

# Results of Long-Term Carcinogenicity Bioassay on Vinyl Acetate Monomer in Sprague-Dawley Rats

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**ABSTRACT:** Vinyl acetate monomer (VAM) was administered in drinking water supplied *ad libitum* at doses of 5,000, 1,000, and 0 ppm (v/v) to 17-week-old Sprague-Dawley rats (breeders) and to 12-day embryos (offspring). Treatment lasted for 104 weeks; thereafter, animals were kept under control conditions until spontaneous death. VAM was found to cause an increase in total malignant tumors and in carcinomas and/or precursor lesions of the oral cavity, lips, tongue, esophagus, and forestomach. Based on these data, VAM must be considered a multipotent carcinogen.

**KEYWORDS:** vinyl acetate monomer; carcinogenicity; long-term bioassay; rat

## INTRODUCTION

Vinyl acetate monomer (VAM) is an important compound in the plastics industry. VAM (C<sub>4</sub>H<sub>6</sub>O<sub>2</sub>) has a molecular weight of 86.09. Industrial production of VAM started in the United States in 1928.<sup>1</sup> VAM is produced mainly by two processes: (1) In a process used since the 1920s, acetylene and acetic acid are reacted in the vapor phase over a catalyst bed,<sup>2</sup> (2) In another process, largely used since the 1970s, ethylene is reacted with acetic acid in the presence of oxygen.<sup>3</sup> The world production of VAM is over 2.5 million tons per year.<sup>4</sup>

The only commercial use of VAM is in the production of polymers (polyvinyl acetate, polyvinyl alcohol, polyvinyl acetals) and copolymers (ethylene-vinyl acetate and polyvinyl-acetate chloride).<sup>3</sup>

Polyvinyl acetate is mainly used in adhesives for paper, wood, glass, metals, and porcelain. It is also used in latex water paint, for paper coating, for textile and leather finishing, as a base for inks and lacquers, in heat-sealing films, in shatterproof photographic bulbs, as an emulsifying agent in cosmetics, pesticide formulations, and pharmaceuticals, and as a food additive.<sup>5,6</sup> Polyvinyl acetate is used as a component

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in the production of chewing gum. The amount of polymer used in the U.S. is about 5% of the final product; in some European countries, the amount is higher.

Polyvinyl alcohol is the most highly produced synthetic, water-soluble plastic in the world, used in sizing for textile warp and yarn, in laminating adhesives, photo-sensitive films, and cements, and as a binder and emulsifying agent.<sup>5,6</sup> Polyvinyl acetals are produced by the condensation of polyvinyl alcohol with an aldehyde. Commonly used aldehydes are formaldehyde, acetaldehyde, and butyraldehyde. Polyvinyl formal, polyvinyl acetals and polyvinyl butyrals are used in adhesives, paints, lacquers, and films. Polyvinyl butyral is also used in sheet form as an inter-layer in safety glasses and shatter-resistant acrylic protection in aircraft.<sup>5</sup> Ethylene-vinyl acetate copolymers improve the adhesive properties of hot-melt and pressure-sensitive adhesives. They are also used in medical tubing, milk packaging, and beer-dispensing equipment. Plastic containers with barrier layers of ethylene-vinyl alcohol copolymers are replacing many glass and metal containers for packaging food.<sup>5,6</sup> Polyvinyl chloride-acetate copolymers, compounded with plasticizers, are used for cable and wire coverings, in chemical plants and in protective garments.<sup>6</sup>

VAM is not known to occur in nature. In the workplace, it may be present wherever its polymers are produced, used, and stocked. Concentrations of 0.25–2 mg/m<sup>3</sup> have been measured<sup>7</sup> in air where vinyl acetate manufacturing or processing facilities are located. In areas near chemical waste disposal sites, concentrations of 0.5 µg/m<sup>3</sup> have been detected.<sup>8</sup> VAM has been found at concentrations of 50 mg/L in wastewater effluents from a polyvinyl acetate plant.<sup>9</sup> VAM was among the volatile chemicals released from food packaging during heating in microwave ovens. A concentration of 0.002–0.14 µg/cm<sup>2</sup> has been detected.<sup>10</sup> VAM has also been detected in cigarette smoke, at concentrations of 400 ng/cigarette.<sup>11</sup>

