# Using Aquatic Macroinvertebrates as Indicators of Stream Flow Duration

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## Goals

The goal of this project is to provide a recommended aquatic invertebrate taxa list, community composition indicators, field methodology, and scoring protocol that will enable practitioners to use invertebrates as one component of a multimetric assessment process to evaluate stream flow duration in Oregon. It is anticipated that most users will not be professional entomologists, but will undergo training in the entire stream assessment process. For speed and ease of assessment and for field applicability, it is further anticipated that the lowest taxonomic level to which most invertebrates will be identified is family. Identification to genus and/or species level is the most informative, but is impractical in the field, as it requires the use of a stereomicroscope as well as the skills of taxonomic experts. Also, species identification is not possible for immature larval forms, as they lack many of the distinguishing characteristics present in mature larvae and are too small to see easily.

The most basic question to be answered, therefore, is whether there are sufficient family-level differences in the macroinvertebrate communities typically inhabiting perennial, intermittent, and ephemeral waters to allow the use of these organisms to discriminate between stream types.

## Challenges

Clifford (1966) observed, "…the gradation from perennial streams to intermittent or especially from intermittent to ephemeral stream is often difficult to resolve objectively." Some challenges to be overcome in successfully using macroinvertebrates as indicators of stream duration include:

### *Between-stream variability*

It is desired that the assessment method and community characteristics be applicable to streams across the state of Oregon. However, a great deal of temporal and spatial variation exists normally both within and between stream reaches, making it difficult to determine invertebrate community indicators that will be robust enough to be used in streams across Oregon's multiple and varied ecoregions.

### *Within-stream variability*

Biological communities vary randomly to some extent, as well as varying in response to biotic interactions, abiotic factors, habitat differences, and environmental influences. Different communities tend to be present at different times of the year, and in different types of in-stream habitats (i.e. riffles versus pools). Macroinvertebrate indicators must be found that are applicable to multiple different reaches within a single stream.

### *Taxonomic constraints*

Most macroinvertebrate families contain genera and species with a variety of life history characteristics. Some genera possess adaptations that allow them to resist desiccation and develop successfully in intermittent waters, while other genera in the same family may lack such adaptations and be restricted to permanent streams. The family-level identification that is desired for the Oregon Draft Stream Assessment will not be able to take such genus- and species-level differences in life histories into account.

### *Key indicator taxa*

Wiggins *et al*. (1980) maintains that the harsher, more variable environmental conditions in temporary waters impose a high enough degree of constraint that only a limited subset of invertebrate species can survive in these habitats. However, a thorough literature review reveals few key taxonomic groups that differentiate consistently, predictably, and unequivocally between perennial, intermittent, and ephemeral streams.

## Invertebrate communities in perennial vs. temporary waters

Much of the literature addressing invertebrates and stream duration compares invertebrate communities between perennial and intermittent streams or in intermittent streams during drought versus flood years. Literature addressing macroinvertebrates in ephemeral streams is limited (Savage & Rabe, 1979; Bonada *et al*., 2007). Comparative studies of invertebrate community composition in perennial and intermittent streams frequently reveals a high degree of overlap (Williams, 1987; Delucchi & Peckarsky, 1989; Boulton & Lake, 1992; Dieterich, 1992; Feminella, 1996; Banks, 2005; Beche *et al*., 2006), with the majority of taxa being common to both types of streams. Many species in these studies could be classified as "facultative", able to occupy both perennial and intermittent streams. This similarity in fauna is due in part to colonization of intermittent streams by aerial movement of invertebrates from nearby perennial waters, particularly those possessing adaptations that render them better able to survive in temporary environments, such as a univoltine life cycle, highly mobile adults, rapid growth in winter and spring (i.e. during the wet seasons), and deposition of eggs in moist substrate in early summer (Clifford, 1966). Ephemeral streams are generally considered to be either completely lacking in aquatic invertebrates, or to have a limited number of adventitious species that can complete their life cycles rapidly before the stream dries (Dieterich, 1992; Ohio EPA, 2002; Bonada *et al.*, 2007).

### *Seasonal changes*

One complicating factor in identifying key indicator taxa for perennial and intermittent waters is the normal replacement of taxa that occurs seasonally, as flow velocities and water levels change in both perennial and intermittent channels. Many types of streams show characteristic differences in the taxa present during the wet-season versus the dry-season (Clifford, 1966; McElravy *et al*., 1989; Boulton & Lake, 1992; Miller & Golladay, 1996; Bonada, 2003; Boulton, 2003; Beche *et al*., 2006; Bogan & Lytle, 2007). Macroinvertebrate community composition in intermittent streams is thought to follow a seasonal succession pattern, as the streams move from conditions of high flow in the fall and winter, though a low flow to standing pool stage in spring, and a summer-dry stage where standing water is absent and terrestrial invertebrates can colonize the dry channel (Williams & Hynes, 1976b; Williams, 1996). Changes in water levels, even in streams that maintain year-round flow, can lead to multiple associated physico-chemical changes. Flooding and drying is accompanied by changes in pH, dissolved oxygen, conductivity, siltation level, and concentrations of ions, toxins, or pollutants (Williams, 1987; Stanley *et al*., 1994: Lake, 2000), and succession to different vegetation types and densities. These changes in turn affect the taxonomic composition and biotic interactions of the macroinvertebrate community. These differences may be manifested even within a single reach; isolated pools that formed a few meters apart in one intermittent stream reach differed substantially in nutrient concentrations and dissolved oxygen levels as drying progressed, and supported different macroinvertebrate communities (Stanley *et al*., 1997). A decrease in dissolved oxygen during stream drying may favor the survival of invertebrates that breathe atmospheric oxygen, such as Coleoptera (beetles) and Hemiptera (true bugs), and their predatory activities can further alter the density, abundance, and composition of the remaining community (Stanley *et al*., 1994). Biotic factors such as inter- and intraspecific competition as well as the presence of predators such as fish, amphibians, and birds can also affect the abundance, density, and taxonomic composition of the macroinvertebrate community.

## *Taxa replacement*

Because intermittent streams generally have a higher degree of seasonal habitat variation than perennial streams, greater differences in invertebrate abundance and diversity should occur in intermittent streams across the wet and dry seasons as seasonal replacement of organisms occurs (Boulton & Lake, 1992; Beche *et al*., 2006; Boulton, 2003; Bogan & Lytle, 2007). Seasonal changes in community composition are larger and more significant in intermittent streams, with rheophilic (requiring water for their entire life cycle) wet-season species gradually being replaced during the dry season by winged air-breathing species of true bugs and beetles (Williams, 1987). Bogan & Lytle (2007) identified Capniidae (small winter stoneflies), Simuliidae (blackflies) and Corydalidae (dobsonflies) as significant indicators of high-flow riffles in montane desert streams that did not have permanent headwaters, while low-flow pools of streams with and without permanent headwaters contained more beetles and true bugs. Intermittent streams in Australia had higher proportions of Simuliidae (blackflies), Chironomidae (non-biting midges), Plecoptera (stoneflies), Ephemeroptera (mayflies), and Elmidae (riffle beetles) in the high-flow conditions of the early wet season, but saw a community shift to Dytiscidae (predaceous diving beetles), Corixidae (water boatmen), Notonectidae (backswimmers), Odonata (dragonflies), Chironomidae (non-biting midges), and Leptoceridae (longhorned caddisflies) larvae, and atyid shrimp, in pools that persisted during the dry season (Boulton, 2003). In a Spanish Mediterranean river network, both perennial and intermittent streams exhibited a higher proportion of EPT (Ephemeroptera (mayflies); Plecoptera (stoneflies); and Trichoptera (caddisflies)) during winter high-flow conditions, with a community shift to more dragonflies, beetles, and true bugs (OCH: Odonata, Coleoptera, Hemiptera) during summer low-flow conditions (Bonada *et al*., 2007). The types of organisms present will thus tend to differ depending on the time of year at which stream sampling is done, although taxa differences between intermittent and perennial streams are still expected.

