

# A (brief) introduction to ordination and the vegan package

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Datasets  
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Unconstrained Ordination  
ooooooooooooooooooooooo

Constrained Ordination  
oooooooooooo

Permutation Tests  
o

End

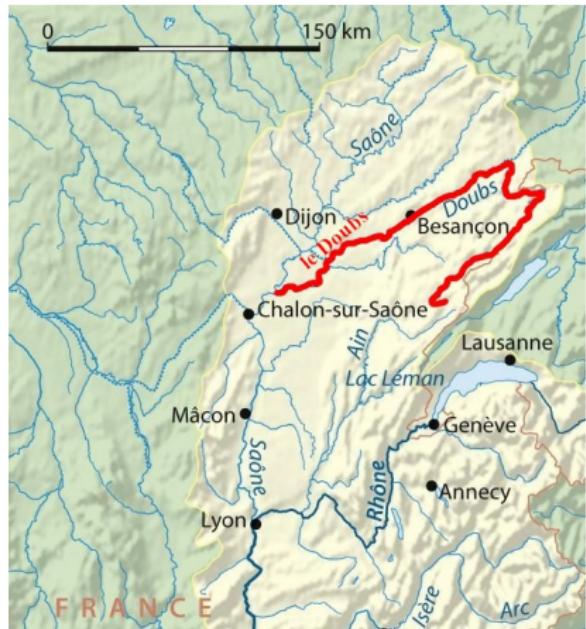
	Raw data	Transformed data	Unimodal	Distance-based	Model-based
Unconstrained	PCA		CA, DCA	PCoA, NMDS	MM, LVM
Constrained	RDA	tb-PCA tb-RDA	CCA	db-RDA	CAO, CQO
Other				Permanova, Dispersion	manyglm

(Nearly) no maths today ;)

# Datasets

# Demonstration: Doubs river fish communities

5 / 46



- ▶ Fish
- ▶ 30 sites along the Doubs River

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Verneaux, J. (1973) Cours d'eau de Franche-Comté (Massif du Jura). Recherches écologiques sur le réseau hydrographique du Doubs. Essai de biotypologie. These d'état, Besançon. 1–257.

Datasets



Unconstrained Ordination



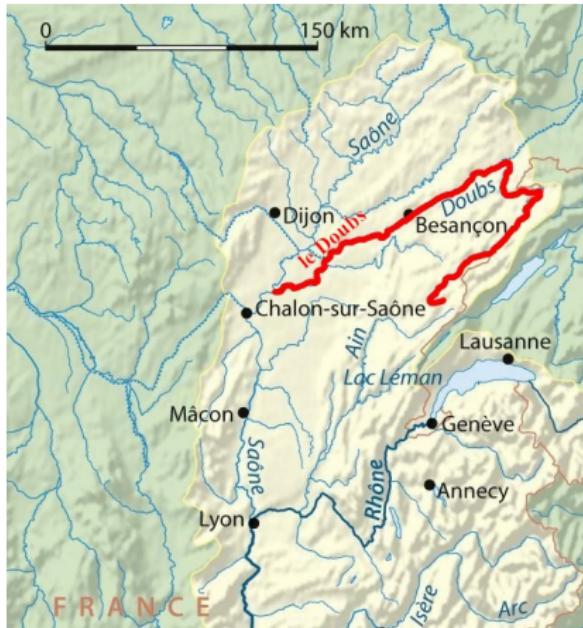
Constrained Ordination



Permutation Tests



End



- ▶ Fish
  - ▶ 30 sites along the Doubs River

## Questions

- ▶ How does fish composition change downstream?
  - ▶ Environmental drivers?

Verneaux, J. (1973) Cours d'eau de Franche-Comte (Massif du Jura). Recherches ecologiques sur le reseau hydrographique du Doubs. Essai de biotypologie. These d'etat, Besancon. 1-257.

```
Dabu <- read.table('doubsAbu.csv', sep = ',', header = TRUE)
Denv <- read.table('doubsEnv.csv', sep = ',', header = TRUE)
Dspa <- read.table('doubsSpa.csv', sep = ',', header = TRUE)
```

**dim(Dabu)**

[1] 30 27

30 sites, 27 taxa

```
head(Dabu[, 1:18])
```

	CHA	TRU	VAI	LOC	OMB	BLA	HOT	TOX	VAN	CHE	BAR	SPI	GOU	BRO	PER	BOU	PSO	ROT
1	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
2	0	5	4	3	0	0	0	0	0	0	0	0	0	0	0	0	0	
3	0	5	5	5	0	0	0	0	0	0	0	0	0	1	0	0	0	
4	0	4	5	5	0	0	0	0	0	1	0	0	1	2	2	0	0	
5	0	2	3	2	0	0	0	0	5	2	0	0	2	4	4	0	0	
6	0	3	4	5	0	0	0	0	1	2	0	0	1	1	1	0	0	

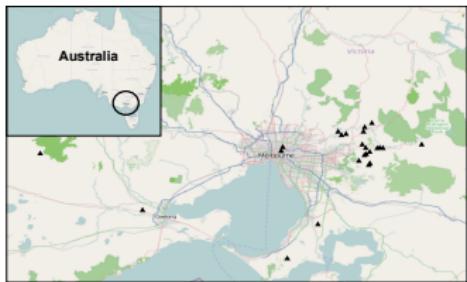
```
# Dimension and first rows of Environmental data  
dim(Denv)
```

[1] 30 11

30 sites, 11 variables

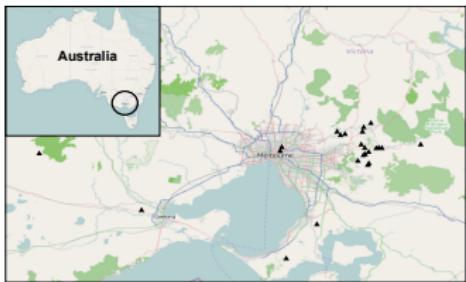
**head(Denv)**

	das	alt	pen	deb	pH	dur	pho	nit	amm	oxy	dbo
1	0.3	934	48.0	0.84	7.9	45	0.01	0.20	0.00	12.2	2.7
2	2.2	932	3.0	1.00	8.0	40	0.02	0.20	0.10	10.3	1.9
3	10.2	914	3.7	1.80	8.3	52	0.05	0.22	0.05	10.5	3.5
4	18.5	854	3.2	2.53	8.0	72	0.10	0.21	0.00	11.0	1.3
5	21.5	849	2.3	2.64	8.1	84	0.38	0.52	0.20	8.0	6.2
6	32.4	846	3.2	2.86	7.9	60	0.20	0.15	0.00	10.2	5.3



- ▶ Macroinvertebrates
  - ▶ 24 sites
  - ▶ covering a salinity and toxicity gradient

The dataset is published in: Szöcs, E., Kefford, B.J., Schäfer, R.B., 2012. Is there an interaction of the effects of salinity and pesticides on the community structure of macroinvertebrates? *Science of the Total Environment* 437, 121–126.



- ▶ Macroinvertebrates
  - ▶ 24 sites
  - ▶ covering a salinity and toxicity gradient

## Questions:

- ▶ Interaction between salinization and pesticides?
  - ▶ Which species are affected?
  - ▶ Other influences?

