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### Use of thermal preference metrics to examine state biomonitoring data for climate change effects

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Abstract. Analyses of long-term data are an important component of climate-change research because they can help further our understanding of the effects of climate change and can help establish expectations for biological responses to future climate changes. We used macroinvertebrate data to assess whether biological trends associated with directional climate change could be detected in routine biomonitoring data from Maine, North Carolina, and Utah. We analyzed data from 8 long-term biomonitoring sites that had 9 to 22 y of data, and focused on thermal-preference metrics based on coldand warm-water-preference trait groups. The thermal-preference metrics were derived primarily from weighted-average or generalized-linear-model inferences based on data from each state database and are region specific. Long-term trends varied across sites and regions. At some sites, the thermal-preference metrics showed significant patterns that could be interpreted as being related to directional climate change, whereas at others, patterns were not as expected or were not evident. The strongest trends occurred at 2 Utah sites that had ≥14 y of data. At these sites, cold-water taxa were negatively correlated with air temperature, and, when years were grouped into hottest- and coldest-year samples, were strongly reduced in the hottest-year samples. Results suggest that thermal-preference metrics show promise for application in a biomonitoring context to differentiate climate-related responses from other stressors.

**Key words:** traits, climate change, macroinvertebrates, biomonitoring, temperature, hydrology, coldwater taxa, warm-water taxa.

Water-quality agencies use biological indicators to assess the status and health of ecosystems as required by the Clean Water Act in the US and other laws in Europe and Australia. These biomonitoring assessments are comprehensive and direct measures of the ability of a water body to support aquatic life and are anchored in comparisons to regionally established reference benchmarks of ecological condition. The assessments are used to gauge effects of stressors and

are a basis for making regulatory and resourcemanagement decisions.

Metrics based on abundance or richness of taxonomic groups, such as Ephemeroptera, Plecoptera, and Trichoptera, are used often in bioassessments. Another framework is a traits-based approach. The traits-based approach attempts to link species attributes from various trait groups (e.g., life history, morphology, mobility, ecology) to environmental conditions. A cornerstone of the traits-based approach is the Habitat Templet Concept (Southwood 1977, 1988), which predicts that habitat and environmental conditions select organisms with particular life-history strategies and biological traits. The develop-

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ment of the River Habitat Templet (Townsend and Hildrew 1994) further advanced this theory for aquatic ecosystems, and many studies have since demonstrated that patterns in the traits of species can be related to environmental conditions (e.g., Statzner et al. 1994, Richards et al. 1997, Townsend et al. 1997, Van Kleef et al. 2006).

Traits-based biomonitoring approaches have been applied most extensively in Europe (e.g., Dolédec et al. 1999, Charvet et al. 2000, Usseglio-Polatera et al. 2000, Gayraud et al. 2003, Bady et al. 2005). In recent years, as part of the Development and Testing of an Integrated Assessment System for the Ecological Quality of Streams and Rivers throughout Europe using Benthic Macroinvertebrates (AQEM) project (www.aqem.de), Europeans have compiled comprehensive species trait information for >12,000 European freshwater organisms, including macroinvertebrates, fish, macrophytes, diatoms, and phytoplankton, into the Taxa and Autecology Database for Freshwater Organisms, which is accessible online (www.freshwaterecology.info) (Schmidt-Kloiber et al. 2006). Traits data for many North American taxa also are available online in formats ready to analyze. In 2006, the US Geological Survey (USGS) published a database of lotic invertebrate traits for North America that was the first comprehensive web-accessible summary of traits for North American invertebrate taxa (Vieira et al. 2006).

We used macroinvertebrate traits data to assess whether biological trends associated with directional climate change could be detected in routine biomonitoring data in Maine, North Carolina, and Utah. Our study is unique in that it is one of the first to evaluate routine state biomonitoring data in the US for longterm climate-induced trends. We analyzed traits data from 8 long-term biomonitoring sites that had 9 to 22 y of data. Long-term data are an important component of climate-change research because they can help further our understanding of the effects of climate change and can help establish expectations for biological responses to future climatic changes. Advantages of using traits data in these types of long-term and regional trend analyses are that they can be less susceptible to taxonomic ambiguities or inconsistencies (Moulton et al. 2000), they can detect changes in functional community characteristics (e.g., Bonada et al. 2006, 2007, Bêche and Resh 2007), and they provide a consistent framework for assessing community responses to gradients across local and regional scales (Vieira et al. 2006).

We focused on thermal-preference traits because metrics based on cold- and warm-water-preference trait groups were more responsive to existing levels of climate-induced changes in the regions examined than were other biological traits and trait groups (USEPA 2010). Moreover, temperature effects on stream organisms have been well documented. Direct effects of rising temperatures include changes in metabolism, growth, development, phenology, and distribution of organisms (Hogg and Williams 1996, Daufresne et al. 2004, Harper and Peckarsky 2006). Some cold-water organisms could face local extinctions, particularly in high-elevation headwater streams where habitat and dispersal options are limited (Sweeney et al. 1992, Poff et al. 2002). Indirect effects include alterations to dissolved O2 levels, primary production, organic decomposition, and litter processing (Richardson 1992, Durance and Ormerod 2008).

