Mean	1.20	0.65	17.64
Standard error	0.08	0.10	1.60
Median	1.00	0.00	13.52
Mode	0.75	0.00	0.00
Standard deviation	0.83	1.05	16.12
Sample variance	0.686547515	1.106872452	259.7113113
Kurtosis	0.94	7.91	1.48
Skewness	0.99	2.51	1.26
Range	4.00	6.00	74.55
Minimum	0.00	0.00	0.00
Maximum	4.00	6.00	74.55
Sum	122.75	66.00	1799.39
Count	102	102	102

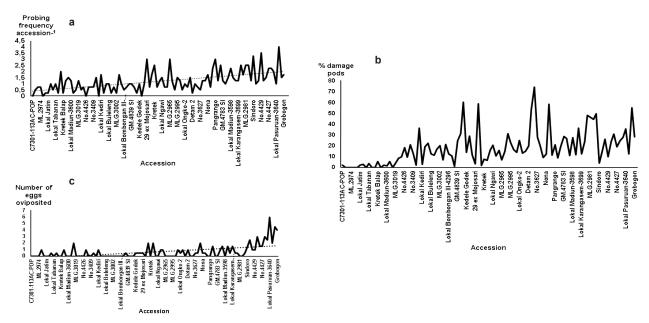


Fig. 4. Probing frequency (a), damage pods (b), and oviposition (c) of 100 soybean accessions infested with Riptortus spp.









Fig. 5. Damage pods of soybean caused by Riptortus spp. in the greenhouse assessment.

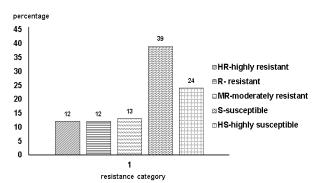


Fig. 6. The proportion of resistant level of 100 soybean accessions to Riptortus spp. based on scoring of damage pod percentage.

# Correlation Between Pod Sucking Bug Evaluation and Yield

Results showed that 100 grain weight, probing activity, oviposition and percentage of damage pods generally had unsignificant low correlation according to Pearson

correlation analysis with p value of 0.01 (Table 7 and 8). Only seed size (small, medium, and big) and 100 grain weight showed significantly high correlation, although negative (Table 7 and 8). The 100 grain weight and percentage of damage pods also showed a negative correlation, where the higher the 100 grain weight (which means the bigger the seed size) the lower number of pods attacked by insects (damage pods).

Based on ANOVA analysis, all variables tested (mean of probing, oviposition, damage pods, and genotype) contributed significantly to the resistance of soybean accessions (p-value = 1.5747E-80 and alpha 0.05). Both nymphs and imago of Riptortus spp. cause damage on soybeans through stylet inserted into leaves, stalks, flowers, pods and seeds (Chen et al. 2018). As a result, these plant parts lose fluids or nutrients, slowly wilt or turn yellow, dry up or fall off and fall. When the insect jabs and sucks the liquid and nutrients from the pods, the loss of yield will be very noticeable. Li et al.

Table 7. Correlation analysis of soybean accession resistance to Riptortus spp.

Variable	Genotype	Average of probing activity	Average of oviposition	Percentage of damage pods	Weight of 100 grains
Average of probing activity	0.131				
Average of oviposition	-0.056	0.181			
Percentage of damage pods	0.118	0.046	0.114		
Weight of 100 grains	-0.008	0.124	0.160	-0.055	
Seed size	0.063	-0.073	-0.196	0.036	-0.896

Table 8. Pairwise Pearson correlation of soybean accession resistance to Riptortus spp.

