vol. 174, no. 4 the american naturalist october 2009

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Facultative versus Obligate Nitrogen Fixation Strategies and Their Ecosystem Consequences

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Submitted January 23, 2009; Accepted May 14, 2009; Electronically published August 20, 2009

Online enhancement: appendix.

abstract: Symbiotic nitrogen (N) fixers are critical components of many terrestrial ecosystems. There is evidence that some N fixers fix N at the same rate regardless of environmental conditions (a strategy we call obligate), while others adjust N fixation to meet their needs (a strategy we call facultative). Although these strategies are likely to have qualitatively different impacts on their environment, the relative effectiveness and ecosystem-level impacts of each strategy have not been explored. Using a simple mathematical model, we determine the best facultative strategy and show that it excludes any obligate strategy (fixer or nonfixer) in our basic model. To provide an explanation for the existence of nonfixers and obligate fixers, we show that both costs of being facultative and time lags inherent in the process of N fixation can select against facultative N fixers and also produce the seemingly paradoxical patterns of sustained N limitation and N richness. Finally, we speculate on why the costs and lags may differ between temperate and tropical regions and thus whether they can explain patterns in both biomes simultaneously.

Keywords: nitrogen fixation, nitrogen limitation, model, evolutionary ecology.

Introduction

Symbioses between certain angiosperm species and nitrogen (N)-fixing bacteria (hereafter, we refer to these symbioses and the plants themselves as N fixers) play a unique and critical role in many terrestrial ecosystems. They can be by far the largest natural N source, bringing in more than 50 kg N ha^{-1} $year^{-1}$ in some ecosystems (Binkley et al. 1992; Uliassi and Ruess 2002), which can facilitate N-limited competitors (and thus succession), speed up the development of nutrient cycles, and increase primary production. Their activity, or lack thereof, likely plays a crucial role in two mysteries in ecosystem ecology. The chronic N limitation that pervades mature temperate and boreal forests could easily be overcome by N fixers, who are conspicuous in these ecosystems in their absence only (Vitousek and Howarth 1991; Vitousek and Field 1999; Rastetter et al. 2001; Vitousek et al. 2002; Menge et al. 2008). In contrast, chronic N richness in many tropical forests may result from biological N fixation (BNF) by leguminous trees, which are ubiquitous in the tropics, but the potential reasons for fixing more than is necessary (overfixation) are at present unclear (Jenny 1950; Vitousek et al. 2002; Hedin et al. 2003; Barron 2007).

Nitrogen fixers are the only ecosystem components that have the capacity to regulate N inputs on the basis of soil N availability (an index of ecosystem-level N demand), and this regulation likely has important implications for the intriguing patterns of N limitation and N richness. However, the extent to which they regulate N inputs and the resulting effects on N limitation and N richness depend on their BNF strategy. We consider two broad strategy classes, obligate and facultative N fixers. By our definition, obligate types fix N at the same rate per unit of biomass regardless of their environment—and thus can regulate N inputs only via changes in their biomass—whereas facultative types adjust BNF per unit of biomass in response to environmental conditions. In the mutualism literature, these strategies are termed "fixed" or "nonconditional" (our "obligate") and "context dependent" or "conditional" (our "facultative"; Bronstein 1994; Heath and Tiffin 2007). Our definition of "obligate" specifies a constant rate because it is more tractable and provides a better comparison for our study.

Although conclusive field tests of the BNF strategy employed by different N fixers are lacking, there is evidence that some are obligate and some are facultative. In many temperate and boreal forests, actinorhizal N fixers (nonleguminous plants that form symbioses with actinomycete bacteria; Huss-Danell 1997) dominate early to midsuccessional habitats before being excluded by nonfixers (Wardle 1980; Binkley et al. 1992; Walker 1993; Chapin et al. 1994; D. N. L. Menge, J. L. DeNoyer, and J. W. Lichstein, unpublished manuscript), and the limited evidence sug-

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Am. Nat. 2009. Vol. 174, pp. 465–477. © 2009 by The University of Chicago. 0003-0147/2009/17404-51025$15.00. All rights reserved.
DOI: 10.1086/605377

gests that they are obligate N fixers. In Oregon and Washington, BNF rates by *Alnus rubra* (red alder) from 50-year-old to 85-year-old sites exceeded average N accretion rates, despite large losses of plant-available N in streams (Binkley et al. 1992). In Alaska, *Alnus tenuifolia* (thin-leaf alder) BNF rates per basal area were nearly identical in early succession and midsuccession, despite lower light availability in the older sites (and thus, presumably, less energy to spend on BNF; Uliassi and Ruess 2002). In New Zealand, *Coriaria arborea* (tutu) fixed N at the same rate (per tutu basal area and per ground area) in 7-year-old and 60-year-old sites (Menge and Hedin 2009), despite the fact that N availability at these two sites spanned the full range across the 120,000-year soil chronosequence (Richardson et al. 2004; Menge and Hedin 2009). Although by no means conclusive, these studies suggest that, at least across the range of conditions seen at these sites, BNF by these actinorhizal N fixers is obligate.

In contrast to temperate and boreal forests, tropical forests are commonly dominated by leguminous trees (many of which form symbioses with the N-fixing bacteria rhizobia; Vitousek et al. 2002), and the limited evidence supports the idea that these legumes are facultative N fixers. In a plantation in Hawaii, *Acacia koa* (koa) fixed less N as soil N availability increased with forest age (Pearson and Vitousek 2001). In Panama, species in the genus *Inga* fixed much more N in gaps and in disturbed forests than in mature forests, the last of which had higher N availability (Barron 2007).

Here we present a simple model of plant growth and BNF in an ecosystem context to explore the relative competitive ability of different BNF strategies under different conditions and the potential importance of these disparate strategies for the fundamental ecosystem-level patterns of N limitation and N richness. With this model we investigate the following questions: (i) Which obligate and facultative N fixers are the best competitors in different environments? (ii) How do obligate and facultative N fixers compare against each other? (iii) How do costs of being facultative affect the answer to ii? (iv) How do time lags inherent in the process of BNF affect the answer to ii? and (v) How do the different strategies influence N losses, a key index of both N limitation and N richness? In the "Discussion" we address which types of N fixers could produce the various ecosystem-level patterns and combine this with our strategy comparison results to make testable predictions about different N fixers in different ecosystems.

Model and Analysis

Model Description

The model we use here builds on our previous work (Menge et al. 2008, 2009). The full model includes biomass B_i (in units of mass or carbon per area) of different N-fixer types i (with BNF rates F_i), organically bound N (D_N) and another nutrient (D_R, where R is a generic resource, which could be phosphorus, P, or any other soil-derived resource) in soil detritus, and the plant-available forms of N (A_N) and the other resource (A_R; fig. 1).

The model works for an arbitrary number of N-fixer types, but we will generally consider only one or two in our analyses. The growth rate for each type, $g_i(A_N, A_R)$, depends on acquisition of the two resources (through uptake from the soil, $\nu_j(A_j)$, and/or BNF, F_i) and the nutrient use efficiencies (NUEs), ω_j (equivalent to the biomass-to-nutrient or carbon-to-nutrient ratios of litter; Vitousek 1982). Growth is governed by Liebig's law of the minimum function (e.g., Von Liebig 1840; Tilman 1982), so it can be limited by a single nutrient or by both at certain points. Plant turnover occurs at a base rate, μ, and is augmented by increasing BNF. The costs of BNF, θ_g and θ_μ, are implemented as a decrease in the growth rate and an increase in the turnover rate, respectively (both proportional to the rate of BNF). This formulation reflects trade-offs between BNF and nutrient uptake, NUE, and turnover (similar to Menge et al. 2008).

The model satisfies the equations

$$\frac{dB_i}{dt} = B_i[g_i(A_N, A_R) - (\mu + \theta_\mu F_i)], \tag{1}$$

$$\frac{dD_N}{dt} = \sum_i \frac{(\mu + \theta_\mu F_i)B_i}{\omega_N} - m_N(D_N) - \phi_N(D_N), \tag{2}$$

$$\frac{dD_R}{dt} = \sum_i \frac{(\mu + \theta_\mu F_i)B_i}{\omega_R} - m_R(D_R) - \phi_R(D_R), \tag{3}$$

$$\frac{dA_N}{dt} = I_N - k_N(A_N) + m_N(D_N) - \sum_i \frac{B_i}{\omega_N}[g_i(A_N, A_R) - \omega_N F_i], \tag{4}$$

$$\frac{dA_R}{dt} = I_R - k_R(A_R) + m_R(D_R) - \sum_i \frac{B_i}{\omega_R} g_i(A_N, A_R), \tag{5}$$

$$g_i(A_N, A_R) = \min\{\omega_N[\nu_N(A_N) + F_i], \omega_R\nu_R(A_R)\} - \theta_g F_i. \tag{6}$$

In addition to the variables and parameters described above, $m_j(D_j)$ are the mineralization functions, $\phi_j(D_j)$ are the organic loss functions, I_j are the abiotic nutrient inputs, and $k_j(A_j)$ are the available nutrient loss functions (see

