Constrained ordination

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Welcome

Today's topics

- Constrained ordination
 - Canonical Correspondence Analysis
 - Redundancy Analysis
 - Partial constrained ordination
- Model Building
 - Model selection
- Permutation tests

Constrained Ordination

CCA

Canonical Correspondence Analysis

CCA is the constrained form of CA; fitted using cca()

Two interfaces for specifying models

- basic; cca1 ← cca(X = varespec, Y = varechem)
- formula; cca1 ← cca(varespec ~ ., data = varechem)

RDA is the constrained form of PCA; fitted using rda()

Formula interface is the more powerful — recommended

Canonical Correspondence Analysis

```
cca1 ← cca(varespec ~ ., data = varechem)
cca1
```

```
## Call: cca(formula = varespec ~ N + P + K + Ca + Mg + S + Al + Fe + Mn +
## Zn + Mo + Baresoil + Humdepth + pH, data = varechem)
###
              Inertia Proportion Rank
           2.0832
## Total
                          1.0000
## Constrained 1.4415 0.6920 14
## Unconstrained 0.6417 0.3080
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
           CCA2
                  CCA3
                         CCA4 CCA5
                                      CCA6
    CCA1
                                             CCA7
                                                    CCA8
                                                           CCA9
                                                                CCA10
                                                                       CCA11
## 0.4389 0.2918 0.1628 0.1421 0.1180 0.0890 0.0703 0.0584 0.0311 0.0133 0.0084
   CCA12 CCA13 CCA14
## 0.0065 0.0062 0.0047
## Eigenvalues for unconstrained axes:
      CA1
              CA2
                      CA3
                              CA4
                                     CA5
                                             CA6
                                                             CA8
##
                                                     CA7
                                                                    CA9
## 0.19776 0.14193 0.10117 0.07079 0.05330 0.03330 0.01887 0.01510 0.00949
```

Redundancy Analysis

```
rda1 ← rda(varespec ~ ., data = varechem)
rda1
```

```
## Call: rda(formula = varespec ~ N + P + K + Ca + Mg + S + Al + Fe + Mn +
## Zn + Mo + Baresoil + Humdepth + pH, data = varechem)
##
                 Inertia Proportion Rank
## Total
           1825,6594
                             1,0000
## Constrained 1459.8891
                         0.7997
                                    14
## Unconstrained 365.7704
                             0.2003
## Inertia is variance
## Eigenvalues for constrained axes:
         RDA2 RDA3
                    RDA4 RDA5 RDA6
                                     RDA7
                                           RDA8
                                                 RDA9 RDA10 RDA11 RDA12 RDA13
   RDA1
## 820.1 399.3 102.6 47.6 26.8 24.0 19.1 10.2
                                                4.4 2.3 1.5
                                                                  0.9
## RDA14
    0.3
## Eigenvalues for unconstrained axes:
     PC1
            PC2
                   PC3
                         PC4
                                PC5
                                       PC6
                                             PC7
                                                          PC9
###
                                                    PC8
## 186.19 88.46 38.19 18.40 12.84 10.55
                                            5.52
                                                   4.52
                                                         1.09
```

The cca.object

- Objects of class "cca" are complex with many components
- Entire class described in ?cca.object
- Depending on what analysis performed some components may be NULL
- Used for (C)CA, PCA, RDA, CAP (capscale()), and dbRDA (dbrda())

The cca.object

cca1 has a large number of components

- \$call how the function was called
- \$grand.total in (C)CA sum of rowsum
- \$rowsum the row sums
- \$colsum the column sums
- \$tot.chi total inertia, sum of Eigenvalues
- \$pCCA Conditioned (partial-ed out) components
- \$CCA Constrained components
- \$CA Unconstrained components
- \$method Ordination method used
- \$inertia Description of what inertia is

The cca.object

Depending on how one called cca() etc some of these components will be NULL

\$pCCA is only filled in if a partial constrained ordination fitted

rda() returns objects with classes "rda" and "cca", but in most cases
those objects work like those of class "cca"

The Eigenvalues and axis scores are now spread about the \$CA and \$CCA components (also \$pCCA if a partial CCA)

