Vinyl Chloride Carcinogenicity: An Experimental Model for Carcinogenesis Studies

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Long-term experimental bioassays of vinyl chloride (VC) carcinogenesis have supplied us with a body of knowledge concerning, on the one hand, the factors and mechanisms of carcinogenesis in general and, on the other, the value of experimental carcinogenesis bioassays in identifying oncogenic agents in the human environment, in assessing the degree of the risk, in predicting their effects on man, and in providing the basis for adequate measures of prevention. The achievements of these results have been favored, in our opinion, by the fact that these experimental studies have been performed by a single institution, under controlled conditions, which were kept uniform during the experiments with reference to material, type of animals and their care, methods of treatment, and the recording of biological effects.

The experimental project on VC carcinogenesis started at the Experimental Unit of the Bologna Institute of Oncology and Tumour Center on July 1, 1971. It included a series of 18 correlated and integrated experiments, started in sequence. At present, part of the experiments have already ended, but several are still going on. It is foreseen that the program will end by June 1978.

This report will consider mainly the terminated or the advanced experiments with significant results.

EXPERIMENTAL PROCEDURES

The experiments were planned to study the effects of VC, administered through different routes (inhalation, ingestion, and peritoneal and subcutaneous injection), at different concentrations, for varying periods of time, by continuous or intermittent treatment, on animals of different species (rats, mice, hamsters), strains (Sprague-Dawley and Wistar rats), sex, and age (adults, newborns, embryos).

The chambers of exposure are built basically of stainless steel and glass. They have been designed to deliver concentrations varying from 30,000 to 1 ppm of VC and to treat simultaneously nearly 4500 rodents. The VC concentration in the chambers is controlled by continuous gas chromatography.

The VC is supplied by Montedison. Each stock is analyzed in the Montedison Research Unit. The impurities and their maximal levels in the VC employed have been as follows:

H_2O	100 ppm
Acetic aldehyde	5 ppm
Acetylene	2 ppm
Allene	5 ppm
Butane	8 ppm
1,3-Butadiene	10 ppm
Chlorophene	10 ppm
Diacetylene	4 ppm
Vinyl acetylene	10 ppm
Propine	3 ppm
Methyl chloride	100 ppm

The experiments utilize mainly Sprague-Dawley rats; Wistar rats, Swiss mice, and hamsters have also been used. All the animals, except the hamsters, have been bred in our Institute for years, and, whatever their use, all are examined at death by complete autopsy, giving us extensive information concerning their current pathology.

The animals were weaned and classified by sex when 4–5 weeks old, at which time they were numbered by ear punch and divided into groups by litter distribution. From weaning, the animals have been fed, ad libitum, an adequate commercial diet. The animals are kept in groups of 5 in makrolon cages with tops made of stainless-steel wire, except those in the inhalation experiments, which, during the treatment period, are housed in groups of 10 in stainless-steel wire cages with solid bottoms of the same metal.

The animals are controlled weekly and weighed every 2 weeks during the period of treatment and monthly after the treatment is over. All the detectable gross pathological changes are recorded during the control. All the animals are kept under observation until spontaneous death. The animals, when moribund, are isolated in order to avoid cannibalism.

A complete autopsy is made on each animal. Histological examinations are performed on Zymbal glands, interscapular brown fat, salivary glands, tongue, lungs, liver, kidneys, spleen, stomach, different segments of the intestine, bladder, brain, bones of the legs and feet, and any other organ with pathological lesions.

All the animals exposed to the highest doses (30,000 and 10,000 ppm for 52 weeks), with or without tumors, were examined radiologically during treatment and/or at death; moreover, radiologic examinations have also been made on several animals bearing tumors even though these animals had been exposed to the lower doses.

The plan of the experiments is shown in Tables 1 to 7.[1]

[1] All Tables have been grouped at the end of this paper.

RESULTS

The results of our experiments have been reported in several previous papers (Maltoni 1974; Maltoni and Lefemine 1974a,b, 1975; Maltoni et al. 1974).

Inhalation Studies

Adult Sprague-Dawley Rats

EXPOSED FOR 52 WEEKS. VC produces on Sprague-Dawley rats Zymbal gland carcinomas, nephroblastomas, liver and other site angiosarcomas, subcutaneous angiomas, skin carcinomas, hepatomas, brain neuroblastomas, and mammary carcinomas (Tables 8 and 9).

In April 1974 we proved that VC was effective from 30,000 ppm down to 50 ppm. A dose–response relationship has been found. Moreover, the dose affects the prevalence of some tumors and, on the whole, the relative distribution of the various types of tumors (Table 10).

The carcinogenic effect of VC at the concentration of 50 ppm, under our experimental conditions, is now being re-proved definitively (Table 11).

As far as the doses below 50 ppm are concerned, the up-to-date review of our material, at 87 weeks from the start of the experiment, has proved that VC shows carcinogenic effects also at lower doses, namely at 25 ppm, with onset of liver angiosarcomas (Table 12).

EXPOSED FOR SHORTER PERIODS. The reduction of length of treatment sharply reduces the onset of several types of tumors, particularly of liver angiosarcomas (Table 13). When treated for only 5 weeks at 10,000 and 6000 ppm (BT10), no liver angiosarcomas were observed.

