

THE USE OF CARBON DIOXIDE-BAITED TICK TRAPS FOR SAMPLING *IXODES DAMMINI* (ACARI : IXODIDAE)

BY RICHARD C. FALCO^{1, 2} and DURLAND FISH^{2, 3}

IMMATURE
IXODES
DAMMINI
CAPTURES
CARBON
DIOXIDE
LYME
DISEASE

ABSTRACT : Carbon dioxide-baited tick traps were used to collect larval *Ixodes dammini* Spielman, Clifford, Piesman, and Corwin in the summer of 1983 at 5 sites in Westchester County, New York. Larval collections ranged from 0 at one site to 712 at another during 1 week sampling periods between 26 July and 26 August. There was no significant difference between morning and evening collections, indicating that larvae are constantly active. No correlation was found between the number of larvae on tick traps and the number of larvae found on white-footed mice, *Peromyscus leucopus*. CO₂-baited tick traps are effective in sampling immature *I. dammini* and may be useful in determining human risk for Lyme disease.

CAPTURES
D'IMMATURES
D'IXODES
DAMMINI
DIOXYDE
DE CARBONE
MALADIE
DE LYME

RÉSUMÉ : Des trappes à tiques, excitées par le gaz carbonique, ont été utilisées pour récolter des larves d'*Ixodes dammini* Spielman, Clifford, Piesman, et Corwin, pendant l'été de 1983, dans 5 localités de Westchester County, New York. Le nombre de spécimens dans ces récoltes a varia de 0 pour une localité jusqu'à 712 pour une autre localité, pendant des périodes hebdomadaires, du 26 juillet au 26 août. Il n'y avait pas de différence significative entre les récoltes du matin et celles du soir, montrant que les larves sont actives continuellement. Aucune corrélation n'a été trouvée entre le nombre de larves dans les trappes et sur le mulot, *Peromyscus leucopus*. Les trappes à excitation par gaz carbonique sont efficaces pour la récolte d'*Ixodes dammini* immature, et peuvent être utiles pour déterminer les risques de maladie de Lyme pour l'homme.

INTRODUCTION

Lyme disease is a tick-borne ailment caused by a spirochete, *Borrelia burgdorferi*. Initial symptoms may include an expanding skin rash (erythema migrans), fatigue, myalgia, malaise, fever, "headache", and joint pain (STEERE *et al.* 1983a). Some

cases may develop severe neurologic (REIK *et al.* 1979) and cardiac (STEERE *et al.* 1980) abnormalities, as well as chronic arthritis (STEERE *et al.* 1979). Because of the seriousness of Lyme disease and its prevalence in the Midwestern and Northeastern United States, it has become a primary concern among public health officials.

1. Bureau of Disease Control, Westchester County Dept. of Health, White Plains, NY 10601, U.S.A.

2. Dept. of Biological Sciences, Fordham University, Bronx, NY 10458, U.S.A.

3. Medical Entomology Laboratory, Division of Infectious Diseases, Dept. of Medicine, New York Medical College, Valhalla, NY 10595, U.S.A.

Westchester County, New York, located to the immediate north of New York City, annually has a particularly high prevalence of Lyme disease. The number of cases has increased dramatically over the last several years, with the number of reported cases increasing from 60 cases in 1982 to 150 cases in 1983 (WILLIAMS *et al.* 1986). This trend appears to correspond to the abundance of *Ixodes dammini* Spielman, Clifford, Piesman, and Corwin, a vector of Lyme disease in the Northeast (STEERE *et al.* 1978, WALLIS *et al.* 1978, STEERE and MALAWISTA 1979).

In order to assess residential or recreational areas for risk of human exposure to *I. dammini* and *B. burgdorferi*, two parameters must be determined : 1) the potential of acquiring a tick bite as measured by the abundance of host-seeking ticks and : 2) the probability of a given tick being infected with *B. burgdorferi* as measured by the prevalence of spirochetes in host-seeking ticks. A single sampling method that can collect ticks which can be used to measure both of these parameters would be the most efficient and cost-effective way to assess risk.

