Environmental Microbiology

ENVIRONMENTAL MICROBIOLOGY

REVIEW OPEN ACCESS

The Bacterial Genus *Ramlibacter*: Betaproteobacteria Capable of Surviving in Oligotrophic Environments Thanks to Several Shared Genetic Adaptation Traits

Gilles De Luca | Mohamed Barakat | André Verméglio | Wafa Achouak | Thierry Heulin

Aix Marseille Univ, CEA, CNRS, BIAM, Lab Microbial Ecology of the Rhizosphere (LEMiRE), Saint-Paul-Lez-Durance, France

Correspondence: Gilles De Luca (gilles.de-luca@cea.fr)

Received: 20 November 2024 | **Revised:** 17 January 2025 | **Accepted:** 20 January 2025

Keywords: ecology | oligotrophy | phylogeny | *Ramlibacter*

ABSTRACT

Ramlibacter tataouinensis, the type species of the genus *Ramlibacter*, is renowned for its ability to thrive in hot, arid and nutrient-poor desert soils. To investigate whether its adaptive properties are shared across all 20 currently described *Ramlibacter* species found in diverse terrestrial and aquatic habitats worldwide, we conducted a comprehensive analysis of 16S rRNA sequences and genomic information available from the literature. Our study encompassed approximately 40 deposited genomes, allowing us to propose a genomic phylogeny that aligns with the 16S rRNA phylogeny. Our findings reveal several conserved features across the genus *Ramlibacter*. This includes the presence of light sensors, environmental sensing networks, organic carbon and phosphate acquisition systems and the ability to store carbon and energy in the form of polyhydroxyalkanoate or polyphosphate granules. These shared traits rationalise the widespread distribution of *Ramlibacter* in oligotrophic terrestrial and aquatic environments. They also explain the genus' ability to withstand desiccation, endure extended periods of starvation, and survive in nutrient-depleted conditions. Notably, certain adaptive features are further enhanced in several species by their pleiomorphism and ability to form cysts. Overall, our study not only highlights the ecological adaptations of *Ramlibacter* species but also extends our understanding of microbial ecology in oligotrophic environments.

1 | Introduction

The genus *Ramlibacter* was first identified on fragments of the Tataouine meteorite, which impacted in a semi-arid desert, near the town of Tataouine in southern Tunisia in 1931 (Lacroix 1931). Large meteorite fragments were collected the day after its fall and preserved at the Muséum National d'Histoire Naturelle in Paris, France. In 1994, researchers collected weathered fragments of the meteorite to analyse the effects of 63 years of terrestrial exposure (Barrat et al. 1998). Scanning electron microscopy revealed distinct alteration zones on the meteorite's surface, particularly on pyroxene and chromite, and secondary calcite formation indicative of terrestrial weathering (Barrat et al. 1998, 1999; Gillet et al. 2000). To identify the microbial agents responsible for these modifications, two bacterial strains were isolated from a meteorite fragment embedded in sandy soil. Subsequent taxonomic analysis classified these two strains as representatives of a novel genus within the *Betaproteobacteria* and type strains of two distinct species, *Ramlibacter tataouinensis* and *Ramlibacter henchirensis* (Heulin et al. 2003). Chanal et al. confirmed the presence of *Ramlibacter* in the sandy soil of Tataouine using a culture-independent method (cloning-sequencing of 16S rDNA) (Chanal et al. 2006). This discovery initiated extensive research on this bacterial genus, revealing various molecular and physiological characteristics that elucidate its adaptation to the desert environment, particularly its ability to thrive under oligotrophic conditions and endure extended periods of dormancy due to desiccation and/or starvation (Gommeaux et al. 2005; De Luca

et al. 2011, 2019). Although these bacteria are easily cultivated, their molecular genetic manipulations are limited (construction of insertion mutants by electropermeation of a suicide plasmid, replicative plasmid) (De Luca et al. 2019).

Since this initial discovery, the *Ramlibacter* genus has proven to be remarkably widespread and prevalent across diverse worldwide environments (Delgado-Baquerizo et al. 2018). Both culture-independent and culture-dependent methods have demonstrated the ubiquity and adaptability of this genus. Culture-independent methods, such as PCR amplification of 16S rRNA followed by cloning or fingerprinting, have uncovered *Ramlibacter* in various habitats. This includes semiarid soil (Rutz and Kieft 2004), trichloroethene-contaminated groundwater undergoing propane-stimulated bioremediation (Connon et al. 2005), soil crusts of the Colorado Plateau (Gundlapally and Garcia-Pichel 2006), association with *Gigaspora* spores (Long et al. 2008), soil from agricultural systems (Jangid et al. 2008), heavy metal-contaminated acidic waters originating from zinc mine residues (Almeida et al. 2009), or *m*-xylene-contaminated soil (Xie et al. 2010). Additionally, classical isolation methods successfully retrieved *Ramlibacter* strains from soil samples (Shrestha et al. 2007). Advanced sequencing technologies have further expanded our understanding of *Ramlibacter* distribution. Sequences of the *Ramlibacter* 16S rRNA gene were identified through pyrosequencing in the murine caecum (Barfod et al. 2013), during micro-aerobic hydrolysis of activated sludges (Zhou et al. 2014) and in the injection water of oil fields in Algeria (Lenchi et al. 2013).

Since the initial description of *R. tataouinensis* and *R. henchirensis*, the genus has therefore expanded considerably, now encompassing 20 officially named and described species. In 2012, *R. ginsenosidimutans*, isolated from Korean cultivated soil, was the first additional species characterised in the genus (Wang, An, et al. 2012). Two years later, in 2014, *R. solisilvae* was identified from Vietnamese forest soil, leading to an emended description of the *Ramlibacter* genus (Lee et al. 2014). Subsequently, 16 new *Ramlibacter* species were officially named and described, bringing the total to 20 species. These species were found in a wide range of environments, such as soil habitats (Wang, An, et al. 2012; Lee et al. 2014; Kang et al. 2022; Lim et al. 2023; Lee and Cha 2017; Dahal et al. 2022; Yan et al. 2017; Akter et al. 2020; Chaudhary and Kim 2017; Zhang et al. 2019; Khan et al. 2021) and aquatic environments (Kang et al. 2022; Props et al. 2019; Yu and Leadbetter 2020; Kim et al. 2021). The widespread presence of *Ramlibacter* is evidenced by over 380 PubMed Central publications referencing *Ramlibacter* in their study samples, by geographical analysis of seventy 16S rRNA sequences and by bacterial strains deposited in the NCBI Taxonomy Browser. The ability of *Ramlibacter* species to thrive in diverse environments, ranging from hot and cold deserts to various oligotrophic ecosystems, underlines their exceptional adaptability and ecological importance. This widespread distribution not only demonstrates their colonisation success but also highlights the shared ecological versatility and adaptive strategies among *Ramlibacter* species.

This review aims to comprehensively analyse the microbial ecology and adaptation mechanisms of *Ramlibacter* species in oligotrophic conditions, providing insights into their ecological significance. We will explore the localisation and distribution of *Ramlibacter* species, examine the genus's taxonomy and phylogeny, investigate key processes and mechanisms involved in their survival, and analyse their adaptation strategies, with a particular focus on *R. tataouinensis* adaptation to semi-arid desert.

This comprehensive analysis will not only enhance our understanding of the ecological significance of *Ramlibacter* but also contribute to the broader field of microbial ecology, particularly in the context of bacterial adaptation to extreme and nutrient-limited environments.

2 | Biogeography and Phylogeny of the *Ramlibacter* Genus: *Betaproteobacteria* From Oligotrophic Environments

2.1 | Census and Biogeography: Ubiquitous Soil Bacteria Found in Terrestrial and Aquatic Environments

Our understanding of the genus *Ramlibacter*, initially described in 2003, requires significant re-evaluation in light of discoveries over the past two decades (Table S1, https://doi.org/10.5281/zenodo.14761071). Recent research revealed the widespread presence of *Ramlibacter*, identifying it as one of the 500 most abundant and ubiquitous phylotypes in soils across six continents. A comprehensive study of 237 locations found *Ramlibacter* present in 90.7% of sites, with an average abundance of 0.05% (Delgado-Baquerizo et al. 2018). *Ramlibacter* sequences were recovered from a wide range of soil types worldwide, including paddy soils (Shrestha et al. 2007), hot deserts (Heulin et al. 2003; Chanal et al. 2006; Rutz and Kieft 2004; Gundlapally and Garcia-Pichel 2006; Hegde et al. 2015; Meier et al. 2021; Molina-Menor et al. 2020), Arctic (Parks et al. 2017) and Antarctic deserts (Chan-Yam et al. 2019). Furthermore, the genus *Ramlibacter* was found less frequently in freshwater (Vilardi et al. 2022), ground water (Pedron et al. 2019), as well as in terrestrial hot spring habitats (Mehetre et al. 2018; Saxena et al. 2016) and Nagorno-Karabakh geothermal spring (Saghatelyan et al. 2021). *Ramlibacter* was also found associated with plant roots (Grönemeyer et al. 2012), pond freshwater sediment (Huang, Straub, et al. 2021), frond of pond aquatic plant (Iwashita et al. 2020), phyllosphere of tree (Bai et al. 2015; Kobayashi and Aoyagi 2019), waste water (Schneider et al. 2021), activated sludges (Huang, Deng, et al. 2021), anaerobic digesters (Campanaro et al. 2020), termite gut (Butera et al. 2016) and air (Wang, Ma, et al. 2012). Notably, 14 out of 20 of the type strains of *Ramlibacter* species were isolated from soils in Southeast Asia, specifically South Korea, China and Vietnam (Wang, An, et al. 2012; Lee et al. 2014; Kang et al. 2022; Lim et al. 2023; Lee and Cha 2017; Dahal et al. 2022; Yan et al. 2017; Akter et al. 2020; Chaudhary and Kim 2017; Zhang et al. 2019; Khan et al. 2021). The type strains of the three most recently described species, *R. aquaticus* (Props et al. 2019), *R. lithotrophicus* (Yu and Leadbetter 2020) and *R. algicola* (Kim et al. 2021), were obtained from freshwater samples, and *R. cellulosilyticus* (Kang et al. 2022) isolated from a shallow coastal seawater sample.

To provide a comprehensive overview of the global distribution of genus *Ramlibacter*, we compiled these diverse datasets into

an atlas (Figure 1) illustrating the geographical distribution of *Ramlibacter* strains sampled worldwide.

