

Biogeographical patterns of soil microbial communities at the scale of French metropolitan territory

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PS.23.001 SPATIAL DISTRIBUTION OF PHOTOSYNTHETIC EFFICIENCY IN MICROBIAL MAT ECOSYSTEMS

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The use of microsensors has revolutionized our understanding of the function of microbial communities and how microbial activity is regulated by biotic and abiotic factors. Although microsensors allow measurements on a µm scale, they represent a limited amount of point measurements that describe complex and inherently heterogeneous microbial communities from a one-dimensional point of view. Thus, it is rather difficult to extrapolate towards quantitative relationships in three dimensions at larger scales [1, 2]. Here, we applied variable chlorophyll fluorescence and hyperspectral imaging for a detailed analysis of the spatial heterogeneity in oxygenic photosynthesis and pigment composition of microbial mats. Additionally, Automated Ribosomal Intergenic Spacer Analysis (ARISA) and statistical methods were used to assess changes in bacterial community structure upon changes in functional and contextual parameters. The combined imaging approach enabled us to co-localize variations in the activity (PSII quantum yield) with the corresponding pigment content for each pixel in the image. Furthermore, statistical analysis quantified activity variations within the same mat sample as well as within samples collected from the same and different geographic regions. Variations in microbial community composition in samples of different geographical origin were explained mainly by quantum yield, E1/2 (the irradiance where half of the maximal quantum yield was observed) and the time between sample collection and measurements. Furthermore, we found a significant variation between samples from the same region. Biogeographical variations between mats collected from different geographical locations, were best described by microbial composition, light adaptations of the microbial mat ecosystems, and the time of sampling. References

1. Jørgensen, B.B. and J.D. des Marais. Limnology and Oceanography, 1990.

2.Kühl, M. and L. Polerecky. quatic Microbial Ecology, 2008.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.002 SMALL SCALE SPATIAL DISTRIBUTION OF MCPA MINERALISATION CHANGES WITH DEPTH IN A DANISH AGRICULTURAL SOIL PROFILE

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Background: Spatial distribution of mineralisation potentials of xenobiotics in soil is typically studied using homogenised soil samples, while little is known about small-scale spatial distribution of specific degradation potentials. Our aim was to study small-scale spatial distributions of phenoxyalkanoic acid (MCPA) mineralisation potentials in a Danish agricultural soil profile.

Method: Arrays of 96 small soil cores (300 µl soil) were sampled in 7 x 10.5 cm grids at five

different depths (8, 28, 48, 85 and 115 cm). Subsamples of 0.20 g from the cores were transferred to deep-well micro plates (96 wells) according to their original sample position. Samples were spiked with ¹⁴C-labelled MCPA (10 mg l⁻¹, 2500 dpm per well) and the plates were sealed with real-time PCR sealing tapes. Each sealing tape was fitted with an array of 96 Ca(OH)₂ impregnated filters to trap the evolved ¹⁴CO₂, and the trapped radioactivity was quantified by digital autoradiography. Real Time qPCR was used to estimate the total microbial populations (16S rDNA genes) and numbers of MCPA degraders (*tfdA* genes) at the five depths. Results: 16S rDNA copy number decreased from 4.3x10⁸ in the top soil to 9.4x10⁷ at a depth of 115cm. The *tfdA* gene copy numbers decreased from 1.2x10⁴ to < 1x10³ MCPA was mineralised in all wells in the top soil (8cm), whereas MCPA was only mineralised in 60% of the samples at 28cm, 4% of the samples at 48cm, 10% of the samples at 85cm and 0% of the samples at 115cm. Geostatistics was applied on data to analyse spatial variation.

Conclusions: The mineralisation of MCPA at each depth was unevenly distributed within the sampling grid with clearly defined hotspots and cold spots. Our study demonstrated an uneven distribution of degrader organism activity and that spatial heterogeneity is most profound in the intermediate soil layers (depth 28-85cm), whereas the top (8cm) and sub soil (115cm) are more homogeneous in respect to MCPA mineralis

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PS.23.003 IMPACT OF PROTOZOA ON 2-METHYL-4-CHLOROPHENOXYACETIC ACID (MCPA) MINERALISATION IN SMALL-VOLUME SOIL SLURRIES

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Mineralisation of xenobiotics is often studied in slurries of soil and water, where the soil structure is broken up and grazing protozoa may have easy access to soil bacteria. We have previously experienced that mineralisation of the phenoxyalcanoic acid herbicide MCPA was considerably lower in soil slurries compared to intact soil, probably due to protozoan grazing. The aim of the present study was to investigate the impact of protozoa on the mineralisation of MCPA in soil slurry samples.

Small-volume soil slurries (200 μ l) and sieved soil samples (200mg) of agricultural top soil (8cm depth) and underlying subsoil (48cm depth) were compared. The impact of protozoan grazing in soil slurry was studied using the protozoan inhibitor cycloheximide (400 mg l⁻¹). Mineralisation was studied by use of ¹⁴C-labelled MCPA, and the numbers of culturable protozoa and MCPA degraders were determined by most probable number methods.

High densities of protozoa $(20.000~g^{-1})$ and MCPA degraders $(2000~g^{-1})$ where found in the top soil, whereas the subsoil contained only low densities of both protozoa $(2400~g^{-1})$ and MCPA degraders $(15~g^{-1})$. Addition of cycloheximide inhibited protozoan activity considerably in both top- and subsoil slurries, but the inhibition led to increased MCPA mineralisation only in the

subsoil. A 100-fold dilution of the top soil, resulting in densities of protozoa and MCPA degraders similar to the subsoil, showed MCPA mineralisation only when the protozoa where inhibited by cycloheximide.

Protozoan grazing caused a severe inhibition of MCPA mineralisation in soil slurry. The inhibition was most pronounced when the density of degrader bacteria was low such as in the subsoil slurry and the 100-fold diluted top soil. Therefore, soil slurry samples should be used with caution in mineralisation studies, especially for soils with low densities of degrader bacteria.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.004 BIOLOGICAL ACTIVITIES IN INDIVIDUAL SOIL AGGREGATES

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Soil is a three-dimensional system made up physically of aggregates of mineral and organic material. The pore spaces within these aggregates serve as potential sites of microbial establishment. However, the small (sub-millimeter) scale of these aggregates makes them difficult to study, even though they may be discrete habitats for microbial communities. We measured the distribution of biomass (measured as ATP) and enzyme activity (β -glucosidase) in 90-member populations of individual aggregates, from each of three size classes (250-425 μ m, 425-841 μ m, and 841-1000 μ m diameter). Aggregate weight, enzyme activity and ATP content were measured sequentially on each individual aggregate within each size population. Additional experiments measuring β -N-acetylglucosaminidase, leucine aminopeptidase, and lipase activity on individual aggregates were also performed. Total activity (per aggregate) was greater for largest size class. . However, when the activities were expressed as a function of the aggregate volume, the smallest aggregates revealed the most intense enzyme activities. There was a weak, but significant, relationship between total ATP (microbial biomass) and β -glucosidase activities; R^2 values were 0.22, 0.51, and 0.43 for the small, medium, and large aggregate categories, respectively (P<0.001).

