A portable chamber for experimental observations of *Bactericera cockerelli* on plant seedlings and leaves

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Bactericera cockerelli Šulc (Hemiptera: Triozidae), the potato-tomato psyllid, is the pest of most concern in solanaceous crops, as a vector of *Liberibacter* (Hyphomicrobiales: Rhizobiaceae), causal agents of severe diseases. Laboratory research on biology, behavior, and physiology of *B. cockerelli* is required (Reyes-Corral et al. 2021). Experimental methods with live insects should be adaptable, simple, and repeatable. Insect bioassays often are performed on complete plants (Liu & Trumble 2005; Echegaray et al. 2016). On detached leaves (Lehman 1930; Wagan et al. 2018), observations can be done for only a few d due to leaf decay. With the clip chamber technique (insects confined in small chambers attached to leaves on whole plants) observations may be difficult because manipulation is cumbersome (Liu & Trumble 2005; Echegaray et al. 2016; Szczepaniec et al. 2019).

A portable chamber was produced to facilitate laboratory observation of processes like oviposition, hatching, and development of *B. cockerelli*, and the effect of variables like plant material (leaflets or rootless seedlings) and substrate (peat moss or water).

This chamber was based on the design of author Romero-Castillo. On Figure 1, one can see that it is composed of: two bottoms (A and B) of transparent polyethylene Petri dishes (100 mm diam × 15 mm deep); a 5 mL cryovial tube (D) with screw cap (C) (Corning PD1013, Corning, New York, USA); organza cloth; Parafilm® (Bemis, Mexico City, Mexico), silicone bars (Modatelas, Saltillo, Mexico); and non-toxic modeling clay (Baco, Mexico City, Mexico). Each bottom (A and B) has a 1.5 cm diam hole (a) for ventilation, covered with organza fabric sealed with silicone. The bottom A has a lateral hole of diameter equal to the tube cap (b). The cap (C) of this tube has a hole (c) 0.7 mm in diam. Assemblage: the cap (C) of the cryovial tube was inserted in hole (b) of bottom A, and the contact of cap and bottom sealed with clay. The tube has water or substrate added, then covered with the perforated cap.

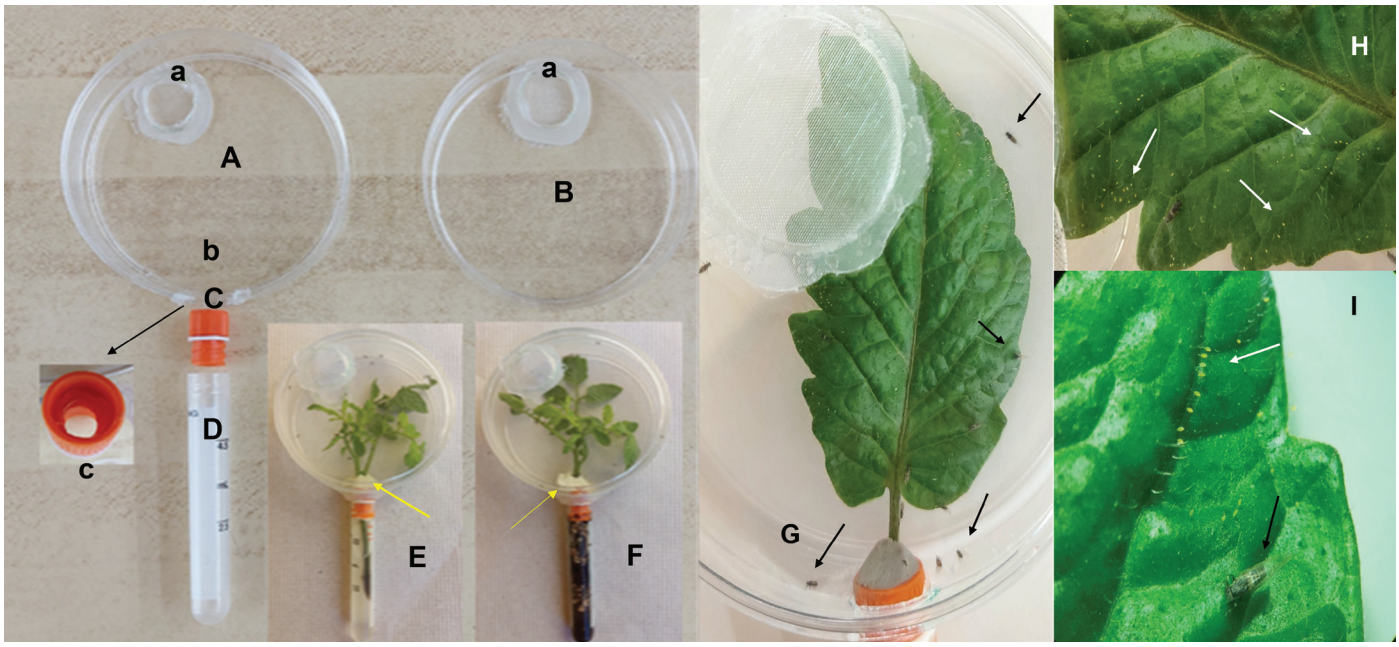

Fig. 1. Portable chamber. Bottoms A and B of chamber, with holes (a) and (b); cryovial cap (C) positioned in hole b; cap C with perforation (c); cryovial tube (D). Two chambers complete with plants, with water (E) or substrate (F) and modeling clay (yellow arrows) sealing the plant in tube. Adults (G) and eggs (H, I) (black and white arrows, respectively) of *Bactericera cockerelli*, and tomato leaflet inside chamber.

