

Dynamics of Evolution and Resistance to Starvation of *Triatoma vitticeps* (Stal 1859) (Reduviidae: Triatominae), Submitted to Two Different Regimens of Food Deprivation

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The resistance to starvation of Triatoma vitticeps has been analyzed comparatively, according to different regimens of food deprivation under laboratory conditions. One cohort, composed of 100 specimens of each evolutionary nymphal stage, was submitted to continuous fasting until death; the second group, arranged in the same way, was fed once on chicken. Through this work, it was possible not only to compare the results obtained from the first group with other authors' results but, also, to analyze the dynamics of evolution, molting, longevity and the increase in insect longevity, in the second group. The average values recorded for survival time revealed statistical differences between the two groups. Among the important results detected, there is one that deserves to be emphasized: the incredible increase in longevity among insects that received only one feed – an average increase in survival time that reached 2.95 to 3.30 times in nymphs of 3rd and 4th stages, respectively. One 5th stage nymph survived for up to 350 days and the females may triplicate their survival rate, what represents an important epidemiological factor. The knowledge about this type of biological characteristic of T. vitticeps may contribute to prevent the domiciliation of this species, what seems to be incipient in some municipal districts, in Brazil.

Key words: *Triatoma vitticeps* - life cycle - resistance in starvation - triatomine longevity - Brazil

Triatominae are insects belonging to the Hemiptera Order, Family Reduviidae, and Subfamily Triatominae. Nowadays, there are about 131 vector species known, distributed among 6 tribus and 17 genera (Carcavallo et al. 2001). All of them are absolute bloodsuckers, regardless of age, sex or evolutionary stage. They are susceptible to infections by *Trypanosoma cruzi* (Chagas, 1909) and acquire the infection after feeding from an infected mammal, thus becoming vectors of Chagas disease, which today affects thousands of people, mainly in South America.

Triatoma vitticeps (Stal, 1859) is found in four Brazilian states: Bahia, Minas Gerais, Espírito Santo and Rio de Janeiro. Basically, it is still considered a wild species for two reasons: (a) the high number of adult insects captured in human dwellings infected with the *T. cruzi* (Santos et al. 1969b, Silveira et al. 1983, Ferreira et al. 1986, Sessa & Carias 1986, Dias et al. 1989); (b) the low rates of fortuitous infections in humans, in these areas (Santos et al. 1969 a, c, Pinto et al. 1969, 1986, Barros et al. 1975, Peçanha et al. 1983, Silveira et al. 1983, Dias et al. 1989). This indicates that the infection is acquired outside the residences, in places that have already been described as natural biotopes. However, Santos et al. (1969b) had already found domestic specimens in the State of Espírito Santo, where, at the occasion, there was a suspicion that the disease transmission had occurred through the action of the insect vector.

Silveira et al. (1984) registered the occurrence of this species in 21 cities of the State of Espírito Santo. In two towns, Cachoeiro do Itapemirim and Guarapari, the process of domiciliation was confirmed by the finding of household nymphs.

In the State of Rio de Janeiro, Ferreira et al. (1986), with the collaboration of the Superintendence of Public Health Campaigns, identified the presence of *T. vitticeps* in 4 municipal districts of the northern area (Campos, Itaocara, Porciúncula and São João da Barra); 3 of the metropolitan area (Maricá, Niterói and Rio de Janeiro) and 2 of the highland area (Santa Maria Madalena and São Sebastião do Alto). Eighteen adult insects among the 51 tested presented *cruzi*-like flagellated forms. In the municipal district of Santa Maria Madalena, 8 household nymphs were collected.

Recently, Gonçalves et al. (1998) captured 465 specimens of *T. vitticeps* (98% were captured indoors), in an eco-epidemiological study performed in the municipal district of Santa Maria Madalena. From this total, 202 (111 alive and 91 dead) were examined and infection by *Trypanosoma cruzi* was confirmed in 88 live bugs and in 31 of the dead ones.

