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Research article

## Contact effects of kaolin and other minerals on *Aphis gossypii* Glover (Hemiptera: Aphididae)

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#### Abstract

**Importance of the work**: The cotton aphid *Aphis gossypii* is a polyphagous insect found in tropical, subtropical and temperate regions. Most farmers take preventive or curative measures by applying synthetic insecticides that are usually effective in reducing aphid populations but should be used sparingly to minimize undesirable side effects. The use of natural products, such as minerals, offers a safer alternative for insect control.

<u>**Objectives**</u>: To investigate the direct contact effect of kaolin and other minerals on the mortality of *A. gossypii*, as well as the residual contact effects of kaolin.

<u>Materials & Methods</u>: In the direct contact test, each mineral suspension at a concentration of 2% (weight per volume) was sprayed onto *A. gossypii* adults. In the residual contact test, the adults of *A. gossypii* were exposed to a kaolin-sprayed chili plant.

**Results**: Out of the 10 minerals tested, 6 had significant direct contact effects on aphid mortality, with the strongest ones being kaolin and calcite. Further testing of kaolin revealed that this mineral also has significant residual contact effects against *A. gossypii*, causing mortality and a reduction in the progeny numbers of *A. gossypii*. The residual contact effects of kaolin were weaker than its direct contact effect and were significantly affected by the age of the residue.

**Main finding**: Kaolin could be applied as a curative as well as a preventive measure to reduce *A. gossypii* populations on chili plants. However, the application of kaolin (2%) needs to be repeated every 7 d to ensure its effectiveness in reducing the survival of *A. gossypii*.

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#### Introduction

Aphis gossypii Glover (Hemiptera: Aphididae), commonly known as the cotton aphid or melon aphid, is a polyphagous insect pest often found in tropical, subtropical and temperate regions (Liu et al., 2017). This insect prefers to feed on young leaves and shoots or flower buds of its host plants by sucking phloem sap using its needle-like mouthpart, resulting in the inhibition of normal plant growth, especially when the aphid population is high (Singh et al., 2014). The presence of A. gossypii is usually followed by the growth of sooty mold that utilizes the honeydew secreted by this aphid (Rondon et al., 2005). Furthermore, A. gossypii is known to be the vector of some plant viruses (Carmo-Sousa et al., 2016; Murakami and Kawano, 2017). Therefore, its population on host plants needs to be controlled immediately to avoid widespread infestation of viruses.

Preventive and curative measures can be implemented to avoid or reduce the population of *A. gossypii* on host plants. Preventive measures may include the use of resistant plant varieties (da Costa et al., 2011; Daryanto et al., 2017) and yellow traps (Buragohain et al., 2017). However, most farmers take preventive or curative measures by applying synthetic insecticides that are usually effective in reducing aphid populations but should be used sparingly to minimize undesirable side effects, such as the death of non-target organisms (Varghese and Mathew, 2012).

The use of natural products, such as minerals, offers a safer alternative for insect control and the mineral kaolin has been reported to be active against several insect pests, such as fruit flies (Saour and Makee, 2004; Braham et al., 2007), caterpillars (Alavo et al., 2010), whiteflies (Núñez-López et al., 2015; Baiomy, 2017), leafhoppers (Tacoli et al., 2017), thrips (Larentzaki et al., 2008; Tyler-Julian et al., 2018) and aphids (Barker et al., 2007; Alavo et al., 2011). Other minerals that have insect control properties include sulfur (Pérez-Guerrero and Molina, 2016), bentonite (Abd El-Aziz, 2013), calcium carbonate (Ramírez-Godoy et al., 2018), calcium oxide (Smitha and Mathew, 2010), zeolite (Rumbos et al., 2016) and zinc oxide (Khooshe-Bast et al., 2016). However, very little is known about the effect of other minerals on *A. gossypii* and other aphids.

