DEPARTMENT OF THE ARMY
U.S. ARMY SOLDIER AND BIOLOGICAL CHEMICAL COMMAND
5183 BLACKHAWK ROAD
ABERDEEN PROVING GROUND, MARYLAND 21010-5423

REPLY TO
ATTENTION OF

August 16, 1999

Freedom of Information and Privacy Act Office

Mr. John Greenewald, Jr.
[redacted]

Dear Mr. Greenewald:

In response to your June 24, 1999, Freedom of Information Act (FOIA) request, I have enclosed a sanitized copy of "Exposure of Human Subjects to Aerosols of Large-Sized EA 3528 Particulates". This document has been reviewed by our Chief Scientist and by our attorneys. Subject names and initials are being withheld under FOIA Exemption Number 6; technical data is being withheld under FOIA Exemption Number 3.

If you are not satisfied with the documents as provided, please contact me again in writing. Upon receipt of your correspondence, I will forward your request to our Initial Denial Authority (IDA) who is our legal Chief Counsel for final review and action.

Fees incurred while processing this request have been waived.

Sincerely,

Cheryl S. Fields
Freedom of Information and
Privacy Act Officer

Enclosure

EDGEWOOD ARSENAL
TECHNICAL REPORT

EATR 4262

EXPOSURE OF HUMAN SUBJECTS TO AEROSOLS OF LARGE-SIZED EA 3528 PARTICULATES (U)

by

John W. Simmonds
Chester W. Gottlieb
Kragg P. Kysor
John T. Weimer
Nicholas Montanarelli

March 1969

DEPARTMENT OF THE ARMY
EDGEWOOD ARSENAL
Research Laboratories
Medical Research Laboratory
Edgewood Arsenal, Maryland 21010

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EDGEWOOD ARSENAL TECHNICAL REPORT

EATR 4262

EXPOSURE OF HUMAN SUBJECTS TO AEROSOLS OF LARGE-SIZED EA 3528 PARTICULATES (U)

by

John W. Simmonds, CPT, MC
Chester W. Gottlieb, CPT, MC
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John T. Weimer
Nicholas Montanarelli

Clinical Research Department

March 1969

Task 1B522301A07908

DEPARTMENT OF THE ARMY
EDGEWOOD ARSENAL
Research Laboratories
Medical Research Laboratory
Edgewood Arsenal, Maryland 21010

(U) FOREWORD

The work described in this report was authorized under Task 1B562602A07908, Non-Defense Medical Aspects of Chemical Agents, Incapacitating and Riot Control Agents (U). This work was started in February 1966 and completed in November 1966.

The volunteers in these tests are enlisted US Army personnel. These tests are governed by the principles, policies, and rules for medical volunteers as established in AR 70-25.

Case numbers of all subjects used in this study are appended.

(C) [redacted]

(C) [redacted]

1. (C) [redacted]

2. (U) The inhalation ID50 (retained dose) of the 12-micron particle is equivalent in size to the incapacitating intravenous or oral dose of EA 3528; the ICt50 is one-third as large as the incapacitating dose of an 0.8-micron (MMD) particle aerosol;

3. (U) Retention of the 12-micron (MMD) particle aerosol by the respiratory passages is essentially complete;

4. (U) Significant agent plasma levels are achieved within 15 minutes when the route of administration of agent is inhalation and the absorptive surfaces are the respiratory passages.

(U) CONTENTS

(U) LIST OF FIGURES

(U) LIST OF TABLES

(C) EXPOSURE OF HUMAN SUBJECTS TO AEROSOLS OF LARGE-SIZED EA 3528 PARTICULATES (U)

I. (C) INTRODUCTION.

(C) [redacted]

(U) In the aerosol studies the calculated retention of agent varied from 60 to 92 percent with a mean retention of approximately 80 percent. This figure is considerably higher than the 50 to 60 percent retention anticipated from established data in the literature relating pulmonary retention to particle size;[3] such disparity may account for the differences between the incapacitating doses by the inhalation, oral, and intravenous routes. Technical considerations suggest that the particles became electrostatically charged during aerosol dissemination and thus increased deposition in the respiratory tract.

(U) The question arose of whether it is necessary to keep the particle size of EA 3528 within narrow limits (MMD of 1 micron). If such a requirement were not stringent, manufacturing costs could be decreased. A study was therefore undertaken to compare the relative effectiveness of large (10- to 12-micron MMD) and small (0.8- to 1.0-micron MMD) particulate aerosols of EA 3528. This report summarizes data on the exposure of 20 volunteers to 12-micron MMD EA 3528 particulate aerosols.

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*(U) Incapacitation is defined here as two or more successive Number Facility (NF) scores below 10 percent of the baseline value. This performance level correlates well with the clinical impression that individuals at this level of performance are unable to carry out normal activity or to obey simple commands.

**(U) Corrected Ct is the Ct adjusted for standard conditions of a 75-kilogram man breathing 15 liters in a minute.

†(U) The free base equivalent of EA 3528 is 0.75 times the maleate by weight. Concentration, Ct and corrected Ct, are expressed as the maleate in this report so that values may be compared with those reported earlier.

II. (U) MATERIALS AND METHODS.

A. Agent.

EA 3528 of 98 percent purity was preground to yield a distribution of particles with an MMD of 12 microns. The mass distribution of particles (table I) was measured in a calibration trial from a Rochester Cascade Impactor placed in the wind tunnel at the same location and distance from the disseminating nozzle as the volunteer's head. MMD is that diameter above which and below which 50 percent of the agent mass occurs. Table II is a list of the distribution of actual particle sizes derived from microscopic particle sizing and counting. The range of particle sizes is 2.5 to 25.0 microns. The median diameter is smaller than the MMD because a single large particle will contribute more to the overall mass of agent than a small particle.

