Microbial Ecology
https://doi.org/10.1007/s00248-023-02202-0

NOTE

Diversity of Microbiomes Across a 13,000-Year-Old Amazon Sediment

Cristiane C. Thompson[1] · Diogo Tschoeke[1,2] · Felipe H. Coutinho[1,3] · Luciana Leomil[1] · Gizele D. Garcia[1,4] · Koko Otsuki[1] · Bruno J. Turcq[5] · Luciane S. Moreira[6] · Patrícia F. M. Turcq[5] · Renato C. Cordeiro[6] · Nils E. Asp[7] · Fabiano L. Thompson[1]

Received: 8 September 2022 / Accepted: 23 February 2023

Abstract

The microbiome is fundamental for understanding bacterial activities in sediments. However, only a limited number of studies have addressed the microbial diversity of Amazonian sediments. Here, we studied the microbiome of sediments from a 13,000-year BP core retrieved in a floodplain lake in Amazonia using metagenomics and biogeochemistry. Our aim was to evaluate the possible environmental influence over a river to a lake transition using a core sample. To this end, we sampled a core in the Airo Lake, a floodplain lake in the Negro River basin. The Negro River is the largest tributary of the Amazon River. The obtained core was divided into three strata: (i) surface, almost complete separation of the Airo Lake from the Negro River when the environment becomes more lentic with greater deposition of organic matter (black-colored sediment); (ii) transitional environment (reddish brown); and (iii) deep, environment with a tendency for greater past influence of the Negro River (brown color). The deepest sample possibly had the greatest influence of the Negro River as it represented the bottom of this river in the past, while the surface sample is the current Airo Lake bottom. In total, six metagenomes were obtained from the three different depth strata (total number of reads: 10.560.701; sequence length: 538 ± 24, mean $\pm$ standard deviation). The older (deeper) sediment strata contained a higher abundance of *Burkholderia*, *Chitinophaga*, *Mucilaginibacter*, and *Geobacter*, which represented ~ 25% of the metagenomic sequences. On the other hand, the more recent sediment strata had mainly *Thermococcus*, *Termophilum*, *Sulfolobus*, *Archaeoglobus*, and *Methanosarcina* (in total 11% of the metagenomic sequences). The sequence data were binned into metagenome-assembled genomes (MAGs). The majority of the obtained MAGs ($n = 16$) corresponded to unknown taxa, suggesting they may belong to new species. The older strata sediment microbiome was enriched with sulfur cycle genes, TCA cycle, YgfZ, and ATP-dependent proteolysis in bacteria. Meanwhile, serine-glyoxylate cycle, stress response genes, bacterial cell division, cell division-ribosomal stress protein cluster, and oxidative stress increased in the younger strata. Metal resistance and antimicrobial resistance genes were found across the entire core, including genes coding for fluoroquinolones, polymyxin, vancomycin, and multidrug resistance transporters. These findings depict the possible microbial diversity during the depositional past events and provided clues of the past microbial metabolism throughout time.

Keywords Microbiome · 13,000-year-old Amazon sediment · Antibiotic resistance genes

✉ Cristiane C. Thompson
thompsoncristiane@gmail.com

✉ Fabiano L. Thompson
fabianothompson1@gmail.com

[1] Institute of Biology, Universidade Federal do Rio de Janeiro (UFRJ), Rio de Janeiro, Brazil

[2] Biomedical Engineer Program, COPPE (UFRJ), Rio de Janeiro, Brazil

[3] Institut de Ciències del Mar (ICM-CSIC), Barcelona, Spain

[4] Instituto de Ciências Médicas, Centro Multidisciplinar UFRJ Macae, Universidade Federal do Rio de Janeiro (UFRJ), RJ, Macae, Brazil

[5] Institute de Recherche pour Le Dévelopment-Sorbonne, Université (UPMC, CNRS, IRD, MNHN) LOCEAN - Centre IRD France Nord, Bondy, France

[6] Programa de Geoquímica, Universidade Federal Fluminense (UFF), Niterói, Rio de Janeiro, Brazil

[7] Instituto de Estudos Costeiros (IECOS), Universidade Federal do Pará (UFPA), Bragança, Brazil

Published online: 05 April 2023

Introduction

Microbial diversity may be influenced by biogeochemical properties of sediment and soils, precipitation seasonality-related changes, sediment nutrient, moisture regimes, and deforestation [4, 5]. The majority of the previous microbiome studies have focused on Amazon soils. In terrestrial ecosystems, decomposers, anaerobic saprophytes, and N_2 fixers are more abundant during the rainy season. An increase in sediment drying may increase methane production [11, 12, 23], with proliferation of *Methanocellales*, *Methanosarcinaceae*, and *Clostridiales*. Deforestation impacts nitrification and denitrification in Amazon soils [25]. Higher litter abundance possibly leads to higher abundance of nitrogen fixers-*Beijerinckiaceae*, suggesting canopy phenology patterns drive microbial abundance and soil nutrient patterns. *Proteobacteria* (*Burkholderiaceae*, *Pseudomonadaceae*), *Acidobacteria*, and *Bacillaceae* are typical groups at the soil surface, and *Bathyarchaeota* and *Thaumarchaeota* appear in deep soils [33]. Antibiotic and metal resistance genes are found in recent Amazon Forest soils [39], as a possible contribution of *Proteobacteria*. However, it is unclear if these resistance genes also occur in deeper sediment layers as a possible effect of microbial competition for sediment habitats.

