Kasetsart J. (Nat. Sci.) 47 : 323 - 332 (2013)

Selection of the Most Appropriate Coating Particle Film for Improving Photosynthesis in Mango

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ABSTRACT

Incremental irradiation, especially during summer, decreases net photosynthesis. Sunburn causes damage to leaves and fruit and consequently decreases the fruit yield and quality. Trials have reported success in using particle film technology for coating fruit tree leaves to remedy these adverse effects of strong summer irradiation. The objectives of this experiment were to compare the suspension properties, precipitation and light transmission of four coating materials—kaolin, bentonite, calcium carbonate and dolomite—and to study the leaf coating properties by measuring photosynthesis after the application of materials. The results showed that 60 $g.L^{-1}$ kaolin had good suspension in water and was the slowest to precipitate. Kaolin was the most effective leaf coating material to reduce transmission through a glass plate by 70.14%. The average percentage of photosynthetic photon flux through a glass plate for bentonite, calcium carbonate and dolomite was 93.50, 92.28 and 86.78%, respectively. Mango leaves sprayed with kaolin had higher average net photosynthesis (P_n), stomatal conductance (g_s) and transpiration rate (E) than in bentonite and untreated leaves which had significantly different average values for P_n, g_s and E. Therefore, kaolin was suitable for use as a coating material on mango leaves.
Keywords: kaolin, bentonite, calcium carbonate, dolomite, gas exchange

INTRODUCTION

Mango (*Mangifera indica* L.) is one of the major economic fruits in Thailand and continues to be one of the major export crops in many tropical countries. Mango trees are cultivated in all parts of Thailand. However, the major growers face low yields and poor quality due to a reduction in photosynthesis and increased diseases (Sangchote, 1987; Spreer *et al*., 2009).

Severe climate change may result in high temperatures and severe drought, particularly during summer, which can result in a midday depression of photosynthesis. The main physiological processes responsible for the midday depression of photosynthesis are stomatal closure or photosystem II (PSII) photoinhibition or both (Muraoka *et al*., 2000). Flexas and Medrano (2002) reported that under severe drought stress, C_3 plants had to close stomata indicating a dominant limitation on photosynthesis.

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Received date : 18/01/13 Accepted date : 02/05/13

Reflective materials can be applied as a leaf or fruit particle film coating to reduce solar heat stress, especially in areas with hot or sunny weather for a substantial part of the year. Such coatings can reduce heat stress, the extent of solar-injured fruit and water stress, and are involved in pest control and the suppression of disease incidence (Glenn and Puterka, 2005). Some of the reflective materials that may be used as leaf coating material include kaolin, bentonite and calcium carbonate. Kaolin or kaolinite, a white aluminosilicate clay with fine, porous and non-abrasive particles, disperses easily in water (Glenn and Puterka, 2005). This inert mineral is used in the paper, paint and plastic industries, depending on the processing method and the characteristic size, dimensions and brightness of the particles. It is used in pharmaceutical drugs due to its safety for ingestion (McBride, 2000). It is also a component of toothpaste, cosmetics and alimentary products (Glenn and Puterka, 2005). Bentonite is a naturally occurring plastic clay composed mainly of montmorillonite. Its volume increases several times on contact with water, creating a gelatinous and viscous fluid (Stoykov *et al.*, 2007). Bentonite's properties of water absorption and viscosity make it a valuable material for a wide range of applications and it is frequently used as a binding, sealing, absorbing and lubricating agent (World Health Organization, 2005). Calcium carbonate ($CaCO_3$) is a white, nontoxic, odorless solid. It is one of the most common and widely dispersed minerals occurring in eggshells, limestone, marble, seashells and other biominerals (McGregor, 1963). Because of its harmlessness and low cost, calcium carbonate has been used for a variety of purposes such as a neutralizing agent, filler, flux and in cement (McGregor, 1963; Sheikholeslami and Ong, 2003).

