

# Mantel Test

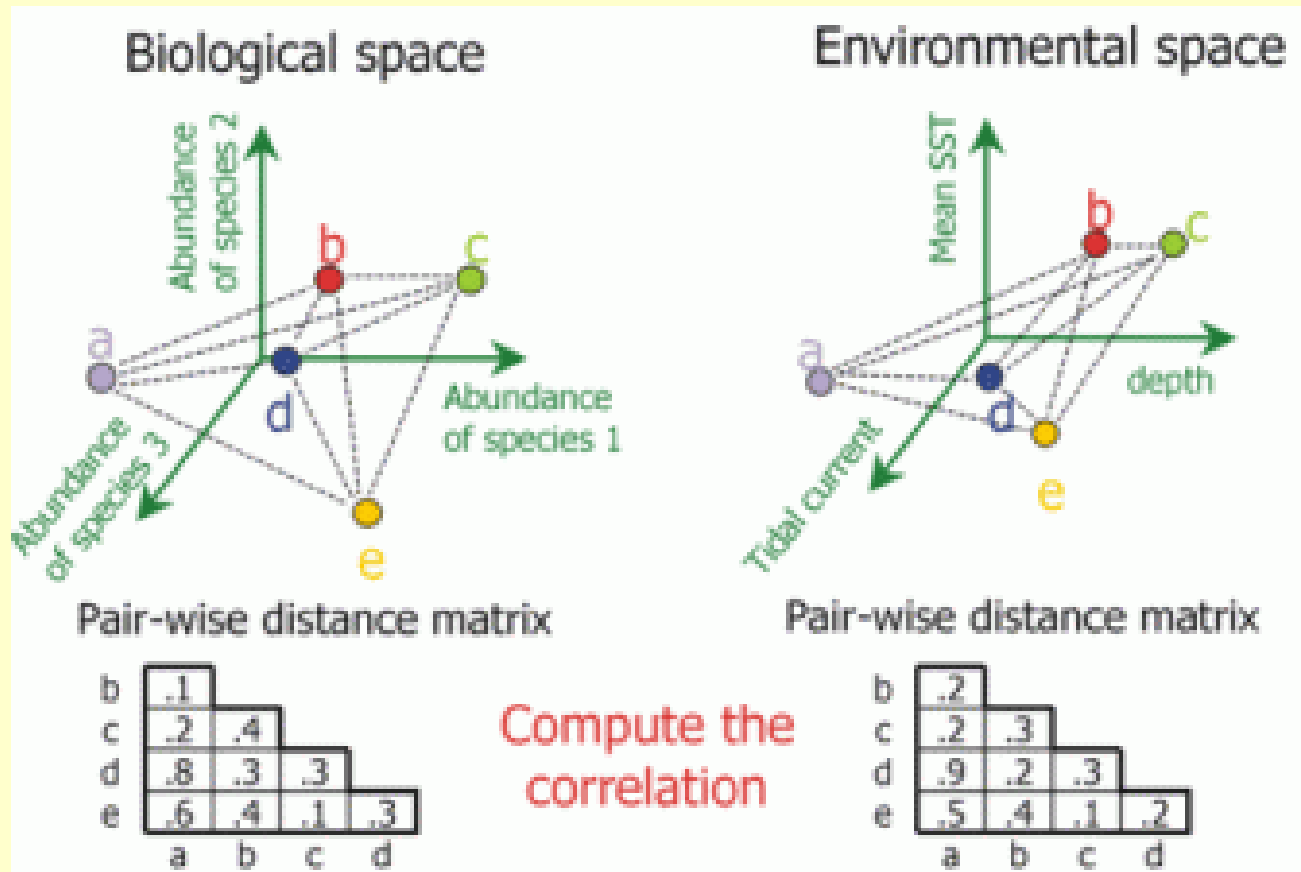
➤ *Objectives:*

Discuss the rationale for the Mantel Test

Illustrate the use of the Mantel Test

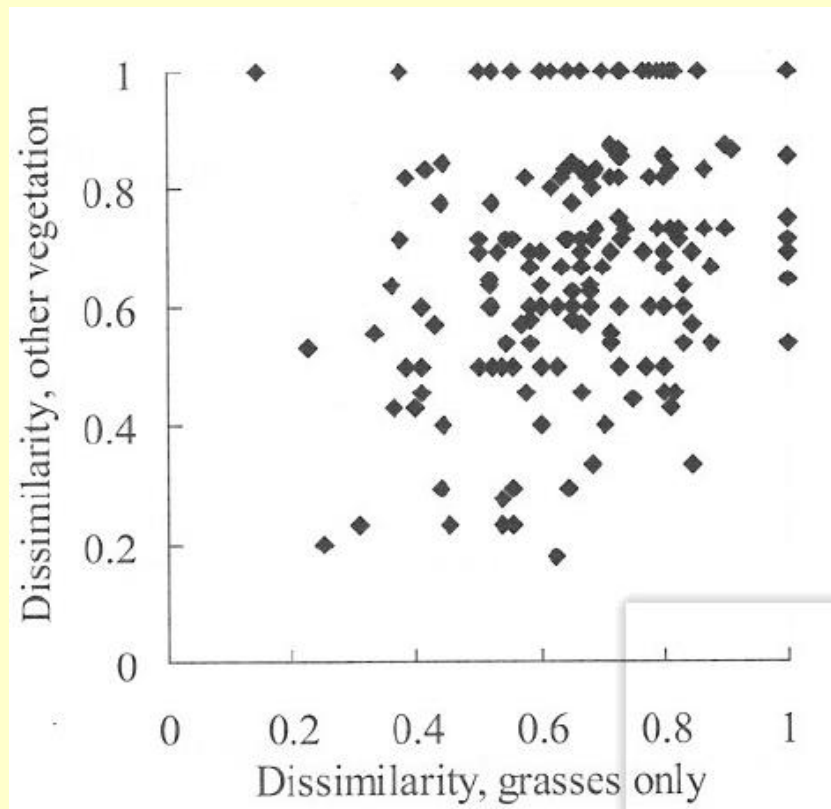
# Mantel Test - Intro

- *Objective* : Compare distances between all pairs of samples (plots) in terms of their species / environment



# Mantel Test - Intro

Mantel's (1967) test is based on a regression analysis, in which the variables are distance dissimilarity (or similarity) matrices summarizing all pairwise sample combinations.



# Mantel Test - Intro

The operative question is: "Do samples that are similar in terms of the predictor (environmental) variables also tend to be similar in terms of the dependent (species) variable ?"

The power and versatility of Mantel's test derives from the various ways that the distance matrices and the regression can be framed.

One important case the Mantel test considers explicitly is the scenario where the predictor variable is space itself, measured as geographic location (e.g., as (lat, long) coordinates). In this case, the operational question is:

"Are closer samples compositionally more similar ?"

# Mantel Test - Approach

Mantel's statistic based on a simple cross-product term:

$$z = \sum_{i=1}^n \sum_{j=1}^n x_{ij} y_{ij}$$

And is normalized as follows: (to consider variables of different measurement units in same framework)