VAM has an irritative effect on the upper respiratory system in humans. After subchronic and chronic exposure by inhalation, in experimental animals it causes hyperplasia and metaplasia of the respiratory epithelium, bronchitis, and bronchiolitis.<sup>1</sup> Experimental studies on reproductive and prenatal effects have shown that VAM causes parental toxicity (including decreased fertility), developmental toxicity, and minor skeletal alterations.<sup>1</sup>

Rats exposed to VAM exhaled acetaldehyde as a result of hydrolysis by esterase.<sup>12,13</sup> Acetaldehyde is known to be carcinogenic in experimental animals,<sup>14</sup> and its carcinogenic potential was clearly demonstrated in a study performed in our laboratory, the results of which are reported in another paper in this volume.

The experimental studies on rodents conducted until 1997 proved in one way or another inadequate to evaluate the carcinogenic potential of VAM. To date, seven carcinogenicity studies have been published in the scientific literature. In the first study, 96 male and female Sprague-Dawley rats were exposed to 2500 ppm VAM by inhalation for 52 weeks. Early mortality was high: only 49 animals survived for 26 or more weeks. No tumors related to VAM were reported during 135 weeks.<sup>15–17</sup> Because of the poor survival rate, this study was inadequate for detecting the carcinogenic potential of the monomer.

In a second study, 60 male and 60 female Sprague-Dawley rats were exposed to 0–600 ppm vinyl acetate for about 104 weeks. A slight increase in benign and malignant nasal cavity tumors was found.<sup>18</sup>

VAM was administered at doses of 0–2500 mg/L in drinking water for 100 weeks, to 20 male and 20 female Fischer F344 rats. An increase in liver neoplastic nodules,

in uterine adenocarcinomas and polyps, and in thyroid C-cell adenomas was observed.<sup>19</sup> The number of the animals tested was small and the histopathological examination was limited to gross lesions and major organs only.

Male ( $n = 72$ ) and female ( $n = 144$ ) Sprague-Dawley rats received 0–5000 mg/L VAM in drinking water. Treatment began 10 weeks before mating and was continued for an additional four weeks for males and throughout mating, gestation, and lactation for females. Sixty male and 60 female F<sub>1</sub> pups were administered 0–5000 mg/L vinyl acetate in drinking water for 104 weeks. No treatment-related increase in tumor incidence was observed.<sup>20</sup>

Fifty male and 50 female F344 rats received 0–10,000 ppm vinyl acetate (98% pure) for 104 weeks. Statistically significant increases in preneoplastic changes and squamous cell neoplasms were observed at several sites in the upper digestive tract, but only at the 10,000 ppm dose (unpublished data).<sup>21</sup>

The last two studies were performed on mice exposed to VAM by inhalation. Swiss mice exposed to 0–600 ppm vinyl acetate for about 104 weeks showed no treatment-related increase in tumor incidence.<sup>18</sup> BDF<sub>1</sub> mice exposed to 0–10,000 ppm vinyl acetate for 104 weeks showed statistically significant increases in preneoplastic changes and squamous cell neoplasms at several sites in the upper digestive tract, but only at the 10,000 ppm dose (unpublished data).<sup>21</sup>

In a cohort study aimed at identifying the specific exposure associated with an excess of lung cancer risk in humans in a synthetic chemical plant, 19 chemicals were studied: the subgroup with undifferentiated large-cell lung cancer had slightly higher cumulative exposure to VAM.<sup>22</sup> In a nested case-control study on a cohort of 29,139 men employed in two U.S. facilities, who died of lymphomas or leukemias, no significant association was reported.<sup>23</sup>

VAM showed genotoxic effects in both human and rodent cells.<sup>1</sup> *In vitro*, VAM produced a dose-related, statistically significant increase of sister chromatid exchanges and chromosomal aberrations in human lymphocytes and whole blood and an increase of sister chromatid exchanges in ovarian cells of Chinese hamsters.<sup>1</sup>

In the 1980s, a research project on VAM consisting of three experiments conducted with the same protocol, using Sprague-Dawley and Wistar rats and Swiss mice, was started at the Cancer Research Center of the Ramazzini Foundation (CRC/RF). The results of the experiment on Swiss mice have been published,<sup>24</sup> and those on Wistar rats are in publication.<sup>25</sup> Results of the experiment on Sprague-Dawley rats are reported herein for the first time.

