concentration (MPC) of 1 ppm several hundredfold, but more recently styrene concentrations had been reduced in most places to less than the MPC. In this shop, there were elevated serum bilirubin concentrations in 20% of the examinees, elevated beta-lipoproteins in an unstated number (but not beyond physiologic limits), and an increase in SGPT in 13%. Leukopenia, stated to be a characteristic effect of styrene exposure, was found in 30% of these workers and in 18% of the controls; almost identical incidences of moderate leukopenia had been found in workers in the first plant and in their controls.

There were various effects attributed to functional changes in the liver. These changes did not reach pathological proportions in the majority of the cases; however, the changes suggested to Veretinskaya et al. [96] definite metabolic disturbances in the liver cells. The relationship of styrene to these effects is difficult to evaluate in this study because of the similarity in responses in areas with airborne styrene exposures believed almost always to be below the MPC (1 ppm) and in other areas with styrene exposures exceeding the MPC several hundredfold.

(b) Plastics Applications (Mainly Production of RP/C)

A clinical study of two factories in Czechoslovakia where reinforced plastics were made was reported in 1960 by Bardodej et al. [97]. In both facilities the styrene polyester resin was applied to wooden molds by hand; in one factory, however, the resin was sometimes sprayed on. Regular medical examinations (not specified) were given to the workers for a 3-year period.

Apparently, area rather than breathing zone air samples for the determination of styrene were collected on numerous occasions during an entire workshift in various work areas and analyzed by UV spectrophotometry [98], spectrophotometry after nitration [98], and polarography after formation of alpha-nitroso-beta-nitroethylbenzene [99]. The method of collection was not specified. The styrene concentrations found averaged about 50 ppm.

The investigators [97] measured benzoic acid and phenol in urine collected at the end of the workshifts from 58 workers and 23 controls. Elevated concentrations of substances measured as benzoic acid were demonstrated (800 vs. 400 mg/l). In a later publication, Bardodej and Bardodejova [100] stated that, with the analysis method used, both hippuric acid and mandelic acid were oxidized to benzoic acid. No increases in urinary phenols were found. There were no major medical findings in the RP/C workers except for four cases of dermatitis that were attributed to other agents. All but five of the workers reported developing increased fatigue and drowsiness toward the end of the workshift.

In 1972, Dzyuba [101] published the results of an investigation of a Russian reinforced plastics plant. Three groups of workers were evaluated over a 3-year period for neurological dysfunctions. Group I consisted of 70

workers whose primary exposure was to styrene; Group II comprised 30 workers exposed primarily to phenol and aniline. Occupational exposure levels were not given. Fifty workers from an instrument manufacturing facility served as a control group. Results of these neurological examinations are presented in Table IV-6.

TABLE IV-6

RESULTS OF NEUROLOGIC EXAMINATION OF WORKERS EXPOSED TO STYRENE

Type of Findings	Percent With Positive Findings		
	Group I Styrene	Group II Phenol/Aniline	Controls
Subjective findings			
Constant headache increasing throughout the day with fatigue and sleepiness increasing after workshift	97	30	8
Nausea, dizziness, heart pain	66	20	2
Objective findings			
Emotional instability	91	60	10
Asthenia (weakness)	23	*	4
Cerebral nerve insufficiency	26	10	*
Unsteadiness in Romberg test	69	*	*
Finger tremors of extended hand	61	20	4
Increased tendon reflexes	100	33	16
Anisoreflexia	11	*	*
Sympathetic nervous system suppression	100	*	*

*Data was not given
Taken from Dzyuba [101]

Among Group I workers, a so-called asthenic-vegetative syndrome was observed in 22% in the first year, 64% in the second year, and 92% in the third year [101]. This neurasthenic syndrome was found in 52% of Group II (workers exposed to phenol and aniline) and in 5% of the controls. When the workers in Group I returned to work after having several days off, only a slight decrease in the manifestation of this condition was noticed, but it substantially decreased or disappeared in Group II workers.

In a factory in France where electronic filters for washing machines were produced, Bernard [102] in 1966 found evidence of blood abnormalities and CNS disturbances in workers. The filters were made by placing a resistor and a capacitor in a flexible plastic mold filled with a solution of polyester in styrene monomer. No information was presented concerning airborne concentrations at the time of the study, but after the introduction of ventilation equipment that changed the room air 50 times per hour, breathing zone concentrations of styrene were 40 ppm for workers who molded

parts and 100 ppm for those who removed the mold. Sampling and analytical methods were not described, nor were concentrations of trichloroethylene used as a cleaning solvent reported [102].

The workers encountered few skin problems, but used a skin cream to prevent skin dryness. There were, however, frequent reports of anorexia, asthenia (weakness), and headache. One worker also complained of nausea, vomiting, gastralgia, and vertigo. Subicteric conjunctivitis, slight leukopenia, neutropenia, lymphocytosis, and a slight anemia were found in this worker. Three other workers had severe anemia (red blood cell count (RBC) was about 3,000,000) that disappeared rapidly after the workers were given jobs away from possible styrene exposure [102]. Whether these effects preceded or followed installation of controls was not reported, though it seems likely that they were the motivation for the changes. Because of the absence of precise information concerning this sequence of events, this study cannot be used to help establish a recommended exposure limit.

Studies of four workshops in Sweden where reinforced plastics were produced were reported in 1972 by Gotell et al. [35] and by Axelson and Gustavson [103] in 1978. The 17 male workers studied had a median age of 28 years (range, 21-57 years) and duration of styrene exposure ranging from a few days to 12 years. Procedures used to manufacture reinforced plastic products involved coating wooden molds with wax, covering the molds with fibrous glass, and applying the styrene-modified polyester resin by either hand rolling or a combination of spraying and hand rolling. Based on their observations, Gotell et al. [35] judged the skin absorption of styrene under these conditions to be fairly low. The studies were conducted in the spring and fall so that the workers could be examined during moderate climatic conditions. Measurements were taken on a day in the middle of the week.

Styrene concentrations in the workers' breathing zones were determined by several methods. In two factories, the samples were collected for 30-minute periods in impingers containing ethanol and analyzed by gas chromatography; in another factory, air was sampled simultaneously by absorption in ethanol and analyzed by gas chromatography, by colorimetric indicator tubes, and by a combustible gas indicator. In the other factory, samples were collected for 24-66 minutes on charcoal and analyzed by gas chromatography. Although Gotell et al. [35] reported that some workers were exposed at 500-800 ppm for certain 1-hour periods, the 8-hour TWA exposure concentrations calculated for each worker ranged from 17-292 ppm. However, due to the nature of these processes, high concentrations of styrene vapor were often briefly encountered; in this study, concentrations of about 1,500 ppm were found for periods of 5-10 minutes. The workers were divided into three groups based on TWA exposures to styrene: (I) 235-292 ppm, (II) 89-139 ppm, and (III) 17-32 ppm.

All workers were given a brief neurological examination that included an evaluation of the knee and Achilles tendon reflexes, pupillary light

reflexes, and vibration sensibility; results of these tests were all normal. A modified Romberg test (one foot in front of the other) was given before and after work. Two workers showed a slightly unsteady Romberg test in the morning before work but not after exposure. A comparison group of 17 men from a motor workshop matched in age to the study group was used for the reaction time test. Reaction times of workers exposed to styrene at concentrations greater than 235 ppm were longer than those of workers exposed at less than 139 ppm and those of the age-matched control subjects, but statistical treatment of these data was not described. Differences between pre- and post-shift reaction time measurements were not significant in any group [35].

Lung function tests consisting of forced expiratory volume in one second (FEV_1) and vital capacity were normal in the morning and did not change during the day [35,103]. Blood samples were taken from 35 male workers in one plant, who had been exposed to less than about 100 ppm styrene. When compared with a group of 12 healthy males in a manufacturing industry, elevated levels of the liver enzymes aspartate-amino-tranferase (ASAT) and alanine-amino-tranferase (ALAT) were found in the styrene workers [103].

Complaints of irritation of the eyes and nasopharynx were common [35]. The concentration of styrene giving rise to these complaints varied from person to person. The tolerance to the irritant effects of styrene vapor may increase with exposure time, because workers complained of only minor to moderate irritation during exposures at 500-800 ppm for several hours, while the investigators [35] who were in the plant only during the study could not tolerate such concentrations for more than 1 or 2 minutes. It was also mentioned that several workers had quit their jobs because of dermatosis. Gotell et al. [35] speculated that the agents causing the dermatosis could have been the peroxide or cobalt compounds used as accelerators and catalysts.

In 1973, Bodner et al. [104] conducted a health hazard survey of a U.S. factory where reinforced plastic bathtubs were manufactured. The work involved spraying a fibrous glass-polyester resin mixture onto molds and then hand-rolling, laminating, and shaping. Although local exhaust ventilation was used in some of the operations, the makeup air was insufficient; thus, some workers wore respirators. Workers were exposed to styrene at 45-550 ppm as determined by breathing zone samples collected on charcoal for unspecified periods. In one operation where a foaming agent was sprayed, workers were exposed to methylene bisphenyl isocyanate (MDI) at 0.02-0.27 mg/cu m. The OSHA ceiling standard for MDI is 0.2 mg/cu m (CFR 1910.1000). Exposure to airborne fibrous glass was not measured.

Thirty-five workers (21 women, 14 men) were interviewed; their average age was 34 years. The average length of employment with the company was 3.7 years; however, this particular facility had only been operating for about 2 years. Sixteen of the workers (46%) smoked at least 1/2 pack of cigarettes a day. Four of the office workers, one man and three women, were used as a comparison group for the investigation. Complaints of some form of eye,

nose, or throat irritation were made by 34 of the 35 workers (97%) examined; 17 (49%) complained of wheezing, shortness of breath, or chest tightness, and 14 (40%) complained of skin rashes, hives, darkening skin color, or skin sores. Other complaints included nose bleeds, anorexia, excessive thirst, numbness of extremities, frequent headaches, occasional vomiting, and upset stomachs. None of these complaints were made by the four office workers who were interviewed for comparison. Since MDI was also present and is known to cause irritant effects similar to those observed among these workers, a direct attribution to styrene cannot be made.

In 1966, Simko et al. [105] reported the effects of styrene on 128 workers (101 women, 27 men) in three Czechoslovakian factories where reinforced plastics (chairs, small parts) were made. Average work experience was 1.8 years, with 20 of the workers having had more than 3 years of exposure. Operations were essentially the same in all three factories, and no respiratory protection was used. Styrene exposures in the three factories ranged from 4-195 ppm; methods of sampling and analysis were not reported. Table IV-7 presents the environmental styrene and urinary mandelic acid concentrations found.

TABLE IV-7

ENVIRONMENTAL STYRENE AND URINARY MANDELIC ACID CONCENTRATIONS

Factory	Year(s) of Exposure	Year(s) Measured	Styrene ppm	Mandelic Acid mg/l
I	5	1960-65	8-195	72-2,620
II	1	1965	5-165	80-2,100
III	1	1965	4-41	36-1,215

Taken from Simko et al. [105]

Clinical and neurological examinations of the workers at the beginning of the study included gynecologic examinations of the women and determinations of SGOT, SGPT, serum cholesterol, albumin, bilirubin, and urinary creatinine in all the workers. In factories I and II, where worker exposure to styrene was the greatest, all results from measurements of blood serum components were within normal limits; no signs of liver or gall bladder injury were found. The subjective symptoms reported included headache (20%), tiredness (15%), and drowsiness at work (13%). Hypertension was found in 23% of the workers. Simko et al. [105] concluded that the primary health hazard of styrene exposure was development of the

neurasthenic syndrome (a term not defined by the investigators), which was found in 33% of the 20 workers exposed to styrene for more than 3 years, and in 13% of the workers overall.

In 1962, Klimkova-Deutschova [106] reported neurological studies of 35 styrene-exposed workers (30 women, 5 men) in two Czechoslovakian RP/C plants. The workers had an average age of 38.5 years and an average exposure duration of 1.9 years. Styrene exposures in the two facilities were 43-131 ppm and 19-98 ppm. Dibutyl phthalate, trichloroethylene, and cobalt naphthenate were also present in the work environment. A catalyst, which was translated as being cyclohexyl peroxide, was also present. Methods of sampling and analysis were not reported. Workers were excluded in which earlier diseases such as hepatitis or working with other solvents could have played a role [107]. Each worker was given a clinical examination, and 17 had EEGs recorded.

The most frequent complaints were fatigue (41% of the workers), headaches (51%), and drowsiness with increased need for sleep (34%). Neurological effects reported included cranial nerve disturbances (91% of the workers), diminished reflexes (86%), and autonomic nervous system disorders (34%). Performance of a Romberg test (maintaining balance with eyes closed) and a Hautant test (walking with eyes closed) was impaired in 83% of the styrene workers. Of the 18 EEGs of 17 workers, 5 EEGs were judged to be normal [106].