Studies on the reactions of *A. gossypii* and other aphid species to kaolin have reported very different results. Alavo et al. (2011) showed that the application of kaolin particles at a concentration of 5% (weight per volume, w/v) significantly reduced the *A. gossypii* population on cotton. In contrast, Showler and Amstrong (2007) suggested that the use of kaolin (6%; w/v) could exacerbate infestations of this aphid on cotton. Furthermore,

Marko et al. (2008) found that the application of kaolin in apple orchards resulted in a significantly lower number of green apple aphid (*Aphis pomi*) colonies, but it led to a significantly higher number of rosy apple aphid (*Dysaphis plantaginea*) colonies.

Very little is known about the mechanisms of kaolin against *A. gossypii*. However, kaolin is unlikely to act as a stomach poison because it may not be able to enter leaf phloem, where *A. gossypii* usually obtains plant sap by its stylet. Therefore, the bioactivities of kaolin most likely affect *A. gossypii* through contact either directly or indirectly (residual effects) or both. The current study aimed to investigate the direct contact effect of kaolin and other minerals on the mortality of *A. gossypii*, as well as the residual contact effects of kaolin. Investigated minerals were selected based on their relatively low impacts on humans and the environment, availability in nature and their bioactivities to insect pests.

#### **Materials and Methods**

#### Minerals

The minerals used in this study were purchased from PT Brataco Chemica (kaolin, talc, zinc oxide, bentonite, calcium oxide, and calcium hydroxide), PT Sakura Medikal (calcium carbonate), Kimia Mart (sulfur), Berkah Jaya Farm Shop (dolomite) and Kucingbilly Shop (zeolite). All the minerals were locally manufactured in Indonesia, except for kaolin (Takehara Kagaku Kogyo, Japan) and talc (Haichen Da Feng Mineral Factory, China). The minerals were passed through a 200-mesh sieve and then a 400-mesh sieve to ensure the maximum particle size was 37 µm. After that, each mineral (80% w/w) was mixed with a wetting agent—sodium lauryl sulfate (10%; w/w)—and sodium naphthalene sulfonate formaldehyde condensates (10%; w/w) as a dispersant agent.

#### Aphis gossypii rearing

A colony of *A. gossypii* was collected from a chili plant grown in a field and then transferred to a rearing cage containing aphid-free chili plants. The aphid species was determined using an identification key (Blackman and Eastop, 2000). The chili plants used for the experiment were grown in polybags filled with a mixture of soil, fertilizer and compost (2:1 v/v) and watered regularly. The cage was covered with mesh on the side walls and transparent plastic on the top. Insect rearing was conducted inside a glasshouse.

#### Direct contact test of kaolin and other minerals to A. gossypii

Ten mineral suspensions were made by mixing each mineral formulation with water at a concentration of 2% (w/v). For comparison, an emulsion of synthetic insecticide formulation (deltamethrin 25 g/L) was made at a concentration of 1 mL/L. Water was used for the control. Each mineral suspension (1 mL) was sprayed by hand onto 20 adults of A. gossypii placed on tissue paper. Before use, each hand sprayer was calibrated to have the same flow rate. The position of the sprayer (nozzle) was kept approximately 20 cm from the aphids to be sprayed. After a few minutes, the treated A. gossypii were transferred to dry tissue paper for further air drying. Next, the treated aphids were placed on a chili leaf. Wet cotton and plastic zipper bags were used to wrap the base of the leaf stalk to maintain its freshness. This experiment was replicated four times in a randomized block design. Observations were recorded 24 hr and 48 hr after treatment regarding the numbers of dead A. gossypii that were used to calculate the mortality rates of the treated Aphid. Schneider-Orelli's formula was applied to correct the mortality of A. gossypii in treatments in comparison to the control (water) (Puntener, 1981): Corrected mortality = ([% mortality in treatment - % mortality in control]/ [100 - % mortality in control]) x 100.