Table I (U). Mass Median Diameter* of EA 3528 Particles as Derived From the Rochester Cascade Impactor in Large Particle Studies: Average of Four Determinations

Stage Calibration		Mass Distribution	Actual Distribution	Cumulative Distribution
Stage No.	Particle Size μ	μg	%	%
I	12.6	130	44	44
II	5.3	72	24	68
III	2.7	30	12	80
IV	1.5	20	7	87
V	0.8	24	8	95
VI	0.4	15	5	100

*Mass median Diameter is 12 microns.

Table II (U). Microscopic Particle Sizing and Count of Large EA 3528 Particles With a Probit Analysis

Particle Size μ	Actual Occurrence %	Cumulative Occurrence %	Statistical Analysis						
			P	(ED(P)	Lower	Upper	Prob Y	Log X	Std Error
2.5	2	2	1	2.06	1.69	2.51	1.43	3.98	2.34
5.0	22	24	16	4.45	3.97	4.99			
7.5	23	47	30	5.84	5.34	6.39			
10.0	18	65	50	7.91	7.38	8.47			
12.5	11	76	84	14.06	13.06	15.14			
15.0	9	85	99	30.39	28.29	35.13			
17.8	4	89							
20.0	7	96							
25.0	4	100							

Total number of particles sized is 1300.
Median diameter is 7.9 microns.
Average particle diameter is 10.8 microns.

B. Disseminating Vehicle.

EA 3528 was suspended in Freon 11 in a covered flask and constantly agitated with a magnetic stirring device. The concentration of the agent in the Freon was approximately 2 percent.

C. Disseminating Device.

A pneumatic spray nozzle under a pressure varying between 15 and 35 psi was used to disseminate the particulate cloud. A fairly uniform spray was produced although the spray occasionally became erratic because of partial plugging of the nozzle orifice. The spray was visually directed at the target area where the volunteer's head was positioned. The 1-1/2 by 2-foot outdoor rectangular wind tunnel was operated at a 5-mph wind speed.

D. Measurement of Agent Concentration During Exposure.

Three filter paper samples positioned around the subject's head collected agent during calibration runs and exposures. Aerosol-laden air was drawn through the sampling devices at a flow rate of 5 l/min. The amount of agent in each sampler was then calculated by eluting the agent from the filter paper and determining agent concentration by UV absorption spectroscopy at 314 millimicrons against known agent standards. Considerable variation occurred in the total agent collected per sampler in each run. The mean of the content of the three samplers was used to calculate the concentration of agent in the inspired air.

E. Regulation of Breathing Rate and Volume.

An oscilloscope was set at a recurring sweep duration of 5 seconds; i.e., 12 sweeps a minute, and was calibrated to reflect inspiratory airflow. The calibration was determined by a differential pressure flowmeter attached to the mouthpiece assembly. A pressure transducer, preamplifier, and transducer monitor coupler provided the necessary conversion of the flow rate into an electronic signal (figure 1). The airflow meter was calibrated against known flow rates, and a curve representing the desired inspiratory flow rate was constructed and inscribed on the oscilloscope screen. The curve, 2 seconds in duration, was so constructed that, when identically transcribed by the volunteer's breathing pattern, an inspired volume of approximately 1.2 l/breath, or about 15 l/min, was achieved. After repetitive practice sessions, all volunteers were able to modify their inspiratory depth and rate to follow the inscribed pattern closely. A permanent record of the inspiratory flow changes was obtained using a DC-coupling pre-amp and paper write-out monitor. Subsequent analysis of this graphic record permitted indirect estimates of inspired volume. The reliability of these estimates, however, is not known.

F. Measurement of Expired Volume.

A manually controlled valve assembly was constructed to facilitate collection and measurement of expired air and expired agent. This assembly enabled each volunteer to inhale aerosol-laden air through one corridor of the Y-shaped assembly and expel exhaled air through

