

Integrated metabolome–metagenome analysis provides new insights into the impacts of sea cucumber (*Apostichopus japonicus*) aestivation on marine sediment composition



大连海洋大学
DALIAN OCEAN UNIVERSITY

Wenhong Mai^a, Jingxian Sun^{a,b}, Haolin Wang^a, Chengda Li^a, Zengdong Wang^c, Cao Cheng^c, Ange Zou^c, Yaoyao Zhan^{a*}, Yaqing Chang^{a,b*}

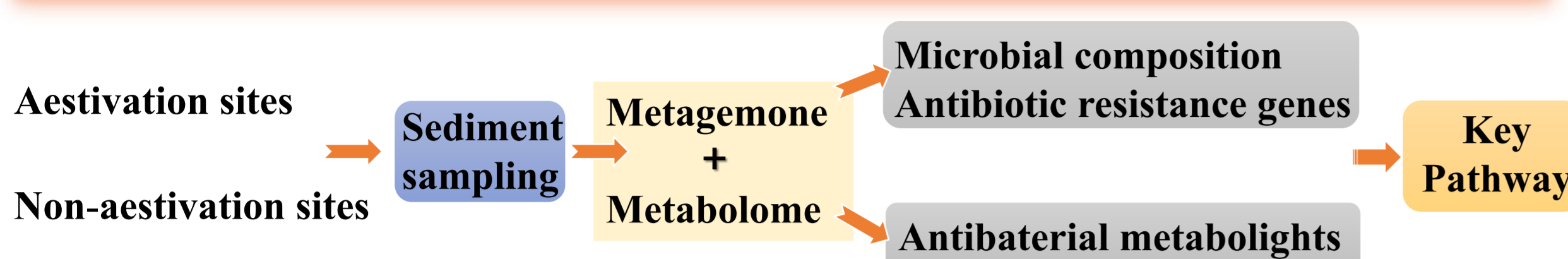
^a Key Laboratory of Mariculture & Stock Enhancement in North China's Sea, Ministry of Agriculture and Rural Affairs, Dalian Ocean University, Dalian, Liaoning, P. R. China, 116023

^b College of Life Science, Liaoning Normal University, Dalian, Liaoning, P. R. China, 116029

^c Shandong Anyuan Seed Industry Technology Co., Ltd., Yantai, Shandong, P. R. China, 265617

* Correspondence: zydlou@hotmail.com and yqkeylab@hotmail.com.