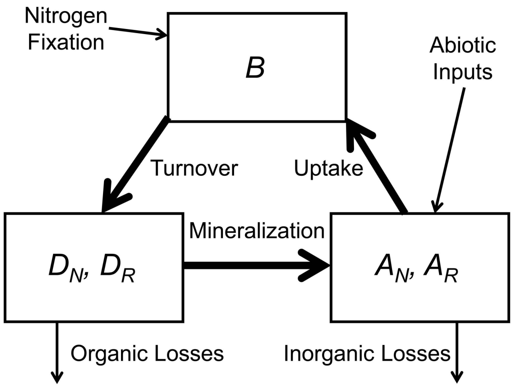

Figure 1: Model system described in equations (1)–(5). Boxes are the nutrient pools in the system: living plant biomass (B), plant-unavailable soil nutrients (D_N, D_R), and plant-available soil nutrients (A_N, A_R), where R is a potentially limiting nutrient other than N. Within-system fluxes include plant uptake, turnover, and net mineralization. The fluxes into and out of the model ecosystem are biological N fixation, abiotic inputs to the available pools, losses of plant-unavailable nutrients, and losses of plant-available nutrients.

table 1 for a list of variables, parameters, and strategies). We assume that the uptake, mineralization, and loss functions equal 0 when their arguments are 0 and increase monotonically. For all BNF strategies, F_i, we specify bounds for biological realism. BNF cannot be negative or exceed a maximum physiological rate. Within these bounds, F is constant for obligate BNF strategies but depends on state variables in the system for facultative BNF strategies.

From equation (6), the BNF rate that makes plant growth colimited by the two resources at any point in time, which we call F_C, is

$$F_C = \frac{\omega_R}{\omega_N} \nu_R(A_R) - \nu_N(A_N). \tag{7}$$

The quantity F_C increases with A_R and decreases with A_N. The exact shape depends on the shape of the nutrient uptake functions, $\nu_j(A_j)$. It is also the dividing point between N limitation and R limitation: when $F_i < F_C$, B_i is not fixing enough N, so it is N limited at that point in time. When $F_i > F_C$, B_i is overfixing and R limited. The quantity F_C is the same for all N fixers or nonfixers because

Table 1: Variables, functions, parameters, and strategies

Symbol	Definition	Units	Equation
B_i	Plant biomass of type i	kg C ha^{-1}	(1)
D_j	Plant-unavailable soil nutrients	kg j ha^{-1}	(2), (3)
A_j	Plant-available soil nutrients	kg j ha^{-1}	(4), (5)
$g_i(A_N, A_R)$	Growth function	year^{-1}	(6)
ω_j	Nutrient use efficiencies	kg C kg j^{-1}	
$\nu_j(A_j)$	Nutrient uptake functions	kg j kg C^{-1} year^{-1}	
μ	Biomass turnover rate	year^{-1}	
θ_g	Growth cost of BNF	kg C kg N^{-1}	
θ_μ	Turnover cost of BNF	kg C kg N^{-1}	
$\theta_{g,F}$	Per-BNF growth cost of being facultative	kg C kg N^{-1}	(10)
$\theta_{\mu,F}$	Per-BNF turnover cost of being facultative	kg C kg N^{-1}	(10)
$m_j(D_j)$	Net mineralization functions	kg j ha^{-1} year^{-1}	
$\phi_j(D_j)$	Plant-unavailable nutrient loss functions	kg j ha^{-1} year^{-1}	
I_j	Nonfixation nutrient input fluxes	kg j ha^{-1} year^{-1}	
$k_j(A_j)$	Plant-available nutrient loss functions	kg j ha^{-1} year^{-1}	
γ_g	Fixed growth cost of being facultative	year^{-1}	(10)
γ_μ	Fixed turnover cost of being facultative	year^{-1}	(10)
F_i	BNF rate under strategy i (per biomass C)	kg N kg C^{-1} year^{-1}	
F_C	F_i that gives colimitation (var)	kg N kg C^{-1} year^{-1}	(7)
$\overline{F}_C$	F_C at equilibrium (const)	kg N kg C^{-1} year^{-1}	
F_O	Generic obligate strategy (const)	kg N kg C^{-1} year^{-1}	
F_{max}	Maximum possible F_i (const)	kg N kg C^{-1} year^{-1}	
$\hat{F}_O$	Obligate CSS: bounded $\overline{F}_C$ (const)	kg N kg C^{-1} year^{-1}	(8)
F_F	Best facultative F_i (var)	kg N kg C^{-1} year^{-1}	(9)
$F_{O,max}$	Obligate $F_O = F_{max}$ (const)	kg N kg C^{-1} year^{-1}	
F_L	Time lag constrained facultative F_i (var)	kg N kg C^{-1} year^{-1}	(11)–(13)

Note: BNF, biological N fixation; CSS, continuously stable strategy; i refers to a strategy type (and thus a plant type), whereas j refers to a nutrient (N or R); "var" means F depends on A_N and A_R, whereas "const" means it is a constant.

all types have access to the same resource pools and because we assumed that stoichiometries and uptake rates do not differ between types (we assumed this to isolate the effects of different strategies from other potential differences between types). We will also refer to $\overline{F}_C$, the constant BNF rate that gives colimitation at equilibrium (F_C evaluated at $\overline{A}_N$, $\overline{A}_R$). Mathematically, both F_C and $\overline{F}_C$ can be negative or arbitrarily high, which is biologically unrealistic. Therefore, F_C and $\overline{F}_C$ can be BNF strategies only when modified to remain within biologically sensible bounds (table 1).

Model Analysis

We use a combination of techniques to examine how the different BNF strategies compare against each other. To compare obligate BNF strategies against each other in an equilibrium environment, we search for evolutionarily stable (cannot be invaded once established), convergence stable (will be evolutionarily approached from anywhere), and continuously stable (both evolutionarily and convergence stable) strategies (Eshel 1983; Geritz et al. 1997; Levin and Muller-Landau 2000). These techniques from adaptive dynamics (Geritz et al. 1998) assume that a strategy that is constant over ecological time (like obligate BNF) can change over evolutionary time and that ecological systems come to equilibrium between evolutionary events (e.g., mutations in the population). When these assumptions are met, these are powerful analytical techniques, allowing us to determine which types will evolve and outcompete other types.

However, these assumptions are not always met. Biogeochemical systems can take a long time to equilibrate (Walker and Syers 1976; Vitousek 2004; Menge et al. 2009), and we are interested in the dynamics of different BNF strategies in transient environments as well as equilibrium environments. To compare different types against each other in transient environments, we first compute the relative growth rates (RGRs), $(1/B_i)(dB_i/dt)$. This technique is less powerful than adaptive dynamics but has many fewer assumptions (notably, it is relevant everywhere rather than only at equilibrium). In certain conditions, comparing RGRs is quite powerful: if one type always has a higher RGR than another type, it will eventually drive the other type to extinction if they are limited by the same nutrient (including one type being colimited). However, if the rank of RGRs differs under different environmental conditions, there is no clear winner in fluctuating environments. To illustrate possible outcomes in these unclear cases and complement the analytical results, we simulate the system with Matlab's ode45 and dde23 functions, using realistic parameter values (parameterizing R as phosphorus) and starting conditions (found in the appendix in the online edition of the *American Naturalist*).

To examine the environmental impacts of different BNF strategies, we determine how available N loss rates depend on BNF strategies, using equilibrium and quasi-equilibrium analyses as well as simulations. Equilibrium loss rates are easy to calculate, depend only on parameters in the system, and would be observed in ecosystems that have been undisturbed for at least many centuries, such as mature forests without fire, large storms, or other large-scale disturbances (Menge et al. 2009). However, many ecosystems have much shorter disturbance return intervals, so to investigate loss rates in these ecosystems we calculate short timescale quasi-equilibrium loss rates (as in Menge et al. 2009). The quasi-equilibrium analysis assumes that available nutrients in the soil track biomass and soil organic matter, which happens within hours to weeks (Jackson et al. 1989; Schimel et al. 1989; Perakis and Hedin 2001; Providoli et al. 2006) and thus is relevant for ecosystems that have been undisturbed for this shorter period. However, unlike equilibrium analyses, quasi-equilibrium analyses depend on the current state of biomass and organically bound soil nutrients and thus require different information. As with the competition analysis, we complement the analytical loss results by simulating the system to illustrate numerically the effects of different BNF strategies on plant-available N losses.

Results

If the costs of BNF ($\theta_g + \theta_\mu = \theta$) exceed the benefits ($\omega_N$), there is obviously no advantage to BNF, and obligate nonfixers always exclude any N fixer. Because our purpose here is to compare different types of N fixers, we hereafter assume that $\omega_N > \theta$.

Obligate BNF

In an equilibrium environment, the continuously stable strategy (CSS) for obligate N fixers is the constant BNF rate that yields colimitation at equilibrium when possible,

$$\hat{F}_O = \min(\overline{F}_C^+, F_{\max}). \tag{8}$$

Here, $\overline{F}_C^+$ indicates $\overline{F}_C$ if it is positive and 0 if it is not. The CSS $\hat{F}_O$ may range from nonfixation (0) to the maximal BNF rate ($F_{\max}$). Following the definition of CSS, this type will be approached evolutionarily from any starting point and will not be invaded (see the appendix for details of the CSS analysis).

