



ACUTE ORAL TOXICITY STUDY OF DELTAMETHRIN IN COMBINATION WITH INSECT REPELLENTS, N,N - DIETHYL BENZAMIDE, N,N - DIETHYL PHENYLACETAMIDE AND N,N - DIETHYL METATOLUAMIDE IN RATS

**ANITHA MAGESH*, R. VIJAYARAGHAVAN, S. SENTHIL KUMAR,
ANITHA ROY AND R.V. GEETHA**

Department of Research and Development, Saveetha University, Chennai - 602105

ABSTRACT

To control a variety of vector borne diseases insecticides and insect repellents are used. They share a common mode of toxicity, and are used together inside and outside the living areas. The present study was designed to evaluate the insect repellents N,N-diethyl benzamide (Deb), N,N-diethyl phenylacetamide (Depa) and N,N,-diethyl metatoluamide (Deet) in combination with insecticide deltamethrin (Del) in rats. The study was conducted in 2 phases. Phase I was determination of LD₅₀ of Del by Gad and Weil moving average method and LD₅₀ of Deb, Depa and Deet by Dixon Up and Down method. Phase II was determination of LD₅₀ of insect repellents in combination with insecticide by Dixon Up and Down method in rats. Among the four, Del was more toxic than the insect repellents. Among the repellents, Deet was least toxic chemical. The estimated LD₅₀ of Del, Deb, Depa and Deet were 129.7, 1035.1, 333.3 and 1894.1 mg/kg respectively. The extrapolated LD₅₀ estimate of Del in presence of one LD₅₀ of Deb, Depa and Deet was 65.9, 40.3 and 20.2 mg/kg respectively. The LD₅₀ were additive and no potentiation was observed between the insecticide and the insect repellents showing they can be used together.

KEYWORDS: Deltamethrin, Deb, Depa, Deet, combined toxicity, LD₅₀, rats.



ANITHA MAGESH*

Department of Research and Development,
Saveetha University, Chennai – 602105.

Received on : 25-01-2017

Revised and Accepted on : 25-04-2017

DOI: <http://dx.doi.org/10.22376/ijpbs.2017.8.2.p426-431>

INTRODUCTION

There are many methods to control mosquitoes viz., physical, biological, chemical and personal protection to prevent from vector borne diseases like malaria, dengue, chikungunya, yellow fever, filariasis and Japanese encephalitis¹. The personal protection methods are insect repellents that are available in the form cream, spray, lotion and coil. When applied on body, the chemical which is present prevent the mosquitoes from biting for a certain period of time. The repellents are, plant derivatives like citronella, neem, and peppermint², and synthetic chemicals like N,N-diethyl benzamide (Deb), N,N-diethyl phenylacetamide (Depa) and N,N,-diethyl metatoluamide (Deet)^{3,4}. Deltamethrin (Del) is an insecticide which is widely used against agricultural and household pests. It is a pyrethroid and an organic compound⁵. It is similar to natural pyrethrins extracted from pyrethrum flowers. The action of the insecticide is either by direct contact or by ingestion. Del action on insect is by affecting the ion channel in the nervous system due to the phosphorylation state and delays inhibition of sodium channel activation gate. Due to this there is a prolonged sodium in nerve that results in repeated nerve signals in sensory organ, nerves and muscles⁶. The toxicity of xenobiotics are of two types, acute toxicity i.e., single large dose which is fatal, and acute or chronic toxicity i.e., small or repeated doses which is fatal⁷. LD₅₀ (lethal dose 50) means the administration of a chemical for a group of laboratory animals and when 50% of animals die than this dose is considered as LD₅₀⁸. If the LD₅₀ value is small the toxicity is high and if the LD₅₀ value is high than the toxicity is low. The LD₅₀ of new chemicals and drugs are determined by various route of administration viz., oral, dermal, inhalation and parenteral routes⁹. The determination of the LD₅₀ provides information of hazards from exposure to the chemicals and it also provides classifying and determining the dosage for both subacute and chronic toxicity studies¹⁰. In general the LD₅₀ estimates of individual chemicals are reported, but in combination the information is not available¹¹. If a chemical potentiates the toxic effect of another chemicals it will be more hazardous¹². For controlling a variety of vector borne diseases insecticides and insect repellents are used. They share a common mode of toxicity, and are used together inside and outside the living areas. The present study was designed to evaluate the insect repellents, Deb, Depa and Deet in combination with insecticide, Del in rats.

