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Research Article

Genomic characterization of *Pseudomonas wenzhouensis* A.M.S.S. isolated from oil-contaminated soil and its metabolic potential for bioremediation

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Abstract
Petroleum-contaminated soil is useful for environmental bioremediation studies because it contains many different types of microorganisms that can hydrolyze hydrocarbons and degrade sulfur-contaminated environments. This study included the isolation of *Pseudomonas wenzhouensis* A.M.S.S. from oil-contaminated soil in Mosul, Iraq; its taxonomic status and potential use were determined through extensive whole-genome sequencing. Whole-genome sequencing, Genome Assembly and Annotation, *16S rRNA* gene phylogenetic tree, and in silico DNA-DNA Hybridization and Genetic analysis were performed on the isolate to assess its ability to degrade hydrocarbon and sulfur compounds. Genomic analysis showed that the GC content was 61.9 percent, 4.228 projected protein-coding sequences, and 50 *tRNA* genes, and the 4.65 Mb draft genome produced by de novo assembly demonstrated significant metabolic potential. A.M.S.S. is a unique strain and possibly a new species within the genus, according to anin silico DNA–DNA hybridization value of 60.9%, which is below the 70% species criterion, even though phylogenomic analysis (TYGS) placed the isolate closest to *Pseudomonas wenzhouensis* A20. A vast gene repertoire for co factor biosynthesis, stress adaptation, and the metabolism of amino acids and carbohydrates was identified through subsystem annotation. Notably, the genome encodes several aromatic compound degradation pathways, and reduction of inorganic sulfate, supporting its ability to catabolize pollutants derived from petroleum and indicating a genomic potential for the breakdown of contaminants produced from petroleum. The study provides important clues about the ecological breadth of this strain and points to its possible use in environmental biotechnology, specifically for the bioremediation of polluted ecosystems.

Keywords: Aromatic compound degradation, Comparative genomics, Genomic prediction of bioremediation, Sulfur metabolism

INTRODUCTION

The genus Pseudomonas, one of the most common and diverse groupings of bacteria, was first described by Professor Migula in 1894. According to Peix *et al*. (2009), it is present in polluted plants, human clinical specimens, and a range of natural environments. This genus contains rod-shaped, aerobic, motile, spore-free Gram-negative bacteria. Usually, they have one or more polar flagella to aid in their mobility (Palleroni, 2015). With a high degree of genetic and metabolic diversity, the genus Pseudomonas occupies a wide range of ecological niches, including soil, water, and the tissues of animals and plants (Peix *et al*., 2009; Palleroni, 2015). Pseudomonas has been considered one of the largest bacterial genera, with legal names published for over 300 species (Parte *et al*., 2020) and over 396 species, including 21 subspecies, registered in the List of Prokaryotic Names with Standing in Nomenclature (Parte, 2014). The Pseudomonas genus contains several species, including *Pseudomonas stutzeri, Pseudomonas fluorescens, Pseudomonas aeruginosa, Pseudomonas entomophila, and Pseudomonas putida*, each with important ecological and economic importance and frequently used in bioremediation (Rossi *et al*., 2021).

The *Pseudomonas* genus shows great diversity in both gene content and environmental adaptation strategies,

according to comparative genomic research. As an illustration of direct genomic comparison for understanding environmental specialization, extensive investigations of thousands of Pseudomonas genomes have shown genetic characteristics linked to specific ecological niches (Saati-Santamaría *et al*., 2022). studies on certain strains relevant to bioremediation, such as *P. pseudoalcaligenes* and *P. putida* LS46, have shown that comparative genomic analysis can differentiate between core and accessory genes associated with pollutant-degrading pathways among closely related strains (Safari *et al*., 2019).

The evolution of whole-genome sequencing has now enabled researchers to build phylogenetic trees based on the most comprehensive sets of genes and proteins. These kinds of genomic-scale sequence data further support the discovery of conserved signature indels (CSIs), which are distinguishing characteristics shared by a number of monophyletic clades of organisms (Parks *et al*., 2018).

According to Gupta *et al*. (2020), molecular delimitation of species clades and taxa can be accurately achieved because multiple distinct molecular synapomorphies exist. A new methodology for grouping members of highly polyphyletic taxa, for instance Bacillus, has been developed by combining them with phylogenomic analyses.

More than 300 Pseudomonas species are now available in the NCBI Genome Database, each with its nucleotide sequence (Sayers *et al*., 2019). In this study, *P. wenzhouensis* was obtained from oil-contaminated soil and identified using *16S rRNA* gene sequencing. A whole-genome analysis of *P. wenzhouensis* A.M.S.S. was then conducted comprehensively to identify, classify, and characterize genes involved in important biological processes.

