INFERENTIAL STATISTICS AND HYPOTHESIS TESTING

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Introduction

Inferential statistics are used to **draw conclusions from data**.

The aim: To formulate hypotheses and test these in order to be able to make generalisations concerning **populations** from **samples**.

The procedure: Using random sampling practices and hypotheses testing procedures to judge validity of previously established hypotheses.

Inferential statistics often invoke **measures of statistical significance**.

Methods & Quirks

Information is handed to inferential statistics in a multitude of different forms (e.g. vectors, matrices, data frames). This information is used to:

Establish Hypotheses:

- Null/Alternative
- (Non-)Directional
- (Non-)Specified
- Difference
- Equivalence
- Relationship

Test Hypotheses:

- *Non-Parametric Tests*
  - Nominal and Correlation tests
  - Ordinal and Metric tests
  - ...
- *Parametric Tests*
  - t-test
  - ANOVA
  - ...

Inferential statistics **allow generalisation** beyond the data at hand!

Hypotheses And Their Importance

What is a hypothesis?

In the case of inferential statistics, a *hypothesis* presents some *rationale* about patterns within the natural world and hence the data.

What's the fuss?

Hypotheses are *simplifications* of possible norms of natural processes and *make things testable*.

So?

Getting the right answers always comes down to asking the right questions.

Hypotheses are, more or less, **educated guesses**.

Null vs. Alternative Hypotheses (Theory)

This is **the most basic format** of hypotheses upon which every other type of hypothesis is built.

Null Hypothesis:

- Represents a **base assumption** $(X = Y)$
- Can either be *accepted* or *rejected*

Alternative Hypothesis:

- Represents the **negation of the null hypothesis** $(X \neq Y)$
- Will be *accepted* or *rejected* based on whether the null hypothesis is found to be correct or not

$\rightarrow$ Usually, you will refer to every type of hypothesis in this context.

Null vs. Alternative Hypotheses (Example)

- *"Our null expectation, if climate niche is expanding randomly or equally on all niche peripheries, [...]. This would result in an EI (Expansion Index) value of 0."*
  Ralston, J. et al. (2016) ’Population trends influence species ability to track climate change’, Global Change Biology, pp. 1-10. doi: 10.1111/gcb.13478.
- *"[...] detect vegetation cover [...]. Chi squared test was applied to test the null hypothesis of no effects. [...] logistic regression model performs better than the null model [...]"*
  Nioti, F. et al. (2015) ’A Remote Sensing and GIS Approach to Study the Long-Term Vegetation Recovery of a Fire-Affected Pine Forest in Southern Greece’, Remote Sensing, 7(6), pp. 7712-7731. doi: 10.3390/rs70607712.

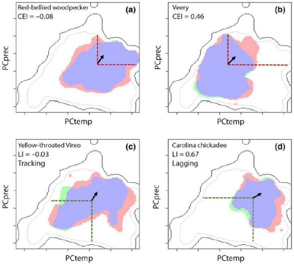


Fig. 2 Climate niche diagrams for four example species demonstrating the range of Expansion Index (EI) and Lagging Index (LI) values, and the relative influence of climate change (black arrow) on these climate response indices. (a) Species with low EI values tended to be geographically expanding with niche expansion (red shading) on all niche margins. (b) Species with high EI tended to be declining with niche expansion confined to the Climate Change Quadrant (red dashed line). (c) Species with low LI values showed very little niche unfilling (green shading) in the Climate Change Opposite Quadrant (CCOQ; green dashed line), indicating an ability to track their climate niche to new sites. (d) Species with high LI values had a larger proportion of unfilling falling within the CCOQ, indicating lagging. a and c represent species with relatively little influence of climate change on niche expansion and unfilling, respectively. b and d represent species with greater influence of climate change on niche expansion and unfilling, respectively. Climate niche diagrams for all 46 species of North American birds used in our analyses are included in the Appendix S3.

Ralston, J. et al. (2016) ’Population trends influence species ability to track climate change’, Global Change Biology, pp. 1-10. doi: 10.1111/gcb.13478.

Difference Hypotheses

This format of hypotheses is built upon postulated **differences in variable parameters within samples**.

