

Preliminary data on larval morphology and life cycle of *Nitidula carnaria* (Coleoptera: Nitidulidae), a species of forensic interest

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Datos preliminares de la morfología de la larva y del ciclo biológico de *Nitidula carnaria* (Coleoptera: Nitidulidae), una especie de interés forense

RESUMEN. La especie *Nitidula carnaria* (Schaller) (Coleoptera: Nitidulidae) tiene importancia económica y forense. Por primera vez, se describe la larva madura criada bajo condiciones controladas de laboratorio. Además, se suministra información preliminar del ciclo de vida. Desde la formación de parejas reproductivas hasta obtener el imago, el tiempo registrado fue de $59 \pm 2,4$ días. Esta publicación es el primer reporte de *N. carnaria* en Sudamérica.

PALABRAS CLAVE. Argentina. Coleópteros carroñeros. Entomología forense. Ciclo de vida.

ABSTRACT. The species *Nitidula carnaria* (Schaller) (Coleoptera: Nitidulidae) has economic and forensics importance. The mature larva, reared under controlled conditions of laboratory, was described for the first time. Moreover, preliminary information about the life cycle is provided. The time recorded from the instance mates placed together to the emergence of imago was 59 ± 2.4 days. This publication represents the first record of *N. carnaria* in South America.

KEY WORDS. Argentina. Carrion beetles. Forensic entomology. Life cycle.

INTRODUCTION

Nitidulidae is a family with species distributed in various habitats, that feed on materials of different origin such as flowers, fruits, sap, fungi, stored products, decaying and fermenting plant tissues or dead animal tissue (Parsons, 1943; Hayashi, 1978). This family comprises nine subfamilies, excluding Cybocephalidae/Cybocephalinae, and more than 100 genera. Although there are many species of sap beetles, there is more information on genera or species of Nitidulidae that attack fruit or vegetables, for which they are regarded as pests (Leschen & Marris, 2005).

Some Nitidulidae species were found in ca-

davers of animals in different regions such as South America (Mavárez-Cardozo *et al.*, 2005), North America (William *et al.*, 1992; Watson & Carlton, 2005), Spain (Castillo-Miralbés, 2001) and Japan (Hayashi, 1978), and specimens were also collected from human corpses (Adair & Kondratieff, 1996).

In South America, few works have mentioned the presence of Nitidulidae in cadavers. A study was performed in Colombia under field conditions where entomofauna succession was analyzed (Olaya Másmela, 2001). A review involving records from forensic cases where *Carpophilus hemipterus* Linnaeus was found in the medullar cavities of bones was published in Argentina (Oliva, 2001). Furthermore, *Carmo-*

philus and three unidentified species of Nitidulidae were collected on a pig carcass during a survey of Coleoptera carried out in the South of Brazil (Mise *et al.*, 2007). No records of *Nitidula carnaria* Schaller in South America have been published so far.

Nitidula Fabricius, *Omosita* Erichson and *Carpophilus* Stephens include species that have been found on carrion, and can thus be useful in forensic research (Smith, 1986). In general, those species were associated to advanced stages of decomposition (Payne & King, 1970; Olaya Másmela, 2001; García-Rojo & Honorato, 2006), although some of them were seen in active decay stages (García-Rojo, 2004).

The proper identification of arthropod species of forensic importance is crucial in forensic entomology. Inaccurate determinations invalidate the estimation of the postmortem interval (Byrd & Castner, 2001). There is a lack of knowledge of life cycles and habits of Nitidulidae, particularly of species of forensic relevance. Information on the biology of species involved with dead bodies under certain rearing conditions will be helpful in forensic entomology, allowing the estimation of the postmortem interval and other patterns related with the discipline. The redescription of the adult morphology has been performed by Audisio (1993). The aims of this study were to report for the first time *N. carnaria* in South America, to describe for the first time the morphology of mature larvae and to present preliminary data on the life cycle of *N. carnaria*.

MATERIALS AND METHODS

For the purpose of studying adults and mature larvae of *N. carnaria*, field succession experiments were performed using pig carcasses (Centeno *et al.*, 2002) exposed to direct sunlight and other natural conditions on the ground, in a semi-urban area of Bahía Blanca (38° 43' 2''S - 62° 15' 54''W), Buenos Aires province, Argentina. Three female pigs selected from the same litter were killed by a stab to the heart and put inside a cage made of wood and wire mesh.