The dataset is published in: Szöcs, E., Kefford, B.J., Schäfer, R.B., 2012. Is there an interaction of the effects of salinity and pesticides on the community structure of macroinvertebrates? *Science of the Total Environment* 437, 121–126.

## Exercise: Salinization and Pesticides

```
# setwd('3-Ordination/data/')
abu <- read.table('melbourneAbu.csv', sep = ';', header = TRUE)
env <- read.table('melbourneEnv.csv', sep = ';', header = TRUE)
```

```
# dimensions of data.frame  
dim(env)
```

[1] 24 23

**dim(abu)**

[1] 24 76

24 sites, 22 environmental variables, 75 taxa

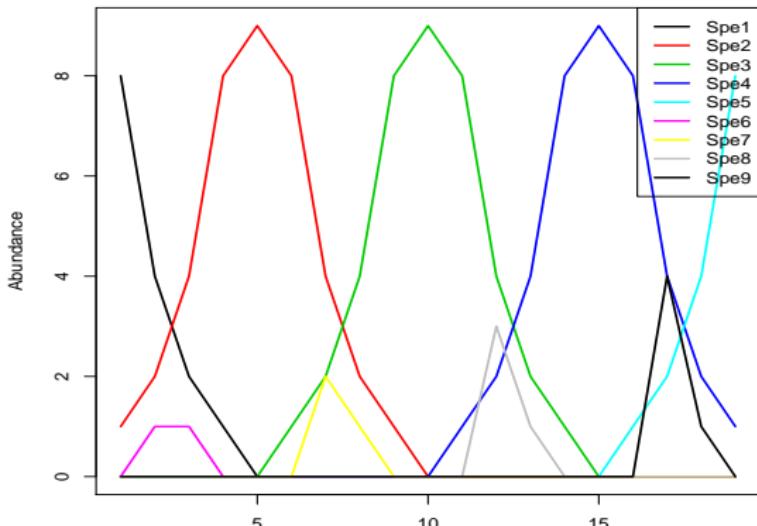
```
head(env[ , 1:10])
```

ID	T	pH	oxygen	Depth	maxwidth	minwidth	rifperc	poolperc	Bedrock	
1	1-11	16.8	7.67	80.1	0.9	15	12.0	0	100	0
2	2-11	16.5	7.29	83.0	0.9	30	15.0	0	100	0
3	3-11	17.3	7.20	77.9	0.4	4	2.5	0	100	0
4	4-11	15.6	7.84	72.0	0.7	8	2.5	0	100	0
5	5-11	17.2	6.97	69.9	0.9	7	4.0	0	100	0
6	6-11	15.5	7.26	80.0	0.2	3	2.0	5	95	0

# Exercise: Dummy abundances

10 / 46

```
# Load dummy data
dummy <- read.table('dummydata.csv', header = TRUE, sep = ';')
# plot dummy data
matplot(dummy[ , -1], type = 'l', xlab = 'Site', ylab = 'Abundance',
        lty = 'solid', lwd = 2, col = 1:9)
legend('topright', legend = colnames(dummy)[-1],
       col = 1:9, lty = 'solid', lwd = 2)
```



Datasets

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Unconstrained Ordination

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Constrained Ordination

ooooooooooooooo

Permutation Tests

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End

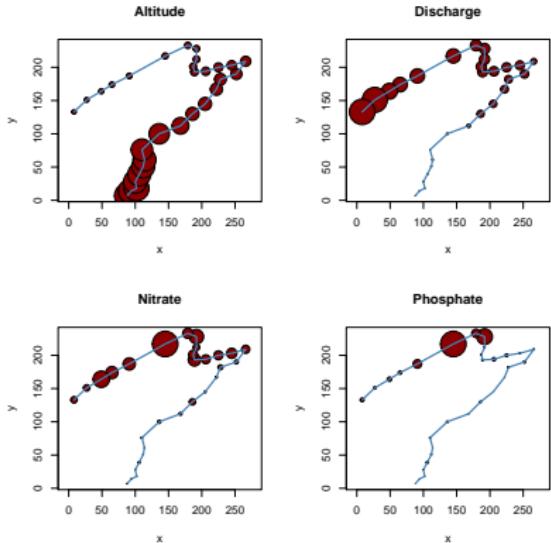
# Unconstrained Ordination

Principal Components Analysis (PCA)

Principal coordinates analysis (PCoA)

Nonmetric Multidimensional Scaling (NMDS)

## Principal Components Analysis (PCA) — Why?

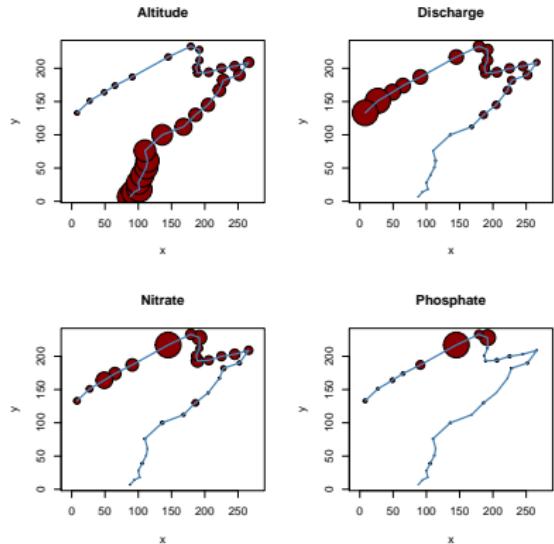


- ## ► 11 variables

## Questions:

- ▶ Which variables are correlated?
  - ▶ Which sites have similar conditions?
  - ▶ How do conditions change downstream?

## Solutions?



- ▶ 11 variables

## Questions:

- ▶ Which variables are correlated?
- ▶ Which sites have similar conditions?
- ▶ How do conditions change downstream?

## Solutions?

- ▶ pairwise comparisons
- ▶ 3D possible
- ▶ more than 3 dimensions?

- ▶ *"Look from another angle on the data"*
- ▶ PCA is just a rotation of the coordinate system
- ▶ The rotation is done so that the first axis contains as much variation as possible
- ▶ Second axis than most of remaining variation

Short Demo.

- ▶ *"Look from another angle on the data"*
- ▶ PCA is just a rotation of the coordinate system
- ▶ The rotation is done so that the first axis contains as much variation as possible
- ▶ Second axis than most of remaining variation

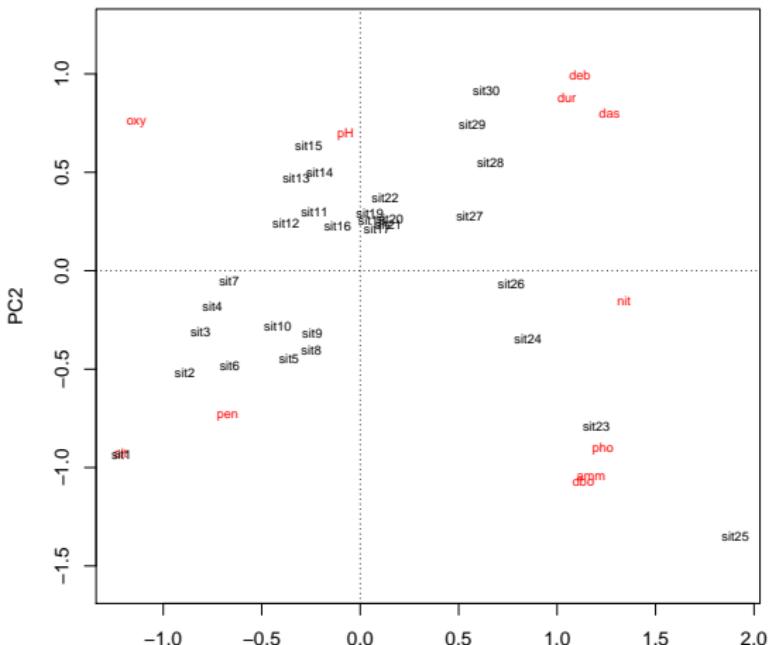
## Short Demo.

