III. BIOLOGIC EFFECTS OF EXPOSURE

Extent of Exposure

The first major industrial use of benzene was as a solvent in the rubber industry just preceding World War I. [1] During World War I, benzene production was stimulated greatly by the demand for toluene in the manufacture of explosives. The large quantities of benzene which were produced resulted in its more widespread use as a starting point for the manufacture of various organic compounds. This situation led to greatly increased uses of benzene as a solvent in the artificial leather, rubber goods, and rotogravure printing industries, and as a starting material in organic syntheses. [1]

Benzene is a clear, colorless, noncorrosive, highly flammable liquid with a strong, rather pleasant odor. Its physical properties are given in Table XII-1. Today, it is obtained primarily from the petroleum industry where it is produced as a petrochemical from paraffinic hydrocarbons. [2,3] It is also recovered from the gases and coal tar in coke oven operations. The major impurities in commercial benzene (benzol) are toluene and xylene although the commercial form may also be contaminated with phenol, thiophene, carbon disulfide, acetyl nitrile, pyridine, and other substances. "Benzol 90" contains from 80-85% benzene, 13-15% toluene, and 2-3% xylene. The "90" designation refers to the percent of total liquid, by volume, which distills below 100 C.

Industries and processes using benzene include coke and gas, chemical, printing and lithography, paint, rubber, dry cleaning, adhesives, petroleum, and coatings. [2,4] Benzene is also used extensively in

chemical laboratories as a solvent and reactant in numerous chemical applications. [1,5]

During 1967, nearly 800 million gallons of benzene were produced in the United States [3] and by 1969, this figure had increased to 1,185 million gallons with approximately 16% of the production derived from coal. [6] About 87% of the benzene output is used chiefly as an intermediate in producing other organic chemicals such as phenol, cyclohexane, and styrene (see Table XII-2). [3] The remaining amount (13%) is used primarily in the manufacture of detergents and pesticides with small amounts of benzene being used in solvents and paint removal formulations. Benzene is also present in gasoline. [7,8] Petrols (gasolines) in the United Kingdom were reported by Sherwood [9] to be as high as 6% in benzene content and an ad hoc report [10] on European gasolines showed that most of the gasolines tested during 1970 to 1972 were in the 5% range with some up to 16%. Benzene analyses reported in 1972 [11] of 37 unleaded and low-lead gasolines from 15 companies in the United States showed a range from 0.3-2.0% benzene content by volume with an average of 0.8%.

Benzene may also be a component in commercial grades of toluene, xylene, and multicomponent solvent mixtures whose composition varies with intended usage. [5] It is a significant component, ie, 3% or more, in numerous hydrocarbon mixtures such as the aromatic petroleum naphthas whose boiling ranges encompass that of benzene. [5,12]

Although benzene is used generally in enclosed systems wherever possible, exposures can occur from liquid transfer operations, from equipment leakage, from carryover losses, and in maintenance operations.

Exposures also occur from its use as a solvent component in small plant open systems. [1,5]

NIOSH estimates that 2,000,000 persons in the work force have potential exposure to benzene.

Historical Reports

The early uses of benzene, particularly as a solvent, resulted in widespread exposures of workers to its vapor with levels regularly around 500 ppm and some in excess of 1,000 ppm (3,200 mg/cu m). [13]

In 1909, three 14-year old Maryland girls became ill and, within 1 month, 2 of them died following exposure for a period of 4-5 months to the vapors of a commercial grade of benzene used as a rubber solvent in sealing tin cans. [14] Leukopenia was the most striking feature of the blood examination. One girl entered the hospital with a leukocyte count of 1,280 cells/ cu mm which dropped to 480-600/cu mm before death. The second girl was hospitalized with a count of 560 which dropped to 140/cu mm before death. In both cases, there were relative decreases in the polymorphonuclear elements of 43% and 18%, respectively, and the red blood cell counts dropped to 640,000/cu mm in the first girl, and to 1,150,000/cu mm in the second. Both deaths occurred 6-7 days after admission to the hospital. No mention was made of the outcome of the third case.

Numerous other early reports of fatal cases of benzene poisoning have been mentioned in review articles by Greenburg in 1926 [1] and Hamilton in 1931. [15]

Early cases of chronic benzene poisoning include those reported in England by Legge in 1920 [13] of 2 men engaged in spreading balloon fabric

with rubber. Legge's report provided the first measurements of benzene levels in workroom atmospheres to which workers were exposed on a chronic basis. The exposure levels were determined by the firm's chemist and for many of the operations they ranged from 2.1-8 parts/ 10,000 (210-800 ppm) with a peak concentration of 1,050 ppm measured "in front of a fan and at back of machine, machines on both sides, both spreading." However, Legge pointed out that considering the amount of benzene which was being consumed in the poorly ventilated spreading room, the concentration at the end of 1 hour could theoretically have been as high as 16,800 ppm.

With the expanded use of benzene in industry after World War I, an increasing number of reports of chronic benzene poisoning of workers began to appear in the literature. [1,16-18] Because of the seriousness of benzene poisoning, investigations were directed to the many aspects of the cause, recognition, and control of the disease; the results from this research were prominent in the occupational health literature. [19-22] The growing recognition of the hazard associated with the use of benzene led gradually to the substitution of other solvents and an accompanying decrease in the incidence of cases of benzene poisoning.

Effects on Humans

(a) Effects of Inhalation

Browning [23] reported that fatal cases have usually occurred when benzene was inhaled in enclosed spaces such as in tanks containing residues of benzene, and that 13 such cases were reported in Great Britain between 1941 and 1959. The effects observed following such severe exposures were convulsive movements and paralysis followed by unconsciousness. Milder

forms of acute intoxication produced an initial state of euphoria followed by giddiness, headache, nausea, a staggering gait and, if not removed from exposure, a state of unconsciousness. Recovery depended upon the severity of the exposure.

Gerarde [24] noted that breathlessness, nervous irritability, and unsteadiness in walking may persist for a period of 2-3 weeks; furthermore, delayed effects may arise and persist long after the acute incident. The postmortem findings in cases of acute benzene poisoning include extensive petechial hemorrhage in the brain, pleurae, pericardium, urinary tract, mucous membranes, and skin.

Flury [25] stated that single exposures to benzene vapor in the atmosphere at 20,000 ppm may be fatal within 5-10 minutes; 7,500 ppm will produce toxic effects if inhaled for 0.5-1 hour and an exposure to 3,000 ppm may only be tolerated for 0.5-1 hour.

(b) Effects of Oral Exposure

Cases of illness or death resulting from the accidental ingestion of a fluid containing benzene have been reported. [26,27] Liquid benzene causes a local irritation of the mucous membranes of the mouth, throat, esophagus, and stomach. [24] The subsequent absorption of ingested benzene into the blood leads to signs and symptoms of systemic intoxication. [24] The ingestion of a tablespoonful of benzene has been known to cause collapse, bronchitis, and pneumonia. Ingestion as a route of entry of benzene in industrial situations is unlikely except in accidental or intentional situations.

