



Protocol for Monitoring Spring Communities at Ozark National Scenic Riverways, Missouri

Natural Resource Report NPS/HTLN/NRR—2008/029

A Heartland Network Monitoring Protocol



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Natural Resource Monitoring

ON THE COVER

Herbert Hoover birthplace cottage at Herbert Hoover NHS, prescribed fire at Tallgrass Prairie NP, aquatic invertebrate monitoring at George Washington Carver NM, the Mississippi River at Effigy Mounds NM.

Protocol for Monitoring Spring Communities at Ozark National Scenic Riverways, Missouri

David E. Bowles, Hope R. Dodd, Michael H. Williams, Lloyd W. Morrison, Kevin James,
Michael D. DeBacker, Catherine E. Ciak, Janice A. Hinsey, Gareth A. Rowell, and Jennifer L.
Haack

National Park Service
Heartland I&M Network
Wilson's Creek National Battlefield
6424 West Farm Road 182
Republic, Missouri 65738

March 2008

U.S. Department of the Interior
National Park Service
Natural Resource Program Center
Fort Collins, Colorado

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Please cite this publication as:

Bowles, D. E., H. R. Dodd, M. H. Williams, L. M. Morrison, K. James, M. D. DeBacker, C. E. Ciak, J. A. Hinsey, G. A. Rowell, and J. L. Haack. 2008. Protocol for Monitoring Spring Communities at Ozark National Scenic Riverways, Missouri. Natural Resource Report NPS/HTLN/NRR—2008/029. National Park Service, Fort Collins, Colorado.

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I. Background and Objectives

Issues Being Addressed and Rationale for Monitoring Springs

Ozark National Scenic Riverways (OZAR) was established to preserve and interpret the free-flowing Current and Jacks Fork rivers. The park was designated for river corridor protection with narrow National Park Service (NPS) jurisdictional boundaries around the Current and Jacks Fork rivers. The jurisdictional boundary of OZAR encompasses only about 5% of the watershed of the Current and Jacks Fork Rivers with over 50% of the watershed in private ownership. Because a large portion of the watershed lies outside park boundaries, much of the watershed is left unprotected, placing the water quality of the rivers and tributaries at risk (Panfil and Jacobson, 2001). Aquatic ecosystem condition and health are dependent on processes occurring in the entire watershed as well as in riparian and floodplain areas and cannot be manipulated independently of this inter-relationship (Doppelt *et al.*, 1993). The extensive karst topography in the area surrounding OZAR is conducive to the formation of springs, making these aquatic systems vulnerable to contaminated groundwater recharge and interbasin transfer of groundwater from adjacent watersheds. Because springs at OZAR receive their baseflows from groundwater, contamination of groundwater is of special concern due to the rapid recharge and transport of contaminants through the soluble bedrock system of caves, springs, and sinkholes.

Land use, particularly land clearing practices and associated increases in sediment load, nutrient loading, and other point and nonpoint sources, has been reported as the largest long-term threat to streams and springs in the Ozark Highlands (Duchrow, 1977; Mott, 1997; Scott and Udouj, 1999). Land use practices at the watershed level appear to overwhelm localized protection of stream corridors. For example, measures of land use and riparian vegetation at larger spatial scales (watershed level) were superior to local measures at predicting stream conditions within a Midwestern watershed (Roth *et al.*, 1996). Land use activities in the Ozark Highlands include timber management, landfills, grazing, swine and poultry operations, urbanization, gravel mining, stream channelization, removal of riparian vegetation, and lead-zinc mining. Impacts to stream integrity include disruptions in channel geomorphology, increased suspended and deposited sediments, bank erosion, increased light penetration and water temperature, higher periphyton biomass, and decreases in leaf litter and woody debris. Aquatic communities can be impacted from land use practices in the watershed, particularly from the conversion of forestland to pasture (Sweeney, 1995). Increased bank erosion rates and changes in channel morphology through time have also been correlated with increased land clearing of steep uplands within a tributary basin (Stephenson and Mott, 1992) as well as historical riparian land clearing (Jacobson and Primm, 1997).

The NPS has mandated that the managers of OZAR establish baseline data or “vital signs” and long-term monitoring programs for the natural resources found within the park. This information will be used to address any current resource problems, while allowing managers to anticipate and plan for future resource problems. A specific mandate of this legislation is the “preservation of springs.” Within the karst terrain of OZAR there are over 250 springs. Although most of these springs are relatively small, six are considered 1st and 2nd magnitude that is defined as having discharges of at least 2.8 m³/second and 0.28 to 2.8 m³/second, respectively (Meinzer, 1927).

The largest (Big Spring) has a maximum recorded discharge greater than 36 m³/sec and is ranked among the five largest springs in North America.

Assessment of chemical/physical characteristics in lotic systems is a common practice used to monitor aquatic conditions and determine potential areas of degradation or resource problems. This type of water quality assessment gives investigators immediate results, but requires that sampling occur during or soon after a disturbance (such as high inputs of sediment or nutrients). Thus, chemical analysis six months or a year after a major disturbance may not indicate a problem. Because the “most direct and effective measure of the integrity of a water body is the status of its living systems” (Karr and Chu, 1999), it follows that the main focus for a system of vital sign monitoring of spring resources should be the living component—thus providing the rationale for the following biological monitoring protocol. A comprehensive monitoring program should include biotic indicators (vegetation, invertebrates, and fish) that respond or are linked to the physical and chemical conditions within the system. Information obtained from monitoring vegetation, invertebrates and fish, together with chemical and physical data, provides the most integrated and robust assessment of water quality and ecosystem integrity.

In order to assess the natural and anthropogenic processes influencing aquatic vegetation, invertebrate, and fish communities, this protocol has been designed to incorporate the spatial relationship of these biotic indicators with chemical constituents and physical habitat. Local variables, such as conductivity, water temperature, pH, dissolved oxygen, turbidity, current velocity, substrate size, and other habitat variables will be measured. Springs have a mosaic structure, a high degree of individuality, and typically have an azonal character attributed to their physiochemical stability (Cantonati *et al.*, 2006). Because of the previously mentioned threats to the ecological functioning of spring systems, these unique habitats are imperiled. Therefore, the framework for monitoring the large springs at OZAR is directed towards maintaining their ecological integrity and this will be assessed through periodic monitoring of aquatic vegetation, invertebrates, and fish communities.

Mosses, algae, and higher plants are particularly important structural and biological constituents of springs and aquatic systems in general (Hannan and Dorris, 1970; Cushing and Wolf, 1984; Carpenter and Lodge, 1986; Durarte and Canfield, 1990; Stream Bryophyte Group, 1999; Cantonati *et al.*, 2006) and they often have complex relationships with some fish and aquatic invertebrates (Cyr and Downing, 1988; Xie *et al.*, 2005, 2006). Additionally, aquatic plants are important biological filters of a variety of chemical contaminants and nutrients (Demars and Harper, 1998; Cantonati *et al.*, 2006; Vardanyan and Ingok, 2006). Growth of aquatic vegetation is influenced by a number of factors including light and nutrient availability, water chemistry and dissolved gasses, temperature, herbivory, and a broad array of physical characteristics including channel size, canopy cover, water depth, substrate size, and current velocity (Spence, 1967; Haslam, 1978; Dawson and Kern-Hansen, 1978, 1979; Chambers *et al.*, 1991; Onaindia *et al.*, 1996; Barendregt and Bio, 2003; Bernez *et al.*, 2004a). As such, aquatic vegetation communities in spring ecosystems are vulnerable to a broad variety of anthropogenic disturbances (Sanford, 1979; Englund, 1991; Schütz, 1995; Preston *et al.*, 2003). Because of these vulnerabilities, monitoring changes in aquatic vegetation has long been used as an indicator of anthropogenic disturbance throughout Europe, although this approach has received little attention in the United States (Romero and Onaindia, 1995; Tremp and Kohler, 1995; Small *et al.*, 1996; Bartodziej and

Ludlow, 1997; Ali *et al.*, 1999; Schorer *et al.*, 2000; Bernez *et al.*, 2001, 2004b; Haury *et al.*, 2002; Scott *et al.*, 2002; Daniel *et al.*, 2005; Haslam, 2006). Previous studies have found that certain hydrophytes and mosses are more sensitive to anthropogenic disturbance than others (Haslam, 1982; Carbiener *et al.*, 1990; Poole and Bowles, 1999; Bernez *et al.*, 2001, 2004b; Haury *et al.*, 2002). As Haslam (2006) stated, “the use of river plants as bioindicators is often undervalued. If autecology and the country of origin are known, and the species list long enough, interpretation is astonishingly accurate, for both natural factors and human impact”.

Invertebrates are an important tool for understanding and detecting changes in aquatic ecosystem integrity, and they can be used to reflect cumulative impacts that cannot otherwise be detected through traditional water quality monitoring. The broad diversity of invertebrate species occurring in aquatic systems similarly demonstrates a broad range of responses to different environmental stressors. Benthic invertebrates are relatively easy to collect, and they can be analyzed at many different levels of precision. They are sensitive to a wide variety of impacts that occur in the Ozark Highlands, such as changes in chemical constituents (including metals), hydrological alterations, sedimentation and bank erosion, and land use and other changes in the watershed. Furthermore, changes in the diversity and community structure of benthic invertebrates are relatively simple to communicate to resource managers, administrators, and park visitors, because the loss of biological communities is of interest and concern to these groups. Benthic community structure can be quantified to reflect stream integrity in several ways, including the absence of pollution sensitive taxa, dominance by a particular taxon combined with low overall taxa richness, or appreciable shifts in community composition relative to the reference condition (Plafkin *et al.*, 1989).

Fish communities of Ozark lotic systems are important components of their aquatic ecosystems. The Ozark Highlands is one of the richest areas of the United States for fish species. More than 175 native and introduced species occur in the Ozarks with several of these species being unique to this region (Petersen, 1998). The Current River basin is considered a “hot spot” for fish species that are designated as vulnerable or imperiled by The Nature Conservancy and the Natural Heritage Network (Master *et al.*, 1998). Because many of these species are considered intolerant of habitat alterations (Robison and Buchanan, 1988; Pflieger, 1997; Dauwalter *et al.*, 2003), fish community assemblages serve as a monitoring tool to assess changes in water and habitat quality for a number of ecological studies within the park (Hoefs, 1989; Petersen, 1998, 2004). In addition to their importance as environmental indicators, direct economic value can also be associated with several fish species that are actively sought by anglers. Because the public is familiar with fish as both an environmental indicator and as a recreational opportunity, the status and change in fish diversity is of concern to resource managers and is easily interpreted to park visitors.

Habitat conditions within a spring are based on its groundwater sources and a matrix of other factors (Danks and Williams, 1991). Terrain features shaped by the geology and topography determine the amount and variability of the water supply and the levels and variability of the temperature and water chemistry. Conditions above the surface are modified by the local climate, size of the spring, habitat diversity, and vegetation (both within and surrounding the spring). Species compositions associated with springs are influenced by differences in surrounding vegetation, substrate, pH, and other factors such as water chemistry (Williams and

Danks, 1991). Recent studies of the possible effects of global warming indicate that spring communities may be impacted due to increases in annual average water temperature (Hogg and Williams, 1996) and changes in discharge (Erman and Erman, 1995; Williams *et al.*, 1995).

History of Monitoring Springs at Ozark National Scenic Riverways

Biomonitoring of springs has not received the level of attention given to wadeable streams in North America. Initial biomonitoring efforts on surface waterways focused on detecting point source disturbances rather than vague non-point source problems. Karst systems are considered to be highly vulnerable to pollution and have limited self-purification potential due to reduced adsorption, making them vulnerable to biological and chemical degradation (Gibert, 1990). Thus, due to their vast and often unknown recharge areas, karst springs may represent the ultimate challenge in water quality protection.

Despite a large amount of literature on groundwater contamination and its complexity, there is almost no information on what happens when contaminated groundwater emerges at the surface through a spring and how it may affect surface water biota (Williams, 1991; Notenboom *et al.*, 1994; van der Kamp, 1995). Potential groundwater contaminants include toxic compounds such as heavy metals, pesticides and xenobiotic organics, nitrogen and phosphorous compounds, and organic matter from sewage or other animal wastes. All of these contaminants may cause adverse effects on groundwater fauna such as bioaccumulation, biotransformation, increased density, and distress by anoxia.

Early works on springs in North America provided comprehensive information on single spring systems (Davidson and Wilding, 1943; Sloan, 1956; Odum, 1957; Teal, 1957; Minckley, 1963; Minshall, 1968; Tilly, 1968; Stern and Stern, 1969; Wilhm, 1970), but there is a general lack of knowledge regarding the biological communities inhabiting different types of springs (Matthews *et al.*, 1983; Ferrington, 1995). This may have been because spring communities typically are represented by fewer species and have less diversity than streams due to relatively constant temperature regimes, mineralization (high dissolved solids), low dissolved oxygen, absence of plankton as a food source, and depauperate habitats (Van Gundy, 1973; Williams and Danks, 1991). However, over the previous two decades, there has been a resurgence of interest in the biodiversity of permanent springs in North America (Glazier and Gooch, 1987; Danks and Williams, 1991; Glazier, 1991; Gooch and Glazier, 1991; Williams and Danks, 1991; Erman, 1992; Blackwood *et al.*, 1995; Erman and Erman, 1995; Hargis, 1995; Mattson *et al.*, 1995; Webb *et al.*, 1995, 1998; Williams *et al.*, 1997; Poole and Bowles, 1999; Williams and Williams, 1999; Bowles *et al.*, 2003, Bowles *et al.*, 2007). This may be due to a growing interest in biodiversity and the accelerating loss of spring habitats worldwide (Erman and Erman, 1995; Bowles *et al.*, 2007).

Because of the complicated nature of monitoring large springs such as those at OZAR, most state agencies, including Missouri, have not addressed biomonitoring protocols for springs within their states. As of yet, there have not been any publications or protocols detailing relationships among the invertebrates, fish, vegetation, and water quality of the springs proposed for monitoring.

History of Aquatic Vegetation Monitoring

No long-term biomonitoring programs for aquatic vegetation have been conducted for the springs at OZAR. However, there have been some studies of algae and aquatic vegetation in these springs that collectively serve as a useful baseline for developing this protocol. Drouet (1933) reported on the algae found in Alley, Big, and Round springs in addition to listing a few unsubstantiated records for vascular plants. Steyermark (1941) conducted the first known inventory of aquatic vegetation in Missouri springs including several springs at OZAR and corrected several misidentifications apparently made by Drouet (1933). Currier (1990a, b) conducted floristic inventories for Alley, Big, Blue, and Round springs. These studies were intended to compare the aquatic plant diversity of these springs to Steyermark's study (Steyermark 1941). Currier concluded that the plant composition in several springs had changed since Steyermark's study, with some species disappearing and others documented for the first time. For example, Currier (1990b) noted that *Zannichellia palustris*, *Potamogeton foliosus*, and submerged stands of the non-native *Poa annua* documented from Big Spring in Steyermark's study were absent in his study. Lipscomb (undated) also conducted a floristic inventory of the aquatic plants occurring in the large springs at OZAR. His findings were similar to those of the previous studies, and he found these three species were present. Steyermark (1941) did not record *Lemna trisulca* or *Ranunculus longirostris* from Big Spring, but these species are now present and abundant there (Currier, 1990b; Lipscomb, undated). The findings of Steyermark (1941), Currier (1990a, b) and Lipscomb (undated) suggest the aquatic plant community in these springs is dynamic. Redfearn *et al.* (undated) conducted a botanical survey of OZAR and listed some aquatic species. However, their effort focused primarily on upland and wetland species. Conrad and Redfearn (1979), in a broad treatment of the mosses and liverworts, addressed some aquatic species known to occur at OZAR. Redfearn (1981) addressed the rich diversity of bryophytes occurring in some Missouri springs, including all of the springs included in this protocol. These springs each hold eight or more species of mosses and liverworts.

Converse (1994) conducted a study of macrophyte production in relation to water chemistry and nutrient dynamics at Big Spring. The focus of his research was on the production of three species: *Ranunculus longirostris*, *Nasturtium officinale*, and *Veronica comosa* (syn. *V. anagallis-aquatica*). Converse (1994) also reported water quality data for the large springs at OZAR, showing they were quite similar and stable (Table 1). Other constituents reported by Converse (1994) (*i.e.*, total particulate phosphorus, soluble reactive phosphorus, particulate organic nitrogen, dissolved organic carbon, calcium, magnesium, sodium chloride, alkalinity, and turbidity) were not constant seasonally. This data suggests that the aquatic plant communities of Big Spring, and other springs at OZAR, are vulnerable to disturbances and eutrophication associated with stormwater runoff. Converse (1994) further noted that the macrophyte production in Big Spring was one of the highest values ever reported for lotic ecosystems ($15 \text{ g-dw m}^{-2} \text{ d}^{-1}$), and the aquatic vegetation in the springs demonstrated a high biomass turnover.

Table 1. Physical-chemical parameters for large springs at OZAR (from Converse, 1994).

Spring		Temperature (°C)	Dissolved Oxygen (mg/l)	pH	Specific Conductance (µm/sec)	Turbidity (NTU)
Alley	Mean	14.16	9.38	n/a	310.58	0.73
	Median	14.2	9.33	7.21	311.65	0.69
	Range	14.00-14.30	9.11-9.83	7.03-7.45	296.00-320.30	0.53-1.26
	Variance	0.01	0.07	n/a	61.86	0.03
	N	8	8	8	8	8
Big	Mean	14.60	8.76	n/a	341.97	1.27
	Median	14.60	8.67	7.18	343.90	1.19
	Range	14.40-14.70	8.54-9.31	7.03-7.36	333.10-349.00	0.96-2.26
	Variance	0.01	0.06	n/a	27.57	0.17
	N	9	9	9	9	9
Blue	Mean	13.90	9.25	n/a	302.46	0.37
	Median	13.90	9.13	7.24	306.20	0.27
	Range	13.50-14.20	8.70-9.82	6.93-7.60	276.70-312.70	0.23-0.70
	Variance	0.04	0.15	n/a	135.19	0.03
	N	9	9	9	9	9
Round	Mean	14.48	8.87	n/a	332.81	0.68
	Median	14.45	8.88	7.24	336.50	0.69
	Range	14.20-14.70	7.59-9.89	6.98-7.60	313.6-342.10	0.53-0.85
	Variance	0.02	0.45	n/a	90.23	0.01
	N	8	8	8	8	8
Welch	Mean	13.98	8.83	n/a	331.60	0.56
	Median	14.00	8.75	7.15	336.80	0.58
	Range	13.9-14.10	8.58-9.34	6.95-7.41	309.50-338.90	0.43-0.72
	Variance	0.00	0.06	n/a	105.23	0.01
	N	8	8	8	8	8

Edwards (2002) surveyed the aquatic plants of the Jacks Fork River and included Alley Spring. He reported 19 species from Alley Spring, of which 11 were wetland type plants that were found on the banks and margins of the spring-run, while only eight species were truly aquatic. Edwards (2002) noted the occurrence of distinct plant communities occurring in the Jacks Fork River Basin and he further described the community at Alley Springs as being characterized as a *Cardamine bulbosa-Veronica catenata* (junior synonym of *V. anagallis-aquatica*) community. However, pilot surveys by HTLN staff during 2005 and 2006 showed the diversity of aquatic plants occurring at Alley Spring is higher than reported by Edwards (2002) and the community there is better characterized as a *Veronica anagallis-aquatica-Elodea nuttalli-Sparganium americanum* complex.

Padgett (2001) reported on the first occurrence of the exotic hydrophyte *Myriophyllum spicatum* (Eurasian watermilfoil) from several locations in Missouri including some from OZAR. Other non-native species known to occur in the springs include *Nasturtium officinale*, *Poa annua*, (Steyermark, 1941), and *Mentha piperita*.

History of Aquatic Invertebrate Monitoring

Several studies have been done on invertebrates occurring in Missouri springs outside of OZAR (Sullivan, 1928; Bonham, 1962; Doisy, 1984; Sarver and Kondratieff, 1997; Mathis, 1999), and in contiguous states such as Arkansas (Robison, 1981; Mathis, 1994; Hargis, 1995; Bowles, 1998; Jackson, 2001; Usrey, 2001), Iowa (Kennedy and Miller 1990), Illinois (Webb *et al.*, 1995, 1998; Bade *et al.*, 2002), Oklahoma (Matthews *et al.*, 1983; Bass, 2000; Gaskin and Bass, 2000; Rudisill and Bass, 2005), Kentucky (Minckley, 1963; Rayburn and Freeze, 1978), and Kansas (Blackwood *et al.*, 1995; Ferrington *et al.*, 1995). Other broader regional treatments of the Ozark Plateau include those of Holsinger (1989) and Koppelman and Figg (1995).

Published studies concerning invertebrates that include the springs and spring-runs of OZAR include information on their assemblages (Vineyard *et al.*, 1974; Gardner, 1984; Gardner and Taft, 1984; Gardner, 1986; Nielsen, 1996; Ferro and Sites, 2008) and specific families, orders, or classes such as Chironomidae (Blackwood, 2001), Ephemeroptera (Wiersema and Burian, 1999), Plecoptera (Poulton and Stewart, 1991), Trichoptera (Moulton and Stewart, 1996), Odonata (Trial and Belshe, 2002), Decapoda (Pflieger, 1996), Amphipoda (Sarver and Lister, 2004), and Gastropoda (Wu *et al.*, 1997). The Missouri Department of Conservation maintains a database with all known collection records for the state (Missouri Department of Conservation, 2003), and their Missouri Species of Conservation Concern Checklist (Missouri Natural History Program, 2003) lists several species of invertebrates from springs within the state. Species listed as rare and uncommon within Missouri that are known to occur in either Shannon or Carter counties include a stonefly, *Allocaenia pygmaea* (collected from Round Spring, date unknown) and the Salem Cave Crayfish (*Cambarus hubrichti*). Although numerous studies have addressed aquatic invertebrates at OZAR, no previous long-term monitoring of aquatic invertebrate communities have been conducted for the large springs.

History of Fish Monitoring

The Ozark Plateau has a rich diversity of freshwater fishes with 112 species of fish reported to occur in or near OZAR (National Park Service, 2005). Fish communities of Ozark streams and springs are important components of their respective ecosystems. Several species (including several darters, minnows, and madtoms) occurring at OZAR are considered intolerant of habitat alterations and poor water quality conditions (Robison and Buchanan, 1988; Pflieger, 1997; Dauwalter *et al.*, 2003). Therefore, fish assemblages are a useful monitoring tool to assess changes in water and habitat quality for a number of ecological studies within the park (Hoefs, 1989; Petersen, 1998, 2004). Large springs, such as those at OZAR, have been shown to serve as key thermal refugia for some stream fishes during winter and summer months when harsh temperature fluctuations occur in mainstem streams (Peterson and Rabeni, 1996). Although the fish communities of the Current River Basin are generally well known, there have been no exhaustive surveys or long-term monitoring conducted on the fish assemblages of the large

springs in this basin. Moreover, there have been few studies of spring-dwelling fish communities for the entire Ozark region (Matthews *et al.*, 1985).

Rationale for Selecting this Resource to Monitor

OZAR lies within the Salem Plateau, one of three subdivisions of the Ozark Plateau, and is composed predominantly of dolomite, limestone, and sandstone, with elevations below 245 m (Nigh and Schroeder, 2002). Accordingly, springs in this region typically are well-buffered with high pH, usually in the 7.5 – 8.5 range. Within the Ozark Plateau, the main factors affecting water quality include geology, land use, and population density. The geology of this area is dominated by mineral dissolution, ion exchange, and oxidation-reduction reactions (Adamski *et al.*, 1995). The Ozark Plateau is characterized by “karst” topography (springs, sinkholes, and caves), meaning that the surface and groundwater are integrally connected. One difference in the groundwater from karst systems is that it may be oxygenated, due to contact with air spaces within channels and caves. Another difference is that unlike groundwater that is filtered through dense soil layers, groundwater in karst systems often moves rapidly through underground channels that fail to provide effective natural filtration and absorption. As a result, these waters often contain contaminants and pollutants not found in groundwater from other types of systems.

Several studies have addressed the water quality of various springs and groundwater within OZAR (Missouri Bureau of Geology and Mines, 1926; Beckman and Hinchey, 1944; Aley, 1973a, b; Vineyard *et al.*, 1974; Aley, 1975, 1976a, b 1978; Barks, 1978; Aley and Foster, 1979; Aley and Aley, 1982; Aley, 1987; Converse, 1994; USGS, 1995; Adamski, 1996; Bell *et al.*, 1996; Adamski, 1997). Nutrient data from 395 groundwater samples in the Ozark Plateau showed that nutrient concentrations, in particular nitrite plus nitrate concentrations, were related to hydrogeology and land use. Approximately 4% of these samples had nitrite plus nitrate concentrations that exceeded the maximum concentration level allowed in drinking water by the EPA (Davis *et al.*, 1995). Additional studies of this problem reported that nitrite plus nitrate was the nutrient most often found, and indicated a relationship between the nutrients in groundwater and land use in the area (Steele *et al.*, 1987; Adamski, 1997).

Aley and others have provided an extensive series of reports delineating the groundwater recharge areas and the resultant potential problems for many of the springs within OZAR (Aley, 1973a, b, 1975, 1976a, b, Vineyard *et al.*, 1974; Aley and Foster, 1979; Aley and Aley, 1982; Aley, 1987). The crux of the problem for OZAR managers is that almost all of the recharge areas for the major springs within the park are outside the control of the NPS. A study of the OZAR watershed identified 378 point-source hazard areas within these privately held lands (Aley, 1987). These included: 1) sewage disposal facilities, 2) dumps, landfills, and salvage yards, 3) industrial sites, 4) transportation routes, including major pipelines, 5) petroleum storage sites, including service stations, and 6) chemical storage sites. Of specific concern are the effects of mining and waste disposal in the recharge zones, conversion of forestland to pasture, pipeline leakage, and impacts from recreational use such as increased nutrient input and bank erosion (Davis and Barr, 2006).

Due to monetary, logistical, and staffing constraints, this protocol will focus on the spring-runs of the permanent, high discharge, and coldwater springs of OZAR. Reasons for the choice of the

large springs include their value as major tourist attractions within the state, detailed mapping of recharge areas for the larger springs, and contribution of base flow to the Current and Jacks Fork rivers. The low variability in water chemistry and temperature of these spring-runs results in more stable biotic communities than those in smaller springs, aiding in higher statistical precision with less sampling effort.

Surveys of aquatic vegetation, fish, and invertebrate communities are important to researchers and managers to establish inventories and aid in the diagnosis of ecosystem disturbances such as water quality degradation and introduction of non-native (*i.e.*, exotic) species. The methods we propose here for monitoring are repeatable, straight-forward, and easy to understand, thus allowing more precise information to be collected by various personnel in a time-effective manner. This protocol will be a primary aid in determining the ecological integrity of the large springs at OZAR. Specifically, data collected from the springs will provide a baseline for assessing the potential for, or extent of, a variety of anthropogenic and natural disturbances.

Monitoring Objectives Addressed by the Protocol

Two broad objectives are addressed by this protocol: 1) Determine the annual status and trends in species diversity, abundance, and community metrics for vegetation, invertebrates, and fish occurring in the large springs at OZAR, and 2) Relate the community data to overall water quality and habitat condition (DeBacker *et al.*, 2005).

Justification/Rationale for These Objectives

The structure of biotic communities of OZAR springs has not been consistently inventoried or monitored. The respective watersheds and recharge areas of the springs remain largely unprotected, leaving the springs vulnerable to disturbance. Estimating natural variability of the vegetation, invertebrate, and fish communities within the springs will further aid in defining sample precision, the number of samples required, and the minimum detectable difference in mean community metric values. The initial years of data collection should adequately address the questions regarding natural variability and baseline water quality conditions. Measuring water quality, habitat structure and availability, and watershed land use patterns and correlating these with aquatic vegetation, invertebrate, and fish community composition will allow insight into the influences these variables have on the natural integrity of the springs at OZAR. A negative decline or trend from the baseline data could be indicative of impairment or disturbance of the springs and could be a basis for more targeted studies.

II. Sampling Design

Spatial Design

Establishing the sample frame

We have developed an integrated aquatic monitoring plan for springs at OZAR, which includes the co-location and co-visitation of invertebrate, fish, and vegetation sampling. This protocol only addresses spring communities occurring within NPS jurisdictional boundaries (Table 2).

This protocol is intended to evaluate the biological integrity of the springs downstream of the spring source and is not intended to assess the entire spring-run. Therefore, our common sample unit definition is a ‘reach’ of the spring-run of some minimum and maximum length. Because of physical limitations of accessing the spring sources, including extreme depth and crew safety, sampling will be conducted on equally spaced transects beginning at the first accessible area of each spring. The effective sampling area of each spring-run will be based on safety of personnel, accessibility, ability to co-locate sampling areas for measured parameters, and other pertinent factors determined by the investigators at the time of sampling.

Table 2. OZAR springs chosen for the spring monitoring protocol.

Spring name	Spring magnitude and type (Meinzer 1927)	County	Quad Map	GPS coordinates	Rate of Flow (m ³ /sec) (Vineyard <i>et al.</i> 1974)
Alley Spring	1st , conduit	Shannon	Alley Spring	N37° 9.247' W91° 26.499'	1.51 – 29.68
Big	1st , conduit	Carter	Big Spring	N36° 57.137' W90° 59.646'	6.61 – 36.40 (est.)
Blue	1st , conduit	Shannon	Powder Mill	N37° 9.956' W91° 9.789'	1.74 – 6.61
Round	2 nd , conduit	Shannon	Round Spring	N37° 16.953' W91° 24.458'	0.28 – 14.56
Pulltite	2 nd , conduit	Shannon	Round Sec. 5	N37° 20.103' W91° 29.408'	0.16 – 3.98
Welch	1st , conduit	Shannon	Cedar Grove	N37° 23.631' W91° 34.459'	1.96 – 9.27

Sampling Reaches within Springs

The source or mouth of a spring usually maintains a constant or near-constant temperature, while the downstream spring-run may vary in temperature due to distance from the source, riparian canopy, and other factors. This increase in downstream temperature variability can impose very different constraints on biological communities, resulting in longitudinal zonation of many of the taxa (Williams and Hogg, 1988; Erman, 1998; Cantonati *et al.*, 2006). Such longitudinal effects may introduce confounding information into biological sampling programs. Although many studies have focused on the spring source in an effort to increase sampling consistency, the sources of the large springs at OZAR are too deep to sample practically or safely. Therefore, this protocol proposes that the spring-run or brook immediately below the source will be sampled. Sampling multiple habitats often provides more comprehensive information about the invertebrate fauna compared to single habitat samples (Lenat and Barbour, 1994). The majority of habitats in the spring-runs, however, are runs and riffles.

At each spring, a sampling reach will be established that satisfies specific requirements necessary to obtain a representative and unbiased sample for all abiotic and biotic parameters being monitored (see SOP #3 for details on reach selection). The portion of the spring-run to be sampled is unique for each spring because of their unique sizes and other physical characteristics. To ensure a uniform sampling effort that is representative of each spring's total area, we applied a weighting factor that accounts for variation among average widths (Table 3). The weighting factor uses the minimum average width of 15 m recorded for Pulltite Spring and Welch Spring. To determine the weighting factor, the average width of the spring is divided by 15. This weighting factor is multiplied by 150 m, the minimum reach recommended by NAWQA (Moulton *et al.*, 2002), to determine sampling reach length. The designated sampling reach lengths will allow for inclusion of representative macrohabitats (riffle, run, and pool habitats) present within the springs, although run habitats dominate all other habitat types. Because the lengths of the springs are relatively short, the reach will begin as close to the spring source as possible. Once located, this reach will become a permanent sampling site barring dramatic alterations in spring morphology that would require re-establishing the sampling reach.

Table 3. Weighting factors and length of sampling reaches for large springs at OZAR.

Spring	Total Length (meters)	Average Width (meters)	Weight Factor (WF)	150 m X WF	Transect Interval (m)
Alley	800	19	$19/15 \approx 1.3$	190 m	19
Big	600	46	$46/15 \approx 3.1$	460 m	46
Blue	250	16	$16/15 \approx 1.1$	160 m	16
Pulltite	250	15	$15/15 \approx 1.0$	150 m	15
Round	300	24	$24/15 \approx 1.6$	240 m	24
Welch	36	15	$15/15 \approx 1.0$	36 m	18

Once the sampling reach is established, it is divided into 11 equally spaced transects beginning at the first accessible area of each spring and proceeding downstream (Fig. 1). The only exception to this will be Welch Spring, where only three transects will be used because the entire spring-run is less than 100 meters in length. Procedures for establishing sample points within each spring-run are described in SOP #9 (Physical Habitat Measurements).

Temporal design

For each spring at OZAR, the revisit design will have an annual revisit panel for invertebrates and aquatic vegetation, and a three year rotating panel for fish (Table 4). This strategy will yield maximum information on trend without inflicting damage to ecologically sensitive springs.

Little is known about the biotic communities of OZAR springs. Although these communities are quite diverse, the dynamic nature of their respective populations has never been addressed. Because of the thermal constancy of the springs and spring-runs, the sampling index period does not have to be constrained to any particular time period. Most of the aquatic hydrophytes occurring in the springs flower during mid-summer through early fall. Therefore, to ensure maximum association of invertebrate and fish communities and to allow for finding hydrophytes

in flower to aid field identifications, this protocol proposes to monitor aquatic vegetation concurrently with aquatic invertebrates and fish during the period 15 July-31 August. To the extent possible, temporal consistency will be maintained through successive years as well as between sample types.

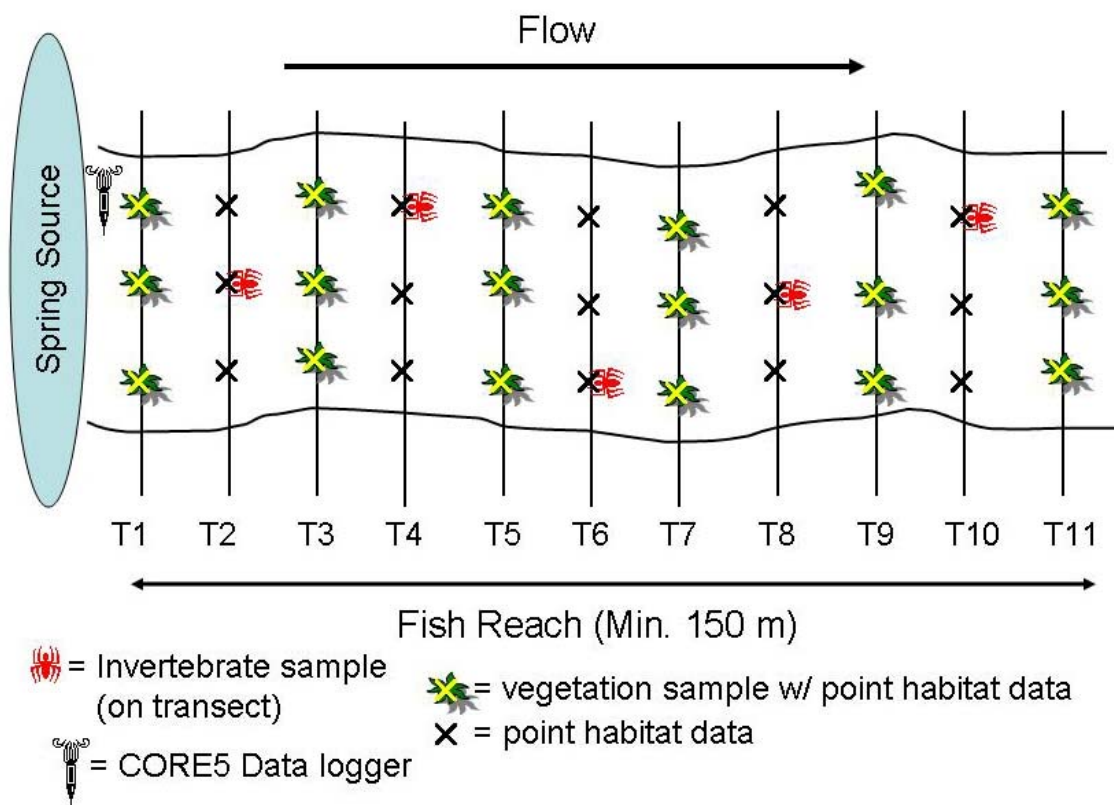


Figure 1. Transect location and layout within a spring-run.

Response Design

The sampling approaches for collecting physical habitat, invertebrates, and fish in this protocol are comparable to those of the US Environmental Protection Agency (EPA) Environmental Monitoring and Assessment Program (EMAP) (Lazorchak *et al.*, 1998; McCormick and Hughes 1998, 2000) and the US Geological Survey (USGS) National Water Quality Assessment (NAWQA) program (Fitzpatrick *et al.*, 1998; Moulton *et al.*, 2002). However, this protocol has been adapted to meet the specific objectives of the long-term monitoring program while considering limitations of staff size and budget. Modifications from these national-level protocols for the purposes of this monitoring program were taken from literature pertaining to sampling of biotic communities in lotic systems.

Table 4. Proposed revisit plans for monitoring studies at six large springs at OZAR. A black square (■) indicates all sample units in that panel are to be visited that year.

Study	Spring	Year												
		Revisit Notation	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018
Vegetation & Invertebrates	ALL	[1-0]	■	■	■	■	■	■	■	■	■	■	■	■
Fish	Welch, Alley	[1-2]	■			■			■			■		
	Round, Blue	[1-2]		■			■			■			■	
	Big, Pulltite	[1-2]			■			■			■			■

Habitat and Water Quality

Habitat incorporates all aspects of physical and chemical constituents and their interactions. Variables such as current velocity, discharge, dominant substrate size, embeddedness, water chemistry, and presence of periphyton, filamentous algae and aquatic plants play key roles in the microhabitat structure and distribution of aquatic invertebrates and fish (Rosenberg and Resh, 1993; Allan, 1995; Hauer and Lamberti, 1996). Other habitat variables such as woody debris, boulders, canopy cover, and bank condition (*e.g.*, height, angle, dominant substrate, degree of undercut, and vegetative cover) also are important for assessing spring condition. We propose to monitor all of the aforementioned habitat variables at our sampling sites.

Aquatic Vegetation

We propose to collect cover estimates of the aquatic hydrophytes, mosses, and certain filamentous and globular algae among 1 m² sampling plots located along fixed transects (Fig. 1) to evaluate vegetation community changes through time (SOP #5 “Aquatic Vegetation Sampling”). Cover estimates within the sample plots will be collected using the Daubenmire cover classes (Daubenmire, 1959) that partitions all possible percentage values into seven categories. Percent cover estimates allow us to obtain information on trends in vegetation communities and relate those to changes in habitat.

Aquatic Invertebrates

The sampling approach described here for invertebrates is based on the EPA EMAP protocols (Lazorchak *et al.*, 1998). Minor modifications to the EMAP approach are made in this protocol to account for program-specific goals related to long-term monitoring of invertebrates, limitations posed by staff size, and logistical and budgetary constraints. The EMAP approach focuses on evaluating ecological conditions on regional and national scales although such an application is not appropriate for the springs at OZAR, although the transect-based methods remain appropriate.

There is an overriding concern that collecting too many benthic samples annually from these delicate springs could cause long-term or permanent damage. For example, Englund (1991) found that the moss community in disturbed plots in a spring-run took over one year to recover following disturbance. Doisy and Rabeni (2004) collected 6 samples from each of the large OZAR springs but only analyzed three of those samples. They also used benthic subsampling procedures that were similar to those recommended in this protocol (SOP #7, Laboratory Processing and Identification of Invertebrates). Their analyses showed the various community metrics were fairly consistent among samples within spring-runs (Table 5). They also found that data from the subsamples adequately represented the contents of the entire samples (Table 6). However, Doisy and Rabeni (2004) restricted their sampling to the area of the spring-runs immediately downstream of the source. They did not collect samples from throughout the entire spring-run as proposed in this protocol. Other studies have similarly shown that three benthic samples are generally adequate to characterize the invertebrate communities occurring in a single habitat type (Canton and Chadwick, 1988; Bowles, 1989; Mathis, 2001; Usrey and Hinsey, 2006). However, to better characterize the spring-runs and their available habitats along their respective continuums, this protocol recommends collecting five benthic invertebrate samples per spring with the individual samples being collected on transects 2, 4, 6, 8, and 10 (Fig. 1). Sampling on successive transects will begin at river left, then mid-channel, and then river right. This sequence is repeated until all samples are completed. In subsequent sampling years, the sampling order is shifted (*i.e.*, sampling at year two will begin at mid-channel). This will result in a specific location on a given transect being sampled every three years and will minimize long-term disturbance to the spring-run.

Fish

Collection of fish community data will closely follow USGS NAWQA protocols (Moulton *et al.*, 2002). Fish will be collected within one reach of each large OZAR spring. The reach will be representative of the spring-run, containing various instream habitats and geomorphic channel units characteristic of the spring-run. Many spring-runs consist primarily of run or riffle channel units. Therefore, the representative reach may only contain these channel units.

Electrofishing techniques will be used to collect fish. A single pass will be used for wadeable reaches (those less than 1.5 m deep) and two passes will be used in non-wadeable (those deeper than 1.5 m) reaches. Depending on the size of the spring-run (width and depth), communities will be sampled using backpack or towed barge gear in wadeable reaches and boat electrofishing

Table 5. Metrics for invertebrates based on three samples collected from each spring in Fall 2003 (mean and 95% confidence intervals). Welch Spring data is based on only one sample.

Spring	Taxa Richness	EPT Richness	Mayfly Richness	Shannon Diversity Index	Biotic Index	Ratio of Mayflies/ Total Abundance	Ratio of Shredders/ Total Abundance
Alley	25.3 (23-27.7)	11.3 (10.7-12)	3.3 (2.7-4)	1.74 (1.58-1.9)	2.43 (2.23-2.63)	35.3 (28.6-42)	46.8 (41.2-52.4)
Big	12 (9.7-14.3)	2.3 (1.7-3)	0.7 (0.01-1.3)	0.94 (0.71-1.18)	2.02 (1.3-2.74)	0.1 (0-0.2)	74 (66.4-81.6)
Blue	14.3 (13-15.6)	3.3 (2-7.4)	0.7 (0.01-1.3)	1.37 (1.17-1.56)	2.35 (1.65-3.05)	0.3 (0.04-0.5)	49 (36-61.3)
Pulltite	20 (18.9-21.1)	6.7 (4.3-9)	3.3 (2.7-4)	1.67 (1.58-1.77)	5.32 (5.08-5.56)	16.2 (12.9-19.4)	4.9 (4.5-5.4)
Round	16.3 (15-17.6)	4 (1.7-6.3)	1 (1.2-2.09)	1.65 (1.2-2.09)	6.39 (5.72-7.05)	0.9 (0.3-1.4)	4.3 (0-8.7)
Welch	24	4	3	1.80	4.5	8	40

Table 6. Comparisons of benthic metrics for whole invertebrate samples and 400 specimen subsamples.

