

Population Parameters of *Triatoma spinolai* (Heteroptera: Reduviidae) Under Different Environmental Conditions and Densities

MILDRED J. EHRENFELD, MAURICIO CANALS, AND PEDRO E. CATTAN

Departamento de Ciencias Biológicas Animales, Universidad de Chile, Casilla 2, Correo 15, Santiago, Chile

J. Med. Entomol. 35(5): 740-744 (1998)

ABSTRACT Population parameters of *Triatoma spinolai* Porter were studied using specimens collected in the north and central region of Chile. Two cohorts of 17 and 44 first instars were maintained at a constant temperature of 28°C and 70% RH. Two similar cohorts of bugs were exposed to 16–24°C and 55–75% RH and maintained under a photoperiod of 14:10 (L:D) h for 16 mo. The preimaginal period ranged between 285 and 372 d under constant conditions. The lower-density cohort required 9.5 mo to reach the adult stage compared with 12.4 mo for the high-density cohort. Bugs placed under variable temperature and relative humidity conditions did not survive long. Cohorts with higher densities had similar mortality rates with greater mortality occurring in cohorts that had lower numbers of bugs. Cohorts under constant temperature and relative humidity reproduced and basic reproduction rates (R_0 , intrinsic growth rate [r], and generation time [G]) were estimated. Cohorts with higher numbers of bugs had higher R_0 and r , whereas values of G were similar for both groups. Apparently, 25°C was a critical temperature threshold for *T. spinolai* and there appeared to be a minimal population density that allowed reproduction.

KEY WORDS *Triatoma spinolai*, wild bugs, population dynamics, R_0 , Chagas disease, Chile

THERE ARE TWO vectors of Chagas disease in Chile: *Triatoma infestans* Klug and *T. spinolai* Porter. *T. infestans* is considered to be a domiciliary vector and is the most important species. The sylvatic vector *T. spinolai* has historically been considered as a low-risk species relative to human health. However, an average of 7–11% of wild individuals has been determined to be infected with *Trypanosoma cruzi* in 1980. Ordenes et al. (1996) reported that 26% of the bugs were infected. Furthermore, *T. spinolai* has been detected in periurban areas (Schenone et al. 1980) and occasionally in human dwellings (Apt and Reyes 1986). Human populations in rural areas increase the probability of interaction between domiciliary and sylvatic bug–Chagas disease. Our objectives were to clarify the importance and vector potential for humans and to identify vulnerable points in the biology of the bugs that could be used in a pest management program.

Previous studies on temperature preference and activity cycle of *T. spinolai* (Canals et al. 1997) and on survival (Canals et al. 1994) suggested that this species has the capacity to colonize domiciliary environments in Chile. However, we do not know of any cohort studies that yield useful population parameters. The goals of the current work were to study the cohort dynamics of *T. spinolai* under different experimental conditions and to characterize the effect of temper-

ature and relative humidity on life cycle stages, reproduction, and survival.

Materials and Methods

Four cohorts of *T. spinolai* were used: 2 cohorts of 44 first instars were designated A1 and B1, and 2 cohorts of 17 first instars were called A2 and B2. The bugs were obtained from the insectary in the ecology laboratory at the Veterinary College, University of Chile. Each cohort was placed in a 1-liter glass jar, with absorbent paper used as a refuge. The top of the jar was covered with a fine mesh net. The cohorts were exposed to selected environmental conditions as follows: the A1 and A2 cohorts were maintained at 28°C and 70% RH throughout the experiment. The jar was placed in a computer-controlled climatic chamber. The B1 and B2 cohorts were maintained on shelves and exposed to ambient conditions that were comparable to those of human dwellings in Santiago, Chile (16–24°C and 55–75% RH). A photoperiod of 14:10 (L:D) h was maintained for all cohorts. The bugs were fed weekly on adult chickens.

The cohorts were observed for a period of 16 mo during which time egg counts, dead bugs, and survivors of each instar were recorded twice each month. At each observation, all the bugs, cast skins, and eggs were removed.

Preimaginal periods and duration of each instar were determined. By using Probit's method (Canals et al. 1994), we determined the mean time required for 50% (MT_{50}) of the bugs to reach each instar. The

The experimental procedures using chickens were in agreement with the International Guiding Principles for Biomedical Research Involving Animals (NIH) and comply with the "Principles of Animal Care" publication No. 86-23 (1995) of the National Institutes of Health.

