Using hybrid automata modelling to study phenotypic plasticity and allocation strategies in the plant mycorrhizal mutualism

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A B S T R A C T

Plants are exemplified by high plasticity in resource allocation strategies which allows them to maximize their fitness under changing resource conditions. In many plant species, obtaining resources involves mutualistic interactions with arbuscular mycorrhizal fungi (AMF), where plants provide the AMF with sugars in exchange for soil nutrients like phosphorus (P). These nutrient exchange rates have high context dependency, influenced by both the cooperative level of the AMF species involved (a qualitative trait) and the ambient nutrient concentrations in the soil (a quantitative value). Because this context dependency arises from a mix of both quantitative and qualitative factors, standard ordinary differential equation (ODE) modeling methods often complicate the representation of resource allocation strategies. Here, we explore the utility of a hybrid automata modeling framework that can intuitively combine the qualitative AMF traits and quantitative nutrient concentrations. This allows for a better analysis and understanding of phenotypic plasticity in resource allocation in the plant–AMF, and other nutrient exchange mutualisms. We consider a focal strategy in which nutrients are allocated to growth at times of nutrient limitations, and to storage otherwise and ask how this changes plant allocation to growth vs. storage. We first model this system dynamically to show how the plant responds to different environmental conditions and interacts with AMF and show that our hybrid automata model can replicate experimental data from the plant–AMF system. From our work, novel perceptions into the well-studied plant–AMF symbiosis and testable hypotheses can be underlined: (1) leaf biomass does not increase proportionally with the level of AMF cooperation; (2) in the context of multiple AMF simultaneously colonizing a host–plant, a narrow variance of response is observed and explained by an auction-like mechanism of the AMF to acquire C from the plant.

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1. Introduction

Phenotypic plasticity is the ability of an organism to change its phenotype based on environmental conditions such as nutrient availability and temperature (West-Eberhard, 1989). For example, in plants, high phenotypic plasticity in root growth is observed related to resource distributions or neighbor interactions (e.g. Hodge, 2004; Hutchings and de Kroon, 1994). As a result of this plasticity, plants can balance their carbon (C) investment to plant shoots and roots depending on the growing conditions.

Plant–phenotypic plasticity can also result from the symbiosis between plants and arbuscular mycorrhizal fungi (AMF) (Streitwolf-Engel et al., 1997). Because 80% of land plants are involved in AMF symbiosis (Smith and Read, 2008), the impact of this interaction on plant trait plasticity is likely substantial, yet has not been extensively studied. In this nutrient-exchange symbiosis, the plant is a facultative mutualist that provides photosynthetically-derived sugars to its obligate mutualist partner, AMF, in exchange for soil nutrients like phosphorus (P). Our aim is to examine the allocation strategies, i.e. the phenotypic plasticity such as root and leaf growth, of the plant depending on changes with its fungal partner. In general, a plant has different allocation strategies based on the environmental conditions, namely it allocates resources to growth vs. storage depending on the value of limiting nutrients (Poorter et al., 2012; Schachman et al., 1998). For example, a low atmospheric C concentration induces investment in the
growth of leaf biomass to increase the uptake of C, whereas shading of the host plant can result in changes in the amount of C the host allocates to the root and fungal partner (Felibaum et al., 2014 and references therein).

A mathematical model of these dynamic processes is of great interest to explore how the environment influences the potential phenotypes of the organism formally by means of functions and variables. However, accurately representing the switching between qualitative phenotypes (e.g. discrete allocation strategies) and quantitative concentrations (e.g. changes in nutrients in the environment) can be difficult and complex if using standard modeling approaches such as ordinary differential equation (ODE) modeling methods (Zhai et al., 2014; Suzuki and Arita, 2013; Thornley and Parsons, 2014). In particular, dynamical equations often remain difficult to parametrize and are unable to capture both qualitative and quantitative features of available experiments. Rather than to model the details of these strategies in terms of specific chemical reactions and processes (Meyer et al., 2012), we use hybrid automata to describe the strategies, namely allocating resources to growth or storage, qualitatively, while basing these strategies on quantitative concentrations.

