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The Genetics of Ecologically Relevant Interactions between Various Plants

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Abstract

Plant interactions have long been acknowledged as a key factor in plant community dynamics and crop output. Surprisingly, we still know very little about the ecological genetics underlying variation in plant-plant interactions. The international Plantcom network selected four pertinent questions to promote a deeper understanding of the mechanisms mediating plant assemblages in this opinion piece by scientists from complementary disciplines. We suggest that in order to more accurately predict genotype-by-genotype interactions with the environment and model stable and productive plant assemblages in both wild habitats and agricultural fields, it is necessary to identify the key relationships between the phenotypic traits involved in plant-plant interactions and the underlying adaptive genetic and molecular pathways [1].

Keywords: Pleiotropy; Quantitative genetics; Univariate; Multivariate; Environmental stresses; Plant communities

Introduction

Most plant species, including nearly all of the world's significant crops, are symbiotically connected to fungi of the subphylum Glomeromycotina through arbuscular mycorrhizal (AM) symbiosis. Agriculture 2, 3, as well as defining plant fitness, variety, and cohabitation in natural groups 4, 5, all depend on the symbiosis because of its impacts on plant nutrient acquisition and growth. Here, we argue that quantitative genetics, and more specifically, an integrative approach combining quantitative genetics of the plant host and its fungal partner, could be used to better use the symbiosis to improve agricultural production or to better apply AM fungi (AMF) in ecological restoration or conservation. We look at how quantitative genetics has developed and how it might be applied to both partners before providing a conceptual framework in order to integrate them. We recognize that many ecosystem ecologists and agronomists probably do not find the subject or technical details of quantitative genetics techniques to be particularly approachable, but we hope to increase their awareness of the potential benefits of using an integrated quantitative genetics approach to understand the symbiosis in agriculture and ecosystem ecology [2].

Approaches in quantitative genetics are not new. This theoretical and practical framework for identifying genes causing traits that do not inherit according to simple Mendelian principles was developed by Fisher and Wright in the early 20th century6, 7. These traits are those caused by the cumulative effects of numerous genes. The factors influencing the phenotype could be predicted by taking into account continuously varying phenotypes from various individuals in one type of organism and connecting them to minute variations in each individual's genetic make-up. For instance, height is a quantitative feature that is influenced by between 50 and 200 genes in both plants and animals 8, 9. It is crucial to identify the genes that cooperate to produce an observed characteristic, especially if the trait is crucial to agricultural or environmental activities. A desired trait in symbiotic relationships, such as the AM symbiosis, may result from a combination of genes from both the plant and the fungus, not just one of the partners. Such a feature might be used in agriculture to increase crop productivity or a crop's resistance to challenges like infections and drought. In ecosystem ecology, desirable characteristics can include those that promote increased nutrient cycling or better below-ground delivery of organic carbon, for example. We support research efforts in this area because of the improved possibility to comprehend the AM symbiosis utilising genetic data that could be offered by quantitative genomics methodologies.

Further association studies can be carried out to uncover additional genetic loci that interact with the initial gene candidate, starting with a known gene candidate that may have been developed using a forward genetics strategy. Starting with the plant gene CASTOR, which is known to play a role in the growth of the AM symbiosis, a recent study serves as an example of this. To maximize the crop's agriculturally advantageous traits, other genetic loci (i.e., combining gene candidates) that interact with the presence of CASTOR (i.e., mycorrhization) in maize were discovered using a quantitative genetics approach on a population made up of half-plants with intact CASTOR and half without. The loci of interest were classified as either changing the advantage of maize production or its dependence on the presence of CASTOR. Given that maize is currently the second-most produced crop in the world in terms of tones, this is obviously of relevance. In this illustration, only plant genes are used. However, given the significant impact of both fungal variation and plant genotype on the outcome of the symbiosis, we anticipate that the key to engineering such traits of interest to agriculture and ecosystem ecology will be finding gene combinations of both partners that act synergistically [3].

