Consideration of mass effect processes in bioindication allows more accurate bioassessment of water quality

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Consideration of mass effect processes in bioindication allows more accurate bioassessment of water quality

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A R T I C L E I N F O

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A B S T R A C T

Bioassessment is widely used to measure ecological integrity of natural habitats following anthropogenic disturbances and modifications. Traditionally, bioassessment has been based exclusively on species-environment interactions, *i.e.* niche processes. However, dispersal processes, and in particular mass effect, could mask the influence of niche processes and lead to erroneous conclusions about ecosystem health. To circumvent this problem, we identified 40 diatom species with distributions driven primarily by mass effect and propose an alternative version of the Biological Diatom Index (IBD_{2007}) excluding these species. We tested the environmental responses of both the original IBD (IBD_{2007}) and the modified IBD (IBD_{mod}) with a benthic diatom dataset from France, collected between 2007 and 2013 and including 9487 samples from 3913 spatially distinct localities. Our results indicate a better relationship between the IBD_{mod} scores and environmental conditions, compared to the IBD_{2007} scores, leading to a more accurate determination of river ecological status, especially in conditions of moderate nutrient enrichment. This study supports the idea that mass effect may result in biased evaluation of water quality. It is advocated that this process is considered in other diatom-based indices, and by extension, in any biotic index.

1. Introduction

Bioassessment is broadly implemented by state and government agencies in an effort to measure how species communities respond to anthropogenic stressors, *e.g.* EU Water Framework Directive (European Parliament, 2000). A key instrument of bioassessment are biotic indices, which have been traditionally derived from species tolerance and sensitivity to anthropogenic perturbations (Moog et al., 2018). In freshwater biomonitoring, diatom-based indices are commonly used to capture species and community responses to environmental conditions, such as eutrophication, acidification, and organic pollution (Stevenson and Bahls, 1999; Passy and Bode, 2004) or even to specific stressors (Vilmi et al., 2016) in order to determine the level of habitat impairment. The Biological Diatom Index (IBD_{2007}) (Lenoir and Coste, 1996; Coste et al., 2009) is a standardized method (AFNOR, 2007) created and widely used in France for the monitoring of water quality in accordance with the Water Framework Directive requirements.

Environmental bioassessment is typically performed using information on species tolerance and sensitivity to disturbance and measures how community composition changes under anthropogenic influences. However, community composition is driven not only by environmental factors but also by dispersal (Leibold et al., 2004). Therefore, to accurately determine ecological status, dispersal processes must be integrated in bioassessment metrics (Brown et al., 2011; Heino et al., 2013). Depending on species traits and environmental settings, dispersal can be limited or unlimited, the latter leading to mass effect. Dispersal limitation, preventing species from reaching suitable habitats, is now considered in new environmental assessment methods, focusing on smaller regions and appropriate scales (Pont et al., 2006; Smucker and Vis, 2011). Mass effect, on the other hand, allowing species persistence in unfavorable conditions due to dispersal from source habitats, has not been considered in bioassessment, despite evidence for its importance in freshwaters (Bottin et al., 2016; Goldenberg Vilar et al., 2014; Jamoneau et al., 2018; Leboucher et al., 2020). There is thus a need to evaluate the potential of mass effect to distort bioassessment results.

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A combination of spatial and trait analyses can measure the importance of mass effect in structuring active disperser communities, *e.g.*, benthic macroinvertebrates (Brown and Swan, 2010). However, this type of approach cannot be used for passive dispersers such as microalgae, due to the absence of functional traits clearly related to the dispersal capabilities of these organisms. This hampers our ability to evaluate the potential influence of mass effect in diatom-based bioassessment methods. Here we propose to circumvent this problem by implementing an innovative method, based on a species distribution modeling, with environmental and spatial factors computed from asymmetric eigenvector maps, coupled with negative co-occurrence patterns (Leboucher et al., 2020). Using this method, we identified 40 species as being influenced by mass effect, referred to as "mass effect species" henceforth.

Our objective was to evaluate whether mass effect species can weaken bioassessment and generate erroneous stream classification in terms of water quality. To achieve this goal, we examined the sensitivity to eutrophication of IBD_{2007} and modified IBD_{2007} (IBD_{mod}) after the removal of mass effect species.

