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New Cases of Gynandromorphism in *Xylocopa* Latreille, 1802 (Hymenoptera: Apidae)

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ABSTRACT

Gynandromorphism is the most common case of sexual anomaly reported in bees and is characterized by individuals that show male and female traits simultaneously in the body. Gynandromorphic cases have been reported for 140 species of bees, an underestimated number comparing to the twenty thousand bee species described nowadays. Here we describe and illustrate the first case of a gynandromorphic Xylocopa darwini *Cockerell, 1926 and the fourth case of* Xylocopa varipuncta *Patton, 1879. The specimens show a mixed form of gynandromorphism with predominantly female features and with all its male traits concentrated in one side of the body, right side in* X. darwini *and left side in* X. varipuncta. *The gynanders of* X. darwini *and* X. varipuncta *were collected on Isabela Island (Galapagos – Ecuador) and Riverside (California – USA), and were deposited in Smithsonian Collection and California Academy of Sciences, respectively. Including this work, eighteen cases of gynandromorphism were reported to* Xylocopa *and twelve were recorded from* Neoxylocopa *subgenus.*

Key-Words: Carpenter bee; Gynandromorphy; *Neoxylocopa;* New World; Xylocopini.

INTRODUCTION

Sexual anomalies were first reported for bees in 1857, when Sichel described a gynandromorphic form of *Bombus lapidarius* (Linnaeus, 1758) (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Silveira *et al.,* 2012). Gynandromorphs are individuals that have male and female traits simultaneously in the body, and the causes of this anomaly are discussed in relation to problems in fertilization, chromosome damages or loss, and association with symbionts and parasites (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Narita *et al.,* 2010; Camargo & Gonçalves, 2013; Ugajin *et al.,* 2016).

Two types of classification are in use to distinguish the distribution of the gynandromorphic features in the body (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Narita *et al.,* 2010). The oldest classification was proposed by Dalla Torre & Friese (1899), who considered four types of gynandromorphs in nature: mixed, lateral, transverse and antero-posterior (Wcislo *et al.,* 2004; Gonzalez, 2004; Lucia *et al.,* 2009; Silveira *et al.,* 2012; Camargo & Gonçalves, 2013; Lucia & Gonzalez, 2013; Alvarez *et al.,* 2014; Coelho *et al.,*

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2016). More recently, only three categories started to be used by some mellitologists: bilateral, transverse and mosaic (Michez *et al.,* 2009; Giangarelli & Sofia, 2011; Vivallo, 2015; Ugajin *et al.,* 2016).

To date, gynandromorphism has been reported for approximately 140 species of bees in 35 genera (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Fateryga *et al.,* 2011; Hinojosa-Díaz *et al.,* 2012; Lucia *et al.,* 2012; Silveira *et al.,* 2012; Camargo & Gonçalves, 2013; Lucia & Gonzalez, 2013; Alvarez *et al.,* 2014; Lucia *et al.,* 2015; Suzuki *et al.,* 2015; Vivallo, 2015; Coelho *et al.,* 2016). These are representatives of six families but especially Megachilidae (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012; Lucia & Gonzalez, 2013; Alvarez *et al.,* 2014; Coelho *et al.,* 2016). In Apidae, most cases were recorded in *Bombus* Latreille, 1802 and *Xylocopa* Latreille, 1802, among the largest bees in the family (Michener, 2007; Michez *et al.,* 2009). Lucia & Gonzalez (2013) provided a revision of gynandromorphism in the carpenter bees genus *Xylocopa.* They listed 14 records of gynandromorphs from 12 species in five subgenera of *Xylocopa,* three of them from the Old World and two from the New World. Recently, two new cases of gynanders were described, the second case for *Xylocopa frontalis* (Olivier, 1789) (Vivallo, 2015) and the first case for *Xylocopa augusti* Lepeletier, 1841 (Lucia *et al.,* 2015). In short, ten of the 16 cases of *Xylocopa* gynandromorphs belong to species of *Neoxylocopa,* a neotropical subgenus that shows an evident sexual dimorphism in its representatives, males being yellowish to ferruginous and females blackish to dark brownish (Silveira *et al.,* 2002; Michener, 2007).