This species had already demonstrated to be suitable as an experimental model for xenodiagnostic tests (Perlowagora-Szumlewicz et al. 1990); this means that it behaves as a proper vector species, presenting high infectivity rates and good parasitic density, for periods of up to 120 days after the infection, when infected by 7 different strains of *T. cruzi*. It was also tested in endemic areas of Chagas disease, in the State of Minas Gerais (Moreira & Perlowagora-Szumlewicz 1997), with better results than *T. infestans* (Klug, 1834). During that study, it was also verified a high level of resistance demonstrated

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by the low mortality rate. However, other characteristics are important to define a good vectorial species, as for the defecation timing during or after feeding, which affects the possibility of transmission of *T. cruzi* to man (Gonçalves et al. 1988).

The domiciliation process of *T. vitticeps* seems to be due to environmental modifications of those areas, as the increase of residences and concomitant shortage of insects' alimentary sources, condition already detected by Barreto (1976) and Silveira et al. (1983). However, there are still few pertinent data concerning the biology of this species and the present work intends to contribute with more information, through original observations, that may help in the efforts to avoid a possible domiciliation of this species.

MATERIALS AND METHODS

All the insects used in this study were derived from our laboratory colony, maintained in the Núcleo de Interação *T. cruzi*-Vetores da Doença de Chagas, Departamento de Medicina Tropical, Instituto Oswaldo Cruz, Fundação Oswaldo Cruz.

Cohorts of 100 specimens of each nymphal stage recently emerged were submitted to continuous fast until death.

More than 100 specimens of each evolutionary stage were fed only one time on chicken (*Gallus gallus*), and 100 well engorged specimens were separated, forming another cohort. This second group was separated according to the feeding date, so that it should be observed the evolution dynamic of the nymphs as well as the mean capacity of blood ingestion per stage.

The bugs were kept in 235 mm x 655 mm glass cylinders, covered with cheesecloth. The bottom of the cylinders were covered with a circle of filter paper (K40) and, in their interior, there was an accordion-pleated strip of the same paper, in order to increase the area of contact of the insects and to remove the excess of humidity, in all cases of the insects were fed a single time. The adult insects and those ones in the 5th nymph instar were divided between two glass jars, to avoid overcrowding.

All groups were daily checked for mortality and ecdysis.

During the accomplishment of the experiment, the bugs were maintained at controlled environmental temperature and humidity, with averages of $26^{\circ}\text{C} \pm 2^{\circ}\text{C}$ and $75\% \pm 5\%$, respectively.

For statistical comparisons, we used t Student test.

RESULTS

The increasing of survival time is proportional to the nymphal stage maintained under conditions of complete fasting until death. Nymphs of the first stage presented the largest pre-mortality period: 39 days. Adult insects presented a lower resistance average, as compared to other evolutionary stages: 26.7 and 33.6 days for males and females, respectively (Table I). In Table I it is also demonstrated that the averages varied in an increasing manner, except for the 1st stage, which presented a superior average (50.6) as compared to the two following stages. The shortest resistance was observed in the 2nd and 5th stages (7 days) and the longest, in the 5th stage (151 days).

Table II shows results obtained when nymphs were maintained in starvation, after a single feeding after molting, for each evolutionary stage. The survival average rate increased, according to the stage of development until the 4th stage, at which point the rate was found to be a marginally superior than at the 5th stage (168.4 and 165.9, respectively). The shortest resistance was observed at the 1st stage (15 days), in contrast to fasting nymphs, and the longest one was once again verified in nymphs at the 5th stage (347 days). All differences among the averages presented in Tables I and II, tested separately for each stage, using the t-test of Student ($p < 0.05$), are statistically significant.

The percentage of molted nymphs (Table III) varied from 39% to 75% at the 4th and 2nd stages, respectively.

