# **Developmental Stages and Host Range Confirmation of Crapemyrtle Bark Scale (***Acanthococcus lagerstromiae***)**

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#### Abstract

Crapemyrtle Bark Scale [(CMBS) Acanthococcus lagerstroemiae] is an exotic pest species that is causing aesthetic and economic damages to crapemyrtles and posing potential threats to other horticultural crops. Although previous studies had reported on the life history of this insect pest, much information is still unclear or missing in terms of the insect developmental process and its ability to infest alternative hosts. In this study, two different insect rearing methods were utilized to obtain a detailed documentation of each developmental stages of CMBS. Infestations of CMBS were confirmed on apple (*Malus domestica*), *Malus angustifolia*, *Chaenomeles speciosa*, *Disopyros rhombifolia*, *Heimia salicifolia* and *Lagerstroemia* 'Spiced Plum'. No sign of CMBS infestation was observed on *Rubus* 'Arapaho', *Rubus* 'Navaho', *Rubus idaeus* 'Dorman Red', *Rubus fruticosus*, *Buxus microphylla* var. *koreana x B. sempervirens*, *Buxus harlandii, and Diospyros virginiana* during a 14-week experiment period.

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## **INTRODUCTION**

Crapemyrtle Scale [(CMBS) Bark Acanthococcus lagerstroemiae] originates from East Asia and has been reported to infest important horticultural crops, such as crapemyrtles and pomegranate, in Japan, Korea, India, and China (Egolf and Andrick, 1978; Gu et al., 2014a). Since its first discovery in 2004 in Richardson, TX, the infestation of this scale insect has been confirmed in New Mexico, Oklahoma, Texas, Arkansas, Louisiana, Tennessee, Mississippi, Alabama, Georgia, North Carolina, Virginia (EDDMapS, 2019), and Washington (Wang, et al. 2016). As an exotic pest species, its wide spread is also posing threats to other potential alternative host crops, especially native plant species in the U.S. The most common host of CMBS, crapemyrtles (Lagerstroemia spp.), have been utilized as a very important landscape tree in the southern U.S., which generates around \$66 million of wholesale market value per year (Wang et al., 2016). The wide use of crapemyrtle could be partly due to its ease to propagate, grow and manage. However, infestation of CMBS can greatly affect performance of crapemyrtle, causing aesthetic and economic damages.

According to previous studies and observations, the development and biology of crapemyrtle bark scale is categorized as incomplete metamorphosis, with a life cycle of 56 to 83 days (Jiang and Xu, 1998). The number of generations of the scale ranges from two to four depending on the climate zones (Gu et al., 2014b; He et al., 2008; Jiang and Xu, 1998). In the U.S., up to four generations were observed in the field in Dallas, TX (Gu, et al., 2014b). Under greenhouse condition, the development of males involves five nymph stages, and three nymph stages were found in females (Wang, et al., 2015). However, the underlying mechanism of the insect development, including the defining differentiation and the lengths (the numbers of days within a life cycle) of each developmental stage of CMBS - remains elusive. The difficult in defining the developmental stages is largely due to the small size (ranging from 0.3 mm to 3 mm depending on the age and sex) of the nymph, which constitutes a large portion of its entire life cycle. The better understanding of CMBS life cycle is important for understanding its entire developmental process. This is crucial for investigating the factors that affect insect mortality in terms of effectively controlling this pest.

In this study, two experiments have been designed to investigate the developmental stages of CMBS, and its host ranges - in order to develop effective Integrated Pest Management (IPM) programs for controlling this pest.