Although this is a clear answer for equilibrium environments, terrestrial ecosystem nutrient cycles take a long time to approach equilibrium—thousands of years for N (Menge et al. 2009) and possibly much longer for other nutrients such as P (Walker and Syers 1976; Vitousek 2004)—and in many locations disturbances occur frequently, relative to this slow timescale. Therefore, an N fixer with strategy $\hat{F}_O$ would rarely experience the environment for which it is best suited. In this case the best obligate strategy would depend on the frequency and magnitude of disturbance and its effects on N and the other resource.

As an example of a dynamic case, figure 2*A* and 2*B* shows a simulation of primary succession with an obligate N fixer (F_{max}; fig. 2*A*, *solid line*) and a nonfixer (fig. 2*A*, *dashed line*) in competition. A nonfixer in this model is an obligate type with $F = 0$. Initially, there is no N in soil, N is strongly limiting (as indicated by the minimal losses of available N in fig. 2*B*), and the type with the highest RGR is F_{max}. After 10 years of rapid growth, enough N has been fixed to overcome N limitation. At this point, available N losses rapidly increase, and the nonfixer has the highest RGR. Nonfixer biomass approaches and, after 50 years, overtakes the fixer's (F_{max}). Nitrogen ceases to leak out in large quantities after about 85 years (fig. 2*B*). Therefore, if an ecosystem with these parameter values has disturbances that cause large N losses frequently enough, obligate N fixers and nonfixers can coexist.

Facultative BNF

In a dynamic environment, the facultative N fixer with the highest RGR is the one that fixes just enough N to be colimited, as seen via the derivatives of the RGR with respect to F: $\omega_N - \theta$ when N limited (a positive derivative; increasing BNF increases the RGR) and $-\theta$ when R limited (which is negative; decreasing BNF increases the RGR). Taking the bounds of BNF into account, the best facultative strategy is therefore the strategy in which the N fixer instantaneously adjusts its BNF rate to meet but not exceed its N demand:

$$F_F = \min(F_C^+, F_{max}). \tag{9}$$

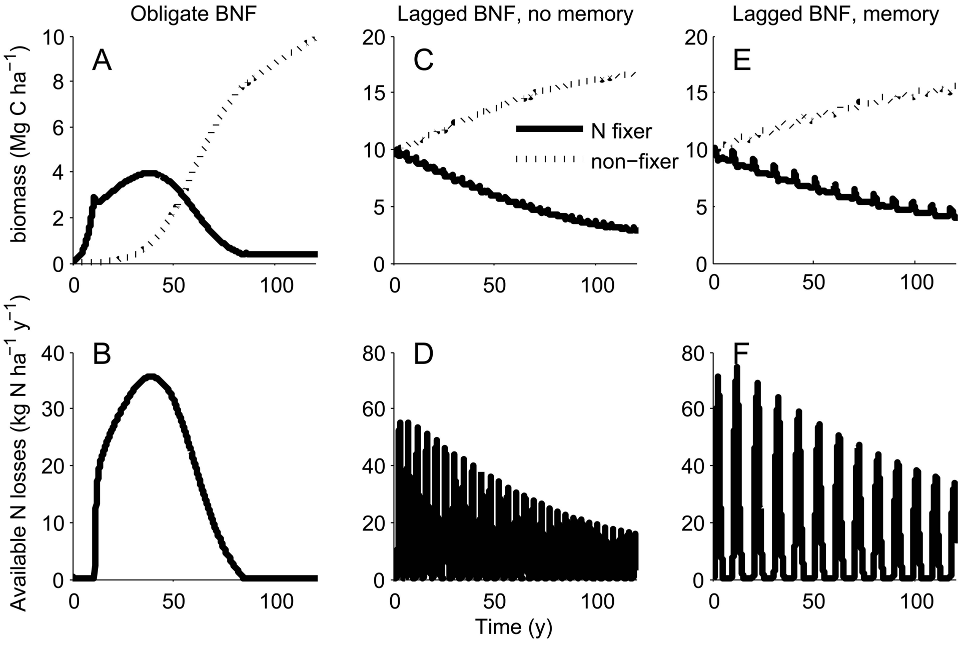

Figure 2: Plant biomass (*A*, *C*, *E*) and available N losses (*B*, *D*, *F*) during competition between a nonfixer and an obligate N fixer (*A*, *B*), a time-lagged facultative N fixer without memory (*C*, *D*), and a time-lagged facultative N fixer with memory (*E*, *F*). The time lag in *C*–*F* is 1 year, although lags of less than 2 days (with no memory) or 3 months (with memory) produce qualitatively similar results with the parameters used here (found in the appendix in the online edition of the *American Naturalist*). *A*, *B*, The obligate N fixer F_{max} dominates early primary succession, leading to an abundance of available N. Nitrogen ceases to limit growth at about 10 years (when N begins to leak), at which point the nonfixer's relative growth rate exceeds the fixer's. After 50 years the fixer is overtaken by the nonfixer. The nonfixer continues to dominate after N becomes limiting again around 85 years. *C*–*F*, Because of the time lag, N fixers overfix after N ceases to limit growth and do not begin fixing until well after N begins to limit growth, causing oscillations with large N losses and an overall lower growth rate than that of the nonfixer. The nonfixer outcompetes both lagged N fixers by not paying the cost of fixing N when it is not limiting. The capacity to integrate past information helps the lagged N fixer, but with a sufficiently large time lag it is still outcompeted by the nonfixer. BNF = biological N fixation.

Obligate versus Facultative BNF

The RGR of the best facultative N fixer (F_F) matches or exceeds that of any obligate N fixer, as shown in the appendix. In this simple model, a facultative N fixer can invade and exclude any obligate N fixer, but some N fixers in real ecosystems seem to be obligate. Why might obligate N fixers exist? One option is that they have not evolved the capability of being facultative, but this answer is hardly satisfying. In the next two sections, we examine two physiological mechanisms that could give obligate types an advantage: fixed costs of being facultative and time lags inherent to BNF.

Costs of Being Facultative

To adjust BNF to local environmental conditions, it is necessary to respond to the environment (or some index thereof, such as internal nutrient stores in a plant) and to have the physiological machinery to increase or decrease the rate of BNF. Such infrastructure may carry a cost (van Kleunen and Fischer 2005), which would result in a fixed cost that decreases the growth rate (or increases the turnover rate or both) regardless of the amount of BNF. It is also possible that the per-unit cost of BNF is higher for facultative N fixers. For example, changing the BNF rate could involve the creation/destruction of nodules or the activation/deactivation of symbionts, which may carry a greater cost than for obligate N fixers that keep a steady nodule density or activity level. Here we examine how these costs of being facultative, which could arise from these mechanisms or others, influence competition between a facultative N fixer and an obligate N fixer.

Let there be fixed and variable costs of being facultative that decrease the growth rate (γ_g and $\theta_{g,\mathrm{F}}$) and increase the turnover rate (γ_μ and $\theta_{\mu,\mathrm{F}}$). Defining $\gamma \equiv \gamma_g + \gamma_\mu$ and $\theta_F \equiv \theta_{g,\mathrm{F}} + \theta_{\mu,\mathrm{F}}$ (i.e., combining the growth and mortality costs, which have identical effects on the analyses we present here), the rate of change of B_F is now

$$\frac{dB_F}{dt} = B_F(\min\{\omega_N[\nu_N(A_N) + F_F], \omega_R\nu_R(A_R)\} - \mu - (\theta + \theta_F)F_F - \gamma). \tag{10}$$

Incorporating costs of being facultative yields an environmental region where any obligate N fixer, F_O, has a higher RGR (fig. 3, *solid lines*) and can also yield regions where F_F has a higher RGR (fig. 3, *hatched lines*). The *X*-axis of figure 3 is F_C (eq. [7]), which depends on A_N and A_R, determines whether the obligate N fixer (a fixed point on the axis) is N limited or R limited, and gives the BNF rate of F_F (as long as it is between 0 and $F_{\max}$). When the costs of being facultative exist but are relatively small, an intermediate F_O has a higher RGR in environments in which it is colimited or close to colimitation (between $F_C = [F_O - (\gamma/\theta)][\theta/(\theta + \theta_F)]$ and $F_C = \{F_O + [\gamma/(\omega_N - \theta)]\}[(\omega_N - \theta)/(\omega_N - \theta - \theta_F)]$), but F_F has a higher RGR outside this region, when F_O is farther from colimitation (fig. 3*A*). An obligate nonfixer grows faster whenever both it and F_F are R limited or colimited or when F_O is mildly N limited (when $F_C < \gamma/(\omega_N - \theta - \theta_F)$; fig. 3*B*). Conversely, $F_{O,\max}$ grows faster when both it and F_F are N limited or colimited or when $F_{O,\max}$ is mildly R limited (when $F_C > [F_{\max} - (\gamma/\theta)][\theta/(\theta + \theta_F)]$; fig. 3*C*). More time spent away from F_O is better for the facultative N fixer, but ironically, F_F might bring F_C toward F_O (e.g., if approaching equilibrium and $F_O = \hat{F}_O$).