Maths:

- ▶ The covariance (or correlation) matrix is decomposed into its Eigenvectors and Eigenvalues.
- ▶ The Eigenvectors give the rotation needed
- ▶ The Eigenvalues stretch the axes

## Principal Components Analysis (PCA) — How?

```
require(vegan)  
PCA <- rda(Denv, scale = TRUE)  
plot(PCA, scaling = 3)
```



## Datasets

## Unconstrained Ordination

#### Unconstrained Orientation

Constrained Ordination

PCP

## Permutation Tests

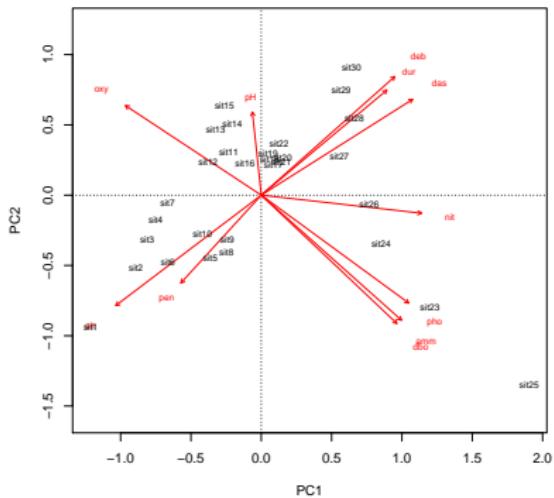
1

End

# Principal Components Analysis (PCA) — Interpretation? (I)

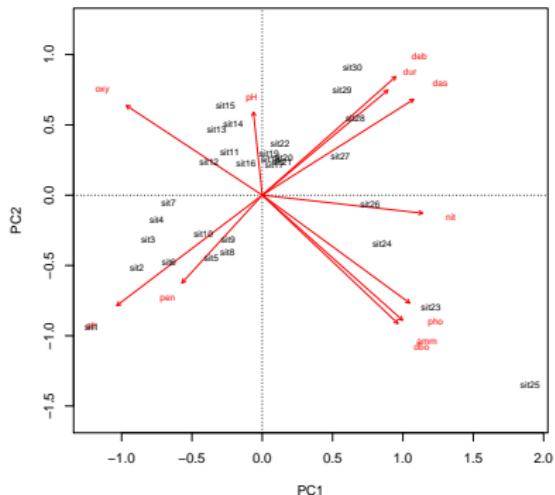
15 / 46

```
biplot(PCA, cex = 5, scaling = 3)
```

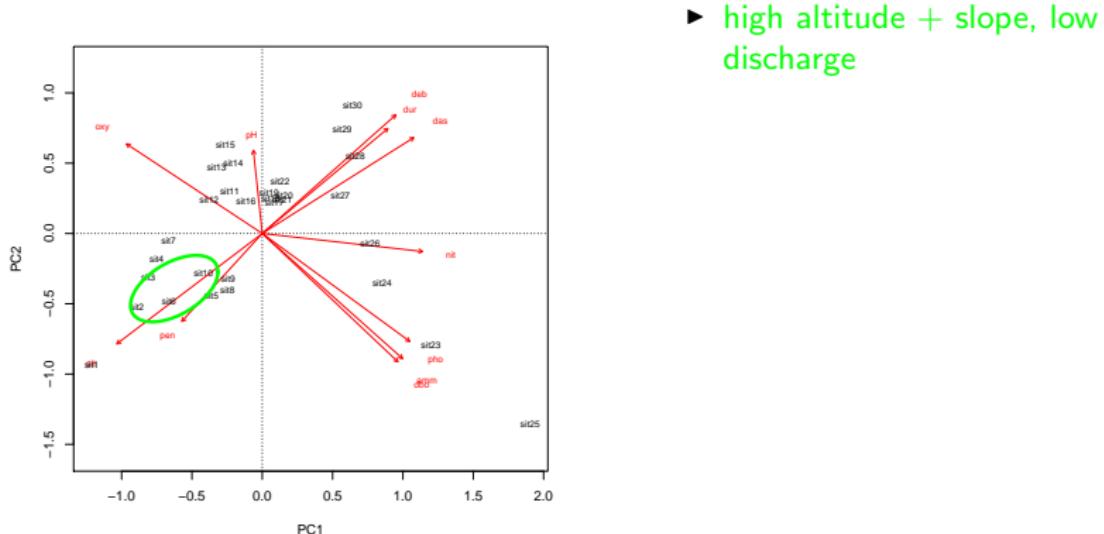


- ▶ angle between variables **approx.** their correlation
- ▶ distance between sites **approx.** their euclidean distance
- ▶ projecting a site on a variable **approx.** the relative value

```
biplot(PCA, cex = 5, scaling = 3)
```

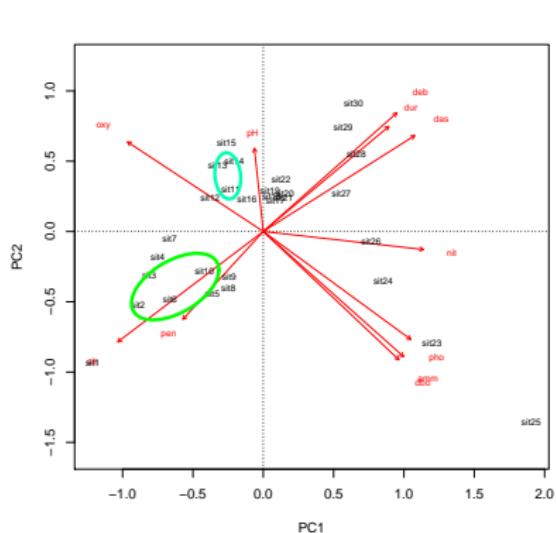


- ▶ angle between variables **approx.** their correlation
  - ▶ distance between sites **approx.** their euclidean distance
  - ▶ projecting a site on a variable **approx.** the relative value
- 
- ▶ scaling = 1 - to interpret (only) distances between sites
  - ▶ scaling = 2 - to interpret (only) correlations between variables



