Biological Condition Gradient (BCG) Level 1 Biodiversity Index

Draft for review by the Pacific NW BCG Expert Panel (version 1, 4/19/2019)

Using benthic macroinvertebrate biomonitoring data to flag stream sites in the maritime Pacific Northwest that may possess exemplary biodiversity.

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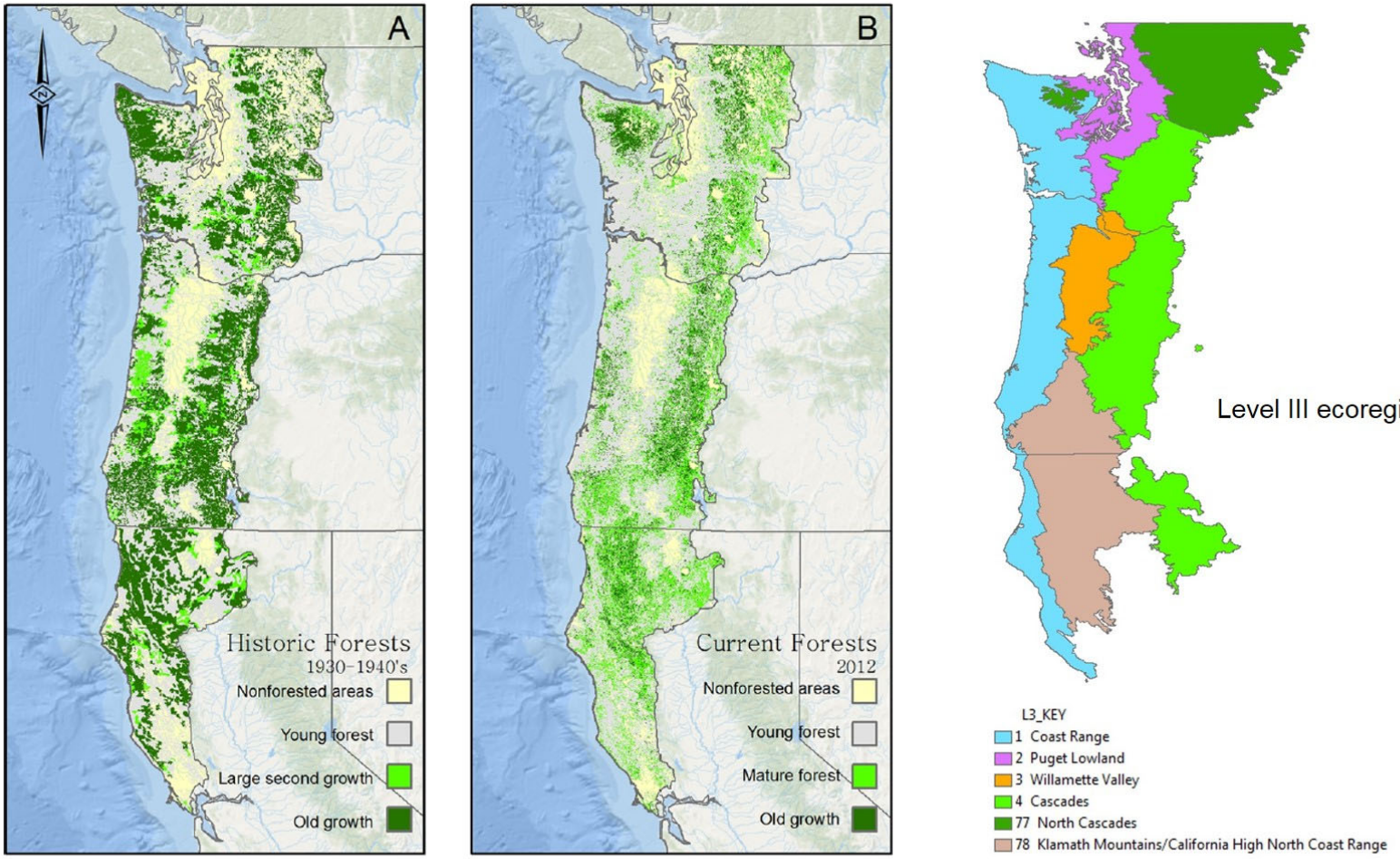
1 Background

Natural, geologic and recent human history have created a mosaic of mid-order, forested watersheds with varying expressions of aquatic invertebrate (AI) biodiversity in the maritime Pacific Northwest (Figure 1). At one extreme are homogenized AI communities composed of a few tolerant taxa that are widespread and common across North American or globally, e.g. urbanized or valley bottom agricultural streams. At the other end of the biodiversity spectrum are streams with a rich, resilient, layered habitat structure supporting high taxa richness, a variety of taxonomic groups, and many uncommon species whose geographic range is more regionally limited.

Here we add further to the narrative of what constitutes a Biological Condition Gradient (BCG) Level 1 watershed in the maritime Pacific Northwest (PNW), to be ones that harbor exemplary biodiversity, with AI communities representing the best of their kind (Figure 2, Table 1). **These are watersheds that have experienced minimal human disturbance, but also recognized is that the substantial number of near pristine, minimally disturbed watersheds still extant in the PNW vary greatly in their expression of AI biodiversity**. For example, glacial meltwater streams often support few species and in many ways their communities are as stressed and homogenized as any urban stream. Benthic biomonitoring data that we have reviewed from pristine (or nearly so) streams in National Parks and wilderness areas located along the Cascade Crest Montane Forest Ecoregion (US EPA 2016) indicate most streams have only moderate taxa richness, with most taxa being widespread and common in western North America. Only about 12,000 years have lapsed since glaciers retreated and streams formed in most of this high elevation zone, leaving little time for an endemic fauna to develop. In our experience, highest AI diversity is most often encountered in forested streams at mid-elevations (i.e. 300-1200 meters) in the maritime PNW. Minimally disturbed, lower elevation, forested streams that may have had exemplary biodiversity are now rare in the region. Within this mid-elevation band, BCG level 1 streams are ones having high habitat complexity and resilience to disturbance, and in turn support a high richness of AI taxa, particularly species endemic to a smaller geographic region. We propose here a Biodiversity Index that can help identify those sites that support an AI community with high and/or unique taxa richness representative of a BCG level 1 stream.



From Penaluma et al. 2017 (original source: Davis et al. 2015)

Figure 1. For purposes of this project, the maritime Pacific Northwest refers to the humid temperate areas extending from western Washington into northern California (Cascade Mountain crest to the Pacific Ocean), as depicted in the two maps on the left, which show historic forest cover (1930-1940's) and more current (2012) conditions (from Penaluma et al. 2017; originally from Davis et al. 2015). The crest of the Cascades generally divides the Humid Temperate Domain from the Dry Domain of the Pacific Northwest (Kimerling & Jackson 1985). The forest boundaries in the maps on the left correspond closely with the Level III Ecoregions shown on the right (Omernik 1987, 1995).