A subsequent report in 1973 by Klimkova-Deutschova et al. [108] described findings in 21 workers examined for a period of three years from the start of exposure. The age distribution showed a marked predominance in the range of 40-49 years. The authors [108] found an increase in reports of fatigue, drowsiness, headaches, symptoms indicative of autonomic nervous system disorders, and hyporeflexia. Also, some workers had abnormal EEGs after 3 years of styrene exposure, whereas no abnormal EEGs were found during the pre-exposure examinations [108]. These effects were manifested in those workers with mandelic acid levels greater than 600 mg/l. Workplace exposure levels to styrene or other substances were not reported.

In 1967, Huzl et al. [109] reported an investigation of five separate RP/C facilities in Czechoslovakia. The average age of the 55 workers (34 women, 21 men) was 38.4 years; average exposure duration was 1 year. In four of the five facilities investigated, styrene polyester resin was applied by hand; the material was sprayed in the fifth facility. Only "primitive" engineering controls were in use in the four areas where the resin was spread by hand; gloves were used only occasionally. In addition to styrene, the workers were also exposed to cyclohexene peroxide, a resin catalyst, and to cobalt naphthenate, an accelerator. Styrene concentrations in the four hand lay-up facilities were found to be 47-94 ppm. In the fifth facility (spray-up), the ventilation was much better, and styrene concentrations were about 6 ppm. A comparison group examined in an outpatient clinic had previous occupational exposure to a variety of substances, including organic solvents, but not styrene. Medical histories

were taken and liver function tests performed. Based on urine specimens collected at the end of the workweek, Huzl et al. [109] found no simple statistical correlation between urinary mandelic acid levels and styrene exposure. However, Huzl et al. [109] believed that a urinary mandelic acid concentration greater than 300 mg/l indicated high occupational exposure to styrene. Mandelic acid values greater than 300 mg/l were found in 24% of the workers examined. The subjective complaints and objective findings of workers from the hand lay-up sites are presented in Table IV-8.

TABLE IV-8

SUBJECTIVE COMPLAINTS AND OBJECTIVE FINDINGS IN STYRENE-EXPOSED RP/C WORKERS FROM FOUR HAND LAY-UP SITES

Complaint or Finding	Percent of Workers	Percent of Controls
Headaches	36.4	21.8
Drowsiness, fatigue	23.6	20.0
Dyspeptic problems	10.9	*
Occupational eczema	7.3	*
Prolonged Weltman's reaction	48.1	23.6
Hyperbilirubinemia	17.1	*

*Data was not given
Taken from Huzl et al. [109]

Medical histories of the workers, according to Huzl et al. [109], revealed no cause other than styrene exposure for the prolonged Weltman's reactions noted, a nonspecific test which the investigators considered not necessarily indicative of liver disease. However, Huzl et al. [109] recommended that prospective workers with a history of liver disease and hepatitis not be allowed to work in areas with potential exposure to styrene.

In 1964, Zielhuis et al. [110] described a clinical study of workers in three factories in the Netherlands where reinforced plastic items (including boats, automobile bodies, and small objects) were manufactured. Boats were constructed using techniques in which layers of fibrous glass were impregnated by hand with a styrene-polyester mixture. The authors [110] noted that as the boats reached the final stages of construction, and the work area became more enclosed, the ventilation decreased. Workers had been exposed to styrene for 2-5 years. Breathing zone concentrations of styrene found during various operations in the factories, together with the authors' [110] classification of workers by job description, are listed in Table IV-9. However, precise information concerning methods of sampling was not provided by the investigators. Three groups of unexposed workers served as controls.

TABLE IV-9

AVERAGE STYRENE CONCENTRATIONS AT THREE FACTORIES

Factory	Group	Number of Workers	Worksite	Styrene ppm
A	Aa	5	Pilot plant Boat construction	24-94
	Ab	6	Pilot plant Laboratory worker	7
	Ac	5	Pilot plant Maintenance	Negligible
B	Ba	8	Steel Works Boat Construction	24-94
	Bb	8	Steel Works Car body construction	24-94
	Bc	22	Steel Works Small object construction	7
	Bd	21	Steel Works Upholsterers and steel workers	Negligible
C	Ca	6	Ship Wharf Boat construction	24-94
	Cb	5	Ship Wharf Carpenters	Negligible

Taken from Zielhuis et al. [110]

Although physical examinations (including blood counts, organ function tests, and urinalysis) revealed no abnormalities that could be attributed to the work situation, many of the workers in factories B and C reported having symptoms during their work with styrene. Workers' symptoms in factories B and C are presented in Table IV-10. Data reported in Table IV-10 were obtained through the use of a questionnaire given at the end of the shift on the days that the air samples were taken. The number of symptoms noted by the workers decreased with decreasing exposure. The authors [110]

concluded that a relationship existed between the workers' feelings of discomfort and styrene exposure. The most striking symptoms of discomfort were mucosal irritation and drowsiness. The workers reported that symptoms disappeared rapidly when they left the work area.

TABLE IV-10

SYMPTOMS OF WORKERS AT TWO PLASTICS PLANTS

Symptoms	Percentage* of Workers Reporting Symptoms Factory and Group				
	Ba N=8	Bb N=8	Bc N=22	Ca N=6	Cb N=5
Drowsiness	90	90	70	50	0
Apathy	70	70	50	10	0
Mental fatigue	50	90	30	0	0
Anorexia	70	30	10	50	0
Dizziness	30	70	10	50	10
Headache	70	30	50	30	30
Feeling groggy	10	50	10	30	0
Nervousness	30	10	10	10	10
Agitation	30	30	10	0	10
Nervous tension	50	30	50	0	10
Gastric pain	10	10	10	10	0
Tearflow	90	70	50	0	10
Eye irritation	90	90	50	90	0
Sneezing/coughing	70	30	30	70	10

*Actual value is the range of the tabulated value $\pm$10; e.g., 90 means that the percentage of workers reporting symptoms was between 80 and 100.
Taken from Zielhuis et al. [110]

In 1975, Gamberale et al. [111] studied 106 workers in four Swedish RP/C boat plants. Styrene-exposed workers included plastics workers and mechanics. The comparison group consisted of 36 workers in the same locality as two of the boat plants, not exposed to styrene. Styrene exposures varied from 10-120 ppm for the plastics workers and from 6-60 for the mechanics who were exposed at levels up to 312 ppm for 24 minutes, while working in narrow spaces.

Reaction time was measured by having the subject rest his wrist and forearm on a table, with his fingertips in contact with a pressure plate. The time between seeing a light signal and pressing the plate with the fingertips was the reaction time, expressed as the mean of 160 responses over a 10-minute period. Workers exposed to styrene had significantly longer reaction times than age-matched controls. This difference

was evident before work started in the morning, and showed a tendency to increase during the workday [111]. The data were not collected in a manner to allow inferences on concentration-response relationships to be made.

In 1977, Bergman and Lindberg [112] conducted industrial hygiene and medical studies at four Swedish factories where boats were made of fibrous glass-reinforced plastics. TWA styrene exposures ranged from 3-312 ppm for 39 of the workers with 29 of the workers having TWA exposures less than 100 ppm. Samples were collected with charcoal tubes or glass syringes and analyzed with a gas chromatograph. Interviews and medical examinations were given to 81 workers exposed to styrene and 32 workers with no styrene exposures. The 32 controls included some workers from other industries. Subjective symptoms elicited from the 81 styrene workers included fatigue that was not attributed to the heaviness or intensity of the work (60%), confusion or dizziness (38%), nausea (14%), headache (25%), and poor memory (21%). Only one of the controls mentioned any of the above subjective symptoms in connection with their work.

Rosensteel and Meyer [113] evaluated health hazards in a U.S. facility where reinforced plastic boats were manufactured. An initial survey in 1975 demonstrated that some exposure concentrations of styrene were greater than 100 ppm and that the workers were also exposed to acetone, methylene chloride, methyl ethyl ketone, naphtha, toluene, xylene, and asbestos. Twenty-one personal air samples in the lamination area were obtained in 1976 by the collection on charcoal for about 7 hours. TWA styrene exposures averaged 69 ppm and ranged from 9-111 ppm. These same workers also had average TWA exposures of 56 ppm acetone and 2 ppm methylene chloride (OSHA standards are 1,000 ppm and 500 ppm, respectively).

During one of the surveys, 9 of 14 workers interviewed reported symptoms of styrene exposure on the day of the interview, and all 14 stated that they had, at some time in this factory, experienced symptoms of styrene exposure such as eye and respiratory irritation [113]. These data are presented in Table IV-11.

TABLE IV-11

WORKERS WHO EXPERIENCED EFFECTS WHILE MANUFACTURING REINFORCED PLASTICS

Symptom	No. of Workers	Percent
Eye irritation	13	93
Skin rash	8	57
Nose irritation	7	50
Headache	7	50
Throat irritation	4	29
Chest pain	4	29
Dizziness	2	14
Fatigue/drowsiness	2	14
Cracked hands	2	14
Irritability/nervousness	1	7
Cough	1	7

Adapted from Rosensteel and Meyer [113]

During the last survey in 1976, serial detector tube sampling procedures were used by Rosensteel and Meyer [113] to determine 5-minute peak concentrations of styrene in workers' breathing zones; concentrations of 50-400 ppm were found. Analyses of samples collected during hull-stiffening, hull spraying, fibrous glass application, and hull roll-out areas, showed that styrene concentrations for 5-minute periods ranged from 200-400 ppm.

During the medical study, 41 workers were examined; 22 workers (16 men and 6 women with average ages of 28.7 years and 35.2 years, respectively) were from the lamination area. The other 19 examined for comparison were from other areas of the factory with little or no styrene exposure (13 men, average age 29.6 years; 6 women, average age 24.0 years). Workers from the lamination areas were matched with the control group on the basis of age, sex, and smoking history. However, because of the frequency of complaints of upper respiratory and eye irritation expressed by this group, perhaps from exposure to other irritants, their use as a comparison group may have been inappropriate. Air samples obtained over 6-7.5 hours from 10 of the 19 controls indicated TWA exposures to styrene below the level of detection for 6, 9-14 ppm for 3, and 62 ppm for 1. A medical history was obtained from each worker, and a physical and clinical examination of each worker was made. All results from blood analyses were within normal limits. Although the lamination workers had significantly elevated concentrations ($p<0.05$) of serum uric acid with respect to the comparison group, the levels were within normal limits [113].

At the time of the evaluation, all 22 lamination workers had work-related symptoms as did 55% of the comparison group. Subjective complaints that the workers reported occurring during their work with styrene and objective findings at the time of the study are presented in Table IV-12.

TABLE IV-12

SUBJECTIVE COMPLAINTS AND OBJECTIVE FINDINGS OF WORKERS WHO MADE REINFORCED PLASTICS

	Percentage of Workers With Complaints	
	Exposed	Control
Subjective complaints		
Eye irritation	45	26
Nose irritation	95	37
Nasal congestion	82	37
Cough	23	21
Chest tightness	23	16
Wheezing	18	5
Shortness of breath	54	11
Nausea and vomiting	0	0
Muscle weakness	4	5
Fatigue	36	5
Headache	14	5
Objective findings		
Skin rash	14	0
Conjunctival erythema	41	5
Nasal erythema	86	63
Mouth and throat erythema	45	32
Abnormal thyroid size	14	5
Rales and wheezes	9	0

Taken from Rosensteel and Meyer [113]

The average maximal mid-expiratory flow-rate (MMEF) of lamination workers that smoked was significantly less ($p<0.05$) at the end of the shift than at the beginning when compared with the smokers in the control group. Pulmonary function tests (FVC, FEV_1) did not reveal any other significant differences between styrene-exposed workers and those used for comparison. A number of suggestions were made concerning improvement of work practices and engineering controls; one recommendation made was that the workers in the high exposure areas be discouraged from smoking [113].

In 1968, Matsushita et al. [59] reported on 14 male production workers and 10 male office workers used as controls from a factory where plywood was laminated with a styrene-polyester resin. The men had been employed for 3-6 years; their average age was 29.8 years. Concentrations of styrene associated with different operations in the coating process ranged from 50-600 ppm; in other areas, no styrene was detected. Other substances including toluene normally were detected only in trace amounts. Toluene (50-550 ppm), ethyl acetate (3-160 ppm), and methanol (100-1,000 ppm) were present in the air when the equipment was cleaned daily during a 30-40 minute period; the workers took turns cleaning the equipment. The method of sampling and the number of samples taken were not reported. However, the substances were analyzed by gas chromatography.