Table 1. Mean time (MT₅₀) in which *T. spinolai* reached each instar

Instar	A1		A2		B1		B2	
	MT ₅₀	SD	MT ₅₀	SD	MT ₅₀	SD	MT ₅₀	SD
2	1.7	0.54	1.6	0.4	1.9	0.25	1.0	0.25
3	3.1	0.99	2.9	0.6	2.2	0.39	2.5	0.78
4	4.8	1.29	3.9	0.6	3.5	0.41	10.5	4.23
5	8.8	3.15	8.2	2.4	8.8	2.50	—	—
Adult	12.4	1.31	9.5	0.4	—	—	—	—

Mean time and standard deviation were obtained with Probit's method (Pike and Thompson 1986).

MT₅₀s were compared using Friedman test wherein cohorts A1, A2, B1, and B2 were considered as treatments and 2nd, 3rd, and 4th instars were considered as blocks.

Life tables were constructed that included survival (l_x), fecundity (m_x), mortality (q_x), life span (e_x), and reproductive values (v_x). Also, we estimated basic reproductive rates ($R_0 = \sum l_x m_x$), generation time ($G = \sum x l_x m_x / \sum l_x m_x$), and intrinsic growth rate ($r = \ln R_0 / G$). By using mortality (q_x), fecundity (m_x) and previous reported data of viability of eggs, we constructed a Leslie matrix and estimated the stable age vector as described by Pielou (1969).

To estimate mortality rates (μ), exponential models were fit to survival curves by means of regression analysis. Survival curves were compared with 2 methods: the Logrank Peto and Peto's method that uses modified chi-square tests to interpret survival data (Pike and Thompson 1986), and the homogeneity of slopes (mortality rates) with analysis of covariance (ANCOVA) analysis, considering cohorts A1, A2, B1, and B2 as treatments; survival as response variable; and time as a covariate. Multiple comparisons with Tukey test were performed (Zar 1996).

Results

Life Cycle. The preimaginal period of *T. spinolai* ranged between 285 and 372 d under constant temperature and relative humidity. The individuals remained as 1st instars for 28–59 d, 2nd instars for 40–67 d, 3rd instars for 32–49 d, 4th instars for 120–159 d, and 5th instars for 36–108 d. The lower-density cohort (A2) maintained under constant temperature and relative humidity took 9.5 mo to reach the adult stage, whereas cohort A1 took 12.4 mo. Cohorts maintained under variable temperature and relative humidity conditions were unable to reach the adult stage. The B1 cohort took 8.8 mo to reach the 5th instar and remained at that stage until the end of the 16-mo study. The B2 cohort reach the 4th instar in 10.5 mo.

Development. The MT₅₀ necessary to reach each instar is shown in Table 1. No differences were found in MT₅₀ when considering A1, A2, B1, and B2 as treatments and instars 2, 3, and 4 as blocks (Friedman test: 1.8, $P = 0.64$). Under the variable temperature and relative humidity the developmental process was interrupted at the 5th and 4th instar in cohorts B1 and B2, respectively.

Survival. Differences in survival curves were found (Logrank: 78.9, $P < 0.01$). All curves were fitted to

exponential decay (Fig. 1). Mortality rates were different ($F = 17.04$, $df = 3, 35$; $P < 0.01$) and the mean number that died ranged between 0.0038 and 0.0049 individuals per day. From the Tukey multiple comparisons we found that mortality rates of cohorts with the same density were similar (Table 2). In contrast, cohorts with lower densities had higher mortality rates.

Life Tables. The life tables of cohorts under constant conditions (A1 and A2) provided more information compared with fluctuating-temperature cohorts because these individuals were able to reproduce. We estimated R_0 , r , and G . Furthermore, V_x was estimated for the A1 cohort. In the A1 cohort, reproduction started at month 11, which is 4 mo earlier than was observed for the A2 cohort. In A1 cohort, R_0 and r were higher ($R_0 = 22.95$, $r = 0.24$) than A2 cohort ($R_0 = 3.29$, $r = 0.08$). However the generation times were similar in both A1 and A2 cohorts (13.2 and 15.8 mo, respectively). Individuals from cohorts that had lower densities also had shorter life spans after birth (e_0), which was consistent with our mortality data (Tables 3 and 4).