Sample	#1		#2	
Metric	Whole Sample (CV)	Mean of subsamples	Whole sample	Mean of subsamples (CV)
Biotic Index	2.34	2.34 (7%)	2.60	2.62 (4%)
Shannon Diversity Index	1.53	1.49 (8%)	1.86	1.74 (7%)
Ratio of mayfly/total abundance (%)	36.5	36.8 (24%)	37.4	37.0 (13%)
Ratio of shredders/total abundance (%)	50.0	49.8 (17%)	41.6	41.8 (13%)

CV=Coefficient of variation

equipment in non-wadeable reaches (see Table 7). Some portions of non-wadeable spring-runs may be shallow and require use of towed barge or backpack electrofishing equipment to collect small benthic species. Techniques for applying these sampling methods are presented in SOP #8 (Fish Community Sampling).

Fish processing will closely follow NAWQA procedures. At a sample reach, a subsample of fish from each species will be measured (lengths and weights) and anomalies recorded; the remaining fish of each species will be counted for abundance estimates. For details on fish processing and data recording see SOP #8.

Table 7. List of spring reaches sampled for fish communities and list of gear used and percent effort by gear for each reach.

Spring	Panel	Reach Type	Gear Used	% Effort by Gear
Welch	1	Wadeable	Backpack	100
Alley	1	Wadeable	Towed Barge	100
Round	2	Wadeable	Backpack	100
Blue	2	Wadeable	Towed Barge	100
Big	3	Non-wadeable	Boat, Towed Barge	50, 50
Pulltite	3	Wadeable	Backpack	100

Rationale for the Sampling Design

Biomonitoring methodologies are constantly being developed, refined, and debated in an effort to achieve the most efficient and effective assessments of the relations of water quality to invertebrate, fish, and vegetation communities. However, there have not been any published protocols designed specifically for biomonitoring of springs. While several biomonitoring methodologies have been developed for use on streams within the Ozarks (Rabeni *et al.*, 1997; Doisy and Rabeni, 1999; Rabeni *et al.*, 1999; Mathis, 2001; Rabeni and Wang, 2001; Zweig and Rabeni, 2001; Sarver *et al.*, 2002; Bowles *et al.*, 2007)), none are entirely appropriate for the springs at OZAR and are not used in this protocol. Therefore, the sampling design described in this protocol is primarily based on existing national level stream biomonitoring protocols (Lazorak *et al.*, 1998; Barbour *et al.*, 1999; Moulton *et al.*, 2002) with minor modifications based on pilot data collected from the springs at OZAR in October of 2003 and 2005.

Habitat and Water Quality

Habitat features are major, often limiting, determinants of invertebrate community structure and accordingly they are especially important for proper determination of biomonitoring results and assessment of ecological integrity (Barbour *et al.*, 1999). Although habitat incorporates all aspects of physical and chemical constituents and their interactions, variables such as current velocity, substrate size, embeddedness, water chemistry, sediment deposition, and presence of periphyton, filamentous algae, and aquatic plants play key roles in the microhabitat structure and distribution of aquatic invertebrates and fish (Rosenberg and Resh, 1993; Allan, 1995; Hauer and Lamberti, 1996).

Biological and environmental correlates of water quality and habitat structure compared across time are powerful tools for assessing disturbances related to natural and anthropogenic impacts on aquatic communities. As such, they are useful for detecting change and elucidating patterns

and trends in long-term data sets (Moulton *et al.*, 2002). For example, as habitat conditions degrade (*e.g.*, water quality decreases, embeddedness increases), degradation of the benthic spring communities are expected to follow. However, the relationship of cause and effect of these variables on aquatic community structure can be difficult to assess and analyze because there often is a broad response range among the resident species (Norris and Georges, 1993). Therefore, any association of community structure with these variables or their combinations must be interpreted cautiously and be based on real biological properties. These limitations withstanding, spring community structure, when viewed in association with environmental variables, can be an effective indicator of ecosystem change (Reice and Wohlenberg, 1993). In combination, such data are useful for providing managers an integrated assessment of water quality.

Aquatic Vegetation

There are no existing national or regional-level protocols that address monitoring aquatic vegetation in springs. Those protocols developed for stream vegetation monitoring in the United States and Europe (Yin *et al.*, 2000; Scott *et al.*, 2002; Veit and Kohler, 2003) often employ destructive sampling methods or are aimed at control or management rather than conservation. Because the springs at OZAR are fragile and prone to long-term damage from destructive sampling, we consider the previously referenced protocols inappropriate and will not use such methods.

Cover estimates allow for describing changes in vegetative cover, and allow for exploration of the correlative relationships between compositional changes and habitat attributes. Use of cover estimates are essential for many species where individuals of a species can rarely be identified and counted. Specific estimation of percent cover done by eye can be subject to problems of observer-bias. However, use of cover classes reduces the problem of observer-bias through partitioning all possible values into percentage categories. To reduce potential observer-bias, Daubenmire cover class estimates are used in this protocol for all cover estimates. Although cover class estimation is superior to percent cover estimates, the effect of different observers can be an important contributor to variability in the data set (Kercher *et al.*, 2003). On the other hand, Klimeš (2003) noted that variation of total plant cover estimates among observers tended to decrease as a function of increasing plot size. The 1 m² used in this study are well within the size range for plots that tended to exhibit the lowest amount of variation among observers (Klimeš, 2003). Although Klimeš (2003) recommended that such error can be minimized by using the estimates of at least three observers, we feel that using multiple 1 m² plots coupled with annual training requirements and detailed guidance for aquatic vegetation monitoring (SOPs #2 and 5) serves to minimize potential variation among observers.

Aquatic Invertebrates

Biomonitoring methodologies are constantly being developed, refined, and debated in an effort to achieve the most efficient and effective assessments of the relations of water quality to invertebrate, fish, and vegetation communities. However, there have not been any protocols published that were designed specifically for biomonitoring of springs. While several biomonitoring methodologies have been developed for use on streams within the Ozarks (Rabeni

et al., 1997; Doisy and Rabeni, 1999; Rabeni *et al.*, 1999; Mathis, 2001; Rabeni and Wang, 2001; Zweig and Rabeni, 2001; Sarver *et al.*, 2002) none are entirely appropriate for the springs at OZAR and are not used in this protocol. Therefore, the sampling design described in this protocol is primarily based on existing national level stream biomonitoring protocols (Lazorak *et al.*, 1998; Barbour *et al.*, 1999; Moulton *et al.*, 2002) with minor modifications based on pilot data collected from the springs at OZAR.

The EMAP program uses probabilistically selected sites where individual sampling sites are assessed using a transect-based design and community biological metrics are tied to habitat structure. Kick-net samples collected from flowing water habitats (*e.g.*, riffles, runs) are combined into a single composite sample for the stream reach while kick net samples collected from pool habitats are combined into a separate composite sample. This protocol does not propose to composite samples by habitat, and samples will be processed individually to gain a better estimate of intra- and inter-transect variability and allow a broader suite of analytical options. The “kick net” used in the EMAP method is effectively the same net as a Slack-Surber sampler used in the USGS NAWQA program (Moulton *et al.*, 2002) minus the frame delineating the sampling area in front of the net. We have opted to use the NAWQA style Slack Surber sampler because it is more useful for delineating the sampling area. Data are analyzed following Barbour *et al.* (1999) and the use of either multimetric or multivariate approaches. In addition, some programs use O/E (Observed/Expected) Ratio of Taxa Loss to assess invertebrate community degradation. This tool is a ratio comparing the number of taxa expected (E) to exist at a site to the number that are actually observed (O). The taxa expected at individual sites are based on models developed from data collected at reference sites. The current protocol does not use O/E ratios particularly because there are no reference sites available for establishing expected taxa. The EPA’s Wadeable Streams Assessment Program is based on the EMAP approach and is not considered separately here (USEPA 2004a, b, c, d, 2006).

Other EPA monitoring programs for assessing water quality using invertebrate communities in wadeable streams include the Rapid Bioassessment Protocols for Use in Streams and Rivers (Barbour *et al.*, 1999). This approach uses either single habitat (*e.g.*, riffles) or multi-habitat approaches and both involve collecting samples from a 100 m reach determined by the investigator to be representative of the characteristics of the stream. The single habitat approach involves sampling using a kick-net with a 1 meter area sampled in front of the net and taking 2-3 kicks using foot agitation. The multi-habitat approach uses 20 jabs or kicks taken from different representative habitat types using a D-frame dipnet. For both approaches, samples are composited for analysis and metrics are the same or comparable to those used in this protocol (Barbour *et al.*, 1999). An additional set of protocols designed for larger non-wadeable rivers (Flotemersch *et al.*, 2006) generally are not applicable to the spring-runs addressed in this protocol and are not further addressed here.

The general basis of the NAWQA program is to collect biological, physical, and chemical data at sites that represent major natural and anthropogenic factors considered responsible for controlling water quality in a river basin (Moulton *et al.*, 2002). The NAWQA sampling design for benthic invertebrates includes two types of sampling sites: basic fixed sites and synoptic sites. The fixed sites are those where parameters are measured over long periods of time and, as such, they are analogous to the sampling sites used in this monitoring protocol. The NAWQA

synoptic sites are used for one-time collections and therefore are not included in this protocol. Additionally, the NAWQA program conducts water-quality assessments in sampling reaches defined as the presence of two repeating geomorphic channel units such as a sequence of pool-riffle-pool-riffle. From these sampling reaches, two broad types of benthic samples are collected to characterize the invertebrate community: 1) semi-quantitative benthic samples collected from targeted habitat types, and 2) a composite qualitative sample collected from a broad variety of habitats from throughout the reach. The semi-quantitative benthic samples recommended by NAWQA are collected from richest-targeted habitat type (riffles for Ozark streams) using a Slack-Surber sampler (Moulton *et al.*, 2002). The number of individual benthic samples to be collected is not specified in the NAWQA protocol and depends on study objectives. Collected samples are partially processed in the field and subsequently composited into a single bulk sample. The NAWQA protocol allows for location of sites based on whether or not the site is representative of the local area and support objectives, thus giving the site investigator flexibility in establishing site boundaries depending on local conditions. The sampling design in this protocol, by comparison, employs a fixed transect design due to the limited relative length of the spring-runs.

Overall, our study design is closest to that of EMAP, and we opted not to use strict EPA or NAWQA monitoring approaches. However, there are some similarities among the EPA and NAWQA approaches and the present protocol will allow for comparison of data. Indeed, Peterson and Zumberge (2006) generally found no significant differences between invertebrate samples collected from riffles using the NAWQA and EMAP protocols. Because this protocol uses many of the same metrics employed in the former two protocols, we contend that the individual metrics and multimetric indices will be comparable among all three protocols.

Fish

There are no existing protocols specifically for sampling fish in large spring systems. However, several different sampling approaches or protocols have been used by state and federal agencies to quantify status and trends of fish communities in streams. Rapid Bioassessment Protocols developed by the EPA (Barbour *et al.*, 1999) have been used by many agencies to evaluate fish communities in streams. These protocols are designed to give a quick, broad picture of stream quality and fish assemblages throughout a region with minimal field and laboratory efforts. Other monitoring groups use the EPA EMAP protocols for wadeable (McCormick and Hughes, 1998) and non-wadeable streams (McCormick and Hughes, 2000), and the USGS NAWQA protocols (Moulton *et al.*, 2002). These latter two protocols have more rigorous data collection (*i.e.*, collection of fish lengths and weights) and quantitative methods (*i.e.*, designated reach length), giving a more complete picture of fish assemblage composition and structure. Therefore, this protocol follows methods similar to NAWQA protocols with minor modifications based on EMAP and other relevant literature. These modifications were necessary to meet specific requirements of this monitoring program under staffing and budgetary limitations, to ensure that sampling effort (*i.e.*, proportion of area sampled) was consistent across springs of different sizes, and to account for logistical constraints of sampling spring-runs.

The fish community protocol for sampling spring-runs is most similar to NAWQA in terms of site selection, minimum reach length required, electrofishing gear used, and data collection for

fish communities, but differs in terms of reach length sampled, gear used, and electrofishing effort. NAWQA sites are selected based on professional judgment and other criteria such as access, presence of streamflow instrumentation, land use characteristics, and other specific objectives. Use of professional judgement and safe access to the spring-run is used here to determine the upstream boundary of the sample reach within a spring-run. NAWQA uses a reach length of 20 times mean wetted stream width (at low flow) with a minimum length of 150 m and a maximum of 1000 m. This protocol also uses a minimum reach of 150 m; however, a weighting factor based on average width is used to establish the reach length. Applying NAWQA's multiplier to springs at OZAR would result in reach lengths for some of over 1000 m. Because sampling a reach length of this size is logistically and monetarily impractical, and most spring-runs at OZAR are less than 500 m, this protocol uses the NAWQA minimum sampling reach of 150 m multiplied by a width-based weighting factor to allow for scale-dependent sampling among the springs. A second modification from NAWQA is the strict use of electrofishing techniques in this protocol. NAWQA protocols allow for use of seining in riffles and shallow margins of pools and run habitat, but dense aquatic vegetation in these springs precludes effective seining. Thus, only electrofishing methods will be used.

A third modification is the use of single pass electrofishing in wadeable reaches. NAWQA protocols specify that two passes be used in wadeable streams. Single pass electrofishing corresponds with methods described in EMAP protocols (McCormick and Hughes, 1998, 2000). The advantage of single pass electrofishing is that a site can be sampled using fewer person-hours, thus reducing costs while increasing time efficiency. Simonson and Lyons (1995) found that catch per effort (*i.e.*, catch per unit time) in one pass provided the same values for species richness and percent species composition as depletion sampling (3-4 passes), and took only one quarter the time required for depletion sampling. However, a study by Meador *et al.* (2003) found the number of species collected after two passes was greater than the number of species collected after a single pass in 50.3 percent of the samples. Meador *et al.* (2003) also concluded that multiple pass electrofishing at a large number of sites across a large geographic area may not be cost effective.

Suitability of Survey Design to Meet Study Objectives

Monitoring objectives are integral to defining the sampling design. The sample design for spring community monitoring is driven by the study objectives discussed above. The overall survey design is deemed suitable for several reasons:

1. *Appropriate for long-term monitoring of aquatic communities.* A transect based approach for sampling and monitoring aquatic communities is consistent with other national level protocols (Lazorchak *et al.*, 1998; USEPA, 2004a, b, c, d). Sampling multiple habitats provides more comprehensive information compared to single habitat samples (Lenat and Barbour, 1994; Moulton *et al.*, 2002). However, run habitat is predominant in the large springs at OZAR.
2. *Appropriate for Ozark springs.* The data generated from this study design will be directly comparable to those of other regional (state and federal) monitoring programs, where applicable, that employ similar methodologies.

3. *Accommodates springs of varying size.* The sample design allows for unbiased estimates of community condition applicable to the entire spring-run regardless of length. While the sampled stretches must be long enough to accommodate unbiased estimates for all studies, they do not have to be the same size among springs.

4. *Easy to learn and use.* Field procedures are easy to use and repeatable over time by different sampling crews. Implementation does not require extensive time or costly equipment.

5. *Sequence of sampling events and revisit design allows for the greatest amount of field work to be accomplished per year while minimizing cost.* Because staff available for manning field crews is limited, and travel costs associated with monitoring are high, this strategy allows cost effective monitoring of the springs.

III. Field and Laboratory Methods

Field work conducted using this protocol will normally take two days per spring to accomplish. The first day will include deploying the datasonde for collecting CORE 5 measurements, assessing point and instream habitat and aquatic vegetation, and collecting invertebrate samples. The second day is devoted to collecting fish samples and assessing riparian habitat variables. A work flow diagram for collecting samples is shown in Fig. 2.

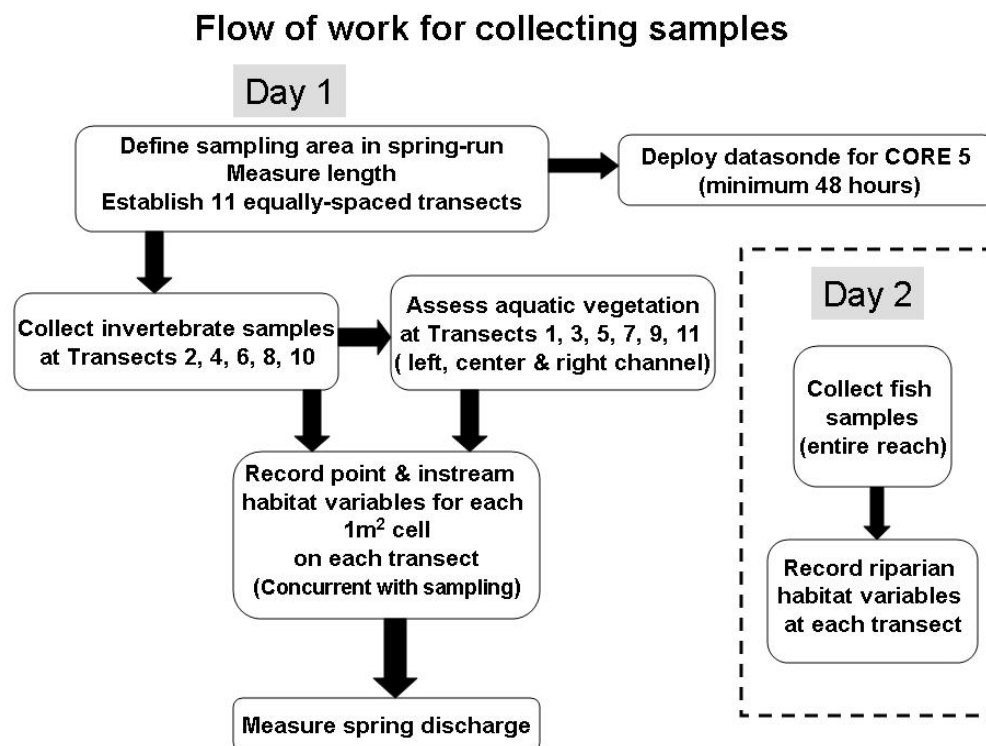


Figure 2. Flow of work diagram for collecting samples in the field.

Field Season Preparations, Field Schedule, and Equipment Setup

Procedures for field season preparations, including preparation of a field sampling schedule and equipment setup, are described in SOP #1 (Preparation for Sampling). Team leaders should ensure that team members have read and understand the protocol and supporting SOPs prior to sampling and that all required equipment and supplies have been ordered and are in proper working condition. Fieldwork must be scheduled in advance so that crews can be assigned. Time spent at a sampling reach will vary, but eight or more hours are typical. Sampling should occur in July-August when spring flows are low allowing for efficient and safe sampling. The team leaders will prepare and maintain a field notebook detailing all sampling-related activities and staff participation during monitoring trips to ensure that trip reports are complete and accurate. Finally, the team leader should ensure that all required scientific collection permits have been obtained.

Measuring Core 5 Water Quality and Spring Discharge

CORE 5 water quality parameters (temperature, dissolved oxygen, specific conductance, and pH, turbidity) will be recorded using a data logger or sonde. The datalogger will be deployed immediately upstream of the sampling area and allowed to operate for a minimum of 48 hours. Instructions for using the datalogger are located in SOP #4 (Documenting CORE 5 Water Quality Variables). Spring discharge will be measured at each site after invertebrate collections have been completed and preferably upstream of the sampling site. Instructions for measuring spring discharge are in SOP #10 (Measuring Spring Discharge).

Assessing Point and Instream Habitat

At each transect, channel unit type (*i.e.*, riffle, run, glide, pool) and pool form (if applicable) are recorded. In-stream habitat is collected within three 1 m² plots at center channel and half the distance between center and the left and right banks. Procedures for locating sample points within each spring-run are described in SOP #3 (Reach Selection). Wetted width of each transect is measured and depth, and current velocity are measured immediately in front of the sampling frame (see SOP #9). Dominant substrate (Wentworth Scale, Wentworth, 1922) is assessed within the sample plot. Additional categorical variables to be assessed include percent periphyton, percent filamentous algae, percent organic material, and canopy cover. Standard classes for all percentage estimates will be as follows: 0 = Absent (0%), 1 = Sparse (<10%), 2 = Moderate (10-40%), 3 = Heavy (40-75%), and 4 = Very Heavy (>75%).

Fish-Related Habitat Data Collection

Habitat composition within a stream is an important component in shaping aquatic communities. The type and abundance of specific habitats (*i.e.*, riffles, woody debris, pools, etc.) will influence species presence and relative abundance, as well as size structure, of the populations. Physical habitat data therefore will be collected every third year in conjunction with fish sampling to examine relationships between environmental conditions and fish communities. These methods

have been modified from NAWQA protocols (Fitzpatrick *et al.*, 1998) to meet the objectives of the NPS. Fish cover is assessed at each transect. Boulders and any artificial cover are assessed at each plot where depth, velocity, and substrate are collected. Small and large woody debris are assessed within a 1 m band on either side of a transect on the left and right sides of center channel. Cover along the banks is assessed within 1 m upstream and downstream of a transect and includes trees/roots, overhanging vegetation, undercut banks, and bluffs (within 5 m of wetted edge). Bank characteristics are observed at each transect to determine bank stability. Bank angle and dominant substrate are observed from the bottom of the bank (*i.e.*, at wetted edge or at the top base of the bar, if one is present), and the category code is recorded. Percent vegetative cover, bank height, and bank cover are assessed from the bank bottom to 10 meters into the bank. Bank cover categories include large trees, small trees/shrubs, grass/forbs, bare sediment, and artificial cover.

Assessing the Aquatic Vegetation Community

Percentage composition of the aquatic vegetation community will be assessed in three, equally spaced 1 m² sample cells located along each of 6 transects (Fig. 1). Welch Spring will have only three transects because of its short length. A modified Daubenmire scale (Daubenmire, 1959) is used to categorically estimate species areal coverage. Standard classes for all cover estimates will be as follows: 1= 0-0.99%, 2= 1-5%, 3= 5-25%, 4= 25-50%, 5= 50-75%, 6= 75-95%, and 7= 95-100%. Refer to SOP #5 (Aquatic Vegetation Sampling) for further details related to assessing the spring plant communities.

Collecting Benthic Invertebrate Samples

Procedures for collecting benthic invertebrate samples and documenting habitat data are presented in SOP #6 (Aquatic Invertebrate Sampling). Additionally, we propose collecting a substrate sample from within the invertebrate net sampling frame for measurement (SOP #9 (Physical Habitat Measurements)).

One invertebrate sample will be collected from sample points located on each of five transects in each spring as described in SOP #6. Only three benthic samples will be collected from Welch Spring because of the short spring-run. Samples will be collected with a Slack Surber sampler (Moulton *et al.*, 2002). Water may flow over the top of the net in deep runs potentially allowing some invertebrates dislodged from the substrate to wash over the net. Because the metrics to be calculated are largely percentage-based, such losses of invertebrates should not negatively affect the metric scores and their loss must be considered relative to the restrictions quantitative sampling gear (*i.e.*, Hess sampler) would impose on sampling effort. Each discrete sample is collected while progressing in an upstream direction. Sampling procedures will be the same for each sampling point and, whenever possible, samples should be collected by the same person to limit variability in sample techniques.

Substrate Size Assessment

Substrate assessments provide a unique characterization of the streambed composition at the time sampling takes place. Therefore, substrate will be measured from the area within the sampling

frame of the net (0.25m²) following collection of habitat data listed above, and it will be measured for every sample. The intent of this substrate assessment is to characterize the dominant substrate for individual samples, not to fully characterize all sediments present. This assessment will help us describe the prevailing microhabitat conditions that influence the structure of invertebrate communities and may help explain variability between sample points. Substrate will be measured based on the standard Wentworth scale. Procedures for collecting and measuring substrate samples are provided in SOP #9 (Physical Habitat Measurements).

Benthic Sample Processing and Specimen Identification

Procedures for processing benthic samples and identifying specimens are described in SOP #7 (Laboratory Processing and Identification of Invertebrates). Methods for preparing samples for sorting and subsampling generally follow those presented in Moulton *et al.* (2002). A list of the aquatic invertebrate taxa known from the greater Ozarks region is shown in SOP #12 (Data Analysis).

Subsampling Benthic Samples

Because of the relatively high densities of aquatic invertebrates occurring in the samples, subsampling individual samples will be necessary to process the samples in a time and cost effective manner. The routine for subsampling benthic samples is presented in SOP #7. The method of subsampling will involve the fixed fraction approach, with 25% of each sample being sorted following thorough washing, agitation, sieving, and elutriation of the entire sample (Moulton *et al.*, 2002). Additionally, a “large and/or rare” taxa component will be included where large or rare taxa that clearly are not in the sorted fraction are removed and stored in a separate vial for the purpose of reflecting accurate sample species richness estimates and calculating specific metrics such as EPT. A fixed fraction subsampling routine was selected over a fixed count routine because some metrics to be calculated from samples are related to specimen density that cannot be obtained with the latter method. Subsampled fraction debris will be subjected to QA/QC analysis (SOP #7) and should be kept until QA/QC is complete for that batch of samples and the Program Leader authorizes disposal of the debris.

Collecting Fish Samples

Fish communities will be sampled using various electrofishing methods (SOP #8) and associated habitat will be measured (Fig. 2). The size of the spring-run (width and depth) will determine the type of electrofishing gear used. Wadeable reaches will be sampled using backpack or towed barge electrofishing units. Non-wadeable reaches will be sampled with boat electrofishing equipment. At sites where depth requires that boat electrofishing be used, towed barge or backpack electrofishing will also be used to collect fish in shallow areas to obtain a representative sample of the reach.

When monitoring, it is important to note that gear type and gear efficiency have been shown to affect fish community data. In a study of fish data from 55 NAWQA sites, Meador and McIntyre (2003) found that among electrofishing methods (backpack, towed barge, and boat), Jaccard’s (similarity) index and percent similarity index values between years and between

multiple reaches were significantly greatest for backpack electrofishing. These results suggest that data collected using different gear (or different combinations of multiple types of gear) will be affected by gear type.

There are three alternatives to resolve the problem of analyzing data collected by different gear. First, the data can be considered to be affected primarily by the size of the spring-run when gear usage is based on the spring-run size; and therefore, data are treated as equivalent across gear types and combined for analysis. Second, data can be compared only with other data collected using the same gear types. Third, the raw data can be corrected for differing gear efficiencies before making comparisons across sites associated with different gear types.

This protocol is concerned with monitoring each spring-run over time, not comparing across springs that may use different gears or combination of gears. Therefore, when monitoring temporal changes in communities, it is imperative that gear type (or combination of gear types) used in a spring-run and sampling effort for each gear type be consistent across years (see Table 7). For reaches where multiple gears are used, we will combine the data across gear types for analysis because samples collected with electrofishing gear are based on time (*i.e.*, effort) and percent effort by gear is consistent across years. However, there may be specific monitoring questions where analyzing data by electrofishing gear is necessary. Therefore, in the field, data from different electrofishing gear will be kept separate.

When processing samples and recording data, all sample data (gear used, time spent sampling, electrofishing settings, length of the spring-run sampled, and species data collected with the gear type) will be recorded separately for each method. To the extent practical, individuals will be identified in the field using appropriate fish identification keys and other information. Specimens that cannot be reliably identified in the field will be preserved for identification in the laboratory (see SOP #8). Individual lengths and weights will be collected on a subsample of each species at a site to estimate the size structure and community composition; anomalies will also be recorded to determine the occurrence of diseases and deformities in the fish populations.

Sample Storage and Reference Collection

Aquatic Vegetation

A reference collection of each species identified in the field will be mounted and labeled on herbarium sheets and stored at the Missouri State University Herbarium, Springfield, MO.

Aquatic Invertebrates

Identified invertebrate samples are stored in 4 dram, glass vials with polycone caps and filled with 70% isopropyl alcohol. Specimen vials will be labeled with the taxon name, date collected, park and site names/code, and name of identifier. Organisms will be retained for at least three years and stored at the NPS HTLN office located at Missouri State University, Springfield, MO. A reference collection consisting of a few representative specimens of each taxon will be prepared and stored in properly labeled vials containing 70% isopropyl alcohol. One set of vials will be stored at the NPS HTLN office located at Missouri State University, Springfield, MO.

Additional sets of specimens should be maintained in the laboratory where identifications are performed for use as reference and training.

Fish

A reference collection of identified fish species is kept at the NPS HTLN office located at Missouri State University. All other fish collected during monitoring will be returned to the springs from which they were collected.

Post Season Procedures

Procedures for the end of the sample season are found in SOP #14 “Procedures and Equipment Storage after the Field Season” and are not further described here.

IV. Data Management

Overview of Database Design

All data management activities related to this protocol are described in SOP #11 “Data Management.” Microsoft Access 2003 is the primary software environment for managing spring community and habitat data. ESRI ArcInfo 9 serves as a tool for validation of spatial data residing in Access. Data products will be posted at the NPS I&M website:

<http://science.nature.nps.gov/im/monitor/protocoldb.cfm>. Metadata for springs community monitoring will be available on the NPS I&M application server:

<http://science.nature.nps.gov/im/apps/index.cfm>. QA/QC guidelines in this document are based on recommendations of Rowell *et al.* (2005) and the National Park Service Inventory & Monitoring Program (<http://science.nature.nps.gov/im/monitor>).

The general data model for spring community monitoring consists of two core sets of tables. One set manages species attribute data (species names, replicate number, count, etc.) and the other associated habitat data. Species attribute and habitat data are linked in time and space by way of standardized event and location tables. The primary table for storing species attribute data contains information about the species. Supporting tables include taxonomic information, observers, and equipment information. A locations table provides detailed location information associated with each sampling point. Look-up tables are linked to relevant tables to provide the values for pick-lists on data-entry forms, thereby reducing possible error during data entry (see Data Verification and Editing below).

Data Entry

A number of features have been designed into the database to minimize errors that occur when field data are transcribed to the database for storage and analysis. Forms are used as portals for data entry into the database. Standardized identifiers (*e.g.*, sample location and event) are selected from a list of easily interpreted codes. Species and habitat data are entered into fields linked to appropriate tables. Look-up tables contain project-specific data and prohibit entry of data into a field if a corresponding value is not included in the look-up table. Consequently, only

valid names or measures may be entered and spelling mistakes are eliminated. Species or habitat measures are selected using a pick list or by typing the beginning of the name.

Data Verification and Editing

Data verification immediately follows data entry and involves checking the accuracy of computerized records against the original source, usually paper field records. While the goal of data entry is to achieve 100% correct entries, this is rarely accomplished. To minimize transcription errors, our policy is to verify 100% of records to their original source by staff familiar with project design and field implementation. Further, 10% of records are reviewed a second time by the Project Manager and the results of that comparison reported with the data. If errors are found in the Project Manager's review, then the entire data set is verified again. Once the computerized data are verified as accurately reflecting the original field data, the paper forms are archived and the electronic version is used for all subsequent data activities.

Although data may be correctly transcribed from the original field forms, they may not be accurate or logical. For example, an invertebrate count of 3,325 instead of 325 may be illogical and almost certainly incorrect, whether or not it was properly transcribed from field forms. The process of reviewing computerized data for range and logic errors is the validation stage. Certain components of data validation are built into data entry forms (*e.g.*, range limits). Data validation can also be extended into the design and structure of the database. As much as possible, values for data-entry forms have been limited to valid entries stored in the look-up tables.

Additional data validation can be accomplished during verification, if the operator is sufficiently knowledgeable about the data. The Project Manager will validate the data after verification is complete. Validation procedures seek to identify generic errors (*e.g.*, missing, mismatched, or duplicate records) as well as errors specific to particular projects. For example, one database query detects records with a location ID from a spring and a period ID from a different spring. Another query counts the number of transects sampled per site to be sure all data were collected and entered.

During the entry, verification, and validation phases, the Project Manager is responsible for the data. The Project Manager must assure consistency between field forms and the database by noting how and why any changes were made to the data on the original field forms. In general, changes made to the field forms should not be made via erasure, but rather through marginal notes or attached explanations. Once validation is complete, the data set is turned over to the Data Manager for archiving and storage.

Spatial validation of database sample coordinates can be accomplished using ArcGIS (ESRI, Inc.). Because this is an Access-maintained database, it can be integrated directly with ArcCatalog (ArcGIS, ESRI, Inc.) as an OLE DB object. Coordinate data (UTM northing and easting) of the locations table can then be used to validate the UTM coordinate values for sample locations stored in Access against the original GPS coordinates.

Metadata Procedures

Metadata for project data are developed using ESRI ArcCatalog 9 and follow current Federal Geographic Data Committee (FGDC) standards. This requires conversion of the Access database to an ESRI personal geodatabase. Metadata are then exported into text and Extensible Markup Language (xml) format. Text-based metadata are then parsed using a USGS metadata parser to check for errors in formal FGDC metadata.

Database Versions

Changes in database structure and functionality require a versioning system. This allows for the tracking of changes over time. With proper controls and communication, versioning ensures that only the most current version is used in any analysis. Versioning of archived data sets is handled by adding a two digit number separated by a period to the file name, with the first version being numbered XXXXXX1.0. Minor changes such as revisions in forms and report content should be noted by an increase of the number to the right of the period. Major changes such as migration between Access versions or database normalization across multiple tables should be indicated by an increase in the number to the left of the period. Frequent users of the data are notified of the updates, and provided with a copy of the most recent archived version.

Database Security

Secure data archiving is essential for protecting data files from corruption. No versions of the database should be deleted under any circumstance. Monitoring databases are small and do not require significant computer drive space or resources. On the other hand, they represent primary data and are expensive to create and impossible to replace. Multiple backup copies of all program data are maintained at the HTLN offices, at the Wilson's Creek visitor center, and at the Missouri State University campus offices. Tape backups of the databases are made weekly. Each weekly full backup copy is maintained at the Wilson's Creek National Battlefield Visitor Center, Republic, MO. Once a month, one tape copy is stored offsite.

Currently, data are available for research and management applications on request, for database versions where all QA/QC has been completed and the data have been archived. Most data requests are currently met using FTP services. Portions of the monitoring data collected under this protocol will be made available for download directly from the NPS I&M Monitoring webpage. Information related to location and persistence of species determined to be threatened or endangered will not be made available for download by the general public. In addition, metadata will be available directly from the NPS I&M NR-GIS Metadata and Data metadata server (<http://science.nature.nps.gov/nrdata>). Data requests should be directed to:

Data Manager, Wilson's Creek National Battlefield, 6424 W. Farm Road 182, Republic, MO 65738-9514, (417) 732-6438

V. Analysis and Reporting

Aquatic Vegetation

The intent of this monitoring protocol is to track community composition and species occurrences through time along fixed transects within a defined sampling reach. Degradation of aquatic vegetation in the springs could signal anthropogenic disturbance, particularly in light of analogous changes in invertebrate and fish communities. Evidence suggests that the aquatic vegetation communities of the springs at OZAR have changed little during the past century (Drouet, 1933; Steyermark, 1941; Currier 1990a, b; Converse, 1994; Edwards, 2002; Lipscomb, undated).

We propose to analyze aquatic vegetation community data from the springs using several metrics. Once estimates for all parameters have been obtained for each plot, they are averaged to obtain a measure of variability (standard deviation) for a transect and then averaged across transects to obtain an estimate of variability (standard error of the mean) for the sample reach. Procedures for calculating these metrics are shown in SOP # 12 (Data Analysis). Summary indices and variables will provide information to park managers on the status of the target communities.

Individual species frequency and percent foliar cover are calculated for each transect and averaged across transects to get estimates for sample reach. Frequency is defined as the number of times a species is present in a given number of plots of a particular size (Raunkiaer, 1934). Foliar cover serves as an estimate of abundance for herbaceous species. To calculate these metrics, the cover class intervals are first converted to median values to estimate percent cover for each species. Mean percent cover is then calculated as the species percent cover for a transect ($n=3$), and for the reach ($n=18$). Plots with zero values for a species are excluded. From these basic estimates of foliar cover and frequency are generated the following metrics for each sample unit: (1) species relative cover, (2) species relative frequency and (3) species importance value.

Diversity of aquatic vegetation is calculated for each transect and averaged across the sample reach using three measures: species richness (S), Simpson's index (D), and the Shannon diversity index (H). Species richness is calculated as the total number of plant taxa recorded per transect and includes all species (native and exotic) in the estimate. Simpson's index of diversity (D) is best described as a dominance index because it weights toward the abundance of the most common species. It gives the probability of any two individuals drawn at random from an infinitely large community belonging to different species (McCune and Grace, 2002). Shannon's index accounts for both abundance and evenness of the species present, and it is more robust when all species are equally abundant or have high evenness. An additional metric, Species Distribution Evenness (E') (also known as the Shannon's Evenness Index) is a measure of the distribution of species within a community as compared to equal distribution and maximum diversity (Pielou, 1969). It is calculated using the values of the Shannon Diversity index (SOP #12).

When interested in measuring diversity in a single community it is best to use all three diversity measures to most accurately reflect diversity (Joust, 2006). At the most base level of species diversity, species richness provides a total number of distinct species sampled per unit area. Richness is insensitive to species abundance. Therefore a single individual species occurring only once in a community is treated the same as a species with thousands of individuals in the community. This measure is an indicator of species diversity but does not provide any information about the composition of species within the community. Shannon diversity index weights species by their abundance. It is an intermediate between species richness and Simpson's diversity index in its sensitivity to rare species. Therefore this diversity measure provides information on both the count of unique species and their abundance or density in the community. Simpson's diversity index goes one step further by disproportionately favoring dominant species based on species abundance and is little affected by gain or loss of rare species.

Shannon and Simpson's diversity index values are converted into effective number of species for each community (H_e and D_e , respectively; Hill, 1973) (see SOP #12). This allows for both diversity measures to be compared directly to species richness of the sites within and among sample years based on count of distinct species in the community (Joust, 2006). Dominance takes into account the species abundance and evenness of species distribution in the community. The degree of species dominance in the community is reflected by the degree that species richness of the site is greater than the effective number of species for each community based on the Simpson's and Shannon's indices ($S > H_e > D_e$) when evenness remains constant in a single community. The difference in number of species between the diversity measures reflects both how each metric considers uncommon species and how species diversity is partitioned within the community among years. If all species occurred in equal abundance in the community within and among sample years then $S = H_e = D_e$. Effective number of species for each diversity measure therefore reflects the number of species found in a similar community when all species occur in equal density.

A final metric that will be calculated is the Ratio of Exotic to Native Taxa. Since exotic species have the potential to disrupt the aquatic vegetation community in OZAR springs, this metric will serve as an important gage of their relative frequency and abundance. This ratio is calculated for each transect and averaged across the sample reach.

Aquatic Invertebrates

There is little information on the best methods for monitoring and assessing invertebrate communities of springs. Consequently, the metrics that we chose to use are based on evaluations done for stream biomonitoring studies, with a focus on metrics that are sensitive to factors such as organic pollution and inorganic toxicants rather than fine sediment. Early biomonitoring programs focused on one or two specific attributes of the communities within a system. For example, the Indicator Species concept that dominated biological evaluations for a long time (Kremen, 1992) operated on the premise that a particular perturbation could be detected by monitoring for a single sensitive species. This view of a system may be of value in detecting selected anthropogenic effects, while being unsuccessful in detecting complex, cumulative impacts (Karr, 1991). This is because an aquatic system is composed of an array of communities, each clearly defined by many different attributes. This has led to the development

of the alternative, multimetric approach that uses an array of measures or metrics, each of which reflects the health of a particular biological attribute such as community structure, community balance, or biological condition. These metrics may be grouped into several categories (Resh and Jackson, 1993) including:

- Richness measures that count the number of distinct specified taxonomic units such as families or species. Examples include taxa richness and number of Ephemeroptera, Plecoptera, and Trichoptera (EPT) taxa.
- Enumeration measures that count the total number of individuals or estimate the relative abundance of all the individuals (or a particular group) from the collection. Examples include total abundance and % dominant taxon.
- Community diversity indices, such as the Shannon Diversity Index, combine richness and relative abundance into a summary statistic.
- Similarity indices that use either qualitative or quantitative data to estimate the percentage similarity of two different communities. Examples include the Sørensen Similarity Index (qualitative) and the Quantitative Similarity Index (quantitative).
- Biotic indices that combine pre-established water-quality tolerance values for the collected taxa with their relative abundance into a summary statistic. Examples include the Hilsenhoff Biotic Index (Biotic Index) and Family Biotic Index.
- Functional feeding group ratios that divide the individuals into different groups based on the morphological structure of their mouth parts and food acquisition behavior. Examples include the ratio of scrapers to collector-filterers and the ratio of shredders to the total abundance.

Dozens of metrics have been developed for each of these various categories resulting in difficulty in attempts to compare results. In recent years, several authors have evaluated the usefulness of many of these metrics in an effort to standardize their use. Barbour *et al.* (1992) analyzed data from 10 ecoregions within the United States to assess the variation found in the rapid bioassessment metrics used by the EPA. They found that EPT richness was the best (least variable and most sensitive) measure of community structure, the Biotic Index was the best measure of community balance, and the ratio of Total Shredders/Total Abundance was the best measure of the functional feeding groups. Rabeni *et al.* (1997), using data from the major ecoregions of Missouri, determined that various metrics had less variability and were more sensitive to either organically enriched or habitat degraded impacts. Organically affected sites were best detected with Taxa Richness, EPT Richness, the Biotic Index, and the Shannon Diversity Index. Habitat degraded sites were more difficult to detect with metrics, but the most sensitive were the Shannon Diversity Index and the Percent Dominant Taxon. The assumption behind the assessment of the Percent Dominant Taxon is that a community that is heavily dominated by one species is impaired. Since dominance by a few taxa is a common and normal occurrence in these springs, this metric will not be used for this protocol (see Table 7).

Multi-metric indices are designed to look at community structure through examination of multiple components of the invertebrate community and their level of change due to disturbance. Individual metrics are chosen based on the specific and predictable response of organisms to landscape changes. Additionally, they are sensitive to a range of factors that stress biological systems and are relatively easy to measure and interpret (Karr and Chu, 1999). Scores of individual metrics are normalized into a single integrated score, reducing the influence of one metric on the overall score and making results less ambiguous for resource managers. Bonada *et al.* (2006), in a comparative analysis of recent bioassessment approaches, showed that multi-metric approaches rate among the best performers for 10 of 12 criteria they tested. However, we are unable to recommend an index-based scoring system for OZAR springs because there are insufficient data to develop such an index. After a few years of data collection from the springs, it may be possible to develop such an index.

