دراسة التأثير السبتوكومائى فى حالة التسمم بمادة السيولين على أرانب الشنشلا

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أجريت تجارب على أرانب الشنشلا وذلك باعطاء جرعات مختلفة لمجموعات منها من مادة السيولين وقد قسمنا هذا البحث الى ثلاث أجزاء :-

الجزء الأول : وقد أختص هذا الجزء بتحديد الجرعة السامة التى تقتل ٥٠٪ من أرانب الشنشلا وقد وجدت ٧ره مجم/كجم بحدود ١٩ / ٢٠ درجة ثقه هى ٢٣ره - ٧٩ر٦ مجم/كجم .

الجزء الثانى : أحتوى هذا الجزء على دراسة الأعراض والصفه التشريحية كذلك بعض التحاليل لأنزيم كولين استريز المصلى وكرات الدم الحمراء بالأضافة الى تعيين نسبة السكر بالدم وقد دلت النتائج ان مادة السيولين تسبب تثبيط فى هذا الانزيم كذلك وجود انخفاض فى معدل السكر وقد أوصينا باعطاء الجلوكوز بالاضافة الى العلاجات المعروفة للتسمم بالمبيدات الحشرية الفسفورية فى حالة التسمم بهذه الماده .

الجزء الثالث : صممت هذه التجربة لمعرفة تأثير مادة السيولين على مكونات الدم وقد دلت النتائج على وجود اختلاف معنوى فى عدد كرات الدم الحمراء والبيضاء ولكن هذا الأختلاف لم يكن يتناسب مع الجرعة المعطاه للحيوان .

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THE CYTOCHEMICAL CHANGES INDUCED BY CYOLANE IN CHINCHILLA RABBITS

(With 9 Figures)

By

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(Received at 19/6/1977)

SUMMARY

The oral LD_{50} of Cyolane was found to be 5.7 mg/Kg which grouped the compound under very toxic insecticide.

The effect of Cyolane showed to have an inhibitory effect on both red cells and serum cholinesterase and depression in blood glucose. There was no characteristic post mortum finding except the lungs showing congestion.

Haematological studies showed that Cyolane cause an increase in R.B.Cs. and W.B.Cs counts.

INTRODUCTION

Since the chlorinated hydrocarbon accumulates unchanged in animal tissues and have adverse effect on ecology. They are now replaced by low toxic organophosphates insecticides which are relatively rapid hydrolyzed and hence have less cumulative and ecological effect (FOWLER and MAHAN 1971, LOWLESS et al. 1972). However, they may cause contamination of the atmosphere, water surfaces and animal food in the vicinity of cotton field treated with pesticides by aircraft or ground spray.

Agricultural practices in Egypt include irrigation water. Egyptian farmers are unaware of the dangerous effect of feeding animals on sprayed plants, this ecological factors increases the possibility of accidental intoxication of animales and birds. Animals and birds living in or near agricultural areas may also be exposed to contamination when

pesticides are applied to cotton field by aircraft.

GOLD et al. (1957) studied the metabolic changes following acute cholinesterase inhibition by parathion and sarine in dog which developed both respiratory and marked elevation of inorganic phosphorus and glucose. They concluded that excessive release of adrenaline is probably responsible for hyperglycaemia. MURPHY (1966) showed also the hyperglycaemia effect of Delnav and Deptrex in rat.

Increased blood glucose level and possible correlation between the hyperglycaemia and degree of exposure to Malathion in fowls has been studied by UPPEL (1970). He concluded that the peak of glulose can be estimated at evidence of poisoning.

KAMEL et al., (1973) reported that after oral administration of Phosvel to rabbits in dose of LD_{50} found no significant rise in blood glucose.

Local application of Malathion, Lindan, Dieldrin and DPT have been reported to cause significant changes in haemoglobin and erythrocyes (STRIVASTAVA et al. 1960).

REHFELD et al. (1969) mentioned that there was no apparent effect on the haemoglobin level in one day old chicks after feeding Malathion at the level of 1000 P.P.M.

Some haematological studies of blood of 7 rabbits under the effect of Trichlorophon (NEGAVON) were carried out by PETRITCHEV and LAZANOV (1969) tested before and after 2,4,24, and 72 hours after I/V injection of 50 mg/Kg.

