

## Chapter 2: Ecoregion 5.2.1 Northern Lakes and Forests: Marcell Experimental Forest, Minnesota

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### Introduction

The Marcell Experimental Forest (MEF) has a rich and long history of research focused on peatland hydrology and biogeochemistry (Kolka et al. 2011c). Research at MEF has encompassed atmospheric deposition chemistry, peatland nutrient cycling, mercury (Hg) cycling, soil properties, tree-stand dynamics, and climate change. Various watershed treatments have been applied to uplands, bogs, or fens to investigate impacts of management practices on water yield, peak streamflow, water quality, and nutrient processing (Kolka et al. 2011b, Verry et al. 2011a). Biogeochemical research at MEF has focused on nitrogen (N), phosphorus (P), dissolved organic carbon (DOC), Hg, and sulfur (S) cycling (Urban et al. 2011). This chapter reviews monitoring and research that has occurred at the MEF related to nutrient biogeochemistry and biological responses to nutrients.

### Site Description

The MEF is located in north central Minnesota (lat. 47° 31' 52" N, long. 93° 28' 07" W) (Verry and Janssens 2011). Elevations in the 1141-ha site range from 412 to 440 m. MEF straddles a continental divide and contains headwaters of rivers draining south to the Gulf of Mexico, and north to James Bay. MEF is the only site in the experimental forest and range (EFR) network of the U.S. Department of Agriculture, Forest Service (USFS) that focuses on the hydrology and biogeochemistry of naturally occurring peatlands in boreal forests (Sebestyen et al. 2011a).

The climate at MEF is continental, with moist warm summers and dry cold winters (Sebestyen et al. 2011a, Verry and Janssens 2011). The mean annual temperature is 3.4 °C (Sebestyen et al. 2011a). Monthly average temperatures range between 19 °C during July and -15 °C during January. Since 1961, the mean annual air temperature has risen by 0.4 °C per decade with the greatest increase in temperature occurring during the winter months (Sebestyen et al. 2011a). Annual precipitation averages 78 cm, of which 33 percent occurs as snow. Mean monthly

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precipitation ranges from 2.1 cm in February to 8.9 cm in August. Total annual precipitation has not changed significantly since 1961 (Sebestyen et al. 2011a).

Landforms of the MEF are a result of past glaciation (Sebestyen et al. 2011a). Lakes and peatlands formed from ice-block depressions that filled with organic soils of 3 to 10 m depth (Verry and Jannsens 2011). MEF contains forested bogs, fens, and poor fens developed on various types of peat. Bedrock geology consists of Ely greenstone and Canadian Shield granite overlain by 8 m of till and sandy outwash (Paulson 1968). Sandy loam soils comprise most of the uplands at MEF with sandy outwash soils also common.

Hydrology at MEF is influenced by glacial deposits that form a large regional aquifer. Bogs at MEF are perched above the aquifer and are fed by precipitation (Bay 1968, 1969; Verry and Boelter 1975; Verry et al. 2011c). The centers of most bogs are raised slightly and are surrounded by wet lagg zones (Sebestyen et al. 2011a). Water from the uplands and bog centers flow into the lagg zones, which drain from the bog through short (30 to 200 m) outlet streams. Although lagg zones are small in area, they are very important hydrologically in bogs (Verry et al. 2011c). Most outlet streams draining the bogs are intermittent, flowing only for a few months after snowmelt or during high precipitation periods. Flows from fens are less variable than those from bogs (Verry et al. 2011c). In fens, outlet streams are perennial, as a result of inputs from precipitation and groundwater from the aquifer. All of the outlet streams are low gradient (<0.5 percent), shallow, shaded, and with peat and silt substrate on the stream bottom.

Vegetation in forested uplands consists of mixed stands of white spruce (*Picea glauca* (Moench) Voss), trembling aspen (*Populus tremuloides* Michx.), bigtooth aspen (*P. grandidentata* Michx.), white birch (*Betula papyrifera* Marshall), balsam fir (*Abies balsamea* (L.) Mill.), red pine (*Pinus resinosa* Aiton), and jack pine (*P. banksiana* Lamb.) (Perala and Verry 2011, Verry 1969). Forested bogs contain the tree species black spruce (*Picea mariana* (Mill.) Britton, Sterns & Poggenb.), eastern tamarack (*Larix laricina* (Du Roi) K. Koch), and northern white cedar (*Thuja occidentalis* L.) as well as various species of sphagnum mosses (*Sphagnum* spp.) and ericaceous shrubs (Sebestyen et al. 2011a). Speckled alder (*Alnus incana* (L.) Moench.) often is found in the lagg zone of these bogs. Open fens also contain speckled alder and a variety of understory species, including sedges (*Carex* spp.), marsh marigold (*Caltha palustris* L.), and cattails (*Typha* spp.).

## Research History

Forests in the MEF region were logged between 1865 and 1897, prior to the establishment of MEF (Perala and Verry 2011). The area burned during widespread fires

in 1917, leaving the land unsuitable for agriculture and other uses (Sommer 2008). The Civilian Conservation Corps replanted red pine in a large portion of the southern unit of the MEF area between 1934 and 1941 (Perala and Verry 2011). MEF was formally established in 1962 as an experimental area of bogs, fens, moraines, and sandy drift, and it was divided into north and south units (Verry et al. 2011a). The site has six gaged watersheds, including two reference watersheds (S2 and S5) (fig. 2.1). The first streamflow gage was established at watershed S2 (Swamp 2) in 1960. Climate monitoring began in 1961, and water chemistry sampling began in 1966 (Verry et al. 2011a).

