See Dow storage # 1193 1701 alley for more info

CHEMICAL BIOLOGY RESEARCH — DOW CHEMICAL U.S.A

FILE VB T34.19-DD54-1

SUBMITTED BY	CHARGE	DATE	K NUMBER
F. D. Axe		4/17/72	DD-54

RECEIVED OCT 06 1988 J. M. GOOTEE

RAT AND HAMSTER INTRATRACHEAL AND INTRAPERITONEAL INJECTION-STUDIES ON TITANIUM PHOSPHATE AND ASBESTOS

REPORTED BY J. M. Norris - G. L. Sparschu CHECKED BY P. J. Gehring

Titanium phosphate ($Ti_3(PO_4)_4$) fibers are being considered as a replacement for asbestos in commercial applications such as brake linings and construct. materials. A study employing the Sherman rat and the Golden hamster, herein reported, was conducted on a sample of titanium phosphate, prepared at the Western Division, to determine whether the fibers would produce a fibrogenic response or compound-induced neoplasms when introduced into the lung. To assure deposition of known quantities of the titanium phosphat the material was administered to the animals by intratracheal injection. Subsequently, an intraperitoneal injection study, using rats and hamsters, was conducted to assess and compare the effects from the two routes of administration.

Crude chrysotile asbestos was used as a positive control in all phases of the investigation.

Titanium phosphate, under the experimental conditions of the study herein reported, produced a granulomatous foreign body response of a non-fibrogenic nature in intratracheally and intraperitoneally treated rats and hamsters. No compound-induced neoplasms were observed in the animals so treated.

The pulmonary response to the titanium phosphate in the intratracheally treated animals consisted of:

(1) A moderate macrophage response
(2) Multifocal inflammatory reaction in the rat and a diffuse inflammatory reaction in the hamster
(3) sequestration of the fibers by atelectasis
(4) focal obliteration of alveolar structure in the animals receiving the top dose and preservation of the alveolar structure in the animals receiving the lower dose.
(5) slow clearance of the fibers from the lung.

ST0000100

The granulomatous response in the peritoneal cavity of the rats and hamsters was accompanied by minimal collagen deposition.

In contrast to these responses, asbestos produced mesotheliomas in the intraperitoneally treated rats and a fibrogenic response in intratracheally treated rats and intraperitoneally treated hamsters.

The fibrogenic response to asbestos was characterized by (1) a reticulin stromal reaction followed by collagen deposition, (2) sequestration by scar tissue, (3) abundant macrophage response, and, in the lung,

(OVER)

DISTRIBUTION

D. W. Richards, Larkin Lab

J. E. Johnson, H. H. McIntyre, C. E. Kimmel, L. K. Freyel, D. D. McCollister (2), F. D. Axe, B. E. Burgert, H. L. Gordon, MD, E. H. Blair, A. J. Schwarz, MD, S. M. MacCutcheon, D. Kilian, MD, B. Horvath, H. Edwards, R. J. Shaver, V. B. Robinson, B. Holder, MD, L. Silverstein, L. Pitchforth, C. A. Goring

Western Division
R. H. Bailes
M. D. Yeaman
J. P. Surls
R. S. Long
H. D. Ledbetter

PLAINTIFF'S EXHIBIT

(4) destruction of alveolar structure, and (5) slow clearance.

In contract to the responses in the rat lung, asbestos in the hamster lung produced a mononuclear inflammatory reaction without collagen deposition.

Chemical Biology Research

DOW CHEMICAL U.S.A.

Midland, Michigan

RAT AND HAMSTER INTRATRACHEAL AND INTRAPERITONEAL INJECTION STUDIES ON TITANIUM PHOSPHATE AND ASBESTOS

by

J. M. Norris and G. L. Sparschu

April 1972

TABLE OF CONTENTS

INTRODUCTION

Titanium phosphate ($Ti_3(PO_4)_4$) fibers are being considered as a replacement for asbestos in commercial applications such as brake linings and construction materials. A study employing the Sherman rat and the Golden hamster, herein reported, was conducte on a sample of titanium phosphate, prepared at the Western Division, to determine whether the fibers would produce a fibrogenic response or compound-induced neoplasms when introduced into the lung. To assure deposition of known quantities of the titanium phosphate, the material was administered to the animals by intratracheal injection. Subsequently, an intraperitoneal injection study, using rats and hamsters, was conducted to assess and compare the effects from the two routes of administration.

Crude chrysotile asbestos was used as a positive control in all phases of the investigation.

SUMMARY OF RESULTS AND DISCUSSION OF THE INTRATRACHEAL AND INTRAPERITONEAL INJECTION STUDIES

Titanium phosphate, under the experimental conditions of the study herein reported, produced a granulomatous foreign body response of a non-fibrogenic nature in intratracheally and intraperitoneally treated rats and hamsters. No compound-induced neoplasms were observed in the animals so treated.

The pulmonary response to the titanium phosphate in the intratracheally treated animals consisted of:

(1) A moderate macrophage response
(2) Multifocal inflammatory reaction in the rat and a diffuse inflammatory reaction in the hamster
(3) sequestration of the fibers by atelectasis
(4) focal obliteration of alveolar structure in the animals receivin the top dose and preservation of the alveolar structure in the animals receiving the lower dose.
(5) slow clearance of the fibers from the lung.

The granulomatous response in the peritoneal cavity of the rats and hamsters was accompanied by minimal collagen deposition.

In contrast to these responses, asbestos produced mesotheliomas in the intraperitoneally treated rats and a fibrogenic response in intratracheally treated rats and intraperitoneally treated hamsters. Observation of mesotheliomas in the peritoneal cavity of rats has been reported by Wagner.[1]

The fibrogenic response to asbestos was characterized by (1) a reticulin stromal reaction followed by collagen deposition, (2) sequestration by scar tissue, (3) abundant macrophage response, and, in the lung, (4) destruction of alveolar structure, and (5) slow clearance.

