

NOTES ON GEOGRAPHIC DISTRIBUTION

Check List 18 (1): 91–102 https://doi.org/10.15560/18.1.91



The exotic snail *Rumina decollata* (Linnaeus, 1758) (Gastropoda, Achatinidae, Rumininae) in Argentina: new records, range extension, and areas of origin of Argentine populations

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Abstract

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The decollate land snail *Rumina decollata* (Linnaeus, 1758), which is native to the Mediterranean region, has been introduced to several countries worldwide. We report here three new records of *R. decollata* in Argentina: two from Córdoba Province and the first one from Misiones Province, which also constitutes the northernmost record for the country and extends its northeastern geographic range. Species-specific identification was achieved based on anatomical, conchological, and molecular information. DNA data showed that different haplotypes are present in Argentina, which originated from distinct source areas within the native range.

Keywords

Decollate snail, COI, invasive species, mitochondrial haplotypes, reproductive system, South America

Academic editor: Carl Christensen | Received 15 September 2021 | Accepted 10 January 2022 | Published 19 January 2022

Citation: Rau AI, Beltramino AB, Serniotti EN, Pizá J, Caffetti JD, Vogler RE (2022) The exotic snail *Rumina decollata* (Linnaeus, 1758) (Gastropoda, Achatinidae, Rumininae) in Argentina: new records, range extension, and areas of origin of Argentine populations. Check List 18 (1): 91–102. https://doi.org/10.15560/18.1.91

Introduction

Rumina Risso, 1826 is a genus of hermaphroditic land snails characterized by decollate shells, facultative self-fertilization, and omnivorous feeding habits (Batts 1957;

Selander and Kaufman 1973; Dundee 1986; Örstan 2008; Prévot et al. 2014). Four morphospecies are currently recognized within the genus: *R. decollata* (Linnaeus, 1758),

R. saharica Pallary, 1901, R. paivae (Lowe, 1860), and R. iamonae Quintana Cardona, 2017 (Mienis 2008; Prévot et al. 2015; Quintana Cardona 2017). Descriptions of the morphospecies were based on morphological features of the shell and genital system for R. decollata, R. saharica, and R. iamonae, and shell size for R. paivae (Bank and Gittenberger 1993; Carr 2002; Mienis 2008; Prévot et al. 2015; Quintana Cardona 2017). Molecular studies have suggested that Rumina comprises at least seven phylogenetic species, in which several molecular operational taxonomic units (MOTUs) have been delimited, one corresponding to R. saharica and six others under R. decollata (namely MOTUs A-F), with DNA data rejecting the species status of R. paivae (Prévot et al. 2013, 2014). In addition, based on genital and shell characters, Prévot et al. (2015) found no significant differentiation among individuals of R. decollata from the different MOTUs, also suggesting that R. paivae represents a large phenotype of R. decollata. To date, no molecular data are available for R. iamonae, which is currently recognized as a species based on morpho-anatomical characters.

Rumina decollata is native to the Mediterranean region, inhabiting the Iberian Peninsula, southern France, Italy, western Balkans, and northern Africa (Prévot et al. 2014). The species is now widely distributed and established in other parts of the world, e.g., eastern Asia, southern North America, some of the Caribbean islands, and southern South America (Selander and Kaufman 1973; Cowie 2001; Bar-Zeev and Mienis 2007; Prévot et al. 2014 and references therein; Naranjo-García and Castillo-Rodríguez 2017; Ishida 2020). As R. decollata is known to prey on other mollusks, it has been promoted as a biocontrol agent for the brown garden snail Cornu aspersum (Müller, 1774) and other helicid snails in citrus groves of California, USA, where it does not cause damage to citrus, despite the fact that it may itself become a plant pest (Johnson 1900; Cowie 2001; Tupen and Roth 2001; Sakovich 2002; Herbert 2010). In this sense, it has been reported as an agricultural and horticultural pest in non-native areas, being documented to cause damages in plantations of chayote, onion, and cucumber in Mexico (Correa Sandoval 1993), beetroot, carrot, chard, lettuce, mango, napa cabbage, and sorghum in Cuba (Matamoros 2014a, 2014b), and strawberry crops in Brazil (Landal et al. 2019). In relation to animal and human health, R. decollata can serve as an intermediate host of the trematode of rodents Brachylaima ruminae Mas-Coma & Montoliu, 1986 (Mas-Coma and Montoliu 1986). In addition, it has been also recorded as an intermediate host of Aelurostrongylus abstrusus (Railliet, 1898), a lungworm that causes bronchopneumonia in wild and domestic cats (Cardillo et al. 2014, 2018; Valente et al. 2017), and as a paratenic host of *Toxocara cati* (Schrank, 1788), a common intestinal worm of felids causing toxocariasis, in which humans can act as accidental hosts (Cardillo et al. 2016). Also, R. decollata has been recorded as an intermediate host of Angiostrongylus malaysiensis Bhaibulaya & Cross, 1971, a nematode parasite of various rat species, and a potential zoonotic pathogen of human angiostrongyliasis (Sawabe and Makiya 1994, 1995; Yong et al. 2016; Watthanakulpanich et al. 2021).

