ComGenR: Community Genetics Analyses in R

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Community Genetics is a field that works at the interface between ecological genetics and community ecology. Being inherently multi-disciplinary, the analytics involved have developed in separate fields. ComGenR is intended to synthesize these analytical techniques and facilitate new analytically and computationally driven research tools. Here, we present an introduction to the package, broken into five main sections:

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1 Community Genetics Data

Two primary aims of Community Genetics (CG) research are to test and quantify how genetic variation influences the distribution of species in a community [1]. These studies have typically examined the composition of a community of organisms associated with individuals of a focal species (e.g. [2] [3] [4]), which is most often a foundation species [5]. Thus, CG datasets tend to be in a community ecology form with sets individuals with multivariate observations of the abundances of associated species and phenotypic and/or genetic information. These data are most often compiled and curated using spreadsheets (e.g. Microsoft Excel).

When working in **R**, these data are most easily managed and imported if they are in a standardized column format, where the first column is a set of labels for each column. For more detailed introduction to **R**, a quick google search for "ecological analysis in R" will guide you to many resources, including my introductory course here.

In order allow for users to extend **R**'s functionality, functions are grouped together into "packages" by programmers. This allows for **R**to be reduced to a core set of software that can be added to by obtaining and loading these other packages. At current count (March 2012), there are over 4000 packages contributed by the **R**software community. This means, however, that any package that is not contained in the core **R**distribution must be downloaded initially and each time **R**is opened the package must be "libraried" (i.e. loaded into the working memory).

Here is the easiest way to do this:

> install.packages('ComGenR')

>

2

> library(ComGenR)

>

Once you have run install.packages, you'll only need to run library when you open up \mathbf{R} .

You can get some quick information on the package and any function by using the question mark symbol:

> ?ComGenR

>

Here, we will use an example dataset of a set of trees with genotypic and community data called $cg_data.csv$. Let's load the data and take a quick look at it's properties:

```
> the.data <- read.csv('cg_data.csv')
>
```

> colnames(the.data)

[1]	"tree.id"	"geno"	"pheno"	"S1"	"S2"	"S3"	"S4"
[8]	"S5"	"S6"	"S7"	"S8"	"S9"	"S10"	"S11"
[15]	"S12"	"S13"	"S14"	"S15"	"S16"	"S17"	"S18"
[22]	"S19"	"S20"	"S21"	"S22"	"S23"	"S24"	"S25"

>

> summary(the.data)

tree.id	geno	pheno	S1	
tree_1 : 1 N	Min. : 1.0	Min. :11.00	Min. : 0.	00001
tree_10: 1	1st Qu.: 3.0	1st Qu.:13.75	1st Qu.: 0.	00001
tree_11: 1 N	Median : 5.5	Median :15.62	Median: 13.	68308
tree_12: 1 N	Mean : 5.5	Mean :15.62	Mean : 34.	01201
tree_13: 1 3	3rd Qu.: 8.0	3rd Qu.:17.50	3rd Qu.: 69.	16127
tree_14: 1 N	Max. :10.0	Max. :21.00	Max. :114.	37323
(Other):44				
S2	S3		S4	S5
Min. : 0.46	626 Min. :	0.00001 Min	n. : 0.00001	Min. : 14.45
1st Qu.: 67.29	984 1st Qu.:	0.00001 1st	t Qu.: 70.77429	1st Qu.: 71.11
Median : 81.80	078 Median :	20.49474 Med	dian : 90.58988	Median : 85.99
Mean : 82.82	257 Mean :	35.23070 Mea	an : 84.21616	Mean : 84.70
3rd Qu.:107.58	350 3rd Qu.:	64.73125 3rd	d Qu.:102.75975	3rd Qu.:105.23
Max. :129.14	476 Max. ::	110.66000 Max	x.:125.48962	Max. :123.51
S6	S7		S8	S9
Min. : 0.35	502 Min. :	0.00001 Min	n. : 9.496	Min. : 0.00001
1st Qu.: 69.38	803 1st Qu.:	0.00001 1st	t Qu.: 72.335	1st Qu.: 44.23739
Median : 91.23	332 Median:	26.69620 Med	dian : 92.194	Median : 78.05749
Mean : 85.45	561 Mean :	39.75816 Mea	an : 88.753	Mean : 70.28158
3rd Qu.:105.50	072 3rd Qu.:	79.23763 3rd	d Qu.:106.059	3rd Qu.: 99.39094
Max. :126.05	525 Max. :	121.37132 Max	x.:128.524	Max. :126.78093
S10	S1:	1	S12	S13

Min. : 0.00001 Min. : 0.00001 Min. : 0.00001 Min. : 19.57

1st Qu.: 0.00001 1st Qu.: 42.00070 1st Qu.: 43.93994 1st Qu.: 67.95 Median: 80.82798 Median: 0.00001 Median: 79.87761 Median: 88.40 Mean : 71.06607 Mean : 75.58593 Mean : 24.74326 Mean : 84.82 3rd Qu.: 99.64315 3rd Qu.: 37.08868 3rd Qu.:104.02991 3rd Qu.:107.96 Max. :115.04995 Max. :112.32940 Max. :126.79579 Max. :125.99

S14 S15 S16 Min. : 0.00001 Min. : 0.00001 Min. : 0.00001 1st Qu.: 58.44259 1st Qu.: 54.28787 1st Qu.: 1.63454 Median: 84.05494 Median : 75.11145 Median: 33.23479 Mean : 77.95023 Mean : 67.32181 Mean : 43.38108 3rd Qu.: 97.15230 3rd Qu.: 88.26893 3rd Qu.: 78.32547 Max. :125.91562 Max. :116.66159 Max. :116.76917

