

# Testing for Homogeneity of Multivariate Dispersions Using Dissimilarity Measures

Irène Gijbels<sup>1,\*</sup> and Marek Omelka<sup>2,\*\*</sup>

<sup>1</sup>Department of Mathematics and Leuven Statistics Research Center (LStat), Katholieke Universiteit Leuven, Celestijnenlaan 200B, B-3001 Leuven (Heverlee), Belgium

<sup>2</sup>Department of Probability and Mathematical Statistics, Faculty of Mathematics and Physics, Charles University in Prague, Sokolovská 83, 186 75, Praha 8, Czech Republic

\**email:* Irene.Gijbels@wis.kuleuven.be

\*\**email:* omelka@karlin.mff.cuni.cz

**SUMMARY.** Testing homogeneity of dispersions may be of its own scientific interest as well as an important auxiliary step verifying assumptions of a main analysis. The problem is that many biological and ecological data are highly skewed and zero-inflated. Also the number of variables often exceeds the sample size. Thus data analysts often do not rely on parametric assumptions, but use a particular dissimilarity measure to calculate a matrix of pairwise differences. This matrix is then the basis for further statistical inference. Anderson (2006) proposed a distance-based test of homogeneity of multivariate dispersions for a one-way ANOVA design, for which a matrix of pairwise dissimilarities can be taken as an input. The key idea, like in Levene's test, is to replace each observation with its distance to an estimated group centre. In this paper we suggest an alternative approach that is based on the means of within-group distances and does not require group centre calculations to obtain the test statistic. We show that this approach can have theoretical as well as practical advantages. A permutation procedure that gives type I error close to the prescribed value even in small samples is described.

**KEY WORDS:** ANOVA; dissimilarity measure; Levene's test; multivariate analysis; permutation tests; principal coordinates; spatial median; U-statistics.

## 1. Introduction

When analysing multivariate data it is often of crucial importance to know if groups of observations (e.g., as defined by different treatments) differ in their relative dispersions. This question may be of interest on its own (e.g., to find the treatments having stabilising/destabilising effects) or an auxiliary step when verifying assumptions or interpreting results of a main analysis.

Data coming from many biological applications, in particular ecological community data, are often very skewed, possibly containing many zeroes, which makes the assumption of normality (as well as any other parametric assumptions) very difficult to justify. Also the number of variables is usually not small in comparison to the sample size. That is why an analyst often uses an appropriate dissimilarity (distance) measure to calculate the matrix of pairwise dissimilarities (distances) and the statistical inference is based on that matrix.

In this paper, we will consider the simple one-way ANOVA design, where each observation belongs to exactly one treatment. The usual question of scientific interest is to find out which treatments have effects on outcome. To answer this question several tests based on a dissimilarity matrix have been proposed, e.g., Mantel and Valand (1970), Mielke et al. (1981), Smith, Pontasch, and Cairns (1990), Excoffier, Smouse, and Quattro (1992), Clarke (1993), Pillar and Orlóci

(1996), Gower and Krzanowski (1999), Legendre and Anderson (1999), and McArdle and Anderson (2001). Roughly speaking, all the above tests are based on comparing the within-group against between-group dissimilarities and in particular they aim at finding the differences in centres of the multivariate distributions underlying the observed data. A significant result is usually interpreted as the tendency of the observations belonging to the same treatment to 'cluster together' around different group centres. However this interpretation may not be correct as all the tests are to some extent also omnibus tests and a significant result may be purely the effect of differences in group dispersions.

Anderson (2006) developed a distance-based test for homogeneity of multivariate dispersions which is inspired by the popular Levene's test in univariate ANOVA. The key idea is to replace each observation with its distance to an estimated group centre.

The aim of this paper is to suggest an alternative test, which is directly based on the means of distances within the same group and does not require group centre calculations to obtain the test statistic. This test overcomes several difficulties of the test of Anderson (2006). First, it can be easily and explicitly stated in terms of the distance matrix which feature of the groups is compared. Second, a large sample version of this test can be calculated for a general dissimilarity matrix

without the need for calculating the principal coordinate representation. Third, the permutation version of the test does not require re-calculation of the centre of the data with each permutation. Last but not least, our simulation experiences show that the proposed tests control slightly better the type I error for comparisons of small samples (up to 20 observations per group).

Note that in ecology the concept of dispersion can be very useful when analysing species diversity. If one observation represents the composition of species for a given site and observations are coming from several locations, then testing the homogeneity of dispersions of the groups is useful when testing for differences between locations in terms of species composition, because differences between locations (beta diversity) depend on dispersions within locations (alpha diversity). Observations can also be compared between years instead of locations. On the other hand, if an observation stands for characteristics of an individual (animals, plants, ...) and the observations are coming from different species (or even well defined groups within a given species), then the presented testing problem can be used in the analysis of genetic or phenotypic differences between species' populations.

With no loss of generality, we talk only about dissimilarity matrices in this paper, with the understanding that similarity matrices can be handled in a similar fashion, after applying a suitable transformation to dissimilarities.

The paper is organized as follows. Section 2 introduces the tests. Permutation procedures to estimate  $p$ -values of the tests are discussed in Section 3. In Section 4, the suggested tests are compared with the tests of Anderson (2006) in a simulation study. Section 5 summarizes and discusses the results. Detailed simulation results are provided in a Web Appendix.

## 2. Description of the Test

In a simple one-way ANOVA design each observation is associated with exactly one of the  $K$  treatments and the sample sizes of the corresponding groups are  $n_1, \dots, n_K$  with the total sample size  $n = n_1 + \dots + n_K$ . Let  $\mathbf{Y}_i^{(k)} = (Y_{i1}^{(k)}, \dots, Y_{ip}^{(k)})^\top$  be the vector of length  $p$  corresponding to the  $i$ th observation in the  $k$ th group for each  $p$  variables and suppose that all the observation vectors are independent and the  $p$ -length observation vectors in the same group follow the same multivariate distribution.

To describe the tests we use two of the data sets discussed in Anderson (2006). We refer to that paper and the references therein for a more detailed description of the data.

