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On synergised and unsynergised pyrethrum. Their toxicity to *Dysdercus cingulatus* FABRICIUS and persistency of the toxic effect of the films

With 1 textfigure

Unfortunately a great confusion exists regarding the terminology for the joint action of an insecticide and any other substance capable of increasing the toxicity of the insecticide when applied in mixture or successively allowing a suitable calculated interval between the two treatments, as compared to the toxicities of both, the insecticide and the other substance when applied quite independently.

MACHT in (1929) expressed his views on 'Synergism' by suggesting that it is not a mere summation of the effects produced by two or more individual components but either one component may show antagonised action over the other or may be potentiated by the other.

Another term 'activation' was suggested by INMAN in the same year i.e. 1929 to express the increase in the physical efficiency of nicotine by adding another compound having the properties of an activator. Since then these two terms have been subjected to a great analysis and gave rise to various views suggesting different terms, in the hand of various toxicologists. BLISS (1939) suggested to accept two different types of actions, additive and synergistic. He further went on to classify the joint action into 3 categories, independent, similar and synergistic; antagonism was considered as negative synergism. SWISHER (1944) did not seem to accept the classification put forward by BLISS (1939) pointing out that BLISS's coneptionc of synergism fails to include many examples which should be supposed, of synergism; to this he quoted an example of *Tribolium* sp. giving 5.4% and 4.1 % kill when treated with sulphur dioxide and acetone separately but 89.5% mortality on subjecting to the mixture of both at the same concentrations and dose as that applied independently. He preferred the term activation, defining an activator as a substance capable of rendering another one more active. FINNEY (1947) suggested that since similar although separate effects are produced in a similar joint action, it is quite possible to substitute one poison for the other at a fixed proportion. HORSEFALL (1945) even went to the extent of saying that a tendency of loose use of such words exists. He thought of a sub-division of synergism into potentiated and supplementary types; attributing physical improvement of the performance of a toxic chemical and the exercise of similar physiological action at the same locus to the supplementary type of synergism.

Thus instead of making the whole thing simpler it is now getting more and more complicated and confusing. The authors feel that a simple clear definition incorporating all the aspects, as far as the classification of some of the substances

DOI: 10.21248/contrib.entomol.15.1-2.167-174

capable of increasing the toxicity of the other when applied in mixture or successively after allowing a definite calculated interval between the application, might prove useful and with this view we bring here some other views appealing us.

HEWLETT (1960) reviewed the joint action of insecticides and expressed his opinion that in cases when a component is inactive or negligible active on independent application but the mixture with the second component is more active than the second component alone, the first component may be categorised as synergist i. e. synergising the second component. This definition suffers the drawback that it does not include the aspect when two toxic components on mixing give a toxicity higher than what is given on independent application. CHADWICK (1963) proposed a definition for pyrethrum synergists almost on the lines similar to HEWLETT (1960), thus having the same lacuna. He thought that any substance which is not primarily an insecticide but which is capable of increasing the toxicity of pyrethrum independently of any effects external to the insect, may be termed as pyrethrins synergist. WADLEY (1945 & 1949) generalised the definition and included those synergists as well which are toxic even when applied independently i.e. insecticides. He defined synergism as the joint action of two materials producing the total effect greater than the sum of the two effects produced on applying independently. The authors although agreeing to a great extent with HEWLETT (1960) and CHADWICK (1963) but feel to incline towards WADLEY (1945 & 1949) whose conception of synergism is quite clear and accepts any substance, whether toxic or inactive, but capable of enhancing the toxicity of the mixture when mixed with another component, as synergist.

With this conception of synergism the authors in the present paper have touched two aspects; the toxicity of synergised and unsynergised pyrethrum to *Dysdercus cingulatus* and the persistency of the toxic effect of their films.

Material and Method

The resinous substances were precipitated by adding an equal volume of absolute alchohol to commercial 20% pyrethrum extract and leaving it for several hours. The resin free 20% pyrethrum was then obtained by filtering the solutions and was diluted with the solvent (Analar Petroleum Ether b.p. 100—120 and liquid paraffin mixed in the ratio of 4:1) to prepare a 5% stock solution.

Sulphoxide technical supplied by S. B. PENICK & Co., New York, was diluted with solvent and 2% stock solution was prepared.

The stock solutions were then further diluted to obtain the required different concentrations.

Dysdercus cingulatus FABRICIUS collected from the botanical garden of University MAHARANI's College for Women were bred in the laboratory at room temperature 28 °C ± 2. About 50 insects were allowed to breed in muslin topped big beakers and were provided with either branches with leaves, seeds and flowers or with small malvaceous plants. After allowing them a suitable period to lay eggs, the adults were removed to other beakers. The nymphs thus were allowed to grow

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freely. About 50 days were required to complete the life cycle. Some soil and leaves were also kept at the bottom of the beaker to provide the insects with natural surroundings to lay eggs.