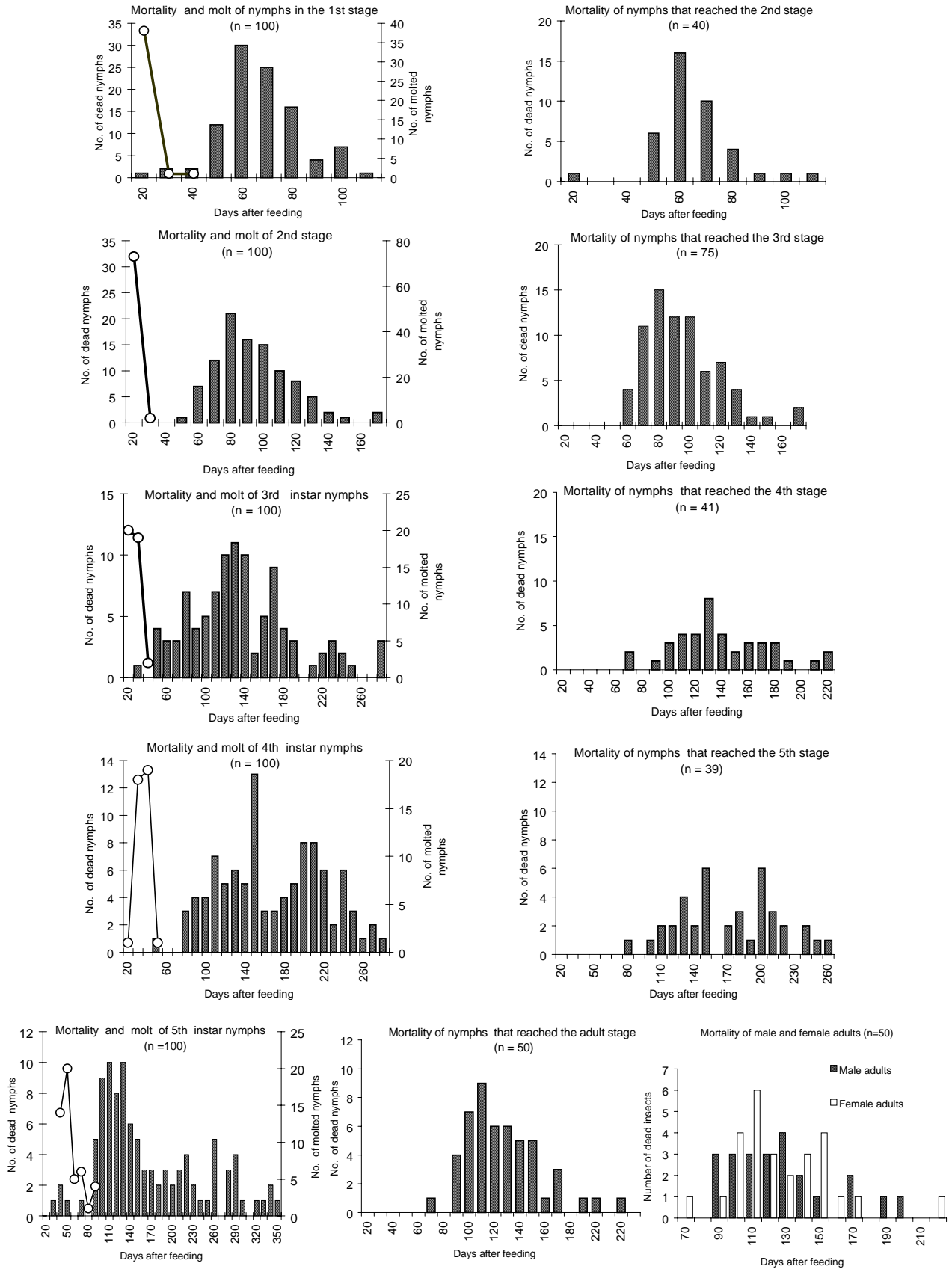
The dynamics of evolution, mortality and molting of nymphs and adults fed for a single time also varied in agreement with nymphal stages (Figure). The time of survival of nymphs that molted to a subsequent stage, as compared to those that stayed at the initial stage of development (in the selection of insects for this study), can be also verified in the Figure. This period was slightly lower in nymphs at 4th stage (proceeding from the 3rd stage) and at the adult stage (proceeding from the 5th stage), compared with nymphs that did not move on to subsequent stages. However, at the 5th stage this difference was more accentuated (350 vs 220 days).