As promoted in Systems Biology for studying single cell molecular systems, hybrid automata is an alternative modeling method (Henzinger, 1996; Henzinger et al., 1996) that we advocate for studying organisms displaying phenotypic plasticity. Hybrid methods are hybrid because they are able to combine qualitative (e.g. discrete allocation strategies) with quantitative (e.g. fluctuations in concentrations) behaviors. A hybrid automaton is a hybrid method that switches between allocation strategies via the transitions, which are triggered when conditions on variables are satisfied. In that sense, any system displaying phenotypic plasticity could be modeled using hybrid automata. For example, an organism that changes behavior with respect to the seasons can be intuitively modeled using hybrid automata. Hybrid automata are implemented in systems biology (Siebert and Bockmayr, 2008; Ahmad et al., 2009, 2008), where the qualitative feature provides a convenient representation of the system when detailed or quantitative knowledge is lacking. In this paper, we show the potential of the hybrid automata modeling approach by considering a system that exhibits phenotypic plasticity. Although automata and hybrid methods have been individually applied to other systems such as cells and economies (Gronewold and Sonnenschein, 1998; Liu et al., 2014), hybrid automata have not yet been applied to an ecological system, such as a symbiotic relationship.

Using an ecological example of the plant–AMF symbiosis, hybrid automata are able to describe the fluctuation of carbon (C) and phosphorus (P) concentrations over time with changing allocation strategies. For example, allocation of storage could decrease the concentration of C and in turn, decreasing C concentration could switch resource allocation from storage to leaf growth (Fig. 1). In other words, representing the system as a hybrid automata intuitively combines the quantitative change in concentrations with the qualitative change in allocation strategies.

Our dedicated hybrid automata model accurately represents the plant–AMF C-P exchange system in order to investigate the plant’s behavior of altering resource allocation to growth vs. storage according to which nutrients are limiting. In particular, we aim to determine when the switching between different strategies occurs. The challenge arises in coordinating the strategies of above- and belowground structures with the C and P that they both depend on.

We simulate scenarios where a host plant is exchanging nutrients with a fungal partner. We observe different strengths of symbiosis, i.e., rates of nutrient exchange between organisms, based on data from empirical work (Kiels et al., 2011). The formal construction of the model allows us to examine the link between data from experimental set-ups and the processes of resource allocation and exchange with AMF. Since the differential behavioral types for both the plant and AMF is completely lacking (Cahill and McNickle, 2011), this work looks to bridge the gap between the behaviors of the organism and their underlying allocation strategies.

2. Methods

2.1. Variables

The variables used in our model (Table 1) are associated with the plants allocation strategies with AMF symbiosis, namely obtaining C and P and allocating the nutrients in the processes of growth, storage and exchange. We assume that each variable can only take positive finite values due to physical restrictions of the plant. \(C_p\) and \(P_p\) are used by the plant for general maintenance, growth, and exchange. \(S_{C}\) and \(S_{P}\) are the forms of carbon and phosphorus that are not available for use in maintenance, growth or exchange (e.g. starches). Qualitatively, we distinguish low and high values of \(C_p\) and \(P_p\) using the thresholds, \(\theta_C\) and \(\theta_P\) respectively (Fig. 1).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Definition</th>
<th>Value Unit</th>
</tr>
</thead>
<tbody>
<tr>
<td>(C_p)</td>
<td>Concentration of freely available carbon</td>
<td>(1) mmol g(^{-1})</td>
</tr>
<tr>
<td>(P_p)</td>
<td>Concentration of freely available phosphorus</td>
<td>(1) mmol g(^{-1})</td>
</tr>
<tr>
<td>(b_0)</td>
<td>Biomass of aboveground structures</td>
<td>(10) g</td>
</tr>
<tr>
<td>(b_i)</td>
<td>Biomass of belowground structures</td>
<td>(10) g</td>
</tr>
<tr>
<td>(S_C)</td>
<td>Stored carbon</td>
<td>(0) mmol g(^{-1})</td>
</tr>
<tr>
<td>(S_P)</td>
<td>Stored phosphorus</td>
<td>(0) mmol g(^{-1})</td>
</tr>
<tr>
<td>(\theta_C)</td>
<td>Threshold of carbon</td>
<td>(30) mmol g(^{-1})</td>
</tr>
<tr>
<td>(\theta_P)</td>
<td>Threshold of phosphorus</td>
<td>(30) mmol g(^{-1})</td>
</tr>
</tbody>
</table>
cycling from decomposer activity, we address the potential for spatially heterogeneous P distributions (Boswell and Davidson, 2012) as a point of future work.

