

UNCLASSIFIED

AD NUMBER

ADC950071

CLASSIFICATION CHANGES

TO: unclassified

FROM: confidential

LIMITATION CHANGES

TO:

Approved for public release, distribution unlimited

FROM:

Distribution authorized to DoD and DoD contractors only; Foreign Government Information; SEP 1974. Other requests shall be referred to The British Embassy, 3100 Massachusetts Avenue, NW, Washington, DC 20008.

AUTHORITY

DSTL, WO 189/4985, 8 Oct 2009; DSTL, WO 189/4985, 8 Oct 2009

THIS PAGE IS UNCLASSIFIED

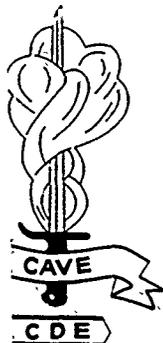
ADC950071

UNANNOUNCED

CONFIDENTIAL

CDE-TP-159

Copy No. 78 of 128 copies



AD No.
 DDC FILE COPY

1

CHANGES IN THE BEHAVIOURAL CHARACTERISTICS AND BRAIN ACETYLCHOLINESTERASE ACTIVITY OF RATS RECOVERING FROM GD POISONING



by

D.A. Buxton, J.J. Gordon and L. Leadbeater

Technical Paper No. 159

September 1974

Chemical Defence Establishment,
Porton Down, Salisbury, Wilts.

DDC
RECEIVED
MAR 27 1975
RECEIVED

CONFIDENTIAL

CONFIDENTIAL

ACCESSION for	
NTIS	White Section <input type="checkbox"/>
DDC	Buff-Section <input type="checkbox"/>
UNANNOUNCED	<input checked="" type="checkbox"/>
JUSTIFICATION.....	
BY.....	
DISTRIBUTION/AVAILABILITY CODES	
Dist.	AVAIL. and/or SPECIAL
/b	

THIS DOCUMENT IS THE PROPERTY OF HER
BRITANNIC MAJESTY'S GOVERNMENT, and is
issued for the information of such persons
only as need to know its contents in the
course of their official duties.

1. This information is released by the UK Government to the recipient Government for defence purposes only.
2. This information must be accorded the same degree of security protection as that accorded thereto by the UK Government.
3. This information may be disclosed only within the Defence Departments of the recipient Government and to its defence contractors within its own territory, except as otherwise authorised by the Ministry of Defence. Such recipients shall be required to accept the information on the same conditions as the recipient Government.
4. This information may be subject to privately-owned rights.

CONFIDENTIAL

(9) Technical papers

CDE TECHNICAL PAPER NO 159
DATE: SEPTEMBER 1974

7

CHANGES IN THE BEHAVIOURAL CHARACTERISTICS AND BRAIN ACETYLCHOLINESTERASE
ACTIVITY OF RATS RECOVERING FROM GD POISONING

[C]-8

by

(10)

D.A. BUXTON, J.J. GORDON

L. LEADBEATER

Sept 74 ✓

(12) 22 p. 1

SUMMARY

1. It has previously been noted that guinea pigs protected against many times the lethal dose of GD by pre-treatment with a carbamate, supported by atropine therapy, had virtually no measurable acetylcholinesterase activity in the brain although the animals appeared normal. The object of the present investigation was to establish whether animals surviving poisoning by GD were normal, mentally and physically. Since behavioural studies cannot be done with guinea pigs, a dose schedule was devised to protect rats (which are resistant to organophosphate poisoning and also resistant to therapy for that poisoning) against GD poisoning. The animals retained about 10% of the brain acetylcholinesterase activity 24 hours after poisoning.
2. Significant change in the behaviour of the animals was observed 48 and 96 hours after poisoning. The rats appeared to be fully recovered 14 days after poisoning.
3. The regeneration of brain acetylcholinesterase activity to within the normal limits occurred within 6 to 8 weeks. Plasma and erythrocyte acetylcholinesterase activity, which was depressed to a similar extent to brain 24 hours after poisoning recovered very much more rapidly.
4. The implications of these results are discussed.

elle

"A"

(Sgd) L. LEADBEATER
Superintendent
Biology Division

CHANGES IN THE BEHAVIOURAL CHARACTERISTICS AND BRAIN ACETYLCHOLINESTERASE
ACTIVITY OF RATS RECOVERING FROM GD POISONING

by

D.A. BUXTON, J.J. GORDON

and

L. LEADBEATER

INTRODUCTION

During studies of the protection of guinea pigs against GD poisoning by carbamate prophylactic treatment (1.2), it was observed that animals protected against as much as eight times the LD₅₀ of GD appeared to be quite normal 24 hours after poisoning. GD-inhibited acetylcholinesterase (AChE, EC 3.1.1.7) ages rapidly and cannot be reactivated by oximes (3.4). Thus it seemed possible that animals surviving poisoning by GD would have little or no acetylcholinesterase activity in their tissues. Preliminary experiments have confirmed that guinea pigs surviving poisoning by 2.3 x LD₅₀ of GD have less than 10% of the normal tissue AChE activity. In view of the central role ascribed to AChE in nervous transmission, it was of great importance to establish whether the animals surviving GD poisoning, with greatly reduced brain AChE levels, were in fact normal, physically and mentally.

Preliminary studies using conditioned avoidance and the spontaneous activity tests, as well as an attempt to devise a "startle response" test, confirmed that guinea pigs are not suitable subjects for behavioural studies. It was therefore necessary to establish a drug regimen which would protect rats against GD poisoning. Rats as a species are relatively unresponsive to nerve agent poisoning and to treatment against such poisoning, hence a treatment schedule was devised which gave a reasonably high proportion of survivors with not more than a slight degree of retention of brain AChE activity. Behavioural studies have been performed on rats protected against poisoning by lethal doses of GD and parallel studies of the AChE activity of brain and blood were also made.

