



Data Article

Draft genome sequence of an ubiquinone-10 producing *Methylobacterium durans* LRY1-08 isolated from lichen in Thailand



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ABSTRACT

A ubiquitous and pink-pigmented facultatively methylophilic bacterium, designated LRY1-08 (=JCM 33120), was isolated from a lichen in Thailand. Strain LRY1-08 and *Methylobacterium durans* NBRC 112876^T shared 99.92% similarity based on the 16S rRNA gene sequence. The draft genome of LRY1-08 was 5.26 Mbp with 4,952 protein-coding sequences and an average G + C content of 70.0 mol%. Comparing strain LRY1-08 to *M. durans* NBRC 112876^T, the ANiB, ANIm, AAI, and digital DNA-DNA hybridization values were 96.29%, 97.10%, 96.7%, and 82.29%, respectively. Based on the phenotypic characteristics and genome analysis, it was identified as *M. durans*. Its genomic sequence data revealed the PHB and CoQ10 biosynthesis genes. Therefore, the results offer suggestions for further investigation into possible applications of this bacterium in biotechnology. The draft genome was deposited at DDBJ/EMBL/GenBank (DNA Databank of Japan/European Molecular Biology Laboratory/Genbank) (JAY-EEX000000000).

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Specifications Table

Subject	Biology
Specific subject area	Microbiology, Genomics, Biotechnology
Type of data	Table, Figure
Data collection	<i>Methylobacterium</i> sp. LRY1–8 was cultivated on ISP 2 medium. Genomic DNA was extracted from a pure culture of <i>Methylobacterium</i> sp. LRY1–8 using a DNeasy UltraClean Microbial Kit (Qiagen). A DNA library [a QIAseq FX DNA library kit (Qiagen) and an Illumina MiSeq platform with a MiSeq reagent kit version 3] and generation of contigs [trimmomatic tool (https://astrobiomike.github.io/genomics/de_novo_assembly#trimmomatic), and SPAdes version 3.12] were performed. The JSpeciesWS web service, enveomics (http://enve-omics.ce.gatech.edu/aai/) and the genome-to-genome distance calculator (GGDC 2.1) with the BLAST+ method (formula 2, identities/HSP length) were selected for genomic identification of strain LRY1–08. Genome was annotated according to The DFAST server, Rapid Annotation Server Technology (RAST) and the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) using Prokka version 1.12 for analyzing polyhydroxybutyrate- and ubiquinone-10- associated genes.
Data source location	Source: Lichen Location (City/Town/Region): Huai Yang Subdistrict, Klaeng District, Rayong province (12°45'20.7"N 101°34'57.1"E), Eastern part Country: Thailand
Data accessibility	The 16S rRNA gene sequence was deposited at DDBJ/EMBL/GenBank under the accession number PP065492 https://www.ncbi.nlm.nih.gov/nucleotide/PP065492 The whole genome was deposited at DDBJ/EMBL/GenBank under the accession number JAYEEX000000000. https://www.ncbi.nlm.nih.gov/nucleotide/JAYEEX000000000 The prediction of the PHB biosynthesis pathway https://figshare.com/articles/dataset/File-1-Prediction-the-PHB-biosynthesis-pathway_pdf/25658919 The prediction of the CoQ10 biosynthesis pathway https://figshare.com/articles/dataset/File2_Prediction_of_the_CoQ10_biosynthesis_pathway_pdf/25658931
Related research article	Mingrapoch N, Tanasupawat S, Charnvanich D, Cell lysis methods and coenzyme Q10 production of <i>Methylobacterium</i> strains. Thai J. Pharm. Sci. 44 (4) (2020) 256–260. https://digital.car.chula.ac.th/tjps/vol44/iss4/8 .

1. Value of the Data

- These data provide the source for the description of *Methylobacterium durans* that was originally published only single isolate.
- These data are fundamental to environmental and clinical microbiology.
- These data serve to conduct comparative genomics in a ubiquitous and pink-pigmented facultatively methylotroph related gene and allow a better understanding of the mechanisms involved in ubiquinone-10 pathway.
- This information provides a resource for studying gene structure and function.