Once in the lab a total of 12 *N. carnaria* adults of both sexes were placed inside a plastic container measuring 12 × 8 cm and filled with approximately 3 cm of sand. To provide protection and a source of humidity and water, we introduced a piece of cotton soaked with

distilled water. Semi-dry beef was supplied as a food source. The containers were maintained in a stove at 25 ± 3 °C, 54 ± 2% relative humidity and 12:12 hour D:L cycle. We made daily observations on the culture, which was maintained for three generations. Five mature larvae were examined; more specimens were not extracted so as not to affect the culture, since it had to be used in subsequent studies. Specimens were killed and preserved in 70% ethanol. Terminology for describing structures of larvae was taken from Carlton & Leschen (2007). Examination and photographs of larvae were obtained using a Leica S6D stereoscopic binocular microscope connected to a digital Sony AGO09 camera. Drawings were made with an Olympus CH20 microscope with an Olympus tubular drawing accessory. The length of the period from the day that adults were placed together until the emergence of imago was estimated as the average of the three generations obtained.

RESULTS

Nitidula carnaria (Schaller, 1783)

Nitidula quadripustulata Fabricius, 1792: 72 (lista)

Nitidula variata Stephens, 1830: 72 (lista) (From Mann, 2012)

Description of larvae.

Mature larvae: 4-4.1 mm long and 0.5 mm wide across abdominal fourth segment. Body sub-parallel getting constricted at segment 9, which bears pregomphi and urogomphi. Color: orange-white; head orange; mouthparts darker except maxillary palpi, lighter; antennae light orange; dorsal plates of thoracic and abdominal segments orange, except tenth segment, white; spiracles orange; legs soft orange almost hyaline; ventral surface white. Tegument cracked.

Head: 0.8-1 mm long and 1.1 mm wide; prognathus; surface weakly wrinkled and shining; two stemmata of different size on each side. Behind stemmata three pairs of setae of different length in dorsal region, the shortest in the middle. Antennae three-segmented with some setae. The last antennomere thinner than the first and second, finishing in a seta located just in the middle (Fig. 1). Palpi three-segmented. Labrum with two pairs of setae, the larger located near laterals.

Thorax: Prothorax slightly narrower and

longer than meso- and metathorax. Prothoracic plate occupying most of dorsum. Meso- and metathorax of similar length. Thoracic segments with short setae in the middle and two pairs of large and slender setae on dorsolateral region. The first pair (shortest) located 1/3 at the beginning of the segment and the second (longest) almost 3/4 of it. Ventrally, two pairs of setae with the large one behind the short one.

Legs: Five-segmented with setae and notable separation between segments. The first is the widest, the third is the largest and the fifth is the smallest finishing in a nail (Fig. 2).

Spiracles: Located laterally in mesothorax and metathorax on papillae or tubercles, and on the first eight abdominal segments (Fig. 3).

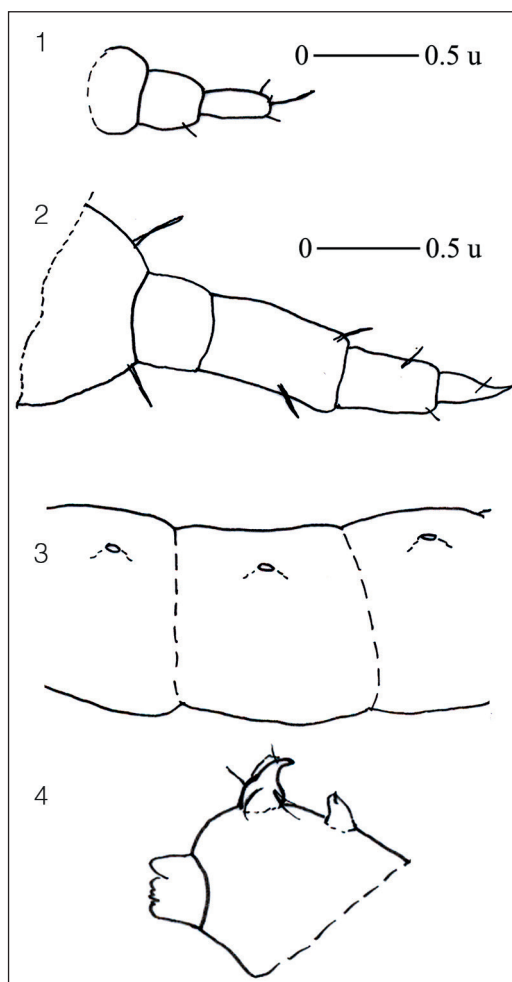
Abdomen: Abdominal segments 1 to 8 similar to meso- and metathorax. Segment 9 narrower than 8. Pregomphi and urogomphi located on sclerotized last plate. Pregomphi smaller than urogomphi with a terminal seta. Urogomphi softly curved, gradually decreasing in width towards apex and with four lateral setae on a stout tubercle and one near the edge (Fig. 4). Tenth segment circular and soft. Sternal areas soft, membranous and with long and short setae with the same disposition as in thoracic segments.

