Univ. Sci. 29(3): 187–205, 2024
doi: 10.11144/Javeriana.SC292.sdob
ORIGINAL ARTICLE

Spatial distribution of black fly (Diptera: Simuliidae) immature taxocenoses from the Pedra Branca State Park, Brazil

Tayanna Rodrigues da Costa[1], Tatiana Nascimento Docile[2,3], Christina de Albuquerque Ferreira[1,4], Tainá Maria Miranda Souza-Martins[1,5], Ivyn Karla Lima-de-Sousa[1,5], Leonardo Henrique Gil-Azevedo[5], Ronaldo Figueiró*[1]

Edited by
Angela Johana Espejo Mojica
editorus@javeriana.edu.co

1. Laboratório de Meio Ambiente e Saúde(LabMAS), Departamento de Biologia (DEPBIO), Faculdade de Ciências Biológicas e Saúde (FCBS), Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, Brazil.
2. Departamento de Ciências da Natureza, Instituto de Aplicação Fernando Rodrigues da Silveira (Cap-UERJ), Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, Brazil.
3. Laboratório de Educação Profissional em Vigilância em Saúde (LAVSA), Escola Politécnica de Saúde Joaquim Venâncio, Fundação Oswaldo Cruz (FIOCRUZ), Rio de Janeiro, Brazil.
4. Centro de Biologia Experimental Oceanus, Instituto Oceanus, Rio de Janeiro, Brazil.
5. Departamento de Entomologia, Museu Nacional, Universidade Federal do Rio de Janeiro (UFRJ), Brazil.

*ronaldofigueiro@gmail.com

Received: 27-10-2023
Accepted: 1-08-2024
Published online: 18-09-2024

Citation: Rodriguez da Costa T, Nascimiento Docile T, de Albuquerque Ferreira C, Miranda Souza-Martins TM, Lima-de-Sousa IK, Gil-Azevedo, Figueiró Ronaldo. Spatial distribution of black fly (Diptera: Simuliidae) immature taxocenoses from the Pedra Branca State Park, Brazil, *Universitas Scientiarum*, 29(3): 187 - 205, 2024
doi: 10.11144/Javeriana.SC292.sdob

Funding: Fundação de Amparo a Pesquisa do Estado do Rio de Janeiro

Electronic supplementary material: n.a.

Abstract

To understand the relationship between environmental factors and black fly (Simuliidae) species' distributions, we sampled immature individuals in Pedra Branca State Park, Rio de Janeiro, Brazil, during the dry season in June 2018. We then correlated environmental variable values with black fly larvae abundances via redundancy analyses. The abundances of *Simulium pertinax* Kollar 1832, *Simulium subpallidum* Lutz 1910, *Simulium (Thyrsopelma)* spp., *Simulium (Psaroniocompsa)* spp., and *Simulium (Inaequalium)* spp. were negatively correlated with light and temperature, while Simulium (Hemicne tha) spp.'s abundance correlated positively with these variables. The abundances of Simulium pertinax Kollar 1832, Simulium (Hemicnetha) spp., and Simulium (Thyrsopelma) spp. were positively associated with altitude, while the abundances of the other species revealed negative correlations with this factor. Species richness was determined using pupae in a linear regression with the Ephemeroptera, Plecoptera Trichoptera (EPT) index. This analysis showed that the most deteriorated sites have a higher species richness than more conserved sites within Pedra Branca State Park. Our results suggest that simuliid species richness reflects habitat integrity, thus becoming a reliable indicator of habitat conservation status.

Keywords: Bioindicators; Community ecology; Macroinvertebrates; Simuliids

1. Introduction

In the past few decades, lotic and lentic systems have been strongly affected by several anthropogenic factors [1], including increased pollution from sediments and chemical and solid waste, as well as disturbances due to plumbing and soil impermeabilization. These factors have altered ecological community structures and ecosystem processes [2, 3]. Consequently, water quality has markedly decreased, leading to aquatic biodiversity impoverishment and changes in the dynamics of biological communities [2, 4].

Different organisms serve as bioindicators to assess environmental impacts on aquatic ecosystems, for instance black flies (Diptera: Simuliidae). Distributed worldwide, Simuliids include 2 415 known species, most of which are hematophagous and act as vectors of diseases such as onchocerciasis, mansonellosis, and pemphigus foliaceus [5, 6]. The geographic range of simulid species is limited to sites with appropriate lotic conditions for breeding and development of black fly immature forms [7]. Black fly larvae are filter-feeders that are strongly dependent on the conditions of the water flow due to their morphological adaptations to fixation and feeding [8, 9, 10]. Therefore, their local microdistribution is limited to sites within streams and rivers with suitable water current velocity and substrate type [11, 12, 13, 14, 15].

These insects are considered key organisms in the boreal biome [17] due to their role as a trophic link in lotic food webs, transforming tiny particles of organic matter into larger fecal pellets that are rich in organic matter due to blackflies' low digestive efficiency, thus making resources available to a wide spectrum of other filter-feeding organisms [18, 19], thus impacting lotic system biodiversity. Another feature of black flies that makes them key organisms in lotic systems is the secretion of silk strands from their salivary glands that form "silk pads" [20], which facilitate the recovery of macroinvertebrate communities [21].

Simuliid species ecology and their potential as bioindicators remain poorly studied in the Neotropical Region [5]. Although these organisms are seen as bioindicators [22, 23], few studies have actually approached this aspect of their biology (e.g., [23, 24, 25, 26, 27]), focusing instead on their vectorial activity [28], behavior [29, 30, 31], natural infections [32, 33, 34] or ecological patterns [35, 36, 37, 38, 39, 40, 41, 42]. The fauna of Pedra Branca State Park (in the state of Rio de Janeiro - Brazil) is abundant, harboring abundant black flies. Currently most of the faunal studies in this park focus on its vertebrates (e.g., [43]; [44]), and only a handful focus on its invertebrates (e.g., [45]) with only one addressing its simuliid fauna [37].

The present study investigated the relationships between environmental factors and black fly larvae distribution and between habitat quality and black fly species richness in Pedra Branca State Park. To this end, we tested the following hypotheses: (i) Black fly species have different microhabitat requirements and are thus associated with other abiotic factors. (ii) Habitat integrity determines black fly larvae species richness, and (iii) land use influences black fly larvae community structure.

2. Materials and methods

2.1. Study area

The Pedra Branca State Park is one of the world's largest urban forests. It was established in 1974 in the Rio de Janeiro municipality. It is an important protected area that comprises eight hydrological basins and 53 microbasins that divide the area into three different hillsides: North (Guanabara Bay), East (Coastal lagoons), and West (Sepetiba Bay) [46].

2.2. Sampling sites

Larvae sampling was conducted in the Divisa/Moinho River (sites 1–4) and Grande River (sites 5–7) during the dry season in June 2018 (**Fig. 1**). The Divisa/Moinho River is located at Vargem Grande, where it is protected and managed by the regional Institute of Environmental Studies (INEA) and the Quilombola Community Cafundá Astrogilda. In this area, the Quilombola population practices sustainable agriculture involving environmental conservation activities and ecotourism. The Grande River is located at Pau da Fome, in the neighborhood of Taquara, where it feeds a water catchment system managed by INEA and the State Company for Water and Waste Management (CEDAE). Access to this area is less restricted than in the Divisa/Moinho River and is also subject to environmental conservation activities and ecotourism.

