III. BIOLOGIC EFFECTS OF EXPOSURE

Extent of Exposure

Benzoyl peroxide, $(C_6H_5CO)_2O_2$, also called dibenzoyl peroxide, is a rhombic crystalline solid at room temperature [1,2]. Benzoyl peroxide is a flammable, solid, diacyl organic peroxide, which may decompose explosively if subjected to excessive heat, friction, or sudden shock [3-5]. If benzoyl peroxide is exposed to temperatures of 75-80 C for prolonged periods, it becomes unstable and may spontaneously decompose [4]. This type of sudden decomposition, a deflagration, is the rapid spreading of fire through a mass of reactive material [6]. This decomposition is accompanied by a 200-fold increase in volume [5] and yields a dense white smoke consisting of benzoic acid, phenyl benzoate, terphenyls, biphenyls, benzene, and carbon dioxide [7]. The resulting biphenyls promote the further decomposition of benzoyl peroxide [5,7] into products which can catch fire and ignite the remaining benzoyl peroxide. If this happens, or if the benzoyl peroxide itself ignites, a dense black smoke results [8].

The peroxide reacts violently with various organic and inorganic acids, amines, alcohols, metallic naphthanates, and other chemicals that are easily oxidized. Benzoyl peroxide also reacts violently with polymerization accelerators [4].

The presence of small quantities of water diminishes some of the hazardous properties of benzoyl peroxide [9]. During a series of tests on the ease of ignition of pure benzoyl peroxide and benzoyl peroxide with various proportions of water, pure benzoyl peroxide was shown to ignite violently with a loud noise, but benzoyl peroxide containing 5% water did

not ignite at all. It was also observed that during this ignition test [9] it did not make any difference whether the total moisture content of the sample was equally divided between each granule or concentrated in 10-20% of the granules, as long as those granules were uniformly dispersed throughout the sample. Additional chemical and physical properties of benzoyl peroxide are presented in Table XIV-1 [1,2].

Benzoyl peroxide is synthesized commercially by a reaction of benzoyl chloride, sodium hydroxide, and hydrogen peroxide [10 (pp 14,85,187),11]. Excess water is removed to obtain pure benzoyl peroxide; the trace impurities remaining are benzoic acid and water. Water, plasticizers, corn starch, or other diluents are added to make the numerous commercial products containing benzoyl peroxide. Benzoyl peroxide has been produced commercially in the United States since 1927 [12]. By 1954, its yearly production was 1,768,000 pounds [13]; 8,829,000 pounds, 9,092,000 pounds, and 7,885,000 pounds were produced in 1973 [14], 1974 [15], and 1975 [16], respectively.

Since benzoyl peroxide is a good source of free radicals, it is used in a number of industrial processes, particularly in the manufacture of plastics [5]. Benzoyl peroxide is a curing agent for silicone rubber [17], a source of free radicals in the resin cements used in dentistry [18], automobile body putty [10 (p 283),19], and roof bolting systems in the mining industry [20], and an initiator in the synthesis of polyvinyl chloride [3]. It is also a component of flour and cheese bleaches [21,22]. In the early 1900's, benzoyl peroxide was used to bleach edible oils, but this practice is now rare [10 (p 276)]. In the past, textiles and paper were also treated with it [11]. In medicine, it now is used in the

treatment of acne [23] and of decubitus ulcers (bed sores) [24]. Formerly, it was applied as an aid in the treatment of poison ivy [25].

NIOSH estimates that 25,000 workers in the United States are potentially exposed to benzoyl peroxide or its formulations. Occupations involving possible exposure to benzoyl peroxide are listed in Table XIV-2.

Historical Reports

Little was known about benzoyl peroxide until the end of the 19th century. In the Encyclopedia of Chemical Technology, Hooft [11] noted that Brodie synthesized benzoyl peroxide in 1858. One of the earliest references to benzoyl peroxide appeared in 1899 when Nencki and Zaleski [26] reported that it was converted to benzoic acid in the intestines of dogs. As early as 1921, benzoyl peroxide was used in Germany as a fixing agent in light microscopy [27]. It was also used at that time as an antiseptic and local anesthetic in the treatment of burns and ulcers, as reported by Farmer [27]. Benzoyl peroxide had previously been taken internally, but that practice was discontinued because of its poisonous action on the blood, which was not specifically described. However, in 1964, Tiunov [28] noted that Smirnova, using unspecified chemical methods, found that benzoyl peroxide had almost no hemolytic action.

In 1930, Lamson [25] stated that powdered benzoyl peroxide was a theoretically ideal treatment for skin lesions caused by poison ivy because it reduced the spread of the rash and relieved itching. The flammability hazard of benzoyl peroxide treatment was not mentioned in the literature until 1931 when it was reported that a man whose poison ivy rash was being treated with benzoyl peroxide was injured by the ignition of bandages that

were covering the powdered benzoyl peroxide on his hands [29,30]. When the bandages were ignited by a lighted cigarette, the benzoyl peroxide exploded, and the skin and several muscles of his right hand were destroyed. The author [29] retracted his recommendation of powdered benzoyl peroxide as a useful therapeutic agent, emphasizing its explosive properties; he recommended, instead, an ointment of an unspecified concentration of benzoyl peroxide in lubricating jelly, which he considered neither explosive nor extremely flammable. No references have been found indicating further use of benzoyl peroxide for the treatment of poison ivy.

Effects on Humans

The effects of occupational exposure to and treatment with benzoyl peroxide have been examined. Inhalation and skin contact are the most frequent routes of exposure.

In 1950, Moskowitz and Burke [31] described the inspection of a factory that used benzoyl peroxide in the production of flour bleach. The powdered bleach contained 32% benzoyl peroxide; the remaining 68% consisted of unspecified proportions of potassium aluminum sulfate (alum) and magnesium carbonate. Over a 3-day period, standard (Greenburg-Smith) impingers containing water collected nine air samples at six different work areas in the factory. The sampling was performed for 20 minutes, 2 to 3 times/shift, on all 3 days. The water from the impingers was analyzed by unspecified methods for benzoyl peroxide and alum. No analyses for magnesium carbonate were performed. Two air samples were taken near grinders on the 1st day. One reportedly contained 1.34 mg of benzoyl peroxide and 2.58 mg of alum/cu m, and the other had 5.17 mg of benzoyl

peroxide and 5.33 mg of alum/cu m. Nose and throat irritation were experienced by the two inspectors who were taking the air samples.

