

Acute Toxicity of Lambda-Cyhalothrin (Synthetic Pyrethroid) to the Juvenile and Adult Crayfish (*Astacus leptodactylus* Eschscholtz, 1823)

Selcuk Berber, Selcuk Turel and Hasan Kaya
Department of Basic Sciences, Faculty of Marine Sciences and Technology,
Canakkale Onsekiz Mart University, Canakkale, Turkey

Abstract: In this study, the acute toxicity of lambda-cyhalothrin, a synthetic pyrethroid pesticides heavily used in the paddy fields, to the juvenile and adults of *Astacus leptodactylus* was investigated. The two different experiments, one for the juvenile (0.417±0.001 g) and one for the adult (39±2.2 g) crayfish were conducted. Acute toxicity tests was performed in 3 L glass jars for the juvenile and in aquariums for the adults. Acute toxicity tests with a static control and 6 different concentrations with 3 replications were tested. About 10 healthy individuals were placed in each. During the experiments, the water temperature was kept at 20°C and average pH was constant at 7.8. At the end of the 96 h experiments, the LC₅₀ value of lambda-cyhalothrin for *Astacus leptodactylus* were found as 0.84 ng L⁻¹ for the juveniles and as 0.207 µg L⁻¹ for the adults. As a result, the heavily used lambda-cyhalothrin in the paddy fields was determined to be very toxic for juveniles and for adult of *Astacus leptodactylus*.

Key words: *Astacus leptodactylus*, lambda-cyhalothrin, acute toxicity, adults, LC₅₀

INTRODUCTION

The synthetic pyrethroid pesticides have been in widespread use for >30 years in the world in agriculture industry to get rid of the pests and to increase the yield of product used in households for public health reasons and used in ovine animals against ectoparasite. The fact that this product shows lower toxicity in mammals increased the use of such compounds and in mid 90's 23% of the total insecticide market was comprised of pyrethroid compounds (Soderlund *et al.*, 2002; Piner, 2009). The pyrethroids are the second most commonly used compounds behind the organo-phosphorus pesticides.

Lambda-cyhalothrin is a synthetic pyrethroid insecticide and acaricide that was developed in 1977. Due to the cyano group that is present in its molecular structure, it is considered as a type 2 pyrethroid (WHO, 1990). Its chemical formula is (α -cyano-3-phenoxybenzyl 3-(2-chloro-3,3,3-trifluoro-1-propenyl)-2,2-dimethylcyclopropanecarboxylate). The lambda-cyhalothrin is in widespread use against the aphids in cotton, grains, potato, various fruit and vegetables, against the coleopterous species that consume the leaves and against the butterfly larvae (Kidd and James, 1991). In addition, it is also utilized against the cockroach, mosquito, tick and flies that pose a threat to human health as well. The

lambda-cyhalothrin poses a high level of toxicity for many fish and aquatic invertebrate species (Extonet, 1996). The wooden bedstead is one of the model organisms that are used to determine the effects of the contamination. *A. leptodactylus* (Eschscholtz, 1823) which is also known as the Turkish crayfish, its widespread consumption and distribution is one of the significant freshwater species of Europe. This species may adapt into the different habitats such as deep and shallow lakes, small waters, rivers, ponds, dams as well as into the river mouth living conditions (Berber, 2009; Ahn *et al.*, 2006). The possible environmental alteration that may occur in aquatic environments may have a negative impact on crayfish species as well.

The crayfish-rice policulture has been implemented throughout the world for about 60 years. By this method, a freshwater crayfish production with a yield of 350-1000 kg ha⁻¹ can be achieved (Chien and Avault, 1980; Brunson and Griffin, 1988; Huner, 1988; McClain *et al.*, 1998). In the country, the freshwater crayfish production is only made through hunting from natural or artificial water bodies. There is no current study, conducted on the practicability of the crayfish-rice policulture model in the country, available so far. In addition to this, as of 2012, a total area of 120,000 ha is utilized for rice farming. Since, the possibility of the production of 1000 kg crayfish per hectare via

crayfish-rice policulture method is documented, there is a significant potential of crayfish production. When practicing crayfish-rice alternating farming production model, the effects of the pesticides that are used for rice plants on the crayfish should be studied carefully.

This study is partaking of a preparatory study in order to ensure that the crayfish-rice production model will be able to be implemented in the country in the future. Revealing the effects of the agricultural pesticides that are used in rice fields on the freshwater crayfish.