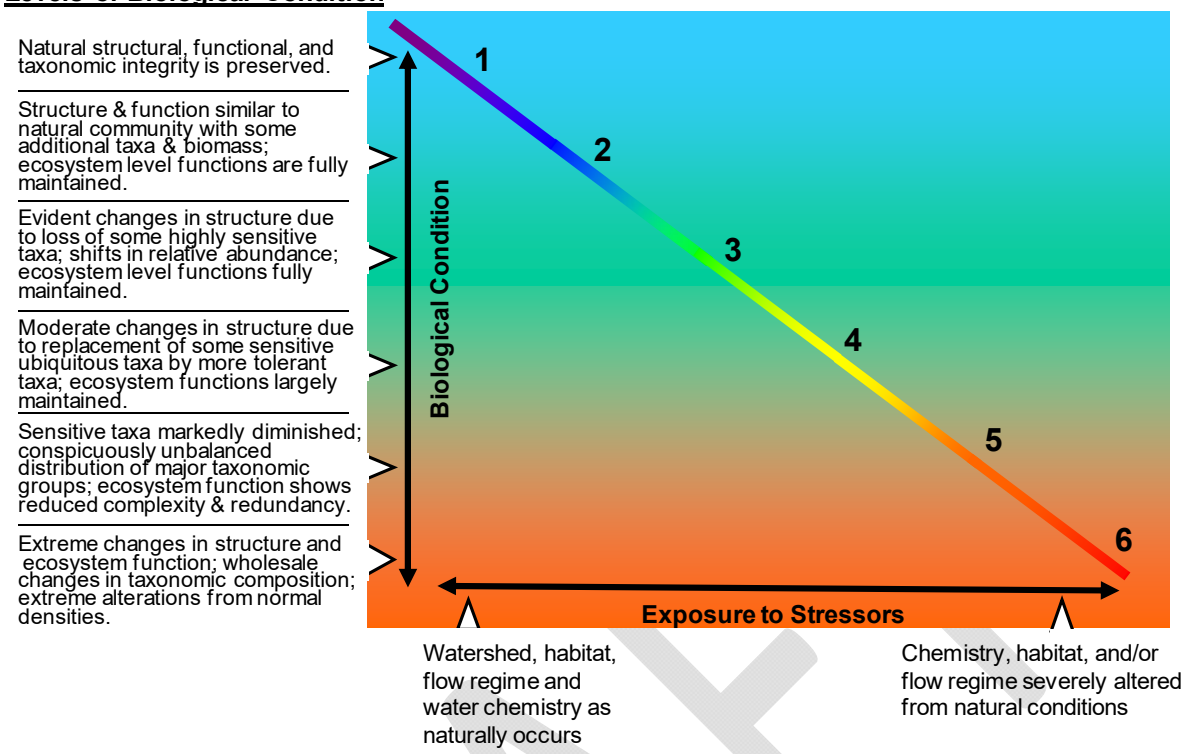


Figure 2. The Biological Condition Gradient (BCG), modified from Davies and Jackson 2006. The BCG was developed to serve as a scientific framework to synthesize expert knowledge with empirical observations and develop testable hypotheses on the response of aquatic biota to increasing levels of stress.

Table 1. The fundamental characteristics of BCG level 1, written by three BCG panelists (Rick Hafele, Rob Plotnikoff and Bob Wisseman) who researched and described, in as much detail as possible, conceptual expectations for BCG level 1 ("as naturally occurs") in perennial, freshwater wadeable (1 to 100 mi2) streams in the Puget Lowlands and Willamette Valley Level III ecoregions. This table was developed through an iterative process (refined with research and dialogue among the other BCG panelists).

Fundamental Characteristics	**Description**
Stream channel	Channel connected to hyporheos and flood plain including wetlands, beaver ponds, etc.; diverse habitats present (e.g. braided channels, side channels, debris jams, mixture of steps and pools consistent with stream gradient); wood debris typically present and may be abundant; quality habitat and refugia persists during periods of both low and high stream-flows.
Riparian & watershed	Riparian zone supports intact community of overstory, understory and groundcover plants (including a mixture of mature conifer and hardwood trees with a diverse age structure in forested watersheds); upper watershed vegetation intact, supporting delivery of water of high chemical and thermal quality to lower reaches.
Hydrologic regime	Hydrologic regime natural, without alteration from dams and/or irrigation withdrawals or return flow; cool-cold water common from springs, groundwater accretion, and/or natural runoff; perennial surface or subsurface flow. Re-charge in the watershed sustains flow, especially during years of extreme drought. Perennial surface water in some portion of watersheds maintain endemic taxa that serve as recolonization sources sustaining high biodiversity at select locations. These locations promote resiliency in stream reaches that are periodically de-watered.
Disturbance regime and resilience	Natural seasonal range of high and low stream-flows present, which enhances and maintains channel and habitat complexity. Natural sediment transport based on local geology, soils and stream gradient. High resilience (ability to recover from disturbance) to natural and anthropogenic watershed stressors (Flotemersch et al. 2016). Watershed integrity maintains disturbance levels within ranges tolerable by endemic taxa and promotes connectivity for purpose of recolonization.
Ecosystem function	Watershed supports full range of ecological processes and functions essential to maintaining high biodiversity provided by a minimally disturbed ecosystem. Food web, nutrient and energy flow linkages between aquatic and terrestrial environments fully supported.
Biodiversity	Benthic macroinvertebrate community typically with high taxa richness, including many micro-habitat specialist taxa and taxa sensitive to human disturbance. Habitat complexity results in diversity of both rheophilic and lotic-depositional taxa. Non-native, invasive taxa not present.

As reservoirs of biotic diversity, BCG level 1 streams and their AI communities are worthy of recognition for conservation planning, but they must be identified first (Vorosmarty et al. 2010). How many BCG level 1 streams exist today in the maritime PNW (Figure 1) and how threatened they are is unknown. Their occurrence is likely to be sporadic in the landscape, often due to human factors as land ownership or difficulty of access, and natural watershed variables such as a primarily spring-fed hydrology or north facing aspect.

Time is of the essence for identifying and securing reservoirs of freshwater biodiversity into a protected area network. For example, McRae et al. (2017) estimated that global populations of vertebrate species associated with freshwater ecosystems have declined by 81% in the Anthropocene. BCG level 1 watersheds are biodiversity museums in the landscape that maintain a broader spectrum of populations and species, and hopefully for dispersal into and recolonization of restored habitat. As Aldo Leopold aptly noted, "The first rule of intelligent tinkering is save all the pieces".

Howard et al. (2018) prioritize California watersheds for conservation planning by analysis and review of a broad array of watershed level information along with faunistic and floristic inventories. Their faunistic inventories focus primarily on herpetofauna, fishes, and freshwater-dependent mammals. Selected invertebrate data is reviewed at a coarse taxonomic level (family). California's comprehensive framework identifying a network of watersheds for priority conservation is the result of years of study and the cooperation of many stakeholders. Conservation planning for freshwater biodiversity in the maritime PNW has focused primarily on salmonid fish. Incorporating AI into freshwater biodiveristy assessments in the PNW has usually entailed a species by species approach to inventory for a short list of sensitive species that are perceived to be rare and vulnerable , e.g. the Interagency Special Status/Sensitive Species Program (ISSSSP) of the U.S. Forest Service and Bureau of Land Management, https://www.fs.fed.us/r6/sfpnw/issssp/ .

We propose using community level benthic macroinvertebrate (BMI) data collected for a variety of reasons over the past 4-5 decades in the PNW to help identify watersheds with exemplary BCG level 1 biodiversity. Outside of fish census data, BMI data may be the most readily available snapshot of freshwater life across much of North America. The BCG level 1 Biodiversity Index outlined here mines BMI data to detect a signal when a BCG level 1 watershed with exemplary biodiversity has potentially been encountered. A signal, in that a "positive hit" in a particular watershed is in no way definitive on its own, but will hopefully instigate a more thorough inventory of the biota and habitat. Objectives and utility for the index are:

1. For regions such as California that are more advanced in gathering information on aquatic biodiversity statewide, the index is an ancillary tool for incorporating BMI data into the inventory and decision making process.

2. In regions that do not have a formalized process for identifying high priority watersheds for conservation, this index can be used by itself to alert stakeholders of sites with probable exemplary biodiverity. Hopefully a fuller investigation is triggered.

This BCG level 1 signal presumes that if exemplary biotic diversity is found in a stream reach used for biomonitoring, that it is usually not localized; it most likely extends into riparian and upslope habitats, and upstream into headwater tributary channels, springs and seeps. Gravity's consequences and the cumulative nature of lotic ecosystems, make this hypothesis highly probable. However, there may be

examples where islands of biodiversity exist downstream from upper watersheds extensively disturbed by human activities, e.g. where habitat heterogeneity is augmented by side-channel restoration.

An important caveat for using this index is that it will produce false negatives, meaning it failed to signal the presence of exemplary biodiversity at a site. Current agency and institutional protocols for collecting and processing BMI samples means a limited area is sampled at a stream site (usually 3-11 square feet); may or may not focus on a single mid-stream habitat type like rifles; typically subsamples the field collected sample down to 300-600 organisms; and taxonomic resolution may be coarse in many invertebrate groups. Thus, the snapshot taken of the BMI community has a limited field of view. More recently applied protocols randomly sample a variety of stream habitats, acquire a more robust subsample, and identify the bulk of the taxa present to the genus, species group, or species level.

Though developed more specifically for the maritime Pacific Northwest region, the index is more generally aplicable to mid-order, perennial, forested, montane streams in the Northwestern Forested Mountains and Marine West Coast Forest Ecoregions (Level 1 Ecoregions, US EPA 2016). Mid-order, wadeable streams is the general habitat where BMI biomonitoring sampling has been concentrated. It is not appropriate to apply it to small seasonal or perennial streams of <1 m base flow width, springs, seeps, and riverine habitat (i.e. base flow width >15 meters). Many of the noteworthy taxa identified here are associated with spring and small stream habitat, but usually their longitudinal range extends into mid-order stream reaches.