On the 2nd day, another air sample taken during a bag-changing operation at one of the grinders contained 2.91 mg of benzoyl peroxide and 3.12 mg of alum/cu m [31]. Another air sample taken on the 2nd day, near a worker emptying a tumbling barrel from one of the grinders, contained 17.0 mg of benzoyl peroxide and 18.8 mg of alum/cu m. In the same location 16 minutes after the barrel had been emptied, the concentrations of benzoyl peroxide and alum were 1.45 mg/cu m and 1.96 mg/cu m, respectively. A fourth sample taken during a bag-changing operation at a grinder contained 5.25 mg of benzoyl peroxide and 5.4 mg of alum/cu m. Again the inspectors had symptoms of nose and throat irritation. The factory workers wore cotton-pad dust respirators during potentially dusty operations and did not complain of nose or throat irritation.

On the 3rd day, during the filling of 100-lb fiber drums at a tumbling barrel, an air sample contained 12.2 mg of benzoyl peroxide and 8.26 mg of alum/cu m [31]. A second air sample taken near a worker emptying a tumbling barrel contained 82.5 mg of benzoyl peroxide and 44.9 mg alum/cu m. The inspectors reported nose and throat irritation at these higher concentrations of airborne dust. A third sample was taken near the grinders 6 minutes after the tumbling barrel had been emptied; also, the floor was being swept near the impinger during the sampling operation, and this action may have increased the airborne dust concentrations. The concentrations in this sample were 2.58 mg of benzoyl peroxide and 3.05 mg of alum/cu m. Nose, eye, and throat irritation occurred duri g the changing of bags and the emptying and filling operations at the grinders

when the concentrations of airborne benzoyl peroxide were between 2.58 mg/cu m and 82.5 mg/cu m. These dust levels could have caused some of the irritation experienced by the inspectors. There was natural ventilation in the plant from open windows, especially when there were strong winds. On days when no wind blew through the working areas, the dustiness increased, and the workers experienced eye irritation. Alum has astringent properties and could have possibly caused the irritation.

The authors [31] made no specific conclusions about the possible irritating effects of benzoyl peroxide. They indicated that the airborne dust was irritating on all 3 days when it contained benzoyl peroxide at concentrations between 1.34 and 82.5 mg/cu m. The reported concentrations of airborne benzoyl peroxide and alum are questionable because no data were given which defined the efficiency of standard impingers containing water for collecting benzoyl peroxide and alum, and there was insufficient analytical information to assess the reliability of the determinations. In addition, it was noted that the proportions of benzoyl peroxide to alum were extremely variable and did not reflect the proportion of the two chemicals in the flour bleach being processed.

In 1945, Baird [32] reported that a young male baker suffered from asthmatic wheezing and severe dermatitis of the face, neck, chest, shoulders, and arms. Although the author was not certain whether these symptoms were caused by occupational skin contact or ingestion of benzoyl peroxide-treated flour, when wheat flour was removed from the baker's diet, he improved rapidly. When wheat flour without improving agents was reintroduced in his diet, he had no further allergic reactions. However, he later worked with treated wheat flour, and the dermatitis reappeared.

Patch tests with different kinds of flour performed on the baker gave positive results if the flours contained improving agents; areas tested with unimproved flours showed no reaction [32]. Patch tests performed on the baker with potassium bromate and benzoic acid in water gave no definitive reaction. A patch test of 6% benzoic acid in liquid petrolatum was positive; a control patch with petrolatum alone was negative.

Baird [32] concluded from information provided by the Canadian Department of Agriculture that the use of benzoyl peroxide in flour produced a benzoic acid residue of 18-45 ppm. Perhaps this is the reason that no patch tests were performed with benzoyl peroxide. However, in 1953, Knight and Kent-Jones [33] stated that, although most of the benzoyl peroxide used to bleach flour decomposes to benzoic acid within a few days, a small amount remains unchanged for several weeks.

Two years after being seen, the baker used benzoyl peroxide-treated flour again and promptly developed dermatitis [32]. Baird concluded that the baker's allergy was the result of benzoic acid, the residue remaining from benzoyl peroxide, but he did not develop data to rule out the role of some other chemical allergen in the diet.

In 1957, Malten [34] outlined a dermatologic study of aircraft factory workers in the Netherlands who suffered from occupational dermatitis. Patch tests were performed with many agents, including benzoyl peroxide, as test materials. Three of 30 polyester processors had hypersensitive skin responses to the benzoyl peroxide patch tests. The total number of workers or the percentage of workers having occupational dermatitis was not specified. Malten stated that no new cases of dermatitis were reported after improved ventilation and changes in work

practices went into effect.

In 1960, Jirasek and Kalensky [35] evaluated 34 of an unspecified number of workers in Czechoslovakia who had been exposed to various epoxy resins and had experienced some degree of irritation from at least one of the epoxy resins or the materials used to make the resins. Benzoyl peroxide was used as a hardener and was one of the compounds tested on the workers. Eight of the 34 showed an unspecified toxic reaction to benzoyl peroxide. The authors also observed that patients with sensitive skin showed signs of slight irritation when tested with benzoyl peroxide at concentrations of 20-100% in an unspecified solvent. Work histories of the patients were not provided.

Morley [24] treated 180 patients who had decubitus ulcers with repeated applications of what was described as a water-in-oil emulsion containing 20% benzoyl peroxide. The benzoyl peroxide-treated dressing was applied to the ulcer, covered with a sheet of plastic, and held in place with an elastic net or body stocking. The dressing was changed every 12 hours. This treatment was continued until the ulcer was healed. Treatment was discontinued in one patient because of irritation. It was necessary to surgically repair only one of the treated ulcers.

