PRIMER-e
empowering research

Presenter: Dr Paul Somerfield (Plymouth Marine Laboratory, UK)

Venue: The Laboratory of Marine Biotechnology, Institute of Bioscience (IBS), Universiti Putra Malaysia (UPM)

Date: 3rd – 7th December 2018

PROVISIONAL PROGRAMME

Monday, 3rd December

08:30-08:45	Introduction
08:45-10:15	Lecture: *Measures of resemblance (similarity/dissimilarity/distance) in multivariate structure for assemblage and environmental data, including shade plots used to assess the effects of pre-treatment options (e.g. standardisation, transformation, normalisation, dispersion weighting), and guidelines for different coefficient choices for different data types, and experimental purpose and protocols*
10:15-10:45	**Coffee break**
10:45-12:45	Lecture: *Clustering of samples by hierarchical agglomerative (CLUSTER) and unconstrained divisive (UNCTREE) methods; also non-hierarchical ('flat') clustering for specified numbers of groups. Includes discussion of a global test for the presence of any multivariate structure in <u>a priori</u> unstructured biotic or abiotic samples, using similarity profiles (Type 1 SIMPROF tests on samples)*
12:45-14:00	**Lunch break**
14:00-14:30	Introduction to PRIMER 7 software
14:30-15:45	Lab session on transforms, similarity options, alternative clustering methods and SIMPROF tests
15:45-16:00	**Coffee break**
16:00-16:45	Lecture: *Ordination (for environmental data) by Principal Components Analysis (PCA)*
16:45-17:30	Lab session on ensuring participants can structure their own data to take into PRIMER*

Tuesday, 4th December

08:30-09:30	Lab session on ordination by PCA
09:30-11:30	Lecture: *Ordination (of assemblage data) by non-metric Multi-Dimensional Scaling (nMDS,) and MDS diagnostics (e.g. stress, MST, cluster overlay) for adequacy of low-d representation. Also how this relates, through the Shepard diagram, to metric MDS (mMDS), useful for abiotic data (Euclidean distances) and for means plots from biotic data with few points* (**includes coffee break c. 10:15-10:45**)
11:30-12:45	Lab session on ordination by nMDS (and mMDS), animating the convergence, examining Shepard plots and other diagnostics, and display tools on MDS plots
12:45-14:00	**Lunch break**
14:00-14:45	Continuation of the lab session on MDS
14:45-15:45	Lecture: *Global hypothesis tests of no agreement between two resemblance matrices (RELATE), comparing assemblage (or environmental) structure with linear or cyclic models in space and time*
15:45-16:00	**Coffee break**
16:00-17:30	Lab session on RELATE tests for simple seriation without replication and a cyclic model with replication, including 'fixing' MDS collapse and dispersion weighting

Wednesday, 5th December

08:30-09:30	Lecture: *Multivariate testing for differences among <u>a priori</u> specified groups of samples (1-way ANOSIM and pairwise). Multivariate means plots where global test has established differences. Brief discussion of approximate region estimates for means, in mMDS plots, using bootstrap averaging.*
09:30-10:15	Lab session on 1-way ANOSIM, means plots and mMDS of region plots for means

10:15-10:45	**Coffee break**
10:45-11:45	Lecture: *Ordered ANOSIM tests, 2- and 3-way ANOSIM designs and testing (unordered) factors in crossed ANOSIM designs without replication, by comparing patterns*
11:45-12:45	Lab session on ordered and multi-way ANOSIM
12:45-14:00	**Lunch break**
14:00-15:15	Lecture: *Linking potential environmental drivers to an observed assemblage pattern, via the matching of multivariate structures (the BEST procedure). Test of no evidence for a biota-environment link, allowing for selection effects in finding an optimum match (global BEST test)*
15:15-16:30	Lab session on PCA and the BEST routine for linking to environmental variables, and global BEST test **(includes coffee break c. 15:45-16:00)**
16:30-17:00	Lecture: *Linkage trees – a further technique for 'explaining' assemblage patterns by environmental variables (LINKTREE), and its relation to unconstrained divisive clustering (UNCTREE)*
17:00-17:30	Lab session on link to abiotic variables (LINKTREE) and comparison to unconstrained cluster