### 2.1 Using the Euclidean Distance

The Bumpus' sparrow data set consists of five morphological characteristics of sparrows measured in Rhode Island after a severe storm. The sparrows are divided into two groups, those that survived the storm and those that died. The data are a subset of the original data recorded by Bumpus (1899) and can be found in Manly (2005).

As the general theory of stabilizing selection (Campbell et al., 2008) suggests that the characteristics of nonsurvivors should be more dispersed than the characteristics of survivors, one of the questions of interest is to test for a difference in the multivariate dispersions of these two groups. Thus, we require a test for homogeneity of multivariate dispersions.

The first option is the traditional likelihood ratio test described, e.g., in Rencher (1998, pp. 138–140). This test assumes multivariate normality of the observations and tests the null hypothesis

$$H_0 : \Sigma_1 = \dots = \Sigma_K, \quad (1)$$

where  $\Sigma_k$  is the variance–covariance matrix of random variables underlying the observations in the  $k$ th group (note that  $K = 2$  in our example). As the resulting test is rather sensitive to the assumption of multivariate normality, more robust procedures have been developed, see e.g., Tiku and Balakrishnan (1985) and O'Brien (1992). In the latter paper it was also suggested that, instead of trying to test the very specific hypothesis (1), it is often reasonable to concentrate on the overall level of dispersion. This basically means that one constructs a simple measure that aims at summarizing the dispersion of each group and then tests for the equality of these measures of dispersion among groups.

The test suggested in Anderson (2006) follows the idea of concentrating on the overall level of dispersion. Although the test can be based on any dissimilarity measure, it is instructive to illustrate it with the Euclidean distance  $d_E$ . The core idea is that if group  $A$  is more dispersed than another group  $B$ , then the distances of the observations to the centre in group  $A$  tend to be larger than those in group  $B$ . To be more precise, let  $\hat{\mathbf{t}}_k$  stand for an estimated centre of the  $k$ th group and define

$$\mathbf{X} = (X_1^{(1)}, \dots, X_{n_1}^{(1)}, X_1^{(2)}, \dots, X_{n_2}^{(2)}, \dots, X_1^{(K)}, \dots, X_{n_K}^{(K)})^\top,$$

where  $X_i^{(k)} = d_E(\mathbf{Y}_i^{(k)}, \hat{\mathbf{t}}_k)$ .

Now, the vector  $\mathbf{X}$  is treated as  $K$  independent random samples and the traditional ANOVA  $F$ -statistic is used to compare the means across  $K$  groups.

A  $p$ -values for this  $F$ -statistic is obtained either by using an  $F$ -distribution or by using the following permutation procedure: (i) permute the “residuals”  $\mathbf{r}_i^{(k)} = \mathbf{Y}_i^{(k)} - \hat{\mathbf{t}}_k$ ; (ii) calculate the new group centres  $\hat{\mathbf{t}}_k^*$  based on the permuted residuals; (iii) take the distances of the permuted data from the new centres and recalculate the  $F$ -statistic.

Anderson (2006) suggested that either centroids (component-wise means) or spatial medians can be taken as the group centres ( $\hat{\mathbf{t}}_k$ 's). As there are many definitions of a multivariate median in the literature, by a spatial median of data-points in this paper we understand a point (say  $\hat{\mathbf{t}}$ ) that minimizes the sum of the Euclidean distances of data-points from  $\hat{\mathbf{t}}$  (Haldane, 1948).

The test  $F_{\text{And}}$  is very appealing as it seems to be a very natural multivariate analogue to Levene's test (see Van Valen (1978) and Manly (2005) for similar suggestions of a multivariate Levene's test). The test is also intuitive and simple to understand.

Despite these nice properties of the test  $F_{\text{And}}$ , one may feel uncomfortable that the test statistic depends on the choice of the centre of the groups. This could be of particular concern if a centroid and a spatial median are not close to each other which is often the case if the data are asymmetric or contain outliers. A straightforward alternative to calculating distances from group centres is to consider interpoint distances within groups. If for example the group of nonsurvivors is

more dispersed than the group of survivors, then one would expect that the interpoint distances within the group of non-survivors are, on average, larger than those within the group of survivors.

Let us make these ideas more precise. Let

$$d_{ij}^{(k)} = d(\mathbf{Y}_i^{(k)}, \mathbf{Y}_j^{(k)}),$$

where  $d$  stands for the used dissimilarity measure (the Euclidean distance in our example), be the dissimilarity between observations  $i$  and  $j$ . Means of the within-group distances can be calculated as

$$\bar{d}_k = \frac{1}{\binom{n_k}{2}} \sum_{i=1}^{n_k-1} \sum_{j=i+1}^{n_k} d_{ij}^{(k)}, \quad k = 1, \dots, K. \quad (2)$$

Note that if the dispersions in the groups are the same, the quantities  $\bar{d}_1, \dots, \bar{d}_K$  are expected to be close to each other. Thus, as in a standard ANOVA, we want to test the equality of these  $K$  means. **The only thing we have to be careful about is that each of the  $\bar{d}_k$  is not a mean of independent random variables, but rather is a  $U$ -statistic of degree two (see chapter 5 of Serfling, 1980).** The asymptotic variance of  $\bar{d}_k$  is usually estimated with the jackknife estimator (formula (9) of Callaert and Veraverbeke, 1981)

$$\hat{\sigma}_k^2 = \frac{S_k^2}{n_k}, \text{ where } S_k^2 = \frac{4(n_k - 1)}{(n_k - 2)^2} \sum_{i=1}^{n_k} (\hat{D}_i^{(k)} - \bar{d}_k)^2, \quad (3)$$

$$k = 1, \dots, K.$$

where the average distance from observation  $i$  in group  $k$  to every other observation within that group, denoted by  $\hat{D}_i^{(k)}$ , is given by

$$\hat{D}_i^{(k)} = \frac{1}{n_k - 1} \sum_{j=1, j \neq i}^{n_k} d_{ij}^{(k)}, \quad i = 1, \dots, n_k, \quad k = 1, \dots, K. \quad (4)$$

Finally, the test statistic is given by

$$F_{\bar{d}} = \frac{\sum_{k=1}^K n_k (\bar{d}_k - \bar{d})^2}{(K - 1) \hat{\sigma}^2}, \quad (5)$$

where

$$\bar{d} = \frac{1}{K} \sum_{k=1}^K n_k \bar{d}_k \quad \text{and} \quad \hat{\sigma}^2 = \frac{\sum_{k=1}^K (n_k - 1) S_k^2}{n - K}.$$

The null hypothesis is rejected when  $F_{\bar{d}}$  exceeds the  $(1 - \alpha)$ -quantile of an  $F$ -distribution with  $K - 1$  and  $n - K$  degrees of freedom. The asymptotic validity of this test follows from independence and the asymptotic normality of each of the quantities  $\bar{d}_1, \dots, \bar{d}_K$ . In Section 3, we describe a permutation procedure that improves the small sample properties of this test.