A number of cases of skin reactions to benzoyl peroxide-containing formulations used in the treatment of acne have been described [36-38]. In 1968, Eaglstein [37] described two patients with allergic dermatitis from benzoyl peroxide. One, a 15-year-old girl, who had previously used a topical antiacne preparation containing benzoyl peroxide and had experienced severe skin irritation, redness, and edema, tried another ointment, which contained 5% benzoyl peroxide and 2% sulfur. It produced

severe edema, redness, and a burning sensation in about 12 hours. She had positive patch-test reactions to all tested preparations containing benzoyl peroxide; the preparations were not described.

The other patient, a 21-year-old woman, was treated for superficial acne lesions with a lotion containing 5% benzoyl peroxide; the other ingredients were not specified [37]. After the second overnight facial application, the patient noted marked erythema and a burning sensation on the face. Patch tests with 5% benzoyl peroxide in petrolatum were positive.

To evaluate the meaning of these positive reactions, Eaglstein [37] conducted patch tests with 5% benzoyl peroxide in petrolatum, with petrolatum alone, and with untreated control patches on 41 patients hospitalized for various skin conditions. After 48 hours, only one patient, who had not used benzoyl peroxide previously, had a positive reaction to benzoyl peroxide. It is unlikely that responses in this control group made up of patients with dermatologic conditions would be representative of the general population.

In 1970, Poole et al [36] conducted a three-part study of experimental contact sensitization with benzoyl peroxide. In the first test, 10 volunteers underwent patch tests for irritation from benzoyl peroxide at 3 concentrations. Each was given single applications of an unspecified amount of polyethylene glycol containing 1% sulfur and 1%, 5%, or 10% benzoyl peroxide on separate sites on the arms. Because it had sufficiently low potential for causing irritation, the ointment containing 10% benzoyl peroxide and 1% sulfur was selected for a large-scale repeated-insult patch test. In a second test, each of 69 volunteers received on 1

arm, during a 3-week period, nine 24-hour applications of 0.25 g of polyethylene glycol containing 10% benzoyl peroxide and 1% sulfur. They were tested simultaneously with polyethylene glycol alone.

Two of the 69 subjects had minor reactions to the test materials during the first 24-hour treatment period [36]. By the 3rd week of the testing period, an unspecified number of subjects had positive reactions to the test materials but not to the control vehicle, polyethylene glycol. If there was a positive reaction at the test site and benzoyl peroxide was applied once to another site on the same person, it also showed a positive reaction, demonstrating general skin sensitivity. At the end of the 3-week period, 25 of 69 subjects showed severe, eczematous skin reactions when challenged with the test material. Another six subjects had responses stronger than those seen on the single induction exposures but which were not classified as sensitization.

The third part of the study occurred 2 months after the conclusion of the repeated patch tests when 10 subjects who had shown moderate sensitivity to the benzoyl peroxide and sulfur test material were tested with a single 24-hour patch test of each of the following: (1) polyethylene glycol, (2) polyethylene glycol containing 1% sulfur, (3) polyethylene glycol containing 10% benzoyl peroxide, and (4) polyethylene glycol containing 1% sulfur and 10% benzoyl peroxide [36]. All the subjects reacted to the benzoyl peroxide whether or not sulfur was present, but none reacted to the polyethylene glycol or sulfur.

In 1973, Ede [38] discussed a double-blind study of 196 acne patients who were randomly divided into 4 groups. Three acne lotions and a placebo were tested. The lotions contained 5.5% benzoyl peroxide, 0.25%

chlorohydroxyquinoline, and 0.5% hydrocortisone; 5.5% benzoyl peroxide and 0.25% chlorohydroxyquinoline; or 5.5% benzoyl peroxide. The placebo contained only the base lotion. The lotion was applied to affected areas 1 to 4 times daily for 4 weeks; however, the mean number of applications/day for the groups ranged from 2.2 to 2.5. The lotion was left on the skin for at least 3-4 hours. None of the patients exhibited any skin sensitivity to the lotions containing benzoyl peroxide at the end of the 4 weeks; however, 10 patients dropped out of the study for unspecified reasons.

The following laboratory tests were performed during the study [38] on the blood and urine of 20 of the patients, 10 men and 10 women, to determine whether there were any systemic effects of the lotions: calcium, inorganic phosphorus, glucose, blood urea nitrogen, uric acid, cholesterol, total protein, albumin, and total bilirubin concentrations; activities of alkaline phosphatase, lactate dehydrogenase, and serum glutamic-oxaloacetic transaminase; complete blood count (hemoglobin, hematocrit, RBC, WBC with differential count) and urinalysis (specific gravity, pH, color, appearance, sugar, microscopic examination, albumin, and acetone). The results were within the normal ranges and indicated no systemic effects from any of the lotions.

Bloom [19], in 1975, reported that welders employed in the manufacture of diesel locomotives were exposed to a plastic body filler made of a talc-polyester resin and benzoyl peroxide. Two of four welders who were interviewed thought that the coughing they experienced during the day was caused by exposure to welding fumes and to plastic body filler dust. There was no evidence of skin irritation or sensitization.

A NIOSH Health Hazard Evaluation Determination discussed by Kingsley [39], indicated that telephone repair workers were exposed to a styrene hardener containing 50% benzoyl peroxide and 50% butyl benzoyl phthalate when new and replacement telephone cables were installed. A worker who was wearing disposable gloves would add the hardener to the polyester, manually knead the mass until it was the right consistency, and drop it down into a vault where another gloved worker would shape the compound around the splice. Each such operation required two or three tubes of hardener and took about 30 minutes. One crew normally coated splices once or twice a week. The vaults were naturally ventilated through the manhole covers. The workers did not report adverse effects from using the compound.

Accidents

Hazardous properties of benzoyl peroxide, such as explosion and flammability, have resulted in accidents and serious injuries or death. The following incidents demonstrate that injuries were usually caused by ignorance of the hazards or by negligent handling. Other accidents that did not produce injury are discussed in Chapter V.

