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COMBINED EFFECT OF CORN IN THE BARRIER CROP AND PLANT EXTRACTS AGAINST *Cowpea mild mottle virus* INFECTING SOYBEAN [*Glycine max* (L.) Merr.] IN THE FIELD

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Abstract

Cowpea mild mottle virus (CpMMV) is one of the problems that can decrease soybean production. The research was conducted on the combined effects of corn in the barrier crop with plant extracts against CpMMV infecting soybean in the field. The field data was conducted using a Completely Randomized Design. The mean of disease incidence and disease severity is measured from total plants in each replicate plot on each treatment. Planting one or two of corn lines were grown at the edge four weeks before planting soybeans. Cashew nut shell (CNS), pagoda leaf, and rhizome of ginger extracts were applied using the sprayer and applied at 24 h before virus acquisition and transmission by whiteflies. The result showed that the virus incubation period ranged from 9−38 days after transmission longer than the untreated control. Planting two corn lines at the edge with CNS extract as bioactivator on soybean was the most extended incubation period of the virus and the lowest absorbance value DAS-ELISA of 0.20. There was a 73.11 % increase in the relative inhibition level of the virus. Planting corn at the edge with CNS extract proved to be more effective than soybean monoculture with CNS extract. However, soybean monoculture with CNS extract provides a better relative inhibition level of the virus (64.32 %) than planting two rows of corn on the edge combined with ginger of rhizome extract and planting two rows of corn on the edge with pagoda leaf extract as bioactivator on a soybean plant.

Keywords: *Bemisia tabaci.* Bioactivators. Tolerance Inductor. Windbreaks.

1. Introduction

The Soybean [*Glycine max* (L.) Merr.] is the essential agriculture commodity as the major source of protein. *Cowpea mild mottle virus* (CpMMV) is one of the problems that can decrease soybean production. Whiteflies (*Bemisia tabaci*) are known as vectors of CpMMV and spread from plant to plant by wind. They transmitted CpMMV manner, causing severe (CpMMV-S) and mild (CpMMV-M) disease symptoms (Andayanie et al. 2019). The virus can be transmitted either by seed or mechanical inoculation. The viruliferous population of whitefly in the tropical areas turned out to be the outbreak of CpMMV. The CpMMV is widely distributed in Indonesia and cause a variety of symptoms on soybean such as mild chlorotic blotches, green mosaic, distortion, blistering, and stunting.

The number of resistant varieties and virus-free seeds is not sufficient for farmers' needs. Control strategies are used to anticipate more significant losses due to whitefly infestation. Management of CpMMV usually controls its vector using insecticide, which harms the environment, non-target insects, and

phytotoxicity. Bioactivators of ginger rhizome, cashew nut shell, and neem leaf extracts helped decrease the disease incidence and severity, symptoms, and virus concentration, respectively in soybean plants. Moreover, the whitefly nymphs of different stages (first to the third instar) could be suppressed by cashew nut shell extracts (Andayanie and Ermawati 2019).

Barrier crops are used to windbreaks around the main crop and considered decreasing the ability of virus infections transmitted non-persistent because the total number of viruliferous aphids landing on the protected crop is significantly reduced (Boiteau et al. 2009; Hooks and Fereres 2006). Consequently, barrier crop may be potential alternatives aphid landing before taking off to host for the virus and the vector. Insect vectors are diverted to eat the main crops and control several viruses such as the *Potato leaf roll virus* (PLRV), *Potato virus Y* (PVY), and *Bean yellow mosaic virus* (BYMV) (Jones 2005; Gobiye et al. 2016). Barrier crop as corn (*Zea mays* L.) was also effective against *Pepper veinal mottle virus* (PVMV) by planting of corn on the edge, and intercrops (Kapoor 2012). Barrier crops should decrease the ability of nonpersistently aphid to transmit viruses to the primary crop. However, the corn potential for the successful use of barrier plants as vector insect management tactics still receives less attention than other management strategies. This is attributable to the fact that commercial soybean farmers commonly used insecticides such as Imidacloprid. El-Sawy et al. (2017) and Aderiye et al. (2015) noted that plant extract's bioactive compound reduces inoculum resources of viral diseases. The bioactivator could play a key role in systemic resistance against plant viruses. This was also supported by Andayanie et al. (2019) who developed *Anacardium occidentale* Linn, *Azadirachta indica* A. Juss. and *Zingiber officinale* Rosc. as a source of tolerance inductor for CpMMV infection on a soybean plant. However, the use of tolerance inductor to CpMMV is still limited to a screen house experiment. Therefore, this study's objectives were to reduce the ability of whitefly-transmitted diseases and suppress CpMMV infection that leads to sustainable and environmentally friendly by barrier crop as windbreaks and plant extract as bioactivator on soybean in the fields.