**REMARK 1.** As pointed out by one of the referees, the test statistic  $F_{\bar{d}}$  can be easily calculated by the standard ANOVA  $F$ -test applied to jackknifed “pseudo-values” (Callaert and Veraverbeke, 1981). In our situation the jackknifed “pseudo-

values” are for  $k = 1, \dots, K$  given by

$$P_i^{(k)} = \frac{2}{n_k - 2} \sum_{j=1, j \neq i}^{n_k} d_{ij}^{(k)} - \frac{n_k}{n_k - 2} \bar{d}_k, \quad i = 1, \dots, n_k.$$

The fact that one does not need to specify the group centres has some methodological advantages. The main advantage is that one can easily specify the null hypothesis in terms of the pairwise distances. Let  $\mathcal{L}_k$  stand for the distribution of the within sample distances in the  $k$ th group. Although in the sequel we will be interested in the following null hypothesis

$$H_0 : \mathcal{L}_1 = \mathcal{L}_2 = \dots = \mathcal{L}_K, \quad (6)$$

different null hypotheses may be of interest. For instance a researcher may be interested in a very broad null hypothesis stating that only the mean values of the distributions  $\mathcal{L}_1, \mathcal{L}_2, \dots, \mathcal{L}_K$  coincide. The corresponding test could be constructed by a modification of an ANOVA test for unequal variances (see e.g., Volaufová, 2009, and references therein).

**REMARK 2.** Note that the test statistic  $F_{\bar{d}}$  defined in (5) is not the only way to test the null hypothesis (6). When using  $F_{\bar{d}}$  one hopes that a difference in distributions  $\mathcal{L}_1, \mathcal{L}_2, \dots, \mathcal{L}_K$  can be discovered as a difference in their mean values. But generally speaking one can take any  $K$ -sample test and modify it for the within sample distances.

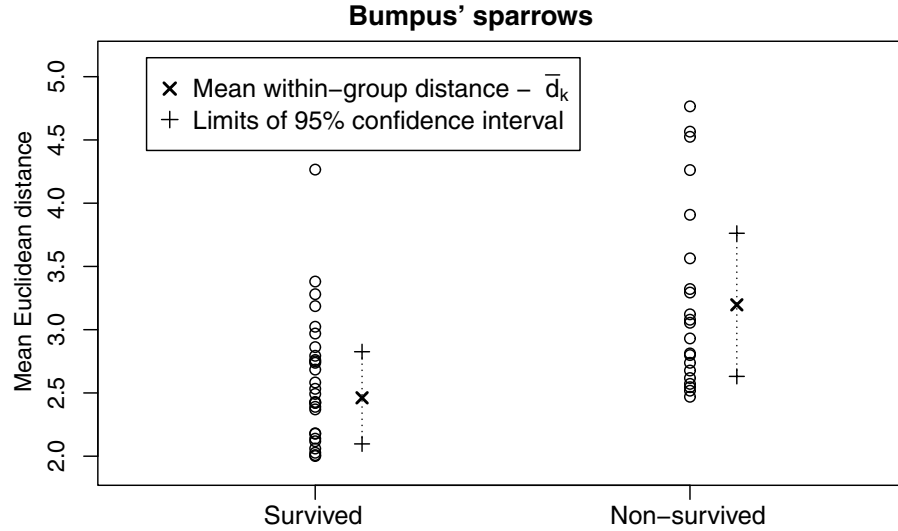
**REMARK 3.** As in fact we test for equality of dispersions, it seems reasonable to expect that the variances of the distributions  $\mathcal{L}_1, \mathcal{L}_2, \dots, \mathcal{L}_K$  increase with the means. Thus one can use a logarithmic transformation of  $\bar{d}_k$  to stabilize the variances. By the delta method (see e.g., chapter 3.1 of Serfling, 1980) it follows that the resulting test statistic  $F_{\bar{d}}^{\log}$  is given by (5) with  $\bar{d}_k$  replaced with  $\log(\bar{d}_k)$  and  $S_k^2$  with  $S_k^2/(\bar{d}_k)^2$ .

**REMARK 4.** The test procedure can be visualized with the help of  $\hat{D}_i^{(k)}$  defined in (4). This quantity gives an average distance from the  $i$ th observation to the other data points in the  $k$ th group. A small/large value of  $\hat{D}_i^{(k)}$  means that the  $i$ th observation is close/far from the “centre” of the  $k$ -group. Further note that the mean within-group distance  $\bar{d}_k$  defined in (2) can also be calculated as  $\bar{d}_k = \frac{1}{n_k} \sum_{i=1}^{n_k} \hat{D}_i^{(k)}$ . Figure 1 plots  $\hat{D}_i^{(k)}$  together with their means  $\bar{d}_k$  and an approximate 95% confidence interval for the mean  $\mu_k$  of the distributions  $\mathcal{L}_k$ . The confidence interval is based on the asymptotic normality of  $\bar{d}_k$  and is given by

$$(\bar{d}_k - u_{0.975} \hat{\sigma}_k, \bar{d}_k + u_{0.975} \hat{\sigma}_k),$$

where  $u_p$  is the  $p$ -quantile of the standard normal distribution and  $\hat{\sigma}_k$  is the estimate of the standard derivation of  $\bar{d}_k$  given in (3). Note that nonoverlapping confidence intervals in Figure 1 would already indicate that the null hypothesis (6) does not hold. As for our data there is a degree of overlap in the intervals, a formal test is necessary to decide about the null hypothesis.

