A/C Pipe
Producers Association

Bill Parrott

Angela J. Bayless

PUBLIC AFFAIRS

Public Affairs encompass those research, educational and promotional (REP) programs which provide the basis for public education activities, field problem containment and regulatory affairs initiatives. Accountability for program planning, review and implementation is assigned to the Public Affairs Committee (PAC); approved scope appended as MIS-08-80. AACPP's Director, Public Affairs serves as PAC program coordinator and staff liaison.

PAC Program Strategy

PAC program strategies are predicted on the premise AACPP member companies will:

1) mitigate market erosion by confronting asbestos health issues whenever, wherever and by whomever raised.

2) build an affirmative case for the A/C pipe sector in asbestos rulemaking proceedings.

Strategically determining which objective merits more attention is difficult. Regulation accelerates market erosion; market erosion facilitates regulation ... an interdependency regulators are exploiting. Nonetheless, with limited resources a choice must be made. It is axiomatic that without a market for A/C pipe the industry cannot survive despite any long term success in the regulatory arena. Thus, PAC program strategy is to concentrate on public education programs on a budgeted basis, while proceeding with selected regulatory affairs initiatives on an "as needed, pay as you go" basis via special assessments.

"Target audiences" are numerous and diverse. The "bullseye" is comprised of A/C pipe customers i.e., appointed public works officials who purchase pipe. In the next ring are elected municipal officials, i.e., final arbiters of procurement policy. The next ring represents a mixed bag of vested interests i.e., public health officials, news media, public interest groups, etc. Although one step removed from the procurement process, they still influence municipal policy and mold opinion in the outer ring, the general public.

Each "target audience" has its own perceptions about A/C pipe and program implementation will reflect this diversity. Tactically, the object is to shoot at the "bullseye" continuously, hit the municipal official ring as frequently as possible, and pepper the vested interest and general public target areas when necessary.

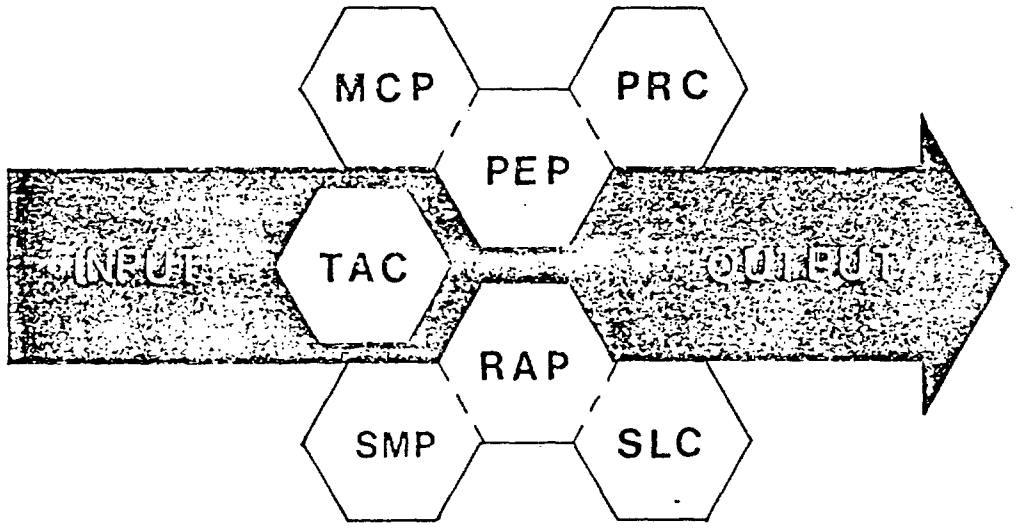
In sum, PAC program strategy is to retain public relations counsel and expand public education and monitoring programs to safeguard A/C pipe "core markets." The need to develop an independent affirmative case for the A/C pipe sector in rulemaking proceedings will be reviewed and funded on a contingency basis.

PAC Program Implementation

PAC program implementation hinges on financial commitment and use of outside experts i.e., public relations counsel, special legal counsel, and state legislative monitoring services to meet strategic objectives.

With program control vested in outside experts to varying degrees, cooperative decision-making is prerequisite to timely and relevant program output. Two other prerequisites, technical support and member company involvement will maximize return on investment. A final prerequisite is a program structure which will provide program flexibility (diversity), program cohesion (cooperation) and program autonomy (options).

PAC program structure is based on a series of interlocking program and support modules to reflect these basic prerequisites plus generate program momentum and synergism. The interaction of PAC program modules is illustrated below:



The Public Education Programs (PEP) module is an output module designed to educate "target audiences" about A/C pipe safety-benefits. The number and composition of individual program elements will fluctuate to reflect regulatory developments, new medical data, containment problems and program feedback (PAC-02-80).

The Regulatory Affairs Program (RAP) module is an output module designed to build an affirmative case for the A/C pipe in asbestos rulemaking proceedings. The number and composition of program elements are finite and will be activated on an "as needed" basis (PAC-01-80).

The State Monitoring Program (SMP) is an input-output module to monitor state legislative activity and to retain special counsel on an "as needed" basis.

The Public Relations Counsel (PRC) and Special Legal Counsel (SLC) modules are input-output modules representing out-of-house support for PAC activities.

The Technical Affairs Committee (TAC) and Member Company Programs (MCP) modules are input and output modules respectively, representing in-house technical and sales support for PAC activities.

Current PAC program status is summarized on the opposite page with individual program module worksheets appended in numerical order.

PROGRAM STATUS

Committee	PAC
Issue Date	JAN. 1981

Program		Schedule			Cost
NUMBER	MODULE/TASK	1980	1981	1982	P-Plan A-Actual
PAC-01-80	RAP/Task 1				P-$105,000 A- 90,785
PAC-02-80	PEP/Task 3A				P-$ 27,150 A- 2,881
	PEP/Task 3B				P-$ 10,150 A- 2,448
	PEP/Task 3C				P-$ 6,650 A-$ 1,458
	PEP/Task 3D				P-$ 17,150 A- 0
	PEP/Task 3E				P-$ 62,775 A- 3,261
	PEP/Task 4A				P-$ 3,150 A- 1,438
	PEP/Task 4B				P-$ 7,150 A- 1,438
	PEP/Task 4C				P-$ 25,300 A- 2,753
	PEP/Task 4D				P-$ 25,750 A- 0
PAC-04-80	SMP/Pending				P-$ 15,000 A- 0

Cash Flow $					
	1	6000*	61195		*Prior FY 80 Programs
	2	3000*	43890		
	3	52500	49475		
	4	65300	32865		
	T	126800	187425		

YTD Expense $					
	1	6000*			*Prior FY 80 Programs
	2	2771*			
	3	51305			
	4	55283			
	T	115359			

