Results of Long-Term Experimental Studies on the Carcinogenicity of Formaldehyde and Acetaldehyde in Rats

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ABSTRACT: Formaldehyde was administered for 104 weeks in drinking water supplied *ad libitum* at concentrations of 1500, 1000, 500, 100, 50, 10, or 0 mg/L to groups of 50 male and 50 female Sprague-Dawley rats beginning at seven weeks of age. Control animals (100 males and 100 females) received tap water only. Acetaldehyde was administered to 50 male and 50 female Sprague-Dawley rats beginning at six weeks of age at concentrations of 2,500, 1,500, 500, 250, 50, or 0 mg/L. Animals were kept under observation until spontaneous death. Formaldehyde and acetaldehyde were found to produce an increase in total malignant tumors in the treated groups and showed specific carcinogenic effects on various organs and tissues.

KEYWORDS: formaldehyde; acetaldehyde; carcinogenicity; long-term bioassay; rat

INTRODUCTION

Formaldehyde and acetaldehyde are two compounds produced in large amounts, employed almost universally, and diffused ubiquitously. Little is known about their carcinogenicity. Formaldehyde and acetaldehyde develop from combustion of gasoline, methanol, and ethanol fuels. Because of the increased production and use of oxygenated gasolines, particularly those with added methyl *tert*-butyl ether (MTBE) or ethyl *tert*-butyl ether (ETBE), large numbers of people may be exposed to formaldehyde and acetaldehyde daily. This report outlines the final results of the carcinogenicity experiments on formaldehyde and acetaldehyde performed at the Cancer Research Center of the European Ramazzini Foundation (CRC/RF) at the Castle of Bentivoglio.

Formaldehyde (CH_2O) is a colorless gas with a molecular weight of 30, and acetaldehyde (CH_3CHO) is a colorless, mobile liquid with a molecular weight of 44.05 that is miscible with water and most common organic solvents.

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FORMALDEHYDE

Two methods are used to produce formaldehyde: the silver catalyst and metal oxide catalyst process and the metal oxide (Formox) process.[1,2] By-products are carbon monoxide, dimethyl ether, and small amounts of carbon dioxide and formic acid.

The worldwide production of formaldehyde in 1992 was 12 million tons.[3] The widest use of formaldehyde is in the production of resins with urea, phenol, and melamine. Formaldehyde-based resins are used as adhesives and impregnating resins in the manufacture of particle board, plywood, furniture, and other wood products.[4] Formaldehyde is used as an intermediate for synthesizing other industrial chemical compounds, such as 1,4-butanediol, trimethylopropane, and neopentyl glycol, which are used in the manufacture of polyurethane and polyester plastics, synthetic resin coatings, synthetic lubricating oil, and plasticizers. Formaldehyde itself is used as a preservative and disinfectant. It is used as an antimicrobial agent in many cosmetics products, hair preparations, deodorants, lotions, make-up, mouthwashes, and nail products.[5]

From 1981 to 1983, about 1,500,000 workers in the United States were estimated to have been exposed to formaldehyde during all or part of that period, representing about 0.6% of the population. In factories producing formaldehyde-based resins, a mean concentration of <1 to >10 ppm has been reported. More or less the same concentrations have been reported in wood products and paper factories, and those of textile garments, and other activities.[5]

Ambient levels are generally <1 $\mu g/m^3$ in remote areas[6] and 1–20 $\mu g/m^3$[3,4,6–9] in urban environments. A major source of formaldehyde is incomplete combustion of hydrocarbon fuels, especially from vehicle emissions. The introduction of MTBE as a gasoline additive increased formaldehyde emissions. A study by the state of Utah Department of Environmental Quality found that during the months of December 1994 and January 1995, when 2.7% oxygen-content-ethanol– and/or MTBE–oxygenated fuel was used, air concentrations of formaldehyde were increased by 300% to 1,800%, and those of acetaldehyde were increased by 470% to 860%.[10]

Indoor levels of formaldehyde are higher than outdoor levels, with the concentrations depending on the sources of formaldehyde that are present, the age of the source materials, ventilation, temperature, and humidity.[6] Cigarette smoke has been reported to contain levels of a few micrograms to several milligrams of formaldehyde per cigarette.[4] The sweetening agent aspartame hydrolyzes in the gastrointestinal tract to become free methyl alcohol, which is metabolized in the liver to formaldehyde, formic acid, and CO_2.[11]

Formaldehyde induces gene mutation in bacteria, fungi, yeast, *Drosophila larvae*, and cultured rodent and human cells. It also causes single-strand breaks in DNA, sister chromatid exchanges, chromosome aberrations, and the transformation of rodent cells in a variety of *in vitro* assays.[12–15] Formaldehyde induces cytogenetic damage in tissues that are locally exposed, either by gavage or inhalation. Sprague-Dawley rats treated orally with formaldehyde and sacrificed 16–30 h after treatment had a greater than fivefold increase in the frequency of micronucleated cells in the stomach, duodenum, ileum, and colon.[16] Male Sprague-Dawley rats exposed by inhalation to formaldehyde vapor for one and eight weeks showed a significant increase in chromosomal aberrations in pulmonary lavage cells.[17] Fisher 344 rats and B6C3F1 mice who received whole-body exposure to formaldehyde vapor for up to 24 months showed nasal cavity malignancies.[18]