Using questionnaires and interviews, it was found that all 14 of the styrene-exposed workers experienced throat pain, that 12 of them (86%) tired easily, and that 11 (79%) caught colds easily. Eye pain, bad breath during work, heaviness of the head, and feeling unwell during work were each reported by 10 workers (71%). Nose pain, headache, dizziness, palpitation and an oppressive sensation of the chest, anorexia, drowsiness during work, loss of weight, and severe forgetfulness were reported by 6-9 of the styrene-exposed workers (43-64%). Heaviness of the head (three complaints) was the most common complaint of the comparison group of 10 office workers; all other complaints were reported by only 1 or 2 of this group. While 8 of the styrene-exposed workers (57%) complained of decreased vision, ophthalmologic examinations confirmed this in only 4. Contracted fields of vision without fundus changes were found in 7. Twelve of the styrene workers were examined by a neurologist; increased knee jerk reflexes were found in 9, hyperreflexia of the Achilles tendon in 5, hyperreflexia of the upper limbs in 3, and sensation disorders in 4. Four styrene workers with severe symptoms were given EEG examinations; there were no abnormal findings. Three workers with severe symptoms had EMG abnormalities. Matsushita et al. [59] concluded that styrene had a major role in causing the effects found in these workers [59].

A series of reports by a group of Polish investigators [114,115,116,117,118,119,120] discussed results of studies of workers exposed to styrene during production of reinforced plastics. One group of 101 workers from two factories had been exposed for about a year; their styrene exposures in 1972 were about 25-75 ppm [115]. Twenty-one workers in another group had been exposed to styrene for about 10 years at concentrations that were about 75 ppm. Methods of sampling were not reported; analysis, however, was by colorimetric methods.

Urine samples were collected for determination of mandelic acid by the nonspecific, colorimetric method of Ohtsuji and Ikeda [121] for the same period during which the air samples were collected. Mandelic acid concentrations of up to 150 mg/l were considered normal, and concentrations greater than 400 mg/l were considered as definite indications of exposure to styrene. Urinary mandelic acid concentrations of the 101 workers who had been exposed for about 1 year (short-term workers) averaged 287 mg/l, with

concentrations of about 150 mg/l being found in 50 workers, 150-300 mg/l in 8 workers, and 300-494 mg/l in the remaining 43 workers [114]. The average concentration of mandelic acid in the urine of the 21 workers who had been exposed about 10 years (long-term workers) was 504 mg/l (range, 325-625 mg/l) [114,116]. Hippuric acid concentrations in the urine did not exceed the normal range [116].

In the 101 short-term workers, four cases of upper respiratory catarrh were noted by Chmielewski and Renke [114]. In 26 workers there were signs that the investigators classified as vegetative nervous system disturbances, which included skin marbleization, asymmetric body warming, prolonged blood vessel filling time, diaphoresis, excitability, hypoesthesia, whitening of fingers, trembling of hands, weakened reactions, cat's eye pupils, and nystagmus [115].

Clinical studies of these workers were performed that included blood cell and platelet counts [117]; examination of serum proteins, lipids, enzymes, bilirubin, and cholesterol [119]; glucose tolerance [116,120]; and 24-hour excretion of 17-ketosteroids [115,116,120]. Pulmonary function [114], blood clotting [117], and EEGs [118] were also studied in the long-term workers, and EEGs were studied in 43 of the short-term workers [118].

In the short-term workers, the significant clinical finding was that abnormal EEGs were noted in 31 of 43 examinations [118]. The abnormal EEGs showed discharges of sharp waves and high-voltage slow waves in the temporal regions that intensified with hyperventilation. Almost all short-term workers complained of "neurotic troubles." Signs such as intensified or abated deep reflexes, vestigial nystagmus, and tremors were interpreted by Dolmierski et al. [118] as being minor symptoms indicating that the nervous system was affected.

Average blood glucose in 53 short-term workers in one of the factories studied was lower than in a control group of 20 individuals [115,120]. Subsequently, blood glucose concentrations were studied in 40 workers in the other factory and in 18 controls using 50 g of glucose taken orally, followed by another 50 g of glucose taken orally 90 minutes later [115,120]. The results showed a heightened glucose tolerance in persons exposed to styrene. There was an indication that a tendency toward increased glucose tolerance was greater among 20 of the workers with concentrations of urinary mandelic acid greater than 400 mg/l and among 27 workers with a reduced 24-hour excretion of 17-ketosteroids (i.e., 5.78 mg/24 h on the average).

In another investigation of these same workers, Chmielewski [116] obtained somewhat different glucose tolerance test results from 21 long-term workers as compared to 40 short-term workers. The maximum concentration of blood glucose in the long-term workers occurred half an hour later on the average after administering glucose and did not rise after the second dose. The average response was due to three classes of blood glucose curves:

hypoglycemic, normal, and hyperglycemic. However, only the hypoglycemic workers responded to the second dose of glucose or to cortisone administered before the test. All workers had glucose assimilation coefficients below control values [116].

Other aberrant clinical findings in the long-term workers included reduced FEV_1/FVC in 4 of 21 workers examined [114], as well as reduced amounts of serum alpha- and beta-lipoproteins [119]. Twelve of the long-term workers had normal EEG patterns, two had borderline patterns, and four had abnormal patterns that were symmetrical in the temporal regions and characterized by low to medium voltage theta waves [118]. Most complaints by workers with abnormal EEGs were of fatigue, a sensation of weakness, and drowsiness. Compared with a nonexposed control group, the long-term group had a reduced number of blood platelets, an increased coagulation time, a reduction in prothrombin ratio, a shorter euglobulin fibrinolysis time, and an increase in blood platelet adhesion that was significantly correlated with beta-lipoprotein concentrations [117]. It was not mentioned whether the comparison group was age-matched to the exposure group, an important point in light of the comparison of serum lipoprotein levels between short-term and long-term workers; thus, the effect of age on these comparisons is not clear.

Occupational dermatoses were reported in 1975 by Golebiowska-Podgorczyk [122] among 70 workers in a Polish factory where boats were built from fibrous glass-reinforced plastic. The workers ranged from 20-64 years of age and had worked at this factory for 1-15 years. The group studied was composed of 15 carpenters, 37 molder-artists, 13 iron workers, and 5 unskilled laborers. The workers were exposed to a variety of polyester resins dissolved in styrene, epoxide resins dissolved in acetone or ether, hardeners that included cyclohexanone hyperoxide, dibutyl phthalate, methyl ethyl glycol hydroxide, and triethylenetetramine, cobalt naphthenate, organic dyes, and various glues. No industrial hygiene monitoring was conducted. None of the workers had a history of, or a predisposition to, any allergic disease, but 18 experienced some type of dermal trauma as a result of their employment. Three of these 18 worked with fibrous glass. No positive reactions to styrene or polyester resins were found by patch testing, although positive reactions were found to the epoxide resins and some of the hardeners. Golebiowska-Podgorczyk [122] found four cases of excessively dry, chapped, and cracked hands and suggested this might have been caused by the defatting action of styrene.

In 1978, Rosen et al. [123] reported, in what was described as a pilot study, the results of neurological examinations of 33 workers from 3 different worksites in Sweden. Thirteen of the workers (aged 24-66 years) had been employed for 1-21 years in the polyester resin boat industry (Group I). Exposures to styrene had been measured the previous year and averaged 125 ppm (range 74-175 ppm), but Rosen et al. [123] stated that during previous years the exposures were significantly higher. Ten of the workers (aged 23-54) had been involved in the production of polyester resin cisterns for 2-14 years (Group II); their average exposure based on measurements

taken 2 years earlier was 47 ppm styrene. The remaining ten workers studied (aged 29-65) were exposed to less than 5 ppm styrene while producing polystyrene for 5-15 years (Group III).

Results of the study were compared with two "control" groups. One group (called the "normal" group) consisted of six men from a hospital transportation service never significantly exposed to organic solvents; the other group (called the "reference" group) consisted of 17 men, many of whom were former painters, reported to have signs of chronic intoxication due to exposure to a mixture of organic solvents. All examinations of styrene workers were performed at least 48 hours after the last exposure, to avoid possible acute effects of styrene on the results. The reported incidences of unusual tiredness, reduced short-term memory, giddiness, headache, paresthesia in fingers or toes, conjunctivitis, throat irritation, and a minor decrease of muscle stretch reflexes were higher in the styrene-exposed workers than in the "normal" group. These results were roughly dose-dependent with the reported effects occurring most among the workers in the higher exposure group (I), but also frequently occurring in the medium exposure group (II), and occasionally among the polystyrene workers, who had the lowest styrene exposures (Group III).

Signs of polyneuropathy or CNS lesions were not found in the styrene-exposed workers. Motor conduction velocities of median, ulnar, fibular, and posterior tibial nerves were recorded, as well as sensory action potential of the median and ulnar nerves. An EEG examination was made while the subjects were awake, and included studies of the effects of arousal, hyperventilation, and intermittent light [123]. No differences in motor conduction velocities were found between the groups. Ten of the 33 styrene-exposed workers had evidence of a mild sensory neuropathy with polyphasic sensory responses of a low amplitude; a similar pattern was found in many of the workers in the solvent-exposed "reference" group. The 10 affected styrene workers were more heavily exposed than those not having signs of neuropathy, but they were also older and had more years of exposure. Based on corrections for age published by others, the investigators [123] believed that age alone could not have accounted for the effects, but speculated that the effects of age and styrene exposure may have been synergistic. Eight of these ten workers had EEG changes consisting of fast activity within the rostral and central parts of the hemispheres; similar changes were common in the solvent-exposed "reference" group, but were seen in only one of the other 23 styrene-exposed workers. Unexpectedly, the highest frequency of diffuse slow activity over both hemispheres was seen in those workers (Group III) exposed at the lowest styrene concentrations. The investigators [123] suggested that the additional exposure of Group III to isopentane might be relevant, but no isopentane determinations were made.

In 1976-1978, a series of reports [124,125,126,127,128] were written by a group of Finnish investigators who examined the results of psychological and neurophysiological tests of 96 workers from 24 different factories where reinforced plastics were made. The workers were 16-54 years old

(mean age, 29.6) and had been exposed to styrene for as little as 6 months or as long as 14 years (mean duration, 5 years). Styrene exposures of these workers who used their hands to spread polyester resin, were evaluated from urinary mandelic acid concentrations determined by the colorimetric method of Ohtsuji and Ikeda [121] in about half of the workers, and by the gas chromatographic method of Engstrom and Rantanen [77] in the remainder. Results from the two methods were transformed to a common scale [79,127]. Urine specimens from each worker were collected at the end of an 8-hour workday, once a week, on a different day each week for 5 weeks. The mean of the five determinations of mandelic acid was used as an index of exposure for each individual. The range of the individual means was 7-4,715 mg/l; the group median was 808 mg/l.

Neurophysiological findings of these workers were reported by Seppalainen and Harkonen [124], psychological functions and data relating to alcohol consumption were reported by Lindstrom et al. [127,128], and effects on the nervous system were reported by Harkonen [126] and Harkonen et al. [125]. EEGs were recorded for all 96 of the workers at least 20 hours after the last exposure to styrene. The recording period lasted 30 minutes, and a 3-minute hyperventilation and a photic stimulator were used as EEG pattern activators. All EEGs were interpreted by one investigator who had no knowledge of the subjects' exposure histories. The findings were normal in 73 of the workers. Abnormal EEG patterns were found in 23 workers with 14 having local slow activity, 8 with diffuse theta activity, and 2 with bilateral spike and wave discharges (1 worker had 2 abnormalities). Based on the available literature on EEGs, about 10% abnormal patterns were expected by the investigators [124] for a normal population. The incidence of abnormal EEGs found (24%) was significantly greater ($p<0.01$) than expected. Of the 23 workers who had abnormal EEGs, 19 had urinary mandelic acid concentrations greater than 700 mg/l. In a subsequent analysis of the data, Harkonen et al. [125] presented evidence that this concentration of mandelic acid corresponded on the average to an 8-hour TWA styrene exposure of 31 ppm.

Possible peripheral nerve dysfunction in 40 of the workers (average age 29.6 years) in the study group with the most severe complaints was also investigated by Seppalainen and Harkonen [124] using nerve conduction velocity measurements. Thirty healthy, age-matched men with no history of occupational exposure to toxic chemicals were used for comparison. An electromyograph was used to measure maximal motor conduction velocity of the median, ulnar, deep peroneal, and posterior tibial nerves; conduction velocity of slower motor fibers of the ulnar and deep peroneal nerves; and the sensory conduction velocity of the median and ulnar nerves. There were no statistically significant differences between the styrene-exposed workers and the comparison group in average nerve conduction velocities. However, slightly abnormal conduction velocities were found in 9 of the 40 workers; the criteria used for judgment were not stated. Five of these workers displayed mononeuropathy, and the other four exhibited polyneuropathy. In four of the workers with mononeuropathy there were possible causes other than styrene exposure; in the fifth worker, no other cause for the

neuropathy could be found. In three of the workers with polyneuropathy, no cause other than styrene exposure could be found. There was no association between urinary mandelic acid concentration and nerve conduction velocity in the four subjects with unexplained abnormalities.