#### Residual contact test of kaolin to A. gossypii

Kaolin residues of various ages on the young chili plants were evaluated for their effects on the mortality rates and progeny numbers of A. gossypii. A kaolin suspension (2 mL) at a concentration of 2% (w/v) was sprayed by hand on the young chili plants at 7 d, 5 d, 3 d, 1 d and 0 d before the infestation of the plants with A. gossypii. For the treatment of 0 d residue (fresh), the plant was sprayed with kaolin suspension about 1 hr before aphid infestation. Two controls were provided. First, a young chili plant was sprayed with water containing the wetting and dispersant agents of the kaolin formulation. For the second, a young chili plant was sprayed only with water. As in the direct contact test, the chili plants were grown in polybags (11 cm in diameter) filled with a mixture of soil and compost (2:1 v/v). Each chili plant was infested with 20 adults (females) of A. gossypii. This experiment was replicated four times and arranged in a randomized complete block design. Observations on the mortality of A. gossypii and the number of A. gossypii progeny were recorded daily for 6 d. Chili plant shoots and leaves were carefully checked using a magnifying glass to count the number of aphid progenies.

#### Statistical analysis

All data were analyzed using the Minitab Version 18 statistical software (Minitab LLC, Pennsylvania, PA, USA). Transformation of data was applied when requirements for normality and homogeneity of variance were not satisfied. A general linear model for the analysis of variance was used to determine whether there were any significant differences between treatments based on Tukey's test at the 95% confidence level. Mortality data at 24 hr after treatment were transformed using ASIN (SQRT(X/100)) ×180 / (22/7) prior to analysis by GLM of ANOVA.

#### Results

### Direct contact effect of kaolin and other minerals on mortality of A. gossypii

The results showed that some minerals had significant effects on the mortality of A. gossypii at 24 hr ( $F_{11, 33} = 9.92$ ; p = 0.000) and 48 hr ( $F_{11, 33} = 8.88$ ; p = 0.000) after spraying (Table 1). Of the 10 minerals tested, the most effective ones were kaolin and calcite of which the mortality of A. gossypii was not significantly different with the control (deltamethrin spray at 1 mL/L). Spraying of the two minerals resulted in 70.00% and 71.43% corrected mortality at 48 hr after treatment, respectively. Other minerals that had significant effects on aphid mortality were tale, zinc oxide, sulfur and dolomite.

#### Residual contact effect of kaolin on mortality of A. gossypii

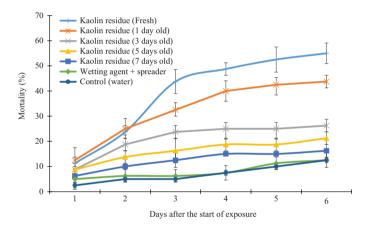
The presence of kaolin particles on the chili plants caused the deaths of some adults of *A. gossypii* at various rates, depending on the age of the residue and the duration of the aphids' exposure (Fig. 1). The older the residue of kaolin, the lower the effect on aphid mortality. At the same time, the longer the exposure, the stronger the effect on aphid mortality.

Based on data from the last observation (6 d after exposure), kaolin residue had significant effects on *A. gossypii* mortality ( $F_{6,18} = 119.35$ ; p = 0.000), as shown in Fig. 2. Fresh kaolin residue yielded the highest aphid mortality (55%). The kaolin residues that were aged 1 d, 3 d and 5 d resulted in lower aphid mortality compared to the fresh kaolin residue (0 d), but they were still significantly different from the control. On the other hand, the kaolin residue aged 7 d failed to have a significant effect on aphid mortality. The wetting agent and spreader used in the kaolin formula had no significant effect on aphid mortality after 6 d of exposure.

Table 1	Mortality (± SD)	of Aphis gossyn	ii sprayed with min	eral suspension at	2% concentration