2. Background

The genus *Methylobacterium*, a rod- and pink-pigmented facultatively methylotroph (PPFM) and ubiquitous bacterium, was proposed by Patt *et al.* [1] and *Methylobacterium organophilum* was the type species. According to the taxonomic analysis of species boundary lines, identities [ANib, ANim, AAI (>95%)] and dDDH value (>70%), and polyphasic characteristics, the genus

currently comprises 50 species with validly published and correct names. (<http://www.bacterio.net/methylo-bacterium.html>, March 7, 2024) [2–5]. Strain LRY1–08 (=JCM 33,120) was isolated from a lichen (*Parmotrema* sp.) collected from Rayong province, Thailand. It showed the highest yield of coenzyme Q10 or ubiquinone-10 (CoQ10) at 0.9173 ± 0.05 mg/L [6], similar to *Methylobacterium* sp. XJLW (22.28 mg of CoQ10/L), revealing many white particles within the cells [7]. Nevertheless, biochemical properties and genomic data of strain LRY1–08 have not yet been studied. The aim of this work was to characterize the phenotypic and genotypic features of strain LRY1–08. The draft genome was analyzed and annotated for further elucidation of bioactive compound pathways related to cosmeceutical applications such as polyhydroxybutyrate (PHB) and CoQ10.

3. Data Description

Cells of strain LRY1–08 are rod-shaped (Fig. 1). It is positive for urease and β -galactosidase but negative for capric acid and adipic acid assimilation. The phenotypic characteristics of strain and *M. durans* NBRC 112876^T are shown in Table 1.

The phylogenetic tree of 16S rRNA gene sequences of strain LRY1–08 to other *Methylobacterium* species is shown in Fig. 2. The highest 16S rRNA gene similarity was 99.8% with *M. durans* NBRC 112876^T. Its draft genome sequence was 5263,245 bp with a genomic G + C content of 70.0%. The genome statistics of strain LRY1–08 and related type strains were summarized in Table 2. ANiB, ANiM, AAI and DNA-DNA hybridization (dDDH) values between strain LRY1–08 and *M. durans* NBRC 112876^T, were 96.10%, 96.29%, 96.70% and 82.29% (C.I. 69.8–75.7%), respectively (Table 3).

Additionally, strain LRY1–08 was observed under a transmission electron microscope (TEM) exhibited numerous white particles with high refraction inside cells (Fig. 1). The genome analysis indicating the presence of genes involved in synthesis of isoprenoid compounds, including ubiquinone-10 and polyhydroxybutyrate (PHB) (Fig. 3).

Its genome possesses genes for the key enzymes for the PHB biosynthesis pathway; acetyl-CoA C-acetyltransferase (*phaA*), acetoacetyl-CoA reductase (*phaB*), and poly (3-hydroxy-

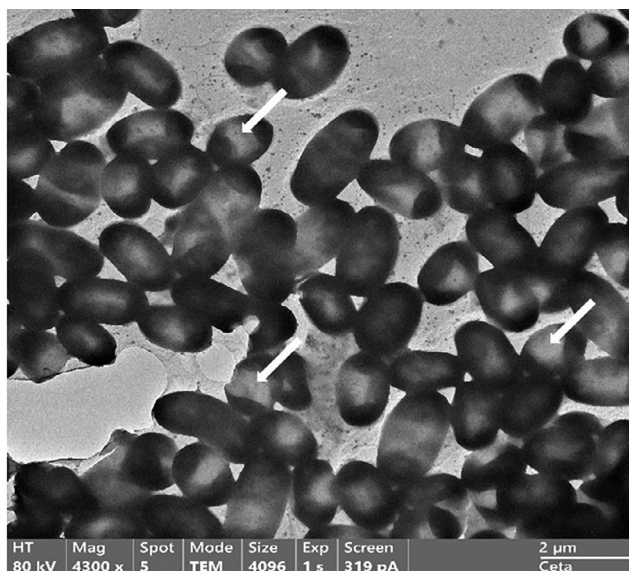


Fig. 1. Transmission electron micrograph of LRY1–08 grown on ISP 2 agar plate for 5 days.

Table 1Phenotypic characteristics of strain LRY1-08 and *M. durans* NBRC 112876^T.