If the costs of being facultative are sufficiently high, facultative N fixers cannot exist regardless of the environmental conditions. Specifically, if $\gamma > F_O\theta$ and $\gamma + \theta_F F_{\max} > (F_{\max} - F_O)(\omega_N - \theta)$, any F_O will always exclude F_F (fig. 3*D*). If the only possible obligate strategies are nonfixers and $F_{O,\max}$, nonfixers will always exclude any facultative N fixer when $\gamma + \theta_F F_{\max} > F_{\max}(\omega_N - \theta)$ (fig. 3*E*), and $F_{O,\max}$ will always exclude any facultative N fixers when $\gamma > F_{\max}\theta$ (fig. 3*F*). If the nonfixer and $F_{O,\max}$ can coexist, they exclude the facultative N fixer under the same conditions as in figure 3*D*. Furthermore, even if the costs of being facultative are relatively low, as in figure 3*A*, a coalition of obligate types could exclude F_F if the obligate types can coexist (fig. 3*G*).

Time Lags Inherent to BNF

Even without direct costs of being facultative, facultative N fixers may be at a disadvantage because of time lags in the process of fixing N. In the analysis above, we assumed that facultative N fixers adjust their BNF rate to meet their needs instantaneously, relative to other changes in the system. However, available nutrient pools in the soil change very rapidly (Jackson et al. 1989; Schimel et al. 1989; Perakis and Hedin 2001; Providoli et al. 2006), and altering the rate of BNF—which involves either building/shedding root nodules or altering carbon or oxygen flow to the nodules—may take an appreciable amount of time, relative to the changes in available nutrients.

To incorporate a time lag into the model, we include a constant T that represents the time it takes to up- or downregulate BNF. To our knowledge, the magnitude of T in real N fixers is unknown, but in this model we can vary T from nonexistent to arbitrarily large to investigate how different time lags affect facultative N fixers' competitive abilities and ecosystem-level consequences. With the time lag, the plant's BNF at the current time, t, depends on the action it put in motion at time $t - T$. We assume that the plant fixes N on the basis of the environmental

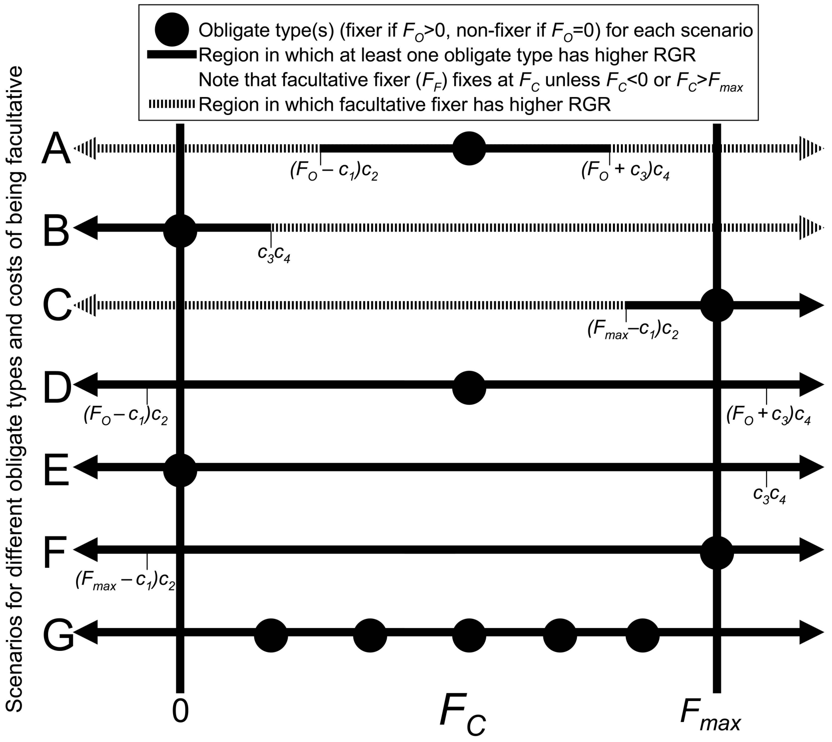

Figure 3: Effects of costs of being facultative on relative growth rates (RGRs) of obligate versus facultative types (i.e., N fixers or nonfixers). The *X*-axis is F_C (eq. [7]), the amount of fixation needed to be colimited, which changes with the environment: it decreases as soil N increases and increases as the other resource (R) increases. When $F_i < F_C$, B_i is underfixing and N limited, whereas when $F_i > F_C$, it is overfixing and R limited. The quantity F_C can be negative or above F_{max}, but F_i cannot, so the facultative N fixer we consider, F_F, equals F_C where possible but is bounded by 0 and F_{max}. Obligate N fixers, F_O, are constant points on the F_C axis. Solid lines indicate a higher RGR for F_O, whereas hatched lines indicate a higher RGR for F_F. The constants, which indicate the cost of being facultative, are $c_1 = \gamma/\theta$, $c_2 = \theta/(\theta + \theta_F)$, $c_3 = \gamma/(\omega_N - \theta)$, and $c_4 = (\omega_N - \theta)/(\omega_N - \theta - \theta_F)$. When F_F pays a fixed cost γ or a variable cost θ_F of being facultative, it can have a higher RGR when F_C is sufficiently far from F_O (*A–C*), but if γ or θ_F are sufficiently high relative to the net gain of fixing N, F_F always has a lower RGR (*D–G*). For instance, in *B*, the obligate type is a nonfixer. If soil N and R availability are such that the nonfixer would be R limited ($F_C < 0$), the nonfixer has a higher RGR because, although neither it nor the facultative type is fixing, the facultative type pays a cost to be facultative. If soil N and R availability are such that the nonfixer is weakly N limited relative to the cost of being facultative ($0 < F_C < c_3c_4$), the nonfixer still has a higher RGR, but if the nonfixer is strongly N limited relative to the cost of being facultative ($0 < c_3c_4 < F_C$), the facultative fixer has a higher RGR.

conditions at or before $t - T$ and let the constrained facultative N fixer, F_L, (L for lag) fix N in such a way as to balance the resource availabilities about which it has information. The simplest extension of the unlagged facultative fixer F_F is that F_L fixes enough N at t such that it would be colimited at $t - T$:

$$F_L(t) = \min(F_C(t - T)^+, F_{max}). \tag{11}$$

Alternatively, plants may have the capacity to integrate past information, for example, if internal nutrient stores are used to regulate BNF, and they depend on nutrient availabilities before $t - T$. We call this integration of past information “memory,” for lack of a better term. To include memory in the model, we let F_L fix N so as to balance the resource availabilities from before $t - T$ to $t - T$, weighting information from the recent past more than the distant. If λ is the discounting/weighting factor, the amount fixed at time t is

$$F_L(t) = \int_{-\infty}^{t-T} F_C(\tau)\lambda e^{-\lambda(t-T-\tau)}d\tau, \tag{12}$$

which is still subject to the bounds of 0 and F_{max}. Equation (11) is a special case of equation (12). Differentiating equation (12) gives

$$\frac{dF_L}{dt} = \lambda[F_C(t - T) - F_L]. \tag{13}$$

Equation (13) can be coupled to equations (1)–(5), with

F_L (from eq. [12]) in place of F and with appropriate initial conditions, to make a set of delayed differential equations. It is not trivial to determine the optimal (or evolutionarily/convergence stable) facultative BNF strategy with the time lag constraint, and we make no claim that either version of F_L we present here is the best strategy. However, they are useful comparisons with F_F and F_O.

Unlike the unconstrained facultative N fixer F_F, F_L (with or without memory) is unlikely to be colimited at any given point in time. Which nutrient limits F_L depends on the history of nutrients in the soil as well as the current state. As shown in the appendix, the time-lagged facultative N fixer (F_L) never has a RGR higher than that of the unlagged facultative N fixer (F_F). Compared with an arbitrary obligate N fixer F_O, F_L may have a higher or lower RGR at any point in time, and therefore the competitive outcome is not immediately clear. If T is small and the recent information is strongly weighted, F_L is similar to F_F and therefore is likely to outcompete F_O. However, if the inherent time lag T is quite large, F_L is less likely to outcompete F_O and can even be outcompeted by F_O.

As our simulations demonstrate, sufficiently large time lags can permit an obligate nonfixer to outcompete time-lagged facultative N fixers without (fig. 2*C*, 2*D*) and with (fig. 2*E*, 2*F*) memory. (By "outcompete," we mean that it grows to a higher biomass level. We have proven nothing about coexistence, and simulations suggest that F_L can invade when it is rare, even in many scenarios in which it is outcompeted.) In figure 2*C*–2*F* the time lag is 1 year, which means that the N fixer adjusts its N fixation to meet the demand it had last year. This leads to pronounced differences in growth between the obligate nonfixer and lagged facultative N fixers (fig. 2*C*, 2*E*), as well as large N losses (fig. 2*D*, 2*F*). As has been known for some time (Cunningham 1954), sufficiently large time lags can destabilize otherwise stable systems. In our system as well, the time lag produces oscillations. The lagged facultative fixer continues fixing N after it is no longer N limited, then stops fixing N, and does not fix N again until it has been N limited for a period equal to the delay. This consistent overfixing or underfixing leads to its lower competitive ability (fig. 2*C*, 2*E*) and high N losses (fig. 2*D*, 2*F*).

