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**Review Article** 

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# Research Progress on Genomic Characteristics and Environmental Adaptability of *Eriocheir sinensis*

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## Abstract

*Eriocheir sinensis* is an important economic crustacean in China. Its unique migratory habits confer strong adaptability to environmental factors such as salinity and temperature. This paper reviews the genomic characteristics and environmental adaptability mechanisms of *E. sinensis*. Its 1.67 Gb genome contains 20,286 protein-coding genes, and significant expansions of gene families — such as F-ATPase, heat shock proteins (HSP), and carbonic anhydrase (CA) — are closely related to its environmental adaptation. In osmoregulation, ion transport genes such as Na+/K+-ATPase regulate ionic balance through gill tissues. Temperature adaptation involves the HSP family in maintaining protein homeostasis, supported by energy metabolism and antioxidant defenses. Acid-base balance regulation relies on carbonic anhydrase (CA)-catalyzed conversion of CO<sub>2</sub> to HCO<sub>3</sub>-, along with dynamic adjustments in metabolic pathways. Additionally, expansions of immune-related genes (such as Spätzle and Dscam) enhance disease resistance. Recent studies have also suggested that genomic repetitive sequences and transposable elements may play important roles in environmental adaptation, providing a theoretical foundation for understanding ecological adaptation mechanisms in crustaceans and informing stress-resistant breeding strategies.

Keywords: Eriocheir sinensis; genome; environmental adaptability; gene family expansion; osmoregulation; temperature adaptation; immune defense

# Introduction

Eriocheir sinensis, also known as the Chinese mitten crab, is one of the most economically valuable species in China's freshwater aquaculture, with its annual output accounting for a significant share of the national crustacean farming industry. As a typical catadromous species, E. sinensis has a life cycle that spans both freshwater and marine environments. Adult crabs breed in estuarine brackish water areas, while larvae (megalopae) migrate upstream to inland lakes for growth. This unique ecological habit has enabled the species to evolve exceptionally strong adaptability to environmental factors, including salinity, temperature, and dissolved

oxygen [1]. Its distribution covers major water systems in eastern China, from the Yangtze River basin to the Pearl River Delta, and from natural wetlands to artificial aquaculture ponds. It has even established wild populations in Europe, North America, and other regions as an invasive species, demonstrating remarkable environmental plasticity [2].

In recent years, breakthroughs in third-generation sequencing technologies and multi-omics analyses have led to landmark advances in the genomic research of *E. sinensis*. A chromosome-level genome map published in *Nature Communications* in 2021 revealed



that the 1.67 Gb genome of *E. sinensis* contains 20,286 protein-coding genes. Notably, significant expansions of gene families such as F-ATPase and heat shock proteins (HSP70) provide key clues for dissecting the molecular basis of its environmental adaptation [1]. These studies have not only revealed the Hox gene regulatory network underlying crustacean brachyurization (e.g., tail shortening), but also clarified the evolutionary pathways of adaptive traits such as osmoregulation and stress response through comparative genomics [3]. Current global climate change and water pollution pose severe challenges to aquaculture. Meanwhile, the survival mechanisms of *E. sinensis* under stresses such as high salinity, low temperature, and hypoxia have become research hotspots in genomics and evolutionary biology.

Studies have shown that antioxidant-related genes (such as thioredoxin TXN) in the E. sinensis genome are significantly upregulated during oxidative stress, and that ion transport genes such as Na $^+$ /K $^+$ -ATPase and F-ATPase work together to maintain osmotic homeostasis [4]. This paper systematically reviews the genomic structural characteristics of E. sinensis, the adaptive evolution of key functional gene families, and its molecular response mechanisms under environmental stresses such as osmotic pressure, temperature, and acid-base imbalance. These findings provide a theoretical basis for understanding the ecological adaptation strategies of crustaceans and for promoting stress-resistant breeding in aquaculture.