Doisy and Rabeni (2004) suggested seven metrics as measures of community structure and balance in springs at OZAR. The procedures for calculating and scoring these metrics are included in SOP #12 (Data Analysis). They include Taxa Richness, EPT (Ephemeroptera, Plecoptera, Trichoptera) Richness, Ephemeroptera Richness, Ratio of Total Ephemeroptera Abundance/Total Abundance, Shannon's Diversity Index, Biotic Index (BI), and Ratio of Shredder Abundance/Total Abundance. We are including Percent Dominant Taxon and Percent Intolerant Taxa as additional metrics. The basis of the Percent Intolerant metric is that as the level of pollution increases, the number of pollution intolerant species should decrease. The assumption behind the assessment of Percent Dominant Taxon is that a community that is heavily dominated by one species is impaired. This metric was selected because none of the Springs at OZAR currently appear to be grossly polluted to the point where they would be totally dominated by a single taxon. The invertebrate communities of the large springs at OZAR are dominated by largely intolerant taxa making this two metrics valuable for evaluating these systems (Table 8).

These nine metrics are generally considered sufficiently sensitive to detect a variety of potential pollution problems in Ozark springs. Some of the potential disturbances that can be detected using these metrics include (after Doisy and Rabeni, 1999):

- Gross organic pollution- Hilsenhoff (1982) listed all of these as indicators of gross organic pollution.
- Thermal impairment- Resh and Jackson (1993) analyzed data from California streams and found that measures such as Taxa Richness and EPT Richness were quite accurate in detecting impairment.
- Agriculturally developed catchments- Ephemeroptera and Plecoptera have shown reductions in abundance or richness (Quinn and Hickey, 1990; Lenat and Crawford, 1994).
- Increases in acidity- Taxa richness, EPT taxa, and the Shannon Diversity Index typically decrease in response to increasing acidity (Hildrew *et al.*, 1984; MacKay and Kersey, 1985; Resh and Jackson, 1993). Mayflies are especially sensitive to low pH (Peterson *et al.*, 1985).
- Heavy metal pollution- Taxa richness and EPT richness (Winner *et al.*, 1980; Chadwick *et al.*, 1986) have been shown to decrease in response to this type of pollution. However,

further research indicates that mayflies may decrease in richness and abundance while caddisflies increase under these conditions, resulting in a static EPT. Hickey and Clements (1998) found that abundance and species richness of mayflies, EPT Richness, and Taxa Richness were strong indicators of heavy metal contamination in streams. Yuan and Norton (2003) analyzed the effects of common anthropogenic stressors on streams and reported that Ephemeroptera (mayfly) richness was the most sensitive indicator of elevated metals or ion concentrations (aluminum and conductivity). Other studies have shown that Taxa Richness, EPT Richness, and Abundance were not affected by heavy metals because of the replacement of sensitive taxa by tolerant taxa (Clements and Kiffney, 1994).

- Insecticides- Wallace *et al.* (1996) found that the EPT index detected disturbances to a stream treated with certain insecticides.
- Habitat degradation in the Ozark Highlands- Rabeni *et al.* (1997) found that Shannon's Index for high gradient riffles typically decreased with disturbance.
- Rabeni *et al.* (1999) also found an increase in the percentage of collector/filterers with increased embeddedness in Ozark Highland streams.
- Baseline community taxa lists offer a direct comparison to future community data to check for the loss or replacement of indicator species with less sensitive species. The reduction in relative abundance of indicator species is a sign of a chronic condition, whereas the replacement of indicator species by less sensitive ones may indicate an acute problem.

Table 8. The five dominant taxa and their relative mean percentages among three samples collected at OZAR springs during Fall 2003. Taxa in bold font are considered pollution intolerant.

% Composition of Dominant Taxa					
Spring	1	2	3	4	5
Alley	Lepidostoma (44.7)	Serratella (24.9)	<i>Dipheter</i> (7)	Hydrobiidae/ <i>Elimia</i> (7)	Chironomidae (5)
Big	Lepidostoma (74)	<i>Gammarus</i> (12)	<i>Gammarus</i> (6)	Oligochaeta (4)	Nematoda (3)
Blue	Lepidostoma (48.6)	Hydrobiidae/ <i>Elimia</i> (31)	Optioservus (6.4)	Hydracarina (5)	<i>Gammarus</i> (3)
Pulltite	Hydrobiidae/ <i>Elimia</i> (51)	<i>Gammarus</i> (16)	<i>Baetis</i> (8)	Serratella (6.4)	Lepidostoma (4.7)
Round	<i>Gammarus</i> (50)	Hydrobiidae/ <i>Elimia</i> (14)	Hydracarina (10)	Oligochaeta (5)	Optioservus (4.7)
Welch	Lepidostoma (38)	<i>Gammarus</i> (27)	<i>Baetis</i> (7)	Chironomidae (6)	Hydracarina (5)

Fish

Several parameters and analysis techniques have been used to detect trends in fish communities and investigate the relationships between fish communities and environmental conditions. Two common approaches are parameter estimations/metric calculations (or combining metrics to form a biological index) (Plafkin, *et al.*, 1989; Hughes and Oberdorff, 1998; Barbour *et al.*, 1999; Simon, 1999) and multivariate statistics (for examples applying to Ozark fish communities see Petersen, 1998, 2004). Using multiple analytic approaches will provide multiple lines of evidence, increasing the validity and confidence of study conclusions. A detailed summary of calculated metrics and data analyses are given in SOP #12 (Data Analysis).

Biological metrics are commonly used by scientists to compare the condition of the biological community at multiple sites (Simon, 1999) or across time. A metric is a characteristic of the biota that changes in a predictable way with increased human disturbance (Barbour *et al.*, 1999). Attributes of the fish community such as habitat and substrate preferences, trophic guilds, spawning preferences, and degree of tolerance to disturbance are measures frequently reflected in metrics making it possible to determine relationships between biological communities and environmental conditions. Metrics used for analysis and reporting are listed in SOP #12.

An extension of the metric approach is to combine multiple metrics into an Index of Biotic Integrity (IBI). This index is used as an indicator of overall stream quality, enabling investigators to compare conditions at multiple sites (Karr, 1981; Barbour *et al.*, 1999; Simon, 1999) or at a single site across time. Prior to use of fish communities as bioindicators, aquatic invertebrate communities were (and still are) used as indicators of stream quality (Hilsenhoff, 1977). Although fish communities have been largely ignored in spring-runs, the popularity of fish with the general public and stakeholders have made them the most commonly used bioindicator for investigating ecological relationships using the IBI approach in streams (Barbour *et al.*, 1999; Simon, 1999).

One of the first fish IBIs developed by Karr (1981) has been modified for use in rivers and streams in many other regions and countries (Hughes and Oberdorff, 1998; Simon, 1999). IBIs have been created for Ozark Highland streams (Hoefs, 1989; Dauwalter *et al.*, 2003; Matt Combes, Missouri Department of Conservation, written communication, 2006). Hoefs (1989) modified metrics and scoring criteria from Karr's (1981) original index for use in the Current River basin. Hoefs' IBI, however, is specific to the Current River (not its spring-runs) and has not been submitted for rigorous peer review. The existing IBI used by Missouri Department of Conservation (MDC) contains modified metrics from various IBIs to assess fish communities in the Ozarks (Matt Combes, Missouri Department of Conservation, written comm., 2006). Again, this Missouri IBI has not been rigorously peer reviewed and may not be applicable to HTLN monitoring data because this IBI is based on methods and equipment used specifically in the MDC stream assessment program. Within the next five years, a regionally based IBI for Missouri will be developed through joint efforts of MDC and the University of Missouri (Matt Combes, Missouri Department of Conservation, verbal communication, 2006). Once developed and peer reviewed, the new Missouri IBI will be evaluated for use in the spring-runs of OZAR. An IBI by Dauwalter *et al.* (2003) was developed for warm water streams in the larger Ozark region and has been peer reviewed, but may or may not be appropriate for uses in springs.

Currently, an IBI for Ozark Highland springs has not been developed. Fish community composition within spring-runs potentially could be different from that of the river due to the presence of species that require or are adapted to more stable habitat conditions (*i.e.*, temperature, pH, specific nutrient concentrations) or due to the absence of species typically found in warm water systems. Because certain metrics used in the Dauwalter IBI depend on the presence and relative abundance of specific species that may not naturally occur in springs, we will assess the applicability of this IBI for use in spring-runs.

Data Analysis

In determining the appropriate statistical approaches for this monitoring protocol, it is important to take into account the primary audience of the various reports that will result. This audience will consist of park resource managers, park superintendents, and other park staff. Park resource managers and staff may not have an in-depth background in statistical methods, and park superintendents may have limited time to devote to such reports. Additionally, protocols such as this may provide much data on many different types of variables. Thus it is important, to the extent possible, that our core data analyses and presentation methods provide a standard format for evaluation of numerous variables, are relatively straightforward to interpret, can be quickly updated whenever additional data become available, and can be used for many different types of indicators, whether univariate or multivariate. Additionally, the type and magnitude of variability or uncertainty associated with the results should be easily discernible, and a threshold for potential management action ideally will be indicated.

There are three main statistical approaches that could be employed with data from long-term monitoring projects such as this: (1) hypotheses testing, (2) parameter estimation, and (3) application of Bayesian methods. When analyzing ecological data, statisticians predominantly employ frequentist methods, and thus many resource managers are not familiar with the interpretation of Bayesian approaches. Bayesian methods are not widely used because they are often difficult to apply, and many researchers are not comfortable specifying subjective degrees of belief in their hypotheses (Utts, 1988; Hoenig and Heisey, 2001). Thus, we do not advocate a Bayesian approach as our main method of data analysis.

Most hypothesis testing approaches involve a null hypothesis of no difference or no change. The problem with such approaches is that the hypothesis under test is thus trivial (Cherry, 1998; Johnson, 1999; Anderson *et al.*, 2000, 2001). No populations or communities will be exactly the same at different times. Thus, we are not really interested in whether these are changing *per se*, but rather in the magnitude of change, and whether it represents something biologically important. Null hypothesis significance testing relies heavily on *P*-values, and results primarily in yes – no decisions (reject or fail to reject the null hypothesis). *P*-values are strongly influenced by sample size, however, and one may, with a large enough sample size, obtain a statistically ‘significant’ result that is not biologically important. Alternatively, with a small sample size, one may determine that a biologically important result is not statistically significant (Yoccoz, 1991). Thus, traditional null hypothesis testing places the emphasis on the *P*-value (which is dependent on sample size) and rejection of the null hypothesis, whereas we should be more concerned whether the data support our scientific hypotheses and are biologically significant (Kirk, 1996; Hoenig and Heisey, 2001).

Parameter estimation, including the use of confidence intervals, provides more information than hypothesis testing, is more straightforward to interpret, and easier to compute (*e.g.*, Steidl *et al.*, 1997; Gerard *et al.*, 1998; Johnson, 1999; Anderson *et al.*, 2000, 2001; Colegrave and Ruxton, 2003; Nakagawa and Foster, 2004). Parameter estimation emphasizes the magnitude of effects, and the biological significance of the results, rather than making binary decisions (Shaver, 1993; Stoeckner, 1999). There is no formal classification of error associated with parameter estimation. It is assumed that the estimate is not accurate, and the width of the confidence interval provides information on the degree of uncertainty (Simberloff, 1990). One of the primary recommendations from a workshop on environmental monitoring organized by the Ecological Society of America was that trend studies should focus on description of trends and their uncertainty, rather than hypothesis testing (Olsen *et al.*, 1997). Thus, most of our data analyses will take the form of parameter estimation with associated confidence intervals, rather than null hypothesis significance testing.

We will also employ control charts in data organization and analysis. Control charts, developed for industrial applications, indicate when a system is going ‘out of control’, by plotting through time some measure of a stochastic process with reference to its expected value (*e.g.*, Beauregard *et al.*, 1992; Gyron, 2001; Montgomery, 2001). Control charts may be univariate or multivariate, and can represent many different types of variables. Control charts have been applied to ecological data (McBean and Rovers, 1998; Manly, 2001), including fish communities (Pettersson, 1998; Anderson and Thompson, 2004) and natural resources within the I&M program (Atkinson *et al.*, 2003). Control charts contain upper and lower control limits specifying thresholds beyond which variability in the indicator reveals a biologically important change is occurring, and warns that management may need to act. Control limits can be set to any desired level.

We did not conduct a formal power analysis for this protocol for three reasons: (1) the primary purpose of conducting a prospective power analysis is to determine whether the proposed sample size is adequate. Because our sample size is limited by the number of first and second magnitude springs in the park, we would not be able to increase it regardless of the result of any power analysis. Furthermore, in many analyses sample size will equate with number of years; in this case, analyses will simply become more powerful over time. (2) Statistical power is dependent upon the hypothesis under test and the statistical test used. Over the course of this long-term monitoring program, we will be interested in many different questions and could potentially evaluate a number of different hypotheses. Thus there is no single ‘power’ relevant to the overall protocol. Estimating power at this point in the context of such a long-term, multifaceted monitoring program could be potentially misleading, as the test this power is based upon may rarely (or never) actually be employed. (3) Most of our data analyses will take the form of parameter estimation with associated confidence intervals, rather than null hypothesis significance testing. When estimating parameters, there is no associated statistical power. It is assumed that the estimate is not accurate, and the width of the confidence interval provides information on the degree of uncertainty. In general, statistical power analyses are frequently mis-used and misinterpreted in ecological contexts (Morrison, 2007), and alternative approaches

to evaluating the degree of uncertainty associated with our data will be evaluated and used when applicable.

Although our primary approach to organizing and analyzing data will consist of parameter estimation with associated confidence intervals, combined with the use of control charts, we do not entirely rule out the use of any statistical methods at this time. Because of the nature of this long-term monitoring program, other approaches (some of which may not have even been developed yet!) may be appropriate at different points in time, depending upon the needs of the resource managers and questions of interest.

At times, depending upon the question of interest to resource managers, a hypothesis testing framework may be used. Because data from studies of aquatic insects is often not normally distributed, non-parametric approaches may need to be employed. For example, if it is desirable to test for differences between transects, a Kruskal-Wallis ANOVA, Friedman's non-parametric two-way ANOVA, or Cochran's Q test could be used. Repeated measures analysis also would be appropriate for analyzing the data for trends. Of course, normality of the data will be evaluated prior to any tests, and transformations may be performed if useful prior to tests requiring normal distributions.

Multivariate analyses are another commonly used statistical method to explain variability in community data and attribute that variability to specific environmental variables or gradients (Gauch, 1982; Jongman *et al.*, 1995; Everitt and Dunn, 2001; Timm, 2002). Multivariate techniques differ from univariate or bivariate analyses in that the former techniques generate a hypothesis from the biological data rather than disproving a null hypothesis, and the effectiveness improves as the number of variables increase (Williams and Gillard, 1971). Two techniques used to analyze community data include ordination and classification (Gauch, 1982; Jongman *et al.*, 1995; Everitt and Dunn, 2001; Timm, 2002).

These approaches and others are described in SOP #12 (Data Analysis).

Reporting

Reports summarizing monitoring data collected during the year should be prepared annually. These reports should include an update on the status of the resources in addition to documenting related data management activities and data summaries. In an effort to disseminate findings in a timely manner, annual summary reports should be completed by September 30th of the year following data collection. Comprehensive trends analysis and synthesis reports should be completed every five to ten years depending on observed impacts in the watershed and how critical summary information is for setting management goals influencing spring condition. Executive summaries should be prepared for all types of reports. Refer to SOP #13 "Data Reporting" for details on types of reports and their primary audiences, report structure and style, and review procedures.

VI. Personnel Requirements and Training

Roles and Responsibilities

The project manager is the Aquatic Program Leader for the HTLN and this person bears responsibility for implementing this monitoring protocol. Because consistency is essential to implementation of the protocol, the project manager will usually lead field data collection efforts unless technicians have several years of experience collecting the data related to this protocol as determined by the project manager. The project manager will oversee all laboratory work including all QA/QC requirements. The data management aspect of the monitoring effort is the shared responsibility of the project manager and the data manager. Typically, the project manager is responsible for data collection, data entry, data verification and validation, data summary, analysis, and reporting. The data manager is responsible for data archiving, data security, dissemination, and database design. The data manager, in collaboration with the project manager, also develops data entry forms and other database features as part of quality assurance and automates report generation. The data manager is ultimately responsible to ensure that adequate QA/QC procedures are built into the database management system and appropriate data handling procedures followed. The Program Fisheries Biologist and Aquatic Ecologists will be responsible for field collection and laboratory processing, equipment maintenance, purchasing of supplies, and sample storage. For invertebrate samples, at least one technician with taxonomic experience will be responsible for identification to the genus level. For fish monitoring, the fisheries biologist (or a technician with skills in taxonomic identification) will be responsible for identifying fish to the species level in the field and the laboratory.

Qualifications and Training

Training is an essential component for collection of credible data. Training for consistency and accuracy should be emphasized for both the field and laboratory aspects of the protocol. SOP #2 “Training for Field Sampling and Laboratory Processing” describes the training requirements for new technicians. The project manager should oversee this training and ensure that each technician is adequately prepared to collect data. Taxonomic identifications for invertebrates and fish may be performed by a technician with several years of experience, but initial identifications should be checked by expert taxonomists.

VII. Operational Requirements

Annual Workload and Field Schedule

Samples will be taken once a year during the summer index period of 15 July-31 August. Sampling should begin at relatively the same time each year and samples should be collected within the shortest time frame possible (4 weeks) to minimize the effects of seasonal change. For aquatic vegetation, invertebrate, and habitat sampling, a minimum of two people will be required, but three people make the process much more efficient. For fish monitoring a minimum crew of 4-5 people will be needed depending on the gear used. Because of travel considerations, only one site can be sampled per day under normal circumstances.

Laboratory processing time per benthic invertebrate sample, including sorting, identification, counting, and entry into the database, will require approximately 6 hours per sample, or a total of 25 laboratory days to complete all samples. These tasks primarily will be accomplished by the Program Aquatic Ecologist stationed at OZAR although our Program staff may assist with this effort. Processing of all fish samples preserved for laboratory identification will take approximately 5 laboratory days.

Facility and Equipment Requirements

Field and lab equipment listed in SOP #1 “Preparation for Field Sampling and Laboratory Processing” are only for one sampling crew. Beyond normal office and equipment storage space, facility needs include access to a wet laboratory. Additional equipment requirements include access to a canoe or motorboat.

Startup Costs and Budget Considerations

Estimated costs for conducting annual monitoring at OZAR springs are shown in Table 9. Personnel expenses for fieldwork are based on a crew of two to three to complete vegetation and invertebrate work and a crew of four to five for fish (a professional aquatic ecologist or fisheries biologist to coordinate and oversee the fieldwork and two to three seasonal biological science technicians to assist in field data collection). Assistance with field work from other agencies and park personnel is welcomed to the extent it is available. Field costs may vary somewhat from year to year depending on the skill level and size of crew. Data management personnel expenses include staff time of Seasonal Biotechnicians, the Aquatic Ecologist, and Data Manager.

Table 9. Estimated annual costs for conducting annual monitoring at the large spring ecosystems at OZAR.

Project Area	Estimated Costs
Staff Salary	\$79,643.80
Admin support to WICR	\$1,176.00
Overhead to MSU	\$500.00
Field work travel	\$2,888.00
Computer hardware and software	\$760.00
Vehicle lease	\$1,824.00
Field / office equipment	\$1,368.00
Supplies	\$912.00
Lab fees	\$2,000.00
TOTAL	\$91,071.80

VIII. Procedures for Protocol Revision

Revisions to both the Protocol Narrative and to specific Standard Operating Procedures (SOPs) are to be expected. Careful documentation of changes to the protocol and a library of previous protocol versions are essential for maintaining consistency in data collection and for appropriate treatment of the data during data summary and analysis. The Microsoft Access® database for each monitoring component contains a field that identifies the protocol version used when the data were collected.

The rationale for dividing a sampling protocol into a Protocol Narrative with supporting SOPs is based on the following:

- The Protocol Narrative is a general overview of the protocol that gives the history and justification for doing the work and an overview of the sampling methods, but that does not provide all of the methodological details. The Protocol Narrative will only be revised if major changes are made to the protocol.
- SOPs, in contrast, are very specific step-by-step instructions for performing a given task. They are expected to be revised more frequently than the protocol narrative.
- When an SOP is revised it usually is not necessary to revise the Protocol Narrative to reflect the specific changes made to the SOP.
- All versions of the Protocol Narrative and SOPs will be archived in a Protocol Library.

The steps for changing the protocol (either the Protocol Narrative or the SOPs) are outlined in SOP #15 (Revising the Protocol). Each SOP contains a Revision History Log that should be filled out each time a SOP is revised to explain why the change was made, and to assign a new Version Number to the revised SOP. The new version of the SOP or Protocol Narrative should then be archived in the HTLN Protocol Library under the appropriate folder.

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X. Standard Operating Procedures (SOPs)

Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri. Heartland I&M Network and Prairie Cluster Prototype Monitoring Program

SOP 1: Preparation for Field Sampling and Laboratory Processing

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP provides information to prepare for the field season, including lists required of field and laboratory equipment. It also provides information on keeping records of staff time spent on sampling trips, checking water levels at the parks, and obtaining collecting permits. A list of required data sheets with a brief explanation of their purpose is provided.

I. General Preparations

- A. Prior to the field season all crew members should review the entire protocol, including SOPs. The following list includes key points to consider in preparing for the upcoming field season.
- B. The team leader of each sampling crew must prepare a field notebook for the survey year. The notebook should contain entries for observer names, field hours, and unique happenings that may influence how the data is reported. Information included in trip reports is based on what is recorded in field notebooks, so it is imperative that they are clearly organized for ease of field note entry. Notebook entries should be recorded daily to ensure accuracy. An example of a notebook log is shown in Fig. 1.

Date	Travel time (hours)	Field time (hours)	Non-project time (hours)	Lunch (hours)
30 Sep 2006	3	8	0	0.5
1 Oct 2006	0	8	1	0.5
2 Oct 2006	3	8	0	0.5
Notes: J. Smith and H. Simpson traveled to OZAR to conduct spring monitoring at Welch Spring. Field assistance was provided by B. Jones and K. Adams. Returned to headquarters. Non-project time included discussing other projects with Park staff.				

Figure 1. Example of a field note book for recording scheduling, travel and field time, and personnel information.

- C. Monitoring will be conducted during the period 15 July-31 August. Inclement weather and personnel workloads will preclude the scheduling of sampling events to specific annual dates. Sampling dates should be scheduled and logistics organized prior to the start of each field season. Monitoring efforts will require a three person crew (two people to sample and one to record data and provide general assistance).
- D. An equipment list will be compiled and equipment organized and made ready for the field season several weeks prior to the first sampling tour to make sure that all supplies are available and all equipment is in working condition. This allows time to make required repairs and order replacement equipment. Inspect the sample nets and wash bucket to ensure there are no tears in the nets or screen. Ensure water quality meters can be calibrated and are properly functioning as described in SOP #4. Equipment and supplies for field and laboratory use is shown in Table 1.

Table 1. Field equipment and supplies.

Number Required	Description
General	
Per Person	Waders and boots Life jackets Rain Gear Polarized sun glasses First aid kit Insect repellent & sunscreen
1	Two-way radio & charger
1	Directions to sample sites, sample site maps, list of GPS coordinates
1	Clip board
5	Pencils
Complete set	Data sheets and bottle labels printed on waterproof paper
1	GPS unit
1	Digital camera
1	Field log book
1	Flagging tape, roll
1	Tool box with miscellaneous tools
Habitat Assessment & Water Quality	
1	1.0 m ² PVC sample cell frame
2	Laminated plastic substrate sheet with Wentworth scale codes

Table 1. Continued.

Number Required	Description
1	Tape measure
1	Range finder
1	Velocity meter (Marsh-McBirney or USGS pygmy) with copies of operations manuals
1	Datasonde for water quality
1	Top-setting wading rod, 1.5 m
Aquatic Vegetation Sampling	
1	Water scope or view bucket
1	1.0 m ² PVC sample cell frame
1	10x magnifying hand lens
1	Plant press with blotter sheets and newspaper for pressing reference plants
6	1-quart storage bottles for temporary storage of reference plants
Invertebrate Sampling	
1	Sample net: EPA style kick net (i.e., Slack Surber) with 500µm mesh net fitted with 0.25 m ² sample frame
1	Nylon scrub brush
1	Hand rake or the "Garden claw"
5	Labeled sample bottles (plastic 1 gallon wide mouth bottles)
Varies per # of samples	Preservative-99% isopropyl alcohol or 10% buffered formalin and 1-L plastic Nalgene bottles for carrying, bring extra quantities to ensure enough is available to preserve all samples. Recommend minimum of 3 gallons of alcohol per spring. If alcohol is used, label container "flammable"
2	Forceps
2	Polypropylene wash bottles
1	U.S. Standard sieve, 500-µm mesh size
2	5-gallon white buckets
1	Benthic sample wash bucket with 500-µm mesh size
1	Shallow white pan
Invertebrate Sample Processing	
1	U.S. Standard sieve 500-µm mesh sieve
1	White plastic pan for elutriating samples
1	Petri dish or similar sorting container
3	Multi-well Petri dishes for separating sorted insects
1	Dissecting stereomicroscope
Several	Forceps (fine point)
Several	Clean plastic bottles with labels inside and out for storage of processed samples
As needed	Preservative (75% isopropyl alcohol)
Several	Storage vials (4 dram or larger) with labels for reference collection specimens
Fish Sampling	
1-2	Backpack electrofishing unit (backpack, cathode, anode, batteries, battery charger)
1	Tow barge electrofishing unit (barge, generator, pulse box, cathode, anode, gas can with gas, oil)
1	Electrofishing boat (generator, pulse box & cradle, boom & cords)
1	Boat motor, 5 hp, gas can with gas and oil
2	Holding pens
3	Aquarium nets
3	Aerators, battery powered
4	Dipnets
1	Dipnet, long-handled (for electrofishing boat)
5 sets	Electrofishing gloves
Varies per # of samples	10% buffered formalin and 1-L plastic Nalgene bottles for carrying, bring extra quantities to ensure enough is available to preserve all samples.
5	5-gallon white buckets
12	1-gallon jugs
1	Shallow white pan
1	Measuring board
1	Hanging scales
1	Weighing balance, large with rechargeable batteries
1	Weighing balance, small with rechargeable batteries
1	Battery charger for rechargeable batteries
12	Extra batteries for water quality meters (size AA)

Table 1. Continued.

Number Required	Description
1	Volt/Ohm meter
1	Fish collecting permit for State of Missouri

II. Field and Laboratory Forms

Print copies of field sheets and labels on waterproof paper. Data should be recorded with waterproof ink or #2 lead pencil. Example data sheets are provided as attachments to their corresponding SOPs.

Table 2. List of field forms and data sheets required for spring community monitoring for the sampling season.

TITLE	PURPOSE	NUMBER OF COPIES NEEDED PER REACH
In-stream habitat assessment form	Recording physical data from spring-runs	1
Sample cell substrate sheet	Recording substrate sizes from within invertebrate sample net frame	1
Aquatic vegetation assessment form	Recording the percent occurrence of aquatic vegetation from sample cells	6
Unknown species form	Recording unknown species of aquatic vegetation	1
Fish collection field form 1	Recording site and weather	1
Fish collection field form 2	Recording fish collection data	As needed
Riparian habitat field form	Recording riparian habitat data	1
Bank habitat form	Recording bank habitat data	1
Discharge form	Recording stream discharge	1
Aquatic invertebrate identification & enumeration sheets	Recording names of identified taxa and their densities	As needed

IV. Collecting Permits

Collecting permits for sampling fish in Missouri must be obtained from the Missouri Department of Conservation. The state of Missouri does not currently require a collecting permit for stream invertebrates exclusive of crayfish. Contact information for applying for permits is:

Missouri Department of Conservation, P.O. Box 180, Jefferson City, MO 65102-0180, (573) 751-4115

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 2: Training for Field Sampling and Laboratory Processing

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP explains the training procedures for using field equipment properly, collecting spring community and habitat data, and processing invertebrates in the laboratory. This training ensures a high level of consistency among samplers and processors. Prior to training, all personnel should review the general protocol and each SOP. Someone familiar with the protocol and experience with the sampling and processing procedures should supervise the training.

I. Field Sampling and Habitat Data Collection

Procedure:

- A. All field personnel will review the plant identification guide included in SOP #5 and be familiar with common species of aquatic vegetation occurring in the springs prior to monitoring. Monitoring techniques described in SOP #5 (Aquatic Vegetation Sampling) also will be reviewed prior to sampling, and team members should practice assigning Daubenmire cover classes to vegetation in practice plots. Find a nearby stream to practice collecting benthic samples and habitat data. Follow the procedures outlined in SOP #6 Aquatic Invertebrate Sampling, and collect several practice samples for processing in the lab. Ensure that each person is comfortable with all aspects of the sampling routine.
- B. All personnel also will review photographs, descriptions, and a reference collection of common fish species occurring in the springs and be familiar with collecting methods described in SOP #8 (Fish Community Sampling).
- C. All personnel will review SOP #9 (Physical Habitat Measurements) prior to monitoring in relation to the above community monitoring.
- D. After collecting the samples, practice taking spring discharge and using the water quality instruments. Practice discharge measurements following procedures described in SOP

#10 (Measuring Spring Discharge). Each person should be familiar with the instruments and be able to calibrate each instrument as outlined in SOP #4 (Documenting CORE 5 Water Quality Variables).

II. Laboratory Processing of Invertebrates

Procedure:

- A. Follow steps outlined in SOP #7 (Laboratory Processing and Identification of Invertebrates) using practice samples collected through field sampling training.
- B. Material left over after sorting is completed should be checked by someone skilled in processing benthic samples to determine recovery efficiency. The number of invertebrates recovered should be expressed as a percentage of the total number of invertebrates. The standard for recovery is 95%.

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 3: Reach Selection

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP describes procedures for establishing sample reaches and transects for monitoring aquatic vegetation, habitat, invertebrates, and fish in spring-runs.

I. Establishing Sampling Transects within the Spring-run

Procedure:

- A. The sample unit is the sample reach located downstream of the source. At OZAR, the source of large springs is characterized by a deep pool area, while the spring-run is the shallower (and often narrower) area of the spring with higher velocity. Sampling will be restricted to a sample reach within the spring-run.
- B. Locate the reach of the spring-run to be sampled:
 1. The upstream boundary of the sampling reach will be located at the first wadeable area downstream of the spring source. The reach will begin as close to the spring source as possible with the starting point based on accessibility and crew safety.
 2. Reach lengths were established using a weighting factor that is based on the minimum average width of 15 m (width of Pulltite Spring and Welch Spring) and a minimum reach length of 150 m (the minimum reach recommended by NAWQA; Moulton *et al.*, 2002).
 3. The weighting factor for each spring was determined by dividing its average width by 15.
 4. The weighting factor was multiplied by 150 m to determine sampling reach length (Table 1).

5. The lower boundary is located downstream of the upper boundary at the distance specified in Table 1.

Table 1. Weighting factors and length of sampling reaches for large springs at OZAR.

Spring	Total Length (meters)	Average Width (meters)	Weight Factor (WF)	150 m X WF	Transect Interval (m)
Alley	800	19	$19/15 \approx 1.3$	190 m	19
Big	600	46	$46/15 \approx 3.1$	460 m	46
Blue	250	16	$16/15 \approx 1.1$	160 m	16
Pulltite	250	15	$15/15 \approx 1.0$	150 m	15
Round	300	24	$24/15 \approx 1.6$	240 m	24
Welch	36	15	$15/15 \approx 1.0$	36 m	18

6. The established reach will become a permanent sampling site barring dramatic alterations in spring morphology that would require re-establishing the sampling reach. Locations of the upper and lower reach boundaries are recorded using GPS. For instructions on using the GPS, refer to the GPS SOP located at: http://www1.nature.nps.gov/im/units/htln/data_management.cfm
- C. Establish 11 transects for each spring-run using the appropriate spacing increments shown in Table 1. The transect spacing interval is determined by dividing the reach length by 10 (Fig. 1). Due to the short length of the spring-run at Welch Spring, only 3 transects will be used. The upstream-most transect and the downstream-most transects fall on the upper and lower reach boundaries, respectively.
 - D. Aquatic vegetation, invertebrate, and physical habitat data will be collected at points along each transect (see Fig. 1). Fish will be collected within the entire reach.
 - E. When establishing transects, avoid walking in the spring-run and use a range finder to measure distances over 20 m.
 - F. Site names will be the actual names of the springs. They are: Alley Spring, Big Spring, Blue Spring, Pulltite Spring, Round Spring, and Welch Spring.

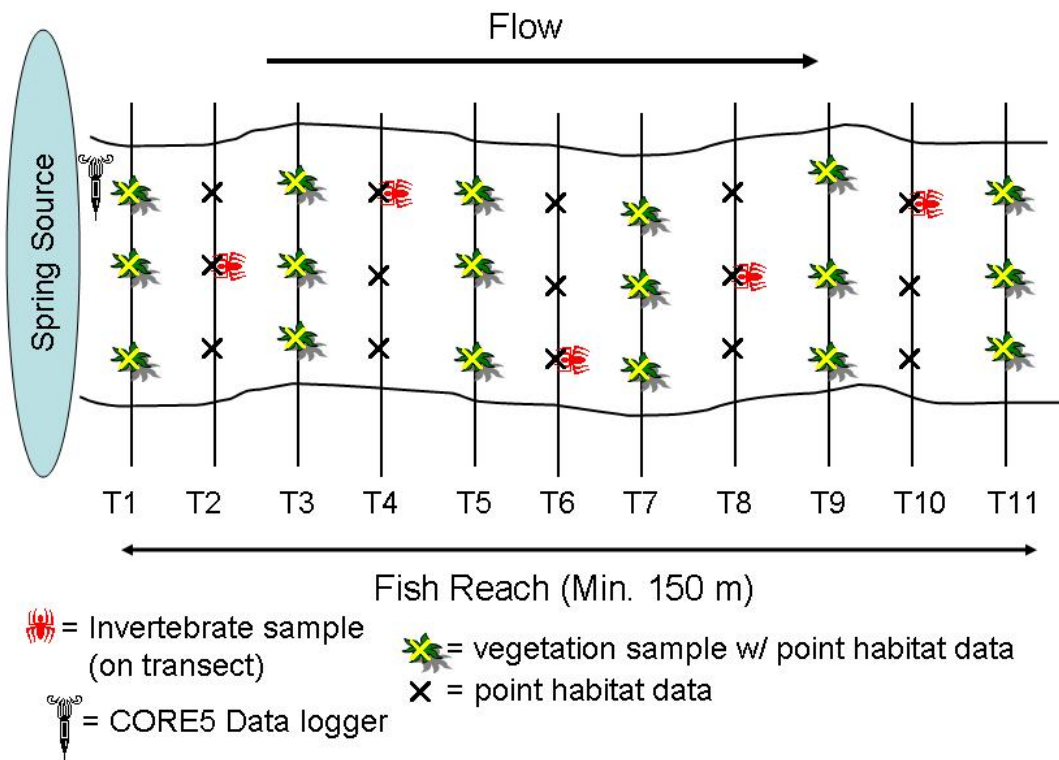


Figure 1. Diagram of transect layout in a spring-run showing locations of vegetation assessment plots, invertebrate samples, fish collection, and physical habitat data.

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 4: Documenting CORE 5 Water Quality Variables

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP addresses the equipment and methods required to measure CORE 5 water quality variables (temperature, dissolved oxygen, specific conductance, pH, and turbidity) in association with all aquatic monitoring in Network parks. Detailed guidance for measuring CORE 5 parameters, including training, calibration, QA/QC, data archiving, meter specifications, field measurements, and trouble shooting, can be found in the Documenting CORE 5 Water Quality Variables SOP located at: <http://www1.nature.nps.gov/im/units/htln/aquabugs.cfm>. This SOP is based on guidance from NPS Water Resources Division (2003), and Wagner *et al.* (2006).

I. Unattended CORE 5 measurements

Unattended CORE 5 data will be recorded using dataloggers or sondes. CORE 5 water quality parameters measured with small intervals (*i.e.*, minutes to hours) between repeated measurements are considered continuous because few if any significant water quality changes are likely to go unrecorded. When the goal is to characterize events of short duration, but such events are difficult to capture manually using discrete measurements (see above), continuous monitoring is appropriate. Continuous monitoring of core parameters helps address questions concerning daily or seasonal variability, or short-term changes (*e.g.*, precipitation related events) that might not be apparent or may prevent accurate understanding of long-term data. Continuous monitoring also provides the most comprehensive temporal data set to assess variability through time. Such information is necessary to document correlations, possible cause and effect relationships, and differentiate natural variability from anthropogenic induced change to an aquatic system. Because loggers give more comprehensive data and are easily deployed in these spring systems, and discrete samples would duplicate effort, we will use loggers exclusively to collect water quality data. Data logging of CORE 5 parameters will be conducted for a minimum of 48 hours at each spring during the sampling period.

II. Analysis and Reporting

CORE 5 data will be analyzed using summary statistics (mean, median, range, standard deviation, standard error) for each site and date. This information will be presented in summary and synthesis reports to support invertebrate collection data.

III. NPS STORET

Collected water quality data that has been successfully subjected to QA/QC will be exported to NPS STORET (see SOP #11 Data Management). Only summary data for a site and collection period in addition to pertinent metadata will be submitted. Instructions for preparing and exporting water quality data to this archival facility can be found at the following website:

<http://nrdata.nps.gov/Programs/Water/NPStoret/>

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 5: Aquatic Vegetation Sampling

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP describes field procedures for assessing aquatic vegetation in the spring-runs.

I. Assessing the Aquatic Vegetation Community

Procedures:

- A. Prior to assessing the aquatic vegetation community and taking habitat measurements, always complete data sheet information for spring name, date and time of survey, and initials of personnel who collect the samples.
- B. Aquatic vegetation will be assessed in three equally-spaced 1 m² sample cells located on transects 1, 3, 5, 7, 9 and 11 (see Fig. 1 in SOP #3, Reach Selection).
- C. Place sampling frame securely on substrate centered on the transect (Fig. 1). The observer should be positioned downstream of the sampling frame. Use a view bucket to assess the coverage of aquatic vegetation within the frame.



Figure 1. Placement of the sampling frame on the stream bottom and using a view bucket to assess coverage. A second team member records data.

1. Record composition of vegetation using a modified Daubenmire scale (Daubenmire 1959, Table 1) within each sampling cell (Fig. 2). The sampling frame is divided into four equal quarters that the observer estimates percentage composition within the entire sampling area. Combined cover may exceed 100% due to overlapping vegetation of different species.

Table 1. Modified Daubenmire cover value scale.

Cover Class Codes	Range of Cover (%)
7	95-100
6	75-95
5	50-75
4	25-50
3	5-25
2	1-5
1	0-0.99

2. Only rooted vegetation and floating species (*e.g.*, *Lemna*) are counted as occurring in a given plot. Vegetation rooted outside of the plot, but with leaves floating over the plot is not counted.

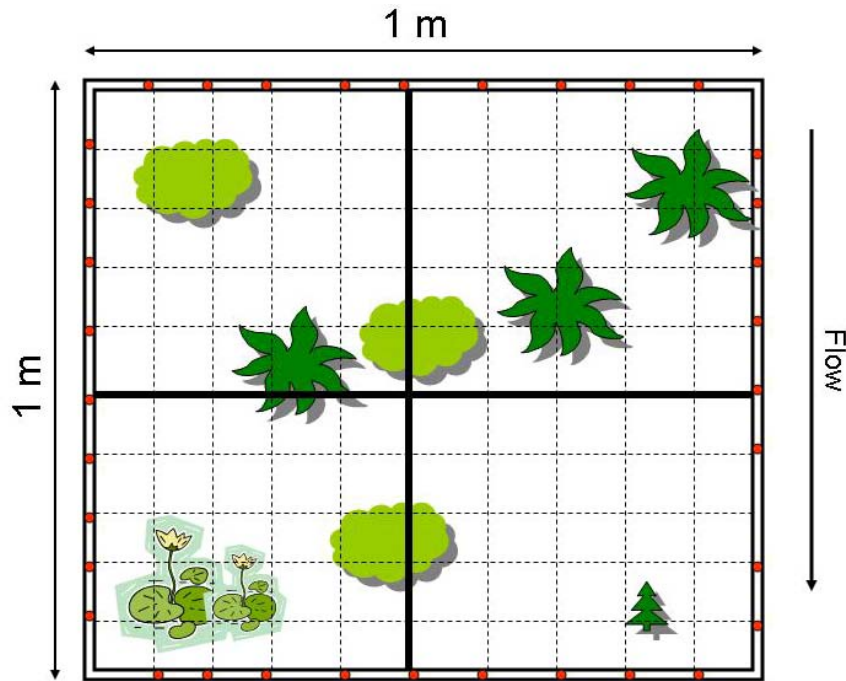


Figure 2. Sampling frame showing hypothetical distribution of aquatic vegetation in sampling area. Dashed lines are only intended to show the approximate visual grid the observer uses to estimate percentage composition. Each subsection equals 1% of the total sample cell area.

3. Mosses and liverworts are classified into those two broad groups, respectively. Algae are classified as macroalgae (*Nitella*), filamentous red algae (*Batrachospermum*), filamentous green algae (*Draparnaldia*, *Chaetophora*), other filamentous algae, and globular algae (*Nostoc*). All other aquatic plants are identified to species.
 4. When the field crew is unable to make a specific determination of an aquatic plant found in a survey plot, the plant should be properly recorded on the Unknown Species Sheet, and a specimen should be collected for later examination in the lab or for sending to an aquatic plant expert.
- D. A list of aquatic vegetation known to or suspected of occurring in large springs at OZAR is presented in Table 2. Suspected occurrence is based on records of those species in the regional proximity of OZAR.
 - E. An identification guide to the aquatic vegetation of the large springs at OZAR is presented in Table 3. However, this guide is not intended to be a definitive identification source.
 - F. Preferred taxonomic keys for identifying aquatic plants in the field or lab include Correll and Correll (1975), Godfrey and Wooten (1979, 1981), Redfearn (1981), and Yatskievych (1999, 2006).