In two experiments, male Syrian Golden hamsters exposed to 30 ppm or 10 ppm of formaldehyde for 5 h/day, 1 or 5 days/week for life showed no nasal tumors.[19] Wistar rats who received drinking water containing formaldehyde for up to 24 months at concentrations of 0–82 mg/kg bw per day for males, and 0–109 mg/kg bw per day for females, showed no treatment-related neoplastic changes.[20]

In a lifetime study performed in our laboratory, formaldehyde was administered in drinking water to male and female Sprague-Dawley rats. In this study, male and female breeder rats were given formaldehyde at 2,500 or 0 ppm for 104 weeks. The offspring were initially exposed to 2,500 or 0 ppm formaldehyde *in utero* starting on day 13 of gestation and then received these levels in drinking water for 104 weeks. Preliminary results demonstrated an increase in a variety of malignant and benign tumors of the stomach and intestine in treated animals. It was also noted that, although the incidence of intestinal tract tumors was low, there were no comparable tumors in the control groups in this study; some of these tumors were reported to be uncommon among historical controls.[21]

A comprehensive review of the epidemiological studies conducted on industrial and other professional groups exposed to formaldehyde has been reported.[5] Overall, epidemiological studies may only suggest a causal relationship between formaldehyde exposure and nasopharyngeal cancer. No excess risks have been shown for oropharyngeal, laryngeal, and lung cancer; low or no risks have been shown for lymphatic or hematopoietic cancer.

ACETALDEHYDE

The production capacity for acetaldehyde in the United States in 1989 was 443,000 tons/year.[23] Acetaldehyde is used principally as a chemical intermediate, predominantly of acetic acid, which is primarily used to make vinyl acetate, cellulose acetate, and other acetic esters.[24] Acetaldehyde is also used in silvering mirrors, leather tanning, as a denaturant for alcohol, in fuel mixtures, as a hardener for gelatin fibers, in glue and casein products, as a preservative for fish and fruit, as well as in the paper industry and as a synthetic rubber.[25]

Acetaldehyde is a natural product of combustion and photo-oxidation of hydrocarbons and has been detected at low levels in drinking water, surface water, rainwater, effluents, engine exhaust, and ambient and indoor air samples. It is present in small amounts in alcoholic beverages such as beer, wine and spirits, and in plant juices, essential oils, and roasted coffee.[23,25–26] Cigarette smoke has been reported to contain acetaldehyde at the level of 980 μg/cigarette.[27] The concentration of acetaldehyde in the whole blood of normal fasting human subjects was reported by Lynch *et al*.[28] to be 1.30 μmol/L (57 μg/L). Acetaldehyde has been detected in mothers' milk.[29] Acetaldehyde is formed during the intracellular oxidation of ethanol.[30] It is metabolized to acetic acid by NAD^+-dependent aldehyde dehydrogenases.[31]

Fetal malformations have been found in mice and rats treated with acetaldehyde.[32] Acetaldehyde causes gene mutations in bacteria and gene mutation, sister chromatid exchanges, micronuclei and aneuploidy in cultured mammalian cells, without metabolic activation. *In vivo*, it causes mutation in *Drosophyla melanogaster* but not micronuclei in mouse germ cells. It causes DNA damage in cultured mammalian cells and in mice *in vivo*.[33] Acetaldehyde-DNA adducts have been found in white blood cells from human alcohol abusers.[34]

In long-term carcinogenicity bioassays, WU albino Wistar rats treated by inhalation showed dose-related increases in nasal carcinomas in both sexes.[35] In hamsters, inhalation of acetaldehyde enhanced the incidence of respiratory tract tumors produced by intratracheal instillation of benzo[*a*]pyrene.[36]

To date, the available epidemiological studies are inadequate for evaluation of the carcinogenicity of acetaldehyde to humans.

MATERIALS AND METHODS

Formaldehyde was supplied in aqueous solution by Montedison S.p.A., Italy, at a concentration of 30.0 ± 0.2%. The impurities included iron 0.6 mg/L; lead 0.1 mg/L; sulphur <5.0 mg/L; chlorine <5.0 mg/L; methyl alcohol (stabilizer) 0.3%. Acetaldehyde was supplied by FLUKA Chemica-Biokemica, AG, Switzerland; its purity was higher than 99.0%. During experiments, formaldehyde and acetaldehyde were supplied every two months and were always kept in the dark at a temperature of 14–20°C and 4–5°C, respectively.

Formaldehyde was administered for 104 weeks in the drinking water to 7-week-old Sprague-Dawley rats ($n = 50$ males and $n = 50$ females) and supplied *ad libitum* at concentrations of 1,500, 1,000, 500, 100, 50, or 10 mg/L. One group of 50 males and 50 females received methyl alcohol in 15 mg/L of drinking water. Control animals (100 males and 100 females) received tap water. Acetaldehyde was administered at concentrations of 2,500, 1,500, 500, 250, 50, or 0 mg/L to Sprague-Dawley rats (50 males and 50 females), beginning at age 6 weeks. The studies on formaldehyde and acetaldehyde ended with the death of the last animal at 163 and 161 weeks of age, respectively.

Experiments were performed according to the Good Laboratory Practices (GLP) and the Standard Operating Procedure (SOP) of the CRC/RF. Detailed experimental methods and protocol for counting tumors have been described previously.[37] Statistical analysis was performed using the χ^2 test to evaluate differences in tumor incidence between treated and control groups. The Cochrane Armitage test was used to evaluate dose-response relationships.

