Sarin exposures in a cohort of British military participants in human experimental research at Porton Down 1945-1987

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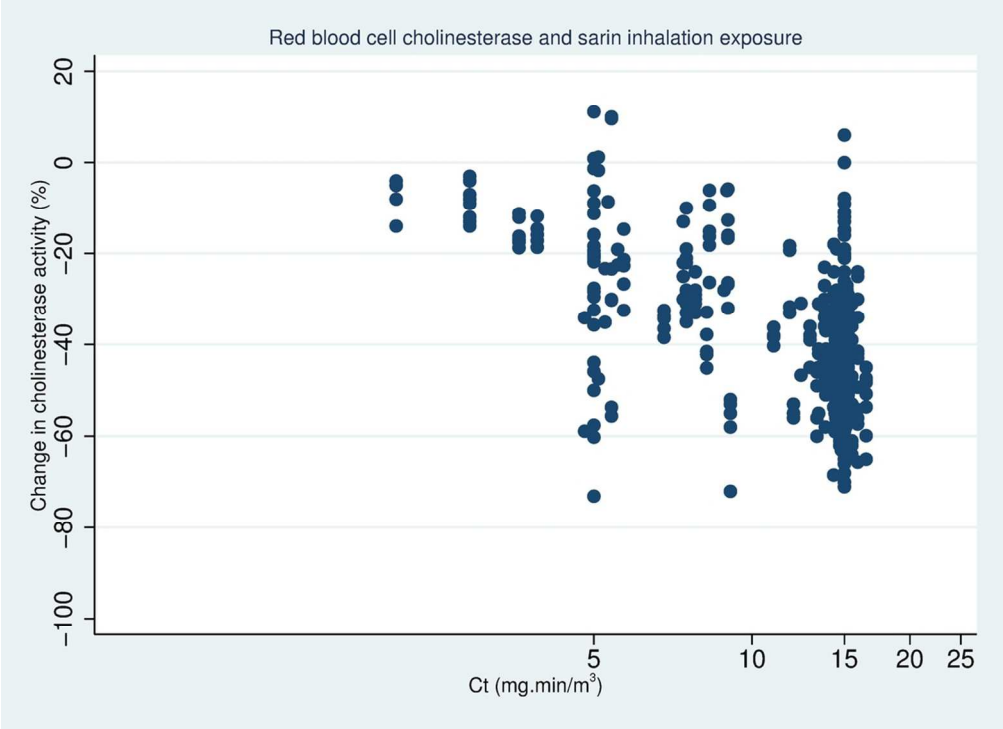

Figure 1 Scatter plots of sarin inhalation exposure and percentage change in activity of a) red blood cell, b) plasma, c) whole blood, and d) unspecified cholinesterase. e) Sarin dermal exposure and percentage change in activity of unspecified cholinesterase.

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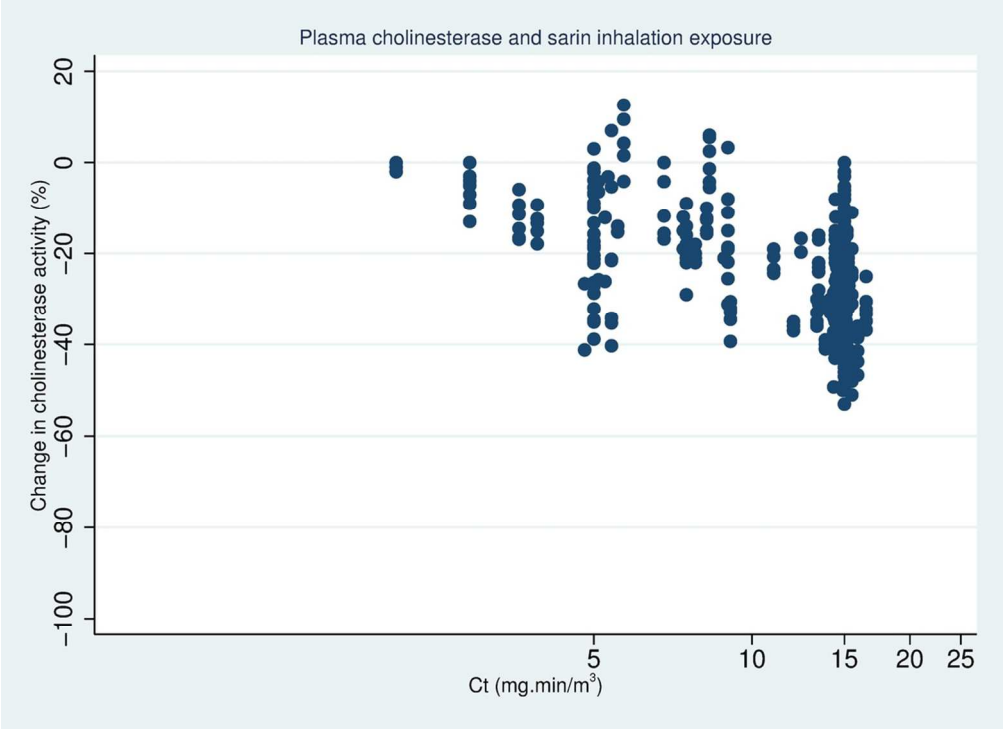

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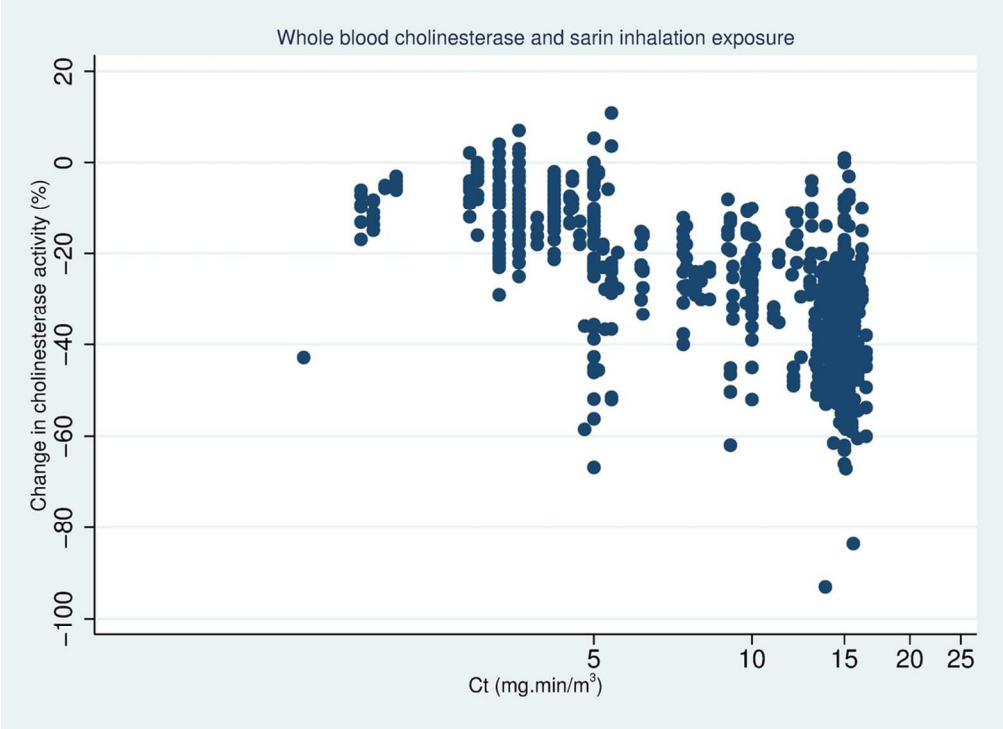

