ECOLOGY, BEHAVIOR AND BIONOMICS

Life Cycle of *Goeldichironomus holoprasinus* Goeldi (Diptera: Chironomidae) in Laboratory

Florencia Zilli[1], Mercedes Marchese[1], Analía Paggi[2]

[1]Instituto Nacional de Limnología (Consejo Nacional de Investigaciones Científicas y Técnicas, Univ. Nacional del Litoral), Ciudad Universitaria, Santa Fe, Argentina; Facultad de Humanidades y Ciencias, Univ. Nacional del Litoral, Santa Fe, Argentina; [2]Instituto de Limnología "Dr. Raúl A. Ringuelet" (Consejo Nacional de Investigaciones Científicas y Técnicas, Univ. Nacional de La Plata), Florencio Varela, Buenos Aires, Argentina

Edited by Stephen Lapointe – USDA

Neotropical Entomology 38(4):472-476 (2009)

Ciclo de Vida de *Goeldichironomus holoprasinus* Goeldi (Diptera: Chironomidae) en Laboratorio

RESUMEN - Las larvas de Chironomidae son muy importantes en los ambientes acuáticos continentales por su abundancia y por su rol en las tramas tróficas. *Goeldichironomus holoprasinus* Goeldi es una de las especies más abundantes de Chironomidae en ambientes urbanos y naturales, siendo indicadora de calidad de ambientes. Por su tolerancia a factores ambientales estresantes muchos estudios se han realizado en relación con los efectos en su anatomía y atributos bionómicos. El objetivo principal de este estudio fue establecer las características del ciclo de vida de *G. holoprasinus* en condiciones de laboratorio. El crecimiento de las dimensiones cefálicas entre estadios tuvo una razón de 1,64 en promedio (proporción de Dyar), mientras que el crecimiento en longitud fue continuo. La temperatura promedio medida en el laboratorio de cría fue de 26ºC (18-33ºC). En estas condiciones el valor de *D* (tiempo de desarrollo desde la oviposición a la emergencia del primer imago) fue de 13 días y el valor de G mínimo (intervalo desde la oviposición a la primer progenie de la siguiente generación) fue de 16 días.

PALABRAS-CLAVE: Región Neotropical, Argentina, estadío

ABSTRACT - The larvae of Chironomidae are very important due to their density and role in aquatic food webs. *Goeldichironomus holoprasinus* Goeldi is one of the most abundant species of Chironomidae in man-made and natural systems, being widely used in water quality assessments. Because of its tolerance to stressing environmental factors, many studies have investigated the effects of stressors on *G. holoprasinus* anatomy and bionomic attributes. The main aim of this work was to describe *G. holoprasinus* life cycle attributes in laboratory conditions. The cephalic capsule growth among instars was 1.64 in average (Dyar proportion), whereas the total size growth was continuous. The average room temperature registered was 26ºC (18-33°C). In this conditions the *D* value (time from oviposition to first imago emergence) was 13 days, whereas the minimum *G* value (interval from the oviposition to the first progeny of the next generation) was 16 days. The emergence lasted four days, which determined that average G of 18 days. Thus, at registered temperatures *G. holoprasinus* has a short life cycle.

KEY WORDS: Neotropical Region, Argentina, stage

The immature stages (larvae and pupae) of Chironomidae inhabit a great variety of freshwater environments, being one of the more abundant insects (Pinder 1983). They can be found in lotic and lentic systems, stable or temporary, colonizing bottom sediments, weeds (Trivinho-Strixino & Strixino 1991, Marchese & Paggi 2004, Poi de Neiff & Neiff 2006) and also phytotelmatas (Ospina-Bautista *et al* 2004, Liria 2007), what reflects the wide adaptive strategies of this taxonomic group (Trivinho-Strixino & Strixino 1999). Besides, they are of great importance in the energetic metabolism of ecosystems, transferring organic matter and energy to the aquatic and aquatic-terrestrial food webs (Masaferro *et al* 1991, Paggi 1998).