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.005 FUNCTIONAL SIGNIFICANCE OF NITRIFIER AND DENITRIFIER SPATIAL VARIABILITY IN THREE ARCTIC ECOSYSTEMS

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Permafrost soil ecosystems dominate about one-fifth of the world's and the majority (40%) of the Canadian landscape. Permafrost regions are likely to undergo the greatest degree of climate change and sequestered soil organic matter may be released to the atmosphere in the form of greenhouse gases. Soil microbial transformations contribute 60 to 90% of the world's total nitrous oxide (a potent greenhouse gas) emissions. Owing to the characteristic variability of soil physical and chemical properties nitrifier and denitrifier abundance and their activities may vary across multiple spatial scales. A thorough understanding of this spatial variability is important to assess the relationships between nitrifier and denitrifier communities and their functions. The objective of this study was to investigate the spatial association between nitrifier and denitrifier abundance and nitrification and denitrification potential in three Arctic ecosystems. Following a variable lag-distance design along three transects 93 soil samples were collected at each site. For each soil sample, nitrification and denitrification potential was measured and microbial abundance was estimated by real time PCR. Spatial variability and correlation was assessed by using geostatistical techniques. Ammonia oxidizing bacterial (10^5-10^7) and archaeal (10^6-10^8) and denitrifying bacterial population size (10^5-10^9) is similar to other ecosystems. The potential nitrification and denitrification in Arctic soils is in the similar range as reported in agricultural soils. Nitrifier and denitrifier population in Arctic soils is spatially well structured and they have a significant spatial correlation with nitrification and denitrification potential. This study highlights that nitrifier and denitrifier spatial heterogeneity can play a key role in regulating nitrification and denitrification in Arctic soils.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.006 BACTERIAL COMMUNITY STRUCTURE IN SOIL MICROAGGREGATES AND ON PARTICULATE ORGANIC MATTER FRACTIONS LOCATED OUTSIDE OR INSIDE SOIL MACROAGGREGATES

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Soil aggregates and particulate organic matter (POM) are thought to represent distinct, specific soil habitats for microbial communities. Soil macroaggregates (> 200 μ m), microaggregates and smaller soil fractions (< 200 μ m) have been also described as potentially, relatively stable, habitats for microorganisms. This study investigated whether POM and organo-mineral soil fractions represent distinct microbial habitats and whether the location of these habitats outside or inside macroaggregates affects features such as the amount and quality of organic matter and the structure of bacterial communities. Organic carbon (OC) content and quality expressed as the C:N ratio, and bacterial communities structure (DGGE profiles) were determined for three organo-mineral soil fraction sizes (0-20 μ m, 20-50 μ m, 50-200 μ m) and two POM sizes (> 200 μ m: coarse POM and < 200 μ m: fine POM) located outside or inside water-stable macroaggregates of a well-aggregated cultivated soil. DGGE profiles mainly revealed different bacterial communities on POM fractions and to a lesser extent in organo-mineral soil fractions in comparison to unfractionated soil. Bacterial community structures were strongly correlated to C content (P=0.001, ρ =0.64) and only slightly to C:N ratios (P=0.005, ρ =0.28). There was little

difference in C content, C:N ratios or DGGE profiles in organo-mineral fractions. POM fractions showed different DGGE profiles than organo-mineral fractions. Only coarse POM showed slightly different C:N ratios but clearly different DGGE profiles according to its location. This study showed that POM provides distinct microhabitats, harboring specific bacterial communities in comparison to organo-mineral soil fractions. This may be due to high soil aggregate turnovers in the soil studied or to the C resources in different soil fractions being too similar to reduce the differentiation of the bacterial community structures.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.007 CONNECTIVITY IN INTERMITTENT STREAM SEDIMENTS SUPPORT DYNAMIC BACTERIAL COMMUNITIES

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Watersheds comprise hundreds of small, headwater streams within their boundaries and their biodiversity remains largely underestimated. We assessed the influence of water connectivity on bacterial community composition in an intermittent headwater stream. In situ sediment bacterial communities were described across consecutive seasons in 2007 and 2008 (i.e., dry and wet years, respectively). Although the stream did not hold surface water at several times during the study, sediments remained saturated, especially deeper into the hyporheic zone. Significant variations in bacterial community composition were attributed to spatial and seasonal changes in environmental factors. Prior to the onset of flooding, bacteria were isolated in pools or at certain depths in the hyporheic zone. Communities were observed to change from being rich in operational taxonomic units (OTUs) in isolated pockets at the surface, to a few OTUs overall, including an overall decline in both common and rare bacterial forms. We examined the plausibility of the hyporheic refuge hypothesis for bacteria, which predicts a migration of biota into hyporheic sediments following periods of drought. Only 15% of all OTUs existed in both surface and sediment samples, suggesting that only a few phylotypes may use hyporheic sediments as refuge and were likely transported through connected pore spaces, or isolation at the streambed surface. Changes to connectivity impacted bacterial diversity on rapid timescales (i.e., within days), below-ground and across the sediment-water interface (i.e., the hyporheic zone). Due to the coupling of intermittent streams and their sediments to the surrounding watershed, we stress the importance of understanding connectivity at the pore-scale, consequences to biodiversity and over longer time periods.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.008 MULTI-TEMPORAL SCALE VARIATION OF ENZYME ACTIVITY AND MICROBIAL COMMUNITY COMPOSITION

Hatosy, S*; Neino, V; Lee, J; Tran, T; Bui, H; Heetland, A; Ho, D; League, C; Takagi, B; Kathuria, S; Allison, S; Martiny, A University of California, Irvine, United States

