

SYSTEMATIC REVISION OF *CHILINA* GRAY (GASTROPODA: PULMONATA)
FROM NORTHWESTERN ARGENTINA AND DESCRIPTION OF A NEW SPECIES

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ABSTRACT

The family Chiliniidae in Argentina has been thought to include 17 species, most being found in Patagonia. All the original descriptions were based mainly on shell characteristics. The present work contains information about the shells, the radulae, and the reproductive and nervous systems in Chiliniidae species from northwestern region, along with the description of a new species. This new species from Tucumán Province is characterized by the shell and a penial morphology that includes pustules and transverse lamellae with irregular contours and longitudinal folds. The taxonomic position of *Chilina tucumanensis* is also reviewed and a redescription of *Chilina portillensis* is provided. Additional information is provided on their habitats and distributions. These records in northwestern Argentina now increase the total number of species of *Chilina* inhabiting Argentina to 19.

Key words: Chiliniidae, taxonomy, *Chilina lilloi*, n. sp., freshwater snail, South America.

INTRODUCTION

The Chiliniidae (Gastropoda: Pulmonata) is one of the oldest freshwater families currently known. Such primitive features as the presence of one chiasmoneuric nervous system, horizontal lamellar tentacles, a noncontractile pneumostome, and an incomplete division of male and female ducts (Haeckel, 1911; Harry, 1964) have indicated a relationship to different groups of gastropods (Hubendick, 1945, 1978). Dayrat et al. (2001) published a molecular phylogeny of Euthyneura that argued for the monophyly of Hygrophila and proposed the Chiliniidae as a basal group. Klussmann et al. (2008) distinguished two clades within the Hygrophila, the first including *Chilina* Gray, 1828, and *Latia* Gray, 1850, and the second comprising higher limnic Basommatophora. More recently, Dayrat et al. (2011) on the basis of molecular analyses concluded that the Hygrophila was not a monophyletic group and proposed a relationship between the Chilinoidea and the Amphiboiloidea, although this contention was not well supported.

Chiliniidae is endemic in southern South America, extending from the coasts of Peru to Cape Horn (Pilsbry, 1911; Castellanos &

Gaillard, 1981) and also reaching the Falkland Islands (Brown & Pullan, 1987). This family consists in only a single genus, *Chilina*, with 32 nominal species, 17 of which are found in Argentina (Núñez et al., 2010). Most Argentine species of *Chilina* were originally described on the basis of shell characteristics alone, but more recently anatomical studies of certain species inhabiting Argentina were performed (Miquel, 1984, 1987; Ituarte, 1997; Gutiérrez Gregoric & Rumi, 2008; Gutiérrez Gregoric, 2010).

Chilina species can be found in quite different types of habitats – such as lakes, lagoons, dams, waterfalls, streams, rivers, canals, and estuaries; all generally with clean oxygenated water and variable temperature ranges (Gutiérrez Gregoric, 2008; Cuezco, 2009).

The northwest region of Argentina, extending between latitudes 21° and 30°S and longitudes 62° and 69°W, covers an area of approximately 470,184 km² and is an ecologic region characterized by the presence to four of the 14 ecoregions of Argentina (Puna, Altos Andes, Chaco, and Yungas). Northwestern Argentina and southern Bolivia constitute the boundary of the distribution of Andean Yungas forest within South America and forming a biogeographic

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as well as an ecologic and social unit (Grau & Brown, 2000). Of the freshwater gastropods, 13 species of pulmonates within the families Physidae, Planorbidae, Lymnaeidae, Ancyliidae, and Chiliniidae have been documented in northwestern Argentina (Rumi et al., 2008). Of the Chiliniidae, three species have been reported: *Chilina parchappii* (d'Orbigny 1835), *Chilina portillensis* Hidalgo, 1880, and *Chilina fluminea tucumanensis* Castellanos & Miquel, 1980. *Chilina parchappii* was recorded in Salta Province (Castellanos & Miquel, 1980) and is a species associated with lotic environments and mesohaline bodies of water of the southern pampas (Tietze & De Francesco, 2010). *Chilina portillensis*, cited in the Salta and Jujuy provinces, is considered endemic in Argentina, with a restricted and discontinuous distribution (Rumi et al., 2006; Núñez et al., 2010). *Chilina fluminea tucumanensis*, from Tucumán and Córdoba provinces, was established by Castellanos & Miquel (1980) as a subspecies of *Chilina fluminea* (Maton, 1809) based on specimens collected in Tucumán Province.

The aim of the present study was to provide information on the anatomy of the species of *Chilina* present in northwestern Argentina and to review the taxonomic status of all species of this genus in that area. Finally, during the examination of the collected specimens along with museum materials, a new species was identified. In this report, we therefore name this new member of the genus and present a complete description of the species.

MATERIALS AND METHODS

The specimens studied come from malacological collections at the Museo Argentino de Ciencias Naturales, Ciudad de Buenos Aires (MACN); the Museo de La Plata, Buenos Aires (MLP); and the Instituto Fundación Miguel Lillo, Tucumán (IFML). Additional material was collected during the field work carried out in northwestern Argentina during summer-autumn seasons from 2010 to 2011. Adult specimens collected were kept in 96% (v/v) aqueous ethanol, after a relaxation in menthol (12 h), followed by an immersion in hot water (70°C). For anatomical studies of the reproductive and pallial systems, the methodology proposed by Cuezco (1997) was followed. Dissections were made under a Leica MZ6 stereoscopic microscope and anatomical systems drawn with the help of a *camera lucida*. Terminology used for the anatomical descriptions follows

that of Harry (1964) and for the descriptions of the nervous system that of Ituarte (1997). Differences in lengths of the right and left nervous connectives were assessed by the Student t test (Gutiérrez Gregoric, 2010).

Radulae were separated from the buccal mass, cleaned by immersion in a sodium hypochlorite solution (Clorox™) and mounted for scanning electron microscopy. The radular-dentition formula gives the number of teeth per row: [(number of left and right teeth) / (number of cusps) + (number of central teeth) / (number of cusps)] number of transversal rows or their lower and maximum number. Seven shell measurements were taken according to Gutiérrez Gregoric (2010): total length (TL), length of the last whorl (LWL), aperture length (AL), total width (TW), aperture width (AW), aperture projection (AP), and spire length (SP). In order to obtain size-free variables to facilitate comparisons between different individuals and species, measurements of the soft parts were expressed as a proportion of the length of the last whorl.

SYSTEMATICS

Family Chiliniidae

Genus *Chilina* Gray, 1828

Diplicaria Rafinesque, 1833: 165.

Linneus d'Orbigny, 1835: 24, in part.

Dombeia d'Orbigny, 1843: 325: pl. 43, figs. 1–20.

Pseudochilina Dall, 1870: 357.

Acyrogonia Rochebrune & Mabille, 1889: 25.

Type Species

Auricula (Chilina) fluctuosa Gray, 1828 (subsequent designation of Gray, 1847).

Original Diagnosis

“Shell ovate, thin; aperture large, expanded; columella flattened in front, with an oblique central fold; outer lip thin. Fresh running water”.

Diagnosis

Oval (oblong to ventricose) shell with an expanded last whorl. Vestigial chiastoneury. Incomplete division of male and female ducts. Calcareous granules in the vaginal lumen and presence of a secondary bursa copulatrix or accessory seminal receptacle. Penial terminal lamellas with cuticularized teeth-like structures.

