HORSE-BAITED INSECT TRAP AND MOBILE INSECT SORTING TABLE USED IN A DISEASE VECTOR IDENTIFICATION STUDY

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ABSTRACT. A horse-baited trap and a mobile insect sorting table were used to conduct an arthropod survey for potential vectors of Potomac Horse Fever in southern Maryland and northern Virginia. The trap and table worked effectively for the live collection and sorting of haemophagous Diptera such as: *Simulium* spp., *Stomoxys calcitrans, Musca autumnalis, Tabanus* spp. and *Chrysops* spp. during the diurnal collections periods, and *Culicoides* spp. during the crepuscular periods. The trap was not as convenient for live collection of mosquito species during the nocturnal period. Modifications to the trap design was suggested for mosquito live trapping.

INTRODUCTION

Potomac Horse Fever, a disease of horses was first identified in 1979 along the Potomac River in Maryland and Virginia (Knowles et al. 1984). The etiological agent of Potomac Horse Fever is a rickettsia, *Ehrlichia risticii* (Holland et al. 1985) and is believed to be transmitted by an arthropod, since other organisms of the genus *Ehrlichia* have had a history of tick transmission (Scott 1978) and the epidemiology of this disease is consistent with that of other arthropod transmitted diseases (Knowles et al. 1984, Perry et al., 1984, 1985, 1986; Whitlock et al. 1985).

In 1984, we began a survey of the arthropods attacking horses to collect, sort, and identify living specimens of certain potential vectors of Potomac Horse Fever in southern Maryland and northern Virginia along the Potomac River basin. The purpose of the survey was to determine the identity and seasonal occurrence of arthropods attacking horses in the endemic area.

Published information on the collection of Diptera from horses is limited. Jones et al. (1977) collected mosquitoes directly from tethered ponies and donkeys in the southwestern U.S.A. as part of an equine encephalitis survey. This direct method has been shown by others to yield a lower proportion of some species of haemophagous Diptera (Bennett 1960, Zimmerman and Turner 1983). This method is also difficult to quantify, being very sensitive to techniques by different collectors and bait animal behavior. Other methods reported by Bram (1978), such as stable traps and elevated drop traps also yield low numbers for certain species of haemophagous Diptera and are not practical when working with full size horses. We therefore designed a trap to accommodate a full size horse as bait that would expose the animal to many different species of haemophagous Diptera in as natural a situation as possible. This trap was not based upon, but uses the same principles as a trap described by McCreadie et al. (1984). This paper will deal with the description of the horse-baited (H-B) trap and a mobile insect sorting table.

In vector identification studies, it is important that samples be identified as quickly as possible. If the samples are to be used for pathogen isolation studies, the insects must be stored in dry ice (solid CO_2) or liquid nitrogen while they are still alive or within a few minutes after death. We designed an insect sorting table that would mount inside a passenger van. This mobile table permitted immediate identification and processing of the Diptera samples.

MATERIALS AND METHODS

Design and operation of H-B trap: This trap was designed so that when open (Fig. 1) a horse was exposed as naturally as possible to haemophagous Diptera, and when closed (Fig. 2) would capture all the flying insects on or around the animal at that time. The dimensions were: length 3.05 m (10 ft.), width 1.22 m (4 ft.), height 2.29 m (7.5 ft.). The trap was of 5 × 5 cm (2 × 2 in.) wood frame construction with 5 × 30 cm (2 × 12 in.) boards attached by the edges around the bottom to serve as a base. This also provided strength and support. The top and lower 1.98 m (6.5 ft.) of the sides and ends were covered with 40 mesh Saran® screening. The upper 30 cm (1 ft.) of the sides were covered with clear 4 mil polyethylene plastic (Fig. 3) and contained 3 collection holes, 20 × 15 cm (8 × 6 in.) spaced equally on each side. Each hole was lined with 46 cm (18 in.) sleeves of orthopedic stockinette to prevent accidental escape of insects. The top of the trap was constructed into a shallow "V" shape which caused the insects to concentrate along the edge of the trap, usually toward the prevailing light, thereby easing their removal. The seam between the two sides of the trap was sealed with 1.9 cm (¾ in.) foam weather strip-