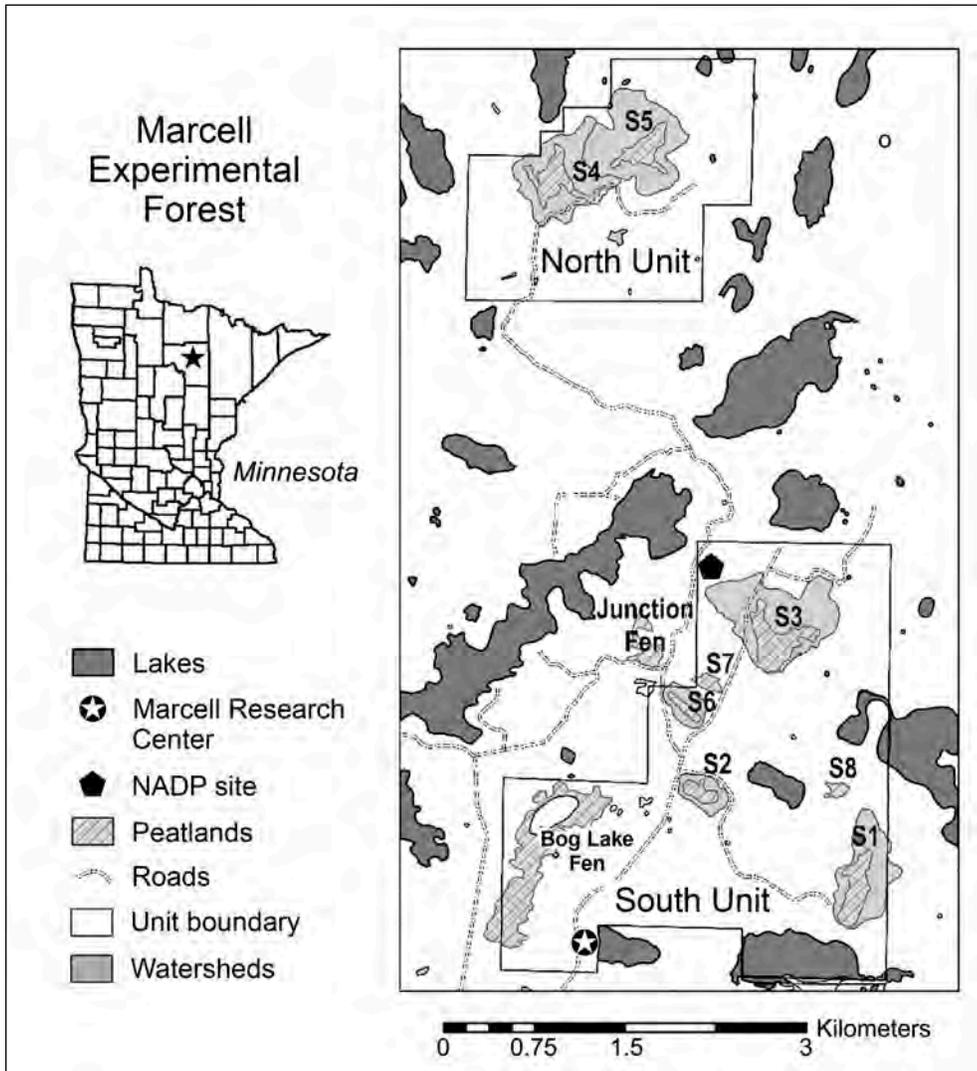


Figure 2.1—Watersheds in north and south units studied since 1961 at Marcell Experimental Forest (MEF), Minnesota. S1 through S8 designate the locations of the eight study watersheds at MEF. NADP = National Atmospheric Deposition Program. Map source: Marcell Experimental Forest, C. Dorrance, cartographer.

**Past research—**

Past research at MEF focused on both basic knowledge about hydrologic and biogeochemical characteristics of northern peatlands and applied studies related to forest management practices in peatlands. Watersheds at MEF were sites of the first detailed studies of water and solute budgets in bogs and fens in the Great Lake states (e.g., Bay 1967; Knighton and Stiegler 1981; Kolka et al. 2001; Nichols and Verry 2001; Urban 1983; Urban and Eisenreich 1988; Urban et al. 1987, 1989a, 1989b, 1995; Verry 1975; Verry and Urban 1992/1993). An analysis of long-term records of geology, soils, groundwater wells, water chemistry, peat-core dating, pollen analysis, and a review of glaciation records for the S2 reference watershed at MEF was used to interpret 12,000 years of ice-block peatland development (Verry and Janssens 2011). Peat from MEF watersheds was instrumental in research characterizing the physical properties of organic soils and their relationships to water (e.g., Boelter 1964, 1965, 1969; Nichols and Boelter 1984; Verry et al. 2011b). Diurnal and seasonal albedo variations and evaporation characteristics of various types of forested and harvested peatlands were documented at MEF by Berglund and Mace (1972, 1976) and Brooks et al. (2011).

Aquatic invertebrates were surveyed in April 2009 in outlet streams of watersheds S2, S3, and S6. A fish survey was conducted in April 2009 in the permanent outlet stream of S3. Data on periphyton standing crop as chlorophyll *a* and factors influencing benthic periphyton in streams were measured once from June to July 2012 in outlet streams of S1 through S6 and are presented below.

Watershed studies at MEF have evaluated hydrological and water chemistry responses to a variety of forest management techniques. Experimental manipulations have included upland clearcutting of aspen, N fertilization, soil compaction impacts, strip and clearcutting of black spruce, prescribed fire in a harvested fen, and conversion of upland aspen forest to conifer forests using cattle grazing and herbicides (e.g., Knighton and Stiegler 1981; Kolka et al. 2011b; Page-Dumroese et al. 2006; Perala and Verry 2011; Sebestyen and Verry 2011; Sebestyen et al. 2011b; Verry 1972, 1981). MEF was a pilot site for the North American Long-Term Soil Productivity (LTSP) experiment and one of four sites in the Great Lake states that demonstrated the effects of severe soil compaction and removal of organic matter on aspen productivity (e.g., Perala and Verry 2011, Powers et al. 2005). Results from these studies have been used to develop forestry best management practices for the Great Lake states (Verry 1975, 1976; Verry et al. 1983, 2000).

**Current research—**

Current research at the MEF focuses on hydrology, carbon (C), and Hg cycling, and climate change in northern peatlands. New hydrological studies have been initiated to investigate how rainfall interception, evapotranspiration, and subsurface flow differ between reference and managed watersheds and how climate variations affect tree growth (Kolka et al. 2011b). Long-term data collection continues on the rate of emissions of methane and carbon dioxide (CO<sub>2</sub>) from bog and fen peatlands, the rates of carbon storage in peatlands, and variation in DOC composition among hydrologic flow paths (e.g., Grigal et al. 2011, Kolka et al. 2011b, Olson et al. 2013, Shurpali and Verma 1998, Shurpali et al. 1993). A large-scale, 10-year climate change experiment, named Spruce and Peatland Responses Under Climatic and Environmental Change (ORNL 2017), is underway in watershed S1. Responses of vegetation (above- and belowground), lichens, microbes, microinvertebrates, hydrology, and C, N, P, iron (Fe), methylmercury (MeHg), and S cycling to increased CO<sub>2</sub> concentrations and temperature are being monitored. Another recent study examined C, N, and P limitation and other environmental drivers on microbial processes and organic matter decomposition in a fen and bog at MEF (Hill et al. 2014, Seifert-Monson et al. 2014). Results from detailed studies of the inputs and fate of both total Hg and MeHg and interactions with S inputs in an 8-year, large-scale experiment continue to show the close relationship between sulfate (SO<sub>4</sub><sup>2-</sup>) and MeHg production in bogs (e.g., Coleman Wasik et al. 2012; Kolka et al. 1999a, 1999b, 2001; Mitchell et al. 2008a, 2008b, 2008c, 2009). In 2012, a study examining the effects of upland clearcutting and biomass harvesting on Hg cycling using enriched stable Hg isotope tracers was begun (Mazur et al. 2014). Results from each of these studies are critical for understanding mechanisms of biogeochemical cycling and predicting the consequences of global climate change and land management in peatland landscapes.