Max. :128.48

Max. :125.91441

S19 S17 S18 S20 Min. : 0.00001 Min. : 40.22 Min. : 0.00001 Min. : 4.761 1st Qu.: 41.17744 1st Qu.: 73.69 1st Qu.: 16.96985 1st Qu.: 62.486 Median : 86.50 Median : 55.39803 Median: 74.84425 Median: 84.989 Mean : 67.76906 Mean : 89.54 Mean : 56.13915 Mean : 84.040 3rd Qu.: 89.32233 3rd Qu.: 97.72487 3rd Qu.:106.55 3rd Qu.:108.528

Max. :122.85165

Max. :129.071

 S21
 S22
 S23
 S24

 Min. : 16.68
 Min. : 12.86
 Min. : 0.00001
 Min. : 0.00001

 1st Qu.: 67.52
 1st Qu.: 68.16
 1st Qu.: 0.00001
 1st Qu.: 47.33374

 Median : 84.09
 Median : 91.39
 Median : 0.25435
 Median : 75.17726

Mean : 29.67761 : 82.88 Mean : 86.55 Mean Mean : 71.78652 3rd Qu.:106.30 3rd Qu.:101.49 3rd Qu.: 55.34534 3rd Qu.: 98.85638 :122.77 :128.81 Max. :106.12659 :121.11751 Max. Max. Max.

S25

Min. : 0.00001

1st Qu.: 54.46735

Median : 80.85409

Mean : 71.28334

3rd Qu.:100.82608

:121.03105

>

Max.

> head(the.data)

tree.id geno pheno S1 S2 S3 S4 S5 1 11.0 55.143075 66.5302804 69.60858 81.94837 73.15117 1 tree_1 2 tree_2 1 11.0 79.698932 38.7225631 80.25098 91.42431 87.69399 tree_3 1 11.0 80.761653 43.2485420 90.15421 104.61598 99.44019 tree_4 1 11.0 65.588369 114.0737494 57.83248 123.67524 123.50614 tree_5 1 11.0 83.601577 0.4625481 84.94255 101.33062 100.60255 2 12.5 7.694761 73.3390908 59.34273 100.64526 80.08185 tree_6 **S7 S8** S12 S6 S9 S10 S11 1 95.11142 79.62777 88.174283 74.472865 35.86047 25.92224 75.06928 2 65.68281 84.98665 66.346524 7.133347 13.04068 81.25744 34.62507 3 81.14893 90.96872 9.495741 0.000010 0.00001 70.19710 0.00001

```
92.11793
             50.38426 102.253264 50.373852
                                             38.70863
                                                       47.14492
                                                                 42.31913
                       17.464870 0.000010
  81.83055 121.37132
                                              0.00001 102.34208
                                                                  0.00001
6 116.48559
             47.97244
                       73.555096 99.620426 107.45411
                                                        0.00001 104.42196
        S13
                  S14
                            S15
                                       S16
                                                 S17
                                                           S18
                                                                      S19
1 117.12440 125.91562
                       67.84136
                                 80.79821 114.16074 106.01641
            73.89958 104.66712 100.51018 125.61742 77.67268 111.13336
2 100.25315
  67.04595
             60.63683 102.63896
                                 75.39869
                                            96.84453
                                                      66.70796
                                                                87.00148
  83.60133 103.65438
                       81.83632
                                 70.44718 103.77101 106.03700
                                                                93.91817
             96.45811
                       72.85438 102.94310 104.26641
                                                      40.22166 117.97061
5
  58.11346
  83.35166 104.38062
                       97.65935
                                  74.75413
                                            73.05425
                                                      72.83551
                                                                46.22320
        S20
                  S21
                            S22
                                       S23
                                                 S24
                                                           S25
1 114.04734 110.98139 101.04959
                                  60.11494
                                            99.24277
                                                      86.03105
             55.53267
                       80.41737 106.12659
2 123.50268
                                            70.96756
                                                      97.61656
3 108.85162
                                  75.23601 114.26938 100.76147
             88.41112
                       45.44620
  78.61885
             92.40699
                                  43.81558 101.82187 101.21749
                       91.48787
  65.95159
             35.38746
                       12.85607
                                 84.10541 102.33353 104.66011
6 115.56595
             82.18089 116.06115
                                  0.00001
                                           79.84856
                                                      81.01749
```

For ease of conducting analyses, it is best to isolate the community and the "environmental" (i.e. tree ID, genotype and phenotype) data. This can be done in many ways, but we'll do it here by selecting the columns containing species abundance data (i.e. columns 4 to 28) and the genotype data (i.e. column 2) creating two new objects ("com" and "geno"):

```
> com <- the.data[,4:28]</pre>
```

>

> colnames(com)

- [1] "S1" "S2" "S3" "S4" "S5" "S6" "S7" "S8" "S9" "S10" "S11" "S12"
- [13] "S13" "S14" "S15" "S16" "S17" "S18" "S19" "S20" "S21" "S22" "S23" "S24"
- [25] "S25"

> summary(com)

