

New species of cave-restricted *Hyaella* (Crustacea: Amphipoda) for Brazil, with the first records from the Amazon

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Abstract

New species of the freshwater amphipod genus *Hyaella* have been discovered in groundwater habitats across Brazil. Three of these species were found in ferruginous caves (two in the state of Pará and one in Minas Gerais) while a fourth species was identified in a cave in Rio Grande do Sul. The discovery of the *Hyaella* species in Pará is particularly noteworthy, as it represents the first record of this genus in the Amazon biome, where no *Hyaella* species had been documented until now.

Résumé

De nouvelles espèces d'amphipodes d'eau douce du genre *Hyaella* ont été découvertes dans des habitats souterrains à travers le Brésil. Trois de ces espèces ont été trouvées dans des grottes ferrugineuses, deux dans l'État du Pará et une dans celui du Minas Gerais. Une quatrième espèce a été identifiée dans une grotte de l'État du Rio Grande do Sul. La découverte des espèces du Pará est particulièrement significative, car elle constitue le premier enregistrement de ce genre dans le biome amazonien, où les espèces de *Hyaella* n'avaient jusqu'à présent jamais été documentées.

Resumo

Novas espécies do anfípodo de água doce do gênero *Hyaella* foram descobertos para águas subterrâneas do Brasil. Três espécies foram coletadas em cavernas ferruginosas, sendo duas delas no estado do Pará, e uma no estado de Minas Gerais. A quarta espécie foi coletada em uma caverna do Rio Grande do Sul. As espécies do Pará representam um importante registro no bioma amazônico, onde até o momento não havia ocorrências de espécies deste gênero.

1. Introduction

The genus *Hyaella* comprises over 100 described species, with more than 80 endemic to South America (Marrón-Becerra & Hermoso-Salazar, 2023; Marrón-Becerra et al., 2023; Peralta & Verónica, 2023; Reis et al., 2023; Tomikawa et al., 2023). While most research has focused on Argentina and southern Brazil, recent discoveries in Mexico, Chile, and Paraguay suggest that this apparent distribution bias may be influenced by differences in sampling efforts and the proximity of research institutions (Reis et al., 2020, 2023; Mussini et al., 2024).

Hyaella is a genus of freshwater amphipods found in both epigeal and hypogean environments, including caves, groundwater, and shallow subterranean habitats (Rodrigues & Bueno, 2012; Bastos-Pereira et al., 2018; Peralta & Verónica, 2023). In Brazil, six species have been recorded in subterranean habitats: *H. caeca* Pereira, 1989; *H. spelaea* Bueno & Cardoso, 2011; *H. epikarstica* Rodrigues, Bueno & Ferreira, 2014; *H. sumida* Penoni & Bueno, 2025; and *H. ceciliae* Penoni & Bueno,

2025 from São Paulo; *H. formosa* Cardoso & Araujo, 2014, from Paraná; *H. veredae* Cardoso & Bueno, 2014, from Minas Gerais; and *H. imbya* Rodrigues & Bueno, 2012, from a wetland in the municipality of Roque Gonzales, Rio Grande do Sul (Pereira, 1989; Cardoso et al., 2011, 2014; Rodrigues & Bueno, 2012; Penoni et al., 2025a,b). All these species are considered stygobitic.

This study significantly expands the known distribution of *Hyaella* in Brazil, providing new records for the Amazon region. Two new species were discovered in the state of Pará, collected from iron ore caves in the Serra dos Carajás region. Additionally, a third species was found in an iron ore cave in Minas Gerais, while a fourth species was identified in a carbonate cave in Rio Grande do Sul. These findings highlight the importance of subterranean environments in harboring previously undocumented *Hyaella* diversity.

2. Materials and methods

The newly discovered *Hyaella* species were preserved in 70% ethanol and deposited in the Collection of Subterranean Invertebrates of Lavras (ISLA) at the Federal University of Lavras (UFLA), Brazil. Specimens were dissected and mounted on slides using Hoyer's medium

for detailed morphological examination. Analyses were conducted using a ZEISS Scope A1 optical microscope equipped with an Axio 105 Color camera, with species descriptions based on photographs of the paratypes. Habitus images of both males and females were captured

using a ZEISS Axio ZoomV16 stereomicroscope coupled with an Axio Cam 506 Color camera. Setae nomenclature followed the classification system proposed by Zimmer et al. (2009).