Deforestation decreased sediment organic matter content and factors linked to sediment acidity and raised sediment pH, base saturation, and exchangeable bases, and increased diversity of *Actinomycetales* [34]. A decrease in the relative abundances of *Chlamydiae*, *Planctomycetes*, and *Verrucomicrobia* was also observed in the deforested sediments. A co-occurrence microbial network analysis disclosed possible pairs: *Planctomycetes* and aluminum content, and *Actinobacteria* and nitrogen sources in Amazon sediments. Modulation of biodiversity is clear after deforestation, suggesting a microbial buffer capacity. Apparently, healthy Amazon Forest sediments contain mycelium-forming actinomycete (*Catenulispora acidiphila*), nitrite-oxidizing rhizobia bacteria (*Nitrobacter hamburgensis*), the photoautotrophic/photoheterotrophic/chemoautotrophic/chemoheterotrophic and nitrogen fixer Alphaproteobacteria (*Rhodopseudomonas palustris*), and chemoautotrophic (*Oligotropha carboxidovorans*) [18]. It is unclear if these taxonomic groups or guilds can be found in old Amazon sediments which would also reflect past healthy conditions. Forest-to-pasture conversion leads to microbial diversity changes [38]. Potential recovery of metagenomic functional attributes may occur in secondary forest after pasture abandonment. Loss of microbial diversity occurs in soybean plantation with a decrease in *Acidobacteria*, *Actinobacteria*, and *Proteobacteria* [21]. However, diazotrophic microbial diversity may recover in secondary forests after agriculture is stopped [32].

Despite all studies on the Amazon Forest developed so far, most of the previous studies focused on the microbial diversity of surface soils with different levels of anthropogenic use and impact. The aim of the present study was to determine the microbial diversity and antibiotic/metal resistance gene contents across a sediment core. The study of sediment cores is a useful tool to unlock the microbial diversity of past events, and the original microbiome of old sediments (e.g., Holocene sediments). The study of these sediment samples may also shed light on the microbial metabolic repertoire throughout time. Here, the taxonomic and functional profiles of such old Amazon sediment microbiome were analyzed by means of metagenomics. Bulk organic matter geochemical analyses were also performed in the sediment core to determine the major features of the different sediment strata.

Materials and Methods

Study Area

The Airo Lake is a floodplain lake in the Negro River basin (0°19′37.225″S, 66°8′33.266″W), Municipality of São Gabriel da Cachoeira, Amazonas State (Fig. S1). The Negro River is the largest tributary of the Amazon River, considered the fifth largest river in the world [19]. In the Negro River basin, Podzol soil type develops as a result of the intense weathering of typical Amazonian Latosols [9]. This natural process leads to the formation of a deep soil horizon rich in organic matter and iron and aluminum oxides characteristic of Podzols. This organic matter is exported to the rivers, hence their typical black color. The Airo Lake has approximately 3 m of depth, maximum width of 300 m, pH around 3.5, and nowadays has almost no connection with the Negro River except during the most extreme floods. Thus, the Airo Lake is located in the equatorial region, in the Amazon Forest biome, with a hot and humid climate. The annual rainfall average at São Gabriel da Cachoeira city is 2853 mm [40].

Sediment Core and Geochemical Analysis

Aluminum tubes, 3 inches in diameter, mounted with "vibra-core" system retrieved an AIRO 12/01 148-cm-long sediment core (AIRO 12/01 0°19′37.225″S 66°8′33.266″W). The AIRO 12/01 sediment core samples from different depths (12–17, 34–37, 44–47, 64–67, 97–101, and 137–141 cm) were analyzed. The texture and color of the sediment (Munsell Sediment Color Chart) were described. The Munsell Sediment Color Chart

based a sediment color and texture core description. The sediment core was sliced into 1-cm sections which were stored in plastic bags. Geochemical characterization of the sediments was performed as described previously [14, 15, 48]. The bulk density of each layer was obtained by removing 8 cm^3 of a wet sediment section and drying it at 60 °C to the constant weight. Seven accelerator mass spectrometry (AMS) radiocarbon dates from bulk organic matter were used to determine chronology at the Laboratoire de Mesure du Carbone 14, (LMC14-France). The 14C AMS ages were transformed into calendar ages using INTCAL98 for ages less than 24,000 ^{14}C year before present (year BP) [45]. A laser particle analyzer (CILAS 1064) was used to measure the grain size distribution (particles between 0.04 and 500 µm) after the organic matter and carbonate destruction with H_2O_2 and HCl, respectively, and it is dispersing in a $Na_4P_2O_7$ solution and sonification. To determine TOC, total organic nitrogen (TN), $\delta^{13}C$, and $\delta^{15}N$, the samples were treated with a 0.5 mol L^{-1} HCl solution. The treated sediment samples were analyzed for their elemental (carbon and nitrogen) and isotopic (^{13}C and ^{15}N) composition using the elemental analyzer PDZ Europa ANCA-GSL interfaced to a mass spectrometer PDZ Europa 20–20 isotope ratio (SERCON Ltd. Cheshire, UK) (UC Davies Laboratory). Chlorophyll derivatives were extracted using 90% acetone, and their absorbances were determined using a scanning spectrophotometer at the 350–800 nm interval. Background correction was performed through baseline subtraction from 500 to 800 nm. Pigment concentrations are reported as Sedimentary Pigments Degradation Units (SPDU) [46], an arbitrary unit, where 1 SPDU represents an absorbance of 1.0 in a 10-cm cell using 100 mL of acetone as a solvent for 1.0 g of OM [13].

DNA Extraction

DNA was extracted from 2 g of sediment from each sample using Power Soil DNA Isolation kit (Mo Bio, Carlsbad, CA, USA). DNA integrity was quantified in a NanoDrop ND 1000 instrument (Thermo Scientific, DE, USA) and with a fluorimetric QuBit dsDNA High Sensitivity Assay system (Life Technologies, Waltham, MA USA). DNA was stored at – 20 °C until library construction. GenomiPhi amplification prior Nextera XT sample preparation was required. GenomiPhi reactions (containing 10–100 ng of template DNA) were performed to produce ~ 4 µg of amplified DNA, using the Illustra GenomiPhi DNA v2 Kit (GE Healthcare). Afterwards, DNA was purified by Power Clean – DNA Clean Up kit (Mo Bio), and again, quantified, prior library construction (16, 49).