In contrast to the responses in the rat lung, asbestos in the hamster lung produced a mononuclear inflammatory reaction without collagen deposition. Gross and deTreville noted these pulmonary observations with asbestos and ascribed them to a less effective pulmonary clearance mechanism and greater reactivity of the pulmonary tissue in the hamster than in the rat.[2] Furthermore, the results obtained on the intratracheally treated rats and hamsters in the study herein reported are the same as reported by Gross et al.[2]

In discussing the possible mechanisms of carcinogenic action of asbestos, Harington and Roe have associated the activity with the presence of (1) natural oils found on asbestos fibers and (2) metal complexes in the asbestos. For this reason, the asbestos used in the present study was crude and non-acid washed. The iron and iron complexes in mined asbestos are suspect carcinogenic agents. Hematite, which contains 70% of ferric iron, has been suspected of causing lung cancer in man. Harington and Roe also suggest that the ratio of ferric to ferrous iron may be of significance, in that a low $Fe^{+++}:Fe^{++}$ may be associated with carcinogenicity. The sample used in the present study was, therefore, analyzed for various trace elements including iron compound and the ratio of iron in the ferric and ferrous states. The ratio in the case of each of the three iron compounds found in the asbestos was high ferric to ferrous. The asbestos, however, did produce a fibrogenic response in the peritoneal cavity of the rats and hamsters and in the lungs of the rats, and diffuse abdominal mesotheliomas in the rats. The association of low $Fe^{+++}:Fe^{++}$ ratio with carcinogenicity, therefore, appears questionable.

MATERIALS

The titanium phosphate sample used in these investigations was identified by reference #GP12-66-67 and batch #30. The fibers which measured 10-20μ by 0.2-0.3μ are acidic, insoluble in water at room temperature and have a density of 2.76 + 0.02 (Midland Analytical Laboratories report AL579-476). Trace elements in the sample were identified by emission spectroscopy (Spectrographic report 195058):

Aluminum	0.07%
Copper	<0.005
Iron	<0.001
Magnesium	<0.1
Manganese	Present but <0.01%
Nickel	<0.005
Silicon	0.03
Titanium	>6.0
Vanadium	<0.01

The sample of chrysotile asbestos used in these investigations was identified as "Hooker #1" non-acid washed asbestos that was mined in Morrisville, Vermont by the G.A.F. Corporation. An aqueous slurry of the asbestos was prepared and, with the use

of a Waring blender, the asbestos was ground to a fiber length of less than 100μ. The water was then separated from the asbestos by suction aspiration through #40 Whatman filter. The asbestos was washed three times with 100% ethanol, dried by washing with ether, and ground through a #40 Tyler mesh screen. The ground asbestos fibers were morphologically characterized by electron microscopy (Midland Analytical Laboratories report, AlS86-352). The fibers measured 150-400Å by 500Å-70μ and were commonly found in bundles. The sample contained electron dense particles, measuring from 0.1μ to 10μ, which could not be identified from morphological characteristics.

Analysis of the sample for iron compound(s) content revealed three such compounds, one of which was magnetite (Fe_3O_4), one an unidentified ferric compound and the remaining one, an unidentified ferrous compound (Letter Report File #68115-01, Radiochemistry Research Laboratory). The relative amounts of the iron compounds were A/B/C = (1.0±0.1)/(2.01±0.2)/(7±1); where A = unidentified ferrous, B = unidentified ferric and C = magnetite. The ratio of the Fe^{++} to Fe^{++} was 2.1±0.3 in the magnetite and 1.8±0.2 in the two unidentified compounds.

INTRATRACHEAL INJECTION STUDY

Preparations Administered to Animals

Suspensions of the titanium phosphate were prepared with 0.85% saline solution so that 0.5 ml of the suspensions would deliver 50, 10, and 2.5 mg of the titanium phosphate/kg body weight of the animal.

Suspensions of the ground asbestos were prepared in 0.85% saline solution so that 0.5 ml of the suspension would deliver 10 mg of the asbestos/kg body weight of the animal.

Experimental Design

The study was conducted in two phases. Phase I was a short term study, using the hamster, that was designed to follow the course of the inflammatory response over an eight week period subsequent to a single intratracheal injection of the titanium phosphate suspensions, the asbestos suspension or the saline solution. Phase II was a long term study to determine whether the titanium phosphate and asbestos fibers would produce a fibrogenic response in the hamster as well as in the rat. The animals used in these studies were randomly selected. The design of both these studies has been summarized in Chart I.

Experimental Procedure

The rats and hamsters used in the Intratracheal Injection Study were housed three and five per cage, respectively, and a standard laboratory chow and water was made accessible to them at all times.

The animals, prior to treatment, were lightly anesthetized with methoxyflurane and then were treated intratracheally with titanium phosphate suspenion or asbestos suspension, or saline solution. The technique used involved insertion of a sterile trocar into the trachea to within approximately 0.5 cm of the bifurcation of the trachea. A dose of 0.5 ml of the test suspension or saline solution were slowly deposited, using a tuberculin syringe attached to a piece of 11 mm plastic tubing, in the designated region of the bifurcation. The rats were placed on an inclined plane after dosing and whenever necessary, given artificial respiration until breathing appeared normal or the rats had recovered from the anesthesia. To facilitate uniform dispersion of the various suspensions or the saline solution throughout the three lobes of the lungs of the hamsters, these animals were rotated laterally immediately after deposition. The hamsters were then handled in a manner similar to the rat.

Spontaneous deaths were recorded. Body weights were taken weekly and the terminal body weights were analyzed by the Student's "t" test.

In the short term study, at the designated necropsy times, the hamsters were lightly anesthetized with methoxyflurane, the trachea clamped off using a bull-dog clamp and the animals killed by decapitation. The lungs and mediastinal lymph nodes were removed and fixed in 10% formalin, sectioned at 6μ and stained with hematoxylin and eosin for histopathologic examination.

In the long term study, at the designated necropsy times, the hamsters and rats were killed by decapitation and examined for gross pathological changes. The lungs and mediastinal lymph nodes and portions of the following organs and tissues were removed and fixed in 10% formalin, sectioned at 6μ and stained with hematoxylin and eosin for histopathologic examination: pituitary, aorta, brain, adrenal, accessory sex glands, mesenteric lymph node, stomach, small intestine, pancreas, thyroid, colon, kidney, heart, testes, spleen, liver, spinal cord, peripheral nerve, urinary bladder, thymus, skeletal muscle, and all lesions that were observed.

Results

PHASE I - Short Term Study Using Hamsters

Titanium Phosphate Study

The hamsters in the various treatment regimens, except for two hamsters that had been treated with 50 mg titanium phosphate/kg and one that had been treated with 10 mg asbestos/kg, survived till the designated necropsy times.

Histopathological examination of sections of lungs removed one hour after treatment showed the titanium phosphate fibers clumping

in the terminal respiratory bronchioles and adjacent alveoli. Within 24 hours polymorphonuclear leukocytes (PMNs) and macrophages were present causing obliteration of alveolar architecture, pulmonary edema, and focal areas of atelectasis containing titanium phosphate fibers and macrophages. The edema was less severe at the 10 mg/kg levels than at the 50 mg/kg level.

