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Exploration of heavy metal resistance in the yeast *Wickerhamomyces anomalus*: Implications for bioremediation

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INTRODUCTION

The inadequate disposal of wastewater containing toxic heavy metals and industrial contaminants has become a critical issue today, posing serious risks to the health of humans, animals, and the environment. Numerous studies have investigated how heavy metals are absorbed, accumulated, and transformed using microorganisms (1). The yeast strain *Wickerhamomyces anomalus* M10 has demonstrated notable resistance to several heavy metals, showing promising potential for bioremediation (2). Therefore, through computational methods (In Silico studies) to investigate metal resistance mechanisms, relevant genes related to this resistance were identified. Specialized databases like BacMet and InterPro were used (3), compiling genes from bacteria and fungi that enhance survival under environmental stress.

MATERIALS AND METHODS

Genome Sequencing

DNA of *Wickerhamomhyces anomalus* M10 were sent to Macrogen for sequencing using the Illumina platform (NovaSeq, 150PE). The yeast genome size is 14.2 Mb.

Raw Data Processing

Read quality was assessed using FastQC, and reads were processed using Trimommatic to remove adapters while adhering to a Phred score of 20 as the quality threshold for the removal of low-quality reads.

Genome Assembly

Whole-genome assembly was performed using the Redundans pipeline, leveraging a reference genome obtained from JGI-Mycocosm. Assembly quality was assessed using QUAST.

Structural Annotation

Gene prediction was carried out using the FUNgap pipeline, which incorporates the BUSCO tool for quality assessment.

Functional Annotation

Two tools were employed for functional annotation. First, a curated experimental database called BACmet, containing heavy metal resistance genes, was used for a blast search. Entries with over 50% coverage were retained. Additionally, the InterProScan tool was used to identify protein domains related to heavy metal resistance.

Image 1. Interpro main interface

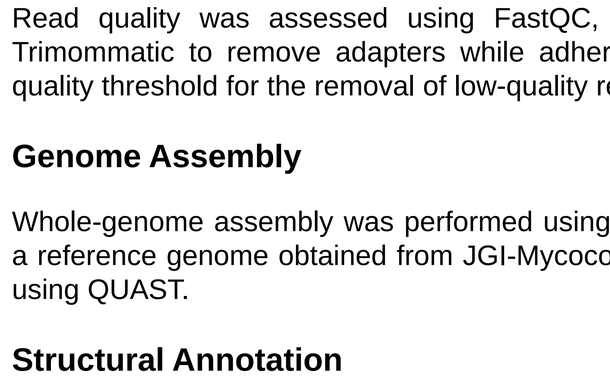
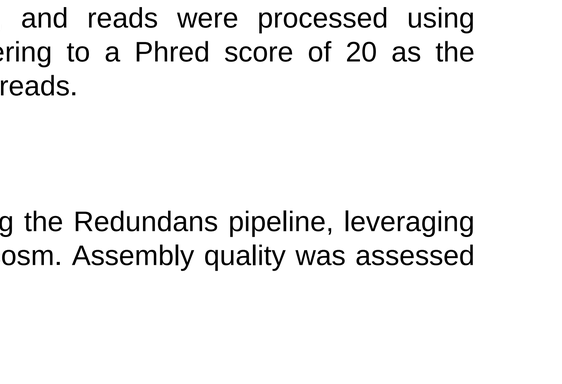


Image 2. BACmet main interface



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RESULTS

Table 1. Genome assembly statistics.