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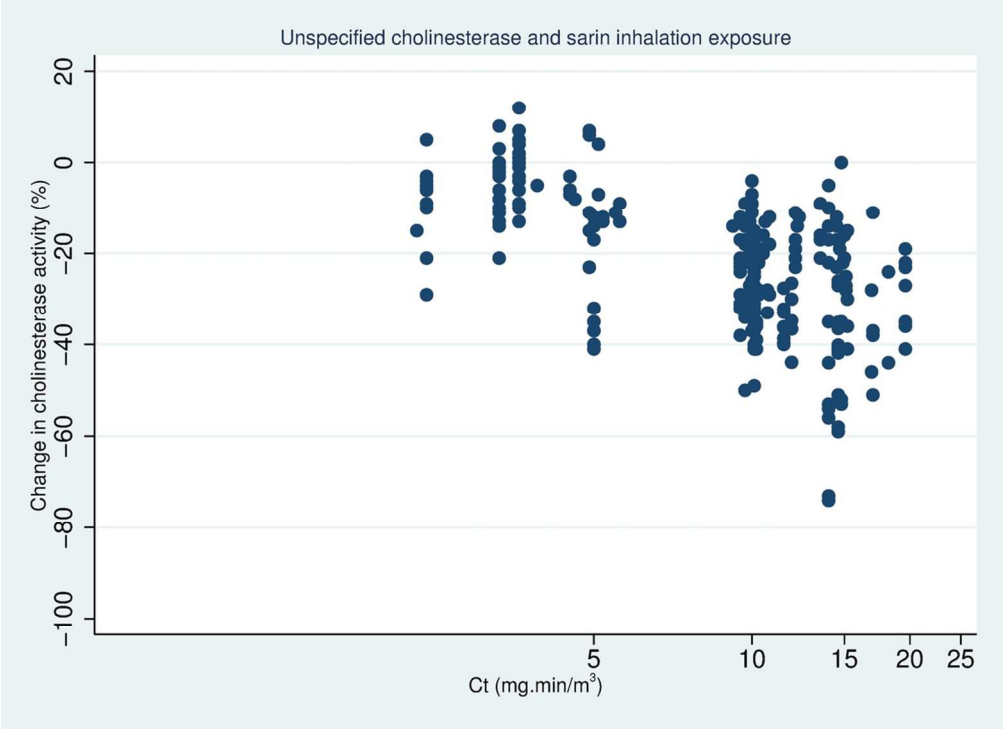

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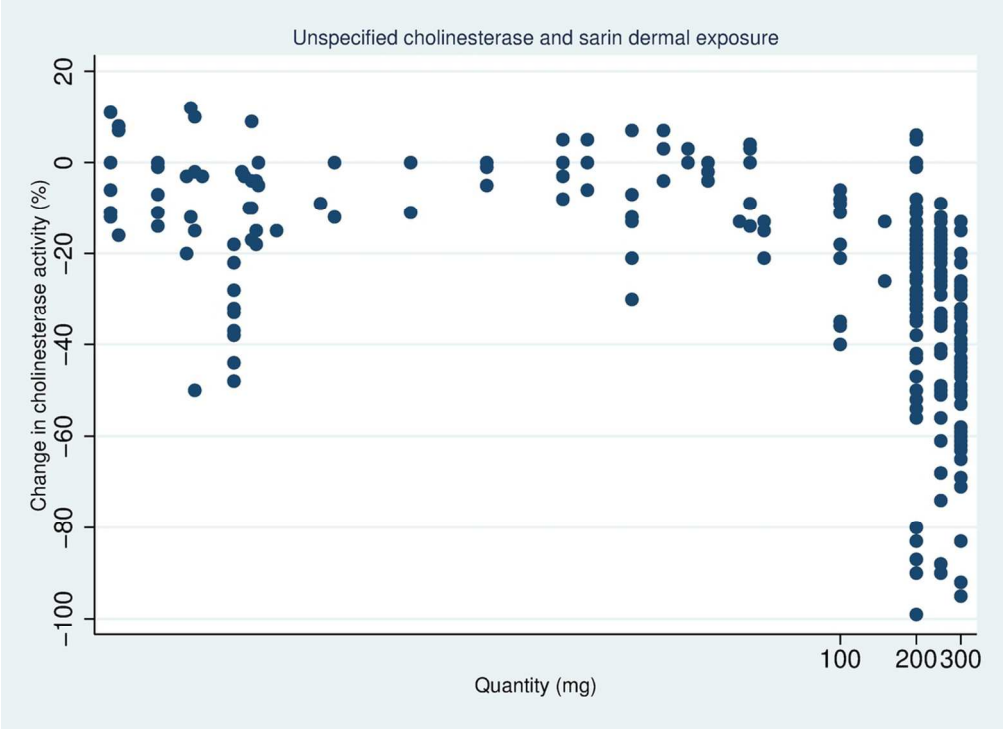

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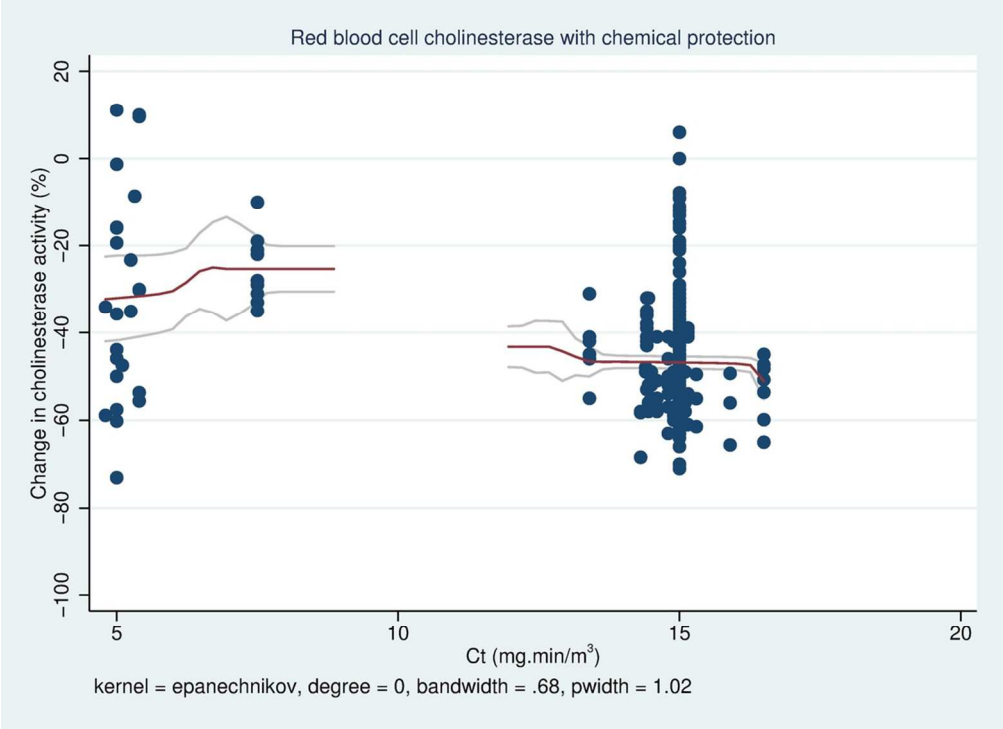

Figure 2 Non-parametric kernel regression estimate and 95% CIs for sarin inhalation exposure and percentage change in red blood cell cholinesterase activity, with a) chemical protection (n = 326) and b) no protection (n = 332).