Variable 1	Variable 2	Correlation	95% CI for ρ	P-Value
Average of probing activity	Genotype	0.131	(-0.065; 0.318)	0.188
Average of oviposition number	Genotype	-0.056	(-0.247; 0.140)	0.578
Percentage of damage pods	Genotype	0.118	(-0.079; 0.305)	0.239
Weight of 100 grains (g)	Genotype	-0.008	(-0.203; 0.186)	0.933
Seed size	Genotype	0.063	(-0.133; 0.255)	0.528
Average of oviposition number	Average of probing activity	0.181	(-0.014; 0.363)	0.068
Percentage of damage pods	Average of probing activity	0.046	(-0.150; 0.238)	0.647
Weight of 100 grains (g)	Average of probing activity	0.124	(-0.072; 0.311)	0.214
Seed size	Average of probing activity	-0.073	(-0.264; 0.123)	0.464
Percentage of damage pods	Average of oviposition number	0.114	(-0.083; 0.301)	0.256
Weight of 100 grains (g)	Average of oviposition number	0.16	(-0.036; 0.343)	0.109
Seed size	Average of oviposition number	-0.196	(-0.376; -0.002)	0.048
Weight of 100 grains (g)	Percentage of damage pods	-0.055	(-0.247; 0.141)	0.583
Seed size	Percentage of damage pods	0.036	(-0.159; 0.229)	0.717
Seed size	Weight of 100 grains (g)	-0.896	(-0.929; -0.849)	0

(2021) reported that R. pedestris infestation in soybean caused seed development stagnation, interference in signalling for senescene and leaf development regulation, interference in photosynthetic transport signal, stay-green leaves, and shriveled seeds. In addition, several Riptortus species are also vectors of certain fungal diseases (Kimura et al. 2008) which cause the problem to become more complex.

# Grouping and Selecting the Resistant Candidate

Grouping of resistant level of 100 soybean accessions tested based on cumulative value (N cum) of scoring of damage pods, probing activity and oviposition of *Riptortus* sp. has successfully selected 12 resistant accessions, i.e. C7301-113AC-POP, Lokal Madiun-3549, Lokal Klungkung, ML.2974, Singgalang, Lokal Jepara, Lokal Jatim, Lokal Trenggalek, Lokal Tabanan, Lokal Blitar, and Lokal Kuningan 10 (Table 9). These accessions were more resistant than the popular released variety such as Wilis, Grobogan, Detam 2, and Gepak Ijo. Kuswantoro et al. (2020) reported that Grobogan is

one of the varieties that responds susceptible to pod sucking pest *N. viridula* (stink bug), where 70% of the pods become empty due to the infestation of these sucking insects. Thus results of the study add valuable information that Grobogan is not only susceptible to *N. viridula*, but also to *Riptortus* spp.

Classification of resistant levels based on cumulative scoring value also obtained 28 accessions that are moderately resistant, 58 accessions susceptible, and 2 accessions highly susceptible to *Riptortus* spp. (Table 9). There is interesting point which, of the 12 resistant accessions selected, six of them were local soybeans originating from East Java. This implicity shows that East Java has the potential as the source of local soybean varieties which are resistant to pod sucking bugs.

Another interesting point is grouping based on cumulative scoring turned out to produce a list of accessions with different resistance levels compared to grouping based on scoring of the percentage of damaged pods. Based on scoring value of the percentage of damaged pods, we obtained large number of resistant accessions and highly susceptible accessions (12 highly resistant, 12 resistant, 13 moderately resistant,

Table 9. Grouping of resistant level of 100 soybean accessions based on cumulative scoring value (N cum).

Name of accessions	Resistant category	Total number of acessions
C7301-113AC-POP, Lokal Madiun-3549, Lokal Klungkung, ML.2974, Singgalang, Lokal Jepara, Lokal Jatim, Lokal Trenggalek, Lokal Tulungagung, Lokal Tabanan, Lokal Blitar, Lokal Kuningan 10	Resistant	12
Kretek Balap, No.29 Ex Bogor, Lokal Magetan, Lokal Madiun-3600, Lokal Sumenep, Lokal Pasuruan-3665, MLG.3019, M.3289, GM.378 SI, No.4226, MLG2627, Kepet Godek, No.3409, Lokal Jombang, No.3596, Lokal Kediri, MLG.2759, Lokal Banyuwangi, Lokal Buleleng, Lokal Madiun-3732, M.2996, MLG.3002, ML.3024, Lokal Bima Hijau, Lokal Bombongan III-429, Lokal Bombongan II, GM.374 SI, GM.4839 SI	Moderately Resistant	28
MLG.2984, Kepet Hitam, Kedele Godek, Kedele Hitam, GM.920 SI, 29 ex Mojosari, Lokal Nganjuk, Lokal Pasuruan-3641, Kretek, No.3654, Lokal Pasuruan-3662, Lokal Ngawi, Lokal Bojonegoro-3737, Otok, MLG.2965, No.3762, MLG.2995, MLG.3025, M.3031, Lokal Ongko-2, Rajabasa, Ijen, Detam 2, Gepak Ijo, MLG.3017, No.3627, Lokal Jember, Lokal Lumajang, Nona, Si Nyonya, Lokal Pangrango, Lokal Bombongan III-4295, GM.4779 SI, GM.4783 SI, GM.363 SI, Hitam, Lokal Madiun-3598, Lokal Bangkalan, Lokal Pasuruan-3666, Lokal Karangasem-3699, Lokal Karangasem-3701, Lokal Bojonegoro-3736, MLG.2981, No.3764, ML.3028, LB-80, Sindoro, GM.4596 SI, Lokal Cikapak, No.4429, Wilis, Kedelai Langkat, No.4227, Keee Susu, Lokal Pasuruan-3612, Lokal Pasuruan-3640, Lokal Badung	Susceptible	58
Lokal Karangasem-3700, Grobogan	Highly susceptible	2

