Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-01 会場:105 時間:5 月 26 日 09:00-09:15

富士山麓における降雨の湧水への直接的な影響のシグナルを追跡するための複合的な解析
Multiple analyses to chase the signature of direct impact of rainfall into groundwater in Mt. Fuji

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A huge amount of groundwater is stored in subsurface environment of Mt. Fuji, the largest volcanic (basalt) mountain in Japan. There distribute many springs flushing out between lava (high permeability) and underlying older lava (low permeability) at various points of the foot. Based on the concept of piston flow transport an apparent residence time was estimated to ca. 30 years by ^{36}Cl/Cl ratio (Tosaki et al., 2011). However, this number represents an averaged value of the residence time of groundwater mixed before flushing out. On the other hand, we found that pH of spring water in the lower part of the foot of Mt. Fuji decreased shortly after the typhoon in August 2011 which suggested the newly supplied rainwater was mixed into groundwater. Thus, we try to chase signature of direct impact of rainfall into groundwater from multiple analyses to elucidate the routes of groundwater under the torrential rainfall. Though analyses of groundwater chemistry show just an averaged value, microbial DNA analysis could suggest the routes of transport; if thermophilic microbial DNA is detected this suggests at least a part of groundwater must originated from the environment >ca. 40^{o}C (=600 m deep in Mt. Fuji). Thus, we employed three different tracers; stable isotopic analysis (δ^{18}O and δD), chemical analysis (concentration of silica) and microbial DNA analysis.

Stable oxygen isotopic ratio of shallow groundwater became higher than usual value reflecting torrential rainfall and the concentration of silica decreased after the torrential rainfall amounting more than 300 mm. In addition, the density of Prokaryotes in shallow groundwater apparently increased. These findings indicate a direct impact of rainfall into groundwater was observed after torrential rainfall with more than 300 mm in the studied geological setting. This did not appear when rainfall did not exceed 100 mm/day. Increase in the density of Archaea at deep groundwater after the torrential rainfall suggests a possible mixing of deep groundwater which was pushed by piston flow transport as an indirect impact of rainfall into deep groundwater, if it is true to this geological setting that Archaea is predominant in deep subsurface environment as was suggested. Microbial DNA can give possible information about the route of groundwater.

キーワード: 地下水, 強い降雨, 湧水の異常出水
Keywords: groundwater, strong rainfall, abnormal flash out

Japan Geoscience Union Meeting 2015
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BGM22-02 会場:105 時間:5 月 26 日 09:15-09:30

環境中には未知なる脱窒微生物が豊富に存在している
Higher diversity and abundance of denitrifying microorganisms in environments than considered previously

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Denitrification is an important process in the global nitrogen cycle. The genes encoding NirK and NirS (*nirK* and nirS), which catalyze the reduction of nitrite to nitric oxide, have been used as marker genes to study the ecological behavior of denitrifiers in environments. However, conventional polymerase chain reaction (PCR) primers can only detect a limited range of the phylogenetically diverse *nirK* and *nirS*. Thus, we developed new PCR primers covering the diverse nirK and nirS. Clone library and qPCR analysis using the primers showed that nirK and nirS in terrestrial environments are more phylogenetically diverse and 2-6 times more abundant than those revealed with the conventional primers. RNA- and culture-based analyses using a cropland soil also suggested that microorganisms with previously unconsidered *nirK* or *nirS* are responsible for denitrification in the soil. PCR techniques still have a greater capacity for the deep analysis of target genes than PCR-independent methods including metagenome analysis, although efforts are needed to minimize the PCR biases. The methodology and the insights obtained here should allow us to achieve a more precise understanding of the ecological behavior of denitrifiers and facilitate more precise estimate of denitrification in environments.

キーワード: 脱窒, 亜硝酸還元酵素遺伝子, nirS, nirK
Keywords: denitrification, nitrite reductase gene, nirS, nirK

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-03 会場:105 時間:5 月 26 日 09:30-09:45

土壌微生物バイオマスの窒素安定同位体比：ストイキオメトリーと同位体比の接点
Soil microbes shape nitrogen isotopic signatures of soils: a linkage between the ecological stoichiometry and d15N.