PROGRAM STATUS	Committee	PAC
	Issue Date	Jan., 1981

Program		STAFF UPDATE
NUMBER	MODULE	
PAC-01-80	RAP	Task 1, Economic Impact Analysis, has been completed and a draft forwarded to counsel on December 16, 1980. Staff and counsel will provide comments by January 31, 1981. A program element of the impact analysis, an Opinion Research Corp. survey of 300 water utilities, was released to Executive Committee on December 16, 1980. Initiation of other RAP tasks continues to be deferred pending rulemaking strategy changes resulting from Reagan election, as well as decisions by AIA/NA to proceed on similar regulatory initiatives. The possibility of "negotiated regulations" with EPA has been explored with Agency representatives. Proposal that A/C pipe sector proceed independently or as a model for other product sectors was discussed with Executive Committee on December 16, 1980; approval pending.
PAC-02-80	PEP	PR counsel has prepared draft materials for (a) press kit and releases, (b) suggested target audiences for receipt of press kit, (c) special inserts or "tip ons" for "A/C Pipe and Drinking Water" booklet, (d) a "letter to the editor", (e) an op-ed article, (f) a revised protocol for handling field problems, (g) a proposal for professional spokesperson training, (h) an advocacy ad and a market pre-test methodology, (i) a safety ad. These materials will be finalized during January for review and approval by the Public Affairs Committee in February. No action has been taken on revision of A/V programs, utility initiative, state of the art paper, or SWAT team.
PAC-03-80	EPA Reg-Neg	See PAC-01-80. Further Staff action pending Executive Committee approval to proceed.
PAC-04-80	SMP	Staff awaiting recommendations by counsel.

PUBLIC AFFAIRS PROGRAM	Number	PAC-01-80
	Issued	9/12/80
	Revised	2/23/31

Title REGULATORY AFFAIRS PROGRAM

Background

This program will coordinate the collection, analysis and presentation of data necessary to develop and prosecute an affirmative case for the A/C pipe sector in EPA, OSHA and CPSC rulemaking proceedings.

Program elements required to develop an affirmative case include: economic impact analyses, risk assessments of substitutes and product life cycle audits of asbestos exposure levels. The technological feasibility and economic impact of reduced exposures must be evaluated and risks quantified for low-level exposures in both the occupational and general environment. Finally, special legal counsel must prepare formal comment and otherwise prosecute the industry's case.

Primary responsibility for planning and preparing industry responses to current rulemaking resides with AIA's special counsel, Kirkland & Ellis. Within their general fact-finding framework, AACPP is acting as an independent subcommittee to coordinate and collect information relevant to the A/C pipe product sector. AACPP's role will continue to be one of actively cooperating and financially supporting essential information-gathering exercises while relying on management oversight to assure A/C pipe's case is not weakened in the broad defense of other product sectors and reserving the option of proceeding independently if desired or necessary.

Since this program is proceeding on a contingency basis, program funding and timetables necessarily reflect that rationale i.e., implementation of additional program elements will be approved on an "as needed -- pay as you go" basis via special assessments rather than budgeted in advance.

Objective

To build in advance of rulemaking an affirmative case for A/C pipe based on:

1) Economic impact analysis of a possible ban on A/C pipe
2) Risk assessment of substitute pipe materials
3) Audit of asbestos exposure levels during product life cycle
4) Technical/economic feasibility studies of reduced exposures
5) Risk assessment of low-level asbestos exposures
6) Retention of legal counsel

PAC-01-80

Tactics & Tasks

Program Review & Initiation

Review quarterly with Executive Committee status of AIA/AACPP information gathering. Resolve "go - no go" decision whether to proceed independently with selected program elements or in toto. If affirmative, identify candidate contractors. Prepare and distribute request for proposal (RFP). Review proposals and select contractor. Implement legal safeguards and let contract. Monitor progress monthly.

1. Economic Impact Analysis of Total or Partial Ban on A/C Pipe

   RFP Issuance: March 3, 1980
   Contractor Selection: May 6, 1980; Arthur D. Little, Inc.
   Program Initiation: June 13, 1980
   Status: In progress, analysis to be completed by November, 1980.

2. Life Cycle Risk Assessment of Substitute Pipe Materials

   RFP Issuance: May 9, 1980
   Contractor Selection: Pending; Arthur D. Little, Inc. and MRI
   Program Initiation: Pending
   Status: Deferred by Executive Committee July 17, 1980 pending further rulemaking developments.

3. Audit of Asbestos Exposure Levels During Product Life Cycle

   RFP Issuance: May, 1980
   Contractor Selection: Pending; Arthur D. Little, Inc.
   Program Initiation: Pending
   Status: Deferred by Executive Committee July 17, 1980 since AIA contemplating audit via EPA TSCA Section 8(a) reporting form and supplementary questionnaires for primary asbestos manufacturing sectors.

4. Technical/Economic Feasibility Studies of Reduced Exposures

   RFP Issuance: May, 1980
   Contractor Selection: Pending; Arthur D. Little, Inc.
   Program Initiation: Pending
   Status: Contractor to submit revised proposal and cost for AIA Executive Committee, September 16, 1980.

5. Medical Evidence/Risk Assessment of Low-Level Asbestos Exposures

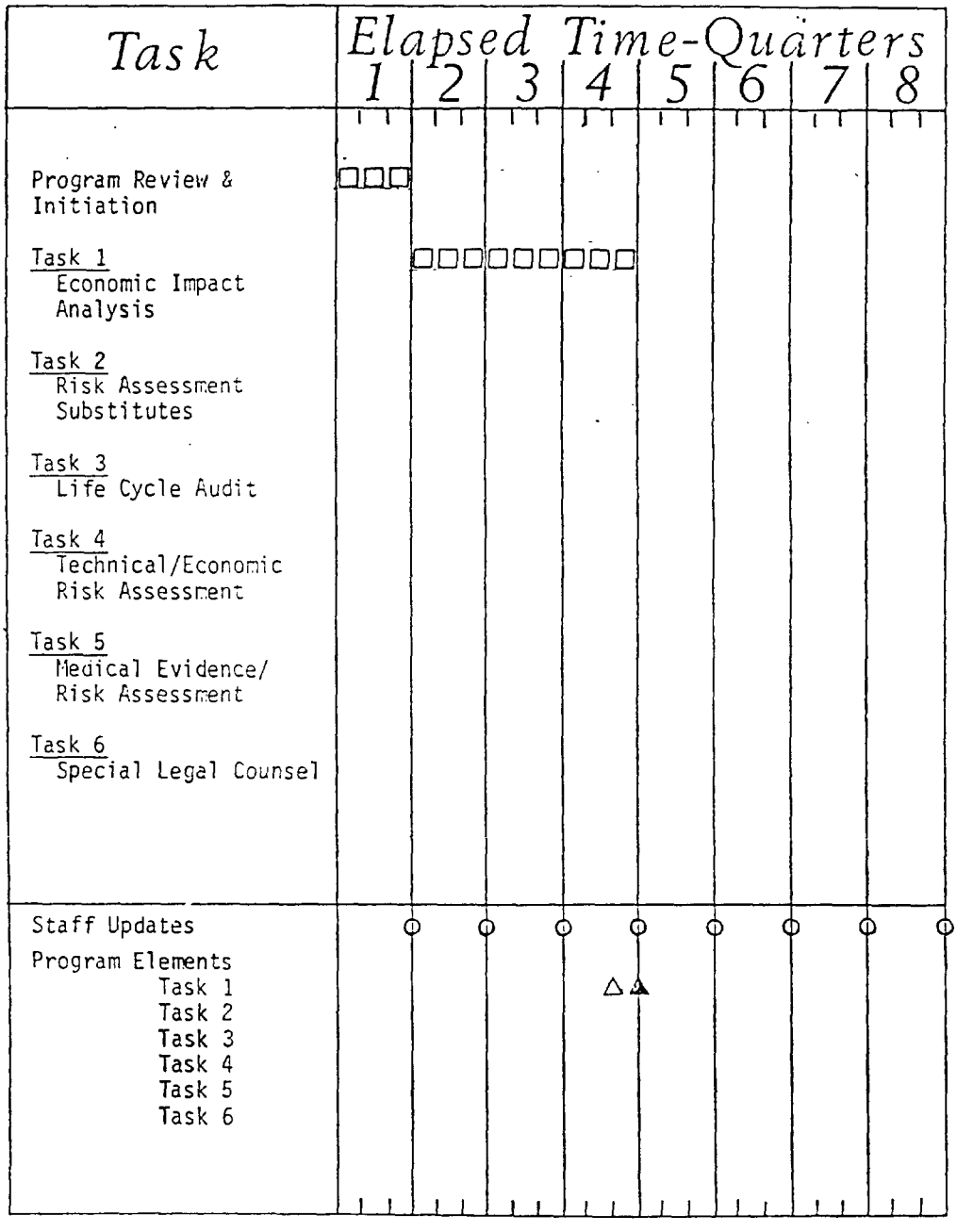
   RFP Issuance: Pending (AIA/NA)
   Contractor Selection: Pending
   Program Initiation: Pending
   Status: Exercise to be coordinated by Dr. Kotin (Johns-Manville).