Researchers have studied the effects of these reflective clay particle films on plant physiology processes. For example, kaolin particle film is associated with increased water use efficiency in citrus under excessive heat stresses (Jifon and Syvertsen, 2003) and kaolin applications appear to reduce the effect of high temperatures on apple fruit thereby increasing carbon assimilation and stomatal conductance (Glenn *et al*., 2003). In addition, kaolin treatment increased fruit weight and redness of 'Empire' apples (Glenn *et al.*, 2002, 2003). Although calcium carbonate has none of the positive effects of kaolin particle film, it reflected more photosynthetically active radiation (PAR) than kaolin (Glenn *et al*., 2003). Spraying citrus (*Citrus auranticola* (Christm.) Swing.) plants with kaolin or mixtures of kaolin and bentonite (montmorillonite) reduced natural spiraea aphid (*Aphis citricola* van der Goot) colonization, due to the white clay coating deposit on the leaves (Bar-Joseph and Frenkel, 1983). There has been no report on the application of bentonite as a leaf coating material. However, bentonite (a white-colored particle) can increases the adhesion of clay suspensions on the leaves (Bar-Joseph and Frenkel, 1983). Thus, bentonite was selected to test for its leaf coating properties.

The physical properties of a film can reduce damage from insects and plant pathogens, while enhancing photosynthesis, yield and fruit quality, particularly in a hot and dry climate. For example, Glenn and Puterka (2005) advised that the mineral particle film must have the following properties: (1) chemically inert, (2) particle diameter less than 2 μm, (3) creation of a uniform film, (4) formation of a porous film allowing leaf gas exchange, (5) transmission of PAR and reflection of ultraviolet (UV) and infrared radiation, (6) interference with insect or pathogen behavior and (7) ability to be washed off from the harvested products.

Thailand is one of the largest current producers of clay minerals in Asia (World Health Organization, 2005). Mango is one of the nation's major export fruits (Mendoza and Wills, 1984; Litz, 1997); thus, mango fruit yield and quality should be improved in order to increase the economic value. No publications were identified on the use of clay minerals in Thailand as a reflective particle film. The objectives of this experiment were to study the suspension, precipitation and light transmission of four coating leaf materials—bentonite, calcium carbonate, dolomite and kaolin clays—for developing a leaf coating material for mango. In addition, the leaf coating abilities of these materials were also investigated by measuring photosynthesis after application.

MATERIALS AND METHODS

The effectiveness for photosynthesis improvement was investigated using mango leaf coatings of kaolin, bentonite, calcium carbonate and dolomite with particle sizes of 325, 240, 240 and 200 mesh, respectively. All four materials were purchased from Bullazia Agrifluids Co. Ltd., Nonthaburi, Thailand. Laboratory and field tests were performed at Kasetsart University, Bangkok, Thailand (13°50'50.39"N, 100°34'19.44"E). The suspendability, precipitation time of the precipitable particles, photon transmission through the particle film, leaf coating ability and gas exchange of the coated leaves were investigated. The following four experiments were performed.

Suspendability and precipitation time of tested substances

Suspendability in water and precipitation times of the dispersing particles were studied. According to previous work by the current authors (data not shown), the concentration of each material was varied from 50 to 70 $g.L^{-1}$. It was found that 60 $g.L^{-1}$ of each material was the optimum concentration to produce suspendability in water with the precipitation time of the dispersing particles being 2 h. Consequently, each material at the rate of 60 $g.L^{-1}$ was well stirred in water with the addition of 1–2 drops of sodium silicate to retard precipitation. The suspensions, after thorough stirring, were left undisturbed for 3 h to allow the precipitable particles to sink to the bottom and the precipitation times were recorded. The average precipitation time from three repetitions was taken for each material.

Photon transmission of coating films

Each leaf coating material was sprayed at the rate of 60 $g.L^{-1}$ as a coating onto a glass plate 12 × 20 cm and allowed to dry at ambient temperature. The glass plate coated with the particle film was placed under direct sunlight and the photosynthetic photon flux (PPF) was recorded continuously using a quantum light sensor (Watch Dog model 450; Spectrum Technologies, Inc.; Plainfield, IL, USA) placed 10 cm below the plate. The mean percentage PPF indicating the photon transmittance of coated glass plate was determined as the percentage transmittance of photons through an uncoated glass plate.