Here we described and illustrated the first case of a gynandromorphic *Xylocopa darwini* Cockerell, 1926, an endemic bee and the most important pollinator species of the Galapagos Islands (Jaramillo *et al.,* 2010; Chamorro *et al.,* 2012; Ascher & Pickering, 2014; Vargas *et al.,* 2015); and the fourth case of *Xylocopa varipuncta* Patton, 1879, one of the few number of large carpenter bees recorded from Mexico and United States (Moure, 2012; Ascher & Pickering, 2014).

MATERIAL AND METHODS

The bees were found in dry preservation in the entomological collections of Smithsonian and California Academy of Sciences. The external morphology of *X. darwini* was studied using a Zeiss Stemi SV8 stereomicroscope and the pictures were taken with a Canon EOS-60D camera attached to a Microptics ML-1000 Digital Imaging System at the American Museum of Natural History, NY – USA. *Xylocopa varipuncta* morphology was studied using a Leica M125 stereomicroscope in the Insect Systematics Lab (UFMG). The pictures of this specimen were taken with a Leica DFC 295 camera attached to a Leica M205C stereomicroscope and images were assembled using LAS 3.8 software at the Arachnology lab (UFMG).

The morphological terminology employed here follows Hurd & Moure (1963) and Michener (2007). The abbreviations T, S and F are used for terga, sterna, and flagellomeres, respectively. The measurements are expressed in millimeters. The body length was taken in dorsal view, with the length of metasoma measured separately and added to the length of the head plus mesosoma; the head length was the distance between the vertex and the apex of the clypeus; the head width was the maximum distance between the outer margins of the compound eyes; the width of the mesosoma was the distance between the inner margins of the tegulae; the metasomal width was the maximum width of T2 and the wing length was measured between the base of the tegula and the apex of the wings.

The terminalia were removed from bees after 24-48 hours in a relaxing chamber. They were extracted using fine pins and forceps and were cleared for half or one hour in heated KOH. After that, they were rinsed in distilled water and preserved in glycerin.

RESULTS

Xylocopa (Neoxylocopa) darwini Cockerell, 1926 (Fig. 1)

Description (Fig. 1A-D): Body length 22.1, head length 5.0; head width 7.0, mesosoma width 5.5, metasoma width 9.2; right forewing length 19.8; left forewing length 20.3. ***Head*** (Fig. 1B): entirely female-like. ***Mesosoma*** (Fig. 1A,C,D): female-like on the left side, with some male traits on right side. Integument predominantly black with black, plumose and long pubescence as in normal females; mesoscutum integument black with yellow hairs on right lateral margin; scutellum on right side brownish to ferruginous with yellow hairs; metanotum black as in females; propodeum vertical and black as in females but propodeal triangle brownish red, smooth and shiny on right side, black, sculptured and dull on left side; right lateral of mesosoma, behind insertion of foreleg, dark, covered by yellow, plumose and long hairs as in normal males; in ventral view, right

side behind foreleg, mostly male-like with black integument covered by yellow, plumose and very long hairs, but triangular area with sparse, simple, black hairs on middle of mesepisternum, before middle leg; legs on left side and foreleg on right side, female-like; middle right leg male-like; hind right leg with black trochanter and femur as in males, trochanter almost glabrous, shining on inner surface, with expanded apical margin; femur inner surface with tuft of yellow hairs on middle of very strong depression; hind right tibia and basitarsus female-like on outer surface, with black integument and black hairs, male-like on inner surface, with ferruginous integument and yellow hairs; inner surface of tibia with irregular margin as in normal male; remaining tarsomeres and claws on right hind leg missing; right tegula ferruginous, humeral plate almost white, as in males; right forewing male-like, remaining three wings female-like. ***Metasoma*** (Fig. 1A,C,D): mostly female-like with male traits on right side. Six exposed terga and sterna as in female; terga asymmetric, left side longer than right side, left halves more round, right halves more straight; left side of tergites female-like, right side mixed but predominantly male-like: integument of T1 and T2 predominantly black with asymmetric ferruginous areas covered by dense, yellow hairs on right side; T1 fringes black as in females; T2 fringes yellow as in males; right side of T3 more related to males than T1 and T2, almost ferruginous covered by yellow, simple and long hairs; integument of T4 ferruginous covered by yellow hairs, except for its right lateral with black integument and small ferruginous area, hair fringe with mixture of long, yellow, simple hairs and very long, black, simple and branched hairs; integument of T5 dorsally ferruginous basally and darker apically, completely black on right lateral with dense fringe formed by long, simple and plumose, black hairs; left side of T6 female-like with half of pygidial plate and right side male-like with completely ferruginous integument covered by very long, yellow and ferruginous, simple and plumose hairs; right side of T6, with slightly emarginate apical margin medially; metasomal sterna female-like but very asymmetric in shape; integument black to dark-brown, reddish on apical margins, covered by black and simple hairs; all segments with conspicuous mid-longitudinal carina, typical of female *X. (Neoxylocopa);* male features of sterna only noticeable in S2, as irregular ferruginous areas and some yellow hairs on right lateral margin. ***Terminalia:*** female sting apparatus; sting apparatus

