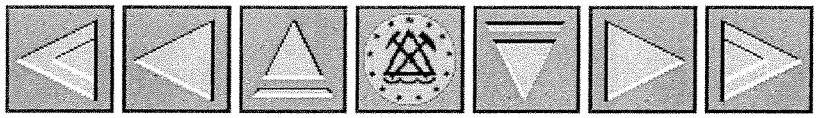

METHODS FOR COLLECTING BENTHIC INVERTEBRATE SAMPLES AS PART OF THE NATIONAL WATER-QUALITY ASSESSMENT PROGRAM

By Thomas F. Cuffney, Martin E. Gurtz, and Michael R. Meador

U.S. Geological Survey

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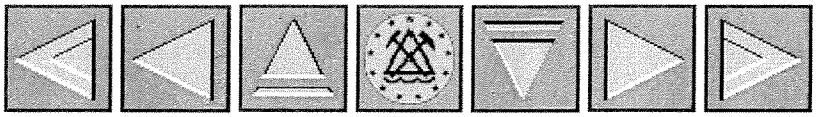
1993



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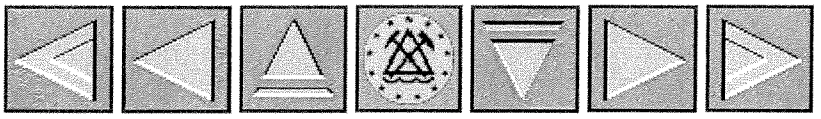

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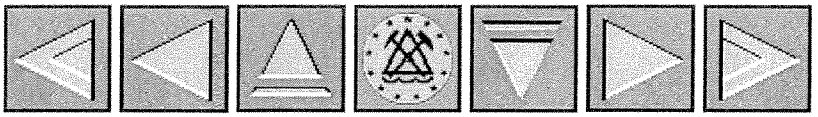
ILLUSTRATIONS

2. Matrix of instream habitat types defined by the three-tiered hierarchy of major geomorphic channel units, channel boundaries, and channel features.
3. Examples of sampling equipment used to collect benthic invertebrates.



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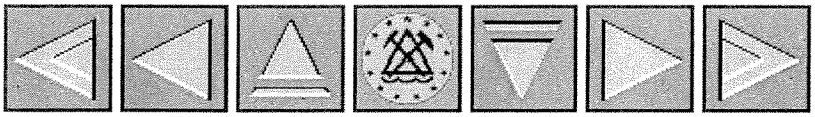

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CONVERSION FACTORS AND ABBREVIATIONS

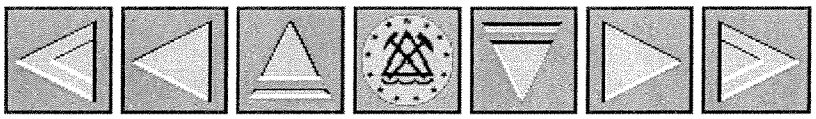
CONVERSION FACTORS AND ABBREVIATIONS

Multiply	By	To obtain
	Length	
micron (µm)	0.00003937	inch
millimeter (mm)	0.03937	inch
centimeter (cm)	0.3937	inch
meter (m)	3.281	foot
	Area	
square centimeter (cm^2)	0.155	square inch
square meter (m^2)	10.7639	square foot
square kilometer (km^2)	0.3861	square mile
	Volume	
liter (L)	0.264	gallon
	33.82	ounce, fluid
	Flow	
centimeter per second (cm/s)	0.0328	foot per second

Additional abbreviations used in this report:

gal	gallon
in.	inch
mL	milliliter
oz	ounce

Any use of trade, product, or firm names is for descriptive purposes only and does not imply endorsement by the U.S. Government.



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METHODS FOR COLLECTING BENTHIC INVERTEBRATE SAMPLES AS PART OF THE NATIONAL WATER-QUALITY ASSESSMENT PROGRAM

By Thomas F. Cuffney, Martin E. Gurtz, and Michael R. Meador

ABSTRACT

Benthic invertebrate communities are evaluated as part of the ecological survey component of the U.S. Geological Survey's National Water-Quality Assessment Program. These biological data are collected along with physical and chemical data to assess water-quality conditions and to develop an understanding of the factors that affect water-quality conditions locally, regionally, and nationally. The objectives of benthic invertebrate community characterizations are to (1) develop for each site a list of tax a within the associated stream reach and (2) determine the structure of benthic invertebrate communities within selected habitats of that reach. A nationally consistent approach is used to achieve these objectives. This approach provides guidance on site, reach, and habitat selection and methods and equipment for qualitative multihabitat sampling and semi-quantitative single habitat sampling. Appropriate quality-assurance and quality-control guidelines are used to maximize the ability to analyze data within and among study units.

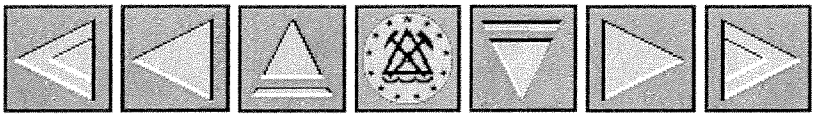
INTRODUCTION

The U.S. Geological Survey's (USGS) National Water-Quality Assessment (NAWQA) Program is designed to assess status and trends in the Nation's water quality and to develop an understanding of the major factors that affect observed water-quality conditions and trends (Hirsch and others, 1988; Leahy and others, 1990). This is accomplished by collecting biological, physical, and chemical data at sites that represent major natural and anthropogenic factors thought to control water quality in a river basin. Together these data are used to provide an integrated assessment of water quality within selected environmental settings, to assess trends in water quality, and to investigate major natural and anthropogenic factors, such as ecoregion, land use, stream size, hydrology, and geology, that influence water quality.

Background

The biological components of the NAWQA Program, ecological surveys (Cuffney and others, 1993; Meador, Cuffney, and Gurtz, 1993; Meador, Hupp, and others, 1993; Porter and others, 1993) and tissue contaminants (Crawford and Luoma, 1993) offer a number of advantages over monitoring physical and chemical water-quality constituents (Price, 1978). These include (1) increased sensitivity to a wide

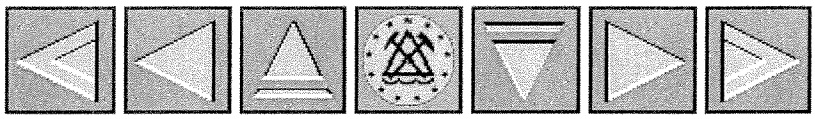
variety of natural and anthropogenic



environmental influences, such as chemical contaminants, hydrologic modifications, sedimentation, and thermal pollution; (2) greater ability to measure biological effects directly without the need to extrapolate from chemical measurements and laboratory effect studies; (3) increased analytical sensitivity as a result of bioconcentration of certain contaminants [For example, bioconcentration by a factor greater than 10,000 is possible (Phillips, 1980).]; (4) greater integration of exposure over multiple temporal and spatial scales (For example, algae integrate exposure over several millimeters and for periods of several weeks, whereas fish may integrate exposure over many kilometers and for a decade or more.); and (5) a high degree of public interest and concern, particularly regarding the consumption of contaminated fish and mollusks as well as the protection of threatened and endangered species. These characteristics make biological investigations an important supplement to the basin-wide physical and chemical water-quality investigations of the NAWQA Program.

The objective of the ecological survey component of the NAWQA Program is to characterize benthic invertebrate, fish (Meador, Cuffney, and Gurtz, 1993) and algal communities (Porter and others, 1993), as well as associated instream and riparian habitats (Meador, Hupp, and others, 1993). Benthic invertebrates (insects, mollusks, crustaceans, and worms) are important elements of ecological surveys because they tend to (1) live in, on, or near streambed sediments; (2) have, with the exception of most mollusks, life cycles (months to a few years) that are intermediate to fish (years to decades) and algae (days to weeks); and (3) be relatively sessile compared to larger organisms, such as fish. This combination of characteristics ensures that benthic invertebrates (1) respond to natural and anthropogenic environmental conditions that physically or chemically alter streambed sediments (for example, sedimentation, xenobiotics, eutrophication, or hydrologic modifications), (2) integrate effects over an approximately annual time period, and (3) characterize effects over a relatively small spatial area in contrast with fish, which may travel over long distances. These factors make benthic invertebrates well suited for use in assessing site-specific water quality and comparing spatial patterns of water quality at multiple sites, and for integrating effects that represent 6 months to a year of exposure at a site. Benthic invertebrates also are particularly useful for monitoring cumulative effects imparted to a site by conditions in the entire upstream landscape (Hynes, 1975). Consequently, these organisms are used increasingly by State and Federal agencies as a cost-effective method (Tesmer and Wefring, 1981) of assessing water-quality conditions in a regulatory context (Lenat, 1988; Ohio Environmental Protection Agency, 1988; Shackleford, 1988; Plafkin and others, 1989; U. S. Environmental Protection Agency, 1990).

Community analysis offers a number of advantages for large-scale water-quality assessments when compared with toxicity testing (American Society for Testing of Materials, 1988), biochemical characterization (Day and Scott, 1990; Hontela and others, 1991; Monod and Vindimian, 1991; Schoor and others, 1991), or direct measurement of ecological processes. For example, community surveys from natural substrates (1) directly relate to actual ambient conditions, (2) take into account a large range of species representing a variety of exposure pathways, (3) eliminate the need to culture and maintain test organisms, and (4) incorporate secondary effects that arise from the interactions of populations through competitive and predator-prey interactions. Community surveys remain the only means of



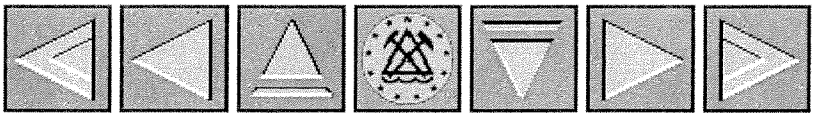
directly assessing the biological integrity of a site and the only method that is sensitive to toxicological influences and habitat degradation resulting from changes in land use or such instream disturbances as floods, navigation improvements, or substrate instability.

The distribution of benthic invertebrate species responds to natural and anthropogenic influences. Natural changes in physical and chemical conditions occur along the longitudinal axis of the river (Vannote and others, 1980), altering environmental variables (for example, riparian conditions, thermal regimes, discharge patterns, light penetration, channel gradients, sediment conditions, water and sediment chemistry) and causing benthic invertebrate communities to change. In addition, each location along the river continuum will contain a variety of habitats, such as riffles, pools, sloughs, bars, and backwaters, that differ in respect to substrate type and stability, current velocity, and water depth. Therefore, each location in the river has a range of natural conditions that, when coupled with environmental requirements of the invertebrate species, determine whether a given organism can live in a particular habitat at a particular point along the river continuum.

These patterns of species distribution are affected by natural and anthropogenic influences that alter the landscape (for example, wild fires, logging, earthquakes, agriculture, volcanic eruptions, and urbanization), modify hydrologic conditions (changes in evapotranspiration and runoff or construction of reservoirs and irrigation diversions), alter habitats (snagging operations, channel dredging, sedimentation, hurricanes), or add chemicals that are toxic or that elevate nutrient or organic loads. The challenge of ecological surveys in the NAWQA Program is to separate changes caused by natural and anthropogenic factors and relate them to water quality. This is accomplished by comparing distributions of organisms among sites that vary in natural and anthropogenic influences, including relatively pristine streams, and relating patterns of distribution to patterns of physical and chemical factors.

Purpose and Scope

The sampling methods and procedures presented here are intended to give guidance to study-unit biologists collecting benthic invertebrates as part of the USGS's NAWQA Program. Various sample-collection techniques, equipment, and data forms are presented for use at basic fixed sampling sites. These methods and techniques can be adapted for use in other components of the NAWQA Program, such as synoptic and case study sampling, or where needed in other programs of the USGS's Water Resources Division. Additional discussions and descriptions of benthic invertebrate sampling devices and methods are reported in Hynes (1970), Mason (1978), Rosenberg (1978), Adamus (1984), Merritt and Cummins (1984), Britton and Greeson (1988), and Klemm and others (1990).



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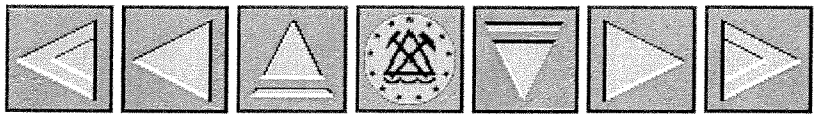
NATIONAL WATER-QUALITY ASSESSMENT PROGRAM SAMPLING DESIGN

The NAWQA Program sampling design emphasizes a multidisciplinary approach using physical, chemical, and biological tools to provide multiple lines of evidence with which to evaluate water-quality conditions. The Program focuses on a broad spectrum of at tributes and sampling approaches to collect data on (1) benthic invertebrate, fish, and algal communities; (2) stream habitats; (3) water-column measures of inorganic constituents (major ions, trace elements, nutrients), physical characteristics (suspended sediment, conductance, temperature), radionuclides, and organic compounds; (4) trace elements and organic compounds in bed material and aquatic biota; and (5) hydrology. The foundation of the Program's sampling design is the 60 study units distributed across the conterminous United States, Alaska, and Hawaii. Each study unit consists of one or more coupled river-basin aquifer systems encompassing from 3,100 to more than 155,000 km*2* (Leahy and Wilber, 1991). Study units conduct water-quality investigations for 4 to 5 years, followed by 5 years of low-level monitoring, with the cycle repeated perennially (Leahy and others, 1990). Activities are staggered so that approximately one-third of the study units are in an intensive data-collection phase each year. Study-unit investigations consist of four main components: (1) retrospective analysis and reconnaissance, (2) occurrence and distribution assessment, (3) assessment of long-term trends and changes, and (4) source, transport, fate, and effect studies.

Retrospective analysis consists of a review and analysis of existing water-quality data (physical, chemical, and biological) within the study unit. This effort provides an historical perspective on water-quality conditions and assists in the identification of major natural and anthropogenic factors that control water quality within the study unit. Analysis of existing information allows project personnel to examine a wide range of environmental-variable combinations associated with specific land areas and provides baseline information to assist in identifying candidate sampling locations within this range of environmental-variable combinations. Sampling locations are chosen following a reconnaissance of candidate sampling locations.

A reconnaissance consists of a rapid visual assessment of a location, including evaluation of stream access, stream habitat conditions, proximity of major natural or anthropogenic stream influences, and methods and equipment appropriate for conducting various types of sampling at that location. The reconnaissance is used by project personnel to gain familiarity with watershed features of the study unit and to evaluate and select candidate locations for subsequent sampling to determine biological, chemical, and physical characteristics of streams. This subsequent, integrated sampling effort is known as an occurrence and distribution assessment.

The occurrence and distribution assessment characterizes geographic and seasonal distributions of water-quality conditions in relation to major natural and anthropogenic features. This assessment fills crucial gaps in existing data for each study unit. The design of water-quality investigations conducted during occurrence and distribution assessment represents a balance between providing study units with the flexibility to address issues of local importance and maintaining the consistency in data collec tion, sampling approaches,



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and spatial and temporal resolution to allow for comparisons among study units (national synthesis). The occurrence and distribution assessment serves as a basis for designing field activities to evaluate long-term trends in water-quality conditions as well as for designing source, transport, fate, and effect studies.

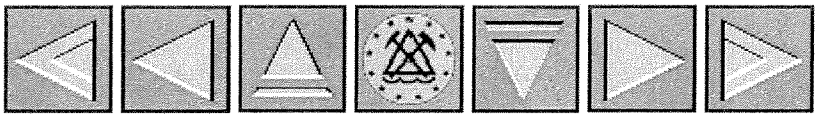
Long-term trends and changes in selected water-quality characteristics are assessed by time series of regular measurements or repeated samplings. In many study units, assessments of long-term trends and changes are conducted in a few basins that are chosen to represent selected environmental settings. Locations selected for monitoring of long-term trends and changes are chosen on the basis of results of the retrospective analysis, reconnaissance, and occurrence and distribution assessment.

Source, transport, fate, and effect studies are conducted to test hypotheses and examine specific issues about characteristics and causes of any water-quality degradation. These studies are directed at high-priority water-quality issues for individual study units and the Nation. The results of these studies are accumulated from multiple study units and used to link assessments of water-quality status and trends to specific causes and processes by example and inference. Source, transport, fate, and effect studies are designed by project personnel in individual study units and are conducted at a wide range of spatial and temporal scales.

Occurrence and distribution sampling includes two distinct types of sampling sites: basic fixed sites and synoptic sites. Basic fixed sites are geographically "fixed" sites at which a broad suite of chemical constituents, along with continuous discharge measurements and ecological surveys, is measured over relatively long time periods (generally, 6 months to 3 years, depending on the constituent). Basic fixed sites form the basis for long-term trend, transport, and integrated physical, chemical, and biological studies within and among cycles of the NAWQA Program. Synoptic sites are typically nongaged sites where one-time collections of a limited number of chemical and biological measurements are made with the objective of answering questions regarding source, occurrence, or spatial distribution.

SAMPLING DESIGN FOR BENTHIC INVERTEBRATES

Ecological surveys characterize biological communities (fish, algae, and benthic invertebrates) and stream habitats at basic fixed sites chosen to represent combinations of major natural and human engendered factors thought to significantly influence water quality nationally and within the study unit. The communities and habitat conditions associated with a site are characterized within a defined length of the stream referred to as the "sampling reach." This approach provides a common spatial scale upon which to assess community and habitat characteristics.



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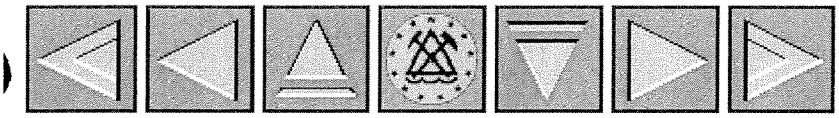
Establishing Sampling Reaches

The location and length of the sampling reach are determined by a combination of geomorphic characteristics (that is, repeating geomorphic channel units (Meador, Hupp, and others, 1993) and fish sampling considerations (Meador, Cuffney, and Gurtz, 1993). Composite qualitative and semi-quantitative samples are collected within each sampling reach to characterize the benthic invertebrate community. Typically, a single sampling reach is established at each site. However, three sampling reaches are established at a subset of sites (intensive ecological assessment sites) in order to assess variability among sampling reaches.

The primary determinant of the length of the sampling reach is the presence of repetitions of two geomorphic channel units such as a sequence of pool, riffle, pool, riffle. Only those geomorphic channel units (riffle, run, and pool) that cover greater th an 50 percent of the active channel width are considered when determining the length of the reach. When repetitions of geomorphic channel units are not present or occur at intervals of more than 1,000 m, then the length of the reach is determined to be 20 channel widths, based on the width of the channel at the boundary of the reach. Theoretically, this length represents at least one complete meander wavelength (Leopold and Wolman, 1957). Regardless of the method used to establish sampling reach length, the minimum and maximum acceptable lengths are 150 m and 300-500 m, respectively, for wadeable sites and 500 m and 1,000 m for nonwadeable sites (Meador, Hupp, and others, 1993).

The location of each sampling reach is related to a durable reference point, such as a stream gage or bridge pier (Meador, Hupp, and others, 1993), that is used to permanently define the location of the sampling reach. Sampling reaches are located where conditions, primarily instream and riparian habitats, are representative of the local area and support objectives for which the site was chosen (for example, representativeness of a certain land use, agricultural practice, or reference condition). In order to meet these objectives, the sampling reach might be located above, below, or adjacent to the site location as long as the water chemistry and hydrologic data collected at the site accurately reflect conditions within the sampling reach or reaches.

A hypothetical intensive ecological assessment (IEA) site consisting of a basic fixed site associated with multiple sampling reaches is shown in figure 1. Each sampling reach is composed of repeating geomorphic units, two pools (shaded areas) and two riffles (unshaded areas). In this example, sampling reach "A" is located above, sampling reach "B" is located at, and sampling reach "C" is located below the basic fixed site. Alternatively, the study-unit biologist might decide to locate all three sampling reaches above or below the basic fixed site, so long as there are no significant intervening changes in water chemistry, hydrology, or habitat conditions among sampling reaches. Where possible, multiple sampling reaches are separated by a minimum of 15 0 m.



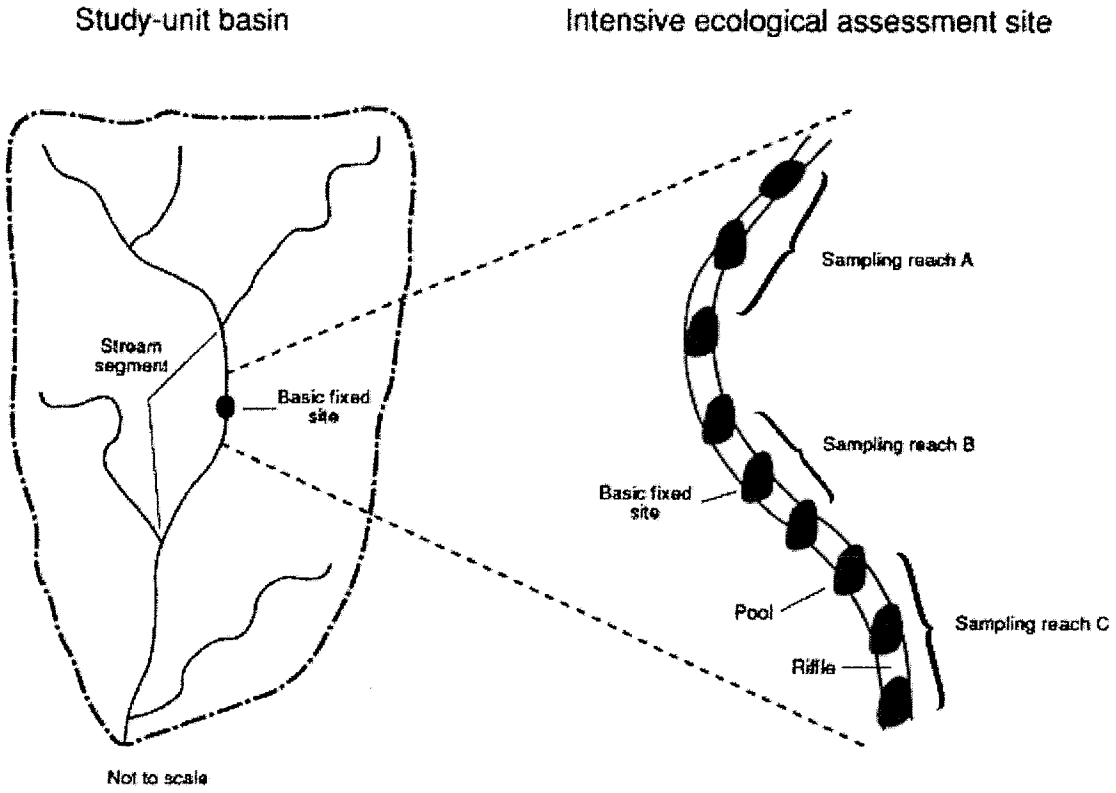
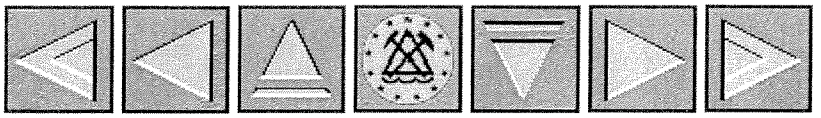


Figure 1.--Hypothetical location of a basic fixed site and three associated sampling reaches used for intensive ecological assessments.



