

ESTIMATING THE GONOTROPHIC CYCLE AND SURVIVORSHIP OF *SIMULIUM OCHRACEUM* (DIPTERA: SIMULIIDAE) DURING ROUTINE VECTOR SURVEILLANCE IN SOUTHERN MEXICO

MARIO A. RODRIGUEZ-PEREZ,^{1,2} FILIBERTO REYES-VILLANUEVA³ AND MARIO H. RODRIGUEZ²

ABSTRACT. The gonotrophic cycle, survivorship, and daily parity rate of *Simulium ochraceum* were estimated from specimens collected during routine vector surveillance in southern Mexico, using a vertical (time-specific) method. Series of sequential data analysis on parity of the entomological data obtained in February 1994 showed the highest significant ($P < 0.05\alpha$) correlation indices ($r = 0.63$ and $r = 0.67$) for a 4-day time lag, indicating that the mean gonotrophic cycle length corresponded to 4 days. Daily survival rate was estimated to be 0.80, the survival to infective age (12 days) to be 6.9%, and the daily parity rate to be 0.41. These estimations are in accordance with those previously obtained by others from mark-release-recapture experiments (horizontal method) in Guatemala.

Simulium ochraceum Walker is the primary vector of *Onchocerca volvulus* (Leuckart) in southern Mexico and Guatemala. Its extensive distribution with relative multifocal isolation suggest the possibility of interspecific diversity (Hirai et al. 1994). However, the possibility of variation in this species' vectorial capacity across its geographical distribution has not been investigated and no information about the gonotrophic cycle and survivorship (important components to estimate the vectorial capacity) in the onchocerciasis-endemic areas of Mexico is available. Herewith we present estimates of the length of the gonotrophic cycle, survivorship, and the daily parity rate for *S. ochraceum* from specimens collected during routine vector surveillance in southern Mexico using vertical (time-specific) methods. These estimates were compared with those obtained from mark-release-recapture studies (horizontal method) in neighboring areas of Guatemala (Porter and Collins 1985, Collins et al. 1992).

The study was conducted in Las Golondrinas (15°25'59"N, 92°39'06"W), an onchocerciasis-hyperendemic (prevalence >50%) village situated at an elevation at 890 m in the Chiapas mountain range in southern Chiapas, Mexico.

As part of vector infection monitoring, female *S. ochraceum* collections were carried out during 20, 15, and 15 days of February (peak transmission occurs from January to March [Rodri-

guez-Perez and Rivas-Alcala 1992]) in 1992, 1993, and 1994, respectively. Capture periods, covering the blood-seeking parous female daily activity peak (Collins et al. 1981), included the first 30 min of each hour from 0800 to 1030 h. Collections were carried out by 2 teams of 2 village volunteers: while one person served as a human landing bait, the other used a mouth aspirator to capture the flies as they landed. One team was placed at the village center and the other in a coffee plantation 800 m away from the village. All specimens of *S. ochraceum* were dissected in 0.9% saline solution and classified as nulliparous or parous by the presence of sacculate dilatation and follicular relics in the ovarian tunic (Cupp and Collins 1979).

Time series for each 20- or 15-day sampling period in each collection site (village center and coffee plantation) were constructed. Each series was constructed using the number of parous (P_i) as the dependent variable (Y) and total females (T_i) as the independent variable (X). A cross-correlation analysis (Birley and Rajagopalan 1981) was applied to each series. The r -coefficient for day 0 represents the correlation between P_i and T_i data pairs from flies captured the same day (20-15 data pairs according to sampling period). The r -coefficient for day 1 was obtained by pairing daily P_i data with the corresponding T_i data of 1 day before. The day 2 coefficient was calculated by pairing daily P_i data with the corresponding T_i data of 2 days before, and so on. It was assumed that a significant cross-correlation coefficient (r) between the time series expresses a time delay (u) equivalent to the oviposition cycle (biting frequency). The highest significant cross-correlation coefficient (r) obtained after day zero ($u = 0$) indicated the number of days per gonotrophic cycle, with descending peaks occurring at multiples of this interval. A t -test to the highest cross-corre-

¹ Centro de Investigaciones Ecologicas del Sureste, World Health Organization Collaborating Center for Studies on Onchocerciasis, Tapachula, Chiapas, Mexico.

² Centro de Investigacion de Paludismo, Ministry of Health, Apartado Postal 537 Tapachula, Chiapas 30700 Mexico.

³ Universidad Autonoma de Nuevo Leon, Laboratorio de Entomologia Medica, San Nicolas de los Garza, N.L., Mexico.