III. Collecting and Preserving Aquatic Plants

- A. A representative reference collection of aquatic plant species occurring in the various springs will be collected, properly preserved, and stored in the Missouri State University Herbarium, Springfield, MO.
- B. Plant specimens should be placed in a plant press with an identification label indicating state (Missouri), county, spring name, GPS coordinates, date, collector, and general habitat condition. After the first day plants have been pressed, the blotters should be changed to ensure maximum drying potential. When the plants have dried sufficiently, they should be mounted on white card stock, about 11 x 16 inches (~279 x 406 mm), labeled, and stored in herbarium cabinets.
- C. Alternatively, aquatic plants can be stored in 4% formalin for short term storage until positive identification can be made, or they can be placed in zip-lock type storage bags along with a small amount of water, and placed in a cooler or refrigerator. Plants stored in this manner ideally should be examined within 24 hours of collection.

AQUATIC VEGETATION ASSESSMENT FORM

Daubenmire Scale Range of Cover*

Spring Name: _____ Transect No. 11 9 7 5 3 1

Crew Members: _____

Date: _____

LOCATION	L	M	R
Floating			
Lemna minor			
Lemna minuta			
Lemna trisulca			

Mosses, Liverworts, Algae

Mosses			
Liverworts			
Batrachospermum			
Chaetophora			
Draparnaldia			
Nostoc			
Filamentous green algae			
Nitella acuminata			

Rooted-Aquatic Zone

Agrostis stolonifera			
Callitriche heterophylla			
Cardamine bulbosa			
Ceratophyllum demersum			
Chara			
Eleocharis acicularis			
Elodea nuttallii			
Glyceria striata			
Heteranthera dubia			
Justicia americana			
Ludwigia palustris			
Ludwigia repens			
Mentha piperita			
Myriophyllum heterophyllum			
Myriophyllum pinnatum			
Myriophyllum spicatum			
Najas guadalupensis			
Nasturtium officinale			

LOCATION	L	M	R
Nuphar lutea			
Plantago cordata			
Poa annua			
Polygonum hydropiperoides			
Potamogeton foliosus			
Potamogeton illinoensis			
Potamogeton nodosus			
Ranunculus longirostris			
Rumex obtusifolius			
Sagittaria latifolia			
Sparganium americanum			
Veronica anagallis-aquatica			
Zannichellia palustris			
UNKNOWN			

Wetland & Marginal Rooted (if in spring-run)

Alisma subcordatum			
Cyperus spp.			
Carex spp.			
Impatiens capensis			
Juncus acuminatus			
Juncus dudleyi			
Juncus macer			
Lindernia dubia			
Penthorum sedoides			
Physostegia virginiana			
Pilea pumila			
Polygonum spp.			
Samolus parviflorus			
Scirpus spp.			

BARE SUBSTRATE

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*1= 0-0.99. 2= 1-5. 3= 5-25. 4= 25-50. 5= 50-75. 6= 75-95. 7= 95-100

Notes:

AQUATIC VEGETATION Unknown Species Sheet

[illegible]

Table 2. List of Aquatic Vegetation Known to or Suspected of Occurring in Large Springs at Ozark National Scenic Riverways, Missouri^{1, 2}. A black square (■) indicates known occurrences.

NOMENCLATURE		SPRINGS						HABITAT FORM ³			
Taxon	Common Name	Alley Spring	Big Spring	Blue Spring	Pulltite Spring	Round Spring	Welch Spring	Floating Not-rooted	Floating Rooted	Rooted Submerged	Rooted Emergent
Algae											
Characeae											
<i>Nitella acuminata</i>	Nitella		■							■	
Mosses, Liverworts											
Amblystegiaceae											
<i>Leptodictyum fluviatile</i>	Moss		■		■	■	■			■	■
<i>Leptodictyum laxirete</i>	Moss	■								■	■
<i>Leptodictyum noterophilum</i>	Moss			■						■	■
<i>Leptodictyum riparium</i>	Moss	■		■	■	■	■			■	■
<i>Leptodictyum tenax</i>	Moss	■	■	■						■	■
Brachytheciaceae											
<i>Brachythecium rivulare</i>	Moss	■	■	■	■		■			■	■
<i>Rhynchostegium riparoides</i>	Moss		■							■	■
Fontinalaceae											

<i>Fissidens fontanus</i>	Moss	■	■	■						■	■
<i>Fissidens grandifons</i>	Moss	■	■	■	■	■				■	■
<i>Fissidens duriaei</i>	Moss	■	■	■	■	■	■			■	■
<i>Fissidens filiformis</i>	Moss			■						■	■
<i>Fissidens minutulus</i>	Moss	■	■	■						■	■
<i>Fissidens missourica</i>	Moss			■						■	■
Pottiaceae											
<i>Hyophila involuta</i>	Moss	■	■		■		■			■	■
Thuidiaceae											
<i>Thuidium pygmaeum</i>	Moss		■		■					■	■
Conocephalaceae											
<i>Conocephalum conicum</i>	Liverwort		■		■	■				■	■
Marchantiaceae											
<i>Dumortiera hirsute</i>	Liverwort		■	■	■					■	■
<i>Marchantia polymorpha</i>	Liverwort	■	■			■	■			■	■
Porellaceae											
<i>Porella pinnata</i>	Liverwort	■	■				■			■	■
Vascular Plants											
Acanthaceae											
<i>Justicia americana</i>	American water					■				■	■

	willow										
Alismataceae											
<i>Sagittaria latifolia</i>	Broad-leaved arrowhead					■			■	■	
Apiaceae											
<i>Hydrocotyle umbellata</i>	Water pennywort					■				■	■
Brassicaceae											
<i>Cardamine bulbosa</i>	Spring-cress	■	■	■	■	■				■	■
<i>Nasturtium officinale</i> ¹	Water-cress	■	■	■	■	■	■			■	■
Callitrichaceae											
<i>Callitriche heterophylla</i>	Water starwort	■	■	■	■	■	■			■	■
Ceratophyllaceae											
<i>Ceratophyllum demersum</i>	Coon's tail or hornwort	■	■							■	
Cyperaceae											
<i>Eleocharis acicularis</i>	Slender spike rush			■							■
Haloragidaceae											
<i>Myriophyllum heterophyllum</i>	Twoleaf water milfoil	■	■		■	■	■			■	■
<i>Myriophyllum spicatum</i> ¹	Eurasian watermilfoil									■	■
Hydrocharitaceae											
<i>Elodea</i>	Canadian									■	

<i>canadensis</i>	waterweed										
<i>Elodea nuttallii</i>	Western waterweed	■	■	■	■	■	■			■	
Lamiaceae											
<i>Mentha piperita</i> ¹	Peppermint	■					■			■	■
<i>Physostegia virginiana</i>	False dragon-head			■						■	■
Lemnaceae											
<i>Lemna minor</i>	Duckweed	■						■			
<i>Lemna trisulca</i>	Star duckweed	■	■	■	■			■			
Onagraceae											
<i>Ludwigia palustris</i>	Marsh seedbox		■			■				■	■
<i>Ludwigia repens</i>	Creeping primrose-willow					■				■	■
Plantaginaceae											
<i>Veronica anagallis-aquatica</i>	Water speedwell	■	■	■	■	■	■			■	■
Poaceae											
<i>Agrostis stolonifera</i> ¹	Creeping bentgrass			■						■	■
<i>Glyceria striata</i>	Fowl meadow grass, Manna grass			■		■				■	■

<i>Poa annua</i> ¹	Blue grass	■	■			■	■			■	■
Pontederiaceae											
<i>Heteranthera dubia</i>	Water star-grass		■	■						■	
Potamogetonaceae											
<i>Potamogeton foliosus</i>	Narrow-leaved pondweed	■	■		■	■			■	■	
<i>Potamogeton illinoensis</i>	Illinois pondweed	■	■	■		■	■			■	
Ranunculaceae											
<i>Ranunculus longirostris</i>	Longbeak buttercup	■	■	■	■		■			■	
Sparganiaceae											
<i>Sparganium americanum</i>	Bur-reed	■	■	■	■	■	■			■	■
Zannichelliaceae											
<i>Zannichellia palustris</i>	Horned pondweed		■	■		■	■			■	
Wetland Plants ²											
Alismataceae											
<i>Alisma subcordatum</i>	American water plantain		■				■				■
Balsaminaceae											
<i>Impatiens capensis</i>	Jewelweed	■				■					■
Crassulaceae											
<i>Penthorum</i>	Ditch			■							■

<i>sedoides</i>	stonecrop										
Juncaceae											
<i>Juncus acuminatus</i>	Tapertip rush					■	■				■
<i>Juncus dudleyi</i>	Dudley's Rush						■				■
<i>Juncus tenuis</i>	Slender rush						■				■
Polygonaceae											
<i>Polygonum</i> spp.	Smartweeds	■	■	■	■	■	■				■
<i>Polygonum hydropiperoides</i>	Swamp smartweed		■								■
Primulaceae											
<i>Samolus parviflorus</i>	Brook pimpernel					■					■
Scrophulariaceae											
<i>Lindernia dubia</i>	False pimpernel						■				■
Urticaceae											
<i>Pilea pumila</i>	Canadian clearweed	■									■

¹ Non-native species known or possibly occurring in OZAR spring-runs. Some researchers consider *Nasturtium officinale* to be a naturalized species introduced from Europe, but this has not been conclusively demonstrated. *Myriophyllum spicatum* has not yet been reported from the springs proper, but it is quite possible that it may eventually be introduced into some of the springs.

² Wetland species that may be at waters edge.

³ Most mosses and bryophytes do not have true roots, but have rhizoids (thread-like structures) used for attachment to the substrate.

Table 3. Identification Guide to Aquatic Vegetation Occurring at Large Springs at Ozark National Scenic Riverways

Identification Guide to Aquatic Vegetation at Large Springs at OZAR*

Filamentous Algae

Globular – A ball of algal growth that is usually gelatinous or jelly-like. These are usually green algae (such as *Chaetophora*) or blue-green algae (such as *Nostoc*).

Short to medium filaments – Filamentous algae form hairlike strands that are branched, to give a bushy appearance, or unbranched. Some filaments are quite tenacious and difficult to break while others break apart easily.

Green – Filamentous green algae are quite diverse. Green filaments that are coarse to the touch and difficult to break are usually *Cladophora* or *Rhizoclonium*. *Vaucheria* (actually classified as a yellow-green alga) forms a felt-like mat of filaments that may or may not be attached to the substrate. Attached filaments that are slippery or gelatinous to the touch may be *Ulothrix*, *Draparnaldia*, or *Stigeoclonium*.

Brown – Short filaments that are reddish brown and break apart when touched are likely diatoms and are usually found in slow flows. Filamentous green algae that are senescent (dead or dying) usually appear brown because of the breakdown of their pigments. Brown color may also indicate a filamentous green alga that is covered by epiphytic diatoms (diatoms that grow on other plants: ‘epi’ – on, ‘phyte’ – plant).

Black – Black filaments indicate blue green algae. Common filamentous blue-greens include *Lyngbya* and *Oscillatoria*.

Reddish to olive – Blue-green algae can appear reddish or olive (especially in the sunlight) due to the presence of phycoerythrin or phycocyanin pigments. Freshwater red algae also contain these pigments and are particularly found in clean, clear, cool water. *Batrachospermum* is a particularly attractive red alga that is slippery or gelatinous to the touch and has a characteristic beaded appearance (like a strand of beads) at close inspection.

Long filaments – Most algae described above can develop filaments or tufts longer than three inches. Large areas of long, green filaments almost always indicate nutrient enrichment (either prolonged high levels or frequent pulses of nutrients). In extreme cases, the filaments can cover the entire stream bottom.

Free floating – Many filamentous algae can form free-floating, unattached, mats or cloud-like growths in the water column in slow-flow areas. These are almost always green or yellowish-green and are usually formed by *Spirogyra*, *Zygnema*, *Oedogonium*, *Mougeotia*, or *Vaucheria*.

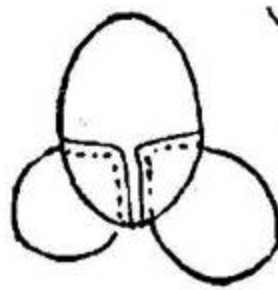
Aquatic Plants and Plant-like Algae

Lemna* and *Spirodela

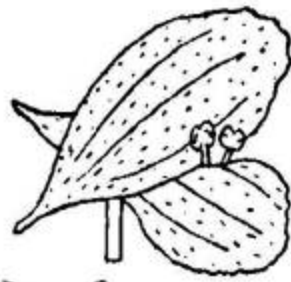
Plants with one root.....*Lemna*
 Plants with several roots.....*Spirodela polyrhiza*



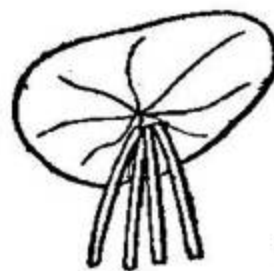
Lemna minor



Lemna minuta



Lemna trisulca



Spirodela polyrhiza

Plants with one faint vein, extending only about half the distance from root to apex; tips rounded.....*Lemna minuta*

Leaves with three distinct veins, tips rounded, with a single root. Always floating.....*Lemna minor*

Leaves with three distinct veins, oblong-lanceolate to elliptic, tapering at base into a long, slender stipe (stalk); tips pointed Rootless or with a single root. Generally submersed with vegetative plant body flattened..... *Lemna trisulca*

Elodea

Leaves 2 (1 to 4) mm wide, tapered abruptly to a blunt point; white female flowers stalked, male flowers a vase-shaped spathe borne on a long stalk*Elodea canadensis*



Elodea canadensis

Leaves 1.5 (0.3 to 1.5) mm wide, tapered to a slender point; white female flowers stalked, male flowers sessile..... ***Elodea nuttallii***



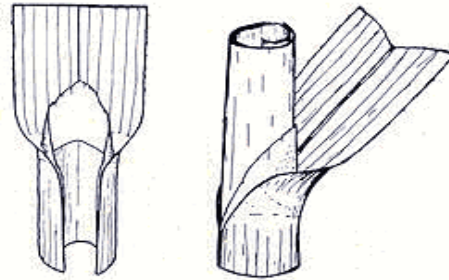
Elodea nuttallii

Grasses

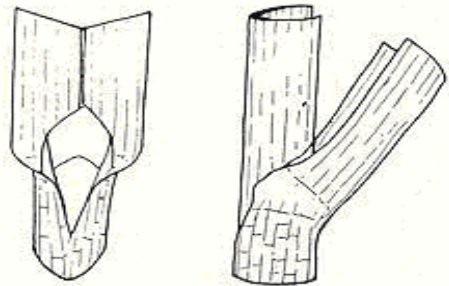
Leaves long and unequal in length, originating from base of plant, 2.5 to 5 mm wide, 5 to 25 cm long, flat or V-shaped, abruptly acute and boat-shaped at tip, glabrous, faintly ridged, two conspicuous median lines; color blue- or gray-green, not glossy. Sheath flattened or elliptical but not keeled, slightly scabrous, pale-green to purple or tinged with purple, prominently cross-nerved, closed almost to the summit but splitting easily due to its membranous nature. Collar not conspicuous, glabrous, pale, divided by midrib; Ligule thin-membranous, 2 to 4 mm long, acute entire..... ***Agrostis stolonifera***

Leaves of equal length, appearing all along stem, 2 to 7 mm wide, 5 to 20 cm long, tapering to a sharp point, prominently ridged on upper surface, midrib distinct below, surfaces scabrous or smooth; color grass green. Sheath not compressed or keeled, glabrous, green, longer than the internode on the basal shoots, split with margins overlapping. Collar prominent, glabrous, pale green, V-shaped, usually oblique. Ligule membranous, thin, 1.5 to 4.0 mm long, rounded to acute, irregularly toothed or split, minutely hairy on back.....***Glyceria striata***

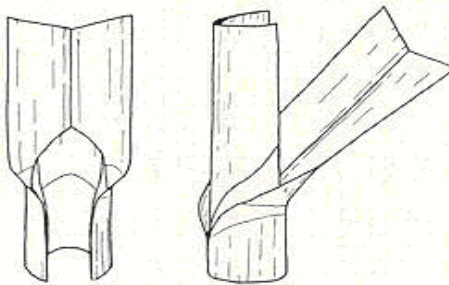
Leaves mostly occurring on lower half or third of stem; roots at nodes, 1.5 to 4 mm wide, 2 to 8 cm long, flat or V-shaped, two distinct light lines may be seen along the midvein, margins glabrous, slightly scabrous towards tip; color pale blue-green, not glossy. Sheath compressed and slightly keeled, glabrous, light green, split part way only; margins membranous and usually overlapping. Collar distinct, glabrous, pale green, V-shaped. Ligule conspicuous membranous, white, 1.0 to 3.0 mm (usually 1.2 to 1.8 mm) long, acute, entire.....***Poa annua***



Agrostis stolonifera



Glyceria striata



Poa annua

Potamogeton

All *Potamogeton* have alternate leaves

Leaves entirely submerged, grass-like, up to 2 mm wide, with 3-5 main veins.....***Potamogeton foliosus***



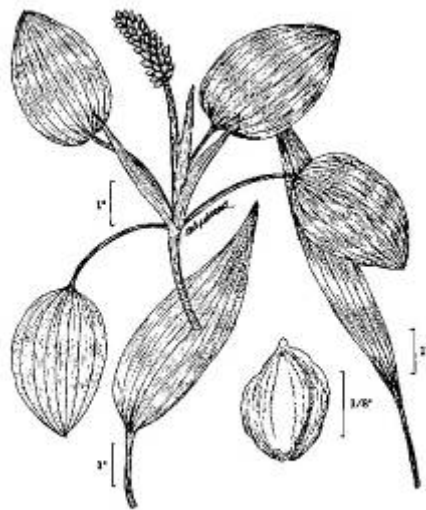
Potamogeton foliosus

Leaves usually entirely submerged and folded, up to 50 mm wide, with 9-19 main veins.....***Potamogeton illinoensis***



Potamogeton illinoensis

Leaves both floating and submerged, submerged leaves strongly folded, up to 75 mm wide, with 25-39 leaf veins.....*Potamogeton amplifolius*



Potamogeton amplifolius

Leaves both floating and submerged, submerged leaves flat, up to 25 mm wide, with 7-15 leaf veins.....*Potamogeton nodosus*



Potamogeton nodosus

Chara-Nitella

With a “main stalk” with regularly arranged whorls of shorter branches; abrasive and rough to the touch; distinct musky odor.....***Chara***



Chara

With many branches but no apparent “main stalk”, leaflets arranged in numerous whorls;
smooth to the touch; without musky odor.....***Nitella***



Nitella

Myriophyllum

Foliage of leaves with 12-28 capillary divisions; submersed leaves in whorls of 5 to 6; primary bracts or bracteal leaves ovate-lanceolate to lanceolate in shape, margins with short serrations or the lower ones entire..... ***Myriophyllum heterophyllum***



Myriophyllum heterophyllum

Foliage of leaves with 8-10 capillary divisions; submersed leaves in whorls of 3 to 4; primary bracts or bracteal leaves with long serrations, usually sub-pectinately toothed.....***Myriophyllum pinnatum***



Myriophyllum pinnatum

Foliage of leaves with a grayish cast, the segments distributed on the axis of the stem in pair or sub-opposite, those on each side extending outward nearly paralleling each other giving the plant a feathery appearance..... ***Myriophyllum spicatum***



Myriophyllum spicatum

Ludwigia

Leaf blade narrowly elliptic to subovate..... ***Ludwigia palustris***



Ludwigia palustris

Leaf blade narrowly elliptic to round.....***Ludwigia repens***



Ludwigia repens

***Heteranthera-Najas-Zannichellia* group**

Leaves mostly alternate, tips pointed, no conspicuous midrib, up to 5mm in width; flowers at end of stem and yellow.....***Heteranthera dubia***



Heteranthera dubia

Leaves mostly opposite, but sometimes appearing alternate or whorled, 2-8 cm long, margins smooth, without conspicuous midrib; flowers and fruits sessile and located at base of leaf axil giving the appearance of small “horns”.....***Zannichellia palustris***



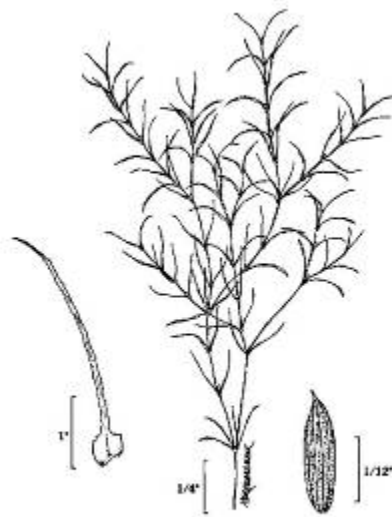
Zannichellia palustris

Leaves mostly opposite, 0.5-3.0 cm long, tips rounded, with margins minutely serrate (not visible with eye).....*Najas guadalupensis*



Najas guadalupensis

Leave opposite, 0.5-3.5 cm long, tips pointed, margins toothed (clearly visible by eye)..... *Najas minor*



Najas minor

Sedges and Rushes

Sedges-usually triangular in cross-section, *Scirpus* appear round

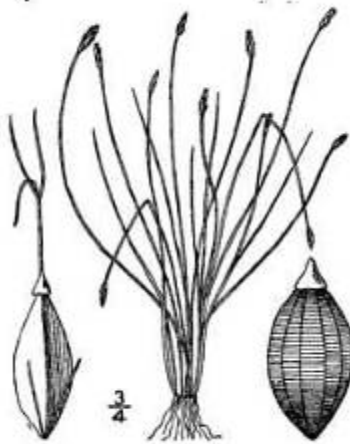
Nutlets enclosed in a distinct saclike case, leaves evident.....***Carex***



Carex

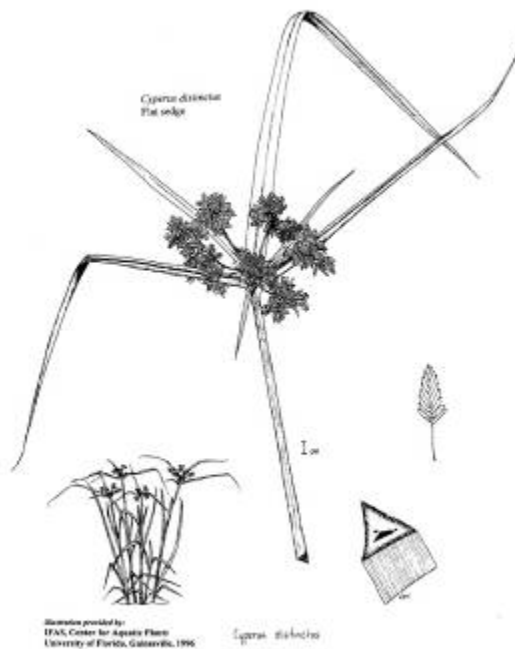
Nutlets without saclike case, naked; leaves evident.***Scirpus* and *Cyperus***

Nutlet as one terminal spikelet; plants essentially leafless.....***Eleocharis***



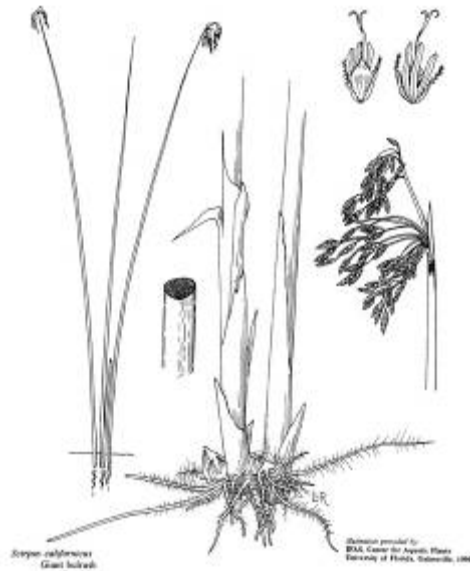
Eleocharis

Spikelet scales two-ranked, somewhat flattened.....*Cyperus*



Cyperus

Spikelet scales several ranked in an overlapping spiral pattern, not flattened.....*Scirpus*



Scirpus

Rushes-round in cross-section

Plants grass-like; base of stem with sheath, no ligule at junction of blade and sheath.....*Juncus*



Juncus

Leaf sheaths without blades, apiculate or mucronate; inflorescences appearing lateral on the stems.*Juncus effusus*

Leaf sheaths with definite blades; inflorescences terminal.....other *Juncus*

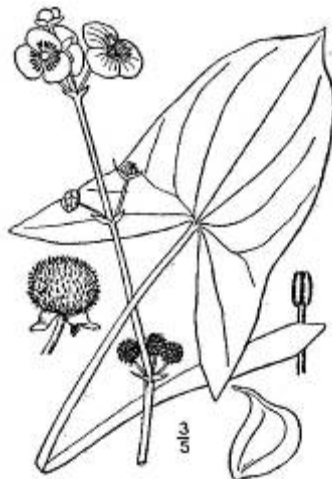
Summit of sheath extended into two white, thin papery lobes (auricles) about 1-3.5 mm long.....*Juncus tenuis*

Sheath with lobes (auricles) white tan or gray and papery, but less than 1 mm long.....*Juncus acuminatus*

Summit of sheath extended into two short rounded, firm, yellow- to copper-colored lobes (auricles).....*Juncus dudleyi*

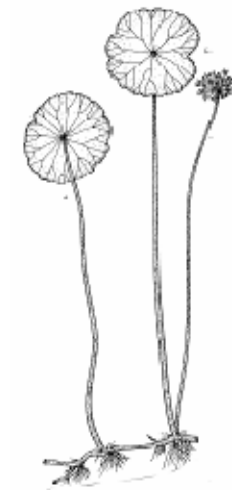
Miscellaneous Plants

Leaves arrow-shaped, emergent.....*Sagittaria latifolia*



Sagittaria latifolia

Leaves circular in shape (up to 2.5 cm in diameter) with crenulated margin; emerged or emergent..... *Hydrocotyle umbellata*



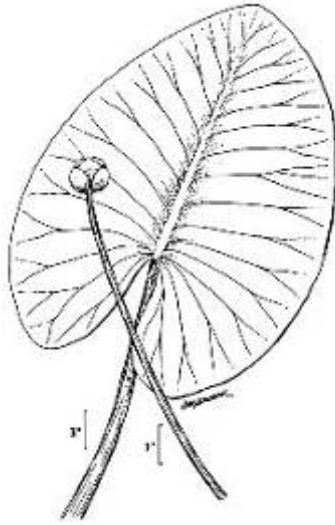
Hydrocotyle umbellata

Leaves heart-shaped to ovate, emersed or emergent, not floating.....***Plantago cordata***



Plantago cordata

Leaves heart-shaped, floating.....***Nuphar lutea***



Nuphar lutea

Line drawings adapted and modified from Correll and Correll (1975); Godfrey and Wooten (1979, 1981); U.S. Department of Agriculture Plant Database (<http://plants.usda.gov/index.html>); and West Virginia University Extension Service (<http://www.caf.wvu.edu/~forage/library/cangrass/content.htm>)

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 6: Aquatic Invertebrate Sampling

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP describes field procedures for collecting and processing invertebrate samples in the field. Appropriate QA/QC requirements are highlighted within the procedure descriptions.

I. Collecting Benthic Samples and Associated Habitat Data

Procedure:

- A. Prior to collecting benthic samples and taking habitat measurements, always complete data sheet information for spring name, date and time of survey, and initials of personnel who collect the samples.
- B. Collect one random benthic sample on transects 2, 4, 6, 8, and 10 for a total of five samples per spring. Only three such samples will be collected at Welch Spring where only three transects can be established. Benthic invertebrate samples will always be taken in an upstream direction. Individual benthic samples from the spring-run will be collected using the following *a priori* randomization procedure:
 1. Collect individual benthic samples centered on transects starting at river left, then alternating to the middle, and finally the right. If the crew is working/collecting data in an upstream direction, river right is on the workers' left, and river left is on the right. This sequence is repeated for the remaining transects. Note: the invertebrate sampling frame is centered within the area delineated by the 1m² sampling frame used for assessing physical habitat (see SOP #9).
 2. The starting sequence (left, middle, right) will be altered in each successive year so that the substrate on each transect will be sampled in a specific area only once ever third year. Note: Sample sequence in any given year should be altered only if the original starting point presents danger to the collector or if it is not accessible.

3. Samples will be collected with a Slack Surber sampler (analogous to the EPA-style rectangular benthic net) with a ~500 μm mesh net fitted with a sampling area guide (0.5 x 0.5 m rectangle of PVC pipe placed immediately in front of the net) to delineate the approximate sampling area of 0.25 m^2 (Fig. 1). Place the collection net and sample frame firmly against the substrate so that the opening is oriented directly into the spring current.



Figure 1. Invertebrate sampling net with sampling frame attached.

C. Benthic sample collection

1. While one team member holds the net firmly against the stream bottom facing the current, a second team member uses a garden cultivation tool to agitate the entire area within the net sample frame for 2 minutes (Fig. 2). At the end of 2 minutes the net can be lifted from the stream bottom, but with sufficient caution so as not to spill the sample.
2. Remove any large debris or rocks from the net and inspect for attached organisms. Scrub individual substrate pieces into the collecting net with a soft-bristled brush as needed to remove moss and attached organisms. These pieces can be discarded after any invertebrates have been removed (Note: ensure that any substrate removed from the sample area is returned to its approximate original location immediately following sample collection). Pour and rinse the contents of the net into a wash bucket with a 500 μm mesh sieve and rinse the sample by swirling the bucket in water using care not to submerge the bucket (Fig. 2). Continue this process until all the fine silt and other sediments have been washed from the sample. Remove the sample contents from the wash bucket and place them in the sample container (Fig. 2). Inspect the net, bucket, and sieve for any remaining organisms and carefully place them in the sample container. Excess water in the

sample container can be drained into the sieve and inspected for organisms so that the preservative is not overly diluted.



Figure 2. Placement of sampling net on stream bottom and agitation of substrate with garden tool (top left), washing the fine sediments from the sample using the wash bucket (top right), and transferring sample from wash bucket to sample container (bottom). Note that sample container is always held over wash bucket to ensure that sample is not accidentally spilled.

3. Once all organisms have been removed from the net, fill the jar with preservative (99% isopropyl alcohol, or 10% buffered formalin), ensure that the container is properly labeled, insert a completed sample label, and tightly close the lid.
Always complete sample labels in permanent, waterproof ink written on 100% ragbond paper. Sample debris must be completely covered by preservative. Example sample labels are provided at the end of this SOP.
 4. Repeat this procedure for each discrete sample. Prior to leaving the site, check the samples to ensure they have been properly labeled and tightly closed, and ensure data sheets are properly completed.
- D. Collect associated habitat data for each sample. Refer to SOP #9 (Physical Habitat Measurements) for descriptions and procedures for collecting habitat data.

Sample Labels

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

Ozark National Scenic Riverways Spring Invertebrate Sample

Spring Name: _____

Transect Number (2, 4, 6, 8 or 10): _____

Sample Location (L, M, R): _____

Date Collected: _____

Collectors: _____

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 7: Laboratory Processing and Identification of Invertebrates

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP explains procedures for processing and storing samples after field collection as well as identification of specimens. Procedures for storing reference specimens are also described.

I. Preparing the Sample for Processing

Processing procedures apply to all benthic samples. This is an important and time consuming step. Particular care should be taken to ensure that samples are being processed thoroughly and efficiently. The purpose of sorting is to remove invertebrates from other material in the sample.

Procedure:

- A. Sample processing begins by pouring the sample into a USGS standard sieve (500-µm) placed in a catch pan. The preservative should be kept and stored in a storage container for eventual rehydration of sample debris prior to QA/QC.
- B. Rinse the sample contents in the sieve with tap water to flush the residual preservative. Large organic material (>2 cm) should be removed by hand and rinsed into the sieve. Each piece of debris removed from the bulk sample should be carefully inspected to ensure that all attached organisms are removed. The rinsed organic material should then be kept separate from the rest of the sample or placed in the original sample container.
- C. Through elutriation, the organic debris should be separated from the inorganic content (sand and gravel). This may take several washes to accomplish. Carefully examine the inorganic content for the presence of remaining invertebrates (especially mollusk shells and Trichoptera cases). Add these specimens to the organic debris portion. Return the inorganic portion to the original sample container.
- D. Inspect the sieve to ensure no invertebrates remain following rinsing. If present, remove any specimens and add them to the organic sample fraction.

- E. The organic sample portion should then be returned to a clean 500- μ m USGS-type sieve marked into eight (8) equal portions (Fig. 1). The sieve should be placed in a shallow pan of water, and the contents floated until they are evenly distributed on the pan bottom. The sieve should then be carefully lifted from the water so that contents are not redistributed. The separated sample is now ready for sorting.

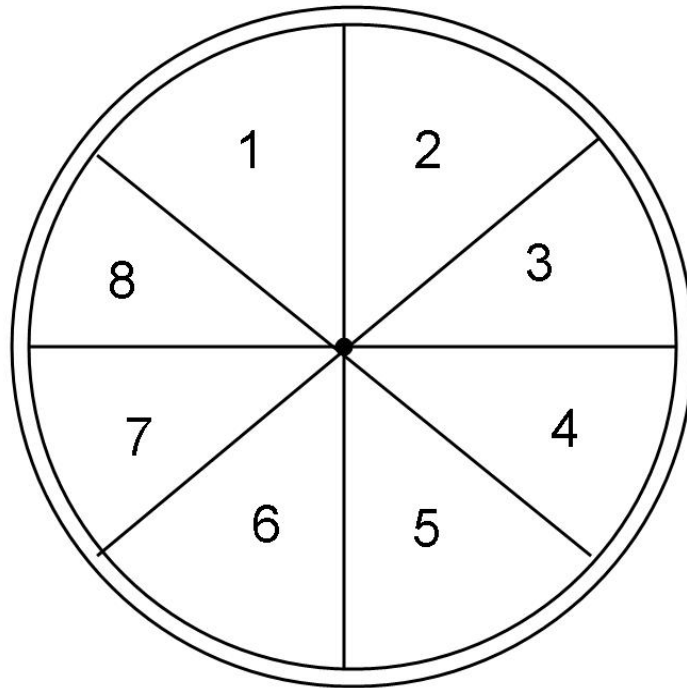


Figure 1. Diagram of a 500- μ m USGS sieve marked into eight equal fractions.

- F. Notes regarding any difficulties with sample sorting should be written on the Aquatic Invertebrate Identification & Enumeration Sheet (an example is located at the end of this SOP).

II. Subsampling

In order to ensure that the subsample adequately represents the contents of the whole sample, a minimum of 200 organisms, if present, will be removed from the sample.

Procedure:

- A. Using the random table below (Fig. 2), the sorter will randomly pick one of the one-eighth fractions of the sample in the sieve to represent a minimum 12.5% subsample.
Note: Random numbers should be selected from top to bottom.

5	7	2	5	3	2	1	2	2	1	1	7
4	6	7	2	8	3	5	8	5	5	7	8
6	3	3	7	7	8	6	1	7	2	4	3
1	8	4	8	5	5	4	3	1	7	3	1
2	2	8	1	1	6	3	5	6	8	8	5
7	1	6	4	4	4	2	7	3	4	2	4
8	4	5	6	6	1	8	6	8	6	6	2
3	5	1	3	2	7	7	4	4	3	5	6

Figure 2. Randomly generated integer sequences between 1 and 8. Sequences are oriented from top to bottom.

- B. Using a putty knife or similar tool, the fraction contents are carefully scraped from the sieve and added to a white sorting pan containing water. The bottom of the sieve in the area where the subsample was removed should then be carefully inspected to ensure that no invertebrates remain.
- C. All samples will be sorted under a minimum 10X magnification.
- D. As invertebrates are removed from the fraction, they should be counted with a hand-held enumerator. When this fraction has been completely sorted, and 200 or more organisms have been removed, no additional sorting is necessary. If less than 200 organisms were removed, the sorter should remove another randomly selected quarter fraction from the sieve and sort it. This process is repeated until a minimum of 200 organisms have been removed or the entire sample has been sorted.
- E. Always completely sort the removed sample fractions regardless of how many organisms are present (*e.g.*, the first fraction removed possibly could contain 300 or more organisms).
- F. The sorter should clearly indicate on the specimen label and lab identification sheet how many fractions of the whole sample were sorted. **This information is critical for estimating benthic densities.** For example, to estimate density for the entire sample if only one eighth section is sorted, the number of specimens in this fraction must be multiplied by a factor of 8. Other fractions and their multiplication factors are: 2=4, 3=2.67, 4=2, 5=1.6, 6=1.33, 7=1.14, and 8=1.
- G. Additionally, a “large and/or rare” taxa component is included in the subsampling routine. Large and/or rare taxa remaining in the sample that clearly were not in the sorted fraction are removed and stored in a separate vial. These specimens will be used for reflecting accurate sample diversity estimates and calculating specific metrics such as EPT.
 1. A large and/or rare additional taxa search will be completed following the subsample routine. Any large and/or clearly rare organisms (*e.g.*, *Corydalus cornutus*, *Pteronarcys picketii*, tabanids, tipulids, dragonfly larvae, crayfish,

gordian worms, large beetles, other unusual species, *etc.*) in the sample that clearly were not in the subsampled fraction will be removed, placed in a separate vial, and labeled appropriately (Fig. 3). There may be several or no specimens depending on the sample.

2. Just because a creature is large does not mean it should be removed during this process. It must fit the criterion that it was not present in the subsample.
 3. A large and/or rare collection may not be necessary for all samples.
- H. Any invertebrates present in the subsampled squares will be stored in a separate storage vial, preserved, and properly labeled (see below).
- I. Organic debris from the subsampled portion will be retained in a separate container until QA/QC checks are completed.

III. Sample Preservation and Labeling

- A. Invertebrates removed from the bulk samples will be stored in 75-80% isopropyl alcohol.
- B. Labels will be written only on rag bond paper in permanent water proof ink (both supplied). Labels written in pencil are not acceptable.
- C. Label data should be printed neatly and include the following: site name (*e.g.*, Blue Spring), transect number (2, 4, 6, 8, 10), sample location (L, M, R), sampling date, and collector initials (Fig. 3). All of this information is on the original sample label.
- D. The vial label for “large and rare” taxa should be specifically labeled as such and also include the original sample data as well.
- E. Do not crowd the collected specimens excessively as it inhibits long term preservation.

<u>Bulk Sample:</u>	Park Name: OZAR Site Name: Round Spring Sample: Transect 1, Left Collection Date: 12 Dec 06 Collector Initials: DEB, HRD
<u>Large & Rare:</u>	LARGE & RARE Park Name: OZAR Site Name: Round Spring Sample: Transect 1, Left Collection Date: 12 Dec 06 Collector Initials: DEB, HRD
<u>Identification:</u>	Park Name: OZAR Site Name: Round Spring Sample: Transect 1, Left Collection Date: 12 Dec 06 Taxon Name: Chimarra Det. D. E. Bowles

Figure 3. Example specimen labels.

IV. Identification of Invertebrates

To the extent possible, all invertebrates should be identified to genus exclusive of the groups and selected conditions indicated below.

Procedure:

- A. A dissecting microscope and taxonomic keys are used to identify each specimen to the genus level whenever possible. For some taxa only higher taxonomic levels can be obtained, and some of these are listed below. In most cases, an entire sample can be identified to the required level, but an occasional sample may contain early instars or damaged specimens. In such cases, the specimen should be identified to the lowest level possible, as indicated in Table 1.
- B. The primary keys will be Merritt *et al.* (2008) for identification of insects and Pennak (1989) for identification of non-insect invertebrates. Additional taxonomic references for specific groups include Moulton and Stewart (1996), Poulton and Stewart (1991) and Wiggins (1995). A list of taxa known or likely to occur at OZAR is included in SOP #12 (Data Analysis). Accuracy of scientific names will be checked at the Interagency Taxonomic Information System (ITIS) website at: http://www.itis.usda.gov/advanced_search.html.

Table 1. Taxonomic levels for identification when not the Genus level.

Phylum Nematoda
Phylum Nematomorpha
Phylum Annelida, Class Hirudinea, Families
Glossiphoniidae, Piscicolidae, Hirudinidae, Erpobdellidae
Phylum Annelida, Class Oligochaeta, Families
Aeolosomatidae, Opistocystidae, Naididae, Haplotaxidae,
Enchytraeidae, Lumbriculidae, Tubificidae
Phylum Arthropoda, Class Arachnoidea, Order Hydracarina
Phylum Arthropoda, Class Crustacea, Order Ostracoda
Phylum Arthropoda, Class Insecta, Order Diptera, Family
Chironomidae

- C. A running total of each taxon for each sample should be recorded on the laboratory bench sheet that is signed or initialed by the identifier. Final counts of each taxon should be entered on the bench sheet when a sample is complete.
- D. If **damaged organisms** can be identified, they are counted ONLY if:
1. The fragment includes the head, and, in the case of arthropods, the thorax. For Oligochaetes, count only fragments that include a rounded end that could be a head or tail end.
 2. The mollusk shell (bivalve or gastropod) is occupied by a specimen.
 3. The specimen is the sole representative of a taxon in the sample.
- E. If **early instar or juvenile** specimens can be identified, they are counted ONLY if:
1. With confidence, they can be associated with one or more mature specimens that have a more developed morphology
 2. The specimen is the sole representative of a taxon in the sample.

V. Sample Storage and Reference Collection

Procedure:

- A. Identified specimens are stored in vials with 75-80% isopropyl alcohol and labeled with the taxon name, date collected, park and spring name, and name of identifier. Organisms will be retained for at least three years and stored at the NPS HTLN office located at Missouri State University, Springfield, MO.

- B. A reference collection consisting of a few representative specimens of each taxon should be prepared and stored in properly labeled vials containing 75-80% isopropyl alcohol. Vials should be labeled as above (Fig. 3).
- C. Regional or other taxonomist specialists should review the identifications for accuracy. One set of vials should be stored at the NPS HTLN office located at Missouri State University, Springfield, MO. Additional sets of specimens should be maintained in the laboratory where identifications are performed for use as reference and training.

VI. QA/QC

Procedures:

Sample processing and sorting

The following QA/QC procedure (adapted from USEPA, 2004b) will be used on sorted invertebrate samples.

