Supplement of

Geodiversity influences limnological conditions and freshwater ostracode species distributions across broad spatial scales in the northern Neotropics

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S1. Structural Equation Modelling, models performance and results.

The Structural Equation Modelling (SEM) technique was used to evaluate the direct and indirect influence of geodiversity of the northern Neotropics on limnological conditions and ostracode species richness and community composition (as a function of distribution). Five different SEM models were fitted with a set of uncorrelated variables in a covariance data matrix. Model performances were evaluated with the Chi-square test, the Root Mean Square Error of Approximation (RMSEA), comparative fit index (CFI) and standardized root mean squared residuals (SRMR).

Model 1: This model evaluates how geodiversity as an exogenous and latent variable influences two endogenous and latent variables: “limnological conditions” and “species composition”. In addition, it explores whether limnological conditions explains species composition. Geodiversity included the following variables: bedrock type and mineralogy as it was hypothesized that different bedrock types will display a mineralogical signature in aquatic systems and therefore influence water chemistry. Limnological conditions included major ions magnesium and bicarbonates recovered as relevant for YG and GSHN in the PCA, as well as, temperature, and pH assuming that species respond to temperature (ranging from 12.5 °C in the highlands to 33.7 °C in the lowlands) and pH (ranging from 6.9 to 9.9) gradients and that water chemistry limits species distributions. Species composition was represented by groups identified in the NMDS ordination. Global fit of this model: CFI 0.79, RMSEA 0.19, and SRMR 0.12. For this model the default maximum likelihood estimator was used.

Model 2: Geodiversity was considered the unique exogenous variable and constructed as in model 1, except for elevation which was included. Elevation is an important characteristic in the GSHN limnological region, ranging from sea level to more than 2800 m a.s.l., and is associated to changes in water temperature and presence of conservative ions such as carbonates-bicarbonates. Limnological conditions was considered as an exogenous variable and constructed as in model 1. The direct influence of geodiversity over limnology was tested statistically in the model, as was the direct effect of geodiversity on species richness and composition. We also evaluated the individual influence of TOC in lake sediments on species distribution, because in the Yucatán Peninsula and northern Guatemala there is a north-south gradient of increasing organic matter content, likely related to precipitation amount, vegetation stature and productivity, and soil development. This model was based on the assumption that limnological conditions exerts a primary effect on species richness and composition and that geodiversity influence over species is limited. Global fit of this model: CFI 0.79, RMSEA 0.13, and SRMR 0.14. For this model, we used weighted least squares as estimator, with the aim of getting robust standard errors and a scaled test statistic.

Model 3: This model evaluated the individual influence of two exogenous variables, geodiversity and limnological conditions, on two endogenous variable, species composition (latent) and richness (observed). We consider geodiversity and limnology to be independent. Our aim was to estimate whether the influence of each exogenous variables was statistically significant, and which variable was of greater significance for community composition and richness. For this model, geodiversity and limnology were constructed as in model 1, except for limnology, for which TOC and conductivity were included. This model produced the following metrics of global fit CFI 0.94, RMSEA 0.06, and SRMR 0.14, which are better scores than in model 1 and 2, but the algorithm did not compute the Gamma matrix, as the data set was too small to calculate the significance of both exogenous variables. This precluded meaningful interpretation of the resulting metrics of fit.

Model 4: In this model the influence of geodiversity over limnological conditions was evaluated, and then, the direct influence of limnology and direct/indirect influence of geodiversity on species composition and richness was tested. For the construction of the geodiversity variable, we used a reduced data set compared with model 3, as we excluded elevation, whereas for the limnological conditions, conductivity was excluded. The individual influence of elevation and conductivity was then tested on species composition and richness, as they are responsible for environmental gradients in the northern Neotropics. For instance, conductivity was recovered as the most relevant variable in the YG and GSHN limnological regions, and the variable-specific interpolated maps revealed gradients in the Yucatán Peninsula by the presence of carbonates in the northern-central part of the Peninsula and by the presence
of chloride in coastal areas. In Central America, elevation gradients are relevant, particularly in the Guatemalan mountain systems such as the Sistema de los Cuchumatanes and Sistema de la Sierra Madre, where elevation may exceed 2800 m a.s.l. Metrics of global fit are as follows CFI 0.94, RMSEA 0.01, and SRMR 0.03.