(c) Effects of Skin Exposure

Dermal contact with liquid benzene may cause erythema and blistering of the skin and a dry, scaly dermatitis may develop on prolonged or repeated exposure. [24] Investigations of the percutaneous absorption of benzene have been very limited and from which only qualified estimates can be made.

In 1946, Cesaro [28] reported no observable change in the urinary inorganic sulfate to total sulfate ratio as evidence of absorption of benzene during 20- to 30-minute exposures of the arms or whole bodies of male human subjects to cotton soaked with benzene.

Conca and Maltagliati [29] in 1955, also reported no urinary sulfate changes and detected no benzene in the expired breath of 3 men whose arms had been immersed in benzene for 25-35 minutes. A colorimetric method of unstated sensitivity was used for the breath analyses.

In 1961, Hanke et al [30] reported the rate of human skin absorption of liquid benzene applied under a closed cup as 0.4 mg/sq cm/hr using an ultraviolet spectrophotometric method to determine the amount of benzene remaining from a known quantity exposed to the skin for 10-15 minutes under controlled conditions. This compared with later findings by his coworkers of 22-23 mg/sq cm/hr for ethylbenzene [31] and 14-23 mg/sq cm/hr for toluene. [32] These findings support the belief that liquid benzene is poorly absorbed through the intact skin.

(d) Absorption, Distribution, Metabolism, and Excretion

Srbova et al [33] in 1950 reported on 23 human volunteers exposed to benzene vapor at levels ranging from 47-110 ppm. The subjects inhaled a mixture of air and benzene, usually for 2 hours (occasionally for as long

benzene vapor at levels ranging from 47-110 ppm. The subjects inhaled a

as 3 hours), during which time samples of inhaled and exhaled air were taken every 15 minutes and analyzed polarographically. Blood and urine samples were also collected and analyzed at different times. The absorption of benzene was reported to be greatest during the first 5 minutes, decreasing rapidly thereafter, and becoming constant after approximately 15 minutes of exposure. After 1 hour, approximately 50% of the inhaled benzene was absorbed. Following benzene exposure, 30-50% of the absorbed benzene was eliminated through the lungs, only 0.1-0.2% was eliminated unchanged through the kidneys, and the remainder was metabolized. Complete equilibrium between the concentrations of benzene in the air and in the blood was not achieved because the duration of the experiments was too short. Benzene removal through of the lungs was also followed in 10 subjects with 16.4-41.6% of the retained benzene being eliminated within 5-7 hours. The rate of benzene elimination was greatest during the first hour and decreased slowly thereafter.

Teisinger et al [34] in 1952 reported on exposing 15 human subjects to an average benzene vapor concentration of approximately 100 ppm for 5 hours. They reported an average retention of 46% of the inhaled benzene, elimination of 12% through the lungs following exposure, and only 0.1-0.2% of the unmetabolized benzene eliminated in the urine. Approximately 29% of the absorbed benzene was metabolized and excreted in the urine in the form of phenol, 2.9% as pyrocatechol, and 1% as hydroquinone.

In 1946, Duvoir et al [35] reported that in subjects exposed to 6,000 ppm of benzene 28-34% was retained and absorbed into the blood. The authors found that approximately 55-60% of the benzene in the blood became fixed in the bone marrow, fatty tissues, and the liver. The remaining 40-

45% was excreted unchanged through the lungs. The absorbed portion was then metabolized through oxidation to phenol and diphenols and eliminated as such or as esters of sulfuric and glucuronic acids. Through this metabolic process, benzene decreased the organic sulfate reserve.

Hunter, [36] using gas chromatographic analysis, stated that at a benzene concentration of approximately 35 ppm a healthy adult male reached a relatively steady state in approximately 5-7 minutes. Approximately 47% of the benzene in the inhaled air was absorbed. The major portion of the urinary phenols was conjugated with glycine, sulfuric acid, or glucuronic acid with up to 8% of the phenols being excreted in the free form.

According to Gerarde, [2] benzene saturation of the circulating blood is very rapid, reaching a 70-80% saturation level within 30 minutes. Relatively complete saturation, however, may require as much as 2-3 days. The author suggested that the fatty tissue, which has a great affinity for benzene, removes and stores the benzene carried by the blood; however, this fatty tissue in many instances has a very meager blood supply and requires a relatively long period to attain equilibrium.

Benzene is best known in industrial exposure situations for its chronic forms of poisoning and specifically for its injurious effect on the hematopoietic system.

Erf and Rhoads [20] presented in 1939 the results of blood findings in 9 individuals, 6 of whom were rotogravure printers employed in a plant from which Greenburg et al [17] also reported on an investigation (see Epidemiologic Studies). The authors stated that "no correlation between the severity of disease and the intensity of exposure can be made." The duration of exposure ranged from 6 months to 3 years, with the symptoms of

poisoning being present from 1-6 months before medical aid was sought. The hematologic findings varied; however, anemia, leukopenia, thrombocytopenia, and elevated reticulocyte levels were present in all cases. Biopsy tissue from the sternal bone marrow of 8 of the patients showed microscopic changes varying from a hypoplasia with immature cellular elements to a hyperplasia with normal maturation. Following 2-5 months of treatment, 8 of the 9 patients were clinically improved; the ninth subject developed leukemia and died. In 1918, the man had worked with his brother in a studio where benzene was used. During that year his brother developed epistaxis and anemia and died. The man then changed his occupation without further exposure to benzene until 17 years later when he obtained employment in the rotogravure plant where he was exposed to benzene vapor for 14 hours a day. Upon hospitalization, leukemia was diagnosed. He did not respond to treatment, the WBC increased in number to 137,000/cu mm with the majority being identified as myelocytes, and the spleen and all peripheral lymph nodes became enlarged. After 5 weeks of therapy he left town and died 2 months later in another city. Post-mortem examination revealed a diffuse infiltration of the organs with immature myeloid cells, a finding typical of myeloid leukemia.

In 1967, Stewart et al presented information concerning 10 chronically exposed benzene workers. Only an abstract of the paper was reported [37]; subsequent publication of the data of potential importance has not resulted. The workers, chronically exposed to benzene (less than 25 ppm) for several years, were accidentally overexposed (85-115 ppm) to benzene for a period of 3 months. Six complained of fatigue and all showed signs of mild anemia. Nine of the 10 recovered in 4-8 months after which

time they were returned to the benzene area and were maintained under a strict medical surveillance program. It included continuous breathing-zone monitoring and frequent analysis of breath for benzene vapor.

