Resistance and tolerance against infections in hybrid mice

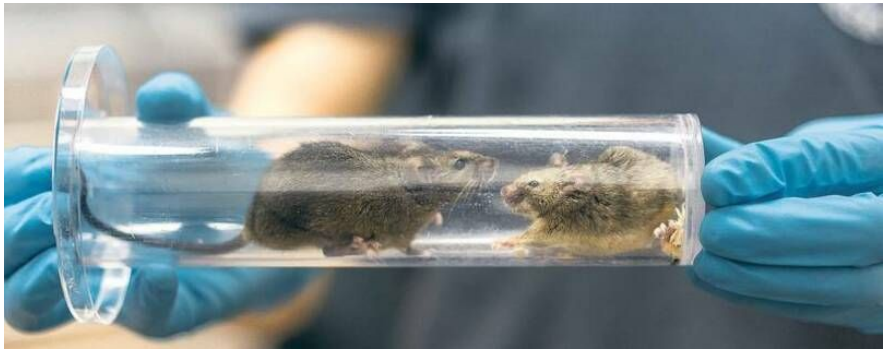
Using immune parameters to predict the impact of hybridization on health

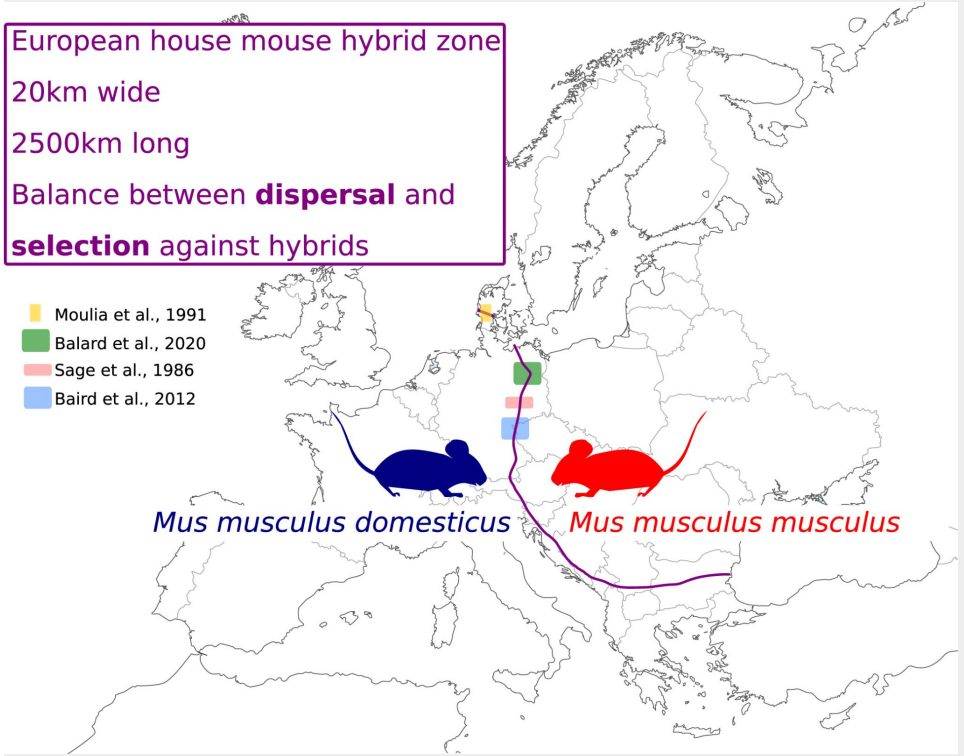
Fay Webster

Emanuel Heitlinger's Lab

1. Introduction:

The european house mouse hybrid zone

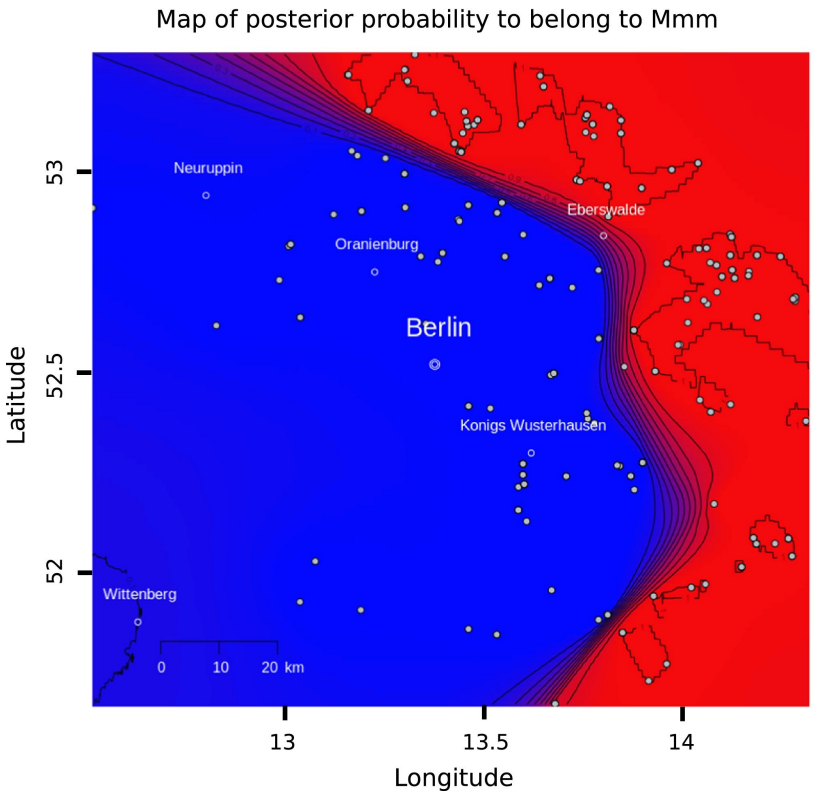




Balard et al., 2019

Study area:

