| Analysis Phase | Research Question | Model ID | Statistical Method |
|---|---|---|---|
| **Laboratory Development** | | | |
| Discovery | Can immune genes predict infection costs? | **DISC-1** | Linear regression (PC1, PC2 → weig loss) |
| Optimization | Can machine learning improve prediction? | **DISC-2** | Random forest (19 genes → weight loss) |
| Validation | Is the model reliable? | **DISC-3** | Train-test cross-validation |
| **Cross-Population Translation** | | | |
| Gene Validation | Which genes show consistent responses across populations? | **TRANS-1** | Linear regression per gene (lab vs field) |
| **Field Translation** | | | |
| Detection | Does the model work in wild populations? | **FIELD-1** | Predicted vs. observed infection status |
| Discrimination | Can it distinguish parasite species? | **FIELD-2** | Predicted loss by species identity |
| Scaling | Does it correlate with infection severity? | **FIELD-3** | Predicted loss vs. parasite load |
| **Biological Validation** | | | |
| Physiological relevance | Does it capture real health impacts? | **PROOF-1** | Predicted loss vs. body condition |
| Specificity | Is the response Eimeria-specific? | **PROOF-2** | Predicted loss vs. parasite communi |

Framework demonstrates progression from basic linear prediction ($R^2$ = 0.106) through machine learning optir relevance. Cross-population translation validates 3/19 genes as conserved biomarkers.

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|---|---|---|---|

[1] Significance levels: *p < 0.05, **p < 0.01, ***p < 0.001

[2] Train-test validation: 70% training (n=96), 30% testing (n=40)

[3] Cross-validated genes: CXCL9 (both species), TICAM1 (E. falciformis), PRF1 (E. falciformis)

[4] E.f: Eimeria falciformis; E.r: E. ferrisi

[5] Parasite community model tested: Eimeria (significant), Aspiculuris, Syphacia, Trichuris, Mastophorus (all non-

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