Table 1 gives the values of the test statistics and  $p$ -values for the sparrow data set (after standardization for each variable).  $F_{\bar{d}}$  stands for the test (5) and  $F_{\bar{d}}^{\log}$  for the



**Figure 1.** Plot of the pseudo-observations  $\hat{D}_i^{(k)}$  for Bumpus' sparrow data set along with mean within-group distance and 95% confidence interval.

**Table 1**

Test statistics and  $p$ -values for the Bumpus' sparrow (*Sp.*) and Tikus Islands coral data set (*Co.*).

		$F_d^c$	$F_d^{\log}$	$F_{\text{And}}^c$	$F_{\text{And}}^m$
Sp.	statist.	4.319	4.956	3.869	3.818
	pval <sub>as</sub>	0.043	0.031	0.055	0.057
	pval <sub>perm</sub>	0.041	0.034	0.055	0.048
	statist.	6.920	5.193	9.097	6.292
Co.	pval <sub>as</sub>	<0.0001	0.0006	<0.0001	0.0001
	pval <sub>perm</sub>	<0.0001	0.0004	<0.0001	0.0001

“log-transformed” test described in Remark 3. The test statistics of Anderson (2006) are denoted by  $F_{\text{And}}^c$  when centred by a centroid or  $F_{\text{And}}^m$  when centred by a spatial median. Further, “pval<sub>as</sub>” stands for the  $p$ -values given by a standard  $F$ -distribution and “pval<sub>perm</sub>” for the  $p$ -values given by the permutation test with 99 999 permutations. The permutation procedure used to get the  $p$ -values  $F_d^{\log}(p)$  and  $F_d^{\log}(p)$  is described in Section 3.

Note that the  $p$ -values of all the tests are “borderline,” i.e., close to 0.05 indicating a possible difference between the two groups of sparrows in the dispersions of these measured morphological characteristics.

## 2.2 Using a General Dissimilarity Measure

The Tikus Island coral data set consists of the percentage cover of each 75 coral species along each of 10 replicate transects in six different years from 1981 to 1988. Differences among the coral assemblages in different years are expected as an El Niño event occurred in 1982–1983. The data are given as a data set called `tikus` in the R-package `mvabund` (Wang et al., 2012). As these data include many zeroes and are highly skewed, the approach that uses the Euclidean distance does not seem to be appropriate here. Following the analysis of Warwick, Clarke, and Suharsono (1990) the matrix of pair-

wise Bray–Curtis dissimilarities (given by (8)) calculated from square-root transformed data was used as a starting point of the analysis.

To be more precise, let  $\mathbf{Y}_i^{(k)}$  stand for already square-root transformed observations. Denote the joint sample as

$$(\mathbf{Z}_1, \dots, \mathbf{Z}_n)^T = (\mathbf{Y}_1^{(1)}, \dots, \mathbf{Y}_{n_1}^{(1)}, \mathbf{Y}_1^{(2)}, \dots, \mathbf{Y}_{n_2}^{(2)}, \dots, \mathbf{Y}_1^{(K)}, \dots, \mathbf{Y}_{n_K}^{(K)})^T. \quad (7)$$

The dissimilarity matrix  $\mathbb{D}$  is a matrix of pair-wise distances with the elements  $d_{ij} = d(\mathbf{Z}_i, \mathbf{Z}_j)$ , where  $d$  now stands for the Bray–Curtis dissimilarity given by

$$d_{BC}(\mathbf{Z}_i, \mathbf{Z}_j) = \frac{\sum_{q=1}^p |Z_{iq} - Z_{jq}|}{\sum_{q=1}^p |Z_{iq} + Z_{jq}|}. \quad (8)$$

For the situation when the analyst starts with a matrix  $\mathbb{D}$  of pairwise-dissimilarities Anderson (2006) suggests the following way of calculating the test  $F_{\text{And}}$ :

1. Using principal coordinate analysis (PCoA, see e.g., Legendre and Legendre, 1998, pp. 424–438) find a representation (say  $\mathbf{U}$ ) of the dissimilarity matrix  $\mathbb{D}$ . The observation  $\mathbf{Z}_i$  is now represented by the vector  $\mathbf{u}_i = (\mathbf{u}_i^+, \mathbf{u}_i^-)^T$  (the  $i$ th row of the matrix  $\mathbf{U}$ ), where  $\mathbf{u}_i^+$  ( $\mathbf{u}_i^-$ ) stands for the coordinates of the vectors that corresponds to real (imaginary) axes of the representation  $\mathbf{U}$ .
2. If  $\mathbf{U}$  contains only real axes, the analysis of Section 2.1 can be directly used with the original data being replaced with the representation  $\mathbf{U}$ . If there are also some imaginary axes, then define

$$X_i^{(k)} = \sqrt{d_E^2(\mathbf{u}_i^+, \hat{\mathbf{t}}_k^+) - d_E^2(\mathbf{u}_i^-, \hat{\mathbf{t}}_k^-)},$$

where  $\hat{\mathbf{t}}_k = (\hat{\mathbf{t}}_k^+, \hat{\mathbf{t}}_k^-)$  stands for the corresponding group centre (either a centroid or a spatial median) with  $\hat{\mathbf{t}}_k^+$

( $\hat{\mathbf{t}}_k^-$ ) being the coordinates corresponding to real (imaginary) axes.

3. Analogously as in Section 2.1 an  $F$ -statistic is computed and its significance is assessed either with the help of an  $F$ -distribution or via a permutation method where  $\mathbf{r}_i = \mathbf{u}_i - \hat{\mathbf{t}}_k$  are permuted.

Although the procedure seems to give reasonable results in practice, it is not without difficulties. Although one intuitively feels that the distances from the group centres in the PCoA representation should reflect the within-group variability, it is not at all straightforward to write down rigorously what feature of the original data is tested and how to formulate the null hypothesis. This is particularly true when the representation  $\mathbf{U}$  also includes imaginary axes.

The test statistics based directly on pairwise distances, suggested in this paper, enable an analyst to specify the null hypothesis by reference to the interpoint distances directly, and decide which feature of within-group distances to compare. For instance when using the statistic  $F_{\bar{d}}$  defined in (5) the analyst concentrates on comparing the means of within-sample dissimilarities.