Metagenomic Sequencing and Data Analyses

Metagenomic libraries were prepared using the Nextera XT DNA Sample Preparation Kit, according to manufacturer's instructions. Library DNA was purified using AMPure XP beads and quantified using the fluorimetric Qubit dsDNA High Sensitivity Assay system (Life Technologies). Quantification of libraries was performed with the 7500 Real-Time PCR (Applied Biosystems, Foster City, CA, USA) and the KAPA Library Quantification Kit (Kapa Biosystems, Wilmington, MA, USA). Library size distribution was accessed using the 2100 Bioanalyzer (Agilent). All ten metagenomes were sequenced by Illumina MiSeq—paired-end sequencing (2 × 300 base pairs).

Low base quality (Phred average quality score < 30) and duplicated sequences were removed using PRINSEQ [42]. The resulting forward and reverse paired-end sequences were merged with SHERA [41]. Taxonomic and functional annotation of merged reads were performed using MG-RAST [31].

We used all metagenomes to build metagenome-assembled genomes (MAGs) [20, 48] using metaSPAdes v. 3.10 [35]. The genome assembly starts with metagenomic binning, in which assembly DNA reads into contigs. These contigs are then binned into putative partial or complete MAGs [3]. In the next step, we evaluated the MAGs' completeness and contamination using checkM [37] and we used GTDB-Tk [8] to perform a taxonomic classification.

Results and Discussion

Airo Sedimentological and Geochemical Features

Core AIRO 12/01 is composed of sand from the base to 54 cm. This unit is very compacted, with water contents of 20 to 40%, making it difficult for microorganisms to migrate. From 54 to 29 cm, more silty layers appear indicating a decrease in the hydrodynamics of the river at the sampling site. From 29 cm to the top of the core, the sediment is clayey-silt indicating the end of the direct influence of the river, the sediment arriving only in suspension during the highest floods. Between 230 cm and the top, bulk density decreased and water content (%) increased. Values of TOC, C:N, $\delta^{13}C$, and $\delta^{15}N$ show clear differences along the sediment profile (Table S1). TOC ranged between 0.6% (depth) and 33.17% (surface), while the highest C:N ratio occurred in the deep sediment strata (57.72) (Table S1; Fig. 1). These extremely high C:N ratios are characteristic of the organic matter from the Podzols of the Negro River basin that give the river its black color [2]. $\delta^{15}N$ showed significant changes between surface (1.90‰) and depth (0.07‰), indicating a relatively higher autochthonous

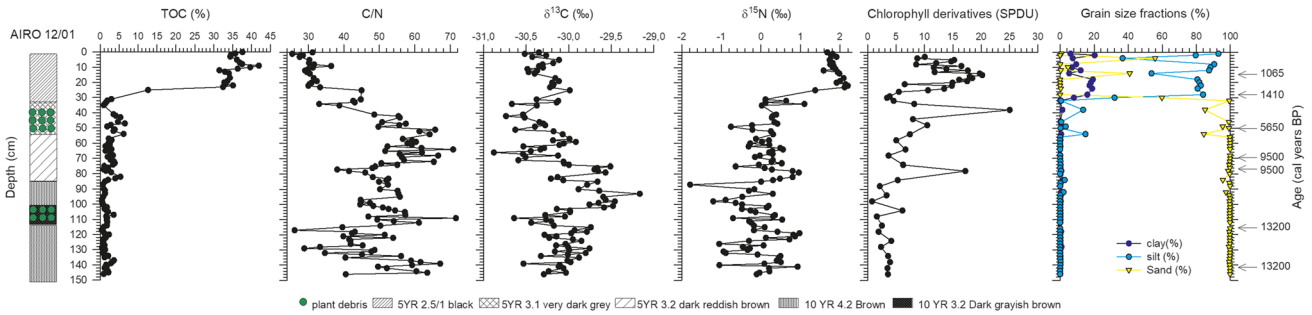

Fig. 1 Geochemical features of the AIRO 12/01 core. Total organic carbon (TOC), carbon:nitrogen molar ratios (C:N), nitrogen stable isotopes ($\delta^{13}C$ and $\delta^{15}N$), chlorophyll derivatives, and grain size distribution

productivity, since less organic matter came from the river, and perhaps a more active recycling of nitrogen at the time of deposition of the upper 24 cm of sediment [22, 47], and total nitrogen had an opposite trend (surface: 1.10%,depth: 0.03% (Table S1; Fig. 1). The age of the stratigraphic profile varied from 13,245 to the present to calibrated years before present (cal year BP; years BP thereafter). This corresponds to the Holocene, including the Pleistocene/Holocene transition. The core was divided into three phases according to the sedimentological and geochemical characteristics: (i) surface (black-colored sediment) (29–0 cm; 1.800–0 cal year BP). It is the period of Lake Airo formation with an almost complete separation from the Negro River except during extreme flood events. The environment becomes more lentic with greater deposition of organic matter from the lacustrine vegetation; (ii) transitional environment (reddish brown) (84–29 cm; 10.800–1.800 cal year BP). During this phase, we observe a progressive decrease of the hydrodynamics of the paleochannel of the Negro River whose abandonment will form the Airo Lake; and (iii) deep (brown color) (148–84 cm; 13.250–10.800 cal year BP) (Table S1; Fig. 1). During this period, the Negro River had a much stronger hydrodynamic force than today and transported a large quantity of sand from the erosion of the soils of its drainage basin. It is the period of construction of the Negro River floodplain by a braided river made up of several channels. One of these channels, very active, passed at the location of the current Airo Lake where it deposited the sandy sediment and organic matter from the Podzols.