Model 5: This model’s performance is similar to that of model 4, except for the fact that we tested the individual influence of the variables TOC and latitude. We selected TOC, as it displays an environmental gradient north-south in Yucatán Peninsula-northern Guatemala, with lower values in the north and progressively increasing values to the south. Latitude was used to test the influence of latitudinal gradients, particularly on species richness. Although the study region is considered tropical, the northern Neotropics is, in fact, part of the transition between the two ecoregions of the American continent, and besides latitude, other gradients associated occur, such as precipitation. We aim to investigate positive/negative correlations between the number of species and latitude to ascertain whether ostracode species numbers increase or decrease at lower latitudes. Metrics of global fit of this model are as follows, CFI 0.62, RMSEA 0.20, and SRMR 0.17.

Output table of model 4 is displaced

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| Estimator       | ML       |
|-----------------|----------|
| Optimization method | NLMINB  |
| Number of model parameters | 16      |
| Number of observations | 66      |

Model Test User Model:

| Test statistic | 2.523 |
|----------------|-------|
| Degrees of freedom | 5     |
| P-value (Chi-square) | 0.735 |

Model Test Baseline Model:

| Test statistic | 75.341 |
|----------------|-------|
| Degrees of freedom | 15    |
| P-value | 0.000  |

User Model versus Baseline Model:

| Comparative Fit Index (CFI) | 0.941 |
|-----------------------------|-------|
| Tucker-Lewis Index (TLI)    | 0.963 |

Loglikelihood and Information Criteria:

| Loglikelihood user model (H0) | -320.248 |
| Loglikelihood unrestricted model (H1) | -322.115 |
Akaike (AIC) 682.503
Bayesian (BIC) 717.624
Sample-size adjusted Bayesian (BIC) bb/0.86

Root Mean Square Error of Approximation:
RMSEA 0.013
90 Percent confidence interval - lower 0.000
90 Percent confidence interval - upper 0.135
P-value RMSEA <= 0.05 0.014

Standardized Root Mean Square Residual:
SRMR 0.035

Parameter Estimates:

Latent Variables:

| Latent Variable | Estimate | Std.Err | z-value | P>|z| | Std.lv | Std.all |
|-----------------|----------|---------|---------|------|-------|--------|
| geodiversity    |          |         |         |      |       |        |
| Carbonates      | 1.320    | 0.275   | 0.275   |      | 0.013 | 0.919  |
| Feldspars       | -1.854   | 0.751   | -2.255  | 0.021| -0.441| -0.441 |
| Bedrock         | 2.743    | 0.823   | 3.250   | 0.000| 0.794 | 0.794  |
| limnology       |          |         |         |      |       |        |
| Mg              | 1.000    |         |         | 0.103| 0.103 | 1.525  |
| HCO3            | 2.390    | 3.220   | 0.831   | 0.033| 0.266 | 0.266  |
| Temp            | 3.481    | 4.430   | 0.822   | 0.013| 0.364 | 0.364  |
| pH              | -1.714   | 2.187   | -0.801  | 0.328| -0.187| -0.187 |
| TOC             | 2.425    | 3.054   | 0.795   | 0.041| 0.250 | 0.250  |
| spedist         |          |         |         |      |       |        |
| NMDs            | 1.000    |         |         | 1.000| 1.000 |        |