Time lags much smaller than a year can also permit an obligate type to outcompete a lagged N fixer, even when there are no direct costs of being facultative. With a realistic set of parameters and starting conditions (see appendix), simulations reveal that the threshold lags that determine whether a nonfixer (one particular obligate type) outgrows F_L are between 1 and 2 days when F_L has no memory and 2 and 3 months when F_L has memory. For the large range of the memory effect parameter we tried (λ ranging from 10^{-4} to 10^{4} year^{-1}, lags ranging from a minute to a year), having memory consistently improves the competitive ability of F_L since the threshold time lag that determines whether F_L outcompetes the nonfixer was consistently higher when F_L had memory.

Effects of BNF Strategy and Time Lags on Available N Losses

Now that we have a sense for the effects of BNF strategy on competition, we investigate how the different strategies influence a fundamental ecosystem property and a key index of N richness: losses of available N from the soil. At equilibrium, available N losses in a system with an N-limited obligate N fixer slightly decrease as BNF increases. This happens because BNF increases equilibrium biomass, which drives $\overline{A}_N$ slightly lower. When an obligate N fixer is R limited, equilibrium N losses increase dramatically with increasing BNF for realistic parameters (see the appendix for details of these analyses). The whole equilibrium picture (fig. 4*A*), therefore, shows that available N losses decline very slightly with rising BNF until $F_O = \overline{F}_C$ (the kink) and then rise with overfixation. Figure 4*A* shows the effect of varying amounts of BNF on plant-available N losses at equilibrium (*solid line*) and quasi equilibrium (*dotted lines*, for two different states of plant biomass and soil organic nutrients) for a realistic set of parameters, flux functions, and starting conditions (see appendix). In each line the kink is at F_C, that is, where limitation switches from N limitation to R limitation and where the facultative N fixer F_F would be.

Many forests are not at the long-term equilibrium of this model, so we also examined N loss rates at quasi equilibrium, when soil-available nutrients are assumed to equilibrate rapidly relative to soil organic matter and plant biomass. When the plant is N limited, quasi-equilibrium N losses increase slightly with BNF. (The decrease seen in the equilibrium case results from longer-term feedbacks of increased biomass with BNF.) When an obligate N fixer is R limited, quasi-equilibrium N losses rise substantially with BNF, as illustrated in the dotted lines in figure 4*A* to the left (N limited) and the right (R limited) of the kink at $F_O = F_C$ (see appendix for details).

In the higher dotted line in figure 4*A*, plants need to fix N only at a miniscule rate to be colimited (the kink in the curve is at approximately 0.005 mg N g C^{-1} year^{-1}) because the N-to-R ratio in soil organic matter (mineralized and made available to the plant population) is higher than at equilibrium. Therefore, obligate N fixers with a higher BNF rate (such as 0.025 mg N g C^{-1} year^{-1}, which would give colimitation at equilibrium) would be R limited and allow about 3 kg N ha^{-1} year^{-1} to be lost as available N. The lower dotted line represents a state of the system that is more N poor than at equilibrium, as might be seen

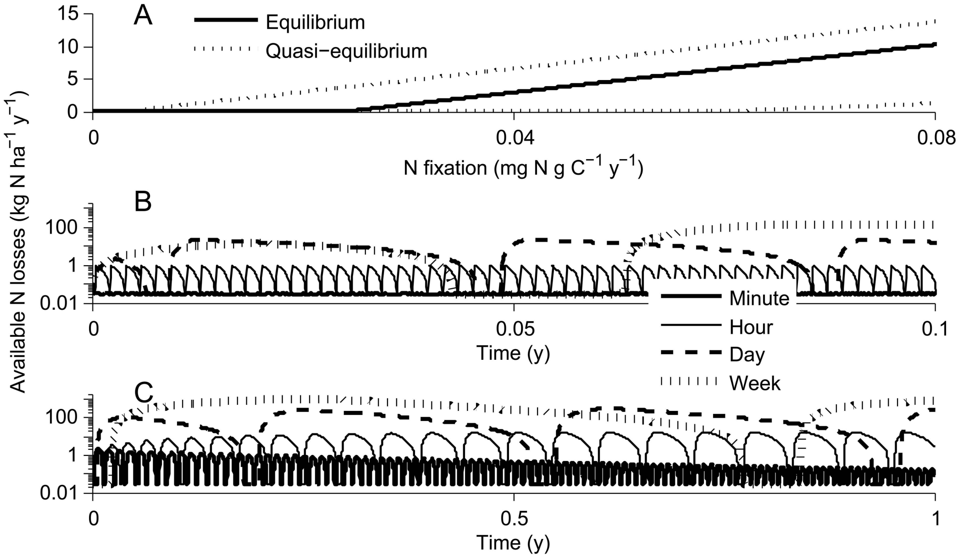

Figure 4: Effects of different levels of obligate biological N fixation (BNF; *A*), time lags without memory (*B*), and time lags with memory (*C*) on plant-available N loss. *A*, Equilibrium (*solid line*) and quasi-equilibrium (two dotted lines for differing amounts of plant and detritus) N losses as functions of the amount of BNF. In each line the kink represents F_C. Any $F_i < F_C$ yields N limitation and negligible available N losses, whereas any $F_i > F_C$ yields R limitation and increasing available N losses with BNF. *B*, Oscillating N losses over time from a time-lagged N fixer (eq. [11]) with four different delay magnitudes: 1 minute (*thick solid line*), 1 hour (*thin solid line*), 1 day (*dashed line*), and 1 week (*dotted line*). Note the logarithmic scale on the vertical axis. *C*, Same as *B*, but the plant has the capacity to integrate past information (eq. [12]). Note that the horizontal scale differs from that of *B*.

after a disturbance that enhances N loss (e.g., fire). Obligate N fixers fixing at a high enough rate to be colimited at this state (slightly over 0.06 mg N g C^{-1} year^{-1}) would yield negligible available N losses in this state but would overfix in the equilibrium and other quasi-equilibrium states, yielding available N losses over 5 kg N ha^{-1} year^{-1} at equilibrium and nearly 10 kg N ha^{-1} year^{-1} at the N-rich quasi equilibrium. The facultative N fixer without time lags would fix at F_C for any quasi-equilibrium or equilibrium state and, therefore, would yield low available N losses.

For the facultative N fixer with a time lag constraint without (fig. 4*B*) and with (fig. 4*C*) memory, N losses exhibit oscillatory patterns, as seen in the competition figure (fig. 2*D*, 2*F*). Both the frequency and the amplitude of the oscillations depend on the length of the time lag. For the parameter values we chose and an N fixer without memory, a delay of 1 min (fig. 4*B*, *thick solid line*) yields available N losses of approximately 0.03 kg N ha^{-1} year^{-1} (with very small oscillations), similar to no delay. A delay of an hour (fig. 4*B*, *thin solid line*) yields losses that oscillate up to 1 kg N ha^{-1} year^{-1}, with an average near 0.5 kg N ha^{-1} year^{-1}. Longer delays yield substantially larger losses: a delay of a day (fig. 4*B*, *dashed line*) and a week (fig. 4*B*, *dotted line*) yield losses that aggregate to about 10 and more than 50 kg N ha^{-1} year^{-1}, respectively.

Even though including memory increases an N fixer's competitive ability, it results in higher N losses with longer oscillations (note longer timescale on horizontal axis of fig. 4*C*) for a given time lag. A delay of 1 minute (fig. 4*C*, *thick solid line*) yields average losses of 0.5 kg N ha^{-1} year^{-1}, whereas a 1-h lag (fig. 4*C*, *thin solid line*) yields average losses close to 10 kg N ha^{-1} year^{-1}, a 1-day delay yields average losses close to 100 kg N ha^{-1} year^{-1}, and a 1-week delay yields losses of multiple hundreds of kilograms of N per hectare per year. The oscillations engendered by adding memory are consistently longer than without memory and substantially longer than the time lags themselves. For example, a time lag of 1 week yields an oscillation amplitude near 1 year with memory, compared with a couple of weeks without memory. Furthermore, the oscillation amplitudes and maxima for an hour or longer are growing much faster through time for the N fixer with memory.

Discussion

In an ecosystem with long periods between disturbances, a single BNF rate results in colimitation to the N fixer at equilibrium. An obligate N fixer that fixes this much ($\hat{F}_O$) beats all other obligate N fixers and nonfixers because it is not overfixing or underfixing in this stable environment.

In dynamic or transient environments, however, the best obligate BNF strategy is not clear, and facultative N fixers are more likely to win as long as the costs of being facultative are small. In fact, if BNF can be adjusted very rapidly, one type of facultative N fixer always fixes just enough to be colimited by N and another resource. This strategy (F_F) also yields very small N losses because it never overfixes. If there are no costs to being facultative, F_F is equivalent to the best obligate type in a stable environment and superior to any obligate type in dynamic environments because it is colimited whenever possible.