Mineral (2%)	After 24 hr		After 48 hr	
_	Mortality (%)	Corrected mortality (%)	Mortality (%)	Corrected mortality (%)
Kaolin	50.00±23.45 <sup>cde</sup>	49.37	73.75±6.29 <sup>cd</sup>	70.00
Talc	$15.00 \pm 5.77^{abc}$	13.92	$61.25 \pm 17.02^{bcd}$	55.71
Zink oxide	$32.50 \pm 6.45^{bcd}$	31.65	$55.00\pm10.80^{bcd}$	48.57
Bentonite	$27.50 \pm 6.45^{abc}$	26.58	$35.00 \pm 12.25^{ab}$	25.71
Sulfur	$23.75{\pm}8.54^{abc}$	22.78	$55.00\pm14.72^{bcd}$	48.57
Dolomite	$38.75 \pm 6.29^{bcd}$	37.97	$67.50\pm22.17^{bcd}$	62.86
Calcium oxide	$12.50 \pm 15.55^{ab}$	11.39	$38.75 \pm 18.43^{ab}$	30.00
Calcium hydroxide	$22.50{\pm}16.58^{abc}$	21.52	$43.75\pm19.74^{abc}$	35.71
Calcite	$56.25 \pm 12.50^{de}$	55.70	$75.00\pm14.72^{cd}$	71.43
Zeolite	$26.25{\pm}2.50^{abc}$	25.32	$36.25 \pm 6.29^{ab}$	27.14
Deltamethrin 25 g/L (1 mL/L)	68.75±13.77e	68.35	83.75±4.79 <sup>d</sup>	81.43
Control	$1.25\pm2.50^a$	-	12.50±6.45a	-

Mean $\pm$ SD within a column superscripted with different lowercase letters are significantly (p < 0.05) different



**Fig. 1** Mean (±SD) mortality of *Aphis gossypii* adults after 1–6 d exposure to kaolin particle film

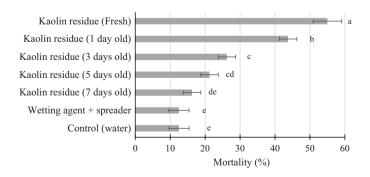
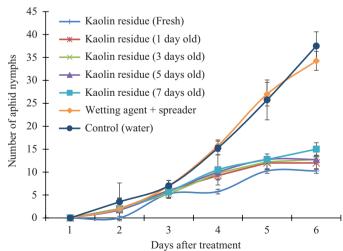


Fig. 2 Mean ( $\pm$ SD) mortality of *Aphis gossypii* adults after 6 d of exposure to kaolin particle film. Means labelled with different lowercase letters are significantly (p < 0.05) different

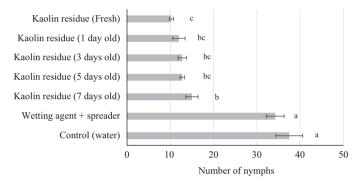
## Residual contact effect of kaolin on number of A. gossypii progenies

As Fig. 3 shows, the number of *A. gossypii* nymphs was affected by the presence of kaolin particles on chili plants. For the two controls (water and the wetting agent and spreader), the number of nymphs continued to rise during the observation period with a sharp increase at 3–6 d after leaf infestation. On the other hand, on the plants treated with kaolin particles, the number of nymphs grew slowly, and population growth even stagnated 5–6 d after exposure when treated with kaolin residue aged no more than 5 d.



**Fig. 3** Mean (±SD) number of *Aphis gossypii* progeny on kaolin-sprayed chili plant after 1–6 d of exposure

As shown by the last observation (Fig. 4), all the kaolin residue treatments significantly reduced the population of *A. gossypii* nymphs compared to the control ( $F_{6, 18} = 197.96$ ; p = 0.000). The lowest number of nymphs was found on the chili plants previously treated with kaolin on the day of treatment (10.25 nymphs) and the highest was on the control (37.50 nymphs) plants. There was no significant difference in the number of nymphs recorded on plants treated with kaolin residue ages of 0 (fresh), 1, 3, and 5 days old (10.25–12.75 nymphs). The wetting agent and spreader in the formulation of kaolin did not have a significant effect on the number of *A. gossypii* nymphs.



**Fig. 4** Mean ( $\pm$ SD) number of *Aphis gossypii* progeny on kaolin-sprayed chili plant after 6 d of exposure, where means superscripted with different lowercase letters are significantly (p < 0.05) different

#### Discussion

The current study found that kaolin and other minerals, namely calcite, dolomite, talc, zinc oxide and sulfur had direct contact effects on the mortality of *A. gossypii*. Of these, the ones with the strongest effect and thus the most potent were kaolin and calcite. Kaolin has the advantage of a near-neutral pH compared with calcite which is basic. Therefore, kaolin is less likely to cause a marked change in soil pH after its field application for insect controls.