Trichlorophon showed arise in total leucocytic count about the fourth hour,followed by a slow fall tobelow the initial level. The erythrocyte sedementation rate was accelerated after 2 and 24 hours. Moreover,HOTHI, (1970) observed a decrease in haemoglobin, erythrocytes counts and packed cell volume and increase in leucocytic count and erythrocytic sedementation rate in buffalo calves after exposure to toxic doses of

Malathion. POLOZ and KOKHTYUK (1971) studied the effect of Dimethoatal on the haemogram of sheep after chronic poisoning. They showed that white cell counts increased while red cell counts and haemoglobin concentration were reduced. GUPTA and PAUL (1972) investigate the effect of dietary intake of Malathion on blood picture and plasma electrolytes in chicken and found the W.B.Cs count increase slightly while there was no change in R.B.Cs count, P.C.V. and E.S.R. and blood clotting time. VARTIC et al. (1972) studied the effect of Bubulin in sheep found that the Hb. content increased during the period of observation. Moreover, there was steady rise in monocytes, basophils and neutrophils in dog.

Although comprehensive, knowledge is available on distribution and concentration of pesticides in Egypt, information on detailed toxicological effect which are likely to be produced on physiology, behaviour and breeding sucess are largely speculative and need further studies.

Cyolane (2 diethoxyphosphinylimino) 1, 3-dithiolane) acts on insects as contact and stomach poison. It is used on commercial scale in the Middle East for control of egyptian contton leafworm.

In the available literature, it could be found no infrormation concerning the specific toxic effects of Cyolane. Therefore it was deemed necessary to evaluate its toxic effects in order to shed more light upon its toxicity.

MATERIALS AND METHODS

Cyolane (2-" diethoxyphenylimino " 1, 3-dithiolane) 99.5% purity supplied by American Cyanimide Co. was given orally to rabbits in 0.5% aquous sol. using stomach tube all over our work. Chinchilla rabbits were obtained from Public Health and Vaccine Institute (Egypt) kept in metal cages giving food and water ad-libitum.

Eighty rabbits were used for LD_{50} determination (LITCHFIELD and WILLCOXON, 1949).

For biochemical,haemattological and symptomatological studies three experiments were carried out.

Exp. I:

Eight groups, each contain 8 rabbits administered 3 mg/Kg Cyolane, then each group slaughtered at different period (1,3,6, 12,24,48, 72,and 120 hours).

Exp. II:

Three groups each contain 3 animals adminstered one dose of Cyolane (6,12,and 18 mg/Km), all animals were slaughtetered one hour postadminstration.

Exp. III:

Three groups of rabbits each contains 10 animals given 7 doses at two days intervals. The first group was given 0.5, 0.7, 0.9, 1.1, 1.3, 1.7, 1.9 mg/Kg, the second group given 0.5 mg/Kg at each interval, the thrid group given 0.5, 0.4, 0.3, 0.2, 0.1, 0.08, 0.06, and 0.04 mg/Kg.

Animals were slaughtered at the end of each experiment.

Blood obtained in each experiment was examined for glucose (FRANK, et al. 1950), Red cells CHE (FLEISHER and POPE 1954) serum CHE (BIGGS et al., 1958) also haematological studies of R.B.Cs, W.B.Cs counts, Hb. conentration, P.C.V.,E.S.R. and leucocytic differential count were done.

The results obtained were compared with control group in each experiment which adminstered distilled water only.

RESULTS

The oral LD_{50} of CYOLANE to chinchilla rabbits was found to be 5.7 mg/Kg with 19/20 confidence limits of 5.23-5.79.

The effect of dosing animals with 3 mg/Kg showing statistical significant lowering, at probability in true and pseudocholinesterase activities while R.B.Cs, W.B.Cs counts, E.S.R., Hb., P.C.V. and glucose

level in the blood are significaly raised (Fig. 1, 2) and the results of leucocytic differential count are showning Fig. 3.