Material and Methods

The field study was carried out in a freshwater marshland in Anqing City, Anhui Province, which has a subtropical monsoon climate (116°59′27″ E, 30°28′08″ N). The average annual temperature is 16.7°C, and the average annual precipitation is 1500 mm. The Yangtze River is close by the location. Due to microtides, this marsh regularly experienced flooding from May to the middle of August. Phragmites australis (Cav.) Trin. ex Steud, Polygonum pubescens Blume,

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Kummerowia striata (Thunb.) Schindl, Leonurus artemisia (Lour.) S. Y. Hu, Ixeris polycephala Cass, and Conyza canadensis (Linn.) Cronq make up the majority of the vegetation.

From April to August 2012, a field experiment was carried out. The selection process involved five co-occurring plant species that varied in their distribution densities and environmental optimums. Forty 0.5 m 0.5 m quadrats were set up in the field in the middle of April for each plant species. Each species' 40 quadrats were divided into 10 blocks. Quadrats were then at random assigned to one of the following two combinations for each block: (2) Two levels of neighbor treatment: neighbor removal versus neighbor present. (1) Two degrees of AMF: benomyl application as opposed to control.

By contrasting the performance of the target plants with and without neighbors, the neighbor removal treatment was utilised to evaluate plant-plant interactions. For each of the five target plants, individuals with the same shoot size and leaf count (within species) were chosen in April, the start of the growing season [4]. In order to reduce the impact of surrounding pairings, plant pairs were placed between 1 and 3 metres apart to minimize variations in microclimate. Following a random selection of the target individuals, the neighbour missing treatment involved clipping the aboveground portion of neighboring plants and removing them.

In the benomyl application treatment, the soil in each quadrat of that treatment was treated with the fungicide benomyl (2 g material, with 50% active ingredient, diluted in 2 L of tap water). This treatment's goal was to reduce AM colonisation. The control quadrats received the same volume of plain tap water without any fungicide added. Since the study location (marsh) was submerged from May to the middle of August due to microtides and benomyl cannot be restricted inside the treatment quadrats, benomyl was not applied.

On September 10, after a season of waterlogging, the experiment's target plants were harvested. Separated from the roots, the shoots were oven dried at 80°C for 48 hours before being weighed. The type and level of plant interactions were reflected by relative interaction intensity (RII). The target biomasses Xe and Xr are determined by the presence or absence of neighbors, respectively, and are used to calculate RII. Limits [1, +1] are used to define the RII. Positive RII values suggest that facilitation is in control, whereas negative RII values suggest that competition is in control.

Root samples from the target plants were taken to test for mycorrhizal colonisation in order to gauge the efficacy of the fungicide benomyl. Giovannetti and Mosse's modified gridline intersection approach was used to determine root colonisation by AMF. Shortly after being cleaned in 10% KOH (w/v), stained in acid fuchsin, and scored for the presence or absence of mycorrhizal infection (arbuscules, vesicles, coils, or hyphae) under a compound microscope at a magnification of around 200, the roots were examined. AMF colonisation (%) = number of intersections colonized (hyphae, arbuscules, vesicles, and hyphal coils)/total intersections examined 100% was used to calculate the AM colonisation level [5].

Soil samples from control and fungicide application plots were collected in order to assess potential adverse effects of the fungicide on soil nutrients and soil microbes. Measurements were made of soil nitrogen and phosphorus, enzyme activity, and culturable fungal units. ANOVA with randomised blocks was used to analyses the data for shoot biomass, RII, and arbuscular mycorrhizal fungal colonisation, with four treatments making up each block. For each analysis, the homogeneity of group variance and the normality of model residuals were examined.

I. polycephala was selected as the test plant in this study to determine whether the effect of AMF was dependent on soil oxygen levels and whether the presence of P. australis could raise soil oxygen levels and support the function of AMF. I. polycephala's growth was heavily reliant on AMF, and I. polycephala has with P. australis there is a strong interspecies relationship. The vertical distribution of I. polycephala's root system overlaps with that of P. australis (both are largely located between 0.1 m and 0.3 m), which is another factor in the selection.