$$r = \frac{1}{(n-1)} \sum_{i=1}^n \sum_{j=1}^n \frac{(x_{ij} - \bar{x})}{s_x} \cdot \frac{(y_{ij} - \bar{y})}{s_y}$$

Where  $x$  and  $y$  are variables measured at locations  $i$  and  $j$ ,  
 $n$  is the number of elements in the distance matrices  
( $n = m * (m-1) / 2$  for  $m$  sample locations)  
and  $s_x$  and  $s_y$  are standard deviations for variable  $x$  and  $y$ .

# Mantel Test - Approach

## Remember:

Mantel test is based on linear correlation and hence subject to the same assumptions of Pearson correlation

$$r = \frac{1}{(n-1)} \sum_{i=1}^n \sum_{j=1}^n \frac{(x_{ij} - \bar{X})}{s_x} \cdot \frac{(y_{ij} - \bar{Y})}{s_y}$$

Because of this limitation, permutation methods are often used for significance testing (when assumptions not met):

- distribution-free method
- empirical p value calculation

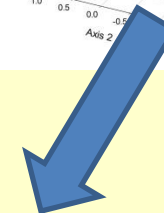
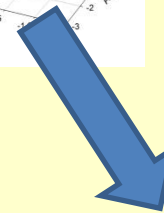
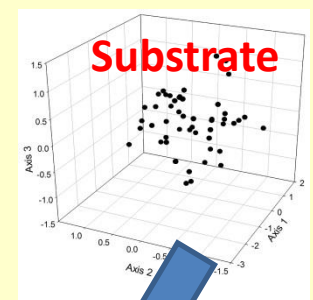
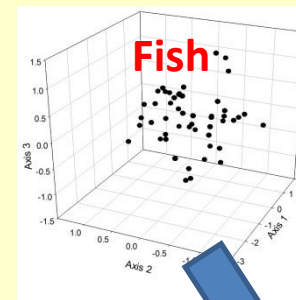
# Mantel Test – Example

Analyze fish / substrate data using two separate NMDS (Sorensen distance)

Performed Mantel test to assess null hypothesis of no relationship between similarity of fish community (matrix 1) and similarity of substrate (matrix 2).

Mantel test measures the strength of the relationship, using standardized Mantel test statistic ( $r$ ), ranging from: -1 (neg.) to 0 (no effect) to +1 (pos.).