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Nitrogen (N) is an essential, although ecologically limiting, nutrient in many terrestrial ecosystems. It is thus critically important to understand N cycles in terrestrial ecosystems to project their responses to expected changes in environments such as the increase in anthropogenic N input and CO2 concentration. Natural abundance of N isotopes (d15N) has been used to get insights into N cycles in the ecosystems because the d15N signature can provide unique information on the naturally-occuring processes in the intact ecosystem. Interpretations of global dataset of plant d15N (e.g. Craine et al. 2009) and soil d15N (e.g. Houlton and Bai 2009, Craine et al. 2015) have been proposed to explore the important flux/parameter in N cycles which are difficult to measure (such as N availability and denitrification loss). In most of these cases, the rule of the thumb in d15N interpretation is that soil loses 15N-depleted N during decomposition (more strictly, mineralization and leaching/denitrification loss), which is also the fundamental concept for marine sediment d15N (e.g. Robinson et al. 2012, Tesdal et al. 2013). Even this ”15N-depleted N loss” concept is easy to follow, the direct (experimental) evidence for the isotopic fractionation during N mineralization or decomposition is suprisingly scarse. Although long-term lab incubation of soil samples revealed the expected d15N increase with the decrease in N concentration (Nadelhoffer and Fry 1988), litter-bag experiments (Melillo et al. 1989; Connin et al. 2001) did not show this expected d15N change during litter decomposition. Thus the gap between field observations and lab experiments in the d15N trend calls the review of the fundamental concept for the interpretation of soil d15N.

In the presentation, I will summarize the d15N data we obtained in the last five years on soil bulk N, several extractable organic N (EON), extractable inorganic N (EIN) in soils and soil microbial biomass (SMB), which are now relatively easy to measure with denitrifier method (Sigman et al. 2001, Houlton et al. 2006). The d15N of SMB is generally higher than d15N of other N compounds, which should be interpreted as a consequence of carbon and N stoichiometry (or N mineralization; Dijkstra et al. 2008). This high d15N of SMB can complement the interpretationo of soil d15N variations – the large d15N differences between organic layers and mineral soils often observed in soil profiles, the low d15N in wet/cold ecosystems and the high d15N in dry/hot ecosystems in the global soil d15N trend, and the high d15N of the microbially-processed soil fractions.

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BGM22-04 会場:105 時間:5 月 26 日 09:45-10:00

アナモックスの優占する活性汚泥における同位体分別
Isotopic fractionations during nitrogen removal in the activated sludge

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Anammox is considered to be an important nitrogen removal pathway in the ecosystem. However, it is still unknown how much the anammox can contribute to the total nitrogen loss in the ecosystem. Natural abundance of stable isotopes can be a promising tool to investigate the relative contribution of anammox and denitrification in the intact ecosystem, although the isotopic fractionation factors during anammox which are necessary to interpret isotopic signatures are not fully known. Here we reported nitrogen and oxygen isotopic fractionation factors during anammox occuring in the activated sludge. We incubated the sludge anaerobically to trace the changes in concentrations and isotopic signatures of ammonium, nitrite and nitrate during the anammox process. We found the large isotopic fractionations for ammonium oxidation and nitrite reduction by anammox. In addition, the inverse isotopic fractionation during nitrite oxidation to nitrate was observed. We will discuss these factors with comparison of the latest study on anammox isotopic systematics (Brunner et al. 2013) in the presentation.

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BGM22-05 会場:105 時間:5 月 26 日 10:00-10:15

1 細胞ゲノム解析から見る海洋性アンモニア酸化アーキアの環境適応機構
Single cell genomic analysis for the marine ammonia-oxidizing archaea

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アンモニア酸化アーキア (AOA) は、光が充分に届かない海洋水塊中微生物生態系において最も優占する系統群であり、しばしば、全微生物数の数十%を占める。これまでの研究により、水塊中に棲息する海洋性 AOA は、系統群毎に適した電子供与体（アンモニア・尿素等）濃度・フラックスに応じた棲み分けを行っていることが示唆されている。一方において、これまでに培養された海洋性 AOA は、比較的高いアンモニア濃度・フラックスに適応した特定系統群に限られており、その性状に関する情報は極めて限られている。

1 細胞ゲノム増幅及びそのシーケンス解析は、未培養系統群の代謝、ゲノム構造、そして進化を明らかにする有力な手段の一つである。本研究では、伊豆小笠原海溝の海洋表層から海溝底（9697 m）に至る計 6 深度から 1 細胞ゲノム増幅ライブラリーを構築し、SSU rRNA 遺伝子の PCR 増幅により、AOA 由来のゲノム増幅産物を確定した。さらに、アンモニア酸化に関わる ammonia monooxygenase や urease 遺伝子を対象に PCR 増幅を試みた。そして、一連の遺伝子系統解析情報に基づき、505・5010・9,697 m の 3 深度において、それぞれ優占する遺伝子型群を示すゲノム増幅産物を対象に、全ゲノムシーケンス解析を進めている。 1 連の解析より明らかにされた深海環境に棲息する AOA の棲み分けの仕組み、その背景にあるゲノム上の特徴について紹介する。