Although facultative N fixers always win in the simplest model, including costs of being facultative or time lags inherent to facultative BNF in the model can have strong effects on the competitive ability of facultative N fixers. Two types of cost we investigated were a fixed cost of being facultative and a variable cost of being facultative that increases the per-unit cost of BNF. A fixed cost could arise from the need to have infrastructure to sense and respond to the environment. All plants have such infrastructure for certain functions (e.g., stomatal guard cells that open and close in response to light and water availability), and the capacity to alter BNF may require additional infrastructure that would carry fixed structural, respiration, and opportunity costs. Facultative N fixers may also have higher per-unit costs of BNF. For example, building nodules requires carbon for structure as well as the metabolic cost of fixing N, and facultative N fixers that turn nodules on and off may be paying a higher structural cost per unit of N fixed than are obligate N fixers that keep their nodules for long periods. These costs of being facultative, if they are large enough, can prevent facultative BNF from evolving or succeeding. Therefore, these costs can also result in large N losses or severe N limitation because the success of obligate N fixers means overfixation in certain environments and underfixation in others.

Time lags inherent to BNF can also hinder the competitive ability of facultative N fixers and affect ecosystem-level N dynamics. Building nodules and altering carbon flow to nodules takes time, so adjusting BNF cannot be instantaneous. Because of feedbacks and delays among BNF, litterfall, and decomposition, even small lags in BNF can severely hinder the competitive ability of facultative N fixers. Moreover, because lags induce periods of N saturation to the plant as large quantities of N are being liberated from decomposing organic matter, and, conversely, periods of N starvation before BNF is active, they can lead to large losses of plant-available N and periods of severe N limitation, much like obligate N fixers. Therefore, both lagged facultative N fixers and obligate N fixers can push an ecosystem over thresholds into N richness or N limitation, with a key difference in timescale. Facultative N fixers adjust BNF on physiological timescales, which are much shorter than the timescale of community dynamics at which obligate BNF strategies affect N dynamics.

In addition to the costs we considered and time lags, a constraint on BNF that we did not include in our model may also affect N dynamics and the fitness of facultative N fixers. Rapid changes in the rate of BNF may be costly because of the construction cost of nodules, above and beyond the higher per-unit BNF cost we examined here. If this is true, an explicit incorporation of the cost of changing the BNF rate would likely reinforce our result that obligate N fixers can outcompete facultative N fixers if the cost of being facultative is high enough. However, it could also result in a benefit to certain types of time lags, which our current model does not allow. In particular, a condition that specifies that the plant waits for a large change in soil nutrient conditions before adjusting BNF could yield fewer large swings in BNF than are seen in instantaneously adjusting BNF to be colimited. In this case it is also possible that an intermediate lag is most beneficial.

Other model omissions may also affect our results and should be examined in future studies. For instance, distinguishing between fine roots and other plant tissues, including the possibility of light limitation, allowing plant stoichiometry to be flexible, and using more detailed growth equations would be more realistic and may alter cyclical dynamics or cost/benefit calculations. Despite these simplifications, our model results may shed light on BNF strategies and biome-level nutrient and biogeographic patterns.

Our model suggests that, without any constraints, all plants should be facultative N fixers. The constraints we examine here—which are by no means the only possible constraints—allow a much richer array of BNF strategies, as seems to occur in nature, and a correspondingly rich array of ecosystem N dynamics. Although some of the patterns themselves are poorly known (e.g., BNF strategies employed by different organisms in different biomes) and there are other potential explanations for biome-level differences (Houlton et al. 2008), we now speculate on the extent to which costs of being facultative and time lags inherent to BNF could influence biome-level patterns of BNF and N dynamics.

Temperate and Boreal Forest Pattern

The successional pattern in temperate and boreal forests is consistent with the following scenario: no facultative N fixer can invade any system because the costs of being facultative are sufficiently high (as in fig. 3*D*–3*F*). Early in succession, when N is strongly limiting, an obligate N fixer fixing at the maximal rate has the highest RGR, so it dominates early successional habitats (fig. 2*A*). As it brings N into the system, the limiting nutrient flips from N to R, at which point available N starts leaking in large quantities (fig. 2*B*; as seen in Binkley et al. 1992; Compton et al. 2003). As limitation switches from N to R, nonfixers have a higher RGR because they are not paying BNF costs. Some time after this switch in limitation, on the timescale of community dynamics, nonfixers overtake obligate N fixers and become dominant (fig. 2*A*). Without the BNF input, uncontrollable N losses bring N limitation back as the forest ages, and available N losses decrease to a minimum in a mature forest (fig. 2*B*; Hedin et al. 1995). This by no means proves that these mechanisms cause these patterns, but the extent to which the model results agree with observed patterns warrants further investigation into more poorly known aspects, such as whether actinorhizal N fixers are indeed obligate and the costs of being facultative.

Tropical Forest Pattern

The pattern starting to emerge in tropical forests—facultative N fixers coexisting with nonfixers and high available N losses (Jenny 1950; Vitousek et al. 2002; Hedin et al. 2003; Barron 2007)—is consistent with a time lag effect, and costs of being facultative could also play a role. Either constraint, if it has a moderate effect, could potentially allow coexistence over some time interval since both types have a higher RGR in certain regions of nutrient availability. For instance, if there is a small cost of being facultative (as in fig. 3*A*–3*C*), facultative N fixers would have the advantage in newly disturbed areas (such as treefall gaps) where N limitation is more likely. In undisturbed, N-rich areas, obligate nonfixers would have a higher RGR, even if the facultative N fixers are not fixing N (so long as there is a fixed cost of being facultative).

Time lags can also explain the coexistence of facultative N fixers with nonfixers (e.g., in fig. 2*C*, 2*E* both types coexist for a long time) and could explain the N richness seen in tropical forests (figs. 2*D*, 2*F*, 4*B*, 4*C*). Even a small time lag in facultative BNF produces substantial overfixation. In any given location this overfixation emerges as pulses in N richness following switches from N to R limitation (fig. 4*B*, 4*C*), but averaged over a landscape this could produce chronic N richness. Given the large effect of the time lag magnitude on N losses and the competitive ability of facultative N fixers, studies investigating the value of this time lag are sorely needed.

Why Facultative BNF in the Tropics and Obligate BNF in Temperate/Boreal Zones?

According to our model, costs of being facultative and time lags inherent to BNF can produce the patterns of obligate BNF in temperate/boreal forests and facultative BNF in tropical forests. To explain both patterns simultaneously, at least one of the constraints must be relatively greater outside the tropics. Two key differences in the two biomes may affect the magnitude of costs or time lags. First, because tropical forests are nearer the equator, they generally have higher temperatures and longer growing seasons than their poleward counterparts. Second, the dominant N-fixing symbioses differ phylogenetically: legumes with rhizobia dominate in the tropics, whereas actinorhizal symbioses between nonleguminous plants and actinomycete bacteria are more common in temperate and boreal forests.

Environmental effects may influence time lags and costs of being facultative. Enzyme activity depends on temperature, and because all processes involved in up- or down-regulating N fixation must depend on enzymes, colder temperatures away from the equator may push facultative N fixation over a threshold from net benefit to net cost. Given that the realized cost of being facultative depends on the maximum BNF rate ($F_{\max}$; fig. 3), lower $F_{\max}$ in lower temperatures may also render being facultative more costly. On a longer timescale, even if it takes a similar amount of time to build nodules in tropical and temperate forests, shorter growing seasons away from the equator may disfavor facultative nodulation because the active period is shorter relative to the building period.

Both partners in the symbiosis and the symbiotic structure itself (the nodule) differ between the two types of symbiosis (Huss-Danell 1997). Actinorhizal nodules are harder than rhizobial nodules and can be large, occasionally exceeding a 5-cm diameter (D. Menge, personal observation). This suggests that they may take longer to build or shed than the softer rhizobial nodules, consistent with a longer time lag in actinorhizal symbioses. Because the interior environment (in particular the oxygen content, which is known to be a key control on nitrogenase efficiency; Leigh 2002) of the actinorhizal nodule is less well regulated than that of the rhizobial nodule, the effective cost of each unit of BNF may also be higher for actinorhizal symbioses. Any combination of a longer time lag and a higher per-unit N cost of BNF in actinorhizal symbioses—if sufficiently large—could explain the difference between tropical and temperate forests. However, this is a proximate (as opposed to ultimate) explanation since it does not explain the biogeography of each symbiosis or why the actinorhizal and rhizobial structures differ. These may be because of historical accidents but may also be because of deterministic environmental effects such as those mentioned above.

Leguminous N-fixing trees exist in temperate zones (e.g., *Robinia pseudoacacia* [black locust]), and actinorhizal N fixers exist in the tropics (e.g., *Morella faya* [fire tree]), so it may be possible to tease apart environmental and phylogenetic effects. Data on the costs and time lags of

BNF are urgently needed to test the hypotheses from this work, which could help explain fundamental differences in community and ecosystem structure and dynamics between temperate/boreal and tropical forests.

Acknowledgments

This work was supported by a National Science Foundation (NSF) graduate research fellowship to D.N.L.M., NSF Doctoral Dissertation Improvement grant DEB-0608267, and NSF grant DEB-0614116. A. Barron, S. Batterman, and two anonymous reviewers gave helpful comments.