MEF also continues to participate in various long-term data networks. In 1978, MEF was established as one of the first National Atmospheric Deposition Program (NADP) (NADP 2017) sites in the United States. MEF was also a pilot site for measuring Hg deposition in the Mercury Deposition Network and developing methods for the LTSP Network. MEF also participates in the National Phenology Network, the EcoTrends Project Network, the United States-China Carbon Consortium, and the USFS EFR synthesis network (Kolka et al. 2011b). For more details about MEF and research conducted at MEF, see Kolka et al. (2011b, 2011c), Verry et al. (2011a), and USDA FS (2017a).

**Availability of publications and data—**

A partial bibliography of MEF scientific publications can be found online (USDA FS 2017d). A hydrology database is available (USDA FS 2017b). Metadata about the hydrology and climate database (air temperature, water tables, stream runoff, precipitation, soil properties, frost, and snow) are available online (USDA FS 2017c). Data from the Marcell NADP site (precipitation-weighted mean concentrations for calcium, magnesium, potassium, sodium, ammonium ( $\text{NH}_4^+$ ), nitrate ( $\text{NO}_3^-$ ), chloride and sulfate, pH, and conductivity) are also available online (NADP 2017). Long-term MEF daily streamflow and meteorological data, formatted for easy comparisons with a large number of other EFR and LTER sites, can be downloaded from the ClimDB/HydroDB website (LTER Network 2017).

## **Biological Responses to Stream Nutrients N and P**

### **Issues of Concern**

Nutrients such as N and P often limit the growth of benthic algae (periphyton) in streams and phytoplankton in lakes. Eutrophication results from excess dissolved N and P, which can stimulate algal production in headwaters such as those at MEF or in downstream waterbodies. The water standard for  $\text{NO}_3^-$  of 10 mg  $\text{NO}_3^-$ -N/L is based on the concentrations in drinking water that can cause methemoglobinemia in humans (Fan and Steinberg 1996). Multiple forms of N may also be toxic to aquatic life (Camargo et al. 2005). Biologically active forms of N,  $\text{NH}_4^+$ , and  $\text{NO}_3^-$ , may be elevated in peatland streams by atmospheric deposition. Total nitrogen (TN) in wet deposition at MEF is similar to that reported from eastern forests (Aber et al. 2003, Driscoll et al. 2003). There has been no significant change in  $\text{NH}_4^+$  in wet deposition at MEF since 1979, but  $\text{NO}_3^-$  concentrations in wet deposition have decreased over that same time period (Sebestyen et al. 2011a, Urban et al. 2011) corresponding with declines in regional emissions of N oxides (McDonald et al. 2010). Total phosphorus (TP) is of less concern than N in peatland streams as P leached from the canopy in throughfall and stemflow is strongly retained by soils, which are a net sink for P, and concentrations in peatland outlet streams are low (Urban et al. 2011, Verry 1975, Verry and Urban 1992/1993).

### **Findings From Studies**

**Nutrient levels in MEF peatland streams—**

Past research on N cycling in a MEF bog (S2) showed that high C:N ratios within bog peat (Malmer and Holm 1984, Urban and Eisenreich 1988) led to about 90 percent retention of inorganic N of inputs to S2 (Urban et al. 2011). Peak concentrations of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  in streamflow occur usually during early snowmelt, while

minimum concentrations occur in summer (Urban et al. 2011). Spring snowmelt is the only time of year when  $\text{NO}_3^-$  is measurable in the outflow from S2. Hill et al. (2014) reported  $\text{NO}_3^-$  concentrations at the outflow of a MEF bog was 0.711 mg/L (annual TN yield of 0.70 kg/ha/yr) and from a fen was 0.409 mg/L (annual TN yield of 0.73 kg/ha/yr), during May–October, from 2010 to 2012. Studies by Grigal (1991) at MEF found that perched bogs, which formed by lake filling, were less N limited than raised bogs, which formed by landscape swamping. Urban et al. (2011) projected that if the rate of atmospheric inputs of N continues its current decline of 1 percent per year at MEF, peatland watersheds would likely become even more retentive over time and experience decreases in terrestrial primary production, litter quality, and overall rates of N cycling.

Fewer data are available for P cycling in MEF watersheds. Urban et al. (2011) summarized data from Verry (1975) for total dissolved phosphorus solute concentrations in streamflow from a MEF perched bog (0.19 mg/L), groundwater fen (0.09 mg/L), and poor fen (0.47 mg/L) for the period 1968–1972. Hill et al. (2014) reported TP concentrations at the outflow of a MEF bog and fen of 0.301 mg/L (annual yield of 0.18 kg/ha/yr) and 0.032 mg/L (annual yield of 0.51 kg/ha/yr), respectively, during May–October from 2010 to 2012. Element budgets constructed for N and P suggest that 56 percent of N inputs and 38 percent of P inputs are retained in the S2 watershed on an annual basis (Urban and Eisenreich 1988, Urban et al. 2011).

#### **Algal responses to stream nutrients—**

Long-term studies of algal response to nutrients have not been done at MEF nor have such studies been done in other peatlands in the Great Lakes region, but a month-long study in 2012 suggested that nitrate may influence algal growth in MEF outlet streams. Periphyton standing crops in outlet streams were measured in June 2012 in the six MEF peatland watersheds with differing ambient stream water N and P concentrations to determine whether N and P, canopy cover, flow, water depth, water temperature, dissolved anions, dissolved cations, pH, and conductivity significantly influence periphyton growth. Tiles were placed in outlet streams and retrieved after 30 days for chlorophyll *a* analysis. Median periphyton standing crop during the June–July 2012 period in the six outlet streams ranged from 3.8 to 53.9 mg chlorophyll *a*/m<sup>2</sup> in the six MEF outlet streams (fig. 2.2). These results were below the benthic chlorophyll *a* values of >150 mg/m<sup>2</sup> in more than 1 year in 10 that the state of Minnesota considers to be excessive in Class 2B rivers and streams (Minnesota Administrative Rule 2018), but are similar to values measured in an Alaskan rich fen by another short-term study (Wyatt et al. 2012). Correlation analysis from the short-term monitoring showed that only  $\text{NO}_3^-$  was positively ( $p = 0.03$ ) related to periphyton standing crops across all six watersheds (table 2.1). Total N