Application of insecticides

The following method was employed for applying the insecticides. 7 cm. filter papers were impregnated with 0.5 ml. of solution by means of a pipette and a thin film of the insecticide was provided on the rough surface of the filter paper. Petroleum ether present in the solution facilitated even distribution of the insecticide on the filter papers. A few hours were allowed to let the petroleum ether to evaporate from impregnated surface. The experimental insects were thus left to crawl over the treated surface for the desired period confined within the muslin topped glass funnels. It was ascertained that the insects were all the time in contact with the treated surface and did not crawl on the walls of the funnel.

Assessment of the toxic effect

Knockdown response was taken to assess the toxic effect. All those insects which were found lying on their backs after the treatment were taken as effected or knockdown. Those, which were on their legs but fell on their backs on a slight touch never to stand again on their legs, were also included as effected.

The experiments were conducted at room temperature 28 °C ± 2. Angular transformed values were plotted on ordinates against the concentrations spaced logarithmically as abscissae and the regression lines were drawn. The median response doses were calculated to compare the toxicity.

Design of Experiments

Experiment No. 1 (with pyrethrum)

Out of 225 adult *Dysdercus cingulatus* taken at random from the population, 3 batches of 25 each were assayed against .125%, .25%, & .5% pyrethrum. The insects were exposed to the treated surface for 45 minutes. Two replicates each of 25 insects were run for each concentration. The knockdown percentage was calculated after the treatment (Table 1).

The treated filter papers were then kept and it was ensured that the treated surface does not come in contact or rub with any substance.

Table 1
Knockdown response of *Dysdercus cingulatus* treated with pyrethrum

	Concentrations					
	.125%		.25%		.5%	
	A	B	A	B	A	B
Percentage response	0	0	24	0	48	24
Replica I	0	0	20	0	68	0
Replica II	8	0	0	20	36	36

A = Fresh insecticide film.
B = 6 days old insecticide film.

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Experiment No. 2 (with pyrethrum)

After 6 days a similar experiment with 225 insects was repeated by subjecting the adults to 6 days old insecticide films on the treated filter papers, and knockdown percentage was calculated (Table 1).

Experiment No. 3 (with pyrethrum + sulphoxide)

.125%, .25% and .5% pyrethrum was mixed with the same concentrations of sulphoxide, in the ratio of 1:1 by volume.

225 insects were taken at random from a population for treatment. 3 batches of 25 each were treated with (.125% pyrethrum + .125% sulph.), (.25% pyrethrum + .25% sulph.), & (.5% pyrethrum + .5% sulph.). The insects were exposed to the treated surface for 45 minutes and two replicates each of 25 insects were run for each mixture. The response recorded was knockdown percentage after the treatment (Table 2).

The treated filter papers were then kept for 6 days as in the experiment no. 1.

Experiment No. 4 (with pyrethrum + sulphoxide)

After 6 days an experiment similar to experiment no. 3 was repeated with 6 days old insecticide films spread on the filter papers and knockdown response in percent was calculated (Table 2).

Table 2
Knockdown response of *Dysdercus cingulatus* treated with pyrethrum + sulphoxide

	Concentrations					
	.125% + .125%		.25% + .25%		.5% + .5%	
	A	B	A	B	A	B
Percentage response	8	0	36	0	76	24
Replica I	0	0	24	0	36	0
Replica II	12	0	0	36	68	60

A = Fresh insecticide film.
B = 6 days old insecticide film.

Results

The concentration response regression lines and the median response concentration calculated for pyrethrum and pyrethrum + sulphoxide for *Dysdercus cingulatus* assayed against both are given in figure 1 and table 3 respectively, which also include the concentration response regression lines and the median response concentration for 6 days old films of both pyrethrum and the mixture of pyrethrum and sulphoxide.

Non-toxicity of sulphoxide concentrations used in the present investigation was ascertained by the preliminary experiments.