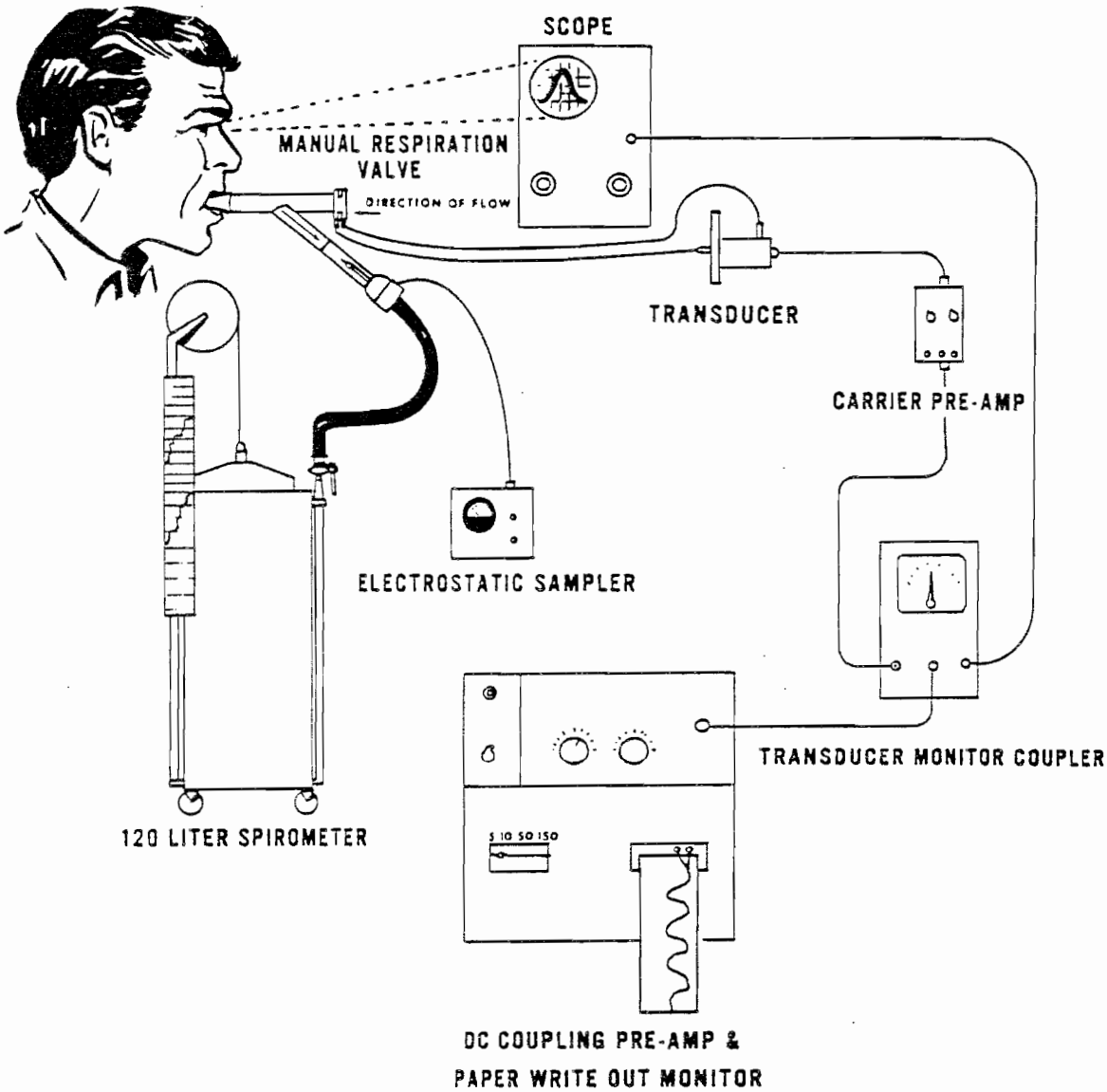


Figure 1 (U). Apparatus for Regulating and Measuring Breathing Rate and Volume

the other corridor (figure 2). Expired air was directed through an electrostatic precipitator into a Collins chain-compensated gasometer (spirometer). As indicated in figure 1, movement of the tank occurred only when the valve was open to the side (exit) corridor; ideally, only during expiration. However, if opening of the manually operated valve was slightly delayed or the open valve was closed prematurely, an underestimate of expired volume resulted. Any such underestimate of expired volume probably did not exceed 10 to 15 percent of the total air volume measured.

G. Measurement of Expired Agent.

Agent present in the exhaled air was collected in an electrostatic precipitator located just beyond the outflow valve. The collection tube was then rinsed twice in 20 milliliters of dilute HCl and the agent sample was analyzed by photofluorometric methods to determine the amount of agent in the expired air.

H. Approximation of Dose.

An approximation of the dose to be delivered for each man was made from a knowledge of the agent concentration in prior tunnel calibration runs, the man's weight, and the expected degree of agent retention. Any given exposure was terminated when the expired volume, reflected by the spirometer, reached a preselected value that would deliver approximately the desired dose.

I. Measurement of Plasma Concentration of Agent.

Samples of blood were drawn and placed in heparinized tubes prior to agent exposure (control) and at the following experimental times after exposure: 0005, 0015, 0030, 0100, 0200, 0300, and 0600. The plasma agent level was subsequently determined by a modification of the spectrophotofluorometric method.[4] This same method was employed to determine agent concentration in the expired air. Occasionally, it was not possible to obtain blood samples from the volunteers because of behavioral disturbances caused by the agent. Also, in several instances, the quantity of blood drawn was insufficient for analysis; on one test day laboratory determinations on four men (DO, ZN, AM, BU) were discarded because technical difficulties invalidated them.

J. Laboratory Examinations.

Normal control values for all subjects were obtained for the following: total and differential white blood cell (WBC) count, hematocrit (Hct), hemoglobin (Hbg), blood urea nitrogen (BUN), bilirubin, alkaline phosphatase, sulfobromophthalein, serum glutamic oxaloacetic transaminase (SGOT), total protein, albumin, thymol turbidity, and urinalysis. Additional laboratory examinations one and seven days after drug administration included values for total and differential WBC count, Hct, Hbg, BUN, bilirubin, SGOT, alkaline phosphatase, and urinalysis.