To study psychological functions in these workers, Lindstrom et al. [127] selected a comparison group of 43 men who worked with reinforced concrete. The comparison group was similar to the styrene-exposed group in age distribution, educational level, and geographic location. Psychological functions which were measured by 30 tests included general intelligence (5 tests), visuomotor speed (8 tests), visuomotor accuracy (3 tests), memory (5 tests), vigilance (2 tests), psychomotor performance (3 tests), and personality (4 tests). Comparisons were made between averages of RP/C workers and the comparison group scores. When comparing these two groups, performances on two tests, one a measure of visuomotor accuracy and the other a measure of response time in Rorschach inkblot tests, were significantly impaired ($p<0.05$) in the RP/C workers. When the 19 styrene-exposed workers with urinary mandelic acid concentrations greater than 1,762 mg/l were compared with the 36 workers who had urinary mandelic acid concentrations less than 674 mg/l, greater visuomotor inaccuracy and poorer psychomotor performance were found in the workers with the higher mandelic acid concentrations. Step-wise multiple regression analyses were used to study relationships of variables to urinary mandelic acid concentrations, duration of exposures, and the combination of mandelic acid concentration and duration of exposure. Duration of exposure had only a slight relationship to disturbances in psychological functions. One measure of visuomotor speed and one measure of visual memory correlated with the duration of exposure; their joint partial correlation with duration of exposure, when the effect of age was eliminated, was 0.28 ($p<0.05$). High urinary mandelic acid concentration was related to visuomotor inaccuracy ($p<0.01$), and it had a slight relationship to vigilance and psychomotor performance ($p<0.13$). The product of duration of exposure and urinary mandelic acid concentration was related to visuomotor inaccuracy and one Rorschach variable, long latency time in answering.

For the study of subjective symptoms experienced by these workers, Harkonen [126] selected a comparison group of male postal workers and electricians (mean age, 29.3 years) with no reported previous exposure to styrene. Both the workers and the comparison group were requested to complete questionnaires dealing with subjective symptoms felt during the workday. In contrast to the comparison group, the styrene-exposed workers felt tired more often in the morning and excessively tired after work, and they also reported more difficulties in concentrating and more frequent loss of appetite. During the workday, the RP/C workers frequently experienced irritation of the eyes, nose, and skin, and many felt nauseated and intoxicated. No correlation was found between urinary mandelic acid concentrations and the magnitudes of the symptoms scored on a scale of 1-3. Harkonen [126] suggested that no correlation was found because acute symptoms may be associated with peak exposures, whereas mandelic acid concentrations reflect the average exposure of the day.

In 1978, Harkonen et al. [125] summarized these studies [124,126,127] and extended some of the analyses. By plotting the log of the urinary mandelic acid concentration against the log of the 8-hour TWA styrene concentration, a significant correlation ($r=0.92$, $p<0.001$) was found. A urinary mandelic acid concentration of 700 mg/l corresponded to 31 ppm of styrene, 800 mg/l to 36 ppm, 1,200 mg/l to 55 ppm, and 1,600 mg/l to 74 ppm. Urinary mandelic acid concentrations were related to the percentage of EEG abnormalities in the group. Repeated slow wave activity and bilateral spike and wave discharges were used to indicate EEG abnormalities. In 38 workers whose urinary mandelic acid concentration was less than 700 mg/l, the percentage with abnormal EEGs was comparable with the general population (about 10%). Of 58 workers with mandelic acid concentrations greater than 700 mg/l, about a third exhibited EEG abnormalities. There was a significant degree of visuomotor inaccuracy with the symmetry of drawing test in workers whose mean urinary mandelic acid concentration was 800 mg/l. However, using the Bourdon-Wiersman test, visuomotor inaccuracy was statistically significant ($p<0.05$) only when the mean mandelic acid concentration was greater than 2,000 mg/l. A statistically significant decline in psychomotor performance (Mira Test) was found at mandelic acid concentrations greater than 1,200 mg/l (equivalent to about 55 ppm styrene). Harkonen et al. [125] concluded that although visuomotor accuracy and unimpaired psychomotor performance may be important in certain demanding operations and a prerequisite for safety at work, impairment may not necessarily affect a worker's performance under normal conditions, but may have an indirect impact by demanding more adaptation and more energy for compensation.

Lindstrom et al. [128] interviewed these same styrene-exposed workers about their alcohol consumption. A quantity of alcohol sufficient to produce a slightly intoxicated state was consumed daily by 1 worker, twice weekly by 22, once or twice a month by 56, and less frequently by the other 19 workers. Frequency of alcohol consumption for the group was about the same both before and after styrene exposure began. Decreased tolerance to alcohol was reported by 32% of the workers, about the same as in a comparison group of painters exposed to other solvents, but greater than in a group of railroad workers having no reported solvent or styrene exposure.

The amount of, and changes in, alcohol consumption and decreased tolerance to alcohol were not statistically related to the duration of exposure to styrene or to the concentration of urinary mandelic acid [128]. Overtime work, another index of styrene exposure, was related to alcohol consumption. The workers with high alcohol consumption were characterized as having straying thoughts and difficulties in staying asleep. Hand tremors and tiredness were related ($p<0.05$) to both the amount and frequency of alcohol consumption. Of all psychological functions, only lowered visuomotor speed was related to the amount of alcohol consumed [128]. Visuomotor inaccuracy was not the function that was previously related to high styrene exposure by Lindstrom et al. [127]; thus, Lindstrom et al. [128] concluded that the psychologic symptoms and signs related to alcohol behavioral variables were not related to styrene exposure.

In 1977, Meretoja et al. [129] reported an increase in the rate of chromosomal aberrations of the lymphocytes in peripheral blood. The ten men studied were 20-41 years old and from a Finnish factory where polyester plastic laminates were made. These workers had been exposed to styrene for 0.6-8.5 years. Before testing began, a complete health history was taken from each worker, followed by clinical and psychological examinations. No previous or present evidence of diabetes, epilepsy, or periods of unconsciousness lasting more than 30 minutes was found in any worker. Five healthy men with no known exposure to styrene or to any agent with known clastogenic activity were used as a comparison group.

No industrial hygiene sampling was conducted, but urinary mandelic acid concentrations were determined from specimens obtained at the end of an 8-hour shift to evaluate styrene exposure. A record was made of past exposures to any known clastogen, and a CBC was also performed [129]. The chromosomes of lymphocytes from peripheral blood of the styrene-exposed workers and the comparison group were studied. At the time of study all the workers were reported to be in good health. Results are presented in Table IV-13.

TABLE IV-13

ANEUPLOIDY AND CHROMOSOMAL ABERRATIONS
IN THE LYMPHOCYTES OF STYRENE-EXPOSED WORKERS

Subject No.	Age Yrs.	Years of Styrene Exposure	Mandelic Acid *	Interphase Cells: Micronuclei **	Interphase Cells: Nuclear Bridges **	Aneuploidy ***	Polyploidy ***	Chromosomal Aberrations ***
				Plant 1				
1	24	0.7	833	8	5	2	-	11
2	20	0.6	229	7	2	4	-	11
				Plant 2				
3	21	1.5	3,257	14	8	2	-	26
4	37	8	23	9	3	3	-	25
5	41	2.5	219	12	0	5	-	13
6	27	2	1,452	6	3	7	-	15
7	21	1	422	6	4	6	1	16
				Plant 3				
8	32	8.5	75	7	0	3	1	17
9	23	3	645	12	6	5	2	17
10	26	4	55	7	11	4	-	15
				Comparison Group				
11	32	0	0	2	0	1	-	2
12	35	0	0	0	0	3	-	4
13	33	0	0	0	2	3	-	1
14	30	0	0	2	2	1	2	1
15	30	0	0	0	0	2	-	1

*mg/g creatinine in urine
**Aberrations/1,000 interphase cells
***No./100 metaphase cells
Taken from Meretoja et al. [129]

There was a statistically greater incidence ($p<0.001$) of cells with chromosomal aberrations in lymphocytes of workers who had been exposed to styrene than in the comparison group (16.7 vs. 1.8 cells, on the average), but the biological significance of this difference is not known. An

increase in the frequency of micronuclei and nuclear bridges between cells was also observed. The incidence of aberrant cells ranged from 11-26% in the lymphocytes of the styrene-exposed workers and was only 1-4% in the lymphocytes of the unexposed comparison group [129].

In a related study in 1978, Meretoja et al. [130] examined the lymphocytes from the peripheral blood of 16 workers (which included 8 workers from the previous study [131]) exposed to styrene in two reinforced plastics factories. No data was given regarding occupational exposures. The subjects, all men 21-51 years of age, had been employed mainly in laminating work for 1-15 years. A statistically significant increase ($p<0.001$) in the incidence of chromosomal aberrations, mainly breaks, was found when compared to the comparison group (15.1 vs. 2.0%), and this was confirmed when 10 of these 16 workers were reexamined a year later and the incidence was 16.2%. The comparison group consisted of six men from outside the factory environment. However, the frequency of sister chromatid exchanges (SCE) was not significantly increased in these styrene workers (5.3 vs. 4.4 SCE/cell in the comparison group). SCE reflect intrachromosome rearrangements of the DNA helices, and as such are a sensitive indicator of damage to the DNA.

In 1979, Hogstedt et al. [131] also found an increased frequency of chromosomal aberrations among reinforced plastics workers in Sweden. Six male workers from a plant manufacturing polyester resin boats, aged 21-56 years with a mean age of 33, were matched by age and sex with 6 workers from a nearby paper factory without exposure to chemicals. In the plastics workers, there was an average of 10.8 aberrations per 100 cells in chromosomes from venous lymphocytes, a statistically significant increase ($p<0.001$) over that in the reference group, 5.2/100 cells. There was also an excess in gaps (3.2 vs. 2.4/100 cells, not significant), isochromatid and chromatid breaks (6.9 vs. 2.5/100 cells, $p=0.008$), and hyperdiploidy (0.7 vs. 0.3/100 cells, not significant). Airborne TWA styrene concentrations measured in previous years ranged from 14-73 ppm, with occasional concentrations as high as 188 ppm, for short periods of time. The investigators [131] speculated that, although styrene was the probable cause of the changes, exposure to other chemicals might have contributed; information was not given on these "other" chemicals.

In 1980, Andersson et al. [132] studied 39 men occupationally exposed to styrene in a plastic boat factory. The total exposures of these workers over a 6-year period were measured with personal TWA samples analyzed by gas chromatography and expressed as a concentration multiplied by the number of years of employment. A low-dose group (average 32 ppm-yr) and a high-dose group (average 283 ppm-yr) were identified. TWA styrene exposures for the various spraying, rolling, and casting operations ranged from 39-71 ppm (on the average), with exposures during assembly being about 8 ppm. TWA styrene exposure during spraying were sometimes as high as 158 ppm. Blood samples were taken from 36 of the styrene-exposed workers and from 37 age-matched workers (i.e., controls) in the same factory who were not exposed to styrene. Lymphocytes in peripheral blood were cultured and examined for

chromosome aberrations and SCE. The styrene-exposed workers had a significantly higher ($p<0.001$) number of chromosomal aberrations compared with the controls (7.9 vs. 3.2 aberrations/100 cells). There was no significant difference between the average numbers of chromosomal aberrations of the highly exposed and the less exposed styrene workers. There was a significant increase ($p<0.05$) in the average frequency of SCE in cells originating from 20 styrene-exposed workers in comparison with 21 controls (8.4 vs. 7.5 SCE/cell). Again there was no difference between the highly and less exposed groups. Interviews were performed when the blood samples were taken. Multiple regression analysis showed that among 9 factors introduced into the analysis (frequency of chromosomal aberrations, age, duration of employment, exposure to styrene, smoking habits, alcohol consumption, exposure to diagnostic X-rays, other solvents, use of breathing mask), only exposure to styrene showed a high positive correlation to the frequency of chromosomal aberrations [132].

In 1978, Fleig and Thiess [76] and Thiess and Friedheim [75] studied 14 workers employed for 2-24 years in three plants processing unsaturated polyester resins. At the time of the study, styrene concentrations as measured by colorimetric indicator tubes ranged from less than 50 to 300 ppm [75]. Mandelic acid concentrations in urine samples from the workers ranged from 100 to 1,500 mg/l [76]. Five of the 14 workers had increased GGTP (>28 units/ml) [75]. The investigators [76] found a significant excess (level of significance was not given) of chromosomal aberrations in the lymphocytes of peripheral blood from the styrene workers as compared to a control group of 20 workers from the same plant not exposed to styrene (9.2 vs. 5.5%). Fleig and Theiss [76] suggested that the chromosomal changes in these workers were probably due to styrene oxide or other exposures such as methylene chloride, rather than to styrene. Mention was made of a finding that styrene oxide was generated due to the use of peroxides, but data was not given on styrene oxide exposures.

