Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Strong meiofaunal differentiation despite weak links to vegetation patch structure in emerging dunes

Paul M.J. Berghuis
Royal Netherlands Institute for Sea Research
Hannah ter Horst
Naturalis Biodiversity Center
Jan-Niklas Macher
jan.macher@naturalis.nl

Naturalis Biodiversity Center

Research Article

Keywords: coastal dunes, landscape connectivity, meiofauna, community composition, beta diversity, habitat patches

Posted Date: November 18th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-11460525/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License

Additional Declarations: No competing interests reported.

Abstract

Context

Landscape studies often assume that visible habitat patches correspond to meaningful biological units, but this need not be the case. Young coastal dunes provide a useful test because pioneer vegetation forms discrete patches that contribute directly to dune development.

Objectives

We asked whether meiofaunal biodiversity follows this vegetation-defined landscape structure, and whether spatial patterns are consistent across major taxonomic groups.

Methods

We surveyed meiofaunal assemblages in standardized samples from 137 dune-grass patches across four geographically separated dune fields along the Dutch North Sea coast. We related local richness and community composition to patch area, vegetation-based connectivity, broader spatial context and measured habitat conditions. We also quantified how diversity accumulated across patches and dune fields and partitioned compositional differentiation into turnover and nestedness.

Results

Patch area and the surrounding vegetation network explained little additional variation in richness or community composition once broader spatial context and measured habitat conditions were accounted for. This result was similar across connectivity decay distances of 5–25 m. Broader spatial context was more strongly associated with community composition, particularly differences among dune fields. Assemblages nevertheless differed strongly among individual patches, and sampling additional patches and dune fields recovered substantial additional richness. Most compositional differentiation reflected taxon turnover. Annelids, arachnids and collembolans broadly followed the whole-community pattern, while nematodes showed weaker spatial structuring and collembolans a somewhat stronger association with the surrounding patch network.

Conclusions

Vegetation configuration is important for dune development, but corresponds only weakly to the spatial organization of sediment faunal diversity. The same landscape can therefore be structured differently by different ecological processes.

Introduction

Understanding whether the spatial units visible in a landscape are also the units across which biodiversity is organized is a central problem in landscape ecology (Wiens 1989; Turner 1989; Chave 2013; Fu et al. 2025). Local habitat patches are embedded within broader landscapes, and their ecological context can be defined at several nested spatial scales(Kotliar and Wiens 1990; Wu and Loucks 1995). Yet the patches that are visually obvious to the human eye are not always the spatial units that matter most to biological communities(Manning et al. 2004; Girvetz and Greco 2007). Communities can be shaped by conditions within a site, movement among nearby sites, and differences among the sets of taxa occurring in broader regions(Leibold et al. 2004; Cottenie 2005; Logue et al. 2011; Thompson et al. 2020). A landscape can therefore be strongly divided in physical space while biodiversity does not follow the same pattern. Determining whether apparent landscape units correspond to the scales at which communities actually differ is important for identifying which aspects of landscape structure are ecologically meaningful(Wu 2026).

Habitat-patch theory gives several reasons to expect landscape structure to matter. Larger patches can support larger populations and more resources, while stronger connections among patches can increase colonisation and reduce local extinction(Hanski 1998; Fahrig 2003). Movement among neighbouring patches may also make their communities more similar(Mouquet and Loreau 2003). If mapped patches and their surrounding networks represent biologically meaningful habitat structure, variation in patch area and connectivity should be reflected in local diversity and community composition. However, organisms do not always experience habitat in the same way that it appears on a map(Dennis et al. 2026a, b), and apparent connections among patches may say little about how easily organisms actually move between them(Taylor et al. 1993; Tischendorf and Fahrig 2000; Vanreusel and Van Dyck 2007). Fine-scale patch structure may therefore explain relatively little of the variation in community composition, while broader spatial context can be more important(Riva and Fahrig 2023). Richness alone cannot distinguish these patterns, because sites with similar numbers of taxa may contain very different assemblages(Hillebrand et al. 2018). Understanding how biodiversity is organized across a landscape therefore requires considering both local diversity and the additional diversity arising among patches, landscapes and regions(Wagner et al. 2000; Crist et al. 2003).

Young coastal dune systems provide a useful setting to test this relationship because their vegetation structure is itself functionally important. Pioneer vegetation traps wind-blown sand, and interactions among neighbouring dune-grass patches can influence dune formation(Hesp 1991; Maun 1998; Bonte et al. 2021; Berghuis et al. 2026a). Growth of dune-building grasses is also strongly affected by sand burial, creating a feedback between vegetation development and sand accumulation(Nolet et al. 2018). The vegetation patch arrangement contributes directly to the development of the dune landscape. This raises the question of whether the same structure is also important for the organisms living within the young dune landscape. Individual patches occur within developing dune fields, which in turn are embedded within geographically distinct coastal regions. Animal communities in dunes are known to vary with local habitat characteristics, disturbance and broader geographic context(Bonte et al. 2003, 2006; Van De

Walle et al. 2023, 2025), but much less is known about whether their spatial organization actually follows the vegetation structure involved in dune development. A weak relationship with patch area or connectivity could mean that communities differ little among patches. Alternatively, patches could contain different communities, but those differences may not follow the mapped vegetation structure.

We examined this question using meiofauna inhabiting dune-grass patches. Meiofauna comprise diverse taxonomic groups with very different dispersal capacities and modes, including passive transport in at least some groups(Ptatscheck et al. 2018). Beach meiofaunal communities also respond strongly to environmental conditions, while major taxonomic groups can show different spatial patterns(Pichler et al. 2025). They therefore provide a useful way to test both how strongly community structure follows the physical organization of the dune landscape and whether the same pattern is shared among major taxonomic groups.

Here, we used meiofaunal communities from incipient dune-grass patches across four coastal regions to ask how closely the spatial organization of biodiversity follows the nested physical structure of the landscape. We addressed three linked questions. First, how much variation in local richness and community composition is associated with patch area and structural connectivity, relative to broader spatial context and local habitat conditions? Second, at which spatial levels does additional biodiversity arise? We partitioned diversity within patches, among patches within dune fields, and among regional dune fields, and asked whether compositional differences at broader scales were mainly due to nestedness or taxon turnover. Third, do the major meiofaunal groups show similar spatial patterns, or do different groups respond differently to the same landscape structure? Together, these analyses tested whether the visible structure of emerging dunes provides a meaningful map of how biodiversity is organized across the landscape.

Methods

Study area and sampling design

Sampling was conducted in four regions along the Dutch North Sea coast: Zeeland, the Zandmotor, Texel, and Ameland. Each region contained a field of incipient dunes formed by patches of sand couch grass (*Elytrigia juncea*) and marram grass (*Ammophila arenaria*) surrounded by unvegetated sand. The regions were geographically separated and each contained substantial variation in the size and spatial arrangement of vegetation patches. This provided a nested spatial design in which individual vegetation patches were embedded within local patch networks and one dune field per region, with the four dune fields representing geographically separated regional settings (Fig. 1).

Incipient dune fields were mapped using a DJI Phantom 4 RTK unmanned aerial vehicle. Orthomosaic and digital elevation maps (with a horizontal resolution of 0.05 m) were generated using Agisoft Metashape Pro. Vegetation patches were delineated from the resulting imagery using the geospatial machine-learning platform Picterra. A custom classification model was trained to identify dune-grass

vegetation and extract the position and spatial extent of individual patches. Ground control and the positions of sampled patches were recorded using a Trimble Catalyst DA2 GNSS receiver (Trimble Inc., Westminster, CO, USA), operating with a 1 cm Catalyst subscription.

