LRH: J. Van Sickle and R.M. Hughes
RRH: Classification strengths for aquatic vertebrates

Classification strengths of ecoregions, catchments and geographic clusters for aquatic vertebrates in Oregon

John Van Sickle[1]

US Environmental Protection Agency, Western Ecology Division, National Health and Environmental Effects Research Laboratory, 200 SW 35th St., Corvallis, Oregon 97333 USA

Robert M. Hughes

Dynamac, Inc., 200 SW 35th St., Corvallis, Oregon 97333 USA

Journal of the North American Benthological Society, 2000, 19(3):370-384

[1] E-mail address: VanSickle.John@epa.gov

Abstract. The usefulness of ecoregions and catchments (hydrologic units) as bases for classifying aquatic vertebrate assemblages in western Oregon were compared using samples collected by electrofishing from 137 wadeable stream sites distributed evenly throughout the region. The classification strengths of these regionalizations were also compared with neutral-model classifications that were based either on intersite proximities in geographic distance or on intersite similarities in the sampled vertebrate assemblages. The strength of each classification was assessed by the extent to which average within-class assemblage similarities exceeded the average similarity between classes. Mean similarity dendrograms were used as a concise graphical comparison of between- and within-class similarities for alternative classifications, whether they were specified a priori or constructed by clustering. For each a priori classification, a permutation test of the *no class structure* hypothesis was performed. Classification strengths were assessed using the Sorenson-Dice (presence/absence) and Bray-Curtis (relative abundance) similarity measures, applied to both species-level and family-level assemblage characterizations, as well as for the Bray-Curtis measure applied to a set of 5 assemblage metrics that were designed to reflect stream impairment. For all 5 measures of assemblage similarity, ecoregions had higher classification strengths than did large catchments, and large catchments had about the same strength as a stream-order classification. A catchment classification with 1 of the 3 catchments split into 2 ecoregions separated assemblages as strongly as ecoregions alone. A neutral-model classification based solely on geographic site proximity classified assemblages with about the same strength as ecoregions. Another neutral-model classification of sites, based solely on their sampled assemblages, was at least twice as strong as any of the geographic classifications. Intermediate strength was seen in site groupings derived from a composite measure of between-site assemblage and geographic dissimilarities. Our results suggest that ecoregions and large catchments do indeed have utility for classifying stream vertebrate assemblages. However much of their classification strength may be a result of spatial autocorrelation effects, rather than ecological factors that determine their particular boundaries. Our similarity analyses also suggest that geographic partitions can be expected to account for only a minor portion of the total variation seen in stream vertebrate assemblages across a large region.

Key words: streams, fish, amphibians, assemblages, communities, ecoregions, drainage basins, classification, western Oregon, cluster analysis, dendrogram, similarity.

Geographic classification has become an integral part of efforts to study, monitor and manage ecosystems regionally. The partitioning of a large area into relatively homogeneous regions provides spatial order and structure for assessing complex patterns of ecosystem variation across a broad range of spatial scales (Huang and Ferng 1990). For example, multimetric indices that measure impacts on lake and stream biota can be applied across large areas because they incorporate reference conditions that strongly differ by regions, usually ecoregions (Omernik 1987), hydrologic units, or large catchments (Fausch et al. 1990, Davis and Simon 1995, Gerritson 1995). Ecoregions and large catchments are also useful for extrapolating biological conditions when ecological data are sparsely and unevenly distributed over space.

Because large catchments and ecoregions are usually used as multiple-purpose classifications (Omernik and Bailey 1997), they are unlikely to provide optimal separation for any single ecosystem component, such as a faunal assemblage (Sokal 1974). However, there is a clear, practical interest in assessing geographic partitions for their within-region homogeneity and between-region differentiation of 1 or more key ecosystem components. Such assessments aid us in deciding whether geographical partitions are at all helpful in modeling large-scale spatial variation of individual components and, if so, which alternatives are most helpful.

Ecoregions and large catchments are rarely congruent, so they usually offer distinctly different geographic frameworks for classifying stream ecosystems. Both frameworks are strongly grounded in ecological principles. Ecoregion boundaries are based on land forms, potential vegetation, soils, and land use, and are intended to delineate regions containing similar ecosystems (Omernik 1987, Omernik and Bailey 1997). As a consequence, ecoregions have been advocated as natural geographic units for aquatic ecosystem management, research, and assessment (Omernik and Griffith 1991). Evidence for the classificatory value of ecoregions has been provided by numerous assessments of between-region differences in aquatic system attributes such as water quality parameters and assemblages of aquatic vertebrates or invertebrates (Hughes et al. 1994). However, for aquatic vertebrates, one might also expect an increased similarity of assemblages within large catchments, because of the greater probabilities of within-catchment, as opposed to between-catchment, migration (Hocutt and Wiley 1986, Hughes et al. 1987, Matthews 1998). Omernik and Griffith (1991) and Omernik and Bailey (1997) have clarified the broad circumstances under which either or both of these 2 regional frameworks might be more suitable for particular regions, ecosystems, and classification objectives, but few direct quantitative comparisons of the 2 approaches are available across large areas.

We define and apply a measure of classification strength that is a function of within-class homogeneity and between-class separation. We then use this measure to compare the classification strengths, for aquatic vertebrate assemblages (fish and amphibians) of ecoregional and hydrologic-unit partitions of western Oregon (Fig.1).

Methods

Alternative stream classifications

There are a multitude of schemes, both geographical and nongeographical, that one might use to classify streams (Naiman et al. 1992). Here we explored several alternatives to western Oregon’s ecoregions and hydrologic units, chosen to provide a context for comparing these 2 geographic schemes. For the 1st alternative, we subdivided 1 of our 3 hydrologic units into 2 ecoregions, in an effort to combine the strengths of the 2 classification systems (Omernik and Bailey 1997). We also classified sites by Strahler stream order, a key nongeographic feature that is often correlated with fish assemblage structure (Kuehne 1962, Platts 1979, Beecher et al. 1988). As a 3rd alternative, we grouped sites according to the 3 separate sampling efforts that were undertaken to collect our assemblage data, to see whether this factor might confound our comparisons of regional partitions.

We also constructed neutral-model classifications that were designed to compete with ecoregions and hydrologic units for classifying stream vertebrates. Neutral-model classifications are constructed using site-grouping rules that deliberately ignore the geographic or ecological factors underlying ecoregions and hydrologic units (Caswell 1976, Gardner et al. 1987). If the neutral model explains patterns in a data set as well as they can be explained by ecoregions or hydrologic units, then that data set gives little support for the classification utility of the underlying ecological factors (Caswell 1976).

The simplest neutral model for assessing any classification is the null hypothesis of no class structure, which is based on a completely random assignment of sites to groups, regardless of their location or other attributes. We carried out statistical tests of our candidate classifications against this null hypothesis to assess their baseline classification utility.

We constructed a more challenging neutral model for ecoregions and hydrologic units by clustering sites into 4 groups based solely on intersite geographic distances. These distance clusters yielded 4 contiguous, spatially compact regions, albeit with imprecise boundaries, at the same spatial scale as ecoregions and hydrologic units. If the distance clusters classify assemblages as strongly as do ecoregions and hydrologic units, then the classification strengths of the latter could arise largely from spatial autocorrelation effects at subregional scales, rather than from strong ecological separability conferred by regional boundaries. Jenerette et al. (1998) created a similar neutral model by generating sets of circular, randomly located regions at a variety of scales. They showed that these regions, on average, had within-region homogeneity in water quality attributes that was as great as that seen in several proposed regionalizations, including ecoregions and catchments. Legendre et al. (1990) also discuss neutral models for regional evaluations, and present alternative algorithms for their generation.

We also constructed a classification model that was spatially neutral. Using cluster analysis, sites were assigned to 1 of 4 groups based on the compositional dissimilarities of their vertebrate assemblages, without regard to site location. Because these groups were constructed to have minimum within-group variation and maximum between-group separability for vertebrate assemblages, their classification strengths as assessed by those same assemblages were near the maximum achievable values for any 4-group classification of our data set. Thus, the vertebrate assemblage clusters give an upper bound to the classification strength that could be expected from any geographical grouping of the sites. The use of direct assemblage clustering, rather than geographic regions, to classify sites is a key feature differentiating multivariate impact assessment approaches (Wright et al. 1984, Reynoldson et al. 1995, Parsons and Norris 1996) from multimetric approaches.

As a final neutral-model classification, we used cluster analysis to generate composite groups based on a combination of intersite geographical distance and assemblage dissimilarity, thus yielding spatially proximate site groups with similar vertebrate assemblages. Legendre and

Legendre (1984) and Hawkes et al. (1986) give other applications of the general approach. These composite groups may be viewed as a data-driven attempt to construct a special-purpose set of partially overlapping regions that is optimal only for vertebrates, as an alternative to general-purpose geographic partitions such as ecoregions, hydrologic units, and stream order. We would expect to see the composite groups classify vertebrate assemblages more strongly than ecoregions and hydrologic units, but the general-purpose regions are hypothesized to be stronger, on average, across multiple characteristics of aquatic ecosystems (Hughes at al. 1994, Omernik and Bailey 1997). In summary, we compared 8 different classification schemes for western Oregon's aquatic vertebrate assemblages: 1) ecoregions, 2) hydrologic units, 3) a combined hydrologic unit/ecoregion partition, 4) stream order, 5) sampling effort, 6) a geographic cluster model that is neutral for hydrologic and ecoregional boundaries, 7) a vertebrate assemblage cluster model that is spatially neutral, and 8) a composite of 6 and 7.