Thankfully we can use extractor functions to get at such things

Eigenvalues

Use eigenvals() to extract Eigenvalues from a fitted ordination object

```
eigenvals(cca1)
                             CCA3
                                                 CCA5
        CCA1
                  CCA2
                                       CCA4
                                                            CCA6
                                                                      CCA7
                                                                                 CCA8
## 0.4388704 0.2917753 0.1628465 0.1421302 0.1179519 0.0890291 0.0702945 0.0583592
        CCA9
                 CCA10
                           CCA11
                                      CCA12
                                                CCA13
                                                           CCA14
                                                                       CA1
                                                                                 CA2
## 0.0311408 0.0132944 0.0083644 0.0065385 0.0061563 0.0047332 0.1977645 0.1419256
         CA3
                   CA4
                              CA5
                                        CA6
                                                  CA7
                                                             CA8
###
                                                                       CA9
## 0.1011741 0.0707868 0.0533034 0.0332994 0.0188676 0.0151044 0.0094876
```

Example

- Fit a CCA model to the lichen pasture data. The model should include, N, P, and K only.
- Save the model in object mycca1
- How much variance is explained by this model?
- Extract the eigenvalues, how many constrained axes are there?

```
library("vegan")
 data(varechem, varespec)
mycca1 \leftarrow cca(varespec \sim N + P + K, data = varechem)
mvcca1
## Call: cca(formula = varespec ~ N + P + K, data = varechem)
                Inertia Proportion Rank
## Total
                 2.0832
                            1.0000
## Constrained 0.4464 0.2143
                                      3
## Unconstrained 1.6368 0.7857
                                     20
## Inertia is scaled Chi-square
###
## Eigenvalues for constrained axes:
     CCA1
             CCA2
                     CCA3
###
## 0.19309 0.16271 0.09060
## Eigenvalues for unconstrained axes:
     CA1
            CA2
                        CA4
                                 CA5
                   CA3
                                        CA6
                                               CA7
                                                      CA8
## 0.4495 0.2870 0.1877 0.1675 0.1280 0.1050 0.0750 0.0629
```

(Showing 8 of 20 unconstrained eigenvalues)

Extracting axis scores

To extract a range of scores from a fitted ordination use scores()

- takes an ordination object as the first argument
- choices which axes? Defaults to c(1,2)
- display which type(s) of scores to return
 - "sites" or "wa": scores for samples in response matrix
 - "species": scores for variables/columns in response
 - "lc": linear combination site scores
 - "bp": biplot scores (coords of arrow tip)
 - "cn": centroid scores (coords of factor centroids)

Extracting axis scores

```
str(scores(cca1, choices = 1:4, display = c("species", "sites")), max = 1)
```

```
## List of 2
## $ species: num [1:44, 1:4] 0.0753 -0.1813 -1.0535 -1.2774 -0.1526 ...
## ..- attr(*, "dimnames")=List of 2
## $ sites : num [1:24, 1:4] 0.178 -0.97 -1.28 -1.501 -0.598 ...
## ..- attr(*, "dimnames")=List of 2
```

```
head(scores(cca1, choices = 1:2, display = "sites"))
```

```
## CCA1 CCA2
## 18 0.1784733 -1.0598842
## 15 -0.9702382 -0.1971387
## 24 -1.2798478 0.4764498
## 27 -1.5009195 0.6521559
## 23 -0.5980933 -0.1840362
## 19 -0.1102881 0.7143142
```

Scalings...

When we draw the results of many ordinations we display 2 or more sets of data

Can't display all of these and maintain relationships between the scores

Solution scale one set of scores relative to the other via the scaling argument

Scalings...

- scaling = 1 Focus on sites, scale site scores by λ_i
- ullet scaling = 2 Focus on species, scale species scores by λ_i
- scaling = 3 Symmetric scaling, scale both scores by $\sqrt{\lambda_i}$
- scaling = -1 As above, but
- scaling = -2 For cca() multiply results by $\sqrt{(1/(1-\lambda_i))}$
- scaling = -3 this is Hill's scaling
- scaling < 0 For rda() divide species scores by species' σ
- scaling = 0 raw scores

```
scores(cca1, choices = 1:2, display = "species", scaling = 3)
```

Scalings...