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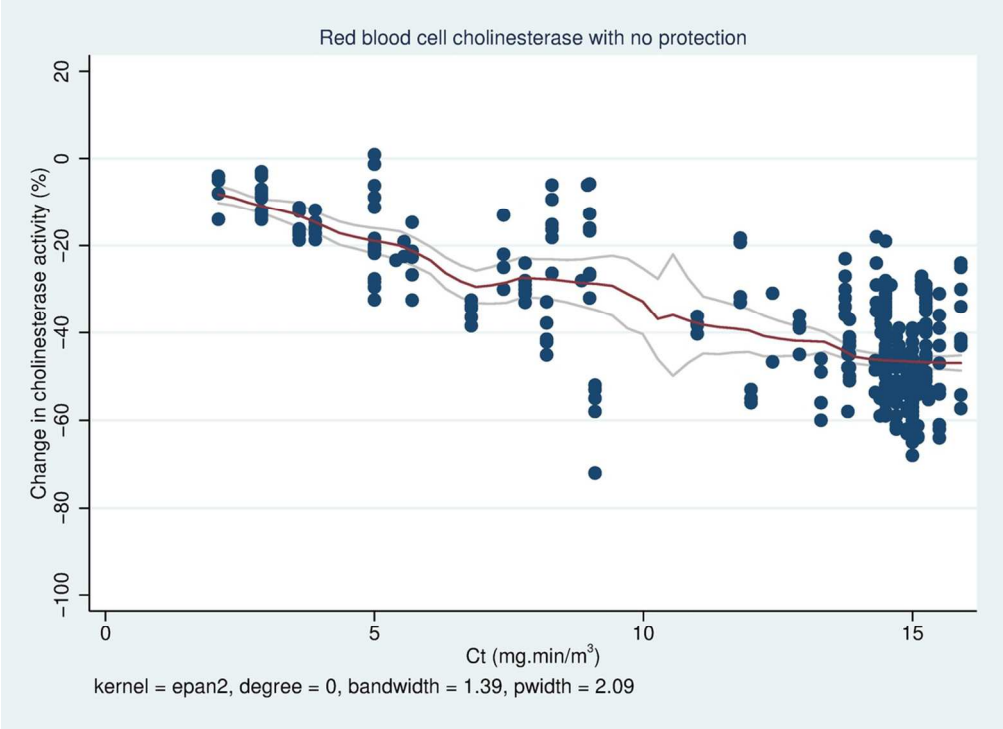

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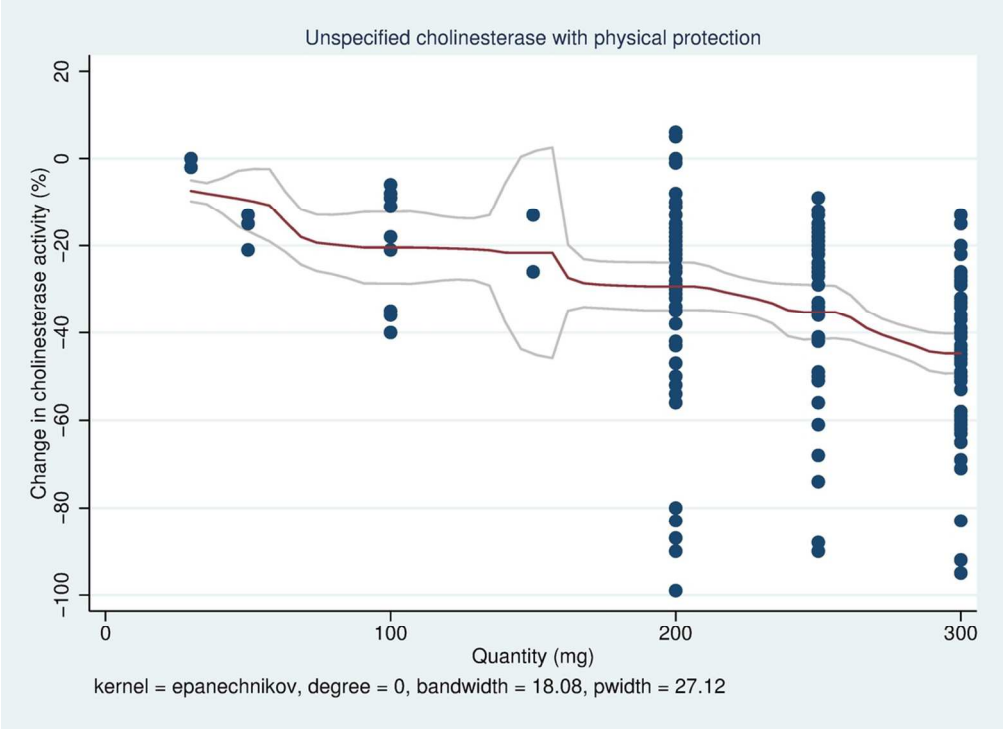

Figure 3 Non-parametric kernel regression estimate and 95% CIs for sarin dermal exposure and percentage change in unspecified cholinesterase activity, with a) physical protection (n = 185), and b) no protection (n = 88).

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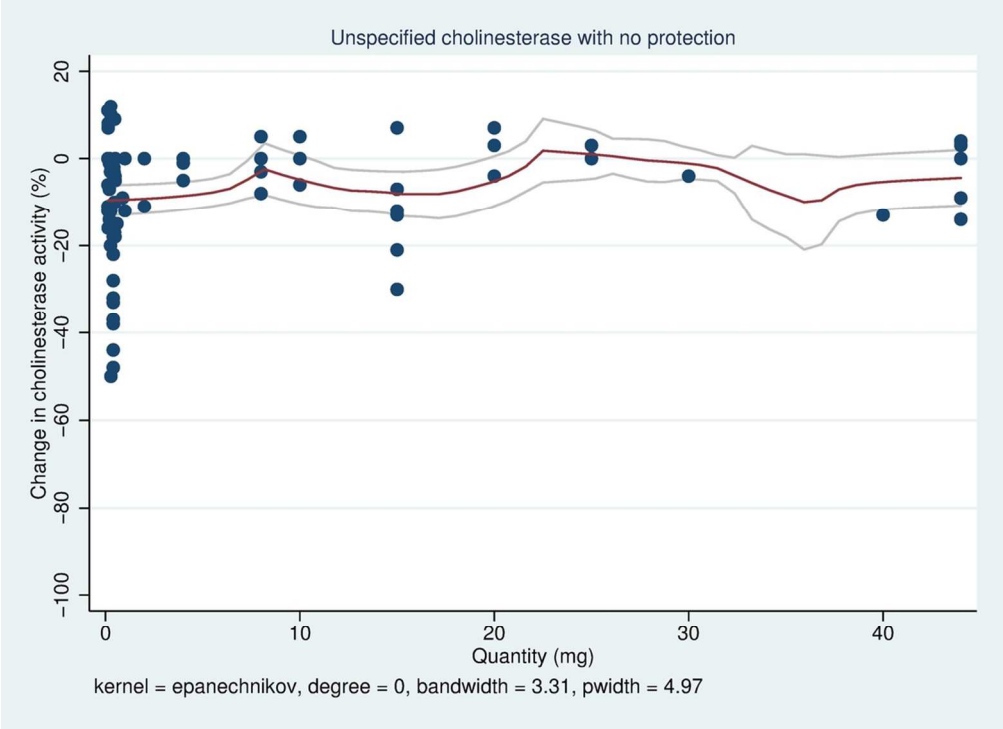

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Sarin exposures in a cohort of British military participants in human experimental research at Porton Down 1945-1987

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Running title: Human sarin exposures at Porton Down

Key words: Chemical weapons, nerve agents, sarin, epidemiology, exposure, exposure-response

Abstract

Background

The effects of exposure to chemical warfare agents in humans are topical. Porton Down is the UK's centre for research on chemical warfare where, since WWI, a programme of experiments involving approximately 30,000 participants drawn from the UK armed services has been undertaken.

Objectives

Our aim is to report on exposures to nerve agents, particularly sarin, using detailed exposure data not explored in a previous analysis.

Methods

In this paper we have used existing data on exposures to servicemen who attended the human volunteer programme at Porton Down to examine exposures to nerve agents in general and to sarin in particular.