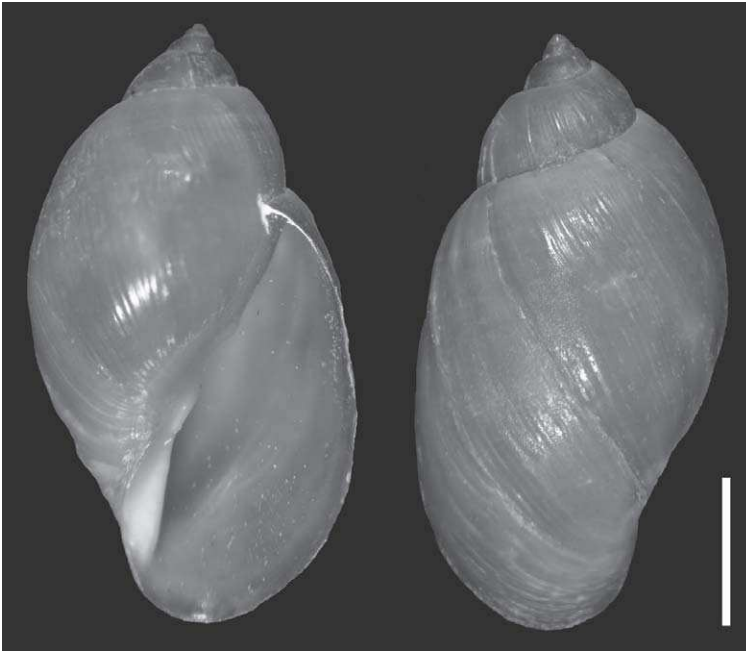


FIG. 1. Shell of *Chilina lilloi*, n. sp. (Holotype). Scale bar = 2 mm.

***Chilina lilloi*, n. sp.**

Type Locality

Juan Bautista Alberdi; unnamed stream on way to Escaba Dam; 27°40'06"S, 65°45'01"W; 1,580 m; Tucumán Province, Argentina.

Type Material

Holotype: IFML 15536 from type locality, 10/11/1999, M. G. Cuezco Leg (dry shell).

Paratypes: IFML 15537 from type locality, 10/11/1999, M. G. Cuezco leg (3 dry shells and 10 preserved specimens). MLP 13343 from type locality, 10/11/1999, M. G. Cuezco leg (3 preserved specimens).

Other Material Examined

Argentina, Tucumán Province: MLP 6887 (3 preserved specimens) and MLP 13343 (3 preserved specimens): Juan Bautista Alberdi Dept., unnamed stream on way to Escaba Dam; 27°40'06"S, 65°45'01"W; M. G. Cuezco leg.

Etymology

The name of the species was given after the scholar Miguel Lillo, from Tucumán.

Diagnosis

Shell with two columellar teeth, upper tooth situated in inner position and poorly developed. Penis sheath $1\frac{1}{4}$ times length of prepuce. Inner sculpture of penis sheath differentiated in three regions. Inner sculpture of prepuce with numerous longitudinal folds, smooth and tightly pressed. Gross nervous connectives. Right pleuroparietal connective preceding penis sheath.

Description

Shell (Fig. 1): Oval, light brown. Five convex whorls, each whorl larger than previous ones. Suture simple, deep not grooved. Protoconch and first whorl dark brown or, in some cases, almost reddish brown. Shell surface with pigmented longitudinal bands, regularly arranged. Last whorl well developed, higher than

TABLE 1. Mean and range of seven measurements for *Chilina lilloi*, n. sp., *C. tucumanensis* and *C. portillensis* from Tucumán Province, Argentina. TL, total length; LWL, last whorl length; SL, spire length; AL, aperture length; TW, total width; AW, aperture width; AP, aperture projection; SL, spire length; n/d, no data.

Species		TL	LWL	AL	TW	AW	AP	SL
<i>Chilina lilloi</i> (n = 15)	Mean	10.93	9.82	8.07	6.22	3.99	1.31	0.63
	SD	0.84	0.79	0.57	0.39	0.20	0.36	0.05
	Max	12.37	11.22	9.11	6.70	4.34	2.39	0.74
	Min	9.62	8.42	6.80	5.16	3.64	1.06	0.58
<i>Chilina tucumanensis</i> (n = 42)	Mean	13.00	11.99	10.06	7.98	4.83	1.97	1.13
	SD	0.92	0.88	0.71	0.63	0.58	0.51	0.23
	Max	14.53	13.72	11.48	9.96	6.25	3.80	1.39
	Min	11.70	10.70	8.90	6.94	4.22	3.17	0.81
<i>Chilina portillensis</i> (n = 4)	Mean	n/d	10.97	9.52	7.48	5.35	2.31	n/d
	SD	n/d	1.67	1.26	1.03	0.37	0.21	n/d
	Max	n/d	13.24	11.21	8.84	5.89	2.60	n/d
	Min	n/d	9.23	8.19	6.36	5.10	2.16	n/d

wide. Aperture oval, slightly excavated at outer edge; aperture basal edge reddish, with smooth expansion outwards. Parietal callus whitish, thick, with its terminal portion slightly widened, flattened. Two columellar teeth, lower tooth more prominent and developed than upper, innermost on columella. Dimensions of 15 specimens taken from the material collected (Table 1).

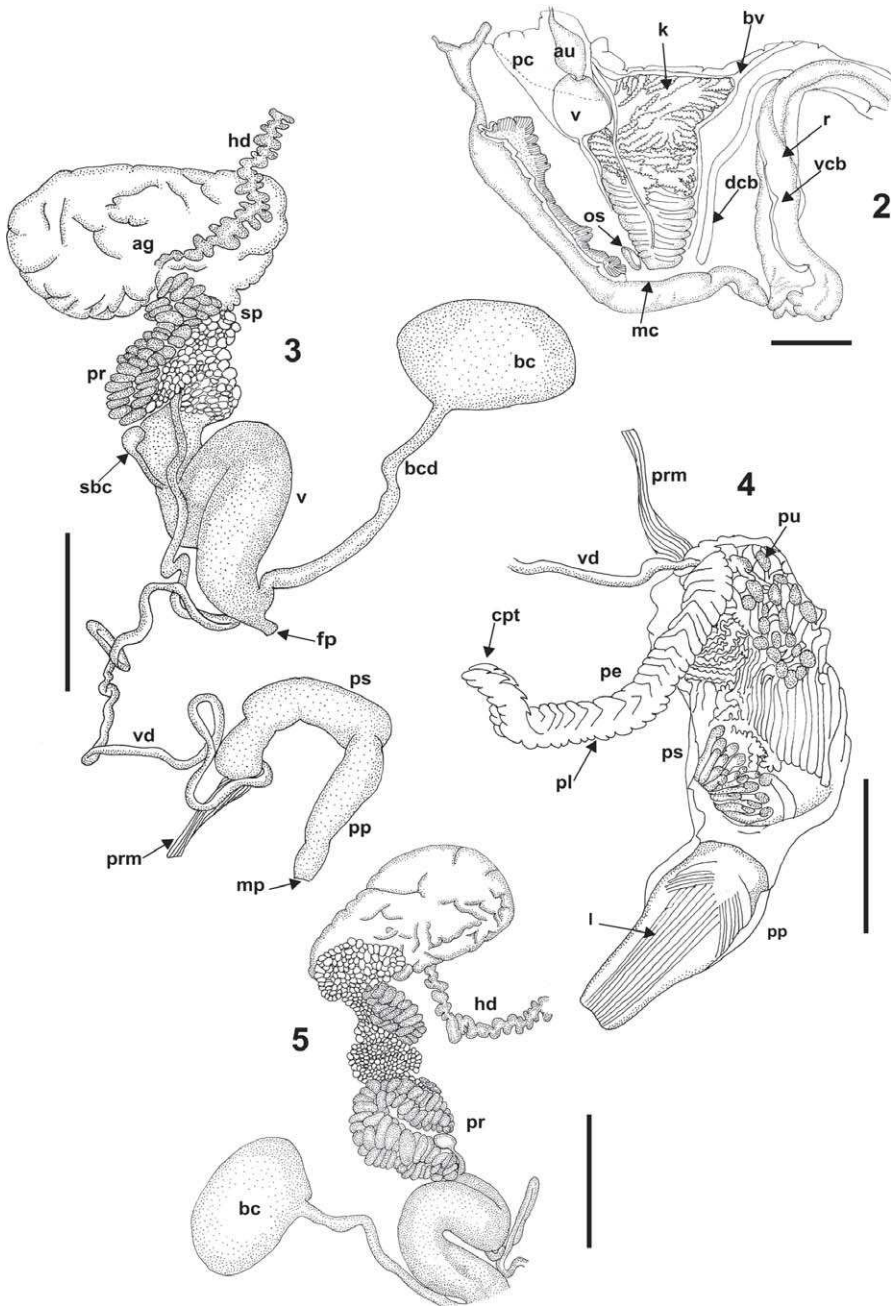
Pallial System (Fig. 2): Extending to half of body whorl. Pulmonary roof pigmented, uniform dark gray color. Kidney occupying almost entire pulmonary roof length. Kidney inner sculpture with numerous trabeculae, irregular contour, transverse, anastomosing. Trabeculae lower portion fused, giving a lobed appearance. Renal pore situated at distal end portion of kidney level with pneumostome. True ureter absent. Pericardium pear-shaped with length $\frac{1}{2}$ of kidney length. Pulmonary vein parallel to left margin of kidney without branching. Two blood vessels running through top and right margins of kidney. Pallial cavity lamella divided into dorsal and rectal portion, dorsal portion located on lung roof on right side of kidney, rectal portion $\frac{3}{4}$ of rectum length, running on rectum surface. Rectum on right side of mantle cavity running until pneumostome.

