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ARTÍCULO DE INVESTIGACIÓN / RESEARCH ARTICLE

SPATIAL AND TEMPORAL DISTRIBUTION OF TWO ANACRONEURIA SPECIES (PLECOPTERA: PERLIDAE) IN A NEOTROPICAL RIVER OF COLOMBIA

Distribución espacial y temporal de dos especies de Anacroneuria (Plecoptera: Perlidae) en un río Neotropical de Colombia

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ABSTRACT

Ecological studies on habitat use patterns on the distribution of Plecoptera species facilitate the understanding of this population dynamics. Our objective was to determine the distribution and abundance of *Anacroneuria caraca* Stark, 1995 and *A. marta* Zúñiga and Stark, 2002 in different microhabitats in the Gaira River during an annual hydrological cycle. Species were identified according to a nymph-adult association based on the male genitalia of mature and pre-emergent nymphs. Univariate analyses were performed to compare the abundance of the two species during the samplings and microhabitats. The highest abundance of both species and the number of pre-emergent nymphs were observed during the dry period and the beginning of the rainy period. Differences were not found in species abundances between samplings or microhabitats ($p > 0.05$); however, *A. marta* presented the highest abundance in the study. Regarding microhabitats, the highest abundance was recorded in the leaf litter with 638 individuals, while similar values were observed in the slow current pool and riffle with 194 and 294 individuals, respectively. This study showed that the leaf litter microhabitat is one of the most important Gaira River niches to both species. Additionally, similar abundance of the two species in the leaf litter may suggest the availability of sufficient resources (food and refuge) and the superposition of niches between them.

Keywords: microhabitats, population dynamics, rainfall regime, stoneflies.

RESUMEN

Los estudios ecológicos relacionados con los patrones de uso del hábitat en la distribución de especies de Plecoptera, facilitan la comprensión de su dinámica poblacional, aspectos poco conocidos en ecosistemas acuáticos tropicales. El objetivo de este trabajo fue determinar la distribución de ninfas de *Anacroneuria caraca* Stark, 1995 y *A. marta* Zúñiga y Stark, 2002 entre microhábitats en el río Gaira durante un ciclo hidrológico anual (julio 2014 – junio 2015). Las especies se identificaron de acuerdo con asociaciones ninfa-adulto mediante el reconocimiento de los genitales masculinos de ninfas maduras preemergentes. Se realizaron análisis univariados para comparar la abundancia de las dos especies durante los muestreos y entre los microhábitats. La mayor abundancia de ambas especies y el mayor número de ninfas preemergentes se presentaron durante la época seca y el inicio de las lluvias. No se encontraron diferencias significativas en la abundancia de especies entre muestreos o microhábitats ($p > 0,05$); sin embargo, *A. marta* presentó la mayor abundancia. En cuanto a los microhábitats, la mayor abundancia se registró en hojarasca con 638 individuos, mientras que se observaron valores similares en lento y rápido con 194 y 294 individuos respectivamente. Este estudio evidenció que la hojarasca

representa uno de los microhábitats más importantes para las dos especies de plecópteros en el río Gaira. Adicionalmente, la similitud en las abundancias en este microhábitat sugiere la disponibilidad recursos (alimento y refugio) y una superposición de nichos entre ellas.

Palabras clave: dinámica poblacional, microhábitats, plecópteros, régimen de lluvias.

INTRODUCTION

Plecoptera is among the most abundant groups of aquatic insects dwelling in small tropical rivers. They are considered good indicators of the ecological health of bodies of water due to their sensitivity to changes in physicochemical variables (Zúñiga and Cardona, 2009). Many plecopteran species, such as those of the genus *Anacraneuria* Klapálek, 1909 (Perlidae), are predators that regulate the growth and abundance of other benthic invertebrates (Allan, 1983). Therefore, they are closely involved in the energy flow and cycling of nutrients that are then transported and linked to the terrestrial systems and other trophic levels of the aquatic ecosystem (Chará-Serna et al., 2010; Gutiérrez-Fonseca, 2010). Plecopterans preferentially inhabit rivers with cold, rapid and highly oxygenated currents, but some species can be found in lentic, oligotrophic, seasonal systems or in deep lakes (Yule, 1985; Gutiérrez-Fonseca, 2010).

