

Mud crab *glutamate dehydrogenase: molecular cloning, tissue expression and response to hyposmotic stress*

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Abstract The glutamate dehydrogenase (GDH) gene from the mud crab *Decapoda Scylla serrata* was cloned and characterized. The full-length cDNA sequence of *S. serrata* GDH (SsGDH) was 1,800 bp in length, encoding a protein of 600 amino acids. The deduced amino acid sequence of SsGDH showed a high homology with other crustacean GDHs. The expression of SsGDH in various tissues was analyzed by RT-PCR. The results showed that SsGDH was widely expressed in all tissues examined, with the highest expression in the muscle. The expression of SsGDH in the muscle was significantly up-regulated in response to hyposmotic stress, indicating that SsGDH may play an important role in the osmotic regulation of the mud crab.

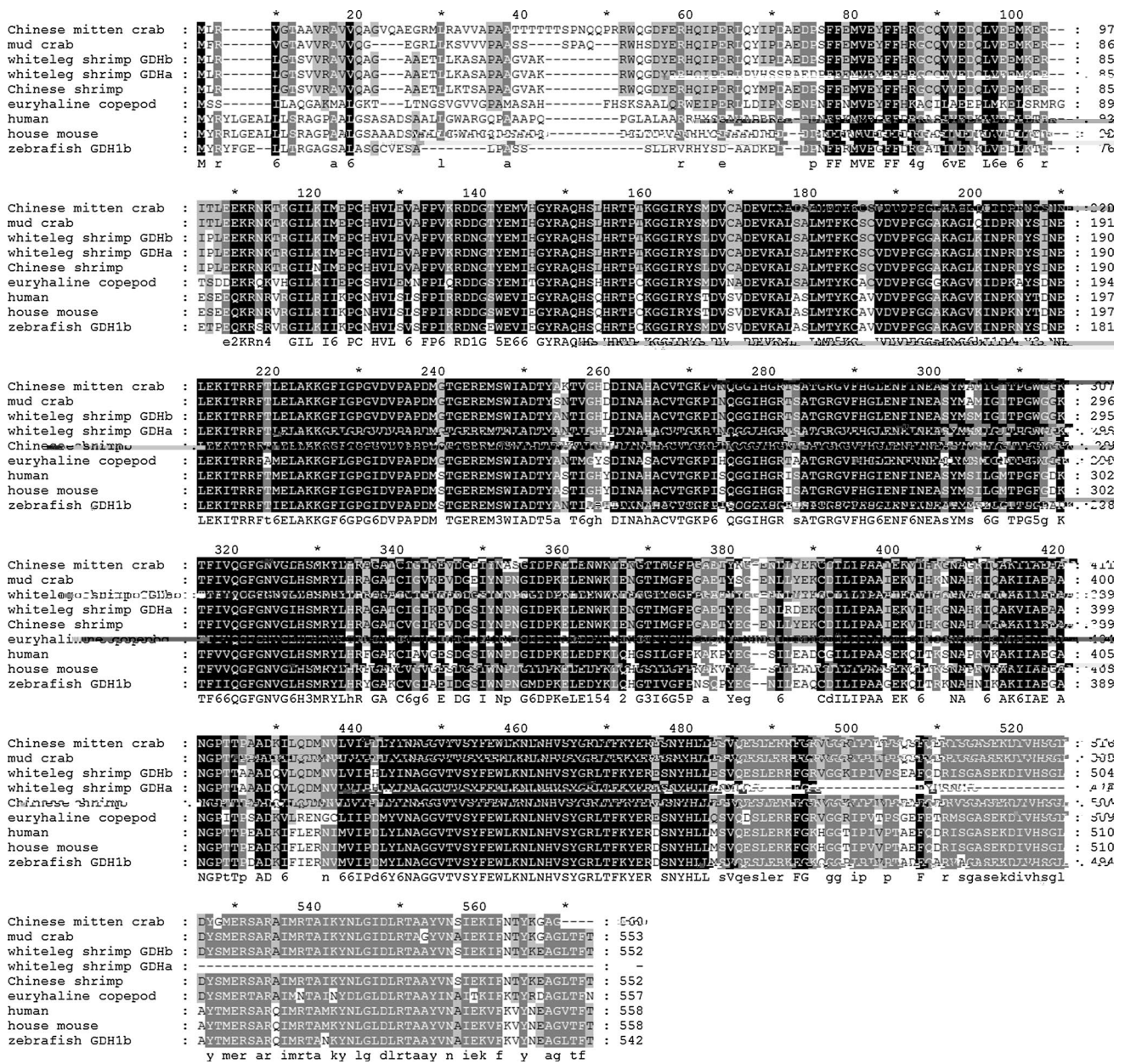


