

## Use of Artificial Egg Laying Substrate to Detect California Tiger Salamanders (*Ambystoma californiense*)

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Many techniques have been used when surveying for the presence of California tiger salamanders (*Ambystoma californiense*) and other amphibians. Night spot-lighting, dip nets, seines, cast nets, drift fences, pit-fall traps, cover boards, and visual and auditory observations have all been used to determine amphibian presence (Bean 1999; Brode 1997; Corben and Fellers 2001; Fellers and Freel 1995; Heyer et al. 1994; Langton 1989; pers. obs.). In 1997 a protocol was developed by the California Department of Fish and Game to survey for the California tiger salamander (Brode 1997). The protocol recommended using land transects for adults, and aquatic larval surveys using dip nets and seines. In 1999 an addition was made to the protocol, which included the use of minnow traps to detect larvae (Bean 1999). The minnow traps facilitated the detection of larvae in deep or heavily vegetated ponds and wetlands.

Although many amphibians demonstrate species-specific preferences with respect to oviposition sites (Beebee 1997), *A. californiense* may deposit its eggs on stones, twigs, submergent and emergent vegetation, and other debris on the bottom of ponds or wetlands (Stebbins 2003). This behavior offers the opportunity to supply artificial oviposition sites that can then be used to detect the presence of this salamander. Recommended here is a technique that allows for detection of *A. californiense* without risk of mortality to salamanders and other amphibian larvae, and with a great reduction in time expended.

A grid was constructed from 13-mm (1/2 inch) polyvinyl chloride (PVC) tubing and 3-mm (1/8 inch) nylon cord. Tubing was cut into two 1-m lengths and two 0.5-m lengths and joined together in a rectangular shape with PVC glue and fittings (Fig. 1.). Around the perimeter of the rectangle, 6-mm (1/4 inch) holes were drilled at intervals of 10 cm. The nylon cord was drawn through the holes, woven over and under one another, and tied at the ends to keep the cord in place. A 45-cm (18 inch) length of foam pipe insulation (used by plumbers to prevent 1/2 inch pipe from freezing) was glued around one of the shorter lengths of the grid tubing.

Egg grids were placed in perennial and ephemeral ponds prior to and during the breeding period for *A. californiense*. Typically the grid was placed within 2 m of the shoreline and secured to a small wooden stake with a length of nylon cord. One end of the grid floated while the other end sank toward the bottom (Fig. 2.). Grids were

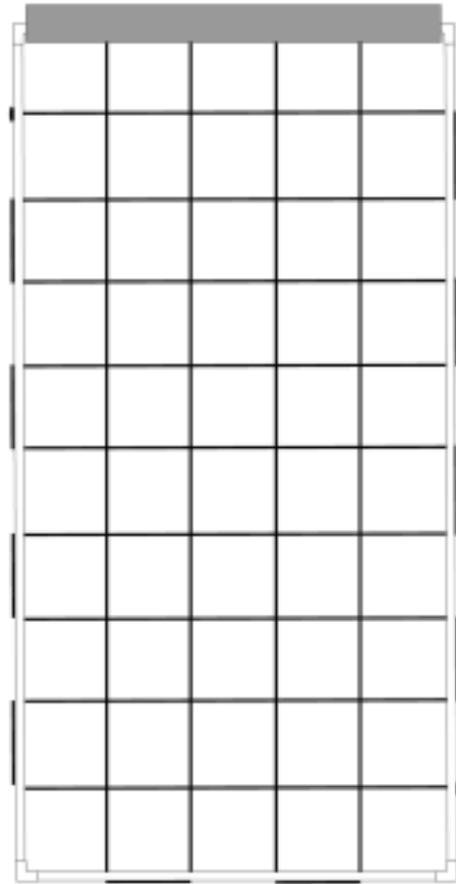


FIG. 1. Rectangular PVC egg grid with nylon cord alternately woven as shown. Foam insulation attached to the upper portion provides flotation.

checked every 7–10 days and eggs were identified and counted.

