Systematics, Morphology and Biogeography

An estimate of the potential number of mayfly species (Ephemeroptera, Insecta) still to be described in Brazil

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ABSTRACT

This study reviewed the data on the Brazilian Ephemeroptera, based on the studies published before July, 2013, estimated the number of species still to be described, and identified which regions of the country have been the subject of least research. More than half the species are known from the description of only one developmental stage, with imagos being described more frequently than nymphs. The Brazilian Northeast is the region with the weakest database. Body size affected description rates, with a strong tendency for the larger species to be described first. The estimated number of unknown Brazilian species was accentuated by the fact that so few species have been described so far. The steep slope of the asymptote and the considerable confidence interval of the estimate reinforce the conclusion that a large number of species are still to be described. This emphasizes the need for investments in the training of specialists in systematics and ecology for all regions of Brazil to correct these deficiencies, given the role of published papers as a primary source of information, and the fundamental importance of taxonomic knowledge for the development of effective measures for the conservation of ephemeropteran and the aquatic ecosystems they depend on.

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Introduction

A number of studies – see review in Nabout et al. (2013) – have concluded that the vast majority of the world’s biodiversity, that is, the total number of species, has yet to be documented. Despite this, ongoing anthropogenic impacts continue to provoke the loss of many species, which has negative consequences for the structure and functioning of ecosystems (Cardinale et al., 2012). The development of effective measures to mitigate these losses will depend not only on the systematic understanding of the species present in the environment, but also on the ecological importance of these taxa for the functioning of the ecosystem. However, the number of new species that have been found in recent years, even in groups considered to be well-studied, reinforces the idea that estimates of the total diversity of most taxa need to be revised considerably (Barratt et al., 1997; Williams et al., 2006).

Given the ongoing and unlimited exploitation of natural resources by human populations, the conservation of biological diversity is a global challenge. In this context, the loss and fragmentation of habitats and environmental pollution are the principal factors responsible for species extinctions (Loyola and Levinsohn, 2009). This challenge is even greater in the Neotropical region, which is characterized by high levels of diversity and relatively incomplete evidence on species diversity, in comparison with most other regions (Kier et al., 2005; Whittaker et al., 2005). Worse still, the Neotropics are also characterized by a relative paucity of biological research, especially in comparison with the accelerated rates of deforestation occurring in most regions (Freitag and Van Jaarsveld, 1998; Myers et al., 2000). This determines high rates of loss of biological diversity – up to three species per hour, by some estimates (Lawton and May, 1995; Wilson, 1999; Chapin et al., 2000) – especially in the more sensitive environments that have been most affected by anthropogenic impacts.

Despite being recognized as a “megadiverse” country, Brazil continues to lose its pristine natural habitats at high rates, even in regions, such as the Amazon Forest, where much of the environment is protected by law. Other biomes, such as the cerrado, caatinga, and Atlantic Forest, have only 36.73%, 24.39%, and 21.80%, respectively, of the original forest cover intact (MMA, 2014). This reinforces the need for inventories, taxonomic studies, and...
additional research into the distribution and abundance of species, in order to understand the gaps in the data, as well as providing sound taxonomic and ecological guidelines for the definition of research and conservation priorities (Pimm et al., 2001; De Marco and Vianna, 2005).

Aquatic macroinvertebrates are among the organisms most affected by the ongoing alteration of natural habitats. This is because they may be influenced not only by physical modifications of the environment, on a landscape scale, but also by changes in the chemical composition of the water, given that such a large proportion of their life cycle is aquatic (Yoshimura, 2012). These organisms are also highly diverse and abundant, have a relatively long life cycle, and can respond to relatively subtle changes in habitat characteristics and the intensity of impacts (Lenat and Barbour, 1994; Alba-Tercedor unpublished, 1996; Oliver et al., 1998; Lewinsohn et al., 2005).

The mayflies (Ephemeroptera) constitute one of the groups of aquatic macroinvertebrates most used as bio-indicators of environmental quality (Callisto et al., 2001; Salles et al., 2010), due primarily to the fact that these insects are highly sensitive to alterations in the physical structure and water quality of streams (Rosenberg and Resh, 1993), and are found in the proximity of both lentic and lotic bodies of water. These insects also represent prey items for a diversity of aquatic predators, both vertebrates and invertebrates, and thus constitute an important component of the trophic networks of tropical river systems, in terms of their contribution to the transformation of organic matter, energy flow, and nutrient cycling (Dodd, 2002).

