Integrating intermittent streams into watershed assessments: Applicability of an index of biotic integrity

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Abstract

Nonperennial streams are often excluded from biomonitoring programs because of inadequate knowledge about their biological and hydrological characteristics and variability, but applying bioassessment indices to nonperennial streams would greatly expand the reach of these programs. To determine if a benthic macroinvertebrate assessment index (the Southern California Index of Biotic Integrity; IBI) developed for perennial streams could work in nonperennial streams, 12 nonperennial streams (3 of which were minimally stressed) in the San Diego hydrologic region were sampled multiple times. For comparison, three low-stress perennial streams were also sampled. Continuous water-level loggers and repeat site visits revealed that hydrologic regimes varied considerably among streams, with gradual drying evident at some and multiple drying/rewetting events evident at others; in addition, streams that were nonperennial one year were perennial in another. IBI scores from low-stress nonperennial streams were similar to those for low-stress perennial streams, and false indications of impairment (i.e., low IBI scores) were never observed. Furthermore, IBI scores declined as stress increased, suggesting that the IBI responds as expected in nonperennial streams. IBI scores were stable at most sites both within and between years, but midsummer declines were observed at high-stress sites. These declines were associated with declines in discharge, fast water habitat, and increases in sands and fines and macroalgae cover. These findings suggest that an assessment tool developed for perennial streams can accurately assess condition at certain nonperennial streams, and that biomonitoring programs can provide more comprehensive watershed assessments by including nonperennial streams in their surveys.

Introduction

Although nonperennial streams comprise large portions of watersheds in arid and temperate parts of the world, their ecology is not well understood (Williams 2008), and are often excluded from bioassessment programs (e.g., Hall *et al.* 1998, Peck 2006). This exclusion is primarily motivated by inadequate knowledge about the applicability of bioassessment tools that were calibrated with perennial streams and questions about whether index scores can be interpreted correctly (Fritz *et al.* 2008). As a result, many surveys of ambient stream condition are incomplete, and water quality programs do not comprehensively evaluate stream health or provide complete assessments of watershed or regional conditions.

Watershed management and resource protection may only have limited success if nonperennial streams are excluded from assessments. Not only do

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nonperennial streams comprise large portions of watersheds in both arid and wet climates (Tooth 2000, Larned *et al.* 2010), but they also may be among the most sensitive to environmental impacts. Flow intermittence affects several ecosystem processes, such as leaf litter breakdown (e.g., Herbst and Reice 1982, Datry *et al.* 2011), nutrient cycling (e.g., Gómez *et al.* 2012), and biomass production (e.g., Tronstad *et al.* 2010); therefore, degradation of nonperennial reaches may have disproportionate impacts on the health of the entire watershed. Furthermore, they represent the greatest interface (in terms of surface area) between waterbodies and terrestrial ecosystems, where most disturbances occur. Comprehensive assessment and management of watersheds in any climate should include nonperennial streams (Fritz *et al.* 2008, Steward *et al.* 2012).

Nonperennial streams present stressful environments for benthic macroinvertebrates because abiotic and biotic conditions change dramatically, both seasonally and between years (e.g., Bêche *et al.* 2006). These changes are principally driven by changes to the hydrologic regime, as the stream passes from a eurheic state to an arheic, hyporheic, or edaphic state (following the terminology of Gallart *et al.* 2012). As the stream shifts from one state to another, certain microhabitats (especially riffles) become scarce or disappear entirely, reducing the abundance of species that depend on them.

Changes in hydrology may also lead to other environmental changes that affect community composition. When water levels decline, available substrate composition, chemical concentrations, and pollutants (if present) may change and increase environmental stress. Temperature, pH, and dissolved oxygen concentration can fluctuate over short time periods (Gasith and Resh 1999). Biotic pressures can also intensify, as predation and competition become more important as space and resources become limited (Robson *et al.* 2011). These changes may lead to differences in community composition during drying, confounding the ability to discern changes in biotic community due to natural variability in environmental conditions from those due to anthropogenic stress (Morais *et al.* 2004).

Many of the life history traits of benthic macroinvertebrates that enable survival in nonperennial streams (such as tolerance of low oxygen or high conductance conditions) are similar to those used to survive in degraded streams. Therefore, indices calibrated with perennial streams (such as the Southern California Index of Biotic Integrity (IBI), Ode *et al.* 2005) might be expected to give false indications of impairment at nonperennial streams under natural conditions. For example, Mazor *et al.* (2009) found that two bioassessment indices (a multimetric and a multivariate index) indicated impairment in two minimally disturbed nonperennial streams in northern California. Additionally, Morais *et al.* (2004) found that intra-annual variability in bioassessment was particularly high in nonperennial streams in Portugal, meaning that both false positive and false negative findings of impairment may be common in nonperennial streams. To date, it has been unclear if bioassessment indices developed for perennial streams function in nonperennial streams and if the indices have the ability to differentiate changes due to anthropogenic stress from those associated with seasonal fluctuations.

The goal of this study was to determine if standard assessment tools developed for perennial streams can be applied to nonperennial streams in the San Diego region of Southern California, USA. The primary objective of the study was to evaluate whether an assessment index developed for perennial streams would correctly assess the condition of nonperennial streams. To meet this objective, this study investigated the following: 1) How do hydrologic regimes at nonperennial streams differ from those at perennial streams? 2) How does biological community structure vary over time? and 3) Which environmental variables associated with hydrologic regimes are related to these changes in biological community structure and environmental indices? Hydrologic regimes were characterized through flow measurements and deployment of water-level loggers at selected sites. Biological communities were characterized by sampling benthic macroinvertebrates and calculating the IBI or by multivariate ordination. Environmental variables were characterized through measurement of the physical habitat using standard protocols (Ode 2007) and by analyzing the water chemistry.

Methods

Study Area

Southern California has a Mediterranean climate, with hot, dry summers, and cool, wet winters. Intermittent streams are typical of the region. Much of the coastal, lower elevation areas have been converted to agricultural or urban land use, where water importation, runoff, and effluent discharges have perennialized most streams (Mazor *et al.* 2012); however, most of the upper elevations of watersheds remain undeveloped, with chaparral, grassland and oak or pine forest land cover. Streams in these undeveloped portions are mostly nonperennial, although short lengths of perennial streams can be found near surficial bedrock or spring sources. Much of the region is comprised of young, erodible sedimentary geology.

Sampling

Twelve nonperennial and three perennial streams in Southern California were selected for sampling of benthic macroinvertebrates and physical habitat. For this study, nonperennial streams are defined as streams that lack surface flow for at least several days per year in most years. This definition encompasses a wide variety of streams, from ephemeral washes and headwaters that flow for only a few hours after storms, to those with sustained flows lasting nearly all year (and even have perennial flow in a year with heavy rainfall). However, all nonperennial streams in this study exhibited flow from groundwater discharge that persisted at least a month past the last storm; following Yavercovski *et al.* (2004), these streams would be classified as seasonal or near-permanent.

The twelve sites were selected to represent a range of natural conditions (e.g., short and long duration of flows; high and low gradient) and as well as anthropogenic stress (e.g., urban development, grazing, water diversion, etc.). Three sites were selected for repeat sampling over multiple years. At a subset of sites, continuous data loggers were deployed to measure water level and temperature throughout the course of one year of sampling (not enough loggers were available for deployment at every site or every year of sampling).

Sampling at nonperennial was scheduled to target the drying phase, typically beginning in March or April and ending when flow was insufficient for further sample collection (i.e., no surface water at > 50% of the sampling reach. In the terminology of Gallart *et al.* (2012), sampling began in the eurheic phase and continued through the oligorheic (or rarely the ahrehic) phases, ending when the stream was observed to be in a drier hydrologic state (i.e., the hyporheic or edaphic phases). Sites were revisited approximately every two to four weeks; when possible, frequency of site visits was increased to weekly towards the end of the drying phase. Sampling at perennial sites was scheduled for monthly visits beginning in April, overlapping the normal index period for bioassessment in perennial streams in Southern California (SMC 2007). Sampling for nonperennial streams was predominant funded under one program, which began in 2008 and was suspended in 2009 for budgetary reasons before resuming in 2010. Sampling for perennial streams was predominantly funded under a second program, which began in 2009 and continued in 2010.

