

Carrion consumption and its importance in a freshwater trophic generalist: the invasive apple snail *Pomacea canaliculata*

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Abstract. Trophic flexibility is a relevant trait in the potential for organisms to establish widely, maintain high abundances and spread after invasion. *Pomacea canaliculata* is an apple snail that feeds primarily on aquatic macrophytes, although it also consumes other trophic resources that likely play an important role in its persistence and contribute to its effects in invaded wetlands. In the present study we determined the ingestion rates in *P. canaliculata* for carrion and subsequent effects on growth, and performed field and laboratory experiments to investigate the mechanism of carrion detection. We observed *P. canaliculata* snails of all sizes feeding on carrion. The specific ingestion rates of carrion decreased with snail size and were 20 times lower than when feeding on lettuce. The growth rates of snails feeding only on carrion were 15–30% higher than those of fasting snails and 30% of those snails feeding on lettuce or lettuce and carrion. We found no evidence of distant chemoreception of carrion. The importance of carrion for *P. canaliculata* is mostly as an alternative resource when its preferred food is absent, and not as a complementary resource that could enhance growth. Nevertheless, the ability of *P. canaliculata* to profit from carrion may help explain its potential to establish widely and to have effects on aquatic vegetation.

Additional keywords: Ampullariidae, gastropod, invasion, trophic ecology.

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Introduction

Trophic flexibility is recognised as an important trait in the potential for animals to establish populations and spread in areas where they are not native (Weis 2010; Hayden *et al.* 2014; Hill *et al.* 2015) because it increases the probability of finding unexploited resources in the recipient ecosystems. Specialised carrion-feeding arthropods and molluscs are a widespread and important guild in marine environments, but they do not seem to have any freshwater equivalents (Dillon 2000; Fenoglio *et al.* 2014). Nevertheless, many species of freshwater snails are able to profit opportunistically on carrion (Dillon 2000; Lyabzina 2013).

Pomacea canaliculata (Lamarck, 1822) is an apple snail that has been translocated to many areas around the world. Interest in the trophic ecology of *P. canaliculata* has increased as it has become a pest on aquatic crops because of its voracious consumption of aquatic macrophytes, its main food resource (Cowie 2002). As a pest, this snail has been responsible for strong effects on agriculture and wetland trophic states (Horgan *et al.* 2014), as well as economic losses (Nghiem *et al.* 2013). Further, it is responsible for emergent health problems as an intermediate host of parasites (Lv *et al.* 2011). *P. canaliculata* is able to persist at high densities in invaded wetlands even after it

has depleted all the submerged and floating macrophytes (Carlsson *et al.* 2004), probably by consuming alternative trophic resources through diverse feeding mechanisms (Saveanu and Martín 2013, 2014, 2015).

Carrion, defined here as the corpses or remains of animals of a size that a snail cannot ingest in one piece, could represent an alternative trophic resource, obtained by the snail by shredding–scraping using its jaws and radula, as well as pedal surface collecting (Saveanu and Martín 2013). Many ampullariids have been observed to consume carrion under laboratory conditions (e.g. Cazzaniga and Estebenet 1984; Aditya and Raut 2001; Hayes *et al.* 2015). In addition, *P. canaliculata* feeds on recently dead small fish (Alonso and Ageitos de Castellanos 1949), bryozoans (Wood *et al.* 2006) and on the adults, juveniles and eggs of other snails (Cazzaniga and Estebenet 1984; Kwong *et al.* 2009). Karraker and Dudgeon (2014) reported *P. canaliculata* consumption of amphibian eggs and Saveanu and Martín (2014) found it feeding cannibalistically on its own egg masses. To date, only laboratory studies have documented the consumption of animal materials, with the exception of two field studies that found scarce invertebrate remains in *P. canaliculata* gut contents (Kwong *et al.* 2010; Ocon *et al.* 2013).

Different studies have reported that the rate of ingestion of macrophytes by *Pomacea* spp. is highly dependent of size (Carlsson and Brönmark 2006; Boland *et al.* 2007; Tamburi and Martín 2009), as with other trophic resources, such as neuston and its own eggs (Saveanu and Martín 2013, 2014). In addition, *P. canaliculata* also exhibits sex-specific differences in ingestion and assimilation rates when feeding on lettuce by shredding–scraping (Tamburi and Martín 2009), but not when feeding on simulated neuston by pedal surface collecting (Saveanu and Martín 2013). The patterns of carrion feeding remain uninvestigated.