6. Retention of Special Legal Counsel

   Status: To be reviewed quarterly by AACPP Executive Committee

PAC-01 -80

Schedule



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PAC- 01-80

Estimated Costs

Implementation Developmental

Required Input

1.	Economic Impact Analysis	$105,000
2.	Risk Assessment - Substitutes	$ 90,000
3.	Life Cycle Audit*	$ 70,000
4.	Technical/Economic Feasibility Studies*	$ 40,000
5.	Medical Evidence/Risk Assessment	**
6.	Special Legal Counsel	**

* Inter-dependent studies
** To be determined

Planned Output

Cash Flow - Quarters

1	2	3	4	5	6	7	8	
$52,500	$52,500							$105,000
		$80,000	$80,000	$40,000				$200,000
		**	**	**	**	**	**	**
$52,500	$52,500							$105,000
		$80,000	$80,000	$40,000				$200,000

PUBLIC AFFAIRS PROGRAM	Number	PAC-02-80
	Issued	9/12/80
	Revised	2/23/31

Title

PUBLIC EDUCATION PROGRAMS

Background

This program will expand current efforts to educate selected municipalities, consumers and public officials about relevant asbestos health issues and cost-benefits of A/C water pipe through the coordinated use of public relations counsel.

Currently, AACPP's education strategy is predicated on the use of knowledgeable industry and staff personnel. Whenever feasible, prepackaged educational programs (e.g. "A/C Pipe and Drinking Water") are used. When field problems develop, a standard protocol is followed to provide a prompt response and assign accountability to either a designated member company, AACPP Staff or special counsel. The education strategy and response protocol are effective within limits with results varying from crisis to crisis depending upon individual expertise, lead time and media exposure.

A growing consumer awareness of potential asbestos hazards, sensationalist media coverage and adverse publicity generated by government pronouncements have undermined consumer confidence and eroded product loyalty toward A/C pipe. This ebb of consumer confidence must be stemmed in order to protect A/C pipe's market share and extend its product life-cycle.

Thus, there is an immediate need to develop and implement broad-based public education programs which will create a receptive environment for A/C pipe. To facilitate this effort, the use of public relations counsel is recommended.

Planning is based on initiating priority public education program modules in FY 80.

Objective

To develop in conjunction with public relations counsel, a modular public education program which can be implemented selectively to safeguard "core markets" for A/C pipe, yet be adaptable to field problems in other parts of the country.

PAC-02-80

Tactics & Tasks

1. Program Initiation

   Approve modular PR concept. Select agency; commence Task 2.

2. Module Selection

   Resolve program module priorities and implementation methodology. Define accountabilities; commence developmental work on Tasks 3-11.

3. Module Priorities - PR Counsel Accountabilities

   A. Fact Book/Inserts
   Prepare "A/C Pipe and Drinking Water" fact book plus a series of special inserts adaptable to a variety of target audiences.

   B. Media Backgrounder
   Prepare press kit on risk-benefits of A/C pipe. Utilize for field problem containment and after Public Affairs Committee approval, distribute to press in selected "core markets."

   C. Seminars
   Arrange industry presentations to AWWA Section and other selected meetings.

   D. Audio-Visual Programs
   Evaluate existing A/V Program; modify accordingly. Update scientific data. Prepare short introduction speech and Q & A sheet to fortify presentation.

   E. Advertising
   Prepare options paper on how to assess impact of advocacy safety ads for A/C pipe. Develop and pre-market test ads if necessary. Make "go - no go" decision. Prepare ads on cost-performance (TAC-02-80). Approve and place.

4. Module Priorities - Staff Accountabilities

   A. Utility Initiative
   Establish state/utility priority list for "core markets." Broaden corporate educational efforts in advance of crises. Prepare camera ready mailer suitable for use by water utilities with field problems.