MATERIALS AND METHODS

Process and collection of samples

Soil samples contaminated with petroleum products were collected at three sites around an electricity generator in Mosul city, and each sample was placed in a sterile container. In the laboratory 1 g of each soil sample mixed with 9 ml sterile distilled water and then subsequently, serial dilutions were performed: 0.1 ml of dilution 10^3, 10^4 and 10^5 were replaced in the Nutrient agar plates and incubated at 37 ^oC temperatures in a span of 24-48 hrs, Al-Sanjary and Burhan (2025).

Genomic DNA extraction and purification

The bacterial isolates' DNA was extracted using the Geneaid Genomic DNA Purification kit. Analysis by a Nanodrop spectrophotometer was performed to determine the quantity and purity of DNA based on the absorbance ratio at 280/260 nm.

Genome submissions to NCBI GenBank

The *Pseudomonas wenzhouensis* A.M.S.S. genome sequence has been submitted to DDBJ/ENA/GenBank (accession no. JAUDTJ000000000).

Genome assembly and annotation

The raw reads were *de novo* assembled into contigs using the SPAdes 3.5 bioinformatics tool (Bankevich *et al*., 2012) with k-mer lengths of 21, 33, 55, and 77. QUAST software (Gurevich *et al*., 2013) was used to generate assembly statistics. The assembled genome was annotated using the RAST server (Aziz *et al*., 2008). The SEED tool (Overbeek *et al*., 2014) was used for predicting functional genes in subsystem categories.

Whole genome based phylogenetic tree

The Type Strain Genome Server (TYGS) was used to build a phylogenetic tree of *P. wenzhouensis* A.M.S.S. and its closest relative based on complete genomes (Meier-Kolthoff and Gopfert, 2019). The default loading of the whole genome file in the fasta format was loaded to the server. The phylogeny itself was inferred by FastME 2.0 (Lefort *et al*., 2015), which was included in the TYGS.

In silico DNA-DNA (*is*DDH) hybridization analysis

The GGDH bioinformatics tool was used in the study by Meier-Kolthoff *et al*. (2022) to determine isDDH values indicating *P. wenzhouensis* A.M.S.S. relative to its closest strains (whole-genome sequences).

16S rRNA gene phylogenetic tree analysis

The Nucleotide Basic Local Alignment Search Tool (BLASTn) program was used to query the *P. wenzhouensis* A.M.S.S. sequence for homology with sequences available in the NCBI GenBank database. The bootstrap (100X) analysis carried out using MEGA-11 software, allowed the reconstruction of the phylogenetic tree (Tamura *et al*., 2021).

Circular genome map

The genome map of *Pseudomonas wenzhouensis* A.M.S.S in a circular form was made with the help of the CGView Comparison Tool (Stothard *et al*., 2019).

Genome comparisons

The BLAST Ring Image Generator (BRIG) software (Alikhan *et al*., 2011) was used to align *P.wenzhouensis* A.M.S.S. with the most closely related Pseudomonas species to generate an image showing the differences and similarities between the *P. wenzhouensis* A.M.S.S. genome sequence and other se-

Table 1. An overview of the general genome characteristics of *Pseudomonas wenzhouensis* A.M.S.S. was prepared with the help of QUAST software and RAST server

Feature	Value
Genome total length (bp)	4,658,189
Number of contigs	210
Largest contig (bp)	173,402
Smallest contig (bp)	1036
GC content (%)	61.92
Total of protein-coding sequences (CDSs)	4,228
Number of tRNA genes	50
N50	52,065

quences as a set of concentric rings.

RESULTS AND DISCUSSION

Psuedomonas wenzhouensis possesses several useful properties.The general genomic characteristics provided by QUAST software and RAST server of A.M.S.S. strain are indicated in Table 1. Genome is comprised of 61.92 percent of GC. The genome was in an order of magnitude ; its genomic length is estimated to be approximately 4.65 million base pairs. This genome is rather complex, owing to its 210 contigs. This strain had 4.228 protein-coding sequences with 50 *tRNA* genes , highlighting the genetic potential of the strain further and potentially hinting at a high propensity of protein synthesis. The last indicator of a high-quality assembly is the N50 value of 52.065, which displays longer contigs.

More importantly, the analysis is enhanced by Fig. 1, which presents a pie chart of RAST server data, dividing the subsystem proteins (green bars) from the non-subsystem proteins (blue bars). The complete composition of a genome in the context of an organism's overall metabolism and physical nature, forms the entire picture, as this analysis provides the fraction of the total genome dedicated to a specific metabolic process.