In September 2012, soil oxygen concentrations were measured and root samples of I. polycephala were collected from 30 randomly selected quadrats (1 m 1 m) in the low marsh (nearly submerged daily, c.0.3 m above mean low tide), in order to quantify the relationship between the oxygen concentration and mycorrhizal status. We dug up roots to collect them. transporting the entire plant to the lab after digging the soil down to a depth of 0.2 m. Only the roots that were plainly attached to shoots were utilised to quantify AM fungal colonisation in order to prevent the mixing of roots from different plant species. By gently pressing a Clark type glass microelectrode (500 m tip, Unisense A/S Aarhus N, Denmark) into the silt, oxygen concentrations were determined. A micromanipulator was used to place the microelectrode, and a picoammeter (PA2000, Unisense A/S) was used to detect the sensor current. Both air- and oxygen-free N2-saturated water were used to calibrate the microelectrode at the same temperature as the sediment.

In September 2012, P. australis populations were measured in 30 quadrats (1 m 1 m) randomly placed in the low marsh (nearly daily inundated, c.0.3 m above mean low tide) to quantify the link between P. australis density and oxygen content. As mentioned above, soil oxygen levels were tested.

A separate field experiment was carried out (April 2012 to September 2012) to determine if the presence of P. australis affected the oxygen concentration and mycorrhizal status of I. polycephala. This was done to experimentally test for the effects of P. australis on soil oxygen concentration and AMF colonisation of I. polycephala. Twenty 1 m 1 m randomly chosen plots were chosen in a marshy location. P. australis was eradicated by cutting the aboveground in ten of these plots in April [6]. The microelectrode was placed in the plot's centre, close to the root of I. polycephala, to monitor the oxygen level after five months. I. Polycephala roots were harvested from each plot, and the AMF association was measured as previously mentioned.

A factorial experiment was carried out to test the idea that P. australis influences mycorrhizal mutualism, which in turn impacts I. polycephala growth (from April 2012 to September 2012). In the middle of April, forty 0.5 m 0.5 m quadrats were set up in the field. Ten blocks were created from the 40 quadrats. Quadrats were then at random assigned to one of the following two combinations for each block: Two levels of neighbor treatment (all neighbors removed versus all neighbors removed but with P. australis present) and two levels of AMF (benomyl application versus control) are described here. Application of benomyl was altered as in experiment 1. For the P. australis neighbor present treatment, P. australis individuals were kept in the plot and all neighbors of the other species were removed, and then the neighbor effect reflected the interaction from the P. australis neighbor. In the all neighbors' removal treatment, an individual of I. polycephala was chosen as the target plant, and all of the neighbors were removed by cutting the aboveground part. In September, I. polycephala individuals that were targets were harvested in order to weigh shoot biomass and gauge AMF colonization [7].

SAS software was used to conduct linear correlations between oxygen concentration and mycorrhizal status as well as between P. australis density and oxygen concentration (SAS Institute Inc., NC, USA). The elimination of P. australis on soil oxygen and the colonisation of AMF on I. polycephala were both tested using the t-test. In the two-factor design, the GLM techniques were employed to compare the AMF colonisation and shoot biomass of the target I. polycephala.

Discussion

The field study was carried out in a freshwater marshland in Anqing City, Anhui Province, which has a subtropical monsoon climate (116°59′27″ E, 30°28′08″ N). The average annual temperature is 16.7°C, and the average annual precipitation is 1500 mm. The Yangtze River is close by the location. Due to microtides, this marsh regularly experienced flooding from May to the middle of August. Phragmites australis (Cav.) Trin. Ex Steud, Polygonum pubescens Blume, Kummerowia striata (Thunb.) Schindl, Leonurus artemisia (Lour.) S. Y. Hu, Ixeris polycephala Cass, and Conyza canadensis (Linn.) Cronq make up the majority of the vegetation [8].

From April to August 2012, a field experiment was carried out. Five co-occurring plant species that varied in their distribution densities and environmental optimums were picked. Forty 0.5 m 0.5 m quadrats were set up in the field in the middle of April for each plant species. Each species' 40 quadrats were divided into 10 blocks. Quadrats were then at random assigned to one of the following two combinations for each block: (2) Two levels of neighbor treatment: neighbor removal versus neighbor present. (1) Two degrees of AMF: benomyl application versus control.

By contrasting the performance of the target plants with and without neighbors, the neighbor removal treatment was used to evaluate plant-plant interactions. For each of the five target plants, individuals with the same shoot size and leaf count (within species) were chosen in April, the start of the growing season. In order to reduce the impact of nearby pairs, plant pairs were placed between 1 and 3 metres apart to minimize variations in microclimate. Following a random selection of the target individuals, the neighbor absent treatment involved cutting the aboveground portion of neighboring plants and removing them.