Results

Six principal nerve agents were tested on humans between 1945 and 1987. Of all 4,299 nerve agent tests recorded, 3,511 (82%) were with sarin, most commonly in an exposure chamber, with inhalation being the commonest exposure route (85%). Biological response to sarin exposure was expressed as percentage change in cholinesterase activity and, less commonly, change in pupil size. For red blood cell cholinesterase, median inhibition for inhalation tests was 41% (IQR 28-51%), with a maximum of 87%. For dermal exposures the maximum inhibition recorded was 99%. There was a clear association between increasing exposure to sarin and depression of cholinesterase activity but the strength and direction of the association varied by exposure route and the presence of chemical or physical protection.

Pupil size decreased with increased exposure but this relationship was less clear when modifiers, such as atropine drops, were present.

Conclusions

These results, drawn from high quality experimental data, offer a unique insight into the effects of these chemical agents on humans.

Introduction

Nerve agents are organophosphate chemicals designed for use as chemical weapons. They act by the irreversible inhibition of acetyl cholinesterase in synapses and neuromuscular junctions, resulting in a build-up of toxic levels of acetylcholine. The effects of nerve agents, and other organophosphate compounds, can be counteracted by atropine and pralidoxime chloride. Atropine antagonises the muscarinic effects and some central nervous system effects of nerve agent exposure. Pralidoxime and other oximes act by reversing acetylcholinesterase phosphorylation. These effects are seen in organs with nicotinic receptors, such as skeletal muscle. Before exposure, pyridostigmine, which reversibly binds to acetyl cholinesterase, may be administered as a pre-treatment (Sidell and Borak 1992).

Nerve agents were first developed by German chemists in the 1930s. Their discovery was the result of research into possible new pesticides (Coleman 2005). They were weaponised by German forces in WWII but were not used in combat during the war. Since then, nerve agents have been produced in considerable quantities by some countries, but rarely used; they were used in attacks against Iranian troops by Iraq (1980-88) and have been used against civilians, most notably against Kurdish civilians in Iraq at Halabja in 1988, on the Tokyo underground in 1995, and more recently in Syria (Organisation for the Prohibition of Chemical Weapons 2017, 2017). Although the use and stockpiling of all chemical weapons is prohibited by the Chemical Weapons Convention, the possibility that they might be used is a recurrent concern (Organisation for the Prohibition of Chemical Weapons 1997).

Porton Down is the UK's centre for research on chemical warfare. Since WWI it has undertaken a programme of experiments using participants drawn from the UK armed services. Since 1916, approximately 30,000 service personnel have taken part. Before WWII, Porton Down scientists had determined that organophosphates were a class of chemicals from

which a chemical weapon might be derived but the focus of their research, iso-propyl fluorophosphate (PF-3), was not considered toxic enough to be a useful weapon (Ministry of Defence 2006). The first nerve agent tested at Porton Down was from a captured German shell in 1945 (Hinsley and Howard 1990) and after April 1945 Porton Down included nerve agents in the human testing programme. Over 4,000 tests with six nerve agents were recorded on over 3,000 participants between 1945 and 1987 (Keegan et al. 2009). Most tests involved sarin (GB) and these took place during the 1950s (Keegan et al. 2009). Three other nerve agents (GA, GD and GE) were tested only in the 1940s and 1950s while two (GF and VX) were not extensively tested on humans and were not tested after the end of the 1960s.

In our previous paper we reported on exposures to all chemicals in the 'human volunteer programme' (Keegan et al. 2009). In that study, we documented every test carried out as part of the programme at Porton Down 1941-1989, grouped documented chemicals into broad categories based on intended use (vesicant, nerve agent, lachrymator etc.), and assessed the number of people exposed to each chemical. We included servicemen in our analyses of mortality and cancer incidence, excluding a small number of servicewomen and civilians (Carpenter et al. 2009; Venables et al. 2009). In the current paper, we concentrate on exposures to nerve agents, and in particular sarin, for which we collected additional detailed exposure data not explored in the previous analysis.

In this paper we report on all sarin exposures recorded in cohort members at Porton Down for the complete period of nerve agent testing, when the sarin tests took place and on the intensity and duration of exposure. Sarin exposures account for over 80% of nerve agent exposure to humans at Porton Down. Additionally, we describe the acute biological response to nerve agent exposure with and without the presence of exposure modifiers (e.g. pharmacological pre-treatments) or physical barriers (e.g. protective clothing). However, the paper focuses on the experience of volunteers in nerve agent tests at Porton Down rather than

on refining human nerve agent exposures, and an important feature of the paper is that little of the material reported here has previously appeared in the public domain.

Methods

Data abstraction

Briefly, the UK Ministry of Defence made available a collection of archival material of contemporaneous records of experiments which were part of the programme at Porton Down between 1941 and 1989. Details of the sources of exposure data are documented elsewhere (Keegan et al. 2007; Keegan et al. 2009).

For all chemical tests, the research team recorded which chemical or chemical mixture was the focus of the test (ignoring other chemicals such as diluents or skin cleansers), exposure state (e.g. solid or liquid) and route of exposure (e.g. dermal, inhalation). For nerve agents, more detailed exposure information was collected. Details of data abstraction procedures have been published previously (Keegan et al. 2009).

Exposure measures

Exposure route was coded, by the researchers, as dermal only, ocular only, or not recorded. For any nerve agent test in which a person was exposed to a nerve agent gas/vapour in a chamber while wearing no protective equipment, the exposure route was coded as inhalation (± other routes), since it was assumed that in these circumstances the principal exposure route would be via the respiratory tract.

Most nerve agent exposures took place in an exposure chamber, and chamber exposure was documented contemporaneously as the product of the concentration of sarin in the chamber air and the duration of exposure (Ct) expressed in mg.min/m^3. Although Ct is a cumulative exposure parameter we have termed it exposure 'intensity' because exposures were usually of

short duration. Where Ct was available from both the experiment record and the separate chamber record, the chamber value was used. If necessary, exposure as Ct was calculated from concentration (as mg/m^3) and duration (as minutes) from recorded data. From 661 inhalation tests for which a Ct was not documented, we were calculate 47 further Cts.

Dermal exposures were applied in drops, usually to the skin of the inside forearm, and were mostly quantified as a known weight of nerve agent in mg. These dermal tests also took place in a chamber.

Chemical classification

The methods used to identify and categorise chemicals have been published (Keegan et al. 2009). Briefly, individual chemical names documented in the Porton Down experiment books initially identified as possible nerve agents by the research team were confirmed (or not) by Porton Down scientists with relevant expertise. Chemical names which were synonyms or structural analogues were grouped together under a project agent name. For example: the nerve agent soman (CAS registry number: 96-64-0) documented as 'soman', was assigned as the project agent name. Synonyms, such as GD, pinacolylmethylphosphonyl fluoride or 1,2,2trimethylpropofluoro(methyl)phosphine oxide, were coded as soman, as were functional analogues, such as tridecylmethylphospono fluoridate. In this paper nerve agents are reported using their project agent name (see table 1).

Protection status

For antidotes or pre-treatments, only the presence or absence of the chemical was recorded. Data relating to whether a test exposure was modified by physical protection were combined in this study as: yes to respirator use, non-protective or protective clothing or equipment. For respirator use, the categories 'no' and 'not known' were combined. No information was available on the type of respirator used.

Data relating to whether a test exposure was modified by chemical protection were combined. Typical chemicals documented as having been used in tests with sarin were atropine and pralidoxime.

Cholinesterase

Porton Down assayed three types of cholinesterase over time: unspecified, whole blood, red blood cell and plasma. We have taken red blood cell cholinesterase as the primary measure, where available. As units of cholinesterase measurement were not consistently applied over the study period, the percentage change in cholinesterase activity was used as the measure of acute biological effect. Whenever possible this was calculated from the pre- and post-exposure values. Percentage change was abstracted when this was the only measure recorded. Where more than one post-exposure value was documented, the lowest was used.