- A. Initially, a QA officer (initially the Aquatic Program Leader) will use a microscope to check **all** sorted fractions from the first five samples processed by a sorter to ensure that all organisms were removed from the detritus. This will not only apply to inexperienced sorters, but also to those initially deemed as “experienced.” Qualification will only occur when sorters are consistent in achieving >90% sorting efficiency after at least five samples have been checked. Samples will be checked using 10x magnification.
- B. The Aquatic Program Leader or other QA Officer will calculate percent sorting efficiency (PSE) for each sample as follows:

$$PSE = A/A+B (100)$$

where A = number of organisms found by the primary sorter, and B = number of recoveries (organisms missed by the primary sort and found by the QC check).

If the sorting efficiency for each of these five consecutive samples is >90% for a particular individual, this individual is considered “experienced” and can serve as a QC Officer. In the event that an individual fails to achieve >90% sorting efficiency, they will be required to sort an additional five samples and his sorting efficiency will continue to be monitored. If they show marked improvement in their sorting efficiency prior to completion of the next five samples, however, and reaches the >90% sorting efficiency, the QA Officer may, at their discretion, consider this individual to be “experienced.” Sorting efficiency should not be calculated for samples processed by more than one individual.

- C. After individuals acquire a >90% sorting efficiency, 10% (1 out of 10) of their samples will be checked.
- D. If an “experienced” individual fails to maintain a >90% sorting efficiency as determined by QC checks, QC checks will be performed on every fraction of five consecutive samples until a >90% sorting efficiency is achieved. During this time, that individual will not be able to perform QC checks.
- E. Residues of sorted bulk samples should be returned to their original sample containers and rehydrated with the alcohol saved during sample preparation. All sorted samples will be kept until QA/QC checks are complete and permission is given by the Program Leader to discard the material.

Specimen Identification

- A. The identification for 10% of the samples must be checked for quality control. The QC sample will be randomly chosen.
- B. Specimens from the QC samples will be analyzed by another trained and experienced individual within the same laboratory (*e.g.*, Program Leader). Percent similarity of the two samples must exceed 90%.
- C. If an individual fails to maintain a >90% identification efficiency as determined by QC checks, QC checks will be performed on every identified specimen of five consecutive samples until a >90% sorting efficiency is achieved.
- D. If any specimens are incorrectly identified, all specimens assigned to that taxon will be reexamined.
- E. If a taxon is especially difficult to identify, specimens will be sent to a person with taxonomic expertise in invertebrates of the region. It is also important that the taxonomist maintains contact with other taxonomists through professional societies and other interactions, and stays current with the pertinent published literature.

VII. Bulk Sample Disposition

When the sample is finished, return the remainder of the bulk sample to its original container with its original label and old alcohol, and return to storage. Mark the top with an X or other mark to identify the sample as one that has been picked. These samples will be subjected to a QA/QC analysis by the Program Leader.

Page ____ of ____

State: _____
Date of Det: _____
Determiner(s): _____

ORDER (or higher)	FAMILY	GENUS	SPECIES	TALLEY/COUNT	Tot Cnt	RC
Diptera						
Coleoptera						
Ephemeroptera						
Plecoptera						
Trichoptera						
Amphipoda						
Other						

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 8: Fish Community Sampling

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

The fish sampling protocols described in this SOP present methods for collecting a representative sample of the fish community from a spring-run. Such a sample should contain most, if not all, species in the spring-run at the time of sampling, in numbers proportional to their actual abundance.

Methodology presented below follows the NAWQA protocols described in Meador *et al.* (1993), Walsh and Meador (1998), and Moulton *et al.* (2002). The methods have been modified where appropriate to meet the specific objectives of the National Park Service. Further, only those portions of the NAWQA protocols applicable to sampling fish at the large springs at OZAR are included here. Electrofishing will be the only method of collecting fish in the springs and it is conducted in a single pass of the sampling reach for wadeable reaches. This SOP presents both techniques for fish sampling and procedures for processing fish.

I. Site and Sampling Conditions

Procedures:

- A. Record the spring name, date, reach length, and reach description on the Fish Field Form 1 (included below).
- B. Record weather conditions (cloud cover, wind, and precipitation) and note any other weather or site conditions that might affect sampling efficiency.
- C. When establishing the permanent reach, record GPS coordinates of upper and lower reach boundaries (see SOP #3, Reach Selection). This should be done only on the first visit to a spring and not again thereafter.
- D. Record the total discharge on the fish field form (see SOP #10, Measuring Discharge).

II. Electrofishing Methods

Overview

Electrofishing is the use of electricity to capture fish. A high-voltage potential is applied between two or more electrodes that are placed in the water. The voltage potential is created with either direct current or alternating current; only direct current is used in the NAWQA Program and will be used in this protocol. Direct current produces a unidirectional, constant electrical current. Pulsed direct current, a modified direct current, produces a unidirectional electrical current composed of a sequence of cyclic impulses (Meador *et al.*, 1993). The frequency of the pulses produced when using pulsed direct current can be adjusted by the operator.

Procedures for Setting Electrofishing Gear:

- A. Prior to sampling, measure conductivity of the water to aid in selecting voltage, frequency, and duty cycle for the electrofishing gear being used. Note: Effectiveness of electrofishing gear is primarily affected by water conductivity.
 - 1. Low conductivity water is resistant to flow, reducing the amount of electrical current traveling through the water.
 - 2. High conductivity water concentrates a narrow electrical field between the electrodes (Meador *et al.*, 1993).
- B. General Rule for Voltage: output of electrofishing gear should be ~ 3,000 watts. Voltage times amperage equals wattage. If the gear is without ammeters, some pre-sampling experimentation is necessary.
 - 1. In low conductivity water, high voltage and low amperage are needed.
 - 2. In high conductivity water, low voltage and high amperage are needed.
- C. General Rule for Frequency: a pulse rate range from 30 to 60 pulses per second (pps) should be used. Frequencies > 60 pulses per second (pps) are effective in collecting fish but can cause injuries, especially to larger fish (Coffelt Manufacturing, Inc., cited in Meador *et al.*, 1993; McMichael *et al.*, 1998). Pulse rates < 30 pps have caused low incidence of injury, but are generally ineffective in collecting fish. Therefore, pulse rate should be set to produce an effective collection of all fish species and sizes while minimizing injury.
- D. General Rule for Duty Cycle: a range of 25% - 100% should be used. Duty cycle is calculated as: pulse frequency X pulse duration X 100. Electrofishing with intermediate to high duty cycles reduces injury and mortality to fish (Dolan and Miranda, 2004).

Electrofishing Wadeable Reaches

General Procedures:

- A. Channel width, depth, and access should be considered before choosing between backpack and towed electrofishing methods (Fig. 1).
 1. Backpack electrofishing (with a single anode) should be used in shallow (< 1 m) and narrow (< 5 m wide) reaches. A crew of three is needed.
 2. Towed electrofishing gear (multiple anodes) is more effective in wide (> 5 m) wadeable reaches with pools deeper than 1 m. A crew of 5 is needed.
- B. All crewmembers must wear low voltage rubber gloves and waders to protect them from the electrical current and wear polarized glasses to enhance their ability to see fish.
- C. Techniques for collecting samples using either backpack or towed electrofishing gear are generally similar.



Figure 1. Backpack (left) and towed barge (right) electrofishing gear used in wadeable spring-runs.

Sampling Direction:

- A. Regardless of the gear used, a single electrofishing pass is made in the reach (Fig. 2).
- B. Sampling begins at the downstream boundary of the sampling reach and proceeds upstream. Sampling in an upstream direction in wadeable streams is preferred because disturbance of the streambed by crewmembers increases turbidity and reduces visibility of the stunned fish (Hendricks *et al.*, 1980).
- C. Sampling requires alternating between banks in a “zigzag” pattern in order to cover habitat features as one proceeds upstream (Fig. 2). Using the zigzag pattern, every effort

is made to sample all geomorphic channel units and instream habitat features, such as woody snags, undercut banks, macrophyte beds, or large boulders.

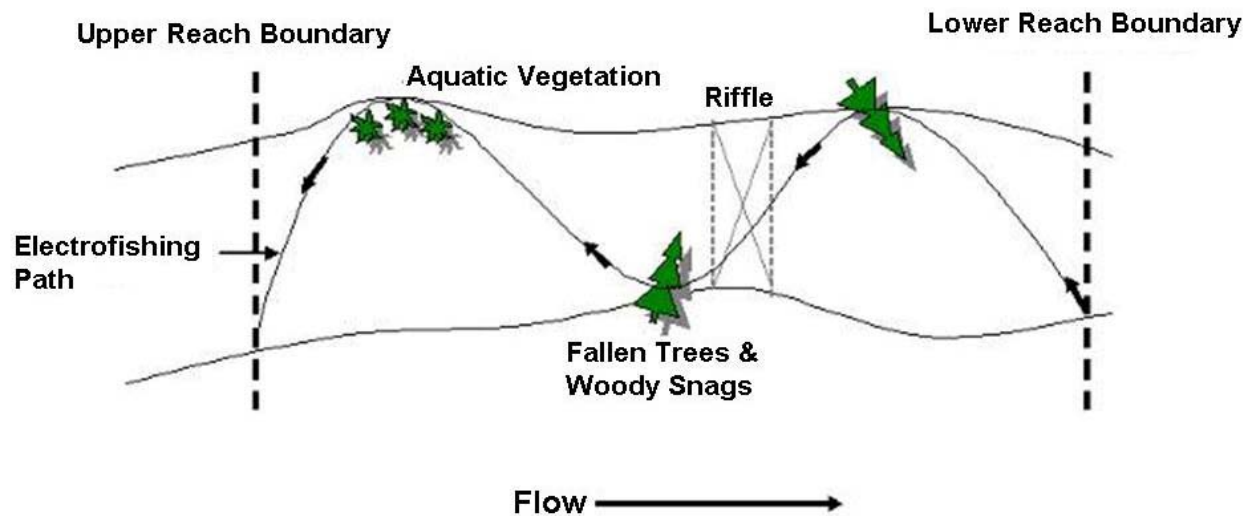


Figure 2. Single pass zigzag sampling technique used in backpack or towed electrofishing for wadeable portions of the spring-run.

Electrofishing Techniques:

- A. Continuous application of electrical current and herding of fish by the operator is used in open run habitats. Fish generally respond to continuous electrical current by avoiding exposure to the electrical field, resulting in fish moving just ahead and away from the operator. The operator uses the current to herd the fish into natural barriers such as banks, bars, or shallow riffles to facilitate capture.
- B. Intermittent application of electrical current and herding fish to the operator can be used in runs or long pools in narrow reaches. This technique requires a crewmember (or two) to enter the reach upstream of the operator. The crewmember(s) moves downstream toward the operator, creating a disturbance and driving fish downstream. The electricity is turned on once the fish are visible and close to the anode.
- C. To sample shallow riffles, the operator sweeps the anode across the riffle from upstream to downstream while walking across the riffle. Crewmembers with nets are positioned downstream of the operator to collect stunned fish.
- D. The ambush technique should be used when a reach has complex instream habitat such as woody debris, boulder fields, etc. Using this technique, the operator approaches the habitat feature with the electrical current off. In quick succession, the anode is thrust close to the habitat feature, the current is turned on, and the anode is withdrawn from the feature. This produces galvanotaxis, where the current forces the fish to swim out from the habitat feature and towards the anode (Meador *et al.*, 1993).

Electrofishing Nonwadeable Reaches

Procedures:

- A. Nonwadeable reaches are sampled using an electrofishing boat (Fig. 3) with supplemental use of backpack or towed electrofishing gear in wadeable areas.
- B. A boat crew consists of a driver and one or two persons who collect the fish with dip nets.
- C. For safety, all crewmembers must wear low voltage gloves, waders, and life jackets. Polarized sunglasses should also be worn.



Figure 3. Boat electrofishing gear used in nonwadeable reaches.

Sampling Direction and Speed

- A. Two separate electrofishing passes, one along each bank, are made in the reach (Fig. 4).
- B. Sampling begins at the upstream boundary of the sampling reach and proceeds downstream by maneuvering the boat along one shoreline. Sampling is conducted downstream because when fish are stunned they are carried downstream by the flow, providing greater opportunity for capture by netters standing on the bow of the boat.
- C. A zigzag pattern between the bank and center channel is used to collect fish from all instream habitats (Fig. 4). Once one side is complete, the other bank is sampled.
- D. The electrofishing boat is operated at a speed equal to or slightly greater than the current velocity. The boat should be positioned so the bow is angled downstream and

toward the bank. This allows the boat operator to reverse direction (generally upstream and away from the banks) and not pass over stunned fish.

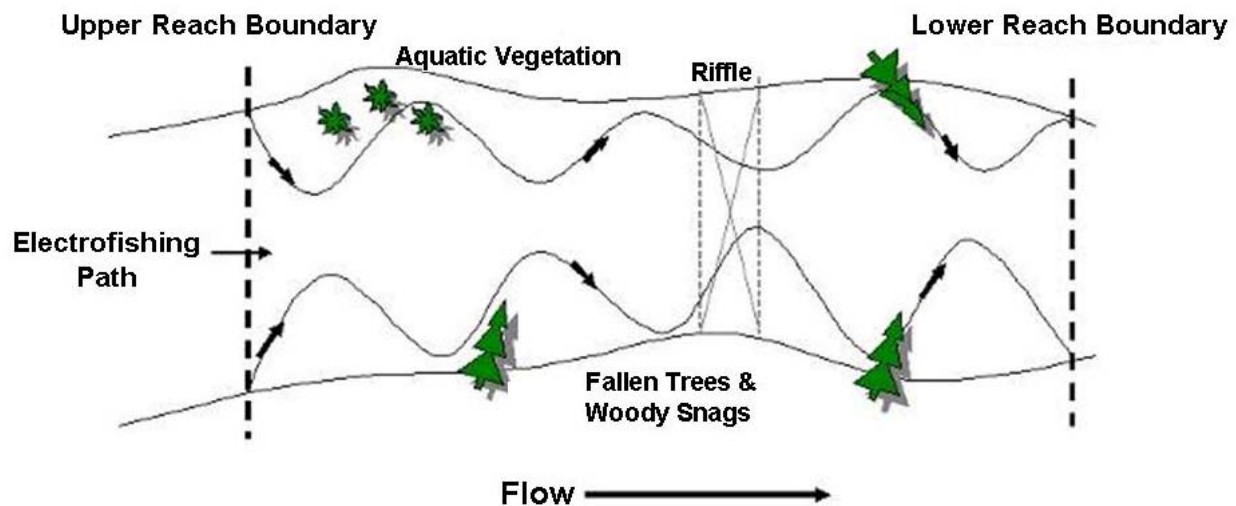


Figure 4. Double pass zigzag sampling technique used in boat electrofishing for non-wadeable portions of the spring-run.

Electrofishing Techniques

1. Continuous application of current is used for most nonwadeable reaches.
2. Intermittent application of current may be more effective in areas with clear or shallow water. Fish are approached with the current off. When the anodes are in position near the fish, the current is applied.
3. Ambush technique (similar to that in wadeable reaches) is used for sampling instream habitat features along the shoreline. The boat is maneuvered close to the habitat feature with the electrical current off. As the anode is placed near the habitat feature, the electrical field is generated and the boat is backed away from the habitat feature.

III. Sample Processing

The goal of processing fish in the field is to collect information on taxonomic identification, length, weight, abundance, and the presence of external anomalies with minimal harm to specimens that will be released alive back into the spring-run. Many species are too small or difficult to identify in the field. These specimens must be brought back to the laboratory for processing.

Procedures:

- A. If different types of gear are used within a reach, fish samples from different gear types are kept separate.

- B. Hold fish in a manner consistent with minimizing stress or death. Captured fish are placed in a live cage or aerated holding tanks during processing to reduce stress and mortality. Regardless of the effort made to minimize handling and stress to fish, some mortality will occur.
- C. Sort fish into identifiable and unidentifiable groups. Identifications are made to species level by a crewmember that is familiar with the fish species commonly found in the study area. Taxonomic nomenclature follows that established by the American Fisheries Society's Committee on Names of Fishes (Robins *et al.*, 1991). Process threatened and endangered (T&E) or sensitive fish species before other identifiable species.
- D. Obtain a subsample of 30 individuals of each identifiable species for measurement.
 - 1. A "blind grab" technique is used where a dip net is used to make a pass through the entire bucket or holding tank to ensure fish of various sizes are captured with each "grab".
 - 2. For species that have a large size range with different sizes being collected by specific gear (*e.g.*, large sunfish collected with boat and small sunfish collected with backpack), a blind grab will be taken from samples collected by each gear type and a representative subsample will be measured.
- E. Measure total length (Fig. 5) and weight of 30 individuals for each and record any anomalies.
 - 1. Length measurements are determined by using a measuring board consisting of a linear metric scale on a flat wooden or plastic base with a stop at the zero point. Total length is taken from tip of snout (with mouth closed) to end of caudal fin (while depressed).
 - 2. Weight measurements are obtained by using portable electronic or hanging scales.
 - 3. A batch weight will be recorded for smaller species (*e.g.*, minnows, darters, sculpins, and madtoms). Individual weights will be recorded for species that obtain large sizes (*e.g.*, bass, sunfish, catfish, suckers).
 - 4. Record anomalies and presence of black spot (*Neascus* spp.), a fish parasite, for each fish measured. Anomalies are externally visible skin or subcutaneous disorders or parasites (Ohio Environmental Protection Agency, 1987). They include deformities, eroded fins, lesions, and tumors, collectively referred to as "DELT anomalies" (Sanders *et al.*, 1999). Other external anomalies, such as anchor worm (*Lernaea* spp.), and popeye disease should also be noted.

5. Once 30 fish of a species have been measured, the remaining specimens are counted.

F. Record data on the Fish Field Data Sheet #2.

1. Record the spring name, date, gear used, sampling effort (time spent electrofishing).
2. Record the three letter initials of all crewmembers. Record the person identifying/measuring fish, the person recording the data, and those who operated the electrofishing equipment and netted fish.
3. For each individual of a species, record the length/weight and any anomalies. For larger fish, record individual weights. For smaller fish, batch weigh 30 specimens.
4. Record the additional number of fish collected for each species under "Species Count". For example, if 46 white suckers were collected at a reach, 30 fish would be measured and weighed and the remaining 16 would be counted (*i.e.*, Species Count = 16).

G. Preserve selected specimens for identification in the laboratory (those too small or difficult to identify in the field) or for a reference collection. Specimens are preserved in 10 percent buffered formalin.

1. Make a small incision along the body for specimens larger than 100 mm. This allows formalin to penetrate the body cavity.
2. All unidentified specimens collected with the same gear type can be preserved in a single jar with a label that contains the spring name, date, gear type, and sampling effort.
3. All preserved specimens will be identified at the HTLN Aquatic Program laboratory at Missouri State University using a microscope and the *Fishes of Missouri* identification key (Pflieger, 1997). Specimens not used for a reference collection will be disposed of after one year.



Figure 5. Total length measurements for a fish.

Fish Community Field Data Sheet 1

Spring Name: _____

Date: _____

Crew: _____

Weather Conditions: Cloud cover: _____ % Wind: *Calm* *Light* *Moderate* *Gusty*

Precipitation: *None* *Rain* *Sleet* *Snow* Intensity: *N/A* *Light* *Moderate* *Heavy*

Other Weather: _____

Reach Data:

Fish Reach Coordinates: Lower UTM Coordinates _____

Upper UTM Coordinates _____

Discharge (m^3/s): _____ (record points on Discharge datasheet & transfer total discharge here)

Additional Comments: _____

	Date	Initials
Data Entered		
Data Verified		

Fish Community Data Sheet 2

page ____ of ____

Spring Name: _____ Date: _____ IDed by: _____ Recorder: _____

Gear: Backpack Barge Boat

Crew: Shocker(s):

Effort (sec.): _____

Bout/Barge: _____

Netter(s): _____

Seine: _____

Anomalies: D = deformities, E = eroded fins, L = Lesions, T = tumors, B = Blackspot

[illegible][illegible][illegible][illegible][illegible][illegible][illegible][illegible][illegible][illegible][illegible][illegible]

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 9: Physical Habitat Measurements

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

Habitat composition within a stream is an important component in shaping biotic communities. The type and abundance of specific habitat characteristics (*i.e.*, woody debris, substrate size, *etc.*) will influence species presence and relative abundance, as well as size structure of the populations. Because of its importance to invertebrates and fish, physical habitat data will be collected as part of this protocol to examine relationships between environmental conditions and biotic communities. The methods described in this SOP have been modified from EMAP (Lazorchak *et al.*, 1998) and NAWQA protocols (Fitzpatrick *et al.*, 1998) to meet the objectives of the National Park Service.

I. Collecting Instream Point Habitat Data

Procedures:

- A. Prior to collecting samples and taking habitat measurements, always complete data sheet information for spring name, date and time of survey, and initials of personnel who collect the samples.
- B. A crew of two to three persons will collect physical habitat data.
- C. Equipment necessary to complete habitat sampling is found in SOP #1 (Preparation for Sampling).
- D. When collecting data, stream banks are referred to in a downstream perspective. Therefore, if the crew is working/collecting data in an upstream direction, river right is on the workers' left, and river left is on the right.

- E. At each transect, measure wetted width and record channel unit and pool form (if applicable). Channel unit definitions are as follows:
1. Riffle. An area of the stream with steepest slope and shallowest depth, often rocky substrate, and a swift moving current. Thalweg is usually poorly defined.
 2. Run or Race. Differ from riffles in that depth of flow is typically greater and slope of the bed is less than that of riffles. Runs will often have a well defined thalweg.
 3. Glide. Normally located immediately downstream of pools. The slope of the channel bed through a glide is negative while the slope of the water surface is positive. The head of the glide can be difficult to identify. Use the following characteristics to help you locate the head of the glide:
 - a. A location of increased flow velocity coming out of the pool,
 - b. A location with a steeply sloped bed rising out of the pool and decreasing to a lesser gradient,
 - c. A location where the thalweg coming out of the pool becomes less well defined and essentially fades completely.
 - d. A location approximately the same elevation as the tail of the run.
 4. Pool. Has a relatively slow current and is usually found at stream channel bends, upstream of riffles, or on the downstream side of obstructions such as boulders or fallen trees. The stream bottom in a pool is often bowl shaped and represents the deepest locations of the reach. Water surface slope of pools at below bankfull flows is near zero.
- F. Canopy cover is visually observed by looking directly overhead at each point and categorizing the percentage cover within one meter upstream and downstream of a transect. If a bridge or other manmade structure is producing the canopy, this should be clearly indicated in the comments section.
- G. In-stream habitat is collected at three points along each transect: center channel (or middle) and half the distance between center and the left and right banks (Fig. 1).
- H. Place 1m² sampling frame on bottom of spring-run centered on the transect.

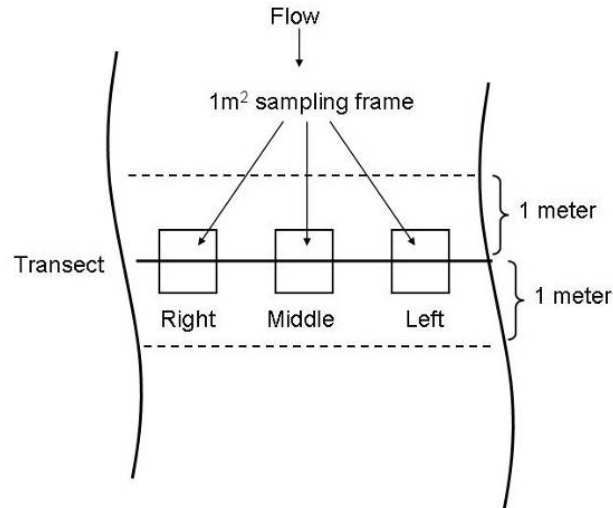


Figure 1. Location of in-stream habitat and fish cover collection at a transect.

1. Velocity (meters/second) and depth (centimeters) are measured concurrently at each sample point and immediately in front of the sample frame (Fig. 2). Depth and velocity can be measured by a third team member while the other two members collect the benthic sample described in SOP #5. Measurements are done using a FLO-MATE Model 2000 or USGS Pygmy current meter attached to a top-setting wading rod. The rod allows for quick and easy measurements of depth with incremental markings and an adjustable arm that places the current meter at the proper depth for measuring velocity (60% of the depth from the surface of the water). Velocity should be recorded in meters per second and increments on the wading rod are in centimeters. Greater detail regarding use of the FLO-MATE 2000 is provided in SOP #10 (Measuring Spring Discharge).



Figure 2. Measuring depth and current velocity in front of a sampling frame.

2. Record measurements for the following habitat variables based on visual estimates from within the sample frame. Percentage categories are: 0= none (0%), 1= sparse (<10%), 2= moderate (10-40%), 3= heavy (40-75%), 4= very heavy (>75):
 - a. Percent embeddedness of the substrate (*e.g.*, bedrock & hardpan clay = 0% embeddedness; sand, clay, silt = 100% embeddedness)
 - b. Percent periphyton
 - c. Percent aquatic vegetation
 - d. Percent filamentous algae
 - e. Percent organic material
3. Within the sampling frame, visually assess dominant substrate size (most common size) present using the Wentworth Scale. The substrate size category field sheet (see below) can aid in visually assessing size categories. A piece of substrate belongs to the smallest size box through which it will fit on any axis. Record the corresponding substrate code on the data sheet. For substrate codes 1-3, there are no boxes shown on the field sheet to estimate their respective sizes because these substrates are so small. For these three substrate codes, it is necessary to grab a sample from the plot for assessment. The general rule is:
 - a. Code 1 (silt/clay) feels slick between thumb and finger with no evidence of grit.
 - b. Code 2 (very fine sand) has a barely perceptible gritty feel.
 - c. Code 3 (fine sand) has a distinct gritty texture.

Note: This technique is used to assess the dominant substrate in the sample area. It is not intended to fully characterize the substrate profile of the stream bottom.

Table 1. Substrate size classes to be used for characterizing substrate based on the Wentworth Scale.

Size Code	Particle Diameter Range (mm)	Category
1	<0.062	Silt/clay
2	0.062-0.125	Very fine sand
3	0.125-0.25	Fine sand
4	0.25-0.50	Medium sand
5	0.50-1	Course sand
6	1-2	Coarse sand
7	2-4	Fine gravel
8	4-5.7	Medium gravel
9	5.7-8	Medium gravel
10	8-11.3	Coarse gravel
11	11.3-16	Coarse gravel
12	16-22.6	Small pebble
13	22.6-32	Small pebble
14	32-45	Large pebble
15	45-64	Large pebble
16	64-90	Small cobble
17	90-128	Small cobble
18	128-180	Large cobble
19	180-256	Large cobble
20	256-362	Boulder
21	362-512	Boulder
22	512-1024	Boulder
23	>1024	Boulder
24	Bedrock	Bedrock

III. Collecting Fish Cover and Bank Habitat Data

Procedures:

- A. Fish cover and bank habitat data are collected **only** in conjunction with fish sampling at the spring-runs or every third year.
- B. Fish cover and bank habitat measurements are collected in relation to the 11 equally-spaced transects described in SOP #3 (Reach Selection).

Fish Cover

It is important to document the presence of fish cover in a spring because different species have affinities for various cover types.

1. Assess fish cover by recording all cover types present along each transect. Filamentous algae, hydrophytes, boulders (sizes 21 to 23 on the Wentworth substrate sheet), and any artificial cover are assessed within a 1 m² plot. If artificial cover (*e.g.*, cinder block, car tire, *etc.*) is present, the type of cover should be noted in the comments section.
2. Assess small and large woody debris along a 1m belt along the transect (1 m on each side of the transect), dividing the belt into left and right side of center (Fig. 1). Small woody debris is defined as being less than or equal to 10 cm in diameter at its largest end, and large woody debris is greater than 10 cm in diameter at its largest end.
3. Fish cover along the banks is assessed within 1 meter upstream and downstream of the transect. Cover along the banks include trees/roots, overhanging vegetation, undercut banks, and bluffs (within 5 m of wetted edge).

Bank Measurements

Characteristics of both the bank and riparian areas can affect spring processes and fish habitat. For example, banks that are bare with steep angles are likely to erode during high flow events, increasing the amount of fine sediment entering a spring and degrading habitat for benthic species (both fish and invertebrates) by burying large gravel/cobble substrates.

The bank is defined as the area of steep sloping ground bordering the stream that confines the water within the channel at normal water levels, and is located between the channel and the floodplain (Fig. 7, Fitzpatrick *et al.*, 1998). The floodplain is defined as a flat or gently sloping depositional area adjacent to the stream. At low flows, it may be difficult to determine the location of the bank due to the presence of bars. Bars are defined as areas usually devoid of woody vegetation (such as small trees and shrubs), and contain coarse materials such as large gravel or cobble. These areas will be covered by water during normal flow and therefore are not considered part of the bank. Each bank measurement begins at the “true” bank (*i.e.*, area of steep slope). In some instances the bank will begin at the wetted edge. If gravel or sand bars are present at a transect, these will not be included in the bank assessment, but will be noted in the comments section by recording the length of the bar from water’s edge to the bank.

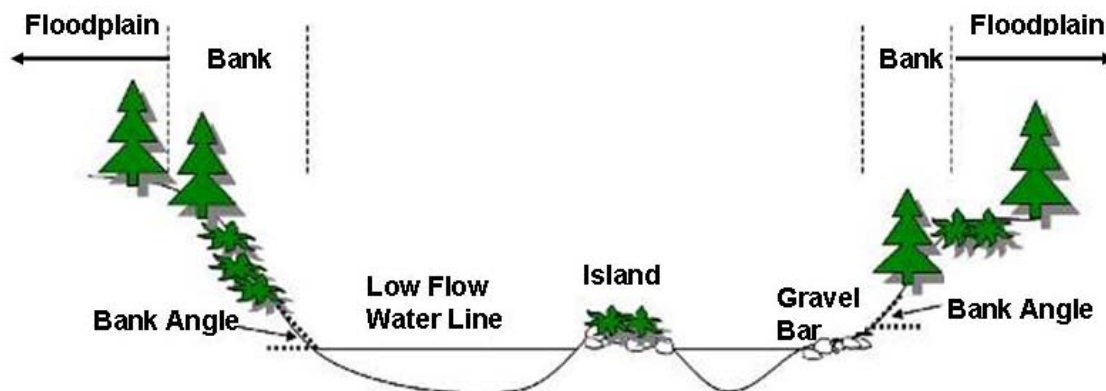


Figure 7. Illustration showing banks and floodplains of a stream.

1. Assess bank stability at each transect and record the category for each characteristic (see Bank Cover field sheet below). Bank characteristics used to measure bank stability are: Angle, Substrate, Percent Vegetative Cover, and Height.
 - a. Bank angle and substrate are observed from the bottom of the bank (*i.e.*, at wetted edge or at the top base of the bar, if one is present), and the category code is recorded. To assess the bank substrate, the Wentworth scale is used to visually estimate the dominant substrate type, and the corresponding category code on the data sheet (**not** the Wentworth code) is recorded.
 - b. Percent vegetative cover and bank height are assessed from the bank bottom to 10 meters into the bank.
2. Assess bank cover for each bank from the bottom of the bank to 10 meters into the bank. Bank cover categories include large trees, small trees/shrubs, grass/forbs, bare sediment, and artificial cover. If artificial cover is present on the bank (*i.e.*, rip-rap, concrete structures), the type of cover should be noted in the comments section. NOTE: For bank cover, more than one cover type may be recorded if two cover types are relatively equal in abundance.

IV. Miscellaneous Actions

- A. Record any necessary notes about the collection site or specific samples.
- B. Take digital photographs of the reach sampled from upstream and downstream perspectives at mid-channel.

In-stream Habitat Assessment Form

Page ____ of ____

Spring Name:

Date:

Crew:

Reach Length:

Transect Spacing Interval:

Trans	Channel Unit	Pool Form	Width (m)	Sample Cell	Depth (cm)	Velocity (m/sec)	Dominant Substrate	Embeddedness	Periphyton	Filamentous Algae	Aquatic Vegetation	Organic Debris	Canopy Cover
11*				Left									
				Middle									
				Right									
10				Left									
				Middle									
				Right									
9				Left									
				Middle									
				Right									
8				Left									
				Middle									
				Right									
7				Left									
				Middle									
				Right									
6				Left									
				Middle									
				Right									
5				Left									
				Middle									
				Right									
4				Left									
				Middle									
				Right									
3				Left									
				Middle									
				Right									
2				Left									
				Middle									
				Right									
1				Left									
				Middle									
				Right									

CHANNEL UNIT CODES		POOL FORM CODES		Embeddedness & Canopy Cover	
GL	Glide	B	Backwater Pool	0 = Absent (0%)	
RI	Riffle	F	Bluff Pool	1 = Sparse (<10%)	
RU	Run/Race	I	Impoundment	2 = Moderate (10-40%)	
PO	Pool	L	Lateral Pool	3 = Heavy (40-75%)	
RRX	Riffle-Run complex	M	Mid-Channel Pool	4 = Very Heavy (>75%)	
PGX	Pool-Glide complex	O	Obstruction Pool	(Canopy within 1m on each side of transect)	

Transects are equally spaced as determined by dividing the reach length by 10.

*Transect 1 is located at the upstream end of the reach; Transect 11 is located at the downstream end of the reach

WENTWORTH SUBSTRATE CODES

1= <0.062 (silt/clay)

2= 0.062-0.125 (very fine sand

3= 0.125-0.25 (fine sand)

.

4

.

5



6



7



8



9



10



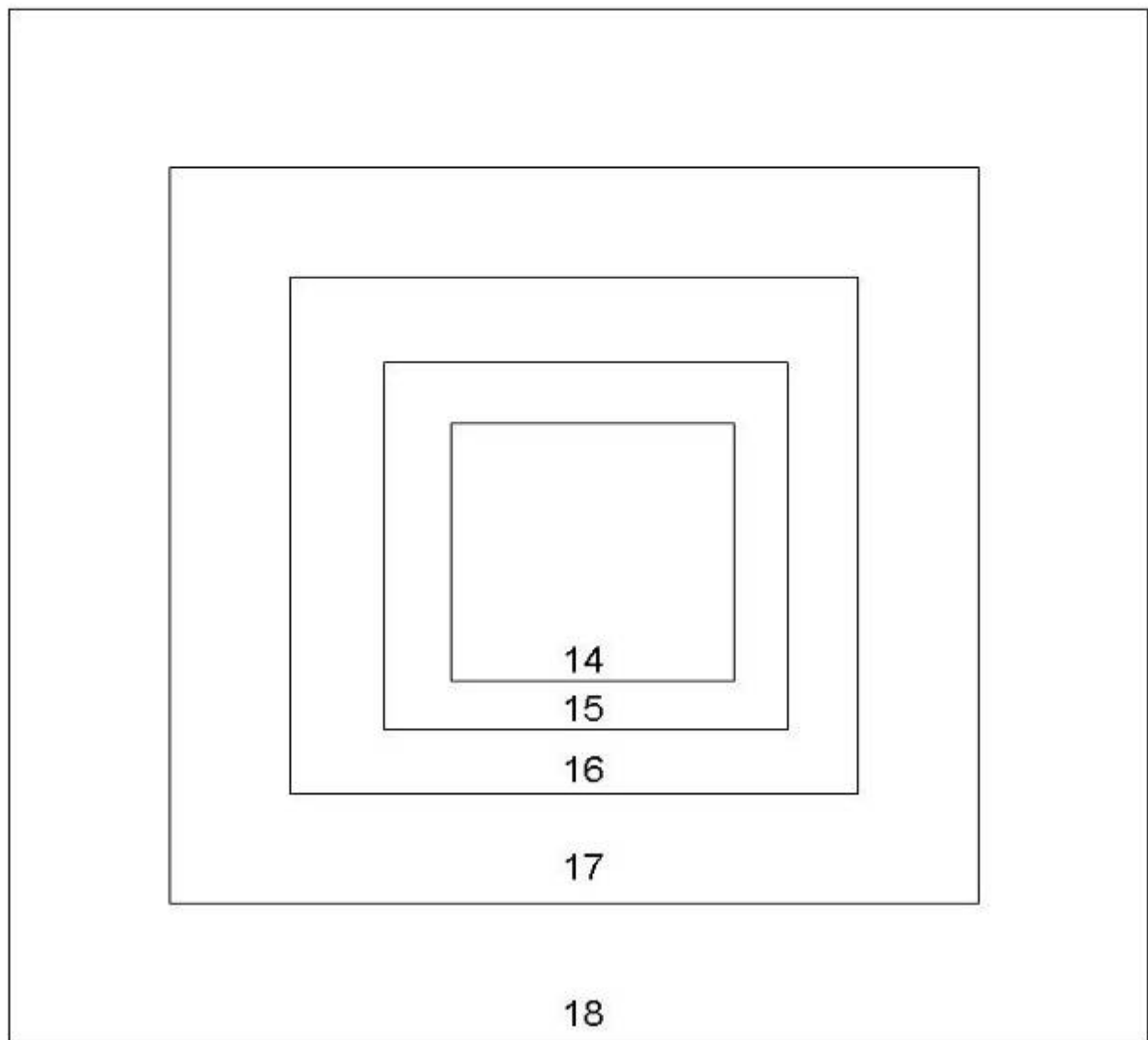
11



12



13



19=180-256 mm, **20**=256-362 mm, **21**=362-512 mm, **22**=512-1024 mm, **23**= >1024 mm, **24**=Bedrock

Fish Cover Form

Spring Name:

Date:

Crew:

Reach Length:

Transect Spacing Interval:

Fish Cover*										
Trans.	Circle all cover types present.								Comment	
11	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
10	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
9	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
8	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
7	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
6	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
5	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
4	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
3	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
2	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	
1	L	BO	AR	SWD	LWD	T/R	OV	UC	BL	
	M	BO	AR							
	R	BO	AR	SWD	LWD	T/R	OV	UC	BL	

Fish Cover Types* BO = Boulders AR = Artificial SWD = Small Woody Debris LWD = Large Woody Debris T/R = Trees/Roots OV = Overhanging Vegetation UC = Undercut bank BL = Bluff within 5m of water	Additional comments: BO, AR are assessed within a 1m² plot on transect SWD is < 10 cm in diameter at largest end; LWD is >10 cm at largest end SWD & LWD assessed on a 1 m belt along transect on left & right side of channel T/R, OV, UC, BL are assessed within 1 m on either side of transect along bank  10 cm or 0.1m
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Bank Measurement Form

Spring Name:

Date:

Crew:

Reach Length:

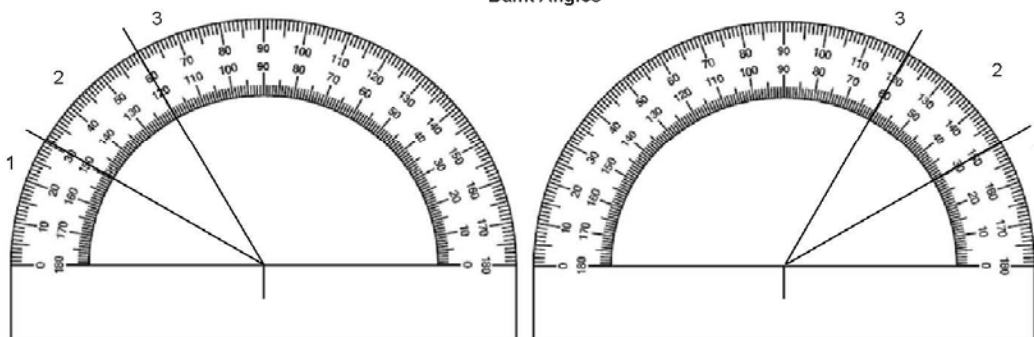
Transect Spacing Interval:

Trans.		Bank Stability			Bank Cover*					Comment
		Angle	Veg	Height	Sub	Circle	Dominant (>50%)	Cover		
11	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
10	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
9	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
8	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
7	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
6	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
5	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
4	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
3	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
2	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR
1	L					TR	SH	GR	BA	AR
	R					TR	SH	GR	BA	AR

*Bank cover is assessed within 1 m on each side of transect and 10 m up the bank from wetted edge

Bank Angle, Degrees	Vegetative Cover (%)	Height (m)	Dominant Substrate	Bank Cover Types*
1 = 0 - 30	1 = >80	1 = 0-1	1 = Bedrock/Artificial	TR = Large trees (> 3 in. dbh)
2 = 31-60	2 = 50-80	2 = 1.1-2	2 = Boulder/Cobble	SH = Small trees and shrubs
3 = >60	3 = 20-49	3 = 2.1-3	5 = Silt	GR = Grass and Forbs
	4 = <20	4 = 3.1-4	8 = Sand	BA = Bare rock/sediment
		5 = >4	10 = Gravel/Sand	AR = Artificial

Bank Angles



**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 10: Measuring Spring Discharge

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP is guidance for measuring discharge in springs at OZAR. The SOP describes sampling procedures, calibration, and general maintenance procedures. If other meters are used, field personnel should review the instruction manual for instrument-specific guidance on how to calibrate and operate those particular meters.

I. Background Information

Velocity and depth are measured using a current meter attached to a wading rod. The rod allows for quick and easy measurements of depth with incremental markings, and has an adjustable arm that places the current meter at the proper depth for measuring velocity (60% of the depth from the surface of the water; Carter and Davidian, 1969). Some current meters have rotating cups (Pygmy and Price models) while others have a pair of electronic contacts on a small head (FLO-MATE 2000) to measure velocity. The sensor in the Marsh-McBirney FLO-MATE 2000 is equipped with an electromagnetic coil that produces a magnetic field. A pair of carbon electrodes measure the voltage produced by the velocity of the conductor, which in this case is the flowing water. Internal electronics process measured voltages and output them as linear measurements of velocity. Velocity is displayed as either feet per second or meters per second.

Spring discharge (Q) is the volume of water passing a cross-section per unit of time and is generally expressed in cubic feet per second (ft³/s) or cubic meters per second (m³/sec). Discharge is estimated by multiplying current velocity by the cross-sectional area (Carter and Davidian, 1969). Cross sectional area is determined by first measuring the width of the stream channel. The cross section is then divided into smaller increments (usually 15 to 20 intervals) and depth and velocity are measured at each increment. The depth and width of the interval are multiplied to get an area for each interval and then each interval area and velocity is multiplied to produce a discharge for each interval. These discharges are summed to produce a total discharge for that cross section of the stream. This process will be described in greater, step-by-step detail in the “Procedures” section.