Andrews (1965) did not report any enzymes involved in protein digestion in *P. canaliculata*, but a diverse array of proteases was recently found in snails fed high-protein food (Godoy *et al.* 2013). Processed animal proteins (e.g. casein, gelatin and fish meal) enhance growth when offered as a complementary component to a plant-based diet (Mendoza *et al.* 1999; Ramnarine 2004), despite the primarily macrophytophagous habit of *Pomacea* snails. However, there are no studies on assimilation and growth when feeding on high-protein natural sources, like the flesh or corpses of aquatic animals.

Carrion is a ubiquitous trophic resource in natural waterbodies (Lyabzina 2013; Fenoglio *et al.* 2014). However, its availability is highly unpredictable in time and space, and decreases quickly with its consumption (Daleo *et al.* 2005) and physical and chemical degradation, especially in lotic environments (Fenoglio *et al.* 2014). Hence, its rapid detection can be important for obligate and facultative carrion feeders when other resources are scarce (Himmelman 1988). In laboratory experiments, *P. canaliculata* was able to detect the presence of macrophytes (Estebenet 1995) and conspecific snails (Takeichi *et al.* 2007) from short distances by chemoreception. As far as we know, it has not been established whether the same mechanism may be used to detect carrion.

The primary aim of the present study was to estimate the importance of carrion as an alternative or complementary trophic resource to macrophytes for *P. canaliculata* from within its native range at the Río de la Plata basin, which is probably the source from which they have been translocated to South-east Asia (Hayes *et al.* 2008). Information about the trophic flexibility of this apple snail obtained from the native range would help us understand how it has become established in a diverse range of natural and managed habitats in its invaded range. In particular, we wanted to determine the rate of ingestion of carrion relative to size and sex, and its effects on growth under laboratory conditions. In addition, we performed a field experiment to investigate the existence of a carrion-detection mechanism. Finally, under laboratory conditions, we tested whether previous experience with carrion increases the ability to detect and use this resource.

Materials and methods

Origin of snails, rearing conditions and food items

P. canaliculata is the only apple snail that inhabits the Encadenadas del Oeste basin, Buenos Aires Province, Argentina (Martín *et al.* 2001). The identity of the snails from the populations used in previous studies and in the present study has been confirmed by Hayes *et al.* (2012).

Adult snails were hand collected in streams from the Encadenadas del Oeste basin and reared in collective 20-L aquaria. Juvenile snails, obtained in the laboratory from egg masses laid by field snails, were reared individually in 200-mL aquaria until used in the experiments. All snails were maintained and the experiments performed in a rearing room at $25 \pm 2^\circ\text{C}$, under a 14-h light–10-h dark cycle. Unless stated otherwise, CaCO_3 -saturated tap water was used and snails were fed on fresh lettuce. Aquaria were cleaned and the water changed once a week.

During each experiment, both in the laboratory and the field, fresh lettuce was offered as a surrogate for a nutritive and palatable macrophyte and pieces of fish flesh (0.8 ± 0.1 g of thawed hake fillet, kept frozen at -18°C until 2 h before the beginning of the trials) were offered as a surrogate for carrion. The freezing and thawing procedure was chosen both to maintain the properties of the hake fillet and to increase the leakage of intracellular fluids to facilitate detection by chemoreception. The choice of lettuce and hake fillets as surrogates for macrophytes and carrion respectively was made, first, because they represent available standardised sources of food and, second, to avoid any variability due to the use of different materials.

Carrion feeding rates: ontogenetic and sex-specific variation

The carrion ingestion rate was studied in the laboratory over a wide range of snail sizes (1.99–56.3 mm, including hatchlings, juveniles, males and females) from El Huáscar stream ($36^\circ55'50''\text{S}$, $61^\circ35'48''\text{W}$). All snails used were measured from the apex to the furthest point of the aperture (shell length (SL), mm), using a stereoscopic microscope with a micrometre lens if the snail was smaller than 10 mm SL or with a Vernier calliper if it was larger. To avoid the amounts of carrion ingested being undetectable on the scale used (precision 0.1 mg), carrion ingestion in snails smaller than 15 mm was estimated for groups of three to 21 snails of similar size (CV for shell length <13%; Tamburi and Martín 2009). Accordingly, live weight (LW, g) was recorded individually for snails larger than 15 mm SL and as a group for smaller individuals, after the snails had crawled onto a dry plastic surface to drain water from the pallial cavity (Tamburi and Martín 2009, 2016).