Globally, this order contains 442 genera and 3269 species, according to the review of Barber-James et al. (2013). In Brazil, a total of 10 families have been recorded up to now, containing 73 genera and 291 species (see http://ephemeroptera.com.br/). This corresponds to 8.87% of global ephemeropteran diversity. The first description of a Brazilian mayfly was that of Campsurius albicans by Guérin and Percheron (1838), although most of the early studies were conducted in the first half of the twentieth century (Da-Salva and Salles, 2012). Until the 1980s, most of the published studies focused on taxonomy and systematics, and few data were provided on the biology or geographic distribution of the species (e.g. Walker, 1853; Froehlich, 1969; Da-Silva, 1997; Salles et al., 2004a; Falcão et al., 2011).

In Brazil, research on mayflies is still characterized by Wallacean shortfalls, that is, a lack of data on species distinctions, as well as many Linnaean deficits, due to the paucity of taxonomic and phylogenetic data, despite the fact that most published studies have focused on these topics (Diniz-Filho et al., 2013). The fact that this order is a bio-indicator which constitutes an important tool for the bio-monitoring of environmental quality (Rosenberg and Resh, 1993; Bauernfeind and Moog, 2000; Buss and Salles, 2007) demands that reliable data are available on its taxonomy, life cycle, and geographic distribution in regions with distinct characteristics (Thomanzin and Thomanzini, 2000; Büchs, 2003). However, the lack of reliable information represents a fundamental drawback for the development of effective conservation policies and strategies (Pimm and Brown, 2004; Grand et al., 2007). Even simple measures, such as the production of lists of endangered species, are still unavailable for the Ephemeroptera.

Given the enormous size of the country – more than eight million square kilometers – and the quantity and heterogeneity of its aquatic environments, many authors have concluded that the real number of species that occur in Brazil is considerably higher than the available estimates of diversity suggest (Lewinsohn and Prado, 2002, 2005; Agostinho et al., 2005; Lewinsohn et al., 2005; Mittermeier et al., 2005). This not only hampers conservation and management efforts for aquatic environments, but also limits the scope of phylogenetic and zoogeographic studies of the ephemeropterans themselves. This lack of data also hinders the identification of priority areas for research and conservation planning, especially considering the scant resources available for such efforts.

Based on these conclusions, the present study estimated the potential number of ephemeropteran species still to be described in Brazil, and investigated the influence of body size on the discovery of species. The analyses were based on four main questions: (i) have nymphs and imagos been described in the same proportion? (ii) Does body size influence the probability of species detection? (iii) How many Brazilian ephemeropteran species have yet to be described? and (iv) which Brazilian region is most deficient in ephemeropteran research?

The answers to these questions will be essential for the understanding of the biological richness of this group, and will contribute to the development of more effective research in the fields of systematics, ecology, and phylogeny. It is also hoped that these answers may stimulate further scientific interest in the mayflies, given the challenges of research in the natural environment, and in particular the development of conservation measures, which are still incipient, even though a few ephemeropteran species are already listed as endangered (Lewinsohn et al., 2005).

Material and methods

We consulted the actual species list of Brazilian Ephemeroptera, available in Brazilian Ephemeroptera site. In this site, all species registered to Brazil are listed and synonyms are cited, so we could control duplicate data. We used all papers providing descriptions of ephemeropteran species from Brazil published between 1838 and July, 2013 (see Supplementary Material 1), were consulted through the following sites: Ephemeroptera Galactica (www.ephemeroptera-galactica.com/), Ephemeroptera of the World (www.insecta.bio.spbu.ru/?Eph-spp/Contents.htm), Scielo (www.scielo.com.br), Web of Science (wwwapps.webofknowledge.com), JStor (www.jstor.org/), Limnology Journal (www.jlimnol.it/index.php/jlimnol), Taylor & Francis Online (www.tandfonline.com/), Wiley Online Library (http://onlinelibrary.wiley.com/), Cambridge Journals (http://journals.cambridge.org), and Zootaxa (www.zootaxa.com.br). When availability of a following information was tabulated: (i) the year of the description, (ii) holotype locality, (iii) presence or absence of description of each life stage, and (iv) mean size of the nymph and the male and female imagines. When body size classes were presented, the mean of these values was calculated.

Geographic data were based on the localities at which the holotypes were collected. For analysis, these sites were allocated to one of the five Brazilian regions – North, Northeast, Midwest, South, and Southeast.