The symptoms observed in animals received 3.0 mg/Kg B.W. began 45 minutes post administration and continued for 3 hours after which the animal return normal. Symptoms were mainly of nicotinic type (Slight clonic convulsion, with eruction of the ears). Animals huddle in the corner of the cages showing increase susceptability to external stimuli, somewhat constricted eye pupiles.

Animals received 6, 12 and 18 mg/Kg B.W. the symptoms were of muscarinic (Salivation, constricted pupils, diarrhea, urination, and respiratory destress) and nicotinic (Clonic and tonic convulsion) in addition to paralysis of the hind limbs. The severity and onest of symptoms were dose related.

The results of bicohemical and haematological investigation were in Fig. 4,5,6.

Animals received gradual increasing, constant and decreasing doses showed clinical manifestation after the 5th., 7th and 10 days of adminstration respectively and symptoms were as previously described.

The results of biochemical and haematological studies of experimental III showen Fig. 7, 8, & 9.

The post mortem finding of animals died was not charactteristic only congestion of lungs, petechial hameorrages in the liver with distended gall bladder, other wise all organs appeared normal.

DISCUSSION

In Egypt there is no standard system of evaluation and registration of pesticides from the toxicological point of view especially those used for agricultural purposes.

The LD 50 determination indicate that Cyolane is a very toxic insecticide and can be listed under the extreemly toxic substances used

for agricultural purposes (KHAN and HAUFE, 1972).

Although the cholinesterase inhibition is not the main factor responsible for symptoms produce at a dose of 3 mg/Kg it is a sensitive indication of "Cyolane" toxicity as poisoned animal after the first three hours showed no symptoms while pseudo and red cell cholinesterase still have significantly injibited, so it can be used as a useful test for diagnosis of such type of toxicity.

The inhibition of cholinesterases in experiment II was dose related and the muscarinic symptoms not observed in 3 mg/Kg dose (Exp.1) was observed in addition to nicotine-like and other C.N.S. effects manifested by paralysis of the limbs which is motor but not sensory as the animals respond to external stimuli has been observed.The effect of Cyolane as neurotoxic organophosphorus needs further studies to investigat the enzymological or pathological changes which may accompany this type of paralysis.

In experiment III all animals receiving doses during 15-days showed the symptoms of acetylcholine poisoning. Although the total dose received by group (3) was only 1.66 mg/Kg yet showed symptoms of muscarinic and nicotinic effects which are not observed by giving, 3 mg in one dose during acute toxicity (Exp.1) this explains that animals in case of subchronic cyolane poisoning differ in their response to symptomatic dose than acute toxicity.

From our previous results it can be suggested that cyolane may have an effect on adrenal cortex specially in zona fasciculate and zona reticulata with resultant hindering of the biosynthesis of glucosteroids which normally have insuline antagonistic proparties and the first rise observed in results (Exp. 1) may be due to impaired peripheral utilization of glucose, than at 120 hours nearly all Cyolane has been excreted and animals returned to normal.

In experiment II dose of 6 mg/Kg resulted in hyperglycaemia possibly due to exerting a stimulant effect on adrenal medulla cousing a release

of adrenaline, while 12 and 18 mg/Kg resulted in hypoglycaemia due to adrenal medulla depression.

In experimental III the total of 1.68 mg/Kg was not sufficient to cause alteration in carbohydrate metabolism while the total dose of 4.0 and 9.6 mg/Kg have an effect on adrenal gland and produce the effect through impairment of hormonal function.

Whatever the cause of hypoglycaemia, we advise to give glucose injections in treatment of Cyloane poisoning as it may have a beneficial effect beside the well known parameters in treatment of organophosphorus insectisides poisoning.

The difference between our result and earlier ones (GOLD et al., 1967, MURPHY by 1966, UPPEL 1970, Kamel et al.1973) may be due to either difference in chemical structure and or optimal doses responsible for this effect.

Our results indicate that there was a correlation between CHE level and heamatological picture and this is in agreement with SWITSKY et al. (1949).

Many clinical laboratories have attempted to use the changes observed in haematological pictures during intoxication by pesticides (STRIVASTAVA et al. 1966; HOTHI, 1970, and VARTIC et al.1970) to demonstrate the state of the subject under study. The fallacy of employing these changes as diagnostic aids is that many diseases and physiological alteration produce similar changes in the blood picture and therefore can not be considered specific for any one abnormal conditions.