This expanded ecological range demonstrates the adaptability of *Ramlibacter* species and highlights their potential significance in global biogeochemical cycles and ecosystem functioning.

2.2 | Phylogenic Analysis and Taxonomy of the *Ramlibacter* Genus

Since its initial description in 2003, the *Ramlibacter* genus, belonging to the family *Comamonadaceae*, order *Burkholderiales* and class *Betaproteobacteria*, has expanded significantly. To date, 20 species have been described, with 19 species validly published according to the List of Prokaryotic names with Standing in Nomenclature (LPSN), excluding *R. lithotrophicus* (Yu and Leadbetter 2020). The majority of these species were described within the last 6 years (Tables S1 and S2, https://doi.org/10.5281/zenodo.14761071). Members of the genus *Ramlibacter* have a gram-negative cell wall, rod- or coccoid-shaped morphology, aerobic metabolism, contain ubiquinone (Q-8) as a major respiratory quinone and a high G+C content ranging from 66.6 to 69.9 mol% [24; 25; Tables S1 and S2, https://doi.org/10.5281/zenodo.14761071]. Recent phylogenetic studies by Kim et al. (2021), Dahal et al. (2022), Kang et al. (2022) and Lim et al. (2023) provided valuable insights into the phylogenetic position of *Ramlibacter* within the *Comamonadaceae*, but none of them include the 20 type strains in a single phylogenetic tree. Based on these results, we conducted a comprehensive phylogenetic analysis incorporating two new maximum-likelihood trees using 16S rRNA gene sequences (Figures 2 and S1), adapted from the GGDC web server (Meier-Kolthoff et al. 2022) from the most exhaustive work of Kim et al. (2021) (see Supporting Information: Appendix for details). Our analysis incorporates more than 80 16S rRNA sequences from *Ramlibacter* strains available on the NCBI Taxonomy Browser (Table S1, https://doi.org/10.5281/zenodo.14761071). This comprehensive dataset included 20 well-characterised *Ramlibacter* species (Figure 2, Tables S1 and S2, https://doi.org/10.5281/zenodo.14761071) and 27 additional strains reported in the literature (Table S1, https://doi.org/10.5281/zenodo.14761071). To enhance the diversity of our analysis, we included sequences of uncultured *Ramlibacter* obtained through cloning, metabarcoding, and metagenomics studies. Additionally, we incorporated 42 non-redundant draft genome and genomes from metagenomics studies or isolated bacteria deposited until the beginning of 2022, including 24 previously published genomes from various environments, with a focus on soils (Table S1, https://doi.org/10.5281/zenodo.14761071). To ensure accurate taxonomic classification, we selected a subset of sequences longer than 1349 bp from our larger dataset that were computed with the GGDC web server, for the 20 type strains (Figure 2) and 13 additional published strains (Figure S1) (see Table S1, https://doi.org/10.5281/zenodo.14761071and Supporting Information: Appendix for details). Our phylogenetic analysis, based on 16S rRNA sequences, confirms the positioning of *Ramlibacter* species within a distinct phylogenetic group and the proximity of some of these uncharacterised strains to *Caenimonas*.

This comprehensive phylogenetic analysis provides a robust framework for understanding the evolutionary relationships within the *Ramlibacter* genus and its position within the broader

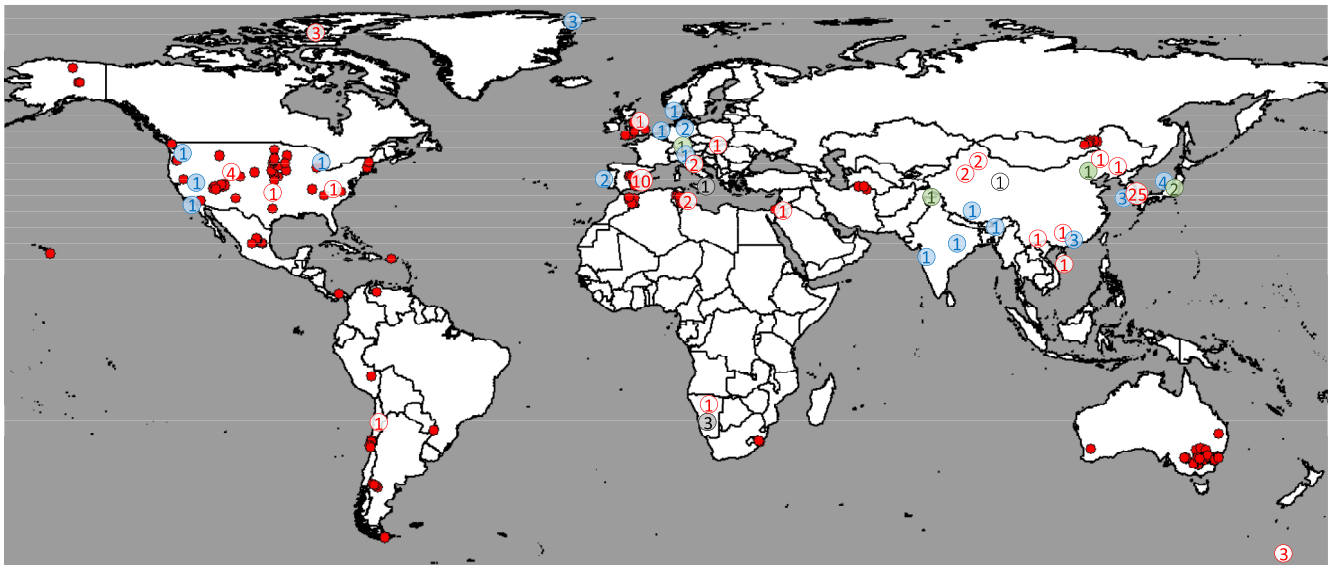

FIGURE 1 | Atlas of *Ramlibacter* isolates and clones found worldwide (reprinted and modified from (Delgado-Baquerizo et al. 2018) with permission from AAAS and the authors). The sampling location of soil clones (mainly from (Delgado-Baquerizo et al. 2018) with permission from AAAS) are represented by solid red stars, and location of isolates or metagenome assembly genomes (MAGs) (103 strains from Table S1) by coloured circles [red: Soil; blue: Aquatic, wastewater sludge (Mumbay, Seattle, Denmark, Germany and Hong-Kong), or Greenland glacier; green: *Arabidopsis* phyllosphere (Swiss), bark of Japanese maple (Japan) and endophytic bacteria from apple tree (Kashmir Valley, India) or soybean stalk (China), pale grey: Termite gut (Italy) or spider nest (Namibia), black: Cave air (China)], and the number inscribed in the circle represents the number of isolates from the same region or geographical area (for example: 10 and 25 soil strains in southern Spain and South Korea, respectively, and 3 and 4 aquatic strains in South Korea and Japan, respectively; see Table S1 for details).

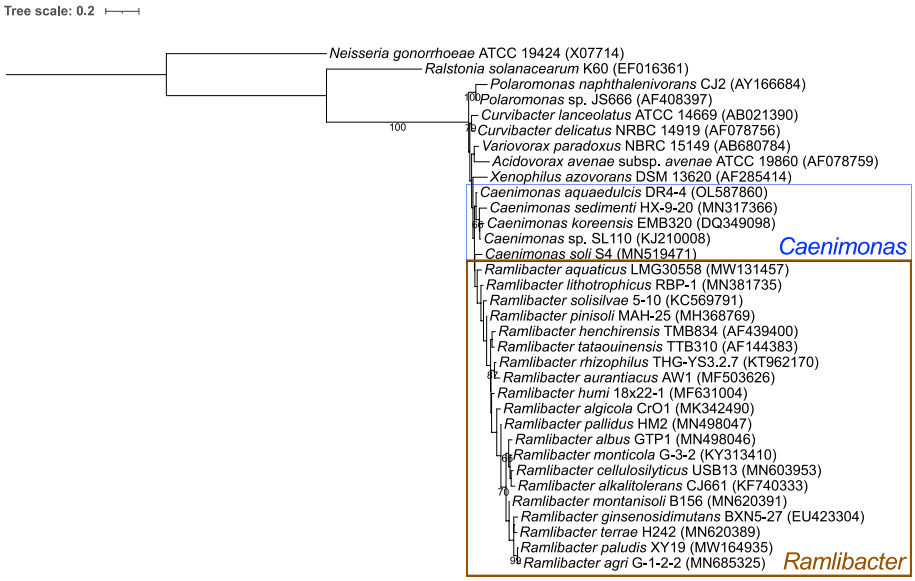

FIGURE 2 | A maximum likelihood tree based on 16S rRNA gene sequences showing phylogenetic relationships between the type strains of 20 *Ramlibacter* species. Only *Caenimonas* sp. SL110 is not a type strain. GenBank accession numbers for the 16S rRNA gene sequences are indicated in parentheses. *Neisseria gonorrhoeae* ATCC 19424^{T} was used as an outgroup. Bar, 0.2 nucleotide substitutions per position. Deduced maximum likelihood tree according to the GTR + GAMMA model and rooted by midpoint-rooting. Branches are scaled according to the expected number of substitutions per site. The numbers above the branches are the support values when greater than 60% according to maximum parsimony bootstrapping (adapted from (Kim et al. 2021) with the GGDC web server (Meier-Kolthoff et al. 2022), see Supporting Information: Appendix for details).

context of *Betaproteobacteria*. It also highlights the need for continued taxonomic refinement as new strains are discovered and characterised.