Table IV shows, in a comparative way, the longevity differences between nymphs that were fed a single time and those maintained under conditions of fasting until death. The major longevity difference among the last live specimen registered for each group was observed at the

TABLE I
Resistance period (days) in complete starvation in *Triatoma vitticeps*

Nymphal stage	N	Amplitude		Mean	SD
		Minimum	Maximum		
1st	100	39	63	50.6	5.1
2nd	100	7	64	42.5	13.3
3rd	100	10	70	46.2	14.4
4th	100	13	95	51	19.7
5th	100	7	151	71.5	37.4
Males	41	30	90	26.7	20.5
Females	59	30	90	33.6	20.2

SD: standard deviation



Dynamic of evolution of the nymphs and adults, fed for a single time. Blanched circles in composed graphs exhibit the nymphs that molted to a superior stage. Columns represent mortalities.

TABLE II
Resistance period (days) in starvation after a single feeding subsequent to molting stage in *Triatoma vitticeps*

Nymphal stage	N	Amplitude		Mean	SD
		Minimum	Maximum		
1st	100	15	107	67.6	16.2
2nd	100	41	162	94.1	23.6
3rd	100	23	272	136.2	54
4th	100	43	280	168.4	52.6
5th	100	21	347	165.9	75
Males	46	90	200	127.8	31
Females	54	70	220	126.3	30.1

SD: standard deviation

3rd (202 days) and 5th stages (196 days). At the 5th stage, this difference is of greater importance, since this stage precedes the adult phase.

In this study, the average gain in the lifetime of the insects turned the differences statistically significant ($p < 0.05$), in comparison with the nymphs maintained in total starvation. The increase in longevity was proportional to the increase in the developmental stages.

The nymphs mean capacity for blood ingestion is shown in Table V. Likewise other already analyzed parameters, the capacity of blood ingestion showed a continued increase until the 5th stage.

DISCUSSION

According to Neiva and Lent (1936), the first reports concerning the capacity to resist conditions of fasting observed in insects were made by Charles Darwin, who noticed a specimen that resisted lack of food for four months. Leboubène observed a still longer period and Porter registered a specimen of *T. infestans* that survived for 17 months (apud Galvão et al. 1996).

Uribe (1926) reported the survival of one 3rd stage nymph of *Rhodnius prolixus* Stal, 1859 during 5 months and, since then, several authors have described the fasting capacity of these insects. Pellegrino (1952) emphasized the epidemiological importance of this characteristic in Triatominae and, according to this author, the resistance to starvation provides the passive transport of the insect to another place and its subsequent development, so that it soon finds a favorable environment for such. It also represents an escape mechanism against the action of insecticides, which are applied in endemic areas.

According to Dias (1965) and Perlowagora-Szumlewicz (1969), this resistance might be of great importance and directly affect the control campaigns against these vectors, since domesticated species can hide in places, like wall openings, that are inaccessible to the action of insecticides or to its possible residual effects (the insects would be protected from the action of insecticides during the time that the residual effect of the product lasted); thus, the possibility of re-colonization is increased.

In this work, the resistance averages found were lower than those reported by Silva (1985) and Gonçalves et al. (1988). The exceptions were the averages found by those authors in nymphs of the 1st stage, maintained under star-

TABLE III
Percentile of molted nymphs after one feeding

Nymphal stage	Percentile (%)
1st	40
2nd	75
3rd	41
4th	39
5th	50

vation. These different results were probably influenced by laboratory conditions, such as different blood sources supplied and variations in environmental conditions (humidity and temperature). The highest averages were reported by Gonçalves et al. (1989), who used mice as alimentary source.

In our study, adult insects demonstrated a lower resistance in comparison with other stages. In this last stage the female insect was more resistant than the male (33.6 and 23.7 days, respectively). Data reported by Silva (1985) and Gonçalves et al. (1989) demonstrate alternating results: sometimes higher for the male and sometimes higher for the female.