In Theory:

- A difference in certain variable parameters (see seminar 4) between multiple samples is postulated
- $X \neq Y$

In Practice:

- *"[...] difference in the rate of treated bleeding events [...] between [...] prophylaxis (group A) and [...] no prophylaxis (group B) [...]"*
  Oldenberg, J. et al. (2017) 'Emicizumab prophylaxis in hemophilia A with inhibitors', N.Engl.J Med., pp. 1-10. doi: 10.1056/NEJMoa1703068.
- *"[...] could enable the plant to react differently to the next frost spell"*
  Walter, J. et al. (2013) 'Ecological stress memory and cross stress tolerance in plants in the face of climate extremes', Environmental and Experimental Botany. Elsevier B.V., 94, pp. 3-8. doi: 10.1016/j.envexpbot.2012.02.009.

Equivalence Hypotheses

This format of hypotheses is built upon postulated **equivalence of variable parameters within samples**.

In Theory:

- An equivalence of certain variable parameters (see seminar 4) between multiple samples is postulated
- $X \approx Y$

In Practice:

- *"Thresholds are equivalent to tipping points [...]"*
  Angeler, D. G. and Allen, C. R. (2016) 'Quantifying Resilience', Applied Ecology, pp. 617-624. doi: 10.1111/1365-2664.12649.
- *"Just as LAI is the canopy equivalent of leaf area, so ϵ_g^* is the canopy equivalent of the quantum yield."*
  Prince, S. D. and Goward, S. N. (1995) 'Global primary production: a remote sensing approach', Journal of Biogeography, pp. 815-835. doi: Doi 10.2307/2845983.

Relationship Hypotheses

This format of hypotheses is built upon postulated **relationships of variables within a population**.

In Theory:

- A relationship of multiple variables within a population is postulated
- $X \sim Y$

In Practice:

- *"[...] yield significant relationships between GPP and tree diversity."*
  Nightingale, J. M. et al. (2008) 'PREDICTING TREE DIVERSITY ACROSS THE UNITED STATES AS A FUNCTION OF MODELED GROSS PRIMARY PRODUCTION', Ecological Applications, 18(1), p. 93. Available at: http://dx.doi.org/10.1890/07-0693.1.
- *"[...] test for a series of hypothetical relationships (i.e., linear through to threshold) between ecological response variable and environment [...]"*
  Seddon, A. et al. (2014) 'A quantitative framework for analysis of regime shifts in a Galapagos coastal lagoon', Ecology, 95(11), pp. 3046-3055. doi: 10.1890/13-1974.1.

Directional vs. Non-Directional Hypotheses (Theory)

This format of hypotheses is built on postulated **connections and/or differences of variables within samples**.

Directional Hypothesis:

- Statement about the direction these connections or differences are postulated to function along
- $X > Y$; $X \leq Y$; $X < Y$; $X \geq Y$

Non-Directional Hypothesis:

- No statement about the direction these connections or differences are postulated to function along
- $X \neq Y$ with $X \quad ? \quad Y$

Directional vs. Non-Directional Hypotheses (Example)

- *"[...] a tenfold variation in mineralization rates from sand dunes to fertilized meadows (Ellenberg 1977) was associated with a 12-fold increase in ANPP (Poorter & de Jong 1999) [...]"*
  Lavorel, S. and Garnier, E. (2002) 'Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the Holy Grail', Functional Ecology, 16(Essay Review), pp. 545-556. doi: Doi 10.1046/J.1365-2435.2002.00664.X.
- *"Individual tropical trees show incredibly strong and persistent variation in long-term growth rates, resulting in a fourfold variation in the ages of similarly sized trees."*
  Brienen, R. J. W., Sch, J. and Zuidema, P. A. (2016) 'Tree Rings in the Tropics: Insights into the Ecology and Climate Sensitivity of Tropical Trees', in Tropical Tree Physiology. doi: 10.1007/978-3-319-27422-5.

Table 2. Nitrogen mineralization rate, above-ground net primary productivity (ANPP) and leaf characteristics of dominant species taken from various vegetation types in Central Europe

Vegetation type/ dominant species	Net N mineralization (kg N ha^{-1} $year^{-1}$)	Minimum above-ground biomass (g m^{-2})	ANPP (g m^{-2} $year^{-1}$)	Mean SLA (m^2 kg^{-1})	Mean leaf N concentration (mg g^{-1})	Estimated A_{max} (nmol g^{-1} s^{-1})
Ellenberg/Poorter						
Sand dunes	12–19	≈0	90	9·9	13·5	67·2
Heath	11–30	≈700*	210	18·7	16·7	124
Chalk grasslands	20–30	≈0	330	21·3	15·7	130
Fertilized meadows	130–160	≈0	1080	31·8	36·1	328
Aerts and co-workers						
Wet heathland:						
Erica tetralix	4·4	600	376	8·0†	12·6	54·8
Molinia caerulea	7·8	117	867	21·3†	19·3	152
Dry heathland:						
Calluna vulgaris	6·2	710	540	8·0†	na	na
Molinia caerulea	10·9	56	614	22·7†	14·0	125

10x

12x

Lavorel, S. and Garnier, E. (2002) 'Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the Holy Grail', Functional Ecology, 16(Essay Review), pp. 545-556. doi: Doi 10.1046/J.1365-2435.2002.00664.X.