RESULTS

Formaldehyde

During the experiment, the intake of liquids decreased in males treated at the highest dose and in females treated at 1,500, 1,000, and 500 mg/L of formaldehyde. No differences were observed in daily feed consumption, body weight, behavior, or survival between treated and control animals. Yellow hair coat was observed in animals exposed to formaldehyde, mainly at the highest concentration. No treatment-related non-oncological pathological changes were detected by gross inspection or histopathological examination.

TABLE 1. Long-term carcinogenicity bioassays on formaldehyde administered with drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats

A. NUMBER AND PERCENTAGE OF MALE AND FEMALE SPRAGUE-DAWLEY RATS BEARING VARIOUS TYPES OF BENIGN AND MALIGNANT TUMORS[a]

Site	Groups																															
	I: 1,500 mg/l				II: 1,000 mg/l				III: 500 mg/l				IV: 100 mg/l				V: 50 mg/l				VI: 10 mg/l				VII: 0[b]				VIII: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Skin																																
Cheratoacanthoma	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Dermatofibroma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0
Squamous cell carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-
Basocellular carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0	0	-
Subcutaneous tissue																																
Fibroma	0	-	0	-	0	-	0	-	1	2.0	0	-	2	4.0	0	-	1	2.0	0	-	3	6.0	0	-	0	-	0	-	0	-	0	-
Lipoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	2	2.0	0	-
Fibrosarcoma	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Liposarcoma	0	-	0	-	1	2.0	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	2	2.0	0	-
Angiosarcoma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Mammary glands																																
Fibroma and fibroadenoma	2	4.0	25(28)	50.0	3	6.0	20(23)	40.0	2	4.0	19(20)	38.0	1	2.0	14(20)	28.0	4(5)	8.0	12(14)	24.0	2	4.0	9	18.0	4	8.0	16(20)	32.0	6(8)	6.0	45(61)	45.0
Fibrolipoma	0	-	0	-	0	-	0	-	1	2.0	1	2.0	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Lipoma	0	-	0	-	0	-	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	1	2.0	11(15)	22.0	0	-	9(10)	18.0	0	-	3	6.0	0	-	8(12)	16.0	0	-	4	8.0	0	-	2	4.0	0	-	7(9)	14.0	1	1.0	11(12)	11.0
Fibrosarcoma	0	-	0	-	0	-	1	2.0	0	-	1	2.0	0	-	2	4.0	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-	0	-
Liposarcoma	0	-	1	2.0	0	-	0	-	0	-	2	4.0	1	2.0	0	-	0	-	1	2.0	0	-	0	-	0	-	1	2.0	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Harderian gland																																
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Zymbal glands																																
Sebaceous adenoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	1	1.0
Carcinoma	2(3)	4.0	0	-	3	6.0	0	-	4	8.0	1	2.0	1	2.0	0	-	1	2.0	0	-	2	4.0	0	-	3	6.0	0	-	3	3.0	1	1.0
Ear ducts																																
Carcinoma	0	-	0	-	3	6.0	3	6.0	0	-	1	2.0	0	-	0	-	1	2.0	2	4.0	1	2.0	1	2.0	1	2.0	1	2.0	7	7.0	3	3.0
Nasal cavities																																
Carcinoma	1	2.0	0	-	1	2.0	1	2.0	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-
Neuroblastoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Oral cavity lips and tongue																																
Acanthoma	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Carcinoma	3	6.0	1	2.0	1	2.0	3	6.0	1	2.0	0	-	1	2.0	1	2.0	1	2.0	1	2.0	3	6.0	1	2.0	2	4.0	1	2.0	5	5.0	3	3.0
Lung																																
Adenoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	2	4.0	0	-	0	-	0	-	0	-	1	2.0	0	-	2	2.0	1	1.0
Angioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	1	2.0	1	1.0	0	-
Esophagus																																
Carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Stomach																																
- Forestomach																																
Acanthoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	1	2.0	3	6.0	0	-	0	-	0	-	2	2.0	2	2.0
Squamous cell carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Leiomyosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-