Goeldichironomus holoprasinus Goeldi has a Neotropical and Neartic distribution (Cranston *et al* in Wiederholm 1989) and usually presents dense populations in natural and man-made habitats, especially in those that are organically enriched, colonizing ephemeral habitats as pioneer species (Epler 2001).

Despite the importance of chironomids in biomonitoring studies and laboratory toxicity tests (Rosenberg & Resh 1993, Nazarova *et al* 2004), little is known on their life

cycle attributes (Lindergaard & Mortensen 1988, Jackson & Sweeney 1995, Corbi & Trivinho-Strixino 2006). The main aim of this analysis was to describe the *G. holoprasinus* life cycle attributes in laboratory conditions.

Material and Methods

Four egg masses of *G. holoprasinus* were collected in lentic urban environments of Santo Tomé city (Santa Fe, Argentina, 31° 40' 2.54" S 60° 45' 13.09" W) in February 2007 and transported to the laboratory in recipients with water from the collecting site. They were conditioned in petri dishes up to the eclosion when first larval instar left the mucilaginous mass. The number of eggs per egg mass was counted and measured (width and length) with a micrometric scale under an optic microscope.

The larvae were placed in plastic aquariums (12 cm x 21 cm x 6 cm) with permanently oxygenated water (1L) at room temperature (18-33ºC). The room temperature (maximum and minimum) was daily measured with a standard thermometer. The larvae were fed with finely ground suspension of flaked fish food (TetraMin®) every two days (Trivinho-Strixino & Strixino 1982).

A sample of larvae was daily collected from the rearing containers, fixed in 5% formaldehyde 5% and preserved in 70% alcohol. Larvae were measured with a micrometric scale under an optic microscope. The cephalic capsule width (CCW: maximum ventral width of the cephalic capsule measured transverse to the major axis of the body), the ventral length (VL: ventral length of the cephalic capsule measured from the anterior margin of the mentum to the posterior margin of the cephalic capsule) and total body length (TBL: length measured from the anterior margin of the cephalic capsule to the final portion of the last abdominal segment) were measured for each collected larva. Larvae were separated into instars by the ratio between consecutive measurements of cephalic capsule and body length. The growth proportion (r) between larval instars (Dyar 1890) was calculated considering its wide application in arthropods. The growth curve was obtained by relating total body length and time, adjusting the regression curve to determine the model giving the best fit.

The duration of the larval instars, pupal and imaginal stages were determined as a population average of the four replicates. In order to determine the minimum immature development time (D) as the time when first individual imago emerged, the life time of adult (without feeding), the time of last individual emergence and the average generation time (G), the rearing aquariums were covered to retain the adults.

Results and Discussion

Egg masses. Each egg mass had an average of 382 ± 10 eggs. In a study carried out in Brazil by Corbi & Trivinho-Strixino (2006), the egg masses of *G. maculatus* presented 780 eggs, whereas *G. luridus* Trivinho-Strixino & Strixino had an average of 600 eggs. Similar differences were also reported between the tropical *Chironomus xanthus* (Rempel) and the subtropical *Chironomus calligraphus* (Goeldi) (Zilli *et al* 2008). These differences could be a consequence of diverse adaptive strategies of Chironomidae from different climates. The eggs of *G. holoprasinus* were 228.9 ± 11.74 µm long and 85.0 ± 9.27 µm wide.

Eggs of *G. holoprasinus* are laid in flat floating masses similar to those of *G. maculatus* Strixino & Strixino (Corbi & Trivinho-Strixino 2006). Once conditioned, the egg masses fell down to the bottom of the container, where eclosion began in approximately 36h. Despite the fact that many eggs eclosed in 24-36h, while other eggs in the masses took as long as three days (average of 2.3 days).

Larval instars. For *G. holoprasinus*, the abdominal tubules appeared in instar II and the anterior pair bifurcated in instar III, making difficult to identify earlier stages. However, the larvae can be identified from other species of genus by a fourth inner mandible tooth (Epler 2001).