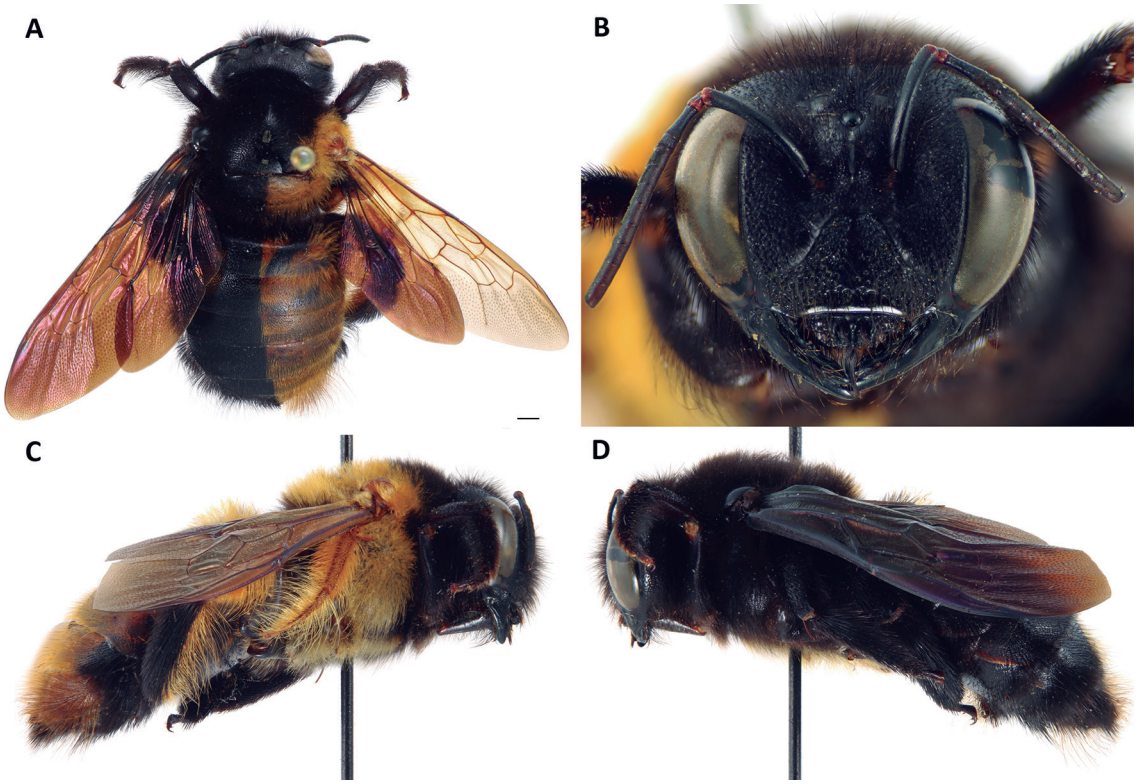

FIGURE 1: Gynandromorph of *Xylocopa (Neoxylocopa) darwini* Cockerell, 1926: **(A)** Dorsal habitus; **(B)** Head in frontal view; **(C)** Right lateral habitus; **(D)** Left lateral habitus. Scale bars = 1 mm. (Photos: Hadel Go).

much similar to that of normal *X. darwini* female without recognizable male structure, just the plate corresponding to S7 on right side differs from that on left side.