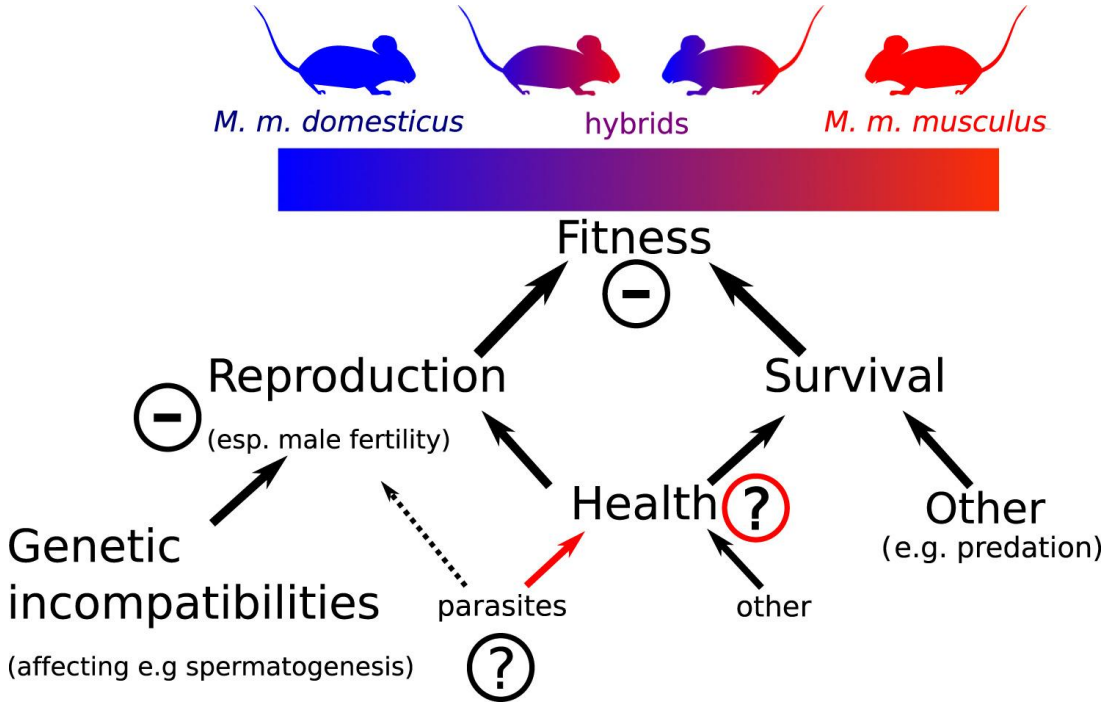
M. musculus domesticus



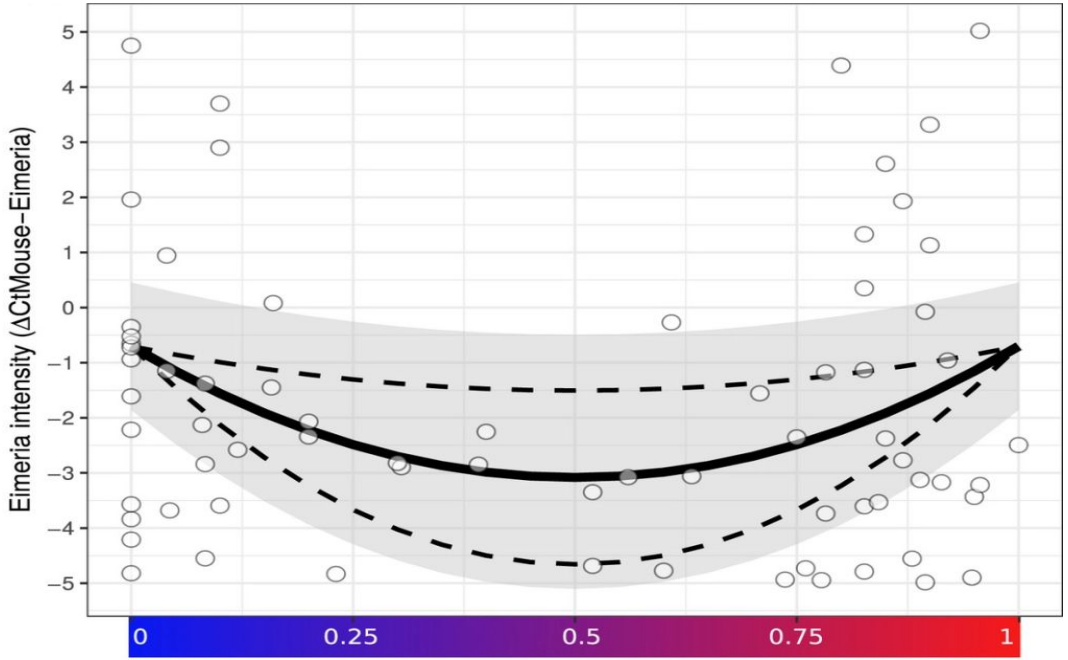
M. musculus musculus

Balard et al., 2019

Fitness components of hybrid mice



Intensity of infections with *Eimeria* are reduced in hybrids

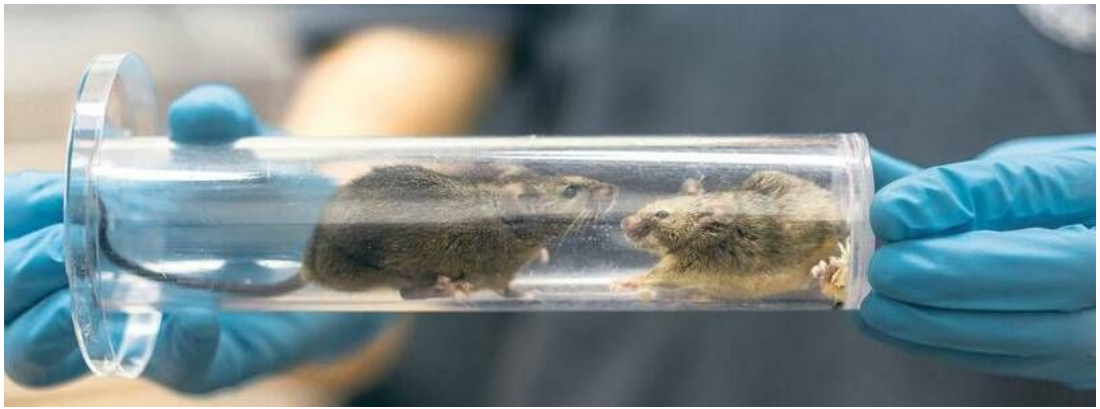


Graph: Balard et al., 2019

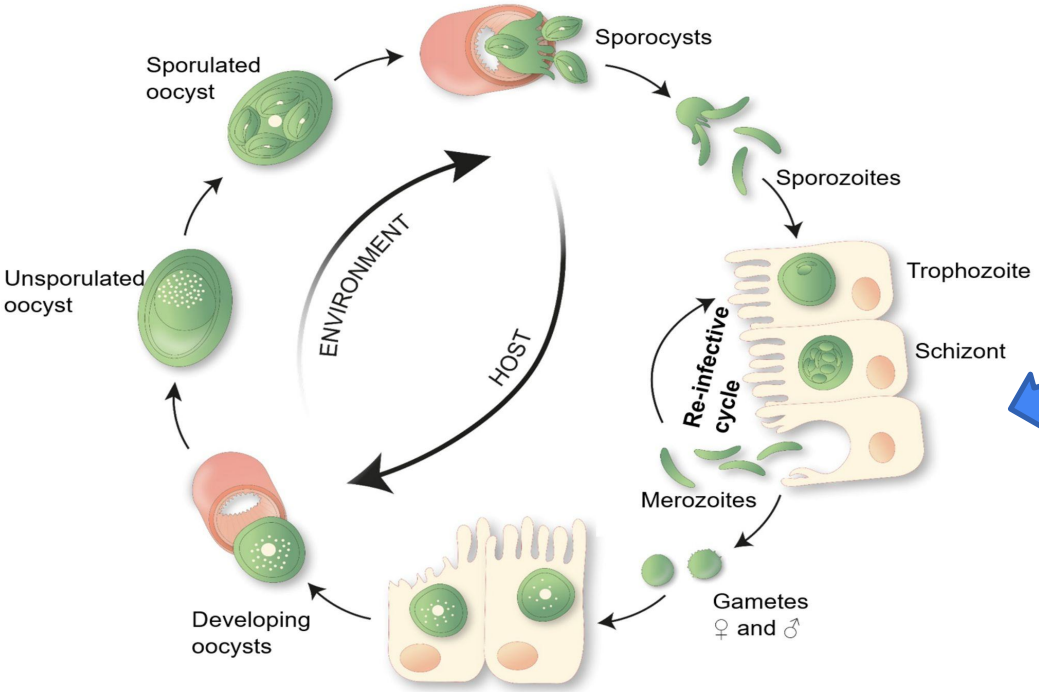
Method: Baird et al., 2012

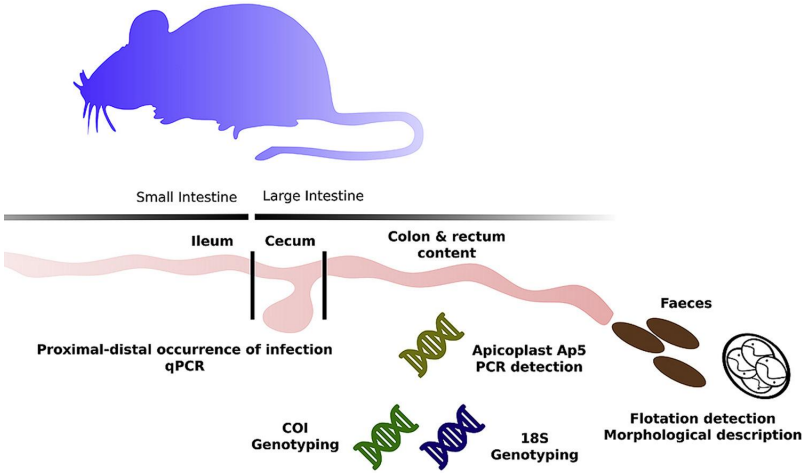
Research question:

Is tolerance modulating the fitness of hybrids?



Life cycle of Eimeria

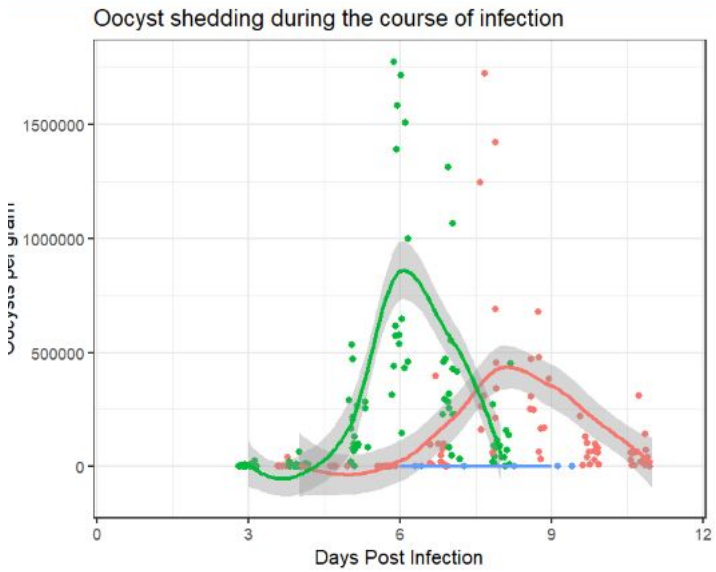


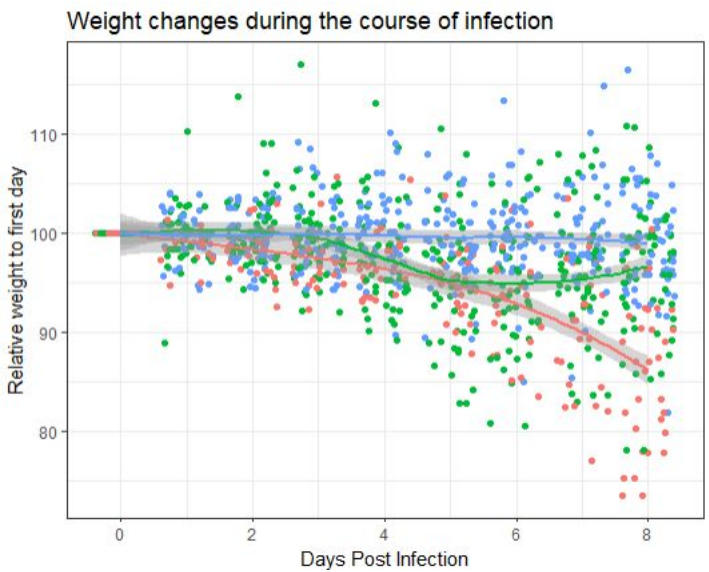


Figueiredo-Campos, P et al., 2019

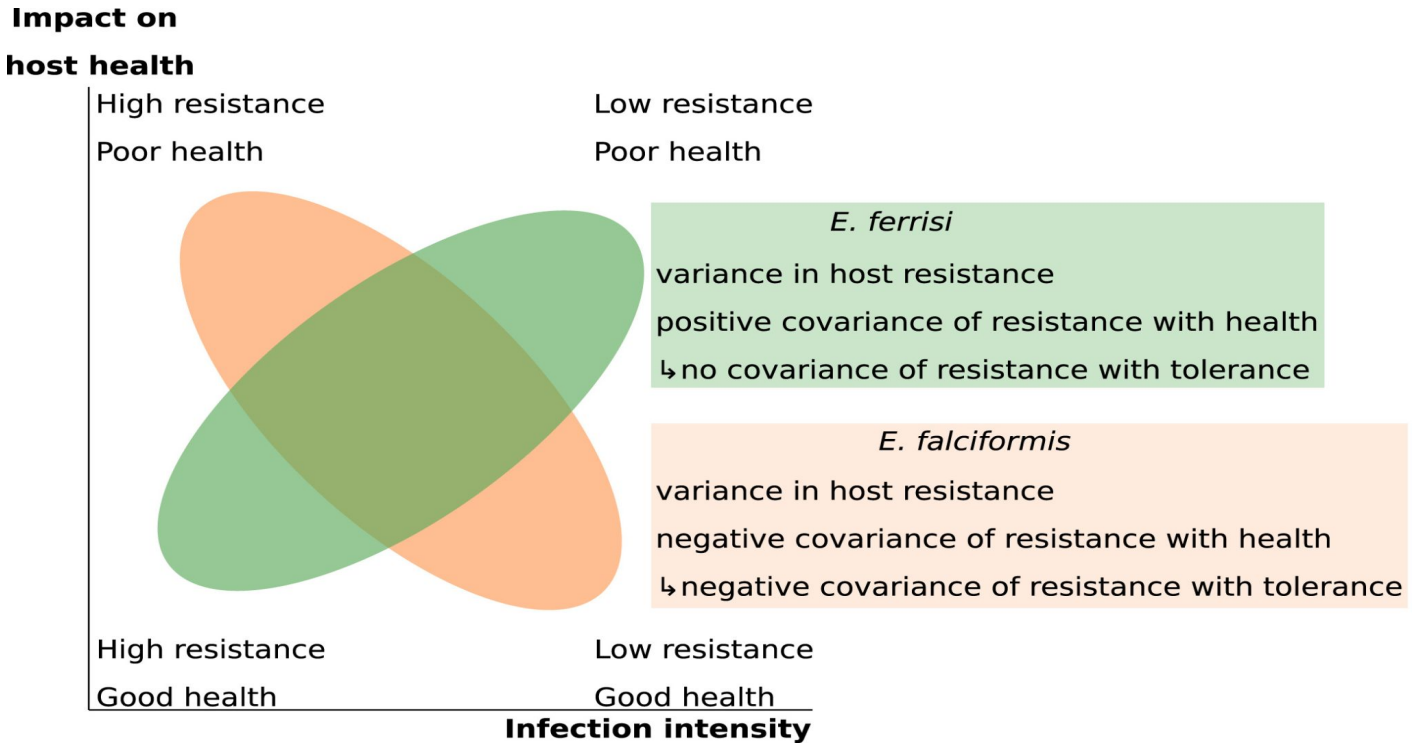
Jarquín-Díaz et al., 2019

Life history differences between two *Eimeria* species





Resistance and Tolerance of E. ferrisi / falciformis



E. ferrisi

variance in host resistance

positive covariance of resistance with health

↳no covariance of resistance with tolerance

E. falciformis

variance in host resistance

negative covariance of resistance with health

↳negative covariance of resistance with tolerance

Objective:

Predicting health impact: An approach using commonly used immune parameters

2. Methods

Experimental design

- Infection groups:
  - *E. falciformis: 22*
  - *E. ferris:* 47
  - Uninfected: 47
  - Total: 116

2. Methods: Experimental design

Infection start:

Infection with 150 sporulated oocysts

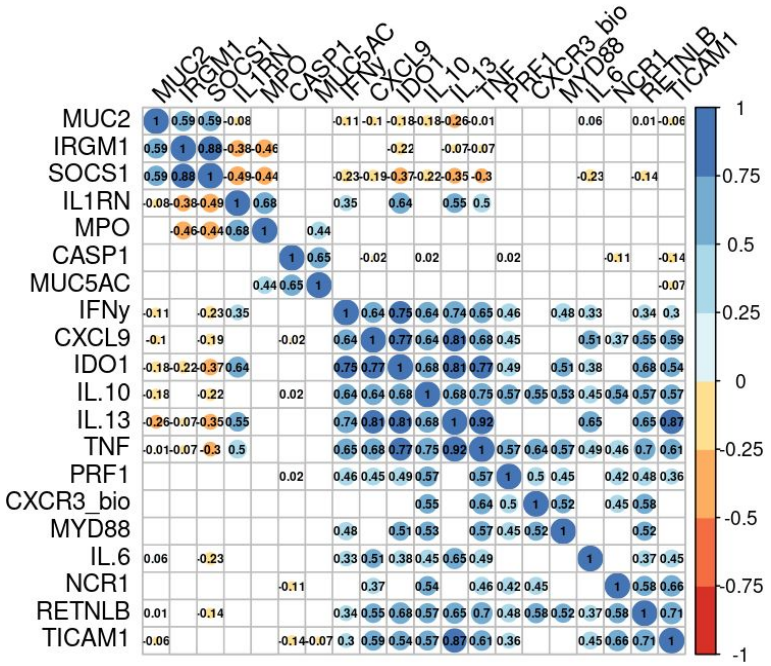
Daily measurements:

1. Weight changes
2. Flotation (OPG)

Dissection

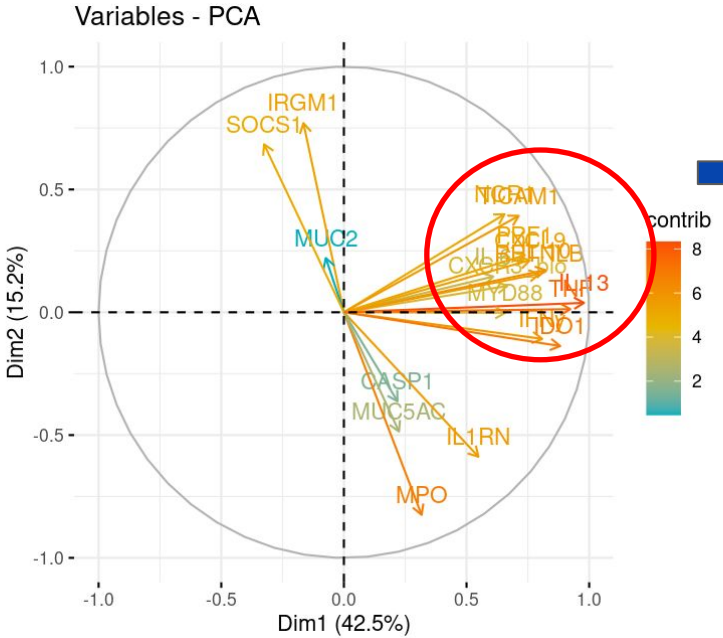
1. Cell sorting (FACS)
2. Immune gene expression Rt qPCR

2. Methods: Immune gene expression



Can the immune measures predict the impact of the infection on host health?

