LabEx BASC
Biodiversité, Agroécosystèmes, Société, Climat

EVOFUNGI

Assessing the evolvability of a pathogenic fungus in the context of global change

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Objective

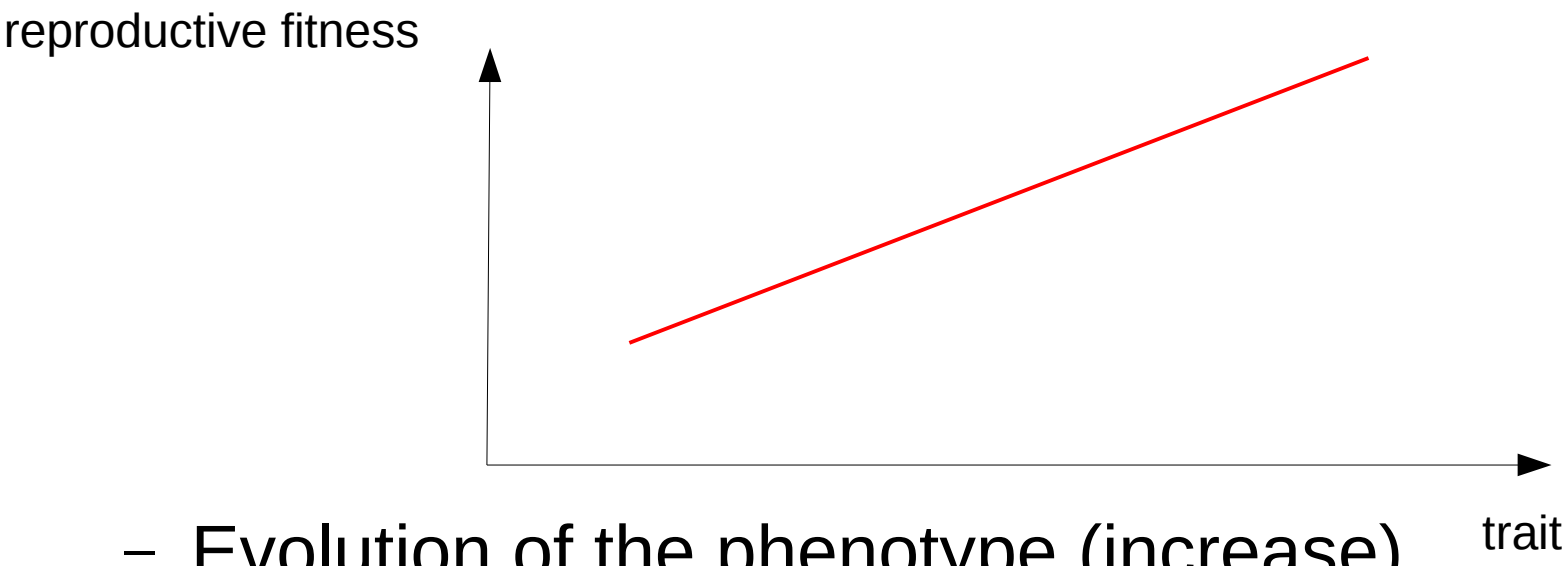
- Describe and understand the evolution of genetic architectures under fluctuating selection