Epidemiologic Studies

The signs and symptoms of chronic benzene poisoning can effectively be described from a report by Helmer [38] in 1944. Because of the difficulties in importing appropriate solvents during World War II, the use of benzene increased markedly in a Swedish plant which manufactured rubber raincoats. Work was performed on a conveyor belt with alternate sewing and gumming, the latter being done mainly by hand on open tables using a solution of 10% rubber in heated benzene. The total amount of benzene used was unspecified; however, at full worker capacity, about 50 kg of benzene evaporated in an 8 1/2-hr workday. The atmospheric benzene concentration was estimated to be approximately 17 mg/liter (5,320 ppm) based upon its rate of consumption and even distribution over the premises. The work force had been reduced to one-third of normal along with reduction in the total output (8 kg benzene consumption/workday) when inspections were conducted by the State Institute of Public Health. With the decreased benzene consumption and fan-installed improved ventilation, environmental analysis showed a benzene content of 0.44-0.70 mg/liter of air (140-220 ppm). There had been no mechanical provisions employed for exhaust ventilation before cases of benzene poisoning were encountered. The study showed that 184 workers (169 women and 15 men) from the rubber plant, of which 60 workers (58 women and 2 men) were entered on the sick list suffering from chronic benzene poisoning. In those workers afflicted,

headaches (73%) and fatigue (88%) were prominent, persisting for many months, even after the blood picture had improved markedly. The subjects complained of having to lie down after performing very simple household chores and not even being able to take short walks. There was an increased tendency to bleed; cutaneous hemorrhages were noted (48%), mainly in the legs and arms. The hemorrhages were often large with initial spreading, and would appear without demonstrable trauma. Other abnormal bleeding occurred in the gums and nose, as well as irregular, sometimes more frequent and copious menses. The latter affected only women who had suffered more serious blood changes. Other common troubles included nervousness, vertigo, somnolence or sleeplessness, shortness of breath, and palpitations. Dyspeptic disorders, nausea, vomiting, and loss of appetite appeared in 22% of the cases. Two subjects complained of a benzene taste in the mouth. Skin changes were manifested in the form of itching, possibly with pruriginous papules or slight dermatitic changes. There was a loss of weight in 9 cases, some up to 10 kg (approximately 22 pounds) in one year. There were reports of prickling sensations in the arms and legs. Eight subjects complained of smarting in the eyes. It was pointed out, both by Helmer [38] in this study and by Greenburg et al, [17] that symptomatic effects often do not correlate with objective findings. Symptomatic effects may be absent, even in serious cases of chronic benzene poisoning.

The unique aspect of chronic benzene poisoning resulting from exposure to benzene vapor over prolonged periods of time is its effect on the blood-forming system. There is a distressing lack of exposure-effect data in the literature; therefore, only approximations of worker exposure

can be made. One exception is the early account in 1926 by Greenburg [19] in which he reported on the complete blood counts (CBC) of workers exposed to concentrations of benzene ranging from 90-1800 ppm (undescribed sampling and analytical method) in 18 workrooms during both winter and summer seasons. The data enable estimates of the effects of local ventilation and seasonal changes on the benzene content in the air. Even so, concentrations were reported as averages; ranges are unknown but may have an important bearing on individual workers, especially those determined to be positive (see below). Summaries of the blood findings and average benzene-in-air concentrations are presented in Tables XII-3 and XII-4. Originally, the most important early sign of benzene poisoning was thought to be the change in the white blood cell (WBC) count. Greenburg considered cases as positive which showed less than 5,500 WBC/cu mm (7,500-9,000 was considered the normal count). A reduction of the WBC count to less than 4,000 and the red blood cell (RBC) count to less than 4 million /cu mm was found in 10 of 26 workers. Three of 8 workers studied in detail showed less than 50% polymorphonuclear leukocytes, 2 showed a lymphocyte population greater than 45%, and 2 showed eosinophils of more than 5%. In all groups listed in Table XII-3, there were workers who showed a picture of chronic benzene poisoning as judged by reductions in the WBC count. The hazard from the use of benzene was evidently not entirely removed at average benzene concentrations in the vicinity of 70-90 ppm. Variations were noted in the individual susceptibility to benzene poisoning as well as to wide variations in the quantities of benzene used during the year; therefore, only qualified estimates can be made to correlate the benzene exposure levels with the clinical findings.

unreliable. Only qualified estimates can be made to correlate the benzene

In 1939, Greenburg et al [17] reported on the results of blood examinations performed on 332 workers exposed to benzene vapor in 3 rotogravure plants. In Plant A, exposures ranged from 50 to over 1,000 ppm; in Plant B, from 24-675 ppm, and in Plant C, from 11-57 ppm in the first floor pressroom, from 182-298 ppm in the proofroom, and from 25-200 ppm in the 11th floor pressroom. The method of sampling and analysis was not described. The extensive results of the blood tests were grouped for the workers from the 3 plants which prevents relating the blood findings to the reported exposure levels in the separate process areas. Of the 332 workers examined, 130 were found to be suffering with varying degrees of benzene poisoning, 22 of these to a severe extent whereas 43 were early cases. The RBC count was less than 4.5 million in 48% of these subjects, the platelets were less than 100,000 in 33%, and the WBC count was less than 5,000 in 15% of the total workers, in 30% of the early cases, and in 86% of the 22 severe cases of poisoning. The hemoglobin (Hgb) was less than 13 g/100ml of blood in 15% of 235 workers examined.

From the detailed blood studies which were performed on 102 of the workers, the incidence of significant abnormalities has been summarized and is presented in Table XII-5. In the early mild cases, the most frequent changes were a reduction in RBC (72.1%) and an increase in average cell size (58.1%). Since macrocytosis was also shown (24.3%) in the 9 otherwise negative cases, it was suggested that an increase in mean corpuscular volume (MCV) and a reduction in the RBC count constituted a more sensitive index of benzene poisoning than did WBC reduction. Various combinations of the 5 most commonly used blood tests (Table XII-6) indicated that 82% of poisoning cases could be revealed by a combination of MCV and RBC

determinations. It was possible to detect poisoning in even more workers when the determinations were combined with WBC counts (93%) and finally, 97% when thrombocyte examinations were added.

Savilahti [39] described in 1956 the clinical findings of 147 workers exposed to benzene for more than 10 years in a shoe factory where air analyses at 3 working stations 6 months preceding the medical study provided average benzene values of 318, 433, and 470 ppm, respectively. Hematologic abnormalities were found in 73% of the workers; thrombocytopenia, 62%; leukopenia, 32%; anemia, 35%; and anemia, leukopenia, and thrombocytopenia simultaneously in 31 subjects. Of those affected, 1 died and 120 became asymptomatic within 3 months following removal from exposure to benzene. Of the remainder, 1 patient was still in the hospital after 1 year, 6 were at home on sick leave, and 20 continued to show relatively minor hematologic symptoms.

Juzwiak [40] published in 1969 the results of blood examinations on 585 persons employed in 13 shoe plants where they were exposed to benzene vapor. From 1960 to 1963, "Butapren" glue was used, consisting of (literally translated) 40% extraction benzene (probably petroleum benzin), 26% technical benzene, 2% toluene, 26% ethyl and butyl acetates, and other "harmless components." Fluctuations in mean benzene concentrations from 0.1-0.5 mg/liter of air (31-156 ppm) were reported. In 1964, the toluene content of the glue was increased to 29% to replace the technical benzene entirely; nevertheless, mean benzene concentrations of 0.13-0.14 mg/liter (41-44 ppm) were still recorded. In addition to the toluene, gasoline was also known to be present in the chemical composition of the glue. Commercial toluene and gasoline regularly contain benzene. Seventy-three

present in the chemical composition of the glue.

percent of the workers were reported to have reduced RBC counts, 8.5% had reduced WBC counts, and 91% had reduced Hgb levels. Again, the lack of adequately documented environmental data in support of medical findings precludes any confident correlation of exposure and effect.