#### Genomic Characteristics of E. sinensis

## **Genome Size and Structure**

The genome size of *E. sinensis* is approximately 1.2-1.8 Gb, placing it among the larger genomes of crustaceans. An integrated analysis based on third-generation sequencing technologies (such as PacBio) and Hi-C chromosome conformation capture revealed a high-quality chromosome- level genome assembly of 1.67 Gb, with 94.4% coverage and 20,286 protein-coding genes [1]. Compared with other crustaceans, the number of protein-coding genes in E. sinensis is moderate. However, the expansion of certain gene families-such as F-ATPase and heat shock protein (HSP70)-is remarkable and closely associated with its environmental adaptability [1,5]. Repetitive sequences account for 40%-50% of the genome, with transposable elements (TEs) as the main component. Studies have found that PIF-Harbinger-like transposons are significantly expanded in the E. sinensis genome, and their expression patterns vary markedly under stresses such as air exposure, suggesting that TEs may contribute to environmental adaptation through genome rearrangement [2]. In addition, microsatellites and SNP loci are widely distributed in the genome, providing abundant markers for population genetic analysis and molecular breeding [6]. Chromosome collinearity analysis shows that the E. sinensis genome shares highly conserved chromosomal structures with species such as Portunus trituberculatus, especially in the Hox gene cluster, further confirming the evolutionary relationships among brachyuran crustaceans [7].

## **Gene Family Expansions and Contractions**

In addition to the F-ATPase and GST gene families, the expansion of heat shock protein (HSP) families in the  $\it E. sinensis$  genome is particularly notable. Fifty-six HSP genes were identified, including members of the HSP100, HSP70, and HSP60 families. Among these, HSP70 genes form multi-copy arrays through tandem duplication and are significantly upregulated under low temperature, starvation, and desiccation stresses, enhancing stress resistance by maintaining protein folding homeostasis [4]. The carbonic anhydrase (CA) gene family also shows adaptive expansion, with a total of 15 CA genes identified, distributed across 12 chromosomes. Notably,  $\it EsiCA7$  plays a dominant role in acid-base regulation by maintaining internal pH balance through catalyzing the interconversion of  $\it CO_2$  and  $\it HCO_3^-$ , facilitating adaptation to carbonic alkaline environments [3].

The expansion of immune-related gene families further enhances *E. sinensis*'s environmental adaptability. For example, three new members (EsSpz1-3) of the Spätzle gene family were identified; they induce antibacterial peptide synthesis by activating the Toll signaling pathway and play a key role in resisting White Spot Syndrome Virus (WSSV) infection [8]. Additionally, the Down syndrome cell adhesion molecule (Dscam) gene produces more than 50,000 protein variants through alternative splicing, significantly enhancing the ability to recognize diverse pathogens [9]. Notably, genes encoding enzymes related to the tricarboxylic acid (TCA) cycle, which is involved in energy metabolism, shows downregulated expression in gill tissues. This contraction of metabolic pathways may represent an adaptive strategy to cope with high alkali stress [10]. The dynamic evolution of these gene families collectively forms the molecular basis for *E. sinensis*'s response to complex environments.

#### Genome and Evolutionary Relationships

Through comparative analysis of the *E. sinensis* genome with those of other crustaceans, its evolutionary status and mechanisms of adaptive evolution have been systematically elucidated. Chromosome-level genome assembly reveals that *E. sinensis* shares highly conserved chromosomal collinearity with brachyuran crabs such as *Portunus trituberculatus*, particularly in the arrangement of the Hox gene cluster (e.g., *lab*, *ftz*, and *Abd-B*), confirming the presence of common ancestral features among brachyuran species during evolution [1,7]. Phylogenetic analysis further indicates that within crustacean evolution, Branchiopoda and Hexapoda form a sister group, whereas Malacostraca, as an independent lineage, diverged earlier than the split between Branchiopoda and Hexapoda—a conclusion supported by molecular clock analysis of 1,406 single-copy orthologous genes [5].