Overall, the study period was drier than the long-term average for the region. Normal rainfall for Lindbergh Airport in San Diego is 10.8 inches, and the three years of sampling each received 7.2, 9.2, and 10.6 inches, respectively. Furthermore, drought was even more severe in the two years preceding the study, with only 5.4 inches of precipitation falling in 2006 and 3.9 inches in 2007 (http://www.sdcwa.org/annual-rainfall-lindbergh-field).

Stressor Assessment

Nonperennial sites were initially selected to represent a gradient of stress using best professional judgment, but *a priori* designations were verified or modified by using a checklist of 50 stressors associated with the California Rapid Assessment Method (CRAM; CWMW 2012), a measure of riverine wetland condition. A total of 50 stressors (covering a variety of hydrologic, physical, land-use, and biological stressors) were evaluated for each site. A stressor was given a score of 0 if it was not observed, a score of 0.5 if the stressor was likely to have a negative impact on the stream, and a score of 1 if the impact was likely to be large. The distribution of scores was examined to identity three groups. Low stress (scores ≤ 1), moderate stress (scores between 2 and 7), and high stress (scores between 7.5 and 9; higher scores were not observed in this study). Although a maximum score of 50 is theoretically possible, scores higher than 6 are uncommon, except at highly developed sites; the highest score observed in a statewide probabilistic data set of 924 sites was 21, and only 25% of this statewide dataset would be classified as high stress (data not shown). This approach to establishing a stressor gradient by enumerating stressors is similar to the approach used by Sánchez-Montoya *et al.* (2009a) to identify reference sites. See Mazor *et al.* (2012) and Supplement Information (SI) Table SI-1 (ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/AnnualReports/2013AnnualReport/

ar13_357_375SI.pdf) for details on stressors identified at each site.

Assessment of Hydrologic Regimes

Hydrologic regimes and stressors were evaluated using a combination of data sources: continuous data loggers, direct measurements, and visual observation during site visits. Continuous water-level loggers were deployed at a subset of sites at the first sampling event and retrieved after the final sampling event. At four sites (BC, AN, CD, and NC), water level was determined by correcting for air pressure measured by a second logger deployed at the site above the water line. At the other sites, nearby weather stations were used to correct for air pressure. Direct measurements of stream discharge were recorded during most sampling events by measuring water velocity using an electromagnetic or propeller-type velocity meter, although flotation time using a neutrally buoyant object was used when conditions were too slow or shallow for the velocity meter. Finally, during site visits it was noted whether streams were flowing. These sources of data were used to identify periods when the reaches contained flowing water, and when they were dry (or intermittently dry), with visual observation being given the highest priority, followed by direct measurements. Long periods (>6 hours) where loggers recorded water-level readings less than 0 were interpreted as times when the stream was dry.

Benthic Macroinvertebrate Collection

Benthic macroinvertebrates were sampled using standard protocols for bioassessment in California (Ode 2007), which are derived from those developed by the US Environmental Protection Agency for national stream surveys (Peck *et al.* 2006). Briefly, a 150-m reach was divided into 11 equidistant transects. At each transect, a 500-µm D-frame kick net was used to sample 1 ft^2 of streambed at 25, 50, or 75% along the transect distance. At three low-gradient sites where stable habitats were restricted to the stream banks (AC, SJ, and PC), samples were collected at 0, 50, and 100% of the transect distance (Mazor *et al.* 2010). Transects were composited into a single sample and preserved in 70% ethanol. For each sample, ~600 individual invertebrates were removed from surrounding detritus and identified to Standard Taxonomic Effort Level 2 (i.e., mostly to species with Chironomidae to genus, where possible) established by the Southwest Association of Freshwater Invertebrates (Richards and Rogers 2011). Benthic macroinvertebrate collection occurred only when flow was sufficient for sampling; that is, the stream was in a eurheic or oligorheic state (sensu Gallart *et al.* 2012) for at least 50% of the reach.

Habitat Characterization

Physical habitat was measured using the protocol referenced above (Ode 2007), with a few modifications for the study. First, the 11 transects were established at fixed locations that did not vary over the course of the study. Thus, sampling extended well beyond the period at which a site would be rejected for sampling due to lack of wet habitat, as mandated by the sampling protocol (Ode 2007). Second, certain aspects of physical habitat were assumed to be stable, and were only measured during one sampling event per year: slope, gradient, bank width, and bank height. Third, the algae cover components of physical habitat were added in 2009 and 2010, following the publication of standard methods of estimation (Fetscher *et al.* 2009).

Physical habitat data were analyzed by calculating metrics following Kaufmann *et al.* (1999), where possible. Other measurements (e.g., data from field probes, such as dissolved oxygen), were analyzed without modification.

Landscape Variables

Landscape variables (specifically, % impervious surface, and road density) were calculated for drainage area upstream of each site. National Landcover Data from 2006 (http://www.epa.gov/mrlc/nlcd-2006.html) were used to assess imperviousness, and road density was derived from a custom road layer.

Data Analysis

Benthic Macroinvertebrate Community Characterization

Benthic macroinvertebrate community structure was examined in two ways: First, with a multimetric bioassessment index (i.e., an IBI), and second, through community composition and structure, analyzed using nonmetric multidimensional scaling (NMS). These measurements of community structure were plotted over time, and compared with environmental variables to examine changes in benthic communities over the course of the study.

Metrics for the Southern California Index of Biotic Integrity (IBI) were calculated as described in Ode *et al.* (2005). In order to maintain consistent sample sizes, each sample was reduced to 500 specimens using random subsampling, and then aggregated to the required level of taxonomy (i.e., Level 1 in Richards and Rogers 2011): most groups to genus with Chironomidae to family. Subsequently, the following 7 metrics were calculated: 1) Coleoptera richness; 2) Ephemeroptera, Plecoptera, and Trichoptera richness; 3) predator richness; 4) % collectors; 5) % intolerant individuals; 6) % non-insect taxa; and 7) % tolerant taxa. These metrics were scored so that higher scores reflect less degraded condition. Metric scores were then summed, and rescaled to a 100-point scale, as described in Ode *et al.* (2005). Values were compared to a threshold of 39, which is two standard deviations below the reference calibration mean, as described in Ode *et al.* (2005).

To examine differences in assemblage composition directly, NMS was run on a presence-absence dataset containing all samples using PC-ORD version 5.12 (McCune and Mefford 2006). Bray-Curtis distance was used, and up to 4 axes were tested using 100 runs with both real and randomized data. Axis selection followed the default procedure of PC-ORD; that is, the highest dimensionality is selected that reduces stress by 5 or more (on a scale of 0 to 100) and has less stress than 95% of runs with randomized data. Maximum number of iterations was set to 250, and the stability criterion was set to 0.000001. Step length was set to 0.2. Final axes were rotated using varimax rotation, and taxon scores were calculated with weighted averaging.

A linear, mixed-effects model was used to determine if flow and stress status affected IBI scores. Using the nlme package in R (Pinheiro *et al.* 2013), a mixed effects model was created for each biological response variable based on four classes of flow and stress (i.e., perennial low stress, nonperennial low stress, nonperennial moderate stress, and nonperennial high stress), with a random effect specified as date nested within site. Subsequently, pairwise significant differences based on flow and stress were tested using the glht function in the multcomp package in R (Hothorn *et al.* 2008). Differences in variability between low-stress perennial and nonperennial sites was examined with an F-test.

Relationships between Biological and Environmental Variables

To examine the relationship between environmental variables and biological assemblage composition (i.e., IBI scores and NMS axis scores), two separate analyses were conducted. First, Spearman rank correlations between within-site means of environmental and biological variables was used to evaluate gradients associated with among-site differences. Second, to evaluate the gradients associated with within-site changes, Spearman rank correlations were calculated on variables after subtracting the within-site mean. Only a subset of environmental variables was used for the second analysis—specifically, those assumed to vary with changing hydrological conditions. Variables assumed to be constant over the course of the study (e.g., all landscape variables, bank dimensions, etc.) were excluded from this analysis. Because the goal of this descriptive analysis was to characterize the relationships between biological and environmental variables, statistical significance of these correlation coefficients was not tested (McCune and Grace 2002).