2. Material and Methods

The experiment was conducted at Merdeka Madiun University, Indonesia - Department of Agrotechnology, Plant Protection Laboratory. The field experiment was initiated in March 2020 on alluvial soil at the regional Research Center (21°29'S and longitude 31°50'E) in the village Jenggrik, District Kedunggalar, Ngawi, Indonesia.

Propagation of *Cowpea mild mottle virus*

The CpMMV isolates were obtained from a collection of the Plant Protection Laboratory. The inoculum was propagated in the soybean Wilis variety. The leaf surface of soybeans seven days after planting was dusted with carborundum and smeared. The CpMMV isolate was inoculated by smeared and maintained in the greenhouse at 29 °C, with 12 h dark, 12 h light period as a source of inoculum. Leaf mosaic and rugose mosaic symptoms appeared during the eight days after inoculation and were used as an inoculum source.

Preparation of plant extracts

Shells were collected from waste product of cashew nut (*Anacardium occidentale* L.) processing in Wonogiri-Indonesia. Pagoda leaf (*Clerodendrum japonicum* Thunb.) and rhizome ginger (*Zingiber officinale* Ros.) were harvested at 90 days after planting (DAP). The different parts of plants include cashew nutshell, pagoda leaf and rhizome ginger washed in tap water and rinsed with distilled water for drying. The dried samples of cashew nutshell were ground to fine powdered form. Extraction of powdered cashew nutshell was carried out by maceration with n-Hexane in a ratio of 1:10 (w/v). The extract was filtered and evaporated with a rotary vacuum evaporator at a temperature of 55−60 ^{o}C at low pressure (450−500 mm Hg). Filter was re-macerated with chloroform solvent and re-evaporated under low pressure. The viscous extract was mixed with distilled water in a ratio of 0.3:10 and added 0.5 mg Tween 80 with stirring (Andayanie et al 2018; Andayanie et al 2019). The extractions of pagoda leaf (*Clerodendrum japonicum* Thunb.) and ginger (*Zingiber officinale* Ros.) rhizome were carried out by maceration with distilled water in a ratio of 1:10 (w/v). Cashew

nutshell, pagoda leaf, and rhizome of ginger are used as bioactivators against CpMMV in the soybean plant. The plant extract was applied using the sprayer and applied at 24 h before virus acquisition and transmission by *Bemisia tabaci.*

Preparation of viruliferous aphids and transmission of CpMMV

Bemisia tabaci from the field was released from taro leaves overnight in the cage net. After that, a large colony of the first instar was kept on healthy soybean plants until a winged population appears. The *Bemisia tabaci* of second instar nymphs were removed to soybean plants infected with CpMMV. The plants were covered with the cage net. Transmission of the virus is carried out by releasing *Bemisia tabaci* adults that have eaten the acquisition of soybean plants infected with CpMMV at four corner points of the experimental field.

Observation of variables

The observational variables were observed as follows:

1. Incubation periods and characteristics symptom. The incubation period was calculated from the first symptoms of soybean-based on visual observation due to the treatment.
2. Disease incidence carried out since the first time the symptoms appeared to the sixth week at intervals once a week. The disease incidence is measured from the total main plants in each replicate plot on each treatment. It was calculated using the formula:

$$DI = \frac{a}{N} \times 100\%$$

Where,
DI : disease incidence (%)
a : numbers of the plant with CpMMV symptoms per plot
N : numbers of the plant observed per plot