The larval instars were clearly separated when comparing the relative size between the cephalic capsule (VL and CCW) and the total body length (TBL) (Fig 1 a,b). It is really difficult

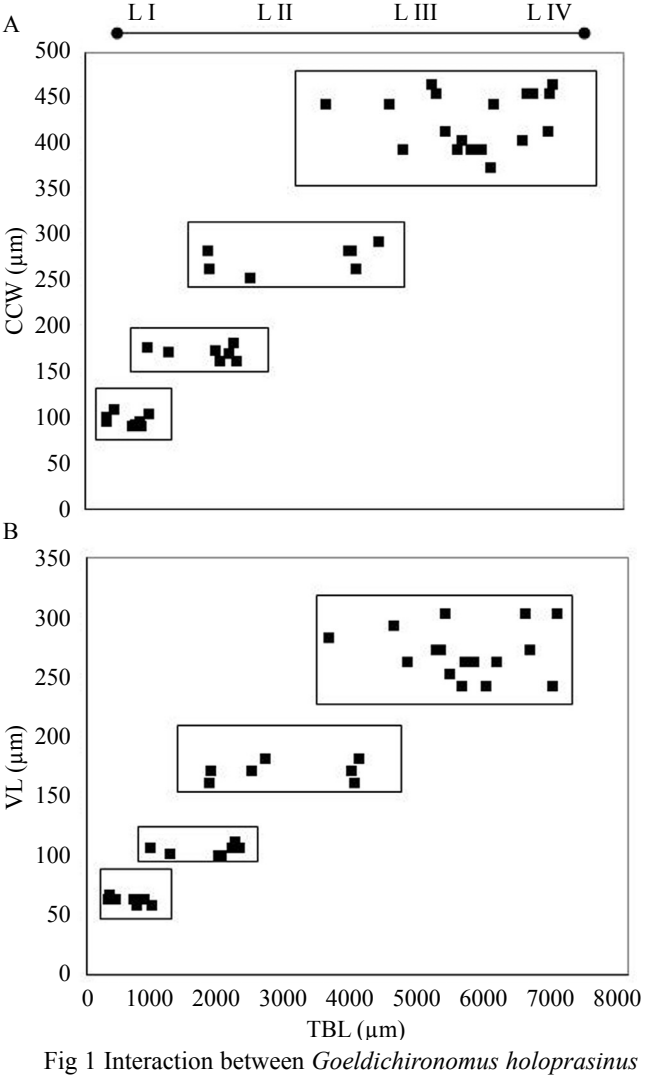

Fig 1 Interaction between *Goeldichironomus holoprasinus* larvae cephalic capsule ventral length (VL) and total body length (TBL). Larval instars are enclosed by squares.

to collect first instars of chironomids developing on macrophytes or sediments. However, the size of first instars can be estimated applying the Dyar´s rule. The growth proportions (Table 1) were very similar for cephalic capsule measurements (1.65 CCW and 1.63 VL) and an average value of 1.64 could be determined. In contrast, the growth proportion for TBL had a high variability (Table 1) with an average r of 2.14.

As stated by many authors, the more sclerotized structures (as the cephalic capsule) grow with gaps between moultings (Strixino & Trivinho-Strixino 1985), while the less sclerotized structures (as the abdominal segments) show continuous growth. Thus, the continuous length growth was the result of the resource exploitation capability of the organism, whereas the changes between instars were showed by cephalic increments.

The larval growth accounted for an average of 893% of the initial length, whereas for the cephalic capsule the total increment was approximately half of it (437%). Species such as *G. maculatus* and *G. luridus* (Trivinho-Strixino & Strixino) showed higher increments (Corbi & Trivinho-Strixino 2006), probably as a consequence of a longer larval developmental time. Moreover, *C. calligraphus* larvae (with immature stages developing in the same habitat) showed almost the same increment than *G. holoprassinus*, but the instars length was always higher (Zilli *et al* 2008). These differences in the development of immature stages among species in the same habitat show the different adaptive life cycle strategies used; while some species expend more time exploiting the food resources, others (such as *G. holoprassinus*) may enhance their fitness reducing the immature development time and the emergence period by synchronization, and probably by increasing the number of generations.