The most common published method of estimating the abundance of adult and immature *I. dammini* is to collect ticks found parasitizing mammalian hosts (CAREY *et al.* 1980, MAIN *et al.* 1982, MAGNARELLI *et al.* 1984, WILSON *et al.* 1984). Because this method samples ticks already on hosts, we do not feel it is useful for comparing relative densities of ticks available to parasitize man or the prevalence of spirochetes in host-seeking ticks. We feel that a direct measure of host-seeking ticks more accurately represents the potential for human risk of Lyme disease.

Traps using dry ice as a source of CO₂ gas have previously been used to collect all stages of *Amblyomma americanum* L. (WILSON *et al.* 1972, KOCH and MCNEW 1982), *Dermacentor andersoni* Stiles (GARCIA 1965), and *I. ricinus* L. (GRAY 1985), as well as adult *D. variabilis* Say and *A. maculatum* Koch (SEMTNER and HAIR 1975). ANDERSON and MAGNARELLI (1980) used CO₂-baited traps as an aid in determining the distribution of nymphal and adult *I. dammini* in Connecticut. Our study was designed to test the effectiveness of CO₂-baited traps for sampling *I. dammini*. The study was

conducted during a period of larval activity in late summer because larvae are more abundant than later stages and therefore should be a more sensitive indicator of *I. dammini* presence than either nymphs or adults.

MATERIALS AND METHODS

Our traps were a modification of a design by WILSON *et al.* (1972). Trap bases were constructed of 33.5 × 33.5 × 3.5 cm treated pine with 40° beveled sides. A dry ice reservoir (14.5 × 14.5 × 17.5 cm) was constructed entirely of 1.5 cm thick styrofoam with a removable top. A 5 mm hole was drilled in the center of each side, approximately 3.5 cm from the base of the reservoir box, to allow escape of CO₂ gas. Masking tape (5 cm width) was placed over the top of the angled base, extending half way over the edge with the adhesive side facing down.

Traps were filled with approximately 1 kg of dry ice and checked twice daily between 0700-1000 hours and 1600-1900 hours. At that time, captured ticks were collected from the masking tape, reservoirs were recharged with dry ice, and the masking tape was replaced if adhesiveness was lost due to age or moisture.

A study site was established in each of 5 Westchester County towns : North Castle (Armonk Village), New Castle (Chappaqua Hamlet), Bedford (Katonah Village), Mount Kisco (Mount Kisco Village), and Pound Ridge. The Armonk, Chappaqua, and Katonah sites were located in the immediate vicinity of Lyme disease cases, as reported to the Westchester County Dept. of Health. All sites were located in wooded habitats dominated by oak (*Quercus* spp.), hickory (*Carya* spp.), and maple (*Acer* spp.).

One CO₂-baited trap was placed at each corner of a 30 × 30 m quadrat, selected arbitrarily at each site. Each site was sampled for 4 continuous days between 26 July and 26 August, 1983. Each 4 day sampling period consisted of 4 morning and 3 evening collections for each of the 4 traps set at each site. Morning and evening collections at the Mount Kisco and Chappaqua sites were compared statistically by the Mann-Whitney-Wilcoxon test to

determine if *I. dammini* larvae were constantly active.

In order to compare the relative abundance of larval *I. dammini* collected from the CO₂-baited traps with tick abundance on small mammal hosts, white-footed mice (*Peromyscus leucopus*) were also sampled and immature ticks were identified for each site. This rodent species is a common host for larval *I. dammini* (CAREY *et al.* 1980, MAIN *et al.* 1982) and is a reservoir of *B. burgdorferi* (LEVINE *et al.* 1985).

A 7 × 7 grid of 49 traps, each 10 m apart, was superimposed over the CO₂ trapping square. Both small (5.1 × 6.4 × 16.5 cm) and large (7.6 × 8.9 × 22.9 cm) Sherman live traps (H. B. Sherman Inc., Tallahassee, Florida, USA) were used and all traps were checked daily. Captured mice were anesthetized with ether, sexed, aged, tagged by toe clipping, and examined for ticks. All ticks were removed, and tagged mice were released at the site of capture. Only those mice caught for the first time each week at each site were included in our results in order to eliminate the effects of removal sampling. The relationship between the mean number of larval ticks collected by CO₂-baited tick traps and the number collected from captured mice was examined by the Spearman rank correlation test.