II. Prior to the Field

- A. Standard wading rods come in both metric and English standard units (feet). Discharge measurements are generally recorded in English units as cubic feet per second. Whatever units are used, ensure that there is consistency between the settings on the velocity meter, the wading rod, and the tape measure, and that the units are clearly recorded on the data sheet. English standard units are easily converted to metric units when required.
- B. Ensure new batteries are placed in units that require them.
- C. Calibrate velocity meters (FLO-MATE 2000 or USGS pygmy) according to instructions in the manufacturer's operations manuals. Photocopies of the operations manuals should be taken to the field.
- D. Equipment maintenance and storage should follow guidance issued by the manufacturers.

III. In the Field

Discharge measurements require wading across the stream and may stir up sediments, disrupting accurate measurement of other parameters.

Quantitative Discharge Procedure:

- A. Prior to taking any measurements, the location where discharge will be measured must be determined. An ideal cross-section in the sample reach will have the following qualities:
 - 1. The stream channel directly above and below the cross-section is straight.
 - 2. There is measurable stream flow, with a stream depth preferable greater than 10 cm and velocities generally greater than 0.15 meter/second.
 - 3. The streambed is a uniform "U" shape, free of large boulders, woody debris, and dense aquatic vegetation (although this may be difficult in spring-runs where vegetation is dense).
 - 4. The stream flow is laminar and relatively uniform with no eddies, backwaters, or excessive turbulence.

Note: The cross section will not likely meet all these qualifications but the best location should be selected based on these standards. Record (or draw a diagram) on the data sheet a description of any discrepancies with the cross section.

- B. Once the cross section is established, measure the width of the stream with a tape measure to the nearest 0.1 meter and secure the tape across the stream for the duration of the discharge measurement.
- C. Divide the stream into equal intervals across the width of the cross section, usually 15 to 20. A minimum of 10 intervals is recommended. A velocity and depth measurement will be recorded for each interval across the stream at the center of each interval. For example, if the stream is 10 meters wide, 10 velocity and depth measurements will be taken at one meter intervals. The first measurement will be taken a half meter from the water's edge, the second will be taken at 1 ½ m from the water's edge, etc., as shown in Fig. 1.
- D. Attach the sensor to the wading rod and ensure that the sensor is securely screwed onto the rod and facing upright.

10 m wide cross section—10 m intervals

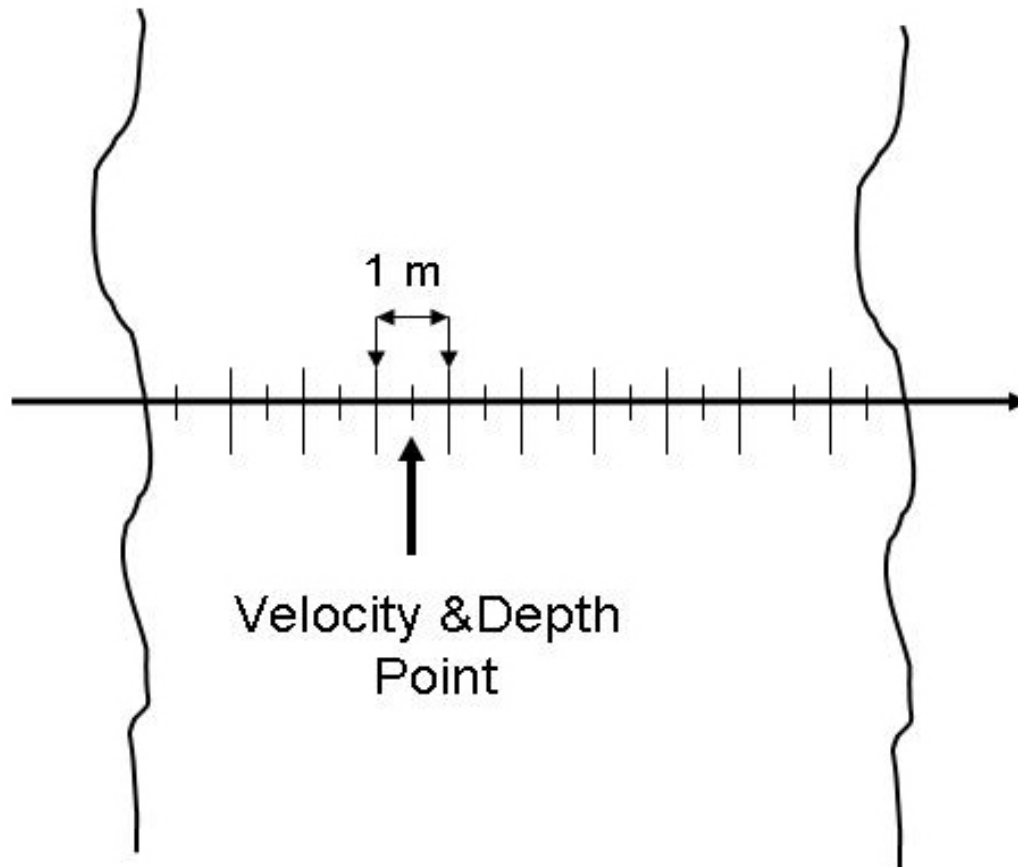


Figure 1. Cross section diagram.

- E. One person should measure discharge and one person should remain on the bank, recording data. The first readings are taken at water's edge and are recorded as

depth=0 and velocity =0. Proceed to the next interval and record readings. Place the wading rod as level as possible and hold perpendicular to the water level. Read depth from the wading rod to the nearest centimeter. The rod will have graduated marks along its length, with single marks indicating two centimeters, double marks indicating 10 centimeters, and triple marks indicating one-half meter increments.

- F. Once depth has been read, adjust the arm of the sliding rod with the sensor attached to 60% of the water depth. The wading rod will place the sensor at 60% of the depth from the surface of the water when properly adjusted.

Note: For example, if the depth is 2.6 meters, line up the 2 on the meter scale (sliding rod) with the 6 on the tenth scale (increments on handle of fixed rod). The sensor is now located at 60% of the water depth.

- G. Stand behind the sensor and make sure there is no disturbance (including the sensor cord) around the sensor that interferes with the velocity measurement. The meter may be adjusted slightly up or downstream to avoid boulders or other interferences.

Note: Make sure the sensor directly faces the flow of the water. This may not always be directly parallel with the water's edge; the rod and sensor may need to be turned slightly with each measurement.

- H. Allow the instrument enough time to get an accurate reading--generally around a minute. Watch the time bar complete two full cycles and then take the velocity reading. If something happens during the measurement, such as accidental movement of the wading rod, the reading should be repeated.

- I. Call out the distance from the water's edge, the depth, and then the velocity to the person recording data. Continue moving across the stream until measurements have been taken at all intervals.

Note: If the water velocity increases greatly between intervals, additional measurements can be taken to shorten the width of the intervals within this area of high velocity. Be sure to change the interval width for these measurements in the calculation of discharge.

- J. When finished, detach the sensor from the wading rod and place it back in the mesh side pocket for transportation. If you do not expect to use the meter for several days, turn the meter off, clean the sensor, and store properly.

SPRING DISCHARGE

SPRING NAME: _____ DATE: _____

CREW MEMBERS: _____

Taken at upper end of sampling area

	Distance Units ft ☐ m ☐	Depth Units ft ☐ cm ☐	Velocity Units ☐ ft/s XX.X ☐ m/s X.XX	
	Start at River Right; Final measurement should be at river left bank			
	Distance from Bank	Depth	Velocity (@0.6 depth)	FLAG
1	0	0	0	
2				
3				
4				
5				
6				
7				
8				
9				
10				
11				
12				
13				
14				
15				
16				
17				
18				
19				
20				

Flags:

Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri. Heartland I&M Network and Prairie Cluster Prototype Monitoring Program

SOP 11: Data Management

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP explains procedures for data management of Ozark National Scenic Riverways spring community (fish and macroinvertebrate) sampling data. It includes a general description of the data model and procedures for data entry, data verification and validation, and data integrity for the primary fish and macroinvertebrate data.

Data management can be divided into (a) the initial design phase that involves defining the data model, its entities and their relationships and (b) the procedures necessary to implement the database. Microsoft (MS) Access 2003 is the primary software used for maintaining fish and macroinvertebrate community data. Water quality data will be stored in the National Park Service's NPStoret database. Environmental Systems Research Institute (ESRI) ArcInfo 9.x is used for managing spatial data associated with field sampling locations. Data products derived from this project will be available at the NPS I&M Data Store and EPA Storet National Data Warehouse. QA/QC guidelines in this document are based on recommendations of Rowell *et al.* (2005) and citations therein.

I. Data Model

The NPS I&M program has designed the Natural Resource Database Template (NRDT) to be used as a database model for storing vital signs monitoring data in MS Access (NPS 2006). The template has a core database structure that standardizes location and observation data to facilitate the integration of datasets. Developed in MS Access, the database allows users to enter, edit, display, summarize, and generate reports as well as integrate with other Natural Resource data systems such as NPStoret.

Note: The NRDT data dictionary follows standards identified in the NRDT phase 2 and is modified where required. Naming standards follow I&M recommended procedures.

The aquatics program relies on distributed databases to allow remote users to enter data and is accomplished via database replication consisting of a design master and replicas. The design

master is stored on the server at Missouri State University and can be directly accessed for local data entry/revisions or design changes. A replica is created for users without access to the server and is distributed for data entry activities. When a user has finished data activities with the replica, it is returned and synchronized with the design master. NPS Water Resource Division (WRD) has also designed the NPStoret database to facilitate archiving NPS data in the EPA Storet database. NPStoret is a series of Access-based templates patterned after the NRDT and includes data entry templates and an import module. It supports the core data management objectives of data entry and verification/validation in a referentially constrained environment (*i.e.*, related locations, events, and primary data elements (NPS/WRD 2007).

A generalized NRDT entity relationship diagram of the OZAR spring community database is given in Fig. 1. The sampling events (*tbl_SamplingEvents*) and locations (*tbl_Locations*) tables are the two core tables and contain general information pertaining to the field sample occasion (the when and where of the sample). This includes information such as date and time, spring name, UTM coordinates, and park/project codes. Detailed information pertaining to the community sampled is maintained in *tbl_Count* (macroinvertebrate count data) and in *tbl_FishCommSppIndiv* (*tbl_FishCommunitySampling* serves as the organizing hub for fish data). Other tables include habitat data (*e.g.*, substrate, discharge), and associated lookup tables (*e.g.*, Wentworth substrate codes, taxonomic data).

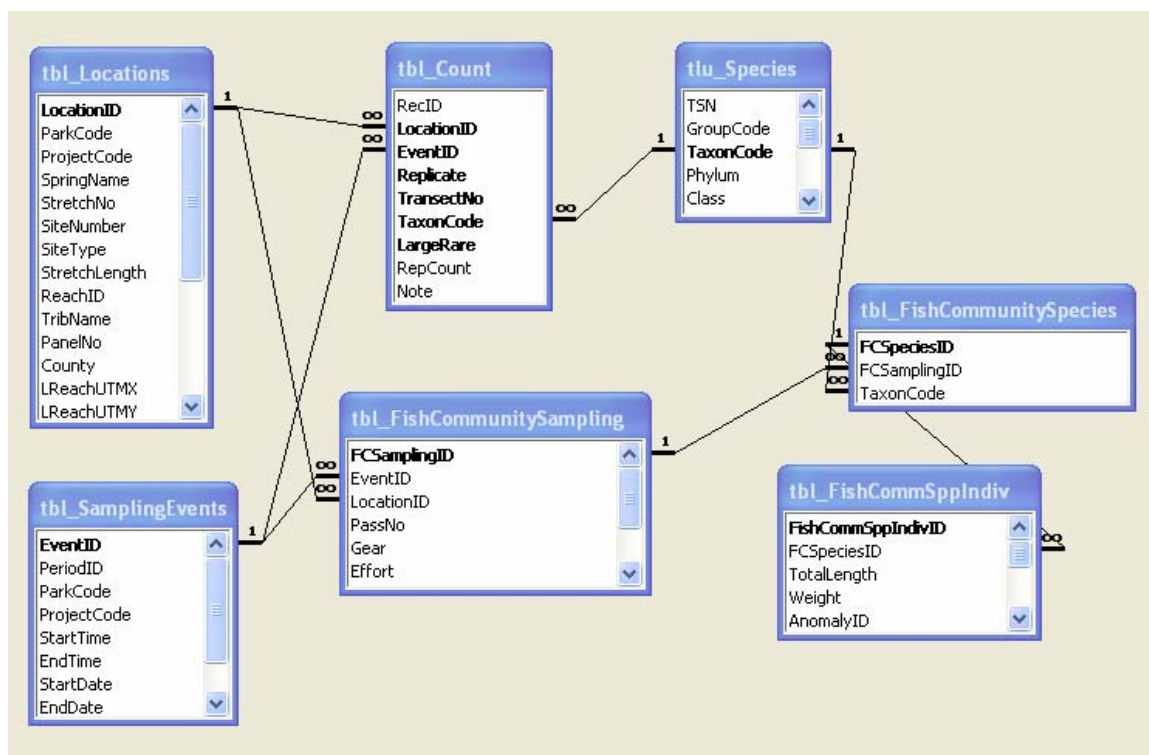


Figure 1. Data model for OZAR spring community monitoring database. Note *tbl_Locations* tracks GPS spatial and non-spatial field data while *tbl_SamplingEvents* tracks events. The data entry forms (below) follow these two levels of information organization.

II. Data Preparation

Quality assurance and quality control procedures related to data recording are important components of any project. Sampling data (*i.e.*, sample methods, effort, weather/water quality conditions, and species abundance data) are recorded and checked for completeness either before leaving a site or within 24 hours of data recording. This will aid in verification and validation of the data after entry into the database. To prevent the complete loss of field form data due to unforeseen circumstances (*i.e.*, fire or flood in the workplace), all field sheets are photocopied and a hard copy located in a separate location as the original. Field sheets are scanned into a computer and electronic copies of the data sheets stored on the HTLN server located at Missouri State University, Springfield, MO. This will ensure that at least one copy of the field sheets is available for data entry and verification.

III. Data Entry

Data entry is accomplished in either the design master or replica databases using a tiered set of forms. Upon opening the database, the user is presented with a switchboard (Fig. 2). A preliminary set of forms define the sampling occasion and requires the input of period and event IDs *prior* to entry of additional data. The other forms address the details of fish or macroinvertebrate occurrence, habitat, observers, etc. and have data entry instructions. Once all fields for the preliminary set of forms have been completed, data can be entered for the remaining forms. Additional forms document sampling personnel for each occasion and their specific hours related to the project (sampling hours, travel hours, *etc.*). When using replicas for data entry, the replica is synchronized with the master database so that new data are added. Note: The “Prevent Deletes” option is enforced in replica databases to ensure data are not inadvertently deleted.

Several features are “built-in” to form properties that enable the user to maximize data entry efforts while minimizing error. These include data input masks for ease of viewing multi-part data (*i.e.*, park/project codes and date in PeriodID or EventID), “fill-in-as-you-type” to automatically complete a field, limiting input values to known ranges (or restricting null values) or providing “drop-down boxes”, highlighting data entry boxes being edited, and tab indexes to control the order of data entry. Forms also contain fields that require data input and/or are constrained to properties and integrity of related tables.

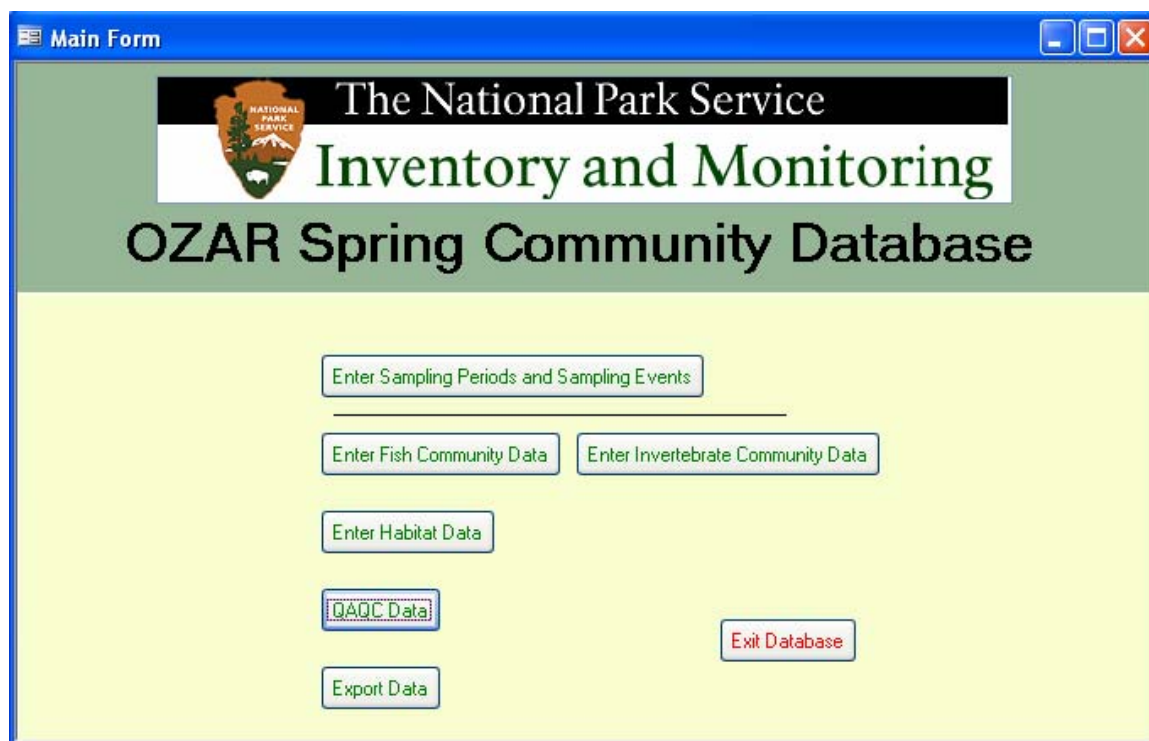


Figure 2. Spring community database switchboard. Note the primary data entry form button (top).

Entering Sampling Occasion Data

Procedure: Sampling occasion data

- A. Open the Sampling Period form (Fig. 3) and use the calendars to select the start and end dates of the sampling period time frame to develop a PeriodID.
- B. Clicking the “Generate Sampling PeriodID” button passes the user data to a secondary form where the user enters the Season and accepts (or revises) the Protocol Version and then clicks the “Verify” button.

PeriodID Data Entry Form

1. Select Start Date and End Date

Start Date

Jul 2007

Sun	Mon	Tue	Wed	Thu	Fri	Sat
24	25	26	27	28	29	30
1	2	3	4	5	6	7
8	9	10	11	12	13	14
15	16	17	18	19	20	21
22	23	24	25	26	27	28
29	30	31	1	2	3	4

End Date

Jul 2007

Sun	Mon	Tue	Wed	Thu	Fri	Sat
24	25	26	27	28	29	30
1	2	3	4	5	6	7
8	9	10	11	12	13	14
15	16	17	18	19	20	21
22	23	24	25	26	27	28
29	30	31	1	2	3	4

2. Generate a PeriodID

PeriodID Data Entry Form

Park Code

Project Code

Period ID

Start Date

End Date

These data cannot be modified

1. Enter the Season
2. Enter the Protocol Version
3. Click the "Verify" button

Season

Protocol Version

(required)

Figure 3. Sampling period forms for spring community database.

- C. Open the Sampling Events forms (Fig. 4), select the appropriate PeriodID, then use the calendars to select the start and end dates of the sampling event to develop an EventID.
- D. Clicking the "Generate Sampling EventID" button passes the user data to a secondary form where the user enters additional event data.

EventID Data Entry Form

1. Choose PeriodID

Period ID:

2. Select Start Date and End Date

Start Date

Jul 2007

Sun	Mon	Tue	Wed	Thu	Fri	Sat
24	25	26	27	28	29	30
1	2	3	4	5	6	7
8	9	10	11	12	13	14
15	16	17	18	19	20	21
22	23	24	25	26	27	28
29	30	31	1	2	3	4

End Date

Jul 2007

Sun	Mon	Tue	Wed	Thu	Fri	Sat
24	25	26	27	28	29	30
1	2	3	4	5	6	7
8	9	10	11	12	13	14
15	16	17	18	19	20	21
22	23	24	25	26	27	28
29	30	31	1	2	3	4

3. Click to generate an EventID

EventID Data Entry Form

Park Code: Project Code:

Period ID:

Start Date: End Date:

Event ID:

These data cannot be modified.

1. Enter Start and End Time
2. Choose Recorder
3. Enter any comments
4. Click "Close Form" to exit the form

Start Time: End Time: Recorder:

Additional Comments:

Figure 4. Sampling event forms for spring community database.

After inputting sampling occasion data (periods and events), the user can begin to enter additional spring community data. The following demonstrates data entry for the additional data and can be used in any order.

Procedure: Fish community data

- A. Click the "Enter Fish Community Data" button on the main switchboard to open the fish community data entry form (Fig. 5).

- B. The appropriate LocationID, EventID, and Gear are selected and Pass Number, Effort, Seine Reach Length and Number of Seine Hauls are entered specifically for that sampling occasion.
- C. Click the “Fish ID Crew” button and select the observer and duty (identifier or recorder). Click the “Fish Shocking Crew” button and select the observer and duty (shocker, seiner, *etc.*). Clicking the “Continue” button in either form will allow the addition of multiple entries and clicking the “Return” button will allow the user to exit the form.
- D. The user then selects species information by selecting the species name and entering additional data specific to the one species. A new species can be added to the taxa lookup table if necessary.

Species data are comprised of three types: regular fish, batch weighed fish, and extra fish data. Regular fish data consist of 30 subsampled individuals of each species that were measured (total length), weighed, and any anomalies noted (Anomaly Description is set to default to N, no anomaly). After selecting a scientific name, the user enters total length and weight and selects Anomaly Description (if required). If the individual was retained (*i.e.*, saved for later, preserved) the Voucher box is checked. Number refers to the number of individuals and defaults to one. The additional batch fields are ignored (*i.e.* tabbed through) and any comments to that individual fish are noted. The user is then returned to the Total Length field and continues entering data for that species. Batch weighed fish consist of those smaller species that were measured (total length) and weighed in groups. Similar to regular fish data, the user selects a scientific name and enters the total length. Weight is left blank and any anomaly for that specific individual is documented by selecting the Anomaly Description and, if retained, the Voucher box checked. As in the regular fish, Number defaults to one. The Batch Weight box is checked for a BatchID (1, 2, 3, *etc.*) and the weight of those batch weighed fish is entered in Batch Weight. For subsequent entry of batch weighed fish the Batch Weight box is left unchecked, BatchID is set to that batch number, and Batch Weight defaults to zero. Any comments to that individual fish are noted and the user is returned to the Total Length field to continue entering data for that species. Extra fish are those collected and merely counted. The user selects a scientific name and enters the number collected in the Number field and any comments for those extra fish (other fields are ignored). When all fish species data are entered for that fish community the user clicks the “Close Form” button to close the form.

Fish Community Species Data Entry Form

Enter entire data sheet before closing form!

Location ID: Big Spring

Event ID: OZAR_SPRNGS_2007-JUL-25

Gear: seine

Fish ID Crew:

Fish Shocking Crew:

Pass #: 1 Effort (total seconds): 2400 **required**

Seine Reach Length: 0 # Seine Hauls: 0

Species

Scientific Name: Ozark madtom

Individual

Total Length	Weight	Anomaly Description	Voucher	Number:
45	3	N	☐	1

Batch Weight? ☐ Batch ID: 0 Batch Weight: 0 **Note: Enter Batch Weight only once!**

Comments:

Record: 2 of 2

Record: 1 of 1

Add New Fish Taxa

Return to previous menu

Record: 1 of 1

Figure 5. Fish community data entry form.

Procedure: Invertebrate count data

- Click the “Enter Invertebrate Community Data” button on the main switchboard to enter invertebrate data.
- Select the appropriate LocationID, Transect #, EventID, and % Subsampled (Fig. 6) and click the “Open Count Form” button to enter occurrence data (Fig. 7).

Macroinvertebrate Occasion Data Entry Form

Spring Aquatic Invertebrate Enumeration Form

1. Select LocationID, Transect #, EventID
2. Change % Subsamped (if required)
3. Click "Open Count Form" to add count data
4. Click "Return to previous menu" to exit the form

LocationID: Big Spring
 Transect #: 2
 EventID: OZAR_SPRNGS_2007-JUL-25
 % Subsamped: 25

Replicate: ☐ L

Open Count Form **Return to previous menu**

Figure 6. Preliminary data entry form for macroinvertebrate counts. Data are selected from drop down boxes and upon clicking the “Open Count Form” button are passed to the macroinvertebrate count data form (Fig. 7).

- C. Select the Taxa from the drop down box. Upon selection, the TaxonCode remains visible and the remaining values are hidden (inset of TaxonCode, Phylum, etc. added to demonstrate selection).
- D. After selecting Taxa, enter the count.
- E. Clicking the “Continue” button will prompt for the next Taxa.

Macroinvertebrate Data Entry Form

LocationID: OZAR_SPRNGS_SITE02
 Transect #: 2
 Replicate: L
 EventID: OZAR_SPRNGS_2007-JUL-25

These data cannot be modified.

1. Select Taxa and enter Count
2. Check Large/Rare (if required)
3. Click "Continue" to add more data
4. Click "Add Taxa" to add a taxon
5. Click "Return to previous menu" to exit the form

Taxa: Add Taxa **Taxa can be added at any time**

TaxonCode	Phylum	Class	Order	Family	Genus
CHLOHA	Arthropoda	Insecta	Plecoptera	Chloroperlidae	Haploperla
LEUC	Arthropoda	Insecta	Plecoptera	Leuctridae	Leuctra
LEUCLE	Arthropoda	Insecta	Plecoptera	Leuctridae	Leuctra
LEUCZE	Arthropoda	Insecta	Plecoptera	Leuctridae	Zealeuctra
NEMO	Arthropoda	Insecta	Plecoptera	Nemouridae	Nemoura
NEMODAM	Arthropoda	Insecta	Plecoptera	Nemouridae	Amphinemura
NEMOPR	Arthropoda	Insecta	Plecoptera	Nemouridae	Prostoia

Count:
 Large/Rare? ☐

Figure 7. Taxa count form for macroinvertebrate data.

Entering Habitat and Discharge Data

Habitat data are entered by clicking the “Enter Habitat Data” button on the main switchboard and selecting the appropriate button for habitat type (see Fig. 8). The user is then presented with selection boxes (LocationID, EventID, etc.) for the appropriate habitat type and this data is passed to subforms where specific replicate data is entered. When finished entering habitat data the user will be returned to the initial habitat form and exits the form by clicking the “Return to previous menu” button.

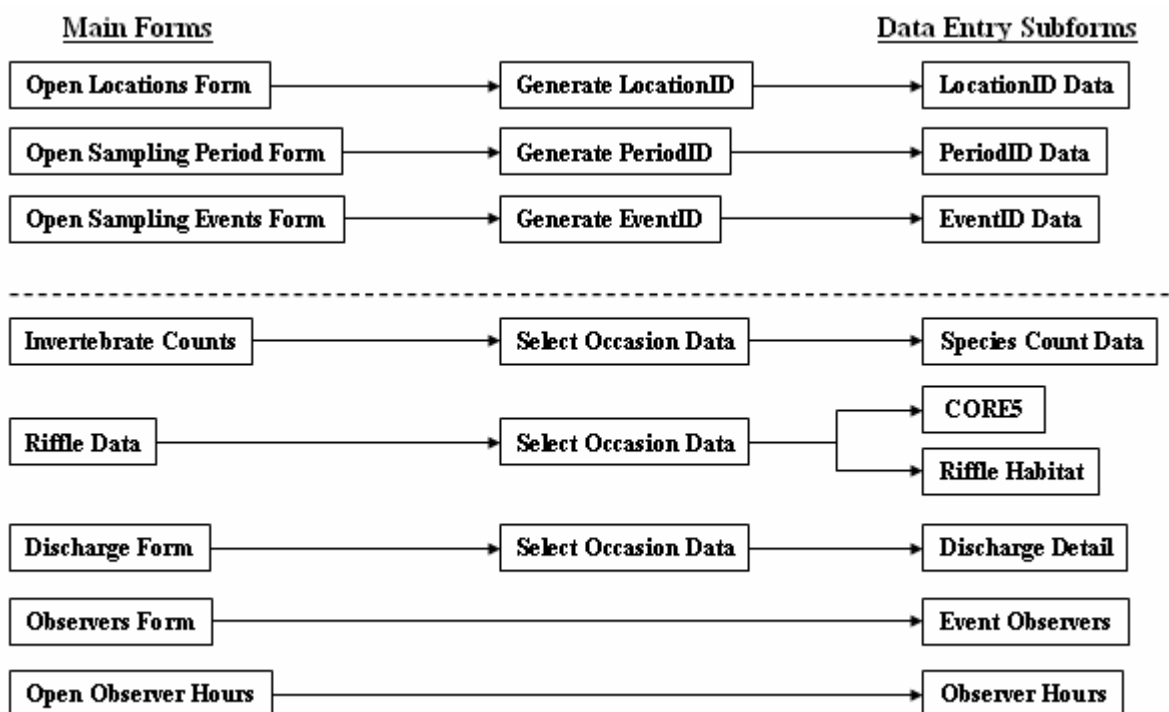


Figure 8. Outline of spring community data forms. Note: Forms are selected from the main switchboard and data entered into data entry subforms.

Entering Water Quality Data

Water quality data collected by unattended CORE 5 data loggers (sondes) are uploaded from the logger using the manufacturer’s accompanying software program and saved in MS Excel. Data are then edited to correct any missing data due to logger maintenance (down time) and validated to determine if the data meet the expected range requirements or critical limits. CORE 5 water quality summary data are then entered into NPStoret either by using the direct data entry templates or the import module. Metadata is then entered for each characteristic/parameter. Coordinate data for logger locations are collected in accordance with the current HTLN spatial data collection techniques and entered into NPStoret. An NPStoret database is then sent to the WRD staff on an annual basis for initial QA/QC and subsequent upload into the WRD master copy of the EPA STORET.

IV. Data Verification

Data verification immediately follows data entry and involves checking the accuracy of computerized records against the original source, usually paper field records. Data tables are queried to produce specific sets of data (*i.e.*, fish or macroinvertebrate data, habitat data, etc.) and exported to Excel worksheets (via the “QAQC Spring Community Data” button, Fig. 2). These worksheets can then be compared to the original source (*i.e.*, field data sheets) to identify missing, mismatched, or redundant records. The design master is then used to correct and/or delete errors via data edit forms. As data are edited, built in table triggers store the original record in a backup audit table and can be recovered where necessary (audit tables mirror the data table). The project manager then rechecks 10% of the records and, if errors remain, repeats steps 2-4 (below).

Procedure:

- A. Print pertinent data.
- B. Compare data with original field forms.
- C. Reconcile errors in database.
- D. Recheck 10% of records.

V. Data Validation

Although data may be correctly transcribed from the original field forms, they may not be accurate or logical. For example, field crews collect data per occasion (location and event) and the resident data should reflect this. At any given occasion a query of these data should reflect these conditions and confirm the coincident (*i.e.*, relational) nature of the database. Missing or mismatched records can then be corrected.

As annual data are amassed in the database, validation is conducted via query and comparison among years to identify gross differences. For example, species A may be recorded at a location this year, but not in previous years, thus representing a possible new locality. The design master is then used to correct and/or delete errors via data edit forms. Once verification and validation is complete, the data set is turned over to the Data Manager for archiving and storage. The data can then be used for all subsequent data activities.

Procedure:

- A. Query pertinent data to insure resident data match existing protocols for each parameter.
- B. Archive validated database.

Spatial Data Validation

Spatial validation of sample coordinates can be accomplished using the ArcMap component of ArcGIS. Coordinate data are maintained in the Access database and can be added to an ArcMap project and compared with existing features (*i.e.*, park boundaries, USGS Digital Orthophoto Quarter Quadrangles, National Hydrography Dataset hydrography, *etc.*) to confirm that coordinate data are valid.

Procedure:

- A. Develop testing project within ArcMap constrained to appropriate UTM zone and projection (15N, NAD83).
- B. Add park unit boundaries and any necessary spatial data (roads, water, contour, *etc.*).
- C. Add relevant site coordinate data to testing project and validate against known features.
- D. Correct mistakes, if necessary, and re-validate.
- E. Develop metadata for final spatial dataset.

VI. File Organization

The various databases, reports and GIS coverages used and generated by the Heartland Network create a large number of files and folders to manage. Poor file organization can lead to confusion and data corruption. As a standard data management technique, files pertaining to the project are managed in their own folder: Analysis, for data analysis; Data, for copies of archived data as well as data sheets; Documents, for supporting materials related to the project; and, Spatial info, for various spatial data. The database is managed in the Databases folder and contains prior versions of the database in a subfolder.

VII. Version Control

Prior to any major changes of a data set, a copy is stored with the appropriate version number. This allows for the tracking of changes over time. With proper controls and communication, versioning ensures that only the most current version is used in any analysis. Versioning of archived data sets is handled by adding a floating-point number to the file name, with the first version being numbered 1.00. Each major version is assigned a sequentially higher whole number. Each minor version is assigned a sequentially higher .1 number. Major version changes include migrations across Access versions and complete rebuilds of front-ends and analysis tools. Minor version changes include bug fixes in front-end and analysis tools. Frequent users of the data are notified of the updates, and provided with a copy of the most recent archived version.

VIII. Backups

Secure data archiving is essential for protecting data files from corruption. Once a data set has passed the QA/QC procedures specified in the protocol, a new metadata record is created using the NPS Metadata Tools (stand alone or within ArcCatalog) and/or Dataset Catalog. Backup copies of the data are maintained at both on- and off-site locations. An additional digital copy is forwarded to the NPS I&M Data Store. Tape backups of all data are made at regular intervals in accordance with current HTLN backup standard operating procedures and will be made minimally, once per week, with semi-annual tapes permanently archived.

Procedure:

- A. Create metadata record pursuant to data archiving.
- B. Backup data.
- C. Store backup copies on- and off-site and forward a copy to the I&M Data Store.
- D. Administer regularly scheduled backups of data.

IX. Data Availability

Currently, data are available for research and management applications for those database versions where all QA/QC has been completed and the data have been archived. Data can be transferred using ftp or by e-mail (where files are smaller than a few megabytes). Monitoring data will become generally available for download directly from the NPS I&M Data Store. Metadata for the spring community database are developed using ESRI ArcCatalog 9.x and the NPS Metadata Tools and Editor extension and will be available at the NPS I&M GIS server (<http://science.nature.nps.gov/nrddata>). Water quality data will be stored at the EPA STORET National Data Warehouse (EPA 2006) and be publicly accessible via the Internet. Additionally, data requests can be directed to:

Heartland I&M Network
Attn. Data Manager
Wilson's Creek National Battlefield
6424 W. Farm Road 182
Republic, MO 65738-9514
417-836-5313

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 12: Data Analysis

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

Conclusions of ecological studies based on biological, chemical, and physical data are used by resource managers to better comprehend underlying system processes and develop environmental/management policies that best serve the resource. A critical component of any long-term monitoring protocol is a consistent and systematic approach to analyzing and reporting data. This information must describe the current condition, or status, of a community, and be robust enough to detect community changes through time. This, in turn, could have substantial ecological repercussions and should be an important consideration for investigators responsible for data interpretation. Therefore, every effort should be made to collect reliable data and use statistical analyses that are straightforward and will result in confident interpretations. This SOP describes the metrics to be calculated for invertebrate, fish, and aquatic vegetation data collected from springs at OZAR, and statistical analyses for interpreting those metrics.

I. Metrics and Estimated Variables for Analysis of Aquatic Vegetation

Summary indices and variables will provide information to park managers on the status of the target communities. As additional data are collected, trend analysis using time-series analysis or repeated measures may be used to detect temporal trends in vegetation community data. Control charts also will be used to evaluate data trends (see below) when appropriate.

For analysis, transects are used as the sample unit of replication and secondary sample units (1 m² plots) are pooled or averaged. Once estimates for all parameters have been obtained for each transect, averages and a measure of variability (standard error of the mean) among transects are obtained for sampling reaches individual springs.

The following metrics will be used to assess the invertebrate communities of springs at OZAR.

Individual species abundances

Individual Species Frequency. Species frequency is reported as the proportion (or percentage) of plots in which the species occurs.

Percent Foliar Cover: This metric serves as an estimate of abundance. To calculate percent foliar cover, the cover class intervals are converted to median values to estimate percent cover for each species. Mean percent cover is then calculated as the species percent cover for each transect, averaged across all transects. Plots within a sampling unit with zero values for a species are excluded.

From these basic estimates of foliar cover and frequency are generated the following metrics for each transect (1) **species relative cover**, (2) **species relative frequency**, and (3) **species importance value**. Relative frequency and relative cover are calculated using the following formulas:

$$\text{Relative cover, species } i = \frac{\text{cover species } i}{\Sigma \text{ cover all species}}$$

$$\text{Relative frequency, species } i = \frac{\text{occurrences species } i}{\Sigma \text{ occurrences all species}}$$

Where occurrence is the number of plots a species is present within a transect. Species importance value (IV) is an index derived from relative cover and relative frequency, [*relative cover* + *relative frequency*)/2]*100]. The importance value gives an overall estimate of the influence or importance of a plant species in the community.

Plant species richness, diversity and evenness

Species Richness. This metric is determined as the total number of plant taxa recorded per transect and averaged for the sampling reach. Species richness (S) is calculated with all species (native and exotic) included in the estimate.

Shannon Diversity Index. Plant diversity for each transect in a spring is calculated using this index. For this index, the higher the score the greater is the degree of uncertainty that the next species will be the same as the previous one. It is calculated as:

$$H' = -\text{SUM}\{ p_i \cdot \ln(p_i) \}$$

or

$$H = - \sum_{i=1}^S p_i \log p_i$$

where p_i is the relative cover of species i (Shannon, 1948). Note: While any base may be used, the natural log (ln) is the most common.

Species Distribution Evenness. This metric (E') is a measure of distribution of species within a community as compared to equal distribution and maximum diversity. It is also known as Shannon's Evenness Index. It is calculated as:

$$E' = H' / H_{\max},$$

where H' is the Shannon diversity index and $H_{\max}$ is the maximum possible diversity for a given number of species if all species are present in equal numbers ((ln(species richness)).

Simpson's Index of Diversity. This metric estimates the likelihood that two randomly chosen individuals from a site will be different species and emphasizes common species. It is calculated using the complement of Simpson's original index of dominance:

$$D = \sum((n^2 - n)/(N^2 - N))$$

or

$$1 - \sum_i^n p_i^2$$

where n = number of individuals of i th species, and $N = \sum n$.

Effective Number of Species. Shannon and Simpson's diversity index values are converted into effective number of species for each community (H_e and D_e , respectively). This allows for both diversity measures to be compared directly to species richness of the sites (S) within and among sample years based on count of distinct species in the community. Shannon diversity index was converted into effective number of species (H_e) using the following formula:

$$H_e = e^H$$

or

$$H_e = \exp^H$$

where H is the Shannon diversity index value.

Effective number of species based on Simpson's diversity index (D_e) is the inverse of the index value or:

$$D_e = 1/(1-D)$$

where D is the Simpson's diversity index value.

Dominance takes into account the species abundance and evenness of species distribution in the community. The degree of species dominance in the community is reflected by the degree to which $S > H_e > D_e$ when evenness (E) remains constant in a single community. The difference in number of species between the diversity measures reflects both how each metric considers uncommon species and how species diversity is partitioned within the community among years. If all species occurred in equal abundance in the community within and among sample years then $S = H_e = D_e$. Effective number of species for each diversity measure reflects the number of species found in a similar community when all species occur in equal density. For example, if $S = 100$ and D_e is equal to 20, then the community is dominated by 20 species and 80 species occur in low abundance. Such a community would be equivalent to a community with just 20 species all occurring in equal abundance.

Ratio of Exotic to Native Taxa. Exotic species have the potential to disrupt the aquatic vegetation community in OZAR springs, this metric will serve as an important gage of their relative frequency and abundance. It is calculated as the simple ratio of the number of exotic species to number of native species for a transect and averaged across the sample reach.

Other Approaches

The simplest, yet perhaps most important method of long-term monitoring data analysis is simply graphing data through time with an estimate of variability. Trend is easily inferred from a graphical presentation. Graphs are easily interpretable, and, as such, serve as useful tools for interpreting monitoring results to managers. The repeated measures linear regression model is also appropriate because the analysis allows for the significance of trends generated by the sequential sampling of plots to be tested (Lesica and Steele, 1996). The repeated measures linear regression model is based on differences in mean values among sample units (*e.g.*, transects) between time periods.

II. Metrics and Estimated Variables for Analysis of Aquatic Invertebrates

Summary indices and variables will provide information to park managers on the status of the target communities. As additional data are collected, trend analysis using time-series analysis or repeated measures may be used to detect temporal trends in invertebrate community data. Control charts also will be used to evaluate data trends (see below) when appropriate.

For analysis, transects (one 0.25 m² sample per transect) are used as the unit of replication. Once estimates for all parameters have been obtained for each transect, averages and a measure of variability (standard deviation or standard error of the mean) among transects are obtained for individual springs.

Multi-metrics

The following metrics will be used to assess the invertebrate communities of springs at OZAR.

Taxa Richness. This metric is a measure of the total number of genera or other designated taxonomic level of identification. Taxa richness generally increases with improving water quality,

habitat diversity, or habitat suitability (Rabeni *et al.*, 1997). Calculate as the total number of genera or other designated taxonomic level of identification.

EPT Richness. This metric is a measure of the number of genera within the aquatic insect orders Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies). Members of these orders are generally considered pollution-sensitive organisms and a decrease in their total number may indicate impairment. Calculate as the total number of different genera within the aquatic insect orders Ephemeroptera, Plecoptera, and Trichoptera.

Ephemeroptera Richness. This metric is a measure of the number of genera within the aquatic insect order Ephemeroptera (mayflies).

Ratio of Total Ephemeroptera Abundance /Total Abundance. This metric is calculated as the total Ephemeroptera density (number/m²) divided by the total density of the sample.

Shannon's Taxa Diversity Index. A variety of community diversity and similarity indices have been used in biological monitoring of water quality including Shannon's index, Simpson's index, the Coefficient of Community Loss, Jaccard Coefficient, and Pinkham-Pearson Community Similarity index (Resh and Jackson, 1993). The formula for calculating this statistic is shown above, but with the distinction where p_i = relative abundance of each species, calculated as the proportion (decimal fraction) of individuals belonging to a given species.

Taxa diversity generally decreases with declining water quality because of reductions in both richness and evenness.