## *Relationship to stream flow duration*

Multiple studies have revealed a large overlap in species identity between perennial and intermittent streams (Williams, 1987; Delucchi & Peckarsky, 1989; Boulton & Lake, 1992; Dieterich, 1992; Feminella, 1996; Shivoga, 2001; Banks, 2005; Beche *et al*., 2006). Although some invertebrate species are more abundant or common in intermittent vs. perennial streams (Smith & Wood, 2002; Flinders & Magoulick, 2003; Wood *et al*., 2005; Meyer & Meyer, 2007), intermittent streams do not have a unique fauna *per se* (Delucchi & Peckarsky, 1989), and few species can be classified as “obligate perennial” or “obligate temporary”. The species composition of intermittent streams during the wet season is often more comparable to that of perennial streams (Shivoga, 2001; Boulton, 2003); for example, the invertebrate community in an intermittent stream riffle during the wet season is more likely to resemble that of a perennial stream riffle, while a small pool remaining as the intermittent stream dries will harbor an invertebrate community more similar to that of an ephemeral habitat. The extent of the average annual dry period in an intermittent stream can also influence invertebrate community composition; in Oregon, both Dieterich (1992) and Banks (2005) found that a continuous flow period of 4-5 months or longer in intermittent headwater streams was correlated with a macroinvertebrate community more similar to that of perennial headwaters in the same region. In addition, intermittent streams with substantial connectivity or proximity to perennial waters are likely to be re-colonized by aerial adults, upstream or downstream movements of aquatic forms, or by upward movement of organisms from deeper substrates (Williams & Hynes, 1976a; Williams, 1977; Gray & Fisher, 1981; Shivoga, 2001).

### *Influence of additional abiotic factors*

Flow duration is a strong driver of macroinvertebrate community composition, but additional abiotic factors also have a significant effect on faunal assemblages. Environmental variables such as stream bed composition, water velocity, predictability of drying/wetting regime, extent of drying (i.e. completely dry vs. small seeps, pools, or moist interstitial spaces), and amount of woody debris, leaf litter, and streambank vegetation strongly affect macroinvertebrate persistence and community composition. The macroinvertebrate community typically inhabiting a first-order, high-elevation, forested headwater stream, for example, would be expected to differ from that in a higher-order, open meadow stream meandering through a valley, even if both streams are intermittent.

Channel substrate/type (i.e. bedrock, boulder cascade, pool, riffle, or rapid) was found to be a strong determinant of invertebrate distribution (Halwas *et al*., 2005), as it affected the degree of stream bed drying and types of invertebrate refugia available. Positive correlations were observed between the degree of sediment moisture and the number of invertebrates in soil cores in a dry desert stream bed (Stanley *et al*., 1994). The presence or absence of a shading canopy has also been seen as an important driver of invertebrate community structure in streams of differing flow duration (Dieterich, 1992; Dieterich *et al*., 1997), because the level of sunlight (insolation) regulates the amount and type of food base present. Studies conducted in Oregon have shown that macroinvertebrate density and diversity can differ in streams flowing through clearcut vs. forested areas (Banks, 2005; Herlihy *et al*., 2005; Banks *et. al*, 2007) as the result of the canopy reduction and increased sedimentation associated with logging.

### *Indicator assemblages*

Despite numerous contradictions in the literature, some commonalities emerge that make it possible to discriminate between invertebrates in streams with differing flow regimes. At the broadest level, taxa diversity (number of taxa and their relative abundance) and/or richness (total number of taxa present) tends to be higher in perennial streams compared to intermittent (Wright *et al*., 1984; Feminella, 1996; Williams, 1996; del Rosario & Resh, 2000; Meyer & Meyer, 2000; Fritz & Dodds, 2005; Wood *et al.*, 2005). Both diversity and abundance are lowest in ephemeral streams (Dieterich, 1992; Price *et al*., 2003; Halwas *et al*., 2005). However, it should be noted that some studies have found similar diversity in perennial and intermittent streams (Bottorff & Knight, 1988; Delucchi, 1988; Miller & Golladay, 1996; Shivoga, 2001; Price *et al*., 2003). Results for the density of organisms in different classes of stream may also differ; Miller & Golladay (1996) found that total invertebrate density was consistently about twice as high in perennial streams, but other studies have found greater invertebrate densities in intermittent streams, as the drying process reduces and fragments wetted habitat, leaving remaining pools crowded with surviving organisms (Boulton & Lake, 1992b; Stanley *et al*., 1994). Conflicting results may be due to the fact that intermittent streams can have greater taxa richness or diversity during periods of high flow, when the habitat is more similar to a perennial stream, but the number of taxa decreases as the stream dries down and organisms die, migrate, or take refuge (Boulton, 2003).

Invertebrate abundance and diversity in temporary streams can also decrease following floods that scour out occupied habitat (Wright *et al*., 1984; Stanley *et al*., 1994; Miller & Golladay, 1996). Fritz & Dodds (2005) found that intermittent prairie streams experiencing harsher conditions, including a long dry period, low flow predictability, high flood frequency, and/or low surface connectivity, had consistently lower taxa richness. This suggests that sites may vary annually in richness depending on differences in precipitation patterns (i.e. drought years vs. flood years). In addition, the drying regime in intermittent streams is affected by the types of habitat available; for example, intermittent streams with exposed bedrock channels retain fewer moist pools and seeps that could serve as refugia during the dry phase than intermittent streams with abundant gravel, cobble, or woody debris (Clifford, 1966; Chadwick & Huryn, 2007).

Although streams with differing flow duration often have many species in common, a small subset of taxa may predominate in either the perennial or intermittent system. Spanish Mediterranean streams with different flow categories showed distinct biological differences, with intermittent streams dominated by taxa with pool-like strategies and ephemeral streams dominated by taxa with life-history adaptations for surviving floods and droughts (Bonada *et al*., 2007). Taxa in permanent streams in this study exhibited few significant biological traits, most likely due to greater habitat stability (i.e. continuing presence of both riffles and pools), and perennial and intermittent streams had similar taxa richness, although both exhibited greater richness than ephemeral sites (Bonada *et al*., 2007). It should be noted that even when intermittent and perennial streams are observed to have similar abundance and diversity of major taxonomic groups, less similarity is seen when species-level identifications are made (Bottorff & Knight, 1988; Wood *et al*., 2005), although this level of taxonomic resolution is not always possible.

### *Community composition*

A positive relationship has been noted between the proportion and/or abundance of EPT taxa and increasing flow permanence (Feminella, 1996; Smith *et al*. 2003; Wood *et al*., 2005). This is not surprising, as these orders are known in general to require cold, well-oxygenated, fast-flowing water (Merritt *et al*., 2007; Wiggins, 1996; Stewart & Stark, 2002). However, some taxa in these orders possess adaptations that render them more tolerant of temporary and/or slow-moving waters. Several species within these three orders have been found in temporary streams, in some instances as dominant components of the invertebrate community (Lehmkuhl, 1971; Tew, 1971;

McElravy *et al*., 1989; Dieterich, 1992; Dieterich & Anderson, 1995; Jacobi & Cary, 1996; Feminella, 1996; Anderson, 1997; Shivoga, 2001; Halwas *et al*., 2005). One confounding factor is the fact that many EPT taxa are intolerant of pollution, so a stream that is negatively impacted by human activities would be expected to have a lower proportion of EPT, even if the stream has year-round flow. Thus, presence of EPT alone is not a conclusive indicator of flow duration, although the presence or dominance of specific families within these orders can be more revealing (see *Life History Adaptations* below).

Predator densities also tend to rise as temporary waters dry down. Several studies have noted higher proportions of certain families of beetles (dytiscids and hydrophilids) and true bugs (notonectids) in intermittent streams (Stanley *et al*., 1994; Boulton, 2003; Bogan & Lytle, 2007), especially as the drying process creates isolated pools (Wood *et al*., 2005). This is likely due to the fact that adults of these orders are strong fliers, capable of colonizing from perennial waters, and most are predators that are attracted to the increased prey density created as streams dry down and surviving organisms are crowded into smaller wetted spaces.

