How phylogenetic relatedness and floral traits are involved in heterospecifc pollen effect in an artificial co-flowering community

Jose B. Lanuza [1,2] true

[1] *US Environmental Protection Agency, Office of Research and Development, National Health and Environmental Effects Research Laboratory, Atlantic Ecology Division, 27 Tarzwell Drive Narragansett, RI, 02882, USA*

[2] *Big Name University, Department of R, City, BN, 01020, USA*

[3] *Estacion Biologica de Donana (EBD-CSIC), E-41092 Sevilla, Spain*

[*] *corresponding author: barragansljose@gmail.com*

Possible journals to publish: New phytologist, journal of ecology, oikos...

Keywords: heterospecific pollen, plant reproduction, fitness, competition

INTRODUCTION

Paragraph 1

In natural systems plant species normally coexist and share their floral visitors with other species (Bascompte et al., 2003). This pollinator sharing from the plant perpective can be negative due to competition (refs) or positive due to facilitation (refs). Moreover, once the pollinator has landed on the stigma some other issues for the species fitness may arise, the arrival of foreign pollen and conspecific pollen loss.

Paragraph 2

The effect of heterospecific pollen has been widely studied (refs). Native species are supposed to have greater negative effects than native ones Arceo-Gómez and Ashman (2016)

species-specific. What kind of pollen interaction? Size just mechanical?

Paragraph 3

Relatedness. What we know?

Paragraph 4

The great difficulty of working with pollen in a coflowering community make the understanding of heterospecifc pollen effect a real challenge. For this reason we have created an artificial co-flowering community in a glasshouse to test the effect with all the possible combinations among them. Where we test the folowing hypothesis: 1) Does heterospecific pollen reduce seed set, if so, 2) Does heterospecific pollen effect depend on the relatedness of the species, 3) Does heterospecific pollen effect depend on any floral trait?

METHODS

The study was conducted in a glasshouse at University of New England (Armidale, Australia) from November 2017 to March 2018. Rooms were temperature controlled depending on the requirements of the species with day and night temperature differences. The species selected (Table 1) belonged to three different families, Solanaceae, Brassicaceae and Convolvulaceae. The criteria of species/family selection was based on close/distant related species (see phylogenetic tree for relatedness fig 1), heterogeneous traits, low structural flower complexity and fast life cycle. For the purpose of the experiment all the species where considered as pollen recipient and as pollen donor (see interaction matrix, fig 2). Species were watered once or twice per day and fertilized weekly (NPK 23: 3.95: 14).

Hand-pollination

Foreign pollen effect was studied through two different treatments, one with 50% conspecific pollen and 50% heterospecific pollen and a second one with 100% foreign pollen (N=10). The proxy of effect were seed set and “pollen tubes”. Moreover, hand cross pollination, hand self pollination, apomixis (bagged emasculated flowers) and natural selfing were tested (N=10). Flowers were emasculated the day prior anthesis and hand pollinated next day with a toothpick. Had-pollination was realized with 3-4 gentle touches on the surface of the stigma. The mixes of pollen were performed on an eppendorf based on the pollen counts maded with Neubaeur chamber (each anther was counted 4 times for 20 different anthers per species).

Evolutive distance

Two types of evolutive distances were calculated with MEGA7 thow kinds of markers: 1) Internal transcribed spacer (ITS) and 2) ribulose-bisphosphate carboxylase (RBCL)

Traits

Several traits of the ten species were measured. Pollen per anther was counted, number of ovules, stigma width and length and stigmatic area, style width and length, ovary width and length. Moreover stigma type was tested. Self-incompatibility was

We used the statistical language `R` (R Core Team 2018) for all our analyses. These were implemented in dynamic rmarkdown documents using `knitr` (Xie 2014, 2015, 2018) and `rmarkdown` (Allaire et al. 2018) packages. All the multilevel models were fitted with `lme4` (Bates et al. 2015).

RESULTS

DISCUSSION

Discuss.

CONCLUSIONS

ACKNOWLEDGEMENTS

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