MATERIALS AND METHODS

Chemicals

Deltamethrin (Del), N,N'-diethylbenzamide (Deb), and N,N'-diethyl phenylacetamide (Depa) were obtained from M/s Tagros Chemicals India Ltd (Chennai, India) and N,N'- diethyl metatoluamide (Deet) from Alkyl Amine Chemicals Ltd. (Mumbai, India). All other chemicals used were of analytical grade.

Animals

Adult female Wistar rats (*Rattus norvegicus*) weighing between 150 to 200 g from Biomedical Research Unit and Laboratory Animal Facility (BRULAC) were used. The animals were housed as per the guidelines of the "Committee for the Purpose of Control and Supervision of Experiments on Animals" (CPCSEA, India). The rats were fed with standard pellet diet and given filtered water ad libitum. They were maintained at natural light and dark cycle. The study protocol was approved by the Institutional Animal Ethics Committee (SU/BRULAC/RD009/2014).

LD₅₀ determination

The LD₅₀ was determined in two phases, (i) in the phase I the LD₅₀ of Del, Deb, Depa and Deet were estimated and (ii) in phase II the LD₅₀ in combination was estimated.

LD₅₀ determination of Del

The LD₅₀ of Del was estimated by Gad and Weil moving average method¹³. Del was made into a suspension with olive oil and was administered orally using a 16 gauge oral feeding cannula (about 0.2 mL/100 g). Four logarithmic doses of 50, 100, 200 and 400 mg/kg of Del were given and for each dose 4 rats were used. Food and water were withheld for a period of 2 hr. The rats were weighed daily and mortality was observed for a period of 14 days.

LD₅₀ determination of Deb, Depa and Deet

The LD₅₀ of Deb, Depa and Deet were estimated by Dixon Up and Down method¹⁴. The insect repellents were dissolved in olive oil and were administered orally using a 16 gauge oral feeding cannula (about 0.2 mL/100 g). By this method, the animals were administered the dose on a logarithmic scale. If the dose produced mortality a lower logarithmic dose was given and if the animal survived a higher logarithmic dose was given. For each insect repellent 6 to 8 animals were used for estimating the LD₅₀. The rats were weighed daily, and survival or mortality for a period of 14 days was considered.

LD₅₀ determination of combination of Del with Deb, Depa and Deet

The LD₅₀ of Deb, Depa and Deet in combination with Del were estimated by Dixon Up and Down method¹⁴. For the combined LD₅₀, three combinations for each insect repellent was taken. Del+insect repellent = 1/4+3/4 LD₅₀, 1/2+1/2 LD₅₀ and 3/4+1/4 LD₅₀. The required dose for 100 g body weight of the animal was made in 0.2 mL of olive oil and administered orally using a 16 gauge oral feeding cannula, about 0.2 mL/100 g as shown in Fig. If the dose produced a mortality a lower logarithmic dose (volume) was given and if the animal survived a higher logarithmic dose (volume) was given. The following logarithmic doses (volume) were used, 0.10, 0.14, 0.20, 0.28 and 0.40 mL. For each combination 6 to 8 animals were used for estimating the LD₅₀. The rats were weighed daily, and survival or mortality for a period of 14 days was considered.



Statistics

The survived animals body weight was calculated as percent change and expressed as mean \pm SE. One way ANOVA with Dunnett's test was used for comparing with control. SigmaPlot 13 was used for statistical analysis and for plotting the graph (Systat Software, USA). The LD₅₀ was estimated either by Gad and Weil method or by Dixon Up and Down method.