2.2. Reactions

The plant processes such as nutrient uptake, storage, and exchange can be simplified as first order reactions. In particular, we focus on the reactions in the plant that play a direct role in symbiosis with AMF (Table 2; Fig. 2).

The uptake of C models photosynthesis, where C is taken up from the atmosphere by the plant and made into freely available carbon, $C_p$, in the plant. Plants take up P either directly from the soil via their roots or receive it from an exchange with AMF. In both growth reactions, a unit of C and a unit of P are required for an increase in biomass. The storage reactions represent the process of $C_p$ or $P_p$ being converted into their more stable stored forms. The unstable $C_p$ can degrade or be used for general plant maintenance, hence the degradation reactions. Finally, we consider the exchange reaction with the AMF.

During exchange, the AMF is modeled assuming a particular AMF strategy, i.e. cooperative or less cooperative based on C to P exchange (Kiers et al., 2011). In particular, the exchange reaction can be described as one C unit in receiving x P units, where $x=10$ can be classed as the most cooperative AMF and $x=1$ as the least cooperative, and other values of x lie along this cooperation continuum (Johnson et al., 1997). There are two exchange reaction rates, namely the plant exchange rate, which is the amount of C that the plant is giving to the AMF, and the AMF exchange rate, which is the amount of P given back to the plant. The AMF exchange rate, however, can be calculated by multiplying the plant exchange rate with the AMF strategy. For this reason, exchange rate henceforth refers to the plant exchange rate with an AMF that has a given level of cooperation.

When leaves grow, we assume that all aboveground structures are increasing, i.e. the biomass $B_l$ increases. Moreover the mass of aboveground structures is assumed to be proportional to the leaf surface area, which, in turn, determines the rate of C uptake. In other words, the uptake of C increases when there is an increase in leaf biomass. To model the change in exchange rate, we used a simple mass action law, i.e. the rate of C uptake, $a_C$, is proportional to $C_p - B_l$. The same reasoning and mass action equation holds for the growing of roots, belowground structures, and uptake of P.

Growth and storage are dependent on whether a nutrient is limiting (Fig. 1). That is, we describe the allocation strategies of the plant quantitatively: namely growth of leaves occurs when $C_p < B_l$ and storage of C occurs otherwise. Similarly, growth of roots occurs when $P_p < B_l$ and storage of P occurs otherwise. Degradation and maintenance are always occurring.

Concerning the exchange rate, we include the rewarding mechanism (Kiers et al., 2011), also termed ‘linear proportional discrimination’ (Wyatt et al., 2014) into the model. In particular, we say that over a set time period, the plant exchange rate, $\epsilon$, with a given AMF increases at a steady rate proportional to the value $x$ associated with that AMF. For example, in a set time period, the $x=10$ AMF would receive an increase in the exchange rate by 10 whereas the $x=1$ AMF only receives an increase of 1 (Fig. A.1). However, we assume that the rate of exchange is only increasing during phases of growth, which is representative of annual plants. This is an important assumption that will determine the model results. The plant’s exchange with fungi is determined by the surface area of the roots, which does not grow exponentially. In order to distinguish between different funga a linear model is preferred to a logistic model.

### Table 2

<table>
<thead>
<tr>
<th>Rate</th>
<th>Reaction</th>
<th>Description</th>
<th>Value</th>
<th>Unit</th>
</tr>
</thead>
<tbody>
<tr>
<td>$a_C$</td>
<td>$C_p \rightarrow B_l$</td>
<td>uptake of C</td>
<td>$B_l$</td>
<td>mmol m$^{-1}$ s$^{-1}$</td>
</tr>
<tr>
<td>$a_P$</td>
<td>$P_p \rightarrow B_l$</td>
<td>uptake of P</td>
<td>$B_l$</td>
<td>mmol m$^{-1}$ s$^{-1}$</td>
</tr>
<tr>
<td>$b_C$</td>
<td>$C_p + P_p \rightarrow B_l$</td>
<td>growth of leaves</td>
<td>100</td>
<td>mg m$^{-1}$ day$^{-1}$</td>
</tr>
<tr>
<td>$\gamma$</td>
<td>$C_p + P_p \rightarrow B_l$</td>
<td>growth of roots</td>
<td>100</td>
<td>mg m$^{-1}$</td>
</tr>
<tr>
<td>$\gamma'$</td>
<td>$C_p \rightarrow S_t$</td>
<td>storage of C</td>
<td>70</td>
<td>mmol m$^{-1}$</td>
</tr>
<tr>
<td>$\gamma''$</td>
<td>$P_p \rightarrow S_t$</td>
<td>storage of P</td>
<td>70</td>
<td>mmol m$^{-1}$</td>
</tr>
<tr>
<td>$\beta$</td>
<td>$C_p \rightarrow C_{dep}$</td>
<td>degradation/maintenance</td>
<td>30</td>
<td>mmol m$^{-1}$</td>
</tr>
<tr>
<td>$\nu$</td>
<td>$P_p \rightarrow P_{dep}$</td>
<td>degradation/maintenance</td>
<td>30</td>
<td>mmol m$^{-1}$</td>
</tr>
<tr>
<td>$\epsilon$</td>
<td>$C_p \rightarrow xP_p$</td>
<td>C is exchanged for x units of P</td>
<td>10$x$</td>
<td>mmol m$^{-1}$</td>
</tr>
</tbody>
</table>