Snails (alone or in groups) were maintained over a 24-h period in 3-L glass aquaria (with tap water but without additional CaCO_3) with lettuce and carrion to homogenise the satiation level and to give the snails experience with carrion. They were then maintained without any food for another 24 h. The aquaria were cleaned and the water changed after each 24-h period. A weighed piece of carrion well in excess of individual ingestion capabilities (~ 1 g) was then provided to each aquarium as the only available resource for 24 h. The snails feeding on carrion (i.e. moving the radula and jaws on it) were recorded during the first hour. After 24 h, the unconsumed carrion remains were drained and weighed to obtain the amount of carrion consumed by each snail or group. To correct for hydration effects of carrion remains, the drained weight of similar pieces of carrion after 24-h submergence in control aquaria without any snails ($n=8$) was used.

Individual daily specific ingestion rates (SIR, $\text{g g}^{-1} \text{day}^{-1}$) were estimated as the corrected weight of carrion consumed by the snail or group in 24 h divided by the live weight of the

snail or group. The observations ($n = 80$ snails or groups) were performed on 6 days over 2 months, with 12–15 snails or groups tested on each occasion; each snail was used only once.

To evaluate ontogenetic variation in carrion ingestion, the simple allometry model ($SIR = a \times SL^b$, where a is the scale factor and b is the allometric coefficient) was adjusted by a least-squares regression analysis of the variables, which were logarithmically transformed to linearise the relationship ($\log_{10} SIR = \log_{10} a + b \times \log_{10} SL$). Differences in carrion ingestion rates between males and females were compared by t -test after being square root transformed due to rejection of homoscedasticity (Levene's test, $P < 0.04$).

Growth of juveniles fed on carrion

To estimate the effects on growth of carrion consumption as an alternative or complementary resource to macrophytes (lettuce), juvenile snails (15.4–19.6 mm SL) were fed with one of the following four food regimens for 4 weeks: lettuce (L), lettuce and carrion (L+C), carrion (C) or no food (NF). The L and NF treatments were used as controls for maximum and minimum growth respectively. These snails ($n = 10$ in each food regimen) were also from El Huáscar stream. Juveniles were used because of their higher growth rates relative to adults (Saveanu and Martín 2014) and the lower measurement error in their length and weight relative to hatchlings.

Snails were reared individually in 3-L glass aquaria with tap water without additional CaCO_3 . Each week, the aquaria were cleaned and the water changed, and SL and LW were recorded as described above for snails larger than 15 mm. Each aquarium had a 1.0-cm mesh grid 2 cm from the bottom to avoid coprophagy between the weekly water changes. Specific growth rates of SL (SLR, $\text{mm mm}^{-1} \text{day}^{-1}$) and LW (LWR, $\text{g g}^{-1} \text{day}^{-1}$) were calculated as the difference between the final and initial values of each week, divided by the initial value. At the end of the experiment, snails were killed by immersion in boiling water and then each snail was dried at 70°C for 48 h to obtain the total dry weight (TDW, g). Snails were then incinerated at 600°C for 4 h to obtain the ash-free dry weight (AFDW, g).

Both carrion and lettuce were provided *ad libitum* in pieces. Each piece of carrion was weighed at the beginning of the experiment and was hung from a fish hook inside the aquarium in contact with the grid at the bottom. After 1 week, carrion remains were drained and weighed to obtain the weight of the carrion consumed. Hydration effects were corrected to calculate daily SIR, as in the previous experiment ($n = 4$ aquaria without snails).

Initial SL and LW and final SL, LW, TDW and AFDW were compared using one-way analysis of variance (ANOVA); least significant difference (LSD) tests were performed to find differences between regimens. The initial and final SL and LW of each food regimen were compared by pairwise comparisons. Some variables were transformed after rejection of Levene's test of homoscedasticity ($P < 0.05$). Growth rates (SLR and LWR) were compared using repeated-measures ANOVA, with food regimens as the main factor and weekly measurements as the within-subject factor. The degrees of freedom were corrected by Greenhouse–Geisser's correction due to a significant non-sphericity in LWR (Mauchly's W -test,

$P < 0.05$). Specific carrion ingestion rates were compared by t -test between the L+C and C regimens.

Carrion detection in the field

To determine whether *P. canaliculata* uses a specific mechanism to detect carrion, and the relationship of such a mechanism with the one used for the detection of vegetal material, an experiment was performed in El Huáscar stream using recently thawed carrion. El Huáscar stream is 20–25 m wide and 0.4–0.5 m deep and has a stony bottom with scattered mats of *Myriophyllum*.