RESULTS

The animals did not show any sign and symptom immediately after the oral administration of Del or Deb, Depa and Deet. After 4 hr the animals showed slow activity and sometimes they were hyperactive. The animals showed gasping and death occurred within 24 hr. No mortality was observed after that. The individual oral LD₅₀ estimate of Del, Deb, Depa and Deet are given in Table 1. Among the four the pyrethroid insecticide (Del) was more toxic than the insect repellents. Among the repellents Deet was the least toxic chemical. The estimated LD₅₀ of Del, Deb, Depa and Deet was 129.7, 1035.1, 333.3 and 1864.1 mg/kg respectively. The dilutions used for the combined toxicity are shown in Table 2. The dose was calculated for 100 g rat for a oral feeding volume of 0.2 mL. The individual chemicals were weighed and made into a suspension in olive oil. This method of dilution was simple and easy to handle. Table 3 shows the combined oral LD₅₀ of Del with Deb, Depa and Deet. In the combination the LD₅₀ were dose dependent. The LD₅₀ estimate of the combinations of Del with Deb, Depa and Deet is shown graphically in Figure 1. Good correlation was observed between the combinations of LD₅₀. The LD₅₀ was additive in this experiment and no potentiation was observed between the insecticide and the insect repellents. The LD₅₀ slope of Deb, Depa and Deet in presence of Del was 0.031, 0.403 and 0.043 respectively. The extrapolated LD₅₀ estimate of Del in presence of one LD₅₀ of Deb, Depa and Deet was 65.9, 40.3 and 20.2 mg/kg respectively. Table 4 shows the body weight changes of Del, Deb, Depa and Deet as independent chemical. Though, death occurred within 24 hr in all the chemicals recovery was observed after that. No change in body weight was observed among the survived animals except Depa showed a lower body weight compared to the control. Table 5 shows the body weight changes of Del in combination with Deb, Depa and Deet. No change in body weight was observed in any of the combinations, among the survived animals.

DISCUSSION

Pesticides and insect repellents are widely used but most of them show high degree of toxicity because they were designed to kill. Same time it can cause health hazards to human health which is of great concern¹³. In modern life, the use of pesticides and insect repellent are increased as people are very prone to mosquito bites. Normally the available mosquito repellents contain Deb, Depa or Deet. Therefore, in this study the various repellents were tested on rats to find out LD₅₀ of the individual agents and in combination with the insecticide. There is a general tendency to use the insecticide, Del and the insect repellents like Deb, Depa or Deet together at home and at outside locations. Both the insecticide and the insect repellents are neurotoxic^{14,15}. Acute toxicity experiments are very essential to assess the LD₅₀¹⁶. The three logarithmic concentrations of individual agent was used to study the LD₅₀ and later their combination in three different ratio Del + insect repellent, 1/4+3/4 LD₅₀, 1/2+1/2 LD₅₀ and 3/4+1/4 LD₅₀. Among the four, the insecticide Del was more toxic than the repellents and Deet was the least toxic. The acute inhalation toxicity studies on Deb, Depa and Deet showed that they have low mammalian toxicity³. The combination of Del with other repellents was not showing any potentiation. The extrapolated LD₅₀ of Del in presence of one LD₅₀ of Deb, Depa and Deet gave a positive value showing the effect is additive. A negative value would have shown potentiation. The pyrethroid poisoning effect is on sodium and chloride channels. It alters the sodium voltage channels by prolonging its closure and also decreasing the chloride in the chloride voltage channel¹⁷. This mechanism brings about the toxic action. The effect of Del is on the muscles and non-myelinated nerve fibres¹⁸. Studies conducted on Deet proved that it has low toxic effect. Unlike some insecticides it has no inhibitory action on acetylcholinesterase in insects and in mammals¹⁹. But, it may cause neuroexcitation. Information on Deb and Depa are not available. The present study shows that the toxic effect of insecticide, Del and the insect repellents, Deb, Depa or Deet were additive and no potentiation was observed. They can be used together within the permissible limits inside and outside the house. However, detailed sub-acute toxicological studies are required