2.3 | Phylogenomic Analysis of the *Ramlibacter* Genus

Recent availability of the genomic sequences of the 20 *Ramlibacter* type strains has enabled comprehensive phylogenomic analyses, complementing 16S rRNA phylogeny and providing deeper insight into the evolutionary relationships among closely related strains. Data published by Kang et al. (2022) and Lim et al. (2023) revealed that the genomes of the type strains of *R. cellulosilyticus* and *R. paludis*, respectively, exhibit average nucleotide identity (ANI) values of around 80% with the other *Ramlibacter* type strains, and average amino-acid identity (AAI) values of between 66% and 80%. These percentages, significantly below the 95% species-level cut-off threshold, indicate genus-level similarity but distinct species. To address the limitations of previously phylogenomic trees, which excluded several type strains (6 and 15 type strains, respectively), we performed an analysis of the 47 *Ramlibacter* genomes available in Genbank (NCBI) using the Genoscope microbial genome annotation and analysis platform 'MicroScope' (Vallenet et al. 2020). We employed the integrated MASH software package [58, and see Supporting Information: Appendix for details] to assess genomic similarities by comparing complete genome sequences pairwise through *k*-mer homology analysis. This approach calculates a genomic distance (D) correlated to the ANI value ($D \approx 1 - \text{ANI}$). The resulting tree topology reflects the genomic relationship among the analysed strains (Figure 3), with 19 *Ramlibacter* type strains clustered, consistent with our 16S rRNA gene-based tree (Figure 2). The species *R. lithotrophicus*, which has not been validly published, is excluded from this cluster, suggesting that this species should be validated using a polyphasic taxonomy approach. In addition, the presence of the *Caenimonas sedimenti* type strain (Xu et al. 2020) within the *Ramlibacter* cluster raises questions as to the assignment of the genus to this species and warrants further investigation. Furthermore, the taxonomic classification of 8 *Ramlibacter* strains associated with the *Caenimonas* cluster, as well as 6 other '*Ramlibacter*' strains outside the *Ramlibacter* cluster, will require careful examination (Figure 3). To validate our tree topology, we utilised the DSMZ Type Genome Server (TYGS), which employs a Genome Blast Distance Phylogeny (GBDP) approach, based on genome-wide homology searches using BLASTN, followed by a distance matrix computation and tree construction (Meier-Kolthoff and Göker 2019; Meier-Kolthoff et al. 2022) (see Supporting Information: Appendix for details). This analysis, which automatically includes genomes of closely related type strains (Figure S2), confirmed the unexpected positioning of *C. sedimenti* and the exclusion of *R. lithotrophicus* from the *Ramlibacter* cluster.

The GTDB taxonomy (Parks et al. 2018), based on genome trees inferred using FastTree (Price et al. 2009) from an aligned concatenated set of 120 ubiquitous bacterial single-copy marker proteins, and its derived GTDB toolkit (Chaumeil et al. 2019), based on a combination of the placement of a new genome in the GTDB reference tree, its relative evolutionary divergence (RED) and its ANI to reference genomes, proposes classifying the *Caenimonas* strains included in their database (*C. koreensis*, *C. soli* and *Caenimonas* sp. SL110) or not (*C. sedimenti*, *C. aquaedulcis*) within the genus *Ramlibacter*. On the basis of genomic data alone, it is difficult to distinguish clearly between the genera *Ramlibacter* and *Caenimonas*, in contradiction with the data from a polyphasic approach clearly separating the two genera (Ryu et al. 2008).

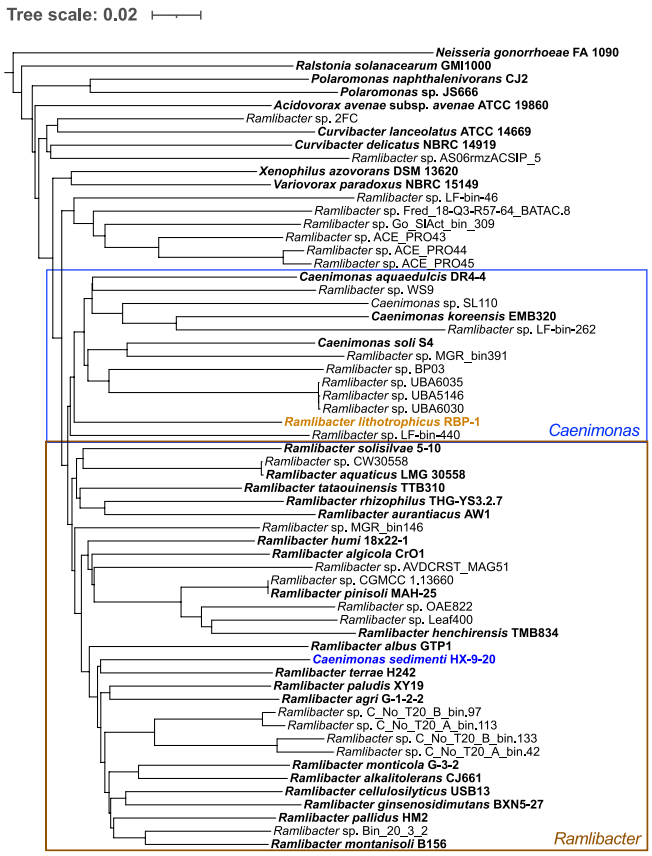

FIGURE 3 | Neighbour-joining genome clustering tree based on pairwise distances calculation of a set of genomes showing genomic similarity between 47 *Ramlibacter* genomes deposited in GenBank and some *Comamonadaceae* genomes. Genomic similarity was estimated using Mash, a software that calculates the distance between two genomes (Ondov et al. 2016), integrated in the 'MicroScope' genome server (Vallenet et al. 2020). *Neisseria gonorrhoeae* FA 1090 was used as an outgroup. Bar, 0.02. Type strains are shown in bold (see Supporting Information: Appendix for details).

These findings highlight the need for a comprehensive re-evaluation of the taxonomic relationships within and between the *Ramlibacter* and *Caenimonas* genera, emphasising the importance of integrating a polyphasic approach for accurate bacterial taxonomy.

2.4 | Unique Traits of *Ramlibacter* Species: Metals Tolerance and Catabolic Versatility

This section highlights the unique traits of *Ramlibacter* strains, focusing on their metal tolerance and catabolic versatility.

R. tataouinensis demonstrates its ability to colonise orthopyroxene, the main component of the Tataouine meteorite, reducing mineral alteration by creating an amorphous layer upon attachment (Benzerara, Barakat, et al. 2004). Additionally, *R. tataouinensis* plays a role in calcium phosphate mineralisation through the activity of five alkaline phosphatases capable of hydrolysing a wide range of organic phosphorus substrates (Benzerara, Menguy, et al. 2004; Skouri-Panet et al. 2017).

Several *Ramlibacter* strains have demonstrated tolerance to various metals (copper (Bernard et al. 2009), zinc (Luo et al. 2018), nickel (Remenár et al. 2017) and chromium) but displayed sensitivity to cadmium and lead (Caliz et al. 2012; Cáliz et al. 2013). *R. ginsenosidimutans* exhibits diverse catabolic capabilities such as the transformation of ginsenoside, an active component of ginseng (Wang, An, et al. 2012). *R. sp.* T-55 (isolated from a termite gut microbiota) and *R. cellulosilyticus* (from coastal seawater) can degrade carboxymethyl cellulose and cellulose, respectively (Kang et al. 2022; Butera et al. 2016). DNA-SIP studies have revealed the ability of various *Ramlibacter* species to metabolise various organic molecules, including the herbicide 2,4-D (2,4-dichlorophenoxyacetic acid) (Cupples and Sims 2007), butane (Deng et al. 2018) or isoprene (Larke-Mejía et al. 2019). This volatile compound emitted by trees is decomposed by *Ramlibacter* sp. strain WS9 isolated from a willow soil (Larke-Mejía et al. 2019).

Several *Ramlibacter* strains show potential for degrading various aromatic organic pollutants. In silico genomics studies identified two gene clusters in *R. tataouinensis* TTB310^T potentially enabling terephthalic acid degradation (*tph*) (Salvador et al. 2019) and protocatechuate (3,4-dihydroxybenzoate) degradation to pyruvate and oxaloacetate (*pmdEFDABC*) (Hickey et al. 2012). Supporting their involvement in aromatic compound degradation, *Ramlibacter* clones were found in a DNA-SIP library from microcosms of river sediment contaminated with the organochlorine aromatic compound (Sul et al. 2009). One *Ramlibacter* clone was identified as the dominant *m*-xylene degrader in sediment from a site contaminated with gasoline (Xie et al. 2010). Finally, among the most abundant bacteria, the POES5 isolate, identified as belonging to the genus *Ramlibacter*, has demonstrated the ability to degrade POESH (polyoxyethylene sorbitol hexaoleate) surfactant as well as phenanthrene (3-benzene ring compound) (Singleton et al. 2016).

These diverse metabolic capabilities highlight the ecological significance of *Ramlibacter* species in various environments, particularly in the context of bioremediation and biogeochemical cycling. Further research into the molecular mechanisms underlying these traits could provide valuable insights into bacterial adaptation and potential biotechnological applications.

3 | Adaptation of *R. tataouinensis* to Desert Environments: Survival Strategies in Nutrient-Poor and Desiccation-Prone Conditions

R. tataouinensis exhibits adaptive traits to survive in nutrient-poor environments and to resist prolonged desiccation. This bacterium's unique life cycle and genomic features provide insights into its survival strategies in semi-desert conditions (Heulin et al. 2017).

3.1 | Complex Cell Cycle of *R. tataouinensis* Including a Cyst-Dividing Stage

R. tataouinensis TTB310^T is distinguished by its pleiomorphism, characterised by the coexistence of two bacterial morphotypes:

a spherical or coccoid form surrounded by a protective exopolysaccharide (Jivkova et al. 2022), resistant to desiccation and capable of dividing (known as the cyst-dividing form), and a rod-shaped form (also capable of dividing) that presents variable size and type of motility on nutrient agar (Heulin et al. 2003; Benzerara, Barakat, et al. 2004). Both forms of *R. tataouinensis* contain granules of polyhydroxyalkanoate (PHA) for carbon and energy storage (Heulin et al. 2003). A morphological transition between the cyst form and the rod form requires the lysis of the exopolysaccharide and a reduction in the cyst volume by about two-thirds but no significant change in cell surface area (Gommeaux et al. 2005). This process, known as 'cyst-rod-cyst differentiation', consists of cyst division leading from one cyst to the formation of two motile rods within 3 h and differs significantly from the germination of *Azotobacter* cysts (Heulin et al. 2003; Gommeaux et al. 2005; Sadoff 1975). In 2011, a more detailed cell cycle model was proposed on the basis that rods can divide without going through the cyst-division phase (Figure S3) (De Luca et al. 2011). In this revised model, *R. tataouinensis*, growing on nutritive agar in the dark, develops mainly as microcolonies of cysts (cyst-division), generating motile rods at the periphery of the microcolonies. After a brief period of exploration on the plate surface, these rods become immobilised, transformed into a coccoid form, and give rise to new cyst microcolonies (Heulin et al. 2003; Gommeaux et al. 2005; Benzerara, Barakat, et al. 2004). This alternating pattern in cyst microcolonies and short periods of exploration by mobile rods appears to be an effective adaptive strategy for discovering favourable environment and periods of development in a hot and dry environment.

