

The role of environmental and spatial factors in the assembly of aquatic insect communities in southern Brazilian temporary ponds

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Abstract Metacommunity organisation in temporary ponds is assembled by spatiotemporally varying processes that are also contingent on the species' dispersal ability and scale of observation. Aquatic insects are useful models to study the relative contribution of environmental and spatial factors to community assembly in view of the differing dispersal ability existing in the species from this group. We assessed the metacommunity organisation of aquatic insects and subsets of strong- and weak-flying insects in relation to environmental (habitat structure; water chemistry; climate) and spatial factors in temporary ponds ranging along a latitudinal gradient in southern Brazil. Local and climate environmental factors (water chemistry and temperature) along with fine-scale spatial factors were the main drivers of the total insect community, although their relative importance shifted between subsets of weak- and strong-flying insects. The composition of strong-flying insects was structured by climate and fine-scale spatial factors, while weak-flying insects, by local (water chemistry) and spatially structured climate. This suggests that strong-flying insects showed stronger signatures of mass effects at finer scales, while weak-flying insects were more strongly affected by species sorting coupled to local environmental factors and regional climate. In summary, our results indicate that the relative importance of assembly processes for metacommunity organisation of aquatic insects in temporary ponds is contingent on dispersal ability.

Key words: assembly processes, dispersal ability, insects, metacommunity organisation, seasonal wetlands.

INTRODUCTION

Local communities are potentially connected by dispersal (metacommunity, *sensu* Leibold *et al.* 2004), and changes in their composition are driven by processes related to selection (biotic and abiotic), dispersal limitation and drift (Vellend 2016). Metacommunity ecology emerged as a framework that attempts to explain the relative importance of assembly processes (Leibold & Chase 2018). Leibold *et al.* (2004) originally proposed four different perspectives (species sorting, patch dynamics, mass effects and neutral) that integrated the role of the aforementioned processes under different dynamics to explain the spatial patterns of metacommunity organisation. Winegardner *et al.* (2012) argued that these perspectives could be viewed as scenarios that differently integrated the role of species sorting (the influence of the differences in local environmental factors (abiotic and/or biotic) in driving community composition) and dispersal-related processes. The latter processes cover scenarios of dispersal limitation

(where a subset of the species pool cannot reach all available patches due to increasing distances) or mass effects (communities spatially closer show similar composition due to homogenising effects of high dispersal; Cottenie 2005; Logue *et al.* 2011; Winegardner *et al.* 2012; Heino *et al.* 2015). The influence of dispersal processes is usually studied through proxies that include a variety of spatial factors (e.g. geographic distances, connectivity, spatial eigenvectors; Heino *et al.* 2015; Hill *et al.* 2017). Many authors have increasingly acknowledged that the relative importance of the assembly processes can vary spatially and temporally (Logue *et al.* 2011; Winegardner *et al.* 2012; Heino *et al.* 2015; Leibold & Chase 2018). Currently, many studies assess species distributions along spatial and environmental gradients to untangle the relative importance of environmental- and dispersal-related processes (Soininen 2014; Leibold & Chase 2018).

Metacommunity organisation is dependent on several contingencies (Soininen 2014; Heino *et al.* 2015; Leibold & Chase 2018). The observed pattern of metacommunity organisation can be affected by the scale of observation and varies according to groups of species with different dispersal abilities (Declerck

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Accepted for publication October 2020.

et al. 2011; De Bie *et al.* 2012; Heino *et al.* 2015). Organisms with higher dispersal ability could more easily access available patches that are geographically closer and thus be more prone to show the signature of mass effects (and more homogenous composition) at finer spatial scales, whereas organisms with reduced dispersal ability should be more limited by dispersal even at finer spatial scales (Van de Meutter *et al.* 2007; De Bie *et al.* 2012). Therefore, assessments that account for multiple scales of observation (such as spatial eigenvectors that capture different spatial scales) are necessary to more reliably assess the relative importance of assembly processes (Declerck *et al.* 2011; Heino *et al.* 2015).

Temporary ponds show a wide variation in local abiotic and biotic environmental factors that affect the establishment of the biota in these ecosystems (Williams 2006). In addition, temporary ponds represent patches that vary both spatially and temporally and thus constitute excellent models for testing the relative influence of assembly processes to metacommunity organisation (Jeffries *et al.* 2016). Many authors postulated that dispersal processes and dispersal ability should be important for the metacommunity organisation in these ecosystems (Jeffries 2005; De Bie *et al.* 2012; Jeffries *et al.* 2016). The relative importance of dispersal limitation processes is assumedly enhanced (over environmental ones) because their temporary condition should hinder part of the species pool from reaching all suitable patches (De Bie *et al.* 2012; Heino *et al.* 2015). While some studies did find predominance of spatial processes for some groups in wetlands (De Bie *et al.* 2012; Bertuzzi *et al.* 2019), other authors found higher importance of environmental processes to invertebrate metacommunity organisation in intermittent ponds (Florencio *et al.* 2011, 2014; Hill *et al.* 2017; Epele *et al.* 2019). This apparent inconsistency can be a result of the dynamic nature of intermittent ecosystems (Datry *et al.* 2016). That is, the relative importance of assembly processes in ponds can vary over their hydrological cycle (Boix & Batzer 2016; Datry *et al.* 2016).

Aquatic insects are the most diverse group of invertebrates in intermittent ponds (Batzer & Wissinger 1996; Batzer 2013). The influence of local environmental factors on the composition of pond insects has been demonstrated by many authors (see review in Batzer 2013). For instance, hydroperiod variation limits the establishment of desiccation-intolerant insects (Wellborn *et al.* 1996; Wissinger *et al.* 1999; Jeffries 2005). Other important factors include habitat structure (e.g. plant cover, canopy, pond depth) and water chemistry (e.g. conductivity, oxygen concentration, pH, water temperature, etc.) that can affect either the establishment or the development of the larval stages and eventually drive local

composition (Batzer & Wissinger 1996; Battle *et al.* 2001; Batzer *et al.* 2004; Stenert *et al.* 2008; Bischof *et al.* 2013). Taken together, these environmental predictors potentially contribute to structure insect communities according to species-sorting processes at finer scales.