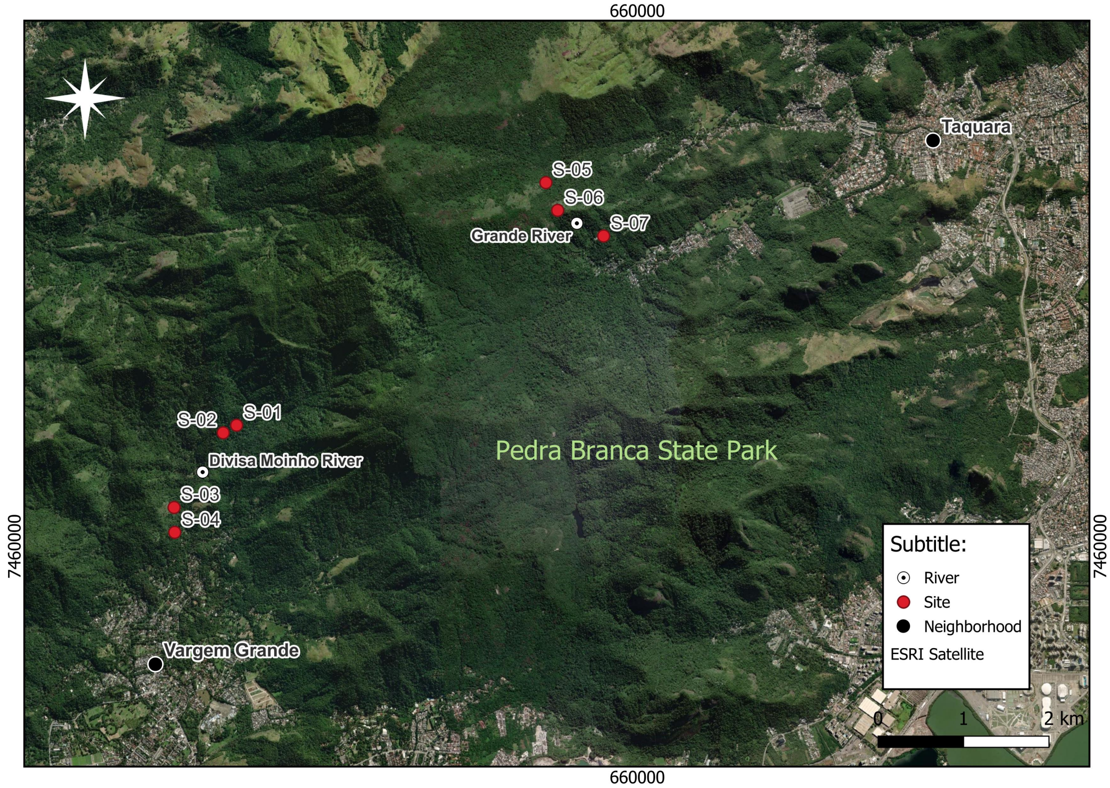

Figure 1. Collection sites within Pedra Branca State Park, Rio de Janeiro, Brazil. Map created in QGIS 3.34.5 using the ESRI cartographic base.

2.3. Larvae sampling and environmental factor assessment

Immature black flies and their associated macroinvertebrate fauna were hand-sampled and immediately stored in 70% ethanol, following previously described methods [47]. Final instar black fly larvae were stored for later morphological study, while pupae were kept for later identification. At each sampling site, which consisted of 15-m stream/river sections, ten randomly placed quadrats (30 cm x 30 cm) were sampled. Luminosity was determined using a luximeter (EQUITHERM LUX 204), temperature was recorded using a digital thermometer, and pH was measured using a portable pH meter (HANNA INSTRUMENTS HI 98130). Altitude was determined using a GPS GARMIN eTREX 30, and water depth was measured with a ruler.

2.4. Larval identification

Macroinvertebrate specimens were identified and quantified using pertinent literature [48, 49, 50, 51, 52, 53, 54, 55, 56]. Black fly larvae were first sorted into different morphotypes in the laboratory and then identified to species level, whenever possible, or to subgenus level where species-level identification was not possible using the available taxonomic guides [57, 58]. Pupae were identified to species level following a previous report [59]. Larvae identified only to subgenus level were treated as taxonomic entities.

2.5. Statistical analyses

To test the first hypothesis, namely the relationships between later instar black fly larvae and the assessed environmental factors, we conducted redundancy and detrended correspondence analyses, both of which were performed in CANOCO 4.5. Significant variables were determined after 5 000 Monte Carlo permutations. To test the second hypothesis, we evaluated habitat integrity with the EPT index [60], which was calculated using macroinvertebrate data (from at least 200 macroinvertebrate specimens per site) (**Table 1**). Black fly species richness was determined based on the identified pupae (**Table 2**) and used as the dependent variable in a linear regression with EPT index values, as the independent variable. This analysis was conducted in Prism 5. We identified and excluded outliers based on Cook's distances. To test the third hypothesis, we submitted later instar black fly larvae abundances (**Table 3**) to non-metric multidimensional scaling (NMDS), using the Bray–Curtis index to compare the different areas of the park, and subsequently subjected the with Bray-Curtis index data to a one-way PERMANOVA with 999 permutations. A cluster analysis was used with the same aim. These latter analyses were performed in PAST 4.1.

Table 1. Abiotic factor values and EPT index scores from Vargem Grande (1-4) and Pau da Fome (5-7) in Pedra Branca State Park, Brazil.

Area	Site	Altitude (m)	pH	Temperature (°C)	Luminosity (Lux)	Depth (cm)	EPT index
Vargem Grande	1	131	7.4	22	1011.8	21	6
Vargem Grande	2	178	6.3	24	3261.9	44	5
Vargem Grande	3	154	5.8	23	2543	27	7
Vargem Grande	4	149	5.6	25	1501.1	11	6
Pau da Fome	5	115	5.3	19.2	1738.3	10	3
Pau da Fome	6	148	5.8	19.8	498.9	10	6
Pau da Fome	7	151	5.7	18.2	3433.6	30	3

Table 2. Black fly species presence (1) and absence (0) determined using identified pupae from Vargem Grande (1-4) and Pau da Fome (5-7) in Pedra Branca State Park, Brazil.

Species/ Sites	1	2	3	4	5	6	7
Simulium pertinax Kollar 1832	1	1	1	1	1	1	1
Simulium anamariae Vulcano 1962	1	1	0	1	1	1	0
Simulium limbatum Knab 1915	1	1	0	1	1	1	0
Simulium inaequale Peterson & Shannon 1927	1	1	0	1	1	1	1
Simulium subnigrum Lutz 1910	1	1	0	1	1	1	1
S. rubrithorax Lutz 1909	0	1	0	0	1	1	1
Simulium botulibranchium Lutz 1910	0	0	0	0	1	1	1
Simulium brachycladum Lutz & Pinto 1931	0	0	0	0	1	1	1
Simulium scutistriatum Lutz 1909	0	0	0	0	1	1	1
Simulium subpallidum Lutz 1910	0	0	0	0	0	1	1
Species Richness	5	6	1	5	9	10	8

Table 3. Black fly larvae species and taxonomic entity frequencies in the sampling sites from Vargem Grande (1-4) and Pau da Fome (5-7) in Pedra Branca State Park, Brazil.