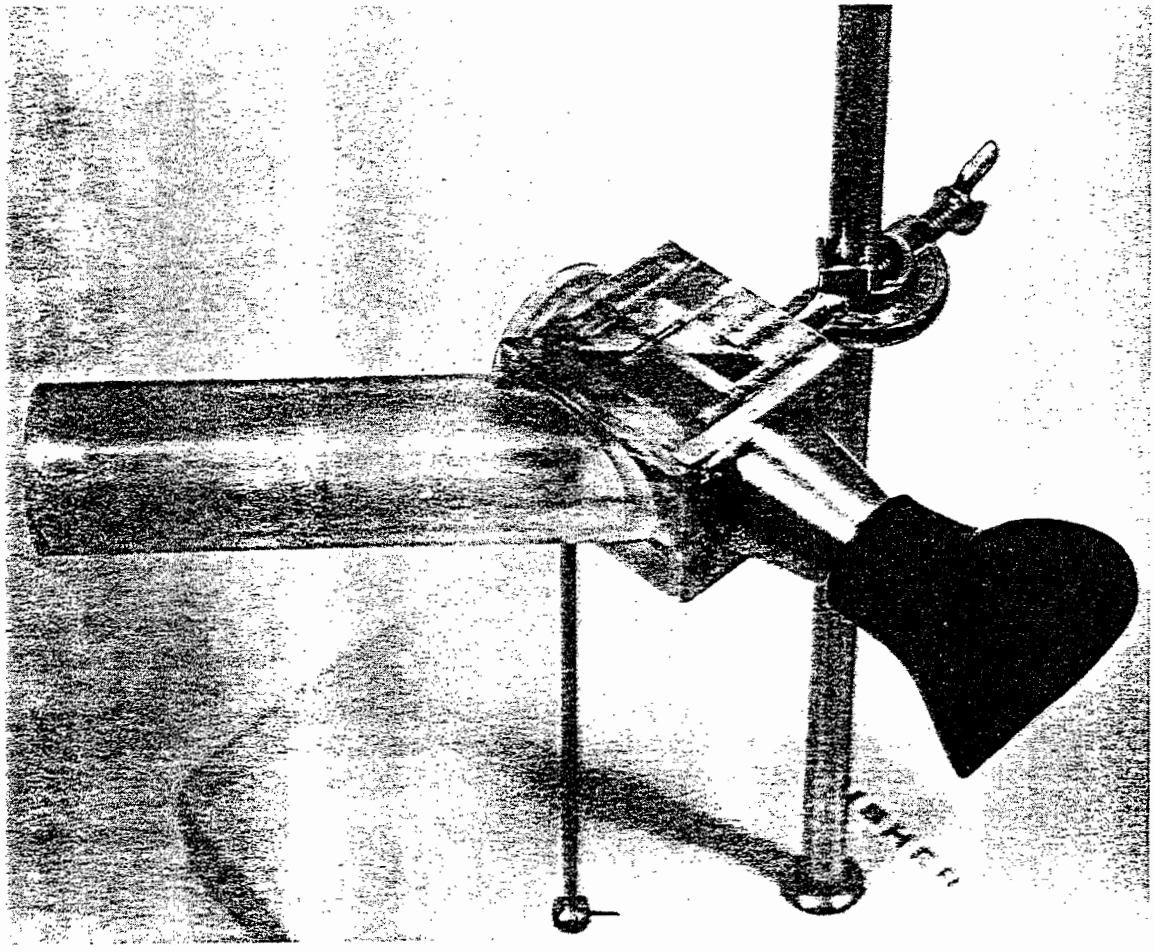


Figure 2 (U). Valve Assembly for Collecting Expired Air

K. Physiological Measurements.

Baseline physiological measurements of supine blood pressure and pulse, axillary temperature, respiratory rate, and pupil size were obtained on the evening before and the morning of testing. These parameters were monitored at frequent intervals after drug administration.

L. Performance Measures.

In the pretest period, all volunteers completed 20 NF tests, with an additional two tests on the evening before and the morning of testing. The best three of the first 20 and the best two of the last four scores were averaged to determine the baseline score for each man. Following exposure, NF tests were administered at the following experimental times: 0015, 0030, 0100, 0130, 0200, 0300, 0400, 0500, 0600, 0800, 1000, 1200, 1600, 2000, and 2400.

M. Behavioral Tests.

Each volunteer underwent family background analysis, the Minnesota Multiphasic Personality Inventory, and a psychiatric interview prior to testing. Pretest briefings included information regarding the nature of anticipated reaction. Observations recorded by the nurses and medical aides throughout the test period included a behavior checklist. The volunteer completed a symptom check list and a written summary of his experience 24 hours after drug administration.

N. Additional Studies.

Electroencephalograms were recorded on all men prior to testing and on a few men during the course of testing.

O. Conduct and Experiment.

After completion of medical, psychiatric, and laboratory screening, the volunteers were briefed about the nature of the experiment. Those who volunteered for testing were given additional baseline performance tests and were trained to breathe according to the pattern previously described. On the evening prior to testing, volunteers reported to the ward area for final physiological and behavioral baseline determinations. After a light breakfast and final baseline measurement, each volunteer was transported to the wind tunnel area by ambulance. Wind tunnel calibrations were established and confirmed prior to testing. The volunteers then established a satisfactory breathing pattern in the wind tunnel, and the aerosol exposure was initiated. The length of exposure varied from 1 to 4 minutes. At the termination of exposure, the volunteer was accompanied to a waiting ambulance where the first blood sample was drawn. He was then transported to the psychopharmacology protected ward area where the testing procedures were carried out for the ensuing 24 hours. Followup laboratory data were obtained 24 hours and 7 days after exposure.

III. (C) [redacted]

A. (C) [redacted]

(C) [redacted]

(U) Table III shows each volunteer's weight, exposure time, minute volume, and the total volume of expired air. The inspired volume was not used in the calculations because it would only have been extrapolated from the polygraph record of instantaneous air flow. The recorded value of expired air was an estimated 10 percent less than the actual amount of inspired air. The expired volume was used for calculations. As shown in table III, the minute volumes differed considerably from the desired 15 l/min, with a mean for the 20 volunteers of 13.1 l/min and a range of 6.8 to 18.6 l/min. The mean exposure time was 104 seconds with a range of 55 to 228 seconds.

(U) Table IV shows the relationship between retained dose, Ct, and incapacitation. The data are listed in order of increasing retained dose expressed as the free base equivalent. Incapacitation is defined in this study as two or more successive NF scores below 10 percent of baseline. This criterion correlates well with clinical estimates of incapacitation. At this NF level the volunteers were unable to perform simple tasks. This definition of incapacitation permits comparison with previous drug studies in which NF performance was measured.

(C) [redacted]