The growth curve obtained based on the TBL of larvae and time, showed a first phase of low increment for the instar I, followed by a rapid increase corresponding to instars II, III and IV. Finally, it reached an asymptote which corresponded to the last instar. The regression equation that best fitted to the data set was TBL = 6216.3 + (662.7 - 6216.3) / (1 + (time / 16.9)$^{4.6}$) (R^2 = 0.928) (Fig 2).

Table 1 Mean cephalic capsule width (CCW), ventral length (VL) and total body length (TBL). Standard deviations are indicated between brackets (n=128). The growth proportions (r) between instars are shown for each measurement.

Instars	CCW (μm)	r	VL (μm)	r	TBL (μm)	r
I	96.4 (6.61)		62.6 (2.38)		640.9 (224.83)	
		1.77		1.67		2.92
II	170.8 (6.96)		104.3 (3.90)		1869.8 (529.25)	
		1.60		1.66		1.66
III	273.9 (13.70)		172.9 (8.43)		3099.4 (1047.41)	
		1.56		1.56		1.85
IV	427.4 (29.93)		270.0 (19.59)		5720.3 (898.78)	
Average r		1.65		1.63		2.14

In the studied replicates, first instars were registered for an average of 2.5 day (1.5-3), second instars (II) registered for 3.0 days (2-4), third instars (III) for an average of 3.5 days (2-4) and fourth instars (IV) for an average of 6.5 days (5-7). As larvae at different instars coexisted in the

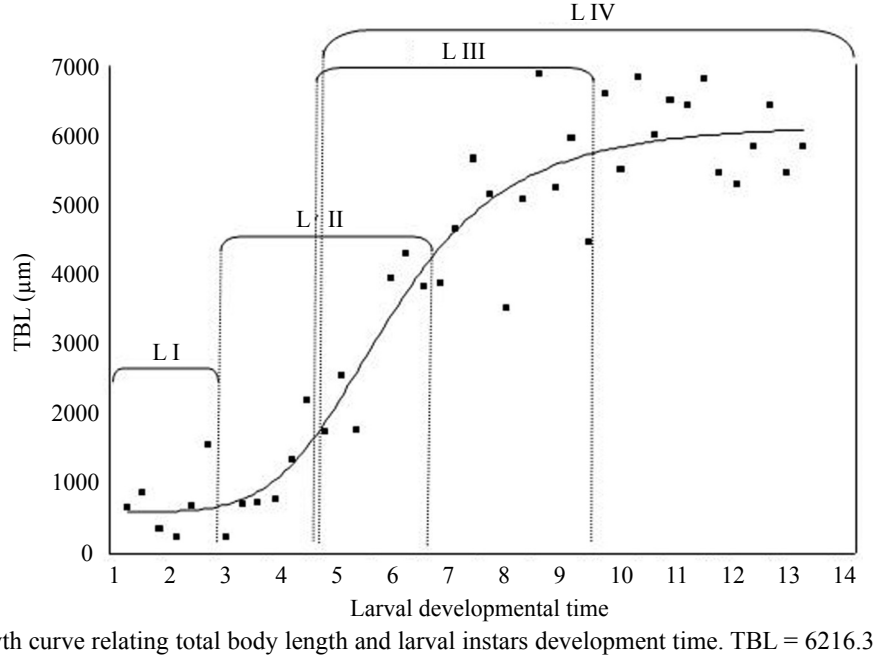

Fig 2 Larval growth curve relating total body length and larval instars development time. TBL = 6216.3 + (662.7 - 6216.3) / (1 + (time / 16.9)$^{4.6}$).