Thursday, 6th December

08:30-09:30	Lecture: *Species contributions to sample patterns: stepwise form of BEST to identify minimal-sized species subsets reconstructing the full assemblage pattern (a whole pattern approach), and species contributions to similarities (SIMPER, a pairwise approach for statistically established groups)*
09:30-10:15	Lab session on various methods for identifying species contributions (Matrix display, stepwise BEST, bubble plots and SIMPER – the latter on groups from 'flat' clustering)
10:15-10:45	**Coffee break**
10:45-11:30	Lecture: *Direct analysis of species (or other variables) through species resemblances: techniques for identifying coherent groups of species (or other variables) in their response across samples*
11:30-12:15	Lab session on coherent variable sets
12:15-14:00	**Lunch break**
14:00-14:45	Lecture: *Diversity measures (DIVERSE) and multivariate treatment of multiple indices; dominance plots and testing differences between sets of curves (e.g. DOMDIS)*
14:45-15:45	Lab session on DIVERSE, dominance plots, and multivariate analyses of multiple diversity indices
15:45-16:00	**Coffee break**
16:00-17:00	Lecture: *Taxonomic (or phylogenetic) diversity and distinctness for quantitative data, or simple species lists, as valid biodiversity measures (DIVERSE) over broad spatial and temporal scales; sampling properties and testing structures (TAXDTEST)*
17:00-17:30	Lab session on TAXDTEST

Friday, 7th December

08:30-09:30	Lecture: *Second-stage analysis (2STAGE) to compare taxonomic levels and transformation etc; also for a possible testing framework in some repeated measures designs*
09:30-10:15	Lab session on 2STAGE
10:15-10:45	**Coffee break**
10:45-11:45	Lecture: *Any methods that have not arisen in earlier discussion (e.g. further resemblance options: modifying Bray-Curtis for denuded samples; resemblance calculations when some data are missing; perhaps dissimilarity measures based on taxonomic distinctness, etc.)*
11:45-12:15	Wrap up of formal programme – brief introduction to PERMANOVA+
12:15-14:45	**Lunch break**
14:45-17:30	Main lab session on analysing own data using PRIMER*

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* Throughout, participants will be given real data sets to analyse, but they may also wish to bring their own data. These should be in numeric, rectangular arrays, with variables (e.g. species) as rows, samples as columns, or vice-versa, in an Excel spreadsheet or text file. Non-numeric sets of information (factors) on each sample are placed below (or to the side of) this table, separated by a blank row (or blank column). There is also a 3-column format (sample label, variable label, non-zero entry) suitable for very large arrays. Participants should take the opportunities of all lab sessions and breaks to discuss their own analyses with the lecturer, and not leave *all* their questions until the final afternoon session!

Some key and well-cited papers on PRIMER and PERMANOVA+ methodology

PRIMER

Clarke KR (1990) Comparisons of dominance curves. *J Exp Mar Biol Ecol* 138: 143-157

Clarke KR (1993) Non-parametric multivariate analyses of changes in community structure. *Aust J Ecol* 18: 117-143

Clarke KR (1999) Non-metric multivariate analysis in community-level ecotoxicology. *Environ Toxicol Chem* 18: 118-127

Clarke KR, Ainsworth M (1993) A method of linking multivariate community structure to environmental variables. *Mar Ecol Prog Ser* 92: 205-219

Clarke KR, Chapman MG, Somerfield PJ, Needham HR (2006) Dispersion-based weighting of species counts in assemblage analyses. *Mar Ecol Prog Ser* 320: 11-27