As ectothermic organisms, many life-history aspects important to the local performance of insects in temporary ponds (desiccation tolerance, growth and emergence patterns, flight initiation) are dependent on climate (Boix *et al.* 2011; Kingsolver *et al.* 2011). Thus, climate gradients are key drivers of the spatial distribution of wetland insects (Boix & Batzer 2016). The composition of wetland insect communities was indeed found to vary along climate gradients (Batzer *et al.* 2005; Ruhí *et al.* 2013, 2014; Pires *et al.* 2018; Epele *et al.* 2019). Climate features (e.g. mean and seasonal precipitation and temperature) are also very important to the hydrology of temporary ponds (Jackson *et al.* 2014). Despite the difficulty to detect spatially independent effects of climate (on account of the spatial structuration of climate at coarser scales; Dodds *et al.* 2019), it could be viewed as an additional filter of insect distribution by affecting either wetland hydrology or trait composition. As a result, climate gradients (such as temperature and precipitation) can be expected to lead to spatial patterns of insect distribution that match the species-sorting perspective.

Although insects are generally considered good dispersers because of their ability to fly, several differences occur among insect taxa regarding flight (and consequently, dispersal) ability (Bilton *et al.* 2001). Many taxa of Odonata, Hemiptera and Coleoptera are strong aerial dispersers with active flight, capable of long-distance dispersal, while taxa of Diptera, Ephemeroptera, Trichoptera fly for shorter distances and depend more heavily on wind for dispersal (Bilton *et al.* 2001; Kelly *et al.* 2001; Petersen *et al.* 2004) and are usually considered as taxa with weaker flight abilities. These differences should affect the metacommunity organisation of insects with varying dispersal-related traits. In fact, Heino (2013) detected distinct signatures of environmental and spatial processes on the community structure of strong- and weak-flying lake insects. Similar evidence was detected by Patrick *et al.* (2014) in floodplains. However, to our knowledge, there have been no studies on the relative importance of environmental and spatial processes focusing the different dispersal abilities among active dispersers such as weak- and strong-flying insects in temporary ponds. Understanding the metacommunity organisation of pond insect communities can improve the knowledge on the processes that influence insect establishment in these ecosystems. Such knowledge can be useful to many applied and conservation issues in pond

management because aquatic insects play important roles in the ecological functioning of ponds, for example, secondary production and food source of waterfowl (Batzer & Wissinger 1996; Boix & Batzer 2016).

In this study, we assessed the relative importance of spatial and environmental (climate, habitat structure and water chemistry) factors to aquatic insect composition in temporary ponds ranging along a latitudinal gradient in southern Brazil. We further assessed the patterns for subsets of insects that assumedly differ as to their dispersal ability (i.e., ‘weak-’ and ‘strong-flying’ insects). Assuming that insects with active and strong flight ability are capable of flying for longer distances and of more easily accessing patches farther apart in comparison with insects that more heavily rely on passive flight associated with climate for dispersal, we hypothesised that the metacommunity organisation would differ between strong- and weak-flying insects (Patrick *et al.* 2014; Heino *et al.* 2015) and according to scale of observation. We expected that the composition of weak-flying insects would be more strongly affected

by broadscale spatial factors and species sorting associated with climate, while the composition of strong-flying insects would be more strongly affected by species sorting coupled to local environmental factors.

METHODS

Study region

The samplings were conducted in the southern Brazilian Coastal Plain, a lowland region (altitudes below 20 m a.s.l.) with sandy geomorphology (Villwock & Tomazelli 2006) running along ~640 km of coastline (~80 km wide) of the states of Rio Grande do Sul and Santa Catarina (Fig. 1). Along its extension, the climate in the Coastal Plain ranges from subhumid subtropical in the north to humid temperate subtropical in the south (Maluf 2000). The annual rainfall ranges from 1000 to 1500 mm and the average temperature from 12 in the coldest season to 22°C in the warmest seasons of the year (Villwock & Tomazelli 2006). The original vegetation types in the region include evergreen (ombrophilous) forests occurring in the northern region (state of Santa Catarina) and coastal *restinga* (scrub-like vegetation and white-sand forests) occurring in the

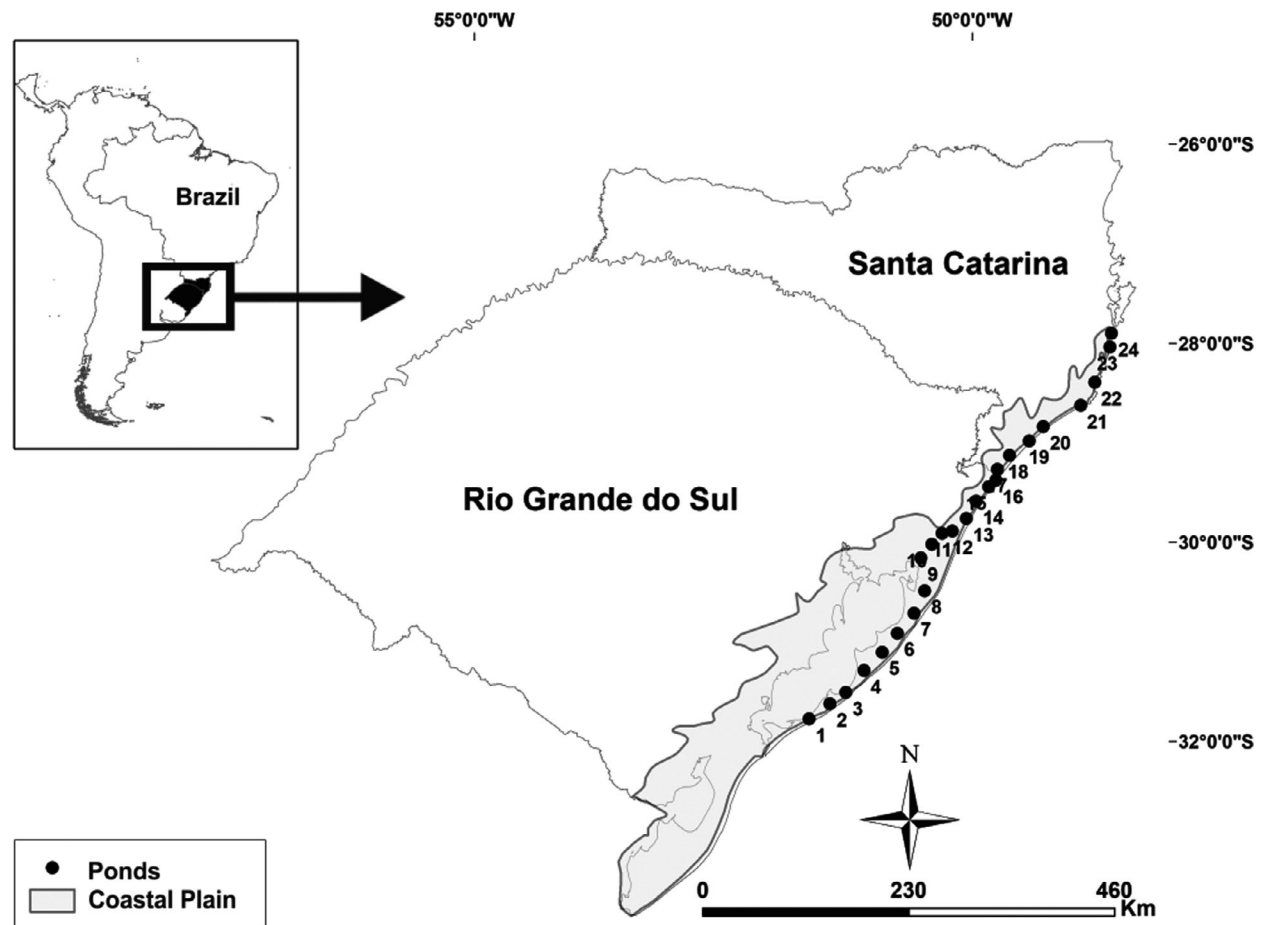


Fig. 1. Location of the sampled ponds across the study region.