Table 1
Oral LD₅₀ of Del, Deb, Depa and Deet in rat

Abbre-iation	Dose (mg/kg)	Test Result Died out of 5 rats	Test out come; Lowest Dose (mg/kg)	Dose Difference (log)	f value	LD ₅₀ (mg/kg)	Confidence Limit (mg/kg)
Del	50	0	0,3,3,4; 50	0.301	0.375	129.7	66.4 -253.2
	100	3					
	200	3					
	400	4					
Abbre-iation	Dose (mg/kg)	Test Result O = survived X = Died	Test out come, Last Dose (mg/kg)	Dose Difference (log)	K value	LD ₅₀ (mg/kg)	Confidence Limit (mg/kg)
Deb	400	OO	OOXXOO	0.301	0.372	1035.1	425.6 - 2517.7
	800	XOO					
Depa	1600	X	800	0.301	0.737	333.3	137.1 – 811.0
	100	OO	OOXOXO				
	200	OOO	200				
Deet	400	XX	OOOXXOX	0.301	0.244	1894.1	899.5 – 3981.1
	800	OO					
	1600	OXX					
	3200	OX					
	6400	X	1600				

LD₅₀ of Del by Gad and Weil method and Deb, Depa and Deet by Dixon Up and Down method

Table 2
Dilutions used for Del, Deb, Depa and Deet in olive oil for combined oral LD₅₀ in rat

Chemical	Parameter	1/4 LD ₅₀	1/2 LD ₅₀	3/4 LD ₅₀
		A	B	C
Del	Dose	32.4	64.9	97.3
	For 100 g rat	3.24	6.49	9.72
	Volume	0.2 mL	0.2 mL	0.2 mL
	Strength (mg/mL)	16.2	32.4	48.6
Chemical	Parameter	3/4 LD ₅₀	1/2 LD ₅₀	1/4 LD ₅₀
		A	B	C
Deb	Dose	776.4	517.6	258.8
	For 100 g rat	77.63	51.76	25.88
	Volume	0.2 mL	0.2 mL	0.2 mL
	Strength (mg/mL)	388.2	258.8	129.4
Depa	Dose	250.0	166.7	83.3
	For 100 g rat	25.0	16.67	8.33
	Volume	0.2 mL	0.2 mL	0.2 mL
	Strength (mg/mL)	125.0	83.4	41.7
Deet	Dose	1420.6	947.1	473.5
	For 100 g rat	142.0	94.7	47.4
	Volume	0.2 mL	0.2 mL	0.2 mL
	Strength (mg/mL)	710.0	474.0	273.0

The volumes of the combination administered for the determination of LD₅₀ are 0.10, 0.14, 0.28 and 0.40 mL.

