RESEARCH ARTICLE

Prevention of urea toxicity in DNA and RNA content of a fresh water field crab, *Barytelphusa guerini* by using Sulphur containing amino acid *Methionine* as an additive

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Manuscript details:

Received: 18.05.2016
Accepted: 28.06.2016
Published : 23.07.2016
Reprinted : 23.08.2016

Editor: Dr. Arvind Chavhan

Cite this article as:
Kulkarni Arvind Ratnakar Pravina and Balkhande Jayvardhan (2016) Prevention of urea toxicity in DNA and RNA content of a fresh water field crab, *barytelphusa guerini* by using sulphur containing amino acid *methionine* as an additive, *International J. of Life Sciences*, 4(2): 263-266.

Acknowledgement
First author thankful to S. R. T. M. U. Nanded for providing financial assistance in the form of Minor Research Project. Authors are very much thankful to Principal N. E. S. Science College, Nanded for continuous encouragement during the project work.

ABSTRACT

The objective of the present study was to investigate the prevention of toxicity of urea in DNA and RNA content of a fresh water field crab, *Barytelphusa guerini* by using sulphur containing amino acid Methionine as an additive. If fertilizer (urea) are used in high concentration than the concentration of urea above the tolerance limit is toxic to the aquatic animal including crab. In present study quantity of RNA was less than the DNA in both Male and Female. Sulphur containing amino acid Methionine an additive is useful to prevention of urea toxicity to a fresh water crab *Barytelphusa guerini*. This is the baseline work, more molecular work is needed for prevention of this type of toxicity in aquatic life.

Key words: Urea Toxicity, DNA, RNA, *Barytelphusa guerini*

INTRODUCTION

Water pollution is increasing day by day different pollutants like heavy metals, pesticide, antifouling agent, fertilizers etc and has adverse effect on growth and survival of aquatic animals. These pollutants directly indirectly enter in the body of aquatic animals and affect on them and disturb biochemical and physiological patterns of the animals. The heavy metals like Copper, Chromium, Iron etc causes maximum effects on non target organisms resulting in imbalance of the ecosystem (Maharajan *et. al*., 2012). Fertilizers pollute the aquatic ecosystem and affect the aquatic biota. Nitrogen pollution is a major problem today (Vidal *et. al*, 2000; Haygarth and Jarvis, 2002; Randall and Tsui, 2002; Yadav *et. al*, 2007).

Crabs constitute a significant portion of the freshwater ecosystem. Very often they become the victim of fertilizers used in agriculture. Therefore their population in this area was found decreasing during the last decade. Urea which is used as fertilizer and protein for ruminant animals enters in the aquatic system from various ways. It accumulates in the soil and pollutes the water which affects the flora and fauna. Urea ulter the

biochemical and physiological pattern at cellular level of aquatic animals including crab. The present work was undertaken to study the toxic effect of urea on DNA and RNA content of fresh water field crab, *Barytelphusa guerini* and prevention of toxicity by using additives. This will provide the scientific information for aquaculturists to minimize the effect by using sulphur containing amino acid *Methionine* as an additive.

MATERIALS AND METHODS

Fresh water field crab, *Barytelphusa guerini* collected from river Godavari near Nanded, and brought to laboratory for acclimatization and kept in an aerated plastic pools for 8 to 10 days. A layer of sand 4 to 5 inches was spreads over the bottom and water was added in it, during this period they were fed with dry prawn powder. Uneaten food and dead animals were removed. After 8 days animals were exposed to various concentrations of urea ranging from 5 to 40 g/ L to estimate LC_{50} value for 96 hrs. The container was maintained by changing the old water and adding new fresh water after every 24 hrs exposure period and LC_{50} value 96 hrs was calculated by using graphical method of Litchfield and Willcoxon (1949) and it was 25 gm/ L. Then animals were exposed to 3 different sets for 96 hrs. First set was control (without urea), second set was with sub lethal concentration of urea (10 mg / L) and third with urea and additive *Methionine* (1 mg/ L). After 24 hrs and 96 hrs of exposure period, animals were sacrificed to estimate DNA and RNA from Hapatopancreas. DNA was estimated by using Diphenylamine whereas RNA was estimated by using Oricinol as suggested by Jayaraman (1996).

RESULTS

After exposure to urea (10 mg/L) for 24 and 96 hrs. Hapatopancreas were removed for the estimation of DNA and RNA. Obtained Results are shown in Table and Graph No. 1 and No. 2. Quantity of RNA was less than the DNA in both Male and Female. RNA and DNA found decreased when compared with control. In third set (10 mg/L urea with additive *Methionine*) both RNA and DNA were increased than the exposed animals (10 mg/L).

Table 1: Estimation of DNA content in Hapatopancreas of urea treated and urea + additive treated crab, *Barytelphusa guerini* (µg/mg).