3. Disease severity is measured from total main plants in each replicate plot on each treatment. It was calculated from the first week to the sixth week after CpMMV infected and based on a scale of 0-5 as described early by Andayanie et al. (2019); 1 = plants healthy without visible symptoms on all leaves; 2 = mild chlorotic blotches symptoms (10 to 30 % of the leaves infected) ; 3 = moderate symptom (green mosaic and distortion) (30 to 50% of leaves infected); 4 = prominent symptom with blistering, and stunting (51 to 70% of leaves infected); 5 = highly severe symptoms with stunting (> 71 %). Symptom score values were converted to disease severity scores. The percentage of disease severity was calculated by the following formula:

$$DS = \frac{\Sigma (n \times v)}{N \times V} \times 100\%$$

Where,
DS: the disease severity (%)
n : the sum of infected leaves in each category
v : value score of each category
N : total number of observed leaves per plant
V : the highest category

4. Disease severity data were used to calculate the area under the disease progress curve (AUDPC) as described by Campbell and Madden (1990). The AUDPC value was calculating according to the formula by Asare-Bediako et al. (2018).

$$AUDPC = \sum_{i=1}^{n} \frac{[y_1 + y_{i+1}]}{2} (t_{1+1} - t_i)$$

Where,

AUDPC : Area under disease progress curve
Y_i : an assessment of disease (%) at the i th observation
t_i : time (in weeks) at the *i* th observation
n : the total number of observation

The percentage relative inhibition level (RIL) of disease for the test at each treatment is calculated using the formula:

$$\text{RIL disease} = \left| \frac{1 - \text{AUDPC treatment}}{\text{AUDPC control}} \right| \times 100\%$$

Where,
RIL disease: The relative inhibition level of disease (%)
AUDPC : Area Under Disease Progress Curve

Serological CpMMV detection

CpMMV titers are detected serologically in samples aged four weeks after the transmission (WAT). Double-antibody sandwich-enzyme linked immunosorbent assay (DAS) ELISA method using *antibody kit* CpMMV (plus positive control) with procedures according to the antiserum manufacturer's guidelines (DSMZ, Braunschweig, Germany) was performed to identify CpMMV in the main plot. Test samples were taken from 10 plant samples from each treatment plot. Each treatment was detected by composite each of six composite samples representing replications of each treatment. The test is positive if the absorbance value of ELISA (AVE) of the test plant is twice the negative control (healthy) of the absorbance value of ELISA. The percentage relative inhibition level (RIL) of virus for the test at each treatment is calculated using the formula:

$$\text{RIL virus} = \frac{\text{AVE control} - \text{AVE treatment}}{\text{AVE control}} \times 100\%$$

Where,
RIL virus = the relative inhibition level of virus (%)
AVE = the absorbance value of ELISA

Statistical analyses

The field data was conducted using a *Completely Randomized Design* with ten treatments. Six plots on each treatment were used as replicates. The mean of disease incidence and disease severity are measured from total plants in each replicate plot on each treatment. The experiment was done in each plot size 2.6 x 6 m and spacing 60 cm with two seeds/per hole for soybean plant. Planting one or two corn lines were grown at the edge four weeks before planting soybeans. The data were subjected to analysis of variance (ANOVA) using the statistical program of SPSS. Treatment means the difference was determined by Duncan's Multiple Range Test (DMRT) at $P< 0.05$.

3. Results

The symptoms varied from healthy, presumably healthy, mild mottling, mild chlorotic blotches, moderate mosaic. Mosaic, rugose mosaic, and stunting symptoms were seen on the untreated control. The incubation period of treatment plants ranged from 9−38 days after transmission longer than the untreated control. There was no significant difference ($p < 0.05$) with the untreated control on the incubation period of CpMMV (Table 1).

The effect of planting corn lines at the edge and plant extract as bioactivator treatments in disease incidence was observed at the 1st to 7th weeks after the transmission (WAT) of *Bemisia tabaci*. These results suggest that planting corn lines at the edge with plant extract as bioactivator on soybean significantly reduced the disease incidence. This study showed that corn border crop with plant extract had a significant difference in the disease incidence from 3rd to 7th WAT. The control treatment had the highest CpMMV

disease incidence at the seventh WAT. There was no significant difference ($p < 0.05$) with soybean monoculture plus pagoda leaf extract as bioactivator on soybean (SMC + PL) from 6th to 7th WAT. Planting two corn lines at the edgewith cashew nutshell extract as bioactivator on soybean (BC 2 R + CNS) recorded the lowest disease incidence during the 6th and 7th WAT. During the 6th and 7th WAT, the stability of the disease incidence was noted on BC 2 R + CNS. There was no significant difference in disease incidence for the difference corn densities at both weeks' recordings (Table 2).