The stable vector factor showed that a greater proportion of the individual bugs was in a lower age class. Considering 27.7% of eggs as being fertile (Canals et al. 1994) at $24 \pm 2.7^\circ\text{C}$ and $73 \pm 7\%$ RH and 1 mo as

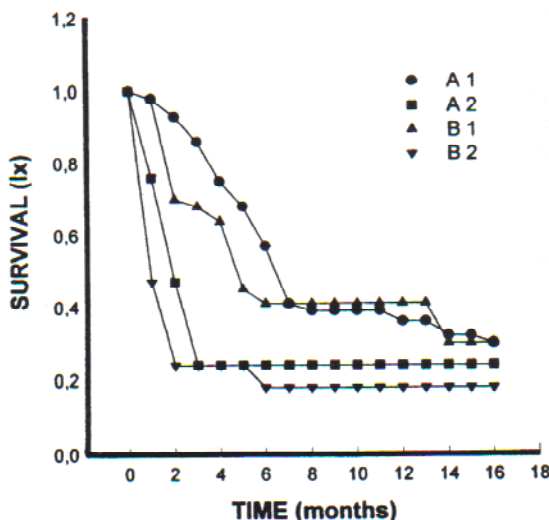


Fig. 1. Survival curves for cohorts of *T. spinolai* exposed to different environmental conditions and densities.

Table 2. Mortality rates of different cohorts of *T. spinolai*

Source of variation		F	P
Covariate (time)		86.64	<0.001
Main effect (cohorts)		17.32	<0.001
Cohort	μ	r^2	Tukey
A1	0.0041	94.1	a(*)
A2	0.0049	64.7	b
B1	0.0038	89.0	a
B2	0.0049	66.4	b

Analysis of covariance of survival considering the cohorts as treatments and time as a covariate. The slope of survival exponential decay represents the mortality rate (μ) of each cohort. The Tukey multiple comparisons of mortality rates.

the eclosion time, we found that a stable vector was reached. The observed proportions of 43.3% of eggs and 56.7% of bugs were stable (Table 5).

Discussion

When considering 1 mo as the time of bug eclosion, the life cycle of *T. spinolai* ranged between 315 and 402 d, similar to the 299–492 d reported by Lent and Jurberg (1967). The life cycle of this species was longer than that of other triatomines maintained under similar temperature and relative humidity conditions. Under constant temperatures of 25°C, the longest life cycles recorded for *Triatoma infestans* Klug, *Triatoma rubrovaria* Blanchard, and *Triatoma sordida* Stål were 167, 197, and 264 d, respectively. Variable temperature and relative humidity affected development. In contrast, bugs maintained under constant conditions and higher temperature reached the adult stage and several bugs reproduced. When nymphs were held under variable conditions and lower temperature, development stopped at the 4th or 5th instar. Individuals in the B1 cohort stopped development at the 5th instar. Bugs in the B2 cohort showed delayed MT_{50s} and further development was detained at the

Table 4. Life table of cohorts of *T. spinolai* under variable environmental conditions (16–24°C; 55–75% RH)

Month	B1 (44)			B2 (17)		
	l_x	d_x	e_x	l_x	d_x	e_x
0	1.00	0.02	8.6	1.00	0.53	4.4
1	0.98	0.27	7.8	0.47	0.24	7.5
2	0.70	0.02	9.5	0.24	0.00	12.0
3	0.68	0.05	8.7	0.24	0.00	11.0
4	0.64	0.18	8.2	0.24	0.00	10.0
5	0.45	0.05	10.2	0.24	0.06	9.1
6	0.41	0.00	10.1	0.18	0.00	11.0
7	0.41	0.00	9.1	0.18	0.00	9.8
8	0.41	0.00	8.2	0.18	0.00	8.8
9	0.41	0.00	7.2	0.18	0.00	7.8
10	0.41	0.00	6.2	0.18	0.00	6.9
11	0.41	0.00	5.2	0.18	0.00	5.9
12	0.41	0.00	4.2	0.18	0.00	4.9
13	0.41	0.11	3.2	0.18	0.00	3.9
14	0.30	0.00	3.0	0.18	0.00	2.9
15	0.30	0.00	2.0	0.18	0.00	2.0
16	0.30	—	—	0.18	—	—

