



## Preliminary evidence for snail deformation from a Eutrophic lake



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### ABSTRACT

The incidence of deformities in snails *Bellamya aeruginosa* was investigated in a typical eutrophicated lake – Taihu Lake. A total of 15 105 specimens were collected, and 0.18–0.93% of the snails exhibited abnormal tentacle bifurcations. Abnormally developed snails were all female and were found in regions with relatively high *Chlorophyll a* levels ( $12.40 \pm 7.23 \mu\text{g/L}$ ). As tentacles are sexually dimorphic in *B. aeruginosa*, we postulated that factors associated with eutrophication might be responsible for the partial masculinization of tentacles in females. Differential gene expression analyses revealed that a number of unigenes were significantly up-regulated or down-regulated in snails sampled from three locations having high *Chlorophyll a* levels compared with snails sampled from the region with lower *Chlorophyll a* level ( $2.95 \mu\text{g/L}$ ). Thus, transcriptomic profiling revealed potential molecular signal of eutrophication that can lead to developmental abnormalities in this species.

### 1. Introduction

Eutrophication has become a global concern in many freshwater, estuarine, and marine ecosystems over the last several decades, and is expected to continue due to the use of nitrogen and phosphorus fertilizers in agricultural production (Lafferty and Holt 2003). Consequently, the increasing frequency and intensity of algal blooms associated with eutrophication poses a serious risk to aquatic ecosystems due to the release of toxins from some algal species and overall declines in water quality. Thus, understanding the biological responses in aquatic organism inhabiting in eutrophicated environments is crucial for the performance of sagacious ecological risk assessments.

Benthic mollusks are used commonly as sensitive bioindicators of aquatic ecosystem pollution, and appear increasingly in global monitoring networks (Gagnaire et al., 2009; Gust et al., 2014). In addition to their use as sentinels in biological monitoring, freshwater snails are also used in hazard and risk assessments (Matthiessen 2008). However, compared with the abundant information on aquatic vertebrates, knowledge about the endocrinology and hormonal functions of invertebrates, especially mollusks, is very limited (LaFont 2000). Sequencing of the Pacific oyster *Crassostrea gigas* genome (Zhang et al.,

2012) has assisted in the identification of potential targets and modes of action of environmental pollutants (Vogeler et al., 2014). For example, the absence of an NR3A ortholog in the Pacific oyster suggested that environmental estrogens do not regulate gene expression through the NR3A pathway; whereas, the discovery of PPAR (NR1C) and RXR (NR2B) homologs opened novel windows for scientists to evaluate the effects of various synthetic chemicals on mollusks (Vogeler et al., 2014). However, the expression of a single gene does not explain the extent and complexity of the mechanisms in organisms exposed to environmental toxicants or stressors. The omics profiling of transcripts provides a powerful tool for discriminating classes of chemicals and understanding modes of action (Iguchi et al., 2006). Further, the transcriptome provides a far more precise measurement of transcript levels, and the numbers of sequence reads or assembled unigenes often vary by several orders of magnitude, reflecting the differential expression among samples (Francis et al., 2013). This makes it possible to identify specific groups of genes, rather than a single gene, which could capture the full mechanism of the molecular responses of an organism exposed to various chemicals or stressors (Chapman et al., 2011).

*Bellamya aeruginosa* is distributed widely across streams, ponds, reservoirs, and other freshwater bodies in East Asia. This species is an

**Abbreviations:** Vtg, Vitellogenin; PPAR, peroxisome proliferator-activated receptor delta-like; GPCR, G protein-couple receptor; RXR, retinoid X receptor; ERR, estrogen receptor-related receptor; RPL 7, ribosomal protein 7; ORF, open reading frame; RPKM, reads per kilobase per million mapped reads; nr, non-redundant dataset