south (for most of the state of Rio Grande do Sul; Leão *et al.* 2014). The landscape of the Coastal Plain is also marked by the frequent occurrence of small temporary wetland ecosystems (Maltchik *et al.* 2003).

Sampling design and collection procedures

Twenty-four intermittent ponds distributed along a latitudinal range (S 27°–31°) covering ~530 km in the study region were selected to be sampled (Fig. 1). The ponds were selected according to two major criteria: (i) original condition (i.e., man-made farm ponds were not selected for sampling) and (ii) low anthropic impact in their surroundings. Specifically, the ponds were located in nonheavily urbanised areas (at least 1 km away from the large and common urban concentrations in the region). Most of the ponds were located in farmlands with extensive livestock (animal farming system with low herd number and without overgrazing). The minimum geographic distance between the closest ponds was 10.1 km. All ponds had maximum flooded area of 1 ha and habitat types characterised by emergent and/or floating vegetation (Pires *et al.* 2018). The occurrence of potential predators (e.g. fish and amphibians) was recorded in all ponds (Pires *et al.* 2018; Knauth *et al.* 2019).

Insect collection was carried out in the early spring of 2015 (from September to October; austral season), during the wet phase of the ponds (Bertuzzi *et al.* 2019; Knauth *et al.* 2019). This is also the season that is known to match the peak in the abundance and richness of many aquatic invertebrate taxa in the region (Lima *et al.* 2013). Specimens were collected with a frame dip-net (30-cm wide; 250- μ m mesh size). Four subsamples were taken at each pond. Each subsample consisted of three 1-m sweeps in the water performed after kicking the substrate of the littoral zone of the ponds. The collected material was preserved *in situ* with 10% formalin, and the subsamples were posteriorly pooled into a single sample. In the laboratory, the specimens were collected under stereomicroscope and fixed in 80% ethanol. Specimens were determined to the lowest taxonomic level (genus level whenever possible, except for Chironomidae) using specialised literature (e.g. Fernández & Domínguez 2001).

Explanatory datasets

Environmental factors

We assembled information from three types of environmental predictors to be employed as proxies of environmental assembly processes in this study: water chemistry, habitat structure (pond-level) and climatic characteristics of the study region. They are hereafter referred to as 'environmental' dataset, described as follows. The water chemistry predictors included the following: dissolved oxygen (DO; mg/L), oxygen saturation (%), electrical conductivity (EC; μ S/cm); pH, total dissolved solids (TDS; mg/L), turbidity (NTU) and water temperature (WT; °C). These variables were measured *in situ* with a multiparameter probe (U-50; Horiba Tokyo, Japan). The habitat structure predictors

included the following: canopy cover in the perimeter (up to ~50 m) of each pond (Pires *et al.* 2018); pond area (ha), calculated with a GPS (Garmin) after walking the perimeter of the surface flooded area of each pond; and mean water depth, measured with a gauge in ten different points of each pond.

The climatic characteristics of the study region were represented by the set of bioclimatic variables available at the WorldClim database (Hijmans *et al.* 2005). This database consists of a set of 19 global climate layers derived from temperature and precipitation. The raw values of each variable were downloaded from the database at the spatial resolution of ~1 km² and obtained for the corresponding geographic location of each pond (lat-long, decimal degrees). Instead of the full climatic dataset, we explored a subset of seven variables that we considered of particular importance to aquatic insects (i.e., with biological relevance in terms of their physiology) and also to the hydroperiod of seasonal ponds. In this regard, we considered the following: annual mean temperature, mean diurnal temperature range, isothermality, temperature seasonality, temperature annual range, annual precipitation and precipitation seasonality. Variables related to seasonal patterns in temperature were chosen because of their general association with the spatial distribution of aquatic insects and wetland invertebrates (Batzer *et al.* 2005; Bonada *et al.* 2007; Ruhí *et al.* 2013; Epele *et al.* 2019). Annual and seasonal patterns of precipitation are of high importance to the hydrology of seasonal ecosystems, with important effects on the community structure of pond invertebrates (Batzer 2013). The original values of the environmental predictors are given in Table S1.

Spatial factors

As spatial factors, we used distance-based Moran's eigenvector Maps (db-MEM) derived from spatial eigenfunction analysis (Borcard *et al.* 2004; Griffith & Peres-Neto 2006; Legendre & Legendre 2012) to represent the geographic relationships among the studied ponds and assess spatial processes. The db-MEM method is based on a pairwise distance matrix containing Euclidean distances between the studied ponds (derived from their geographic coordinates). This distance matrix is then truncated at a given threshold that connects all sites and subject to an ordination based on principal coordinate analysis (PCoA). As criterion for truncation of the distance matrix, we used the longest distance connecting each pair of ponds in a minimum spanning tree (43.5 km). This procedure yielded nine positive db-MEM eigenvectors by the eigenfunction analysis, which constituted the explanatory matrix of spatial factors. After visual inspections of the one-dimensional line plots of each vector, we arbitrarily defined the first three eigenvectors (db-MEM 1–3) as proxies of 'broad-scale' geographic variation in the study region, db-MEM 4–6 as 'medium-scale' and db-MEM 7–9 as 'fine-scale' variation.