Work flow



Results and Discussion

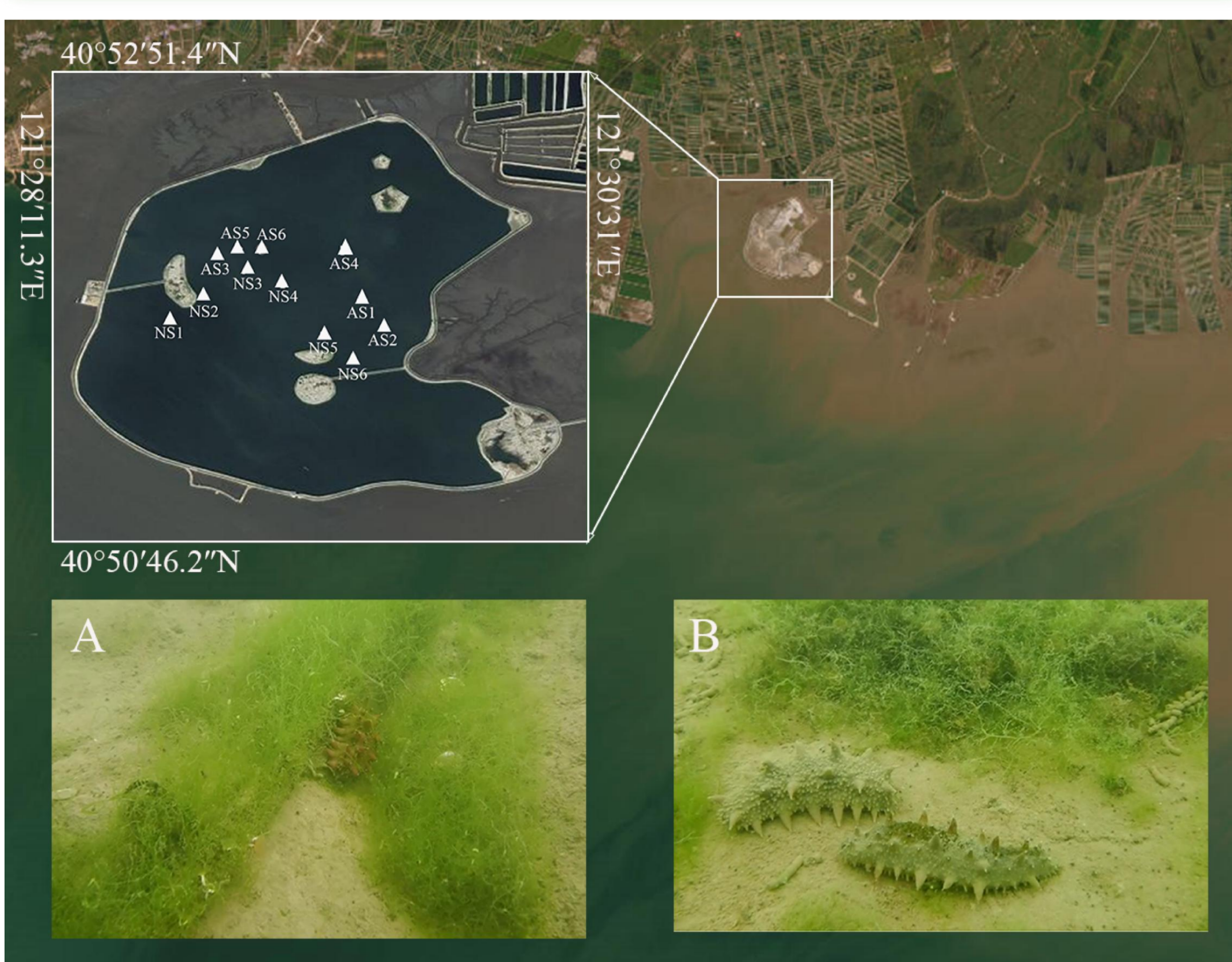


Fig. 1. Sampling sites and the morphology and habitat of aestivated (A) and non-aestivated (B) sea cucumbers. The satellite map was obtained from ArcGIS (<https://www.arcgis.com/index.html>), powered by Esri.