3.2 | Additional Features of Adaptation to Desert Conditions and Desiccation Evidenced by Genomic Data: The Special Role of Light in the Cell Cycle

Genomic sequencing of *R. tataouinensis* TTB310^T has shed light on the use of conventional and specialised mechanisms for adaptation to semi-desert life (De Luca et al. 2011). Genome analysis revealed the presence of glycosyltransferases and glycosylhydrolases, suggesting their potential involvement, respectively, in the synthesis and the hydrolysis of the exopolysaccharide surrounding the cysts (Jivkova et al. 2022). Additionally, the synthesis of the compatible solute trehalose, known to aid in desiccation tolerance, was observed. This result is significant, given the likely scarcity of such molecules in the desert environment, which is by definition very poor in organic matter. In addition, *R. tataouinensis* possesses PHA, an energy storage molecule also found in *Azotobacter* cysts (Heulin et al. 2003; Sadoff 1975). To respond effectively to the dynamic changes in the external environment, *R. tataouinensis* has also evolved a large and distinctive two-component signalling network comprising 131 proteins and representing 3.5% of its genes (Figures 4 and S4). This network has intriguing characteristics, including a probable convergence with more histidine kinases than response regulators, one half dedicated to intracellular signal-sensing, one part regulated by post-transcriptional regulations (allowing a faster adaptation compared with transcriptional regulations), and finally has two chemotaxis systems dedicated to a form of gliding and/or twitching mobility involving type IV pili (Heulin et al. 2003; De Luca et al. 2011; Benzerara, Barakat, et al. 2004).

More intriguingly, *R. tataouinensis* exhibits one blue-light-sensing histidine kinase (Rta_12790) and two red/infrared-light-sensing ones (bacteriophytochromes BphP1 Rta_25470 and BphP2 Rta_28950), as well as various 'Blue Light-sensing Using Flavins' (BLUF) proteins, potentially involved in wavelength-specific sensing (Figures 4, 5 and S4). The latter and/or the blue-light-sensing histidine kinase could be involved in the strong growth inhibition of growth by relatively low-intensity white or blue light illumination observed in *R. tataouinensis* TTB310^T (De Luca et al. 2019). The bacteriophytochromes BphP1 and BphP2 themselves play an essential role in the cell cycle of this strain (De Luca et al. 2019; Baker et al. 2016). Studies have shown that rod divisions predominate under red or far-red light (De Luca et al. 2019), while cyst divisions and the complete 'cyst-rod-cyst' cycle predominate in the dark, as previously stated (Heulin et al. 2003; Gommeaux et al. 2005; Benzerara, Barakat, et al. 2004). Mutant phenotypes resulting from the inactivation of genes encoding bacteriophytochromes or heme oxygenase clearly indicate the involvement of these bacteriophytochromes in the regulation of rod division. This process may facilitate rapid rod division at sunrise, after dew formation, but before the progressive onset of desiccation. The potential presence of a rudimentary hourglass is also suggested by the presence of a gene encoding a phosphorylatable KaiC homologue, followed by a histidine kinase (Figures 4 and S4) (De Luca et al. 2011). This hourglass could have a synchronising function and be used to anticipate water availability during dew formation at the middle or at the end of winter nights in the desert, thus aligning the growth window with cyclical periods of water availability.

4 | Bibliographic, Pan-Genome and Data Mining Analyses of *Ramlibacter* Species: Phenotypic Traits and Gene Repertoire for Oligotrophic Adaptation

4.1 | Bibliographic Data: Shared Adaptive Phenotypic Traits for Starvation and Desiccation

Pleiomorphism, previously observed in *R. tataouinensis* and *R. henchirensis* (Heulin et al. 2003), was identified in 7 out of the 18 well-characterised species, while the remaining 12 were described solely as rod-shaped cells (Table S2, https://doi.org/10.5281/zenodo.14761071). Although detailed cell cycle data for other *Ramlibacter* species is lacking, several strains (Hegde et al. 2015; Molina-Menor et al. 2020) or clones (Chanal et al. 2006; Rutz and Kieft 2004; Gundlapally and Garcia-Pichel 2006; Meier et al. 2021) have been found in desert environments, suggesting this adaptation is relatively shared within this genus. Some well-characterised strains from non-desert environments exhibit features similar to those of *R. tataouinensis*. For instance, a *R. aquaticus* strain isolated from freshwater produces coccoid cyst-like cells, accumulates polyphosphate and PHA granules and demonstrates considerable plasticity by adjusting its cell size in response to nutrient concentrations (Props et al. 2019). Microscopy images of a *R. solisilvae* strain isolated from soil reveal rods, coccoids and coccoid aggregates containing PHA granules, closely resembling *R. tataouinensis* TTB310^T (Lee et al. 2014). PHA granules were also observed in the pleiomorphic species, including *R. solisilvae*, *R. monticola* and *R. humi* isolated from forest soils, as well as *R. aquaticus*

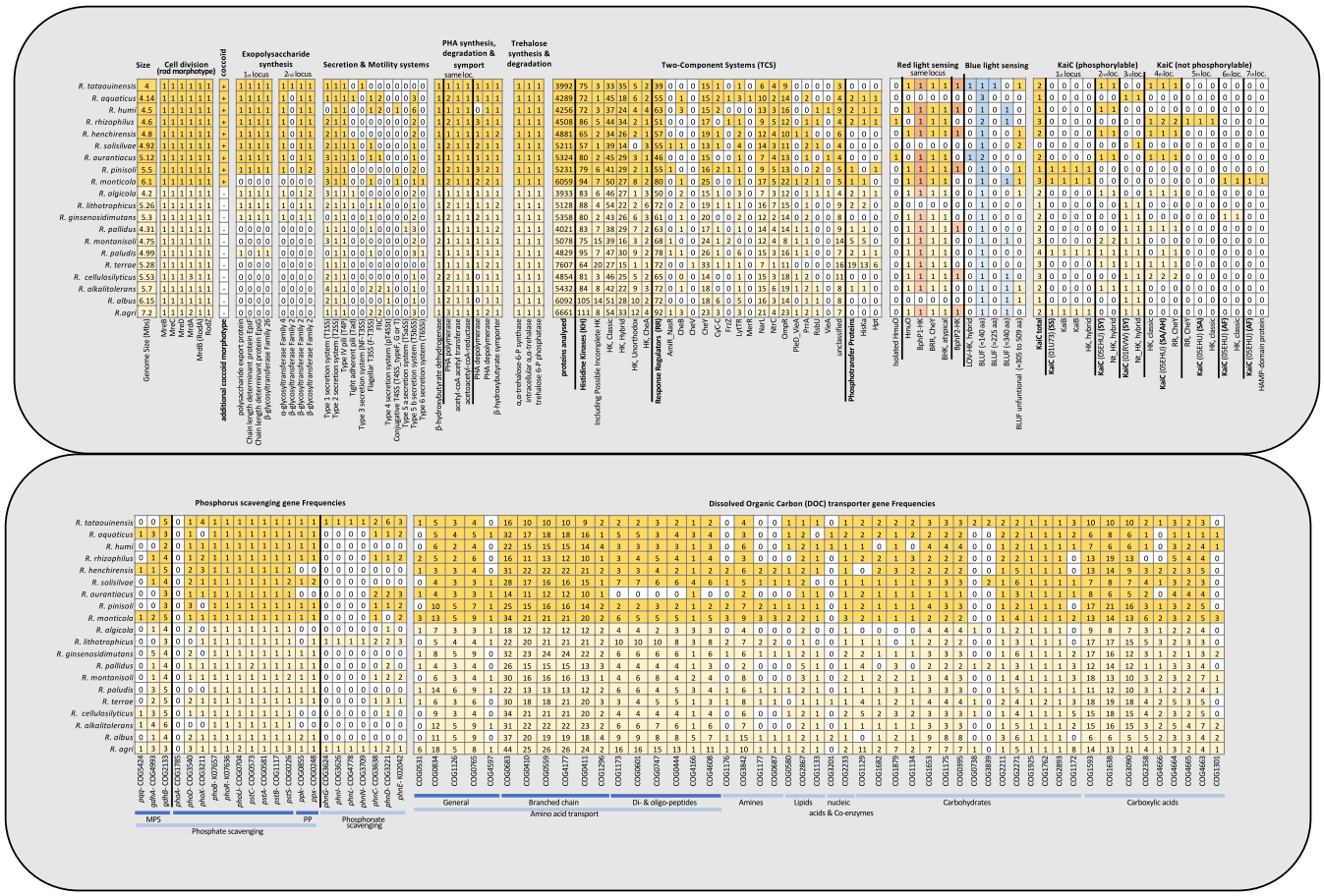

FIGURE 4 | Frequencies of some 'key' systems, proteins or genes in the genomes of the 20 *Ramlibacter-type* strains. The genomes of type strains are grouped according to their ability to coexist in coccoid and rod forms (first nine lines), to be pure rods, and to possess two conserved polysaccharide synthesis loci (next three lines), or to be pure rods and to exhibit an absence or a partial conservation of loci for the synthesis of this specific polysaccharide (last eight lines), inter-ranked according to genome size in ascending order. White boxes with a (–) indicate the absence of coccoid forms in the species description ('pure rods'), and those with a (0) indicate the absence of a gene or a complete system. Dark yellow boxes with a (+) sign indicate the coexistence of cocci and rods in the species, while yellow boxes (dark and light) with a number indicate the presence of one or more genes or a complete system. Depending on nucleotide sequence coverage and error rate, the number of artefact-free CoDing Sequences for some strains may differ more or less from genome size (see Table S3 for details). Boxes indicating the frequency of proteins sensing red or blue light are coloured red and blue, respectively. Key proteins involved in rod morphogenesis in *E. coli* [MreBCDMrdA operon, MrdB (RodA) and RodZ)] and absence (–) or coexistence of a coccoid morphotype (+) are indicated. For secretion and motility systems, the number in brackets indicates the number of proteins present [flagellin (FliC) and type 5 secretion system, single protein] or the number of complete systems composed of several proteins. For KaiC proteins, the number in brackets indicates the protein's EGGNOG category and the letters the amino acids names corresponding to the two phosphorylatable residues found in KaiC of *Synechococcus elongatus* (ST, Ser Thr). For phosphorus scavenging and dissolved organic carbon (DOC) transporter genes, each column represents a single COG. BHK, bacteriophytochrome histidine kinase; BHK, bacteriophytochrome histidine kinase (or red-light-sensing histidine kinase); BLUF, blue light sensor protein using flavin; BRR, bacteriophytochrome response regulator; HK, histidine kinase; LOV-HK, light oxygen voltage domain containing histidine kinase (or blue-light-sensing histidine kinase); MPS, mineral phosphate solubilisation; PhoA, acid phosphatase; PhoD and PhoX, alkaline phosphatases; Ppk, polyphosphate kinase; Ppx, exopolyphosphatase; RR, response regulator.