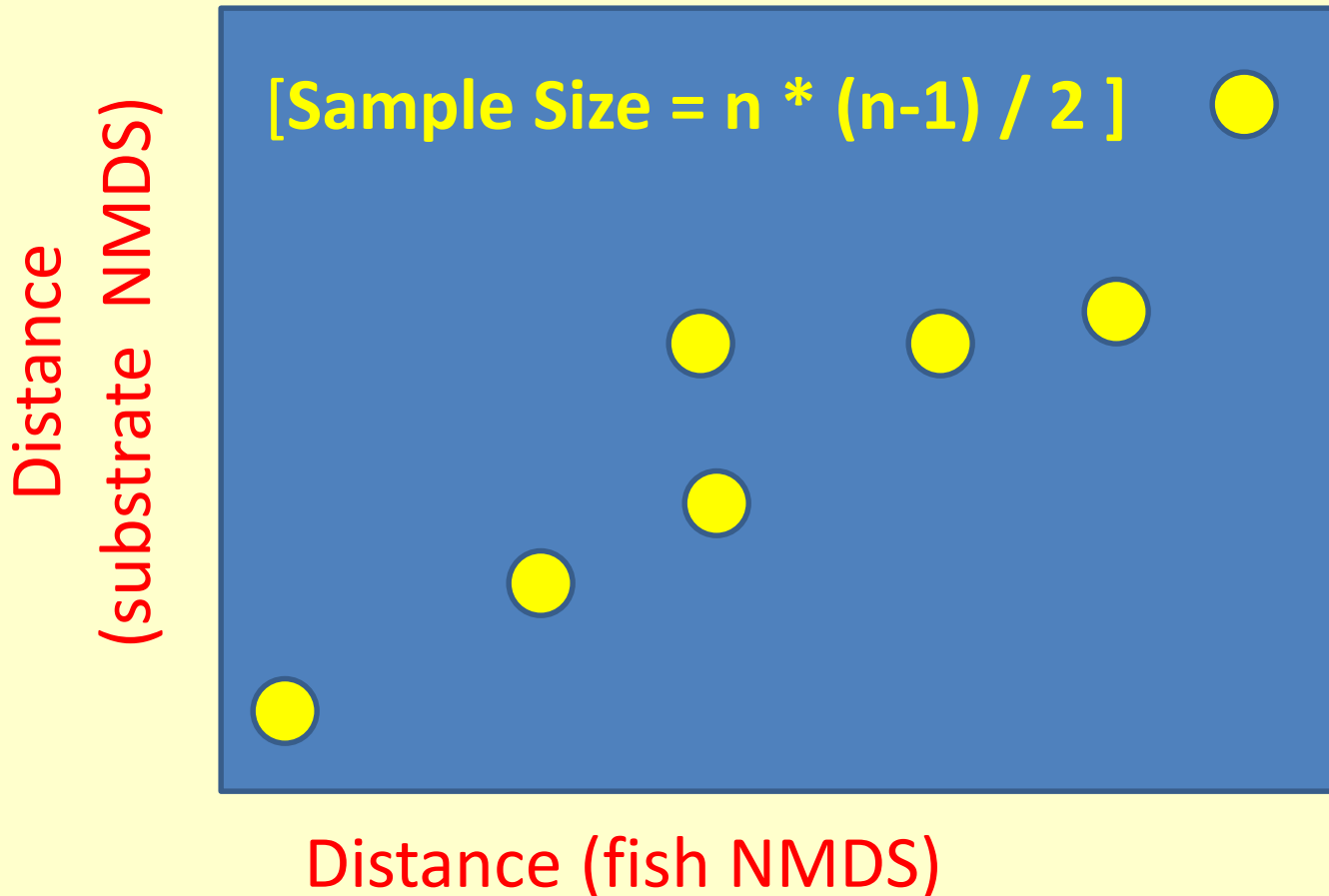
Associated  $p$  value from 999 randomizations.



$r$

# Mantel Test – Example

Significant positive relationship between structure of fish assemblages and substrates ( $r = 0.20$ ,  $p < 0.001$ , 999 runs).



**Remember:**

Ecological  
vs  
Statistical  
Significance

( $r^2$  vs  $p$ )



# Mantel Test - Setup

1) How many variables ?

Simple  
(2-way)

or

Partial  
(3-way)

2) Select distance measure

Mantel Test Setup

**Main Distance Measure**

- Sorensen (Bray-Curtis)
- Relative Sorensen
- Jaccard
- Euclidean (Pythagorean)
- Relative Euclidean
- Correlation
- Chi-squared
- Squared Euclidean

Main is already a distance matrix

**Second Distance Measure**

- Sorensen (Bray-Curtis)
- Relative Sorensen
- Jaccard
- Euclidean (Pythagorean)
- Relative Euclidean
- Correlation
- Chi-squared
- Squared Euclidean

Second is already a distance matrix

Partial mantel test

**Control Distance Measure**

- Sorensen (Bray-Curtis)
- Relative Sorensen
- Jaccard
- Euclidean (Pythagorean)
- Relative Euclidean
- Correlation
- Chi-squared
- Squared Euclidean

Control is already a distance matrix

**Output Options**

- List first distance matrix
- List second distance matrix
- List control distance matrix
- Write matrices in list format

**Method for Calculating P-value**

- Mantel's asymptotic approximation
- Randomization test

**Control Matrix**

History

# Mantel Test - Setup

## Two type of matrices

Normally the matrices will be typical PC-ORD data sets. If, however, one or more of the matrices is already a distance matrix, then select the appropriate checkboxes:

**Main matrix is already a distance matrix**

**Second matrix is already a distance matrix**

**Control matrix is already a distance matrix**

# Mantel Test - Setup

## Output Options

Use checkboxes to select:

**List first distance matrix**

**List second distance matrix**

**List control distance matrix** (relevant only with partial test)

**Write matrices in list format.**

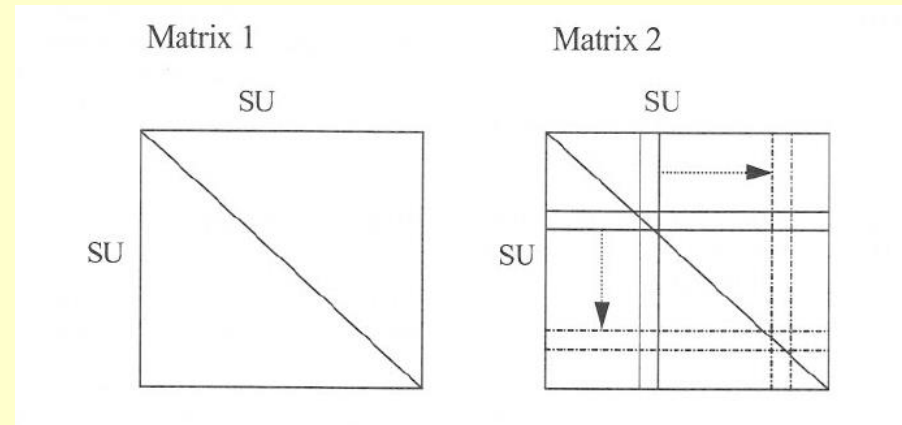
Non-redundant values in the distance matrix written as a list, in addition to the full matrix. If you select more than one distance matrices, corresponding distances are listed together.