The averages observed in the 2nd and 3rd instar nymphs were also inferior to the value observed in the 1st stage (Table I). This result was influenced either by food reserves carried over from the eggs, that are maintained by nymphs throughout their entire life, until death, as by the premature mortality of some nymphs (6 nymphs died during the first 10 days, at the second stage, and 2 nymphs died on the 10th and 20th days, at the 3rd stage). Precocious mortality as observed in nymphs at the 2nd stage was also observed by Galvão et al. (1996), when they studied the resistance to starvation in *T. nitida* Usinger, 1939; by Cortez and Gonçalves (1998) for *T. rubrofasciata* De Geer, 1773; and by Martinez-Ibarra and Kathhain-Duchateau (1999) for *T. pallidipenis* Stal, 1872. In our case, it seems to be due mainly to the incapacity to ingest the blood meal, since dead bugs had no alimentary contents in their digestive tracts.

Another important aspect was the evolution process of nymphs fed only once in each stage (Figure). This kind

of study begins to be valued by some authors, and not only the study of resistance to absolute fast, because this factor associated with the capacity of the insects to feed from several alimentary sources constitute an important survival strategy. In this context, Dias-Lima and Sherlock (2001) performed a comparative study on resistance to starvation, after a single blood meal by stage, in six species of vectors. They verified different resistance rates for different evolutionary stages, drawing attention to *T. matogrossensis* (Leite and Barbosa, 1953), which resisted up to 575 days, at the 4th nymph stage.

According to Galvão et al. (1995) some triatomine species can reach the 4th stage with just one feeding in each stage, which is in agreement with the findings of Brasileiro and Perondini (1974) results with *T. brasiliensis* Neiva, 1911. On the other hand, Heitzmann-Fontenelle (1972) and Gonçalves et al. (1997), both working with *T. pseudomaculata* Corrêa & Espínola, 1964, demonstrated the requirement for more than one feeding at each stage for the successful development of this species. The present results indicate that the percentage of molted nymphs varied from 39% to 75%, at 4th and 2nd stages, respectively (Table III).

The largest rate of nymphs that molted to a superior evolutionary stage was observed in nymphs at the second stage, followed by 5th stage nymphs.

The time of survival in the cohorts of selected nymphs was longer than of those that passed through ecdysis, for nymphs at the same stages (the first ones belonging to the previously separated cohorts at the beginning of experiments and the last ones originating from ecdysis), which can be attributed to the metabolic costs associated with the process of ecdysis. This can be observed in Figure comparing, for example, the graph that shows mortality in nymphs that reached the 2nd stage (n = 40) with the graph that demonstrates the mortality and molting of nymphs at the same stage (n = 100), and so forth. These differences can also be explained by the larger quantities of blood ingested by the nymphs that stayed at the initial stage, at the moment that the cohorts were created (Table V).

An important result was the average increase in survival rates for insects that fed only a single time (Table IV). These insects presented an average gain of survival time that varied from 1.34 times (1st stage) to 4.79 times (males). The females may triplicate their survival rate. Similar date were seen with nymphs at the 3rd and 4th

stages, what represents another important epidemiological factor. The great survival capacity demonstrated by the 5th instar nymphs where the last insect died within 350 days, after a single feeding, must be emphasized. Thirty nine percent of the nymphs fed at the 4th stage passed on to the 5th stage (Figure) and that, in this last stage, 10% of the nymphs survived 275 days after the single feeding. If their survivors manage to feed at this stage, 50% of them will pass on to the adult phase and will survive long enough to reproduce (10% of the adults can survive up to 160 days).

Lima et al. (1991) tested experimentally the toxicity of several synthetic insecticides (hexachlorocyclohexane, DDT, tricolorfon, malathion, cypermetrin, deltametrin and permethrin). She added them to the food intake of six triatomine species (*Panstrongylus megistus*, *T. infestans*, *T. pseudomaculata*, *T. vitticeps*, *T. brasiliensis* and *Rhodnius prolixus*) and, among all these species, *T. vitticeps* demonstrated the largest level of resistance to any tested insecticide, except for HCH in concentration of 1 g/l, in spite of the fact that *T. vitticeps* is not a household species. This could indicate that this species is naturally resistant.