Regressions:

| Regression     | Estimate | Std.Err | z-value | P>|z| | Std.lv | Std.all |
|----------------|----------|---------|---------|------|-------|--------|
| limnology ~    |          |         |         |      |       |        |
| geodiversity   | 0.555    | 0.650   | 0.851   | 0.039| 1.525 | 1.525  |
| spedist ~      | -1.532   | 2.046   | -0.743  | 0.045| -0.157| -0.157 |
| limnology ~    | 0.015    | 1.265   | 0.009   | 0.993| 0.001 | 0.001  |

Covariances:

| Covariance     | Estimate | Std.Err | z-value | P>|z| | Std.lv | Std.all |
|----------------|----------|---------|---------|------|-------|--------|
| spedist ~~     | -0.173   | 0.079   | -2.105  | 0.035| -0.697| -0.697 |
| .geodiversity  |          |         |         |      |       |        |
| .spedist       | -0.502   | 0.103   | -2.254  | 0.024| -0.279| -0.288 |
| .cond          | -0.071   | 0.078   | 0.903   | 0.036| 0.085 | 0.102  |
| .elevation     | 0.049    | 0.065   | 0.717   | 0.043| 0.163 | 0.163  |
| .geodiversity  | 0.022    | 0.139   | 0.156   | 0.876| 0.022 | 0.022  |
| .richness      | -0.200   | 0.119   | -1.682  | 0.093| -0.200| -0.241 |
| .cond          | 2.475    | 0.141   | 0.672   | 0.033| 0.307 | 0.307  |
| .elevation     | 0.889    | 0.062   | 0.715   | 0.041| 0.163 | 0.163  |

Variances:

| Variance       | Estimate | Std.Err | z-value | P>|z| | Std.lv | Std.all |
|----------------|----------|---------|---------|------|-------|--------|
| .Carbonates    | 0.905    | 0.432   | 2.126   | 0.031| 0.919 | 0.919  |
| .Feldspars     | 0.772    | 0.354   | 2.217   | 0.025| 0.784 | 0.784  |
| .Bedrock       | 0.313    | 0.133   | 2.302   | 0.022| 0.307 | 0.307  |
| .Elevation     | 0.687    | 0.215   | 3.194   | 0.001| 0.687 | 0.687  |
| .Mg            | 0.989    | 0.166   | 5.951   | 0.000| 0.989 | 0.989  |
| .HCO3          | 0.929    | 0.164   | 5.682   | 0.000| 0.929 | 0.929  |
| .Temp          | 0.868    | 0.144   | 6.038   | 0.000| 0.868 | 0.868  |
| .pH            | 0.935    | 0.142   | 6.796   | 0.001| 0.965 | 0.965  |
| .TOC           | 0.921    | 0.236   | 3.979   | 0.000| 0.938 | 0.938  |
| .cond          | 0.936    | 0.358   | 2.613   | 0.009| 0.936 | 0.936  |
|               | Estimate | R-Square: |
|---------------|----------|-----------|
| Carbonates    | 0.081    |           |
| Feldspars     | 0.216    |           |
| Bedrock       | 0.693    |           |
| Altitude      | 0.313    |           |
| Mg            | 0.011    |           |
| HCO3          | 0.071    |           |
| Temp          | 0.132    |           |
| pH            | 0.035    |           |
| TOC           | 0.062    |           |
| cond          | 0.064    |           |
| NMDS          | 1.000    |           |
| richness      | 0.001    |           |
| limnology     | NA       |           |
| spedist       | 0.303    |           |
Figure S1. Ternary plots showing major cations and anion proportions [%] of 76 aquatic systems of the northern Neotropical region. Major anions (A) and major cations (B) from YG limnological group; Abbreviations correspond to those in Table S1, colors representing limnological subregions, correspond to the cluster analysis dendrogram (Fig. 2).
Figure S2. Ternary plots showing major cations and anion proportions [%] of 76 aquatic systems of the northern Neotropical region. A) major anion from GSHN limnological group; d) major cation from GSHN limnological group. Abbreviations correspond to those in Table S1 and colors, representing limnological subregions, correspond to the cluster analysis dendrogram (Fig.)
Table S2.1. Loading values of the 13 variables used for principal component analysis of the YG region (P <0.05). Significant scores for components 1 and 2 in bold.