METHODS

Animals

Male rats of initial body weight 180-220 g were used.

Materials

GD (at least 95% pure), pyridostigmine and P2S were synthesised within the Establishment. Atropine sulphate was obtained from BDH Limited. Solutions were made in isotonic saline (0.85% NaCl) and administered in a volume of 1 ml per kg.

Treatment of Rats

Rats were treated in groups of four. They were given P2S (15 mg per kg, i.m.) followed by pyridostigmine (0.05 mg per kg, i.m.) 10 minutes later. GD (200 µg per kg, approximately twice the LD₅₀, s.c.) was given 15 seconds later followed by intramuscular therapy with P2S (15 mg per kg) mixed with atropine sulphate (17.4 mg per kg) at the earliest signs of organophosphate poisoning. Additional doses of atropine sulphate (booster atropine) were administered (i.p.) when needed, according to the severity of the signs of poisoning. (The use of the i.m. route for booster atropine was not practicable because of the greater strength of the booster dose solutions. Attempts to use the subcutaneous route at this stage were unsuccessful, since tissue reaction was caused at the sites of injection. The intraperitoneal route was free from that disadvantage.) The first booster dose of atropine (139 mg per kg) was given as signs of poisoning first developed, shortly after the injection of P2S and atropine. If the signs of poisoning, after becoming transiently more marked, did not begin to moderate after 5 min, a second booster dose was given, otherwise a second dose was given in the event of a relapse. In some instances a third booster of atropine was necessary, but none of the rats used were given more than three booster doses in all. The proportion of rats surviving poisoning when given this treatment varied from group to group but over the whole investigation the survival rate was 62%.

Because of the large amounts of atropine administered to the GD treated rats (even where only one booster dose was found necessary) it was important to perform behavioural tests on rats treated with similar doses of atropine, in the absence of GD. Rats for these purposes were first given P2S and pyridostigmine, as in the full treatment, followed by P2S and atropine 3 min after the pyridostigmine, and were then given booster doses of atropine 1 min (70 mg per kg) 15 min (105 mg per kg) and 30 min (70 mg per kg) after the P2S - atropine injection.

Measurement of AChE activity

Rats were killed by exposure to fluothane. The brain was removed, washed in 0.85% NaCl to remove traces of blood, drained and weighed. It was then homogenized in isotonic saline using a glass-teflon homogenizer and the homogenate was dispersed into a total volume of 10 ml of saline.

Blood was drawn into a heparinized tube by cardiac puncture. It was centrifuged at 2000 x g for 5 min and the plasma removed and stored at 0-4°C. The erythrocytes were washed twice in isotonic saline and the packed cells were haemolysed by addition of 0.2 ml of 10% saponin. The haemolysate was made up to the original blood volume with 0.85% NaCl.

AChE activity was estimated at 37° by the Warburg manometric technique (5) in a medium containing NaHCO₃ (0.02M) under 5% CO₂/95% N₂. The amounts of tissue used per vessel were: 0.1 - 0.5 ml plasma, 0.5 - 1.0 ml erythrocyte haemolysate and 0.5 - 2.0 ml brain homogenate, the exact amount depending upon the extent of the inhibition anticipated. Acetylcholine chloride was added, after equilibration of the vessels, from the side arm in 0.2 ml solution to give a final concentration of 0.012 M (brain and erythrocyte) or 0.03 M (plasma). The total volume of solution and tissue preparation was adjusted to 3.0 ml with isotonic saline.

AChE activities were expressed as µmol acetylcholine hydrolysed per 30 minutes per mg (wet weight) brain or per ml of blood.

Behavioural Tests

In each test three groups of animals were used:

- A. untreated animals
- B. animals treated with P2S, pyridostigmine and atropine, and
- C. animals protected against GD poisoning.

The rats were tested at 48 hours, 96 hours and 14 days after poisoning.

The behavioural tests chosen were:

1. The open field test (6). This test was originally designed to measure emotional activity in rats but it gives several useful measures of behaviour. It has been used successfully to detect small behavioural changes produced by a variety of drugs acting on the central nervous system (7, 8).
2. Spontaneous locomotor activity. Changes in locomotor activity are often seen during depression or stimulation of the CNS. This test was only employed on the 48 hour group of rats. (Unfortunately the apparatus broke down and it

was not available for the 96 hour and 14 day experiments.)

3. Conditioned avoidance behaviour in a shuttle box. There is good evidence to suggest cholinergic involvement in conditioned behaviour (9, 10, 11) and depression or stimulation of the CNS via other mechanisms are often reflected as changes in conditioned behaviour.
4. Rotarod test. This is a useful means of quantifying physical incapacitation.

Each animal was examined in the tests in the order described.

Testing procedure

1. Open field. The apparatus was similar to that previously described (8). The animals were placed in the open field for a 3 minute period and the following items of behaviour were scored:

Preening, rearing, number of times defecating, and the number of floor squares traversed in the central area of the field.

2. Spontaneous locomotor activity. Activity recording cages were used which measured the movement of rats across a grid floor by detecting small changes in the electrical impedance caused by changing patterns of feet on the bars. Each animal was allowed 60 minutes in the activity box, the activities scored in 5 minute intervals were recorded and the mean activity plotted for each group.

3. Conditioned avoidance. The shuttle box apparatus used in this experiment has been described previously (8). Rats were trained to make the response of crossing from one side of the shuttle box to the other when presented with an audible signal (conditioned stimulus), in order to avoid an electric shock which followed it (unconditioned stimulus). A total of 80 training trials (i.e. 50 presentation of the signal followed by shock) in one 30 minute session were given to each rat. The total number of conditioned avoidance responses made during this session were computed and the number of occasions on which animals failed to cross the shuttle box even when shock was presented (termed failures) were also noted. The former gives a measure of learning ability and probably also of behavioural arousal, whilst a high failure score, gives a measure of behavioural depression or physical incapacitation.