Rumina decollata has fusiform and elongated shells of moderate size (typically up to 4 cm in height). Shells are generally light grey to dark brown, exhibiting moderately deep sutures, and weak growth lines on the external surface (Herbert 2010; Prévot et al. 2015; Landal et al. 2019). The apex is missing (decollate) in adult shells, thus appearing broken, with last 3-6 whorls remaining (usually 4) (Herbert 2010; Prévot et al. 2015). Based on the body and foot, five color morphs have been characterized for R. decollata (Prévot et al. 2015), two of them previously referred to as the light and the dark morphs by Selander and Hudson (1976); the former characterized by a light grey body with a black medio-dorsal line and a pale yellowish foot, and the latter by a black body and a dull olive-grey foot (Prévot et al. 2015). This dark phenotype was found to be distinctive of specimens of MOTU A defined by Prévot et al. (2013), to which all worldwide introduced populations belong (Prévot et al. 2014). Anatomically, the reproductive system is broadly characterized by an elongated penis with the distal portion containing transverse lamellae, which separate into prominent papillae towards the center and proximal part of the penis. The vagina is wide, short, exhibiting inner longitudinal crenulated lamellae (Prévot et al. 2015), and the bursa copulatrix is a well-developed sac with a short narrow duct (Schileyko 1999).

In South America, R. decollata has been introduced and established in Argentina, Brazil, and Uruguay (Miquel et al. 1995; Scarabino 2003; De Francesco and Lagiglia 2007; Oliveira and Abreu 2013). Its presence in Chile was mentioned by Rumi et al. (2010) without further information other than the general statement as occurring in that country, and not providing precise data (Araya 2015; Darrigran et al. 2020). In Argentina, specimens of R. decollata were first reported by Miquel (1988) in two localities from Buenos Aires Province, close to Buenos Aires city, gradually spreading and becoming established into new areas of that province (Virgillito 2012; Bogan et al. 2021). In the mid-2000s, the species was recorded in the semiarid region of La Pampa and Mendoza provinces in central-western Argentina (De Francesco and Lagiglia 2007). Subsequently, Reyna and Gordillo (2018) reported its occurrence in Córdoba city, Córdoba Province, in central Argentina, and more recently Pérez and Tissot (2021) reported its presence for Chubut and Río Negro provinces in the Patagonian region.

In this study, we report three new records for *R. decollata* in Argentina: two from Córdoba Province and the first one from Misiones Province, which also constitutes the northernmost record for the species in Argentina and extends its northeastern geographic range in the country. The anatomical data obtained here represent the first information of this species from Argentina.

Phylogenetic analyses based on partial cytochrome c oxidase subunit I (COI) sequences were also carried out to explore its geographical origins. New data presented here are expected to help determine invasion pathways of this snail in South America.

Methods

Individuals and shells of *Rumina decollata* were collected by handpicking in urban domestic gardens from the Argentine cities of Posadas, Misiones Province, and Río Tercero and Villa María, Córdoba Province. Geographic coordinates were recorded with GPS Garmin eTrex Legend. Living specimens were relaxed in water with menthol crystals for about 8 h, subsequently immersed in hot water (80 °C), and finally fixed and preserved in 96% ethanol. For the morpho-anatomical studies, soft parts of adult individuals were separated from the shell. Voucher material was deposited in the Malacological Collection of the Instituto de Biología Subtropical (IBS-Ma), CONICET – Universidad Nacional de Misiones, Misiones Province, Argentina.

Shells were cleaned in water using an ultrasonic bath Codyson CD4810 for 5-10 min, air dried, and then photographed in apertural, dorsal, and lateral views. Specimens were identified based on external shell morphology and genital anatomy following Wille (1915), Pilsbry (1946), Carr (2002), Mienis (2008), Herbert (2010), and Prévot et al. (2015). Six shell measurements were taken from specimens of each locality according to availability following Prévot et al. (2015): shell height (SH), shell width (SW), aperture transversal length (AT), aperture width (AW), height of body whorl (FwH), and number of whorls (WN). The soft parts of four adult individuals (IBS-Ma 383-2, 383-5, 565-3, 565-4) were dissected using a Zeiss Stemi SV6 stereomicroscope with a camera lucida device. Terms proximal and distal refers to the position of an organ or part of an organ in relation to the gamete flow from ovotestis (proximal) to genital pore (distal) (Cuezzo et al. 2018).