Circular wire quadrats (30-cm diameter) were deployed along 60 m of the shore in five blocks. Each block consisted of three quadrats 20 cm apart aligned perpendicularly to the stream flow. Each quadrat was divided into upstream and downstream halves; a lace tied from the centre of the quadrat was used to determine stream flow direction. The baits of each block, lettuce, carrion and control, were randomly located and tethered at the centre of each quadrat. Water temperature and current velocity in each block were recorded using a thermometer and a digital flowmeter (FP101 Global Water, Xylem Inc., Gold River, CA, USA).

Each block was observed by one operator over 3 h, recording the snails' movements in the stream. The number of snails that entered into the different quadrats and the residence time of each snail were recorded and compared by one-way ANOVA. In addition, the direction of entrance into the quadrat (upstream or downstream) was recorded for each snail and these frequencies were compared using the Chi-Square test.

Effect of previous experience in carrion detection

To assess whether previous experience of adult *P. canaliculata* with carrion increases their capacity to detect, or their attraction towards, carrion or the use of carrion as a trophic resource, an experiment was performed using T-maze aquaria (Burela and Martín 2014). In each trial, the behaviour of carrion-experienced *v.* carrion-naïve snails was recorded using carrion as bait in one arm and no bait in the other. Seven days before the trial, two snails were individually placed in 3-L glass aquaria with tap water, one with lettuce only (carrion-naïve snail) and the other with lettuce and carrion (carrion-experienced snail); 24 h before the initiation of the trial, each snail was transferred to a new aquarium without food. A total of 21 pairs of snails (31.3–52.3 mm SL; 18 males, 24 females) collected from the Pigüé-Venado channel were tested. Blocks of two trials were performed sequentially (three times per day and three times per week).

The piece of carrion offered as bait was maintained at 25°C for 24 h before observation, and placed in a filter paper bag to allow smell diffusion and to prevent direct contact of the snail with the carrion and its consumption. The bag with carrion was hung in one of the opposite arms of the T-maze and an empty bag was hung in the other arm as a control; the position of carrion in the arms of each T-maze, and snails between both T-mazes, were randomly determined before the experiment; sexes were balanced between carrion-experienced and carrion-naïve snails. The experiment was performed in a room at 25°C during diurnal hours, without direct sunlight but with a lamp over each maze to homogenise illumination.

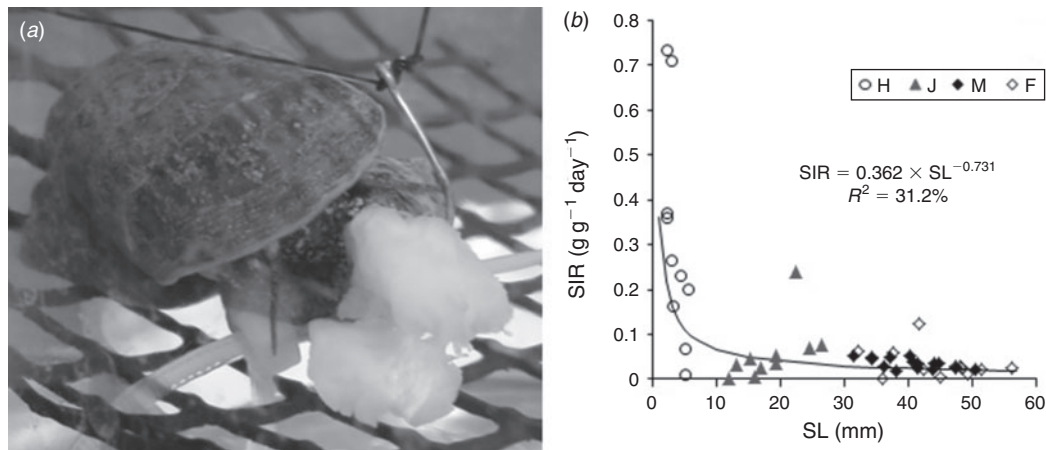


Fig. 1. Carrion consumption by *Pomacea canaliculata*. (a) An adult feeding on carrion under laboratory conditions. (b) Specific ingestion rates (SIR) for carrion relative to shell length (SL) for hatchlings (H), juveniles (J), males (M) and females (F).