Fig. 3 Multiple sequence alignment of GDH protein from various species. The alignment is shown in blocks of 20 amino acids, with positions 20, 40, 60, 80, 100, 120, 140, 160, 180, 200, 220, 240, 260, 280, 300, 320, 340, 360, 380, 400, 420, 440, 460, 480, 500, 520, 540, and 560 marked. Species include Chinese mitten crab, mud crab, whiteleg shrimp, Chinese shrimp, euryhaline copepod, human, house mouse, and zebrafish. Conserved residues are indicated by asterisks (*). A consensus sequence is shown at the bottom of each block.

Chinese mitten crab GDH
mud crab GDH
whiteleg shrimp GDHb GDH
whiteleg shrimp GDHa GDH
Chinese shrimp GDH
euryhaline copepod GDH
human GDH
house mouse GDH
zebrafish GDH1b GDH

Chinese mitten crab GDH
mud crab GDH
whiteleg shrimp GDHb GDH
whiteleg shrimp GDHa GDH
Chinese shrimp GDH
euryhaline copepod GDH
human GDH
house mouse GDH
zebrafish GDH1b GDH

Chinese mitten crab GDH
mud crab GDH
whiteleg shrimp GDHb GDH
whiteleg shrimp GDHa GDH
Chinese shrimp GDH
euryhaline copepod GDH
human GDH
house mouse GDH
zebrafish GDH1b GDH