Figure 2 presents the visualization of the test procedure and the  $p$ -values for the coral data set are given in Table 1. To estimate the  $p$ -values of the permutation tests, 99 999 random permutations were used. The results suggest that there is a statistically significant difference in the means of group dissimilarities. Already the visual inspection of Figure 2 reveals that multivariate dispersion, as measured by the Bray-Curtis dissimilarity, is significantly higher in 1983, which corresponds to the year of the El Niño event. Analogously as in Anderson (2006) one can now proceed and try to find which pairs of years are significantly different. The findings (not presented here) when using either  $F_{\bar{d}}$  or  $F_{\bar{d}}^{\log}$  are similar to the findings based on  $F_{\text{And}}$ .

### 2.3 Interpreting Results

It is worth mentioning that, as in O'Brien (1992) and Anderson (2006), the test statistics proposed in this paper concentrate on the overall level of dispersions as measured by the mean within-group dissimilarities. Thus, it might happen that the null hypothesis (6) is retained even if (1) is not true. Thus, for example, differences in rotations of the groups cannot be detected by the suggested tests when a rotation invariant dissimilarity measure is used. But this can be viewed also as a desirable feature of the test when analysing beta diversity (see Section 5).

Generally speaking, when using distance-based tests one should be careful when interpreting the results. In our case, rejecting the null hypothesis (6) says that the distributions  $\mathcal{L}_1, \dots, \mathcal{L}_K$  differ in their means. This can be safely interpreted as the difference in multivariate dispersions if a location invariant dissimilarity measure is used (e.g., Euclidean or Manhattan) and the underlying distribution of the data can be considered to belong to a multivariate location-scale family. This seems to be reasonable for Bumpus' sparrow data set, but not for the coral data set. When one is dealing with data sets that represent a percentage coverage (such as the coral data set) or with abundance data, the interpretation of any distance-based test is difficult because of the intrinsic mean-

variance relationships inherent in the distribution of counts or species abundances (Warton, Wright, and Wang, 2012).

Further, one should be aware that the results of tests are usually strongly influenced by the choice of a dissimilarity measure and by transformation/normalization of the data. This can be viewed also as an advantage though. As argued and illustrated in Anderson, Ellingsen, and McArdle (2006), by using different dissimilarity measures together with different transformations of the data an analyst can explore various aspects of data. Rejecting the null hypothesis (6) however always indicates that there is a difference among groups. Differences in dispersion may or may not be detected by the traditional ANOSIM-type procedures (see e.g., Clarke, 1993; Anderson, 2001) as these techniques target, in particular, location differences.

### 3. Resampling Procedures

So far, the new proposed tests have been described where inferences rely only on asymptotic distributions of the test statistics. It is well known that finite-sample properties of these tests can often be improved through resampling procedures. Several resampling algorithms can be proven to be asymptotically valid for our problem. In this section we describe a modified permutation procedure that works very well in all situations we have encountered so far and that we recommend for general use.

#### 3.1 The Standard Permutation Procedure

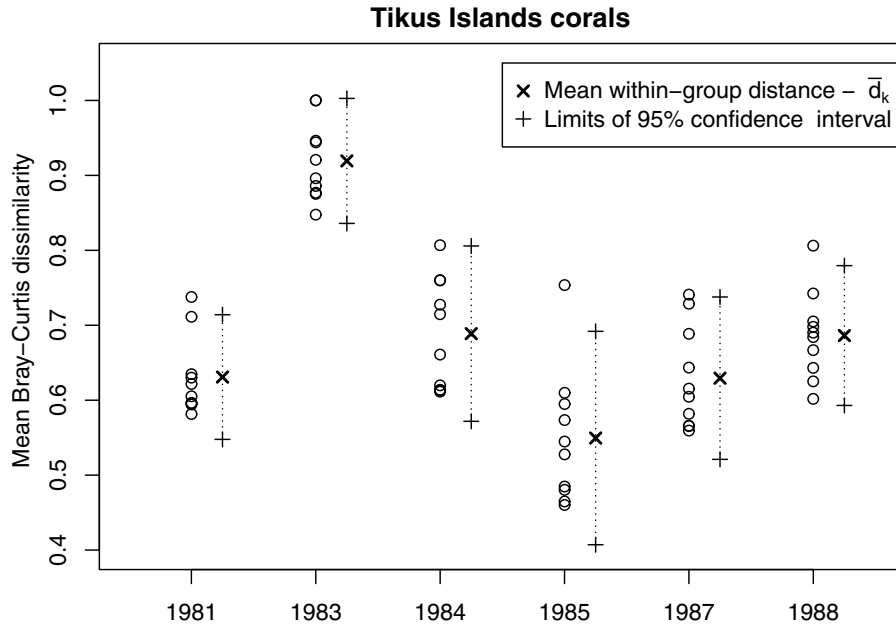
The standard permutation approach consists of permuting the original observations  $\mathbf{Z}_1, \dots, \mathbf{Z}_n$ . Note that this is equivalent to permuting both the rows and the columns of the matrix  $\mathbb{D}$ . But this method gives an exact permutation test only if the distribution of the original observations is the same in all the groups. But as the null hypothesis (6) allows (among others) for different locations of the groups, this method cannot be generally recommended. Our experience is that this standard permutation approach does not hold the type I error if the between-group dissimilarities are much bigger than the within group distances. That is why we recommend the "centred" permutation procedure described below.