The thermal-preference metrics used in our study are unique in that they were derived primarily from weighted-average or generalized-linear-model inferences based on data from each state database, and, therefore, they are region specific. Weighted-average inferences are used in ecology as a simple, robust approach for estimating the central tendencies of different taxa, or in this case, of thermal optima and tolerance values (ter Braak and Looman 1986). We investigated whether climatic trends could be detected in routine state biomonitoring data with metrics based on cold- and warm-water-preference trait groups. We examined yearly trends in thermalpreference metrics and associations between thermal-preference metrics and temperature. We expected that cold-water-preference metrics would be negatively associated with temperature and warm-waterpreference metrics would be positively associated with temperature.

#### Methods

Study area

We selected Utah, Maine, and North Carolina for our study because they have long-term data that were collected and reported using consistent techniques, and they represent regions of the US with different climate, geography, topography, geology, and hydrology. Utah is in the southwestern region and has a semiarid to arid climate. Maine is in the northeastern region and has a continental climate with cold winters and warm summers in its interior zone, whereas its coastal zone has more moderate summer and winter temperatures (Jacobson et al. 2009). North Carolina is in the southeastern region and has a subtropical and humid climate with long summers and short, mild winters. Warming trends have been observed in each of these states over the past several decades (USEPA

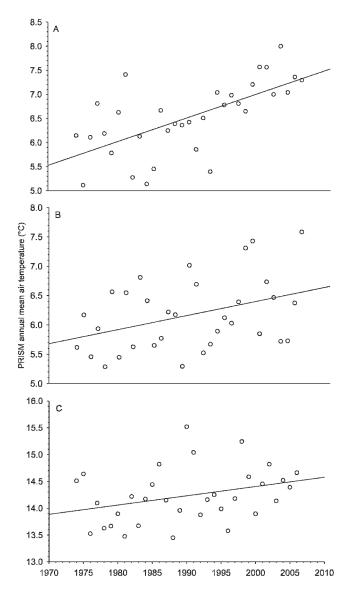


Fig. 1. Annual Parameter-elevation Regressions on Independent Slopes Model (PRISM) air temperatures (°C) from 1974 to 2006 (averaged across all biological sampling sites) in Utah ( $r^2 = 0.42$ , p < 0.01) (A), Maine ( $r^2 = 0.15$ , p = 0.03) (B), and North Carolina ( $r^2 = 0.11$ , p = 0.06) (C).

2010). The strongest trend has occurred in Utah, whereas trends in Maine and North Carolina have been more gradual (Fig. 1A–C). The HadCM3 model projects that temperatures will increase from 1.1 to 2.9°C in each of these regions by 2050, with increases varying regionally and seasonally (NCAR 2008; Table 1).

Even though these states have some of the longest records of state biomonitoring data in the US, long-term (defined here as  $\geq 9$  y of data) monitoring sites are scarce. We selected 8 best-available or minimally disturbed (Bailey et al. 2004, Stoddard et al. 2006)

long-term monitoring sites based on guidance from the respective state agencies. We screened these sites based on chemistry and habitat data (where available), and examined 2001 National Land Cover Data (NLCD; Vogelmann et al. 2001) within a 1-km buffer zone around each site to evaluate the degree to which the long-term trends might have been influenced by anthropogenic stressors. We aggregated landuse classifications into broad categories (e.g., urban and agricultural). Urban land uses at these sites generally consisted of low-intensity and open-space development, whereas agricultural land uses were mostly pasture/hay, with occasional cultivated crops. Anthropogenic influences were higher than desired (>5% urban or >10% agricultural) at 6 of the 8 sites (Utah [UT]-1, UT-3, Maine [ME]-1, ME-2, ME-3 and North Carolina [NC]-1; Table 2), but we analyzed data from these sites because they represented the best-available long-term data in the state databases.

Two sites were in the Wasatch and Uinta Mountain level III ecoregion of Utah, which has a core area of high, precipitous mountains with narrow crests and valleys flanked in some areas by dissected plateaus and open high mountains (Omernik 1987, USEPA 2002). Two other Utah sites were in the Colorado Plateaus level III ecoregion, which is characterized by rugged tableland topography (Omernik 1987, USEPA 2002). Three sites were in the Laurentian Plains and Hills level III ecoregion in eastern Maine (Maine/New Brunswick Plains and Hills). This ecoregion is mostly forested and has dense concentrations of continental glacial lakes (Omernik 1987, USEPA 2002). The site in North Carolina was in the western part of the state in the Blue Ridge level III ecoregion, where terrain ranges from narrow ridges to hilly plateaus to mountainous areas with high peaks. Its high-gradient, cool, clear streams have high diversity of flora and fauna (Omernik 1987, USEPA 2002).

The number of years of data available for these sites varied. ME-1 had the longest-term record (22 y of continuous data, 1985–2006), followed by UT-1 (17 y, 1985–2005) (Table 3). UT-4 and ME-3 had the fewest years of data (9, mid-1990s to 2005). Elevations and stream sizes varied across sites. The highest elevation sites (>1300 m) were in Utah, whereas the lowest elevation sites (<75 m) were in Maine (Table 2). ME-3 and ME-2 had the smallest drainage areas (12.8 km² and 38.1 km², respectively), whereas NC-1 had the largest (835 km²).

#### Data preparation

We compiled biomonitoring data from each state into separate Ecological Data Application System

TABLE 1. Climate-change projections for US northeastern, southeastern, and southwestern regions. Data were downloaded from the Regional Climate-Change Projections from Multi-Model Ensembles (RCPM) project website (http://www.rcpm.ucar. edu/)<sup>a</sup>. Values represent average temperature changes (°C) projected to occur across a range of low- and high-emission scenarios during different seasons, as indicated by the HadCM3 model from the Hadley Centre. 2050 and 2090 are the middle years of the 2-decade future spans that were considered in these analyses and are compared to average temperatures that occurred from 1980 to 1999.

