AIS INVENTORY AND MONITORING FRAMEWORK FOR THE GYA

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ABSTRACT

Seven of the nation's top experts in the field of aquatic invasive species (AIS) convened June 2009 to develop an AIS inventory and monitoring framework for the Greater Yellowstone Area (GYA). The framework includes a list of focus species, suggested methodologies, and a prioritization scheme. Inventory and monitoring is an important element to rapid response and successful control of threatening invasions and will provide baseline knowledge that will be helpful in our understanding of the spread and impacts of AIS upon ecosystems within the GYA. AIS surveys have been conducted by biologists from agencies, universities, non-governmental organizations (NGOs), and private individuals using various survey methodologies. Many of these previous surveys focused upon a particular species or genus and may have lacked a holistic, prioritized approach.

This paper provides a list of focal AIS, currently thought to be a high threat to the GYA, which could be used in survey efforts. However, surveyors should be cognizant of other, unexpected invasive species that may be encountered during fieldwork. Surveys should be rigorous enough for early detection of invasive species at a stage at which there may be a higher potential of control or eradication. However, there's value in both qualitative and quantitative survey methods. Physical parameters to document include GPS coordinates, temperature, calcium concentration, hardness, alkalinity, pH, nitrate and phosphate concentrations, turbidity, chlorophyll *a* concentration, flow velocity, substrate type, and habitat condition.

A probabilistic sampling approach that includes a stratified survey design is recommended. A stratified survey design that includes fixed annual high-probability sites and occasional low-probability sites would be stratified by aquatic habitat types such as rivers, lakes, and wetlands. The sites would be selected through a probability design, so that each site has a known probability of being sampled within a given year and each year, a unique set of streams, lakes and wetlands would be sampled. As a whole, the sites within each habitat type would be statistically representative of the aquatic resources within the GYA. Biological and physical habitat survey methods for each habitat type are presented in this paper. High priority survey sites will be based upon potential introduction vectors, the pristine nature of the water body, and the likelihood of further invasion/downstream spread. The specific study design will be weighted between greater use sites with a high vector potential (2/3) and pristine sites with potential risk of spread downstream (1/3).

The GYA and 100th Meridian boater assessment programs and angler use data are valuable tools to identify high probability sites to focus priority surveys. Lower use sites should also be surveyed, although less frequently than high use areas, to increase the level of survey certainty and to gather baseline data. Knowledge of focal species biology and life history are necessary to foster an informed, efficient, and effective monitoring strategy.

The use of databases such as Nonindigenous Aquatic Species (NAS) (in combination with the Nonindigenous Species Database Network (NISbase)) and predictive monitoring systems such as Global Organism Detection and Monitoring system (GODM) will be necessary to effectively use and share data.

INTRODUCTION

Inventory and monitoring is one of four primary goals of the Greater Yellowstone Area Aquatic Invasive Species Cooperative as identified in the strategic plan and further defined in the implementation plan. There is a need for clear knowledge of the location of existing AIS populations as well as the identification of uninfected waters. In early June 2009, 7 AIS experts convened at the AMK Ranch in Grand Teton National Park to develop a draft of this AIS inventory and monitoring framework for the GYA. Participating visitors were Dave Britton (USFWS), Robert Hall (University of Wyoming), Sunil Kumar (Colorado State University), Robert McMahon (The University of Texas at Arlington), Sarah Spaulding (USGS), Mark Sytsma (Portland State University), and Erin Williams (USFWS). GYA AIS Cooperative members James Capurso (USFS), Aïda Farag (USGS), and Susan O'Ney (NPS) also contributed to this effort.

The intent of this framework is to provide entities within the GYA AIS Cooperative a uniform approach to inventory and monitoring for aquatic invasive species that includes focus species, appropriate methodologies, and a prioritization scheme. It was not our intent to re-invent an inventory and monitoring program, but to evaluate existing efforts, incorporate current programs, and supplement with needed surveys. That is why experts involved in the creation of other AIS Plans on larger scales were invited to participate in the creation of this framework. At the time of its development, most AIS monitoring plans were species or genus specific. Our intent was to develop a holistic approach that would document a diversity of existing plant and animal AIS that threaten the GYA while maintaining sensitivity to the possibility of encountering newly discovered AIS species.

This AIS inventory and monitoring framework for the GYA was reviewed and approved by the GYA AIS Cooperative. It is not intended to eclipse the authorities, priorities, or prerogatives of entities that participate in the GYA AIS Cooperative. Rather, its intent is to provide the group, as a whole, methods and priorities for action. This will aid the Cooperative in prioritizing the application of their collective but limited inventory and monitoring resources.

The expansion of non-native aquatic species into new habitats is so great in extent as to be termed an "ecological revolution" (Lodge et al. 2006). At the same time that this "revolution" is underway, the earth is undergoing anthropomorphic ecological changes. Direct and indirect impacts to aquatic and riparian habitats, including changes in temperature, precipitation, surface flow, and water quality, are the very factors that are likely to alter the distribution, abundance, and rate of new introductions of aquatic invasive species (Kolar and Lodge, 2000; Lee et al. 2008; Rahel and Olden, 2008). For example, the annual mean water temperature in Yellowstone Lake has increased by 1.0° C over the past thirty years (Tronstad 2008). In the western USA, climatic warming is influencing the proportion of snow and rain in precipitation (Knowles et al. 2006) and potentially the season available for invasion by aquatic species. Aquatic organisms are sensitive to changes in temperature, and those at margins of their range will likely expand or contract in response (Hellman et al. 2008). Field inventories and monitoring are crucial to detect species shifts related to climate change and invasion dynamics.

As of the writing of this framework, AIS infestation in the GYA has been limited but where found, has been profound (e.g. lake trout effects upon the native biota of Yellowstone Lake). Currently the spread of harmful non-native mussels brings them closer to the GYA. We believe inventory and monitoring is an important element to rapid response and successful control of these types of threatening invasions. In addition, a sound inventory and monitoring framework will provide baseline knowledge that will be helpful in our understanding of the spread and impacts of invasive species upon ecosystems within the GYA. This framework may also be applicable and helpful to other regional efforts throughout the country that seek ways to effectively and holistically inventory and monitor for AIS.

CONTEXT WITHIN THE WEST

The GYA encompasses Yellowstone and Grand Teton National Parks, parts of Idaho, Montana and Wyoming, and six National Forests (including Caribou-Targhee, Beaverhead, Gallatin, Custer, Shoshone, and Bridger-Teton), and the U.S. Fish and Wildlife Service National Elk Refuge. One of the four goals in the GYA AIS Cooperative Strategic Plan centers upon creating an inventory and monitoring protocol that is uniform for the GYA and integrated with existing national reporting systems. While there have been various efforts to inventory and monitor species in the GYA, to date, there has not been comprehensive monitoring for aquatic invasive species, or baseline monitoring for all species in the GYA. The sampling or monitoring efforts that have occurred in the GYA are summarized below.

Monitoring is a focus of many AIS efforts in the western United States. However, there are no known broad-based, regularly recurring sampling programs for AIS in the western United States. Portland State University conducts extensive AIS monitoring for the Columbia River system. California initiated several baseline field surveys of ports and bays along the California coast in 2000 and 2002, expanded that baseline to include outer coast sites in 2004, and is now monitoring those sites for new introductions. Some inventory efforts are species-specific (e.g. *Dreissenid* (quagga or zebra mussels), *Spartina,* New Zealand mudsnail (*Potamopyrgus antipodarum*) and Tamarisk *(Tamarix* spp.)), while others are geographically-specific (e.g. Lake Tahoe or Sacramento-San Joaquin Delta) and many are only one or two-year projects.

100th Meridian Initiative boater assessments were conducted in 2008 in some waters surrounding the GYA by Lilias Jarding at the University of Wyoming. Boater assessments are conducted annually in Montana, the upper Snake River in Wyoming, Henrys Lake in Idaho, and Grand Teton and Yellowstone National Parks. Boater assessments are an effort to gather information at boat ramps as to where recreational boats last launched, and where they plan to launch next. This information can provide vital information about potential pathways for AIS that are moved from one waterbody to another via boats that have not been decontaminated or dried, which can assist an administrator in management actions to reduce the potential of an introduction. For example, if a boat that was last used in Lake Mead, which is heavily infested with quagga mussels, was about to launch in Jackson Lake, an agency administrator could require decontamination or quarantine of that boat in order to prevent the spread of species that can survive overland transport.

EXISTING GYA AIS INVENTORY AND MONITORING

Some AIS inventory and monitoring has occurred in the GYA prior to this framework. Data were collected by biologists from agencies, universities, NGOs, and private individuals. Surveys were conducted during efforts to study fish, invertebrates, plants, and disease. In some cases, these data can be used as baseline data that can be compared with future monitoring data collected at the same location. Future inventory and monitoring efforts should give consideration to past GYA surveys to use them as baseline when possible.