## MATERIALS AND METHODS

VAM, purity >99%, was supplied by an Italian chemical plant. The impurities were: benzene 30–45 ppm; methyl and ethyl acetate 50 ppm; crotonaldehyde 6–16 ppm; acetaldehyde 2–11 ppm; acetone 330–500 ppm.

VAM was administered in drinking water supplied *ad libitum* at concentrations of 5000, 1000, or 0 ppm to 17-week-old male and female Sprague-Dawley rats (breeders) and 12-day embryos (offspring). Control animals received tap water without VAM. After 104 weeks of treatment, all animals received regular tap water.

The experimental protocol, including method of tumor reporting and statistical methods, are reported in detail elsewhere in this volume.<sup>26</sup>

## RESULTS

There were no substantial differences between treated animals and controls in mean body weight, survival, behavior, or treatment-related nononcological pathological changes.

The occurrence of benign and malignant tumors is shown in TABLE 1. Differences observed between treated and control animals were:

(1) an increase in total malignant tumors per 100 animals in male breeders and in male and female offspring of the VAM-treated group (TABLE 2);

(2) an increased incidence of squamous cell carcinomas of the oral cavity and lips in female breeders treated at two dose levels, and in male and female offspring treated at 5000 ppm (TABLE 3);

(3) an increased incidence of squamous cell carcinomas of the tongue in female breeders and in male and female offspring exposed at 5000 ppm; an increased incidence of squamous cell dysplasias was observed in all treated female breeders and offspring and in male offspring treated at 5000 ppm (TABLE 4);

(4) a dose-related increase in the incidence of squamous cell dysplasia of the esophagus in male offspring and female breeders, in male offspring treated at 5000 ppm, and in the female offspring of both treated groups; one case of squamous cell carcinoma occurred in a male offspring treated at 5000 ppm (TABLE 5);

(5) an increased incidence of squamous cell carcinomas of the forestomach in male and female breeders treated at the higher dose and also in male breeders treated at the lower dose; a dose-related increase in incidence occurred in male and female offspring; the same trend was observed for squamous dysplasias (TABLE 6);

(6) when squamous cell dysplasias and carcinomas of the upper gastrointestinal tract were considered as a whole, a highly significant dose-related increase was observed in male and female breeders and offspring (TABLE 7).

## CONCLUSIONS

VAM caused an increase in total malignant tumors and tumors at several body sites. The increase in squamous cell carcinomas of the oral cavity and lips, tongue, esophagus and forestomach is of particular significance for two reasons: 1) the same oncological lesions were found in Swiss mice and Wistar rats during the experiments performed in our laboratory following the same experimental protocols; and 2) because the sites of these tumors were tissues most directly exposed to VAM.

The results of this experiment, together with those of the experiment on Swiss mice<sup>24</sup> and on Wistar rats,<sup>25</sup> show that VAM is a multipotent carcinogen. As reported in another paper of this volume, most tumors arose after 112 weeks of age in the present study. Had we stopped our experiment at 112 weeks of age it is unlikely that we would have found the multipotent carcinogenic activity of VAM.

Based on these experimental findings, regulatory measures must be undertaken to prevent the carcinogenic risk of VAM among workers exposed and consumers of goods containing the monomer. Of particular concern is the current use of VAM-based polymers for containers of food and beverages, as VAM has been found to migrate from plastic material into wine<sup>27</sup> and water.<sup>28</sup> Based on this, the use of VAM-based polymers for the production of chewing gum must also be of concern, as one cannot exclude migration into saliva and other biological fluids.