However, there have been studies showing no significant increases in chromosome aberrations among styrene-exposed workers. An investigation of 24 styrene-exposed workers (20 men, 4 women) assigned to laboratory and technical service operations in a German polyester processing plant was reported in 1979 by Theiss and Friedheim [133] and in 1980 by Theiss et al. [134]. Their age range was 23-59 years, and they had been working with styrene for 4-27 years. The average styrene exposure was 6 ppm (range 1-12 ppm) in the laboratory and 58 ppm (range 1-178 ppm) in the technical service operation. Samples were collected with a "Personal Air Sampler" and analyzed with a gas chromatograph. The workers were given thorough clinical examinations that included personal history, chest X-rays, and laboratory studies including body plethysmography, EEGs, ECGs, blood counts, and blood chemistry determinations [133]. Findings were not relatable to styrene exposure, and there were no patterns of disease found. Except for one worker known to imbibe alcohol excessively, the neurological status of all workers was normal. One of the women, a diabetic treated with insulin, had given birth to a stillborn child. Another woman had aborted in the second month of pregnancy. Chromosome analyses of lymphocytes in peripheral blood

were carried out for all 24 workers as well as 24 controls from the same plant. The control group was comprised of medical department and office staff, and plant maintenance workers, none of whom were exposed to radiation or suspected chemicals at the time of testing. Urinary mandelic acid excretion did not exceed 350 mg/g of creatinine in any case; 19 of 22 workers tested had values below 80 mg/g. The investigators [133,134] considered 350 mg mandelic acid/g of creatinine to be a tolerable limit from the standpoint of occupational medicine. The mean frequency of chromosomal aberrations (i.e., chromatid and isochromatid gaps, breaks, fragments, chromatid interchanges, and dicentric chromosomes) was 5.1% in the styrene workers vs. 3.8% in the controls, a non-significant difference as determined by Theiss et al. [134]. However, Norppa et al. [135], in a 1981 discussion of this study [134], considered the results to show an increased frequency of cells with aberrations, when gaps were included.

In 1981, Watanabe et al. [136] examined peripheral blood lymphocytes of 16 workers occupationally exposed to styrene in two facilities (designated as I and II) where TWA styrene exposures were about 70 ppm and 35 ppm, respectively, as determined by carbon felt dosimeters and gas chromatography. Mandelic acid concentrations in the urine collected at the end of the work shift ranged from 90-4,300 mg/l (mean 647 mg/l) at Facility I and 300-1,360 mg/l (mean 526 mg/l) at Facility II. As compared to age- and sex-matched controls, the styrene-exposed workers showed no statistically significant increase in chromosomal aberrations (3.3% in Facility I and 3.6% in Facility II vs. 2.9% in the controls) or SCE frequencies (7.8% in Facility I and 6.7% in Facility II vs. 7.6%).

In 1982, Pero et al. [137] examined lymphocytes from Swedish workers exposed to styrene for genotoxic effects using unscheduled DNA synthesis (UDS) as the indicator of DNA damage. Heparinized blood specimens were taken by venous puncture from 38 male workers in a fibrous glass-reinforced polyester plastics factory where 8-hour TWA styrene exposures, as determined by gas chromatography, were 1-40 ppm. The workers (average age, 38.7 years) had been exposed to styrene for 1-23 years (average, 8.1 years). Twenty workers (average age, 36.2 years) were selected as a control group from a mechanical industry in the same town. Age distributions and smoking habits were similar in both the styrene-exposed and control groups.

Pero et al. [137] examined the possibility that lymphocytes isolated from styrene-exposed workers might have an altered level of unscheduled DNA synthesis (UDS) when the UDS was induced *in vitro* by either N-acetoxy-2-acetylaminofluorene (NA-AAF) or ultraviolet radiation (UV). The mean level of NA-AAF-induced UDS was significantly increased ($p<0.001$) for the styrene-exposed workers when compared to the mean level for the unexposed controls. There was no significant effect on UV-induced UDS from the *in vivo* styrene exposure. These results, in addition to lymphocyte cultures exposed *in vitro* to styrene, indicated to the investigators [137] that styrene exposure did not alter the efficiency of DNA repair synthesis, but rather predisposed lymphocytes to an increased risk for DNA damage from subsequent exposures to genotoxic agents that are dependent on cellular metabolism.

During 1977, Brooks et al. [91] studied 152 styrene-exposed workers (82 women and 70 men) and compared them with 34 female workers with no current styrene exposure. The styrene-exposed workers made reinforced plastic boats. Some of the workers sprayed the styrene-containing resin; others performed hand lay-up or other functions. Workers in the comparison group produced electronic circuit boards and, although they were exposed to many chemicals, their TWA exposures were always much lower than the respective OSHA limits. Table IV-14 contains information that describes the workers from both the study and comparison groups.

TABLE IV-14

DESCRIPTION OF WORKERS FROM STUDY AND COMPARISON GROUPS

Sex	Study Group Female	Study Group Male	Comparison Group All Female
Number	82	70	34
Mean age, years	41	37	38
Age range, years	19-72	18-76	19-56
Smokers	38	36	17
Exsmokers	7	18	1
Nonsmokers	36*	16	16
Average time on present job, months	77	85	62

*One woman did not respond
Taken from Brooks et al. [91]

Information about the workers was obtained from medical histories and physical examinations, psychomotor tests, CBCs, and pulmonary function tests. Additional information included data about the environmental concentrations of styrene and other contaminants from three industrial hygiene studies, and concentrations of mandelic and phenylglyoxylic acids in urine and of styrene in the blood and breath. Brooks et al. [91] combined the environmental data from all three surveys to characterize the styrene exposure for each job category; these data are presented in Table IV-15.

TABLE IV-15

STYRENE CONCENTRATIONS IN THE AIR OF THE STUDY FACTORY

Job Category	Number of Samples	8-hour TWA Styrene Exposure, ppm Mean ± SD
Prefabrication	12	2.8 ± 1.4
Gel coat spraying	6	68.5 ± 59.7
Hand lay-up	63	83.4 ± 42.7
Hand lay-up, other areas	18	25.7 ± 26.2
Woodwork/upholstery	11	3.4 ± 2.1
Final assembly	18	3.2 ± 1.8
Custom molding	18	6.5 ± 3.9
Small boat assembly	4	4.1 ± 1.0
Miscellaneous	2	3.9 ± 3.1

SD = standard deviation
Taken from Brooks et al. [91]

Data in Table IV-15 were obtained from the gas chromatographic analysis of 152 charcoal tube personal samples. In addition to these TWAs, the investigators recorded peak styrene concentrations in the range of 200-800 ppm. These peaks occurred during spraying operations.

To describe styrene absorption and excretion more completely, Brooks et al. [91] determined the concentration of styrene in the workers' blood and expired air. A statistically significant relationship ($r=0.79$, $p<0.001$) between the post-shift concentration of styrene in exhaled air and the styrene concentration of inspired air over the workshift was found. The data indicated that an 8-hour TWA exposure at 50 ppm styrene would result in a post-shift expired breath concentration of about 5.7 µg/liter. There was also an excellent correlation ($r=0.74$, $p<0.001$) between the concentration of styrene in inspired air during a workshift and that in venous blood at the end of a shift.

Brooks et al. [91] found that the styrene concentration in the expired air increased rapidly during the first 2 hours of exposure, remained relatively constant during the shift, then decreased slowly over the next 16 hours. The concentration of styrene in blood, however, rose continuously throughout the exposure period. Styrene concentrations in the blood declined to pre-shift values during the 16 hours following exposure [91].

Because of their hand lay-up work, the 152 workers studied had significant skin contact with the resin mixture [91]. Based on complaints, 41% of the styrene workers had a rash during the past year, and 19% had a rash at the time of the study, both significantly different ($p<0.05$) than the control group. These rashes were on the forearms in 29% of the workers; on the back of the hands in 15%; on the trunk in 14%; and on the upper arms in 12%. In the comparison group, only four workers (12%) reported a rash during the past year and one (3%) at the time of the study. A dermatologist diagnosed the styrene workers' rashes as follows: 18 workers with fibrous glass dermatitis, 6 with contact dermatitis, 7 with nonspecific dermatitis, 10 with acne vulgaris, and 3 workers with other inflammations.

Medical histories revealed a lower incidence of colitis, kidney or bladder infections, and anemia among workers of the study group than in the comparison group. There were no significant differences in the incidence of eye, nose, and throat irritation between the study and the comparison groups, nor were there any significant differences in the incidence of symptoms indicative of cardiovascular or neurological damage, nor were the reproductive histories of the women in the study and comparison group different.

Brooks et al. [91] did find statistically significant differences in the lung function of workers in the study and control groups. Seven workers (4.6%) in the study plant had FVC less than 80%, 7 (4.6%) had FEV_1/FVC X 100 below 70, 13 (8.6%) had FEV_1 less than 80%, and 24 (15.9%) had forced expiratory flow between 25 and 75% of vital capacity (FEF (25-75)), less than 70% of predicted values. No workers in the control plant had abnormal pulmonary function as indicated by FEV_1/FVC, FVC, or FEV_1; one of the subjects (2.9%) in the control group had a FEF (25-75) below 70%. Brooks et al. [91] concluded that since some subjects with abnormal pulmonary function were nonsmokers, the statistical difference between the study and control groups could suggest an occupational origin for the abnormality.

Possible acute effects of styrene exposure were studied by examining pre- and post-shift psychomotor performances of the exposed and comparison groups. The results of the tests for choice reaction time, Flanagan Coordination, Neisser letter search, and digit span were significantly worse for the styrene-exposed group as compared to the control group. However, as noted by the authors [91], a bias may have been introduced since a pre-requisite of employment at the control plant was the passing of a dexterity test. Performance on each test administered showed either no change or slight-to-moderate improvement post-shift in both the styrene-exposed group and the control group when compared to their pre-shift performances. When the styrene-exposed workers were divided into two

groups, one exposed at a mean TWA concentration of about 9 ppm (range, 4-120 ppm) and the other at about 82 ppm (range, 9-244 ppm), no significant difference between the groups was found in terms of effects of styrene on pre- or post-workshift performance of psychomotor tests.

Additional analysis (correcting for workers' ages) by Brooks et al. [91] of the psychomotor data, revealed a significant correlation ($p<0.025$) between decreased performance on the Neisser letter search test and duration of employment for the group with an average TWA styrene exposure of about 82 ppm. For the same group, impaired performance of both the Digit Span test and the Flanagan Coordination test was associated with duration of employment, although it was not statistically significant. Based on these results, Brooks et al. [91] concluded that exposure to styrene at an average concentration of about 82 ppm impaired workers' performance on the Neisser letter search test and, possibly, tests of coordination and memory.

Brooks et al. [91] also determined the concentrations of mandelic and phenylglyoxylic acids in the urine of the styrene-exposed workers. When the log of the sum of the mandelic acid and phenylglyoxylic acid concentrations in post-shift urine was plotted against the log of the TWA styrene concentration for that shift, the correlation coefficient r was 0.925 ($p<0.00001$); when only the concentration of mandelic acid was used, the correlation coefficient was 0.93 ($p<0.00001$). There were also excellent correlations between the log of the post-shift urinary mandelic acid concentration and the log of the post-shift venous blood styrene concentration ($r=0.899$, $p<0.001$) and the log of the styrene concentration in post-shift expired breath ($r=0.877$, $p<0.001$).

In 1979, Kjellberg et al. [392] studied 7 boat-fabrication shop workers and compared them with 7 workers in a mechanical industry in the same Swedish town. The boat workers were exposed to styrene while making the boats and to acetone during the cleaning of equipment. TWA exposures were found to be 3-14 ppm of styrene and 8-60 ppm of acetone. Of the behavioral tests applied, namely, reaction time, Bourdon-Wiersman, and reaction time additions, only the reaction time test showed a significant difference ($p<0.05$) between the RP/C workers and the comparison group. This reaction time test involved measurement of the time between the illumination of a lamp and the press of a button by the subject. Kjellberg et al. [392] concluded that work exposure had caused the decreased reaction times, but were unable to determine whether styrene, acetone, or their combined effect was responsible.