Taxonomic Features of Metagenomes Across Sediment Strata

Bacteria were the most abundant domain (52.5 to 79.7% of the total metagenomic sequences) in all depths, followed by Archaea (10.1 to 49.2% of the total metagenomic sequences) (Table S2). Bacterial sequences were more abundant at deep samples ($p < 0008$); meanwhile, Archaea were more abundant at surface ($p < 0.0002$). Virus (mean ± standard deviation: 0.21 ± 0.2) and Eukarya (mean ± standard deviation: 3.9 ± 5.6) represented a smaller fraction of the metagenomes. Eukarya was significantly more abundant at deeper layers (mean ± standard deviation: 0.9% and 9.8%; $p < 0.046$). *Ascomycota* was found in the two sampling points (97 cm: 5.9%; 137 cm: 1.5%). *Basidiomycota* (0.62%), *Phaeophyceae* (0.9%), and *Streptophyta* (0.75%) occurred at 97 cm depth, but not at 137 cm depth (Table S2).

Archaeal members of *Euryarchaeota* (6.8 to 33.0% of abundance) and *Crenarchaeota* were dominant (2 to 3.7% of abundance) at all depths followed by *Thaumarchaeota* (0 to 3% of abundance) (Table S2). Abundance of Archaea in sediment cores related to reduced availability of organic matter was observed [4, 17, 20]. We obtained 16 MAGs (13 archaeal and 3 bacterial). The *Proteobacteria Burkholderiaceae*, *Chitinophagaceae*, and *Sphingobacteriales* occurred among the 16 identified MAGs; however, the majority of the MAGs did not have a taxonomic identification as they may belong to new groups (Table S3). Further work is required to determine the exact taxonomic position of these unidentified MAGs.

Two major clusters were found based on the taxonomic profiles obtained in MG-RAST (Table S2). The surface cluster comprised mainly of *Thermococcus* (2.9%), *Termophilum* (2.4%), *Sulfolobus* (2%), *Archaeoglobus* (1.8%), and *Methanosarcina* (1.7%). *Thermococcus*species are strictly anaerobes, thermophilic, and found in a variety of depths [1, 6]. *Thermococcus* are described as heterotrophic, chemotrophic, and organotrophic sulfanogens, using elemental sulfur and carbon sources. Some *Thermococcus* species produce CO_2, H_2, and H_2S as products of metabolism and respiration. The releases of these molecules are used by other autotrophic species [43]. *Sulfolobus* are sulfur-oxidizing bacteria, and *Archaeoglobus* are sulfate-reducing Archaea, coupling the reduction of sulfate to sulfide with the oxidation of many different organic carbon sources [26, 49]. *Methanosarcina* species are known as anaerobic methanogens that produce methane using all three metabolic pathways for methanogenesis.

Burkholderia (12%), *Chitinophaga* (8.3%), *Mucilaginibacter* (2.4%), and *Geobacter* (2.4%) were more abundant in the deep cluster. *Burkholderia*are common soil inhabitants and some species can oxidize sulfur [7]. *Chitinophaga*can be found in soil and have the capability to hydrolyze chitin and some species contained deferoxamine producing genes [24].*Mucilaginibacter* are heterotrophic bacteria capable of degrading pectin, xylan, laminarin, and some other polysaccharides [36]. *Geobacter*species have been found in anaerobic conditions in soils and aquatic sediment [30] and can oxidize organic compounds and metals, including iron, radioactive metals, and petroleum compounds into carbon dioxide [10].

Transition from River to Lake Influenced Sediment Microbiome

An evident change of gene content across the sediment strata was observed (Fig. 2a, b). Sulfur metabolism genes (0.92% at deep strata; 0.27% surface) were more abundant in the deep strata than in the surface (Fig. 2a). Today, the Negro River contains very little sulfate

