SHORT-NOTE

Hidden impacts of environmental stressors on freshwater: Cytoscape® correlation networks for community ecotoxicology analysis.

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Abstract

Freshwater systems are affected by multiple stressors, ecotoxicological studies using communities or highest ecological levels commonly focus on abundances analysis. This proposal suggests incorporating correlation networks to bring to light possible hidden impacts on the community connections.

Keywords Stressors; Freshwater; Community, Correlation networks

Introduction

Freshwater systems are exposed to multiple stressors as pesticides, microplastics, heavy metals, wastewater, industrial residues, biological invasions, nutrification and physical factors such as the temperature increase as a consequence of global warming (Jackson et al. 2016). Aquatic ecosystems are composed by several communities that could be direct or indirect affected and interact through trophic relationships (Kattwinkel et al. 2010). The stressors

impacts on freshwater communities can be detected by structural changes (specific abundances, diversity index, etc.) and/or by physiological effects on organisms, leading or not to, to functional changes (productivity, gas emissions, etc.) (Havens, 1994). Correlation networks using very simple interfase using Citoscape® (Shannon et al. 2003) could help us to understand the relationships between the differents components of the communities face to an impact (Lozano et al. 2019), and to elucidate ecological mechanisms of mitigation or exacerbation. A new mode of response through community or highers levels could appear after an impact by the modification of the relationships between the different components of a system. In this work, it is proposed a community-based correlation analyze using specific abundances to attend possible hidden effects of environmental stressors with focus in freshwater systems.

Methods

Theorical values of specific abundances results in a community after an impact were used to simulate possible hidden effects of one environment stressor. Spearman correlation coeficients were calculated with InfoStat v.2008 and correlation networks were built with Cytoscape® v.3.7.1., incorporating only significant correlation coeficients ($p<0.05$) Other theorical examples were built directly on Cytoscape® to illustrate proposals.

Results and discussion

Hypothetical values of specific and total abundances from a community treated or not with a stressor are shown on Table I, with means and standar

desviation of 4 replicates from control (C) and treatment (T). Non-statistical differences are seen under a classical abundance analyze (Fig. I).

	Sp.1	Sp.2	Sp.3	Sp.4	Total
C1	4	4	8	3	19
C2	2	2	4	1	9
C3	2	2	4	1	9
C4	2	2	4	1	9
Mean	2.5	2.5	5.0	1.5	11.5
SD	1.0	1.0	2.0	1.0	5.0
T1	2	4	4	1	11
T2	2	2	8	1	13
T3	4	2	4	3	13
T4	2	2	4	1	9
Mean	2.5	2.5	5.0	1.5	11.5
SD	1.0	1.0	2.0	1.0	1.9

Table I: Simulated values of specific abundances of controls (C) and treated systems (T).