As aquatic ecologists in the PNW intimately involved with benthic macroinvertebrate biomonitoring, from time-to-time we have stumbled upon watersheds with outstanding and exemplary biodiversity on public lands. How to convey our excitement of this discovery to land managers and stakeholders is problematic. Numerical indices of biological integrity, though useful for trend monitoring and other purposes, don't contribute much to an understanding of the full biotic diversity possible in a catchment and the potential conservation significance. Narratives and definitions attached to BCG level 1 and hopefully the Biodiversity Index may be more digestible by the general public and inform management decisions on conservation and protection. Through this BCG Level 1 Biodiversity Index, we attempt to define measurable biological indicators, or signals, for a benthic macroinvertebrate community comparable to BCG level 1, an exemplary community that has not been impacted by human disturbance.

2 BCG Level 1 Biodiversity Index

The BCG level 1 Biodiversity Index presented here (draft, version 1) is comprised of twelve metrics (Table 2). The metrics are derived from standard benthic biomonitoring data. I believe these metrics are positively correlated with overall habitat complexity/resilience and high biodiversity (a belief based on 35 years of an informal matching of field observations on habitat structure of PNW streams with a parallel analysis of their AI community from biomonitoring samples). It is a starting point and hypothesis awaiting a more formal quantitative analysis.

We evaluated the distribution of metric values across BCG levels in the Puget Lowlands/Willamette Valley (PL/WV) BCG calibration dataset to help inform metric selection and scoring (see box plots below). The metrics are scored individually and then summed to get an overall score (with higher scores indicating higher biodiversity/stronger BCG level 1 signal). The overall score is broken into three ratings -

high, medium and low (Table 3). Descriptions of each of the twelve metrics (and rationale for inclusion) are included below. The index scores were calculated using the 'metric_scores' function in the BioMonTools R package (https://github.com/leppott/BioMonTools).

One of the metrics – the 'noteworthy taxa richness' - was developed specifically for the BCG level 1 Biodiversity Index for the maritime Pacific Northwest. A list of the noteworthy taxa and the rationale for including them can be found in Attachment A.

Noteworthy taxa are -

- Often rare or occasional in stream biomonitoring samples. Many are believed to be truly rare as demonstrated by decades of targeted surveys and collecting efforts by regional experts in aquatic ecology.

- Most have a geographic distribution (range) restricted to a portion of the Northwestern Forested Mountains and Marine West Coast Forest Ecoregions (Figure 1); or populations have disjunct or patchy distributions in western North America; or have a total range isolated to a few watersheds.

- Includes taxa with lotic-rheophilc , lotic-depositional, and lotic-specialist habitat affiliation (lotic-specialists for example are associated with hyporheic, debris jams, wetted margins, madicolous, torrential, liverwort or moss microhabitats).

- Symptomatic of high habitat heterogeneity and resilience in a watershed.

- May have unusual life history adaptations.

- Include many taxa indicative of habitat stability and resilience, e.g. that are semivoltine; medium to large-size at maturity; and prefer cold stenothermal or cool eurythermal habitats (Poff et al. 2006).

When applying the index, it is important to keep in mind the following caveats -

- The BCG level 1 Biodiversity Index is based purely on my best professional judgement based on 35 years of field experience and research throughout the maritime Pacific Northwest Region. Metrics, scoring criteria and noteworthy taxa will undoubtedly require adjustment (particularly in version 1). Your review with specific comments, edits and recommendations is requested.

- Scoring criteria is set to be more inclusive of streams at all elevations in the maritime Pacific Northwest, not just lower elevations in the Willamette Valley & Puget Lowlands.

- **500-600 count subsample**. The scoring thresholds are based on routine, 500-600 count subsamples, which is a limited inventory of the total AI biota present.

- An uneven taxonomic effort is applied to these samples, from species to phyla level identification.

- **Stream size**. Pertains to mid-order watersheds with perennial channels from about 1 meter to 15 meters wide at base flow, i.e. not very small headwater streams, or larger, more open streams and small rivers.

- **Gradient and substrate**. Application is limited to moderate to high gradient streams with coarse mineral substrates and extensive erosional habitat. Low gradient streams with mostly fine substrates are not included here, as we currently have a poor understanding of their AI communities and biodiversity potential.

- False negatives in the signal are likely to occur often because of the limited census of the total biota that the typical biomonitoring sample provides. False positives are unlikely, as sites with a strong signal based on a 500 organism subsample are at least exceptional, if not exemplary.

- In PNW watersheds BCG level 1 Biodiversity Index scores may decrease with increasing stream size, because fewer noteworthy taxa are likely to be encountered in larger streams. Most noteworthy taxa are associated with smaller streams, seeps and springs. Highest signal scores will likely be associated with mid-size streams (2-3 m width at base flow) where cumulative points from the twelve community composition metrics can be high, along with increased chances of encountering **noteworthy taxa**.

Table 2. BCG Level 1 Biodiversity Index (draft, version 1) - community composition metrics and scoring thresholds.

Metric	Scoring criteria (points)			
	0	**1**	**2**	**3**
Total taxa richness	<40	40-49	50-59	≥ 60
EPT taxa richness	<20	20-24	25-29	≥ 30
BCG attribute 1i, 1m & 2 taxa	<2	2-5	6-8	≥ 9
Shannon-Weaver diversity ($\log_e x$)	<2.75	2.75-2.99	3.0-3.24	≥ 3.25
Long-lived taxa richness	<6	6-8	9-11	≥ 12
Ephemerellidae taxa richness	0-2	3	4	≥ 5
Heptageniidae taxa richness	0-2	3	4	≥ 5
Nemouridae taxa richness	0-2	3	4	≥ 5
Perlidae taxa richness	0	1	2	≥ 3
Rhyacophila taxa richness	0-2	3-4	5	≥ 6
Predator taxa richness	<10	10-12	13-15	≥ 16
Noteworthy taxa richness	Add an additional score point for each noteworthy taxa present			

*The list of noteworthy taxa and rationale for their inclusion can be found in Attachment A

Table 3. BCG Level 1 Biodiversity Index (draft, version 1) – overall scores and ratings, as well as recommendations.

Rating	Score	Description
High	≥ 30	Exemplary biodiversity and high habitat complexity and resilience probable. Acquire additional information and data on the site and watershed that is readily available. Alert stakeholders, including government and non-government organization conservation agencies.
Medium	21-29	Moderate habitat complexity/resilience and biodiversity indicated. Further evaluation of the site and watershed is recommended.
Low	<20	Unexceptional biodiversity indicated. Mostly widespread and common taxa present.

Input metrics

Metric # 1 - Total taxa richness

A lengthy taxa list in itself does not automatically confer exemplary biodiversity, though it is a positive signal. Taxa lists from stressed habitats can be padded by mostly widespread, common, generally tolerant, habitat generalist taxa of little conservation concern. In general though, increasing levels of human stress on lotic ecosystems translates to lower total taxa richness (Figure 3).

Scoring criteria used here is based on biomonitoring samples where a 500-600 organism subsample is taken, processed with 500 micron mesh, and an approximate Pacific Northwest Aquatic Monitoring Partnership (PNAMP.org) PNAMP level 2 standard taxonomic effort is applied. An upward creep in total taxa richness over the past few decades because of finer level identifications being applied to certain groups in more recent years is noted.

We have encountered candidate BCG level 1 stream sites at mid-elevations in western Oregon and northern California that have 80-95 total taxa in a single biomonitoring sample.

