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# Effect of spark EC 36 [combination pesticide] on the AchEase activity in plasma and Brain of wistar rats

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#### ABSTRACT

The organophosphates and pyretheroids presently used as mixed pesticide in agricultural practice, since the organophosphates are known to produce neurotoxicity combination pesticides one prepared by mixing pyretheroids with organophosphates to reduce its toxicity on mammals and at the same time it will have best pesticidal property due to the synergetic effect of its constituents. Hence a 28 days sub acute studies were designed by administering 1/10,1/20 and 1/40 <sup>th</sup> of LD50 dosage daily through oral gavages for 28 days. The Blood, Plasma and Brain samples were collected from both experimental and control Rats; the results were quite interesting to note that males were affected only at the higher doses whereas females showed dose dependent AchEase inhibition. When we refer the earlier works on the constituent pesticide of the combination it shows AchEase inhibition in all the experimental groups of both male and female. Hence it is evident that a short time sub acute toxicity studies on the biomarkers will give all the required data obtained in the long term study, Such type of short time sub acute studies will help us to locate type of a combination pesticide which is eco friendly, best pesticidal property with low mammalian toxicity. **Keywords:** AchEase, pesticide, brian



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#### INTRODUCTION

We are that there are number of possible ways in which humans can also be exposed to pesticides. Residues of pesticides contribute significantly to contamination of air, water, soil and food [1] and thus the toxic effects of these substances may have consequences even for consumers of food indirectly. In the olden days more acutely toxic pesticides have been used for suicide and murder [2]. The Organo- phosphates after entering the body of an organism reaches the cholinergic sites of the nervous system and inhibit the activity of the AChE by binding at its active sites. AChE inhibition, thus leads to the accumulation of Ach at nerve endings which in turn cause the disruption of the nervous activity resulting in excitation, paralysis and finally the death of the organism [3]. Hence recently new combination of pesticides have been introduced to reduce environmental pollution and at the same time to have a maximum action in killing the pest animals. The recent trend is to mix organophosphate along with pyrethroids in bringing out combination pesticide. The combination pesticides are gaining popularity in pest control programmes as they exhibit a broad spectrum of activity coupled with better efficacy and economy. But for the registration of these pesticides, acute toxicity data is sufficient and therefore the longterm toxicity of these compounds remains unexplored. Alternatives are required for long term studies as they are difficult to carryout, time consuming and expensive. Hence such type of study can explore all toxicity profile as expected in long term study.

#### MATERIALS AND METHODS

The present study was carried out to evaluate the short-term sub acute (28 days) toxicity and neurotoxicity of combination pesticide Spark EC 36 [triazophos 35% + deltamethrin 1%] in Wistar rats.The acute toxicity of these combination pesticides were carried out to arrive at the LD50 dose [4] from the LD50 dosage the sub acute doses were fixed as 1/10, 1/20 and 1/40 <sup>th</sup> of LD 50 dosage (4, 8 and 16 mg/kg B.W.) These dosed pesticides were dissolved in water and administered orally using oral gavage for 28 days.On the 29 <sup>th</sup> day the animals were sacrificed to collect brain and blood sample to screen the AchEase activity. AChE, a sensitive promarker of neurotoxicity which is widely distributed within the central nervous system (CNS) [5]. Some sub cortical areas like nucleus caudatus and globus pallidus are particularly rich in this enzyme [6]. It exists in several molecular forms (Taylor et al., 1987) of which the soluble globular form is present in the brain. AChE was estimated in whole blood plasma and tissues extracts by Ellmans's method [7]. This method estimates AChE using acetylcholine iodide (substrate) and dithiobisnitro benzoic acid. The enzymatic activity is measured by the yellow colour produced by thiocholine when it reacts with dithiobis nitrobenzoate ion.

Acetylcholine  $\xrightarrow{\text{enzyme}}$  Thiocholine +Acetate. Thiocholine + dithiobisnitro  $\longrightarrow$  benzoate yellow color.

The colour intensity can be measured on a spectrophotometer and the enzyme activity expressed as the rate of reaction per minute.



#### RESULTS

. In the results we see a marked inhibition in the blood and brain AchEase enzyme activity in the treated male and female animals..The present results explaining that sex also plays vital role in the AchEase activity, because male AChEase markedly differs from the female enzyme inhibition.In male rats, a significant decrease of AChE activity in blood was observed in the highest dose group (G IV) when compared to untreated control.Whreras in other doses there is less significant results observed but in case of female a dose dependent decrease of AChE activity was observed (Table 1, Figure 1 and 2). In plasma a dose dependent decrease of AChE activity was observed in both male and female rats treated with the pesticide (Table 1; Figure 1 and 2).Whereas in brain, a dose dependent steady decrease of AChE activity was observed in both male and female rats treated with the pesticide when compared with the untreated control (Table 1].