Marine microbial communities in the Southern California Bight are subjected to various influences throughout the year. Upwelling and terrestrial runoff deliver nutrients, seasonal variation affects water temperature, and tidal changes can affect dissolved oxygen and nutrient concentrations. Changes in these influences can result in variations in microbial community composition and enzymatic activity. Similar to spatial biogeographic patterns, is community composition more similar when sampled over shorter time intervals, and does enzyme activity vary at the same time scales as community composition? To address these questions, water samples were collected at time intervals ranging from thirty minutes to months over a one-year period. The water was filtered for enzyme, nutrient, and genetic analysis of microbial communities. Enzyme activity was measured using a fluorometric assay, and communities were analyzed by sequencing the 16S rDNA region. Enzyme activity showed seasonal variation. Of the four enzymes measured most had sharp increases of activity in the winter and gradual increases over spring with declining activity over the summer. Bacterial communities also varied seasonally. The winter was dominated by alphaproteobacteria and cyanobacteria. Bacteroidetes was the dominant taxa during the spring, and the summer was dominated by bacteroidetes and cyanobacteria. Based on the enzyme analysis and the community analysis, both factors follow patterns of seasonal variation which seem to correspond to increased rain-fall in the winter and plankton bloom conditions in the spring. However, further analysis is being completed using similarity matrices. Changes in community composition over various temporal scales will be determined using a Mantel test. Future work will investigate links between community variation and changes in enzyme activity. This work will attempt to understand how changes in community composition influence ecosystem function.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.009 AGGREGATES PROVIDE IMPORTANT MICROHABITATS FOR MICROBIAL COMMUNITIES IN NO-TILL AND CONVENTIONALLY-TILLED AGRICULTURAL SOILS

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Aggregates provide important physical microhabitats for the growth and activity of microorganisms in soil. The three-dimensional structure of soil influences the spatial distribution of water, nutrients and organisms themselves. Tillage management can affect soil microorganisms through changes to the broader soil structure, redistribution of crop residues and by physical disruption of aggregates themselves. We studied microbial biomass and community structure in a long-term tillage experiment comparing no-till and conventional tillage practices at Swift Current, Saskatchewan, Canada. Sampling strategy included the collection of 0-15 cm

cores, segmented into 5 cm depth increments, as well as 0-10 cm samples dry-sieved into five aggregate size fractions. Phospholipid fatty acid analysis was conducted on both bulk soils and aggregates to assess microbial biomass and community structure. Community fingerprinting of 16S and 18S rDNA using denaturing gradient gel electrophoresis was also used to assess bacterial and fungal community structure in the bulk soils. Total microbial biomass and biomass of different functional groups was greater in no-till than conventional-till bulk soils and aggregates. Conversely, no significant tillage-induced shift in microbial community structure was detected in the bulk soils. However, a significant shift was observed between communities in no-till vs. conventional-till aggregates, indicating that tillage management was in fact an important determinant of community structure. Our results highlight the importance of aggregates as microbial habitats under different tillage management practices. Further investigation is needed to elucidate how tillage management affects microbially-driven processes that occur within aggregates in this semi-arid prairie agroecosystem.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.010 IMPROVED *IN SITU* METHODS FOR THE CO-LOCALISATION OF MICROBES IN SOIL

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The physical structure of soil is highly complex and it is this complexity that permits the coexistence of air and water in soil that is essential for supporting terrestrial life. That same structure provides a habitat for plant roots and soil microbial community that represent the most diverse component of the terrestrial biosphere, and is responsible for processing the soil carbon store. Recently, X-ray CT imaging has captured the details of soil pore geometry from which information on water, gas and nutrient delivery can be predicted and related to function using 3D modelling. However, a key limitation of this approach lies in locating the position of microorganisms within the soil. This could be overcome by labelling soil microorganisms with a biomarker able to attenuate X-rays, such as, heavy metals. It may be then possible to co-locate microorganisms that have accumulated these heavy metals using the CT system. To this end, we explored the possibility of using Alexa Fluor 594 Nanogold probe to co-locate micro-organisms within the soil. We initially developed an in situ hybridisation (ISH) protocol for nanogold probing on pure culture (*Pseudomonas*) using combined Fluorescence and Electron microscopy. The combined ISH and Electron Microscopy approach makes it possible to distinguish microorganisms from soil particles and thereby overcoming the problems of autofluorescence normally associated with conventional Fluorescent in situ hybridisation (FISH). The protocol was developed further to locate microorganisms within soil plugs (soil embedded in agarose) and root plugs. This ensured that protocol and sample preparation was optimised before attempting to co-localise microorganisms using CT approaches.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.011 BIOGEOGRAPHIC PATTERNS IN SOIL BACTERIAL AND FUNGAL COMMUNITIES, WITHIN AND AMONG SIX TERRESTRIAL BIOMES

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The relevant spatial scales and biogeochemical factors influencing soil microbial community structure remain unclear. We examined the influence of plant cover, edaphic factors, and distance on variability of fungal and bacterial communities at the hectare scale within six terrestrial biomes (pine, sweetgum, aspen, desert, scrub oak/palmetto, estuary). We also compared compositional similarity between the regionally separated biomes.

Highly replicated rDNA bacterial and fungal community sequence surveys (n=12-18 per site) were compared within and across biomes using multidimensional visualization tools and resampling tests. At two sites, the influence of soil depth was also assessed.

Within each site, we identified edaphic factors that contributed to spatial heterogeneity. At the desert site where plants and soil crusts are patchily distributed, the presence of plants or crusts had a major effect on underlying soil community structure. In the sweetgum, scrub oak, and aspen sites, communities were highly structured by soil depth. Although the trees within the sweetgum, aspen and pine sites were of identical age and spacing, the bacterial community in each replicate sample diverged. Despite the within-site variability, bacterial and fungal community composition was more similar within biomes than between biomes. Each biome contained a distinctive fungal community. In contrast, some overlap occurred with the bacterial communities across the forest sites. Conclusions. Spatial heterogeneity occurs at different scales in these biomes, and is strongly influenced by both plant and soil conditions. Despite local heterogeneity, there appears to be bacterial and fungal meta-communities specific for each biome.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.012 EXTREME HETEROGENEITY AMONG DRY VALLEY SOIL BACTERIAL COMMUNITIES—ANTARCTIC MICROBIOLOGY REVAMPED

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Although recent reports on the microbial ecology of McMurdo Dry Valley soils have rejected the notion that Antarctic Dry Valley soils contain limited microbiota, little is known regarding how these microbial communities are influenced by harsh physicochemical conditions that preclude the existence of vascular plants and macrofauna. Comprising of only 0.03% of Antarctica, the