## Principal Components Analysis (PCA) — Interpretation? (II)

16 / 46



- ▶ high altitude + slope, low discharge
  - ▶ high oxygen, low nutrient

## Datasets

## Unconstrained Ordination

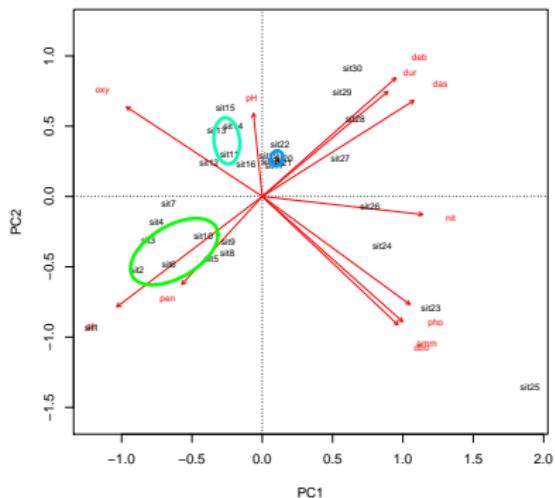
## Constrained Ordination

## Permutation Tests

End

## Principal Components Analysis (PCA) — Interpretation? (II)

16 / 46



- ▶ high altitude + slope, low discharge
  - ▶ high oxygen, low nutrient
  - ▶ intermediate

## Datasets

## Unconstrained Ordination

## Constrained Ordination

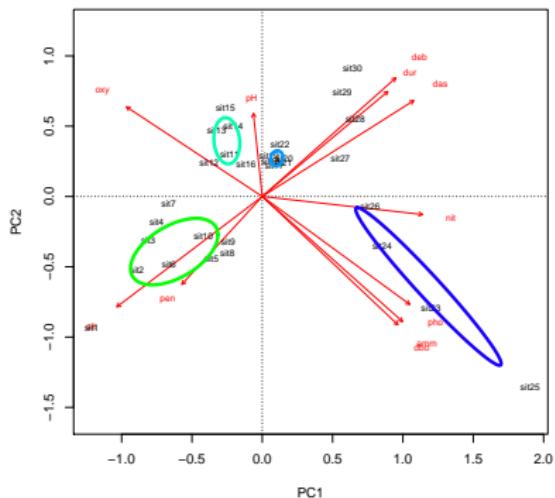


## Permutation Tests

End

## Principal Components Analysis (PCA) — Interpretation? (II)

16 / 46



- ▶ high altitude + slope, low discharge
  - ▶ high oxygen, low nutrient
  - ▶ intermediate
  - ▶ nutrient rich

## Datasets



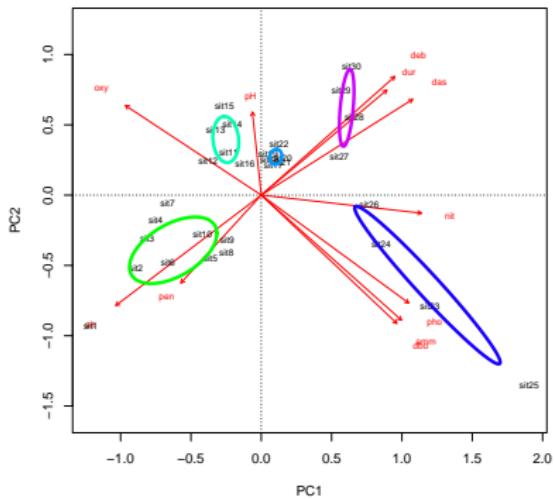
## Unconstrained Ordination

## Constrained Ordination



## Permutation Tests

End



- ▶ high altitude + slope, low discharge
  - ▶ high oxygen, low nutrient
  - ▶ intermediate
  - ▶ nutrient rich
  - ▶ high discharge, low altitude, medium nutrient

## Datasets

## Unconstrained Ordination

## Constrained Ordination

oooooooooooo

## Permutation Tests

End

# Principal Components Analysis (PCA) — Interpretation? (III)

```
summary(PCA, display = NULL, scaling = 3)
```

Call:

```
rda(X = Denv, scale = TRUE)
```

Partitioning of correlations:

	Inertia	Proportion
Total	11	1
Unconstrained	11	1

Eigenvalues, and their contribution to the correlations

Importance of components:

	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Eigenvalue	5.9687	2.1638	1.06516	0.73873	0.40027	0.33565	0.1727
Proportion Explained	0.5426	0.1967	0.09683	0.06716	0.03639	0.03051	0.0157
Cumulative Proportion	0.5426	0.7393	0.83616	0.90331	0.93970	0.97022	0.9859
	PC8	PC9	PC10	PC11			
Eigenvalue	0.10821	0.02368	0.01707	0.005993			
Proportion Explained	0.00984	0.00215	0.00155	0.000540			
Cumulative Proportion	0.99575	0.99790	0.99946	1.000000			

Scaling 3 for species and site scores

- \* Both sites and species are scaled proportional to eigenvalues on all dimensions
- \* General scaling constant of scores:

# Your turn!

Load the Melbourne dataset (only environmental variables).

**Exclude** the variables ID, logCond and logmaxTU.

Perform a PCA.

Which variables are correlated?

How much variance is explained by the first 2 axes?

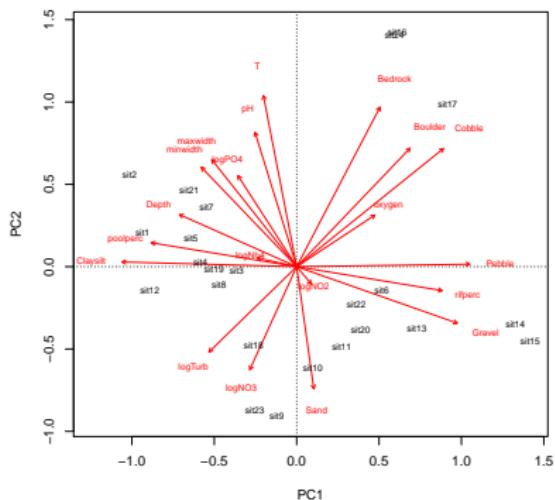
How could the two PCA axes be interpreted?

# Exercise

```
take <- env[ , !names(env) %in% c('ID', 'logCond', 'logmaxTU')]
PCA <- rda(take, scale = TRUE)
cumsum(PCA$CA$eig / PCA$tot.chi)[1:2]
```

PC1	PC2
0.2839873	0.4537452

```
biplot(PCA, scaling = 3)
```



- ▶ multiple variables interrelated
- ▶ 1st axis can be interpreted as *hydrological gradient*
- ▶ 2nd axis can be interpreted as *chemistry gradient*

## Question:

- ▶ How is diversity related to salinity, pesticides and other variables?

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- ▶ How is diversity related to salinity, pesticides and other variables?

## Problem:

- ▶ Only 24 sites
  - ▶ but 22 (potentially correlated) explanatory variables
  - ▶ strong hypotheses about salinity and pesticides

## A Solution:

## Question:

- ▶ How is diversity related to salinity, pesticides and other variables?