Study area

Pará state (PA)

The species *Hyaella* n. sp. 1 (Fig. 1C, 2A) (Fig. 1C) and *Hyaella* n. sp. 2 (Fig. 1D, 2B) were collected from ferruginous caves located in Serra do Tarzan and Serra Sul, respectively, in the state of Pará, Brazil. These areas belong to the Serra dos Carajás Speleological Unit (Fig. 1A,B), which contains an extensive network of over 2,000 ferruginous caves and is renowned for its vast mineral reserves, including iron, nickel, copper, and gold (Piló et al., 2015).

The region has a tropical climate, with temperatures ranging from 23°C to 25°C and an average annual precipitation of approximately 2,400 mm. The dry season extends from June to September, with monthly rainfall ranging from 10 to 90 mm, while the rainy season lasts from October to April, with precipitation levels between 160 and 340 mm per month (Sahoo et al., 2016).

Serra do Tarzan is located within two federally protected conservation units: the Campos Ferruginosos National Park and the Carajás National Forest (FLONA Carajás). Consequently, this geomorphological area remains relatively pristine compared to other parts of Carajás, which are more affected by mining activities.

In contrast, Serra Sul, despite being within FLONA Carajás, hosts the

world's largest open-pit mining operation, producing up to 90 million tons of iron ore annually (Matlaba et al., 2017) (Fig. 1C, D).

Minas Gerais state (MG)

Hyaella n. sp. 3 (Fig. 1E,F, 2C) was identified in an iron ore cave within the Iron Quadrangle region of Minas Gerais, a major hub for iron ore and gold extraction in Brazil. This region represents a transitional zone between two global biodiversity hotspots—the Brazilian Savannah (Cerrado) and the Atlantic Forest (Myers et al., 2000). The landscape is a mosaic of native vegetation interspersed with areas impacted by human activities, including mining and agriculture (Gomes et al., 2019). The local climate is characterized by dry winters and wet summers, with monthly average temperatures ranging from 16.5°C to 23.2°C (Gomes et al., 2019).

Rio Grande do Sul state

Hyaella n. sp. 4 was collected in Serra Geral (Fig. 2D), within caves formed in basaltic rock formations resulting from ancient volcanic flows (Reginato et al., 2013). Although cave research in this region remains in its early stages, at least 14 species of *Hyaella* have already been described, highlighting the region's high potential for amphipod diversity (Talhaferro et al., 2021).

The regional climate is characterized by temperatures ranging from 15°C to 18°C. Peak rainfall occurs at the end of spring (September–October, 160–180 mm/month) and during summer (January, ~160 mm/month) (Buriol et al., 2007).