Thankfully we can use alternative descrpitors to extract scores:

- "none"
- "sites"
- "species"
- "symmetric"

Two modifiers select negative scores depending on whether the model is CCA or RDA:

- hill = TRUE
- correlation = TRUE

Example

 Using the CCA model you fitted, extract the site scores for axes 2 and 3 with Hill's scaling, focusing on the sites

```
## CCA2 CCA3
## 18 0.21507383 -0.22617222
## 15 -0.53564592 -0.14736699
## 24 -0.28328352 -0.56306912
## 27 -0.79825273 -0.35205393
## 23 -0.06029273 -0.09438971
## 19 0.04742753 0.09586591
```

Partial constrained ordinations

Partial constrained ordinations remove the effect of one or more variables then fit model of interest

Argument Z is used for a data frame of variables to partial out

Or with the formula interface use the Condition() function

```
pcca ← cca(varespec ~ Ca + Condition(pH), data = varechem) ## easier!
```

Partial constrained ordinations

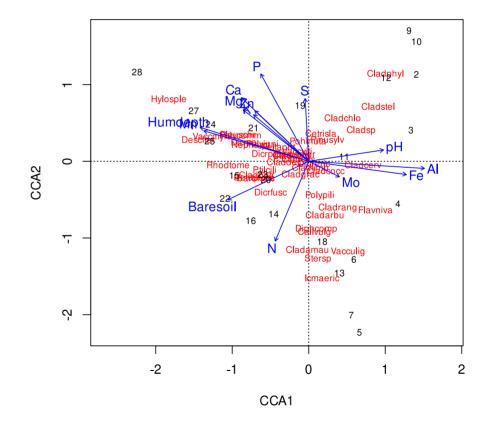
```
pcca ← cca(varespec ~ Ca + Condition(pH), data = varechem) ## easier!
pcca
```

```
## Call: cca(formula = varespec ~ Ca + Condition(pH), data = varechem)
###
                Inertia Proportion Rank
## Total
                2.0832
                          1.0000
## Conditional 0.1458 0.0700
                                    1
## Constrained 0.1827 0.0877 1
## Unconstrained 1.7547 0.8423
                                   21
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
     CCA1
## 0.18269
## Eigenvalues for unconstrained axes:
     CA1
            CA2
                   CA3
                                CA5
                         CA4
                                       CA6
                                              CA7
                                                    CAS
## 0.3834 0.2749 0.2123 0.1760 0.1701 0.1161 0.1089 0.0880
## (Showing 8 of 21 unconstrained eigenvalues)
```

Triplots

Triplots will generally produce a mess; we can really only display a couple of bits approximately anyway Trying to cram three things in is a recipe for a mess... but we can do it

plot(cca1)



LC vs WA scores

In constrained ordinations there are two sets of "site" scores

- 1. linear combination scores
- 2. weighted average (or weighted summation) scores

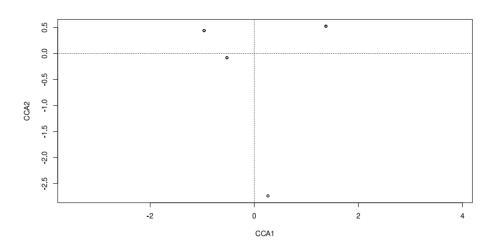
The LC scores are weighted & scaled versions of the constraints (predictors)

The WA scores are weighted averages of the species (responses) that are as close to the LC scores as possible

LC vs WA scores

Because the LC scores are based on the constraints you can get odd results if you plot them for some models

```
# example from Design Decision vignette
data(dune, dune.env, package = "vegan")
ord ← cca(dune ~ Moisture, data = dune.env)
plot(ord, display = "lc", type = "points")
```

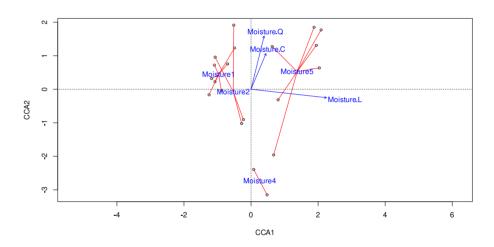


Moisture doesn't vary within observations of the same Moisture class (or level) so no variation in the LC scores for samples in each class.