Based on a literature search, this may be the first published report on the direct contact effects of the above minerals on the mortality of *A. gossypii*. Two previous studies have shown that kaolin can significantly reduce the number of *A. gossypii* settling on kaolin-treated plants (Garzo et al., 2003; Alavo et al., 2011). However, the reductions in *A. gossypii* population in these two studies are more likely to involve residual effects of kaolin particles deposited on the plant surface.

Little is known about the direct contact effects of kaolin on aphids, while even less is known about the effects of calcite. Barker et al. (2007) reported that spraying kaolin suspension on green peach aphids *Myzus persicae* (Sulzer) did not have a significant impact on their subsequent survival or reproduction on untreated plants. Similarly, a laboratory study by Tacoli et al. (2017) indicated that kaolin spray with a concentration of 4% (w/v) did not significantly affect the survival of *Empoasca vitis* Goethe nymphs during the 7 d of observation.

Nevertheless, it has been observed that kaolin can affect the cuticular permeability of an arthropod (Cook et al., 2008), which may trigger excessive loss of water from the body. Swamiappan et al. (1976) revealed that the application of kaolinitic clay caused the adsorption of lipoid particles in the epicuticular wax of storage insects; subsequently, this resulted in a loss in body weight. Therefore, the death of *A. gossypii* in our study could be attributed to desiccation. No reports have been found about the direct contact effect of calcite on *A. gossypii*. However, colloidal (< 10 μm) and nano-sized (20–100 nm) particles of calcite (calcium carbonate) had direct contact effects on oriental fruit flies (*Bactrocera dorsalis* Hendel) with LC<sub>50</sub> values of 9,330 mg/mL and 6,530 mg/L, respectively (Hua et al., 2015).

In addition to these effects from direct contact, the current study also found that kaolin had residual contact effects on A. gossypii consistent with reports on A. gossypii and other species of aphids, using different host plants. Garzo et al. (2003) demonstrated that kaolin particle film can reduce the settlement of A. gossypii on treated melon plants. Another study found that kaolin-based particle film reduced the number of black pecan aphids (Melanocallis carvaefoliae Davis) visiting the treated foliage and it caused mortality, as well as the reduction of progeny (Cottrell et al., 2002). Furthermore, both choice and no-choice tests (field cages) showed that branches treated with processed-kaolin particle film received lower numbers of the rosy apple aphid Dysaphis plantaginea (Pass.) compared to the untreated control (Bürgel et al., 2005). By contrast, Barker et al. (2007) showed that in a no-choice test, the survival and reproduction of M. persicae were not affected by kaolin-treated host plants, although they preferred the untreated leaf areas in a choice test.

Kaolin residue has also been reported to be effective against other insect pests. Miranda et al. (2018) reported that kaolin residue reduced the number of the Asian citrus psyllid *Diaphorina citri* Kuwayama settling on kaolin-treated sweet orange seedlings and trees. Similarly, Unruh et al. (2000) found that kaolin residue reduced oviposition of the codling moth *Cydia pomonella* (L.) on kaolin-treated apple fruits and reduced fruit infestation on kaolin-treated trees. In addition,

Kuhar et al. (2019) found fewer brown marmorated stink bugs *Halyomorpha halys* (Stal) settling on the fruit and leaves of cherry tomato plants that had been treated with kaolin compared to those on an untreated control. Furthermore, kaolin application to a peach tree was comparable with or better than standard pesticides in controlling populations of the oriental fruit moth *Grapholita molesta* (Busck), plum curculio *Conotrachelus nenuphar* (Herbst) and the Japanese beetle, *Popillia japonica* Newman (Lalancette et al., 2005).

The current study indicated that the residual effects of kaolin are affected by the age of the residue (particle film). The older the kaolin residue was, the less potent the effects on A. gossypii, especially mortality. Kaolin residue aged up to 5 d had significant effects on A. gossypii mortality, while the residue aged up to 7 d significantly affected the progeny number. It seemed that the reduction in the number of aphid progenies (nymphs) in the current investigation was influenced mainly by the decline in the number of surviving aphids and to a certain extent by the sub-lethal effect of the kaolin residue. The latter was likely to occur on aphids treated with kaolin residue aged 3 d and 5 d, where they had a higher number of surviving aphids but produced equal numbers of progenies compared to the fresh kaolin residue and kaolin residue aged 1 d. Nevertheless, further studies need to be conducted to reveal the extent of the sub-lethal effect of kaolin residue on the number of aphid progenies.