Plecoptera is an ancient order of aquatic insects encompassing 3500 species distributed among 16 families with 286 genera covering the whole world except Antarctica (Fochetti and Tierno de Figueroa, 2008; DeWalt et al., 2017). There are six Plecoptera families in South America including the particularly speciose Perlidae (Stark et al., 2009; Froehlich, 2010; DeWalt et al., 2017). In Colombia, stoneflies are represented by two families (Zúñiga, 2010) with Gripopterygidae restricted to the high mountain systems of two localities in the departments of Tolima and Nariño (Barreto-Vargas et al., 2005, Zúñiga and Cardona., 2009), while Perlidae is widespread across different natural regions of the country with a large predominance of *Anacroneuria,* which is a good biological model for populations studies and is a group indicator of water quality by this high sensibility to pollution and changes in the physicochemical variables like dissolved oxygen, pH, flow and temperature (Zúñiga, 2010). The latter is the most diverse and abundant genus with 64 described species (Stark and Zúñiga, 2014). In the Sierra Nevada de Santa Marta (SNSM), north-western Colombia, *Anacroneuria* remain the only the stonefly genus recorded and it is represented by five species: *A. caraca* Stark, 1995; *A. paleta* Stark, 1995; *A. marta* Zúñiga and Stark, 2002; *A. choco* Stark and Bersosa, 2006 and *A. tayrona* Zúñiga and Tamaris, 2007 (Stark, 1995; Zúñiga and Stark, 2002; Zúñiga et al., 2006; Stark and Zúñiga, 2014). *Anacroneuria marta* and *A. caraca* are species with the highest abundance in the middle reaches of the Gaira River, a strategic watershed of the SNSM.

Stoneflies are often found in substrates with gravel, rocks, and leaf litter. However, such observations are typically based on occasional or low-intensity sampling (Tamaris-Turizo et al., 2007; Guzmán-Soto and Tamaris-Turizo, 2014; Barragán et al., 2016). Moreover, the abundance of nymphs for the two investigated species (*A. marta* and *A. caraca*) appears to vary with period (Sierra-Labastidas et al., 2017); therefore, longer-term studies are required to determine their population dynamics and responses to environmental parameters (Ridl et al., 2018). Studies that use biological traits of the organisms have been used with more frequency during the last decade and could be a better tool to describe environmental dynamics and responses of populations (Verberk et al., 2013).

Nymphs of the genus *Anacroneuria* are morphologically very similar among the different species and only ~10 % of the species recorded in South America and the Neotropics are taxonomically well-known (Stark et al. 2009; Froehlich, 2010). Consequently, information on the ecology of these species is very limited. The purpose of this study was to describe the spatial (between microhabitats) and temporal (during an one-year period) distribution of *A. marta* and *A. caraca* in the middle basin of the Gaira River, Sierra Nevada de Santa Marta, Colombia. For both species, we expect that higher nymph abundance would be recorded during the dry period and at leaf litter substrate since the dry period is characterized by the most stable flow and leaf litter offers more availability of food items than gravel (Tamaris-Turizo et al., 2007; Guzmán-Soto and Tamaris-Turizo, 2014; Barragán et al., 2016; Sierra-Labastidas et al., 2017; Ridl et al., 2018; Tamaris-Turizo et al., 2020b). However, our hypothesis could be affected by the sampling done during the El Niño phenomena, a scenario that affects the abundance of aquatic insects (particularly the EPT) (Godoy et al., 2022). Finally, we do not expect to find differences between species, regardless of the period and microhabitat due are congeneric species with similar spatial distribution and use of resources.