Dry Valley soil ecosystem is exceptionally simple and provides a highly tractable framework for elucidating interactions between abiotic factors and soil microbial communities. Reported dominance of aeolian transport in biota distribution and extremely low estimated turnover rates contribute to an assumption that Dry Valley soil microbial communities are relatively homogeneous despite clear physicochemical heterogeneity within the region. Here we present evidence that challenges this belief and reveals remarkably localized phylogenetic diversity in Dry Valley soil microbiota. Soil samples collected in four Dry Valleys were analyzed using biogeochemical analyses and DNA-based molecular tools, including pyrosequencing of bacterial 16S rRNA gene PCR amplicons. The results show that the four communities are structurally and phylogenetically distinct, and possess significantly different levels of diversity. Geochemical analyses reveal correlation between physicochemical parameters and compositions of bacterial and cyanobacterial communities. Our results indicate that physical and geochemical factors play major roles in shaping microbiology of ice-free areas of Antarctica, and the astonishingly localized diversities of Dry Valley soil microbial communities indicate extraordinary levels of spatial heterogeneity in an area widely believed to harbor a homogeneous microbial population. These observations underscore the need to re-evaluate current ideas of microbial biogeography and raise issues regarding potential loss of biodiversity induced by changes in macroclimatic conditions in Antarctica.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.013 EFFECTS OF LONG-TERM FERTILIZATION ON SPATIAL DISTRIBUTION PATTERNS OF MICROBIAL FUNCTIONAL GENES FROM THE ROTHAMSTED EXPERIMENTAL SITE

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Understanding the diversity and spatial patterns of microbial communities and the mechanisms shaping biotic communities is a central goal in microbial community ecology. The tendency that species richness increases with area (taxa-area relationships, TARs) is a well-known law in microbial ecology. Here, to test the TARs of soil microbial communities and the effects of long term fertilization and liming, a nested sampling scheme was designed at Rothamted Park Grass field plots with a more than 150-year fertilization history. Soil cores (0-10 cm) were taken at the four corners of the 5 nested squares at increased areas (10 cm², 25 cm², 1 m², 2.5 m², and 5 m²). Plant species in the same area at the same scale were taken to relate the above-ground botanical diversity to the microbial functional diversity. The same sampling scheme was applied on two plots, plot 11/2C with ammonium sulphate and triple superphosphate as fertilizer, and plot 12D with no fertilizer and organic manure. Lime was applied to plot 11/2C since 1863 onto adjust the pH to the same level as that of plot 12D. GeoChip 3.0, covering >37,700 gene sequences from 290 gene families involved in N, C, S, P cycling and other important biogeochemical processes, was used to determine the gene-area relationship (GAR) for both functional and phylogenetic genes. The functional gene numbers were significantly different between the two plots by t-test (P = 0.004). Detrended corresponding analysis (DCA) also indicated that long term fertilization

profoundly changed the microbial functional pattern that the microbial communities were separated by fertilization and non-fertilization. Soil microbial communities exhibited higher z value with long term fertilization, $z_{11/2C} = 0.0864$, than non-fertilization, $z_{11D} = 0.0742$ (P < 0.05), which was in accord with the tendency of plant diversity turnover, $z_{11/2C} = 0.419$ and $z_{11D} = 0.225$.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.014 AQUIFERS - HOMOGENEOUS SATURATED SYSTEMS OR MICROBIAL HABITATS WITH SURPRISING FINE-SCALE SPATIAL AND TEMPORAL HETEROGENEITY?

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Aguifers deliver a number of important ecosystem services to our society, amongst the most important of which are the sustaining of water resources and the attenuation of anthropogenic contamination. In dichotomy to this relevance, the scientific understanding of the ecological principles controlling aquifer functioning, especially as habitats for a diverse intrinsic microbiota, is still in its infancy. Classically, aquifers are considered as systems low in spatial or temporal heterogeneity. Here, we challenge this perception based on fine-scale qualitative and quantitative microbial community data obtained by depth- and time-resolved sampling of sediments at a BTEX-contaminated aquifer. By T-RFLP fingerprinting, qPCR and 454 Pyrosequencing of ribosomal and functional marker gene fragments we show, that pronounced cm-scale distinctions actually govern the structure and distribution of aquifer microbiota. Especially in different compartments of the BTEX-plume, as well as at biogeochemical redox gradients, characteristic microbial community patterns were established. When functionally dissected for dominating respiratory and catabolic potentials, these were surprisingly well correlated to the fine-scale localization of respective processes. Thus, plume zones characterized by highest anaerobic BTEX degradation activity were coupled to the dominance of distinct anoxic degrader populations, as well as to high catabolic (bssA) gene abundances. At the oxic/anoxic capillary fringe, cm-scale distinctions in the sequential detectability of typical aerobic degrader lineages, thiotrophic chemolithoautotrophs, as well as anaerobic respiratory guilds unveiled unparalleled fine-scale heterogeneities in aquifer microbial community distribution. We discuss how these findings can help to understand the controls of natural attenuation at contaminated sites, and whether similar heterogeneity may be found also in noncontaminated groundwater environments.

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23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.015 CENTIMETER SCALE ANOXIC SITES AND N₂O DYNAMICS IN SOIL Markfoged, R*¹; Nielsen, L¹; Nyord, T²; Ottosen, L¹; Revsbech, N¹

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Periodic depletions of O₂ due to rainfall or application of animal slurry is an essential factor in the control of N₂O production in the soil environment. In this study distribution of O₂ and N₂O in the soil was monitored with microsensors during a four week period following injection of animal slurry in tracks five cm below the surface. Emission rates of N₂O from the soil surface were measured by the closed chamber method. A combination of high O₂ consumption and a low diffusivity (8*10⁻⁴ m⁻² s⁻¹) created an anoxic 3-cm thick cylindrical soil volume which persisted for 7 days. Two N₂O emission peaks from the soil surface were detected with the emission chamber; first a high transient peak 1 day after slurry injection and secondly a longer lasting peak after 6-17 days. However, with the microsensors N₂O was only detected during the first two days and only within the anoxic cylinder. The anoxic cylinder functioned as a hotspot for denitrification of the NO₃ originally in the soil, and denitrification was responsible for the N₂O production causing the first emission peak. Transport limitation in the saturated cylinder resulted in accumulation of N₂O within the anoxic volume and delayed the N₂O emission peak as compared to the net N₂O production. Depletion of the NO₃ in the cylinder two days after injection caused a shift from net N₂O production to net consumption and thereby resulted in a rapid exhaustion of the accumulated N₂O and a consequently cease of the N₂O emission. It was calculated that 20-30% of the NO₃ initially present in the anoxic volume was emitted as N₂O. The second hot-moment of net N₂O production and emission was ascribed to increasing nitrification in the aerated soil and coupled nitrification-denitrification in the periphery of the anoxic cylinder. This study illustrates directly how the N₂O emission from the soil is controlled by temporal and spatial variations in diffusivity as well as the availability of O₂, NO₃, NH₄ and organic carbon