#### 3.2 The "Centred" Permutation Procedure

Improvement of the properties of the standard resampling procedure can be obtained by reducing the distances among groups. To achieve this we make use of the principal coordinate representation (PCoA) of the distance matrix  $\mathbb{D}$ . In order to prevent the imaginary axes of the representation that raises difficulties in interpretation, we use the *Correction method 2* of Legendre and Legendre (1998, pp. 434–435). This method adds the smallest positive constant  $c$  to all nondiagonal elements of the matrix  $\mathbb{D}$ , such that the new matrix ( $\mathbb{D}^c$ ) has PCoA representation with only real axes. As proved by Cailliez (1983) the constant  $c$  can be found as the largest positive eigenvalue of the matrix

$$\begin{bmatrix} \mathbf{0} & 2\mathbf{\Delta}_1 \\ -\mathbf{I} & -4\mathbf{\Delta}_2 \end{bmatrix},$$

where  $\mathbf{0}$  is a  $n \times n$  null matrix and  $\mathbf{I}$  is a  $n \times n$  identity matrix. Let matrix  $\mathbf{A}$  of elements  $\{a_{ij}\}$  be defined element-wise as  $a_{ij} = -0.5 d_{ij}^2$ . Matrix  $\mathbf{\Delta}_1$  with elements  $\{\delta_{ij}\}$  is defined as:



**Figure 2.** Plot of the pseudo-observations  $\hat{D}_i^{(k)}$  for Tikus Island coral data set along with mean within-group distance and 95% confidence interval.

$\delta_{ij} = a_{ij} - \bar{a}_{i.} - \bar{a}_{.j} + \bar{a}_{..}$ , where  $\bar{a}_{i.}$ ,  $\bar{a}_{.j}$  and  $\bar{a}_{..}$  are the means for row  $i$ , column  $j$  and the overall mean, respectively, from matrix  $\mathbf{A}$ . Matrix  $\mathbf{\Delta}_2$  is defined in precisely the same way, but where matrix  $\mathbf{A}$  contains, instead, the elements  $a_{ij} = -0.5 d_{ij}$ .

The PCoA applied on the matrix  $\mathbb{D}^c$  gives a matrix  $\mathbf{U}$  such that for each  $i, j \in \{1, \dots, n\}$

$$d_{ij} + c = d_E(\mathbf{u}_i, \mathbf{u}_j), \quad \text{for } i \neq j,$$

where  $d_E$  stands for the Euclidean distance and  $\mathbf{u}_k$  is the  $k$ th row of the matrix  $\mathbf{U}$ .

Put  $\mathbf{U}_1$  for the first  $n_1$  rows of the matrix  $\mathbf{U}$ ,  $\mathbf{U}_2$  for the next  $n_2$  rows of the matrix  $\mathbf{U}$  and so on. Thus, one can write

$$\mathbf{U} = [\mathbf{U}_1^T, \mathbf{U}_2^T, \dots, \mathbf{U}_K^T]^T, \quad (9)$$

with  $\mathbf{U}_k$  being the representation for the  $k$ th sample. Now, put  $\tilde{\mathbf{U}}_k$  for  $\mathbf{U}_k$  centred with either a centroid or a group spatial median defined as

$$\hat{\mathbf{t}}_k = \arg \min_{\mathbf{t}} \sum_{i=1}^{n_k} d_E(\mathbf{u}_i^{(k)}, \mathbf{t}), \quad (10)$$

where  $\mathbf{u}_i^{(k)}$  is the  $i$ th row of the matrix  $\mathbf{U}_k$ . Replacing  $\mathbf{U}_k$  with  $\tilde{\mathbf{U}}_k$  in (9) one gets the ‘centred’ representation  $\tilde{\mathbf{U}}$ , that is used to construct the ‘centred’ distance matrix  $\tilde{\mathbb{D}}^c$  with the elements

$$\tilde{d}_{ij}^c = d_E(\tilde{\mathbf{u}}_i, \tilde{\mathbf{u}}_j).$$

Finally, remove the constant  $c$  from all the nondiagonal elements of the matrix  $\tilde{\mathbb{D}}^c$  and use the resulting matrix ( $\tilde{\mathbb{D}}$ ) for the standard permutation approach (instead of the matrix of the original distances  $\mathbb{D}$ ) of Section 3.1.

REMARK 5. Note that

$$\tilde{d}_{ij} = d_{ij} \quad \text{if } \mathbf{Z}_i \text{ and } \mathbf{Z}_j \text{ belong to the same group.}$$

Thus switching from  $\mathbb{D}$  to  $\tilde{\mathbb{D}}$  modifies only the dissimilarities between observations from different groups.

Further note for the test statistic  $F_{\tilde{d}}$  defined in (5) one can already resample the matrix  $\tilde{\mathbb{D}}^c$  as adding a constant to all distances does not affect the statistic  $F_{\tilde{d}}$ .

REMARK 6. Note that PCoA is used only as a vehicle to estimate a sampling distribution of the test statistic  $F_{\tilde{d}}$  ( $F_{\tilde{d}}^{\log}$ ) under the null hypothesis. In contrast with the test  $F_{\text{And}}$ , PCoA is not necessary to calculate the values of the test statistics. The user thus has the possibility to use the method of adding a constant to the original dissimilarity matrix to have a PCoA with only real axes. But for  $F_{\text{And}}$  the dissimilarity matrix  $\mathbb{D}^c$  gives generally a different value of the test statistic than when using the original distance matrix  $\mathbb{D}$ .

### 3.3 Centring the Original Data

The construction of the matrix  $\tilde{\mathbb{D}}$  described above may be summarized as

$$\text{DATA} \rightarrow \mathbb{D} \rightarrow \mathbb{D}^c \rightarrow \mathbf{U} \rightarrow \tilde{\mathbf{U}} \rightarrow \tilde{\mathbb{D}}^c \rightarrow \tilde{\mathbb{D}}.$$

Sometimes it seems even more natural to calculate the matrix  $\tilde{\mathbb{D}}$  from the original observations that are appropriately centred rather than with the help of PCoA.

This approach has basically two requirements. First, the centring of the observations has to be a reasonable operation. Note that this is true for the sparrow data set of Section 2.1, but not for the abundance-type of data of Section 2.2. Second, the chosen dissimilarity measure must be location-invariant, so that centring of the observations does not affect the within



sample distances. In this situation one can centre data either with a centroid or with a spatial median, where the latter is recommended if outliers are present in the data.

A general class of dissimilarity measures that are location invariant is generated by the *Minkowski distance* defined as

$$d_r(\mathbf{Z}_i, \mathbf{Z}_j) = \left( \sum_{q=1}^p |Z_{iq} - Z_{jq}|^r \right)^{1/r}, \quad \text{where } r > 0.$$

Note that for  $r = 2$  the Minkowski distance  $d_2$  reduces to the *Euclidean distance*  $d_E$ , and for  $r = 1$  the Minkowski distance results in the *Manhattan distance*.

