

The experimental test addresses the question of the adaptation of an insect plant-parasite to a local host, both domestic and wild, and to its local climate. The experimental design aims to answer five questions related to the adaptation of the parasitic apple aphid *Dysaphis plantaginea* on apple tree species *Malus sp.* As a large-scale study with replication at three garden orchard sites and many individual replicates, this multi-genotypic cross-infection experiment has very promising potential. The whole set-up and preparation of the material for the experimental test scheduled for March 2021 already began in 2019 with the growth of wild or domesticated apple scions and the breeding of aphids. The plants were transferred in early 2020 to each site. Global planting was carried out with a randomization of the plots to avoid any plot combination effect as much as possible and will be included in the model. I believe the study was well thought and measured at a large scale in order to optimize the repetability and homogeneity between experimental sites, apple trees and aphid treatments and preparation. As it is an ambitious project, the work load is very important and the inoculating experiment will thus be conducted over 18 days. As this may as well cause important variability, the days will be integrated in the statistical model as a random effect.

I have though some minor comments / concerns about the experimental analyse.

Fitness variables. I believe authors should be more accurate about the fitness variable acquisition and measure. Authors specify : "We will estimate rosy apple aphid fitness as the growth rate of the colony (cumulative number of nymphs produced per surviving females produced on the infested plant; Warneys et al., 2018), and the insect life cycle (aphid stages (L1 to L5), apterous adults, nymphs, and winged forms (Angeli and Simoni, 2006))."

First, it is not completely clear to me at what time points the fitness measurement is occurring or in other words are you following the cumulative number every days since inoculation and for how long?

Second, it is not described how experimentators will combine growth number (nymph/viable mother) and cycle development (aphid stages). They should indicate if they will be using both data independently for modelling and what they expect for their distribution. One particular question is : how do you account for the 5 aphid stages in the model? Do you model for 5 dependant variables? or do you run multivariate models?

Apple trees description. Since Chen et al is not available yet, authors should precise how the choice of wild apple tree was done for this approach. It seems to me according to authors' previous data (Cornille et al. 2015) that *Malus sylvestris* shows about so five distinct european genetic clusters among which only three are present in Spain/France/Belgium. Are all the selected wild genotype among those three groups?

Also, while it is not the main focus of the study, is there any other easy accessible genomic data that could be utilised regarding resistance such as known R genes / QTLs presence on those tree accounting for parasite fitness?

Multigenotype test per single trees and systemic response. Authors should consider the systemic response within individuals and maybe to a less extent between individuals (supposedly through metabolite exudation as reported in some studies about rhizosphere plant/plant communication). Testing 9 genotypes per tree will induce systemic responses and possibly decrease quite some variability that could be perceived testing single inoculation per tree. As much as I understand the setup of the study to maximise the number of combinaisons with minimal number of tree, it has to be specified and accounted for.

Tree ecophysiological traits to be included in the model. Authors should indicate how they will account for tree development and ecophysiological traits (measurements on modality 2). It does not

seemed to be appearing in any of the tested models. Could it be added in the model as nested(?) covariates for each apple tree genotype?

Overall this project seems very promising and will certainly result in important and inovative findings regarding parasite/plant adaptation and speciation in wild and domesticated trees. I am looking forward to seeing the results. Also, as findings may result in large datasets that could be further decomposed in several publication, autors will have to indicate any change in their expected publication scheme.