4th nymphal instar. These responses to variable and restrictive environmental conditions were similar to those of *T. infestans*. Canals et al. (1992) found that under temperatures from 15 to 32°C and 40 to 90% RH, *T. infestans* delayed its development because of a longer preimaginal period (14.3 mo), increasing its generation time to 14.7 mo and reducing its fecundity and R_0 to 1.9 and 1.36 eggs per female, respectively. The climatic effects were evident when these values were compared with a preimaginal period of 6.1 mo; generation time of 7.1 mo; fecundity of 5.4 eggs per female-week; and R_0 of 25.04 at 26°C and 70% RH reported by Rabinovich (1972).

Previous work showed that at $24 \pm 2.7^\circ\text{C}$ and $73 \pm 7\%$ RH, *T. infestans* and *T. spinolai* delayed development. Under these conditions, the preimaginal period of *T. spinolai* lasted ≤ 541 d. Nevertheless, reproduction by newly developed adults was observed by Canals et al. (1994). In the current study, the lower tem-

Table 3. Life table of cohorts of *T. spinolai* under constant environmental conditions (28°C, 70% RH)

Month	A1 (44)					A2 (17)			
	l_x	d_x	e_x	m_x	v_x	l_x	d_x	e_x	m_x
0	1.00	0.02	9.4	0	23.0	1.00	0.24	5.5	0
1	0.98	0.05	8.6	0	23.4	0.76	0.29	5.9	0
2	0.93	0.07	8.0	0	24.7	0.47	0.24	8.0	0
3	0.86	0.11	7.5	0	26.7	0.24	0.00	13.7	0
4	0.75	0.07	7.5	0	30.6	0.24	0.00	12.8	0
5	0.68	0.11	7.1	0	33.8	0.24	0.00	12.8	0
6	0.57	0.16	7.3	0	40.3	0.24	0.00	10.8	0
7	0.41	0.02	8.8	0	56.0	0.24	0.00	9.8	0
8	0.39	0.00	8.2	0	58.9	0.24	0.00	8.8	0
9	0.39	0.00	7.2	0	58.9	0.24	0.00	7.8	0
10	0.39	0.00	6.2	0	58.9	0.24	0.00	6.9	0
11	0.39	0.02	5.3	9	58.9	0.24	0.00	5.9	0
12	0.36	0.00	4.6	20	51.3	0.24	0.00	4.9	0
13	0.36	0.05	3.6	7	33.9	0.24	0.00	3.9	0
14	0.32	0.00	2.9	8	30.2	0.24	0.00	2.9	0
15	0.32	0.02	1.9	14	22.2	0.24	0.00	2.0	3
16	0.30	—	—	9	8.9	0.24	—	—	11
R_0			22.95					3.29	
G (month)			13.20					15.80	
r			0.24					0.08	

Table 5. Stable age vector of *T. spinolai* under constant environmental conditions

Stage	Month	%	% ^a	% ^b	Instar
Egg	0	43.3	—		
	1	10.8	19.1		
	2	9.3	16.5		
	3	7.8	13.8	49.4	≤2
	4	6.4	11.3		
	5	5.0	8.8	20.1	3
	6	4.0	7.0		
	7	3.0	5.3		
	8	1.9	3.4		
	9	1.6	2.8	18.5	4
	10	1.4	2.5		
Nymph	11	1.2	2.1		
	12	1.1	1.9	6.5	5
	13	0.9	1.6		
	14	0.8	1.4		
	15	0.6	1.1		
	16	0.5	0.9		
Adult	17	0.4	0.7		
	18	0.4	0.6	5.7	

^a Nymphs + adults.^b Accumulated percentage.

perature rather than the relative humidity appeared to be the parameter responsible for delayed development in variable environments (cohorts B1 and B2) because relative humidity was similar to that of the cohorts maintained under a constant environment (A1 and A2). In contrast, temperatures that ranged from 16 to 24°C were lower than the constant environment temperature of 28°C and of those reported in previous studies ($24 \pm 2.7^\circ\text{C}$) (Canals et al. 1994). *T. infestans* and related species showed a decrease in developmental rate at lower temperatures and stop growth at 16°C (Schofield 1994). Jorg (1989) showed that hematophagous activity decreased at temperatures between 10 and 20°C, which limits development.