At the Los Vaqueros Watershed, Contra Costa County, California, visual surveys, minnow traps, seines, and dip nets were used to determine the presence of *A. californiense* in (up to) 90 perennial and ephemeral ponds. Surveys for the presence of *A. californiense* were made during six different years between 1989 and 1999. Within those years, the number of ponds that were reported to have *A. californiense* ranged from 2 to 30 (average = 13.8 ponds). During the winter of 2000–2001, the same ponds were surveyed using egg grids and the presence of *A. californiense*

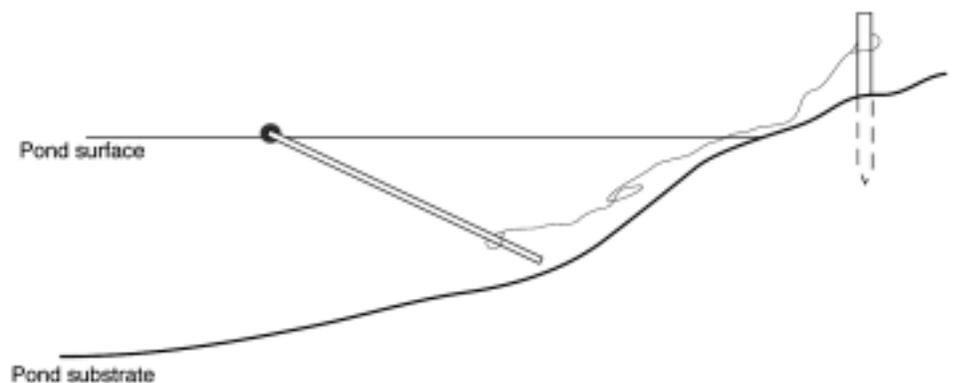


FIG. 2. Placement of the egg grid within a pond with an attachment to a stake.

**Proximate Cues for Ovarian Recrudescence and Ovulation in the Brown Treesnake (*Boiga irregularis*) Under Laboratory Conditions**TOM MATHIES\*<sup>†,1</sup>  
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was observed in 69 of 90 perennial and ephemeral ponds. Eggs were present on grids from mid-November through late-February. Presence of eggs on the egg grid was the sole or primary source of identification in 49 of the 69 ponds. Egg identification confirmation was made by the examination of larvae later in the season.

A note of caution: considerable care should be taken when examining habitat within the range of the introduced tiger salamander (*A. tigrinum*; Bean 1999). Under these circumstances neither eggs nor larvae should be used to identify the species of salamander. Genetic testing should be used to discriminate between these species.

The addition of this technique to the survey protocol may increase detection of *A. californiense*, particularly in turbid water bodies. Additionally, it can reduce or eliminate the need for Federal 10(a)(1)(A) permit for take of California red-legged frog (*Rana aurora draytonii*) larvae that are sympatric with *A. californiense*. Further, many ponds can be surveyed simultaneously with relatively little time expended and a marked reduction in potential mortality.

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The reproductive biology of the brown treesnake (*Boiga irregularis*), an invasive tropical species known primarily for the extensive ecological damage it has caused on the island of Guam (Savage 1987), is poorly understood. This is not for lack of sampling effort by researchers—it is simply because reproduction by both sexes apparently occurs in all months of the year (Rodda et al. 1999) and periodic sampling in such systems cannot detect the phenology of major reproductive events (e.g., ovarian recrudescence, ovulation, oviposition) and other important life history attributes (e.g., frequency of reproduction). In such systems, repeated observations on individuals may be the only recourse for obtaining such information. Accordingly, we recently established a captive colony of brown treesnakes at our facilities and were able to successfully induce reproduction in a number of females (Mathies and Miller 2003). Here, in our second effort to induce reproduction in this colony, we report on the phenology of ovarian recrudescence, the conditions that initiate this process, and the possibility that copulation may be necessary to induce ovulation.

Details on origin, husbandry, and procedures for mating of the snakes in this study are the same, or similar, to those given in Mathies and Miller (2003). Snakes in our colony (10 males, 15 females) were collected as adults on Guam and are the same individuals used in a previous breeding study (Mathies and Miller 2003). Because of space limitations, snakes were housed in two adjacent rooms. Temperatures in the rooms were thermostatically controlled and no other heat sources were available to snakes. Differences in air temperatures between the two rooms were unintended and due to equipment malfunction (Fig. 1). Overall mean temperatures during the study period, however, were similar (Room A: 22.8°C; Room B: 22.9°C). Relative humidity in both rooms was maintained at about 80%. Room lighting was provided by fluorescent bulbs and the photoperiod was 12L:12D. The time of year this study was conducted was chosen out of convenience. Dates provided herein should not necessarily be taken to imply that the brown treesnake is more likely to become reproductive at