The T-maze was completely covered with a nylon sheet before each trial to avoid the presence of mucous tracks or odour traces in the following trials and was filled with tap water at $25 \pm 1^\circ\text{C}$ (depth 6 cm). Five minutes after the bags had been placed in the ends of the arms, one snail of each type was put at the base of each T-maze and both mazes were videotaped from above for 2 h. An observation period of 1 h was defined for each snail after its foot was in full contact with bottom; this period is long enough for an adult snail (40 mm SL) to cover more than seven times the total length of both arms and the base at an average crawling velocity of 0.24 cm s^{-1} (Seuffert and Martín 2010).

During the observation period of each trial, the first choice between the opposite arms, contact with the bag with carrion and the number of entries into each arm were recorded and compared between the carrion-experienced and carrion-naïve snails using Fisher's exact tests. In addition, the time in the base of the T-maze and in each arm, with and without carrion, and the time spent using this food resource (considered as the time in contact with the bag with carrion) were recorded. The significance of differences between the carrion-experienced and carrion-naïve snails was analysed using *t*-tests.

Results

Spontaneous field observations of carrion consumption

During the course of the different sampling campaigns in the field, we encountered two instances of carrion consumption. Interestingly, in both cases the snails were adults partially or totally out of the water while eating the flesh remains attached to an operculum of *P. canaliculata* or to a bird corpse.

Carrion feeding rates: ontogenetic and sex-specific variations

Snails of all sizes performed radular and jaw movements on carrion (93% of adults (Fig. 1a), 56.5% of juveniles and 69.2% of hatchlings). Hatchlings had the highest SIR ($0.313 \pm 0.244 \text{ g g}^{-1} \text{ day}^{-1}$; $n = 10$; Fig. 1b), whereas juveniles and adult snails has lower rates with less variation (0.058 ± 0.068

($n = 10$) and $0.035 \pm 0.024 \text{ g g}^{-1} \text{ day}^{-1}$ ($n = 27$) respectively). The regression model for carrion ingestion rates relative to SL was significant, although it explained a low percentage of the variability ($F_{1,44} = 19.940$; $P < 0.001$; $R^2 = 31.2\%$). SIRs decreased significantly with SL ($b = -0.731$; 95% confidence interval (CI) $-1.061, -0.401$) and did not differ between sexes ($F_{1,25} = 0.643$; $P = 0.430$).

Growth of juveniles fed on carrion

Initial SL and LW did not differ significantly between different food regimens ($F_{3,36} = 0.552$, $P = 0.650$; and $F_{3,36} = 0.816$, $P = 0.494$ respectively). None of the snails (including those without food) died during the experiment. Snails reared with food for 4 weeks (L, L+C or C) grew in terms of both SL and LW (paired *t*-tests, $P < 0.001$); snails without food (NF) grew in SL ($P < 0.001$) but not LW ($P = 0.625$).

Growth rates relative to shell length differed significantly between different food regimens, as well as over the different weeks of the experiment (between subjects $F_{3,36} = 70.866$, $P < 0.001$; within subjects $F_{3,108} = 166.371$, $P < 0.001$); the same pattern of effects was detected for LW (between subjects $F_{3,36} = 60.932$, $P < 0.001$; within subjects $F_{1,63} = 139.705$, $P < 0.001$). Comparing the 95% CI, we observed that snails fed on carrion grew significantly more in SL and LW compared with those fasting only in the first week (Fig. 2a, b). Carrion did not produce any significant additional growth in SL or LW relative to lettuce. Although growth rates relative to the LW of snails reared without food were negative in the last 2 weeks of the experiment, none of them was significantly different from zero (*t*-test, $P > 0.242$). Fasting snails only grew in SL in the first week (*t*-test, $P = 0.025$).

Final SLs of snails fed on carrion did not differ significantly from those fasting, and in both cases was significantly lower than that of snails reared with lettuce, with or without carrion (Fig. 3a). Conversely, LW, TDW and AFDW of snails only fed with carrion were significantly higher than those of fasting snails and significantly lower than those of snails fed on lettuce