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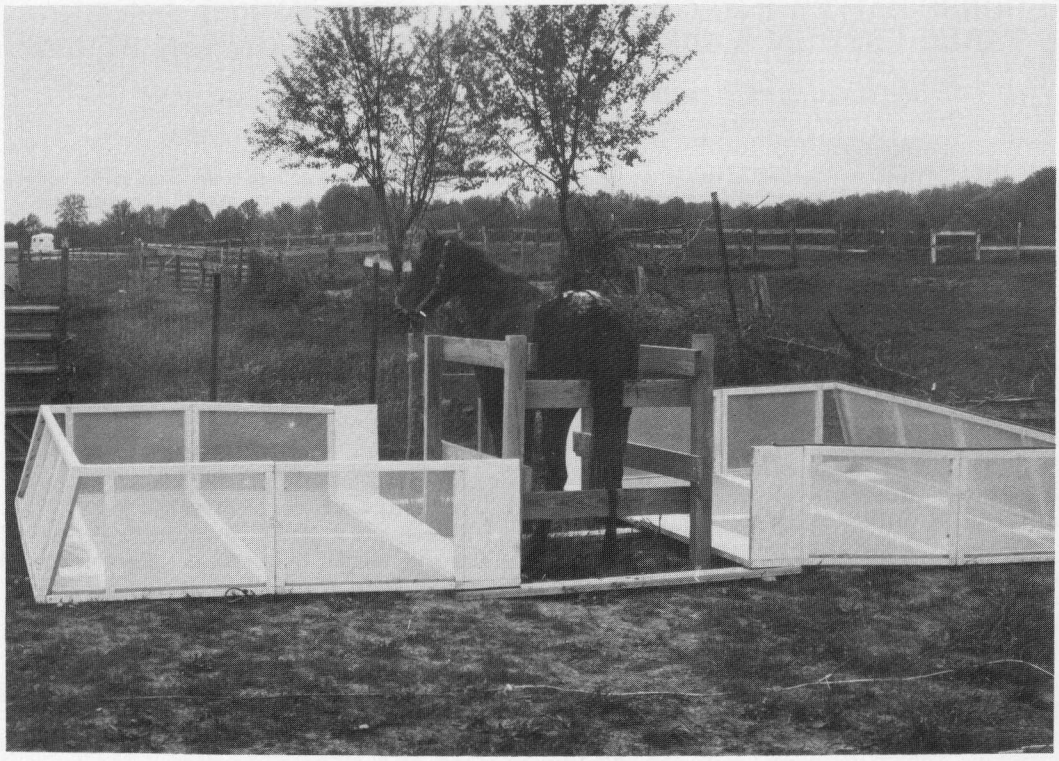

Fig. 1. Animal baited enclosure trap open showing stall which confined the horse.

ping. The 2 sides of the trap were held together tightly by elastic straps.

The trapped insects were collected by using a 12 volt hand-held vacuum aspirator (modified Black and Decker Car-Vac, Raleigh, NC 27625), similar to that described by Meek et al. (1985). The collector inserted the aspirator through one of the 3 collection holes. A flashlight was used to locate the specimens in the trap during the nocturnal collection periods.

To prevent the horse from damaging the trap, a permanent restraining stall was constructed at each collection site (Fig. 1). This stall consisted of 4 treated 10 × 10 cm (4 × 4 in.) posts planted firmly in the ground and connected at the sides and the ends by 2 horizontal 5 × 15.24 cm (2 × 6 in.) boards.