RESULTS

The mean number of larval *I. dammini* recovered per CO₂-baited trap ranged from 0 at Pound Ridge to 25.4 at Mount Kisco (Table 1). A total of 919 larvae were collected through the trapping period, with 712 larvae (77.5 %) collected from the Mount Kisco site.

TABLE 1 : Total *I. dammini* larvae collected by CO₂-baited traps

Site	No. larvae		Total no. larvae	Larvae/trap Mean + SD
	day ^a	night ^b		
Mt. Kisco	350	362	712	25.4 ± 29.24
Chappaqua	94	94	188	6.7 ± 9.99
Katonah	5	12	17	0.6 ± 1.91
Armonk	1	1	2	0.1 ± 0.26
Pound Ridge	0	0	0	0.0 ± 0.00
Total	450	469	919	6.6 ± 16.80

^a 12 trap days; ^b 16 trap nights.

A total of 469 larvae were collected in 80 trap nights and 450 larvae were collected in 60 trap days. Differences in morning and evening collections within the Mount Kisco and Chappaqua sites (which accounted for 97.9 % of the ticks collected in the study) were not significant as determined by the Mann-Whitney-Wilcoxon test ($p > .05$, $n_1 = 16$, $n_2 = 12$).

I. dammini larvae were collected from *P. leucopus* at all sites. The mean number of ticks per mouse ranged from 2.6 at Pound Ridge to 11.9 at Chappaqua (Table 2). Chappaqua and Katonah provided the most mice (19 each), with Chappaqua having the most larvae (226). The Armonk site, although having the least number of mice (3), had the second highest mean ticks per mouse (11.7).

TABLE 2 : *I. dammini* larvae collected from *P. leucopus*^a

Site	No. mice	No. larvae	Larvae/mouse Mean + SD
Mt. Kisco	10	62	6.2 ± 4.89
Chappaqua	19	226	11.9 ± 16.07
Katonah	19	176	9.3 ± 11.35
Armonk	3	35	11.7 ± 10.97
Pound Ridge	9	23	2.6 ± 4.53

^a Includes weekly first capture mice only.

The relationship between the mean number of larval ticks collected by CO₂-baited tick traps and the number collected from captured mice was examined by the Spearman rank correlation test. There was no significant correlation between the mean ticks per tick trap and the mean ticks per mouse among all sites ($r_s = 0.3$, $p > .05$, $n = 5$).

DISCUSSION

This study demonstrates that *I. dammini* can be collected with CO₂-baited traps. The collection of 712 larvae at Mount Kisco and 188 larvae at Chappaqua suggests that these traps are capable of attracting and capturing large numbers of ticks. The relatively few larvae collected from CO₂-baited traps at Katonah, Pound Ridge, and Armonk might be due in part to the temporal differences in

sampling, the spatial placement of traps independent of differences in tick population density, or it might reflect actual differences in the tick populations between sites.

Because CO₂-baited traps attract ticks that are seeking hosts, the absence of any significant difference in number of captures between day and night trap periods indicates that *I. dammini* larvae seek hosts during both periods. If nymphs and adults behave similarly, people may always be at risk of tick bites in areas where *I. dammini* is present.

There are several advantages to using CO₂-baited traps over alternative tick collection methods. CO₂-baited traps are relatively easy to maintain and do not require the skills associated with the trapping of small mammals. In addition, sampling with these traps is less time consuming and less labor intensive than mammal trapping. More areas can therefore be sampled in a given amount of time.

Although there was no correlation between the mean number of ticks caught in CO₂-baited traps and that collected from mice, there is presently no way to determine which sampling method most accurately estimates population densities of ticks in the field.

