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Hydrology and Earth System Sciences
Discussions

Interactive comment on "Interacting effects of climate and agriculture on fluvial DOM in temperate and subtropical catchments" *by* D. Graeber et al.

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article graphicx

Reply to the comments by Anonymous Referee 1

29 January 2015

1 Replies to the referee comments

1) Indicate sample sizes for statistical tests. No sample sizes are mentioned anywhere in the paper.

Reply: We thank the reviewer for raising our attention to the missing sample sizes. Please see the end of this document for an overview of the sample sizes. We will include the sample sizes at the appropriate positions of the revised manuscript.

2) It seems to me that after converting some data to ratios, the statistical analysis has proceeded in much the same way as it would have for the raw dataset. Some, but not all, variables were transformed to improve their distributions (relative to the assumption of normality), then ANOVA, PCA were performed. I However, methods for statistical analysis of compositional data /ratios are special due to the constraint that the data sum to one (closed data/constant sum constraint). There is a whole field of multivariate statistical analysis devoted to the analysis of compositional data, e.g. in the field of geology. There is an R

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package called composition, and several other packages, specifically directed at analysing compositional data (incl. imputing missing data). The logratio transformation is often used prior to linear modelling. See papers by J. Aitchison starting in the 1980s. Also a very readable R tutorial about the problem with ratios at http://advan.physiology.org/content/37/3/213.

Reply: The reviewer is right that the data from the PARAFAC and SEC were converted to ratios. However, no classical ANOVAs or linear models were applied in this study. Please see the points below for a description of the used test types and their relationship to ratio data:

1. We performed permutative MANOVAs (PERMANOVAs, often called ANOSIM or analysis of similarity) , a non-parametric alternative to MANOVAs and permutative multivariate tests of dispersal (PERMDISP), a multivariate non-parametric alternative to Levene's test (described in lines 238–247 of the submitted manuscript). According to the literature (Anderson 2001, see submitted manuscript for reference), no special treatment is needed for compositional data when using PERMANOVAs, since the data is converted into a dissimilarity matrix before applying the statistic (in our case Euclidean distances). Since the data for PERMDISPs is also translated into a dissimilarity matrix before the statistic is done, the same applies to PERMDISP. Both statistics can be based on ratio data or presence-absence data and on different dissimilarity indexes. We only transformed some of the data, to keep the same transformations as in the PCA and to make the PERMANOVAs / PERMDISPs representative for the data depicted in the PCA.

2. As stated in the first part of the reply, a part of the data for the PCA and PERMANOVAs / PERMDISPs was transformed prior to its application, to allow the application of the linear relationships on which a PCA is based. We agree with the reviewer that the commonly used log transformation is problematic with ratios with fixed limits $(0..1)$. Thus, we used logit transformations

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($logit(x) = -log(1/x - 1)$)for the three variables which were ratios with fixed limits and which had to be transformed to reach normal distribution: $HMWS_N$, $HMWS_C$, and HS_N and repeated the PERMANOVAs and PCA. No difference could be detected for the PERMANOVAs or PERMDISPs. Only very small differences could be detected for the PCA, most notably a change from 18.7 to 18.8% explained variance for the 4th PCA axis. To prove that no significant changes occurred, we show the PCA results from the submitted manuscript version (Fig.1 of supplement pdf) and the PCA results with the revised transformation (Fig.2 of supplement pdf) below. We will include the PCA based on the revised data transformations in the revised manuscript.

3. No linear models were applied in this study. Instead, we used Spearman rank correlations, but not for ratio data with fixed limits.

4. Differences in univariate data were assessed with permutative one-way statistics (often called Monte-Carlo tests). Hence, neither normal distribution nor homoscedacity was assumed for the data. This test works well with ratio data, since it has no assumption on the data probability distribution.

5. Levene's test based on the median was used to assess the variability of univariate data, but never for ratio data with fixed limits.

âĂČ **3) Currently, some data used in the PCA are bounded by (0..1) and some are not (e.g. fluorescence index) , but overall the dataset does not sum to 100% (as it would in a typical compositional dataset). This does not sound like a good situation for starting a PCA. A simple approach would be to autoscale the raw (not compositional) data prior to PCA (transformation of some variables might still be advisable), which takes care of differences in scale between different variables, produces readily interpretable plots, and has other useful properties as described by Bro and Smilde (2014) in their recent PCA tutorial. The autoscaling**

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will allow the PCA to reveal compositional differences between samples, which was the motivation for generating ratio data.