#### MATERIALS AND METHODS

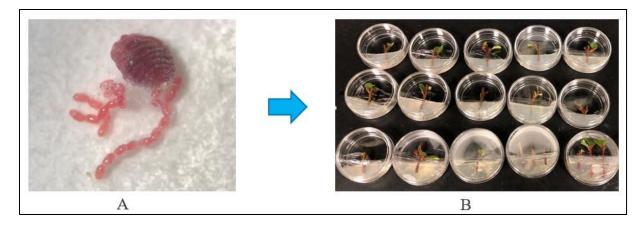
# Experiment 1: Developmental stages of crapemyrtle bark scale:

#### Insect source and plant material

Branches/twigs infested with CMBS were collected from crapemyrtle trees on campus (Texas A&M University, College Station, TX), and stored in zip-lock bags under constant temperature (25°C). The gravid females collected from infested plants were used for the experiments immediately, or within one or two days after the collection.

#### Construction of feeding chamber

Feeding chambers were constructed with small petri dishes and clear plastic food wrap. Around half of petri dish was wrapped by clear plastic to create space for medium. Water agar was poured into the bottom of the petri dish at a 70-degree angle to fill around one third portion of the petri dish. One piece of detached stem from *Lagerstroemia fauriei* 'Kiowa' (food source) was stuck in agar medium to stay turgid (Figure 1).



**Figure 1**. (A) Female CMBS producing eggs within 24 hr; (B) CMBS feeding chambers containing detached plant stems (from *L. fauriei* 'Kiowa') as food source and agar medium.

#### Collecting CMBS eggs

Gravid female was obtained by gently opening white ovisac on infested branches using a fine pin. All existing eggs were removed, and the gravid female were transferred onto a wet filter paper placed in a petri dish. All eggs laid by gravid female were collected in the next day (after 24 hours) and used as day-1 eggs in the rearing experiment (Fig. 1). Day-1 eggs were collected and kept under 25°C until hatched.

#### Insect rearing experiment

Detached crapemyrtle stems with leaves and bud nodes was used as food sources and placed in the feeding chamber as described above. One or two newly hatched (within one day) crawlers/nymphs were transferred by using a fine brush on detached stem with separate feeding chambers. Daily observation was made to record the settling status for nymphs. Feeding chambers with nymphs that failed to settle on plant were discarded when the mortality or escaping were confirmed.

#### Data collection

Egg incubation time and the number of settling (feeding) nymphs were first recorded to evaluate to mortality rate of insects at the

early nymph stages. Daily observation continued as nymphs start feeding, and the duration of each developmental stage (including nymph stages, pupa, and adult stages) were recorded. When a male reached the adult stage, it will be transferred to pair with a female for mating in order to complete the life cycle of the female. Fecundity data (the number of eggs that an adult female produces), and longevity (the number of days a female lives) were recorded as gravid females completed their life cycles. The developmental stages of both male and female CMBS were determined by the number of times the nymphs molted, which was determined by keeping track of the exuviates.

### **Experiment 2: Host Range Confirma**tion of crapemyrtle bark scale:

#### Insect source and plant material

The branches/twigs infested with CMBS were collected from crapemyrtle trees on campus (Texas A&M University, College Station, TX), and stored in zip-lock bags under constant temperature (25° C). The CMBS collected from infested plants were used immediately for experiments, or within one or two days after the collection. Fourteen plant

species and cultivars from seven genera, including Malus domestica 'Fuji', M. domestica 'Red Delicious', M. angustifolia, Rubus 'Arapaho', R. 'Navaho', R. idaeus, 'Dorman Red', R. fruticosus, Buxus harlandii, B. microphylla var. speciosa, Chaenomeles speciosa, **Diospyros** rhombifolia, D. virginiana, Heimia salicifolia, and Lagerstroemia 'Spiced Plum', planted in 1gal pots were used in the experiment.

#### Construction of plant chamber

Plant chambers [1.3 m x 1.3 m x 1.2 m  $(4.2 \text{ ft} \times 4.2 \text{ ft} \times 3.8 \text{ ft})$ ] were constructed using PVC pipes as frame and enclosed by mesh fabric sown together with a sewing machine. Zippers were sown onto mesh fabric to form an opening for transporting the plants (Fig. 2).