Examined material: one gynandromorph, one male and three females collected in May 9th, 1964 by D.Q. Cavagnaro on North Coast of Isabella Island, Galapagos, Ecuador. The bees are deposited in the Smithsonian Collection – Washington DC, USA.

Xylocopa (Neoxylocopa) varipuncta Patton, 1879 (Fig. 2)

Description (Fig. 2A-D): Body length 23.1; head length 4.5; head width 6.6; mesosoma width 5.9; metasoma width 10; right forewing length 20.6; left forewing length 20.4. ***Head*** (Fig. 2B): mixed, female-like on right and male on left side, except for some mixed characters on left side of galea and mandible, one black area and hairs on left paraocular area and frons. ***Mesosoma*** (Fig. 2A,C,D): predominantly female with some male traits; one vertical line of ferruginous integument with yellow hairs on left middle of mesoscutum; sparse yellow hairs on left margin of mesoscutum; ferruginous integument and yellow hairs on left apical margin of scutellum, left middle part of metanotum, left basal half of pronotum; some yellow hairs on lateral and left ventral surface of mesepisternum; left anterior leg with mixed femur, male tibia and tarsal segments; left middle leg predominantly female-like with some yellow integument and hairs on ventral surface of femur, tibia and tarsal segments; left hind leg with tufts of short yellow hairs on ventral surface of tarsal segments 3-5 and claws; left tegula predominantly ferruginous with some black areas on lateral and posterior margins; some yellow hairs on basal outer margin of anterior left wing; forewings predominantly female-like but left slightly short than right. ***Metasoma*** (Fig. 2C-D): predominantly female-like with few features of male on left side; T1 left half male-like except for some black integument and hairs laterally; T2 and T3 female-like with some sparse ferruginous areas covered by yellow hairs; T6 with ferruginous integument and yellow hairs on middle half of left side, pygidial plate just on right side; left side of S2 with a big yellowish white area on middle part. ***Terminalia:*** female sting apparatus.