Table 1. Effect of treatments on the incubation period and typical symptom.

Treatments	Incubation periods (day)	Typical symptoms
BC 1 R + CNS	25.87 ± 2.33^{b}	Healthy
BC 2 R + CNS	38.61 ± 2.17^{a}	Healthy
SMC + CNS	23.49 ± 2.72^{b}	Healthy
BC 1 R + PL	12.70 ± 3.41^{e}	Mild chlorotic blotches
BC 2 R + PL	16.20 ± 2.70^{d}	Healthy, presumably healthy, early signs of senescing
SMC + PL	9.85 ± 1.65^{f}	Mild chlorotic blotches, moderate mosaic
BC 1 R + RG	19.55 ± 2.69^{c}	Healthy
BC 2 R + RG	21.68 ± 2.76^{c}	Healthy, early signs of senescing
SMC + RG	16.47 ± 3.19^{d}	Presumably healthy, mild chlorotic blotches
Control	8.11 ± 1.76^{f}	Mosaic, rugose mosaic, and stunting

Note: BC 1 R + CNS= planting one corn line at the edge with cashew nut shell extract as bioactivator on soybean; BC 2 R + CNS= planting two corn lines at the edge with cashew nut shell extract as bioactivator on soybean; SMC + CNS = soybean monoculture with cashew nut shell extract as bioactivator on soybean; BC 1 R + PL= planting one corn lineat the edge with pagoda leaf extract as bioactivator on soybean; BC 2 R + PL= planting two corn lines the edge with pagoda leaf extract as bioactivator on soybean; SMC + PL = soybean monoculture with pagoda leaf extract as bioactivator on soybean; BC 1 R + RG= planting one corn lines at the edge with rhizome of ginger extract as bioactivator on soybean; BC 2 R + RG= planting two corn lines at the edge with rhizome of ginger extract as bioactivator on soybean; SMC + RG = soybean monoculture with rhizome of ginger extract as bioactivator on soybean; Control = soybean monoculture without plant extract as bioactivator on soybean. According to the Duncan Multiple Range Test, values sharing the same letters differ non significantly (P < 0.05).

Table 2. The development of disease incidence at weeks after the transmission.

Treatments	Incidence of *Cowpea mild mottle virus* from ... WAT), %						
	1	2	3	4	5	6	7
BC 1 R + CNS	0.00^{b}	0.00^{c}	2.88^{d}	3.14^{e}	4.12^{f}	5.78^{e}	6.94^{e}
BC 2 R + CNS	0.00^{b}	0.00^{c}	2.64^{d}	2.91^{e}	3.98^{f}	4.15^{e}	4.15^{e}
SMC + CNS	0.00^{b}	2.10^{c}	6.42^{c}	9.55^{d}	12.76^{e}	16.20^{d}	18.25^{d}
BC 1 R + PL	0.00^{b}	9.83^{ab}	18.26^{bc}	22.80^{b}	32.95^{c}	45.85^{b}	59.12^{b}
BC 2 R + PL	0.00^{b}	4.92 b^{c}	15.54^{bc}	16.12^{c}	35.26^{c}	36.52^{c}	42.87 b^{c}
SMC + PL	0.00^{b}	11.56^{ab}	24.70^{ab}	33.75^{a}	47.09^{b}	53.17^{a}	70.11^{a}
BC 1 R + RG	0.00^{b}	5.27^{bc}	10.73^{c}	20.60^{b}	24.39^{d}	33.08^{c}	36.62^{c}
BC 2 R + RG	0.00^{b}	2.98^{c}	8.85^{c}	11.82^{c}	15.64^{e}	18.45^{d}	21.12^{d}
SMC + RG	0.00^{b}	6.75^{b}	17.07^{b}	24.96^{b}	26.78^{d}	47.14^{b}	58.66^{b}
Control	9.31^{a}	17.48^{a}	31.05^{a}	38.23^{a}	54.71^{a}	56.82^{a}	73.29^{a}

Note: WAT = weeks after the transmission. According to the Duncan Multiple Range Test, values sharing the same letters differ non significantly (P < 0.05).