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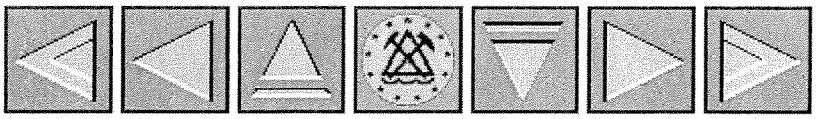
Types of Samples Collected

Qualitative and semi-quantitative sampling is conducted within each sampling reach associated with a basic fixed site. Qualitative sampling, which involves collecting invertebrates from as many different instream habitats as possible, is intended to provide a list of taxa present in the sampling reach. Semi-quantitative sampling is intended to provide a measure of the relative abundance of each taxon present in two contrasting habitat types within the sampling reach. These samples, along with the corresponding chemical and physical data, are used to (1) characterize the community within the sampling reach, (2) compare reaches among environmental settings, (3) compare changes in communities over time, and (4) couple physical and chemical water-quality characteristics with biological characteristics.

More intensive sampling is conducted at a subset of four to six sites to assess spatial variability among reaches and short-term temporal variability at a site. At these IEA sites, three sampling reaches are established to represent environmental conditions associated with the basic fixed site. One sampling reach is sampled in each of 3 successive years to estimate short-term temporal variability. Two additional sampling reaches are sampled in 1 year to assess the magnitude of reach-to-reach variability. Sampling at IEA sites supplements ecological surveys by providing a means to assess and compare variability locally, regionally, and nationally. These sites are chosen, to the extent possible, with the intent of characterizing variability across the range of conditions exemplified by the study unit.

In the example presented in figure 1, the sampling reach containing the basic fixed site (sampling reach B) is sampled in all 3 years to assess short-term temporal variability. The other two reaches (A and C) are sampled during only 1 year, typically the first or second year of sampling, to assess reach-to-reach variability. The study-unit biologist selects which one of the three sampling reaches to use for multiple-year sampling based on such criteria as ease of access and representativeness of the sam pling reaches.

The national scope of the NAWQA Program ensures that characterizations of benthic invertebrate communities are done in streams and rivers where physical and chemical characteristics vary widely in response to local and regional differences in environmental settings and human influences. Consequently, no single sampling technique or device is appropriate for all sites and instream habitats across the Nation. Therefore, a variety of techniques and sampling gear is recommended, based on the type of sample being collected (qualitative or semi-quantitative) and the physical conditions at the collecting site, such as water depth, current velocity, and bed materials. Likewise, no "standard," nationally consistent instream habitat, such as a riffle, exists. Consequently, sampling focuses on a qualitative characterization of the sampling reach supplemented with semi-quantitative characterization of standard "response" habitats. Standard response habitats include the taxonomically richest instream habitat with in the sampling reach and a contrasting faunistically impoverished habitat, which is typically, but not always, a slow-flowing, depositional habitat.



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Selecting Sampling Sites and Habitat Types

Sample site selection involves three elements: (1) locating the basic fixed site where chemical and flow data are available; (2) establishing one or more sampling reaches; and (3) identifying and selecting specific locations of instream habitat types with in each sampling reach from which invertebrate samples are taken. The first element of site selection is a cooperative effort among the discipline groups of the study-unit team to locate sites that represent the set of environmental conditions deemed important for controlling water quality in the basin. Retrospective information is an important component of this element of site selection, as is input from liaison committees and other local experts. This element of site selection is crucial to the success of local, regional, and national synthesis efforts. Sites are chosen to represent combinations of natural and anthropogenic factors thought to collectively influence the biological, physical, and chemical characteristics of water quality in the study unit and to be of importance locally, regionally, or nationally.

The second element involves establishing sampling reaches that are used to characterize conditions associated with the basic fixed sites. The length of the sampling reach is established by guidelines set forth in the stream habitat assessment protocol (Meador, Hupp, and others, 1993). Ideally, each sampling reach includes multiple examples of the major geomorphic features of the stream segment (for example, two pool-riffle sequences) and might be located entirely above or below the basic fixed site, or might encompass it. Major discontinuities in channel or riparian characteristics and intervening point sources within or among the sampling reaches associated with a basic fixed site are avoided. Theoretically, ecological survey sampling is preceded by at least 1 year of antecedent physical and chemical data collection to maximize the integration of these data sets. The reconnaissance effort plays a major role in locating and selecting sampling reaches that represent conditions typical of the areas associated with basic fixed sites.

The third element of site selection involves identifying and locating appropriate instream habitat types. Instream habitat types are broadly defined on the basis of a hierarchical grouping consisting of three tiers: major geomorphic channel units, major channel boundaries, and major channel features (table 1). The highest level of habitat organization is the major geomorphic channel unit: riffle, run, or pool. The second level is based on the influence of margins on the distribution of organisms, particularly in large rivers, and subdivides riffles, runs, and pools into channel, channel margin, and island margin areas (Thorp, 1992). Margins, loosely defined as instream areas associated with the edges of main or secondary channels and islands, are typically depositional, subhorizontal fluvial surfaces, with reduced current velocity as compared with the adjacent channel area. Margins are influenced heavily by the streambanks and typically contain elements directly derived from the streambank, such as root wads, snags, and terrestrial vegetation that trails into the water. Channels tend to be less influenced by the channel banks and represent the main or secondary flow path of the river. The extent of the margin will be influenced by river stage, the size of the river, and channel characteristics. For example, margins might be (1) a substantial proportion of the width of small streams but only a very small proportion of the width of large rivers, (2) greatly reduced on the outside of stream meanders but extensive on the inside of stream meanders, and (3) abundant at low flows but inaccessible at

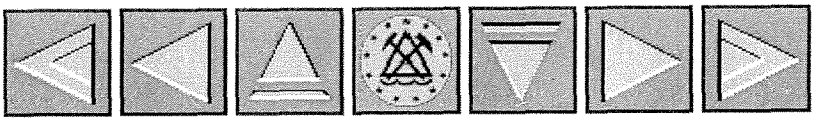

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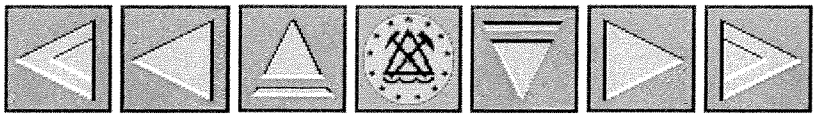
Table 1.--*Description of the elements that define the three-tiered hierarchy of instream habitat types targeted for qualitative and semi-quantitative sampling*

Tier I	
Major geomorphic channel units	**Description**
Riffle	Turbulent flow; shallow, coarse-grained substrate
Run	Laminar flow, less turbulent; variable depth and substrate
Pool	Very low current velocity; relatively deep, depositional, accumulations of fine sediment particles.
Tier II	
Major channel boundaries	**Description**
Channel	Flow paths associated with the main and secondary river channels
Channel margin	Sub-horizontal, fluvial areas associated with the streambanks
Island margin	Sub-horizontal, fluvial areas associated with the banks of islands
Tier III	
Major channel features	**Description**
Natural bed	Natural bed materials without extensive macrophyte beds
Manufactured bed	Revetments, riprap, or other manmade bed materials predominate.
Slough	Remnants of abandoned river channels that connect with the main channel even at normal low flows.
Macrophyte bed	Growths of emergent or submergent aquatic macrophytes
Woody snag	Trees, branches, or other woody debris of terrestrial origin that extend into the water column.
Bars	Shallow, gently sloping sand or gravel bars primarily associated with channel edges or major changes in water velocity.



high flows. The importance of differentiating margins is that marginal areas may contain richer communities of organisms than channel areas as a result of the increased complexity of the stream margins. This circumstance is particularly true of large, deep rivers.

The final level of the habitat hierarchy deals with major channel features that are thought to be important in the distribution of benthic invertebrates and that can be sampled discretely for invertebrates. Six categories are recognized: natural bed, ma nufactured bed, slough, macrophyte bed, woody snag, and bar. Natural bed refers to areas where natural bed materials predominate and where macrophytes are not a dominant feature. Manufactured bed refers to substrates that are created by man, such as revetments, levees, junk cars, riprap, dams, fish weirs, and bridge piers. Sloughs are remnants of abandoned river channels that are connected with the main channel even at normal low flows. Sloughs that are isolated at low flows tend to diverge biologically and chemically from conditions in the river and are not considered here as an instream habitat. Macrophyte beds are areas where emergent or submergent aquatic plants dominate and invertebrate communities are expected to contain organisms dependent upon such plants. Woody snags refer to trees, branches, or other woody debris of terrestrial origin that extend into the water column either from the streambank or streambed. Bars are shallow, gently sloping sand or gravel ridges primarily associated with channel edges or major changes in water velocity. Bars, when exposed at low flows and vegetated, can resemble islands. However, islands typically have woody vegetation and are at an elevation equal to or above that of the surrounding flood plain. Collectively, this three-tiered hierarchy describes 54 possible habitat types. However, sloughs are restricted to channel and island margins, so only 51 habitat types are available for qualitative and semi-quantitative sampling (fig. 2).

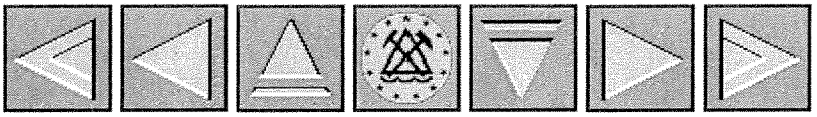
Major geomorphic channel units	Major channel boundaries	Major channel features					
		Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Riffle	Main channel			■			
	Channel margin						
	Island margin						
Run	Main channel			■			
	Channel margin						
	Island margin						
Pool	Main channel			■			
	Channel margin						
	Island margin						

EXPLANATION

□ HABITAT TYPES THAT ARE POSSIBLE

■ HABITAT TYPES THAT ARE NOT POSSIBLE

Figure 2.--Matrix of instream habitat types defined by the tree-tiered hierarchy of major geomorphic channel units, channel boundaries, and channel features.



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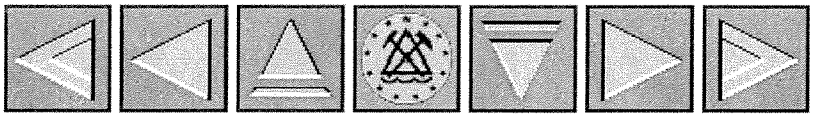
Following guidelines for establishing the size and location of the sampling reach (Meador, Hupp, and others, 1993) should lead to the inclusion of the majority of instream habitat types typical of the stream at a given location. Rarely will all 51 instre am habitat types be present. Consequently, it is a good practice to use the instream habitat type matrix during site reconnaissance to determine which habitats are present and what type of equipment is needed to sample each one. Because qualitative sampling involves all instream habitat types that are present and accessible within the sampling reach, no guidance is required for selecting one habitat type over another, as contrasted with semi-quantitative sampling. However, the types of habitats present and their abundance within the sampling reach are important in determining the proper approach to qualitative multihabitat (QMH) sampling.

Because of the complexity and uncertainties associated with reach-based proportional qualitative sampling, the NAWQA Program limits the objective of qualitative sampling to the development of a taxonomic list of organisms present within the sampling reach . Consequently, roughly equivalent effort (for example, time spent sampling, number of samples collected, or area sampled) is put into sampling each habitat type. This approach maximizes the representation of small habitats and prevents the over-represe ntation of areally extensive habitat types. The instream habitat types sampled and the sampling gear are recorded on the qualitative-sampling field data sheet.

The selection of appropriate instream habitat types for semi-quantitative sampling requires a considerable amount of guidance to determine which of the many habitat types present within a sampling reach are sampled. Semi-quantitative samples supplement q ualitative samples by providing data on the presence and relative abundance of invertebrates in two contrasting instream response habitats: (1) a habitat that supports, in the absence of human influences, the richest assemblage of invertebrates within the sampling reach (for example, a riffle in shallow, coarse-grained, high-gradient streams; or snags in sandy-bottomed, Coastal Plain streams); and (2) a habitat where organisms are most likely to be exposed to sediment-borne contaminants for extended periods of time, typically a depositional area such as a pool where particulate-borne contaminants tend to accumulate.

Characterizing communities in these two contrasting instream habitat types is important because these habitat types differ widely in the duration and pathways of contaminant exposure and potential for species loss. The species-rich habitat is expected to be highly sensitive to water-quality changes because it can support a diverse assemblage of species that display a wide range of sensitivities to water-quality changes. Therefore, the community in this habitat has the potential for greater change than less species-rich communities. However, species-rich habitats typically occur in erosional areas and may not be sites where exposure to sediment-borne contaminants, in either concentration or duration, is greatest.

The fauna of depositional areas are of interest because, even though they are generally less rich taxonomically, they are probably exposed to greater concentrations of sediment-borne contaminants for longer periods of time than the fauna of erosional areas. Consequently, communities in depositional areas may respond to contaminants before the



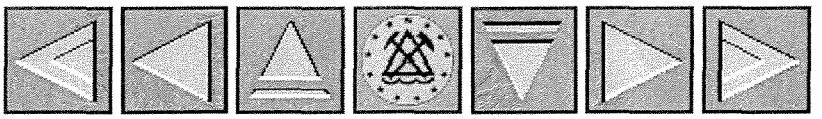
more sensitive, but less exposed, faunistically richest instream habitat types. In addition, depositional habitats, such as pools, represent the most prevalent types of habitat across the Nation and may facilitate national and regional comparisons. Consequently, the structures of both types of communities are important to the determination of biological water-quality conditions (Kerans and others, 1992).

The types of instream habitats sampled, particularly the habitat identified as being faunistically richest, vary with the geographic location of the stream and the longitudinal position of the sampling reach along that stream. In wadeable streams and rivers, the richest habitats are probably found in coarse-grained, fast-flowing riffles (the riffle, main-channel, natural-bed habitat type in fig. 2), whereas fine-grained, organically rich pools offer the highest likelihood of exposure to sediment-borne co ntaminants (the pool, channel-margin, natural-bed habitat type in fig. 2). Larger, nonwadeable rivers, and sandy-bottomed, Coastal Plain streams probably do not have coarse-grained riffles but still contain fine-grained, organically rich pools. Consequently, an alternative richest habitat is identified and sampled in these systems. Such alternative habitats are chosen from the list presented in figure 2, in consultation with the regional biologist, liaison teams, and local biologists. (Details are provided in the section on Recommendations for Sampling Benthic Invertebrates.) Prior to collection of samples, each basic fixed site is visited to determine access, verify which of the 51 types of habitats are available for qualitative and semi-quantitative sampling, and establish the locations of the three sampling reaches. Most of these tasks should be accomplished during the on-site reconnaissance.

Appropriate Season and Hydrologic Conditions for Sampling

The appropriate season and hydrologic conditions for sampling are determined primarily by the life-history characteristics of the aquatic insects that dominate, at least numerically, in most riverine benthic invertebrate communities of North America. Various environmental factors influence insect life-history patterns and are considered in the selection of an appropriate sampling time (Hynes, 1970; Sweeney, 1984). Ideally, sampling occurs at a time of year when the majority of insects are at or near maturity and few species are in early instars or resting stages (for ex ample, eggs, pupae, or diapausing larvae). Early instars are problematic because they generally lack the morphological features necessary for identification to genus or species and may be difficult to collect because of their small size. The resting stages of most insects are either difficult to identify because there are no standardized taxonomic keys (for example, eggs) or difficult to collect because many pupae or diapausing larvae move into the hyporheos or streambanks where they are missed by stand ard collection procedures. Therefore, site-to-site differences in community development caused by differences in physical factors, such as temperature (Vannote and Sweeney, 1980; Ward and Stanford, 1982), dissolved oxygen (Nagell, 1981), and discharge (Patterson and Vannote, 1979; Wiggins and others, 1980), need to be considered in the selection of an appropriate sampling season and in the interpretation of biological data.

Water temperature is the primary physical factor directly influencing the rate of development of invertebrates and reproduction (Vannote and Sweeney, 1980). Development is commonly expressed as the cumulative number of degree days required to complete the aquatic portion of an insect's life cycle. Cumulative degree days are calculated as the sum of



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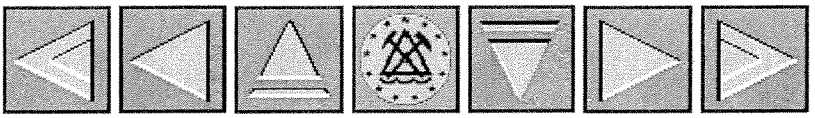
the average daily water temperatures to which organisms are exposed. A species in a warmer downstream reach typically matures earlier than does an upstream population of the same species and may actually have multiple generations in a year compared to only one generation a year at the colder upstream site (Lehmkuhl, 1973; Newell and Minshall, 1978; Mackay, 1979). In temperate climates, the period of maximum community maturity and richness typically ranges from late fall to early spring, depending upon local and regional factors that influence temperature, such as elevation and latitude. In addition to this fall-winter community, most temperate streams have a spring-summer community that is generally less rich taxonomically but whose period of maximum maturity closely approximates summer low-flow periods. Characterization of both fall-winter and spring-summer communities is desirable. However, the choices of when and how often to sample must be made in conjunction with the other elements of the NAWQA Program.

Because of the importance of water temperature in insect development, the timing of sample collection must take water temperature into consideration to account for developmental differences among sites. Water temperatures, at least daily maximum and minimum, need to be monitored at ecological survey sites associated with basic fixed sites. These data are useful in scheduling sampling activities, making comparisons within and among basins, and interpreting study-unit, regional, and national differences.

Current and antecedent discharge conditions must also be considered in determining the appropriate time to collect samples. Access to the sampling site can be limited during seasonal high-flow conditions. Unusually high flows can wash out or bury substantial parts of the benthic invertebrate community and redistribute, create, or eliminate instream and riparian habitats within the sampling reach (Irvine, 1986; Resh and others, 1988; Poff and Ward, 1989). Invertebrate communities require a period of time to adjust following such unusually high discharges. Therefore, sampling should be delayed approximately 4 weeks following a discharge event with a recurrence interval greater than 5 years. This delay reduces the likelihood of misinterpreting channel habitat characteristics and making errors such as sampling substrates that were dry prior to the increase in flow. Extreme flows with recurrence intervals greater than 10 years can so alter the physical conditions in the sample reach that many years or de cades can be required for communities to return to "normal" conditions. In such cases, recovery of the community probably would not occur within the current NAWQA Program cycle, so additional delays in sampling would make little difference in assessing the community. Subsequent interpretation of community structure in these circumstances must take into consideration the unusual antecedent discharge and altered habitat conditions. Selecting a season of minimum-flow variability for ecological sampling minimizes the probability of encountering an extremely high discharge before or during sampling.

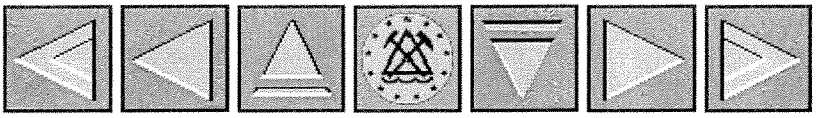
The logistical challenges of organizing field crews, especially when arrangements have been made for assistance from other agencies or researchers, can cause major difficulties in implementing the strategy of maximizing community development and minimizing the influence of antecedent high flows. Postponement of a field effort can be complicated by an even greater flow in the intervening period, especially when local climatic conditions are highly variable. Also, flow conditions are seldom uniform across basins; thus, part of a basin



may experience a high flow that exceeds a 10-year recurrence interval while the majority of the basin experiences no significant change. In these cases, the planned sequence of sampling sites may need to be altered to allow those sites that experienced the high flow to be sampled near the end of the field sampling period. Professional judgment and a fair amount of luck are required in making decisions about postponing field sampling. It is important to consider that peak flows do influence community structure and that this influence depends on factors such as the magnitude of the peak flow, duration of the high flow, and frequency of high flows that scour the channel. When sampling cannot be postponed after extreme flows, quality of the data can decline.