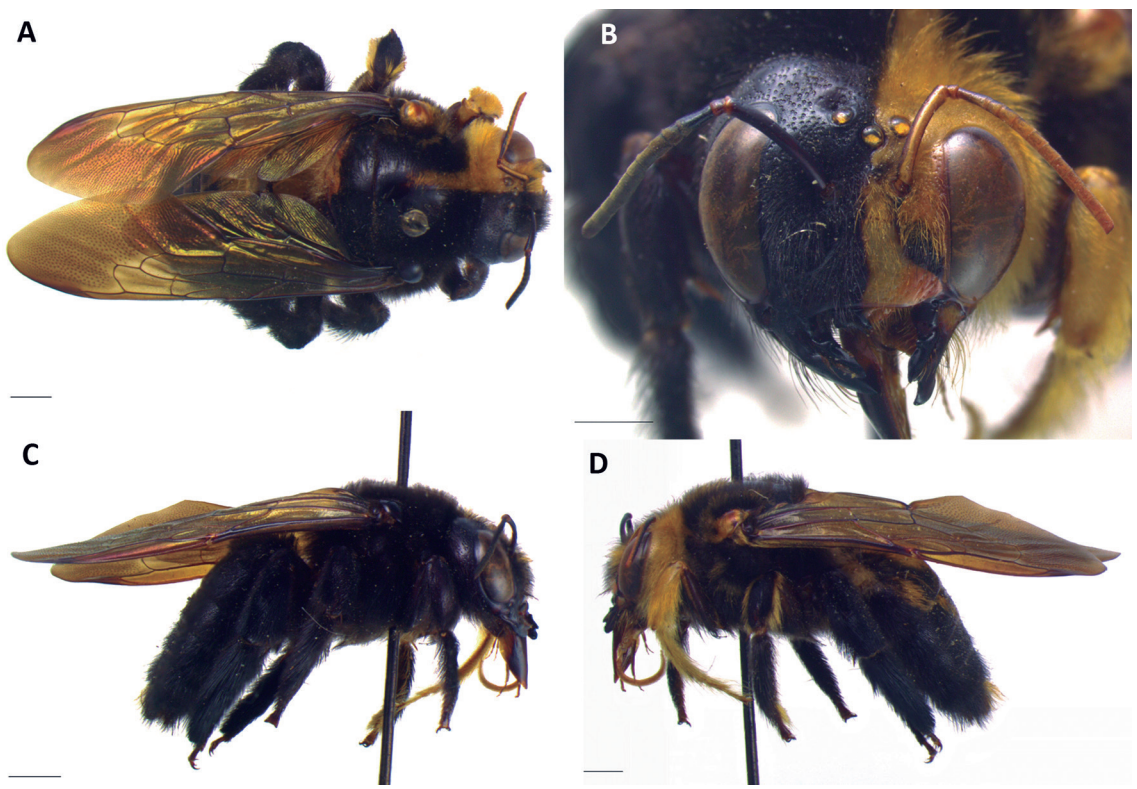

FIGURE 2: Gynandromorph of *Xylocopa (Neoxylocopa) varipuncta* Patton, 1879: **(A)** Dorsal habitus; **(B)** Head in frontal view; **(C)** Right lateral habitus; **(D)** Left lateral habitus. Scale bars = 1 mm. (Photos: Paula C. Zama).

Examined material: one gynandromorph, two males and two females collected in August, 1929 by L. Peirce Coy on Riverside, California, USA. The bees are deposited in the Entomological Collection of California Academy of Sciences – San Francisco, CA, USA.

DISCUSSION

New cases of gynandromorphism in bees have been reported since the beginning of the last century (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012). In despite of this, the number of gynandromorphs for bees is still around 180 specimens (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Fateryga *et al.,* 2011; Hinojosa-Díaz *et al.,* 2012; Lucia *et al.,* 2012; Silveira *et al.,* 2012; Camargo & Gonçalves, 2013; Lucia & Gonzalez, 2013; Alvarez *et al.,* 2014; Lucia *et al.,* 2015; Suzuki *et al.,* 2015; Vivallo, 2015; Coelho *et al.,* 2016; Ugajin *et al.,* 2016), an underestimated number comparing to total number of described bee species nowadays, something around 20.000 (Danforth *et al.,* 2013). As the majority of the records, gynanders of *X. darwini* and *X. varipuncta* were found in the museums collections, in dry preservation, and the descriptions were based only in external morphology (Bonnet, 1952; Carcasson, 1965; Lucia *et al.,* 2009; Lucia *et al.,* 2012; Camargo & Gonçalves, 2013; Lucia & Gonzalez, 2013; Alvarez *et al.,* 2014; Lucia *et al.,* 2015; Vivallo, 2015; Coelho *et al.,* 2016). Just a few observations about biology and behavior of gynandromorphic forms have been made (Gordh & Gulmahamad, 1975; Wcislo *et al.,* 2004; Michez *et al.,* 2009; Giangarelli & Sofia, 2011; Hinojosa-Díaz *et al.,* 2012; Silveira *et al.,* 2012; Ugajin *et al.,* 2016).

The majority of the bees show some level of sexual dimorphism and in some bees, like *X. (Neoxylocopa),* they show very evident dimorphic characteristics (Silveira *et al.,* 2002; Michener 2007). The difference of color on integument and pilosity of males and females of *X. (Neoxylocopa)* is probably the more easily perceivable character to diagnose gynandromorphic cases. The specimens herein described increase the cases of gynandromorphism to 18 in the genus and to 12 in the subgenus, with 14 different species in *Xylocopa* and eight in *X. (Neoxylocopa).*

Both gynandromorphic bees described here are predominantly-female and have male traits concentrated in one side of their body. Taking this into consideration, they can be treated as partially bilateral, except for female head in *X. darwini* and for intermingled characteristics of both sexes in the right side in *X. darwini* and left side in *X. varipuncta.* As in many other gynandromorphic bees mentioned in the literature, the distribution of opposite sexual characters is not completely symmetrical (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012; Lucia & Gonzalez, 2013).