39 susceptible and 24 accessions were very susceptible, respectively). The difference in scoring determination is the main cause of the result's difference. However, based on our opinion, scoring using N-cumulative provides more comprehensive assessments so that it will be more stringent in selecting resistant accessions. The implication is, we confidently suggested that this scoring method will provide more valid results.

# Plant Type, Seed Size, Seed Weight and Resistance

Based on the observation results, 76 accessions had determinate plant growth types, 24 accessions were semi-determinate, and only one accession was indeterminate (Figure 7 and Appendix 1). The control varieties, Cikuray and PI-092734, both had determinate growth types. Mostly soybean accessions in this study were determinate type which showed a resistant response to insect infestations (Figure 8b). It was suggested that plant type might play important role related to resistance against pod sucking pests.

The types of growth of soybean related to their biomass accumulation. The determinate type was characterized by: size of the tip of the plant stem is almost as large as the middle stem; the flowering takes place simultaneously; vegetative growth stops after the plant flowers; the plant height is short to medium; and the upper leaves have the same size as the middle leaves (Rukmana and Yuniarsih 1996). For indeterminate type, size of the tip of the plant is smaller than the middle stem; the length of

the stem is long and twisted; the flowering takes place gradually from the base to the upper stem; the plant has a continuous vegetative growth after flowering; plant height is moderate to high; and the upper leaves are smaller than the middle leaves (Rukmana and Yuniarsih, 1996). Meanwhile, the semi-determinate type has the characteristics between both determinate and indeterminate. Most of cultivated soybean in Indonesia are classified into determinate and semi-determinate types.

So far there have been no reports on the correlation between plant types and their resistance to pod sucking insects in soybean. Mostly resistant and susceptible accessions in this study were obtained from determinate soybeans. It seems that the determinate type is not the underlying factor in plant resistance to *Riptortus* spp. In this study, only one accession had indeterminate type, and it was found as resistant against *Riptortus* spp. This is quite interesting, because it might possible that the indeterminate-type character is one of the factors of soybean resistance to this pest. Unfortunately, the limitation of the indeterminate type in this study cannot confirm this suggestion.

The results of seed observations showed that 25 accessions have big seed size, 21 accessions have medium size, and 54 accessions have small seed size (Figure 7). Further analysis on resistant accessions showed that 8 accessions have small seed size, 3 accessions have big seed size, and one accession have medium seed size (Figure 8a). Bae et al. (2014) reported that seed size is closely related to the nutritional

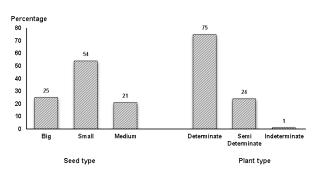


Fig. 7. Seed size and plant type of 100 soybean accessions used in this study.

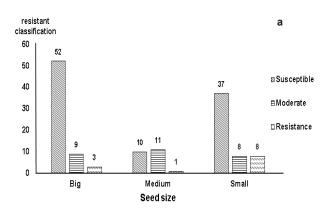
content. Logically, if seed size is closely related to nutritional content, then the smaller beans will have the lower nutrition. Thus the nutritional needs of insects will be more fulfilled by soybeans with big seed size. That is, it should be the big size accessions will be preferred by insect pests. In other words, accessions with big seed size

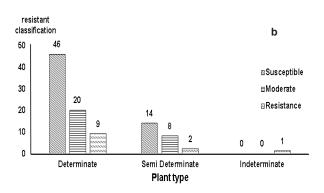
will be more susceptible than those having small seed size. Using this understanding, the results of this study are relevant where most of the susceptible accessions were obtained from soybeans with big seed size, reached 52 accessions respectively (Figure 8a).