Cases of leukemia reportedly due to benzene exposures first appeared in the literature of the 1930's. [17,22] Mallory et al, [22] in 1939, presented necropsy or biopsy protocols from 19 cases having chronic exposure to benzene. They pointed out that early phases of benzene poisoning were not exemplified in their report since all but 3 of the cases were fatal and the early death of 1 of the remainder was expected, based on a diagnosis of aleukemic leukemia. None of the reported cases had less than 6 months' contact with benzene vapor and only 4 cases had less than a year's exposure. Table XII-7 lists the presumptive duration of contact and interval between the last contact and death or biopsy in the 19 cases of chronic benzene poisoning. Of special note are 2 cases of verified leukemia:

(1) The subject had been exposed to benzene for 10 years and, according to the authors, had shown hematologic evidence of benzene poisoning. He developed a typical picture of acute myeloid leukemia in the last 3 months of his life. Autopsy showed the characteristic myeloid infiltration of the liver, spleen, and bone marrow. In addition, a true leukemic tumor, 4 cm in diameter, was localized in the liver.

(2) A boy of 12, a painter's son, played in his father's shop, frequently using a paint remover known to contain benzene to remove coats of paint from toys. He developed a clinical picture of aplastic

anemia but tissue biopsy revealed a typical leukemic replacement of the marrow.

The authors described "a neoplastic tendency" for benzene as evidenced by the degree of aplasia, excessive mitotic figures, and all development having no counterpart in normal tissues but common to malignant tumors. They concluded that the evidence that chronic exposure to benzene produced leukemia in human beings was incomplete but sufficient to command serious consideration.

Vigliani and Saita [41] reported in 1964 on 6 cases of benzene-associated leukemia which had been observed by them. Meager environmental data were presented which applied to 2 of the cases.

(1) A 38-year-old man became an operator in a rotogravure firm which used inks containing 40% benzene. According to the authors, the benzene concentration in the department where the man worked varied between 0.60 and 2.10 mg/liter of air (190-660 ppm). Four years later, he was hospitalized with generalized depression of the formed elements of the blood. Post-mortem findings were myeloid metaplasia of the liver and spleen.

(2) A 24-year-old man began work in the same rotogravure department as the operator described above. Seven years later (1945), when the first worker died of leukemia, some of the other workers in the department showed signs of benzene poisoning and were examined; however, benzene poisoning was apparently not suspected in this man and he was not examined. It is not known whether he had been examined prior to the death of his colleague. In 1949, at the same time the plant replaced benzene with other solvents, he showed a slight leukopenia. Subsequent quarterly

routine medical examinations showed him normal until 1961 when serious signs and symptoms of leukemia appeared. In spite of intensive therapy, death occurred approximately 1 year after the clinical appearance of the disease.

The incidence of benzene-induced chromosome changes in peripheral blood lymphocytes or bone marrow has received increased interest. [42-49] Significantly increased rates of "unstable" and "stable" chromosome aberrations were observed and were still present several years after cessation of exposure to benzene. [48] Followup studies showed a tendency toward a decrease in unstable chromosome changes and a persistence or an increase in stable changes. Occasional abnormal clone formations were observed. These changes are similar to those reported in individuals with past exposure to ionizing radiations, both therapeutic [50] and accidental. [51-53] According to Forni et al, [48] the implications of the chromosome findings with respect to the problem of benzene leukemia are not clear. Persistent chromosome changes in lymphocytes seem simply to indicate that damage has occurred. However, stable chromosome changes in the bone marrow might give rise to leukemic clones, as has been demonstrated in 2 reported cases of benzene-associated leukemia. [44,45] An interesting case involving pregnancy was reported by Forni et al [48] in which a patient, while severely pancytopenic and with severe hemorrhagic problems, delivered an apparently normal boy. All chromosome studies in the patient showed an increased rate of chromosome aberrations; however, a cytogenetic study of the peripheral blood performed on the newborn boy did not show chromosomal abnormalities. In 1969, the patient had another pregnancy and delivered a normal daughter.

abnormalities. In this the

In reviewing reports of chromosomal aberrations observed by Pollini and Columbi, [42] Vigliani and Saita [41] suggested a possible mutagenic effect on blood cells which may help to explain the appearance of leukemia during the course of a benzene hypoplastic anemia. The authors made no conclusion concerning the existence of a true "benzene leukemia" because of a lack of extensive analysis of the incidence of leukemia among workers exposed to benzene as compared with that of a carefully evaluated control group. In a leukemia survey reported by Thorpe, [54] emphasis was placed on the need for improvement in the recording and storage of biological observations, job histories, occupational exposures, and demographic data. A comparison by Vigliani and Saita [41] of the incidence of acute leukemia among the general population in Milan from 1959 to 1961, however, indicated an incidence of about 1 case among 20,000 people. Statistics from the Italian National Institute for Insurance Against Accidents and Occupational Diseases as reported by Vigliani and Saita [41] showed a sharp rise in the reported cases of acute leukemia among local residents in 1962 to 1963, coinciding with the increase in cases of benzene poisoning in workers. The rise in leukemia cases was about 20 times higher than expected. The incidence of leukemia was especially striking when the fatal general population cases were considered: out of 26 deaths, 11 were due to leukemia and 15 to aplastic anemia. [41] Their figures corresponded well with those of Cavignaux [55] for 1960 and 1961 which pointed out the high incidence of leukemia among cases of benzene poisoning in France. Vigliani and Saita [41] emphasized that, "Great caution must be exercised before admitting the benzene etiology of chronic myeloid or lymphatic types of leukemia."

admitting the benzene etiology of chronic myeloid or lymphatic types of

Browning [23] has tabulated 60 cases of leukemia among benzene workers in her 1965 text. She found no correlation between the original authors' reported medical findings and benzene exposure levels.

In a 1969 report of the health status of 765 female workers in the leather industry in Lodz, Poland, Butarewicz et al [56] provided data showing blood changes in 18.6% of 350 workers exposed to a benzene-containing adhesive, as compared with blood changes in only 5% of 246 workers exposed to a benzene-free glue and 3.5% in 169 control female workers who were not exposed to either of the adhesives. The atmospheric benzene levels were not well defined, the highest concentrations noted in one of the zones being reported as more than 1.2 times (37 ppm) the maximum permissible concentration of 100 mg/cu m (31 ppm). There were wide variations in the air analyses as a function of the season, room temperature, the number of dryers operating, and the efficiency of the ventilation; therefore, essentially no exposure-effect comparisons can be made. Such a large population of workers exposed to benzene in recent years would have provided an excellent opportunity for the development of correlative data between blood changes and other medical findings with measured environmental exposures to benzene.