S1 S2 S3 S4

Min. : 0.00001 Min. : 0.4626 Min. : 0.00001 Min. : 0.00001

1st Qu.: 0.00001 1st Qu.: 67.2984 1st Qu.: 0.00001 1st Qu.: 70.77429

Median: 13.68308 Median: 81.8078 Median: 20.49474 Median: 90.58988

Mean : 34.01201 Mean : 82.8257 Mean : 35.23070 Mean : 84.21616

3rd Qu.: 69.16127 3rd Qu.:107.5850 3rd Qu.: 64.73125 3rd Qu.:102.75975

Max. :114.37323 Max. :129.1476 Max. :110.66000 Max. :125.48962

S5 S6 S7 S8

1st Qu.: 71.11 1st Qu.: 69.3803 1st Qu.: 0.00001 1st Qu.: 72.335

Median: 85.99 Median: 91.2332 Median: 26.69620 Median: 92.194

Mean : 84.70 Mean : 85.4561 Mean : 39.75816 Mean : 88.753

3rd Qu.:105.23 3rd Qu.:105.5072 3rd Qu.: 79.23763 3rd Qu.:106.059

Max. :123.51 Max. :126.0525 Max. :121.37132 Max. :128.524

S9 S10 S11

Min. : 0.00001 Min. : 0.00001 Min. : 0.00001

1st Qu.: 44.23739 1st Qu.: 42.00070 1st Qu.: 0.00001

Median: 78.05749 Median: 80.82798 Median: 0.00001

Mean : 70.28158 Mean : 71.06607 Mean : 24.74326

3rd Qu.: 99.39094 3rd Qu.: 99.64315 3rd Qu.: 37.08868

Max. :126.78093 Max. :115.04995 Max. :112.32940

S12	S13	S14	S15
Min. : 0.00001	Min. : 19.57	Min. : 0.00001	Min. : 0.00001
1st Qu.: 43.93994	1st Qu.: 67.95	1st Qu.: 58.44259	1st Qu.: 54.28787
Median : 79.87761	Median : 88.40	Median : 84.05494	Median : 75.11145
Mean : 75.58593	Mean : 84.82	Mean : 77.95023	Mean : 67.32181
3rd Qu.:104.02991	3rd Qu.:107.96	3rd Qu.: 97.15230	3rd Qu.: 88.26893
Max. :126.79579	Max. :125.99	Max. :125.91562	Max. :116.66159
S16	S17	S18	S19
Min. : 0.00001	Min. : 0.000	001 Min. : 40.22	Min. : 0.00001
1st Qu.: 1.63454	1st Qu.: 41.17	744 1st Qu.: 73.69	1st Qu.: 16.96985
Median : 33.23479	Median : 74.84	425 Median: 86.50	Median : 55.39803
Mean : 43.38108	Mean : 67.769	906 Mean : 89.54	Mean : 56.13915
3rd Qu.: 78.32547	3rd Qu.: 97.72	487 3rd Qu.:106.55	3rd Qu.: 89.32233
Max. :116.76917	Max. :125.91	441 Max. :128.48	Max. :122.85165
S20	S21	S22	S23
Min. : 4.761	Min. : 16.68	Min. : 12.86 Min	. : 0.00001
1st Qu.: 62.486	1st Qu.: 67.52	1st Qu.: 68.16 1st	Qu.: 0.00001
Median : 84.989	Median : 84.09	Median: 91.39 Med	lian : 0.25435
Mean : 84.040	Mean : 82.88	Mean : 86.55 Mea	n : 29.67761
3rd Qu.:108.528	3rd Qu.:106.30	3rd Qu.:101.49 3rd	Qu.: 55.34534
Max. :129.071	Max. :122.77	Max. :128.81 Max	:. :106.12659
S24	S25		
Min. : 0.00001	Min. : 0.000	001	
1st Qu.: 47.33374	1st Qu.: 54.46	735	

Median: 75.17726 Median: 80.85409

Mean : 71.78652 Mean : 71.28334

3rd Qu.: 98.85638 3rd Qu.:100.82608

Max. :121.11751 Max. :121.03105

- > geno <- the.data[,2]</pre>
- > summary(geno)

Min. 1st Qu. Median Mean 3rd Qu. Max.

1.0 3.0 5.5 5.5 8.0 10.0

> geno

9 10 10 10 10 10 [26]

>

Note that **R**is treating genotype as a set of numbers instead of genotypic categories. It is important that we change this in order to avoid in-correct analyses later on. This is easily done with the following code:

- > geno <- factor(geno)
- > summary(geno)

1 2 3 4 5 6 7 8 9 10

5 5 5 5 5 5 5 5 5

> geno

[26] 6 6 6 6 6 8 8 10 10 10 10 10 7 7

Levels: 1 2 3 4 5 6 7 8 9 10

>

We can tell that **R**is now treating our "geno" values as categorical because it returns a list of the levels present in our "geno" object.

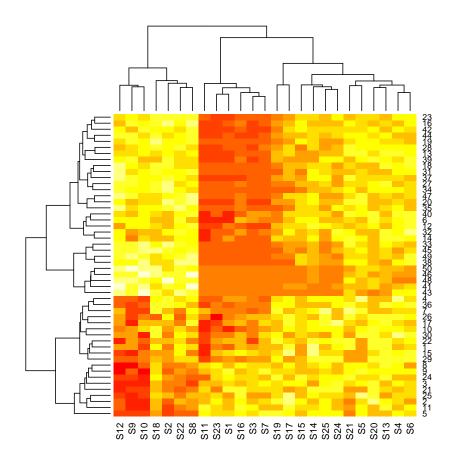
2 Community Composition

Now the we have imported, checked and corrected the format of our data, we can start conducting analyses. A good first step is a visual analysis of the data. As community data are inherently multivariate, direct observation of the data requires the aid of sophisticated visualizations. Two useful approaches are heatmaps and ordinations.

Heatmap

> heatmap(com)

>



NMDS Ordination

Non-metric Multidimensional Scaling (NMDS) ordination plots are a much more common, albeit abstract means of visualizing community data. In CG studies, it has also been used as a way to generate a trait-like vector that can be used in quantitative genetic analyses. We can quickly do this in Rusing functions from the *vegan* and *ecodist* packages:

- > d <- vegdist(com)</pre>
- > nms <- nmds(d,mindim=2,maxdim=2,nits=3)</pre>

```
Using random start configuration
Using random start configuration
Using random start configuration
>
```

Note here that we first calculate the Bray-Curtis dissimilarity scores for each observation (which we call "d"). This distance matrix in then used to conduct the ordination. Here we set the "mindim" and "maxdim" arguments in the function to 2 so that we will get a set of ordinations with that dimensionality. Because the NMDS procedure starts with a randomly generated set of numbers that are then adjusted until best represent the structure of the original distances of the data, we have also specified the "nits" argument to be 3, which will have the function output 3 ordinations. We then select the lowest "stress" (i.e. the best fitted) ordination from our set of three.