At 72 hours the giant cells were forming and much of the titanium phosphate was within alveolar macrophages. The alveoli in the terminal respiratory bronchiole areas were obliterated by minimal edema, macrophages and PMNs and proteinaceous debries.

At the end of one week there was no pulmonary edema, but there was an increase in the number of giant cells. The inflammatory reaction to 10 mg titanium phosphate/kg was less severe than to 50 mg titanium phosphate/kg.

By 8 weeks there was a granulomatous response consisting of mononuclear cells, giant cells containing the titanium phosphate fibers, and a minimal number of PMNs. The granulomatous inflammatory response focally obliterated the alveolar architecture.

Asbestos Study

Histopathological examination of sections of lungs from hamsters treated with asbestos revealed severe pulmonary edema and numerous PMNs within 24 hours. Throughout the first week the inflammatory reaction was diffuse and consisted predominantly of PMNs. After 2 weeks, foci of mononuclear cells, and PMNs, were present and by 8 weeks these inflammatory foci consisted of mononuclear cells with scattered lymphocytes, plasma cells, and golden structures interpreted as asbestos bodies.

PHASE II - Long Term Study Using Rats and Hamsters

Mortality - The mortality data on the rats on the long-term study are presented in Table I. The study was concluded after 17 months due to the increase in spontaneous deaths primarily caused by endemic chronic murine pneumonia. The number of rats surviving until the designated necropsy times are given in Table II. Several animals were sacrificed prior to their designated necropsy times due to injury and/or apparent ill health. The times at which these animals were necropsied are also included in Table II.

The spontaneous mortality data on the hamsters on the long term study is presented in Table III. The mortality data of the titanium phosphate and asbestos treated hamsters is similar to the saline controls. The random mortality distribution suggests that there is no direct correlation with treatment. The study was concluded at 18 months to be consistent with the long term rat study. The number of hamsters that survived until the designated necropsy times are given in Table IV.

Body Weight - The terminal body weight data for the 6, 12, and 18 month necropsies are presented in Tables V (rats) and VI (hamsters). The statistically significant body weight difference in the rat weights are interpreted as not of practical significance with respect to treatment. The male hamsters treated with 50 and 2 mg/kg titanium phosphate, and those treated with 10 mg/kg of asbestos and necropsied at 12 months had significantly decreased body weights. The females treated with 10 mg/kg asbestos had significantly increased body weights at the 6 month necropsy. The significant body weight differences in the hamster weights are interpreted as not being of practical significance as there was no dose response in the titanium phosphate treated males at 12 months and no significant body weight differences in either sex dosed with the titanium phosphate or asbestos at the 17-18 month necropsies.

Gross Pathology - The principal lesion seen grossly in the rats and hamsters treated with titanium phosphate, asbestos or saline solution and necropsied at the designated times consisted of focal consolidation of the lung (Table VII). Pulmonary consolidatic was a consistent finding in control rats and reflected the high incidence of chronic murine respiratory disease in these animals. Consolidated pulmonary foci were not observed in hamsters treated with 10 or 2 mg/kg of titanium phosphate.

Histopathology

Titanium Treated Animals - Histopathologically, the pulmonary response in rats to intratracheal injection of 50 mg titanium phosphate/kg at 6 months consisted of multifocal granulomas, macrophages and giant cells containing titanium phosphate fibers with scattered PMNs. The areas of inflammation were discrete foci that obliterate the alveolar architecture. The granulomas did not contain collagen. Scattered focal areas of atelectasis containing titanium phosphate fibers were observed.

The inflammatory response at the 10 and 2 mg/kg levels consisted of scattered macrophages and giant cells, containing the titanium phosphate fibers, within the alveoli and alveolar wall in the area of the terminal respiratory bronchiole. The alveolar architecture was not destroyed.

The reaction to the titanium phosphate was essentially the same at 12 and 17 months as at 6 months. At 17 months there was minimal amount of collagen within the focal granulomas at the 50 mg/kg level. The titanium phosphate fibers did not appear to be readily cleared from the lung as evidenced by their presence at the 10 and 2 mg/kg levels after 17 months.

At 6 months and throughout the remainder of the study, titanium phosphate fibers were present within macrophages of the mediastinal lymph nodes of the rat. There was no proliferative or inflammatory response to these engulfed fibers.

The spontaneous rat neoplasms are tabulated in Table VIII. No neoplasms were associated with the intratracheal titanium phosphate treatment in the rat.

Histopathologically, the pulmonary response to titanium phosphate in the hamster was similar for the 6, 12, and 18 month test period. The response was mononuclear in nature and not unlike that observed at 8 weeks. The titanium phosphate was present within macrophages and giant cells and focally the granulomatous reaction at the 50 mg/kg dose obliterated alveolar architecture. The reaction at the 50 mg/kg dose was more diffusely scattered throughout the lung, whereas, the reaction at the 10 and 2 mg/kg doses was located mainly in the areas of the terminal respiratory bronchioles.

The titanium phosphate fibers did not appear to be readily removed from the hamster lung, but were, rather, engulfed by macrophages and sequestered in the lung. There was no evidence of a fibrogenic response or collagen deposition in relation to the titanium phosphate fibers. After 8 weeks, scattered macrophages containing the titanium phosphate fibers were found in the mediastinal lymph nodes. No proliferative reaction was associated with this finding throughout the 18 month test period. The spontaneously occurring neoplasms in the hamster are given in Table IX. No treatment related neoplasms were observed.

Asbestos Treated Animals - Histopathological examination of the lungs of the rats treated with asbestos and necropsied after 6 months showed scattered circumscribed collagenous nodules and a collagenous thickening of alveolar walls throughout the lungs but more commonly near terminal respiratory bronchioles. Associated with the areas of collagen deposition were asbestos bodies and scattered mononuclear cells. Morphologically, the lesions in the lungs were the same at 12 and 17 months. Asbestos produced no lesions in the mediastinal lymph nodes. Spontaneously occurring rat neoplasms are tabulated in Table VIII. There were no neoplasms induced by the intratracheal injection of asbestos in the rat.

Histopathologically, the hamster treated with asbestos and necropsied at 6 and 12 months showed multifocal areas, mainly near terminal respiratory bronchioles, of mononuclear cells with a few PMNs and asbestos bodies. A scant amount of collagen deposition was associated with the mononuclear reaction. At 18 months there was a slight increase in the amount of collagen present within the inflammatory foci, but the reaction was predominantly mononuclear. Asbestos bodies or inflammatory foci were not observed within the mediastinal lymph nodes. The spontaneously occurring neoplasms in the hamster are given in Table IX. No neoplasms were associated with treatment.