The study of Hardy and Elkins [57] in 1948 emphasized that benzene poisoning, as evidenced by laboratory blood studies, frequently occurred without any indication of clinical signs or symptoms. A small Massachusetts rubber coating firm experienced the death of an employee which was diagnosed as benzene poisoning. Subsequently, a blood study was performed on all 52 workers employed by the company. Sixteen workers showed deviations in more than 1 blood element. The results of air

analyses, taken on 3 different occasions, are listed in Table XII-8 and indicate what the authors termed remarkable uniformity. There had been no significant changes in the ventilation of the plant or in the plant's operational procedures during the 8-year period preceding the complete blood study; however, during the wartime period of 1942-1946, most of the men had put in considerable overtime work, "averaging much more than eight hours a day." According to the authors, [57] of the 16 men with abnormal blood pictures, 6 worked in the coating room and, in all probability, were exposed to average benzene concentrations of not over 60 ppm; 2 men who worked in the mixing room were possibly exposed to 80 ppm; and 1 man who cleaned the cans may have had a considerably higher exposure. Followup studies of the 16 workers, either 4 months or 10 months later, showed only 4 workers with relatively normal hematologic values at the time the report was written, 10 months after benzene exposure ceased entirely. It was concluded from this study that the maximum allowable concentration of 75 ppm of benzene which was accepted by the State of Massachusetts at that time was too high, and subsequently the figure of 35 ppm was used.

Additional data on effects of benzene on the blood picture were provided in 1961 by Pagnotto et al [12] from a study of the Massachusetts rubber coating industry. By that time, the use of benzene as an industrial solvent in large quantities had diminished considerably; however, petroleum naphthas containing varying amounts of benzene up to 9.3% were used heavily in the rubber coating industry. Their study covered 11 plants which involved practically all the large Massachusetts plants and some of the smaller ones. Of 65 environmental determinations taken, only 4 were recorded above 40 ppm, the highest being 125 ppm. Air sampling and

analyses were performed by collecting the benzene vapor on silica gel and analyzing the desorbed benzene by ultraviolet spectrophotometry. In addition, urinary phenol determinations were performed on 162 workers and compared with the atmospheric exposure levels as shown in Figure XII-1. According to the authors, it was apparent from the air-urine correlation that the phenol test was a good index of benzene exposure. A limited number of Hgb determinations were also performed on workers in 3 of the plants. The results of the blood tests on 47 men representing spreader, saturator, and churn operations showed in Plant A, 5 out of 27 workers had Hgb levels below 13.5 g/ 100 ml of blood and 2 out of 32 had RBC under 4 million/cu mm. In Plant B, 1 worker out of 9 examined showed deviations in Hgb and RBC below the criteria listed for Plant A. No abnormal findings were reported in Plant C.

The domestic and foreign literature dealing with the effects of benzene on exposed workers consists primarily of medical reports rather than documented, comprehensive frequency and distribution studies encompassing both medical and environmental findings. Published definitive epidemiologic data have been difficult to find on workers exposed to benzene vapor at specific concentrations for prolonged periods of time.

Kozlova and Volkova [58] in 1960 reported on observed changes of the formed elements of the blood and phagocytic activity of leukocytes in workers exposed to benzene in a leatherette factory over a study period of 5 years, 1953-1957. The blood changes were studied in 252 production workers; phagocytic activity of leukocytes was also determined from 157 of the subjects. Environmental concentrations of benzene fluctuated from 150-1,000 mg/cu m (47-310 ppm) during the first 3 years of the study. At the

end of 1955, environmental concentrations were reduced due to installation of improved control measures, and in 1957, exposure levels reportedly did not exceed 80-150 mg/cu m (25-47 ppm) average concentrations. Worker job assignments remained relatively unchanged during the course of the 5-year period. Phagocytic activity of leukocytes was measured by the average number of bacteria engulfed by neutrophils in a 1 1/2 billion suspension of Bacillus Fridmani. For analysis of the data, 3 groups were selected based upon environmental benzene levels in the plant areas. Group I, consisting of 121 workers, was exposed at benzene concentrations exceeding 250 mg/cu m (about 78 ppm) which was more than 5 times the then existing maximum permissible concentrations of 50 mg/cu m (16 ppm). Group II consisted of 60 workers exposed to concentrations of 100-200 mg/cu m (31-62 ppm), 2-4 times the maximum permissible concentration, and Group III, 71 workers not having production assignments involving benzene but whose exposures were from 75-125 mg/cu m (24-39 ppm) due to benzene vapor in the proximity of the main production areas. The Group III exposure levels were 1 1/2 to 2/1/2 times the maximum permissible concentration. Marked alterations of blood formed elements were reported for all groups and the authors [58] noted that decreased leukocyte and thrombocyte counts were observed more frequently in workers employed 5-10 years than were changes in erythrocyte composition. Furthermore, it was stated that with prolonged contact with small concentrations of benzene (Group III exposure levels), leukocyte depression occurred prior to depression of the erythrocytes. In Groups I and II, the prevalence of neutropenia was closely related to the length of worker employment. Because of the leukocyte depressions observed, and particularly with the neutrophils, it was believed that phagocytic activity

provided a measure of individual susceptibility to benzene. Phagocytic activity was decreased in 86% of Group I workers and 60% of Group III workers. It was found that phagocytic activity of the leukocytes, as a rule, decreased in the majority of workers sooner than blood alterations were noted. It was suggested that phagocytic activity of the leukocytes was a more sensitive test for benzene poisoning than was observation of morphological changes of the blood.

Horiuchi et al [59] presented in 1963 the effects of benzene exposure in 373 male workers engaged in paint manufacture of coating operations in 14 workshops. Thirty-minute breathing zone samples were taken along with clinical laboratory tests which included complete blood cell counts, specific gravities of whole blood and serum, urinary coproporphyrin and total sulfate ratios, and subjective clinical symptoms obtained by a questionnaire. Workshops were grouped according to the range of benzene concentrations encountered as: Group I, 6.6-78.5 ppm; Group II, 3.4-35.9 ppm; Group III, 0.3-22.1 ppm; and Group IV, trace-1.8 ppm. From the frequency of "abnormal" findings (unspecified as to what constituted abnormal) it was concluded that effects noted in Group IV were essentially the same as for workers not exposed to benzene. Based on the higher incidence of abnormal findings in Groups I and II compared with Group III, the authors concluded that the maximium allowable concentration of benzene in the workroom air should be below the maximum encountered in Group III, that is, approximately 20 ppm.

Followup studies [LD Pagnotto, written communication, October 1972] of the rubber coating industry originally reported by Pagnotto et al [12] were made through 1963 and results are summarized in Table XII-9 for Plant

A for which the most complete information was available. Two naphtha solvents were used in Plant A, (3% and 7.5% benzene by volume) until 1965 when toluene-containing solvents were introduced. Table XII-9 lists 12 of 35 workers who were involved in the study. Although information is minimal, the table provides information as to environmental benzene concentrations and worker exposures as measured by urine phenol levels over a 3 1/2 year period from 1960 through 1963. Environmental benzene concentrations for spreader and churn operations consistently averaged between 20 and 25 ppm and frequently were lower. The saturator operation indicated fluctuations as high as 140 ppm and it appears that saturator environmental levels in the vicinity of 70-90 ppm were encountered rather frequently. Of particular significance is the generally close agreement observed between environmental benzene concentrations obtained from laboratory analysis and equivalent air levels for individual worker exposures as determined from Table XII-10. This again indicated the value of urinary phenol determinations as a measure of exposure to benzene.