Although pairwise genome comparisons (isDDH) with other Pseudomonas species ranged from 39.7% to 53.1%, *P. wenzhouensis* A.M.S.S. and *P. wenzhouensis* A20 exhibited the highest values (60.9%) (Table 2). Compared with other related species, *P. wenzhouensis* A.M.S.S. had a smaller genome, measuring 4.452.100 bp and encoding 4.068 proteins. Its G+C content was 62.2%, and strain's δ-values, (indicate the level of genetic distance between strains) varied from 0.21 to 0.33 (Meier-Kolthoff *et al*., 2013). Based on these genomic results, it has a unique taxonomic position and is most closely related to *P. wenzhouensis* A20.

Based on Fig. 2, bootstrap value of 100 % implies a high genetic similarity between *P.wenzhouensis* A.M.S.S. and *P. wenzhouensis* A20.The phylogenetic tree also demonstrates the differences between this isolate and other Pseudomonas species, including *P. mendocina* and *P. hydrolytica.*

Table 3. Several genes involved in sulfur metabolism are found in the genome of *P. wenzhouensis* A.M.S.S. The several genes involved in inorganic sulfur assimilation include sulfate transporters (*CysT*, *CysW*, *CysA*, and *CysP*), sulfite reductase, ferredoxin, sulfate adenylyltransferase, adenylylsulfate kinase, and related oxidoreductases. These include thioredoxin-disulfide reductases, including thiol peroxidases and alkyl hydroperoxide reductase. This suggests the strain has the genetic capacity to utilize and reduce sulfur.

Table 4. Several genes linked to the possible breakdown of chemicals generated from oil are encoded in the genome of *P. wenzhouensis* A.M.S.S., since its genome includes essential genes involved in the breakdown of aromatic hydrocarbons. A number of enzymes are found to play significant roles in the bacterial catechol degradation pathway, especially 4β-ketoadipate enol-lactone hydrolase, which catalyzes the homogentisate pathway, along with fumarylacetoacetase, homogentisate 1,2-dioxygenase, and maleylacetoacetate isomerase. Pathogenic strains also contain biphenyl-2,3-diol dioxygenase and a set of enzymes that can catalyze the breakdown of n-phenylalkanoic acid, namely enoyl-CoA hydratase and 3-ketoacyl-CoA thiolase. Togetherthese enzymes increase the organism's ability to metabolize complex aromatic compounds of petroleum origin (Arias-Barrau *et al*., 2004).

Fig. 3 shows a circular genome map of *P. wenzhouensis* A.M.S.S., derived using the CG View Comparison Tool. The centermost ring is used to give a guide of chromosomal position in the form of genomic co-ordinates. It is encircled in a sequence of concentric rings, each marked with a certain genetic characteristic. Each functional category is assigned a distinct hue to easily identify RNA genes, protein-coding genes, and other genomic regions. Such representation not only helps us understand gene distribution and gene functional clustering along the circular genome but also provides a better understanding of the genome's structural architecture.

Using the BRIG software, Fig. 4 shows a comparative circular genome map of *P. wenzhouensis* A.M.S.S. coupled with five closely related Pseudomonas species. Following rings that indicate GC content and GC skew, the innermost gray ring represents the reference genome of *P. wenzhouensis* A.M.S.S. Other species' genomes are shown by coloured rings outside, where unique parts are represented by gaps and shared are-