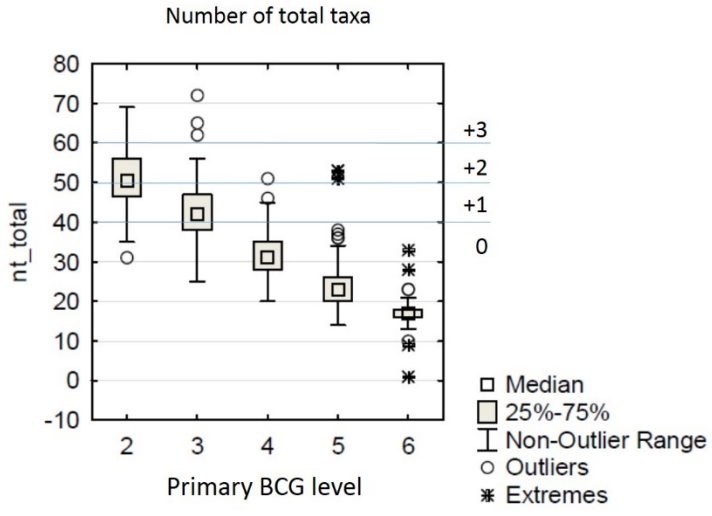


Figure 3. Box plot of number of total taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 2 - EPT taxa richness

Ephemeroptera+Plecoptera+Trichoptera (EPT) taxa richness is a widely used metric for assessing benthic macroinvertebrate communities in lotic ecosystems. Taxa in these insect orders are often dominant numerically and taxonomically in benthic biomonitoring samples from mid-order streams. These orders contain many sensitive and/or noteworthy taxa, and are comprised mainly of lotic-rheophilic, lotic-depositional, and lotic-specialist taxa. Compared with total taxa richness, this metric is probably less noisy for evaluating trends or detecting signals. Refinements and nomenclature upgrades of the standard taxonomic effort over the past few decades means there has been some upward creep in the average EPT richness values.

In general, increasing levels of human stress on lotic ecosystems translates to lower EPT taxa richness (Figure 4). BCG level 2 streams in the Puget Lowlands and Willamette Valley have a median value of about 24 EPT taxa. We have encountered exceptional streams at mid-elevations in western Oregon with 40-50 EPT in a single biomonitoring sample.

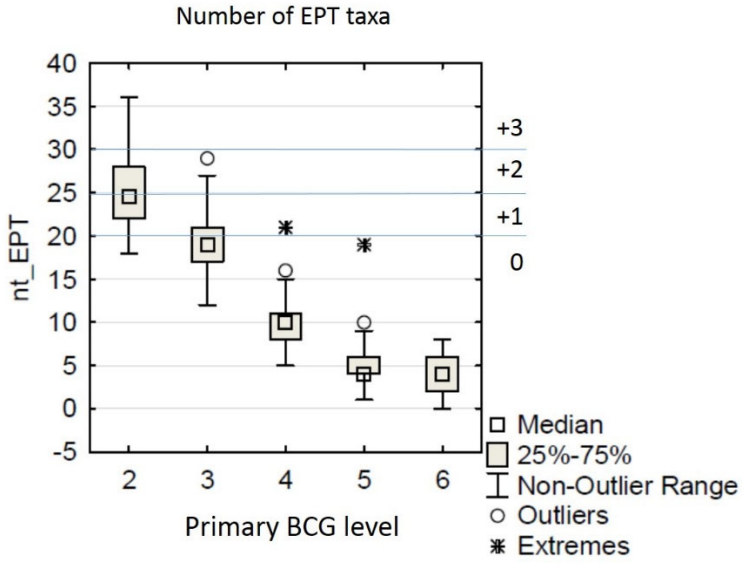


Figure 4. Box plot of number of EPT taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 3 - BCG attribute Ii, 1m & II taxa richness

Attribute Ii, Im and II taxa (BCG report – Stamp and Gerritsen 2019) are expected to be a dominant component of Level 1 stream communities. These are often regionally endemic or uncommon taxa of particular conservation concern. Most of these taxa also appear on the noteworthy taxa list. Along the human disturbance gradient in the Puget Lowlands and Willamette Valley, attribute Ii, 1m & II taxa richness drops precipitously between Level 2 & 3 streams and are mostly absent in the more disturbed BCG level 4-6 streams (Figure 5).

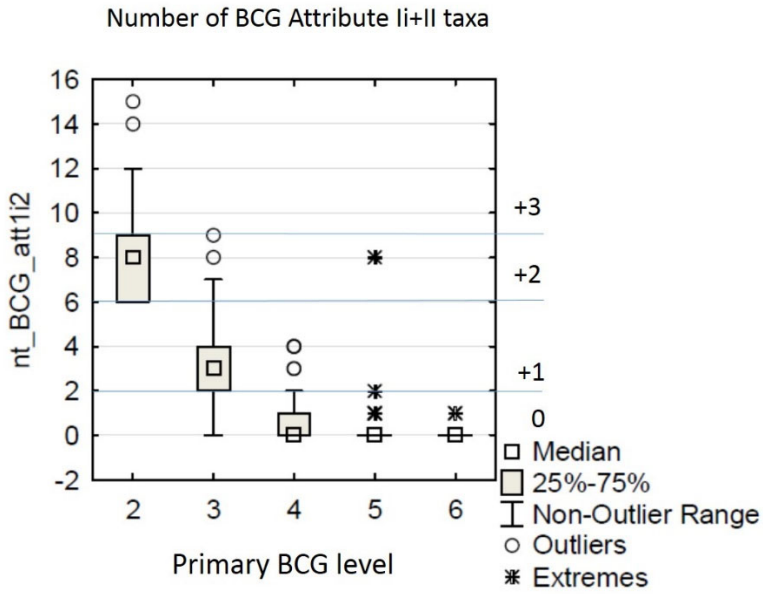


Figure 5. Box plot of number of BCG attribute Ii+II taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 4 - Shannon-Weaver Diversity Index (loge)

We have observed over the years that benthic macroinvertebrate communities from least disturbed sites with high habitat complexity and high resilience to environmental extremes such as fire, floods and droughts, usually have species abundance distributions showing a more gradual tapering of abundance than highly disturbed sites. Human disturbance typically leads to a more homogeneous habitat structure, and species abundance displays a rapid drop in abundance after the first few super-dominant species.

Ecological diversity indices mathematically describe the species abundance distribution of a community. We chose the Shannon-Weaver Diversity Index (loge) to incorporate into the BCG Level 1 signal. Index values are negatively related to increasing levels of human disturbance (Figure 6). Variation in index values increases with increasing levels of human stress.

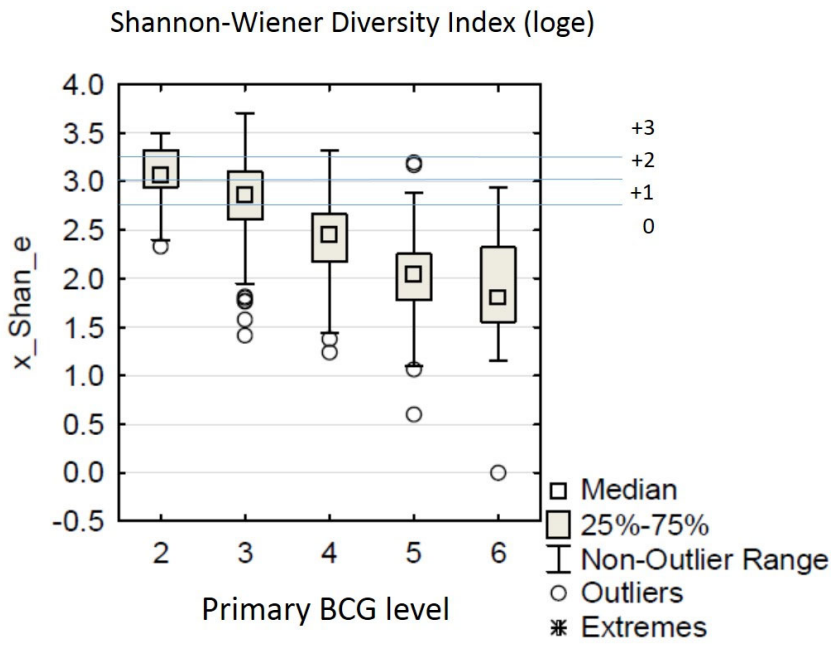


Figure 6. Box plot of Shannon Wiener diversity index (loge) vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 5 - Long-lived taxa richness

Long-lived or semivoltine taxa have a substantial proportion of their populations requiring more than one year to complete a life cycle. Stream channels and their riparian corridors that have a multitude of moderately to highly stable (resilient to disturbance) habitats and micro-habitats support more semivoltine taxa, many of which are noteworthy taxa. High long-lived taxa richness appears to be correlated with sites displaying exemplary biodiversity. A high proportion of the AI species that are considered to be vulnerable or imperiled are long-lived (NatureServe.org , see noteworthy taxa list).

There is a strong negative relationship between long-lived taxa richness and increasing levels of human disturbance in the Willamette Valley and Puget Lowlands calibration data set (Figure 7). A median value of 10 long-lived taxa is found in Level 2 streams. At high quality stream sites at mid-elevations in western Oregon and Washington, 15-20 long-lived taxa may be present in a single biomonitoring sample.