4. Rotarod. The rotarod apparatus used consisted of a rubber covered rod of diameter 10 cm, rotating at a speed of 12 Hz. Each rat was placed on the rotating rod and replaced if it fell. Control animals rapidly learned to walk

in order to stay on and by plotting falls a minute for 10 minutes a learning curve was obtained. Ataxia and other forms of physical incapacitation produced changes in the shape and level of the learning curve.

RESULTS

Acetylcholinesterase Activity of the Tissues

Little AChE activity was present in the brains of animals 24 hours after GD poisoning (Table 1). The proportion of AChE activity remaining was variable, the mean being 13.9% of the control value. Similar results were found for erythrocyte AChE but appreciably greater amounts of plasma cholinesterase were retained.

The recovery of AChE activity in rats after poisoning is shown in Figures 1-3. The recovery of plasma cholinesterase was extremely rapid, rising to the normal control level in 3 days. The recovery of erythrocyte AChE activity was slightly less rapid. An interesting feature of the results for both erythrocytes and plasma is that activities continued to rise to temporarily much higher values than the normal levels. Over the eight week period of the experiment, the control values for both plasma and erythrocyte rose by 48-50%.

Brain AChE activity recovered at a very much slower rate than that of blood. It was 6 weeks before values came back to within the normal limits. In this case, the temporary rise to super normal values was not observed, the activity tending to level off after 6 weeks.

Behavioural Studies

1. Open Field Tests

The results of the open field test on normal control (A), atropine control (B) and GD treated animals are shown in Table 2. Severe depression of most types of behaviour was seen at 48 hours and to a slightly lesser extent at 96 hours after GD poisoning. The behavioural effects seen were typical of depression of the CNS and included reduced preening, rearing, ambulation and defecation. Fourteen days after poisoning there was an unexpected increase in the defecation score and a reduced ambulation. It is important to note here that there was also a significant difference in ambulation scores between the two control groups, apparently due to an unusually high score in the untreated control group (A).

2. Spontaneous locomotor activity

This parameter was only measured in animals tested 48 hours after poisoning. The two control groups did not differ from each other but the GD poisoned rats had

activity scores that were significantly lower (Student's t test, $P < 0.01$) from both the control scores over the period up to 30 min after introduction into the box.

3. Conditioned Avoidance

Large reductions in the conditioned avoidance response were seen in GD poisoned animals when tested at 48 or 96 hours after exposure (Table 3). Not only was the learning behaviour depressed but general behavioural depression or physical incapacitation was evident by the high failure scores in the GD groups at these times. However, after 14 days there was no significant difference between the GD treated and the control animals, although a suggestion of a remaining depression could be seen in their scores.

4. Rotarod

In Figure 4 and Table 4 and 5, three statistical comparisons are made. First, comparisons were made of the mean falls in the first minute of training and also of the total falls over the 10 min period. These give measures of physical incapacitation, the first one containing little or no learning component. Also a statistical comparison of the slopes was made to compare the learning ability under physical incapacitation, thus providing a measure of the degree of the effect. It may be possible, for example, for ataxic rats to learn to overcome their disability and produce a learning curve that is not significantly different from a control one, but which is comprised of a greater total number of falls than a control curve. Thus, the slope for the 48 hour group is not different from that of the controls but the mean of the first minute falls and of the total number of falls are significantly different from those of both the control groups. The 96 hour slopes are all similar but the total number of falls shown in the GD poisoned groups is significantly higher than in the control. In the 14 day group none of the comparisons showed significant differences between the groups.

AChE activities in Rats used for Behavioural Tests

Brain AChE estimations were performed on a representative sample of the rats immediately after they had been tested in the behavioural experiments for both control and GD treated animals (Table 6). The data for the AChE activity from GD poisoned rats were consistent with those found previously (Figure 1) although the brain AChE activity of the 96 hour survivors was markedly below that found previously. The results for the atropine controls were not significantly different from the normal controls at any of the time intervals employed.

DISCUSSION

There were significant changes in the behavioural pattern of rats 48 and 96 hours after poisoning by GD. These changes had largely disappeared 14 days after poisoning, the only significant differences seen between GD poisoned and control animals at that stage was in the open field test. There was a significant increase in defecation and a significant decrease in ambulation. As the direction of the change in the first parameter was opposite to that previously observed it is difficult to assess its importance but it is unlikely to be related to the treatment which the animals had received. The decrease in ambulation was only significant when compared with one of the two control groups in the experiment in which the ambulation score was unusually high. Physical incapacitation, measured by the rotarod technique, followed a similar time course to the behavioural changes, the animals being completely normal 14 days after poisoning.

It is tempting to speculate that the changes in the behavioural activity of the animals is related to the tissue levels of AChE. However the data presented here and by other workers (12) cannot support such a correlation. In the present work the AChE activity was measured on homogenates of whole brain. To attempt to correlate changes in tissue AChE activity with behaviour the experiments must be repeated measuring the AChE activity in discreet functional areas of a brain. It should be borne in mind that GD can inhibit a number of enzymes of importance in metabolism of the brain (13) and therefore the severe depression of the AChE activity may only be an index of the penetration of GD into the brain tissue.

The recovery of brain AChE activity after GD poisoning followed a similar time course to that reported previously for rats chronically exposed to GB vapour (14). In the present experiments the recovery of erythrocyte and plasma AChE activity was very much more rapid than was reported in the GB experiments. The temporary super normal values of blood AChE activity on recovery from nerve agent poisoning was greater after GD than was found after GB poisoning.

The rotarod experiments demonstrated that the animals poisoned by GD were ataxic for up to at least 4 days after poisoning but that they had recovered completely at 14 days. This result suggests that GD poisoning has no permanent effect, that it is not neurotoxic. However it must be borne in mind that the animals used in these experiments were only poisoned by about twice the lethal dose of GD whereas it is now possible to protect animals (and probably man) against up to 20 or more times the LD₅₀ of GD.