Planting two corn lines at the edge with extract of cashew nutshell as bioactivator on soybean (BC 2 R + CNS) recorded the lowest disease severity during the 3rd to the 7th WAT. It was shown a similar pattern with planting one corn line at the edge with extract of cashew nutshell on soybean (BC 1 R + CNS) on disease severity during the 1st to the 7th WAT. There were significant differences when compared to soybean monoculture with CNS extract (SMC + CNS) at the 6th and the 7th WAT. During the 6th and the 7th WAT, the worst performance was planting soybean monoculture with pagoda leaf extract. It showed no significant difference from the control treatment. However, planting two corn lines at the edge with pagoda leaf extract as bioactivator on soybean (BC 2 R + PL) and planting two corn lines at the edge with rhizome of ginger extract as bioactivator on soybean (BC 2 R + RG) performed better than one corn line with pagoda leaf extract as bioactivator on soybean (BC 1 R + PL) and planting one corn line at the edge with rhizome of ginger extract as bioactivator on soybean (BC 1 R + RG), respectively (Table 3).

The analysis at the 4th WAT of data in Table 4 revealed that planting two corn lines at the edge with cashew nut shell extract as bioactivator on soybean (BC2R + CNS) had the lowest area under the disease progress curve (AUDPC) of 02.72. There was no significant difference (P<0.05) from planting one corn at the

edge with cashew nutshell extract as bioactivator on soybean (BC1R + CNS) but was significantly different (P<0.05) from the remaining eight treatments. Planting two corn lines at the edge with cashew nutshell extract as bioactivator on soybean (BC 2 R + CNS) had the highest relative inhibition level (RIL) of disease (73.11 %) and RIL of virus (61.79 %). There was the lowest absorbance value of ELISA (AVE). The titer of CPMMV concentration reached 0.20 while the AVE of the negative sample was 0.28 (the data is not shown in Table 4). The treatments BC 2 R + PL, BC 1 R + RG, SMC + RG provided no significant differences ($p < 0.05$). Among test treatments, the absorbance value test sample of control, planting one corn line at the edge with pagoda leaf extract as bioactivator on soybean (BC 1 R + PL) and soybean monoculture with pagoda leaf extract as bioactivator (SMC + PL) showed positive reaction with absorbance value of two times larger than healthy control plants.

Table 3. The development of disease severity at weeks after the transmission.

Treatments	Disease severity of *Cowpea mild mottle virus* from ... WAT), %						
	1	2	3	4	5	6	7
BC 1 R + CNS	0.00^{b}	0.00^{d}	0.83^{f}	1.19^{e}	2.26^{d}	4.86^{e}	4.86^{f}
BC 2 R + CNS	0.00^{b}	0.00^{d}	0.00^{f}	0.71^{e}	0.74^{d}	1.22^{e}	1.24^{f}
SMC + CNS	0.00^{b}	0.03^{d}	2.25^{ef}	3.40^{e}	1.57^{d}	10.78^{d}	12.48^{e}
BC 1 R + PL	0.00^{b}	3.19^{c}	14.48^{c}	19.33^{c}	31.65^{b}	44.65^{b}	53.29^{b}
BC 2 R + PL	0.00^{b}	2.88^{c}	10.15^{d}	10.88^{d}	20.11^{c}	28.78^{c}	36.60^{c}
SMC + PL	0.00^{b}	4.72^{b}	18.94^{b}	27.12^{b}	38.45^{b}	53.01^{a}	71.47^{a}
BC 1 R + RG	0.00^{b}	2.66^{c}	9.22^{d}	12.18^{d}	11.02^{cd}	25.21^{c}	29.98^{d}
BC 2 R + RG	0.00^{b}	2.15^{c}	4.78^{e}	4.78^{e}	9.26^{c}	14.62^{d}	14.99^{e}
SMC + RG	0.00^{b}	3.94^{b}	9.51^{d}	10.28^{d}	14.75^{cd}	31.10^{c}	38.05^{c}
Control	3.66^{a}	9.01^{a}	26.42^{a}	40.90^{a}	51.69^{a}	58.34^{a}	77.81^{a}

Note: WAT = weeks after the transmission. According to the Duncan Multiple Range Test, values sharing the same letters differ non significantly ($P < 0.05$).