Mango leaf coating ability and coated leaf gas exchange measurements

Kaolin and bentonite which previously had been found to form satisfactory films and produce satisfactory photon transmission were used in this experiment. Each of the materials was suspended well in water at a rate of 60 $g.L^{-1}$. The aqueous suspension was sprayed on each mango tree twice a week using a hand-held sprayer. In total, 12 applications were made from 1 November to 16 December 2008 to maintain a uniform coating of the film on the leaves of treated trees throughout the study period.

Leaf gas exchange was measured with a portable photosynthetic system (LI-6400; LI-COR Inc.; Lincoln, NE, USA). The leaf sampled was clamped inside the leaf chamber of a portable photosynthesis system with the upper leaf surface inside the chamber fully exposed to direct sunlight. Leaf samples were chosen from the first mature leaves at exterior canopy positions after kaolin spraying. The measurements were performed on selected, clear, sunny days. Measurements of leaf gas exchange were made during the period from 1130 to 1330 hours. The net photosynthesis (P_n), stomatal conductance (g_s), transpiration rate (E), leaf temperature (T_{leaf}), leaf to air vapor pressure deficit ($VPD_{leaf\text{-}air}$), intercellular CO_2 concentration (C_i), air CO_2 concentration (C_a), ratio of intercellular to air CO_2 concentration (C_i:C_a), photosynthetic photon flux (PPF), air temperature (T_{air}) and relative humidity (RH) were measured under ambient conditions. All measurements were performed at the CO_2 concentration of 400 μmol.mol^{-1}. The air vapor pressure deficit (VPD_{air}) was calculated from T_{air} and RH, according to Goudriaan and van Laar (1994). VPD_{air} was calculated using Equation 1:

$$VPD_{air} = e^0 - e \quad (1)$$

where e^0 is the saturation vapor pressure (kPa). e^0 can be calculated (LI-COR ,1990) using Equation 2:

$$e^0(T_{air}) = 0.61083 \times 10^{\frac{7.6448T}{242.62+T}} \quad (2)$$

where T is the dewpoint temperature (°C).

When e is the actual vapor pressure (kPa), e can be calculated using Equation 3:

$$e = e^0_{Tair} \times (1 - RH_{air}) \quad (3)$$

where T_{air} is the air temperature (°C) and RH_{air} is the percentage of the amount of water that the air can hold at a given temperature (%). RH and T_{air} must be taken in the same time period.

Light response curve of mango

Measurements of light response curves of photosynthetic characteristics were carried out on the same leaves that were evaluated for diurnal changes in gas exchange. A light response curve was obtained with PPF values of 2000, 1600, 1200, 1000, 800, 600, 400, 200, 100, 75, 50, 25 and 0 μmol PPF m^{-2}.s^{-1}, adjusted automatically by a red-blue light-emitting diode light source (LI-6400; LI-COR Inc.; Lincoln, NE, USA). The CO_2 concentration and temperature in the leaf chamber were maintained at 400 μmol CO_2 mol^{-1} and 37 °C, respectively. Leaves were allowed to stabilize at each light step for a minimum of 3 min and then data were locked after the steady-state condition was achieved. The data were fitted to a non-rectangular hyperbolic function (Thornley and Johnson, 1990) shown in Equation 4:

$$A = \frac{1}{2\theta}\left[\alpha I + P_m - \sqrt{(\alpha I + P_m)^2 + 4\theta\alpha I P_m}\right] - R_d \quad (4)$$

where A is the net photosynthesis (μmol CO_2 m^{-2}.s^{-1}), I is photosynthetic photon flux (μmol PPF m^{-2}.s^{-1}), P_m is the maximum net photosynthesis rate (μmol CO_2 m^{-2}.s^{-1}), R_d is the darkness respiration rate (μmol CO_2 m^{-2}.s^{-1}), α is the quantum or photochemical efficiency (mol CO_2 mol^{-1} PPF) and θ is the curvature factor.

Each parameter of the leaf photosynthetic response to PPF was recorded and the average determined from four leaves. The average values of each parameter were used in the model of the leaf photosynthetic response to PPF to describe the curve.