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.016 SPATIAL VARIABILITY OF NITROUS-OXIDE PRODUCTION, DENITRIFICATION GENES AND DENITRIFICATION ENZYME ACTIVITY IN A TABLE GRAPE VINEYARD

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Nitrous-oxide (N₂O), a potent greenhouse gas contributes to ozone layer depletion and global climate change. Denitrifying bacteria are major controllers of N₂O, through their ability to perform either complete or incomplete denitrification based on their production of either dinitrogen or N₂O as an end product, respectively. Previous studies, in a California table grape vineyard had determined large N₂O fluxes in the berm compared to the row. The goal of this study was to determine if the ratio of denitrification genes (*nir:nos*) would correlate with N₂O fluxes. Denitrification enzyme activity in the berm ranged from 10.3-11.5 ng N₂O-N min⁻¹ g dry soil⁻¹ and from 1.1-1.8 ng N₂O-N min⁻¹ g dry soil⁻¹ in rows. This corresponds to a 7.5 fold increase in denitrification activity in the berm. Denitrification genes were quantified using quantitative polymerase chain reaction and reported as a percent of 16S rRNA. There were 7-10% nirS genes in the row and 20-50% in the berm; 0.6-0.7% nirK genes in the row and 1-1% in the Berm; 0.4-0.5% nosZ genes in the row and 0.3% in the Berm. N₂O reductase (nosZ) was

enriched 1.7 times in the row compared to the berm whereas nitrite reductase, nirS and nirK, were enriched 3.0 and 1.6 times in the berm compared to the row, respectively. The ratio of nirS:nirK indicated that there was a preference for nirS in the berm. The ratio of nir:nos was 17-21 in the row and 80-244 in the berm which correlates with the N_2O fluxes measured in the field and the denitrification enzyme activity data. Whereas nosZ gene abundances were inversely correlated with denitrification enzyme activity and N_2O fluxes recorded in the field. The abundance of denitrification genes can be used to predict overall denitrification rates and incomplete denitrification, N_2O production.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.017 ARE MEASURED NITRIFICATION RATES A GOOD INDICATOR OF SOIL HEALTH?

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The concept of soil health encompasses the complex interactions of plants, hydrology, and nutrient cycling. Microbial indicators have been highlighted as superior in determining the healthy functioning of the soil microhabitat, due to their rapid response to environmental change. Modern molecular methods allow for rigorous assessment of soil health through their ability to measure both biodiversity and functional processes in the soil environment. Microbial health restoration and maintenance in new soils created from Brownfield land remediation, is important for flexible and safe end uses such as recreation and residential housing. Currently soil health guidelines do not exist for reclamation of contaminated land. This study aimed to define rapid high throughput indicators for use in contaminated land assessment and remediation that directly link functional metabolic processes to community diversity. A range of soils have been analysed to evaluate the effects of pollutants on basal soil health characteristics. Soil health has been monitored through the measurement of three main soil functions: respiration, nitrification and denitrification rates. Key populations related to soil health have been assessed using cultureindependent community analysis (Polymerase Chain Reaction, real time PCR and Denaturing Gel Gradient Electrophoresis) to target 16S rRNA and functional genes (amoA, nirS/K). In this study, Potential Nitrification Rates (PNR) did not indicate significant effects of contamination upon function despite measurable changes in community diversity. PNR measurements of highly contaminated soils show rates, often equating to and exceeding uncontaminated soils. Functional gene profiling of Ammonia oxidizing Bacteria and Archaea identified strong reductions in diversity with contamination. This indicates that PNR measurements may be insensitive to impacts on soil health in polluted environments.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.018 COMMUNITY DIVERSITY IN INDIVIDUAL SOIL AGGREGATES

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Soil aggregates are one example of a discrete habitat experienced by soil microbial communities. These aggregates are the building blocks of soils, and highly stable individual microaggregates bind together to form more dynamic, larger aggregates (up to 2 mm diameter and larger). These individual and compound aggregates thus potentially form different habitat types, and may feature different microbial community structures and functions. We therefore examined the microbial community structures in samples of individual aggregates, organized by size (250-425 µm, 425-841 µm, and 841-1000 µm). We developed an approach to extract DNA from individual soil aggregates, weighing between 0.03 and 0.5 mg. The 16S rDNA from 16 individual aggregates, as well as pools of aggregates, was amplified and sequenced using barcoded universal primers (27F and 338R). Over 314,000 sequences were obtained, assigned to Operational Taxonomic Units (OTUs), and used to examine the community diversity among and between individual aggregates. There was substantial variability in the number and identity of OTUs identified in individual aggregates, indicating considerable beta-diversity (differences in community composition between sites) at the scale of soil aggregates.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.019 DISTURBANCE FREQUENCY AFFECTS MORPHOLOGY AND COMMUNITY COMPOSITION OF A BIOFILM

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Biofilms are spatially heterogeneous at the scale of micrometers in terms of microbial composition, physiological state, and biofilm morphology. It is often neglected that maturing biofilms in a flow field also develop a heterogeneous spatial structure on the order of millimetres and centimetres. At this larger scale, the heterogeneity of biofilms is likely an important parameter affecting substrate diffusion into the biofilm and consequently influences the availability of substrate to the microbial community. How disturbance frequency, in our case the interval of monochloramine addition, influences the evolution of biofilm morphology and bacterial community structure on a larger scale is the objective of our study.

Multispecies biofilms are grown on defined synthetic substrate in three 5-liters bubble column reactors. While one reactor serves as control without biocide addition, two reactors receive a biocide pulses daily and every other day. In each reactor, over a period of 2 months, biofilm is sampled on 133 removable coupons for the acquisition of images with a size of nearly 4 cm2 and for DNA extractions. DNA and images allow us to perform community fingerprinting using single strand conformation polymorphisms and automated image analysis of biofilm morphology.

We showed in initial experiments that a minimum image size is necessary to capture heterogeneities in biofilm morphology relevant for mass transfer and thus substrate availability. This size is larger than typical dimensions of images acquired with e.g. confocal microscopes.