Results

Stressor Assessment

Stress scores confirmed that the perennial (AN, BC, and CD) and three of the twelve nonperennial sites (TE, AC, and CC) were low stress (Table 1). Among the nonperennial sites, six were moderate stress (NC, AS, SR, CV, PC, and SY), and three were high stress (OF, SJ, TC). The most stressed site (TC) had a score of 8.5. Stressors affecting stressed sites included grazing (at SY, SR, and CV), nutrient impacts (at SY, SJ, OF, and TC), and urban runoff (at PC and TC). Evidence of non-natural flow regimes (e.g., floods unaccompanied by precipitation at site AS) were observed at several sites. Communication with nearby land managers indicated groundwater diversions that affect sites SJ and OF. Complete details about the stressors observed at these sites are available in Mazor *et al.* (2012) and in Table SI-1.

Patterns in Hydrologic Regimes

Examination of data from water-level loggers revealed several different hydrologic patterns among the sites (Figure 1). Even among the three perennial sites, several patterns were evident. Hydrographs were most stable at perennial sites (particularly SA), and at certain nonperennial sites (i.e., SY,

Table 1. Sites sampled in the present study.

Site	Name	Stress Score	Major Stressors	Watershed Area (km^2)	Elevation (m)	Gradient (%)	Ecoregion
Perennial, Low Stress							
AN	South Fork Santa Ana River	0.0	None	11	2447	>2	Mountains
BC	Bear Canyon	0.0	None	65	639	>2	Mountains
CD	Cedar Creek	0.0	None	55	522	>2	Chaparral
Nonperennial, Low Stress							
TE	Temescal Creek	0.0	None	22	333	>2	Chaparral
AC	Agua Caliente Creek	0.5	None	46	918	<1	Mountains
CC	Carney Canyon	0.5	None	19	312	>2	Chaparral
Nonperennial, Moderate Stress							
NC	Noble Canyon	2.0	Altered flow	39	1169	>2	Chaparral
AS	Arroyo Seco	2.5	Altered flow	34	494	<1	Chaparral
SR	San Diego River Headwaters	3.0	Grazing	2	1038	>2	Mountains
CV	Cañada Verde	4.5	Grazing	14	954	>2	Mountains
PC	Pine Valley Creek	5.0	Runoff	74	1132	<1	Chaparral
SY	Santa Ysabel Creek	7.0	Grazing	32	902	1 - 2	Mountains
Nonperennial, High Stress							
OF	Ortega Falls	7.5	Altered flow, runoff	16	575	>2	Chaparral
SJ	San Juan Mainstem	8.0	Altered flow, runoff	103	181	<1	Chaparral
TC	Trabuco Creek	8.5	Runoff	58	237	1 - 2	Chaparral

SR, and NC), where water levels were stable until they lowered abruptly. In contrast, two sites (i.e., SJ and OF) showed periods of large fluctuations. Subsequent communication with the manufacturer of the loggers (Onset) indicated that these fluctuations may indicate periods of extremely low water levels, during which the logger may be partially exposed to air; during these periods, intermittent drying and rewetting of the logger are likely. Intermittent drying was also evident at AS. Additionally, this site also experienced a few short high flow events, despite the lack of recent precipitation. Loggers were not deployed at any low-stress nonperennial site.

Hydrologic characteristics varied from year to year, as revealed by direct observation at three sites with multiple years of sampling (Figure 2). All three sites selected for repeat visits dried in June or July during the first year of sampling (i.e., 2008 for SY and SR, and 2009 for NC). However, only one site (i.e., SR) showed a similar pattern in 2010, as it dried in June both years. In contrast, flow began to dry only in September at SY, and never ceased at NC. Data from multi-year deployment of water-level loggers were not available. Precipitation data from Lindbergh Airport in the City of San Diego indicate that 2010 received more rainfall (10.6”) than 2008 (7.2”) or 2009 (9.2”), although all years received less precipitation than normal (10.8”) (source: http://www.sdcwa.org/annual-rainfall-lindbergh-field).

Trends and Patterns in IBI Scores

IBI scores were high at low-stress sites, regardless of flow status (Figure 3; Table 2) or observed discharge (Figure 4). The three low-stress nonperennial sites had IBI scores that are comparable to low-stress perennial sites, and no sample from these sites was observed below the reference threshold of 39. The linear mixed-effect model showed a small (6.6 points) and non-significant ($p = 0.65$) effect of flow status on IBI score, although power was low with only 3 sites in each low-stress group. Trends for each site are presented in Table SI-2.

IBI scores responded to stress at nonperennial sites in an expected manner (Figure 3; Table 2). Mean scores declined as site quality declined (Spearman rank correlation: -0.91). For example, low-stress sites had higher mean IBI scores than moderate stressed sites (40.3), which in turn had higher scores than high stress sites (26.8). In fact, nearly all samples from all high stress sites had scores well below the reference threshold of 39.

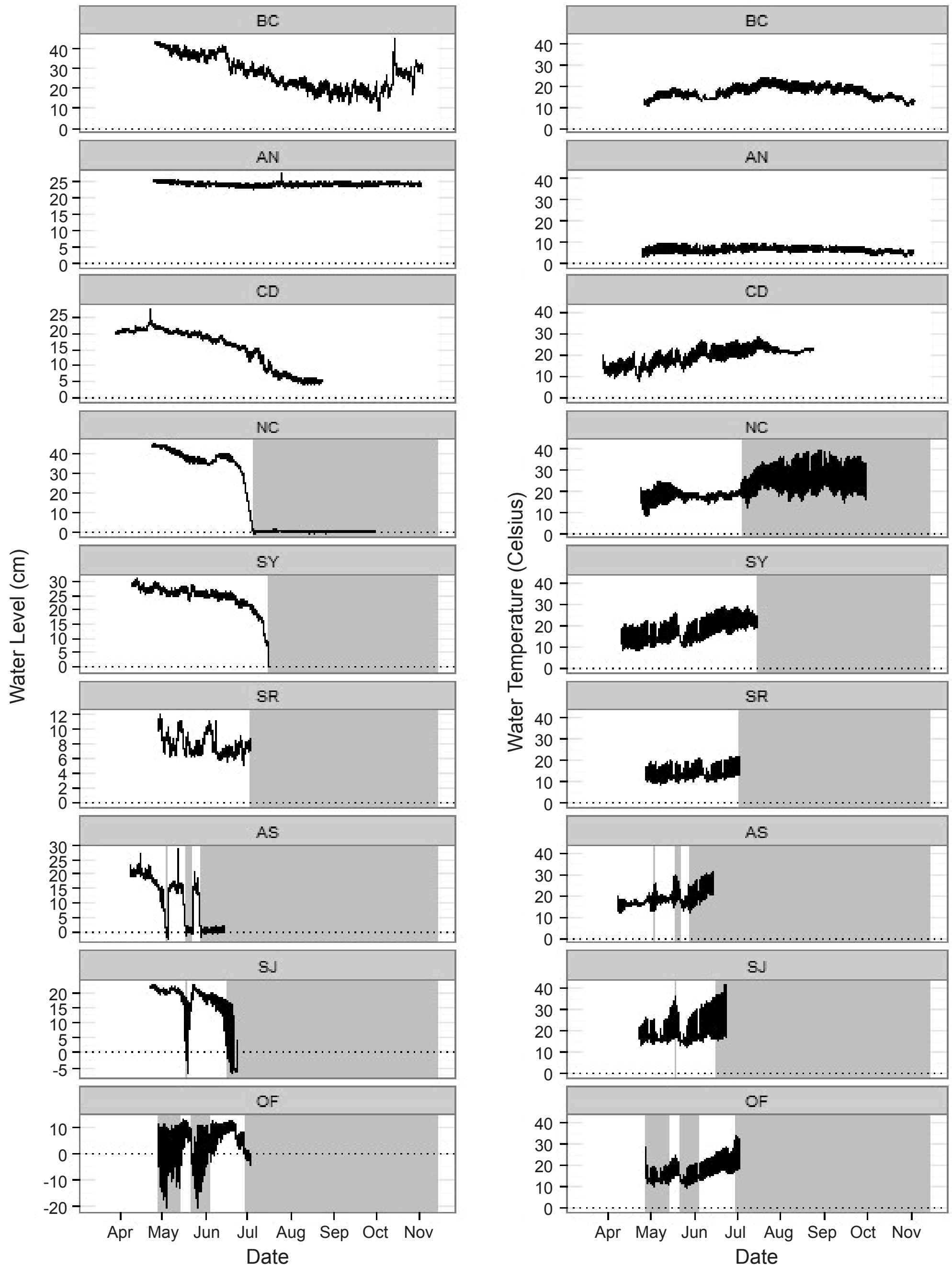

Figure 1. Water surface levels (left) and temperature (right) at a subset of sites. Background color indicates inferred flow status: A white background indicates wet periods, and a gray background indicates dry (or fluctuating) periods. Data for SY, SR, AS, and OF were obtained for 2008. Data for BC, SA, and NC were for 2009. Data for CD were for 2010. Where water levels are greater than 0 and background color is gray (as at sites SJ and OF), surface water is inferred to be present during a period of water level fluctuation.