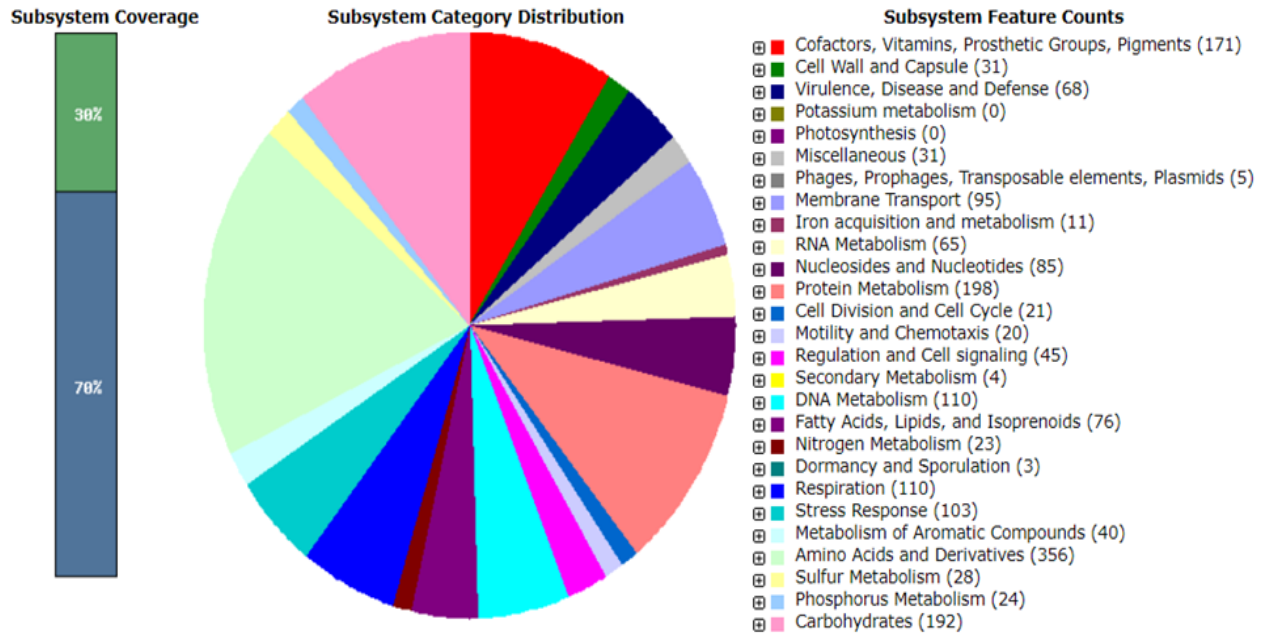

Fig. 1. *Statistics of the category distribution of subsystems of Pseudomonas wenzhouensis A.M.S.S. Analysis of genome was achieved using Rapid Annotation System Technology (RAST) server. There were numerical counts of each subsystem feature which was extracted using the SEED viewer with the pie chart displaying the subsystem coverage. The green bar on the subsystem coverage indicates the percentage of proteins present in the subsystems whereas blue bar indicates the percentage of proteins not present in the subsystems*

Table 2. Genome comparison of Pseudomonas wenzhouensis A.M.S.S. genome – vs. type strain genomes using isDDH, GC content, δ- value, genome size and number of proteins

***P. wenzhouensis A.M.S.S.* vs. type strain genome**	**Digital *is*DDH value (%)**	**Percent G+C (%)**	**δ- value**	**Genome Size (pb)**	**Number of proteins**
Pseudomonas wenzhouensis A20	60.9	62.2	0.25	4,452,100	4068
Pseudomonas pseudoalcaligenes NBRC 14167	53.1	62.25	0.27	4,702,414	4507
Pseudomonas indoloxydans JCM 14246	52.5	62.25	0.26	5,198,577	4666
Pseudomonas chengduensis DSM 26382	44.4	62.32	0.21	5,412,239	5096
Pseudomonas sihuiensis KCTC 32246T	45.4	62.54	0.22	5,476,267	5044
Pseudomonas toyotomiensis JCM 15604	44.6	62.61	0.26	5,489,804	5160
Pseudomonas alcaliphila JCM 10630	43.1	62.88	0.33	5,282,641	4874
Pseudomonas sediminis PI11	44.9	62.48	0.32	4,878,472	4443
Pseudomonas mendocina NBRC 14162	43.1	62.83	0.28	5,124,692	4711
Pseudomonas guguanensis JCM 18416	45.5	64.21	0.23	5,077,861	4577
Pseudomonas hydrolytica DSWY01	41.4	64.47	0.22	5,387,415	2702
Pseudomonas khazarica TBZ2T	39.7	64.96	0.24	5,200,365	4806