**TABLE 1. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

NUMBER AND PERCENTAGE OF MALE AND FEMALE SPRAGUE-DAWLEY RATS BEARING VARIOUS TYPES OF BENIGN AND MALIGNANT TUMORS (a)

Site	Groups																							
	I: 5,000 ppm								II: 1,000 ppm								III: 0 (control)							
	Breeders				Offspring				Breeders				Offspring				Breeders		Offspring					
	Male		Female		Male		Female		Male		Female		Male		Female		Male	Female	Male	Female				
No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%			
Skin																								
Acanthoma	0	-	0	-	0	-	1	1.8	0	-	0	-	0	-	0	-	0	-	0	-	0	-		
Dermatofibroma	2(3)	15.4	0	-	2	3.8	0	-	0	-	0	-	1	1.2	0	-	0	-	0	-	2	1.9	1	1.0
Squamous cell carcinoma	0	-	0	-	0	-	0	-	0	-	1	2.7	1	1.2	0	-	0	-	0	-	0	-	0	-
Carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0
Basocellular carcinoma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Siringocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	1	1.2	0	-	0	-	0	-	0	-	0	-
Sebaceous adenocarcinoma	0	-	0	-	0	-	0	-	1	7.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Fibrosarcoma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Subcutaneous tissue																								
Fibroma	0	-	0	-	2	3.8	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	0.9	0	-
Lipoma and fibrolipoma	0	-	0	-	0	-	0	-	1	7.7	0	-	0	-	0	-	0	-	0	-	2	1.9	0	-
Fibrosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	7.1	0	-	0	-	0	-
Liposarcoma	1	7.7	0	-	1	1.9	0	-	1	7.7	0	-	1	1.2	1	1.1	0	-	0	-	3	2.8	0	-
Mammary glands																								
Fibroma & fibroadenoma	0	-	15(22)	40.5	8	15.1	29(42)	50.9	2	15.4	20(32)	54.1	2	2.4	50(62)	57.5	1	7.1	22(28)	59.5	9(10)	8.4	69(98)	69.7
Lipoma & fibrolipoma	0	-	0	-	1	1.9	0	-	1	7.7	0	-	3(4)	3.6	2	2.3	2	14.3	0	-	5	4.7	1	1.0
Adenocarcinoma	0	-	6	16.2	0	-	13(16)	22.8	0	-	7	18.9	1	1.2	12(19)	13.8	0	-	7(9)	18.9	3	2.8	17(21)	17.2
Carcinosarcoma	0	-	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	1	1.0
Fibrosarcoma	0	-	0	-	0	-	1	1.8	0	-	0	-	0	-	3	3.4	0	-	3(4)	8.1	0	-	4	4.0
Liposarcoma	1	7.7	0	-	0	-	2	3.5	0	-	0	-	2	2.4	2	2.3	0	-	1	2.7	2	1.9	2	2.0
Rhabdomyosarcoma	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Harderian glands																								
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	1	1.2	0	-	0	-	0	-	0	-	0	-
Zymbal glands																								
Sebaceous adenoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-
Carcinoma	0	-	0	-	1	1.9	0	-	1	7.7	0	-	3	3.6	1	1.1	1	7.1	0	-	2(3)	1.9	0	-
Ear ducts																								
Carcinoma	0	-	0	-	2	3.8	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-

— Continued

TABLE 1. *Continued*

Site	Groups																							
	I: 5,000 ppm						II: 1,000 ppm						III: 0 (control)											
	Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Nasal cavities	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Olfactory neuroblastoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Oral cavity & lips <sup>b</sup>																								
Acanthoma	0	-	1	1.9	1	1.8	0	-	1	2.7	0	-	0	-	0	-	1	7.1	1	2.7	1	2.7	1	0.9
Carcinoma	0	-	2	5.4	13	24.5	9	15.8	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	2	1.9
Fibrosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	0.9
Tongue <sup>c</sup>																								
Granulosa cell tumor	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-
(Abrikossoff's tumor)																								
Carcinoma	0	-	1	2.7	1	1.9	2	3.5	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-
Pharynx																								
Carcinoma	0	-	0	-	1	1.9	1	1.8	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Lung																								
Adenoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Fibrosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Pleura																								
Mesothelioma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Oesophagus <sup>d</sup>																								
Carcinoma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Stomach																								
- Forestomach <sup>e</sup>																								
Acanthoma	1	7.7	1	2.7	6	11.3	2	3.5	3	23.1	0	-	7	8.4	5	5.7	2	14.3	1	2.7	9	8.4	8	8.1
Carcinoma	1	7.7	3	8.1	7	13.2	4	7.0	1	7.7	0	-	6	7.2	3	3.4	0	-	0	-	0	-	0	-
Intestine																								
Leiomyoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-
Fibrosarcoma	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-