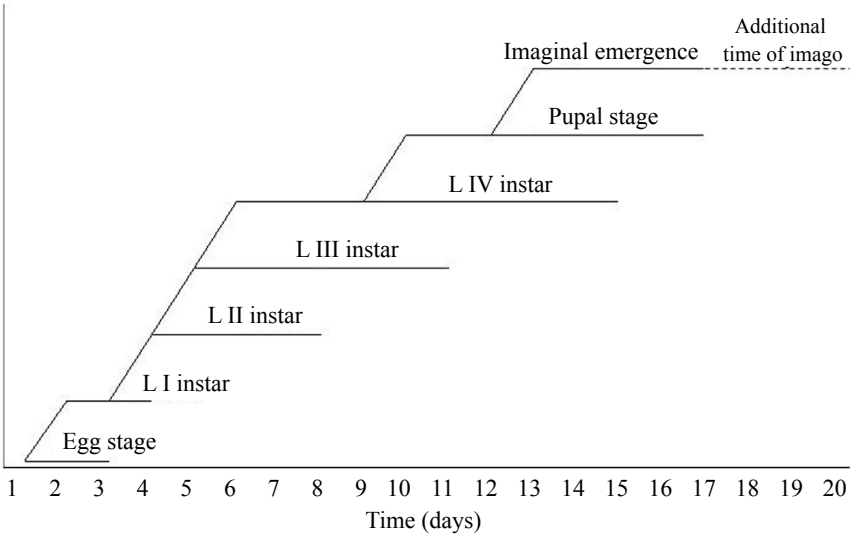

Fig 3 Duration of the different development stages and imago emergence of *Goeldichironomus holoprasinus*. The additional life time of emerged imagoes (without feeding) is showed (pointed line).

replicates (Fig 3), *G. holoprassinus* completed its larval developmental time in an average of 12.0 days (11-14).

Pupal and imaginal stages. Pupae were registered for an average of 4.5 days (3-5). The individual pupae developed in the tube built by fourth instars, and then swam actively to the surface up to the moment when imago emerged short after (usually a few minutes).

As the overlapping between larval instars and pupal stages was relatively high (Fig 3), the first emerged imago was registered in the 13th day (D). Imago emergence lasted only four days (up to 17th day) and as the last emerged imagoes lived three days without feeding, this additional time was considered the life time of individual imagoes (Fig 3). Thus, the minimum and maximum G were of 16 and 20 days respectively, and the average life cycle duration of *G. holoprassinus* was 18 days. The highest density of adults was observed at the 14th day in agreement with an increment in temperature (26ºC to 29ºC).

The effects of temperature and food availability on life history traits of chironomids are widely reported (Trivinho-Strixino & Strixino 1982, Strixino & Trivinho-Strixino 1985, Lindergaard & Mortensen 1988, Danks 2006). At high temperatures, larval development tends be more synchronized, favoring a short period for emergence (Trivinho-Strixino & Strixino 1982). In our study, high overlapping was observed when third and fourth instars and pupae coexisted (Fig 3), mainly because of the duration of fouth instars. However, a fast synchronically response probably due to the increment in temperature, determined a short emergence time (four days).

Some authors consider that a life cycle is short when last less than 12 days (G) (Danks 2006) and others establish a rank of 14 to 30 days as short (Strixino & Trivinho-Strixino 1985). In our study the average duration of *G. holoprasinus* generation time (G) indicates that this species has a short life cycle in temperatures around 26°C.

Acknowledgments

This study was supported by Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) and Universidad Nacional del Litoral (UNL).

References

Corbi J J, Trivinho-Strixino S (2006) Ciclo de vida de duas espécies de G*oeldichironomus* (Diptera, Chironomidae). Rev Bras Entomol 50: 72-75.

Cranston P S, Dillon M E, Pinder L C V, Reiss F (1989) The adult males of Chironominae (Diptera: Chironomidae) of the Holartic region-keys and diagnoses, p 353-502. In Wiederholm T (ed) The adult males of the Chironomidae (Diptera) of the Holarctic region - keys and diagnoses. Entomol Scand Suppl, Sandby, 532p.

Danks H V (2006) Short life cycles in insects and mites. Can Entomol 138: 407-463.