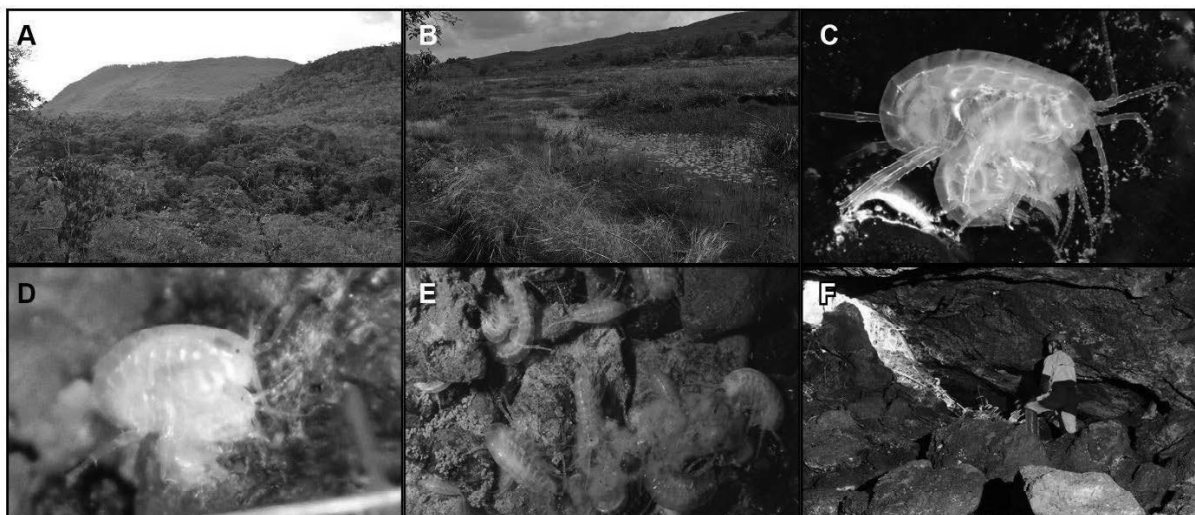


Figure 1: A, Carajás Mountain Range; B, altitude lake in Carajás Mountain Range; C, couple in amplexus of *Hyaella* n. sp. 1; D, couple in amplexus of *Hyaella* n. sp. 2; E, *Hyaella* n. sp. 3; F, Cave entrance where *Hyaella* n. sp. 3 was collected.