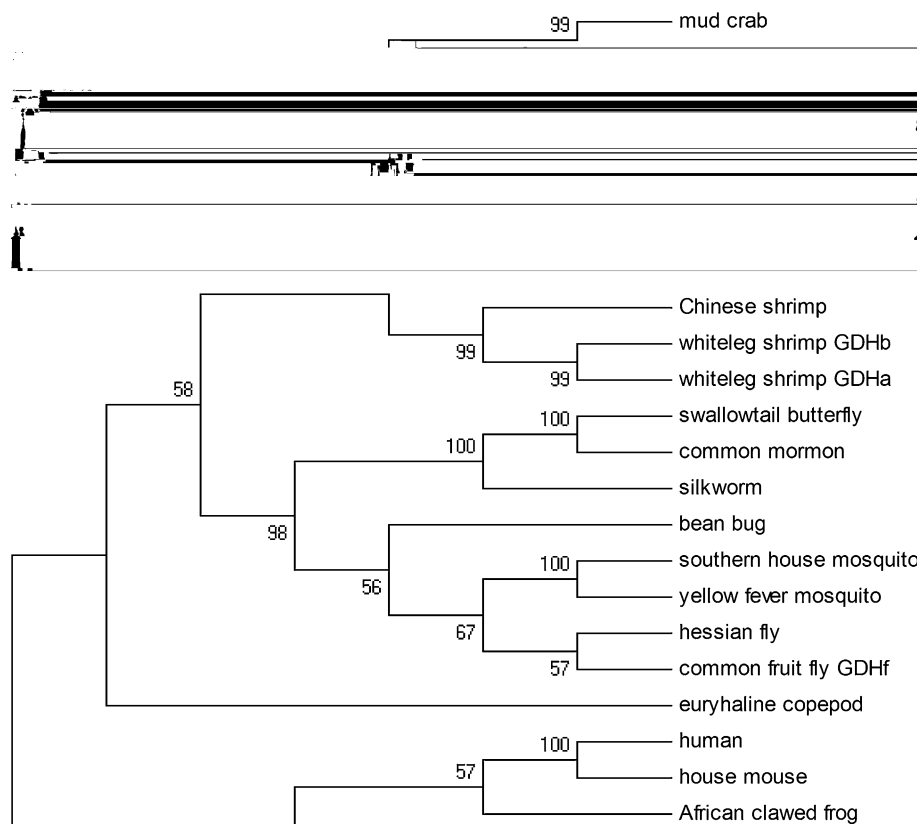


Fig. 4 *GDH* protein sequence alignment. The alignment shows conserved regions across species including *Scylla paramamosain*, *Xenopus laevis*, *Salmo salar*, *Riptortus pedestris*, *Eriocher sinensis*, *Fenneropenaeus chinensis*, *Papilio xuthus*, *Drosophila melanogaster*, *Misgurnus anguillicaudatus*, and *Tigriopus californicus*.

Mayetiola destructo, *Mus musculus*, *Oncorhynchus mykiss*, *Homo sapiens*, *Monopterus albus*, *Carassius auratus red var*, *Bombyx mori*, *Culex quinquefasciatus*, *S. paramamosain*, *Papilio xuthus*, *Litopenaeus vannamei*, *L. vannamei*, *Aedes aegypti*, and *Danio rerio*.

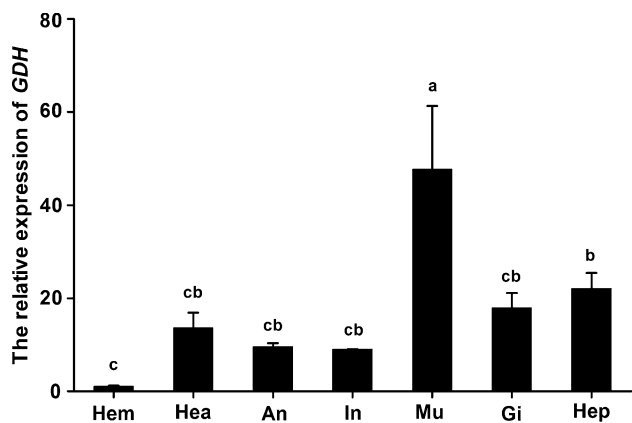
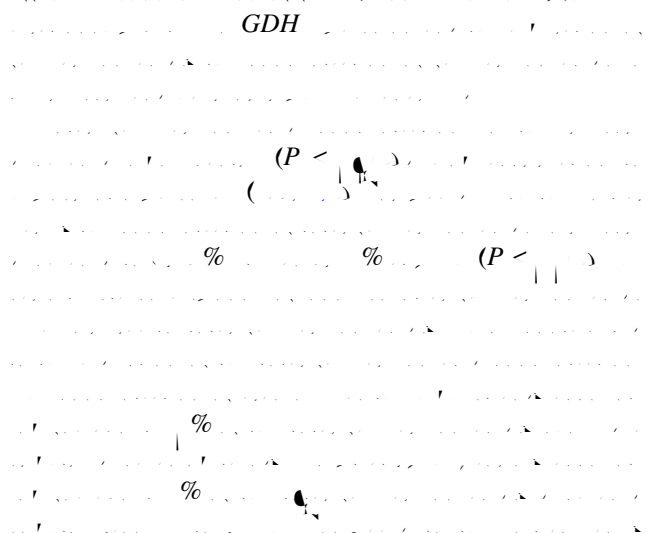


Fig. 5 *GDH* expression in different tissues of *Scylla paramamosain*. Hem: Hemolymph, Hea: Heart, An: Antenna, In: Intestine, Mu: Muscle, Gi: Gut, Hep: Hepatopancreas. Bars represent relative expression levels.