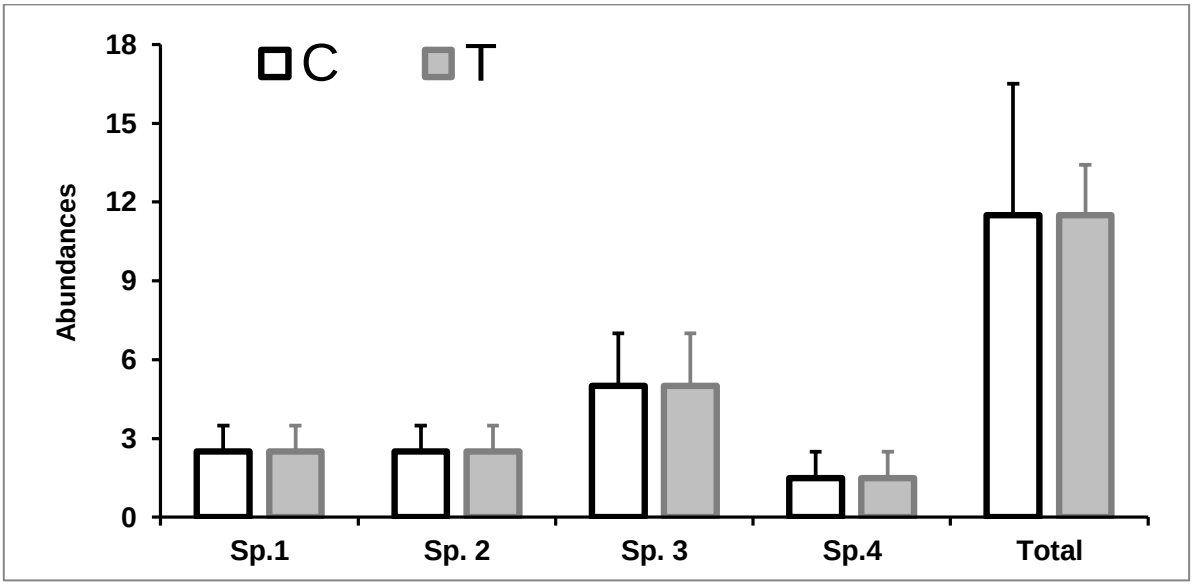

Fig. I: Abundances of simulated control (C) and treated (T) systems, non-significant variations are found.

Using the specific abundances of the Table I, Spearman correlations were calculated and used to build networks with Cytoscape®. Hidden effects of the stressor that change the relationships between the populations are shown (Fig. II). Considering the number of replicates (n=4) and the number of specific abundances (4), the maximum possible correlations was 6 (4*3:2) and the minimun was 0 if any specific abundace was correlated. Results show that a community with non-significant specific abundances variations could be strongly affected on its connectivity, in this exemple; going from a situation where all populations from a community are positively correlated (A) to other situation where each population is totally disconnected (B).

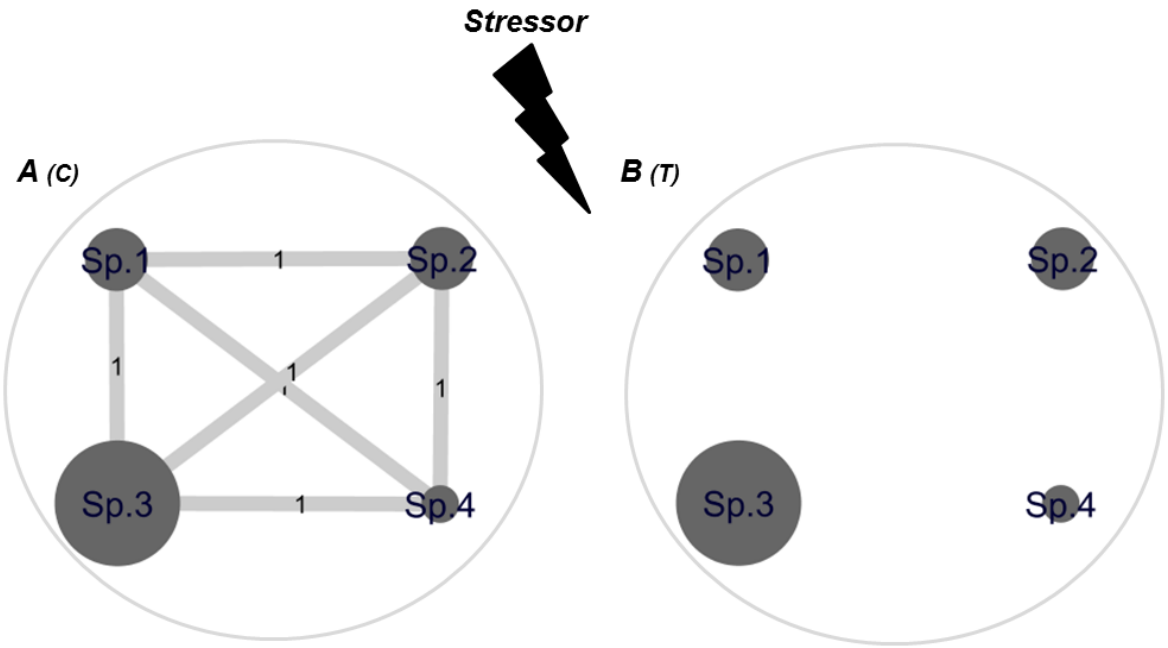

Fig. II: Hidden effects on the community caused by a stressor are exposed by the correlation network analyze. Significant Spearman correlation values are shown as edges ($p<0.001$), balls are related to relative specific abundances.