キーワード: ゲノム, アンモニア酸化, アーキア
Keywords: genome, ammonia oxidation, archaea

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-06 会場:105 時間:5 月 26 日 10:15-10:30

軽希土類元素と細菌 C1 代謝：Bradyrhizobium 属細菌のメタノールデヒドロゲナーゼ
Rare earth elements and bacterial C1 metabolic system

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軽希土類元素（La, Ce, Nd など）はレアアースとも呼ばれ、宇宙存在比はケイ素の 100 万分の 1 程度と極めて低いが、土壌も含めた地殻存在度はその数十倍で、数十 ppm に達する。Methylobacterium. extorquens AM1 のメタノールデヒドロゲナーゼ遺伝子 xoxF の産物は、Ca によって活性を得る MxaF と異なり、La や Ce のような希土類元素存在下で活性を示すことが報告された (Nakagawa et al. 2012)。Bradyrhizobium 属細菌には、マメ科植物の根粒菌だけでなく、非マメ科植物のイネ根などにエンドファイトとして生息している。そこで、本研究では、これらの Bradyrhizobium 属細菌の C1 代謝系に及ぼす希土類元素の影響を検討した。Bradyrhizobium 属細菌における xoxF, mxaF の有無を確認するために、クサネム根粒菌である B. oligotrophicum S58, Bradyrhizobium sp. BTAi1, Bradyrhizobium sp. ORS278、イネ根から分離された Bradyrhizobium sp. RP5, Bradyrhizobium sp. RP7, Bradyrhizobium sp. WD16 の 6 株のゲノム情報より、xoxF と mxaF (M. extorquens AM1 株由来) の配列を対象とした BLASTN による遺伝子検索を行った。その結果、対象菌株全てが xoxF を有すること、およびイネ根から分離された 3 株 (RP5, RP7, WD16) は xoxF と mxaF の両方を有することが予想された。供試菌のメタノール資化能に及ぼす希土類元素の影響を検討した。炭素源を除いた液体の HM 培地 5 ml にメタノールを 0, 0.1, 1, 10% (v/v) の 4 段階添加し、La 添加区 (30 μ M) と Ce 添加区 (30 μ M)、非添加区を設けた。6 株 (S58, BTAi1, ORS278, RP5, RP7, WD16) を液体 HM 培地で前培養し、OD660 を 24 時間毎に測定した。その結果、xoxF のみを有するクサネム根粒菌 3 株 (S58, BTAi1, ORS278) では La もしくは Ce を添加し且つメタノール濃度 0.1% と 1%の条件で生育が観察された。xoxF と mxaF の両方を有するイネ根分離菌 3 株 (RP5, RP7, WD16) は La, Ce の添加の有無に関わらずメタノール濃度 0.1 と 1%の条件で生育が見られた。B. oligotrophicum S58 株を対象に、ΔxoxF::Ω (xoxF 遺伝子のΩカセット挿入変異) と ΔxoxF (xoxF 遺伝子のマーカーレス欠損変異) の 2 種類の変異株を作製し、培養実験を行った。その結果、ΔxoxF::Ω変異株では希土類元素存在下で野生株に比べ生育は低下したが、ΔxoxF 変異株では逆に野生株よりも生育が促進された。野生株において La 添加区メタノール濃度 0.1%の系で、濁度の上昇と共に培養液中のメタノール濃度が減少した。一方、ΔxoxF::Ω変異株では野生株に比べメタノール濃度の減少は抑制され、ΔxoxF 変異株は野生株と同様のメタノール濃度の減少が見られた。供試したクサネム根粒菌 Bradyrhizobium 属 3 株 (S58、BTAi1、ORS278) は mxaF を持たず xoxF を有し、0.1~1%のメタノールを添加した HM 培養液において La, Ce 依存的な生育を示した。一方、イネ根分離 Bradyrhizobium 属 3 株 (RP5、RP7、WD16) は、mxaF と xoxF を保有し、La, Ce 非依存的な生育を示した。この結果より、xoxF のみを持つクサネム根粒菌 3 株がメタノール依存的な生育に La, Ce が必要であることが明らかになった。しかし、S58 株の?xoxF::Ω変異株はメタノール依存的な生育能力が低下し、ΔxoxF 変異株ではメタノール資化能が失われず、かつ La, Ce 依存的に生育の促進が観察された。これらのことから、S58 株が xoxF とは異なる新規の希土類元素依存的メタノールデヒドロゲナーゼ遺伝子を保持していることが強く示唆された。また ΔxoxF::Ω変異株の生育能力の低下は、Ωカセット挿入によってアルデヒドデヒドロゲナーゼなどの C1 代謝系に関わる下流遺伝子の転写が抑制されたことによると考えられた。以上より、植物共生 Bradyrhizobium 属細菌には広く xoxF 遺伝子が存在し、特にクサネム根粒菌は希土類元素依存的なメタノール代謝能を示すこと、および新規の希土類元素依存的メタノール資化に関わる遺伝子の存在が示唆された。本発表では、希土類元素と微生物の関係について生物地球化学視点からも議論したい。