The abnormalities which were observed by the previous authers are compared to values which are accepted as being normal.

Since it is rare to find a completely normal subject and many differnce appear in blood pictures of healthye subjects who for all practical purposes are normal as the difference in sex, age, physiological

state, dietary intake and analytical procedure employed make the comparison even more difficult along with the degree of abnormalities.

The determination of normal values should be made from accumulation of data from significant number of apparently healthy animals kept under similar conditions. It is also important that the same technique be employed throughout and preferably the same individual should be responsible for the analysis. We can conclude that the determination of blood pictures in organophosphorus poisoning can not be used as a main factor in diagnosis of poisoning but only to demonstrate the state the subject under study.

As the examination of P.M. shows no specific lesions, it can not be used niether as a main or additional factor in diagnosis of Cyolane poisoning.

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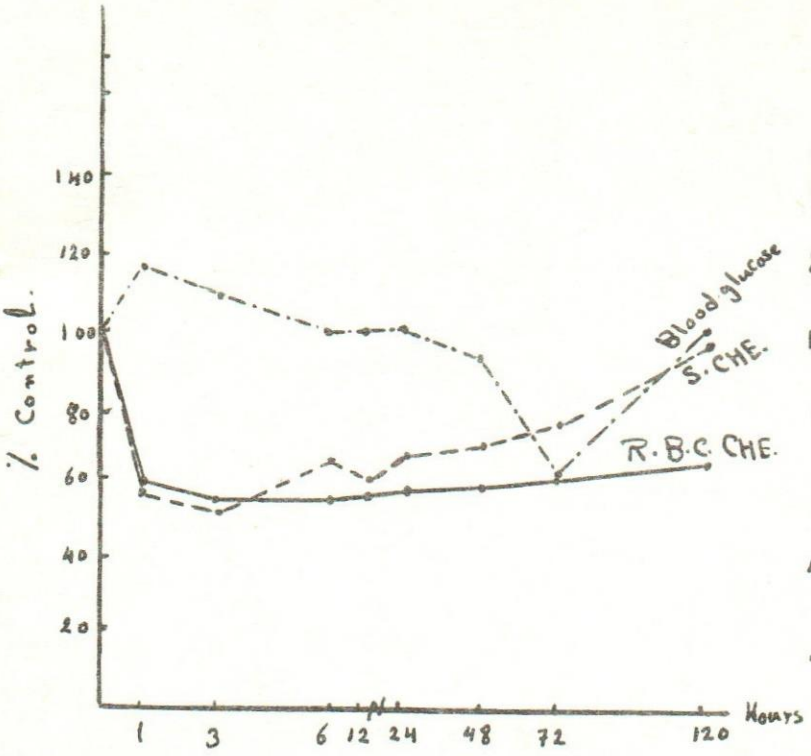

Fig. 1 – Changes in CHE.s. and blood glucose at different periods after oral administration of 3 mg/Kgm.

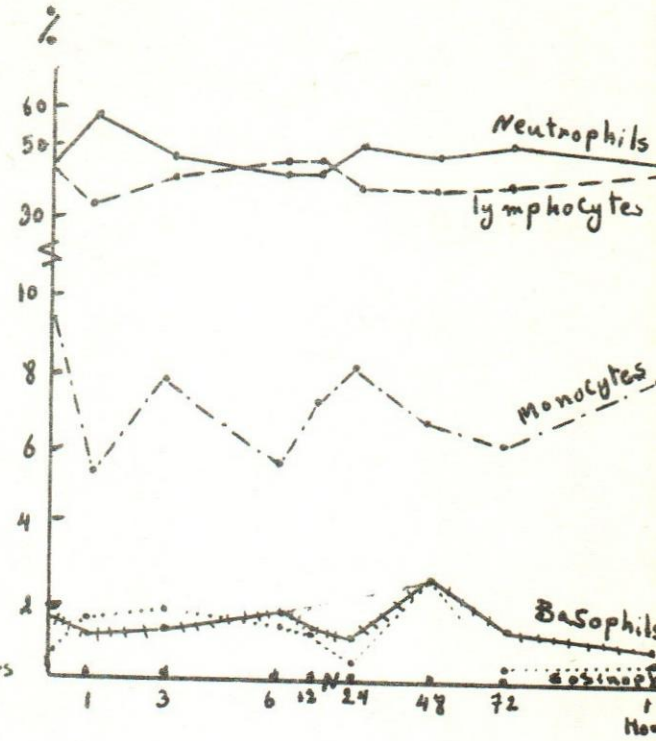

Fig-3. Leucocyte differential count at different periods after administration of 3 mg/Kgm.