MATERIALS AND METHODS

Juvenile crayfish experiment: Within the scope of the study, the spawners that were used to obtain juveniles were supplied from the local fishermen of Lake Porsuk. The naturally obtained spawners were fed with the crustasae food in the fostering tanks of the Faculty of the Marine Sciences and Technologies until they produce their offsprings. The crayfish offsprings that were obtained from the spawners were raised for 2 months in aquarium conditions and their adaptation was ensured. The juvenile individual trials were conducted in 3 L glass jars. A design that includes 10 juvenile which are selected from the healthy individuals, being placed in each jar, mean weight is 0.417 ± 0.001 g with 3 repetitions was set up. The acute toxicity test was carried out for a period of 96 h by conducting a static experiment with one control and 6 different concentrations (0.5, 0.6, 0.7, 0.8, 0.9 and 1 ng L^{-1}) In the trial, the water temperature was measured as $21 \pm 0.1^\circ\text{C}$, its pH value was measured as 7.8 ± 0.05 , its dissolved oxygen ratio was determined as $6.80 \pm 0.08 \text{ mg L}^{-1}$ and the total ammonia was measured as $0.12 \pm 0.01 \text{ mg L}^{-1}$. During the trials, no food was provided to crayfish juveniles. At the end of the 96 h experiment, the LC_{50} value which killed off 50% of the juvenile individuals and the safety range of 95% were calculated via the computer software called EPA Probit Analysis Program Used For Calculating LC/EC Values Version 1.5.

Adult crayfish experiment: The adult individuals of *A. leptodactylus* which were used in the experiment (mean weight: 39 ± 2.2 g) were supplied from the local fishermen of Lake Porsuk and by being kept in 18 stock aquarium with 20 L of capacity in the with the size of $45 \times 28 \times 80$ in the Faculty of the Marine Sciences and Technologies Laboratories for 1 month, adopted to the ambient conditions. In the study, the individuals were separated into 18 trial aquarium in the way that there would be 10 crayfish in each tank and a test design of 3 replicates was created. The acute toxicity test was carried out for a period of 96 h by conducting a static

experiment with one control and 6 different concentrations (0.05, 0.01, 0.15, 0.2, 0.3, $0.4 \text{ } \mu\text{g L}^{-1}$). In the trial, the water temperature was measured as $21 \pm 0.1^\circ\text{C}$, its pH value was measured as 7.72 ± 0.06 , its dissolved oxygen ratio was determined as $6.65 \pm 0.1 \text{ mg L}^{-1}$ and the total ammonia was measured as $0.125 \pm 0.01 \text{ mg L}^{-1}$. During the trials, no food was provided to adult crayfish. At the end of the 96 h experiment, the LC_{50} value which killed off 50% of the adult individuals and the safety range of 95% were calculated via the computer software called EPA Probit Analysis Program used for calculating LC/EC values Version 1.5.

Preparation of the lambda-cyhalothrin stock solutions and dosing: In the trial, the analytic standard active substance with the brand Sigma-Aldrich (Stenheim, Germany-99.5%) was used. The stock solution was extracted by solving the active substance within the acetone (APHA/AWWA/WEF, 1998). Both the stock and the dose solutions were kept at 4°C until being used. By heating it up to the ambient temperature, the stock solution was administered into the experiment jars and aquariums.

RESULTS AND DISCUSSION

After the 96 h trial, in order to identify the lethal concentration, the number of the dead individuals and their mortality ratio were determined and by using the computer software, those figures were converted into the probit values (Anonymous, 1998). In the study while the LC_{50} value of the lambda cyhalothrin on the juvenile individuals of *A. leptodactylus* was revealed to be 0.84 ng L^{-1} (Table 1 and Fig. 1), the LC_{50} value on the adult individuals were identified as $0.207 \text{ } \mu\text{g L}^{-1}$ (Table 2 and Fig. 2). While only one death was observed in the control group during the juvenile trials, no death occurred in control group in adult trial.