Examination of scores over time indicated that IBI scores were least variable at low-stress sites, (Figure 3). Within low-stress sites, flow status did not have a large effect on variability (i.e., mean within-site standard deviation was 2.9 at perennial sites and 3.9 at low-stress nonperennial sites, $F = 0.97$, $p = 0.38$). In neither perennial nor nonperennial low-stress sites was a directional trend evident. In contrast, sharp declines were observed at several stressed nonperennial sites (e.g., OF, AS, SJ), and erratic fluctuations at others (e.g., CV, NC). Changes in flow did not show a strong relationship with changes in IBI score (Figure 4).

At two of the sites that were resampled over two years (SR and SY), a mid-spring decline in IBI scores was followed by an early summer increase, and this

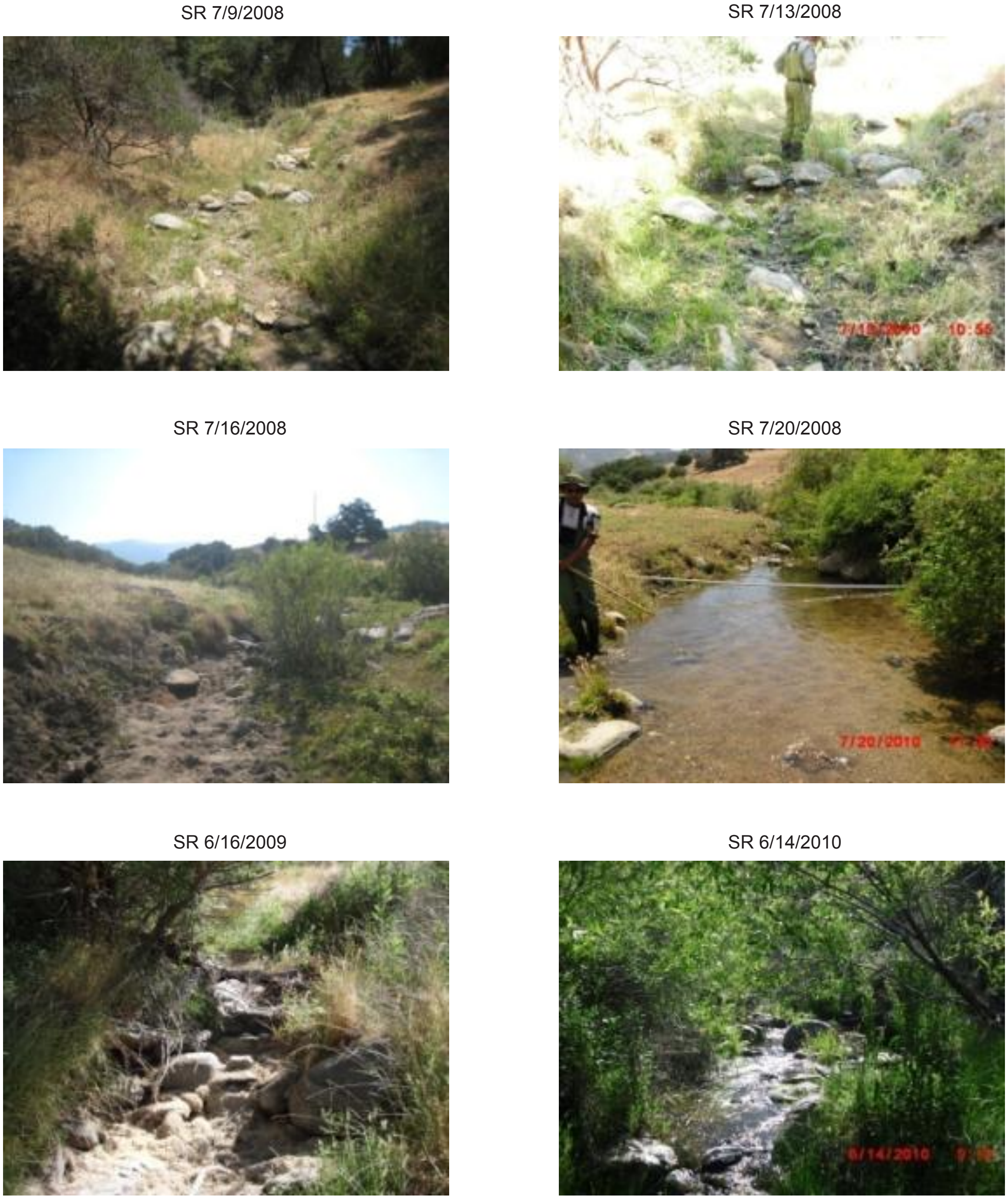

Figure 2. Photos of sites sampled over multiple years. Each photo was taken in the same month of each year at approximately the same location in each reach.

pattern was similar in both years of sampling, although no individual metrics appeared to consistently influence this pattern. The third replicated site (NC) did not display similar patterns in each year, perhaps due to the different hydrologic regimes observed each year (explained below).

Examination of metric scores over time showed that stability in IBI scores arose from patterns that varied across sites (Figure 5). In some cases, stability in the IBI score arose from stability in the underlying metrics (site AN is an extreme example of this stability). In other cases, decreases in some metric scores were offset by increases in others (e.g., site CD). More common, however, were large fluctuations in metric scores yielding smaller fluctuations in the IBI score, as observed at site TE. At sites where the IBI declined (e.g., site OF), metric scores declined in unison near the end of the sampling period.

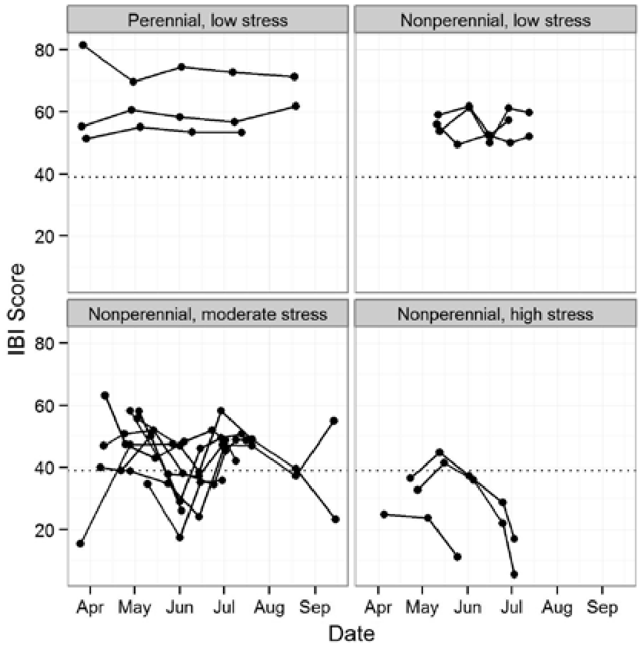

Figure 3. Trends in IBI scores at each site. Each trajectory represents samples from a single site in a single year of sampling. To view trends for individual sites, refer to Supplement 2.