We introduce a novel strategy for large-scale biofilm imaging to relate biofilm inhabitants to macroscopic heterogeneities on the order of millimetres and centimetres. In three parallel reactors, we relate microbial diversity to disturbance frequency and biofilm morphology.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.020 LINKING BACTERIAL ABUNDANCE TO HUMAN DENSITY AT A CONTINENTAL SCALE

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Accessing the global importance of bacteria and their major regulating factors require knowledge of their abundance at large spatial scales, only by which general patterns could emerge. Published comparative analysis at large spatial scales were performed using local environmental factors as independent variables, but no attempt was performed using a broader, landscape approach, that also includes the human impacts on aquatic ecosystems. We demonstrated that there is a strong-positive relation between bacterial and human densities when analyzing a unique large-continental data set. Bacterioplankton samples (N = 1,160) were collected in large rivers, lakes and reservoirs of the 12 largest Brazilian hydrographical regions (from Brazil das Águas project, www.brasildasaguas.com.br), which encompass the overall Brazilian territory and approximately 50% of South American continent. Bacteria were counted by flow-cytometry, while human density in each region was obtained by the last Brazilian census. We observed a positive-linear relation between aquatic bacterial abundances and human densities in hydrographic regions. Overall, almost 50% of variation of aquatic bacterial abundance may be explained by this sole regional factor, which is surprising since several local factors may independently, or combined, regulate natural bacterial abundances. This whole idea seems to be trivial or well-documented, however the direct relationship between these smallest organisms of from aquatic ecosystems and human occupation in drainage areas is unlikely and was never shown, since several local factors acting on lower scales usually regulate bacterial abundances and processes

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.021 NEOTROPICAL ISLAND BIOGEOGRAPHY FOR SULFATE-REDUCING BACTERIA ACROSS MANGROVES

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Global phylogeography across disturbed sediments has revealed a patchy distribution for sulfatereducing bacteria (SRB). However, a distinctive cluster was formed by several Neotropical mangrove samples. Mangroves swamps represent a major habitat for SRB while prevailing in the tropical and subtropical shorelines. Our objective is to examine the heterogeneity of SRB in mangroves along the coast of Puerto Rico to assess their island biogeography. Sulfidogenic community composition was characterized with terminal restriction fragment length polymorphisms of the dissimilatory sulfite reductase gene (dsr-TRFLP) amplified from soil/sediment samples. A total of 2640 phylotypes (TRF) were detected (representing 373 different TRF). The sulfidogenic community ranged from 8 (Vieques) to 103 (Loiza). No TRF was common to all samples analyzed so far. In contrast, 14% were detected only in one sample (potential endemic taxa). Few major peaks and minor peaks dominated the overall communities. Similarity analysis, based on the Sorensen's index, illustrated clustering together with minimal geographical proximity. Additional samples to process and application of multivariate analyses will provide a more complete description of the island biogeography for SRB in Neotropical mangroves. Sulfidogenic communities sustain a patchy distribution within the continue contour of the island likely due to biogeochemical conditions, geomorphological characteristics, and hydrological regimes of each sites. The application of dsr-TRFLP has provided a description of the functional diversity for SRB in one of the most relevant habitats in Earth. The genetic pool being disclosed will assist in understanding the prevailing taxa and resilience of the sites to environmental disturbances and changes.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.022 DIVERSITY AND DISTRIBUTION OF SULFATE-REDUCING BACTERIA AT THE JOBOS BAY NATIONAL ESTUARINE RESERVE (PUERTO RICO, USA)

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The Jobos Bay National Estuarine Research Reserve has suffered anthropogenic and natural disturbances that affect environmental quality compromising its sustainability. Government agencies monitor its water quality and sediments chemistry, and benthic habitats. The chemical character of water and sediments is greatly influenced by microbial activities. Sulfate-reducing bacteria (SRB) predominate in marine sediments and play key roles in biogeochemical cycles, including transformations of pollutants. In order to understand the contributions of SRB to the environmental quality of the sediments, we examine the diversity and distribution of SRB along transect of sediments monitored. SRB were described using terminal restriction fragment length polymorphism analysis of dissimilatory sulfite reductase genes (dsr-TRFLP). A total of 1388 phylotypes (TRF) were detected (representing 273 different TRF). The sulfidogenic community ranged from 10 (most polluted area) to 98 (most pristine area) TRF. No TRF was common to all samples analyzed so far. In contrast, 22.7% were detected only in one sample (potential endemic taxa). Few major peaks and many minor peaks dominated the overall communities. Similarity analysis, based on the Sorensen's index, illustrated clustering together with minimal geographical proximity. Additional samples to process, clonal sequencing, and application of multivariate

analyses coupled to chemical data will provide a more complete description of the benthic biogeography for SRB in the estuary. Sulfidogenic communities sustain a patchy distribution within the sediments examined likely due to biogeochemical conditions influenced by anthropogenic influences. This study will help to understand SRB's roles in natural attenuation of contaminants and dynamics within the marine sediments of the Jobos Bay, specifically the impact of non-point source pollution in SRB diversity and distribution.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.023 EFFECTS OF DISTURBANCE AND RECOLONIZATION ON BENTHIC MICROALGAL COMMUNITY STRUCTURE AND SPATIAL PATTERNS

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Benthic microalgae (BMA) are important primary producers in intertidal and shallow subtidal sediments, serving as a vital food resource for heterotrophs. BMA also release extracellular polymeric secretions that stabilize sediments. Primary objectives of the research reported here were to assess the effects of biotic and abiotic disturbances on microalgal community structure in sediments, and to determine mechanisms by which BMA recover from such disturbances.

Using field comparative studies, we characterized the effects on BMA of two prominent types of disturbance: macroinvertebrate ingestion and tidal resuspension. BMA biomass and composition in fecal materials from the enteropneust, *Balanoglossus aurantiacus*, and sediments were followed through time using fluorometry, microscopy and molecular techniques (DGGE and sequencing). We also determined experimentally the mechanisms and rates of recolonization of disturbed sediment patches, comparing regrowth, recruitment and immigration. Last, we examined effects of disturbance on spatial patterns of BMA biomass over the larger sedimentary landscape using correlative studies.