Reproductive System (Figs. 3–5): Ovary constituted by several branches of small, rounded follicles flowing into a common duct. Ovary embedded in digestive gland at apex

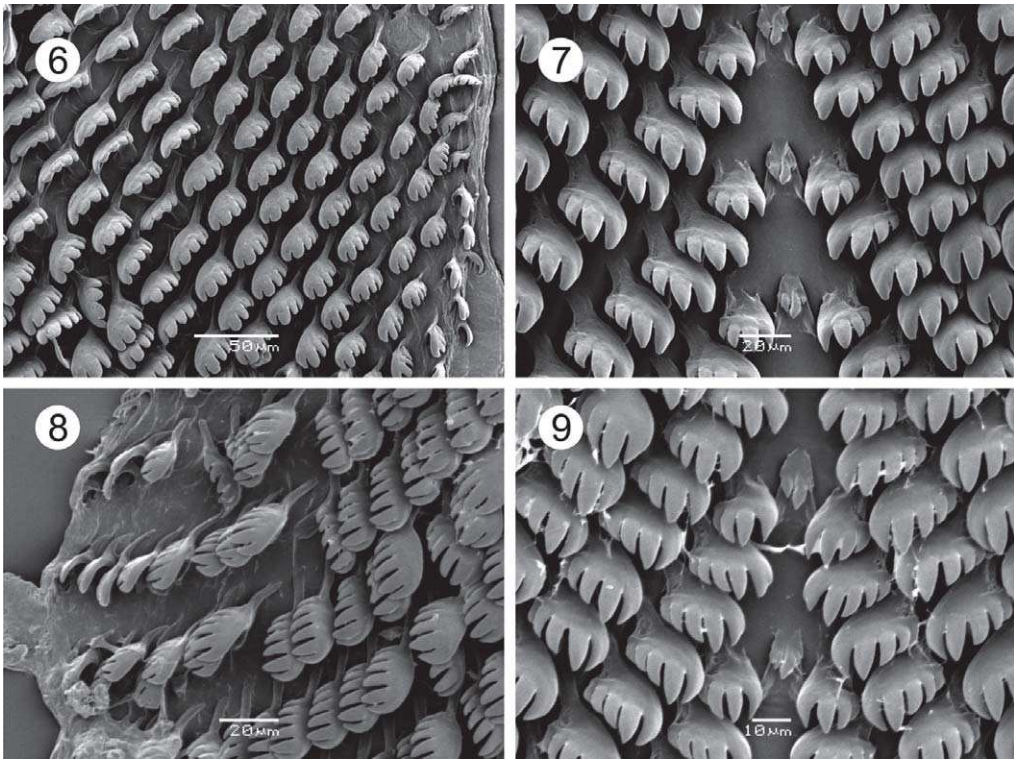
of shell. Common duct flowing into hermaphrodite duct, with irregular contours on both sides. Albumen gland elongated, glandular, twice as big as uterus and becoming sperm duct. Uterus proximal portion with glandular walls, folded over itself and on ventral part of albumen gland.

Female Genital System: Transparent, glandular-looking uterus wall continuing through free oviduct. Bursa copulatrix consisting of cylindrical tube with uniform diameter, inserting into distal free oviduct. Bursa copulatrix duct length $1\frac{1}{2}$ times the diameter of its sac. Bursa copulatrix sac on left side of cephalopodal hemocoel between pericardial cavity and columella base. Secondary bursa copulatrix short, emerging from uterus base (25% bursa copulatrix duct length), cylindrical, expanded in its distal portion. Vagina shortest and narrow portion between free oviduct and female genital atrium. Vagina cylindrical, folded over free oviduct and entering female atrium. Female genital atrium opening outward through female pore on right side of head above male pore.

Male Genital System: Prostate extending to lower half of uterus and consisting of variable size and cylindrical-shaped acini. Vas deferens, emerging from terminal portion of prostate, showing a constant diameter throughout its length and running parallel to right distal portion of oviduct to penis complex, over uterus. Vas deferens entering body wall along an external



FIGS. 2-5. *Chilina lilloi*, n. sp. FIG. 2: Ventral view of pallial system dissected out. Scale bar = 2 mm; FIG. 3: Dorsal view of reproductive system. Scale bar = 1 mm; FIG. 4: Ventral view of reproductive system without ovotestis; FIG. 5: Penis inner wall. Scale bar = 1 mm. Abbreviations: ag, albumen gland; au, auricle; bc, bursa copulatrix; bcd, bursa copulatrix duct; bv, blood vein; k, kidney; l, lamellas; mc, mantle collar; mp, male pore; os, osphradium; pc, pericardic cavity; pe, penis; pl, penis lamella; pp, preputium; pr, prostate; prm, penis retractor muscle; ps, penis sheath; pu, pustules; r, rectum; rp, renal pore; sbc, secondary bursa copulatrix; sp, spermiduct; va, vagina; vcb, ventral ciliar band; vd, vas deferens; ve, ventricle.



FIGS. 6–9. Radula of *Chilina lilloi*, n. sp., from stream on way to Escaba Dam, Tucumán, Argentina. FIGS. 6, 8: Lateral teeth. Scale bars = 50 µm and 20 µm; FIGS. 7, 9: Central tooth and first lateral teeth. Scale bars = 20 µm and 10 µm.

TABLE 2. Ratio between ganglia length and last whorls length in *Chilina lilloi*, n. sp. (n = 7), *Chilina tucumanensis* (n = 4) and *C. portillensis* (n = 4). Abbreviations for each ganglion: c, cerebral; lc, left cerebral; lp, left parietal; lpe, left pedal; lpl, left pleural; p, pedal; rc, right cerebral; rp, right parietal; rpe, right pedal; rpl, right pleural; si, subintestinal; v, visceral.

	<i>Chilina lilloi</i>			<i>Chilina tucumanensis</i>			<i>Chilina portillensis</i>		
	Ratio	Mean (mm)	SD	Ratio	Mean (mm)	SD	Ratio	Mean (mm)	SD
lc – rc	13.11	1.28	0.14	16.70	1.86	0.44	13.46	1.48	0.21
lpe – rpe	3.68	0.36	0.04	4.99	0.56	0.08	3.85	0.42	0.03
lc – lpl	3.97	0.39	0.09	7.26	0.81	0.30	6.98	0.77	0.03
rc – rpl	3.26	0.32	0.13	6.20	0.69	0.25	5.34	0.59	0.04
c – p	8.37	0.82	0.23	10.70	1.19	0.36	6.74	0.74	0.17
rpl – rp	11.55	1.13	0.23	13.79	1.54	0.31	16.24	1.78	0.42
lpl – lp	2.47	0.24	0.13	4.52	0.50	0.09	4.99	0.55	0.20
lp – si	15.92	1.56	0.14	18.23	2.03	0.77	16.14	1.77	0.24
rp – v	16.19	1.58	0.40	20.90	2.33	0.15	19.85	2.18	0.25
si – v	4.86	0.48	0.30	3.63	0.40	0.05	6.55	0.72	0.11