Set No.	Concentration	24 Hours		96 Hours	
		Male	Female	Male	Female
1	Control	2271.26	1589.87	2244.13	2044.13
2	Urea 10 gm/L	1362.74	1358.74	1190.82	1148.82
3	Urea 10 gm/L + *Methionine* 1 mg/L	**1444.13**	**1472.40**	**1271.26**	**1284.87**

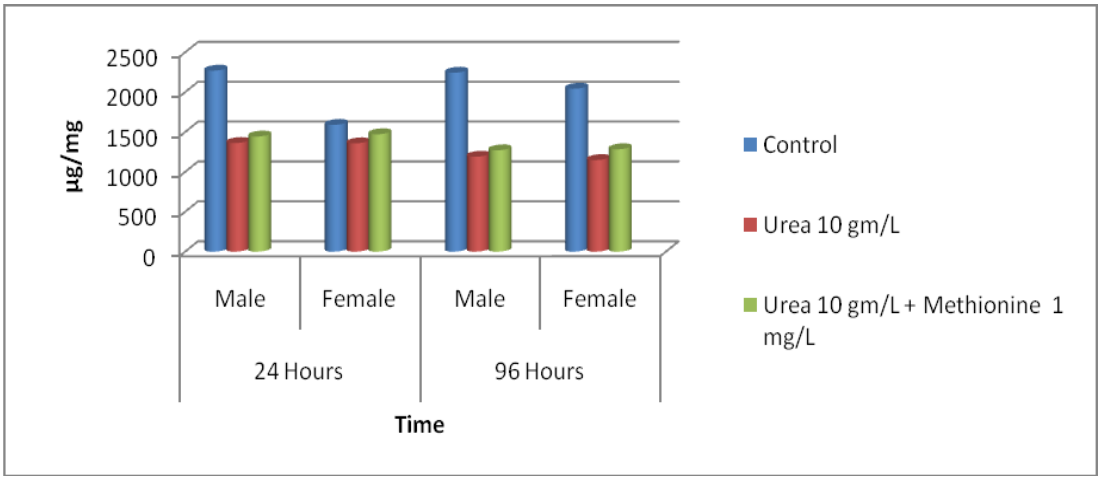

Fig. 1: Estimation of DNA content in Hapatopancreas of urea treated and urea + additive treated crab, *Barytelphusa guerini* (µg/mg).

Table 2: Estimation of RNA content in Hapatopancreas of urea treated and urea + additive treated crab, *Barytelphusa guerini* (µg/mg).

Set No.	Concentration	24 Hours		96 Hours	
		Male	Female	Male	Female
1	Control	321.35	437.97	147.95	173.96
2	Urea 10 gm/L	80.16	106.96	73.46	100.26
3	Urea 10 gm/L + *Methionine* 1 mg/L	**132.42**	**153.86**	**83.46**	**108.75**

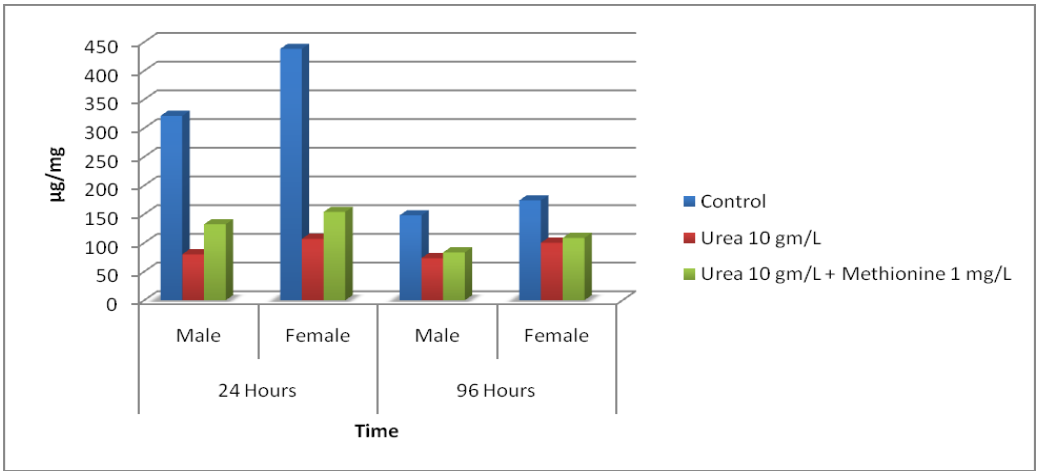

Fig. 2: Estimation of RNA content in Hapatopancreas of urea treated and urea + additive treated crab, *Barytelphusa guerini* (µg/mg).

Ravikiran and Kulkarni (2012) observed DNA and RNA content decreasing in different tissues of fresh water fish *Notopterus notopterus* when exposed to $CuSo_4$ and this may be due to the stress condition induced by Copper sulphate. Asfia and Vasanta (1985) observed depletion of RNA and DNA in tissue of Endosulfan treated fresh water fish, *Clarias batrachus*. In Present study decrease in RNA and DNA in Hapatopancreas of crab was due to stress condition of urea whereas RNA and DNA were found increase in additive treated crab.

The freshwater environment is going to be polluted by various pollutants which have adverse effects on aquatic organisms. The freshwater organism such as crabs is susceptible to these pollutants. Since, their habitats are confined and escape from such polluted habitats is impossible.

Thus it was concluded that sulphur containing amino acid *Methionine* an additive is useful to prevention of urea toxicity to a fresh water crab *Barytelphusa guerini*. This study can be useful to reduce the toxicity of urea at cellular level. As the urea destabilizing DNA and RNA content, we observed decreased in DNA and RNA content whereas sulphur containing amino acid *Methionine* inhibits destabilizing of DNA and RNA. Hence in *Methionine* treated animal recovery of DNA and RNA composition at the cellular level was observed. This is the preliminary work hence more work should be focused on this track and which will helpful to reduce the toxicity among all aquatic animals.

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