DOI: 10.21248/contrib.entomol.15.1-2.167-174

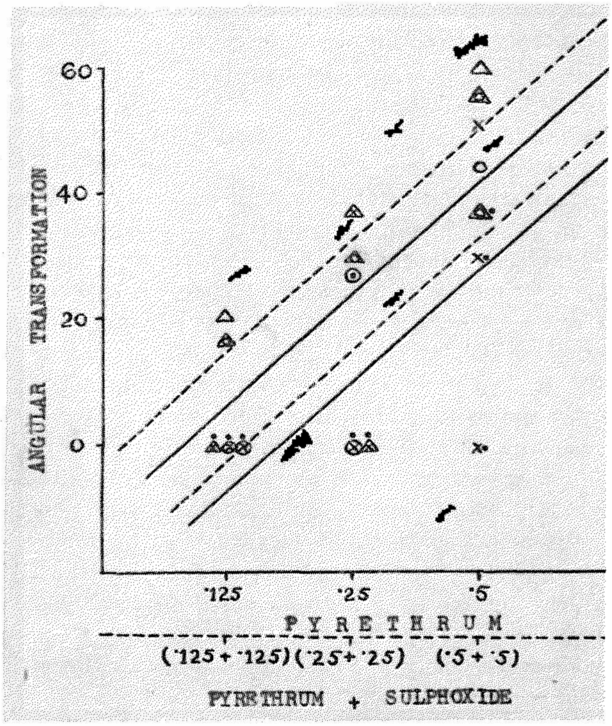

Fig. 1. Response of *Dysdercus cingulatus* FABRICIUS to pyrethrum and pyrethrum + sulphoxide.

○, ——— Pyrethrum treated (on 1st day of treatment);
●, ——— Pyrethrum treated (on 7th day of treatment);
△, - - - - - Pyrethrum + Sulphoxid treated (on 1st day of treatment);
×, - - - - - Pyrethrum + Sulphoxid treated (on 7th day of treatment)

Table 3

Median Response Concentrations

Pyrethrum		Pyrethrum + Sulphoxide	
Fresh film	6 days old film	Fresh film	6 days old film
.6%	1.05%	.425%	.858%

Discussion

The laboratory experiments have proved pyrethrum very effective to *Dysdercus cingulatus* a notorious pest of cotton and other malvaceous plants. On an average above 50% *D. cingulatus* were knockdown, most of them never to recover, on giving a contact treatment with .5% pyrethrum for 45 minutes only. Most of the authors in our country (SAXENA, 1955; LAL, 1955) have recommended either dusting with 5% BHC or spraying with .25% DDT to control this pest but hardly any mention has been made of pyrethrum. May be it is due to high cost

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of pyrethrum which thus may not be economical. Experiments with synergist sulphoxide in mixture with pyrethrum were therefore performed in order to find out whether the mixture proves to be more toxic than what pyrethrum alone is, so that by reducing the pyrethrum concentration by adding sulphoxide the cost may also be brought down. The concentration response regression lines drawn for pyrethrum and pyrethrum + sulphoxide are not coincident and the median response concentrations calculated for both show that a lower concentration of pyrethrum + sulphoxide as compared to the concentrations of pyrethrum alone can give 50% knockdown. The median response concentration (MRC) for pyrethrum .6% and for pyrethrum + sulphoxide .425% clearly show that the sulphoxide acts as synergist and the mixture becomes much more toxic than what pyrethrum alone is. This should also be considered that in the present work, to .125%, .25% & .5% pyrethrum concentrations, the sulphoxide at the same concentrations in the ratio of 1:1 was added. The mixtures thus were having half the percentage of original pyrethrum concentrations, i.e. .0625%, .125% & .25% but the results show a high knockdown percentage than what was obtained with .125%, .25% & .5% pyrethrum. This again clearly indicates that the toxicity of pyrethrum was considerably increased, as shown by the fall in median response concentration as well, by adding sulphoxide, which thus acted as synergist. Though the authors have not calculated the cost as experiments on large scale could not be performed due to certain reasons, yet considering the low price of sulphoxide we strongly feel that the use of pyrethrum in mixture with sulphoxide should prove quite economical.

An experiment was conducted to make a comparative study of the persistence of the toxic effect of the films of synergised and unsynergised pyrethrum. The concentration response regression lines do not coincide and the median response concentrations (MRC) calculated show considerable difference. MRC of .6% for fresh unsynergised pyrethrum film goes up to 1.05% for 6 days old pyrethrum film, showing a considerable decrease in the toxicity on 7th day. Similarly MRC of .425% for fresh synergised pyrethrum film (pyrethrum + sulphoxide), is increased to .858% for 6 days old synergised pyrethrum film. As discussed above the addition of similar concentrations of sulphoxide to pyrethrum in the ratio of 1:1 leave with half the original concentrations of both in the mixture (i.e. half of .125%, .25% & .5% of pyrethrum and sulphoxide). But even these half concentrations are more toxic than the original pyrethrum concentrations (.125%, .25% & .5%); thus the fall in toxicity of synergised pyrethrum, (when the concentrations were half of the original ones), as shown by rise in MRC from .425% to .858% as compared to the decrease in the toxicity of unsynergised pyrethrum shown by rise in MRC from .6% to 1.05% apparently points to the longer persistence of the toxicity of synergised films. But the authors feel that since the mixture of pyrethrum and sulphoxide although at a level of ½ the concentrations of both (on mixing) becomes more toxic than the original concentrations, obviously will take a longer period to loose the toxicity. SAXENA (1958) pointed out that the higher the concentration for a given time of exposure to the insecticides, the longer is the duration of its toxic effect. HEWLETT (1951) studied the effect of piperonyl