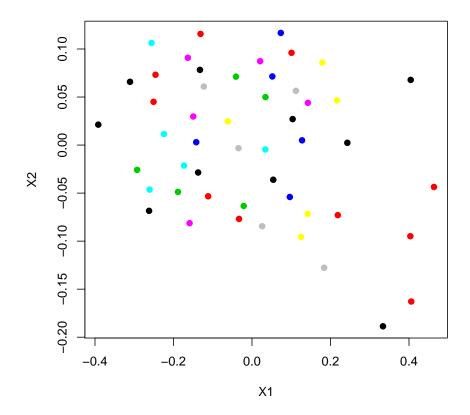
```
> nms <- nmds.min(nms)

Minimum stress for given dimensionality: 0.1102475
r^2 for minimum stress configuration: 0.9698926</pre>
```

Note first that the fit is below the arbitrary threshold of 0.2 and that the low number of iterations used here has been chosen purely for example's sake. Run?nmds to get a more detailed description of the NMDS and how to customize its functionality.

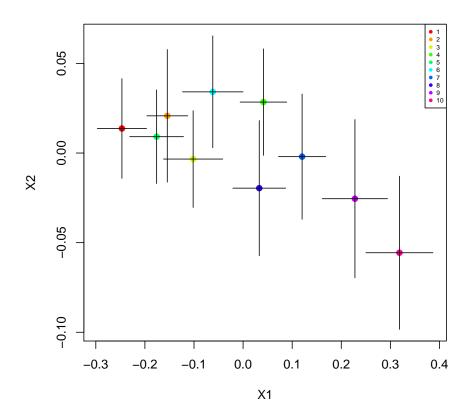
We can now plot our ordination, overlaying our genotype information:

```
> plot(nms,col=as.numeric(geno),pch=19)
>
```



Although the stress of the ordination is low, it is still difficult to see the patterns of the genotypes. Another method can be used to plot our ordination using the centroids (i.e. multivariate means) and the standard errors. This can be done easily with this function from the ComGenR pacakge:

```
> ch.plot(nms,geno,plot.legend=TRUE,loc='topright')
>
```

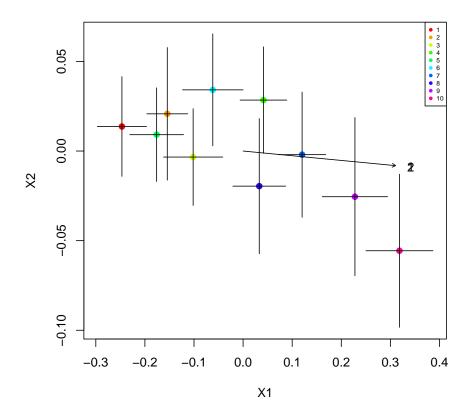


Vectors

>

It is also easy to overlay other information (such as out phenotype) onto our ordination using vectors:

```
> pheno <- the.data[,3]
> pheno.vector <- envfit(nms,pheno)
> ch.plot(nms,geno,plot.legend=TRUE,loc='topright')
> plot(pheno.vector,col='black')
```



${\bf PerMANOVA}$

Permutational Multivariate Analysis of Variance (PerMANOVA) has been developed by ecologists, namely Marti Andersen, to address the need for a multivariate test of compositional effects that accommodates the often non-normal distributions of community data. We can execute it easily in Rusing the interestingly named adonis function from the *vegan* package:

> adonis(com~geno)

The *ComGenR* package provides an additional function for conducting pairwise PerMANOVAs for levels of a single factor, such as genotype, in order to identify the statistical differences among pairs of levels:

```
> pp.results <- pair.permanova(x=com,f=geno,nits=999)</pre>
```

- [1] "1 vs 2"
- [1] "1 vs 3"
- [1] "1 vs 4"
- [1] "1 vs 5"
- [1] "1 vs 6"
- [1] "1 vs 7"
- [1] "1 vs 8"
- [1] "1 vs 9"

- [1] "1 vs 10"
- [1] "2 vs 3"
- [1] "2 vs 4"
- [1] "2 vs 5"
- [1] "2 vs 6"
- [1] "2 vs 7"
- [1] "2 vs 8"
- [1] "2 vs 9"
- [1] "2 vs 10"
- [1] "3 vs 4"
- [1] "3 vs 5"
- [1] "3 vs 6"
- [1] "3 vs 7"
- [1] "3 vs 8"
- [1] "3 vs 9"
- [1] "3 vs 10"
- [1] "4 vs 5"
- [1] "4 vs 6"
- [1] "4 vs 7"
- [1] "4 vs 8"
- [1] "4 vs 9"
- [1] "4 vs 10"
- [1] "5 vs 6"
- [1] "5 vs 7"
- [1] "5 vs 8"
- [1] "5 vs 9"

- [1] "5 vs 10"
- [1] "6 vs 7"
- [1] "6 vs 8"
- [1] "6 vs 9"
- [1] "6 vs 10"
- [1] "7 vs 8"
- [1] "7 vs 9"
- [1] "7 vs 10"
- [1] "8 vs 9"
- [1] "8 vs 10"
- [1] "9 vs 10"

> pp.results\$p.mat

1 2 3 4 5 6 7 8 10 9 NA 0.261 0.106 0.025 0.388 0.077 0.010 0.014 0.014 0.013 NA 0.542 0.034 0.941 0.337 0.008 0.049 0.007 0.004 2 NANA 0.102 0.342 0.603 0.035 0.157 0.010 0.009 3 NANANA 0.027 0.193 0.293 0.965 0.075 0.022 4 NANANANA 0.181 0.019 0.037 0.013 0.010 5 NANANANANA 0.054 0.304 0.026 0.015 NANANANA6 NANANANANA 0.325 0.268 0.093 7 NANANANANANANANANANANA 0.084 0.025 9 NANANANANANANANANA 0.348 10 NA NANANANANANANANANA

>

Note, these p-values are not adjusted for multiple tests. It has been stated that given the permutational nature of the test statistic used in PerMANOVA, that this is not necessary [6]. They can, however, be easily adjusted in **R**, see the ?p.adjust.

