



## Role of L-ascorbic acid in the protection of the hepatopancreas of an experimental model, fresh water bivalve, *Lamellidens marginallis* against the methomyl induced alterations

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### ABSTRACT

Excessive use of pesticides resulted in serious ecological and environmental problems as well as health hazards. Histopathological changes are mostly confined to organs directly involved in their metabolism and detoxification. In Molluscs, hepatopancreas is the main site of action, degradation and detoxification of pesticide, hence hepatopancreas is chosen as test organs. Ascorbic acid is main dietary antioxidant. For different physiological acts vitamins are essential, although required in trace amount. Therefore, the present study was conducted to evaluate the effectiveness of L-ascorbic acid in Methomyl -induced toxicity in an experimental model, the fresh water bivalve, *Lamellidens marginallis*. The animals were exposed to acute dose (35 PPM LC 50/2 value of 96 h) of Methomyl, Methomyl in same dose along with 50 mg/L of L- ascorbic acid and Methomyl in same dose along with 100mg/L of L-ascorbic acid for 96 hours. Control animals were maintained in normal water. Due to Methomyl intoxication damage to the hepatopancreas was extensive resulting epithelial hyperplasia, necrotic changes in basement membrane and intertubular connective tissue at 24 hours of exposure. The severity of damage progressed with longer exposure, after 96 hours of exposure to Methomyl, swelling, rupture and flattening of epithelial cells along with displacement of nuclei and widening of tubular lumen were noted. Exposure to Methomyl in combination with 50 mg/l of L-ascorbic acid showed considerable reduction in nature of damage, while with 100 mg/l of L-ascorbic acid showed almost normal histological structure of hepatopancreas.

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### Introduction

Pollution an undesirable changes in the physical, chemical and biological characteristic of air, water and soil, which is harmful to living organisms. Whatever may be the mode of contamination polluted environment is not suitable for existing life forms. The use of various classes of pesticides as organophosphate, organochlorine, carbamate and pyrethroids have been increased many fold for the last 10 years (Wolansky et al., 2006). Out of these Organophosphates Pesticides (OP's) have been widely used to control agricultural pests but are frequently harmful to non target aquatic organisms due to contamination of the aquatic environment by drainage from agricultural areas (Roche et. al., 2001-2007; Joseph and Raj, 2011). Methomyl is one of the class of chemicals called Carbamates and its trade name includes Lannate, Lanox, Methavin and Nudrin. It is classified as Restricted Use Pesticide (by Environmental Protection Agency) because of its acute toxicity to humans. Methomyl is highly toxic to aquatic invertebrates, when absorbed through the mucous membrane of the respiratory tract, resulting in systemic intoxication.

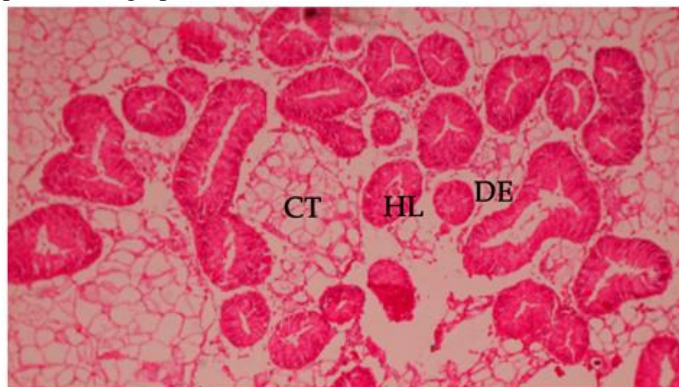
### Materials and methods:

Medium sized healthy, live freshwater bivalve, *Lamellidens marginallis* were acclimatized for a week to dechlorinated tap water. The animals were exposed to acute dose (35 PPM LC 50/2 value of 96 h) of methomyl, Methomyl in same dose along with 50 mg/L and 100mg/L of L-ascorbic acid for 96 h. Control animals were maintained in normal water. During experimentation, animals were fed on fresh water algae and

every day solutions were changed. After 24 and 96 h of exposure, animals were dissected, hepatopancreas were fixed in aqueous Bouin's fluid for 24 h, processed by usual way, serial sections were cut at six micron thickness and were stained with haematoxyline-eosine stain.