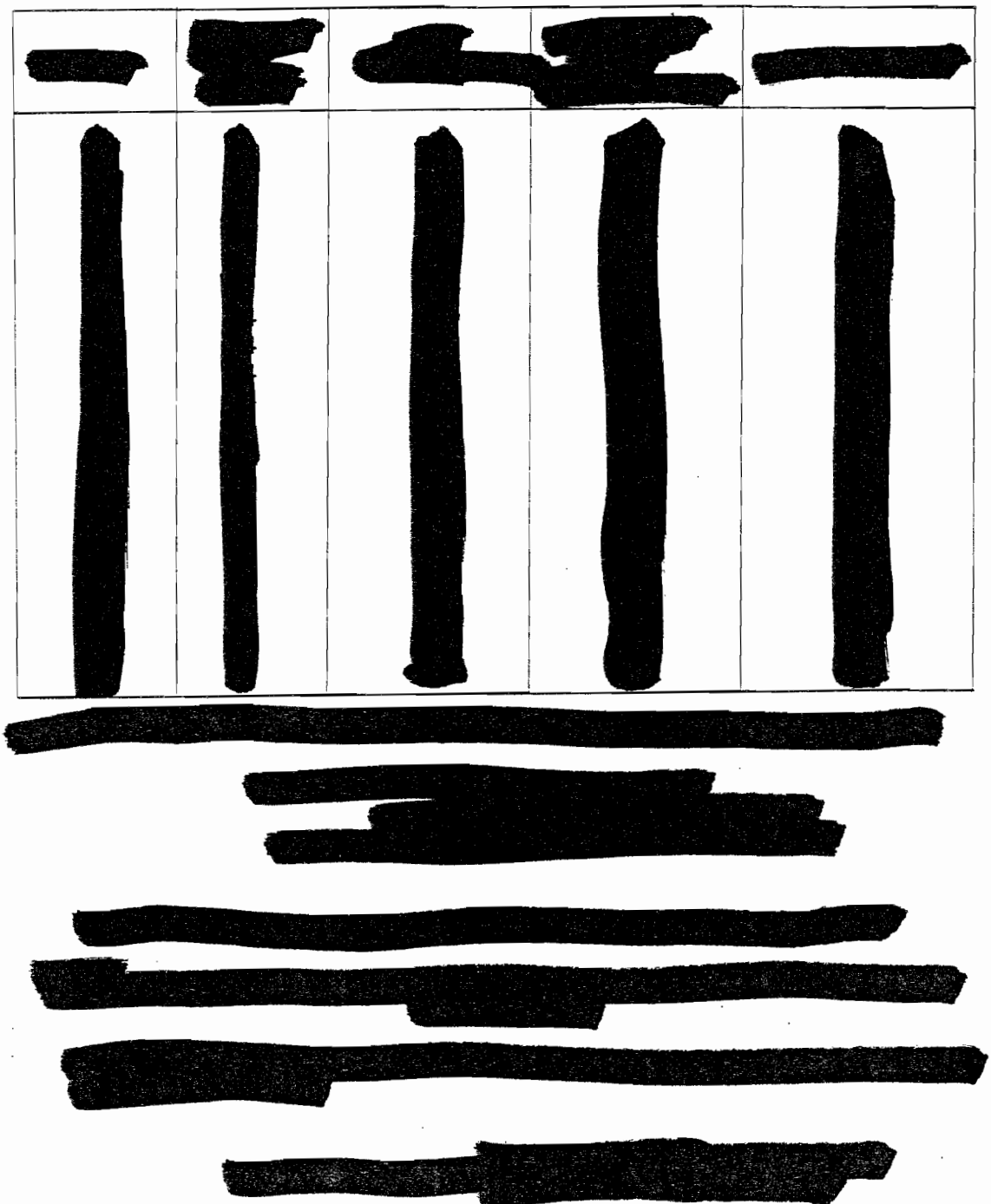
(U) The amount of agent actually administered may be quantitated more precisely by expressing it as the retained dose, which is the total quantity of agent retained by the individual, expressed as micrograms of agent per kilogram of body weight. Retained dose was calculated according to the following formula:

$$\text{Retained dose} = \frac{(\text{agent conc. mg/cu m}) \times (\text{expired vol., cu m}) - (\text{expired agent, mg})}{\text{body weight, kg}}$$

Table III (U). Inhalation Data for Volunteers Exposed to EA 3528

Volunteer	Weight kg	Exposure Time sec	Agent Concentration mg/cu m	Minute Volume l	Volume Air Expired l/min
[redacted]	62.7	83	9.7	10.5	14.5
	71.3	114	10.0	8.7	16.5
	89.0	59	18.3	12.1	11.9
	75.9	100	9.9	11.5	19.1
	70.9	60	14.1	13.2	13.2
	66.7	95	17.1	6.8	10.8
	90.5	148	11.0	10.6	26.0
	82.2	65	18.4	13.2	14.3
	60.4	86	12.9	10.4	14.9
	67.7	87	12.9	11.7	17.0
	80.4	121	15.3	9.8	19.6
	72.3	118	12.5	11.2	21.9
	68.6	55	16.9	17.1	15.7
	87.7	96	13.7	17.2	27.5
	72.7	82	13.2	17.5	23.8
	82.2	228	10.0	12.2	46.5
	82.7	150	13.7	14.3	35.7
	67.7	105	13.7	17.1	29.9
	76.8	112	13.7	18.6	34.6
	81.4	120	17.8	18.3	36.6

Table IV (C). Relationship Between Retained Dose, Ct, and Incapacitation when Volunteers are Exposed to EA 3528 (U)



[c](U) Incapacitation is defined as two or more sucessive NF scores below 10 percent of baseline.

For these 20 men, the quantity of expired agent was not sufficient to significantly affect the calculation of retained dose, so the retained dose was calculated as 100 percent. With small particle aerosols, the dose retained is significantly less than 100 percent, and the quantity of expired agent thus represents a significant fraction of the total dose delivered. In the present study, retained dose, expressed as the free base equivalent, ranged from 1.7 μg/kg to 6.0 μg/kg. A probit analysis of retained dose versus incapacitation yields an ID50 of 2.3 μg/kg with 95 percent confidence limits of 1.4 to 3.7 μg/kg.

(U) The relation between retained dose and Ct is plotted in figure 3. A correlation coefficient (r) of 0.71 ($p < 0.01$) was obtained between these two measures and reflects in part the technical errors inherent in the present methodology. When retained dose is plotted against corrected Ct (figure 4), a nearly perfect correspondence is seen, with a correlation coefficient (r)* of 0.99 ($p < 0.001$). The reason for this high correlation becomes apparent when the various factors constituting retained dose and corrected Ct are analyzed. If those variables that are contained in both the corrected Ct and retained dose are evaluated, the graph becomes a plot of air breathed (reflected in the retained dose) against the actual time spent breathing (reflected in the Ct). The graph thus illustrates that the present system of breathing-control produces a remarkably constant volume of air breathed per unit time for the same individual. Doubling the breathing time essentially will double the volume of air breathed.