Sampling during normal low- and stable-flow periods, if compatible with life-history characteristics, is the preferred time for benthic invertebrate sampling. Sampling during low flow has a number of advantages including increased accessibility to the river, a reduced dependency on labor-intensive deep-water sampling techniques, and increased confidence that all parts of the wetted channel have been continuously part of the aquatic habitat. In contrast, some instream habitats, such as snags and stream m argins, that are accessible during high water may actually be out of water for most of the year. However, sampling during periods of abnormally low flow should be avoided because extreme low flows may affect invertebrate communities by reducing current velocities, decreasing the amount of instream habitat, increasing water temperature, decreasing dissolved-oxygen concentrations, increasing contaminant concentrations, and indirectly altering food resources and biotic interactions. Consequently, unusual l ow flows need to be identified and factored into the interpretation of the biological data in much the same manner as unusually high discharges. Regional characteristics must also be considered, particularly where streams are ephemeral or where flow characteristics, such as current velocity, change dramatically during normal summer low flows. For example, Coastal Plain swamp streams are best sampled at higher winter flows when conditions are suitable for supporting aquatic communities. Sampling these systems during normal summer low flows is generally not productive because conditions, such as current velocity, dissolved-oxygen concentration, and temperature, are not suitable for sustaining aquatic communities even at unimpaired reference sites (D.R. Lenat, North Carolina Division of Environmental Management, oral commun., 1992). To aid in interpreting discharge effects, all ecological survey sites associated with basic fixed sites should be continuously gaged for at least 6 months prior to sampling and throughout the intensive sampling period.

Other factors to consider in selecting the appropriate sampling season include life-history characteristics of other aquatic taxa, seasonal human activities, and site access. Spawning and migration periods of anadromous fish, especially threatened or end angered species, should probably be avoided. Seasonal agricultural practices, such as applications of pesticides and fertilizers, soil preparation, and irrigation patterns, that affect loads of sediment, nutrients, and pesticides must be considered in the selection of an appropriate sampling season. In addition, seasonal road conditions may limit site access during certain parts of the year. Subbasins with different climatic or hydrologic characteristics may need to be sampled at different times of the year. If this is the case, then an attempt must be made to link the subbasin sampling on the basis of temperature-dependent organism development (that is, sampling at periods of similar accumulated degree-days). The final selection of



sampling season is a compromise based on all of the above factors and considers retrospective data and input from the liaison committees, regional biologists, North Carolina Ecology Group, local biologists, and national synthesis teams. Once the appropriate sampling seas on has been determined for each subbasin, all sites are sampled within a 3- or 4-week period. Ecological surveys in subsequent years or subsequent NAWQA Program cycles are conducted within the chosen season (consistent accumulated degree-days) or include that season to ensure year-to-year data comparability. Eventually, sampling should be expanded to include two seasonal periods (fall-winter and spring-summer) to accommodate seasonal shifts in community structure, particularly in heavily forested stream basins.

METHODS FOR COLLECTING BENTHIC INVERTEBRATES

Many types of sampling equipment and various techniques have been developed for the collection of benthic invertebrate samples. The proper choice of sampling equipment and technique depends on the water depth, current velocity, and type of bed material to be sampled and on whether the sample is intended to provide qualitative or semi-quantitative data. The following sections establish guidelines for collecting samples. The study-unit biologists are responsible for applying and modifying these guidelines as conditions within their study areas warrant. These modifications are made in consultation with regional biologists, North Carolina Ecology Group, and local invertebrate biologists.

Qualitative Multihabitat Sampling Methods

The objective of qualitative multihabitat (QMH) sampling is to obtain as complete a list of invertebrate taxa present in a sampling reach as is possible in the time available, usually about 1 hour. To accomplish this, individual samples are composited f rom as many of the 51 instream habitat types (fig. 2) as are present and accessible within the sampling reach. This composited sample, together with the semi-quantitative samples, represents the aggregation of organisms that exist in the sampling reach.

The primary sampling gear used to collect QMH samples in wadeable streams is a D-frame kick net (fig. *a*3A) equipped with a 210-µm mesh net. This net is used to collect samples by kicking, dipping, or sweeping in a manner appropriate for the instream habitat type being sampled. QMH sampling encompasses as many habitat types as possible, including those habitat types sampled by semi-quantitative methods. When possible, equal sampling effort is applied to each habitat type within the sampling reach. This is usually accomplished by dividing the available 1-hour sampling time equally among the instream habitat types. This strategy can be adjusted to accommodate complicated collection methods, such as deep-water and diver-assisted sampling. In these situations, dividing sampling effort on the basis of the area or number of samples collected is preferred.

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*a*Photographs of samplers A-C, F-J, and L are courtesy of Wildlife Supply Company, Saginaw, Mich.; sampler K is modified from Gale and Thompson (1975); samplers D-E and M-O are modified from Merritt and Cummins (1984).

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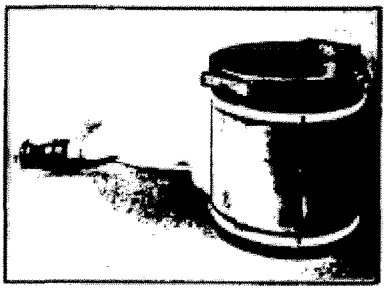
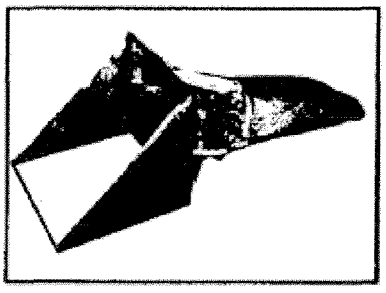
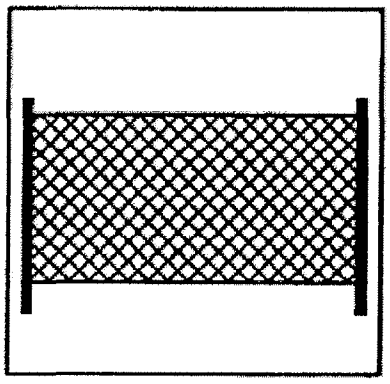
A. D-frame net and handle



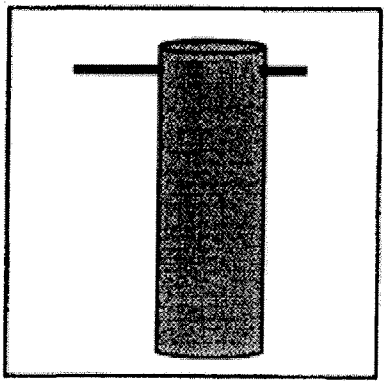
B. Hess sampler



C. Surber sampler

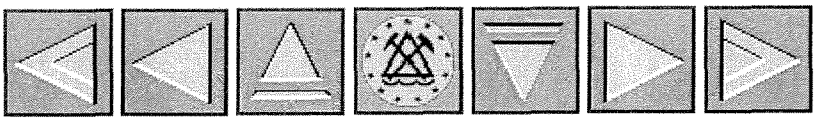


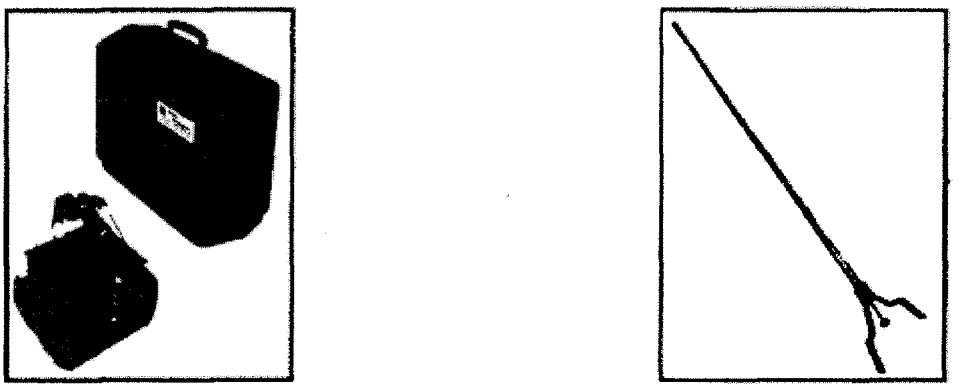
D. Hand-screen collector



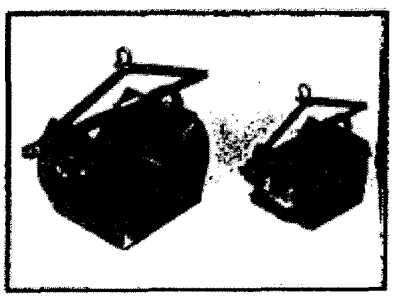
E. Stovepipe core sampler

Figure 3.--Examples of sampling equipment used to collect benthic invertebrates (A-C, photographs courtesy of Wildlife Supply Company, Saginaw, Mich.; D-E, modified from Merritt and Cummins, 1984).

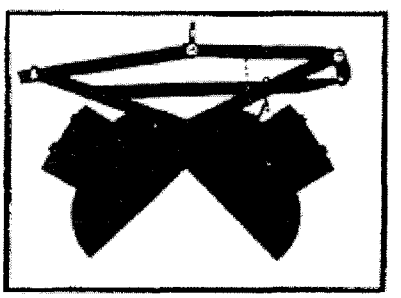




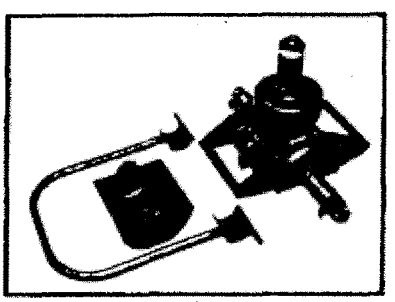
F. Ekman grab and carrying case on the left, and handle on the right.



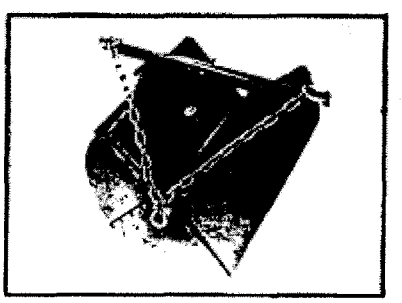
G. Ponar grab



H. Petersen grab



I. Shipek grab



J. Van Veen grab

Figure 3.--Examples of sampling equipment used to collect benthic invertebrates (F-J, photographs courtesy of Wildlife Supply Company, Saginaw, Mich.)--Continued.

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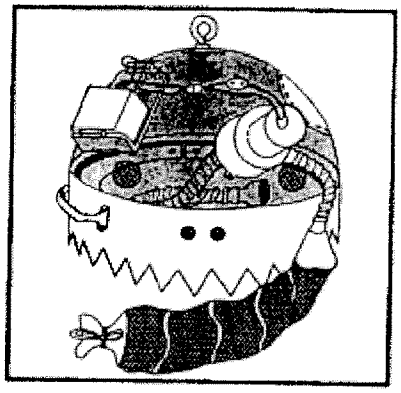
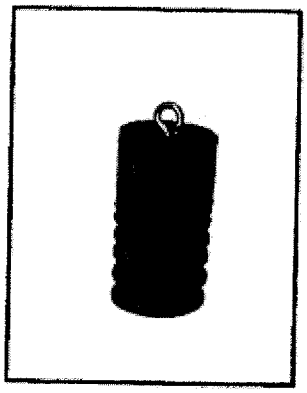
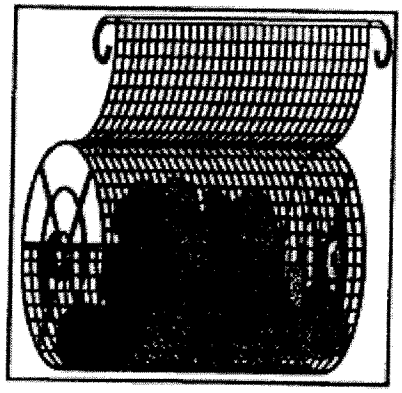


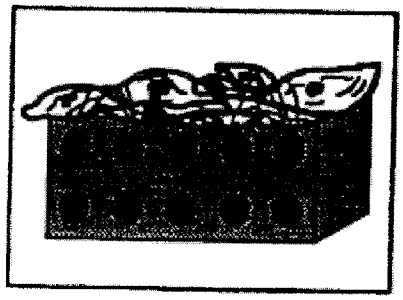
K. Dome sampler



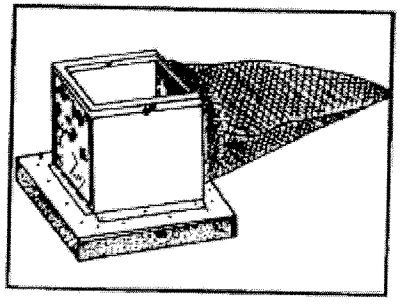
L. Multiple artificial substrate sampler



M. Barbeque basket artificial substrate sampler

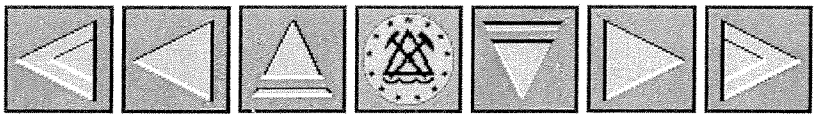


N. Leaf pack artificial substrate sampler



O. Box sampler

Figure 3.--Examples of sampling equipment used to collect benthic invertebrates (K, modified from Gale and Thompson, 1975; L, photograph courtesy of Wildlife Supply Company, Saginaw, Mich.; M-O, modified from Merritt and Cummins, 1984)--Continued.



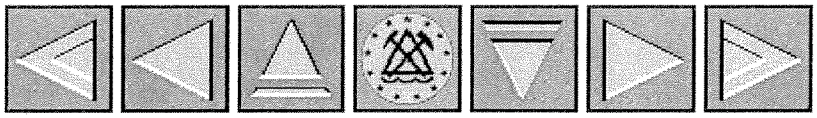
The D-frame kick net collections are supplemented with visual collections and, where appropriate, with seines to collect highly-motile invertebrates. Visual collections involve manually collecting large rocks, coarse organic debris, clay from stream marg ns, root wads, and macrophytes or other substrates, and visually locating and removing any associated organisms. This method is particularly useful for collecting firmly attached organisms, such as sponges, and organisms that burrow into hard substrates (*Tortopus*), plant tissues (*Donacia*), or sponges (*Climacia*).

Seining along point bars and islands in mid- to high-order streams is an effective means of collecting larger, highly-motile invertebrates such as many of the amphipods, siphlonurid mayflies, and freshwater prawns. A common minnow seine with 3.2-mm (1/8-in.) mesh can be used for this purpose. Specimens collected using seines and visual methods are placed in an appropriate container and labeled. (See section on Sample Processing and Labeling for details.) Other sampling techniques and equipment, such as Hess samplers (fig. 3B), Surber samplers (fig. 3C), hand screens (fig. 3D), and core samplers (fig. 3E), are used in wadeable habitats as appropriate.

The choice of collection method for QMH samples from nonwadeable habitats depends upon the depth of the water, current velocity, and bed material. For example, grab samplers (grabs), such as Ekman (fig. 3F), Ponar (fig. 3G), or Petersen (fig. 3H) grabs, are suitable for sand or fine gravel substrates in moderate-current conditions and waters of medium depths. Shipek (fig. 3I) and Van Veen (fig. 3J) samplers are useful in extremely deep and fast rivers with sand or fine gravel bottoms (Wells and Demas, 1 979). However, grabs and Shipek samplers do not work well in deep rivers where the bed material is composed of large gravel, cobble, boulder, or bedrock. A diver-operated Dome sampler (fig. 3K) or artificial substrates (fig. 3L, -M, and -N) are required in these situations. Where appropriate, baited traps may also be used to obtain difficult-to-catch organisms, such as crayfish, in large rivers with coarse substrates.

QMH sample collections in large, nonwadeable rivers are distributed among the different instream habitat types and along at least two channel transects. Transect sampling is usually done with appropriate grab samplers or artificial substrates. Samples a recollected at the ends of the transects (near the shoreline), in the middle of the channel, and at points midway between the in-shore and mid-channel samples (total of five samples). Transect sampling is usually combined with the habitat assessment of large, nonwadeable rivers.

Qualitative sampling employs a variety of samplers and techniques, including visual collections of leaves, wood, and rocks. When the collection technique involves using a net, the mesh size of the net must be 210 µm. Samples are elutriated, sieved, and split in the field to reduce the bulk of the composite sample to less than 0.75 L. This processing is done using 212-µm sieves [USA Standard Testing Sieve, American Society for Testing of Materials (ASTM) number 70 or Tyler equivalent 65 mesh]. The composited qualitative sample is placed in an appropriate sample container, preserved in 10-percent formalin, and properly labeled.



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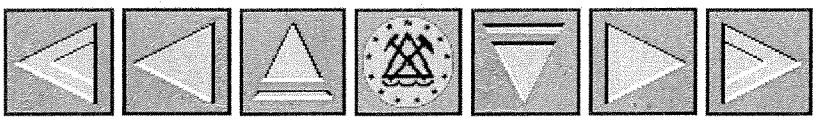
Semi-Quantitative Targeted-Habitat Sampling Methods

The objective of semi-quantitative targeted-habitat sampling is to obtain representative samples of benthic invertebrate communities from two instream habitat types: (1) a habitat supporting the faunistically richest community of benthic invertebrates (r ichest-targeted habitat, RTH), usually a fast-flowing, coarse-grained riffle; and (2) a fine-grained, organically rich depositional habitat (depositional-targeted habitat, DTH), usually a pool. Semi-quantitative sampling methods characterize the structure of invertebrate communities in terms of the relative abundances of each taxon rather than absolute density. Information on community structure is useful in constructing a variety of biological indexes, such as diversity, community similarity, and funct ional and trophic groupings. These groupings are used to (1) interpret how the community is functioning (For example, what is the source of its energy?); (2) compare sites (For example, do sites with the same physical and chemical characteristics have the same community structure?); and (3) relate community structure to physical, chemical, and land-use characteristics affecting water quality (For example, do community characteristics correlate with land-use and chemical characteristics?).

The type of sampler used to collect a semi-quantitative sample depends upon the depth, velocity, and substrate within the instream habitat type sampled. The choice of sampler is made with the advice of the regional biologist, North Carolina Ecology Group, and local biologists. Artificial substrates, such as multiplate samplers (fig. 3L), substrate basket samplers (fig. 3M), artificial leaf packs (fig. 3N), and artificial snags are used in situations where natural substrates cannot be sampled because of inaccessibility of the habitat, cost of sample collection, or safety issues associated with collecting the samples (for example, the use of divers in large, fast-flowing rivers). However, artificial substrates have a number of limitations that should be factored into their use: (1) they require two trips to the sampling site (installation and removal) separated by an interval of time to allow for colonization of the substrates, which depends on season, discharge, temperature, and other environmental variables; (2) they are susceptible to loss and vandalism; (3) they are biased toward species that are actively colonizing at the time of placement; (4) they often do not accurately depict the types or relative abundances of the benthic invertebrates at a site; and (5) they may not be sensitive to changes in water quality associated with changes in land use.

Under certain circumstances, such as in large, deep rivers with cobble, boulder, or bedrock substrate, artificial substrates may offer the only viable means of obtaining community samples. The preference in these circumstances is to use artificial substrates that mimic natural substrates such as "barbecue baskets" (fig. 3M) filled with indigenous rocks (Britton and Greeson, 1988) or artificial snags floating at the stream margins. All artificial substrates are allowed to colonize for a minimum of 6 weeks unless locally available data suggest that a longer or shorter colonization period is more appropriate.

Generally, habitats associated with deep, sandy-bottomed, fast-flowing rivers do not yield sufficient numbers of taxa to warrant the effort required to obtain representative samples. In these situations, habitats associated with the margins of the main channel and islands, such as snags and macrophyte beds, generally support accessible and faunistically rich assemblages of invertebrates (Benke and others, 1984; Thorp, 1992). The use of these



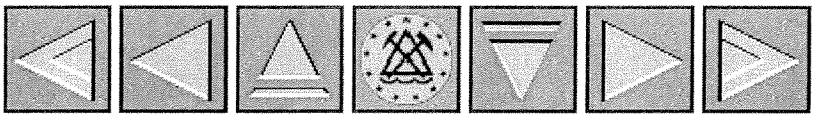
habitats for semi-quantitative sampling should be investigated prior to committing to the use of artificial substrates for semi-quantitative sampling of the richest-targeted habitat.

All nets and screens used in the collection of semi-quantitative samples must have a mesh size of 425 µm (USA Standard Testing Sieve, ASTM number 40 or Tyler equivalent 35 mesh). Samples are elutriated, sieved, and split in the field to reduce the bulk o f the composite sample to less than 0.75 L. Samples collected and processed in this manner are placed in appropriate sample containers, preserved in 10-percent formalin, and properly labeled.

Recommendations for Sampling Benthic Invertebrates

Specific, nationally consistent recommendations on which habitat type to select for semi-quantitative sampling in each study unit are difficult to devise, particularly for selection of the RTH. The instream habitat types presented in figure 2 represent a generalized classification scheme, broadly based on habitat features of relevance to benthic invertebrates, that can be used for local, regional, and national aggregation of data. However, this classification scheme cannot account for local factors, such as substrate condition, depth, current velocity, hydrologic management, and accessibility, that may make one habitat type more suitable under one set of site conditions than another. For example, in large rivers with unstable erosional zones that lack snags or macrophyte beds, pool habitats may represent the faunistically richest habitat as well as the habitat where exposure to particle-borne contaminants is greatest. Under these circumstances, pool habitats are sampled as the RTH and a contrasting ha bitat type is chosen as the DTH. The selection of the appropriate instream habitat type for RTH and DTH sampling is based on national guidance supplemented with information derived during the retrospective data analysis, input from the study-unit liaison committee, consultation with the regional biologists and North Carolina Ecology Group, and reconnaissance sampling.

National guidance on choosing appropriate instream habitat types (fig. 4) is provided by prioritizing the elements within each level of the hierarchy used to define the matrix of instream habitat types. Characteristics of the hierarchy are ranked from hi ghest priority (1) to lowest priority (6). Based on these rankings, the highest priority for RTH sampling is a riffle, main-channel, natural-bed instream habitat type, whereas the lowest priority is given to a pool, island-margin, slough instream habitat type. Similarly, the highest priority for DTH sampling is a pool, main-channel, natural-bed instream habitat type, and the lowest priority is given to a riffle, island-margin, manufactured-bed instream habitat type. These priorities apply only within a level of the hierarchy, such as major channel feature, and not across levels, such as comparing priority levels for channel margins with woody snags. The study-unit biologist determines which of the instream habitat types present in the sampling reach b est meets the objectives of semi-quantitative sampling with regard to local conditions and available resources. The guidance provided in figure 4 represents a starting point for determining the appropriate habitat type to sample within a study unit.