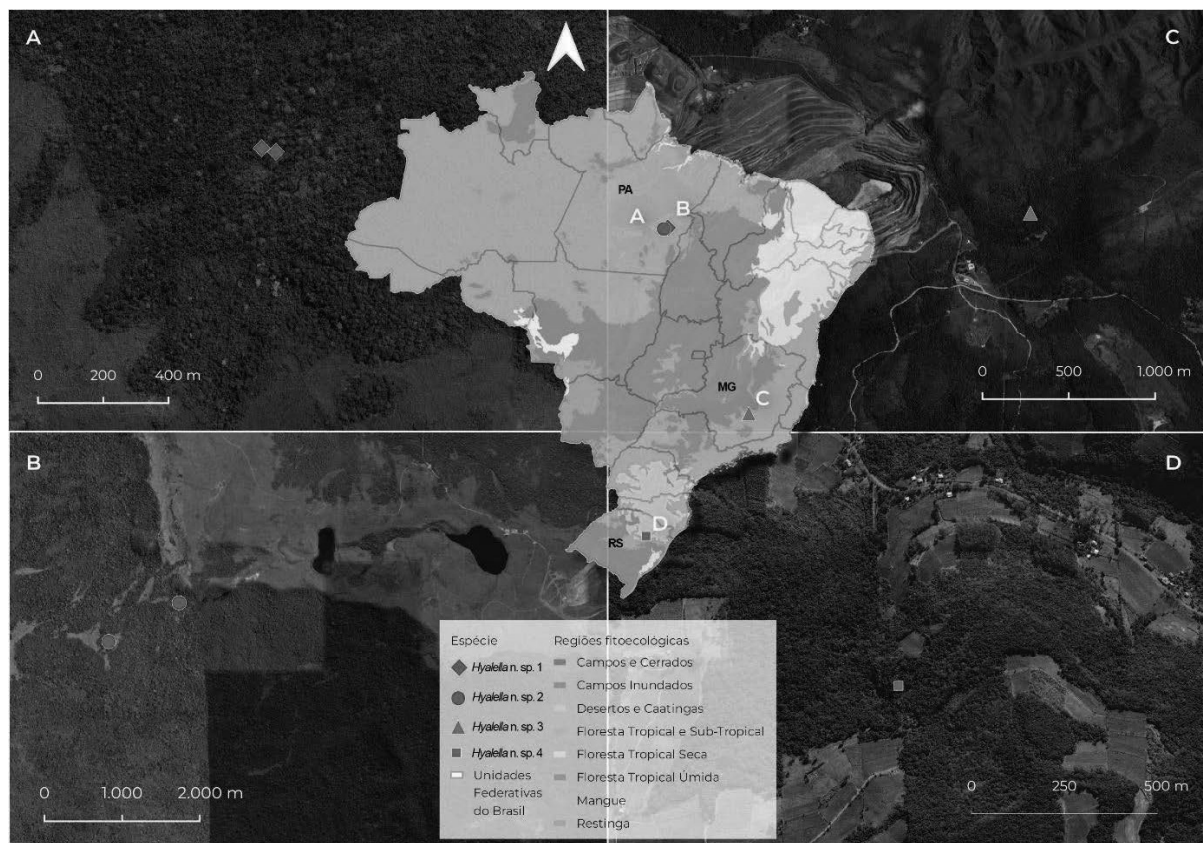


Figure 2: Distribution map of the new *Hyalella* species in Brazil. Abbreviations: PA, Pará state; MG, Minas Gerais state; RS, Rio Grande do Sul state.

3. Results

Hyalella n. sp. 1 (Figs 3A, 4A) differs from the other species by the absence of eyes, by the length of the antennae 1 and 2, by the shape of male gnathopod and by the number and type of setae on telson. Comparing with the other stygobite *Hyalella* species from Brazil, *Hyalella* n. sp. 1 resembles *H. caeca* and *H. spelaea* by the absence of a curved seta on uropod 1, however differs by the number of setae on telson, with two simple setae on the middle plus one cuspidate seta on each side, while both *H. caeca* and *H. spelaea* have only two simple setae apically. By the similar length of the antennae 1 and 2, *Hyalella* n. sp. 1 resembles *H. veredae*, however, *H. veredae* presents one curved seta on uropod 1.

Hyalella n. sp. 2 (Figs 3B, C, 4B) differs from *Hyalella* n. sp. 1 by the reduced number of articles on the antennae (antenna 1 with 6 and antenna 2 with 8 versus antenna 1 with 14 and antenna 2 with 11 in *Hyalella* n. sp. 1), and the length of the antennae with antenna 1 shorter than antenna 2 versus antenna 1 and antenna 2 with similar length in *Hyalella* n. sp. 1. The telson differs with two cuspidate setae versus two simple and two cuspidate setae on *Hyalella* n. sp. 1.

Hyalella n. sp. 3 (Figs 3D, 4C) resembles *H. troglolugia*, that occurs in the nearby municipality of Nova Lima, in Minas Gerais state. Both species

lack the curved setae on uropod 1, and present 2 cuspidate setae on telson. However, *Hyalella* n. sp. 3 differs from *H. troglolugia*, by the presence of reduced eyes, that are absent in *H. troglolugia*; and the number of articles on antennae, 6-7 on antenna 1 and 11 on antenna 2; while the number of articles on *H. troglolugia* are 9-11 and 9-12 respectively; the number of setae in gnathopod 1 propodus, 7 in *Hyalella* n. sp. 3 and *Hyalella* n. sp. 4 in *H. troglolugia*; and the shape of gnathopod 2 propodus, slender in *Hyalella* n. sp. 3 (1.5 times longer than wide).

Hyalella n. sp. 4 (Figs 3E, 4D) resembles *H. imbya* in its troglomorphic characteristics, such as the absence of eyes and body pigmentation. It also shares the relative development of antenna 1, which is longer and has more articles compared to antenna 2, as well as the presence of two simple setae on telson. However, the two species differ in the number of articles in their antennae: antenna 1 has 15-16 articles in *Hyalella* n. sp. 4 and 18-23 articles in *H. imbya*, while antenna 2 has 10 articles in *Hyalella* n. sp. 4 and 14-16 in *H. imbya*. Additionally, an important distinguishing feature is the presence of curved seta on uropod 1 in *H. imbya*, which is absent in *Hyalella* n. sp. 4.