— Contintued

TABLE 1. *Continued*

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

	Groups																															
	I: 1,500 mg/l				II: 1,000 mg/l				III: 500 mg/l				IV: 100 mg/l				V: 50 mg/l				VI: 10 mg/l				VII: 0[b]				VIII: 0 (control)			
Site	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
- Glandular stomach																																
Adenomatous polyp	6	12.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	1	2.0	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-
Leiomyosarcoma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Intestine																																
Adenomatous polyp	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Leiomyoma	0	-	3	6.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	2	4.0	0	-	0	-	0	-	0	-
Adenocarcinoma	3	6.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-	0	-	0	-	0	-
Leiomyosarcoma	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Salivary glands																																
Adenocarcinoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Liver																																
Cholangioma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	1	2.0	1	2.0	1	2.0	0	-	0	-	1	2.0	0	-	1	1.0	0	-
Hepatocarcinoma	2	4.0	0	-	0	-	1	2.0	1	2.0	1	2.0	0	-	2	4.0	0	-	0	-	0	-	0	-	1	2.0	0	-	2	2.0	0	-
Cholangiocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0	1	1.0
Embryonal sarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-
Pancreas																																
Exocrine adenoma	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-	1	1.0
Islet cell adenoma	1	2.0	0	-	3	6.0	1	2.0	3	6.0	2	4.0	0	-	2	4.0	1	2.0	2	4.0	1	2.0	1	2.0	2	4.0	2	4.0	5	5.0	3	3.0
Islet cell carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-
Kidneys																																
Adenoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0	0	-
Lipoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Nephroblastoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0
Pelvis and ureters																																
Transitional cell carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Bladder																																
Angioma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Prostate																																
Adenoma	1	2.0			0	-			0	-			0	-			0	-			0	-			0	-			1	1.0		
Testes																																
Interstitial cell adenoma	9(11)	18.0			12(21)	24.0			10(14)	20.0			6(7)	12.0			6(11)	12.0			3(5)	6.0			3(5)	6.0			10(12)	10.0		
Interstitial cell malignant tumor	0	-			0	-			1	2.0			0	-			0	-			0	-			0	-			0	-		
Ovaries																																
Cystadenoma			0	-			0	-			1	2.0			0	-			1	2.0			0	-			0	-			0	-
Granulosa cell tumor			2(4)	4.0			3(5)	6.0			2(4)	4.0			0	-			0	-			1(2)	2.0			2(3)	4.0			4(7)	4.0
Granulosa and theca cell tumor			2	4.0			0	-			0	-			0	-			0	-			0	-			0	-			2	2.0
Sertoli cell tumor			0	-			1	2.0			1	2.0			0	-			0	-			0	-			0	-			0	-
Benign arrhenoblastoma			0	-			0	-			0	-			0	-			0	-			0	-			0	-			1	1.0
Adenocarcinoma			1	2.0			0	-			0	-			0	-			0	-			0	-			0	-			0	-

— *Continued*

TABLE 1. *Continued*

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

Site	Groups																															
	I: 1,500 mg/l				II: 1,000 mg/l				III: 500 mg/l				IV: 100 mg/l				V: 50 mg/l				VI: 10 mg/l				VII: 0[b]				VIII: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Uterus																																
Polyp			2	4.0			6	12.0			5	10.0			2	4.0			5	10.0			4	8.0			2	4.0			10	10.0
Leiomyoma			0	-			0	-			0	-			0	-			0	-			1	2.0			0	-			0	-
Fibroangioma			0	-			0	-			0	-			0	-			1	2.0			0	-			1	2.0			0	-
Angioma			0	-			0	-			0	-			1	2.0			0	-			0	-			0	-			0	-
Squamous cell carcinoma			0	-			0	-			1	2.0			0	-			0	-			0	-			1	2.0			1	1.0
Adenocarcinoma			3	6.0			3	6.0			3	6.0			8	16.0			2	4.0			2	4.0			1	2.0			3	3.0
Fibrosarcoma			0	-			0	-			2	4.0			0	-			0	-			0	-			0	-			0	-
Leiomyosarcoma			0	-			0	-			1	2.0			0	-			0	-			0	-			0	-			0	-
Angiosarcoma			1	2.0			0	-			0	-			0	-			0	-			0	-			0	-			0	-
Uterus & Vagina																																
Malignant Schwannoma			0	-			0	-			0	-			0	-			0	-			2	4.0			0	-			3	3.0
Vagina																																
Malignant Schwannoma			0	-			0	-			0	-			0	-			0	-			1	2.0			0	-			0	-
Peritoneum																																
Lipoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Mesothelioma	1	2.0	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	1	1.0
Pituitary gland																																
Adenoma	5	10.0	20	40.0	9	18.0	19	38.0	8	16.0	31	62.0	9	18.0	30	60.0	6	12.0	17	34.0	3	6.0	19	38.0	6	12.0	27	54.0	22	22.0	44	44.0
Thyroid gland																																
C-cell adenoma	2	4.0	2	4.0	0	-	0	-	1	2.0	2	4.0	0	-	1	2.0	1	2.0	1	2.0	1	2.0	1	2.0	0	-	2	4.0	9	9.0	2	2.0
Follicular carcinoma	0	-	0	-	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
C-cell carcinoma	0	-	1	2.0	0	-	1	2.0	0	-	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-	0	-	0	-	2	2.0	0	-
Adrenal glands																																
Cortical adenoma	0	-	0	-	0	-	3	6.0	2	4.0	3	6.0	0	-	3	6.0	1	2.0	3	6.0	0	-	2	4.0	0	-	3	6.0	2	2.0	10(11)	10.0
Pheochromocytoma	11(16)	22.0	12(16)	24.0	22(33)	44.0	8(11)	16.0	19(26)	38.0	20(26)	40.0	25(36)	50.0	16(26)	32.0	25(37)	50.0	17(22)	34.0	21(30)	42.0	11(17)	22.0	16(24)	32.0	10(14)	20.0	53(77)	53.0	36(47)	36.0
Cortical adenocarcinoma	0	-	4(7)	8.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	2	4.0	0	-	0	-	0	-	1	2.0	0	-	4	4.0
Pheochromoblastoma	4(5)	8.0	2	4.0	1	2.0	0	-	8(10)	16.0	1	2.0	3	6.0	1	2.0	0	-	1	2.0	2	4.0	0	-	5(6)	10.0	1	2.0	5(6)	5.0	6	6.0
Central nervous system																																
- Brain																																
Oligodendroglioma	0	-	0	-	1	2.0	1	2.0	1	2.0	0	-	1	2.0	1	2.0	0	-	0	-	1	2.0	2	4.0	0	-	5	10.0	3	3.0	2	2.0
- Meninges																																
Malignant meningioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	2	4.0	2	2.0	0	-
Peripheral nervous system																																
- Major peripheral nerves																																
Malignant Schwannoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0
- Ganglia																																
Pheochromocytoma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Bones																																
- Head																																
Osteosarcoma	3	6.0	1	2.0	3	6.0	0	-	2	4.0	0	-	3	6.0	3	6.0	0	-	0	-	1	2.0	2	4.0	1	2.0	2	4.0	4	4.0	0	-
- Other																																
Osteosarcoma	0	-	1	2.0	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	2	2.0	0	-