Table 3
Combined oral LD₅₀ of Del with Deb, Depa and Deet in rat

Combi- nation	Dose in volume (log value)					Test out come O = Survived X = Died	Last dose (log)	Dose Difference (log)	K Value
	0.1 mL (- 1.0)	0.14 mL (-0.854)	0.20 mL (-0.699)	0.28 mL (-0.553)	0.40 mL (-0.398)				
Del-Deb									
A	O	OO	OO	OOO	XX	OOOXXOXO	-0.553	0.150	0.741
B	O	OOO	XXO	X		OOOXXOXO	-0.854	0.150	0.952
C	OO	OO	XOO	XX		OOOXXOOX	-0.553	0.150	-0.753
Del-Depa									
A		OO	OX	OX	X	OOOXXOXO	-0.854	0.150	1.544
B		O	OO	OX	X	OOOXXOXO	-0.699	0.150	1.139
C		O	OOO	XX	X	OOXOXO	-0.699	0.150	0.737
Del-Deet									
A		OO	OXO	X		OOXXOO	-0.699	0.150	0.372
B		OO	XX	OX		OXOX	-0.699	0.150	-0.500
C		OO	OXX	XX		OOXXOX	-0.699	0.150	-0.169
Combi- nation	LD ₅₀ Calculation log LD ₅₀ = log Last Dose + k value (log Dose Difference) = anti log (mL)					Del Dose (mg/kg) *	Insect Repellent Dose (mg/kg) *		
A	LD ₅₀	-0.553 + 0.741 (0.150)			- 0.44185 = 0.3615 mL		Del	58.6	Deb
B	LD ₅₀	-0.854 + 0.952 (0.150)			- 0.7112 = 0.1944 mL	Del	63.1	Deb	503.1
C	LD ₅₀	-0.553 + -0.753 (0.150)			- 0.66595 = 0.2157 mL	Del	104.9	Deb	279.1
A	LD ₅₀	-0.854 + 1.544 (0.150)			- 0.6224 = 0.2386 mL	Del	38.7	Depa	298.3
B	LD ₅₀	-0.699 + 1.139 (0.150)			- 0.52815 = 0.2964 mL	Del	96.2	Depa	247.0

C	LD ₅₀	-0.699 + 0.737 (0.150)	- 0.5884 = 0.2580 mL	Del	125.5	Depa	107.5
A	LD ₅₀	-0.699 + 0.372 (0.150)	- 0.6432 = 0.2274 mL	Del	36.8	Deet	1615.2
B	LD ₅₀	-0.699 + -0.500 (0.150)	- 0.7740 = 0.1683 mL	Del	54.6	Deet	797.0
C	LD ₅₀	-0.699 + -0.169 (0.150)	- 0.72435 = 0.1886 mL	Del	91.8	Deet	446.5
Combinations (mg/kg)			Del (0.2 mL)	Deb (0.2 mL)	Depa (0.2 mL)	Deet (0.2 mL)	
A Combination	1/4 Del + 3/4 Insect Repellent		32.4	776.3	250.0		1420.6
B Combination	1/2 Del + 1/2 Insect Repellent		64.9	517.6	166.7		947.1
C Combination	3/4 Del + 1/4 Insect Repellent		97.3	258.8	83.3		473.5

*The Del, Deb, Depa and Deet doses were used for plotting the figure 1.

Table 4
Body weight (percent) of rats following p.o. administration of Del, Depa and Deet.

Groups	3 days		7 days		14 days	
	Mean	SE	Mean	SE	Mean	SE
Control	102.9	1.7	99.1	2.9	105.4	3.6
Del	99.2	0.8	101.5	1.0	103.6	1.3
Deb	96.7	3.2	94.7	2.5	100.2	3.5
Depa	99.3	0.7	101.3	2.3	90.3*	4.3
Deet	98.6	1.1	99.6	1.7	99.7	3.4
F	1.73		1.97		3.22	
P	NS		NS		< 0.05	

Values are mean ± SE (n = 4 to 6) of survived animals.

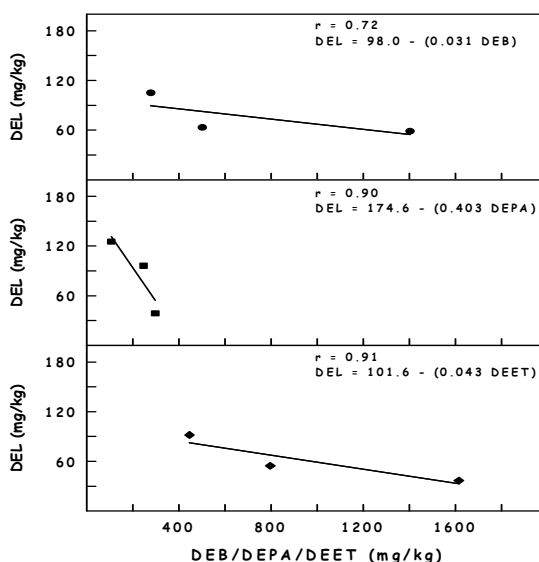
*Statistically significant from control by one way ANOVA with Dunnett's test.