The trap was constructed in sections for transportation. The upper half was divided into 2 sections and the lower half in four "L" shaped sections (Fig. 2). To assemble the trap, three 5 × 20 cm (2 × 8 in.) boards 1.83 m (6 ft.) long were laid on the ground. The 4 bottom sections of the trap were assembled over the three 5 × 20 cm (2 × 8 in.) boards, then 2 pieces on the same side of the trap were locked together with two 0.64 × 10 cm (¼ × 4 in.) carriage bolts. The 2 top sections were put into place and each section was locked to the sections below it with eight 0.64 × 10 cm (¼ × 4 in.) carriage bolts. The 5 × 30 cm (2 × 12 in.) base of the trap was attached to the three 5 × 20 cm (2 × 8 in.) boards with three 10 cm (4 in.) door hinges on each side. Then the 2 sides of the trap could be let down to open it and expose the bait horse. When the trap was disassembled, the process was reversed except that the hinges were left in place, merely removing the pins that held the hinges together. Assembly and disassembly of the trap was made easier by using wing nuts on the carriage bolts. This trap, not including the restraining stall, could be assembled or disassembled in about 30 minutes making transportation from one pre-assembled restraining stall to another possible. When the trap was disassembled, the lower "L" shaped section was stacked inside the 2 upper sections. The whole trap and the materials to construct the restraining stall will fit in the bed of a standard full-size pickup truck.

The horses used in the traps were supplied by the cooperators of the individual farms. Because large and medium size horses in the Potomac Horse Fever area were available, they were chosen over small ponies as bait animals. Horses were selected by size (400–550 kg), color (dark) and disposition (calm). None of the horses used were disturbed by the opening and closing of the trap.

The traps were operated on one of 3 farms

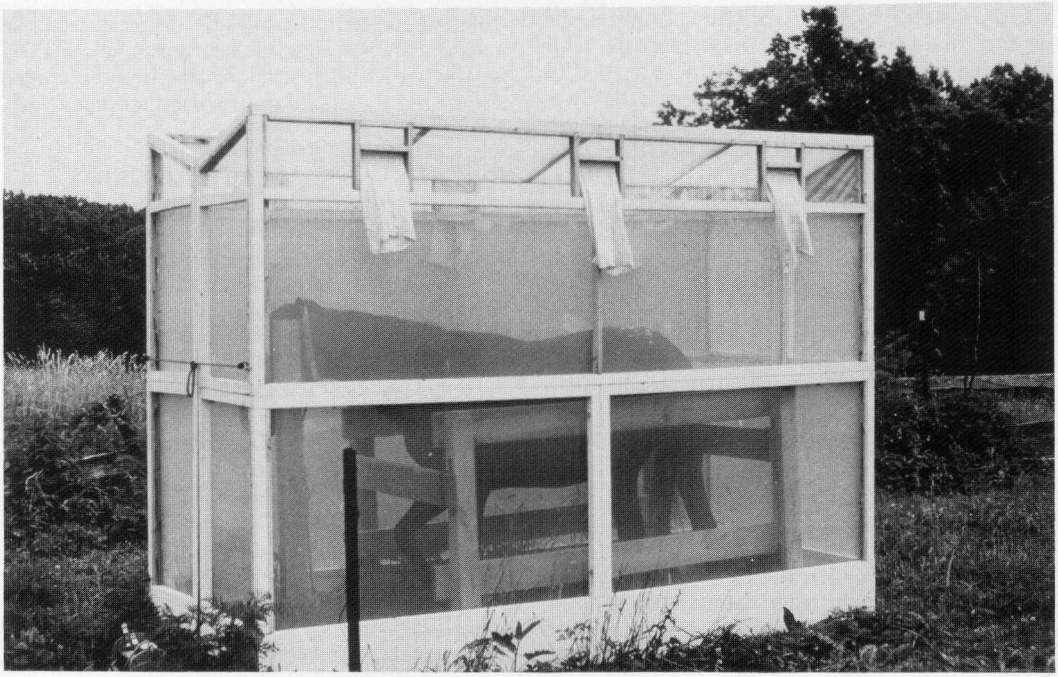

Fig. 2. Animal baited enclosure trap closed to trap haemophagous Diptera.