As the result of the study, the active substance of the insecticide lambda-cyhalothrin was revealed to be severely toxic for both the juvenile and the adult

Table 1: The 96 h acute toxicity of Lambda cyhalothrin on crayfish juveniles 95% confidence interval (Min.-Max. value)

Points	Concentration (ng L ⁻¹)	Low (ng L ⁻¹)	High (ng L ⁻¹)
LC/EC 1.00	0.293	0.156	0.388
LC/EC 5.00	0.399	0.259	0.485
LC/EC 10.00	0.470	0.338	0.549
LC/EC 15.00	0.526	0.405	0.597
LC/EC 50.00	0.840	0.768	0.955
LC/EC 85.00	1.340	1.119	1.990
LC/EC 90.00	1.497	1.215	2.385
LC/EC 95.00	1.764	1.370	3.122
LC/EC 99.00	2.400	1.713	5.185

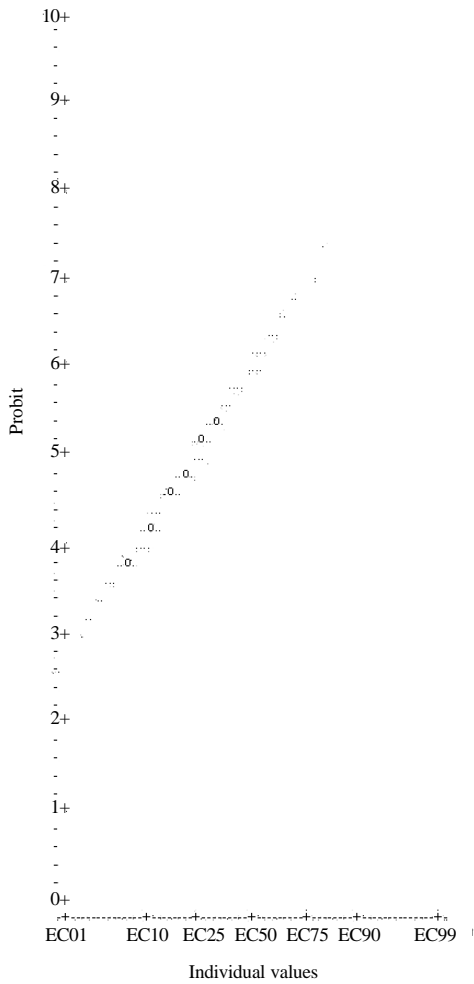


Fig. 1: Plot of adjusted probits and predicted regression line for juvenile caryfish



Fig. 2: Plot of adjusted probits and predicted regression line for adult caryfish

individuals of *Astacus leptodactylus* species. The findings obtained by this hereby study are found out to be consistent with the results of the toxicity researches in the literature, conducted regarding the impacts of the active substance of lambda-cyhalothrin on fish and aquatic invertebrate species. The LC_{50} values of Lambda-cyhalothrin was determined to as $0.21 \mu\text{g L}^{-1}$ for bluegill sunfish (*Lepomis macrochirus*) (Kidd and James, 1991) $0.24 \mu\text{g L}^{-1}$ for the rainbow trout (Anonymous, 1998) 4.9 ng L^{-1} (Anonymous, 1998) for mysid prawn and $0.36 \mu\text{g L}^{-1}$ (Anonymous, 1998) for *Daphnia magna*. In addition, the LC_{50} value for the young adults of *Procambarus clarkii* which is another freshwater crayfish (3 months) was determined as $0.16 \mu\text{g L}^{-1}$ (Barbee *et al.*, 2009). The findings of this study suggest that the *A. leptodactylus* is more susceptible to lambda-cyhalothrine than *P. clarkii*.

Table 2: The 96 h acute toxicity of lambda-cyhalothrin on adult crayfish 95% confidence interval (Min.-Max. value)

Points	Concentration ($\mu\text{g L}^{-1}$)	Low ($\mu\text{g L}^{-1}$)	High ($\mu\text{g L}^{-1}$)
LC/EC 1.00	0.034	0.013	0.055
LC/EC 5.00	0.058	0.029	0.083
LC/EC 10.00	0.077	0.044	0.103
LC/EC 15.00	0.093	0.059	0.120
LC/EC 50.00	0.207	0.169	0.260
LC/EC 85.00	0.462	0.346	0.795
LC/EC 90.00	0.558	0.401	1.057
LC/EC 95.00	0.739	0.498	1.618
LC/EC 99.00	1.253	0.744	3.620

In this study, it was determined that the active substance of lambda-cyhalothrin which is densely used in rice fields is highly toxic to the juvenile and adult individuals of *A. leptodactylus* species. Although, the synthetic pyrethroids are a kind of pesticides that are

used and favored more due to their fast decomposition, rapid absorption by the soil and its lower toxicity in mammals, it is also ominous that they have at the same time very high toxicity level for the aquatic organisms. For a successful and productive crayfish-rice rotation application, the trials or the development of a new substance which would have significantly lower acute toxicity, instead of the lambda-cyhalothrin pesticide is foreseen.