The modification of the relationships between specific fractions from a system after a stressor could affect a posterior community response face to another stressor (Ashauer et al. 2017). In this sense, if the Sp.4 from our example corresponds to a bloom-former cyanobacteria, it is probably that its disconnection from the community (Fig. III B) will enhance the possibility of a bloom face to another stressor or opportunity (Fig. III C). This proposal attends to motivate the incorporation in the ecotoxicological assays, that use at least a community level, the analysis of correlation networks. It is important to highlight that conclusions should be made paying attention to the inherent number of correlation possibilities, which is determined by the number of replicates and the number of components, and working always under a biological hypothese. Correlation values must be calculated after normality verification, if it is the case, Pearson coefficients should be used, in other case Spearman rank approach is recommended. Morever, multi-trophic studies analyze could also be improved with this kind of approach (Fig. IV).

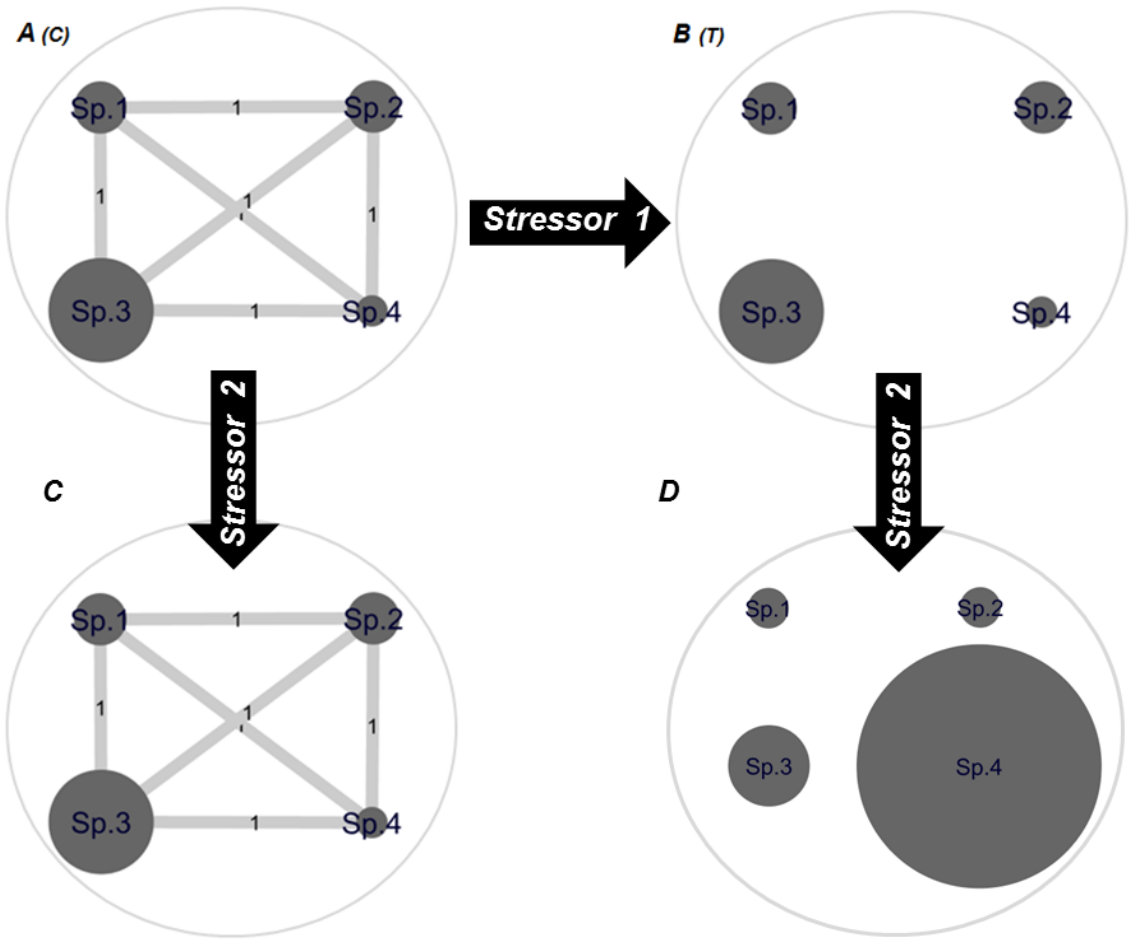

Fig. III: Possible consequences of changes in networks behaviour face to secuential stressors.

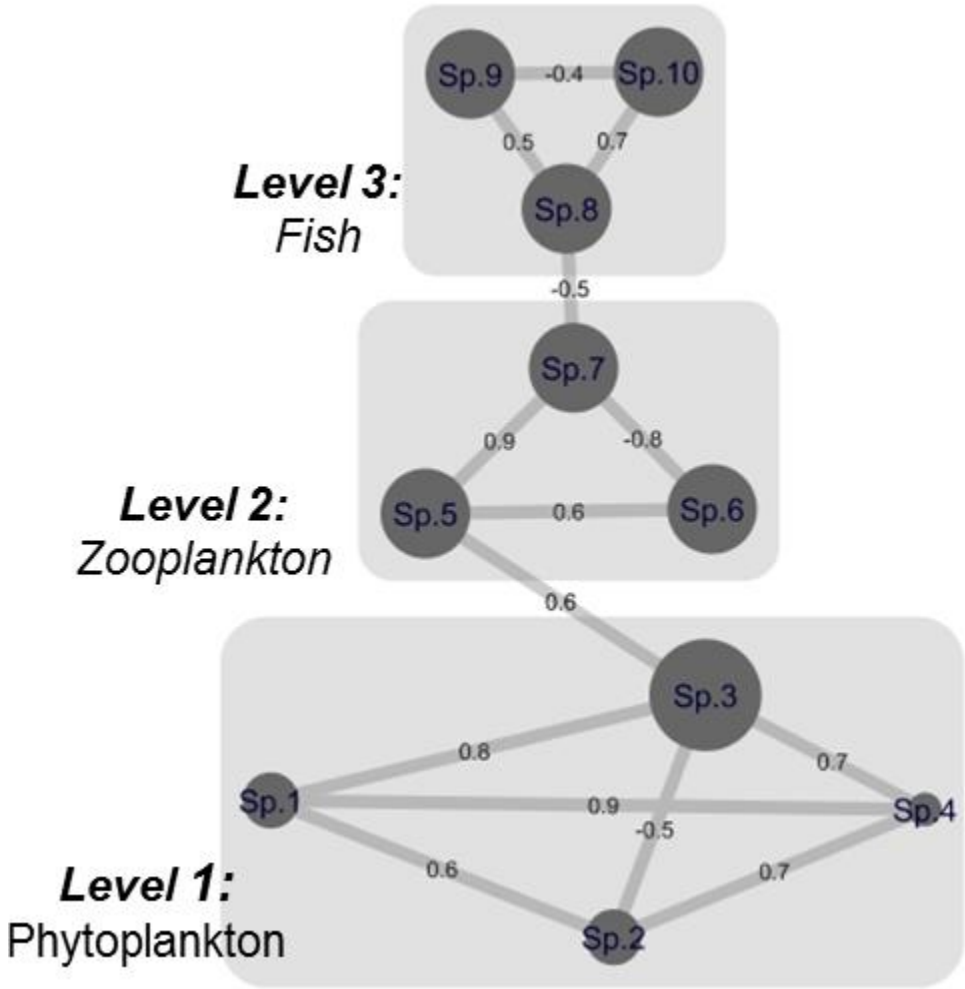

Fig. IV: Multi-trophic correlation network.

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