Data analysis

A completely randomized design was performed for precipitation and photosynthetic gas exchange. Where significant differences were found due to treatment, Duncan's multiple range test was applied. Differences were considered significant at $P \leq 0.05$.

RESULTS

Suspendability and precipitability

Table 1 shows the average precipitation

times of suspended particles of kaolin, bentonite, calcium carbonate and dolomite. The kaolin particles exhibited the longest precipitation times of 75.66 min followed by bentonite, calcium carbonate and dolomite particles with 48.33, 27.66 and 25.66 min, respectively.

Transmission of particle film coated

The results on transmission of photons through a suspended material coating on a glass plate (Figure 1 and Table 2) indicated that kaolin was the most effective coating material based on the PPF values as the kaolin coating was the lowest. The average percentage of PPF for kaolin was 70.14%, while those for bentonite, calcium carbonate and dolomite were almost the same. However, the PPF values of light in these three treatments were higher than for kaolin. The average percentage PPF through a coating on a glass plate of bentonite and calcium carbonate were 93.50% and 92.28%, respectively. Furthermore, the average percentage PPF through the dolomite-coated glass plate was 86.78%.

Leaf coating ability

The microclimate during the period of measurement was characterized as hot and dry**.** The average midday PPF was 1,462.67 µmol PPF $m^{-2}.s^{-1}$. The average T_{air} reached 41.49 °C, while the average RH was 20.78%. The VPD_{air} value was 6.34 kPa (Table 3).

Table 1 Average times of precipitation of suspended particles of kaolin, bentonite, calcium carbonate and dolomite.

Coating material	Precipitation time (min)
Kaolin	75.66 ± 2.33^{a}
Bentonite	48.33 ± 1.66^{b}
Calcium carbonate	27.66 ± 1.45^{c}
Dolomite	25.66 ± 0.66^{c}

Means followed by the same lowercase superscript letter within a column are not significantly different by Duncan's multiple range test at the $P \leq 0.05$ level.

Table 2 Average percentage of photosynthetic photon flux (PPF) through a glass plate coated with different materials.

Coating material	PPF transmittance (%)
Kaolin	70.14
Bentonite	93.50
Calcium carbonate	92.28
Dolomite	86.78

Table 3 Microclimatic parameters measured from 1130 to 1330 hours.

Parameter	Average*
PPF (µmol PPF $m^{-2}.s^{-1}$)	1462.67 ± 25.67
T_{air} (°C)	41.49 ± 0.32
RH (%)	20.78 ± 0.47
VPD_{air} (kPa)	6.34 ± 0.14

PPF = Photosynthetic photon flux, T_{air} = Air temperature, RH = Relative humidity, VPD_{air} = The air vapor pressure deficit.
* = Mean value ±SE of 12 replications.

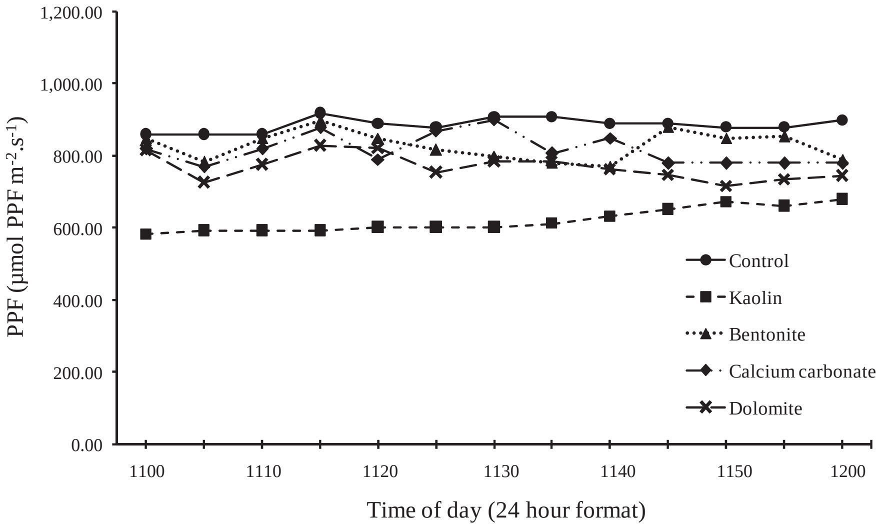

Figure 1 Photosynthetic photon flux (PPF) of light passing through glass coated by different coating materials.