DOI: 10.21248/contrib.entomol.15.1-2.167-174

butoxide on pyrethrum film persistence, and thought it useless to compare 1.3% pyrethrum + 5% piperonyl butoxide with that of 1.3% pyrethrum alone as the mixture being more toxic obviously has more toxicity to loose. Hence the authors inspite of various views, DOVE (1947) claiming that piperonyl butoxide stabilizes pyrethrins in surface films, PHIPERS & WOOD (1957) favouring the reported stabilization of pyrethrins by piperonyl butoxide, BROOKE (1958) accepting the stabilization by piperonyl butoxide in water and GLYNNE JONES (1961) suggesting that in sunlight PB is an effective stabilizer for pyrethrum, support O'FARRELL, JONES & BRETT (1949), HEWLETT (1951) and BLACKITH (1952) and incline to accept HEWLETT (1951) that since the toxicity of the synergised pyrethrum is increased, the mixture has more toxicity to loose thus could be expected to produce more persistent films irrespective of the possibility that sulphoxide may have some special stabilizing effect upon the pyrethrum.

Acknowledgements

We wish to express our deep gratitude to Mrs. S. BHARTIYA, the Director, University MAHARANI's College for Women, Jaipur, for her prompt and liberal, moral as well as financial help, keen interest and encouragement which enabled us to complete this investigation. Thanks are also due to Prof. L. S. RAMASWAMI, Head of Zoology Department for his encouragement throughout the work.

Summary

Dysdercus cingulatus FABRICIUS has been found very susceptible to pyrethrum and pyrethrum + sulphoxide. Sulphoxide when mixed with the same concentrations of pyrethrum in the ratio of 1:1, act as synergist increasing the toxicity of the mixture and thereby giving a higher knockdown percentage of *Dysdercus cingulatus*. It is felt that this mixture may prove economical to control this pest, of cotton and other malvaceous plants.

In view of the suggestion that sulphoxide increases the persistence of the toxicity of pyrethrum film, an experiment was specifically designed and it is suggested that irrespective of whether sulphoxide has any stabilizing effect on pyrethrum, since the pyrethrum becomes more toxic on adding sulphoxide, a longer persistent effect could be expected as the mixture of pyrethrum and sulphoxide being more toxic would take longer to loose its toxicity.

Zusammenfassung

Es wurde festgestellt, daß *Dysdercus cingulatus* FABRICIUS sehr anfällig gegen Pyrethrum und Pyrethrum plus Sulphoxid ist. Wenn man Sulphoxid mit der gleichen Konzentration von Pyrethrum im Verhältnis 1:1 mischt, wirkt es synergistisch und verstärkt die Toxizität der Mischung, wodurch ein höherer Prozentsatz in der Vernichtung von *Dysdercus cingulatus* erzielt wird. Man kann annehmen, daß sich diese Mischung als ein wirtschaftliches Mittel zur Bekämpfung dieses Schädlings erweisen wird, der Baumwolle und andere Malvengewächse befällt.

In Anbetracht der Vermutung, daß Sulphoxid die Beständigkeit der Giftwirkung der Pyrethrumschicht erhöht, wurde ein Versuch speziell hierfür entwickelt. Es wird die Ansicht vertreten, daß unabhängig davon, ob Sulphoxid einen stabilisierenden Einfluß auf Pyrethrum ausübt, eine längerdauernde Wirksamkeit erwartet werden kann, da das Pyrethrum durch den Zusatz von Sulphoxid giftiger wird und deshalb die giftigere Mischung von Pyrethrum und Sulphoxid ihre Giftwirkung langsamer verliert.

Резюме

Было установлено, что *Dysdercus cingulatus* FABRICIUS очень восприимчив к пиретруму и пиретруму с сульфоксидом. Если смешать сульфоксид с пиретрумом той же концентрации в соотношении 1:1, то проявляется синергизм

DOI: 10.21248/contrib.entomol.15.1-2.167-174

действия и усиливается токсичность смеси, чем достигается более высокий процент уничтожения *Dysdercus cingulatus*. Можно предполагать, что эта смесь окажется выгодным средством для борьбы с этим вредителем, который поражает хлопок и другие мальвовые растения.

В связи с предположением, что сульфоксид повышает устойчивость ядовитости слоя пиретрума, был проведен специально для этого опыт. Автор того мнения, что независимо, оказывает ли сульфоксид стабилизирующее действие на пиретрум или нет, можно ожидать более длительного действия, так как пиретрум в результате прибавки сульфоксида становится ядовитее. Эта более ядовитая смесь пиретрума и сульфоксида медленнее теряет действенность.

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DOI: 10.21248/contrib.entomol.15.1-2.167-174