MATERIALS AND METHODS

Study site

The Gaira River basin is located to the northwest of the Sierra Nevada de Santa Marta (an isolated mountain range in northern Colombia). It has a total surface area of 10,464.3 ha and its main channel is 32.53 km long (Frayter et al., 2000). The river is characterized by rapid to slow flow, exhibits fluctuations in its flow, ranging between 0.41 and 0.88 $m3/s^{-1}$, with an average depth of 0.35 m and presents great geomorphological diversity conducive to a variety of microhabitats (such as rocky and sandy, among others)

which enable the establishment of different biological communities (Tamaris-Turizo, et al., 2013; Hurtado-Borrero and Tamaris-Turizo, 2023). The water's average pH is 7.48, with a dissolved oxygen content of 7.68 mg/L-1, and temperature averages 18.9 °C (Oliveros-Villanueva et al., 2020). Sampling was conducted at the site known as Hacienda La Victoria, located between 11°07'44.2" N and 74°05'35.8" W at an altitude of 900 m. in the Gaira River middle basin. The riverbank area (70 to 100 m) is well preserved despite large extensions of adjacent coffee plantations (Tamaris-Turizo, et al., 2020a). The study section encompassed a variety of microhabitats dominated by large rocks (60 %, more than 20 cm diameter), gravel (30 %, between 10 and 20 cm diameter), and leaf litter (ten percent) in agree the characterization proposed by Rosgen (1994) (Fig. 1).

Collection and identification of organisms

Sampling campaigns were conducted every 20 days (14 campaigns in total) between July 2014 and June 2015, a timeframe that included "El Niño" a climate event in Colombia and across the tropical Pacific. A stretch of 100 m, representing the study site microhabitat variability, was selected. Nymphs were collected from the following microhabitats: leaf litter, pool, and riffle (Hawkins et al. 1993; Hauer and Lamberti, 2011). Slow sections were characterized by flow speeds < 2 m/s while rapids had water velocities > 3 m/s. For the leaf litter sample, a hand net was used to collect approximately 500 g of wet weight. For gravel located in slow and rapid currents, a Surber net (area: 0.09 m^2; mesh size: 250 µm) was used at countercurrent for three minutes (Wantzen and Rueda-Delgado, 2009; Zúñiga and Cardona, 2009). Six replicates were collected in each microhabitat. The stonefly nymphs were separated at the laboratory and preserved in 96 % ethanol. During the sampling carried out on December 6th, 2014, there was abundant rainfall with flood risk and only three replicas were collected in each microhabitat.

Mature nymphs were identified as species based on the taxonomic characteristics cited by Stark (1995), Zúñiga et al., (2007), and Hurtado-Borrero et al., (2020). These identifications were later confirmed by extracting male genitalia from mature and pre-emergent nymphs and comparing them to corresponding taxonomic descriptions (Stark, 1995; Zúñiga and Stark, 2002). Approximately ten percent of the nymphs could not be used in the study because they were very young, and their morphological features were not yet well defined.

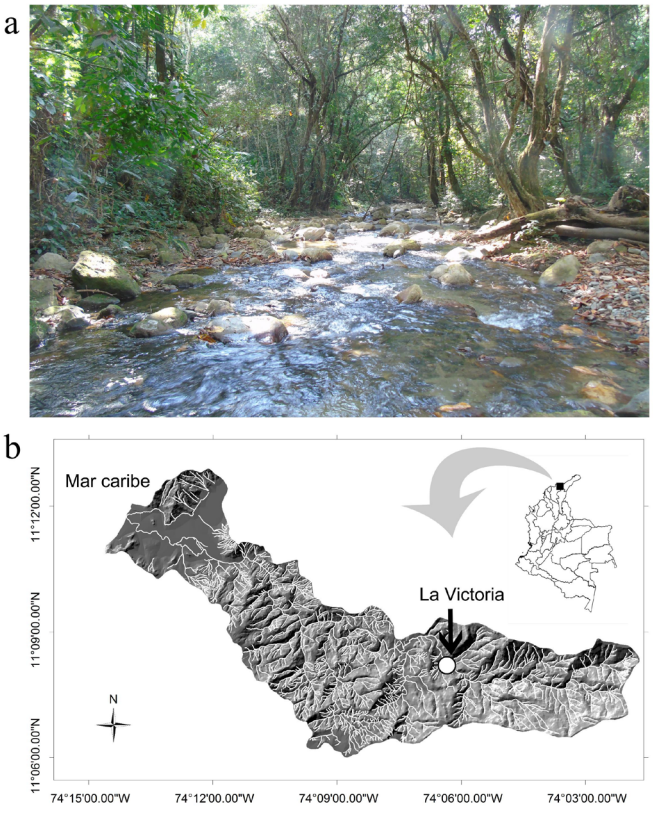

Figure 1. Gaira River basin (Sierra Nevada de Santa Marta - Colombia). a) show details of the selected reach. b) location of the study site in the basin.