— *Contintued*

TABLE 1. *Continued*

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

Site	Groups																															
	I: 1,500 mg/l				II: 1,000 mg/l				III: 500 mg/l				IV: 100 mg/l				V: 50 mg/l				VI: 10 mg/l				VII: 0[b]				VIII: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Soft tissues																																
Fibroangioma	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	1.0	0	-
Liposarcoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Heart																																
Malignant Schwannoma	2	4.0	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Thymus																																
Benign thymoma[c]	0	-	0	-	0	-	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Malignant thymoma[c]	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Spleen																																
Angioma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Angiosarcoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Subcutaneous lymph nodes																																
Fibroangioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1(2)	1.0	0	-
Pancreatic lymph nodes																																
Fibroangioma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Mesenteric lymph nodes																																
Angioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	2	2.0
Fibroangioma	1	2.0	0	-	0	-	0	-	1	2.0	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Hemolymphoreticular tissues[d,e]																																
Histiocytoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-
Lymphomas and leukemias	23	46.0	10	20.0	11	22.0	11	22.0	12	24.0	7	14.0	13	26.0	8	16.0	10	20.0	7	14.0	4	8.0	5	10.0	10	20.0	5	10.0	8	8.0	7	7.0

[a] Number in parentheses are the number of total tumours; one animal can bear more than one tumour

[b] 15 mg of methyl alcohol / liter of drinking water

[c] In 96% of cases the tumor itself is composed of a mixture in varying proportions of epithelial cells and lymphocytes. In the remaining 4%, only epithelial cells are present. We consider that a tumor composed exclusively of lymphocytes should not be classified as a thymoma but as lymphoma involving the thymus.

[d] Including thymus, spleen, subcutaneous, pancreatic and mesenteric lymph nodes

[e] See table 3

TABLE 2. Long-term carcinogenicity bioassays on formaldehyde administered with drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats

TOTAL MALIGNANT TUMORS

Group	Concentration	Animals		Malignant tumors: Tumor-bearing animals		Malignant tumors: Tumors	
No.	(mg/l)	Sex	No.	No.	%	No.	Per 100 animals
I	1,500	M	50	36	**72.0** **	56	**112.0** **
		F	50	27	**54.0**	48	**96.0** **
		M+F	100	63	**63.0**	104	**104.0**
II	1,000	M	50	23	46.0	30	60.0
		F	50	29	**58.0**	39	**78.0** **
		M+F	100	52	52.0	69	69.0
III	500	M	50	24	48.0	36	**72.0** *
		F	50	19	38.0	25	50.0
		M+F	100	43	43.0	61	61.0
IV	100	M	50	22	44.0	23	46.0
		F	50	25	50.0	41	**82.0** **
		M+F	100	47	47.0	64	64.0
V	50	M	50	12	24.0	15	30.0
		F	50	20	40.0	26	52.0
		M+F	100	32	32.0	41	41.0
VI	10	M	50	14	28.0	19	38.0
		F	50	20	40.0	22	44.0
		M+F	100	34	34.0	41	41.0
VII	0[a]	M	50	21	42.0	29	**58.0**
		F	50	23	46.0	32	**64.0**
		M+F	100	44	44.0	61	**61.0**
VIII	0	M	100	38	38.0	50	50.0
		F	100	43	43.0	49	49.0
		M+F	200	81	40.5	99	49.5

[a] 15 mg of methyl alcohol / liter of drinking water

* $p<0.05$ using χ^2 test

** $p<0.01$ using χ^2 test

The occurrence of benign and malignant tumors in animals exposed to formaldehyde is shown in TABLE 1. Differences observed between formaldehyde-treated and control animals are as follows:

(1) The number of total malignant tumors was highest in males and females treated with 1,500 and 1,000 mg/L, in males treated with 500 mg/L, and in females treated with 100 mg/L (TABLE 2). An increase in total malignant

TABLE 3. Long-term carcinogenicity bioassays on formaldehyde administered with drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats

HEMOLYMPHORETICULAR NEOPLASIAS

Group No.	Concentration (mg/l)	Animals		Animals with hemolymphoreticular neoplasias		
		Sex	No.	No.	%	Latency[a] (weeks)
I	1,500	M	50	23	**46.0** **	98.2
		F	50	10	**20.0** *	110.5
		M+F	100	33	**33.0**	104.4
II	1,000	M	50	11	**22.0** *	101.5
		F	50	11	**22.0** *	118.0
		M+F	100	22	**22.0**	109.8
III	500	M	50	12	**24.0** *	121.2
		F	50	7	**14.0**	113.6
		M+F	100	19	**19.0**	117.4
IV	100	M	50	13	**26.0** **	106.3
		F	50	8	**16.0**	118.6
		M+F	100	21	**21.0**	112.5
V	50	M	50	10	**20.0**	111.3
		F	50	7	**14.0**	104.3
		M+F	100	17	**17.0**	107.8
VI	10	M	50	4	8.0	118.7
		F	50	5	10.0	88.6
		M+F	100	9	9.0	103.7
VII	0[b]	M	50	10	**20.0**	105.6
		F	50	5	10.0	106.8
		M+F	100	15	15.0	106.2
VIII	0	M	100	8	8.0	108.7
		F	100	7	7.0	103.0
		M+F	200	15	7.5	105.9

[a] Age at death
[b] 15 mg of methyl alcohol / liter of drinking water
* $p<0.05$ using χ^2 test
** $p<0.01$ using χ^2 test

tumors was also detected in males and females treated with methyl alcohol alone.