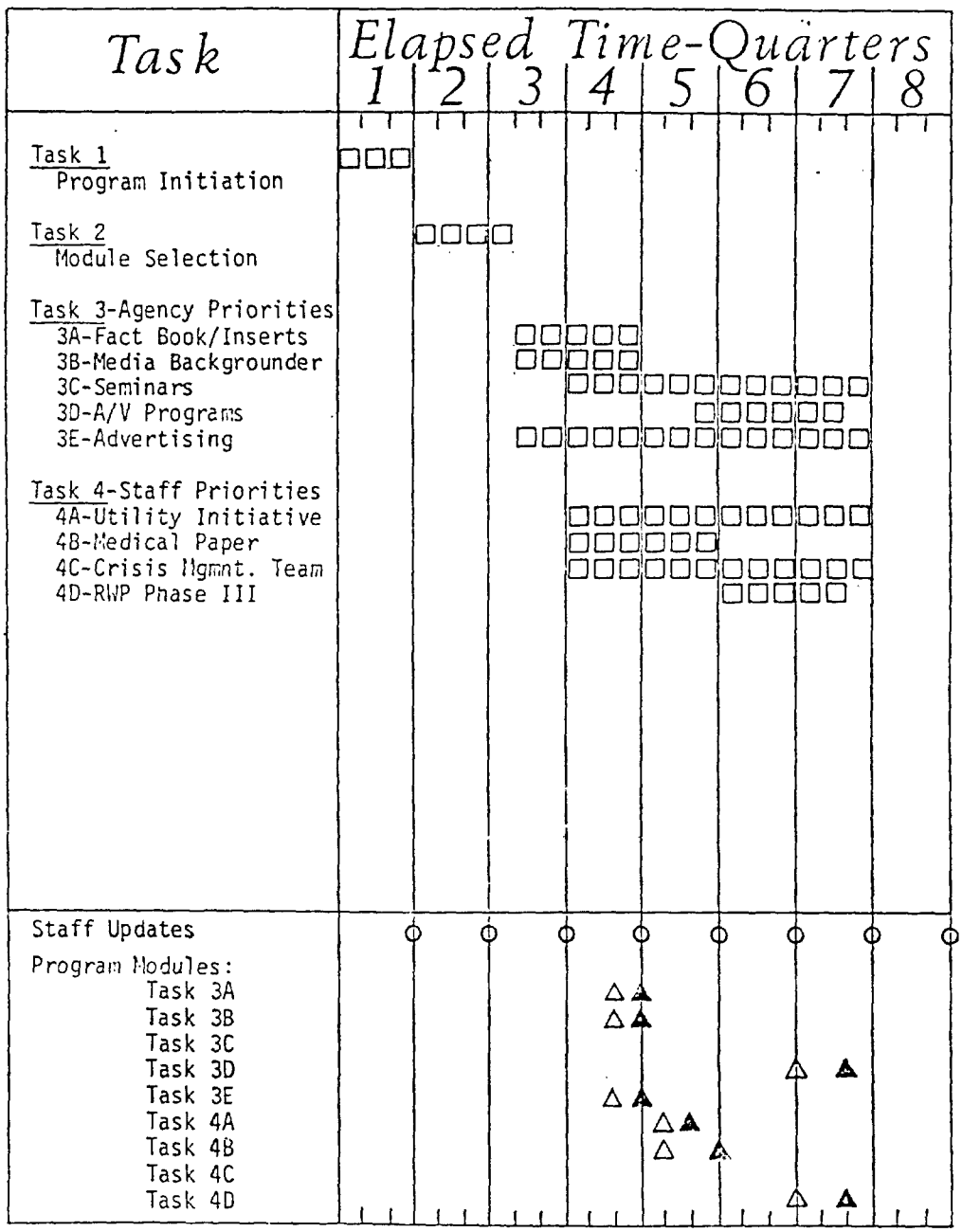
   B. Medical Paper
   Prepare "state of the art" medical paper supporting industry position. Direct mail to public health officials. Use in field problem containment.

   C. Crisis Management Team
   Organize and train a crisis management team to coordinate industry response to field problems (e.g. media specialist, scientist, etc.).

   D. Recommended Work Practices (RWP): Phase III
   Revise RWP to reflect recent developments (e.g., TAC-02-80), new data and suggested revisions. Develop "contractors certification form."

PAC-02-80

Schedule



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PAC-02-80

Estimated Costs	Implementation Developmental		
Required Input			
1. AACPP Staff Travel			
A. Agency Management			$ 1,500
B. AIA Communications Advisory Panel and International Affairs Committee			$ 3,500
C. Seminar Presentations			$ 3,000
Planned Output			
1. PR Counsel Accountabilities			
A. Fact Book and Inserts		$11,500	$15,500
B. Media Backgrounder		$ 2,000	$ 8,000
C. Seminars		-	-
D. A/V Programs		$14,000	$ 3,000
E. Advertising - Cost Performance		$13,000	$35,200
- Product Safety		$ 6,500	$ 7,925
2. Staff Accountabilities			
A. Utility Initiative		$ 2,500	$ 500
B. Medical Paper		$ 5,000	$ 2,000
C. Crisis Management Team (incl. advocacy ads)		$13,000	$12,000
D. Work Practices - Phase III		$ 4,000	$21,600

Cash Flow - Quarters									
1	2	3	4	5	6	7	8		
$12,500	$36,000	$ 6,500	$16,500	-				$71,500	
$ 300	$21,445	$33,640	$29,225	$29,115					$113,725
$12,800	$57,445	$40,140	$45,725	$29,115				$185,225	

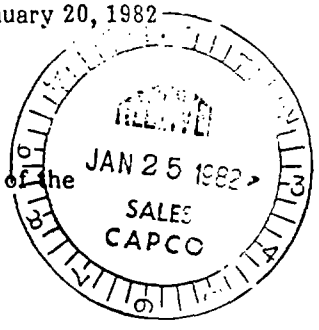
A/C Pipe
Producers Association
Public Affairs Committee
International Affairs Committee

Internal Correspondence

TO

FROM J. F. Welch, Vice President

DATE January 20, 1982



SUBJECT EPA Study on Asbestos Penetration of the Gastrointestinal Tract

REF: JFW correspondence, Study on Asbestos Fiber Penetration of the Gastrointestinal Tract, April 3, 1980.

ACTION REQUIRED: Review for information

The above reference, a research study entitled "Presence of Fibers in the Urine of a Baboon Gavaged with Chrysotile Asbestos," appeared to lend substantial credence to the theory that ingested asbestos fibers could penetrate the gastrointestinal tract. It supported similar findings in studies involving animals (Cunningham, et al, 1977, Brown and Storeygard, 1978, Sebastien, 1980) and humans (Cook and Olson, 1979). Such movement presumably enabled residence in the gastrointestinal tract and/or transport, via the circulatory or lymphatic system, to other organs.

The enclosed study attempted to duplicate the author's previous work by expanding asbestos analysis to a larger number of body sites. After sacrifice of the test animals, twenty-two tissues were collected for analysis. And in comparison to the first study, two types of asbestos fiber (chrysotile and crocidolite) were used rather than one. The researchers conclude:

> Given the above limitations, the low fiber counts observed in the present comprehensive study do not support the hypothesis that ingested asbestos fibers can penetrate the gastrointestinal tract of the baboon and migrate to various tissues.

It is interesting to note that research funded by the U.S. Environmental Protection Agency (EPA) also has failed to replicate the Cook and Olson findings of asbestos fibers in human urine. Preliminary results of a study using asbestos containing water from the Puget Sound Region of Washington State indicate that asbestos is not found in the urine of subjects consuming such water. These results and the recent Hallenbeck study thus raise serious doubts about the ability of fibers to penetrate the gastrointestinal tract. It is possible, indeed likely, that fiber size may be a governing factor in the penetration/migration mechanism i.e. only sub-micron length fibers are capable of such movement.