Species/ Sites	**1**	**2**	**3**	**4**	**5**	**6**	**7**
Simulium pertinax Kollar 1832	5	3	1	6	1	20	27
Simulium sp.	9	2	1	1	4	0	0
Simulium (Inaequalium) sp.	4	4	0	2	14	39	2
Simulium (Trichodagmia) sp.	0	1	0	0	3	152	19
Simulium (Psaroniocompsa) sp.	0	0	0	0	1	26	0
Simulium subpallidum Lutz 1910	0	0	0	0	0	13	1

3. Results and Discussion

The redundancy analysis on identified black fly larvae revealed that the abundances of *Simulium pertinax* Kollar 1832, *Simulium subpallidum*, *S.(Thyrsopelma)* spp., *Simulium (Psaroniocompsa)* spp., and *Simulium (Inaequalium)* spp. were negatively correlated with luminosity (p = 0.0018) and temperature (p = 0.0016); in contrast, *Simulium (Hemicnetha)* spp. correlated positively with these two variables. The abundance of *Simulium pertinax* Kollar 1832, *Simulium (Hemicnetha)* spp., and *Simulium (Thyrsopelma)* spp. correlated positively with altitude (p = 0.0012), and the opposite was observed for *Simulium subpallidum* Lutz 1910, *Simulium (Psaroniocompsa)* spp., and *Simulium (Inaequalium)* spp. (**Fig. 2**).

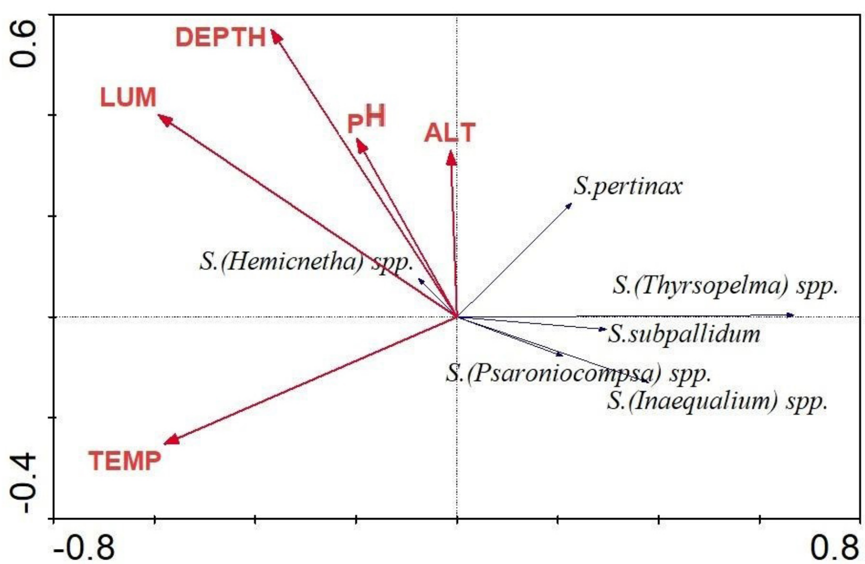

Figure 2. Redundancy Analysis (RDA) results indicating that the presence of *Simulium pertinax* Kollar 1832, *Simulium subpallidum* Lutz 1910, *Simulium (Thyrsopelma*) spp., *Simulium (Psaroniocompsa*) spp. and *Simulium (Inaequalium*) spp. larvae were negatively correlated with luminosity (p=0.0018) and temperature (p=0.0016), while *Simulium (Hemicnetha*) spp. correlated positively with those variables; *Simulium pertinax* Kollar 1832, *Simulium (Hemicnetha*) spp. and *Simulium (Thyrsopelma*) spp. correlated positively with altitude (p=0.0012), while *Simulium subpallidum* Lutz 1910, *Simulium (Psaroniocompsa*) spp. and *Simulium (Inaequalium*) spp correlated negatively with this factor.

Redundancy analysis outcomes corroborated our first hypothesis that different species have different associations with abiotic factors, highlighting that luminosity, altitude, and temperature were the only variables significantly determining the distribution of collected simuliid species. Interestingly, most black fly species were abundant in dim places, as aligned with previous studies [30, 47, 62]. Our redundancy analysis also revealed altitude-dependent species composition variation. Temperature, water current speed, pH, and luminosity can influence species composition, and several previous studies have approached different abiotic variable sets and consistently found that high-altitude regions harbor different species compared to the lower-altitude areas [47, 63].

The association between luminosity and black fly larvae from different species has often been reported as one of the most important factors influencing their spatial distribution. Some authors have noted that a small canopy likely benefits black flies, contrary to the most common findings in the literature [61], because it buffers temperature changes, as more exposed sites are prone to daily temperature fluctuations [47]. However, other authors have found that denser canopy covers, and hence lower luminosity, are better for black fly species since a denser canopy cover is also correlated with higher levels of riffle litter, a substrate often harboring black fly larvae.

Simuliid species are widely tolerant to a wide range of environmental factors, with species occurring in water bodies of different temperatures (0 °C to 35 °C) and within variable altitude environments, from sea level to altitudes above 4,000 meters [64]. Thus, black flies respond to given factors, which determine their dominance in a particular location [65]. The study of aquatic environment communication suggests that an optimal thermal regime exists for each species and directly interferes with adult size and fertility [66]. Temperature is one of the main factors influencing simuliid microdistribution [67].

The linear regression of EPT scores versus black fly species richness showed a significant negative correlation ($r^2 = 0.97$, $p < 0.001$, $r = -0.98$), indicating that more deteriorated sites contain higher black fly species richness than more preserved sites (**Fig. 3**).

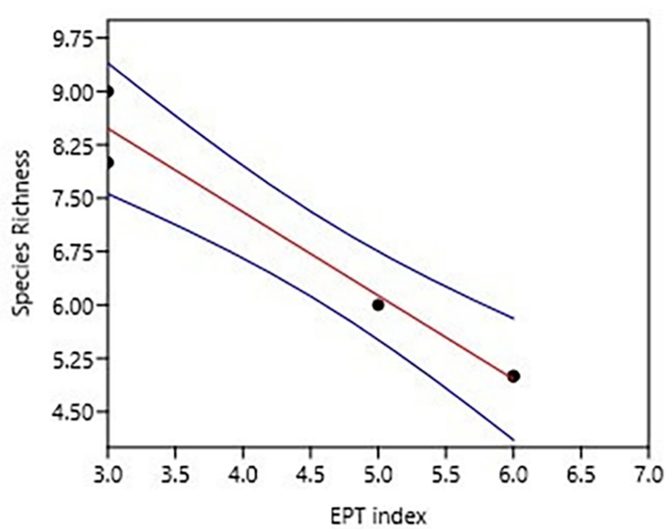

Figure 3. Linear regression on EPT index scores versus black fly species richness suggesting a pattern towards higher species richness values at lower habitat quality sites ($r^2 = 0,97$, $p < 0,001$, $r = -0.98$).