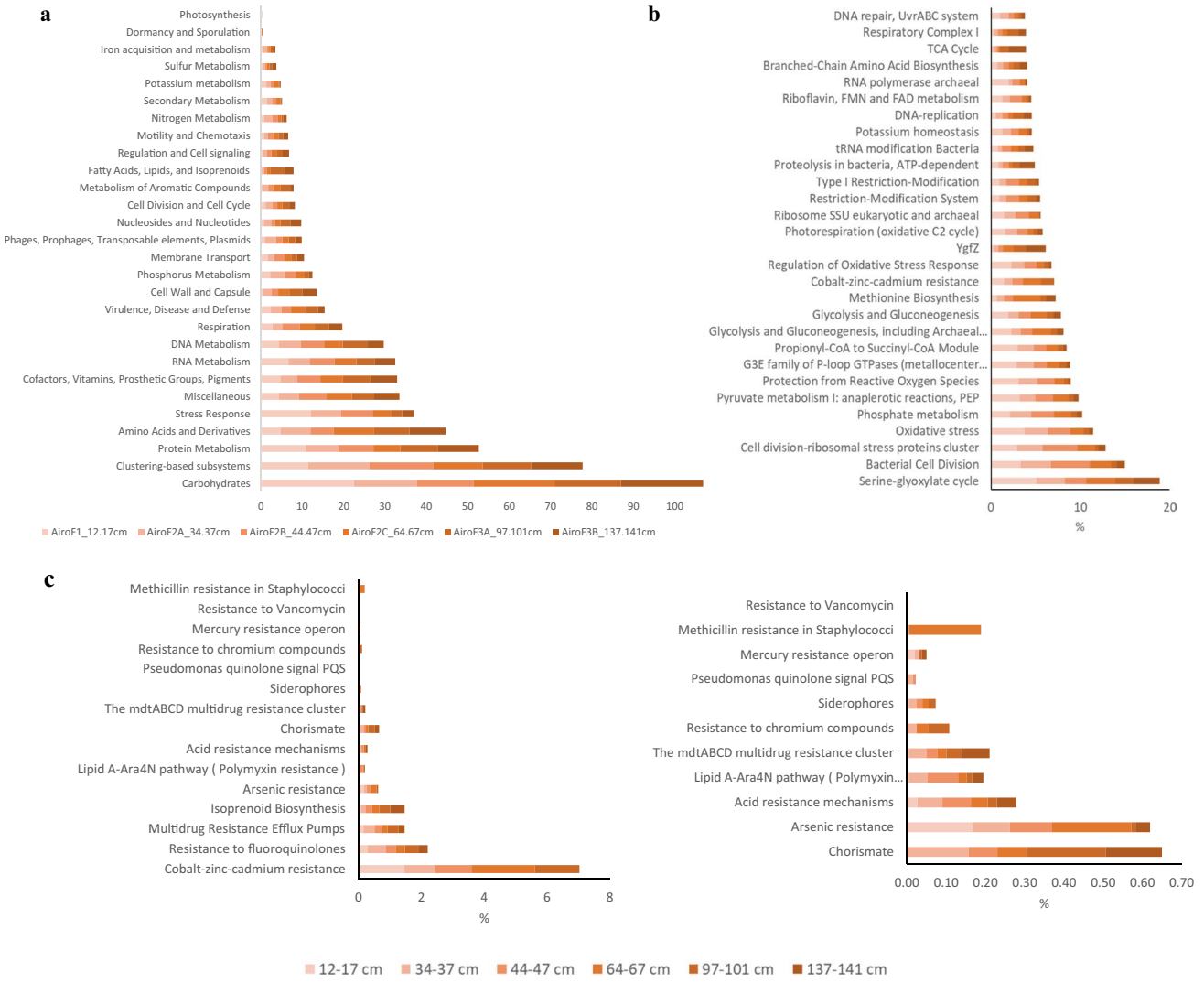

Fig. 2 Functional identification of metagenomes using MG-RAST. **a** The most abundant subsystems were carbohydrates and clustering-based subsystems that amounted to 30.75% relative abundance across metagenomes, followed by protein metabolism (8.77% relative abundance) and amino acid and derivatives (7.43% relative abundance). **b** Serine-glyoxylate cycle ranging from 2.01% (97–101 cm) to 5.10% (12–17 cm) followed by bacterial cell division ranging from 4.31% (44–47 cm) to 0.60% (97–101 cm), cell division-ribosomal stress protein cluster (from 3.87% at 44–47 cm to 0.43% at 97–101 cm), and oxidative stress from 3.74% (12–17 cm) to 0.43% (137–141 cm) which were the most abundant level 3 subsystems and decreased with depth. TCA cycle, YgfZ, proteolysis in bacteria–ATP-dependent were increased in high depths. **c** Antimicrobial and metal resistance genes using MG-RAST. Siderophore = siderophore assembly kit, pyoverdine, yersiniabactin, bacillibactin, anthrachelin, staphylobactin, achromobactin. chorismate: intermediate for synthesis of tryptophan, PAPA antibiotics

(0.002 mmol l^{-1}) compared to the Amazon downstream (Obidos: 0.05 mmol l^{-1}) [44]. The presence of bacteria using sulfur compounds in the basal unit suggests that the ionic composition of the Negro River waters was very different at the beginning of the Holocene, which corresponds to a phase of intense erosion of the Negro River basin soil [29]. TCA cycle, YgfZ, and ATP-dependent proteolysis in bacteria were also increased in deep strata. On the other hand, serine-glyoxylate cycle (2.01% at deep strata; 5.10% at surface), stress response genes (12% in the surface; 2.8% in deep), bacterial cell division (4.31% at intermediate; 0.60% at deep strata), cell division-ribosomal stress protein cluster (3.87% at intermediate; 0.43% at deep strata), and oxidative stress (3.74% at surface; 0.43% at deep strata) decreased with depth (Fig. 2b). The greater environmental stress in the upper part of the core may be related to the fact that the lake is then isolated from the Negro River. It is a small aquatic system that responds easily to climatic and hydrological variability. These results are consistent with an environment with low microbial growth and abundance at deep strata (9.3Kybp). The variation of gene content across the core profile suggests major environmental changes throughout time which influenced the biochemical properties of the sediments. The results indicate a greater influence of the Negro River in deep strata and minor influence of this river in surface strata. The presence of a diverse arsenal of genes related to resistance to metals and antibiotics was also clear from the results of this study (Fig. 2c).

Genes responsible for antibiotic resistance were found in small amounts (up to 0.06%) across the entire sediment core (polymyxin resistance protein ArnA_DH, UDP-glucuronic acid decarboxylase EC 4.1.1.-, polymyxin resistance protein PmrJ, predicted deacetylase, polymyxin resistance protein ArnT, undecaprenyl phosphate-alpha-L-Ara4N transferase, polymyxin resistance protein ArnA_FT, UDP-4-amino-4-deoxy-L-arabinose formylase EC 2.1.2.-, polymyxin resistance protein PmrL, sucrose-6 phosphate hydrolase, polymyxin resistance protein ArnC, glycosyl transferase EC 2.4.-.-, ABC-type transport system involved in resistance to organic solvents, permease component USSDB6A, vancomycin resistance protein VanH, multidrug resistance transporter, and the Bcr/CflA family). Genes related to metal resistance were also found in small amounts (up to 0.06%; mercuric resistance operon regulatory protein, arsenic resistance protein ArsH, mercuric resistance operon coregulator, cobalt-zinc-cadmium resistance protein CzcA) (Fig. 2c). These findings suggest antibiotic and metal resistance genes may play roles in bacterial niche occupancy in sediments as antibiotic- and metal-resistant microbes may have a competitive advantage to colonize certain types of sediments throughout time. Metal and antibiotic resistance genes occur in proximal genomic regions and have co-evolved in certain bacteria [16, 27, 28]. Certain microbial species are relevant bioindicators of contamination as they are both metal and antibiotic resistant [16].