## *Life history adaptations*

It is postulated that life history traits of macroinvertebrate communities such as respiration, reproduction, locomotion, development rate, and dispersal capacity will differ according to stream flow duration (Williams, 1996), with species that persist through the dry season in temporary habitats more likely to have life history adaptations that allow them to resist or avoid desiccation. Williams & Hynes (1977) postulated three main classes of organisms inhabiting temporary streams in southern Canada, namely: permanent stream species with wide enough tolerance ranges to survive in temporary streams; facultative species able to exploit both permanent and temporary waters; and species specialized to, and potentially restricted to, temporary habitats. Wiggins *et al.* (1980) considered four major life history strategies that allow species to successfully exploit temporary waters: 1) year-round residents that are desiccation-resistant and non-dispersing; 2) spring recruits that oviposit in water but aestivate and overwinter in the dry basin; 3) summer recruits that oviposit in the dry basin and overwinter as desiccation-resistant eggs or larvae; and 4) non-wintering migrants that leave the habitat before it dries, and return when flow resumes. Such species would be better able to survive sudden or unpredictable changes such as atypical flooding or drought events compared to obligate perennial species, and would also be likely to occur as part of the annual faunal community of streams that experience regular and predictable cycles of dry-down and re-wetting (Clifford, 1966; Williams, 1996).

### Desiccation resistance

Numerous examples of invertebrates with life history adaptations that confer resistance to drying have been reported in the literature. Some limnephilid caddisflies lay desiccation-resistant eggs encased in a gelatinous mass in humid protected areas at the edges of the dry stream channel, such as the undersides of logs or rocks. The eggs undergo diapause until they are wetted and stimulated to hatch by the resumption of flow in intermittent streams (Wiggins, 1973). Several species of limnephilid caddisflies have been reported as occurring with greater abundance or richness in intermittent streams in a variety of geographic regions (Tew, 1971; Meyer & Meyer, 2000; Anderson, 1997; Smith & Wood, 2002; Wood *et al*., 2005).

## Dispersal

Adult invertebrates with strong flight capability and high dispersal capacity, especially beetles (Coleoptera) and true bugs (Hemiptera), are often more abundant and diverse in intermittent streams (Williams, 1996; Boulton, 2003; Bogan & Lytle, 2007; Bonada *et al*., 2007). Adult dytiscids (predaceous diving beetles) or hydrophilids (water scavenger beetles) can fly in from nearby permanent waters to feed in intermittent streams as prey density rises in remaining pools, then depart as conditions become unfavorable. Similar behavior is seen in Hemiptera such as notonectids (backswimmers) and corixids (water boatmen), although the latter are not predaceous. Crayfish, which have a longer life span and are entirely aquatic, would be expected to be a more typical perennial group, but they are also highly mobile and can survive drying by moving into pools, burrowing into the substrate, or migrating to new habitat, and may be more abundant in intermittent streams than in perennial (Williams & Hynes, 1976b; Flinders & Magoulick, 2003).

## In-stream refugia

Individuals may also survive during the dry season by burrowing into the hyporheic zone (HZ), a subsurface volume of sediment with porous spaces that acts as a region of active exchange of water, nutrients, organic materials, and dissolved oxygen (Boulton *et al*., 1998). It is generally considered to constitute the region below and adjacent to the stream bed, and is an area in which water from the channel and water from stream bed sediments mix and are exchanged. Spaces between sediment particles in the HZ provide habitat for a hyporheic community of small crustaceans, worms, and water mites, and for the immature stages of many aquatic insects (Clifford, 1966; Clinton *et al.*, 1996; Boulton *et al*., 1998; Smith, 2005). The HZ may also serve as a refuge for benthic invertebrates, which are considered temporary hyporheic zone residents, during periods of low flow or even in apparently dry stream beds (Williams & Hynes, 1974; Williams & Hynes, 1976b; Williams 1987; Beffy, 1997). Data are conflicting, however, as other studies indicate that benthic invertebrates do not tend to use the hyporheic zone as a refuge during stream drying (Delucchi, 1989; Stanley *et. al*., 1994; Boulton & Stanley, 1995; Clinton *et al.*, 1996; del Rosario & Resh, 2000).

## Obligate perennial taxa

Because so many invertebrate taxa are common to both intermittent and perennial streams, for the purposes of stream duration assessment it may be more revealing to focus on a few perennial obligate taxa whose life history characteristics render them unable to persist in temporary waters. Chadwick & Huryn (2007) noted that channel drying typically excludes large-bodied aquatic taxa whose long generation times and high biomass requires perennial flow to complete their life cycle, specifically freshwater mussels (Margaritiferidae, Unionidae), some odonates (Aeshnidae, Corduligasteridae, Gomphidae), and some families of stonefly (Pteronarcyidae, Perlidae). In the Pacific Northwest, freshwater mussels, some aquatic snails, and most species of mayflies are considered perennial obligate taxa. *Juga*, an aquatic pleurocerid snail with a long life cycle (3-7 years) that feeds on algae, leaves, and detritus, can be considered an obligate perennial stream species (Hawkins & Furnish, 1987; Dieterich, 1992; Bob Wisseman, Aquatic Biology Associates, pers. comm.), although it should be noted that this snail may be absent from cold, high-elevation streams (Hawkins & Furnish, 1987). Freshwater mussels, which are entirely aquatic and can live for up to 100 years, are also confined to permanent water bodies (Burch, 1973), although they may be present at lower abundance or absent from high-gradient streams where fast flow and rocky substrate inhibits the establishment of juveniles. Some species of mussels are also sensitive to pollution and may therefore be absent from impaired waters, or from streams that lack the fish hosts required by larval mussels to develop to the juvenile stage.

### *Special considerations for Oregon*

Multiple studies conducted in Oregon streams indicate that whereas sharply delineated perennial-obligate or temporary-obligate communities do not exist, certain families of macroinvertebrates are present in higher abundances in perennial vs. intermittent or ephemeral streams. This may be due in part to the greater predictability of Oregon's distinct wet and dry seasons, which could drive development of a more characteristic faunal community in temporary streams (Dieterich, 1992). Many studies have been conducted in the eastern foothills of the Coast Range around Corvallis Oregon, in intermittent forested or meadow streams that maintain flow from about November through May. During the dry season, the meadow channels tended to dry down rapidly and completely, while the forest streams usually dried out more gradually and retained regions with small pools or seeps, even when dry along much of their length (Dieterich, 1992; Dieterich & Anderson, 1998).

Many findings regarding the macroinvertebrate communities of summer-dry streams in Oregon are similar to those reported above in other geographic areas. A great deal of overlap is seen in the fauna of perennial and temporary streams, but some groups are found more commonly in intermittent or ephemeral sites, and a smaller subset prefer or are restricted to perennial habitats (Dieterich, 1992; Banks, 2005). Some perennial headwater streams in western Oregon had a greater abundance of organisms than intermittent, although the species richness (total number of taxa) was similar or even greater in intermittent. Ephemeral streams had only about 25-33% as many species as the other two stream types (Dieterich, 1992). In addition to flow duration, in Oregon timber harvest can be a strong driver of macroinvertebrate community composition, as logging can alter the degree of sunlight reaching the stream and contribute to increased sedimentation. Differences in adult insect emergence rates, taxa richness, and density have been observed in perennial and intermittent streams flowing through logged vs. forested sites in western Oregon (Banks, 2005; Banks *et al*., 2007).