Biotic Index (BI). The basis of the BI is that each taxon is assigned a pollution tolerance value related to its assumed or known tolerance of water quality degradation (Hilsenhoff, 1982, 1988). Tolerance values for taxa occurring at OZAR will follow Hilsenhoff (1982, 1988) and Lenat (1993) and are presented in Table 1 of this SOP. The BI increases with increasing impairment.

Calculate as:

$$BI = \sum (X_i T_i) / N$$

X_i = number of individuals within each genus

T_i = tolerance value of that species

N = total number of organisms in the sample

Ratio of Shredder Abundance/Total Abundance. This metric is calculated as the total density of the shredder functional feeding group (number/m²) divided by the total density of the sample.

Percent Dominant Taxa. This metric provides some insight into the evenness (relative abundance of the taxa to one another) of an invertebrate community. As water quality declines, a few taxa benefit from the disturbed conditions and increase dramatically in number. Most other taxa decline in abundance. For this protocol, percent dominance is based on the three most abundant taxa because none of the springs at OZAR currently appear to be grossly polluted to the point where they would be totally dominated by a single taxon.

Calculate as:

$$\text{Percent Dominance} = (D_1 + D_2 + D_3) / N * 100$$

D_1 , D_2 , and D_3 = abundances of the first, second, and third most numerous taxa in a sample
 N = total abundance of invertebrates in that sample.

Percent Intolerant. The tolerance values given in Lenat (1993) will be used here and they are presented in Table 1 below. Many of the taxa present in the springs at OZAR are the same as those presented in Lenat (1993). Tolerance values for species not addressed by Lenat (1993) are presented as the mean tolerance value for all species in the genus. Genera occurring in the springs at OZAR not listed by Lenat (1993) are not assigned tolerance values until their specific tolerances can be determined. The basis of this metric is that as the level of pollution increases, the number of pollution intolerant species should decrease. Calculate as a percentage of the number of specimens in the sample considered intolerant. Only those taxa with a tolerance value of zero or one are used to calculate this metric.

Parameter estimation with confidence intervals

Constructing confidence intervals around a parameter estimate is relatively straightforward, and covered in most elementary statistics texts. The primary consideration in constructing confidence intervals is that one must use the appropriate formula(e) for the particular distribution (*i.e.*, normal, Poisson) from which the data are obtained. Otherwise, one may transform the data; Kutner *et al.* (2005) discuss different data transformations and the situations where each may be appropriate. Distribution-free confidence intervals may also be constructed, although these will usually be relatively wide and more difficult to calculate (Conover, 1999).

In the simplest presentation of data, each parameter should be estimated in each year that data are available, and confidence intervals calculated. Confidence intervals in such cases are usually 90-95%, but could assume some other value, depending upon the amount of uncertainty that can be tolerated. Probabilistic thresholds other than confidence intervals (*e.g.*, McBean and Rovers, 1998) could also be used, if these provide more appropriate information. Finally, rather than some statistically based estimate of uncertainty, one could simply plot the range of variability observed for each parameter.

Other methods

If a hypothesis testing approach is deemed appropriate, many tests may be employed, depending upon the question being asked and the structure of the data. For example, a Kruskal-Wallis ANOVA may be used to test for significant differences among transects using metric scores from each of the pseudoreplicates. If there is reason to compare more than two variables among samples, Friedman's non-parametric two-way analysis of variance should be used. Alternatively, Cochran's Q is particularly useful for measuring changes in frequencies (proportions) across time.

Quantitative Similarity analyses can be performed on each possible pairing of samples to produce a similarity matrix. Then spatial and temporal analysis with previous years data can be done using a Mean Similarity Analysis (Van Sickle, 1997) or Wilcoxon's matched pairs test.

A Detrended Correspondence Analysis (DCA) and other multivariate techniques may be used to look for spatial relationships. This produces a two-dimensional graph in which samples of similar taxonomic composition are near each other and dissimilar entities are far apart. Equal distances in the ordination correspond to equal differences in species composition. Samples from the previous years can be compared over time and samples can be compared from different locations in the same year. In addition, environmental gradients can be inferred from the species composition data by performing a Spearman rank correlation between the rankings of the sample scores on the first two axes and rankings of the environmental variables (water quality and habitat data).

III. Metrics and Estimated Variables for Analysis of Fish Communities

Primary approaches to analyzing fish data from OZAR springs will include metric estimation/calculations with use of control charts and multivariate statistics. Biological metrics are commonly used by investigators at all levels (*e.g.*, private, state, tribal, and federal) to compare the condition of the biological community at multiple sites (Simon, 1999) or examine trends across time. Barbour *et al.* (1999) define a metric as a characteristic of the biota that changes in an expected direction with increased anthropogenic disturbance. Using these characteristics (Table 2) allows investigators to determine the importance of environmental conditions, clarify which habitat factors play a large role in shaping fish communities, and identify specific sources of impairment (Karr, 1981).

By combining multiple metrics (and results for those metrics) into a single index of biotic integrity (IBI), investigators can determine the overall quality of the fish community. An IBI can also be used to compare overall ecological conditions over time and among sites, providing the selected metrics are related to variables responsible for impairment (Karr, 1981; Barbour *et al.*, 1999; Simon, 1999). To assess fish community condition, the IBI developed for Ozark Highland streams by Dauwalter *et al.* (2003) will be evaluated for use at springs in OZAR.

Metrics

Species Richness. The number of species collected for the entire sample reach. Typically, species richness declines with increases in human disturbance.

Simpson's Diversity Index. This index uses both richness and abundance to calculate diversity of the fish community. This index is preferable to the Shannon diversity index and will be used for data analysis because this index is independent of sample size. This index decreases with poor water quality and habitat conditions. Simpson's Diversity Index is calculated with the formula shown above.

Catch per Unit Effort. Catch per unit effort (CPUE) can be calculated as either Catch per Time or Catch per Area sampled. When using electrofishing gear to collect community data, CPUE is typically calculated as Catch per Time. We will use catch per minute to obtain relative abundance. Total CPUE will be calculated using the total number of fish collected at a reach. CPUE for each individual species will also be calculated at each reach.

Size Structure. Size structures of fish populations within the community can be indicative of a disturbance or resource problem. A community with primarily larger fish indicates that there is little recruitment to keep the community self-sustaining. A community with primarily smaller fish

could indicate inadequate resources (*e.g.*, food resources) that limit growth. Average length and weight (and ranges) for each species at the reach will be calculated.

Percent Composition. Percent composition by biomass will be calculated for each species in the reach. To calculate this:

First, calculate individual biomass for each species (B_i):

$$B_i = W_i N_i$$

W_i = average weight of fish species i

N_i = number of individuals of species i

Second, calculate biomass per area for each species (BPA_i) and total biomass per area (BPA_t):

$$BPA_i = B_i / A$$
$$BPA_t = \sum BPA_i$$

A = area of the sample reach

Lastly, calculate percent composition for each species (C_i)

$$C_i = BPA_i / BPA_t * 100$$

Index of Biotic Integrity. The IBI developed by Dauwalter *et al.* 2003 will be evaluated for use in springs at OZAR. This IBI consists of seven metrics that are calculated and scored. Individual metric scores are totaled for an IBI score at that sample reach. The metrics are: (1) percent of individuals as algivorous/herbivorous, invertivorous, and piscivorous; (2) percent of individuals with black spot or an anomaly; (3) percent of individuals as green sunfish, bluegill, yellow bullhead, and channel catfish; (4) percent of individuals as invertivorous; (5) percent of individuals as top carnivores; (6) number of darter, sculpin, and madtom species; (7) number of lithophilic spawning species. Specifics on calculation of and scoring of metrics can be found in Dauwalter and others (2003).

Multivariate Analysis

Multivariate analysis is another frequently used analysis technique and involves methods used to explain variability in community data and to identify the environmental variables that best explain, and have an assumed responsibility for, the variability measured (Gauch, 1982; Jongman *et al.*, 1995; Everitt and Dunn, 2001; Timm, 2002). Multivariate techniques elicit a hypothesis from the biological data rather than disproving a null hypothesis. Two commonly used multivariate techniques include: ordination (such as principal components analysis, canonical correspondence analysis, and detrended correspondence analysis) and classification (such as two-way indicator species analysis). Detailed discussion of these methods can be found in several texts (Gauch, 1982; Jongman *et al.*, 1995; Everitt and Dunn, 2001; Timm, 2002).

IV. Control Charts

The construction and interpretation of control charts is covered in many texts focusing on quality control in industry (*e.g.*, Beauregard *et al.*, 1992; Gyra, 2001; Montgomery 2001). The application of control charts for ecological purposes, however, is relatively straightforward and therefore will be used for the interpretation of all spring community data collected under this protocol. The use of control charts in environmental monitoring is discussed in texts by McBean and Rovers (1998) and Manly (2001), although not in as great detail as the texts referenced above focusing on industrial applications. Many different types of control charts could be constructed, depending upon the type of information desired. For example, control charts can be used to evaluate variables or attributes (*i.e.*, count or frequency data), focus on measures of central tendency or dispersion, and be used in univariate or multivariate analyses.

Although some of the above-mentioned texts discuss the use of multivariate control charts (using the Hotelling T^2 statistic), this approach is only practical for a small number of variables, and assumes a multivariate normal distribution. In general, species abundances are not distributed as multivariate normal (Taylor, 1961), and traditional multivariate procedures are frequently not robust to violations of this assumption (Mardia, 1971; Olson, 1974). A new type of multivariate control chart has recently been described for use with complex ecological communities (Anderson and Thompson, 2004). A software application entitled *ControlChart.exe* is available for constructing these types of multivariate control chart (see Anderson and Thompson, 2004). Multivariate temporal autocorrelation will violate the assumption of stochasticity that this method is based upon, and it is important to test for temporal autocorrelation using Mantel correlograms prior to using this method.

Table 1. Ozarks region invertebrate taxa list with functional feeding groups and tolerance values.

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
	6	C			Branchiobdellida	Hirudinea	Annelida
	7.8	Pa		Erpobdellidae	Arhynchobdellida	Hirudinea	Annelida
<i>Erpobdella punctata</i>		Pa		Erpobdellidae	Arhynchobdellida	Hirudinea	Annelida
<i>Mooreobdella microstoma</i>		Pa		Erpobdellidae	Arhynchobdellida	Hirudinea	Annelida
	7	Pr		Glossiphoniidae	Rhynchobdellida	Hirudinea	Annelida
<i>Glossophonia complanata</i>				Glossiphoniidae	Rhynchobdellida	Hirudinea	Annelida
<i>Helobdella triserialis</i>	8.9			Glossiphoniidae	Rhynchobdellida	Hirudinea	Annelida
<i>Placobdella ornate</i>				Glossiphoniidae	Rhynchobdellida	Hirudinea	Annelida
<i>Placobdella parasitica</i>	6.6	C		Glossiphoniidae	Rhynchobdellida	Hirudinea	Annelida
		Pa		Piscicolidae	Rhynchobdellida	Hirudinea	Annelida
<i>Pisciola punctata</i>		Pa		Piscicolidae	Rhynchobdellida	Hirudinea	Annelida
<i>Haplotaxis</i>		C		Haplotaxidae	Haplotaxida	Oligochaeta	Annelida
	7.3	C		Lumbricidae	Lumbricina	Oligochaeta	Annelida
<i>Lumbriculus variegates</i>	8	C		Lumbriculidae	Lumbriculida	Oligochaeta	Annelida
	10	C		Enchytraeidae	Tubificida	Oligochaeta	Annelida
	9.2	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Aulodrilus</i>	8	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Branchiura sowerbyi</i>	8.4	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Ilyodrilus templetoni</i>	9.4	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Limnodrilus angustipenis</i>		C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Limnodrilus cervix</i>	9.8	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Limnodrilus claparedeianus</i>	10	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Limnodrilus hoffmeisteri</i>	9.8	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Potomothrix bavaricus</i>		C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Quistadrilus multisetosus</i>	10	C		Tubificidae	Tubificida	Oligochaeta	Annelida
<i>Tubifex tubifex</i>	10	C		Tubificidae	Tubificida	Oligochaeta	Annelida
	5.7	Pa, Pr			Hydracarina	Arachnoidea	Arthropoda
<i>Allocrangonyx</i>		C		Allocrangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Allocrangonyx hubrichtti</i>		C		Allocrangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Bactrurus</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Batrurus brachycaudus</i>	8	C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Crangonyx</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Crangonyx forbesi</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Stygobromus</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Stygobromus albamensis</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Stygobromus onandagensis</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Stygobromus ozarkensis</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Stygonectes</i>		C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Synurella</i>	6.9	C		Crangonyctidae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus</i>		C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus fasciatus</i>		C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus minus</i>		C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus pseudolimnaeus</i>	7.9	C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus pulex</i>		C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Gammarus troglophilus</i>		C		Gammaridae	Amphipoda	Crustacea	Arthropoda
<i>Hyaella azteca</i> Sassure		C		Hyaellidae	Amphipoda	Crustacea	Arthropoda
<i>Caecidotea</i>	8	C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea (epigean)</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea antricola</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea brevicauda</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea ancyla</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea dimorpha</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea foxi</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea fustis</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea salemensis</i>	7.7	C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea serrata</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Caecidotea stygia</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Lirceus garmani</i>		C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Lirceus hoppinae</i>	7.7	C		Asellidae	Isopoda	Crustacea	Arthropoda
<i>Cylisticus convexus</i>		C		Cylistidae	Isopoda	Crustacea	Arthropoda
<i>Metroponorthus</i>		C		Porcellionidae	Isopoda	Crustacea	Arthropoda
<i>Porcellio</i>		C		Porcellionidae	Isopoda	Crustacea	Arthropoda
<i>Caucasonethes</i>		C		Trichoniscidae	Isopoda	Crustacea	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Miktoniscus</i>	8.1	C		Trichoniscidae	Isopoda	Crustacea	Arthropoda
<i>Cambarellus puer</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarellus shufeldtii</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus diogenes diogenes</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus hubbsi</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus hubrichti</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus maculatus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Cambarus setosus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Fallicambarus fodiens</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Faxonella clypeata</i>	2.7	C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes eupunctus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes harrisonii</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes hylas</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes immunis</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes lancifer</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes longidigitus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes luteus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes macrus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes marchandi</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes medius</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes meeki</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes neglectus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes neglectus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes neglectus neglectus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes ozarkae</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes palmeri palmeri</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes peruncus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes punctimanus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes quadruncus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes rusticus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes virilis</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Orconectes williamsi</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Procambarus</i>	9.5	C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Procambarus acutus acutus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Procambarus clarkii</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Procambarus gracilis</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Procambarus liberorum</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Procambarus viaeveridus</i>		C		Cambaridae	Decapoda	Crustacea	Arthropoda
<i>Palaemonetes kadiakensis</i>	6.7	C		Palaemonidae	Decapoda	Crustacea	Arthropoda
		C		Entomobrydiae	Collembola	Insecta	Arthropoda
		C		Isotomidae	Collembola	Insecta	Arthropoda
		C		Sminthuridae	Collembola	Insecta	Arthropoda
<i>Helichus</i>	5.4	C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Helichus basalis</i>	5.5	C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Helichus fastigiatus</i>	5.5	C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Helichus lithophilus</i>	5.5	C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Helichus striatus</i>	4.6	C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Acilius</i>		C, Sc		Dryopidae	Coleoptera	Insecta	Arthropoda
<i>Agabus</i>	5	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Agabus amplus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Brachyvatus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Copelatus</i>	9.1	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Coptotomus interrogatus</i>	9	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Coptotomus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Cybister</i>	4.6	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Desmopachria</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Dytiscus</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Graphoderus</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hoperius</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydaticus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydroporus</i>	8.9	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydroporus niger</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydroporus pulcher</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydroporus undulatus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydroporus vilis</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Hydrovatus</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Hygrotus</i>	1.9	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Ilybius</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Laccophilus</i>	10	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Liodessus</i>		Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Oreodytes</i>	4.6	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Rhantus</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Thermonectus</i>	3.7	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Uvarus</i>	4.6	Pr		Dytiscidae	Coleoptera	Insecta	Arthropoda
<i>Ancyronyx variegata</i>	6.9	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Dubiraphia</i>	6.4	C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Dubiraphia vittata</i>		C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Gonielmis dietrichi</i>		C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Heterelmis</i>		C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Macronychus glabratus</i>	4.7	C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Microcylloepus</i>	2.1	C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Microcylloepus pusillus</i>		C		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Optioservus</i>	2.7	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Optioservus immunis</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Optioservus ozarkensis</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis</i>	5.4	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis beameri</i>	4.6	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis bicarinata</i>	5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis cheryl</i>	5.5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis convexula</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis crenata</i>	5.5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis decorata</i>	5.5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis exigua</i>	5.5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis exilis</i>	5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis grossa</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis knobeli</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis lateralis</i>	4.6	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis lignicola</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis mera</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis sandersoni</i>	5.5	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Stenelmis sexlineata</i>	6.4	C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Stenelmis xylonastis</i>		C, Sc		Elmidae	Coleoptera	Insecta	Arthropoda
<i>Dineutus</i>	5.5	Pr		Gyrinidae	Coleoptera	Insecta	Arthropoda
<i>Gyretes</i>	3.7	Pr		Gyrinidae	Coleoptera	Insecta	Arthropoda
<i>Gyretes sinatus</i>		Pr		Gyrinidae	Coleoptera	Insecta	Arthropoda
<i>Gyrinus</i>	6.3	Pr		Gyrinidae	Coleoptera	Insecta	Arthropoda
<i>Halipilus</i>	5			Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes</i>	8.5	He		Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes edentulus</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes litoralis</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes lengi</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes muticus</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes sexmaculatus</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Peltodytes tortulosus</i>				Halipilidae	Coleoptera	Insecta	Arthropoda
<i>Helophorus</i>	7.9	Sh		Helophoridae	Coleoptera	Insecta	Arthropoda
<i>Heterocerus</i>				Heteroceridae	Coleoptera	Insecta	Arthropoda
<i>Hydraena</i>		C, Sc		Hydraenidae	Coleoptera	Insecta	Arthropoda
<i>Ochthebus</i>		C, Sc		Hydraenidae	Coleoptera	Insecta	Arthropoda
<i>Hydrochus</i>	4.6	Sh		Hydrochidae	Coleoptera	Insecta	Arthropoda
<i>Berosus</i>	8.6	He, C		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Chaetarthria</i>	5.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Crenitis</i>		C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Cymbiodyta</i>	5.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Dibolocelus</i>	5.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Enochrus</i>	8.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Helobata</i>				Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Helochares</i>	5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Helocombus</i>	5.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Hydrobius</i>	5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Hydrochara</i>	6	C		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Hydrophilus</i>	4.6	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Laccobius</i>	8	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Paracymus</i>	7.3	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Sperchopsis</i>	6.5	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Tropisternus</i>	9.8	C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Tropisternus lateralis</i>		C, Pr		Hydrophilidae	Coleoptera	Insecta	Arthropoda
<i>Lutrochus</i>	2.75			Lutrochidae	Coleoptera	Insecta	Arthropoda
<i>Lutrochus laticeps</i>				Lutrochidae	Coleoptera	Insecta	Arthropoda
<i>Hydrocanthus</i>	6.9	Pr		Noteridae	Coleoptera	Insecta	Arthropoda
<i>Suphisellus</i>		Pr		Noteridae	Coleoptera	Insecta	Arthropoda
<i>Ectopria nervosa</i>	4.3	Sc		Psephenidae	Coleoptera	Insecta	Arthropoda
<i>Psephenus herricki</i>	2.5	Sc		Psephenidae	Coleoptera	Insecta	Arthropoda
<i>Cyphon</i>		Sh		Salpingidae	Coleoptera	Insecta	Arthropoda
<i>Scirtes</i>	5	Sh		Scirtidae	Coleoptera	Insecta	Arthropoda
<i>Atherix</i>	2.1	Pr		Athericidae	Diptera	Insecta	Arthropoda
<i>Atherix lantha</i>		Pr		Athericidae	Diptera	Insecta	Arthropoda
<i>Atherix variegata</i>		Pr		Athericidae	Diptera	Insecta	Arthropoda
	6	C, Pr, Sc					
<i>Bezzia</i>		Pr	Ceratopogoninae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Culicoides</i>		C, Pr	Ceratopogoninae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Mallochohelea</i>		Pr	Ceratopogoninae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Palpomyia</i>		C, Pr	Ceratopogoninae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Probezzia</i>		Pr	Ceratopogoninae	Ceratopogonidae	Diptera	Insecta	Arthropoda
	6	C, Sc					
<i>Dasyhelea</i>		C, Sc	Dashyheleinae	Ceratopogonidae	Diptera	Insecta	Arthropoda
	6	C					
<i>Atrichopogon</i>		C	Forcipomyiinae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Chaoborus</i>	8.5	Pr	Forcipomyiinae	Ceratopogonidae	Diptera	Insecta	Arthropoda
<i>Eucorethea underwoodi</i>		Pr		Chaoboridae	Diptera	Insecta	Arthropoda
<i>Acalcarella</i>		C	Chironominae	Chironomidae	Diptera	Insecta	Arthropoda
<i>Apedilum</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Axarus</i>	6	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Chironomus</i>	9.8	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cladopelma</i>	2.5	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cladotanytarsus</i>	3.7	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cladotanytarsus</i>	3.7	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cladotanytarsus</i>	5.5	C, F		Chironomidae	Diptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Constempellina</i>	4	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cryptochironomous</i>	7.4	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cryptotendipes</i>	6.1	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Demicryptochironomus</i>	2.1	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Dicrotendipes</i>	7.9	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Endochironomus</i>	7.5	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Endotribelos</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Glyptotendipes</i>	8.5	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Goeldichironomus</i>	9			Chironomidae	Diptera	Insecta	Arthropoda
<i>Harnischia</i>	7.5			Chironomidae	Diptera	Insecta	Arthropoda
<i>Hyporhygma</i>		Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Kiefferulus</i>	10	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Lauterborniella</i>	5.5	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Lipiniella</i>		C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Microchironomous</i>	8	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Micropsectra</i>	1.4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Microtendipes</i>	6.2	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Nilothauma</i>	5.5			Chironomidae	Diptera	Insecta	Arthropoda
<i>Omisus</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Pagastiella</i>	2.6			Chironomidae	Diptera	Insecta	Arthropoda
<i>Parachironomus</i>	9.2	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Paracladopelma</i>	4.8	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Paralauterborniella</i>	8			Chironomidae	Diptera	Insecta	Arthropoda
<i>Paratanytarsus</i>	7.7	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Paratendipes</i>	5.3	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Phaenopsectra</i>	6.2	C, Sc		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum</i>	7.4	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum convictum grp</i>	5.3	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum fallax grp</i>	6.7	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum halterale grp</i>	7.2	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum illinoense grp</i>	9.2	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Polypedilum scaleanum grp</i>	8.7	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Pseudochironomus</i>	4.2	Co		Chironomidae	Diptera	Insecta	Arthropoda
<i>Rheotanytarsus</i>	6.4	Fi		Chironomidae	Diptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Robackia</i>	3.4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Saetheria</i>	8.1	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Stelechomyia</i>	4.6			Chironomidae	Diptera	Insecta	Arthropoda
<i>Stempellina</i>	2	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Stempellinella</i>	5.3	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Stenochironomus</i>	6.4	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Stictochironomus</i>	6.7	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Sublettea</i>	1.7	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Tanytarsus</i>	6.7	C, F		Chironomidae	Diptera	Insecta	Arthropoda
<i>Thienemanniola</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Tribelos</i>	6.6	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Xenochironomus</i>	7	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Zavreliella</i>	7			Chironomidae	Diptera	Insecta	Arthropoda
<i>Diamesa</i>	7.7	C	Diamesinae	Chironomidae	Diptera	Insecta	Arthropoda
<i>Potthastia</i>	4.7	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Pseudodiamesa</i>	4.6	C	Orthoclaadiinae	Chironomidae	Diptera	Insecta	Arthropoda
<i>Sympotthastia</i>	5.7	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Syndiamesa</i>		C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Brillia</i>	5.2	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cardiocladius</i>	6.2	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Corynoneura</i>	6.2	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cricotopus bicintus</i>	8.7	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Cricotopus/Orthocladus</i>	6.5	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Diplocladius</i>	7.7	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Epoicocladius</i>	4	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella</i>	4	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella brehmi grp</i>	3.7	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella brevicar grp</i>	4	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella claripennis grp</i>	5.7	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella coerulesens grp</i>	4	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella cyanea grp</i>		C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella devonica grp</i>	2.6	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella gracei grp</i>	2.7	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Eukiefferiella pseudomontana</i> grp	8	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella rectangularis</i> grp		C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Eukiefferiella similis</i> grp		C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Heterotrissocladius</i>	5.4	C, Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Hydrobaenus</i>	9.6	C, Sc		Chironomidae	Diptera	Insecta	Arthropoda
<i>Krenosmittia</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Lymnophyes</i>	8	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Mesocricotopus</i>		C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Mesosmittia</i>	7			Chironomidae	Diptera	Insecta	Arthropoda
<i>Nanocladius</i>	7.2	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Oliveridia</i>				Chironomidae	Diptera	Insecta	Arthropoda
<i>Orthocladius</i> (<i>Euorthocladius</i>)	6.3	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Orthocladius</i> (<i>Symposiocladius</i>)	5.4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Parakiefferiella</i>	5.9	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Parametriocnemus</i>	3.7	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Paraphaenocladius</i>	4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Parorthocladius</i>		C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Psectrocladius</i>	3.8	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Pseudorthocladius</i>		C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Pseudosmittia</i>	4			Chironomidae	Diptera	Insecta	Arthropoda
<i>Rheocricotopus</i>	7.3	C, Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Smittia</i>	4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Synorthocladius</i>	4.7			Chironomidae	Diptera	Insecta	Arthropoda
<i>Thienemanniella</i>	6			Chironomidae	Diptera	Insecta	Arthropoda
<i>Tvetenia</i>	4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Tvetenia bavarica</i> grp	3.9	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Tvetenia discoloripes</i> grp	4	C		Chironomidae	Diptera	Insecta	Arthropoda
<i>Xylotopus</i>	6.6	Sh		Chironomidae	Diptera	Insecta	Arthropoda
<i>Monodiamesa</i>	7	C	Prodiamesinae	Chironomidae	Diptera	Insecta	Arthropoda
<i>Ablabesmyia</i>	6.4	Pr	Tanypodinae	Chironomidae	Diptera	Insecta	Arthropoda
<i>Clinotanypus</i>	9.1	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Coelotanypus</i>	6.2	Pr		Chironomidae	Diptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Djalmabatista</i>	6.4	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Krenopelopia</i>		Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Labrundinia</i>	5.3	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Larsia</i>	83	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Monopelopia</i>		Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Natarsia</i>	8	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Nilotanypus</i>	6	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Paramerina</i>	2.8	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Pentaneura</i>	4.6	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Procladius</i>	9.3	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Psectrotanypus</i>	10	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Tanypus</i>	9.6	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Thienemannimyia</i> grp.	6	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Zavrelymia</i>	9.3	Pr		Chironomidae	Diptera	Insecta	Arthropoda
<i>Aedes</i>	5.5	Pr		Culicidae	Diptera	Insecta	Arthropoda
<i>Anopheles</i>	9.1	Fi		Culicidae	Diptera	Insecta	Arthropoda
<i>Culex</i>	10	Fi		Culicidae	Diptera	Insecta	Arthropoda
<i>Culiseta</i>	5.5	C, F		Culicidae	Diptera	Insecta	Arthropoda
<i>Mansonia</i>		C, F		Culicidae	Diptera	Insecta	Arthropoda
<i>Dixa</i>	2.8	C, Pr		Dixidae	Diptera	Insecta	Arthropoda
<i>Dixella</i>		C		Dixidae	Diptera	Insecta	Arthropoda
	9.7	Pr		Dolichopodidae	Diptera	Insecta	Arthropoda
<i>Chelifera</i>	6	Pr		Empididae	Diptera	Insecta	Arthropoda
<i>Clinocera</i>	6	Pr		Empididae	Diptera	Insecta	Arthropoda
<i>Hemerodromia</i>	6	C, Pr		Empididae	Diptera	Insecta	Arthropoda
<i>Rhamphomyia</i>		Pr		Empididae	Diptera	Insecta	Arthropoda
<i>Roederiodes</i>		Pr		Empididae	Diptera	Insecta	Arthropoda
	5.5	C, Sh		Ephydriidae	Diptera	Insecta	Arthropoda
	6	Pr		Muscidae	Diptera	Insecta	Arthropoda
<i>Pericoma</i>	4	C		Psychodidae	Diptera	Insecta	Arthropoda
<i>Pericoma fuliginosa</i>		C		Psychodidae	Diptera	Insecta	Arthropoda
<i>Pericoma freyii</i>		C		Psychodidae	Diptera	Insecta	Arthropoda
<i>Pericoma mutua</i>		C		Psychodidae	Diptera	Insecta	Arthropoda
<i>Psychoda</i>	9.9	C		Psychodidae	Diptera	Insecta	Arthropoda

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<i>Telmatoscopus</i>		C		Psychodidae	Diptera	Insecta	Arthropoda
		He,		Scathophagidae	Diptera	Insecta	Arthropoda
		Sh		Sciomyzidae	Diptera	Insecta	Arthropoda
		Pr		Simuliidae	Diptera	Insecta	Arthropoda
<i>Cnephia</i>	4	F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Prosimulium</i>	2.6	F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Simulium</i>	4.4	F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Simulium luggeri</i>		F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Simulium venustum</i>		F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Stegapterna</i>	5	F		Simuliidae	Diptera	Insecta	Arthropoda
<i>Allognosta</i>				Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Caloparyphus</i>		C		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Euparyphus</i>		C, Sc		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Myxosargus</i>		C		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Nemotelus</i>	7.3	C		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Odontomyia</i>	7.3	C		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Oxycera</i>		Sc		Stratiomyidae	Diptera	Insecta	Arthropoda
<i>Stratiomys</i>	7.3	C		Stratiomyidae	Diptera	Insecta	Arthropoda
	8.25	C		Syrphidae	Diptera	Insecta	Arthropoda
<i>Chrysops</i>	7.3	Pr		Tabanidae	Diptera	Insecta	Arthropoda
<i>Hybomitra</i>		Pr		Tabanidae	Diptera	Insecta	Arthropoda
<i>Silvius</i>		Pr		Tabanidae	Diptera	Insecta	Arthropoda
<i>Tabanus</i>	9.7	Pr		Tabanidae	Diptera	Insecta	Arthropoda
<i>Protoplasia fitchii</i>				Tanyderidae	Diptera	Insecta	Arthropoda
<i>Antocha</i>	4.6	C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Cryptolabis</i>				Tipulidae	Diptera	Insecta	Arthropoda
<i>Dactylolabis</i>	3.7	C, Sh		Tipulidae	Diptera	Insecta	Arthropoda
<i>Dicranota</i>	0	Pr		Tipulidae	Diptera	Insecta	Arthropoda
<i>Erioptera</i>	5.5	C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Gonomyia</i>	5.5	C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Holorusia</i>		Sh		Tipulidae	Diptera	Insecta	Arthropoda
<i>Hexatoma</i>	4.7	Pr		Tipulidae	Diptera	Insecta	Arthropoda
<i>Limnophila</i>	4.6	Pr		Tipulidae	Diptera	Insecta	Arthropoda
<i>Limonia</i>	10	Sh		Tipulidae	Diptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Lipsothrix</i>		Sh		Tipulidae	Diptera	Insecta	Arthropoda
<i>Molophilus</i>				Tipulidae	Diptera	Insecta	Arthropoda
<i>Ormosia</i>	4.6	C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Paradelphomyia</i>				Tipulidae	Diptera	Insecta	Arthropoda
<i>Pedicia</i>	4.6	Pr		Tipulidae	Diptera	Insecta	Arthropoda
<i>Pilaria</i>	7			Tipulidae	Diptera	Insecta	Arthropoda
<i>Prionocera</i>		Sh		Tipulidae	Diptera	Insecta	Arthropoda
<i>Pseudolimnophila</i>	7.3	C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Rhabdomastix</i>		C		Tipulidae	Diptera	Insecta	Arthropoda
<i>Tipula</i>	7.7	C, Sh		Tipulidae	Diptera	Insecta	Arthropoda
<i>Thaumalea</i>		Sc		Thaumaleidae	Diptera	Insecta	Arthropoda
<i>Ameletus</i>	7	C, Sc		Ameletidae	Ephemeroptera	Insecta	Arthropoda
<i>Ameletus lineatus</i>	2.1	C, Sc		Ameletidae	Ephemeroptera	Insecta	Arthropoda
<i>Ameletus ludens</i>	0	C, Sc		Ameletidae	Ephemeroptera	Insecta	Arthropoda
	4	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Acentrella</i>	3.6	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Acerpenna</i>	3.7	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Apobaetis</i>	6	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Baetis</i>	6	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Barbaetis</i>		C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Callibaetis</i>	9.3	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Camelobaetis</i>	9.3	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Centroptilum</i>	6.3	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Centroptilum ozakensum</i>		C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Cloeon</i>	7.4	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Diphetor</i>	5			Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Fallceon</i>	6	Pr		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Heterocloeon</i>	2	Sc		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Labiobaetis</i>	6	Pr		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Paracloeodes</i>	5	Sc		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Plauditus</i>	6	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Procloeon</i>	6.3	C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Pseudocentroptiloides</i>		C		Baetidae	Ephemeroptera	Insecta	Arthropoda
<i>Pseudocloeon</i>		C		Baetidae	Ephemeroptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Baetisca gibbera</i>	1.4	C, Sc		Baetiscidae	Ephemeroptera	Insecta	Arthropoda
<i>Baetisca lacustris</i>	4	C, Sc		Baetiscidae	Ephemeroptera	Insecta	Arthropoda
<i>Baetisca obesa</i>	4	C, Sc		Baetiscidae	Ephemeroptera	Insecta	Arthropoda
<i>Amercaenis</i>		F		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Brachycercus</i>	3.5	C		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis amica</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis anceps</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis hilaris</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis latipennis</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis maccafferti</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis punctata</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Caenis tardata</i>	7.6	C, Sc		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Cercobrachys</i>		C		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Cercobrachys serpentis</i>		C		Caenidae	Ephemeroptera	Insecta	Arthropoda
<i>Attenella attenuata</i>	1	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella</i>	1.7	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella argo</i>		C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella aurivillii</i>	0	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella catawba</i>	4	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella dorothea</i>	1	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella excrucians</i>	2	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella invaria</i>	2.2	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella needhami</i>	0	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella subvaria</i>	1	C, Sc		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella</i>	3	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella aestiva</i>	5	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella bicolor</i>	5.1	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella enoensis</i>	5	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella funeralis</i>	2.3	Sh		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella macdunnoughi</i>		C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Eurylophella versimilis</i>	0.3	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Serratella</i>	1.9	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Serratella deficiens</i>	2.7	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda

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<i>Serratella frisoni</i>		C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Serratella serratooides</i>	1.5	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Serratella sordida</i>	2	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Timpanoga</i>	2	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Timpanoga lita</i>	0	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Timpanoga provonshali</i>		C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Timpanoga simplex</i>	3.9	C		Ephemerellidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella</i>	2.2	C, Pr		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella guttulata</i>	2	C, Pr		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella simulans</i>	3.7	C, Pr		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella traversae</i>		C, Pr		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Ephemerella varia</i>		C, Pr		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Hexagenia</i>	4.7	C		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Hexagenia atrocaudata</i>	3.7	C		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Hexagenia bilineata</i>	3.7	C		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Hexagenia limbata</i>	4.6	C		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Hexagenia rigida</i>	5.5	C		Ephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Anepeorus</i>		Pr		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Epeorus</i>	1.2	C		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Heptagenia</i>	2.8	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Leucrocuta</i>	0	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Macdunnoa</i>	4.6	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Nixe</i>	2	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Rhithrogena</i>	0.4	C		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenacron</i>	7.1	C		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema</i>	3.4	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema bednariki</i>	3.4	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema exiguum</i>	5	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema femoratum</i>	7.5	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema integrum</i>	5.5	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema luteum</i>		C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema mediopunctatum</i>	1.7	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema modestum</i>	5.8	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema pulchellum</i>	3	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda

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<i>Stenonema terminatum</i>	4.5	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Stenonema vicarium</i>	1	C, Sc		Heptageniidae	Ephemeroptera	Insecta	Arthropoda
<i>Isonychia</i>	3.8	F		Isonychidae	Ephemeroptera	Insecta	Arthropoda
<i>Isonychia bicolor</i>	2	F		Isonychidae	Ephemeroptera	Insecta	Arthropoda
<i>Isonychia rufa</i>	3.7	F		Isonychidae	Ephemeroptera	Insecta	Arthropoda
<i>Isonychia sayi</i>		F		Isonychidae	Ephemeroptera	Insecta	Arthropoda
<i>Isonychia sicca</i>	3.7	F		Isonychidae	Ephemeroptera	Insecta	Arthropoda
<i>Choroterpes</i>	2	C, Sc		Leptophlebiidae	Ephemeroptera	Insecta	Arthropoda
<i>Habrophlebiodes</i>	6	C, Sc		Leptophlebiidae	Ephemeroptera	Insecta	Arthropoda
<i>Leptophlebia</i>	6.4	C		Leptophlebiidae	Ephemeroptera	Insecta	Arthropoda
<i>Neochoroterpes</i>		C, Sc		Leptophlebiidae	Ephemeroptera	Insecta	Arthropoda
<i>Paraleptophlebia</i>	1.2	C, Sh		Leptophlebiidae	Ephemeroptera	Insecta	Arthropoda
<i>Neoephemera</i>		C		Neoephemeridae	Ephemeroptera	Insecta	Arthropoda
<i>Pentagenia vittigera</i>	6.4	C		Palingeniidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephoron</i>	4.6	C		Polymitarciidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephoron album</i>	4.6	C		Polymitarciidae	Ephemeroptera	Insecta	Arthropoda
<i>Ephoron leukon</i>	1.5	C		Polymitarciidae	Ephemeroptera	Insecta	Arthropoda
<i>Tortopus primus</i>	5.5	C		Polymitarciidae	Ephemeroptera	Insecta	Arthropoda
<i>Anthopotamus</i>	1.6	F		Potamanthidae	Ephemeroptera	Insecta	Arthropoda
<i>Pseudiron centralis</i>	5	Pr		Pseudironidae	Ephemeroptera	Insecta	Arthropoda
<i>Siphonurus</i>	2.6	C, Sc		Siphonuridae	Ephemeroptera	Insecta	Arthropoda
<i>Tricorythodes</i>	5.4	C		Tricorythidae	Ephemeroptera	Insecta	Arthropoda
<i>Belostoma</i>	9.8	Pr		Belostomatidae	Hemiptera	Insecta	Arthropoda
<i>Lethocerus</i>	4.6	Pr		Belostomatidae	Hemiptera	Insecta	Arthropoda
<i>Corisella</i>	6.4	Pr		Corixidae	Hemiptera	Insecta	Arthropoda
<i>Hesperocorixa</i>	5	Mp		Corixidae	Hemiptera	Insecta	Arthropoda
<i>Palmacorixa</i>	5.5			Corixidae	Hemiptera	Insecta	Arthropoda
		Mp,					
<i>Sigara</i>	4.6	C		Corixidae	Hemiptera	Insecta	Arthropoda
<i>Trichocorixa</i>	5.5	C, Pr		Corixidae	Hemiptera	Insecta	Arthropoda
<i>Gelastocoris</i>	7.3	Pr		Gelastocoridae	Hemiptera	Insecta	Arthropoda
<i>Aquarius</i>	6.4	Pr		Gerridae	Hemiptera	Insecta	Arthropoda
<i>Gerris</i>	6.4	Pr		Gerridae	Hemiptera	Insecta	Arthropoda
<i>Metrobates</i>	6.4	Pr		Gerridae	Hemiptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Rheumatobates</i>	6.4	Pr		Gerridae	Hemiptera	Insecta	Arthropoda
<i>Trepobates</i>	6.4	Pr		Gerridae	Hemiptera	Insecta	Arthropoda
<i>Hebrus</i>	6.4	Pr		Hebridae	Hemiptera	Insecta	Arthropoda
<i>Merragata</i>	7.3	Pr		Hebridae	Hemiptera	Insecta	Arthropoda
<i>Hydrometra</i>	7.3	Pr		Hydrometridae	Hemiptera	Insecta	Arthropoda
<i>Mesovelis</i>	6.4	Pr		Mesoveliidae	Hemiptera	Insecta	Arthropoda
<i>Pelocoris</i>		Pr		Naucoridae	Hemiptera	Insecta	Arthropoda
<i>Nepa</i>	4.6	Pr		Nepidae	Hemiptera	Insecta	Arthropoda
<i>Ranatra</i>	7.5	Pr		Nepidae	Hemiptera	Insecta	Arthropoda
<i>Ranatra fusca</i>	7.3	Pr		Nepidae	Hemiptera	Insecta	Arthropoda
<i>Ranatra kirkaldyi</i>	5.5	Pr		Nepidae	Hemiptera	Insecta	Arthropoda
<i>Ranatra nigra</i>	6.4	Pr		Nepidae	Hemiptera	Insecta	Arthropoda
<i>Buenoa</i>	5.5	Pr		Notonectidae	Hemiptera	Insecta	Arthropoda
<i>Notonecta</i>	5.5	Pr		Notonectidae	Hemiptera	Insecta	Arthropoda
<i>Neoplea</i>	5.5	Pr		Pleidae	Hemiptera	Insecta	Arthropoda
<i>Micracanthia</i>	5.5	Pr		Saldidae	Hemiptera	Insecta	Arthropoda
<i>Pentacora</i>	6.4	Pr		Saldidae	Hemiptera	Insecta	Arthropoda
<i>Saldula</i>	6.4	Pr		Saldidae	Hemiptera	Insecta	Arthropoda
<i>Microvelia</i>	6.4	Pr		Veliidae	Hemiptera	Insecta	Arthropoda
<i>Platyvelia</i>		Pr		Veliidae	Hemiptera	Insecta	Arthropoda
<i>Rhagovelia</i>	7.3	Pr		Veliidae	Hemiptera	Insecta	Arthropoda
<i>Steinovelis</i>		Pr		Veliidae	Hemiptera	Insecta	Arthropoda
		He		Cossidae	Lepidoptera	Insecta	Arthropoda
<i>Nepticula</i>		He		Nepticulidae	Lepidoptera	Insecta	Arthropoda
<i>Bellura</i>		He		Noctuidae	Lepidoptera	Insecta	Arthropoda
<i>Simyra</i>		He		Noctuidae	Lepidoptera	Insecta	Arthropoda
<i>Nymphula</i>	7	He		Crambidae	Lepidoptera	Insecta	Arthropoda
<i>Parapoynx</i>	5	He		Crambidae	Lepidoptera	Insecta	Arthropoda
		He,					
<i>Petrophila</i>	1.8	Sc		Crambidae	Lepidoptera	Insecta	Arthropoda
<i>Schoenobius</i>		He		Crambidae	Lepidoptera	Insecta	Arthropoda
<i>Archips</i>		Sh		Totidae	Lepidoptera	Insecta	Arthropoda
<i>Chauliodes</i>	4	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Chauliodes pectinicornis</i>	4	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Chauliodes rastricornis</i>	4	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Corydalus</i>	5.6	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Corydalus cornutus</i>	5.6	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Nigronia fasciatus</i>	6.2	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Nigronia serricornis</i>	5.5	Pr		Corydalidae	Megaloptera	Insecta	Arthropoda
<i>Sialis</i>	7.5	Pr		Sialidae	Megaloptera	Insecta	Arthropoda
<i>Sialis velata</i>		Pr		Sialidae	Megaloptera	Insecta	Arthropoda
<i>Climacia</i>	6.5	Pr		Sisyridae	Neuroptera	Insecta	Arthropoda
<i>Sisyra</i>		Pr		Sisyridae	Neuroptera	Insecta	Arthropoda
<i>Sisyra vicaria</i>		Pr		Sisyridae	Neuroptera	Insecta	Arthropoda
<i>Aeshna</i>	6.4	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Anax</i>	6.4	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Basiaeschna janata</i>	7.7	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Boyeria</i>	6.3	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Epiaeschna heros</i>	3.7	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Nasiaeschna pentacantha</i>	8	Pr		Aeshnidae	Odonata	Insecta	Arthropoda
<i>Calopteryx</i>	8.3	Pr		Calopterygidae	Odonata	Insecta	Arthropoda
<i>Hetaerina</i>	6.2	Pr		Calopterygidae	Odonata	Insecta	Arthropoda
<i>Amphiagrion</i>	2.8	Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Argia</i>	8.7	Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Chromagrion</i>		Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Enallagma</i>	9	Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Ischnura</i>	9.4	Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Nehalennia</i>		Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Telebasis</i>		Pr		Coenagrionidae	Odonata	Insecta	Arthropoda
<i>Cordulegaster</i>	6.1	Pr		Cordulegastridae	Odonata	Insecta	Arthropoda
	7	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Arigomphus</i>	6.4	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Dromogomphus</i>	6.3	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Erpetogomphus</i>	5.5	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Gomphus</i>	6.2	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Hagenius brevistylus</i>	4	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Lanthus parvulus</i>	2.7	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Lanthus vernalis</i>	2.7	Pr		Gomphidae	Odonata	Insecta	Arthropoda