Table 4. Effect treatments on AUDPC, absorbance value of ELISA, disease, and virus inhibition at fourth weeks after the transmission.

Treatments	AUDPC	RIL of disease (%)	Absorbance value of ELISA	RIL of virus (%)
BC 1 R + CNS	04.33^{g}	56.48	0.24^{e}	68.03
BC 2 R + CNS	02.72^{g}	61.79	0.20^{e}	73.11
SMC + CNS	13.85^{f}	54.95	0.28^{de}	64.32
BC 1 R + PL	35.21^{c}	27.68	0.65^{b}	29.41
BC 2 R + PL	26.04^{d}	34.42	0.51^{c}	31.93
SMC + PL	41.93^{b}	19.60	0.70^{a}	24.82
BC 1 R + RG	20.38^{e}	48.27	0.48^{c}	51.03
BC 2 R + RG	15.97^{f}	51.92	0.36^{d}	59.70
SMC + RG	24.50^{d}	40.83	0.59^{b}	35.95
Control	46.15^{a}	00.00	0.74^{a}	0.00

Note: AUDPC = Area Under the Disease Progress Curve; RIL = Relative Inhibition Level; ELISA = Enzyme Linked Immunosorbent Assay According to the Duncan Multiple Range Test, values sharing the same letters differ non significantly ($P < 0.05$).

4. Discussion

Planting two corn lines at the edge with cashew nutshell extract as bioactivator on soybean proved to be more effective because of the ability bioactive compound on CNS in reducing CPMMV symptoms. Moreover, the use of two corn lines at the edge can prevent the spread of *B. tabaci* to the main crop by the wind. This is in agreement with earlier findings by Andayanie et al. 2019 that the bioactive component in cashew nutshell extract consisted mainly of anacardic acid (6-pentadecylsalicylic acid) of 76.93%. Anacardic acid is a derivative of salicylic acid. The bioactive compound was assumed to play a role in increasing resistance to CPMMV infection. Other studies carried out by Andayanie and Ermawati (2019) have shown that the first to the second nymphal instar population density of *B. tabaci* could be reduced until approximately 90 % after using CNS extract at a concentration of 2.00 % in a screen house. The same trend has been observed with *Aphis gossypii* that a high-density border is associated with low population aphids in cotton crops (Gobiye et al. 2016). Similarly, in a study conducted by Damicone et al. (2007), no border crop could be associated with increasing aphid landing on plots. Thus, the main crop will have an increased

level of virus symptoms.

Planting one or two corn lines with CNS extract seems to affect the incidence of viral disease. There was an ability to attract *B. tabaci* that will prevent the migration of aphid to the main crop. Systemic acquired resistance against plant viruses can also be enhanced by natural elicitors such as bioactivator. From this study, it is evident that planting one or two corn lines with CNS extract proved to be more effective in the incidence of virl diseases. The same trend has been observed with *B. tabaci* on soybean plants where CNS extract had an antifeedant effect (Andayanie & Ermawati, 2019). On the other hand, Hooks and Fereres (2006) noted that the mechanism of suppression of a viral disease incidence could be expressed as follows: (1) barrier plants acted as natural sinks for non-persistent virus vectors that lose their ability to transmit viruses after the acquisition because of these viruses are lost on barrier plants (2) barrier plants may act as a physical barrier to transmit the virus if plants were taller than the main crop, and (3) barrier plants acted as camouflage or mask of host plants against the insect. Moreover, cashew nutshell, *Zingiber officinale*, and *Clerodendrum inerme* extracts significantly induced systemic resistance against CpMMV, BCMMV, *Tomato yellow leaf curl virus* (TYLCV), respectively compared to untreated plants (El-Sawy et al. 2017).