Study region and vertebrate assemblage data

We used aquatic vertebrate assemblage data collected from 137 wadeable streams (Strahler orders 1-3, on 1:100,000 scale maps) in western Oregon (Fig. 1) during summer baseflow conditions. The data set was assembled from 3 separate sampling efforts: an Environmental Monitoring and Assessment Program (EMAP) study (57 sites in 1997), a Regional EMAP study of the Oregon and Washington Coast Range (49 sites in 1996), and the Oregon State University pilot study (31 sites in 1996; Herlihy et al. 1997).

Each of the 3 sampling efforts selected sites at random from all mapped stream kilometers in the study region, resulting in a fairly even spatial distribution of sites (Fig.1). The sites are distributed throughout the 4 Level III Ecoregions (Omernik 1987; Fig. 1). To define hydrologic units, we used the 3 Hydrologic Accounting Units (HAUs; Seaber et al. 1987), that compose Oregon west of the Cascade Mountains crest (Fig. 1). Of the 3 HAUs, only the Willamette Basin can be viewed as a single connected catchment (Fig.1). However, the 2 Coastal HAUs do represent sets of smaller catchments having greater probability of migration within sets than between sets. In addition, the 3 HAUs are commonly used by state agencies as management and planning units.

For our HAU/ecoregion classification, we split the Willamette Basin HAU (Fig. 1) into a Willamette Valley ecoregion component (agricultural lowlands), and a Cascades ecoregion component (forested uplands). These 2 regions, plus the North Coast and South Coast HAUs, gave a 4-region partition of western Oregon.

The 137 sampled streams span a full range of human impacts and few can be regarded as "reference" sites (sensu Hughes 1995). Thus, assemblage variability across these sites is a result of some combination of natural variation and human impacts. Because of the large size and topographic diversity of western Oregon, with its valleys and mountains, we believe that natural gradients are at least as important as anthropogenic ones in structuring spatial variation of stream vertebrates. For example, Rathert et al. (1999) found that climatic and hydrologic variables could account for > 60% of the variation in fish species richness across the state of Oregon.

Aquatic vertebrate assemblages at all sites were sampled with the same protocol (McCormick and Hughes 1998). Each site was electrofished by 2 persons for a distance 40 times the mean wetted width of the channel or a minimum of 150 m; this level of effort produced repeatable estimates of species richness in Oregon streams (Hughes et al. 1998) and elsewhere (Lyons 1992, Angermeier and Smogor 1995, Paller 1995). Battery powered Smith-Root backpack electrofishers were set at 50-60 Hz and 700-1000 V, and each site was fished for 2 to 3 h in an afternoon. Both native and alien fishes and aquatic amphibians were collected and analyzed because both are important top carnivores in small streams. There are few native fish species in coldwater streams, and amphibians tend to replace fishes in the headwaters (Moyle and Marchetti 1998).

Assemblage similarity measures

For every pair of sites, we computed 5 similarity measures. Sorenson-Dice similarity is the ratio between the number of taxa common to 2 sites and the average number of taxa per site that was found at the 2 sites; the measure can be interpreted as the proportion of the assemblages found at 2 sites that are shared by the sites. Bray-Curtis similarity is analogously computed from proportional, untransformed abundances of taxa at 2 sites, and can be interpreted as the shared relative abundance at 2 sites (Digby and Kempton 1987, Ludwig and Reynolds 1988). Both similarities lie in the range from 0 to 1, and here we express them as percentages. We computed both similarities for family-level and again for species-level identifications of the sampled vertebrates, resulting in 4 measures of taxonomically based similarity.

For each assemblage we also computed a set of metrics that are probable indicators of human impacts : % coldwater individuals, % coolwater individuals, % alien individuals, % tolerant individuals, and % anadromous species. We used only these 5 metrics because our small streams often supported only ≤5 species, unlike the more speciose midwestern streams having 20-30 species and commonly characterized by 10 to 12 metrics. We expressed all metrics as percentage of species or individuals in the total collection to avoid having to adjust assemblage size for catchment area (Fausch et al. 1984). Two metrics, % coldwater individuals (salmonids and some sculpins) and % anadromous species (most salmonids and a lamprey) were selected to reflect higher-gradient and minimally impaired conditions. Both tend to decline as streams warm or are modified by dams and physical habitat degradation. At the other end of the impairment gradient, % alien individuals and % tolerant individuals increase with anthropogenic disturbances that increase water temperature and sedimentation; % aliens also directly indicates the effects of stocking nonnative species. Aliens include sunfishes, bullheads, and brook trout; tolerants include sunfishes, bullheads, and a few native minnows. We also used % coolwater individuals (mostly lampreys, sculpins, and native minnows) to reflect moderate levels of impairment. Although our objective was not site assessment per se, we wanted to ensure that we did not bias our site classifications by evaluating patterns evident from selecting metrics sensitive at only 1 impairment extreme. To express beween-site similarities for the set of 5 metrics, we used the Bray-Curtis measure with untransformed metric percentages.

Analysis of similarities

We assessed the strength of each classification by comparing within- and between-class similarities. In a strong classification, similarities between sites that are in the same class tend to be substantially greater than similarities between sites that are in different classes. To evaluate the compactness and isolation of the classes composing 1 single classification, Smith et al. (1990) suggested examining a matrix of means of between-site similarities within each class and between each pair of classes (see Rohm et al. 1987 for an example).

Here, we wished to compare the strengths of several alternative classifications. Mean similarity dendrograms (Van Sickle 1997) offer a compact graphical format for such comparisons. To construct the dendrogram for a single classification, we 1st calculated the mean of all between-class similarities ($\overline{B}$) and also the within-class mean similarity W_i for each class $i=1,2...k$. The dendrogram node is then plotted at $\overline{B}$ along a similarity axis. The dendrogram has k branches, 1 for each class, with each branch end plotted at W_i for that class. If a classification is strong, then its mean similarity dendrogram has relatively long branches. That is, $\overline{B}$ is low, and for each class, W_i is high.

The overall weighted mean ($\overline{W}$) of within-class similarities can be calculated as $\overline{W} = \sum_i (n_i / N)W_i$, where n_i is the number of sites in class i, and N is the total number in all classes (Van Sickle 1997). Then a single measure of overall classification strength (CS) is provided by statistics such as the ratio $\overline{B}/\overline{W}$ (Smith et al. 1990, Van Sickle 1997) or the difference ($\overline{W} - \overline{B}$). Here we use $CS = (\overline{W} - \overline{B})$ because it can be interpreted as the (weighted) average length of all branches in a dendrogram.

We used a permutation procedure to test whether the overall strength of a specific a priori site classification, such as ecoregions or HAUs, was significant in the sense of being greater than would be seen in random groups of sites (Mielke et al. 1976, Clarke and Green 1988, Smith et al. 1990). The statistic $CS = (\overline{W} - \overline{B})$ was recalculated for each of 10,000 randomly-chosen reassignments of sites to groups of the same size as the tested classification (Jackson and Somers 1989). The p-value giving evidence against the null hypothesis of no class structure was then estimated as the proportion of the 10,000 trials having CS at least as large as the value observed for the tested classification. The p-value of the test gives evidence for significant separation between at least 2 of several groups, but does not reveal which pairs of groups are particularly far apart.

We wrote special-purpose software to compute $\overline{B}$, W_i, and $\overline{W}$, and to perform the permutation tests (MEANSIM6, available at http://www.epa.gov/wed). Nearly equivalent permutation tests of $\overline{W}$ can be carried out using the MRPP method of Mielke, et al. (1976), available in the BLOSSOM software of the US Geological Survey, and in PC-ORD (B. McCune and M. J. Mefford, 1997. PC-ORD, Multivariate analysis of ecological data, Version 3.0. MjM Software Design, Gleneden Beach, Oregon). The ANOSIM method of Clarke and Green (1988), available in PRIMER (M.R. Carr, 1995. PRIMER user manual. Plymouth Marine Laboratory, Plymouth UK.) and in PATN (L. Belbin, 1992. PATN Technical reference. CSIRO, Division of Wildlife and Ecology, Canberra, Australia) also compares statistics of within- and between-class similarities to test the no class structure null hypothesis.

Clustering sites

We used the flexible-Beta clustering strategy, with Beta = -0.1, to create neutral-model site clusters (Lance and Williams 1967, Legendre and Legendre 1983, Belbin et al.1992). The strategy equally weights site groups, regardless of their size, in deciding which groups are mutually closest and should be fused, during the agglomerative clustering process (Lance and Williams 1967). With the strategy's Beta parameter set = - 0.1, clusters move slightly away from one another as they grow in size (Lance and Williams 1967), and the approach performs well across a range of data (Milligan 1989, Belbin et al. 1992). Following each clustering, the full hierarchical dendrogram was pruned to yield the 4 highest-level clusters, thus creating a classification having the same order (number of classes) as our ecoregion classification.

The neutral-model vertebrate assemblage clusters were based on the assemblage dissimilarity measure VERT = (100% - similarity), calculated for all site pairs (Digby and

Kempton 1987). One matrix of VERT dissimilarities was calculated for each of our 5 similarity measures (Bray-Curtis for species, families, and metrics, and Sorenson-Dice for species and families). Each matrix was then used to cluster sites, resulting in 5 alternative site classifications based on VERT dissimilarities. The neutral-model geographic distance clusters were based on a matrix (DIST) of between-site overland Euclidean distances.