The percentage change in pupil diameter was used to assess the acute effect of nerve agent exposure. Documented measures (in mm) were for both right and left eye, for right or left eye alone, or for a single (unspecified) eye. The primary measure used here was change in right eye pupil size. If not available, change in left eye pupil size was used and, if that was not available, single (unspecified) eye. Correlation between left and right eye measurements was good (Pearson's coefficient 0.89). Our data indicate that the resolution of pupil diameter measurement was approximately 0.25mm.

Statistical analysis

Statistical analysis of data was performed using STATA v11.0 (StataCorp 2009). As distributions of many of the exposure parameters were positively skewed, these were primarily described using a median and interquartile range (IQR). Maximum exposure intensities and durations are also reported. Non-parametric kernel regression was used to model the association between exposure as the predictor variable and acute biological

response (the outcome variable). The kernel regression was carried out using the *lpoly* command in STATA, on untransformed data using the default Epanechnikov kernel function and 0 degrees of smoothing). These regressions give a non-parametric smoothed regression estimate which does not make any assumptions about the functional form of the relationship (Royston et al. 1999).

Ethical approval

Ethical approval for the epidemiological studies of Porton Down veterans was granted by the South East Multicentre Research Ethics Committee, the Defence Medical Services Clinical Research Committee, and the Patient Information Advisory Group. The Ministry of Defence provided access to data and was represented on the Medical Research Council's project monitoring group but had no role in the abstraction of data, analysis, or interpretation of the results.

Results

Chemicals tested and dates of tests

Human tests with nerve agents at Porton Down took place between April 1945 and December 1987 (Table 1). During that period six nerve agents were tested in a total of 4,299 tests, on 3,597 individuals. Most (75%) tests with nerve agents were completed by 1959. The most commonly-tested nerve agent was sarin, which accounted for 3,511 (82%) of the 4,299 nerve agent tests. The second most-tested nerve agent was tabun (GA), tested 398 times (9% of tests) on 262 individuals. Other nerve agents tested were soman (GD), cyclo-sarin (GF) and VX.

Sarin exposure route and protection use

Of all nerve agent tests carried out at Porton Down (n = 4,299), 80% (n = 3,511) were with Sarin. Most sarin tests (2,860 of 3,511, 82%) took place in an experimental chamber. Inhalation was the most common exposure route (2,972, 85% of all sarin tests, Table 2). Only 288 (8%) of tests were dermal. A small number of sarin tests were directly to the eye (110, 3%).

Forty-seven percent of sarin tests took place with some kind of physical or chemical protection (Table 2). In 665 (19%) sarin tests the participant was wearing a respirator, and this figure was slightly lower in tests by the inhalation route (470, 16%). Information on the type of respirator used was not available in this dataset. Physical protection (such as a patch of fabric under test) was present in 897 (25%) of sarin tests, and chemical protection (such as treatment with pralidoxime) in 577 (16%).. In 53% of sarin tests no modifier was present.

Exposure intensity

Median sarin inhalation exposure intensity was 12.8 mg.min/m^3 (Table 3). Most (2,229, 75%) of the participants in sarin tests had exposures to sarin of $\leq$15 mg.min/m^3. The maximum inhalation exposure to sarin was recorded as 48.5 mg.min/m^3 (in 1967), but all tests with sarin at exposure intensities over 15 mg.min/m^3 were in the presence of (physical) protection, except 22 single breath inhalation tests (range 15 to 19.6 mg.min/m^3). Median Ct varied by protection status. It was lowest in tests with physical protection present and highest in tests with chemical protection only.

Median dermal exposure intensity for sarin exposures was 200 mg (IQR 8-250 mg). The maximum dermal sarin exposure was 300 mg (Table 3). Most (188, 65%) dermal exposures had physical protection present, often a small piece of fabric taped to the skin onto which the drop of sarin was administered. All dermal sarin exposures took place before February 1953.

Duration of exposure

The median duration of all sarin exposures, and of inhalation exposures, was 15 minutes. The duration was shorter in inhalation tests where physical protection was present (2 mins). The duration of sarin dermal exposures was 30 mins in all but one test (Table 4). There were 153 unprotected sarin inhalation tests whose duration was greater than 60 minutes but in none of these did the exposure intensity exceed 16.2 mg.min/m^3. There were two tests with recorded durations of 400 minutes and these were with low chamber concentrations of 0.005 mg/m^3, resulting in an exposure of 2 mg.min/m^3.

Measures of acute biological effect

Table 5 shows the numbers of sarin inhalation and dermal tests for which data were available on both exposure and acute biological response. Unspecified cholinesterase was a measure used only in the 1950s, in 692 tests. All dermal tests where a biological effect was assessed had cholinesterase inhibition measured using unspecified cholinesterase. In most tests after the end of the 1950s three cholinesterase types were measured: red blood cell, plasma and whole blood, though more whole blood measures overall were made as it was the sole measure of biological effect in some tests in the 1950s.

Table 6 shows the median and maximum cholinesterase inhibition by cholinesterase type and exposure route. For dermal exposures, only unspecified cholinesterase was available. For inhalation exposures the median cholinesterase depression was greatest for red blood cell cholinesterase. There are marked differences in the cholinesterase inhibition by cholinesterase type for ocular exposures.

For sarin inhalation exposures overall there was a median red blood cell cholinesterase inhibition of 41% (IQR 24-51). When stratified by protection type little difference was noted in this median inhibition, though it was least (33%) in tests where no protection was present

(Table 7). In dermal tests with physical protection the median cholinesterase depression was 31%.

Relationship between sarin exposure intensity and cholinesterase activity

There were 2,353 sarin inhalation tests with a measure of exposure intensity (Ct) and in 1,436 (61%) of these a cholinesterase measure had also been documented. For dermal tests the figures were 282 and 273 (96%) respectively (Table 5). In sarin inhalation tests, red blood cell cholinesterase activity decreased with increasing unprotected exposures (Figure 1). Non-parametric kernel regression of the untransformed data indicates a exposure-response relationship between unprotected inhalation exposure to sarin and red blood cell cholinesterase inhibition (Figure 2). For dermal sarin tests (Figure 3) with physical protection there was a exposure-response relationship between exposure and cholinesterase inhibition. Figure 3 also shows that in dermal exposures ≥200 mg with physical protection there was pronounced variation in inhibition of cholinesterase activity. In 11 of these tests the maximum inhibition was ≥80% in and >90% in 5 tests. When there was no protection present, maximum dermal exposure did not exceed 44 mg and the maximum decrease in cholinesterase activity was 50%.

Pupil size

Between May 1949 and December 1980 pupil size was one of the measures used to assess the effect of nerve agent exposure, 90% of these tests taking place before 1973. Pupil diameter decreased with increasing exposure, and with decreased red blood cell cholinesterase activity in unprotected inhalation tests (Table 8). The direction and magnitude of the relationships was less clear, or even reversed, when protection was present.

Discussion

In our earlier papers, we documented the numbers of participants exposed to nerve agents, and examined the cause specific mortality and cancer incidence in those exposed (Carpenter et al. 2009; Keegan et al. 2009; Venables et al. 2009). This paper now presents in greater detail information on tests with nerve agents, including exposure-response relationships for acute effects.