# Mantel Test - Setup

## Method for calculating p value

Two methods are available:

1) Randomization test by  
by conducting permutations  
of the distance matrix



2) Asymptotic approximation method  
(faster with large data sets)

## NOTE:

For partial Mantel test, asymptotic approximation not available.

# Mantel Test - Setup

## Matrix Shuffling

Preserves internal symmetry of dataset:

	SU1	SU2	SU3	SU4
SU1	11	12	13	14
SU2	21	22	23	24
SU3	31	32	33	34
SU4	41	42	43	44

Select SU3 at random to swap with first item. Swap rows 1 and 3

	SU1	SU2	SU3	SU4
SU3	31	32	33	34
SU2	21	22	23	24
SU1	11	12	13	14
SU4	41	42	43	44

Swap columns 1 and 3

	SU3	SU2	SU1	SU4
SU3	33	32	31	34
SU2	23	22	21	24
SU1	13	12	11	14
SU4	43	42	41	44

# Mantel Test

## ➤ Example 1:

### **Simple Mantel's Test on a Predictor Matrix.**

If the dependent matrix is species similarity and the predictor matrix is a similarity matrix based on a set of environmental variables, then the simple test is for a correlation between the two matrices.

Such correlation would indicate that locations that are similar environmentally tend to be similar compositionally.

# Mantel Test - Output

First ordination: 14 points in ordination, 3 axes

Distance measure for first matrix = Euclidean (Pythagorean)

Second ordination: 14 points in ordination, 3 axes

Distance measure for second matrix = Euclidean (Pythagorean)

REDUNDANCY BETWEEN TWO SETS OF ORDINATION SCORES:

**0.007195 = r-squared** = Squared standardized Mantel statistic comparing Euclidean distances among points in one ordination with the Euclidean distances among points in the other ordination.

**0.719492 = Percent redundancy (shared pattern)**

# Mantel Test - Output

TEST STATISTIC: t-distribution with infinite degrees of freedom using asymptotic approximation of Mantel (1967).

If  $t < 0$ , then negative association is indicated.

If  $t > 0$ , then positive association is indicated.

MANTEL TEST RESULTS: asymptotic approximation method

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0.084823 = r = Standardized Mantel statistic

0.908917E+02 = Observed Z

0.899204E+02 = Expected Z

0.192620E+01 = Variance of Z

0.138788E+01 = Standard error of Z

0.699835E+00 = t

0.484254 = p (type I error)

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# Mantel Test

## ➤ Example 2:

### **Simple Mantel Test between Observed and Model Matrix.**

The model distance matrix might be provided as a simple binary matrix of 0's and 1's; alternatively, distances might be based on a more complicated model.

A simple example would be a case where the samples are each assigned to a group (e.g., community type) and the predictor variables are measured environmental variables.

The question is: are samples in the same group (community type) also similar in terms of the environmental variables? In this case two samples are similar (distance = 0) if they are both assigned to the same group, otherwise they are dissimilar (distance = 1).

# Mantel Test

## ➤ Simple Mantel Test between Observed Matrix and Model Matrix.

Show how  
Mantel tests  
used to test  
genetics  
distance  
Hypotheses:



*Genetics and Molecular Biology*, 25, 4, 435-439 (2002)  
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[www.sbg.org.br](http://www.sbg.org.br)

Research Article

Hypothesis testing of genetic similarity based on RAPD data using  
Mantel tests and model matrices

Flávia Melo Rodrigues<sup>1</sup>, José Alexandre Felizola Diniz-Filho<sup>2</sup>, Luiz Artur Mendes Bataus<sup>3</sup>  
and Rogério Pereira Bastos<sup>2</sup>

Mantel Test Calculation:

$$Z = \sum_i \sum_j (E_{ij} * M_{ij})$$

(Tested with Permutations)

# Mantel Test

## ➤ Simple Mantel Test between Observed Matrix and Model Matrix.

Model I: with a value of one if a given pair of species belonged to the same genus and zero elsewhere.

Model II: matrix representing connections among species in terms of their families.