Saline Solution Treated Animals - Histopathologically, the rats treated with saline solution showed the existence of chronic murine respiratory disease. The spontaneous lesions,

which were found in all groups of rats from 6 months on, consisted of peribronchiole and perivascular lymphoid collections throughout the lung. There were focal areas of hyperplasia of the bronchiole mucosa with mucus and PMNs on the mucosa and in the lumen. Occasionally there were focal areas of PMNs and mucus in the alveoli. Much of the interstitial tissue of the alveolar walls was thickened with mononuclear cells. Spontaneously occurring neoplasms in the rats are given in Table IX.

Through the 18 month test period, a minimal number of control hamsters treated with saline solution had small foci of PMNs scattered within and on the bronchial mucosa. Occasionally an animal would have peribronchial lymphoid infiltration. No neoplasms were observed among this group of hamsters.

INTRAPERITONEAL INJECTION STUDY

Preparations Administered to Animals

Suspensions of titanium phosphate and asbestos were prepared with 0.85% saline solution so that 0.5 ml of each would deliver 10 and 2 mg/kg body weight of the animals, respectively.

Experimental Design

The Intraperitoneal Injection Study was designed to follow the course of the response to titanium phosphate and asbestos when suspensions of these materials were injected into rats and hamsters. A group of rats and hamsters that were left untreated served as control animals. The design of the Intraperitoneal Injection Study has been summarized in Chart II.

Experimental Procedure

The rats and hamsters on the intraperitoneal injection study received one injection of either a sterile suspension of titanium phosphate or asbestos in saline solution. A group of untreated rats and hamsters were used as controls. The rats were housed three/cage and the hamsters were housed five/cage. Standard laboratory chow and water were made accessible to the animals at all times. Spontaneous deaths were recorded. At the designated necropsy times, the animals were killed by decapitation and examined for gross pathological changes. The organs and tissues saved from these animals and the histological techniques used were the same as described for the intratracheal injection study.

Results

Mortality - The spontaneous mortality of the rats on the intraperitoneal injection study are given in Table X. The study

was concluded in the 17th month because of the increased incidence of spontaneous deaths. The major cause of spontaneous death was endemic chronic murine pneumonia. Due to the increased mortality among the control rats during the 13-17 months of the study, no definitive statement relating mortality to treatment can be made. The number of rats surviving until the designated necropsy times are given in Table XI.

The spontaneous mortality data on the hamsters on the intraperitoneal injection study is given in Table XII. There was no excess mortality associated with the titanium phosphate or asbestos treatments. The number of rats surviving until the designated necropsy times are given in Table XIII.

Gross Pathology

Titanium Phosphate Treated Animals - Gross pathological examination of the titanium phosphate treated rats revealed scattered abdominal adhesions and white raised foci on the abdominal viscera. The gross observations were similar at each necropsy from 6 weeks to 17 months.

Gross pathological examination of the titanium phosphate treated hamsters revealed minimal scattered abdominal adhesions and grey-white, slightly elevated foci on abdominal viscera. The gross observations were similar at all the various necropsy times.

Asbestos Treated Animals - Gross examination of the asbestos treated rats revealed scattered abdominal adhesions and smooth, slightly elevated white foci. A number of rats that were necropsied at 12 and 17 months and those dying spontaneously during that period of time had white nodular masses throughout the serosal surface of the abdominal cavity.

Gross examination of the asbestos intraperitoneally treated hamsters revealed minimal abdominal adhesions and slightly raised scattered white foci on visceral surfaces. The gross observations were similar at each necropsy from 6 weeks to 18 months.

Histopathology

Titanium Phosphate Treated Animals - Histopathologically, the titanium fibers invoked a granulomatous foreign body inflammator response in the omentum and on the serosal surface of the abdominal viscera in the rat. The inflammatory reaction consisted of giant cells and macrophages containing the titanium phosphate fibers, along with scattered plasma cells, lymphocytes and occasionally focal areas of PMNs. The response was similar for each necropsy interval and there was very little collagen deposition present.

There were no titanium phosphate treatment-related neoplasms in the rat. A male rat necropsied at 17 months had a pulmonary

reticulum cell sarcoma and another male rat had a subcutaneous fibroma of the axilla (Table XIV).

Histopathologically, the inflammatory reaction in the hamster induced by the titanium phosphate fibers at 18 months was similar to that seen at six weeks. Titanium phosphate invoked a granulomatous foreign body reaction characterized by the fibers being present within giant cells surrounded by macrophages, and scattered plasma cells, lymphocytes and occasionally focal areas of PMNs. The granulomatous reactions were not encapsulated and there was very minimal collagen deposition.

One male hamster at 10 mg/kg, had a leiomyosarcoma of the small intestine after 12 months on test. Malignancy was based on the extreme anaplastic nature of the cells in conjunction with several mitotic figures. The neoplasm appeared to have originated in the smooth muscle of the intestinal wall. The leiomyosarcoma was interpreted as not being related to treatment. A female that received 10 mg/kg titanium phosphate died after 14 months on test with generalized reticulum cell sarcoma, a spontaneous neoplasm (Table XV).

Asbestos Treated Animals - Histopathological examination of the rats necropsied throughout the test period revealed asbestos produced collagenous nodules and plaques on the abdominal serosal surfaces. Asbestos bodies and scattered mononuclear cells were associated with the areas of collagen deposition.

In addition to the inflammatory reaction, 8 male and 6 female rats had diffuse abdominal mesotheliomas that were interpreted as being related to treatment. The first mesothelioma was observed after 8 months on test (Table XIV).

All of the mesotheliomas were predominantly of a mesenchymal type with oval to spindle shaped cells. Several mesotheliomas had tubulopapillary areas with epithelial type cells. Mitoses were common and areas of collagen were present within the neoplasms. The mesotheliomas tended to spread along serosal surfaces with infiltration of contiguous structures. Occasionally liver metastasis were found.

Histopathologically, the lesions in the hamster produced by asbestos consisted of mature collagen surrounded by a minimal number of mononuclear cells, plasma cells and lymphocytes. Asbestos bodies were observed embedded in the collagenous foci. The fibrogenic response produced by asbestos in the peritoneal cavity of the hamster was not severe or progressive in nature. No neoplasms were observed in the asbestos intraperitoneally treated hamsters(Table XV).

No neoplasms were observed in the control animals (Tables XIV and XV).