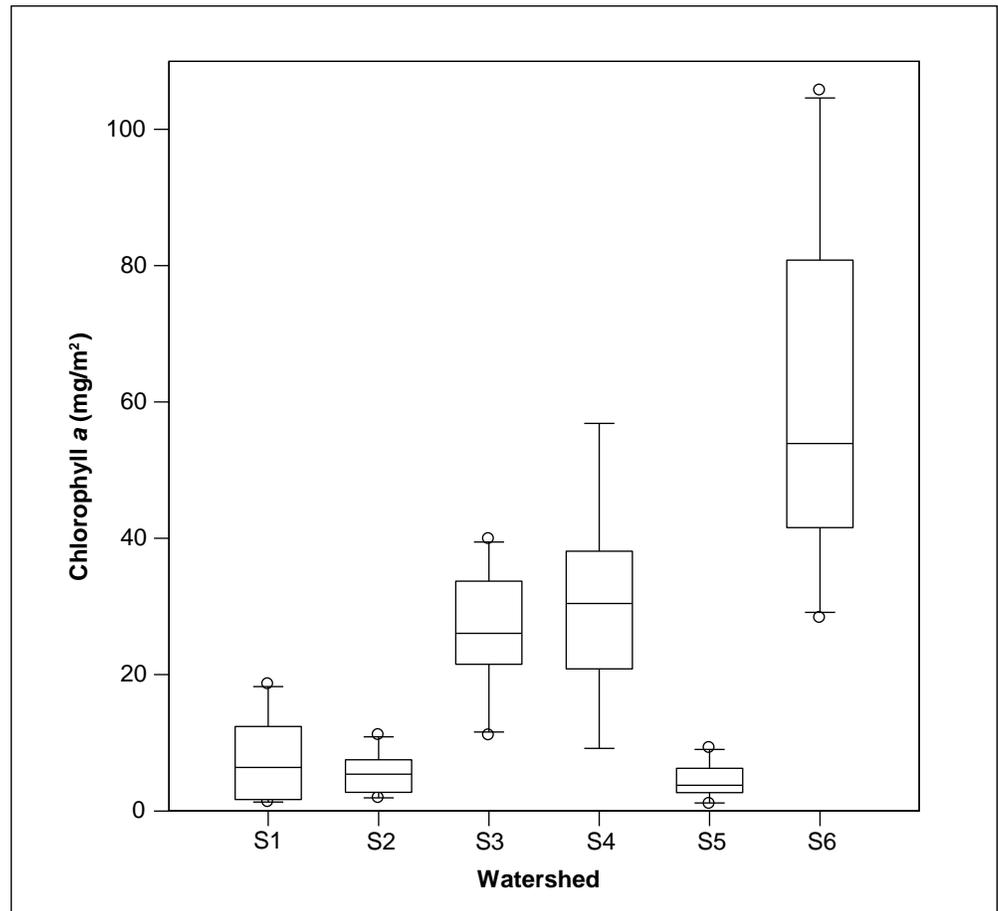


Figure 2.2—Median benthic chlorophyll *a* concentrations and 25<sup>th</sup> and 75<sup>th</sup> percentiles for six Marcell Experimental Forest outlet streams with a range of ambient nitrate concentrations (0.01 to 0.59 mg/L) in June 2012. Error bars = 10<sup>th</sup> and 90<sup>th</sup> percentiles. N = 10 tiles per watershed.

**Table 2.1—Correlations among mean periphyton standing crops measured as chlorophyll *a* concentrations on tiles placed in stream bottoms, physical habitat variables, and water quality variables for six outlet streams (S1 through S6) with a range of ambient nitrate concentrations at Marcell Experimental Forest in June 2012<sup>a</sup>**

Factor	r	p	Factor	r	p
Percent canopy cover	-0.43	0.39	Nitrate	0.84	0.03*
Water depth	-0.46	0.36	Sulfate	-0.44	0.39
Current velocity	-0.10	0.86	Chloride	-0.68	0.14
pH	0.22	0.68	Calcium	0.15	0.78
Conductivity	0.02	0.98	Iron	0.13	0.81
Water temperature	0.68	0.14	Potassium	-0.70	0.12
Total nitrogen	0.75	0.09	Magnesium	0.22	0.68
Total phosphorus	0.19	0.72	Sodium	0.29	0.58
Dissolved organic carbon	-0.12	0.82	Silica	-0.29	0.58
Dissolved inorganic phosphorus	0.28	0.59			

<sup>a</sup> Periphyton standing crop has significant relationship ( $p < 0.05$ ) with factor of interest.

was positively but not significantly ( $p = 0.09$ ) related to periphyton standing crops. No significant relationships between TP ( $p = 0.72$ ) or dissolved inorganic phosphorus ( $p = 0.59$ ) and periphyton standing crops were found. Multiple regression models relating chlorophyll *a* standing crop to important variables showed that only  $\text{NO}_3^-$  was a significant predictor of periphyton standing crops in peatland outlet streams (Chlorophyll *a* [ $\text{mg}/\text{m}^2$ ] =  $14.128 + [77.230 \times \text{NO}_3^-]$ ,  $r^2 = 0.71$ ,  $p = 0.03$ ).

#### **Microbial responses to nutrients within peatlands—**

Recent work by Hill et al. (2014) examined N, P, and C limitations on the microbial processes of respiration and coenzyme activity and organic matter processes within MEF fens and bogs. They found that microbial respiration, coenzyme activity, and decomposition were more closely related to soil pools of C, N, and P than to atmospheric sources of these elements, and suggested that localized nutrient availability may be a better predictor of peatland microbial processes in a changing climate than regional climate models.

### **Other Factors Relevant to Biological Responses to Stream Nutrients N and P**

Many other factors that may limit biological responses to stream N and P have been measured or monitored at MEF, including long-term streamflow, meteorology, atmospheric deposition, and DOC concentrations.

Short-term results from the periphyton colonization study described above suggested that none of the nonnutrient variables that were measured during the 30-day period, including canopy cover, flow, water depth, water temperature, and other water quality parameters of anions other than nitrate, cations, pH, DOC, and conductivity, were significantly related to periphyton standing crops in MEF outlet streams.

### **Dose-Response Studies**

The only dose-response studies that have been conducted at MEF have been the short-term study of the response of periphyton to ambient stream nutrient levels described above.

### **Reference Watersheds**

Two reference watersheds (S2 and S5) were established at MEF in 1960 and 1961, respectively (Sebestyen et al. 2011a). These watersheds are representative of relatively undisturbed bog watersheds in the region. Both watersheds contain second-growth upland forests that were previously logged in the late 1800s and have experienced multiple wildland fires since then, as have most watersheds in the region (Verry and Janssens 2011). Argerich et al. (2013) found no long-term

trends in nitrate or ammonium concentrations in stream water from these MEF reference watersheds, despite long-term declines in atmospheric deposition of N. This response differed from what has been found in mountainous reference watersheds in the Appalachians, where stream nitrate fluxes tracked long-term trends in atmospheric deposition of N (see chapters 3 and 14 on Hubbard Brook Experimental Forest and Coweeta Hydrologic Laboratory, respectively). No long-term trends in P in stream water from MEF reference watersheds have been reported.