Response datasets

We used as response dataset the presence–absence matrix of insect taxa rather than the semi-quantitative data. The site-by-taxon matrix was Hellinger-transformed prior to the

major procedures (Legendre & Gallagher 2001). For the assignment of each taxon as ‘strong-’ or ‘weak-flying’, we consulted both general (Bilton *et al.* 2001; Kelly *et al.* 2001) and specific databases of biological traits of freshwater invertebrate taxa (Poff *et al.* 2006; Vieira *et al.* 2006). As these sources were originally developed for lotic invertebrates, we rechecked the classification of studies that used the same approach in lentic ecosystems (Heino 2013; Patrick *et al.* 2014). When information for specific taxa was unavailable, we assign these taxa to the same category of the closest taxonomic unit (genera from the same family, or family from the same order). In brief, the subset of strong-flying insects included families of Anisoptera (Odonata) and most families of Coleoptera (Hydrophilidae, Dytiscidae) and Hemiptera, while weak-flying insects comprised all families of Diptera (including Chironomidae), families of Zygoptera (Odonata), Ephemeroptera, Lepidoptera, Trichoptera and some families of Coleoptera (e.g. Elmidae and Psephenidae) and Hemiptera (Pleidae). The category to which each taxon was assigned to is shown in Table S2.

Data analysis

Before the major procedures, the environmental predictors of continuous nature (bioclimatic and water chemistry) were standardised (zero-scaled mean and unit variance), while the habitat structure predictors were arcsin-transformed. On account of the large number of predictors (higher than sampling units) and multicollinearity within the environmental dataset, we used variance inflation factors (VIF; Legendre & Legendre 2012) to remove redundant variables from the environmental dataset. We established a threshold value of 5 and sequentially removed variables until all remaining ones had VIF below this value. This procedure resulted in the retaining of annual mean temperature, mean diurnal range, isothermality temperature seasonality, temperature annual range, annual precipitation, dissolved oxygen, oxygen saturation, pH, pond area and water temperature, which constituted the environmental dataset in the major procedures.

The influence of environmental and spatial factors on aquatic insect composition was assessed with redundancy analysis (RDA; Legendre & Legendre 2012). Prior to the final model, we separately tested the significance of the association of the composition datasets (total insect community, ‘weak-’ and ‘strong-flying’ insects) with the full set of predictors within each explanatory dataset (environmental and spatial factors, or global models). Only if the global model was significant ($P < 0.05$), we proceeded to additional searches for subsets of best-fit variables within each explanatory dataset (spatial and environmental factors). This additional search was carried out using forward selection procedures (Blanchet *et al.* 2008). The forward selection procedures considered two stopping criteria for selection of variables: (i) a significance value no higher than 0.05 (after 999 permutations); and (ii) explained variance ($R^2_{\text{adj.}}$ values) by the preselected variables no higher than the model containing all variables within each explanatory dataset.

We built final RDA models with the variables retained by the forward selection procedures. We used the variation

partitioning procedures to estimate the pure and shared amounts of variance in community composition explained by environmental and spatial factors (Legendre & Legendre 2012). We used a permutation-based approach (Monte Carlo test; 999 permutations) to assess the significance of the final models as well as of the pure environmental and spatial fractions (Peres-Neto *et al.* 2006). To further dissect the spatial variation in the study region, we first ran ordinary least-square (OLS) multiple linear regression models between the selected db-MEM (response variable) and environmental predictors (explanatory variable; linearly detrended values) to investigate whether the spatial variation was related to a specific environmental predictor. We repeated the abovementioned procedures for the datasets of the total insect community and subsets of weak- and strong-flying insects. All analyses were run in the statistical environment R version 3.5.3 (R Core Team 2019), using the functions *rda* and *varpart* from the *vegan* v.2.5-6 (Oksanen *et al.* 2019) and functions *dbmem* and *forward.sel* from the *adespatial* package v.0.3-8 (Dray *et al.* 2020). The full workflow of the undertaken analytical procedures is given in Fig. S1.

RESULTS

We collected 32998 specimens from 66 taxa (57 genera and nine additional taxa identified to family level) in the region. Chironomidae (Diptera) was the dominant taxon (46.7% of the total abundance), followed by *Callibaetis* (Navás 1924) (Ephemeroptera, Baetidae; 16.9%). Coleoptera was the richest order (31 taxa), followed by Odonata and Diptera (13 taxa each; Table S2).

The global environmental and spatial models were significantly associated with all composition data sets (total insect community; subsets of weak- and strong-flying insects; $P < 0.05$). Broad- and fine-scale spatial predictors (db-MEM3 and 7, respectively), oxygen saturation, annual temperature and isothermality explained 13% of the composition of the total insect community. The pure environmental fraction accounted for most part of this variation (7%; $P = 0.005$), followed by the pure fine-scale fraction (~4%; $P = 0.016$). The pure broadscale spatial fraction was not significant ($P > 0.05$) (Table 1; Fig. 2 a).

For the subset of strong-flying insects, broad- and fine-scale spatial predictors (db-MEM3 and 7, respectively), along with isothermality, explained ~9% of the variation in their composition. The relative contributions of the spatial and environmental fractions were very similar to the pattern observed in the total community (Table 1; Fig. 2b). For the subset of weak-flying insects, medium-scale spatial predictors (db-MEM5), water temperature and isothermality 9% explained of the variation in composition. The pure environmental fraction accounted for most part of this variation (5%; $P = 0.005$),

Table 1. Output of the global and final (reduced) RDA models for the influence of environmental and spatial factors on the community structure of aquatic insect communities (and subsets of weak- and strong-flying insects) in the studied ponds

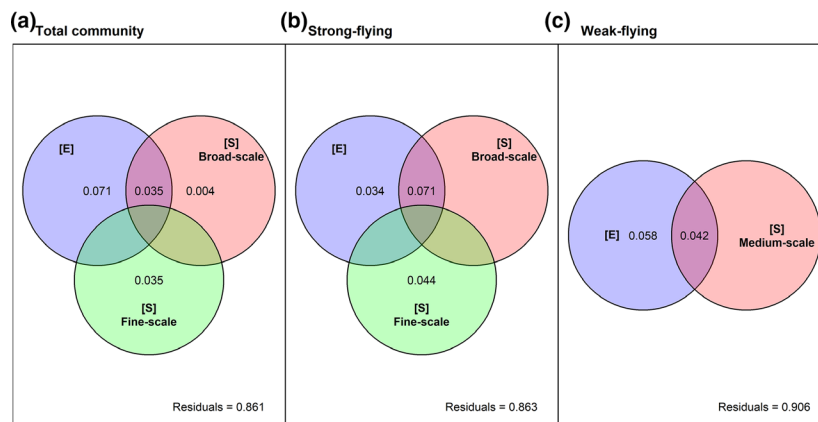
	Spatial <i>P</i> [global]	Broadscale		Medium scale		Fine scale		Environmental				
		Selected predictor	$R^2_{adj.}$	Selected predictor	$R^2_{adj.}$	Selected predictor	$R^2_{adj.}$	<i>P</i> [global]	Selected predictor	$R^2_{adj.}$	$R^2_{adj.}$ [E-S]	[<i>d</i>]
Total community	**	db-MEM3	0.004	–	–	db-MEM7	0.035*	**	DO_Sat; Bio1; Bio3	0.071**	0.035	0.862
Strong-flying	**	db-MEM3	<0	–	–	db-MEM7	0.044**	*	Bio1	<0	0.071	0.88
Weak-flying	*	–	–	db-MEM5	<0	–	–	*	WT; Bio3	0.058**	0.042	0.91

Bio1 = annual temperature; Bio3 = isothermality; db-MEM = distance-based Moran eigenvector map; DO_Sat = oxygen saturation (%); WT = water temperature (°C).