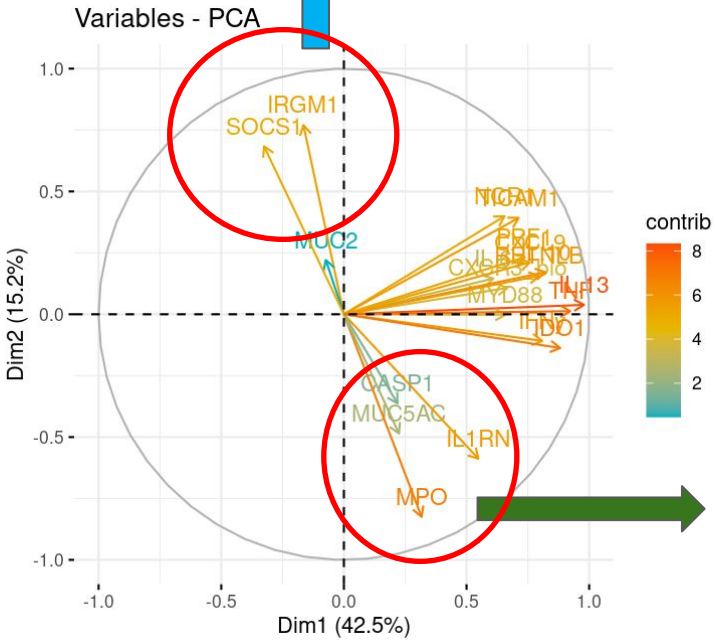
2. Methods: PCA of immune gene expression



pro-inflammatory response

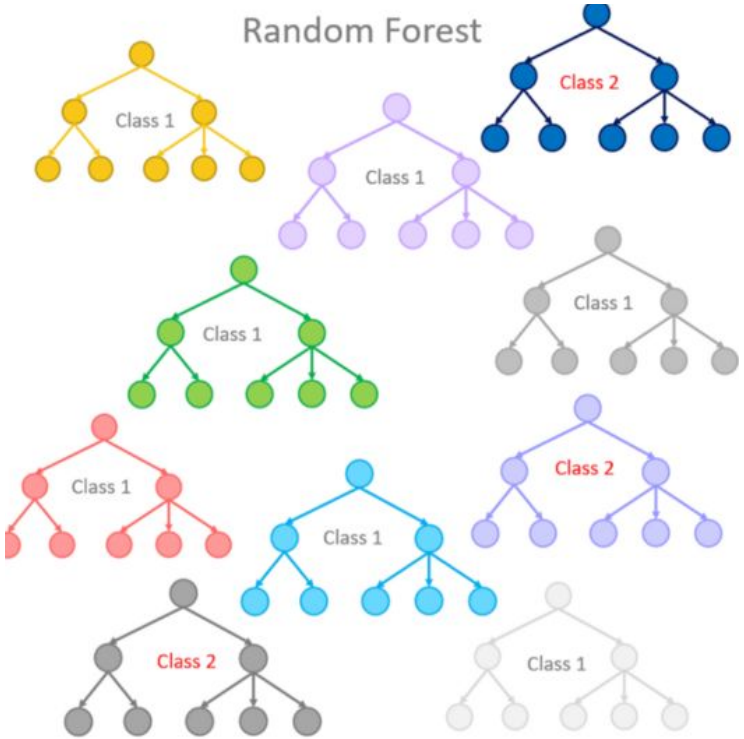
2. Methods: PCA of immune gene expression