Conclusions

The gene content variation across the core profile suggests major environmental changes throughout time which influenced the biochemical properties of the sediments. These changes imprinted clear modifications in the sediment microbiomes, separating the sediment in different groups. The results indicate a greater influence of the Negro River in deep strata and minor influence of this river in surface strata in agreement with sedimentological and geochemical proxies. The presence of a diverse pool of genes related to resistance to metals and antibiotics was also clear from the results of this study, which contribute to the diversification of microbiomes throughout time. The results of this study depict the possible microbial diversity during the depositional past events, and provided clues of the past microbial metabolism throughout time.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s00248-023-02202-0.

Author Contribution C.C.T. and F.L.T. conceived and designed the study. Material preparation, data collection, and analysis were performed by D. T., F. H. C., L. L., G. D. G, K. O., B. J. T., L. S. M., P. F. M.T., R. C. C., and N.E.A. The first draft of the manuscript was written by C.C.T. and F.L.T. and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Funding The study was financially supported by CNPQ, FAPERJ, and CAPES.

Declarations

Conflict of Interest The authors declare no competing interests.

References

1. Amenábar MJ, Flores PA, Pugin B, Boehmwald FA, Blamey JM (2013) Archaeal diversity from hydrothermal systems of Deception Island. Antarctica Polar Biol 36(3):373–380. https://doi.org/10.1007/s00300-012-1267-3.S2CID11705986
2. Bardy M, Derenne S, Allard T, Benedetti MF, Fritsch E (2011) Podzolisation and exportation of organic matter in black waters of the Rio Negro (upper Amazon basin, Brazil). Biogeochem 106(1):71–88. https://doi.org/10.1007/s10533-010-9564-9
3. Bowers RM, Kyrpides NC, Stepanauskas R, Harmon-Smith M, Doud D, Reddy TBK, Schulz F, Jarett J, Rivers AR, Eloe-Fadrosh EA et al (2017) Minimum information about a single amplified genome (MISAG) and a metagenome-assembled genome (MIMAG) of bacteria and archaea. Nat Biotechnol 35:725–731
4. Buessecker S, Zamora Z, Sarno AF, Finn DR, Hoyt AM, van Haren J, Urquiza Muñoz JD, Cadillo-Quiroz H (2021) Microbial