$R^2_{adj.}$ [E] and [S] variance explained by the environmental and spatial fractions of reduced model, respectively; $R^2_{adj.}$ [E-S] variance explained by the spatially structured environmental fraction of the model; [*d*] unexplained variance.

* $P < 0.05$.

** $P < 0.01$.

**Fig. 2.** Venn diagrams showing the fractions of explained variation of aquatic insect composition by environmental and spatial factors. (a) Total insect community; (b) subset of strong-flying insects; (c) subset of weak-flying insects; [e] environmental fraction of the model. For the specific environmental predictors selected in each model, see Table 1.

followed by the spatially structured environmental variation (~4%). The pure spatial fraction had no significant contribution ($P > 0.05$; Table 1; Fig. 2c). The OLS linear regressions detected a significant relationship between annual temperature and isothermality with the medium-scale spatial predictor ($P < 0.001$). The remaining environmental predictors showed no spatial dependence (Table S3).

DISCUSSION

Environmental (climate and water chemistry) and fine-scale spatial factors influenced the composition of aquatic insects. Our results thus evidenced that species sorting and dispersal-related processes jointly assembled the metacommunity organisation of aquatic insects in the temporary ponds studied. The pure

environmental fraction accounted for a larger portion of the explained variation in the composition of the total insect community compared to the spatial fraction; however, the relative contribution of environmental and scale-specific spatial factors differed between strong- and weak-dispersing insects. Fine-scale spatial factors and spatially structured climate influenced the composition of strong-flying insects, while water chemistry and spatially structured climate influenced the composition of weak-flying insects. This suggests that species sorting (associated with local pond characteristics and regional climate) and fine-scale dispersal processes jointly structured the metacommunity organisation of aquatic insects in the study region, although the relative roles of these assembly processes differed between insects with different dispersal abilities, in accordance with our general hypothesis.

The pure environmental fraction accounted for most part of the variation in insect composition, suggesting a predominant role of species-sorting processes. At first, this result contrasts with the postulation that dispersal limitation processes predominate to explain the metacommunity organisation in temporary ponds (De Bie *et al.* 2012; Heino *et al.* 2015). These authors showed that spatial factors explained most part of the community structure of some taxa in ponds (e.g. anurans, submerged macrophytes and zooplankton; Declerck *et al.* 2011; De Bie *et al.* 2012; Trindade *et al.* 2018; Bertuzzi *et al.* 2019; Knauth *et al.* 2019). However, the relative importance of spatial and environmental factors may vary according to the focal organism (Soininen 2014; Heino *et al.* 2015), and insects can be a remarkable exception to this expected pattern. Under a metacommunity framework, the congruence between species composition and environmental conditions is expected to be maximised at intermediate dispersal rates, which allow the species to track their optimal requirements and lead to the predominance of species-sorting processes (Heino *et al.* 2015; Leibold & Chase 2018). In addition, broader scales of observation can increase the probability of detecting the effects of dispersal limitation (Heino *et al.* 2015). However, we did not detect pure effects of broad- and medium-scale spatial factors to insect composition, suggesting that insect communities were not dispersal limited in this study (Heino *et al.* 2015). Many wetland insect taxa have high mobility and dispersal ability, which favour their adaptation to temporary ecosystems (Williams 1996). For instance, the adults of many generalist insect taxa such as Coleoptera, Hemiptera and Odonata migrate in dry periods and oviposit once ponds reflow (Batzer & Wissinger 1996; Williams 1997; Bilton *et al.* 2001). In this sense, the aerial dispersal stage and the corresponding ability to cyclically recolonise allow their continuous occurrence in wetlands (Batzer & Wissinger 1996; Wissinger 1997; Boix & Batzer 2016), which could allow insects to select more suitable sites. In fact, many authors detected exclusive (or higher) effect of environmental over spatial factors to insect composition in temporary ponds (Van de Meutter *et al.* 2007; De Bie *et al.* 2012). Other studies of invertebrate communities also described a higher influence of environmental factors to the composition of active-dispersing invertebrates (mostly insects) in ponds (Florenco *et al.* 2011, 2014; Hill *et al.* 2017; Epele *et al.* 2019). Our results followed these findings, as most part of the variation in insect composition in this study was explained by the pure environmental fraction.

Another circumstance that likely explains the predominance of environmental processes is the specific

phase of the hydrological cycle of the studied ponds. Recent reviews on the metacommunity organisation of temporary ecosystems stressed that assembly processes vary over time (Datry *et al.* 2016). These authors stated that dispersal-related processes dominate in the colonisation phase; after most colonists establish, environmental processes would dominate. In intermittent ponds, it is implied that dispersal-related processes dominate in the early wet phase, and environmental ones, in the mid- and late-wet phases. As our design matched the last phase (Bertuzzi *et al.* 2019; Knauth *et al.* 2019), we covered the phase that was most likely structured by environmental (species sorting) processes.

In this study, climate (annual mean temperature and isothermality) and water chemistry (oxygen) accounted for the variation in aquatic insect composition. Temperature is an important driver of many aspects of the life history of aquatic insects, such as development and flight initiation (Batzer & Wissinger 1996; Williams 1996; Boix *et al.* 2011), and thus gradients in temperature can constrain the distribution aquatic insects (Bonada *et al.* 2007; Boix & Batzer 2016). In fact, climate gradients were associated with the spatial distribution of wetland insects (Batzer *et al.* 2005; Ruhí *et al.* 2013, 2014; Pires *et al.* 2018). Nevertheless, the contribution of climate was spatially structured. The distribution of climate is associated with geography (Dodds *et al.* 2019), and this is likely a contingency of the sampling design of this study, which targeted a study region (southern Brazilian Coastal Plain) that covers a latitudinal gradient of climatic conditions (Villwock & Tomazelli 2006). As for the role of water chemistry, a wide number of studies detected significant association of specific water physicochemical variables with insect composition in ponds, including oxygen content (Batzer & Wissinger 1996; Batzer *et al.* 2004; Stenert *et al.* 2008; Hill *et al.* 2017). Oxygen can be a limiting factor for the development of wetland insects, although the concentration varies along the hydrological cycle and its exact effect is usually observed in short-term temporal range (Batzer & Wissinger 1996; Battle & Golladay 2001). Because this study consisted of a single collection, we might have matched the period of changing oxygen along the specific phase of the hydrological cycle of some of the wetlands.