In a 1980 study of 27 British men engaged in boat building, Cherry et al. [138] found effects attributable to CNS depression. The styrene-exposed workers were compared to a control group of workers from the same plant, with no styrene exposures, of almost the same average age (23 years in exposed workers vs. 26 years in referents). Personal TWA exposures averaged 117 ppm styrene in the morning and 52 ppm in the afternoon, for an overall average of 92 ppm. Blood styrene averaged 6.9 µmol/liter in the exposed and 0.6 µmol/liter in the control workers; urinary mandelic acid

averaged 581 μmol/mmol creatinine in the styrene-exposed workers, with no value being given for controls. Changes in mood were reported in both groups, but more so in those workers exposed to styrene; changes in mood were correlated with blood styrene concentrations. The reaction times of styrene-exposed workers were slower than the referents in the morning, but their reaction time increased during the day so that in the afternoon they were similar to those of referents. The styrene-exposed workers also fared worse in other behavioral tests than did the controls, but the differences were minor. The styrene-exposed workers reported being more fatigued than did the men in the comparison group, and they also reported being more tired on Friday evening than on Monday evening.

In 1980, Hemminki et al. [139], described finding a greater number of spontaneous abortions among female chemical workers than would be expected from the rate among all Finnish women, with part of this excess occurring among styrene workers. The investigators [139] obtained information on 9,000 female workers during the period 1973-1976 from union and national Registry files, and found that 52 of the women had reported spontaneous abortions. The national Registry was based on general hospital inpatients, not including aborting women not treated, women treated on an outpatient basis, or women treated in private hospitals, which provided only about 2.2% of the obstetrics-gynecology beds in Finland. Abortion rates were expressed as the ratio of spontaneous abortions to pregnancies (SA/P) and as the ratio of spontaneous abortions to births (SA/B). Hemminki et al. [139] used both ratios because they believed the former index underestimated the risk, inasmuch as some induced abortions included in total pregnancies would have miscarried, and the latter index overestimated the risk because induced abortions were not included; there is a high rate of induced abortions in Finland. The results of the study are shown in Table IV-16.

TABLE IV-16

SPONTANEOUS ABORTIONS OF
FINNISH CHEMICAL WORKERS IN 1973-1976

Branch of Employment	Number of Spontaneous Abortions (SA)	SA x 100 / Pregnancies (SA/P)	SA x 100 / Births (SA/B)
All women in Finland	15,482	5.52	7.98
Union of Chemical Workers	52	8.54**	15.57***
Plastics Industry	21	8.94*	17.80***
Styrene Production and Use	6	15.00**	31.59***
Viscose Rayon Industry	9	11.25*	22.50***
Laundries	7	10.14	16.67*
Pharmaceutical Industry	5	10.20	22.72*

*significant difference from "All women in Finland," $p<0.05$
**significant difference from "All women in Finland," $p<0.01$
***significant difference from "All women in Finland," $p<0.001$
Taken from Hemminki et al. [139]

There were statistically significant excesses in spontaneous abortions among members of the Union of Chemical Workers and among several subgroups. There was a significant excess among plastics workers, especially among styrene and viscose rayon workers. There were also excesses, significant only in terms of the SA/B index, among workers in laundries and in the pharmaceutical industry. There were 6 spontaneous abortions among styrene workers, resulting in an SA/P index of 15.00 ($p<0.01$) and an SA/B index of 31.59 ($p<0.001$), the highest indices of any group tabulated in the report [139], compared to 5.52 and 7.98, respectively, in the overall Finnish population. Hemminki et al. [139] stated that the styrene workplaces included mainly reinforced plastics workshops.

In 1982, Harkonen and Holmberg [140] studied 67 female lamination workers occupationally exposed to styrene to evaluate the possible embryotoxic effects of styrene. The average age of the workers at the 6 Finnish factories manufacturing reinforced plastics was 30 and ranged from 19-40 years. The duration of past styrene exposure was 0.5 to 10 years, and averaged 4.5 years. No styrene measurements were made in this study, but the investigators [140] stated that in a previous study in the Finnish polyester plastics industry, the median TWA exposure to styrene in lamination work was 66 ppm. Each of the laminators was matched by age with textile or food production workers of a similar social class with no occupational solvent exposures. A questionnaire was personnally administered by the same interviewer during 1979-1980. The obstetric

histories of the subjects (laminators) were divided according to the time prior to styrene exposure and the period of styrene exposure. The obstetric histories of the controls were likewise divided to correspond to the time periods of their age-matched exposed subjects. Table IV-17 presents data on pregnancies, births, spontaneous abortions, and induced abortions.

TABLE IV-17

NUMBER OF PREGNANCIES, BIRTHS, SPONTANEOUS ABORTIONS, AND INDUCED ABORTIONS AMONG 67 LAMINATION WORKERS AND MATCHED CONTROLS

	Before Styrene Exposure		During Styrene Exposure	
	Laminators	Controls	Laminators	Controls
Pregnancies	48 (84)	48 (80)	12 (16)	20 (22)
Births	40 (69)	39 (67)	3 (4)*	14 (14)
Spontaneous abortions	8 (8)	5 (8)	3 (4)	4 (4)
Induced abortions	6 (7)	5 (5)	8 (8)	4 (4)

Note: The number of occurrences is given in parentheses.
*different from controls, $p<0.01$
Taken from Harkonen and Holmberg [140].

Prior to the period of styrene exposure, the number of women with pregnancies, births, spontaneous abortions, and induced abortions (or the number of occurrences of each) did not differ significantly for the styrene-exposed and control groups. During styrene exposure, the number of pregnancies was not significantly different but the number of births among the exposed subjects was significantly less than the controls (4 vs. 14, $p<0.01$). One cause leading to this difference was the higher number of induced abortions (8 vs. 4); this difference, however, was not significant. There were two birth defects reported by both the laminators and the controls. The two groups did not differ in the use of contraceptives or drugs, but smoking and the consumption of alcohol during pregnancy were more common in the styrene-exposed group [140].

To summarize this subsection, these studies of workers in factories where styrene copolymers (mainly RP/C) were produced have demonstrated effects that have been attributed to styrene exposure despite potential exposure of the workers to other chemicals. It is likely that some of the reported effects on worker health such as eye and respiratory tract irritation, dizziness, headache, nausea, and feelings of intoxication were due to styrene exposures at peak concentrations. Some other effects such as

abnormal EEG patterns, increased incidence of chromosomal aberrations and sister chromatid exchanges, increased incidence of spontaneous abortions, visuomotor inaccuracy, and impaired psychomotor performance have also been reported.

Epidemiological Studies

A retrospective cohort mortality study of the German styrene and polystyrene production plant previously described in the Clinical Studies Section [75,76,77], was reported in 1978 by Frentzel-Beyme et al. [78]. Airborne styrene concentrations in production areas were about 1 ppm in 1975 [75]. In the mortality study, records of 1,960 past and present workers were reviewed; causes of death as listed on death certificates of 74 workers that died during 1956-1976 were compared with age-specific mortality of the population of the whole country. The proportion of deaths due to liver and digestive organ disease was higher in the styrene-exposed workers than in the overall population of the country. However, the investigators [78] did not identify their criteria of diagnosis of liver and digestive organ diseases. Frentzel-Beyme et al. [78] suggested that the higher incidence of liver disease might be because these workers were from the primary wine-producing region of the country, implying a greater than average alcohol consumption. However, no data on the alcohol consumption of either the styrene-exposed or unexposed groups were given. A total of 12 deaths from cancer were recorded among the styrene workers (3 lung, 2 stomach, 2 pancreas, 2 colon, 1 rectum, 1 spleen, and 1 probable kidney cancer). The incidence of these tumors did not differ from that of the overall population of the country. The proportion of deaths due to cardiovascular diseases was also less than in the country as a whole.

In 1974, Maier et al. [84] reported the results of a proportional mortality study of a U.S. styrene and polystyrene plant previously described in the Clinical Studies Section [58,81,82,83,84,85,86,87]. Production of styrene and butadiene monomers began at this plant in 1943, with butadiene produced until about 1950; since 1950, the principal product had been polystyrene [82]. Prior to 1962, benzene was used as a raw material in the on-site production of ethylbenzene; subsequently, ethylbenzene for styrene monomer production had been shipped into the plant [141]. In a 1973 industrial hygiene survey, Maier et al. [84] determined that exposures to styrene, benzene, ethylbenzene, or toluene were each usually less than 10 ppm. Death certificates for 46 workers from this plant who had died during the previous 5 years were analyzed by Maier et al. [84] by a comparison to the mortality of males, aged 60-64 years, in the nation as a whole. Of the 46 deaths, 1 was a suicide and 4 were accidents. Grouping the remaining 41 death certificates into either "Coronary Disease," "All Forms of Cancer," "Cerebral Vascular Disease," "Respiratory Disease," "Diseases of the Digestive System," or "All Other" revealed no significant differences from the expected proportion of deaths in any classification. Due to the small number of deaths involved, as well as the use of mortality rates from males aged 60-64 years for comparison, this study is difficult to interpret.

However, these findings were verified by a more extensive mortality study reported in 1978 by Nicholson et al. [83]. The vital status in 1975 of 560 men who were working in this factory on May 1, 1960 and who had been employed at least 5 years was determined. The investigators [83] expected about 106 deaths but 83 were found. Deaths from "Heart and Circulatory Diseases," "Respiratory Diseases," "Cancer," and "Other Causes" categories were not greater than expected. Because of potential benzene exposures, special attention was given to leukemia as a cause of death. Two cases of leukemia and one lymphoma were recorded on the 83 death certificates. Death certificates provided by the company of 361 other workers who had been employed at this factory for 6 months or more were examined, and five additional cases of leukemia and four additional cases involving the lymph system were recorded on these. Although the information available from the 361 randomly collected death certificates was "suggestive" of an excess risk of death from leukemia or lymphoma in this factory, Nicholson et al. [83] concluded that the data was not definitive.

In 1980, Ott et al. [31] studied the mortality rate of workers at four different locations of a U.S. manufacturer of styrene and styrene products. Work activities were mainly in styrene production, polystyrene production, copolymerization of styrene with butadiene or acrylonitrile, color mixing, resin extrusion, and in support activities such as research, pilot plant development, and product development. Based on surveys conducted between 1962 and 1975, estimated TWA styrene exposures were all below 10 ppm, including one survey in which there were excursions to 50 ppm. In some units, there were exposures to benzene which were estimated to have been below 15 ppm during the period 1953-1972, but much higher earlier. There were other exposures recorded, such as ethylbenzene (less than 10 ppm), ammonia (about 15 ppm), acrylonitrile (less than 10 ppm), vinylidene chloride (less than 1 ppm), formaldehyde (about 3 ppm), cleaning solvents, and various pigments and dyes.

The earliest operation was 40 years old, but how many, if any, workers had worked with styrene that long was not stated; however, 2,904 workers had been employed for at least 1 year in styrene operations. Of the total cohort, 2,360 workers (81%) were from the company's Michigan location, with the remaining workers from plants in Texas, Connecticut, and California. Verification of vital status was completed for all but 88 former workers [31].

During the period 1940-1975, there were 303 deaths found among styrene workers (professional and nonprofessional research workers, supervisors, and production workers), compared with 425 expected in the U.S. white male population; this total of 303 did not include 17 deaths among 164 former workers exposed to arsenic, asbestos, or high levels of vinyl chloride. There were 58 deaths from the "All Malignant Neoplasms" category compared with 76.5 expected. Except as discussed below, the number of deaths from specific causes did not exceed the expected number. There was a slight excess of deaths from the category "Bronchitis, Emphysema, and Asthma" (10 observed vs. 7.5 expected), "Malignant Neoplasms of the Lymphatic and

Hematopoietic Tissue except Leukemia" (7 observed vs. 5.3 expected), and "Leukemia" (6 observed vs. 3.4 expected). In addition to the 13 cases in which leukemia or lymphoma (malignant neoplasms of the blood forming organs) were reported as the cause of death, there were 2 cases where the presence of leukemia or lymphoma was mentioned on the death certificate but where another cause of death was listed, and 6 cases identified as still living. The incidence of lymphocytic leukemia cases were significantly greater ($p<0.05$) than expected on the basis of age-specific incidence rates from the Third National Cancer Survey (7 observed vs. 1.6 expected). Of these cases, 5 cases (1 of whom was still living) vs. 0.26 cases expected ($p<0.05$) occurred in workers in operations involving colorant blending and roll compounding or extrusion of plastics, areas where polymer dusts, solvents, colorants, and extrusion fumes composed of vapors including styrene and ethylbenzene were present [31].

The production and nonprofessional research workers (2,310 men) were also studied separately. There were 282 deaths in this group vs. 357.8 expected from U.S. white male data and 287.6 expected from data published in 1954 on workers from the company's Michigan location. (There was an overlap between the two cohorts as 1,333 of the 8,171 workers from the 1954 comparison group were included in the later study group.) Of the 282 deaths, 55 were due to "Malignant Neoplasms," vs. 64.2 expected in the U.S. population and 65 in the 1954 company comparison group. There were 6 deaths from "Leukemia," a significant difference ($p<0.05$) from the 1.6 expected in the company comparison group but not from the 2.9 expected in the U.S. population [31].

