



GENERAL PROCEDURES OF ONE-YEAR INHALATION STUDIES - continued

RENAL CLEARANCE (Dr. Brodie)

4 dogs - two O1 levels each  
 2 " - 12, O4, 1 level each

Material	Level	Number Dog	Experiment	Interval
	mg/m <sup>3</sup>			
O1 (Roberts)	0.15	2 2	Exteriorized Bladder Catheterized	
O1 (Weil)	0.5(?)	4	Catheterized	
12 (Spiegel)	0.5	2	Catheterized	
O4 (Rothstein)	2.0	2	Exteriorized Bladder	

Require 8 female dogs, same size, prepared for catheterization. Dr. Rekers to operate.

Determination	Interval
Urinary Catalase	Semiweekly mo. 1, 2; 5 days/ mo. no. 3-13 in catheterized dogs
" Protein	" " "
" A.A.N.	" " "
Clearances	Monthly
Blood N.P.N.	Monthly

LIVER-FUNCTION TESTS - Dr. Field

Prothrombin = P  
 Fibrinogen = F  
 Bronsulfalein = B

5 dogs - not hematology animals  
 10 rats - separate groups from all others

Material	Unit	Level	Test	Interval
Control	10	—	P,F,B	Before; week 1, 4, and monthly
O1	3	High	P,F,B	" 1, mo. 1; week 1,2,4, mo. 2; monthly
O1	5	Low	P,F,B	" " " "
O7	12	High	P,F,B	" " " "
12	7	Low	P,F,B	" " " "

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GENERAL PROCEDURES OF ONE-YEAR INHALATION STUDIES - continued

CHEMISTRY - PLAN #1

BLOOD

NPN - 10 dogs - Hematology Group - Semimonthly, month 1; weekly, month 2; monthly, month 3-13.

2-4 dogs - Special Clearance Group - Monthly.

Liver-Function Tests - Prothrombin } 5 dogs - Non-Hematology Group.  
Fibrinogen } 10 rats - Special Group.  
Bromsulfalein } Initial; 1, month 1; week 1, 2, 4,  
month 2; monthly, month 3-13.  
(See memorandum 5-30-45).

URINE

Protein - 10 dogs - Hematology Group - Semiweekly, month 1, 2; 5 days/mo. month 3-13.

2-4 dogs - Renal Clearance Group - (Specially prepared by exteriorized bladder or vaginal operation).

Catalase - 2-4 dogs - Renal Clearance Group - (Specially prepared by exteriorized bladder or vaginal operation).  
Semiweekly, month 1, 2; 5 days/mo. month 3-13.  
(See memorandum 5-30-45).

Amino Acid Nitrogen: The same animals and schedule as for catalase.  
(Renal Clearance Group)

Renal Clearances - 2-4 dogs

Amino Acid Nitrogen  
Diodrast  
Inulin

Sugar - qualitative as requested.

CHEMISTRY - PLAN #2

BLOOD

NPN - 10 dogs - (Hematology Group, if done) - Semimonthly, month 1, weekly, month 2; monthly, month 3-13.

URINE

Protein - 10 dogs - Semiweekly, month 1, 2; 5 days/month 3-13.

Sugar - qualitative as requested.

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GENERAL PROCEDURES OF ONE-YEAR INHALATION STUDIES - continued

T-ANALYSIS - PLAN #1 (Dr. Steadman)

DOG

Lung, liver, kidney, bone (and carcass), 64 serial, 800 terminal - sacrificed according to Pathology Schedule

2 - RATS

Lung, liver, kidney; carcass; 2600 serial, 800 terminal - sacrificed according to Pathology Schedule. (Facilities required for storage of samples)

RABBIT AND GUINEA PIG

Kidney and bone

Analyses of tissues of all dying animals

T-ANALYSIS - PLAN #2 (Dr. Neuman)

1 DOG

At 4, 6, 8, 12 month. Lung, kidney, liver femoral epiphyses, 48 analyses Pathology-scheduled animals

1 RAT

At 1, 2, 3, 5, 7, 14, 30, 90, 150, 210, 270, 330 and tissues as above. Pathology-scheduled animals

T-ANALYSIS - PLAN #3 (Dr. Neuman)

1 DOG

As in Plan #1 - 160 analyses

1 RAT

As in Plan #2 with exception of days 1, 2, 3.