Blood Hgb levels of some of the workers are listed in Table XII-11. The data are very meager and no firm conclusions can be drawn from the information. In one of the 1961 studies, 27 workers were tested and 6 were found to have Hgb levels below 13.5 g/100 ml of blood. In the 1963 study, 12 of 24 workers examined were reported to have normal blood pictures. The remainder were said by the company to show minor deviations from normal, although these differences were unspecified and the information was unavailable. Blood studies from Worker L were reported to have had an appreciable deviation from the normal. There was no knowledge of his blood picture prior to being hired by the rubber coating firm. He was removed

appreciable deviation from the normal. There was no knowledge of his blood

from the job in 1963. Worker H was the saturator operator reported in Table XII-9 and had the highest benzene exposures. He was still working at the saturator job in June, 1967.

(a) Sex, Pregnancy, and Age

In 1939, Hunter [21] reported on a study of 70 male and 19 female workers exposed to benzene whose histories, physical examinations, routine urine and complete blood examinations were conducted over a period of 4 years. The workers were divided into 3 main classifications consisting of group 1, those showing a normal blood picture; group 2, those showing only 1 abnormal feature; and group 3, those showing 2 or more deviations from the normal. Two of the 10 fatal cases in the study were female. In one factory where 43 workers were studied, although the only fatal case was a young girl, 26 of the workers were men (60.5%) and about 28 of them (64%) had a depression of the polynuclear percentage. The author found no supporting evidence that women were more liable to development of benzene poisoning than men and suggested that facts from the study cast considerable doubt on whether a female hypersusceptibility to benzene existed. Hunter emphasized that the early diagnosis of benzene poisoning depends upon an evaluation of the complete blood picture rather than upon the existence of a leukopenia alone. No environmental exposure data were given in this report.

Mallory et al [22] in a companion paper to that of Hunter suggested from either necropsy or biopsy material obtained from 4 females and 12 males that hypoplastic reactions might be more common in the female than in the male, thus supporting an opinion at the time that females were more

susceptible than males to the effects of benzene poisoning. The authors recognized that their figures were too few to be conclusive.

In 1928, Smith [16] reported on a study of 79 women, 25 with confirmed chronic benzene poisoning and 5 suspected cases. Complete histories were obtained which were designed to reveal past and present exposure to benzene. In addition, complete blood counts were taken, consisting of Hgb, RBC, and WBC, including differential counts. Findings were not compared with those of male workers but the results did not differ from those reported in men by other investigations. The age spread of the women studied was quite evenly distributed from 17-52 years. The author found that susceptibility to benzene poisoning appeared to be about equally marked among young and older women and that youth did not seem to be a predisposing factor in the development of benzene poisoning. In addition, the menstrual function was undisturbed in the majority of positive and suspected cases. The few incidences of menstrual irregularities were not considered to be of concern.

In 1956, Cassan and Baron [60] mentioned the greater susceptibility of women to benzene poisoning, particularly when they were pregnant. Because the study involved women exclusively, no firm conclusions may be drawn from this report regarding sex differences in susceptibility to benzene poisoning.

Savilahti [39] reported in 1956 that he did not find any correlation between age, sex, and symptoms of benzene poisoning in the study of 147 workers (98 women, 49 men). Subjects showing abnormal blood changes ranged in age from 16-66 (average=36) while those unaffected were from ages 18-66 with an identical average age. Twenty-four of the 41 female workers and 18

in age from 16-66 (average=36) while those unaffected were from

of the 35 male workers became ill; thus, no significant differences were observed between age or sex.

Animal Toxicity

(a) Acute Exposures

Lazarew et al [61] reported in 1931 that liquid benzene could be absorbed through a rabbit's paw by measuring the weight increase resulting from the absorption of benzene vapor by activated silica gel from the exhaled air after preliminary removal of water and carbon dioxide by phosphorus pentoxide and soda lime. The animal served as its own control by providing an exhaled air sample through a tracheal canula for analysis preceding the 30- to 60-minute exposure. The weight increases of the silica gel adsorption tubes during the experimental runs were 2-3 times greater than the controls. No quantitative data were available and the degree of benzene absorption is unknown.

In 1944, Carpenter et al [62] described their observations of ten rabbits undergoing anesthesia with 35,000-45,000 ppm of benzene vapor in air. The average time required for light anesthesia was 3.7 minutes, 5.0 minutes for excitation and tremors; death ensued in approximately 36 minutes.

In 1965, Jonek et al [63] reported on a histochemical study of enzymatic changes in the central nervous system of mice subjected to acute benzene poisoning by single inhalation exposures at the 60 mg/liter (18,750 ppm) and compared the findings to those observed in a control group. Changes in the activity of enzymes suggested a selective influence of benzene on oxidation in neurons. This reaction was not the same in all

neurons; some showed enzymatic activity similar to that observed in the control group, while in others, activity was less than that in the controls or even absent. The authors postulated that the observed changes in the enzyme activities may be the result of a direct lesion of the lipoprotein membranes of the structural elements of the cells by benzene or the products of benzene metabolism.

(b) Chronic Exposures

In 1941, Schrenk et al [64] reported, in an extensive study of dogs exposed continuously (24 hr/day), intermittently (4- or 8-hr/day), and singly to benzene vapor, that (1) the initial absorption of benzene was extremely rapid (nearly complete within 30 minutes) with later absorption approaching equilibrium more slowly (over several hours), (2) a linear relationship existed between the concentration of benzene in the air and the equilibrium concentration in the blood, (3) distribution of benzene throughout the body occurred rapidly, (4) the fat, bone marrow, and urine contained approximately 20 times the concentration of benzene as the blood, (5) benzene concentration in the muscle and vital organs was 1-3 times that in the blood, and (6) the RBC contained approximately twice the amount of benzene found in the plasma. The blood benzene values were determined by either a nitration method or a rather involved method in which carbon dioxide was formed from the combustion of benzene and ultimately measured by changes in electrical conductivity through precipitation as barium carbonate from a barium hydroxide solution. The method provided 70-112% recoveries of added known amounts of benzene. The blood samples were drawn before and after the repeated daily exposures to benzene. The exposures ranged from 2-37.5 hours for different animals. The results of this work

before and after the repeated daily exposures to b

showed that for each 100 ppm of benzene vapor in air, the equilibrium blood concentration in terms of milligrams of benzene/100 ml of blood was 0.21. When the concentration of benzene in blood and air were calculated in mg/liter, the coefficient of distribution obtained by dividing the blood concentration by the air concentration was found to be 6.58.