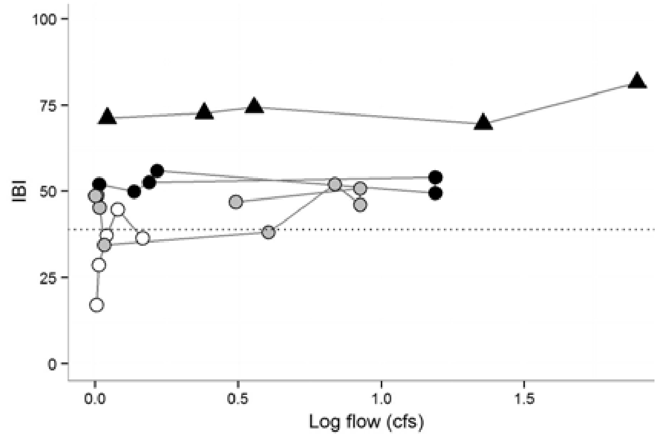

Figure 4. IBI scores versus measured flow. For clarity, only a subset of sites are shown. Black triangles represent BC, a perennial low-stress. Black circles represent AC, a nonperennial low-stress site. Gray circles represent SY (2008 data), a nonperennial moderate-stress site. White circles represent SJ, a nonperennial high-stress site. Trajectories connect samples within sites in order of sampling date.

Table 2. Mean IBI scores for sites in the study.

Site (Stress Score)	Mean IBI	SD	n	n >39
Perennial				
Low Stress	61.1	10.7		
BC (0.0)	73.9	4.6	5	5
AN (0.0)	58.6	2.7	5	5
CD (0.0)	53.3	1.5	4	4
Nonperennial				
Low Stress	55.6	3.1		
TE (0.0)	57.2	5.1	5	5
AC (0.5)	52.0	2.6	6	6
CC (0.5)	57.6	3.9	5	5
Moderate Stress	40.3	6.4		
NC (2.0)	43.0	12.0	10	6
AS (2.5)	38.7	9.8	4	2
SR (3.0)	48.2	8.4	13	12
CV (4.5)	30.8	8.9	5	1
PC (5.0)	36.1	9.4	5	2
SY (7.0)	45.1	9.0	16	12
High Stress	31.4	10.6		
OF (7.5)	27.5	14.2	5	1
SJ (8.0)	32.8	10.5	5	1
TC (8.5)	20.0	5.1	3	0

Trends and Patterns in Community Structure

Nonmetric multidimensional scaling showed that perennial and nonperennial streams did not represent distinct community types, and instead represented extremes along a continuous biological gradient (Figure 6). NMS resulted in a 3-axis solution that represented over 80% of the total variance, with a final stress of 17.8. Axis 3 represented more variance (41%) than the other axes (20 and 19% for Axes 1 and 2, respectively). Visual inspection of this ordination showed a weak segregation of sites by flow status along both Axes 2 and 3. Although this segregation was substantially driven by one site (i.e., BC on Axis 2 and AN on Axis 3), samples from the other two perennial sites were dispersed among the nonperennial sites. Samples also segregated weakly along a stressor gradient, with low-stress sites clustered at the positive ends of Axes 2 and 3, and the negative end of Axis 1.

Plotting weighted averages for species showed that taxa segregated primarily along Axis 3, with mayflies, stoneflies, and caddisflies being more common in sites with high positive values on this axis (Figure 6). Coleoptera occupied a slightly lower position along Axis 3, suggesting a shift from "EPT" taxa to beetles as sites moved down this axis. In contrast, major taxonomic groups were strongly interspersed along Axes 1 and 2. Consistent with this taxonomic segregation, tolerance values were

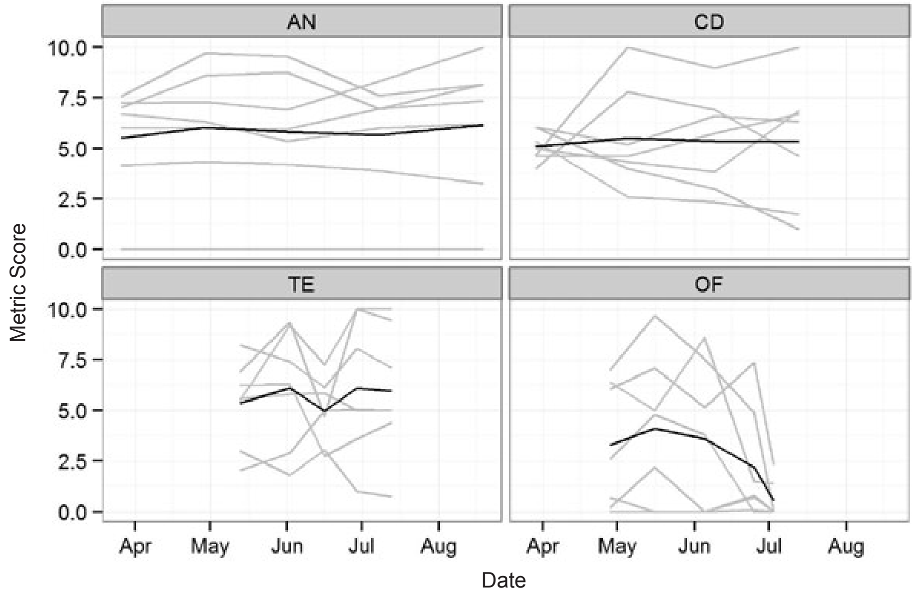

Figure 5. Trends in metric scores at selected sites. Gray lines represent trends in scores for each of the seven metrics in the IBI. The black line represents the mean metric score.

more strongly correlated with species scores along Axis 3 (Spearman's Rho: -0.49) than Axis 1 (Rho: 0.36) or Axis 2 (Rho: -0.9). No segregation of taxa by functional feeding group was evident along any axis.

Replotting trajectories in ordination scores as change from initial sampling revealed that community composition at most sites was either stable or changed in unison (Figure 7). For example, all trajectories were either constant or moved in a positive direction along Axes 1 and 2, or in a negative direction along Axis 3. Thus, if sites changed at all, they changed towards communities more characteristic of nonperennial and high-stress sites, with fewer "EPT" taxa and more Coleoptera, Diptera, and non-insects. In general, trajectories were shortest for perennial and low-stress nonperennial sites, and longest for high-stress nonperennial sites.

Relationships between Biological and Environmental Variables

Many environmental variables were associated with biological differences among sites, but few were associated with differences within sites (Figure 8; Table 3). For example, of the 28 environmental variables evaluated, 11 had strong relationships (|Rho| greater than 0.5) with IBI scores, as did 16 with at least one of the three ordination axes. For IBI scores, the strongest relationships were observed for stressor scores (Rho = -0.91), a few habitat (e.g., % fast-water habitat: Rho = 0.75; % shading: Rho = 0.68), and specific conductance (Rho = -0.69). For ordination axes, stressor scores were less important (i.e., strongest Rho was -0.53, with Axis 2), but a large number of habitat variables showed strong relationships with site differences, particularly those related to substrate (e.g., % cobble embeddedness: Rho = -0.85 with Axis 3), hydrology (e.g., % fast water habitat: Rho = 0.70 with Axis 2), and riparian vegetation (e.g., mean riparian vegetation cover: Rho = 0.70 with Axis 3). Specific conductance also had a strong relationship with ordination axes (e.g., Rho = 0.65 with Axis 3).