- Two research lines:
  - Experimental evolution
  - Theoretical models

- Timing: 2014 - 2016

Theoretical context

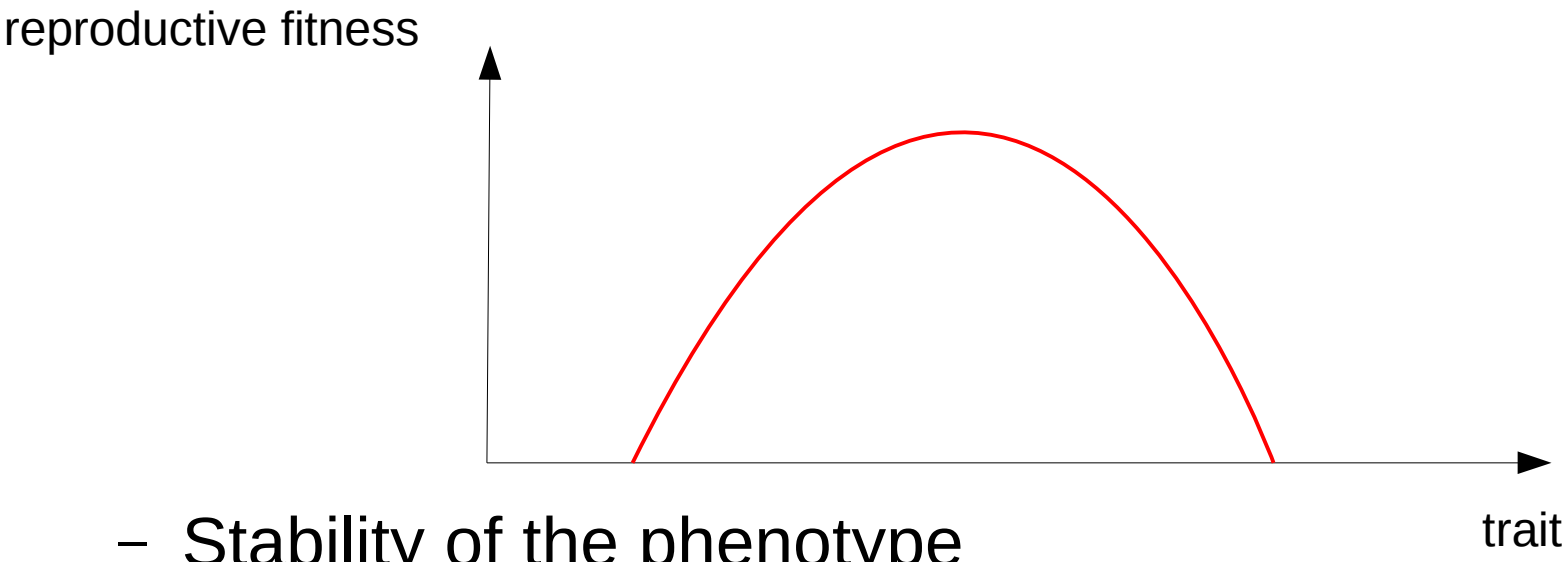
- Several patterns for natural (artificial) selection:
  - Directional selection



  - Evolution of the phenotype (increase)
  - Evolution of the genetic architecture (decanalization)

Theoretical context

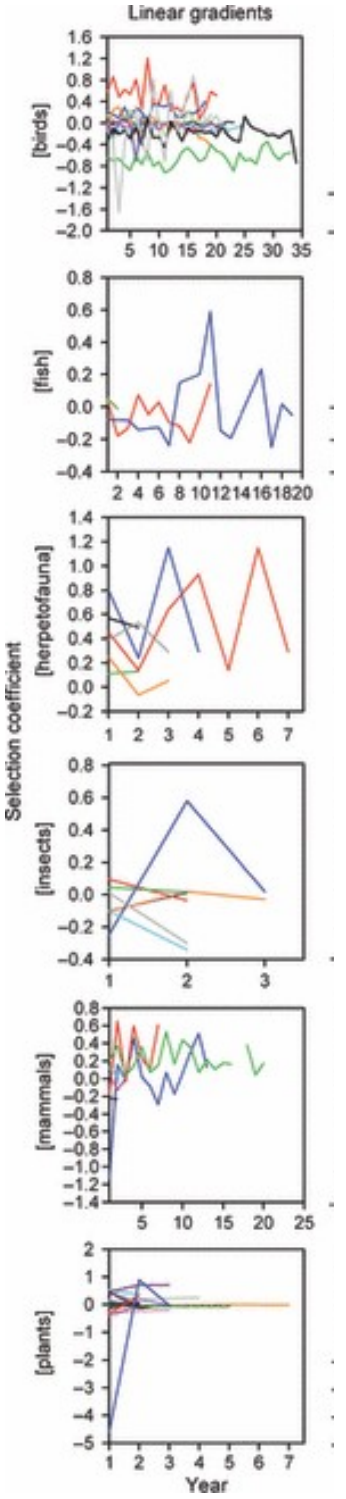
- Several patterns for natural (artificial) selection:
  - Directional selection
  - Stabilizing selection



