Abstract Surface waters in forested watersheds in the Adirondack Mountains and northern New York State are susceptible to nitrogen (N) saturation. Atmospheric deposition of N to watersheds in this region has been measured but the extent of internal N inputs from symbiotic N₂ fixation in alder-dominated wetlands is not known. We estimated N₂ fixation by speckled alder in these wetlands by the ¹⁵N natural abundance method and by acetylene reduction using a flow-through system. Foliar N derived from fixation (%Ndfa) was estimated for five wetlands. The δ¹⁵N of speckled alder foliage from four of the five sites did not differ significantly (P ≤ 0.05) from that of nodulated speckled alders grown in N-free water culture (–1.2±0.1‰). Estimates from the ¹⁵N natural abundance method indicated that alders at these sites derive 85–100% of their foliar N from N₂ fixation. At one of the sites, we also measured biomass and N content and estimated that the alder foliage contained 43 kg N ha⁻¹ of fixed N in 1997. This estimate was based on a foliar N content of 55.4±7 kg N ha⁻¹ (mean±SE), 86±4% Ndfa, and an assumption that 10% of foliar N was derived from reserves in woody tissues. At this site, we further estimated via acetylene reduction that 37±10 kg N ha⁻¹ was fixed by speckled alders in 1998. This estimate used the theoretical 4:1 C₂H₂ reduction to N₂ fixation ratio and assumed no night-time fixation late in the season. Nitrogen inputs in wet and dry deposition at this site are approximately 8 kg N ha⁻¹ year⁻¹. We conclude that speckled alder in wetlands of northern New York State relies heavily on N₂ fixation to meet N demands, and symbiotic N₂ fixation in speckled alders adds substantial amounts of N to alder-dominated wetlands in the Adirondack Mountains. These additions may be important for watershed N budgets, where alder-dominated wetlands occupy a large proportion of watershed area.

Keywords Actinorhizal plants · Alnus · Nitrogen fixation · Adirondack Mountains

Introduction

The actinorhizal shrub Alnus incana ssp. rugosa (speckled alder) forms a root nodule symbiosis with N₂-fixing actinomycetes of the genus Frankia, and dominates some shrub and riparian wetlands of northeast North America (Furlow 1979). Speckled alder is the dominant species of the second largest wetland covertype [Scrub-Shrub 1 (Cowardin et al. 1979)] in the Adirondack Mountains (Roy et al. 1996), a region with surface waters that are susceptible to N saturation or excess N (Stoddard 1994). Nevertheless, N₂ fixation in speckled alder has not been quantified in Adirondack wetlands.

A wide range of N₂ fixation has been reported for speckled alder in other ecosystems. Voigt and Stueckel (1969) and Daly (1966) measured N accretion of 85–167 kg N ha⁻¹ year⁻¹ in speckled alder stands, indicating high rates of N₂ fixation. In contrast, low rates of fixation have been estimated in speckled alder in pure stands (5 kg N ha⁻¹ year⁻¹) or mixed stands with Populus tremuloides (1 kg N ha⁻¹ year⁻¹) in northern Wisconsin (Younger and Kapustka 1983).

There are two practical ways of estimating N₂ fixation in woody plants in the field: ¹⁵N natural abundance and acetylene reduction. Like all estimates of N₂ fixation in the field, they are accompanied by high levels of uncertainty (Winship and Tjepkema 1990; Binkley et al. 1994). The success of ¹⁵N natural abundance to estimate
tissue N derived from fixation (Shearer and Kohl 1993) relies on significant differences in the $^{15}$N of soil N, incorporated into tissues of non-fixing reference plants, and N$_2$ incorporated in tissues of fixing plants grown without other N sources. The method is also susceptible to error based on characteristics of non-N$_2$-fixing reference plants (Högberg 1997). The acetylene reduction assay estimates nitrogenase activity over short time periods, and so must be repeated to capture seasonal and diurnal patterns. Acetylene reduction involves reduction of C$_2$H$_2$ to C$_2$H$_4$ by nitrogenase, and so requires a conversion ratio for acetylene reduction to N$_2$ fixation. This ratio may be determined experimentally using $^{15}$N$_2$, but more often is assumed based on the minimum theoretical value of 4:1 (Schwintzer and Tjepkema 1994). Because speckled alder nodules exhibit a pronounced acetylene-induced decline, the initial peak rate is the only reliable measure of nitrogenase activity, and this can only be measured in an open flow-through system (Schwintzer and Tjepkema 1997).

Fixation of N$_2$ by alders, as well as by other actinorhizal plants such as Myrica gale (Schwintzer 1983), may be an important N input to wetlands and forested watersheds of the Adirondack Mountains and other watersheds of eastern North America. Accounting for natural N inputs in wetland areas is critical if we are to attempt to predict effects of anthropogenically derived N on sensitive ecosystems, particularly if riparian or wetland ecotones are considered control points for watershed N transport. Watershed-level predictions based on up-slope forest characteristics and downstream chemistry (e.g., time to or stage of N saturation, and wetland influence on watershed N retention) could be altered substantially by speckled alder wetlands juxtaposed between these points, where alders may add N through fixation, accelerate nitrification processes (Van Miegroet and Cole 1984), and use little soil-derived N (Mead and Preston 1992).