B. (U) Plasma Levels of Agent.

Table V presents the concentration of agent in plasma, expressed as nanograms (millimicrograms, 10^{-9} gm) per milliliter of plasma (ng/ml) at the indicated experimental times, for 16 of the 20 men. Technical difficulties prevented accurate measurement of agent plasma level for the remaining four men (DO, ZN, AM, BU). Table V shows the significant blood levels of agent achieved within a short time after initiation of exposure.

Median plasma concentrations of agent for different experimental times and the corresponding NF scores are graphically depicted in figure 5. There was a rapid rise in agent plasma level within minutes after exposure. The agent plasma level peaked at 30 minutes to 1 hour, and then gradually declined. The rise in agent plasma level was associated with a rapid decrement in NF performance. Some severely affected individuals scored below 10 percent on the first NF test performed 15 minutes after exposure. An NF score of 10 percent or below generally reflected an agent plasma concentration of 4 to 6 ng/ml or greater. NF scores tended to rise rapidly after the plasma concentration had fallen below 4.0 ng/ml, and NF scores of those individuals who received a dose near the ID50 had returned to about 75 percent of baseline in 8 to 12 hours. By 24 hours scores for all subjects were at or above baseline levels. The half time ($T_{1/2}$) for disappearance of this agent from plasma, as determined from the linear decrease in plasma concentrations from 2 to 6 hours after exposure, was 200 minutes. This value agrees with a previous estimate of 170 minutes for the plasma $T_{1/2}$ after intravenous administration of the agent.[4]

*All correlations reported in this study are Pearson product-moment correlation coefficients.