Description of life cycle.

We obtained some preliminary results of the life cycle of *N. carnaria* under controlled conditions in laboratory. Table I shows the length of different stages of the life cycle. After mating, females laid eggs but these could not be observed. Approximately, 13 days after the adults were placed together, the first larval stage appeared. The larvae were found under the semi-dry beef and buried in the sand. It took 35-40 days for pupation to start. No pupal chambers were seen, instead pupae were found between the sand substrate and the piece of cotton. The emergence of the imago occurred 8-10 days after pupation. At first the coloration of the adults was lighter, but it darkened after 24 hours. A total 59 ± 2.4 days elapsed from the moment that adults were placed together to the emergence of imago.

DISCUSSION

Knowledge on the biology of the genus *Nitidula* is scarce. This publication represents the first record of *N. carnaria* as a species of foren-



Figs. 1-4. Some structures of mature larva. 1, antenna; 2, leg; 3, spiracles (schematic); 4, pregomphi, urogomphi and tenth segment (schematic).

sic importance in South America, and provides the first data on its rearing in controlled conditions. This is important because the laboratory rearing of insects collected from a death scene is a key step in the analysis of entomological evidence, and it should not be overlooked (Byrd & Castern, 2001). We could not observe eggs, but they were supposed to have been laid on or under the food source, as is the case for other species of Nitidulidae whose eggs are deposited on or near decomposing plant material (Sanford & Luckmann, 1963). This paper contributes with preliminary biological and forensic information. Finally, further research will be conducted on the biology of *N. carnaria*, in order to determine the exact length of each stage, which will be of

Table I. Length (days) of the life cycle stages (mean \pm standard error) per generation at $25 \pm 3^\circ\text{C}$, $54 \pm 2\%$ relative humidity and 12:12 hour D:L cycle.

	Adults together/ Eclosion	Larval period	Pupal period	Total Length
Generation 1	13	40	8	61
Generation 2	13	40	10	63
Generation 3	12	35	8	55
Mean \pm SE	12.7 \pm 0.3	38.3 \pm 1.7	8.7 \pm 0.7	59.7 \pm 2.4

*Period from the moment adults were placed together until eclosion of eggs.

great value to the further improvement of the PMI estimation and other forensic implications.

ACKNOWLEDGEMENTS

The authors wish to thank Agr. Eng. Barriga-Tuñón for corroborating the taxonomic determination of adult specimens, Dr. Daniel Tanzola for allowing us the use of a camera lucida to obtain morphological drawings and Dra. Mariana Chani-Posse for her helpful comments on the manuscript. This research was funded by Universidad Nacional de Quilmes. Noelia Inés Zanetti thanks CONICET for her fellowship.