Fishes

State and federal agencies, universities, and organizations have been monitoring fish populations in the GYA for decades. These surveys were conducted with a variety of methods and for a variety of reasons, but seldom exclusively for determining the range of non-native fish species. However, the residual non-native fish distribution and population density data provide an insight into their occurrence and spread. For example, the Caribou-Targhee National Forest Fisheries Program has performed fish distribution surveys on most streams within the National Forest between 1997 and 2003. In their attempt to document native fish stronghold populations, non-native fish distribution was also documented. The Caribou and Targhee Forest Plans direct the Forest to return to documented stronghold streams every 10 years. Return visits have documented the invasion of non-native fish and their replacement of native fish populations in some of the native fish stronghold streams over a 10 year period. For example, the 2007 return to the 1997 Yellowstone cutthroat trout strongholds in the Sinks Drainages of the upper Snake River Plain in SE Idaho (near Dubois, ID) documented the conversion of 5 out of the 10 stronghold streams from native to non-native fish dominated systems.

Data for the distribution of non-native fish in the GYA are in various databases, reports, and files throughout agency, university, and NGO offices. Possibly the most complete collection of the data exists in interagency status assessment databases developed for each of the cutthroat trout subspecies that occur in the GYA. Rangewide interagency conservation teams have been established for Yellowstone, Colorado River, Bonneville, and Westslope cutthroat trout and they have developed distribution maps and databases for each of these subspecies. The presence and threat of non-native fish species are available from those sources.

Although aquatic habitats in the GYA are currently not as invaded as the North American Great Lakes, there are some notable invasions that have greatly altered native biota. Lake trout and whirling disease have dramatically lowered Yellowstone cutthroat trout (*Oncorhynchus clarki bouveri*) populations in Yellowstone Lake (Koel et al. 2005). The impacts of whirling disease and lake trout have extended beyond the lake to terrestrial predators of cutthroat trout (Crait and Ben-David 2007) and lowered nutrient transport to spawning streams (Tronstad 2008).

Invertebrates

Dan Gustafson, Montana State University Biology Department, has sampled waters of the GYA extensively for freshwater invertebrates. His work has generated valuable distribution maps of New Zealand mudsnail. These maps and references from his work are available at:
http://www.esg.montana.edu/aim/mollusca/nzms/status.html

New Zealand mudsnails have invaded several streams and rivers within the GYA. These snails achieve extremely high biomass and secondary production relative to native invertebrates suggesting high impact in invaded streams (Hall et al. 2006). Snail production is sufficient in Polecat Creek for the species to constitute the largest fluxes of carbon and nitrogen in the stream (Hall et al. 2003). Mudsnails compete asymmetrically with native, endemic Jackson Lake spring snails (*Pyrgulopsis robusta*). Spring snails facilitate mud snail growth, yet the presence of mud snails inhibits spring snail growth, suggesting that mud snails may competitively exclude the native snails (Riley et al. 2008). Based on the impact of the above invasions, there is evidence that the spread of these present non-native species into new habitats, or the invasion of new species into the GYA, will have effect upon populations of native invertebrates there.

Montana has been sampling for AIS statewide since 2004. While Montana does focus effort on sampling for adult and juvenile *dreissenid* mussels, they also have a broad sampling protocol that includes multiple methods in order to assess many taxa. Montana also maintains a veliger laboratory that utilizes cross-polarized light microscopy to look for d*reissenid* veligers in samples from Montana, North Dakota, South Dakota, Iowa, Kansas, Nebraska, and Missouri. Mussel survey reports from Montana are available on the internet at: http://mtnhp.org/Reports.asp?key=1

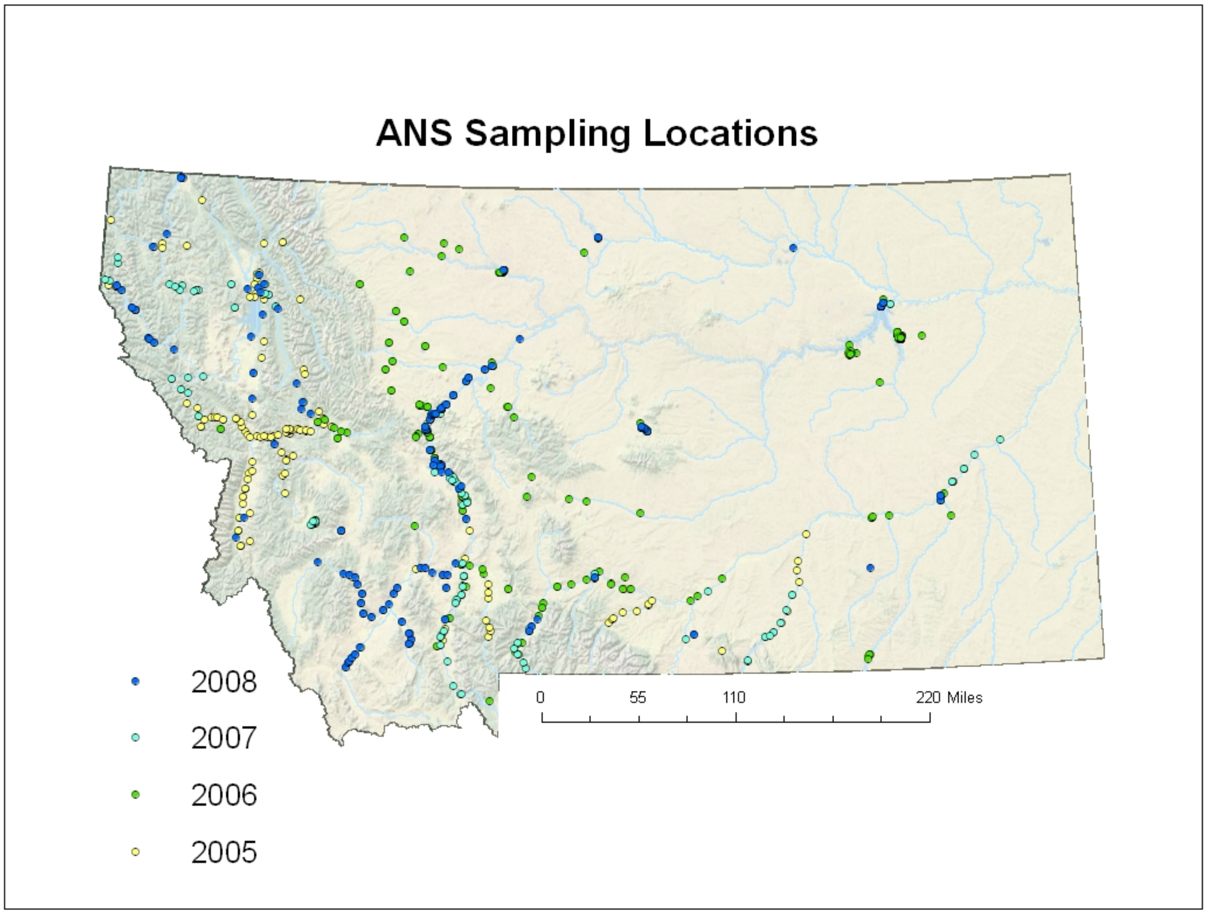

Figure 1. AIS Sampling Locations in Montana conducted by MTFWP.

Wyoming will start a substrate sampling program for mussels in 2009. Idaho also has a monitoring program that focuses primarily on high-risk waterbodies for d*reissenid* mussel introduction. The Bureau of Reclamation will begin sampling Jackson Lake, Fontenelle, Boysen, Big Horn, and Buffalo Bill reservoirs in 2009 in the GYA. These waters will be sampled monthly with plankton tows and with one substrate sampler for d*reissenid* mussels. State Departments of Environmental Quality have sampled extensively throughout the GYA per their Beneficial Use Reconnaissance Project (BURP) Surveys. The BURP surveys collect water quality, benthic macroinvertebrate, and fish distribution data.

Plants

As part of their terrestrial weed program, some counties in the GYA, including Bonneville and Bear Lake counties in Idaho and Teton County in Wyoming, have documented and treated riparian AIS such as salt cedar and purple loosestrife.

Diatoms

The diatom didymo (*Didymosphenia geminate)* is emerging as an organism with an extraordinary capacity to impact stream ecosystems on a global scale. In recent years, streams in New Zealand, North America, Europe, and Asia have been colonized by unprecedented masses of "didymo" and its extracellular stalks. This diatom is able to dominate stream surfaces by covering up to 100% of substrate with thicknesses of greater than 20 cm, greatly altering physical and biological conditions within streams. Although it is considered native to high latitude and high elevation sites in North America, this species is believed to be expanding its geographic range. Two other species of concern, *Cymbella mexicana* and *C. janischii*, historically inhabit western streams and rivers (Patrick & Reimer 1966, Wellnitz et al. 1996, Bahls 2004). The typical growth habit of all these species includes the episodic formation of large masses (Cleve 1894-1896, Skvortzow 1935, Skulberg 1982) and the rate of reported nuisance blooms has increased.