Sprague-Dawley Rats of Different Ages—Embryos and Newborns

VC has a transplacental effect. Exposure of breeders from days 12 to 18 of pregnancy is sufficient to produce VC-dependent tumors in offspring (Table 14).

The effect of age, however, is striking when comparing the onset of liver angiosarcomas and hepatomas in newborn and adult animals after 5 weeks of treatment at the same concentration (Table 15). The incidence of hepatomas among newborn rats is exceptionally high when compared with what we observed in all the other experiments of the project performed in adult animals.

Wistar Rats

In this strain of rats also, when exposed for 52 weeks at different concentrations, VC appears to produce broadly the same kinds of tumors it produces in Sprague-Dawley rats; however, the relative response of different organs and tissues appears to differ in the two strains (Table 16).

Swiss Mice and Golden Hamsters

VC exposure for 30 weeks, at different doses, produces in mice lung tumors, mammary carcinomas, liver angiosarcomas, vascular tumors of other types and/or site, and epithelial tumors of the skin (Table 17). It has proved to be effective at doses as low as 50 ppm.

In Golden hamsters, VC produces liver angiosarcomas, skin trichoepi-

theliomas, melanomas, and forestomach epithelial tumors; it also anticipated the onset of lymphomas, whose latency time is 48 weeks in treated animals and 82 weeks in controls (Table 18).

Ingestion Studies on Sprague-Dawley Rats

VC ingested by stomach tube for 52 weeks or more produces in Sprague-Dawley rats a large spectrum of the tumors it produces by inhalation (Table 19). No carcinogenic effects have been detected at lower doses after 57 weeks (Table 20).

Injection Studies on Sprague-Dawley Rats

One nephroblastoma and one subcutaneous angiosarcoma have been found among 240 animals which received from one to four intraperitoneal injections of 4.25 mg of VC.

One nephroblastoma has been observed among 75 animals which received a subcutaneous injection of 4.25 mg of VC.

Morphological Observations

The VC-dependent tumors are of the rare and current type. Extensive iconography has been given in previous publications (Maltoni and Lefemine 1974a; Maltoni et al. 1974).

Apart from tumors, other tissue changes have been observed in treated animals. We plan to review all our slides in order to record extraneoplastic lesions.

However, we want to point out here a current finding, i.e., vascular changes and correlated pathological lesions. In VC-treated animals, we observed in different anatomical sites vascular or sinusoidal ectasia and endothelial cell hyperplasia, dysplasia, and atypia (which may be considered the precursors of angioblastic tumors). Furthermore, in a high percentage of animals, local or systemic perivascular edema and hemorrhages were found. Hemorrhages are unusually frequent in the brain.

CONCLUSIONS

From the available data provided by long-term experimental bioassays, which have been partly presented here, and from the history of VC carcinogenesis, the following conclusions can be drawn:

1. VC is a multipotential carcinogen, meaning that it may produce tumors of different types at different sites. This fact and the increasing evidence from other carcinogenic models suggest that carcinogens in general should be considered multipotential.
2. The neoplastic response to VC depends largely on the species and strain of the animal used. If for some tumors the influence of the agent has been prominent, as is the case of liver angiosarcomas, for other types of tumors,

like Zymbal gland carcinomas and lung tumors among others, the species and strain of the animals have a determinant role.

3. The age of the animals is an extremely important factor in determining not only the general incidence of tumors, but also their relative distribution.
4. The dose affects, at least in given ranges, the incidence of tumors and their own relative distribution.
5. VC administered by inhalation has been found to produce angiosarcomas of the liver in rats at concentrations as low as 25 ppm.
6. The carcinogenicity of VC in man, as well as the most important target organs, have been predicted by experimental bioassays. All human tumors which at present have been correlated to the exposure to VC had been predicted by experimental evidence (Table 21).

   To our knowledge, 63 liver angiosarcomas in workers in the VC-PVC industry have been documented throughout the world (Table 22). Epidemiologic data had exposed a high incidence of brain tumors, large-cell carcinomas of the lung, lymphomas, and leukemias. Early cases of hepatomas are also being found among exposed workers. A cytological examination of sputa from nearly 4000 Italian workers in the VC-PVC industry has shown a high incidence of changes in bronchial epithelia, including squamous dysplasia and atypical adenomatous proliferation.
7. Finally, experimental bioassays have provided indications on the level of the risk that have led to the adoption of new rules and measures of prevention.

We wish, as we did in the past, that proper experimental bioassays would be performed, following a list of priorities, on all the agents already produced, used, and widespread in the human environment, and on the new ones destined for production.

In our Institute, we now have under study several compounds that are widely produced and used.

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Table 1
Plan of the Experiments on Vinyl Chloride Oncogenesis: Effects of Inhalation of Different Doses for 1 Year

		Animals: Sprague-Dawley rats				
			no.			
Exp. no.	*Treatment:[a] doses VC*	*age (weeks)*	♀	♂	*total*	*per group*
BT1	10,000, 6000, 2500, 500, 250, 50 ppm; untreated controls; treated controls: VA 2500 ppm	13	268	309	577	64–96
BT2	200, 150, 100 ppm; untreated controls	13	280	265	545	120–185
BT6	30,000 ppm	17	30	30	60	60
BT9	50 ppm; untreated controls	11	200	200	400	100–300
BT15	25, 10, 5, 1 ppm; untreated controls	13	300	300	600	120

[a] The duration of treatment in each case was 4 hr daily, 5 days weekly.