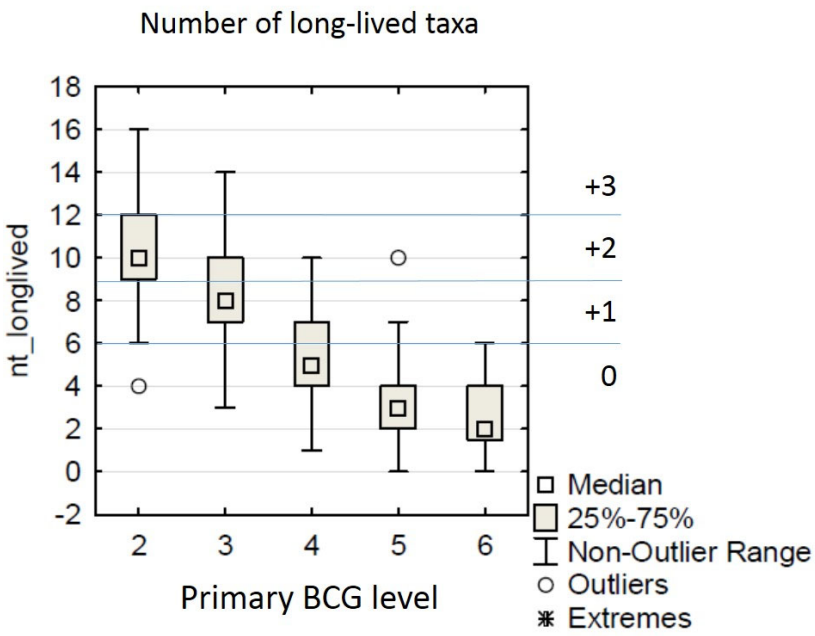


Figure 7. Box plot of number of long-lived taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 6 - Ephemerellidae taxa richness

Mayflies of the family Ephemerellidae are ubiquitous in forested, montane streams in temperate zones around the world. There is an exceptional richness of ephemerellid species in high quality streams in the Marine West Coast Forest and Northwestern Forested Mountains Ecoregions, with peak diversity occurring in mid-order streams. Many of these species are sensitive taxa (attribute Ii, Im, 2, and 3); most are associated with lotic-rheophilic habitats, but some are lotic-depositional; some are stellar cold-cool water indicators since they are nearly ubiquitous in western montane streams; and collectively they have a diversity in spatially and temporally related life history and feeding strategies.

High ephemerellid taxa richness in our experience is correlated with sites with high habitat complexity and resilience and with high overall biodiversity. Species tend to disappear in a predictable manner along a disturbance gradient, whether the disturbance is human or naturally derived. In the lower elevation and collectively more human disturbed Willamette Valley and Puget Lowland stream sites, ephemerellids are found almost exclusively in Level 2 and 3 streams, with a median value of ephemerellid taxa richness of 2.5 and 1, respectively (Figure 8). Higher quality streams at mid-elevations in the maritime PNW typically have 4-6 ephemerellid taxa per 500-600 count biomonitoring sample. PNAMP-level 2 standard taxonomic effort for ephemerellids in biomonitoring samples is mostly species or species groups.

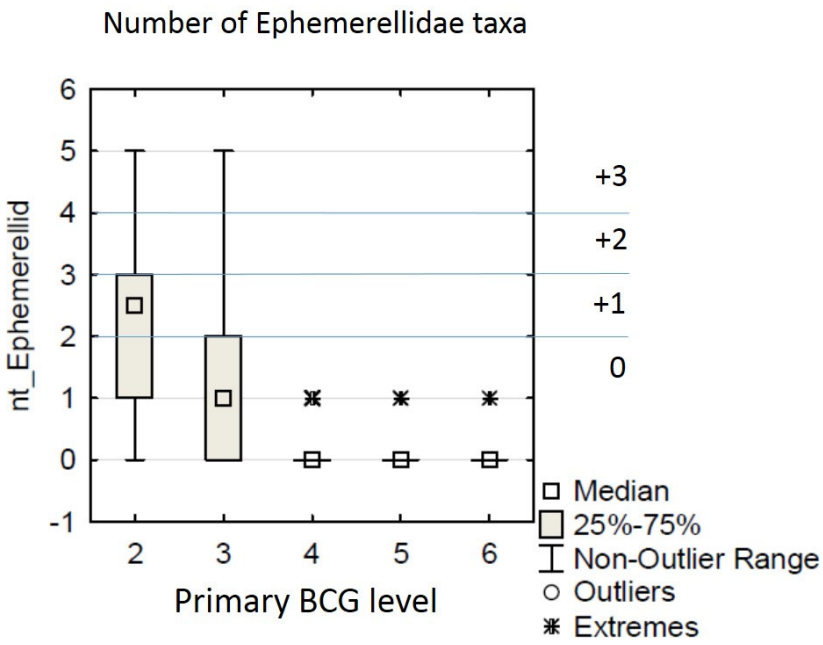


Figure 8. Box plot of number of Ephemerellidae taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 7 - Heptageniidae taxa richness

Mayflies of the family Heptageniidae are also ubiquitous in forested, montane streams in temperate zones around the world, and also have exceptional richness of species in high quality streams in the Marine West Coast Forest and Northwestern Forested Mountains Ecoregions, with peak diversity occurring in mid-order streams. Many of these species are sensitive taxa (attribute Ii, Im, 2, and 3); all are associated with lotic-rheophilic habitats; all are of the scraper feeding group; taxa like *Epeorus grandis group* are stellar cold-cool water indicators because of their ubiquity in western, montane streams; and collectively they have a diversity in spatially and temporally related life history strategies.

High heptageniid taxa richness in our experience is correlated with sites with high habitat complexity and resilience and with high overall biodiversity. Species tend to disappear in a predictable manner along a disturbance gradient, whether the disturbance is human or naturally derived. In the lower elevation and collectively more human disturbed Willamette Valley and Puget Lowland stream sites, heptageniids are found in Level 2, 3, and 4 streams, with a median value of ephemerellid taxa richness of 2.5, 2, and 0.5 respectively (Figure 9). Higher quality streams at mid- and higher elevations in the maritime PNW typically have 4-6 heptageniid taxa per 500-600 count biomonitoring sample. PNAMP-level 2 standard taxonomic effort for heptageniids in biomonitoring samples is genus, species group or species.

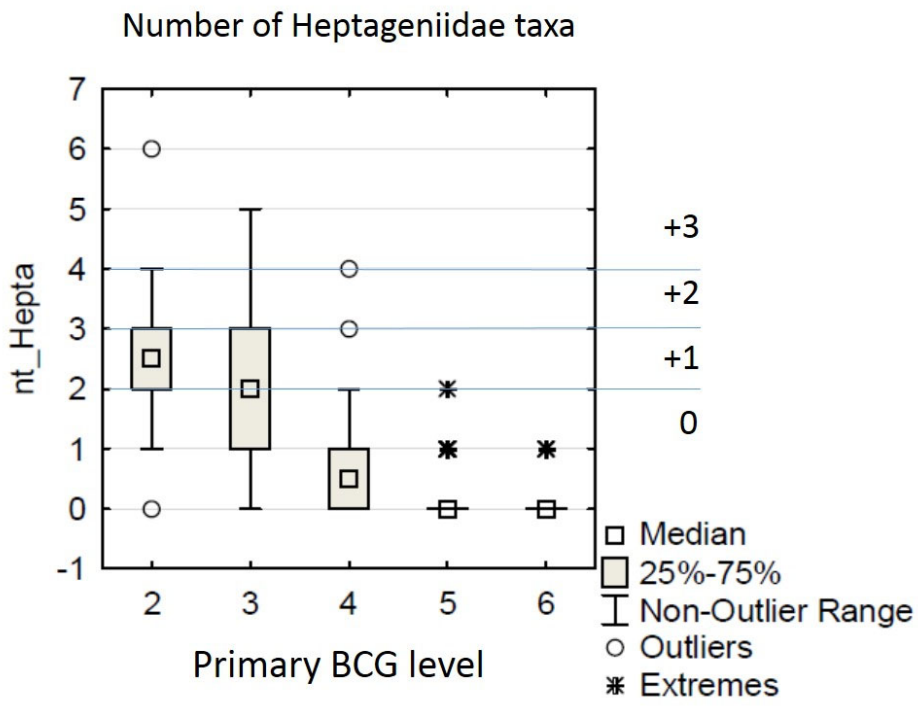


Figure 9. Box plot of number of Heptageniidae taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 8 - Nemouridae taxa richness

Nemourid stoneflies are again ubiquitous in forested, montane streams in temperate zones around the world, and also have exceptional richness of species in high quality streams in the Marine West Coast Forest and Northwestern Forested Mountains Ecoregions, with peak diversity occurring in mid-order streams. The family also has numerous species associated with seasonal and very small perennial streams, and there are also species that extend downstream into riverine environments. Some nemourids are sensitive taxa (attribute Ii, Im, 2, and 3); are associated with lotic-rheophilic, lotic-depositional and even lentic habitats; all are of the shredder feeding group; taxa like *Zapada columbiana* and *Visoka cataractae* are stellar cold-cool water indicators because of their ubiquity in western, montane streams; and collectively they have a diversity in spatially and temporally related life history strategies.