Statistical analyses

Paired t tests were used to evaluate possible differences in the description of life stages (nymph vs. imago) and sexes (adult males vs. adult females), based on the year each species was published. To verify the possible variation in the rate of descriptions based on either nymphs or imagos over time, a simple linear regression was applied once the homoscedasticity of the residuals and the homogeneity of their variance was established (Zar, 2010).

A simple linear regression was also used to verify whether the body size of the ephemeropteran species influenced the probability of description over time, using the year of publication as the response variable. In comparative studies of closely-related species, it is important to take phylogenetic patterns into account, given that the lack of independence among the taxa may bias the statistical analyses (Felsenstein, 1985; Freckleton et al., 2002; Martins
et al., 2002). Given this, the phylogenetic relationships among the 291 mayfly species were defined based on the literature and research at specific sites (see Supplementary Material 2 and 3). The independence of the data was evaluated using a linear regression of the phylogenetic pattern, and the residual of this regression was tested using Moran’s I coefficient, based on correlograms with five distance classes (Sokal and Oden, 1978). The I values vary from $-1.0$ to 1.0, and the analysis is equivalent to a correlation coefficient, in other words, values close to 0.1 indicate that species allocated to a given phylogenetic class tend to be more similar to one another in relation to the trait analyzed than expected by chance, whereas values tending toward $-1.0$ indicate dissimilarities among the species. Moran’s index of phylogenetic distances given by:

$$I = \frac{n}{N} \left( \frac{\sum_{i=1}^{n} \sum_{j=1}^{n} (y_i - \bar{y}) (y_j - \bar{y}) w_{ij}}{\sum_{i=1}^{n} (y_i - \bar{y})^2} \right)$$

where $n$ = the number of species analyzed; $y_i$ and $y_j$ = values of $y$ for species $i$ and $j$, $\bar{y}$ = the mean value of $y$, $w_{ij}$ = the element of the symmetric square matrix $W$, which expresses the phylogenetic relationships among the $n$ species, the sum of which, between $i$ and $j$ is equal to $S$.

Significant positive values of $I$ for the first distance class indicate the existence of a phylogenetic pattern in the residual of the regression and thus an increase in type I errors. Where evidence of a phylogenetic pattern was found in the residuals, we generated phylogenetic filters using the Phylogenetic Eiingen-vector Regression or PFR approach (Diniz-Filho et al., 1998, 2009), with three filters – 1, 3, and 4 – which minimize the phylogenetic autocorrelation in the residuals – being selected. These three filters were used as predictors for a partial multiple regression, for which the response variable was the year in which the species was described, and the predictor variables were body size and the phylogenetic filters. The filters were thus used to control for phylogenetic dependence. In this multiple regression, four partial components were obtained: [a] variation in the year of description explained solely by body size, [b] variation explained solely by the phylogenetic structure, [c] the component shared between [a] and [c], and [d] the residual variation. All these analyses were run in the Spatial Analysis and Macroecology (SAM) program (Rangel et al., 2010).

Variation in the rates of species description by year among the five Brazilian regions (North, Northeast, Midwest, South, and Southeast) was evaluated using a one-way Analysis of Variance (ANOVA), given the assumptions of normality and homoscedasticity. To estimate the number of ephemeropteran species found in Brazil, the date (year) of the description and the number of species described per year were recorded. These two variables were used to estimate the cumulative species curve for Brazil. In this case, it is important to remember that a finite number of species exist on the planet (Cabral-Saúdo and Lobo, 2003), and that, as the number of recorded species nears the total number of species in existence, it becomes increasingly difficult to discover new species (Diamond, 1985; Medellín and Soberón, 1999; Cabral-Saúdo and Lobo, 2003). Given this, the cumulative number of described species was related to the year of description, and finally, was adjusted to three curvilinear models – the extreme (Williams, 1995), logistic, and Gompertz (Ratkowsky, 1990) models used Statistica 12 program (StatSoft Inc., 2014). Both functions were adjusted using the Quasi-Newton method and criterion of convergence 0.0001. We used default values of the program for starting values (0.1) and sept-width (0.5) for all parameters. These default values always led to a satisfactory fit.