Characteristics	LRY1-08	NBRC 112876 ^T
Oxidase	+	+
Catalase	+	+
Nitrate reduction	+	+
Urease	+	+
β -Galactosidase	-	-
Arginine dihydrolase	w	+
Hydrolysis of gelatin	+	-
Indol production from tryptophan	-	-
Glucose fermentation	-	-
Assimilation of		
Capric acid	-	-
Adipic acid	-	-
G + C content (mol%)	70.0	68.7

+, positive; w, weak positive; -, negative reaction.

Table 2

Genome statistics of strain LRY1-08 and related type strains.

Genome of	Features						
	Accession no.	Genome size (bp)	G + C content (%)	No. of Contigs	Protein coding genes	rRNA gene	tRNA gene
1	JAYEEX000000000	5263,245	70.0	79	4952	3	57
2 ^a	CP029550	6788,375	68.7	1	6917	12	68
3 ^a	VZZK000000000	6288,989	70.0	343	6193	3	56
4 ^a	BJZU000000000	6648,145	68.5	111	6514	1	57
5 ^a	BPQD000000000	5495,114	67.8	175	5379	4	51
6 ^a	BPQP000000000	4677,674	68.5	61	4385	2	50
7 ^a	FOPM000000000	4524,655	68.7	77	4211	5	54

Strains: 1, LRY1-08; 2, *Methylobacterium durans* NBRC 112876^T; 3, *Methylobacterium oxalidis* DSM 24028^T; 4, *Methylobacterium soli* DSM 21955^T; 5, *Methylobacterium iners* JCM 16407^T; 6, *Methylobacterium adhaesivum* DSM 17169^T; 7, *Methylobacterium gossipiicola* NRRL B-51692^T. ^a Data obtained from GenBank.

Table 3

ANIb, ANIm and dDDH values among draft genomes of strain LRY1-08 and related type strains.

Genome	Reference genome	ANIb%	ANIm%	%dDDH (Formula2) ^a	Prob.DDH \geq 70 %	Model C.I.
1	2	96.29	97.10	82.29	82.95	[69.8 - 75.7 %]
	3	86.14	88.37	0.18	0.18	[28.9 - 33.8 %]
	4	81.13	86.30	0.01	0.01	[22.5 - 27.3 %]
	5	78.61	85.35	0	0	[20.9 - 25.7 %]
	6	77.59	84.70	0	0	[19.9 - 24.6 %]
	7	77.57	84.66	0	0	[19.8 - 24.5 %]

Strains: 1, LRY1-08; 2, *Methylobacterium durans* NBRC 112876^T; 3, *Methylobacterium oxalidis* DSM 24028^T; 4, *Methylobacterium soli* DSM 21955^T; 5, *Methylobacterium iners* JCM 16407^T; 6, *Methylobacterium adhaesivum* DSM 17169^T; 7, *Methylobacterium gossipiicola* NRRL B-51692^T. All data were obtained from this study.

^a Recommended formula (identities/HSP length) liberated of genome length and thus prosperous against the use of incomplete draft genomes.

alkanoate) polymerase subunit (*phaC*) (Table 4) [5,8]. For the ubiquinone pathway, the most significant genes, *Dxs*, *ubiA*, *UbiG*, and *ispA*, involved in biosynthesis are presented in Table 5 [9-10]. The prediction of the isoprenoid biosynthesis pathway is described in supplementary data (Figs. S1 and S2).

Table 4

Polyhydroxybutyrate-associated genes by DFAST and RAST: +, detected genes: -, not detected genes.

Biosynthesis of	Genes	Functions	LRY1-08 ¹	XJLW ²	ATH 2.4.1 ³
Polyhydroxybutyrate	<i>yngG</i>	Hydroxymethylglutaryl-CoA lyase	-	+	+
	<i>phaA</i>	Acetyl-CoA C-acetyltransferase	+	+	+
	<i>phaB</i>	Acetoacetyl-CoA reductase	+	+(RAST)	-
	<i>paah</i>	3-Hydroxybutyryl-CoA dehydrogenase	+	+	+
	<i>bdh</i>	3-Hydroxybutyrate dehydrogenase	+	+	+
	<i>fadJ</i>	Enoyl-CoA hydratase/isomerase	+	+	+
	<i>bdh</i>	3-Hydroxybutyrate dehydrogenase	+	+	+
	<i>fadN</i>	3-Hydroxyacyl-CoA dehydrogenase	+	+	+
	<i>phaC</i>	Poly(3-hydroxyalkanoate) polymerase	+	+	-
	<i>phaZ</i>	Poly(3-hydroxyalkanoate) depolymerase	-	-	+

¹ LRY1-08.² *Methylobacterium* sp. XJLW.³ *Cereibacter sphaeroides*.**Table 5**

Ubiquinone-associated genes by DFAST and RAST: +, detected genes: -, not detected genes.