| Variable     | PC1 (23.47 %) | PC2 (14.63 %) |
|--------------|---------------|---------------|
| Temperature  | 0.02          | 0.35          |
| DO           | 0.06          | 0.45          |
| pH           | 0.01          | **0.73**      |
| Conductivity | **0.77**      | 0.04          |
| HCO₃⁻        | 0.13          | 0.05          |
| Cl⁻          | **0.64**      | 0.06          |
| Na⁺          | **0.72**      | 0.04          |
| Ca           | 0.51          | 0.11          |
| Mg²⁺         | **0.65**      | 0.04          |
| TOC          | 0.10          | 0.13          |
| Altitude     | 0.40          | 0.12          |
| SO₄²⁻        | 0.47          | 0.12          |
| Age          | 0.43          | 0.47          |

Table S2.2. Loading values of the 13 variables used for principal component analysis of the GSHN limnological region (P <0.05). Significant scores for components 1 and 2 in bold.

| Variable     | PC1 (26.83 %) | PC2 (22.89 %) |
|--------------|---------------|---------------|
| Conductivity | 0.57          | 0             |
| HCO₃⁻        | **0.70**      | 0.11          |
| Cl⁻          | **0.62**      | 0.08          |
| Na⁺          | **0.72**      | 0.07          |
| Ca           | 0.33          | 0.15          |
| Mg²⁺         | 0.48          | 0.05          |
| TN           | 0.01          | 0.62          |
| TOC          | 0.01          | **0.79**      |
| Phyllosilicates | 0.11      | 0.20          |
| Altitude     | 0.11          | 0.09          |
| CO₃⁻         | **0.56**      | 0.17          |
| Bedrock      | 0.12          | **0.75**      |
| Age          | 0.23          | **0.80**      |
### Table S3. List of ostracode species found in our study.