CONCLUSION

The estimated LC_{50} values that are determined for the crayfish in laboratory conditions, provides information to the researches regarding the maximum exposure concentrations and lethal dose. However, in the field conditions in which more than 1 factor are effective, it is possible that the toxicity of such substances may change and vary. In addition, the formulation of the pesticide, the usage (seed therapy or leaf administration), frequency of application and time, the pH value and temperature of the water, the life cycle of the crayfish, the food status and the size of the organism affect the toxicity of the pesticides (Mayer and Ellersieck, 1988; Morgan and Brunson, 2002). In order to be able to conduct a realistic environmental impact study by presenting the impact of the pesticides, long term observation studies on the concentrations that the organisms are exposed to in the nature.

ACKNOWLEDGEMENT

This study was supported by the Çanakkale Onsekiz Mart University Scientific Foundation (BAP) (Project No: 2011/67).

REFERENCES

- APHA/AWWA/WEF, 1998. Standard Methods for the Examination of Water and Wastewater. 20th Edn., American Public Health Association/American Water Works Association/Water Environment Federation, Washington, DC., USA., ISBN-13: 9780875532356, Pages: 1220.
- Ahn, D.H., T. Kawai, S.J. Kim, H.S. Rho and J.W. Jung *et al.*, 2006. Phylogeny of Northern hemisphere freshwater crayfishes based on 16S rRNA gene analysis. Korean J. Genet., 28: 185-192.
- Anonymous, 1998. US environmental protection agency. Fact Sheet No. 171, Karate, Washington, DC, USA.
- Barbee, G.C., W.R. McClain, S.K. Lanka and M.J. Stout, 2009. Acute toxicity of chlorantraniliprole to non-target crayfish (*Procambarus clarkii*) associated with rice-crayfish cropping systems. Pest Manag. Sci., 66: 996-1001.
- Berber, S., 2009. Natural resources conservation and management and protection of crayfish stocks in Turkey. 25-26 July, Egirdir, Isparta.
- Brunson, M.W. and J.L. Griffin, 1988. Comparison of rice-crayfish and grain sorghum-crayfish double cropping systems. Aquaculture, 72: 265-272.
- Chien, Y.H. and J.W. Jr. Avault, 1980. Production of crayfish in rice fields. Prog. Fish. Cult., 42: 67-71.
- Extonet, 1996. Extension toxicology network: Pesticide information profiles. <http://extoxnet.orst.edu/pips/cypermet.htm>.
- Huner, J.V., 1988. *Procambarus* in North America and Elsewhere. In: Freshwater Crayfish Biology: Management and Exploitation, Holdich, D.M. (Ed.). 1st Edn., Croom Helm, London, UK., pp: 239-261.
- Kidd, H. and D.R. James, 1991. The Agrochemicals Handbook. 3rd Edn., Royal Society of Chemistry Information Services, Cambridge, UK., pp: 2-13.
- Mayer Jr., F.L. and M.R. Ellersieck, 1988. Experiences with single-species tests for acute toxic effects on freshwater animals. Ambiology, 17: 367-375.
- McClain, W.R., J.L. Avery and R.P. Romaine, 1998. Crawfish production: Production systems and forages. SRAC Publication No. 241, Southern Regional Aquaculture Center (SRAC). <https://srac.tamu.edu/index.cfm/event/getFactSheet/whichfactsheet/45/>.
- Morgan, E.R. and M.W. Brunson, 2002. Toxicities of agricultural pesticides to selected aquatic organisms. SRAC Publication No. 4600, Southern Regional Aquaculture Center (SRAC). <https://srac.tamu.edu/index.cfm/event/getFactSheet/whichfactsheet/166/>.
- Piner, P., 2009. Determination of oxidative stress potential of lambda-cyhalothrin in the presence of PBO and its effects on stress proteins and apoptosis in the Liver of *Oreochromis niloticus*. Ph.D. Thesis, Department of Biology, Institute of Natural and Applied Sciences Cukurova University, Turkey.
- Soderlund, D.M., J.M. Clark, L.P. Sheets, L.S. Mullin and V.J. Piccirillo *et al.*, 2002. Mechanisms of pyrethroid neurotoxicity: Implications for cumulative risk assessment. Toxicology, 171: 3-59.
- WHO, 1990. International programme on chemical safety. Environmental Health Criteria No. 99, Cyhalothrin, World Health Organization, Geneva, Switzerland.