キーワード: 希土類元素, 細菌, C1 代謝, Bradyrhizobium, メタノール, メタノールデヒドロゲナーゼ
Keywords: Rare earth elements, Bacteria, C1 compound metabolism, Bradyrhizobium, Methanol, Methanol dehydrogenase

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-07 会場:105 時間:5 月 26 日 11:00-11:15

クロロフィルの光毒性：光合成生物からのエネルギーフラックスを制約する光生化学的因子
Phototoxicity of chlorophylls: a major photobiochemical constraint on the energy flux from photosyntheis

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光合成の仕組みを系内に獲得して以降，太陽からの光のエネルギーを主要なソースとしたエネルギーフラックスが地球生命圏を駆動する状態へとシフトした。この地球生命の光合成は，クロロフィル類という光増感剤を用いたエネルギー転換の機構が根幹をなしている。

【光毒性問題：酸素とクロロフィル光合成のパラドックス】
光合成電子伝達においては細胞外から電子供与体が要求されるが，初期の光合成では硫化水素などの還元的分子種が利用された。これに代わって遍在する水分子から電子を奪う仕組み，すなわち酸素発生型光合成の登場に至って，光合成からのエネルギーフラックスが飛躍的に重要度を増したと概略的には看做される。しかし，その代償として発生する分子酸素は，クロロフィル類とは非常に相性が悪い。すなわち，クロロフィル類は通常の分子酸素［三重項酸素］を強力な活性酸素［一重項酸素］に励起させる光増感作用を有し[1]，これは生命に致死的なダメージを与えうる（クロロフィルの光毒性）。現在の植物（シアノバクテリアと真核植物）はこのクロロフィルの光毒性を回避する巧妙な仕組みを備えているが[1]，最初に還元的な大気下で酸素発生型光合成を始めた生物は，あらかじめその「巧妙な仕組み」を備えていなければならなかったはずである。

【クロロフィルの「解毒」：植物の捕食と光毒性問題】
光合成からのエネルギーフラックスが生態系には，植物が産生する有機物が従属栄養生物の細胞内に取り込まれるステップが必須である。現在の水圏環境でも重要な役割を果たす捕食性のプロティスト（単細胞体制の真核生物）は，食胞作用を通して光合成生物を補食するが，多細胞生物登場以前の初期の水圏生態系においては，これが特に重要なプロセスであったことは想像に難くない。しかしこれは，クロロフィルを含有する物体を光が透過する細胞内に取り込む行為であり，光毒性に対する厳重な制御機構が必須である。しかし見かけ上，これら生物にクロロフィルの光毒性は顕在化しない。我々は近年，植食性のプロティストがクロロフィルの光毒性を無効化する代謝プロセスを発見し，その仕組みの一端がようやく明かされてきた。すなわち，水圏環境で微細藻類を捕食するプロティストは食胞形成と消化の進行の過程で，クロロフィル類を光毒性のない有色化合物 13^2,17^3-シクロフェオフォルバイドエノール（シクロエノール）に代謝する[2,3]。この代謝は，ほぼ全ての真核生物のスーパーグループ間に共有されていることも分かった。シクロエノールは分析条件下では非常に不安定であり，従来，定量分析はおろか検出すら困難であったが，実際にはあらゆる水圏環境に多量に存在する（底泥中にはクロロフィルを遙かに凌駕する量が含まれる）。一連の発見は，（１）微細藻類を捕食するプロティストが現在の水圏環境においても量的に重要であることを示し，（２）それを可能とする「シクロエノール代謝」が，真核生物による酸素発生型光合成生物（初期には特にシアノバクテリア？）の直接捕食を通した繁栄と多様化を駆動する重要な因子であったことをも示唆する。

