Original Article
Toxicology International, *Sept-Dec 2016 / Vol-23 / Issue-3, 260-264*
ISSN (Print) : 0971-6580
ISSN (Online) : 0976-5131
DOI: 10.22506/ti/2016/v23/i3/146720

Alteration in the biochemical contents of the fresh water fish *Cyprinus carpio* after chronic exposure to Sevin

Panhale A V[1*] and Muley D V[2]

Department of Zoology,
[1]Shivaji Univeristy, Kolhapur,
[2]Krantishinh Nana Patil College, Walwa, Sangli.
*panhale.arjun@rediffmail.com

Abstract

Sevin is pesticide containing active ingredients of carbaryl. It is largely used to control various agricultural pests of vegetables, citrus, cotton, nuts and ornamentals. In present study, effect of Sevin on biochemical contents was studied in gill, liver, muscle and kidney of common carp, *Cyprinus carpio*. The fish were exposed to 0.6 ppm (1/10th of LC50) and 0.3 ppm (1/20th of LC50) concentrations for a period of 30 days. Alterations in the biochemical contents of fish were observed in target tissues. The decrease in glycogen, protein and lipid content were observed in all tissues after chronic exposure at 0.6 ppm and 0.3 ppm concentrations over control respectively. Comparatively, less depletion in biochemical content was observed at 0.3 ppm than 0.6 ppm concentration.

Keywords: Sevin, *Cyprinus carpio*, Glycogen, Protein, Lipid, LC50.

Received: 04-07-2016 **Revised:** 26-10-2016 **Accepted:** 05-11-2016

1. Introduction

Pesticides, fertilizers, heavy metals are causing major contamination of aquatic resources. Water bodies used for aquaculture in tropical countries are under threat of such toxic pollutants [1]. Pesticides are reaching aquatic sources directly or indirectly by different means [2,3]. There are 234 kinds of pesticides used in India [4].Pesticides and heavy metals are common environmental contaminants which leads to an alteration in biochemical parameters of fish [5].

Sevin (1-naphthyl methyl carbamate insecticide) containing active ingredients of carbaryl. It is used to control pests of vegetables, gardens, landscape plants, lawns, citrus, cotton, nuts and ornamentals. There are several reports regarding toxicological effects of pesticides on biochemical aspects of the fish. Tilak et al., [6] have studied the biochemical changes induced by Fenvalerate in the fresh water fish *Chana punctatus*. Bhavan and Geraldine [7] have been studied carbaryl induced alterations in the biochemical metabolism of the prawn *M. malcomsonni*. Shrivastava and Singh [8] reported changes in protein level of the muscle in *Heteropneustes fossilis* after exposure to carbaryl. Saxena and Gupta [9] have studied the impact of carbamates on glycogen contents in the muscle of fresh water fish *Channa punctatus*. Mishra et al., [10] shown the effect of Malathion on lipid content of liver and muscles of *Anabas testudiens*. The biochemical changes due to the toxic stress of pesticides have been reported by [11,12]. Kamble and Muley [13] have studied the effects of acute exposure of Endosulfan and chlorpyriphos on the biochemical composition of *Sarotherodon mossambicus*. Biochemical contents like glycogen, protein and lipid play a major role as precursors in fish under stress conditions [1].

Plenty of work has been carried out by different researchers regarding the effect of pesticides on biochemical contents in different fish. In the present

* *Author for correspondence*

investigation, attempts have been made to analyze the effects of Sevin on biochemical contents of *Cyprinus carpio*, as biochemical content acts as biomarker under stress condition.[14].

2. Materials and methods

A collection of test animals was carried out from local sources. After acclimation, fishes were used for experimentation . Experimental fishes were having the average length of 6-9 cm and the weight about 10-14 grams. Sevin 50% EC was used as toxicant and 96h LC50 of Sevin to Cyprinus carpio was determined by using static bioassay test. After determining LC50 value, the fish were exposed to 0.6 ppm ($1/10^{th}$ of LC50) and 0.3 ppm ($1/20^{th}$ of LC50) concentrations for a period of 30 days. Everyday water was changed to maintain pesticide concentration. Another group of fish was maintained as a control group. After completion of the exposure period, the fishes were dissected out immediately and tissues like gill, liver, kidney and muscle were used for biochemical estimations. The glycogen content was estimated by DeZwaan and Zandee [15] method using anthrone reagent. The protein content was estimated by lowry's et al.,[16] method using follin phenol reagent and total lipids was estimated by Barnes and Black-Stock [17] method using vanilline reagent.