Year	US region	December–February	March–May	June-August	September–November
2050	Northeast	1.1–2.4	1.6–2.3	1.8–2.8	1.9–2.4
	Southeast	1.1–2.0	1.6–2.2	1.9–2.4	1.5–2.2
2090	Southwest	1.6–2.1	1.2–2.1	1.9–2.9	2.1–2.6
	Northeast	3.1–4.1	2.7–4.2	3.2–5.7	3.0–4.5
	Southeast	2.3–3.4	2.4–4.5	2.8–5.1	2.7–4.3
	Southwest	2.4–3.8	2.4–4.5	3.5–5.7	3.6–4.9

<sup>&</sup>lt;sup>a</sup> The RCPM project uses data obtained from the World Climate Research Programme's (WCRP's) Coupled Model Intercomparison Project phase 3 (CMIP3) multimodel data set. RCPM gratefully acknowledges the modeling groups for making their simulations available for analysis, the Program for Climate Model Diagnosis and Intercomparison (PCMDI) for collecting and archiving the CMIP3 model output, and the WCRP's Working Group on Coupled Modelling (WGCM) for organizing the model data analysis activity. The WCRP CMIP3 multimodel data set is supported by the Department of Energy Office of Science.

databases in Microsoft Office Access (2007; Microsoft Corporation, Redmond, Washington). We screened data to minimize the chance of detecting false trends caused by changes in field and laboratory protocols (e.g., differences in collection methods, sample processing/subsampling methods, taxonomists, or taxonomic keys). For Utah, we used only autumn (September–November) kick-method samples collected in riffle habitats (UTDWQ 2006). For Maine, we used only samples collected with rock bags or rock baskets during late-summer (July-September), lowflow periods (Davies and Tsomides 2002). For North Carolina, we used only samples collected during summer (July-August) with the standard qualitative (full-scale) method, which consists of 2 kick, 3 sweep, 1 leaf-pack, 2 fine-mesh-rock or log-wash, and 1 sand sample (NCDENR 2006). In addition, crew members did visual collections during which they walked the stream reach and sampled habitats and substrate types that might have been missed or under-sampled by the other collection techniques (NCDENR 2006).

We followed guidelines in Cuffney et al. (2007) to develop operational taxonomic units (OTUs) for each database to exclude ambiguous taxa from the analyses and to include only distinct/unique taxa. Because of taxonomic ambiguities in the long-term data, genuslevel OTUs were generally most appropriate, although there were some exceptions. Traits analyses based on genus- and family-level taxonomy have been used successfully to characterize aquatic communities for bioassessment purposes (Dolédec et al. 2000, Gayraud et al. 2003), and congeneric species often have similar functional trait niches (Poff et al. 2006).

#### Temperature data

We attempted to acquire all available site-specific water-temperature data for each site. The data we were able to gather consisted primarily of instantaneous water-temperature measurements that were made at the time of the sampling event. We would have preferred to use continuous temperature data recorded by temperature loggers because continuous data capture more aspects of the thermal regime, such as timing, duration, and frequency of extremes, but such data were unavailable.

We supplemented the limited water temperature data by using Geographic Information System (GIS) software (ArcGIS 9.2; ESRI, Redlands, California) to obtain Parameter-elevation Regressions on Independent Slopes Model (PRISM) annual average maximum and minimum air-temperature data for each site from 1974 to 2006 (PRISM Climate Group, Oregon State University, Corvallis, Oregon; http://www. prismclimate.org). PRISM uses a digital elevation model and point measurements of climate data to generate estimates of annual, monthly, and eventbased climatic elements. For purposes of our study, maximum and minimum air temperature values were averaged to derive what we refer to as mean annual air temperature. Air temperatures can track water temperatures closely in the absence of large effects from evaporative cooling, warm-water additions, or groundwater damping (Caissie 2006). These factors are unlikely to have influenced water temperatures at the study sites, but we lacked the data necessary to confirm this.

Site characteristics for the long-term biological monitoring stations in Utah (UT), Maine (ME), and North Carolina (NC). Percent urban and agricultural (ag) apply to a 1-km buffer zone around each site and are based on 2001 National Land Cover Data. EPA = Environmental Protection Agency. W.Br. West Branch Table 2.

	,		•			Drainage		1
Site ID	Water body	Longitude (°)	Latitude (°)	EPA level 3 ecoregion	Elevation (m)	area (km²)	% urban	% ag
$UT-1^a$	Weber	111.37358	40.75294	Wasatch and Uinta Mountains	1846.6	740.7	4.5	21.1
UT-2	Virgin	112.94808	37.28483	Colorado Plateaus	1369.2	756.3	3.4	0.5
UT-3	Duchesne	110.83000	40.46139	Colorado Plateaus	2123.5	489.5	4.8	10.3
UT-4	Beaver	112.56711	38.28000	Wasatch and Uinta Mountains	1904.8	236.2	3.9	0.0
ME-1	Sheepscot	69.59334	44.22319	Laurentian Plains and Hills	31.6	362.8	16.4	23
ME-2	W.Br.Sheepscot	69.53129	44.36791	Laurentian Plains and Hills	70.1	38.1	9.1	18.5
ME-3	Duck	68.23461	44.39340	Laurentian Plains and Hills	54.6	12.8	15.9	0
NC-1	New	81.18330	36.55220	Blue Ridge	713.6	835.0	25.0	13.4