2. Materials & methods

2.1. Diatom and environmental condition datasets

We analyzed a benthic diatom dataset from France, including 9487 samples collected between 2007 and 2013 from 3913 spatially distinct localities (Fig. 1), following a standardized protocol (NFT 90-354, (AFNOR, 2007). Diatoms were sampled on hard substrates between May and October, in order to reduce seasonal variability in species assemblages. Cells were identified at 1000× magnification by examining 400 cleaned diatom frustules in permanent slides using among others Krammer and Lange-Bertalot (1986-1991, 1995-2015, 2000-2013) as identification references. A taxonomic homogenization at the species level was performed with Omnidia 5.3 software (Lecointe et al., 1993).

Corresponding physico-chemical data were gathered by the French Water Agencies. Physico-chemical variables included water pH, specific conductance (at 25 °C, $mS.cm^{-1}$), biological oxygen demand (mg of O_2 consumed per liter during 5 days of incubation at 20 °C), concentrations of total phosphorous ($mg.L^{-1}$ of $_{15}P$), orthophosphate ($mg.L^{-1}$ of $_{15}P$), ammonium ($mg.L^{-1}$ of $_{7}N$), nitrate ($mg.L^{-1}$ of $_{7}N$) and nitrite ($mg.L^{-1}$ of $_{7}N$). We used the median values of all environmental variable measurements obtained during the 30 days before and the 15 days after the diatom sampling date.

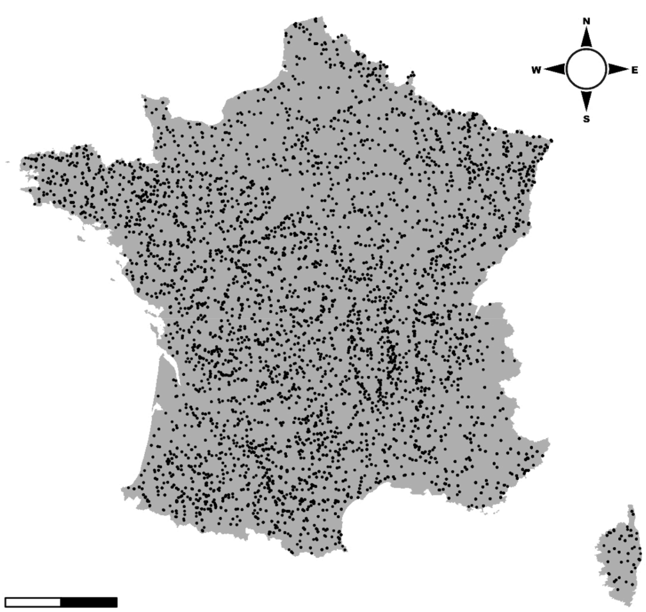

Fig. 1. Map of the studied area, including 3913 spatially distinct localities.

2.2. Biological Diatom Index (IBD_{2007})

The IBD_{2007} (Coste et al., 2009) is calculated using the sum of the presence probability of indicator taxa along seven water quality classes following the equation

$$BDI = \sum_{j=1}^{7} j \times \frac{\sum_{i=1}^{n} A_i \times V_i \times P_{ij}}{\sum_{i=1}^{n} A_i \times V_i}$$

where A_i is the abundance of taxon i, V_i is the degree of stenoecy (*i.e.* tolerance) of the taxon i, P_{ij} is the presence probability of the taxon i for the quality class j.

IBD_{2007} scores are related to reference values for the type of river considered, to calculate corresponding Ecological Quality Ratios (EQR) and to assign an ecological status class to the considered site (JORF, 2015). These 5 ecological status classes are, from the best to the worst, high, good, moderate, poor and bad. A modified IBD (IBD_{mod}) was calculated after removing from the list of indicator taxa 40 species (Table 1) previously identified as influenced by mass effect (Leboucher et al., 2020). To ensure that the removal of the 40 species does not generate a mathematical artifact, we also calculated 999 alternative versions of the IBD_{2007} (referred to as randomized IBD_{2007}) after removing 40 species randomly drawn among the indicator species not subjected to mass effect.

Table 1
List of the 40 indicator taxa identified as influenced by mass effect in Leboucher et al. (2020).