In contrast, few environmental variables were associated with within-site biological changes, and only two were associated with IBI scores a |Rho| greater than 0.2 (temperature: -0.31; wetted width: 0.27). Relationships with ordination axes were stronger, but only 4 of the 22 variables considered had |Rho| greater than 0.5. The strongest relationships were for variables related to water quality (e.g., temperature: Rho = 0.66 with Axis 2; dissolved oxygen: Rho = 0.57 with Axis 3) and hydrology (e.g., Flow: Rho = -0.57 with Axis 1, wetted width: Rho = -0.67 with Axis 2). Physical habitat variables not directly related to water availability had much weaker relationships with within-site biological changes; among these variables, the strongest relationships were observed for

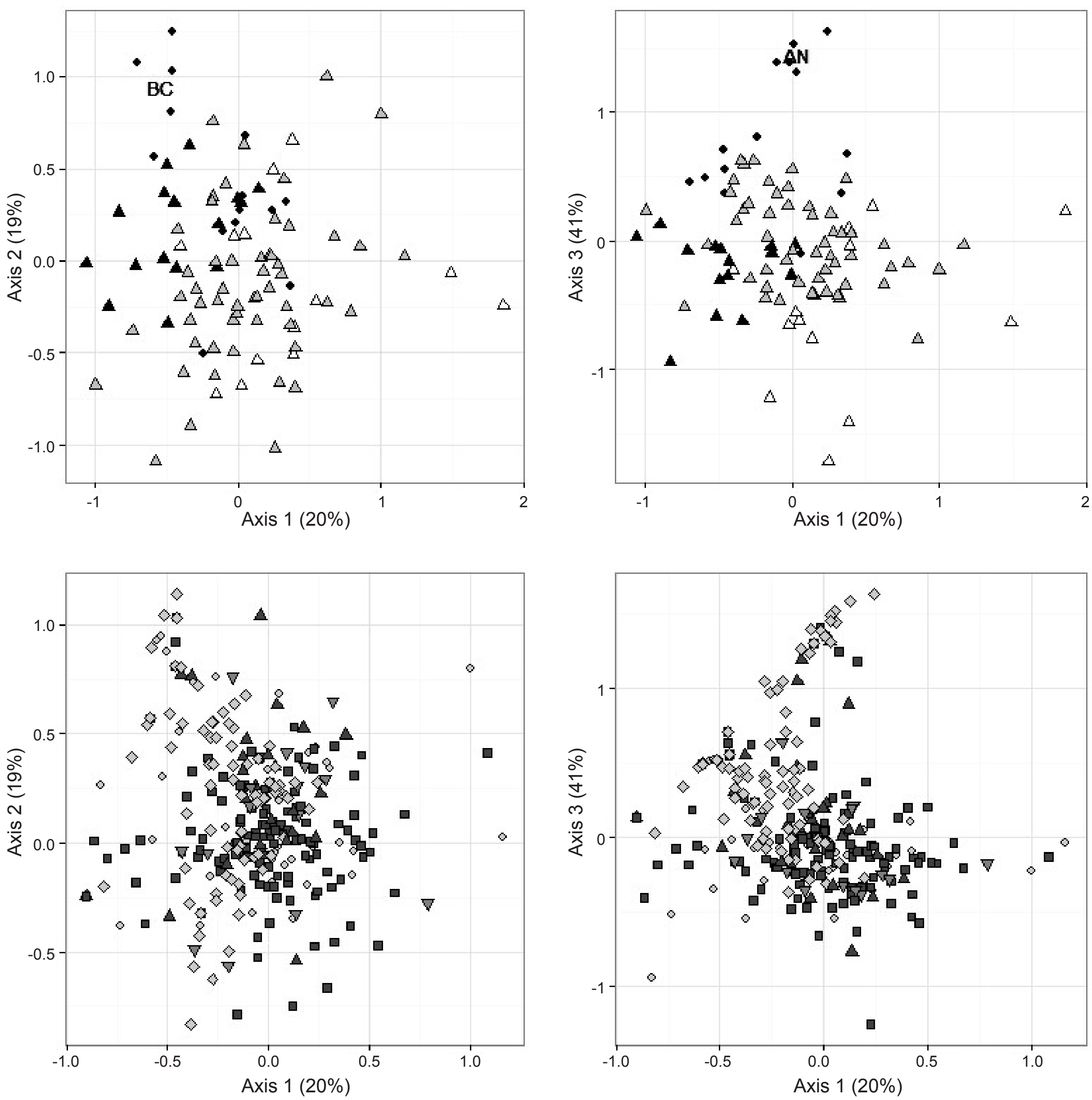

Figure 6. Results of an NMS ordination of presence-absence data. Top row: Ordination scores for each sample in the study. Samples from perennial sites are represented as circles, and samples from nonperennial sites are represented as triangles. Black symbols indicate low-stress sites, gray symbols indicate moderate-stress sites, and white symbols indicate high-stress sites. Centroids for two sites referenced in the text (AN and BC) are annotated. Numbers in axis labels show the percent of the total variance represented in the axis. Bottom row: weighted average scores for taxa included in the ordination. Each symbol represents a different taxon: White circles are Coleoptera; black squares are Diptera; gray diamonds are Ephemeroptera, Plecoptera, and Trichoptera; gray down-triangles are other insect orders; and black up-triangles are non-insects.

% sands and fines (Rho = -0.32 with Axis 1) and % shading (Rho = 0.43 with Axis 2). The strength and sometimes even the direction of these relationships varied from site to site. For example, although within-site correlations between IBI scores and temperature were negative for most sites, Rho was positive at TE (Rho = 0.1), and strongly positive at NC (Rho = 1.0).

Discussion

As this study demonstrates, the wholesale exclusion of nonperennial streams from bioassessment programs is not justified, despite their biological differences from perennial streams. First, the assessment index used in this study correctly assessed the condition of low-stress nonperennial streams.

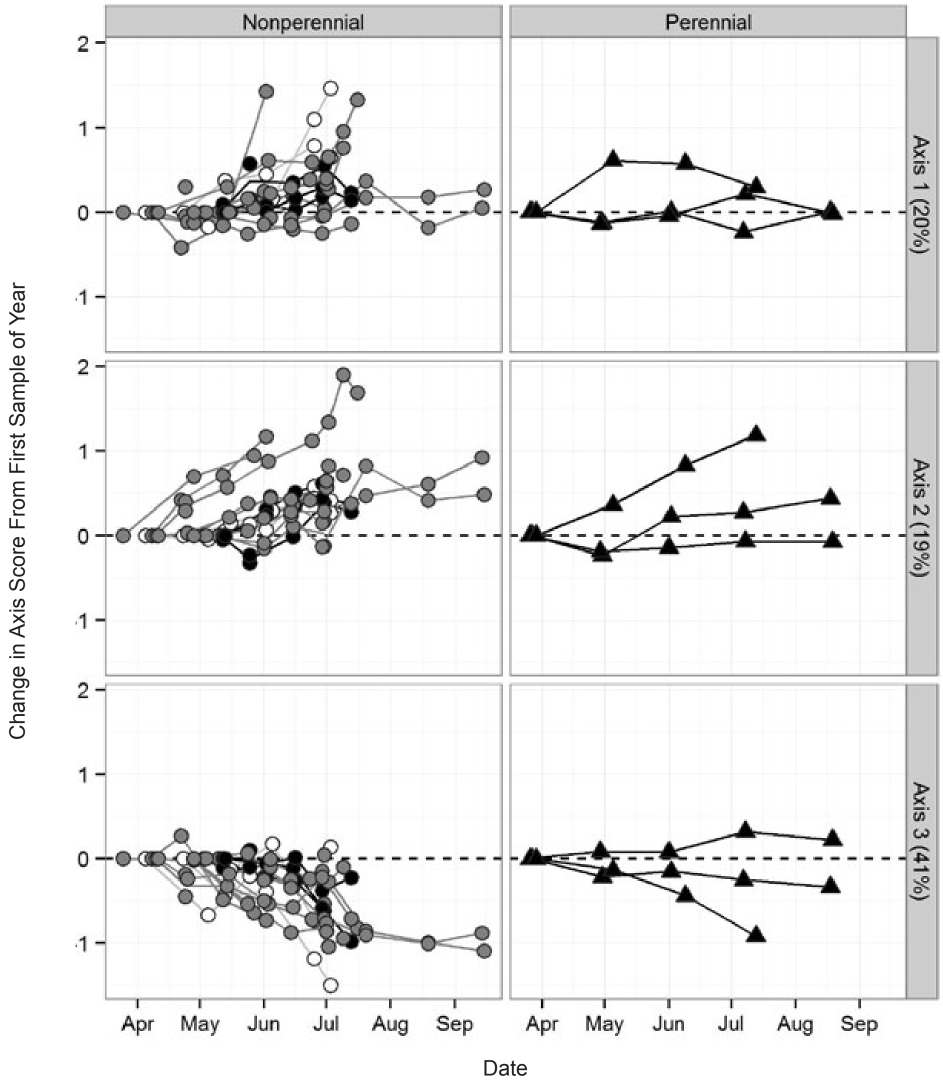

Figure 7. Trajectories in ordination space for each site and year plotted against time. In each panel, scores for the initial sampling event were subtracted from all subsequent samples at a site and year. Triangles represent perennial sites and circles represent nonperennial sites. White symbols indicate high-stress sites. Gray symbols indicate moderate-stress sites. Black symbols indicate low-stress sites.