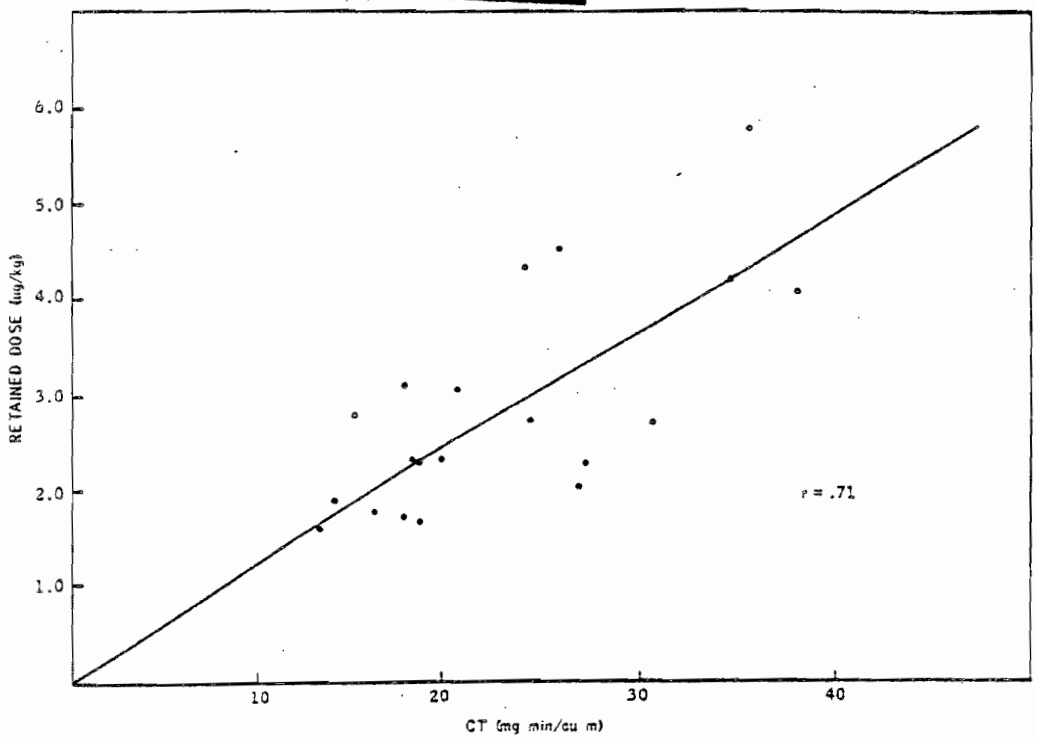


Figure 3 (U). Retained Dose of EA 3528 Vs. Ct

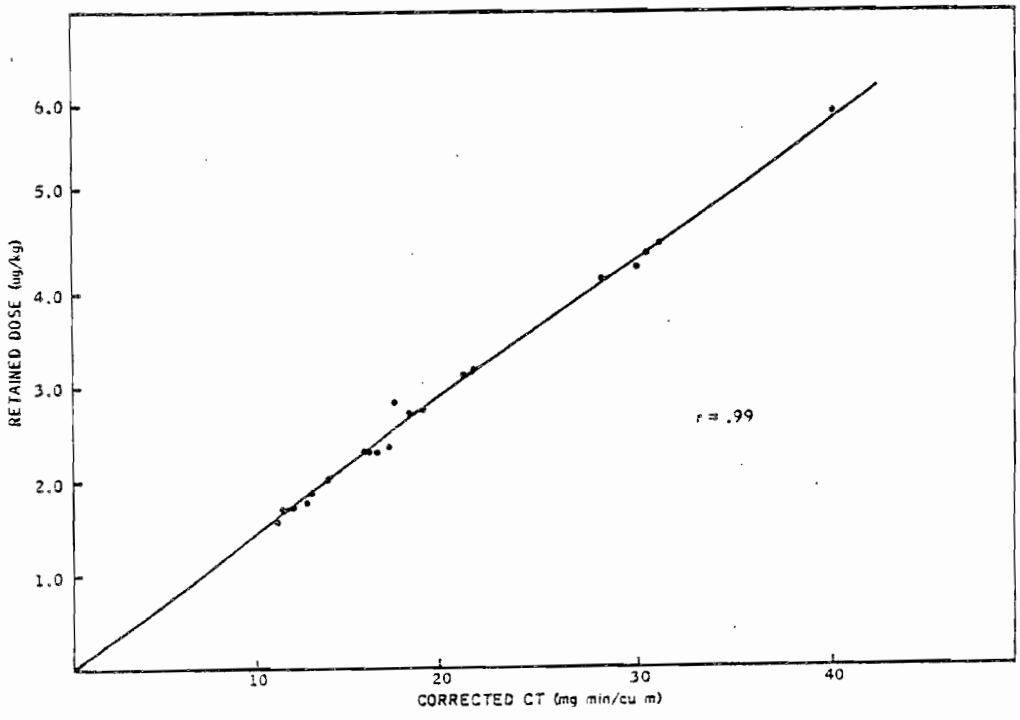


Figure 4 (U). Retained Dose of EA 3528 Vs. Corrected Ct

Table V (U). Plasma Levels of EA 3528

Name	Retained Dose µg/kg	Plasma Levels ng/ml Time						
		0005	0015	0030	0100	0200	0300	0600
[redacted]	1.7	3.1	7.8	5.9	7.1	4.4	4.0	1.6
	1.7	2.0	4.8	4.2	2.9	2.7	2.7	0.5
	1.8	1.5	2.7	3.6	2.8	1.7	2.1	---
	1.9	6.3	10.0	9.5	8.3	7.6	6.1	3.6
	2.0	5.3	5.6	5.9	4.5	4.4	4.0	2.2
	2.1	2.9	5.3	6.0	6.2	5.2	4.4	1.8
	2.4	---	---	---	---	---	---	---
	2.4	1.4	4.1	4.5	3.5	2.7	2.9	1.5
	2.4	3.9	7.9	5.9	5.9	4.9	3.9	1.5
	2.4	3.1	5.8	6.6	5.3	4.7	3.2	2.2
	2.8	1.3	4.8	5.0	5.0	3.8	2.5	1.1
	2.8	---	---	---	---	---	---	---
	2.9	2.1	4.1	3.5	3.7	3.4	2.6	0.9
	3.2	---	---	---	---	---	---	---
	3.2	---	---	---	---	---	---	---
	4.2	9.4	12.5	12.3	11.9	9.0	8.7	2.7
	4.4	4.4	11.1	15.7	9.9	7.6	6.5	3.4
	4.5	3.1	11.2	10.2	7.6	7.3	5.7	4.2
	4.6	---	11.2	12.0	10.3	---	6.0	3.1
	6.0	7.4	8.2	20.9	18.9	16.0	12.3	6.2

Figure 5 (C). Median Plasma Concentrations of EA 3528 and NF Scores (U)

C. (C) [redacted]

(U) No consistent changes were seen in axillary temperature or respiratory rate. During periods of marked anxiety as a consequence of drug effects, a transitory rise in respiratory rate was observed. One volunteer who received this agent in a prior study showed hyperventilation with carpopedal spasm. Pulse rate of the subjects showed no consistent changes except when associated with overt signs of anxiety. Mild increases in both systolic and diastolic blood pressure (supine) persisted for up to 8 hours after drug administration. Enlargement of pupil diameter (mydriasis), up to 2.5 millimeters above baseline, generally appeared within 15 to 30 minutes, reached a maximum by 1 to 3 hours, and returned to baseline levels within 12 to 16 hours. Blurring of near vision, commonly seen with glycolate compounds, did not occur. The duration of mydrasis appeared to parallel the disturbances in central nervous system function and behavior produced by the drug.

(C) [redacted]

D. (C) [redacted]

(C) [redacted]

(U) The subjects in this study received higher doses than those in previous studies. The higher dose levels produced some qualitative and quantitative differences in performance decrement and in observed behavior. Of interest was an increased incidence of hostile and aggressive behavior and perhaps a higher incidence of anxiety. Three of 12 men who received an incapacitating dose displayed contrary behavior directed either at the physical environment or, in one instance, toward the personnel in attendence. An analysis of the nurses' notes

Figure 6 (C). Laboratory Physiological and Performance Measures on a Single Volunteer (WA) After EA 3528 (U)

revealed that approximately 58% (7 out of 12) of those volunteers who were incapacitated by NF criteria showed what might be considered evidence of latent or overt contrary behavior. No difference between contrary and noncontrary men could be determined on subsequent review of psychiatric screening interviews and personality inventories. The small sample in this study does not permit adequate statistical comparison, but this problem will be reviewed more thoroughly in a separate report.* Table VI shows the quotations from the nurses' notes which are considered evidence of adverse behavior.

(U) A second behavioral observation concerned two men who were incapacitated by the drug and displayed unusual symptoms lasting up to 2 weeks after drug administration. One man [redacted] complained of increased nervousness and frequent daydreams in the week following exposure. These symptoms were alleged to be responsible for a marked increase in alcohol consumption. Detailed postexposure psychiatric evaluation suggested the drug experience was traumatic for this volunteer and may have related to loss of control of coordinated thinking experience while under agent effect. A second volunteer (CA) exhibited unexplained symptoms of mild depression and social withdrawal for 1 to 2 weeks after exposure. Review of the screening psychiatric interviews and personality interviews of both these volunteers failed to suggest any specific difference from the other volunteers.

E. (U) Laboratory Measures.

All control laboratory measurements were within normal limits. Repeated laboratory tests 24 hours and 7 days after examination were within normal limits except for an elevated BUN of 28 mg/100 ml for one volunteer [redacted] who had a control level of 20 mg/100 ml. Repeat determinations of the BUN at 10 and 14 days and creatinine clearance and urinalysis between days 7 and 14 were normal on OL.