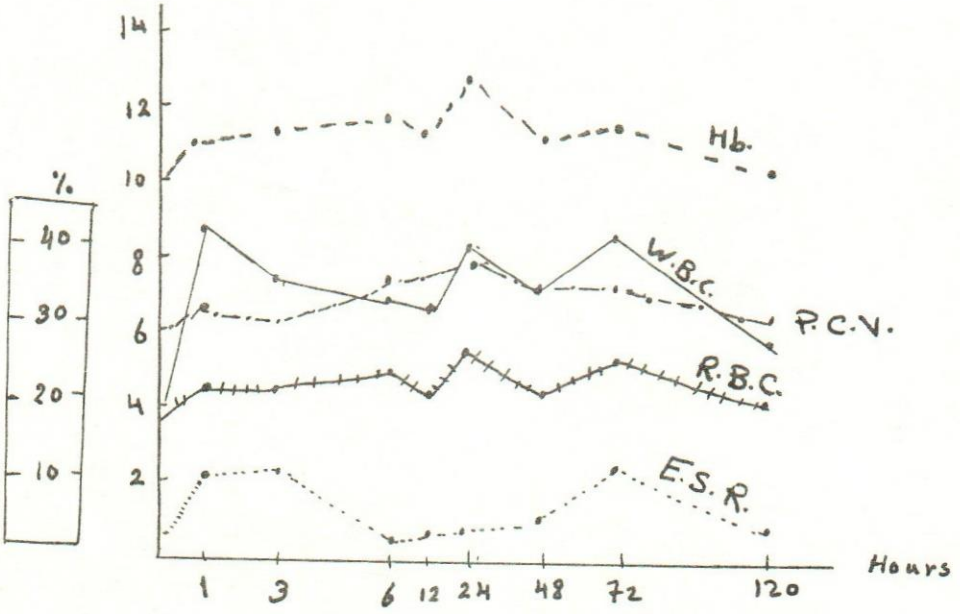

Fig-2. Changes in Haemogram after dosing of 3 mg/Kgm. at diffenent periods.

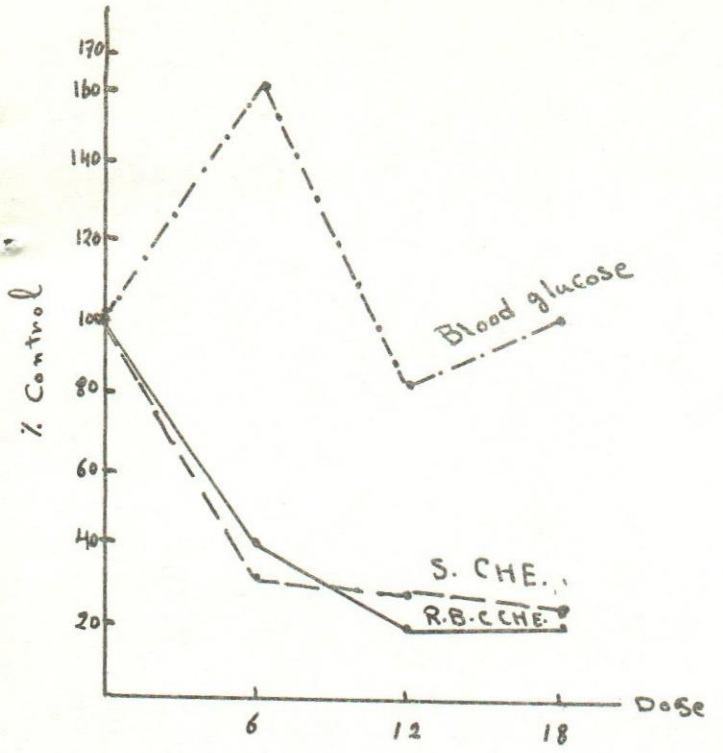

Fig. 4- Effect of different doses of Cyolane on Cholinesterase and blood glucose level.