LC vs WA scores

Weighted average scores include the variation in the species composition

```
# example from Design Decision vignette
data(dune, dune.env, package = "vegan")
ord ← cca(dune ~ Moisture, data = dune.env)
plot(ord, display = "wa", type = "points")
ordispider(ord, col = "red")
text(ord, display = "cn", col = "blue")
```



Model building

Building constrained ordination models

If we don't want to think it's easy to fit a poor model with many constraints — That's what I did with cca1 and rda1

Remember, CCA and RDA are *just regression methods* — everything you know about regression applies here

A better approach is to *think* about the important variables and include only those

The formula interface allows you to create interaction or quadratic terms easily (though be careful with latter)

It also handles factor or class constraints automatically unlike the basic interface

Building constrained ordination models

```
vare.cca ← cca(varespec ~ Al + P*(K + Baresoil), data = varechem)
vare.cca
```

```
## Call: cca(formula = varespec ~ Al + P * (K + Baresoil), data =
## varechem)
##
              Inertia Proportion Rank
## Total
           2.083
                            1,000
## Constrained 1.046 0.502 6
## Unconstrained 1.038 0.498 17
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
         CCA2
                  CCA3
                        CCA4
                             CCA5
    CCA1
                                     CCA6
## 0.3756 0.2342 0.1407 0.1323 0.1068 0.0561
## Eigenvalues for unconstrained axes:
      CA1
              CA2
                     CA3
                             CA4
                                     CA5
                                            CA6
                                                    CA7
                                                           CA8
## 0.27577 0.15411 0.13536 0.11803 0.08887 0.05511 0.04919 0.03781
## (Showing 8 of 17 unconstrained eigenvalues)
```

Building constrained ordination models

For CCA, RDA etc we have little choice but to do

- 1. Fit well-chosen set of candidate models & compare, or
- 2. Fit a *full* model of well-chosen variables & then do stepwise selection

Automatic approaches to model building should be used cautiously!

The standard step() function can be used as *vegan* provides two helper methods, deviance() and extractAIC(), used by step()

vegan also provides methods for class "cca" for add1() and drop1()

Variance inflation factors

Linear dependencies between constraints can be investigated with VIF

VIF is a measure of how much the variance of $\hat{\beta}_j$ is inflated by presence of other covariates

Lots of rules of thumb

- VIF >= 20 indicates strong collinearity in constraints
- VIF >= 10 potentially of concern & should be looked at

Computed via vif.cca()

```
vif.cca(cca1)

## N P K Ca Mg S Al Fe
## 1.981742 6.028515 12.009357 9.925801 9.810609 18.378794 21.192739 9.127762
## Mn Zn Mo Baresoil Humdepth pH
## 5.380432 7.739664 4.320346 2.253683 6.012537 7.389267
```

step() uses AIC which is a fudge for RDA/CCA — use ordistep()

- 1. Define an upper and lower model scope, say the full model and the null model
- 2. To step from the lower scope or null model we use

```
upr ← cca(varespec ~ ., data = varechem)
lwr ← cca(varespec ~ 1, data = varechem)
set.seed(1)
mods ← ordistep(lwr, scope = formula(upr), trace = 0)
```

trace = 0 is used here to turn off printing of progress

Permutation tests are used (more on these later); the theory for an AIC for ordination is somewhat loose

(Showing 8 of 20 unconstrained eigenvalues)

The object returned by step() is a standard "cca" object with an extra component \$anova

```
mods
## Call: cca(formula = varespec ~ Al + P + K, data = varechem)
                Inertia Proportion Rank
## Total
              2.0832
                            1,0000
## Constrained 0.6441 0.3092
## Unconstrained 1.4391
                        0.6908
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
    CCA1
         CCA2
                  CCA3
## 0.3616 0.1700 0.1126
## Eigenvalues for unconstrained axes:
                                 CA5
     CA1
            CA2
                   CA3
                          CA4
                                       CA6
                                              CA7
                                                     CA8
## 0.3500 0.2201 0.1851 0.1551 0.1351 0.1003 0.0773 0.0537
```

The \$anova component contains a summary of the steps involved in automatic model building

```
mods$anova

## Df AIC F Pr(>F)

## + Al 1 128.61 3.6749 0.005 **

## + P 1 127.91 2.5001 0.005 **

## + K 1 127.44 2.1688 0.050 *

## ---

## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

- 1. Al added first, then
- 2. P, followed by
- 3. K, then stopped

Step-wise model selection is fairly fragile; if we start from the full model we won't end up with the same final model

Stepwise selection in CCA