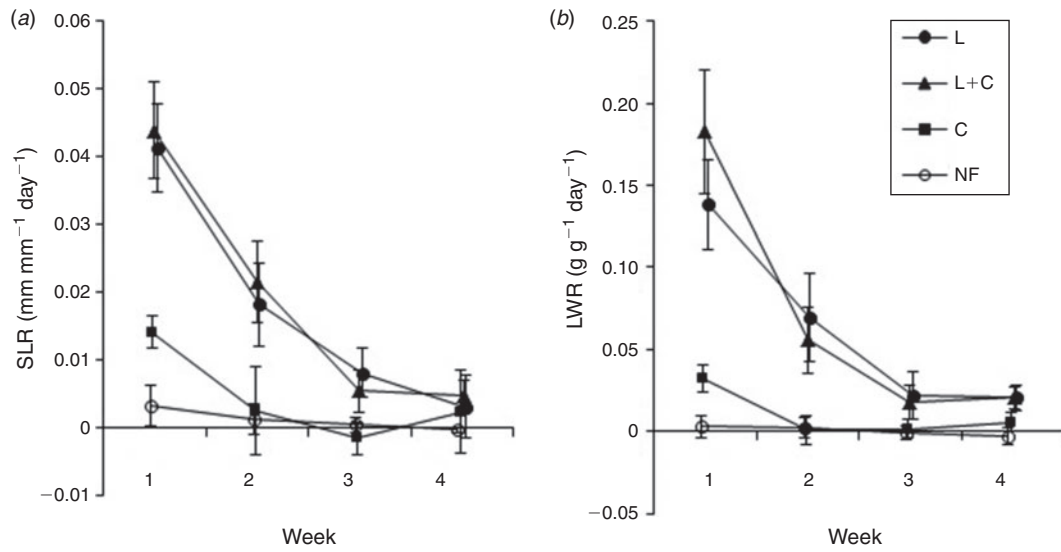


Fig. 2. Mean ($\pm 95\%$ confidence intervals) of specific growth rates for juveniles of *Pomacea canaliculata* under four different food regimens: L, lettuce; L+C, lettuce and carrion; C, carrion; NF, no food. (a) Shell length rate (SLR) and (b) live weight rate (LWR).

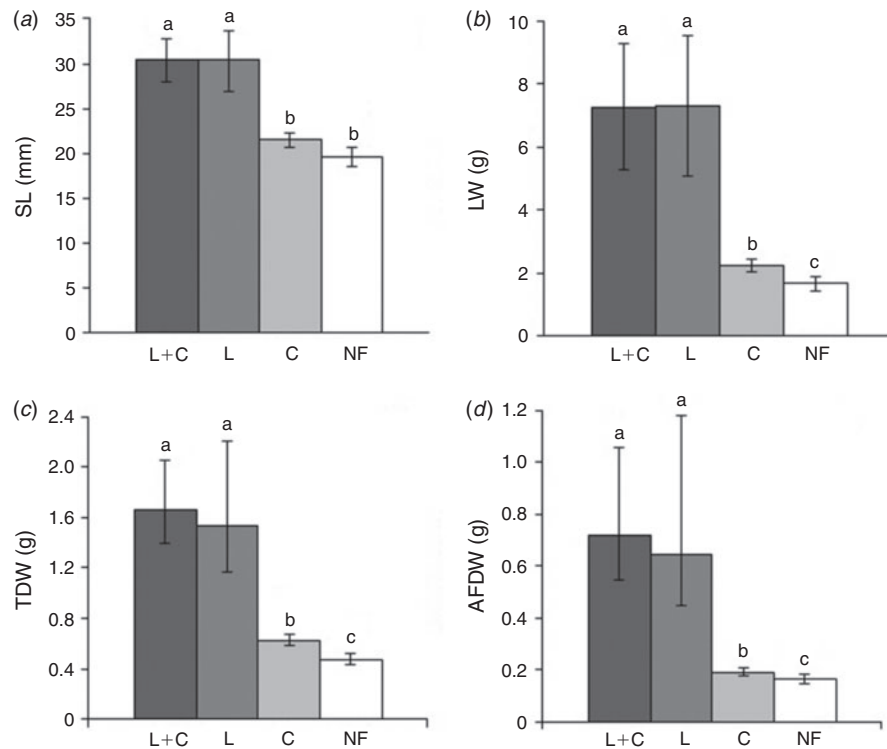


Fig. 3. Mean ($\pm 95\%$ confidence intervals) of final sizes for juveniles of *Pomacea canaliculata* under four food regimens: lettuce (L), lettuce and carrion (L+C), carrion (C) and no food (NF). Different letters indicate significant differences between mean values (LSD test, $P < 0.05$). (a) Shell length (SL); (b) live weight (LW); (c) total dry weight (TDW); and (d) ash-free dry weight (AFDW). SL was \log_{10} transformed, whereas TDW and AFDW were transformed to their inverses; the mean values are shown back-transformed.