Resistant genotypes of soybean obtained in this study mostly had 100 seed weight of 4.5 - 6 g (Figure 8c). Those results are different from that reported by Takashi et al. (2006) that pods with smaller seeds are susceptible to pod suckers, so usually in the tropics there will be many pod sucking pests in the small seed genotypes (Takashi et al. 2006). This difference in results indicates that seed size might not the only character determining the soybean resistance to pod sucker insects.

Seed size and shape are important traits for determining yield and quality of soybean (Hina et al. 2020). Several reports stated that the ability of soybeans to compensate for damage is included in its resistance in the field to soy-sucking pests, including seed size. However their contribution or correlation to plant resistance against *Riptortus* spp. remains an interesting and open area of inquiry.

To determine how the values of N1, N2 and N3 contribute in selecting resistant accessions, we noted that for the resistant category, the N1 value had the highest proportion compared to N2 and N3. However, in moderately resistant and susceptible categories, the N3 value had the largest proportion (Figure 9). N1 is a scoring based on insect probing activity, while N3 is determined from pod damage. So it is reasonable that both N1 and N3 determine the most to the resistance or susceptibility of the accessions. As for N2, which is scored based on oviposition activity, the proportion





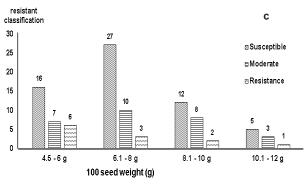


Fig. 8. Resistant level of soybean accessions based on seed size (a), plant type (b) and seed weight (c).

tends to be lower. This might be because, during the insects infestations in the screenhouse, males and females insects were mixed, thus the existence of males becomes the correcting factor (males do not ovipose).

Screening for germplasm accessions to obtain resistant candidates is admittedly a very inefficient activity in terms of time, requires a lot of effort and money. Moreover, by limiting the observation variables which usually should involve several yield components, the duration of the research must be carried out throughout the life of the plant and even continued after harvest. With the large number of accession collections, which might reach thousands

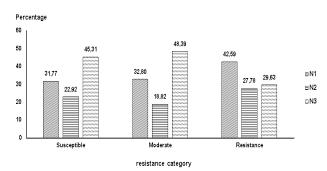


Fig. 9 . Proportion of N1, N2 and N3 to determine resistant level of soybean accessions to *Riptortus spp*.

number in Gene Bank, at the same point, this activity will become difficult to maintain. Related to this, specifically to address the problem of pests from pod sucking insect (Hemiptera) in soybean, there is actually another suggested approach which focusing on searching for tolerant genotypes.

Plant with tolerance mechanisms has very different defense systems from resistance mechanisms. When we use resistant approached then the focus will be related on antixenosis and antibiosis; but if we use tolerant approached then we will pay attention more on the ability of the plant to recover (Li et al. 2021) and accelerate its growth to anticipate damage caused by insect infestations. Or at least, tolerant plants are those that are less susceptible even though they are attacked by high density pests. Based on this, the screening carried out will be more fruitful, because the standards set are below the resistant plant standard.

Related with the above thought, the evaluation of 100 soybean accessions to *Riptortus* spp. in the screen house basically attempted to carry out a more practical but accurate screening method (by releasing a large number of pod sucking insects in the screenhouse as the selection pressure), and included several insect behaviors (probing and oviposition activities) as new approaches in determining plant resistance. In addition, the scoring system used in this study is also novel, which we integrated the insect behavior as weighting into the cumulative scoring determination. In fact, this approach is very relevant according to the integrated pest management (IPM) which was popular as the best control system for pests (Marwoto 2006).