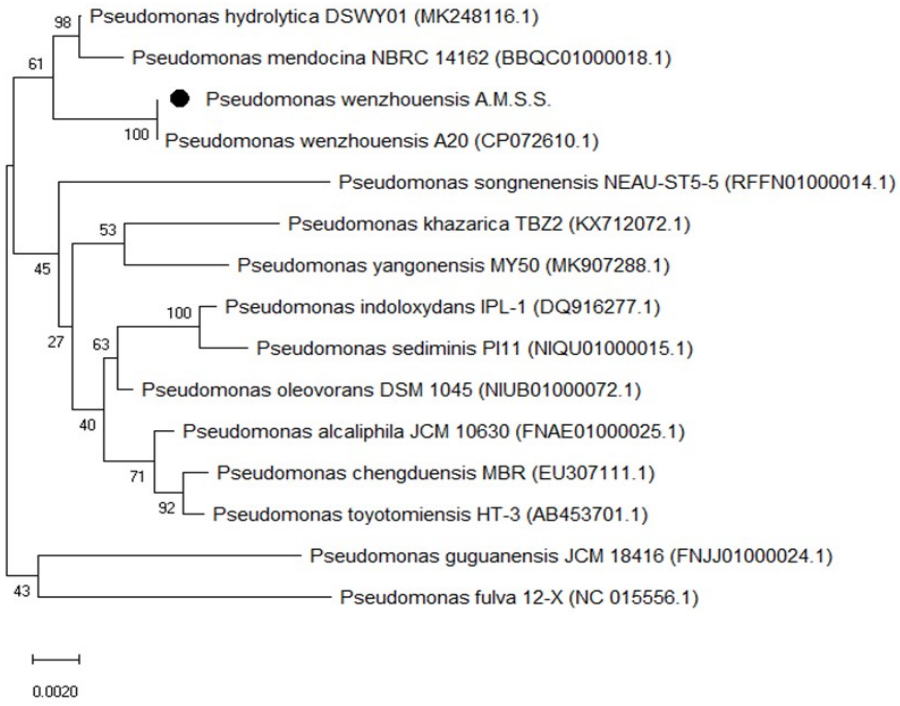

Fig. 2. *Neighbor-joining phylogenetic trees with 16S rRNA sequences of Pseudomonas wenzhouensis A.M.S.S. (numbered in black circle) and also depicting their related closely related strains using MEGA-11 software with the scale length being 0.002. The proportion of replicate trees where the corresponding strains grouped together, in the bootstrap test (100 replicates) are tabulated beside the branches.*

as by shared colours. Both strain-specific and conserved genomic areas are highlighted in this Fig. to illustrate the genetic similarities and differences between *P. wenzhouensis* A.M.S.S. and its closest relatives.

The present study provides a complete genomic characterization of *P. wenzhouensis* A.M.S.S., describing its genetic material, metabolic capabilities, and potential ecological habitat. Combining *16S rRNA* gene sequencing with whole-genome analysis through an integrative strategy enabled accurate taxonomic identification and revealed genomic features suggesting potential metabolic versatility. The genome includes 50 *tRNA* genes and 4.228 protein-coding genes is 4.65 Mb, and has a GC content of 61.92 %. This structure, as demonstrated in a previous work by Zhang *et al*. (2021), is indicative of that of GC-rich, highly metabolically versatile Pseudomonas genomes.

RAST subsystem annotation identified highly interconnected sets of genes related to amino acid metabolism, cofactor biosynthesis, and glucose consumption , reflecting the strain's high metabolic flexibility. Such flexibility is presumably beneficial for survival and adaptation under nutrient-limited or environmentally stressed conditions, including polluted environments. Additionally, the presence of genes committed to vitamin and pigment biosynthesis indicates important ecological functions, notably for protection against oxidative stresses and mediation of microbial consortium interactions. These observations are in agreement with the reports of de Sousa *et al*. (2021).

In silico DNA-DNA hybridization (isDDH) value between *P. wenzhouensis* A.M.S.S. and *P. wenzhouensis* A20 is approximately 60.9%, which is below the standard 70% threshold used to distinguish bacterial species (Meier-Kolthoff *et al.* 2013).The strains A.M.S.S. and A20 cluster closely related, according to whole-genome-based phylogenetic analysis using TYGS; however, there are some differences in genome size, δ-values, and the number of coding sequences. Existing taxonomic standards for the genus *Pseudomonas*, the strain is currently kept as *P. wenzhouensis* A.M.S.S. based on these findings. However, additional experimental and phenotyp research is needed to confirm the possibility of a new species.

Moreover, a *16S rRNA* phylogenetic comparison revealed high sequence similarity (>98.7%) among several Pseudomonas species, namely *P. hydrolytica, P. oleovorans*, and *P. mendocina*. Nonetheless, the observation underscores the importance of avoiding reliance on *16S rRNA* alone or species delimitation in bacterial taxonomy, given the observed inconsistency between *16S rRNA* similarity and isDDH values (Chun *et al*., 2018). Whole-genome-based methods such as average nucleotide identity (ANI) and in silico DNA-DNA hybridization (isDDH) have become more powerful and