According to Silva et al. (2001), *T. vitticeps* is invading residences also in the State of Minas Gerais, where it is sometimes lodged, also presenting high infection rates by *T. cruzi*. In spite of the fact that a human being seldom is infected by the parasite, its circulation is happening in reservoirs and there is risk for becoming endemic, by linking the wild cycle to the domestic one.

The resistance to insecticides and resistance to fasting after a single blood meal at each nymph stage, as the

TABLE V
Nymphs mean capacity of blood ingestion (mg)

Stage	Mean of blood intake	SD
1st	9.26	1.75
2nd	36.74	7.56
3rd	118.24	20.23
4th	209.49	30.77
5th	411.67	37.89
Adult	149.19	10.76

SD: standard deviation

TABLE IV
Comparison of survival time between fed and unfed nymphs (in days)

Nymphal stages	Unfed nymphs Min - Max	Unfed nymphs (average ± SD)	Fed nymphs Min - Max	Fed nymphs (average ± SD)	Average gain in insects life (x)
1st	39 - 63	50.6 ± 5.1	15 - 107	67.6 ± 16.2	1.34
2nd	7 - 64	42.5 ± 13.3	41 - 162	94.1 ± 23.6	2.21
3rd	10 - 70	46.2 ± 14.4	23 - 272	136.2 ± 54	2.95
4th	13 - 95	51 ± 19.7	43 - 280	168.4 ± 52.6	3.30
5th	7 - 151	71.5 ± 37.4	21 - 347	165.9 ± 75	2.32
Males	30 - 90	26.7 ± 20.5	90 - 200	127.8 ± 31	4.79
Females	30 - 90	33.6 ± 20.2	70 - 220	126.3 ± 30.1	3.76

SD: standard deviation

present work has demonstrated, may increase the risk of domiciliation of *T. vitticeps* in some areas. Therefore, we conclude that an epidemiological policy is vital in those areas under risk, mainly because of the inevitable environmental changes caused by man. Hence if the necessary actions to control these insects are needed, the knowledge of the time that the species may resist to starvation after a single blood meal be of importance for elaboration of such actions, thus avoiding flaws in the methodology.