  - Stability of the phenotype
  - Evolution of the genetic architecture (canalization)

Theoretical context

- However, “real” natural selection might be more complex.
- Fluctuating selection might be pervasive.



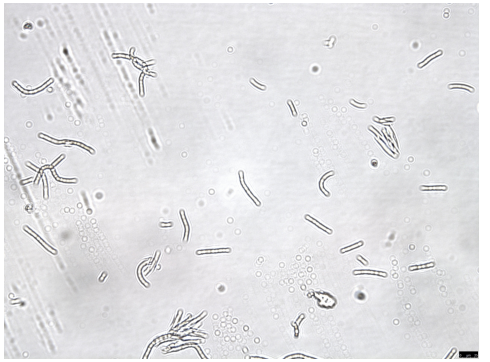
Review by AM Siepielski, JD DiBattista & SM Carlson Ecology Letters 2009

Theoretical context

- However, “real” natural selection might be more complex.
- Fluctuating selection might be pervasive.
- No clear theoretical background
  - “Intuition”: adaptive decanalization
  - Simple model results: similar to stabilizing selection

Le Rouzic, Alvarez-Castro, Hansen (2013) Evolutionary Biology

Experimental setup



- Experimental evolution in *Mycosphaerella graminicola*
  - One of the main pest of wheat
  - Small haploid genome (40 Mb, 13 + 8 chromosomes), reference genome available
  - A lot of standing genetic variation (7 SNP/100bp!)
  - We have the full sequence of our lab strains

Experimental setup

- Experimental evolution in *Mycosphaerella graminicola*: 2 strains X 3 treatments

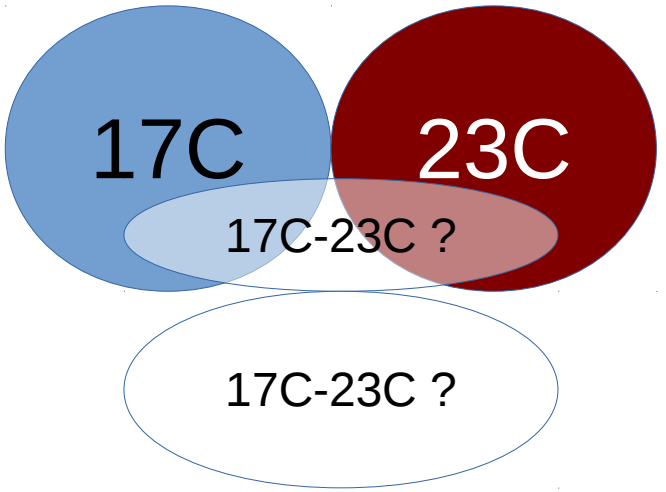
Test two selection regimes:

- Stable selection (constant temperature: 17C or 23C)
- Fluctuation selection (temperature fluctuates between 17 and 23C: switch every 3 days)

Duration of the experiment: One year (expectations: ~ 300 generations: hypothesis it is enough time to select new mutations and for the mutations to reach fixation)

Experimental setup

- “Phenotypic” measurements:
  - RNA seq at the end of the experiment
    - Change in gene expression
    - Change in gene sequences