As it has been well known, IPM strongly places monitoring of pest populations as consideration to carry out the control actions. By knowing the actual number of pests infested in crop, control actions will be determined in accordance with the control threshold (usually related to economic losses) that

has been determined. IPM is currently the most applicable, effective, and safe control strategy for the environment which broadly adapted by farmers in Indonesia (Marwoto 2006). Unfortunately, so far there have been no studies to integrate data on insect pest populations as a variable to measure or estimate plant resistance to these insect pests. Therefore, the scoring method in this study is new effort to develop new scoring system which adopted the monitoring of pest population (based on insect's probing and oviposition observations) as variable for measure plant resistance against pest.

It appears that in our proposed method, the selection pressure of insect pests in the treatment with insect infestation was able to select resistant accessions from those are susceptible. However, according to these results, the number of infested insects needs to be increased to obtain higher selection pressure, including a minimum density of 500 imago per unit treatment. In addition, the scoring method using N-cumulative (which incorporate N1, N2 and N3 as its component score) is also able to provide better selection results. Although this method still requires further validation and improvement, it hopefully will add contribution to support research on plant tolerance mechanism to pod sucking insects, *Riptortus* spp. in the future.

## CONCLUSION

Twelve soybean accessions showed resistance to pod sucker Riptortus spp., namely C.730-1113-4-C-O, Lokal Jatim, Lokal Madiun, Lokal Trenggalek, Lokal Tulung Agung, Lokal Klungkung, Lokal Tabanan, Lokal Blitar, MLG-2974, MLG.3019, Lokal Kuningan 10, and Lokal Jepara. These accessions were more resistant than the popular released variety such as Wilis, Grobogan, Detam 2, and Gepak Ijo. Small seed size was not a major determinant of soybean resistant to pod suckers. Indeterminate plant types still require further validation as to whether they contribute significantly to plant resistance against pod sucking insects. The selection pressure of insect pests in the treatment with insect infestation was able to select resistant accessions from those are susceptible. The scoring method using N-cumulative (which incorporate N1, N2 and N3 as its component score) was also able to provide better selection results. Crop damage was indirectly influenced by the high frequency of probing and oviposition, although its relation to plant tolerant mechanisms still needs further investigation.

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Appendix 1. Weight of 100 grains, Seed size, growth type and resistance level of 100 soybean accessions in this study.

Name of accession	Weight of 100 seeds	Seed size	Growth type		sification of resistance (Code - category)
MLG2627	11	big	Determinate	R	Resistant
C7301-113AC-POP	4.7	small	Indeterminate	R	Resistant
MLG2984	6.4	small	Determinate	R	Resistant
Kepet Hitam	6	small	Determinate	R	Resistant
Kepet Godek	4.7	small	Determinate	R	Resistant
Kedele Godek	4.6	small	Determinate	R	Resistant
Kedele Susu	9	big	Semi determinate	R	Resistant
No.3409	4.7	small	Determinate	R	Resistant
Wilis	9.4	big	Determinate	R	Resistant
Kedele Hitam	7.6	medium	Determinate	R	Resistant
Lokal Jatim	4.7	small	Semi determinate	R	Resistant
Hitam	6.8	small	Determinate	R	Resistant
Kretek Balap	7.7	medium	Determinate	MR	Moderately resistant
Lokal Madiun-3549	8.6	medium	Determinate	MR	Moderately resistant
GM.920 SI	8.5	medium	Determinate	MR	Moderately resistant
No.29 Ex Bogor	6	small	Determinate	MR	Moderately resistant
29 ex Mojosari	6	small	Determinate	MR	Moderately resistant
Lokal Jombang	8	medium	Determinate	MR	Moderately resistant
Lokal Nganjuk	7.5	medium	Determinate	MR	Moderately resistant
Lokal Magetan	8	medium	Determinate	MR	Moderately resistant
No.3596	12	big	Determinate	MR	Moderately resistant
Lokal Madiun-3598	5.3	small	Semi determinate	MR	Moderately resistant
Lokal Madiun-3600	5	small	Determinate	MR	Moderately resistant
Lokal Trenggalek	5	small	Semi determinate	MR	Moderately resistant
Lokal Tulungangung	8	medium	Determinate	MR	Moderately resistant
Lokal Kediri	8	medium	Semi determinate	MR	Moderately resistant
Lokal Pasuruan-3612	8	medium	Determinate	MR	Moderately resistant
Lokal Sumenep	6.9	small	Determinate	MR	Moderately resistant
Lokal Bangkalan	11	big	Determinate	MR	Moderately resistant
No.3627	11	big	Determinate	MR	Moderately resistant
MLG.2759	4.5	small	Determinate	MR	Moderately resistant
Lokal Pasuruan-3640	9	big	Semi determinate	MR	Moderately resistant
Lokal Pasuruan-3641	10	big	Semi determinate	MR	Moderately resistant
Lokal Banyuwangi	9	big	Semi determinate	MR	Moderately resistant
Lokal Jember	8	medium	Semi determinate	MR	Moderately resistant
Kretek	9	big	Determinate	MR	Moderately resistant
No.3654	4.7	small	Determinate	MR	Moderately resistant
Lokal Lumajang	9	big	Determinate	MR	Moderately resistant
Lokal Pasuruan-3662	9	big	Semi determinate	MR	Moderately resistant
Lokal Pasuruan-3665	7.5	medium	Determinate	MR	Moderately resistant
Lokal Pasuruan-3666	5.4	small	Determinate	S	Susceptible
Lokal Klungkung	12	big	Determinate	S	Susceptible
Lokal Tabanan	12	big	Determinate	S	Susceptible
Lokal Badung	7	small	Determinate	S	Susceptible
Lokal Karangasem-3699	8	medium	Semi determinate	S	Susceptible
Lokal Karangasem-3700	8	medium	Semi determinate	S	Susceptible
Lokal Karangasem-3701	8.2	medium	Determinate	S	Susceptible
Lokal Buleleng	9.3	big	Determinate	S	Susceptible
Lokal Blitar	6.8	small	Determinate	S	Susceptible
Lokal Madiun-3732	5.5	small	Semi determinate	S	Susceptible
Lokai iviaululi-3/32	٥.٥	Sinali	seim determinate	3	Susceptible