Twelve pounds of pure benzoyl peroxide being added through a stainless steel funnel into a polymerization kettle exploded, killing the operator [3]. There were three possible reasons for the explosion: (1) the funnel may have become heated during the operation, so that excessive heat may have caused the peroxide to explode; (2) the peroxide may have become contaminated with residual vinyl acetate from the polymerization reaction; or (3) a static discharge may have occurred.

In another case, an employee escaped serious injury when a flash fire

erupted in a 1-pound container of benzoyl peroxide and covered his safety glasses with melted benzoyl peroxide [3]. He was using a glass spatula to transfer benzoyl peroxide from the container to a laboratory scale [3]. As the spatula, which had just been cleaned and dried, was inserted into the container, the benzoyl peroxide burst into flame. The account of the accident indicated that contamination of the benzoyl peroxide may have caused the fire. It is also possible that the friction from the insertion of the spatula may have started it.

In still another case, the owner of a plant that manufactured benzoyl peroxide sustained second degree burns from a fire started by an unknown quantity of benzoyl peroxide dust exposed to an arcing electric light switch [3]. The fire generated smoke and chemical fumes; eventually, there was an explosion.

Lappin [40] found that a laboratory worker received hand injuries and lacerations when benzoyl peroxide in a 4-ounce, brown-glass container exploded as the plastic screwcap was being removed. The author thought that some benzoyl peroxide, along with other organic dust present in the laboratory, was caught in the threads and, as the cap was unscrewed, the friction caused the top layer of peroxide in the bottle to explode.

The explosiveness of benzoyl peroxide was further illustrated when several thousand pounds of the compound exploded in a truck, causing severe property damage within a radius of several city blocks and injuring four people, one seriously [41]. A fire was seen seconds before the explosion occurred, but the exact cause of the accident was unknown. Investigators speculated that perhaps other chemicals had come in contact with the cargo of benzoyl peroxide or that an all-day exposure to hot sun had caused

drying of the benzoyl peroxide. Another possibility was that the truck might have been bumped, dislodging the cargo.

Animal Toxicity

There are few data on the effects of benzoyl peroxide on animals. The effects of inhalation, ingestion, skin painting, and injection of benzoyl peroxide have been examined.

Two eye irritation tests with granular 78% benzoyl peroxide were conducted on eight albino rabbits by Wazeter and Goldenthal [42]. Though not specified in the report, 78% benzoyl peroxide granules commonly consist of 22% water and benzoic acid. Sodium fluorescein was put into the eyes when they were examined under ultraviolet light so that corneal damage could be detected. The eyes were examined before treatment with benzoyl peroxide and periodically afterwards. In the one test, 111.4 mg of 78% benzoyl peroxide (0.1 ml measured by volume) was put in the cupped conjunctival sac of the right eye of each of five rabbits; the eyelid was held shut for 1 second. The left eyes served as controls. After 5 minutes, the test eyes were washed with a gentle stream of water, regulated to deliver 300 ml in 2 minutes.

The corneas showed no ulceration or opacity after 1, 24, 48, or 72 hours or after 7 days [42]. The irises appeared unaffected. The conjunctivae of two rabbits showed slight redness 1 hour and 24 hours after the washing, but this disappeared in 48 hours. Three of five rabbits exhibited conjunctival edema 1 hour after the washing, but this was not apparent at 24 hours. The authors concluded that, under these test

conditions, benzoyl peroxide was not irritating or corrosive to the eyes.

In another eye irritation test [42], 120.7 mg of 78% benzoyl peroxide was placed in the cupped conjunctival sac of the right eye of each of three rabbits where it remained for 24 hours; the left eyes were controls. After 24 hours, the benzoyl peroxide was washed out with 300 ml of water for 2 minutes. The eyes were examined under ultraviolet light as described in the first test. The irises appeared normal after 1, 24, 48, and 72 hours and after 7 days. The conjunctivae of the rabbits exhibited various degrees of redness and conjunctival edema at 1, 24, 48, and 72 hours, but all adverse effects disappeared in 7 days. One rabbit had blanched conjunctival tissue at 1 hour, but normal color had returned within 24 hours. Examinations under ultraviolet light showed corneal opacity in the three rabbits after 24 hours but no corneal opacities at 48 hours. The only corneal damage in this experiment was revealed in one rabbit by the eye examinations done at 72 hours, and it had disappeared by the 7th day.

Wazeter and Goldenthal [42] concluded that benzoyl peroxide was neither irritating nor corrosive to the eyes of albino rabbits if it was washed out within 5 minutes after being placed in the conjunctival sac; however, if 78% benzoyl peroxide was not washed out until 24 hours later, it proved to be a strongly irritating substance. It was not considered corrosive because corneal opacity lasted less than 6 days.

In a third experiment, Wazeter and Goldenthal [42] tested the skin irritation potential of benzoyl peroxide on three male and three female New Zealand white rabbits. No control animals were mentioned. The hair was shaved from an area on the back of each rabbit, and the skin was then abraded with a scalpel blade. Five hundred milligrams of 78% benzoyl

peroxide was applied to each patch of skin and held in place for 4 hours with a gauze bandage. After 4 hours, the bandages were removed and the exposed areas washed with lukewarm water. The skin was examined for any injury or irritation from benzoyl peroxide at 4, 24, and 72 hours. The skin on the six rabbits appeared unaffected. The authors concluded that 78% benzoyl peroxide was neither a primary skin irritant nor a corrosive substance.

Wazeter and Goldenthal [42] also performed a short-term inhalation study on 10 male Spartan rats housed in groups of 2 or 3. The rats were exposed at an atmospheric concentration of 24.3 mg/liter of 78% benzoyl peroxide added to a 59.1-liter glass test chamber supplied by two Wright dust feeders with a regulated airflow.

None of the rats died during the test or the subsequent 14-day observation period [42]. The rats showed the following signs during the 4-hour exposure period: eye squint, increased and decreased respiratory rates, difficulty in breathing, salivation, lacrimation, erythema (location unspecified), and an increase followed by a decrease in motor activity. All of the rats appeared normal at 24 and 48 hours. An unspecified number of rats exhibited signs of eye irritation consisting of corneal opacity and ulceration from the 5th to the 14th day. The authors concluded that 78% benzoyl peroxide was not highly toxic by the inhalation route of administration under the conditions of the experiment.