| #  | Species name                                         | Species code | #  | Species name                                         | Species code |
|----|------------------------------------------------------|--------------|----|------------------------------------------------------|--------------|
| 1  | Alicinula serricaudata (Klie, 1935a)                 | ASE          | 36 | Hemicypris sp.1                                      | Hemicypris sp.|
| 2  | Alicinula yucatanensis sp. nov.                      | AYU          | 37 | Heterocypris nicaraguensis Hartmann 1959            | HNI          |
| 3  | Candonia sp.                                         | Candonia sp. | 38 | Heterocypris punctata (Keyser, 1975)                | HPU          |
| 4  | Cypretta campechensis Cuhu-Durán et al., 2013        | CCA          | 39 | Keysercypris sp.1                                   | Keysercypris sp.1 |
| 5  | Cypretta cf. campechensis Cuhu-Durán et al., 2013    | CCAF         | 40 | Keysercypris sp.2                                   | Keysercypris sp.2 |
| 6  | Chlamydotheca cf. colombiensis Roessler 1985         | CCOF         | 41 | Keysercypris sp.3                                   | Keysercypris sp.3 |
| 7  | Cypretta elongata sp. nov.                           | CEL          | 42 | Keysercypris sp.4                                   | Keysercypris sp.4 |
| 8  | Cypretta cf. elongata                                | CELf         | 43 | Keysercypris sp.5                                   | Keysercypris sp.5 |
| 9  | Cypria gibbera Furtos, 1936*                         | CGI          | 44 | Keysercypris granulata (Hartmann 1959)             | KGR          |
| 10 | Chlamydotheca sp. 1                                 | Chlamydotheca sp. | 45 | Limnocythere floridensis Keyser, 1975               | LFL          |
| 11 | Cytheridella ilnovayi Daday, 1905                    | CIL          | 46 | Limnocythere sp.1                                   | Limnocythere sp. |
| 12 | Cypretta maya Cuhu-Durán et al., 2013               | CMA          | 47 | Limnocythereina royi Hartmann 1959                  | LRO          |
| 13 | Cypria petenensis Ferguson et al., 1964             | CPE          | 48 | Limnocythere cf. stationis Vávra, 1891             | LST          |
| 14 | Cypretta spinosa Cuhu-Durán et al., 2013            | CSP          | 49 | Neocypridopsis sp.1                                 | Neocypridopsis sp. |
| 15 | Cyprinotus unispinifera Furtos, 1936b              | CUN          | 50 | Pseudocandona antilliana Broodbakker, 1983c       | PAN          |
| 16 | Chlamydotheca unispinosa (Baird, 1862)             | CUNI         | 51 | Peryssocytheridea cf. cribrosa Klie, 1933a         | PCRf         |
| 17 | Cypridopsis sp. [Ca 1]                               | CVI sp 1     | 52 | Penthasilenula sp.1                                 | Penthasilenula sp. |
| 18 | Cypridopsis sp. [Ca 2]                               | CVI sp 2     | 53 | Potamocypris islagrandensis Hoff 1943b             | PIS          |
| 19 | Cypridopsis sp. [Ca 3]                               | CVI sp 3     | 54 | Pericythere marginata Hartmann, 1959               | PMA          |
| 20 | Cypridopsis sp. 4                                   | CVI sp 4     | 55 | Paracythereis opesta (Brehm, 1939)                 | POP          |
| 21 | Cypridopsis sp. 5                                   | CVI sp 5     | 56 | Potamocypris sp.1                                  | Potamocypris sp.1 |
| 22 | Cypridopsis sp. 6                                   | CVI sp 6     | 57 | Potamocypris sp.2                                  | Potamocypris sp.2 |
| 23 | Cypridopsis sp. 7                                   | CVI sp 7     | 58 | Potamocypris sp.3                                  | Potamocypris sp.3 |
| 24 | Cypricerinaceae sp. 1                               | CYP 1        | 59 | Pseudocandona sp.1                                 | Pseudocandona sp.1 |
| 25 | Cypricerinaceae sp. 2                               | CYP 2        | 60 | Pseudocandona sp.2                                 | Pseudocandona sp.2 |
| 26 | Cypricerinaceae sp. 3                               | CYP 3        | 61 | Pseudostrandesia sp.1                             | Pseudostrandesia sp.1 |
| 27 | Cyprinotinae sp. 1                                  | CYP 4        | 62 | Pseudostrandesia sp.2                             | Pseudostrandesia sp.2 |
| 28 | Cypris sp. 1                                        | Cypria sp. 1 | 63 | Strandesia bicuspis (Claus 1892)                   | SBI          |
| 29 | Cypris sp. 4                                        | Cypria sp. 4 | 64 | Stenocypris cylindrical major (Baird, 1859b)      | SCY          |
| 30 | Cypris sp. 5                                        | Cypria sp. 5 | 65 | Strandesia intrepida Furtos, 1936b                | SIN          |
| 31 | Cyprideis cf. salebrosa                             | Cyprideis sp. | 66 | Strandesia sp.1                                   | Strandesia sp. |
| 32 | Cyprididae sp. [Ca 1]                               | Cyprididae sp. | 67 | Tanycypris sp.1                                   | Tanycypris sp. |
| 33 | Desc. 1                                             | Cyprididae sp.2 | 68 | Thalassicypria sp.1                               | Thalassicypria sp. |
| 34 | Diaphanocypris meridana (Furtos, 1936b)             | DME          | 69 | Vestalenula sp.1                                  | Vestalenula sp. |
| 35 | Darwinula stevensoni (Brady & Robertson, 1885)      | DST          | 70 | Vestalenulaflagioli (Pinto and Kotzian 1961)      | VPA          |