In 2008, a nuisance bloom of the diatom didymo was present in Lake Creek, from the outlet of Phelps Lake to approximately 1 km downstream of the Rockefeller Preserve in GTNP. This bloom was considered "excessive" because the coverage of the stream substrate was 70% or above for greater than 1 km. At these sites the total amount of periphyton biomass was up to six-fold the biomass found at sites without didymo. In addition to Lake Creek, Kaufmann and Taggart creeks had visible masses of didymo, but the stream extent and coverage was not quantified. The diatom was also confirmed in Fish Creek by microscopic examination but no visible masses were documented during the observation period.

Although there have been attempts to relate the occurrence of didymo to specific water chemistry and geologic influence, chemical and physical factors seem to represent only a portion of the control on growth and distribution (Lindstrom 1991, Sherbot & Bothwell 1993, Jónsson et al. 2000, Kilroy 2004). Therefore, it is difficult to state whether water chemistry is controlling the formation of the blooms in GTNP. However, there is indication that these species may be favored under some conditions of increased human impact. Types of human influence that are associated with *D. geminata* blooms include increased nutrient concentration (Kara and Şahin 2001, Kawecka and Sanecki 2003, Noga 2003, Subakov-Simić and Cvijan 2004), stable flow below impoundments (Dufford et al. 1987, Holderman and Hardy 2004, Shelby 2006), low flows (Kilroy et al. 2005a), and spread by aquatic recreationalists (Kilroy et al. 2005b, Kilroy et al. 2006, Larned et al. 2006). In particular, recent blooms on the east coast of the USA in sites with heavy fishing pressure point to spread by humans. The relationship, however, has not been fully documented (Bothwell 2008, personal communication).

Didymo is able to survive out of water, and it may be transported on the gear of recreationalists (Kilroy et al. 2005b). In Grand Teton National Park, this diatom was found in a high visitor use area, and there is the potential for the species to spread by anglers to other sites within the national park and other public and private lands. Although there are several factors that appear to influence its distribution, recent nuisance blooms of this species suggest popular angling sites are often sites of nuisance blooms. Decontamination of aquatic gear by recreationalists may be appropriate as a management response within the national park system and adjacent public and private water bodies.

Disease

In December 1994, whirling disease (WD), caused by the Myxobolus cerebralis parasite, was confirmed in Montana's Madison River. The disease was believed to be the cause of a substantial decline in the Madison's rainbow trout populations. This discovery resulted in increased sampling and monitoring of rivers and streams and continued inspection of private, federal, and state hatcheries by fish and game agencies in the GYA. In addition, the Whirling Disease Steering Committee of the National Partnership for the Management of Wild and Native Cold Water Fisheries which was run through the Water Center at Montana State University began a competitive grant program offering funding provided by the US Fish and Wildlife Service for whirling disease research projects.

In most GYA waters, whirling disease has not caused the severe population impacts seen in Montana's Madison River. However, WD continues to spread within the GYA and remains a serious fish health issue. Montana Fish, Wildlife, and Parks maintains an extensive sentinel cage monitoring program placing approximately 110 cages in Montana's rivers and streams. MTFWP annually inspects all state, federal, and private hatcheries, and no WD has been detected in Montana's hatcheries. Dr. Billie Kerans, researcher at Montana State University in Bozeman, has received an NSF grant for continuing WD research on the Madison River, and MTFWP biologist Ron Pierce is doing research in the Blackfoot River on the impact of WD on Bull trout and Mountain whitefish. In Yellowstone National Park, while there is currently no whirling disease monitoring program or ongoing whirling disease research, funds are being sought by YNP to do additional WD monitoring in Yellowstone Lake and its tributaries. In Wyoming, the Game and Fish Department has not noted any population declines in WY trout populations due to whirling disease. The department does annual hatchery inspections and has had problems with WD in two hatcheries. As a result of an appropriation provided by the Wyoming legislature, Wyoming Fish and Game has invested nearly $20 million in hatchery improvements which includes protection of hatchery water sources and converting hatcheries from high risk surface water sources to ground water. Idaho Fish and Game has no active WD projects in the Yellowstone/Upper Snake River area at this time. IDFG did a distribution/prevalence/intensity project on the Teton River in 2003-04 using sentinel fish in live-boxes and a less intensive survey on the lower Bear River. Because the USFWS has reported positive findings in the upper Bear River, IDFG plans to do another live-box trial in that area next year. The only new confirmation made in the GYA was from the Falls River in 2007. IDFG does extensive WD inspections on its own hatchery populations, but does only limited work with the private sector. Additionally, they also do extensive sampling of all anadromous and feral brood stocks. IDFG has made two significant hatchery modifications over the years due to the microscopic parasite *Myxobolus cerebralis* (MC) presence in the water sources. The first was at Hayspur Hatchery, a rainbow trout production and egg-take station. Here IDFG drilled wells and built covered concrete ponds for brood fish containment, and eliminated nearly all on-station fish production for stocking. Hayspur is now an egg-take facility and catchable-trout redistribution station. The other modified hatchery occurred at the Pahsimeroi Hatchery. Historically, Chinook salmon were reared in Pahsimeroi River water which was found to be highly infective for WD in the early 1990s. The first production modification involved shipping all eggs taken on site to another hatchery for early rearing, but the fish were returned at about 3 inches because the other facility did not have room or water to hold them longer. The fish were exposed and infected later in life so that clinical signs were reduced. Idaho Power has constructed new incubation and rearing facilities with enough well water to keep the fish from exposure much longer and not encroach on the capacities of other hatcheries.

Existing More Holistic Monitoring

Some AIS surveys in the GYA were more inclusive than others. Some surveys such as the Portland State University AIS survey, the Montana survey, the Environmental Protection Agency Environmental Monitoring and Assessment Program (EMAP), and USGS National Water Quality Assessment (NAWQA) include several AIS plant and animal species in the same survey and database.

Portland State University conducted surveys in six lakes and at 22 sites in rivers and streams in the GYA in 2008 (Sytsma and Howard 2008). Lake sampling included characterization of aquatic macrophytes and benthic invertebrates in lakes and streams and collection of dreissenid mussel veliger samples in lakes. Riparian weeds were noted when encountered during lake and stream sampling. Surveys occurred from mid-July through mid-August. Target species included New Zealand mud snail, zebra and quagga mussels, rusty crayfish (*Orconectes rusticus*), and Asian clam (*Corbicula fluminea*), Eurasian watermilfoil (*Myriophyllum spicatum*), curly-leaf pondweed (*Potamogeton crispus*), water chestnut (*Trapa natans*), water thyme (*Hydrilla verticillata*), parrotfeather (*Myriophyllum aquaticum*), Brazilian elodea (*Egeria densa*), purple loosestrife (*Lythrum salicaria*), saltcedar (*Tamarix* spp.), perennial pepperweed (*Lepidium latifolium*), and (*Euphorbia esula*). None of the target species were detected. Additional leafy spurge sampling is planned for 2009 in Fremont Lake and the upper Green River.

Ask Eileen if she could insert a paragraph summarizing the State of Montana's holistic survey effort, including sample locations. Her presentation at the crew training in Jackson had summary info that could be valuable here. THIS HAS BEEN REQUESTED OF EILEEN.

Over 1500 samples on 1340 perennial streams in the western US were collected in 2000-2004 by the EPA EMAP Program (Stoddard et al. 2005b). The sites were randomly chosen to be representative of flowing waters in the West, or selected as reference sites, or "best possible condition". The EMAP survey design uses a probabilistic sample design which allows estimates of population abundance over particular geographic regions with a known confidence level. Sites were sampled by crews trained to use identical sampling methods to facilitate comparisons across the region, and all of the data were subject to strict quality assurance procedures. Samples were collected and analyzed for water chemistry, physical habitat parameters, fish, benthic macroinvertebrates and periphyton, with inclusion of invasive and non-native species (Stoddard et al. 2005b). In particular, the program provided measures of the total stream kilometers impacted by non-native vertebrates, non-native crayfish, and Asian clams. The EMAP survey included approximately 60 sites in the GYA (Figure 2), and data are available from the EPA Corvallis Laboratory http://oaspub.epa.gov/emap/webdev_emap.search Additional surveys of lakes (2008 National Lakes Survey, NLS, www.epa.gov/owow/lakes/lakessurvey/) and streams (2009 National Stream and River Assessment, NSRA, www.epa.gov/owow/streamsurvey/) are in progress.

