Supplementary Material

Long-term mercury contamination does not affect the microbial gene potential for C and N cycling in soils but enhances detoxification gene abundance

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Supplementary Tables: 3 Supplementary Figure: 1
 Table 1. Statistics of the genome assembly.

Parameter	Size
Assembly size (bp ⁺)	12,375,076,959
Number of high-quality reads after filtering	786,878,084
Number of MEGAHIT assembly of reads into contigs	17,743,321
Maximum contig length (bp)	207,300
Minimum contig length (bp)	200
Mean contig length (bp)	697
N ₅₀ contig length (bp)	782
Number of predicted protein-coding genes with MetaGeneMark [†]	26,237,952
Number of predicted protein-coding genes annotated with EggNOG database *	12,468,915
Number of predicted protein-coding genes annotated with CAZy database [#]	262,248
Number of predicted protein-coding genes annotated with NCyc database [‡]	42,859

⁺ bp: base pairs

[†] MetaGeneMark: Tool for gene prediction in metagenomes, utilizing metagenome parameters and gene prediction

* EggNOG: Database of evolutionary genealogy of genes: non-supervised orthologous groups

[#] CAZy: Carbohydrate-active enzymes database

[‡] NCyc: Curated integrative database for fast and accurate metagenomic profiling of N cycling genes

Table 2. Richness and Shannon index of normalized counts for all predicted genes and for genes ofthe EggNOG, CAZy, and NCyc databases.

	Richness			Shannon			
Database	Low Hg	Moderate Hg	High Hg	Low Hg	Moderate Hg	High Hg	
All predicted genes	12,063,152	12,493,639	11,582,793	15.6	15.8	15.7	
EggNOG	6,846,271	7,099,416	6,594,610	15.0	15.2	15.1	
CAZy	183,586	190,025	177,268	11.3	11.5	11.4	
NCyc	28,109	29,496	28,146	9.4	9.7	9.6	

Table 3. Main effects on functional gene alpha-diversity (Richness and Shannon index) and betadiversity of all predicted genes and the genes annotated with the EggNOG, CAZy, and NCyc databases for soils with different Hg contamination levels (low, moderate, high).

			Alpha-d	liversity*			
		Richness Shannon		nnon	Beta-diversity ⁺		
Databases	DF [#]	F	Р	F	Р	Pseudo-F	Р
All predicted genes	2	2.98	0.42	3.06	0.16	3.08	0.011
EggNOG	2	3.01	0.43	3.21	0.15	2.76	0.016
CAZy	2	2.93	0.48	3.11	0.17	3.03	0.013
NCyc	2	3.10	0.39	3.30	0.09	2.72	0.015

* Effect of Hg contamination assessed by analysis of variance (ANOVA).

⁺ Effect of Hg contamination assessed by permutational multivariate analysis of variance (PERMANOVA).

[#] Values represent degrees of freedom (*DF*), *F*-ratio (*F*), pseudo-*F* ratio (Pseudo-*F*), and the level of significance (*P*); significant values (P < 0.05) are in bold.



Figure 1: Schematic pathways of Hg²⁺, MeHg⁺, RHg⁺, and Hg⁰, and the location and functions of MerA to MerG, MerP, MerR, and MerT proteins (circles) and of HgcA and HgcB proteins (squares) in a bacterial cell, following the schemes of Dash et al. (2012) and Date et al. (2019). Purple shading indicates proteins of significantly overrepresented genes at the high Hg contamination level, as indicated in Table 9. Cp: cytoplasm, Im: inner membrane, MeHg⁺: methyl Hg, Om: outer membrane, Op: *mer* operon, Pp: periplasm, RHg⁺: organic form of Hg.