Appendix 1. (continued).

Name of accession	Weight of 100 seeds	Seed size	size Growth type		sification of resistance (Code - category)
Lokal Ngawi	4.7	small	Semi determinate	S	Susceptible
Lokal Bojonegoro-3736	5.7	small	Semi determinate	S	Susceptible
okal Bojonegoro-3737	6.3	small	Semi determinate	S	Susceptible
Iona	6.7	small	Semi determinate	S	Susceptible
tok	7.2	small	Determinate	S	Susceptible
i Nyonya	8.5	medium	Determinate	S	Susceptible
ILG.2965	5	small	Semi Determinate	S	Susceptible
IL.2974	6.1	small	Determinate	S	Susceptible
0.3762	6	small	Determinate	S	Susceptible
ILG.2981	9	big	Determinate	S	Susceptible
o. 3764	6.4	small	Determinate	S	Susceptible
LG.2995	6.4	small	Determinate	S	Susceptible
1.2996	7	small	Semi determinate	S	Susceptible
okal	11.5	big	Determinate	S	Susceptible
ILG.3002	6	small	Determinate	S	Susceptible
ILG.3019	7.1	small	Determinate	S	Susceptible
IL.3024	8.8	medium	Determinate	S	=
				S	Susceptible
ILG.3025	8	medium	Semi determinate		Susceptible
IL.3028	9	big	Semi determinate	S	Susceptible
1.3031	6.1	small	Semi determinate	S	Susceptible
1.3289	6.2	small	Determinate	S	Susceptible
B-80	6.1	small	Determinate	S	Susceptible
angrango	7	small	Determinate	S	Susceptible
okal Ongko-2	8.2	medium	Determinate	S	Susceptible
okal Bima Hijau	5	small	Determinate	S	Susceptible
edelai Langkat	7.3	small	Determinate	S	Susceptible
inggalang	9.8	big	Semi determinate	S	Susceptible
okal Bombongan III-4295	6.8	small	Semi determinate	S	Susceptible
okal Bombongan III-4296	4.8	small	Determinate	S	Susceptible
okal Bombongan II	4.7	small	Determinate	S	Susceptible
indoro	7	small	Determinate	S	Susceptible
M.374 SI	8	medium	Determinate	S	Susceptible
M.378 SI	9	big	Determinate	S	Susceptible
M.4779 SI	7	small	Determinate	S	Susceptible
M.4783 SI	10	big	Determinate	S	Susceptible
M.4596 SI	8	medium	Determinate	S	Susceptible
M.4839 SI	4.7	small	Determinate	S	Susceptible
M.363 SI	7	small	Determinate	S	Susceptible
okal Kuningan 10	7	small	Determinate	S	Susceptible
okal Cikapak	5	small	Determinate	S	Susceptible
ajabasa	5	small	Determinate	S	Susceptible
okal Jepara	9.8	big	Determinate	S	Susceptible
en	9	big	Determinate	S	Susceptible
o.4426	4.7	small	Determinate	S	Susceptible
0.4427	4.6	small	Determinate	S	Susceptible
0.4429	11.5	big	Determinate	S	Susceptible
etam 2	11	big	Determinate	S	Susceptible
epak Ijo	4.5	small	Determinate	S	Susceptible
ILG.3017	7	small	Determinate	HS	Highly susceptible
robogon	8	medium	Determinate	HS	Highly susceptible