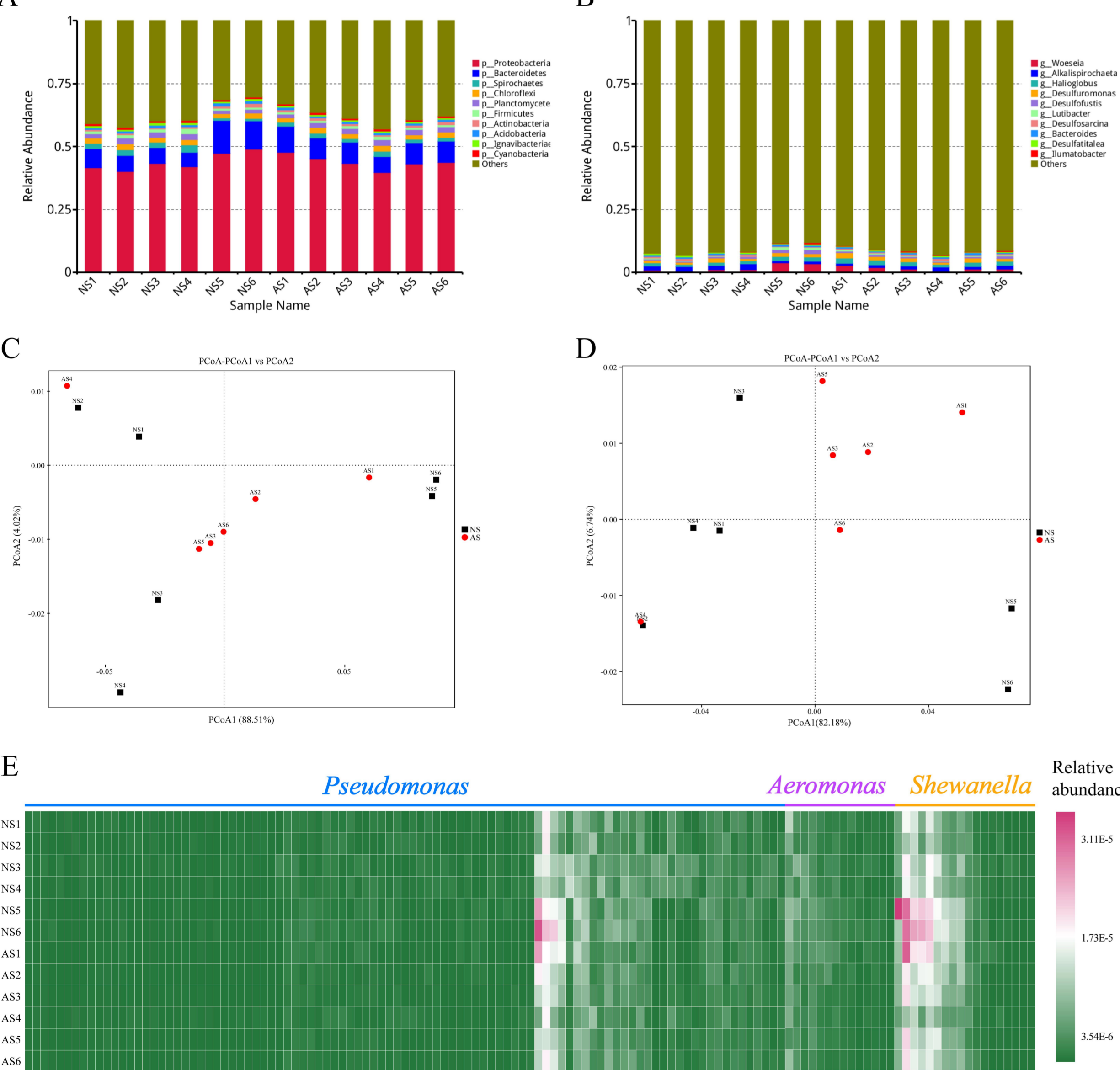


Fig. 2. Bacterial taxonomic profiles and principal coordinates analysis (PCoA) of the taxonomy of the microbial community in sediments from non-aestivation sites (NSs) and aestivation sites (ASs). (A) Relative abundances of phyla at the overall community level. (B) Relative abundances of dominant genera. (C) PCoA based on phylum level. (D) PCoA based on genus level. (E) Heat map of the relative abundances of bacterial species in genera closely correlated with sea cucumber disease and aquaculture in the aestivation site (AS) and non-aestivation site (NS) groups.