LITERATURE CITED

- ADAIR, T. W. & B. C. KONDRATIEFF. 1996. The occurrence of *Nitidula flavomaculata* (Coleoptera: Nitidulidae) on a human corpse. *Entomological News* 107: 233-236.
- AUDISIO, P. 1993. *Fauna d'Italia XXXII. Coleoptera Nitidulidae - Kateretidae*. Calderini, Bologna.
- BYRD, J. H. & J. L. CASTNER. 2001. *Forensic Entomology: The utility of arthropods in legal investigations*. CRC Press, Boca Raton, Florida.
- CARLTON, C. & R. A. B. LESCHEN. 2007. Description of *Soronia complex* (Coleoptera: Nitidulidae: Nitidulinae) larvae of New Zealand with comments on life history and taxonomy. *New Zealand Entomologist* 30: 41-51.
- CASTILLO-MIRALBÉS, M. 2001. Artrópodos presentes en carroña de cerdos en la Comarca de la Litera (Huesca). *Boletín de la Sociedad Entomológica Aragonesa* 28: 133-140.
- CENTENO, N., M. MALDONADO & A. OLIVA. 2002. Seasonal patterns of arthropods occurring on sheltered and unsheltered pig carcasses in Buenos Aires Province (Argentina). *Forensic Science International* 126: 63-70.
- GARCÍA-ROJO, A. M. 2004. Estudio de la sucesión de insectos en cadáveres en Alcalá De Henares (Comunidad Autónoma de Madrid) utilizando cerdos domésticos como modelos animales. *Boletín de la Sociedad Entomológica Aragonesa* 34: 263-269.
- GARCÍA-ROJO, A. M. & L. HONORATO. 2006. La Entomología Forense y la práctica policial en España: estimación del intervalo post-mortem en un cadáver hallado en el interior de una arqueta en la comunidad de Madrid. *Ciencia Forense* 8: 57-62.
- HAYASHI, N. 1978. A contribution to the knowledge of the larvae of Nitidulidae occurring in Japan (Coleoptera: Cucujoidea). *Insecta Matsumurana* (N. S.) 14: 1-97.
- LESCHEN, R. A. B. & J. W. M. MARRIS. 2005. *Carpophilus* (Coleoptera: Nitidulidae) of New Zealand with notes on Australian species. Landcare Research Contract Report: LCO405/153: 1-40.
- MANN, D. J. 2012. Family Scarabaeidae Latreille, 1802. *En: Duff, A. G. (ed.), Checklist of Beetles of the British Isles. 2nd edition*, Pemberley Books, Iver, pp. 55-58.
- MAVÁREZ-CARDOZO, M. G., A. I. ESPINA DE FERREIRA, F. A. BARRIOS-FERRER & J. L. FERREIRA-PAZ. 2005. La Entomología forense y el Neotrópico. *Cuadernos de Medicina Forense* 11: 23-33.
- MISE, K. M., L. M. ALMEIDA & M. O. MOURA. 2007. Levantamento da fauna de Coleoptera que habita a carcaca de *Sus scrofa* L., em Curitiba, Parana. *Revista Brasileira de Entomologia* 51: 358-368.
- OLAYA MÁSMELA, L. A. 2001. Entomofauna sucesional en el cadáver de un cánido en condiciones de campo en la Universidad del Valle (Cali-Colombia). *Cuadernos de Medicina Forense* 23: 5-14.
- OLIVA, A. 2001. Insects of forensic significance in Argentina. *Forensic Science International* 120: 145-154.
- PARSONS, C. T. 1943. A revision of Nearctic Nitidulidae (Coleoptera). *Bulletin of the Museum of Comparative Zoology* 92: 119-278.
- PAYNE, J. A. & E. W. KING. 1970. Coleoptera associated with pig carrion. *Entomologist's Monthly Magazine* 105: 224-232.
- SANFORD, J. W. & W. H. LUCKMAN. 1963. Observations on the biology and control of the dusky sap beetle in Illinois. *Proceedings of the North-Central Branch of the Entomological Society of America* 18: 39-43.
- SCHALLER, J. G. 1783. Neue Insekten. *Abhandlungen der Hallischen Naturforschenden Gesellschaft* 1: 217-332.
- SMITH, K. G. V. 1986. *A Manual of Forensic Entomology*. London British Museum of Natural History, Cornell Univ. Press, London.
- WATSON, E. J. & C. E. CARLTON. 2005. Insect succession and decomposition of wildlife carcasses during fall and winter in Louisiana. *Journal of Medical Entomology* 42: 193-203.
- WILLIAMS, R. N., J. L. BLACKMER, D. S. RICHMOND & M. S. ELLIS. 1992. Nitidulidae (Coleoptera) diversity in three natural preserves in Portage County, Ohio. *Ohio Journal of Science* 92: 82-87.