Within each region, 36 marram grass patches were selected across the observed gradients in patch area and local structural connectivity, resulting in 144 sampled patches. Sampling was stratified across both gradients to avoid concentrating observations in the most common patch configurations. Field sampling was conducted in August and September 2025. Three sediment cores were collected from each vegetation patch: one from the centre of the vegetation, one at an intermediate position, and one near the vegetation edge. Cores were collected vertically using a cylindrical corer with an internal diameter of 5 cm to a depth of 10 cm. The three cores were pooled to represent the meiofaunal assemblage of the focal patch, yielding approximately 600 mL of sediment per patch.

Separate sediment cores were collected from the same three positions for physicochemical measurements. These cores had an internal diameter of 1 cm and were inserted to a depth of 10 cm, corresponding to approximately 8 mL of sediment per core. The three cores were pooled in a sterile 50-mL polypropylene tube. All samples were transported to Naturalis Biodiversity Center immediately after collection, and meiofauna extraction was initiated within 12 h.

Sediment settlement time was used as a standardized proxy for relative sediment composition. For each measurement, 1 mL of sediment was added to a 15-cm-high settling column containing 13 mL of water, and the time until the sediment had completely settled was recorded. Measurements were repeated three times per sample and averaged. After the sediment–water mixture had remained undisturbed for 5 h, pH of the overlying water was measured using a Hanna Instruments pH probe (Hanna Instruments, Woonsocket, USA).

Meiofauna were extracted from the pooled sediment samples by freshwater flotation and decantation. Each sample was mixed with 2 L of freshwater and the sediment was swirled ten times to suspend organisms and fine material. After 20 s of settlement, the supernatant was decanted through a sieve cascade consisting of a 1-mm sieve above a 41-µm sieve. This procedure was repeated three times. Material retained on the 41-µm sieve was rinsed into a sterile 50-mL polypropylene tube, preserved in 90% molecular-grade ethanol, and stored at − 20°C until DNA extraction.

Landscape structure, local habitat and spatial context

Predictors were organized according to the spatial hierarchy tested in the study rather than treated as a single set of competing environmental variables. We distinguished four levels of structural or spatial context: (1) the focal vegetation patch, (2) its immediate patch network, (3) broader position within the dune field, and (4) regional dune-field context. Measured sediment and vegetation properties were retained separately as a local-habitat benchmark.

At the focal-patch level, structural habitat was represented by patch area derived from the mapped vegetation polygons. Immediate patch-network structure was quantified using an area-weighted distance-decay connectivity index. Larger neighbouring patches contributed more strongly to the index, while their contribution declined exponentially with increasing Euclidean distance from the focal patch. Connectivity was calculated separately within each region, with the focal patch excluded. A decay distance of 10 m was specified a priori for the primary analysis, and the resulting connectivity index was transformed as log(x + 1) before standardization. To assess whether the neighbourhood association depended on the spatial scale at which connectivity was defined, we additionally calculated the same index using decay distances of 5 and 25 m. These were treated as scale-sensitivity analyses around the predefined 10 m primary scale rather than as a procedure for selecting an optimal scale.

Broader within-dune-field context was represented by the position of each patch within its dune field. This included distance to the coastline, distance to established dunes, and two spatial covariates derived from geographic coordinates. Dutch national-grid coordinates were centred separately within each region and subjected to principal component analysis; the first two axes were retained as descriptors of relative spatial position within dune fields. Because coordinates were centred within regions, these axes represented fine- to intermediate-scale spatial structure independently of the broad geographical separation among Zeeland, Zandmotor, Texel, and Ameland. Region itself represented the broadest spatial context.

Local habitat conditions were characterized using sediment pH, mean settlement time, and percentage grass cover within 10 m of the focal patch. These variables were included as a benchmark against which correspondence with the structural hierarchy could be evaluated, rather than being interpreted as an additional spatial level.

Predictor screening and standardization

Candidate variables initially comprised sediment pH, sediment settlement time, distance to the coastline, distance to established dunes, patch area, surrounding grass cover, structural connectivity, patch elevation, and local patch density. Collinearity was assessed using Pearson correlations calculated after centring predictors within regions, so that correlations reflected covariation among patches within dune fields rather than differences among the four regions. Predictor pairs with $|r| \geq 0.70$ were considered strongly collinear. Where this threshold was exceeded, one variable was retained on the basis of its ecological relevance to the predefined spatial hierarchy and the redundant variable was excluded.

Patch elevation was strongly correlated with grass cover ($r = 0.753$), and local patch density was likewise strongly correlated with grass cover ($r = 0.923$); elevation and patch density were therefore excluded from the primary analyses. The retained variables were assigned to the predefined focal-patch, neighbourhood, within-field, regional, or local-habitat components described above. All retained continuous predictors were centred and scaled to unit variance before analysis, such that model effects corresponded to a one-standard-deviation difference in the predictor.

DNA extraction and metabarcoding

Before DNA extraction, the ethanol was evaporated by placing the open sample tubes in a sterile drying cabinet at 50°C overnight. The dried meiofauna fractions were then transferred to sterile 2 mL microcentrifuge tubes. DNA was extracted using the NucleoSpin Soil kit (Macherey-Nagel, Düren, Germany) following the standard protocol supplied by the manufacturer. The protocol was modified by including an overnight Proteinase K digestion to improve the lysis of meiofaunal tissue. For this step, 50 µL of Proteinase K solution at a concentration of 250 µg mL^{-1} (Thermo Fisher Scientific, Waltham, USA) was added to the lysis buffer, following previous approaches (Weigand and Macher 2018; Macher et al. 2024).

We used a two-step PCR metabarcoding protocol targeting a 313 bp fragment of the mitochondrial cytochrome *c* oxidase subunit I gene. The fragment was amplified using the LerayXT primer set, which provides broad amplification across metazoan and other eukaryotic taxa (Leray et al. 2013; Wangensteen et al. 2018). The first PCR was performed in a total reaction volume of 20 µL. Each reaction contained 11.7 µL of molecular-grade water, 2 µL of CoralLoad PCR buffer (10×; Qiagen, Hilden, Germany), 0.4 µL of $MgCl_2$ (25 mM; Qiagen), 0.8 µL of bovine serum albumin (10 mg mL^{-1}), 0.4 µL of dNTP mixture (2.5 mM), 0.2 µL of Qiagen Taq DNA polymerase (5 U $µL^{-1}$), 1 µL of each Nextera-tailed primer (10 pmol $µL^{-1}$), and 2.5 µL of DNA template. Thermal cycling began with an initial denaturation at 96°C for 3 min, followed by 30 cycles of denaturation at 96°C for 15 s, annealing at 50°C for 30 s, and extension at 72°C for 40 s. A final extension was performed at 72°C for 5 min. Six negative controls containing molecular-grade water instead of DNA template were processed alongside the samples to detect potential contamination. No reads were recovered from the negative controls. Products from the first PCR were purified using AMPure XP beads (Beckman Coulter, Brea, USA) at a bead-to-sample ratio of 0.9:1, removing primer dimers and other short fragments. A second PCR was used to attach individual sample indices. Reaction and thermal cycling conditions were the same as for the first PCR, but amplification was limited to 10 cycles and 1 µL of purified product from the first PCR was used as template. Fragment size and DNA concentration were measured using a Fragment Analyzer with the High Sensitivity NGS Fragment Analysis kit (Agilent Technologies, Santa Clara, USA). Products were pooled at equimolar concentrations, and the final library was purified with AMPure XP beads using the same bead-to-sample ratio as described above. The pooled library was sequenced on an AVITI platform using a paired-end 2 × 300 bp high-output sequencing kit.