DNA was isolated from muscle tissue of six individuals (IBS-Ma 383-1, 383-2, 383-3, 565-2, 565-3, 565-4) using a CTAB protocol (Beltramino et al. 2018). Partial sequences of the mitochondrial COI marker were amplified by PCR using the universal primers LCO1490 and HCO2198 (Folmer et al. 1994). PCR master mix and thermal profile were conducted as in Guzmán et al. (2018). The PCR products were purified with an AccuPrep PCR Purification Kit (Bioneer, Korea), and bidirectionally sequenced by Macrogen Inc. (Seoul, Korea). Sequences were trimmed to remove the primers, and consensus sequences were assembled using BIOEDIT 7.2.5 (Hall 1999). Consensus sequences were compared with reference sequences from GenBank through BLASTn algorithm (Altschul et al. 1990), in order to confirm the morphology-based identification of the specimens.

Phylogenetic analyses using maximum likelihood (ML), and Bayesian inference (BI) were also conducted

to make inferences about the source location. We used the COI sequences of almost all specimens from various locations represented in Prévot et al. (2013, 2014), with Subulina octona (Bruguière, 1789) as the outgroup. The ML analysis was conducted using MEGA X (Kumar et al. 2018) with the Nearest-Neighbor Interchange branch swapping algorithm. Optimal nucleotide substitution model (GTR+G) was selected using the jModelTest 2.1.10 program (Darriba et al. 2012) by means of the corrected Akaike Information Criterion. Nodal support values were computed by bootstrapping with 1000 replicates (Felsenstein 1985). The BI was performed in MrBayes 3.2.6 (Ronquist et al. 2012) with the same substitution model used in the ML analysis. Two runs were performed simultaneously with four Markov chains for 2 million generations, sampling every 200 generations. The first 1001 samples of each run were removed as burn-in, with the remaining 18000 trees used to estimate posterior probabilities. In addition, in order to better visualize the relationships among haplotypes of introduced populations, a dataset containing the COI sequences of MOTU A specimens was used to construct a median-joining network with the Network 10.2.0.0 software (Bandelt et al. 1999). DNA sequences were deposited in GenBank under the accession numbers MZ605490 to MZ605495.

Results

Taxonomic account

Gastropoda Cuvier, 1795 Stylommatophora A. Schmidt, 1855 Achatinidae Swainson, 1840 Rumininae Wenz, 1923 Rumina Risso, 1826

Rumina decollata (Linnaeus, 1758)

New records. ARGENTINA — Misiones • Posadas; 27.3740°S, 055.8896°W; 17.VII.2019; E.N. Serniotti leg.; 3 spec., 23 dry shells (22 juveniles), IBS-Ma 565 — Córdoba • Río Tercero; 32.1822°S, 064.1194°W; 15.I.2019; R.E. Vogler, A.A. Beltramino leg.; 19 spec. (2 juveniles), IBS-Ma 383 • Villa María; 32.4179°S, 063.2262°W; 01.I.2019; E.N. Serniotti, E.A. Giovannini, E.E.A. Mellano leg.; 14 dry shells (11 broken), IBS-Ma 388 (Table 1, Fig. 1).

Identification. The snails were firstly identified as belonging to the genus *Rumina* by their typically decollate shells. Examined shells are rather thick, elongated, mid-brown in color, having 4–6½ slightly convex whorls, with surface sculptured mostly by irregular growth lines. The aperture is ovate and with the outer lip simple (Fig. 2). Shell measurements (Table 2) fitted well with those reported for *R. decollata* in previous studies (e.g., Prévot et al. 2015). Specimens examined showed the black body and the dull olive-gray foot typical of the dark morph of *R. decollata* (Fig. 2). In addition, species-specific

Table 1. Reports of *Rumina decollata* obtained in the present study and literature records for the species in Argentina. CFA-IN: Invertebrate Collection at Fundación de Historia Natural Félix de Azara, Buenos Aires, Argentina. CNP-INV: General Invertebrate Collection of Instituto de Biología de Organismos Marinos (IBIOMAR), CONICET-CENPAT, Puerto Madryn, Chubut. IBS-Ma: Malacological Collection of the Instituto de Biología Subtropical, CONICET-Universidad Nacional de Misiones, Posadas, Argentina. MACN-In: Malacological Collection of the Museo Argentino de Ciencias Naturales Bernardino Rivadavia, Buenos Aires, Argentina. MLP-Ma: Malacological Collection at the Museo de La Plata, La Plata, Argentina. MMHNSR: Museo Municipal de Historia Natural de San Rafael, San Rafael, Argentina. *Error of locations estimated following the point-radius method of Wieczorek et al. (2004) when georeferencing data were absent in the consulted sources.

