



Data in Brief

Transcriptome response to copper heavy metal stress in hard-shelled mussel (*Mytilus coruscus*)



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ABSTRACT

The hard-shelled mussel (*Mytilus coruscus*) has considerably one of the most economically important marine shellfish worldwide and considered as a good invertebrate model for ecotoxicity study for a long time. In the present study, we used Illumina sequencing technology (HiSeq2000) to sequence, assemble and annotate the transcriptome of the hard-shelled mussel which challenged with copper pollution. A total of 21,723,913 paired-end clean reads (NCBI SRA database SRX1411195) were generated from HiSeq2000 sequencer and 96,403 contigs (with N50 = 1118 bp) were obtained after de novo assembling with Trinity software. Digital gene expression analysis reveals 1156 unigenes are upregulated and 1681 unigenes are downregulated when challenged with copper. By KEGG pathway enrichment analysis, we found that unigenes in four KEGG pathways (aminoacyl-tRNA biosynthesis, apoptosis, DNA replication and mismatch repair) show significant differential expressed between control and copper treated groups. We hope that the gill transcriptome in copper treated hard-shelled mussel can give useful information to understand how mussel handles with heavy metal stress at molecular level.

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Specifications [standardized info for the reader]

Organism/cell line/tissue	<i>Mytilus coruscus</i> /whole organism or gill
Sex	N/A
Sequencer or array type	Illumina HiSeq2000
Data format	Raw and processed
Experimental factors	Transcriptome profiling of gill tissue on copper treatment
Experimental features	Gill tissues from control and copper treated groups were dissected for total RNA extraction. Prepared cDNA libraries were paired-end sequenced by HiSeq2000 system. The obtained data was subjected for de novo transcriptome assembly using Trinity, and coding regions were predicted by BLAST. We performed BLASTP against the NR, NT, Swiss-Prot, KEGG, COG and GO database using BLAST with an e-value cut off of 1e-6 to annotate identified proteins.
Consent	N/A
Sample source location	Samples were collected from Qingbang island, Dongji, Zhoushan in Zhejiang province, China.

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1. Direct link to deposited data

<http://www.ncbi.nlm.nih.gov/sra/SRX1411195> SRX1411195 for contig assembly.

<http://www.ncbi.nlm.nih.gov/sra/SRX1411193> SRX1411193 for control gill.

<http://www.ncbi.nlm.nih.gov/sra/SRX1411194> for copper treated gill.

2. Introduction

Cd²⁺ and Cu²⁺ are two major toxic contaminants in marine environment which can generate oxidative stress, DNA damage, apoptosis and protein denature in various living organisms [14,17]. The gene expression profiling is frequently used to monitor the impact of heavy metal exposure in diverse vertebrate [12,13,19] and invertebrate species [1,6,11,21]. Among those diverse animal species, the mussel has been long been used as good sentinel organism that accumulates pollutants in their tissues from the surrounding environment or via food chain [16]. Mussels are filter feeders and can concentrate metals in their gills or other tissues. In the US, the blue mussel (*Mytilus edulis*) became a popular model for monitoring the levels of pollution. The hard-shelled mussel (*Mytilus coruscus*) is a common species inhabiting the temperate zone along the coastal waters of East Asia, Korea and Japan. In China, this

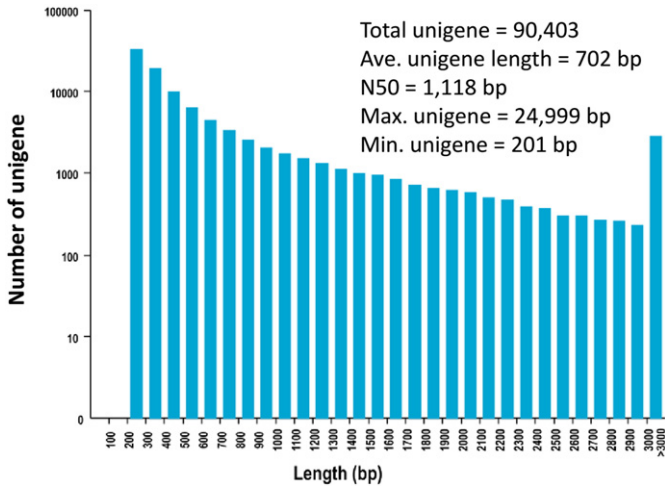


Fig. 1. Length distribution of the assembled unigene of hard-shelled mussel (*Mytilus coruscus*) transcriptome.

species is one of the most heavily commercially exploited marine bivalves, particularly within the Zhejiang Province [20]. In previous studies, *M. coruscus* has been utilized as new ecotoxicity model for marine pollution studies [7–9]. In this study, we aimed to explore the gene expression profiling between control and copper exposure *M. coruscus* by using RNAseq approach.

3. Experimental design, materials and methods

3.1. RNA extraction

Specimens of *M. coruscus* were collected from Qingbang island, Dongji, Zhoushan in Zhejiang province, China. Mussel was approximately 6 months old with an average weight of 30 g (range from 25 g to 40 g) and an average body length of 11.5 cm (range from 9.5 cm to 15.4 cm). Before Cu²⁺ challenge, mussels were acclimated indoor with room temperature for 15 days. Later, mussels were challenged

with copper sulfate at 0.2 ppm. The control mussels were cultured in the seawater without copper sulfate treatment. All dissected tissues were flash-frozen in liquid nitrogen and then stored at –80 °C prior to RNA extraction. Total RNAs were extracted by using the TRIZOL Kit (Invitrogen, Carlsbad, CA, USA) following the manufacturer's instructions. Total RNA samples were then digested by DNase I to remove potential genomic DNA contamination. Integrity and size distribution were checked with a Bioanalyzer 2100 (Agilent technologies, Santa Clara, CA, USA). Equal amounts of the high quality RNA samples from each tissue were then proceed to perform cDNA synthesis and next generation sequencing.