Composite site groupings were based on the composite dissimilarity measure COMP = $\delta*$DIST + $(1-\delta)*$VERT, after normalizing the DIST matrix so that its largest distance had a value of 100%. We set δ equal to 0.5, thus assigning equal weightings to DIST and VERT dissimilarities. Composite dissimilarity measures, created as weighted averages of ≥ 2 dissimilarities, were 1st proposed by Gower (1971), and Legendre (1987) discusses the use of COMP-type measures in performing distance-constrained clustering. We constructed 5 COMP matrices by combining DIST with each of the VERT matrices in turn and, by clustering on each COMP matrix, generated 5 alternative COMP-model site groupings.

Assessing CS

We computed $CS = (\overline{W} - \overline{B})$ and tested the no class structure hypothesis for the ecoregion, HAU, HAU/ecoregion, stream order, and sampling effort classifications, using each of the 5 similarity measures. Classification strength was also assessed for each of the 11 clustering attempts (5 VERT-based classifications + 1 DIST-based classification + 5 COMP-based classifications). Each VERT or COMP classification was assessed using only the similarity measure corresponding to the VERT dissimilarity measure used in constructing that classification. Because the VERT assessments (and partly, the COMP assessments) were based on exactly the same similarity measure used to optimally choose the 4 site classes, the *CS* values for each of these classifications was near its maximum possible value for that particular similarity measure and number of classes. Thus, the permutation test of no class structure is invalid for these cases and was not performed. However, the classification derived solely from intersite geographic distance can be regarded as a priori, from the standpoint of vertebrate similarities; hence, the no class structure test of the DIST-derived groups are valid and were performed.

Results

Relative strengths of classifications

With only 1 exception, all a priori classifications, as assessed by all 5 vertebrate similarity measures, showed statistical evidence ($p < 0.02$) of greater *CS* than would be seen for randomly grouped sites (Table 1). However, these *CS* values were not numerically large. Ecoregions, with *CS* ranging from 4% to 13% across similarity measures, were consistently stronger classifiers than HAUs, which ranged from *CS* = 1% to 5%. Table 1 also reports $\overline{B}/\overline{W}$, to facilitate comparisons with other papers in this series. Values of $\overline{B}/\overline{W}$ near 1.0 indicate a weak classification, and *CS* increases progressively as $\overline{B}/\overline{W}$ decreases from 1.0 towards 0.

No branch of the ecoregion dendrogram was very short, and branch lengths were not greatly unequal, showing that the 4 ecoregions have non-negligible and comparable levels of within-class homogeneity in species presence/absence (Fig. 2). We found similar patterns for the other similarity measures, except for the metrics similarity (Fig. 3).

Mean similarity dendrograms for species presence/absence (Fig. 2) showed that within-HAU similarity was high for the North Coast and South Coast HAUs, relative to the mean similarity within the Willamette Basin HAU. However, the ecoregions dendrogram showed that

Willamette Valley assemblages in the Willamette Basin were more similar amongst themselves, as were the Cascade assemblages within the Basin. This result suggests that the HAU/ecoregion partition might be stronger than either ecoregions or HAUs alone. In fact, the HAU/ecoregion partition had *CS*s very close to that of the ecoregion-only classification, across the 5 similarity measures (Table 1).

The 3 HAUs classified vertebrates about as well as stream order (Table 1), and slightly better than the 3 sampling effort groups. The low *CS* values for sampling effort groups (2 to 4%; Table 1) showed that there was relatively little difference between the 3 sampling efforts arising from differences in sampling years and other possible factors. This result reassured us that combining data from the 3 studies resulted in negligible confounding of the estimates of strength for other classifications.

The DIST clustering formed distinct spatial groups, as intended (Fig. 4). The strength of this neutral classification was within a few % of the ecoregion and HAU/ecoregion partitions for every similarity measure (Table 1). These small differences are not surprising if one compares the spatial patterns of sites for the DIST and ecoregion groupings (Figs. 4 and 5). Both partitions defined regions corresponding to the Cascade Mountains and the Southern Oregon Klamath Mountains, and their major differences are only their partitionings of Willamette Valley and Coastal streams (Fig. 1). The ecoregions map split these streams into east-west groupings (Fig. 5), whereas the comparable DIST classes had a north-south separation (Fig. 4).

The VERT classifications had *CS*s ranging from 23 to 47% for the 5 similarity measures, representing upper bounds for the *CS* that can be achieved by 4-class partitions of these 137 sites (Table 1). However, these groupings have little geographic coherence, as illustrated by a map of the groups derived from the species presence/absence VERT matrix (Fig. 6).

The COMP classifications showed some geographic coherence, but at a cost in *CS*, which decreased to 19% for species presence/absence (Fig. 7, Table 1). COMP classes overlapped considerably in space and did not offer a clear partitioning of western Oregon. However they did suggest approximate areas within which one might expect to find similar assemblages.

Figures 6 and 7 were derived all or in part from species presence/absence VERT dissimilarities. VERT-derived classifications based on the other 4 dissimilarity measures displayed the same trend of increasing geographic coherence (maps not shown) and decreasing vertebrate classification strength (Table 1), as one moved from VERT classes, to COMP classes, and finally to distinct DIST regions.

Sensitivity of CSs to similarity measure

For a single classification scheme, whether cluster-derived or a priori, a consistent increase in $\overline{W}$ is clear as one scans the 5 similarity measures from species abundance to species P/A to family abundance to family P/A, and finally to metrics abundance (left to right across Table 1). This same sequence of measures represents a gradient of detail, from more to less, in the set of assemblage descriptors that determine similarity. This result is consistent with the expectation that, for any pair of assemblages, a greater degree of matching (higher similarity) would occur for measures using less detail (metrics abundance) than for those using more detail (species abundance).

However, $\overline{B}$ tended to increase in the same direction, and for the same reasons, as did $\overline{W}$, across similarity measures. As a result, for any single classification, *CS* itself showed no clear trend across the sequence of similarity measures, and in most cases it varied little (Table 1).

For ecoregions, the stability of *CS* relative to similarity measures is illustrated by the way that mean similarity dendrograms of the 4 (species, family) x (abundance, presence/absence) measures are progressively shifted along the similarity axis, while showing little variation in average branch length (Fig. 3). In short, our overall *CS* measure appeared to be fairly robust, relative to the level of assemblage compositional detail that was expressed by various similarity measures.

The 5-metrics abundance measure did not clearly conform to this pattern. For this measure, *CS* was high for VERT and COMP classifications, but it was also so low for all a priori classifications that they could not be reliably ranked amongst themselves (Table 1). The low value of *CS* for the 5-metrics measure is exemplified by its ecoregion dendrogram (Fig. 3), which showed that mean similarity within the Willamette Valley was actually slightly lower than $\overline{B}$.

Discussion

Ecoregions and HAUs

The 4 ecoregions were more effective than the 3 HAUs for classifying our 137 wadeable stream sites, in the sense of having consistently higher *CS*s across a gradient of taxonomic information on vertebrate assemblages. Our HAU/ecoregion classification, which conferred notably higher *CS* on the 3 HAUs simply by dividing 1 of the 3 along ecoregion lines, illustrates the value of using both regionalizations and not regarding the two as an either/or choice (Omernik and Bailey 1997).

Consistent with their *CS* values, ecoregions do offer a useful framework for examining patterns in the raw species assemblage data. Differences between ecoregions arise because of a small number of species that are common in 1 region but absent or rare in others. Four species (Cottus beldingi, Ascaphus truei, Bufo boreas, Salvelinus fontinalis) characterize the Cascades ecoregion. The 1st is a sculpin that prefers large substrates and cold water (Zaroban et al. 1999), and the tailed frog prefers similar conditions but at the stream edge or in cascades (Stebbins 1954), whereas the western toad is typically found near water in meadows; brook trout is an alien species commonly introduced to cold headwater streams. The Cascades are also characterized by the absence of several species that commonly occur in other regions (Cottus perplexus, Lampetra tridentata, Rhinichthys osculus, Oncorhynchus kisutch). The first 3 tolerate warmer water and higher sediment loads (Zaroban et al. 1999), whereas the coho salmon (O. kisutch) is restricted from the region by dams, falls, and flows too low for adult migration. The Willamette Valley ecoregion supports the most speciose streams and is characterized by alien warmwater taxa (Ameiurus natalis, Micropterus salmoides, Gambusia affinis, Rana catesbiana) as well as the sandroller (Percopsis transmontana), a species that can tolerate high levels of bed sediments and prefers vegetative cover such as roots, submerged brush, and macrophytes (Zaroban et al. 1999). The Coast Range ecoregion might be considered a sculpin region with 3 species (Cottus aleuticus, C. asper, and C. gulosus) found only or largely there. These species along with O. kisutch thrive in cool water with easy access to the Pacific Ocean (Zaroban et al. 1999). The Klamath ecoregion shares most of its species with the Cascades and Coast Range, but differs from both by sharing Richardsonius balteatus with the Willamette Valley and from the Cascades by supporting O. kisutch. Richardsonius balteatus is a cool to warmwater minnow (Zaroban et al. 1999) found in the agricultural valleys of the region, whereas the coho salmon benefits from the Rogue and Umpqua Rivers, which provide passage between the Pacific Ocean and Cascade

Mountains.