These models are considered sigmoid models and present three parameters, of which the first indicates the asymptote, while the other two form the curve. Dengler (2009) reviewed these models (among others), detailing each equation. The parameters $b$ indicates the upper asymptote. Thus to estimate the number of species we adjusted the models considering all criteria of the models and observed the asymptote parameter. All three models are frequently used to describe cumulative species curves (Tjurve, 2003). Finally the models were compared using Akaike’s Information Criterion, or AICc (Motulsky and Christopoulos, 2004):

$$AICc = N \times \ln \left( \frac{SS}{N} \right) + 2K + \left( \frac{2K(K+1)}{N-K-1} \right)$$

where $N$ = number of records; $SS$ = sum of squares, and $K$ = the number of model parameters.

**Results**

A total of 178 published papers were identified which describe 286 of the 291 mayfly species known to occur in Brazil (Supplementary Material 1). Five species – Campsurus burmeisteri (Ulmer, 1942), Campsurus indivisus (Ulmer, 1942), Campsurus melanoccephalus (Pereira and Da-Silva, 1991), Lachlania bonanovae (Da-Silva and Pereira, 1993), and Thraulodes limbatus (Navás, 1936) – were omitted from the analysis because it was impossible to consult the original descriptions. Descriptions that did not present data pertinent to the present analysis were omitted from our study.

In the data set as a whole, only 81 species were described based on the analysis of both life stages, while there were 40 descriptions only from imagos male, 10 exclusively with imagos female and 83 only from nymphs (Fig. 1), representing a significant difference ($t = 2.012, df = 132, p = 0.046$). In general, however, only the descriptions of species of the families Baetidae, Leptophlebiidae and Oligoneuriidae were based more on nymphs than adults (Table 1), and the description of all the life stages of each species was only available for monotypic families.

In only 114 of the 291 species descriptions, both male and female imagos were analyzed. A larger number of descriptions included male imagos (184) in comparison with those including females (133) (Fig. 2), although the difference in the sexes among years was not significant ($t$ (unequal variances) = 1.454, df = 116, $p = 0.148$).

Body size appeared to have an effect on description rates (Fig. 3A), with the first species described being significantly larger, on average, than those described more recently ($r^2 = 0.18, b = -0.43, p < 0.001$). However, a phylogenetic pattern was found in the regression residual (Fig. 3B), with a phylogenetic correlation being found between the residual of the regression and body size ($r = 0.882, p < 0.001$). In order to control for this apparent effect...
Table 1
Proportion of the descriptions of species belonging to the different ephemeropteran families published up until July, 2013, based on different life stages (nymph, male and female imagos).

<table>
<thead>
<tr>
<th>Family</th>
<th>Nymphs</th>
<th>Male imagos</th>
<th>Female imagos</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baeotidae</td>
<td>91.4</td>
<td>38.71</td>
<td>36.56</td>
</tr>
<tr>
<td>Caenidae</td>
<td>63.16</td>
<td>84.21</td>
<td>42.10</td>
</tr>
<tr>
<td>Leptophlebiidae</td>
<td>86.95</td>
<td>63.04</td>
<td>47.82</td>
</tr>
<tr>
<td>Coryphoridae</td>
<td>65.38</td>
<td>78.20</td>
<td>46.15</td>
</tr>
<tr>
<td>Oligoneuriidae</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
</tr>
<tr>
<td>Ephemerida</td>
<td>60.00</td>
<td>50.00</td>
<td>60.00</td>
</tr>
<tr>
<td>Leptophlebiidae</td>
<td>13.16</td>
<td>78.94</td>
<td>55.26</td>
</tr>
<tr>
<td>Coryphoridae</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
</tr>
<tr>
<td>Oligoneuriidae</td>
<td>25.00</td>
<td>100.00</td>
<td>75.00</td>
</tr>
<tr>
<td>Euthyplocidae</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
</tr>
</tbody>
</table>

Fig. 2. Simple linear regression between the number of ephemeropteran species described based on male/female specimens and the year the description was published.

of phylogenetic dependence, phylogenetic filters were generated using the PVR approach (see Methods section), and inserted into the multiple regression, for which the partial components were investigated. This partial regression upheld the conclusion that body size was an important factor in species description rates (Table 2). The residual of this new regression had no phylogenetic dependence (Moran’s I, first class = 0.01; p = 0.09).

Table 2
Partial components of the partial multiple regression: a = component explained by the environment; b = shared component; c = component explained by phylogeny; d = residual.