Table 1. Number of captured *Simulium ochraceum* females in 15 days of sampling in a coffee plantation at Las Golondrinas, Chiapas, Mexico, February 1994.

Catch day	Total no. of females	No. parous females	Σ total no. of females	Σ no. parous females	Σ no. parous females/Σ total no. of females
1	86	48			0.56
2	132	52	218	100	0.46
3	60	39	278	139	0.50
4	45	26	323	165	0.51
5	122	45	445	210	0.47
6	82	45	527	255	0.48
7	98	38	625	293	0.47
8	72	27	697	320	0.46
9	78	67	775	387	0.50
10	33	19	808	306	0.38
11	46	22	854	328	0.38
12	44	19	898	347	0.39
13	29	19	927	366	0.39
14	27	21	954	387	0.41
15	36	18	990	405	0.41

lation coefficient in the time series was applied to goodness of fit. It was considered that values exceeding $\pm 1.96/\sqrt{n}$ were significant with $P < 0.05$.

In order to avoid false peaks in the correlation coefficients caused by uncontrolled factors (inconsistency in day-to-day collections or collections dominated by disturbances), the original data was transformed (filtration) by an auto-regressive equation with a time delay of 1 day of $Z_t = X_t - \beta(X_{t-1})$ where Z_t is the transformed data, X_t is the time series to be filtered, and β is the estimated auto-regressive parameter (Holmes and Birley 1987). The equation $P_t = S(T_{t-u})$ was

fixed through the origin (by regression analysis) and the value of β (slope) was the estimated survival rate (S).

Daily parity rate for the 1994 study was estimated using the formula $\Sigma P/\Sigma T$ (sum of parous females/sum of total female). The daily survival rate was also calculated using the root n of the daily parity rate: $DSR = \sqrt[n]{DPR}$ (Davidson 1954), where n is the number of days of the gonotrophic cycle.

After data filtration, only time series constructed from data collected in the coffee plantation during 1994 showed a definite pattern in the cross-autocorrelation analyses between total and parous females. Therefore, all other series were discarded from the analysis. A total of 990 females was collected with a parity average of 0.41 (405 parous females). The lowest capture occurred on day 14 (27 flies), and the highest capture (132 flies) occurred on day 2 (Table 1). The first significant peak ($P < 0.05\alpha$) appeared on day 4, using both the original and filtered data. The cross-correlation coefficients were $r = 0.63$ and $r = 0.67$ for this time delay ($u = 4$) (Table 2). In addition, 1992 original data collected from a coffee plantation showed 2 significant ($P < 0.05\alpha$) peaks on day 5 and 10, but this was not supported by filtered data. These results are in accordance to 5 and 3-4 days described in other studies (Watanabe et al. 1980, Porter and Collins 1985, Collins et al. 1992) in neighboring onchocerciasis-endemic areas in Guatemala.

The slopes obtained from the linear regression

Table 2. Cross-correlation values between total and parous *Simulium ochraceum* females for a time series of 15 days of sampling in a coffee plantation at Las Golondrinas, Chiapas, Mexico, February 1994.

Day	<i>r</i>	
	Original data	Filtered data
0	0.771	0.652
1	0.474	0.205
2	0.359	0.197
3	0.246	-0.020
4	0.635 ¹	0.671 ¹
5	-0.163	-0.247
6	-0.225	-0.333
7	0.598	0.598
8	-0.080	-0.647

¹ Statistically significant, $P < 0.05$.

analyses using original and filtered data were $Y = 5.88 - 0.32X$ and $Y = 8.86 - 0.31X$, respectively. However, these values could not be considered an adequate estimation of the survival per gonotrophic cycle because intercept values were statistically different from zero. On the other hand, using Davidson's formula (1954), the daily survival rate for 1994 was estimated to be 0.80 using a daily parity rate of 0.41 (Table 1). Considering at least 11–12 days for *O. volvulus* 3rd-stage larvae (L_3) to develop (Porter and Collins 1985), the survival to infective age was calculated to be $0.80^{12} = 6.9\%$. The same value of 0.80 elevated to the 4th potency was 40.9%, the survival rate for the 4th day (first gonotrophic cycle). Using experimental infections and field findings, Takaoka et al. (1981) and Porter and Collins (1988) observed that 3 gonotrophic cycles were needed for *S. ochraceum* from Guatemala to reach infectious stage. The surviving females per cycle was calculated as 39% and the survival of females to infectivity (0.39^3) as 6.3%. Those are very similar to our estimations in Mexican flies using time series analysis.