Staff Analysis

As experimental pathologist W. E. Smith, M.D. has pointed out on numerous occasions, the observation of asbestos penetration of the gastrointestinal tract is primarily one of theoretical interest. The real issue is not whether ingested fibers penetrate the gut and migrate to various body sites, but whether those fibers produce an adverse health effect. AACPP's position has been that penetration may be physiologically plausible, but has little relevance in the absence of disease. The

consistent finding of no increased incidence of tumors in animals fed asbestos and similar findings in human populations exposed to asbestos in drinking water continues to support the validity of this position.

Staff plans to prepare an "A/C Advisory" on this study. If you have any questions, please do not hesitate to call.

JFW/ajb

Enclosures

cc: A. Kahn, Esq.
N. Rahn, Esq.
Special Counsel (7)
Asbestos International Association
Asbestos Information Association/NA
W. E. Smith, M.D.
Brian Commins, Ph.D.

copies to:

Public Affairs Committee	International Affairs Committee
H. Olson	R. Dorner
B. Cook	E. van der Rest
R. LeGault	B. Giboin
W. Perrell	N. Officer
J. Woods	R. Hobbs
J. Baker	A. Saoulis
T. Dougherty	R. Jalan
D. Stinson	V. Pattabhi
W. McCallie	H. Hudson
B. Collier	C. Barton
	S. Al-Tarkait

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Chrono

ENVIRONMENTAL RESEARCH 25, 349–360 (1981)

Analyses of Tissue, Blood, and Urine Samples from a Baboon Gavaged with Chrysotile and Crocidolite Asbestos

WILLIAM H. HALLENBECK, DONALD R. MARKEY, AND DAVID G. DOLAN

School of Public Health, University of Illinois Medical Center, P.O. Box 6998, Chicago, Illinois 60680

Received September 15, 1980

Tissue, blood, and urine samples from one test and one control baboon were analyzed by transmission electron microscopy for the presence of chrysotile and crocidolite asbestos. The test animal had been gavaged with cumulative doses of 800 mg each of chrysotile and crocidolite asbestos. Urine and blood samples were collected during the 16-day gavage and 21-day postgavage period. Twenty-two tissues were collected at sacrifice. None of the analyses of tissue, blood, or urine samples from the test animal exceeded the expected level of background contamination for chrysotile. One crocidolite bundle was observed in a test sample. The results of this study indicate that asbestos fibers do not penetrate the gastrointestinal tract of the baboon and migrate to various tissues.

INTRODUCTION

The presence of asbestos fibers in drinking water supplies (Millette, *et al.*, 1980) has raised questions concerning human health: (1) Can ingested fibers penetrate into the wall of the gastrointestinal (GI) tract? (2) Can ingested fibers penetrate through the wall of the GI tract and migrate systemically? (3) Can the ingested fibers cause cancer in one or more target organs? Known sites of human cancer associated with asbestos exposure are the lung, GI tract, pleura, and peritoneum (Selikoff *et al.*, 1979). The extrapulmonary cancers observed by Selikoff *et al.* (1979) may have been related to inhaled asbestos fibers which were cleared from the lungs and swallowed. Some of the ingested fibers may have penetrated into and through the GI tract and caused cancer of the GI tract and peritoneum.

The following is a chronological summary of studies which have involved the ingestion of asbestos by animals or humans. Prior to 1976, animal ingestion studies provided inconclusive answers to the three questions raised above (Hallenbeck and Hesse, 1977). Several significant studies have been reported recently. Gut clearance experiments reported by Bolton and Davis (1976) demonstrated that the transit time for ingested chrysotile, crocidolite, and amosite was 72 hr in rats. Also, they reported that there was little or no evidence of fiber penetration into the gut wall and no sign of damage to the GI tracts of rats fed 250–300 mg/week of asbestos for up to 1 year. Gibel *et al.* (1976) produced a significant excess incidence of malignant tumors (kidney, lung, liver, and reticular cell) in rats fed 50 mg/kg-BW/day of a powdered filter material composed of 53% chrysotile and unspecified amounts of sulfate cellulose and condensation resin. Cunningham *et al.* (1977) presented evidence which indicated that ingested chrysotile fibers may penetrate

0013-9351/81/040349-12$02.00/0

through the GI tract of the rat and migrate to various sites (omentum, lung, kidney, and brain). Cook and Olson (1979) analyzed urine samples from residents of the Duluth area and found that levels of amphibole fibers in the urine corresponded to levels of exposure in drinking water. Kanarek *et al.* (1980) found associations between the chrysotile asbestos content of drinking water and human cancer of the lung, gall bladder, pancreas, peritoneum, esophagus, pleura, kidney, and stomach. However, they stated that their regression results must be viewed with caution due to low multiple correlation coefficients. A study by Sebastien *et al.* (1980) indicated that orally administered chrysotile and crocidolite was found in lymph fluid collected from the thoracic lymph duct of rats.

Recent studies indicated that orally administered chrysotile fibers may penetrate through the GI tract of the baboon and be recovered in the urine (Hallenbeck and Patel-Mandlik, 1979) and various tissues (Patel-Mandlik *et al.*, 1979; Patel-Mandlik and Millette, 1980). The limitations of these studies can be summarized as follows: (1) a relatively small number of fibers were observed in the urine and tissue preparations; (2) the time allowed for possible penetration and migration was relatively short (23 days in the urine study and 9 days in the tissue study); (3) urine samples were not collected during the 4-day gavage period, urine samples were collected on only 5 days during the 19-day postgavage period; (4) no blood samples were collected; (5) relatively low cumulative doses were administered (20 mg/kg-BW in the urine study and 30 mg/kg-BW in the tissue study); (6) only chrysotile asbestos was administered; (7) elevated fiber counts were not reproduced. Chrysotile is a relatively common environmental contaminant and as such is known to cause spurious laboratory results. Therefore, it is important to administer another type of asbestos which is not a common environmental contaminant.

The objective of the present baboon study was to improve upon the previous baboon studies and to determine whether the previous results could be corroborated.

MATERIALS AND METHODS

Two healthy female baboons (*Papio anubis*) were used. The test animal was 38 months old and weighed 7.9 kg; the control animal was 40 months old and weighed 6.3 kg. Prior to gavage, blood and urine samples were taken from each animal on each of 3 consecutive days. Analyses of these 12 pregavage samples for asbestos indicated that there was no detectable background level of asbestos imparted by the animals' environment. The gavage and postgavage period covered 37 days. During the 16-day gavage period, 200 mg of asbestos (100 mg of chrysotile and 100 mg of crocidolite) was administered to the test animal on approximately every second day. Food was withheld for 20 hr before gavage and for 3–4 hr after gavage. The dose was delivered by adding 30 ml of filtered water to a syringe containing 100 mg of each type of asbestos, injecting the suspension into a stomach tube, and rinsing the tube twice with 10 ml of water. On each day of gavage, the control animal received 50 ml of filtered water by stomach tube. The cumulative dose for the test animal was 101 mg/kg-BW of chrysotile (8 days × 100 mg/day/7.9 kg) and 101 mg/kg-BW of crocidolite. Characterization of the length and diameter distributions of the fibrils and bundles administered by stomach tube are given in Table 1. The length and diameter distributions were similar to those for