The P_n values of the kaolin-sprayed leaves were greater than for the bentonite and untreated leaves throughout the measurement interval (Figure 2). The average P_n value in kaolin-sprayed leaves was significantly higher than in other treatments. Moreover, g_s and E values were similarly higher in kaolin-sprayed leaves than in bentonite and untreated leaves. The values of C_i and C_i:C_a in kaolin-treated leaves were higher than those in untreated leaves, but were lower than those of the bentonite treatment. Mango leaves treated with kaolin had lower average T_{leaf} and $VPD_{leaf\text{-}air}$ values than in the bentonite and untreated leaves which had average T_{leaf} and $VPD_{leaf\text{-}air}$ values that were not significantly different (Table 4).

On the contrary, the P_n values for the bentonite-sprayed leaves generally were lower than in the koalin and untreated leaves throughout the measurement interval (Figure 2). Furthermore, the P_n values in the bentonite-sprayed leaves were significantly the lowest (Table 4). In addition, the values of g_s and E in bentonite were significantly lower than those in the kaolin and untreated leaves. Nevertheless, the values of C_i and C_i:C_a in the bentonite treatment were significantly higher than those of the kaolin treatments (Table 4).

Light response curves of P_n

The light response curve of mango leaves exhibited a saturation phenomenon with the relationship between P_n and PPF. The P_n value increased rapidly as PPF increased to 200 µmol PPF $m^{-2}.s^{-1}$ and then increased slowly to the maximum, followed by a slow decrease as PPF increased to 2,000 µmol PPF $m^{-2}.s^{-1}$ (Figure 3). The value of P_n was found to be saturated at around 600 µmol PPF $m^{-2}.s^{-1}$. The maximum rate of photosynthesis (P_{max}) was determined at 3.5 µmol CO_2 $m^{-2}.s^{-1}$ (Figure 3). However, when the data were fitted to a non-rectangular hyperbola function (Thornley and Johnson, 1990), the light saturation point was calculated as 232.93. µmol PPF $m^{-2}.s^{-1}$.

DISCUSSION

Kaolin suspended well in water and precipitated slowly. The precipitation time of a coating substance depends on its particle size—the larger the particle size, the shorter the precipitation

Table 4 Physiological variables related to the photosynthesis of uncoated and coated mango leaves.

Treatment	Physiological variables*							
	T_{leaf} (°C)	$VPD_{leaf-air}$ (kPa)	P_n (µmol CO_2 $m^{-2}.s^{-1}$)	g_s (mmol H_2O $m^{-2}.s^{-1}$)	E (mmol H_2O $m^{-2}.s^{-1}$)	C_i (µmol CO_2 mol^{-1})	C_a (µmol CO_2 mol^{-1})	C_i:C_a
Control	41.51±0.41	6.04±0.16	2.34±1.34[b]	45.07±4.16[b]	2.81±0.32[b]	261.00±5.64[c]	382.90±0.46	0.68±0.01[c1/]
Kaolin	41.46±0.63	5.73±0.29	3.65±0.33[a]	91.97±7.75[a]	5.25±0.13[a]	281.75±2.65[b]	385.12±1.34	0.73±0.01[b]
Bentonite	41.48±0.74	6.21±0.31	0.31±0.06[c]	21.75±1.94[c]	1.40±0.13[c]	325.75±3.25[a]	385.99±2.43	0.84±0.01[a]

T_{leaf} = Leaf temperature, $VPD_{leaf-air}$ = Leaf to air vapor pressure deficit, P_n = Average net photosynthesis, g_s = Stomatal conductance, E = Transpiration rate, C_i = Intercellular CO_2 concentration, C_a = Air CO_2 concentration, C_i:C_a = Ratio of intercellular to air CO_2 concentration.