Most nerve agent tests were with sarin, and for most of those tests the exposure route was via inhalation, with dermal tests also taking place between October 1951 and May 1953. The resultant change in cholinesterase activity was high in some tests (a depression of around 90%). Indeed, in May 1953, a national serviceman, Ronald Maddison, died as a result of his exposure to sarin (Ministry of Defence 2006). He had taken part in a dermal test with an exposure of 200 mg of sarin. A subsequent report into the tragedy, and more widely into testing involving servicemen, was compiled by a committee led by the then president of the Royal Society, Lord Adrian. It concluded that nerve agent exposures should not exceed a Ct of 15 mg.min/m^3 and should be to sarin only (Ministry of Defence 2006). The inquest into Ronald Maddison's death, held *in camera*, returned a verdict of accidental death. However, the inquest was reopened and in 2004 returned a verdict of unlawful killing (Schmidt 2006).

These data give a rare insight into cholinesterase activity after nerve agent exposure and indicate that in unprotected tests there was a clear relationship between increasing exposure and decrease in red blood cell cholinesterase, and one of the strengths of our study is that not only was red blood cell cholinesterase reported consistently across three decades but it is also a good predictor of synaptic cholinesterase activity (Bajgar 1992; Bajgar 2004). The use of just one of the three measure of cholinesterase activity in our analysis of the exposure-response relationship was intended to minimise uncertainly in validity of exposure measures

which would have arisen with use of different biomarkers of cholinesterase activity. Of the three cholinesterase measures available to us, whole blood cholinesterase activity is considered an acceptable marker for organophosphate exposure but further inference from it is limited (Bajgar 2004). Both red blood cell cholinesterase and plasma cholinesterase are informative but plasma cholinesterase is understood to capture change in the activity of butrylcholinesterase, whose physiological function is unknown (Marrs et al. 2007).

Variability of cholinesterase response to sarin exposures, both inhaled and dermal, was high. For example, in sarin exposures of 15 mg.min/m^3, the cholinesterase inhibition varied between 5% to 60%. The variability when either chemical or physical protection was present, for both inhalation and dermal exposure routes, was greater at higher exposure intensities. This may not reflect inability of protective measures to prevent exposure, but the wide range of types of protection, particularly physical. Details of the exact chemical modifier present during each test were not captured in this dataset but could be examined in a future analysis.

Part of the variability in response might be biological. Whilst the range of natural annual within-individual variation in red blood cell cholinesterase is 8% of the annual mean (Sidell and Kaminskis 1975), annual within-individual variability in plasma cholinesterase is higher and may be 25%. It has been suggested that, in an individual, a change in red blood cell cholinesterase of more than 8% should 'alert one to look for a cause' (Sidell and Kaminskis 1975). Our data did not allow for an assessment of within-individual variation because our analysis is at the level of the test and repeated exposures to individuals were uncommon.

In a study of workers exposed to organophosphate pesticides, full body protection (not including respiratory protection) significantly attenuated the effect of the pesticides on cholinesterase activity whereas face and glove protection alone did not (Lander and Hinke 1992). The effect of physical protection was not clear in our data, which may be related to the

heterogeneity of the physical materials placed between the exposure and the skin. No clear effect of chemical protection was seen in our data; again, there was heterogeneity in types of chemical protection employed.

One known ocular effect of organophosphates is constriction of the pupil, or miosis (Nozaki et al. 1997). Our results show that in unprotected tests the pupil constricted as exposure intensity increased. Sarin also causes eye pain and visual disturbances (Sidell and Borak 1992) which add to its impact on military effectiveness (Ministry of Defence 2006). Research at Porton Down in the 1940s and early 1950s had shown that the single exposure threshold at which miosis, but no other symptom, was apparent was 3.3 mg.min/m^3, leading to a recommendation that miosis be considered a marker for exposure to a low concentration of nerve agent (Ministry of Defence 2006). Other early studies at Porton Down on the effects of tabun on visual acuity showed a linear relationship between tabun exposure and decrease in pupil diameter ((Marrs et al. 2007). Miosis is still a clinically-relevant marker of exposure: in a study of people exposed to sarin in the terrorist attack on the Tokyo underground in 1995, it was found that red blood cell cholinesterase activity was significantly lower in people who were miotic (pupil size <3mm) than those who were not (Nozaki et al. 1997).

The response measure percentage change in pupil diameter was calculated from before- and after-exposure measurements of pupil diameter in millimetres. The most commonly-noted changes were 0 mm (15%), 1 mm (15%), 1.25 mm (18%) and 1.5 mm (16%). Pupil size was used as a response measure over a ten-year period (1949-1959).

In our data the effect on the eye of unmodified exposure to sarin was to decrease pupil diameter. This decrease in pupil diameter was not apparent when the exposure to sarin was modified by chemical protection; the effect then was to increase the diameter of the pupil. This may be because in-test protection against nerve agents was provided by pre-treatment

with pyridostigmine which, like sarin, is an acetylcholine antagonist (but which unlike sarin has an effect that is reversible). The effect of exposure with physical protection was statistically significantly smaller than that observed with no protection. Physical protection comprises a variety of protective measures, including respirator use, and this result would indicate that measures were successful in limiting exposure. However, some materials coded as protective, such as textiles used for military uniforms, were under test for their protective efficacy but later were found to increase dermal exposure by limiting evaporation of nerve agents (Ministry of Defence 2006; Organisation for the Prohibition of Chemical Weapons 2015). The increase in cholinesterase depression with dermal exposure could be explained by this effect. The lack of cholinesterase depression shown in unprotected dermal exposures is likely the result of the smaller quantities administered (maximum 40 mg) compared to those given with protection (maximum 300 mg).

Chamber exposures were expressed as 'Ct', the product of concentration (C) and time (t). While Haber's law would imply that the identical products of C x t will provoke identical biological responses, this is often true only for a set range of concentrations (Witschi 1999). A high concentration of nerve agent administered in a short time could be lethal and a low concentration for a longer time effectively harmless. For example, an exposure of 300 mg/m^3 for 3 seconds and exposure of 0.15 mg/m^3 for 100 minutes both result in a Ct of 15 mg.min/m^3, but the former would result in a chamber concentration of 48 ppm sarin, the latter 0.025 ppm. One estimate for the human LD_{50} of sarin is 1.2ppm (Ellison 2000). In tests where the duration was 30 minutes, the chamber air concentrations during the tests would have been on average, 0.5 mg/m^3 (0.08 ppm). It has also been assumed that identical cumulative exposures, calculated as the product of different combinations of time and exposure concentration, are associated with identical risk of disease, but this too is an assumption that needs refining (Smith and Kriebel 2013).

Since nerve agents are banned under the Chemical Weapons Convention (Organisation for the Prohibition of Chemical Weapons 1997) and any human testing is likely to have been done by governments, and secretly, there is little in the open scientific literature with which to compare our findings. However, the US National Research Council has reported on the mortality of US veterans of a chemical testing programme at Edgewood Arsenal, finding no increase in all-cause or cause-specific mortality in 846 veterans of tests with anti-cholinesterases (National Research Council and Committee on Toxicology 1985). Their exposure data are difficult to compare to ours because only a sample of all exposures was reported, and no acute biological response data were reported.

Detailed internal technical reports on experiments with nerve agents were prepared at Porton Down and many are declassified and available from the UK National Archive. Our data, containing as it does results of many experiments spanning decades, is unusual in its scale. However, it is acknowledged that the individual experiments were not designed to a standard template and there are therefore limitations to the inferences we can draw in an analysis of them as a single body. Notwithstanding that, these results offer a unique insight into the experiments carried out with nerve agents at Porton Down in the five decades following WWII. While chemical weapons have not been widely used in major conflicts during that period, their use, especially against civilians, has increased as formerly stable states possessing chemical arsenals have become destabilised.

Table 1 Number of participants exposed to nerve agents at Porton Down 1945-1987.