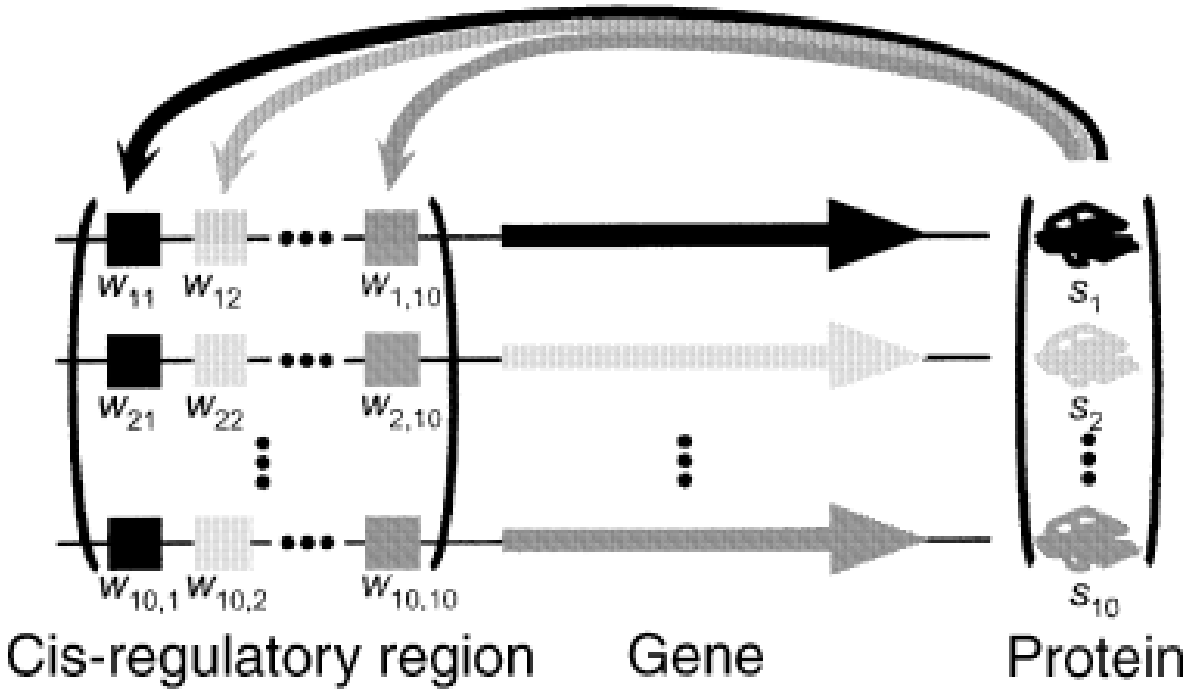


Experimental setup

- “Phenotypic” measurements:
  - RNA seq at the end of the experiment
    - Change in gene expression
    - Change in gene sequences
  - Fitness change during selection (adaptation)
    - Exponential growth rate
    - Competitive fitness
- *in plantae* tests
  - Does fluctuating selection promote evolvability in general?