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Associate Editor: Judith L. Bronstein
Editor: Donald L. DeAngelis

Top, temperate forest with no nitrogen-fixing trees, Sequoia National Park; *bottom*, *Coriaria arborea* nodules, where nitrogen fixation happens, New Zealand. Photographs by Duncan N. L. Menge.

Appendix from D. N. L. Menge et al., “Facultative versus Obligate Nitrogen Fixation Strategies and Their Ecosystem Consequences”

(Am. Nat., vol. 174, no. 4, p. 465)

Additional Calculations and Simulation Details

Parameter Values and Starting Conditions for Simulations

The functions we use to calculate N losses are Michaelis-Menten for uptake and linear for nutrient net mineralization and losses, as shown in table A1. The parameters we use are in table A2.

Table A1

Functional forms used to make N loss figure

Process	Function
Nutrient uptake	$\nu_j(A_j) = \nu_j A_j/(\beta_j + A_j)$
Net mineralization	$m_j(D_j) = m_j D_j$
Plant-unavailable nutrient loss	$\phi_j(D_j) = \phi_j D_j$
Plant-available nutrient loss	$k_j(A_j) = k_j A_j$

Table A2

Parameter values used for simulations

Parameter	Value/N	P	Units
ω_j	50	600	kg C kg j^{-1}
ν_j	.2	.2	kg j kg C^{-1} year^{-1}
β_j	.1	.01	kg j ha^{-1}
μ	.5	. . .	year^{-1}
θ_g	5	. . .	kg C kg N^{-1}
θ_μ	5	. . .	kg C kg N^{-1}
γ_g	.025	. . .	year^{-1}
γ_μ	.025	. . .	year^{-1}
m_j	.3	.4	year^{-1}
ϕ_j	.001	.001	year^{-1}
I_j	2	.4	kg j ha^{-1} year^{-1}
k_j	5	5	year^{-1}
$F_{\max}$	.01	. . .	kg N kg C^{-1} year^{-1}
λ	1	. . .	year^{-1}

Note: R is parameterized as phosphorus (P); j refers to a nutrient. Parameter values for figure 4 are as shown; parameter values for figure 2 are as shown, except for γ_g and γ_μ (both 0), ν_{P} (0.02), β_{P} (0.1), k_{N} (1), and k_{P} (0.005); λ is relevant only for figures 2*E*–2*F* and 4*C*; ω and μ were chosen to represent common C : N ratios, C : P ratios, and leaf life spans (e.g., Wright et al. 2004); I was chosen to represent unpolluted ecosystems (e.g., Smil 2000; Galloway et al. 2004); $F_{\max}$ was chosen to yield, along with steady state N-fixer monoculture biomass, biological N fixation input rates near the maximum reported in a global synthesis (Cleveland et al. 1999); ν, β, m, ϕ, and k were chosen to fit N and P fluxes to standard fluxes for forests (e.g., Vann et al. 2002). The growth cost of fixation (θ_g) comes from the metabolic cost (Gutschick 1981), the turnover cost (θ_μ) was assumed to be similar, and the costs of being facultative (γ) were our best guesses; we know of no data from which to estimate them.

The simulation in figure 2*A* and 2*B* is meant to be primary succession; hence the starting conditions are 1 kg C ha^{-1} each for the obligate N fixer and the nonfixer, 1 kg N ha^{-1} of mineralizable soil N, 5 kg P ha^{-1} of mineralizable P, 0.001 kg N ha^{-1} of available N, and 0.1 kg P ha^{-1} of available P. The simulations for figure

2*B*–2*F* begin with both the facultative fixer and the nonfixer at 50%, detritus N and P at 95% and 101%, and available N and P at 90% and 105% of the equilibrium for the system with the facultative N fixer alone. Simulations for figure 4*B* and 4*C* are the same as for figure 2*B*–2*F*, except that the facultative N fixer begins at 95% of its equilibrium biomass and the nonfixer is absent. For figure 4*A*, starting conditions for the equilibrium run were the equilibrium given these functions and parameters (analytically solved by setting eqq. [1]–[5] equal to 0). For the first quasi-equilibrium run we started at the equilibrium for the N-limited nonfixer, and for the second we removed some plant biomass and soil organic P from the equilibrium for a slightly N-limited obligate N fixer.

Obligate Continuously Stable Strategy Calculations

To show that $\hat{F}_{\mathrm{O}}$ is a continuously stable strategy, we must show that it is both evolutionarily stable and convergence stable (Eshel 1983). To show convergence stability, we must show that it will be approached evolutionarily from any starting population at ecological equilibrium, that is, from any N-limited or R-limited population. In an equilibrium environment, N-limited N-fixing mutants F_{m} can invade residents F_{r} when

$$\left.\frac{1}{B_{\mathrm{m}}}\frac{dB_{\mathrm{m}}}{dt}\right|_{\bar{A}_{\mathrm{N,r}}} = (\omega_{\mathrm{N}} - \theta)(F_{\mathrm{m}} - F_{\mathrm{r}}). \tag{A1}$$

The quantity $\bar{A}_{\mathrm{N,r}}$ is the equilibrium soil-available N pool in the ecosystem with the resident alone and is a constant. Because we have assumed that the growth benefit of N fixation (the nutrient use efficiency, ω_{N}) exceeds the cost (θ), N-limited mutants fixing more than the residents will always invade, and thus $\hat{F}_{\mathrm{O}}$ will be approached from any N-limited population. This is similar to what we found in an earlier study (Menge et al. 2008), but in that work we examined what the costs could be and how high they would need to be.

The R-limited N-fixing mutants invade residents when

$$\left.\frac{1}{B_{\mathrm{m}}}\frac{dB_{\mathrm{m}}}{dt}\right|_{\bar{A}_{\mathrm{N,r}}} = \theta(F_{\mathrm{r}} - F_{\mathrm{m}}), \tag{A2}$$

so mutants fixing less than residents will always invade when they are R limited. Therefore, in an equilibrium environment, selection will push a population toward the N fixation rate that yields colimitation, so $\hat{F}_{\mathrm{O}}$ is convergence stable.

To show evolutionary stability, we must show that $\hat{F}_{\mathrm{O}}$ cannot be invaded once it is established. At equilibrium, a population of obligate N fixers fixing at $\hat{F}_{\mathrm{O}}$ is colimited by N and R. A mutant F_{m^+} fixing more N than the colimited resident F_{r} (with $F_{\mathrm{m}^+} > F_{\mathrm{r}}$) would be R limited, with a lower relative growth rate (RGR) than the resident:

$$\frac{1}{B_{\mathrm{m}^+}}\frac{dB_{\mathrm{m}^+}}{dt} = \omega_{\mathrm{R}}\nu_{\mathrm{R}}(\bar{A}_{\mathrm{R,r}}) - \theta F_{\mathrm{m}^+}, \tag{A3}$$

$$\frac{1}{B_{\mathrm{r}}}\frac{dB_{\mathrm{r}}}{dt} = \omega_{\mathrm{R}}\nu_{\mathrm{R}}(\bar{A}_{\mathrm{R,r}}) - \theta F_{\mathrm{r}}. \tag{A4}$$

Because $F_{\mathrm{m}^+} > F_{\mathrm{r}}$, $(1/B_{\mathrm{r}})(dB_{\mathrm{r}}/dt) > (1/B_{\mathrm{m}^+})(dB_{\mathrm{m}^+}/dt)$. Conversely, a mutant F_{m^-} fixing less N ($F_{\mathrm{m}^-} < F_{\mathrm{r}}$) would be N limited but would still have a lower RGR than the resident:

$$\frac{1}{B_{\mathrm{m}^-}}\frac{dB_{\mathrm{m}^-}}{dt} = \omega_{\mathrm{N}}\nu_{\mathrm{N}}(\bar{A}_{\mathrm{N,r}}) + (\omega_{\mathrm{N}} - \theta)F_{\mathrm{m}^-}, \tag{A5}$$

$$\frac{1}{B_{\mathrm{r}}}\frac{dB_{\mathrm{r}}}{dt} = \omega_{\mathrm{N}}\nu_{\mathrm{N}}(\bar{A}_{\mathrm{N,r}}) + (\omega_{\mathrm{N}} - \theta)F_{\mathrm{r}}. \tag{A6}$$

In both cases, the mutant has a lower RGR than the resident, which by definition is 0 at equilibrium, so no mutants have a positive growth rate at equilibrium. Therefore, no mutant can invade $\hat{F}_O$, so it is evolutionarily stable and therefore a continuously stable strategy.

RGRs of F_F versus Other N Fixers without Costs of Being Facultative or Time Lags

It seems intuitive that the facultative fixer F_F (see eq. [9]) should always be at least as competitive as any other N fixer as long as there is no cost to being facultative and there are no time lags. Comparing the RGRs (from eq. [1]) of the F_F with those of an arbitrary F_i proves that this intuition is right. Here, F_i can be an obligate fixer (F_O), a time-lagged facultative N fixer (F_L), or any N fixer other than F_F, all of which would typically be overfixing or underfixing (and therefore not colimited by N and R).