ID	Species
DVUL	*Diatoma vulgaris* Bory
ADEU	*Achnanthidium eutrophilum* (Lange-Bertalot) Lange-Bertalot
ACOP	*Amphora copulata* (Kützing) Schoeman & Archibald
AOVA	*Amphora ovalis* (Kützing) Kützing
APED	*Amphora pediculus* (Kützing) Grunow
CNTH	*Cocconeis neothumensis* Krammer
CPED	*Cocconeis pediculus* Ehrenberg
CPLA	*Cocconeis placentula* Ehrenberg
COPL	*Cocconeis pseudolineata* (Geitler) Lange-Bertalot
DMON	*Diatoma moniliformis* Kützing
DOCU	*Diploneis oculata* (Brebisson) Cleve
ENLB	*Encyonema lange-bertalotii* Krammer
ECPM	*Encyonopsis minuta* Krammer & Reichardt
EOMI	*Eolimna minima* (Grunow) Lange-Bertalot
FPYG	*Fallacia pygmaea* (Kützing) Stickle & Mann
FSLU	*Fallacia sublucidula* (Hustedt) Mann
GMIN	*Gomphonema minutum* (Agardh)
GOLI	*Gomphonema olivaceum* (Hornemann) Brubisson
GPUM	*Gomphonema pumilum* Reichardt & Lange-Bertalot
GYAT	*Gyrosigma attenuatum* (Kützing) Rabenhorst
KAPL	*Karayevia ploenensis* (Hustedt) Bukhtiyarova
LGOE	*Luticola goeppertiana* (Bleisch in Rabenhorst) Mann
NANT	*Navicula antonii* Lange-Bertalot
NCPR	*Navicula capitatoradiata* Germain
NCTE	*Navicula cryptotenella* Lange-Bertalot
NGER	*Navicula germainii* Wallace
NAMP	*Nitzschia amphibia* Grunow
NCPL	*Nitzschia capitellata* Hustedt in Schmidt
NFON	*Nitzschia fonticola* Grunow
NREC	*Nitzschia recta* Hantzsch in Rabenhorst
NSOC	*Nitzschia sociabilis* Hustedt
NSUA	*Nitzschia subacicularis* Hustedt in Schmidt
NTUB	*Nitzschia tubicola* Grunow
PGRN	*Planothidium granum* (Hohn & Hellerman) Lange-Bertalot
PRST	*Planothidium rostratum* (Oestrup) Lange-Bertalot
PTCO	*Platessa conspicua* (Mayer) Lange-Bertalot
PBTG	*Pseudostaurosira brevistriata* (Grunow in Van Heurck) Williams & Round
RUNI	*Reimeria uniseriata* Sala Guerrero & Ferrario
RABB	*Rhoicosphenia abbreviata* (Agardh) Lange-Bertalot
SIDE	*Simonsenia delognei* Lange-Bertalot

2.3. Relationships between IBD_{2007} indices and environmental conditions

We performed a principal component analysis (PCA) on power-transformed physico-chemical variables. We used Box-Cox maximum likelihood estimation (Box and Cox, 1964) to select the power-transformation best approximating normality. We simplified the analysis by retaining only the components with eigenvalues greater than those randomly generated by a broken-stick model (Jackson, 1993; Legendre and Legendre, 2012). We used sample coordinates on the principal component axes (PC) selected by the broken-stick model to evaluate the sample site position on the environmental gradient of the dataset.

Akaike information criterion (minimum theoretical information criterion, AIC) (Akaike, 1974) was implemented to compare linear models between the different versions of IBD (IBD_{2007}, IBD_{mod}, randomized IBD_{2007}) and the PCA coordinates. The smallest value of AIC indicates the best model, and therefore, the IBD version with the best response to environmental conditions (original, modified and the randomized IBD_{2007}). Difference between the AICs of randomized IBD_{2007} and the AICs of the original and modified IBD were tested using z-scores with the criterion $-2 < Z < 2$.

2.4. Comparison of ecological status assessment

We compared the number of samples per water quality category among the original and the modified IBD with a χ^2 test. We also analyzed the differences in ecological status assessment among the different index versions, and whether these differences were correlated with the nutrient gradient (derived from the PCA coordinates) with ANOVA, followed by Tukey post-hoc tests. Only categories with at least 30 values were tested.