The S2 reference watershed contains a 3.2-ha black spruce bog and a 6.5-ha aspen and birch upland (fig. 2.1) (Sebestyen et al. 2011a). The S2 watershed has been used as a reference for experiments conducted on the S1, S3, and S6 watersheds. The S5 watershed contains a 6.1-ha black spruce bog and a 46.5-ha upland of aspen and balsam fir (fig. 2.1). The S5 watershed is used as a control watershed for the S1 and S4 watersheds. Discharge from both reference watersheds is monitored with v-notch weirs at their outlets. Biweekly water chemistry samples have been collected upstream of the weirs since 1975 in S2 and S5, except for 1983-1988 when samples were not collected in S5. In watershed S2, chemical analyses have also been performed on samples collected during storms at the weir since 2008 and monthly water samples from a groundwater well in the upland since 1987 (Sebestyen et al. 2011a). Nutrient transport in surface runoff and interflow from S2 was reported by Timmons et al. (1977) and Verry and Timmons (1982) and is currently being monitored. Upland and bog wells have been monitored for water table elevations in both watersheds since the early 1960s. Meteorological stations in the uplands of S2 and S5 have recorded maximum and minimum air temperature, relative humidity, and precipitation in the watersheds since the early 1960s, and continuous air temperature has been measured since 1997. Snow and frost depth and snow water equivalents have also been measured in both watersheds since 1962. Peat and upland soil temperatures and upland soil moistures have also been measured in the reference watersheds at various intervals for varying periods of time. A complete list of hydrologic and climate variables measured in S2 and S5 over time is in Sebestyen et al. (2011a). Aquatic invertebrates were surveyed at the S2 outlet stream in April 2009 (Batzer et al. 2016) and periphyton standing crop as chlorophyll *a* and important habitat variables were measured in June–July 2012 in both S2 and S5 outlet streams.

### Cross-Site and Regional Studies

MEF data on  $\text{NO}_3^-$  and  $\text{NH}_4^+$  concentrations in forested peatlands were included in a synthesis study examining long-term trends in stream nitrogen concentrations for forest reference catchments across the United States (Argerich et al. 2013). A synthesis of streamflow responses to climate change by Jones et al. (2012) included

data collected at MEF. Impacts of forest management, such as harvesting and regeneration, on stream water yields at MEF were included in a cross-site comparison study of four experimental forests in the northeastern United States (Hornbeck et al. 1993). The S2 watershed served as an undisturbed reference site in a network of sites used to assess nutrient runoff over an agricultural gradient in Minnesota during the 1970s (Verry 2015). Long-term hydrology and climate data from MEF were included in a synthesis study examining interactions among climate, streamflow, and forest management practices at 10 U.S. USFS EFRs (Vose et al. 2012). The role that MEF played in the development of ecosystem science and biogeochemical cycling research at USFS EFRs was discussed in Vose et al. (2014).

## Responses to Management

### **Effects of harvest and regeneration of upland aspen forests on stream chemistry—**

To better understand the effects of aspen harvesting on water quality and stand regeneration, the uplands of the S4 fen were whole-tree clearcut during 1970 and 1971 (Perala and Verry 2011). After the seventh year of regeneration, the uplands were aerially fertilized with ammonium nitrate (340 kg/ha) (Berguson and Perala 1988). Water yields and solute concentrations in one of two outlet streams of the harvested S4 watershed and the outlet stream of the S5 reference watershed were monitored from 1967 to 1982 and from 1989 to present (Sebestyen and Verry 2011). Water yields from S4 were elevated for 10 years after harvest (Sebestyen and Verry 2011, Sebestyen et al. 2011b, Verry 1972).  $\text{NO}_3^-$  + nitrite and  $\text{NH}_4^+$  concentrations (annual volume weighted) and yield ratios (i.e., ratio of treatment to reference solute yields) did not increase in the treatment stream relative to the reference stream until the treatment watershed was fertilized in 1978 (Sebestyen and Verry 2011). Immediately after fertilization, the treatment to reference ratios of stream  $\text{NO}_3^-$  concentrations increased 97-fold compared to reference ratios, while treatment to reference ratios of ammonium concentration ratios increased 6-fold immediately after fertilization. Treatment ratios declined over 2 years but remained elevated over reference levels for 10 years. Biological responses to stream water N and P were not included in this study.

### **Conversion of an upland forest from hardwoods to conifers—**

The uplands of the S6 bog were harvested by whole-tree clearcut in 1980 to test the efficiency of herbicide vs. cattle grazing in the preparation of hardwood sites for conversion to conifers (Perala and Verry 2011). Following harvesting, a portion of the S6 uplands was treated with the herbicide Weedone 170, while another portion was grazed by cattle (2.5 individuals/ha) for 3 years to prevent the regrowth of aspen. In

1983, the uplands were replanted with red pine and white spruce (Perala and Verry 2011). The S2 watershed served as a reference site for hydrologic and solute comparisons (Sebestyen and Verry 2011). While water yield increased immediately in S6 and remained high for 10 years (Sebestyen et al. 2011b), solute responses were delayed. Nitrate + nitrite concentrations and yield ratios increased by an order of magnitude during year 2 after clearcutting and declined immediately to levels observed in the S2 reference stream (Sebestyen and Verry 2011). Ammonium concentrations and yield ratios increased threefold during year 2 following clearcutting. Total organic N concentrations and yield ratios peaked during years 4 and 5 after harvest. Total P concentrations and yield ratios increased during the first 7 years after harvest (Sebestyen and Verry 2011). Fecal coliform counts were measured for 6 years to monitor the effects of cattle on stream water quality during the conversion from hardwoods to conifers (Sebestyen and Verry 2011). Fecal coliform counts were ~three times greater at the grazed S6 weir than at the reference S2 weir. Below the S6 weir, coliform counts were also higher in the outlet stream as a result of cattle entering the stream. Biological responses to stream water N and P were not measured in this study.

#### **Response of phosphorus to clearcutting in a bog and fen—**

Several experiments were conducted at MEF peatlands during the 1970s to examine harvesting effects on water yields and P concentrations in bog and fen watersheds (Sebestyen and Verry 2011). In the S1 bog, black spruce was whole-tree clearcut in alternating strips that left uncut trees to serve as seed source for regeneration in the cut strips (Verry and Elling 1978). Five years after regeneration began, the remaining uncut strips of black spruce were harvested. The S3 fen was clearcut harvested in the winter of 1972 and reseeded after burning (Sebestyen and Verry 2011). The hardwood uplands were not harvested during either experiment. Solute samples were collected from the pools of water backed up behind the weirs and not from the outlet streams. Results from the harvesting experiments were reported in Knighton and Stiegler (1981) and Sebestyen and Verry (2011). Water yields did not change in either the bog or fen after harvest (Sebestyen et al. 2011b). Total phosphorus concentrations above the weirs increased in both the fen and bog after clearcutting. The duration of the increase in total P in the fen lasted at least 6 years, after which data collection ended, while in the bog it lasted only 1 year. Biological responses to stream water phosphorus were not included in either of these studies.