Second, scores were consistent over time, at least at low-stress sites. And third, although seasonal (within-site) changes affected biological community structure, they did not affect assessment scores. Therefore, biomonitoring programs may be able to integrate nonperennial streams and assess them using some of the same multimetric indices used in perennial streams. Study results indicate that two factors contribute to the comparability of bioassessments observed here: first, the regional fauna may be adapted to nonperennial flow regimes; second, the relative stability and predictability of nonperennial streams in this study increases the biological similarity of perennial and nonperennial streams.

Assessment Tools Work in Nonperennial Streams

Although nonperennial streams are widely described as supporting biological communities that are distinct from those found in perennial streams (e.g., Álvarez and Pardo 2007, Datry 2011, Bogan *et al.* 2013), fauna from the streams in this study appear to occupy different positions along a continuous gradient, rather than discrete community types. For example, samples from perennial and nonperennial streams were interspersed in ordination space, emphasizing the overall similarity of these stream types. The relative similarity of perennial and nonperennial streams observed in this study contrasts with several other studies (e.g., Arscott *et al.* 2010).

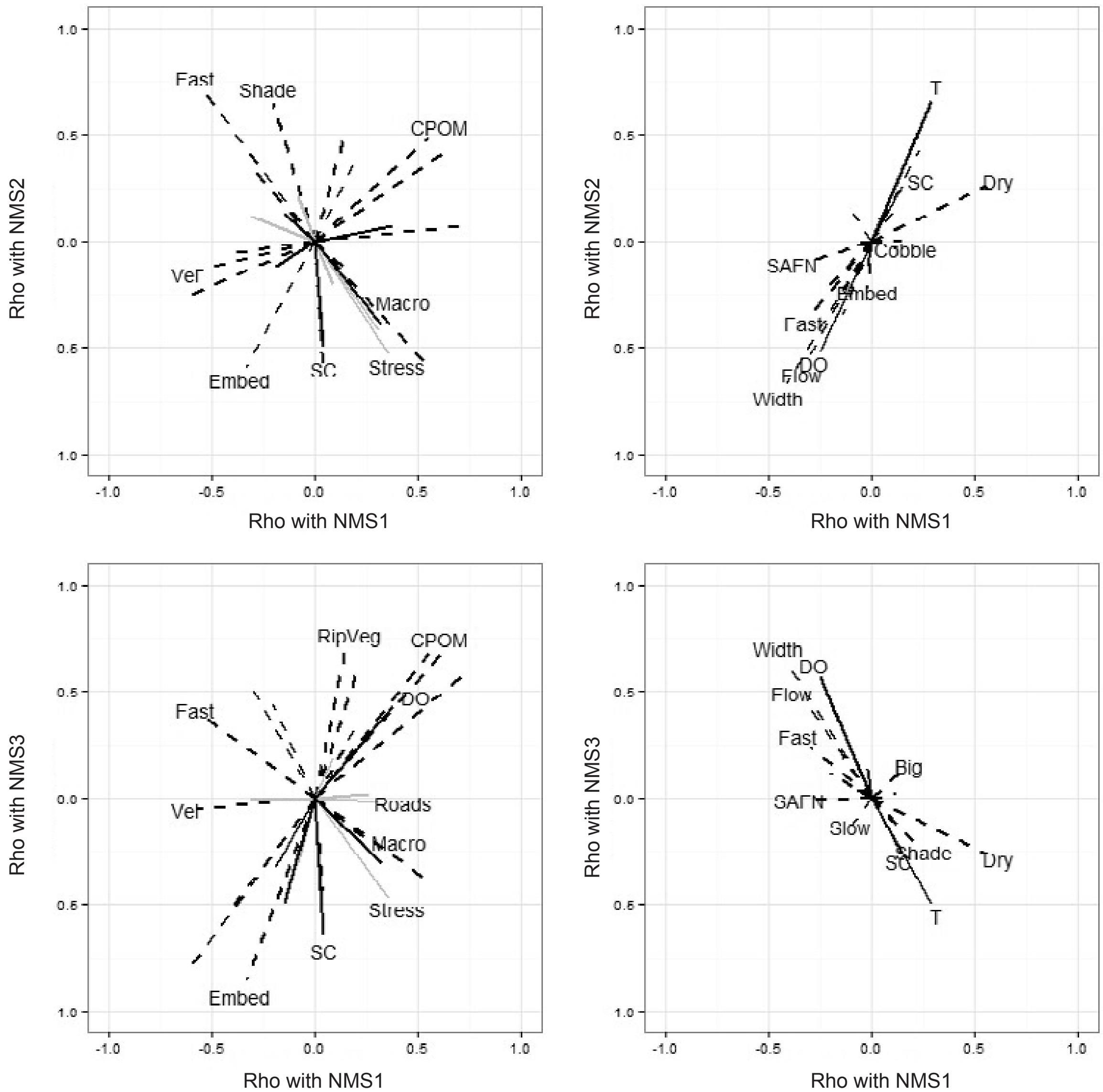

Figure 8. Relationships between environmental variables and ordination axes. Top row: Among-site correlations (Rho), calculated from within-site means. Bottom row: Within-site correlations (Rho), calculated from variables minus within-site means. Length of each line represents the strength of correlation (Rho) with the ordination axes. Solid black lines represent water quality variables; dashed black lines represent habitat variables; and solid gray lines (top row only) represent landscape- or site-based variables. Selected variables are labeled. Fast: % fast-water habitat. Slow: % slow-water habitat. Shade: % shading. CPOM: % cover by coarse particulate organic matter. Vel: Mean water velocity. Flow: Discharge. Macro: % macroalgae cover. Embed: Mean embeddedness. SC: Specific conductance. T: Water temperature. Stress: Stressor score. RipVeg: Mean riparian vegetation cover. Roads: Road density. Dry: % dry habitat. Big: % particles larger than cobble. SAFN: % sands and fines. Cobble: % cobbles. Width: mean wetted width. DO: Dissolved oxygen.

However, many of these studies had limited spatial (e.g., Bêche *et al.* 2006) or temporal (e.g., Lunde *et al.* 2013) replication, and these limits alone could exaggerate the apparent distinctness of nonperennial streams. These findings are supported by those of Gallart *et al.* (2012), who suggest that ecological status may be assessed in the same way at temporary streams as at perennial streams if flow permanence and seasonal predictability are relatively high.

The comparability of bioassessments between perennial and nonperennial streams may be limited to regions like southern California, where nonperennial streams are the dominant stream type. Benthic macroinvertebates collected at a site represent a subset of the regional fauna that can tolerate the local environmental conditions (Southwood 1977, Townsend and Hildrew 1994, Statzner *et al.* 1997). In predominantly arid regions, taxa that are

Table 3. Among- and within-site correlations (Spearman's Rho) between environmental and biological variables.