When $F_C \leq 0$, all plants are R limited, and F_i may pay a higher cost than F_F:

$$\frac{1}{B_F}\frac{dB_F}{dt} = \omega_R \nu_R(A_R) - \mu, \tag{A7}$$

$$\frac{1}{B_i}\frac{dB_i}{dt} = \begin{cases} \omega_R \nu_R(A_R) - \mu & \text{if } F_i = 0 \\ \omega_R \nu_R(A_R) - \mu - \theta F_i & \text{if } F_i > 0 \end{cases}. \tag{A8}$$

When $F_C \geq F_{max}$, all plants are N limited, and F_i may not get the same gain as F_F:

$$\frac{1}{B_F}\frac{dB_F}{dt} = \omega_N \nu_N(A_N) - \mu + (\omega_N - \theta)F_{max}, \tag{A9}$$

$$\frac{1}{B_i}\frac{dB_i}{dt} = \begin{cases} \omega_N \nu_N(A_N) - \mu + (\omega_N - \theta)F_i & \text{if } F_i < F_{max} \\ \omega_N \nu_N(A_N) - \mu + (\omega_N - \theta)F_{max} & \text{if } F_i = F_{max} \end{cases}. \tag{A10}$$

When $0 < F_C < F_{max}$, F_F is always colimited, but F_i could be R limited or N limited:

$$\frac{1}{B_F}\frac{dB_F}{dt} = \omega_N \nu_N(A_N) - \mu + (\omega_N - \theta)F_C = \omega_R \nu_R(A_R) - \mu - \theta F_C, \tag{A11}$$

$$\frac{1}{B_i}\frac{dB_i}{dt} = \begin{cases} \omega_N \nu_N(A_N) - \mu + (\omega_N - \theta)F_i & \text{if } F_i < F_C \\ \omega_R \nu_R(A_R) - \mu - \theta F_i & \text{if } F_i > F_C \end{cases}. \tag{A12}$$

When $F_C < 0$ and $F_i = 0$, $F_C > F_{max}$ and $F_i = F_{max}$, or $F_C = F_i$, the RGRs for F_i and F_F are identical, but everywhere else F_F has a higher RGR.

Equilibrium and Quasi-Equilibrium N Loss Calculations

Equilibrium N Losses

When the plant is N limited, equilibrium available N losses are given by

$$k_N(\bar{A}_N) = k_N\left\{\nu_N^{-1}\left[\frac{\mu}{\omega_N} - F_O\left(1 - \frac{\theta}{\omega_N}\right)\right]\right\}. \tag{A13}$$

The functions k_N and ν_N^{-1} are monotonically increasing (assuming the inverse function ν_N^{-1} exists, which it must for an equilibrium $\bar{A}_N$ to exist), and $\omega_N > \theta$, so equation (A13) shows that available N losses decrease as biological N fixation (BNF) increases. Exploration of reasonable parameter ranges show that this decrease is very small relative to changes in N losses when the plant is R limited.

When the plant is R limited, equilibrium available N losses are given by the balance of inputs and the plant-unavailable N loss:

$$k_N(\bar{A}_N) = I_N - \phi_N(\bar{D}_N) + \bar{B}F_O. \tag{A14}$$

To determine how increasing BNF affects equilibrium losses of plant-available N, we need to know how it affects the equilibrium values of state variables.

To do this, we first derive equilibrium expressions from equations (1)–(5), using the R-limited version of equation (6). We leave the functions $\nu_j(A_j)$, $m_j(D_j)$, $\phi_j(D_j)$, and $k_j(D_j)$ unspecified for generality, still assuming monotonicity and intersection of the origin. From setting equation (1) equal to 0,

$$\omega_R \nu_R(\overline{A}_R) = \mu + \theta F_O, \tag{A15}$$

$$\overline{A}_R = \nu_R^{-1}\left(\frac{\mu + \theta F_O}{\omega_R}\right), \tag{A16}$$

assuming ν_R^{-1} exists (which it must for $\overline{A}_R$ to exist and therefore for the R-limited equilibrium to be biologically relevant). Because $\nu_R(A_R)$ is monotonically increasing, ν_R^{-1} is also monotonically increasing, and therefore

$$\frac{\partial \overline{A}_R}{\partial F_O} > 0. \tag{A17}$$

From setting equation (3) equal to 0,

$$\overline{B} = \frac{\omega_R}{\mu}[m_R(\overline{D}_R) + \phi_R(\overline{D}_R)]. \tag{A18}$$

Plugging equations (A16) and (A18) into equation (5) and setting it equal to 0,

$$I_R = k_R\left(\nu_R^{-1}\left(\frac{\mu + \theta F_O}{\omega_R}\right)\right) + \phi_R(\overline{D}_R). \tag{A19}$$

Therefore,

$$\frac{\partial \overline{D}_R}{\partial F_O} < 0, \tag{A20}$$

and from equation (A18) and condition (A20),

$$\frac{\partial \overline{B}}{\partial F_O} < 0. \tag{A21}$$

From setting equation (2) equal to 0,

$$\frac{\mu \overline{B}}{\omega_N} = m_N(\overline{D}_N) + \phi_N(\overline{D}_N), \tag{A22}$$

and using condition (A21),

$$\frac{\partial \overline{D}_N}{\partial F_O} < 0. \tag{A23}$$

At equilibrium, increasing BNF when the plant is R limited decreases plant biomass ($\overline{B}$), soil organic N ($\overline{D}_N$), and soil organic R ($\overline{D}_R$) but increases plant-available R in the soil ($\overline{A}_R$). Therefore, the second term on the right-hand side of equation (A14) increases with BNF, but whether the last term increases or decreases with BNF depends on the magnitude of the change in $\overline{B}$ with BNF. Specifically, the partial derivative of equilibrium available N losses ($k_N(A_N)$; eq. [A14]) with respect to F_O is

$$\frac{\partial k_{\mathrm{N}}(\bar{A}_{\mathrm{N}})}{\partial F_{\mathrm{O}}} = -\frac{\partial \phi_{\mathrm{N}}(\bar{D}_{\mathrm{N}})}{\partial F_{\mathrm{O}}} + F_{\mathrm{O}}\left(\frac{\partial \bar{B}}{\partial F_{\mathrm{O}}} + \frac{\bar{B}}{F_{\mathrm{O}}}\right). \tag{A24}$$

The first term is positive (because of monotonicity and condition [A23]), and the second depends on the sign of $(\partial\bar{B}/\partial F_{\mathrm{O}}) + (\bar{B}/F_{\mathrm{O}})$. If

$$\left|\frac{\partial \bar{B}}{\partial F_{\mathrm{O}}}\right| \leq \frac{\bar{B}}{F_{\mathrm{O}}}, \tag{A25}$$

$$\frac{\partial k_{\mathrm{N}}(\bar{A}_{\mathrm{N}})}{\partial F_{\mathrm{O}}} > 0. \tag{A26}$$

If condition (A25) is met, then a graph for the equilibrium scenario (with F_{O} on the horizontal and $k_{\mathrm{N}}(\bar{A}_{\mathrm{N}})$ on the vertical) decreases until F_{C} and then increases thereafter. For all realistic values we have tried, condition (A25) is met.

Quasi-Equilibrium N *Losses*

To examine how plant-available N losses depend on BNF away from equilibrium, we analyzed N losses at quasi equilibrium. In this analysis we assume that plant-available nutrients in the soil equilibrate rapidly relative to plant biomass and organic nutrients in the soil, which is generally the case in terrestrial ecosystems (see Menge et al. 2009). Using the notation $\hat{A}_{\mathrm{N}}$ for the quasi equilibrium of A_{N}, the effect of BNF on quasi-equilibrium losses of available N in a system with an N-limited obligate N fixer is given by

$$\frac{\partial}{\partial F_{\mathrm{O}}}[k_{\mathrm{N}}(\hat{A}_{\mathrm{N}}) + B\nu_{\mathrm{N}}(\hat{A}_{\mathrm{N}})] = B\left(\frac{\theta_g}{\omega_{\mathrm{N}}}\right), \tag{A27}$$

where B is treated as a constant. Because of the trade-off between BNF and N uptake from the soil, per-biomass plant uptake decreases with increasing BNF. Therefore, at the short timescale during which plant biomass is effectively constant, N losses increase with increasing BNF. Our exploration of parameter values shows that this is a very slight increase.

When the obligate N fixer is R limited, the effect of BNF on available N losses is greater than when it is N limited:

$$\frac{\partial}{\partial F_{\mathrm{O}}}\left[k_{\mathrm{N}}(\hat{A}_{\mathrm{N}}) + B\frac{\omega_{\mathrm{R}}}{\omega_{\mathrm{N}}}\nu_{\mathrm{R}}(\hat{A}_{\mathrm{R}})\right] = B\left(\frac{\theta_g}{\omega_{\mathrm{N}}} + 1\right). \tag{A28}$$

Because $\theta_g/\omega_{\mathrm{N}} \ll 1$, the rise in N losses when an obligate N fixer is R limited is substantially greater than when it is N limited. Therefore, as in the equilibrium case, BNF has little effect on plant-available N losses when the plant is N limited but a strong positive effect on N losses when the plant is R limited.

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