

2Hp12 Microbial population dynamics in an acidophilic nitrifying process in the presence of streptomycin

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[Purposes] Generally, nitrification takes place under neutrophilic conditions. Nitrifying bacteria so far isolated are sensitive to pH and are unable to grow at pH 4 and below. However, we have succeeded to construct an acidophilic nitrifying sequencing batch reactor that keeps activity at pH ≤ 4. Based on the bacterial community analysis of the acidophilic reactor, we found that members of the candidate phylum TM7 became predominant at the end of operation. TM7 bacteria are known to be resistant to streptomycin. Therefore, the main purpose of this study is to confirm acidophilic nitrifying activity of the reactor in the presence of streptomycin.

[Methods & Results] Acidophilic sequencing batch reactors were operated with mineral medium at pH 4.0. Streptomycin at a concentration of 10, 30 and 50 mg/l was added to the reactors. The concentration of ammonia was measured by a colorimetric method. Nitrate and nitrite were measured by ion chromatography. Ammonia oxidation was observed in all test reactors, although the activity decreased with increasing concentrations of streptomycin. These results suggested that streptomycin-resistant bacteria, i.e., TM7 bacteria, or ammonia-oxidizing archaea are involved in acidophilic nitrification. 16S rRNA gene-targeted PCR-DGGE and FISH analyses showed that TM7 bacteria predominated in the reactors.

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Key words acidophilic ammonia oxidation, streptomycin, TM7 bacteria

2Hp14 黄砂バイオエアロゾルのサンプラー開発と生物分析

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[目的] 黄砂は、主として疫学的手法による健康影響に関する研究が展開され、その原因として生物由来の有機物質あるいは微生物そのものが黄砂とともにあるいは黄砂粒子に付着しているのではないかと、すなわち、「黄砂バイオエアロゾル」の存在が疑われている。しかしながら、その存在証拠を提示した研究はほとんどない。本研究では、上空に浮遊している微生物を採取するためのバイオエアロゾルサンプラーを開発し、実際に係留気球を用いて直接採集した。さらに、採取したサンプルの生物学的分析により微生物飛来の実相調査を行った。

[実験方法及び結果] 黄砂発生源地域である中国敦煌市と黄砂沈着地域である日本石川県金沢市・珠洲市にて、係留気球と独自に開発したバイオエアロゾルサンプラーを用いて黄砂バイオエアロゾルの直接採集を実施した。得られたサンプルの分離培養・同定や直接ゲノム解析などを行い、種々の菌株が上空まで舞い上がっていることが確認され、黄砂バイオエアロゾルの存在を実験的に明らかにした。

Development of sampler and bioanalysis of KOSA bioaerosol

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Key words KOSA (Yellow dust), bioaerosol, sampler, bioanalysis

2Hp13 Characterization of predominant uncultured bacteria affiliated with the candidate phylum TM7 in an acidophilic nitrifying reactor

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[Purposes] There is general agreement that nitrification takes place in neutrophilic conditions and is inhibited at acidic pH. However, we have established an acidophilic nitrification reactor in which ammonia oxidizing activity remains continuously at around pH 4.0. The reactor operated under steady-state conditions contained members of the class *Gammaproteobacteria* and the candidate phylum TM7 as the predominant bacteria. Especially, the TM7-type microbes became predominant gradually with time of operation, suggesting that they play an important role in acidophilic nitrification. Therefore, this study aimed to reveal ecophysiological roles of the TM7-type microbes in the acidophilic nitrification reactor. In this report, we attempted to investigate diversity, visualization, and isolation of the TM7-type microbes in the reactor.

[Methods & Results] 16S rRNA gene-targeted PCR-DGGE and clone library analyses for understanding the microbial diversity in the reactor revealed that at least four species were relevant to the formation of microbial community structure in the reactor. Fluorescence in situ hybridization using TM7-specific probes showed that the all TM7 microbes present in the reactor are rod morphotypes with a relatively small shape. Subsequent cultivation procedures with mineral media were successful to enrich TM7 microbes with others such as microbes affiliated with the classes *Actinobacteria* and *Gammaproteobacteria*.

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Key words TM7, acidophilic nitrification, ammonia oxidation

2Hp15 黄砂バイオエアロゾルの沈着地域における直接採集・同定および生態系影響調査

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[目的] 黄砂バイオエアロゾルとは、黄砂粒子に付着または単体で浮遊し黄砂と挙動を共にする生物粒子で、沈着地域のヒトの健康や生態系に影響を及ぼしている可能性がある。本研究では黄砂バイオエアロゾルを直接採集・同定し、生態系影響調査を試みた。

[実験方法及び結果] 係留気球を用いて石川県珠洲市上空に浮遊する黄砂バイオエアロゾルの直接採集を行った。採集した菌を単離し、18S rRNA 遺伝子解析・BLAST検索した結果、糸状菌の一種の *Bjerkandera* sp. であることがわかった。沈着地域の黄砂バイオエアロゾルとして糸状菌が単離されたことから、生態系影響として糸状菌食性線虫が原因の枯死を想定し、黄砂との関連を統計学的手法を用いて検討した。その結果、黄砂観測日数と枯死被害量の相関係数に高い正の相関かつ 95 %水準の有意性が認められた。

Direct sampling and identification of KOSA bioaerosol over a deposit region for the research of ecosystem effects

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Key words KOSA, 18S rRNA, Ecosystem effect