Data analysis

Species abundance among the campaign 14 events, was compared using the Kruskal-Wallis test (H') followed by a Tukey Test (*post-hoc*). Since the residuals did not meet the assumptions of normality and homoscedasticity, samples included organisms collected from all substrates (pool, riffle, and leaf litter) to compare the abundances of both species between seasons. The sampling design was unbalanced (an unequal number of replicates between treatments) because only three replicates were taken during the December sampling. Nevertheless, this analysis allowed for comparisons among all the treatments (sampling campaigns) and we, therefore, considered a sampling unit as the sum of samples taken in the substrates during a monthly sampling campaign. To compare the abundance of nymphs for each *Anacroneuria* species between climatic periods and between pool and riffle microhabitats, the Mann-Whitney U test was performed. All analyses were performed using the StatR package in RWizard v. Beta 2.3 (Guisande et al., 2014).

RESULTS

A total of 1126 individuals were collected and identified, 294 were found in riffle, 194 in pool, and 638 in leaf litter (Table 1). Both species showed significant changes in abundances between samplings (*A. marta*: H = 57.52; df = 15; $p < 0.05$. *A. caraca*: H = 53.27; df = 15; $p < 0.05$). The highest abundances for both species were observed during the dry period and the beginning of the rainy period (December 2014 to June 2015) while the lowest abundances were observed during the months of highest rainfall (August

to November 2014 and July 2015). On the other hand, the total abundance of both species in each sampling event did not show significant differences ($p > 0.05$) (subsamples taken at microhabitats were pooled together) even when *A. marta* presented a greater abundance than *A. caraca* (628 and 498 individuals, respectively) (Fig. 2).

Table 1. Total abundance (number of individuals) of the two species of *Anacroneuria* in each microhabitat of the Gaira River, north-west Colombia during the sampling campaigns (July 2014 – June 2015).

Microhabitat	*A. marta*	*A. caraca*	Total
Riffle	155	139	294
Pool	90	104	194
Leaf litter	383	255	638

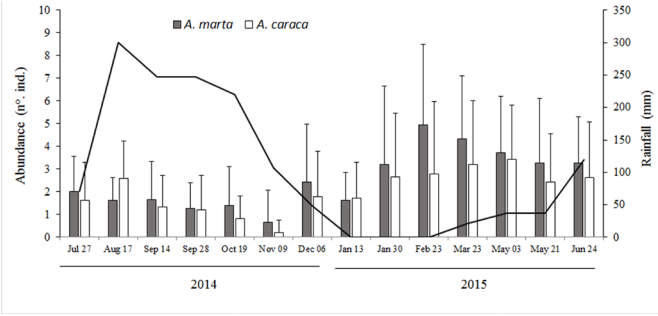

Figure 2. Mean abundance of nymphs of *A. marta* and *A. caraca* during the study and this relation with the annual precipitation (solid black line). Error bars indicate the standard deviation of the mean.

The abundance of *Anacroneuria* (both species together) in pool and riffle during all sampling events did not show significant differences ($p > 0.05$). However, when comparing these microhabitats at each of the samplings, differences were observed in the samples taken on July 27 ($W = 102$; $p < 0.05$), December 6 ($W = 109$; $p < 0.05$), and January 13 ($W = 116$; $p < 0.05$). In all cases, the highest abundances among the riffle microhabitat occurred in the rapid current (Fig. 3a). The pool microhabitat recorded the lowest abundance of both species during the whole sampling period (Fig. 3b), although no significant differences were found ($p > 0.05$). The leaf litter microhabitat showed little seasonal variation in the abundance (Fig. 3c), despite registering the highest number of nymphs collected for both species; a tendency that was observed throughout the year.