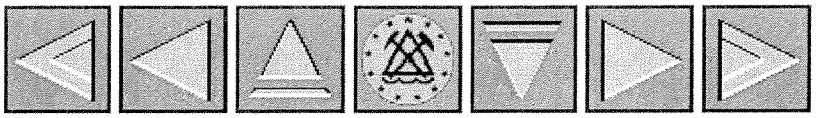
Richest-Targeted Habitat (RTH) Type

Major geomorphic channel unit		Major channel boundary		Major channel feature	
Priority level	**Descriptor**	**Priority level**	**Descriptor**	**Priority level**	**Descriptor**
1	Riffle	1	Main channel	1	Natural bed
2	Run	2	Channel margin	2	Woody snag
3	Pool	3	Island margin	2	Macrophyte bed
				3	Bar
				4	Manufactured bed
				5	Slough

Depositional-Targeted Habitat (DTH) Type

Major geomorphic channel unit		Major channel boundary		Major channel feature	
Priority level	**Descriptor**	**Priority level**	**Descriptor**	**Priority level**	**Descriptor**
1	Pool	1	Main channel	1	Natural bed
2	Run	2	Channel margin	2	Slough
3	Riffle	3	Island margin	3	Bar
				4	Macrophyte bed
				5	Woody snag
				6	Manufactured bed

Figure 4.--Recommended priority levels for determining the appropriate habitat type to select for semi-quantitative sampling.



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Once the appropriate habitat type has been identified for RTH and DTH sampling, then a suitable sampler is selected, based on the depth, velocity, and substrate conditions within the habitat type. The following recommendations on semi-quantitative sampling techniques and gear are organized on the basis of wadeability of sites and dominant substrate condition (coarse or fine-grained). Wadeable sites are generally considered to be areas where water depth is less than 1 m, whereas depths at nonwadeable sites are typically greater than 1 m. Coarse-grained substrates are defined as those that are typically dominated by medium to large gravel, cobbles, or boulders, or by bedrock. Fine-grained substrates are defined as those that are dominated by small grave l, sand, silt, or clay. As with the discussion of habitat types, the recommendations on quantitative sampling gear and procedures are matched to local conditions and modified as needed to better characterize the benthic invertebrate community. The proce dures and equipment recommended here are equally applicable to QMH sampling.

Wadeable Coarse-Grained Substrates

Disturbance-removal sampling techniques are the most appropriate method for sampling wadeable coarse-grained substrates with current velocities greater than 5 cm/s. These techniques involve defining a specific area, disturbing the substrate within that a rea to dislodge invertebrates into a sampler or downstream net, and then removing the larger substrate elements to acquire any specimens that are adhering tightly to the rocks. Hess samplers (fig. 3B), Surber samplers (fig. 3C), stovepipe corers (fig. 3E), and box samplers (fig. 3O) are examples of the types of samplers that can be used.

The Slack sampler, a modification of the Surber sampler, proved very useful for sampling riffles and runs during the NAWQA Program pilot studies. This sampler, also referred to as a "Surber-on-a-stick," was developed by Keith Slack, USGS, Menlo Park, Calif. It requires a minimum of two people to operate and employs a 0.5-m wide rectangular kick-net frame to which a Nitex net with 425-µm mesh openings is attached (fig. 5). The sampler is held perpendicular to the direction of flow and pressed tightly ag ainst the stream bottom. It may be necessary to move cobbles aside or to add a self-sticking foam strip to the bottom of the sampler (particularly when working on rock outcrops) in order to achieve a tight seal. Benthic invertebrates are collected from an area of approximately 0.25 m*2* immediately upstream of the Slack sampler. The sampling area is delineated using either a guide rod or frame that attaches to the sampler. A combination guide rod and digging tool can be fashioned from 0.61 m (2 ft) of 9.5-mm (3/8-in.) diameter threaded rod (Karen Murray, U.S. Geological Survey, oral commun., 1992). Two nuts are threaded onto the rod to divide it into 0.5-m and 0.1-m lengths. One side of the sampling area is delineated by laying the rod perpendicular to the Slack sampler with the longer length facing upstream and the nuts in contact with the side of the sampler. The other dimensions of the sampling area are visually approximated. Alternatively, the sampling area can be delineated using a guide frame fashioned by bending a 6.35-mm (1/4-in.) wide by 3.18-mm (1/8-in.) thick flat aluminum strip to form three 0.5-m long arms joined at right angles. This frame is then attached to the front of the Slack sampler and used to define the sampling area.

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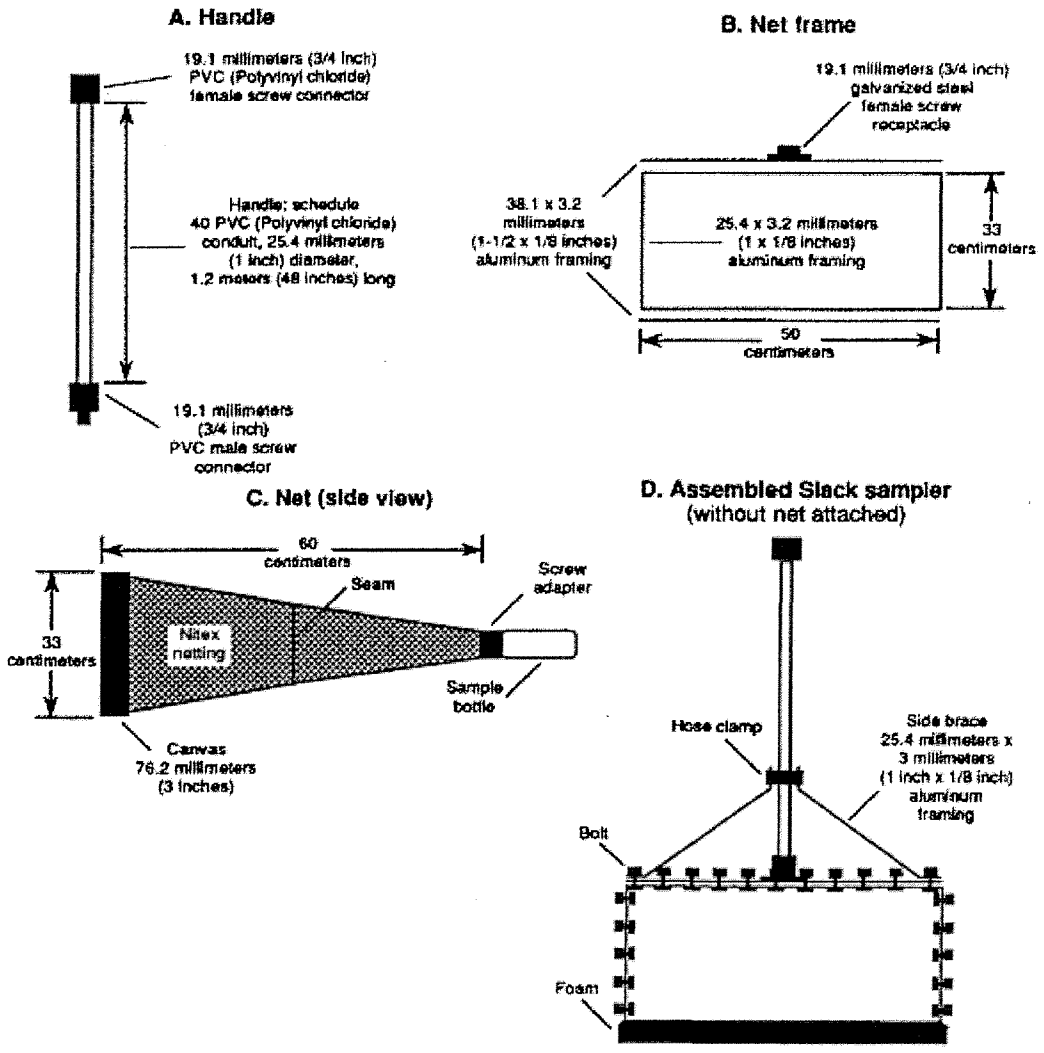
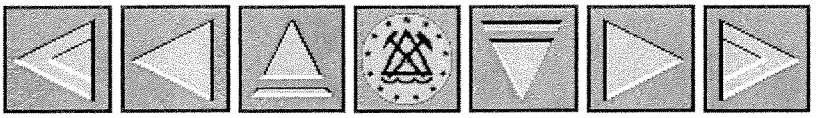


Figure 5.--Schematic diagram of the Slack sampler used to collect benthic invertebrates from wadeable, coarse-grained substrates.



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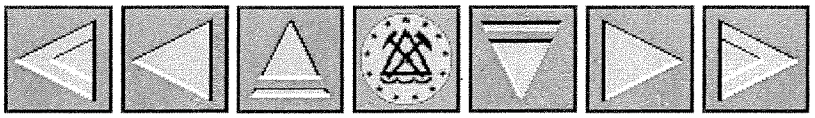
If 50 percent or more of a rock lies within the sampling area, it is removed and held in front of the net opening, and attached organisms are dislodged into the net by gently brushing the surface of the rock with the hand and then with a fingernail brush. After a rock is brushed, it is examined to determine if any closely adhering organisms, such as *Leucotrichia* (microcaddisfly) or *Parargyractis* (aquatic lepidopteran), are present. Such organisms are removed from the rock surfaces using fo rceps and placed into a separate vial holding the large-rare sample component. This sample component contains large organisms that can interfere with sample splitting and rare organisms that might be lost during sample splitting. After the large rocks (fist size and larger) are removed, the sampling area is dug to a depth of about 0.1 m. The guide rod makes an effective tool for digging to this depth. The short end of the guide rod is set to the depth criterion (0.1 m), and the long end provides subst antial leverage for digging into consolidated materials. When it is not possible to achieve the 0.1-m depth, digging is done as deeply as is practical. Any remaining organisms are dislodged into the net by kicking the substrate within the sample area fo r a period of 30 seconds. The material collected in the net is then rinsed into the bottle attached to the sampler and transferred to an appropriate container, usually a 19-L (5-gal) plastic bucket or dishpan, for further field processing. Subsequent elements of the composite sample are added to this container and then processed, or the separate elements may be processed and then composited.

Nonwadeable Coarse-Grained Substrates

Coarse substrates in water deeper than approximately 0.50-0.75 m cannot be effectively sampled using most disturbance-removal type samplers. A diver-operated dome sampler (fig. 3K) can be used in such situations. This sampler contains a battery-operated pump that empties material into a Nitex bag with 425-µm mesh openings. The material in the dome sampler is dislodged by the diver, sucked up by the pump, and deposited in the mesh bag. As with the Slack sampler, the diver brushes invertebrates from the surfaces of the rocks first using his hands and then a fingernail brush. These rocks are taken out of the sampler and returned to the surface to remove tightly attached invertebrates. The substrate remaining in the sampler is then disturbed by hand for 30 seconds to a depth of about 0.1 m. Substrate samples are collected and returned to the surface for substrate characterization and inspection for remaining invertebrates. After the pump has cleared the dome sampler of suspended debris and invertebrates, the sampler is returned to the surface where the Nitex bag is removed and the contents washed into a suitable sample container and held for field processing.

Artificial substrates, such as rock-filled barbecue baskets (fig. 3M), may also be used in sampling coarse substrates in nonwadeable areas (Britton and Greeson, 1988). The barbecue baskets are filled uniformly with indigenous rocks from the river or with rocks that are geologically similar to the local river rocks. Filled baskets are placed on the bottom of the river within the appropriate instream habitat type and tied to floats or stream-side vegetation. The baskets are allowed to colonize for a mini mum of 6 weeks and then are removed with the aid of a 425-µm net to catch organisms dislodged during transfer of the



baskets to the surface. After retrieval, the contents of the basket and the collection net are emptied into a bucket, and the associated organisms are removed by scrubbing the substrate with a fingernail brush. This material is then concentrated on a 425-µm mesh sieve and composited with other basket samplers prior to field processing. Baited traps or baited artificial substrate samplers are appropriate for QMH sampling in deep, rocky rivers but not for RTH or DTH sampling. Baited traps can be particularly effective for collecting crayfish under conditions where other methods are ineffective.

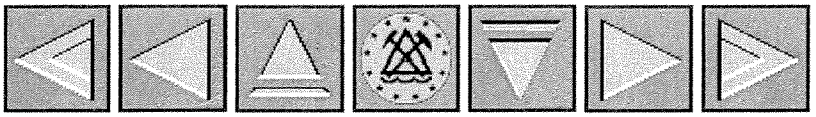
Stovepipe samplers (fig. 3E) may also work in this type of habitat in water less than 0.75 m deep. For this technique, approximately 1 m of large-diameter 30-cm polyvinyl chloride (PVC) pipe with a beveled bottom edge is used. This sampler is driven int o the substrate deeply enough to produce a good seal around the bottom of the sampler. Substrate is removed from the sampler by hand, if physically possible, and composited. More typically, the substrate is too deep to reach by hand, so organisms are dislodged from the substrate using long-handled brushes and scrapers attached to poles. A hand- or battery-operated diaphragm pump equipped with suitable hoses and a PVC pipe is used to pump the water and suspended invertebrates into a 425-µm mesh net. In dividual samples are composited to obtain a representation of the habitat type. The disadvantages of this type of sampler are that it is (1) often difficult to handle the sampler in very fast-flowing water, (2) usually impossible to assess the degree to which the substrate has been cleaned of invertebrates, and (3) often difficult to estimate substrate characteristics. However, despite these shortcomings, this type of sampler and its modifications provide one of the few means to collect samples on rock outcrops in water 50-75 cm deep (Voshell and others, 1992).

Wadeable Fine-Grained Substrates

Grab samplers (fig. 3F, -G, -H, and -J) are appropriate for use in shallow, fine-grained riffles or pools. A pole-mounted Ekman grab is particularly useful in wadeable streams with sand or silt substrates, whereas a Ponar grab is a better choice for fine-gravel substrates. All screening on the grab should have mesh openings of 425 µm or smaller. The appropriate screening mesh can be accomplished by gluing or sewing smaller mesh fabric over the existing larger mesh panels of grabs such as the Ponar. Grab samplers are lowered carefully to the stream bottom and released to avoid disturbing the sediments prior to contact and to aid in establishing a uniformity of substrate penetration. Additional weights can be added to grabs to achieve better and more uniform substrate penetration. Recovered grabs are carefully checked to make sure that sample material was not lost because of rocks, sticks, or other debris catching in the jaws of the grab. Each of the samples to be composited is taken within the sam e instream habitat type but at sufficient distances apart to avoid interference among samples. Individual samples can be composited in a suitable container prior to field processing or processed and then composited.

Nonwadeable Fine-Grained Substrates

Grab samplers, such as Ponar (fig. 3G), Petersen (fig. 3H), Van Veen (fig. 3J), or Shipek (fig. 3I) grabs, can be used from boats to obtain samples from deep rivers with fine-grained substrates. A hand or power winch is recommended for sampling in deep waters or using weighted grab samplers. All screening on the grab sampler should have mesh openings of



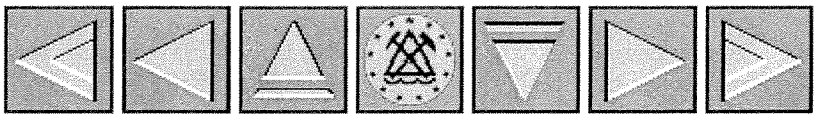
425 µm or smaller. Grab samplers are lowered to within 3 m of the streambed, stopped, and then allowed to drop to the streambed. This process helps minimize disturbance of the substrate by the descending sampler and achieves a more uniform depth of substrate penetration. Recovered grab samplers are carefully checked to make sure that sample material was not lost because of rocks, sticks, or other debr is catching in the jaws of the sampler. Individual grab samples are composited in a suitable sample container, such as a 19-L (5-gal) plastic bucket or a large dishpan. Each of the samples to be composited is taken within the same instream habitat type but at sufficient distances apart to avoid interference among samples. Simple sonar units, such as those used by sport fishermen to locate schooling fish, are used to coarsely approximate stream bottom conditions and depth, to help define the limits of the instream habitat type, and to determine the depth at which the sampler is released.

In large, sandy-bottomed rivers, the faunistically richest instream habitat type may be woody snags or macrophyte beds associated with the margins of riffles or runs, and the appropriateness of these habitats should be evaluated prior to committing resources to quantitatively sampling deep, sandy substrates. The selection of the appropriate instream habitat type for RTH and DTH sampling is based on national guidance supplemented with information derived from the retrospective data analysis and reconnaiss ance sampling or obtained by consulting the study-unit liaison committee, the regional biologists, and the North Carolina Ecology Group.

Woody Snags and Macrophytes

Special attention is given to sampling woody snags and macrophyte beds in large, sandy-bottomed rivers or streams with otherwise unstable substrates. Under these conditions snags and macrophyte beds can support the faunistically richest communities of or ganisms within the sampling reach. Snags are sampled by cutting off sections of tree limbs with a saw or lopping shears, removing the limb from the water, and collecting the attached invertebrates by hand-picking and brushing the limb surface and cavities. Losses of mobile or loosely attached organisms can be minimized by placing a net (for example, a D-frame dip net or the Slack sampler) downstream from the limb to catch dislodged organisms or by placing the limb in a specialized snag sampler (Thorp an d others, 1992). The Thorp and others (1992) snag sampler clamps the limb between an upstream and downstream net that minimizes the loss of mobile organisms while the sample is cut away from the main body of the snag. If the limb is too large to cut, it is sampled in place by brushing its surface with a fingernail brush and catching the dislodged invertebrates in a net placed immediately downstream from the snag. The lengths and diameters of the limbs sampled should be entered on the field data sheet.

Macrophyte beds can be sampled with disturbance-removal samplers (Slack sampler, grab, or stovepipe samplers). Net samplers, such as the Slack sampler (fig. 5), can be used if there is sufficient current to wash the dislodged plant and animal material in to the net. A knife or trowel is used to dislodge the plant material from the substrate. Grabs can be used to sample low-growing submergent macrophytes on sandy or silty substrates in areas with low current velocities. However, stovepipe samplers may prove more effective and should



be used when the macrophytes are too tall to allow use of a dredge. The macrophytes that are removed should be inspected carefully for attached invertebrates and for invertebrates that burrow into stems.

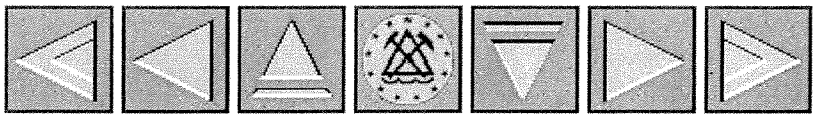
Field Sampling Activities

Benthic invertebrate sampling, algal sampling, habitat assessment, and measurements of water-chemistry and physical properties, such as discharge, nutrients, dissolved-oxygen concentration, conductivity, and pH, are activities that are typically carried out on visits to a basic fixed site. Consequently, there must be close coordination among sampling teams to minimize interference while keeping the time spent at a site to a minimum. Once on site, the habitat crew establishes the beginning and ending points of the sampling reach(es) and conducts riparian and flood-plain characterizations while the algal and invertebrate teams cooperatively collect samples. Alternatively, the habitat crew can establish the sampling reach(es) and conduct all or part of the riparian and flood-plain characterization procedures on one or more prior visits to the sampling site. This approach greatly facilitates the efficiency of the ecological survey sampling. Upstream disturbance is minimized during sample collection by be ginning with the most downstream sampling location and progressing upstream. When collections are made at sites with multiple sampling reaches, all sampling activities are completed within a sampling reach before proceeding to the next reach.

The combined invertebrate and algal sampling teams start by identifying the locations of appropriate instream habitat types for the semi-quantitative (RTH and DTH) and qualitative (QMH) sampling of invertebrates and algae. When the appropriate habitat type for RTH sampling can be determined in advance (through literature review or input from local biologists), then RTH samples are collected first followed by DTH and QMH samples. If the appropriate habitat type for RTH sampling cannot be determined in advance (for example, in many large rivers), then the QMH samples are collected first and the appropriate RTH and DTH instream habitat types are identified based on the QMH sampling. If fish are to be collected at the same site, fish sampling is done after all invertebrate samples are collected to avoid disturbing the site.

Care must be exercised to ensure that sampling sites are typical of the chosen instream habitat type within the sample reach--that is, reasonably similar with respect to substrate type, current velocity, depth, and debris accumulation. Sites below upstream obstructions and along the edges of adjacent instream habitats are avoided. A minimum of five samples, apportioned within and among examples of the targeted instream habitat type, are composited into a single RTH or DTH sample. Examples of the target ed habitat type are collected from across the length and width of the sampling reach.

A hypothetical example of semi-quantitative samples collected from a sampling reach consisting of a sequence of alternating pools and riffles is illustrated in figure 6. In this example, six samples are collected with a Hess sampler from each of the two riffles (total area sampled is 0.51 m*2*), and six samples are collected from the two pools using a petite Ponar sampler (total area sampled is 0.14 m*2*). The samples from each habitat are



composited to yield two samples, one for the riffle, channel, natural-bed instream habitat type and one for the pool, channel, natural-bed habitat type. The appropriate instream habitat characterization data for each sample of the composite are recorded on the appropriate field data sheet.