Neutral-model clusters

The DIST classification had at least twice the *CS* as HAUs for the 4 taxa-based similarities, and also had about the same *CS*s as those of ecoregions or the HAU/ecoregion hybrid. Thus, our data showed no evidence that ecoregional or HAU boundaries confered any additional *CS* for stream vertebrate assemblages over that achieved by spatial correlation effects. This conclusion must be qualified by the fact that our distance-based clustering approach only generated a single neutral-model partition of western Oregon (Fig. 4), which turned out to be not radically different in site composition from the ecoregion grouping (Fig. 5). In fact, any such neutral model is restricted by the relatively few ways that a 2-dimensional space can be partitioned into compact subregions having approximately equal areas. The randomly located circles used by Jenerette et al. (1998) offer a richer set of neutral-model regions, but because these circles are assessed one at a time and can overlap, they do not generate full regional partitions. The random circles can thus be used as a neutral model for assessing within-region homogeneity of individual catchments or ecoregions, but their applicability to the overall *CS* of a geographic partition is unclear. There is clearly room for improvement in the available formulations of neutral models for regional partitions.

These results should neither surprise nor deter users of geographic frameworks such as ecoregions and large catchments. First, it bears repeating that ecoregions and large catchments are intended for use in classifying whole ecosystems and their entire biophysical setting. Although very helpful for this purpose, these regionalizations are, at the same time, unlikely to strongly classify any 1 ecosystem component such as a faunal assemblage. Second, the development of nongeographic classification models requires large sets of internally consistent data, obtained from carefully planned, spatially distributed sampling efforts. Such databases are rarely available, and corresponding models of whole-ecosystem classification have yet to be developed. In the absence of these tools, ecoregions and catchments offer geographic partitions within which to expect somewhat similar conditions and responses to management activities.

VERT classifications had much higher *CS*s than any geographic classification of the same order. In other words, there is a substantial amount of variability in our vertebrate assemblage data that is not explained by regional factors. A thorough exploration of possible explanatory factors is beyond the scope of this paper, but our results for the similarity measure based on assemblage impairment metrics offered some indirect evidence that human disturbance factors are likely candidates. The high *CS* of the metrics-based VERT classification (Table 1) showed that the metrics are capable of defining distinct groups of sites. At the same time, the very low metrics-based *CS* values of geographic partitions and stream order (Table 1) suggest that the metrics are relatively insensitive to biogeographic factors, as was intended in their design. Lastly, we note that the metric-based W_i for the Willamette Valley ecoregion was unusually low, compared with that of other ecoregions (Fig. 3). Because the Willamette Valley, with its mix of agricultural, forested, and urbanized areas, is likely to have the greatest disturbance variability of the 4 ecoregions, it is also likely to show the greatest within-ecoregion variation (i.e., lowest mean similarity) in disturbance-sensitive assemblage metrics.

Our COMP classifications were based on an average of the taxonomic dissimilarity and geographic distance between sampled assemblages. They are useful for studying the large-scale geographic patterns that are often observed in species assemblage data sets, relative to the patterns of biophysical factors that may influence assemblage composition (Hughes et al. 1987). The COMP dissimilarity measure allows for any desired relative weighting of geographic distance and assemblage dissimilarity. As one adjusts the weighting parameter δ from 0 to 1, the *CS* value of the 4 resulting COMP-based clusters smoothly decreases from the maximum levels seen for purely assemblage-based (VERT) clusters to the lower strength seen for a purely geographic (DIST) grouping. At the same time, the COMP-based clusters progressively coalesce from the scatter seen in Fig. 6 to the distinct regions of Fig. 4.

Robustness of CS to choice of similarity measure

Our 5 assemblage similarity indices represented the spectrum of measures commonly used for aquatic vertebrate and invertebrate sample assemblages. Four of these indices ranked several classifications in a consistent order with respect to our *CS* measure, suggesting that this statistic is fairly robust relative to the level of assemblage compositional detail that goes into measuring assemblage similarity. As we have already suggested, the 5th index (metric similarity) is likely responding to human disturbance rather than biogeographic patterns, giving it somewhat different behavior.

Our assessment of *CS* robustness to the choice of similarity measure and taxonomic level of the sample assemblage adds a small piece to the substantial and growing literature on such comparisons (e.g., Cao et al. 1997). So far, this literature has succeeded in articulating only a few clear, broadly applicable patterns of behavior for similarity measures. Here, we have tried to follow Green's (1980) advice and place strongest reliance on classification results that are qualitatively consistent across multiple measures of assemblage similarity.

Statistical vs ecological significance

In recent years, permutation tests of the no class structure null hypothesis, based on within- and between-class similarities, have been increasingly used as they become widely available in statistical ecology software. Our experience suggests that such permutation tests are too powerful for moderate and larger sample sizes (>50 sites); that is, a very small *p*-value will result even if an observed *CS* differs only slightly in magnitude from the value expected under chance assignments of sites to classes.

Furthermore, evidence against the no class structure null hypothesis is, by itself, not particularly informative (Green 1980, Gordon 1981). One can learn a great deal more by assessing the biological significance of plausible, competing models than by testing the statistical significance of a single model against an unrealistic and uninformative null (Yoccoz 1991, Stewart-Oaten et al. 1992, Hilborn and Mangel 1997). For these reasons, our analysis emphasizes comparisons of the relative magnitudes of *CS* statistics for several classifications, including neutral models, which represent alternative plausible models of assemblage variation over space.

Our mean similarity dendrograms expand on the *CS* comparisons by illustrating within-class homogeneities relative to a single measure of average between-class similarity. However, they are of limited help in aggregating regions to create simplified, special-purpose regionalizations with little loss in *CS* (Hughes et al. 1987, Barbour et al. 1996). Natural candidates for aggregation would be region pairs with relatively high between-region mean similarity, and except for some special cases (Van Sickle 1997), these candidates are only revealed by inspecting the full matrix of mean between- and within-region similarities (Smith et al. 1990). This examination can be difficult for > 3-4 regions, but graphical methods can still be of assistance. A multidimensional scaling ordination (Digby and Kempton 1987) provides a 2 or 3-dimensional display of the approximate relative mean dissimilarities between regions. One could also carry out a cluster analysis on the mean similarity matrix itself to reveal whole-region neighbor relations.

In conclusion, vertebrate assemblages in western Oregon's wadeable streams were more similar, on average, within ecoregions than they were between ecoregions. Ecoregions and the HAU/ecoregion partition were consistently stronger classifiers than HAUs alone, but not by much. None of these ecologically based geographic partitions showed greater strength in classifying our data than did a neutral partition whose strength derived mainly from spatial autocorrelation effects.

In short, choosing between similarly ordered geographic partitions (ecoregions vs catchments) appears to be a fairly minor issue, if one's goal is to find an optimal way to classify stream ecosystems. A much broader range of classification strengths can be found by loosening, either partly or entirely, geographic constraints on grouping streams. Constructing these improved classifications, however, will require the acquisition of quantitative environmental and biological data from many sites.

Acknowledgements

We thank Francie Faure for graphics assistance and Shay Howlin for statistical programming. Leska Fore, Chuck Hawkins, Phil Larsen, Kelly Moore, Scott Urquhart , and an anonymous reviewer provided valuable suggestions on the manuscript. The research described in this article has been funded by the US Environmental Protection Agency and is a joint contribution of the Pacific Northwest Research and the Environmental Monitoring and Assessment programs. This document was prepared at the EPA National Health and Environmental Effects Research Laboratory in Corvallis, Oregon, in part through contract No. 68-C5-0005 to Dynamac . It has been subjected to the Agency's peer review and approved for publication. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

Literature Cited

Angermeier, P. L., and R. A. Smogor. 1995. Estimating number of species and relative abundances in stream-fish communities: effects of sampling effort and discontinuous spatial distributions. Canadian Journal of Fisheries and Aquatic Sciences 52:936-949.

Barbour, M. T., J. Gerritsen, G. E. Griffith, R. Frydenborg, E. McCarron, J. S. White, and M. L. Bastian. 1996. A framework for biological criteria for Florida streams using benthic macroinvertebrates. Journal of the North American Benthological Society 15:185-211.

Beecher, H. A., E. R. Dott, and R. F. Fernau. 1988. Fish species richness and stream order in Washington state streams. Environmental Biology of Fishes 22:193-209.

Belbin, L., D. P. Faith, and G.W. Milligan. 1992. A comparison of two approaches to Beta-flexible clustering. Multivariate Behavioral Research 27:417-433.

Cao, Y., W. P. Williams, and A. W. Bark. 1997. Similarity measure bias in river benthic aufwuchs community analysis. Water Environment Research 69:95-106.

Caswell, H. 1976. Community structure: a neutral model approach. Ecological Monographs 46:327-354.

Clarke, K. R., and R. H. Green. 1988. Statistical design and analysis for a 'biological effects' study. Marine Ecology Progress Series 46:213-226.

Davis, W. S., and T. P. Simon (EDITORS). 1995. Biological assessment and criteria. Tools for water resource planning and decision making. Lewis Press, Boca Raton, Florida.

Digby, P. G. N., and R. A. Kempton. 1987. Multivariate analysis of ecological communities. Chapman and Hall, London, UK.

Fausch, K. D., J. R. Karr, and P. R. Yant. 1984. Regional application of an index of biotic integrity based on stream fish communities. Transactions of the American Fisheries Society 113:39-55.

Fausch, K. D., J. Lyons, J. R. Karr, and P. L. Angermeier. 1990. Fish communities as indicators of environmental degradation. American Fisheries Society Symposium 8:123-144.