(inferred from genomic data), *R. alkalitolerans*, and *R. agri*, two species exclusively present in rod forms (Table S2, https://doi.org/10.5281/zenodo.14761071). Thus, pleiomorphism and PHA presence in both coccoid and rod forms are shared among *Ramlibacter* species.

This adaptation to nutrient-depleted environments appears prevalent among most sequenced *Ramlibacter* species, as detailed below (Figures 4 and S4). The combination of pleiomorphism and PHA accumulation likely confers a significant advantage in oligotrophic conditions, allowing these bacteria to persist in diverse nutrient-poor habitats.

4.2 | Pan-Genome Analysis: Limited Repertoire of Species-Specific Genes With Predominantly Hypothetical Functions

To identify genes potentially involved in adaptation to diverse ecological niches, we conducted a pan-genomic analysis on 29 genomes within the strictly defined *Ramlibacter* clade (Figure 3), including the two type strains *R. lithotrophicus* and *R. solisilvae* (Table S3, https://doi.org/10.5281/zenodo.14761071). The pan-genome was computed using the Genoscope's Pan/Core-Genome tool (Vallenet et al. 2020), from a set of selected genomes, comprising the core genome (genes families shared by

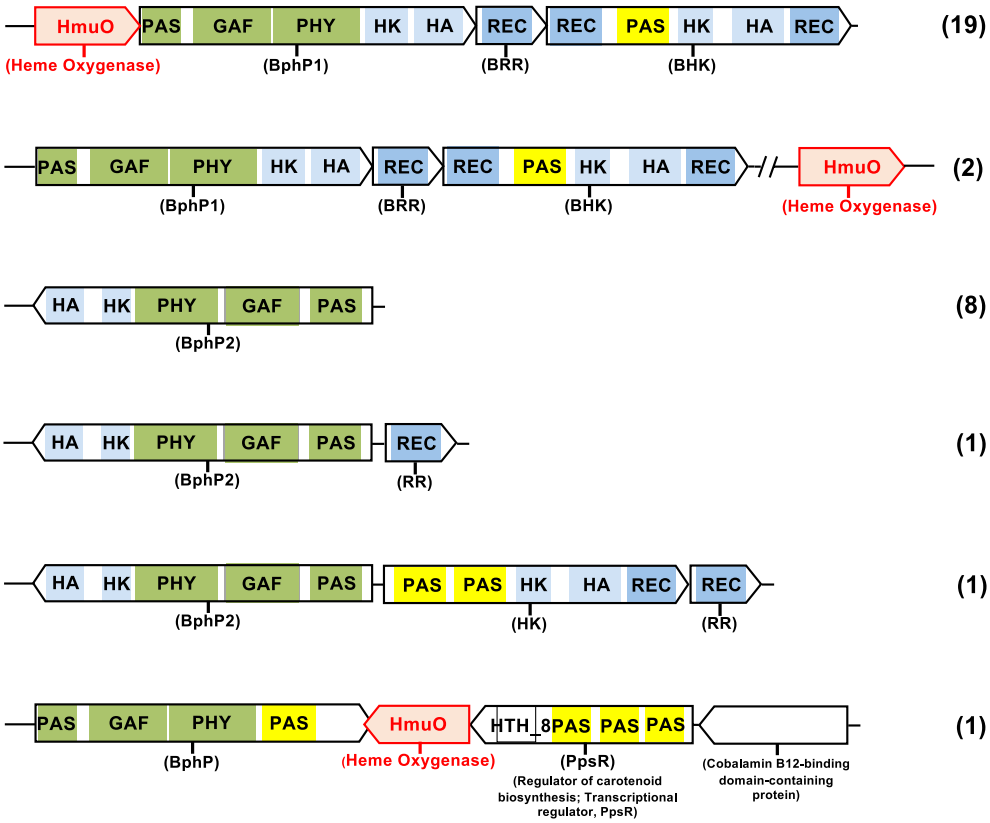

FIGURE 5 | Genetic background and structural organisation of bacteriophytochromes and their cognate regulators found in *Ramlibacter* species. The various BphPs and cognate regulators were compared to those of *R. tataouinensis* TTB310 (*Rt*BphP1 and *Rt*BphP2). The number in brackets indicates the number of strains with the same type of bacteriophytochrome and the same organisation, in order of appearance: (19) nineteen BphP1 with the same operonic structure present in *R. tataouinensis* (around 62% to 70% identity with all BphP1), *R. henchirensis*, *R. ginsenosidimutans*, *R. alkalitolerans*, *R. monticola*, *R. humi*, *R. pinisoli*, *R. algicola*, *R. terrae*, *R. montanisoli*, *R. agri*, *R. cellulosilyticus* USB13, *R. pallidus* HM2, *R. paludis* XY19, *R.* sp. Leaf400, *R.* sp. OAE822, *R.* sp. C_No_T20_B_bin.97, *R.* sp. C_No_T20_A_bin.113 and *R.* sp. C_No_T20_B_bin.133, (2) two BphP1 with *hmuO* outside the operon present in *R. rhizophilus* and *R. aurantiacus* AW1, (8) eight additional isolated BphP2 present in *R. tataouinensis* (33% identity with *Rt*BphP1), *R. humi*, *R. agri*, *R. cellulosilyticus* USB13, *R. pallidus* HM2, *R. paludis* XY19, *R.* sp. OAE822 and *R.* sp. LF-Bin-46, (1) one additional BphP2 present in *R. pinisoli* next to a response regulator, (1) one additional BphP2 present in *R. henchirensis* next to an histidine kinase hybrid and a response regulator, (1) a second and 'atypical' BphP found next to an heme oxygenase and a transcriptional regulator in *R.* sp. LF-Bin-46. The Photosensory Core Domain contains PAS, GAF and PHY subdomains (in green) and a Cys residue located near the N-terminus that covalently binds the biliverdin chromophore. The Histidine Kinase signal transduction domain is located at the C-terminus of the protein. BHK: bacteriophytochrome atypical hybrid His; BRR: bacteriophytochrome Response Regulator; GAF: CGMP-specific phosphodiesterases, Adenylyl cyclases and FhlA domain; HA: HATPase_c Histidine kinase-like ATPase, C-terminal domain; HK: HisKA (histidine kinase A phosphoacceptor) domain; HmuO: heme oxygenase involved in chromophore maturation; PAS: Per-Arnt-Sim domain; PHY: phytochrome domain; REC: CheY-homologous regulator domain.

all organisms) and the variable genome (genes families shared by two or more organisms and strain-specific genes), employing a criterion of 50% identity and 80% alignment coverage for protein conservation. Strain-specific genes ranged from 6% (264 out of 4129 total CDS in *Ramlibacter* sp. Leaf400) to 28% (2141 out of 7520 total CDS in *R. agri*) of the total CoDing Sequences (CDS) with an average of 20% in the analysed subset of 10 genomes (Table S3, https://doi.org/10.5281/zenodo.14761071). Notably, the majority of the 'specific' genes (average 71%) encoded hypothetical or conserved hypothetical proteins, ranging from approximately 52% in *R. lithotrophicus* (423 CDS) to 78% in *R. humi* (981 CDS).

Comparing the genomes of two species from the same semi-desert soil, *R. tataouinensis* and *R. henchirensis*, revealed 2160 shared gene families (2645 and 2778 core CDS, respectively, considering the presence of several orthologs in the same gene family), and 1126 variable or strain-specific gene families for *R. tataouinensis* (1153 CDS) and 1908 for *R. henchirensis* (2012 CDS). *R. tataouinensis* shares 70% of its genes with *R. henchirensis* (66% in synteny), and has 30% strain-specific genes. Conversely, *R. henchirensis*, with about 1000 more genes than *R. tataouinensis* (4790 CDS vs. 3798), shares 58% of its genes with *R. tataouinensis* (54% in synteny), and has 42% strain-specific genes. Interestingly, two strains isolated from a plant leaf from European soil (*Ramlibacter* sp. Leaf400) and North American soil (*Ramlibacter* sp. OAE822) appear to be genetically closer to the North African desert species *R. henchirensis* than *R. tataouinensis* (Figure 3). *Ramlibacter* sp. Leaf400, isolated from *Arabidopsis* sampled in Switzerland, shares 82% of its genes (77% in synteny with *Ramlibacter* sp. OAE822), isolated from an Oklahoma soil (USA) (3391 and 3364 core CDS, respectively) and possesses 727 strain-specific gene families (738 CDS) compared to 1679 (1710 CDS) for *Ramlibacter* sp. OAE822.

Despite geographical distance and different living environments, further analysis revealed that strains within the genus *Ramlibacter* share a significant repertoire of genes, largely contributing to their common adaptive traits in oligotrophic characterised by nutrient scarcity and desiccation. However, the uncertainty surrounding the functions of strain-specific hypothetical proteins limits our understanding of their specific ecological roles.

4.3 | Data-Mining Informed by Bibliographic Analysis: Shared Traits of Adaptation to Desert Environment, Oligotrophy and Light-Sensing

We utilised the 'MicroScope' tool from Genoscope (Vallenet et al. 2020) to analyse the genomes of *Ramlibacter* strains (Figures 4, S4, and S5), completing data obtained from previous publications on *R. tataouinensis* (De Luca et al. 2011), *R. aquaticus* (Props et al. 2019), *R. lithotrophicus* (Yu and Leadbetter 2020), *R. algicola* (Kim et al. 2021) and an uncultured *Ramlibacter* sp. AVDCRST_MAG51 from a Negev desert biological crust (Meier et al. 2021). This analysis focused on potential adaptations to drought and scarcity.

The 43 non-redundant genomes or metagenome-assembled genomes in NCBI databases exhibited a high G% + C% (around 70%) and relatively large genome sizes ranging from 2.3 to 7.2 Mb (average 4.6 Mb) (Figure S6, Tables S2 and S3, https://doi.org/10.5281/zenodo.14761071). This suggests an adaptive strategy favouring gene number extension rather than genome streamlining in these oligotrophic bacteria, as observed in *R. aquaticus* (5.3 Mb), isolated from an oligotrophic freshwater environment (Props et al. 2019).