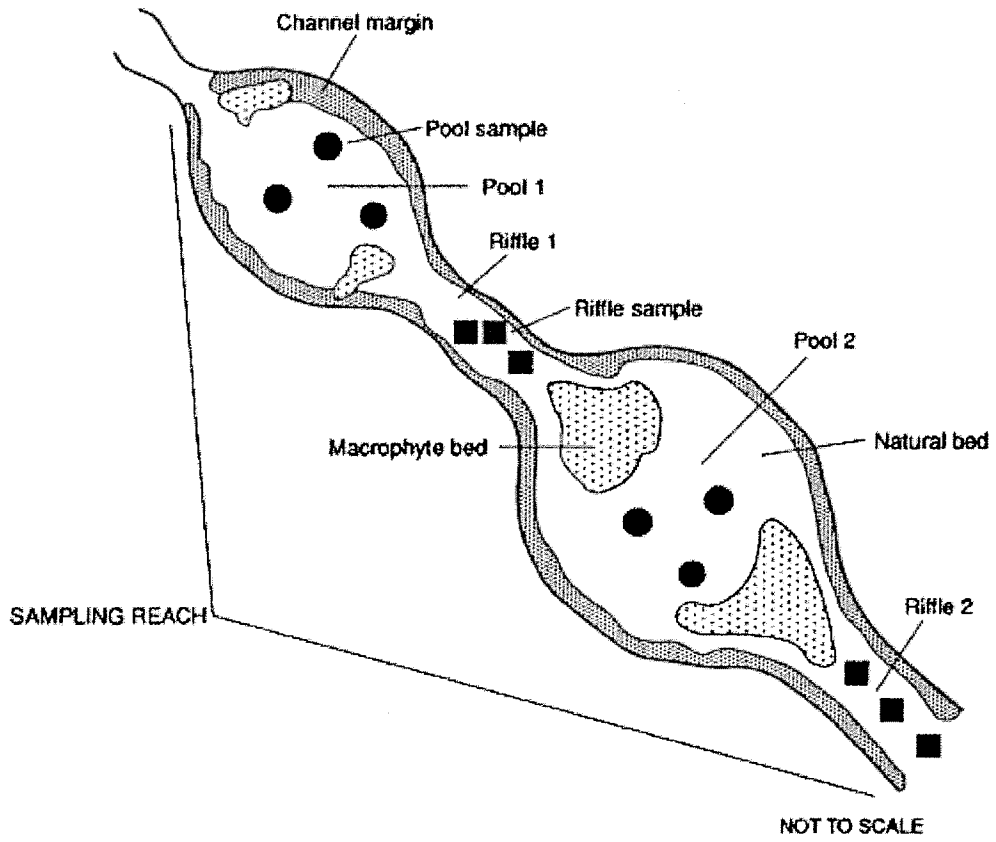
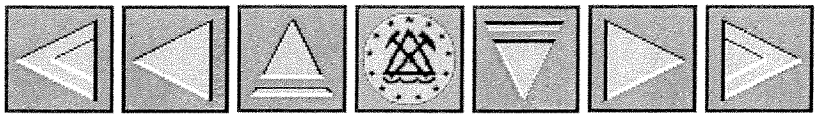


Figure 6.--Diagram illustrating how semi-quantitative samples could be distributed within a sampling reach.



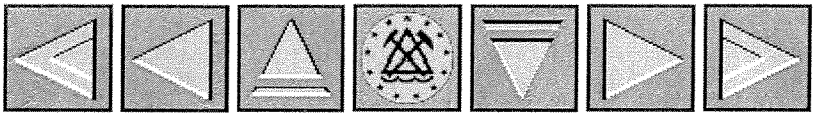
MAINTENANCE OF SAMPLING EQUIPMENT

An important component of field quality assurance and quality control is the maintenance of field equipment. All nets and sieves must be inspected for damage at least daily and repaired as soon as damage is discovered. Nitex netting can be repaired easi ly using thermoelectric glue guns that are available in corded and cordless models. The locations of holes and worn spots are determined by visual inspection and marked. Hot glue is then applied to the damaged spot from both sides of the net and allowed to cool. This procedure produces a quick, permanent, and durable repair. Small torn places in net seams are repaired in a similar manner, although it is usually advisable to re-sew the seam before sealing it. Similarly, brass and stainless steel sieve s also can be temporarily repaired in this fashion, though permanent repairs are made by having the sieves professionally soldered. The canvas scuff guards present on many samplers, such as D-frame dip nets, Surber samplers, and Hess samplers, are inspected for damage daily and repaired by oversewing with similar canvas materials. Nets are replaced when damage is severe or repairs are impossible. Replacement nets should be part of standard field equipment.

Grab samplers are cleaned after every use to prevent cross contamination of samples and to ensure reliable operation. In most cases, lubrication of grab samplers is not necessary if they are kept clean and are stored properly. Lubrication should be avoided if the sampler is ever intended to be used for collecting chemical samples because of the risk of contamination. Screening on the samplers is inspected and repaired on a daily basis, and samplers are transported in sturdy boxes to prevent damage. All cables are inspected daily to ensure that they are in good condition and are replaced as necessary. Winches, boats, motors, and safety gear such as personal floatation devices (PFD's), fire extinguishers, and first-aid kits are periodically inspected, maintained, and repaired as necessary.

SAMPLE PROCESSING AND LABELING

The combining of multiple benthic samples into a single composite sample may result in an unacceptably large volume of material (more than 0.75 L). Consequently, samples are field processed to reduce the volume of each sample component so that it fits in to a 1-L sample container with ample room for preservative. Sample volume reductions are accomplished by removing large debris, elutriating to remove inorganic sediments, and then splitting the elutriated samples (fig. 7). Field processing is applied ei ther to individual samples as they are collected or to the entire composite sample. The latter approach is feasible if each sample produces only a small volume of material. However, if each sample produces a relatively large volume of material, it will be faster to process each sample individually and composite the sample components. The study-unit biologist determines which approach is more appropriate for conditions at the site. Field processing can result in the production of four sample components from each composite sample: large-rare, main- body, elutriate, and split-sample components.



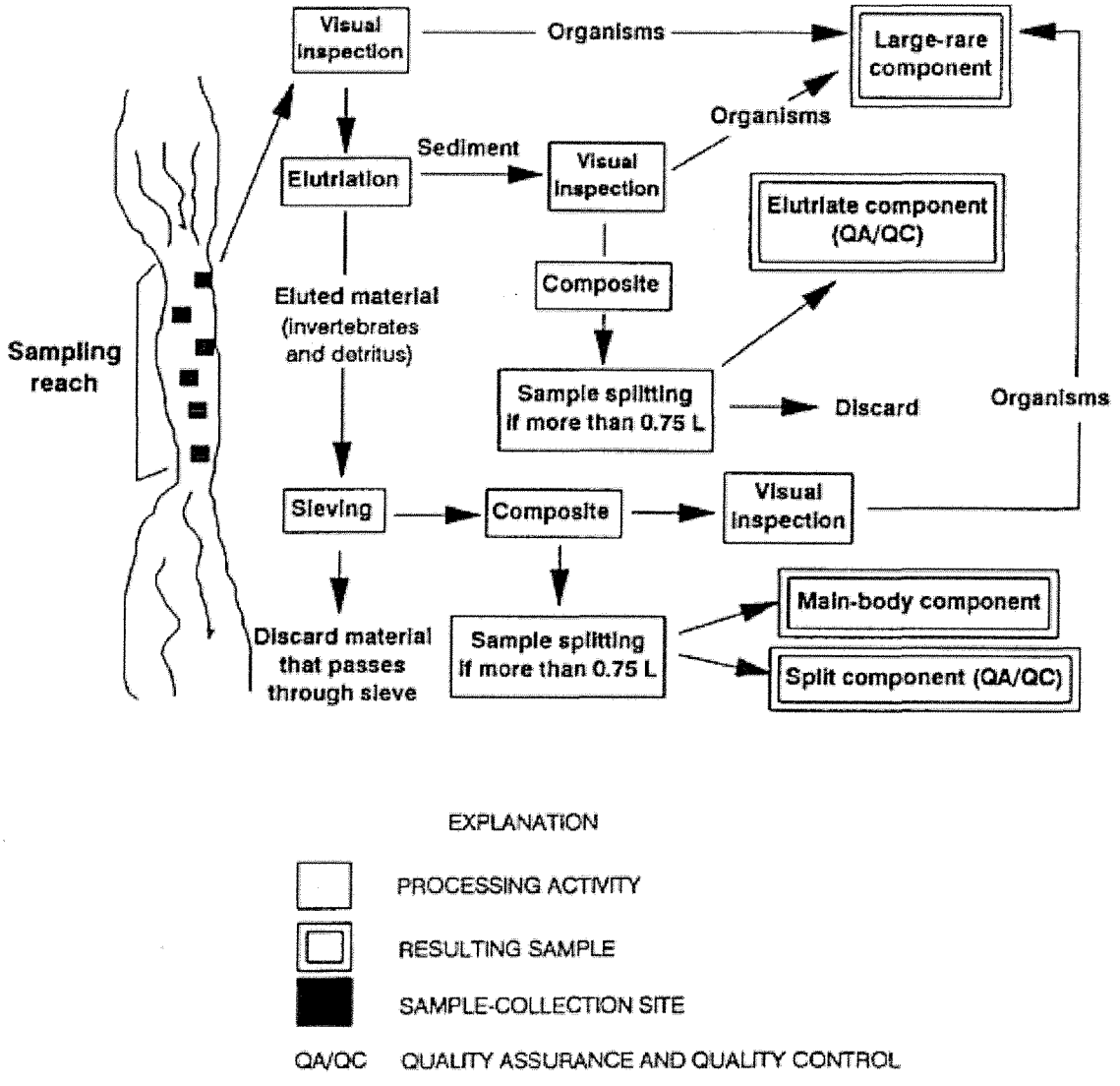
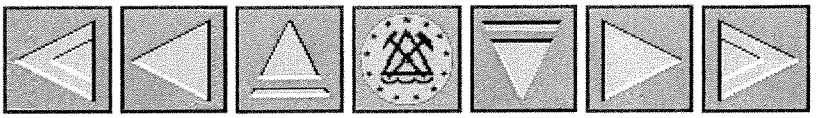


Figure 7.--Flow chart indicating how benthic invertebrate samples are processed in the field.



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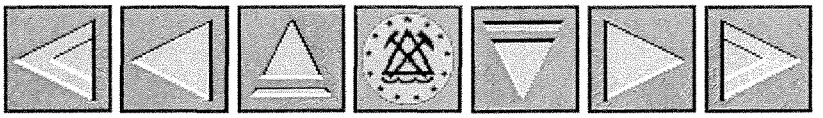
Field processing begins with the removal of large rocks and organic debris, such as leaves, twigs, and roots, from the sample. These materials are discarded after checking to ensure that all attached invertebrates have been removed. The remaining material is quickly examined for large, rare organisms that could be lost during subsequent sample splitting. These large-rare organisms are removed and placed in a separate, labeled container that is identified as the "large-rare" sample component. All organ isms that are picked from the sample by hand prior to sample splitting are added to the large-rare sample component.

The remaining sample material is elutriated onto an appropriately sized sieve (425-µm mesh for semi-quantitative samples and 212-µm mesh for qualitative samples) to separate the lighter organic material from the heavier sand and gravel. Elutriation is usually accomplished by placing the sample in a deep bucket (a 19-L plastic bucket works well for this purpose) filled about one-fourth to one-half with water. The contents of the bucket are stirred by hand to suspend as much material as possible. The buc ket is picked up, swirled, and then gently decanted onto an appropriate sieve while the advancing sediment front is carefully watched. Sieving effectiveness is increased and clogging is decreased if the sieve is kept in constant motion while the sample is decanted. An effective motion is created by holding the sieve in one hand with the thumb on the top lip of the sieve and the other fingers supporting the bottom of the sieve. The wrist is used to impart a back and forth motion perpendicular to the flow of water onto the sieve. Decanting stops when the sediment front reaches the lip of the bucket. A backup container, such as a wash tub or another 19-L bucket, is placed under the sieve to catch any material that is spilled over the edge of the sieve d uring the elutriation. If material is spilled into the backup container, the contents of the backup container are returned to the sample bucket and the elutriation is repeated. If no material is spilled, then the contents of the backup container are dis carded after each decantation. The elutriation process is repeated until it appears that only sand and gravel remain in the elutriation bucket.

The sand, gravel, and small pebbles remaining in the bucket are visually examined for invertebrates, particularly case-building caddisflies and small mollusks such as *Corbicula*. Visual inspections are usually easier and faster if only small amounts of the sample are examined at a time and(or) the material is sieved through nested large-meshed sieves (for example, 4-, 2-, and 1-mm mesh sizes). A shallow white pan or tray containing approximately 1 cm depth of water is ideal for visual inspections . Invertebrates that are removed during this process are added to the large-rare sample component. Sieving this material often speeds up the examination process. Once free of invertebrates, the left-over sand and gravel is retained as a quality-assuran ce check on the efficiency of elutriation. This "elutriate" sample component is split, if necessary, preserved in 10-percent buffered formalin, labeled, and shipped to the USGS Quality Management Group's Biological Quality-Assurance Unit (BQAU) located at the National Water-Quality Laboratory (NWQL) in Arvada, Colo., to determine the effectiveness of elutriation. The BQAU is responsible for handling quality assurance and quality control (QA/QC) for laboratory analysis of the samples.



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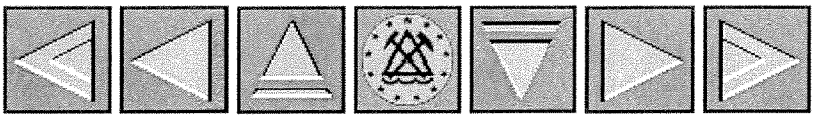
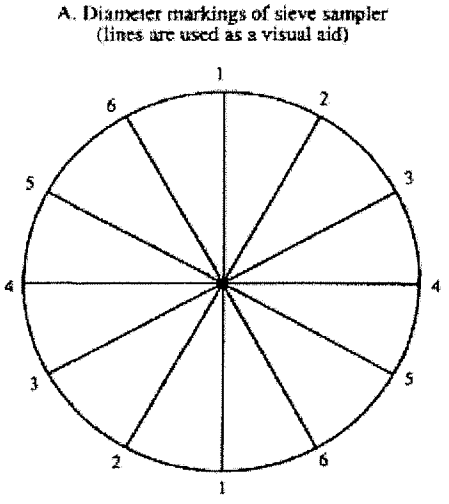
Elutriated material retained on the sieve is quickly examined for large, rare organisms that are added to the large-rare sample component. This procedure helps to ensure that these organisms are not lost during subsequent sample processing. The material remaining on the sieve is then washed by repeatedly dipping the sieve into a dishpan or bucket half filled with water and gently swirling the sieve to wash material through it. Large commercially available sieves, such as the wash bucket and Ponar wash frame (Wildlife Supply Company Catalog, 1980), can be used effectively to screen large-volume samples. Alternatively, large-volume samples can be divided into smaller portions and washed using standard-sized sieves. In the latter case, the respective sample components (large-rare, elutriate, and main-body) resulting from each washing are combined to form the components of the composite sample.

If, after elutriation and compositing, the volume of material constituting the main-body or elutriate sample component exceeds 0.75 L, that sample component is split in the field. Any debris or large organisms that remain in the sample must be removed lest they interfere with the sample-splitting process. Organisms so removed are added to the large-rare sample component, whereas debris is discarded after any attached invertebrates are removed.

Sample splitting is accomplished by using either a special sieve sample splitter (Mason, 1991) or a sieve diameter splitting method. The sieve sample splitter consists of a Plexiglas box with a mesh bottom formed by two equal compartments. The two compartments are latched tightly together, and the sample is placed on the sieve, immersed in water, and gently agitated to distribute the sample uniformly over the surface of the sieve. The sieve is then gently removed from the water, drained, and unlatched to produce two subsamples.

In contrast, the sieve diameter splitting method uses a standard 20-cm (8-in.) diameter metal or plastic sieve marked with six equally spaced (30 degrees apart) diameters (fig. 8A). The diameter markings are extended up the inside walls of the sieve and numbered 1 through 6 (fig. 8B). The sample is placed on the marked sieve, immersed in water, and gently agitated. Next, the sample is distributed uniformly across the sieve, which is then gently removed from the water and drained. A die is rolled to obtain an unbiased determination of which diameter (1-6) to use for splitting the sample. A metal-edged ruler (20 cm long for a standard sieve) is used to divide the sieve contents into halves by aligning the ruler with the appropriate inscribed diameter markings (diameter 5 in the example presented in fig. 8B) and pressing the ruler against the bottom of the sieve. A small scraper, such as a putty knife, is used to help separate the sample into halves by pulling material away from the ruler. This process is completed using a wash bottle to remove and concentrate any remaining sample material.





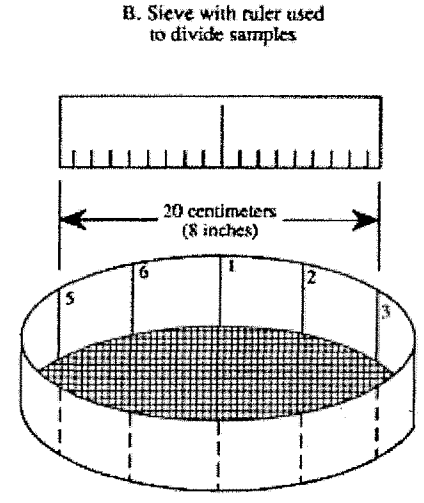
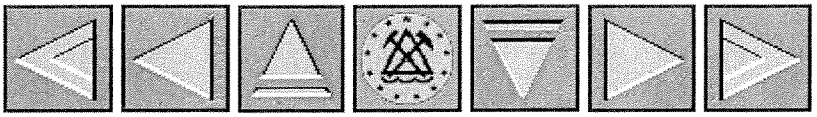


Figure 8.--Diagram of the diameter markings (A) and the alignment of the sample divider (B) used in the sieve diameter sample splitting procedure.



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Once the sample has been split, a coin toss (heads indicates the right half, tails the left half) or throw of a die (even indicates the right half, odd the left half) is used to randomly select half of the sample. If the sample being processed is an elutriate sample, then the half of the sample selected is retained for analysis and the other half is discarded. If the sample being processed is a main-body sample, then the half of the sample selected is designated as the main-body component and the other half is designated as the "split" sample component. The split-sample component is retained for QA/QC purposes and shipped to the BQAU. All sample components must be labeled and preserved properly.

Some particularly large samples may require repeated splitting to obtain suitable volumes (less than or equal to 0.75 L) of main-body, split, and elutriate sample components. These samples may also be larger than the capacity of the sieve used for splitt ing the sample. In these circumstances, manageable portions of the sample are split until the entire sample has been split into halves. The multiple portions of split sample that are produced are assigned to their component halves in an unbiased manner (that is, by coin toss or roll of the die). If the resulting split-sample component (elutriate, split, or main-body) exceeds 0.75 L, it is split again. Careful records of the number of splits performed and the portion of the original sample retained for analysis are kept and entered on the appropriate field data sheet.

A flow chart showing how the various sample components are split and reduced to one-fourth of their original volume is illustrated in figure 9. In actual field situations further sample splitting may be necessary. Each time the elutriate sample component is split, a portion of it is discarded. In contrast, when the main-body sample component is initially split (one-half split), an additional sample component is generated (split) and no sample material is discarded. When subsequent splits are necessary , they are applied to the "split" and "main-body" sample components and portions of each are discarded (for example, the one-fourth split). The main-body and split-sample components are processed in parallel so that the split-sample component sent to the BQAU is, in theory, identical to the main-body sample component sent to the contract laboratory. Consequently, the split samples can be processed and used to evaluate the performance of contract laboratories.

After samples have been processed, they are transferred to appropriately sized plastic sample containers. Wide-mouth high-density polyethylene plastic 0.5-L (19-oz) and 1-L (32-oz) jars are recommended. These are closed with compatible unbreakable plast ic screw-on lids with liners.

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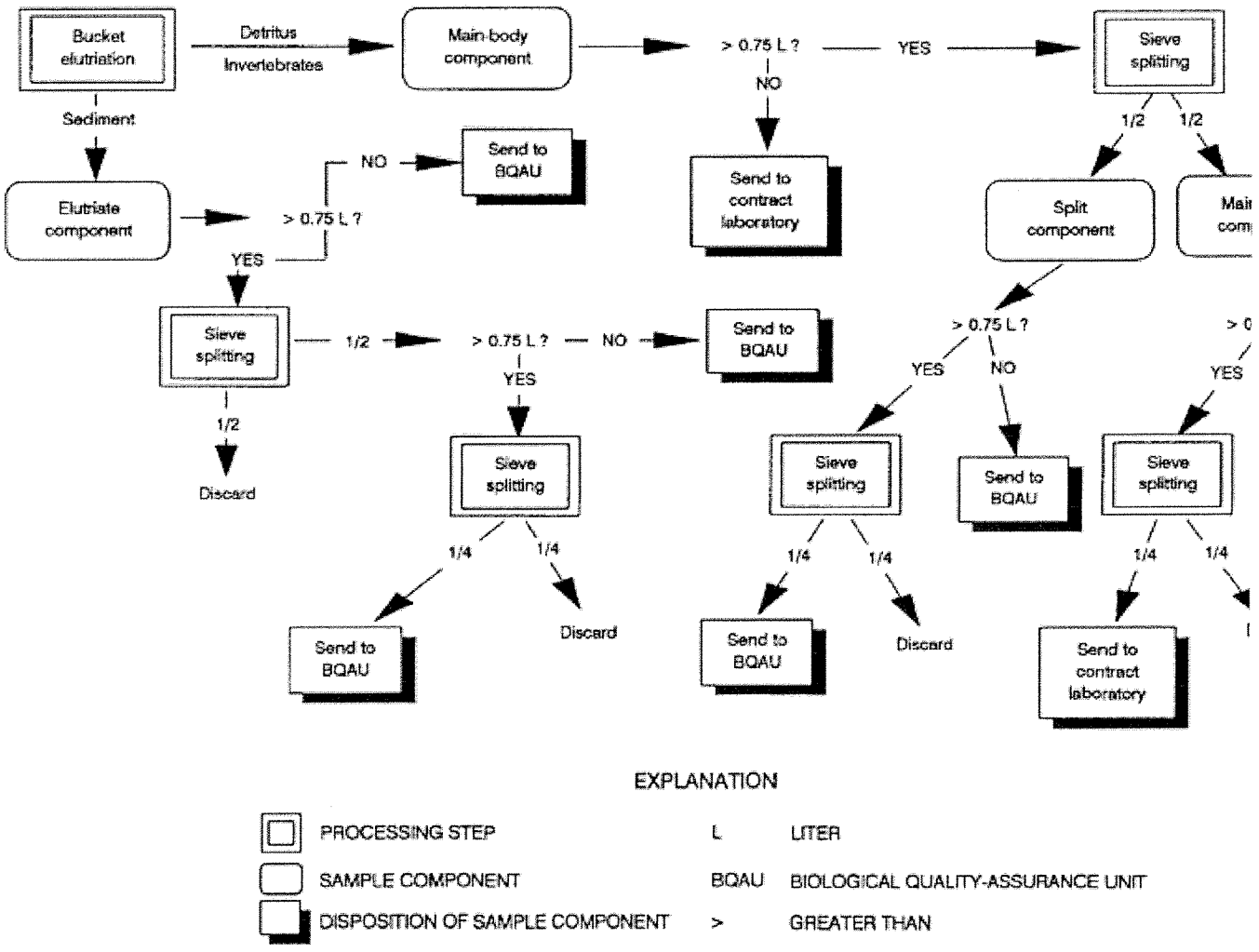
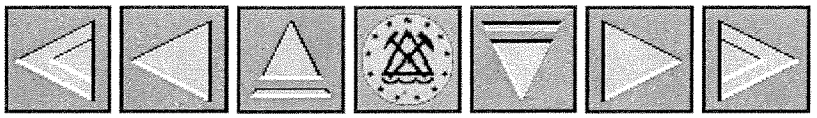


Figure 9.--Flow chart illustrating the repeated splitting of elutriate and main-body benthic invertebrate sample components to achieve appropriate sample volumes.