【真核植物の進化とクロロフィル光毒性の制御】
複雑な体制を構築できる真核生物が，シアノバクテリアを細胞内取り込み制御することで酸素発生型の光合成の機構を獲得したことで（葉緑体の獲得：一次植物の進化），光合成からのエネルギーフラックスは飛躍的に重要度を増したと想像される。色素体の獲得は大量のクロロフィル類を細胞内で保持する大きなリスクを負うため，それらの光毒性を制御する生理機構を発達させる必要がある。さらに，様々な系統の真核生物は，緑色植物や紅色植物の一次植物を細胞内に取り込み葉緑体化することで，「植物化」のという進化を繰り返してきた（二次植物の進化）。二次植物化は，食胞作用による植物の細胞内への取り込み過程の延長線上にあったとみなされている。いくつかの二次植物の系統では，自ら細胞内でシクロエノールを産生するが[4,5]，これは光毒性の制御との関連性が考えられる。特に，光栄養性のユーグレノイドにおいては，自己の色素体の分解に際し不要となるクロロフィルを光無毒性のシクロエノールへ代謝しており，祖先的な捕食性のユーグレノイドのシクロエノール代謝の仕組みを，自らの細胞内で産生されるようになったクロロフィルに対する「安全装置」として維持することで植物化を可能としたのかもしれない。

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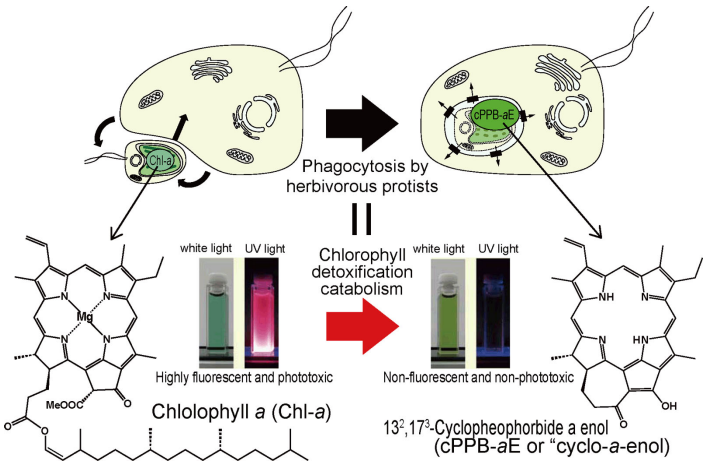
キーワード: クロロフィル, 酸素, 光毒性, プロティスト, 微細藻類, シクロフェオフォルバイドエノール
Keywords: chlorophyll, oxygen, phototoxicity, protist, microalgae, cyclopheophorbide enol



Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-08 会場:105 時間:5 月 26 日 11:15-11:30

アンチモン微生物変換と環境動態における影響
Microbiological transformation of antimony and its geochemical implications

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Antimony (Sb) is a naturally occurring toxic element and is considered to be a priority pollutant of interest by the USEPA. Although the concentrations of Sb in soils are generally low ($<$1 mg kg^{-1}), elevated levels of Sb have been released via mining activities and other anthropogenic activities due to its increasing industrial use. Antimony is commonly associated with arsenic (As) in the environment and both elements have similar chemistry and toxicity. Increasing numbers of studies have focused on microbial roles in As transformations, while microbial-Sb interactions are still not well understood. To gain insight into microbial roles in the geochemical cycling of Sb, soils from an old stibnite (Sb_2S_3) mine tailing area (Ichinokawa mine, Ehime, Japan) were characterized geochemically and examined for the presence of Sb-transforming microbial populations. Total concentrations of Sb and As were higher in the surface soil (0-3 cm: 2280 and 1240 mg kg^{-1}, respectively) and decreased with depth (9-12 cm: 330 and 133 mg kg^{-1}). Bacterial community profiles, examined by cultivation-independent analysis using 16S rRNA gene-based denaturing gradient gel electrophoresis, did not show substantial differences through depth (0-12 cm). After the aerobic enrichment culturing with Sb(III) (100 μM), pure cultures of *Pseudomonas*- and *Stenotrophomonas*-related isolates with Sb(III) oxidation activities were obtained. Anaerobic enrichment cultures capable of reducing Sb(V) (2 mM) were also obtained, in which the precipitation of antimonite [Sb(III)] as antimony trioxide was observed. These results demonstrate that indigenous microorganisms associated with stibnite mine soils are capable of Sb redox transformations and contribute to the speciation and mobility of Sb *in situ*.