— *Continued*

TABLE 1. Continued

Site	Groups																							
	I. 5,000 ppm						II. 1,000 ppm						III. 0 (control)											
	Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring					
No.	%	Male	Female	No.	%	Male	Female	No.	%	Male	Female	No.	%	Male	Female	No.	%	Male	Female	No.	%			
Liver																								
Cholangioma	0	-	0	-	1	1.9	1	1.8	0	-	0	-	0	-	1	1.1	0	-	0	-	1	0.9	1	1.0
Angioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	7.1	0	-	0	-	0	-
Hepatocarcinoma	0	-	2	5.4	3	5.7	1	1.8	0	-	1	2.7	1	1.2	2	2.3	0	-	0	-	5	4.7	5	5.1
Angiosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	0.9	1	1.0
Pancreas																								
Exocrine adenoma	0	-	0	-	1	1.9	0	-	3	23.1	1	2.7	5	6.0	2	2.3	1	7.1	0	-	0	-	1	1.0
Istlet cell adenoma	4	30.8	0	-	2	3.8	1	1.8	1	7.7	1	2.7	9	10.8	2	2.3	0	-	2	5.4	10	9.3	3	3.0
Exocrine adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.1	0	-	0	-	0	-	0	-
Istlet cell carcinoma	1	7.7	1	2.7	0	-	2	3.5	0	-	0	-	1	1.2	2	2.3	0	-	0	-	2	1.9	0	-
Kidneys																								
Adenoma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	1	2.7	0	-	0	-
Adenocarcinoma	0	-	0	-	1	1.9	0	-	0	-	0	-	0	-	0	-	0	-	0	-	2	1.9	0	-
Liposarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.1	0	-	0	-	0	-	1	1.0
Angiosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	1	1.2	0	-	0	-	0	-	0	-	0	-
Pelvis and ureters																								
Transitional cell papilloma	0	-	0	-	0	-	0	-	0	-	0	-	1	1.2	0	-	0	-	0	-	0	-	0	-
Bladder																								
Leiomyosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.1	0	-	0	-	0	-	0	-
Testes																								
Leydig cell tumor	0	-	4(6)	7.5	0	-	0	-	2(3)	2.4	0	-	0	-	0	-	0	-	0	-	5(7)	4.7	0	-
Malignant Leydig cell tumor	1	7.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Ovaries																								
Luteoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1(2)	1.1	0	-	0	-	0	-	0	-
Granulosa cell tumor	3(4)	8.1	4(5)	7.0	0	-	3(4)	8.1	0	-	0	-	1	1.1	0	-	0	-	0	-	2(3)	2.0	0	-
Granulosa and theca cell tumor	1	2.7	0	-	0	-	0	-	0	-	0	-	2(3)	2.3	0	-	1	2.7	0	-	2	2.0	0	-
Sertoli cell tumor	0	-	1(2)	1.8	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Fibroma	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0
Granulosa cell malignant tumor	1	2.7	0	-	0	-	1	2.7	0	-	1	2.7	0	-	0	-	0	-	0	-	0	-	0	-

— Continued



TABLE 1. Continued

Site	Groups																			
	I: 5,000 ppm						II: 1,000 ppm						III: 0 (control)							
	Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring		Breeders		Offspring	
Male No.	Female No.	Male %	Female %	Male No.	Female No.	Male %	Female %	Male No.	Female No.	Male %	Female %	Male No.	Female No.	Male %	Female %	Male No.	Female No.	Male %	Female %	
Peripheral nervous system																				
- Major peripheral nerves	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Benign Schwannoma	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Malignant Schwannoma																				
Bones																				
- Head																				
Osteosarcoma	0	1	2.7	1	1.9	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
- Other																				
Osteosarcoma	0	0	0	1	1.9	0	0	0	1	2.7	3	3.6	1	1.1	0	0	0	0	0	2
Soft tissues																				
Lipoma	0	1	2.7	0	0	0	0	0	0	0	0	1	1.2	0	0	0	0	0	1	0.9
Liposarcoma	0	0	0	1	1.9	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0.9
Thymus																				
Malignant thymoma <sup>f</sup>	1	7.7	0	1	1.9	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Spleen																				
Fibrosarcoma	1	7.7	0	1	1.9	0	0	0	0	0	0	0	1	1.1	0	0	0	0	0	0
Mesenteric lymph nodes																				
Fibrosarcoma	0	0	0	0	0	0	0	0	0	0	0	1	1.2	1	1.1	1	7.1	0	0	0
Hemolymphoreticular tissues <sup>g</sup>																				
Lymphomas and leukemias	1	7.7	2	5.4	6	11.3	7	12.3	4	30.8	10	27.0	20	24.1	19	21.8	0	8	21.6	13