High nemourid taxa richness in our experience is correlated with sites with high habitat complexity and resilience and with high overall biodiversity. Species tend to disappear in a predictable manner along a disturbance gradient, whether the disturbance is human or naturally derived. In the lower elevation and collectively more human disturbed Willamette Valley and Puget Lowland stream sites a strong negative relationship with the human disturbance gradient is not evident (Figure 10). Nemourid taxa are found across all BCG levels and median taxa richness varies between 1 and 2 taxa. *Malenka* and *Zapada cinctipes* are tolerant taxa that occur across all BCG levels. However, higher quality streams at mid- and higher elevations in the maritime PNW typically have 3-5 nemourid taxa per 500-600 count

biomonitoring sample. PNAMP-level 2 standard taxonomic effort for nemourids in biomonitoring samples is genus, species group or species.

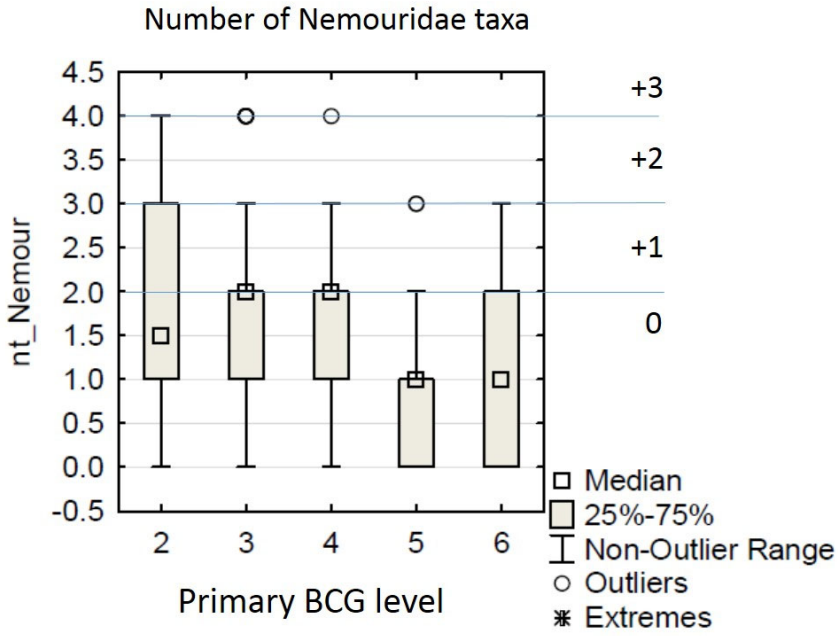


Figure 10. Box plot of number of Nemouridae taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 9 - Perlidae taxa richness

Perlid stoneflies are large, predaceous, semivoltine taxa that are ubiquitous in forested, montane streams in temperate zones around the world. Maximum species diversity in Marine West Coast Forest and Northwestern Forested Mountains Ecoregion streams is about 3 taxa; and includes *Calineuria californica*; 2 geographically separated *Doroneuria* species; a widely distributed western montane species, *Hesperoperla pacifica*; and a species isolated in the Sierra Nevada Mountains, *Hesperoperla hoguei*. Colder, high elevation streams are typically restricted to just *Doroneuria*.

Perlid taxa richness of 2-3 species in our experience is correlated with sites with high habitat complexity and resilience and with high overall biodiversity. An absence of perlids in hard-bottomed streams in montane western North America is a definite signal of disturbance of some kind. Willamette Valley and Puget Lowland stream sites have a median value of 2 species for Level 2 streams, 1 for Level 3, and absent for levels 4-6 (Figure 11). Mid-elevation sites with 3 perlid species almost always display high overall biodiversity. PNAMP-level 2 standard taxonomic effort for perlids in biomonitoring samples is effectively species.

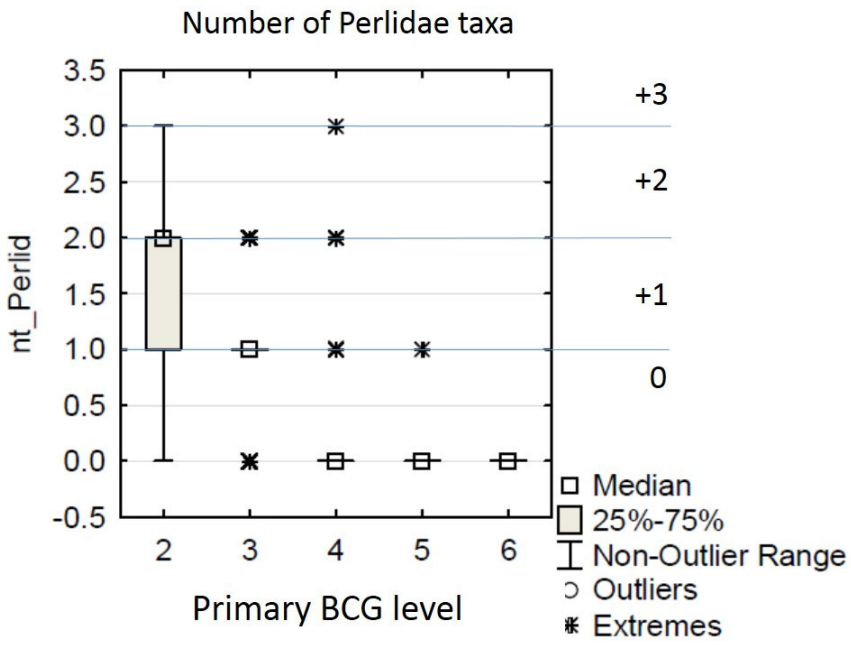


Figure 11. Box plot of number of Perlidae taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 10 - Rhyacophila taxa richness

Rhyacophila is a diverse genus of free-living caddisflies found in montane streams and rivers throughout the northern hemisphere. They are mostly predaceous, primarily associated with lotic-rheophilic habitats, and are primarily cold or cool water associated. Collectively they have a diversity in spatially and temporally related life history strategies and habitat associations. There are over 100 *Rhyacophila* species known from western North America. Many species are widespread and common in western North America. Conversely, many are noteworthy taxa with Restricted or Isolated distributions.

Species tend to appear or disappear in a predictable manner along gradients of human disturbance, stream water temperature, stream slope, and longitudinal and altitudinal gradients. High *Rhyacophila* taxa richness in our experience is correlated with sites with high habitat complexity and resilience and with high overall biodiversity. Willamette Valley and Puget Lowland stream sites have a median value of 3.5 species for Level 2 streams, 3 for Level 3, 1 for Level 4, and none for Levels 5-6 (Figure 12). There are some *Rhyacophila* taxa that are relatively tolerant of human disturbance and are nearly ubiquitous in all but the most severely disturbed valley bottom and foothill streams. High quality mid- and higher elevation forested, montane streams typically have 5-8 *Rhyacophila taxa*. PNAMP-level 2 standard taxonomic effort for *Rhyacophila* in biomonitoring samples is species group and species.