(2) The number of mammary malignant tumors increased in females treated with 1,500 ($P < 0.05$), 1,000, and 100 ($P < 0.01$) mg/L and were also increased in females treated with methyl alcohol alone.

(3) Sporadic cases of oncological lesions of the stomach, including two leiomyosarcomas, which have a very rare spontaneous incidence in the colony of Sprague-Dawley rats used, were observed in males treated with formaldehyde at concentrations of 1,000 and 10 mg/L, but were not found in animals treated with methyl alcohol alone or in the control group.

(4) Oncological lesions of the intestine were detected in the treated groups, particularly at the highest dose. Among the lesions observed were leiomyomas and leiomyosarcomas, very rare tumors in the Sprague-Dawley colony of rats used at the CRC/RF.[21] These tumors were not found in animals treated with methyl alcohol alone or in the control group.

(5) The number of testicular interstitial cell adenomas increased in males treated with 1,500, 1,000 ($P < 0.05$), and 500 mg/L of formaldehyde.

(6) The number of hemolymphoreticular neoplasias increased in both sexes treated with 1,500, 1,000, 500, 100, and 50 mg/L, and a slight increase was observed in females treated with 10 mg/L (TABLE 3). This increase shows a dose-response relationship. An increase in hemolymphoreticular neoplasias was also observed in males treated with methyl alcohol alone when compared to nontreated animals.

Acetaldehyde

No significant differences in the daily consumption of beverages and feed, behavior, body weight, or survival were observed between treated and control animals, nor were any treatment-related nononcological pathological changes detected by gross inspection or histopathological examination.

The occurrence of benign and malignant tumors in acetaldehyde-treated animals is shown in TABLE 4. Differences observed between treated and control animals are as follows:

(1) The number of total malignant tumors was increased in all of the treated groups, except for males treated with 250 mg/L (TABLE 5).

(2) The number of malignant mammary tumors increased in all treated females, with the exception of those treated with 250 mg/L. This increase was not dose related. It is noteworthy that some malignant mammary tumors were also observed in treated males, except for those treated with 250 mg/L.

(3) The overall incidence of carcinomas of the Zymbal gland, external ear ducts, nasal sinuses, and oral cavity increased in males and females treated with the highest dose.

(4) Sporadic cases of lung adenoma/adenocarcinoma were found in groups treated with the three highest doses.

(5) Sporadic cases of tumors of the stomach and intestine were observed in the treated groups.

TABLE 4. Long-term carcinogenicity bioassays on acetaldehyde administered with 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: 2,500 mg/l				II: 1,500 mg/l				III: 500 mg/l				IV: 250 mg/l				V: 50 mg/l				VI: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Lung																								
Adenoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	1	2.0	1	2.0	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Pleura																								
Mesothelioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Stomach																								
- Forestomach																								
Acanthoma	2	4.0	2	4.0	1	2.0	2	4.0	0	-	0	-	1	2.0	0	-	2	4.0	0	-	0	-	0	-
Squamous cell carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
- Glandular stomach																								
Adenocarcinoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Intestine																								
Fibroma	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	1	2.0	0	-	0	-	0	-	0	-
Liver																								
Hepatocarcinoma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-
Pancreas																								
Exocrine adenoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Islet cell adenoma	9	18.0	2	4.0	2	4.0	3	6.0	7	14.0	3	6.0	5	10.0	2	4.0	4	8.0	3	6.0	2	4.0	0	-
Kidneys																								
Adenoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Fibrolipoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	1	2.0
Bladder																								
Transitional cell carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Seminal vesicles																								
Adenoma	0	-			0	-			0	-			1	2.0			1	2.0			0	-		
Testes																								
Interstitial cell tumor	4(5)	8.0			3(4)	6.0			4(5)	8.0			1	2.0			6(7)	12.0			2	4.0		
Ovaries																								
Cystadenoma			0	-			0	-			0	-			1	2.0			0	-			0	-
Granulosa cell tumor			0	-			1	2.0			0	-			0	-			0	-			0	-
Fibroangioma			0	-			0	-			1	2.0			0	-			0	-			0	-
Adenocarcinoma			0	-			0	-			0	-			1	2.0			0	-			1	2.0
Granulosa cell malignant tumors			0	-			0	-			0	-			1	2.0			1	2.0			0	-
Uterus																								
Polyp			6	12.0			6	12.0			7	14.0			8	16.0			8	16.0			7	14.0
Squamous cell carcinoma			1	2.0			1	2.0			0	-			0	-			1	2.0			0	[illegible]
Adenocarcinoma			1	2.0			2	4.0			1	2.0			5	10.0			0	-			0	-
Fibrosarcoma			0	-			0	-			0	-			0	-			1	2.0			0	-
Angiosarcoma			0	-			0	-			0	-			0	-			0	-			1	2.0