Gardner, R. H., B. T. Milne, M. G. Turner, and R. V. O’Neill. 1987. Neutral models for the analysis of landscape pattern. Landscape Ecology 1:19-28.

Gerritsen, J. 1995. Additive biological indices for resource management. Journal of the North American Benthological Society 14:451-457.

Gordon, A.D. 1981. Classification. Chapman and Hall, London.

Gower, J. C. 1971. A general coefficient of similarity and some of its properties. Biometrics 27:857-872.

Green, R. H. 1980. Multivariate approaches in ecology: the assessment of ecologic similarity. Annual Review of Ecology and Systematics 11:1-14.

Hawkes, C. L, D. L. Miller, and W. G. Layher. 1986. Fish ecoregions of Kansas: stream fish assemblage patterns and associated environmental correlates. Environmental Biology of Fishes 17:267-279.

Herlihy, A. T., P. R. Kaufmann, L. Reynolds, J. Li, and E. G. Robison. 1997. Developing indicators of ecological condition in the Willamette Basin: an overview of the Oregon prepilot study for EPA's EMAP program. Pages 275-282 in A. Laenen and D. A. Dunnette (editors). River quality: dynamics and restoration. Lewis Press, Boca Raton, Florida.

Hilborn, R., and M. Mangel. 1997. The ecological detective: confronting models with data. Princeton University Press, Princeton, New Jersey.

Hocutt, C. H., and E. O. Wiley. 1986. The zoogeography of North American freshwater fishes. John Wiley and Sons, New York.

Huang, S. L., and J. J. Ferng. 1990. Applied land classification for surface water quality management: I. Watershed classification. Journal of Environmental Management 31:107-126.

Hughes, R. M. 1995. Defining acceptable condition. Pages 31-47 in W. Davis and T. Simon (editors). Biological assessment and criteria. Tools for water resource planning and decision making. Lewis Press, Boca Raton, Florida.

Hughes, R. M., S. A. Heiskary, W. J. Matthews, and C. O. Yoder. 1994. Use of ecoregions in biological monitoring. Pages 125-151 in S. L. Loeb and A. Spacie (editors). Biological monitoring of freshwater ecosystems. Lewis Press, Chelsea, Michigan.

Hughes, R. M., P. R. Kaufmann, A. T. Herlihy, T. M. Kincaid, L. Reynolds, and D. P. Larsen. 1998. A process for developing and evaluating indices of fish assemblage integrity. Canadian Journal of Fisheries and Aquatic Sciences 55:1618-1631.

Hughes, R. M., E. Rexstad, and C. E. Bond. 1987. The relationship of aquatic ecoregions, river basins, and physiogeographic provinces to the ichthyogeographic regions of Oregon. Copeia 1987:423-432.

Jackson, D. A., and K. M. Somers. 1989. Are probability estimates from the randomization model of Mantel's test stable? Canadian Journal of Zoology 67:766-769.

Jenerette, D., J. Lee, D. Waller, and R. Carlson. 1998. The effect of spatial dimensions on regionalization of lake water quality data. Pages 256-264 in T. K. Poiker and N. Chrisman (editors). Proceedings of the 8th International Symposium on Spatial Data Handling. Geographic Information Science Study Group, International Geographic Union. Available from: Department of Geography, Simon Fraser University, Burnaby, British Columbia.

Kuehne, R. A. 1962. A classification of streams, illustrated by fish distribution in an eastern Kentucky creek. Ecology 43:608-614.

Lance, G. N., and W. T. Williams. 1967. A general theory of classificatory sorting strategies: I. Hierarchical systems. Computer Journal 9:373-380.

Legendre, L., and P. Legendre. 1983. Numerical ecology. Elsevier, Amsterdam.

Legendre, L., and V. Legendre. 1984. Postglacial dispersal of freshwater fishes in the Quebec

Peninsula. Canadian Journal of Fisheries and Aquatic Sciences 41:1781-1802.

Legendre, P. 1987. Constrained clustering. Pages 289-306. in P. Legendre and L. Legendre (editors). Developments in numerical ecology. Springer-Verlag, Berlin.

Legendre, P., N. L. Oden, R. R. Sokal, A. Vaudor, and J. Kim. 1990. Approximate analysis of variance of spatially autocorrelated regional data. Journal of Classification 7:53-75.

Ludwig, J. A., and J.F. Reynolds. 1988. Statistical ecology. John Wiley and Sons, New York.

Lyons, J. 1992. The length of stream to sample with a towed electrofishing unit when fish species richness is estimated. North American Journal of Fisheries Management 12:198-203.

Matthews, W.J. 1998. Patterns in freshwater fish ecology. Chapman and Hall, New York.

McCormick, F. H., and R. M. Hughes. 1998. Aquatic vertebrates. Pages 159-180 in J. L. Lazorchak, D. J. Klemm, and D. V. Peck (editors). Field operations and methods for measuring the ecological condition of wadeable streams. EPA/620/R-94/004. US Environmental Protection Agency. Washington, DC.

Mielke, P. W., K. J. Berry, and E. S. Johnson. 1976. Multiresponse permutation procedures for a priori classifications. Communications in Statistical Theory and Methods A5:1409-1424.

Milligan, G.W. 1989. A study of the beta-flexible clustering method. Multivariate Behavioral Research 24:163-176.

Moyle, P. B., and M. P. Marchetti. 1998. Applications of indices of biotic integrity to California streams and watersheds. Pages 367-380 in T. P. Simon (editor). Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press, Boca Raton, Florida.

Naiman, R. J., D. G. Lonzarich, T. J. Beechie, and S. C. Ralph. 1992. General principles of classification and the assessment of conservation potential in rivers. Pages 93-123 in P. J. Boon, P. Calow and G. E. Petts (editors). River conservation and management. John Wiley and Sons, New York.

Omernik, J. M. 1987. Ecoregions of the conterminous United States. Annals of the Association of American Geographers 77:118-125.

Omernik, J. M., and R. G. Bailey. 1997. Distinguishing between watersheds and ecoregions. Journal of the American Water Resources Association 33:935-949.

Omernik, J. M., and G. E. Griffith. 1991. Ecological regions versus hydrologic units: frameworks for managing water quality. Journal of Soil and Water Conservation 46:334-340.

Paller, M. H. 1995. Relationships among number of fish species sampled, reach length surveyed, and sampling effort in South Carolina Coastal Plain streams. North American Journal of Fisheries Management 15:110-120.

Parsons, M., and R. H. Norris. 1996. The effect of habitat-specific sampling on biological assessment of water quality using a predictive model. Freshwater Biology 36:419-434.

Platts, W. S. 1979. Relationships among stream order, fish populations, and aquatic geomorphology in an Idaho river drainage. Fisheries 4(2):5-9.

Rathert, D., D. White, J. C. Sifneos, and R. M. Hughes. 1999. Environmental correlates of species richness for native freshwater fish in Oregon, USA. Journal of Biogeography 26:257-273.

Reynoldson, T. B., R. C. Bailey, K. E. Day, and R. H. Norris. 1995. Biological guidelines for freshwater sediment based on **BE**nthic **A**ssessment of **S**edimen**T** (the **BEAST**) using a multivariate approach for predicting biological state. Australian Journal of Ecology 20:198-219.

Rohm, C.M., J. W. Giese, and C. C. Bennett. 1987. Evaluation of an aquatic ecoregion classification of streams in Arkansas. Journal of Freshwater Ecology 4:127-140.

Seaber, P. R., F .P. Kapinos, and G. L. Knapp. 1987. Hydrologic unit maps. US Geological Survey Water Supply Paper 2294, US Geological Survey, Denver, Colorado.

Smith, E. P., K. W. Pontasch, and J. Cairns. 1990. Community similarity and the analysis of multispecies environmental data: a unified statistical approach. Water Research 24:507-514.

Sokal, R.R. 1974. Classification: purposes, principles, progress, prospects. Science 185:1115-1123.

Stebbins, R. C. 1954. Amphibians and reptiles of western North America. McGraw-Hill, New York.

Stewart-Oaten, A., J. R. Bence, and C. W. Osenberg. 1992. Assessing effects of unreplicated perturbations: no simple solutions. Ecology 73:1396-1404.

Van Sickle, J. 1997. Using mean similarity dendrograms to evaluate classifications. Journal of Agricultural, Biological, and Environmental Statistics 2:370-388.

Wright, J. F., D. Moss, P. D. Armitage, and M. T. Furse. 1984. A preliminary classification of running-water sites in Great Britain based on macroinvertebrate species and the prediction of community type using environmental data. Freshwater Biology 14:221-256.

Yoccoz, N. G. 1991. Use, overuse and misuse of significance tests in evolutionary biology and ecology. Bulletin of the Ecological Society of America 72:106-111.

Zaroban, D. W., M. P. Mulvey, T. R. Maret, R. M. Hughes, and G. D. Merritt. 1999. Proposed classification of species attributes for Pacific Northwest freshwater fishes. Northwest Science 73:81-93.

Figures

Fig. 1. Western Oregon, partitioned by ecoregions (solid lines; names vertically aligned in regular text), and by hydrologic accounting units (HAUs; shaded regions; names horizontally aligned in italics). Dots locate sampling sites on 137 wadeable streams.