## Problem:

- ▶ Only 24 sites
- ▶ but 22 (potentially correlated) explanatory variables
- ▶ strong hypotheses about salinity and pesticides

## A Solution:

- ▶ Reduce number of variables to *Principal Components*
- ▶ regress these

## Excursus — principal component regression (PCR)

```
# calculate shannon diversity index
div <- diversity(abu[, -1], index = 'shannon')
pc <- scores(PCA, choices = c(1, 2), scaling = 1, display = 'sites')
model_data <- data.frame(div, pc, logCond = env$logCond, logmaxTU = env$logmaxTU)
model <- lm(div ~ PC1 + PC2 + logCond + logmaxTU, data = model_data)
summary(model)
```

Call:

```
lm(formula = div ~ PC1 + PC2 + logCond + logmaxTU, data = model_data)
```

Residuals:

Min	1Q	Median	3Q	Max
-0.64415	-0.15688	0.02063	0.18219	0.57929

Coefficients:

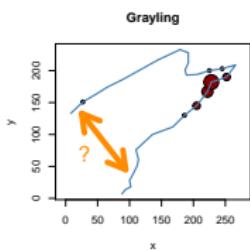
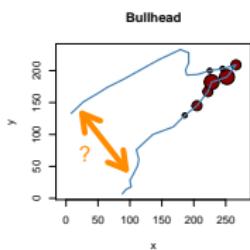
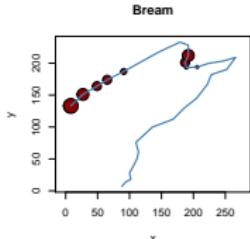
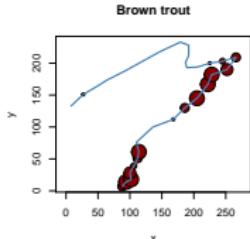
	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	1.83079	0.43429	4.216	0.000468 ***
PC1	0.01971	0.16691	0.118	0.907262
PC2	0.02192	0.19570	0.112	0.911996
logCond	-0.20942	0.13050	-1.605	0.125049
logmaxTU	-0.12572	0.07316	-1.718	0.101994
---				

Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 0.3645 on 19 degrees of freedom

Multiple R-squared: 0.2682, Adjusted R-squared: 0.1141

F-statistic: 1.741 on 4 and 19 DF, p-value: 0.1827



- ▶ Species may be absent due to different factors (too high flow, too saline, etc.)
  - ▶ *Absence* contains less information than *Presence*
  - ▶ PCA preserves the euclidean distance between sites
  - ▶ Need another measure of similarity for (raw) abundances

## Dissimilarity measures — Species abundance paradox

	Spe1	Spe2	Spe3
sit1	0	4	8
sit2	0	1	1
sit3	1	0	0

```
vegdist(mat, method = 'euclidean')

      sit1      sit2
sit2 7.615773
sit3 9.000000 1.732051
```

```
vegdist(mat, method = 'bray')

      sit1      sit2
sit2 0.7142857
sit3 1.0000000 1.0000000
```

---

from: Anderson, M.J., Crist, T.O., et al. , 2011. Navigating the multiple meanings of beta diversity: a roadmap for the practicing ecologist. *Ecology Letters* 14, 19–28.

## Dissimilarity measures — Species abundance paradox

	Spe1	Spe2	Spe3
sit1	0	4	8
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```

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          sit1      sit2
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```

```
vegdist(mat, method = 'bray')
```

```
          sit1      sit2
sit2 0.7142857
sit3 1.0000000 1.0000000
```

Abundances included  
(or cover, biomass, etc.)

Exclude joint absences

Include joint absences

Chi-squared  
Hellinger, Chord  
Euclidean on proportions  
Bray-Curtis, fourth root  
Bray-Curtis, square root  
Bray-Curtis, raw data  
Modified Gower, log10  
Modified Gower, log5  
Modified Gower, log2

Euclidean, log(y+1)  
Euclidean, raw data

*Emphasis:*

Rare species  
Proportions

Composition +  
relative abund.

Composition  
log abundance

Log abundance  
Raw abundance

from: Anderson, M.J., Crist, T.O., et al. , 2011. Navigating the multiple meanings of beta diversity: a roadmap for the practicing ecologist. Ecology Letters 14, 19–28.

- ▶ Works on distance matrices
- ▶ Species can be added as *weighted averages*
- ▶ Eigenvalue based
- ▶ PCoA with euclidean distance == PCA

# Principal coordinates analysis (PCoA)

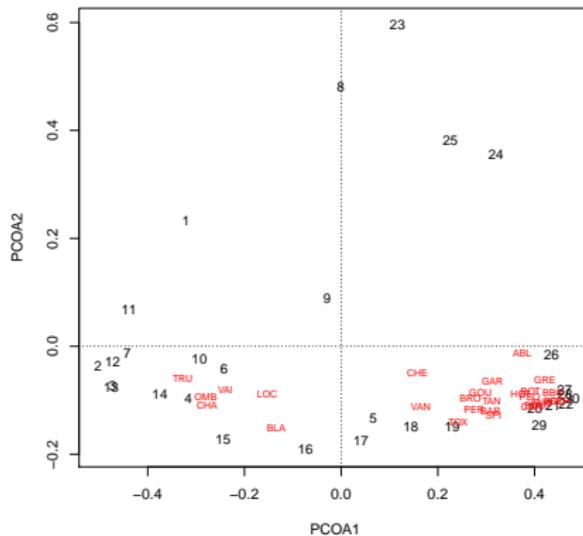
25 / 46

```
# Distance matrix
Dabu_dist <- vegdist(Dabu, method = 'bray')

# PCoA
PCOA <- cmdscale(Dabu_dist, eig = TRUE)

# Create plot
plot(PCOA$points, type = 'n',
      xlab = 'PCOA1', ylab = 'PCOA2')
text(PCOA$points,
     labels = rownames(Dabu), cex = 0.9)
abline(h = 0, lty = 'dotted')
abline(v = 0, lty = 'dotted')
# Add species as weighted averages
wa <- wascores(PCOA$points, Dabu)
text(wa, labels = colnames(Dabu),
     col = 'red', cex = 0.7)
```

```
# explained variance
(PCOA$eig / sum(PCOA$eig))[1:2] * 100
[1] 49.24914 15.95758
```



- ▶ Similar to PCoA
  - ▶ Does not preserve exact distances between objects
  - ▶ Possibly better representation in low dimensions
  - ▶ **Not** eigenvalue based, iterative algorithm
  - ▶ Axes have no meaning, just the relative distances

## Datasets

## Unconstrained Ordination

## Constrained Ordination

## Permutation Tests

End

# Nonmetric Multidimensional Scaling (NMDS)

27 / 46

```
# Distance matrix
Dabu_0 <- Dabu[!rowSums(Dabu) == 0, ]
Dabu_dist <- vegdist(Dabu_0, method = 'bray')

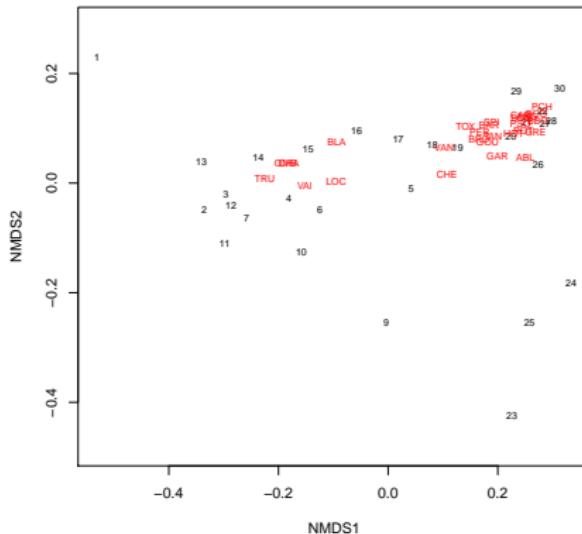
# NMDS
NMDS <- metaMDS(Dabu_dist, k = 2, trace = 0)

# Plot
plot(NMDS, type = 't')

# Add species as weighted averages
wa <- wascores(NMDS$points, Dabu_0)
text(wa, labels = colnames(Dabu),
     col = 'red', cex = 0.7)
```

```
# Stress value
NMDS$stress

[1] 0.07429467
```