Clarke KR, Gorley RN (2001, 2006, 2015) *PRIMER v5, v6, v7: User manual/tutorial.* PRIMER-E, Plymouth, UK, 91pp, 192pp, 296pp

Clarke KR, Green RH (1988) Statistical design and analysis for a 'biological effects' study. *Mar Ecol Prog Ser* 46: 213-226

Clarke KR, Somerfield PJ, Airoldi L, Warwick RM (2006) Exploring interactions by second-stage community analyses. *J Exp Mar Biol Ecol* 338: 179-192

Clarke KR, Somerfield PJ, Chapman MG (2006) On resemblance measures for ecological studies, including taxonomic dissimilarities and a zero-adjusted Bray-Curtis coefficient for denuded assemblages. *J Exp Mar Biol Ecol* 330: 55-80

Clarke KR, Somerfield PJ, Gorley RN (2008). Exploratory null hypothesis testing for community data: similarity profiles and biota-environment linkage. *J Exp Mar Biol Ecol* 366: 56-69

Clarke KR, Somerfield PJ, Gorley RN (2016) Clustering in non-parametric multivariate analyses. *J Exp Mar Biol Ecol* 483: 147-155.

Clarke KR, Tweedley JR, Valesini FJ (2014) Simple shade plots aid better long-term choices of data pre-treatment in multivariate assemblage studies. *J Mar Biol Assoc UK* 94: 1-16

Clarke KR, Warwick RM (1994, 2001, 2014) *Change in Marine Communities: An Approach to Statistical Analysis and Interpretation.* PRIMER-E, Plymouth, UK. *1st ed:* 144pp; *2nd ed:* 172pp. *3rd ed:* (authors: Clarke KR, Gorley RN, Somerfield PJ, Warwick RM) 260pp

Clarke KR, Warwick RM (1998) Quantifying structural redundancy in ecological communities. *Oecologia* 113: 278-289

Clarke KR, Warwick RM (1998) A taxonomic distinctness index and its statistical properties. *J Appl Ecol* 35: 523-531

Clarke KR, Warwick RM (2001) A further biodiversity index applicable to species lists: variation in taxonomic distinctness. *Mar Ecol Prog Ser* 216: 265-278

Field JG, Clarke KR, Warwick RM (1982) A practical strategy for analysing multispecies distribution patterns. *Mar Ecol Prog Ser* 8: 37-52

Somerfield PJ, Clarke KR (1995) Taxonomic levels, in marine community studies, revisited. *Mar Ecol Prog Ser* 127: 113-119

Somerfield PJ, Clarke KR (2013) Inverse analysis in non-parametric multivariate analyses: distinguishing groups of associated species which covary coherently across samples. *J Exp Mar Biol Ecol* 449: 261-273

Somerfield PJ, Clarke KR, Olsgard F (2002) A comparison of the power of categorical and correlational tests applied to community ecology data from gradient studies. *J Anim Ecol* 71: 581-593

Warwick RM, Clarke KR (1991) A comparison of some methods for analysing changes in benthic community structure. *J Mar Biol Ass UK* 71: 225-244

Warwick RM, Clarke KR (1993) Increased variability as a symptom of stress in marine communities. *J Exp Mar Biol Ecol* 172: 215-226

Warwick RM, Clarke KR (1995) New 'biodiversity' measures reveal a decrease in taxonomic distinctness with increasing stress. *Mar Ecol Prog Ser* 129: 301-305

Warwick RM, Clarke KR (1998) Taxonomic distinctness and environmental assessment. *J appl Ecol* 35: 532-543

Warwick RM, Clarke KR (2001) Practical measures of marine biodiversity based on relatedness of species. *Oceanog Mar Biol Ann Rev* 39: 207-231

PERMANOVA+

Anderson MJ (2001) A new method for non-parametric multivariate analysis of variance. *Austral Ecol* 26: 32-46