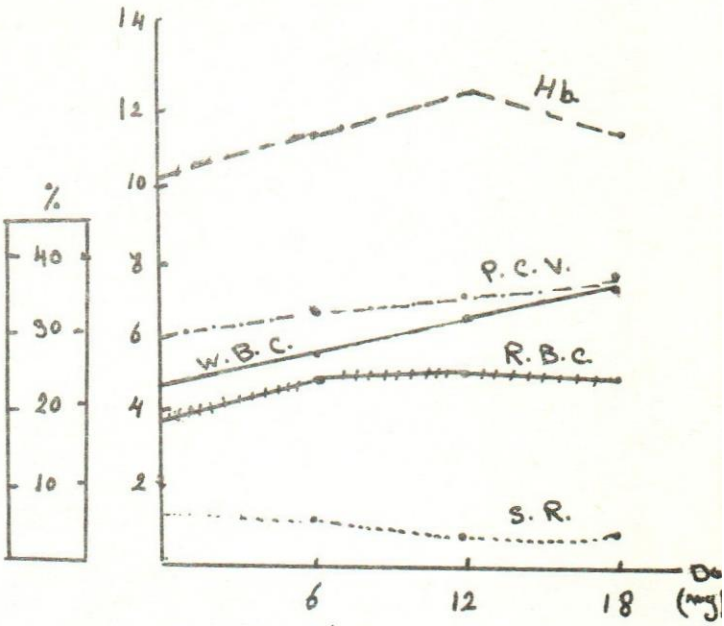

Fig. 5. Changes in Haemogram under the effect of Cyolane in diff doses.

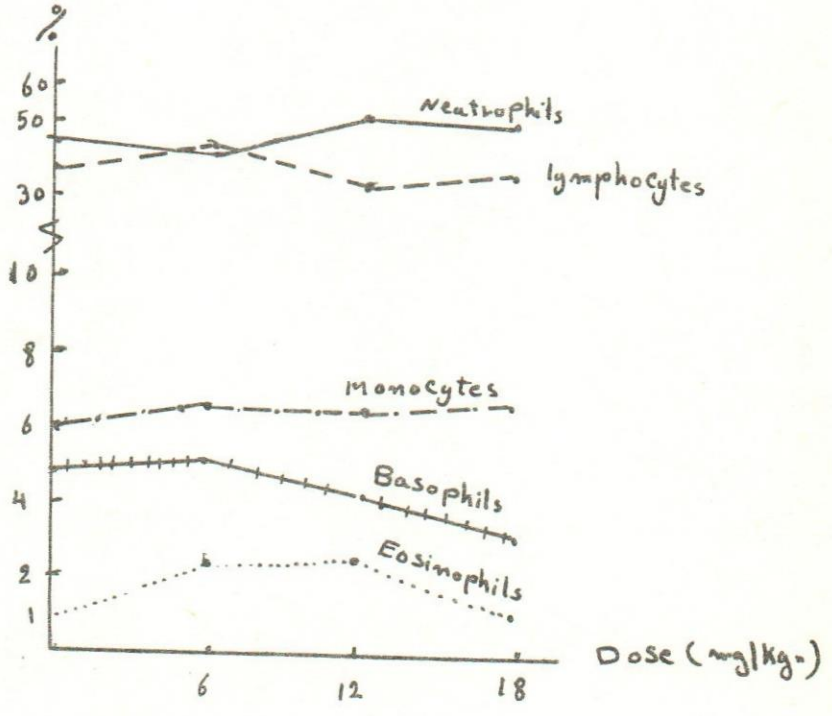

Fig. 6- Differential Leucocytic Counts after adminstration of 3 different doses (6, 12, and 18 mg/Kgm)