Another study found that weekly sprays of kaolin (5%) effectively reduced the population of the aphid *Lipaphis erysimi* Kalt. on cabbage (Alavo and Abagli, 2011). In addition, Smaili et al. (2014) reported that a foliar application of kaolin decreased the density of the aphids *Toxoptera aurantii* Boyer de Fonscolombe and *Aphis spiraecola* Pach for 1 wk. In addition, repeated applications of kaolin significantly reduced *D. plantaginea* females, whereas a single application of kaolin failed to provide a significant effect on the aphid (Bürgel et al., 2005). Therefore, it could be expected that repeated weekly applications of kaolin suspension on chili plants could suppress a population of *A. gossypii*.

This study demonstrated that the kaolin residue on chili leaves had a lower and slower mortality effect on *A. gossypii* than its direct contact application. It seemed that the death of *A. gossypii* during the residual contact test mainly occurred due to the presence of the kaolin particle film on the leaves, which disturbed the feeding activity of this insect. There are several possible mechanisms by which kaolin residue can affect *A. gossypii*. For example, it may interfere with the ability of *A. gossypii* to hold onto the plant and move around.

Such reactions were observed by Hall et al. (2007) when testing the effect of applying kaolin particle film to citrus leaves on adults of the psyllid *Diaphorina citri* Kuwayama. In addition, Unruh et al. (2000) indicated that kaolin residue on leafless apple shoots reduced the mobility of codling moth Cydia pomonella (L.) larvae. Salerno et al. (2020) showed that kaolin nano particles reduced the ability of the southern green stink bug Nezara viridula L. and the Mediterranean fruit fly Ceratitis capitata Wiedemann to attach to both natural and artificial surfaces. In another study, Sackett et al. (2005) demonstrated that kaolin was active against larvae of the oblique-banded leafroller Choristoneura rosaceana (Harris) as a physical barrier to feeding. The irritant effects of kaolin on insects have also been examined (Yee, 2008); kaolin may have disturbed the feeding activities of A. gossypii on the kaolin-treated leaves in the current study. While another study showed that kaolin did not discourage the Asian citrus psyllid Diaphorina citri Kuwayama from probing, this treatment reduced the proportion of psyllid individuals that were able to reach phloem (Miranda et al., 2018). Besides its ability to reduce insect pest infestation and solar injury, kaolin application may have effects on the physiology of the treated plants. A study by Cantore et al. (2009) revealed that kaolintreated tomato plants experienced a reduction in photosynthesis and dry biomass. The photosynthetic rate of kaolin-treated citrus trees was reduced (Ramirez-Godoy et al., 2018). It was suggested that kaolin-coated leaves reflected more light and this resulted in less light available for photosynthesis (Le Grange et al., 2004). Nevertheless, kaolin sprays are effective in alleviating the negative effects of drought, light and heat stresses (Denaxa et al., 2012; Gharaghani et al., 2018; Brito et al., 2021; Mahmoudian et al., 2021). Further studies are required to find the concentration of kaolin and its frequency of application that can effectively control the aphid population on chili plants in the field without causing side effects to the plants.

In summary, out of the 10 minerals tested, kaolin and calcite were the strongest for controlling *A. gossypii* through direct contact. Kaolin had a residual contact effect on the mortality of *A. gossypii* but this was weaker than its direct contact effect and was related to the age of the residue. The presence of the kaolin residue reduced progeny numbers of *A. gossypii*. The current results suggested that kaolin could be applied as part of curative as well as preventive measures to reduce *A. gossypii* populations on chili plants. However, the application of kaolin (2%) needs to be repeated every 7 d to ensure its effectiveness in reducing the survival of *A. gossypii*.

#### **Conflict of Interest**

The authors declare that there are no conflicts of interest.

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