Caddisflies often exhibit very high species diversity in Oregon's summer-dry streams, even though they may comprise a small proportion of the overall invertebrate abundance (Dieterich, 1992; Anderson & Dieterich, 1992; Anderson 1997); this has also been found in other studies in different regions (Mackay & Wiggins, 1979; Smith & Wood, 2002). The Limnephilidae (Northern caddisflies) are the largest caddisfly family in North America; its members exploit a wide variety of habitats, from permanent streams to temporary pools to moist terrestrial habitats (Wiggins, 1993). Several genera in this family are known to have adaptations that allow them to survive periods of summer dryness, including: synchronizing their growth period to the wet season; inhabiting moist seeps that persist in intermittent streams; surviving in moist terrestrial habitats as adults that undergo ovarial diapause, living for 5-7 months and ovipositing when stream flows resume in fall; or laying desiccation-resistant eggs that persist through the dry season and hatch when wetted (Clifford, 1966; Tew, 1971; Wiggins, 1973; Mackay & Wiggins, 1979; Anderson & Dieterich, 1992). Because of these adaptations, limnephilid caddisflies are often an abundant and diverse component of intermittent streams. In studies on Trichoptera conducted by Anderson (1997) on intermittent oak savannah streams near Corvallis OR, the vast majority of all caddisflies collected were limnephilid species (*Pseudostenophylax, Psychoglypha*, and *Limnephilus*). Five species of caddisflies in three families (Limnephilidae, Rhyacophilidae, and Lepidistomatidae) accounted for over three-quarters of the total number of species collected from intermittent streams (Anderson & Dieterich, 1992), with overall species assemblages differing in a characteristic way between forested and meadow streams. Six species in three genera of Limnephilidae, all of which appeared to have life history adaptations for temporary streams, were collected from a small second order intermittent stream near Corvallis (Tew, 1971).

Plecoptera (stoneflies) and Ephemeroptera (mayflies) are more commonly found in fast-flowing perennial waters. An analysis of macroinvertebrate data from 167 small, forested, perennial headwater streams in three different ecoregions in western Oregon showed that 9 of the 16 most common taxa reported were Ephemeroptera, Trichoptera, or Plecoptera (Herlihy *et al*., 2005), with an average 55% of all individuals collected at each site being EPT. A comparative study of small headwater streams in a portion of the Central Oregon Coast Range found a heptageniid (flat headed) mayfly (*Ironodes*) and a hydropsychid (net-spinning) caddisfly (*Parapsyche*) only in the perennial streams (Banks, 2005). However, some EPT taxa in Oregon occur in temporary habitats and possess life history characteristics that allow them to survive periods of drought. Lemkuhl (1971) collected stonefly nymphs in the genus *Nemoura* (Nemouridae; forestflies) and *Capnia* (Capniidae; small winter stoneflies) from a roadside ditch in Corvallis that dried down regularly for 6 months of the year, and hypothesized that eggs or young nymphs present in the wet spring undergo diapause when the habitat is dry, completing development when flow resumes in the fall. Two species of stonefly from the Nemouridae and Chloroperlidae (green stonefly) families (*Ostrocerca* and *Sweltsa*, respectively) and a philopotamid caddisfly (*Wormaldia*) were strongly associated with intermittent flow in small headwater streams in the Central Oregon Coast Range (Banks *et al*., 2007), although a different philopotamid species (*Dolophilodes*) was found more commonly in perennial streams in the same study. Two mayfly species (Leptophlebiidae and Siphlonuridae family) and four stonefly species (Nemouridae, Chloroperlidae, and Perlodidae families) that accounted for >95% of mayfly and stonefly emergence from two temporary forested streams in western Oregon were found to exhibit life history strategies that increased the likelihood of survival in temporary habitats, including asynchronous development, faster development at longer day lengths, and egg diapause during summer-dry periods (Dieterich & Anderson, 1995). Tew (1971) reported two species of Baetidae (small minnow mayflies) as extremely abundant in an intermittent Oregon stream, although these are normally a component of perennial streams, as well as five species of nemourid and capniid stoneflies, although the precise mechanism of their survival during the dry season was not elucidated.

The variety of ecoregions in Oregon can complicate the designation of flow duration indicator taxa and community composition. The invertebrate communities in streams in the Willamette Valley and Cascade Mountain ecoregions in western Oregon show substantial differences, with total taxa richness, diversity, and %EPT composition much higher overall in Cascade streams (Li *et al*., 2001). Additionally, taxonomic composition of stream communities differs depending on stream elevation, gradient, order, and ecoregion. For example, freshwater mussels, *Juga* snails, and gomphid dragonfly nymphs rely on permanently flowing water, but are unlikely to be found in high-gradient forested headwater streams in western Oregon, as substrate, amount of sunlight, and other physico-chemical variables are less suited to the presence of these organisms. Thus, even though several taxa in Oregon streams can be strong indicators of perennial flow, members of these families will not always be present in every perennial stream, depending on the ecoregion, elevation, substrate, canopy, types of surrounding landscape uses, etc.

## *Existing models*

Intermittent and especially ephemeral streams are often overlooked and undervalued as habitat for aquatic invertebrates and other organisms, and few biological assessment techniques take inhabitants of temporary flowing waters into account. Rapid field-based tools that distinguish reliably between ephemeral, intermittent, and perennial streams are critically needed but challenging to develop. For example, different rapid habitat assessment protocols used in forested headwater streams in several eastern and Midwestern states consistently identified ephemeral channels, but were significantly less accurate at distinguishing between intermittent and perennial streams (Fritz *et al*., 2008).

### Ohio

Detailed invertebrate sampling and sample preservation techniques for use in headwater stream permanence assessment have been developed by the EPA (Fitz *et al*., 2006). Aquatic macroinvertebrate metrics have been created as part of a rapid assessment protocol for primary headwater streams in Ohio, which are generally small, have shallow pools (generally <40 cm), drain a watershed less than 1 $mi^2$, and contain reaches that may be perennial, intermittent, or ephemeral (Ohio EPA, 2002). The field evaluation index used for macroinvertebrates in this procedure identifies organisms to order or family level, with the exception of EPT taxa, which must be identified to genus. This method is based on water temperature as a major driver of macroinvertebrate community composition, with three classes delineated: Class III = streams with cool-cold perennial flowing water; Class II = streams with warm-water adapted fauna present either seasonally or annually; and Class I = streams with normally dry channels and little to no aquatic life. Macroinvertebrate sampling may be done at any time of year using this protocol, but is considered most representative during the summer (July through September). All available habitats are sampled for a minimum of 30 minutes or until no new taxa are found.

For the assessment process, each identified taxon collected in the field receives a score based on its expected presence in cool/cold perennial waters (score = 3), intermittent or warm water perennial streams (score= 2), or ephemeral streams (channel usually dry, score = 1). The final summed score is highest for cool-cold perennial waters and lowest for ephemeral. The Ohio assessment procedure weights EPT taxa, fishflies (Corydalidae), and water penny beetle larvae (Psephenidae) as cold water-adapted types; crayfish, dragonfly nymphs, and riffle beetles as warm-water perennial or intermittent types; and worms, sowbugs, scuds, damselfly nymphs, larvae of Chironomidae (non-biting midges), all other Diptera (true flies), all other beetles, and snails and clams as indicators of ephemeral waters. Aquatic bugs (Hemiptera) are not included in the rating, as they are considered highly mobile and able to colonize many types of habitat rapidly (Ken Fritz, pers. comm.). The total number of EPT taxa is also used as a separate indicator for the final designation of a stream as perennial, and requires genus- or species-level identification.

### North Carolina

Aquatic macroinvertebrates are included in methodology developed to determine flow duration in North Carolina streams (NC Division of Water Quality, 2005). A recommended list of perennial taxa has been generated, the majority of which are mayflies, caddisflies, and stoneflies, in addition to a few families of true flies, beetles, and mollusks. Several of the recommended perennial indicator families, such as water penny beetle larvae, riffle beetles, and freshwater mollusks, have also been shown to be more common in or restricted to perennial waters in studies detailed above, and the larger number of EPT families considered as perennial indicators agrees with the overall higher proportion of EPT found in perennial waters in other studies. However, several of the individual EPT families specified as perennial indicators contain genera and/or species that can be considered facultative in Oregon and are not perennial-obligate; some, such as the limnephilid caddisflies, are known to have life history adaptations that allow exploitation of temporary habitats (Clifford, 1966; Tew, 1971; Wiggins, 1973; Mackay & Wiggins, 1979; Anderson & Dieterich, 1992).