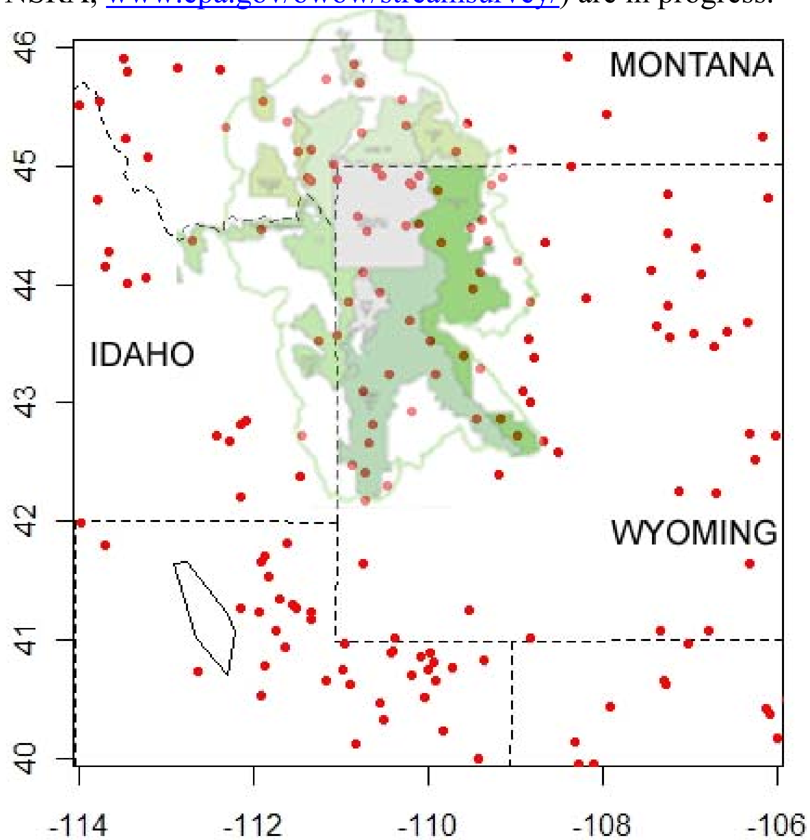

Figure 2. 2000-2004 Western EMAP survey sites within parts of Montana, Idaho and Wyoming. The GYA is shown in colored outline.

The USGS NAWQA Program includes two watersheds that fall within the GYA; the Upper Snake River Basin and the Yellowstone River Basin (Figure 3). The objectives of the NAWQA Program are to measure water quality, determine change in water quality over time, and understand human impacts on water quality. The NAWQA survey design is based on establishment of fixed sites in selected watersheds that are sampled over time. This type of design is appropriate for detecting change over short temporal periods and long-term trends at representative sites. Fixed sites have been sampled from 1998 for fish, benthic macroinvertebrates, and periphyton, as well as water quality parameters. Data are publically available and will be beneficial to not only document past species presence, but to leverage the current effort in the GYA.

Figure 3. Map of the Yellowstone River Basin, a watershed that has been the site for long-term surveying by the USGS NAWQA program.

FOCAL SPECIES

Although the list of potential aquatic invasive species is long, there are some clear threats to aquatic systems in the GYA. These focal species are known to be present in the West or in regions that can serve as sources of propagules to the GYA (Table 1). Focal species can be introduced to the GYA by a variety of vectors such as trailered boats, anglers, and aquarium hobbyists. The vector strength at a particular water body may be used to prioritize detection of focal species.

Table 1. Focal Species, habitat types, and general comments

		Lake/pond benthic	Lake/pond pelagic	River	Stream	Wetlands	Comments
Snails							
	Cipangopaludina chinensis (chinese mystery snail)	x				x	
	Potamopyrgus antipodarum (New Zealand mudsnail)	x		x	x	x	
	Melanoides tuberculata	x			x		stenothermal areas
Bivalves (these species have benthic and pelagic lifestages)							
	Corbicula fluminea (asian clam)	x		x	x		
	Dreissena polymorpha (sebra mussel)	x	x	x		x	Ca requirement >10 mg/L and
	Dreissena bugensis (quagga mussel)	x	x	x		x	rivers downstream from lentic water bodies
Benthic Invertebrates							
	Procambarus clarkii (red swamp crayfish)	x		x	x	x	

	Gammarus fasciatus (scud)	x		x	x	x	
	Orconectes rusticus (rusty cryafish)	x		x	x	x	
	leeches (sp?)	x		x	x	x	
Planktonic crustaceans							
	Bythotrephes cederstroemis (spiny waterflea)		x				
	Cercopagis pengoi (fishhook waterflea)		x				
	Daphnia lumholtzi		x				
Planktonic algae							
	Cylindrospermopsis raciborskii		x				
	Prymnesium parvum (golden algae)		x				high salinity water
Periphyton							
	Didymosphenia geminata (didymo)	x		x	x		no nuisance blooms in lakes and ponds
	Cymbella mexicana	x		x	x		no nuisance blooms in lakes and ponds
	Cymbella janischii	x		x	x		no nuisance blooms in lakes and ponds
Aquatic macrophytes							
	Myriophyllum spicatum (eurasian watermilfoil)	x				x	rooted submersed
	Potomogeton crispus (curly leaf pondweed)	x		x		x	rooted submersed
	Trapa natans (water chestnut)	x				x	floating rosettes
	Hydrilla verticillata (water thyme)	x		x		x	rooted submersed
	Egeria densa (brazilian egeria)	x		x		x	rooted submersed
	Myriophyllum aquaticum (parrotfeather)	x		x		x	shallow water emergent
	Butomus umbellatus (flowering rush)	x		x		x	
Pathogens							
	Viral Hemorrhagic Septicemia (VHS)	x	x	x	x	x	
	Myxobolus cerebralis (whirling disease)	x	x	x	x	x	
	Batrachochytrium dendrobatidis (Bd)	x	x	x	x	x	
Riparian plants							
	Lythrum salicaria (purple loosestrife)	x	x	x	x	x	
	Tamarix sp. (saltcedar)	x	x	x	x	x	
	Lepidium latifolia (perrenial pepperweed)	x	x	x	x	x	
	Euphorbia esula (leafy spurge)	x	x	x	x	x	
	Phalaris arundinacea (reed canary grass)	x	x	x	x	x	
	Phragmites australis sub. Australis (reed)	x	x	x	x	x	
	Elaeagnus angustifolia russian olive)	x	x	x	x	x	
Amphibians, reptiles, mammals							
	Myocastor coypus (nutria)	x		x		x	
	Rana catesbeiana (bullfrog)	x		x	x	x	slow-moving streams
	Macrochelys temminckii (snapping turtle)	x		x	x	x	
Fish							
	Salvelinus fontinalis (brook trout)		x	x	x		
	Oncorhynchus mykiss (rainbow trout)		x	x	x		
	Salvelinus namaycush (lake trout)		x	x	x		
	Rhinogobius brunneus (amur goby)	x	x	x	x	x	
	Neogobius melanostomus (round goby)	x	x	x	x		
	Sander vitreus (walleye)		x	x	x		
	Esox lucius (pike)		x	x	x		
	Micropterus salmoides (largemouth bass)		x	x	x		

	Micropterus dolomieui (smallmouth bass)		x	x	x		
	Perca flavescens (perch)		x	x	x		
	Aquarium		x	x	x		thermal areas
	Cyprinids (minnows as baitfish)		x	x	x	x	
	Lota lota (burbot)	x	x	x	x	x	

SAMPLING PROGRAM DESIGN

Biotic monitoring serves three purposes. 1) detect the presence of invasive species with a high degree of certainty, 2) monitor the changing population size of an established invasive species through time, because the long-term effects of invaders are likely quite different than those immediately following establishment (Strayer et al. 2006), and 3) detect a response by native biota, which under the best circumstances requires data prior to invasion of unwanted species. Thus biotic monitoring surveys should include quantitative estimates of native biota as well as systematic searching for alien species. Given the expense of field campaigns, quantitative collections of native biota will provide much more potential information for little added cost, though we do note that processing these samples can be expensive. If resources to count and identify organisms in pre-invasion samples do not exist, then it is important to archive these samples in case the ecosystem is invaded.

Aquatic sampling methods vary depending on species and habitat sampled and will be described in greater detail below. However, there are commonalities that should be incorporated into any sampling program for indigenous and/or non-indigenous aquatic species. When sampling for invasive species a sampling method must be designed to detect rare as well as common species. Ability to detect rare species at low densities is particularly important for early detection of an invasive species which is likely to occur in very low densities and limited distribution in the initial stages of invasion. A sampling program's ability to detect the early establishment of an invasive species is a key component in the development of a successful early detection, rapid response, and eradication programs.