DISCUSSION

The results found corroborate our hypothesis, due the highest abundance of both *Anacroneuria* species were recorded during the dry period; however, we observed similar abundances of both species during dry and rainy seasons, similar to the registered by Zúñiga et al., (2003) and Zúñiga (2010) who analyzed the emergence patterns and flight periods of four *Anacroneuria* species, including *A. caraca*, from a stream in southwestern Colombia. On another hand, we did not observe seasonal emergence, although the peaks for the greatest numbers of mature nymphs, and emerging adults were reported during the end of the dry period and the beginning of the rainy period, this indicates that populations are begin renewed during all the year, and the support the nearby populations. Similar results were observed in the Rio Frío stream (Valle del Cauca, Colombia), in which a higher abundance of some species of *Anacroneuria* (including *A. caraca*) during their emergence period was found (Zúñiga, 2010).

The similarity in the abundance of the riffle and pool microhabitats indicates that flow velocity in the Gaira River during the study period did not affect the distribution of the species evaluated in these microhabitats (Tamaris-Turizo et al., 2007). In addition, the highest abundances in all microhabitats were recorded during the months corresponding to the dry period and the beginning of the rainy period which are associated with hydraulic stability and reduced flow which was observed by Sierra-Labastidas et al., (2017) in

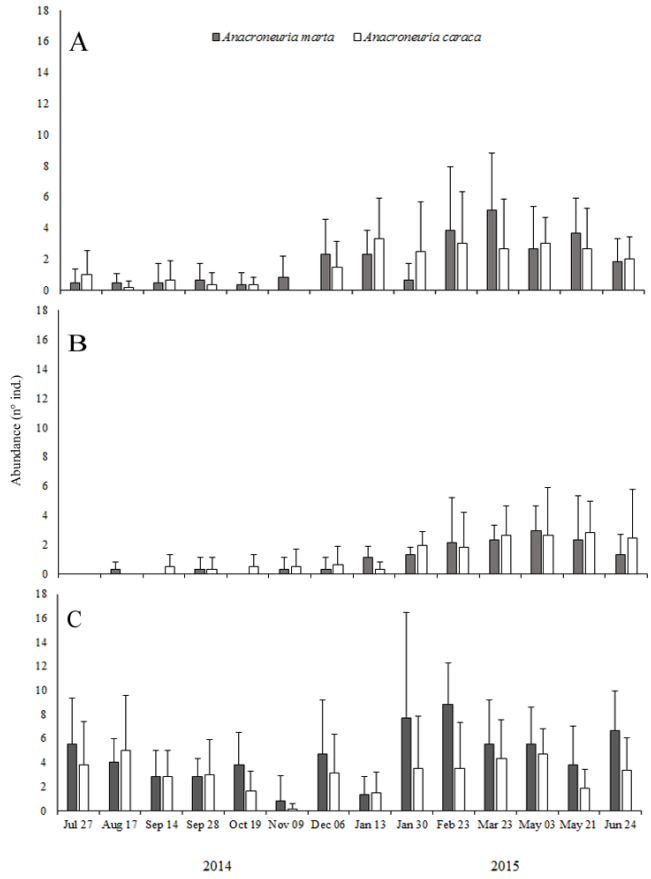

Figure 3. Abundance (mean ± standard deviation) per microhabitat of *A. marta* and *A. caraca* in the Gaira River middle basin, Sierra Nevada de Santa Marta. A) riffle, B) pool; and C) leaf litter.

the same river. Overall, the riffle, pool, and leaf litter microhabitats had high abundances with leaf litter showing consistently the highest values, thereby confirming the niche habitat overlap of these plecopteran species previously documented in the trophic overlap of resources consumed – mainly aquatic insects: dipterans and mayflies (Hurtado-Borrero et al., 2019). Similar abundances of both species at the studied microhabitats could be related to the constant and sufficient supply of food resources previously documented by Rodríguez-Barrios et al., (2011) along the Gaira River basin. The food supply is related to the quantities of available coarse particulate organic matter (CPOM) and fine particulate organic matter (FPOM), mainly coming from different fractions of riparian vegetation (Collantes et al., 2014), and in the Plecoptera order the availability of prey (Rodríguez-Barrios et al., 2011). This could mean that essential food resource contributions may be necessary for the coexistence of the two species because the organic matter input -leaves, fine debris from the riparian plants, offer substrates, refuges such as leaf litter, and food that can be consumed directly or indirectly (i.e., through the consumption of detritivorous insects) (Hurtado-Borrero et al., 2019; Tamaris-Turizo et al., 2020b).