For the assessment process, diversity and abundance of benthic macroinvertebrates found at different ranges of search effort is recorded, with rankings of "absent", (no macroinvertebrates), weak (macroinvertebrates observed after intensive searching, i.e. $\geq$10 min.), moderate (macroinvertebrates observed after moderate searching, i.e. $\geq$1-2 min.), or strong (macroinvertebrates found easily). The procedure recommends sampling a variety of habitats, though little methodology is given, and alternative sampling methods for high flow versus low flow conditions are not addressed. The stated sampling effort times are low, and are thus unlikely to allow representative sampling of different habitats within a stream reach, especially by practitioners who are not trained as entomologists, which could result in a skewed rating. The Fairfax County Perennial Stream Mapping Project (VA) uses a stream identification method based on the North Carolina protocol (www.fairfaxcounty.gov/dpwes/watersheds/ps_protocols.pdf) , but because most macroinvertebrates are identified only to order in this assessment, they are considered to be only a secondary indicator of stream type.

## Macroinvertebrates in Oregon stream duration assessment

### *Taxonomic considerations*

In the majority of stream duration studies, taxa indicated as predominant in a given stream type are identified to genus or species level. Identification to the family level, although it can be done fairly rapidly in the field, introduces an unavoidable aspect of uncertainty. For example, indicator species analyses performed on macroinvertebrates in 20 streams in the Central Oregon Coast Range found that *Dolophilodes*, a genus of caddisfly in the family Philopotamidae, was found most commonly in perennial streams, but *Wormaldia* caddisflies, a genus that is also in the family Philopotamidae, were associated with intermittent sites (Banks, 2005). The need to identify macroinvertebrates to family for the purposes of the Oregon assessment tool will not allow the variety of genus- and species-level differences in life history traits to be taken into account. In addition, estimates of taxa richness will be lower, since multiple species may be present within a single family or order within a stream. For example, a study of intermittent streams in western Oregon (Dieterich, 1992) found the highest species richness comprised by members of just a single family, the Tipulidae (crane flies); the same study found that caddisflies accounted for the 2nd highest species richness, with 26 species in six families, although overall caddisfly abundance in samples was low. Order or family-level identification of invertebrates will thus underestimate the actual number of taxa present. This problem could be compounded by errors in identification by practitioners who lack sufficient entomological experience; for example, a pool may contain larvae of Culicidae (mosquitoes), Dixidae (dixid midges) and Chironomidae (non-biting midges), but members of these families may be difficult for an inexperienced practitioner to distinguish, which could result in the total number of families being scored incorrectly.

Taxa diversity is considered to increase from ephemeral to perennial streams, but the season during which macroinvertebrate sampling is done will also influence the number of taxa present. For example, both perennial and intermittent streams will have lower overall numbers of taxa at low-flow summer periods. In addition, even if multiple isolated pools are found within a reach, families that require actively flowing water, such as philopotamid or hydropsychid caddisflies, are likely to be excluded.

Our recommendations for field methodology, indicator taxa, and scoring take into account the need for rapid field-based assessment that can be done at any time of year, and the accompanying taxonomic constraints.

## *Recommended Indicators*

The recommended numbers of taxa for each flow duration category in Table 1 are based on averages from multiple studies conducted in the United States (Clifford, 1966; Tew, 1971; Savage & Rabe, 1979; Abell, 1984; Feminella, 1996; Miller & Golladay, 1996; del Rosario & Resh, 2000; Chadwick & Huryn, 2007), Europe (Meyer & Meyer, 2000), Africa (Shivoga, 2001), and Canada (Williams & Hynes, 1976b; Williams, 1987), as well as datasets from an assessment of 166 headwater streams in western Oregon, and from 159 perennial streams sampled as part of a national wadeable streams assessment (Alan Herlihy, Oregon State University, unpublished data). Indicator taxa recommendations are based more heavily on results from studies conducted in Oregon and elsewhere in the Northwest, in addition to consultation with multiple regional and national experts, including:

- Larry Eaton, North Carolina Division of Water Quality, Raleigh NC
- Ken Fritz, U. S. Environmental Protection Agency, Cincinnati OH
- Bill Gerth, Faculty Research Assistant, Oregon State University
- Jim Johnson, Odonatologist, Vancouver WA
- Judith Li, Oregon State University, Corvallis OR
- Dennis Paulson, Director Emeritus, Slater Museum of Natural History, Seattle WA
- Perianne Russell, North Carolina Division of Water Quality, Raleigh NC
- Robert Wisseman, Aquatic Biology Associates Inc., Corvallis OR

Photographs of representative specimens of indicator taxa below are provided for reference in Appendix A. It is anticipated that all identification will be done in the field and to the taxonomic level of family, except in such cases where the extreme immaturity or small size of a specimen limits resolution to order. Non-insect taxa will be routinely identified to subclass or order level, such as aquatic earthworms (subclass Oligochaeta), leeches (subclass Hirudinea), scuds (order

Amphipoda), water mites (subclass Hydracarina), and aquatic sowbugs (order Isopoda). An inexpensive, portable, waterproof field guide such as The Xerces Society *Macroinvertebrates of the Pacific Northwest* (2003) and a hand lens is needed to identify organisms to family level. Ideally, we recommend that a macroinvertebrate field guide be developed specifically to accompany this stream duration assessment protocol. Such a guide could be targeted at the specific needs of practitioners, such as identifying the pupal forms of common aquatic macroinvertebrates to family, distinguishing between larvae of different families that are similar in appearance (i.e. mosquito vs. midge), and recognizing common semi-aquatic or terrestrial invertebrates that are most likely to invade a dry channel.

Users should be aware that not all taxa listed as flow indicators are present in every correlated stream class. For example, a perennial stream may lack *Juga* snails, depending on the stream location, elevation, and flow rate. By the same token, many families of caddisflies that do not construct portable cases may complete development successfully in an intermittent stream but leave no trace of their presence once development is completed, unlike the substantial cases that can be left behind by limnephilid caddisflies. However, the presence of a number of the different indicators for each specific stream class below, in combination with the absence or very low level of indicators for alternative stream classes, should provide a more robust estimation of stream duration.

**Table 1. Stream flow duration indicators**

| Perennial | Intermittent | Ephemeral |
|---|---|---|
| *Juga spp.* (pleurocerid snail)<br>Freshwater mussels (Margaritiferidae, Unionidae), but less likely in small high-gradient streams | Larvae/pupae of:<br>Limnephilidae (Northern caddisfly) | Larvae/pupae of:<br>Culicidae (mosquito) |
| Larvae or pupae of:<br>Philopotamidae (finger-net caddisfly)<br>Hydropsychidae (net-spinning caddisfly)<br>Rhyacophilidae (freeliving caddisfly)<br>Glossosomatidae (saddle case-maker caddisfly), esp. in forested headwater streams | Nymphs of:<br>Capniidae (small winter stonefly)<br>Nemouridae (forestfly) | |
| Nymphs of:<br>Pteronarcyidae (giant stonefly)<br>Perlidae (golden stonefly) | Larvae/adults of:<br>Dytiscidae (predaceous diving beetle)<br>Hydrophilidae (water scavenger beetle) | |
| Larvae of:<br>Elmidae (riffle beetle),<br>Psephenidae (water penny), esp. in eastern regions | Nymphs/adults of:<br>Notonectidae (backswimmers) | |
| Larvae/nymphs of:<br>Gomphidae (clubtail dragonfly)<br>Cordulegastridae (biddies)<br>Calopterygidae (broadwinged damselfly)<br>esp. in larger, higher-order streams in eastern OR | Larvae/nymphs of:<br>Lestidae (spread-winged damselfly) | |
| Nymphs of ≥4 different families of Ephemeroptera (mayfly) | | |
| Greatest taxa diversity | Intermediate taxa diversity | Low taxa diversity |
| Highest EPT | Intermediate EPT | Low/no EPT (**exception** is limnephilid cases in a dry channel, which would indicate intermittent flow) |

### *Scoring*

The current Oregon Draft Stream Assessment method uses a character rating system of absent (0), weak (1), moderate (2), or strong (3), with higher scores correlating with increased flow permanence. In the biological assessment portion of the method, fish and macroinvertebrates are given the same weight, with possible scores ranging from 0-3, whereas the score from amphibian monitoring is weighted less heavily (0-1.5). Because of the uncertainties inherent in using macroinvertebrate families as indicators, we recommend that macroinvertebrates receive the same final scoring weight as amphibians in the final assessment, i.e. 0-1.5.