Fig. 2. Mean similarity of species presence/absence, for Ecoregion, HAU (hydrologic accounting units), VERT, DIST, and COMP classifications of 137 western Oregon vertebrate assemblages. VERT classes (V1-V4), DIST classes (D1-D4), and COMP classes (C1-C4) were constructed by clustering on VERT (vertebrate assemblage), DIST (geographic), and COMP ($0.5*$VERT $+0.5*$DIST) dissimilarities, respectively. For each dendrogram, the node is plotted at the overall mean between-class similarity ($\overline{B}$), and each branch end is plotted at the mean within-class similarity (W_i) for that class. Class names on dendrogram branches correspond to the class memberships plotted in Figs 4-7.

Fig. 3. Mean similarity dendrograms of the ecoregion site classification for species, family, and metrics levels of vertebrate assemblage identification. AB = Bray-Curtis relative abundance similarity, P/A = Sorenson presence/absence similarity. Classification strength ($CS = \overline{W} - \overline{B}$) is the weighted average length of dendrogram branches. Branch names in all dendrograms are in same order as for family P/A.

Fig. 4. Classes (D1-D4) of western Oregon stream sites derived by clustering on intersite geographic distances (DIST).

Fig. 5. Western Oregon stream sites classified by ecoregion.

Fig. 6. Classes (V1-V4) of western Oregon stream sites derived by clustering on their dissimilarities (VERT) in presence/absence of aquatic vertebrate species.

Figure 7. Classes (C1-C4) of western Oregon stream sites derived by clustering on their composite measures (COMP) of intersite geographic distance and dissimilarity in species presence/absence.

Table 1. Strengths of 8 classifications for vertebrate assemblages from 137 wadeable stream sites in western Oregon. HAU = hydrologic accounting units. VERT, DIST, and COMP classifications were constructed by clustering on VERT (vertebrate assemblage), DIST (geographic), and COMP (0.5∗VERT +0.5∗DIST) dissimilarities, respectively. Each cell contains the classification strength measure $CS = [\overline{W} - \overline{B}]$ (%), the corresponding measure $\overline{B}/\overline{W}$, and the overall mean within-class similarity $\overline{W}$ (%). $\overline{B}$ = overall mean between-class similarity. Results are given for Bray-Curtis abundance and Sorenson presence/absence (P/A) measures of similarity, applied to species and family-level identifications, and for the Bray-Curtis abundance measure applied to 5 vertebrate assemblage metrics. In cases where the no class structure (NCS) test was performed, resulting *p*-values were <0.02 unless noted otherwise.

Classification	No. of classes	NCS test?	Species abundance	Species P/A	Family abundance	Family P/A	5 metrics abundance
			$\overline{W}-\overline{B}, \overline{B}/\overline{W}, \overline{W}$	$\overline{W}-\overline{B}, \overline{B}/\overline{W}, \overline{W}$	$\overline{W}-\overline{B}, \overline{B}/\overline{W}, \overline{W}$	$\overline{W}-\overline{B}, \overline{B}/\overline{W}, \overline{W}$	$\overline{W}-\overline{B}, \overline{B}/\overline{W}, \overline{W}$
VERT	4	No	35, 0.19, 43	23, 0.48, 44	45, 0.27, 62	32, 0.44, 57	47, 0.44, 84
COMP	4	No	30, 0.25, 40	19, 0.54, 41	34, 0.42, 59	21, 0.65, 60	35, 0.55, 77

DIST	4	Yes	14, 0.50, 28	12, 0.68, 38	11, 0.73, 41	9, 0.82, 51	5, 0.91, 55
Ecoregion	4	Yes	11, 0.58, 26	13, 0.66, 38	7, 0.83, 41	11, 0.79, 52	4, 0.93, 55
HAU	3	Yes	5, 0.76, 21	5, 0.84, 32	3, 0.92, 36	1[a], 0.98, 45	3, 0.94, 54
HAU/ecoregion	4	Yes	12, 0.56, 27	12, 0.68, 38	11, 0.74, 42	9, 0.82, 51	5, 0.91, 55
Stream order	3	Yes	5, 0.76, 21	5, 0.84, 32	4, 0.89, 37	3, 0.94, 47	3, 0.95, 55
Sampling effort	3	Yes	2, 0.89, 19	4, 0.87, 31	3, 0.91, 34	2, 0.96, 46	2, 0.96, 53

[a] $p = 0.09$ for NCS test

Figure 1

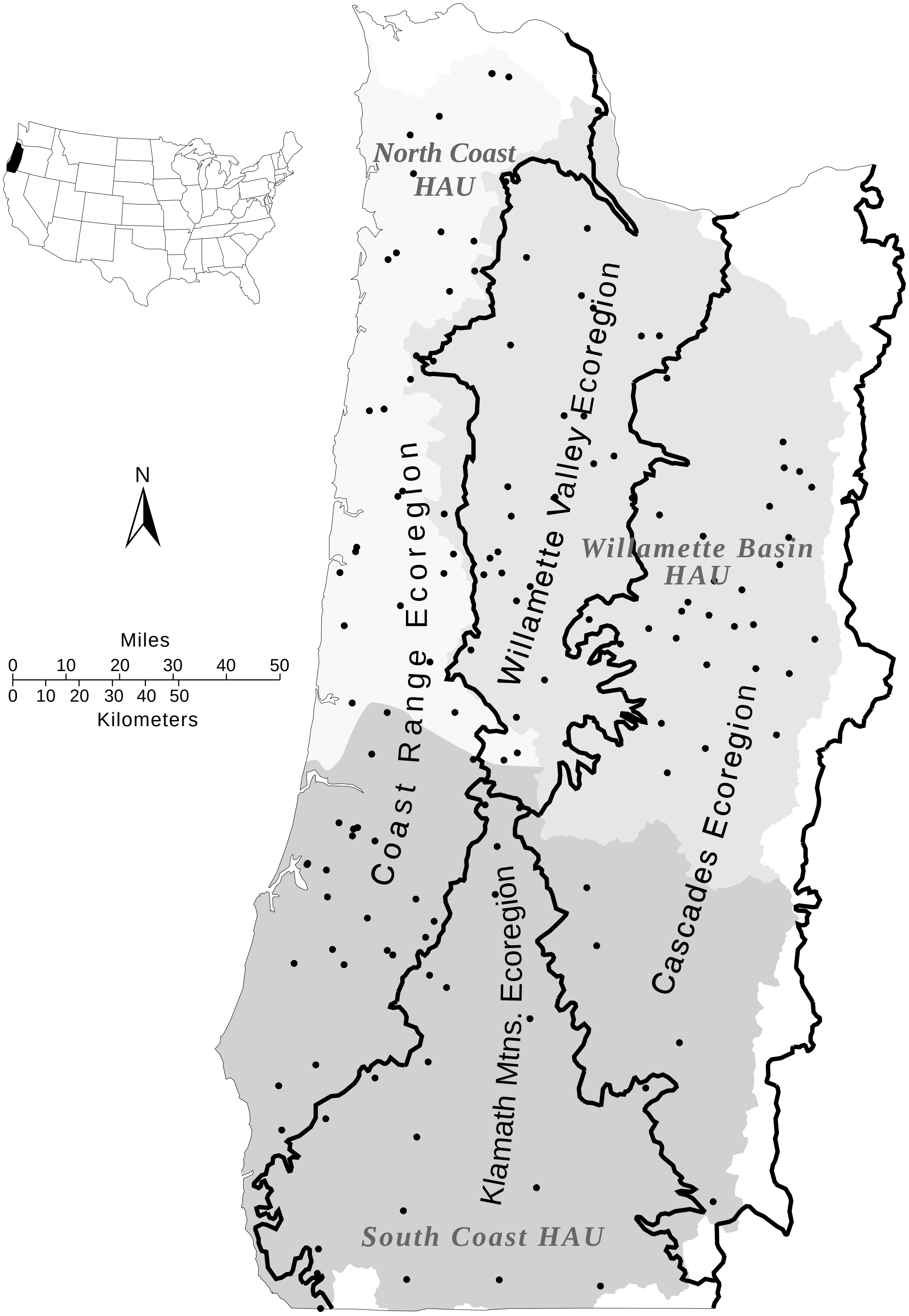
North Coast HAU
Willamette Basin HAU
South Coast HAU
Coast Range Ecoregion
Willamette Valley Ecoregion
Cascades Ecoregion
Klamath Mtns. Ecoregion
N
Miles
0 10 20 30 40 50
0 10 20 30 40 50
Kilometers