Site is 0.8 km above a reservoir

#### Metric calculations

We derived lists of cold- and warm-water-preference taxa primarily from thermal optima values specific to each state or region. We used the guidelines of Yuan (2006) to calculate optima values based on instantaneous water-temperature measurements and occurrences of organisms. We derived optima values for Utah and Maine from weightedaverage inferences. We supplemented the lists for Utah with weighted-average inferences derived from data sets from Idaho (Brandt 2001) and Oregon (Yuan 2006). We used maximum-likelihood inferences in North Carolina because North Carolina Department of Environment and Natural Resources (NCDENR) abundance data are categorical (1 = rare: 1-2 specimens, 3 = common: 3-9 species, 10 = abundant: ≥10 species). To improve model performance, we calculated optima values only for taxa occurring in >9 sites or samples.

Because the methods used to derive the thermal optima values and the specific characteristics of the data sets (e.g., range of collection dates, station locations, elevation) varied, we developed an arbitrary ranking scheme to make results more comparable across data sets. We assigned taxa in each state rankings ranging from 1 to 7 based on percentiles within each data set. Initially, we designated taxa with rankings  $\leq 3$  ( $\leq 40^{th}$  percentile) as cold-water taxa and taxa with rankings  $\geq 5$  ( $\geq 60^{th}$  percentile) as warm-water taxa. Thermal optima values were not available for all taxa, so we used literature, primarily the traits matrix in Poff et al. (2006) and the USGS traits database (Vieira et al. 2006), as a basis for making some additional initial designations.

After making initial cold- and warm-water designations, we refined the lists in each state based on case studies and best professional judgment from regional advisory groups. Thermal tolerance values, which were calculated using the methods described above (Yuan 2006), also were taken into consideration. We thought these additional considerations were necessary because some taxa occurred with greater frequency in warm- or cold-water habitats but were not present exclusively in one or the other. For example, some taxa initially designated as cold-water taxa also were present at sites that had the hottest recorded water temperatures. During the refinement process, we removed these taxa from the cold-water list. Also, we occasionally removed taxa from the lists because regional taxonomists did not think that the literaturebased designations were appropriate for their region. The cold-water-preference lists in Utah, Maine, and North Carolina consisted of 33, 39, and 32 taxa,

TABLE 3. Time periods for which biological data were available at the long-term monitoring sites in Utah (UT), Maine (ME), and North Carolina (NC). Data used in these analyses were limited to autumn (September–November) kick-method samples in the Utah data set, summer (July–September) rock-basket samples in the Maine data set, and summer (July–August) standard qualitative samples in the North Carolina data set.

Site ID	Water body	Number of years of data analyzed	Years
UT-1	Weber	17	1985–1995, 1998, 2000, 2001, 2003–2005
UT-2	Virgin	14	1985–1993, 1996, 2000–2002, 2004
UT-3	Duchesne	12	1985–1993, 1995, 2000, 2001
UT-4	Beaver	9	1996–1998, 2000–2005
ME-1	Sheepscot	22	1985–2006
ME-2	West Branch Sheepscot	12	1995–2006
ME-3	Duck	9	1997–2005
NC-1	New	11	1983–1990, 1993, 1998, 2003

respectively. The warm-water-preference lists in Utah, Maine, and North Carolina consisted of 16, 40, and 27 taxa, respectively. The relatively low number of taxa on the Utah warm-water-preference list was partially a consequence of the need to use a family-level OTU for Chironomidae because of inconsistencies in the long-term data set that arose from a change in taxonomic laboratories. These lists are the basis of the region-specific thermal-preference richness and relative-abundance metrics used in our study (Appendix; available online from: http://dx.doi.org/10.1899/10-003.1.s1)

#### Data analysis

We used Pearson product moment correlation analyses to assess yearly trends in the thermal-preference metrics and to examine associations between the cold- and warm-water-preference trait groups and PRISM mean annual air temperature. We considered associations significant if p < 0.05. The PRISM annual air-temperature data used in these analyses were different from the instantaneous water-temperature measurements used to develop the cold- and warm-water-preference metrics.

We used 1-way analysis of variance (ANOVA) to evaluate differences in mean thermal-preference metric values between samples collected during coldest, normal, and hottest years. We based groupings on 25<sup>th</sup> and 75<sup>th</sup> percentiles of PRISM mean annual air-temperature data for years during which biological samples were collected. Gaps in the biological data prevented us from designating groupings based on the full range of temperature data (1974–2006), which would have been preferable. PRISM mean annual air temperatures for the hottest-year samples were ~1 to 2°C higher than for the coldest-year samples (Table 4), a difference that corresponds with HadCM3 model projections for

2050 (NCAR 2008). We used the Tukey Honestly Significant Difference (HSD) test for unequal sample size (n) (Spjøtvoll and Stoline 1973) to identify significant differences between specific year groups (p < 0.05). If multiple samples or replicates were collected in a year, we averaged metric values across samples to produce 1 value/y. We used Statistica software (version 8.0; StatSoft, Tulsa, Oklahoma) for all analyses.

#### **Results**

Trends in thermal-preference metrics varied across sites and regions. Significant yearly trends occurred at 3 sites (ME-1, UT-1, UT-2). Both cold- and warmwater-preference metrics were positively correlated with year at ME-1 (Table 5). At UT-1 and UT-2, coldwater-preference metrics were negatively correlated with year (Table 5). The warm-water-preference richness metric increased over time at UT-2 (r = 0.85, p < 0.01).