```
## Call: cca(formula = varespec ~ P + K + Mg + S + Mn + Mo + Baresoil +
## Humdepth, data = varechem)
                Inertia Proportion Rank
## Total
              2.0832
                          1.0000
## Constrained 1.1165 0.5360
## Unconstrained 0.9667
                          0.4640 15
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
    CCA1 CCA2
                CCA3 CCA4 CCA5
                                    CCA6
                                             CCA7
                                                    CCA8
## 0.4007 0.2488 0.1488 0.1266 0.0875 0.0661 0.0250 0.0130
###
## Eigenvalues for unconstrained axes:
      CA1
              CA2
                      CA3
                              CA4
                                     CA5
                                             CA6
                                                     CA7
                                                             CA8
                                                                     CA9
                                                                            CA10
## 0.25821 0.18813 0.11927 0.10204 0.08791 0.06085 0.04461 0.02782 0.02691 0.01646
     CA11
             CA12
                     CA13
                             CA14
                                    CA15
## 0.01364 0.00823 0.00655 0.00365 0.00238
```

Adjusted R^2 for ordination models

Ordinary \mathbb{R}^2 is biased for the same reasons as for a linear regression

- ullet adding a variable to constraints will increase R^2
- the larger the number of constraints in the model the larger \mathbb{R}^2 , is due to random correlations

Can attempt to account for this bias via an adjusted \mathbb{R}^2 measure

Adjusted R^2 for ordination models

Can attempt to account for this bias via an adjusted R^2 measure

$$R_{adj}^2 = 1 - rac{n-1}{n-m-1}(1-R^2)$$

- n is number of samples m is number of constraints (model degrees of freedom)
- ullet Can be used up to $\sim M > n/2$ before becomes too conservative
- Can be negative

```
RsquareAdj(cca1)
```

```
## $r.squared
## [1] 0.6919576
##
## $adj.r.squared
## [1] 0.2163163
```

Stepwise selection via adjusted ${\mathbb R}^2$

Problems with stepwise selection are myriad. Affects RDA, CCA, etc

Blanchet $\it et~al~(2008)$ proposed a two-step solution for models where $R^2_{\it adj}$ makes sense

Stepwise selection via adjusted ${\cal R}^2$

- Global test of all constraints
 - Proceed only if this test is significant
 - Helps prevent inflation of overall type I error
- Proceed with forward selection, but with two stopping rules
 - \circ Usual significance threshold lpha
 - \circ The global R^2_{adj}
 - \circ Stop if next candidate model is non-significant or if R^2_{adj} exceeds the global R^2_{adj}

Available in ordiR2step()

Stepwise selection via adjusted ${\cal R}^2$

```
ordiR2step(lwr, upr, trace = FALSE)
```

```
## Call: cca(formula = varespec ~ Al + P + K, data = varechem)
                Inertia Proportion Rank
## Total
            2.0832
                           1.0000
## Constrained 0.6441 0.3092
## Unconstrained 1.4391 0.6908
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
    CCA1 CCA2 CCA3
## 0.3616 0.1700 0.1126
###
## Eigenvalues for unconstrained axes:
            CA2
                   CA3
                         CA4
                                CA5
     CA1
                                       CA6
                                             CA7
                                                    CA8
## 0.3500 0.2201 0.1851 0.1551 0.1351 0.1003 0.0773 0.0537
## (Showing 8 of 20 unconstrained eigenvalues)
```

Permutation tests

RDA has lots of theory behind it, CCA bit less. However, ecological/environmental data invariably violate what little theory we have

Instead we use permutation tests to assess the *importance* of fitted models — the data are shuffled in some way and the model refitted to derive a Null distribution under some hypothesis of *no effect*

What *is* shuffled and *how* is of **paramount** importance for the test to be valid

- No conditioning (partial) variables then rows of the species data are permuted
- With conditioning variables, two options are available, both of which permute residuals from model fits
 - \circ The full model uses residuals from model Y=X+Z+arepsilon
 - \circ The reduced model uses residuals from model Y=Z+arepsilon
- In vegan which is used can be set via argument model with "direct", "full", and "reduced" respectively

A test statistic is required, computed for observed model & each permuted model

 \mathbf{vegan} uses a pseudo F statistic

$$F = rac{\chi^2_{model}/df_{model}}{\chi^2_{resid}/df_{resid}}$$

Evaluate whether F is unusually large relative to the null (permutation) distribution of F