communities and interactions of nitrogen oxides with methanogenesis in diverse peatlands of the Amazon basin. Front Microbiol 12:659079. https://doi.org/10.3389/fmicb.2021.659079
5. Buscardo E, Geml J, Schmidt SK, Freitas H, da Cunha HB, Nagy L (2018) Spatio-temporal dynamics of soil bacterial communities as a function of Amazon forest phenology. Sci Rep 8(1):4382. https://doi.org/10.1038/s41598-018-22380-z
6. Canganella F, Jones WJ, Gambacorta A, Antranikian G (1998) Thermococcus guaymasensis sp. nov. and Thermococcus aggregans sp. nov., two novel thermophilic archaea isolated from the Guaymas Basin hydrothermal vent site. Int J Syst Bacteriol 48 Pt 4(4): 1181–5
7. Chaudhary S, Dhanker R, Singh K, Brar B, Goyal S (2022) Characterization of sulfur-oxidizing bacteria isolated from mustard (Brassica juncea L) rhizosphere having the capability of improving sulfur and nitrogen uptake. J Appl Microbiol 133(5):2814–2825. https://doi.org/10.1111/jam.15742
8. Chaumeil PA, Mussig AJ, Hugenholtz P, Parks DH (2019) GTDB-Tk: a toolkit to classify genomes with the genome taxonomy database. Bioinformatics 36:1925–1927
9. Chauvel A, Lucas Y, Boulet R (1987) On the genesis of the soil mantle of the region of Manaus, Central Amazonia, Brazil. Experientia 43:234–240
10. Childers SE, Ciufo S, ovely DR, (2002) Geobacter metallireducens accesses insoluble Fe (III) oxide by chemotaxis. Nature 416(6882):767–769. https://doi.org/10.1038/416767a.PMID11961561.S2CID2967856
11. Conrad R (2020) Methane production in soil environments-anaerobic biogeochemistry and microbial life between flooding and desiccation. Microorganisms 8(6):881. https://doi.org/10.3390/microorganisms8060881
12. Conrad R, Ji Y, Noll M, Klose M, Claus P, Enrich-Prast A (2014) Response of the methanogenic microbial communities in Amazonian oxbow lake sediments to desiccation stress. Environ Microbiol 16(6):1682–94. https://doi.org/10.1111/1462-2920.12267.
13. Cordeiro RC, Turcq B, Suguio K, Silva AO, Sifeddine A, Ribeiro CV (2008) Holocene fires in East Amazonia (Carajás), new evidences, chronology and relation with paleoclimate. Global and Planetary Change, Amsterdam 61:49–62. https://doi.org/10.1016/j.gloplacha.2007.08.005
14. Cordeiro RC, Machado W, Santelli RE, Figueiredo AG Jr, Seoane JCS, Oliveira EP et al (2015) Geochemical fractionation of metals and semimetals in surface sediments from tropical impacted estuary (Guanabara Bay, Brazil). Environ Earth Sci 74:1363–1378. https://doi.org/10.1007/s12665-015-4127-y
15. Cordeiro RC, Santelli RE, Machado W, Moreira LS, Freire AS, Braz BF, Rizzini Ansari N, Bidone ED, Meniconi MFG (2017) Biogeochemical factors controlling arsenic distribution in a densely populated tropical estuary (Guanabara Bay, RJ, Brazil). Environ Earth Sci 76:561. https://doi.org/10.1007/s12665-017-6888-y
16. Custodio M, Espinoza C, Peñaloza R, Peralta-Ortiz T, Sánchez-Suárez H, Ordinola-Zapata A, Vieyra-Peña E (2022) Microbial diversity in intensively farmed lake sediment contaminated by heavy metals and identification of microbial taxa bioindicators of environmental quality. Sci Rep 12(1):80. https://doi.org/10.1038/s41598-021-03949-7
17. Danovaro R, Snelgrove PV, Tyler P (2014) Challenging the paradigms of deep-sea ecology. Trends Ecol Evol 29(8):465–475. https://doi.org/10.1016/j.tree.2014.06.002
18. Fonseca JP, Hoffmann L, Cabral BCA, Dias VHG, Miranda MR, de Azevedo Martins AC, Boschiero C, Bastos WR, Silva R (2018) Contrasting the microbiomes from forest rhizosphere and deeper bulk soil from an Amazon rainforest reserve. Gene 5(642):389–397. https://doi.org/10.1016/j.gene.2017.11.039
19. Franzinelli E, Igreja H (2002) Modern sedimentation in the Lower Negro River, Amazonas State, Brazil. Geomorphology 44:259–271
20. Freitas L, Appolinario L, Calegario G, Campeão M, Tschoeke D, Garcia G, Venancio IM, Cosenza CAN, Leomil L, Bernardes M, Albuquerque AL, Thompson C, Thompson F (2020) Glacial-interglacial transitions in microbiomes recorded in deep-sea sediments from the western equatorial Atlantic. Sci Total Environ 746:140904. https://doi.org/10.1016/j.scitotenv.2020.140904
21. Goss-Souza D, Mendes LW, Rodrigues JLM, Tsai SM (2020) Ecological processes shaping bulk soil and rhizosphere microbiome assembly in a long-term amazon forest-to- agriculture conversion. Microb Ecol 79(1):110–122. https://doi.org/10.1007/s00248-019-01401-y
22. Granger J, Wankel SD (2016) Isotopic overprinting of nitrification on denitrification as a ubiquitous and unifying feature of environmental nitrogen cycling. Proc Natl Acad Sci USA 113(42):E6391–E6400. https://doi.org/10.1073/pnas.1601383113
23. Hernández M, Klose M, Claus P, Bastviken D, Marotta H, Figueiredo V, Enrich-Prast A, Conrad R (2019) Structure, function and resilience to desiccation of methanogenic microbial communities in temporarily inundated soils of the Amazon rainforest (Cunia Reserve, Rondonia). Environ Microbiol 21(5):1702–1717. https://doi.org/10.1111/1462-2920.14535
24. Jin CZ, Jin L, Kang DH, Liu MJ, Lee JM, Park DJ, Kim CJ (2022) Description of desferrioxamine-producing bacterium Chitinophaga agrisoli sp. nov., isolated from soil. Antonie Van Leeuwenhoek. 114(6):741–750. https://doi.org/10.1007/s10482-021-01554-2. Epub 2021 Mar 10. Erratum in: Antonie Van Leeuwenhoek. 115(6):839–840
25. Khan MAW, Bohannan BJM, Nüsslein K, Tiedje JM, Tringe SG, Parlade E, Barberán A, Rodrigues JLM (2019) Deforestation impacts network co-occurrence patterns of microbial communities in Amazon soils. FEMS Microbiol Ecol 95(2):230. https://doi.org/10.1093/femsec/fiy230
26. Klenk HP, Clayton RA, Tomb JF, White O, Nelson KE, Ketchum KA, Dodson RJ, Gwinn M, Hickey EK, Peterson JD, Richardson DL, Kerlavage AR, Graham DE, Kyrpides NC, Fleischmann RD, Quackenbush J, Lee NH, Sutton GG, Gill S, Kirkness EF, Dougherty BA, McKenney K, Adams MD, Loftus B, Peterson S, Reich CI, McNeil LK, Badger JH, Glodek A, Zhou L, Overbeek R, Gocayne JD, Weidman JF, McDonald L, Utterback T, Cotton MD, Spriggs T, Artiach P, Kaine BP, Sykes SM, Sadow PW, D'Andrea KP, Bowman C, Fujii C, Garland SA, Mason TM, Olsen GJ, Fraser CM, Smith HO, Woese CR, Venter JC (1997) The complete genome sequence of the hyperthermophilic, sulphate-reducing archaeon Archaeoglobus fulgidus. Nature 390(6658):364–370. https://doi.org/10.1038/37052
27. Kumar S, Schmitt C, Gorgette O, Marbouty M, Duchateau M, Giai Gianetto Q, Matondo M, Guigner JM, De Reuse H (2022a) Bacterial membrane vesicles as a novel strategy for extrusion of antimicrobial bismuth drug in Helicobacter pylori. mBio 13(5):0163322. https://doi.org/10.1128/mbio.01633-22
28. Kumar S, Vinella D, De Reuse H (2022b) Nickel, an essential virulence determinant of Helicobacter pylori: transport and trafficking pathways and their targeting by bismuth. Adv Microb Physiol 80:1–33. https://doi.org/10.1016/bs.ampbs.2022.01.001
29. Latrubesse EM, Franzinelli E (2005) The late Quaternary evolution of the Negro River, Amazon, Brazil: implications for island and floodplain formation in large anabranching tropical systems. Geomorphology (Amstrerdam) 70:372–397
30. Lovely DR, Stolz JF, Nord GL, Phillips EJP (1987) Anaerobic production of magnetite by a dissimilatory iron-reducing microorganism (PDF). Nature 350(6145):252–254. https://doi.org/10.1038/330252a0.S2CID4234140