Genotype Means for Species Abundances

Last, it is worth noting here that the *ComGenR* package contains two functions to help calculate the means and standard errors for each species on a set of genotypes. This might be useful for plotting:

> mean.g(com,geno)

	S1	. S2	S3	S4	S5	S6	S7
1	72.95872122	2 52.60754	76.557761	100.59891	96.87881	83.17833	85.4677456
2	56.69297651	68.53598	68.909479	103.05853	106.65108	98.44179	74.8649139
3	51.88369704	65.35430	50.616295	97.79041	92.84703	105.77472	44.1075264
4	18.25755529	109.75559	12.935158	79.39797	86.46640	82.52525	23.1239447
5	75.48528310	78.07419	62.112840	89.81027	87.05418	92.63548	70.6078202
6	44.85606705	94.02754	47.787183	107.50176	95.77763	108.14469	57.9241813
7	8.55379707	101.86531	8.110586	84.77437	70.42539	74.27427	11.8072010
8	11.38816789	96.08158	25.277710	90.53159	89.00881	100.50032	29.2444528
9	0.04378675	82.98088	0.000010	56.71628	65.07748	66.29151	0.4337885
10	0.00001000	78.97367	0.000010	31.98149	56.79711	42.79418	0.0000100
	S8	S9	S10	S11	S12	S13	S14
1	56.74694	26.39602	17.52196 65	5.372756	30.40270 8	35.22766	92.11290
2	76.10238	51.38230	57.33525 54	1.066535	51.98798	99.20441	96.39379
3	89.64142	55.19926	72.18741 30	0.189585 6	33.86459 10	05.04676	96.27458

```
4 108.36642 83.01691 80.04658 6.116174 89.80038 87.69209 84.81264
5
   72.33276 48.19628 39.58385 53.036931 40.14465 94.32357 100.47270
6
   97.64410 74.39221 72.12086 27.326698 77.13680 113.46773 89.38643
7
   85.91710 82.12188 92.83378 4.033882 110.86225 83.40102 68.28648
   99.68091 84.92606 85.77424 7.289999 88.78019 80.91477 70.95691
8
9 103.92340 93.30570 103.10800 0.000010 108.38613 54.35210 42.28541
10 97.17063 103.87917 90.14880 0.000010 94.49367 44.61974 38.52047
                         S17
       S15
                S16
                                  S18 S19
                                                    S20
                                                             S21
1 85.96763 86.019473 108.93202 79.33114 99.18178 98.19442 76.54393
2 89.30438 70.738822 91.18252 98.28993 84.72937 104.93359 100.06243
3 74.09788 56.043769 92.90926 78.58441 74.77891 85.79231 85.97045
4 83.13640 28.943160 65.06201 100.25234 43.46535 78.93218 89.84110
5 88.65563 84.227058 86.86709 94.70526 93.86222 104.96906 91.90441
6 75.73313 55.088124 77.90997 101.88877 76.73125 113.66565 93.43136
7 45.26008 17.237142 41.47292 89.36479 15.87536 67.36296 82.84907
8 64.94430 25.132405 60.66301 84.55315 50.89024 86.03285
                                                         98.27927
9 37.05411 3.200096 33.97264 94.35563 15.03518 56.38830 60.77949
10 29.06456 7.180708 18.71920 74.07238 6.84187 44.13021 49.12444
        S22
                 S23
                          S24
                                   S25
   66.25142 73.879707 97.72702 98.05734
1
2
   79.29482 60.078459 88.12643 103.80586
3
   88.03367 39.732162 82.25411 82.31501
   87.78842 7.451767 69.70087 70.51218
4
```

76.21280 60.644732 106.01405 99.27077

94.14219 30.261926 85.53601 90.15915

98.57979 8.715062 64.58645 48.51186

5

6

7

- 8 102.14983 16.012311 68.66818 77.39797
- 9 84.25041 0.000010 43.63751 30.54391
- 10 88.80604 0.000010 11.61458 12.25931

> se.g(com,geno)

S1 S2 S3 S4 S5 S6 **S7** 5.43739796 18.670058 5.778127 7.004304 8.296997 5.168865 11.3677516 2 19.33808894 6.722551 8.565175 4.272462 6.979069 7.519987 11.9702015 3 14.85762786 11.806658 19.777658 7.139296 6.754016 2.771125 14.4360210 4 13.51048754 9.275191 11.505846 4.700245 8.010242 8.925719 17.4309123 5 20.05753949 11.561569 15.598365 4.780548 9.453669 5.632065 13.0486235 6 15.03419429 12.768953 18.976293 12.665196 9.523529 8.869932 18.7127527 8.55378707 5.631535 8.110576 6.170892 6.561304 12.411438 11.6856331 8 9.25256856 9.351974 14.193028 9.212339 5.771482 10.184360 12.7238895 0.04377675 10.555517 0.000000 11.545928 16.310413 5.925743 0.4337785 9 10 0.00000000 10.195389 0.000000 12.029387 14.440445 19.683846 0.0000000 S8 S9 S10 S11 S12 S13 S14 18.609691 15.249375 8.424058 13.287071 14.148463 10.750736 11.439359 2 8.683733 17.514793 19.384793 17.809221 18.673534 8.763967 6.320158 3 11.849952 17.252835 18.231981 21.668772 12.625390 8.041351 11.002180 9.498094 15.447826 4.362404 6.116164 11.243921 7.954612 5.840242 5 13.046998 16.228164 13.450362 14.667054 12.644679 9.981228 6.680258 4.536860 11.555202 13.455153 14.193351 11.551900 5.057715 11.138096 7 4.748334 8.108177 7.069453 4.033872 4.971996 9.002124 8.033439 8 9.447377 15.950437 12.165406 7.289989 12.373326 13.305234 7.910734 7.501465 8.958307 6.514739 0.000000 5.783432 15.259269 17.668598

```
10 11.834437 8.767616 6.566103 0.000000 10.114481 12.524265 16.201040
        S15
                 S16
                          S17
                                    S18
                                              S19
                                                       S20
                                                                S21
   7.567003 6.629286 5.001266 12.483897 6.514305 11.018625 13.626689
1
2
   5.600815 9.237984 6.976859 8.835930 11.191024 8.131273 6.682346
3
   6.179000 20.450765 10.425164 3.726023 5.034212 6.581304 11.988811
   6.466336 13.869177 12.057774 10.412588 11.321905 7.884235 8.444292
4
5 7.532464 14.975124 12.326897 9.743686 14.279270 5.397849 11.091566
6 12.603375 13.509516 14.671140 12.167747 18.913283 6.094358 10.780627
7 11.476068 11.137053 15.463569 6.814927 11.635601 8.256831 8.762707
8 7.156257 11.114869 17.465461 7.163573 16.764906 12.798263 9.994269
9 18.900828 3.200086 15.159717 13.846307 9.321271 14.583797 11.159802
10 18.218019 7.180698 14.681206 7.146889 4.642727 8.403332 13.257250
        S22
                 S23 S24
                                    S25
1 16.328481 10.582029 7.176362 3.207189
2 11.490827 17.974677 7.989786 7.243416
3 9.118039 18.468054 5.600795 9.194714
4
   9.074147 7.451757 13.733018 10.588776
5 16.334576 16.471093 4.914319 8.206593
6
   8.624746 12.923827 15.426250 12.377842
7
   7.541532 8.715052 12.525196 10.288491
   5.467973 10.587743 13.575491 12.240856
8
9
   6.151666 0.000000 14.992092 17.653522
10 8.582013 0.000000 8.876805 7.967191
```