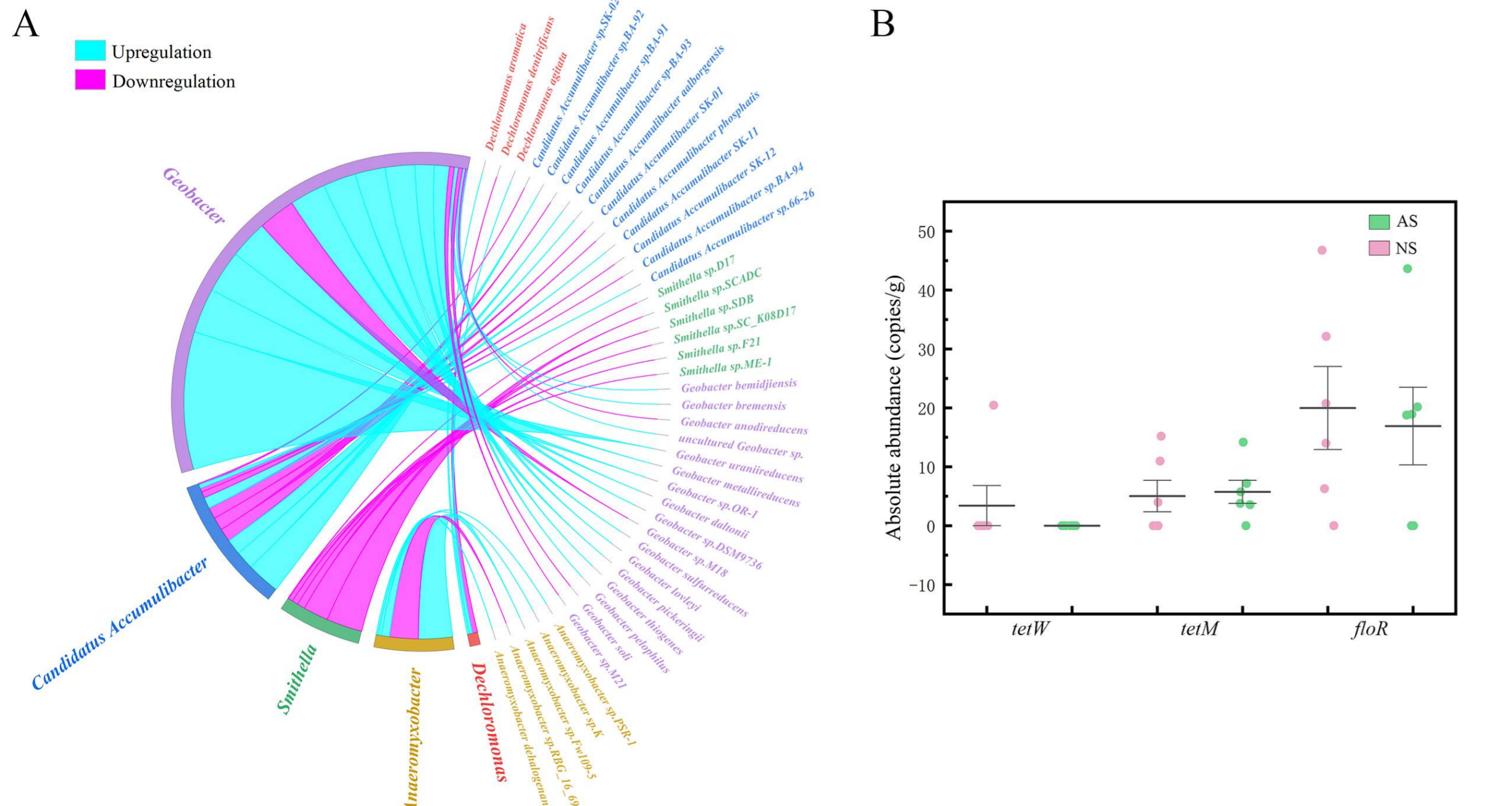


Fig. 3. Potential host genera of antibiotic resistance genes (ARGs) in sediments. (A) Chord diagram of the relative abundances of five potential host genera of ARGs in sediments. "Upregulation" means the relative abundance of the genera shows increases in sediments from aestivation site (AS) samples compared with those from non-aestivation site (NS) samples, and "Downregulation" means the opposite. (B) Scatter plot of the absolute abundance (copies/g) of ARGs in sediments from the AS and NS groups. The abscissa shows ARGs, and the bold black line means the average of the absolute abundance.

Introduction

● Sea cucumbers are typical benthic invertebrates distributed in coastal and deep-sea ecosystems worldwide. *Apostichopus japonicus* is the primary commercially farmed sea cucumber species in China and is cultivated from the coastland of Liaoning and Shandong Provinces to Zhejiang, Jiangsu, Fujian, and Guangdong Provinces. It has been documented that more than 70 species of sea cucumbers are of commercial importance for aquaculture.

● Aestivation is a unique physiological behavior of sea cucumbers. *A. japonicus* aestivation include no feeding, no locomotion, and tending to hide. Although aestivation is a survival strategy for sea cucumbers to adapt to hostile natural environmental conditions, a four-month growth stagnation would undoubtedly prolong the breeding cycle and increase the breeding cost.

● As a typical detritus feeder, *A. japonicus* ingests sediment to obtain nutrients from organic matter, bacteria, and protozoa. Besides, the survival and health status of *A. japonicus* rely heavily on farm sediment. Whether aestivation behavior affects the sediment composition of *A. japonicus* aquaculture ponds remains unknown, as well as whether there is a relationship between *A. japonicus* aestivation and sediment composition.

● This study will enrich the understanding of organism–environment interaction mechanisms and provide new hints for developing high-efficiency breeding and management strategies for sea cucumber aquaculture.