F. (U) Additional Examinations.

Electroencephalograms taken on four men during peak drug effects showed generalized increased high-frequency low-voltage discharge.

G. (U) Statistical Considerations.

The three major parameters studied in the 20 volunteers (plasma level was available in 16 volunteers) were the retained dose of the agent, the consequent plasma level of agent resulting from absorption of the agent retained by the respiratory mucosa, and the ability of that dose to alter NF performance. The relationship between these three parameters is presented in table VII with a separate statistical analysis presented for each unit of time in which these three parameters were simultaneously tested. The correlation coefficient and five best-fit equations with their applicability are shown. The correlation coefficients between the retained dose of agent and the resultant highest agent plasma level, and between retained dose and resultant lowest NF score (values are independent of time) are, respectively, 0.86 ($p<0.01$) and 0.56 ($p<0.05$). The correlation coefficient between the highest agent plasma level in each volunteer and the resultant lowest NF score, independent of time, is 0.60 ($p<0.05$).

*(U)Safer, D. J., CPT, MC, and Allen, R. P., CPT, MSC. Unpublished data.

Table VI (U). Examples of Contrary Behavior

[redacted]	Retained Dose µg/kg	Time	Comments
[redacted]	2.8	0200	Complained of severe pain on each drawing of blood specimen. Only 6 cc obtained because subject moved arm away; refused to let doctor draw a second specimen.
[redacted]	2.0	0200	Subject stated that he wanted to discontinue EEG; he stated that he and the nurse would be in a big fight before the day was over and that when the test was over he was going to hate the nurse.
		0215	Subject refused to continue EEG.
		0230	Subject refused to eat lunch.
		0300	Stated that he wanted people to stop asking him questions and that he wanted to stop being tested.
		0330	Subject quite agitated and becoming hostile; stated that he wanted to stop the test.
		0345	He stated that the reason he wanted to stop the test was he felt like he wanted to hurt the nurse.
[redacted]	4.6	0200	Subject stated that he felt like hitting the nurse; said to the nurse "I don't want to hurt you but I want to hit you." Kept hitting hand with fist, raised fist as though to hit nurse. Nurse left area, subject went from window to window looking at nurse laughing and hitting hand with fist, attempted to leave area.
		0220	Banged on windows with fist, laughed as he did so.
		0235	Subject attempted to grab another nurse, started banging on window when he saw a nurse pass.
		0250	Tore ceiling down.
		0305	Treated with thorazine.

Table VI (U). Continued

[redacted]	Retained Dose µg/kg	Time	Comments
[redacted]	4.4	0005	Upon return to padded area, subject resisted aidman's attempts to dress him in pajamas, he began screaming.
		0015	Subject screamed loudly, "They're killing me."
		0100	Refused to do NF test.
[redacted]	4.5	0100	Refused to return to testing booth.
		0200	Refused to do NF test.
		0312	Attempted to do NF test but wouldnot go back to testing booth, threw clipboard on the floor when asked to continue with the test and said that he was sorry.
		0400	Said he feels like he has been liquidated.
[redacted]	3.2	0015	He flatly refused to do NF and pegboard tests.
		0030	Still refused to do NF test.
		0100	Subject was extremely restless, hitting the walls.
		0110	Treated with thorazine.
		0200	Physically assaulted and struck the physician.
		0240	Did not want to go to sleep because he felt it might bring out his violent tendencies.
[redacted]	6.0	0030	Very difficult to test, he almost refused to attempt test, but in a humorous manner; threw pencil across the room each time it was given to him to continue. He continued in a hilarious state of laughter.

Table VII (C). Intercorrelations and Parameters for Retained Dose, Plasma Level, and Number Facility Scores After EA 3528 (U)

Table VII (C). Continued

[redacted]

IV. (C) [redacted]

(C) [redacted]

(U) The site of deposition of the particulate aerosol within the respiratory tract cannot be determined from this study. Theoretical considerations and prior research on aerosol deposition[3] indicate that a 12-micron MMD particulate aerosol would be completely deposited and retained almost totally within the upper respiratory tree, including the mucous membranes of the mouth, oropharynx, trachea, and larger bronchi.

(U) As expected, retention of agent by the 20 volunteers in this study was essentially complete. Of special interest is the rapidity and apparent completeness of absorption of the agent as measured by its appearance in the plasma. The peak median plasma concentration occurred at 15 minutes, indicating that absorption is quite rapid. Whether subsequent appearance of agent in plasma represents absorption through the mucous membranes of the mouth and upper respiratory tract and/or through the mucosa of the gastrointestinal tract after swallowing cannot be determined from the data at hand.

(U) The physiological, behavioral, and performance aiterations produced by the large particles are similar to those seen when the drug is given as a smaller-sized particle or by the oral or intravenous routes. The qualitative and quantitative differences seen appeared to be a function of higher dose rather than of the route of administration or aerosol characteristics.

V. (C) [redacted]

(C) [redacted]

1. (C) [redacted]

2. (U) The inhalation ID50 (retained dose) of the 12-micron particle is equivalent in size to the incapacitating intravenous or oral dose of EA 3528; the ICt50 is one-third as large as the incapacitating dose of an 0.8-micron (MMD) particle aerosol.

3. (U) Retention of the 12-micron (MMD) particulate aerosol by the respiratory passages is essentially complete.

4. (U) Significant agent plasma levels are achieved within 15 minutes when the route of administration of agent is inhalation and the absorptive surfaces are the respiratory passages.

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(U) APPENDIX

The following are the case numbers of all subjects used in this study:

[redacted]– 1959
[redacted]– 1956
[redacted]– 1929
[redacted]– 1957
[redacted]– 1926
[redacted]– 1927
[redacted]– 1947
[redacted]– 1928
[redacted]– 1933
[redacted]– 1932
[redacted]– 1934
[redacted]– 1949
[redacted]– 1935
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