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An internal sample label (fig. 10) is filled out and placed in the sample container. This label is printed or photocopied on plastic or 100-percent rag, acid-neutralized paper using formalin- and alcohol-resistant inks. All spaces for information on the label are filled in using a pencil or rapidograph containing water-, formalin-, and alcohol-resistant ink. The site names and site identification numbers entered on the labels correspond to official USGS station names and station identification numbers assigned to the basic fixed sites, typically a site name and number for an existing gaging station. The date of collection is entered in two-digit format as month, day, and year. The proportion of the sample retained for processing (subsample) and the names of the people responsible for collecting the sample are entered in the proper place on the label. The appropriate information regarding sample type and mesh size is circled and the sample-identification code entered.

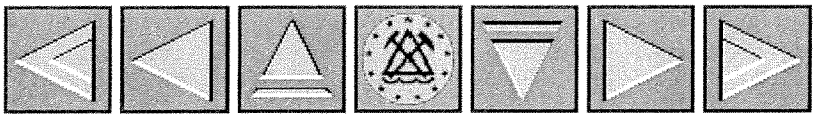
NAWQA INVERTEBRATE SAMPLE

Site Name: Yakima R. blw Toppenish Cr.
Site ID No.: 12507525
Date: 10 / 22 /90 Subsample 1/4
Collected by: Irving M. Bugged,
Bruce N. Othaus, Charlie L. Ambake
Type of Sample:
QMH (RTH) DTH
Large-rare Main Elutriate (Split)
Mesh: (425 µm) 210 µm Reach: A
ID Code: YAKI 1090IRS0142D

EXPLANATION

NAWQA	NATIONAL WATER-QUALITY ASSESSMENT
ID No.	IDENTIFICATION NUMBER
QMH	QUALITATIVE MULTIHABITAT
RTH	RICHEST-TARGETED HABITAT
DTH	DEPOSITIONAL-TARGETED HABITAT
µm	MICRON
YAKI	YAKIMA RIVER BASIN (See table 2)

Figure 10.--Example of a completed internal sample label.



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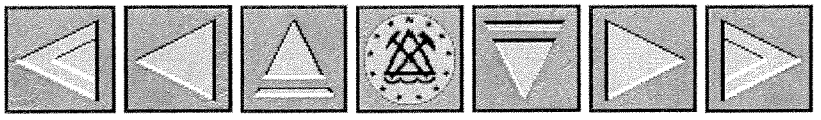
The sample should occupy approximately one-half to three-fourths of the container volume. A solution of 10-percent buffered formalin is added to bring the total volume to within 2 cm of the top of the jar. The jar is then capped and slowly inverted several times to mix the contents of the jar with the formalin solution and to remove any air trapped in the sample matrix. The jar is then opened and topped off with 10-percent buffered formalin. Air in the sample container should be minimized in order to prevent damage to specimens during shipment. As an additional safeguard against leakage, plastic electrical tape is tightly wrapped around the junction between the jar and lid. The outside of the sample container is cleaned and dried, and an appropriate external sample label, indicating the sample-identification code number, is affixed to the outside of the jar with a large piece of transparent packaging tape.

Alcohol- and formalin- resistant inks are specified because contract laboratories will transfer samples fixed in 10-percent buffered formalin to 70-percent ethanol upon receipt. Consequently, study units should ship samples to the contract laboratory as soon as possible. If samples are rich in organic matter and will be held for a week or more before shipping to the contract laboratory, the formalin in the sample needs to be changed to 70-percent ethanol. Waste formalin is regarded as hazardous material and should be disposed of properly (U.S. Environmental Protection Agency, 1981).

The sample-identification code used in the internal (fig. 10) and external (fig. 11) labels is a 16-character sequence that uniquely identifies each sample container. Characters 1-4 are the abbreviation of the study-unit name (table 2) and 5-8 are the collection date (month and year). Characters 9-11 describe the type of sample contained in the sample jar. An "I" indicates an ecological survey benthic invertebrate sample. Other possibilities include algal and fish community samples for ecological surveys ("A" and "F," respectively), bed sediment samples ("S"), and tissue samples ("T"). The subtype codes (characters 10 and 11) listed in figure 11 are specific for the ecological survey benthic invertebrate sample type ("I") and must be interpreted in conjunction with character 9 (that is, letter codes in position 10 and 11 may be repeated among sample types). Character 10 indicates whether the sample is an RTH, a DTH, or a QMH sample. Character 11 identifies the sample component (large-rare, main-body, split, or elutriate). The sample number is represented by characters 12-15 and character 16 represents the bottle sequence code ("A to Z").

The sample-identification code presented in figure 11 indicates the following information about the contents of the jar: they are

1. from the South Platte River Basin study unit (SPLT)
2. from a collection made in August 1992 (0892)
3. part of an ecological survey benthic invertebrate sample (I)
4. a semi-quantitative sample collected from the richest-targeted habitat type (RTH)
5. the main-body sample component that requires picking and identification at a contract laboratory (M)
6. part of sample number 10 (0010, a unique number within a study unit for that year) and
7. the third sample container (C) associated with this sample number.



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These sample-identification codes are used by the study unit and the BQAU to track samples throughout sample processing.

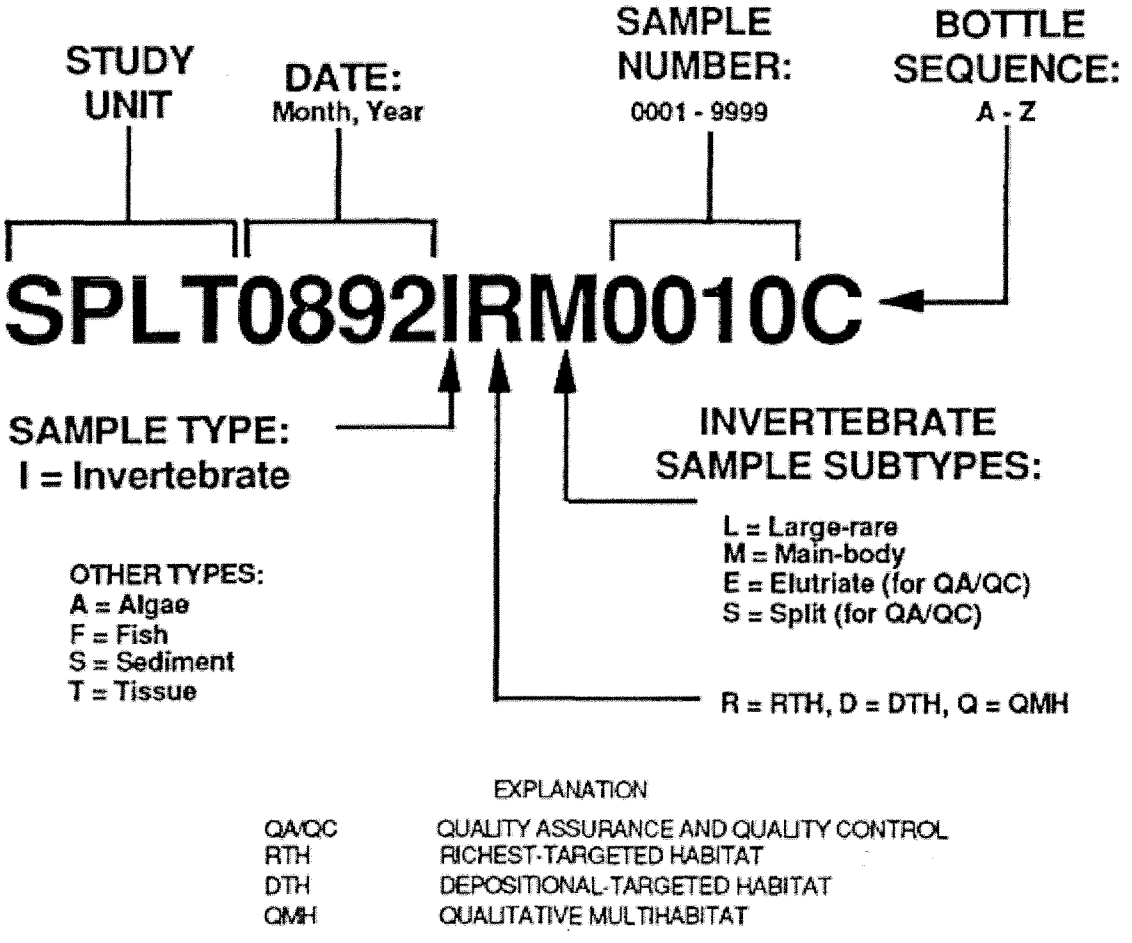

Figure 11.--Example of a completed external sample label with explanation of the 16-character sample-identification code used by the National Water-Quality Assessment Program.

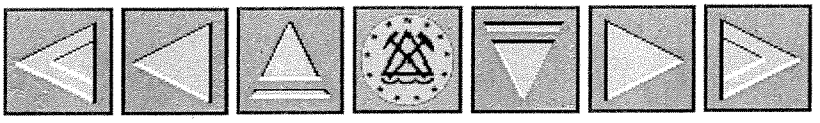

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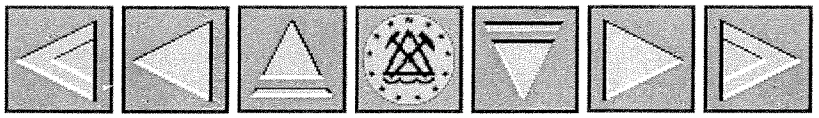
Table 2.--*Abbreviations of study-unit names used in the 16-character sample-identification code*

[Abbr., abbreviation]

Study Unit	Abbr.	Study Unit	Abbr.
Albemarle-Pamlico Drainage	ALBE	Northern Rockies Intermontaine Basins	NROK
Allegheny and Monongahela Basins	ALGH	Oahu	OAHU
Apalachicola-Chattahoochee-Flint River Basin	ACFB	Ozark Plateaus	OZRK
Central Columbia Plateau	CCPT	Potomac River Basin	POTO
Central High Plains	CHPL	Puget Sound Drainages	PUGT
Central Nebraska Basins	CNBR	Red River of the North	REDN
Central Oklahoma	COKL	Rio Grande Valley	RIOG
Cheyenne and Belle	CHEY	Sacramento Basin	SACR
Chicot-Evangeline	CHEV	San Joaquin-Tulare Basins	SANJ
Connecticut, Housatonic, and Thames River Basins	CONN	Santa Ana Basin	SANA
Cook Inlet	COOK	Santee Basin and Coastal Drainage	SANT
Delaware River Basin	DELR	South Central Texas	SCTX
Delmarva Peninsula	DLMV	South Platte River Basin	SPLT
Eastern Iowa Basins	EIWA	Southeastern New England	SENE
Georgia-Florida Coastal Plain	GAFL	Southern Arizona	SOAZ
Great and Little Miami River Basin	MIAM	Southern Florida	SOFL
Great Salt Lake Basin	GRSL	Southern High Plains	SHPL
Hudson River Basin	HDSN	Southern Illinois	SILL
Kanawha-New River Basin	KANA	Trinity River Basin	TRIN
Kansas River Basin	KANS	Upper Arkansas River	UARK
Kentucky River Basin	KNTY	Upper Colorado River Basin	UCOL
Lake Erie-Saint Clair Drainage	LERI	Upper Illinois River Basin	UIRB
Long Island-New Jersey Coastal Plain	LINJ	Upper Mississippi River Basin	UMIS
Lower Susquehanna River Basin	LSUS	Upper Snake River Basin	USNK
Lower Tennessee River Basin	LTEN	Upper Tennessee River Basin	UTEN
Mississippi Embayment	MISE	Western Lake Michigan Drainages	WMIC
Mobile River	MOBL	White River Basin	WHIT
Nevada Basin and Range	NVBR	Willamette Basin	WILL
New Hampshire and Southern Maine Basins	NHME	Yakima River Basin	YAKI
North Platte Basin	NPLT	Yellowstone River Basin	YELL



Sample-identification codes are unique for each sample and sample jar. Figure 12 illustrates the sample-identification codes that might result from processing one composite sample. In this example, the large-rare component from an RTH sample (IRL) was placed into two sample containers (A and B). The main-body (M), elutriate (E), and split (S) sample components are contained in single containers (C, D, and E, respectively). Sample-bottle sequence codes are assigned to sample components in the following sequence: large-rare, main-body, elutriate, and split. Assigning sample-bottle sequence codes in this manner ensures that contract laboratories cannot determine which samples have supporting quality-assurance samples based on gaps in the sample code sequences received by the laboratory.

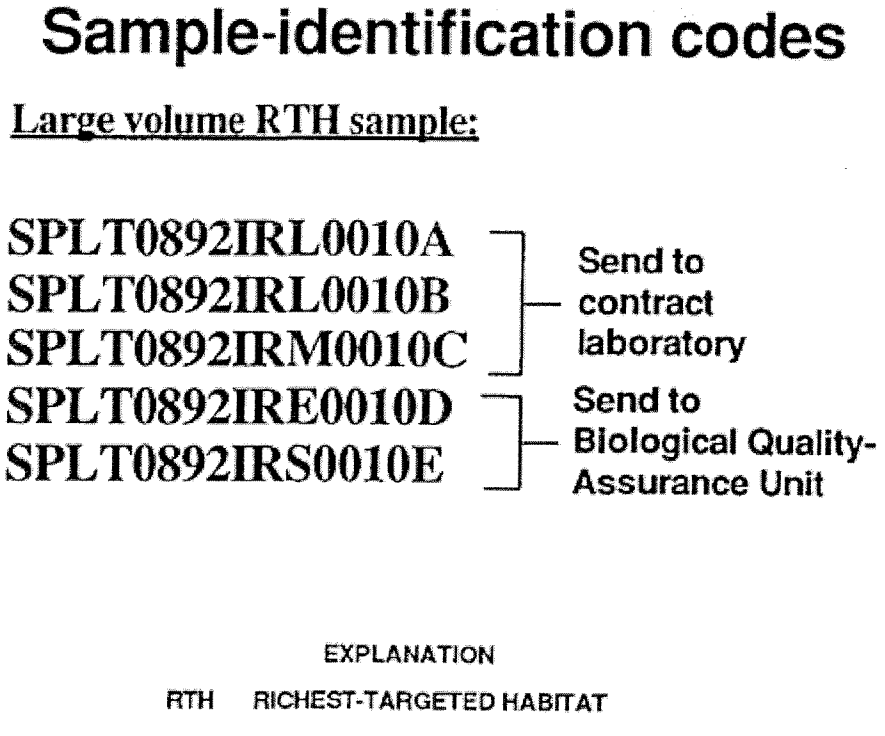
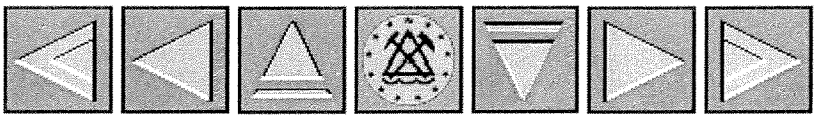


Figure 12.--Examples of sample-identification codes generated from one composite sample.



FIELD DATA SHEETS

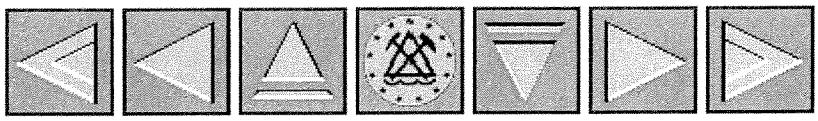
Field data sheets are printed or photocopied on waterproof plastic-coated paper using water-, formalin-, and ethanol-resistant inks. Three field data sheets are needed for invertebrate sampling: one for QMH samples and two for semi-quantitative samples. Each field sheet has a space for entering a page number and a cumulative page number. The page number is used by each field sampling team to consecutively number field data sheets. Therefore, the page number is unique only for a particular field sampl ing team during a specific field sampling effort. The cumulative page number is used by the study unit to consecutively number invertebrate field data sheets obtained from multiple field sampling efforts and teams. This procedure provides a unique number for each data sheet placed in the study-unit biological field data notebook and(or) file.

Qualitative Multihabitat Field Data Sheet

The QMH field data sheet (fig. 13) is divided into eight sections for entering site and sampling information, and information on the instream habitat types sampled and the samplers used. The "SITE INFORMATION" section is for recording the sampling date, the site name, the site identification (ID) number, and the identity of the sampling team members. Dates are entered in double-digit numeric format for month, day, and year (for example, November 9, 1992, is entered as 11/09/92). The site name is either a descriptive name (for example, Yakima River at Kiona, Wash.) or an official USGS station name if one is available. The site identification number is an official USGS station number, such as 12510500. The site name and identification number must be us ed consistently within the study unit and among NAWQA Program cycles. The full names of the sampling team members are entered with the team leader's name enclosed in parentheses.

The section on "RELATED SAMPLING ACTIVITIES" contains a checklist for other sampling activities that co-occur or immediately precede the benthic invertebrate sampling. Activities which occur, but are not listed in this section, are specified in the "Other" category. Listing related sampling activities makes it easier to find other supporting data or to determine possible factors that can interfere with invertebrate sampling.

The "PHYSICAL SITE CONDITIONS" section is for recording weather conditions, water temperature, and river stage. Data on local weather conditions that might affect sampling, such as percent cloud cover, wind direction and speed, and type, relative intensity, and duration of precipitation, are entered in the appropriate spaces. Other relevant weather-related conditions are entered in the space provided for "Other." Water temperature and time of determination are entered for the start and finish of the o verall sampling effort (QMH, RTH, and DTH sampling). All times are entered in a 24-hour format. Finally, the river stage is recorded, as well as the time of determination. Stage data place the hydrologic conditions at the time of sampling in the overall context of the annual hydrograph for gaged sampling sites.



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QUALITATIVE MULTIHABITAT (QMH) SAMPLE

SITE INFORMATION	
Site name:	Date: (MM-DD-YY)
Site ID number:	
Sampling team (leader):	

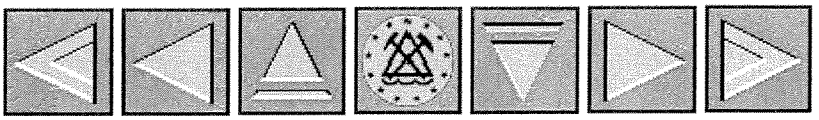
RELATED SAMPLING ACTIVITIES (check appropriate items)							
	Water chemistry		Discharge		Habitat survey		Other (specify):
	Algae survey		Fish survey		Bed sediment		Tissues

PHYSICAL SITE CONDITIONS			
Cloud cover: %	Wind:	Precipitation:	
Other:			
Water temp -- Start: °C @ time h	Finish: °C @ time h		
River stage: ft @ time h			

SAMPLING INFORMATION				
Sampling duration -- Start: h End: h			Reach ID	Mesh size: 210 µm
	CONTRACT LABORATORY SAMPLES		QA/QC SAMPLES	
	Large-rare	Main-body	Elutriate	Split
Sample ID number				
Subsample	1/1			
Elutriation method: Bucket Other (specify):				
Sample-splitting method: Sieve diameter Other (specify):				

SAMPLE AND PHOTOGRAPH NOTES

Figure 13.--A two-page field data sheet used to record sampling information during the collection of qualitative multihabitat (QMH) samples. (Page 1)



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INSTREAM HABITAT TYPES SAMPLED:

Instream Habitat Matrix: Indicate the habitat types sampled by inserting the code for the sampler(s) used. Enter NA if a habitat type is not present or NC if a habitat type is present but not sampled.