mucosal immune response +
inflammatory inhibition



- inflammation
- parasite clearance
- mucus production

2. Methods: Random forest



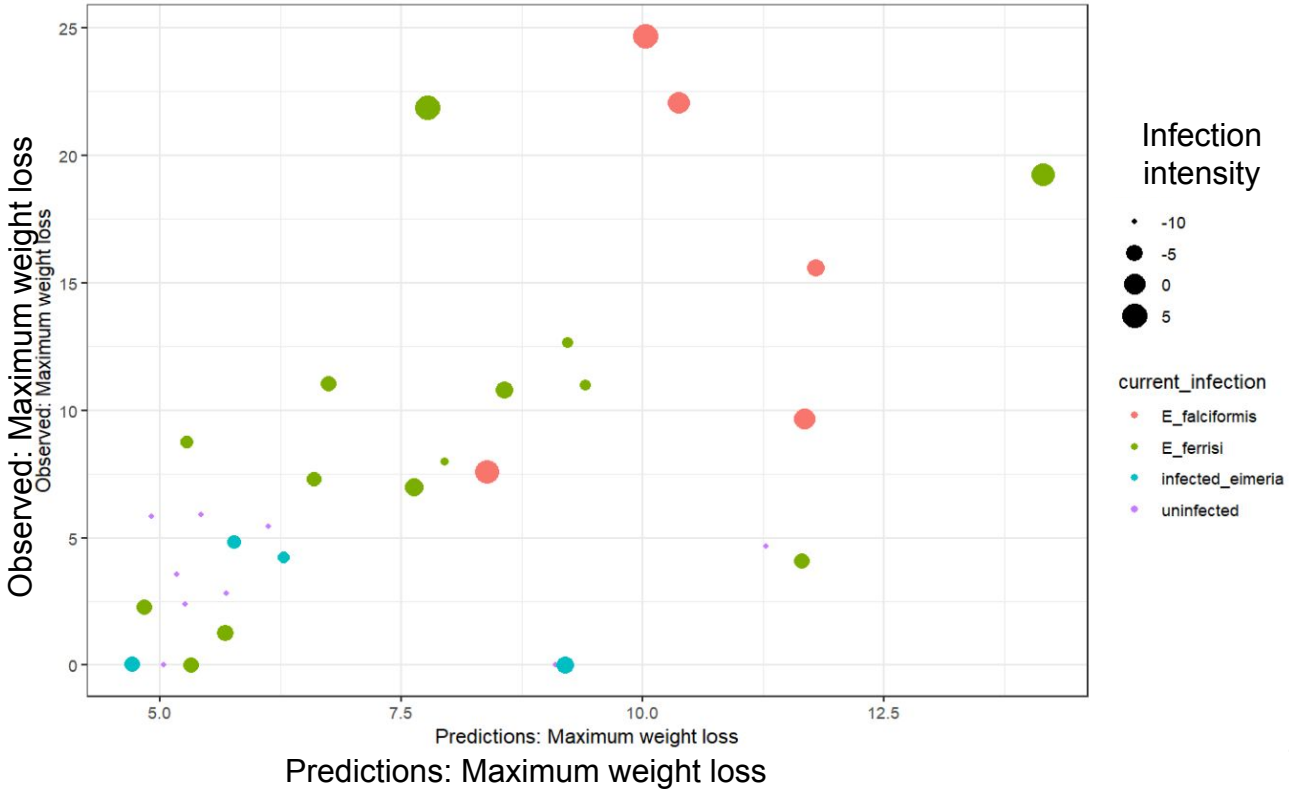
https://towardsdatascience.com/

Random forest: Predicting health impact - lab data

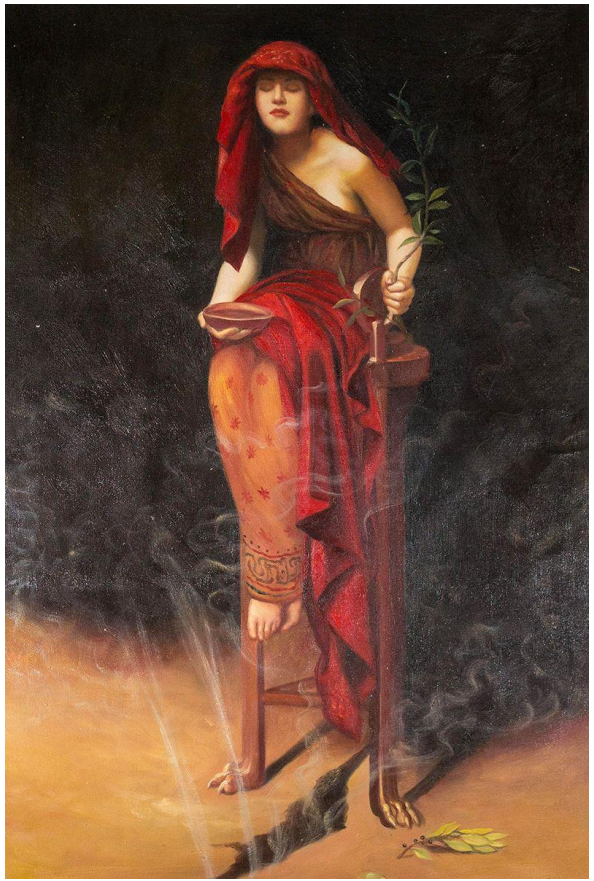
Spearman's rank correlation

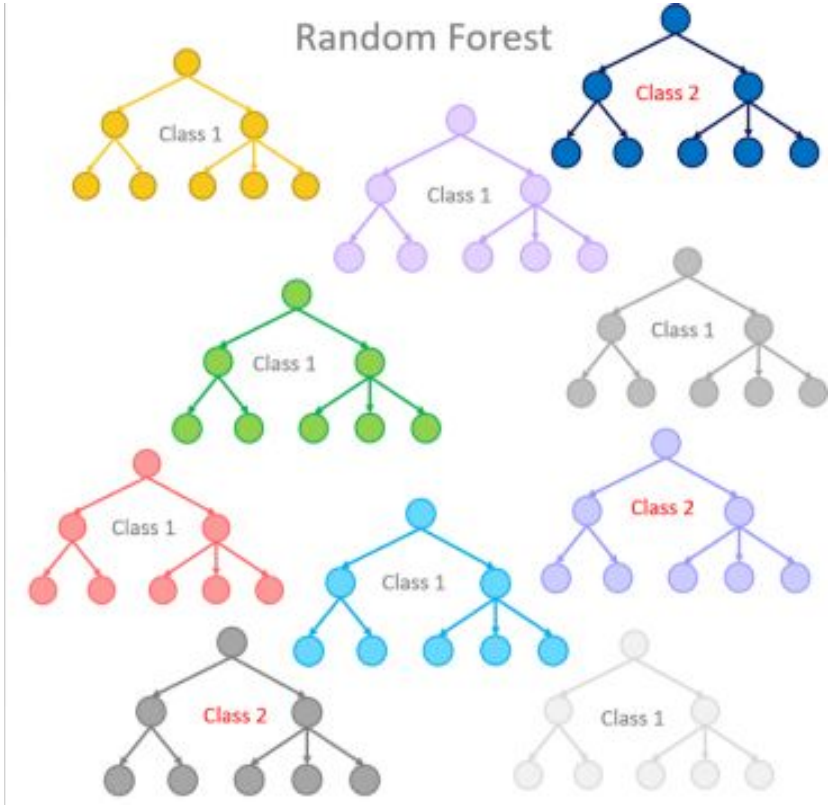
S = 2346.1
p-value = 0.0006603

Rho: 0.5699874

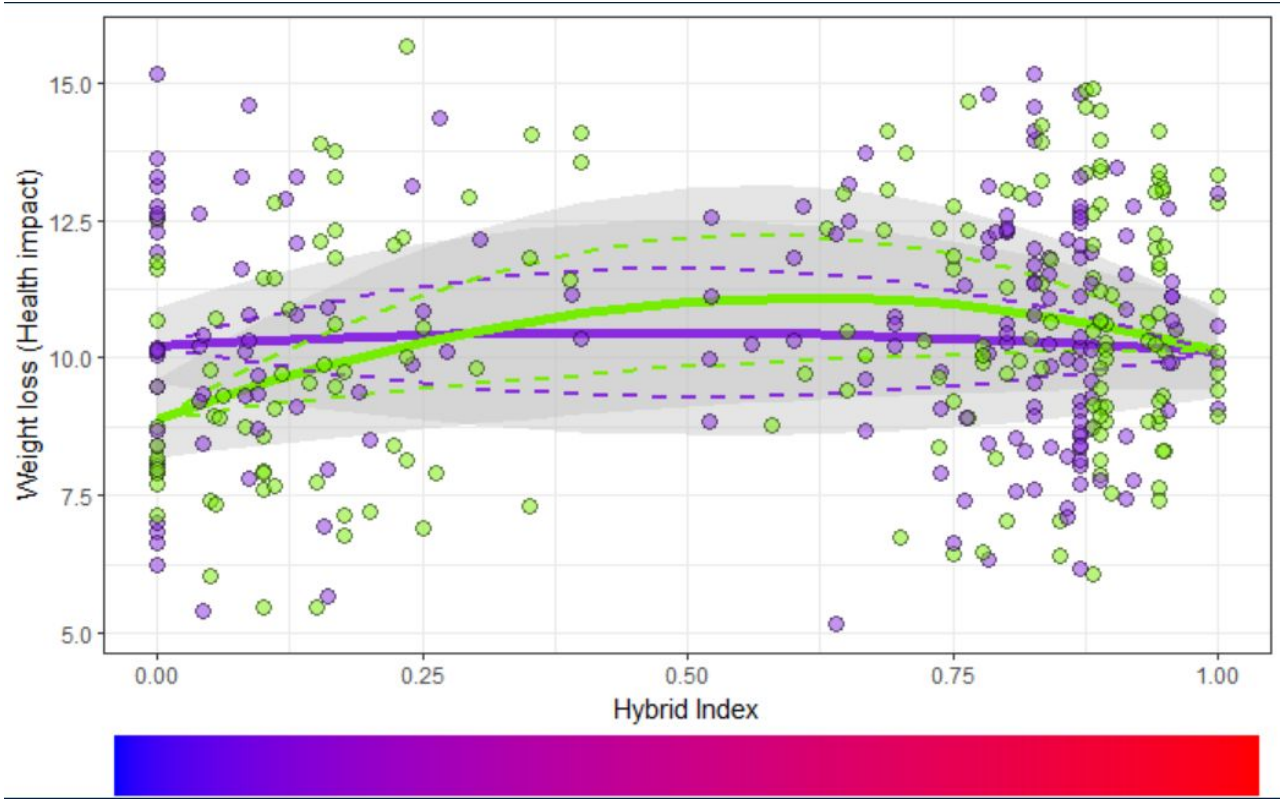


Random forest: Predicting health impact in the field





3. Results: Effect of hybridization on predicted health impact



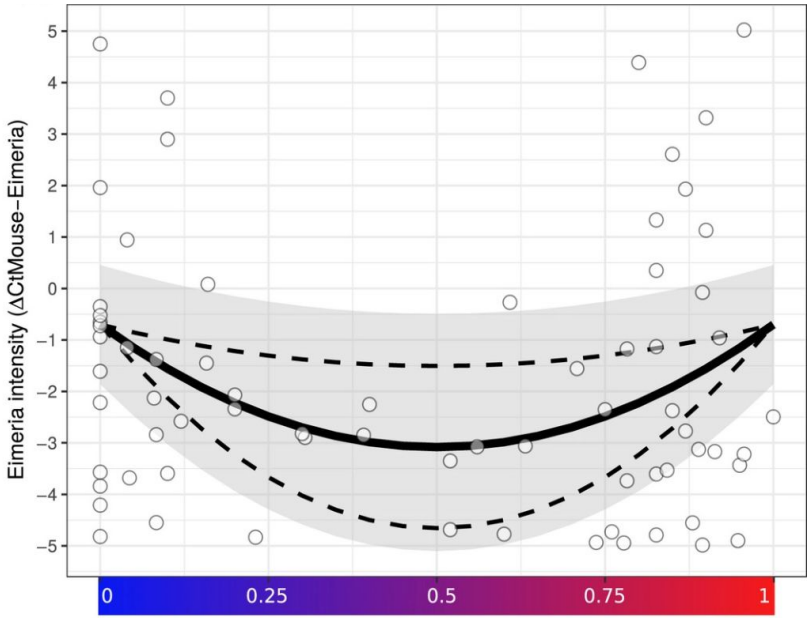
Total mice: 358
Groups: **female** / **male**

Hybridization effect on males:
P value:
0.007448931

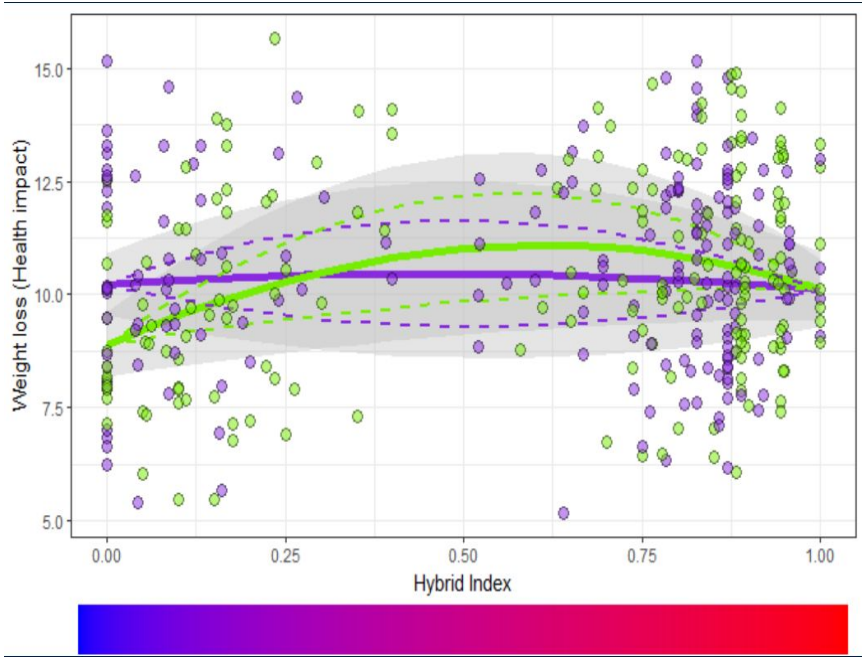
Male effect;
P value:
0.02043259

3. Results: Effect of hybridization on predicted health impact

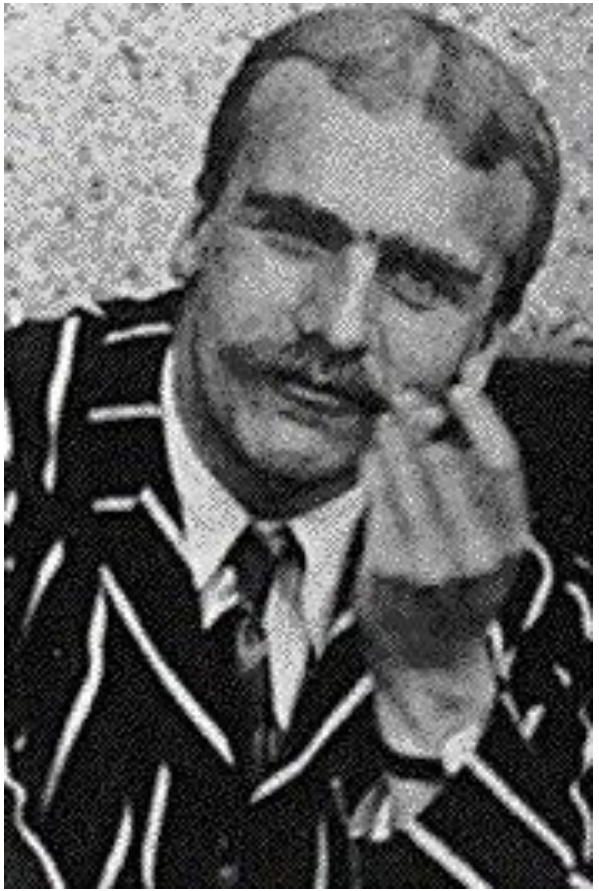
Resistance



Health impact

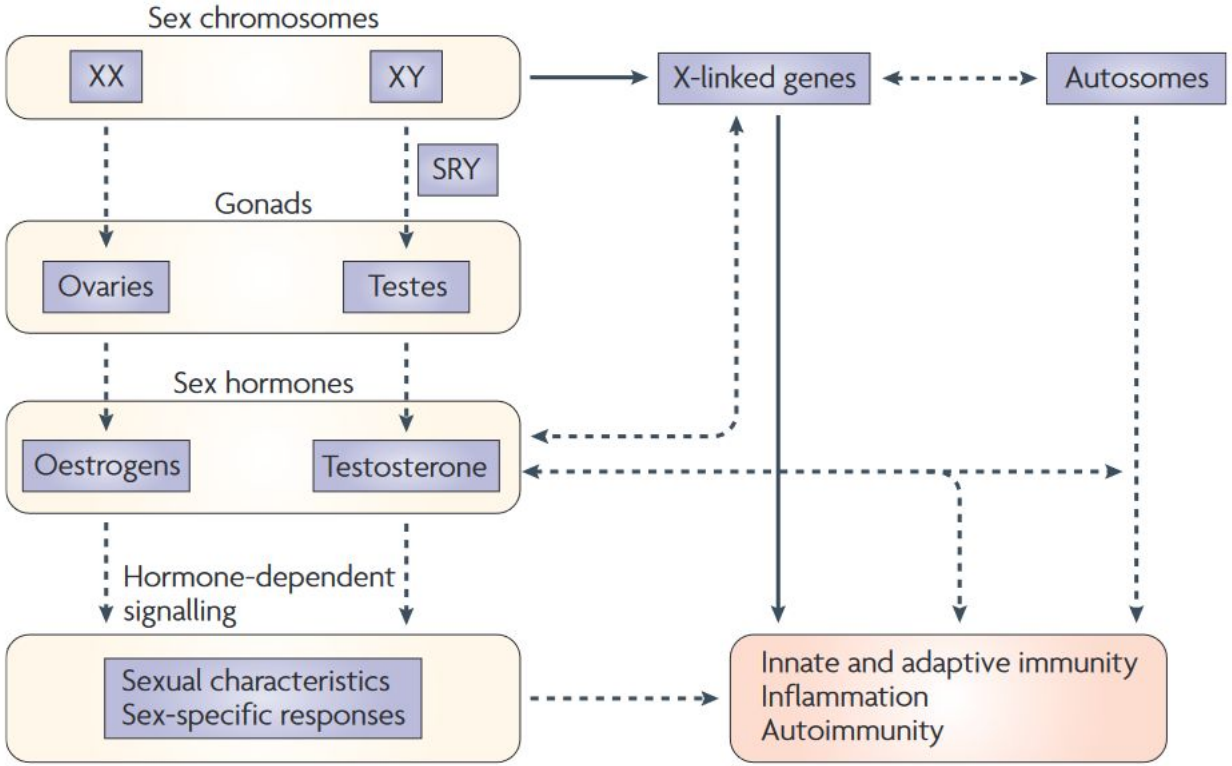


’When in the offspring of two different animal races one sex is absent, rare, or sterile, that sex is the heterozygous sex'



J. B. S. Haldane

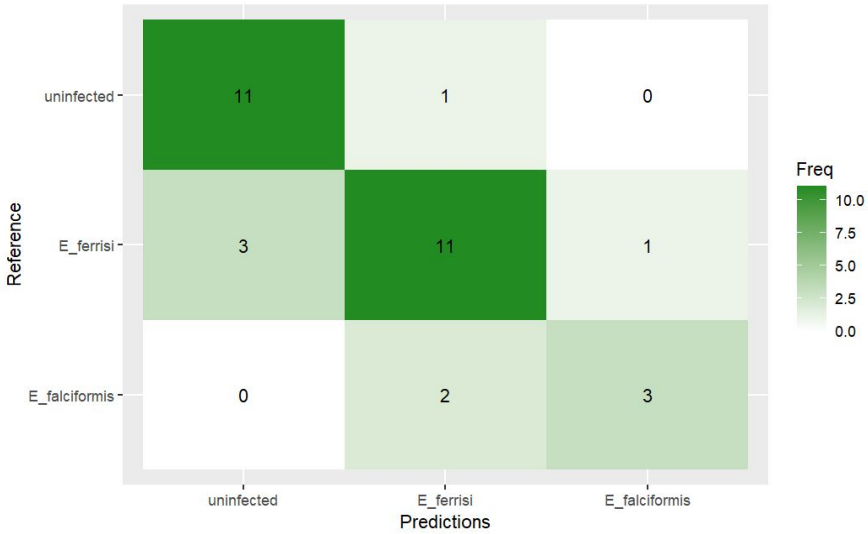
The X chromosome and immunity



Libert et al., 2010

Random forest: Using immune parameters to predict current infection

Lab data: predicting infective parasite



Field data: application of random forest

Acknowledgments

Berlin team:

- Emanuel Heitlinger
- Lubomir Bednar
- Finn Lobnow
- Victor Hugo Jarquín Díaz
- Alice Balard
- Sebastian Rausch
- Hongwei Zhang