TABLE 1

CHARACTERIZATION OF THE LENGTHS AND DIAMETERS OF CHRYSOTILE AND CROCIDOLITE FIBERS[a] ADMINISTERED BY GAVAGE

Length (μm)	Chrysotile diameter (μm)						Crocidolite diameter (μm)				
	<0.021	0.021–0.043	0.044–0.064	0.065–0.115	>0.115	Total [%][b]	<0.065	0.065–0.115	0.116–0.165	>0.165	Total [%]
<0.2	2(0)[c]	23(0)	2(0)	4(0)	0(0)	31 [14]	0(0)	0(0)	0(0)	0(0)	0 [0]
0.2–0.5	3(0)	48(0)	4(0)	22(8)	2(1)	79 [35]	5(0)	14(1)	4(2)	1(0)	24 [18]
0.51–1.0	3(0)	31(1)	2(0)	18(7)	6(6)	60 [27]	6(0)	14(1)	12(2)	4(3)	36 [27]
1.1–2.0	3(0)	13(0)	5(1)	8(4)	7(5)	36 [16]	2(0)	9(1)	11(5)	8(7)	30 [22]
2.1–5.0	2(0)	4(0)	0(0)	3(1)	6(6)	15 [7]	0(0)	4(1)	13(7)	12(11)	29 [22]
>5.0	0(0)	1(0)	0(0)	1(0)	2(2)	4 [2]	0(0)	3(0)	4(2)	9(7)	16 [12]
Total [%]	13[6]	120[53]	13[6]	56[25]	23[10]	N[d] = 225	13[10]	44[33]	44[33]	34[25]	N = 135

	Chrysotile		Crocidolite	
	Length (μm)	Diameter (μm)	Length (μm)	Diameter (μm)
Maximum	34	3.4	34	0.42
Minimum	0.1	0.01	0.2	0.04
Mode	0.2	0.04	0.8	0.08
Median	0.5	0.04	1.2	0.13
Mean	1	0.07	2.7	0.13
SD	2.4	0.23	4.5	0.07
Geom. mean	0.5	0.05	1.4	0.11

[a] Fibers = fibrils and bundles of fibrils. The asbestos was provided by James Millette (USEPA, Cincinnati) and has been used in other animal studies such as the NIEHS "Study of Biological Effects of Ingested Asbestos" (Moore, 1978). A suspension was prepared by swirling a small amount of asbestos in 20 ml of filtered acetone and immediately placing a few μl on a carbon/Formvar-coated grid. The grid was scanned at a magnification of 21,000×.

[b] The proportion of the total number of fibers in a specified length or diameter interval is shown in brackets.

[c] Numbers in parentheses refer to the number of bundles included in the cell total, e.g., 10(2): there are 2 bundles in a total of 10 fibers. For bundles of chrysotile, the maximum lengths and diameters were recorded. The maximum lengths of crocidolite bundles were recorded. Unlike chrysotile bundles, the diameters of crocidolite bundles were uniform.

[d] N = Total number of fibers.

several U.S. water supplies (Millette *et al.*, 1980). In general, the crocidolite fibers used in this study were larger than the chrysotile fibers and were more likely to occur as bundles of fibrils (58% of the crocidolite fibers were greater than 0.115 μm in diameter versus 10% for chrysotile; 56% of the crocidolite fibers were greater than 1.0 μm in length versus 25% for chrysotile; 37% of crocidolite fibers occurred as bundles versus 19% for chrysotile). On each of the 8 days of gavage, 100 ml of urine and 2–5 ml of blood were collected from each animal. Urine and blood were collected during the same time intervals each day (9–11 AM). During urine collection procedures the baboons were anesthetized with Ketaset and Surital, and standard intravenous fluids were administered. Urine was obtained by a urinary catheter which drained into a closed container. Blood samples were obtained by venipuncture and were heparinized to prevent coagulation.

The usual diet of water, various fruits, and Ralston Purina Chow (No. 5037) was given throughout the study. The chow consisted of a biscuit with the following composition: minimum crude protein (15%), minimum crude fat (5%), maximum crude fiber (5%), maximum ash (5%), and maximum added minerals (3%).

During the 21-day postgavage period, urine and blood samples were collected on approximately every second day. On the 37th day of the experiment, the animals were sacrificed and the tissues and organs shown in Table 2 were collected.

Preparation of Urine Samples for Analyses by Transmission Electron Microscopy (TEM)

(1) 100 ml of urine was vacuum filtered through a 0.1-μm polycarbonate filter. The filter was covered during filtration and taken to dryness. Polycarbonate filters were used instead of cellulose acetate filters because the latter exploded in the low temperature plasma asher during the first few minutes of ashing.

(2) The filter was ashed in a low temperature plasma asher (13.56 MHz, 80 W, 1 Torr) for 30 min to 2 hr. To prevent possible cross-contamination, each sample was ashed separately. The asher was equipped with very sensitive air control valves which permitted extremely slow, nonturbulent attainment of either low or atmospheric pressure in the ashing chamber.

(3) The ash was suspended in 5 ml of prefiltered (0.2 μm Fluoropore) acetone by ultrasonication at 55 kHz for 1 min. Ultrasonication was necessary to insure release of fibers adsorbed to tiny unashed particles.

(4) Five microliters of the acetone was immediately withdrawn and deposited on a carbon/Formvar-coated 200-mesh TEM grid. Disposable pipettes were used to prevent cross-contamination among samples. Rapid evaporation of the acetone was effected by use of an infrared heat lamp (250 W) positioned 5 in. above the grid.

(5) For each urine preparation, ten grid openings were randomly selected from each of two grids and scanned using a Philips 300 TEM. Selection of random grid openings was facilitated by using grids with indexed grid openings. Grids were scanned for chrysotile and crocidolite at a magnification of 21,000×. The electron microscopist was provided with coded samples to prevent bias due to awareness of which preparations were from test or control animals. Identification was made

TABLE 2
SAMPLES ANALYZED FROM TEST AND CONTROL BABOONS

Hallenbeck and Patel-Mandlik (1979)	Patel-Mandlik and Millette (1980)	Present study
Urine[a]	Kidney medulla[b] (0.1)	Urine
	Kidney cortex[b] (1.8)	Blood
	Lymph nodes[b] (0.5)	Esophagus
	Spleen[b] (0.3)	Stomach
	Colon[b] (0.2)	Liver
	Esophagus[b] (0.2)	Gall bladder
	Stomach	Pancreas
	Liver	Duodenum
	Duodenum	Jejunum
	Cecum	Ileum
	Heart	Cecum
		Ascending colon
		Descending colon
		Rectum
		Urinary bladder
		Heart
		Spleen
		Peritoneum
		Kidney cortex and medulla
		Omentum
		Parietal pleura
		Lymph nodes—hilar, mediastinal, and mesenteric
		Lung—apical, diaphragmatic, and medial lobes with visceral pleura intact

[a] Positive results were characterized as preliminary.
[b] Positive results were characterized as significant at $P \leq 0.05$ even though the chrysotile fiber counts were low. Fibers per grid opening are given in parentheses.

utilizing primarily morphological characteristics. In most cases electron diffraction patterns were not obtained.