Biosynthesis of	Genes	Functions	Ubiquinone 10		Ubiquinone 8
			LRY1-08	XJLW	NCTC 9112 ⁴
Ubiquinone	<i>dxs</i>	1-Deoxy-D-xylulose-5-phosphate synthase	+	+	+
	<i>dxr</i>	1-Deoxy-D-xylulose-5-phosphate reductoisomerase	+	+	+
	<i>ispD</i>	2-C-methyl-D-erythritol-4-phosphate cytidyltransferase	-	-	+
	<i>ispDF</i>	Bifunctional 2-C-methyl-D-erythritol 4-phosphate cytidyltransferase/2-C-methyl-D-erythritol 2,4-Cyclodiphosphate synthase	+	+	-
	<i>ispE</i>	4-(Cytidine 5'-diphospho)-2-C-methyl-D-erythritol kinase	+	+	+
	<i>ispF</i>	2-C-methyl-D-erythritol 2,4-cyclodiphosphate synthase	-	-	+
	<i>ispG</i>	Flavodoxin-dependent (E)-4-hydroxy-3-methylbut-2-enyl-diphosphate synthase	+	+	+
	<i>ispH</i>	4-Hydroxy-3-methylbut-2-enyl diphosphate reductase	+	+	+
	<i>ispA</i>	Geranylgeranyl diphosphate synthase	+	+	+
	<i>ddsA</i>	Decaprenyl diphosphate synthase	+	+	-
	<i>ispB</i>	Octaprenyl diphosphate synthase	(RAST)	(RAST)	+
	<i>ubiA</i>	4-Hydroxybenzoate octaprenyltransferase	+	+	+
	<i>ubiC</i>	Shikimate dehydrogenase	+	+	+
	<i>ubiD</i>	3-Octaprenyl-4-hydroxybenzoate decarboxylase	-	+	+
	<i>ubiE</i>	2-Methoxy-6-polyprenyl-1,4-benzoquinol	+	+	+
	<i>ubiF</i>	UbiF family hydroxylase	+	+	+
	<i>ubiG</i>	Bifunctional 3-demethylubiquinone 3-O-methyltransferase	+	+	+
	<i>ubil</i>	2-Polyprenylphenol 6-Hydroxylase/2-octaprenylphenol hydroxylase	+	+	+
	<i>ubiH</i>	FAD-dependent monooxygenase	+	+	+
	<i>ubij</i>	Ubiquinone biosynthesis accessory factor UbiJ	-	-	+
	<i>ubiK</i>	Ubiquinone biosynthesis accessory factor UbiK	+	-	+
	<i>ubiX</i>	Flavin prenyltransferase	-	+	+

⁴ *Escherichia coli* NCTC 9112.