# Your turn!

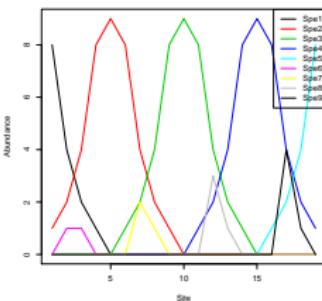
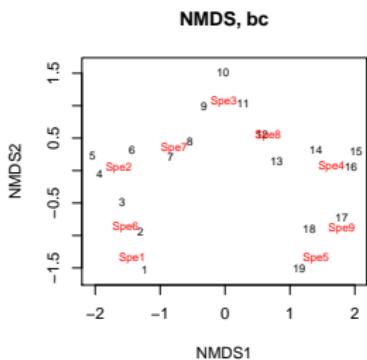
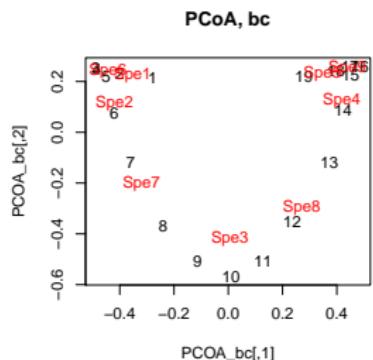
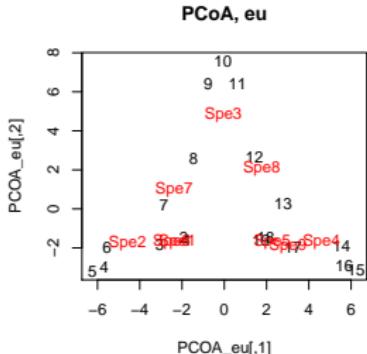
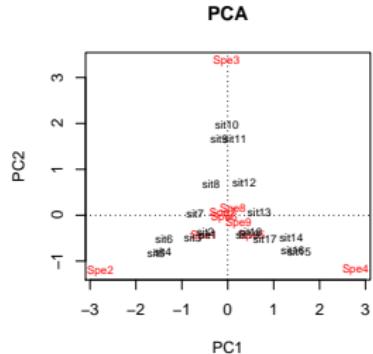
Using the artificial dummy dataset.

Run:

1. PCA
2. PCoA with euclidean distance
3. PCoA with Bray-Curtis dissimilarity
4. NMDS with Bray-Curtis dissimilarity

What are the differences between ordinations?  
Which represent better the underlying gradient?

## Exercise



## Datasets

oooooo

## Unconstrained Ordination

## Constrained Ordination



## Permutation Tests

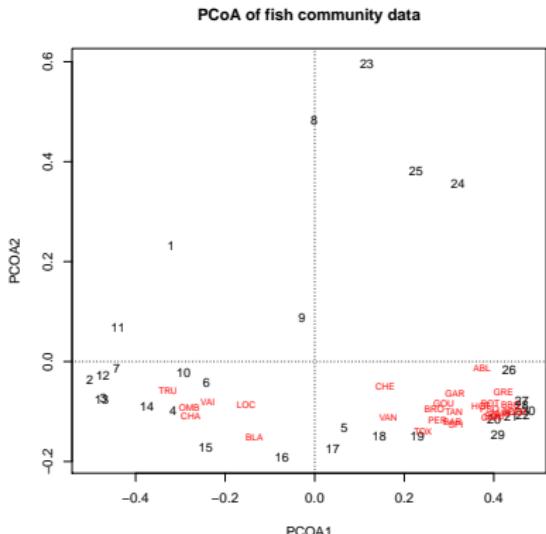
End

- ▶ This ordination is **only** driven by fish community data

## Question:

- ▶ How can we interpret the gradients in community composition?

A solution:



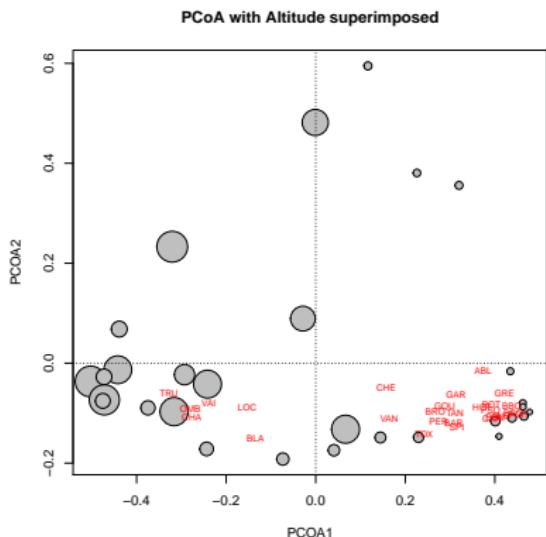
- This ordination is **only** driven by fish community data

### Question:

- How can we interpret the gradients in community composition?

### A solution:

- Superimpose environmental variables

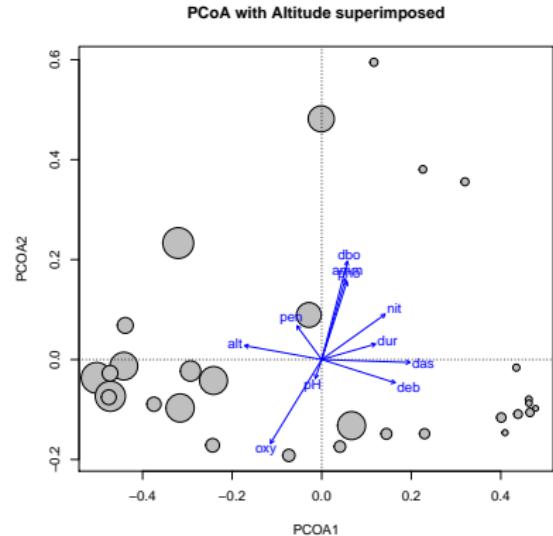


# Fit environmental variables to ordination (II)

```
# PCoA of fish community data
plot(PCOA$points,
      xlab = 'PCoA1', ylab = 'PCoA2',
      cex = 5*Denv$alt / max(Denv$alt),
      main = 'PCoA with Altitude',
      bg = 'grey75', pch = 21)
abline(h = 0 , lty = 'dotted')
abline(v = 0 , lty = 'dotted')

# Fit Altitude to site-scores
ef <- envfit(PCOA, Denv)
plot(ef)
ef # summary

# Fit GAM
ordisurf(PCOA, Denv$alt, add = TRUE)
```



- ▶ Post hoc method
- ▶ non-linearity?