The sample preparation methodology described above yields a uniform distribution of fibers on the grid (Patel-Mandlik *et al.*, 1979). The occurrence of a cluster of fibrils and/or bundles may be indicative of an error in preparation (possibly from unusual background contamination) which resulted in a nonuniform distribution of fibers on the grid. Clustering can lead to serious over- or underestimates of fiber counts. Use of the above methodology produced clustering on only 11 (4%) of the 291 grids scanned. When a cluster was observed in a preparation of urine, blood, and tissue samples, additional grids were prepared from the original acetone suspension. When clustering was observed in a prepara-

tion of tissue samples, a second tissue sample was analyzed in addition to a reanalysis of the original acetone suspension.

Calculation of Fibers per Milliliter of Urine and Detection Limit

The term "fiber" as used in this paper means an individual fibril or bundle of fibrils.

$$\text{Fibers/ml of urine} = \frac{\bar{F} \times A_1 \times V_1}{V \times A_2 \times V_2} = \bar{F} \times 8.8 \times 10^3,$$

where

$\bar{F}$ = average number of fibers per grid opening,
A_1 = 0.07 cm^2 = area of a 200-mesh TEM grid,
A_2 = 8×10^{-5} cm^2 = area of one grid opening,
V_1 = 5 ml = volume of sample suspended in acetone,
V_2 = 0.005 ml = volume of sample suspension deposited on grid,
V = 100 ml = volume of urine collected and filtered.

The conversion factor of 8.8×10^3 is large. Therefore, it is extremely important that investigators are confident that a raw fiber count for a test sample is statistically significantly greater than that for a control sample. Statistical comparison of raw data will prevent the erroneous inflation of insignificant raw fiber counts, possibly due to ambient levels, by conversion to a per milliliter basis.

To calculate the detection limit, substitute the observation of only 1 fiber in 20 grid openings ($\bar{F} = 1/20 = 0.05$) to obtain 440 fibers/ml of urine as the minimum detectable concentration. The detection limit can be improved by decreasing V_1 or increasing V_2, V, or the number of grid openings scanned.

Preparation of Blood Samples for Analysis by TEM

(1) 1 ml of blood as added to 50 ml of prefiltered (0.4 μm polycarbonate) 1% potassium hydroxide, and the solution was placed in a water bath for 2 hr at 38°C.

(2) The digested blood was vacuum-filtered through a 0.4-μm polycarbonate filter. The filter was covered during filtration and taken to dryness.

(3) The rest of the procedure is the same as steps 2 through 5 for urine.

Calculation of Fibers per Milliliter of Blood and Detection Limit

$$\text{Fibers/ml of blood} = \frac{\bar{F} \times A_1 \times V_1}{V \times A_2 \times V_2} = \bar{F} \times 8.8 \times 10^5,$$

where all the definitions are the same as for urine except V = 1 ml. The detection limit is 4.4×10^4 fibers/ml of blood.

Preparation of Tissue Samples for Analysis by TEM

(1) Comparable wet weights (about 1 g) of control and test tissues were dried in a vacuum oven at 90°C for 24 hr. Lung samples were obtained by boring completely through a lobe with a cork borer (11.5 mm internal diameter). Three cor-

ings were taken from each of the three lobes of the right lung for both the test and control baboons. Corings from each lobe were pooled for further preparation. Tubular organs (esophagus, stomach, duodenum, jejunum, ileum, cecum, colon, and rectum) were longitudinally incised, and transverse sections were prepared for analysis. Random pieces of the other tissues were excised for preparation.

(2) 20 ml of prefiltered (0.4 μm polycarbonate) 5% potassium hydroxide was added to the dried tissue, and the mixture was placed in a water bath for 24 hr at 60°C.

(3) The digested tissue was vacuum filtered through a 0.4-μm polycarbonate filter. The filter was covered during filtration and taken to dryness.

(4) The rest of the procedure is the same as steps 2 through 5 for urine.

Calculation of Fibers per Milligram of Dry Tissue and Detection Limit

$$\text{Fibers/mg of dry tissue} = \frac{\bar{F} \times A_1 \times V_1}{M \times A_2 \times V_2} = \frac{\bar{F} \times 8.8 \times 10^5}{M},$$

where all the definitions are the same as for urine except M = dry weight of tissue in milligrams. The detection limit can be calculated by substituting the average value of M. For M = 200 mg, the detection limit is 220 fibers/mg of dry tissue.

RESULTS AND DISCUSSION

At the time of sacrifice, the esophagus and stomach of both baboons were inspected for evidence of irritation caused by the stomach tube and asbestos (test animal) or the stomach tube alone (control animal). No signs of irritation were observed. There was relatively little extraneous background material on the grids containing preparations of tissues and urine. However, the grids containing blood preparations had to be scanned slowly due to the relatively high level of extraneous background material present.

One crocidolite bundle was observed on a preparation of kidney cortex tissue taken from the test animal. This finding could not be repeated upon analysis of a second tissue preparation or reanalysis of the original acetone preparation. It is to be expected that a low count may not be reproduced. However, elevated counts of chrysotile must be reproduced, especially if clustering of fibers was observed, in order to dismiss the possibility of error in sample preparation (especially error due to unusually high background contamination). For example, the first analyses of preparations of four test tissues (jejunum, urinary bladder, peritoneum, and omentum), two test bloods, one control blood, and one control urine showed clustered chrysotile fiber counts which were elevated above the level of expected background contamination. The expected background contamination level, $\bar{F} \leq 0.55$, was derived from the control data in Tables 3 and 4. Note that all results from the control samples in Tables 3 and 4 are ≤ 11 ($\bar{F} \leq 0.55$). This low level of chrysotile was attributed to expected background contamination. The results of analyses of second tissue preparations and reanalyses of the original acetone preparations of the four tissues, three bloods, and one urine clearly indicated that the initial elevated counts were most likely due to unusually high background contamination. Therefore, the elevated counts from the first preparations were

not reported in Tables 3 and 4. If the attempts at replication of the elevated counts had been successful, a third method of confirmation would have been applied. This method would have involved analyses of several different weights for each type of sample. For example, if the analyses of several different dry weights of jejunum resulted in counts which increased in a roughly linear manner with increasing dry weight, it could be concluded that the elevated counts resulted from penetration and migration of gavaged asbestos.

The data in Tables 3 and 4 show that no chrysotile fiber counts above the expected background contamination level were observed for any of the urine, blood, or tissue preparations. Hence, the counts were not converted to a per milliliter or milligram basis. The chrysotile counts reported in Table 3 for blood include fibers which were significantly degraded in morphology. In most blood preparations, a high proportion of observed fibers were degraded.