The Oregon Draft method also incorporates levels of search effort into scoring. We recommend instead that a standardized number of 6 samples encompassing all habitats within a reach be taken at each site. Alternatively, a standardized macroinvertebrate search time of 30 minutes can be used, during which period all possible different habitat types within the reach are sampled (see *Field Methodology* below), although this will not allow as thorough a sampling of the reach to be done.

Recommendations for some metrics have been made with the intended end users of the protocol in mind, specifically for the expected numbers of different aquatic invertebrate taxa found within the reach. For example, we recommend that if practitioners find 15 or more different taxa (i.e. families, subclasses, or orders, where applicable) within a reach, it is considered a strong indicator of perennial flow. An individual with extensive experience in aquatic entomology and stream sampling conducting a scientific study is would be more likely to find at least 20 different taxa, depending on the time of year. However, practitioners using this assessment method will mostly lack this expertise, and a lack of experience in aquatic entomology may also cause users to overlook small or cryptic individuals. In addition, regardless of whether six samples per reach are taken or if sampling is conducted for 30 minutes in a reach, it is very unlikely that all possible taxa will be found. The constraints on time and expertise inherent in this technique result in more of a sub-sampling of each reach, and have therefore led us to recommend slightly lower numbers of taxa as stream duration class indicators. Modifications to this metric may be made based on the results of field testing.

For macroinvertebrate scoring, the number and identity of all taxa collected is recorded, so that the number of different taxa and % EPT can be calculated. Specific organisms whose identity is in question can be preserved and identified later with additional guides and expertise if needed (as described in *Field methodology*, pg. 20). The following rating scale should be used:

**Strong (1.5):**
≥15 different aquatic macroinvertebrate taxa are found within the reach
*OR*
1 or more members of the families of perennial indicator taxa (Table 1) are found
*OR*
≥4 different families of Ephemeroptera (mayfly) are found
*OR*
>5 different families of EPT are found
*OR*
>40% of total taxa are EPT

**Moderate (1.0):**
The above criteria are not met, *AND*
5-14 different aquatic macroinvertebrate taxa are found within the reach
*OR*
1 or more members of the families of intermittent indicator taxa (Table 1) are found
*OR*
2-5 different families EPT are found
*OR*
5-40% of total taxa are EPT
*OR*
1 or more limnephilid caddisfly cases are found in a dry stream bed

**Weak (0.5):**
None of the above criteria are met, *AND*
1-4 different aquatic macroinvertebrate taxa are found within the reach
*OR*
0-1 family of EPT is found
OR
<5% of total taxa are EPT (note exception if limnephilid cases are found in a dry channel)

**Absent (0):**
No macroinvertebrates are found within the reach after sampling

## *Field methodology*

The current Oregon Draft Protocol assigns indicator strengths based in large part on the amount of time spent searching for macroinvertebrates, with search effort ranging from "minimal, 5 minutes" to "significant, 15 or more minutes". We recommend instead that a set number of six samples total be taken at each site across all habitat types identified within the reach as a more thorough and representative means of sampling. For example, in a reach that has one riffle, 2 pools, and a region of large woody debris, two samples each could be taken from the riffle and the woody debris while each pool is sampled once, for a total of six samples representing all available habitats. Studies of headwater streams in the Midwest suggest that the number of different taxa collected from a 30-m stream reach peaks and levels off at eight samples (Fritz *et al*., 2006). Similar results were seen in streams in western Oregon, with the number of taxa collected increasing sharply for the first four to eight samples taken, and then increasing more gradually (Li *et al*., 2001). This study also suggested that the appropriate number of samples to be taken for full taxa representation may differ depending on reach length and ecoregion, as streams in the Willamette Valley had consistently lower taxa richness than Cascade streams, so the number of samples needed to achieve maximum invertebrate representation with minimum sampling effort for the purposes of the Oregon Draft Stream Assessment may need to be standardized experimentally.

Conversely, macroinvertebrate sampling could also be standardized if conducted within a set span that is long enough to reasonably allow a thorough search effort. If this approach is desired, we recommend that 30 minutes minimum be used, as opposed to a varying "level of effort". Note that this time refers <u>only</u> to sampling effort, not to the time needed for specimen sorting and identification. The time ranges in the Draft Protocol represent an extremely minimal span to realistically be able to sample more than one type of habitat in a stream reach. Aquatic macroinvertebrates are typically small, cryptic, and hidden, especially in a dry or drying stream bed that could have many small separated pools, seeps, or areas of moist substrate to be investigated. In addition, since different types of aquatic macroinvertebrates exploit different habitats (i.e. pools, riffles, cobble, woody debris, leaf packs, root wads, upper level of substrate, etc.), five to ten minutes is not sufficient to sample more than one or two habitats in a reach, which could thus skew the data. The upper limit for significant search effort is not specified in the current protocol, which again reduces between-site consistency, as under this designation "significant" effort at one site may be a 15 minute search, while a "significant" effort at a different site may consist of a 30 minute search. Thirty minutes is a standardized time that will allow a more consistent search effort at each site and better enable a more thorough search of multiple regions of potential habitat any type of stream reach, without being too long for the desired rapid assessment process.

## Equipment

Recommended equipment for field sampling includes:

- GPS unit
- 5-gallon bucket
- D-frame net or Surber sampler
- Small hand net or aquarium net
- Wash bottles, 250 mL
- Round metal sieve with 500 µm mesh
- Entomological forceps or fine tweezers
- Small hand trowel or 3-pronged rake
- Small brush
- Knee boots or hip waders
- Shallow white plastic tray or Tupperware container
- Field data sheets on Rite-in-the-Rain waterproof paper
- Hand lens
- Macroinvertebrate identification field guide

## Wet channel

Field sampling techniques have been modified from Fritz (2006). Additional sampling protocols are available in the Water Quality Monitoring Technical Guidebook published by the Oregon Plan for Salmon and Watersheds (www.oregon.gov/OWEB/docs/pubs/wq_mon_guide.pdf). Wetted habitats with sufficient flow can be sampled using a standard D-frame net with 500 µm mesh or a Surber sampler (for smaller, shallower streams). When possible, sample all available habitat types in the reach to obtain a representative invertebrate sample, including riffles, runs, pools, large woody debris, leaf packs, and root wads. Begin sampling at the most downstream point in the reach and move upstream to each new sampling site. Avoid additional disturbance and trampling within the transect when moving from one sampling site to the next by walking as far outside of the sampling area as possible.

To sample riffle or run/glide habitats, hold the net perpendicular to the bottom downstream of the area being sampled, so that the current carries material into the net. Remove any rocks that block close contact between the net and the substrate, so that all organisms dislodged from the benthos will be carried into the net and not be lost underneath the net's edge. Use your fingers or a soft brush to gently rub the rocks while holding them in front of the net to remove and collect any attached organisms. Then, stir and disturb the sediment in a 0.16 $m^2$ (1.7 $ft^2$) sampling area in front (upstream) of the net mouth for 1-2 minutes using hands, boots, or a small three-pronged garden trowel. Small cobble and rocks in the sampling area should be picked up and rubbed or brushed gently while held in the current to dislodge any attached organisms. To sample pools, stir the sediment in the same way and sweep the net upwards from the top of the substrate through the water column to collect dislodged material. Root wads can be sampled by holding the net underneath and kicking or shaking the root wad to dislodge organisms while sweeping the material into the net. Large pieces of woody debris can be sampled in the same manner, or examined visually and attached organisms rinsed off or picked off using entomological forceps or tweezers.