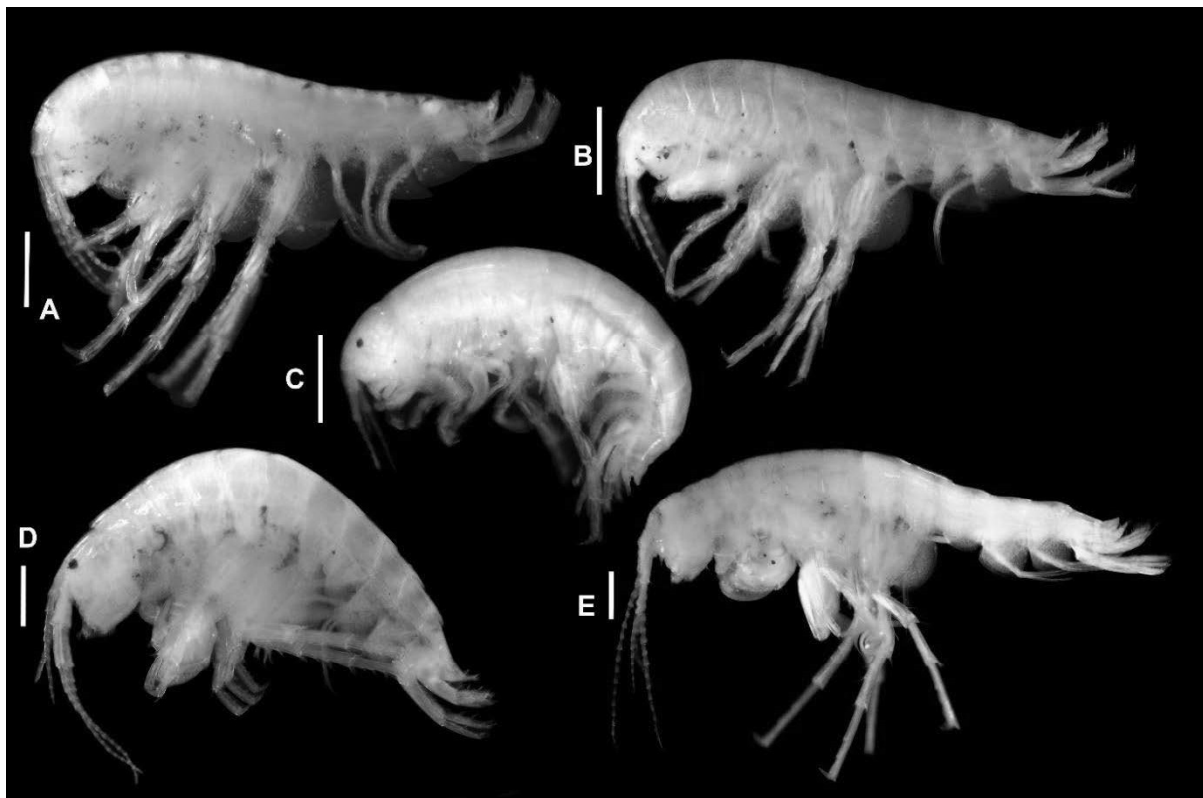


Figure 3: Subterranean species of *Hyaella*. A, male of *Hyaella* n. sp. 1; B, male of *Hyaella* n. sp. 2; C, female of *Hyaella* n. sp. 2; D, male of *Hyaella* n. sp. 3; E, male of *Hyaella* n. sp. 4. Scale bars: 0.5 mm.