<sup>a</sup> Number in brackets indicate the total number of tumors, one animal can bear more than one tumor

<sup>b</sup> See table 3

<sup>c</sup> See table 4

<sup>d</sup> See table 5

<sup>e</sup> See table 6

<sup>f</sup> In 96 percent of cases, the tumor itself is composed of a mixture in varying proportions of epithelial cells and lymphocytes. In the remaining 4 percent, only epithelial cells are present. We consider that a tumor composed exclusively of lymphocytes would not be classified as a thymoma but as a lymphoma involving the thymus

<sup>g</sup> Including thymus, spleen and mesenteric lymph nodes

**TABLE 2. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

TOTAL MALIGNANT TUMORS								
Group dose (ppm, v/v)	Animals			Malignant tumors				
	Age	Sex	No.	Tumor-bearing animals		Tumors		
				No.	%	No.	Per 100 animals	
I 5,000	Breeders (17 weeks old)	M	13	8	<b>61.5</b>	11	<b>84.6 **</b>	
		F	37	18	<b>48.6</b>	27	73.0	
		M+F	50	26	<b>52.0</b>	38	76.0	
	Offspring (Embryos)	M	53	31		<b>58.5 *</b>	59	<b>111.3 **</b>
		F	57	32		<b>56.1</b>	62	<b>108.8 **</b>
		M+F	110	63		<b>57.3</b>	121	<b>110.0</b>
II 1,000	Breeders (17 weeks old)	M	13	7	<b>53.8</b>	9	<b>69.2 *</b>	
		F	37	20	<b>54.1</b>	33	89.2	
		M+F	50	27	<b>54.0</b>	42	84.0	
	Offspring (Embryos)	M	83	38		45.8	55	<b>66.3</b>
		F	87	42		<b>48.3</b>	79	<b>90.8 *</b>
		M+F	170	80		47.1	134	<b>78.8</b>
III 0 <sup>a</sup>	Breeders (17 weeks old)	M	14	5	35.7	7	<b>50.0</b>	
		F	37	22	<b>59.5</b>	34	91.9	
		M+F	51	27	52.9	41	80.4	
	Offspring (Embryos)	M	107	43		40.2	63	58.9
		F	99	43		43.4	66	66.7
		M+F	206	86		41.7	129	62.6

<sup>a</sup>Drinking water alone. \*  $p < 0.05$  using  $\chi^2$  test. \*\*  $p < 0.01$  using  $\chi^2$  test.

**TABLE 3. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

ONCOLOGICAL LESIONS OF THE ORAL CAVITY AND LIPS											
Group/ dose (ppm, v/v)	Animals			Animals with oncological lesions						Total	
	Age	Sex	No.	Acanthomas		SqDy		SqCa		No.	Per 100 animals
				No.	%	No.	%	No.	%		
I (5,000)	Breeders	M	13	0	-	0	-	0	-	0	-
		F	37	0	-	0	-	2	5.4	2	5.4
		M+F	50	0	-	0	-	2	4.0	2	4.0
	Offspring	M	53	1	1.9	3	5.7	13	24.5 **	17	32.1 **
		F	57	1	1.8	0	-	9	15.8 **	10	17.5 ****
		M+F	110	2	1.8	3	2.7	22	20.0	27	24.5
II (1,000)	Breeders	M	13	0	-	0	-	0	-	0	-
		F	37	1	2.7	0	-	1	2.7	2	5.4
		M+F	50	1	2.0	0	-	1	2.0	2	4.0
	Offspring	M	83	0	-	0	-	0	-	0	-
		F	87	0	-	2	2.3	0	-	2	2.3 **
		M+F	170	0	-	2	1.2	0	-	2	1.2
III 0 <sup>a</sup>	Breeders	M	14	1	7.1	0	-	0	-	1	7.1
		F	37	1	2.7	0	-	0	-	1	2.7
		M+F	51	2	3.9	0	-	0	-	2	3.9
	Offspring	M	107	1	0.9	1	0.9	2	1.9	4	3.7
		F	99	0	-	0	-	1	1.0	1	1.0
		M+F	206	1	0.5	1	0.5	3	1.5	5	2.4

<sup>a</sup>Drinking water alone. \*  $p < 0.01$  using  $\chi^2$  test. \*\* $p < 0.01$  using Cochrane-Armitage test for dose-response relationship.