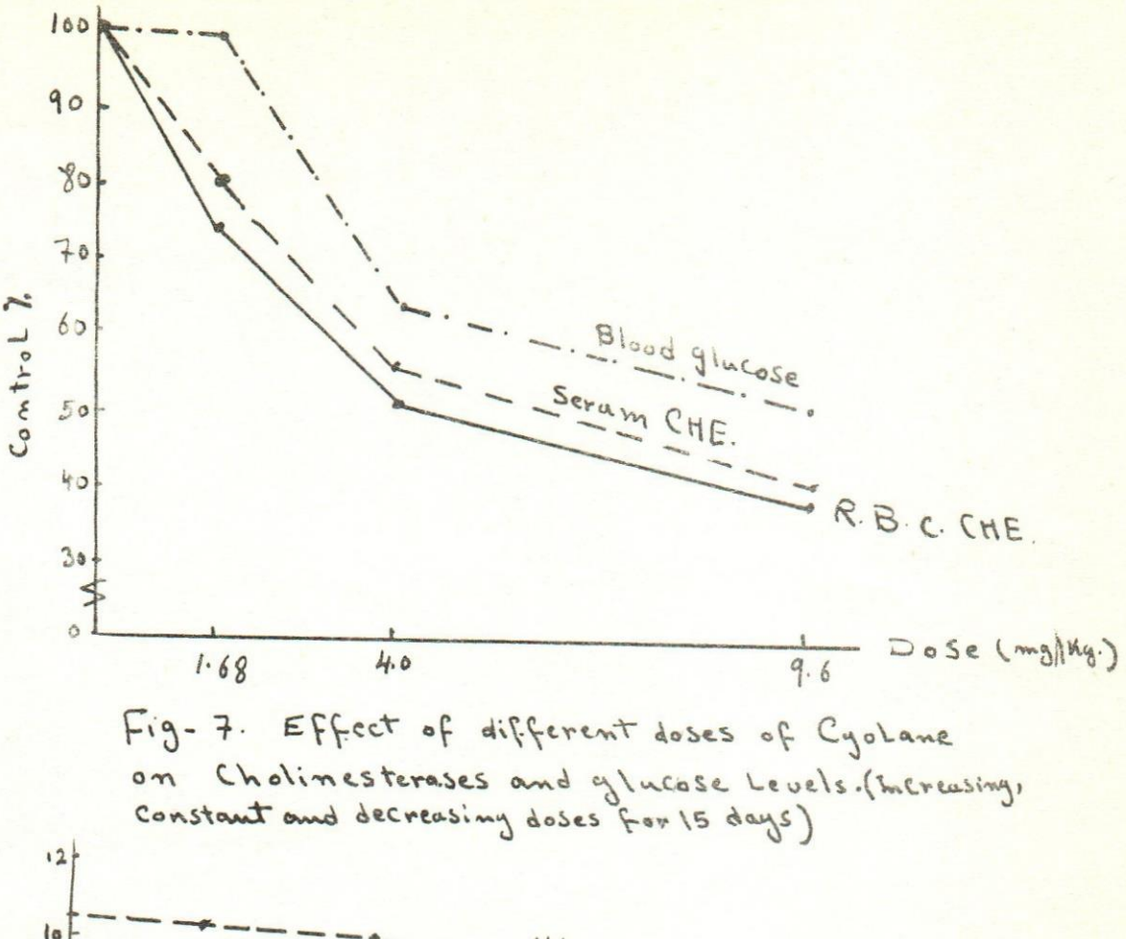

Fig- 7. Effect of different doses of Cyolane on Cholinesterases and glucose Levels. (increasing, constant and decreasing doses for 15 days)