キーワード: アンチモン, ヒ素, アンチモン還元微生物, アンチモン酸化微生物, 土壌微生物群集
Keywords: Antimony, Arsenic, Microbial antimony oxidation, Microbial antimony reduction, Soil bacterial community

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-09 会場:105 時間:5 月 26 日 11:30-11:45

薩摩硫黄島の浅海性熱水マウンドにおける鉄酸化微生物群集の分子生態学的・鉱物学的特徴
Ecological and mineralogical characteristics of Fe-oxidizing microbial communities in a shallow hydrothermal mound

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Microbial Fe-oxidation has been mainly studied at deep-sea hydrothermal vents because the low concentration of oxygen and high concentration of ferrous ion was preferable for Fe-oxidizing bacteria that have to compete with abiotic Fe-Oxidation. However, microbial Fe-oxidization in shallow, fully oxygenated environments has been still largely unknown. In this study, we aim to reveal the ecology and role for mound formation of Fe-oxidizing bacteria at iron oxyhydroxide-rich hydrothermal mounds developing in Nagahama Bay, Satsuma-Iwojima where we observed dense assembly of twisted stalks, typical signature of microbial Fe-oxidation. Core samples were taken from the iron oxyhydroxide-rish mound and used for sequencing and microscopic analysis. Microscopic observation indicated the highest occurrence of stalk structure was observed at around 20 cm from the surface. Sequencing of 16S rRNA gene of prokaryotic communities (>100,000 reads/sample) revealed that Anaerolineae known as obligately anaerobic heterotroph was highly dominated at ˜40% throughout all depths down to 40 cm from the surface of the mound, inferring anaerobic circumstances in the sediment. We also found Fe-oxidizing Zetaproteobacteria in all depths and its population was determined to be up to 4%. Network analysis of microbial communities revealed that appearance of the Zetaproteobacteria coincided with some anaerobic sulfur reducing bacteria, indicating that the Zetaproteobacteira lived in ecological niche of oxic-anoxic interface in the mounds. Seismic data indicated that those mounds grow ˜1cm/yr which is much faster than the abiotic deposition occurring at the surrounding diffuse hydrothermal venting seafloor. Overall, our results indicated that Zetaproteobacteria may accelerate deposition of Fe species in hydrothermal fluid and formation of iron oxyhydroxide-rich mounds in the Nagahama-bay, Satsuma-Iwojima.

キーワード: 熱水マウンド, 鉄酸化細菌, 微生物生態
Keywords: Hydrothermal mound, Fe-oxidizing bacteria, microbial ecology

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-P01 会場:コンベンションホール 時間:5 月 26 日 18:15-19:30

沖縄トラフ熱水プルーム中の微生物群集
Unique microbiome in the hydrothermal plumes in Okinawa Trough sediment hosted back arc hydrothermal systems

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海底ー海洋境界の物質循環において重要な海底熱水が噴出後に海水と混合してできる熱水プルーム中では、熱水中に含まれるイオウ、メタン、水素などの還元物質の酸化によりエネルギーを得る微生物が増殖する。特にイオウをエネルギー源にする SUP05 はさまざまな熱水プルームで報告され、伊豆小笠原弧の熱水プルームでは増加する微生物細胞の８割以上がこの微生物で占められている。一方、沖縄トラフの熱水系では、SUP05 が占める割合は低く、メタンをエネルギー源にする*Methylococcus*が出現することが明らかになっているが、未だに増殖する微生物の最大で約半分はわかっていない。そこで、本研究では今後の海底熱水鉱床開発に向けた熱水域の深海生態系のベースライン調査も含め、これまでに取得した沖縄トラフ熱水系における７サイトの熱水プルーム試料について NGS による高解像度微生物群集解析を行った。プルーム中では、*Thaumarchaeota*種の入れ替わり、*Thermogemmatispora*の増加傾向が認められ、一部のプルームでは、*Surfurimonas*の増加も認められた。熱水由来の無機化学成分利用可能性のあるこれらの微生物に加え、*Caulobacter*、*Sphingomonas*、*Marinobacter*などの従属栄養微生物の増殖も認められた。

キーワード: 熱水プルーム, 次世代シーケンサー, 微生物群集構造解析, 沖縄トラフ
Keywords: hydrothermal plume, MiSeq, microbial community structure, Okinawa Trough

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-P02 会場:コンベンションホール 時間:5 月 26 日 18:15-19:30

微生物による OCS 分解時の同位体分別係数の決定
Sulfur isotopic fractionation in carbonyl sulfide during microbial degradation