### Results:

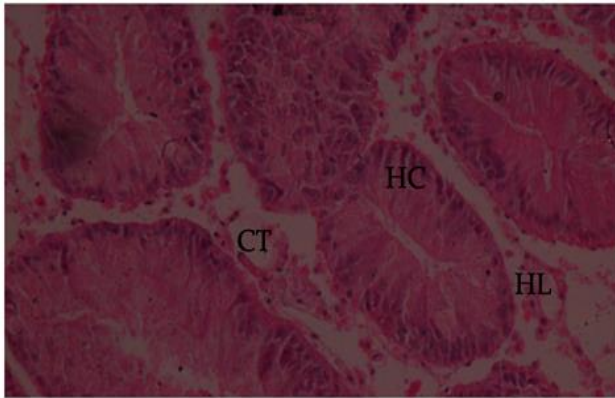
The gross histopathological effects of acute dose of Methomyl alone and with 50mg/L and 100 mg/L of L-ascorbic acid and recovery responses studied in an experimental model fresh water bivalve, *Lamellidens marginallis* are shown in photomicrographs 1-4.



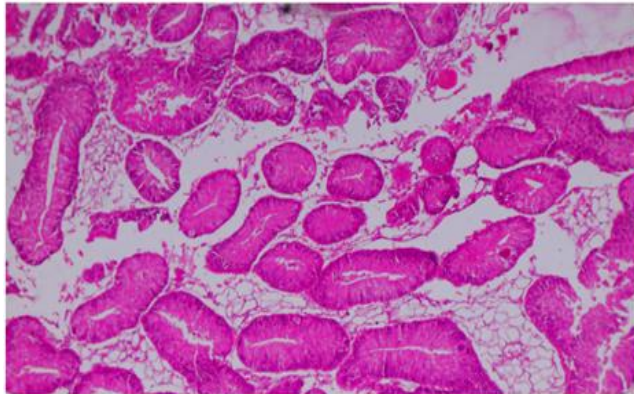
Photomicrograph of Digestive Gland of *Lamellidens marginallis* in control at 96 hrs.[10X]

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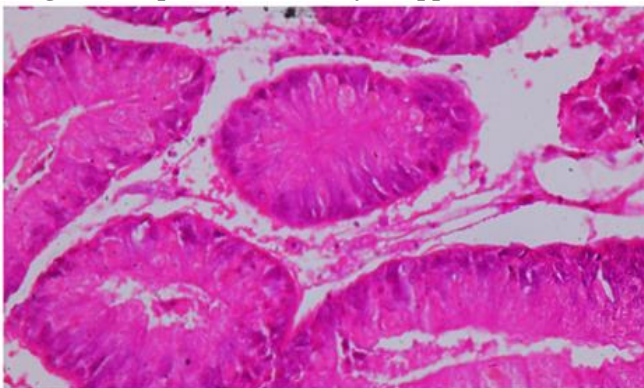
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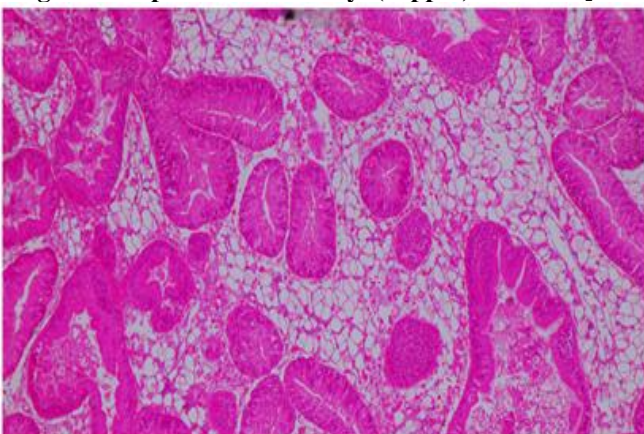
Photomicrograph of Digestive Gland of Lamellidens marginallis in control at 96 hrs.[40X]