We feel that CO₂-baited tick traps are better suited for use in studies to determine the human risk for Lyme disease than is mouse trapping. Spirochete infection rates, expressed as the percentage of ticks containing *B. burgdorferi*, are more indicative of human risk when determined from unfed, host-seeking ticks because ticks on mice have already succeeded in finding a host and are no longer an immediate risk to humans. Additionally, infection rates derived from ticks parasitizing an infected mouse are likely to be higher than in unfed ticks because spirochetes may be acquired while feeding, especially on white-footed mice (LEVINE *et al.* 1985). Consequently, the use of ticks from mice may result in an artificially inflated infection rate.

Because transovarial transmission of the Lyme disease spirochete in *I. dammini* is extremely low (STEERE *et al.* 1983b, PIESMAN *et al.* 1986, MAGNARELLI *et al.* 1987), larvae would not be expected to contain spirochetes and may not pose a significant risk to humans. However, CO₂-baited traps should be equally effective in collecting other stages of *I.*

dammini and infection rates of nymphs and adults are indicative of human risk to Lyme disease. MOUNT and DUNN (1983) have demonstrated that CO₂-baited traps are useful in predicting a threshold for human exposure to *A. americanum*. Therefore, we feel that CO₂-baited traps may have a useful role in the surveillance of both *I. dammini* populations and the risk of Lyme disease.

ACKNOWLEDGEMENTS

We thank Christine FALCO and Luis RUEDAS of the Westchester County Dept. of Health and Fordham University, for their assistance in the collection of data. We also thank William GALA, Fordham University, for the construction of the traps and Dr. Thomas DANIELS, Medical Entomology Laboratory, New York Medical College, for reviewing the manuscript. This research was performed by R. F. in partial fulfillment of requirements for the Ph. D. degree in Biology at Fordham University and was funded in part by the Westchester County Dept. of Health.

REFERENCE

- ANDERSON (J. F.), and MAGNARELLI (L. A.), 1980. — Vertebrate host relationships and distribution of ixodid ticks (Acari : Ixodidae) in Connecticut USA. — *J. Med. Entomol.* **17** : 314-323.
- CAREY (A. B.), KRINSKY (W. L.), and MAIN (A. J.), 1980. — *Ixodes dammini* (Acari : Ixodidae) and associated ixodid ticks in south-central Connecticut, USA. — *J. Med. Entomol.* **17** : 89-99.
- GARCIA (R.), 1965. — Collection of *Dermacentor andersoni* (Stiles) with carbon dioxide and its application in studies of Colorado tick fever virus. — *Am. J. Trop. Med. Hyg.* **14** : 1090-1093.
- GRAY (J. S.), 1985. — A carbon dioxide trap for prolonged sampling of *Ixodes ricinus* L. populations. — *Exp. Appl. Acarol.* **1** : 35-44.
- KOCH (H. G.), and McNEW (R. W.), 1982. — Sampling of lone star ticks (Acari : Ixodidae) : dry ice quantity and capture success. — *Ann. Entomol. Soc. Am.* **75** : 579-582.
- LEVINE (J. F.), WILSON (M. L.), and SPIELMAN (A.), 1985. — Mice as reservoirs of the Lyme disease spirochete. — *Am. J. Trop. Med. Hyg.* **34** : 355-360.
- MAGNARELLI (L. A.), ANDERSON (J. F.), BURGDORFER (W.), and CHAPPELL (W. A.), 1984. — Parasitism by *Ixodes*