Riffle	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel						
Channel margin						
Island margin						
Run	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel						
Channel margin						
Island margin						
Pool	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel						
Channel margin						
Island margin						

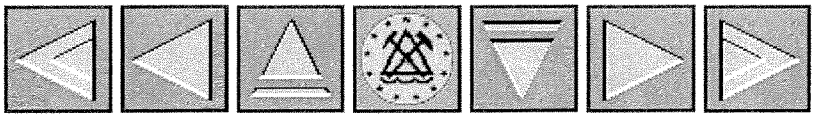
EQUAL SAMPLING EFFORT PROCEDURE

	Time		No. samples		Area		Other (specify):

SAMPLER CODES:

Sampler type	*Code*	*Sampler type*	*Code*	*Sampler type*	*Code*
Standard Surber (929 cm^2)	1	Macan (1,225 cm^2)	12	Visual collection - leaf debris	23
Slack sampler (2,500 cm^2)	2	Standard Ekman (232 cm^2)	13	Visual collection - rocks	24
Hess (855 cm^2)	3	Large Ekman (523 cm^2)	14	Visual collection - other	25
Demne (1,800 cm^2)	4	Extra-large Ekman (929 cm^2)	15	Snag collection with dip net	26
Small stovepipe (182 cm^2)	5	Standard Ponar (523 cm^2)	16	Snag collection with Slack net	27
Box sampler	6	Petite Ponar (232 cm^2)	17		28
D-frame aquatic net	7	Shipek (413 cm^2)	18		29
Seine (1/8 in. mesh)	8	Artificial snags	19		30
Hand screen collector	9	Artificial substrate basket	20		31
Kick sample	10	Hester Dendy	21		32
Thorp snag sampler	11	Visual collection - wood	22		33

Figure 13.--A two-page field data sheet used to record sampling information during the collection of qualitative multihabitat (QMH) samples. (Page 2)



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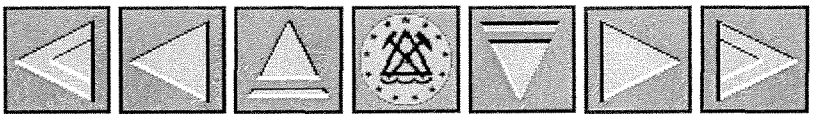
The "SAMPLING INFORMATION" section is for recording the sampling start and end times, the sampling reach identifier (A, B, or C), the mesh size associated with the sample type (210 µm for QMH samples), sample-identification numbers, the proportion of the sample (subsample) retained for analysis, and the methods used for sample elutriation and splitting. The beginning and ending times of the qualitative sampling effort are recorded to provide a chronology of sampling activities in the sampling reach. Inf ormation related to sample processing methods includes the mesh size used (preprinted on the data sheet), the elutriation method used (circle "Bucket" if bucket elutriation is used or specify the appropriate technique next to the "Other" category), and th e sample-splitting method used (circle "Sieve diameter" if the sieve diameter method is used or specify the appropriate technique next to the "Other" category). If sample elutriation or sample-splitting procedures are not used in the processing of the co mposite sample, then "NA" (not applicable) is entered in the appropriate space. Also, enter "NA" in the cells of the matrix associated with sample components that were not generated during processing. For example, if the main-body sample component was 0.75 L or less, then a split-sample component is not generated, and "NA" is entered in the spaces related to the split-sample component and the sample-splitting technique. The general objective in completing field data sheets is to enter information into all data fields, thereby avoiding ambiguities presented by blank data fields.

A unique identification number ("Sample ID number") is entered in the "SAMPLING INFORMATION" section for each sample container produced. This identification number is the 16-character sample-identification code. All containers produced from the processi ng of a single composite sample have identical sample-identification code numbers with the exceptions of the eleventh character, which identifies the sample component type (L, M, S, or E), and the last character, which is unique for each sample container (fig. 11). If multiple jars are used to contain a sample component (typically, the large-rare component only), then the first 15 characters are entered, followed by the appropriate sample container letter codes assigned to each jar, separated by commas. For example, the entry LSUS0992IRL0005A,B,C indicates that the large-rare sample component (L) is contained in three jars: A, B, and C. Any modifications to standard sample-collection, processing, and labeling procedures are recorded in the "SAMPLE AND PHOTOGRAPH NOTES" section.

The "SAMPLING INFORMATION" section also contains an entry space labeled "Subsample." If the sample component (elutriate or main-body) is split, this space is used to record the proportion of the total composite sample retained for processing. The proportion entered here is used to calculate the relative abundances of organisms in the composite sample and to determine the efficacy of sample elutriation and splitting. Consequently, it is extremely important that the information entered here be accurate. The subsample entry for the large-rare sample component (1/1) is preprinted on the data sheet to indicate that this component is never a subsample. Proportions are entered in the same manner as used for the large-rare sample component: 1/1, no sample splitting; 1/2, one-half of the sample is retained for processing; and 1/4, one-fourth of the sample is retained for processing.



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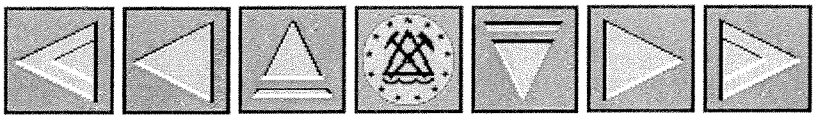
Space is provided in the "SAMPLE AND PHOTOGRAPH NOTES" section for recording site notes and details of photographs taken. The purpose of recording information about photographs is to apprise the data-sheet reviewer of the availability and location of su pporting photographs. Desired information about photographs includes the medium used (for example, prints, slides, or video), the repository for the photographs, and a brief description of the subject of each photograph. Technical details associated wit h the photographs, such as type of camera, exposure settings, focal length, and type of film used, may be entered in a separate photodocumentation data sheet. This section is also used to record information on unusual or unique site conditions, problems encountered in sampling, departures from "normal" sample-collection techniques, and recommendations for modifying sampling procedures and approaches. Such field notes are valuable for interpreting outliers in the data and for improving sampling procedure s and design.

The back of the QMH field data sheet contains three sections. The section labeled "INSTREAM HABITAT TYPES SAMPLED" lists the matrix of 51 instream habitat types that can occur in each sampling reach. The codes for the type(s) of sampler(s) used to sampl e the various instream habitat types are entered in the appropriate cells of this matrix. Sampler codes are provided in the section "SAMPLER CODES" of the QMH field data sheet. There are provisions for adding new samplers in the QMH sampler code matrix after entry 27. These new entries relate to the specific data sheet upon which they are entered. They are not nationally consistent as are sampler codes 1-27. The section labeled "EQUAL SAMPLING EFFORT PROCEDURE" is for recording which procedure is use d to divide sampling effort among the various instream habitats sampled.

Figure 14 illustrates how the appropriate sampler codes are entered into the cells corresponding to the various instream habitat types. If multiple samplers are used within a habitat, the sampler codes are separated by a comma. If a habitat type is not present in the sampling reach, "NA" (not applicable) is entered in the corresponding cell. If the habitat type is present but not sampled, "NC" (not collected) is entered. The example provided in figure 14 indicates that 28 of the 51 instream habitat ty pes were present at this site and 27 were sampled. One type--run, island-margin, natural-bed--was present but not collected. Six different samplers (fig. 13 codes in parentheses) were used: Slack sampler (2), D-frame aquatic net (7), seine (8), petite Ponar (17), visual collection from wood (22), and visual collection from leaf debris (23).



INSTREAM HABITAT TYPES SAMPLED:

Instream Habitat Matrix: Indicate the habitat types sampled by inserting the code for the sampler(s) used. Enter NA if a habitat type is not present or NC if a habitat type is present but not sampled.

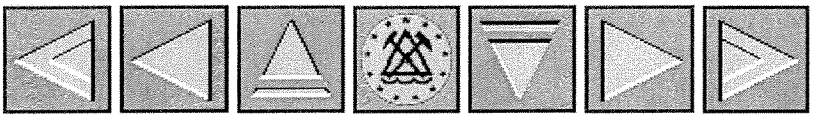
Riffle	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel	2	NA		7	NA	NA
Channel margin	7, 22, 23	NA	7	7	22	8
Island margin	7, 22, 23	NA	NA	7	22	8
Run	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel	2	NA		7	NA	NA
Channel margin	7, 22, 23	NA	7	7	22	8
Island margin	NC	NA	NA	7	22	8
Pool	Natural bed	Manufactured bed	Slough	Macrophyte bed	Woody snag	Bar
Main channel	17	NA		NA	NA	NA
Channel margin	7, 22, 23	NA	7	7	22	8
Island margin	NA	NA	NA	NA	NA	NA

EXPLANATION

7, 22, 23 SAMPLER CODES IN FIGURE 13

COMBINATION OF HABITAT DESCRIPTORS THAT ARE NOT POSSIBLE

Figure 14.--Example of information entered into the instream habitat matrix of the qualitative multihabitat field data sheet (fig. 13).



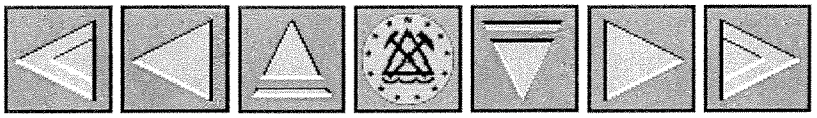
Semi-Quantitative Field Data Sheets

Field data sheets for the semi-quantitative samples (RTH and DTH) (fig. 15 for instream habitat sampling and fig.16 for snag habitat sampling) are divided into eight sections. The first five sections are identical to the first five sections of the QMH fi eld data sheet (fig. 13) with the following two exceptions: the mesh sizes differ and there is an additional line in the "SAMPLING INFORMATION" section of the semi-quantitative sampling field data sheet. This line, "Number of samples constituting compos ite," is used for entering the number of individual samples that were collected to form the composite sample that characterizes the sampling reach. This number should be five or greater.

The sixth section of the semi-quantitative sampling field data sheet shown in figure 15, "INSTREAM HABITAT TYPE SAMPLED," is similar to the corresponding section of the QMH field data sheet in that it contains information on the instream habitat type samp led and the sampling equipment used. However, unlike QMH sampling, semi-quantitative sampling (RTH and DTH) is done in only one instream habitat type using a single sampler type. The instream habitat type sampled is indicated by checking the boxes that correspond to the appropriate geomorphic channel unit, channel boundary, and channel features that define the habitat type. The code for the sampler used is entered on the last line of the block. The "SAMPLER CODES" section, the seventh section, is identical to the corresponding section on the QMH field data sheet.

Space is provided in the "INSTREAM HABITAT TYPE SAMPLED" section (labeled "Other") for describing an instream habitat type that does not correspond to any of the 51 instream habitat types described by the habitat matrix. This provision is designed to address those rare instances when a sampling reach contains a highly unusual instream habitat type that fits the definition of RTH or DTH but cannot be described using the existing instream habitat matrix (fig. 2). This provision is to be used conservativel y and in consultation with the regional biologists, North Carolina Ecology Group, and national synthesis teams.

The last section of the RTH and DTH field data sheet (fig. 15), "MICROHABITAT CHARACTERIZATION," is for recording information on the water depth, current velocity, type of current meter used, and substrate characteristics associated with each of the sampling locations from which samples were collected for the composite sample. Water depths are measured either directly using a meter stick (wadeable sites) or indirectly using a depth finder (nonwadeable sites). Current velocity is measured at six-tenths of the water depth for wadeable sites or at two- and eight-tenths water depth at nonwadeable sites (separate counts and seconds with a '/' on the data sheet and check the type of current meter used). When velocity measurements are made at two- and eight-tenths water depth, the two values are averaged and recorded in the velocity column. Depths and velocities are obtained prior to positioning the sampler. When possible, these measurements are taken at a representative point, avoiding large rocks, eddies, and other features of the sampling area that would yield nonrepresentative results within the area to be sampled, or at multiple points within this area, if time permits (record average depth and velocity). If this is not possible or if the measurements would disturb the sampling site, then the measurements are made in an adjacent area with similar characteristics.



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SEMI-QUANTITATIVE TARGETED-HABITAT SAMPLES: RTH DTH (circle one)

SITE INFORMATION

Site name:	Date: (MM-DD-YY)
Site ID number:	
Sampling team (leader):	

RELATED SAMPLING ACTIVITIES (check appropriate items)

	Water chemistry		Discharge		Habitat survey		Other (specify):
	Algae survey		Fish survey		Bed sediment		Tissues

PHYSICAL SITE CONDITIONS

Cloud cover: %	Wind:	Precipitation:
Other:		
Water temp -- Start: °C @ time h	Finish: °C @ time h	
River stage: ft @ time h		

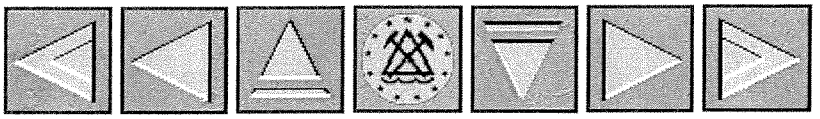
SAMPLING INFORMATION

Sampling duration -- Start: h	End: h	Reach ID	Mesh size: 425 µm	
	CONTRACT LABORATORY SAMPLES		QA/QC SAMPLES	
	Large-rare	Main-body	Elutriate	Split
Sample ID number				
Subsample	1/1			
Number of samples constituting composite:				
Elutriation method:	Bucket	Other (specify):		
Sample-splitting method:	Sieve diameter	Other (specify):		

SAMPLE AND PHOTOGRAPH NOTES

Figure 15.--A two-page field data sheet used to record information during the collection of semi-quantitative samples from the faunistically richest- (RTH) and depositional- (DTH) targeted

habitats. (Page 1)



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INSTREAM HABITAT TYPE SAMPLED	*Check the appropriate boxes.*
GEOMORPHIC CHANNEL UNIT:	☐ Riffle ☐ Run ☐ Pool (check one)
CHANNEL BOUNDARIES:	☐ Main channel ☐ Channel margin ☐ Island margin
CHANNEL FEATURES:	☐ Natural bed ☐ Manufactured bed ☐ Slough ☐ Macrophyte bed ☐ Snag ☐ Bar
Other (specify):	
Sampler used (enter code number):	

SAMPLER CODES:

Sampler type	*Code*	*Sampler type*	*Code*	*Sampler type*	*Code*
Standard Surber (929 cm^2)	1	Macan (1,225 cm^2)	12	Visual collection - leaf debris	23
Slack sampler (2,500 cm^2)	2	Standard Ekman (232 cm^2)	13	Visual collection - rocks	24
Hess (855 cm^2)	3	Large Ekman (523 cm^2)	14	Visual collection - other	25
Dome (1,800 cm^2)	4	Extra-large Ekman (929 cm^2)	15	Snag collection with dip net	26
Small stovepipe (182 cm^2)	5	Standard Ponar (523 cm^2)	16	Snag collection with Slack net	27
Box sampler	6	Petite Ponar (232 cm^2)	17		28
D-frame aquatic net	7	Shipek (413 cm^2)	18		29
Seine (1/8 in. mesh)	8	Artificial snags	19		30
Hand screen collector	9	Artificial substrate basket	20		31
Kick sample	10	Hester Dendy	21		32
Thorp snag sampler	11	Visual collection - wood	22		33

MICROHABITAT CHARACTERIZATION

NA: Characterization element is not applicable. NC: Characterization element is applicable but was not measured.

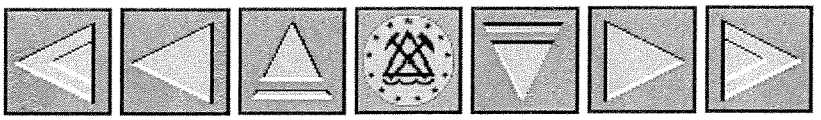
Sampling location number	Water depth (cm)	Substrate type		Substrate embeddedness class	Current velocity		
					Counts/seconds		Velocity (cm/s)
		Dominant	Co-dominant		0.6 or 0.2 Depth	0.8 Depth	

Velocity meter used: ☐ Pygmy ☐ AA ☐ Marsh-McBirney ☐ Other (specify):

Substrate Codes: Bedrock (BR), Boulder (BO), Cobble (CL), Gravel (GV), Sand (SA), Silt (SI), Hardpan (HP), Marl (MA), Detritus (DE), Muck (MU), Artificial (AR).

Figure 15.--A two-page field data sheet used to record information during the collection of semi-

quantitative samples from the faunistically richest- (RTH) and depositional- (DTH) targeted habitats. (Page 2)



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SEMI-QUANTITATIVE WOODY SNAG SAMPLES: RTH DTH (circle one)

SITE INFORMATION	
Site name:	Date: (MM-DD-YY)
Site ID number:	
Sampling team (leader):	

RELATED SAMPLING ACTIVITIES (check appropriate items)							
	Water chemistry		Discharge		Habitat survey		Other (specify):
	Algae survey		Fish survey		Bed sediment		Tissues

PHYSICAL SITE CONDITIONS			
Cloud cover: %	Wind:	Precipitation:	
Other:			
Water temp -- Start: °C @ time h	Finish: °C @ time h		
River stage: ft @ time h			

SAMPLING INFORMATION				
Sampling duration -- Start: h End: h		Reach ID	Mesh size: 425 µm	
	CONTRACT LABORATORY SAMPLES		QA/QC SAMPLES	
	Large-rare	Main-body	Elutriate	Split
Sample ID number				
Subsample	1/1			
Number of samples constituting composite:				
Elutriation method: Bucket Other (specify):				
Sample-splitting method: Sieve diameter Other (specify):				

SAMPLE AND PHOTOGRAPH NOTES

Figure 16.--A two-page field data sheet used to record information during the collection of semi-

quantitative samples from snag habitats. (Page 1)

PAGE # [ ] CUMULATIVE PAGE # [ ]

SNAG HABITAT TYPE SAMPLED *Check the appropriate boxes.*

GEOMORPHIC CHANNEL UNIT: [ ] Riffle [ ] Run [ ] Pool

CHANNEL BOUNDARIES: [ ] Main channel [ ] Channel margin [ ] Island margin

CHANNEL FEATURES: [ ] Natural bed [ ] Manufactured bed [ ] Slough [ ] Macrophyte bed [X] Snag [ ] Bar

Other (specify):

Sampler used (enter code number):

SNAG SAMPLER CODES:

Sampler type	*Code*	*Sampler type*	*Code*	*Sampler type*	*Code*
Thorp snag sampler	11	Visual collection - wood	22	Snag collection with Slack net	27
Artificial snags	19	Snag collection with dip net	26		28

SNAG MICROHABITAT CHARACTERIZATION

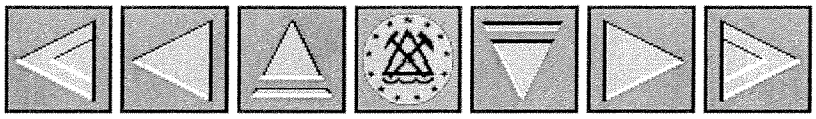
NA: Characterization element is not applicable. NC: Characterization element is applicable but was not measured.

Sampling location number	Branch length (cm)	Branch diameter (cm)	Substrate type	Water depth (cm) from surface to		Current velocity		
						Counts/seconds		Velocity (cm/s)
				Snag	Bottom	0.6 or 0.2 depth	0.8 depth	

Velocity meter used: [ ] Pygmy [ ] AA [ ] Marsh-McBirney [ ] Other (specify):

Substrate Codes: Bedrock (BR), Boulder (BO), Cobble (CL), Gravel (GV), Sand (SA), Silt (SI), Hardpan (HP), Marl (MA), Detritus (DE), Muck (MU), Artificial (AR).

Figure 16.--A two-page field data sheet used to record information during the collection of semi-quantitative samples from snag habitats. (Page 2)



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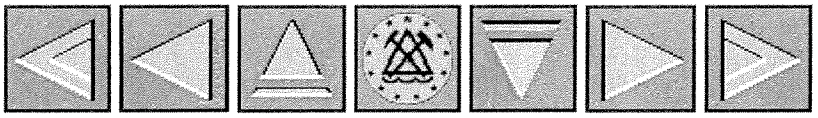
Substrate characteristics are estimated either visually in shallow, clear waters or from examination of sampler contents. Characterization consists of identifying the dominant and co-dominant substrate size fractions and embeddedness class. The substra te classifications and two-letter substrate codes are listed at the bottom of the "MICROHABITAT CHARACTERIZATION" section. The substrate size and embeddedness classifications are identical to those set forth in the habitat protocol (Meador, Hupp, and ot hers, 1993).

Snag sampling differs from other types of semi-quantitative sampling activities. Consequently, there is a separate data sheet for snag sampling (fig. 16). This data sheet differs from other semi-quantitative data sheets in that the "SAMPLER CODES" secti on contains only samplers appropriate for snags, and the "MICROHABITAT CHARACTERIZATION" section includes space for recording approximate length and width of the branches sampled and the depth to the snag and streambed. More than one snag may be collecte d from a given location, such as several branches collected from a single large tree or debris accumulation. The dimensions of each of these branches are entered on the data sheet and associated with the same location number. At least five snag locations (if available), numbered sequentially from the downstream to the upstream end of the reach, are sampled within the sampling reach.

CONTRACT LABORATORIES AND THE BIOLOGICAL QUALITY-ASSURANCE UNIT

Benthic invertebrate sample processing, enumeration, and taxonomic identifications are done by outside contract laboratories under the direction of the USGS Quality Management Group's Biological Quality-Assurance Unit (BQAU), located at the National Water Quality Laboratory (NWQL) in Arvada, Colo. The BQAU oversees and coordinates all contracts for the processing and identification of benthic invertebrate samples according to standardized qualification, processing, and QA/QC criteria (Cuffney and others, 1993). It is responsible for overseeing the quality of samples processed by outside contract laboratories, in terms of the accuracy of enumeration and taxonomic identifications, and for resolving taxonomic issues within and among study units. The BQAU also oversees the entry of contractor data into the National Water Information System-II (NWIS-II) data base, maintains reference collections, and deposits voucher specimens in outside museums.

Study-unit personnel send sample components to the contract laboratory (large-rare and main-body sample components) and the BQAU (elutriate and split-sample components) as soon as possible, preferably directly from the field. This procedure helps to mini mize storage of formalin-containing samples and reduces the damage or loss of specimens and samples during shipment and storage. The study-unit biologist contacts the BQAU, prior to collection of the invertebrate samples, to determine which contract labo ratory will receive the samples. The contract laboratory receives only the large-rare and main-body sample components and is not apprised of the existence or identity of quality-assurance samples.