3. Results and Discussion

Glycogen as the chief source of energy plays a significant role in metabolism. In present findings glycogen observed in different tissues was in the order of liver > muscle > gill > kidney in the control group. Results obtained in the present work are given in the table 1. A significant decrease in glycogen content was found at 0.6 ppm ($1/10^{th}$ of LC50) and 0.3 ppm ($1/20^{th}$ of LC50) concentration. The maximum decrease in glycogen content ($p<0.001$) was observed in the kidney at 0.6 ppm and 0.3 ppm concentration while a minimum decrease was observed ($p<0.05$) in the liver at at 0.6 ppm concentration. Decrease in total glycogen content in different fish tissue, due to pesticidal stress has been reported by many researchers. Breautd et al.,[18] reported carbofuran induced biochemical and behavioral effects in gold fish *Carrassius auratus*. Jagatheeshwari.[19] observed decreasing changes in biochemical contents induced by phosalone. Ganeshwade et al.,[20] reported a decrease in glycogen content in tissue of P. ticto after exposure to dimethioate for 96h and 30 days.

Decrease in glycogen content in all tissue may be due to the utilization of carbohydrates for energy production and induced hypoxia by toxicants.(Somaiha et al.,[4]. Decreased glycogen content in all tissues clearly indicates excessive utilization of carbohydrates to cope up pesticide induced toxicity and stress conditions. Glycogen stored in different tissues is utilized in the form of carbohydrates during acute and chronic exposure or through glycolysis or hexose monophosphate pathway. Decrease in glycogen content is due to inhibition of hormones responsible for glycogen synthesis.[21,22].

In the present investigation, glycogen content found to be decreased in gill, liver , muscle and kidney. Decrease in glycogen content after exposure to pesticidal stress has been reported by many researchers. Rawat et al [23] have also reported continuous depletion in total glycogen content in the liver of *Heteropneustes fossilis* after exposure to endosulfan.

The protein content in different tissues of the fish *Cyprinus carpio* of the control group was in the order of muscle > liver >gill >kidney. Results obtained in the present investigation are given in the table 2 . Decrease in protein content has been found at 0.6 ppm ($1/10^{th}$ of LC50) and 0.3 ppm ($1/20^{th}$ of LC50) concentration after 30 days exposure to sevin. A change in protein content varies

Table 1. Changes in glycogen content after chronic exposure to Sevin (values are expressed in mg/100mg wet tissue)

Tissue	Control	$1/10^{th}$ (0.6ppm)	% change over control	$1/20^{th}$ (0.3ppm)	% change over control
Gill	2.104 ±0.148	1.433 ±0.104**	-31.879	1.455 ±0.131**	-30.817
Liver	6.101 ±0.533	5.054 ±0.367*	-17.157	5.136 ±0.319 N.S	-15.822
Muscle	4.276 ±0.353	3.449 ±0.185*	-19.341	3.466 ±0.255*	-18.955
Kidney	1.814 ±0.097	0.697 ±0.059***	-61.559	0.758 ±0.047***	-58.229

Values are mean ± S.D., *, **, *** indicates significance level $P<0.05$, $P<0.01$, $P<0.001$ and N.S.-Non significant. (n = 3)

Table 2. Changes in protein content after chronic exposure to Sevin Sevin (values are expressed in mg/100mg wet tissue)

Tissue	Control	1/10th (0.6ppm)	% change over control	1/20th (0.3ppm)	% change over control
Gill	8.32 ± 0.46	5.57 ± 0.26***	-33.01	6.05 ± 0.26***	-27.28
Liver	9.51 ± 0.42	6.75 ± 0.36***	-28.96	7.23 ± 0.41***	-23.90
Muscle	11.22 ± 0.26	7.56 ± 0.44***	-32.63	8.52 ± 0.42***	-24.09
Kidney	7.09 ± 0.35	4.96 ± 0.25***	-30.03	5.88 ± 0.27***	-17.01