Associations between thermal-preference metrics and PRISM mean annual air temperature were mixed. At some sites, the metrics showed significant patterns that could be interpreted as being related to directional climate change, whereas at others, patterns were not as expected or were not evident. The strongest expected trends occurred in the coldwater-preference groups at UT-1 and UT-2, where richness metrics were significantly and negatively correlated with PRISM mean annual air temperature (Table 6). Also, at UT-2, the warm-water richness metric was significantly and positively correlated with PRISM mean annual air temperature (r = 0.76, p < 0.01).

Significant patterns occurred at ME-3 and ME-2 but were not as expected. At ME-3, the warm-water richness metric was significantly and negatively correlated with PRISM mean annual air temperature

Table 4. Mean Parameter-elevation Regressions on Independent Slopes Model (PRISM) annual air temperature values in the temperature year groups (coldest, normal, hottest) used in 1-way analysis of variance of data from long-term biological monitoring sites in Utah, Maine, and North Carolina. dT = difference in temperature between samples collected during the hottest and coldest years.

	Nu	ımber of sample	es	Mear	dT (Hottest –		
Site	Coldest	Normal	Hottest	Coldest	Normal	Hottest	coldest)
UT-1	5	7	5	5.9	6.8	7.6	+1.7
UT-2	4	6	4	10.7	11.3	13.4	+2.7
UT-3	3	6	3	3.3	3.9	4.8	+1.5
UT-4	3	3	3	8.0	8.6	9.1	+1.1
ME-1	6	10	6	6.8	7.3	8.3	+1.5
ME-2	3	6	3	6.6	7.1	8.3	+1.7
ME-3	3	3	3	7.2	7.8	8.6	+1.4
NC-1	3	5	3	10.2	10.7	11.7	+1.5

(r = -0.73, p = 0.02), and the warm-water relative-abundance metric showed a similar trend at ME-2 (r = -0.60, p = 0.04) (Table 6). Trends at other sites were not significant. However, the cold-water-preference metrics in Utah and North Carolina consistently showed negative associations with PRISM mean annual air temperature.

Results from the 1-way ANOVA analyses also were mixed. At UT-1 and UT-2, mean numbers of coldwater-preference taxa were significantly (p < 0.05) lower in the hottest-year samples, and % cold-water-preference individuals followed a similar pattern (Table 7, Fig. 2A, B). A similar but nonsignificant pattern occurred with the cold-water richness metric at UT-3, whereas at UT-4, no trend was evident (Fig. 2C, D). Another significant and expected pattern occurred at UT-2, where the mean number of warmwater-preference taxa was significantly higher in hottest-year samples (Table 7). Warm-water-preference taxa either were absent or were present in low numbers at the other Utah sites.

At the Maine sites, assemblages consisted of higher numbers of warm-water-preference taxa than cold-water-preference taxa (Table 7). A significant and expected pattern occurred at ME-2, where % cold-water-preference individuals was significantly (p < 0.05) higher in the coldest-year samples (Fig. 3B). A similar but nonsignificant pattern occurred at ME-3 (Fig. 3C). Nonsignificant and expected patterns occurred at ME-1 and ME-2, where mean numbers of warm-water-preference taxa were lowest in the coldest-year samples (Table 7, Fig. 3A, B). No patterns were evident at NC-1.

#### Discussion

The shifts in cold- and warm-water-preference trait groups that occurred at some of the long-term monitoring sites in Utah and Maine suggest that thermal-preference metrics show promise for application in a biomonitoring context. Thermal-preference metrics are relatively straightforward to interpret,

Table 5. Results of Pearson product moment correlation analyses done to examine associations between year and thermal-preference metrics at long-term biological monitoring sites in Utah (UT), Maine (ME), and North Carolina (NC). Significant (p < 0.05) relationships are shown in bold. NA = not applicable (warm-water-preference taxa absent).

		Cold				Warm			
Number of		Richness		Relative abundance		Richness		Relative abundance	
Site	samples	r	р	r	р	r	р	r	р
UT-1	17	-0.71	0.00	-0.72	0.00	-0.21	0.42	-0.21	0.42
UT-2	14	-0.62	0.02	-0.63	0.02	0.85	0.00	0.41	0.15
UT-3	12	-0.38	0.23	-0.15	0.64	0.38	0.22	0.42	0.17
UT-4	9	-0.64	0.07	-0.12	0.76	NA	NA	NA	NA
ME-1	22	0.49	0.02	0.48	0.03	0.75	0.00	0.51	0.02
ME-2	12	-0.17	0.60	0.29	0.36	-0.51	0.09	-0.48	0.11
ME-3	9	0.54	0.13	0.45	0.23	0.58	0.10	-0.36	0.34
NC-1	11	0.55	0.08	0.57	0.07	-0.58	0.06	-0.04	0.90

Table 6. Results of Pearson product moment correlation analyses done to examine associations between Parameter-elevation Regressions on Independent Slopes Model (PRISM) mean annual air temperatures and thermal-preference metrics at long-term biological monitoring sites in Utah (UT), Maine (ME), and North Carolina (NC). Significant (p < 0.05) relationships are shown in bold. NA = not applicable (warm-water-preference taxa absent).