Specified vs. Non-Specified Hypotheses (Theory)

This format is built on postulated **effect sizes** of treatments/groupings in experimental/observational set-up.

Specified Hypothesis:

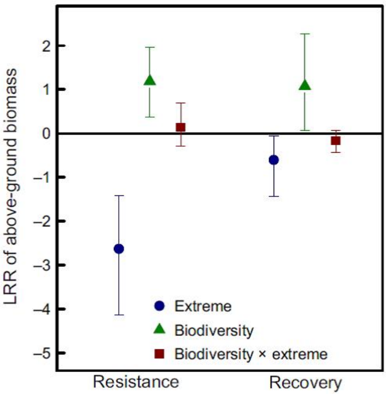
- Statement about an expected effect size/intensity within a set of response variables based upon a set of predictor variables.
- $X = \beta * Y$ with β being some pre-defined coefficient

Non-Specified Hypothesis:

- Statement about an expected effect within a set of response variables based upon a set of predictor variables without a notion of an effect size/intensity.
- $X = \beta * Y$ with β being some undefined coefficient

Specified vs. Non-Specified Hypotheses (Example)

- *"The effect size of diversity (natural log response ratio; LRR) is based on the comparison of high and low species richness levels [...]"*
  De Boeck, H. J. et al. (2017) 'Patterns and drivers of biodiversity-stability relationships under climate extremes', Journal of Ecology, (October), pp. 1-13. doi: 10.1111/1365-2745.12897.
- *"[...] a sample of 51 participants with a withdrawal rate of 10% in the control group would provide a power of more than 95% at a two-sided significance level of 0.05 to detect an effect size of 4/18 = 0.22 (null hypothesis: rate ratio = 1)."*
  Oldenberg, J. et al. (2017) 'Emicizumab prophylaxis in hemophilia A with inhibitors', N.Engl.J Med., pp. 1-10. doi: 10.1056/NEJMoa1703068.

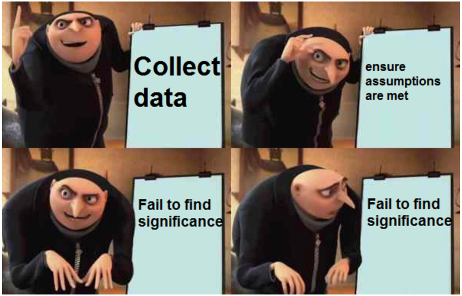


De Boeck, H. J. et al. (2017) 'Patterns and drivers of biodiversity-stability relationships under climate extremes', Journal of Ecology, (October), pp. 1-13. doi: 10.1111/1365-2745.12897.

How To Go About Testing Hypotheses

This process is highly variable but can be broken down into the following general, consecutive steps:

1. Establish a hypothesis (in terms of *Null* and *Alternative*)
2. Plan study and collect data
3. **Testing**
   - *Assumption check*
   - *Exploratory analyses* (seminar 4)
   - *Data visualisation* (seminar 5)
   - *Final analysis*
4. **Exporting results** and final plotting



Planning A Study And Collecting Data

Study design is part of many other courses. A few personal tips:

- Establish a **schedule** for your project
- Use **journal(s)** to record:
  - Weekly ToDo lists
  - Important talks with supervisors/co-authors
  - Note down spontaneous ideas for the project
- **Talk** about it

When **collecting data** ensure that

- **Relevant standards** and **standardised measuring schemes** are used (e.g.: Pérez-Harguindeguy, N. et al. (2013) 'New handbook for standardized measurement of plant functional traits worldwide', Australian Journal of Botany, 61, pp. 167-234. doi: http://dx.doi.org/10.1071/BT12225. for plant functional traits)
- **Relevant details** about data collection make it to the **final manuscript**