>

3 Modeling and Quantifying Heritability

Community Genetics also seeks to quantify how much variation in the community is explained by genetic variation. The *ComGenR* package has several functions for both modeling and quantifying the community level effects of genetic variation as developed in the Shuster et al. 2006 [3] article.

Simulating Community Genetics

In general, simulation modeling is a useful tool for exploring possible mechanistic explainations for patterns. As community geneticists are interested in understanding how genetics influences community patterns, it is useful to have a simple simulation framework. ComGenR provides a set of functions to do this. Described more fully here [3], briefly the model simulates the response of a community of dependent species to selection imposed by genetically based phenotypic variation in a foundation species. This can be done by first creating a set of "trees" and a set of "insects" that form a dependent community. This can be done by hand, but ComGenR provides two functions to easily do this:

```
> trees <- gpmTrees()
> insects <- gpmCom()
>
```

Note the structure of these two matrices:

```
[2,] 1 11.0
```

- [3,] 1 11.0
- [4,] 1 11.0
- [5,] 1 11.0
- [6,] 2 12.5

> head(insects)

- [1,] 2.668155 3.451313
- [2,] 6.721325 7.999297
- [3,] 6.448846 7.297900
- [4,] 5.664091 5.855590
- [5,] 6.870599 7.561521
- [6,] 9.371366 10.567356

The "trees" matrix has two columns: geno and pheno. The "geno" value is the genotype of each tree in each row and "pheno" is the associated phenotype that is used to determine the effect of that tree on the arthropod community. The "insects" matrix has phenotypic values for each insect species in each row. These "insect" values are generated randomly using a heterkaryotic genome model from within a range of user determined values for each of two alleles.

Now, these values can be used to simulate the response of a community of arthropods:

```
> our.sim <- cgSim(trees,insects,reps=1,YY=5,GG=5)</pre>
```

- [1] "1 1 1"
- [1] "1 1 2"

- [1] "1 1 3"
- [1] "1 1 4"
- [1] "1 1 5"
- [1] "1 2 1"
- [1] "1 2 2"
- [1] "1 2 3"
- [1] "1 2 4"
- [1] "1 2 5"
- [1] "1 3 1"
- [1] "1 3 2"
- [1] "1 3 3"
- [1] "1 3 4"
- [1] "1 3 5"
- [1] "1 4 1"
- [1] "1 4 2"
- [1] "1 4 3"
- [1] "1 4 4"
- [1] "1 4 5"
- [1] "1 5 1"
- [1] "1 5 2"
- [1] "1 5 3"
- [1] "1 5 4"
- [1] "1 5 5"

>

This outputs a set of simulated communities. The "reps", "YY" and "GG" arguments determine the number of iterations, environmental scenarios and selection intensity scenarios. For each environmental scenario the effect of the genetic variance is heald constant and the amount of random noise introduced by non-genetic influences is increased. Similarly, each selection intensity scenario increases the effect of genetic variation while holding the influence of the environment constant.

NMDS Community Trait

Per the methods of Shuster et al. 2006, we can take one of our simulated matrices and summarize the variation of the community with an NMDS ordination. This is done in order to be able to treat the multivariate community as a univariate trait that has similar statistical properties as traits analyzed in quantitative genetics (e.g. univariate and normally distributed). We can use the same ordination methods that we used above to get a single NMDS ordination axis for a simulated community:

```
> com.sim <- our.sim[[1]][[1]][[5]]
> d <- vegdist(com.sim)
> nms.sim <- nmds(d,mindim=1,maxdim=1)

Using random start configuration

Using random start configuration</pre>
```

```
Using random start configuration
Using random start configuration

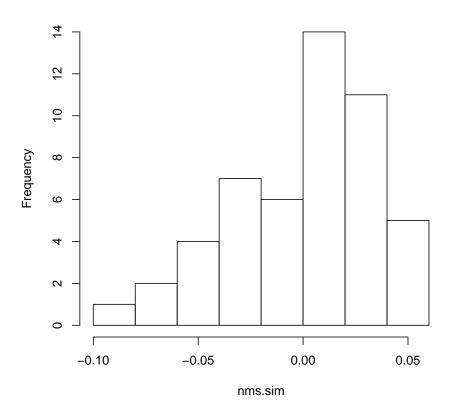
Vsing random start configuration

> nms.sim <- nmds.min(nms.sim)

Minimum stress for given dimensionality: 0.03004015
r^2 for minimum stress configuration: 0.9980397

> nms.sim <- nms.sim[,1]
>
hist(nms.sim)
```

Histogram of nms.sim



In this output, note both the stress and the r^2 of the final configuration. This similarly indicates how well the ordination represents the original data. As one would expect, this representation is never perfect as it is intended to be an abstraction of the original data. The user should be familiar with the meaning of ordinated scores and how they can and should be interpreted.

