RESEARCH NOTE

A Note on Slide-mounting Technique of Unfed Immature Stages of *Amblyomma cajennense* (Fabricius, 1787) (Acari: Ixodidae)

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Several tick slide-mounting techniques have been used for many years with varying degrees of success. The classic Canada balsam technique employed by entomologists and acarologists over many decades is efficient but time consuming (RL Palma 1978 *New Zeal Entomol 6*: 169-170). The classic methods used in Acarology include the water soluble media such as Berlese, Faure and Hoyer (GM Krantz 1978 *A Manual of Acarology*, Oregon State University Book Store, 2nd ed., Oregon, 509pp., A Fain 1980 *Inter J Acarol 6*: 169-170). Water soluble media are inadequate in the humid tropic, even when the slides are ringed with a water-proofing material such as Glyptal. Crystallization of the media, probably due to absorption of water, is the main problem with slide-mounting specimens.

Basic procedures in slide-mounting techniques should involve death and fixing, clarifying and mounting of the arthropods in solutions that will produce minimal alterations of cuticle and its appendages. At some point of the procedure stains might be added to enhance visibility.

The removal of gut contents is the main problem in ticks (even in unfed specimens). To do so an aqueous solution of potassium hydroxide has been used in different concentrations (10 to 20%). However, frequently the gut contents are not totally clarified and setae may break off.

During the past four years we have mounted more than 100 unfed larvae and nymphs of *Amblyomma cajennense* (Fabricius, 1787) for morphological studies using the technique for copepods mentioned in WA Boerger and VE Thatcher (1990 *Syst Parasitol 17:* 133-141), with minor modifications as follows:

1. Unfed larvae and nymphs were obtained under laboratory conditions (25-29°C, 80 ± 10% RH) and kept alive until hardening of the cuticle (15 days for larvae and 24 hr for nymphs). The ticks were killed in either hot water or 70% ethanol (approximately 70°C) and, then preserved in 70% ethanol.

2. Fixed specimens were subsequently transported to lactic acid for 1 hr at 50°C.

3. Specimens were further transferred to an alcoholic solution saturated with phenol. The specimens should be left in phenol solution (approximately 24 hr). After clarification had been finished, specimens were washed in 100% ethanol.

4. Washed specimens were transferred to a solution of pure creosote for approximately 24 hr and then mounted in Canada balsam medium.

Remarks: larvae and nymphs are still in good condition after four years from mounting as shown in Figs 1-3. Three larvae and four nymphs were deposited in Acarological Collection of Instituto Oswaldo Cruz, Rio de Janeiro, Brazil (no. 039).

The time that specimens were left in lactic acid (2) and phenol (4) may be shortened or lengthened in terms of size, gut contents and pigmentation. The phenol dehydrates and stains the specimens. Washing in 100% ethanol avoid later crystallization of phenol.

The technique described in the present article is an attempt to solve the problem of mounting immature *Amblyomma* ticks with water soluble media in the high humid tropics.

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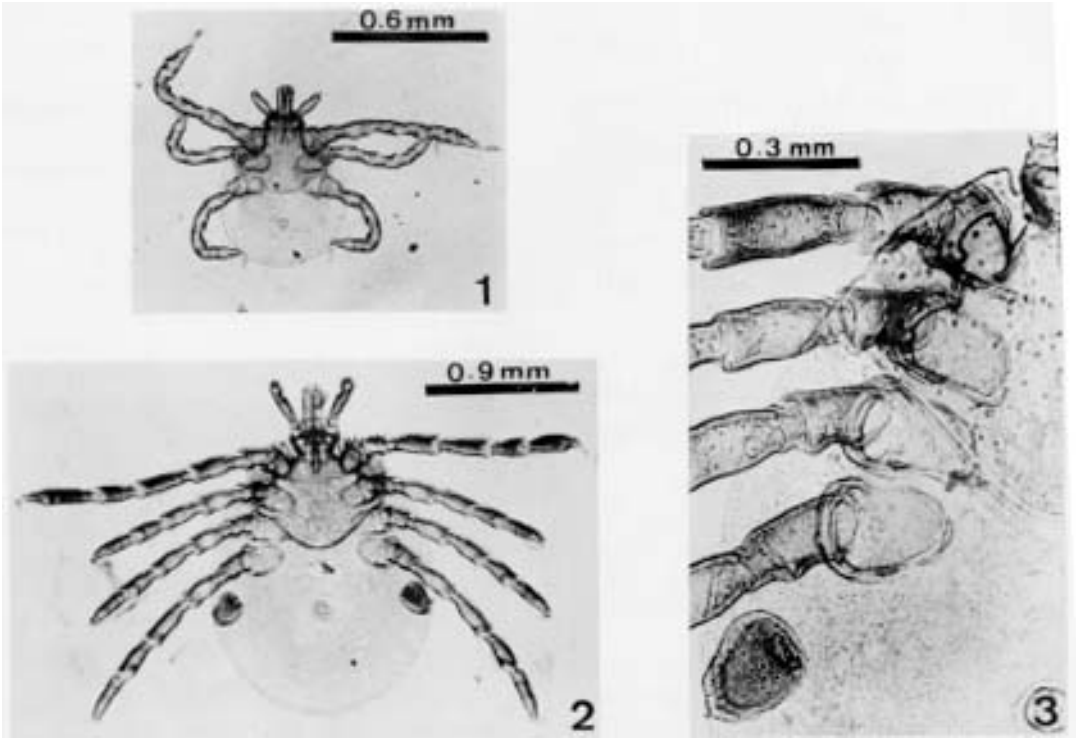

Unfed specimens of *Amblyomma cajennense* after four years from mounting using an adaptation of technique applied formerly to copepods. Fig. 1: larva, Fig. 2: nymph, Fig. 3: detail of nymph's appendages.