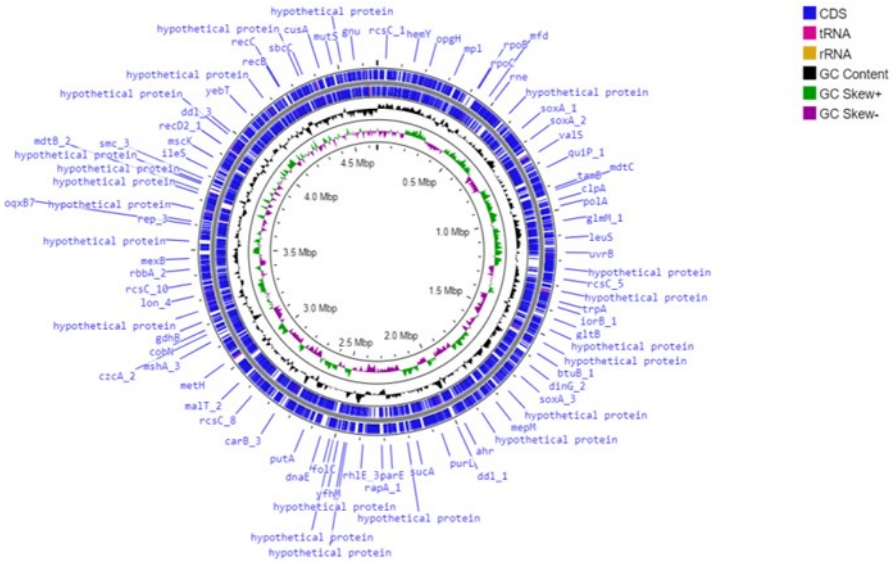

Fig. 3. *Circular genome map of Pseudomonas wenzhouensis A.M.S.S. The innermost ring represents the chromosome position. From the outer to the inner rings, the colored tracks represent coding sequences (CDSs) on the forward strand, CDSs on the reverse strand, tRNA genes, rRNA genes, GC content, and GC skew*

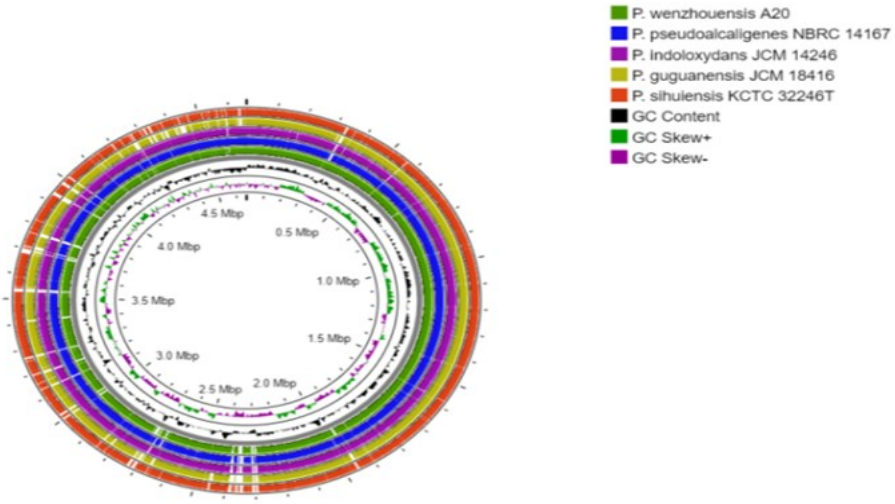

Fig. 4. *Comparative circular genome map of Pseudomonas wenzhouensis A.M.S.S. and five closely related Pseudomonas species. The innermost dark gray circle represents the reference genome of Pseudomonas wenzhouensis A.M.S.S. GC content (black) and GC skew (green/purple) are shown in the inner rings. The outer colored rings represent the genomes of the related Pseudomonas species. Regions lacking color indicate genomic regions that are either absent or highly divergent in the compared genomes relative to the reference genome*

reliable for species-level taxonomic relationships (Richter and Rosselló-Móra, 2009).

Genome analysis of *P. wenzhouensis* A.M.S.S. identified a set of genes related to sulfur metabolism, specifically genes involved in inorganic sulfate reduction. It is noteworthy that genes for the sulfate transport system proteins (*cysT, cysA, cysW*, and *cysP*) were found along with important reductive enzymes like phosphoadenylyl -sulfate reductase and sulfite reductase.

The presence of several stress response and resistance genes in the genome of *P. wenzhouensis* A.M.S.S. supports its ecological resilience. For instance, the strain is probably able to endure and proliferate in sulfur-richhydrocarbon-contaminated environments because of genes involved in hydrocarbon breakdown, oxidative stress response, and sulfur metabolism. Furthermore, the diversity of metabolic pathways indicates genetic flexibility, suggesting the ability to adapt to changing environmental conditions. The idea that these genetic characteristics contribute to the strain's environmental robustness is supported by Yanagita *et al*. (2025), who found comparable correlations between stress response genes and ecological resilience in other Pseudomonas strains.

This conclusion is consistent with a recent publication using a sulfur-based autotrophic denitrification filter

Table 3. Genes associated with sulfur metabolism in *Pseudomonas wenzhouensis* A.M.S.S.