Variable	Among Sites				Within Sites			
	IBI	NMS1	NMS2	NMS3	IBI	NMS1	NMS2	NMS3
Stressor Score	-0.91	0.36	-0.53	-0.47	NA	NA	NA	NA
Landscape								
Latitude	0.01	-0.31	0.12	-0.01	NA	NA	NA	NA
Longitude	0.03	0.09	-0.20	0.19	NA	NA	NA	NA
Catchment Area	-0.20	-0.08	0.21	-0.30	NA	NA	NA	NA
% Impervious Surface	-0.59	0.26	-0.32	0.02	NA	NA	NA	NA
Road Density	-0.59	0.31	-0.42	-0.02	NA	NA	NA	NA
Physical Habitat								
Algae cover								
% Algae Cover	-0.31	0.24	-0.26	-0.19	-0.06	-0.20	-0.20	0.12
% Macrophyte Cover	0.14	0.71	0.07	0.57	0.11	0.10	0.14	-0.11
Natural Habitat Cover Index	0.34	0.61	0.41	0.68	0.12	-0.06	-0.01	0.08
Hydrology								
Flow	0.54	-0.32	0.36	0.54	0.15	-0.32	-0.57	0.45
Velocity	0.20	-0.57	-0.14	-0.05	0.18	-0.14	-0.34	0.19
% Dry Habitat	-0.61	0.04	-0.62	-0.44	-0.12	0.56	0.26	-0.26
% Fast-Water Habitat	0.75	-0.53	0.70	0.38	0.07	-0.30	-0.35	0.24
% Slow-Water Habitat	-0.69	0.56	-0.6	-0.40	0.03	-0.09	0.13	-0.12
Wetted Width	0.07	0.20	0.04	0.34	0.27	-0.41	-0.67	0.64
Riparian Vegetation								
% Shading	0.68	-0.20	0.65	0.44	-0.03	0.23	0.43	-0.23
Mean Riparian Vegetation Cover	0.55	0.15	0.52	0.70	0.01	0.06	0.08	-0.04
% Reach with 3 Vegetation Layers	-0.36	-0.60	-0.25	-0.79	-0.14	0.04	0.12	-0.06
Substrate								
% Large Substrates	0.43	0.20	0.40	0.59	0.05	0.15	0.00	0.13
% Cobble	-0.02	0.17	-0.15	0.27	-0.15	0.12	-0.03	0.02
% CPOM Cover	0.39	0.55	0.49	0.68	-0.02	-0.02	-0.06	0.09
% Sands and Fines	-0.02	-0.39	-0.05	-0.50	-0.07	-0.32	-0.10	-0.01
Mean Embeddedness	-0.48	-0.33	-0.59	-0.85	0.11	-0.02	-0.22	0.02
Water Chemistry								
Alkalinity	0.04	-0.15	0.13	-0.49	0.16	-0.07	-0.02	0.04
Dissolved Oxygen	-0.14	0.38	0.07	0.43	0.01	-0.25	-0.52	0.57
pH	-0.20	-0.19	-0.12	-0.32	-0.16	-0.02	-0.12	0.14
Specific Conductance	-0.69	0.04	-0.50	-0.65	-0.02	0.15	0.26	-0.27
Temperature	-0.51	0.32	-0.39	-0.31	-0.31	0.29	0.66	-0.50

dependent on perennial streams may be excluded from the regional fauna because of the scarcity of habitat (e.g., only 500 km of streams in the San Diego region are perennial, National Hydrography Dataset Plus, www.horizon-systems.com/nhdplus); therefore, the community composition at perennial streams in arid regions may be constrained to be more similar to nonperennial streams. Additionally, the temporal variability in flow status, as observed in this study and elsewhere (e.g., Gasith and Resh 1999), may further limit the regional fauna to taxa adapted to intermittent flow. The richer regional fauna of wetter regions may allow greater divergence between perennial and nonperennial streams. The findings of Arscott *et al.* (2010) support this theory. They observed that communities in intermittent and ephemeral streams in New Zealand were nested subsets of communities found in nearby perennial streams, and reflect a loss of desiccation-sensitive taxa, rather than a gain of desiccation-tolerant taxa.

Additionally, the similarities between perennial and nonperennial streams observed here may be restricted to the types of nonperennial streams included in the study—specifically, long-lasting, seasonal streams that flow for several months nearly every year. Less predictable or more ephemeral streams may support more different biological communities from perennial streams than what was observed in this study. Looking just within nonperennial streams, Anna *et al.* (2008) found large differences in bioassessment indices between ephemeral and intermittent streams. Similarly, Gallart *et al.* (2012) found that unpredictable hydrologic regimes decreased the measured ecological status of temporary streams. Therefore, the applicability of bioassessment tools developed from perennial streams may only provide comparable interpretations at nonperennial streams with sufficient relative flow permanence. Determination of critical thresholds in flow permanence or predictability should be a focus of further research.

Assessment Scores are Consistent over Time

Although multivariate analyses showed large seasonal changes in community composition at most sites, IBI scores were relatively stable, at least at low-stress sites. This stability was particularly evident at perennial sites, where all samples ranged ~5 points at a single site. The consistency was somewhat less pronounced at the low-stress nonperennial sites (with typical ranges of ~10 points). However, at moderate and high stressed nonperennial sites, IBI scores were noticeably unstable, and showed steep declines at several sites, particularly at the end of sampling. Ranges at these sites even exceeded 40 points on the 100-point scale of the IBI. Examining unstressed intermittent streams in Spain, Sánchez-Montoya *et al.* (2009b) similarly found low intra-annual variability in multimetric indices, as well as in certain metric types, such as those derived from life history traits, further suggesting that benthic communities in undisturbed nonperennial streams are relatively resilient to seasonal changes.

Stability in IBI scores at nonperennial sites also extended to interannual replicates, at least at two of the three sites selected for multi-year sampling, a finding consistent with the observations of others (e.g., Morais *et al.* 2004, Sánchez-Montoya *et al.* 2009b). Despite the different weather patterns over the years of study, these two sites exhibited similar patterns in IBI scores as well as trajectories in ordination space. The stability in IBI scores at low-stress sites may result from the use of a broad index period (April to October) in the development of the IBI (Ode *et al.* 2005). Thus, the IBI appears to integrate the temporal variability evident in the ordinations and creates a stable measurement of structural and functional attributes of the benthic macroinvertebrate assemblage. This study suggests that a common assumption underlying the development of many bioassessment indices—the substitution of space for time during index development—may be effective in broadening their applicability (Stoddard *et al.* 2008, Schoolmaster *et al.* 2012). That is, indices calibrated from data covering many sites but only a few years may still be resilient to temporal changes. A large number of reference sites is likely to sample streams at many different stages in their individual pheonologies, which may produce similar biological variability that would result from repeated visits, thus producing resilience to temporal variability in a bioassessment index.

Few Changes in the Environment were Associated with Trends in Biology

Given the overall stability of IBI scores, it is not surprising that few environmental variables were related to within-site changes in scores. Moreover, only a few variables were associated with changes in ordination scores. Unsurprisingly, these variables were mostly related to water quantity or quality (e.g., velocity, % fast water habitat, temperature, dissolved oxygen); relationships with variables related to physical structure (such as substrate composition), primary productivity, and riparian vegetation were weaker. Other studies of temporary streams in Mediterranean-climate regions have found similar relationships between these variables and benthic macroinvertebrate community structure (e.g., Morais *et al.* 2004, Álvarez and Pardo 2007. Garcia-Roger *et al.* (2011) also found that most changes in habitat were site-specific, although those related to quantity of aquatic habitat were strongly associated with changes in community structure. In contrast, among-site differences were stronger and associated with a larger variety of environmental variables, consistent with many studies on stream ecosystems both temporary (e.g., Álvarez and Pardo 2007) and perennial (e.g., Sandin and Johnson 2004, Mazor *et al.* 2006, Yuan *et al.* 2008).

One of the more surprising findings of this study was the apparently high sensitivity of nonperennial streams to non-natural flow regimes. For example, several sites that had very few disturbances apart from altered hydrology (e.g., AS, NC) nonetheless had relatively low IBI scores, and the lowest scores were observed in sites with altered hydrology combined with other stressors (e.g., TC, OF, and SJ). In contrast, sites with substantially higher stress levels but relatively stable hydrographs (e.g., SY, SR) had much higher IBI scores. Skoulikidis *et al.* (2011) also found that biological assemblages were particularly sensitive to modified flow regimes and artificial drying, and recommended distinguishing naturally from artificially nonperennial streams for assessment purposes. Therefore, watershed managers should take care to monitor alterations to hydrologic regimes in nonperennial streams as much as they do for perennial streams.

Implications for Bioassessment Programs

Although limited to a small number of sites, this study illustrates that certain nonperennial streams can be incorporated into routine bioassessment programs. Hydrologically stable and predictable nonperennial streams that are sampled during eurheic or oligorheic states are biologically similar to perennial streams, at least within arid regions like southern California. Expanding water quality assessment programs to include nonperennial streams would give resource managers the ability to manage a greater extent of their streams, and also address impacts to some of the most sensitive portions of their watersheds. Although these changes may be most profound in arid regions, like southern California, the global ubiquity of nonperennial streams (Tooth 2000) suggests that watershed protection in both wet and dry climates can be greatly improved by including nonperennial streams in assessment programs.

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Supplemental Information

Supplemental Information is available at ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/AnnualReports/2013AnnualReport/ar13_357_375SI.pdf.