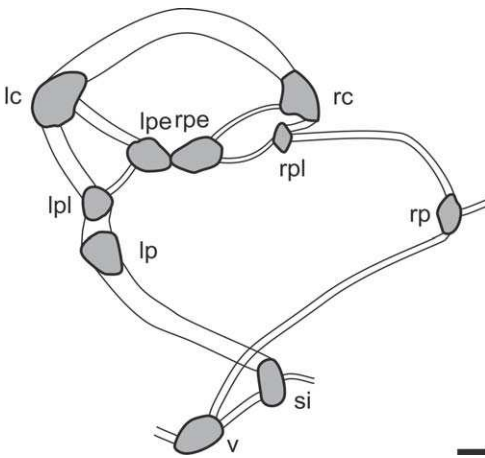


FIG. 10. Diagram of nervous system of *Chilina lilloi*, n. sp. Abbreviations: lc, left cerebral; lpe, left pedal; lp, left parietal; lpl, left pleural; rc, right cerebral; rpe, right pedal; rp, right parietal; rpl, right pleural; si, subintestinal; v, visceral. Scale bar = 1 mm.

partially occluded groove between female and male pores, subsequently reentering body cavity to be inserted into penis complex apex and folding into three loops of variable size before leaving body wall. Muscular penis sheath, $1\frac{1}{4}$ length of prepuce. Penis sheath inner sculpture differentiated into three regions. Upper right portion with pustules, upper left portion with irregularly contoured transverse lamellae, middle portion with longitudinal folds. Penis long ($1\frac{1}{4}$ length of penis sheath), robust, with outer surface cut by transverse lamellae, triangular in cross section. Prepuce cylindrical, thin, with constriction marked by oblique lines arranged in V making connection with penis sheath. Prepuce inner sculpture with numerous smooth, very tight longitudinal folds. Penis retractor muscle long, slender.

Digestive System

Radula (Figs. 6–9): Rows mean number 44 ($n = 4$; range = 42 to 46). Teeth per half row mean number (except for central tooth) 41 ($n = 4$; range = 39 to 43). Central tooth asymmetrical, tricuspid, elongated base as high as wide, mesocone more developed, slightly tilted to left. Cusps more developed giving tooth a pincer-like aspect. First lateral tooth tricuspid or tetracuspid, with endocone (tricuspid) or the inner second cusp (tetracuspid) more developed, base of tooth narrower than the apical part (cusp area), cusps taller than wide. On base of tooth, a groove dividing endocone from

other cusps. Second lateral tooth tetracuspid or pentacuspid, with inner cusp more developed, base of tooth narrower than apical part of tooth. Unlike previous tooth, base more notable. Last teeth with thin base, can even have 5–7 cusps of similar development.

Nervous System (Fig. 10; Table 2)

All connectives between ganglia relatively thick compared to both size of ganglia and system in general. Length of left connective joining cerebral ganglion with pleural ganglion greater than right connective, but the difference does not reach statistical significance (t_8 ; $p > 0.05$; 3.97 vs. 3.26% of LWL). Right pleuroparietal connective preceding penis sheath. Significant differences (t_8 ; $p < 0.001$) in length of pleuroparietal connectives, with left one smaller than right one (2.47 vs. 11.55% of LWL). Long connective (ratio: 15.92 of LWL) linking left parietal ganglion to subintestinal ganglion, located above posterior half of columellar muscle. Long connective (ratio: 16.19 of LWL) linking right parietal ganglion to visceral ganglion. One very short connective (ratio: 4.86 of LWL) linking subintestinal ganglion to visceral ganglion and closing posterior nerve ring.

Distribution & Habitat

Chilina lilloi was detected only at type locality in the Escaba area, Juan Bautista Alberdi Department in Tucumán; this locality is classified within the Yungas biogeographic ecoregion. The new species has been collected from a subtropical mountain stream with a stony substratum.

Remarks

Chilina lilloi is similar to the genus type species, *C. fluctuosa* (Gray, 1828), in general shell morphology (Haeckel, 1911). It differs from the latter in the presence of two small teeth on the columellar side in the aperture, those being absent in *C. fluctuosa*. In the present study, the dissection of several specimens showed three different types of sculpture. Pustules, lamellae, and longitudinal folds are present in the inner penis-sheath wall of *C. lilloi*. On the contrary, Harry (1964) described numerous papillae, some of them fused into lamellae present in *C. fluctuosa*. Other differences are the numerous, slender longitudinal folds found in the prepuce of *C. lilloi* that are not present in *C. fluctuosa* according to Harry (1964). *Chilina lilloi*, n. sp., differs from *C. tucumanensis* in having different shell morphology and a completely different sculpture pattern of the penis sheath.



FIG. 11. Shells of *Chilina tucumanensis*. Scale bar = 2 mm.

Chilina tucumanensis
Castellanos & Miquel, 1980

Chilina fluminea tucumanensis Castellanos & Miquel, 1980: 172.

Diagnosis

Male reproductive system with vas deferens forming two overlapping circles, of different sizes. Penis complex cylindrical, elongated. Penis sheath $2\frac{1}{4}$ times the length of prepuce.

Type Locality

Cochuna River, Tucumán Province, Argentina.

Type Material

Holotype: MACN 9927: from type locality, December 1916, Doello Jurado, M. leg (dry shell).

Paratypes: MACN 9927 (3 dry shells) and MLP 4042: from type locality, December 1916, Doello Jurado, M. leg. MACN 9954: Córdoba, Río Primero, 1917, Bondemberder leg (45 dry shells).

Other Material Examined

Argentina, Tucumán Province: IFML 15540: Concepción Dept; Las Cañas River; Villa

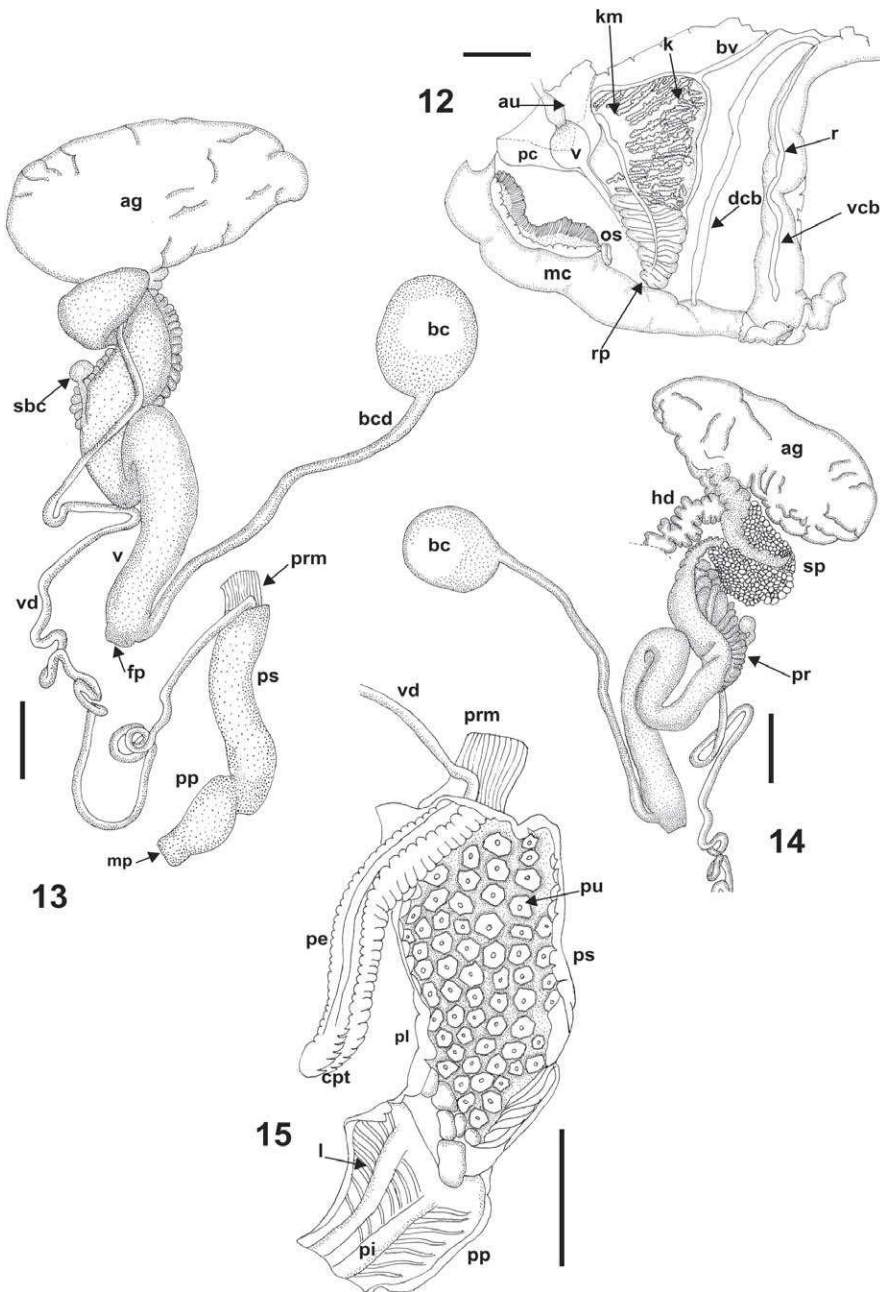
Lola Dam; $27^{\circ}21'40''S$, $65^{\circ}49'13''W$; 655 m; 14/11/2006; Cuezco M.G. leg. (23 preserved specimens). IFML 15541 (55 preserved specimens and 15 dry shells) and MLP 13344 (4 preserved specimens): Concepción; Cañas River; Villa Lola Dam; 09/07/2010, Ovando, X. M. C. leg. IFML 15550: Tafi del Valle, stream on the way to Tafi del Valle village; $27^{\circ}01'45.3''S$, $65^{\circ}39'20.6''W$; 1220 m; 28/03/2011; Ovando, X.M.C. leg. (60 preserved specimens). MACN 9927: Concepción, Cochuna River, 12/1916, Doello Jurado, M. leg (87 dry shells).