The relative contribution of environmental and spatial factors differed between strong- and weak-dispersing insects, suggesting that the metacommunity organisation of each subset of taxa was differently affected by species-sorting and dispersal-related assembly processes. This result is broadly in accordance with our general hypothesis, although our expectation was not fully corroborated. It has been increasingly recognised that the relative importance

of assembly processes related to dispersal differs between taxa with high and low dispersal ability (Van de Meutter *et al.* 2007; De Bie *et al.* 2012; Heino 2013). Under a metacommunity framework, stronger dispersers are expected to show more homogenous composition at finer scales due to mass effects compared to taxa with more restricted dispersal ability (Ng *et al.* 2009; Logue *et al.* 2011; Leibold & Chase 2018). According to the framework provided by Heino *et al.* (2015), the pure influence of fine-scale spatial factors on community composition is an indication of the role of mass effects. Aquatic insects that inhabit temporary ponds differ with respect to life-history traits that affect the efficiency of dispersal by flight. For instance, while many taxa of Coleoptera, Hemiptera and Odonata have adult stages with strong flight ability that allows long-distance, overland dispersal (Williams 1996; Batzer & Wissinger 1996), individuals of Diptera, Ephemeroptera and Trichoptera generally have smaller body sizes, lower flight ability and strongly rely on weather conditions (wind, sunlight and temperature) for dispersal at longer distances (Bilton *et al.* 2001). Our results are broadly in line with the findings of Van de Meutter *et al.* (2007) and Patrick *et al.* (2014), which detected higher similarity in the composition of active- and good-dispersing taxa (i.e., including insects with active strong flight such as Coleoptera and Hemiptera) in ponds. In specific, individuals from many families of strong-flying insects such as Coleoptera and Hemiptera are known to be the earliest colonisers of temporary ponds and effectively establish in these sites via cyclic colonisation, irrespective of their specific ability to tolerate dry periods (Boix & Batzer, 2016). The fine-scale spatial factors were the most important drivers of the composition of the strong-flying insects in this study suggesting that the group with higher dispersal abilities showed mass effects.

Our procedures managed to explain a limited fraction of the variation in insect composition (~10%). Recent reviews on the ecology of aquatic invertebrates in wetland ecosystems (Batzer 2013; Boix & Batzer 2016; Jeffries *et al.* 2016) highlight the complex challenge of explaining assembly processes of this fauna. These reviews state the role of processes that operate at much finer scales (e.g. varying oviposition cues and flight behaviour among populations from the same species), which eventually result in unpredictable responses of insect communities to the environmental and spatial predictors usually included in community ecology studies (Batzer 2013). Another contingency is the seasonal variation in the phenology of many species (Jeffries *et al.* 2016), which is not captured in 'snapshot' studies such as ours. Therefore, assessments that include a more complete fraction of the hydrological

cycle can likely increase the amount of explained variation.

CONCLUSIONS

Environmental (regional patterns in temperature and water chemistry) had higher influence than spatial factors on aquatic insect composition in this study, indicating that species-sorting processes associated with regional climate and local water chemistry predominated over dispersal-related processes in the metacommunity organisation. The predominance of species sorting and the lack of significant effects of broadscale spatial factors suggest little evidence of dispersal limitation in the assembly of this fauna (at least in the geographic scale-covered). However, the relative contributions of environmental and spatial factors differed between strong- and weak-flying insects, indicating that the relative importance of assembly processes was contingent on dispersal ability. Strong-flying insects likely showed mass effects at fine scales, while weak-flying insects were more influenced by species sorting at the site level (water chemistry). Overall, our results suggest a complex interplay of assembly processes to the metacommunity organisation of aquatic insects in the temporary ponds studied and highlights the necessity to account for differences in dispersal ability in this assessment.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Roberta Cozer Bacca: Data curation (lead); Investigation (lead); Methodology (lead); Writing-original draft (supporting). **Mateus Marques Pires:** Conceptualization (equal); Data curation (supporting); Formal analysis (supporting); Investigation (lead); Validation (equal); Visualization (equal); Writing-original draft (supporting); Writing-review & editing (lead). **Leonardo Felipe Bairos Moreira:** Resources (supporting); Writing-review & editing (supporting). **Cristina Stenert:** Conceptualization (lead); Funding acquisition (supporting); Methodology (supporting); Project administration (lead); Resources (lead); Writing-review & editing (equal). **Leonardo Maltchik:** Conceptualization (lead); Funding acquisition (lead); Project administration (lead); Supervision (lead); Writing-review & editing (supporting).

RESEARCH ETHICS

We declare that the data collection complied with current Brazilian environmental laws (SISBIO – n. 36365-2).

DATA AVAILABILITY STATEMENT

Specimens are stored in the collection of the Laboratory of Ecology and Conservation of Aquatic Ecosystems from UNISINOS.