In order to increase the probability of detecting an aquatic invasive species in the early, and possibly, controllable stages of its introduction, a sampling design should emphasize where dispersal vectors are likely to introduce an invasive species. Such areas where known human-aided dispersal vectors are likely to introduce an invasive species include boat launch sites, intensely fished areas, and recreation sites receiving high human traffic, among others. A boater assessment program can be used to determine which launch sites receive the most trailered boat traffic as well as water bodies on which boats were launched prior to entering the GYA. Such information allows assessing the potential for trailered boats to act as vectors for introduction of invasive species to GYA aquatic habitats. It would be efficacious to collect information in a GYA boater survey program similar to that collected in the 100th Meridian Boater Assessment allowing analyses of GYA data in relation to boater movements across the western United States. In addition, creel surveys of wading anglers, gathering information similar to that of boater surveys, could be used to assess the likelihood and probable source habitats for the introduction of aquatic invasive species by this vector and the potential dispersal within the GYA, such as the New Zealand mud snail and didymo. While sites considered likely to receive and support establishment of invasive species should be included and perhaps emphasized in a sampling program, it also important to sample sites considered less susceptible to non-native species invasion. This is because the points of introduction and successful establishment of an invasive species cannot be predicted with 100% certainty. Sampling of such sites is also critical to assess the spread and ecological impacts of an invasive species after it has been introduced and established.

Integrating Invasive Species Life Histories/Phenologies

The efficiency of a sampling program in the early detection of an invasive species can also be improved by integrating the time and frequency of sampling efforts with the life histories/phenologies of the invasive species of concern. This allows a sampling program to be conducted at times and in areas where the probability of detection of an invasive species is maximized. For example, plankton samples for the presence of dreissenid mussel (*Dreissena polymorpha*, zebra mussel and *D. rostriformis bugensis*, quagga mussel) veliger larvae or recently released juveniles of Asian clams are best conducted after surface ambient water temperatures rise above 16-18ºC where reproduction rates in these species are maximized. This increases the probability of detecting veligers or juveniles in the water column. Sampling at lower ambient water temperatures may result in a lack of detection of the planktonic stages of these damaging invasive bivalves. Similarly, the microhabitats of the adults of these bivalves also differ and, thus, require different sampling strategies. Adult dreissenid mussels are most likely to be found byssally attached to hard substrata such as rocks, submerged wood, and other hard-surfaced debris whereas Asian clams burrow in soft sandy and sand/gravel substrata. Thus, the sampling techniques for the adults of these bivalves would differ with inspection, and scraping of individuals from hard surfaces by divers and/or deployment of hard-surfaced settlement plates being most likely to detect the presence of adult dreissenid mussels while adult Asian clams are best detected with soft substrata sampling devices such as grabs and dredges. If only one type of substratum or sampling methodology was utilized in a water body, the early establishment of one of these two groups of invasive bivalves could go undetected.

Statistical Analyses of the Detection Power

As described above, a sampling program must be designed to detect invasive species at low density in the early stages of its invasion to maximize the probability for successful rapid response and eradication. If a sampling design has a relatively low probability of detecting a rare species, it is unlikely to detect individuals of a newly introduced invasive species early enough to allow successful rapid response and eradication to occur. Appropriate methods for statistical power analyses should be applied to determine the number of samples taken at any one sampling site and the number and distribution of sampling sites within the lentic and lotic waters of the GYA. Statistical power analyses would assure a minimum number of samples required to achieve an acceptable probability of detecting an invasive species in the early stages of its colonization. If a sampling design has a relatively low probability of detecting a rare, low density species, it is unlikely to detect individuals of a newly introduced invasive species early enough to allow successful rapid response and eradication. A justified method for statistically evaluating the power of a sampling design to detect a non-indigenous species early in the invasion process should be applied to any sampling procedures adopted in the GYA Invasive Species Inventory and Monitoring Framework.

Qualitative Versus Quantitative Sampling

Sample design can be of two general types. Qualitative sampling identifies the presence of species in a sample without estimating their relative abundance or densities. Quantitative sampling measures the relative abundance of species in samples by counting numbers of individuals of species collected in a specific unit area or volume of habitat allowing estimation of species' densities and dominance. Further analyses of quantitative samples could involve measuring species' biomasses or production per unit area or volume. Quantitative samples involve more handling time and expense to process samples than qualitative samples, but quantitative samples yield much more information on the community structure of the sampled habitat and can provide much richer baseline data with which to evaluate the ecological impacts of an invasive species introduction. Both types of sampling designs can be integrated into an overall GYA program. Regardless of the sampling procedures adopted, they should be maintained through time so that changes in the occurrence and relative densities of both indigenous and invasive species within the waters of the GYA can be directly compared over time. This would allow both the natural variation within indigenous communities and the impacts of invasive species on those communities to be assessed.

Other Variables

In addition to documenting species presence, relative abundance, density, and biomass, other relative data should be recorded for each sample site as it strengthens the database and increases the ability to assess the likelihood of invasion (see Green 1979 for a review of environmental sampling methods and analyses). Among these would be the geographical coordinates of the sample site determined with a GPS unit. GPS coordinates define the exact location of a sample site so that it can be re-sampled in the future. Known sample coordinates also allow sample data to be overlaid on maps for GIS analysis of the sampling data. Physical parameters, such as temperature, calcium concentration, hardness, alkalinity, pH, nitrate and phosphate concentrations, turbidity, chlorophyll *a* concentration, flow velocity, and substrate type (among others as appropriate) should be recorded for each sample site and during each sample period. It is of particular importance to deploy temperature data loggers in key water bodies and sample sites in order to determine their annual temperature profiles. Temperature loggers are relatively inexpensive but record temperature data over long periods of time. Temperature profiles are valuable to predict the susceptibility of aquatic habitats to invasive species that are unlikely to become established in habitats with an annual temperature range that falls outside of their tolerated incipient upper or lower temperature ranges.

It is also important to record other relative data for a sample site such as degree of natural and/or anthropomorphic disturbance and level and type of human access and use. Such data enrich the sampling database and are important to estimate the likelihood for the introduction of invasive species, their potential for dispersal to other areas in the GYA, the potential impacts of introduction, and provide a baseline with which to determine actual ecological consequences if introduction occurs.

Spatial Distribution of Sample Site Locations

Effective sample collection needs to be driven by a survey design that is most likely to detect introductions and establishment of non-native species, include GYA native aquatic species in inventory lists, and monitor populations for change in abundance. The spatial distribution and temporal frequency of sample collection needs to be appropriate for the types of organisms to be monitored and the habitats of greatest vulnerability and concern. In order to inventory and monitor for the detection of non-natives, we recommend a tiered survey design based on habitat type and sites of likely invasion throughout the GYA.

Sites of primary importance for the detection of AIS are those targeted as most likely to be invaded by organisms of highest potential impact to GYA habitats. These sites are areas that receive the most human use and/or highest impact by known vectors. For example, some warm springs have already been infested with NZ mud snail and red-rimmed melania (*Melanoides tuberculata*). Several boat docks on Jackson and Yellowstone lakes are vulnerable to introduction of zebra and quagga mussels by large boats arriving from invested sites such as Lake Mead and the Great Lakes. These high-use sites should be sampled annually with repeated sampling at the same sites over time.

The National Park Service, Inventory and Monitoring Program has implemented a couple probabilistic sampling designs in the Greater Yellowstone Ecosystem and could assist in the development of a similar effort for AIS sampling in the GYA. Contact Inventory and Monitoring Program Manager Cathie Jean for assistance at Cathie_jean@nps.gov or 406-994-7530.

Inventory and Estimate Abundance of Native Aquatic Species

Detection of aquatic species and estimates of population abundance with a defined confidence level would be most appropriate using a probabilistic sample design over the GYA. A stratified survey design that includes sites outside of fixed annual sites, would be based on habitat types such as streams/rivers, lakes, and wetlands/ponds. For example, within this design concept, approximately 20 streams and rivers, 20 lakes, and 20 wetlands and ponds would be included in the probability design to sample each year. The sites would be submitted to selection through a probability design, so that each site has a known probability of being selected for sampling within a given year. That is, each year, a unique set of streams, lakes and wetlands would be sampled. As a whole, the sites within each habitat type would be statistically representative of the aquatic resources within the GYA. Tony Olsen, at EPA Corvallis, supports development of survey designs using this approach (Olsen 2008, personal communication).

BIOLOGICAL SAMPLING OF VARIOUS HABITAT GROUPS

Lake/Pond Benthos

Lake and pond benthos can be sampled using bottom dredges and cores. The efficacy of these samplers depends upon sediment characteristics. Rocky, large-cobbled bottoms cannot be effectively sampled using these devices. Sediment samples collected with cores may be sectioned to examine distribution of organisms with depth. Benthic organisms can be separated from the sediment by vigorously agitating mud, sand, gravel and rock samples in water to suspend organic material and small invertebrates, then elutriating the suspension through a series of mesh sieves (2-mm, 1-mm mesh, and 0.5-mm) to retain suspended organisms. The washing and decanting procedure should be repeated until the majority of organisms in the samples are removed. Sub-samples should be made only when the total volume of organisms retained on the sieves exceeded the volume of the largest sample containers. Abundance of individual species should be expressed as density (# of individuals/m^2). The area sampled by the dredge or core and a general description of sediment texture (e.g., sand, silt, gravel) should be included in the metadata for each sample. Species such as crayfish can be sampled using baited traps. Abundance data for sampling using this method should be expressed as catch per unit effort. Metadata should include period of trap deployment, sediment type, and presence of macrophytes. Invertebrate samples should be preserved in at least 70% EtOH.