Two-component systems, crucial for cell cycle regulation and desert life adaptation in *R. tataouinensis* TTB310^T (De Luca et al. 2011), are highly developed across all analysed *Ramlibacter* genomes as revealed by the P2RP tool (Barakat et al. 2013) (Figures 4 and S4). These bacteria possess multiple chemotaxis systems, ranging from 1 to 6, with the aquatic bacterium *R. lithotrophicus* exhibiting the highest number. Notably, certain terrestrial species such as *R. humi* and *R. agri* also demonstrate a high number of chemotaxis systems, possessing 4 CheA proteins each. Among these systems, the chemotaxis system containing the CheA protein Rta_34130 was present in almost all strains and is likely associated with twitching motility involving type IV pili (Figures 4 and S4). While only a few terrestrial strains may possess them, aquatic strains belonging to *R. aquaticus*, *R. lithotrophicus* and *R. algicola* are distinguished by the presence of 'Tight Adherent Pili'. Two of these species, *R. aquaticus* and *R. lithotrophicus*, harbour the *fliC* gene responsible for encoding flagellin, the pivotal component of flagella (Iino et al. 1988). These features collectively contribute to their enhanced adaptation to aquatic habitats, setting them apart from their terrestrial counterparts.

A previous study by Props et al. (2019) noted that the four publicly available *Ramlibacter* genomes in 2019 exhibited an extensive array of genes involved in the transport of a wide range of dissolved organic carbon molecules, especially branched-chain amino acids and oligopeptides (serving as nitrogen-rich organic carbon source). Genes responsible for polyphosphate granules formation (*ppk* and *ppx* genes) were found in 76% of the strains, while transporters of dissolved organic carbon are highly developed in all analysed strain (Figures 4 and S5).

All strains possessed at least one phosphatase and a high-affinity inorganic phosphate transport system, but only a few exhibited additional abilities to solubilise inorganic phosphate or capture phosphonate (Figures 4 and S5). For *Ramlibacter* free-living oligotrophs, which have experienced evolutionary pressures in low-nutrient environments, the development of these regulatory and metabolic networks likely represents an active strategy to combat starvation. These networks enable sensing and extraction of all available nutrients from the environment, allowing the bacteria to capitalise on brief growth opportunities (Bergkessel and Delavaine 2021; Hobbie and Hobbie 2013).

Most strains possess the genes encoding the synthesis and degradation of PHA, facilitating adaptation to nutrient-poor conditions and starvation. Moreover, genes involved in trehalose synthesis and degradation, associated with desiccation tolerance, were present in all strains (De Luca et al. 2011) (Figures 4 and S4). Two operons containing GT2- and GT4-glycosyltransferases (De Luca et al. 2011), likely implicated in exopolysaccharide synthesis (*Rta_10900* to *Rta_10830*) and transport (*Rta_10750* to *Rta_10820*) in *R. tataouinensis* (Jivkova et al. 2022), were conserved in seven other strains exhibiting coccoid forms, as well as in eight additional strains, including the metagenome-assembled genome MAG from a Negev desert biocrust (Figures 4 and S4). A notable correlation emerges between the presence of these two operons and the coccoid morphotype in most cases. However, certain species (*R. algicola*, *R. lithotrophicus*, *R. ginsenosidimutans*), for which the coccoid form was not evidenced, also possess these operons.

Ramlibacter species exhibit various 'Blue Light-sensing Using Flavins' (BLUF) proteins, with the shortest variant (Rta_31060) being the most frequent and universal. This BLUF protein is also present in a MAG from a Negev desert biocrust (Meier et al. 2021) (Figures 4 and S4). Some strains possess one to two additional duplicated BLUF proteins that are slightly longer, although their functionality is not always evident, as observed in *R. tataouinensis* (Figures 4 and S4). The precise role of these blue light receptors remains unidentified; however, it is noteworthy that relatively low-intensity white or blue light illumination strongly inhibits the growth of *R. tataouinensis*, suggesting their involvement in cells division at least in this particular species (De Luca et al. 2019).

The bacteriophytochrome BphP1 (Rta_25470), initially identified in *R. tataouinensis* TTB310^T (De Luca et al. 2011), is relatively 'common' and found in 21 of the 43 non-redundant genomes analysed, with a consistent operonic arrangement, except for the position of the *hmuO* gene encoding a heme oxygenase found isolated in two strains (Figures 4, 5 and S4). Additionally, another bacteriophytochrome analogous to *R. tataouinensis* BphP2 (Rta_28950) is present in 9 of these 21 strains and in *Ramlibacter* sp. LF-Bin-46. It exhibits a similar configuration, either isolated on the genome for 7 strains or located next to a response regulator in *R. pinisoli*, or in proximity to another histidine kinase and a response regulator in *R. henchirensis* (Figure 5). One MAG

(*Ramlibacter* sp. LF-Bin-46) possesses an equivalent BphP2 and an atypical BphP without a histidine kinase domain, located next to a heme oxygenase and a transcriptional regulator (Figure 5). While it is not possible to attribute to the other bacteriophytochromes the same role as those of *R. tataouinensis* in the cell cycle regulation of the other strains, the relatively common presence of light-sensing proteins in *Ramlibacter* suggests that light serves as an environmental signal potentially playing an important role in many species of this genus, including those inhabiting environments other than semi-arid region. Finally, the potential presence of a rudimentary hourglass, encoded by the *kaiC* gene evidenced in *R. tataouinensis*, also appears to be relatively common among most *Ramlibacter* species (Figures 4 and S4). This wide distribution of light-sensing and timekeeping mechanisms among *Ramlibacter* species underlines their importance in the adaptation and survival of these bacteria in diverse environments.

5 | Conclusion

The analysis of approximately 40 genomes deposited in the databases has enabled us to propose a genomic phylogeny of the *Ramlibacter* genus that is consistent with the 16S rRNA phylogeny. However, identifying specific functions related to adaptation to particular environments remains challenging, as a significant portion of the 'strain-specific' genes encode proteins with unknown functions. Despite this limitation, several shared characteristics are observed within the *Ramlibacter* genus. These include the presence of light sensors, highly developed environmental detection networks, systems for capturing organic carbon and phosphate, and the ability to store carbon and energy in polyhydroxyalkanoate or polyphosphate granules. These common characteristics underlie the presence of bacteria of the genus *Ramlibacter* strains in diverse oligotrophic terrestrial and aquatic environments. They account for their ability to adapt to desiccation, prolonged starvation and survival in nutrient-depleted environments. These adaptive properties are further enhanced by their pleiomorphic and encystment abilities in some species, enabling them to endure and persist in challenging conditions. The conservation of these traits across the genus suggests their importance in the evolutionary success of *Ramlibacter* species. The presence of light-sensing proteins in *Ramlibacter* species indicates that light is an important cue for the physiology and adaptation of the non-photosynthetic bacteria.

Future research should focus on elucidating the functions of the numerous hypothetical proteins identified in strain-specific genomic regions. This could provide further insights into the specific adaptations that allow *Ramlibacter* species to thrive in their respective niches. Additionally, experimental studies on the less-characterised strains could help to validate the predicted functions and adaptations inferred from genomic data. In conclusion, this comprehensive genomic analysis of the *Ramlibacter* genus reveals a remarkable set of adaptations that enable these bacteria to thrive in diverse oligotrophic environments.

Author Contributions

Gilles De Luca: writing – original draft, writing – review and editing, conceptualization, investigation, project administration, visualization. **Mohamed Barakat:** writing – review and editing, investigation, visualization. **André Verméglio:** writing – original draft, writing – review and editing. **Wafa Achouak:** writing – review and editing, visualization, funding acquisition. **Thierry Heulin:** writing – original draft, writing – review and editing, conceptualization, supervision, project administration, visualization, funding acquisition.

Acknowledgements

We would like to thank Philippe Ortet and Marie-Ange Palomares (BIAM) for their technical help and helpful discussions and advice.

Disclosure

Figure 1 was reprinted and modified from Delgado-Baquerizo et al., *Science* 2018 (A global atlas of the dominant bacteria found in soil) with permission from AAAS (Licence Number 5857050562261) and the corresponding author Manuel Delgado-Baquerizo. Figure S3 was reprinted from De Luca et al., *PLoS One* 2011 (The cyst-dividing bacterium *Ramlibacter tataouinensis* TTB310 genome reveals a well-stocked toolbox for adaptation to a desert environment) with permission [Creative Commons Attribution 4.0 International (CC BY) licence].

Ethics Statement

The content and authorship of the submitted manuscript have been approved by all authors, and all prevailing local, national and international regulations and conventions, and normal scientific ethical practices, have been respected.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The authors declare that the data supporting the findings of this study are available within the paper and its Supporting Information files (https://doi.org/10.5281/zenodo.14761071). Should any raw data files be needed in another format, they are available from the corresponding author upon reasonable request. Source data are provided with this paper.

References

Akter, S., N. A. M. Kihong, S.-Y. Lee, et al. 2020. "*Ramlibacter pinisoli* sp. nov., a Novel Bacterial Species Isolated From Pine Garden Soil." *International Journal of Systematic and Evolutionary Microbiology* 70: 5841–5847.

Almeida, W. I., R. P. Vieira, A. M. Cardoso, et al. 2009. "Archaeal and Bacterial Communities of Heavy Metal Contaminated Acidic Waters From Zinc Mine Residues in Sepetiba Bay." *Extremophiles* 13: 263–271.

Bai, Y., D. B. Müller, G. Srinivas, et al. 2015. "Functional Overlap of the *Arabidopsis* Leaf and Root Microbiota." *Nature* 528: 364–369.

Baker, A. W., K. A. Satyshur, N. Moreno Morales, and K. T. Forest. 2016. "Arm-In-Arm Response Regulator Dimers Promote Intermolecular Signal Transduction." *Journal of Bacteriology* 198: 1218–1229.

Barakat, M., P. Ortet, and D. E. Whitworth. 2013. "P2RP: A Web-Based Framework for the Identification and Analysis of Regulatory Proteins in Prokaryotic Genomes." *BMC Genomics* 14: 269.

Barfod, K. K., M. Roggenbuck, L. H. Hansen, et al. 2013. "The Murine Lung Microbiome in Relation to the Intestinal and Vaginal Bacterial Communities." *BMC Microbiology* 13: 303.

Barrat, J. A., P. Gillet, C. Lécuyer, S. M. F. Sheppard, and M. Lesourd. 1998. "Formation of Carbonates in the Tatahouine Meteorite." *Science* 280: 412–414.