REFERENCES

- Battle J., Stephen W. & Golladay S. W. (2001) Water quality and macroinvertebrate assemblages in three types of seasonally inundated limesink wetlands in southwest Georgia. *J. Freshw. Ecol.* **16**, 189–207.
- Battle, J. & Golladay, S. W. (2001) Water Quality and Macroinvertebrate Assemblages in Three Types of Seasonally Inundated Limesink Wetlands in Southwest Georgia. *J. Freshw. Ecol.* **16**, 189–207.
- Batzer D. P. (2013) The seemingly intractable ecological responses of invertebrates in North American wetlands: a review. *Wetlands* **33**, 1–15.
- Batzer D. P., Dietz-Brantley S. E., Taylor B. E. & DeBiase A. E. (2005) Evaluating regional differences in macroinvertebrate communities from forested depressional wetlands across eastern and central North America. *J. N. Am. Benthol. Soc.* **24**, 403–14.
- Batzer D. P., Palik B. J. & Buech R. (2004) Relationships between environmental characteristics and macroinvertebrate communities in seasonal woodland ponds of Minnesota. *J. N. Am. Benthol. Soc.* **23**, 50–68.
- Batzer D. P. & Wissinger S. A. (1996) Ecology of insect communities in nontidal wetlands. *Annu. Rev. Entomol.* **41**, 75–100.
- Bertuzzi T., Marques Pires M. & Maltchik L. (2019) Drivers of the beta diversity of aquatic plant communities along a latitudinal gradient in southern Brazilian coastal ponds. *J. Veg. Sci.* **30**, 281–90.
- Bilton D. T., Freeland J. R. & Okamura B. (2001) Dispersal in freshwater invertebrates. *Annu. Rev. Ecol. Syst.* **32**, 159–81.
- Bischof M. M., Hanson M. A., Fulton M. R., Kolka R. K., Sebestyen S. D. & Butler M. G. (2013) Invertebrate community patterns in seasonal ponds in Minnesota, USA: Response to hydrologic and environmental variability. *Wetlands* **33**, 245–56.
- Blanchet G., Legendre P. & Borcard D. (2008) Forward selection of spatial explanatory variables. *Ecology* **89**, 2623–32.
- Boix D. & Batzer D. (2016) Invertebrate assemblages and their ecological controls across the world's freshwater wetlands. In: *Invertebrates in Freshwater Wetlands: An International Perspective on their Ecology* (eds D. Boix & D. Batzer) pp. 601–40. Springer, Amsterdam.
- Boix, D., Magnusson, A. K., Gascón, S., Sala, J. & Williams, D. D. (2011) Environmental influence on flight activity and arrival patterns of aerial colonizers of temporary ponds. *Wetlands*, **31**, 1227–40.
- Bonada N., Doledec S. & Statzner B. (2007) Taxonomic and biological trait differences of stream macroinvertebrate communities between Mediterranean and temperate regions: implications for future climatic scenarios. *Glob. Change Biol.* **13**, 1658–71.
- Borcard D., Legendre P., Avois-Jacquet C. & Tuomisto H. (2004) Dissecting the spatial structure of ecological data at multiple scales. *Ecology* **85**, 1826–32.
- Cottenie K. (2005) Integrating environmental and spatial processes in ecological community dynamics. *Ecol. Lett.* **8**, 1175–82.
- Datry T., Bonada N. & Heino J. (2016) Towards understanding the organisation of metacommunities in highly dynamic ecological systems. *Oikos* **125**, 149–59.
- De Bie T., De Meester L., Brendonck L. *et al.* (2012) Body size and dispersal mode as key traits determining metacommunity structure of aquatic organisms. *Ecol. Lett.* **15**, 740–47.
- Declerck S. A. J., Coronel J. S., Legendre P. & Brendonck L. (2011) Scale dependency of processes structuring metacommunities of cladocerans in temporary pools of high Andes wetlands. *Ecography* **34**, 296–305.
- Dodds W. K., Batzer D. P., Schechner A. *et al.* (2019) The freshwater biome gradient framework: predicting macroscale properties based on latitude, altitude, and precipitation. *Ecosphere* **10**, e02786.
- Dray S., Bauman D., Blanchet G. *et al.* (2020) Package ‘adespatial: multivariate multiscale spatial analysis’. R package version 0.3-8.
- Epele L., Brand C. & Miserendino M. L. (2019) Ecological drivers of alpha and beta diversity of freshwater invertebrates in arid and semiarid Patagonia (Argentina). *Sci. Total Environ.* **678**, 62–73.
- Fernández H. & Domínguez E. (2001) *Guía para la determinación de los artrópodos bentónicos sudamericanos*. Universidad Nacional de Tucumán, Facultad de Ciencias Naturales, Instituto M. Lillo, Tucuman.
- Florencio M., Díaz-Paniagua C., Gómez-Rodríguez C. & Serrano L. (2014) Biodiversity patterns in a macroinvertebrate community of a temporary pond network. *Insect Conserv. Divers.* **7**, 4–21.
- Florencio M., Díaz-Paniagua C., Serrano L. & Bilton D. T. (2011) Spatio-temporal nested patterns in macroinvertebrate assemblages across a pond network with a wide hydroperiod range. *Oecologia* **166**, 469–83.
- Griffith D. A. & Peres-Neto P. R. (2006) Spatial modelling in ecology: the flexibility of eigenfunction spatial analyses. *Ecology* **87**, 2603–13.
- Heino J. (2013) Does dispersal ability affect the relative importance of environmental control and spatial structuring of littoral macroinvertebrate communities? *Oecologia* **171**, 971–80.
- Heino J., Melo A. S., Siqueira T., Soininen J., Valanko S. & Bini L. M. (2015) Metacommunity organisation, spatial extent and dispersal in aquatic systems: patterns, processes and prospects. *Freshw. Biol.* **60**, 845–69.
- Hijmans R. J., Cameron S. E., Parra J. L., Jones P. G. & Jarvis A. (2005) Very high resolution interpolated climate surfaces for global land areas. *Int. J. Climatol.* **25**, 1965–78.
- Hill M. J., Heino J., Thornhill I., Ryves D. B. & Wood P. J. (2017) Effects of dispersal mode on the environmental and spatial correlates of nestedness and species turnover in pond communities. *Oikos* **126**, 1575–85.

