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Sociedad Argentina para el Estudio de los Mamíferos

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**Use of artificial materials in nest construction
by *Gracilinanus microtarsus* (Wagner, 1842)**

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Citación: CONSTANTINO, W. D., S. M. DE AZEVEDO, J. D. R. ARIAS, G. MACHADO, I. M. SOUZA, L. O. DRUMMOND, & C. BRAGA. 2024. Use of artificial materials in nest construction by *Gracilinanus microtarsus* (Wagner, 1842). *Notas sobre Mamíferos Sudamericanos* 6:e24.06.2.

ABSTRACT

Nests are structures employed by animals for resting, reproduction, and offspring care. The materials involved in the construction of nests vary depending on many aspects (species, environment, availability of resources, among others). Also, there are species that use natural cavities or artificial materials to construct their nests. In this note, we describe a case of a nest fashioned from a PET bottle by an individual of *Gracilinanus microtarsus*, highlighting the opportunistic behavior of this marsupial. This is a unique record that reveals the potential use of artificial nests for ecological studies for arboreal marsupials such as *G. microtarsus*.

Keywords: Atlantic Forest, Didelphidae, marsupials, nidification, small mammals

RESUMO - Uso de materiais artificiais na construção de ninho por *Gracilinanus microtarsus* (Wagner 1842)

Os ninhos são estruturas utilizadas pelos animais para repouso, reprodução e cuidados com a prole. Os materiais envolvidos na construção dos ninhos variam de acordo com diversos aspectos (espécie, ambiente, disponibilidade de recursos, entre outros). Além disso, há espécies que utilizam cavidades naturais ou materiais artificiais como base para a construção de seus ninhos. Nesta nota, descrevemos um caso de um ninho confeccionado a partir de uma garrafa PET por um indivíduo de *Gracilinanus microtarsus*, evidenciando o comportamento oportunista deste marsupial. Este é um registro único que revela o potencial de utilização de ninhos artificiais para estudos ecológicos de marsupiais arborícolas como *G. microtarsus*.

Palavras-chave: Didelphidae, Floresta Atlântica, marsupiais, nidificação, pequenos mamíferos

Nests are commonly defined as sites where animals spend certain moments of the day engaging in activities such as rest, reproduction, or offspring care (Deeming

Recibido el 28 de noviembre de 2023. Aceptado el 30 de abril de 2024. Editor asociado Gabriel Martin.



2023). The construction of nests can vary widely in many aspects, such as size, location, and construction material. This variation primarily depends on the species that build them and environmental conditions, including the availability of materials and suitable locations for construction (Fustec & Cormier 2007; Biddle et al. 2017). While the utility of nests may vary among different taxonomic groups, they play a fundamental role in animal survival and reproductive success (Downing & Jeanne 1986).

Small mammals often make use of pre-existing nests built by other animals such as birds or other mammals (Jordan et al. 2023). Nevertheless, some small mammal species build their own nests, which not only provide a resting place but also offer protection against predators and other external threats during rest or other activities (Hamilton 1982; Voss & Jansa 2021). These structures also play an important role in the protection and development of offspring (Voss & Jansa 2021) and serve as a shelter against severe weather conditions (e.g., extreme heat, storms) (Aquino & Encarnacion 1986; Kimble 1997).

Didelphid marsupials have developed unique reproductive traits, such as their short gestation period, the birth of offspring in an embryonic state, and the completion of the development of the offspring outside the uterus and attached to the mother's teats, whether exposed to the environment (e.g., *Gracilinanus*, *Metachirus*), or protected within a marsupium (e.g., *Didelphis*, *Philander*), providing a safe and nourishing environment for the young (Neto 2013). Nonetheless, didelphids usually still need a nest for their own protection, and to protect the young from predators and adverse weather conditions. Thus, although young didelphid offspring do not remain in the nest as is the case with eutherians of similar size (Ross 2001; Wolff 2007; Deeming 2023), the nest is still necessary as a daily shelter for adults, and to shelter infants from the period in which they detach from their mother's teat until the moment they are weaned.

The choice of nest construction sites and utilization are important aspects of a species' ecology and are primarily influenced by the availability of suitable locations and resources (Smith et al. 2007; Lira et al. 2017). Understanding the types and patterns of nest utilization, as well as the choice of sites and material for nest construction by didelphid marsupials are crucial for comprehending their ecology and natural history.

In undisturbed forests, some didelphid species prefer the use of natural cavities as the basis for nest construction (Tortato & Campbell-Thompson 2006; Pires et al. 2010). Within these cavities, the animals use primarily dry leaves as material for the nest structure (Tubelis 2000). For example, the four-eyed opossum, *Philander quica* (Temminck, 1824), nests in underground burrows, tree cavities, tangles of lianas, and crowns of palm trees, forming natural shelters through the accumulation of dry leaves (Lira et al. 2017). Palms also play an important role in the nesting behavior of the woolly mouse opossum, *Marmosa paraguayana* (Tate, 1931), which builds its nests at sites where leaf stalks insert into the trunk, accumulating dry leaves for better protection against the light (Emmons & Feer 1997; Junior & Chiarello 2005). According to Pires et al. (2010), the Brazilian gracile mouse opossum, *Gracilinanus microtarsus* (Wagner 1842), also utilizes tree cavities, with the potential to take ad-



vantage of abandoned bird nests, like the mottled piculet (*Picumnus nebulosus*) (Cáceres & Pichorim 2003).