```
pstat ← permustats(anova(cca1))
summary(pstat)
```

```
##

## statistic SES mean lower median upper Pr(perm)

## Model 1.4441 2.1270 1.0215 1.0033 1.3824 0.035 *

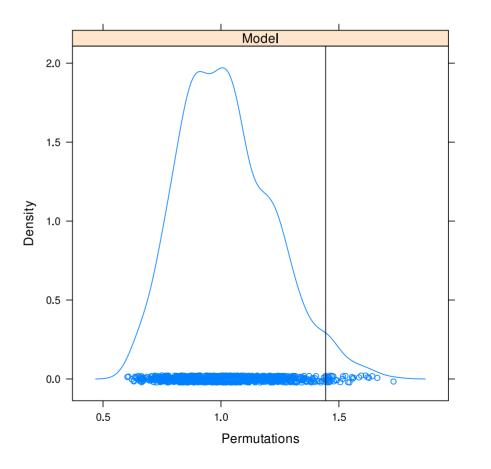
## ---

## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1

##

## (Interval (Upper - Lower) = 0.95)
```

densityplot(pstat)



Permutation tests in vegan: anova()

- The main user function is the anova() method
- It is an interface to the lower-level function permutest.cca()
- At its most simplest, the anova() method tests whether the model
 as a whole is significant

Permutation tests in vegan: anova()

$$F = rac{1.4415/14}{0.6417/9} = 1.4441$$

```
set.seed(42)
(perm ← anova(cca1))
```

Permutation tests in vegan: anova()

anova.cca() has a number of arguments

```
args(anova.cca)

## function (object, ..., permutations = how(nperm = 999), by = NULL,

## model = c("reduced", "direct", "full"), parallel = getOption("mc.cores"),

## strata = NULL, cutoff = 1, scope = NULL)

## NULL
```

object is the fitted ordination

permutations controls what is permuted and how

by determines what is tested; the default is to test the model

Types of permutation test in vegan

A number of types of test can be envisaged

- Testing the overall significance of the model
- Testing constrained (canonical) axes
- Testing individual model terms sequentially
- The marginal effect of a single variable

The first is the default in anova()

The other three can be selected via the argument by

Testing canonical axes

- The constrained (canonical) axes can be individually tests by specifying by = "axis"
- The first axis is tested in terms of variance explained compared to residual variance
- The second axis is tested after partialling out the first axis...
- and so on

Testing canonical axes

```
set.seed(1)
anova(mods, by = "axis")
```

Testing terms sequentially

- The individual terms in the model can be tested using by = "terms"
- The terms are assessed in the order they were specified in the model, sequentially from first to last
- ullet Test is of the additional variance explained by adding the kth variable to the model
- Ordering of the terms will affect the results

Testing terms sequentially

```
set.seed(5)
anova(mods, by = "terms")
```

Testing terms marginal effects

- The marginal effect of a model term can be assessed using by = "margin"
- The marginal *effect* is the effect of a particular term when all other model terms are included in the model

Testing terms marginal effects

```
set.seed(10)
anova(mods, by = "margin")
```

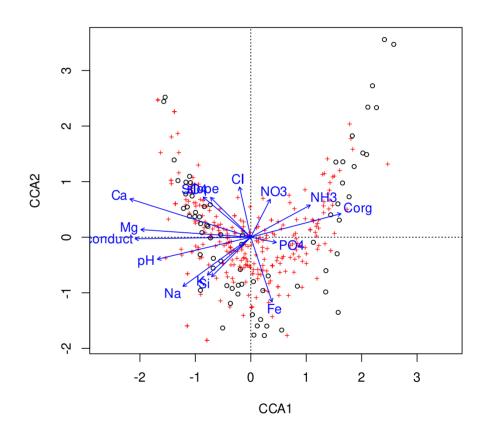
Example & data taken from Leps & Smilauer (2014), Case Study 2

Spring fen meadow vegetation in westernmost Carpathian mountains

CCA a reasonable starting point as the gradient is long here (check with decorana() if you want)

```
m1 ← cca(spp ~ ., data = env)
set.seed(32)
anova(m1)
```

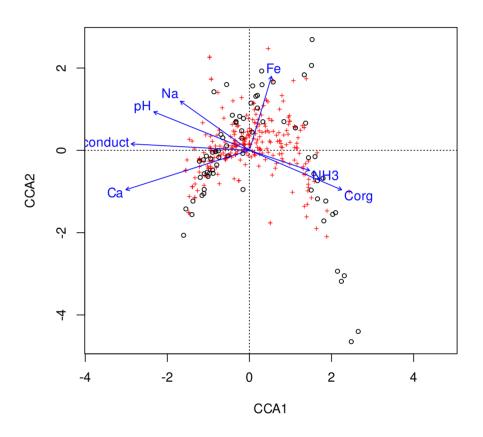
plot(m1)