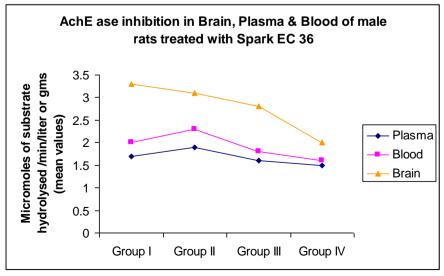
## Table 1. Levels of Acetyl cholinesterase in wistar rats treated with different concentrations of combination pesticide Spark EC 36

		pesi	licide Spark EC :	30		
	AchE					
Groups	(μ moles of substrate hydrolysed/minute/l (or) g)					
	Plasma		Blood		Brain	
	Male	Female	Male	Female	Male	Female
Groups I	1.854ª	1.856ª ±	2.026ª	2.064ª	3.266ª	4.01ª
	±	0.07	±	±	±	±
	0.11	0.07	0.14	0.16	0.13	1.24
Groups II	2.008ª	2.004	2.244ª	2.270	2.02ª	2.12 <sup>b</sup>
	±	±	±	±	±	±
	0.15	0.18	0.31	0.06	0.61	0.18
Groups III	1.674 <sup>b</sup>	1.612 <sup>c</sup>	1.844 <sup>b</sup>	1.720 <sup>c</sup>	2.23 <sup>b</sup>	1.42 <sup>c</sup>
	±	±	±	±	±	±
	0.05	0.06	0.08	0.07	0.27	0.22
Groups IV	1.235 <sup>c</sup>	1.783 <sup>d</sup>	1. 480 <sup>c</sup>	1. 295 <sup>d</sup>	1.857 <sup>c</sup>	1.714 <sup>d</sup>
	±	±	±	±	±	±
	0.16	0.15	0.16	0.12	0.19	0.16

Values are presented as mean ± standard Error

Values having similar superscripts are not statistically significant (p>0.05).

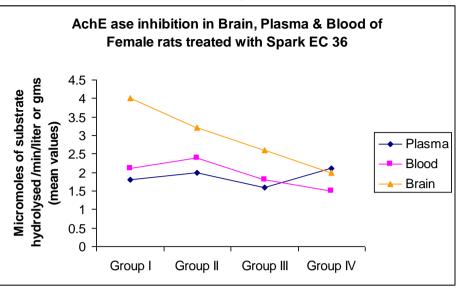
Figure 1



1(4)







#### DISCUSSION

Acute OP poisoning causes various neurological signs in human and experimental animals [8]. This includes behavioral changes; sleep disturbances, tremors, convulsions, coma and respiratory/circulatory failures. Early signs and symptoms of OP poisoning like depression, emotional lability, insomnia and tremors [9], exhibit as a result of the disturbances to the Central Nervous System due to AchEase inhibition and also due to reduced activity of blood AChE. From the present investigation it could be observed that the combination pesticide Spark EC 36, did exhibit the characteristic toxicity signs of its constituent insecticides like suppression of AChE (induced by organophosphates) and convulsions and tremors (induced by deltamethrin). Out of both the sexes it is evident from the present result that the males were affected only in higher doses whereas they dint show any marked inhibition in lower doses. So it could be understood that the toxicity of Spark EC 36 was not the same as that of the constitute pesticides namely triazophos and deltamethrin it may be due to the synergic effect of the constituents which attributes low mammalian toxicity with reference to AchEase inhibition [10,11].

#### **SUMMARY**

The combination pesticides exhibit toxicity in a different fashion compared to the toxicity of its constitute pesticides. From the earlier reports it is understood that long term toxicity of the combination pesticide in most of the cases vary compared to that of the individual pesticide in the combination .Hence long-term toxicity studies are very essential for combination pesticides. The present study is aimed to screen the neurotoxic biomarker AchEase using 28 days short-term sub acute studies.which can be used an alternative in the long-term toxicity studies..From the present study it could be understood that the combination pesticides behave differently in its toxicological profile when compared to the toxicity of the individual pesticides in combination. Spark EC 36 which may be due to the synergetic effect of its constituents.Hence a complete short term sub acute studies coupled with screening of biomarker AChEase will give most of the information obtained through



long term studies. Similar studies can also help to identify combination pesticides that have eco-friendly with lesser mammalian toxicity.

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