Further, a number of dissimilarity measures are ‘Euclidean-transformable’, that is they coincide with the Euclidean distance after appropriate transformations of the data. Examples of such dissimilarity measures are the *Chord dissimilarity measure*, the *Chi-square distance*, the *Distance between species profiles* and the *Hellinger distance* (Legendre and Gallagher, 2001).

On the other hand, the distances that are usually used for abundance-type of data are not location invariant. Among others let us mention the *Canberra metric*, the *Coefficient of divergence*, the *Bray-Curtis dissimilarity* (Legendre and Legendre, 1998) or the *scale-invariant binomial deviance* (Anderson and Millar, 2004)

$$d_{bin}(\mathbf{Z}_i, \mathbf{Z}_j) = \sum_{q=1}^p \frac{1}{S_q} \left[ Z_{iq} \log \left( \frac{Z_{iq}}{S_q} \right) + Z_{jq} \log \left( \frac{Z_{jq}}{S_q} \right) - S_q \log \left( \frac{1}{2} \right) \right], \quad (11)$$

where  $S_q = Z_{iq} + Z_{jq}$ . For these dissimilarity measures, centring must be done using the PCoA representation, as described in Section 3.2.

#### 4. Simulation Study – Findings

In the simulation study we investigated the type I error and power properties of both asymptotic as well as permutation versions of the tests suggested in Section 2.1 and compared them with the performances of the  $F_{\text{And}}$ -tests.

The following test procedures were considered:

1.  $F_{\bar{d}}(as) - F_{\bar{d}}$  given by (5) +  $F$ -distribution;
2.  $F_{\bar{d}}(p_{med}) - F_{\bar{d}}$  + the “centred” (by median) permutation procedures of Section 3.2 or Section 3.3 (if the Euclidean distance is employed);
3.  $F_{\bar{d}}(p_{centr}) - F_{\bar{d}}$  + the “centred” permutation procedure as for  $F_{\bar{d}}(p_{centr})$  but with centring by a centroid instead of a spatial median
4.  $F_{\bar{d}}^{\log}(as) - F_{\bar{d}}^{\log}$  of Remark 3 +  $F$ -distribution;
5.  $F_{\bar{d}}^{\log}(p_{med}) - F_{\bar{d}}^{\log}$  + the “centred” permutation procedure as for  $F_{\bar{d}}^{\log}(p_{med})$
6.  $F_{\bar{d}}^{\log}(p_{centr}) - F_{\bar{d}}^{\log}$  + the “centred” permutation procedure as for  $F_{\bar{d}}^{\log}(p_{centr})$
7.  $F_{\text{And}}(as_{centr})$  – test of Anderson (2006) + centroid +  $F$ -distribution;
8.  $F_{\text{And}}(p_{centr})$  – test of Anderson (2006) + centroid + permutation;
9.  $F_{\text{And}}(as_{med})$  – test of Anderson (2006) + spatial median +  $F$ -distribution;

10.  $F_{\text{And}}(p_{med})$  – test of Anderson (2006) + spatial median + permutation.

#### 4.1 The Models Used to Generate Data

We considered four types of data generation processes. Detailed descriptions of these data generations are given in the Web Appendix.

1. *Sparrows type data* – This data generation process is inspired by the data set of Section 2.1. The simulated data came from a five-dimensional normal distribution and the Euclidean distance  $d_E$  was used.
2. *Fish type data* – This data generation example is inspired by the data coming from a study on spatial variation in temperate reef fish assemblages (Anderson and Millar, 2004). In this data set each observation records abundance of 57 fish species. In this model the samples were simulated from a multivariate Poisson-lognormal distribution (Aitchison and Ho, 1989). As a dissimilarity measure we used the scale-invariant binomial deviance  $d_{bin}$  (11).
3. *Corals type data* – This data generation process is inspired by the Tikus Islands coral data discussed in Section 2.2. In this data set each observation records the percentage cover of 75 coral species. In our simulation model these covers were generated as a mixture of independent normal distributions and zeroes. Here the Bray-Curtis dissimilarity measure  $d_{BC}$  (8) was used.
4. *Gaussian data with outliers* – Here the data were generated from a bivariate normal distribution with 10% of outliers. The Euclidean distance  $d_E$  was used.

The detailed results of the simulation study are given in the Web Appendix. Here we limit ourselves to describing and discussing simulation findings.

#### 4.2 An $F$ -Distribution or a Permutation Test?

Generally we recommend to use permutation tests whenever it is computationally feasible. The tests  $F_{\bar{d}}(as)$  and  $F_{\bar{d}}^{\log}(as)$  tend to be conservative in particular for *fish data* and *coral data*. (Web Tables 2 and 6). Note that  $F_{\bar{d}}^{\log}(as)$  is rather unreliable in terms of type I error when outliers were present and the Euclidean distance was used (Web Table 8).  $F_{\text{And}}(as_{centr})$  tends to exceed the level slightly in balanced samples and considerably in unbalanced samples in all simulation settings. Finally, the test  $F_{\text{And}}(as_{med})$  is usually conservative in balanced samples, but sometimes exceeds the level in unbalanced samples (Web Table 2).

The  $F$ -distribution approximation of critical values seems to work reasonably well when the group sample sizes exceed 50, but one should be aware that even for very large sample sizes  $F_{\text{And}}(as_{centr})$  can exceed the level by about one percent, and  $F_{\text{And}}(as_{med})$  by about a half percent (Web Table 4). The tests  $F_{\bar{d}}(as)$  and  $F_{\bar{d}}^{\log}(as)$  seem to work better in this aspect.

#### 4.3 Centring by a Centroid or a Spatial Median?

Not surprisingly, centring by a spatial median is preferred when there are outlying observations. Particularly in small samples even the permutation test  $F_{\text{And}}(p_{centr})$  is not reliable in terms of type I error. This can be seen from Web Table 8 as well as from Web Table 2; indeed the *fish type* data generation also produced values that are outlying.