- dammini* (Acari : Ixodidae) and antibodies to spirochetes in mammals at Lyme disease foci in Connecticut, USA. — *J. Med. Entomol.* **21** : 52-57.
- MAGNARELLI (L. A.), ANDERSON (J. F.), AND FISH (D.), 1987. — Transovarial transmission of *Borrelia burgdorferi* in *Ixodes dammini*. — *J. Infect. Dis.* **156** : 234-236.
- MAIN (A. J.), CAREY (A. B.), CAREY (M. G.), and GOODWIN (R. H.), 1982. — Immature *Ixodes dammini* (Acari : Ixodidae) on small mammals in Connecticut, USA. — *J. Med. Entomol.* **19** : 655-664.
- MOUNT (G. A.), and DUNN (J. E.), 1983. — Economic thresholds for lone star ticks (Acari : Ixodidae) in recreational areas based on a relationship between CO₂ and human subject sampling. — *J. Econ. Entomol.* **76** : 327-329.
- PIESMAN (J.), DONAHUE (J. G.), MATHER (T. N.), and SPIELMAN (A.), 1986. — Transovarially acquired Lyme disease spirochetes (*Borrelia burgdorferi*) in field-collected larval *Ixodes dammini* (Acari : Ixodidae). — *J. Med. Entomol.* **23** : 219.
- REIK (L.), STEERE (A. C.), BARTENHAGEN (N. H.), SHOPE (R. E.), and MALAWISTA (S. E.), 1979. — Neurologic abnormalities of Lyme disease. — *Medicine.* **58** : 281-294.
- SEMTNER (P. J.), and HAIR (J. A.), 1975. — Evaluation of CO₂-baited traps for survey of *Amblyomma maculatum* Koch and *Dermacentor variabilis* Say (Acarina : Ixodidae). — *J. Med. Entomol.* **12** : 137-138.
- STEERE (A. C.), BARTENHAGEN (N. H.), CRAFT (J. E.), HUTCHINSON (G. J.), NEWMAN (J. H.), RAHN (D. W.), SIGAL (L. H.), SPIELER (P. N.), STENN (K. S.), and MALAWISTA (S. E.), 1983a. — The early clinical manifestations of Lyme disease. — *Ann. Intern. Med.* **99** : 76-82.
- STEERE (A. C.), BATSFORD (W. P.), WEINBERG (M.), ALEXANDER (J.), BERGER (H. J.), WOLFSON (S.), and MALAWISTA (S. E.), 1980. — Lyme carditis : cardiac abnormalities of Lyme disease. — *Ann. Intern. Med.* **93** : 8-16.
- STEERE (A. C.), BRODERICK (T. E.), and MALAWISTA (S. E.), 1978. — Erythema chronicum migrans and Lyme arthritis : epidemiologic evidence for a tick vector. — *Am. J. Epidemiol.* **108** : 312-321.
- STEERE (A. C.), GIBOFSKY (A.), PATARROYO (M. E.), WINCHESTER (R. J.), HARDIN (J. A.), and MALAWISTA (S. E.), 1979. — Chronic Lyme arthritis : clinical and immunogenic differentiation from rheumatoid arthritis. — *Ann. Intern. Med.* **90** : 896-901.
- STEERE (A. C.), GRODZICKI (R. L.), KORNBLATT (A. N.), CRAFT (J. E.), BARBOUR (A. G.), BURGDOFFER (W.), SCHMID (G. P.), JOHNSON (E.), and MALAWISTA (S. E.), 1983b. — The spirochetal etiology of Lyme disease. — *New Engl. J. Med.* **308** : 733-740.
- STEERE (A. C.), and MALAWISTA (S. E.), 1979. — Cases of Lyme disease in the United States : locations correlated with the distribution of *I. dammini*. — *Ann. Intern. Med.* **91** : 730-733.
- WALLIS (R. C.), BROWN (S. F.), KLOTTER (K. O.), and MAIN (A. J.), 1978. — Erythema chronicum migrans and Lyme arthritis : field study of ticks. — *Am. J. Epidemiol.* **108** : 322-327.
- WILLIAMS (C. L.), CURRAN (A. S.), LEE (A. C.), and SOUSA (V. O.), 1986. — Lyme disease : epidemiologic characteristics of an outbreak in Westchester County, N.Y. — *Am. J. Pub. Health.* **76** : 62-65.
- WILSON (J. G.), KINZER (D. R.), SAUER (J. R.), and HAIR (J. A.), 1972. — Chemo-attraction in the lone star tick (Acarina : Ixodidae) : I. Response of different developmental stages to carbon dioxide administered via traps. — *J. Med. Entomol.* **9** : 245-252.
- WILSON (M. L.), LEVINE (J. F.), and SPIELMAN (A.), 1984. — Effect of deer reduction on abundance of the deer tick (*Ixodes dammini*). — *Yale J. Biol. Med.* **57** : 697-705.