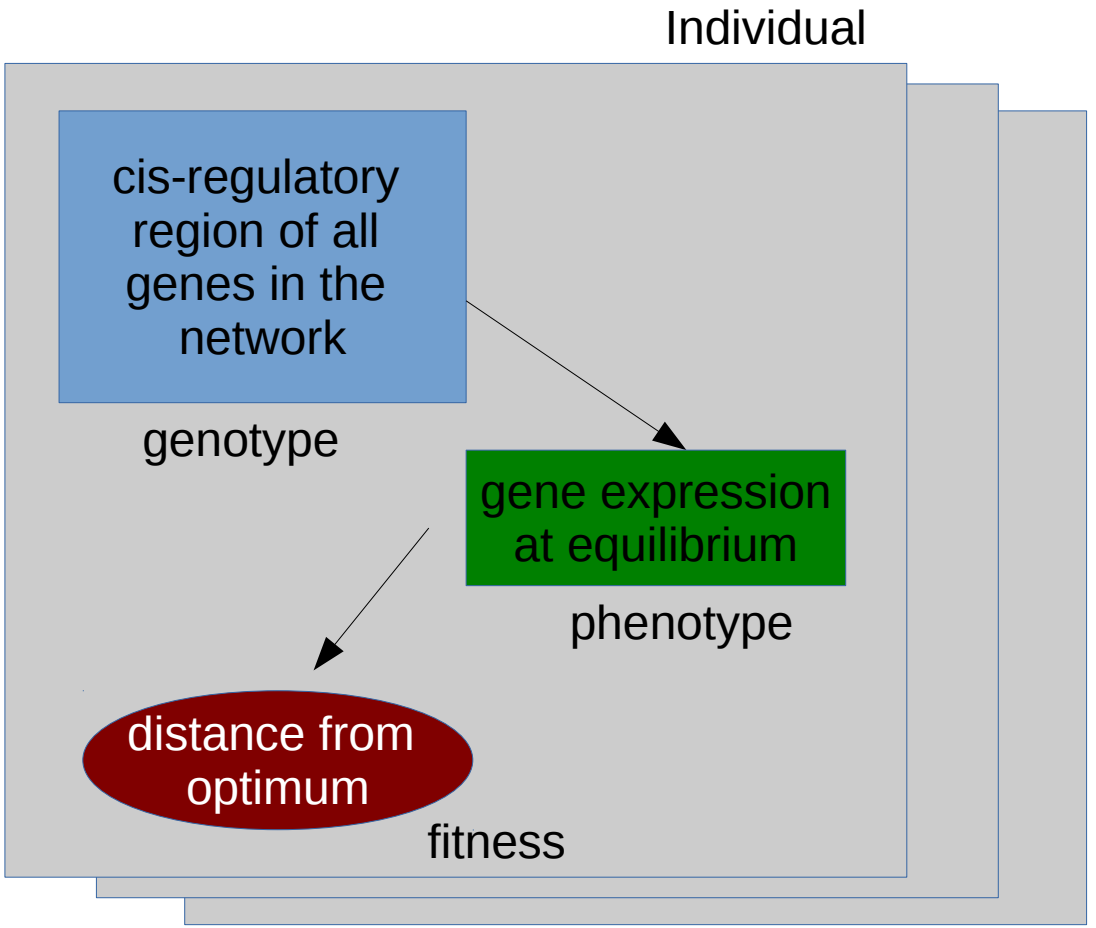
Theoretical models

- Gene regulatory network model



Theoretical models

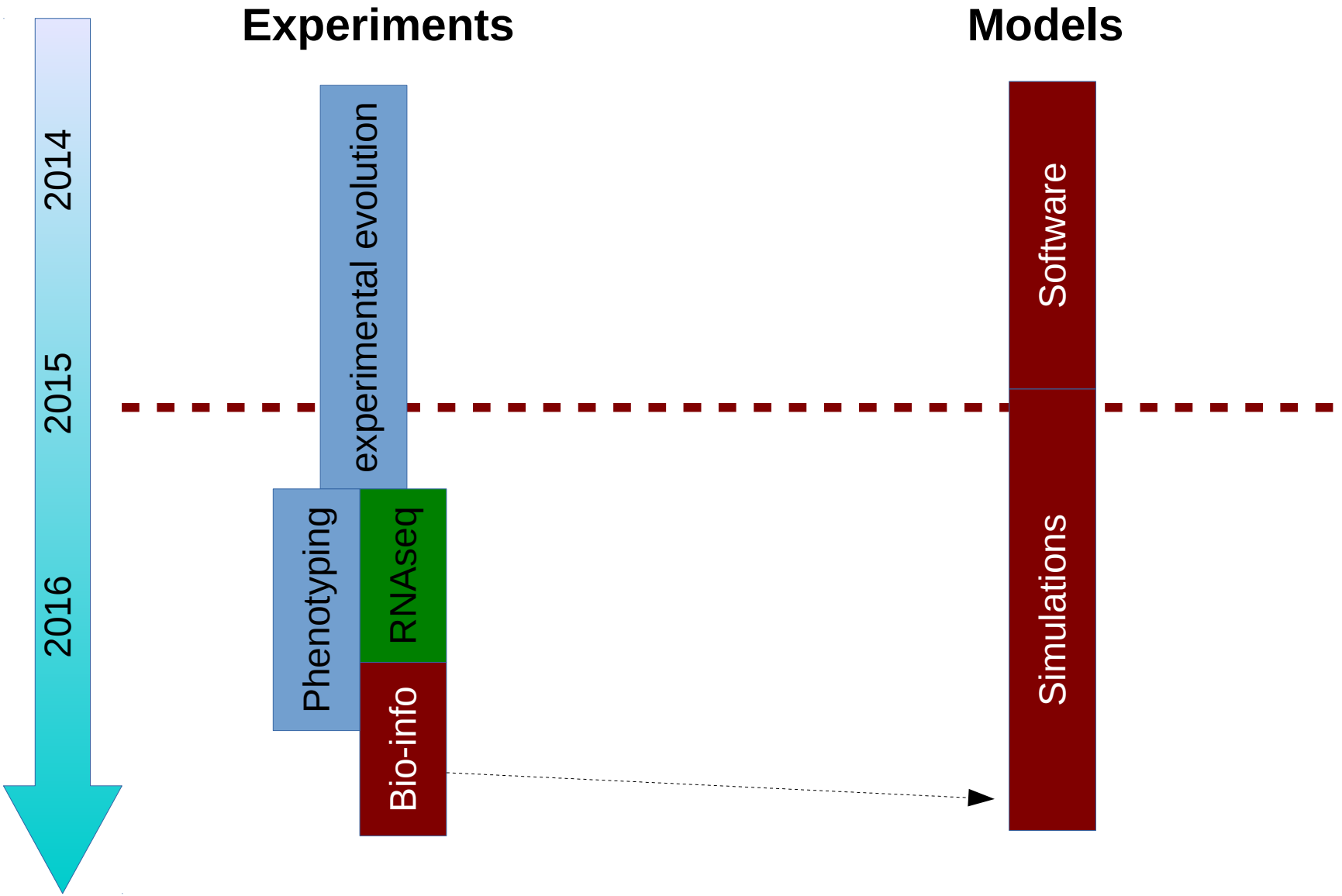
- Gene regulatory network model
- Individual-based simulations



Theoretical models

- Gene regulatory network model
- Individual-based simulations
  - Stabilizing selection for opt1
  - Stabilizing selection for opt2
  - Fluctuating selection between opt1 and opt2
- Gene network
  - Random according to some general rules (connectivity...)
  - From empirical input