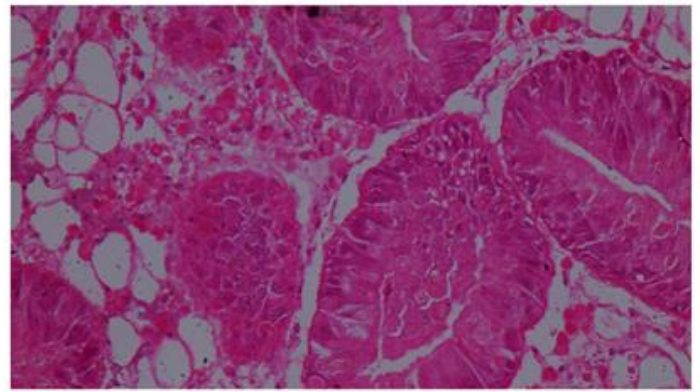
Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) at 96 hrs.[40X]



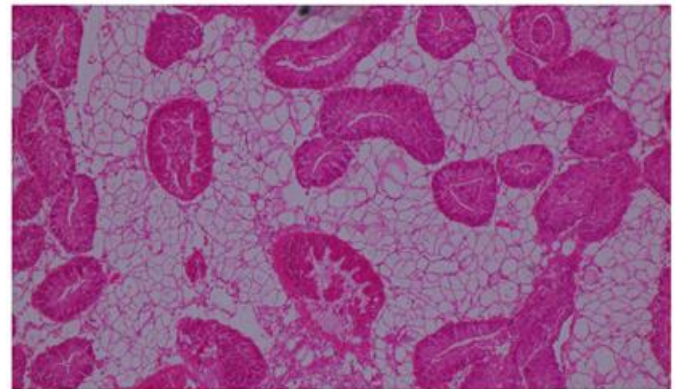
Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) at 96 hrs.[100X]



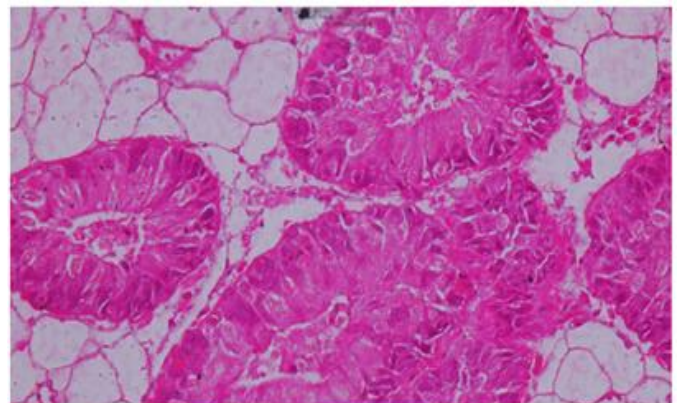
Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) along with 50mg/l. Ascorbic acid at 96 hrs.[40X]



Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) along with 50mg/l. Ascorbic acid at 96 hrs.[100X]



Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) along with 100 mg/l. Ascorbic acid at 96 hrs.(40X)



Photomicrograph of Digestive Gland of Lamellidens marginallis exposed to Methomyl (35ppm) along with 100 mg/l. Ascorbic acid at 96 hrs.(100X)

#### Histology of the normal hepatopancreas of *Lamellidens marginallis*:

Hepatopancreas is composed of tubules, having different shapes and sizes, and are surrounded by the intertubular connective tissues with some muscles, collagen fibres and amoebocytes. The epithelium of the tubules is placed on a thin basement membrane. The epithelial cells have basally situated nuclei.