These experiments with rats recovering from poisoning by relatively low doses

of GD support the data reported for humans poisoned by low doses of organophosphates (15, 16, 17): namely, that recovery, both mental and physical, is complete after a period of about 14 to 21 days. The evidence of depression of the CNS in GD poisoning may have important implications for the management of casualties following a CW attack. This series of experiments was essentially of a preliminary character to point the way to a future programme to establish the behavioural effects of poisoning by a variety of nerve agents over a wide range of dose levels in animals protected by a variety of treatments, including carbamate prophylaxis supported by anti-acetylcholine drugs, oxime and anti-convulsant therapy. Further experiments are also required to establish whether the changes in the behavioural activity of the animals can be related to the depression of the brain AChE activity.

CONCLUSIONS

1. Significant behavioural changes were observed in rats 48 and 96 hours after poisoning by 200 µg GD per kg. The animals appeared to be fully recovered 14 days after poisoning.
2. Blood and brain AChE activities were depressed by about 90% in rats 24 hours after poisoning by GD (200 µg per kg). Regeneration of brain AChE activity to within the normal limits occurred in 6 to 8 weeks. Plasma and erythrocyte activities recovered much more rapidly.

ACKNOWLEDGEMENT

Mr. T. Grace did the AChE assays.

REFERENCES

1. GORDON, J.J. and LEADBEATER, L. (1972) CDE TP 53.
2. GORDON, J.J. and LEADBEATER, L. (1972) CDE TP 66.
3. FLEISCHER, J.H. and HARRIS, L.W. (1965) Biochem. Pharmacol. 14, 641.
4. MICHEL, H.O., HACKLEY, B.E., BERKOWITZ, L., LIST, G., HACKLEY, E.B., GILLILAN, W. and PANKAU, M. (1967) Arch. Biochem. Biophys. 121, 29.
5. AMMON, R. (1938) Arch. ges. Physiol. 233, 486.
6. HALL, C.S. (1934) J. Comp. Psychol. 18, 385.
7. BRIMBLECOMBE, R.W. (1963) Psychopharm. 4, 139.
8. BUXTON, D.A. (1972) Progress in Brain Research 36, 171.
9. RUSSELL, R.W. (1966) "Frontier in Physiol. Psychol." p. 141. Academic Press, New York.
10. CARLTON, P.L. (1963) Psychol. Reviews, 70, 19.
11. LONGO, V.G. (1966) Pharmacol. Reviews. 18, 966.
12. RUSSEL, R.W., WATSON, R.H. and FRANKENHAUSEN, M. (1961). Scand. J. Psychol. 2, 21.
13. JOVIC, R., BACHELARD, H.S., CLARK, A.G. and NICHOLAS, P.C. (1971) Biochem. Pharmacol. 20, 519.
14. OBERST, F.W. and JHRISTENSEN, M.K. (1956). J. Pharm. Exp. Therap. 116, 216.
15. GROB, D. (1956) A.M.A. Archives of Internal Med. 98, 221.
16. FRIBORSKA-WAELSCHOVA, A. (1957) Archiv. Für Gewerbepathol. u. Gewerbehygiene 16. 63 (Porton Translation 230).
17. DURHAM, W.F., WOLFE, H.R. and QUINBY, G.E. (1965) Arch. Env. Health 10, 55.

TABLE 1

AChE activities of tissues from surviving rats, 24 hrs
after GD poisoning, compared with normal rats

Tissue		No. of rats used	AChE activities μMole ACh/30 min/mg or ml		% control
			Range	Mean, with standard deviation	
Brain	Control	9	4.30-5.60	5.02 ±0.44	13.9
	Poisoned	20	0.34-1.75	0.70 ±0.27	
Erythrocytes	Control	10	139-264	179 ±36.9	13.5
	Poisoned	4	12.0-44.0	24.2 ±12.6	
Plasma	Control	10	135-252	201 ±34.4	32.4
	Poisoned	4	43.0-105	65 ±19.3	

TABLE 2

Behavioural tests on rats recovering from GD poisoning:

The Open Field Test

Time after poisoning	48h	96h	14 days
Control (A) X Control (B)	NS	NS	SQ↑
GD Group X Control (A)	P↑SQ↓	R+SQ↓	D↑SQ↓
GD Group X Control (B)	P+D↑SQ↓	NS	NS

NS No significant differences

↑ Significant increase

↓ Significant decrease

P Preening SQ Number of squares traversed

R Rearing D Times defecating

Control (A) Untreated animals

Control (B) Rats treated with pyridostigmine, P2S and atropine

GD Group Animals protected against GD poisoning by pyridostigmine, P2S and atropine treatment.

TABLE 3

Behavioural tests on rats recovering from GD poisoning: the

Conditioned Avoidance Response

<u>Time After Poisoning</u>	<u>Animals</u>	<u>Avoidance Response</u>	<u>Significance</u>	<u>"Failures" (to respond to shock)</u>	<u>Significance</u>
48 hr	Control (A)	14.0 ± 11.4	<p>p < 0.02 p < 0.01</p>	7.8 ± 8.9	<p>p < 0.001 p < 0.001</p>
	Control (B)	15.5 ± 9.9		2.3 ± 2.7	
	GD Group	4.2 ± 5.9		27.3 ± 13.5	
96 hr	Control (A)	16.8 ± 14.2	<p>p < 0.02 p < 0.05</p>	5.0 ± 5.7	<p>p < 0.001 p < 0.001</p>
	Control (B)	10.1 ± 8.9		7.8 ± 5.4	
	GD Group	2.5 ± 0.5		35.8 ± 10.4	
14 days	Control (A)	18.9 ± 11.1		2.1 ± 2.6	
	Control (B)	15.2 ± 6.9		8.0 ± 8.0	
	GD Group	9.0 ± 8.8		10.8 ± 12.2	

Scores are expressed as Means ± St. Deviation and were compared for Significance by Student's t-test. Unmarked comparisons are not significantly different (p > 0.05).