Community Heritability

We can now use this ordinated representation of the community to calculate the community heritability value for this simulated population of trees:

>

The output gives heritability score for the community, as represented by the ordination, along with associated confidence limits.

4 Network Modeling and Co-occurrence Analyses

Community Ecology and Community Genetics deal with complex sets of organisms largely because these fields aknowledge the need to study groups of organisms. A primary motivation for this is that species interact and these interactions contribute to variation in their distributions, abundances and function. Thus, communities are formed by webs or networks of interacting species and a complete understanding of communities requires an understanding of these networks. Thus, the *ComGenR* package provides tools for both modeling and analyzing relationships among species comprising communities.

Null-Model Co-occurrence Analysis

Given the motivation described above, it is unfortunate that interaction data is exceedingly rare and difficult to obtain for ecological studies. Initially developed for

biogrographic studies, co-occurrence analysis was developed to bridge this information gap. At its inception [7], the analysis of species co-occurrence patterns was inteded to generate and test hypotheses about how communities assemble [8]. It was posited that interactions among species influenced the distribution of species in space, namely through competitive exclusion [7], though later work has demonstrated the importance of positive interactions [9].

Analyzing co-occurrence patterns in CG data provides a well developed means to examine the co-variances among species. Once the effect of genetic variation on community composition has been established, co-occurrence analysis can then be used to examine the overarching structure in the community data due in part to that genetic effect. To do this, we use permutation based null modeling tools provided in the *vegan* package. *ComGenR* provides high level access to these functions, so that these analyses can be performed as follows:

++++++++	I	20%
	I	24%
++++++++++	I	28%
-+++++++++++++	1	32%
	1	02/0
++++++++++++++	I	36%
-+++++++++++++++++	1	40%
	·	,0
++++++++++++++++++	1	44%
++++++++++++++++++++++++++++++++++++++	I	48%
	ı	52%
I		
++++++	1	56%
		60%
++++++++++++++++++++++++++++++++++++++		60%
++++++	I	64%
I		
++++++		68%

```
72%
                                 76%
                                 80%
                                 84%
                                 88%
                                 92%
                                 96%
 SES
       lower.p
             upper.p
-42.85706
       0.00000
             1.00000
>
```

It is important to consider a threshold of detection for species prior to running co-occurrence analysis, since it does not use abundance data but presence-absence data (i.e. occurrences and non-occurrences). Here, we set values less than 1 to zero.

Although co-occurrence analyses allow us to test for the average structure of cooccurrence patterns in the community, they do not resolve the structure those patterns. Although the network approach has been employed in ecology for a relatively long time (e.g. [10]), recent developments in analytical methods have expanded utility and scope of this approach [11]. The *ComGenR* package provides several functions for the user to analyze CG data using a network modeling and analytical approach.

First to compliment the co-occurrence analysis, it is extremely useful to plot community data as a bipartite network. This re-representation of the data in this context allows for the examination of co-occurrence patterns. To do this, we use tools from the *bipartite* package:

```
> com. <- com
> com.[com.<=85] <- 0
> com. <- com.[,order(apply(com,2,sum),decreasing=TRUE)]
> rownames(com.) <- the.data$tree.id
> geno.color <- rainbow(nlevels(geno))[as.numeric(geno)]
> plotweb(com.,method='normal',col.low=geno.color,text.rot=90)
>
```

It's useful to note here that previous studies of bipartite networks in ecology have shown that these networks tend to have a nested structure that has potentially stabilizing effects on the community as a whole [12]. The *bipartite* package provides a means to test for this. For more information see ?nestedness.

Next, we can use another network approach to examine these co-occurrence patterns with regard to the relationship *among* species in the community matrix. Before do so, it is important to provide a brief caveat. This approach is meant to explore the data, and, toward this end, it provides a perspective that appears to resolve interactions among species. While this may be the case, this is not testable with the analysis itself. It is up to the user to decide to what extent these results can be used to speak to the structure of true ecological interactions (e.g. trophic or pollination) given the

nature of the data and other information about the community. However, analysis is only useful with appropriate interpretation, and it can be argued that ecological interactions tend to occur locally, and, thus if species are observed at an appropriate scale, it is possible to make inferences about the potential for interactions to occur, given non-random patterns of co-occurrence [11].

Here is how to conduct the co-occurrence based network modeling described in Araujo et al. 2011 [11] in the *ComGenR* package:

> net <- CoNetwork(com.)</pre>

> net

	S18	S8	S22	S6	S13	S5	S4	S20	S21	S2	S14	S12	S24
S18	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S8	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S22	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S6	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S13	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S5	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S4	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S20	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S21	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S2	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S14	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S12	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S24	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.0000000
S25	0	0	0	0	0	0	0	0.3095219	0	0	0.2762403	0.8432432	0.3012886
S10	0	0	0	0	0	0	0	0.0000000	0	0	0.0000000	0.0000000	0.8730844

```
0 0.0000000
S9
     0
        0
            0
               0
                  0
                     0
                                        0 0.0000000 0.3497901 0.0000000
S17
     0
        0
            0
               0
                  0
                     0
                        0 0.0000000
                                     0
                                        0 0.0000000 0.9203663 0.3468235
                                        0 0.0000000 0.0000000 0.0000000
S15
     0
        0
            0
               0
                  0
                     0
                        0 0.0000000
                                     0
S19
                        0 0.0000000
                                        0 0.0000000 0.9571190 0.2851947
        0
            0
               0
                  0
                     0
                                        0 0.0000000 1.0000000 0.0000000
S16
     0
               0
                        0.0000000
S7
                        0 0.0000000
                                        0 0.0000000 0.0000000 0.5461307
     0
        0
            0
               0
                  0
                     0
                                        0 0.0000000 0.0000000 0.0000000
S3
               0
                     0
                        0 0.0000000
     0
        0
            1
                  0
                                     0
               0
                     0
                        0 0.0000000
                                     0
                                        0 0.0000000 0.0000000 0.0000000
S1
     0
        0
            0
                  0
S23
     0
        0
            0
               0
                  0
                     0
                        0 0.0000000
                                     0
                                        0 0.0000000 0.0000000 0.0000000
                                        0 0.0000000 0.0000000 0.0000000
S11
            0
               0
                  0
                     0
                        0 0.0000000
         S25
                             S9
                                     S17 S15
                                                  S19
                                                                      S7
                  S10
                                                            S16
0 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
S8
   0.0000000 \ 0.0000000 \ 0.0000000 \ 0.0000000
0 0.0000000 0.0000000 0.0000000
   0.0000000 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
0 0.0000000 0.0000000 0.0000000
S5
   0.0000000 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
S4
   0.0000000 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
S20 0.3095219 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
0 0.0000000 0.0000000 0.0000000
   0.0000000 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
S14 0.2762403 0.0000000 0.0000000 0.0000000
                                           0 0.0000000 0.0000000 0.0000000
S12 0.8432432 0.0000000 0.3497901 0.9203663
                                           0 0.9571190 1.0000000 0.0000000
S24 0.3012886 0.8730844 0.0000000 0.3468235
                                           0 0.2851947 0.0000000 0.5461307
S25 0.0000000 0.0000000 0.8646999 0.2800404
                                           0 0.3096778 0.5115566 0.0000000
S10 0.0000000 0.0000000 0.3407545 0.0000000
                                           0 0.9569047 1.0000000 0.0000000
```

```
0.8646999 0.3407545 0.0000000 0.9148164
S17 0.2800404 0.0000000 0.9148164 0.0000000
S19 0.3096778 0.9569047 1.0000000 0.4047748
S16 0.5115566 1.0000000 0.0000000 0.4660540
  0.0000000 0.0000000 0.0000000 0.0000000
S3
  0.5536757 0.0000000 0.0000000 0.0000000
  0.0000000 0.0000000 0.0000000 0.0000000
S23 0.6077532 0.0000000 0.0000000 0.5806987
S1
                 S23
                       S11
      S3
0.0000000 0.0000000 0.0000000 0.0000000
S22 1.0000000 0.0000000 0.0000000 0.0000000
  0.0000000 0.0000000 0.0000000 0.0000000
S5
  0.0000000 0.0000000 0.0000000 0.0000000
  0.0000000 0.0000000 0.0000000 0.0000000
S25 0.5536757 0.0000000 0.6077532 0.0000000
```

0 1.0000000 0.0000000 0.0000000 0 0.4047748 0.4660540 0.0000000 0 0.0000000 0.0000000 0.0000000 0 0.0000000 0.0000000 0.5359016 0 0.0000000 0.0000000 0.5548607 0 0.5359016 0.5548607 0.0000000 0 0.0000000 0.4039530 0.3816155 0 0.0000000 0.5697954 0.0000000 0 0.0000000 0.3753787 0.0000000 0 0.0000000 0.3845681 0.5160511

S11 0.4494444 0.0000000 0.6235711 0.0000000

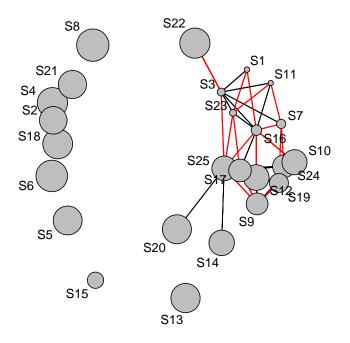
Once this network has been generated, we can now plot. ComGenR provides an easy to use function built on the gplot function in the sna package:

> mgp(net,com.,displaylabels=TRUE)

x y

- [1,] 38.81950 22.02641
- [2,] 41.55542 29.73309
- [3,] 49.52017 29.88970
- [4,] 38.35226 19.52846
- [5,] 48.78940 10.00295
- [6,] 39.59314 16.05584
- [7,] 38.40427 25.24226
- [8,] 48.11713 15.36902
- [9,] 39.96504 26.66322
- [10,] 38.46984 23.86893

- [11,] 51.60774 14.30781
- [12,] 54.30462 19.40306
- [13,] 56.46616 20.28039
- [14,] 51.80492 20.12208
- [15,] 57.28974 20.59832
- [16,] 54.37428 17.34724
- [17,] 53.03839 19.95993
- [18,] 41.76408 11.39318
- [19,] 56.07488 18.98300
- [20,] 54.31746 23.10148
- [21,] 56.25011 23.58857
- [22,] 51.57154 26.07460
- [23,] 53.58076 27.82475
- [24,] 52.49217 24.45522
- [25,] 55.43395 26.77914



5 A Template Analysis

To help guide the user, we present a template for using the package and how one might go about conducting an analysis on a dataset from a CG study.

```
> com <- com.sim[[1]][[5]][[7]]
> geno <- factor(trees[,1])</pre>
                                             #composition
> adonis(com~geno)
> nms <- nmds(vegdist(com),2,2,nits=3)</pre>
> my.nms <- nmds.min(nms)</pre>
> ch.plot(my.nms,g=geno,plot.legend=FALSE)
> top.ten <- com[,order(apply(com,2,sum),decreasing=TRUE)][,1:10]</pre>
> plot(envfit(my.nms,top.ten),add=TRUE,col='darkgrey')
                                             #heritability
>
> getH2C(com,geno)
>
                                             #networks
> net <- CoNetwork(com, threshold=20)</pre>
> mgp(net,com,displaylabels=TRUE)
> mgp(min.net(net,com)[[1]],min.net(net,com)[[2]],displaylabels=TRUE)
>
                                             #co-occurrence
> cnm.results <- cnm.test(com,nits=100,threshold=10)</pre>
> cnm.results
>
```

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