A stream reach may have areas that are wetted but lack sufficient depth to allow the use of a kicknet or Surber sampler. In these cases, a modified bucket sampling technique should be used (see also Fritz *et al.*, 2006). Remove larger rocks and cobble in the designated 0.16 $m^2$ sampling area and gently rub and rinse any attached material into a 5 gallon plastic bucket. Use a small hand trowel to stir the remaining sediment in the sampling area for 1-2 minutes, and collect the suspended material by simultaneously sweeping a small hand net or aquarium net through the water column.

Once a sample has been collected, wash the material in the into a 5 gallon plastic bucket. Large pieces of debris caught in the net (stones, pieces of wood, large intact leaves) can be gently rubbed using fingers or a small soft brush and rinsed into the bucket to dislodge attached organisms, and then discarded. If the net contains an excessive amount of sand or silt, which may occur when sampling in areas with soft substrates, this can be reduced if needed by gently swirling and rinsing down the outside of the net bag in the stream. Once all the sample material has been rinsed into the bucket, it is poured through a 500 μm sieve to collect and concentrate the organisms; additional water may be poured over the sieve to rinse the sample further. Examine the bucket to ensure that no organisms are left behind; snails and leeches often attach to the sides of the bucket, and heavier organisms such as fingernail clams, mollusks, and large caddisflies may remain in sediment left at the bottom of the bucket. Invertebrates can be picked directly from the sieve using entomological forceps or tweezers, but it is preferable to rinse the sieved material into shallow white plastic trays (using a wash bottle) for picking. This will facilitate picking, as the small size and cryptic coloration of many aquatic invertebrates makes them difficult to see when they are not in motion, and organisms are more visible when swimming against a light background.

If any further sample identification is to be performed later or if voucher specimens are desired, invertebrates can be preserved in plastic Nalgene jars containing 95% ethanol. Sample volume and any associated organic material (i.e. algae, leaf debris) should comprise no more than one-half to two-thirds of the sample jar volume to ensure adequate sample preservation (i.e. greater amount of organic material = smaller sample volume per jar); use multiple jars if needed, and label each with stream name, reach location, habitat type in pencil on waterproof paper.

To ensure that each subsequent sample set taken in a different reach is not “contaminated” by invertebrates from the previous site, rinse all equipment thoroughly between each sampling event.  Examine the net closely after rinsing and use forceps or fingers to remove any organisms still clinging to it.

## Dry channel

Our recommendation when assessing a “dry” stream channel is that the reach should first be scouted to ascertain whether it is completely dry, or if areas of standing water such as small pools or seeps that can be sampled for aquatic invertebrates still remain.  Areas with standing water can be sampled as described above; the sampling effort should also include searching in areas of likely refuge such as underneath rocks, in leaf packs, or in areas of moist substrate. Scoring would be done as described above.

If the channel completely lacks standing water, a twenty-minute search effort should be made in which areas of likely refuge such as underneath rocks, in leaf packs, in areas of moist substrate, etc. are investigated.  If NO aquatic organisms, cases, shells, or exuviae are found, then macroinvertebrates should be omitted completely from the assessment process.  Any live invertebrates that are unequivocally aquatic forms may be scored; omit any terrestrial organisms, as these will rapidly invade a dry channel.  If no aquatic invertebrates are found, but a search reveals caddisfly fly casings, aquatic invertebrate exuviae (i.e. dragonfly exuviae), or mussel shells, this should be noted and scored as an indicator of intermittent flow.  This can be a revealing process if sufficient time is taken to adequately search, as the presence of caddisfly cases in a dry channel or a fishfly larva hiding beneath a rock, for example, would be a stronger indicator of more recent flow and thus an intermittent stream.

The decision to retain macroinvertebrates in the assessment metric when the stream channel is dry will depend in part on the amount of time and effort that practitioners are willing to spend searching and sampling.  Searching for invertebrates with adaptations to resist or avoid desiccation in dry or nearly dry streams will require alternative sampling methods.  Individuals may burrow into the hyporheic zone, undergo egg or larval diapause in moist sheltered habitats in the stream bed or edge, or diapause as adults in nearby terrestrial habitat.  Invertebrate sampling during dry periods therefore requires a greater degree of effort, as stream reaches must be scanned for any remaining small seeps or pools as well as areas that might harbor aestivating or diapausing life stages, such as the underside of rocks and branches or the moist substrate of the hyporheic zone.

Identification of invertebrate taxa will also become more problematic, as many invertebrates in dry stream beds are terrestrial, and a practitioner without the necessary entomological expertise identifying organisms in the field may not be able to differentiate between some aquatic and terrestrial invertebrates.  Less error would be incurred by treating the invertebrate metric similar to the fish metric and omitting it entirely from the assessment process if the stream channel is completely dry.  It is anticipated that the redundancy in the other metrics would provide an assessment that would be sufficiently robust even in the absence of macroinvertebrate data.

However, the degree of dryness in both intermittent and ephemeral streams will vary depending on time of year, precipitation, stream size, gradient, etc., and this in turn has a significant impact on the types of macroinvertebrates able to persist in the channel. An ephemeral stream may dry completely in the summer in the absence of rainfall and lack even hyporheic refugia, whereas intermittent streams may retain refugia where some invertebrates persist, such as small pools and seeps, moist/humid areas beneath rocks and debris, or in the hyporheic zone. In addition, ephemeral streams do not tend have as high a proportion of taxa with desiccation-resistant adaptations; rather, they tend to be colonized by adventitious species that have a rapid development time and can mature before complete drying occurs.

If desired, sampling the channel in a dry stream bed can be done by excavating the substrate in 0.16 $m^2$ sampling area to a depth of 20 cm (8 in) using a hand trowel. Additional water will need to be carried in to rinse the sample. The excavated soil is placed in a 5 gallon bucket containing water and allowed to hydrate for 5 minutes. The sample material is then sieved, rinsed, and picked as described above. This is not recommended, as it will be time and labor intensive; substrate excavation may not be possible in areas of bedrock or large cobble and boulders; and practitioners who are not entomologists may be unable to accurately distinguish between aquatic organisms that have taken refuge in the substrate vs. terrestrial invertebrates that have invaded the dry channel.

For purposes of scoring in a dry channel, the section entitled “Drawing Conclusions” in the Oregon Draft Assessment Protocol should be modified per our recommendations to read (addition shown in italics):

“**If the stream segment being evaluated does not meet the above criteria, the stream segment is intermittent when any of the following criteria are met:**

2. A numerical value of at least:
   12 points is determined on the stream identification Form for a Dry Channel
   14 points is determined on the stream identification Form for a Wet Channel

OR

3. A fish is found in the segment

OR

4. More than one individual of an amphibian life stage of larva or further developed associated with the sustained presence of water (Table 3) are present

*OR*

*5. One or more caddisfly cases, mussel shells, or aquatic invertebrate exuviae associated with the sustained presence of water are found in the segment*

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# Appendix A. Representative photographs of indicator taxa

The photographs below are intended only as a quick guide to provide context and a visual reference to individuals who may be unfamiliar with the recommended indicator taxa. As stated earlier, we recommend that a guide be developed specifically for practitioners in the field that could be targeted at issues users of the stream assessment protocol are likely to encounter, such as recognizing and identifying pupae, exuviae, and cases, and differentiating between families whose members may closely resemble one another. Unless otherwise indicated, all photos below are credited to The Xerces Society (Jeff Adams).