In summary, mortality was less than that of the corresponding white male population. Deaths due to malignant neoplasms were fewer than expected, but an increase in lymphatic leukemia was observed among a subgroup of workers who had exposures to polymer extrusion fumes (including styrene), solvents, and colorants. However, the etiology of the lymphatic leukemia could not be established by Ott et al. [31].

In 1978, Ahlmark [142] described the cancer incidence and mortality among a group of workers in Sweden who were exposed to styrene in the manufacture of reinforced plastics. This was a retrospective cohort study in which entry into the cohort was defined by the date of first employment. From as many reinforced plastics companies as could be located and as were willing to participate, persons working with styrene up to the year 1970 were identified. Sufficient information to use in the study was found on 1,114 men and 91 women. Exposure information for these workers was not provided, but Ahlmark [142] estimated, from information on reinforced plastics operations in Sweden, that mean styrene exposures were 250-300 ppm in the late 1960s and early 1970s and were probably higher earlier. The 1,114 men ranged in age from the late teens to the 70's and the 91 women from the 20's to the 60's. Twelve deaths from cancer were found in the study population from the Social Welfare Board Cancer Register for 1959-1976 and the Central Statistical Bureau Mortality Register for 1972-1976, which were less than the 16.6 expected [142].

While no excess of cancer was found in this group of workers, exposed to styrene at concentrations estimated to have been 250-300 ppm and higher, Ahlmark [142] pointed out that latency periods for many of these workers were not great, so, if styrene were a carcinogen, more cases would be expected later. The problem of varying latency was handled in the calculations by computing person-years of employment in each age category in the study cohort. However, only 472 workers (39%) in the cohort had a latency period of 11 years or more, so, although not given, the number of workers with more more than 20 years since first exposure was probably small.

Uptake, Metabolism, and Elimination

Studies were reported by Bardodej et al. [143] in 1961 and Bardodej [144] in 1964 on the uptake, metabolism, and elimination of inhaled styrene by humans. These investigations were later summarized in 1966 and 1970 by Bardodej and Bardodejova [100,145]. Human volunteers, with no history of occupational styrene exposure, were experimentally exposed to styrene in a closed chamber with no provisions for changing chamber air. To study styrene absorption, Bardodej [144] measured each subject's minute volume during exposures to styrene at 21, 49, 106, and 188 ppm. The range of minute volumes was 6-40 liters. Styrene vapor concentrations inside the chamber and in the breath were monitored by ultraviolet spectrophotometry. The percentages of styrene retained by the lungs were calculated from styrene concentrations in the inhaled and the exhaled air. Over the range of styrene concentrations studied, the subjects retained about 60% of the inhaled styrene regardless of minute volumes or duration of exposure. When the subjects were removed from the test atmosphere, styrene concentrations in exhaled air decreased within seconds to less than half the levels found immediately after removal from exposure.

To study styrene metabolism, urine was collected from subjects exposed at 22, 129, and 235 ppm for 8 hours and analyzed for styrene and its metabolites. Styrene concentrations were below the detection limit of the photometric method used. Urine samples were subjected to paper chromatography, using several liquid phases and reagents for detection of mandelic, styrene thiolic, hippuric, and phenylglyoxylic acids, phenylglycol, phenylglycol glucuronide, and omega-hydroxyacetophenone [144]. The presence of phenylglycol, the glucuronide of phenylglycol, omega-hydroxyacetophenone, and styrene thiolic acid could not be demonstrated in the urine of exposed subjects, nor could an increase in hippuric acid concentration [143]. Styrene oxide could not be found in the urine samples by using a spectrophotometric method. Both mandelic and phenylglyoxylic acids were found and confirmed.

Urinary mandelic and phenylglyoxylic acids accounted for 85% and 10% of the absorbed styrene, respectively. Eighty percent of orally administered mandelic acid was excreted as such, with 15 percent excreted in the urine as phenylglyoxylic acid [144]. Concentrations of urinary mandelic acid of subjects at the end of 8-hour exposures to styrene at 22, 129, and 235 ppm

averaged about 500, 1,350, and 2,700 mg/l, respectively [143,144]. After an 8-hour exposure to 22 ppm, urinary mandelic acid, expressed as the ratio of mandelic acid to creatinine, was maximal at the end of exposure (i.e., 0.4) and declined linearly over the next 20 hours to about 0.06.

In 1965, Fiserova-Bergerova and Teisinger [146] reported the retention of styrene in subjects exposed at 24 ppm. Ultraviolet and polarographic methods (which they found equivalent) were used to analyze for styrene in inhaled and exhaled air during 2- and 5-hour exposures. In all experiments, the subjects exhaled through the mouth; they inhaled through the nose in some experiments and through the mouth in others. The average styrene retention from the very beginning of exposure was 66% following nasal inspiration and 59% after oral inspiration, and did not change during the 2 or 5 hours of exposure. In the analysis of a single deep breath, alveolar styrene concentrations were found to be about 6.2% of the inhaled concentration; after an expiration pause (30 seconds), alveolar concentrations were about 5.5%. This indicated that transfer of styrene from the blood to the lungs was minor. No styrene was found in exhaled air one minute after removal from exposure [146].

Studies of pulmonary absorption and the elimination of styrene inhaled at 70, 103, 115, 200, and 206 ppm for 4 or 8 hours were reported by Fernandez and Caperos [147] in 1977 and Caperos et al. [148] in 1979. Pulmonary ventilation, respiratory rate, and alveolar concentrations of styrene were monitored frequently during the exposures. Removal of styrene from inhaled air by blood was practically constant throughout all of the exposures. The amount of styrene absorbed ranged from 90.5% during exposure at 70 ppm for 4 hours to 86.8% during exposure at 206 ppm for 8 hours. The styrene analytical method (gas chromatography) under their sampling conditions had a lower limit of detection of 0.01 ppm in 25 ml of expired air. Some styrene was found in alveolar air for about a week after the exposures. The calculated total amount of styrene eliminated by the lungs accounted for 2.6% (1.5-4.4%) of that absorbed, independent of exposure conditions [147]. On the average 54% was excreted in the urine as mandelic acid and 38% as phenylglyoxylic acid. Half of the urinary mandelic acid and one quarter of the phenylglyoxylic acid formed was excreted during the 8-hour exposure to 103 or 206 ppm styrene with the rest being excreted within 3-5 days [148].

After administering styrene at 50 and 150 ppm through a mouthpiece, Astrand et al. [88] in 1974 measured its concentrations in the alveolar air and arterial and venous blood of human volunteers. These measurements, along with routine cardiac and respiratory tests, were conducted on 2-14 male subjects who were 21-28 years old. Measurements were made at rest and after exercise on a bicycle ergometer at 50, 100, and 150 Watts (W) to study the effects of various workloads on styrene uptake. Mixtures of air that contained 4% carbon dioxide and various amounts of styrene were used to observe the effects of increased respiration on styrene metabolism without the complicating effects of an increased cardiac rate. The styrene concentrations found in the alveolar air and in the arterial and venous blood under different conditions of exposure are presented in Table IV-18.

TABLE IV-18

AVERAGE STYRENE CONCENTRATIONS IN ALVEOLAR AIR AND BLOOD OF SUBJECTS EXPOSED TO STYRENE AT 50 AND 150 ppm

Exposure Conditions				Styrene Concentration (ppm)		
Styrene (ppm)	Workload (W)	CO2	N*	Alveolar Air	Arterial Blood	Venous Blood
50	0	No	6	8.8	0.5	0.3
50	50	No	6	9.5	1.9	1.4
150	0	No	14	23.0	1.8	1.0
150	0	Yes	2	26.3	5.3	2.4
150	50	No	11	29.9	6.5	4.7
150	50	Yes	2	28.8	12.3	7.7
150	100	No	4	32.5	11.6	7.9
150	150	No	4	35.4	15.9	12.4

*Number of subjects
Taken from Astrand et al. [88]

Pulmonary ventilation, heart rate, and oxygen uptake were unaffected by exposure to styrene. Alveolar styrene concentrations rose only slightly while the arterial concentrations almost tripled in response to a 50-Watt workload. The alveolar styrene concentration reached a plateau after only 1 minute of exposure, but the arterial styrene concentration increased during the entirety of each 30-minute exposure period. Further increases in the arterial styrene concentrations also occurred after exposure to styrene in air containing 4% carbon dioxide [88].

Because of the poor relationship between alveolar and arterial styrene concentrations, Astrand et al. [88] concluded that alveolar air was a poor indicator of the extent of exposure to styrene vapor. Astrand et al. [88] recommended that a fingertip blood sample be used to evaluate styrene uptake because styrene concentrations in arterial and capillary blood were in very good agreement. Urinary mandelic acid was determined colorimetrically before exposure and at 3-hour intervals for 24 hours after removal from exposure. The mandelic acid concentrations peaked 3-5 hours after exposure and fell to pre-exposure values 9-13 hours after concluding exposure. In one experiment, the concentrations of styrene in inspired and expired air were continuously measured in two individuals at rest during four consecutive 30-minute styrene exposures at 50, 150, 250, and 350 ppm. More than two-thirds of the styrene inhaled during each 30-minute period was retained by the subjects. The amount of styrene absorbed was 750 mg during this

sequential 2-hour exposure that averaged 200 ppm of styrene, and it was determined that about 50% of the amount absorbed was excreted as mandelic acid. Because a nonspecific method was used for determining mandelic acid, it is likely that not all of the mandelic acid excreted was measured. Astrand et al. [88] concluded that measurement of urinary mandelic acid would not provide an accurate indication of styrene exposure because there was a large variation between subjects at low exposures. However, with the nonspecific method used to determine mandelic acid, background values were large and variable; consequently, slight differences in mandelic acid concentrations might not have been detectable.

Hake et al. [70] in an inhalation study summarized earlier in the Experimental Exposures Section, concluded that blood analysis, metabolite to creatinine ratio in 24-hour urine samples, and alveolar breath analysis 15 minutes after exposure were all useful indicators of styrene exposure, but they preferred breath analysis provided analysis could be performed the same day as exposure occurred. Such a sample taken 15 minutes after exposure and containing more than 2.8 ppm styrene indicated to the investigators [70] a styrene burden injurious to health; this was associated with changes in EEGs, visual evoked response, and pulmonary function, and in the onset of such symptoms as headache, dizziness, and irritation of the eyes and upper respiratory tract. The experimental exposures resulting in styrene concentrations of 2.8 ppm in alveolar air (15 minutes after exposure) were 100 and 125 ppm. No subject exposed at 20 ppm had more than 0.8 ppm styrene in his alveolar breath 15 minutes after exposure.

Gotell et al. [35], in field studies of RP/C production workers summarized earlier in the Clinical Studies Section, collected breath samples 15 minutes, 2 hours, and 5 hours after the end of the workday and analyzed them for styrene by gas chromatography using a flame ionization detector. The workers were divided into three groups based on TWA exposures to styrene: (I) 235-292 ppm, (II) 89-139 ppm, and (III) 17-32 ppm. When the concentrations of styrene in expired air were plotted against the time after exposure, there was a characteristic rate of styrene excretion for each group based on the level of exposure. Five hours after exposure, the concentration of styrene in expired air was about 0.9 ppm for group I, 0.3 ppm for group II, and 0.2 ppm for group III [35].

Urine samples were collected immediately after work for measurement of mandelic and phenylglyoxylic acids by the methods of Ohtsuji and Ikeda [121] with the results adjusted to a urine specific gravity of 1.024. Control values in 27 nonexposed men were: mandelic acid, 78-434 mg/l; and phenylglyoxylic acid, 20-196 mg/l [35]. The overall linear correlation coefficients (Groups I, II, and III) with 8-hour TWA styrene exposure concentrations were 0.75 for mandelic acid and 0.36 for phenylglyoxylic acid. However, the respective correlation coefficients for workers exposed at 8-hour TWA concentrations ranging from 17-139 ppm (Groups I and II) were 0.96 and 0.85, while for the workers in Group III exposed at higher concentrations (i.e., 235-292 ppm), the correlation coefficients were -0.93 and -0.95. These negative correlation coefficients found at the higher

styrene concentrations may reflect an asymptotic curve rather than a real reduction in mandelic or phenylglyoxylic acid concentration. Gotell et al. [35] plotted three linear regression lines to explain the relationship between concentrations of metabolites and airborne styrene. Visual inspection of these plots suggests that the data might best be fitted by a hyperbola. If so, the negative regression would be better expressed as the upper end of such a curve. If these data are indeed properly described by a hyperbola, it appears that excretion of these metabolites approaches a maximum (asymptotically) with increasing dose of absorbed styrene. Whether the limitation is metabolic or excretory is not established, though metabolic limitation seems a more likely explanation.