The obtained linear regressions corroborated our second hypothesis. The patterns observed in this study agreed with those in the literature on black fly community composition to evaluate the conservation status of lotic systems, indicating that environments with different degrees of habitat integrity harbor different black fly assemblages [23, 68]. Rather than assemblage attributes, population indicators should be used to assess the conservation status in rural areas [16].

Our cluster analysis using black fly presence/absence (**Fig. 4**) corroborated the pattern observed by NMDS, while the cluster analysis using the EPT index scores showed that Site 6 was more similar, regarding habitat quality as revealed by its EPT index, to the sites from the Vargem Grande group, which showed better habitat integrity.

The pattern revealed by our linear regression is a likely consequence of this study being held in a protected area. Previous studies have shown black flies to be absent in extremely deteriorated areas [16, 39]. Therefore, the negative correlation between the conservation status, according to the EPT index and Simuliidae species richness possibly can only be observed within a range limited to low to moderately deteriorated sites. While most of the literature on the relationship between black flies and habitat integrity focuses on the community composition rather than the species richness, most of the previous studies on this topic have shown blackflies to be ubiquitous, being absent only in extremely deteriorated sites [23, 68].

NMDS application to our dataset resulted in two significantly distinct groups, as shown by the one-way PERMANOVA (F = 3.18, p = 0.028). One group included the sites from Vargem Grande and the other consisted of the sites from Pau da Fome (**Fig. 5.**), revealing more dissimilarity among sites from Pau da Fome than among the sites from Vargem Grande (stress = 0.05, R2 axis 1 = 0.94, R2 axis 2 = 0.53).

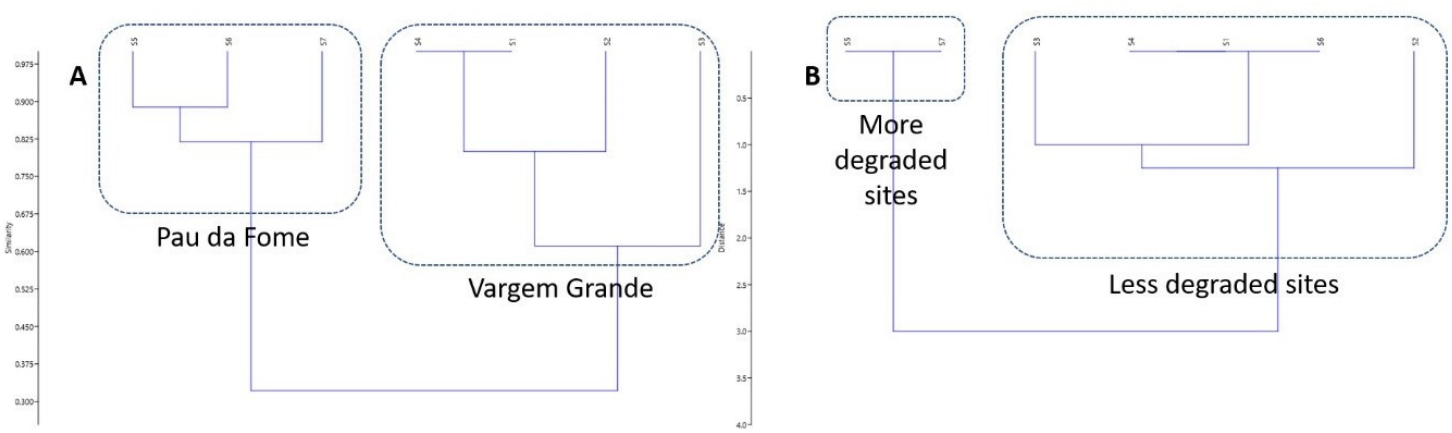

Figure 4. Cluster analyses indicating that the black fly fauna from Vargem Grande (sites 1-4) and Pau da Fome (Sites 5-7) fall into two different groups **(A)**, and that in relation to habitat quality measures by the EPT index site 6 from Pau da Fome is grouped in the better quality group (formed by the sites in Vargem Grande) **(B)**.

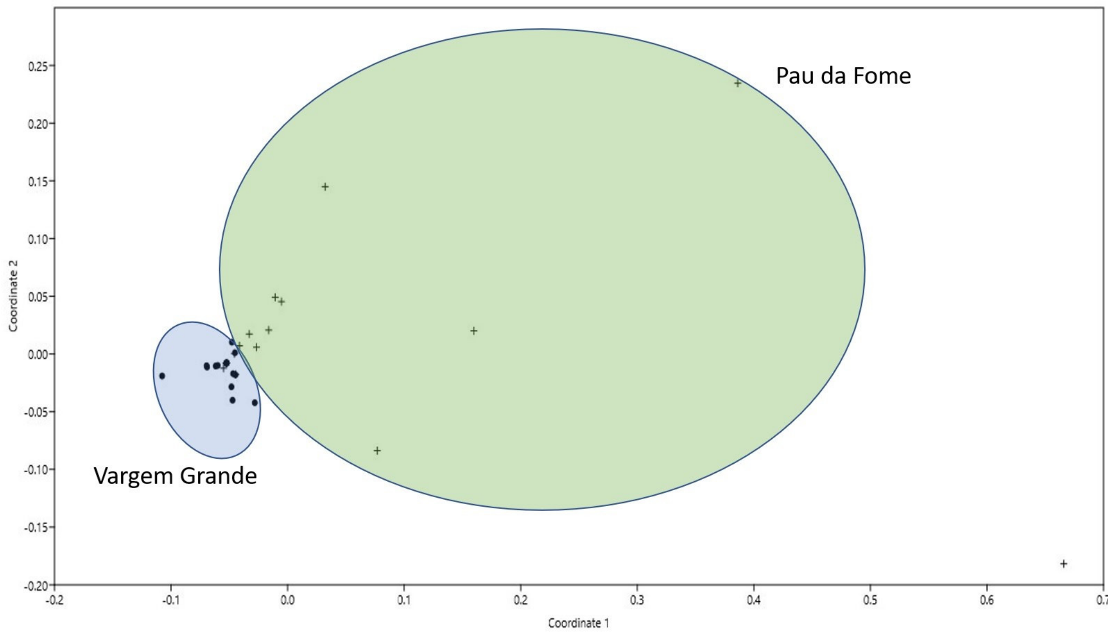

Figure 5. Non-metric multidimensional scaling (NMDS) showing that Vargem Grande and Pau da Fome differ significantly and that black fly species composition differed more in sites within Pau da Fome than within Vargem Grande (Stress = 0,05, R^2 Axis 1 = 0,95, R^2 Axis 2 = 0,53).

The third hypothesis was corroborated via NMDS, resulting in significant differences between the Vargem Grande and Pau da Fome areas within the park. This outcome implies that while both areas are protected, the Pau da Fome (Rio Grande) area, open to visitors, is more disturbed than Vargem Grande (Rio da Divisa/Moinho), which is home to native Quilombola populations and who conduct subsistence practices within their area. The difference between black fly assemblages in both areas is consistent with the relationship between land use and black fly composition reported in previous studies [22, 24], as these areas differ markedly in their land use, even though both are located within the same protected area.