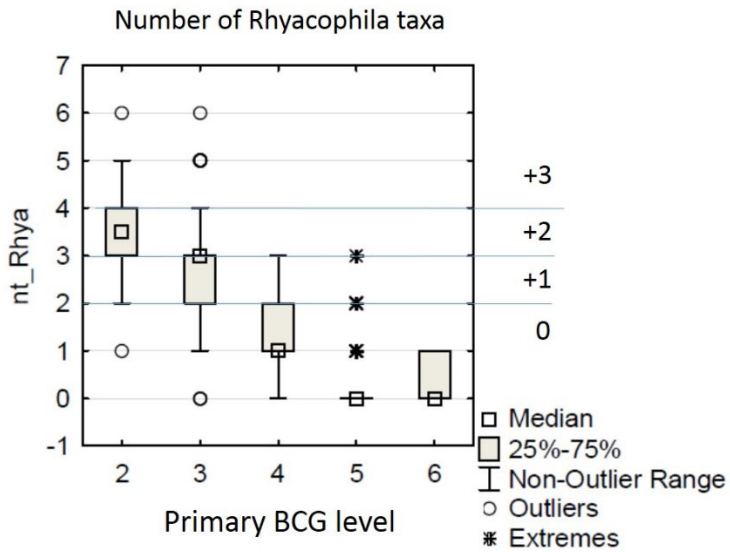


Figure 12. Box plot of number of Rhyacophila taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 11 - Predator taxa richness

The proportion of predators (%) in a benthic macroinvertebrate community has been positively correlated with biological integrity of flowing waters (Karr 1999). Predator taxa richness also appears to be positively correlated with overall habitat complexity and resilience and overall biodiversity. In Willamette Valley and Puget Lowland streams there is a strong positive relation between predator richness and BCG levels, going from a median of 14 taxa in Level 2 streams down to 3 in Level 5 & 6 streams (Figure 13). High quality mid- and higher elevation forested, montane streams can have 20-25 predator taxa.

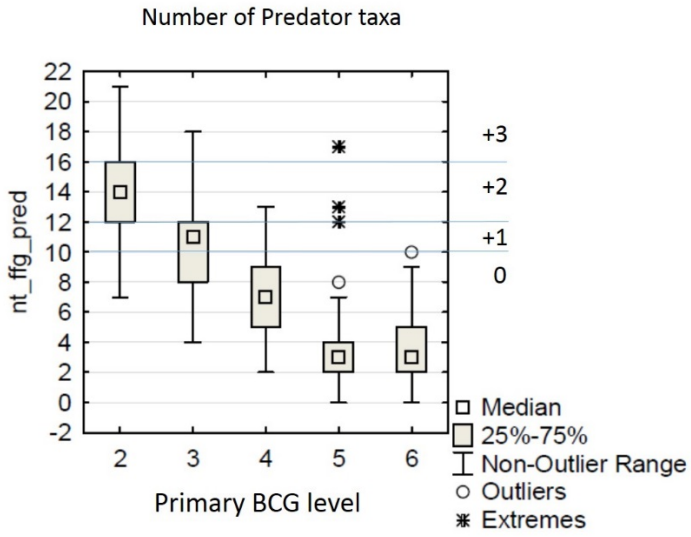


Figure 13. Box plot of number of predator taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

Metric # 12 – Noteworthy taxa richness

Attachment A lists noteworthy taxa that appear to occur most often at sites with complex and resilient habitat structure that supports high overall freshwater biodiversity. Most are rarely or occasionally encountered in benthic macroinvertebrate biomonitoring samples from mid-order, forested streams in the Marine West Coast Forest and Northwestern Forested Mountains Ecoregions.

Figure 14 provides the median number and ranges of noteworthy taxa encountered in the calibration BMI data set from the Willamette Valley and Puget Lowlands. The median value for the few available Level 2 streams is only 0.5 taxa, and for all other BCG levels it is 0. Ranges, outliers, and extremes are high. This result underscores that much of the AI biodiversity in mid-order streams in these two lowland ecoregions has probably disappeared.

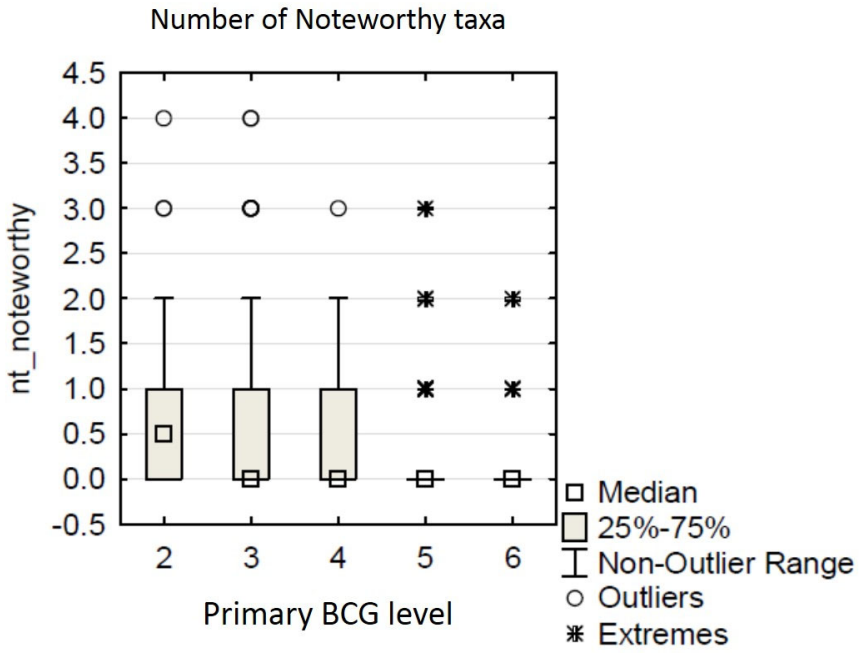
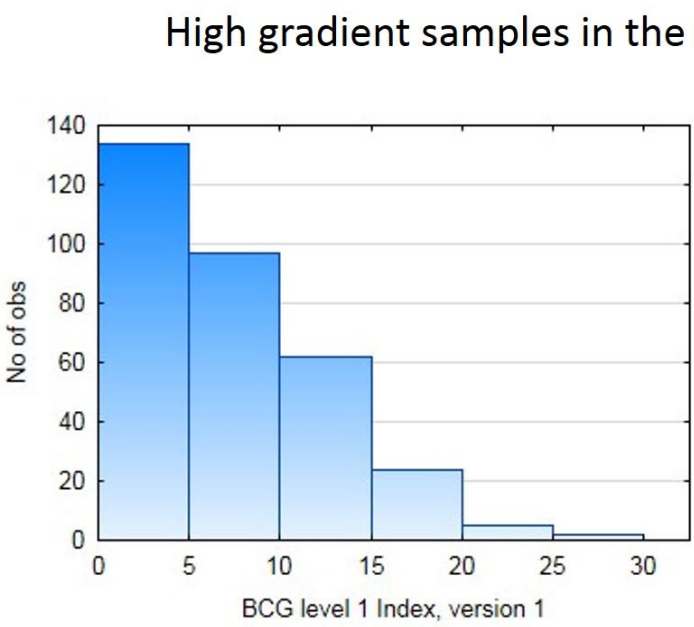


Figure 14. Box plot of number of noteworthy taxa vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the Willamette Valley and Puget Lowlands BCG calibration dataset (sample size = 426).

3 Index Scores for the Puget Lowlands/Willamette Valley BCG Calibration Dataset

When the BCG level 1 Biodiversity Index was calculated for the 426 high gradient samples[1] in the WV/PL calibration data set, scores were distributed as shown in Figure 15. There were no signals for streams with high potential biodiversity, but two sites came close with scores of 27 (BIO06600-TUMW02 in WA and Roaring Creek (Tualatin) Station 12215 in OR, which are shown as large green circles in Figure 16). The preponderance of very low scores mirrors the finding of few noteworthy taxa in streams in this representative data set. The full set of scores for the PL/WV BCG calibration dataset are provided in Attachment B. In addition, we examined the distribution of BCG level 1 Biodiversity Index scores across BCG levels and found good correspondence (index scores followed the expected pattern, with highest median index scores occurring in BCG level 2 samples and lowest in BCG level 5-6 samples; Figure 17).



Rating	Index score	# of samples
High	30+	0
Medium	25-29	2
	20-24	8
Low	15-19	33
	10-14	72
	5-9	95
	1-4	114
Zero	0	102
Total		**426**

Figure 15. Distribution of index scores for samples collected at high gradient sites (≥1% slope), based on the BCG calibration dataset (n=426 samples, from 162 unique sites).

[1] High and low gradient streams were defined based on the NHDPlus V2 flowline slope attribute, using 1% as the threshold (low (<1% slope) and high (≥1% slope).