Chemicals (abbreviated name)	n	Number of tests	Number of tests (%)	Tests per participant (median)	Date range	75% of tests complete by
Sarin (GB)[a]	2,980	3,511	82	1	1949-87	1959
Tabun (GA)[b]	262	398	9	2	1945-52	1951
Soman (GD)[c]	141	149	4	1	1950-63	1952
GE[d]	25	25	1	1	1949-50	1950
Cyclo-sarin (GF)[e]	166	167	4	1	1950-64	1963
VX[f]	48	49	1	1	1960-69	1969
All nerve agents	3,622*	4,299	100	1	1945-87	1959

[a] Iso-propylmethylphosphonofluoridate (CAS
[b] Ethyl N,N- dimethylphosphoramidocyanidate
[c] Pinacolyl methylphosphonofluoridate
d Isopropyl ethylphosphonofluoridate
[e] Cyclohexylmethyl phosphonofluoridate
f O-ethyl S-diisopropylaminoethyl methyl phosphonofluoridate
* Number of veterans exposed to any nerve agent = 3,597. N=25 veterans were exposed to more than one nerve agent.

Table 2 Sarin tests by exposure route and presence of physical or chemical modifiers.

Modifier type	Exposure route				
	Inhalation (%)[a]	Dermal only (%)	Ocular only (%)	Not recorded (%)	Total (%)
Chemical only	567 (19)	0	2 (2)	8 (6)	577 (16)
Physical only	669 (22)	194 (67)	26 (24)	8 (6)	897 (25)
Respirator	468	194	0	1	663
No respirator	201	0	26	7	234
Chemical and physical	144 (5)	0	0	15 (11)	159 (5)
Respirator	2	0	0	0	2
No respirator	142	0	0	15	0
None	1,592 (54)	94 (33)	82 (74)	110 (78)	1,878 (53)
Total	2,972 (100)	288 (100)	110 (100)	141 (100)	3,511 (100)

[a] +/- other exposure routes

Table 3 Exposure intensity of sarin tests by exposure route and protection status.

Exposure route and modifier type	n	Median	IQR	Maximum
Inhalation[a]:	**(mg.min/m^3)**			
Chemical only	545	14.9	12.0-15.0	16.6
Physical only	437	10.7	7.4-13.9	48.5
Chemical and physical	144	12.3	10.3-12.9	44.9
None	1,227	11.8	5.1-14.8	19.6
Total	2,972	12.8	7.5-15.0	48.5
Dermal only:				**(mg)**
Physical only	188	250.0	200-300.0	300.0
None	94	0.50	0.3-8.0	44.0
Total	288	200.0	8.0-250.0	300.0

[a] +/- other exposure routes

Table 4 Duration of sarin tests (mins) by exposure route and protection status.

Exposure route and modifier type	n	Median (mins)	IQR (mins)	Maximum (mins)
Inhalation[a]:				
Chemical only	552	15.0	15.0-15.0	75.0
Physical only	433	2.0	2.0-30.0	150.0
Chemical and physical	144	2.0	1.7-2.0	30.0
None	1,374	15.0	2.0-30.0	400.0
Total (inhalation)	2,503	15.0	0.3-28.0	400.0
Dermal only:				
Physical only	84	30.0	30.0-30.0	30.0
None	1	5.0	n/a	5.0
Total (dermal)	85	30.0	30.0-30.0	30.0

[a] +/- other exposure routes

Table 5 Number of sarin tests with exposure intensity and/or biological response data available, by decade.

Decade	Intensity inhalation[a]		Intensity dermal only		Cholinesterase type				Total sarin inhalation tests	Total sarin dermal tests
	Ct	Ct + ChE	Quantity (mg)	Quantity (mg) + ChE[c]	Whole blood	Red blood cell	Plasma	Unspecified		
1940s	108	0			0	0	0	0	173	
1950s	1,703	1,096	282	273	945	579	420	692	2,154	288
1960s	214	195			266	271	270	0	286	
1970s	236	96			96	96	95	0	249	
1980s	92	49			51	46	46	0	110	
All years	2,353	1,436	282	273	1358	992	831	692	2,972	288

[a] +/- other exposure routes

[b]Cholinesterase

[c] tests which contain a value for exposure intensity (Ct) and at least one measure of percentage change in cholinesterase (ChE) activity

Table 6 Percentage inhibition in cholinesterase activity, by exposure route and type of cholinesterase.

Exposure route and cholinesterase type	n	Median (%)[b]	IQR (%)[2]	Maximum (%)
Inhalation[a]:				
Red blood cell	915	41	24-51	87
Plasma	803	24	15-33	67
Whole blood	1,327	32	18-42	93
Unspecified	370	20	9-30	74
Dermal only:				
Red blood cell	0	.	.	.
Plasma	0	.	.	.
Whole blood	0	.	.	.
Unspecified	279	22	10-37	99
Ocular only:				
Red blood cell	26	-1	-4-3	10
Plasma	1	63	n/a	63
Whole blood	4	40	19-52	58
Unspecified	19	-1	-3-4	30
Not recorded:				
Red blood cell	51	19	10-53	67
Plasma	27	25	2-38	49
Whole blood	27	40	3-51	60
Unspecified	24	10	7-15	22
Total:				
Red blood cell	992	40	21-51	87
Plasma	831	24	15-33	67
Whole blood	1,358	32	18-43	93
Unspecified	692	19	9-32	99

[a] +/- other exposure routes

[b] Negative values indicate an increase in cholinesterase activity.

Table 7 Percentage inhibition in red blood cell and unspecified cholinesterase, by exposure route and modifier type.

Exposure route and protection status	n	Median (%)	IQR (%)	Maximum (%)
Inhalation[a]:	**Red blood cell cholinesterase**			
Chemical only	344	48	39-54	73
Physical only	20	46	21-53	60
Chemical and physical	2	47	40-54	54
None	549	33	19-48	87
Total	915	41	24-51	87
Dermal only:	**Unspecified cholinesterase**			
Physical only	191	31	19-46	99
None	88	5	0-13	50
Total	279	22	10-37	99

[a] +/- other exposure routes

Table 8 Results of linear regression of percentage change in pupil diameter on a) inhalation exposure (mg.min/m^3), and b) percentage change in red blood cell cholinesterase activity, by protection status.

Protection status	n	B[a]	const	r^2	p
a) Pupil diameter v. Ct (mg.min/m^3)					
Chemical only	196	-0.62	61.0	0.02	0.02
Physical only	50	0.58	29.8	0.17	0.001
Chemical and physical	138	0.22	43.2	<0.01	0.166
None	360	2.60	26.0	0.43	<0.001
b) Pupil diameter v. RBC cholinesterase					
Chemical only	55	-0.02	54.2	-0.02	0.87
None	45	0.40	34.7	0.40	<0.001

[a]A negative coefficient indicates an increase in pupil diameter as sarin exposure increased.

Figure 1 Scatter plots of sarin inhalation exposure and percentage change in activity of a) red blood cell, b) plasma, c) whole blood, and d) unspecified cholinesterase. e) Sarin dermal exposure and percentage change in activity of unspecified cholinesterase.