Table 5
Body weight (percent) of rats following p.o. administration of Del in combination with Deb, Depa and Deet.

Groups	Combina-tion LD ₅₀	3 days		7 days		14 days	
		Mean	SE	Mean	SE	Mean	SE
Control	-	102.9	1.7	99.1	2.9	105.4	3.6
Del-Deb	1/4 Del + 3/4 Deb	96.6	2.0	98.5	2.9	105.2	6.3
	1/2 Del + 1/2 Deb	96.9	1.2	96.2	0.9	101.4	1.0
	3/4 Del + 1/4 Deb	100.6	1.8	100.2	1.8	99.8	0.8
Del-Depa	1/4 Del + 3/4 Depa	104.6	1.1	105.1	4.8	110.5	5.9
	1/2 Del + 1/2 Depa	98.4	1.6	103.2	4.5	104.6	3.5
	3/4 Del + 1/4 Depa	98.5	0.7	97.4	0.8	104.9	1.6
Del-Deet	1/4 Del + 3/4 Deet	101.5	3.6	101.8	3.5	100.0	2.5
	1/2 Del + 1/2 Deet	96.3	3.2	96.3	3.7	95.1	5.9
	3/4 Del + 1/4 Deet	100.8	2.1	98.2	1.5	94.5	3.7
F		1.69		1.06		1.73	
P		NS		NS		NS	

Values are mean ± SE (n = 4 to 6) of survived animals.

*Statistically significant from control by one way ANOVA with Dunnett's test.



CONCLUSION

The present study shows the combined LD₅₀ effect of various insect repellents (Deb, Depa and Deet) with pyrethroid insecticide (Del) by oral route in Wistar rats. The study shows that the toxicity of pyrethroid insecticide and the insect repellent are additive and there is no potentiating of toxicity. Hence, they can be used together either indoor or outdoor.

REFERENCES

1. Fankhauser B, Dumont P, Hunter JS 3rd, McCall JW, Kaufmann C, Mathis A, Young DR, Carroll SP, McCall S, Chester ST, Soll MD. Repellent and insecticidal efficacy of a new combination of fipronil and permethrin against three mosquito species. *Parasit Vectors* 2015; 8: 64. 2.
2. Wong KY, Signal FA, Campion SH, Motion RL. Citronella as an Insect Repellent in Food Packaging. *J. Agric. Food Chem.* 2005; 53: 4633-4636.
3. Deb U, Ahmed F, Singh S, Mendki MJ, Vijayaraghavan R. Comparative effects of insect repellent N,N-diethylbenzamide, N,N-diethylphenylacetamide, and N,N-diethyl-3-methylbenzamide aerosols on the breathing pattern and respiratory variables in mice. *Inhal Toxicol.* 2010 22: 469-478.
3. Prakash S, Vijayaraghavan R, Sekhar K. DEPA: Efficacy, safety and use of N,N-diethyl phenylacetamide, a multi-insect repellent, In: *Insect repellents; Principles, methods and uses*, Debboun M, Frances SP and Strickman D (Eds), Taylor and Francis Group, CRC Press, New York: USA, 2006. p341-345
4. Sharma P, Singh R, Jan M. Dose-dependent effect of deltamethrin in testis, liver, and kidney of wistar rats. *Toxicol Int.* 2014; 21(2):131-139.
5. Brown LD, Narahashi T. Modulation of nerve membrane sodium channel activation by deltamethrin. *Brain Res.* 1992; 584(1-2): 71-76.
6. Raj J, Chandra M, Dogra TD, Pahuja M, Raina A. Determination of median Lethal dose of combination of Endosulfan and cypermethrin in wistar rats. *Toxicol Int.* 2013; 20(1):1-5.
7. Adamson RH. The acute lethal dose 50 (LD50) of caffeine in albino rats. *Regul Toxicol Pharmacol.* 2016; 80:274-276.
8. Omidbakhsh N, Sattar SA. Broad-spectrum microbicidal activity, toxicologic assessment, and materials compatibility of a new generation of accelerated hydrogen peroxide-based environmental surface disinfectant. *Am J Infect Control.* 2006; 34(5): 251-257.
9. Chandra M, Raj J, Dogra TD, Rajvanshi AC, Raina A. Determination of median lethal dose of Triazophos with DMSO in wistar rats. *Asian*

