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Effect of biochemical alterations in DNA content of gill and gonad tissues of *Lamellidens marginalis* in Godavari River due to 5-fluorouracil toxicity

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Abstract
The present research paper deals with the study of the effect of biochemical alterations in DNA content of gill and gonad tissues of the freshwater bivalve *Lamellidens marginalis*. Samples were collected from the Godavari river at Pravara Sangam district Ahmednagar, Maharashtra. The study of DNA contents of gill and gonad tissues in *L. marginalis* shows significant decrease in Gill from 4.38 ±0.145 to 3.61 ± 0.246 for 15th day and on 30th day, there is significant decrease from 3.61 ± 0.451 to 2.86 ± 0.351 whereas Gonad shows significant decrease from 3.87 ±0.246 to 3.21 ±0.126on 15th days and on 30th days there is a significant decrease from 3.54 ±0.369 to 2.52 ±0.214, this is due to toxicity of 5- Fluorouracil and mobilization of tissue in the metabolism.

Keywords: DNA content, *Lamellidens marginalis*, 5- Fluorouracil

Introduction
Godavari River is an important river in India. It is second longest river in the country measuring 1465Km long and flows from western to southern India. Pravara Sangam is located 65 kms north of Ahmednagar. In last few decades increase in population density, heavy industrialization and agricultural activities have resulted in more and more waste entering in river. Contamination of fresh water with a wide range of pollutants has become a matter of concern over last few decades. (Vutukuru, 2005) [9]. Heavy metals have devastating effects on ecological balance environment and diversity of aquatic organisms. In order to evaluate the adverse effect of the pollutants on aquatic organisms, there is a worldwide trends to complement physical and chemical parameters with bio markers in aquatic pollution monitoring. (Abdel *et al*. 2012) [1]. Bivalves are used in monitoring programmes due to their ability to concentrate pollutants to several orders of magnitude above ambient levels in sea water. Biochemical modulation is a special type of combination chemotherapy which aims to selectively improve the therapeutic index by increasing the antitumor effect and protecting against toxic side effects. In the past decade a number of biochemical modulation approaches have been tested to improve the activity of 5-fluorouracil. 5FU itself has only modest anticancer activity but has been shown to be a very attractive target for biochemical modulation. The main conclusion is that properly applied biochemical modulation schedules may lead to successful use in the clinic (Peter, 1991) [5]. Excess of one of the deoxyribonucleotide precursors increases the frequency of miscorporation of that deoxyribonucleotide and inhibits the proof reading mechanisms. Increase in the sensitivity to alkylating DNA damaging agents has been observed when the deoxyribonucleotide pools are unbalanced (Phear *et al*. 1987) [6].

It has been observed that heavy metals can cause biochemical alterations such as inhibition of enzymes, metabolic disorder, genetic damage, hypertension and cancer. (Underwood, 1971; Lucky and Venugopal, 1977) [8, 3]. Effects of heavy metals on gills showed lamellar degeneration, epithelial lifting and necrotic changes in epithelial cells. The gills of heavy metal expose group show some epithelial lesions when exposed to heavy metals. The nephrotoxicity, ototoxicity and neurotoxicity of 5- fluorouracil may be due to reactions with cellular molecules other than DNA. The rate of replicative DNA synthesis was unexpectedly increased and the deoxyribonucleotide pools unbalanced (Skog, *et al.,* 1994) [7].

Materials and Methods
Attempts will be made in this study to select Fresh water bivalves, *Lamellidens marginalis* were collected from Godavari river at Pravara Sangam which is about at the distance of 65 kms away from Ahmednagarr City of Maharashtra state. First they are made acclimatized to laboratory conditions and they are washed. The water in the aquarium was changed regularly after every 24 hours. After the acclimatization, bivalves, *Lamellidens marginalis* were divided into two groups with equal numbers of animals. They were kept in separate aquarium for 15 and 30 days. Out of remaining one groups treated by chronic Concentration ($LC_{50/10}$ value of 96 hrs.) of 5-flurouracil (3.716 ppm) on 15th and 30th day of exposure, bivalves from each experimental group were sacrificed and gills, and gonads, were removed. These tissues were dried in oven at 75 ^{0}C to 80 ^{0}C till constant weight was obtained and blended into dry powder. These powders were used for the estimation of biochemical components of DNA to observe Efficacy of 5- fluorouracil. All the precautions recommended by ATSDR, 2003 to minimize risks of sample contamination were followed during collection and treatment.

Results and Discussion

Histopathological changes after 5- fluorouracil exposes are anticipatory in nature. These abnormalities results physiological dis function in the tissues. The nephrotoxicity, ototoxicity and neurotoxicity of 5- fluorouracil may be due to reactions with cellular molecules other than DNA. The Experiment has concluded that the result obtained on 15 & 30 days of gills and gonads with 5- fluorouracil were as follows.

Table 1: Alterations in the DNA content mg/100mg dry weight+ S.E. in gill and gonad tissues of *Lamellidens marginalis* treatment with 5-fluorouracil.

Sr. No.	Tissues	Days	Control	Experimental	Student 't' test 'p' value	% decrease (-)
1	Gill	15	4.38±0.145	3.61± 0.246	P < 0.01	17.58%
		30	3.61±0.451	2.86±0.351	P < 0.01	20.78 %
2	Gonad	15	3.87±0.246	3.21±0.126	P < 0.01	17.06%
		30	3.54±0.369	2.52± 0.214	P < 0.001	28.82 %

Values are expressed as mg/100mg dry weight.
± indicates S. D. of five observations
(-) indicate %decrease over control

15 Days treatment period (Subchronic)

The gill and gonad of *Lamellidens marginalis* shows a significant (P < 0.01) decrease. The gill shows control 4.38±0.145 to treated with 5- Fluorouracil *3.61 ± 0.246* mg/100g wet tissues in treated. The total DNA content in gill shows a decrease by 17.58%. The profile of total DNA content in gonad shows significant decrease from 3.87±0.246 to 3.21±0.126 mg/100 g wet tissues in gonad respectively. The gonad shows a decrease 17.06%. In both cases significantly decreases is recorded.

30 Days treatment period (Chronic)

The total DNA content of gill and gonad in control *Lamellidens marginalis* has been assessed. The result obtained 30th day after treating gills with the dose of 5- Fluorouracil has shown significant decrease from 3.61±0.451to 2.86±0.351 mg/100g wet tissues of gills i.e. 20.78%. Secondly the result of gonad treated with 5-fluorouracil is significant decreases 3.54±0.369 to 2.52± 0.214**mg/g wet tissues i.e. 28.82%. similarly 15 days subchronic exposure of treatment as compare to 30 days exposure shown that are both tissues reveals significant depletion in DNA content. In the present comparative study gill and Gonad shows significant elevation in to the DNA level, depletion in the exposure periods. 5-fluorouracil remains the mainstay of treatment for advanced gastric cancer (AGC) and no standard chemotherapy regimen exists. Combinations of irinotecan with folinic acid and infusional 5-fluorouracil (5-FU) (ILF) have shown good efficacy with acceptable toxicity as hematologic toxicity (anemia, neutropenia and leucopenia), Non-hematologic (nausea/vomiting) and diarrhea in patients with metastatic colorectal cancer (Kunz, 1998). Unbalanced supply deoxyribonucleotide triphosphates affect DNA replication and give rise to a range of genetic effects such as mutations, mitotic recombination's, chromosome aberrations and DNA strand breakage (Memuth, 1989).

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