Etymology

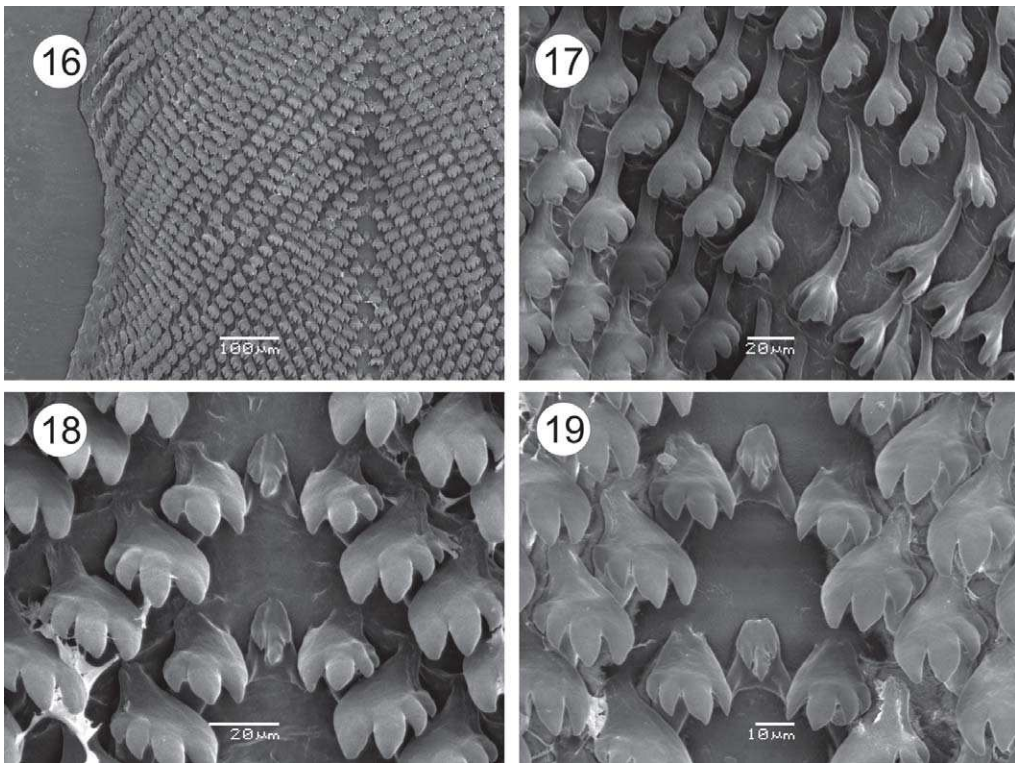
Named for Tucumán Province.

Description

Shell (Fig. 11): Dark brown, with dark brown bands in zigzags or spirals, more evident on the body whorl. Four convex whorls separated by simple suture. Protoconch sometimes eroded. Shells surface with growth lines, tight radial grooved layout. Body whorl prominent, not gibbous, longer than wide. Aperture oval, wider than long. Parietal area smooth, without ridges. Parietal callus wholly whitish, thick, narrow, with fold in the middle, posterior end slightly flattened. Two columellar teeth highly developed.



FIGS. 12–15. *Chilina tucumanensis*. FIG. 12: Ventral view of pallial system dissected out. Scale bar = 2 mm; FIG. 13: Dorsal view of reproductive system. Scale bar = 1 mm; FIG. 14: Ventral view of reproductive system without ovotestis; FIG. 15: Penis inner wall, Scale bar = 1 mm. Abbreviations: ag, albumen gland; au, auricle; bc, bursa copulatrix; bcd, bursa copulatrix duct; bv, blood vein; cpt, cuticular penis tooth; dcb, dorsal ciliar band; fp, female pore; hd, hermaphrodite duct; k, kidney; km, muscle band of kidney; l, lamellas; mc, mantle collar; mp, male pore; os, osphradium; pc, pericardic cavity; pe, penis; pi, pilasters; pl, penis lamella; pp, preputium; pr, prostate; prm, penis retractor muscle; ps, penis sheath; pu, pustules; r, rectum; rp, renal pore; sbc, secondary bursa copulatrix; sp, spermoviduct; va, vagina; vcb, ventral ciliar band; vd, vas deferens; ve, ventricle.



FIGS. 16–19. Radula of *Chilina tucumanensis* from Villa Lola Dam, Tucumán, Argentina. FIG. 16: General view. Scale bar = 100 µm; FIG. 17: Lateral teeth. Scale bar = 20 µm; FIGS. 18, 19: Central tooth and first lateral teeth. Scale bars = 20 µm and 10 µm.

The dimensions of 42 specimens from the collection of MLP were analyzed, including the type and paratype for MACN (Table 1).

Pallial System (Fig. 12): Pallial system dorsal portion light gray in whole area. Kidney triangular, occupying $\frac{3}{4}$ of pulmonary roof total length. Pericardium $\frac{1}{4}$ of kidney length, consisting of an auricle with a noticeable split.

Reproductive System (Figs. 13–15)

Female Genital System: Vagina cylindrical, longer than wide. Bursa copulatrix duct three times bursa sac diameter. Bursa copulatrix sac spherical-shaped. Secondary bursa copulatrix short, cylindrical, spherical (20% the length of bursa copulatrix duct), expanded in its distal portion.

Male Genital System: Prostate extending to lower half of uterus and composed of cylindri-

cal acini. Distal vas deferens folded forming two circles, overlapping; one being smaller near insertion of vas deferens into cylindrical, elongated penis complex. Penis sheath muscular, $2\frac{1}{4}$ length of prepuce, with slight convexity on right side. Penis sheath inner sculpture with regular pustules over entire surface. Pustules raised, polyhedral, with circular, white, well marked center. Pustules increased in size in area joining with foreskin and becoming ellipsoidal in shape. Penis elongated, almost as long as prepuce. Prepuce inner sculpture showing two longitudinal narrow pilasters, diagonal arranged in V at top and side of pilaster. Penis retractor muscle attached to short and stout columellar muscle.