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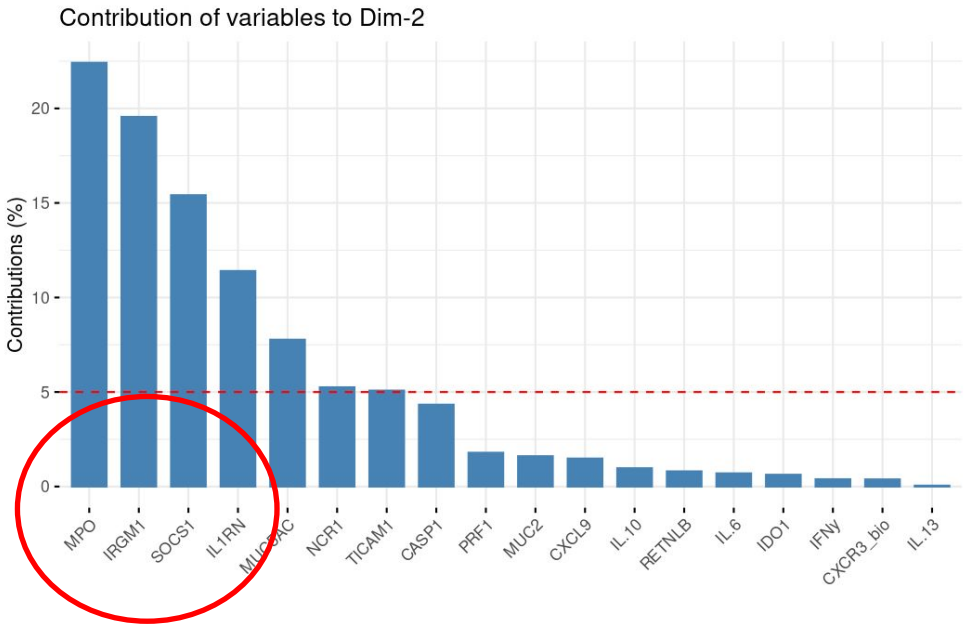
- Jaroslav Piálek
- Miloš Macholán
- Joëlle Goüy De Bellocq
- Ľudovít Ďureje
- Iva Martincová
- Kristina Daniszová

Which principle component better predicts weight loss?

```
## Call:
## lm(formula = max_WL ~ pc1 + pc2, data = imputed_expr)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -16.913  -3.236   1.379   5.127  10.471
##
## Coefficients:
##             Estimate Std. Error t value Pr(>|t|)
## (Intercept)  92.3746     0.6006 153.811   <2e-16 ***
## pc1           0.1702     0.2061   0.826   0.4107
## pc2          -0.7501     0.3448  -2.175   0.0317 *
## ---
```

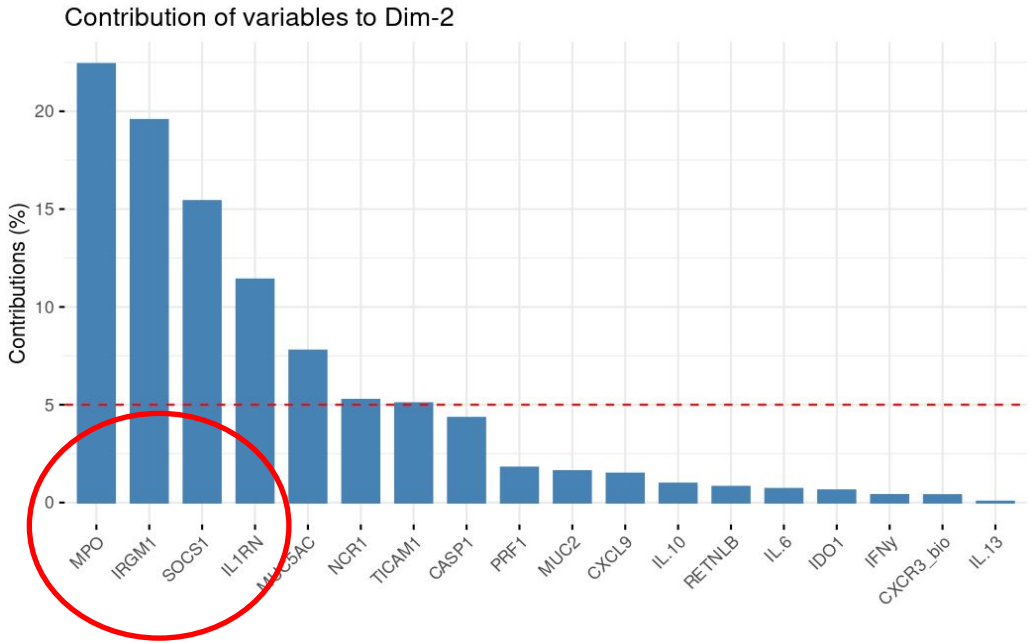


Which principle component better predicts weight loss?

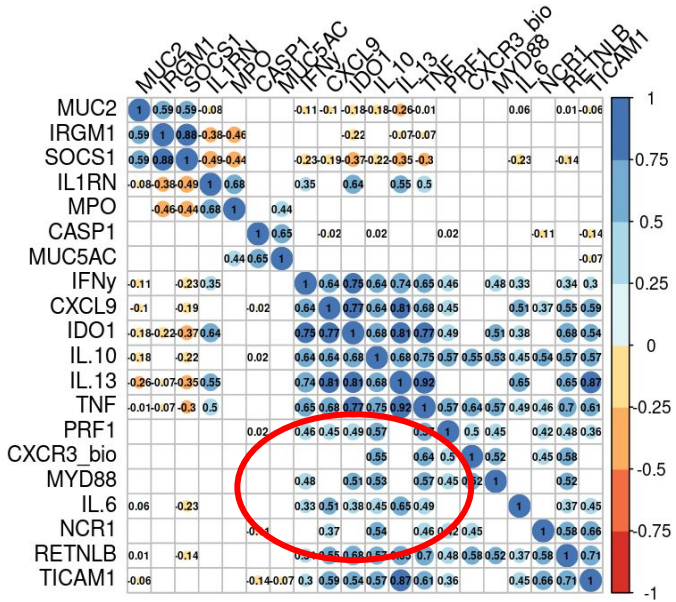
Parasite clearance

mucosal response

inflammatory inhibition



Inflammation, anti-fungal and microbial defense



Field data

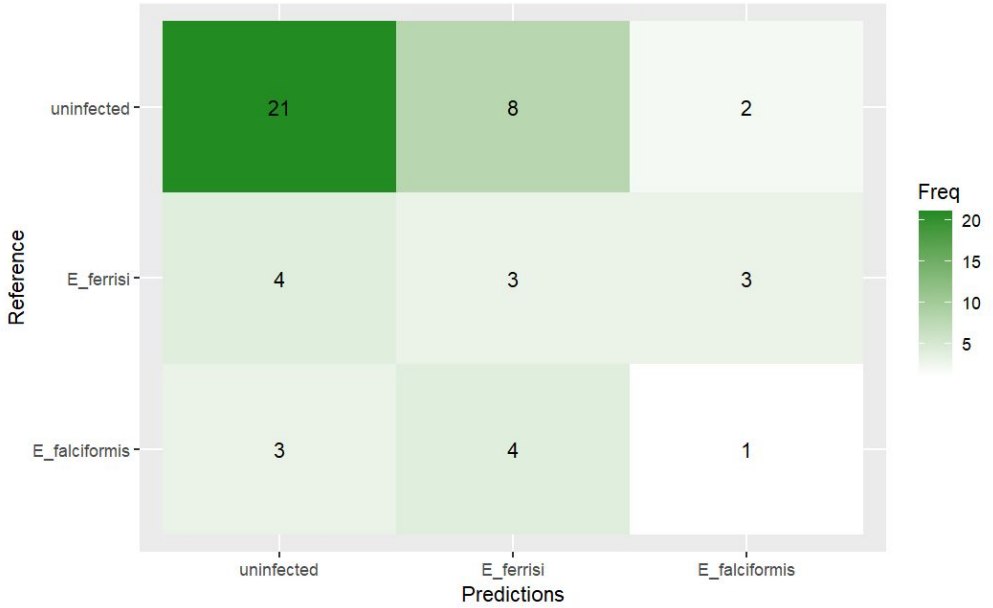
Accuracy : 0.5102
95% CI : (0.3634, 0.6558)
No Information Rate : 0.6327
P-Value [Acc > NIR] : 0.9713

Kappa : 0.1191

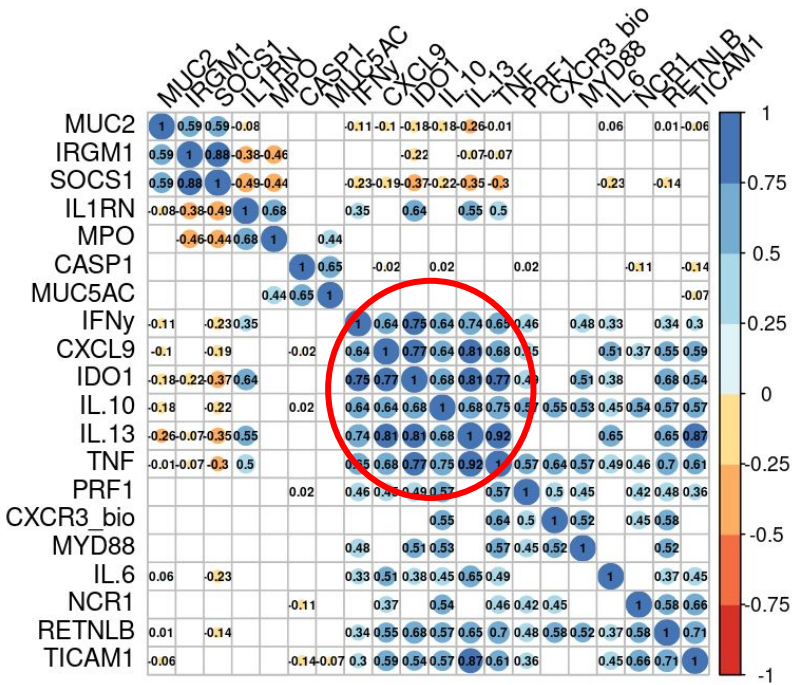
Mcnemar's Test P-Value : 0.6422

Statistics by Class:

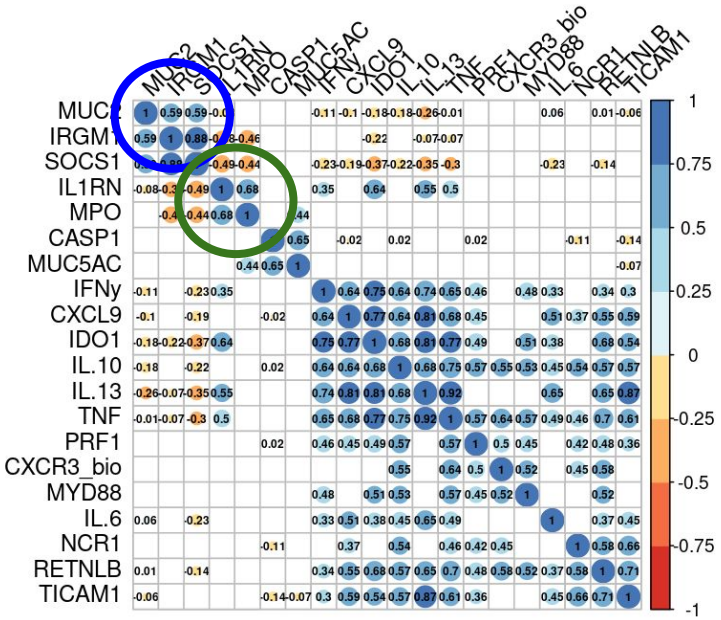
	Class: E_falciformis	Class: E_ferrisi	Class: uninfected
Balanced Accuracy	0.50152	0.49615	0.6443

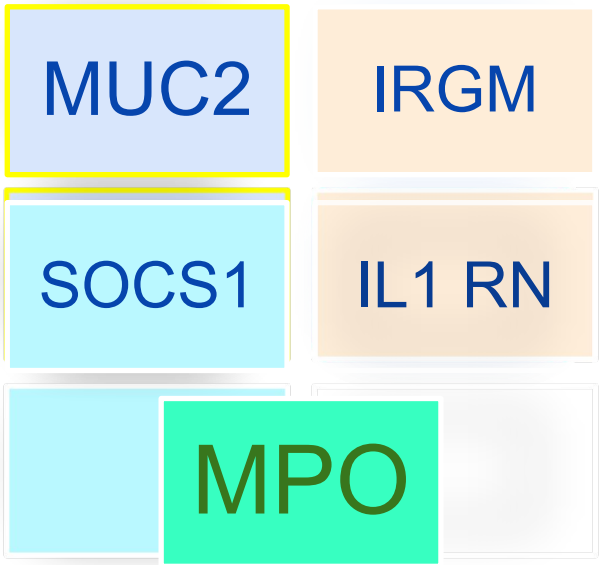


Pro-inflammatory response → parasite clearance, mucus production

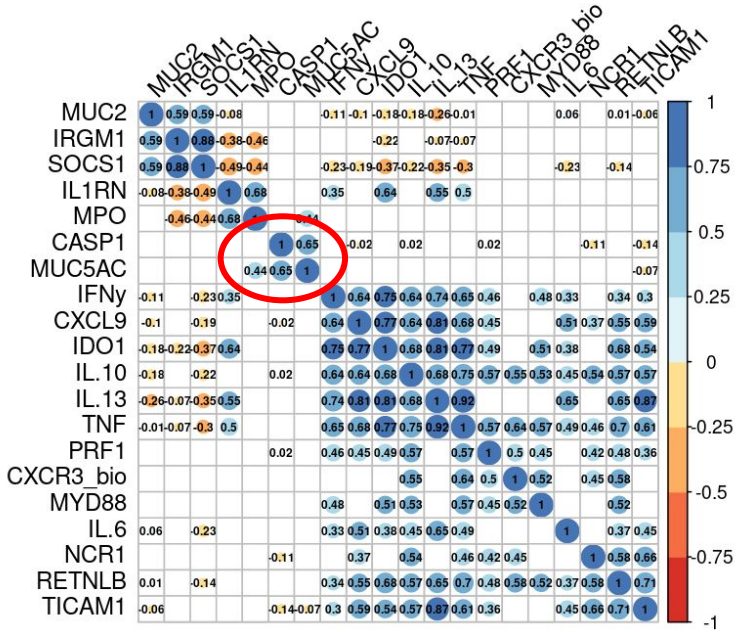


Mucosal immune response + inflammatory inhibition:



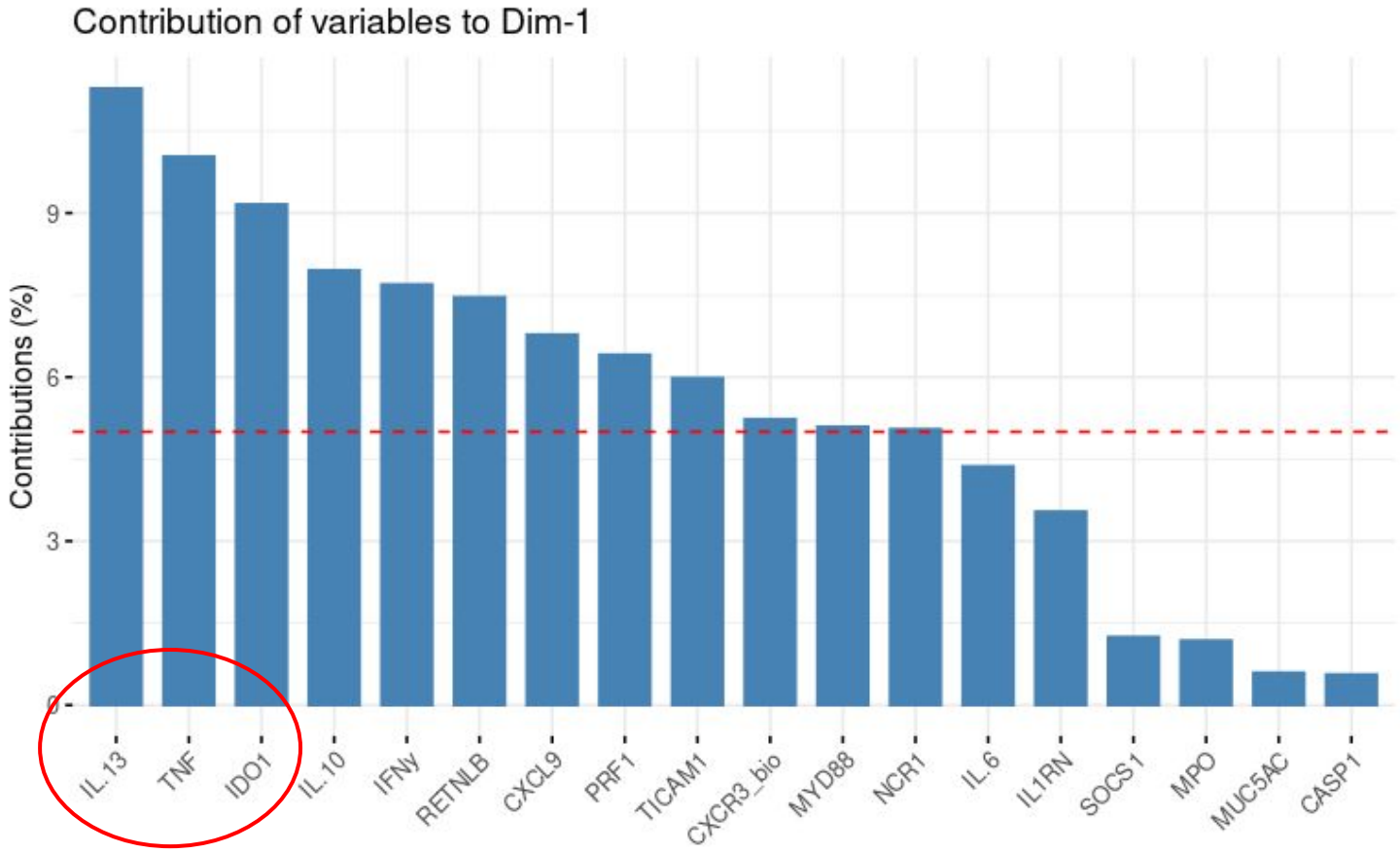


Parasite clearance, Mucus production

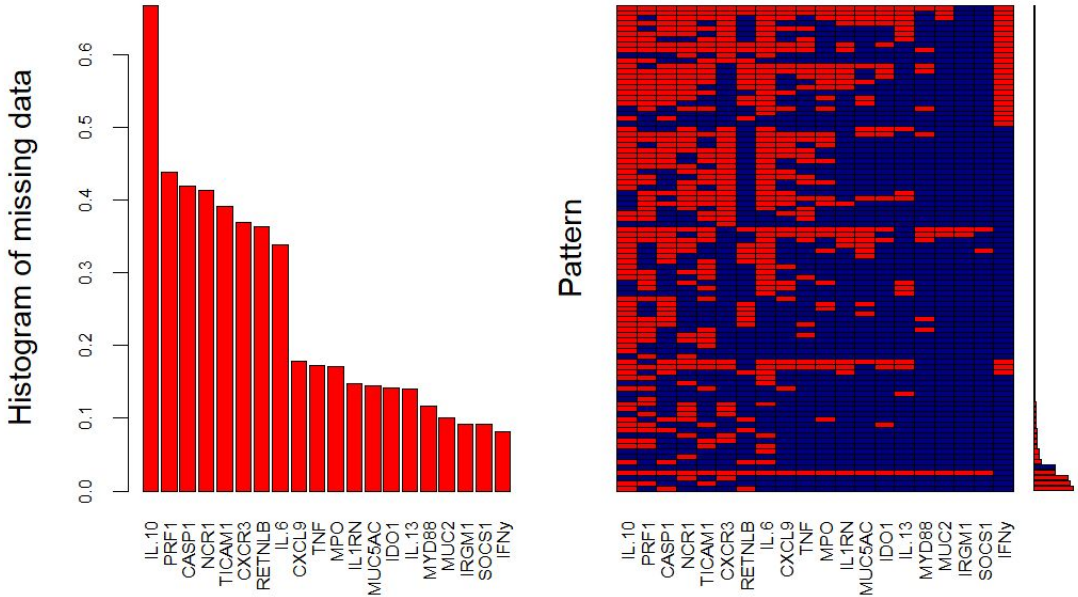


CASP1

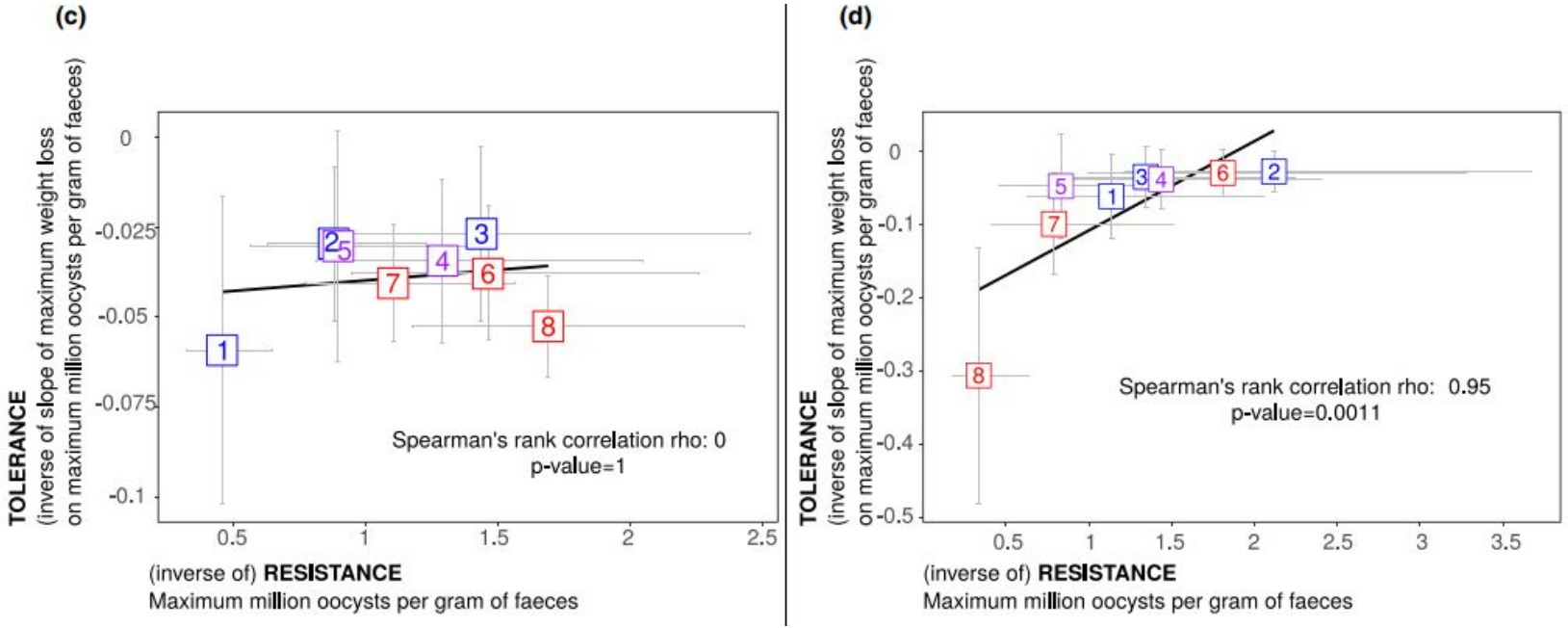
MUC5AC


Contribution of variables to Dim-1
Contributions (%)
9
6
3
0
IL.13
TNF
IDO1
IL.10
IFNγ
RETNLB
CXCL9
PRF1
TICAM1
CXCR3_bio
MYD88
NCR1
IL.6
IL1RN
SOCS1
MPO
MUC5AC
CASP1