Table 2
Plan of the Experiments on Vinyl Chloride Oncogenesis: Effects of Length of Exposure by Inhalation

			Animals: Sprague-Dawley rats				
	Treatment			*no.*			
Exp. no.	*doses VC*	*duration*	*age (weeks)*	♀	♂	*total*	*per group*
BT3	10,000, 6000, 2500, 500, 250, 50 ppm; untreated controls	4 hr daily, 5 days weekly, 17 weeks	21	262	288	550	60–190
BT10	10,000, 6000 ppm; untreated controls	4 hr daily, 5 days weekly, 5 weeks; 4 hr daily, 1 day weekly, 25 weeks; 1 hr daily, 4 days weekly, 25 weeks	11	420	420	840	120

Table 4
Plan of the Experiments on Vinyl Chloride Oncogenesis: Effects of Strain

		Animals: Wistar rats				
			no.			
Exp. no.	*Treatment:[a] doses VC*	*age (weeks)*	♀	♂	*total*	*per group*
BT7	10,000, 6000, 2500, 500, 250, 50, ppm; untreated controls	11	—	220	220	30–40
BT17	1 ppm; untreated controls	13	120	120	240	120

[a] The route in both cases was by inhalation, and the duration of treatment was 4 hr daily, 5 days weekly, 52 weeks.

Table 5
Plan of the Experiments on Vinyl Chloride Oncogenesis: Effects of Species

		Animals						
					no.			
Exp. no.	*Treatment:[a] doses VC*	*species*	*strain*	*age (weeks)*	♀	♂	*total*	*per group*
BT4	10,000, 6000, 2500, 500, 250, 50 ppm; untreated controls	mouse	Swiss	11	250	260	510	60–150
BT8	10,000, 6000, 2500, 500, 250, 50 ppm; untreated controls	hamster	Golden	11	—	268	268	32–70

[a] The route in both cases was by inhalation, and the duration of treatment was 4 hr daily, 5 days weekly, 30 weeks.

Table 6
Plan of the Experiments on Vinyl Chloride Oncogenesis: Experiments by Ingestion

			Animals: Sprague-Dawley rats				
	Treatment				*no.*		
Exp. no.	*doses VC*	*duration*	*age (weeks)*	♀	♂	*total*	*per group*
BT11	50 mg, 16.65 mg, 3.33 mg/kg body weight in olive oil; controls: olive oil	5 times weekly, 52 weeks	13	160	160	320	80
BT27	1 mg, 0.3 mg, 0.03 mg/kg body weight in olive oil; controls: olive oil	5 times weekly, 52 weeks or more	10	300	300	600	150

Table 7
Plan of the Experiments on Vinyl Chloride Oncogenesis: Experiments by Injection

Exp. no.	*Treatment*			*Animals: Sprague-Dawley rats*				
					no.			
	route	*doses VC*	*duration*	*age (weeks)*	♀	♂	*total*	*per group*
BT12	endoperitoneal	4.25 mg in 1.0 ml olive oil; controls: 1.0 ml olive oil	4, 3, 2 times by two months and once	13	150	150	300	60
BT13	subcutaneous	4.25 mg in 1.0 ml olive oil; controls: 1.0 ml olive oil	1 injection	21	80	70	150	75

Table 8
Experiment BT1: Results after 135 Weeks (End of Experiment)

Group, treatment	Animals (S-D rats) total	Animals (S-D rats) corrected no.[a]	Animals with tumors: Zymbal gland carcinomas[b] no.	%	av. latency (weeks)	nephroblastomas[c] no.	%	av. latency (weeks)	angiosarcomas: liver[d] no.	%	av. latency (weeks)	other sites no.	subcutaneous angiomas no.	skin carcinomas no.	hepatomas no.	brain neuroblastomas no.	mammary carcinomas no.	other type and/or site[k] no.	total[n] no.
I VA 2500 ppm	96	49	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
II VC 10,000 ppm	69	61	16	26	50	5	8	59	9	15	64	3[e]	4	3	1	7	3	4[l]	38
III VC 6000 ppm	72	60	7	12	62	4	7	65	13	22	70	3[f]	3	1	1	3	—	8[m]	31
IV VC 2500 ppm	74	59	2	3	33	6	10	74	13	22	78	3[g]	3	1	2	5	1	3[n]	32
V VC 500 ppm	67	59	4	7	79	4	7	83	7	12	81	2[h]	1	1	4	—	1	3[o]	22
VI VC 250 ppm	67	59	—	—	—	6	10	80	4	7	79	2[i]	—	4	—	—	1	2[p]	16
VII VC 50 ppm	64	59	—	—	—	1	2	135	1	2	135	1[j]	1	1	—	—	2	7[q]	10
VIII No treatment	68	58	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	10[r]	6
Total	577	464	29	—	—	26	—	—	47	—	—	14	12	11	8	15	8	37	155

[illegible] 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

[a] Animals alive after 26 weeks, when the first tumor (a Zymbal gland carcinoma) was observed. The percentages refer to the corrected number.

[b] Metastases to lung.