3.2. RNA isolation, library construction and Illumina sequencing

Initially, about 5 µg of starting total RNAs was used to synthesize the cDNA libraries by following the standard protocols of the Illumina TruSeq RNA Sample Preparation Kit (Illumina). The final library had an average fragment size of 250 bp. After KAPA quantitation and dilution, the library was sequenced on an Illumina HiSeq 2000 with 101 bp paired-end reads. A total of 43,447,826 paired-end clean reads was generated with a read length of 101 bp. The raw transcriptome sequences in the present study were deposited in the NCBI SRA database (SRX1411195).

3.3. De novo transcriptome assembly and functional annotation of gill expressed genes

Cleaned reads were de novo assembled into contigs by Trinity software [3] with default parameter settings. The transcriptome was assembled, combining 21,723,913 paired-end clean reads into 96,403 unigenes, ranging from 201 to 24,999 bp in length. The average length was 702 bp, the N50 length was 1118 bp. The unigene length distribution is shown in Fig. 1. The assembled transcriptome unigenes were subjected to similarity search against multiple databases like COG, GO, KEGG, Swissprot or NR databases using BLASTX with an e-value cut off of 1e-6. About 31% assembled unigene (29,804 unigenes) can be assigned at least one annotation terms at either COG, GO, KEGG, Swissprot or NR databases.

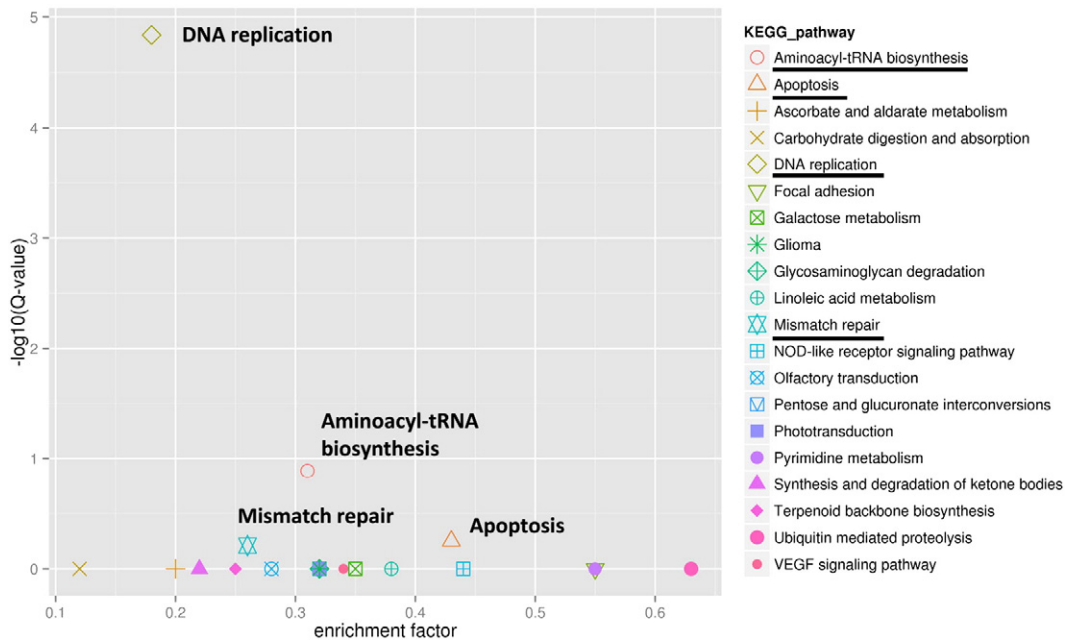


Fig. 2. The enrichment KEGG pathway comparison between control and copper treated groups.

3.4. Gene expression quantification and differential expression analysis

For gene expression comparison, two gill cDNA libraries were established and subjected to generate 8,499,247 (SRX1411193) and 7,987,935 (SRX1411194) single-end reads with 51 bp for control and copper treated groups, respectively. The cleaned reads of each RNA-seq library were mapped to the previous assembled unigenes with Bowtie program [4]. The counting of alignments was done using RSEM [5]. The differential expression statistical analysis was done using the statistical method described in the R package [2]. Differentially expressed gene (fold changes >2 and adjusted p-value < 0.001) between two samples were identified with the software. Mapping results show 2837 unigenes (1156 up and 1681 down) display significant differential expression between the control and copper-treated groups (Table S1). Among the differentially expressed genes, we found C1q domain containing protein MgC1q51, which previously identified to play important role against marine pathogen and heavy metal stress [7], is strongly upregulated in the copper treated group. By KEGG pathway enrichment analysis, we found unigenes in four KEGG pathways (aminoacyl-tRNA biosynthesis, apoptosis, DNA replication and mismatch repair) show significant differential expressed between control and copper treated groups (Fig. 2 and Table S2). All differential expressed unigenes in DNA replication and mismatch repair pathway are strongly upregulated in copper treated group. Previous study on cellular level has shown that the copper exposure will induce DNA damage and oxidative stress to induce cell damage in golden mussel [18]. The strong elevation of gene expression in DNA replication and mismatch repair pathway may play an important role on attenuating the acute copper toxicity. On the contrary, we found that genes in aminoacyl-tRNA biosynthesis pathway are down-regulated. This result is consistent with the finding that shows protein synthesis pathway is negatively affected when mussels are exposed to heavy metal contamination [15]. We hope that the gill transcriptome in copper treated hard-shelled mussel can give useful information to understand how mussel handles with heavy metal stress at molecular level.

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.gdata.2015.12.010>.

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