— Continued

TABLE 4. *Continued*

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

Site	Groups																							
	I: 2,500 mg/l				II: 1,500 mg/l				III: 500 mg/l				IV: 250 mg/l				V: 50 mg/l				VI: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Skin																								
Acanthoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-
Dermatofibroma	0	-	0	-	2	4.0	0	-	1	2.0	0	-	2	4.0	0	-	0	-	0	-	0	-	0	-
Squamous cell carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Basocellular carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Fibrosarcoma	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Subcutaneous tissue																								
Fibroma	1	2.0	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-
Lipoma	0	-	0	-	1	2.0	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-
Liposarcoma	0	-	1	2.0	0	-	0	-	1	2.0	0	-	0	-	1	2.0	0	-	0	-	2	4.0	0	-
Interscapular brown fat pad																								
Lipoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Liposarcoma	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-
Mammary glands																								
Fibroma and fibroadenoma	3	6.0	22(30)	44.0	1	2.0	29(41)	54.0	1	2.0	25(40)	50.0	1	2.0	26(33)	52.0	1	2.0	17(21)	34.0	3	6.0	14(22)	28.0
Fibrolipoma	0	-	1	2.0	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Lipoma	0	-	0	-	1	2.0	1	2.0	0	-	0	-	0	-	0	-	1	2.0	1	2.0	0	-	0	-
Adenocarcinoma	1	2.0	6(10)	12.0	0	-	8	16.0	0	-	10(11)	20.0	0	-	3	6.0	0	-	9(13)	18.0	0	-	3	6.0
Fibrosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	2	4.0	0	-	0	-	0	-
Liposarcoma	0	-	1	2.0	1	2.0	0	-	2	4.0	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-
Carcinosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Harderian gland																								
Adenocarcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-
Zymbal glands																								
Sebaceous adenoma	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Carcinoma	2	4.0	3	6.0	1	2.0	3	6.0	2	4.0	1	2.0	0	-	1	2.0	0	-	1	2.0	1	2.0	0	-
Ear ducts																								
Carcinoma	6	12.0	7(8)	14.0	3	6.0	3	6.0	3	6.0	0	-	2	4.0	4	8.0	1	2.0	5	10.0	4	8.0	3	6.0
Nasal cavities																								
Carcinoma	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Oral cavity lips and tongue																								
Carcinoma	2	4.0	2	4.0	0	-	0	-	1	2.0	0	-	1	2.0	0	-	0	-	0	-	0	-	1	2.0
Pharynx																								
Carcinoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-
Larynx																								
Carcinoma	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-

— *Contintued*

TABLE 4. *Continued*

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

Site	Groups																							
	I: 2,500 mg/l				II: 1,500 mg/l				III: 500 mg/l				IV: 250 mg/l				V: 50 mg/l				VI: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Uterus & Vagina																								
Malignant Schwannoma			0	-			0	-			1	2.0			0	-			1	2.0			2	4.0
Vagina																								
Fibrosarcoma			0	-			0	-			1	2.0			0	-			0	-			0	-
Peritoneum																								
Fibroangioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Liposarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Mesothelioma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Pituitary gland																								
Adenoma	13	26.0	21	42.0	16	32.0	22	44.0	14	28.0	19	38.0	19	38.0	24	48.0	19	38.0	19	38.0	14	28.0	14	28.0
Thyroid gland																								
C-cell adenoma	1	2.0	3	6.0	3	6.0	4	8.0	3	6.0	3	6.0	2	4.0	2	4.0	3	6.0	3	6.0	2	4.0	0	-
Follicular carcinoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	1	2.0	0	-	0	-
C-cell carcinoma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Adrenal glands																								
Cortical adenoma	0	-	4	8.0	1	2.0	7(8)	14.0	1	2.0	2	4.0	0	-	4(5)	8.0	1	2.0	2(3)	4.0	0	-	5(6)	10.0
Pheochromocytoma	17(25)	34.0	8(14)	16.0	15(21)	30.0	12(16)	24.0	13(19)	26.0	12(15)	24.0	20(27)	40.0	11(15)	22.0	22(33)	44.0	10(13)	20.0	17(26)	34.0	10(14)	20.0
Fibroangioma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-
Cortical adenocarcinoma	0	-	0	-	0	-	2	4.0	0	-	1(2)	2.0	0	-	0	-	0	-	3(5)	6.0	0	-	0	-
Pheochromoblastoma	0	-	2	4.0	1	2.0	0	-	2	4.0	0	-	1	2.0	0	-	0	-	3(5)	6.0	0	-	0	-
Central nervous system																								
- Brain																								
Oligodendroglioma	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	2	4.0	0	-	0	-	1	2.0	1	2.0
- Meninges																								
Malignant meningioma	2	4.0	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0
Peripheral nervous system																								
- Major peripheral nerves																								
Neuroblastoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-
Malignant Schwannoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0