Aquatic mammals, amphibians, and reptiles can be monitored opportunistically. Sampling crews should document the presence of all species encountered during a sampling event. Documentation should include species abundance and location. Adaptive monitoring should be implemented upon confirmation of a focal species, which may involve development of species-specific sample protocols.

Lake/Pond Pelagic

Lake and pond pelagic habitats should be sampled using plankton nets. Many sizes and types of nets are available, and most can be used effectively to collect plankton samples. Net mesh determines the organisms that are collected. Generally, a mesh size of <53 µm should be used for zooplankton collection. A 20-µm net should be used for phytoplankton collection. Tows should be made through the entire water column in unstratified lakes. In stratified lakes with orthograde dissolved oxygen profiles, tows should also be made through the entire water column. In stratified lakes with clinograde dissolved oxygen profiles the tows should be made from the top of the metalimnion to the surface. Samples for zooplankton identification should be preserved in at least 70% EtOH. Phytoplankton samples should be preserved with Lugol's solution. Phytoplankton and zooplankton should be enumerated by counting organisms in a subsample. Counting should continue until a minimum of 400 organisms are counted. Dreissenid mussel veligers can be counted using polarized-light microscopy by a trained analyst, using a Flow-cam, or detected by PCR. All veliger detections should be confirmed by a second laboratory. Metadata collected with plankton tows should include mesh size, the diameter of the net opening, length of tow, and depth at beginning and end of the tow.

Pelagic fish can be collected using a variety of nets and traps. Random panel and graduated panel sinking gill nets with mesh sizes of 19, 25, 38, 51 57, 64, 76, 89, and 102 mm can be used in waters like Jackson Lake (Harper et al. 2007). Nets can be set overnight at 17 locations in the lake perpendicular to the shoreline at depths ranging from approximately 5-30 meters. The total length and weight of all fish species captured in gill nets should be recorded. Fike nets and smaller-mesh gill nets, baited minnow traps, and trotlines may be required to collect benthic fish like gobies (Diana et al. 2006).

Lake/Pond Plant Methods

Submersed plants can be collected using a rake sampler tossed from a boat. A thatching rake head attached to a rope provides an ideal sampler for most plants, although some small plants may be under-sampled using this method. A minimum of thirty samples at three sites within a lake should be sampled at depths ranging from shore to the maximum depth of colonization. Optimally a minimum of 100 samples per lake should be sampled. Sampling should occur randomly in areas likely to contain aquatic plant stands (shallow, littoral areas) and in areas near likely sites of introduction of invasive species (marinas). All the plants attached to the rake upon retrieval should be identified to the lowest taxon possible, voucher specimens collected, and relative abundance recorded (rare, common, abundant). Some species, e.g. water milfoil (*Myriophyllum* species), are difficult to identify morphologically. Molecular identification methods may be required for those species. A bottom viewer should be used to scan for plants not collected in rake samples. Emergent and riparian plants can be observed by boat, walking the shoreline, and/or use of binoculars. Unrooted plant fragments found anywhere within the lake should also be collected and identified. Sampling should be conducted annually in water bodies with frequent trailered boat use.

Wetland/Riparian Plant Methods

Riparian plants can be surveyed by observing the shoreline from a boat or on land. Ideally the entire shoreline of a lake or pond should be surveyed. Small wetlands can be surveyed by walking transects through the wetland and recording species presence and cover. Cover can be estimated using random quadrats or line-intersect methods. Large wetlands may be most efficiently surveyed aerially by fixed-wing or helicopter. Aerial surveys for some species may be most effectively conducted during flowering, particularly for species like purple loosestrife.

Sampling for Diseases

Batrachochytrium dendrobatidis (Bd) is collected from amphibians by swabbing the ventral side and toes of animals with a sterile cotton swab to collect epidermal tissue. Samples should be preserved with 95% ETOH in vials labeled with location, species, and lifestage. All samples should be examined using PCR assay (Boyle et al. 2004). Skerratt et al. (2008) recommended sampling 60 individuals per population and a maximum of 15 individuals per site to achieve 95% certainty of detecting one positive frog when the disease prevalence is ≥5%. Each life stage and gender should be sampled (e.g. larvae, juvenile, adult male, adult female).

The American Fisheries Society (AFS) Fish Health Section publishes the procedures and guidelines used to verify the disease status of finfish and shellfish. Their official publication "*Suggested Procedures for the Detection and Identification of Certain Finfish and Shellfish Pathogens*" is referred to as the "Blue Book" and is provided electronically at http://www.fisheries.org/units/fhs/BlueBook_access.html. The Blue Book provides specific recommendations for screening samples for the presence of viral hemorrhagic septicemia (VHS) and whirling disease and, when screening results are positive, confirming the presence of VHS and whirling disease.

To detect VHS, a more robust sample protocol may be required if the effort is funded through USDA-APHIS. USDA-APHIS published an interim rule, effective November 10, 2008, regulating live fish importation and interstate transport to prevent the spread of VHS. For stocking, importation or interstate shipment, methods should follow procedures recommended in the APHIS order for VHS. However, field sample designes for VHS presence/absence may not need to meet the same standards, rigor or scientific certainty as certification for stocking, importation or interstate shipment. If one wanted to screen many water bodies or fish populations to assess whether they remain VHS-free, testing methods which are more rapid than established cell culture methods or included fewer fish could be implemented.

Whirling disease is caused by a microscopic parasite called *Myxobolus cerebralis,* which affects fish in the trout and salmon family. Detection of whirling disease may be difficult because the life cycle of the parasite includes two hosts: salmonids and the aquatic oligochaete worm, *Tubifex tubifex.* Rainbow trout are a highly susceptible species. Brook trout and cutthroat trout are moderately susceptible; brown and bull trout are partially resistant. Conflicting data are present for lake trout but in general, these species may be considered resistant or partially resistant to *M. cerebralis.* Fish that have a high degree of resistance should not be selected for sampling unless they are the only species present. Therefore, a two-pronged program consisting of sampling resident trout populations, and exposing 'sentinel' fry to waters for a prerequisite period and testing for infection is recommended.

Stream Monitoring Methods

Given the two goals of attempting to detect and monitor invasive species with concomitant assessment of native populations, stream monitoring programs should include both systematic searches combined with quantitative sampling.

Stream Benthic Invertebrates

Surveys for benthic invasive invertebrates should begin with systematic searches. Large invertebrates (e.g. crayfish) can be detected by disturbing and dip netting suitable habitats. Smaller invertebrates such as snails, can be searched by scrubbing substrate and visually detecting live animals in the field. It is important to use a pre-defined search time that is long enough to have a high probability of detection (see Power analyses section above). These times will strongly depend on the habitat and the organism searched for and should be determined by the investigators.

In addition to searches, benthic invasive and native invertebrates should be quantitatively sampled using standard samplers, such as Hess samplers (Hauer and Resh 2006). Samplers will depend on habitat type. For example invertebrates in rocky substrates are quantitatively sampled with 6-10 Hess samples (500-µm mesh) for each site. Samples are not composited, but kept separate to estimate uncertainty in population estimates. Soft substrates are best sampled with stovepipe cores where all material is removed from this sample and contents are sieved in the field. Samples are preserved in the field and invertebrates are identified and counted in the laboratory following collection(Hauer and Resh 2006). Organisms should be identified to taxonomic levels that balance understanding of impacts with the reality of processing hundreds of samples. For example, invasive species, macrocrustaceans, and known threatened taxa (e.g. Jackson Lake spring snail) should be identified to species. On the other hand, species such as naiadid oligochaetes and chironomids that are difficult to identify may be identified at family levels. Even if there are not resources to count benthic invertebrate samples from uninvaded habitats, these samples should be archived to provide reference data in the event of future species invasions.

To detect nuisance and invasive algae such as Didymo, standard collections for periphyton are first made to detect microscopic quantities of cells. Because Didymo may grow without forming blooms, a number of substrates (i.e. five rocks of known planar surface area) can be collected and scrubbed to remove the surface biofilm. Second, systematic visual searches are made for macroscopic growths in areas of potential introduction by vectors (Kilroy et al. 2007)(refer to power section). If mats are detected, the spatial extent of mats can be quantified by a transect method, such as the Kilroy Visual Index (Larned et al. 2006). To estimate cell density and mat biomass, material is removed from known surface areas and processed following Larned et al. (2006). Subsamples of the resulting slurry should be preserved and scanned for large invasive algal cells and potentially native algae to detect changes following potential invasion. Further subsample should be used for analysis of chlorophyll a and organic matter (as ash-free dry mass) stocks, which provide measures of biomass and ecosystem change.