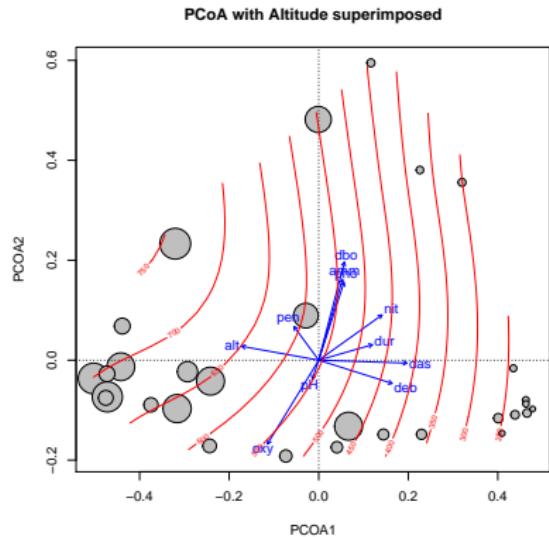
# Fit environmental variables to ordination (II)

31 / 46

```
# PCoA of fish community data
plot(PCOA$points,
     xlab = 'PCOA1', ylab = 'PCOA2',
     cex = 5*Denv$alt / max(Denv$alt),
     main = 'PCoA with Altitude',
     bg = 'grey75', pch = 21)
abline(h = 0 , lty = 'dotted')
abline(v = 0 , lty = 'dotted')

# Fit Altitude to site-scores
ef <- envfit(PCOA, Denv)
plot(ef)
ef # summary

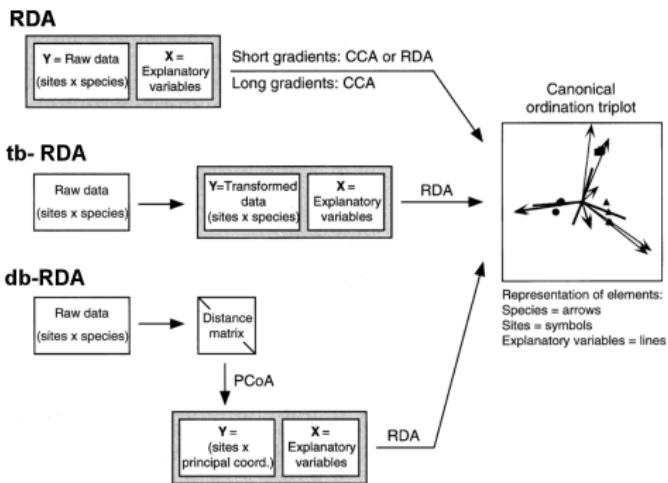
# Fit GAM
ordisurf(PCOA, Denv$alt, add = TRUE)
```



- ▶ Post hoc method
- ▶ non-linearity?
- ▶ be careful with `summary`
- ▶ Constrained ordination a better alternative

# Constrained Ordination

- ▶ Redundancy analysis (RDA)
- ▶ Transformation-based RDA (tb-RDA)
- ▶ Distance-based RDA (db-RDA)



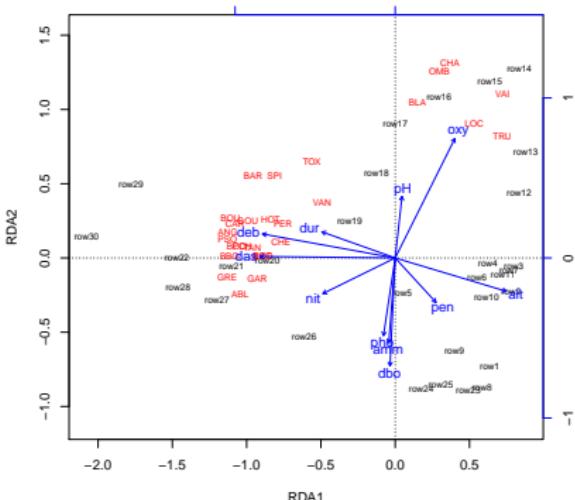
Adapted from: Legendre, P., Gallagher, E.D., 2001. Ecologically meaningful transformations for ordination of species data. *Oecologia* 129, 271–280.

- ▶ Associates both environmental and community data at once
- ▶ Combination of regression and PCA:
  1. Regress explanatory variables on community data
  2. Run PCA on fitted values
- ▶ Can test hypothesis about relationships

```
RDA <- rda(Dabu ~ ., data = Denv,
             scale = TRUE)
plot(RDA, scaling = 3)
```

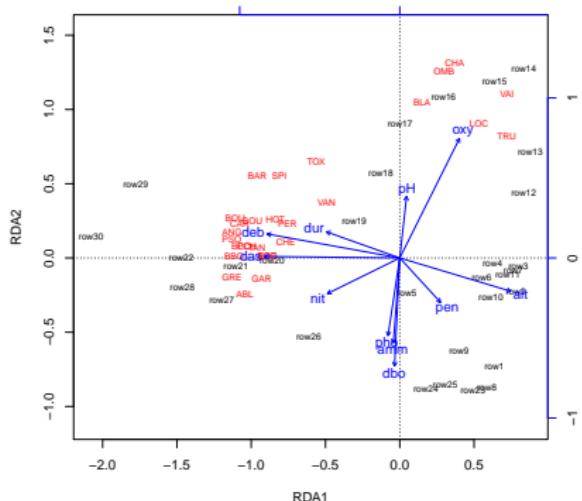
## ► Formula interface

- Left side: Response matrix
- Right side: Response variables from Denv



# Redundancy analysis (RDA) — Interpretation? (I)

36 / 46



- ▶ Similar to PCA
- ▶ Projecting a site on response or explanatory variable **approx.** the value
- ▶ angles between response and expl. variables **approx.** their correlations

```
...
```

Partitioning of correlations:

	Inertia	Proportion
Total	27.000	1.0000
Constrained	20.177	0.7473
Unconstrained	6.823	0.2527

Eigenvalues, and their contribution to the correlations

Importance of components:

	RDA1	RDA2	RDA3	RDA4	RDA5	RDA6	RDA7
Eigenvalue	14.714	2.6433	1.1341	0.76821	0.33807	0.28135	0.09356
Proportion Explained	0.545	0.0979	0.0420	0.02845	0.01252	0.01042	0.00347
Cumulative Proportion	0.545	0.6429	0.6849	0.71333	0.72585	0.73627	0.73974
	RDA8	RDA9	RDA10	RDA11	PC1	PC2	
Eigenvalue	0.08411	0.07592	0.02314	0.02129	2.44703	1.4094	
Proportion Explained	0.00312	0.00281	0.00086	0.00079	0.09063	0.0522	
Cumulative Proportion	0.74285	0.74566	0.74652	0.74731	0.83794	0.8901	

```
...
```

- RDA (as PCA) preserves the euclidean distance.

What about the species abundance paradox?

---

Legendre, P., Gallagher, E.D., 2001. Ecologically meaningful transformations for ordination of species data. *Oecologia* 129, 271–280.

Datasets  
oooooooo

Unconstrained Ordination  
ooooooooooooooooooooooo

Constrained Ordination  
oooooooo●oooooo

Permutation Tests  
○

End

- ▶ RDA (as PCA) preserves the euclidean distance.

What about the species abundance paradox?

- ▶ Can transform data to use with euclidean distance
  - ▶ Remove differences in total abundance, while keeping the variations relative abundance
- ▶ Chord and Hellinger transformations useful.