Under variable temperatures, *T. spinolai* showed a capacity to delay its development for a long time without causing morphological changes. This diapause phenomenon could be useful during period of unfavorable climatic conditions (Jorg 1962). Working with *T. infestans* maintained at 32°C and 52–78% RH, Jorg (1962) found that after 400 d, 10% of individuals had remained in the nymphal stage. Canals et al. (1992) showed that 15% of *T. infestans* remained as 1st instars after 6 mo when held under variable temperatures of 15–32°C and 40–90% RH. Diapause could be representative of an adaptive response that permitted survival until more optimum environmental conditions occurred.

In our study, reproduction occurred only in cohorts maintained under constant temperature and relative humidity. However, it is possible that the other cohorts could have completed development and reproduced if held for a longer time.

The mortality rates observed in our study for *T. spinolai* were comparable to the 0.0055 (24°C and 73% RH) reported by Canals et al. (1993) for *T. spinolai*, 0.0083 (25°C and 75% RH) reported by Rabinovich (1972), and 0.0070 (15–32°C and 40–90% RH) shown by Canals et al. (1992) for *T. infestans*.

In our study, the initial density of cohorts affected bug survival. Higher mortality rates at lower densities were observed. This insect density-dependent effect was not expected. Density probably affects developmental time and reproduction more than it affects mortality (Gorla and Schofield 1989). In our study no effects of density on the developmental process were found, probably due to having population sizes under the threshold necessary to provoke density-dependent effects.

A possible explanation for density-dependent mortality is that a positive social effect occurs in the access to the alimentary source. This could be mediated by the movement of individuals toward the alimentary source, determining that at higher numbers of individuals there was a higher probability of obtaining a food source. Consequently, obtaining a good nutritional basis would lower the mortality level. However, it must be considered that the effect would probably be limited by interference among individuals. Further study is necessary to test such a hypothesis.

R_0 of 22.95 obtained for the A1 cohort represented an increasing population with high reproductive capacity. Each female would produce a mean of 22.95 females in her lifetime. This value is similar to that reported for *T. infestans* ($R_0 = 25$) under 26°C and 70% RH (Rabinovich 1972). The R_0 of 3.29 for A2 cohort also represents a growing population. This value is higher than the R_0 of one proposed for the majority of the domiciliary populations of *T. infestans* by Rabinovich (1972).

In the A2 cohort, although adults were obtained 3 mo before they were found in the A1 cohort, the reproductive process started 4 mo later than that of the A1 cohort. The A2 cohort, with its lower density of bugs, took longer to reach the minimum number of adults necessary for reproduction. Social factors may be involved in the reproductive event and they probably would be triggered under a certain density threshold.

The stable age vector that was obtained showed that 77.8% of the population was ≤4 mo old (including eggs). This was similar to 79% age factor reported for *T. infestans* (Canals et al. 1992).

Environmental conditions of 28°C and 70% RH for A1 and A2 cohorts were almost equal to the 25°C and 70% RH that is considered optimum for populations of *T. infestans* that have been maintained for >30 yr in the University of Chile (Canals et al. 1992). However, the same acclimatization of the *T. spinolai* populations that have been exposed for 3 yr to these conditions was not expected. The food provided to support these bugs in our laboratory (chicken blood) could have been a limiting factor if it was of poor quality or lacked nutritive compounds that are essential for triatomine development. Reports of *T. spinolai* diet are scarce, but what is known reveals that the bugs are euryphagous and feed preferentially on mammals (Knierim et al. 1976, Durán et al. 1989). Lent and Wygodzinsky (1979) did not maintain *T. spinolai* colonies for >2 generations when the colony was fed on pigeon blood. The effect

of the species used to provide blood meals for laboratory colonies must be tested in more detail.

Acknowledgments

We thank C. Dayton Steelman and an anonymous reviewer for helpful suggestions to improve the manuscript. This research was supported by grants 194-0373 and 198-0768 from the Chilean Scientific and Technology National Fund (FONDECYT). The R & D Department, University of Chile, supported the cost of publication.