### 3. Hybrid automata

With the variables and reactions (Tables 1 and 2), we can represent the allocation strategies of the plant as a hybrid automaton (Fig. 2B). The allocation strategies are displayed as boxes, where each strategy contains a subset of reactions from Table 2 that dictate how the variables will change over time. Finally, the transitions between strategies are labeled with conditions on the variables.

The dynamics of the hybrid automaton are as follows: starting in one strategy, the variables change based on the available reactions of that strategy. A transition between strategies occurs when the conditions on the variables are satisfied. For example, starting in the bottom left strategy of Fig. 2B, the variables change due to the reactions until either $C_p > \theta_l$ or $P_p > \theta_l$, which switches the strategy to either the bottom right or top left respectively.

Hybrid automata could be simulated deterministically or non-deterministically. Because of the inherent uncertainties of plant–AMF system, we propose to investigate herein the impact of strategies by using a non-deterministic, namely probabilistic, version of the above hybrid automata.

#### 3.1. Probabilistic hybrid automata

For this purpose, we use a random sampling procedure described as follows: each state of the system (i.e. specific concentration values of every variable) has a set of available reactions corresponding to the states allocation strategy, i.e. the subset of reactions in each box of Fig. 2B. We choose one of these available reactions at random but weight each reaction proportionally to their reaction rate, e.g. if the uptake of P has a rate five times that of the uptake of C, i.e. if $a_C/a_P=5$, then the uptake of C has a five-in-one chance of being chosen over the uptake of P. The chosen reaction is then applied, where the corresponding units of substrates are subtracted and units of products added, putting the system in a new state to repeat the random sampling.

Because of the random sampling of reactions, each trajectory of our probabilistic hybrid automaton has some variation, unlike the deterministic trajectories of ODEs, where from a single initial value there is one unique trajectory. The variation that comes from the random sampling implies the need to repeat simulations analogous to the wet lab experiments. Two example trajectories that use this procedure of random sampling and that trace the concentrations of $C_p$ and $P_p$ are depicted in Fig. A.2.

Note that different initial conditions would influence the final state of the system. We chose the origin as the initial condition because we assume that the strength of symbiosis is best determined when the entire life of the plant is observed. Nevertheless, if the leaves of the plant are suddenly cut off by grazers for example, the plant could revert to its growth phase, which would represent an initial condition with large root biomass and small leaf biomass. Herbivory can induces low to high plant defense syndromes (Agrawal and Fishbein, 2006). Our work does not address this aspect, especially possible high responses against herbivory.
We conducted simulations under 4 levels of P uptake coefficients (1, 1/2, 1/3 and 1/4) and fixed values of all other parameters (Table 2), where the coefficients could represent the high to low P levels in the soil.

3.2. Model checking

For the sake of validation, models that represent uncertainties cannot be simulated alone. Probabilistic models need to be verified given their non-deterministic nature. Using the benefit of the automata framework, we advocate the use of available model checking tools, which are parallel to the formal analysis techniques of ODEs.

Model checking is a formal verification technique used in computer science (Bianco and de Alfaro, 1995), where automata are tested for favorable or unfavorable properties. One of the largest and most successful applications of model checking is verifying designs for large commercial aircraft (Cofer, 2010). In the context of systems biology, model checking has been applied in order to verify and test hypothetical network models (Kwiatkowska et al., 2010; Klarner et al., 2012; Barnat et al., 2012).