The results of the present study are in agreement with the work of Bolton and Davis (1976) and in disagreement with that of Cook and Olson (1979) and Cunningham *et al.* (1977). In order to assess reasons for the disagreement, it is necessary to evaluate raw fiber counts. These data were not given in the papers of Cook and Olson (1979) and Cunningham *et al.* (1977).

It is important to compare the present study to the previous baboon studies carried out in our laboratory. Tables 2 and 5 contain a summary of appropriate

TABLE 3
RESULTS OF ANALYSES OF URINE AND BLOOD SAMPLES BY TEM

Time when samples were collected (day)	Total number of chrysotile fibers[a] observed in 20 grid openings			
	Urine		Blood	
	Test	Control	Test	Control
Gavage period				
1	0	1	0	0
2	0	0	0	2
5	1	1	4	0
7	1	0	3	0
9	1	0	1	0
12	6	7	3	0
14	5	2	0	1
16	7	1	5	0
Postgavage period				
20	1	1	1	5
22	2	11	2	1
26	3	4	5	0
28	3	2	0	1
30	2	3	4	7
33	0	3	10	2
35	0	5	3	1
37	0	0	0	6

Note. 100 ml of urine and 1 ml of blood were prepared for analysis.
[a] No crocidolite fibers were observed. Fiber = individual fibril or bundle of fibrils.

TABLE 4
RESULTS OF ANALYSES OF TISSUE SAMPLES BY TEM

Tissue	Total number of chrysotile fibers[a] observed in 20 grid openings	
	Test	Control
Esophagus	2 (130)[b]	3 (130)
Stomach	0 (160)	6 (130)
Liver	0 (370)	0 (230)
Gall bladder	0 (150)	0 (200)
Pancreas	0 (280)	2 (290)
Duodenum	1 (140)	0 (140)
Jejunum	0 (150)	0 (150)
Ileum	1 (170)	3 (120)
Cecum	3 (160)	0 (190)
Ascending colon	2 (160)	1 (270)
Descending colon	0 (200)	1 (240)
Rectum	1 (180)	0 (110)
Urinary bladder	1 (180)	0 (180)
Heart	1 (190)	4 (210)
Spleen	0 (220)	0 (260)
Peritoneum	2 (520)[c]	9 (410)[c]
Kidney cortex	0 (170)	0 (170)
Kidney medulla	0 (140)	0 (120)
Omentum	0 (810)	0 (820)
Parietal pleura	0 (170)	2 (60)
Lymph nodes[d]	0 (510)	2 (150)
Lung		
Right apical lobe	0 (140)	0 (150)
Right medial lobe	1 (160)	0 (160)
Right diaphragmatic lobe	0 (200)	0 (150)

[a] One crocidolite bundle was observed on a preparation of kidney cortex tissue taken from the test animal. However, this finding could not be reproduced.
[b] Dry weight (mg) shown in parentheses.
[c] This is a wet weight. The dry weight was inadvertently not recorded.
[d] The mass of lymph nodes collected at sacrifice was small. Therefore, the hilar, mediastinal, and mesenteric lymph nodes were combined for analysis.

comparisons. It can be seen that the present study embodies several improvements:

(1) Two types of asbestos were administered. If chrysotile alone were administered, a finding of high levels in the test and control samples would have compromised the experiment. Unlike chrysotile, crocidolite is not a general environmental contaminant. It is unlikely that crocidolite could contaminate test and control samples. Therefore, if both types of asbestos were administered and there was reason to suspect contamination by chrysotile, the results of the experiment could have been interpreted on the basis of crocidolite levels alone.

(2) The cumulative dose, 101 mg/kg-BW of each type of asbestos, was higher than the previous cumulative doses of 20 and 30 mg/kg-BW of chrysotile. There was significant overlap among the length distributions of chrysotile asbestos administered in all the baboon studies conducted in our laboratory.

TABLE 5
GENERAL COMPARISON OF BABOON STUDIES

	Hallenbeck and Patel-Mandlik (1979)	Patel-Mandlik and Millette (1980)	Present study
Age and sex of baboons used as test and control	2-year-old females	Neonate	3-year-old females
Method of dose administration	Stomach tube	Bottle	Stomach tube
Type of asbestos	Chrysotile	Chrysotile	Chrysotile and crocidolite
Cumulative dose (mg/kg-BW)	20	30	101 of each type of asbestos
Period over which dose was administered (days)	4	9	16
Number of days during dose period on which urine and blood samples were taken	0	0	8
Period after end of dose administration but before sacrifice (days)	19	0	21
Number of days during post administration period on which urine and/or blood samples were taken	5	0	8
Period after first dose but before sacrifice (days)	23	9	37
Detection limit (fibers/ml or mg)	440/ml urine	90/mg tissue	440/ml urine 4.4×10^4/ml blood 220/mg tissue
Highest level of chrysotile observed in a control preparation (fiber/grid opening)	0.7	0.16	0.55
Highest level of chrysotile observed in a test preparation (fibers/grid opening)	2.5	1.8	0.5
Were samples with elevated fiber counts reprepared to assess the role of background contamination with chrysotile?	No	No	Yes

(3) The time allowed for possible penetration and migration was greater (37 days versus 23 and 9).
(4) Urine samples were collected on 16 days versus 5.
(5) Blood samples were obtained.
(6) A greater variety of tissues were examined.

(7) Preparations which resulted in elevated fiber counts were checked for reproducibility.

There are several limitations to this study. The first is common to almost every asbestos study and is directly related to the detection limits for urine, blood, and tissues which were 440 fibers/ml, 4.4×10^4 fibers/ml, and 220 fibers/mg, respectively. These detection limits are relatively insensitive, and it is possible that penetration and migration may have gone undetected. Assuming for the moment that penetration and migration occurred, it is not possible from this study to infer the true value of $\bar{F}$ due to penetration and migration because the results of all analyses were in the range of expected background contamination. As an illustration of the effect of a low $\bar{F}$ on detection, assume that the true $\bar{F}$ due to penetration and migration was 0.05. The probability of detection when 20 grid openings were scanned was only 0.63 (Hallenbeck *et al.*, 1977). As the true $\bar{F}$ decreases, the probability of detection of penetration and migration decreases. The probability of detection can be increased by scanning more grid openings. For example, if 50 grid openings were scanned, the probability of detection would be 0.92 for a true $\bar{F} = 0.05$ (Hallenbeck *et al.*, 1977).

The second limitation is caused by the use of only one test animal. There may be individual differences among baboons which allow penetration and migration in some but not all members of a species. Penetration and migration may be a function of age and many other individual variables. Also, frequency of sampling is very important (Sebastien *et al.*, 1980). It is possible that more frequent sampling of the blood and urine may have resulted in the detection of fibers.

A final limitation is related to the length and diameter distributions of the types of asbestos administered. The possibility remains that administration of fibers with different length/diameter distributions may result in penetration and migration.

Given the above limitations, the low fiber counts observed in the present comprehensive study do not support the hypothesis that ingested asbestos fibers can penetrate the gastrointestinal tract of the baboon and migrate to various tissues.

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