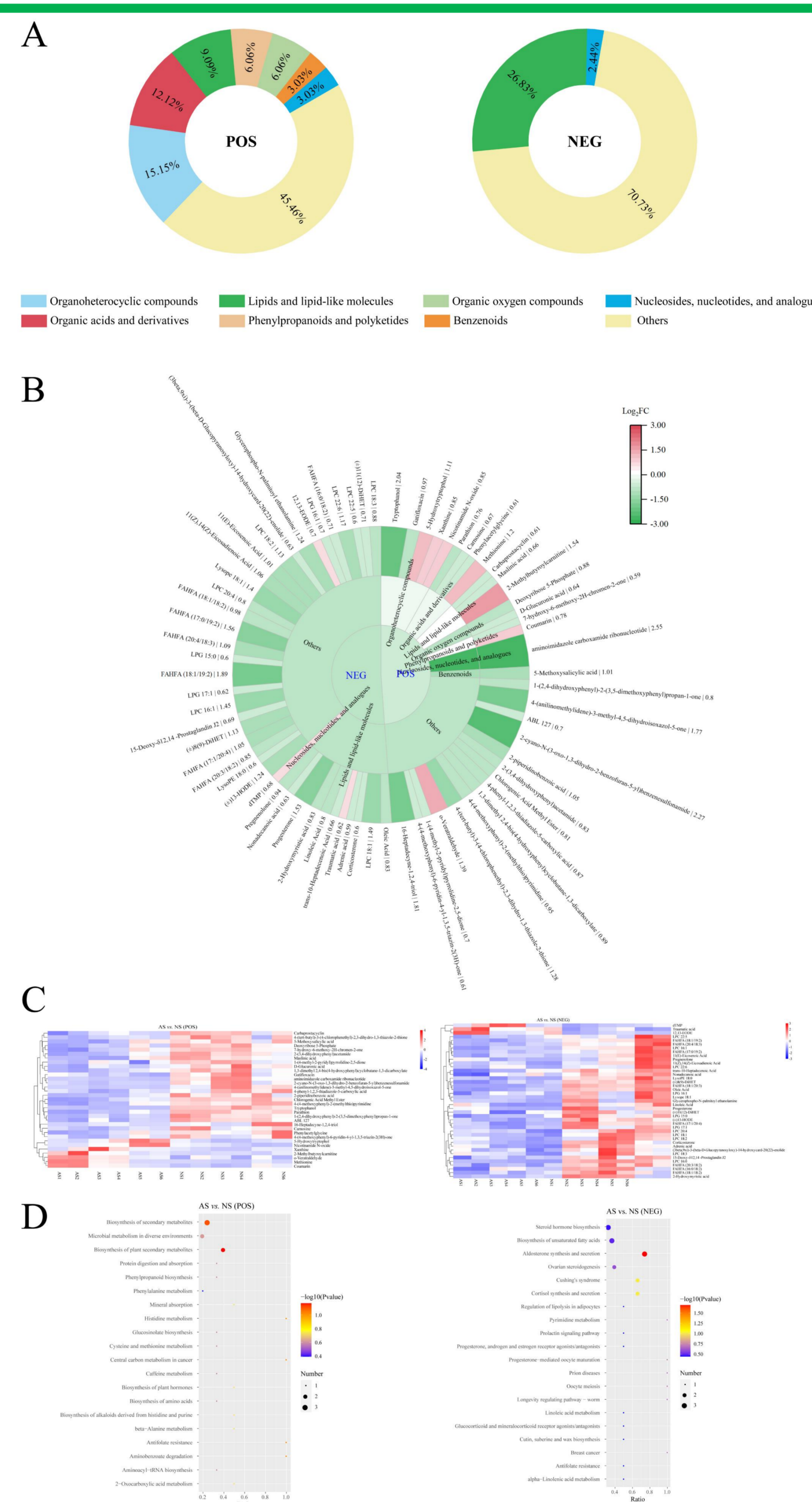


Fig. 5. Classification (A), identification (B), heat map plot (C), and Kyoto Encyclopedia of Genes and Genomes (KEGG) annotation (D) of significantly differentially expressed metabolites (SDMs) observed from a comparative metabolome analysis between sediment from aestivation site (AS) and non-aestivation site (NS) samples.