ID	Location	Voucher (collection year)	Latitude	Longitude	Error (km)*	References
1	Posadas, Misiones Province	IBS-Ma 565 (2019)	27.3740°S	055.8896°W	-	Present study
2	Río Tercero, Córdoba Province	IBS-Ma 383 (2019)	32.1822°S	064.1194°W	-	Present study
3	Villa María, Córdoba Province	IBS-Ma 388 (2019)	32.4179°S	063.2262°W	-	Present study
4	Córdoba, Córdoba Province	Collection not reported (2017)	31.4171°S	064.1978°W	10	Reyna and Gordillo 2018
5	Santa Rosa, La Pampa Province	MMHNSR number not reported (2003 and 2004)	36.6209°S	064.2912°W	5	De Francesco and Lagiglia 2007
6	Mendoza, Mendoza Province	MMHNSR number not reported (2005)	32.8903°S	068.8462°W	4	De Francesco and Lagiglia 2007
7	Lomas de La Lucila, Vicente López, Buenos Aires Province	MACN-In 32920 (1988)	34.4984°S	058.4838°W	1	Miquel 1988
8	Carapachay, Vicente López, Buenos Aires Province	CFA-IN-5586 (date not reported)	34.5266°S	058.5370°W	1	Bogan et al. 2021
9	Villa Ballester, General San Martín, Buenos Aires Province	MLP-Ma 12012 (2005)	34.5528°S	058.5624°W	2.5	Virgillito 2012
10	Reserva Ecológica Costanera Sur, Ciudad Autónoma de Buenos Aires	MACN-In 36261 (1998); MACN-In 37755 (1999)	34.6121°S	058.3516°W	2	Virgillito 2012
11	Ciudad Autónoma de Buenos Aires	MACN-In 38393 (2009); MACN-In 39396 (2011)	34.6215°S	058.4627°W	9	Virgillito 2012
12	Avellaneda, Buenos Aires Province	MACN-In 32921 (1988); MACN-In 37757 (1994)	34.6789°S	058.3446°W	4.4	Miquel 1988; Virgillito 2012
13	Glew, Almirante Brown, Buenos Aires Province	MACN-In 36103 (1992)	34.8868°S	058.3809°W	3.2	Virgillito 2012
14	La Plata, Buenos Aires Province	MLP-Ma 7083 (2002); MLP-Ma 12544 (date not reported)	34.9216°S	057.9519°W	3.5	Virgillito 2012
15	Villa Regina, Río Negro Province	CNP-INV 3860 (2021)	39.0974°S	067.0832°W	-	Pérez and Tissot 2021
16	Puerto Madryn, Chubut Province	CNP-INV 3856 (2021); CNP-INV 3857 (2021)	42.7888°S	065.0610°W	-	Pérez and Tissot 2021
17	Rawson, Chubut Province	CNP-INV 3859 (2021)	43.2982°S	065.0961°W	-	Pérez and Tissot 2021

Table 2. Shell measurements of *Rumina decollata* specimens from Misiones and Córdoba provinces, Argentina. Measurements in mm. SH = shell height (maximum length, mm); SW = shell width (maximum width, mm); AT = aperture length (greatest transverse measurement of aperture); AW = aperture width (maximum horizontal width of aperture, mm); FwH = height of body whorl (at middle of shell, mm); WN = number of whorls.

Locality	Voucher	SH	SW	AT	AW	FwH	WN
Posadas, Misiones Province	IBS-Ma 565-2	28.89	10.88	10.57	7.51	15.24	5 ½
	IBS-Ma 565-3	29.75	10.55	10.75	7.15	16.15	4 1/2
	IBS-Ma 565-4	25.50	10.70	9.95	7.50	15.40	4
	IBS-Ma 565-5	29.91	10.67	10.97	6.62	17.58	6
Río Tercero, Córdoba Province	IBS-Ma 383-1	27.65	10.01	10.39	6.26	15.42	4
	IBS-Ma 383-2	30.75	10.65	10.10	6.65	15.95	6
	IBS-Ma 383-3	26.83	10.62	9.67	5.94	14.98	5
	IBS-Ma 383-4	25.79	10.09	10.19	5.72	15.27	4 1/2
	IBS-Ma 383-5	27.70	10.25	10.05	6.40	16.10	4
	IBS-Ma 383-7	32.55	10.77	11.68	5.98	16.75	6 ½
Villa María, Córdoba Province	IBS-Ma 388-2	29.25	11.11	11.48	5.92	16.48	6
	IBS-Ma 388-3	28.12	11.03	10.67	6.04	15.23	5

identification was achieved based on morphological features of the genital system and partial COI sequences data.

Anatomical description. The pallial complex (Fig. 3A, B) is elongated, 25 mm long on average. A triangular kidney is located proximally in the lung cavity alongside the periaortic intestinal bend and occupies 30% of the lung length. The primary ureter runs along the rectal side of the kidney up to the top of the lung cavity where it turns down along the rectum forming the secondary ureter, which opens in the ureteric pore at the pneumostome. The pericardium, located in the upper columellar side of the pallial system, is about 4 mm long. It is continuous with the prominent pulmonary vein that runs parallel to the rectum and reaches the mantle collar. The pulmonary

vein is about 15 mm long. The vascularization is moderate and presents scattered pigmented spots. The genital system of the specimens studied is shown in Figure 3C, and D. The bursa copulatrix is sacciform, with a short duct with internal straight folds. The vagina is cylindrical, longer and wider than the penis, and about 4.5 times as long as wide. The inner vaginal surface with longitudinal crenulated lamellae (Fig. 4A, B). The vas deferens is a conspicuous tubule of constant diameter and longer than the penis plus the vagina. It runs attached externally to the vagina and penis until it enters the integument at its medial portion. It continues beneath the integument and finally opens into the lumen at the proximal end next to the attachment of the penis retractor muscle. The penis is subcylindrical, slightly curved in the specimens