Barrat, J. A., P. Gillet, M. Lesourd, J. Blichert-Toft, and G. R. Poupeau. 1999. "The Tatahouine Diogenite: Mineralogical and Chemical Effects

of Sixty-Three Years of Terrestrial Residence." *Meteoritics and Planetary Science* 34: 91–97.

Benzerara, K., M. Barakat, N. Menguy, et al. 2004. "Experimental Colonization and Alteration of Orthopyroxene by the Pleomorphic Bacteria *Ramlibacter tataouinensis*." *Geomicrobiology Journal* 21: 341–349.

Benzerara, K., N. Menguy, F. Guyot, et al. 2004. "Biologically Controlled Precipitation of Calcium Phosphate by *Ramlibacter tataouinensis*." *Earth and Planetary Science Letters* 228: 439–449.

Bergkessel, M., and L. Delavaine. 2021. "Diversity in Starvation Survival Strategies and Outcomes Among Heterotrophic Proteobacteria." *Microbial Physiology* 31: 146–162.

Bernard, L., P. A. Maron, C. Mougel, et al. 2009. "Contamination of Soil by Copper Affects the Dynamics, Diversity, and Activity of Soil Bacterial Communities Involved in Wheat Decomposition and Carbon Storage." *Applied and Environmental Microbiology* 75: 7565–7569.

Butera, G., C. Ferraro, G. Alonzo, S. Colazza, and P. Quatrini. 2016. "The Gut Microbiota of the Wood-Feeding Termite Reticulitermes Lucifugus (Isoptera; Rhinotermitidae)." *Annales de Microbiologie* 66: 253–260.

Cáliz, J., G. Montserrat, E. Martí, et al. 2013. "Emerging Resistant Microbiota From an Acidic Soil Exposed to Toxicity of Cr, Cd and Pb Is Mainly Influenced by the Bioavailability of These Metals." *Journal of Soils and Sediments* 13: 413–428.

Caliz, J., G. Montserrat, E. Martí, et al. 2012. "The Exposition of a Calcareous Mediterranean Soil to Toxic Concentrations of Cr, Cd and Pb Produces Changes in the Microbiota Mainly Related to Differential Metal Bioavailability." *Chemosphere* 89: 494–504.

Campanaro, S., L. Treu, L. M. Rodriguez-R, et al. 2020. "New Insights From the Biogas Microbiome by Comprehensive Genome-Resolved Metagenomics of Nearly 1600 Species Originating From Multiple Anaerobic Digesters." *Biotechnology for Biofuels* 13: 25.

Chanal, A., V. Chapon, K. Benzerara, et al. 2006. "The Desert of Tataouine: An Extreme Environment That Hosts a Wide Diversity of Microorganisms and Radiotolerant Bacteria." *Environmental Microbiology* 8: 514–525.

Chan-Yam, K., J. Goordial, C. Greer, A. Davila, C. P. McKay, and L. G. Whyte. 2019. "Microbial Activity and Habitability of an Antarctic Dry Valley Water Track." *Astrobiology* 19: 757–770.

Chaudhary, D. K., and J. Kim. 2017. "*Ramlibacter monticola* sp. nov., Isolated From Forest Soil." *International Journal of Systematic and Evolutionary Microbiology* 67: 4468–4474.

Chaumeil, P. A., A. J. Mussig, P. Hugenholtz, and D. H. Parks. 2019. "GTDB-Tk: A Toolkit to Classify Genomes With the Genome Taxonomy Database." *Bioinformatics* 36: 1925–1927.

Connon, S. A., A. Tovanabootr, M. Dolan, K. Vergin, S. J. Giovannoni, and L. Semprini. 2005. "Bacterial Community Composition Determined by Culture-Independent and -Dependent Methods During Propane-Stimulated Bioremediation in Trichloroethene-Contaminated Groundwater." *Environmental Microbiology* 7: 165–178.

Cupples, A. M., and G. K. Sims. 2007. "Identification of In Situ 2,4-Dichlorophenoxyacetic Acid-Degrading Soil Microorganisms Using DNA-Stable Isotope Probing." *Soil Biology and Biochemistry* 39: 232–238.

Dahal, R. H., J. Kim, D. K. Chaudhary, et al. 2022. "Genome Mining Revealed Polyhydroxybutyrate Biosynthesis by *Ramlibacter agri* sp. nov., Isolated From Agriculture Soil in Korea." *Antonie Van Leeuwenhoek* 115: 563–572.

De Luca, G., M. Barakat, P. Ortet, et al. 2011. "The Cyst-Dividing Bacterium *Ramlibacter tataouinensis* TTB310 Genome Reveals a Well-Stocked Toolbox for Adaptation to a Desert Environment." *PLoS One* 6: e23784.

De Luca, G., S. Fochesato, J. Lavergne, et al. 2019. "Light on the Cell Cycle of the Non-Photosynthetic Bacterium *Ramlibacter tataouinensis*." *Scientific Reports* 9: 16505.

Delgado-Baquerizo, M., A. M. Oliverio, T. E. Brewer, et al. 2018. "A Global Atlas of the Dominant Bacteria Found in Soil." *Science* 359: 320–325.

Deng, Y., C. Deng, J. Yang, B. Li, E. Wang, and H. Yuan. 2018. "Novel Butane-Oxidizing Bacteria and Diversity of *bmoX* Genes in Puguang Gas Field." *Frontiers in Microbiology* 9: 1576.

Gillet, P. h., J. A. Barrat, T. h. Heulin, et al. 2000. "Bacteria in the Tatahouine Meteorite: Nanometric-Scale Life in Rocks." *Earth and Planetary Science Letters* 175: 161–167.

Gommeaux, M., M. Barakat, M. Lesourd, J. Thiéry, and T. Heulin. 2005. "A Morphological Transition in the Pleomorphic Bacterium *Ramlibacter tataouinensis* TTB310." *Research in Microbiology* 156: 1026–1030.

Grönemeyer, J. L., C. S. Burbano, T. Hurek, and B. Reinhold-Hurek. 2012. "Isolation and Characterization of Root-Associated Bacteria From Agricultural Crops in the Kavango Region of Namibia." *Plant and Soil* 356: 67–82.

Gundlapally, S. R., and F. Garcia-Pichel. 2006. "The Community and Phylogenetic Diversity of Biological Soil Crusts in the Colorado Plateau Studied by Molecular Fingerprinting and Intensive Cultivation." *Microbial Ecology* 52: 345–357.

Hegde, S., I. G. Paulino-Lima, R. Kent, L. Kaltenegger, and L. Rothschild. 2015. "Surface Biosignatures of Exo-Earths: Remote Detection of Extraterrestrial Life." *Proceedings of the National Academy of Sciences of the United States of America* 112: 3886–3891.

Heulin, T., M. Barakat, R. Christen, et al. 2003. "*Ramlibacter tataouinensis* gen. nov., sp. nov., and *Ramlibacter henchirensis* sp. nov., Cyst-Producing Bacteria Isolated From Subdesert Soil in Tunisia." *International Journal of Systematic and Evolutionary Microbiology* 53: 589–594.

Heulin, T., G. De Luca, M. Barakat, et al. 2017. "Bacterial Adaptation to Hot and Dry Deserts." In *Adaption of Microbial Life to Environmental Extremes: Novel Research Results and Application*, edited by H. Stan-Lotter and S. Fendrihan, 75–98. Springer International Publishing.

Hickey, W. J., S. Chen, and J. Zhao. 2012. "The Phn Island: A New Genomic Island Encoding Catabolism of Polynuclear Aromatic Hydrocarbons." *Frontiers in Microbiology* 3: 125.

Hobbie, J. E., and E. A. Hobbie. 2013. "Microbes in Nature Are Limited by Carbon and Energy: The Starving-Survival Lifestyle in Soil and Consequences for Estimating Microbial Rates." *Frontiers in Microbiology* 4: 324.

Huang, Y., Y. Deng, J. C.-F. Law, et al. 2021. "Acesulfame Aerobic Biodegradation by Enriched Consortia and *Chelatococcus* spp.: Kinetics, Transformation Products, and Genomic Characterization." *Water Research* 202: 117454.

Huang, Y.-M., D. Straub, A. Kappler, N. Smith, N. Blackwell, and S. Kleindienst. 2021. "A Novel Enrichment Culture Highlights Core Features of Microbial Networks Contributing to Autotrophic Fe(II) Oxidation Coupled to Nitrate Reduction." *Microbial Physiology* 31: 280–295.

Iino, T., Y. Komeda, K. Kutsukake, et al. 1988. "New Unified Nomenclature for the Flagellar Genes of *Escherichia coli* and *Salmonella typhimurium*." *Microbiological Reviews* 52: 533–535.

Iwashita, T., Y. Tanaka, H. Tamaki, et al. 2020. "Comparative Analysis of Microbial Communities in Fronds and Roots of Three Duckweed Species: *Spirodela polyrhiza*, *Lemna minor*, and *Lemna aequinoctialis*." *Microbes and Environments* 35: ME20081.

Jangid, K., M. A. Williams, A. J. Franzluebbers, et al. 2008. "Relative Impacts of Land-Use, Management Intensity and Fertilization Upon Soil Microbial Community Structure in Agricultural Systems." *Soil Biology and Biochemistry* 40: 2843–2853.

Jivkova, D., G. Sathiyanarayanan, M. Harir, et al. 2022. "Production and Characterization of a Novel Exopolysaccharide From *Ramlibacter tataouinensis*." *Molecules* 27: 7172.

Kang, M., G. Chhetri, I. Kim, Y. So, and T. Seo. 2022. "Comparative Genomic Analyses of Four Novel *Ramlibacter* Species and the Cellulose-Degrading Properties of *Ramlibacter cellulosilyticus* sp. nov." *Scientific Reports* 12: 21233.

Khan, S. A., H. M. Kim, J. H. Baek, H. S. Jung, and C. O. Jeon. 2021. "*Ramlibacter terrae* sp. nov. and *Ramlibacter montanisoli* sp. nov., Isolated From Soil." *Journal of Microbiology and Biotechnology* 31: 1210–1217.

Kim, H. M., D. M. Han, B. H. Chun, H. S. Jung, K. H. Kim, and C. O. Jeon. 2021. "*Ramlibacter algicola* sp. nov., Isolated From a Freshwater Alga *Cryptomonas obovoidea*." *International Journal of Systematic and Evolutionary Microbiology* 71: 005010.

Kobayashi, K., and H. Aoyagi. 2019. "Microbial Community Structure Analysis in *Acer palmatum* Bark and Isolation of Novel Bacteria IAD-21 of the Phylum Abditibacteriota (Former Candidate Division FBP)." *PeerJ* 7: e7876.