REFERENCES

1. Gross, P. Westrick, M. L., McNerney J. M. Progression of experimental silicosis in the rat. Industrial Hygiene J., 19:201-204-158.

2. Gross, P. and deTreville, R. T. Experimental asbestosis. Arch. Environ. Health, 15, 638-649, 1967.

BIBLIOGRAPHY

Churg, J., Rosen, S. H., and Moolten, S. Histological characteristi of mesothelioma associated with asbestos. Annals New York Academy of Sciences, 132, 614-622, 1965.

Faulds, J. S. and Steward, M. J. Carcinoma of the lung in hematite workers. J. Path. Bact., 72, 353, 1956.

Gross, P. Chronic pathologic responses of the lung to inhaled materials. Paper presented before Gordon Research Conference, Toxicology and Safety Evaluations Session, July 30, 1969, Meriden, New Hampshire.

Harington, J. S. Chemical studies of asbestos. Annals New York Academy of Sciences, 132, 31-47, 1965.

Harington, J. S., Roe, F. J. C. Studies of carcinogenesis of asbestos fibers and their natural oils. Annals New York Academy of Sciences, 132, 439-449, 1965.

McCaughey, W. T. E. Criteria for diagnosis of diffuse mesothelial tumors. Annals New York Academy of Sciences, 132, 603-613, 1965.

Wagner, J. C. Experimental production of mesothelial tumors of the pleura by implantation of dusts in laboratory animals. Nature, 196: 180, 1962.

CHART I

EXPERIMENTAL DESIGN OF THE TITANIUM PHOSPHATE INTRATRACHEAL INJECTION STUDY

Material Injected	Dose mg/kg	Number of Animals to be Necropsied	
		Phase 1 Necropsy Times	Phase II Necropsy Times
		1 hr. 24 hrs. 72 hrs. 1 wk. 2 wk. 8 wk	6 mo. 1 yr. 1.5 yr. 2 yr.
Titanium Phosphate	50	*5 →	**6/sex →
	10	5 →	" →
	2		" →
Saline (control)	0	3 →	6/sex → (1)
Asbestos (control)	10	3 →	6/sex →

*Hamters/time interval
**Hamters and rats/time interval
(1)Number of hamsters treated for necropsy at 2 years - 5 females, 6 males.

91100001S

CHART II

EXPERIMENTAL DESIGN OF THE TITANIUM PHOSPHATE INTRAPERITONEAL INJECTION STUDY

Animal	Sex	Material Injected	Dose mg/kg	Number of Animals* to be Necropsied at 6 wks.	3 mo.	6 mo.	9 mo.	12 mo.	18 mo.	24 mo.
Rat	M-F	Titanium phosphate	10	6/sex →				6/sex →		
	M-F	Asbestos	2	6/sex →				6/sex →		
	M-F	--	--			6		6	6	6
Hamster	M-F	Titanium Phosphate	10	5/sex →						
	M	Asbestos	2	4 →						
	F	Asbestos	2	5	6 →					
	M-F	--	--					7	7	

*Hamsters and Rats/time interval.

TABLE I

SPONTANEOUS MORTALITY OF RATS ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material	Dose (mg/kg)	Sex	Total No. Animals Dying Spontaneously/ No. Animals Treated	Number of Spontaneous Deaths During Designated Time Intervals			
				1-8 wks	2-6 mo	7-12 mo	13-17 mo
$Ti_3(PO_4)_4$	50	M	13/24	9	0	1	3
		F	11/24	2	0	2	7
	10	M	10/24	3	0	0	7
		F	8/24	4	0	0	4
	2	M	12/24	5	0	2	5
		F	8/24	2	0	0	6
Asbestos	2	M	12/24	10	1	0	1
		F	5/24	0	0	0	5
Saline		M	15/24	3	0	3	9
		F	11/24	2	1	2	6

TABLE II

NECROPSY TIMES OF RATS ON LONG TERM INTRATRACHEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	No. Animals Necropsied/ No. Animals Treated	Number Animals Necropsied 6 mo	10 mo	11 mo	12 mo	17 mo
$Ti_3(PO_4)_4$	50	M	11/24	5	2	1	3	0
		F	13/24	6			4	3
	10	M	14/24	5	1		5	3
		F	15/24	5 + 2[1]	1		5	2
	2	M	12/24	5			2	5[3]
		F	16/24	5		1	5	5[3]
Asbestos	2	M	12/24	4			4	4[3]
		F	19/24	6			6 + 1[2]	6[3]
Saline		M	9/24	5			3	1
		F	13/24	5			5	3

[1]Two animals marked for necropsy in the 18th month were sacrificed during the 7th month.
[2]One animal marked for necropsy in the 24th month was sacrificed during the 14th month.
[3]One animal sacrificed in the 16th month, had bronchopneumonia.

TABLE III

SPONTANEOUS MORTALITY OF HAMSTERS ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material	Dose mg/kg	Sex	Total No. Animals Dying Spontaneously/ No. Animals Treated	Number of Spontaneous Deaths During Designated Time Intervals			
				1-8 wks	2-6 mo	7-12 mo	13-18 mo
$Ti_3(PO_4)_4$	50	M	6/24	2	1	1	2
		F	9/24	0	1	1	7
	10	M	9/24	1	1	1	6
		F	8/24	1	0	4	3
	2	M	4/24	1	0	2	1
		F	14/24	0	7	2	5
Asbestos	10	M	10/24	2	0	4	4
		F	13/24	3	2	3	5
Saline		M	7/24	0	0	3	4
		F	7/23	1	4	0	2

TABLE IV

NECROPSY TIMES FOR HAMSTERS ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	No. Animals Necropsied/ No. Animals Treated	Number Animals Necropsied at 6 months	12 months	18 months
$Ti_3(PO_4)_4$	50	M	18/24	6	5	7
		F	15/24	6	5	4
	10	M	15/24	6	4	5
		F	16/24	5	3	8
	2	M	20/24	6	4	10
		F	10/24	4	2	4
Asbestos	10	M	14/24	4	5	5
		F	11/24	6	4	1
Saline		M	17/24	6	5	6
		F	16/23	5	5	6