Bioinformatic processing

Raw paired-end reads were processed using APSCALE (v 4.2) (Buchner et al. 2022). Paired reads were merged using a maximum difference threshold of 10% and a minimum overlap of 100 bp. Only merged sequences between 300 and 326 bp in length were retained. Sequence variants represented by fewer than five reads before denoising were removed. Sequences were processed as operational taxonomic units (OTUs), which were generated by clustering sequences at 97% similarity. Within each sample, OTUs

representing less than 0.01% of the reads were removed. OTU sequences were compared against the NCBI GenBank database supplemented with curated COI reference sequences generated during taxonomic expert workshops at Naturalis Biodiversity Center, Leiden (Macher et al. 2024), using blastn (Camacho et al. 2009). Assignments with less than 85% sequence identity to the closest reference sequence were excluded from further analyses. OTUs assigned to non-meiofaunal organisms were also removed. The resulting OTU occurrence matrix was converted to presence–absence data for the subsequent community analyses. Of the 144 sampled patches, 7 were not analysed further due to a lack of molecular data after sequencing, bringing the number of analysed samples to 137.

Statistical analyses

All analyses were conducted in R using OTU presence–absence data. Predictors were grouped according to the predefined spatial hierarchy: focal patch (patch area), immediate neighbourhood (structural connectivity), within-dune-field context (spatial PC1 and PC2 plus retained distances to the coastline and established dunes), and regional context (region). Retained sediment and vegetation variables were analysed separately as a local-habitat benchmark. Structural connectivity calculated with a 10 m decay distance was used in all primary analyses, and the 5 and 25 m decay distances were used to assess the scale sensitivity of the neighbourhood effect.

To test structural–biological correspondence, local OTU richness was analysed with negative-binomial generalized linear models and community composition with distance-based redundancy analysis (dbRDA) of binary Jaccard dissimilarities using a Lingoes correction. We quantified the unique contribution of each predictor block after accounting for all other blocks. For comparison, we also quantified model fit for region alone and for the full model containing all predictor blocks. We additionally quantified the joint contribution of patch area and structural connectivity beyond regional context by comparing the region-only model with a model containing region, patch area and connectivity.

Richness effect sizes were expressed as changes in McFadden's pseudo-R^2, with nested models compared by likelihood-ratio tests. Compositional effect sizes were expressed as changes in adjusted R^2. Compositional significance was assessed using 9,999 permutations; tests of patch, neighbourhood, within-dune-field and local-habitat effects used permutations restricted within regions, whereas region was tested using unrestricted permutations. P-values for the predictor-block tests were adjusted within each response using the Benjamini–Hochberg procedure. For the connectivity-scale sensitivity analysis, the unique contribution of the neighbourhood block was recalculated for connectivity defined with 5, 10 and 25 m decay distances while all other predictor blocks were kept unchanged. We compared effect sizes across scales rather than selecting the scale with the strongest association.

To determine where biodiversity accumulated across the spatial hierarchy, total OTU richness was additively partitioned into diversity within patches, additional diversity among patches within dune fields, and additional diversity among regional dune fields. Regions were weighted equally by first calculating mean patch richness and pooled regional richness within each region. We complemented this richness

partition with multisite Sørensen dissimilarity, decomposed into turnover and nestedness using betapart. Multisite beta diversity was calculated among patches within each region and among four regional species pools obtained by pooling OTU occurrences across patches within regions. As supporting analysis, pairwise Sørensen dissimilarity, turnover and nestedness were compared between within- and between-region patch pairs; significance was assessed by permuting region labels among patches 9,999 times rather than treating pairwise observations as independent. Because incidence-based dissimilarity can be sensitive to sparsely detected taxa, we repeated the pairwise beta-diversity analysis after excluding OTUs occurring in only one sample as a sensitivity analysis.

To test taxonomic congruence, the compositional hierarchy test was repeated for major taxonomic groups containing at least 10 OTUs, occurring in at least 30 patches overall and in at least five patches per region. Group-specific composition analyses included only patches in which the focal group occurred, avoiding the treatment of jointly empty patches as compositionally identical. Group-level effect sizes were compared descriptively because groups differed in the number and identity of occupied patches.

Results

Across the four regions, 137 of the 144 sampled patches were retained for community analyses, comprising 350 OTUs. Sample numbers were 36 patches each in Ameland and Texel, 34 at Zandmotor and 31 in Zeeland.

Structural–biological correspondence

Community composition was associated mainly with dune-field and regional context rather than with the fine-scale patch mosaic (Fig. 2A). Region alone accounted for 20.0% of adjusted compositional variation, compared with 23.3% for the full model. Adding patch area and connectivity to the region-only model increased adjusted R^2 from 20.0% to 20.8%, a difference of only 0.82 percentage points. After accounting for the other predictor blocks, regional context had the largest unique contribution (4.37%), followed by position within the dune field (2.10%). The unique contributions of focal patch area and the surrounding patch network were much smaller, at 0.34% and 0.30%, respectively. Both effects were statistically detectable (BH-adjusted P = 0.003 and P = 0.009), despite their small effect sizes. The local habitat benchmark explained a similarly small amount of unique variation (0.26%, P_BH = 0.053; Online Resource 2, Table S1). Changing the spatial scale used to calculate structural connectivity did not substantially alter this result. Connectivity explained 0.31%, 0.30% and 0.15% additional compositional variation at decay distances of 5, 10 and 25 m, respectively (Online Resource 1, Figure S1).

Local richness showed little relationship with fine-scale landscape structure (Fig. 2B). McFadden's pseudo-R^2 increased from 0.0875 in the region-only model to 0.0947 in the full model. The unique contributions of focal patch area, the local patch network and within-dune-field context were 0.03, 0.07 and 0.45 percentage points, respectively, and none remained statistically supported after correction for

multiple testing. Region was the only spatial block with a supported unique contribution to richness (1.18 percentage points, P_BH = 0.036). Overall, both richness and community composition were more strongly associated with broader spatial context than with patch area or the surrounding vegetation network.

Hierarchical accumulation and turnover of diversity

Although patch area and connectivity explained little of the variation in community composition, communities differed strongly among individual patches (Fig. 3). An average patch contained 26.4 OTUs, compared with a mean of 153 OTUs when patches were pooled within a dune field and 350 OTUs across all four regions. In the additive partitioning, 7.5% of total richness occurred within the average patch, while 36.2% accumulated among patches within dune fields and 56.3% among regional dune fields (Fig. 3A;Online Resource 2, Table S2). Regional gamma richness ranged from 95 OTUs in Ameland to 200 at Zandmotor (Online Resource 2, Table S2).

Compositional differences were already large among patches within regions and increased further between regions (Fig. 3B; Online Resource 2, Table S3). Mean pairwise Sørensen dissimilarity was 0.616 for patches from the same region and 0.813 for patches from different regions, a difference of 0.197 (P_perm < 0.001). Most of this dissimilarity reflected taxon turnover. Mean turnover increased from 0.532 within regions to 0.755 between regions (P_perm < 0.001), accounting for 86.3% and 92.8% of total dissimilarity, respectively. Nestedness contributed relatively little and decreased from 0.084 within regions to 0.058 between regions (P_perm < 0.001). The multisite analysis showed a similar pattern, with turnover accounting for 96–98% of within-region dissimilarity and 86% of the differentiation among pooled regional assemblages. Excluding OTUs detected in only a single sample had little effect on these patterns. Mean Sørensen dissimilarity remained 0.602 within fields and 0.807 between fields, and turnover remained the main component of dissimilarity.

Taxonomic congruence of the spatial hierarchy

The same broad spatial pattern was found across the major meiofaunal groups (Fig. 4; Online Resource 2, Table S4). Four groups met the predefined inclusion criteria: Arachnida (139 OTUs), Annelida (96), Nematoda (51) and Collembola (30). Regional context had the largest unique contribution to community composition in all four groups, with within-dune-field context contributing the next largest spatial component. For Arachnida, Annelida and Collembola, regional context explained 4.64%, 3.57% and 3.90% of compositional variation, respectively, compared with 1.77%, 1.54% and 1.77% for within-dune-field context. The pattern was weaker in Nematoda, where regional and within-dune-field context explained 1.25% and 0.63% of compositional variation, respectively.