Category	Subcategory	Subsystem	Role
Sulfur metabolism	Sulfur metabolism - no subcategory	Thioredoxin-disulfide reductase	Thioredoxin reductase (EC 1.8.1.9)
		Thioredoxin-disulfide reductase	Thiol peroxidase, Bcp-type (EC 1.11.1.15)
		Thioredoxin-disulfide reductase	Thiol peroxidase, Tpx-type (EC 1.11.1.15)
		Thioredoxin-disulfide reductase	Alkyl hydroperoxide reductase subunit C-like protein
	Inorganic sulfur assimilation	Inorganic sulfur assimilation	Sulfate transport system permease protein CysT
		Inorganic sulfur assimilation	Oxidoreductase probably involved in sulfite reduction
		Inorganic sulfur assimilation	Phosphoadenylyl-sulfate reductase [thioredoxin] (EC 1.8.4.8)
		Inorganic sulfur assimilation	Putative sulfate permease
		Inorganic sulfur assimilation	Sulfate adenylyltransferase subunit 1 (EC 2.7.7.4)
		Inorganic sulfur assimilation	Adenylylsulfate kinase (EC 2.7.1.25)
		Inorganic sulfur assimilation	Sulfate permease, Trk-type
		Inorganic sulfur assimilation	Sulfite reductase [NADPH] hemoprotein beta-component (EC 1.8.1.2)
		Inorganic sulfur assimilation	Sulfate adenylyltransferase subunit 2 (EC 2.7.7.4)
		Inorganic sulfur assimilation	Ferredoxin
		Inorganic sulfur assimilation	Sulfate and thiosulfate import ATP-binding protein CysA (EC 3.6.3.25)
		Inorganic sulfur assimilation	Ferredoxin--NADP(+) reductase (EC 1.18.1.2)
		Inorganic sulfur assimilation	Sulfate and thiosulfate binding protein CysP
		Inorganic sulfur assimilation	Sulfate transport system permease protein CysW
		Inorganic sulfur assimilation	3'(2'),5'-bisphosphate nucleotidase (EC 3.1.3.7)

(SADF), which supported the conclusion that high sulfate salinity enriched microbial populations with genes for nitrogen and sulfur metabolism, with indications of functional cross-interplay between the pathways (Chen *et al*., 2025). Overall, the present findings, supported by Chen *et al*. (2025), indicate the strain's possible adaptability to salinized or polluted environments, thereby affirming its utility for environmental applications.

The *P. wenzhouensis* A.M.S.S. genome contains several gene clusters for aromatic compound degradation pathways. Among these genes, the homogentisate pathway is of significant value, involving essential enzymes like maleylacetoacetate isomerase and homogentisate 1,2-dioxygenase that play a role in aromatic amino acid catabolism. Apart from this, the catechol branch of the β-ketoadipate pathway was identified allowing for efficient catechol degradation, a key intermediate in aromatic hydrocarbon catabolism. Peripheral catabolic pathways targeting biphenyl, benzoate, n-phenylalkanoic acid, and quinate were also discernedpoint to a genetic potential for the breakdown of a variety of petroleum pyrolysis products. (Hossain *et al*., 2024). Apart from detoxification, these pathways supply important intermediates for major energy-producing processes, thereby relating pollutant catabolism to cell energetics. Moreover, recent investigations indicate that microorganisms not only degrade non-halogenated aromatic hydrocarbons but also disintegrate aromatic halogenated molecules, which represent some of the most persistent environmental pollutants (Pimviriyakul *et al*., 2020). The identified genomic repertoire indicated a broad catabolic potential in *P. wenzhouensis* A.M.S.S., which may be relevant to future

Table 4. Gene involved in aromatic compound metabolism in *Pseudomonas wenzhouensis* A.M.S.S.