Digestive System

Radula (Figs. 16–19): Rows mean number 53 ($n = 3$; range = 46 to 58). Teeth per half row mean number 39 (except for the central tooth) ($n = 4$; range = 36 to 43). Central tooth asym-

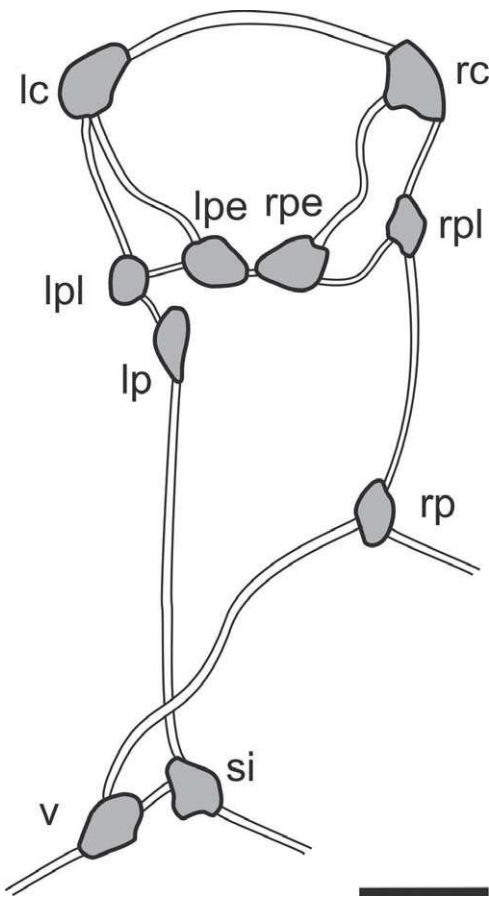


FIG. 20. Diagram of nervous system of *Chilina tucumanensis*. Abbreviations: lc, left cerebral; lpe, left pedal; lp, left parietal; lpl, left pleural; rc, right cerebral; rpe, right pedal; rp, right parietal; rpl, right pleural; si, subintestinal; v, visceral. Scale bar = 1 mm.

metrical, tricuspid, with triangular base as wide as high, mesocone most developed, sawlike edges in the two major cusps. First lateral tooth tetracuspid or tricuspid, with narrower base than apical part of tooth (cusp area), cusps wider than high, inner second cusp (tetracuspid) or mesocone (tricuspid) more developed. Second lateral tooth tetracuspid or tricuspid, with two inner cusps more developed, tooth base narrower than first lateral tooth. Last teeth having a thin base, can even have four to five cusps with similar development. Second inner cusp showing greatest development.

Nervous System (Fig. 20)

All connectives between ganglia relatively thin compared to both size of ganglia and system in general. Length of cerebral-pleural connective without significant statistical difference (7.3 vs. 6.2% of LWL; t_8 ; $p > 0.05$; Table 2). Right pleuroparietal connective running over the penis complex. Lengths of pleuroparietal connectives significantly different (4.52 vs. 13.79% of LWL; t_7 ; $p < 0.001$; Table 2). Parietal-subintestinal connective shorter than parietal-visceral connective (18.23 vs. 20.90 of LWL).

Distribution & Habitat

In addition to the known localities (Cochuna River in Tucumán and Río Primero in Córdoba), specimens were found on stones in two streams in the rain-forest area, the first with stony-sandy substratum and abundant macrophytes in the Las Cañas River and the other with sandy-stony substratum (Tucumán, Tafí del Valle).

Remarks

In the holotype and paratype material, two evident, well-developed columellar teeth were observed. In the rest of the specimens analyzed, however, the upper tooth does not show the same development and occupies an innermost portion of the columellar area. This last feature was also pointed out by Castellanos & Miquel (1980) in one of their figures. In the original description, the reproductive system was not mentioned. The analysis performed here showed differences between this species and *Chilina lilloi*, n. sp., with respect to both the general shape and the inner sculpture of the penis complex. The central tooth of the radula described by Castellanos & Miquel (1980) was tetracuspid with a larger development in one of lateral cusps; whereas in this specimen the tooth is tricuspid, with a more developed central cusp (mesocone).

Chilina portillensis Hidalgo, 1880

Chilina portillensis Hidalgo, 1880: 322; Smith, 1882: 846; Pilsbry, 1911: 547; Castellanos & Gaillard, 1981: 32 in part; Rumi et al., 2008: 82 in part.

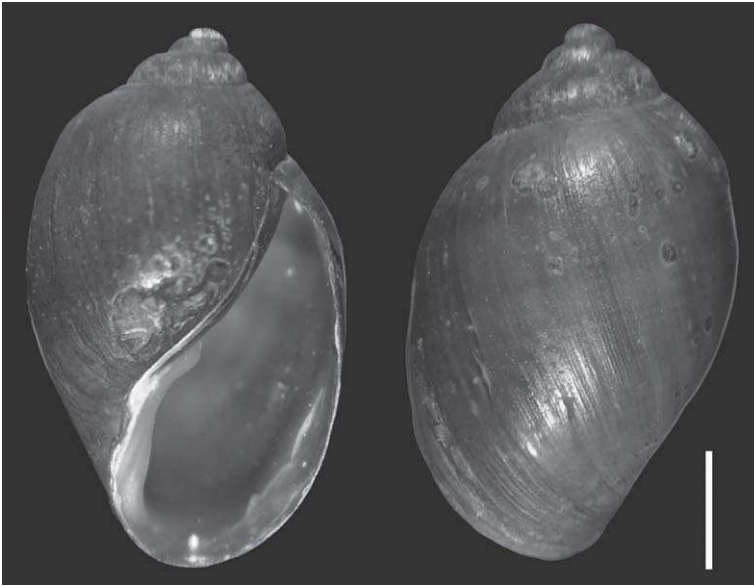


FIG. 21. Shells of *Chilina portillensis*. Scale bar = 2 mm.

Type Locality

Portillo Pass (Andean pass between Argentina and Chile).

Type Material

Syntype (MNHN 23108): from type locality, Paz & Membiela leg.

Other Material Examined

Argentina, Tucumán Province: IFML15538 (22 dry shells and 25 preserved specimens) and MLP 13345 (4 preserved specimens): Concepción Dept., Potrerillo stream; 27°21'6.04"S, 65°50'8.24"W; 706 m; Ovando, X. M. C. leg.

Etymology

Named for Portillo, the Andean pass.

Diagnosis

Shell with two columellar teeth, a little apart. Penis sheath $2\frac{1}{4}$ times the length of the prepuce. Penis sheath inner sculpture displaying star-shaped pustules. Pustules arranged on left and right margins. Penis sheath middle region smooth, without sculpture. Prepuce inner sculpture with two short, broad pilasters.

Description

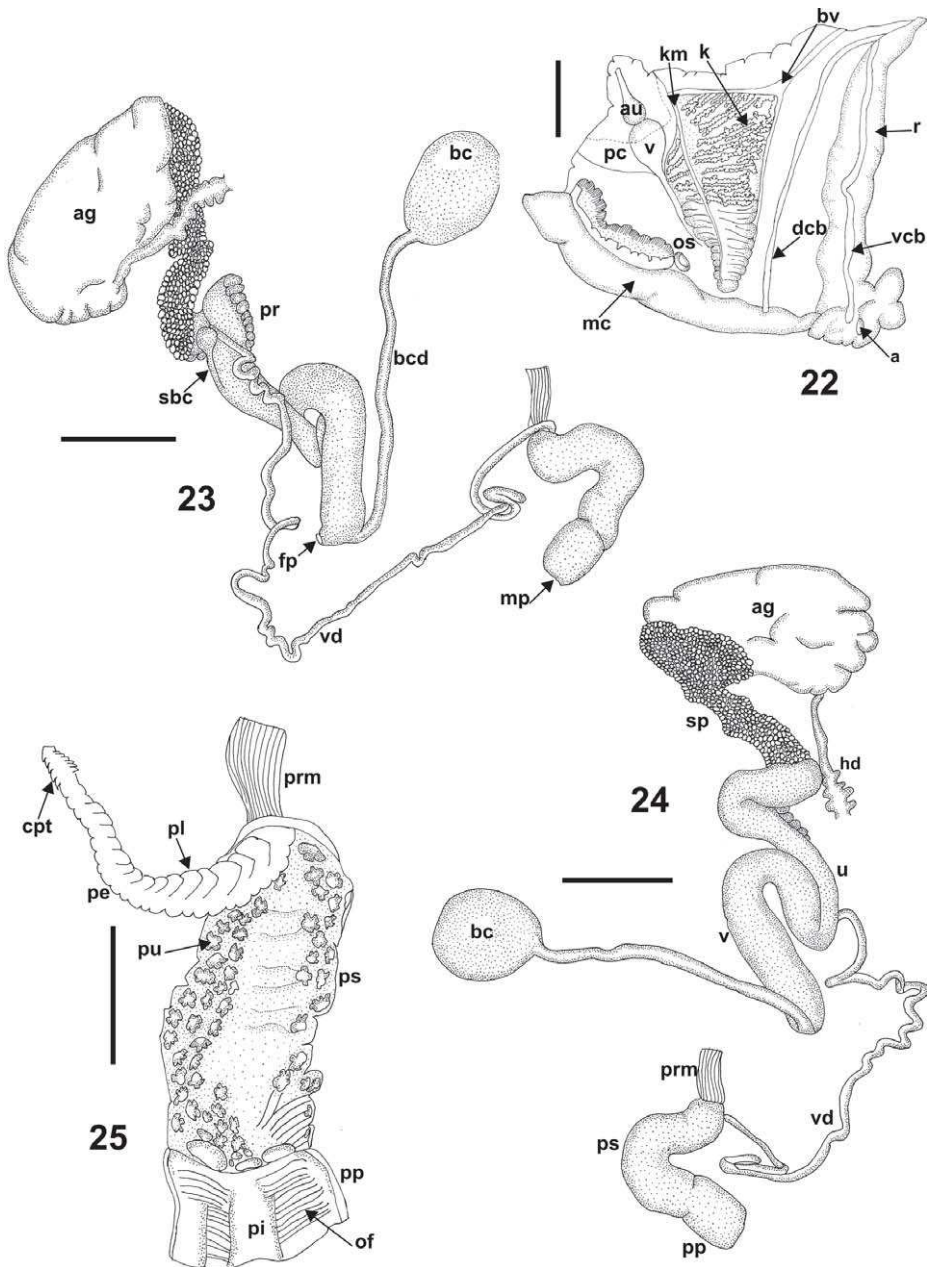
Shell (Fig. 21): Imperforate, ovate, rather solid, dull, almost opaque, smooth, olive-colored, with darker transverse bands slightly marked. Spire short, with eroded apex. Suture simple. Three, convex whorls, rapidly growing, last whorl somewhat distended. Aperture oval, pale reddish inside, with dark transverse lines. Columella thick, whitish, flattened at bottom. Two transverse columellar teeth slightly separated; lower tooth being larger than upper. Dimensions of four specimens of *C. portillensis* are shown in Table 1.