Figure 2

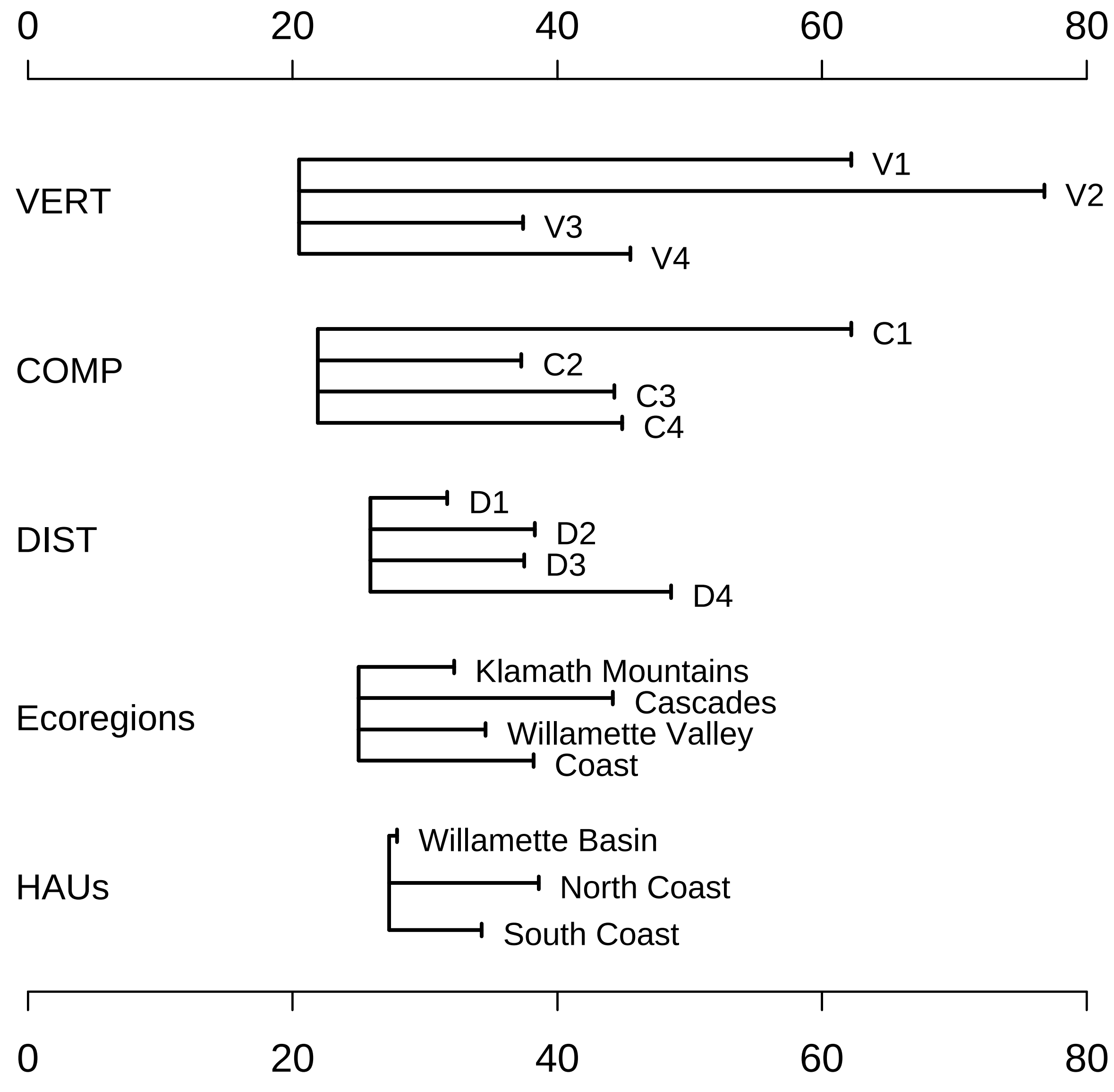
0
20
40
60
80
VERT
V1
V2
V3
V4
COMP
C1
C2
C3
C4
DIST
D1
D2
D3
D4
Ecoregions
Klamath Mountains
Cascades
Willamette Valley
Coast
HAUs
Willamette Basin
North Coast
South Coast
0
20
40
60
80
% of species shared (Sorenson)

Figure 3

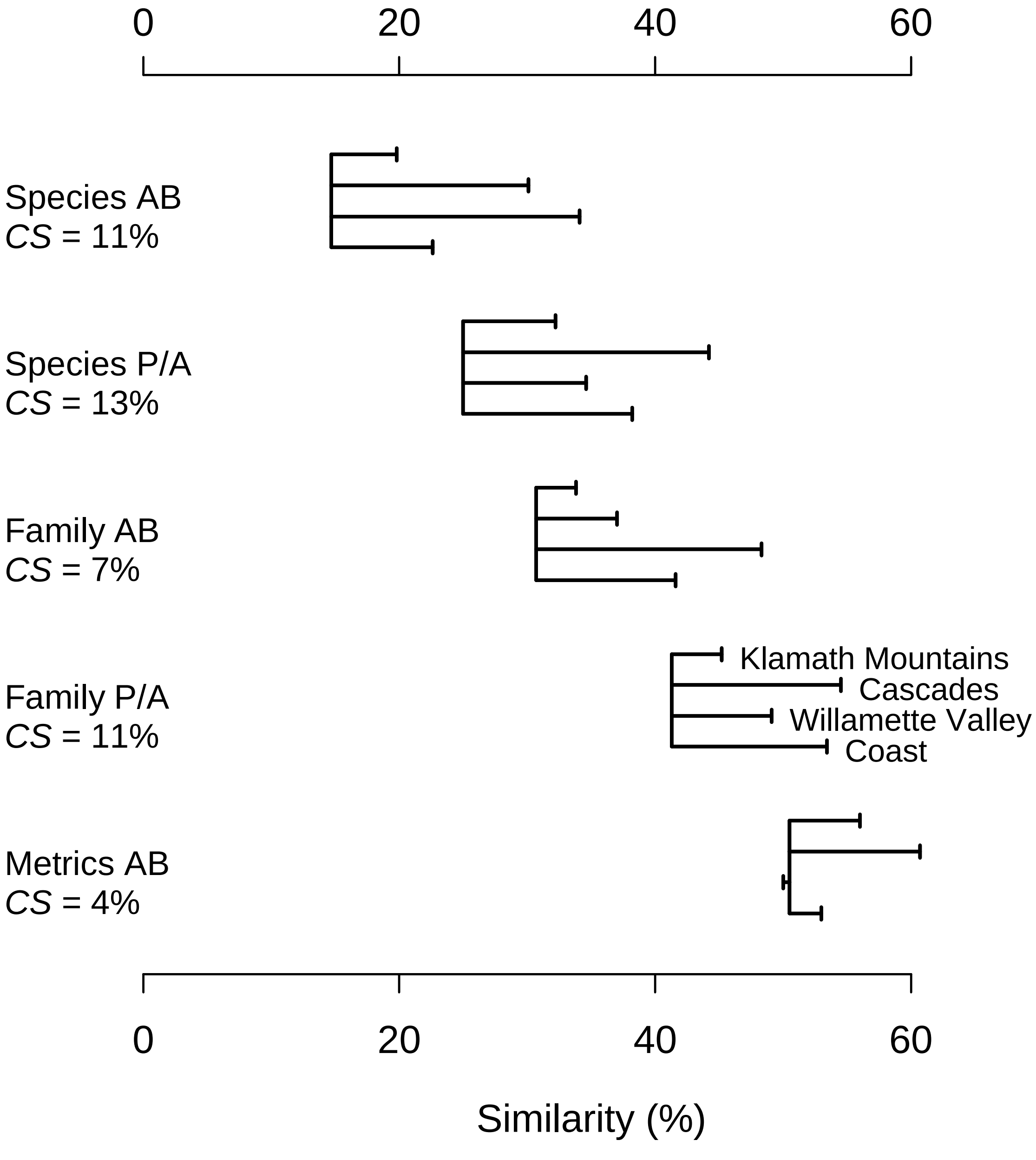
0
20
40
60
Species AB
CS = 11%
Species P/A
CS = 13%
Family AB
CS = 7%
Family P/A
CS = 11%
Klamath Mountains
Cascades
Willamette Valley
Coast
Metrics AB
CS = 4%
0
20
40
60
Similarity (%)