All data analyses were performed with R 3.4.4 (R Core Team, 2018), using the 'vegan' package (Oksanen et al., 2018).

3. Results

Following the broken-stick model, only the first principal component (PC1) axis was significant explaining 40% of the total variance (Fig. 2). PC1 represented a gradient of nutrient enrichment mainly driven by phosphorous (orthophosphates and total phosphorous), nitrogen (nitrites and ammonia) and biological oxygen demand. Only PC1 scores were then used to compare the response of the different IBD indices to environmental conditions. AIC of the linear models between IBD indices and PC1 scores was smaller for the IBD_{mod} (−45477) compared to the IBD_{2007} (−45235) indicating a better relationship.

AIC of the IBD_{2007} was not significantly different from those of the randomized IBD_{2007} (z-score = -0.45). Conversely, AIC of the IBD_{mod} was significantly smaller than AIC of the randomized IBD_{2007} (z-score = -2.30).

Original and modified IBD based ecological status classifications of sites were significantly different (χ^2 test, $p < 0.001$) largely because a significant number of sites originally belonging to average classes (good and moderate) were placed in the extreme classes (high, poor and bad) (Table 2). The overall number of samples originally belonging to high and bad quality status did not significantly change (Table 2).

The IBD_{mod} allowed for a better discrimination of ecological status assessments (Fig. 3), given that the new classes obtained with the IBD_{mod} were better correlated with the nutrient gradient from the PCA. Samples upgraded with the IBD_{mod} to higher ecological status class displayed low nutrient concentrations (low PC1 scores) whereas samples downgraded

Table 2
Number of samples across ecological status classes obtained with the original and the modified version of IBD. The sum of each row represents the number of samples placed in the respective class by the IBD_{2007}, and the sum of each column represents the number of samples placed in the respective class by the IBD_{mod}.

		Modified IBD_{2007}					
		High	Good	Moderate	Poor	Bad	Total
Original IBD_{2007}	High	2061	9	0	0	0	2070
	Good	745	2353	483	2	0	3583
	Moderate	12	297	2379	536	2	3226
	Poor	0	0	12	443	100	555
	Bad	0	0	0	0	53	53
	Total	2818	2659	2874	981	155	9487

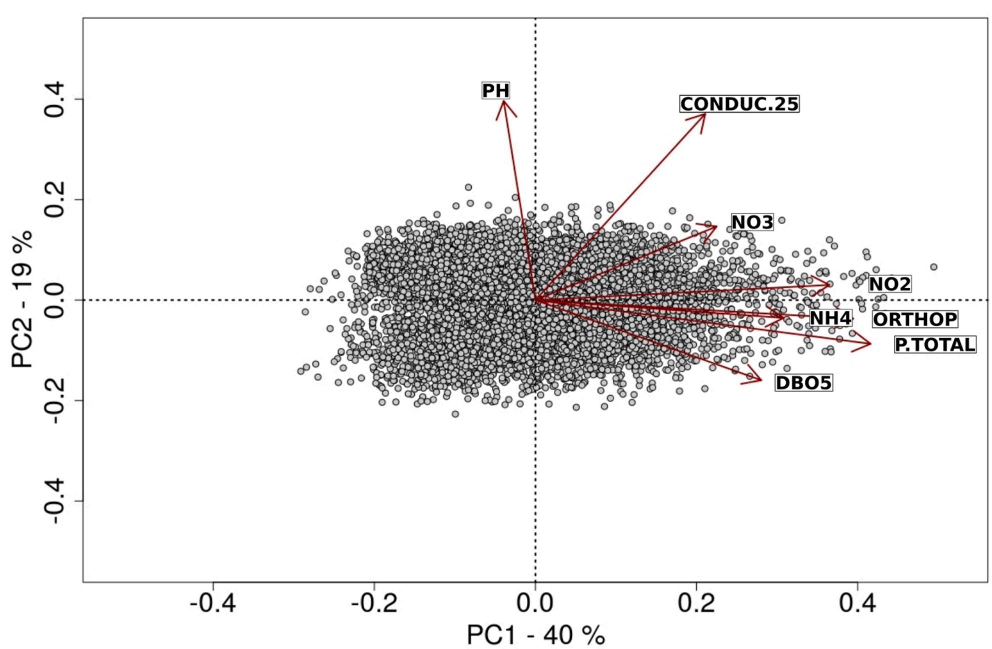

Fig. 2. Principal component analysis biplot showing site distribution along the first two principal component axes. Environmental variables are represented by arrows, inclding CONDUC.25: specific conductance at 25 °C; BDO5: biological oxygen demand; NO2: nitrite concentration; NH4: ammonium concentration; P.TOTAL: total phosphorous concentration; ORTHOP: orthophosphate concentrations.