Pallial System (Fig. 22): Pallial system dark gray on entire surface. Mantle collar edge right margin with continuous line of light brown pigment. Kidney triangular, almost as long as whole lung-cavity roof. Pericardium $\frac{1}{2}$ kidney length.

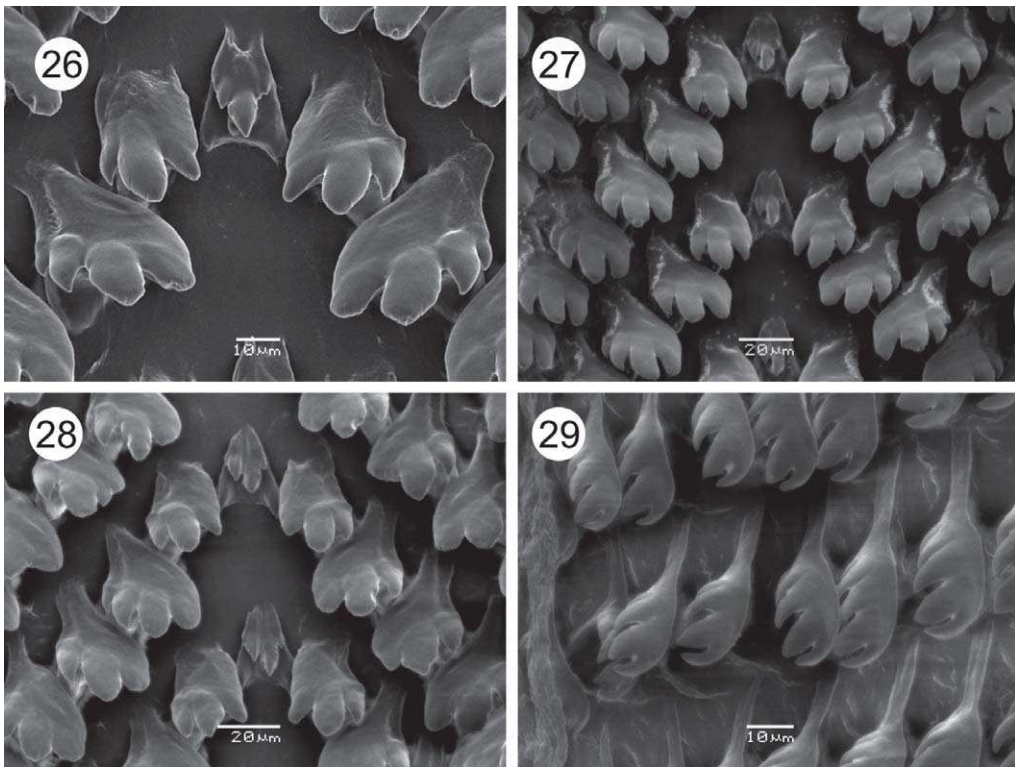
Reproductive System (Figs. 23–25)

Female Genital System: Bursa copulatrix duct length $3\frac{1}{4}$ times bursa copulatrix sac diameter. Secondary bursa copulatrix short, cylindrical, spherical (18% bursa copulatrix duct length), expanded in its terminal portion.

Male Genital System: Prostate extending to middle right side of uterus composed of cylindri-



FIGS. 22–25. *Chilina portillensis*. FIG. 22: Ventral view of pallial system dissected out. Scale bar = 2 mm; FIG. 23: Dorsal view of reproductive system without ovotestis. Scale bar = 1 mm; FIG. 24: Ventral view of reproductive system without ovotestis. Scale bar = 1 mm; FIG. 25: Penis inner wall. Scale bar = 1 mm. Abbreviations: a, anus; ag, albumen gland; au, auricle; bc, bursa copulatrix; bcd, bursa copulatrix duct; bv, blood vein; cpt, cuticular penis tooth; dcb, dorsal ciliar band; fp, female pore; hd, hermaphrodite duct; k, kidney; km, muscle band of kidney; mc, mantle collar; mp, male pore; os, osphradium; pc, pericardium; v, ventricle; r, rectum; sbc, secondary bursa copulatrix; sp, spermatheca; u, uterus; v, vagina; vcb, ventral ciliar band; vd, vas deferens; ve, ventricle.



FIGS. 26–29. Radula of *Chilina portillensis* from Potrerillo stream, Tucumán, Argentina. FIGS. 26–28: Central tooth and first lateral teeth. Scale bars = 10 µm and 20 µm; FIG. 29: Marginal teeth. Scale bar = 10 µm.

cal acini. Vas deferens sinuous all along, folding back on itself near insertion into penis complex. Penis complex cylindrical, curved down, inverted U-shaped. Penis sheath muscular, 2¼ prepuce length, with slight convexity toward right side. Penis sheath inner sculpture displaying star-shaped pustules arranged on penis sheath left and right margins. Penis sheath without sculpture on middle region. Pustules reduced in number to four or three, becoming ellipsoidal towards union with prepuce. Penis elongated, similar length to prepuce. Prepuce inner sculpture with two broad pilasters. Prepuce showing longitudinal grooves, oblique on both sides of pilasters.

Digestive System

Radula (Figs. 26–29): Rows mean number 54 ($n = 4$; range = 50 to 57). Teeth per half row mean 39 (except for the central tooth) ($n = 4$; range = 38 to 41). Central tooth asymmetrical,

bicuspid, triangular base, of equal length and width, central cusp more developed with serrate left edge. First lateral tooth tricuspid, with mesocone more developed, cusps equal in width and height. Second lateral tooth tricuspid or tetracuspid, with endocone and mesocone (tricuspid) or two inner cusps (tetracuspid) more developed. From tooth No 27 on, five cusps can be observed; the last two teeth, however, have four cusps.

Nervous System (Fig. 30)

All connectives between ganglia relatively thin compared to both size of ganglia and system in general. Length of cerebral-pleural connective significantly different (t_8 ; $p < 0.00004$) (6.98 vs. 5.34% of LWL; Table 2). Right pleuro-parietal connective running over penis complex. Lengths of pleuro-parietal connectives significantly different (t_8 ; $p < 0.0002$), left one being smaller than right (4.99 vs. 16.24% of LWL).