The patterns of associations of simulids with abiotic factors found in this study was similar to that of previously published studies, highlighting the consistency of these relationships in different ecological and geographic contexts [23, 47, 62].

4. Conclusion

The patterns of black fly associations with abiotic factors were consistent with those in previous studies, with some species abundances correlating positively and negatively with abiotic factors, as previously seen in the literature. In contrast to most studies, species richness patterns suggested that they may vary significantly with the degree of habitat integrity. Therefore, the observed species richness patterns represent a potential indicator of conservation status in addition to composition assemblage. Although further studies are needed to effectively test the efficiency and ease of application of black fly species richness as bioindicators, the results of the present study suggest that the number of black fly subgenera/species is an alternative to current lotic systems biomonitoring metrics.

5. Acknowledgements

The authors would like to thank the Pedra Branca State Park staff and Nira Beatriz Mendes Ferreira for assisting the Ecologia de Macroinvertebrados Aquáticos e Insetos Vetores research group team, at Universidade Federal do Rio de Janeiro (UFRJ), during field work. The Entomology Department /Museu Nacional at UFRJ supported the authors particularly during data collection and black fly species identification.

6. Conflict of interest

The authors declare that there are no conflicts of interest.

References

[1] Mcallister DE, Hamilton AL, Harvey B. Global freshwater biodiversity: Striving for the integrity of freshwater ecosystems, *Sea Wind*, 11(3): 1-140, 1997.

[2] Shepp DL, Cummins JD. Restoration in an urban watershed: Anacostia River of Maryland and the district of Columbia. In Watershed restoration: principles and practices J. E. Williams, C. A. Wood, & M. P. Dombeck (eds.) American Fisheries Society, Bethesda, 297-317, 1997.

[3] Docile TN, Rosa DCO, Figueiró R, Nessimian J. Urbanization alters the flow of energy through stream food webs, *Insect Conservation and Diversity,* 9 (5): 416-426, 2016.

https://doi.org/10.1111/icad.12176

[4] Goulart MDC, Callisto M. Bioindicadores de qualidade de água como ferramenta em estudos de impacto ambiental, *Revista da Faculdade de Pará de Minas*, 2(1): 153-164, 2003.

[5] Figueiró R, Gil-Azevedo LH. The role of the Neotropical blackflies (Diptera: Simuliidae) as vectors of the onchocerciasis: a short overview of the ecology behind the disease, *Oecologia Austral*, 14(3): 745-755, 2010.

https://doi.org/10.4257/oeco.2010.1403.10

[6] Adler PH. World blackflies (Diptera: Simuliidae): a comprehensive revision of the taxonomic and geographical inventory. Clemson, Clemson University, 2022.

[7] Currie DC, Adler PH. Global diversity of blackflies (Diptera: Simuliidae) in freshwater, *Hydrobiologia*, 595: 469–475, 2008.

https://doi.org/10.1007/s10750-007-9114-1

[8] Palmer RW, Craig DA. An ecological classification of primary labral fans of filter-feeding black fly (Diptera: Simuliidae) larvae, *Canadian Journal of Zoology*, 78(2): 199-218, 2000.

https://doi.org/10.1139/cjz-78-2-199

[9] Figueiró R, Calvet A, Gil-Azevedo LH, Docile TN, Monteiro RF, Maia-Herzog M. Evidence of phenotypic plasticity of larvae of *Simulium subpallidum* Lutz in different streams from the Brazilian Cerrado, *Revista Brasileira Entomologia*, 59(1): 30-33, 2015.

https://doi.org/10.1016/j.rbe.2015.02.003

[10] Figueiró R, Calvet A, Gil-Azevedo LH, Monteiro RF, Maia–Herzog MM. Potential phenotypic plasticity within *Simulium nigrimanum* Macquart, 1838 (Diptera: Simuliidae) larvae, *Universitas Scientiarum*, 26(2): 217-227, 2021.

https://doi.org/10.11144/Javeriana.SC26-2.pppw

[11] Malmqvist B, Zhang Y, Adler PH. Diversity, distribution and larval habitats of North Swedish blackflies (Diptera: Simuliidae), *Freshwater Biology*, 42(2): 301-314, 1999.

https://doi.org/10.1046/j.1365-2427.1999.444497.x

[12] Hamada N, Mccreadie JW, Adler PH. Species richness and spatial distribution of black flies (Diptera: Simuliidae) in streams of central Amazonia, Brazil, *Freshwater Biology*, 47(1): 31-40, 2002.

https://doi.org/10.1046/j.1365-2427.2002.00778.x

[13] Mccreadie JW, Hamada N, Grillet ME. Spatial-temporal distribution of preimaginal blackflies in Neotropical streams, *Hydrobiologia*, 513: 183-196, 2004.

https://doi.org/10.1023/B:hydr.0000018182.69746.f5

[14] Landeiro VL, Pepinelli M, Hamada N. Species richness and distribution of black flies (Diptera: Simuliidae) in the Chapada Diamantina region, Bahia, Brazil, *Neotropical Entomology*, 38(3): 332-339, 2009.

https://doi.org/10.1590/S1519-566X2009000300006

[15] Figueiró R, Gil-Azevedo LH, Maia-Herzog M, Monteiro RF. Diversity and microdistribution of black fly (Diptera: Simuliidae) assemblages in the tropical savanna streams of the Brazilian cerrado, *Memórias do Instituto Oswaldo Cruz*, 107(3): 362-369, 2012.

https://doi.org/10.1590/S0074-02762012000300011

[16] Couceiro SR, Hamada N, Sagot LB, Pepinelli M. Black-fly assemblage distribution patterns in streams in disturbed areas in southern Brazil, *Acta Tropica*, 140: 26-33, 2014.

https://doi.org/10.1016/j.actatropica.2014.07.018

[17] Malmqvist B, Adler PH, Kuusela K, Merritt RW, Wotton RS. Black flies in the boreal biome, key organisms in both terrestrial and aquatic environments: a review, *Écoscience*, 11(2): 187-200, 2004.

https://doi.org/10.1080/11956860.2004.11682824

[18] Wotton RS, Malmqvist B, Muotka T, Larsson K. Fecal pellets from a dense aggregation of suspension-feeders in a stream: an example of ecosystem engineering, *Limnology and Oceanography*, 43: 719-725, 1998.

[19] Malmqvist B, Wotton R, Zhang Y. Suspension feeders transform massive amounts of seston in large northern rivers, *Oikos*, 92: 35-43, 2001.

[20] Crosskey RW. The natural history of blackflies. John Wiley & Sons, Chichester, 711p. 1990.

[21] Hammock BG, Bogan MT. Black fly larvae facilitate community recovery in a mountain stream, *Freshwater Biology* 59: 2162-2171, 2014.

[22] Lautenschlager M, Kiel E. Assessing morphological degradation in running waters using blackfly communities (Diptera, Simuliidae): Can habitat quality be predicted from land use? *Limnologica*, 35: 262-273, 2005.

[23] Docile TN, Figueiró R, Gil-Azevedo LH, Nessimian JL. Water pollution and distribution of the blackfly (Diptera: Simuliidae) in the Atlantic Forest, Brazil, *Revista de Biologia Tropical*, 63: 683–693, 2015.