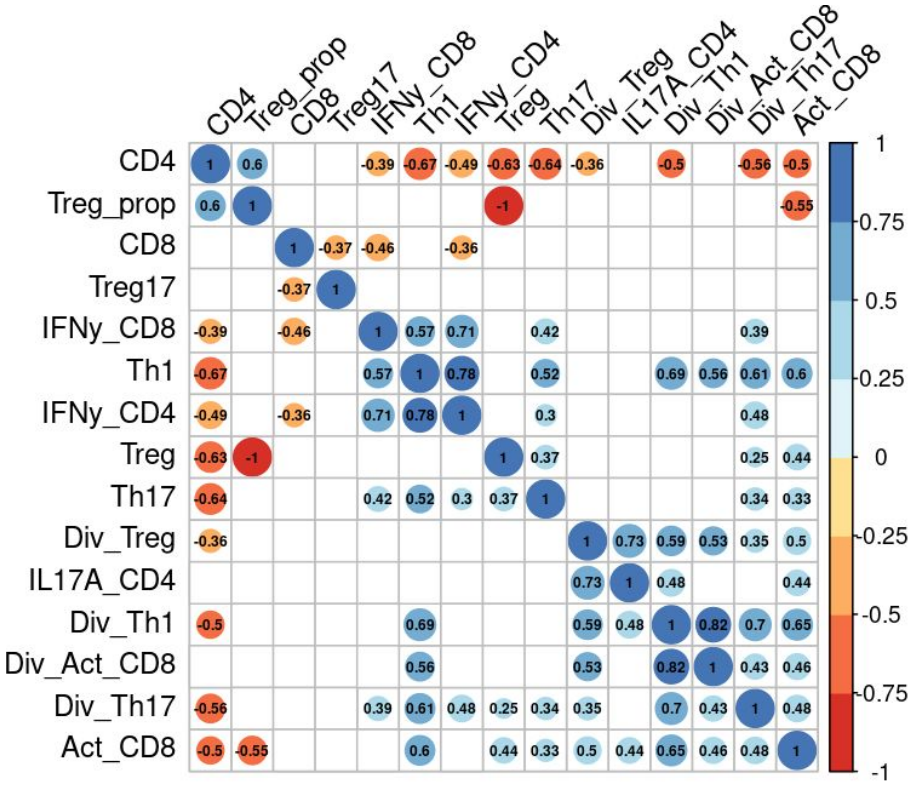
Imputation mice

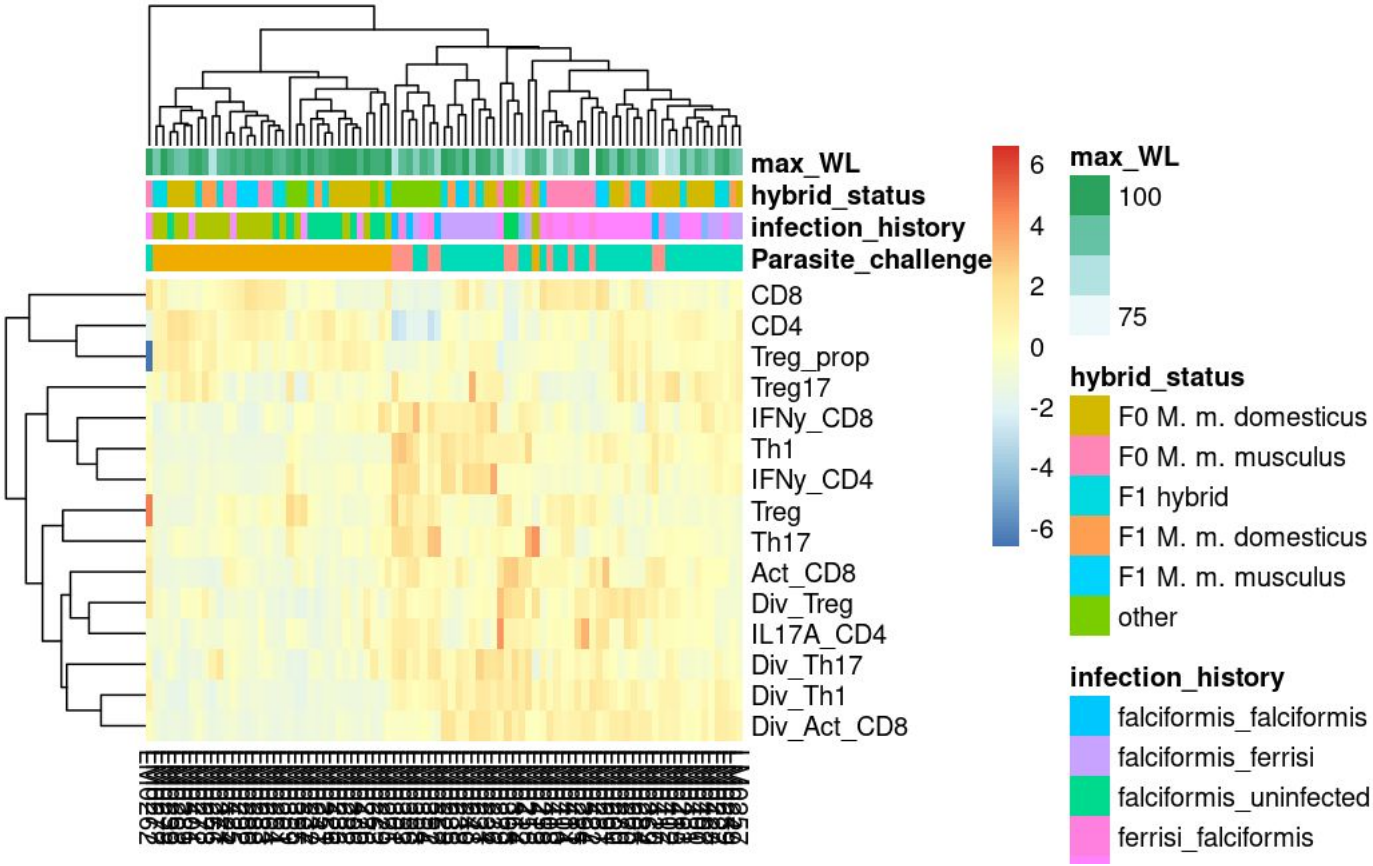


E. ferrisi and E. falciformis: How does resistance and tolerance correlate in each species?

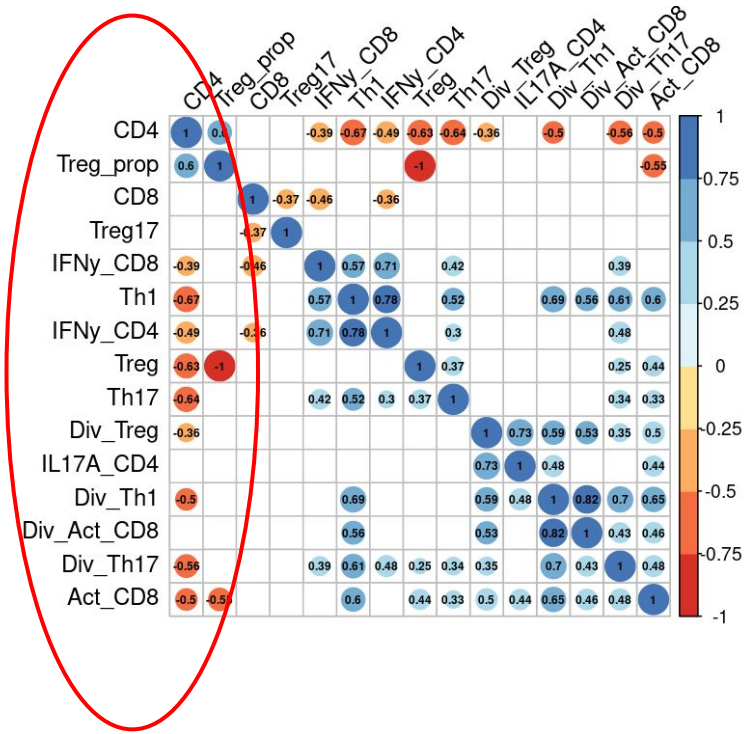


FACS


CD4
Treg_prop
CD8
Treg17
IFNy_CD8
Th1
IFNy_CD4
Treg
Th17
Div_Treg
IL17A_CD4
Div_Th1
Div_Act_CD8
Div_Th17
Act_CD8
1
0.75
0.5
0.25
0
-0.25
-0.5
-0.75
-1

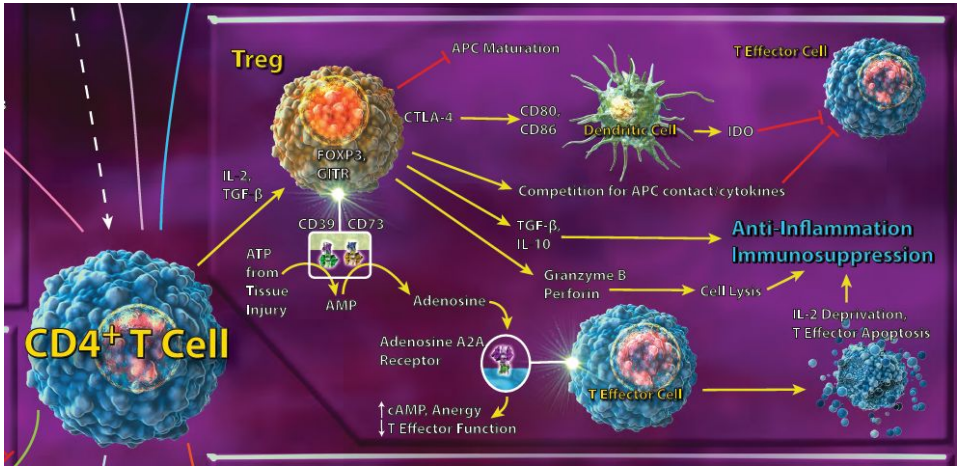

max_WL
hybrid_status
infection_history
Parasite_challenge
CD8
CD4
Treg_prop
Treg17
IFNy_CD8
Th1
IFNy_CD4
Treg
Th17
Act_CD8
Div_Treg
IL17A_CD4
Div_Th17
Div_Th1
Div_Act_CD8
6
4
2
0
-2
-4
-6
max_WL
100
75
hybrid_status
F0 M. m. domesticus
F0 M. m. musculus
F1 hybrid
F1 M. m. domesticus
F1 M. m. musculus
other
infection_history
falciformis_falciformis
falciformis_ferrisi
falciformis_uninfected
ferrisi_falciformis

CD4

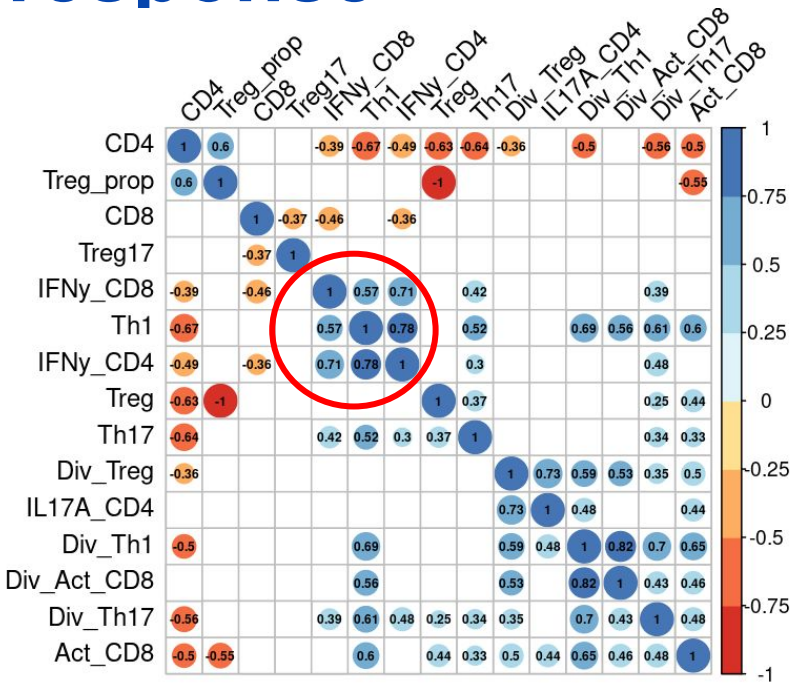


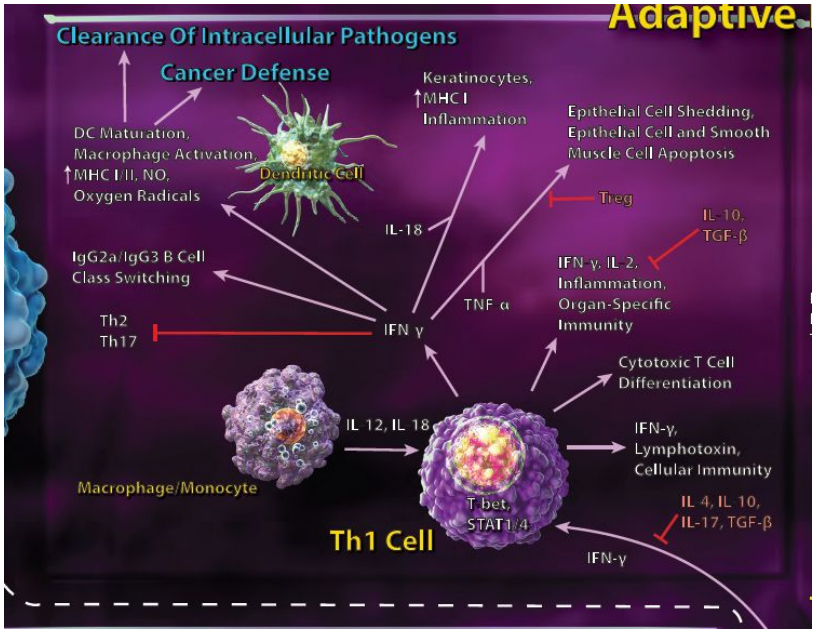
CD4+T:

- activation of the cells of the innate immune system
- critical role in the suppression of immune reaction