DISCUSSION

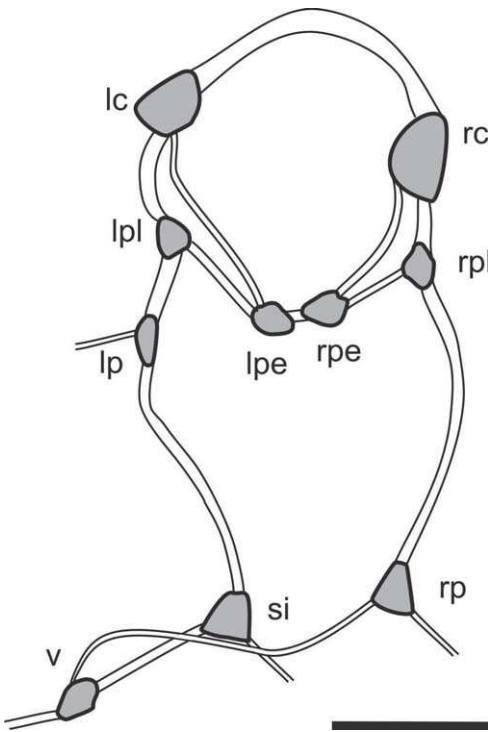


FIG. 30. Diagram of nervous system of *Chilina portillensis*. Abbreviations: lc, left cerebral; lpe, left pedal; lp, left parietal; lpl, left pleural; rc, right cerebral; rpe, right pedal; rp, right parietal; rpl, right pleural; si, subintestinal; v, visceral. Scale bar = 1 mm.

Distribution & Habitat

Chilina portillensis has been recorded in only one locality in the rain-forest area, Concepción Department in Tucumán. The specimen was collected from a stream with a stony substratum.

Remarks

Castellanos & Gaillard (1981), based on specimens deposited in La Plata Museum (MLP 2983 and 2967), mentioned the presence of *Chilina portillensis* in Jujuy and Salta provinces. Those materials have two well-developed columellar teeth, the spire well developed, and other proportions as in the description made by Hidalgo (1880).

This report provides anatomical and distributional information on the species of *Chilina* distributed in northwestern Argentina. On the basis of new anatomical information and the review performed, we have identified a new species within the genus, redescribed *Chilina portillensis*, and raised the subspecies *Chilina fluminea tucumanensis* to the rank of species.

Chilina parchappii, another species recorded from northwestern Argentina (Castellanos & Miquel 1980; Castellanos & Gaillard, 1981), has been found in only a single locality in Cachi, Salta Province (only two dry shells, one being damaged); whereas that species is very common and widely distributed in the southern pampas (Martín, 2003). Despite the exhaustive field work carried out in northwestern Argentina, we were unable to find any live specimens or dry shells and therefore could not further describe or provide new information on this species.

Castellanos & Miquel (1980) concluded that the differences in shell and radula morphology were not enough to consider *C. tucumanensis* at the species level. Nevertheless, in the present reevaluation we found new anatomical evidence to differentiate this taxon from *C. fluminea* (common species inhabiting the Del Plata Basin) at the species level. In *C. tucumanensis*, the vas deferens, before re-entering the haemocoelic space toward the penis complex, folds and forms two overlapping circles; whereas in *C. fluminea* the organ develops two or three large loops (Ituarte, 1997; Gutiérrez Gregoric, 2008). Another significant difference was found in the penis complex, being cylindrical and slender in *C. tucumanensis* but curved in *C. fluminea*. The penis sheath in *C. tucumanensis* is longer than the prepuce, whereas in *C. fluminea* the penis sheath can be almost the same length or else twice the prepuce (Gutiérrez Gregoric, 2008). Within the organization of the nervous system, the left subintestinal parietal connective in *C. fluminea* is longer than the right visceral one (Gutiérrez Gregoric, 2010), whereas in *C. tucumanensis* the reverse is true.

Furthermore, *C. lilloi*, n. sp., and *C. portillensis* can also be differentiated from *C. tucumanensis* by their radulae, nervous systems, and penis complex morphologies. The radula of

C. lilloi, n. sp., has the lowest number of tooth rows (44) as yet described for any known species of the genus in Argentina (Gutiérrez Gregoric, 2010). The radulae of *C. portillensis* and *C. tucumanensis* are similar to each other with respect to both the number of rows and the shape of the teeth. In comparison to *C. fluminea*, however, the number of rows in the *C. tucumanensis* radula is greater, though in *C. fluminea* and *C. tucumanensis* the first lateral tooth has three or four cusps. The number of teeth per half row in *C. tucumanensis* and *C. rushii* Pilsbry, 1896, is the lowest recorded for species of *Chilina*. In addition, the last lateral teeth in *C. lilloi*, n. sp., *C. fluminea*, and *C. rushii* can have up to seven cusps (Gutiérrez Gregoric, 2010).

The penis complex in *C. lilloi*, n. sp., is curved and U-shaped, unlike that of *C. tucumanensis*, where the complex is cylindrical and elongated but never curved. Furthermore, in contrast to the conclusions of Miquel (1987) regarding the homogeneity of the penis-complex inner-wall sculpture, the present results show conspicuous differences among the species under consideration. In *C. lilloi*, n. sp., this sculpture is divided into three well-delimited portions, but in *C. tucumanensis* the inner wall shows only one type of sculpture, consisting in polyhedral pustules in an orderly array. By contrast, in *C. portillensis*, the inner-wall sculpture shows star-shaped pustule arrangements on the left and right margins around a smooth middle portion. Another anatomical difference found in the genitalia concerns the natural folding of the vas deferens, which in *C. lilloi*, n. sp. develops three small loops upon reentering the haemocoelic cavity toward the penial complex, but in *C. tucumanensis* develops two round, overlapping folds.

The organization of the pallial system was similar in all the species analyzed, and this finding is in agreement with previous descriptions provided by Harry (1969) and Brace (1983) for other *Chilina* species of South America.

All the species studied here have the same general pattern of the nervous system. In *C. megastoma* Hylton Scott, 1958, however, a slight swelling, not forming a true ganglion, between the left pleural ganglion and the sub-intestinal ganglion is present (Ituarte, 1997). Another difference observed between the species from the northwest and the Del Plata Basin is in the relative lengths between the left pleural and the parietal ganglia. *Chilina lilloi*, n. sp., shows the lower relative length (55% of the

length of *C. fluminea*). Furthermore, in *C. lilloi*, n. sp., as well as in *C. fluminea*, the length of the pleuro-subintestinal connective is larger than that of the pleuro-visceral one. In the rest of the species (*C. gallardoi* Castellanos & Miquel, 1980; *C. iguazuensis* Gutiérrez Gregoric & Rumi, 2008; *C. megastoma*; *C. portillensis*; *C. rushii*; and *C. tucumanensis*), the reverse is true (Gutiérrez Gregoric, 2010). In addition, in *C. lilloi*, n. sp., the right pleuro-parietal connective runs in front of the penis complex, whereas in the rest of the species that connective runs either over or behind the complex.

The findings from this study have increased the species richness of *Chilina* within the area from three to four and as a consequence have augmented the total number of species of this genus inhabiting Argentina to 19. In northwestern Argentina *Chilina* species were collected in rivers and streams of the rain forests called Yungas. This ecoregion is considered one of the most diverse in the country and has the highest values of plant and vertebrate taxa endemism from Argentina (Brown & Kappelle, 2001).

According to Gutiérrez Gregoric (2008), the family Chiliniidae shows a marked distribution pattern in Argentina, with the highest species richness and abundance in the southernmost areas (i.e., the Patagonia). In northwestern Argentina *Chilina* species richness is notably low, with only scarce localities of occurrences registered for the genus. With respect to the normal distribution gradient characterized by a decrease in freshwater-gastropod diversity and species richness with increased latitude (Rumi et al., 2006), the Chiliniidae appear to have an inverse pattern. Nevertheless, the findings documented in this study of new records for *C. tucumanensis*, *C. lilloi*, and *C. portillensis* enhance the information on the distribution of this family within areas constituting possible objectives for future biogeographical analyses.

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