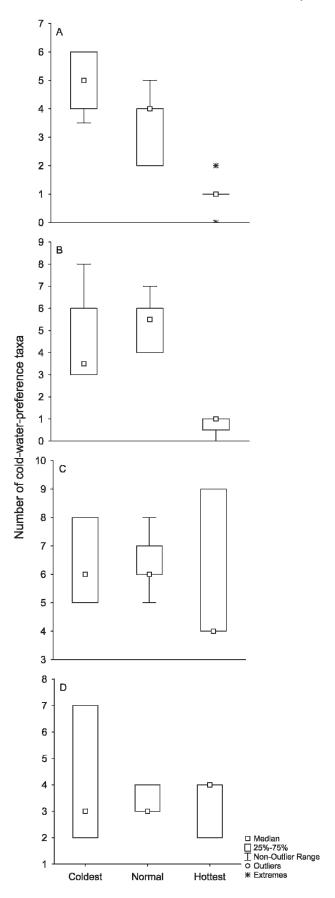
		Cold				Warm			
	Number of	Richness		Relative abundance		Richness		Relative abundance	
Site	samples	r	р	r	р	r	р	r	р
UT-1	17	-0.63	0.01	-0.30	0.24	-0.44	0.08	-0.35	0.17
UT-2	14	-0.73	0.00	-0.56	0.04	0.76	0.00	0.62	0.02
UT-3	12	-0.08	0.82	-0.20	0.53	-0.03	0.93	0.01	0.98
UT-4	9	-0.14	0.73	-0.29	0.46	NA	NA	NA	NA
ME-1	22	0.33	0.13	0.15	0.50	0.27	0.23	0.16	0.49
ME-2	12	0.13	0.69	-0.57	0.05	0.47	0.13	-0.60	0.04
ME-3	9	-0.58	0.10	-0.27	0.48	-0.73	0.02	0.05	0.90
NC-1	11	-0.38	0.25	-0.32	0.34	-0.18	0.59	0.00	0.99

although somewhat masked by interannual variability. They are informative regarding temperature effects and mechanisms, but they do not necessarily indicate a causal link with climate change. They provide a mechanism for tracking and distinguishing climate-change effects from responses potentially

related to other stressors. They can provide information on spatial distributions of cold- and warm-water assemblages. Last, they can be used to modify and adapt traditionally used indices to make them more responsive to altered temperatures (Hamilton et al. 2010).

Table 7. Mean ( $\pm 1$  SD) richness and % individuals with cold- or warm-thermal preferences in coldest (group 1), normal (group 2), and hottest (group 3) years at long-term biological monitoring sites in Utah (UT), Maine (ME), and North Carolina (NC). Year groups were based on Parameter-elevation Regressions on Independent Slopes Model (PRISM) mean annual air-temperature values at each site. One-way analysis of variance was done to evaluate differences in mean thermal-preference metric values. Groups with the same superscripts within a site are not significantly different (p < 0.05). NA = not applicable (warm water preference taxa absent).

		(	Cold		Varm
Site	Group	Richness	% individuals	Richness	% individuals
UT-1	1	$4.9 \pm 1.1^{A}$	$6.5 \pm 5.3^{A}$	$2.3 \pm 0.8^{A}$	$0.6 \pm 0.5^{A}$
	2	$3.4 \pm 1.1^{A}$	$6.7 \pm 7.3^{A}$	$1.1 \pm 0.7^{A}$	$0.4 \pm 0.3^{A}$
	3	$1.0 \pm 0.7^{B}$	$1.0\pm1.1^{ m A}$	$1.0 \pm 1.2^{A}$	$0.3 \pm 0.4^{A}$
UT-2	1	$4.5 \pm 2.4^{A}$	$15.7 \pm 10.9^{AB}$	$1.5 \pm 0.6^{A}$	$7.7 \pm 6.7^{A}$
	2	$5.3 \pm 1.2^{A}$	$23.4 \pm 15.6^{A}$	$1.5 \pm 0.8^{A}$	$18.1 \pm 15.3^{A}$
	3	$0.8 \pm 0.1^{B}$	$0.2 \pm 0.2^{B}$	$3.8 \pm 1.3^{B}$	$27.8 \pm 19.4^{A}$
UT-3	1	$6.3 \pm 1.5^{A}$	$24.3 \pm 4.1^{A}$	$0.3 \pm 0.6^{A}$	$0.03 \pm 0.1^{A}$
	2	$6.3 \pm 1.0^{A}$	$14.9 \pm 6.8^{A}$	$0.7 \pm 0.8^{A}$	$0.1 \pm 0.2^{A}$
	3	$5.7 \pm 2.9^{A}$	$17.7 \pm 8.5^{A}$	$0.7 \pm 1.2^{A}$	$0.1 \pm 0.2^{A}$
UT-4	1	$4.0 \pm 2.6^{A}$	$12.1 \pm 6.2^{A}$	NA	NA
	2	$3.3 \pm 0.6^{A}$	$10.0 \pm 9.2^{A}$	NA	NA
	3	$3.3 \pm 1.2^{A}$	$8.4 \pm 5.9^{A}$	NA	NA
ME-1	1	$0.5 \pm 0.5^{A}$	$0.6 \pm 0.6^{A}$	$6.4 \pm 2.4^{A}$	$15.6 \pm 7.4^{A}$
	2	$0.5 \pm 0.8^{A}$	$0.7 \pm 1.7^{\mathrm{A}}$	$8.0 \pm 1.4^{A}$	$21.2 \pm 11.5^{A}$
	3	$1.1 \pm 0.5^{A}$	$1.0 \pm 0.8^{A}$	$8.5 \pm 2.7^{A}$	$19.6 \pm 10.7^{A}$
ME-2	1	$1.2 \pm 0.4^{A}$	$9.8 \pm 5.9^{A}$	$4.9 \pm 1.0^{AC}$	$51.7 \pm 12.2^{A}$
	2	$1.0 \pm 0.6^{A}$	$1.6 \pm 1.1^{B}$	$8.4 \pm 1.4^{B}$	$38.9 \pm 15.3^{A}$
	3	$1.4 \pm 0.8^{A}$	$1.7 \pm 1.2^{B}$	$7.8 \pm 1.8^{BC}$	$22.9 \pm 14.6^{A}$
ME-3	1	$2.4 \pm 1.2^{A}$	$7.8 \pm 6.4^{A}$	$6.3 \pm 0.6^{A}$	$44.0 \pm 22.5^{A}$
	2	$1.7 \pm 0.3^{A}$	$5.3 \pm 5.9^{A}$	$6.8 \pm 1.5^{A}$	$32.8 \pm 10.8^{A}$
	3	$1.6 \pm 0.7^{A}$	$5.0 \pm 3.3^{A}$	$4.8 \pm 1.3^{A}$	$46.6 \pm 17.6^{A}$
NC-1	1	$4.3 \pm 1.5^{A}$	$2.3 \pm 0.7^{A}$	$8.3 \pm 0.6^{A}$	$7.7 \pm 2.5^{A}$
	2	$5.4 \pm 1.7^{A}$	$3.6 \pm 2.9^{A}$	$7.4 \pm 1.7^{A}$	$7.6 \pm 2.5^{A}$
	3	$4.0 \pm 1.7^{A}$	$2.2 \pm 1.0^{A}$	$7.3 \pm 2.3^{A}$	$7.0 \pm 1.3^{A}$