[c] Metastases to liver, lung, spleen, and brain.

[d] Metastases to lung.

[e] One intra-abdominal angiosarcoma (next to liver); 1 angiosarcoma of the lips; 1 angiosarcoma of the nose.

[f] One angiosarcoma in subcutaneous fibrosing angioma; 1 ossifying parauricular angiosarcoma; 1 intra-abdominal angiosarcoma (next to liver).

[g] One ossifying angiosarcoma of neck; 2 intra-abdominal angiosarcomas (1 next to spleen and 1 next to ovary).

[h] One angiosarcoma of uterus; 1 lung angiosarcoma.

[i] One intra-abdominal angiosarcoma (next to spleen); 1 intrathoracic ossifying angiosarcoma.

[j] One intra-abdominal diffused angiosarcoma.

[k] Several cases of breast fibroadenomas; adrenal and pituitary tumors (generally adenomas) have not been considered since their distribution in the different groups does not vary significantly.

[l] Two Zymbal gland adenomas; 1 ovarian cystoadenocarcinoma; 1 neurilemmoma.

[m] Four Zymbal gland adenomas; 2 hepatic angiomas; 1 peritoneal angioma; 1 salivary gland adenocarcinoma.

[n] One Zymbal gland adenoma; 2 ependymomas.

[o] One pulmonary fibrosarcoma; 2 lymphomas.

[p] One Zymbal gland adenoma; 1 lymphoma.

[q] Three Zymbal gland adenomas; 1 subcutaneous angiopericitoma; 3 uterine adenocarcinomas (1 with sarcomatous component).

[r] One invasive acanthoma of Zymbal gland; 1 subcutaneous fibrosarcoma; 2 peritoneal fibroangiomas; 2 uterine adenocarcinomas (1 with sarcomatous component); 1 uterine leiomyosarcoma; 1 ovarian fibrosarcoma; 1 pulmonary rhabdomyosarcoma; 1 lymphoma.

[s] Several animals with two or more tumors.

Table 9
Experiment BT2: Results after 143 Weeks (End of Experiment)

Group, treatment	Animals (S-D rats)	No. of animals with tumors: Zymbal gland carcinomas	nephroblastomas	angiosarcomas: liver	angiosarcomas: other sites	other type and/or site	total[h]
I VC 200 ppm	120	1	3	12	1[a]	23[d]	30
II VC 150 ppm	120	2	7	5	3[b]	17[e]	29
III VC 100 ppm	120	1	10	1	—	12[f]	20
IV No treatment	185	1	—	—	1[c]	20[g]	21
Total	545	5	20	18	5	72	100

Exposure by inhalation to VC in air at 200, 150, and 100 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

[a] One intra-abdominal angiosarcoma.

[b] Two subcutaneous angiosarcomas; 1 intra-abdominal angiosarcoma.

[c] One subcutaneous angiosarcoma.

[d] Five skin carcinomas; 1 subcutaneous fibroangioma; 4 mammary carcinomas; 2 liver fibroangiomas; 3 liver angiomas; 3 liver hepatomas; 1 liver colangioma; 1 intra-abdominal fibroangioma; 1 leimyosarcoma of uterus; 1 neurilemmoma; 1 orbital fibrosarcoma.

[e] Three skin carcinomas; 5 mammary carcinomas; 1 thymus angioma; 1 nasal osteosarcoma; 1 cranial osteosarcoma; 1 liver colangioma; 2 adenocarcinomas of uterus; 1 fibromixosarcoma of uterus; 1 fibrosarcoma of lung; 1 lymphoma.

[f] One skin carcinoma; 1 subcutaneous fibrosarcoma; 3 mammary carcinomas; 1 intra-abdominal angioma; 3 forestomach papillomas; 1 adenocarcinoma of uterus; 1 intra-abdominal rabdomyosarcoma; 1 kidney adenoma.

[g] One Zymbal gland adenoma; 1 skin adenocarcinoma; 1 skin sebaceous adenoma; 1 subcutaneous fibrosarcoma; 2 subcutaneous fibromixosarcomas; 1 mammary carcinoma; 1 intrathoracic rabdomyosarcoma; 3 forestomach papillomas; 1 adrenal carcinoma; 3 adenocarcinomas of uterus; 1 fibrosarcoma of uterus; 1 fibrosarcoma of scrotum; 3 lymphomas.

[h] Several animals with two or more tumors.

Table 10
Experiment BT6: Results after 63 Weeks (End of Experiment)

Group, treatment	Animals (S-D rats)		Animals with tumors							
			Zymbal gland carcinomas			nephroblastomas	angiosarcomas		other type and/or site	
	total	corrected no.[a]	no.	%	av. latency (weeks)	no.	liver no.	other sites no.	no.	total[d] no.
I										
VC 30,000 ppm	60	60	31	52	43	—	17	1[b]	27[c]	51

Exposure by inhalation to VC in air at 30,000 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

[a] Animals alive after 24 weeks, when the first tumor (a Zymbal gland carcinoma) was observed. The percentages refer to the corrected number.

[b] One lung angiosarcoma.

[c] Seven Zymbal gland adenomas; 1 skin carcinoma; 1 subcutaneous fibroangioma; 2 mammary carcinomas; 1 liver angioma; 1 hepatoma; 11 forestomach papillomas; 1 ovarian angioma; 1 brain neuroblastoma; 1 Harderian gland carcinoma.