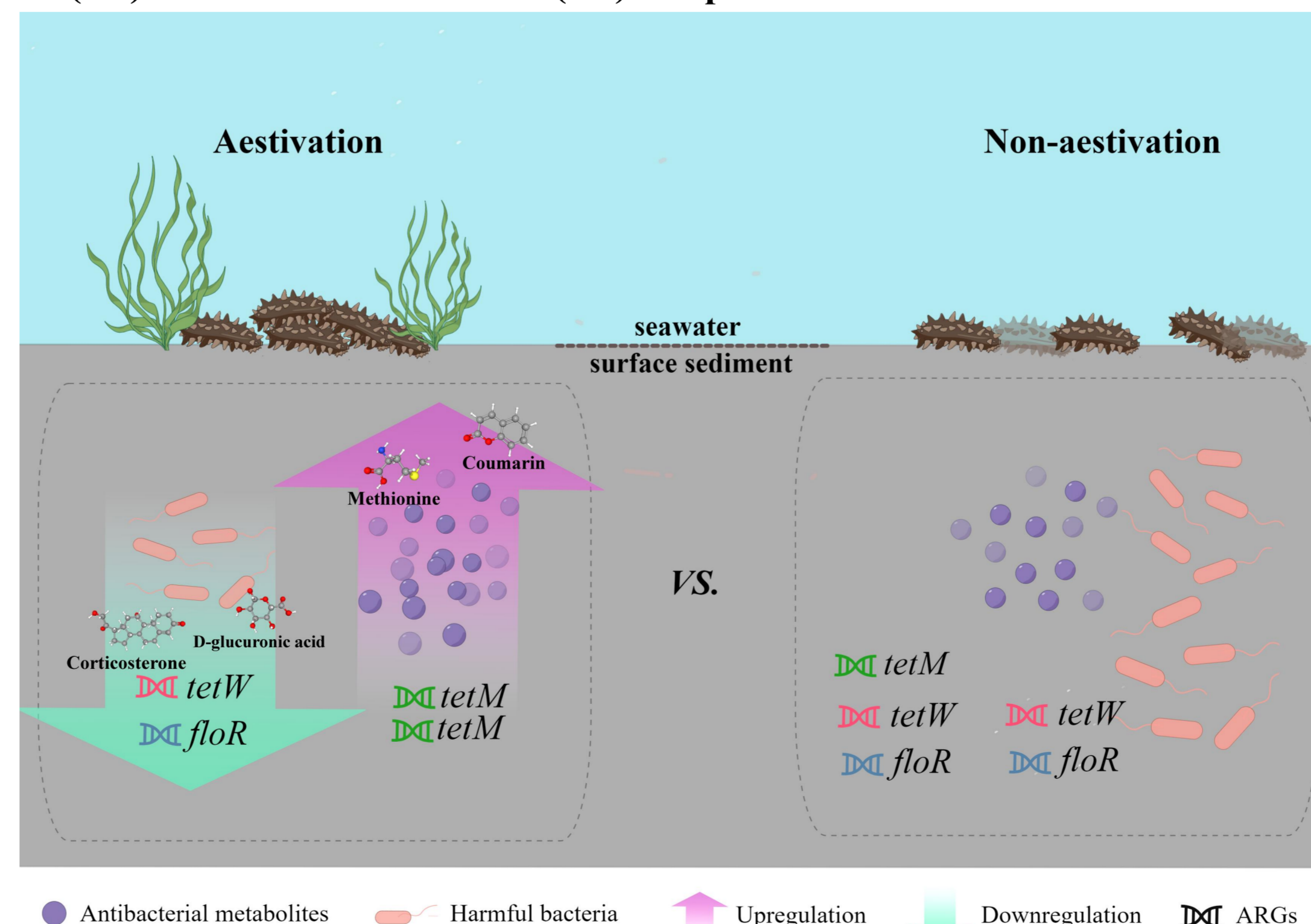


Fig. 7. Schema image of the impacts of sea cucumber aestivation on marine surface sediment composition. The image was drawn using Figdraw (www.figdraw.com). ARGs: antibiotic resistance genes.