**TABLE 4. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

ONCOLOGICAL LESIONS OF THE TONGUE											
Group/ dose (ppm, v/v)	Animals			Animals with oncological lesions						Total	
	Age	Sex	No.	Acanthomas		SqDy		SqCa		No.	Per 100 animals
				No.	%	No.	%	No.	%		
I (5,000)	Breeders	M	13	0	-	0	-	0	-	0	-
		F	37	0	-	7	18.9 *	1	2.7	8	21.6 ****
		M+F	50	0	-	7	14.0	1	2.0	8	16.0
	Offspring	M	53	0	-	3	5.7	1	1.9	4	7.5 *
		F	57	0	-	9	15.8 ****	2	3.5	11	19.3 ****
		M+F	110	0	-	12	10.9	3	2.7	15	13.6
II (1,000)	Breeders	M	13	0	-	0	-	0	-	0	-
		F	37	0	-	3	8.1	1	2.7	4	10.8 **
		M+F	50	0	-	3	6.0	1	2.0	4	8.0
	Offspring	M	83	0	-	0	-	0	-	0	-
		F	87	0	-	2	2.3 **	0	-	2	2.3 **
		M+F	170	0	-	2	1.2	0	-	2	1.2
III 0 <sup>a</sup>	Breeders	M	14	0	-	0	-	0	-	0	-
		F	37	0	-	0	-	0	-	0	-
		M+F	51	0	-	0	-	0	-	0	-
	Offspring	M	107	0	-	0	-	0	-	0	-
		F	99	0	-	1	1.0	0	-	1	1.0
		M+F	206	0	-	1	0.5	0	-	1	0.5

<sup>a</sup>Drinking water alone. \* $p < 0.05$  using  $\chi^2$  test. \*\* $p < 0.01$  using  $\chi^2$  test. \*\*\* $p < 0.01$  using Cochran-Armitage test for dose-response relationship.

**TABLE 5. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

ONCOLOGICAL LESIONS OF THE ESOPHAGUS											
Group/ dose (ppm, v/v)	Animals			Animals with oncological lesions						Total	
	Age	Sex	No.	Acanthomas		SqDy		SqCa		No.	Per 100 animals
				No.	%	No.	%	No.	%		
I (5,000)	Breeders	M	13	0	-	2	15.4	0	-	2	15.4
		F	37	0	-	8	21.6 ***	0	-	8	21.6 ***
		M+F	50	0	-	10	20.0	0	-	10	20.0
	Offspring	M	53	0	-	19	35.8 **	1	1.9	20	37.7 **
		F	57	0	-	23	40.4 ****	0	-	23	40.4 ****
		M+F	110	0	-	42	38.2	1	0.9	43	39.1
II (1,000)	Breeders	M	13	0	-	1	7.7	0	-	1	7.7
		F	37	0	-	2	5.4 **	0	-	2	5.4 **
		M+F	50	0	-	3	6.0	0	-	3	6.0
	Offspring	M	83	0	-	0	-	0	-	0	-
		F	87	0	-	4	4.6 **	0	-	4	4.6 **
		M+F	170	0	-	4	2.4	0	-	4	2.4
III 0 <sup>a</sup>	Breeders	M	14	0	-	0	-	0	-	0	-
		F	37	0	-	1	2.7	0	-	1	2.7
		M+F	51	0	-	1	2.0	0	-	1	2.0
	Offspring	M	107	0	-	0	-	0	-	0	-
		F	99	0	-	0	-	0	-	0	-
		M+F	206	0	-	0	-	0	-	0	-

<sup>a</sup>Drinking water alone. \* $p < 0.05$  using  $\chi^2$  test. \*\* $p < 0.01$  using  $\chi^2$  test. \*\*\* $p < 0.01$  using Cochran-Armitage test for dose-response relationship.