[d] Several animals with two or more tumors.

Table 11
Experiment BT9: Results after 100 Weeks

Groups, treatment	Animals (S-D rats) total	Animals (S-D rats) survivors	No. of animals with tumors: Zymbal gland carcinomas	nephroblastomas	angiosarcomas: liver	angiosarcomas: other sites	mammary tumors: total	mammary tumors: histological examination	mammary tumors: carcinomas	mammary tumors: other malignant tumors	mammary tumors: fibroadenomas	other type and/or site	totals
I													
VC 50 ppm	300	55	6[a]	1	6	7[b]	111	90	43	2[c]	45	28[e]	128
II													
No treatment	100	28	—	1	—	—	20	8	3	2[d]	3	7[f]	24
Total	400	83	6	2	6	7	131	98	46	4	48	35	152

Exposure by inhalation to VC in air at 50 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

[a] One with sarcomatous component.

[b] Two subcutaneous angiosarcomas; 1 mammary angiosarcoma; 1 heart angiosarcoma; 1 thymus angiosarcoma; 1 abdominal angiosarcoma (next to spleen); 1 ovarian angiosarcoma.

[c] Two carcinosarcomas.

[d] Two carcinosarcomas.

[e] Two skin carcinomas; 3 subcutaneous fibrosarcomas; 3 subcutaneous rabdomyosarcomas; 1 subcutaneous leiomyosarcoma; 1 subcutaneous mixed sarcoma; 1 subcutaneous fibroangioma; 1 liver angioma; 1 thymus fibroangioma; 1 mediastinal fibroangioma; 1 spleen fibroangioma; 1 uterine fibroangioma; 2 uterine adenocarcinomas; 2 hypophysis adenomas in malignant deviation; 1 Leydig cells tumor; 1 ovarian fibroma; 1 retroperitoneal liposarcoma; 1 osteosarcoma; 4 leukemias.

[f] One subcutaneous fibrosarcoma; 1 subcutaneous angiopericitosarcoma; 2 uterine fibrosarcomas; 3 leukemias.

[g] Several animals with two or more tumors.

Table 12
Experiment BT15: Results after 87 Weeks

Group, treatment	Animals (S-D rats)		No. of animals with tumors						
	total	survivors	Zymbal gland carcinomas	nephroblastomas	angiosarcomas: liver	angiosarcomas: other sites	mammary carcinomas	other type and/or site	total
I VC 25 ppm	120	45	3	—	3	—	10	7	20
II VC 10 ppm	120	51	1	—	—	—	11	5	16
III VC 5 ppm	120	62	—	—	—	—	13	5	17
IV VC 1 ppm	120	49	—	—	—	—	8	4	12
V No treatment	120	49	—	—	—	—	2	4	6
Total	600	256	4	—	3	—	44	25	71

Exposure by inhalation to VC in air at 25, 10, 5, and 1 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

Table 13
Experiment BT3: Results after 155 Weeks (End of Experiment)

Group, treatment	Animals (S-D rats) total	Animals (S-D rats) survivors	No. of animals with tumors[a] Zymbal gland carcinomas	nephroblastomas	angiosarcomas liver	angiosarcomas other sites	brain neuroblastomas	other type and/or site	total[a]
I VC 10,000 ppm	60	—	7 (16)	1 (5)	— (9)	1[b] (3)	6 (7)	7[b] (18)	21 (38)
II VC 6000 ppm	60	—	6 (7)	1 (4)	1 (13)	1[c] (3)	2 (3)	11[i] (13)	17 (31)
III VC 2500 ppm	60	—	3 (2)	2 (6)	1 (13)	2[d] (3)	2 (5)	6[j] (11)	13 (32)
IV VC 500 ppm	60	—	1 (4)	— (4)	1 (7)	1[e] (2)	—	3[k] (10)	6 (22)
V VC 250 ppm	60	—	1	2 (6)	— (4)	— (2)	—	5[l] (8)	7 (16)
VI VC 50 ppm	60	—	—	1 (1)	— (1)	2[f] (1)	—	5[m] (13)	8 (10)
VII No treatment	190	2	1	—	—	1[g]	—	12[n] (10)	12 (6)
Total	550	2	19 (29)	7 (26)	3 (47)	8 (14)	10 (15)	49 (83)	84 (155)

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 17 weeks.

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 17 weeks.

[a] The number of tumors found in experiment BTf after 135 weeks is shown in parentheses.

[b] One subcutaneous angiosarcoma.

[c] One intra-abdominal angiosarcoma.

[d] One subcutaneous angiosarcoma; 1 intra-abdominal angiosarcoma.

[e] One intra-abdominal angiosarcoma.

[f] One subcutaneous ossifying angiosarcoma; 1 orbital angiosarcoma.

[g] One ovarian angiosarcoma.

[h] Three skin carcinomas; 1 skin papilloma (nose); 1 renal adenoma; 1 lung angioma; 1 parauricular fibrosarcoma.

[i] One Zymbal gland fibroangioma; 5 skin carcinomas; 1 subcutaneous angioma; 1 mammary carcinoma; 1 forestomach papilloma; 1 adenocarcinoma of uterus; 1 cranial osteoma.