Anderson MJ (2001) Permutation tests for univariate or multivariate analysis of variance and regression. *Can J Fish Aquat Sci* 58: 626-639

Anderson MJ (2006) Distance-based tests for homogeneity of multivariate dispersions. *Biometrics* 62: 245-253

Anderson MJ (2008) Animal-sediment relationships revisited: characterising species' distributions along an environmental gradient using canonical analysis and quantile regression splines. *J Exp Mar Biol Ecol* 366: 16-27

Anderson MJ, Crist TO, Chase JM, Vellend M, Inouye BD, Freestone AL, Sanders NJ, Cornell HV, Comita LS, Davies KF, Harrison SP, Kraft NJB, Stegen JC, Swenson NG (2011) Navigating the multiple meanings of b diversity: a roadmap for the practicing ecologist. *Ecol Lett* 14: 19-28.

Anderson MJ, Connell SD, Gillanders BM, Diebel CE, Blom WM, Landers TJ, Saunders JE (2005) Relationships between taxonomic resolution and spatial scales of multivariate variation in kelp holdfast assemblages. *J Anim Ecol* 74: 636-646

Anderson MJ, Diebel CE, Blom WM, Landers TJ (2005) Consistency and variation in kelp holdfast assemblages: spatial patterns of biodiversity for the major phyla at different taxonomic resolutions. *J Exp Mar Biol Ecol* 320: 35-56

Anderson MJ, Ellingsen KE, McArdle BH (2006) Multivariate dispersion as a measure of beta diversity. *Ecol Lett* 9: 683-693

Anderson MJ, Gorley RN, Clarke KR (2008) *PERMANOVA+ for PRIMER: Guide to Software and Statistical Methods*. PRIMER-E: Plymouth, UK, 214pp

Anderson MJ, Gribble NA (1998) Partitioning the variation among spatial, temporal and environmental components in a multivariate data set. *Aust J Ecol* 23: 158-167

Anderson MJ, Legendre P (1999) An empirical comparison of permutation methods for tests of partial regression coefficients in a linear model. *J Statist Comput Sim* 62: 271-303

Anderson MJ, Millar RB (2004) Spatial variation and effects of habitat on temperate reef fish assemblages in northeastern New Zealand. *J Exp Mar Biol Ecol* 305(2): 191-221

Anderson MJ, Robinson J (2003) Generalized discriminant analysis based on distances. *Aust NZ J Stat* 45: 301-318

Anderson MJ, Robinson J (2001) Permutation tests for linear models. *Aust NZ J Stat* 43: 75-88

Anderson MJ, Santana-Garcon J (2015) Measures of precision for dissimilarity-based multivariate analysis of ecological communities. *Ecol Lett* 18: 66-73.

Anderson MJ, ter Braak CJF (2003) Permutation tests for multi-factorial analysis of variance. *J Statist Comput Sim* 73: 85-113

Anderson MJ, Walsh DCI (2013) What null hypothesis are you testing? PERMANOVA, ANOSIM and the Mantel test in the face of heterogeneous dispersions. *Ecol Monogr* 83: 557-574.

Anderson MJ, Walsh DCI, Clarke KR, Gorley RN, Guerra-Castro E (2017) Some solutions to the multivariate Behrens-Fisher problem for dissimilarity-based analyses. *Aust NZ J Stat* 59: 57-79.

Anderson MJ, Willis TJ (2003) Canonical analysis of principal coordinates: a useful method of constrained ordination for ecology. *Ecology* 84: 511-525

Legendre P, Anderson MJ (1999) Distance-based redundancy analysis: testing multispecies responses in multifactorial ecological experiments. *Ecol Monogr* 69: 1-24

McArdle BH, Anderson MJ (2001) Fitting multivariate models to community data: a comment on distance-based redundancy analysis. *Ecology* 82: 290-297

Paul WL, Anderson MJ (2013) Causal modelling with multivariate species data. *J Exp Mar Biol Ecol* 448: 72-84.