In Neotropical lotic systems, a decrease in the density of aquatic insects has been documented during periods of high rainfall (Giacometti and Bersosa, 2006; Cardona et al., 2008; Bohórquez et al., 2011; Roldán et al., 2014; Theodoropoulos et al., 2017; Castillo et al., 2018). The similar decrease recorded in this study could be consistent with the results obtained in other studies conducted in the SNSM (Barragán et al., 2016) and in the Gaira River (Tamaris-Turizo et al., 2007; Sierra-Labastidas et al., 2017; Tamaris-Turizo et al., 2020b). Moreover, the Gaira River is a steep-gradient river, where the formation of substantial pool zones (associated with the slow current) is limited; this condition could be associated with the similarity found in the abundance of both *Anacroneuria* species in the gravel substrate. Because this study coincided with the "El Niño" climatic phenomenon (Instituto de Hidrología, Meteorología y Estudios Ambientales [IDEAM], 2016), there was very little variation in river flow, and this lack of variance led to high hydrologic stability of the flow (Table 2.) which represents a suitable environment for the establishment of stonefly populations (Ruhí et al., 2016). Also, less frequent disturbance can be better for populations with high growth rates, which raises the competition between species as we evidence here (Huston, 1979; Magurran and Huston, 1995; Godoy et al., 2022). Although not found differences between both species, new studies that will include traits functional (morphologic, physiologic, and behaviors) are necessary to improve the understanding of population ecology of aquatic insects (Verberk et al., 2013).

Table 2. Monthly rainfall during 2014, 2015, multiannual rainfall (MP) mean and the standard deviation (SD) from 1962 to 2022. Source: Minca Weather Station data.

MONTH	2014	2015	MP	SD
January	0.0	0.0	48,5	11,5
February	0.0	0.0	15	7,3
March	0.0	21.2	133,6	40,4
April	7.3	24.2	491,1	173,6
May	117.3	36.7	285,1	304,3
June	90.3	119.1	140,1	248,8
July	71.3	68.9	396,2	309,4
August	300.0	124.3	128	316
September	247.2	255.4	563,4	494,5
October	220.1	91.9	581	510,7
November	106.9	131.9	225,6	262,2
December	47.9	8.0	80,9	72
Total annual	1208.3	874.4	2596,8	2713,3

CONCLUSIONS

In the middle basin of the Gaira River, located in the Sierra Nevada de Santa Marta, *Anacroneuria caraca* and *A. marta* nymphs were abundant in the gravel and leaf litter microhabitats during most of the studied period, showing a particular preference for the last one and during the dry period. This could be due to the greater microhabitat heterogeneity and food availability provided by this microhabitat.

This study showed that the leaf litter microhabitat is one of the most important Gaira River niches since it offers protection to both studied species against the mechanical friction of the current that exhibits high slope and flow velocity. Moreover, leaf litter provides adequate food resources for the other organisms that are part of the macroinvertebrate community at the studied sites, including the potential prey for stonefly nymphs evaluated in this study. Additionally, the similar abundance of the two species in the litter may suggest the availability of sufficient resources and the superposition of niches between them (Hurtado-Borrero et al. 2019). We did not observe an emergence period, although the peaks for the greatest numbers of mature nymphs, and emerging adults of both *Anacroneuria* species were reported

during the end of the dry period and beginning of the rainy period, suggesting a considerable influence of the rainfall regime on the area's stonefly populations. Autecological studies are essential to increase understanding of the spatial and temporal dynamics of different populations of macroinvertebrates species that can be useful as bio-indicators of water quality and in defining activities related to river system conservation and management, being the selected plecopteran species good model organisms.

AUTHOR'S PARTICIPATION

Yesely Hurtado-Borrero and Cesar E. Tamaris-Turizo participated in the conceptualization of the work, the collection and processing of biological material, data analysis, and the document's writing. Maria del Carmen Zúñiga contributed to the review and writing of the document.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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