Control (A) Untreated animals
 Control (B) Rats treated with pyridostigmine, P2S and atropine
 GD Group Animals protected against GD poisoning by pyridostigmine, P2S and atropine.

TABLE 4

Behavioural tests on rats recovering from GD poisoning: rotarod test, physical capacity

Time after poisoning	Animals	Total Falls			Falls in First Minute		
		No. of animals	Falls	Significance	Nc. of animals	Falls	Significance
48 hr	Control (A)	10	14.0 ± 2.3	p<0.01	10	3.9 ± 0.5	p<0.005
	Control (B)	10	15.9 ± 4.4				
	GD Group	6	39.0 ± 7.9				
96 hr	Control (A)	8	13.5 ± 3.0	p<0.005	8	2.6 ± 0.4	
	Control (B)	8	22.0 ± 5.5				
	GD Group	9	34.6 ± 5.4				
14 days	Control (A)	8	13.6 ± 4.6		8	3.1 ± 1.0	
	Control (B)	9	21.4 ± 3.3				
	GD Group	7	21.6 ± 6.2				

Scores are expressed as Means ± St. Deviation and were compared by Student's t-test. Unmarked comparisons are not significantly different (p>0.05)

Control (A) Untreated animals
 Control (B) Rats treated with pyridostigmine, P2S and atropine
 GD Group Animals protected against GD poisoning by pyridostigmine, P2S and atropine

CONFIDENTIAL

TABLE 5

Behavioural tests on rats recovering from GD poisoning: rotarod test, a comparison of the learning curves

Time after poisoning	Animals	No. of animals	Gradient (\pm S.D)
48 hr	Control (A)	10	3.64 \pm 2.19
	Control (B)	10	3.96 \pm 2.42
	GD Group	10	5.12 \pm 4.76
96 hr	Control (A)	8	2.34 \pm 1.71
	Control (B)	8	3.15 \pm 2.55
	GD Group	9	2.10 \pm 2.70
14 days	Control (A)	8	3.29 \pm 2.06
	Control (B)	9	4.68 \pm 3.04
	GD Group	7	4.17 \pm 2.80

There was no significant difference between the gradients of the learning curves of any of the 3 groups at the 3 times they were measured.

Control (A) Untreated animals
Control (B) Rats treated with pyridostigmine, P2S and atropine
GD Group Animals protected against GD by pyridostigmine, P2S and atropine treatment

CONFIDENTIAL

TABLE 6

Brain AChE activity in rats immediately after
the behavioural tests

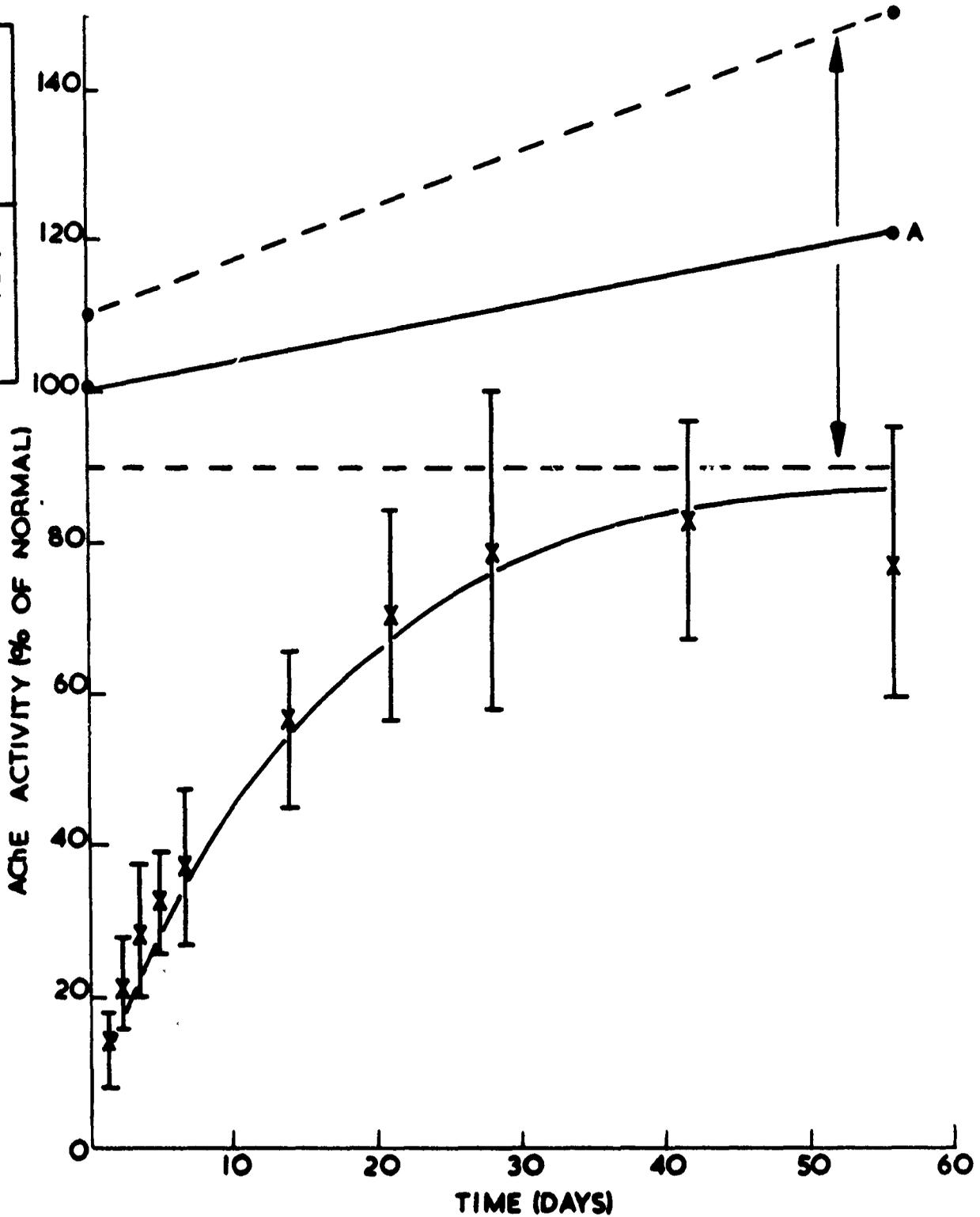
Animals	Time After Treatment	No. of animals	AChE Activity (μ Moles ACh/30 min/mg)		
			Range	Mean \pm S.D	% Control
Control (A)		6	6.37-7.89	7.01 \pm 0.75	100
Control (B)		6	6.04-7.47	6.65 \pm 0.55	95.1
GD Group	48 hr	6	2.06-2.88	2.37 \pm 0.37	33.5
GD Group	96 hr	4	0.60-0.90	0.72 \pm 0.05	10.3
GD Group	14 days	5	1.84-3.97	2.74 \pm 0.90	39.4