A short-term oral toxicity test was performed by Wazeter and Goldenthal [42] with 78% benzoyl peroxide in water on five male Spartan albino rats. Each rat received one 5,000 mg/kg dose of 78% benzoyl peroxide suspended in corn oil. The rats took food and water ad libitum

and were maintained in temperature- and humidity-controlled quarters during the 14-day study. No control animals were reported. Body weights of all the rats were recorded initially and at 14 days. None of the rats died during the study, and all exhibited normal weight gain. Under the test conditions, 78% benzoyl peroxide was not toxic by the oral route of administration.

In 1958, Kuchle [43] described an experiment in which 15 organic peroxides, including benzoyl peroxide, were tested for their effects on rabbits' eyes. A "lentil-sized" amount of an undefined paste containing 50% benzoyl peroxide was placed in the conjunctival sacs of each of several rabbits, and unspecified amounts of a 93% benzoyl peroxide powder were placed in the conjunctival sacs of several other rabbits. No controls were mentioned. After 1 minute, the eyes were rinsed with tapwater, and any solid residues were removed with a cotton swab. The eyes were then examined after 20 minutes, after 24 hours, then every other day for 1 week, and finally twice a week for 6 weeks. Neither form of benzoyl peroxide was considered to have had harmful effects on the rabbits' eyes; no evidence of burning or irritation was observed, and the corneas of the test animals were clear and had no opacities.

Radomski et al [44] published, in 1948, a study in which three dogs were given a diet containing benzoyl peroxide-treated flour for 6 weeks. The purpose of the experiment was to determine the toxicity of candidate replacements, including benzoyl peroxide, for agene, an improving agent used to treat flour, which consisted of 1% nitrogen trichloride in air saturated with water vapor. Benzoyl peroxide was added to the flour (1 oz benzoyl peroxide/100 pounds flour or 0.625 g/kg). A short time before it

was fed to the dogs, the mixture was steamed for 90 minutes, and nutrients were added to it. The nutritionally balanced diet contained 71.6% treated flour on a dry-weight basis. Because the authors did not state the amount of food consumed by each dog, the actual intake of benzoyl peroxide is unknown. The effects of steaming on benzoyl peroxide were not considered.

The authors [44] stated that, since the 1920's, canine hysteria, sometimes called running fits, had been observed in dogs that had eaten agene-treated flour. No canine hysteria was observed in the dogs given the diet containing benzoyl peroxide, and, unlike dogs fed agene-treated flour, they behaved in a normal manner.

In 1949, Arnold [45] described a study in which dogs were provided with a diet in which flour had been treated with 0.8 g of benzoyl peroxide/100 pounds of flour (0.02 g/kg). Chlorine at 20 g/100 pounds (0.44 g/kg), ammonium persulfate at 15 g/100 pounds (0.33 g/kg), and potassium bromate at 5 g/100 pounds (0.11 g/kg) were also used to treat the flour; the amounts were greater than those used commercially in flour bleaching. The diet contained about 80% treated flour on a dry-weight basis. This diet and other experimental diets were given intermittently to six dogs for periods ranging from 21 to 38 days with intervening times of 3-16 days. The dogs were observed for canine hysteria, but it was not seen in those dogs fed benzoyl peroxide-treated flour.

One group of investigators [46] attempted to determine the oral LD50 of benzoyl peroxide in rats. Groups of two fasted rats each were given oral doses of benzoyl peroxide placed on a small amount of pea soup concentrate at 200, 400, and 950 mg/kg. None died. One of the rats that received 400 mg/kg had some vasodilatation, and one that received 950 mg/kg

showed slight muscular weakness. The investigators concluded that the oral LD50 of benzoyl peroxide in rats is greater than 950 mg/kg.

Skin irritation by benzoyl peroxide in an unspecified number of guinea pigs was also tested [46]. Patches of skin were chemically depilated, and pure benzoyl peroxide, in doses ranging from 0.25 to 1.0 g/kg, was held against the depilated skin under patches for 24 hours. The skin under the benzoyl peroxide was examined for any irritation or other injury. Slight erythema with some delayed scarring of the epidermis resulted. There were no deaths. A similar test was run on guinea pigs with a 10% solution of benzoyl peroxide in propylene glycol. The doses ranged from 5 to 20 ml/kg. Only slight erythema was observed; no deaths occurred.

An inhalation test also described in this study [46] showed that an unspecified number of rats had no observable ill effects after being exposed to airborne benzoyl peroxide at an unspecified concentration for 3 hours.

In 1957, Horgan et al [47] gave 12- to 14-week-old female R and CBA hybrid hairless albino mice intraperitoneal (ip) injections of benzoyl peroxide. The injections consisted of 0.1-0.4 ml of unspecified concentrations of benzoyl peroxide in ethyl palmitate. The LD50 was reported to be 20 μmoles (4.8 mg)/mouse.

In 1959, Philpot and Roodyn [48] found the LD50 of benzoyl peroxide in 13- to 14-week-old female R hybrid mice to be 17.1 μmoles (4.1 mg)/mouse or 167 mg/kg.

In 1964, Sharratt et al [49] reported the results of a series of tests to determine the effects of benzoyl peroxide incorporated in the diet or administered by subcutaneous injection or by skin painting on rats and mice. Each test lasted 120 weeks for rats and 80 weeks for mice; moribund animals were killed during the study. The age and weight of the animals at the start of the experiment were not reported.

Three experimental groups, each composed of 25 male and 25 female rats and 25 male and 25 female mice, were given nutritionally balanced diets of wholemeal flour that was treated with a commercial flour bleach consisting of 18% benzoyl peroxide, 78% calcium sulfate, and 4% magnesium carbonate [49]. The control group contained the same number of animals as the experimental group but received untreated flour in their diet. The resulting benzoyl peroxide concentrations in the diet were 2,800 ppm, 280 ppm, and 28 ppm. These concentrations were selected because they were estimated to be 1,000, 100, and 10 times the normal human intake based on a yearly consumption of 200 pounds of flour/person. How much the animals actually ate was not reported, so exact dosages cannot be determined. Weight gains were recorded only for the rats during the first 16 months of the test.