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KAMEZAKI, Kazuki[1*] ; HATTORI, Shohei[1] ; OGAWA, Takahiro[2] ; ISHINO, Sakiko[1] ; TOYODA, Sakae[1] ; KATO, Hiromi[3] ; KATAYAMA, Yoko[2] ; YOSHIDA, Naohiro[1]

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Carbonyl sulfide (OCS) is the most abundant gas containing sulfur in the atmosphere, with an average mixing ratio of 500 p.p.t.v. in the troposphere. OCS is suggested as a sulfur source of the stratospheric sulfate aerosols (SSA) which plays an important role for Earth of radiation budget and for ozone depletion. Therefore, OCS should be validated for prediction of climate change, but the global OCS budget is imbalance. It is known that some microorganisms in soil can degrade OCS, but the mechanism and the contribution to the OCS in the air are still uncertain. Isotopic composition and isotopic enrichment factor are used to trace the sources and transformations of atmospheric trace gases. Recently, we developed new method measuring sulfur isotopic composition of OCS using fragmentation ions S^+ , and this method can be used to investigate its sources and sinks in the troposphere. In order to determine sulfur isotopic enrichment factor of OCS during degradation via microorganisms, we performed laboratory incubation experiments using OCS-degradingmicroorganism.

Bacterium strains, which have OCS degradation activity, were cultured on the slant in a glass tube. After forming the colonies, headspace were replaced with N_2/O_2 (80:20 mixture) and 0.03% of CO_2, and then OCS were added to the batch. The concentrations of OCS were measured using gas chromatograph equipped with a flame photometric detector, and headspace gases were collected in the helium purged vials for isotope analysis at the same time of concentration measurements. For isotope analysis, we injected the OCS samples to the gas chromatography-isotope ratios mass spectrometry system using ^{32}S, ^{33}S, and ^{34}S fragment ions. Isotopic enrichment factor is determined by correspond to the Rayleigh isotope fractionation model.

In this experiment, The isotopic compositions (^{33}S and ^{34}S) of OCS were increased during degradation of OCS, indicated that reaction for $OC^{32}S$ was faster than that for $OC^{33}S$ and $OC^{34}S$. On the basis of the concentration of OCS and its isotopic compositions, Rayleigh isotope fractionation model were applied to determine isotopic enrichment factors ($^{33,34}\varepsilon = (^{33,34}S\text{-}_{33,34}S_{initial})/\ln f$). It is worth noting that$^{33}\varepsilon$ and$^{34}\varepsilon$ values determined by the experiments which showed no significant deviations from mass-dependent relationship, indicating that OCS degradation via microorganisms is not mass-independent fractionation (MIF) process. This result suggests this reaction is not contributed to the MIF signatures observed in sulfur for sulfate aerosol samples and/or Archaean rock records.

At the presentation, the comparison of $^{33}\varepsilon$ and $^{34}\varepsilon$ values using some strains and the atmospheric implications for the OCS degradation in the present atmosphere are discussed.

キーワード: 硫化カルボニル, 同位体分別係数, 微生物
Keywords: Carbonyl sulfide, Isotopic fractionation factor, Microorganism

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-P03 会場:コンベンションホール 時間:5 月 26 日 18:15-19:30

Aspergillus oryzae の窒素安定同位体比における窒素可給性の影響
Nitrogen availability influences natural abundance 15N of Aspergillus oryzae

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Nitrogen availability controls nitrogen mineralization and nitrification which are important reaction for nitrogen cycle in the soil (Schimel and Bennet. 2004). To evaluate nitrogen availability, soil C/N ratio and net nitrogen mineralization are usually used. However, difficulty in extracting the nitrogen source pools for soil microbes and getting the field circumstance information by using laboratory culture experiment create the difficulty in evaluating the nitrogen availability accurately. Then, the natural abundance of 15N (d15N) has been used for evaluating the nitrogen availability as a tool of getting the field circumstance information. Dijkstra et al. (2008) showed negative correlation between D15N which means the difference between d15N of SMB (Soil Microbial Biomass) and d15N of microbial substrate (K2SO4 extractable nitrogen from soil) and microbial substrate C/N, and this result suggested D15N could be a good indicator for nitrogen availability. They explained this phenomenon that mineralization is the dominant process for soil microbes at the high nitrogen availability sites, and SMB becomes enriched in 15N because microbes release NH4+ which is depleted in 15N. However, previous study about the relationship between d15N-biomass and d15N-NH4+ in C/N controlled pure culture is conducted only by Collins et al. (2008) who used E. coli, and they could not detect d15N-NH4+ in a low concentration. Thus, the relationship between nitrogen availability and d15N-biomass is unclear. The purpose of our study is to reevaluate if biomass becomes enriched in 15N when microbes release NH4+ which is depleted in 15N. In this study, we cultured Fungi (Aspergillus oryzae) who has large biomass in the forest soil in C/N controlled pure culture (C/N5, 10, 30, 50, 100) for 4 days. We used glycine and glucose as a nitrogen and carbon source. And we measured mainly changes in d15N-biomass, NH4+ concentration and d15N-NH4+. In C/N5 and 10 where NH4+ concentration increased over time, we found that biomass was strongly enriched in 15N and NH4+ is depleted in 15N. Conversely, in C/N 30, 50 and 100 where microbes hardly released NH4+, we found that d15N-biomass got the almost same value of initial d15N-glycine. In the presentation, we will discuss more detail about the carbon and nitrogen mass balance during our experiment.