Sampling

Depending on how your project is structured, you will need to draw samples from your data. The most common sampling practices are:

Random sampling:

- Most commonly used
- Applicable when *true randomness* is desired

Use the `sample()` function in `R` (see seminar 1)

> Remember to make the sampling **reproducible!**

Stratified sampling:

- Applicable when *pseudo-randomness* is desired
  - Population is divided into groups (**strata**)
  - Random sampling is carried out for each strata
  - Strata samples are combined

Use the `stratified()` function in `R` or in-built functions of certain statistical test functions

Assumptions

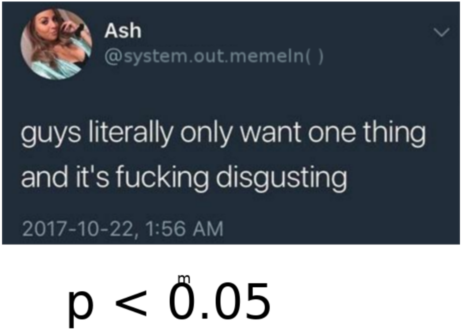
Statistical tests rely on individual *statistical assumptions*. Most prominent:

- **Normality**: Data follow a normal distribution (see seminar 3)
- **Randomness**: Data are truly random (see seminar 1)
- **Independence**: Data are independent
- **Homogeneity of variances**: Data from separate groups have same variance
- **Linearity**: Data have linear relationship

Testing

General procedure:

1. Select appropriate test (this should happen **before data collection**) based on:
   - Data structure
   - Variable scale
   - Statistical assumptions
   - Applicability to the hypothesis
2. Choose an appropriate test statistic (often pre-determined by the choice of test)
3. Test *statistical significance* (usually p-value)



Overview Of Tests

Tests come in a variety of forms. Too much to cover all of them in one seminar series.
$\rightarrow$ We will focus on a select few.

Tests can be classified according to their use of *parameters*:

Parametric Tests

- More *restrictive*
- Make *strict assumptions*
- **Easy to interpret**
- Require *less data*

Non-Parametric Tests

- Less *restrictive*
- Make *little to no assumptions*
- Often a **black box**
- Require *more data*

$\rightarrow$ We will focus on the **more basic tests of both categories**.

Choosing The Appropriate Test I

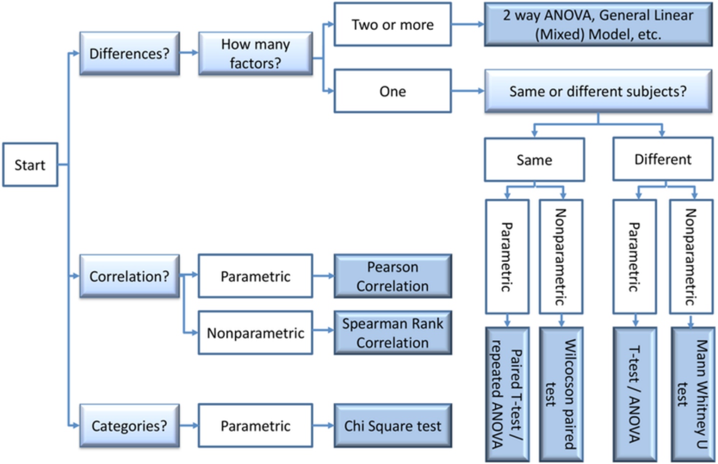
The choice of test depends on:

- The **dependent variable(s)** (≈ *Response(s)*)
  - Scale/Distribution (Type)
  - Number
- The **independent variable(s)** (≈ *Predictor(s)*)
  - Scale/Distribution (Type)
  - Number
- The **measure** employed by descriptive statistics which is to be tested on (seminar 4)