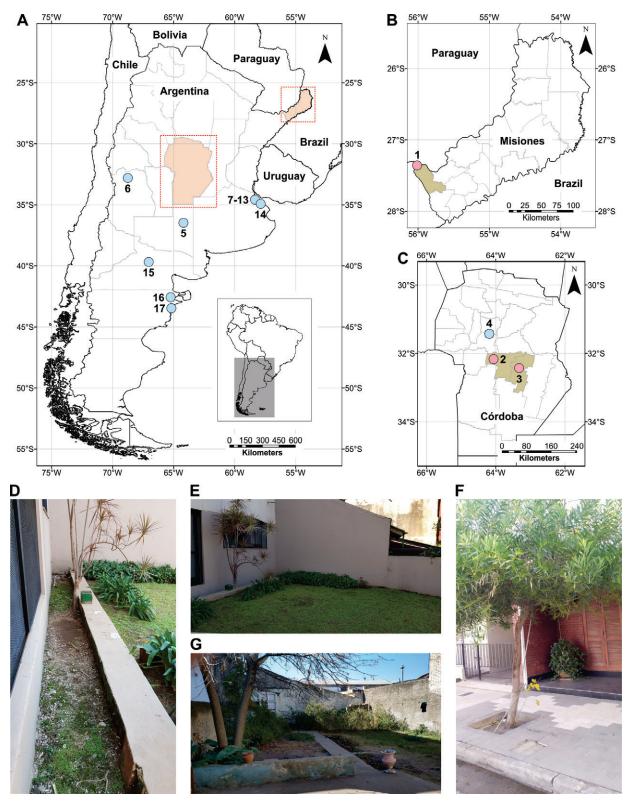


Figure 1. Distribution and environments of the exotic land snail *Rumina decollata* in Argentina. **A–C.** Localities where the species has been recorded in Argentina. **B.** Detail of the new record in Misiones Province. **C.** Detail of the new records in Córdoba Province. Locations in light blue correspond to literature records and the pink circles indicate the new localities where the specimens of *R. decollata* were found. Location numbers correspond to the ID numbers in Table 1. **D–E.** Residential garden where individuals were collected in Posadas, Misiones. **F.** Front yard and sidewalk where individuals were found in Río Tercero, Córdoba. **G.** Backyard of the house where individuals were found in Villa María, Córdoba.

analyzed, with an external constriction in the proximal third. Internally, the penis presents two different areas (Fig. 4C, D); the proximal one bears a voluminous and folded mass and two longitudinal conspicuous pilasters

surrounded by pustules. The distal area, from the mass to the genital atrium, has longitudinal and crenulated folds. **Source area of Argentine populations.** Partial COI sequences were of 655 bp in length for all specimens. The

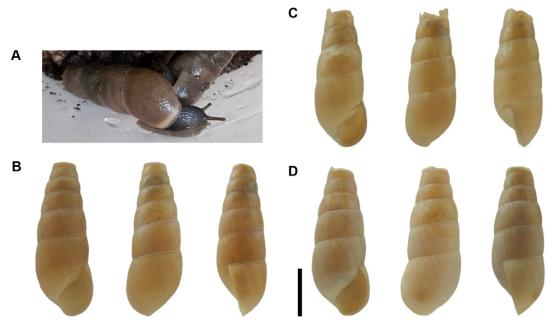


Figure 2. Living specimen and ventral, dorsal, and lateral views of shells of *Rumina decollata* from Argentina. **A.** Living specimen from Río Tercero, Córdoba Province. **B.** Shell from Posadas, Misiones Province (IBS-Ma 565-5). **C.** Shell from Río Tercero, Córdoba Province (IBS-Ma 383-1). **D.** Shell from Villa María, Córdoba Province (IBS-Ma 388-3). Scale bar = 1 cm.