Figure 4

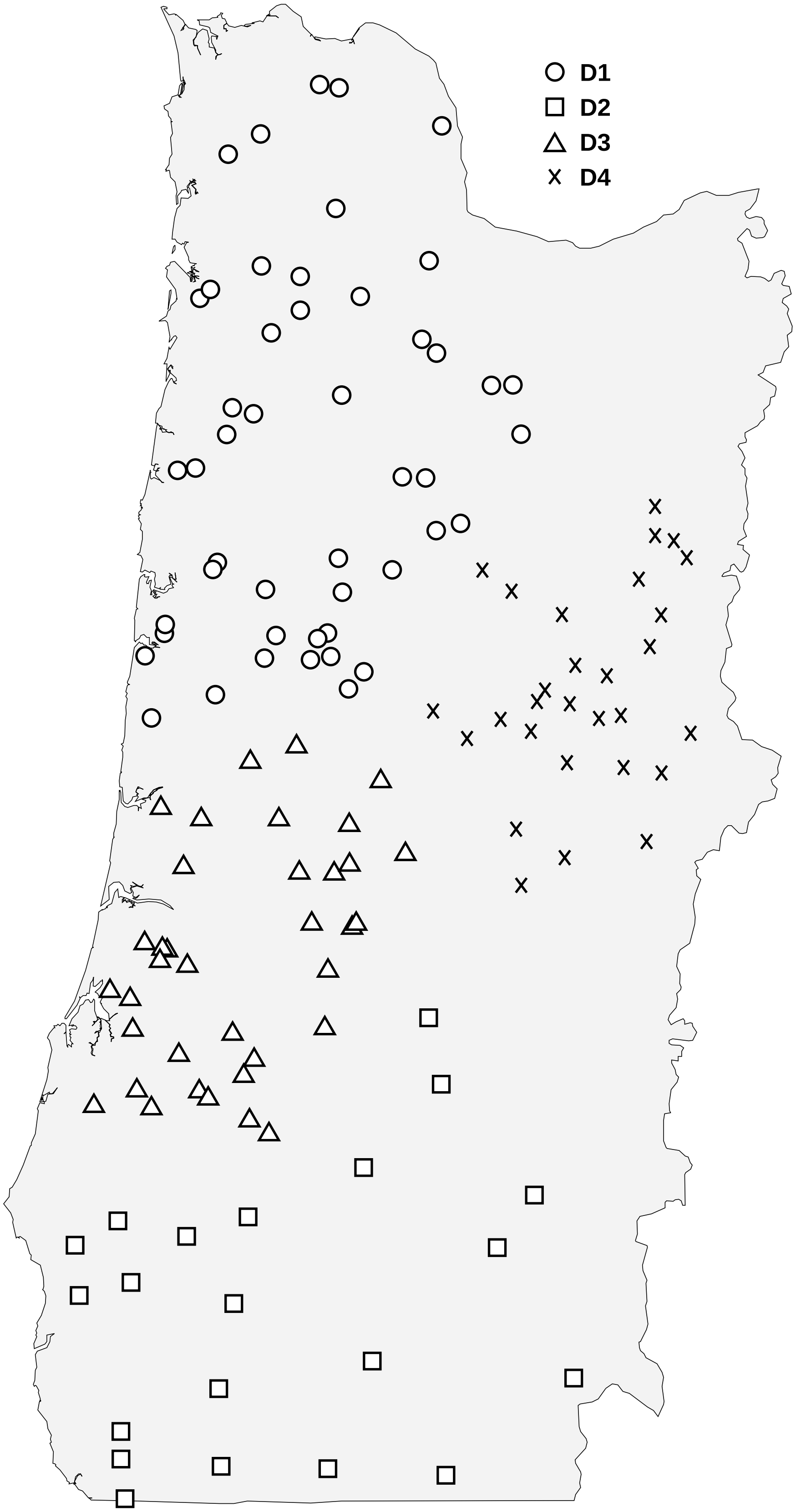

Figure 5

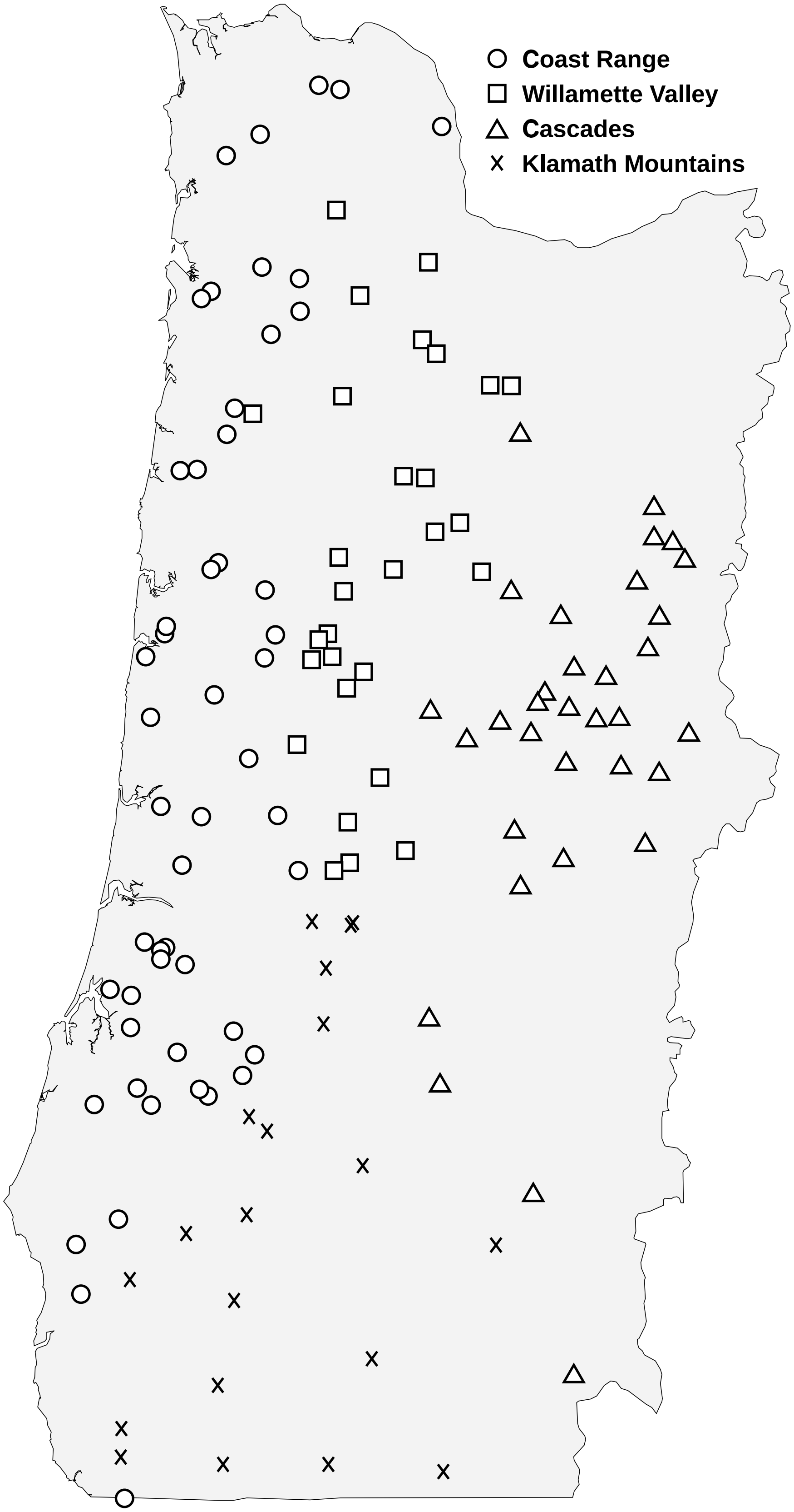
○ Coast Range
□ Willamette Valley
△ Cascades
x Klamath Mountains

Figure 6

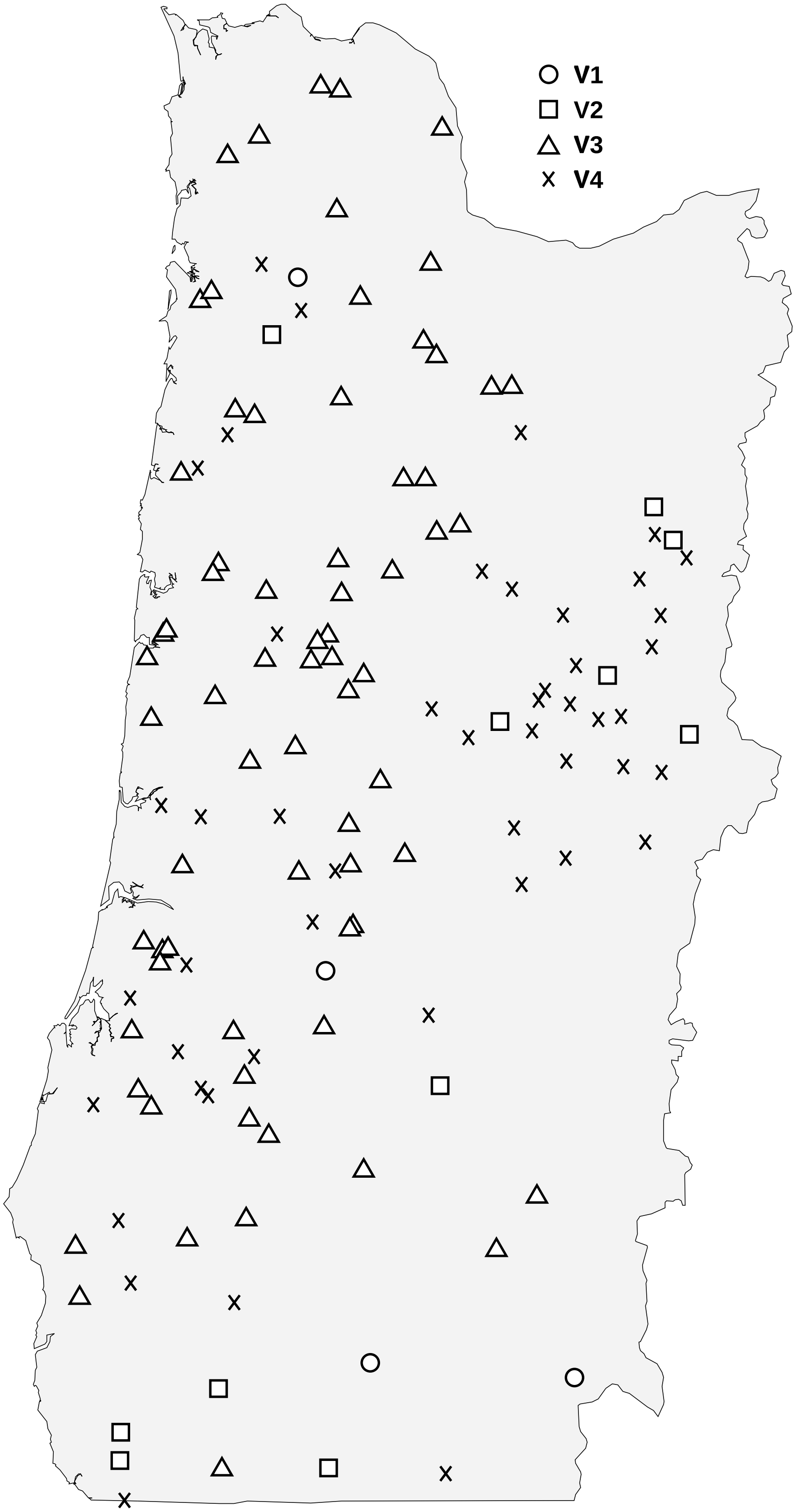

Figure 7