The control plot showed the highest percentage of disease severity from the first WAT onwards. From this study, it is evident that corn as a barrier and plant extract as bioactivator on soybean proved to be more effective than non-treated control plot because of its ability to attract aphid on the barrier crop and antifeedant effect of plant extracts. It may be due to the high population of *Bemisia tabaci* in the control treatment. On the other hand, disease severity increases progressively without plant extract as bioctivator on soybean. Moreover, several bioactive compounds in cashew nut shells as bioactivator could play a key role in this respect. The *same phenomenon* was reported by Andayanie and Ermawati (2019) that cashew nutshell compounds act as antifeedant on the nymphal stage of *B. tabaci*. The previous study was carried out using cashew nutshell extract to cause the antifeedant effect of *B. tabaci* and biactivator of soybean against CpMMV. The results of Andayanie et al. (2019) also showed that cashew nutshell extract at a concentration of 2.00 % could inhibit the landing of adults *Bemisia tabaci* on soybean leaf. Additionally, plant extracts as bioctivator had mobilized salicylic acid content on secondary metabolite production to challenge CpMMV. It can be seen in soybean monoculture with cashew nutshell extract as bioactivator on soybean. This observation could be due to the fact that bioactive compounds in CNS extract had an inhibitory effect on landing and staying for feeding deterrence on soybean leaflets. According to the previous results obtained by Andayanie et al. (2019), the content of anacardic acid in CNS extract had the dominant role as an antifeedant active compound on *B. tabaci.* This implied that planting corn on the edge with pagoda leaf and rhizome of ginger extract, respectively proved to be less effective than no barrier crop with CNS extract because the bioactive compound in CNS extract was able to inhibit the landing and staying of *B. tabaci* to the main crop for feeding deterrence. Conversely, though, the results contradict previous findings by Hooks and Fereres (2006) who suggested that the height of barrier crop is important for non-persistent virus vector control. On the other hand, the retention period of the non-persistent virus is short of several minutes to several hours.

Based on all samples, plants' serological detection from each replicate plot none of BC 2 R + PL extract, BC 2 R + RG, and SMC + RG extract was detected to be positive for CPMMV. However, some plants showed presumably healthy and early signs of senescing symptoms at the 8th WAT. These show that can be controlled CPMMV. However, due to other pathogenic infections occurring naturally in the field, symptoms were different from those of CPMMV infection. In this study, there was no other pathogenic detection, such as fungi that infect naturally because of high soil moisture in the field. Unlike BC 1R + CNS extract, BC 2 R + CNS extract and SMC + CNS extract due to the presence of CNS extract as an antifungal. This concept distinguishes typical symptoms. This is in agreement with Garcia et al. (2018), who concluded that CNSL had the highest antifungal potential in the control of *C. gloeosporioides* and *L. theobromae* in papaya fruits *C. gloeosporioides* and *L. theobromae* caused anthracnose and fruit rot, respectively. The bioactive compound of CNSL can prevent the inhibition of mycelial growth and spore production. Similarly, in a study conducted by Andayanie et al. (2021), CNS extract had a high level of total polyphenolic, flavonoid content, pH, and a low value of titratable acidity be attributed to its potent antifungal activity. The same trend has been observed in bioactive polyphenol compounds, singly or in combination. The compounds could not stimulate fungal and viral growth. The compounds may have destroyed the cell membrane fungi (Rongai et al. 2012).

The use of corn on the edge with plant extracts and soybean monoculture with cashew nutshell extract can inhibit CpMMV infection and infection through *B. tabaci*. There was not found in the BC 1 R + PL treatment. Corn border type at high density causes the main plant target to be less recognized by *B. tabaci* for colonization. However, plant extracts as an impact on resistance also have a role in inhibiting CpMMV infection and disease. Other studies carried out by Elbeshehy (2017) have been used to bioactivator against plant pathogen, including bacteria, fungi, and viruses. The inducers such as CNS extract also effectively acquired soybean against CpMMV (Andayanie et al. 2019). The value of disease severity at four weeks after the transmission was directly related to AUDPC. The virus titer of CpMMV also decreased with the relative decline in symptom severity scale. Similarly, in a study conducted by Asare-Bediako et al. (2018) that the *Cowpea mosaic virus* of disease severity was positively related to AUDPC on cowpeas genotypes resistance.

5. Conclusions

This study showed that planting corn as a barrier crop with CNS extract as bioactivator prevents migrating *B. tabaci* to the main crop and decreasing the relative inhibition level (RIL) of disease and CpMMV. Moreover, the use of CNS extract prevented adults *B. tabaci from* landing on planting soybean monoculture. There was a better decrease in RIL of disease and CpMMV than maize barrier plant with rhizome ginger extract and planting two maize barrier plant with pagoda leaf extract.

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