#### **Reliability and Limitations of Findings**

No long-term research has been conducted on biological responses to nutrients in MEF outlet streams. One short-term study, described above, quantified periphyton accrual in MEF outlet streams with a range of nutrient concentrations. Although

this preliminary investigation suggests a positive relationship between  $\text{NO}_3^-$  concentrations in streamflow and algal biomass, it should not be considered definitive until measurements have been made for longer periods of time to account for seasonal and annual variability. Our knowledge of biological responses to nutrients in northern peatland streams is also limited by a lack of basic knowledge of the diversity, feeding habits, and life histories of organisms inhabiting the peatlands and baseline measurements of ecosystem functions such as primary and secondary production, respiration, nutrient spiraling lengths, and organic matter dynamics in peatland streams.

MEF hydrologic and biogeochemical research findings and datasets are likely to represent reliably upper Great Lake states forested bogs and fens that have relatively light human disturbance and low nutrient enrichment, and headwater streams fed by these peatlands. Outside of settings similar to these, MEF findings should be applied with caution and, where greater confidence is needed, validation studies should be performed to test how well MEF findings and data represent local conditions.

## Research Needs

Research in the MEF has provided valuable insights into the effects of forestry practices on hydrology and nutrient export. Significant science gaps exist for both basic and applied questions regarding biological responses to N and P at multiple spatial and temporal scales. Of these science gaps, the basic information that would add the most to the existing body of MEF science related to biological responses to N and P would be how the structural and functional characteristics of microbial, algal, and invertebrate communities compare among MEF peatlands with differing natural levels of nutrients and how they vary seasonally and interannually.

Minnesota promulgated river eutrophication standards in August 2014, making some prior needs less pressing for regulators. However, long-term monitoring of ecosystem processes, such as primary and secondary production, nutrient spiraling, organic matter breakdown, and community responses of aquatic taxa, such as periphyton, invertebrates, and amphibians, that may be altered by nutrients and other management activities are needed by regulators for northern peatland streams. The concept of reference conditions is a high-priority science gap at MEF and should include not only water chemistry data but also aquatic invertebrate and aquatic plant data. These data are of considerable value for establishing “natural background” and are difficult to define where public and private lands may be co-mingled and collection of these data is the type of work that can only be done in an experimental forest setting. By more fully understanding natural processes, it is possible to establish natural background, which can be an issue in determining

whether a water quality impairment exists and can aid in total maximum daily load (TMDL) development.

To fill this highest priority gap, work is needed to address the following questions: Is periphyton growth controlled by ambient stream  $\text{NO}_3^-$  levels? Does this relationship hold across seasons and multiple years? Are microbial and invertebrate production driven by periphyton production or detrital resources? To address these questions, periphyton, invertebrate, and microbial production along with other selected environmental factors that might affect biological activity, for example stream nutrients, flow, temperature, light, leaf litter, DOC, pH, and conductivity, will need to be measured in six MEF streams for 10+ years. In situ monitoring devices that continuously measure factors such as dissolved oxygen concentrations and water temperature in outlet streams, may provide useful, high-resolution data. Collected data would be analyzed for evidence of specific factors that correlate with biological activity across the spectrum of MEF peatland streams.

### Potential Utility to Water Quality Regulatory Agencies

It is important to understand how land use practices may contribute to excessive levels of periphyton and determine the relative importance of instream factors, such as habitat, light, and nutrients, on periphyton growth. Currently, data regarding biological responses to nutrients associated with land use practices do not exist for MEF streams. Results from future studies of stream responses to land use management at MEF may be useful for developing restoration strategies for total P-based TMDLs for low-order streams and serve as a basis for management of riparian corridors to avoid impairment, for example, from excessive periphyton growth or streambank erosion. Existing MEF datasets and analyses of N and P concentrations in stream water originating from forested peatlands could be used to represent contributions to streams from relatively undisturbed wetlands in TMDL models for this region. These existing data plus new collection efforts, as they become available, will augment the paucity of diel data we have on forested peatland streams and provide valuable biological data that can be used for future water quality standard refinements and Tiered Aquatic Life Use standards implementation.

Key points:

- At MEF, there is a long record of N and P monitoring in stream water draining several common peatland types in the northern Great Lake states.
- Responses of N and P concentrations in streamflow related to forest management activities at MEF generally have been of shorter duration and delayed when compared to results of management studies at other experimental forests.

- There have been preliminary, short-term studies regarding biological responses to ambient levels of nutrients N and P in MEF outlet streams under undisturbed conditions, but none in response to forest management experiments.
- The highest priority science gap at MEF related to N and P from the perspective of the regulatory community is to improve our understanding of natural biotic conditions in bogs and what the most important factors are in the growth and management of excessive levels of periphyton in outlet streams.

## **Biological Responses to Stream Nutrients Other Than N and P**

### **Issues of Concern**

Sulfur is a macronutrient of concern in peatlands owing to its transport into peatlands via atmospheric deposition and its strong influence on the transformation of inorganic Hg to MeHg via microbial  $\text{SO}_4^{2-}$  reduction (Jeremiason et al. 2006, Kolka et al. 2011a). MeHg is of great concern because it bioaccumulates in aquatic food webs and reaches highest concentrations in apex predators, especially those that feed on fish. MeHg poses a risk to humans because it is a neurotoxin that people acquire by consuming MeHg-contaminated fish. Sulfate has been shown to penetrate down to the water table in the S2 bog where it is available to be reduced via dissimilatory  $\text{SO}_4^{2-}$  reduction (Urban and Bayley 1986). Sulfate concentrations in wet deposition at MEF have declined significantly since 1979, following the decline in sulfur dioxide emissions in the upper Midwest (Urban et al. 2011), but Hg deposition and MeHg in Great Lake states aquatic systems continue to be a regional concern (Evers et al. 2011). Apart from  $\text{SO}_4^{2-}$  and its effect on MeHg, there are no other major concerns in this region about biological responses to nutrients other than N and P.

### **Findings From Studies**

#### **Long-term monitoring—**

There has been long-term monitoring of several nutrients other than N and P, including major cations, and anions, and DOC, in both stream water and atmospheric deposition at MEF (e.g., Urban et al. 1989a, 1995; Verry and Timmons 1982; Verry and Urban 1992/1993).