**Figure 2**. Plant chambers  $[1.3 \text{ m x } 1.3 \text{ m x } 1.2 \text{ m } (4.2 \text{ ft} \times 4.2 \text{ ft} \times 3.8 \text{ ft})]$  containing fourteen plant species and cultivars.

#### Host range confirmation

All plants were placed inside the plant chambers before inoculating with CMBS with one set of 14 plant species and cultivars per chamber. One infested crapemyrtle branch with 5 fresh ovisacs was attached each tested plant. To ensure successful inoculation of CMBS, plants were inoculated twice on 13 May and 15 June 2019, respectively. The plants were observed weekly from May to August 2019, and the numbers of male pupae and gravid females were recorded biweekly.

#### **RESULTS AND DISCUSSION**

# Developmental stages of crapemyrtle bark scale

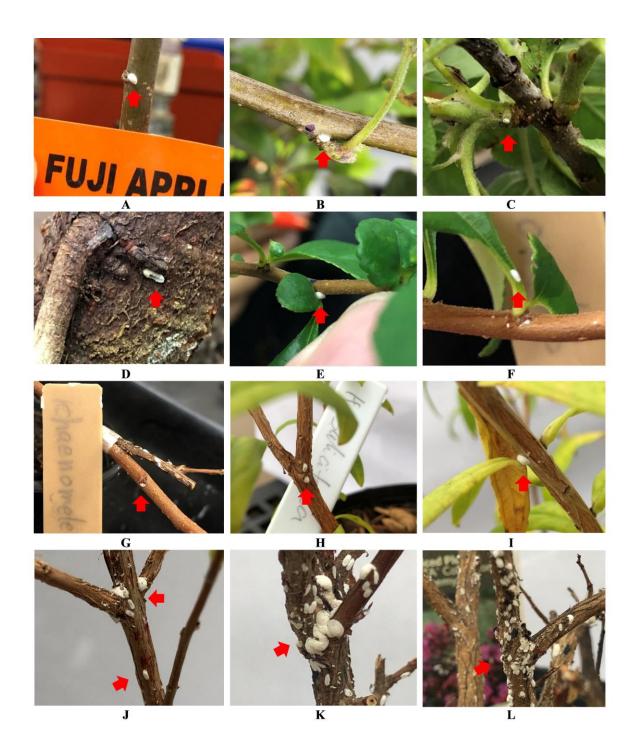
Observations performed in this study confirmed that the developmental stages of adult males consisted of an egg, two nymphal stages (1<sup>st</sup> and 2<sup>nd</sup> instar), three different stages of pupa, and the winged adult stage. The development of a female entails four major stages: egg, two nymph stages (1st and 2nd instar) and an adult stage. The egg incubation time for both males and females is a little over 12 days under 25 °C. For males, the average duration for 1st instar, 2nd instar, pupal, pupa2, pupa3, and adult period is around 14, 9, 3, 2, 4, and 4 days, respectively. For females, the average duration for 1<sup>st</sup> instar, 2<sup>nd</sup> instar, and adult period is around 13, 13, and 31 days, respectively. The males had shorter life span of around one month compared to the females, which lived much longer and produced eggs for up to 57 days. This might be due to the sole purpose of males for mating and indicating their specific role within the sexual reproduction of CMBS.

The rearing experiment was successful in supporting the development CMBS. Insects were able to successfully complete their life cycle under the experimental conditions of the study. The feeding chamber constructed for rearing CMBS allowed for direct monitoring of the entire developmental process of CMBS, which would normally be difficult to achieve under field conditions. Hence, this novel rearing method can be used to further compare the effects of different plant hosts on CMBS. Experiments can be set up to extract data for life table analysis, which is one fundamental tool in the field of entomology that builds up the knowledge foundation and provides insight of the population dynamics of arthropods of interest.