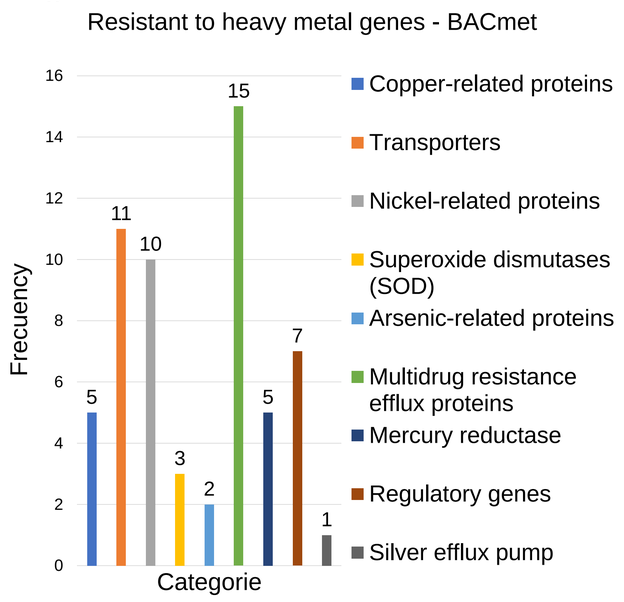
Contigs	411
Contigs (>=0 bp)	703
Contigs (>= 1000 bp)	335
Contigs (>= 5000 bp)	80
Contigs (>= 10000 bp)	24
Contigs (>= 25000 bp)	10
Contigs (>= 50000 bp)	9
Largest contig	3 214 346
Total length	14 343 505
Total length (>=0 bp)	14 438 866
Total length (>=1000 bp)	14 288 948
Total length (>=5000 bp)	13 664 680
Total length (>=10000 bp)	13 272 614
Total length (>=25000 bp)	13 089 063
Total length (>=50000 bp)	13 060 986
N50	1 973 147
N90	455 794
auN	1 847 581
L50	3
L90	8
GC (%)	34.5
Mismatches	
N´s per 100 Kpb	1068.5
N´s	153 260

Table 2. Genome annotation attributes

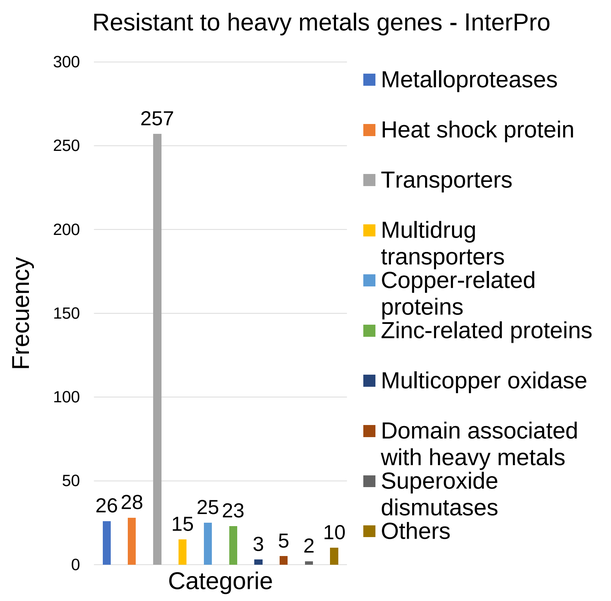
Attributes	Values
Total protein-coding genes	7118
Transcript length (avg/med)	1,382.8/1,165.0
CDS length (avg/med)	1,350.5/1,140.0
Protein length (avg/med)	450.2/380.0
Exon length (avg/med)	1020.0-813.0
Intron length (avg/med)	99.3/57.0
Spliced genes	1656 (23.26%)
Gene density (genes/mb)	492.98
Number of introns	2,307
Number of introns per genes (med)	1.0
Number of exons	9,425
Number of exons per genes (med)	1.0

A total of 7118 protein-coding genes were identified.

Assembly quality. The assembly demonstrates exceptional quality, characterized by a notably high N50 value, a total length consistent with the genome size, and the presence of a limited number of contigs



Graphic 1. Identified genes with BACmet



Graphic 2. Identified genes with InterPro

Using BacMet, we identified 59 genes, including 11 ABC-type transporters, 10 related to resistance and the efflux of multiple drugs, as well as proteins for copper (5), nickel (10), arsenic (2), mercury (5), and silver (1). These proteins perform functions in binding, reduction, and transport, along with superoxide dismutase proteins, DNA regulation, and repair genes. Additionally, through InterPro, we found 394 genes, including 257 transporters, 25 copper related proteins, and 23 zinc-related proteins. 15 multidrug resistance proteins and other DNA regulation and repair genes (10) were detected.

CONCLUSIONS

The bioinformatics analysis of *Wickerhamomyces anomalus* underscores its promising potential for bioremediation in metal-contaminated environments, specifically in addressing nickel and arsenic pollution, thanks to its resistance-related genes. Nevertheless, further in vivo studies are necessary to validate its ability to survive and effectively transform these metals. This research not only uncovers genes that broaden the horizons of microbial bioremediation but also advances the understanding of the significant role yeast strains can play in addressing environmental pollution challenges.

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