Appendix 2. Probing activities, number of oviposition, percentage damage pods, and scoring cumulative (Ncum) to determine resistance level of 100 soybean accessions.

Accession name C7301-113AC-POP	Probing frequency	Number of eggs oviposited	% Damage pods	N cum	Resistancy category	
					R	Resistant
Lokal Madiun-3549	0.5	0	0	4	R	Resistant
Lokal Klungkung	0.75	0	0	4	R	Resistant
ML.2974	0.75	0	0	4	R	Resistant
Singgalang	0	1	0	4	R	Resistant
Lokal Jepara	0.25	0	0	4	R	Resistant
Lokal Jatim	0.25	0	2.33	5	R	Resistant
Lokal Trenggalek	1	0	3.07	5	R	Resistant
Lokal Tulungangung	0.5	0.5	0	5	R	Resistant
Lokal Tabanan	1	0	3.76	5	R	Resistant
Lokal Blitar	0.25	0.5	0	5	R	Resistant
Lokal Kuningan 10	2	0	0	5	R	Resistant
Kretek Balap	0.25	0	5.62	6	MR	Moderately resistant
No.29 Ex Bogor	1.25	1	0	6	MR	Moderately resistant
Lokal Magetan	1.5	0	2.27	6	MR	Moderately resistant
Lokal Madiun-3600	1.25	0	1.42	6	MR	Moderately resistant
Lokal Sumenep	0.25	0	5.59	6	MR	Moderately resistant
Lokal Pasuruan-3665	0.5	2	0	6	MR	Moderately resistant
MLG.3019	1.25	0	4.6	6	MR	Moderately resistant
M.3289	0.5	0	8.71	6	MR	Moderately resistant
GM.378 SI	0.75	0	10	6	MR	Moderately resistant
No.4426	0	0	18.31	6	MR	Moderately resistant
MLG2627	0.75	1	9.63	7	MR	Moderately resistant
Kepet Godek	0.75	0	21.13	7	MR	Moderately resistant
No.3409	0.75	0	12.16	7	MR	Moderately resistant
Lokal Jombang	1.5	0.5	2.17	7	MR	Moderately resistant
No.3596	0	0	36.67	7	MR	Moderately resistant
Lokal Kediri	1.75	1	2.88	7	MR	Moderately resistant
MLG.2759	0.25	0	19.15	7	MR	Moderately resistant
Lokal Banyuwangi	1	0	22.46	7	MR	Moderately resistant
Lokal Buleleng	1	0	12.44	7	MR	Moderately resistant
Lokal Madiun-3732	0.25	0	11.62	7	MR	Moderately resistant
M.2996	0.75	0	17.17	7	MR	Moderately resistant
MLG.3002	0.25	0	20.31	7	MR	Moderately resistant
ML.3024	1.75	0	7.1	7	MR	Moderately resistant
Lokal Bima Hijau	1.73	0	21.26	7	MR	Moderately resistant
Lokal Bombongan III-4296	0.5	0	12.64	7	MR	Moderately resistant
Lokal Bombongan II	0.75	0	10.59	7	MR	Moderately resistant
GM.374 SI	1	0.5	1.08	7	MR	Moderately resistant
GM.4839 SI	1	0.5	24.82	7	MR	Moderately resistant
MLG2984	0.75	0	31.35	8	S	Susceptible
	0.73	0	60.62	8	S	Susceptible
Kepet Hitam						=
Kedele Godek Kedele Hitam	1	0.5 0.5	13.89	8	S S	Susceptible Susceptible
	0		29.21	8		•
GM.920 SI	1	0.5	12.03	8	S	Susceptible
29 ex Mojosari	3	0.5	3.3	8	S	Susceptible
Lokal Nganjuk	0.75	0	59.19	8	S	Susceptible
Lokal Pasuruan-3641	1.75	2	1.72	8	S	Susceptible
Kretek	2.5	0	7.72	8	S	Susceptible
No.3654	0.75	2	6.67	8	S	Susceptible
Lokal Pasuruan-3662	1.25	0	16.21	8	S	Susceptible