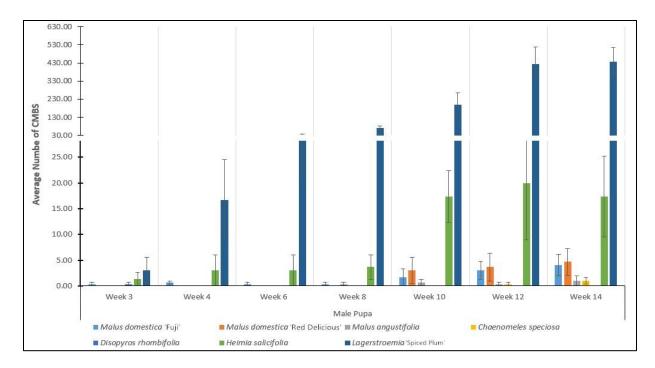
#### Host Range Confirmation of Crapemyrtle Bark Scale

During the 14 weeks (May to August 2019) of the experiment, 7 out of 14 tested plant species and cultivars (*Malus domestica* 'Fuji', *Malus domestica* 'Red Delicious', *Malus angustifolia*, *Chaenomeles speciosa*, *Diospyros rhombifolia*, *Heimia salicifolia*, and *Lagerstroemia* 'Spiced Plum') were confirmed to have different levels of CMBS infestation (Figs. 3, 4 and 5). The control plant *Lagerstroemia* 'Spiced Plum' had the highest number of male pupae and gravid females throughout the 14-week period. Population numbers were highest durng week 14, with 414.7 pupae and 100.3 gravid females (Fig. 4 and 5).

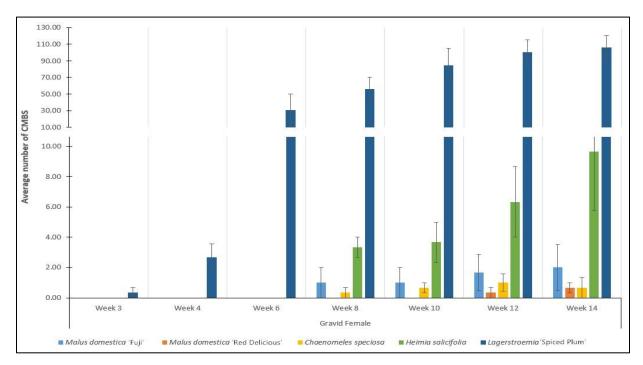
The number of CMBS pupa and gravid females were much lower on *M. angustifolia*, and *Diospyros rhombifolia*, respectively. Gravid females were observed on 'Fuji' (2 gravid females), 'Red Delicious' (0.7 gravid females), *C. speciosa* (0.7 gravid females), and *H. salicifolia* (9.7 gravid females) (Fig. 4 and 5). The lower number on other plant species, indicates that CMBS showed greater feeding preference toward *Lagerstroemia*. However, our study confirmed the ability of CMBS to infest alternative plant species, especially plants within genera of *Malus*, *Chaenomeles*, and *Heimia*.



**Figure 3**. Infestation of (A-B) *A. lagerstromiae* (red arrows) found on *Malus domestica* 'Fuji', (C), *Malus domestica* 'Red Delicious', (D), *Malus angustifolia*, (E-G), *Chaenomeles speciose*, (H-J) *Heimia salicifoliam*, and (K-L) *Lagerstroemia* 'Spiced Plum' recorded from May to August 2019.



**Figure 4**. Average number of male pupae of *A. lagerstromiae* on *Malus domestica* 'Fuji', *Malus domestica* 'Red Delicious', *Malus angustifolia, Chaenomeles speciosa, Diospyros rhombifolia, Heimia salicifoliam*, and *Lagerstroemia* 'Spiced Plum' recorded from May to August 2019.



**Figure 5.** Average number of gravid females of *A. lagerstromiae* on *Malus domestica* 'Fuji', *Malus domestica* 'Red Delicious', *Malus angustifolia, Chaenomeles speciosa, Diospyros rhombifolia, Heimia salicifoliam*, and *Lagerstroemia* 'Spiced Plum' recorded from May to August 2019.

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