Number of Dependent Variables	Number of Independent Variables	Type of Dependent Variable(s)	Type of Independent Variable(s)	Measure	Test(s)
1	0 (1 population)	continuous normal	not applicable (none)	mean	one-sample t-test
		continuous non-normal		median	one-sample median
		categorical		proportions	Chi Square goodness-of-fit, binomial test
	1 (2 independent populations)	normal	2 categories	mean	2 independent sample t-test
		non-normal		medians	Mann Whitney, Wilcoxon rank sum test
		categorical		proportions	Chi square test Fisher's Exact test
	0 (1 population measured twice) or 1 (2 matched populations)	normal	not applicable/ categorical	means	paired t-test
		non-normal		medians	Wilcoxon signed ranks test
		categorical		proportions	McNemar, Chi-square test
	1 (3 or more populations)	normal	categorical	means	one-way ANOVA
		non-normal		medians	Kruskal Wallis
		categorical		proportions	Chi square test
	2 or more (e.g., 2-way ANOVA)	normal	categorical	means	Factorial ANOVA
		non-normal		medians	Friedman test
		categorical		proportions	log-linear, logistic regression
	0 (1 population measured 3 or more times)	normal	not applicable	means	Repeated measures ANOVA
	1	normal	continuous		correlation simple linear regression
		non-normal			non-parametric correlation
		categorical	categorical or continuous		logistic regression
			continuous		discriminant analysis
	2 or more	normal	continuous		multiple linear regression
		non-normal			
		categorical			logistic regression
		normal	mixed categorical and continuous		Analysis of Covariance General Linear Models (regression)
		non-normal			
		categorical			logistic regression
2	2 or more	normal	categorical		MANOVA
2 or more	2 or more	normal	continuous		multivariate multiple linear regression
2 sets of 2 or more	0	normal	not applicable		canonical correlation
2 or more	0	normal	not applicable		factor analysis

Source: James D. Leeper, Ph.D. (University of Alabama)

Choosing The Appropriate Test II

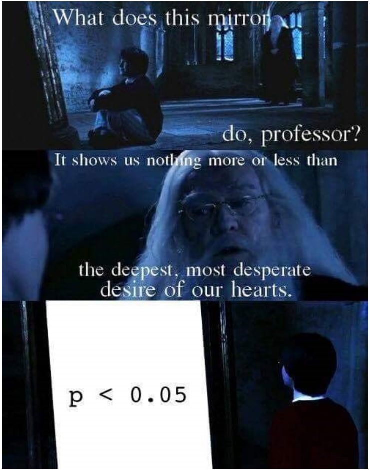


The p-value: Abstraction, Distraction And Action I

The p-value is **the** measure of statistical significance in contemporary science!

A p-value **below** the significance **cut-off value** (usually 0.05) indicates a **significant test metric**

p-values are **subject to** a heated **debate** (seminar 1) as everyone wants significant results and the concept of the p-value is **often misunderstood**



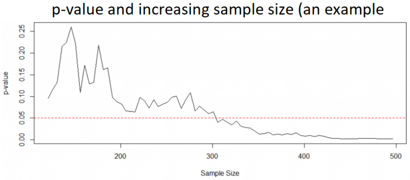
The p-value: Abstraction, Distraction And Action II

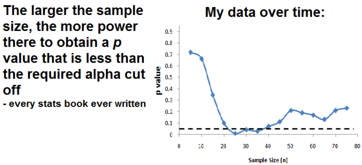
The common **misconceptions**:

- *"The p-value is the probability that the null hypothesis is true"*
- *"The p-value is the probability that the observed effects were produced by random chance alone."*
- *"The p-value does indicate the size or importance of the observed effect."*

The **right interpretation**:

- *"The p-value is the probability of randomly obtaining an effect at least as extreme as the one in your sample data, given the null hypothesis."*





The 0.05 significance level is an arbitrary convention!

Errors I

Uncertainty is an **inherent property** of any statistical method.

Statistical errors can be of:

- **Type I** (*"True Negative"*)
- **Type II** (*"False Positive"*)

Statistical errors are **impossible to avoid** but we can aim to **make as few as possible**.

	The null hypothesis (H_0) is	
Statistical result	True	False
Reject null hypothesis	**Type I error,** α value = probability of falsely rejecting H_0	Probability of correctly rejecting H_0: (1 - ß) = power
Accept null hypothesis	Probability of correctly accepting H_0 : (1 - α)	**Type II error,** ß value = probability of falsely accepting H_0

$\rightarrow$ Optimise α and β cut-off values (there is a trade off between them)

Errors II

Doctor: your diabetes test result is positive, it's Type I

Statistician: Oh that's great news!!

Doctor: How so?

Statistician: that means I don't have diabetes!

OUR RESEARCH PROJECT

Evolution of *Passer domesticus* in Response to Climate Change

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Motivation

Climate Change:

- Increasingly warming temperatures
- Increasing frequency and intensity of climate extremes

De Boeck, H. J. et al. Patterns and drivers of biodiversity-stability relationships under climate extremes. J. Ecol. 1-13 (2017). doi:10.1111/1365-2745.12897

Understanding patterns of evolution caused by climate change is vital for mankind.