In the benomyl application treatment, the soil in each quadrat of that treatment was treated with the fungicide benomyl (2 g material, with 50% active ingredient, diluted in 2 L of tap water). This treatment's goal was to reduce AM colonisation. The control quadrats received the same volume of plain tap water without any fungicide added. Since the study location (marsh) was submerged from May to the middle of August due to microtides and benomyl cannot be restricted inside the treatment quadrats, benomyl was not applied [9].

On September 10, after a season of waterlogging, the experiment's target plants were harvested. Separated from the roots, the shoots were oven dried at 80°C for 48 hours before being weighed. The type and level of plant interactions were reflected by relative interaction intensity (RII). The target biomasses Xe and Xr are determined by the presence or absence of neighbors, respectively, and are used to calculate RII. Limits [1, +1] are used to define the RII. Positive RII values suggest that facilitation is in control, whereas negative RII values suggest that competition is in control [10].

Root samples from the target plants were taken to test for mycorrhizal colonisation in order to gauge the efficacy of the fungicide benomyl. The gridline intersection method as modified by Giovannetti and Mosse was used to determine root colonisation by AMF [52]. Shortly after being cleaned in 10% KOH (w/v), stained in acid fuchsin, and scored for the presence or absence of mycorrhizal infection (arbuscules, vesicles, coils, or hyphae) under a compound microscope at a magnification of around 200, the roots were examined. AMF colonisation (%) = number of crossings colonized (hyphae, arbuscules, vesicles, and hyphal coils)/total intersections inspected 100% was used to establish the AM colonisation level.

Soil samples from control and fungicide application plots were collected in order to assess potential adverse effects of the fungicide on soil nutrients and soil microbes. Measurements were made of soil nitrogen and phosphorus, enzyme activity, and culturable fungal units [11].

ANOVA with randomised blocks was used to analyse the data for shoot biomass, RII, and arbuscular mycorrhizal fungal colonisation, with four treatments making up each block. For each analysis, the homogeneity of group variance and the normality of model residuals were examined. I. polycephala was selected as the test plant in this study to determine whether the effect of AMF was reliant on soil oxygen levels and whether the presence of P. australis could raise soil oxygen levels and support the function of AMF. I polycephala has a strong interspecies connection with P. australis and its growth was heavily dependent on AMF. The vertical distribution of I. polycephala's root system overlaps with that of P. australis (both are largely located between 0.1 m and 0.3 m), which is another factor in the selection.

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A factorial experiment was carried out to test the idea that P.

australis influences mycorrhizal mutualism, which in turn impacts I. polycephala growth (from April 2012 to September 2012). In the middle of April, forty 0.5 m 0.5 m quadrats were set up in the field. Ten blocks were created from the 40 quadrats. Quadrats were then at random assigned to one of the following two combinations for each block: Two levels of neighbor treatment (all neighbors removed versus all neighbors removed but with P. australis present) and two levels of AMF (benomyl application versus control) are described here. Application of Benomyl was manipulated. For the P. australis neighbor present treatment, P. australis individuals were kept in the plot and all neighbors of the other species were removed, and then the neighbor effect reflected the interaction from the P. australis neighbor. In the all neighbors removal treatment, an individual of I. polycephala was chosen as the target plant, and all of the neighbors were removed by cutting the aboveground part. There were 10 blocks created from the 40 quadrats. The following two combinations of the following two factors were then randomly assigned to quadrats for each block: Two levels of neighbor treatment are used in (1) two levels of AMF (benomyl application versus control) and (2) two levels of neighbor removal (all neighbors removed versus all neighbors removed but with P. australis present). Application of Benomyl was tampered with. For the P. australis neighbor present treatment, P. australis individuals were kept in the plot while all neighbors of the other species were removed, and the neighbor effect then reflected the interaction from the P. australis neighbor. For the all neighbors removal treatment, an individual of I. polycephala was chosen as the target plant, and all of the neighbors were removed by cutting the aboveground part.

Conflict of Interest

None

Acknowledgement

None

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