PHYSICAL PARAMETER MONITORING

Along with gathering information about presence/absence of invasive species, it is important to monitor other physical and chemical attributes of aquatic ecosystems for three reasons:

1. To estimate the impact of invaders beyond changes to one level of biological assemblages. For example, water clarity increases following dreissenid invasion (Caraco et al. 1997), and some dominant invaders can alter the nutrient concentrations and processing rates (Hall et al. 2003). Large changes to physical attributes can alter entire biotic assemblages. For example, the introduction of lake trout to Yellowstone Lake may alter not only the cutthroat trout and other fish assemblage, it may affect populations of grizzly bears that utilize this resource.

2. Knowledge about potential links between physical and biological attributes within ecosystems will provide a basis to focus monitoring efforts in specific locations where alien species will potentially be most successful. For example, preliminary data suggest that Didymo is most successful at lake and reservoir outlets which provide stable flows and stable substrate (Kilroy et al. 2007 and Kirkwood et al. 2007). Dreissenid mussel distributions are limited by temperature and dissolved calcium (Whittier et al. 2008, McMahon 1996). Therefore, a monitoring effort that includes sites with stable flows and substrate would enhance the detection of Didymo, if it is present. Sites that possess the temperature and calcium ranges most tolerant for Dreissenid mussels would enhance its detection.

3. Climate change is altering the physical habitat for alien and native species and the long term effects of invaders and the habitat that they will be able to colonize cab change through time (Lee et al. 2008). The response of native biota to these compound stressors are not easily predicted (Paine et al. 1998). Lakes are model ecosystems for detecting climate change, and there are several examples where they have changed both physically (Magnuson et al. 2000) and biologically (Winder and Schindler 2004). Through the

monitoring of the changing physical conditions, resource managers can more completely understand the impacts of invasive species on biota.

Physical and Chemical Measurements in Lakes

Epilimnetic and littoral temperature should be monitored with automatic data loggers which are inexpensive and reliable. Whenever biological data are collected, and at least several times during the ice-free season, we suggest measuring standard limnological data such as temperature and dissolved O_2 profiles. Water clarity should be measured using a Secchi disk (Wetzel and Likens 2000). Measure light penetration as the extinction coefficient calculated from a light profile with lake depth (Wetzel and Likens 2000).

Chemical measures should include basic water chemistry: acid neutralizing capacity, pH, electrical conductivity, and common anions (HCO_3^-, SO_4^{2-}, Cl^-), and dissolved cations (Na^+, Ca^{2+}, K^+, Mg^{2+}) following standard methods (Wetzel and Likens 2000, Eton et al. 2005). Macronutrients that can limit production and algal assemblage structure in lakes (i.e. N, P, and in lakes, Si) (Kilham et al. 1996) should be measured. These nutrients are likely to vary on shorter time scales and have stronger biotic feedbacks than ions such as Na^+ or SO_4^{2-}, and may be monitored at greater temporal frequency in focal habitats, *e.g.* whenever biological data are collected. Analyses should focus on dissolved (soluble reactive P, NH4+ NO3+, SiO4) and total N and P following standard methods (Eaton et al. 2005).

Physical and Chemical Measurements in Streams and Rivers

Like lakes, temperature is easily monitored in focal habitats using thermologers such as I-buttons. Properly installed, these can operate for a year prior to retrieval and downloading.

Stream morphology should be measured as mean width, depth at sampling sites. Stream discharge is measured as mean velocity multiplied by the cross-sectional area with standard approaches (Gordon et al.2004). The type of substrate determines biotic assemblages and should be measured at least once per ecosystem sampled, and possibly more often if the stream or river is subject to human-induced changes in sediment type. Substrate can be sampled with Wolman pebble counts (Gordon et al. 2004). Other physical attributes of streams and rivers (elevation, watershed area, surficial geology, slope, etc.) can be obtained from GIS.

Stream water chemistry, at a minimum, could consist of a grab or an integrated depth sample taken during biological sampling and processed in the same manner as lake water. Temporal sampling could be beneficial at priority sites or could be co-located with on-going monitoring.

DECONTAMINATION OF SAMPLING GEAR AND EQUIPMENT

Sample collection requires movement of sample gear and equipment that has been submerged during the sampling process (i.e., boots, gloves, sampling devices, and associated equipment) between sites. This makes the equipment a potential dispersal vector for invasive species which are established or which may become established in the GYA (see Table 1 for a list of species of concern). A single individual or monitoring team may sample several sites within a single day and many sites within several days. Such rapid movements among sample sites does not allow the equipment or gear to be dried for a sufficient time to assure 100% mortality of all life stages of the invasive species that may become attached or entangled, particularly because many invasive species tolerate extended periods of emersion. Thus, decontamination protocols for sample equipment must be established and followed rigorously by all personnel for all monitoring programs initiated within the GYA. Decontamination procedures should be based on established HACCP (Hazard Analysis and Critical Control Point) applications for aquatic invasive species (for an example see State of Oklahoma Water Resources Board 2005 and Kilroy 2005).

Sytsma and Morgan (2008) sampled six lakes and 22 river access sites in Teton County, WY, for invasive and indigenous species of plants and animals. Prior to leaving each sample site, their team thoroughly scrubbed all equipment to remove any attached organisms and followed with a treatment of a general biocide that consisted of a 3% solution of Sparquat 256®, a proprietary polyquaternary ammonium compound with known biocidal properties. The equipment was then air dried to complete the decontamination procedure prior to subsequent use in a new sampling site. This procedure is commonly used to decontaminate aquatic animals, plants, algae, fungus, and disease organisms on aquatic sampling gear and equipment (U.S. Forest Service 2007) and was approved by the Wyoming Game and Fish Department and the National Park Service prior to initiating the Teton County aquatic sampling program.

DATABASE

A survey and monitoring program for the Greater Yellowstone Area should include a readily accessible account of the results. Access to such an account would be best facilitated if the data were available through an on-line database that is coordinated with the Nonindigenous Species Database Network (NISbase), the National Institute of Invasive Species Science (www.NIISS.org), and the Nonindigenous Aquatic Species (NAS) database.

The NAS database, hosted by the USGS (http://nas.er.usgs.gov), is a central repository for spatially referenced biogeographic accounts of nonindigenous aquatic species. It was established by the federal Aquatic Nuisance Species Task Force under the authority of the Nonindigenous Aquatic Nuisance Species Control and Prevention Act of 1990 (P.L. 101-646) with the primary goal of providing timely, reliable data concerning the distribution of nonindigenous aquatic species within the United States. The NAS database is a spatial information system that is interactive, capable of providing maps, running test-based queries, and providing fact sheets for many nonindigenous species. The NAS information system is coordinated with NISbase, a distributed database system capable of querying multiple nonindigenous-species databases (similar to the NAS system) and compiling the results in a consistent format. NISbase covers all nonindigenous species, and thus, is larger in scope than the NAS system (which is designed specifically for aquatic species). NISbase was designed by the Smithsonian Environmental Research Center and the USGS and built around existing NIS databases to provide a central interface to existing repositories of nonindigenous species records and information. Thus, NISbase serves as a gateway for many researchers into the various existing databases.

The NAS database is part of the NISbase network. Thus, if data collected within the GYA were coordinated with the NAS system, these data would be automatically coordinated with NISbase. However, the NAS system only hosts records of confirmed presence for nonindigenous species. It does not host records for monitoring activities where nothing or only native species were found. Such information is useful to biologists and natural resource managers for many reasons. These include the ability to assess current habitat health and plan on-going or future survey and monitoring activities. Thus, it is recommended that a survey and monitoring program within the Greater Yellowstone Area establish a database that can accommodate all collected data, including data on native and non-indigenous organisms. Meanwhile, the data collected for nonindigenous species should be maintained in a way that is compatible with the existing database structure (schema) of the NAS database to allow for synchronization and compatibility with the NISbase system. Accordingly, database development should be coordinated with the program lead for the Nonindigenous Aquatic Species Program, Dr. Pam Fuller, USGS.

Survey and monitoring activities within the Greater Yellowstone Area may accumulate a substantial amount of data, including habitat parameters, water quality information, native and non-native species presence and density, and other information. This may warrant an independently maintained database, which would require dedicated personnel and an appropriate infrastructure. Alternatively, these data may be incorporated into an existing database that can be expanded to accommodate data specific for the GYA. Again, we recommend consulting with Dr. Pam Fuller of the Nonindigenous Species Program, USGS.