Desoille et al [65] described in 1967 the effects of exposure to benzene on virgin and pregnant guinea pigs from a dose of 0.1 g/kg using benzene in olive oil and injected subcutaneously each day for a period of 9 weeks. A study of the variations of RBC and WBC counts was made before, during, and after pregnancy. Pregnancy did not enhance the hemotoxic effects of benzene.

Deichmann et al [66] in 1963 published results after exposing 8 groups of rats to analyzed benzene concentrations extending from 15-831 ppm for periods ranging from 5 weeks to 7 months. A significant leukopenia resulted after 2-4 weeks of exposure to the 3 highest exposure groups of 831, 65, and 61 ppm. Exposure to mean concentrations of 47 and 44 ppm, 7 hours/day, 5 days/week, in separate experiments induced a moderate but definite leukopenia after 5-8 weeks of exposure. The WBC dropped from 12.1 to 10.4 thousand/cu mm in the males and from 11.3 to 9.4 thousand/cu mm in the females when exposed to 47 ppm (range, 33-55). In the groups exposed to 44 ppm (range 40-50), the WBC dropped from 15.2 to 10.0 thousand/cu mm in the males and from 11.8 to 7.7 thousand/ cu mm in the females. No leukopenia was observed in the groups of rats exposed to average concentrations of 31, 29, or 15 ppm or in the controls.

Nau et al [67] exposed rats to a benzene concentration of 1,000 ppm for 23.5 hours/day, 7 days/week. After 183 hours of exposure, the rats

appeared to be in "poor" condition and suffered a loss in body weight. They hemorrhaged from the nose and mouth, the stomach was distended, the gut was empty, and the blood vessels of the lungs, liver, kidneys, intestines, and omental tissues were engorged. The WBC fell from a mean value of 22,650 to 5,425/cu mm by the 105th day, and there was a reversal of the polymorphonuclear-lymphocyte ratio from 22:57 in the preexposure period to 54:46 at the end of 105 days of exposure. Microscopic studies of the bone marrow showed an increase in the proportion of RBC precursors. Nau et al [67] also showed a decrease of WBC (no values given) after about 90 days of repeated daily exposures of 8 hours/day, 5 days/week at the 200 ppm level, but there was no change in the polymorphonuclear-lymphocyte ratio. Microscopic examination of the bone marrow showed some depression of myelocytic activity and stimulation of erythrocytic activity. Rats similarly exposed at 50 ppm had a decrease in the WBC (no values given).

In addition, the development of bilateral cataracts was reported in 50% of the rats after 600 hours of exposure to the 50-ppm concentration of benzene. However, observation was not found in the rats exposed to 200 ppm of benzene undergoing the same 8-hour/day, 5-day/week regimen after 750 hours of exposure. The rats exposed to 50 ppm also developed, as did the rats at the 200 ppm exposure level, lower leucocyte DNA values, depression of myelocytic activity, and a stimulated erythrocytic activity in the bone marrow.

Wolf et al [68] published in 1956 the results of their toxicologic studies of benzene and certain of its alkylated derivatives. These investigators noted slight histopathologic changes in the blood and testes of rabbits consisting of leukopenia and degeneration of the seminiferous

tubules when exposed to 80 ppm concentrations of benzene for 243 days; in the bone marrow, blood, spleen, and testes of guinea pigs exposed to 88 ppm for 269 days; in the blood and kidneys of guinea pigs exposed to 88 ppm for 32 days; and in the blood and spleen of rats exposed to 88 ppm for 204 days. On the basis of these results, the authors concluded that the no-effect level was below 80 ppm.

Following the belief that the central nervous system has a regulatory effect on hematopoietic activity, and that functional disturbances of the central nervous system preceded changes occurring in either blood morphology or the hematopoietic system to chronic benzene intoxication, Novikov [69] in 1956 reported on changes in conditioned reflex activity in 6 rats exposed at 64 mg/cu m (20 ppm) of benzene vapor and an additional 6 animals exposed at 13 mg/cu m (4 ppm) for 6 hours/day, 6 days/week, for 5 1/2 months. As could be expected with a central nervous system depressant, there was a delay in conditional response time after exposure to benzene; this delay was observed in rats exposed at 20 ppm but not at 4 ppm. It was suggested by the author [69] that the results presented could serve as a physiological basis for an allowable benzene concentration limit in atmospheric air.

Horiuchi et al [70] reported in 1967 on the effects of benzene inhalation on spontaneous behavior in 15 mice as measured by spontaneous motor (wheel-turning) activity. This study was initiated as a result of reports in the USSR literature on behavioral responses. Mice were exposed 6 hours/day for 20 days to benzene concentrations at 10 ppm and 100 ppm. Observations were also made on changes in body weight, RBC's, WBC's, and thrombocyte counts. Thirty days after cessation of benzene exposure,

examinations of the bone marrow, spleen, liver, and kidneys were performed. A decrease in wheel-turning activity was observed by the 5th day in the 5 mice exposed at 100 ppm. The 5 mice exposed at 10 ppm and 100 ppm showed decreases in the RBC and WBC counts, but values overlapped those of the controls. In addition, degenerative changes of the bone marrow were reported in all mice exposed to benzene at 100 ppm and slight degenerative changes in 2 of 5 mice exposed at 10 ppm. The authors emphasized that further study was needed. No attempt was made to relate the animal results to human exposure, but it was concluded that the benzene concentration necessary to prevent effects in mice was below 10 ppm.

(1) Nutrition

Experimental studies with dogs and rats conducted by Shils and Goldwater [71] showed that an inadequate protein intake predisposes to increased susceptibility to benzene poisoning. They stated that no appreciable effect of varying the fat content of the diet has been demonstrated convincingly. On this basis, they recommended that the benzene workers have well balanced meals containing sufficient of high quality protein. This recommendation was made with a view to the role of the sulfur-containing amino acids and of choline in influencing the fat content of the liver and the reparative processes in the liver.

(2) Proneness to Infections

Reports on this subject are limited to several studies made in the 1913-1917 period. Winternitz and Hirschfelder [72] reported that rabbits with leukopenia from benzene had strikingly reduced resistance to pneumonia. Kline and Winternitz [73] emphasized the lowering of resistance that accompanies leukopenia. Animals with leukopenia from benzene

succumbed in 41 hours to 5 ml of a 20-hour culture of pneumococcus introduced intratracheally, while animals injected subcutaneously with 1 ml of toluene/kg of body weight did not have leukopenia, had normal resistance to infection, and survived. Weiskotten and Steensland [74] noted that active acute infection appeared spontaneously in rabbits injected with benzene. The authors suggested that the lowering of resistance by benzene may activate latent or quiescent infection. White and Gammon [75] reported that rabbits exposed to benzene vapor administered from a wide-mouthed bottle with absorbent cotton on the bottom were less resistant to tuberculous infection than were unexposed rabbits. Camp and Baumgartner [76] found that rabbits whose leukocyte counts had been lowered to below 1,000/cu mm succumbed in 1 1/2 to 4 days from inflammatory reactions resulting from irritation of an ear scratch with croton oil or from an intramuscular injection of carmine.