Biological Consequences:

- Pole-ward range shifts of species have been observed

Ralston, J. et al. Population trends influence species ability to track climate change. Glob. Chang. Biol. 1-10 (2016). doi:10.1111/gcb.13478

- Recent evolutionary processes can be linked to climate change

Parmesan, C. Ecological and Evolutionary Responses to Recent Climate Change. Annu. Rev. Ecol. Evol. Syst. 37, 637-669 (2006).

- Mankind relies on ecosystem services which may be affected by climate change

Truchy, A. et al. Linking biodiversity, ecosystem functioning and services, and ecological resilience: Towards an integrative framework for improved management. Advances in Ecological Research 53, (Elsevier Ltd., 2015).

Studying Climate Change

- Climate change is a **temporal** phenomenon
  $\rightarrow$ Usually studied through *time-series based* approaches
- What if we don't have time-series data?
  $\rightarrow$ We can *trade* **time** for **space** making use of the **spatial aspect** of climate change

Warming Effects:

- The spatial equivalent to temporal warming effects of climate change manifests on **latitudinal gradients.** Equatorward placement of organisms imposes a warming effect.

Climate Extremes:

- The spatial equivalent to temporal changes in frequency and intensity of climate extremes can be expressed via **continental** (extreme) **and coastal** (moderate) **climate** patterns

Studying Evolution

- Evolution is an inherently **temporal** as well as **spatial** phenomenon
  $\rightarrow$ Studied through *time-series based* approaches, **phylogenies**, etc.

Temporal Aspects:

- What if we don't have time-series data at one location?
  $\rightarrow$ We can *trade* **time** for **space** as long as there are gradients representing evolutionary forcing
- **Latitudinal and longitudinal gradients** can be regarded as representative of glimpses into future or past environmental conditions of species.

Spatial Aspects:

- Evolution relies on the separation of populations for differences to arise (divergent evolution)
  $\rightarrow$ We need to select **populations** that are **in no feasible reproductive contact**
- **Invasive** vs. **Non-Invasive Populations** can be used to draw conclusions about divergent evolutionary patterns

Our Study Organism

Passer domesticus - The Common House Sparrow

- Present globally
  → Lends itself to **gradient-based approaches**
- Non-migratory & Invasive species in some parts of the world
  → Studies of **divergent evolution** are possible
- Well-researched
  → **Comparative analyses** are possible

Warming Effects

Equatoward location driven warming effects alter the size and bodyweight of individual sparrows.

Variables	
Weight:	Weight is a reliable indicator of how much resources have been amassed by an individual sparrow.
Height:	Height influences exposure to surrounding temperatures through stature and surface area thus indicating heat loss potential of an individual sparrow.
Wing Span:	Wing span is the horizontal analogue to height measurements and can be indicative of heat loss potential of individual sparrows.

According to Bergmann's rule, organisms of the same species tend to grow bigger and heavier in colder climates since larger animals have a lower surface area to volume ratio thus radiating less body heat per unit of mass and conserving energy.

Climate Extremes

Sparrows residing in areas characterised by extreme climate events will differ from sparrows in more stable environments.

Variables	
Weight:	Weight of individual sparrows is representative of energy resources.
Population Size:	Population size is an important factor of carrying capacity of habitats.
No. Eggs:	Number of eggs reflects investment in offspring.
Egg Weight:	Weight of individual eggs is representative of investment in individual offspring.

We expect sparrows in more extreme climatic conditions to have stored vast amounts of energy whilst the habitats themselves exhibit lower carrying capacities.

Competition

Competition is more pronounced in certain areas leading to changes in sparrow physiology and behaviour.

Variables	
Flock Size:	Flock size is an indicator of the rate of resource depletion and resource availability.
Home Range:	Home range is a measure of how far an individual will fly to forage.
Weight:	Individual weight is a measure of how well an individual does in competing for food.
Sex:	Differences in fitness due to competition may be a result of sexual differences.

We expect sparrows in less hospitable habitats to group in smaller flocks with bigger home ranges.

Predation

Presence and type of predator will influence sparrow behaviour and physiology.

Variables	
Predator Presence:	Indicating whether a predator is present.
Predator Type:	Indicating the kind of predator that is present.
Nesting Site:	Where a sparrow nest is located.
Nesting Height:	How height the nesting site is from the ground.
Colour:	Colour is one of the main factors to conspicuousness.
Flock Size:	Flock size is one of the main factors to conspicuousness.