**Sulfate levels in MEF peatland streams—**

The cycling of S within peatlands is very similar to N cycling (Urban et al. 2011). Long-term records of  $\text{SO}_4^{2-}$  fluxes in streamflow from S2 and atmospheric deposition show that  $\text{SO}_4^{2-}$  is retained in MEF bogs (Urban et al. 2011). Peak concentrations of  $\text{SO}_4^{2-}$  in streamflow occur during early snowmelt, while minimum concentrations occur in summer (Urban et al. 2011). The decline in atmospheric inputs of  $\text{SO}_4^{2-}$  over time at MEF has not resulted in any significant declines in  $\text{SO}_4^{2-}$  export in streamflow (Urban et al. 2011). Element budgets constructed for S suggest that 42 percent of S is retained in the S2 watershed on an annual basis (Urban et al. 2011).

**Interactions of S and Hg cycling in MEF peatlands—**

Peatlands are sources of total mercury and MeHg to downstream lakes and rivers via export from outlet streams (Grigal et al. 2000; Kolka et al. 2001, 2011a). MeHg, the chemical species of mercury of greatest concern for human health, is produced in wetlands by biologically mediated processes that respond to the availability of the nutrient S in the form of  $\text{SO}_4^{2-}$ . In 2001, a large-scale experiment was initiated to investigate the effects of increased atmospheric  $\text{SO}_4^{2-}$  deposition on rates of methylation of Hg in peatlands (Jeremiason et al. 2006). Sulfate was delivered at four times the average 1990 deposition rate of  $\text{SO}_4^{2-}$  at MEF through a network of PVC pipelines and sprinklers encompassing the downstream half of the S6 bog for 5 years and compared to a control section located at the upstream half of the bog. Percentage of MeHg increased in porewaters of the treatment section of the bog immediately following  $\text{SO}_4^{2-}$  addition and remained elevated. MeHg increased over time in the peat, which served to retain newly formed MeHg (Coleman Wasik et al. 2012). Along with retention of MeHg by peat, MeHg yields in stream water also increased twofold (Jeremiason et al. 2006). Sulfate additions were discontinued in the upstream end of the treatment section after 5 years to create a recovery section. After ending the  $\text{SO}_4^{2-}$  addition, the percentage of MeHg declined in the porewater and peat in the recovery section relative to the treatment section over the 3-year recovery period, but remained higher than the control section (Coleman Wasik et al. 2012). Concentrations of total Hg in mosquito larvae collected in each experimental treatment paralleled MeHg levels in treatment sections, including the recovery section. This work suggested that  $\text{SO}_4^{2-}$  atmospheric deposition drove MeHg production and that lower regional  $\text{SO}_4^{2-}$  emissions and deposition could result in reduced MeHg contamination in aquatic food webs (Coleman Wasik et al. 2012). This relationship implies that past reductions in  $\text{SO}_4^{2-}$  emissions, which were primarily justified to reduce the acidity of precipitation, may have had an added benefit of

decreasing MeHg production in some aquatic systems. If the relationship between  $\text{SO}_4^{2-}$  deposition and MeHg production at MEF were found to be significant across wide geographic regions, it might provide an additional rationale for further reducing  $\text{SO}_4^{2-}$  emissions in the future.

## Other Factors Relevant to Biological Responses to Stream Nutrients Other Than N and P

Interactions among watershed topography, hydrology, and DOC in peatlands were also shown to be important predictors of MeHg production in the presence of  $\text{SO}_4^{2-}$  (Mitchell et al. 2009). A mesocosm study conducted in the  $\text{SO}_4^{2-}$  addition bog showed that added  $\text{SO}_4^{2-}$  resulted in decreased DOC concentrations (Seifert-Monson et al. 2014). Additional research in two bogs at MEF found MeHg hot spots within lagg zones that coincide with inputs of upland-derived  $\text{SO}_4^{2-}$  and DOC (Mitchell et al. 2008b). An experiment examining the effects of  $\text{SO}_4^{2-}$  and organic carbon on MeHg production demonstrated that the highest MeHg concentrations were reached only in the presence of both  $\text{SO}_4^{2-}$  and DOC (Mitchell et al. 2008a) at concentrations that were similar to those in the MeHg hot spots in the bog lagg zones at MEF (Mitchell et al. 2008b).

## Reference Watersheds

Anions and cations, including  $\text{SO}_4^{2-}$ , have been measured at various times in outlet streams of reference watersheds S2 and S5 since the 1960s (e.g., Sebestyen and Verry 2011; Sebestyen et al. 2011a; Urban et al. 1989b, 1995, 2011; Verry and Timmons 1982). Chloride and  $\text{SO}_4^{2-}$  were measured during ice-free months 2010–2012 at an outlet of a bog and fen (Hill et al. 2014). Total mercury, MeHg, and DOC have been measured at various times in S2 and S5 from 1993 to 1995 and during 2005 (Kolka et al. 1999a, 2001; Mitchell et al. 2008c, 2009).

## Cross-Site and Regional Studies

MEF participates in the National Trends Network, which produces maps of long-term records of precipitation chemistry, including  $\text{SO}_4^{2-}$  concentrations across the United States. MEF is part of a study examining litterfall Hg dry deposition in the Eastern United States (Risch et al. 2012). MEF has also participated in the Mercury Deposition Network (MDN) since 1996. The goal of MDN is to monitor regional Hg levels in precipitation. Total Hg concentrations in weekly precipitation are collected at the MEF NADP site and analyzed by Frontier Geosciences. Hg data from the MDN site were included in a study by Monson (2009), which reported a regional decline in Hg concentrations in fish in the 1980s and increases from the 1990s to 2006. The MEF NADP data are available online (NADP 2017).

## Responses to Management and Natural Disturbance

Apart from the  $\text{SO}_4^{2-}$  addition experiment designed to simulate increased atmospheric deposition (Jeremiason et al. 2006), which was described earlier, effects of forest management and natural disturbance on nutrients other than N and P have not been studied at MEF.

## Reliability and Limitations of Findings

The watershed-scale and long-term nature of the  $\text{SO}_4^{2-}$  addition to S6 was atypical of most studies done elsewhere and increased the reliability and applicability of the results. Biological responses to sulfate addition, such as microbial processes, and the potential bioaccumulation of MeHg in aquatic and terrestrial food webs, were not part of the original research plan. The limited mosquito larvae data that were collected after the  $\text{SO}_4^{2-}$  addition had ended demonstrated that measurements of more biological responses might have produced valuable information had they been originally included.

Research findings and datasets on  $\text{SO}_4^{2-}$  and Hg at MEF are likely to represent expected results in other acidic peatlands in the upper Great Lakes states. Outside of settings similar to MEF, however, results should be applied with caution and, where confidence is required, validation studies should be considered to test how well MEF results predict local responses.