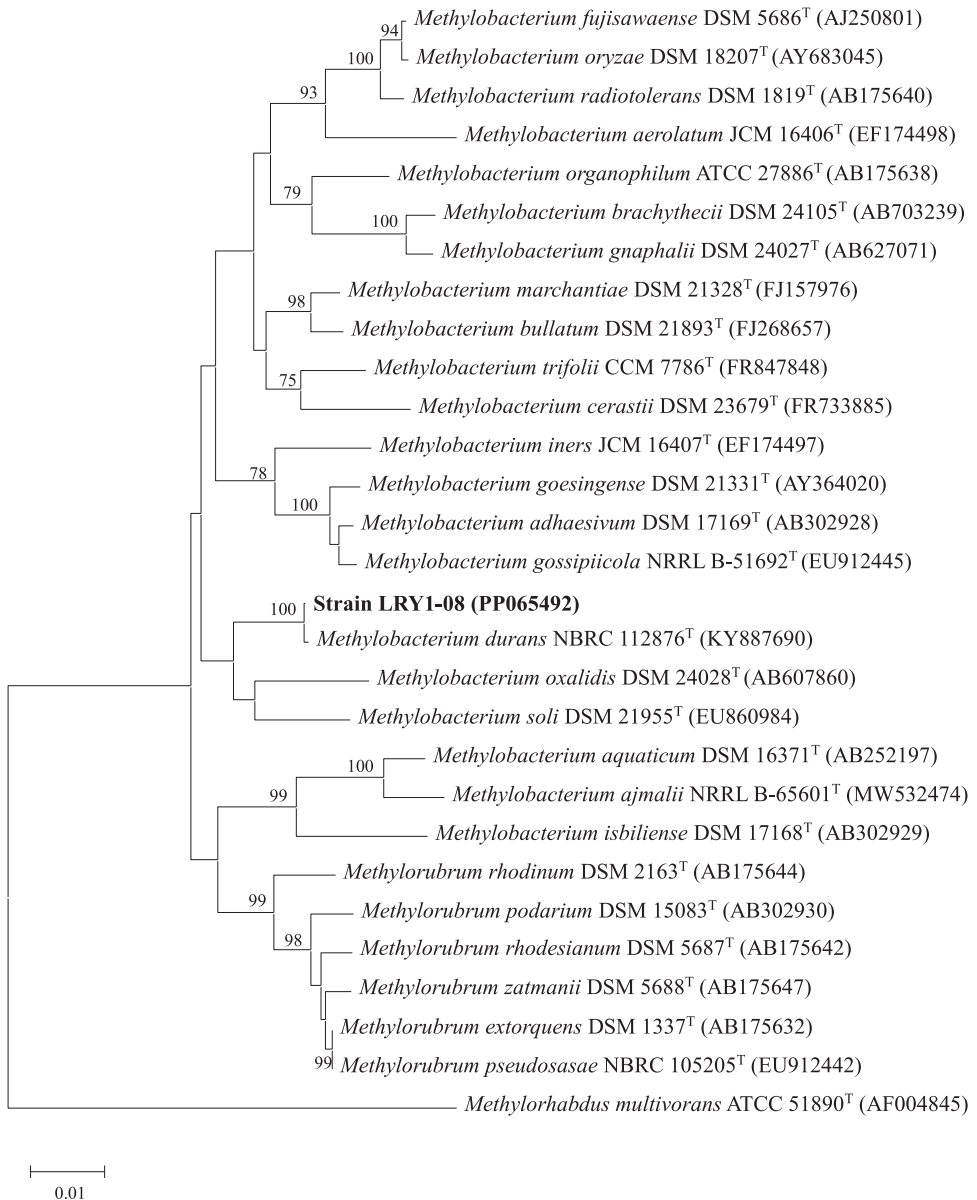


Fig. 2. Phylogenetic tree of strain LRY1-08 and related type strains based on 16S rRNA gene sequences. The branching pattern was generated by the neighbor-joining method. Bar, 10 substitutions per 1000 nucleotide positions.

4. Experimental Design, Materials and Methods

Methylobacterium durans LRY1-08 was isolated from lichen, Thailand by using spread-plate technique duplicate on ISP 2 agar plates as reported previously [2]. This isolate was taxonomic characterized using phenotypic and genotypic characteristics as previously described [1]. Transmission electron microscopy was observed according to the method of Cui *et al.* [7]. Physiological and biochemical properties were tested using the commercial API 20NE systems (bioMérieux), according to the manufacturer's instructions.