[j] Two skin carcinomas; 1 subcutaneous angioma; 1 mammary carcinoma; 1 hepatoma; 1 adenocarcinoma of uterus.

[k] One ovarian carcinoma; 2 lymphomas.

[l] One Zymbal gland adenoma; 1 mammary carcinoma; 1 adenocarcinoma of uterus; 1 Leydig cells tumor; 1 retrobulbar fibroma.

[m] One skin papilloma (nose); 1 mammary carcinoma; 3 adenocarcinomas of uterus.

[n] One Zymbal gland adenoma; 1 skin carcinoma; 3 subcutaneous fibrosarcomas; 1 subcutaneous leiomyosarcoma; 1 mammary carcinoma; 1 ovarian gynandroblastoma; 1 fibrosarcoma of uterus; 1 salivary adenocarcinoma; 2 lymphomas.

[o] Several animals with two or more tumors.

Table 14
Experiment BT5: Results after 143 Weeks (End of Experiment)

Group, treatment	Animals (S-D rats) total	Animals (S-D rats) corrected no.	No. of animals with tumors: Zymbal gland carcinomas	nephroblastomas	angiosarcomas: liver	angiosarcomas: other sites	other type and/or site	total
I VC 10,000 ppm breeders	30	30	1	—	—	1[b]	2[c]	3[h]
II VC 6000 ppm breeders	30	30	—	—	—	—	—	—
III VC 10,000 ppm offspring	54	51	3	1	—	2[e]	2[f]	8
IV VC 6000 ppm offspring	32	32	1	—	—	2[d]	4[g]	6[h]
Total	146	143	5	1	—	5	8	17

Exposure by inhalation to VC in air at 10,000 and 6000 ppm of breeders, 4 hr daily, for 1 week (from 12th to 18th day of pregnancy).

[a] Animals alive after 22 weeks, when the first tumor (a subcutaneous angiosarcoma) arose in an offspring.

[b] One intra-abdominal angiosarcoma.

[c] One subcutaneous angiosarcoma; 1 angiosarcoma of the leg.

[d] One subcutaneous angiosarcoma; 1 intra-abdominal angiosarcoma.

[e] One liver fibroangioma; 1 liver angioma.

[f] One Zymbal gland fibrosarcoma; 1 ovarian leiomyosarcoma.

[g] One Zymbal gland adenoma; 1 skin carcinoma; 1 subcutaneous fibroangioma; 1 mammary carcinoma.

[h] One animal with two tumors.

Table 15
Incidence of Hepatic Tumors among Sprague-Dawley Rats Exposed to VC: Results after 104 Weeks

Exp. no.	Group	No. of animals		Liver tumors		
		total	survivors	angiosarcomas	hepatomas	total animals with liver tumors
BT10	I					
	VC 10,000 ppm	120	16	—	1	
	II					
	VC 6000 ppm	120	15	—	—	
	III					
	no treatment	249	55	—	—	
BT14	I					
	VC 10,000 ppm	46	8	10	15	19
	II					
	VC 6000 ppm	43	5	10	13	17

Exposure in air at 10,000 and 6000 ppm, 4 hr daily, 5 days weekly, for 5 weeks, at different ages: 13 weeks (BT10) and 1 day (BT14).

Table 16
Experiment BT7: Results after 136 Weeks

Group, treatment	Animals (Wistar rats) total	Animals (Wistar rats) survivors	No. of animals with tumors[a] Zymbal gland carcinomas	nephroblastomas	angiosarcomas liver	angiosarcomas other sites	brain neuroblastomas	other type and/or site	total[b]
I VC 10,000 ppm	30	—	1 (10)	1 (3)	8 (4)	— (2)	1 (2)	2[c] (7)	11 (20)
II VC 6000 ppm	30	—	— (3)	3 (3)	2 (3)	1[b] (1)	1 (2)	5[f] (4)	8 (13)
III VC 2500 ppm	30	—	— (1)	— (5)	3 (6)	1[e] (2)	— (2)	2[g] (2)	4 (16)
IV VC 500 ppm	30	—	— (3)	1 (2)	4	—	—	2[h]	7 (6)
V VC 250 ppm	30	—	—	— (2)	1 (2)	1[d] (1)	—	4[i] (3)	4 (6)
VI VC 50 ppm	30	—	—	—	—	— (1)	—	1[j] (1)	1 (2)
VII No treatment	40	3	—	—	—	—	—	4[k]	4
Total	220	3	1 (17)	5 (15)	18 (15)	3 (7)	2 (6)	19 (17)	39 (63)

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 52 weeks.

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 52 weeks.
[a] The number of tumors found in experiment BT1 after 135 weeks is shown in parentheses.
[b] One intra-abdominal angiosarcoma (next to spleen).
[c] One angiosarcoma of the interscapular fat pad.
[d] One scrotal angiosarcoma.
[e] Two Zymbal gland adenomas.
[f] Two liver angiomas; 1 hepatoma; 1 testicular mesothelioma; 1 thymus fibroangioma.
[g] One skin sebaceous adenoma (eye); 1 Leydig cells tumor.
[h] One liver fibroangioma; 1 intra-abdominal fibromixosarcoma.
[i] One skin carcinoma; 1 skin acanthoma; 1 angioma of the caecum; 1 lymphoma.
[j] One lymphoma.
[k] One pericitosarcoma; 1 Leydig cells tumor; 2 lymphomas.
[l] Several cases with two or more tumors.