When there is no evidence for outlying values, the differences in methods are subtle.  $F_{\text{And}}(p_{\text{centr}})$  gives usually slightly higher power results than  $F_{\text{And}}(p_{\text{med}})$ , but in some models the former test exceeds the level for small and unbalanced samples (Web Tables 2, 3, 6 and 7).

The choice of centring seems to be of lesser importance for the permutation versions of the tests  $F_{\bar{d}}$  and  $F_{\bar{d}}^{\log}$  than for the  $F_{\text{And}}$ -tests. This can probably be explained by the fact that the type of centring does not affect the test statistics and it is used only as an adjustment for the data before resampling. The test  $F_{\bar{d}}(p_{\text{centr}})$  ( $F_{\bar{d}}^{\log}(p_{\text{centr}})$ ) usually achieves slightly higher power than its closest competitor  $F_{\bar{d}}(p_{\text{med}})$  ( $F_{\bar{d}}^{\log}(p_{\text{med}})$ ). The type I error properties of the tests  $F_{\bar{d}}(p_{\text{centr}})$  and  $F_{\bar{d}}^{\log}(p_{\text{centr}})$  are also very satisfactory and both the tests hold the level very closely for sample sizes bigger than 10. For very small sample sizes, the tests can exceed the level slightly (but usually not more than by a half percent). On the other hand  $F_{\bar{d}}(p_{\text{med}})$  and  $F_{\bar{d}}^{\log}(p_{\text{med}})$  are sometimes unnecessarily conservative for small samples.

#### 4.4 Distance to Centres or Within Group Distances?

In most of the situations all the permutation tests hold the level quite satisfactorily. Only in small or unbalanced samples the  $F_{\text{And}}$ -tests slightly (usually with about a half or one percent) exceed the level (Web Tables 2 and 6). On the other hand all the permutation tests suggested in this paper hold the level very satisfactorily in all situations we encountered so far.

Regarding power, all the permutation tests give similar powers in balanced samples. This is not so surprising as the tests concentrate on very similar features of the data. For unbalanced samples, the suggested tests are more powerful when bigger groups tend to have smaller dispersions. If it is the other way around, then the tests of Anderson (2006) achieve higher power. This seems to be a small sample feature of the tests that is diminishing with increasing sample size (Web Table 5).

#### 4.5 $F_{\bar{d}}$ -test or $F_{\bar{d}}^{\log}$ -test?

In terms of type I error these two tests are comparable. Our experience is that also in terms of power performances these tests are very close when a scale-invariant dissimilarity measure is used, that is, a dissimilarity measure satisfying  $d(c\mathbf{Z}_i, c\mathbf{Z}_j) = d(\mathbf{Z}_i, \mathbf{Z}_j)$  for each  $c > 0$ . Note that  $d_{BC}$  defined in (8) as well as  $d_{bin}$  defined in (11) are scale-invariant. On the other hand a noticeable difference in the power performances of the tests can be observed for dissimilarity measures that are scale-equivariant, that is  $d(c\mathbf{Z}_i, c\mathbf{Z}_j) = c d(\mathbf{Z}_i, \mathbf{Z}_j)$ . This is the case for the Euclidean measure used for the sparrow-type data (Web Table 1). As the test statistic  $F_{\bar{d}}^{\log}$  transforms a scale effect into an additive effect and the  $F$ -statistic used in ANOVA is constructed to detect additive differences in mean values, we recommend to use  $F_{\bar{d}}^{\log}$  for scale-equivariant dissimilarity measures. However, one should be aware that  $F_{\bar{d}}^{\log}(as)$  can break down when outliers are present (Web Table 8).

#### 4.6 Computational Aspects

Sometimes it might be advantageous that the test statistics  $F_{\bar{d}}$  and  $F_{\bar{d}}^{\log}$  can be computed without the necessity of calculating a PCoA representation. The permutation versions of the

suggested tests also require a PCoA representation, thus from this point of view the suggested procedures are comparable with the tests of Anderson (2006). The amount of calculations needed to recalculate the test statistic is for the suggested test procedure comparable with that for  $F_{\text{And}}(p_{\text{centr}})$ . The test  $F_{\text{And}}(p_{\text{med}})$  is more computationally expensive when the total sample size  $n$  exceeds one hundred and not the Euclidean distance is used as a dissimilarity measure. The reason is that in this situation the PCoA representation has typically  $(n - 1)$  dimensions. Recalculating a spatial median in such a high dimension, with each permutation and in each group, can take a substantial amount of computing time.

## 5. Conclusions

In this work we propose a testing approach that aims at detecting differences in the overall level of dispersion among independent groups of observations. The approach is based on pairwise distances of observations. We compare the suggested tests with the tests introduced in Anderson (2006) from the computational aspect as well as from aspects of performances in power and type I error.

While the tests of Anderson (2006) summarize the overall measure of dispersion by means of the distances from the group centres, the tests proposed in this paper used the mean within group distances to summarize the dispersion. While these concepts are close together when the Euclidean distance is used, the difference becomes more important for other dissimilarity measures. The reason is that in this case tests of Anderson (2006) are based on the distances from the group centres in the PCoA representation and it is less obvious what feature of the original distance matrix is tested. This is in particular true when also imaginary axes are present in the PCoA representation. On the other hand, the approach based on distances from the centroids can be more useful when one also wants to identify outlying observations visually.

As suggested in Anderson et al. (2006) for the mean distance-to-centre, the mean within-group dissimilarity  $\bar{d}_k$  can also be used as a direct measure of beta diversity (e.g., Whittaker, 1960, 1972; Vellend et al., 2007). For example, if one analyses species data like the Tikus Island coral data set, then  $\bar{d}_k$  can be interpreted as a measure of beta diversity for a given year (e.g., Anderson et al., 2011).

The proposed new approach to test for homogeneity of overall dispersions among groups behaves just as well as the tests proposed by Anderson (2006) with respect to type I error and power, has reasonably good asymptotic properties, and is generally easier to compute and to interpret than the methods proposed by Anderson (2006).

## 6. Supplementary Materials

The data sets analysed in Section 2, an R-code implementing the considered tests and the Web Appendix referenced in Section 4 are available with this paper at the Biometrics website on Wiley Online Library.

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