The asymmetry of the features and the use of two classifications can take the definition of the categories of gynandromorphy controversial. According to Dalla Torre & Friese (1899), the cases described here can be classified as lateral gynanders, considering the fact that all male features are concentrated just in one side of the body. But, according to Michez *et al.* (2009), recognizing the asymmetrical features of males, the gynandromorphic forms of *Xylocopa* can be allocated in mosaic/mixed category. Comparing just *Xylocopa* records of gynandromorphy, controversial classifications can be found for three other cases: *Xylocopa (Koptortosoma) nigrita* (Fabricius, 1775) – gynander described by Carcasson (1965); *Xylocopa (Neoxylocopa) mendozana* Enderlein, 1913 – gynander by Enderlein (1913); and *Xylocopa (Schonnherria) micans* Lepeletier, 1841 – described by Maidl (1912). *Xylocopa nigrita* and *X. micans* were classified as bilateral using the old classification of Dalla Torre & Friese (1899) (Maidl, 1912; Carcasson, 1965; Wcislo *et al.,* 2004; Lucia & Gonzalez, 2013), and as transverse using Michez *et al.* (2009). *Xylocopa mendozana* is classified as mixed with the oldest classification and as bilateral with the newer (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Lucia & Gonzalez, 2013). The case of *X. micans* seems more controversial, Maidl (1912) affirms that its head is female-like on the left and male-like on the right side, but inverted on the mesosoma and metasoma. Actually it seems to be a mixed/mosaic-kind of gynandromorph, since it has a heterogeneous distribution of traits in different parts of the body, and not bilateral or transverse as diagnosed in the previous works.

Besides the lack of consensus among the categories of gynandromorphs, other problems can bring some discussion. The high concentration of the gynandromorphic cases in the bees with Holartic distribution seem to be more related to the lack of information in other regions than with a genetic problem with these lineages (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012; Lucia & Gonzalez, 2013; Suzuki *et al.,* 2015). The same apparently to be happening with the great number of gynandromorphs occurring in some genera, as *Megachile* and *Andrena.* Probably, the cause is more related to a sampling artifact (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012; Lucia & Gonzalez, 2013; Suzuki

et al., 2015). Despite these misunderstandings and the fact the gynandromorphism is currently recorded in almost all bee families (Wcislo *et al.,* 2004; Michez *et al.,* 2009; Hinojosa-Díaz *et al.,* 2012), further studies about gynandromorphy are needed. Works including morphology, behavior, ontogenetic or causes of gynandromorphy could be very important to increase the knowledge and the discussion about sexual anomalies, especially in insect.

RESUMO

Ginandromorfismo é o caso mais comum de anomalia sexual registrado para abelhas e destaca-se por indivíduos que apresentam características simultâneas de machos e de fêmeas no corpo. Casos de ginandromorfos foram registrados para aproximadamente 140 espécies de abelhas, um número subestimado comparado às vinte mil espécies de abelhas descritas atualmente. No presente trabalho, o primeiro registro de ginandromorfismo para Xylocopa darwini *Cockerell, 1926 e o quarto caso para* Xylocopa varipuncta *Patton, 1879 são descritos e ilustrados. Os espécimes apresentam a forma mista de ginandromorfo com características de fêmeas predominantes e as de machos concentradas em uma lateral do corpo, lado direito em* X. darwini *e esquerdo em* X. varipuncta. *Os ginandromorfos de* X. darwini *and* X. varipuncta *foram coletados na Ilha Isabela (Galápagos – Ecuador) e em Riverside (California – USA), e estavam depositados nas coleções entomológicas do Smithsonian e da Academia de Ciências da Califórnia, respectivamente. Incluindo as abelhas do presente trabalho, dezoito casos de ginandromorfismo estão registrados para abelhas do gênero* Xylocopa *e doze para o subgênero* Neoxylocopa.

Palavras-Chave: Abelhas carpinteiras; Ginandromorfismo; *Neoxylocopa;* Novo Mundo; Xylocopini.

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