Focal patch area contributed less than 0.35% in each of the analysed groups. The contribution of the surrounding patch network differed somewhat among groups and was largest for Collembola (0.74%, P_BH = 0.006), whereas it contributed effectively no unique variation to Nematoda composition (adjusted

R^2 difference = − 0.09%). Despite these differences in effect size, all four groups were associated more strongly with broader dune-field and regional context than with focal patch area or local network structure.

Discussion

Weak links between patch structure and biological organization

Our results show that spatial structure important for landscape development is not necessarily a good guide to the distribution of biodiversity. In emerging dunes, the arrangement of vegetation patches contributes to dune formation(Bonte et al. 2021; Berghuis et al. 2026a), but patch area and the vegetation-based connectivity index explained little additional variation in meiofaunal composition once broader spatial context and measured habitat conditions were taken into account. At the same time, communities differed strongly among samples from different patches. The key result is therefore not that fine-scale differentiation was absent, but that it corresponded only weakly to the mapped properties of the vegetation patches. This distinction matters because spatial structure that is important for one ecological process may not organize other components of the same system in the same way(Dennis and Huck 2025; Dennis et al. 2026a, b) .

Patch-based approaches often treat discrete habitat patches as biologically meaningful units. This is a reasonable expectation when separation among patches limits colonisation and movement(Hanski 1998; Fahrig 2003). However, patches defined from vegetation or other mapped features may not match the spatial units experienced by the organisms(Dennis and Huck 2025; Dennis et al. 2026a, b). In our system, grass patches are separated by bare sand, but that separation may represent a much weaker barrier for meiofauna than it does in the vegetation map. Small-bodied animals can disperse passively by wind and water(Fontaneto 2019; Ptatscheck and Traunspurger 2020), so even taxa with limited active mobility may move across the distances separating neighbouring grass patches. Moreover, recent work suggests the importance of continuous redistribution of sand by wind-driven erosion and deposition within young dune systems, which potentially shapes the dispersal of small-bodied animals as well(Berghuis et al. 2026b). Structural connectivity based on grass tussocks can therefore differ from the functional connectivity experienced by more mobile organisms(Keeley et al. 2022; Savary et al. 2024), and the relationship between the two may also vary among taxa with different dispersal capacities(Bertellotti et al. 2023).

Previous work has shown that interactions among neighbouring grass patches can amplify dune development(Bonte et al. 2021; Berghuis et al. 2026a). Our analysis asks a different question: whether patch area and a vegetation-based connectivity index are associated with meiofaunal composition. We did not measure either sand accumulation or faunal movement, and our connectivity metric is not identical to those used to quantify interactions among vegetation patches in earlier geomorphic studies. The weak relationship with neighbourhood structure therefore does not show that dispersal is

unimportant. It does show that the mapped vegetation network explained little additional variation in community composition once broader spatial context and measured habitat conditions were included. This result was similar across the connectivity decay distances we tested, making it unlikely that the pattern arose simply because a single inappropriate decay distance was chosen.

Community composition was more strongly associated with broader spatial context, particularly differences among the four sampled dune fields, although these associations do not by themselves reveal the processes behind them. Geographic structuring is well documented in dune invertebrates. Spider assemblages in North Sea dunes differ across local and regional scales(Bonte et al. 2003, 2006), and recent studies of dune nematodes and other marram-associated invertebrates have also identified biogeographic context, together with local habitat conditions, as an important component of community structure(Van De Walle et al. 2023, 2025). Our results fit this broader pattern, but also show that substantial differentiation occurred among samples from different patches within the same dune field, even though this differentiation was poorly explained by patch area or connectivity. Sandy-beach studies have likewise found strong environmental associations with meiofaunal composition(Pichler et al. 2025), although differences in predictors and sampling design make direct comparisons of effect size difficult.

Several processes could contribute to the differences among our four dune fields, including environmental variation, dispersal limitation, historical differences among locations, and differences in the taxa available to colonise local communities(Cornell and Harrison 2014; Thompson et al. 2020). The measured local habitat variables explained little additional variation once spatial context was taken into account, although their contribution to differences among dune fields cannot be separated from regional context in our design. There is also an important limitation to the interpretation of the regional effect, as each region was represented by a single dune field. The regional term therefore includes unmeasured differences among those four dune fields and cannot be treated as a purely biogeographic effect. In the same way, associations with position within a dune field identify a spatial pattern but not whether it is caused by environmental gradients, dispersal or other spatially structured processes. Our design therefore shows more clearly where compositional differences occur than why they occur.

High biological differentiation despite weak links to patch structure

The strongest evidence for a mismatch between mapped patch structure and community organization comes from combining the patch-level models with the diversity and turnover analyses. Patch area and connectivity explained little additional variation in community composition, but this was not because assemblages were similar among patches. Sampling additional patches within a dune field recovered many OTUs that were absent from the average patch sample, and compositional dissimilarity among patches was high. In other words, communities differed substantially from patch to patch, but those differences were poorly captured by patch area and connectivity. This is an important distinction: a weak relationship between community composition and patch properties can reflect either little biological

differentiation among patches or strong differentiation that follows other, unmeasured features of the landscape. Our results are more consistent with the latter(Riva et al. 2024; Perrin et al. 2025).

Several processes could explain this pattern. Patches with similar mapped geometry may differ in habitat characteristics that we did not measure, including moisture, root-associated resources, burial history or developmental stage. Previous studies in dunes have shown that local vegetation conditions and sand dynamics can influence invertebrate communities(Bonte et al. 2006; Van De Walle et al. 2023, 2025). The vegetation map may also be a poor proxy for connectivity if animals move through bare sand or disperse passively across the apparent boundaries between patches(Fontaneto 2019; Ptatscheck and Traunspurger 2020; Keeley et al. 2022). A further possibility is that present-day vegetation geometry does not reflect the conditions under which communities were assembled. Because we sampled each patch once, we cannot tell whether the observed differences persist through time or whether communities respond with a lag as patches grow, change and merge.

Incomplete sampling may also contribute to the observed dissimilarity. A single pooled sample cannot provide a complete inventory of a patch, and incidence-based measures of beta diversity can be sensitive to incomplete detection(Beck et al. 2013). Some of the differences among patches may therefore reflect undetected taxa rather than true ecological turnover. These possibilities are not mutually exclusive, and the present data cannot separate their relative contributions.

Differences among dune fields added a broader level of compositional differentiation. Samples from different fields were more dissimilar than samples from the same field, and most of this increase reflected taxon turnover rather than nested loss or gain. This is consistent with previous work showing strong local-to-regional differentiation in dune animal communities(Bonte et al. 2003, 2006; Van De Walle et al. 2023, 2025). Together with the differences among patches within dune fields, the richness partition shows that sampling additional patches and additional dune fields continued to add taxa that were not represented in the average standardized patch sample. The turnover component describes these differences in taxon identity, but does not reveal whether they arose through environmental filtering, dispersal limitation, colonisation history or temporal change.

A broadly shared pattern across taxonomic groups

The broad spatial pattern was similar across the major taxonomic groups, although the strength of individual associations varied. Nematodes showed weaker spatial structuring overall, while collembolans were more strongly associated with the surrounding patch network. This variation is not unexpected, given the large differences among meiofaunal taxa in dispersal mode, mobility and dependence on the surrounding substrate(Gansfort et al. 2020).