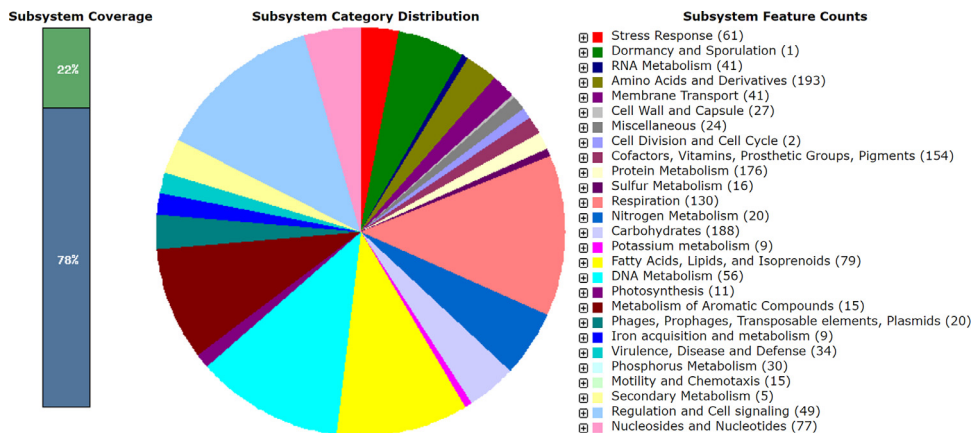


Fig. 3. Subsystem distribution of *Methylobacterium durans* LRY1-08 constructed from the RAST annotation server.

The 16S rRNA gene was amplified according to the standard procedure and the purified PCR products were sequenced on a DNA sequencer (Macrogen, Korea) [11]. The 16S rRNA gene sequence was deposited in the GenBank under the accession number PP065492. The aligned 16S rRNA gene sequence of strain LRY1-08 against sequences of *Methylobacterium* type strains was constructed using a phylogenetic tree based on neighbor-joining algorithms using MEGA 7.0 [12].

Whole-genome sequencing of strain LRY1-08 was performed at the Japan Collection of Microorganisms (JCM) as previously described [13] with minor modifications. In brief, genomic DNAs were extracted from the harvested cells using a DNeasy UltraClean Microbial Kit (Qiagen). A DNA library was constructed using a QIAseq FX DNA library kit (Qiagen), and sequenced on an Illumina MiSeq platform with a MiSeq reagent kit version 3 (600 cycles, 300 bp paired-end reads; Illumina). The generated raw reads were trimmed and filtered using Trimmomatic tool (https://astrobiomike.github.io/genomics/de_novo_assembly#trimmomatic), and then a de novo assembly of the high-quality reads into contigs using SPAdes version 3.12 was accomplished [14]. The assembled genome of strain LRY1-08 was deposited in GenBank (accession number: JAYEEX000000000). The genome was annotated by using the DFAST server [15] and Rapid Annotation Server Technology (RAST) [16] and the NCBI Prokaryotic Genome Annotation Pipeline (PGAP). The genome was annotated using Prokka version 1.12 [17] in line with the NCBI prokaryotic genome annotation pipeline (PGAP). The average amino acid identity (AAI) was calculated by enveomics (<http://enve-omics.ce.gatech.edu/aai/>) [3]. An average nucleotide identity (ANI) was calculated among the genomes of strain LRY1-08 and *Methylobacterium* type strains based on ANI-Blast (ANiB) and ANI-MUMmer (ANIm) algorithms implemented within the JSpeciesWS web service [18]. Values of the digital DNA-DNA hybridization (dDDH) was calculated using the genome-to-genome distance calculator (GGDC 2.1) with the BLAST+ method [19]. Results were based on the recommended formula 2 (identities/HSP length), which is useful when dealing with incomplete draft genomes.

Limitations

None.

Ethics Statement

No ethical issue.

CRedit Author Statement

Sirilak Namwong: Methodology, Formal analysis, Writing- Original draft preparation. **Shingo Kato:** Data curation, Formal analysis, software. **Takao Iino:** Data curation, Formal analysis, software. **Takashi Itoh:** Validation, Writing-review & editing. **Moriya Ohkuma:** Supervision, Project administration. **Pawina Kanchanasin:** Provision, Resources. **Wongsakorn Phongsopitannun:** Methodology, Investigation, resources. **Somboon Tanasupawat:** Conceptualization, Validation, Resources, Methodology, Project administration, Supervision, Funding acquisition.

Data Availability

Genome of LRY1-08 (Original data) (SOL Genomics).
The prediction of the CoQ10 biosynthesis pathway (Original data) (LRY1-08).
16S rRNA gene sequence of LRY1-08 (Original data) (SOL Genomics).
The prediction of the PHB biosynthesis pathway (Original data) (LRY1-08).

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Supplementary Materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.dib.2024.110485.

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