The rats whose diets contained flour treated with 2,800 ppm and 280 ppm benzoyl peroxide gained weight at a slower rate than the controls; the authors reported that this effect was not seen when the rats were caged singly in a diet preference test and an individual caging test [49]. Seventeen mice that received the 280-ppm diet were killed accidentally, and a large number of rats and mice in the entire colony showed signs of infection, the nature of which was not specified by the investigators. For

these reasons, the statistical significance of the results cannot be accurately evaluated.

A diet preference test and an individual caging test were conducted with 10 pairs of male rat littermates to determine if any differences in weight gain in the animals were the result of greater food intake with 1 of the diets [49]. One of each pair of the male littermates was given a flour-based diet containing benzoyl peroxide at 2,800 ppm, and the other was given the same diet without any benzoyl peroxide. Each rat was caged singly.

The weight gain for the two groups was reported to be similar [49]. After 30 weeks, each of the control rats had gained an average of 355 g and had consumed an average of 4,870 g of the supplied diet; the experimental group had gained 350 g each and eaten 4,902 g of the supplied diet. Rats caged singly tended to increase food intake slightly. On the basis of the diet preference test and the caging test, they concluded that concentrations of 1,000 and 100 times the normal human daily intake of benzoyl peroxide in the diets may have reduced the nutritional value of the diet; whereas the diet containing 10 times the normal daily intake of benzoyl peroxide did not.

Sharratt et al [49] provided diets of breadcrumbs made from flour treated with benzoyl peroxide to two groups of animals. The breadcrumbs given to 100 male and 100 female mice and 100 male and 100 female rats were prepared from bread made with flour containing benzoyl peroxide at 28 ppm. A group of 25 male and 25 female mice and 25 male and 25 female rats received a breadcrumb diet in which the flour had contained 2.8 ppm benzoyl peroxide. A control group of 100 male and 100 female mice and 100 male and

100 female rats was given a breadcrumb-based diet made from flour containing no benzoyl peroxide. Weight gains were reported only for the rats during the first 16 months of the test.

There were no significant differences in the body weights of the rats given treated breadcrumbs made with treated flour and those of the controls except at 16 months, when the male rats that received the breadcrumbs made from flour containing 2.8 ppm of benzoyl peroxide weighed significantly more than the male control rats [49]. The authors considered this of doubtful importance, since all rats began to gain and lose weight erratically because of chronic infection in the colony.

In another part of the study [49], rats and mice were given a single subcutaneous injection of what was described as a freshly prepared 20% suspension of benzoyl peroxide in starch solution. The dose for 25 male and 25 female rats was 120 mg of benzoyl peroxide, and, for 25 male and 25 female mice, it was 50 mg of benzoyl peroxide. Control rats and mice, 25 of each sex of each species, were each given an injection of the starch solution. All the rats and mice were provided with a commercial pellet diet. Body weights were reported only for the rats for the first 16 months. There was no difference in the rate of weight gain in the rats administered benzoyl peroxide and in their controls. No tumors were found at the injection sites in any of the rats or mice; there was no significant difference in the tumor incidence in the experimental animals and in the controls.

Sharratt et al [49] also painted benzoyl peroxide on the back of the neck of 25 male and 25 female mice for 6 consecutive days. One drop (about 50 mg) of a freshly prepared 50% suspension of benzoyl peroxide in flour

paste was applied to each animal. A similar number of control mice were painted with only the flour paste. Both groups of mice were fed a commercial pellet diet. No tumors appeared at the sites of painting, and there was no significant difference in the overall tumor incidence between the experimental animals and the controls.

Sharratt et al [49] also administered a multiple treatment to groups of 25 male and 25 female rats and 25 male and 25 female mice. There were no control animals for this part of the experiment. The rats and mice received the flour-based diet containing 2,800 ppm benzoyl peroxide and subcutaneous injections of benzoyl peroxide as in the previously described tests. The mice were also painted with flour paste containing benzoyl peroxide in the manner described previously. Body weights were reported only for the rats for the first 16 months. Except for a slight decrease at the 8th month, the weight gain of the rats in this multiple treatment group was not significantly different from that of the controls in the other tests described previously. No tumors were found at the sites of injection or painting.

Sharratt et al [49] observed that the entire colony of mice and rats used in their experiments with benzoyl peroxide had many abnormal changes irrespective of the test performed on the animal. There was a statistically significant incidence of atrophy of the testicles in the rats given the diet based on flour treated with benzoyl peroxide at 2,800 ppm and in the rats receiving diets of breadcrumbs made with flour treated with benzoyl peroxide at 28 ppm and 2.8 ppm. The authors suggested that this atrophy was caused by benzoyl peroxide, which probably marginally decreased the amount of vitamin E in the diet. This conclusion was not supported by

any analyses of the diets, and the degree of testicular atrophy in each rat was not stated; therefore, no definitive conclusion can be made. While the authors [49] concluded that benzoyl peroxide was not carcinogenic in rats or in mice under the test conditions, it does not seem that this was a definite experiment of carcinogenicity or of other types of chronic toxicity. The length of the observation periods and the experimental design were probably adequate; however, there may have been insufficient numbers of animals to detect carcinogenicity. In addition, it is uncertain how much benzoyl peroxide remained unchanged after it was added to the diets.

Other investigators have studied the action of benzoyl peroxide in animals to ascertain whether it is carcinogenic. Hueper [50] conducted a study to determine if benzoyl peroxide, when used as a polymerization catalyst for silicone rubber, had carcinogenic properties. According to the manufacturer, benzoyl peroxide was totally destroyed in the rubber curing process. A piece of silicone rubber that had been cured with benzoyl peroxide was implanted subcutaneously in the neck of each of 21 male and 14 female Bethesda black rats. In another group of Bethesda black rats, a gelatin capsule containing 50 mg of benzoyl peroxide was implanted subcutaneously in the nape of the neck of 20 males and 15 females. No control animals were used. The rats were observed for 24 months.