Differences in body size, life history, dispersal ability and the presence of resistant stages may all contribute to these contrasts(Bertellotti et al. 2023; Alexandre et al. 2025). Passive transport can be important for microscopic animals, and even groups with limited active mobility may disperse over considerable distances through wind or water transport(Ptatscheck et al. 2018; Fontaneto 2019;

Ptatscheck and Traunspurger 2020). This may be particularly relevant in dynamic coastal environments. Our data cannot distinguish among these mechanisms, however, and testing them will require better trait information and finer taxonomic resolution than is currently available for many meiofaunal groups. Limited functional and genetic information, together with incomplete taxonomic databases, remains a major constraint in meiofauna research(Macher et al. 2024; Martínez et al. 2025). Reference libraries that link DNA sequences to reliable taxonomy and ecological traits would make it possible to test more directly why groups differ in their response to landscape structure.

Importantly, the general ordering of spatial associations was similar across groups, suggesting that the whole-community result was not driven by a single major taxonomic group. These comparisons should nevertheless be treated cautiously. The group-specific analyses contained different sets of occupied patches, and estimated effect sizes may also depend on OTU richness and detection. They therefore describe variation in community composition where each group was present, rather than effects of landscape structure on the probability that a group occurred.

From patch effects to biological organization

Incipient dunes provide a useful test of whether mapped landscape structure corresponds to the way biodiversity is organized. Previous work has shown that interactions among neighbouring dune-grass patches contribute to dune development. Our results address a different question: whether patch area and vegetation-based connectivity also describe the distribution of meiofaunal communities. They did so only weakly once broader spatial context and measured habitat conditions were taken into account. The spatial structure involved in dune development therefore does not necessarily describe the spatial structure of the fauna living within those dunes.

This issue extends beyond coastal dunes. Landscape studies have to define spatial units, but units identified from vegetation maps or other visible features may not correspond to functional habitat for the organisms being studied(Manning et al. 2004; Girvetz and Greco 2007). Here, the vegetation structure has a demonstrated role in dune development, yet variation in its mapped properties explained little of the differentiation among meiofaunal communities. The same landscape can therefore have different spatial structures depending on the ecological process being considered.

Understanding why these patterns differ will require information that was not available in the present study. Repeated sampling or experimental manipulation of patches could show how community composition changes as vegetation develops, while direct measurements of dispersal or population-genetic approaches could test whether connections visible in the vegetation map correspond to movement among patches(Keeley et al. 2022; Zeller et al. 2024; Day et al. 2025; Murphy et al. 2025). More detailed environmental measurements could also reveal habitat variation not captured by patch area, connectivity, pH, sediment settlement time or vegetation cover. Structural habitat measures should therefore be validated against the organisms of interest before they are used as indicators of meiofaunal biodiversity(Pierson et al. 2015).

Across the four dune fields studied here, patch area and vegetation-based connectivity added little explanatory information beyond broader spatial context and measured habitat conditions, even though communities differed strongly among patches. Mapped vegetation structure and meiofaunal composition thus describe different aspects of these developing dune landscapes, and a spatial pattern that matters for one ecological process does not necessarily provide a good map of biodiversity.

Declarations

Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Data Availability

Sequence data have been submitted to the NCBI Sequence Read Archive (SRA); the BioProject accession number will be added prior to publication. Drone imagery is available through DataverseNL at https://doi.org/10.34894/EFGAKP.

Funding

This research was funded by the Stemmler Foundation through the MeioMon project awarded to Jan-Niklas Macher.

Author Contribution

Paul M.J. Berghuis and Jan-Niklas Macher conceived and planned the research and conducted the fieldwork. Hannah ter Horst and Jan-Niklas Macher performed the laboratory analyses. Paul M.J. Berghuis, Hannah ter Horst and Jan-Niklas Macher conducted the bioinformatic analyses. All authors contributed to the interpretation of the results and to writing, reviewing and editing the manuscript. All authors read and approved the final manuscript.

Acknowledgement

We thank Joppe Lodewijks (Stichting Amelander Musea) for permission to conduct drone surveys on Ameland and for assistance with transport, and Thomas van der Es (Staatsbosbeheer) for permission to conduct drone surveys on Texel. We also thank the Naturalis laboratory team for their support with laboratory work.

References

1. Alexandre D, Oliveira Filho LCI de, Goss-Souza D, et al (2025) Landscape fragmentation impacts springtail community assembly in subtropical agroecosystems. Appl Soil Ecol 207:Not Available
2. Beck J, Holloway JD, Schwanghart W (2013) Undersampling and the measurement of beta diversity. Methods Ecol Evol 4:370–382
3. Berghuis PMJ, Reijers VC, van de Koppel J, et al (2026a) A connectivity threshold between grass patches amplifies coastal dune formation. Nat Commun 17.: https://doi.org/10.1038/s41467-026-70552-7
4. Berghuis PMJ, Siteur K, Reijers VC, et al (2026b) Density-dependent aggregation underlies spatial self-organization in coastal dunes. Proceedings of the National Academy of Sciences. https://doi.org/10.1073/pnas.2610711123
5. Bertellotti F, Sommer NR, Schmitz OJ, McCary MA (2023) Impacts of habitat connectivity on grassland arthropod metacommunity structure: A field-based experimental test of theory. Ecol Evol 13:e10686
6. Bonte D, Batsleer F, Provoost S, et al (2021) Biomorphogenic feedbacks and the spatial organization of a dominant grass steer dune development. Front Ecol Evol 9.: https://doi.org/10.3389/fevo.2021.761336
7. Bonte D, Criel P, Van Thournout I, Maelfait J-P (2003) Regional and local variation of spider assemblages (Araneae) from coastal grey dunes along the North Sea. J Biogeogr 30:901–911
8. Bonte D, Lens L, Maelfait J-P (2006) Sand dynamics in coastal dune landscapes constrain diversity and life-history characteristics of spiders: Sand dynamics and spiders. J Appl Ecol 43:735–747
9. Buchner D, Macher T-H, Leese F (2022) APSCALE: advanced pipeline for simple yet comprehensive analyses of DNA metabarcoding data. Bioinformatics 38:4817–4819
10. Camacho C, Coulouris G, Avagyan V, et al (2009) BLAST+: architecture and applications. BMC Bioinformatics 10:421
11. Chave J (2013) The problem of pattern and scale in ecology: what have we learned in 20 years? Ecol Lett 16 Suppl 1:4–16
12. Cornell HV, Harrison SP (2014) What are species pools and when are they important? Annu Rev Ecol Evol Syst 45:45–67
13. Cottenie K (2005) Integrating environmental and spatial processes in ecological community dynamics. Ecol Lett 8:1175–1182
14. Crist TO, Veech JA, Gering JC, Summerville KS (2003) Partitioning species diversity across landscapes and regions: a hierarchical analysis of alpha, beta, and gamma diversity. Am Nat 162:734–743
15. Day CC, Zollner PA, Gilbert JH, Gustafson EJ (2025) Integrating mechanistic models of landscape change and animal behavior to measure functional connectivity. Landsc Ecol 40.: https://doi.org/10.1007/s10980-025-02046-8