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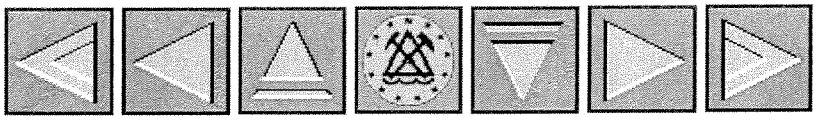
The formaldehyde solution used as a fixative for invertebrates is considered to be a hazardous material; consequently, there are specific Federal guidelines governing the shipment of these samples. In addition, individual shipping companies can have their own more stringent requirements for the packaging and labeling of preserved samples. Therefore, it is important to adhere to the following procedures when packaging, labeling, and shipping preserved samples: (1) consult the shipping company regarding its requirements prior to collecting any samples; (2) make sure that the shipping company understands that the samples contain a solution of 10-percent formalin (not 10-percent formaldehyde); and (3) be prepared to provide information on the maximum amount of preservative in each container and the total in each package. Packaging and labeling standards can require special boxes, packing materials, and labels that need to be ordered well in advance of their use. Therefore, the necessary shipping material s and instructions should be on hand prior to leaving for the field so that samples can be shipped directly from the field to the appropriate contract laboratory.

A complete list of the contents of each package, including appropriate information from the sample-identification code, is placed in the package as a packing list. Copies of the packing list are sent to the contractor and the BQAU, and one copy is retained by the study unit. The field sample log (fig. 17) serves as the basis for the packing list by indicating information on each container returned from the field. Entries listed under the "Sample description" heading include a description of the type of sample (QMH, RTH, and DTH), and the sample component (M is main-body, L is large-rare organisms, E is elutriate, and S is split). The disposition column indicates the date that the containers were shipped and their destination: BQAU or the name of the contract laboratory (XYZ Laboratory). A copy of the field sample log should be sent to the BQAU to aid in inventorying and tracking samples.

Data are returned by the contractor directly to the BQAU, which reviews the data for quality and accuracy. Provisional data are released to the study units by way of NWIS-II and are available only on the local study-unit node. Once appropriate QA/QC che cks have been completed, the BQAU, in consultation with the study-unit chief, releases taxonomic data to general access.

ADAPTING COLLECTION METHODS FOR OTHER NATIONAL WATER-QUALITY ASSESSMENT PROGRAM OBJECTIVES

The sample collection methods and techniques outlined here primarily relate to characterizing invertebrate communities at sites associated with basic fixed sites. However, these methods and procedures are readily adaptable for other objectives of the NAWQA Program that require characterization of the benthic invertebrate community, such as synoptic spatial surveys and case studies. For the most part, sample-collection and processing procedures will not have to be modified; however, samples must be label ed to ensure that they are uniquely identified as to their location and purpose. The BQAU, regional biologists, and North Carolina Ecology Group should be consulted to ensure that unique identifiers are being applied.



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FIELD SAMPLE LOG

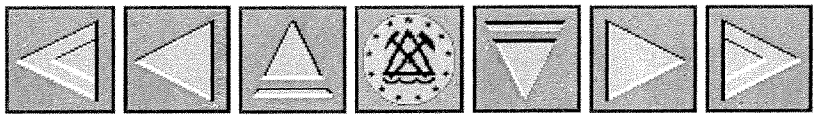
Study Unit: Lower Susquehanna River Basin **Sampling Dates:** **Start: 9-25-92** **Finish: 9-26-92**

Sample identification code	Site code	Collection date (Mo/Day/Yr)	Sample description Type	Component	Disposition
LSUS0992IQL0001A	12510500	09-25-92	QMH	L	XYZ Labs, 10/01/92
LSUS0992IQM0001B	12510500	09-25-92	QMH	M	XYZ Labs, 10/01/92
LSUS0992IQE0001C	12510500	09-25-92	QMH	E	NWQL, 10/01/92
LSUS0992IQS0001D	12510500	09-25-92	QMH	S	NWQL, 10/01/92
LSUS0992IRL0002A	12510500	09-25-92	RTH	L	XYZ Labs, 10/01/92
LSUS0992IRM0002B	12510500	09-25-92	RTH	M	XYZ,Labs 10/01/92
LSUS0992IRE0002C	12510500	09-25-92	RTH	E	NWQL, 10/01/92
LSUS0992IRS0002D	12510500	09-25-92	RTH	S	NWQL, 10/01/92
LSUS0992IDL0003A	12510500	09-25-92	DTH	L	XYZ Labs, 10/01/92
LSUS0992IDM0003B	12510500	09-25-92	DTH	M	XYZ, 10/01/92
LSUS0992IDE0003C	12510500	09-25-92	DTH	E	NWQL, 10/01/92
LSUS0992IDS0003D	12510500	09-25-92	DTH	S	NWQL, 10/01/92
LSUS0992IQL0004A	12510600	09-26-92	QMH	L	XYZ Labs, 10/01/92
LSUS0992IQM0004B	12510600	09-26-92	QMH	M	XYZ Labs, 10/01/92
LSUS0992IQE0004C	12510600	09-26-92	QMH	E	NWQL, 10/01/92
LSUS0992IQS0004D	12510600	09-26-92	QMH	S	NWQL, 10/01/92
LSUS0992IRL0005A	12510600	09-26-92	RTH	L	XYZ Labs, 10/01/92
LSUS0992IRL0005B	12510600	09-26-92	RTH	L	XYZ Labs, 10/01/92
LSUS0992IRL0005C	12510600	09-26-92	RTH	L	XYZ Labs, 10/01/92
LSUS0992IRM0005D	12510600	09-26-92	RTH	M	XYZ Labs, 10/01/92
LSUS0992IRE0005E	12510600	09-26-92	RTH	E	NWQL, 10/01/92
LSUS0992IRS0005F	12510600	09-26-92	RTH	S	NWQL, 10/01/92
LSUS0992IDL0006A	12510600	09-26-92	DTH	L	XYZ Labs, 10/01/92
LSUS0992IDM0006B	12510600	09-26-92	DTH	M	XYZ Labs, 10/01/92
LSUS0992IRL0007A	12510700	09-26-92	RTH	L	XYZ Labs, 10/01/92
LSUS0992IRM0007B	12510700	09-26-92	RTH	M	XYZ Labs, 10/01/92

EXPLANATION

QMH	QUALITATIVE MULTIHABITAT SAMPLE	NWQL	NATIONAL WATER-QUALITY LABORATORY
L	LARGE-RARE SAMPLE COMPONENT	S	SPLIT-SAMPLE COMPONENT
XYZ Labs	NAME OF CONTRACT LABORATORY	RTH	RICHEST-TARGETED HABITAT SAMPLE
M	MAIN-BODY SAMPLE COMPONENT	DTH	DEPOSITIONAL-TARGETED HABITAT SAMPLE
E	ELUTRIATE SAMPLE COMPONENT		

Figure 17.--Example of a field sample log that lists collection and disposition data for all samples collected.



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The primary modifications to the protocol described here would be to determine the degree of temporal and spatial replication necessary to support specific synoptic and case study objectives and to determine if both qualitative and semi-quantitative samplings are necessary. If the objective of the collection is to characterize the presence and absence of organisms in a sampling reach, then only the QMH methods are needed. If the objective is to characterize community structure, then semi-quantitative sampling in one or more appropriate habitat types is necessary. Case studies may require more intensive sampling than synoptic studies and may involve the use of different mesh sizes to meet specific objectives. Additional synoptic and case-study sampling approaches begin with the methods and procedures described here. They can be modified as needed, and the methods used should be carefully documented.

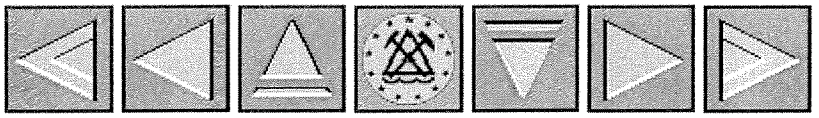
SAFETY AND HEALTH

Field sampling carries with it a potential for personal injury from equipment operation and exposure to environmental hazards. Injuries resulting from the improper use of equipment can be minimized through training in the safe operation of samplers, cars, trucks, trailers, and boats. Injuries from environmental hazards can be minimized by wearing the appropriate safety equipment, handling chemicals safely, and recognizing and avoiding known hazards (for example, poisonous plants, snakes, and insects).

All vehicle operators must have valid drivers' licenses and have attended USGS-recommended driver-training courses. Boat operators must be properly trained in boat handling, safety, and "rules-of-the-road" through participation in USGS-approved U.S. Coast Guard training courses. In addition, boat operators must be familiar with the boats that they will operate and with all of the equipment, such as safety equipment, warning devices, winches, depth finders, and anchors. All boat occupants must have approved PFD's and use them in accordance with established USGS policy. Boats, trailers, outboard motors, and other vehicles must be maintained according to USGS guidelines.

The study-unit biologist is responsible for instructing other study-unit team members in the safe operation of field sampling gear. Many of the grab samplers recommended for use in the NAWQA Program (Ponar, Shipek, Van Veen, Ekman) have jaws, joints, projections, or sufficient mass that could inflict serious injuries upon untrained personnel. All such samplers must have safety catches in place when the sampler is not being used or is in storage. All personnel on the sampling team must be instructed on operation and safe handling and storage of each sampler. Many of the larger samplers require the use of a hand or power winch. Personnel must be instructed in the maintenance and safe operation of these devices. A rope or cable cutter must be immediately accessible during the operation of any winches. This cutter must be capable of cutting the sampler loose quickly if it becomes entangled and threatens the safety of the sampling boat and its occupants.

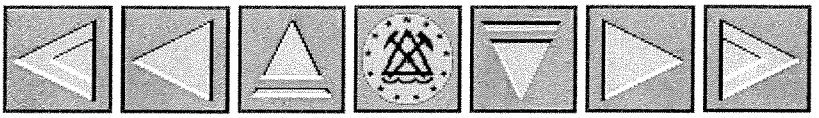
Proper safety equipment must be worn when personnel are in the field. Waders and shoulder-length gloves are worn when there is any risk that sharp, submerged objects, water-borne diseases, or toxic substances may be encountered, such as when working



down stream from a sewage-treatment plant. Because there is almost always a risk of exposure to sharp objects or water-borne diseases during collection of benthic samples, gloves and waders should be worn when these samples are collected and processed. Appro priate vaccinations against water-borne diseases should be considered when there is likelihood that such diseases could be present. Safety glasses or goggles are another consideration when personnel are working in contaminated situations and handling fix atives or preservatives. The mouth should be kept closed or covered during sample collection and processing to avoid accidental ingestion of contaminated water, sediment, or detritus. A sample or sample site is always assumed to be highly contaminated. PFD's are mandatory when working conditions involve swift or deep water. When chestwaders are used, the suspenders are worn on the outside of all clothing, including PFD's, so that the waders can be removed in an emergency, such as accidentally falling into deep or swift water. Wearing a belt around the top of chestwaders is discouraged because this complicates removal of waders in an emergency situation, and air trapped below the waist provides excessive buoyancy that interferes with the correct funct ioning of the PFD.

Preservation of invertebrate samples in the field involves the use of a 10-percent solution of buffered formalin. This material is a suspected carcinogen (U.S. Environmental Protection Agency, 1981) and should be handled only in well ventilated areas while wearing gloves and eye protection. Parts of the body that come in contact with formalin should be flushed with large amounts of water. If the eyes are involved, they should be flushed with plenty of water, and the injured person should receive medical treatment as soon as possible. A suitable eye-wash solution (for example, sterile deionized water) should be available during field work. Formalin should be purchased as 10-percent buffered formalin. The acquisition, storage, and transportation of full-strength formalin (37-percent formaldehyde) is discouraged. Formalin and formalin-preserved samples should not be transported in the passenger compartment of vehicles because of the risk of exposure if a container breaks or leaks. Instead, formalin and formalin-preserved samples should be transported in the back of pick-up trucks, in roof-top carriers, or in other areas where leaking formalin cannot endanger human health. Any waste formalin is disposed of as hazardous waste according to local, State, and Federal guidelines.

Field sampling also involves a number of environmental risks stemming from contact with the local flora and fauna. Field personnel should be instructed on how to recognize and avoid poisonous plants, snakes, and insects. Contact with poisonous plants ty pically occurs near streambanks and can best be avoided by inspecting the bank and keeping away from any suspect plants. Snakes and venomous insects are often found around trees and snags that overhang or protrude from the river bank. Such locations sho uld be approached from downstream and examined at a distance to determine if snakes, wasps, or bees are present. If a snake is present, it may be encouraged to leave by shouting, throwing small stones, or disturbing the tree that it occupies. However, if an active wasp nest or bee hive is observed, it should not be disturbed and an alternative sampling location should be chosen. If someone is thought to have been bitten by a poisonous snake, it is generally better to transport the individual immediatel y to the nearest hospital or doctor for treatment rather than applying treatment in the field prior to transport to a medical facility. Likewise, if someone is stung by a bee or wasp, that person should be observed and taken to a hospital at



the first sign of an adverse reaction (for example, hives or difficulty in breathing). Field personnel who are sensitive or allergic to bee or wasp stings should inform their project chief and avoid situations where they might be stung.

Field teams should always be composed of at least two people; no one should sample alone. All individuals in the field sampling team should be trained in basic first aid and cardiopulmonary resuscitation techniques. Each field team should be equipped wi th a suitable first-aid kit and, when possible, a cellular telephone for emergencies. A list of medical facilities closest to each sampling site should be developed and carried in each field vehicle.

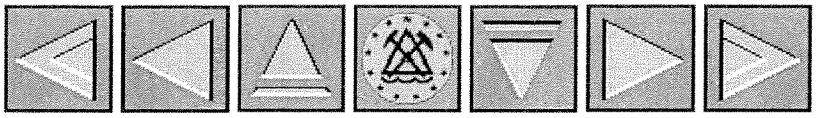
SUMMARY

Benthic invertebrate communities are characterized in the U.S. Geological Survey's National Water-Quality Assessment Program as part of an integrated physical, chemical, and biological assessment of the Nation's water quality. This multidisciplinary approach provides multiple lines of evidence for evaluating water-quality status and trends, and for refining understanding of the factors that control water quality. This is accomplished by integrated, multiyear sampling at sites chosen to represent combina tions of natural and anthropogenic factors that are important in influencing water quality locally, regionally, and nationally.

Benthic invertebrate communities are an important part of biological water-quality assessment because these organisms live in, on, or near streambed sediments where hydrophobic chemicals tend to concentrate. These organisms integrate exposures over a period of approximately a year (depending upon the length of the life cycle) and, because they are relatively sessile, characterize effects over a relatively small spatial area. They also respond to a wide variety of natural and human-engendered influences, including sedimentation, hydrologic changes, thermal pollution, xenobiotics, habitat modification, and eutrophication. These characteristics make them well suited for use in (1) assessing site-specific water-quality conditions, (2) comparing spatial pat terns of water quality, (3) integrating effects over an annual cycle, and (4) relating biological effects to physical and chemical measures of water quality.

The basis for invertebrate community characterization is the sampling reach, which is usually a length of river containing multiple examples of the dominant geomorphic features that characterize the stream segment. Each sampling reach is characterized us ing a combination of qualitative and semi-quantitative methods. A three-level hierarchy of geomorphic and channel characteristics is used to define a matrix of 51 instream habitat types. The habitat matrix supplies information used to determine where qu alitative and semi-quantitative samples are collected in the sampling reach and what types of samplers and collection methods are used.

Qualitative benthic invertebrate samples are collected from as many of the instream habitat types as are present and accessible within the sampling reach. These qualitative



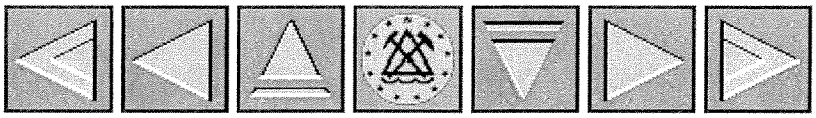
multihabitat samples, together with semi-quantitative samples, are used to develop a detailed list of the taxa present within the sampling reach at the time of collection. Semi-quantitative sampling is used to measure community structure, expressed as relative abundance of each taxon, within standardized instream response habitat types.

Two types of standardized response habitats are sampled semi-quantitatively, a faunistically "richest" (richest-targeted) and a contrasting "depositional" (depositional-targeted) habitat. The "richest" habitat is standardized, within certain limits, on the basis of the anticipated ability of the habitat to support the taxonomically richest benthic invertebrate community within the sampling reach. The "depositional" habitat is standardized as an instream habitat usually characterized by low current velo city and fine-sediment deposition. Typically, habitats selected for richest-targeted habitat sampling are those characterized by coarse substrates and high current velocities. However, in some circumstances, such as in some large rivers, fine-grained, slow-flowing "depositional" habitats may represent the most stable and faunistically "richest" habitats within the sampling reach and are sampled as the richest-targeted habitat. Relevant site information, sampling information, and microhabitat characteristics are recorded on separate field data sheets.

Typically, a single sampling reach at each basic fixed site is sampled once during a NAWQA Program cycle. However, at a subset of basic fixed sites, more intensive sampling is done to assess spatial variability among sampling reaches at a site and short-term temporal variability. Spatial variability is estimated by establishing three sampling reaches at a site and sampling all of them once during 1 year. The spatial variability assessment is usually timed so that the amount of supporting physical and chemical data is maximized. Short-term temporal variability is assessed by choosing one of the three sampling reaches and sampling it once a year for 3 consecutive years. Sites are chosen for the intensive multiple-reach and multiple-year sampling with the intent of encompassing the important sources of variability within the study unit. Such sites include reference conditions, heavily impacted sites, major land uses, and major physiographic areas of the study unit.

Each sampling reach is characterized by collecting and compositing multiple samples for qualitative and semi-quantitative sampling. Qualitative samples are composited with the objective of representing all instream habitat types accessible within the sam pling reach. Semi-quantitative targeted habitats are characterized by collecting and compositing a minimum of five samples from each of the appropriate habitat types. Benthic invertebrates are collected using the sampling method and equipment that are most appropriate for the specific instream habitat type being sampled. Dip nets, kick nets, grabs, seines, and hand-picking of substrates are recommended for obtaining qualitative samples. Slack samplers, Ponar grabs, Surber samplers, large coring devices, and artificial substrate samplers are suggested for obtaining semi-quantitative samples.

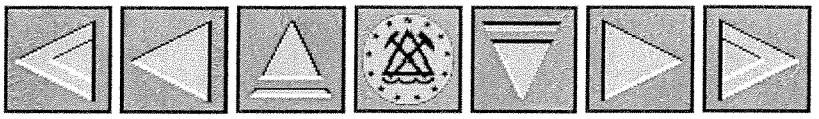
Sample processing in the field focuses on reducing the volume of composited samples to a manageable level while maximizing the number of large and rare taxa collected. Reductions in sample volume are accomplished by removing coarse organic and inorganic debris, removing large and rare taxa to a separate sample container, and elutriating, sieving,



and splitting the remaining sample. Qualitative samples are sieved to a standard size of 210-212 µm, whereas semi-quantitative samples are retained on a sieve with openings of 425 µm. Additional samples using smaller mesh sizes can be collected for semi-quantitative samples if conditions in the study unit warrant collecting smaller invertebrates. For example, using a smaller mesh size can be particularly helpful when collecting samples in streams with unstable sand or fine gravel beds where small oligochaetes and chironomids are expected to dominate the benthic invertebrate communities.

All sample containers are labeled internally and externally using standardized waterproof labels and a unique 16-character code that identifies the study unit, sampling date, type of sample, sample number, and sample component. All samples are fixed in 10-percent buffered formalin and shipped to an outside contractor for identification and enumeration of benthic invertebrates under the guidance of the Biological Quality-Assurance Unit, Arvada, Colo. This Unit has responsibility for contract development, laboratory quality assurance and quality control, entry of contractor data, national coordination of taxonomic identifications, and storage and maintenance of taxonomic collections. In addition, the BQAU monitors the effectiveness of field elutriation and splitting techniques by analyzing the elutriate and split samples from 10 percent of study-unit sample reaches.

Proper safety and health procedures need to be followed when sampling. Field personnel should be trained in the safe and proper operation, storage, transportation, and maintenance of equipment. Safety equipment must be available and must be used to prevent injury when personnel are working in fast or deep water or are handling chemicals such as formalin. Field personnel should be instructed in the recognition of poisonous plants and animals. A list of medical facilities close to each sampling site, as well as a cellular phone, should be kept with each field vehicle for use in medical emergencies.



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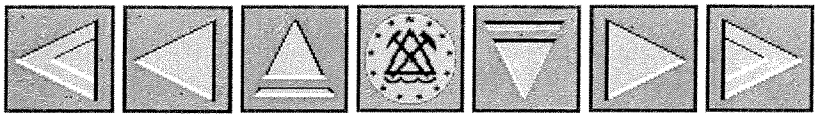

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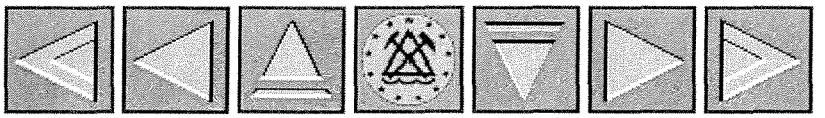

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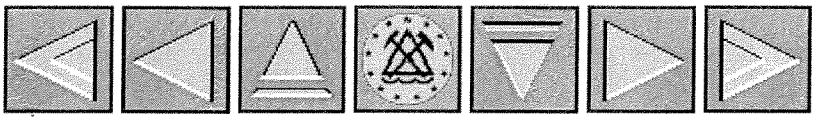

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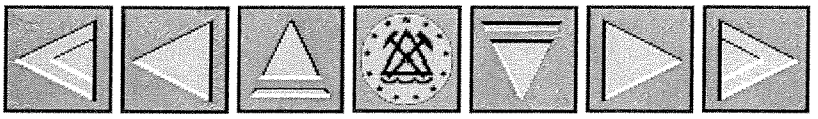

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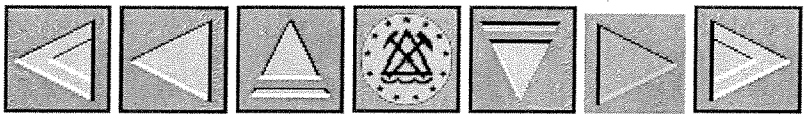

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