— *Contintued*

TABLE 4. *Continued*

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

Site	Groups																							
	I: 2,500 mg/l				II: 1,500 mg/l				III: 500 mg/l				IV: 250 mg/l				V: 50 mg/l				VI: 0 (control)			
	Male		Female		Male		Female		Male		Female		Male		Female		Male		Female		Male		Female	
Histotype	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Bones																								
- Head																								
Osteosarcoma	7	14.0	2	4.0	0	-	2	4.0	2	4.0	0	-	1	2.0	2	4.0	5	10.0	2	4.0	0	-	2	4.0
- Other																								
Osteosarcoma	0	-	0	-	1	2.0	0	-	0	-	1	2.0	0	-	0	-	2	4.0	0	-	1	2.0	0	-
Soft tissues																								
Lipoma	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Fibrosarcoma	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Angiosarcoma	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-
Malignant Schwannoma	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	1	2.0	1	2.0
Spleen																								
Fibroma	0	-	0	-	0	-	0	-	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Fibroangioma	0	-	0	-	2	4.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Subcutaneous lymph nodes																								
Fibroangioma	1	2.0	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-	0	-
Hemolymphoreticular tissues[b,c]																								
Lymphomas and leukemias	8	16.0	7	14.0	15(16)	30.0	3	6.0	9	18.0	4	8.0	10	20.0	8	16.0	14	28.0	5	10.0	6	12.0	2	4.0

[a] Numbers in parentheses indicate the total number of tumours; one animal can bear more than one tumor

[b] Including spleen, mediastinal, subcutaneous and mesenteric lymph nodes

[c] See table 6

TABLE 5. Long-term carcinogenicity bioassays on acetaldehyde administered with drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats

TOTAL MALIGNANT TUMORS

Group No.	Concentration (mg/l)	Animals		Malignant tumors			
				Tumor-bearing animals		Tumors	
		Sex	No.	No.	%	No.	Per 100 animals
I	2,500	M	50	23	**46.0**	33	**66.0** *
		F	50	21	42.0	39	**78.0** *
		M+F	100	44	44.0	72	**72.0**
II	1,500	M	50	21	42.0	27	**54.0**
		F	50	20	40.0	29	**58.0**
		M+F	100	41	41.0	56	**56.0**
III	500	M	50	20	40.0	23	**46.0**
		F	50	19	38.0	25	**50.0**
		M+F	100	39	39.0	48	**48.0**
IV	250	M	50	15	30.0	17	**34.0**
		F	50	26	52.0	33	**66.0**
		M+F	100	41	41.0	50	**50.0**
V	50	M	50	20	40.0	26	**52.0**
		F	50	24	48.0	41	**82.0** *
		M+F	100	44	44.0	67	**67.0**
VI	0	M	50	14	28.0	17	34.0
		F	50	20	40.0	23	46.0
		M+F	100	34	34.0	40	40.0

* $p<0.05$ using χ^2 test

(6) An increased incidence of interstitial cell adenomas of the testis was observed in all treated males except for those in group IV.

(7) An increased incidence of malignant uterine adenocarcinomas was detected in females treated with 250 mg/L of acetaldehyde.

(8) An increased incidence of cranial osteosarcomas occurred in males treated with the highest (P <0.05) and the lowest doses.

(9) An increased incidence of hemolymphoreticular neoplasias was observed to a varying degree in all treated groups (TABLE 6).

CONCLUSION

Formaldehyde, administered with drinking water, was shown to be carcinogenic based on an increased incidence of total malignant tumors and oncological lesions varying in site and histotype. Tumors included malignant mammary tumors, oncological lesions of the stomach and intestine, testicular interstitiae cell adenomas, and hemolymphoreticular neoplasias.

TABLE 6. Long-term carcinogenicity bioassays on acetaldehyde administered with drinking water supplied *ad libitum* to male (M) and female (F) Sprague-Dawley rats

HEMOLYMPHORETICULAR NEOPLASIAS

Group No.	Concentration (mg/l)	Animals		Animals with hemolymphoreticular neoplasias		
		Sex	No.	No	%[a]	Latency[b] (weeks)
I	2,500	M	50	8	**16.0**	105.2
		F	50	7	**14.0**	102.1
		M+F	100	15	**15.0**	103.7
II	1,500	M	50	15 [c]	**30.0**	103.7
		F	50	3	**6.0**	118.7
		M+F	100	18	**18.0**	111.2
III	500	M	50	9	**18.0**	82.2
		F	50	4	**8.0**	60.0
		M+F	100	13	**13.0**	71.1
IV	250	M	50	10	**20.0**	92.2
		F	50	8	**16.0**	91.4
		M+F	100	18	**18.0**	91.8
V	50	M	50	14	**28.0**	100.8
		F	50	5	**10.0**	104.2
		M+F	100	19	**19.0**	102.5
VI	0	M	50	6	12.0	105.7
		F	50	2	4.0	121.0
		M+F	100	8	8.0	113.4

[a] Percentages refer to the number of animals at start
[b] Age at death
[c] One animal bore a lymphoimmunoblastic lymphoma and histiocytic sarcoma

The results reported on acetaldehyde administered in drinking water indicate a carcinogenic effect of the compound on different organs and tissues, even though this effect is often not dose related.

Because of the extreme rarity of lesions of the stomach and intestine in nontreated animals,[21] their onset in formaldehyde-treated animals cannot be considered accidental and should not be underestimated. The production and use of oxygenated compounds as alternative fuels or as additives in reformulated gasolines was started with the intention of reducing the deleterious effects of vehicular fuels on the environmental quality. On the basis of these new data on the carcinogenicity of formaldehyde and acetaldehyde, and on what was already long known,[18,35] we think that this strategy should be carefully revaluated through an up-to-date analysis of risks and benefits.

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