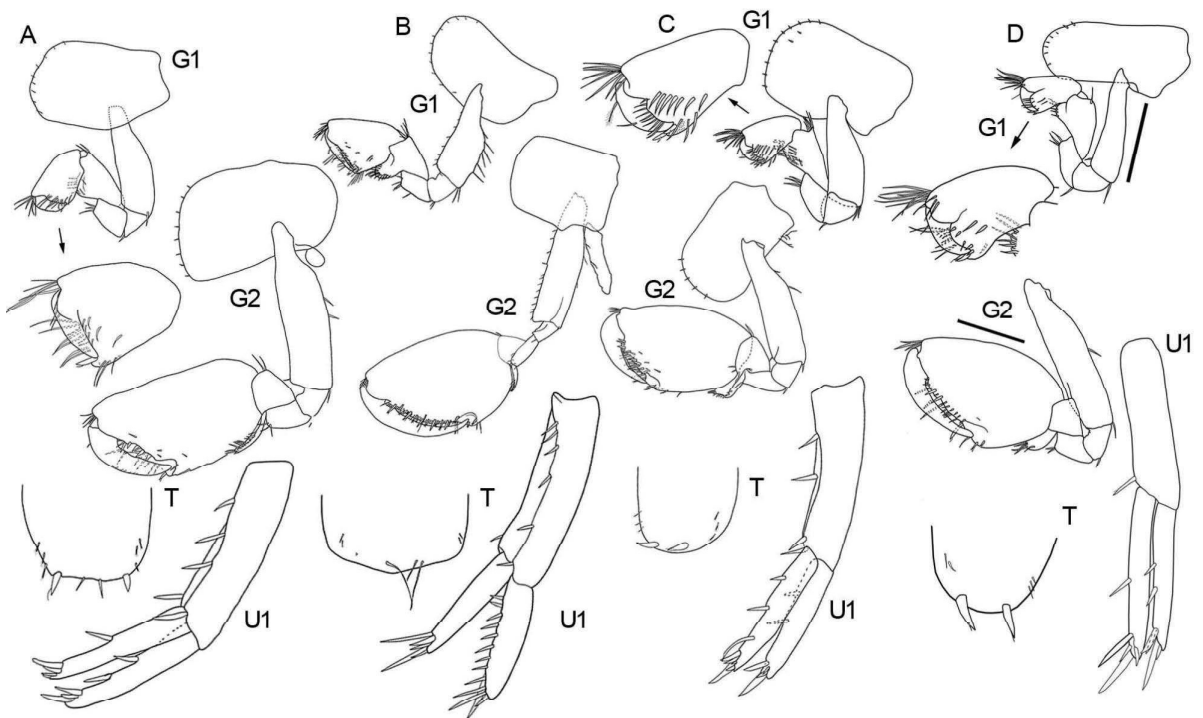


Figure 4: Subterranean species of *Hyaella*. A, male of *Hyaella* n. sp. 1; B, male of *Hyaella* n. sp. 2; C, male of *Hyaella* n. sp. 3; D, male of *Hyaella* n. sp. 2. Scale bars: 0.2 mm. Abbreviations: G1: gnathopod 1; G2: gnathopod 2; U1: uropod 1; T: telson.

4. Discussion

Four new species are recorded in caves from Brazil. Three species are recorded in iron ore caves (Fig. 2).

Hyaella n. sp. 1 and *Hyaella* n. sp. 2 represent two important records for the genus in the Amazon region, and the record in the state of Pará. Providing important information on the distribution of the genus in poorly sampled locations, whether due to difficulty of access or sampling effort. *Hyaella* n. sp. 3 represents the second stygobite species recorded for Minas Gerais state, the first being *H. veredae*. However, in Minas Gerais, two epigeal species present troglomorphic traits such as the variations in the eye size, *H. montana* and *H. troglófugia*, which indicate may suggest that such species inhabit interstitial subterranean spaces (Rodrigues et al., 2017; Bastos-Pereira et al., 2018).

Hyaella n. sp. 4 represents the second troglomorphic species recor-

ded for the Rio Grande do Sul state, the first being the hypohelminorheic species, *H. imbya*.

The newly described species are considered stygobitic, displaying troglomorphic characteristics such as depigmentation and elongated appendages, particularly antenna 1 relative to antenna 2. Eye morphology varies across species: in *Hyaella* n. sp. 1 and *Hyaella* n. sp. 4, the eyes are absent; in *Hyaella* n. sp. 3, they are reduced. Interestingly, in *Hyaella* n. sp. 2, some specimens lack eyes entirely, a variation also observed in certain individuals of *H. veredae* (Zeppon et al., 2021). New records of *H. veredae* were discovered and it was observed a variation in the eyes which may reflect the degree and duration of isolation experienced by each population in subterranean environments (Zeppon et al., 2021).

5. Conclusion

New species are continuously being discovered, highlighting the importance of studying caves. Many of these species are restricted and endemic to specific locations, and their formal description not only advances scientific knowledge but also contributes to the preservation of the

cave where they occur and the entire associated community. Therefore, conducting fauna inventories in underexplored areas is essential, as these discoveries reveal the significant potential for identifying new species.

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