Control (A) Untreated animals

Control (B) Rats treated with pyridostigmine, P2S and atropine

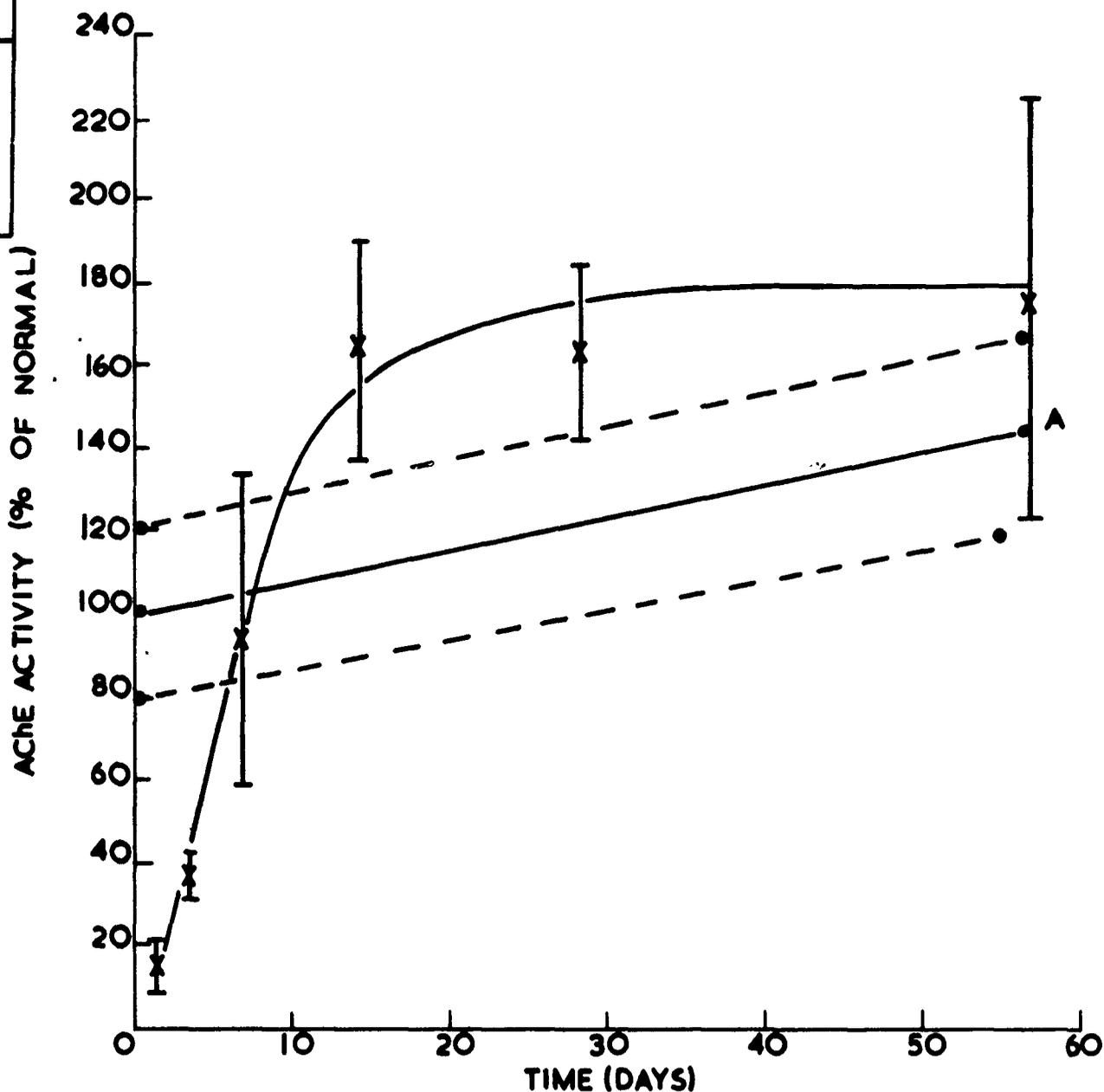
GD Group Animals protected against GD poisoning by pyridostigmine
P2S and atropine treatment

TP 159 PT 6687



RECOVERY OF BRAIN AChE IN RATS PROTECTED AGAINST 2LD₅₀GD. A, NORMAL VALUES, ESTIMATED AT 0 AND AT 56 DAYS ONLY.