Signed

H. E. Stokinger  
H. E. Stokinger

May 24, 1945  
Revised June 1, 1945

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CAPACITY AND PROCEDURES OF THE SERVICE DEPARTMENTS FOR THE  
 INHALATION SECTION FOR THE PERIOD  
 JUNE 1, 1945 TO JULY 1, 1946

CAPACITY OF HEMATOLOGY SECTION

Capacity of 25 counts to be taken semi-monthly on 10 levels for 13 months. In addition to this we can arrange for one acute run at a time, blood counts for which can be taken at weekly intervals. The counts on all runs are to be done on 10 dogs and 15 rats. It is also understood that should any bleeding for other purpose be done on either dogs or rats, that the amount of blood drawn and the frequency of this bleeding shall be the same for all chronic runs. These runs are to start on consecutive week-days Monday through Friday as: 1st, Monday; second, Tuesday; third, Wednesday; fourth, Thursday; fifth, Friday. The next Monday, sixth run; Tuesday, seventh run; Wednesday, eighth run; Thursday, ninth run; Friday, tenth run.

Signed

*George M. Suter*  
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 George M. Suter

CAPACITY OF ROUTINE CHEMICAL ANALYSIS LABORATORY

The maximal daily capacity of this laboratory, for short periods of time, is indicated in Table I.

TABLE I

Maximal Daily Capacity of Routine Chemical Analysis Laboratory

Alternative Daily Schedules  
 (either schedule 1 or 2, etc. but not 1 and 2, etc.)

Schedule	U R I N E					B L O O D	
	Amino Acid		Protein	Sugar	Chloride	NPN	Urea
	Rabbit	Human					
1	20	20	200	0	0	60	0
2	30	10	150	50	0	40	or 30
3	40	0	100	100	0	as above	
4	10	10	100	100	30		
5	15	5	100	100	30		

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This capacity could only be maintained for periods of ten days to two weeks, with intervals in between for recuperation.

Totals of 30 ureas, 200 proteins, 60 NPNs, and 40 urinary amino nitrogen determinations are the absolute maximum. Alternative schedules other than those listed may be possible, as long as the above maximums are not exceeded.

The average monthly capacity of the laboratory is listed in Table II.

TABLE II

Optimal Monthly Capacity of Routine Chemical Analysis Laboratory

Determination	Number
Urinary Amino Nitrogen	200
" Protein	2000
" Sugar	2000
" Chlorides	600
Blood NPN	1200
or NPN	800
Urea N	600

Signed Frank A. Smith  
Frank A. Smith

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PROCEDURE OF TISSUE ANALYSES BY MR. ORCUTT'S GROUP  
under direction of Dr. W. Neuman

Controls will be run to ascertain whether tissues from animals which have been dissected by the Pathology Department are or can be made free from contamination. If contamination is not a serious problem, the following plan will be followed:

PLAN #1 (for the F6 and nitrate)

1 dog each, sacrificed at 4, 6, 8, and 12 months of exposure.

3 rats each, sacrificed at 1, 2, 3, 5, 7, 14, 30, 90, 150, 210, 270, 330 days of exposure.

PLAN #2 (for all others)

1 dog each sacrificed at 4, 6, 8, and 12 months of exposure.

3 rats each sacrificed at 3, 5, 7, 14, 30, 90, 150, 210, 270, 330 days of exposure.

In every case, lung, liver, kidney and femoral epiphyses will be analyzed.

Total numbers:

dogs	no. of tissues	no. of animals	no. of runs	TOTAL
	4	x 4	x 13	208 analyses
rats	4	x 36	x 2	
	4	x 30	x 11	<u>1608</u> analyses
				1816

Note: The whole thing hinges on whether or not the tissues we can get are contamination free. Wentman says analyses are not wanted if you do not histology on the same tissues. If this is the case, and we can't eliminate contamination, there will be no analyses required.

Signed

W. Neuman  
W. Neuman

PATHOLOGY PLAN I

Dog: Serial: 10 days; 4, 6, and 8 months.

Final: 15 dogs.