Unique transposable element (TE) expansion events in the *E. sinensis* genome further illuminate its adaptive evolutionary pathway. For example, PIF-Harbinger-like transposons account for 12.7% of the genome—significantly higher than in non-brachyuran crustaceans such as *Daphnia pulex*—and exhibit stress-specific expression activation under conditions such as air exposure, suggest-

ing their involvement in environmental adaptation through genome rearrangement [2]. In addition, tandem duplication of the F-ATPase gene family (expanded from 4 to 7 copies) and gene rearrangement in the mitochondrial genome endow E. sinensis with metabolic advantages in osmoregulation, representing specific adaptive evolutionary features in migratory crustaceans [1,11]. Evolutionary pressure analysis shows that the immune-related Dscam gene in E. sinensis has undergone positive selection (Ka/Ks = 1.23), and the high mutation rate in its extracellular variable region enables the production of more than 50,000 protein variants. This feature parallels the evolutionary diversification of vertebrate immunoglobulins, providing molecular evidence for adaptive evolution against pathogenic microorganisms [9]. These genomic characteristics collectively form the evolutionary foundation for the unique biological traits of E. sinensis, such as euryhaline adaptation from freshwater to seawater and limb regeneration.

# Environmental Adaptability Mechanisms Osmoregulation

E. sinensis is a euryhaline crustacean with a life history that involves migration from freshwater to seawater, thereby exhibiting strong osmoregulation capabilities. Genomic studies have shown that gene families related to ion transport, such as Na<sup>+</sup>/K<sup>+</sup>-ATPase and Cl- channels, have undergone expansion and adaptive evolution in its genome [1]. These genes are highly expressed in the gills and epithelial tissues, helping maintain internal osmotic balance by regulating ion absorption and excretion. In low-salinity environments, E. sinensis activates ion transport proteins in the gills, such as Na<sup>+</sup>/K<sup>+</sup>-ATPase, to absorb sodium and chloride ions from the surrounding water and compensate for internal ion loss [11]. In high-salinity environments, it maintains osmotic stability by excreting excess ions. Proteomic analysis shows that under high-salinity stress, the expression of energy metabolism-related proteins in the gills of *E. sinensis* is downregulated, while actin and other cytoskeletal proteins are upregulated—changes that may be associated with adjustments in cell morphology and function to adapt to high-salinity conditions [11].

## **Temperature Adaptation**

Temperature is a critical environmental factor influencing the growth, development, and reproduction of *E. sinensis*. This species can tolerate a wide range of temperatures, demonstrating adaptability to both low and high extremes. Genomic studies have revealed that its genome contains an abundant and diverse heat shock protein (HSP) gene family [4]. For example, researchers have identified 56 HSP genes in the E. sinensis genome, including members of the HSP100, HSP90, HSP70, HSP60, and small HSP families. These genes play essential roles in responding to temperature stress by facilitating proper protein folding and preventing protein aggregation, thereby maintaining normal cellular functions. For low-temperature adaptation, E. sinensis regulates energy metabolism and its antioxidant system to cope with cold stress. Transcriptome analysis shows that under low-temperature conditions, genes involved in glycolysis and fatty acid metabolism are upregulated to provide additional energy [12]. Meanwhile, the activities of antioxidant enzymes such as superoxide dismutase (SOD) and catalase (CAT)

are enhanced to reduce oxidative damage induced by cold exposure. Under high-temperature stress, the expression of HSP genes increases significantly, particularly *HSP70* and *HSP90*, which play key roles in cellular protection and the maintenance of protein homeostasis [13].

#### **Acid-Base Adaptation**

Changes in water pH have significant effects on the survival and physiological functions of E. sinensis. This species can tolerate a certain range of both acidic and alkaline conditions; a trait closely associated with acid-base regulatory genes in its genome. For instance, the carbonic anhydrase (CA) gene family plays a key role in maintaining acid-base balance in E. sinensis. Researchers have identified 15 CA genes in its genome, distributed across 12 chromosomes, among which the EsiCA7 gene may play a dominant role in acid-base regulation [3]. In acidic environments, E. sinensis upregulates the expression of CA genes to promote bicarbonate production and facilitate carbon dioxide excretion, thereby maintaining internal pH stability [3]. Meanwhile, the expression of genes involved in ammonia metabolism is also altered, reducing the accumulation of acidic substances by converting ammonia into urea or glutamine [10,14]. In alkaline environments, E. sinensis adapts to elevated pH levels by regulating ion transport and metabolic pathways, such as inhibiting the tricarboxylic acid (TCA) cycle [1,10].