The long-term trend analyses in our study have direct relevance to establishing expectations for biological responses to climate-change effects. Regional differences appear to exist, although direct comparisons are difficult to make because of differences in state sampling methods. The strongest trends occurred at the 2 Utah sites that had  $\geq$  14 y of data. At these sites, cold-water taxa decreased, were negatively correlated with temperature, and, when years were grouped into hottest- and coldest-year samples, were strongly reduced in the hottest-year samples. These patterns, in combination with the significant increase in the number of warm-water-preference taxa at UT-2, indicate that the warmer temperatures could have (directly or indirectly) triggered a significant shift in species composition at these sites. The hottest annual temperatures at these sites occurred during 5 consecutive years (2000-2005), a fact that suggests that the biological responses might be partly explained by short-term climatic patterns. Bêche and Resh (2007) showed that short-term climatic patterns explained much of the temporal variation in composition and abundance of macroinvertebrate communities in Mediterranean-climate streams in northern California.

At the study sites in Maine and North Carolina, temperature trends were more variable. The greater environmental variability might partly explain the lack of directional biological trends at some of the sites. In some longer-term data sets, community variability increases with study length because greater environmental variability can occur over the longer study periods (e.g., Bengtsson et al. 1997, Haddad et al. 2002). Anthropogenic influences also could have masked climate-related trends at some of the sites. For example, at ME-1, where the thermal-preference metrics were significantly associated with year but not with temperature, yearly trends in the biological data probably were influenced by nonpoint-source pollution, but we lack the long-term chemistry data necessary to confirm this possibility.

#### Limitations

Our study had several limitations. One was our inability to separate completely biological responses

Fig. 2. Distributions of cold-water-preference richness values in coldest-, normal-, and hottest-year samples at Utah (UT) sites UT-1 (A), UT-2 (B), UT-3 (C), and UT-4 (D). Groupings are based on Parameter-elevation Regressions on Independent Slopes Model (PRISM) mean annual air temperatures from each site. Average temperatures in hottest-year samples were 1.1 to 2.7°C higher than coldest-year samples (Table 4).

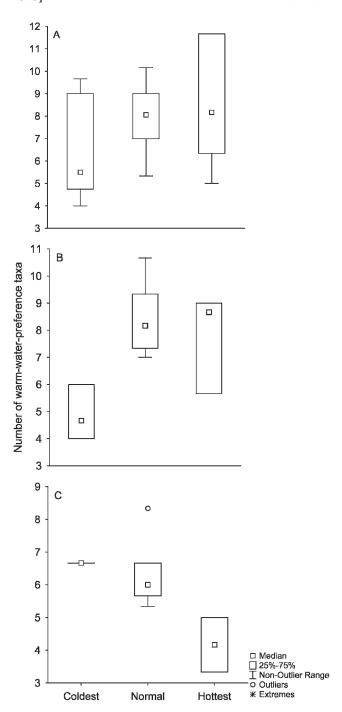


Fig. 3. Distributions of warm-water-preference richness values in coldest-, normal-, and hottest-year samples at Maine (ME) sites ME-1 (A), ME-2 (B), and ME-3 (C). Groupings are based on Parameter-elevation Regressions on Independent Slopes Model (PRISM) mean annual air temperatures from each site. Average temperatures in hottest-year samples were 1.4 to 1.7°C higher than coldest-year samples (Table 4).

to climate-induced changes from responses triggered by other confounding factors. This limitation is generally true in analyses of observational data because covarying factors can confound associations and prevent assignment of cause (Yuan 2010). Non-climatic factors, such as anthropogenic stressors, could have masked climate-related trends at some of the sites. This problem has occurred in studies of other long-term data sets. In Ohio, steady improvements in water quality that occurred simultaneously with climatic changes confounded analyses of long-term trends at reference sites (USEPA 2010). At some river sites in southern England, improvements in water quality also masked climate-related trends (Durance and Ormerod 2008).