Deposit-feeder ingestion was found to significantly reduce BMA biomass and alter BMA composition, although qualitative changes were comparatively less remarkable. BMA biomass recovery was significant in < 3h, with migration dominating. A significant difference was also found between average biomass before and after tidal immersion, a physical disturbance. Spatial autocorrelation revealed heterogeneity in BMA distribution during low tide, and that this distribution correlated with *B. aurantiacus* fecal coils. Samples taken after tidal immersion showed no patchiness, nor was there a correlation between BMA biomass and pre-immersion fecal cover. At least in intertidal habitats, the qualitative and quantitative impacts of deposit feeding, and the resulting landscape-scale patchiness, appear to be short-lived due to frequent tidal resuspension.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.024 BIOGEOGRAPHICAL PATTERNS OF SOIL MICROBIAL COMMUNITIES AT THE SCALE OF FRENCH METROPOLITAN TERRITORY

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Ecologists studying plant and animals have long recognized that the modifications of diversity across a landscape are central for understanding the environmental factors driving the magnitude and the variability of community assembly. Despite the statement that this conceptual vision is also relevant for microorganisms, since it can offer valuable insights into the relative influence of dispersal limitations, environmental heterogeneity, and environmental and evolutionary changes in shaping the structure of ecological communities, studies integrating wide spatial scale have been poorly investigated and the environmental filters structuring microbial biodiversity remain largely unknown. In this context, we have investigated the characterisation of indigenous microbial communities from soils sampled on a broad scale. This characterisation relied on analyses of density, diversity and genetic structure of microbial communities by using molecular tools directly on DNA extracted from the soil library of RMQS ("Rseau de Mesures de la Qualit des Sols" = French soil quality monitoring network) which cross-rules all the French territory with about 2,200 soils sampled. Geo-positioning, physico-chemical characteristics, climatic factors, floristic composition and land use of these soils were also recorded. The geostatistical interpolation of soil microbial density and diversity revealed a heterogeneous distribution of microbial communities on a wide extent which is spatially structured in bio geographical patterns. Microbial density and diversity seems to be more influenced by local filters such as soil properties and land use but is relatively independent of global filters such as climatic factors or the presence of natural barriers. Our study confirms that the biogeography of microorganisms differs fundamentally from the biogeography of "macro-organisms" and that soil management can have significant large-scale effects.

Abstract Category
23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.025 THE ROLE OF SPATIAL HETEROGENEITY ON PRODUCTIVITY/DIVERSITY RELATION IN A MICROBIAL BIOFILM

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A strong correlation between ecosystem's productivity and diversity has been proposed in many studies. However, there is a lack of agreement concerning the pattern of this relation (unimodal, positive linear, negative linear), as well as the processes behind these patterns. Changes on the levels of heterogeneity on resource availability have been proposed to explain, at least, the decrease on diversity levels in an unimodal ("hump-shaped") relation. Nevertheless, due to ecosystem complexity, heterogeneity has not been characterized and therefore its role in shaping these patterns has not been determined. Bacterial systems provide an excellent model where populations and communities can be propagated in controlled environments in order to reduce

ecosystem complexity. In this work we varied the degree of heterogeneity in the limiting resource (oxygen) in tubes with LB broth, by two means: 1) shake the tubes or leave them static 2) using different diameter tubes, changing the area of the air-broth interface. We inoculated a Pseudomonas sp. in the different tubes and after four days it diversifies into different morphs each one with a particular niche. After plating a sample of each tube, we measured both, productivity and diversity. Heterogeneity was calculated as the variance in oxygen concentration (measured with a microelectrode) inside the tubes. Results show that there is no relation between productivity and diversity in homogeneous environments. In contrast, in heterogeneous environments there is a high influence of resource availability on both: diversity and productivity. We conclude that there is an important effect of spatial heterogeneity in shaping the relation between productivity and diversity.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.026 IMPACT OF DISTURBANCE ON SOIL MICROBIAL ACTIVITY IN THE NORTHERN CHIHUAHUAN DESERT

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Cryptobiotic soil crusts in arid regions contribute to ecosystem stability through increased water infiltration, soil aggregate stability, and nutrient cycling between the soil community and vascular plants. These crusts are particularly sensitive to compaction/fracturing disturbances such as livestock grazing, off-road vehicle use, trampling by humans, and drilling and mining activities. Loss of soil crusts is believed to increase the rate of desertification, and recent findings indicate that crusts are extremely slow to recover, on the order of hundreds of years. However, post-disturbance recovery rates for soil bacterial and fungal populations is vastly understudied, and it is suggested that loss of soil crusts leads to decreased abundance and diversity of these non-crust soil biota. Soil microbial activity within and around two natural gas well pads embedded within a livestock grazing area were investigated. During the natural gas extraction process, the shallow surface soil is stockpiled near the well pad. Surface and subsurface soil samples were taken from the heavily compacted well pad, the surface soil stockpile (fallow 12yr), and the grazed area outside of the well pad. Microbial activity was measured using the MicroResp[™] system, which measures respiration of microbes within whole soil samples supplemented with various carbon sources (simple and polymeric sugars, amino acids, carboxylic acids, and fatty acids). Preliminary results indicate slightly reduced activity in heavily disturbed well pad areas compared to grazed areas, although overall activity for all samples was extremely low, and not significantly different from controls. Activity was marginally higher in surface soils (top 5cm) compared to subsurface soils (5-30cm). Greatest microbial activity was measured in the soil stockpile at a depth of 5-30cm. Further testing is underway to compare microbial activity in these disturbed areas to samples from nearby areas with intact crusts.

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.027 KARST POOLS IN SUBSURFACE ENVIRONMENTS: COLLECTORS OF MICROBIAL DIVERSITY OR TEMPORARY RESIDENCE BETWEEN HABITAT TYPES

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Groundwater is a major source of drinking water for a significant proportion of the world's population. Subterraneous karst aquifers are structurally diverse and they may play an important role in groundwater formation. So far little is known about the microorganisms inhabiting karstderived subsurface aquatic habitats and about the factors influencing microbial assemblages in such environment. We studied bacterial diversity and community composition in three shallow pools of a Swiss karst cave system with contrasting hydrological and hydrochemical properties. A comparative analysis of 16S rRNA genes showed remarkable differences between the microbial assemblages in the pools. Only one operational taxonomic unit (OTU, 97% similarity) was shared between the three of them (total OTU number in all pools: 150). Unexpectedly high microbial phylotype richness was found even in the two pools without groundwater contact and with low concentrations of organic carbon and total cell numbers (< 10⁴ ml⁻¹). One of these seepage water fed systems harbored 15 distinct OTUs from several deeply branching lineages of the candidate phylum OP3, whereas representatives of OP3 were not found in the other two pools. A tentative phylogeographic analysis of available OP3-related sequences in the context of our data set revealed that there was generally little agreement between the habitats of origin of closely related sequence types. Two bacterial clades affiliated with the obligate methylamine utilizer Methylotenera mobilis were only found in the pool that was exposed to repeated flooding events. Direct microscopic analyses by fluorescence in situ hybridization revealed that these bacteria formed relatively stable populations of up to 6% of total cell counts over periods of several months irrespective of inundation by groundwater. This suggests that karst water may provide a means of transport for these bacteria from terrestrial to freshwater habitats.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.028 AN INDIVIDUAL-BASED APPROACH TO SURFACE COLONIZATION BY BACTERIA