## Research Needs

The question that would add the most to the existing body of MEF science related to  $\text{SO}_4^{2-}$  and Hg is: How do various environmental factors influence MeHg availability to peatland biota? Filling this gap is of highest priority to regulators because watersheds differ in their efficiency at converting Hg to MeHg, and understanding what environmental factors influence this process might be useful for developing management strategies to control or reduce MeHg levels in biota. Filling this critical information gap at MEF would require continuing NADP  $\text{SO}_4^{2-}$  and Hg deposition monitoring and initiating long-term monitoring of major food web components, such as microbes, algae, detritus, aquatic invertebrates, and apex predators, including fish, amphibians, spiders, birds, and bats. Long-term monitoring of environmental factors, for example water temperature,  $\text{SO}_4^{2-}$  concentrations, atmospheric concentrations of  $\text{CO}_2$ , and water table changes, to track the patterns of MeHg bioaccumulation in the food webs of peatland streams would also be required. The SPRUCE experiment at MEF, using enclosed mesocosms, also has the potential to contribute additional information about the cycling and mobilization of  $\text{SO}_4^{2-}$ , Hg, and MeHg in peatland soils and porewater subjected to warming and elevated  $\text{CO}_2$  conditions and their effects on relatively immobile food web components such as microbes and macroinvertebrates.

## Potential Utility to Water Quality Regulatory Agencies

Methylmercury continues to be a vexing problem. Insights into how MeHg forms and is processed within peatlands are of value for developing management strategies that could effectively reduce the amounts of Hg available for uptake in the food chain. Current total Hg and  $\text{SO}_4^{2-}$  deposition data being collected at the MEF NADP site will continue to be useful for monitoring and mapping regional Hg and  $\text{SO}_4^{2-}$  levels and trends in precipitation.

Key points:

- MEF research and monitoring have shown that  $\text{SO}_4^{2-}$  addition to peatlands results in increased MeHg production.
- There are hot spots of MeHg production, especially in lagg zones of peatlands related to upland runoff of solutes.
- Limited mosquito larvae data suggest that reducing  $\text{SO}_4^{2-}$  emissions might result in decreased MeHg accumulation in aquatic food webs, with potential reduction of risks to human health.
- The highest priority science gap at MEF related to  $\text{SO}_4^{2-}$ , from the perspective of the regulatory community, is the need for scientific information regarding the effects of atmospheric deposition of  $\text{SO}_4^{2-}$  and other environmental factors such as ambient temperature, atmospheric  $\text{CO}_2$ , and changes in water tables on the bioaccumulation of MeHg in aquatic food webs of northern peatlands. The research and monitoring needed to fill this gap is continued monitoring of  $\text{SO}_4^{2-}$  and Hg deposition as well as further research on environmental factors that influence MeHg availability and bioaccumulation in peatland food webs.

## Overview and Synthesis

Nutrient and hydrologic monitoring have been conducted systematically at multiple MEF watersheds for almost five decades. Seasonal and annual trends in N and P concentrations and hydrology have been documented in reference watersheds as well as nutrient concentration changes that occur as a result of atmospheric deposition, upland clearcutting of aspen, N fertilization, strip and clearcutting of black spruce, prescribed fire in a harvested fen, and the conversion of upland aspen forest to conifer forests. Detailed elemental budgets showed that most of the N and P inputs to MEF bogs and fens are retained, and seasonal variations in nutrient export are associated with snowmelt runoff. Changes in nutrient concentrations in response to land management activities at MEF are usually short lived but can differ between types of peatlands and type of disturbance. While there is a long history of chemical and

hydrological monitoring at MEF, there are no long-term biological data for establishing baseline or reference conditions, as well as understanding how forest management, atmospheric deposition, and other anthropogenic activities influence aquatic and riparian ecosystems. Recent short-term biological studies have suggested that  $\text{NO}_3^-$  concentrations may control algal growth in MEF outlet streams, and localized nutrient availability may be a better predictor of peatland microbial processes in a changing climate than regional climate models. However, biological processes in these peatlands have not been examined at larger spatial, or temporal scales.

MEF's long-term  $\text{SO}_4^{2-}$  concentrations in wet deposition have significantly declined, reflecting the decreases in sulfur dioxide emissions in the upper Midwest. Weekly measurements of total Hg levels in precipitation at MEF have also been critical in tracking declines in Hg deposition in the 1980s followed by increases in more recent years. An 8-year, watershed-scale addition of  $\text{SO}_4^{2-}$  at MEF, an experiment unique among EFRs, documented the close interactions between  $\text{SO}_4^{2-}$  and Hg cycles in northern peatlands. Although biological responses to  $\text{SO}_4^{2-}$  addition were not investigated thoroughly, results indicated that environmental  $\text{SO}_4^{2-}$  concentrations drove MeHg production and suggest that reduction in regional  $\text{SO}_4^{2-}$  emissions and deposition might potentially reduce MeHg bioaccumulation in aquatic food webs.

Despite the wealth of data collected at MEF, significant science gaps regarding biological responses to N, P,  $\text{SO}_4^{2-}$ , and Hg in northern peatlands remain. Science gaps of highest priority for the regulatory community include defining "natural background" or reference response of the biological community, including aquatic microbes, plants, and animals, to the range of ambient nutrient availability found in relatively undisturbed northern peatlands, and what environmental factors affect the cycling of N, P,  $\text{SO}_4^{2-}$ , Hg, and MeHg in northern peatland food webs. A better understanding of these relationships might be useful to regulatory agencies for developing standards for protecting water quality in northern peatlands. Information gaps of importance to regulators might be addressed by continuing long-term monitoring of chemical and physical environmental variables and by initiating new studies of biological responses at reference sites and sites that have undergone significant changes through anthropogenic impacts. Also, experiments such as the  $\text{SO}_4^{2-}$  addition experiment done at S6 and the current SPRUCE study can be extremely useful. Any studies done should consider biological as well as biogeochemical and hydrological responses. It could be of value for the USFS to convene a small group of state and federal scientists to design a program of research and monitoring at MEF that would address pressing science needs of mutual interest to water quality regulatory and land management agencies and propose actions for its implementation.

In the upper Great Lakes region, intact reference sites that provide systematic measurements of natural background conditions in streams and wetlands with minimal onsite human disturbance, are rare. The MEF affords water quality regulators with valuable long-term research results and monitoring datasets on chemistry and hydrology of reference streams for comparisons to similar, but humanly affected streams. This type of information along with results of future studies at MEF could benefit Minnesota regulatory agencies as they implement stream eutrophication standards and work toward applying Tiered Aquatic Life Use standards to streams of northern Minnesota.

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