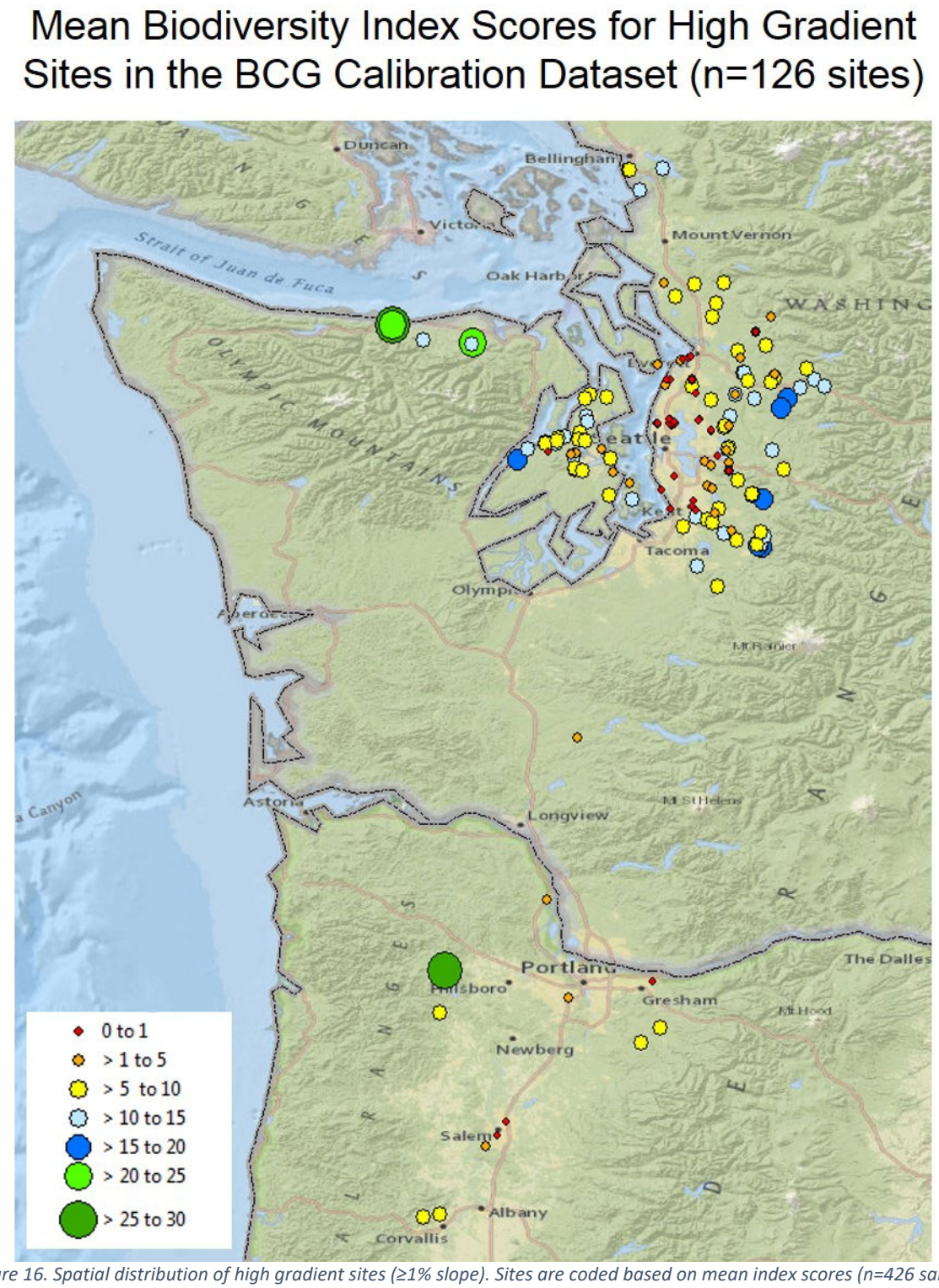


Figure 16. Spatial distribution of high gradient sites (≥1% slope). Sites are coded based on mean index scores (n=426 samples, from 162 unique sites), where sites with the highest index scores are shown with large green circles and sites with the worst scores are shown by small red or orange circles.

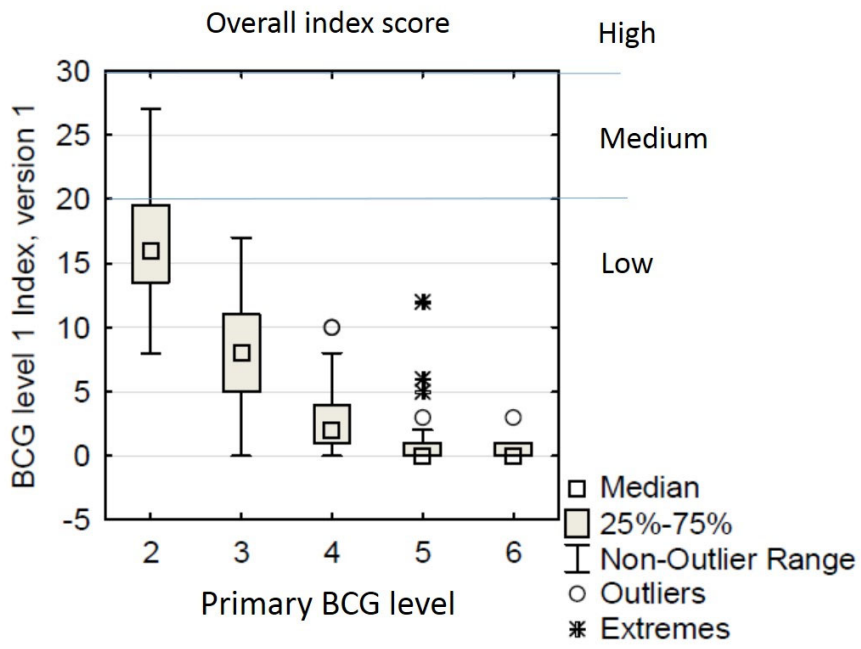


Figure 17. Box plot of the BCG level 1 Biodiversity Index vs. BCG Level, based on high gradient (≥ 1% slope) calibration samples in the PL/WV BCG calibration dataset (sample size = 426).

4 Concluding Remarks

Our experience from processing thousands of biomonitoring samples from the PL/WV Ecoregions is that the typical BMI community is greatly homogenized, composed of taxa that:

- Have a wide geographic range, either common or widespread throughout most of western North America (WEST), or have transcontinental, Holarctic or cosmopolitan distributions (WIDE).
- Are common (COMM) or nearly ubiquitous (UBIQ) in mid-order WV/PL streams.
- Have habitat affiliations dominated by taxa that are generalists (GENE), found in a wide variety of lentic habitats and in slack-water areas of streams and rivers; or lotic generalists (LOGE), found frequently in both depositional and erosional habitats in streams. There are few taxa with more specialized habitat affiliation.
- Multivoltine (MUL) life cycles dominate, followed by univoltine (UNI). Fewer semivoltine (SEM) taxa are present.
- Small (SMA) and medium (MED) size taxa dominate. Few large taxa are present.
- Cool eurythermal (COOL) and broadly eurythermal (WARM) taxa dominate.

The exception to the above are streams in the PL/WV ecoregions that are dominated by the tolerant pleurocerid snail *Juga*, which has a more restricted west coast distribution, is semivoltine, and is large.

After 200 years of increasing human disturbance in the maritime PNW, most of the remaining reservoirs of AI biodiversity reside in the forested, montane watersheds at mid-elevations that surround the PL/WV ecoregions. The low biodiversity index scores and presence of few noteworthy taxa in the PL/WV ecoregions raises some conservation concerns.

If we capitulate on efforts to preserve mid-order stream AI biodiversity within the borders of the PL/WV ecoregions, will species unique to these regions become vulnerable? How important is a network of conservation watersheds in these lowland ecoregions to preserving overall AI biodivesity in the maritime PNW? We don't know. However, our sense is that the vast majority of the species/taxa on the noteworthy taxa list (particularly insects) occur in watersheds from sea level to about 1500 meters elevation. AI taxa unique to the PL/WV ecoregions are more likely to occur in other freshwater habitat types, e.g. riverine or lentic habitats. Still, an assessment of the conservation status of AI species unique to these lowland regions would be timely.

Population and development pressures on freshwater environments in the PL/WV ecoregions are predicted to continue unabated and even accelerate, as is the impacts of climate change. By default mid- elevation watersheds will probably become the reservoir of last resort for AI biodiversity in PNW streams. This index is designed to help identify watersheds, particularly at mid-elevations, that harbor exemplary AI biodiversity, and hopefully bring them into a protected conservation network.

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