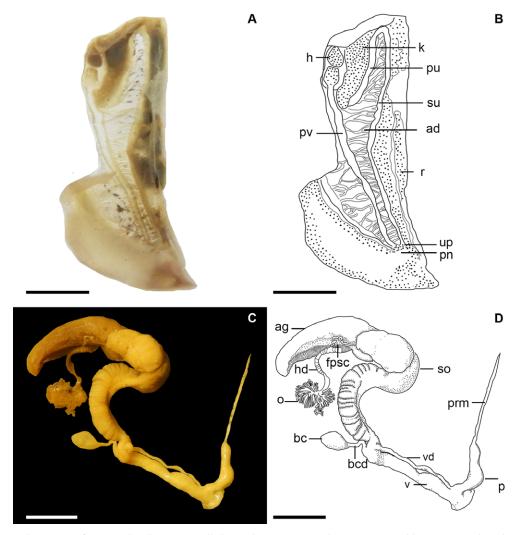


Figure 3. Internal anatomy of *Rumina decollata*. **A, B.** Pallial complex. **C, D.** Reproductive system. Abbreviations: ad = ad-rectal area; ag = albumen gland; bc = bursa copulatrix; bcd = bursa copulatrix duct; fpsc = fertilization pouch–spermathecal complex; h = heart; hd = hermaphroditic duct; k = kidney; o = ovotestis; p = penis; pn = pneumostome; prm = penis retractor muscle; pu = primary ureter; pv = pulmonary vein; r = rectum; so = spermoviduct; su = secondary ureter; up = ureteric pore; v = vagina; vd = vas deferens. Scale bars = 5 mm.

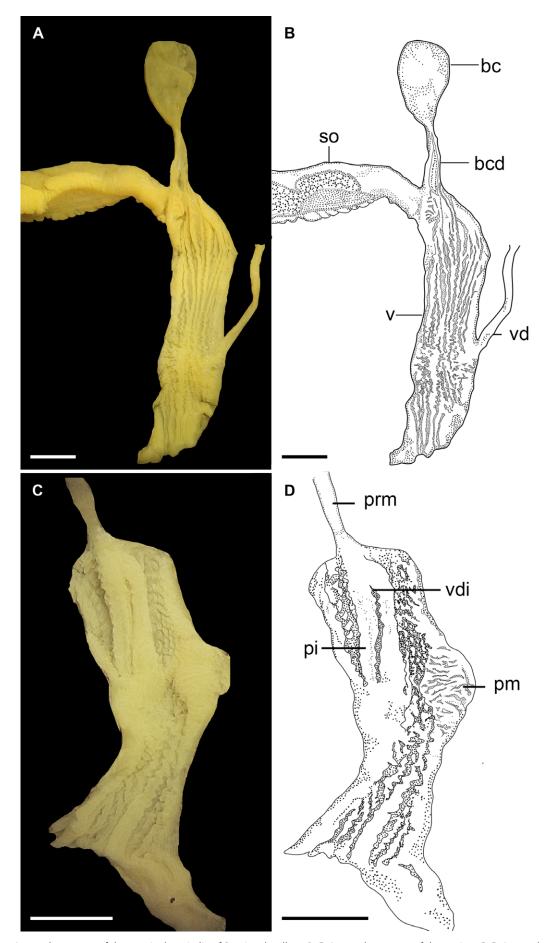


Figure 4. Internal structure of the terminal genitalia of *Rumina decollata*. **A, B.** Internal structure of the vagina. **C, D.** Internal structure of the penis. Abbreviations: bc = bursa copulatrix; bcd = bursa copulatrix duct; prm = penis retractor muscle; pi = pilaster; pm = penial mass; so = spermoviduct; v = vagina; vd = vas deferens; vdi = vas deferens insertion. Scale bars = 2 mm.

BLASTn search results with the COI sequences here obtained confirmed their specific identities as R. decollata, as they showed 100% coverage and top-ranking scores between 99.85 and 100% similarity with the sequences available in the GenBank database. No genetic variation was found within each examined locality, with specimens from Córdoba and Misiones provinces sharing the same single haplotype. Both phylogenetic approaches provided similar results; consequently, we report only the ML tree. Phylogenetic analyses found that R. decollata sequences from Córdoba and Misiones provinces are nested within MOTU A, although sequences obtained here are different from those previously reported for the species in South America (Fig. 5). The phylogenetic trees and haplotype network showed that two different haplotypes are present in the Argentine populations, which

originated from two distinct source areas within the native range: the "Arg" haplotype, previously observed by Prévot et al. (2014) in specimens from Mendoza Province and related to populations from southern France, and the one identified here from Córdoba and Misiones provinces, which was identical to a haplotype from the Iberian Peninsula (Spanish and Portuguese populations).

Discussion

In this study we report for the first time the occurrence of the decollate snail *Rumina decollata* in the Misiones Province, Argentina, from Posadas city. The present finding extends the distribution range of *R. decollata* about 800 km northeastward from the sites where it was first detected in Buenos Aires Province (Miquel 1988),