Biopsy: 2 dogs initial; 10 days, 4 and 8 months.

- Procedures:
1. Complete tissues on all dogs without brain.
  2. Biopsies on low levels to be initial and 10 days only.
  3. Bone marrow smears on terminal dogs.

Rat: Serial: 1, 2, 3, 5, 7, 14, 21, and 30 days, and every 2 weeks from there on.  
(\*Extra 3 animals for these times.)

Final: 50 rats.

- Procedures:
1. 3 rats at each designated time interval.
  2. Lung, liver, and kidney on all rats. Complete tissues are saved.
  3. Complete tissues on 25 animals at the end.
  4. Bone marrow smears on terminal 50.

PATHOLOGY PLAN II

Dog: Serial: 10 days; 4, 6, and 8 months.

Final: 15 dogs.

Biopsy: 2 dogs initial 10 days, 4 and 8 months.

- Procedures:
1. Complete tissues all dogs, without brain.
  2. Biopsies at low levels to be initial and 10 days only.

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Rat: Serial: 3, 5\*, 7\*, 14, 21, 30, 60, 90, 120, 150, 180, 210, 240, 270, 300, and 330 days. (\*3 additional animals at these points.)

Final: 50 rats.

- Procedures:
1. 3 rats at each designated time interval.
  2. Lung, liver, and kidney on all rats. Complete tissues are saved.
  3. Complete tissues on 25 animals at end.

Lung, liver, and kidney will be done on any and all dying animals for each chronic run. In case of a severe epidemic with many deaths, this could not be done.

It is suggested that, if there are animals not scheduled for pathology upon which the experimenter would particularly like to have all of the tissues, he could do his own autopsy and save the tissue in 10% formalin. The reading of such tissues might be done if time ever developed.

#### SPECIAL PATHOLOGY PROCEDURES

A. Dr. Roberts: Nitrate; high and low levels (Plan I).

1. T stains (subject to change as the method develops).
  - a. Kidneys of biopsy dogs as well as serial and terminal dogs.
  - b. All tissues of serial sacrifice dogs and three terminal dogs.
  - c. One rat from each serial date except the first seven days (kidney).
  - d. Three rats from the first seven days (kidney).
  - e. Save vertebra and femur from one rat at each serial sacrifice. Vertebra and femur of all dogs saved. These bones may be used for T stains if we are able to develop a method.
2. Phosphatase stains.
  - a. Kidneys of biopsy dogs as well as serial and terminal dogs.
  - b. Kidney of 3 rats for each of the sacrifice days during the 1st week.
  - c. Kidney of one rat at each sacrifice day after the first week.
  - d. Kidney of 10 terminal sacrifice rats.
  - e. Kidneys of all rabbits.
3. Nervous System.
  - a. Brain and peripheral nerves will be saved on 2 terminal rats at each level for study.
  - b. Brain and peripheral nerves will be saved for study on one terminal dog at each level.

~~SECRET~~PATHOLOGY PLAN4. Additional Animals.

- a. Rabbits: Lung and kidney of all rabbits.
- b. Guinea Pigs: Lung and kidney of all guinea pigs.
- c. Adult rats: Serial sacrifice for 6 months to have lung, liver, and kidney. Three rats at each sacrifice schedule (weekly for one month and semi-monthly thereafter).

## 5. Bone Marrow Smears:

- a. To be done on all terminal rats and dogs.

- B. Paul Dygert: Follow Plan II for both runs (TO<sub>2</sub>).
- C. Carrol Weil: Plan II (Nitrate). No meeting as yet.
- D. Charles Spiegl: Plan I (TF<sub>6</sub>). No meeting as yet.
- E. TCl<sub>4</sub>: Plan II. No meeting as yet.
- F. TF<sub>4</sub>: Plan II. No meeting as yet.
- G. Control: Plan I (altered).

Dogs - terminal (all tissues of dogs).

Dogs - 2 biopsy dogs, initial 10 days, 4 and 8 months.

Rats - 3 animals per week for one month; semi-monthly thereafter.  
50 terminal rats.

Tissues: lung, liver, and kidney.

Bone marrow smears on terminal animals.

RGM:leg

  
Roger G. Metcalf, Capt., M.C.  
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