* = Mean value of 12 replications (±S.E.)

Means followed by the same lowercase superscript letter within a column are not significantly different by Duncan's multiple range test at the $P \leq 0.05$ level.

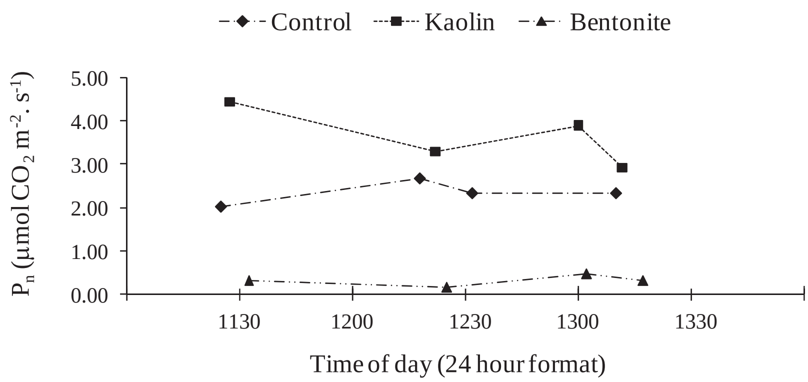

Figure 2 Net rate of photosynthesis (P_n) around midday for different leaf coatings.

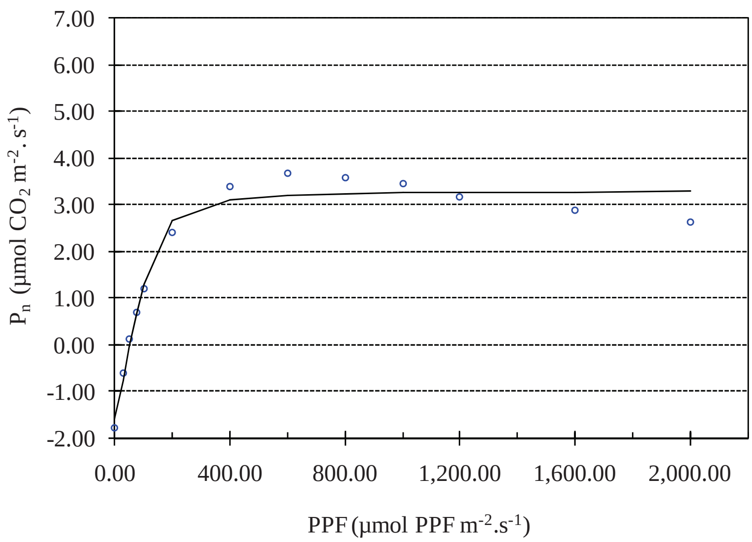

Figure 3 Net rate of photosynthesis (P_n) versus photosynthetic photon flux (PPF) light response curve of mango leaves measured in CO_2 (400 µmol.mol^{-1}) in an atmospheric chamber at leaf temperature of 37 °C.

time. In this research, the precipitation of kaolin was slowest; this was due to it having the smallest particle size.

The results showed that among the four coating materials, kaolin was the best for reducing irradiation as measured by transmission of photons though the glass plate. Kaolin could increase the reflective irradiation, therefore, its measurement of PPF by the transmission of photons through the glass plate showed the lowest PPF percentage. An explanation of the mechanism of kaolin application on a treated plastic Petri plate was reported by Jifon and Syvertsen (2003), who demonstrated that kaolin particle film reduced PAR transmittance by approximately 28%. They also found that the loss of PAR absorption was entirely due to increased PAR reflection. Glenn and Puterka (2005) reported on kaolin sprayed onto tree foliage as a liquid suspension, leaving kaolin as a white, porous, protective, powdery film on the surface of leaves and fruits after water evaporation. The physical properties of the film were a reduction of damage from insects and plant pathogens as well as enhancing photosynthesis, yield and fruit quality, particularly in hot and dry climates. Kaolin particle film has been reported to increase foliage reflectivity and reduce the heat load on plants with some increases in plant productivity (Glenn *et al.*, 2001). The leaf is able to utilize PAR through the particle film, but the film reflects ultraviolet and infrared radiation from the leaf or fruit surface (Glenn and Puterka, 2005). However, the variability in light transmittance probably approximates nonuniform particle deposition on leaves as might occur in the field (Jifon and Syvertsen, 2003; Glenn and Puterka, 2005).