In the rats with silicone rubber implants, 10 sarcomas occurred at the implantation sites, and there were neoplasms at other sites, viz, 4 round cell sarcomas of the ileocecal lymph nodes, 3 mesotheliomas of the peritoneum, and 1 carcinoma of the bladder [50]. There were no tumors at the implantation sites in the rats with the encapsulated benzoyl peroxide,

although seven of these rats had malignancies at other sites, including four round cell sarcomas of the ileocecal lymph nodes, one mesothelioma of the peritoneum, one epidermoid carcinoma of the snout, and one myxosarcoma of the anal region. Benign tumors, including two adenofibromas of the breast and one cystic cholangioma, appeared in three other rats in the group with the benzoyl peroxide implants. Hueper concluded that the absence of tumors at the sites of implantation provided conclusive evidence that benzoyl peroxide was not implicated in the induction of polymer cancers.

Van Duuren and his colleagues [51] studied the carcinogenicity of a group of epoxides, lactones, and peroxides including benzoyl peroxide. The backs of 30 male Swiss-Millerton mice were painted 3 times weekly with about 100 mg of a 5% benzene solution of benzoyl peroxide. Controls were similarly painted 3 times weekly with 100 mg of benzene alone. The median survival times were 292 days for the mice exposed to benzoyl peroxide and 264, 262, 412, and 292 days for the four control groups. The animals were examined regularly for tumors. None of the mice developed carcinomas; one mouse exposed to benzoyl peroxide developed a benign tumor. The authors concluded that benzoyl peroxide showed no carcinogenic activity in this experiment.

In 1972, Epstein et al [52] tested 174 agents, including benzoyl peroxide, for dominant lethal mutations in ICR/Ha Swiss mice. Benzoyl peroxide at doses of 54 and 62 mg/kg was administered by intraperitoneal (ip) injection to seven and nine male mice, respectively. Each animal was then caged with three untreated virgin female mice for 1 week. The females were replaced each week for a total of 8 weeks and then killed and examined

for pregnancy (total implants), early fetal deaths, and late fetal deaths. Since late fetal deaths were very rare, total implants and early fetal deaths were the only implant features analyzed.

The results obtained in the experimental mice were not significantly different from the results in the control mice [52]. Benzoyl peroxide, in the dose range and in the strain of mice used, met none of the screening criteria for these dominant lethal mutations. The authors recommended additional tests to confirm the apparent lack of mutagenicity of benzoyl peroxide.

An evaluation of the mutagenic properties of 78% benzoyl peroxide was reported in 1975 [53]. The yeast Saccharomyces cerevisiae, strain D4, and the bacterium, Salmonella typhimurium, strains TA-1535, TA-1537, and TA-1538, were used in modified Ames assays. Tissue homogenates from mice, rats, and monkeys were added to the culture media to see if benzoyl peroxide might be activated to a mutagenic compound. It was concluded that benzoyl peroxide exhibited no mutagenic activity in any of the in vitro microbial assays performed; this conclusion is consistent with the data presented. However, the benzoyl peroxide was added in dimethylsulfoxide, a solvent in which it is not soluble, although it did, nevertheless, allow the benzoyl peroxide to come in contact with the yeast and bacteria.

Correlation of Exposure and Effect

The one report [31] on the effects of inhalation of airborne dust containing benzoyl peroxide on humans stated that two plant inspectors had symptoms of nose and throat irritation on 2 days when the concentrations of benzoyl peroxide ranged from 1.34 to 17.0 mg/cu m. On the 3rd day, when

the concentrations of airborne benzoyl peroxide were 2.58-82.5 mg/cu m, they had symptoms of eye irritation, as well as of nose and throat irritation. However, no definite conclusions can be made from this report because the analytical information provided is insufficient for the reliability of the determinations to be assessed, so the concentrations of airborne benzoyl peroxide are questionable. Also, the presence of alum in the airborne dusts may have caused or contributed to the irritation.

Eye irritation tests in rabbits [42,43] and skin irritation tests on rabbits [42] and guinea pigs [46] have indicated that benzoyl peroxide is a low-grade irritant. There is some evidence that contact with benzoyl peroxide can cause sensitization in humans, although the incidence of this appeared low. Baird [32], Malten [34], and Jirasek and Kalensky [35] observed cases of occupational or contact dermatitis in humans, which were attributed to exposure to benzoyl peroxide. Benzoyl peroxide has been reported to be an allergen [34]; however, because it is unstable when in solution or in contact with flour and reacts to yield benzoic acid, it is not clear whether benzoic acid or benzoyl peroxide might be the allergen.

Benzoic acid itself is an allergen [32] and, perhaps because of its acid nature, an irritant. Redness and skin irritation occurring after exposure to benzoyl peroxide may be caused by primary irritation or by an allergic response. Baird [32] observed an allergic skin reaction and asthmatic wheezing in a baker who was exposed to benzoyl peroxide-treated flour. Malten [34] and Jirasek and Kalensky [35] diagnosed skin reactions as occupational contact dermatitis in workers who had become sensitized to benzoyl peroxide.

Some patients who used benzoyl peroxide for acne therapy were sensitized after repeated applications [36,37]; others had redness, which could have been primary skin irritation as well as a sensitization, but the authors [37,38] did not differentiate between the two. Morley [24] observed that only 1 of 180 patients treated with benzoyl peroxide could not tolerate the treatment. It was not stated whether this patient had an allergic response or a skin irritation.

There has been no evidence of systemic toxicity caused by benzoyl peroxide. Dogs given diets containing flour treated wih 0.8-28 g of benzoyl peroxide/100 pounds of flour had no apparent adverse effects [44,45]. No data were presented that would indicate the amount of benzoyl peroxide that remained in their diets after they were prepared, which involved steaming the flour treated with the compound. Sharratt and his colleagues [49] noted that male and female rats given benzoyl peroxide at concentrations of 280 or 28 ppm in a flour-based diet gained weight at a slower rate than the control rats; male rats given a diet with breadcrumbs made from flour treated with benzoyl peroxide at a concentration of 2.8 ppm gained weight at a rate similar to that of the controls. In another study [46], single dietary doses of 950, 400, or 200 mg/kg produced no ill effects. Ingestion of benzoyl peroxide in amounts far greater than those normally used to treat commercial flour had no apparent toxic effects in rats and dogs [42,44-46,49]. However, much of the benzoyl peroxide in the diets of these animals may have decomposed to benzoic acid by the time it was consumed.

