## A New Plastic Soda Bottle Cage Useful for Standardizing Biological Studies of Arthropods on Plants

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Entomologists frequently find it necessary to devise new methodologies for testing insect-plant interactions, investigating behavior, or assessing aspects of a study organism's biology (Ratnadass, et al. 2001, Vargas, et al. 1985, Hughes et al. 1966, Mackauer and Bisdee 1965, Eide and McLean 1954, Huffaker 1948, Dustan 1931). Plastic soda bottles previously have been employed for uses such as funnels (Barton 1994), pheromone lures (Smit et al. 1997, Borges et al. 1998, Oliver et al. 2004), clockworks (Stevenson et al. 2006), and aerial interception traps (Carrel 2002) and monitoring devices. Modified plastic soda bottles have been used as cages for studies on various arthropods and plants (Medal et al. 1997, Carrell et al. 2000, Glynn and Larsson 2000). Three-liter soda bottles have been utilized for feeding and oviposition preference studies (Hawthorne, et al. 1992), but have not been modified in such a manner as to enclose a potted plant. Herein, we describe construction of a rigid, plastic sleeve cage constructed from a transparent 3-liter plastic soda bottle. The bottle cage volume is 2,500 cm³ and can fit over a 10 cm diameter pot holding plants ≤ 25 cm in height.

Three-liter, clear, plastic soda bottles were cut with a razor blade at points 5 cm from the bottom and 6 cm from the top (Fig. 1A), respectively, resulting in openings of 12.5 and 8 cm in diameter. Cut ends were pressed for 15 sec on a hot plate (~65°C), covered with aluminum foil, to smooth uneven and jagged edges. A hot glue gun was used at medium-high temperature to glue a 15-cm diameter disk of mesh screen (nylon organdy) to the bottom 12.5-cm diameter opening of the bottle. To create an opening in the side of the bottle, a 10-mm (size 5) metal cork borer, heated over a gas flame, was used to puncture the side of the cage at the desired height (Fig. 1A). The resulting hole had a final diameter of 12 mm and served as ingress and egress for study organisms. A 3-cm piece of 12-mm diameter braided dental cotton was used to plug the cored hole. Supplemental provisions may be offered by dipping the inserted end of the cotton bung into liquid food (e.g., honeywater). Additional holes of any size can be cored for organism access or covered with mesh for increased ventilation.

The bottle cage design allows for placement over a plant contained in a 10-cm diameter pot (Fig. 1B). A larger pot size may be used if the plant size is compatible with cage height. To accommodate taller plants, simply attach an additional bottle cage (without heat or mesh applied to the 12.5-cm diameter opening) as an extension to the bottom of a completed bottle cage. The extension will fit snugly and can be secured with transparent adhesive tape. A bottle cage cut with an 8.5-cm diameter opening, instead of an 8 cm diameter opening mentioned above, may be threaded onto the top of a wide-mouth mason canning jar to provide an experimental

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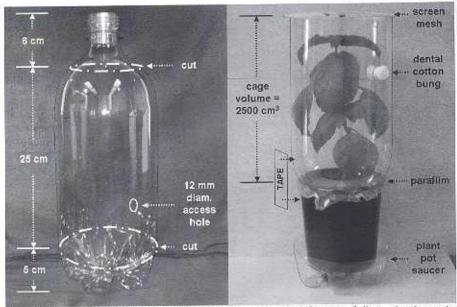


Fig 1A-B. (A) Diagrammatic bottle cage constructed from a 3-liter plastic soda bottle. (B) Completed bottle cage containing a sweet basil plant and *Homalodisca vitripennis* (Germar) (Hemiptera: Cicadellidae).

chamber for studies of arthropods on aquatic and semi-aquatic plants or stored food products. Stretched parafilm can be wrapped around the top of the pot at the base of the plant to cover the soil and help create a seal between the cage and the pot. The parafilm barrier prevents excessive condensation on the interior walls of the cage, maintains arthropods on the plant, and eliminates the possibility of "losing" study organisms in moist potting soil. The bottle cage can be secured to the pot using transparent adhesive tape. The removed bottom 5-cm portion of the bottle can be used as a plant pot saucer to catch excess water after watering caged plants (Fig. 1B). Soda bottles become misshapen under direct sunlight or over-exposure to heat in excess of 65°C (R. Wolfe, Ball Corp., pers. comm.) but are robust enough to be washed multiple times with a soft sponge and a mild detergent solution.

The bottle cage described here is useful in rearing and maintaining arthropods for long durations, such as Isofemale lines utilized in genetic studies. The bottle cage has superior visual clarity, is inexpensive to assemble, is ideal for studies requiring use of a potted plant, and significantly reduces demand for laboratory space. In addition, this cage serves as a standardized and compact experimental unit which can facilitate an increased number of replicates for rigorous experimental analysis. These cages have been utilized successfully for host specificity tests and non-target impact studies between cicadellid leafhoppers and mymarid parasitoids, day-degree studies in temperature cabinets, plant pathogen transmission studies, behavioral observations, and acoustic communication studies (EAB and MSH unpub. data). Table 1 provides descriptive statistics of arthropods and plants utilized to date in experiments involving the bottle cage.

TABLE 1. Arthropod and Plant Families Housed Successfully in Experiments

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	Densitya	Duration <sup>b</sup> (d)	Life / Growth Stages <sup>c</sup>
Arthropod Family <sup>de</sup>	-37-20		
Aleyrodidae	50	45	Egg - Adult
Braconidae	9	45	Egg – Adult
Cicadellidae	20	230	Egg – Adult
Cicadidae	1	4	Adult
Coccidea	30	60	Egg - Adult
Encyrtidae	50	30	Egg – Adult
Mymaridae	40	30	Egg – Adult
Pyralidae	50	45	Larva – Adult
Plant Family			
Chenopodiaceae	1	60	Seedling (8-10 rosette leaves)
Fabaceae	1	60	All
Lamiaceae	4	75	All
Poaceae	5	60	All
Rosaceae	120	45	Nuts with hulls <sup>9</sup>
Rutaceae	1	60	1 yr old grafted seedling
Simmondsiaceae	2	60	Seedling
Vitaceae	1	60	Mature plant ≤ 30 cm

Maximum number of arthropods or plants housed in a single cage.

Maximum length of time (d) that anthropods and plants were housed in a single bottle cage.

"Hymenoptera fed via honey-water saturated dental cotton bung shown in Fig. 18.

\*Contained in a wide-mouth mason canning jar.

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<sup>&</sup>quot;Life stages of arthropods and growth stages of plants successfully housed in a single botile cage."

"Bernisia spp. (Hemiptera: Aleyrodidae), Bracon fiebefor Say (Hymenoptera: Braconidae), Dracoulacophele minerva Ball, Graphocephala atropunctata (Signoret), Homelodisca vitripennis (Germar), H. lifurata Ball, Neoaliturus teneflus (Baker) (Hemiptera: Cicadellidae), Diceroprocta apache Davis (Hemiptera: Cicadelae), Coccus hespendum L. (Hemiptera: Coccidae), Metaphycus spp. (Hymenoptera: Encyrtidae), Polynema sp. Haliday, Gonatocerus ashmeadi Girault, G. fascietus Girault (Hymenoptera: Mymaridae), and Amyelois transitella (Walker) (Lepidoptera: Pyralidae).

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