Category	Subcategory	subsystem	Role
Metabolism of aromatic compounds	Metabolism of Aromatic Compounds–no subcategory	Gentisate degradation	Maleylacetoacetate isomerase (EC 5.2.1.2)
	Metabolism of central aromatic intermediates	Homogentisate pathway of aromatic compound degradation	Aromatic-amino-acid aminotransferase (EC 2.6.1.57)
		Homogentisate pathway of aromatic compound degradation	Homogentisate 1,2-dioxygenase (EC 1.13.11.5)
		Homogentisate pathway of aromatic compound degradation	4-hydroxyphenylpyruvate dioxygenase (EC 1.13.11.27)
		Homogentisate pathway of aromatic compound degradation	Maleylacetoacetate isomerase (EC 5.2.1.2)
		Homogentisate pathway of aromatic compound degradation	Transcriptional regulator, IclR family
		Homogentisate pathway of aromatic compound degradation	Fumarylacetoacetase (EC 3.7.1.2)
		Catechol branch of beta-ketoadipate pathway	Succinyl-CoA:3-ketoacid-coenzyme A transferase subunit B (EC 2.8.3.5)
		Catechol branch of beta-ketoadipate pathway	Succinyl-CoA:3-ketoacid-coenzyme A transferase subunit A (EC 2.8.3.5)
		Catechol branch of beta-ketoadipate pathway	Beta-ketoadipate enol-lactone hydrolase (EC 3.1.1.24)
	Peripheral pathways for catabolism of aromatic compounds	Biphenyl Degradation	4-hydroxy-2-oxovalerate aldolase (EC 4.1.3.39)
		Biphenyl Degradation	biphenyl-2,3-diol 1,2-dioxygenase III-related protein
		Biphenyl Degradation	Acetaldehyde dehydrogenase, acetylating, (EC 1.2.1.10)
		Benzoate degradation	Benzoate transport protein
		n-Phenylalkanoic acid degradation	3-hydroxybutyryl-CoA epimerase (EC 5.1.2.3)
		n-Phenylalkanoic acid degradation	enoyl-CoA hydratase, R-specific
		n-Phenylalkanoic acid degradation	3-hydroxyacyl-CoA dehydrogenase (EC 1.1.1.35)
		n-Phenylalkanoic acid degradation	Long-chain-fatty-acid--CoA ligase (EC 6.2.1.3)
		n-Phenylalkanoic acid degradation	Enoyl-CoA hydratase (EC 4.2.1.17)
		n-Phenylalkanoic acid degradation	Delta(3)-cis-delta(2)-trans-enoyl-CoA isomerase (EC 5.3.3.8)
		n-Phenylalkanoic acid degradation	3-ketoacyl-CoA thiolase (EC 2.3.1.16)
		Quinate degradation	3-dehydroquinate dehydratase II (EC 4.2.1.10)

investigations in engineered bioremediation systems.

The circular map of the genome identified operon-like gene arrangements related to aromatic compound and sulfur metabolism. These gene clusters may represent coordinated functional activities, and such fine tuning is likely to improve the bacterium's capacity to cope with unfavourable environmental conditions. However, this conclusion is based on genetic arrangement rather than direct functional data.

Comparative genomic analysis using BRIG also identified strain-specific genomic regions in *P. wenzhouensis* A.M.S.S. that were absent in closely related *Pseudomonas* species. Horizontal gene transfer (is a key mechanism enabling bacteria to have adaptive traits in polluted environments). In *P. wenzhouensis* A.M.S.S., several strain-specific genomic regions predicted to be acquired via HGT encode putative resistance and catabolic functions. These acquired genes may enhance tolerance to oxidative stress including hydrocarbon toxicity, thereby contributing to ecological resilience in oil-contaminated sites. Similar associations between HGT-acquired functions and adaptation to hydrocarbon-polluted environments have been reported in other microbial systems (Das and Chandran, 2021).

These observations validate earlier studies highlighting at the acquisition of genomic islands, along with mobile

genetic elements, is an important regulatory mechanism that provides the spectacular adaptability of Pseudomonas species occupying various ecological niches (Espinosa-Camacho *et al*., 2022).

The study as a whole establishes the evolutionary and ecological adaptability of *P. wenzhouensis* A.M.S.S., which possesses a metabolically broad repertoire, environmental tolerance and adaptation strategies, and prospects for bioremediation of sulfur- and hydrocarbon -contaminated soil sites. The results also highlight the significance of whole-genome sequencing of a species in proper delicacy and functional characterization in taxa of complex microbes.

Conclusion

The complete genome sequence of *P. wenzhouensis* A.M.S.S. indicated that it belonged to a distinct genomic lineage within the genus Pseudomonas, supporting its classification as a potential new species. The genome contained diverse metabolic pathways, including those involved in the degradation of aromatic compounds, sulfur uptake, and the flexible metabolism of amino acids and carbohydrates. Comparative genomic analyses revealed unique genomic regions may be acquired through horizontal gene transfer, which may contribute to its adaptability in contaminated or environmentally challenging environments. Based on these genomic predictions, future experimental studies are needed, including growth assays with selected aromatic hydrocarbons and alternative sulfur sources to evaluate metabolic functions, as well as transcriptomic analyses under pollution or stress conditions to study the regulation of stress resistance and catabolic pathways. The studies will provide experimental verification of the predicted functional abilities and further illustrate the ecological and biotechnological significance of this strain.

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Conflict of interest

The authors declare that they have no conflict of interest.

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