(c) Metabolism

Phenol is the chief metabolite of benzene in the urine and, to a lesser extent, hydroquinone and catechol have been found [77]; yet, although the toxic effects of benzene have been attributed to its phenolic metabolites, [23, 78] subcutaneous administration of phenol, catechol, and hydroquinone in rats failed to produce any hematopoietic toxicity even at doses approximating an LD50. [79] Posner et al [80] in 1961 demonstrated a microsomal enzyme that metabolized benzene and it has been concluded from studies in rats that metabolism by hepatic microsomal enzymes is necessary for the observed bone marrow toxicity. [79] Also, benzene itself, rather than its hydroxylated derivatives, is probably responsible for the microsomal stimulation. [81] Drew and Fouts [82] in 1974 demonstrated that

pretreatment of rats with phenobarbital increased the rate of hepatic microsomal metabolism of benzene 10-fold. On the basis that the LC50 for inhaled benzene and the LD50 for injected benzene were not affected by pretreatment of rats with phenobarbital, a protective effect from barbiturateinduced microsomal metabolism was not demonstrated. Lee et al [83] in 1974 presented a study which was undertaken to determine which stage in erythrocyte development was most sensitive to benzene in the belief that benzene interfered with erythrocyte production. Based upon the 24- or 72-hour uptake of 59Fe in the circulating erythrocytes of mice having benzene pretreatment at selected time intervals, the possible damage to stem cells, pronormoblasts, normoblasts, or reticulocytes was determined. Using the appearance of 59Fe as an index of red cell development, it was determined that single doses of benzene selectively damaged pronormoblasts and normoblasts without affecting stem cells or reticulocytes. Thus, benzene seemed to damage red cell percursors which underwent both differentiation and maturation, rather than those concerned principally with differentiation (stem cells) or maturation (reticulocytes).

Correlation of Exposure and Effect

Liquid benzene on the skin may cause erythema and blistering, and a dry, scaly dermatitis may develop on prolonged or repeated exposure. [24] Investigations of the percutaneous absorption of benzene have failed to detect changes in the urinary inorganic to total sulfate ratio [28] or of benzene in the expired breath. [29] According to Hanke et al, [30] the rate of benzene absorption through the human skin was found to be 0.4 mg/sq

cm/hr as compared with later findings by others of 14-23 mg/sq cm/hr for toluene. [32] These findings, along with supporting evidence determined in rabbits by Wolf et al [68] indicate that liquid benzene is poorly absorbed through the intact skin. Therefore, skin absorption of benzene is not considered an important route of entry in the occupational situation; however, it is important to avoid skin contact with benzene to prevent local effects. Similarly, ingestion of liquid benzene is generally of concern only in cases of accidental swallowing or attempted suicide.

Absorption of benzene through inhalation is by far the most important route of entry in industrial exposures. Repeated exposures of workers over a prolonged time to high concentrations of benzene have occurred under conditions of poor ventilation combined with heated benzene to accelerate evaporation. [38] In a plant which manufactured rubber raincoats, an atmospheric benzene concentration was estimated to be 5,320 ppm. Sixty workers out of 184 suffered from chronic benzene poisoning in the reported study. [38] Rats exposed experimentally to 1,000 ppm of benzene [67] by Nau et al showed hemorrhaging from the nose and mouth, engorgement of the blood vessels, stomach distention, and markedly reduced WBC levels. Animals were exposed (23.5 hours/day, 7 days/week) for 183 hours to the benzene vapor. Such an exposure is not representative of the normal work schedule but it does indicate the toxic effects of benzene at a concentration to which humans are known to have been exposed.

Worker exposures to benzene concentrations from approximately 300-700 ppm consistently show marked blood dyscrasias. [19,39,41] Reduced WBC and RBC counts were noted by Greenburg [19]; Savilahti [39] found thrombocytopenia, leukopenia, and anemia in from 32-62% of 147 workers; and

Vigliani and Saita [41] described 2 cases of benzene-associated leukemia along with other workers who showed signs of benzene poisoning in a rotogravure plant which used inks containing 40% benzene. Deichmann et al [66] found significant leukopenia in rats exposed to 831 ppm of benzene vapor for periods of 5 weeks and longer. In another animal study, Nau et al [67] showed decreased WBC's and altered myelocytic and erythrocytic activity of the bone marrow in rats exposed to 200 ppm, 8 hours/day, 5 days/week, for 750 hours of exposure.

Alterations in the blood picture also have occurred at benzene exposure levels in the vicinity of 100 ppm. Juzwiak, [40] in 1969, stated the results of blood examinations on 585 persons employed in 13 shoe plants. Exposure levels from benzene contained in a glue mixture fluctuated in mean concentrations from 31-156 ppm. The author found reduced RBC counts, WBC counts, and Hgb levels. Although 91% of the workers had reduced Hgb levels, only 8.5% had reduced WBC counts. It is difficult to correlate the medical findings with the airborne exposures because of the lack of adequately documented environmental data.

Although Greenburg's findings [19] were published in 1926, they represent some of the most meaningful studies available showing approximate correlations of environmental benzene concentrations and chronic benzene poisoning (see Table XII-3). Greenburg considered cases as positive which showed less than 5,500 WBC/cu mm. In all groups studied, there were workers who presented a picture of chronic benzene poisoning as judged by reductions in the WBC count. Greenburg concluded that keeping the average concentration of benzene in the workroom air below 100 ppm (presumably just below this level) still involved a substantial hazard to workers. Effects

at 80-88 ppm were noted in animals by Wolf et al [68] in 1956 in rats, rabbits, and guinea pigs exposed intermittently to benzene vapor for periods varying from 204 to 269 days. On the basis of results from their study, the no-effect level was concluded by the authors to be below 80 ppm.

Hardy and Elkins [57] found evidence of deviations in more than one blood element in 16 of 52 workers from blood studies in a plant using naphtha solvents. For 8 years preceding their blood study, the plant's operational procedures and ventilation had undergone no significant changes. Six of the men with abnormal blood pictures were exposed to average benzene concentrations of not over 60 ppm and two other men were exposed to possibly 80 ppm. Additional data from further studies of the rubber coating industry from 1960 through 1963 were supplied by Pagnotto in a 1972 written communication. Environmental benzene concentrations consistently averaged between 20 and 25 ppm and frequently were lower for spreader and churn operations. From minimal blood data, 6 of 27 workers tested were found to have Hgb levels below 13.5 g/100 ml of blood in one of the 1961 studies (see Table XII-11). In the 1963 study, 12 of 24 workers were said to show minor deviations from normal although these differences were unspecified. Rats exposed by Deichmann et al [66] at mean concentrations of 44 ppm (range 40-50), 7 hours/day, 5 days/week, had WBC levels decreasing from 15.2 (preexposure) to 10.0 thousand/cu mm (after 5-8 weeks) in the males and from 11.8 (preexposure) to 7.7 thousand/cu mm in the females. No leukopenia was observed in rats exposed to average concentrations of 15 or 31 ppm.