From simulations, the biomass (and storage) properties of the plant can be determined. However, the choice of parameter values, i.e. thresholds, growth rates and storage rates, needs to be verified through model checking.

In our work, we used PRISM, a probabilistic model checker (Kwiatkowska et al., 2011), for verification of the plant–AMF model. In essence, model checking gives a true/false statement of whether a property is satisfied, while probabilistic model checking gives a probability of the property being satisfied. For example, we can test in our model whether the leaf biomass is above a certain value after growth has ceased, i.e. neither C nor P is a limiting factor.

4. Results

4.1. Simulations of plant only

It is generally accepted that low P levels in the soil result in larger roots whereas high P levels yield smaller root systems (Hutchings and de Kroon, 1994; Cahill and McNickle, 2011). Although this behavior seems straightforward in terms of the allocation strategies, the interdependence of the above and belowground growth processes may cause different behaviors in the model because of the common need for C and P. For this reason, we need to verify that the model displays the expected behavior.

Across all P uptake coefficients (1, 1/2, 1/3 and 1/4), simulations display the general behavior that growth of the plant in its entirety, i.e. leaves and roots, continues until neither C nor P is a limiting factor, at which time the biomass reaches a steady state (Fig. 3A and B). More specifically, the interdependent allocation strategies can be grouped into the growth phase, which is composed of the three strategies: (i) leaf growth/root growth, (ii) leaf growth/P storage and (iii) root growth/C storage, and the storage phase composed of the single strategy of C storage/P storage (Fig. 1). The switching between the three allocation strategies of the growth phase can be seen in the simulations (Fig. 3A and B), where growth temporarily ceases as shown by the intermediate constant values of the leaf and root biomass. By identifying when switching between strategies occurs, we can determine the duration of each strategy, e.g. how long the plant has been investing in leaf growth. Because we know how long the plant has been growing and the rate at which it is growing, we can determine the plant’s biomass value after completion of the growth phase.

Under the lowest P uptake coefficient, the growth phase is significantly greater than at the highest P uptake coefficient (P < 0.01, data not shown). Consequently, we observe a significant decrease in the
root biomass (Fig. 3D) with an increase in the P uptake coefficient ($P < 0.01$). However, the leaf biomass (Fig. 3C) does not change significantly ($P > 0.05$) with an increase in the P uptake coefficient, but there seems to be an increasing trend. In that sense, the expected behavior of larger roots in low P soil conditions still occurs even though the above and belowground allocation strategies are coupled.

4.2. Simulations of plant and fungi

The plant’s general behavior described in Section 4.1 is extended to incorporate the exchange relationship with AMF. That is, the exchange with AMF that influences the allocation strategies of the plant gives rise to the more complex symbiosis that can now replicate experimental data. We set the model parameters obtained from model checking to mimic the experimental conditions in Kiers et al. (2011) and ran fifty simulations of each set up.

First, Kiers et al. (2011) showed that plant above-ground biomass increased when associated with AMF showing greater cooperativity (i.e. lower C:P ratios) compared to less cooperativity (Fig. 4A and B). We ran simulations of our model under different AMF cooperation levels representing a no-AMF control ($x = 0$), less cooperative ($x = 1$) and more cooperative ($x = 2, 3, 4$) AMF analogous to Kiers et al. (2011). Our simulations displayed the pattern that leaf biomass when hosts are associated with more cooperative AMF is significantly higher than the leaf biomass of the control or less cooperative AMF ($P < 0.01$) (Fig. 4C). However, amongst the more cooperative AMF, the leaf biomass varies, namely the leaf biomass decreases going from cooperation level 2 to 3 and from 3 to 4. The same simulations in Fig. 4C were also used to measure the root biomass for the different cooperation levels (Fig. 4D).

Second, Kiers et al. (2011) demonstrated that plants allocated C to fungal partners according to the fungal species level of cooperation, but that C allocation patterns depended on whether the host was colonized by one or multiple AMF species (Fig. 5). Specifically, less cooperative AMF received more C (as indicated by higher fungal biomass) when they were the sole partner compared to when there was another more cooperative species present (Fig. 5A). We ran simulations of our model using the same low and high cooperation levels $x = 1$ and $x = 4$. Each cooperating AMF was simulated as being (i) the sole partner or (ii) in the presence of the other partner. Our simulation results match the data, where there was no significant difference for the more cooperative AMF under alone and mixed conditions ($P > 0.05$), but the less cooperative AMF received significantly more carbon when alone than in mixed conditions ($P < 0.01$, Fig. 5B).