#### **Immunity and Disease Resistance**

E. sinensis is exposed to a variety of pathogenic microorganisms in its natural environment, and its robust immune defense system is essential for survival and reproduction. Genomic studies have revealed the presence of complex immune-related gene families. For instance, gene families involved in the Toll-like receptor (TLR) signaling pathway, immunoglobulin-like receptors (IgRs), and antimicrobial peptides (AMPs) have undergone significant expansion in the E. sinensis genome [8,15]. Studies have identified multiple Spätzle genes in the E. sinensis genome. The proteins encoded by these genes serve as ligands for the Toll signaling pathway and play a crucial role in antiviral and antibacterial immunity [8]. Upon invasion by pathogenic microorganisms, the expression of Spätzle genes is upregulated, leading to activation of the Toll signaling pathway and induction of antibacterial peptide production. Additionally, E. sinensis possesses a unique Dscam gene that generates a large repertoire of receptor variants through alternative splicing, thereby enhancing its capacity to recognize diverse pathogens [9]. In response to fungal infections, a series of protein expression changes occur in the hemolymph of *E. sinensis*, including upregulation of cytoskeleton-associated proteins, serine proteases, and antioxidant enzymes, alongside downregulation of proteins involved in organ regeneration [16]. Collectively, these changes form the immune response network of E. sinensis against pathogen invasion.

#### **Adaptation to Other Environmental Stresses**

In addition to the aforementioned environmental factors, *E. sinensis* is also subjected to other stresses, such as pollutant exposure and hypoxia. Genomic studies have demonstrated that gene families associated with detoxification and antioxidation, including

cytochrome P450 enzymes (CYP) and glutathione S-transferases (GST), provide a molecular basis for coping with these challenges [17,18]. For instance, under prolonged exposure to the organophosphate pesticide trichlorfon, significant changes occur in the expression of metabolism- and detoxification-related genes in the hepatopancreas of E. sinensis, notably the upregulation of cytochrome P450 enzymes, glutathione S-transferases, and carboxylesterases [19]. These gene expression changes facilitate the metabolism and elimination of pesticide residues, thereby reducing toxic damage. In hypoxic environments, *E. sinensis* adapts to anoxic conditions by modulating energy metabolism and gene expression. Transcriptome analyses reveal that under hypoxic stress, genes involved in glycolysis and anaerobic respiration are upregulated, whereas those associated with aerobic respiration are downregulated [20]. Additionally, certain transcription factors related to hypoxia adaptation, such as homologs of hypoxia-inducible factor (HIF), may participate in this process, although research on HIF in crustaceans remains limited.

# **Conclusions and Prospects**

Significant progress has been made in research on the genomic characteristics and environmental adaptability mechanisms of E. sinensis, revealing the molecular foundations underlying its osmoregulation, temperature adaptation, acid-base balance, immune defense, and other physiological processes. The expansion and adaptive evolution of gene families such as F- ATPase, HSP, CA, and GST provide a genetic basis for its robust environmental resilience. However, many questions remain to be addressed. For instance, the dynamic changes in gene expression across different life stages of E. sinensis-such as larvae and adultsand the role of epigenetic regulation, including DNA methylation and histone modification, in environmental adaptation remain poorly understood [21]. Moreover, the interplay between the microbiome and E. sinensis environmental adaptation represents another promising area for in-depth investigation [22]. Future research should integrate multi-omics approaches—including genomics, transcriptomics, proteomics, and metabolomics—to comprehensively dissect the molecular regulatory networks of E. sinensis under various environmental stresses. Concurrently, functional validation using gene-editing technologies such as CRISPR/Cas9 will be instrumental in elucidating the specific roles of key genes in environmental adaptation [23]. These studies will not only deepen our understanding of the evolutionary and ecological adaptation mechanisms of *E. sinensis* but also provide a theoretical foundation for the aquaculture and conservation of crustaceans.

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