**TABLE 6. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

ONCOLOGICAL LESIONS OF THE FORESTOMACH												
Group/ dose (ppm, v/v)	Animals			Animals with oncological lesions						Total		
	Age	Sex	No.	Acanthomas		SqDy		SqCa		No.	Per 100 animals	
				No.	%	No.	%	No.	%			
I (5,000)	Breeders	M	13	1	7.7	4	<b>30.8</b>	1	<b>7.7</b>	6	<b>46.2</b>	
		F	37	1	2.7	11	<b>29.7 *</b>	3	<b>8.1</b>	15	<b>40.5 **</b>	
		M+F	50	2	4.0	15	<b>30.0</b>	4	<b>8.0</b>	21	<b>42.0</b>	
	Offspring	M	53	6	11.3	13	<b>24.5 ****</b>	7	<b>13.2 **</b>	26	<b>49.1 ****</b>	
		F	57	2	3.5	14	<b>24.6 ****</b>	4	<b>7.0 *</b>	20	<b>35.1 ****</b>	
		M+F	110	8	7.3	27	<b>24.5</b>	11	<b>10.0</b>	46	<b>41.8</b>	
II (1,000)	Breeders	M	13	3	23.1	3	<b>23.1</b>	1	<b>7.7</b>	7	<b>53.8</b>	
		F	37	0	-	2	5.4	0	-	2	<b>5.4</b>	
		M+F	50	3	6.0	5	10.0	1	2.0	9	<b>18.0</b>	
	Offspring	M	83	7	8.4	16	<b>19.3 ****</b>	6	<b>7.2 *</b>	29	<b>34.9 ****</b>	
		F	87	5	5.7	14	<b>16.1 ***</b>	3	<b>3.4</b>	22	<b>25.3 ***</b>	
		M+F	170	12	7.1	30	<b>17.6</b>	9	<b>5.3</b>	51	<b>30.0</b>	
III 0 <sup>a</sup>	Breeders	M	14	2	14.3	1	7.1	0	-	3	21.4	
		F	37	1	2.7	3	8.1	0	-	4	10.8	
		M+F	51	3	5.9	4	7.8	0	-	7	13.7	
	Offspring	M	107	9	8.4	4	3.7	0	-	13	12.1	
		F	99	8	8.1	4	4.0	0	-	12	12.1	
		M+F	206	17	8.3	8	3.9	0	-	25	12.1	

<sup>a</sup>Drinking water alone. \* $p < 0.05$  using  $\chi^2$  test. \*\* $p < 0.01$  using  $\chi^2$  test. <sup>†</sup> $p < 0.01$  using Cochran-Armitage test for dose-response relationship.

**TABLE 7. Long-term carcinogenicity bioassay on vinyl acetate monomer (VAM) in drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats**

UPPER GIT SQUAMOUS CELL CARCINOMAS (SqCa) PLUS THEIR PRECURSOR (SqDy)						
Group No.	Dose (ppm, v/v)	Animals			SqDy + SqCa	
		Age	Sex	No.	No. per 100 animals	
I	5,000	Breeders (17 weeks old)	M	13	<b>53.8 ****</b>	
			F	37		<b>86.5 ****</b>
			M+F	50		
		Offspring (Embryos)	M	53	<b>113.2 ****</b>	
			F	57		<b>107.0 ****</b>
			M+F	110		
II	1,000	Breeders (17 weeks old)	M	13	<b>38.5 ****</b>	
			F	37		<b>24.3 ***</b>
			M+F	50		
		Offspring (Embryos)	M	83	<b>26.5 ****</b>	
			F	87		<b>28.7 ****</b>
			M+F	170		
III	0 <sup>a</sup>	Breeders (17 weeks old)	M	14	7.1	
			F	37	10.8	
			M+F	51	9.8	
		Offspring (Embryos)	M	107	6.5	
			F	99	6.1	
			M+F	206	6.3	

<sup>a</sup>Drinking water alone. \* $p < 0.05$  using  $\chi^2$  test. \*\* $p < 0.01$  using  $\chi^2$  test. \*\*\* $p < 0.01$  using Cochran-Armitage test for dose-response relationship.

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