In 1978, Engstrom et al. [149] found styrene in subcutaneous adipose tissue in volunteer subjects exposed to styrene vapor. Seven male subjects, 22 to 30 years of age, were exposed for two hours to 50 ppm of styrene through a breathing valve and mouthpiece with 30 minutes of rest and three 30-minute work periods on a bicycle ergometer at intensities of 50, 100, and 150 Watts. Average styrene uptake was 490 mg, corresponding to 63% of the amount inspired. Styrene uptake was 5-6 times higher during heavy work than at rest. The average total amount of styrene expired during the 19 hours following exposure was about 3% of the amount retained during exposure.

Needle biopsy of subcutaneous adipose tissue was performed on all of the subjects before exposure and 0.5, 2, 4, and 20-24 hours after the exposure. In addition, four of the men were subjected to biopsies during the 1-2 weeks following exposure. Styrene concentrations in the adipose tissue of the subjects were determined after evaporation of the solvent at 150°C into nitrogen which was continuously exchanged. The gas was collected in 30-ml glass syringes and assayed with gas chromatography. About 24 hours after exposure the average concentration of styrene in adipose tissue was about the same level as 2-4 hours after exposure, i.e., about 3.5 mg/kg. Retention of styrene was noticed as late as 13 days after the 2-hour exposure. The investigators [149] estimated the half-life of styrene in adipose tissue to be 2-4 days.

Also in 1978, Engstrom et al. [150] conducted a similar investigation of three males occupationally exposed to styrene during the processing of polyester tanks. The TWA exposures of the workers were 8, 15, and 20 ppm. The average daily uptake of styrene was estimated to be 193-558 mg. On Monday morning styrene concentrations in adipose tissue were 2.8-8.1 mg/kg and on Friday afternoon were 4.7-11.6 mg/kg. The concentrations were higher in the two workers with the higher exposures (i.e., 15 and 20 ppm) of longer duration (several years) as compared to styrene adipose tissue concentrations in the other worker who had been employed only 2 weeks with an exposure of 8 ppm . Both of the workers with several years of exposure to styrene had a considerable amount of body fat (27 and 41 kg, respectively) as estimated by an anthropometric method. The half-lives of styrene in adipose tissue for these two workers were calculated by Engstrom et al. [150] to be 5.2 and 2.8 days, respectively.

The percutaneous absorption of undiluted and aqueous solutions of styrene was studied in 1968-1969 by Dutkiewicz and Tyras [151,152,153]. With undiluted styrene, either 0.1 ml (88.8 mg) or 0.2 ml (176.8 mg) was applied to the forearms of each of seven subjects. A watch glass was pressed tightly to the skin, and the amount of styrene absorbed was calculated from the difference between the amount applied and the amount recovered after 8-15 minutes. Styrene that was removed from the skin with gauze and from the gauze and watch glass with glacial acetic acid was analyzed colorimetrically. On the average, styrene was in contact with 17.3 sq cm of skin for 11.1 minutes, and 39.4 mg of styrene was absorbed. The average calculated rate of absorption in the seven subjects was 11.9 mg/sq cm/h [151].

In the study of styrene diluted with water, the difference in the concentration of the solution before and after immersing one hand for an hour was directly determined with an ultraviolet spectrophotometer. Evaporation of the solution was prevented by enclosing the beaker and hand in a polyethylene bag. With styrene concentrations of 67.5-264.0 mg/l of water, 14 observations were made on 6 subjects. The rates of styrene absorption increased linearly with styrene concentration. Rates of 38-184 µg/sq cm/h were estimated [151].

Dutkiewicz and Tyras [151] also evaluated urinary mandelic acid and styrene in exhaled breath in the study of percutaneous absorption of styrene. Four subjects each immersed both hands in aqueous styrene solutions (215-220 mg styrene/liter of water) for 2 hours. During the ensuing 24 hours, mandelic acid was determined polarographically after conversion to benzaldehyde. The rate of excretion of mandelic acid in urine decreased exponentially from 3-4 mg/h initially to about 0.7 mg/h at 18-24 hours. On the average, the subjects excreted about 40 mg of mandelic acid in 24 hours or only about 13% of the estimated average amount of styrene absorbed. Traces of styrene were detected in expired air of subjects who had absorbed about 200 mg of styrene. The investigators [151] concluded that contact of both hands with undiluted styrene for 1.5 minutes or with saturated aqueous solutions for 1 hour could result in absorption of styrene equivalent to that absorbed by a worker exposed to airborne styrene at 12 ppm for 8 hours. Whether data from absorption of styrene through the hands can be quantitatively applied to other skin areas is not clear, but at least the data [151] clearly demonstrate that styrene can penetrate human skin.

In 1978, Riihimaki and Pfaffli [154] described the percutaneous absorption of airborne styrene in two male subjects. These subjects were exposed to styrene vapor at a concentration of 600 ppm for 3.5 hours. Each wore socks, pajamas made of a thin cloth, and, to prevent absorption through the lungs, a full-facepiece positive pressure respirator that was monitored for proper operation. They exercised at 100 Watts for 10 minutes of each hour on a bicycle ergometer, and at all other times remained sedentary; this exercise made them perspire for about 15 minutes. Based on amounts of urinary metabolites (mandelic, phenylglyoxylic, and methylhippuric acids) and styrene in expired air, the amount of styrene absorbed through the skin

during the exposure averaged 576.1 μmoles for the two subjects. This amount was estimated to be 19% of the amount that would have been absorbed through the lungs at the same concentrations, assuming a 60% retention rate for styrene at 600 ppm. This amount was twice that absorbed by the lungs during the exposures of three subjects at 10 ppm (576.1 vs. 288.1 μmoles). The small number of subjects and the variability in the data preclude definite conclusions from this experiment, other than to infer that styrene vapor can penetrate intact skin, albeit inefficiently. A question should also be raised about the degree of reliance to be placed on respirators by those working in high concentrations, such as some work in enclosed spaces.

In 1980, Brooks et al. [155] designed a study to determine whether styrene absorption through the skin results in measurable changes in biological indicators of styrene exposure. Eight female workers engaged in hand lay-up operations in fibrous glass boat production during which extensive styrene skin contact occurred were studied. Measurements of expired breath and blood styrene levels and urinary levels of mandelic and phenylglyoxylic acids were made during 4 consecutive days using different experimental conditions including either (1) gloves and protective clothing alone, (2) respirator alone, (3) gloves, protective clothing, and respirator, or (4) no respirator, gloves, or protective clothing. TWA airborne styrene levels ranged from 71-91 ppm, as determined with charcoal tubes and gas chromatographic analysis. Levels of styrene in venous blood and expired breath and excretion of urinary mandelic and phenylglyoxylic acids were no different when gloves and protective clothing were used as protection compared to when no gloves, protective clothing, or respirators were used. Significant reduction in all biological indices studied occurred when respiratory protection was used. Brooks et al. [155] concluded that percutaneous absorption of styrene was not a significant exposure source and did not significantly contribute to the body burden of styrene in the RP/C workers studied who were engaged in hand lay-up operations. The lack of apparent skin absorption was attributed to possible differences in transport phenomena attendant to the use of polyester resins in combination with styrene, which might have inhibited percutaneous absorption [155]. However, Dutkiewicz and Tyras [151] as previously discussed, determined that only 13% of the styrene absorbed percutaneously with hands immersed in an aqueous solution was eliminated as mandelic acid.

In 1978, Guillemin and Bauer [156] reported a study of the excretion of urinary mandelic and phenylglyoxylic acids after experimental exposures to styrene. Nine healthy male volunteers, 21-34 years old, with normal heights, weights, and vital capacities and with no current styrene exposure were used, some several times. The study was conducted in an experimental chamber that had been previously described by Guillemin [157]. It had an internal volume of 10 cu m, and there were provisions for controlling the temperature (22°C) and humidity (50%) and for exchanging the air. Styrene concentrations inside the chamber were monitored continuously with a total hydrocarbon analyzer and periodically by gas chromatography. The experimental parameters are shown in Table IV-19.

TABLE IV-19

STYRENE EXPOSURE SCHEDULE OF HUMAN VOLUNTEERS

Number of Subjects	Number of Exposures	Duration of Exposure (h)	Concentration (ppm)
5	4	4	99-122
4	1	4	200
4	2	8	42-49
9	3	8	100-112
3	2	3.5	99-101

Taken from Guillemin and Bauer [156]

All urine specimens were collected during exposures and for up to 4 days afterwards; urinary mandelic and phenylglyoxylic acids were determined gas chromatographically. The urine samples were divided into two portions with one being analyzed directly for mandelic acid. Phenylglyoxylic acid in the second portion was found indirectly after it was converted to the trimethyl silyl derivative of mandelic acid as described by Guillemin and Bauer [158] and total mandelic acid representing initial mandelic acid and phenylglyoxylic acid was determined. Both mandelic and phenylglyoxylic acids were found in the urine of subjects within an hour of the beginning of styrene exposure [156]. After the 8-hour exposure at 100-112 ppm, the urinary mandelic acid concentration decreased from 1,000 mg/g of creatinine at the end of exposure to about 10 mg/g of creatinine 56 hours later. Urinary excretion of phenylglyoxylic acid decreased exponentially during the 48 hours after exposure.

Urinary mandelic and phenylglyoxylic acid excretion data obtained from individuals exposed to styrene for 8 hours at 50 ppm were indistinguishable from those obtained from individuals exposed for 4 hours at 115 ppm. Thus, these investigators [156] showed that urinary mandelic acid concentrations reflect exposure dose (as estimated by the product of exposure concentration and duration) rather than exposure concentration. Ratios of mandelic acid to creatinine calculated for urine samples collected at the end of exposure and 14 hours later, and total mandelic acid in all urine collected over a 4-day period after exposure were correlated to the exposure dose in terms of ppm times hours of exposure with coefficients of 0.706, 0.668, and 0.873, respectively. The data demonstrated that there is no advantage to measuring phenylglyoxylic acid or mandelic plus phenylglyoxylic acids over measuring mandelic acid only.

In 1974, Ideda et al. [159] measured urinary metabolites of styrene in six workers who were exposed to styrene during the production of electric motor parts in a Japanese factory. On the day the workers were studied, they were exposed at 50-200 ppm for two 80-minute periods with a 200-minute nonexposure interval. One week later, five of these same workers and an additional worker were studied. The workers were exposed that day to styrene at 4-60 ppm for 120 minutes. Styrene concentrations in the workplace were determined by colorimetric detector tubes and by gas-liquid chromatography.

Urinary hippuric acid concentrations following the first exposure (50-200 ppm) reached a maximum a few hours after urinary excretion of mandelic and phenylglyoxylic acids had returned to normal. At the lower level of exposure (i.e., 4-60 ppm) a week later, no increase in hippuric acid above control values could be detected. The reason for the delayed hippuric acid excretion was considered by Ikeda et al. [159] to be due to a slow step in the conversion of mandelic and phenylglyoxylic acids to hippuric acid. The half-lives of mandelic and phenylglyoxylic acids, and therefore styrene, were estimated to be about 8 hours [159].

In 1981, Pfaffli et al. [160] verified by gas chromatography/mass spectrometry the presence of 4-vinylphenol in the urine of workers in two reinforced plastics factories where average airborne styrene concentrations were about 130 ppm. The correlation between mandelic acid and 4-vinylphenol was good (r=0.93); increasing excretion of mandelic acid was also accompanied by increasing amounts of 4-vinylphenol in the urine. The presence of 4-vinylphenol was not detected in urine of unexposed individuals. The presence of 4-vinylphenol in the urine of styrene workers suggested to the investigators [160] that styrene was also metabolized via arene oxidation, with styrene-3,4-oxide probably functioning as an intermediate. Styrene-3,4-oxide was found in 1982 by Watabe et al. [161] to have potent mutagenicity and cytogenicity toward Salmonella typhimurium strain TA 100. However, other pathways to 4-vinylphenol are possibly present which do not involve arene oxides. The metabolic pathway via the oxidation of the vinyl group is at least quantitatively the more important route as compared to arene oxidation, since the amount of 4-vinylphenol was only about 0.3% of the amount of mandelic acid.

Analytical methods for determining mandelic acid and relationships between urinary mandelic acid concentrations and TWA exposures of workers to styrene are discussed further in Chapter V and in Appendix II.

EFFECTS ON ANIMALS

Toxicity

Effects of styrene vapor exposures at 1,300-10,000 ppm on rats, guinea pigs, rabbits, and monkeys were reported in 1942 by Spencer et al. [53]. The styrene contained 0.01% 4-tert-butylcatechol. Rats (405) and guinea