References Cited

- Apt, W., and H. Reyes. 1986. Aspectos epidemiológicos de la enfermedad de Chagas en Chile I. Distribución geográfica, índices de infección en vectores y humanos. *Parasitol. Dia* 10: 94–101.
- Canals, M., P. E. Cattán, R. Solís, and J. Valderas. 1991. Biología de poblaciones de *Triatoma infestans*: Fluctuaciones de la fecundidad y mortalidad. *Rev. Med. Chile* 119: 979–983.
- Canals, M., P. E. Cattán, M. Ehrenfeld, and P. Torres. 1992. Poblaciones experimentales de *T. infestans*: Efectos de condiciones ambientales variables. *Parasitol. Dia* 16: 72–77.
- Canals, M., P. E. Cattán, and M. Ehrenfeld. 1993. Algunas estimaciones numéricas de la importancia epidemiológica de los vectores chilenos de la enfermedad de Chagas en Chile. *Parasitol. Dia* 17: 79–86.
1994. Sobrevivencia de *Triatoma spinolai* en ambiente habitacional. *Parasitol. Dia* 18: 82–87.
- Canals, M., R. Solís, J. Valderas, M. Ehrenfeld, and P. E. Cattán. 1997. Preliminary studies on temperature selection and activity cycles of *Triatoma infestans* and *T. spinolai* (Heteroptera: Reduviidae), Chilean vectors of Chagas disease. *J. Med. Entomol.* 34: 11–17.
- Durán, J., M. Videla, and W. Apt. 1989. Enfermedad de Chagas en una comunidad de pequeños mamíferos simpátricos de la Reserva Nacional Las Chinchillas. *Parasitol. Dia* 13: 15–20.
- Gorla, D. E., and C. J. Schofield. 1989. Population dynamics of *Triatoma infestans* under natural climatic conditions in Argentina. *Med. Vet. Entomol.* 3: 179–194.
- Jorg, M. E. 1962. Influencia de temperaturas fijas en períodos anuales sobre metamorfosis y fertilidad de *Triatoma infestans*. *Bol. Chile Parasitol.* 17: 17–19.
1989. Area termoclimática desfavorable para el desarrollo de la vinchuca *Triatoma infestans* en el sudeste de la provincia de Buenos Aires. *Bol. Acad. Nac. Med.* 67: 382–398.
- Knierim, F., M. Castro, F. Villarroel, and H. Schenone. 1976. Estudio preliminar sobre la fuente de alimentación de *Triatoma infestans* y *T. spinolai* mediante la reacción de doble difusión en gel. *Bol. Chile Parasitol.* 31: 34–36.
- Lent, H., and J. Jurberg. 1967. Algumas informacoes sobre *Triatoma spinolai* com um estudo sobre as genitais externas (Hemiptera: Reduviidae). *Rev. Bras. Biol.* 27: 273–288.
- Lent, H., and P. Wygodzinsky. 1979. Revision of the triatominae (Hemiptera: Reduviidae) and their significance as vectors of Chagas disease. *Bull. Am. Nat. Hist.* 163: 334–335.
- Ordenes, H., M. Ehrenfeld, P. E. Cattán, and M. Canals. 1996. Infección tripano-triatomina de *Triatoma spinolai* en una zona de riesgo epidemiológico. *Rev. Med. Chile* 124: 1053–1057.
- Pielou, E. C. 1969. An introduction to mathematical ecology. Wiley New York.
- Pike, D. A., and J. N. Thompson. 1986. Statistical analyses of survival and removal rate experiments. *Ecology* 67: 240–245.
- Ravinovich, J. E. 1972. Vital statistics of Triatominae under laboratory conditions. *J. Med. Entomol.* 9: 351–370.
- Schofield, C. J. 1994. *Triatominae* biología y control. Eurocommunica, London, UK.
- Schenone, H., F. Villarroel, A. Rojas, and E. Alfaro. 1980. Factores biológicos y ecológicos en la epidemiología de la enfermedad de Chagas en Chile. *Bol. Chile Parasitol.* 35: 42–54.
- Zar, J. H. 1996. Biostatistical analysis, 3rd ed. Prentice-Hall, Englewood Cliffs, NJ.