In this note, we describe the nest of a female Brazilian gracile mouse opossum *Gracilinanus microtarsus* (body length: 87 mm; tail length: 139 mm; weight: 20 g) (Fig. 1) in an artificial cavity at the Reserva Biológica União, hereafter REBIO União (latitude -22.447970; longitude -42.056270; DATUM WGS84), in the State of Rio de Janeiro, Brazil. This reserve is covered mainly by submontane (47%) and lowland (29%) rainforest (ICMBIO 2008). The area where the nest was found is covered by a lowland rainforest with a high local predominance of bamboos. The individual was identified as *G. microtarsus* following external and cranial diagnostic characters from Creighton and Gardner (2008). This is the only species of the genus occurring in the lowlands of Rio de Janeiro (Bergallo et al. 2004; Rocha et al. 2004; Mayer et al. 2023).

The nest was built inside a PET plastic bottle resting against a wooden stake of a pitfall fence, positioned approximately 1 meter above the ground. This 2-liter bottle was cut approximately in its half (measuring 171 mm in height x 104 mm in diameter, measurements taken in the laboratory), and used for water removal from the pitfall traps. The cut bottle is left leaning on the wooden stake of the pitfall fence between field campaigns. Due to the COVID-19 pandemic, the pitfall line remained inactive for 14 months since December 2019. When we restarted the pitfall monitoring in February 2022, we found the bottle, and the enclosed animal and plant materials were visible through the bottle (Fig. 2A).

The individual was euthanized and deposited as skin and skeleton in the Mammal collection of the Institute of Biodiversity and Sustainability at the Federal University of Rio de Janeiro (NUPEM/UFRJ) under the number NPM2059. Collection permit was granted by ICMBio (Number: 64807-9).

The nest was primarily composed of entangled vines and dry leaves (Fig. 2B). Dry leaves were also the materials used by individuals of the same species reported by Tubelis (2000). As shown in Figure 2A, the PET bottle opening was oriented downward, without any support beneath the nest, so the use of lianas was probably due to the lack of support at the bottom of the PET bottle. This configuration demonstrates the complexity in the construction and organization of the nest structure, entangling the materials in a way that the vines impede the falling of leaves from inside the nest (Fig. 2B).

The use of artificial materials highlights the opportunistic character of *G. microtarsus* nesting behavior, especially considering that this behavior was recorded in a preserved and legally protected forest reserve, where the species would be able to easily find natural cavities for nest construction. Pires et al. (2010) also found an individual of *G. microtarsus* in a nest made of dry leaves inside a non-triggered Sherman trap. The opportunistic nest-building habit of *G. microtarsus* is further reinforced by previous records of nesting not only in natural tree hollows, but also in bird nests (Cáceres & Pichorim 2003), artificial nests made from bamboo trees (Tubelis 2000; Loretto & Vieira 2011), and even within a sherman trap that failed to trigger (Pires et al. 2010).

The nests built by species of didelphid marsupials consist predominantly of dry



leaves (Deeming, 2023), although occasionally other materials may be used. Lira et al. (2017) suggested that individuals of *Philander frenatus* found in underground burrows, tree cavities, vine tangles and palm canopies commonly use dry leaves, forming natural shelters in the latter two types of nest. In a study conducted in captivity, Unger (1982) described the short tail mouse opossum *Monodelphis domestica* (Wagner, 1842) building elaborated nests composed of interwoven strips of paper to form a multi-layered structure that resembled leaf-built nests of other species in natural environments. As far as we know, there are no previous studies on didelphids nests describing a complex structure of intertwined vines made by the animal like the one used as a base to support the leaves in the nest described in the present study. Most of the studies described nests built in burrows or inside artificial structures that already have a base to support the animal and the other materials used (Tubelis 2000; Lira et al. 2017).

The use of a simple artificial material as PET bottle by the animal raise the possibility of using this type of material to study marsupial nidification. Artificial nests may be an important tool for studying American marsupials, especially as a complementary method to study scansorial and arboreal species (Loretto 2005). Future studies could evaluate the use of plastic bottles in trees as artificial nests for small mammals, since it is cost-effective, involves simple logistics for transportation to remote study areas, and facilitates replicability for testing and comparisons.



Figure 1. Individual of *Gracilinanus microtarsus* found in an artificial nest in the Reserva Biológica União. Source: Leandro de Oliveira Drummond.



Figure 2. A) Artificial nest built by a female of *G. microtarsus* in a cut PET bottle at the Reserva Biológica União (REBIO União), Rio de Janeiro, Brazil; B) and plant materials entangled used by this female to construct toe nest at the bottle.

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