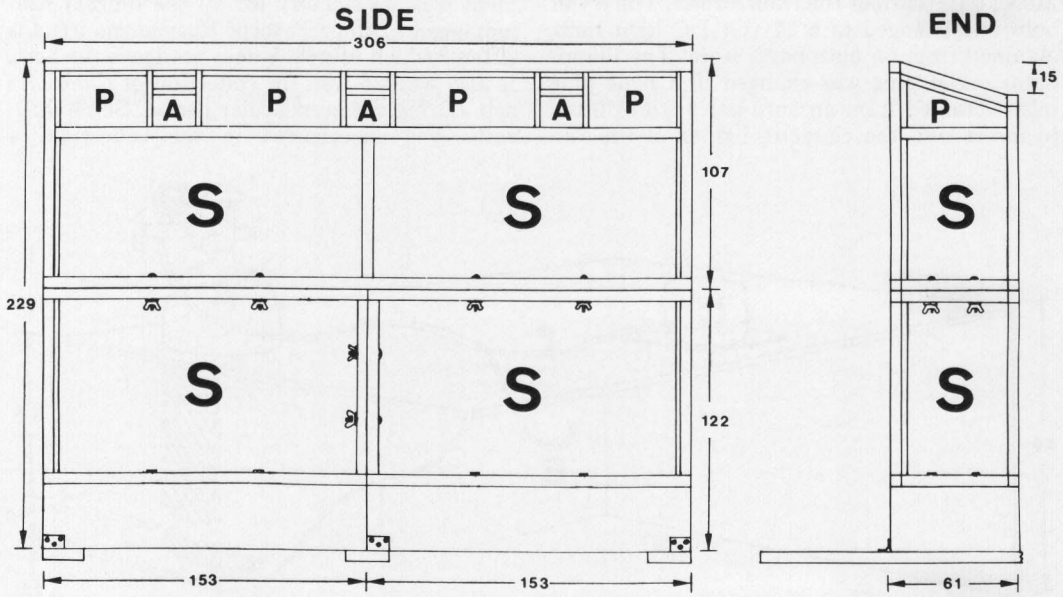

Fig. 3. Diagram of the side and left half of the end of the horse-baited trap used for the collection of haemophagous Diptera. (P) = clear 4 mil polyethylene plastic, (A) = arm holes lined with orthopedic stockinette and (S) = 40 mesh Saran screening.

each week on a rotational basis throughout the observation period during 2 collection periods: (1) diurnal (1300–1500 hr) and (2) crepuscular-nocturnal (45 minutes before sunset-sunset-45 minutes after sunset). For each collection period, the trap was opened and closed 3 times. The trap was opened, exposing the horse for 15 minutes, then closed for ca. 30 minutes. The trap was undisturbed for the first 15 minutes after closing. Following the waiting period, all Diptera were removed through arm holes at the top of the trap using the mechanical aspirator.

Design and operation of sorting table: Following collection, the Diptera were sorted and iden-

tified primarily to family or genus under a dissecting microscope on an insect sorting table mounted inside an 8-passenger van (Fig. 4). The first bench seat behind the driver's seat was removed and the table, constructed of 0.95 cm (⅜ in.) plywood, was fastened to the floor using screw hooks fitted into the same brackets that were used to secure the seat. The second bench seat served as a seat for the table making it possible for 2 people to work at the same time. The dimensions of the table were: height 71 cm (28 in.), length 91.4 cm (36 in.) and width 66 cm (26 in.).

A 22.7 kg (50 lb.) CO_2 cylinder was placed in a circular hole in the top right hand corner of the table. The tubing from the CO_2 tank was divided using a Y-tube, one side of which enabled us to direct CO_2 into the aspirator chamber to anesthetize the insects. They were then poured into a sorting tray. The other side of the tubing was connected to the bottom of this sorting tray into which the CO_2 was allowed to seep slowly over the specimens, thus keeping them inactive while they were being examined under the dissecting microscope.

The light for the microscope was a standard microscope illuminator (American Optical Corp, Model 651) without the transformer. The 6 volt bulb was changed to a 12 volt DC light bulb, obtained from an auto parts store. The illuminator outlet plug was changed to a male plug (also obtained from an auto parts store) fitted to insert into the cigarette lighter of the van which operated on 12 volts DC. Once sorted, the insects were placed in vials either in 70% ETOH for further identification, or into liquid nitrogen and stored for organism isolation studies.