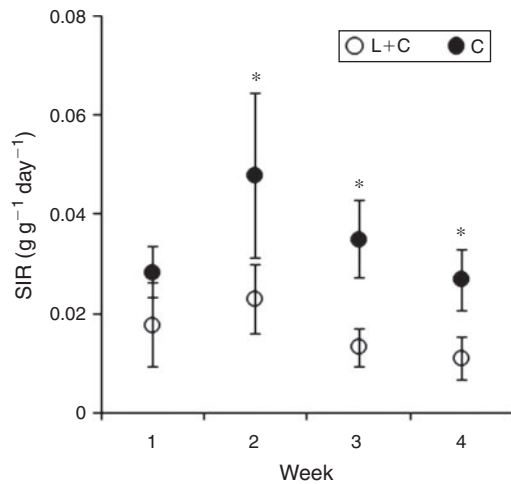


Fig. 4. Mean ($\pm 95\%$ confidence intervals) of carrion daily specific ingestion rates (SIR) of juvenile *Pomacea canaliculata* over 4 weeks under two different food regimens: lettuce and carrion (L+C) and carrion (C). Asterisks indicate a significant difference between the two means in each week ($P < 0.05$).

(Fig. 3b–d); the addition of carrion to lettuce did not significantly affect any of the variables considered.

Carrion was consumed even in the presence of lettuce. However, when carrion was the only resource, SIRs were higher than when lettuce was also present (Fig. 4); these differences were marginally significant in the first week ($P = 0.051$) and highly significant in the following weeks ($P < 0.001$).

In the experiment of carrion feeding rates, the physical state of carrion remained almost unchanged after 24 h at 25°C. However, in the experiment of growth rates, after 1 week in the aquaria, carrion lost turgidity and was covered by a mucilaginous layer. Despite these differences, the ingestion rates were similar in both experiments, indicating that the decomposition of carrion did not affect its consumption.

Carrion detection in the field

During the field experiment, the water temperature was $26 \pm 2^\circ\text{C}$, water depth was 28.3 ± 0.5 cm and current velocity was lower than 0.1 m s^{-1} . Approximately 80 snails entered into the quadrats with the different baits. The presence of bait or bait type did not affect the number of snails entering each quadrat ($F_{2,12} = 0.128$; $P = 0.881$). Residence time in each quadrat did not differ between blocks ($F_{4,6} = 0.560$; $P = 0.697$) or between baits ($F_{2,6} = 1.727$; $P = 0.256$). Baits did not affect the frequency of snails that entered quadrats from upstream or downstream of the bait (χ^2 tests, $P > 0.590$ in all cases). No snail consumed lettuce in the quadrats and only three of 30 snails ingested carrion in the carrion quadrats.

Effect of previous experience in carrion detection

None of the variables analysed relative to carrion detection and its use as food differed between carrion-experienced and carrion-naïve snails ($P > 0.100$ in all cases). The number of snails that entered into the left or right arm did not differ between these types of snails ($P > 0.229$).

Discussion

The observations of the present study represent the first reports of *P. canaliculata* feeding on carrion in natural waterbodies. In addition, our laboratory observations highlight the acceptance and consumption of carrion by snails of a wide range of sizes and the importance of carrion for growth when it is the only trophic resource available.

The ontogenetic decrease found in specific rates of carrion ingestion was similar to the pattern found for lettuce (Tamburi and Martín 2009) and the snails' own eggs (Saveanu and Martín 2014), as well as the pattern of specific capture rates of neuston and surface material by pedal surface collecting (Saveanu and Martín 2013). The regression model explained a low percentage of the variation, probably because of the low absolute amount of carrion consumed by small snails, which was almost undetectable in some cases with the scale used. The results indicate that snails of 5 mm SL had ingestion rates of $0.11 \text{ g g}^{-1} \text{ day}^{-1}$, whereas juveniles of 25 mm SL and adults of 45 mm SL had ingestion rates that were one-quarter and one-fifth of those in the smaller snails (0.03 and $0.02 \text{ g g}^{-1} \text{ day}^{-1}$ respectively).

Carrion ingestion rates were lower than those observed for plant material. In no-choice feeding trials, the SIR of carrion for a snail of 40 mm SL ($0.02 \text{ g g}^{-1} \text{ day}^{-1}$) was one-twentieth that of lettuce ($0.40 \text{ g g}^{-1} \text{ day}^{-1}$; Tamburi and Martín 2009). Moreover, although carrion was consumed when lettuce was available, the ingestion rates were one-quarter of the rates when it was the only available food. Conversely, carrion ingestion rates were fivefold higher than when snails were feeding on their own eggs ($0.004 \text{ g g}^{-1} \text{ day}^{-1}$; Saveanu and Martín 2014), a trophic resource that allows juveniles to grow despite the calcareous eggshells and a perivitelline fluid that is toxic to rats (Dreon et al. 2014).