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<i>Ophiogomphus</i>	6.2	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Progomphus obscurus</i>	8.7	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Stylogomphus albistylus</i>	4.8	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Stylurus</i>	4	Pr		Gomphidae	Odonata	Insecta	Arthropoda
<i>Archilestes</i>	6.4	Pr		Lestidae	Odonata	Insecta	Arthropoda
<i>Lestes</i>	6.4	Pr		Lestidae	Odonata	Insecta	Arthropoda
<i>Epithea (Epicordulia)</i>	5.5	Pr	Corduliinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Epithea (Tetragoneuria)</i>	8.5	Pr	Corduliinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Helocordulia</i>		Pr	Corduliinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Neurocordulia</i>	4	Pr	Corduliinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Somatochlora</i>	8.9	Pr	Corduliinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Brachymesia</i>		Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Celithemis</i>	3.7	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Dythemis</i>	3.7	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Erythemis</i>	7.7	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Erythrodiplax</i>		Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Leucorrhinia</i>	6.4	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Libellula</i>	9.8	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Orthemis ferruginea</i>	4.6	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Pachydiplax longipennis</i>	9.6	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Pantala</i>	6.4	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Perithemis</i>	10	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Plathemis</i>	10	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Sympetrum</i>	7.3	Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Tramea</i>		Pr	Libellulinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Didymops</i>	5.5	Pr	Macromiinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Macromia</i>	6.7	Pr	Macromiinae	Libellulidae	Odonata	Insecta	Arthropoda
<i>Tachopteryx thoreyi</i>	3.7	Pr		Petaluridae	Odonata	Insecta	Arthropoda
<i>Allocapnia</i>	2.8	Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocania forbesi</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia granulata</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia jeanae</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia malverna</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia mohri</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Allocapnia mystica</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia oribata</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia ozarkana</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia peltoides</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocania pygmaea</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia rickeri</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia sandersoni</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia smithi</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia vivipara</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Allocapnia warreni</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Nemocapnia carolina</i>		Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Paracapnia angulata</i>	0.2	Sh		Capniidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla</i>	1.4	Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla caddo</i>		Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla caudata</i>		Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla hamata</i>		Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla leonarda</i>		Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Alloperla ouachita</i>		Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Haploperla</i>	1.4	C, Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Haploperla brevis</i>	1.3	C, Pr		Chloroperlidae	Plecoptera	Insecta	Arthropoda
<i>Leuctra</i>	0.7	Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Leuctra paleo</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Leuctra rickeri</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Leuctra tenuis</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealeuctra</i>	0	Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealeuctra cherokee</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealuctra claasseni</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealuctra fraxina</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealuctra narfi</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealuctra warreni</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Zealuctra wachita</i>		Sh		Leuctridae	Plecoptera	Insecta	Arthropoda
<i>Amphinemura</i>	3.4	Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Amphinemura delosa</i>		Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Amphinemura nigrutta</i>		Sh		Nemouridae	Plecoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Prostoia</i>	6.1	Sc, Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Prostoia completa</i>	6.1	Sc, Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Prostoia similis</i>	6.1	Sc, Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Shipsa rotunda</i>	0.3	Sc, Sh		Nemouridae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria</i>	1.4	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria abnormis</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria evoluta</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria filicis</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria internata</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria mela</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria ozarkensis</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Acroneuria perplexa</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Agneta capitata</i>	1.4	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Agneta flavescens</i>	0	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Attaneuria ruralis</i>	2.75	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla</i>	1.6	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla carlsoni</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla catharae</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla choctaw</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla falayah</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla harpi</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Neoperla osage</i>		Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Paragnetina</i>	1.8	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Paragnetina kansensis</i>	2	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Paragnetina media</i>	1	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlesta</i>	0	C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlesta baumanni</i>		C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlesta browni</i>		C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlesta cinctipes</i>		C, Pr					
<i>Perlesta decipiens</i>		C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Perlesta fusca</i>		C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlesta shubata</i>		C, Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlinella drymo</i>	0	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Perlinella ephyre</i>	0	Pr		Perlidae	Plecoptera	Insecta	Arthropoda
<i>Clioperla clio</i>	4.8	Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Helopicus nalatus</i>	5.75	Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Hydroperla</i>	2.75	Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Hydroperla crosbyi</i>	3.7	Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Hydroperla fugitans</i>	2.75	Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla</i>	2	C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla bilineata</i>	5.5	C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla buski</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla couthatta</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla decepta</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla dicala</i>	2.2	C, Pr					
<i>Isoperla mohri</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla namata</i>	1.8	C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla mohri</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla signata</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Isoperla szczytkoi</i>		C, Pr		Perlodidae	Plecoptera	Insecta	Arthropoda
<i>Pteronarcys pictetii</i>	1.7	Sh		Pteronarcyidae	Plecoptera	Insecta	Arthropoda
<i>Strophopteryx</i>	2.5	Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Strophopteryx arkansae</i>		Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Strophopteryx cucullata</i>		Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Strophopteryx fasciata</i>	3	Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx</i>	6.3	C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx burksi</i>	5.8	C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx lita</i>		C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx lonicera</i>	2	C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx maura</i>		C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Taeniopteryx metequi</i>	1.4	C, Sh		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
		C,					
<i>Taeniopteryx parvula</i>	2	SH		Taeniopterygidae	Plecoptera	Insecta	Arthropoda
<i>Brachycentrus</i>	2.2	F		Brachycentridae	Trichoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Brachycentrus americanus</i>	1	F		Brachycentridae	Trichoptera	Insecta	Arthropoda
<i>Brachycentrus lateralis</i>	0.4	F		Brachycentridae	Trichoptera	Insecta	Arthropoda
<i>Brachycentrus numerosus</i>	1.8	F		Brachycentridae	Trichoptera	Insecta	Arthropoda
<i>Brachycentrus occidentalis</i>	1	F		Brachycentridae	Trichoptera	Insecta	Arthropoda
<i>Micrasema</i>	0.6	Sh		Brachycentridae	Trichoptera	Insecta	Arthropoda
<i>Agapetus</i>	0	C, Sc		Glossosomatidae	Trichoptera	Insecta	Arthropoda
<i>Agapetus illini</i>	0	C, Sc		Glossosomatidae	Trichoptera	Insecta	Arthropoda
<i>Glossosoma</i>	1.5	C, Sc		Glossosomatidae	Trichoptera	Insecta	Arthropoda
<i>Protoptila</i>	2.8	Sc		Glossosomatidae	Trichoptera	Insecta	Arthropoda
<i>Helicopsyche</i>	0	Sc		Helicopsychidae	Trichoptera	Insecta	Arthropoda
<i>Ceratopsyche</i>	1.4	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Ceratopsyche morosa grp</i>	2.95	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Ceratopsyche piatrix</i>		F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Ceratopsyche slosonae</i>	0	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Cheumatopsyche</i>	6.6	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Diplectrona</i>	2.2	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Hydropsyche</i>	4	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Macrostemum</i>	3.6	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Potamyia flava</i>	5	F		Hydropsychidae	Trichoptera	Insecta	Arthropoda
<i>Agraylea</i>	8	He, Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Dibusa angata</i>		Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Hydroptila</i>	6.2	He, Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Ithytrichia</i>	4	Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Leucotrichia</i>	4.3	C, Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Neotrichia</i>	2	Sc		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Ochrotrichia</i>	6.4	C, He		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Orthotrichia</i>	7.2	C, He		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Oxyethira</i>	3	C, He		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Stactobiella</i>	2.75	Sh		Hydroptilidae	Trichoptera	Insecta	Arthropoda
<i>Lepidostoma</i>	1	Sh		Lepidostomatidae	Trichoptera	Insecta	Arthropoda
<i>Ceraclea</i>	2.3	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Leptocerus americanus</i>	4.6	Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Mystacides</i>	3.5	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche</i>	4.1	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche albida</i>	5.5	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche candida</i>	3.8	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche diarina</i>	5.5	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche exquisita</i>	4.2	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche pavida</i>	4.2	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Nectopsyche spiloma</i>	4.6	C, Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Oecetis</i>	5.7	He, Pr		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Setodes</i>	0.9	C, Pr		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Triaenodes</i>	3.7	Sh		Leptoceridae	Trichoptera	Insecta	Arthropoda
<i>Frenesia</i>	0	Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Hesperophylax</i>	3	He, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Hydatophylax</i>	2.3	C, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Ironoquia</i>	7.3	C, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Leptophylax</i>		He, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Limnephilus</i>	2.75	He, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Platycentropus</i>	4	Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Pseudostenophylax</i>	0	C, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Pycnopsyche</i>	2.3	C, Sh		Limnephilidae	Trichoptera	Insecta	Arthropoda
<i>Molanna</i>	3.9	C, Sh		Molannidae	Trichoptera	Insecta	Arthropoda
<i>Marilia</i>		Sh		Odontoceridae	Trichoptera	Insecta	Arthropoda
<i>Psilotreta</i>	0	C, Sc		Odontoceridae	Trichoptera	Insecta	Arthropoda
<i>Chimarra</i>	2.8	F		Philopotamidae	Trichoptera	Insecta	Arthropoda
<i>Dolophilodes</i>	1	F		Philopotamidae	Trichoptera	Insecta	Arthropoda
<i>Wormaldia</i>	0.4	F		Philopotamidae	Trichoptera	Insecta	Arthropoda
<i>Agrypnia</i>	4.6	Sh		Phryganeidae	Trichoptera	Insecta	Arthropoda
<i>Banksiola</i>		Sh		Phryganeidae	Trichoptera	Insecta	Arthropoda
<i>Phryganea</i>	4.6	Sh, Pr		Phryganeidae	Trichoptera	Insecta	Arthropoda

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Ptilostomis</i>	6.7	Sh, Pr		Phryganeidae	Trichoptera	Insecta	Arthropoda
<i>Cernotina</i>	4.6	Pr		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Cyrnellus fraternus</i>	7.4	F		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Neureclipsis</i>	4.4	F, Sh		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Paranyctiophylax</i>	0.9	C, Pr		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Phylocentropus</i>	5.6	F		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Polycentropus</i>	3.5	C, Pr		Polycentropodidae	Trichoptera	Insecta	Arthropoda
<i>Lype diversa</i>	4.3	Sc		Psychomyiidae	Trichoptera	Insecta	Arthropoda
<i>Paduniella neartica</i>	0	C, Sc		Psychomyiidae	Trichoptera	Insecta	Arthropoda
<i>Psychomyia flavida</i>	3.3	C, Sc		Psychomyiidae	Trichoptera	Insecta	Arthropoda
<i>Rhyacophila</i>	0.8	Pr		Rhyacophilidae	Trichoptera	Insecta	Arthropoda
<i>Neophylax</i>	1.6	Sc		Uenoidae	Trichoptera	Insecta	Arthropoda
<i>Neophylax concinnus</i>	1.6	Sc		Uenoidae	Trichoptera	Insecta	Arthropoda
	0	F		Unionidae	Unionida	Bivalvia	Mollusca
<i>Dreissena polymorpha</i>	8	F		Dreissenidae	Cardiida	Bivalvia	Mollusca
<i>Corbicula</i>	6.3	F		Corbiculidae	Veneroidea	Bivalvia	Mollusca
<i>Pisidium</i>	6.8	F		Sphaeriidae	Veneroidea	Bivalvia	Mollusca
<i>Sphaerium/Musculium</i>	7.7	F		Sphaeriidae	Veneroidea	Bivalvia	Mollusca
<i>Ferrissia</i>	6.9	Sc		Ancylidae	Limnophila	Gastropoda	Mollusca
<i>Ferrissia rivularis</i>		Sc		Ancylidae	Limnophila	Gastropoda	Mollusca
<i>Laevapex</i>	7.3	Sc		Ancylidae	Limnophila	Gastropoda	Mollusca
<i>Laevapex fuscus</i>		Sc		Ancylidae	Limnophila	Gastropoda	Mollusca
<i>Rhodaemea</i>		Sc		Ancylidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea</i>	6	Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea</i>		Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea</i>	8	Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea humilis</i>		Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea modicella</i>		Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Lymnaea obrussa</i>		Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Pseudosuccinea</i>	7.2	Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Pseudosuccinea columella</i>		Sc		Lymnaeidae	Limnophila	Gastropoda	Mollusca
<i>Physella</i>	9.1	Sc		Physidae	Limnophila	Gastropoda	Mollusca
<i>Physella anatine</i>		Sc		Physidae	Limnophila	Gastropoda	Mollusca

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Physella goodrichi</i>	8	Sc		Physidae	Limnophila	Gastropoda	Mollusca
<i>Physella gyrina</i>		Sc		Physidae	Limnophila	Gastropoda	Mollusca
<i>Physella heterostropha</i>		Sc		Physidae	Limnophila	Gastropoda	Mollusca
<i>Physella pomilia</i>		Sc		Physidae	Limnophila	Gastropoda	Mollusca
<i>Carychium exile</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Gyraulus</i>	6.5	Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Helisoma</i>	8.4	Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Helisoma subcrenatum</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Helisoma trivolvis</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Menetus</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Menetus sampsoni</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Planorbella</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Planorbula</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Promenetus</i>		Sc		Planorbidae	Limnophila	Gastropoda	Mollusca
<i>Pomatiopsis lapidaria</i>		Sc		Pomatiopsidae	Limnophila	Gastropoda	Mollusca
<i>Fontigens aldrichi</i>	2.5	Sc		Hydrobiidae	Limnophila	Gastropoda	Mollusca
		Sc		Hydrobiidae	Limnophila	Gastropoda	Mollusca
<i>Elimia</i>		Sc		Pleuroceridae	Limnophila	Gastropoda	Mollusca
<i>Elimia protosiensis</i>		Sc		Pleuroceridae	Limnophila	Gastropoda	Mollusca
<i>Goniobasis</i>		Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Goniobasis protosiensis</i>	1.6	Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Goniobasis ozarkensis</i>		Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Leptoxis</i>		Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Lithasia</i>		Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Pleurocera</i>		Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Pleurocera acuta</i>	6	Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Pleurocera alveare</i>	8	Sc		Pleuroceridae	Mesogastropoda	Gastropoda	Mollusca
<i>Valvata</i>		Sc		Valvatidae	Mesogastropoda	Gastropoda	Mollusca
<i>Campeloma</i>		Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca
<i>Campeloma subsolidum</i>		Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca
<i>Cipangopaludina</i>	6	Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca
<i>Lioplax</i>		Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca
<i>Viviparus</i>		Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca
<i>Viviparus georgianus</i>		Sc		Viviparidae	Mesogastropoda	Gastropoda	Mollusca

Genus/Species	Tolerance Value	FFG ¹	Subfamily	Family	Order	Class	Phylum
<i>Macrocotyla grandulosa</i> <i>Dugesia</i>	7.5	Pa		Chordodidae	Tricladida		Nematomorpha
		Pa		Gordiidae			Nematomorpha
		C, Pr		Dendrocoelidae			Platyhelminthes
		C, Pr		Planariidae			Platyhelminthes
		¹ Functional Feeding Groups: C=Collector, F=filterer, He=Herbivore, Pa=Parasite, Pr=Predator, Sc=Scraper, Sh=shredder					

Table 2. Fish species classifications by tolerance, trophic status, habitat, spawning, and substrate preferences.

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
American brook lamprey (ammocoete)	<i>Lampetra appendix</i>	Unk		X		X								X		X				
American brook lamprey adult	<i>Lampetra appendix</i>	I	X					X		X	X	X	X							
American eel	<i>Anguilla rostrata</i>	M-T		X		X													X	X
Arkansas saddled darter	<i>Etheostoma euzonum</i>	Unk	X								X	X								
Banded darter	<i>Etheostoma zonale</i>	I	X				X					X							X	
Banded sculpin	<i>Cottus carolinae</i>	M	X						X		X	X							X	
Bigeye chub	<i>Hybopsis amblops</i>	I-M		X	X		X					X	X						X	
Bigeye shiner	<i>Notropis boops</i>	I		X			X				X								X	
Bigmouth buffalo	<i>Ictiobus cyprinellus</i>	M		X	X	X	X							X					X	
Black buffalo	<i>Ictiobus niger</i>	M-I		X		X	X													
Black bullhead	<i>Ameiurus melas</i>	M-T		X		X			X				X	X					X	X
Black crappie	<i>Pomoxis nigromaculatus</i>	M		X		X			X				X	X	X				X	X
Black redhorse	<i>Moxostoma duquesnei</i>	I			X		X			X	X	X	X						X	

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Rifle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Blackside darter	<i>Percina maculata</i>	M		X			X			X		X	X						X	
Blackspotted topminnow	<i>Fundulus olivaceus</i>	M		X		X	X					X	X			X			X	
Blacktail shiner	<i>Cyprinella venusta</i>	Unk			X			X					X						X	
Bleeding shiner	<i>Luxilus zonatus</i>	Unk									X								X	
Bluegill	<i>Lepomis macrochirus</i>	M-T		X					X						X				X	
Blue sucker	<i>Cycleptus elongatus</i>	I			X		X			X		X	X			X			X	
Bluntnose minnow	<i>Pimephales notatus</i>	T		X		X			X			X	X	X				X		
Brindled madtom	<i>Noturus miurus</i>	I-M		X					X			X	X	X					X	
Brook silverside	<i>Labidesthes sicculus</i>	M-I		X			X										X		X	
Brown trout	<i>Salmo trutta</i>	M-I		X				X		X	X								X	X
Carmine shiner	<i>Notropis percobromus</i>	I		X	X			X			X	X				X		X	X	
Central stoneroller	<i>Campostoma anomalum</i>	M-T	X	X	X			X		X	X					X				
Chain pickerel	<i>Esox niger</i>	M		X			X								X					X

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Checkered madtom	<i>Noturus flavater</i>	Unk																		
Chestnut lamprey adult	<i>Ichthyomyzon castaneus</i>	M			X			X			X	X								X
Common carp	<i>Cyprinus carpio</i>	T		X			X								X			X	X	
Creek chub	<i>Semotilus atromaculatus</i>	T		X				X			X		X						X	X
Creek chubsucker	<i>Erimyzon oblongus</i>	M-T-I		X			X			X		X	X		X				X	
Current darter (traits from orangethroat darter)	<i>Etheostoma uniporum</i>	M		X	X			X				X							X	
Cypress darter	<i>Etheostoma proeliare</i>	M		X		X		X						X	X				X	
Duskystripe shiner	<i>Luxilus pilsbryi</i>	Unk		X	X						X								X	
Emerald shiner	<i>Notropis atherinoides</i>	M		X	X		X						X				X			
Fantail darter	<i>Etheostoma flabellare</i>	M	X						X		X	X							X	
Fathead minnow	<i>Pimephales promelas</i>	T		X					X				X	X				X	X	
Flathead catfish	<i>Pylodictis olivaris</i>	M		X					X										X	X
Flier	<i>Centrarchus macropterus</i>	M		X		X			X					X					X	

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Rifle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Freckled madtom	<i>Noturus nocturnus</i>	M-I	X		X				X			X	X						X	
Freshwater drum	<i>Aplodinotus grunniens</i>	M		X		X	X												X	X
Goldfish	<i>Carassius auratus</i>	T		X		X	X							X	X	X			X	
Gilt darter	<i>Percina evides</i>	I	X				X				X								X	
Gizzard shad	<i>Dorosoma cepedianum</i>	M-T		X	X		X									X				
Golden redhorse	<i>Moxostoma erythrurum</i>	M-I		X			X			X									X	
Golden shiner	<i>Notemigonus crysoleucas</i>	T		X		X	X									X			X	
Goldeye	<i>Hiodon alosoides</i>	I		X		X	X												X	
Gravel chub	<i>Erimystax x-punctatus</i>	M-I	X		X		X					X				X				
Green sunfish	<i>Lepomis cyanellus</i>	T-M		X		X			X						X				X	X
Greenside darter	<i>Etheostoma blennioides</i>	M-I	X					X			X								X	
Highfin carpsucker	<i>Carpiodes velifer</i>	I-M		X		X	X			X		X	X					X		

		Habitat preference					Spawning preference				Substrate Preference				Trophic status					
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Horneyhead chub	<i>Nocomis biguttatus</i>	I-M	X	X				X			X	X				X			X	
Johnny darter	<i>Etheostoma nigrum</i>	M		X					X				X	X					X	
Lake chubsucker	<i>Erimyzon sucetta</i>	M		X		X	X						X	X		X			X	
Largemouth bass	<i>Micropterus salmoides</i>	M-T		X		X			X				X	X	X				X	X
Largescale stoneroller	<i>Campostoma oligolepis</i>	M	X		X			X		X	X					X				
Least brook lamprey adult	<i>Lampetra aepyptera</i>	M-I	X					X				X	X							
Least brook lamprey ammocoete	<i>Lampetra aepyptera</i>	Unk		X		X								X		X				
Logperch	<i>Percina caprodes</i>	M					X					X	X							
Longear sunfish	<i>Lepomis megalotis</i>	I-M		X					X		X		X		X				X	
Longnose gar	<i>Lepisosteus osseus</i>	M		X		X	X								X					X
Mississippi silvery minnow	<i>Hybognathus nuchalis</i>	M-I		X		X	X						X					X		
Mooneye	<i>Hiodon tergisus</i>	I		X		X	X												X	
Mountain madtom	<i>Noturus eleutherus</i>	I	X		X				X		X	X							X	

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Rifle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Mud darter	<i>Etheostoma asprigene</i>	M			X				X			X	X						X	
Northern hog sucker	<i>Hypentelium nigricans</i>	I-M	X		X		X				X	X				X			X	
Northern studfish	<i>Fundulus catenatus</i>	I				X	X						X					X		
Orangespotted sunfish	<i>Lepomis humilis</i>	M		X					X						X				X	
Orangethroat darter	<i>Etheostoma spectabile</i>	M		X	X			X				X							X	
Ozark bass	<i>Ambloplites constellatus</i>	Unk		X					X		X							X		X
Ozark chub	<i>Erimystax harrisi</i>	I		X	X		X				X	X								
Ozark madtom	<i>Noturus albater</i>	Unk	X	X							X								X	
Ozark minnow	<i>Notropis nubilus</i>	I		X		X		X			X	X				X				
Ozark sculpin	<i>Cottus hypselurus</i>	Unk	X									X			X				X	
Ozark shiner	<i>Notropis ozarcanus</i>	Unk		X	X						X		X						X	
Paddlefish	<i>Polyodon spathula</i>	I			X		X			X							X		X	

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Pealip redhorse	<i>Moxostoma pisolabrum</i>	M	X	X	X		X			X		X							X	
Pirate perch	<i>Aphredoderus sayanus</i>	M		X		X			X					X	X				X	X
Pugnose minnow	<i>Opsopoeodus emiliae</i>	I				X			X						X		X	X		
Quillback	<i>Carpiodes cyprinus</i>	M-T		X		X	X			X		X	X					X	X	
Rainbow darter	<i>Etheostoma caeruleum</i>	M-I	X	X				X			X	X							X	
Rainbow trout	<i>Oncorhynchus mykiss</i>	M-I	X	X				X		X		X							X	X
Redear sunfish	<i>Lepomis microlophus</i>	M		X		X			X				X	X					X	
Redfin pickerel	<i>Esox americanus</i>	M		X			X								X				X	X
Redfin shiner	<i>Lythrurus umbratilis</i>	M-T		X				X						X						
Redspotted sunfish	<i>Lepomis miniatus</i>	Unk		X					X				X	X					X	
Ribbon shiner	<i>Lythrurus fumeus</i>	M																		
River carpsucker	<i>Carpiodes carpio</i>	M		X		X	X						X	X			X	X		

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Rifle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
River redhorse	<i>Moxostoma carinatum</i>	I		X	X			X			X	X							X	
Sauger	<i>Sander canadensis</i>	M		X	X	X	X			X		X	X	X				X		X
Shadow bass	<i>Ambloplites ariommus</i>	Unk		X					X			X	X	X				X		X
Shortnose gar	<i>Lepisosteus platostomus</i>	M		X			X													X
Shovelnose sturgeon	<i>Scaphirhyncus platyrhynchus</i>	M			X		X					X	X					X		
Silver redhorse	<i>Moxostoma anisurum</i>	M		X			X			X								X		
Skipjack herring	<i>Alosa chrysochloris</i>	M			X		X			X		X	X				X			
Slender madtom	<i>Noturus exilis</i>	I	X	X	X				X			X						X	X	
Smallmouth bass	<i>Micropterus dolomieu</i>	M-I		X	X				X			X						X		X
Smallmouth buffalo	<i>Ictiobus bubalus</i>	M-I		X	X	X	X									X		X		
Southern cavefish	<i>Typhlichthys subterraneus</i>	Unk																		
Southern redbelly dace	<i>Phoxinus erythrogaster</i>	M-I		X			X					X					X			

		Habitat preference					Spawning preference				Substrate Preference				Trophic status					
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Speckled darter	<i>Etheostoma stigmaeum</i>	Unk		X	X			X				X	X							
Spotted bass	<i>Micropterus punctulatus</i>	M		X	X				X			X							X	X
Spotted gar	<i>Lepisosteus oculatus</i>	M		X		X	X													X
Spotted sucker	<i>Minytrema melanops</i>	M-I		X			X						X	X					X	
Stargarzing darter	<i>Percina uranidea</i>	I			X							X							X	
Steelcolor shiner	<i>Cyprinella whipplei</i>	M-I		X	X			X			X		X						X	
Stippled darter	<i>Etheostoma punctulatum</i>	Unk		X							X								X	
Striped shiner	<i>Luxilus chrysocephalus</i>	M-T		X		X		X				X	X						X	
Suckermouth minnow	<i>Phenacobius mirabilis</i>	M	X		X		X			X	X	X	X						X	
Tadpole madtom	<i>Noturus gyrinus</i>	M-I		X		X			X		X			X			X		X	
Telescope shiner	<i>Notropis telescopus</i>	Unk		X	X						X								X	
Threadfin shad	<i>Dorosoma petenense</i>	M					X										X			

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Rifle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Walleye	<i>Sander vitreus</i>	M		X	X	X	X			X									X	X
Warmouth	<i>Lepomis gulosus</i>	M		X		X			X					X	X				X	X
Wedgespot shiner	<i>Notropis greenei</i>	Unk		X	X						X	X	X							
Weed shiner	<i>Notropis texanus</i>	I		X	X		X						X		X			X		
Western mosquitofish	<i>Gambusia affinis</i>	M-T				X													X	
White bass	<i>Morone chrysops</i>	M-T		X			X			X									X	X
White crappie	<i>Pomoxis annularis</i>	M-T		X		X			X				X	X					X	X
White sucker	<i>Catostomus commersoni</i>	T	X	X			X			X	X	X				X		X		
Whitetail shiner	<i>Cyprinella galuctura</i>	Unk			X			X			X								X	
Yellow bullhead	<i>Ameiurus natalis</i>	T-M		X		X			X				X	X	X				X	X
Yellow perch	<i>Perca flavescens</i>	M		X		X	X								X				X	X

		Habitat preference				Spawning preference				Substrate Preference				Trophic status						
Common name	Scientific name	Tolerance	Riffle	Pool	Run	Backwater	Broadcaster	Simple nester	Complex nester	Migratory	Cobble/rubble (rocky)	Gravel	Sand	Mud (silt, clay, detritus)	Vege-tation	Herbivore	Plank-tivore	Detrit-ivore	Invert-ivore	Carnivore
Yoke darter	<i>Etheostoma juliae</i>	Unk	X								X								X	
* Tolerance: I = Intolerant, M = Moderately Tolerant, T = Tolerant, Unk = Unknown.																				

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 13: Data Reporting

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP gives instructions for reporting on aquatic vegetation, invertebrate, and fish data collected at the large springs of Ozark National Scenic Riverways. The SOP describes the procedure for formatting a report, the review process, and distribution of completed reports. Efficient reporting of monitoring results is critical in assisting park Resource Managers in management decisions.

I. Report Format

Template

The report template for regional natural resource technical reports should be followed (<http://www.nature.nps.gov/publications/NRPM/index.cfm>). Natural resource reports are the designated medium for disseminating high priority, current natural resource management information with managerial application. The natural resource technical reports series is used to disseminate the results of scientific studies in the physical, biological, and social sciences for both the advancement of science and the achievement of the National Park Service's mission.

Style

Standards for scientific writing as recommended in the CBE Style Manual (1994) should be followed. Reports should be direct and concise. Refer to CBE Style Manual Committee (1994), Mack (1986), Goldwasser (1999), Strunk and White (1999), and Day and Gastel (2006).

II. Types of Reports and Review Procedure

Table 1. Summary of types of reports produced and review process. Adapted from DeBacker *et al.* 2005.

Type of Report	Purpose of Report	Primary Audience	Review Process	Frequency
Annual Status Reports for Specific Protocols	Summarize monitoring data collected during the year and provide an update on the status of selected natural resources. Document related data management activities and data summaries.	Park resource managers and external scientists	Internal peer review by HTLN staff	Annually
Executive Summary of Annual Reports for Specific Protocols	Same as Annual Status Reports but summarized to highlight key points for non-technical audiences.	Superintendents, interpreters, and the general public	Internal peer review by HTLN staff	Simultaneous with Annual Status Reports
Comprehensive Trends and Analysis and Synthesis Reports	Describe and interpret trends in individual vital signs. Describe and interpret relationships among observed trends and park management, known stressors, climate, <i>etc.</i> Highlight resources of concern that may require management action.	Park resource managers and external scientists	Internal peer review by HTLN staff	Every 5-7 years
Executive Summary of Comprehensive Trends and Analysis and Synthesis Reports	Same as Comprehensive Trends and Analysis and Synthesis Reports, but summarized to highlight findings and recommendations for non-technical audiences.	Superintendents, interpreters, and the general public	Internal peer review by HTLN staff	Simultaneous with Comprehensive Trends Analysis and Synthesis Reports

III. Distribution Procedure

Annual reports will be provided to the Resource Management staff and the Superintendent of Ozark National Scenic Riverways. Additionally, a copy will be kept on file with the HTLN office of the National Park Service, Republic, Missouri, and made available to all interested parties upon request. All data collected by the HTLN is public property and subject to requests under the Freedom of Information Act (FOIA). Reports also are converted to Adobe portable document format (pdf files) and posted on the HTLN website: (<http://science.nature.nps.gov/im/units/htln/>).

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 14: Procedures and Equipment Storage After the Field Season

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

Maintenance of sampling equipment will be necessary to maximize its life and ensure proper functioning. Poorly maintained equipment adversely affects equipment performance thus decreasing the accuracy of water quality readings and sampling efficiency. This will introduce variability into the data set. This SOP explains procedures that all field observers should be familiar with and follow after the field season is completed.

Procedures:

Equipment

- A. Clean and repair all equipment prior to return to the proper storage areas. Maintenance and storage of electronic equipment should follow manufacturer's instructions.
- B. Check sampling nets and determine if new nets must be ordered prior to the next field season.
- C. Batteries must be removed from all equipment. Rechargeable batteries are used in some equipment (*e.g.*, backpack electrofishing units). These batteries should remain on chargers when not in use.
- D. Clean the insides and outsides of all vehicles used in the field.

Paperwork and Reports

- A. All reference manuals should be re-shelved. Other reference materials and extra data sheets need to be filed in their appropriate filing cabinet.

- B. At the end of each field season, after all sampling has been completed the project manager will file a trip report with the data manager outlining hours worked, field-crew members and their responsibilities on the project, and any unique situations encountered. This information is incorporated in the database and used during data analysis, and it may be useful in identifying causes for discrepancies and inconsistencies in the data.

**Protocol for Monitoring Springs at Ozark National Scenic Riverways, Missouri.
Heartland I&M Network and Prairie Cluster Prototype Monitoring Program**

SOP 15: Revising the Protocol

Version 1.00 (03/5/2008)

Revision History Log:

Previous Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This SOP explains how to make changes to the Spring Communities Monitoring Protocol Narrative and accompanying SOPs and how to track these changes. Anyone asked to edit the Protocol Narrative or SOPs must follow this outlined procedure in order to eliminate confusion in how data is collected and analyzed.

Procedures:

- A. The Spring Communities Monitoring Protocol and accompanying SOPs for Ozark National Scenic Riverways, Missouri have used sound methodologies for collecting and analyzing data. All protocols require editing as new and different information becomes available. Required edits should be made in a timely manner and appropriate reviews undertaken.
- B. All edits require review for clarity and technical soundness. Small changes or additions to existing methods will be reviewed in-house by the Heartland Network and Prairie Cluster LTEM staff. However, if a complete change in methods is sought, then an outside review is required. Regional and national staff of the National Park Service with familiarity in spring community research and data analysis will be utilized as reviewers. Also, experts in spring community research and statistical methodologies outside of the Park Service will be used in the review process.
- C. Document edits and protocol versioning in the Revision History Log that accompanies the Protocol Narrative and each SOP. Log changes in the Protocol Narrative or SOP being edited. Version numbers increase incrementally by hundredths (*e.g.*, version 1.01, version 1.02..., *etc.*) for minor changes. Major revisions should be designated with the next whole number (*e.g.*, version 2.0, 3.0, 4.0..., *etc.*). Record the previous version number, date of revision, author of the revision, identify paragraphs and pages where changes are made, and the reason for making the changes along with the new version number.

- D. Inform the Data Manager about changes to the Protocol Narrative or SOP so the new version number can be incorporated in the Metadata of the project database. The database may have to be edited by the Data Manager to accompany changes in the Protocol Narrative and SOPs.
- E. Post new versions of the protocol on the Heartland Network internet website and forward copies to all individuals with a previous version of the affected Protocol Narrative or SOP.

Appendix A. Map and photographs of large springs at Ozark National Scenic Riverways, Missouri.

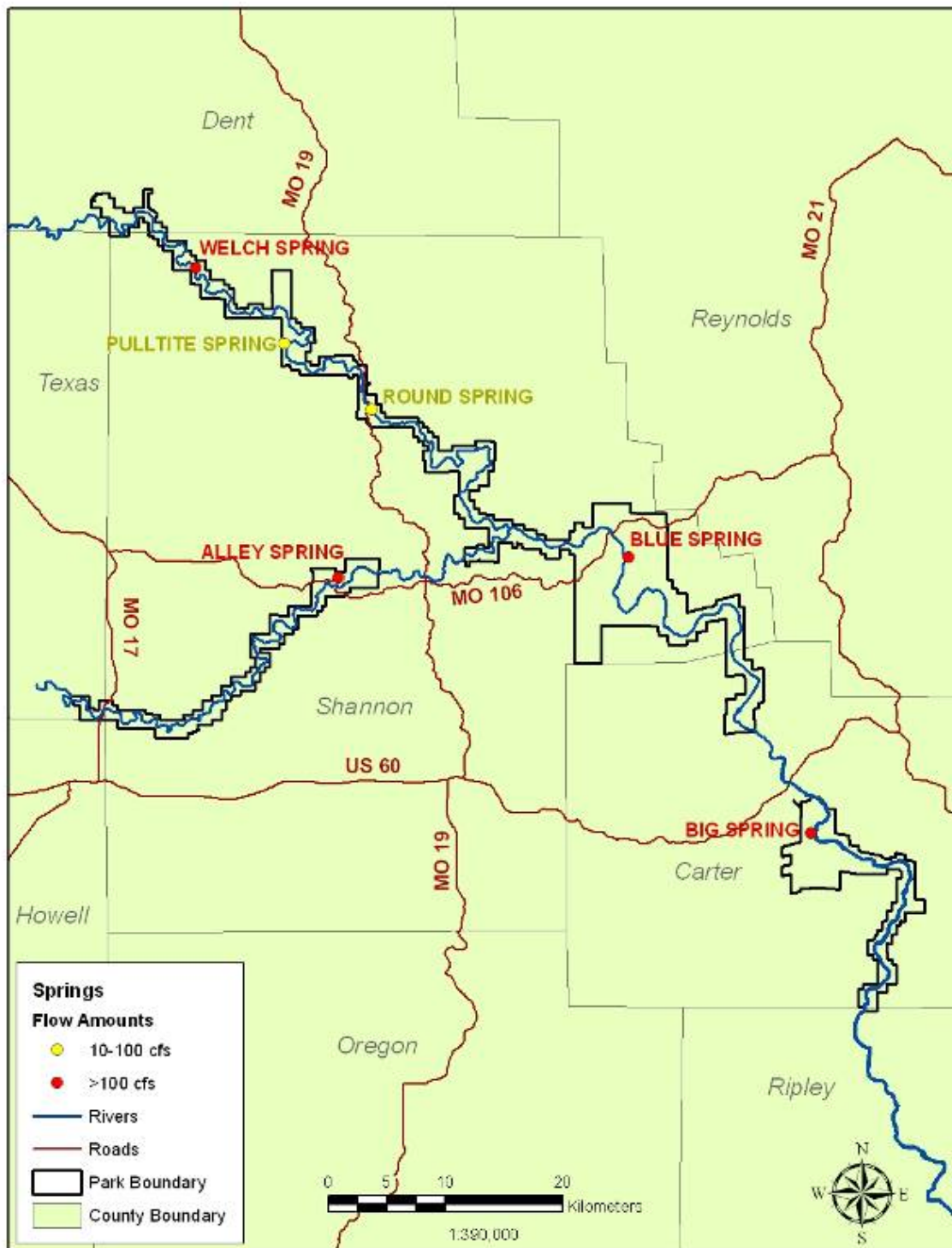


Figure 1. Map showing the general locations of large springs at Ozark National Scenic Riverways, Missouri.



Alley Spring-Downstream



Alley Spring-Upstream



Big Spring-Downstream



Big Spring-Upstream



Blue Spring-Downstream



Blue Spring-Upstream

Figure 2. Photographs of Alley, Big, and Blue springs, Ozark National Scenic Riverways, Missouri



Pulltite Spring-Downstream



Pulltite Spring-Upstream



Round Spring-Downstream



Round Spring-Upstream



Welch Spring-Downstream



Welch Spring-Upstream

Figure 3. Photographs of Pulltite, Round, and Welch springs, Ozark National Scenic Riverways, Missouri.

The NPS has organized its parks with significant natural resources into 32 networks linked by geography and shared natural resource characteristics. HTLN is composed of 15 National Park Service (NPS) units in eight Midwestern states. These parks contain a wide variety of natural and cultural resources including sites focused on commemorating civil war battlefields, Native American heritage, westward expansion, and our U.S. Presidents. The Network is charged with creating inventories of its species and natural features as well as monitoring trends and issues in order to make sound management decisions. Critical inventories help park managers understand the natural resources in their care while monitoring programs help them understand meaningful change in natural systems and to respond accordingly. The Heartland Network helps to link natural and cultural resources by protecting the habitat of our history.

The I&M program bridges the gap between science and management with a third of its efforts aimed at making information accessible. Each network of parks, such as Heartland, has its own multi-disciplinary team of scientists, support personnel, and seasonal field technicians whose system of online databases and reports make information and research results available to all. Greater efficiency is achieved through shared staff and funding as these core groups of professionals augment work done by individual park staff. Through this type of integration and partnership, network parks are able to accomplish more than a single park could on its own.

The mission of the Heartland Network is to collaboratively develop and conduct scientifically credible inventories and long-term monitoring of park “vital signs” and to distribute this information for use by park staff, partners, and the public, thus enhancing understanding which leads to sound decision making in the preservation of natural resources and cultural history held in trust by the National Park Service.

www.nature.nps.gov/im/units/htln/



The Department of the Interior protects and manages the nation’s natural resources and cultural heritage; provides scientific and other information about those resources; and honors its special responsibilities to American Indians, Alaska Natives, and affiliated Island Communities.

**National Park Service
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Natural Resource Program Center

Natural Resource Program Center
1201 Oakridge Drive, Suite 150
Fort Collins, CO 80525

www.nature.nps.gov

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