[24] Ilésová D, Beracko P, Krno I, Halgos J. Effects of land use on black fly assemblages (Diptera: Simuliidae) in submontane rivers (West Carpathians, Slovakia), *Biologia*, 65: 892–898, 2010.

https://doi.org/10.2478/s11756-010-0096-5

[25] Kazanci N, Ertunç Ö. Use of Simuliidae (Insecta: Diptera) species as indicators of aquatic habitat quality of Yeşilırmak River Basin (Turkey), *Review of Hydrobiology*, 3: 27-36. 2010.

https://www.researchgate.net/publication/268224317

[26] Docile TN, Figueiró R, Molina O, Gil-Azevedo LH, Nessimian JL. Effects of Bacillus thuringiensis var. israelensis on the black Fly Communities (Diptera, Simuliidae) in Tropical Streams, Neotropical. *Entomology*, 50(2): 269-281, 2021.

https://doi.org/10.1007/s13744-020-00842-2

[27] Cuadrado LA, Moncada LI, Pinilla GA, Larrañag A, Sotelo AI, Adler PH. Black Fly (Diptera: Simuliidae) Assemblages of High Andean Rivers Respond to Environmental and Pollution Gradients, *Environmental Entomology*, 48(4): 815–825, 2019.

https://doi.org/10.1093/ee/nvz053

[28] Hendy A, Kruger A, Pfarr K, De Witte J, Kibweja A, Mwingira U, Dujardin JC, Post R, Colebunders R, O'neill S, Kalinga A. The blackfly vectors and transmission of Onchocerca volvulus in Mahenge, south eastern Tanzania, *Acta Tropica*, 181: 50–59, 2018.

https://doi.org/10.1016/j.actatropica.2018.01.009

[29] Kiel E. Behavioural Response of blackfly Larvae (Simuliidae, Diptera) to Different Current Velocities, *Limnologica*, 31: 179-83, 2001.

https://doi.org/10.1016/S0075-9511(01)80018-9

[30] Pepinelli M, Siqueira T, Salles FF, Shimbori EM. First report of Simuliidae and Chironomidae (Diptera) living on nymphs of Lachlania Hagen (Ephemeroptera: Oligoneuriidae) in South America, *Biota Neotropica*, 9 (1): 245-247, 2009.

https://doi.org/10.1590/S1676-06032009000100023

[31] Sanchez MO, Gil-Azevedo LH. Comments on the association of immatures of Hemerodromia (Diptera, Empididae) and Simulium (Diptera, Simuliidae), and first record of this association in the Atlantic Forest (Brazil), *Acta tropica*, 163: 7-8, 2016.

https://doi.org/10.1016/j.actatropica.2016.07.015

[32] Ginarte CA, Andrade CF, Campos-Gaona J. Larvas de simulídeos (Diptera: Simuliidae) do Centro Oeste, Sudeste e Sul do Brasil, parasitadas por Microsporídeos (Protozoa) e Mermitídeos (Nematoda), Iheringia, *Série Zoologia*, 93(3): 325–334, 2003.

https://doi.org/10.1590/S0073-47212003000300011

[33] Araújo-Coutinho CJ, Nascimento ES, Figueiró R., Becnel J.J. Seasonality and prevalence rates of microsporidia in Simulium pertinax (Diptera: Simuliidae) larvae in the region of Serra dos Orgãos, Rio de Janeiro, Brasil, *Journal of Invertebrate Pathology*, 85(3):188-191, 2004.

https://doi.org/10.1016/j.jip.2004.02.003

[34] Nascimento ES, Figueiró R, Becnel JJ, Araújo-Coutinho CJPC. Influence of temperature on microsporidia infections in a natural population of Simulium pertinax Kollar, 1832 (Diptera; Simuliidae), *Brazilian Journal of Biology*, 67(3): 519-526, 2007.

https://doi.org/10.1590/S1519-69842007000300017

[35] Figueiró R, Nascimento ES, Gil-Azevedo LH, Maia-Herzog M, Monteiro RF. Local distribution of blackfly (Diptera: Simuliidae) larvae in two adjacentstreams: the role of water current velocity in the diversity of blackfly larvae, *Revista Brasileira de Entomologia*, 52(3): 452-454, 2008.

https://doi.org/10.1590/S0085-56262008000300020

[36] Figueiró R, Maia-Herzog M, Gil-Azevedo LH, Monteiro RF. Seasonal variation in black fly (Diptera: Simuliidae) taxocenoses from the Brazilian Savannah (Tocantins, Brasil), *Journal of Vector Ecology*, 39(2): 321-327, 2014.

https://doi.org/10.1111/jvec.12107

[37] Figueiró R., Santos SS, Docile TN, Costa TR, Albuquerque CF, Gil-Azevedo LH. Preliminary observations on the patterns of co-occurrence of black fly (Diptera: Simuliidae) larvae and some of their potential macroinvertebrate predators, *Revista Brasileira de Entomologia*, 64(3): e20200046, 2020.

https://doi.org/10.1590/1806-9665-RBENT-2020-0046

[38] Carvalho BMC, Wainfas RL, Rodrigues T, Berbert LC, Franklin T, Lima-de-Sousa IK, Figueiró R. Black fly (Diptera:Simuliidae) larvae body size variation along an altitude gradient in the Itatiaia National Park, *Acta Ambiental Catarinense* 13 (1/2): 1-7, 2016.

https://doi.org/10.24021/raac.v13i1.3314

[39] Buitrago-Guacaneme A, Sotelo-Londonõ A, Pinilla-Agudelo GA, García-García A, Moncad LI, Adler PH. Abundance and diversity of black flies (Diptera: Simuliidae) in rivers of the Andean Eastern Hills of Bogotá (Colombia), and its relationship with water stream physicochemical variables, *Universitas Scientiarum*, 23 (2): 291-317, 2018.

https://doi.org/10.11144/Javeriana.SC23-2.aado

[40] Limberger PA, Tataje DAR, Novick D, Junior LCM, Strieder MN. Spatio-temporal distribution of preimaginal black flies (Diptera: Simuliidae) in streams of a sub-basinof the Uruguay river basin, southern Brazil, *Entomobrasilis*, 14: e94, 2021.

https://doi.org/10.12741/ebrasilis.v14.e946

[41] Menzel TC, Hentges SM, Reynalte-Tataje DA, Strieder MN. Diversity and spatial distribution of black flies (Diptera: Simuliidae) in the Ijuí river drainage basin, Rio Grande do Sul, Brazil, *EntomoBrasilis*, 12(2): 47-56, 2019.

https://doi.org/10.12741/ebrasilis.v12i2.845

[42] Menzel TC, Hentges SM, Loebens CM, Reynalte-Tataje DA, Strieder MN. Spatial and temporal distribution of black flies (Diptera: Simuliidae) in the Pampa biome streams, Brazil, *Biologia*, 76: 3711-3720, 2021.

https://doi.org/10.1007/s11756-021-00859-x

[43] Gentile R, Cardoso TS, Costa-Neto SF, Teixeira BR, D'andrea PS. Community structure and population dynamics of small mammals in an urban-sylvatic interface area in Rio de Janeiro, Brazil, *Zoologia*, 35: 1-12, 2018.