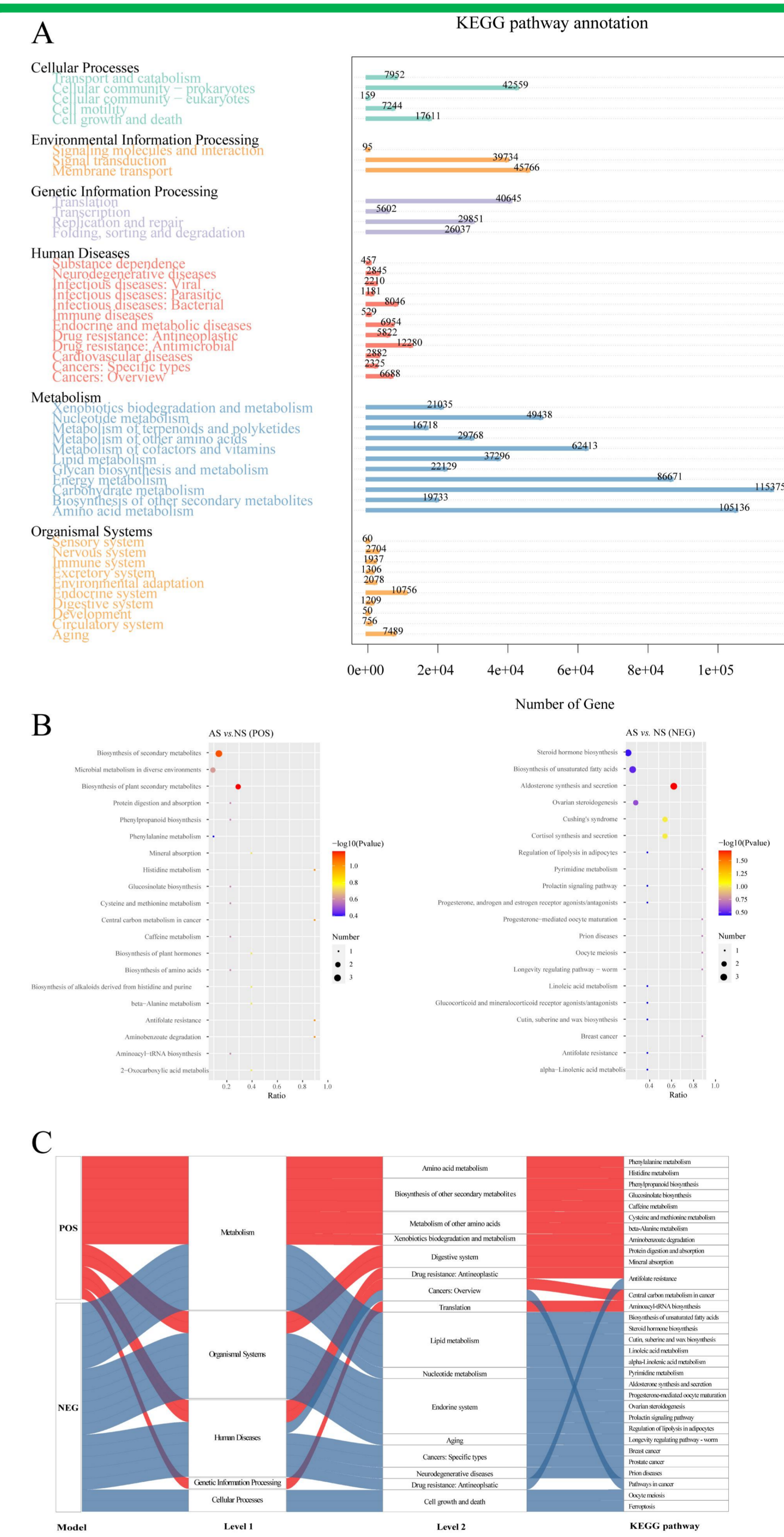


Fig. 6. Integrated metagenome-metabolome Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis. (A) KEGG pathway annotation of the metagenome. (B) Bubble diagram of the top 20 KEGG pathway enrichment of differentially expressed metabolites in the positive model. (C) Bubble diagram of the top 20 KEGG pathway enrichment of differentially expressed metabolites in the negative model. (D) Alluvial diagram of the same KEGG pathways in the metagenome and metabolome.

Conclusions

● Aestivation behavior might not alter the microbial composition of sediments but rather the relative abundances of some pathogenic and spoilage-related bacteria species.

● The upregulation of antibacterial metabolites induced by aestivation behavior might enhance the immune defense of aestivated sea cucumbers and reduce the dissemination of some ARGs (*tetW* and *floR*).

● The increased abundance of *tetM* indicates that this ARG might be considered a key indicator for monitoring and indicating ARG transfer or the sediment status of sea cucumber culture ponds during the aestivation period.

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