Carrion ingestion rates increased significantly in the second week and then returned to values similar to those seen in the first week. This delayed peak of ingestion rate may be due to the need to become accustomed to a regimen with a high protein content (near 20% protein content; Izquierdo Córser et al. 2000) relative to a low-protein diet based on lettuce (8.9% protein; Granval and Gaviola 1991). The subsequent decrease found in the last 2 weeks perhaps indicates that intracellular digestion of the carrion ingested in previous weeks has not been completed, because animal protein is difficult to digest and assimilate by *Pomacea* snail species (Mendoza et al. 1999, 2002; Ramnarine 2004).

The consumption of carrion as the only resource allowed juveniles of *P. canaliculata* to increase their body mass between 15 and 34% relative to fasting snails; the lack of differences in SL may be attributed to the addition of new shell material during fasting, probably by using stored organic substances and incorporating calcium from the water (Zischke et al. 1970). Conversely, juvenile snails grew 30% less when fed only on carrion compared with those fed on lettuce, but the addition of carrion to lettuce did not produce an increase relative to feeding of lettuce alone. However, in other *Pomacea* species, the addition of 25–75% animal proteins (fish meal) to lettuce–spinach resulted in higher growth rates (Mendoza et al. 2002). In the latter case, the food was pelleted and the animal component was thoroughly mixed with the vegetal one. Perhaps the differences between the results of growth trials in the present

study and the study of [Mendoza et al. \(2002\)](#) are due to the choice between lettuce and carrion or the higher digestibility of fish meal relative to fish flesh. The results of the present study support the idea that carrion would mostly provide an alternative resource when preferred food is absent and not a complementary food that can enhance growth in addition to the preferred food (at least when the availability of the preferred food is very high).

In the field experiment, current velocity and water temperature were within the range in which snails are normally active ([Seuffert and Martín 2010, 2012](#); [Seuffert et al. 2010](#); [Saveanu and Martín 2013](#)). The lack of differences in the number of snails entering the quadrats with baits and the control quadrats indicate that snails were not able to detect the bait or were not attracted to it, even when the carrion was partially decomposed. *P. canaliculata* is able to detect macrophytes and conspecific individuals from short distances (up to 30 cm; [Estebenet 1995](#); [Takeichi et al. 2007](#)), but when preying on other snails they apparently detect by random crawling and direct contact ([Kwong et al. 2009](#)). However, when fresh egg masses were submerged, *P. canaliculata* snails congregated and consumed them after a few minutes ([Saveanu and Martín 2014](#)), indicating that they are able to detect them from distance. The egg masses are an alternative trophic resource that is apparently more frequently encountered and consumed than previously thought. This would indicate that previous experience with specific trophic resources is necessary to prompt a response, although our T-maze experiments did not support this hypothesis; moreover, snails in the field feed on fish bait when they come in contact with it for the first time.

P. canaliculata is primarily a macrophytophagous snail and a neuston feeder ([Saveanu and Martín 2013, 2015](#)). However, it is also an opportunistic feeder on different trophic resources (submerged eggs, carrion and other invertebrates) that it randomly encounters while wandering through its habitat in search of macrophytes, mates and sites for oviposition or aerial respiration. Although it apparently does not use any specific search mechanisms for food of animal origin, such food is readily consumed upon encounter, providing the snails with some nutritional and energetic benefits without any extra cost in searching. Carrion consumption probably passes unnoticed in the turbid waters that these snails commonly inhabit ([Martín et al. 2001](#)).

The ability of *P. canaliculata* to sustain growth using alternative resources, such as carrion, may help explain the snails' persistence in high densities even after populations have consumed all the submerged and floating macrophytes ([Carlsson et al. 2004](#)). Moreover, this ability, together with the starvation endurance recently reported (more than 100 days with no mortality; [Tamburi and Martín 2016](#)), may explain, in part, the persistent effects that *P. canaliculata* has on aquatic vegetation. Snails that are starving, or only just surviving on alternative trophic resources, may deter any regrowth of macrophytes from inedible organs or from the seed bank and hence may greatly reduce the resilience of the macrophyte communities.

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