## *Mollusca (snails, mussels)*

- *Juga spp.* (pluerocerid snail): Perennial indicator; shells are dark reddish-brown to black, may be smooth or ridged; may have lighter colored lines spiraling with the coils; 10-30 mm (0.4-1.2 in.) in length

- Freshwater mussels (Margaritiferidae <*Margaritifera* spp.>, Unionidae <*Anodonta* & *Gonidea* spp.>): Perennial indicator; bivalved shell with two oblong halves; can live for 100 years and reach >15 cm (6 in.) in length; more common in larger rivers with softer substrate; larvae (glochidia) must attach to a fish host for development and dispersal before dropping off and burrowing into the substrate. For a detailed guide, see "*Freshwater Mussels of the Pacific Northwest*", published by the Pacific Northwest Native Freshwater Mussel Group.

*Anodonta nuttalliana* (winged floater)

*Margaritifera falcata* (western pearlshell)

*Gonidea angulata* (western ridged mussel)

## *Trichoptera (caddisflies)*

- Philopotamidae (finger-net caddisfly): perennial indicator; yellowish-white fleshy body; reach up to 16 mm (0.6 in.); 1st thoracic segment is sclerotized (hardened) on top and has prominent black band around posterior rim; long anal prolegs (arrow); build finger-like nets on the underside of rocks for shelter and to filter organic particles from the water.

- Hydropsychidae (net-spinning caddisfly): perennial indicator; slightly curved body;

sclerotized plate on tope of each thoracic segment; clusters of gills on underside of abdominal segments and last 2 thoracic segments (may be absent in very small early instar larvae); anal prolegs have tufts of long setae (hairs); up to 30 mm (1.2 in.).

- Rhyacophilidae (freeliving caddisfly): perennial indicator; also called green rock worms due

to pale green color of many species of larvae; fleshy body with prominent constrictions between each segment; top of 1st thoracic segment sclerotized; head and mouthparts directed forwards; generally lack abdominal gills; anal prolegs have large claw.

- Glossosomatidae (saddle case-maker caddisfly): perennial indicator; larvae build portable

domed saddle- or tortoise-shaped case from rock fragments; 1st thoracic segment is sclerotized; no abdominal gills; as larvae grow, each successive instar constructs a new case and abandons the old one; larvae up to 9 mm (0.35 in.).

- Limnephilidae (Northern caddisfly): intermittent indicator; fleshy body; thoracic segments

have sclerotized plates on top; larvae up to 30 mm (1.2 in.) in length; prominent prosternal horn projects from underside of "neck"; portable cases may reach 76 mm (3 in.) in length; cases vary in size and construction, usually made from plant materials such as grass or woody debris fragments.

## *Plecoptera (stoneflies)*

- Pteronarcyidae (giant stonefly): perennial indicator; largest stonefly in North America, up to 50 mm (2 in.); body somewhat stocky, dark; clusters of finger-like gills on underside of at least the first two abdominal segments; two cerci ("tails") at tip of abdomen

- Perlidae (golden or common stonefly): perennial indicator; lighter brown, slightly flattened body with golden patterning; up to 30 mm (1.2 in.); gill clusters around the base of each leg; two cerci ("tails") at tip of abdomen

- Capniidae (snowfly or slender winter stonefly): intermittent indicator; elongate dark body with cylindrical abdomen, slightly bulbous in the mid-region; up to 20 mm (0.8 in.) in length; no gills in "neck" region; two cerci ("tails") at tip of abdomen

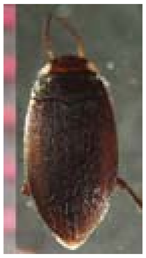
- Nemouridae (forestfly): intermittent indicator; small, stout, hairy bodies; up to 9 mm (0.35 in.) in length; may have gills in "neck" region; both pairs of wing pads diverge at an angle from midline of body; two cerci ("tails") at tip of abdomen

### *Coleoptera (beetles)*

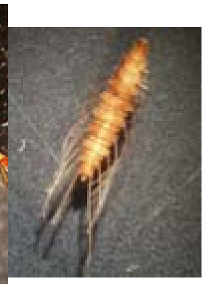
- Elmidae (riffle beetle): perennial indicator; larval body is elongated, cylindrical, hard, dark brown or reddish; up to 16 mm (0.6 in.) long; may be gill tuft at tip of abdomen. Adults have oval to elongate dark brown bodies; up to 6 mm (0.2 in.) long; head is often withdrawn under the pronotum (top of 1$^{st}$ thoracic segment).

- Psephenidae (water penny): perennial indicator; body is flattened, oval to circular, up to 10 mm (0.4 in.) long; head and legs not visible in top view, covered by armored segments,

- Dytiscidae (predaceous diving beetle): intermittent indicator; adults are dark, shiny, convex on top and bottom; may be patterned with lighter patches or fine stripes; up to 40 mm (1.6 in.) in length; long slender antennae; hind and middle legs flattened, covered with dense fringe of swimming hairs. Larvae have elongated bodies, long slender legs; often 2 small cerci ("tails) at the tip of the abdomen; conspicuous broad heads with large curved mandibles



- Hydrophilidae (water scavenger beetle): intermittent indicator; adults resemble Dytiscidae but often have raised keel on underside of body; up to 44 mm (1.6 in.) in length; last 3 segments of antenna swollen to form a club, and segment immediately preceding club may be cup-shaped; Larvae have long cylindrical bodies with short legs; up to 60 mm (2.4 in.) in length; strong curved jaws project at front of head; tip of abdomen is bluntly rounded, lacks cerci.



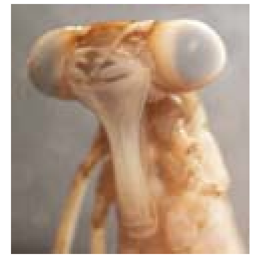

## *Odonata (dragonflies & damselflies)*

- Gomphidae (clubtail dragonfly): perennial indicator; body slightly flattened, up to 42 mm (1.7 in.) in length; tip of abdomen rounded or tapering to blunt point; labium (lower "lip") is flat, not scoop- or spoon-shaped; antennae have 4 segments, with 3rd segment larger, conspicuous, often a different shape, 4th segment small, barely visible

- Cordulegastridae (biddies): perennial indicator; larvae may appear hairy; up to 45 mm (1.8 in.) in length; labium (lower "lip") is scoop shaped and covers much of the front of the head when not extended; short threadlike antennae with 7 segments

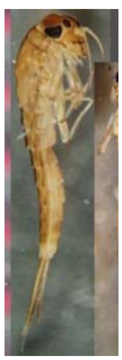
- Calopterygidae (broadwinged damselfly): perennial indicator; body slender, elongated, up to 50 mm (2 in.) in length; long slender legs; 3 gill "plates" at tip of abdomen, with central plate shorter than outer plates; conspicuous antennae, with 1st segment as long as combined length of remaining segments

- Lestidae (spreadwinged damselflies): intermittent indicator; body slender, elongated, up to 29 mm (1.1 in.) in length; 3 gill "plates" at tip of abdomen, all ~same length; labium (lower "lip") is spoonlike, expanded and elongated away from the head; all segments of antennae ~same length



## *Ephemeroptera (mayflies)*

Multiple families: perennial indicator; nymphs have elongated bodies that may be cylindrical or flattened, with three (sometimes 2) long cerci ("tails") at tip of abdomen; up to 20 mm (0.8 in.) in length; plate-like, feathery, or fringed gills at sides of abdomen; conspicuous eyes; slender antennae. From left to right: Leptophlebiidae (pronggill mayfly), Baetidae (small minnow mayfly), Leptohyphidae (little stout crawler mayfly), Heptageniidae (flatheaded mayfly)

## *Hemiptera (true bugs)*

- Notonectidae (backswimmers): elongated body, generally convex on top surface and flattened on bottom; hind legs very long, flattened, oar-like, with fringe of swimming hairs; swim upside down; top surface is lighter colored, often patterned; bottom surface is darker; nymphs resemble adults but have wing pads instead of fully developed wings; short antennae; narrow tubelike beak on underside of head