Values are mean ± S.D., *** indicates significance level P<0.001, (n = 3)

Table 3. Changes in lipid content after chronic exposure to Sevin(values are expressed in mg/100mg wet tissue)

Tissue	Control	1/10th (0.6ppm)	% change over control	1/20th(0.3ppm)	% change over control
Gill	2.174 ± 0.098	1.396 ± 0.040***	-35.774	1.400 ± 0.080***	-35.607
Liver	3.979 ± 0.205	2.682 ± 0.139***	-32.592	2.860 ± 0.072***	-28.125
Muscle	1.672 ± 0.111	1.239 ± 0.054**	-25.902	1.336 ± 0.051**	-20.075
Kidney	1.892 ± 0.106	1.268 ± 0.077***	-32.995	1.497 ± 0.079**	-20.845

Values are mean ± S.D., **, *** indicates significance level P<0.01, P<0.001, (n = 3)

from tissue to tissue. Comparatively, maximum decrease in protein content was observed at 0.6 ppm concentration than 0.3 ppm concentration. The maximum decrease in protein content was noticed (P<0.001) at 0.6 ppm concentration in muscle and gill (-32.63 % and -33.01%) respectively. Similar kind of decreasing trend (P<0.001) was observed in muscle and gill (-24.09 % and -27.28%) at 0.3ppm concentration, while a minimum decrease in protein content was observed in liver (-28.96) at 0.6ppm concentration and in the kidney (-17.01%) at 0.3 ppm concentration. Bindukumari and Vasanthi [24] reported a decrease in protein content of various tissues of fresh water *fish Labeo rohita* treated to dimethoate. Somaih et al., [4] also reported decrease in total protein content of Labeo rohita after exposure to Phenthoate in tissues like brain, gill, kidney, liver, and muscle.

Depletion in protein content is due to its utilization in metabolic processes and resulted in stress of sevin induced toxicity. Pesticidal exposure of test fishes leads to hormonal imbalance, which leads to an adverse effect on protein content. [25].The decrease in protein content may be due to increased catabolism and decreased anabolism of proteins or degradation in protein content may be due to increased proteolysis and inhibition of protein synthesis. [26]. Singh et al.,[27] studied the effect of malathion and carbaryl on biochemical parameters of the fish *Colisa asciatus*. Venkatramana et al., [28] reported the impact of malathion on gobiid fish *Glossogobius giuris* (Ham).

The total lipid content observed in the control group was in the order of liver > gill > kidney > muscle.After 30 days of chronic exposure to sub lethal concentration of sevin, decrease in lipid content was observed at 0.6 ppm and 0.3 ppm concentrations. Comparatively highly significant (P<0.001) decrease in lipid content was observed in gill, liver and kidney while, significant decrease(P<0.01)was recorded in muscle at 0.6 ppm concentration. At 0.3 ppm concentration highly significant (P<0.001) decrease in lipid content were observed in gill and liver while, a significant decrease(P<0.01) in muscle and kidney. The maximum decrease in lipid content was observed in gill (-35.77%) at 0.6 ppm concentration and (-35.60%) at 0.3 ppm concentration. Dhanpackium et al.,[29] also reported decrease in total lipid content of liver and muscle in *Cyprinus carpio* after exposure to malathion

Present findings clearly indicate that Sevin induced toxicity resulted in a decrease of lipid content in different tissues of *C. carpio*. Changes in lipid contents of all target organs is because of utilization of lipid components at the cellular level or increased lipolysis or mitochondrial structural damage impair the function of citric acid and fatty acid mechanism.[29]. Piska et al., [30] have reported a decrease in lipid content of gill, liver and brain of *Cyprinus carpio* exposed to sublethal concentrations of cypermethrin. Mishra et al., [10] have studied the decrease in the level of lipid content in liver and muscle of Anabas testudines due to to malathion. Israel et al.,[31] reported a decrease in lipid content of liver after exposure to fenthion.