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REFERENCES

- Barreto MP 1976. Possible role of wild mammals and triatomines in the transmission of *Trypanosoma cruzi* to man. In International Symposium on New Approaches in American Trypanosomiasis Research, Belo Horizonte, MG, 1975. Proceedings Washington DC, Pan American Health Organization, p. 307-316 (PAHO-Scientific Publication 318).
- Barros GC, Mayrink W, Salgado AA, Barros RCG, Sessa PA 1975. Contribuição para o conhecimento da doença de Chagas autóctone no Espírito Santo. *Rev Inst Med Trop São Paulo* 17: 319-329.
- Brasileiro VLF, Perondini ALP 1974. Biologia de *T. brasiliensis* (Hemiptera, Reduviidae, Triatominae) I. Tempo de sucção e repleção de ninfas de 4° e 5° estádios. *Rev Bras Ent* 18: 43-50.
- Carcavallo RU, Jurberg J, Lent H, Galvão C, Steindel M, Pinto CJC 2001. Nova espécie do complexo *oliveirai* (Nova denominação para o complexo *matogrossensis*) (Hemiptera, Reduviidae, Triatominae) do Estado do Rio Grande do Sul, Brasil. *Mem Inst Oswaldo Cruz* 96: 71-79.
- Cortéz MR, Gonçalves TCM 1998. Resistance to starvation of *Triatoma rubrofasciata* (De Geer, 1773) under laboratory conditions (Hemiptera: Reduviidae: Triatominae). *Mem Inst Oswaldo Cruz* 93: 549-554.
- Dias JCP 1965. Observações sobre o comportamento de triatomíneos brasileiros frente ao jejum, em laboratório. *Rev Bras Malariol D Trop* 17: 55-63.
- Dias-Lima AG, Sherlock IA 2001. Períodos de resistência em jejum de algumas espécies de triatomíneos (Hemiptera: Reduviidae: Triatominae). *Rev Soc Med Trop* 34 (Supl. I): 140.
- Dias JCP, Feitosa VR, Ferraz-Filho AN, Rodrigues VLC, Alencar SA, Sessa PA 1989. Fonte alimentar potencial vetorial de *Triatoma vitticeps* (Stal, 1859) com relação à doença de Chagas humana no Estado do Espírito Santo, Brasil. *Mem Inst Oswaldo Cruz* 84 (Suppl. IV): 165-173.
- Ferreira E, Souza PA de, Fonseca Filho M da, Rocha I 1986. Nota sobre a distribuição geográfica do *Triatoma vitticeps* (Stal, 1859) (Hemiptera, Reduviidae) no Estado do Rio de Janeiro, Brasil. *Rev Bras Malariol D Trop* 38: 11-14.
- Galvão C, Jurberg J, Cunha V, Mello RP 1995. Biologia do *Triatoma nitida* Usinger, 1939 em laboratório (Hemiptera: Reduviidae). *Mem Inst Oswaldo Cruz* 90: 657-663.
- Galvão C, Jurberg J, Lent H 1996. Resistência ao jejum de *Triatoma nitida* Usinger, 1939 em laboratório (Hemiptera: Reduviidae). *Mem Inst Oswaldo Cruz* 91: 639-640.
- Gonçalves TCM, Cunha V, Oliveira E, Jurberg J 1997. Alguns aspectos da biologia de *Triatoma pseudomaculata* Corrêa & Espínola 1964, em condições de laboratório (Hemiptera: Reduviidae: Triatominae). *Mem Inst Oswaldo Cruz* 92: 275-280.
- Gonçalves TCM, Oliveira E, Dias LS, D Almeida M, Nogueira WO, Pires FDA 1998. An investigation on the ecology of *Triatoma vitticeps* (Stal, 1859) and its possible role in the transmission of *Trypanosoma cruzi*, in the locality of Triunfo, Santa Maria Madalena municipal district, State of Rio de Janeiro, Brazil. *Mem Inst Oswaldo Cruz* 93: 711-717.
- Gonçalves TCM, Victório VNM, Jurberg J, Cunha V 1988. Biologia do *Triatoma vitticeps* (Stal, 1859) em condições de laboratório (Hemiptera, Reduviidae, Triatominae) I. Ciclo evolutivo. *Mem Inst Oswaldo Cruz* 83: 519-523.
- Gonçalves TCM, Victório VNM, Jurberg J, Cunha V 1989. Biologia do *Triatoma vitticeps* (Stal, 1859) em condições de laboratório (Hemiptera, Reduviidae, Triatominae). II - Resistência ao jejum. *Mem Inst Oswaldo Cruz* 84: 131-134.
- Heitzmann-Fontenelle T 1972. Bionomia de *Triatoma pseudomaculata* Correa & Espínola, 1964, em laboratório. *Mem Inst Butantan* 36: 251-262.
- Lima MM, Rey L, Mello RP de 1991. Lethality of triatomines (Hemiptera: Reduviidae), vectors of Chagas' disease, feeding on blood baits containing synthetic insecticides, under laboratory conditions. *Rev Inst Med Trop São Paulo* 34: 295-301.
- Martinez-Ibarra J A, Katthain-Duchateau G 1999. Biology of *Triatoma pallidipenis* (Stal, 1872) (Hemiptera: Reduviidae: Triatominae) under laboratory conditions. *Mem Inst Oswaldo Cruz* 94: 837-840.
- Moreira CJC, Perlowagora-Szumlewicz A 1997. Attempts to improve xenodiagnosis: comparative test of sensibility using *Rhodnius neglectus*, *Panstrongylus megistus*, *Triatoma vitticeps* and *Triatoma infestans* in endemic areas of Brazil. *Mem Inst Oswaldo Cruz* 92: 91-96.
- Neiva A, Lent H 1936. Notas e comentários sobre triatomíneos. Lista de espécies e sua distribuição geográfica. *Rev Ent Rio de Janeiro* 6: 153-190.
- Pecanha PM, Sessa PA, Silva FJD, Pereira RS, Almeida IM 1983. Apresentação de três casos autóctones do Estado do Espírito Santo. XIX Congresso da Soc Bras Med Trop, Rio de Janeiro, p. 33.
- Pellegrino J 1952. Observações sobre a resistência do *Triatoma infestans* ao jejum. *Rev Bras Biol* 12: 317-320.
- Perlowagora-Szumlewicz A 1969. Estudos sobre a biologia do *Triatoma infestans*, o principal vetor da doença de Chagas no Brasil (importância de algumas de suas características biológicas no planejamento de esquemas de combate a esse vetor). *Rev Bras Malariol D Trop* 21: 117-159.
- Perlowagora-Szumlewicz A, Muller CA, Moreira CJC 1990. Studies in search of a suitable experimental insect model for xenodiagnosis of hosts with Chagas' disease. 4 - The reflection of parasites stock in the responsiveness of different vector species to chronic infection with different *Trypanosoma cruzi* stocks. *Rev Saúde Públ São Paulo* 24: 165-177.
- Pinto AFS, Santos UM, Murad V, Lima FEP, Zaganelli FL, Almeida AZ 1969. Doença de Chagas no Estado do Espírito Santo. V - Primeiro caso de Chagas diagnosticado no município de Alfredo Chaves, E.S. *Rev Soc Bras Med Trop* 3: 53.
- Pinto AFS, Vieira NER, Milanez MC, Lima Pereira FE, Sessa PA 1986. Doença de Chagas autóctone no Espírito Santo: relato de um caso agudo em gestante com transmissão para o feto. *Rev Soc Bras Med Trop* 19 (Supl.): 78.
- Santos UM, Pinto AFS, Zaganelli FL, Almeida AZ, Martins A, Carancho PV, Vieira HJ 1969a. Doença de Chagas no estado do Espírito Santo. II - Inquérito sorológico. E.S. *Rev Soc Bras Med Trop* 3: 51.
- Santos UM, Pinto AFS, Zaganelli FL, Almeida AZ, Martins A, Carancho PV, Vieira HJ 1969b. Doença de Chagas no estado do Espírito Santo. III - Vetores do tripanosoma. *Rev Soc Bras Med Trop* 3: 51-52.