16. Dennis M, Huck J, Holt C, et al (2026a) Beyond the patch: leveraging functional habitat delineation in fragmentation-biodiversity research. Landsc Ecol 41:37
17. Dennis M, Huck J, Holt C, et al (2026b) In search of Schrödinger's patch: a functional approach to habitat delineation. Landsc Ecol 41:36
18. Dennis M, Huck JJ (2025) The Continuity-Contiguity Problem in Fragmentation-Biodiversity Research. Journal of Biogeography 52:e15077
19. Fahrig L (2003) Effects of habitat fragmentation on biodiversity. Annu Rev Ecol Evol Syst 34:487–515
20. Fontaneto D (2019) Long-distance passive dispersal in microscopic aquatic animals. Mov Ecol 7:10
21. Fu B, Liu Y, Zhao W, Wu J (2025) The emerging "pattern-process-service-sustainability" paradigm in landscape ecology. Landsc Ecol 40.: https://doi.org/10.1007/s10980-025-02063-7
22. Gansfort B, Fontaneto D, Zhai M (2020) Meiofauna as a model to test paradigms of ecological metacommunity theory. Hydrobiologia 847:2645–2663
23. Girvetz EH, Greco SE (2007) How to define a patch: a spatial model for hierarchically delineating organism-specific habitat patches. Landsc Ecol 22:1131–1142
24. Hanski I (1998) Metapopulation dynamics. Nature 396:41–49
25. Hesp PA (1991) Ecological processes and plant adaptations on coastal dunes. J Arid Environ 21:165–191
26. Hillebrand H, Blasius B, Borer ET, et al (2018) Biodiversity change is uncoupled from species richness trends: Consequences for conservation and monitoring. J Appl Ecol 55:169–184
27. Keeley ATH, Beier P, Jenness JS (2022) Connectivity metrics for conservation planning and monitoring. Biol Conserv 255:Not Available
28. Kotliar NB, Wiens JA (1990) Multiple scales of patchiness and patch structure: A hierarchical framework for the study of heterogeneity. Oikos 59:253
29. Leibold MA, Holyoak M, Mouquet N, et al (2004) The metacommunity concept: a framework for multi-scale community ecology. Ecol Lett 7:601–613
30. Leray M, Yang JY, Meyer CP, et al (2013) A new versatile primer set targeting a short fragment of the mitochondrial COI region for metabarcoding metazoan diversity: application for characterizing coral reef fish gut contents. Front Zool 10:34
31. Logue JB, Mouquet N, Peter H, et al (2011) Empirical approaches to metacommunities: a review and comparison with theory. Trends Ecol Evol 26:482–491
32. Macher J-N, Martínez A, Çakir S, et al (2024) Enhancing metabarcoding efficiency and ecological insights through integrated taxonomy and DNA reference barcoding: A case study on beach meiofauna. Mol Ecol Resour 24:e13997
33. Manning AD, Lindenmayer DB, Nix HA (2004) Continua and Umwelt: novel perspectives on viewing landscapes. Oikos 104:621–628

34. Martínez A, Bonaglia S, Di Domenico M, et al (2025) Fundamental questions in meiofauna research highlight how small but ubiquitous animals can improve our understanding of Nature. Commun Biol 8:449
35. Maun MA (1998) Adaptations of plants to burial in coastal sand dunes. Can J Bot 76:713–738
36. Mouquet N, Loreau M (2003) Community patterns in source-sink metacommunities. Am Nat 162:544–557
37. Murphy MO, Farleigh K, Peterman WE, et al (2025) Land-cover patterns differentially affect population genetic structure and connectivity of two anurans. Landsc Ecol 40.: https://doi.org/10.1007/s10980-025-02086-0
38. Nolet C, van Puijenbroek M, Suomalainen J, et al (2018) UAV-imaging to model growth response of marram grass to sand burial: Implications for coastal dune development. Aeolian Res 31:50–61
39. Perrin A, Rein F, Christe P, Pellet J (2025) Habitat fragmentation impact on insect diversity: opposing forces at patch and landscape levels. Landsc Ecol 40:113
40. Pichler M, Creer S, Martínez A, et al (2025) Metacommunity theory and metabarcoding reveal the environmental, spatial and biotic drivers of meiofaunal communities in sandy beaches. Mol Ecol 34:e17733
41. Pierson JC, Barton PS, Lane PW, Lindenmayer DB (2015) Can habitat surrogates predict the response of target species to landscape change? Biol Conserv 184:1–10
42. Ptatscheck C, Gansfort B, Traunspurger W (2018) The extent of wind-mediated dispersal of small metazoans, focusing nematodes. Sci Rep 8:6814
43. Ptatscheck C, Traunspurger W (2020) The ability to get everywhere: dispersal modes of free-living, aquatic nematodes. Hydrobiologia 847:3519–3547
44. Riva F, Fahrig L (2023) Landscape-scale habitat fragmentation is positively related to biodiversity, despite patch-scale ecosystem decay. Ecol Lett 26:268–277
45. Riva F, Pierre E, Guisan A (2024) On the emergence of ecosystem decay: A critical assessment of patch area effects across spatial scales. Biol Conserv 296:Not Available
46. Savary P, Clauzel C, Foltête J-C, et al (2024) Multiple habitat graphs: how connectivity brings forth landscape ecological processes. Landsc Ecol 39.: https://doi.org/10.1007/s10980-024-01947-4
47. Taylor PD, Fahrig L, Henein K, Merriam G (1993) Connectivity is a vital element of landscape structure. Oikos 68:571
48. Thompson PL, Guzman LM, De Meester L, et al (2020) A process-based metacommunity framework linking local and regional scale community ecology. Ecol Lett 23:1314–1329
49. Tischendorf L, Fahrig L (2000) On the usage and measurement of landscape connectivity. Oikos 90:7–19
50. Turner MG (1989) Landscape ecology: The effect of pattern on process. Annu Rev Ecol Syst 20:171–197

51. Van De Walle R, Dahirel M, Langeraert W, et al (2025) Drivers of plant-associated invertebrate community structure in West-European coastal dunes. NPJ Biodivers 4:12
52. Van De Walle R, Massol F, Vandegehuchte ML, Bonte D (2023) The functional composition of dune nematode communities is structured by both biogeographic region and the local marram grass environment. Eur J Soil Biol 117:103511
53. Vanreusel W, Van Dyck H (2007) When functional habitat does not match vegetation types: A resource-based approach to map butterfly habitat. Biol Conserv 135:202–211
54. Wagner HH, Wildi O, Ewald KC (2000) Additive partitioning of plant species diversity in an agricultural mosaic landscape. Landsc Ecol 15:219–227
55. Wangensteen OS, Palacín C, Guardiola M, Turon X (2018) DNA metabarcoding of littoral hard-bottom communities: high diversity and database gaps revealed by two molecular markers. PeerJ 6:e4705
56. Weigand AM, Macher J-N (2018) A DNA metabarcoding protocol for hyporheic freshwater meiofauna: Evaluating highly degenerate COI primers and replication strategy. Metabarcoding Metagenom 2.: https://doi.org/10.3897/mbmg.2.26869
57. Wiens JA (1989) Spatial Scaling in Ecology. Funct Ecol 3:385
58. Wu J (2026) What is landscape scale? Landsc Ecol 41.: https://doi.org/10.1007/s10980-026-02421-z
59. Wu J, Loucks OL (1995) From balance of nature to hierarchical patch dynamics: A paradigm shift in ecology. Q Rev Biol 70:439–466
60. Zeller KA, Compton BW, P. Finn S, Palm EC (2024) Measuring ecological connectivity with ecological distance and dynamic resistant kernels. Landsc Ecol 39.: https://doi.org/10.1007/s10980-024-01890-4