4. Conclusion

The changes in glycogen, protein and lipid content clearly indicate susceptibility of fish to Sevin. From the present study, it is clear that animal tries to cope up with stress conditions of the pesticide. The decrease in contents biochemical parameters clearly suggests alteration in fundamental mechanism of biochemical contents, such alterations in biochemical content affects the fish productivity.

5. Acknowledgement

The authors are thankful to UGC-SAP DRS-Phase I and department of Zoology, Shivaji University Kolhapur, for providing laboratory facilities.

6. Conflict of Interest

None Declared

7. References

1. Thenmozhi, C.V.vignesh, R.Thirumuragan,S. Arun et al.Impact of malathion on mortality and biochemical changes of fresh water fish *Labeo rohita*.Iran J. Environ. Health. Sci Eng 2011; Vol 8 pp-387-394.
2. Dutta HM, Dalal R. The effect of Endosulfan on the ovary of bluegill sunfish: a histopathological study (*Lepomis macrochirus* sp) Int J Environ Res. 2008;2:215–224.
3. Chandran R, Sivakumar AA, Mohandass S, Aruchami M. Effect of Cadmium and Zinc on Antioxidant Enzyme Activity in the Gastropod, Achatina fulica. Comp Biochem Physiol. 2005;140 C:422–426.
4. Somaiah, K., P.V.V.Satish, K.Sunita, B. Nagraju and O.Oyediran Oyebola et al. Toxic impact of phenthoate on protein and glycogen level in certain tissues of Indian major carp *Labeo rohita* (Ham) IOSR journal of Environmental science vol(8) issue 9pp-65-73.
5. Fatma A. S. Mohamed and Nahed,S.Gad (2008):Environmental pollution induced Biochemical changes in tissue of *Tillipia zillii*, *Solea vulgaris* and *Mugil capito* from lake qurun Egypt. Global veternica 2008; 2 (6):327-336
6. Tilak, K. S., Veeraih, K. and Vardhan, K. S et al. Toxicity and residue studies on Fenvalerate to the fresh water fish *Labeo rohita*.2003
7. Bhavan, P.S. and P. Geraldine Carbaryl-induced alterations in Biochemical metabolism of the prawn, *Macrobrachium malcolmsonii*. J. Environ. Biol., 2002;23, 157-162.
8. Shrivastava S, Singh S. Changes in protein content in the muscle of *Heteropneustes fossilis* exposed to carbaryl. J. Ecotoxicol. Environ. Monit. , 2004 14: 119-122.
9. Saxena, K.K and Gupta, P. Impact of carbamates on glycogen contents in the muscles of fresh waterfish *Channa punctatus*. Poll. Res. 2005; 24: 669-670.
10. Mishra,S.K.,Padhi,J. and Shahoo,L. et al. Effect of Malathion on lipid content of liver and muscles of *Anabas testudiens*.J. Appl. Zool.Res. 2004;15(1081-82
11. Khare,A.,Singh,S. Impact of Malathion on Protein content in the Fresh water fish *Clarius batracus*.J.Ecotoxicology and Environmental monitoring. 2002;12(2)129-132.
12. Rao,L.M. and K.Rameshwari Effect of Sublethal stress of Endosulfan and Monocrotophos on the biochemical components of *Labeo rohita*,Mystus vittatus and Chana punctatus. Ecology Environ . Con. 2000; 6(3) 289-296.
13. Kamble, G.B and Muley, D.V. Effect of acute exposure of endosulfon and chloropyrifos on the biochemical composition of the fish *Sarotherodon mossambicus*. Indian. J. Environ. science. 2000; 4(1) 97-102.
14. Agrahari, S., K. Gopal and K.C. Pandey et al. Biomarkers of Monocrotophos in freshwater fish, *Channa punctatus* (Bloch) J Environ Biol 2000; 27, 453-457
15. De Zwaan, A. and D. I. Zandee. Body distribution and seasonal changes in the glycogen content of the common mussel, *Mytilus edulis*. Comparative Biochemistry and physiology, 1972 43 A 53-58.
16. Lowry O. H.., Rosenbrough M. J., Farry A. L. and Randoll R. J. et al. Protein measurement with follin phenol reagent .J.Biol.Chem. 1956;(4)265-275.
17. Barnes, H. and Blacksock, J. Estimation of lipids in marine animals and tissues. Detailed investigation of sulphophos-phovanillin method for total lipids. Journal of Experimental Marine Biology and Ecology, 1973; Vol. 12 (1): 103 - 118.
18. Bretaud, Sandrine, Saglio, Philippe, Saligaut, Christian, Auperin, Benoit et al.Biochemical and behavioural effects of carbofuran in goldfish (*Carassivs auratus*). J. Environ. Toxicol. Chem., 2002; 21(1)175-181.
19. Jagatheeswari, J. Biochemical changes induced by pesticide - phosalone in *Cyprinus carpio* (Linn.) J Aqua Biol 2005; 20(1)123-125.
20. Ganeshwade, R.M., S.M. Pawar, S.R. Sonavane, K.D. Devararooand V B Sutar et al. Effect of Dimethoate on the level of glycogen in tissue of fresh water fish *Puntius ticto* (Ham)The Bioscan 2011 6(11)131-135.
21. Martinez, C.B.R., Nagae, M. Y., Zaia, C.T.B.V. and D.A.M. Zaia et al. Acute morphological and physiological effects of lead in the neotropical fish *Prochilodus lineatus*. Braz. J. Biol., 2004; 64(4)1-23.
22. Patil, I. and A.M. Malwadkar. Glycogen profile in *Puntius sarana sarana* from Pavana, a Tributary of river Bhima, Maharashtra.Bionano Frontier 2012;Vol. 5 (2) 250-252.
23. Rawat D.K..Bais V.S. and Agarwal N.C. et al., A Correrelative study on liver glycogen and endosulfan toxicity in *Heteropneustes fossilis* (Bloch) Journal of Env Biol 2002; 23(2)205-207.