Table 17
Experiment BT4: Results after 81 Weeks (End of Experiment)

Group, treatment	Animals (Swiss mice) total ♂	♀	♂ ♀	corrected number[a] ♂	♀	♂ ♀	Animals with tumors: lung tumors[b] no.	%	av. latency (weeks)	mammary carcinomas[c] no.	%	av. latency (weeks)	liver angiosarcomas no.	other vascular no.	epithelial tumors of the skin no.	other type and/or site no.	total[w] no.
I VC 10,000 ppm	30	30	60	22	28	50	35	70	36	13	47	31	8	9[d]	3[k]	2[p]	36
II VC 6000 ppm	30	30	60	26	28	54	38	70	38	8	28	33	5	9[e]	6[l]	4[q]	39
III VC 2500 ppm	30	30	60	23	30	53	30	57	43	9	30	35	11	12[f]	3[m]	2[r]	31
IV VC 500 ppm	30	30	60	29	29	58	38	66	41	7	24	37	11	18[g]	1[n]	1[s]	43
V VC 250 ppm	30	30	60	29	29	58	33	57	45	11	32	39	11	22[h]	2[o]	3[t]	40
VI VC 50 ppm	30	30	60	27	30	57	2	3.5	51	12	33	43	1	13[i]	—	1[u]	18
VII No treatment	80	70	150	74	67	141	8	6	64	—	—	—	—	1[j]	—	7[v]	13
Total	260	250	510	230	241	471	184	—	—	60	—	—	47	84	15	20	220

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 30 weeks.
a Animals alive after 16 weeks when the first tumor (a mammary carcinoma) was observed. The percentages refer to the corrected number.

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 30 weeks.

[a] Animals alive after 16 weeks, when the first tumor (a mammary carcinoma) was observed. The percentages refer to the corrected number.

[b] Adenomas, some of which undergoing malignant transformation.

[c] In females.

[d] Three subcutaneous angiomas; 4 liver fibroangiomas; 1 heart fibroangioma; 1 ossifying interscapular angioma.

[e] One subcutaneous angiosarcoma; 2 liver fibroangiomas; 4 liver angiomas; 1 renal fibroangioma; 1 thymus angioma.

[f] Three subcutaneous angiosarcomas; 1 subcutaneous angioma; 3 liver angiomas; 2 intra-abdominal angiosarcomas; 2 renal angiosarcomas; 1 lung angioma.

[g] Two subcutaneous angiosarcomas; 1 subcutaneous fibroangioma; 1 subcutaneous angioma; 4 liver fibroangiomas; 1 liver angioma; 3 intra-abdominal angiosarcomas; 1 intra-abdominal fibroangioma; 1 renal angiosarcoma; 1 testicular fibroangioma; 1 angioma of the caecum; 1 lung fibroangioma; 1 lung angioma.

[h] One subcutaneous angioma; 6 liver fibroangiomas; 10 liver angiomas; 2 intra-abdominal angiosarcomas; 1 ovarian angioma; 1 scrotal angioma; 1 lung angioma.

[i] One subcutaneous angiosarcoma; 1 subcutaneous fibroangioma; 2 subcutaneous angiomas; 2 liver fibroangiomas; 3 liver angiomas; 1 intra-abdominal angioma; 1 ovarian angioma; 1 intrathoracic fibroangioma; 1 angioma of the interscapular fat pad.

[j] One angiosarcoma of uterus.

[k] Two squamous carcinomas; 1 invasive acanthoma.

[l] Five squamous carcinomas; 1 acanthoma.

[m] One squamous carcinoma; 2 acanthomas.

[n] One acanthoma.

[o] One skin adenocarcinoma; 1 basalioma.

[p] One Zymbal gland adenoma; 1 forestomach papilloma.

[q] One subcutaneous leiomyosarcoma; 1 forestomach papilloma; 1 Harderian gland adenoma; 1 lymphoma.

[r] One forestomach papilloma; 1 parotid gland mixed tumor.

[s] One Zymbal gland adenoma.

[t] One Zymbal gland adenoma; 1 Leydig cells tumor; 1 lymphoma.

[u] One parotid gland adenocarcinoma.

[v] One subcutaneous leiomyosarcoma; 1 adenoma of colon; 1 ovarian carcinoma; 1 leiomyosarcoma of uterus; 3 lymphomas.

[w] Several cases with two or more tumors.

Table 18
Experiment BT8: Results after 109 Weeks (End of Experiment)

Group, treatment	Animals (Golden hamsters)	No. of animals with tumors						
		liver angiosarcomas	skin trichoepitheliomas and basaliomas[a]	melanomas	lymphomas	forestomach epithelial[b] tumors	other type and/or site	total[g]
I VC 10,000 ppm	35	—	6	1	—	4	2[c]	10
II VC 6000 ppm	32	1	2	2	2	7	7[d]	10
III VC 2500 ppm	33	—	1	1	1	11	3[e]	13
IV VC 500 ppm	33	2	4	—	1	7	2[f]	12
V VC 250 ppm	32	—	3	—	1	2	—	6
VI VC 50 ppm	33	—	6	1	1	4	—	10
VII No treatment	70	—	2	—	2	2	—	7
Total	268	3	24	5	8	37	14	68

Exposure by inhalation to VC in air at 10,000, 6000, 2500, 500, 250, and 50 ppm, 4 hr daily, 5 days weekly, for 30 weeks.