This is the first study we are aware of on the application of a time series method for the estimation of the gonotrophic cycle in *Simulium*. This method has advantages over mark–release–recapture techniques, which could be affected by low survival, behavior modifications induced by the dye, and sample lost due to dispersion (Milby and Reisen 1989). On the other hand, the sampling method in a time series is not always consistent. Dispersion of *S. ochraceum* populations within habitats is not well known; 3-year collections in the village center and data from 2-year collections in the coffee plantation showed inconsistent proportions in the age-structure distribution, and nonsignificant correlation indices for the time series analyses could be documented. A rapidly changing population structure in this species, as suggested by the markedly reduced number of flies captured during the last 6 days of sampling, as well as an inappropriate sampling size could also be responsible of the inconsistency among the time series analyzed.

Interspecific variation has been documented in Central America (Hirai et al. 1994). Interestingly, *S. ochraceum* cytotype A is present in our study area and in the area in Guatemala where previous estimations of the gonotrophic cycle and survivorship were conducted (Porter and Collins 1985, Collins et al. 1992, Hirai et al. 1994). This could explain the consistency in the results obtained. It would be interesting to estimate the gonotrophic cycle and survivorship of cytotypes B and C, which are distributed at different altitudes than cytotype A.

We thank Rene Solis-Franco, Anibal Aguirre-Sanchez, Raul Rodriguez-Perez, Rafael Vazquez-Sanchez, and Dina Gomez-Mendez for their support of our field work. This research was supported by the Colegio de la Frontera Sur (ECO-SUR) and the Consejo Nacional de Ciencia y Tecnologia (CONACyT) under grant number 3187-M.

REFERENCES CITED

- Birley, M. H. and P. K. Rajagopalan. 1981. Estimation of the survival and biting rate of *Culex quinquefasciatus* (Diptera: Culicidae). *J. Med. Entomol.* 18:181–186.
- Collins, R. C., M. Merino and E. W. Cupp. 1981. Seasonal trends and diurnal patterns of man-biting activity of four species of Guatemalan black flies (Simuliidae). *Am. J. Trop. Med. Hyg.* 30:728–733.
- Collins, R. C., J. O. Ochoa, E. W. Cupp, C. Gonzalez-Peralta and C. Porter. 1992. Microepidemiology of onchocerciasis in Guatemala: dispersal and survival of *Simulium ochraceum*. *Am. J. Trop. Med. Hyg.* 47:147–155.
- Cupp, E. W. and R. C. Collins. 1979. The gonotrophic cycle in *Simulium ochraceum*. *Am. J. Trop. Med. Hyg.* 28:422–426.
- Davidson, G. 1954. Estimation of the survival rate of anopheline mosquitoes in nature. *Nature* 174:792–793.
- Hirai, H., W. S. Procunier, J. O. Ochoa and K. Uemoto. 1994. A cytogenetic analysis of the *Simulium ochraceum* species complex (Diptera: Simuliidae) in Central America. *Genome* 37:36–53.
- Holmes, P. R. and M. H. Birley. 1987. An improved method for survival rate analysis from time series of haematophagous dipteran populations. *J. Anim. Ecol.* 56:427–440.
- Milby, M. M. and W. K. Reisen. 1989. Estimation of vectorial capacity: vector survivorship. *Bull. Soc. Vector Ecol.* 14:47–54.
- Porter, C. H. and R. C. Collins. 1985. The gonotrophic cycle of wild *Simulium ochraceum* and the associated development of *Onchocerca volvulus*. *Am. J. Trop. Med. Hyg.* 34:302–309.
- Porter, C. H. and R. C. Collins. 1988. Seasonality of adult black flies and *Onchocerca volvulus* transmission in Guatemala. *Am. J. Trop. Med. Hyg.* 38:153–167.
- Rodriguez-Perez, M. A. and A. R. Rivas-Alcala. 1992. Age structure of *Simulium ochraceum* and transmission of *Onchocerca volvulus* in Mexico. *J. Am. Mosq. Control Assoc.* 8:305–317.
- Takaoka, H., K. M. Hansen, H. Takahashi, J. Ochoa and E. L. Juarez. 1981. Development of *Onchocerca volvulus* larvae in *Simulium ochraceum* at various altitudes in Guatemala with special reference to the ambient temperature. *Jpn. J. Trop. Med. Hyg.* 9:187–197.
- Watanabe, M., I. Tanaka, T. Okazawa, T. Y. Yamagata and A. J. O. Ochoa. 1980. Notes on the age determination, ovariole changes and gonotrophic cycle of *Simulium ochraceum* in Guatemala. *Jpn. J. Sanit. Zool.* 31:215–222.