24. Bindukumari,S and Vasanthi J. The Toxic Effect of Pesticides Dimethoate 30%EC on Protein Metabolism of the fresh water fish, *Labeo rohita*.Int.J.Curr.Microbiol.Appl.Sci. 2013;2(12)79-82.
25. Abdul, N C, Janaiah, D and Venkatashwarlu, P., The effect of Lichocin toxicity on protein metabolism of the fresh water edible fish *Chana punctatus* (Bloch) J of Toxicol and H sci, 2010; 3:18-23.
26. Muley, D V, Karanjkar, D M and Mhaske, S V et al., Impact of industrial effluents on the biochemical composition of the fresh water fish *Labeo rohita*. J. Environ. Biol. 2007; 28(2):243-249.
27. Singh,S.K.,P K Tripathi,R P Yadav D Singh and A Singh et al.,Toxicity of Malathion and Carbaryl Pesticided:Effect on some biochemical profiles of the fresh water fish *Colisa asciatus*.Bulletin of Environmental Contamination and Toxicology 2004; 72-592-599.
28. Venkataramana, G. V., Sandhyarani, P. N. and Murthy, P. S.et al., Impact of malathion on the biochemical parameters of gobiid fish, *Glossogobius giurus* (Ham) Environ Biol 2006; 27(1) pp-119-122.
29. Dhanpackium, P.,N. Fatima bibi and V.K. Ramasamy et al., Effect of Malathion on Fresh Water fish *Cyprinus carpio*.Utterpradesh journal of zoology 2000; 20(1)13-16.
30. Piska, R.S., Sarala, W., Indira, D. 1992. The effect of sublethal concentration of synthetic pyrethroid, cypermethrin to the common carp Cyprinus carpio (linn) fry J Environ Biol 1992; 13: 89 94.
31. Israel,S.S, S.Manohar,Das,S.et al.,Biochemical changes in certain tissue of Cirrhinus mrigala (ham)exposed to fenthion Int J Of Env Sci 2012; Vol 2 no 31268-1276.