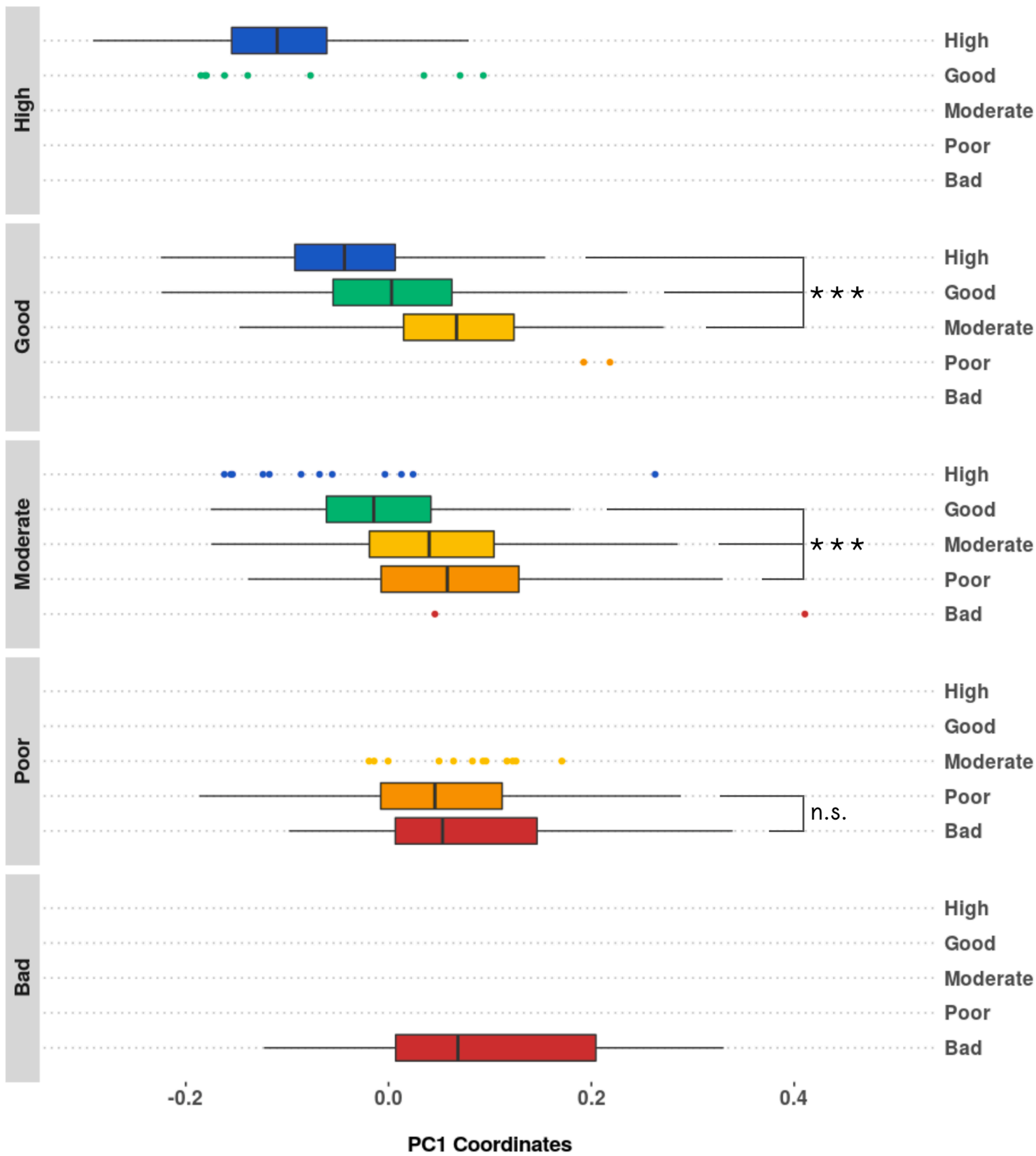

Fig. 3. Boxplot of the site coordinates on PC1 in correspondence with 1) ecological status classes from the original IBD_{2007} version (grey boxes on the left) and 2) ecological status classes from the modified IBD_{2007} version (box and whisker plots on the right). Only categories with at least 30 values were tested with ANOVA followed up by Tukey post-hoc tests when multiple classes were compared (n.s.: not significant; ***: p-value < 0.001).