TABLE V

TERMINAL MEAN BODY WEIGHTS OF RATS ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material and Dose	Sex	6 Month Necropsy		12 Month Necropsy		17 Month Necropsy	
		Number of Rats	Body Weight (g)	Number of Rats	Body Weight (g)	Number of Rats	Body Weight (g)
Saline	M	5	530.00±22.06	3	467.33±42.92	1	494[1]
Ti_3PO_4							
50 mg/kg	M	5	494.40±39.16	3	452.00±22.54	-	---
10 mg/kg	M	5	514.00±68.44	5	536.80±47.17	2	471[1]
2 mg/kg	M	5	560.80±55.68	1	566[1]		506[1]
Asbestos							
2 mg/kg	M	4	553.50±36.90	4	449.25±65.23	3	406[1]
Saline	F	5	276.40±20.01	5	290.20±26.99	3	265.00±32.23
Ti_3PO_4							
50 mg/kg	F	6	273.50±11.54	4	316.50±30.03	2	287.00±49.50
10 mg/kg	F	5	321.40±27.84*	5	308.80±54.51	2	304.00±16.97
2 mg/kg	F	5	314.60±13.88	5	347.40±44.38*	4	323.00±26.81*
Asbestos							
2 mg/kg	F	6	297.33±27.21	6	352.17±38.56*	5	322.40±35.28*

Values marked with asterisks differ significantly (Student's "t" test) from control values:
*$P<0.05$.
[1]Mean body weight.

TABLE VI

TERMINAL MEAN BODY WEIGHT OF HAMSTER ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material and Dose	Sex	6 Month Necropsy		12 Month Necropsy		18 Month Necropsy	
		Number of Hamsters	Body Weight (g)	Number of Hamsters	Body Weight (g)	Number of Hamsters	Body Weight (g)
Saline	M	6	151.80±20.60	5	149.60±7.89	6	115.17±14.36
Ti_3PO_4							
50 mg/kg	M	6	128.50±16.30	5	107.80±15.00*	7	134.57±17.99
10 mg/kg	M	6	182.30±26.40	4	149.25±16.88	5	132.40±27.84
2 mg/kg	M	6	175.30±23.2	4	115.50±18.16*	10	128.40±12.99
Asbestos							
10 mg/kg	M	3	135.30±7.10	5	122.60±21.46*	5	125.40±15.98
Saline	F	5	145.60±12.36	5	120.20±22.12	6	126.67±10.11
Ti_3PO_4							
50 mg/kg	F	6	184.50±29.84	5	103.00±10.34	4	122.25±9.57
10 mg/kg	F	5	144.40±16.80	3	113.68±19.01	8	127.75±13.30
2 mg/kg	F	4	190.50±33.25	1	105.50±7.78	4	127.25±13.82
Asbestos							
10 mg/kg	F	6	175.33±7.63*	4	110.75±13.40	1	123

Values marked with asterisks differ significantly (Student's t test) from control values: *P<0.05.

TABLE VII

INCIDENCE OF PULMONARY CONSOLIDATION IN RATS AND HAMSTERS ON THE LONG TERM INTRATRACHEAL INJECTION STUDY

Material and Dose	Animal[1]	6 Month Necropsy	12 Month Necropsy	17 Month Necropsy	18 Month Necropsy
Saline	Rat	0/10[2]	5/8	4/4	--
Ti_3PO_4					
50 mg/kg		5/11	4/7	2/2	--
10 mg/kg		3/10	2/10	3/4	--
2 mg/kg		1/10	4/6	6/8	--
Asbestos					
2 mg/kg		5/10	5/10	7/8	--
Saline	Hamster	0/11[2]	0/10	---	1/12
Ti_3PO_4					
50 mg/kg		1/12	2/10	---	6/11
10 mg/kg		0/11	0/7	---	0/13
2 mg/kg		0/10	0/6	---	0/14
Asbestos					
10 mg/kg		1/9	1/9	---	1/6

[1]Sexes combined.
[2]Number of observations/number of animals examined.

ST0000124

DOW 00066

TABLE VIII

NEOPLASMS IN RATS ON LONG TERM INTRATRACHEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	Total # Rats Examined*	Neoplasm Classification
$Ti_3(PO_4)_4$	50	M	11	(2)[1] - Reticulum cell sarcoma (9 months and 10 months)[2]
		F	13	(1) - Cortical adenocarcinoma of adrenal (12 months) (1) - Carcinoma of thyroid (17 months) (1) - Uterine myoma (16 months)
	10	M	14	(1) - Diffuse lymphosarcoma (17 months)
		F	15	-----
	2	M	12	(1) - Pheochromocytoma (17 months)
		F	16	-----
Asbestos	10	M	12	-----
		F	19	(2) - Fibroadenoma of mammary gland (17 months)
Saline		M	9	(1) - Abdominal carcinoma with lung metastases (11 months)
		F	13	(1) - Adenocarcinoma of mammary gland (16 months)

*Necropsied or dying spontaneously from 6th-17th month of test.
[1]Number in parenthesis indicates the number of rats with the indicated neoplasm.
[2]Numbers in parenthesis indicate month after treatment in which observation was made.

TABLE IX

NEOPLASMS IN HAMSTERS ON LONG TERM INTRATRACHEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	Total Number Hamsters Examined*	Neoplasm Classification
$Ti_3(PO_4)_4$	50	M	20	(1)[1]-Islet cell adenoma o pancreas (18 month)[2] (1)-Pheochromocytoma (18 month)
		F	17	------
	10	M	17	(1)-Hemangioendotheloma o spleen (6 month) (1)-Pheochromocytoma (18 month)
		F	19	(1)-Islet cell adenoma of pancreas (18 month) (1)-Undifferentiated sarcoma of abdomen (18 month)
	2	M	21	------
		F	14	(1)-Adenocarcinoma of uterus (12 month)
Asbestos	10	M	15	(1)-Pheochromocytoma (18 month) (1)-Adrenal cortical adenocarcinoma (18 mont
		F	15	------
Saline		M	18	------
		F	16	------

*Necropsied or dying spontaneously from 6th-18th month of test.
[1]Number in parenthesis indicates the number of hamsters with the indicated neoplasm.
[2]Numbers in parenthesis indicate month after treatment in which observation was made.