Symbiotic N$_2$ fixation in alders of areas such as the Adirondack Mountains, which receive elevated N in atmospheric deposition, may be reduced if inorganic N accumulates to high soil concentrations. Symbiotic N$_2$ fixation decreases in Alnus spp. subjected experimental- ly to high concentrations of N (Huss-Danell and Hahlin 1988). However, small, incremental N additions may maintain or even stimulate fixation in Alnus spp. (Stewart and Bond 1961; Ingestad 1980; Mackay et al. 1987), and the effects of inorganic N on fixation in Alnus in the field are less clear (Binkley et al. 1994).

This study provides the first field estimates for symbiotic N$_2$ fixation in speckled alder of the Adirondack Mountains. The objectives of this study were to estimate: (1) the fraction of alder foliage N derived from fixation in five alder scrub-shrub wetlands of northern New York State, a region where elevated N from atmospheric deposition might be expected to reduce the dependence of N$_2$-fixing plants on atmospheric N$_2$, and (2) rates of symbiotic N$_2$ fixation in a alder shrub wetland representative of the second largest wetland type in major drainage basins of the Adirondack Mountains (Roy et al. 1996).

Materials and methods

Site locations

Natural abundance $^{15}$N dilution

Five alder wetlands were selected to estimate the contribution of atmospheric N$_2$ to foliar N in alder shrubs by the $^{15}$N natural abundance method (Table 1). Three wetlands were on the Huntington Wildlife Forest in the central Adirondacks, one of which was used for the acetylene reduction assay (Table 1). The Huntington Wildlife Forest is a National Atmospheric Deposition Program and National Trends Network (NADP/NTN) monitoring site (NY20), and has been the locus of many biogeochemical studies (Raynal et al. 1985; Johnson and Lindberg 1992; Mitchell et al. 1994, 1996). Soils, surficial and bedrock geology at Huntington Wildlife Forest are typical of the Adirondack region and are described in Ohrui et al. (1999). Mean annual temperature is 4.4°C, with a dormant season mean of –2.8°C and a growing season mean of 14.3°C. Mean annual precipitation is 101 cm (Shepard et al. 1989). Vegetation on the upper slopes is mixed northern hardwood forest. The lower slopes are characterized by eastern hemlock [Tsuga canadensis (L.) Carr.], red spruce [Picea rubens Sarg.], balsam fir [Abies balsamea (L.) Miller] and yellow birch [Betula alleghaniensis]. Nitrogen inputs in wet deposition at Huntington Wildlife Forest are approximately 5 kg N ha$^{-1}$ year$^{-1}$ [National Atmospheric Deposition Program (NRSP-3)/National Trends Network 1999], with an additional 3 kg N ha$^{-1}$ year$^{-1}$ added in dry deposition (Shepard et al. 1989).

Additional wetlands studied were located near Blue Mountain in the central Adirondacks, and near Altmar, Oswego County, N.Y., between Lake Ontario and the Tug Hill Plateau (Table 1). The latter site is within 8 km of the NADP/NTN Bennett Bridge monitoring site (NY52), which records the greatest N in wet deposition (approximately 10 kg N ha$^{-1}$ year$^{-1}$) of any station in the national network [National Atmospheric Deposition Program (NRSP-3)/National Trends Network 1999].

Biomass estimation and the acetylene reduction assay

Biomass estimates for alder and the acetylene reduction assay were conducted in a 4-ha, riparian wetland dominated by speckled alder along Fishing Brook, at the Huntington Wildlife Forest (Table 1). This wetland is classified as Scrub-Shrub 1-Emergent 1 (SS1/EM1; Cowardin et al. 1979). The range in stem diameters of alders at Fishing Brook, measured 25 cm from the ground surface, was 0.5–12.1 cm, and the estimated stem density was 19,120±4,723 stems ha$^{-1}$ (mean±SE, n=10) within the SS1 component of the wetland.

Biomass estimates

In July 1997, the biomass of above- and below-ground tissues of speckled alder was estimated at ten, randomly located points at Fishing Brook. At each point, stems >0.5 cm diameter were measured in 5×5 m plots. Foliar biomass, above-ground woody biomass, and annual increment were estimated from these measurements using allometric equations developed at Huntington Forest (Bischoff et al., in press; Hurd 1999). Roots and nodules were excavated along with soil to a depth of 25 cm in 0.5×0.5 m plots located 3 m away from large plots. No nodules or fine roots occurred below this depth due to the shallow depth of the watertable. Roots and nodules were washed, and hand sorted into root size classes (0–0.5, 0.5–3.0, >3.0 mm diameter), and living (firm, yellow) or dead (soft, brown) nodules, dried at 65°C, and weighed.