showed high nutrient concentrations (high PC1 scores). These differences were, for the most part, significant (see ANOVA and Tukey tests on Fig. 3).

4. Discussion

Mass effect is an important driver of diatom community structure and thus has the potential to mask the effect of the local environment, and lead to an inaccurate evaluation of water quality (Heino et al., 2015; Jamoneau et al., 2018). However, until recently, there were no statistical methods for identifying species driven by mass effect (Leboucher et al., 2020). In this study, we implemented such method and determined the extent to which mass effect altered water quality assessment. We compared the performance of a broadly used diatom index in its original form and after the removal of species influenced by mass effect. We showed for the first time that accounting for mass effect leads to more reliable estimates of ecological status. We discuss the implications of this finding below.

Research on bioindication is just beginning to recognize the role of spatial effects (Heino, 2013), which may even exceed this of the environment. For example, Vilmi et al. (2016) simultaneously considered environmental and spatial effects on different diatom indices designed to measure water quality. Variance partitioning revealed that score variation across indices was largely associated with pure effects of spatial variables, or shared fractions of spatial variables and physico-chemical variables. In contrast, water chemistry (*i.e.* water quality or the eutrophication) did not capture much of the variation in the diatom indices. Although the results of Vilmi et al. (2016) clearly showed the importance of spatial factors, traditional methods, such as variance partitioning, were unable distinguish between the forms of dispersal, *i.e.* limited or unlimited (Cottenie, 2005), which makes them less useful for bioassessment.

In this study, we specifically targeted species identified as being influenced by mass effect by a newly developed spatial method (Leboucher et al., 2020). Using a large-scale database of diatom samples collected along a eutrophication gradient in rivers throughout France, we demonstrated a significantly improved capability of a common diatom index, IBD_{2007}, to measure water quality after the exclusion of mass effect species. Specifically, the relationship between IBD_{mod} scores and the eutrophication gradient was stronger than for the original index, which was created to respond to this type of gradient (Coste et al., 2009; Prygiel et al., 2002; Karthikeyan et al., 2018; Torrisi and Dell'Uomo, 2006).

IBD_{mod} also better discriminated ecological status of the study streams compared to IBD_{2007} and resulted in reclassification of 2198 out of the 9487 study sites (23.2%). We, therefore, recommend re-evaluation of ecological status generated by IBD_{2007}. The risk of assigning an incorrect ecological status to a site with IBD_{2007} was recently studied by Wach et al. (2019). Authors pointed out the role of the inter-operator variability (human error) in potential misclassification, but we recommend here to investigate the role of mass effect, and we expect a lower variability in ecological status assignment with IBD_{mod} scores. For example, using IBD_{2007}, Szczepocka et al. (2018) reported an unexpected ecological status of good for highly impacted urban sites. Communities from these sites were dominated by one or two taxa, including *Diatoma vulgaris a* species identified here as influenced by mass effect. In such contexts, when communities show both strong abundance inequalities and dominance of mass effect species, we consider IBD_{mod} more appropriate than IBD_{2007}.

In summary, the results presented here from a national database, indicated that mass effect processes can bias ecological status evaluation. Therefore, identification of mass effect species and their removal from biotic indices will provide more accurate bioassessment.

5. Authors' contributions

T.L., supported by J.T.R. and S.P., was responsible for the development of the study. All authors participated in designing the study and developing aims and research questions. T.L. designed methodology, extracted data and made the analyses, supported by L.M., S.B. and M.W. T.L. led the writing of the manuscript supported by J.T.R. and S.P.. All authors contributed critically to the drafts, contributed to the final version of the manuscript, and gave final approval for publication.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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