Model III: combines two previous matrices: a value of 1.0 assigned to pairs of species in the same genus, 0.5 assigned to species in different genera but in the same family, and zero indicated that the pair of species being compared belonged to different families

**(Rodrigues et al. 2002)**

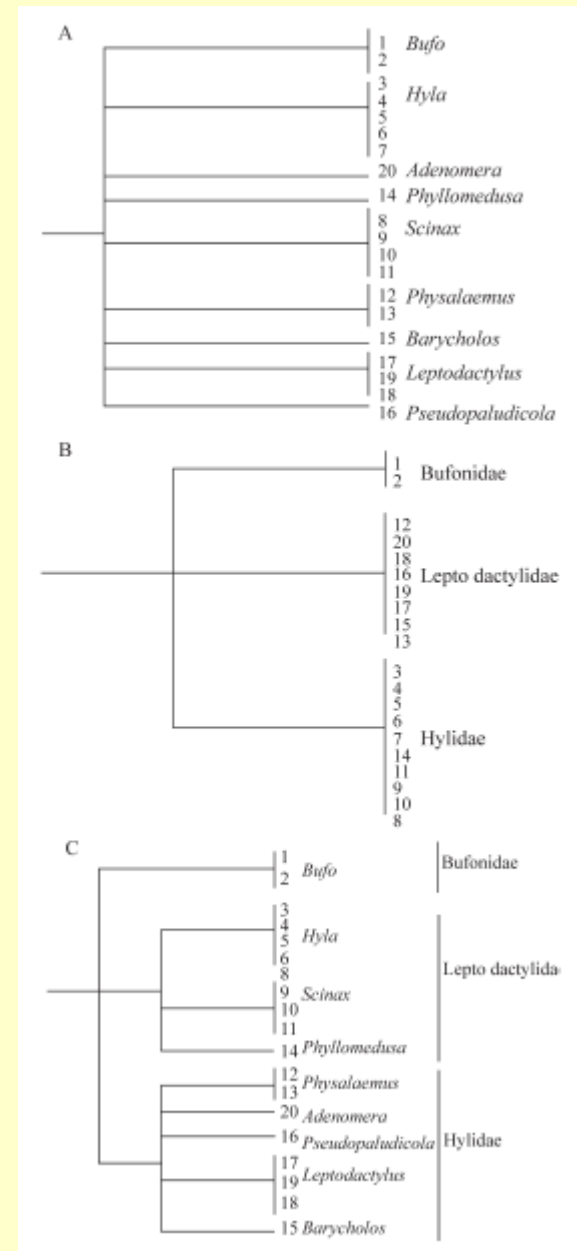
# Mantel Test

## ➤ Simple Mantel Test between Observed Matrix and Model Matrix.

**Model I:** with a value of 1.0 if a given pair of species in same genus and 0.0 elsewhere.

**Model II:** matrix representing connections among species in terms of their families.

**Model III:** combines two previous matrices: 1.0 assigned to pairs of species in same genus, 0.5 pairs of species in different genera but in same family, and 0.0 indicated that pair of species belonged to different families.



(Rodrigues et al. 2002)

# Mantel Test

- Simple Mantel Test between Observed Matrix and Model Matrix.

Primer	Same Genus		Same Family		Same Genus & Family	
	Model I		Model II		Model III	
	R	P	r	P	r	P
62	0.151	0.037	0.128	0.0620	0.160	0.027
63	0.043	0.301	0.021	0.3504	0.035	0.267
64	0.029	0.382	0.064	0.1950	0.059	0.212
OPA-01	0.063	0.421	0.040	0.1026	0.00002	0.346
OPA-03	0.227	0.018	0.148	0.0470	0.209	0.008
OPC-02	0.092	0.111	0.023	0.3370	0.059	0.197
OPC-15	0.077	0.164	0.030	0.3670	0.013	0.375
OPH-11	0.067	0.209	0.101	0.1116	0.103	0.102
OPH-14	0.044	0.426	0.014	0.4490	0.031	0.471
All primers	0.224	0.003	0.082	0.1304	0.161	0.018

(Rodrigues et al. 2002)

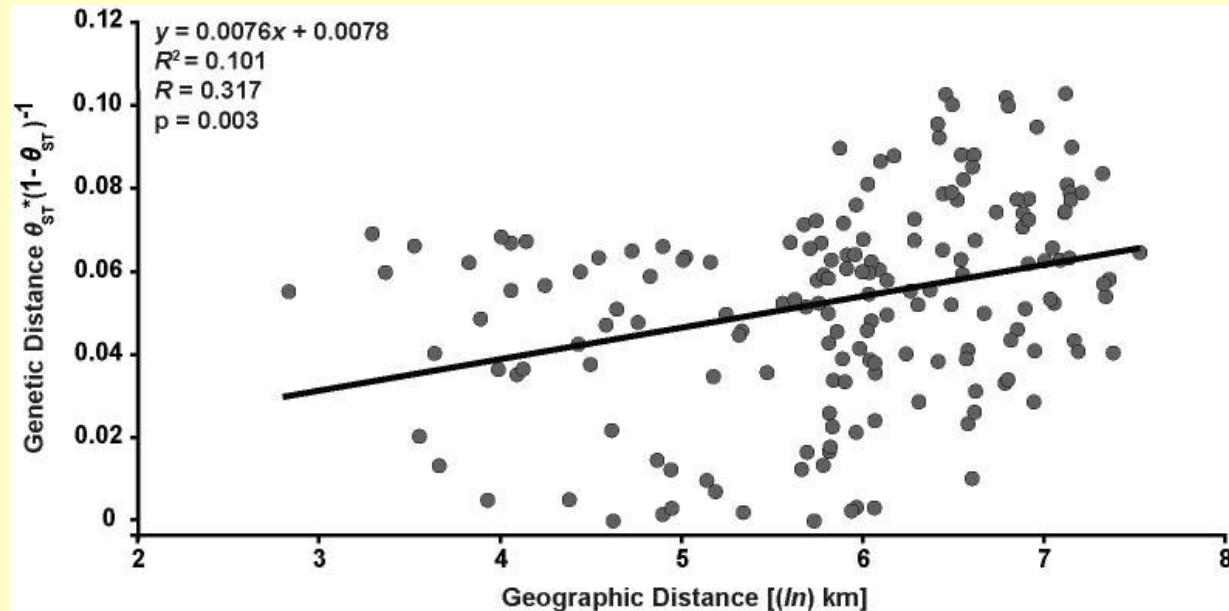
# Mantel Test

## ➤ Example 3: Simple Mantel's Test on Geographic Distance.

If the dependent distance matrix is species similarity and the predictor matrix is geographic distance ("space itself"), the research question is:

"Are samples that are close together also compositionally similar?"

Equivalent to testing for overall autocorrelation in the dependent matrix (i.e., averaged over all the distances).



# Mantel Test

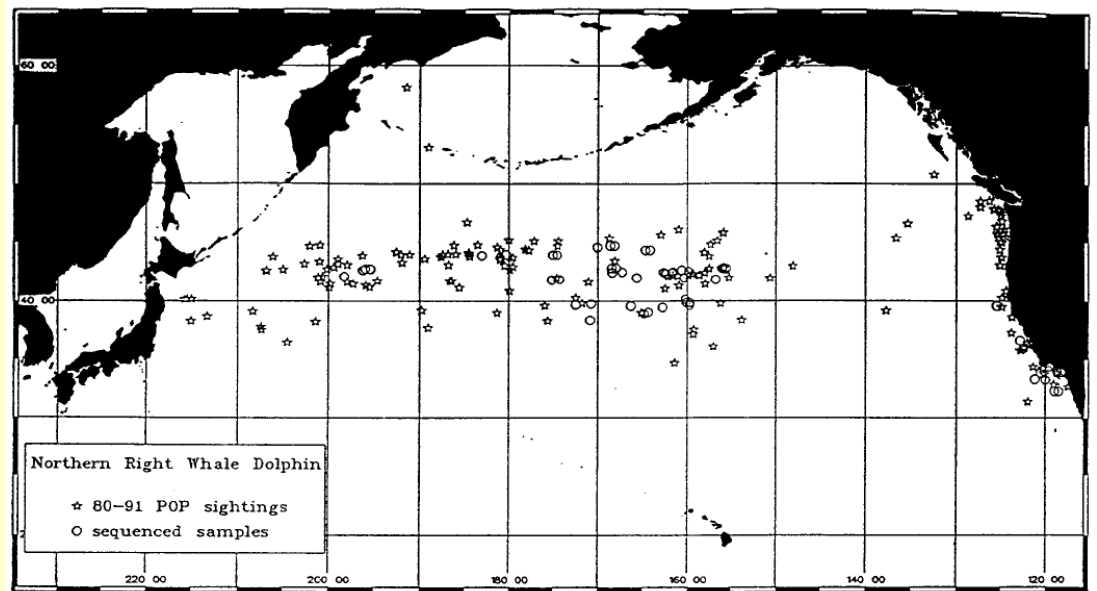
## ➤ Simple Mantel's Test on Geographic Distance.

DIZON ET AL.: GENETIC STOCKS AND STATISTICAL POWER  
CalCOFI Rep., Vol. 35, 1994

### INTRASPECIFIC STRUCTURE OF THE NORTHERN RIGHT WHALE DOLPHIN (*LISSODELPHIS BOREALIS*): THE POWER OF AN ANALYSIS OF MOLECULAR VARIATION FOR DIFFERENTIATING GENETIC STOCKS

ANDREW E. DIZON, CARRIE A. LEDUC<sup>1</sup>, AND R. GENE LEDUC

No correlation existed between genetic and geographic distance (normalized Mantel statistic  $Z = -0.005$ ).



Of 1000 permutations:  
49% had larger  $Z$  values than the sample matrix, 51% smaller ( $p = 0.49$ )

# Mantel Test

## ➤ Simple Mantel's Test on Geographic Distance.

Connectivity can be made more complex – to test specific models:

- some relationships are symmetrical
- some relationships are asymmetrical

e.g. Symmetrical Relationship:  
Group membership (like MRPP)

e.g., MPA connectivity

e.g., River connectivity

e.g., Behavioral ecology

	Plot														
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
plot1	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
plot2	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
plot3	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
plot4	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
plot5	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
plot6	1	1	1	1	1	0	0	0	0	0	1	1	1	1	1
plot7	1	1	1	1	1	0	0	0	0	0	1	1	1	1	1
plot8	1	1	1	1	1	0	0	0	0	0	1	1	1	1	1
plot9	1	1	1	1	1	0	0	0	0	0	1	1	1	1	1
plot10	1	1	1	1	1	0	0	0	0	0	1	1	1	1	1
plot11	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0
plot12	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0
plot13	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0
plot14	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0
plot15	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0