Reply: We agree that the data is on different scales and auto-scaling needs to be applied. In fact, in the submitted version of the manuscript, auto-scaling was applied to the data before making the PCA. For this, we used the parameter scale of the rda() function. Similarly, we auto-scaled the data by using the scale() function before applying the PERMANOVAs. In both cases, the function was mentioned in the submitted version of the manuscript, but we forgot to mention the auto-scaling. We thank the reviewer for raising our attention to the missing description and it will be included in the revised version of the manuscript.

4) Consider also the underlying assumptions of ANOVA, box and whisker plots and other statistical representations in the analysis of ratio data. When comparing ratio/percentages, it is common to arcsin transform the data first or use a chi-squared test.

Reply: We considered all statistics / plots. No further adjustments in addition to the ones explained in reply to comments 1-3 need to be done. We used logit transformation instead of arcsin transformation (see above).

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2 Sample sizes for the statistics

DK = Denmark, UY = Uruguay

2.1 Levene's tests

2.1.1 DOC and DON loads for each country and for land use within country

Statistic described in lines 209-211 of the submitted manuscript

DK = 1414, UY = 1455

Catchment	Number of samples
Intensive UY	728
Extensive UY	727
Intensive DK	707
Extensive DK	707

The high sample numbers were the result of the interpolation of the DOC and DON loads for each day between the sampling occasions (as described in lines 209-211 of the submitted manuscript).

2.1.2 Precipitation

Effect of country or land-use type within country. Whole time period is included. Description of statistic missing in methods section of the submitted version of manuscript. It will be included in the revised version of the manuscript.

DK = 1640, UY = 1991

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Catchment	Number of samples
Intensive UY	996
Extensive UY	995
Intensive DK	820
Extensive DK	920

2.1.3 Discharge

Effect of country or land-use type within country. Whole time period is included. Description of statistic missing in methods section of the submitted version of manuscript. It will be included in the revised version of the manuscript.

DK = 1554, UY = 1990

Catchment	Number of samples
Intensive UY	995
Extensive UY	995
Intensive DK	777
Extensive DK	777

2.2 Permutative one-way tests

2.2.1 Effect of country on DOC and DON concentrations

Statistic described in lines 222-224 of the submitted manuscript.

DK = 98 samples, UY = 95 samples

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2.3 Nemenyi pairwise tests

2.3.1 Effect of the sampled catchment on DOC and DON concentrations

Statistic described in lines 224-225 of the submitted manuscript.

Catchment	Number of samples	
Intensive UY	48	
Extensive UY	47	âĂČ
Intensive DK	49	
Extensive DK	49	

2.3.2 Effect of the sampled catchment on carbon or nitrogen in humic substances, C:N ratio of humic substances, fluorescence index, PARAFAC component C1 and ratio of absorbance curve slopes (Sr)

Statistic shown in Figure 4 of the submitted manuscript but not described in methods section of the submitted manuscript. The description will be included in the methods section of the revised version of the manuscript

Catchment	Number of samples
Intensive UY	48
Extensive UY	47
Intensive DK	49
Extensive DK	49

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2.4 Spearman rank correlations

2.4.1 Correlation between DOC or DON concentrations and discharge values

Statistic described in lines 225-227 of the submitted manuscript.

Catchment	Number of samples
Intensive UY	48
Extensive UY	46
Intensive DK	48
Extensive DK	48

The slightly lower number of samples is a result of the fact that not for all sampling dates discharge values were available.

Due to an error in the data preparation, less samples were included in the Spearman correlations of the submitted manuscript. This was corrected, and the Spearman rank correlations were done again with the sample numbers given above. No significant changes in the results occurred and the slightly changed rho and p values will be included in the revised version of the manuscript.

2.5 Sensitivity analysis of the load calculations

Statistic described in lines 230-237 of the submitted manuscript.

Catchment	Number of samples
Intensive UY	728
Extensive UY	727
Intensive DK	707
Extensive DK	707

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The high sample numbers were the result of the interpolation of the DOC and DON loads for each day between the sampling occasions (as described in lines 209-211 of the submitted manuscript).

2.6 Principal component analysis

Statistic described in lines 238-247 of the submitted manuscript. For the PCA, 193 samples and 20 variables were used, resulting in a sample to variable ratio of 9.65.

2.7 Permutative multivariate analysis of variance, permutative multivariate dispersal tests

Statistic described in lines 248-256 of the submitted manuscript. Same number of samples as in section 2.3.2

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