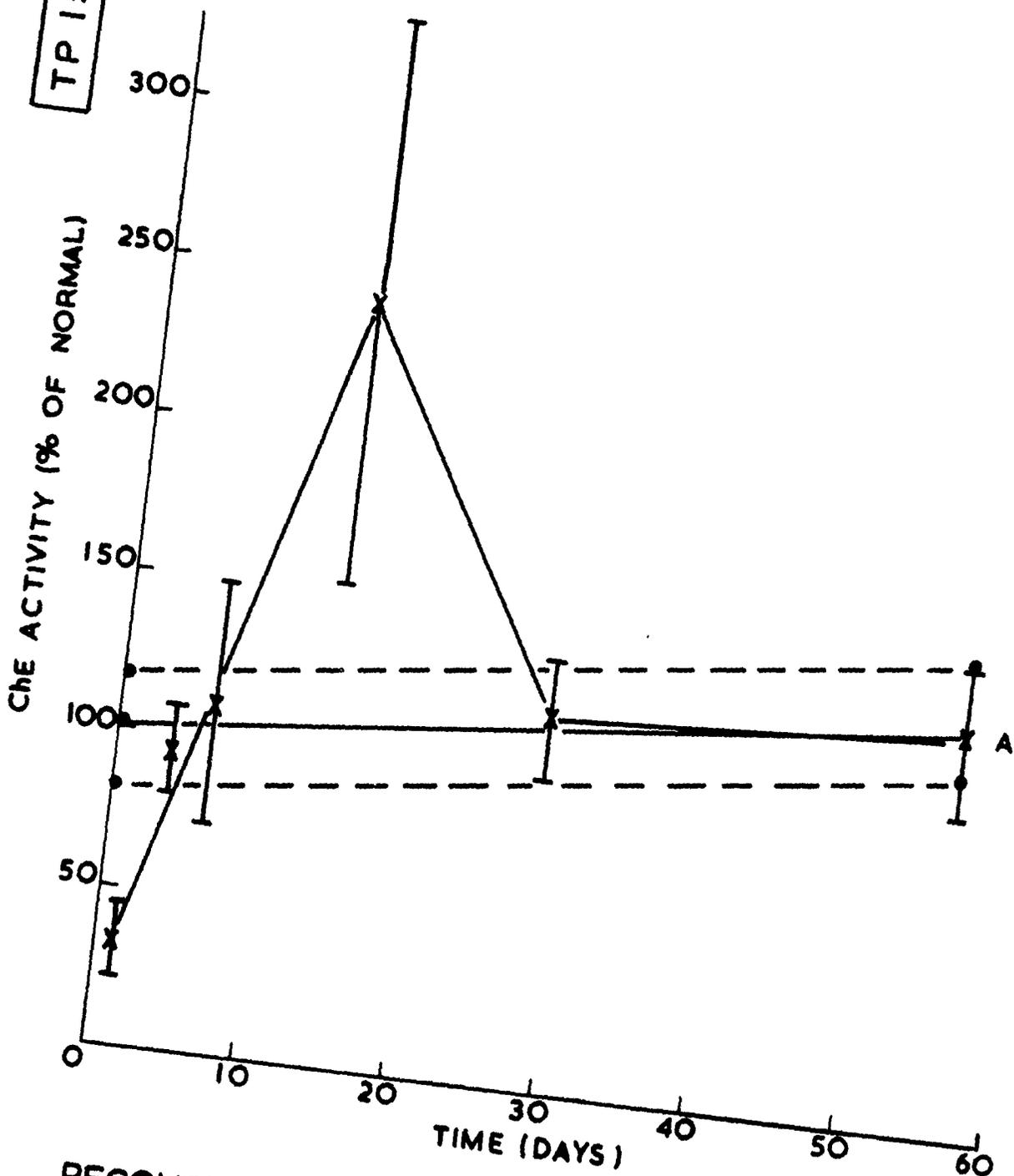
TP 159 PT 6688



RECOVERY OF ERYTHROCYTE AChE IN RATS
PROTECTED AGAINST 2LD₅₀ GD. A, NORMAL VALUES,
ESTIMATED AT 0 AND AT 56 DAYS ONLY

CONFIDENTIAL

TP 159 PT 6689



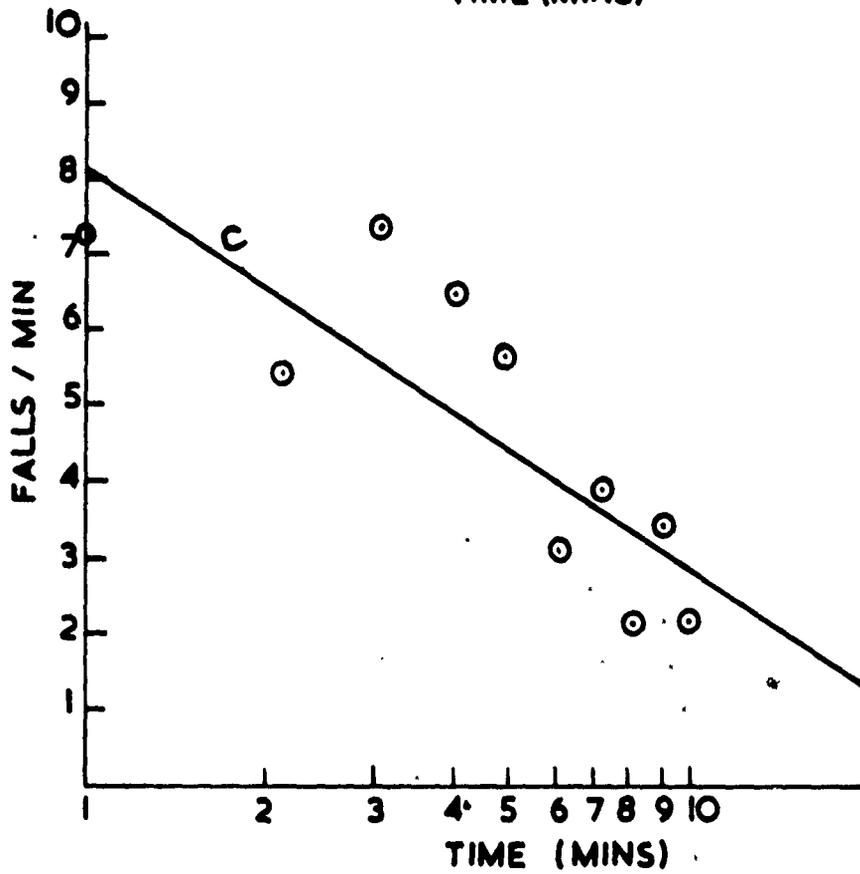
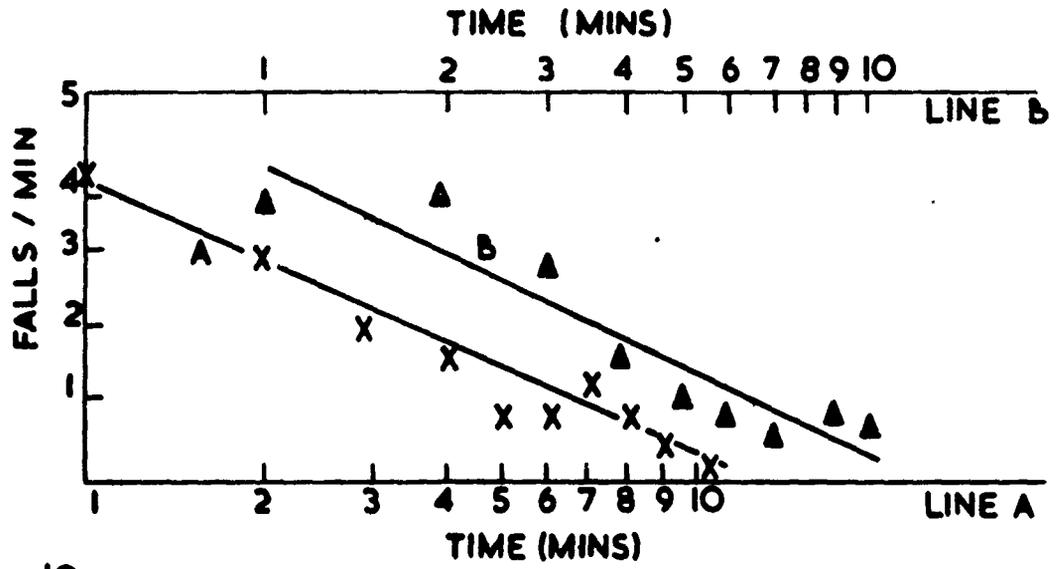
RECOVERY OF PLASMA ChE IN RATS PROTECTED
AGAINST 2LD₅₀ GD. A, NORMAL VALUES, ESTIMATED
AT 0 AND AT 56 DAYS ONLY.

CONFIDENTIAL

FIG. 3.

CONFIDENTIAL

TP 159 PT 6690



ROTARD TESTS. A, NORMAL CONTROLS
B, ATROPINE CONTROLS,
C, GD-TREATED RATS (AT 48 HRS)

CONFIDENTIAL

FIG. 4.

CDE TECHNICAL PAPER 159

DISTRIBUTION LIST

DCS(A)	<u>Australia cont</u>
AD RPB	Royal Australian Air Force
DR/CB (2 copies)	
PM Wpns 8	<u>Canada</u>
	Chairman, Defence Research Board
GS(OR) 4	Defence Research Establishment, Ottawa
AMD5	(2 copies)
Cmdt, Defence NBC School	Defence Research Establishment,
RMCS, Shrivenham	Suffield
DCS(N)	<u>USA</u>
MDG(N)	S.O.(C) to CBNS Washington (2 copies)
	US Naval Tech Liaison Officer
DHR(RAI')	
	<u>Internal</u>
Members of Biology Cttee (18 copies)	CD
Members of Medical Cttee (25 copies)	ERD (2)
	Med D (5)
MPE	Prot D
	TCD
Home Office SAB (3 copies)	PD
(1 copy attn Mr R Cotterill)	AS
	Proc R D (2)
DRIC (2 copies)	SMO
	NEO
<u>Overseas</u>	TIRS (9)
USASR (Porton) (18 copies, 2 for FCST)	Tech Reg (3)
<u>Through DRIC</u>	
<u>Australia</u>	
Defence Standards Laboratories (3 copies)	