31. Meyer F, Paarmann D, D'Souza M, Olson R, Glass EM, Kubal M, Paczian T, Rodriguez A, Stevens R, Wilke A, Wilkening J, Edwards RA (2008) The metagenomics RAST server – a public resource for the automatic phylogenetic and functional analysis of metagenomes. BMC Bioinformatics 19(9):386. https://doi.org/10.1186/1471-2105-9-386
32. Mirza BS, McGlinn DJ, Bohannan BJM, Nüsslein K, Tiedje JM, Rodrigues JLM (2020) Diazotrophs show signs of restoration in Amazon Rain Forest soils with ecosystem rehabilitation. Appl Environ Microbiol 86(10):e00195-e220. https://doi.org/10.1128/AEM.00195-20
33. NascimentoLemos L, Manoharan L, William Mendes L, Monteiro Venturini A, SatlerPylro V, Tsai SM (2020) Metagenome assembled-genomes reveal similar functional profiles of CPR/Patescibacteria phyla in soils. Environ Microbiol Rep 12(6):651–655. https://doi.org/10.1111/1758-2229.12880
34. Navarrete AA, Tsai SM, Mendes LW, Faust K, de Hollander M, Cassman NA, Raes J, van Veen JA (2015) Kuramae EE (2015) Soil microbiome responses to the short-term effects of Amazonian deforestation. Mol Ecol 24(10):2433–2448. https://doi.org/10.1111/mec.13172
35. Nurk S, Meleshko D, Korobeynikov A, Pevzner PA (2017) MetaSPAdes: a new versatile metagenomic assembler. Genome Res 27:824–834
36. Pankratov TA, Tindall BJ, Liesack W, Dedysh SN (2007) Mucilaginibacter paludis gen. nov., sp. nov. and Mucilaginibacter gracilis sp. nov., pectin-, xylan- and laminarin-degrading members of the family Sphingobacteriaceae from acidic Sphagnum peat bog. Int J Syst Evol Microbiol 57(Pt 10):2349–2354. https://doi.org/10.1099/ijs.0.65100-0. Erratum in: Int J Syst Evol Microbiol. 2007;57(Pt12):2979
37. Parks DH, Imelfort M, Skennerton CT, Hugenholtz P, Tyson GW (2015) CheckM: assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes. Genome Res 25:1043–1055
38. Pedrinho A, Mendes LW, Merloti LF, da Fonseca MC, Cannavan FS, Tsai SM (2019) Forest-to-pasture conversion and recovery based on assessment of microbial communities in Eastern Amazon rainforest. FEMS Microbiol Ecol 95(3):236. https://doi.org/10.1093/femsec/fiy236
39. Qian X, Gunturu S, Guo J, Chai B, Cole JR, Gu J, Tiedje JM (2021) Metagenomic analysis reveals the shared and distinct features of the sediment resistome across tundra, temperate prairie, and tropical ecosystems. Microbiome 9(1):108.https://doi.org/10.1186/s40168-021-01047-4
40. Ribeiro A, Villa NA, Lu N et al (1996) Análise do regime pluviométrico da região Amazônica a partir de dados de onze localidades. Rev Bras Meteorol 1:25–35
41. Rodrigue S, Materna AC, Timberlake SC, Blackburn MC, Malmstrom RR, Alm EJ et al (2010) Unlocking short read sequencing for metagenomics. PloS One 5(7):11840. https://doi.org/10.1371/journal.pone.0011840
42. Schmieder R, Edwards R (2011) Quality control and preprocessing of metagenomic datasets. Bioinformatics 27:863–864
43. Schut GJ, Boyd ES, Peters JW, Adams MW (2013) The modular respiratory complexes involved in hydrogen and sulfur metabolism by heterotrophic hyperthermophilic archaea and their evolutionary implications. FEMS Microbiol Rev 37(2):182–203. https://doi.org/10.1111/j.1574-6976.2012.00346.x
44. Seyler and Boaventura (2003) Distribution and partition of trace metals in the Amazon basin. Hydrol Process. https://doi.org/10.1002/hyp.1288
45. Stuiver M, Reimer PJ, Bard E, Beck JW, Burr GS, Hughen KA, Kromer B, McCormac FGV, Der Plight J, SPURK, M (1998) INTCAL98 radiocarbon age calibration, 24,000–0 cal AD. Radiocarbon 40:1041–1083
46. Swain EB (1985) Measurement and interpretation of sedimentary pigments. Freshw Biol 15:53–75
47. Torres IC, Inglett PW, Brenner M, Kenney WF, Reddy KR (2012) Stable isotope (δ 13C and δ 15N) values of sediment organic matter in subtropical lakes of different trophic status. J Paleolimnol 47(4):693–706. https://doi.org/10.1007/s10933-012-9593-6
48. Tschoeke DA, Coutinho FH, Leomil L, Cavalcanti G, Silva BS, Garcia GD, Dos Anjos LC, Nascimento LB, Moreira LS, Otsuki K, Cordeiro RC, Rezende CE, Thompson FL, Thompson CC (2020) New bacterial and archaeal lineages discovered in organic rich sediments of a large tropical Bay. Mar Genomics 54:100789. https://doi.org/10.1016/j.margen.2020.100789
49. von Jan M, Lapidus A, Del Rio TG, Copeland A, Tice H, Cheng JF, Lucas S, Chen F, Nolan M, Goodwin L, Han C, Pitluck S, Liolios K, Ivanova N, Mavromatis K, Ovchinnikova G, Chertkov O, Pati A, Chen A, Palaniappan K, Land M, Hauser L, Chang YJ, Jeffries CD, Saunders E, Brettin T, Detter JC, Chain P, Eichinger K, Huber H, Spring S, Rohde M, Göker M, Wirth R, Woyke T, Bristow J, Eisen JA, Markowitz V, Hugenholtz P, Kyrpides NC, Klenk HP (2010) Complete genome sequence of Archaeoglobus profundus type strain (AV18). Stand Genomic Sci 2(3):327–346. https://doi.org/10.4056/sigs.942153