Japan Geoscience Union Meeting 2015
(May 24th - 28th at Makuhari, Chiba, Japan)

BGM22-P04 会場:コンベンションホール 時間:5 月 26 日 18:15-19:30

埋没腐植層における土壌微生物の無機化ポテンシャルの定量的解析
Biodegradation activity of organic matter in the buried humic horizons in volcanic ash soils

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Over ten thousand years, Hokkaido soils have been formed through deposition of volcanic ash. The surface soil layer in the past has been buried in deeper part of soil profile. The buried humic horizons contain the high amounts of organic matter (OM) and they serve as a large C reservoir. The microbial degradation activity can generally decrease with soil depth, and OM in the deeper soil horizons is stable due to limitations of organic matter input. In contrast, microbial activity in the buried humic horizons may not be low, because the high amounts of OM can fuel soil microbial activity. We aim to examine the microbial mineralization kinetics in the buried humic horizon using ^{14}C-tracer incubation.

Soil samples (volcanic ash soil) were collected from the soil profiles in three forest sites and one pasture site in Hokkaido, Japan. These “fresh” field moist, un-sieved soils were used for mineralization kinetic studies. A ^{14}C-glucose solution (100 μL; 10 - 300000 μM) was added to 1 g of field-moist soil and incubated for 24 h at 20 ℃. The $^{14}CO_2$ production was trapped in the scintillation vial containing NaOH and determined by liquid scintillation counting. The experiments were performed in triplicate. The data of mineralization kinetics were fitted to a single Michaelis-Menten equation: $V = V_{MAX}C/(K_M+C)$, where V is the mineralization rate (nmol g^{-1} h^{-1}), C is the substrate concentration (μM) in soil solution, V_{MAX} is the maximum mineralization rate (nmol g^{-1} h^{-1}), and K_M is the Michaelis constant (μM) representing the concentration at which 1/2 V_{MAX} is achieved. We also measured soil microbial biomass, fungal/bacterial (F/B) ratio, and fine root biomass.

Fine root biomass in the soil profiles decreased with depth, suggestion the low rates of organic substrate supply in the buried humic horizon. Their Michaelis-Menten kinetic parameters (V_{MAX} and K_M) varied widely from 303 to 18598 nmol g^{-1} h^{-1} and 198 to 1294 μM. The parameters decreased with soil depth. The kinetic parameters of the surface soil horizon exhibited high mineralization capacity, while parameters of the buried humic horizons were similar to those of the other soil types (11 to 2406 nmol g^{-1} h^{-1} for V_{MAX} and 198 to 30786 μM for K_M). The both of V_{MAX} and K_M parameters were correlated positively with microbial biomass-C and -N, respectively. This indicates microbial biomass is a primary factor regulating the potential degradation activity in the buried humic horizons. Microbial biomass decreased with soil depth, consistent with the low input of fresh organic substrates in the deeper soil horizons. The higher F/B ratios were observed in the buried humic horizon, compared to the surface horizon. Since F/B ratios has influence on the mineralization kinetic parameters due to differences of substrate use efficiency and growth speed between fungi and bacteria, the high potential degradation activity relative to the other soil types may be due to high F/B ratios in the buried humic horizons. The high potential degradation activities of soil microorganisms in the buried humic horizons suggest that OM decomposition can be accelerated by addition of easily-biodegradable OM which stimulates soil microbial activity.
Keywords: the buried humic horizon, volcanic ash soil, 14C-glucose, microbial mineralization kinetics, soil organic matter