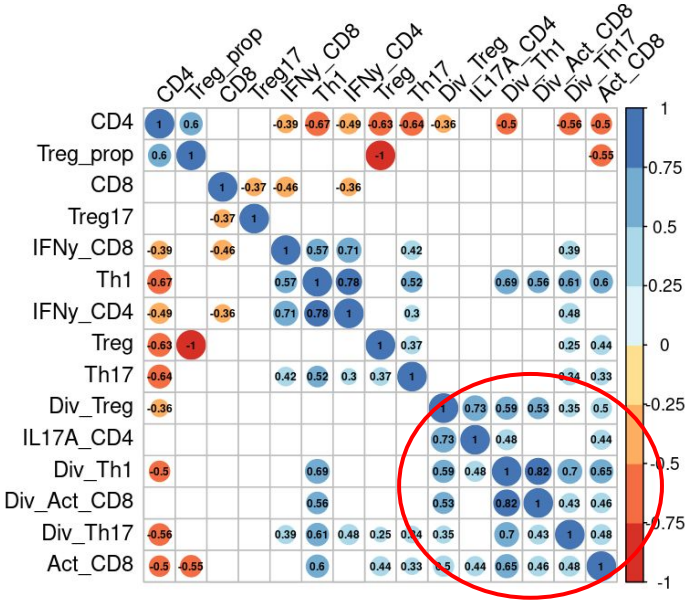
Pro-inflammatory response



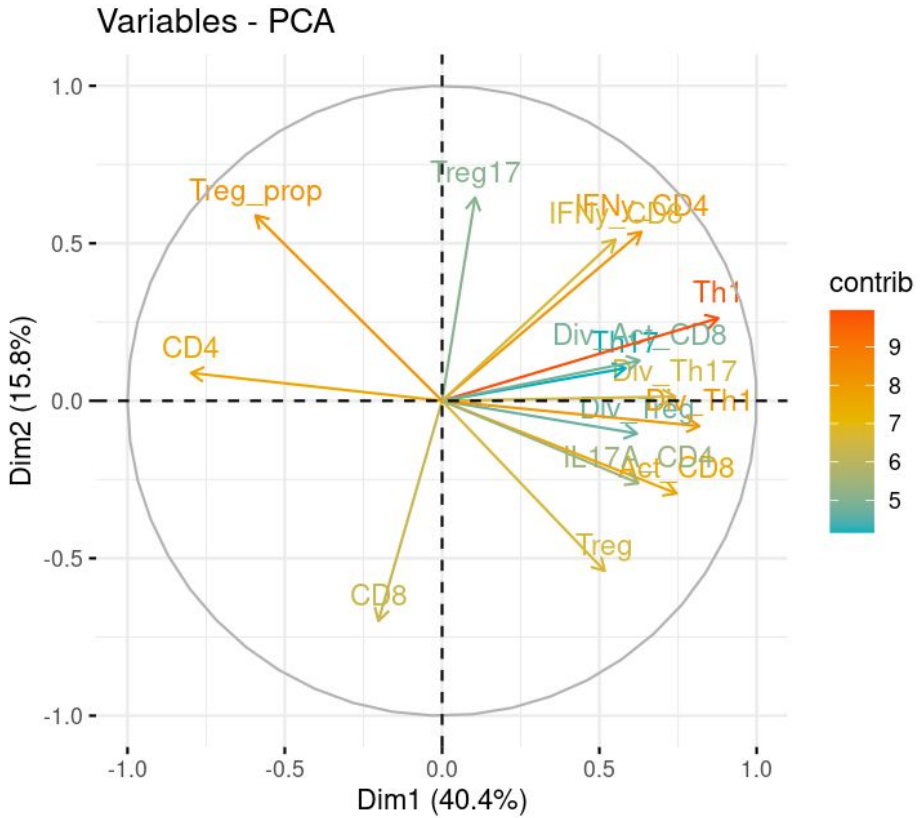


- **IFNy_CD8: pro - inflamatory IFNy**
- **IFNy_CD4: pro - inflamatory IFNy**
- **Th1: increased cell-mediated response**

Dividing cells



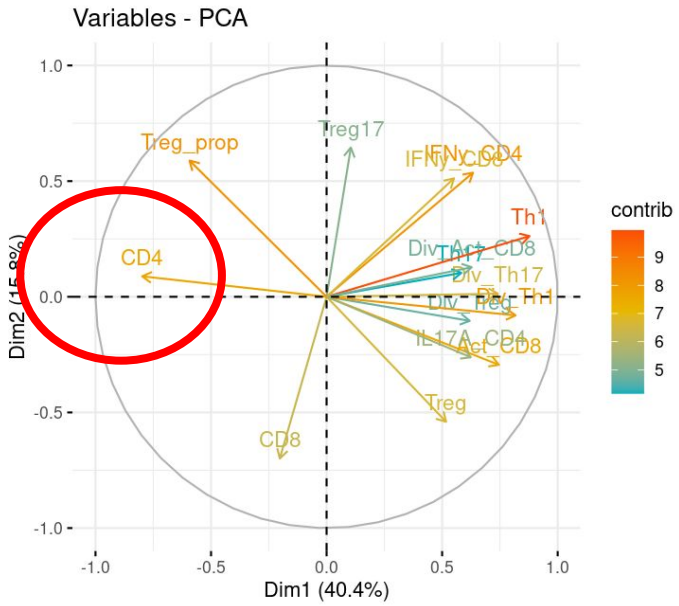
PCA of FACS data

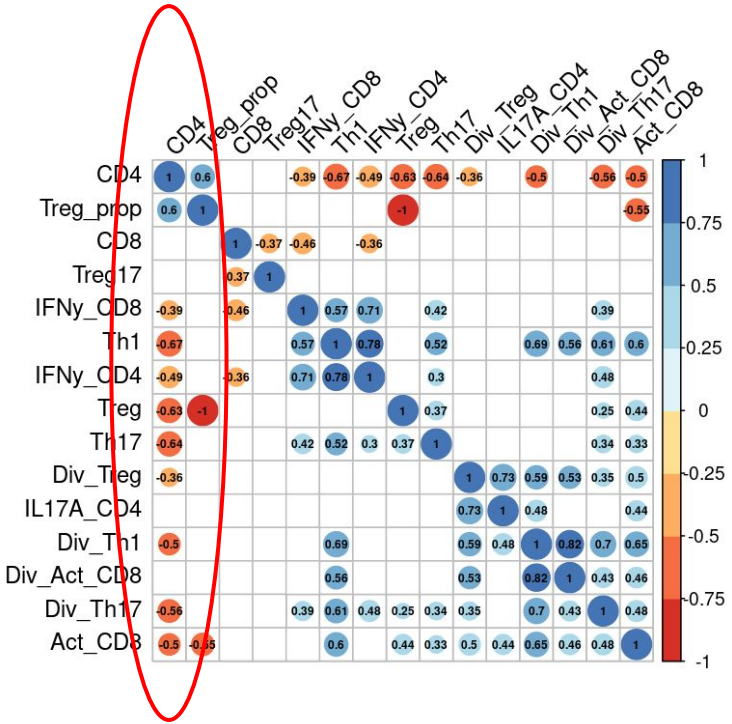


CD4

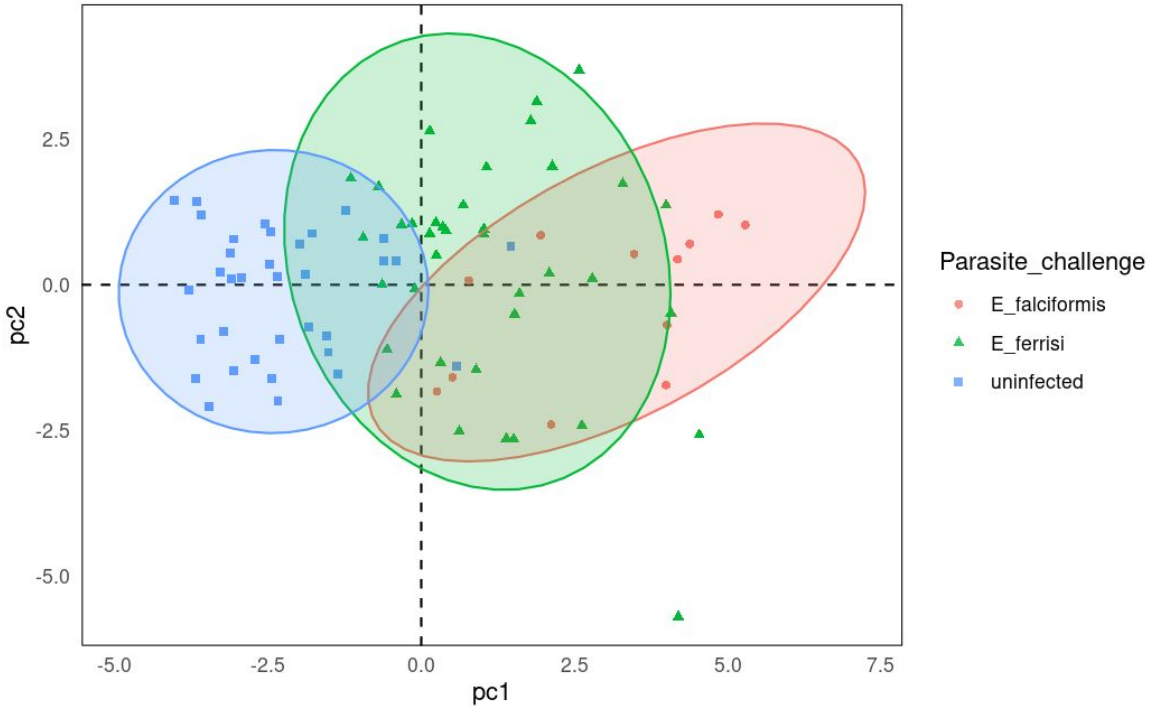
CD4+T:

- activation of the cells of the innate immune system
- critical role in the suppression of immune reaction

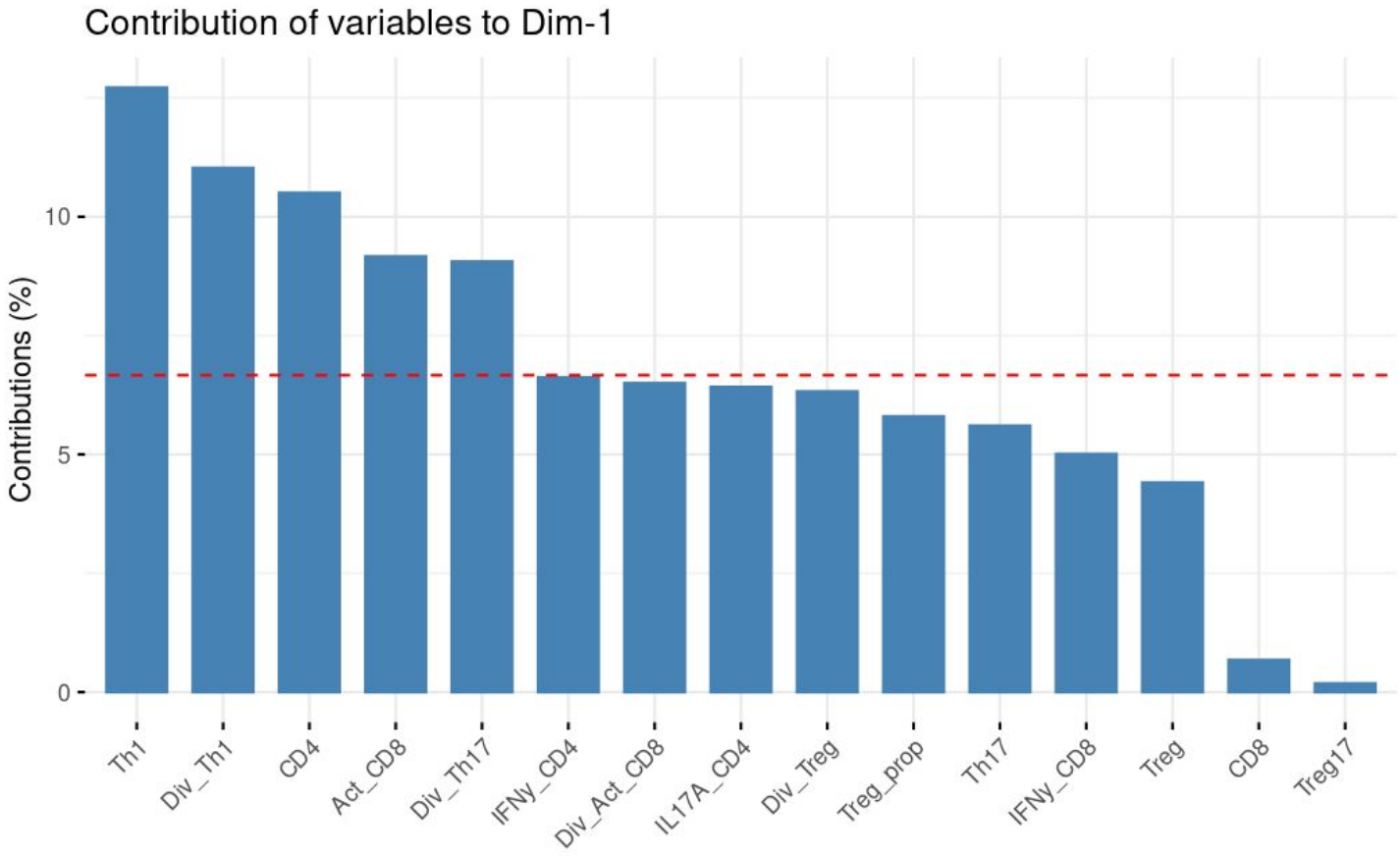




FACS


2.5
0.0
-2.5
-5.0
pc2
-5.0
-2.5
0.0
2.5
5.0
7.5
pc1
Parasite_challenge
E_falciformis
E_ferrisi
uninfected

FACS


Contribution of variables to Dim-1
Contributions (%)
10
5
0
Th1
Div_Th1
CD4
Act_CD8
Div_Th17
IFNγ_CD4
Div_Act_CD8
IL17A_CD4
Div_Treg
Treg_prop
Th17
IFNγ_CD8
Treg
CD8
Treg17

PCA of immune gene expression