A glimpse into the biosynthetic potential and resistome of microbial communities inhabiting sulfidic, chemoautotrophic Movile Cave ecosystem

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Abstract

Background: Microbial secondary metabolites (SM), especially those produced by soil microorganisms, have been a valuable source of antibiotics, antitumor agents, pigments, growth-promoting substances, etc., with tremendous market potential. These molecules are encoded by biosynthetic gene clusters (BGCs) within the bacterial genome. Their synthesis confers survival advantages by facilitating chemical defense, interspecies communication, and adaptation to well-defined ecological niches.

Caves, particularly Movile Cave (Romania) - a sulfidic autotrophic-based ecosystem - can be considered extreme environments suited to investigate and discover novel bioactive microbial molecules. Here, low nutrient availability can lead to resource competition and, consequently, antimicrobial production to deter nearby microbial competitors.

Aim: Our study focused on highlighting the biosynthetic gene clusters (BGCs) and biosynthetic potential of the sediment-associated microbiome in Movile Cave.

Methods: Over 100 high-quality metagenome-assembled genomes (MAGs) were retrieved by whole-community shotgun-sequencing of 7 sediment samples collected in different Movile Cave's galleries (Chiciudean et al. 2022). Detected MAGs were then analyzed for the presence of BGCs by antiSMASH (v. 6.1.1) (Blin et al. 2021) whereas antibiotic resistance genes were predicted by ResFinder (v. 4.1) (Florensa et al. 2022). The statistical analysis of BGCs data was performed by Past (v. 4.03).

Results: We detected 637 BGCs across 106 high-quality MAGs that were affiliated to 22 phyla. The diversity of predicted BGCs varies across sediment samples with no apparent correlation to the number of analyzed MAGs per sample. The MAGs recovered from the sulfidic water-sediment interface (sample code PMV4) had the lowest alpha BGCs diversity among all sampled locations and it was clearly distinct in BGCs composition and

abundance (β-diversity) from dry gallery samples (PMV7 and PMV8). The most abundant BGCs predicted in Movile Cave metagenomic dataset encode for terpenes, non-ribosomal peptides (NRPs) and ribosomally synthesised and post-translationally modified peptides (RIPPs). *Acidobacteriota* and *Chloroflexota*- affiliated MAGs were most enriched in BCG containing 20 and 23 BGCs per MAG, respectively, in contrast with the candidate phylum *Ca.* Patescibacteria-related MAGs that showed no SM biosynthetic capabilities. Two antimicrobial resistance (AR) genes (*ole(C)*, *oqxB*) encoding resistance to antibiotics (i.e. oleandomycin, chloramphenicol, ciprofloxacin, trimethoprim) and disinfectants were identified in MAGs affiliated with the class *Actinomycetia* and *Gammaproteobacteria*.

Based on the analyzed data, the biosynthetic potential of Movile Cave is significant compared with other microbiomes (Donia et al. 2014) and has a pronounced degree of novelty whereas the resistome (that is the genetic potential for antibiotic resistance) is reduced. Considering the uncommon futures of Movile Cave environment, future in-depth analysis of the identified BGCs might lead to the discovery of novel bioactive compounds. Additionally, the seclusion of this environment may provide an exciting opportunity for surveying the occurrence and environmental drivers of natural AR traits.

Keywords

Movile Cave; metagenomes; biosynthetic gene clusters (BGCs); antiSMASH; antimicrobial resistance (AR).

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