Senior Representative, Dept of Supply	
Australian Army Staff, London	



Department for
Foreign Affairs
[dstl] - International
Security
Policy
SIPRI
2004-2018
www.dstl.gov.uk
www.sipri.org

Defense Technical Information Center (DTIC)
8725 John J. Kingman Road, Suit 0944
Fort Belvoir, VA 22060-6218
U.S.A.

AD#: ADC950071

Date of Search: 8 Oct 2009

Record SummaryWO 189/4985

Title: Changes in Behavioural Characteristics and Brain Acetylcholinesterase Activity of Rats Recovering from GD (SOMAN) Poisoning
Availability Open Document, Open Description, Normal Closure before FOI Act: 30 years
Former reference (Department): CDE TP 159
Held by: The National Archives, Kew

This document is now available at the National Archives, Kew, Surrey, United Kingdom.

DTIC has checked the National Archives Catalogue website (<http://www.nationalarchives.gov.uk>) and found the document is available and releasable to the public.

Access to UK public records is governed by statute, namely the Public Records Act, 1958, and the Public Records Act, 1967.

The document has been released under the 30 year rule.

(The vast majority of records selected for permanent preservation are made available to the public when they are 30 years old. This is commonly referred to as the 30 year rule and was established by the Public Records Act of 1967).

This document may be treated as UNLIMITED.