The integration of field data with environmental gradients (e.g. calcium) as well as current and future climatic conditions is needed for potential habitat suitability analyses for aquatic invasive species that would be necessary to support adaptive monitoring and surveys. This can be achieved by either independent ecological modelers trained in species environmental-matching that use niche modeling techniques, or by an automated online system. For example, for terrestrial invasive species such tasks can be accomplished online with the Global Organism Detection and Monitoring system (GODM) at the National Institute of Invasive Species Science (www.NIISS.org) website. The GODM provides users with the capability to upload and store their data (with spatial coordinates and other attributes), manage and update their data, display and overlay the data (using GIS), and generate potential habitat distribution maps for the species of interest at multiple scales. For marine environments, examples of such systems include FishBase (http://www.iobis.org/) and OBIS (http://www.iobis.org/), which are both dynamically linked to Kansas Geological Survey Mapper (KGSMapper, http://drysdale.kgs.ku.edu/website/Specimen_Mapper/). The FishBase, OBIS and other such websites provide coarse resolution predictive maps of species distribution which may not be very useful for resource managers. However, NIISS provides higher resolution (~1km or less) predictive maps. It is also flexible and can be modified for aquatic invasive species. Therefore, the user has much more control over which modeling algorithm to choose and which environmental variables to include in a predictive and potentially suitable habitat model for an aquatic invasive species. More information about NIISS can be obtained by contacting Dr. Jim Graham (jim@nrel.colostate.edu) or Dr. Sunil Kumar (sunil@nrel.colostate.edu), research scientists at Natural Resource Ecology Laboratory, Colorado State University.

PRIORITIZATION OF INVENTORY AND MONITORING

For the efficient use of available funds, priority must be given to monitoring tasks that are potentially the most effective and will detect aquatic invasive species. While many factors will influence the establishment of priorities, there are three primary factors to be given the greatest priority; potential vectors, pristine nature of the water body, and the likelihood of invasion and downstream spread.

There is general consensus that potential vectors of AIS movement in the GYA should be given high priority when developing a monitoring plan is developed. In particular, data support the conclusion that boat and angling use increase the likelihood of aquatic invasive species occurrence and spread. Therefore, a monitoring effort should be designed and conducted in concert with the current boating and angling survey (GYA Implementation Plan 2008) to direct a portion (possibly 2/3) of the sampling effort at areas defined as high use during the survey(s). High priority should be given to water bodies with potential vectors of AIS transport that also have connectivity to other waters, which is often the case in the GYA.

Another priority is the pristine nature of a defined area, therefore, at least a portion of the monitoring effort (possibly 1/3) should be delegated to studying pristine sites. These sites are not only important because of their inherent scientific and public value, but they tend to be upstream and once invaded, would provide a source of invasion to downstream sites. These sites often tend to include Endangered Species Act listed, Sensitive, or Species of Concern.

An additional priority would be the use of the available physical and biological data to define the likelihood of not only the establishment of an invasive species, but subsequent downstream invasion. Within this level, priority would be given to sites where conditions are conducive for an aquatic invasive species. For example, rivers where the bed mobilizes during floods may not provide a conducive environment for the continued survival of New Zealand mudsnails (e.g. mainstem Snake River during spring runoff) as opposed to a slower moving stream (e.g. Polecat Creek during low flow). And the stability of flow in lake outlets may provide a good environment for didymo (e.g. outlet Phelp's Lake, Grand Teton National Park) and the dissolved oxygen (DO) stratum in Jackson Lake may provide a good environment for benthic filter feeders, such as dreissenid mussels. Physical characteristics such as temperature and calcium could also be used for prioritization. For example, the risk of quagga mussel infestation is considered moderate at 20 – 28 mg Ca/L dissolved and high at > 28 mg Ca/L dissolved (Whittier et al. 2008). The temperature range is 9 – 24^{o}C with peak infestation likely at 18 ^{o}C for zebra mussels. But the duration of infestation of aquatic invasive species is likely limited in hot geothermal waters that exist in the GYA.

We should also consider that the three priorities above cannot only assist in the initial prioritization of sites, but can also assist in the adoption of an adaptive monitoring style. With this style, the previously collected information can assist managers to further refine the monitoring framwork. This is not unlike an adaptive management style where the future success of the monitoring framwork is directly linked to lessons learned from the previously collected data.

The three priorities discussed above could be applied to factors that will comprise the specific monitoring study and may include 1.) aquatic invasive organisms of particular concern 2.) sample sites within various habitat types, and 3.) the frequency of sample collection efforts. Top priority and the greatest effort (2/3) of sites would be devoted to detecting the occurrence of known organisms of concern (see focal species list, Table 1) with high vector potential. Particular attention should be given to New Zealand mudsnail, Eurasian watermilfoil, hydrilla, didymo, zebra/quagga mussels, and non-native fish because of the scientific evidence describing their affects on the ecological stability of aquatic environments. As a result, habitats for sampling would include both benthic substrates and the water column. It is important to remember that the monitoring framework is designed to investigate the occurrence of invasive species that have yet to be discovered. Therefore, the short list of aquatic invasive species of special concern in Table 1 does not imply that these are the only invasive species to consider. This short list is provided to define the habitat types that would be necessary to study in the monitoring framework. The sites with high vector potential would require a greater frequency of sample collection in the monitoring design, while pristine sites, though still important, would be sampled less frequently. We should take care not to consider the less frequent sampling effort as a statement that these sites have less priority. Rather, it is unlikely that they will need to be sampled with the same frequency as the high vector potential areas because of their pristine nature (see methodology sample strategy).

In summary, we have defined potential vectors, pristine nature, and likelihood of invasion with downstream spread as three factors with the greatest priority in a monitoring effort. As a result, the specific study design will be weighted between greater use sites with a large vector potential (2/3) and pristine sites with potential risk of spread downstream (1/3). Finally, the specific study should include both benthic and water column sampling efforts because invasive species exist in both habitat types.

CONCLUSIONS

AIS monitoring within the GYA is necessary to detect the presence of AIS to a high degree of certainty, to monitor established AIS populations, and to detect the response by native biota to a future invasion. The intent of the GYA AIS Inventory and Monitoring Framework is help direct limited AIS inventory and monitoring resources in an efficient and effective manner. It will be applied when the GYA AIS Cooperative collaborates on AIS inventory and monitoring. It is not intended to dictate what individual entities such as federal, state, and county agencies do. However, it does encourage collaboration and cooperation whenever possible.

Although specific AIS surveys may occur, AIS data can be collected in conjunction with other aquatic and riparian monitoring efforts. Whenever possible, inventory and monitoring efforts should be holistic, taking advantage of the field visit by using a methodology that detects numerous species and documents multiple parameters. The potential sample methods, locations, and timing require that the life history and habitat preferences of the species of concern be considered. A justified method for statistically evaluating the power of a sampling design to detect a non-indigenous species early in the invasion process should be considered when establishing a monitoring effort. Either qualitative or quantitative monitoring efforts are acceptable, but quantitative efforts provide a better opportunity for comparisons through time. Monitoring efforts should be maintained through time to detect variations in native and non-native population densities over time. Physical habitat parameters and land use impacts should also be documented at monitoring sites.

AIS inventory and monitoring financial and personnel resources are limited, so it is important to prioritize efforts whenever possible. Prioritization could be based upon invasion vectors and probability of invasion. Although areas likely to receive and support establishment of invasive species should be used to prioritize inventory and monitoring survey efforts, consideration should also be given to collecting data at less susceptible sites. The three factors with the greatest priority in our inventory and monitoring efforts are potential vectors, pristine nature, and likelihood of invasion with downstream spread. As a result, the specific study design will be weighted between greater use sites with a large vector potential (2/3) and pristine sites with potential risk of spread downstream (1/3).

There is an essential tie between recreational water user contact surveys and this monitoring/inventory framework. Regular water user contact surveys help identify the highest priority monitoring locations within the GYA. It would be these areas that become annual, high intensity monitoring locations. A stratified survey design that includes sites outside of fixed annual sites, should include a stratification based on habitat types such as streams/rivers, lakes, and wetlands/ponds. Each habitat type should be represented. Standard sampling methodologies should be employed whenever possible.

It is recommended that the GYA Inventory and Monitoring Program utilize existing databases whenever possible. The USGS NAS database should be used to document the occurrence of AIS species detected. However, the NAS database does not accommodate other data such as physical parameters of uninfested waters or population density of native species, so it is recommended the GYA Inventory and Monitoring Program establish a database that can accommodate all collected data, including data on native and non-indigenous organisms. The data collected for nonindigenous species should be maintained in a way that is compatible with the existing database structure (schema) of the NAS database to allow for synchronization and compatibility with the NISbase system. Accordingly, database development should be coordinated with the program lead for the Nonindigenous Aquatic Species Program, Dr. Pam Fuller, USGS.

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