# Mantel Test

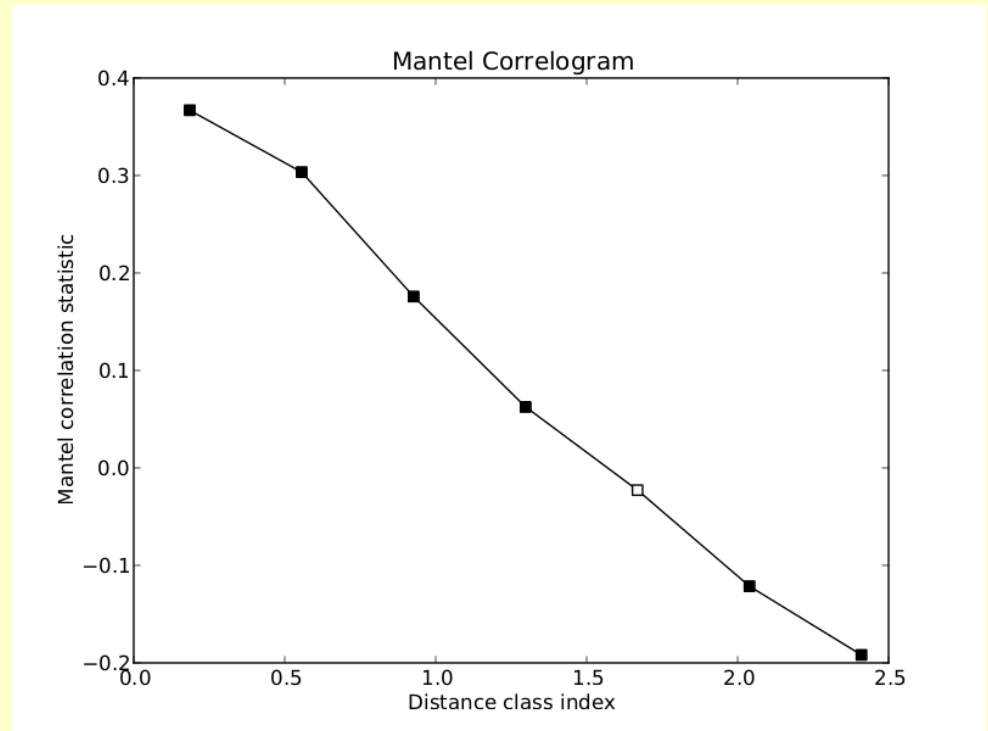
## ➤ Simple Mantel's Test on Geographic Distance.

**Limitation:** What if there is smaller-scale spatial structure ?

Test of spatial dependence averaged over all distances in the simple Mantel test.

Thus, test cannot discover changes in correlation at different distances (scales).

The Mantel correlogram overcomes problem, at extra computational expense.



(Borcard and Legendre 2012)

# Mantel Correlogram

## Case 4. The Mantel Correlogram.

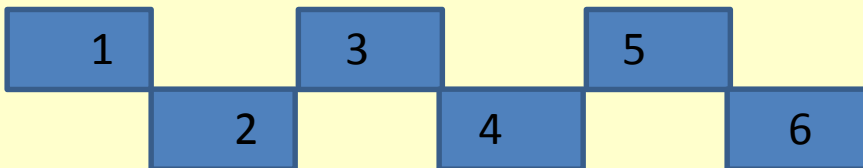
Special approach provides series of "model" adjacencies corresponding to membership within specific distance class.

### Quantifying auto-correlation:

Correlation as a function of the spatial / temporal lag



Lag = 1, Sample = 5 pairs



Lag = 2, Sample = 4 pairs

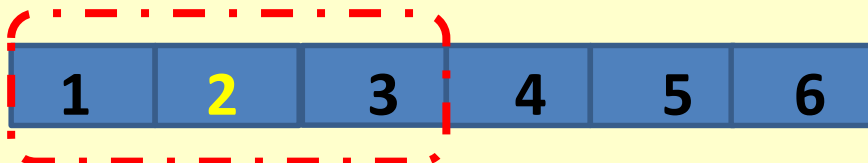
# Mantel Correlogram

## Case 4. The Mantel Correlogram.

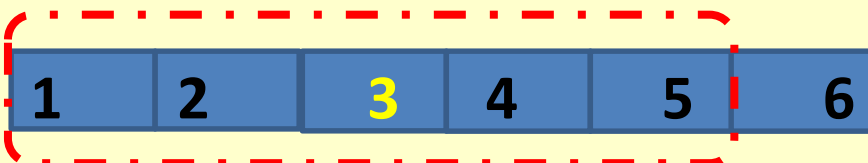
The first distance matrix "turned on" for all pairs of points within the first distance class; second matrix is scored for all pairs of points within second distance interval, and so on.

### Quantifying the Mantel Correlogram:

Correlation as a function of spatial / temporal neighborhood



Lag = 1

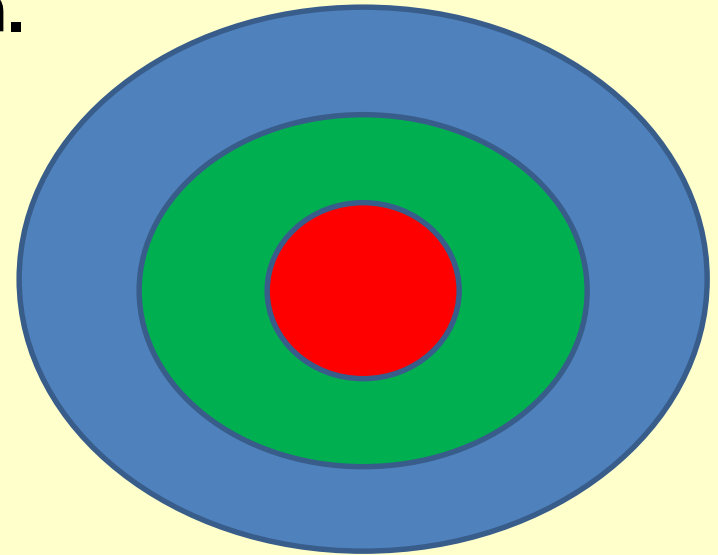


Lag = 2

# Mantel Correlogram

## Case 4. The Mantel Correlogram.

The result of this analysis is a correlogram, analogous to an autocorrelation function but performed on a distance matrix.