TABLE X

SPONTANEOUS MORTALITY OF RATS ON INTRAPERITONEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	Total Number of Spontaneous Deaths/ Number Rats Treated	Number of Spontaneous Deaths During Designated Time Interval			
				1-6 wks	7 wks-6 mo	7-12 mo	13-17 mo
$Ti_3(PO_4)_4$	10	M	10/36	3	0	2	5
		F	4/36	1	0	0	3
Asbestos	2	M	14/36	3	0	4	7
		F	8/36	1	0	4	3
Control		M	15/24	3	0	3	9
		F	11/24	2	1	2	6

TABLE XI

NECROPSY TIMES FOR RATS ON THE INTRAPERITONEAL INJECTION STUDY

Material and Dose	Sex	No. Animals Necropsied/ No. Animals Treated	No. of Rats Necropsied at Each Time Period				
			6 wk.	3 mo.	6 mo.	12 mo.	17 mo.
$Ti_3(PO_4)_4$	M	24/36	5	6	6	3	4
10 mg/kg	F	32/36	6	6	6	6	8
Asbestos	M	17/36	4	6	5	2	0
2 mg/kg	F	26/36	6	6	5	5	4
Control	M	9/24	5			3	1
	F	13/24	5			5	3

TABLE XII

SPONTANEOUS MORTALITY OF HAMSTERS ON INTRAPERITONEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	Total Number of Spontaneous Deaths/ Number Hamsters Treated	Number of Spontaneous Deaths During Designated Time Interval			
				1-6 wks	7 wks-6 mo	7-12 mo	13-18 mo
$Ti_3(PO_4)_4$	10	M	4/30	2	0	2	0
		F	7/30	1	2	2	2
Asbestos	2	M	5/24	4	0	1	0
		F	6/35	0	3	2	1
Control		Mixed	4/14	0	0	2	2

TABLE XIII

NECROPSY TIMES FOR HAMSTERS ON INTRAPERITONEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	No. Animals Necropsied/ No. Animals Treated	Number Animals Necropsied at 6 wks	3 mo	6 mo	9 mo	12 mo	18 mo
$Ti_3(PO_4)_4$	10	M	26/30	5	4	5	4	5	3
		F	23/30	4	4	5	5	3 + 1[1]	1
Asbestos	2	M	19/24	3	4	3	4	3 + 2[2]	-
		F	29/35	5	5	6	4	5 + 4	-
Control		Mixed	10/14					5	5

[1]One animal marked for necropsy in the 18th month was sacrificed in the 14th month.
[2]Two surviving male and four surviving female hamsters marked for necropsy in the 18th month were sacrificed in the 14th month.

TABLE XIV

NEOPLASMS IN RATS ON INTRAPERITONEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	Number Rats Examined*	Neoplasm Classification
$Ti_3PO_4)_4$	10	M	21	(1)[1]-Pulmonary reticulum cell sarcoma (17 month)[2]
				(1)-Subcutaneous fibroma of axilla (17 month)
		F	23	------
Asbestos	2	M	20	(8)-Diffuse abdominal mesothelioma (1-10mo.; 3-11 mo.; 1-14 mo.; 3-16 mo.)
		F	21	(6)-Diffuse abdominal mesothelioma (1-8 mo.; 2-12 mo.; 3-16 mo.)
				(1)-Generalized reticulum cell sarcoma (8 month)
Control		M	9	------
		F	13	------

*Necropsied or dying spontaneously from 6-18 months on test.
[1]Number in parenthesis indicates the number of rats with the indicated neoplasms.
[2]Numbers in parenthesis indicate month after treatment in which observation was made.

TABLE XV

NEOPLASMS IN HAMSTERS ON INTRAPERITONEAL INJECTION STUDY

Material Injected	Dose mg/kg	Sex	No. Hamsters Examined*	Neoplasm Classification
$Ti_3(PO_4)_4$	10	M	18	(1)[1]-Leiomyosarcoma of small intestine (12 month)[2]
		F	19	(1)-Reticulum cell sarcoma (14 month)
Asbestos	2	M	13	----
		F	20	----
Control		Mix	11	----

*Necropsied or dying spontaneously from 6th-18th month of test.
[1]Numbers in parenthesis indicate the number of hamsters with the indicated neoplasms.
[2]Numbers in parenthesis indicate month after treatment in which observation was made.

Toxicology Contact - H.C. Spenc

DOW THE DOW CHEMICAL COMPANY

BIOCHEMICAL RESEARCH LAB. MIDLAND, MICHIGAN 48640

TOX. NO. DD-54

FILE NO. T34.19-DD-54-(

REQUEST FOR TOXICOLOGICAL STUDIES 1803 BLDG.

% OF MATERIAL TO BE TESTED

Titanium Phosphate - $Ti_3(PO_4)_4$

SUBMITTED BY (Signature)	(Date)	(Building)	(Phone)
Present contact - R. G. Heitz ~~F. D. Axe~~	1967	Western Division	
SUPERVISOR (Signature)	(Date and Building)	PROJECT SAFETY COORDINATOR	Date
V. K. Rowe		L. L. Pitchforth	

ESTIMATED COST	ACCOUNT NUMBER	LEDGER / LOCATION GROUP FUNCTION / SECTION SUB FUNC. / SUFFIX	PROBLEM NO. TO BE USED BY TOXICOLOGY	PROBLEM INDEX NO. LAB. / PROBLEM
$40,000		Western PO 74484	4-73-000-7-38-2	9 9 8 0.0 0 5 2 0 9

DO NOT WRITE BELOW THIS LINE COMPLETE ALL INFORMATION ON REVERSE

ROUTE OF ADMINISTRATION: Single dose IP and Intratracheal

LENGTH OF TREATMENT: up to 2 years

SPECIES:

(RAT) ☒M ☒F RABBIT ☐M ☐F GUINEA PIG ☐M ☐F

DOG ☐M ☐F MONKEY ☐M ☐F (HAMSTER) ☒M ☒F

OTHER

DOSES: 0, 50, 10, 2 mg/kg

CHECK POINT	DATE
AMPLE RECEIVED TOXICOLOGY	1967
[redacted] STARTING TIME	5/68
ESTIMATED REPORTED DATE	1970
REVISED ESTIMATE REPORTED	
REPORT DRAFTED	
REPORT ISSUED	

FATE OF SAMPLE	DATE
☐ RETURNED	
☐ DESTROYED	
☐ REFERENCE SAMPLE STORED	
☐ REFERENCE SAMPLE TO K STORAGE	

NOTES

DATE PROJECT COMPLETED

NOTES, CHANGES, ADVERSE EFFECTS NOTED

7-31-69: To date, groups of rats and hamsters, injected IT or IP, have been necropsied 6 weeks 3, 6, 9 or 12 months following treatment. 18 & 24-month necropsies are scheduled.