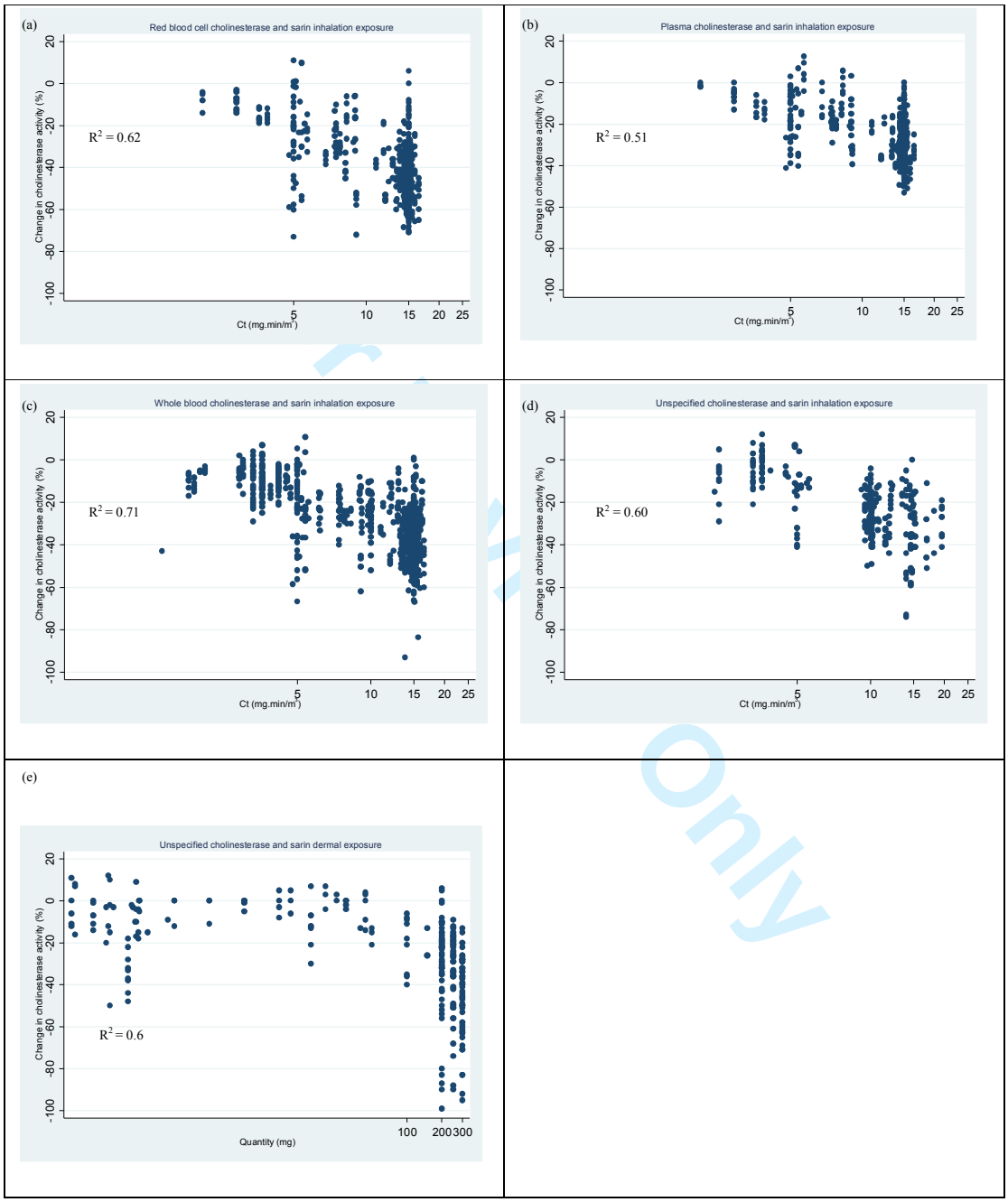


Figure 2 Non-parametric kernel regression estimate and 95% CIs for sarin inhalation exposure and percentage change in red blood cell cholinesterase activity, with a) chemical protection (n = 326) and b) no protection (n = 332).

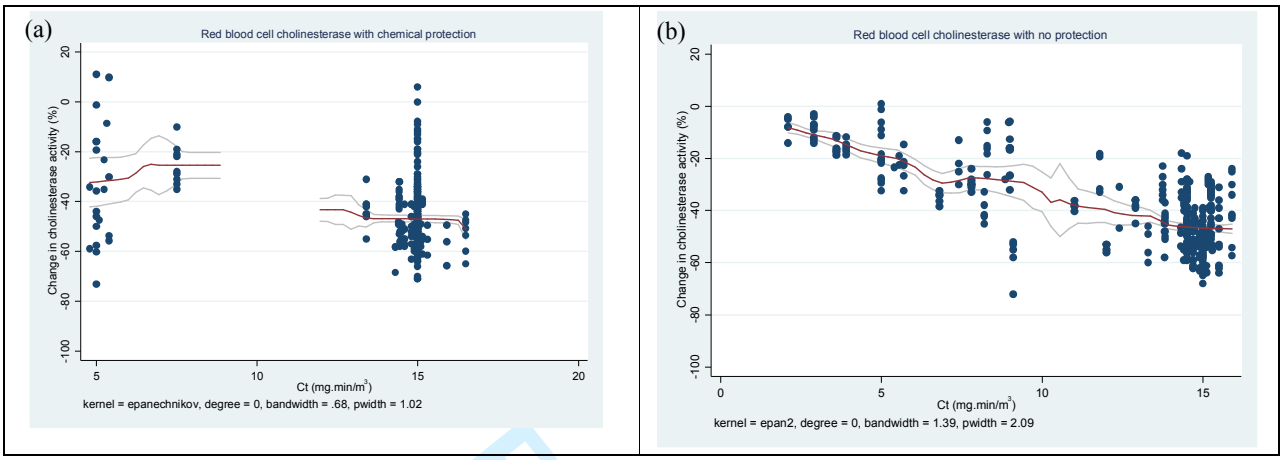
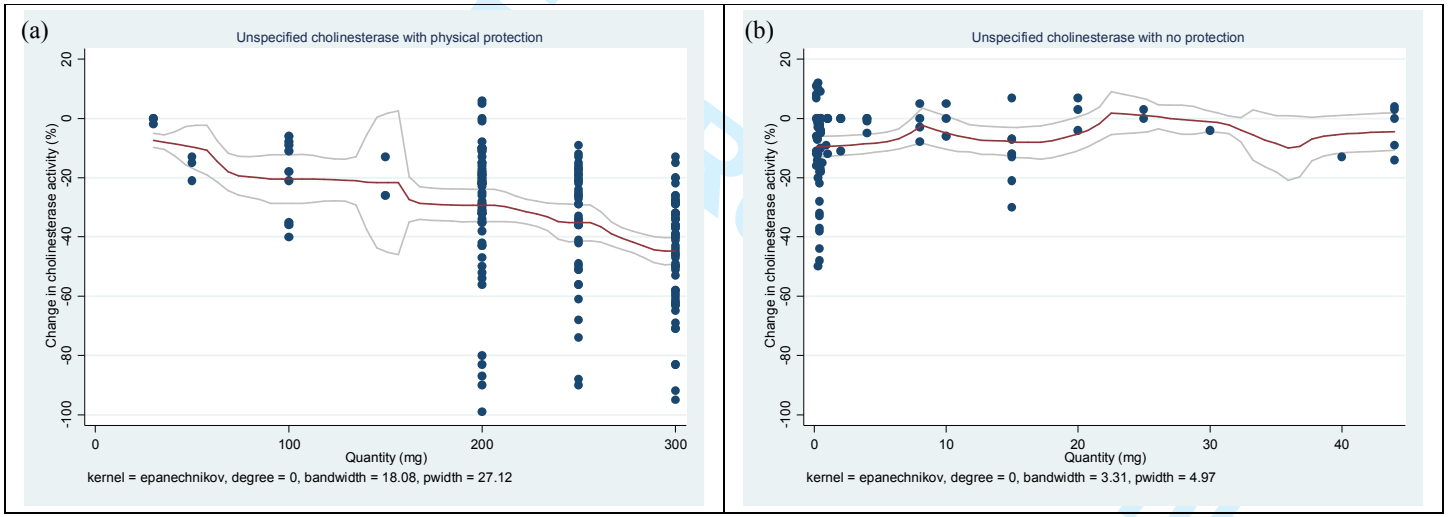


Figure 3 Non-parametric kernel regression estimate and 95% CIs for sarin dermal exposure and percentage change in unspecified cholinesterase activity, with a) physical protection (n = 185), and b) no protection (n = 88).



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