Appendix 2. (continued).

Accession name	Probing frequency	Number of eggs oviposited	% Damage pods 20.72	N cum	Resistancy category	
Lokal Ngawi	1.5				S	Susceptible
Lokal Bojonegoro-3737	0.75	1	10.74	8	S	Susceptible
Otok	0.75	1	15.9	8	S	Susceptible
MLG.2965	3	0	7.17	8	S	Susceptible
N0.3762	1.25	0	14.73	8	S	Susceptible
To.3764	0.5	0	31.35	8	S	Susceptible
ILG.2995	1.5	0	23.34	8	S	Susceptible
ILG.3025	1.25	0	16.74	8	S	Susceptible
1.3031	0.5	1	14.22	8	S	Susceptible
okal Ongko-2	1	0.5	25	8	S	Susceptible
ajabasa	0.75	1	13.15	8	S	Susceptible
en	1.5	0	15.35	8	S	Susceptible
etam 2	0.25	1	20.1	8	S	Susceptible
epak Ijo	1	0	53.79	8	S	Susceptible
LG.3017	0.75	0	74.55	8	S	Susceptible
0.3627	0.5	0.5	28.07	9	S	Susceptible
okal Jember	1.25	0.5	20.48	9	S	Susceptible
okal Lumajang	1.25	2	9.38	9	S	Susceptible
ona	1.75	0.5	12.5	9	S	Susceptible
Nyonya	0.75	0.5	58.73	9	S	Susceptible
kal	2.25	0.5	11.58	9	S	Susceptible
ngrango	3	0	21.02	9	S	Susceptible
kal Bombongan III-4295	1.25	1.5	8.78	9	S	Susceptible
1.4779 SI	2.5	0	6.76 19.94	9	S	Susceptible
1.47/9 SI 1.4783 SI	1.25	0	28.74	9	S	=
1.4783 SI 1.363 SI	1.23	1	26.93	9	S	Susceptible
am	1.5	1.5	11.08	10	S	Susceptible
				10		Susceptible
kal Madiun-3598	2.25	0	27.57		S	Susceptible
kal Bangkalan	1.5	1.5	11.95	10	S	Susceptible
kal Pasuruan-3666	1.25	0.5	36.49	10	S	Susceptible
kal Karangasem-3699	1.25	1.5	12.5	10	S	Susceptible
kal Karangasem-3701	2.5	0.5	23.84	10	S	Susceptible
kal Bojonegoro-3736	1.75	0.5	48.53	10	S	Susceptible
LG.2981	2.5	0	46.62	10	S	Susceptible
L.3028	2.5	0	44.84	10	S	Susceptible
3-80	1.25	0.5	50	10	S	Susceptible
ndoro	3.25	1.5	4.17	10	S	Susceptible
M.4596 SI	1	2.5	15.71	10	S	Susceptible
kal Cikapak	1.25	1	26.24	10	S	Susceptible
0.4429	3.5	1	9.9	10	S	Susceptible
ilis	1.25	3	24.01	11	S	Susceptible
edelai Langkat	1.5	1.5	30.85	11	S	Susceptible
5.4427	2.25	1.5	19.2	11	S	Susceptible
edele Susu	2.25	3	24.83	12	S	Susceptible
okal Pasuruan-3612	2	2.5	28.07	12	S	Susceptible
okal Pasuruan-3640	1	6	35.65	12	S	Susceptible
okal Badung	4	2	12.59	12	S	Susceptible
okal Karangasem-3700	1.5	4.5	55.26	13	HS	Highly susceptibl
robogon	1.75	4	28.33	13	HS	Highly susceptibl