```
set.seed(67)
lwr ← cca(spp ~ 1, data = env)
( m2 ← ordistep(lwr, scope = formula(m1), trace = FALSE) )
```

```
## Call: cca(formula = spp ~ Ca + conduct + Corg + Na + NH3 + Fe + pH,
## data = env)
                Inertia Proportion Rank
             5.3107
## Total
                        1.0000
## Constrained 0.9899 0.1864
## Unconstrained 4.3208
                         0.8136 62
## Inertia is scaled Chi-square
## Eigenvalues for constrained axes:
    CCA1 CCA2 CCA3 CCA4 CCA5 CCA6
                                            CCA7
## 0.4268 0.1447 0.1116 0.0936 0.0760 0.0719 0.0652
###
## Eigenvalues for unconstrained axes:
      CA1
              CA2
                     CA3
                             CA4
                                     CA5
                                            CA6
                                                    CA7
                                                            CA8
## 0.27251 0.19518 0.16703 0.14993 0.14606 0.14168 0.13292 0.12154
## (Showing 8 of 62 unconstrained eigenvalues)
```

plot(m2)



m2\$anova

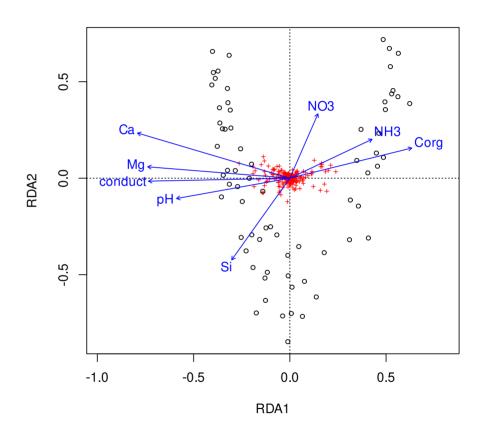
```
F Pr(>F)
           Df
                AIC
       1 453.14 4.7893 0.005 **
## + Ca
## + conduct 1 453.29 1.7915
                          0.005 **
## + Corg 1 453.61 1.6011 0.005 **
       1 453.93 1.5827 0.010 **
## + Na
## + NH3 1 454.36 1.4507 0.020 *
       1 454.89 1.3386 0.040 *
## + Fe
       1 455.46 1.2756 0.040 *
## + pH
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

Alternative is RDA with a transformation

```
m4
```

```
## Call: rda(formula = spph ~ Ca + NH3 + conduct + Si + Corg + NO3 + pH +
## Mg. data = env)
                Inertia Proportion Rank
## Total
                 0.6123
                            1.0000
## Constrained
                 0.1823
                            0.2977
## Unconstrained 0.4300
                            0.7023 61
## Inertia is variance
## Eigenvalues for constrained axes:
             RDA2
                    RDA3
                             RDA4
                                     RDA5
                                             RDA6
## 0.10572 0.02148 0.01224 0.01148 0.00945 0.00891 0.00696 0.00609
## Eigenvalues for unconstrained axes:
              PC2
                      PC3
                              PC4
                                      PC5
                                                              PC8
## 0.04311 0.03026 0.02030 0.01767 0.01649 0.01519 0.01383 0.01346
## (Showing 8 of 61 unconstrained eigenvalues)
```

plot(m4)



Stepwise using R^2_{adj}

```
m5 ← ordiR2step(lwr, scope = formula(m3), trace = FALSE)
m5$anova
```

```
R2.adj Df
                                      F Pr(>F)
                             AIC
                0.12588 1 -41.779 10.9370 0.002 **
## + Ca
## + NH3
                       1 -42.468 2.6242 0.002 **
              0.14628
## + conduct
            0.16322 1 -42.925 2.3570 0.002 **
## + Si
            0.17711 1 -43.164 2.1136 0.002 **
         0.18518 1 -42.940 1.6442 0.014 *
## + Corg
## + NO3
         0.19257 1 -42.680 1.5853 0.010 **
         0.19966 1 -42.417 1.5583 0.012 *
## + pH
## <All variables> 0.20332
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

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