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Table 1. Mean ± SE of *Bactericera cockerelli* oviposition and egg hatching.

Treatments	Eggs per chamber Mean ± SE	Eggs per female Mean ± SE	Hatching % Mean ± SE
Leaflets in water	67.3 ± 18.2 a (13.2–170.3)	13.3 ± 3.6 a (2.6–34.2)	66.8 ± 9.2 a (46.1–94.7)
Plants in water	75.8 ± 7.4 a (38.1–111.3)	15.2 ± 1.5 a (7.6–22.2)	78.9 ± 6.1 a (30.8–95.5)
Plants in peat moss	68.4 ± 10.0 a (22.2–128.4)	13.7 ± 2.0 a (4.4–25.8)	73.28 ± 3.7 a (54.71–98.2)

*means within columns followed by the same letter are not significantly different (α = 0.05). SE = Standard error. Range of observations in parentheses.

Terminal leaflets (6–7 cm) of tomato leaves or seedlings (with cut root) were used. The petiole or stem was inserted in hole (c) of the cap, and the contact of plant and cap was sealed with clay to prevent leaks and insect escape. Bottoms A and B were placed together and the union sealed with Parafilm®, forming the chamber (Fig. 1).

There were 2 sets of observations on *B. cockerelli*. The first observations used 10 replicates (chambers) with tomato leaflets and the petiole in water. To ensure the use of live, vigorous leaflets, these were incubated in chambers for 24 h before starting observations. After this acclimatization period, adult insects (2 males and 3 females) were introduced per chamber, which was quickly assembled to prevent insect escape; the union of bottoms A and B was sealed with Parafilm®. After 48 h adults were removed, and the number of eggs, and eventually nymphs and adults, were recorded over several d. Leaflets were changed twice (14 and 21 d after experiment setup) since nymphs (about 30 per leaflet) caused rapid deterioration of plant material.

In the second set of observations, on tomato (Rio Grande variety), we compared the effect of 3 treatments on oviposition: leaflets in water, plants in water, and plants in moistened substrate (Premier® sphagnum peat moss; Premier, Quakertown, Pennsylvania, USA). Plants were first acclimatized for 24 h as before. Ten *B. cockerelli* adults (5 females and 5 males) were introduced per chamber as described with 10 replicates (chambers) per treatment. Adults were confined in the chamber for 48 h, then removed. After 5 d, we recorded the number of unhatched and hatched eggs. Data (second observations) were processed by analysis of variance and means separation (Tukey's HSD) in the statistical package InfoStat (Di Rienzo et al. 2017).

In the first observations, females oviposited a mean of 15.3 eggs per leaflet. In the second observations, means were 13.3 eggs on leaflets in water, 13.7 on plants in moistened substrate, and 15.2 on plants in water (Table 1). Twelve eggs were observed in a clutch by Lehman (1930).

In the first observations (leaflets with water), the life cycle of *B. cockerelli* took on average 20.2 d (range: 20–27 d). On potted plants in the laboratory, it took 18.7 d (range: 17–27 d) (Yang et al. 2013). In the second observations, 85.9% of eggs hatched, and 74% of eggs became adults across treatments. No significant differences were observed in oviposition (P = 0.8585) or egg hatching (P = 0.7164) between treatments (leaflets in water, seedlings in water, and plant with substrate) (Table 1).

Plants remained in good condition until the second experiment was finished. Both leaflets and plants kept growing and often developed roots in vials. This root development may be useful for experimental evaluations. Leaflets with few (near 10) nymphs remained viable for at least 3 wk; the plants (both in water and substrate), for up to 5 wk.

Kaur et al. (2020) observed *B. cockerelli* on potato leaves, in an arena composed of a plastic cup (29.5 mL) and a 1.5 mL vial; however, its shape and size make observations under the stereoscope difficult. Flat, clear Petri dishes of a small size, and easy manipulation of our chamber allow observations under the stereoscope; leaflets or plants can be used with water or solid substrates. Also it is useful for the following: *B. cockerelli* on potato leaflets, and pepper (*Capsicum annuum* L.; Solanaceae) and nightshade (*Solanum elaeagnifolium* Cav.; Solanaceae) twigs; Asian citrus psyllid (*Diaphorina citri* Kuwayama; Hemiptera: Liviidae), on orange (*Citrus* × *sinensis* Osbeck; Rutaceae) and orange jessamine (*Murraya paniculata* L. Jack; Rutaceae); and western flower thrips (*Frankliniella occidentalis* Pergande; Thysanoptera: Thripidae), and on bean (*Phaseolus vulgaris* L.; Fabaceae) leaves.

This chamber is easy to implement and replicate; it allows examination under the stereoscope of oviposition, hatching, development time, and mortality of insects. It is a simple option for observation of *B. cockerelli* and other insects. These results can be a baseline for experimental analysis of phytophagous insects and plants.

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Summary

We describe a portable, economical plastic chamber for observations on development and reproduction of potato-tomato psyllid, *Bactericera cockerelli* for up to a few wk on plant material under controlled conditions. It can be used for other insects as well.

Key Words: insect; phytophagous; laboratory; Solanaceae

Sumario

Se describe una cámara de plástico portátil y económica para observaciones durante semanas del desarrollo y reproducción de el psílido de la papa y tomate, *Bactericera cockerelli* en material vegetal bajo condiciones controladas. Esta cámara también se puede utilizar para otros insectos.

Palabras Clave: insecto; fitófago; laboratorio; Solanaceae

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