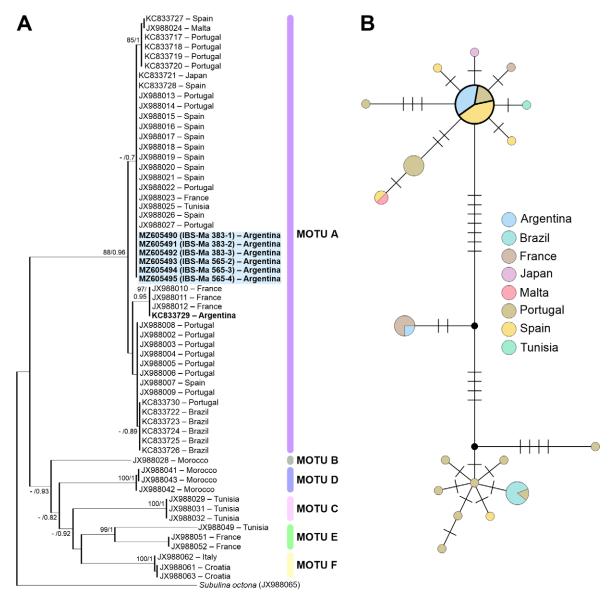


Figure 5. Phylogenetic reconstruction and haplotype network of *Rumina decollata* based on the partial COI gene. **A.** ML tree. Numbers within groups are GenBank accession numbers. The bootstrap values for the ML tree and posterior-probability values for BI are shown above or below the branches; support values below 70/0.7 are not shown. **B.** Haplotype network of the specimens of MOTU A. Haplotypes are colored according to the respective countries, with their size proportional to the number of individuals sharing the same haplotype. Haplotype highlighted with a thicker line corresponds to that of the specimens from Córdoba and Misiones provinces. Small lines indicate the number of mutations separating haplotypes. Small black circles represent undetected/hypothetical intermediate haplotypes.

and more than 900 km in the same direction from the recently published record for the species in Córdoba city, Córdoba Province (Reyna and Gordillo 2018). Although the species may have been present in this region for some time and had gone unnoticed, it seems highly unlikely since the Misiones Province has been the aim of several malacological surveys carried out in recent decades (e.g., Rumi et al. 2006; Núñez et al. 2010; Gutiérrez Gregoric et al. 2013; Beltramino et al. 2018; Serniotti et al. 2019). On the contrary, as is the case with many other widespread invasive species in our country (e.g., Serniotti et al. 2019, 2020), it is difficult to overlook the absence of R. decollata in the regions between Córdoba and Misiones provinces, which may be attributed to a lack of surveys in these provinces. In addition, two new localities were recorded in the Córdoba Province, namely Villa María and Río Tercero, which increase the number of records in central Argentina and suggest the species is well established and successfully reproducing. These new records reinforce the hypothesis that R. decollata is rapidly spreading across Argentina and prove it can withstand a wide range of climatic conditions, ranging from the semiarid zone with temperate-cold climate in the Patagonia, through temperate areas in Córdoba Province, all the way to the subtropical region of the Misiones Province (Selander and Kaufman 1973; Oliveira and Abreu 2013; Reyna and Gordillo 2018; Pérez and Tissot 2021).

Individuals of R. decollata were collected from urban gardens, which are the places the species usually inhabits in anthropized areas, probably due to the high availability of food and protection against natural predators (De Francesco and Lagiglia 2007; Reyna and Gordillo 2018). As it happens with other travelling species (as defined by Robinson 1999), this preference for ornamental plants might be the vector responsible for the widespread of the species in Argentina and, most likely, for its introduction in the Misiones Province. In addition, there is evidence that R. decollata also spreads and colonizes areas far from anthropogenic activity, invading riparian and natural habitats (Selander and Kaufman 1973). Therefore, it is expected that new colonized habitats will be reported for our country in the future, comprising both natural and anthropogenic environments.

In this study we present the first anatomical description of Argentine specimens of *R. decollata* from the Córdoba and Misiones provinces. All the individuals analyzed here presented turriculate shells with broken apices, which is the diagnostic feature of the genus *Rumina*. Based on shell shape as well as body and foot coloration, the specimens examined were distinguished from *R. saharica*, which is characterized by having slender and more cylindrical shells, and a lighter color of the body (Carr 2002; Prévot et al. 2015). Regarding the gross morphology of the reproductive system, Argentine specimens exhibited most of the morphological features of the genital system defined for *R. decollata* as described by Wille (1915), Germain (1930), Pilsbry (1946), Giusti (1970), Carr (2002), and Prévot et al. (2015). The penis

was subcylindrical, slightly curved, with an external constriction in the proximal third, similar to that illustrated by Carr (2002: figs. 1.3, 1.6). The internal structure of penis fits particularly well with that described by Pilsbry (1946), characterized by a granulose proximal region with two strong pilasters between which the vas deferens opens by a simple pore. The pilasters were not figured by Carr (2002). In addition, Argentine specimens showed a vagina with an inner sculpture exhibiting longitudinal crenulated lamellae, which differ from the straight ones present in R. saharica as reported by Carr (2002) and Liberto et al. (2012). On the other hand, R. iamone was dismissed as a possibility for the specimens collected in Córdoba and Misiones provinces, as that species, described by Quintana Cardona (2017), has a penis of constant diameter, a long bursa copulatrix, and a much longer vagina than those observed in this study.