https://doi.org/10.3897/zoologia.35.e13465

[44] Tavares JA, Novaes RLM, Veríssimo I, Kuzel MAA., DA Costa-Neto SF, Rangel CL, Borges M, Medrado H, Alves B, Souza RF, Pinto-Menezes ACD, Menezes-Junior LF, Dias D, De Andreazzi CS, Gentile R, Moratelli R. Bats from the Pedra Branca Forest, Rio de Janeiro, Brazil, *Biodiversity Data Journal*, 9: e77400, 2021.

https://doi.org/10.3897/BDJ.9.e77400

[45] Avelino-Capistrano F, Almeida GL, Barbosa LS. Diversidade e distribuição de ninfas de Plecoptera (Insecta) por substratos em rios do Parque Estadual da Pedra Branca, Rio de Janeiro, *EntomoBrasilis*, 10(2): 76-81, 2017.

https://doi.org/10.12741/ebrasilis.v10i2.680

[46] Vallejo LR, Campos RM, Santos-Junior WM. Parque Estadual da Pedra Branca (RJ): Caracterização, Gestão e Conflitos Territoriais. In: Congresso Brasileiro de Unidades de Conservação. Fundação O Boticário de Proteção à Natureza, Curitiba, 2009.

[47] Figueiró R, Araújo-Coutinho CJPC, Gil-Azevedo LH, Nascimento ES, Monteiro RF. Spatial and temporal distribution of blackflies (Diptera: Simuliidae) in the Itatiaia National Park, Brazil, *Neotropical Entomology*, 35(4): 542-550, 2006.

https://doi.org/10.1590/s1519-566x2006000400018

[48] Angrisano EB. Metodologia para su Estudio. Insecta Trichoptera in Ecosistemas de Aguas Continentales (E.C. Lopretto & G. Tell eds.), 3nd edn., Ediciones Sur, La Plata, Argentina. p. 1199–1224, 1995.

[49] Merritt RW, Cummins KW. Trophic relations of macroinvertebrates. Methods in Stream Ecology (F.R. Hauer & G.A. Lamberti eds.), *Academic Press*, San Diego, California. p. 453–473, 1996.

[50] Wiggins GB. Larvae of the North America Caddisfly Genera (Trichoptera). University of Toronto Press, Toronto, Ontario, 1996.

[51] Carvalho AL, Werneck-de-Carvalho P, Calil ER. Description of the larvae of two species of Dasythemis Karsch, with a key to the genera of Libellulidae occurring in the states of Rio de Janeiro and São Paulo. (Anisoptera), *Odonatologica*, 31: 23–33, 2002.

[52] Da-Silva ER, Salles FF, Nessimian JL, Coelho LBN. A identificação das famılias de Ephemeroptera (Insecta) ocorrentes no Estado do Rio de Janeiro: chave pictórica para as ninfas, *Boletim do Museu Nacional Nova Serie Zoologica*, 508: 1–6, 2003.

[53] Olifiers MH, Dorville LFM, Nessimian JL, Hamada N. A key to Brazilian genera of Plecoptera (Insecta) based on nymphs, *Zootaxa*, 651(1): 1–15, 2004.

https://doi.org/10.11646/zootaxa.651.1.1

[54] Salles FF, Da-Silva ER, Hubbard MD, Serrão J.E. As espécies de Ephemeroptera (Insecta) registradas para o Brasil, *Biota Neotropica*, 4(2): 1–34, 2004.

https://doi.org/10.1590/S1676-06032004000200011

[55] Pes AMO, Hamada N, Nessimian JL. Chaves de identificação de larvas para famılias e gêneros de Trichoptera (Insecta) da Amazônia Central, Brasil, *Revista Brasileira de Entomologia*, 49(2): 181–204, 2005.

https://doi.org/10.1590/S0085-56262005000200002

[56] Passos MIS, Nessimian JL, Ferreira JRN. Chave para a identificação dos gêneros de Elmidae (Coleoptera) ocorrentes no Estado do Rio de Janeiro, Brasil, *Revista Brasileira de Entomologia*, 51: 42–53, 2007.

[57] Hamada N, Adler PH. Bionomia e chave para imaturos e adultos de Simulium (Diptera: Simuliidae) na Amazônia Central, Brasil, *Acta Amazonica*, 31(1): 109-132, 2001.

https://doi.org/10.1590/1809-43922001311132

[58] Coscarón S, Coscarón-Arias CL. Neotropical Simuliidae (Diptera: Insecta). In: Adis J, Arias JR, Rueda-Delgado G, Wantzen KM. (eds.) Aquatic biodiversity in Latin America. Pensoft Publishers, Sofia, Bulgaria, pp. 685, 2007.

[59] Gil-Azevedo LH, Ferreira JRN, Maia-Herzog M. Chave deidentificação de pupas de Simuliidae (Diptera) do sudeste do Brasil, *Revista Brasileira de Zoologia*, 22(3): 742-752, 2005.

https://doi.org/10.1590/S0101-81752005000300035

[60] Lenat DR. Water quality assessment using a qualitative collection method for benthic macroinvertebrates, *Journal of the North American Benthological Society*, 7(3): 222-233, 1988.

https://doi.org/10.2307/1467422

[61] Srisuka W, Takaoka H, Otsuka Y, Fukuda M, Thongsahuang S, Taai K., Choochote W, Saeung A. Seasonal biodiversity of black flies (Diptera: Simuliidae) and evaluation of ecological factors influencing species distribution at Doi Pha Hom Pok National Park, Thailand, *Acta Tropica*, 149: 212–219, 2015.

https://doi.org/10.1016/j.actatropica.2015.05.024

[62] Coppo TL, Lopes J. Diversidade de Simuliidae (Diptera: Nematocera) de três cursos d'água no parque ecológico da Klabin S.A. - Telêmaco Borba, Estado do Paraná, Semina: *Ciências Biológicas da Saúde*, 31(1): 3-14, 2010.

[63] Kuvangkadilok C, Boonkemtong C, Phayuhasena S. Distribution of the larvae of blackflies (Diptera: Simuliidae) at Doi Inthanon National Park, Northern Thailand, *Southeast Asian Journal Tropical Medicine Public Health*, 30(2): 328-326, 1999.

[64] Gaona JC, Andrade CFS. Aspectos da entomologia médica e veterinária dosborrachudos (Diptera: Simuliidae). Biologia, Importância e Controle. LECTA, São Paulo, 17(1): 51-65, 1999.

[65] Coscarón S, Arias CLC. Aquatic Biodiversity in Latin America: Neotropical Simuliidae (Diptera: Insecta). 3. ed. Bulgaria: Pensoft, 2007.

[66] Vannote RL, Sweeney BW. Geographic analysis of thermal equilibria: a conceptual model for evaluating the effect of natural and modified thermal regimes on aquatic communities, *American Naturalist*, 115: 667-695, 1980.