The natural microclimatic conditions indicated that mango leaves were exposed to high levels of PPF, T_{air} and VPD_{air} at midday. Consequently, the T_{leaf} and $VPD_{leaf\text{-}air}$ values in mango leaves increased with these factors leading to the closing of stomata and a midday depression of photosynthesis due to limited CO_2 uptake and decreased CO_2 availability. Furthermore, mango leaves received excess radiation of 1,462.67 µmol PPF $m^{-2}.s^{-1}$ which was higher than the light saturation point measurement in this study of around 600 µmol PPF $m^{-2}.s^{-1}$ or 232.93. µmol PPF $m^{-2}.s^{-1}$ from the calculation. Moreover, the saturated P_n values which were measured from the first mature leaves in exterior canopy positions showed a P_{max} value of 3.5 µmol CO_2 $m^{-2}.s^{-1}$ for the data. However, the P_n value of untreated leaves was 2.34 µmol CO_2 $m^{-2}.s^{-1}$. The decrease in photosynthesis in uncoated leaves due to the high VPD_{air} induced stomatal closure and limited the CO_2 uptake. The carbon metabolism may limit the consumption of photosynthetic energy under high light radiation resulting in excess photon absorption. As a consequence, non-utilized excitation energy can accumulate, promoting reductions in photosynthetic efficiency, which is termed photoinhibition. Thus, this condition could exacerbate photoinhibition in non-kaolin-treated mango leaves due to the excess of photosynthesis (Xu and Shen, 2005). Goldschmidt (1999) found that the excess radiation and high temperatures of leaves caused water deficits and reduced light-use efficiency, leading to photoinhibition, reduced photosynthesis, lower growth, less fruit yield and poorer quality.

The study on physiological variables relating to photosynthesis after applying coating materials to mango leaves showed that kaolin induced the highest P_n, g_s and E values compared with the bentonite and control treatments. In addition, the C_i and C_i:C_a values for the kaolin-coated leaves increased, compared with untreated leaves. It may be assumed that kaolin could help the reflection of excess radiation from the mango leaf. The uncoated leaves accumulated heat load inside the leaves which affected stomatal closure. Bentonite had a higher particle size than kaolin which caused low attachment on the leaves resulting in its poor suitability as a coating material. Its low reflection result led to the lowest P_n value and the highest C_i value since carbon dioxide use was inhibited. The study indicated that kaolin effectively increased P_n since it was able to decrease excess irradiation. Likewise, Glenn and Puterka (2005) suggested that kaolin was reflective to UV radiation but the formulation and particle distribution significantly influenced the degree of its UV reflection. In addition, the physical presence of the clay particles apparently did not inhibit leaf gas exchange, perhaps due to the porous nature of kaolin clay. This desirable quality led to an increase of net carbon assimilation in the kaolin-treated plant. It suggested that an increase in yield and fruit quality on kaolin-treated apple trees was caused by an increase in whole-tree carbon assimilation under heat stress (Glenn *et al*., 2003).

CONCLUSION

Kaolin suspended well in water and showed the slowest rate of precipitation compared with bentonite, calcium carbonate and dolomite. In addition, kaolin was the most effective leaf coating material for reducing transmission as it had the lowest photon transmittance through a glass plate. Mango leaves sprayed with kaolin could increase net photosynthesis, stomatal conductance and the transpiration rate under high temperature and *severe drought*. Therefore, kaolin is suitable for use as a leaf coating material on mango trees grown under hot and dry conditions.

ACKNOWLEDGEMENTS

The authors would like to thank the National Research Council of Thailand and the Office of the Higher Education Commission, Thailand for providing grant funds under the program of Strategic Scholarships for Frontier Research Network for the Joint Ph.D. Program Thai Doctoral Degree for this research. Comments on the manuscript from Dr. Gaysorn Dhavises are gratefully acknowledged.

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