<table>
<thead>
<tr>
<th></th>
<th>R²</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>0.09</td>
<td>0.01</td>
</tr>
<tr>
<td>B</td>
<td>0.09</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>0.06</td>
<td>0.01</td>
</tr>
<tr>
<td>D</td>
<td>0.74</td>
<td></td>
</tr>
</tbody>
</table>

Fig. 4. Analysis of variance between the Brazilian regions in which the ephemeropteran holotypes were collected and the number of species described. Values with different letters are significantly different.

Most of the species were described from holotypes collected in the southeastern (78 species) and northern regions (74) of Brazil, with 43 being recorded in the South, 16 in the Midwest, and 11 in the Northeast. Significant variation was found among regions in the number of taxa described ($F_{4,22} = 6.92, p < 0.001$), with significant differences being confirmed between the Northeast and both the South ($p = 0.013$) and Southeast ($p = 0.001$). The Northeast was represented by 13 fewer descriptions than the South, on average, and 18 fewer than the Southeast (Fig. 4).

The extreme model (Fig. 5) provided the best adjustment of the data between the number of species and the year, given that it returned the lowest AICc (Table 3). As the number of known species is lower than the total number predicted by the asymptote, this model indicates that many new species of Brazilian

Fig. 3. (A) linear regression between mean body size of ephemeropteran species discovered in Brazil, and the year of the species description; (B) phylogenetic correlogram of the residual of the linear regression. The white circle indicates a significant value for Moran’s I ($p < 0.0001$).
The nymphs are often very similar to one another in many morphological parameters, and their descriptions and identification keys often depend on highly specific characteristics, which permit the differentiation of species, but also hamper the whole process of the identification and description of taxa (e.g., Lugo-Ortiz and McCafferty, 1996; Salles and Lima, 2011). The presence of atrophied mouthparts and vestigial legs in adults, related to reproductive adaptations (Da-Silva and Salles, 2012), also contributes to the reduced number of traits available for the description of this stage.

Despite this, three families – Baetidae, Leptohyphidae, and Oligoneuriidae – are characterized by a predominance of species descriptions based on the analysis of the nymphs. We believe that this apparent discrepancy can be accounted for by the fact that most of these descriptions were derived from ecological studies, which tend to focus on the nymphs (Shimano et al., 2013), and thus make up the majority of the specimens forwarded to the specialists for analysis. Ultimately, the studies of these families may be more refined simply because there are more specialists in Brazil.

The similarity in the number of male and female specimens used in the species descriptions probably reflects the efforts of taxonomists to balance the sampling, given that the early studies were based primarily on female specimens. Given this, we believe that the specialists, recognizing this deviation, have spent more time describing males, with the aim of balancing the available data, and subsequently focused more on the males once they realized that the male genitalia provides more diagnostic features for the differentiation of species than that of the females. This would have led to the overall equilibrium between the sexes in species descriptions (Fig. 2).

Overall, only around 17% of species were described based on specimens of both nymphs and adults. It is important to note here that the effective classification of taxa depends on the diagnosis of all life stages, given that many structures are modified or become more or less visible during these different stages. In the specific case of the Ephemeroptera, we believe that the lack of data on the characteristics of one life stage or one of the sexes, in the case of the adults, may often result in errors of identification, especially under field conditions, when ecologists, rather than taxonomists, identify the specimens. This may result in shortcomings and misinterpretations in many areas of research (Mariano and Polegato, 2011), given the fundamental need for an adequate classification of organisms (Marques and Lamas, 2006). In some extreme cases, new taxa may be falsely described due to the lack of data on one or other life stage (Falcão et al., 2011).

The observed tendency for larger-bodied species to be described first was expected, given the limitations of the equipment available historically for the examination of small insects. In fact, the lack of information on the biology of the group and collection procedures may have imposed certain limitations on the early studies. The first species recorded in Brazil was *C. albicans* (Guérin and Percheron, 1838), and while the author did not provide measurements for the specimen, *Eaton* (1871) subsequently reported a body length of 10 mm for a male imago, a relatively large size by the standard of most species described in recent years, which tend to measure 5 mm or less.

While body size has influenced description rates, other factors may also be important, including the number of researchers involved in studies of ephemeropolitan in Brazil, the effectiveness of the materials used historically for collection of specimens and other data, and the lack of information on the distribution of these insects, which is still poorly known in much of the country. In the older studies, in fact, a little or no information was provided on the habitat in which the specimens were collected, and this alone may have hampered the collection of additional studies for the development of more detailed studies on the taxonomy of a given species or
genus. Given this, we would recommend that some species descriptions be revised and/or complemented by data on the body size of the holotype, for example, and the description of life stages or the sex not included in the original study (e.g. Salles and Serrão, 2005; Dominguez et al., 2009; Lima et al., 2010).