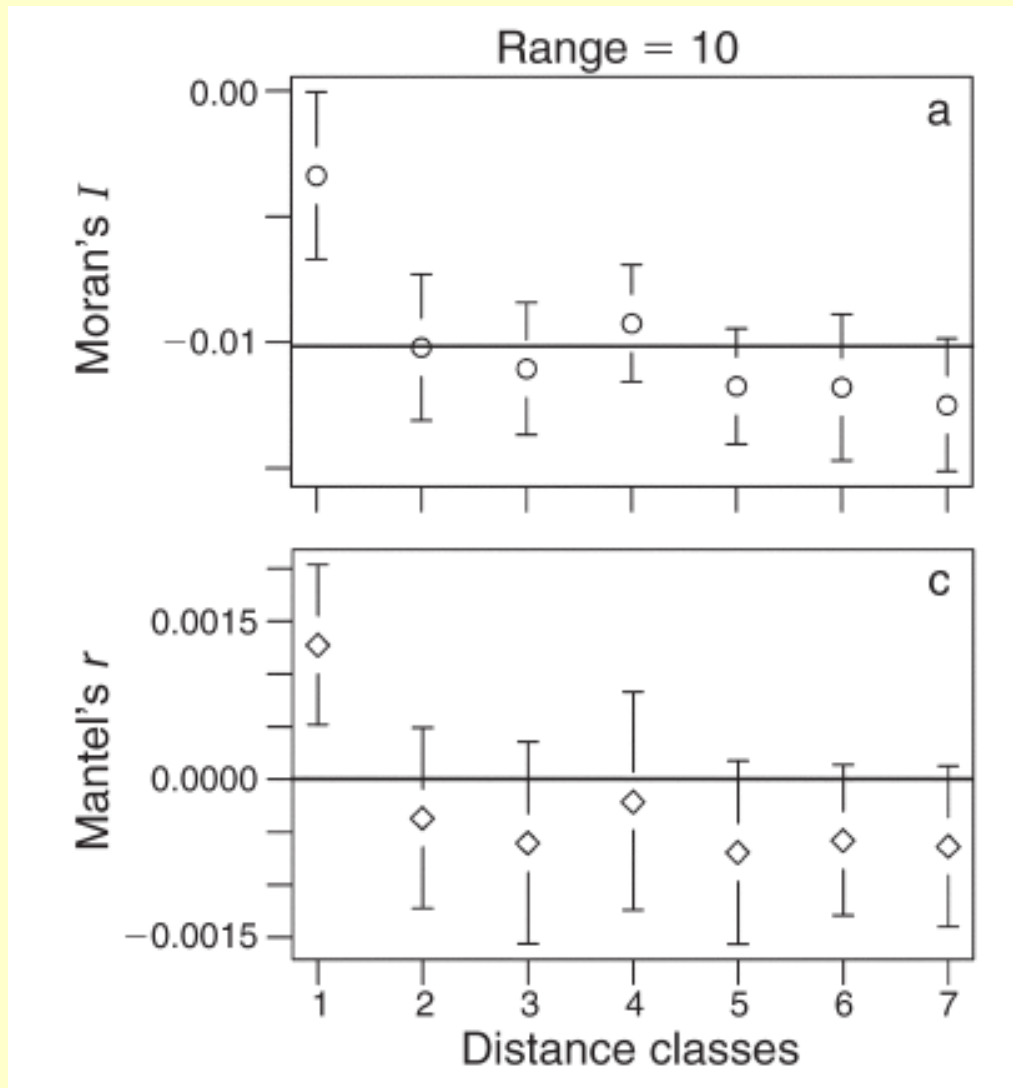
**Approach:** Define Neighborhood

both samples inside – matrix assigned a “1”

one sample “in” and one sample “out” – matrix assigned a “0”

Another appealing feature of the Mantel correlogram is that it may be multivariate – not bi-variate like Moran’s I.

# Mantel Correlogram



Moran's  $I$  measures spatial structure, as a function of the lag (distance classes)

## Model of Spatial Structure:

If closer than distance class, then matrix assigned 1

If farther than distance class, then matrix assigned 0

(Borcard and Legendre 2012)

# Summary

Mantel Test provides flexible approach for comparing two (or more) distance matrices (dissimilarity / similarity)

This approach uses linear regression, with the inherent benefits / limitations of this approach.

Flexibility in methods for establishing similarity (connectivity rules) and significance testing allows for many applications.

A particularly attractive extension is use of spatial distances for geographic analyses and correlation analyses.

# References

Mantel, N. 1967.

The detection of disease clustering and a generalized regression approach. *Cancer Research* 27: 209-220.

Legendre, P., and M-J. Fortin. 1989.

Spatial pattern and ecological analysis. *Vegetatio* 80: 107-138.