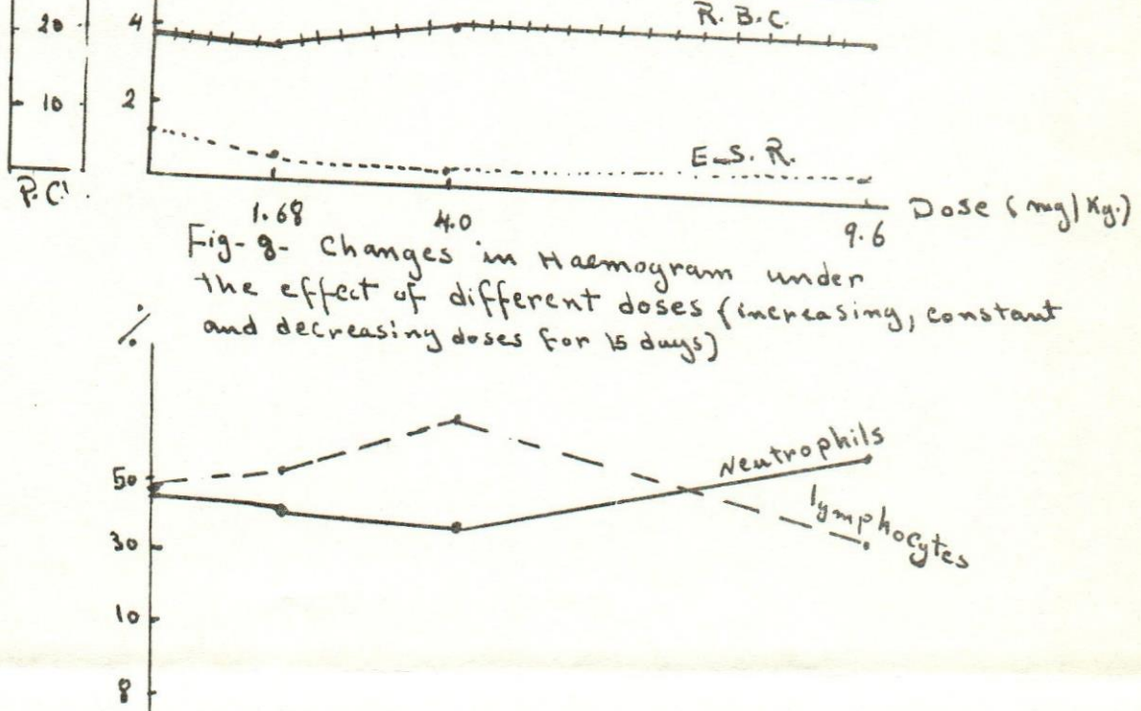

Fig- 8- Changes in Haemogram under the effect of different doses (increasing, constant and decreasing doses for 15 days)

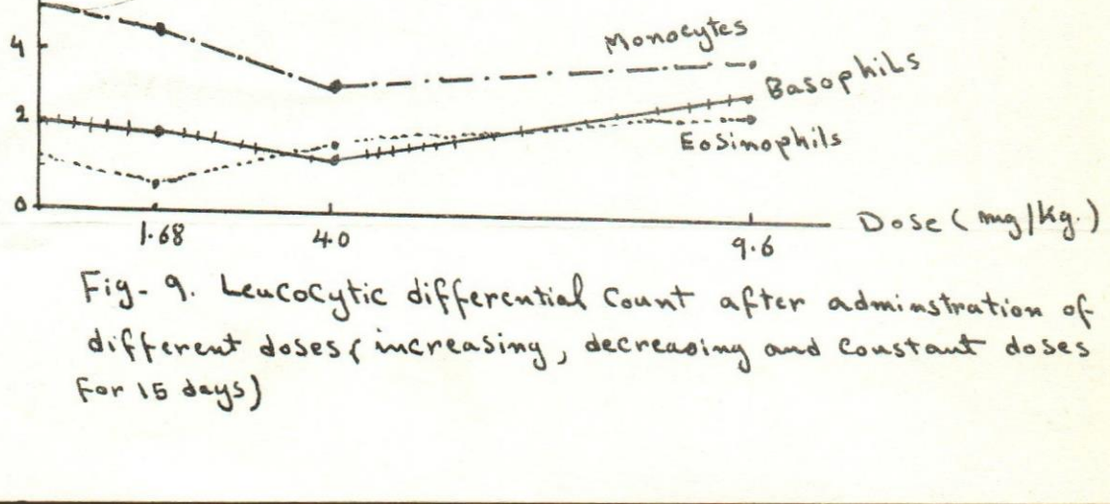

Fig- 9. Leucocytic differential Count after adminstration of different doses (increasing, decreasing and constant doses for 15 days)