Another limitation was the lack of site-specific, continuous water-temperature data. We used annual air-temperature data and instantaneous water temperature as surrogates, but we lacked the data necessary to verify that air and water temperature were strongly correlated at all sites. Other limitations included the small number of long-term monitoring sites, the limited spatial distribution of these sites, and the relatively short durations (often <20 y) of the data sets. Often, samples are not collected from the same sites every year, so many data sets have discontinuities, which make analyzing and detecting trends difficult.

Yet another limitation was that we focused on only one environmental variable, temperature. Warming trends associated with climate change are intertwined with many other factors, such as hydrology, that influence the distribution of species in lotic environments. Altered precipitation patterns, increased frequencies of extreme hydrologic events (floods and droughts), and shifts in the timing of runoff have occurred and are projected to continue to occur as a result of climate change (Bates et al. 2008). In a separate study, we investigated relationships between trait metrics and measures of hydrology (e.g., flow or precipitation as a proxy) in Utah, Maine, and North Carolina and found equivocal results (JDS, AH, LZ, and BB, unpublished data). This outcome probably reflects the significant variability in hydrology and precipitation patterns that was evident in each of the state data sets and that has been documented globally (USEPA 2010, Bates et al. 2008).

#### Recommendations

Climate change affects aquatic ecosystems directly, indirectly, and through interactions with other stressors. All of its complexities are difficult to capture, especially given the limitations and data gaps we

encountered in our study. Nevertheless, given the available data, our study furthers understanding of current detectable effects of climate change on aquatic systems and helps establish expectations for biological responses to future climatic changes. Our study also highlighted knowledge gaps that will help us identify future research needs, both immediate and long-term. We conclude by making the following recommendations:

- 1. Further explore use of thermal-preference metrics in detecting climate-related trends.—As the thermal suitability of the habitats changes, the composition of cold- and warm-water-preference trait groups probably will shift. Use of thermal-preference metrics to monitor for these trends will increase the probability of detecting community responses to warming trends and reduce the likelihood that they will be obscured by taxonomic variability. Thermal-preference metrics also can be used to examine spatial distributions of thermal-preference trait groups. A focus on sites defined on the basis of thermal-preference composition could help refine monitoring approaches for detecting and accounting for climate-change effects.
- 2. Glean additional information from the Utah, Maine, and North Carolina databases.—These data sets could be used to investigate time-lagged biological responses to temperature effects, interannual variability, and climatic shifts associated with the North Atlantic Oscillation, El Niño Southern Oscillation, and Pacific Decadal Oscillation. Such studies could be modeled after studies by Bêche and Resh (2007) and Durance and Ormerod (2007). Additional types of statistical analyses, such as the RLQ ordination technique used in Díaz et al. 2008 (described in Dolédec et al. 1996) could be explored.
- 3. Examine long-term biomonitoring data in other states.—Long-term data can help us understand climate-related effects and will further our understanding of the natural variability in community composition and condition among sites. In addition to macroinvertebrate data, long-term fish and periphyton data sets should be analyzed (if available).
- 4. Continue to refine existing lists of cold- and warm-water-preference taxa and develop lists for more states and regions.—The cold- and warm-water lists developed for our study (Appendix) were a first step, not a final product. They should be refined as more information becomes available and should be compared across states and regions. Refinements can be made by using continuous water-temperature data instead of instantaneous water-temperature data, by calculating propensity scores to help improve the robustness of the analyses (Yuan 2010), and by using species-level OTUs for genera in which differences in

- species-level thermal preferences are known to occur. In addition, it would be valuable to examine variability in thermal optima and tolerance inferences calculated from data sets specific to particular community types (such as those described in Poff et al. 2010) or ecoregions (our calculations were based on data sets that spanned several different ecoregions within each state).
- 5. Conduct case studies to further our understanding of how rare and extreme climate events might affect assemblages.—These studies can be modeled after existing studies. Examples include: Bêche and Resh (2007), which documented the effects and recovery of macroinvertebrate assemblages from prolonged and severe droughts in northern California; Herbst and Cooper (2010), which examined rain-on-snow flooding effects on aquatic invertebrate communities of small streams in the Sierra Nevada, California; and case studies by NCDENR (2004, 2005), which documented effects of long-term drought and catastrophic flooding on aquatic communities in North Carolina streams
- 6. Continue to further our knowledge of traits and how they relate to climate change.—More information is needed about which traits are most important in the context of climate change, the influence of each trait on an organism's ability to adapt, and which combinations of traits are most adaptive to particular environmental conditions. Our knowledge of biological trait responses of lotic invertebrates to individual and multiple stressors continues to grow (reviewed in Statzner and Bêche 2010). A key component of furthering the traits-based framework will be expansion and unification of existing trait databases (Statzner and Bêche 2010).
- 7. Encourage researchers to continue collecting biological samples at long-term monitoring sites (if possible, annually).—Collection of continuous temperature and flow data at these sites would be extremely valuable as well. If these types of data are collected with standardized procedures, they could potentially be integrated into regional databases, and perhaps into a climate-change monitoring network. This type of coordination and collaboration across states and regions would greatly enhance our ability to understand the effects of climate change on aquatic ecosystems.

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