Figures

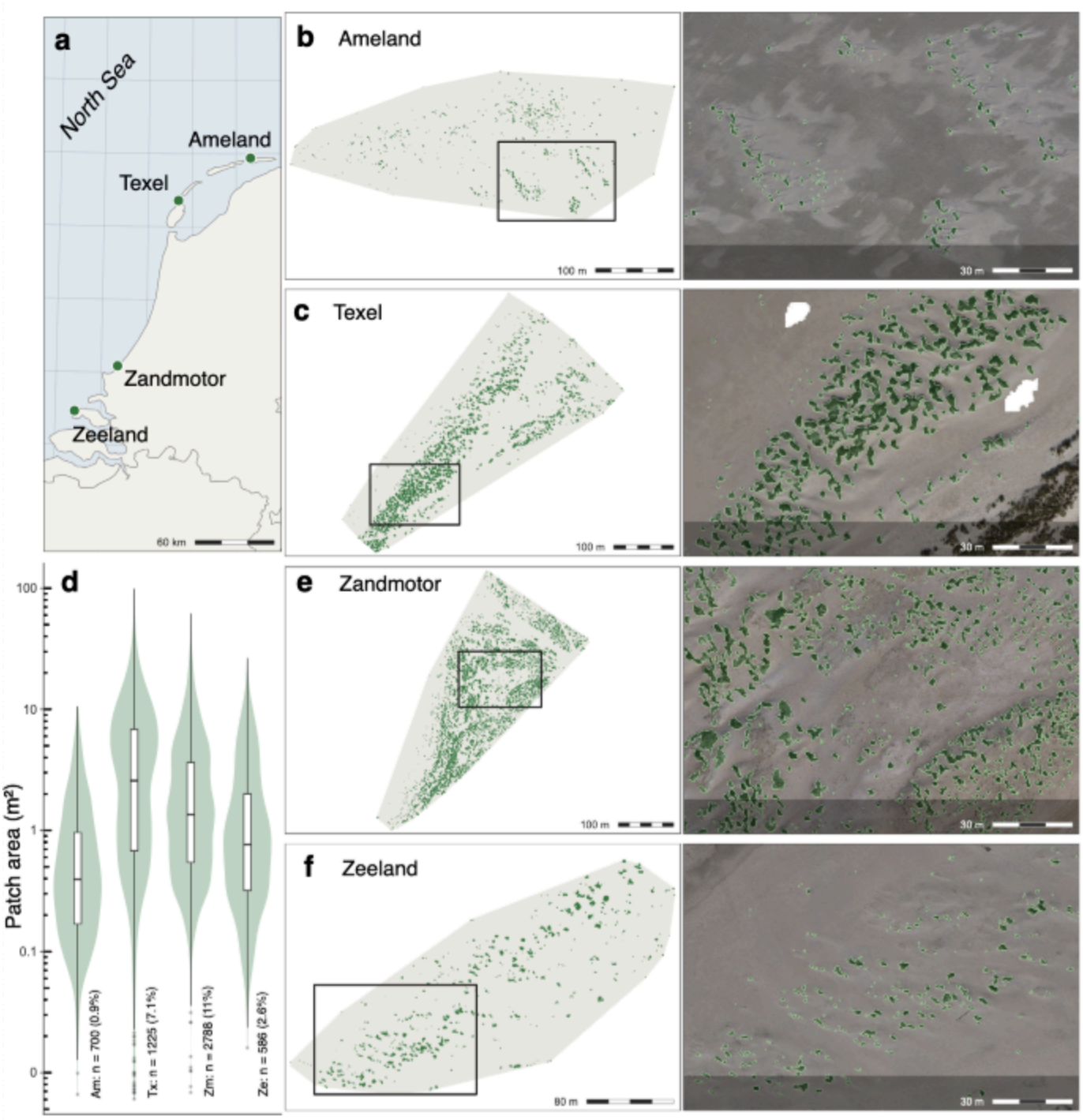


Figure 1

Study regions, mapped dune-grass patches and drone imagery along the Dutch coast. a) Locations of the four study regions, from north to south: Ameland, Texel, the Zandmotor and Zeeland. **b, c, e, f)** Mapped dune-grass patches and corresponding drone imagery for Ameland, Texel, the Zandmotor and Zeeland, respectively. For each region, the left panel shows all mapped dune-grass patches within the surveyed dune field, with the black rectangle indicating the area shown in detail on the right. Right panels show the corresponding orthomosaic with mapped vegetation patches outlined in green. **d)**Distribution of mapped patch areas in each region. Violin plots show the distribution of patch sizes and boxplots indicate the median and interquartile range; note the logarithmic y-axis. The number of mapped patches

(*n*) and the percentage of the surveyed dune-field area covered by vegetation patches are shown for each region

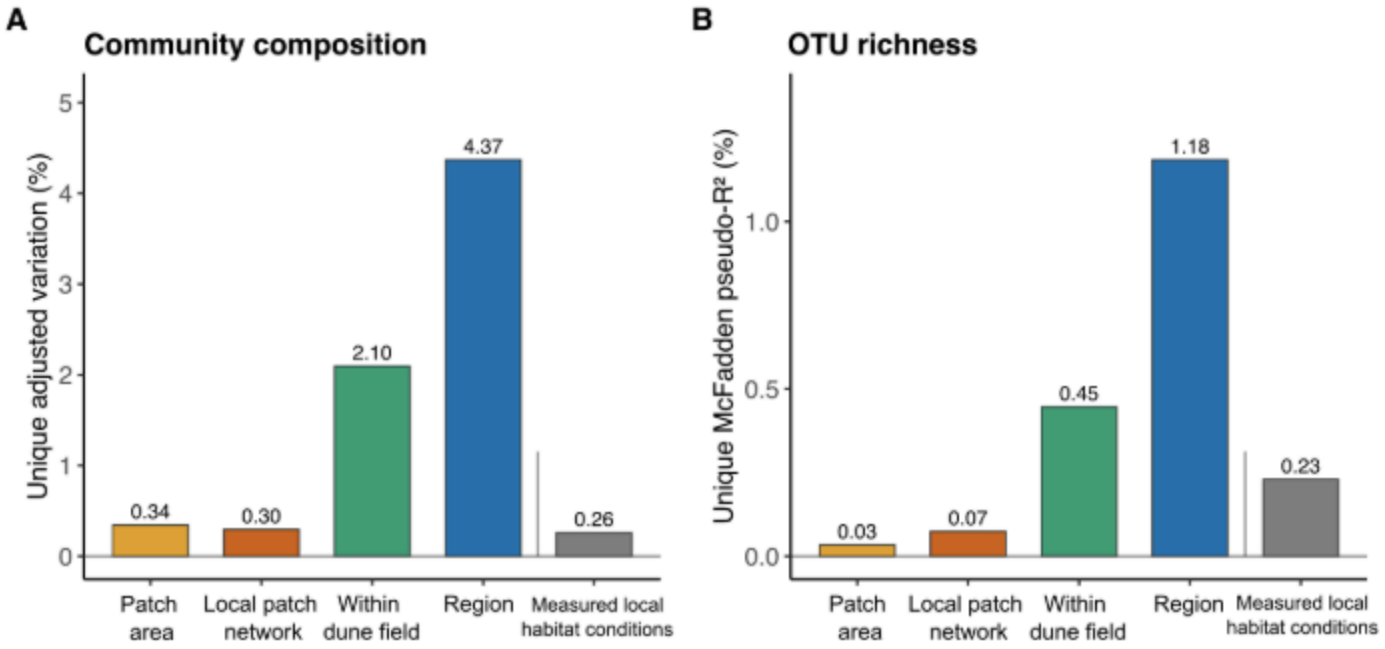


Figure 2

Contribution of landscape structure and local habitat conditions to meiofaunal community composition and OTU richness in incipient dune-grass patches. Bars show the unique contribution of each predictor group after accounting for all other groups. **A)** Community composition was analysed using dbRDA of binary Jaccard dissimilarities, with effect sizes expressed as unique adjusted R^2. **B)** OTU richness was analysed using negative-binomial generalized linear models, with effect sizes expressed as unique changes in McFadden's pseudo-R^2. Patch area represents the focal vegetation patch; local patch network represents structural connectivity with a 10 m decay distance; within-dune-field context includes distance to the coastline, distance to established dunes, and spatial PC1 and PC2; and region represents differences among the four coastal regions. Measured local habitat conditions comprise sediment pH, sediment settlement time and vegetation cover and are shown separately from the spatial hierarchy. Values above bars give the unique effect size for each predictor group, expressed as adjusted R^2 for composition and change in McFadden's pseudo-R^2 for richness