#### Hepatopancreas under Methomyl intoxication:

Histopathological alterations were recorded in the hepatopancreas of fresh water bivalve *Lamellidens marginallis* after acute exposure to Methomyl alone & in combination with 50mg/l & 100mg/l of L-ascorbic acid. Due to Methomyl intoxication damage to the hepatopancreas was extensive resulting epithelial hyperplasia necrotic changes in basement membrane and intertubular connective tissue at 24 hours of

exposure. The severity of damage progressed with longer exposure, after 96 hours of exposure to Methomyl swelling, rupture and flattening of epithelial cells along with displacement of nuclei and widening of tubular lumen were noted. Exposure to Methomyl in combination with 50 mg/l of L-ascorbic acid showed considerable reduction in nature of damage, while with 100 mg/l of L-ascorbic acid showed almost normal histological structure of hepatopancreas. In present study on combined exposure to acute dose of Methomyl along with 50mg/L and 100 mg/L of L-ascorbic acid showed reduction in damages, indicating the protective effect of L-ascorbic acid. Thus the result of the present study clearly demonstrates protective ability of ascorbic acid against pesticide toxicity. Waykar and Tambe (2011) also reported mitigation of cypermethrin induced histological damage in the hepatopancreas of freshwater bivalve, *Parreysia cylindrica* by L-ascorbic acid.

#### Discussion:

Pesticides possibly affect all the body parts of the exposed animals either physiologically or by inducing histopathological changes. Once the pesticides enter the body, the animal tries to metabolise it, so that it is thrown outside the body to detoxify its effect, but where the pesticides could not be metabolised, animals may adopt their ways of detoxification. The pesticidal stress reflects in the cytoarchitecture which is greatly affected due to diffusion of pesticides in the cells. Hepatopancreas is one of the important organ in the body of bivalves. Disintegration or partial damage of exocrine and endocrine portion of hepatopancreas of teleost treated with a variety of pesticides is documented. The histopathological changes as observed on Methomyl exposure were reported after exposure to different pesticides (Thoser et al., 2001; Omiamia, 2004; Waykar, 2006; Saraswathy et al., 2010; Andhale et.al., 2011). Many workers (Amminikutty and Rege, 1977; Clements et.al., 1980; Akarte et.al., 1987; Srivastawa and Maurya 1991; Pillai and Menon, 1998; Shaikh et.al.,2012) studied the effect of different toxicants on the hepatopancreas of various aquatic animals. Probable cause of epithelial damage in most of the pesticide poisoning cases is the destruction of the basement membrane which mainly contains collagen. The reason for collagen damage can be the decrease in ascorbic acid content during pesticide exposure. Number of workers has reported that ascorbic acid content is decreased due to pesticide intoxication (Muley and Mane, 1987; Jadhav et. al; 1996;Waykar and Lomte, 2004). Ascorbic acid also plays a very major role in tissue synthesis and growth process and obviously mediates rapid tissue repair in trauma or disease condition (Halver, 1972).

#### Conclusion

It has also been observed that digestive tubule accumulate maximum pollutants which might be responsible for histopathological alterations. Such damages in the digestive tubule of *Lamellidens marginalis* due to methomyl, disturbs its normal function like secretion, absorption and storage of nutrient materials. These parameters could be used as biomarkers for the assessment of actual health of the organisms living in the polluted aquatic environment.

#### References

**Akarte, S.R. and Mane, U.H. (1988).** Toxicity of folithion to three freshwater bivalve mollusks in different seasons. *J. Envnt. Biol.* **9**, 27.  
**Amminikutty, C.K and M.S. Rege: (1977):** Effects of acute and chronic exposure to pesticides, thiodan 35 EC and agallol 3

on the liver of widowwrtetra, *Gymnocorymbus ternetzi* Boulenger. *Ind. J. Exp. Biol.*, **15**, 197-200.