At the molecular level, a single haplotype of the partial mitochondrial COI gene was found among the analyzed individuals of R. decollata from the Córdoba and Misiones provinces, whose species-specific identity was confirmed by the BLASTn search results. This haplotype, which is included within MOTU A, was previously described from Spanish and Portuguese populations of the Iberian Peninsula by Prévot et al. (2013) and differs from the one found in the Mendoza Province, which instead is related to populations of southern France. This finding suggests at least two different introductions of R. decollata into Argentina, one from southern France, and one from the Iberian Peninsula. To date, the introduction pathway of R. decollata into Argentina remains uncertain but could be related to inadvertent human transport, possibly in association with the commerce of ornamental plants and the horticultural trade (Cowie and Robinson 2003; Cowie et al. 2008). The Misiones province is located in the northeastern corner of Argentina, bordering on Brazil and Paraguay. In this province, other invasive mollusks species—such as the Giant African Snail Lissachatina fulica (Bowdich, 1822), the Jumping Snail Ovachlamys fulgens (Gude, 1900), and the Chinese Slug Meghimatium pictum (Stolyczka, 1873)—have been recorded, and their occurrences have been suggested to be linked to a dispersion from Brazil and/or to the commerce of flora (Gutiérrez Gregoric et al. 2013; Beltramino et al. 2018). Although R. decollata has been reported in the southernmost states of Brazil (Minas Gerais, Paraná, Rio Grande do Sul, São Paulo, and Santa Catarina) (Landal et al. 2019), the haplotype network developed here accounts for a significant genetic distance (13 mutational steps) between the haplotype found in this study and those of Brazil, suggesting that the origin of their introductions are not related. On the contrary, the distribution pattern of R. decollata in Argentina, together with the genetic evidence, points out that dispersal routes from central Argentina are possible. However, further studies comprising more populations from more provinces are needed to elucidate the introduction history and dispersal pathways of *R. decollata* in the country.

Given its generalist habits, its ability to self-fertilize, and its high reproduction rates, R. decollata was to be expected to become a horticultural pest in several countries around the world (e.g., Correa Sandoval 1993; Matamoros 2014a, 2014b; Landal et al. 2019). Despite the fact that it has not been reported causing damage to crops in Argentina, Miquel (1988) stated the species was observed feeding on black mulberries plants (Morus nigra) and ornamental plants such as nasturtium (Tropaeolum majus L.) in the area where it was reported for the first time, which could constitute a serious threat for fruit production. Although these plant species are not part of large-scale production systems in Argentina (SENASA 2020), risk analyses should be undertaken to assess the potential threat of R. decollata to national agroecosystems. In addition, R. decollata may serve as an intermediate host of Aelurostrongylus abstrusus, a causative agent of bronchopneumonia in wild and domestic cats (Cardillo et al. 2018), and as a paratenic host of Toxocara cati, the etiological agent of toxocariasis (Cardillo et al. 2016). The presence of these parasites has already been reported for the Buenos Aires Province, where they were found in both feces of domestic cats and R. decollata snails (Cardillo et al. 2014, 2016, 2018), and for the Misiones Province, where A. abstrusus larvae were recovered from Lissachatina fulica snails (Valente et al. 2017). Although there is no record of these parasites infesting the recently found populations of R. decollata, further studies are needed to assess the co-occurrence of parasites and snails in the Córdoba and Misiones provinces.

Finally, considering the rapid spread documented of *R. decollata* in Argentina, which could have negative consequences on native biota, commerce, and health of animals and humans, we expect management authorities may use this information to establish priority areas for surveillance and to plan preventive actions aimed at monitoring its distribution and limiting the spread and impacts of the decollate snail in Argentina.

Acknowledgements

This study was financially supported by Facultad de Ciencias Exactas, Químicas y Naturales – Universidad Nacional de Misiones (Proyectos de Investigación 16/Q1227-PI and 16/Q1559-PI), Agencia Nacional de Promoción de la Investigación, el Desarrollo Tecnológico y la Innovación (PICT-2017-3961) and Universidad Nacional del Sur (PGI 24/B244). We thank Dr. Carl Christensen and an anonymous reviewer for corrections made during the peer-review process, as well as Robert Forsyth for the constructive comments which greatly improved the manuscript.

Authors' Contributions

Conceptualization: REV AAB, JDC, AIR. Data curation: AIR, AAB. Formal analysis: JP, AIR, AAB, JDC, REV. Funding acquisition: AAB, JP, JDC, REV. Investigation:

AIR, JP, ENS, JDC, AAB, REV. Methodology: JP, AAB, REV, AIR. Resources: REV, JP, ENS. Supervision: REV. Validation: ENS, REV, JP, AAB, AIR. Visualization: AAB, JP, JDC, REV, AIR. Writing – original draft: REV, AAB, JP, AIR, JDC, ENS. Writing – review and editing: JDC, AAB, ENS, AIR, JP, REV.

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