RESULTS AND DISCUSSION

In 46 trapping periods, 7,613 haemophagous insects were collected representing 9 genera and 16 species. The most common genus collected was *Simulium* with a mean/45 minute exposure of 123.1 specimens. *Simulium jenningsi* Malloch, and *Simulium vittatum* Zetterstedt were the only species of the genus identified (62.1 and 52.8, mean/45 min exposure, respectively). We collected 13.2 *Culicoides* (mean/45 min exposure) representing 8 species, of which the most abundant were *C. bigutattus* (Coquillett) and *C. obsoletus* (Meigen) (9.7 and 2.7, mean/45 min. exposure respectively). Other Diptera collected and their numbers (mean/45 min exposure) were: *Musca autumnalis* De Geer (8.6); *Stomoxys calcitrans* (Linn.) (6.8); *Anopheles* spp. (0.8); *Culex* spp. (0.4); and *Aedes* spp. (0.3); *Tabanus* spp. (0.6) and *Chrysops* spp. (0.02).

The trapping, sorting procedures and equipment was satisfactory for all the diurnal haemophagous Diptera, except *Haematobia irritans* (Linn.) which normally does not leave the host. It also worked well for collection of *Culicoides* spp. during the crepuscular period. Some difficulty was experienced in the collection of

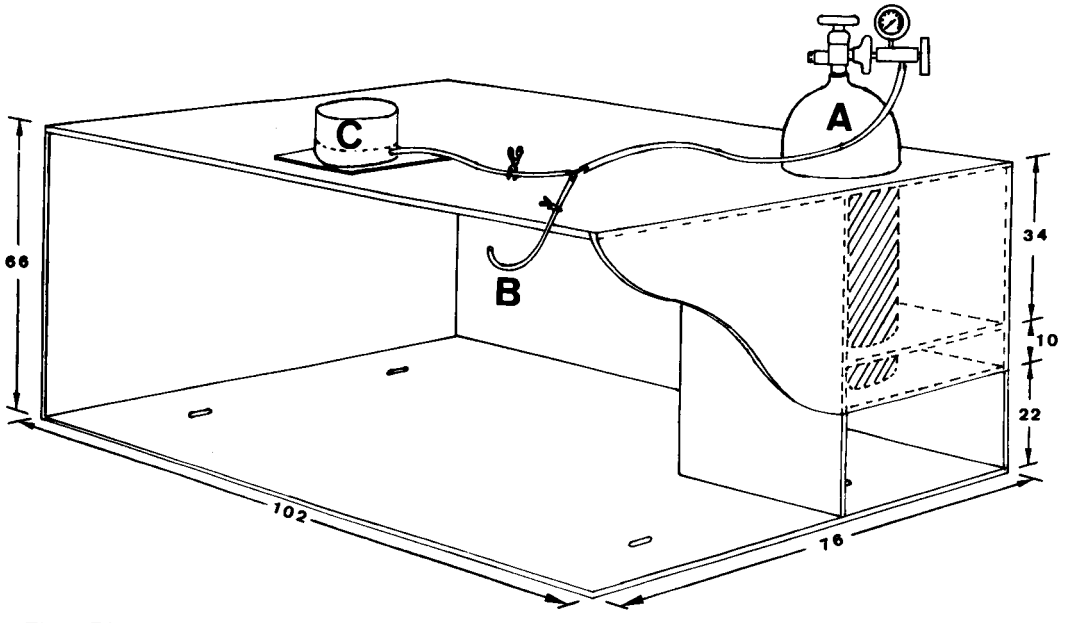

Fig. 4. Diagram of the table mounted in the van for field sorting and classification of haemophagus Diptera. (A) = CO_2 cylinder with regulator, (B) = plastic tube to anesthetize the Diptera in the collection chamber, (C) = CO_2 supplied sorting tray.

trapped nocturnal mosquito species because of a difference we observed in their post-feeding behavior. As previously described, most Diptera would fly to the top of the trap and congregate toward the prevailing light. However, the engorged mosquitoes would simply fly to the side walls and remain there, not moving up. We suggest that adding additional collection holes along the middle of the trap and placing observation ports on the ends of the trap would facilitate the collection of mosquitoes.

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