Horgan et al [47] reported that, in mice, the LD50 of benzoyl peroxide administered through ip injection was 4.8 mg/mouse; later, Philpot

and Roodyn [48] calculated an LD50 in mice of 4.1 mg/mouse for benzoyl peroxide given by ip injection. Sharratt et al [49] reported that a subcutaneous injection of 50 mg of benzoyl peroxide/mouse (2,500 mg/kg) caused an abscess that healed in several weeks; no deaths occurred. Sharratt et al [49] also gave rats 120 mg of benzoyl peroxide by subcutaneous injection with no apparent adverse effects. The absorption of benzoyl peroxide in mice appears to vary greatly depending on the site of injection.

Laboratory tests reported by Ede [38] on 10 men and 10 women using acne medications containing benzoyl peroxide were normal, indicating no systemic effects from dermally applied benzoyl peroxide. No data were found that dealt specifically with absorption of benzoyl peroxide through the skin or from different sites of injection in humans or animals.

The flammability and explosiveness of pure benzoyl peroxide have been the cause of accidents involving serious injuries and fatalities [3,40]. Accidents involving only property damage are summarized in Chapter V.

Carcinogenicity, Mutagenicity, Teratogenicity, and Effects on Reproduction

The results of experiments designed to show if benzoyl peroxide has any carcinogenic activity when it is implanted [50], painted on skin [49,51], or injected [49] were negative. The results of tests to detect mutagenic effects of benzoyl peroxide in a modified dominant-lethal assay with mice [52] and in Ames assays with bacteria and yeast [53] were also negative. No data on teratogenesis or other effects of benzoyl peroxide on reproduction were found.

TABLE III-1

EFFECTS OF BENZOYL PEROXIDE EXPOSURE ON HUMANS

Route of Exposure	Exposure Concentration and Duration	Effects	Reference
Dermal	20%	Irritation in 1 of 180	24
"	1%, 5%, and 10% 9 24-hr applications	Severe eczematous skin reactions in 25 of 69 at end of experiment	36
"	5 % 12 hr	Marked erythema and burning	37
"	5% 48 hr	Severe irritation	37
"	Unknown	Positive patch test and dermatitis in 3 of 30	34
"	20 - 100%	Slight skin irritation	35
Respiratory	1.34-17.0 mg/cu m	Nose and throat irritation	31
"	2.58-82.5 mg/cu m	Eye, nose, and throat irritation	31
Dermal and respiratory	Unknown	Severe dermatitis, asthmatic wheezing	32

TABLE III-2

EFFECTS OF BENZOYL PEROXIDE EXPOSURE ON ANIMALS

Route of Exposure	Species	Exposure Concentration and Duration	Effects	Reference
Inhalation	Rats	Unknown concentration 3 hr	None	46
"	"	24.3 mg/l 4 hr	Eye squint, increased and decreased respiratory rates, salivation, lacrimation, erythema; no effects after 48 hr except lingering eye irritation	42
Oral	"	5,000 mg/kg	None during 14-d observation period	42
"	"	950 mg/kg	Slight muscular weakness in 1 of 2	46
"	"	400 mg/kg	Vasodilatation in 1 of 2	46
"	"	200 mg/kg	None	46
"	"	2,800 ppm in diet 120 wk	Testicular atrophy	49
"	"	280 ppm in diet 120 wk	None	49
"	"	28 ppm in diet 120 wk	"	49
"	"	28 ppm in breadcrumb diet 120 wk	Testicular atrophy	49

TABLE III-2 (CONTINUED)

EFFECTS OF BENZOYL PEROXIDE EXPOSURE ON ANIMALS

Route of Exposure	Species	Exposure Concentration and Duration	Effects	Reference
Oral	Rats	2.8 ppm of breadcrumb diet 120 wk	Testicular atrophy	49
"	Dogs	Benzoyl peroxide-treated* flour 71.6 % of diet for 6 wk	None	44
"	"	Benzoyl peroxide-treated** flour 80% of diet for 21 - 38 d	"	45
ip	Mice	62 mg/kg	"	52
"	"	54 mg/kg	"	52
sc	Rats	120 mg	"	49
"	Mice	50 mg	"	49
Eye contact	Rabbits	111.4 mg of 78% benzoyl peroxide 5 min	Redness of conjunctivae in 2 of 5 lasting up to 48 hr	42
"	"	120.7 mg of 78% benzoyl peroxide 24 hr	Slight opacity of cornea in 3 of 3 lasting up to 48 hr; redness of conjunctivae in 3 of 3 lasting up to 7d	42
"	"	Unknown amount of 93% benzoyl peroxide 1 min	None	43

TABLE III-2 (CONTINUED)

EFFECTS OF BENZOYL PEROXIDE EXPOSURE ON ANIMALS

Route of Exposure	Species	Exposure Concentration and Duration	Effects	Reference
Eye contact	Rabbit	Unknown amount of 50% benzoyl peroxide 1 min	None	43
Dermal	"	500 mg of 78% benzoyl peroxide 4 hr	"	42
"	Guinea pigs	0.25 - 1.0 g/kg 24 hr	Slight erythema, delayed scarring	46
"	"	5 - 20 ml/kg of 10% benzoyl peroxide in propylene glycol 24 hr	Slight erythema	46
"	Mice	50 mg of 50% suspension	No tumors	49
"	"	100 mg of 5% solution	"	51
sc implants	Rats	50 mg 24 mon	No tumors at site of benzoyl peroxide implant; no tumors attributed to benzoyl peroxide	50

*28 g of benzoyl peroxide/100 lb of flour
**0.8 g of benzoyl peroxide/100 lb of flour