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Most microbial habitats are characterized by physical, chemical, and biological conditions that differ significantly along micrometer distances. This environmental heterogeneity at the scale of individual microbes may account for much of the nonrandom distribution of microorganisms observed in natural settings. Here we report on an individual-based approach that combines spatially explicit modeling and bioreporter-driven experimentation to improve our basic understanding of the impact of small-scale heterogeneity on the bacterial colonization of

surfaces. We used bioreporters of *Erwinia herbicola* 299R tagged with the green fluorescent protein (GFP) in two ways. One type of bioreporter constitutively expressed GFP which allowed for easy visualization of the location of bacteria in relation to one another. In the second type of bioreporter, preformed GFP was diluted from cells as they divided, rendering GFP content of daughter cells an inverse function of the reproductive success of their mothers. These bioreporter cells were inoculated on a defined agarose surface and analyzed by fluorescence microscopy to measure bacterial growth and colony formation during incubation. *E. herbicola* cells all grew at the same rate on the surface of the gel medium, regardless of the bacterial spatial density. This suggests that under these conditions, growth of bacteria was not influenced by the presence of others nearby. Fluorescence intensity and surface area measurements of the reproductive success were in good correlation. The pattern simulation matched the experimental data when it introduced initial variations in the cell cycle of colonizers. This approach of validating bioreporter output in simple environments with defined complexity are essential for our ability to explain their performance in more complex environments, such as the natural habitat of *E. herbicola* 299R, i.e. the plant leaf surface.

Abstract Category
23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.029 BIODIVERSITY OF THE WORLD'S MOST ALKALINE ENVIRONMENT: SOIL AND GROUNDWATER COMPARISON

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The microorganisms in the groundwater of the Lake Calumet wetlands, Chicago, IL, have been described as the world's most extreme alkaliphiles. Over the 100 years, the wetlands have been infilled with steel slag. The slags were deposited from local blast furnace mills and were comprised primarily of high-temperature calcium silicate minerals and could contain as much as 50% metallic iron, manganese, and other steel additives (Cr, Mo, V) which can vary in composition within in meters of each other.. The weathering of the calcium silicates within the slag subsequently formed an aquifer with groundwater pH up to 12.8. Previous research on the microbial community has been performed on the groundwater but nothing has been done to examine this community in the soils in contact with this high pH groundwater. The objective of this research project is to determine the structure of the microbial communities at different locations with the Calumet aguifer systems and to determine if the microbial communities are similar in the groundwater and the soils. Preliminary screening of three soils and two groundwater samples with utilizing sole source carbon utilization plates (Biolog® Ecoplates) revealed low microbial diversity. It also determined that the microbial community in the soil was significantly different than that found in the soil especially in highest pH setting. In addition, the spatial distribution of the microbial community in relationship to the mineralogy was determined using scanning electron microscopy. It was determine that the different chemical composition of the aquifer materials impacted the biofilm that formed on the soil associated with the extreme alkaphile conditions. The microbial community will also be examined using different molecular

techniques. The findings of this study will reveal novel insights into the extreme alkaliphilic microbiology.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.030 A LANDSCAPTE PERSPECTIVE ON ANTIBIOTIC RESISTANCE IN SOIL BACTERIA

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The extent, pattern, and causes of antibiotic resistant bacteria in soil are not well understood. Antibiotic resistant in the environment may arise from the use of antibiotics in agricultural practices, from indirect selection for resistance to heavy metals, and from natural interactions among microbes. We performed a novel study of the large-scale distribution of antibiotic resistance in soil bacteria. We determined the relationship of resistance in Enterobacter aerogenes to land use and concentration of metals during July of 2007 and 2009 in Lancaster County, PA, USA. Sites were randomly chosen within each land use. Microbial analyses involved isolation and replica plating of 48 isolates per site. Metal analyses were performed with a MS-ICP. From soils collected from 84 sites in 2007, we found mean proportions of isolates resistant to ampicillin (0.50), chloramphenicol (0.49), kanamycin (0.05), and tetracycline (0.03). From soils collected from 73 sites in 2009, we found mean proportions of isolates resistant to ampicillin (0.64), chloramphenicol (0.12), trimethroprim-sulfamethoxazole (0.04), kanamycin (0.002), and tetracycline (0.02). Levels of resistance were typically lower in forest versus other land uses. In both years, bacteria from forests had significantly less resistance to ampicillin (2007: F3,80 = 6.7, p = 0.001; 2009: F3,69 = 5.6, p = 0.002) than those in residential, crop, or pasture sites. In 2007, bacteria from forests and residential sites had significantly less resistance to kanamycin (F1,79 = 11.88, p = 0.001) than from agricultural sites. We did not detect an effect of soil metal concentration on antibiotic resistance. We document microbial resistance in a human-dominated landscape and provide an assessment of variables affecting that resistance. A greater effort should be made to understand the relationship between land use, soil characteristics, and resistance and the implications of these relationships on public health.

Abstract Category

23 Spatial Heterogeneity - Small Volumes with Big Impacts

PS.23.031 INFLUENCE OF SOIL STRUCTURE ON MICROBIAL INTERACTIONS AND DIVERSITY

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Soil habits are extremely heterogeneous and are inhabited by a tremendous number and diversity of microorganisms. Although recent studies have provided insight into some primary drivers of soil-borne microbial community structure, little is known about how the physical structure of soil

influences the establishment and maintenance of microbial diversity. Soil structure and stability determines the connectivity (flow of chemical compounds such as nutrients, antibiotics, and signaling compounds) of soil-borne microhabitats, thereby influencing bacterial interactions such as resource- and interference competition. In a series of microcosm experiments, we therefore sought to gain insight into the influence of soil structure and soil particle connectivity on microbial diversity. We hypothesized that small-scale spatial isolation fosters greater biodiversity by allowing for the co-existence of otherwise competing species. To test this hypothesis, a series of artificial soil communities were constructed, using different numbers of soil isolates introduced into microcosms of different pore-size-distributions with different levels of mixing. Resulting communities were followed in time, and population densities tracked by specific plate counting and quantitative real-time PCR. These communities were also examined by fluorescent in situ hybridization (FISH) to assess the physical distribution of the different populations in the developing microcosms. Experiments involved inoculation with two different hierarchies of postulated competitive interactions: perfectly intransitive competition (i.e. strain A beats B, B beats C and C beats A) and perfectly hierarchical competition (A beats B, B beats C, etc.). Results show that spatial isolation at the micro-scale can greatly affect competitive relationships, thereby stabilizing microbial community composition and diversity.

Abstract Category
23 Spatial Heterogeneity - Small Volumes with Big Impacts