4.3. Sensitivity analysis of plant–AMF model

We test the model over a range of parameter and property values to determine the probabilities of the leaf biomass being greater than a certain value (Fig. 6A and B).

To illustrate, we determined how the parameters of leaf growth rate and root growth rate affect the expected value of leaf biomass
(all other parameters are constant). Examining these relationships separately, the leaf biomass increases with increasing leaf growth rate, while leaf biomass is more or less invariant to changes in root growth rate (Fig. 6A and B). These data can be combined to generate a landscape of expected leaf biomass values given different root and leaf growth rates (Fig. 6C).

Landscapes such as in Fig. 6C and D can be used for model verification, where we can identify what levels of root and leaf growth rates result in an a priori specified level of leaf biomass. For example, if from experimental observations/data we expect to see a leaf biomass of 45, the landscape indicates that a leaf growth rate of 10 or 11 and any choice of root growth rate could yield a leaf biomass of 45 (Fig. 6C, light blue).

Once the parameters have been verified, the model can be used for model prediction, where we can identify the root and leaf growth rate levels needed to move from one leaf biomass level to another. For example, if we are interested in increasing leaf biomass from 45 to 60, the model tells us that increasing leaf growth rate would achieve the goal rather than changing the root growth rate (Fig. 6C).

Fig. 5. The experimental data (A) from the supplementary material of (Kers et al., 2011) measures the abundance of the different fungal species under similar situations, with the cooperative AMF R. irregularis and the less cooperative AMF G. custos and G. aggregatum. The simulation data (B) measures the amount of C exchanged to the cooperative and less cooperative AMF when only the single AMF is present (light gray) and when both AMF are present (dark gray).
In this example, the parameters of leaf growth rate, of root growth rate and the variable of leaf biomass is used for illustrative purposes, but any combination of parameters and variables can be used to verify the model. We also conducted sensitivity analysis on the parameters of storage, decay and thresholds and found that decay and storage had little effect on the root and leaf biomass whereas either of the thresholds would have greater influence over the root and leaf biomass.

5. Discussion and outlook

There have been many models of plant and AMF interactions that look at the allocation of nutrients based on environmental conditions. Approaches that focus on the teleonomic properties of the plant (Thornley and Parsons, 2014) and the N and C cycling in the soil (Meyer et al., 2012) are just two examples of other models that use differential equations to describe the symbiosis. Thornley and Parsons (2014) uses a teleonomic or optimization assumption, which is trying to induce the strategies from the emergent behavior while we are trying to deduce the growth through the strategies. Meyer et al. (2012) combines multiple approaches that model various processes in the soil including microclimate, soil chemistry and physiology. This does attempt to increase accuracy but at the expense of more parameters.

Here we present a novel approach of modeling phenotypic plasticity using probabilistic hybrid automata. The basic allocation strategy of investing in growth when a nutrient is limiting and in storage otherwise was observed in both the above and below-ground structures. The interdependence of these structures and their mutual dependence on C and P give rise to more complex behaviors when observing the plant as a whole. Nevertheless, the model can predict when the plant switches between the different allocation strategies (based on the initial conditions), and thus determine leaf and root biomass, whose values can then be used to verify the model parameters through model checking.

In particular, when we replicated experimental data derived from empirical work on the plant–AMF symbiosis, we observed two interesting patterns: (1) leaf biomass does not increase proportionally with level of fungal cooperativity (Cavagnaro et al., 2003); (2) there is a potential competition occurring between multiple AMF, where each AMF achieves its optimal yield. We discuss the mechanisms behind these two findings below.
When replicating experimental data, the simulations unexpectedly show that leaf biomass peaked at cooperation level $x=2$ (Fig. 4C). This counterintuitive behavior can be explained with respect to the mechanics of the model. The plant exchange increases at a greater rate with highly cooperative AMF creating a greater demand for $C$. To meet this demand, the plant invests in leaf biomass. In contrast, with a less cooperative AMF, the plant exchange rate does not increase as much; the plant does not have high demands for $C$, and thus the plant does not invest as much in the growth of leaves (Fig. 4C). Based on this reasoning, we would expect that the higher cooperation levels would continue to have an increase in the leaf biomass, which is what is observed in experiments (Figs. 4A and B). However, for cooperation values $x>1$ the growth duration begins to play a role (data not shown).