ACKNOWLEDGEMENT

This research work did not receive any grant from funding agencies in the public, commercial, or non-profit organizations.

CONFLICT OF INTEREST

Conflict of interest declared none.

- Journal of Pharmaceutical and clinical Research 2014; 7(4): 64-67.
10. Garud A, Gautam A, Ganesan K, Kumar P, Prakash S, Jatav PC, Kumar A, Vijayaraghavan R. Acute toxicity studies of safer and more effective analogues of N,N-diethyl-2-phenylacetamide. *J Med Entomol* 2011 48: 1160-1166.
11. Cedergreen N. Quantifying synergy: a systematic review of mixture toxicity studies within environmental toxicology. *PLoS One.* 2014; 9(5): e96580.
12. Gad SC, Weil CS. *Statistics for toxicologists*. In *Principles and methods of toxicology*, Hayes AW (ed), 2nd edition. Raven Press, New York, 1989; 463-467.
13. Dixon WJ. The Up-and-Down Method for Small Samples, *Journal of the American Statistical Association* 1965; 60: 967-978.
14. Sharma VP. Health hazards of mosquito repellents and safe alternatives. *Current science* 2001; 80(3): 341-343.
15. Hughes MF, Edwards BC. In vivo dermal absorption of pyrethroid pesticides in the rat. *Journal of Toxicology and Environmental Health, Part A.* 2016; 79(2): 83-91.
16. Meepagala KM, Bernier UR, Burandt C, Duke SO. Mosquito repellents based on a natural chromene analogue with longer duration of action than N,N-diethyl-meta-toluamide (DEET). *J Agric Food Chem.* 2013; 61(39): 9293-9297.
17. Damalas CA, Eleftherohorinos LG. Pesticides Exposure, safety Issues and Risk assessment indicators. *Int.J.Environ .Res.Public Health* 2011; 8(5): 1402-1419.
18. Bradberry SM, CageSA, Proudfoof AT, Vale JA. Poisoning due to pyrethroids. *Toxicol Rev.* 2005; 24(2): 93-106.
19. Forshaw PJ, Ray DE. A novel action of deltamethrin on membrane resistance in mammalian skeletal muscle and non myelinated nerve fibres. *Neuropharmacology* 1990; 29(1) 71-81.
20. Swale DR, Sun B, Tong F, Bloomquist JR. Neurotoxicity and mode of action N,N Diethyl-meta-toluamide. *PLOS* 2014; 9(8): 1-11.

Reviewers of this article

Dr. Vinesh Kumar

Associate Professor, Pharmacy,LBS
College of Pharmacy, Udai Marg, Tilak
Nagar, Jaipur,India



Prof.Dr.K.Suriaprabha

Asst. Editor , International Journal
of Pharma and Bio sciences.



**Mr. Anubrata Paul M.Sc. Biotech
(Research)**

Affiliation

Department of Biotechnology, Natural
Products Research Laboratory, Centre for
Drug Design Discovery & Development (C-
4D) , SRM University, Delhi-NCR,Sonepat.



Prof.P.Muthuprasanna

Managing Editor , International
Journal of Pharma and Bio sciences.

We sincerely thank the above reviewers for peer reviewing the manuscript