[a] Several cases with acanthosis and some undergoing malignant transformation.
[b] Papillomas, acanthomas, some of which undergoing malignant transformation.
[c] One subcutaneous angioma; 1 gall bladder adenocarcinoma.
[d] Two hepatomas; 2 liver fibroangiomas; 2 liver angiomas; 1 biliducts adenocarcinoma.
[e] One hepatoma; 1 liver fibroangioma; 1 liver angioma.
[f] One subcutaneous angioma; 1 bronchial carcinoma.
[g] Several animals with two or more tumors.

Table 19
Experiment BT11: Results after 120 Weeks

Group, treatment	Animals (S-D rats)		No. of animals with tumors						
			Zymbal gland carcinomas	nephroblastomas	angiosarcomas		mammary carcinomas	other type and/or site	totals[g]
	total	survivors			liver	other sites			
VC 50.00 mg/kg I	80	2	1	2	16	2[a]	5	7[e]	29
VC 16.65 mg/kg II	80	2	2	3	9	—	6	3[d]	21
VC 3.33 mg/kg III	80	4	—	—	—	2[b]	4	3[e]	9
Control: olive oil IV	80	4	1	—	—	—	4	5[f]	9
Total	320	12	4	5	25	4	19	18	68

Exposure by ingestion (stomach tube) to VC in olive oil, at 50.00, 16.65, and 3.33 mg/kg body weight, once daily, 4–5 days weekly, for 52 weeks.

[a] One thymus angiosarcoma; 1 intra-abdominal angiosarcoma (next to spleen).

[b] One lung angiosarcoma; 1 intra-abdominal angiosarcoma (next to kidney).

[c] One skin sebaceous carcinoma; 1 hepatoma; 1 liver angioma; 2 forestomach papillomas; 1 intestinal adenocarcinoma; 1 lymphoma.

[d] One liver angioma; 1 adrenal carcinoma; 1 lymphoma.

[e] One subcutaneous sarcoma with angioblastic component; 1 intra-abdominal ossifying sarcoma with angioblastic component; 1 bladder papilloma.

[f] One skin carcinoma; 1 papilloma of the auditory duct; 1 bladder carcinoma; 1 adrenal carcinoma; 1 reticulosarcoma.

[g] Several animals with two tumors.

Table 20
Experiment BT27: Results after 57 Weeks

Group no.	Treatment: dose	Treatment: length (weeks)	Treatment: ongoing	Animals[a]: no. at start: ♂	no. at start: ♀	no. at start: total	survivors: ♂	survivors: ♀	survivors: total	present age (weeks)	Tumors: mammary tumors: total	mammary tumors: histologically examined: fibroadenomas	mammary tumors: histologically examined: carcinomas	Tumors: others
I	1 mg	57	+	81	77	158	48	67	115	67	11	—	—	1 subcutaneous fibrosarcoma
II	0.3 mg	57	+	82	77	159	51	48	99	67	6	—	—	1 renal pelvis carcinoma
III	0.03 mg	57	+	78	85	163	48	58	106	67	14	1	2	1 malignant histiocitosis
IV	controls: olive oil	57	+	82	77	159	38	57	95	67	6	—	—	1 forestomach papilloma; 1 stomach adenomatous polypus; 2 lymphomas
Total				323	316	639	185	230	415		37	1	2	7

Exposure by ingestion (stomach tube) to VC in olive oil at 1, 0.3, and 0.03 mg/kg body weight, once daily, 4–5 days weekly, for 104 weeks.
[a] Sprague-Dawley rats, 10 weeks old at start.

Table 21
Tumors Presently Correlated to VC Exposure (by Inhalation) on Experimental Rodents and Man

Species	*Angiosarcomas of liver*	*Tumors of brain*	*Tumors of lung*	*Lymphomas and leukemias*	*Hepatomas*	*Angiosarcomas*	*Nephroblastomas*	*Sebaceous cutaneous carcinomas*	*Other cutaneous epithelial tumors*	*Mammary carcinomas*	*Forestomach papillomas and acanthomas*
Rat	+	+			+	+	+	+	+	(+)	+
Mouse	+		+			+		+	+	+	(+)
Hamster	+		(+)	+		(+)			+		+
Man	+	(+)	(+)	(+)	(+)						

Table 22
Reported Cases of Angiosarcoma of the Liver among Workers Exposed to Vinyl Chloride

Country	*Polymerization*	*Other occupation*	*Total*
Belgium	1	—	1
Canada	10	—	10
Czechoslovakia	2	—	2
Federal Republic of Germany	8	1	9
France	5	—	5
Great Britain	2	1	3
Italy	2	1	3
Japan	1	—	1
Norway	1	—	1
Rumania	1	—	1
Sweden	2	1	3
Switzerland	1	—	1
U.S.A.	19	2	21
Yugoslavia	2	—	2
Total	57	6	63