- Jackson C. R., Thompson J. A. & Kolka R. K. (2014) Wetland soils, hydrology, and geomorphology. In: *Ecology of Freshwater and Estuarine Wetlands* (eds D. P. Batzer & R. R. Sharitz) pp. 23–60. University of California Press, Berkeley.
- Jeffries M. (2005) Local-scale turnover of pond insects: intra-habitat quality and inter-pond geometry are both important. *Hydrobiologia* **543**, 207–20.
- Jeffries M. J., Epele L. B., Studinski J. M. & Vad C. F. (2016) Invertebrates in temporary wetland ponds of the temperate biomes. In: *Invertebrates in Freshwater Wetlands: An International Perspective on their Ecology* (eds D. Boix & D. Batzer) pp. 105–40. Springer, Amsterdam.
- Kelly L. C., Bilton D. T. & Rundle S. D. (2001) Genetic differentiation and dispersal in the Canary Island caddisfly *Mesophylax aspersus* (Trichoptera: Limnephilidae). *Heredity* **86**, 370–77.
- Kingsolver J. G., Woods H. A., Buckley L. B., Potter K. A., MacLean H. J. & Higgins J. K. (2011) Complex life cycles and the responses of insects to climate change. *Integr. Comp. Biol.* **51**, 719–32.
- Knauth D. S., Pires M. M., Stenert C. & Maltchik L. (2019) Disentangling the role of niche-based and spatial processes on anuran beta diversity in temporary ponds along a forest–grassland transition. *Aquat. Sci.* **81**, 63.
- Leão T. C., Fonseca C. R., Peres C. A. & Tabarelli M. (2014) Predicting extinction risk of Brazilian Atlantic Forest angiosperms. *Conserv. Biol.* **28**, 1349–59.
- Legendre P. & Gallagher E. D. (2001) Ecologically meaningful transformations for ordination of species data. *Oecologia* **129**, 271–80.
- Legendre P. & Legendre L. F. J. (2012) *Numerical Ecology*. Elsevier, Amsterdam.
- Leibold M. A. & Chase J. (2018) Metacommunity Patterns in Space. In: *Metacommunity Ecology*, Vol. **59** (eds M. A. Leibold & J. Chase) pp. 90–130. Princeton University Press, Princeton.
- Leibold M. A., Holyoak M., Mouquet N. *et al.* (2004) The metacommunity concept: a framework for multi-scale community ecology. *Ecol. Lett.* **7**, 601–13.
- Lima F. B., Schäfer A. E. & Lanzer R. M. (2013) Diversity and spatial and temporal variation of benthic macroinvertebrates with respect to the trophic state of Lake Figueira in the South of Brazil. *Acta Limnol. Bras.* **25**, 429–41.
- Logue J. B., Mouquet N., Peter H., Hillebrand H. & The Metacommunity Working Group (2011) Empirical approaches to metacommunities: a review and comparison with theory. *Trends Ecol. Evol.* **26**, 482–91.
- Maltchik, L., Schneider, E., Becker, G. & Escobar, A. (2003) Inventory of wetlands of Rio Grande do Sul (Brazil). *Pesqui. Botânica*
- Maluf, J. R. T. (2000) A new climatic classification for the state of Rio Grande do Sul, Brazil. *Rev. Bras. Agrometeorol.* **8**, 141–50.
- Ng I. S. Y., Carr C. & Cottenie K. (2009) Hierarchical zooplankton metacommunities: distinguishing between high and limiting dispersal mechanisms. *Hydrobiologia* **619**, 133–43.
- Oksanen J., Blanchet F. G., Friendly M. *et al.* (2019) Package ‘vegan: Community Ecology Package’. R package version 2.5-6.
- Patrick C., Cooper M. & Uzarski D. (2014) Dispersal mode and ability affect the spatial turnover of a wetland macroinvertebrate metacommunity. *Wetlands* **34**, 1133–43.
- Peres-Neto P. R., Legendre P., Dray S. & Borcard D. (2006) Variation partitioning of species data matrices: estimation and comparison of fractions. *Ecology* **87**, 2614–25.
- Petersen I., Masters Z., Hildrew A. G. & Ormerod S. J. (2004) Dispersal of adult aquatic insects in catchments of differing land use. *J. Appl. Ecol.* **41**, 934–50.
- Pires M. M., Stenert C. & Maltchik L. (2018) Drivers of beta diversity of Odonata along a forest–grassland transition in southern Brazilian coastal ponds. *Freshw. Sci.* **37**, 357–66.
- Poff, N. L., Olden, J. D., Vieira, N. K. M., Finn, D. S., Simmons, M. P. & Kondratieff, B. C. (2006) Poffetal_2006_FunctionalTraitNiche_JNABS insects. *J. North Am. Benthol. Soc.* **25**, 730–55.
- R Core Team (2019) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna.
- Ruhí A., Boix D., Gascón S., Sala J. & Batzer D. P. (2013) Functional and phylogenetic relatedness in temporary wetland invertebrates: current macroecological patterns and implications for future climatic change scenarios. *PLoS One* **8**, e81739.
- Ruhí A., Chappuis E., Escoriza D. *et al.* (2014) Environmental filtering determines community patterns in temporary wetlands: a multi-taxon approach. *Hydrobiologia* **723**, 25–39.
- Soininen, J. (2014) A quantitative analysis of species sorting across organisms and ecosystems. *Ecology*, **95**, 3284–92.
- Stenert C., Bacca R. C., Mostardeiro C. C. & Maltchik L. (2008) Environmental predictors of macroinvertebrate communities in coastal wetlands of southern Brazil. *Mar. Freshwater Res.* **59**, 540–48.
- Trindade, C. R. T., Landeiro, V. L. & Schneck, F. (2018) Macrophyte functional groups elucidate the relative role of environmental and spatial factors on species richness and assemblage structure. *Hydrobiologia*, **823**, 217–30.
- Van de Meutter F., De Meester L. & Stoks R. (2007) Metacommunity structure of pond macroinvertebrates: effects of dispersal mode and generation time. *Ecology* **88**, 1687–95.
- Vellend M. (2016) *The Theory of Ecological Communities*. Princeton University Press, Princeton.
- Vieira, N., Poff, N., Carlisle, D., II, S., Koski, M. & Kondratieff, B. (2006) A Database of Lotic Invertebrate Traits for North America. *U.S. Geol. Surv. Data Ser*
- Villwock J. A. & Tomazelli L. J. (2006) Planície Costeira do Rio Grande do Sul: gênese e paisagem atual. In: *Biodiversidade; Regiões da Lagoa do Casamento e dos Butiazais de Tapes, planície costeira do Rio Grande do Sul* (eds F. G. Becker, R. A. Ramos & L. A. Moura) pp. 20–33. Ministério do Meio Ambiente, Brasília.
- Williams, D. D. (1997) Temporary ponds and their invertebrate communities. *Aquat. Conserv. Mar. Freshw. Ecosyst.* **7**, 105–17.
- Williams, D. D. (2006) *The Biology of Temporary Waters*. New York: Oxford University Press
- Winegardner A. K., Jones B. K., Ng I. S., Siqueira T. & Cottenie K. (2012) The terminology of metacommunity ecology. *Trends Ecol. Evol.* **27**, 253–54.
- Wissinger S. A. (1997) Cyclic colonization and predictable disturbance: a template for biological control in ephemeral crop systems. *Biol. Contr.* **10**, 1–15.
- Wissinger S. A., Bohonak A. J., Whiteman H. H. & Brown W. S. (1999) Subalpine wetlands in Colorado: habitat permanence, salamander predation, and invertebrate communities. In: *Invertebrates in Freshwater Wetlands of North America: Ecology*

and Management (eds D. P. Batzer, R. D. Rader & S. A. Wissinger) pp. 757–90. Wiley, New York.

SUPPORTING INFORMATION

Additional supporting information may/can be found online in the supporting information tab for this article.

Fig. S1. General workflow of the statistical procedures undertaken for the assessment of the influence of environmental and spatial factors on the composition of aquatic insects.

Table S1. Geographic location (latitude) and environmental factors (habitat structure, climatic and water chemistry) assessed at the studied ponds.

Table S2. Composition of aquatic insect taxa (and their respective categories of dispersal ability) recorded in the studied ponds.

Table S3. Coefficients of the OLS multiple linear regressions of the environmental predictors (retained by forward selection) used to predict each spatial variable significantly associated with insect composition.