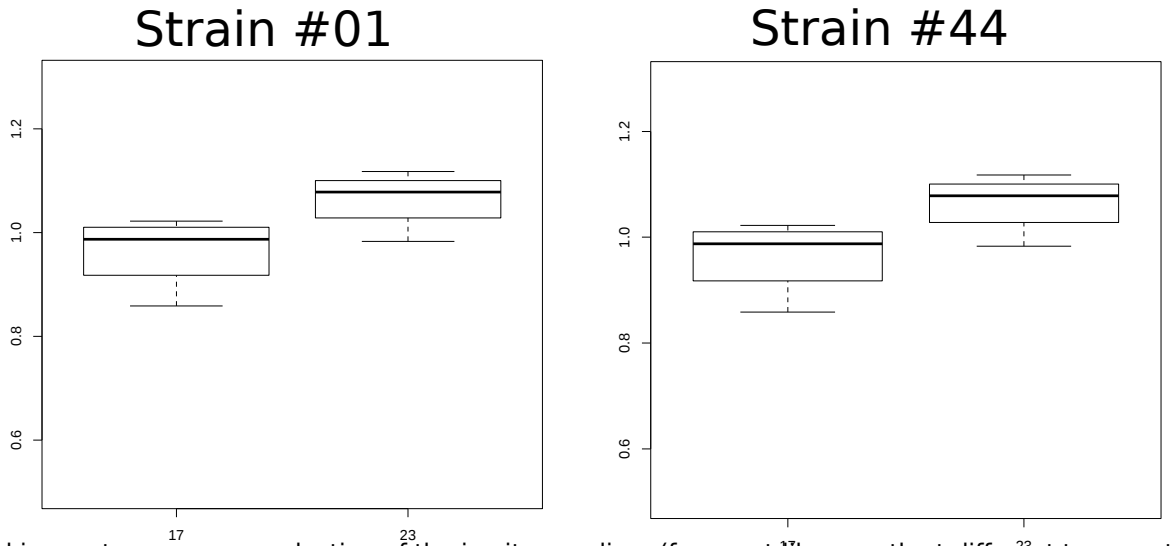
Timeline


Experiments
Models
2014
2015
2016
experimental evolution
Phenotyping
RNAseq
Bio-info
Software
Simulations

Supplementary slides

Experimental Evolution: design

- Two strains of *Mycosphaerella graminicola* from the South of France (# 01 and #44) collected in 2010
- In vitro growth prior to the experiment at 17C and 23C



data from 4 bioreplicates after 10days (y-axis: growth (optical density; x-axis: temeprature)

- Technical issues to overcome: selection of the in vitro medium (for yeast like growth at different temperatures), volume of medium for no genetic drift to occur

- Goals of the experiment: mutation accumulation under selection: mutation occurs through asexual reproduction only (*when in vitro..*)
- phenotype “yeast-like”

in our growth conditions

under the microscope G*100

RNA seq data analysis

- mapping using reference genome (strain ipo323) and DNA re-sequencing data of the selected strains (tool: Bowtie)
- seek genes differentially expressed between samples : target of selection? (tools: Deseq and EdgeR)
- seek mutations in genes (may be *trans* regulators)
- seek regulatory networks (based on co-expression information and link with *trans* effect mutations)); epistasis
- seek the distribution of gene expression, GO overrepresentation in different selection regime

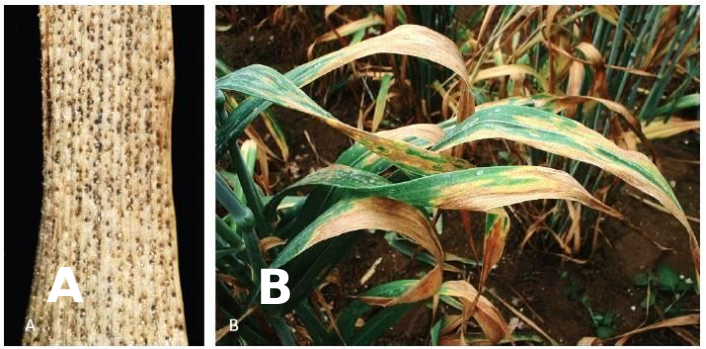
  - > next: re-sequencing of *cis*-regulatory regions of most differentially expressed genes to find new selected mutations
  - > (these data also contributes to the knowledge on natural variation of gene expression)
  - > (these data will also be the first estimate of mutation rate in this species)

fitness of the lines after the selection

Fitness estimates that can be done to find out the effect of the selection: Do the selection regime affect : the growth rate? the stress tolerance? competition? are there pleiotropic effects on other life history traits such as pathogeny?

- Use the time 0, (half experiment ~150G?), end of experiment (300G)

- traits to measure are mainly:
    1. *in vitro* growth performance (liquid and solid medium): count cells and colonies.
    2. competition between the genotypes in liquid medium (genotype-specific alleles are known)
    3. Pathogeny of the strains (surface of damaged area on wheat infected leaves, pycnidia number and number of spores within the pycnidia)



colonies grown on PDA solid medium

A: spores coming out of infected leaves;
B: damaged leaves after infection: chlorosis