Dyar H G (1890) The number of molts of Lepidopterous larvae. Psyche 5: 420-422.

Epler J H (2001) Identification manual for the larval Chironomidae (Diptera) of North and South Carolina. NC Dept Environ Nat Res, Raleigh, NC 526p.

Jackson J K, Sweeney BW (1995) Egg and larval development times for 35 species of tropical stream insects from Costa Rica. J North Am Benthol Soc 14: 115-130.

Lindergaard C, Mortensen E (1988) Abundance, life history and production of Chironomidae (Diptera) in a Danish lowland stream. Arch Hydrobiol Suppl 4: 563-587.

Liria J (2007) Fauna fitotelmata en las bromelias *Aechmea fendleri* André y *Hohenbergia stellata* Schult del Parque Nacional San Esteban, Venezuela. Rev Peru Biol 14: 33-38.

Marchese M, Paggi A C (2004) Diversidad de Oligochaeta (Annelida) y Chironomidae (Diptera) del Litoral fluvial argentino, p.217-223. In Aceñolaza F C (ed) Temas de la biodiversidad del Litoral fluvial argentino. Insugeo, Tucumán, Argentina, 380p.

Masaferro J, Paggi A C, Rodrígues Capítulo A (1991) Estudio poblacional de los quironómidos (Insecta Diptera) de la laguna de Lobos, Provincia de Buenos Aires, Argentina. Graellsia 47: 129-137.

Nazarova L B, Riss H W, Kahlheber A, Werding B (2004) G Some observations of buccal deformities in chironomid larvae (Diptera: Chironomidae) from the Ciénaga grande de Santa Marta, Colombia. Caldasia 26: 275-290.

Ospina Bautista F, Estévez Varón J V, Realpe Rebolledo E (2004) Estructura y composición de macro invertebrados acuáticos asociados a *Tillandsia turneri* Baker (Bromeliaceae) en un bosque alto andino colombiano. Acta Zool Mex 20: 153-166.

Paggi A C (1998) Chironomidae, p.202-207. In Morrone J J, Coscarón S (eds) Biodiversidad de artópodos argentinos. Una perspectiva biotaxonómica. Ediciones Sur, La Plata, Argentina, 599p.

Pinder L C V (1983) The larvae of Chironomidae (Diptera) of the holartic region. Introduction, p.7-10. In Wiederholm T (ed) Chironomidae of the Holartic region - keys and diagnoses. Entomol Scan Supp, Sandby, 457p.

Poi de Neiff A, Neiff J J (2006) Riqueza de especies y similaridad de los invertebrados que viven en plantas flotantes de la planicie de inundación del río Paraná (Argentina). Interciencia 31: 220-225.

Rosenberg D, Resh V (1993) (eds) Freshwater biomonitoring and benthic macroinvertebrates. Chapman & Hall, London, 488p.

Strixino G, Trivinho-Strixino S (1985) A temperatura e o desenvolvimento larval de *Chironomus sancticaroli* (Diptera: Chironomidae). Rev Bras Zool 3: 177-180.

Trivinho-Strixino S, Strixino G (1982) Ciclo de vida de *Chironomus sancticaroli* Strixino & Strixino, (Diptera, Chironomidae). Rev Bras Entomol 26: 183-189.

Trivinho-Strixino S, Strixino G (1991) Estrutura da comunidade de insetos aquáticos associados à *Pontederia lanceolata* Nuttal. Rev Bras Entomol 53: 103-111.

Trivinho-Strixino S, Strixino G (1999) Insectos dípteros quironomídeos, p.141-148. In Ismael D, Valenti W C, Matsumura-Tundisi T Rocha O (eds) Biodiversidade do estado de São Paulo Brasil, 176p.

Zilli F L, Montalto L, Paggi A C, Marchese M R (2008) Biometry and life cycle of *Chironomus calligraphus* Goeldi 1905 (Diptera, Chironomidae) in laboratory conditions. Interciencia 33: 767-770.

Received 11/IV/08. Accepted 11/XII/08.