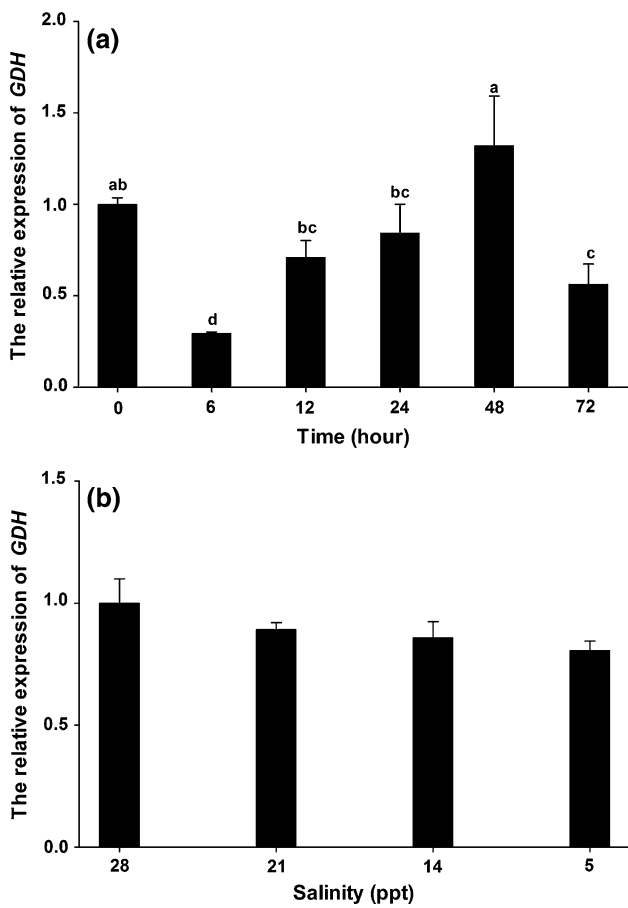


Fig. 6 a *GDH* expression in *Scylla paramamosain* b *GDH* expression in *Scylla paramamosain* H *Scylla paramamosain* Bars represent standard deviation. Different letters indicate significant differences ($P < 0.05$)

($P < 0.05$) ($P < 0.05$) H ($P < 0.05$)

Discussion

H H *S. paramamosain* *Scylla paramamosain* *Macrobrachium amazonicum* *L. vannamei*

Scylla serrata

GDH

GDH *GDH* *S. paramamosain*

Tigriopus californicus *L. vannamei* *E. sinensis*

GDH

Table 2 Percentages of *Scylla paramamosain* individuals with different genotypes (M and H) in the 10 sampling sites (M₁–M₁₀ and H₁–H₁₀)

Sampling site	M ₁ (%)	M ₂ (%)	M ₃ (%)	M ₄ (%)	M ₅ (%)	M ₆ (%)	M ₇ (%)	M ₈ (%)	M ₉ (%)	M ₁₀ (%)	H ₁ (%)	H ₂ (%)	H ₃ (%)	H ₄ (%)	H ₅ (%)	H ₆ (%)	H ₇ (%)	H ₈ (%)	H ₉ (%)	H ₁₀ (%)	
1	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
2	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
3	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
4	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
5	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
6	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
7	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
8	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
9	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
10	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100

... (P < ...)

...

L. vannamei

(P < ...)

(P < ...)

GDH

S. paramamosain

GDH

GDH

S. paramamosain

S. paramamosain

GDH

S. paramamosain

Acknowledgments

References

...

H M (...)

...

H M (...)

Dilocarcinus pagei (...) E E

M (...)

...

M (...)

H M (...)

Macrobrachium amazonicum

M. olfersi (...)

M (...)

M (...)

M (...)

Litopenaeus vannamei E M E

Litopenaeus vannamei

E (...)

+ K+ (...)

Litopenaeus vannamei E (...)

Litopenaeus vannamei E (...)

Litopenaeus vannamei

E (...)

H K M (...)

M (...)

Eriocheir sinensis

Scylla spp

(...)

Penaeus monodon (...)

H (...) M (...)

(M H) (...)

Scylla serrata E

H M (...) K

Litopenaeus vannamei

M (...)

M (...)

Litopenaeus vannamei M (...)

K H (...) K

Scylla paramamosain M E

Scylla paramamosain