We expect sparrows which are under pressure from predation to nest differently than ones which are not.

Sexual Dimorphism

Sexual dimorphism is less/more pronounced in invasive or non-invasive species.

Variables	
Weight:	Differences in weight of individuals of different sexes are key to uncovering sexual dimorphism.
Colour:	Displays of colour greatly influence competition for mates which is often subject to a structure of sexual dimorphism.

We expect sexual dimorphism to be more pronounced in invasive populations of sparrows as these are located in environments which are much less hostile to them due to an initial lack of predators and thus increased fitness (and ability to invest in sexually dimorphic displays).

Study Setup I

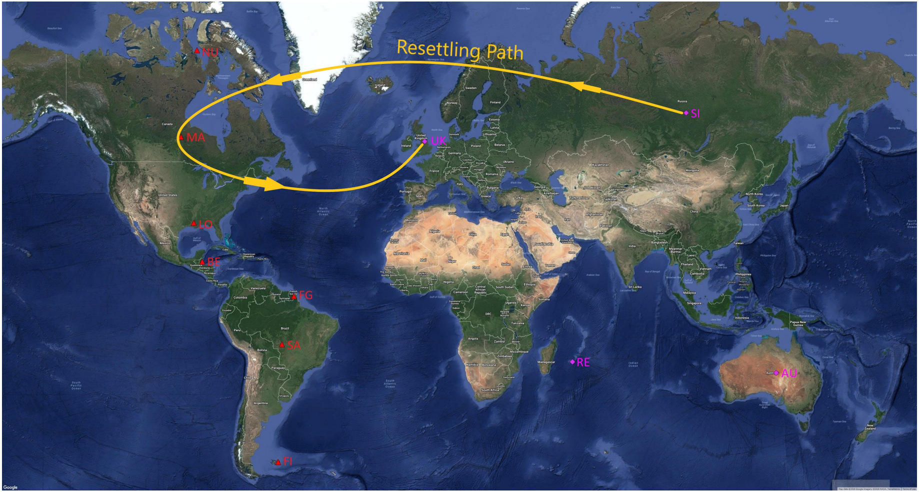
Set-up of **11 Sites** chosen according to **three factors/treatments** (latitude, climate and population status):

Site	Index	Lat [°]	Lon [°]	Climate	Population Status
Siberia	SI	60	100	Continental	Native
United Kingdom	UK	54	-2	Coastal	Native
Australia	AU	-25	135	Continental	Introduced
Reunion	RE	-21.1	55.6	Coastal	Introduced
Nunavut	NU	70	-90	Coastal	Introduced
Manitoba	MA	55	-97	Semi-Coastal	Introduced
Louisiana	LO	31	-92	Coastal	Introduced
Belize	BE	17.25	-88.75	Coastal	Introduced
French Guiana	FG	4	-53	Coastal	Introduced
South America	SA	-14.6	-57.7	Coastal	Introduced
Falkland Isles	FI	-51.75	-59.17	Coastal	Introduced

Source: https://www.cabi.org/isc/datasheet/38975, retrieved 21/01/2018

We also resettle the population of SI to MA and UK

Study Setup II



We have three data files. Check the README file for a data description.

Using Our Data I

ATTENTION!

All the data we will use is simulated!

Data Management and data cleaning will be done throughout seminar 7 (Data Handling and Data Mining).

The actual analyses will be done in the following seminars (8-12) using these statistical approaches:

Nominal Tests (Seminar 8)

- Binomial
- McNemar
- Cochqran's Q
- Chi^2

Correlation Tests (Seminar 9)

- Pearson
- Rank
- Spearman
- Kendall's Tau

Using Our Data II

Analyses covered in this seminar series (continued):

Ordinal/Metric tests for two-sample situations (Seminar 10)

- Mann-Whitney U
- Wilcoxon signed-rank test

Parametric Tests (Seminar 12)

- t-test
- ANOVA
- (M)AN(C)OVA

Ordinal/Metric tests for more than two-sample situations (Seminar 11)

- Kruskal-Wallis H test
- Friedman test

Some **advanced statistical methods** will be touched on in seminar 13 (**Summary, Manuscript Workflow and an Outlook on Advanced Statistics**)

A finalised script (of `R` code) will be produced in seminar 13 (**Summary, Manuscript Workflow and an Outlook on Advanced Statistics**)