**Andhale, A. V.,Bhosale P.A and Zambare S.P.,2011:** Histopathological study of nickel induced alterations in the fresh water bivalve, *Lamellidens marginalis* Journal of Experimental Sciences Vol. 2, Issue 4, Pages 01-03 .www.jexpsciences.com

**Clement, R., Gardner, R. C., & Smythe, P. C. (1980).** Social and individual factors in second language acquisition. *Canadian Journal of Behavioural Science*, **12**, 293-302.

**Halver, J. E. (1972):** The role of ascorbic acid in fish disease and tissue repair. *Bull. Jap. Soc. Sci. Fisheries.* **35**(1): 79-92.

**Jadhav, S., Sontakke, Y.B. and Lomte, V.S. (1996):** Effect of carbaryl on ascorbic acid content in the selected tissues of *Corbicula striatela*. *J. Ecotoxicology. Env. Monit.* **6**(2): 109-112.

**Joseph, B and Raj S. J. (2011):** Impact of pesticide toxicity on selected biomarkers in fishes. *International Journal of Zoological Research.* Vol. 7 issue 2, 212-222.

**Muley, D.V and Mane, U.H. (1987):** Sublethal effect of mercuric chloride on the tissue composition of bivalve mollusc, *Lamellidens marginalis*. *Biological Bulletin of India*, **9**, 31: 40.

**Omiamia, S. E. (2004):** Protective Effect of vitamin C and glutathione against the histopathological changes induced by imidacloprid in the liver and testis of Japanese quail. *The Egyptian J. Hospital Medicine* Vol. **16**: 39 – 54.

**Roche, S. M. ,Jones, A. Q., Majowicz, S. E., McEwen S. A and Pintar, K. D. M. (2001–2007):** Drinking water consumption patterns in Canadian communities *Journal of Water and Health* Vol 10 No 1 pp 69–86 © IWA Publishing 2012 doi:10.2166/wh.2011.051

**Saraswathy, G. R., Maheswari, E. and Thakur, S. (2010).** Effect of Vitamin C Supplementation on Phenytoin Induced Hepatototoxicity. *Global J. Pharmacology.* **4**(3): 127-135.

**Shaikh Yasmeen, Suryawanshi, G.D. and Mane, U.H. (2012):** Histopathological changes in the hepatopancreas of *Lamellidens marginalis* exposed to cadmium in winter. *International Multidisciplinary Research Journal* 2012, **2**(6):41-43 ISSN: 2231-6302. Online: <http://irjrs.info/>

**Shrivastava, V. M .S. and Maurya, R. S. (1991):** Effect of chromium Stress on gill and intestine of *Mystus vittatus* (Block) scanning E. M. Study *J. Ecobiology.* **3**: (1) 59-71.

**Thoser, M. R., Huilgol, N. V. and Lonkar, A. N.( 2001):** Observation on the changes in the structure of hepatopancreas in the fresh water gastropod snail *V. bengalensis*, exposed to sub lethal concentration of insecticide, matasystox. *J. Aqua. Biol.* **16**(2): 42-44.

**Waykar, B. (2006):** Role of L-ascorbic acid in the protection of the hepatopncrease of the fresh water bivalves, *Parreysia cylindrical*.*Nat. J. life sciences.* **3**(supp) -501-508.

**Waykar, B. and Lomte V. S. (2004) :** Carbaryl induced changes in the ascorbic acid content in different tissues of fresh water bivalve *Parreysia cylindrica*. *Asian J. Microbiol.Biotech.Env.Sci.* **6**(3): 181- 189.

**Waykar B. and Tambe, R.(2011):** Mitigation of cypermethrin induced histological damage in the hepatopancreas of freshwater bivalve, *Parreysia cylindrica* by L-ascorbic acid. *The Ecoscan: special issue*, Vol.1,45-51.

**Wolansky, M. J., Gennings, C. and Crofton, K. M. (2006):** Relative potencies for acute effects of pyrethroids on motor function in rats. *Toxicol. Sci.* **89**(1): 271-277.