Lacroix, A. 1931. "Sur La Chute Récente (27 Juin 1931) D'une Météorite Asidérite Dans L'extrême Sud Tunisien." *Comptes Rendus de l'Académie des Sciences* 193: 305–309.

Larke-Mejía, N. L., A. T. Crombie, J. Pratscher, T. J. McGenity, and J. C. Murrell. 2019. "Novel Isoprene-Degrading Proteobacteria From Soil and Leaves Identified by Cultivation and Metagenomics Analysis of Stable Isotope Probing Experiments." *Frontiers in Microbiology* 10: 2700.

Lee, D.-H., and C.-J. Cha. 2017. "*Ramlibacter alkalitolerans* sp. nov., Alkali-Tolerant Bacterium Isolated From Soil of Ginseng." *International Journal of Systematic and Evolutionary Microbiology* 67: 4619–4623.

Lee, H. J., S. H. Lee, S.-S. Lee, et al. 2014. "*Ramlibacter solisilvae* sp. nov., Isolated From Forest Soil, and Emended Description of the Genus *Ramlibacter*." *International Journal of Systematic and Evolutionary Microbiology* 64: 1317–1322.

Lenchi, N., O. Inceoğlu, S. Kebbouche-Gana, et al. 2013. "Diversity of Microbial Communities in Production and Injection Waters of Algerian Oilfields Revealed by 16S rRNA Gene Amplicon 454 Pyrosequencing." *PLoS One* 8: e66588.

Lim, J. E., J. H. Jo, and W. T. Im. 2023. "*Ramlibacter paludis* sp. nov., Isolated From Wetland." *International Journal of Systematic and Evolutionary Microbiology* 73: 005792.

Long, L., H. Zhu, Q. Yao, and Y. Ai. 2008. "Analysis of Bacterial Communities Associated With Spores of *Gigaspora margarita* and *Gigaspora rosea*." *Plant and Soil* 310: 1–9.

Luo, Z., J. Ma, F. Chen, X. Li, and S. Zhang. 2018. "Effects of Pb Smelting on the Soil Bacterial Community Near a Secondary Lead Plant." *International Journal of Environmental Research and Public Health* 15: E1030.

Mehetre, G., M. Shah, S. G. Dastager, and M. S. Dharne. 2018. "Untapped Bacterial Diversity and Metabolic Potential Within Unkeshwar Hot Springs, India." *Archives of Microbiology* 200: 753–770.

Meier, D. V., S. Imminger, O. Gillor, and D. Woebken. 2021. "Distribution of Mixotrophy and Desiccation Survival Mechanisms Across Microbial Genomes in an Arid Biological Soil Crust Community." *mSystems* 6: e00786-20.

Meier-Kolthoff, J. P., J. S. Carbasse, R. L. Peinado-Olarte, and M. Göker. 2022. "TYGS and LPSN: A Database Tandem for Fast and Reliable Genome-Based Classification and Nomenclature of Prokaryotes." *Nucleic Acids Research* 50: D801–D807.

Meier-Kolthoff, J. P., and M. Göker. 2019. "TYGS Is an Automated High-Throughput Platform for State-of-the-Art Genome-Based Taxonomy." *Nature Communications* 10: 2182.

Molina-Menor, E., H. Gimeno-Valero, J. Pascual, J. Peretó, and M. Porcar. 2020. "High Culturable Bacterial Diversity From a European Desert: The Tabernas Desert." *Frontiers in Microbiology* 11: 583120.

Ondov, B. D., T. J. Treangen, P. Melsted, et al. 2016. "Mash: Fast Genome and Metagenome Distance Estimation Using MinHash." *Genome Biology* 17: 132.

Parks, D. H., M. Chuvochina, D. W. Waite, et al. 2018. "Standardized Bacterial Taxonomy Based on Genome Phylogeny Substantially Revises the Tree of Life." *Nature Biotechnology* 36: 996–1004.

Parks, D. H., C. Rinke, M. Chuvochina, et al. 2017. "Recovery of Nearly 8,000 Metagenome-Assembled Genomes Substantially Expands the Tree of Life." *Nature Microbiology* 2: 1533–1542.

Pedron, R., A. Esposito, I. Bianconi, et al. 2019. "Genomic and Metagenomic Insights Into the Microbial Community of a Thermal Spring." *Microbiome* 7: 8.

Price, M. N., P. S. Dehal, and A. P. Arkin. 2009. "FastTree: Computing Large Minimum Evolution Trees With Profiles Instead of a Distance Matrix." *Molecular Biology and Evolution* 26: 1641–1650.

Props, R., P. Monsieurs, P. Vandamme, N. Leys, V. J. Denef, and N. Boon. 2019. "Gene Expansion and Positive Selection as Bacterial Adaptations to Oligotrophic Conditions." *mSphere* 4: e00011-19.

Remenár, M., J. Harichová, M. Zámocký, et al. 2017. "Metagenomics of a Nickel-Resistant Bacterial Community in an Anthropogenic Nickel-Contaminated Soil in Southwest Slovakia." *Biologia* 72: 971–981.

Rutz, B. A., and T. L. Kieft. 2004. "Phylogenetic Characterization of Dwarf Archaea and Bacteria From a Semiarid Soil." *Soil Biology and Biochemistry* 36: 825–833.

Ryu, S. H., D. S. Lee, M. Park, et al. 2008. "*Caenimonas koreensis* gen. nov., sp. nov., Isolated From Activated Sludge." *International Journal of Systematic and Evolutionary Microbiology* 58: 1064–1068.

Sadoff, H. L. 1975. "Encystment and Germination in *Azotobacter vinelandii*." *Bacteriological Reviews* 39: 516–539.

Saghatelyan, A., A. Margaryan, H. Panosyan, and N.-K. Birkeland. 2021. "Microbial Diversity of Terrestrial Geothermal Springs in Armenia and Nagorno-Karabakh: A Review." *Microorganisms* 9: 1473.

Salvador, M., U. Abdulmutalib, J. Gonzalez, et al. 2019. "Microbial Genes for a Circular and Sustainable Bio-PET Economy." *Genes* 10: 373.

Saxena, R., D. B. Dhakan, P. Mittal, et al. 2016. "Metagenomic Analysis of Hot Springs in Central India Reveals Hydrocarbon Degrading Thermophiles and Pathways Essential for Survival in Extreme Environments." *Frontiers in Microbiology* 7: 2123.

Schneider, D., D. Zühlke, A. Poehlein, K. Riedel, and R. Daniel. 2021. "Metagenome-Assembled Genome Sequences From Different Wastewater Treatment Stages in Germany." *Microbiology Resource Announcements* 10: e0050421.

Shrestha, P. M., M. Noll, and W. Liesack. 2007. "Phylogenetic Identity, Growth-Response Time and rRNA Operon Copy Number of Soil Bacteria Indicate Different Stages of Community Succession." *Environmental Microbiology* 9: 2464–2474.

Singleton, D. R., A. C. Adrion, and M. D. Aitken. 2016. "Surfactant-Induced Bacterial Community Changes Correlated With Increased Polycyclic Aromatic Hydrocarbon Degradation in Contaminated Soil." *Applied Microbiology and Biotechnology* 100: 10165–10177.

Skouri-Panet, F., K. Benzerara, J. Cosmidis, et al. 2017. "In Vitro and In Silico Evidence of Phosphatase Diversity in the Biomineralizing Bacterium *Ramlibacter tataouinensis*." *Frontiers in Microbiology* 8: 2592.

Sul, W. J., J. Park, J. F. Quensen, et al. 2009. "DNA-Stable Isotope Probing Integrated With Metagenomics for Retrieval of Biphenyl Dioxygenase Genes From Polychlorinated Biphenyl-Contaminated River Sediment." *Applied and Environmental Microbiology* 75: 5501–5506.

Vallenet, D., A. Calteau, M. Dubois, et al. 2020. "MicroScope: An Integrated Platform for the Annotation and Exploration of Microbial Gene Functions Through Genomic, Pangenomic and Metabolic Comparative Analysis." *Nucleic Acids Research* 48: D579–D589.

Vilardi, K. J., I. Cotto, M. Sevillano, Z. Dai, C. L. Anderson, and A. Pinto. 2022. "Comammox *Nitrospira* Bacteria Outnumber Canonical Nitrifiers Irrespective of Electron Donor Mode and Availability in Biofiltration Systems." *FEMS Microbiology Ecology* 98: fiac032.

Wang, L., D.-S. An, S.-G. Kim, et al. 2012. "Ramlibacter Ginsenosidimutans sp. nov., With Ginsenoside-Converting Activity." *Journal of Microbiology and Biotechnology* 22: 311–315.

Wang, W., Y. Ma, X. Ma, et al. 2012. "Diversity and Seasonal Dynamics of Airborne Bacteria in the Mogao Grottoes, Dunhuang, China." *Aerobiologia* 28: 27–38.

Xie, S., W. Sun, C. Luo, and A. M. Cupples. 2010. "Stable Isotope Probing Identifies Novel m-Xylene Degraders in Soil Microcosms From Contaminated and Uncontaminated Sites." *Water, Air, and Soil Pollution* 212: 113–122.

Xu, J., M. Sheng, Z. Yang, et al. 2020. "*Caenimonas sedimenti* sp. nov., Isolated From Sediment of the Wastewater Outlet of an Agricultural Chemical Plant." *Current Microbiology* 77: 3767–3772.

Yan, Z.-F., H. Trinh, G. Moya, et al. 2017. "*Ramlibacter rhizophilus* sp. nov., Isolated From Rhizosphere Soil of National Flower Mugunghwa From South Korea." *International Journal of Systematic and Evolutionary Microbiology* 67: 3773–3777.

Yu, H., and J. R. Leadbetter. 2020. "Bacterial Chemolithoautotrophy via Manganese Oxidation." *Nature* 583: 453–458.

Zhang, X.-J., G.-D. Feng, Q. Yao, Y.-H. Wang, S.-Z. Yang, and H.-H. Zhu. 2019. "*Ramlibacter humi* sp. nov., Isolated From Tropical Forest Soil." *International Journal of Systematic and Evolutionary Microbiology* 69: 3460–3464.

Zhou, Z., W. Qiao, C. Xing, X. Shen, D. Hu, and L. Wang. 2014. "A Micro-Aerobic Hydrolysis Process for Sludge In Situ Reduction: Performance and Microbial Community Structure." *Bioresource Technology* 173: 452–456.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.