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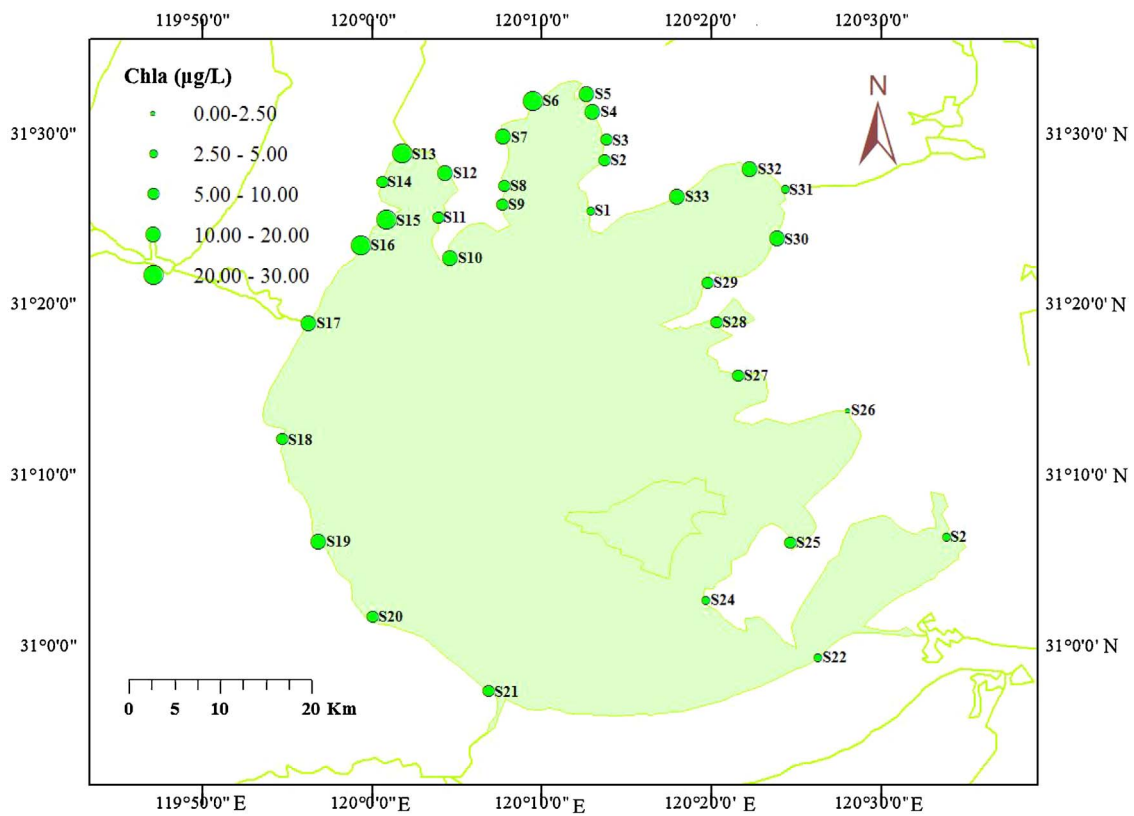


Fig. 1. Sampling sites (green blots) for investigating *Bellamya aeruginosa* along Taihu Lake. The size of green blots also indicated the *Chlorophyll a* level in these sampling sites during the investigation. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1

Water quality parameters and numbers of snails collected along Taihu Lake. Percentage of abnormalities represents the number of deformed/total number of females.

Site	Shoreline	Tem (°C)	Sal. (%)	Chl-a(µg/L)	DO (mg/L)	pH	Numbers of snails sampled		Deformed snails	
							Female	Male	Number	(%)
1	North	25.7	0.23	4.40	8.60	8.83	125	108	0	/
2		26.3	0.23	9.02	10.24	8.70	262	169	1	0.38
3		27.0	0.21	8.70	7.57	8.49	369	156	0	0
4		26.9	0.22	16.60	10.00	8.30	395	246	1	0.25
5		26.0	0.19	10.02	17.23	9.13	276	116	0	/
6		27.3	0.21	20.50	11.23	8.67	215	101	0	/
7		25.3	0.20	12.70	7.18	7.87	303	107	0	/
8		25.6	0.24	7.80	5.59	7.67	551	281	1	0.18
9		26.0	0.25	7.71	6.21	7.80	217	102	0	0
10		25.4	0.20	10.04	8.23	7.80	107	43	1	0.93
11		25.1	0.16	7.20	2.14	7.00	314	262	0	/
12	24.8	0.16	10.50	3.98	7.30	479	312	0	/	
13	24.3	0.18	28.20	5.85	7.60	545	296	1	0.18	
14	24.3	0.20	8.60	2.46	7.24	255	147	0	/	
15	24.5	0.20	29.70	2.86	7.19	375	82	0	/	
16	24.5	0.20	22.70	10.58	8.47	0	0	/	/	
29	26.5	0.24	5.96	8.16	7.84	275	134	0	/	
30	25.9	0.24	13.47	6.33	7.72	256	112	0	/	
31	25.6	0.26	3.87	7.71	8.18	364	169	0	/	
32	24.4	0.25	10.20	4.40	7.86	342	156	1	0.29	
33	23.7	0.26	12.70	0.44	7.40	221	162	0	/	
17	West	24.7	0.16	10.80	5.36	7.37	344	148	0	/
18		23.5	0.16	9.00	8.02	7.91	439	230	0	/
19	South	22.7	0.18	10.70	7.17	7.62	199	143	0	/
20		23.4	0.17	7.46	7.57	7.70	336	209	0	/
21		22.9	0.15	5.80	6.77	7.60	310	97	0	/
22		22.7	0.20	4.47	7.55	7.75	112	51	0	/
23	East	23.7	0.23	2.80	8.05	7.84	196	184	0	/
24		22.9	0.24	2.95	6.81	7.75	302	141	0	/
25		23.2	0.23	8.20	7.89	8.01	353	173	0	/
26		23.8	0.20	2.50	6.82	7.59	260	122	0	/
27		24.8	0.22	5.80	8.55	7.89	333	188	0	/
28		25.5	0.22	7.40	4.24	7.38	516	212	0	/

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