In recent years, taxonomic reviews and re-evaluations have led to changes in the listing of ephemeropteran species, the diagnostic characteristics of the different taxa, and known distributions. The use of advanced microscopy techniques has permitted the discovery of new species, which are often of relatively small body size. Cooperation between research institutions, both in Brazil and abroad, has also helped to reduce Linnaean and Wallacean deficits, contributing to improvements in the reliability and efficiency of mayfly studies. Body size also has an effect on other fields of research – Orlofske and Baird (2013), for example, found that, in bio-monitoring studies, whereas larger specimens tend to be identified to genus or even species, smaller specimens are typically identified to no more than the family level.

Another aspect of the results is the geographic distribution of the holotypes, in particular, the lack of studies from the Brazilian Northeast, a region that includes nine states and makes up almost a fifth (18.27%) of the country's total area. In fact, no holotypes have been described from five of these states, and the total number of species known from this region is highly disproportionate to that of the South and Southeast, which are smaller in area. The paucity of data from the Northeast almost certainly reflects the lack of ephemeropteran researchers based in this region, although in recent years, specialists have become established in the states of Bahia and Pernambuco.

Research in this region began only recently, with the first record of a Baetidae being reported by Lugo-Ortiz et al. (2002), and the first species being described within the past decade (Salles and Serrão, 2005; Salles et al., 2005). This emphasizes the need for further studies in the region, given that most of the area has been surveyed only superficially, when at all. A scarcity of research in the Brazilian Northeast is also typical of other macroinvertebrates, such as the Odonata (De Marco and Vianna, 2005).

By contrast, the largest number of ephemeropteran researchers are based in the Brazilian Southeast, and this is reflected in the numbers of species descriptions. While fewer researchers are based in the northern region, the vast Amazon basin (Ab/Saber, 2002) attracts scientists from other regions, and around the world. This does not mean, however, that these other regions of the country have been surveyed adequately, given the vast size of the country and the complexity of its hydrographic network. Unfortunately, the number of researchers and the resources available for the study of ephemeropteran diversity are still far from adequate, given the apparent complexity of this group of insects. For this reason, efforts should be focused on the description of species as a basic parameter for the understanding of conservation priorities.

The large number of as yet undescribed Brazilian species predicted by the results of the present study was expected, however, especially considering that Da-Silva and Salles (2012) estimated a total of only 8000 species for the whole planet, and only 600 for Brazil, in contrast with 10,864 species predicted for this country here. It is important to remember, however, that a certain amount of variation was found among the estimates, among both methods (non-linear) and groups, probably related to the reduced number of known species and the steepness of the curve.

The paucity of data on Brazilian mayflies is a fundamental problem here, and this is reflected in the broad confidence interval recorded in the present study (Table 2). Whatever the reliability of the estimate presented here – it will almost certainly be altered as data accumulate – what is certain is that many mayfly species have yet to be described in Brazil. The factors that contribute to the considerable disparity between this estimate and the number of known species include (i) the recent development of ephemeropteran research in Brazil, (ii) the paucity of specialists, (iii) the lack of financial resources for fieldwork, (iv) the shortage of government incentives for research, and (v) the long period of time required to train specialists. In addition, the paucity of scientific collections and taxonomic studies means that most tropical species are still unknown (Da-Silva and Salles, 2012).

Current scientific knowledge of the biological diversity of the mayflies (Ephemeroptera) is assumed to be highly deficient, and despite the growth of research in Brazil in recent years (Falcão et al., 2011; Shimano et al., 2013), additional studies will be necessary to overcome the limitations of the available data, as well as covering the considerable gaps that persist in the understanding of the taxonomy of the order (Salles et al., 2004b).

Focusing on the zoogeography of the mayflies may contribute to the management of areas that have suffered high rates of loss of biodiversity, as well as elucidating the relationship between these organisms and the environments they inhabit. In addition to this research, some known species require revision in order to refine the original descriptions. Raising nymphs until the adult stage would also be a way of identifying taxa more precisely. This type of approach would also provide more reliable and informative data for studies in other areas, such as ecology and systematics.

**Conflicts of interest**

The authors declares no conflicts of interest.

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**Appendix A. Supplementary data**

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.rbje.2015.03.014.

**References**