- Santos UM, Pinto AFS, Zaganelli FL, Almeida AZ, Martins A, Carancho PV, Vieira HJ 1969c. Doença de Chagas no estado do Espírito Santo. IV - Primeiro caso de Chagas diagnosticado no município de Rio Novo do Sul, E.S. *Rev Soc Bras Med Trop* 3: 52-53.
- Sessa PA, Carias VRD 1986. Infecção natural em triatomíneos do Espírito Santo por flagelados morfologicamente semelhantes ao *Trypanosoma cruzi*. *Rev Soc Bras Med Trop* 19: 99-100.
- Silva CS, Santos-Mallet JR, Ziccardi M, Gonçalves TCM 2001. Caracterização morfológica e ultraestrutural de amostras de *Trypanosoma cruzi*, isoladas de *Triatoma vitticeps* no estado do Rio de Janeiro. *Rev Soc Med Trop* 34 (Supl. I): 122.
- Silva IG 1985. *Influência da Temperatura na Biologia de 18 Espécies de Triatomíneos (Hemiptera: Reduviidae) e no Xenodiagnóstico*, MSc Thesis, Universidade Federal do Paraná, Curitiba, 169 pp.
- Silveira AC, Alencar TA, Máximo MHC 1983. Sobre o *Triatoma vitticeps* Stal, 1859 (Hemiptera: Reduviidae) no Espírito Santo, Brasil. X Reunião Anual de Pesquisa Básica em Doença de Chagas, 15.
- Silveira AC, Feitosa VR, Borges R 1984. Distribuição de triatomíneos capturados no ambiente domiciliar no período de 1975-1983, Brasil. *Rev Bras Malariol D Trop* 36: 15-312.
- Uribe C 1926. On the biology and the life history of *Rhodnius prolixus* Stal, 1859. *J Parasitol* 13: 129-136.