9-15-69: Animals continue on expt. as schedule
10/13/69: Animals continue on expt. as schedule
11/4/69: "Epidemic" of pneumonia has developed in the past week, with considerable mortality. Tetracycline is being administered in drinking water.
Susan McCollister

11/19,20/69 - Due to the high mortality, it was decided that there would not be enough animals to continue the experiment as originally scheduled. Therefore, the animals are being necropsied these dates (approximately 18 months following treatment). Susan McCollister

12/8/69 - Tissues are being processed for slide preparation. G. Sparschu and Susan McCollister

1/6/70 - Tissue preparation still in progress. Susan McCollister

2/13/70 - No Change. Susan McCollister

3/15/70 No Change. Susan McCollister

4/12/70 - No Change. Susan McCollister

9-1-71- Pathology has been completed and Path report given to J. Norris for final reporting - G. L. Sparschu

ST0001363

S7000 MOD

FORM C-32 '70 PRINTED IN U.S.A. 3-69

SAMPLE INFORMATION

NAME

Titanium Phosphate

MOLECULAR FORMULA	MOLECULAR WEIGHT	SAMPLE REFERENCE, SOURCE AND OR IDENTIFICATION
$Ti_3(PO_4)_4$		

SYNONYMS

STRUCTURAL FORMULA OR COMPOSITION

PHYSICAL AND CHEMICAL PROPERTIES

ESTIMATED PURITY OF SAMPLE	SAMPLE ANALYSIS	RESULTS
	[X] YES [ ] NO	See File (S. McCollister)

BOILING POINT	EXPLOSIVE LIMITS (% BY VOLUME IN AIR)	FLASH POINT	IGNITION TEMP.	MELTING POINT	VAPOR PRESSURE
°C mmHG		°F	°C	°C	mmHG 25°C

CORROSIVENESS (to common metals)	PHYSICAL STATE	COLOR

CHEMICAL REACTIVITY	ODOR (include concentration in air)

STABILITY (to air change, heat, light)

Why are toxicological studies needed? Include stage of development, proposed use, method of handling, and amount handled. Do you know of any adverse effects from human exposure?

This study was requested to evaluate the fibrogenic potential of $Ti_3(PO_4)_4$. Included as controls are asbestos and saline.

ST0000134

	DATE		DATE		DATE
[ ] PROTOCOL COMPLETED		[ ] STATISTICS DONE		[ ] SLIDES TO BE READY	
[ ] ANIMALS ORDERED		[ ] CLINICAL TESTS REPORTED		[ ] SLIDES READ	
[ ] ANIMALS STARTED ON TEST		[ ] HEMATOLOGY TESTS REPORTED		[ ] HISTOPATH REPORTED	
[ ] NECROPSY DONE		[ ] SLIDES STARTED		[ ] WORK ALL DONE, READY TO WRITE	

FORM C-32790 PRINTED IN U.S.A. 3-69

(BACK)

DOW 00076

Rad 401 – cage 10

CAGE: 10
MARK: 10
BIRTH DATE: —
AGE IN DAYS: —
STARVED: no

OD-54-(1)
SLIDES FILED 48 OC
BLOOD COUNTS ( )
BONE MARROW COUNTS ( )

BIOCHEMICAL RESEARCH DEPARTMENT

68-427

TISSUE EXAMINATION

ANIMAL NO.: DD54 - [redacted] 2,10
MATERIAL: $Ti_2(PO_4)_4$ 2 mg/kg

FILE NO. T
EXPT. NO.
RUN NO.
DAYS
DIED ( )
KILLED ( )

GROSS EXAMINATION

DATE: 10-9-68
CONDITION:
BODY WT.: _____ GRAMS
KILLED BY DECAPITATION ( )
OR
EXAMINATION OF ORGANS:
APPEARANCE OF VISCERA:

MICROSCOPIC EXAMINATION

FORMALIN ( ) OR _____
DATE REMOVED FROM FIXATIVE _____
EMBEDDED: PARAFFIN ( ) OR _____
SECTIONED BY _____
STAINS: HEMATOXYLIN AND EOSIN ( )
OR _____
EXAMINATION OF SECTIONS:

WT. (g)	TISSUE SAVED	DEGREE OF DEG.	LUNG (VENTRAL / DORSAL)	SLIDE PREP.	DEGREE OF DEG.	
			LUNG:			LUNG:
			HEART:			HEART:
			LIVER:			LIVER:
			KIDNEYS:			KIDNEYS:
			SPLEEN:			SPLEEN:
			ADRENALS:			ADRENALS:
			PANCREAS:			PANCREAS:
			TESTES:			TESTES:
			BONE MARROW:			BONE MARROW:
			STOMACH:			STOMACH:

ST0000135

DATED _____ SIGNED _____ DATED _____ SIGNED _____

1219-6 PRINTED IN U.S.A.

DOW 00077

GROSS NECROPSY

Animal #DD54 210
brain

There was a subdural hemorrhage approximately 1/2 centimeter in diameter on the left mid-cerebrum. The left ear canal was filled with a yellowish-green somewhat caseous material which extended up into the brain following the route of the 8th nerve.
Diagnosis - Otitis interna.

ST0000136

Philip B. Conran, D.V.M.
October 9, 1968

MICROSCOPIC EXAMINATION
Animal #68-427

brain — This section of brain is taken through the hippocampus and focally there is an area of acute necrosis characterized by coagulation necrosis and focal hemorrhage. Immediately adjacent to this is a moderate amount of sub-dural hemorrhage.

brain — This section of brain is characterized by focal areas of hemorrhage and necrosis with occasional polymorphonuclear inflammatory cells being noted. Focally there appears to be a thrombotic vessel. Immediately adjacent to this vessel are focal areas of hemorrhage.

brain — This section of brain is characterized by having the presence of the third in the two lateral ventricles and suspected of being ante: to the hippocampus section. Focally there is a wedge shaped area of necrosis with pin-point areas of hemorrhage. Associated with this area of necrosis, there is massive subdural hemorrhage. The distribution of the lesion is characteristic of possible vascular occlusion. Within the necrotic area, there are occasional polymorphonculear inflammatory cells noted, however for the most part the lesion is very acute and the tissue appears to have died in situ without inflammatory exudate.

REPORT CONTINUED

P.B. Conran
November 13, 1968

MICROSCOPIC EXAMINATION
Page 2
Animal #68-427 (con't)

brain	This section of brain is taken through the hippocampus and again there is a focal area of necrosis with pinpoint hemorrhage. Within the central area of the necrotic area there is a thrombosed vessel.
brain	This section of brain is taken through the cerebellum and there appears to be severe congestion of the cerellar vessels.

Final Diagnosis:
Infarction, focal, hemorrhagic, acute, cerebrum, brain, due to thrombosed veins.

Philip B. Conran, D.V.M.,
November 13, 1968

ST0001837

DOW 00079