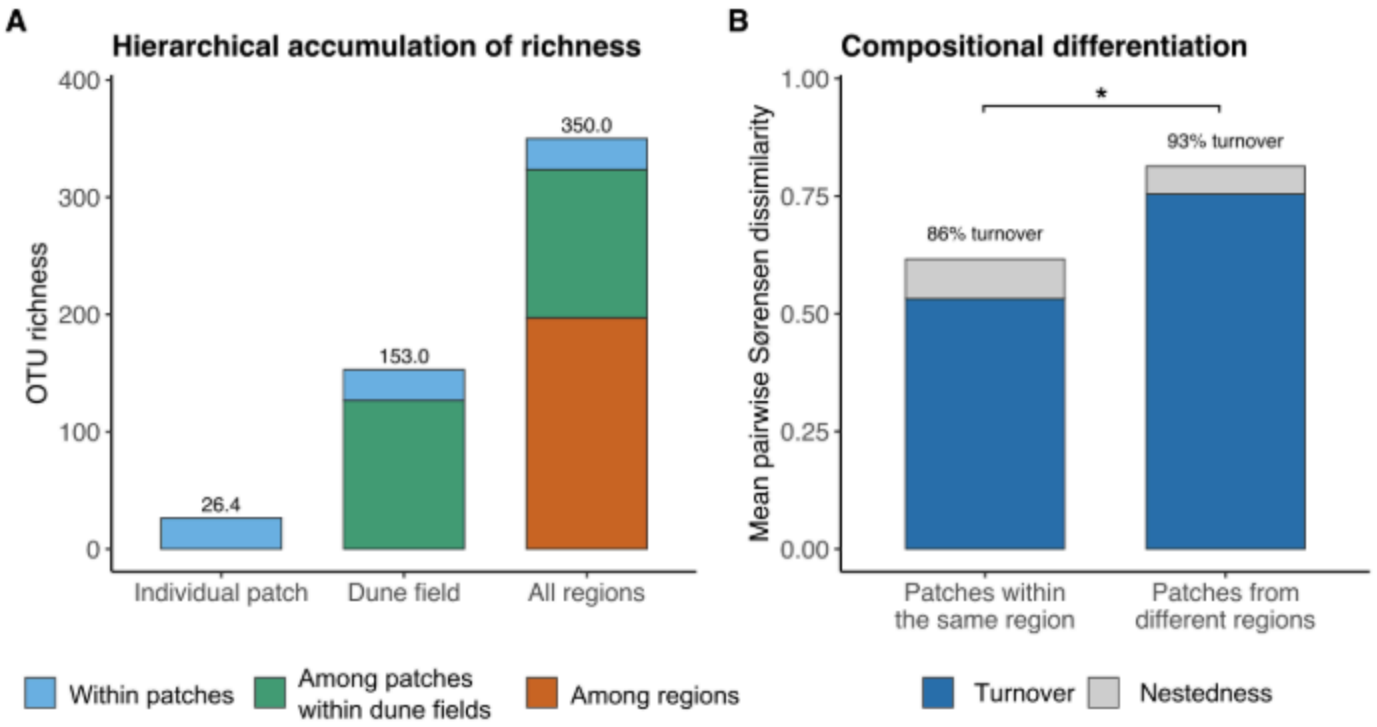


Figure 3

Accumulation and compositional differentiation of diversity across spatial levels.(A) Hierarchical accumulation of OTU richness from individual patches to dune fields and across all four regions. Bars show mean richness within patches, additional richness contributed by differences among patches within dune fields, and additional richness among regions. Values above bars give cumulative OTU richness at each spatial level. Regions were weighted equally in the partition. (B) Mean pairwise Sørensen dissimilarity between patches from the same region and patches from different regions, partitioned into turnover and nestedness components. Percentages above bars indicate the proportion of total dissimilarity attributable to taxon turnover. Patches from different regions were significantly more dissimilar than patches within the same region (Asterisk indicates $P < 0.001$; permutation test with 9,999 permutations)

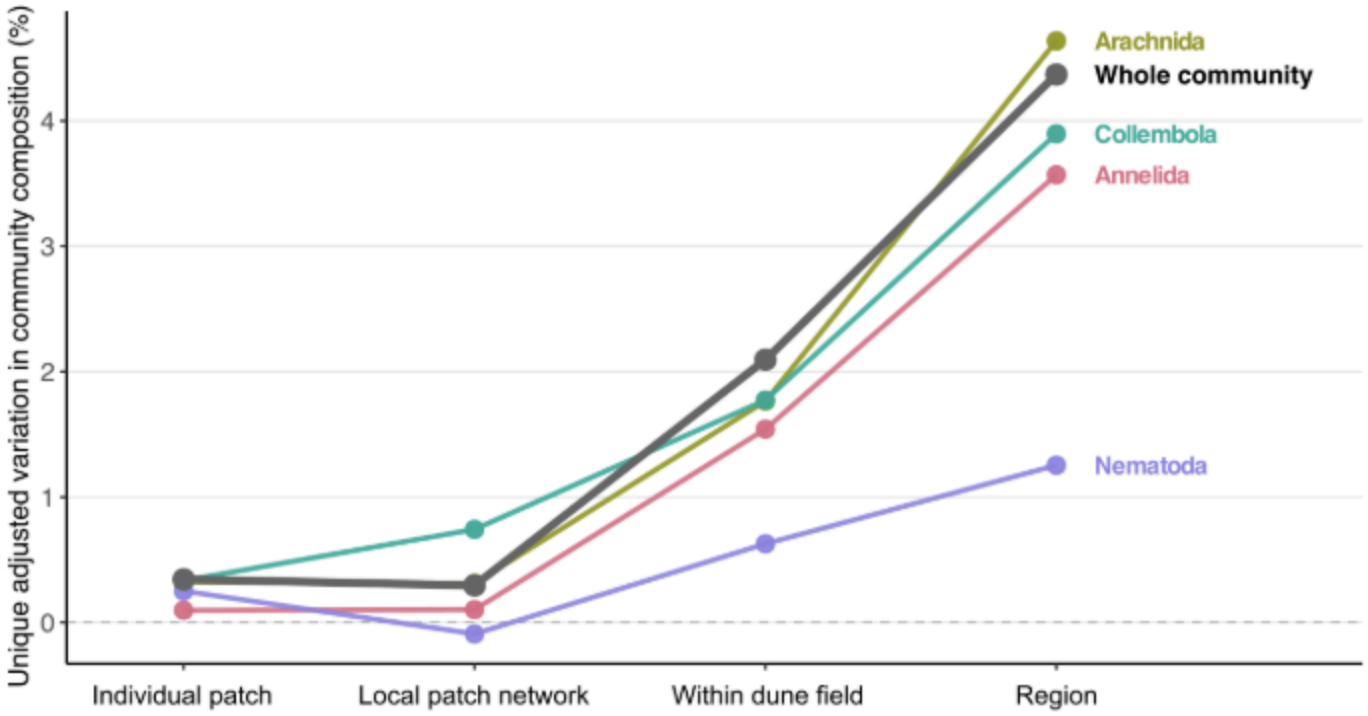


Figure 4

Spatial organization of community composition across major meiofaunal taxonomic groups. Lines show the unique proportion of variation in community composition associated with the focal patch, local patch network, within-dune-field context, and region after accounting for the other spatial levels. Effect sizes are expressed as unique adjusted R^2 from dbRDA of binary Jaccard dissimilarities. Results are shown for the whole meiofaunal community and for the major taxonomic groups retained for group-specific analyses. Most groups showed weak relationships with the individual patch and local patch network and stronger differentiation at broader spatial levels, whereas nematodes showed weaker spatial structuring overall and collembolans somewhat stronger correspondence with the local patch network

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- OnlineResource1FigureS1neighbourhoodradiussensitivity.pdf
- OnlineResource2SupplementaryTables.xlsx