Hellinger:

$$y'_{ij} = \sqrt{\frac{y_{ij}}{y_{i+}}}$$

```
mat
  Spe1  Spe2  Spe3
sit1    0     4     8
sit2    0     1     1
sit3    1     0     0

decostand(mat, 'hellinger')
  Spe1      Spe2      Spe3
sit1  0 0.5773503 0.8164966
sit2  0 0.7071068 0.7071068
sit3  1 0.0000000 0.0000000
attr(,"decostand")
[1] "hellinger"
```

---

Legendre, P., Gallagher, E.D., 2001. Ecologically meaningful transformations for ordination of species data. Oecologia 129, 271–280.

## transformation-based RDA

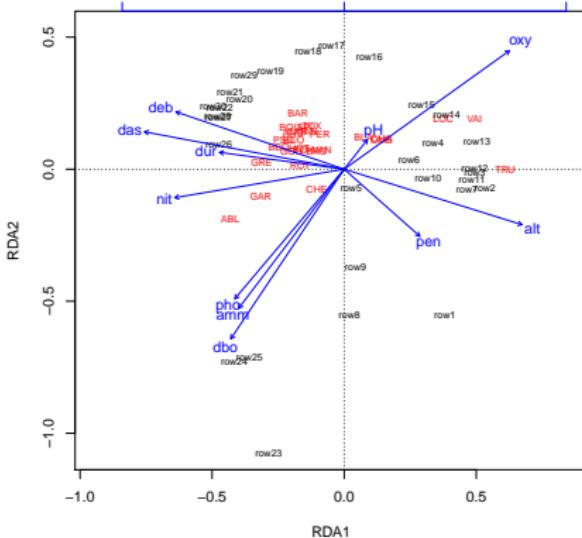
39 / 46

```

# Hellinger transformation
Dabu_h <- decostand(Dabu, 'hellinger')
# RDA on Hellinger transformed abundances
tbRDA <- rda(Dabu_h ~ ., data = Denv)

# Plot
plot(tbRDA, type = 't')

```



## Datasets



## Unconstrained Ordination

## Constrained Ordination

## Permutation Tests

End

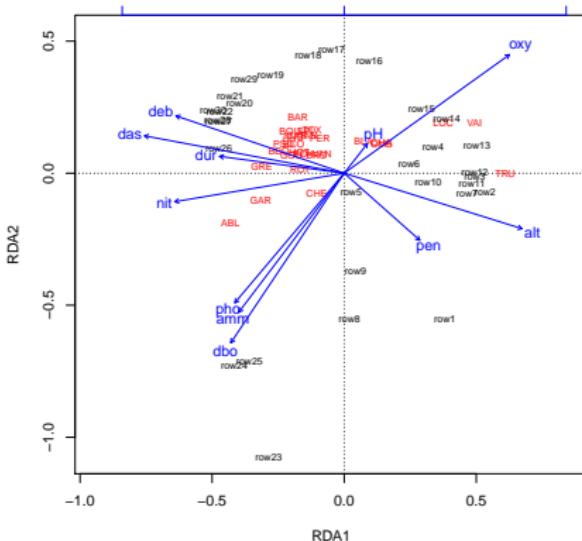
```

# Hellinger transformation
Dabu_h <- decostand(Dabu, 'hellinger')
# RDA on Hellinger transformed abundances
tbRDA <- rda(Dabu_h ~ ., data = Denv)

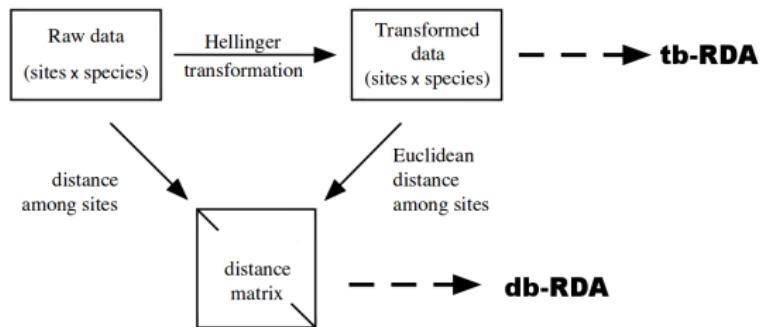
# Plot
plot(tbRDA, type = 't')

```

- ▶ alt, oxy and nutrients important
  - ▶ Trout (TRU) and minnow (VAI) found at high sites with high oxygen
  - ▶ Bleak (ABL) is found a low oxygen and high nutrients
  - ▶ Other species in similar environments



- db-RDA is a related method
- hellinger transformation can also be expressed as distance matrix
- *Constrained PCoA*
- Can use every distance metric



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modified from: Legendre, P., Gallagher, E.D., 2001. Ecologically meaningful transformations for ordination of species data. *Oecologia* 129, 271–280.

# distance-based RDA

41 / 46

```
# dbRDA  
dbRDA <- capscale(Dabu ~ ., data = Denv,  
                     distance = 'bray')  
plot(dbRDA, type = 't')
```

```
summary(dbRDA)
```

```
...
```

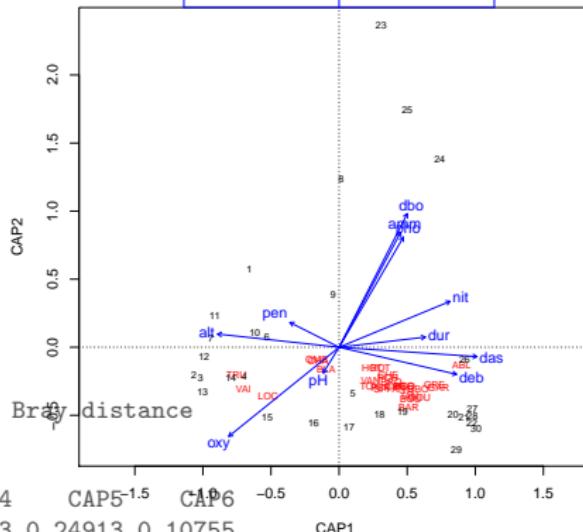
	Inertia	Proportion
Total	7.802	1.0000
Constrained	5.715	0.7325
Unconstrained	2.087	0.2675

Eigenvalues, and their contribution to the squared Bray distance

Importance of components:

	CAP1	CAP2	CAP3	CAP4	CAP5	CAP6
Eigenvalue	3.2627	1.0283	0.53633	0.37863	0.24913	0.10755
Proportion Explained	0.4182	0.1318	0.06874	0.04853	0.03193	0.01379
Cumulative Proportion	0.4182	0.5500	0.61875	0.66729	0.69922	0.71300

```
...
```



# Your turn!

Using the artificial dummy dataset and Site as only constraining variable

Run:

1. RDA
2. tbRDA (Hellinger)
3. dbRDA with Bray-Curtis
4. dbRDA with  $x^{0.25}$  transformed abundances and Bray-Curtis

What ordination presents best the gradient?

What method explains most of variance?

See Demo.

## Datasets

oooooooo

## Unconstrained Ordination

Constrained Ordination

## Permutation Tests

End

# Permutation Tests

# Your turn!

Using the melbourne data.

- ▶ Distance-based hypothesis testing ((PER-)MANOVA, SIMPER, ANOSIM)
- ▶ Dispersion measures ( $\beta$ -Diversity, Functional diversity)
- ▶ Consensus RDA, RLQ (traits), ...
- ▶ Model-based ordination / hypothesis testing (See work of David Warton et al.)