While the effect of being associated with cooperative vs. less-cooperative AMF on root biomass was not reported in Kiessler et al. (2011), we calculated root biomass from the same simulations that generated the leaf biomass data (Fig. 4D). We found that root biomass decreases as the degree of cooperation increases. This is expected because under a high level of cooperation, the $P$ demands of the plant are met by the AMF, reducing the need to invest in root biomass. This drastic decrease in root biomass also corresponds to a drastic decrease in the growth duration. This reduction in the growth duration means that less time is spent generating a demand for $C$ from the AMF and thus the leaf biomass reduces for larger cooperativity levels, $x>1$. In other words, optimizing the leaf biomass does not necessarily correspond to level of cooperation in AMF. Therefore, under our model, the most cooperative fungus in Kiessler et al. (2011) could be considered as any cooperative level $x>1$, while two less cooperative fungi can be described as $x=1$ (Figs. 4A and B and 5).

Fig. 5 shows the performance of the fungi when it is the only partner with the plant and when it is mixed with other fungi, namely a cooperative, $x=4$, and less-cooperative, $x=1$, fungi. An explanation for this behavior based on the model can be shown with a numerical example. Suppose we have the three AMF species that supply 5, 2 and 1 P units per time step and the plant has demands for 40. If all three exchange with the plant simultaneously then in 5 time steps the demands have been reached. If each species exchanges with the plant alone then there would be 8, 20 and 40 time steps respectively before the plant is satisfied. Comparing the time steps required by the more cooperative AMF when alone (8) and when mixed (5) is not significantly different ($P>0.05$). However, for the less cooperative, the time steps alone (20 or 40) and mixed (5) is significantly different ($P<0.01$). Because growth duration determines the amount of exchange with the AMF; these values are proportional to the amount of $C$ given to the AMF (Fig. 5B), which when multiplied by the corresponding exchange rates are proportional to the biomass of the AMF displayed in the experimental data (Fig. 5A).

Although the exchange values of the more cooperative AMF are not significant, we still expect from the numerical example above that the exchange value in the mixed case is less than in the alone case. More specifically, we expect a decrease in the exchange value from the alone to mixed case but with no change in variance (Fig. 5B). However, in the experimental data (Fig. 5A), we observe a much narrower variance in the mixed case than in the alone case. Following the previous argument, the mixed case appears to take the higher values out of the range of values expected from the simulations. The fact that this narrow variance is not replicated by the model could suggest that there is interaction or competition between the AMF that contribute to the plant or AMF’s exchange performance (Vandenkoonhyse et al., 2007; Engelmoer et al., 2014). An analogous example would be bidding for $C$ in an auction, where an AMF does not need to bid high when alone but when there are other AMFs, each AMF is forced to bid to their upper limit in order to still receive $C$ (Wyatt et al., 2014).

Future extensions of the model could include incorporating AMF strategies with varying nutrient concentrations in the environment and single AMF colonizing multiple host-plants. Also, model extensions could distinguish the processes of storage and reproduction and then model the system as it evolves over generations.

Depending on the ecological question, we could include changes in spatial (Boswell and Davidson, 2012) or temporal dimensions, or incorporate more resources such as those relating to global change (e.g. drought stress, nutrient deposition).

Further experimental data could extend and fine tune the model so that the model can be used for prediction. In the agricultural context, for example, we could use the model to test different combinations of plants and AMF in order to design crop combinations: specific variables might be optimized according to the preferred output, e.g. leaf biomass for lettuce or spinach, root biomass for radish or carrot and storage (also associated with reproduction in our model) for potatoes (because of stored starches). In other words, the abstract form of the model gives us the ability to experiment with different crops and crop combinations.

6. Conclusions

From our results, we observed how modeling phenotypic plasticity, namely resource allocation and exchange with AMF, provides enough information to describe some complex behaviors such as the strength of symbiosis with AMF under different experimental conditions (Fig. 5). Furthermore, the theoretical model allows us to discuss, in detail, an alternative interpretation of the data based on the processes and strategies that are present in the system. As many organisms display some form of phenotypic plasticity, there is great potential of the method to be used in modeling of other ecological systems, such as interactions with diverse microbes such as endophyte communities.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ecolmodel.2015.04.021

References