[67] Mccreadie JW, Adler PH, Burger JF. Species assemblages of larval black flies (Diptera: Simuliidae): random or predictable? *Journal of the North American Benthological Society*, 16(4): 760-770. 1997.

https://doi.org/10.2307/1468169

[68] Ciadamidaro S, Mancini L, Rivosecchi L. Black flies (Diptera, Simuliidae) as ecological indicators of stream ecosystem health in an urbanizing area, *Annali dell'Istituto Superiore di Sanità*, 52(2): 269-276, 2016.

https://doi.org/10.4415/ANN_16_02_20

Distribución espacial de taxocenosis inmaduras de moscas negras (Diptera: Simuliidae) en el Parque Estatal de Pedra Branca, Brasil

Resumen: Para comprender la relación entre los factores ambientales y la distribución de especies de moscas negras (Simuliidae), realizamos un muestreo de individuos inmaduros en el Parque Estatal de Pedra Branca, Río de Janeiro, Brasil, durante la temporada seca de junio de 2018. Luego, correlacionamos los valores de las variables ambientales con los valores de abundancia de larvas de mosca negra mediante análisis de redundancia. Los valores de abundancia de *Simulium pertinax* Kollar 1832, *Simulium subpallidum* Lutz 1910, *Simulium (Thyrsopelma)* spp., *Simulium (Psaroniocompsa)* spp. y *Simulium (Inaequalium)* spp. se correlacionaron negativamente con la luz y la temperatura, mientras que la abundancia de *Simulium (Hemicnetha)* spp. se correlacionó positivamente con estas variables. Los valores de abundancia de *Simulium pertinax* Kollar 1832, *Simulium (Hemicnetha)* spp. y *Simulium (Thyrsopelma)* spp. se asociaron positivamente con la altitud, mientras que los valores de las otras especies revelaron correlaciones negativas con este factor. La riqueza de especies se determinó mediante una regresión lineal con el índice Ephemeroptera, Plecoptera Trichoptera (EPT) utilizando los valores de las pupas. Este análisis mostró que los sitios más deteriorados tienen una mayor riqueza de especies que los sitios más conservados dentro del Parque Estatal de Pedra Branca. Nuestros resultados sugieren que la riqueza de especies de simúlidos refleja la integridad del hábitat, convirtiéndose así en un indicador confiable del estado de conservación del mismo.

Palabras Clave: Bioindicadores; Ecología comunitaria; Macroinvertebrados; Simúlidos

Distribuição espacial de taxocenoses imaturas de moscas negras (Diptera: Simuliidae) no Parque Estadual da Pedra Branca, Brasil

Resumo: Para compreender a relação entre os fatores ambientais e a distribuição das espécies de moscas negras (Simuliidae), realizamos uma amostragem de indivíduos imaturos no Parque Estadual da Pedra Branca, Rio de Janeiro, Brasil, durante a estação seca em junho de 2018. Em seguida, correlacionamos os valores das variáveis ambientais com os valores de abundância das larvas de mosca negra por meio de análises de redundância. Os valores de abundância de *Simulium pertinax* Kollar 1832, *Simulium subpallidum* Lutz 1910, *Simulium (Thyrsopelma*) spp., *Simulium (Psaroniocompsa*) spp. e *Simulium (Inaequalium*) spp. apresentaram uma correlação positiva com a luz e a temperatura, enquanto que a abundância de *Simulium (Hemicnetha*) spp. apresentou uma correlação negative com essas variáveis. Os valores de abundância de *Simulium pertinax* Kollar 1832, *Simulium (Hemicnetha)* spp. e *Simulium (Thyrsopelma)* spp. apresentaram om corralação positiva com a altitude, enquanto os valores das outras espécies revelaram correlações negativas com esse fator. A riqueza de espécies foi determinada por meio de uma regressão linear com o índice Ephemeroptera, Plecoptera Trichoptera (EPT) utilizando os valores das pupas. Essa análise mostrou que os locais mais deteriorados apresentam uma maior riqueza de espécies do que os locais mais conservados dentro do Parque Estadual da Pedra Branca. Nossos resultados sugerem que a riqueza de espécies de simuliídeos reflete a integridade do habitat, tornando-se assim um indicador confiável do estado de conservação do mesmo.

Palavras-chave: Bioindicadores; Ecologia de comunidades; Macroinvertebrados; Simuliídeos

Tayanna Rodrigues da Costa

Bachelor's degree in Biological Sciences and Master of Science in Environmental Science and Technology from the State University of Rio de Janeiro (UERJ).

ORCID: 0000-0001-8255-2164

Tatiana Nascimento Docile

She is currently Adjunct Professor at the Fernando Rodrigues da Silveira Application Institute-CAP/UERJ. Graduated in Biological Sciences from Universidade Federal Fluminense (UFF). Master (2014) and PhD (2018) in ecology from the Federal University of Rio de Janeiro (UFRJ).

ORCID: 0000-0001-8964-5662

Christina Albuquerque Ferreira

Supervision, Method Development and Quality Management in the Radiochemistry Laboratory of the CENTRO DE BIOLOGIA EXPERIMANTAL OCEANUS. Biologist, with a Master's degree in Environmental Science and Technology; Specialization in Quality Management and Teaching of Biological Sciences.

ORCID: 0009-0009-5452-3054

Tainá Maria Miranda Souza Martins.

Bachelor's Degree in Biological Sciences from the State University of Rio de Janeiro(UERJ), and Master of Science in Environmental Science and Technology from the same institution. Holds a specialization course in Environmental Sciences at Educational Technical Foundation Souza Marques and currently is a PhD candidate in Biological Sciences - Zoology at the Postgraduate Program of the National Museum - UFRJ.

ORCID: 0000-0002-1285-7031

Ivyn Karla Lima-de-Sousa.

Has a first degree in Biotechnology from Universidade do Estado do Rio de Janeiro (UERJ), and a second degree in Biological Sciences (Licenciatura) from Universidade Cruzeiro do Sul and a Master's degree in Biological Sciences - Zoology from the Graduate Program from the National Museum - UFRJ. Currently is a PhD candidate in Biological Sciences - Zoology at the Postgraduate Program of the National Museum - UFRJ.

ORCID: 0000-0001-5756-805X

Leonardo Henrique Gil Azevedo.

He is currently an Adjunct Professor at the Department of Entomology at the National Museum of the Federal University of Rio de Janeiro (UFRJ), where he is Curator of the Entomological Collection Collection (MNRJ). He has a degree in Biological Sciences from the Federal University of Rio de Janeiro (2003), Master's (2006) and PhD (2010) in Biological Sciences (Zoology) from the National Museum - Federal University of Rio de Janeiro. He has experience in Zoology, with emphasis on Taxonomy, Phylogeny, Morphology and Biology of Diptera (Insecta), with emphasis on Simuliidae, Blephariceridae and other aquatic dipterans.

ORCID: 0000-0002-0389-2185

Ronaldo Figueiró

He is currently an Associate Professor at the Faculty of Biological Sciences and Health (FCBS) of the State University of Rio de Janeiro (UERJ), where he teaches at the undergraduate level and at the Professional Master's Program in Environmental Science and Technology (PPGCTA).

Graduated in Biological Sciences (Bachelor's Degree in Ecology) at the Federal University of Rio de Janeiro, the same institution where he completed his Master of Science and PhD degrees in Ecology. Between 2013 and 2015, he held a postdoctoral position in the Graduate Program in Biodiversity and Health (PPGBS/Fiocruz).

ORCID: 0000-0003-0762-1312