

III. BIOLOGIC EFFECTS OF EXPOSURE

Extent of Exposure

In gaseous form, carbon dioxide is colorless, odorless, relatively inactive, and nonflammable. [1] Carbon dioxide, CO₂, has a molecular weight of 44.01, has been called carbonic acid gas, and, depending on temperature and pressure, may exist as a solid, liquid, or gas. The solid phase is known as dry ice. Table XIV-1 [2] presents some physical properties of carbon dioxide. Carbon dioxide is normally present in the atmosphere at a concentration of approximately 0.03% (300 ppm). [3] It is a normal body constituent arising from cellular respiration.

The bulk of the commercially available carbon dioxide is recovered from industrial processes in which it is generated as a byproduct. [4] Carbon dioxide occurs in natural gas wells and is a product of carbonaceous fuel combustion and of fermentation. [5] Industrial processes in which carbon dioxide gas is generated as a byproduct include synthetic ammonia production, lime kiln operations, and fermentation. [5] Carbon dioxide is a byproduct of synthesis gas (carbon monoxide and hydrogen) in the production of ammonia, methanol, and other chemicals. [5] Carbon dioxide is also generated as a byproduct during the manufacture of ethylene oxide and in the catalytic oxidation of benzene during production of maleic anhydride. [5]

Much of this byproduct carbon dioxide is vented into the atmosphere and not recovered. [4] Nevertheless, commercial manufacture of carbon dioxide in the United States rose from 299,000 short tons (short ton = 2,000 pounds) in 1940 to 1,135,000 short tons in 1973. The 1973 figure

includes 815,000 short tons of liquid and gas and 321,000 short tons of solid. The current manufacturing trend favors the production of the liquid. [4]

Commercial uses of solid carbon dioxide primarily concern the refrigeration of food. [5] Dry ice is a coolant for dairy products, meat, poultry, and frozen foods during transit. [5] Dry ice is also used for industrial applications in which low temperatures are essential, eg, the grinding of heat-sensitive materials, low-temperature testing, chilling of molded elastomer products, and chilling of various pieces of equipment in heavy manufacturing. [4]

Liquid carbon dioxide is used primarily in refrigeration and is replacing the solid phase in applications such as tumbling of rubber products, use in chemical reactions to control temperatures, and by direct injection into foodstuffs as a refrigerant. The vapor pressure of liquid carbon dioxide has been used as a source of power in such operations as remote signal devices and spray painting. Other applications include its use in fire extinguishers and in blasting operations in coal mining (Cardox method). [6]

The gaseous phase of carbon dioxide is used commercially in the carbonation of beverages; as an inert blanket over paints and resins; in shielded-arc welding operations; as a raw material in the production of urea, sodium carbonate, sodium bicarbonate, and sodium salicylate; as an aerosol propellant; as a growth factor in bacteriology; and, in solution as carbonic acid, as a pH-control agent. [7,8] Carbon dioxide is used in hydrocracking and hydrotreating of petroleum products. [4,5] It is used as an "acid egg" (a pocket of carbon dioxide, also known as a blowcase,

which moves another substance by pressure) in moving raw materials and partially formed products within plant pipelines. [9] It has also been used as a coma-inducing agent in psychiatric treatment [10] and is present in therapeutic oxygen. Carbon dioxide in the atmosphere plays a major role in the photosynthetic cycle of green plants.

In specific instances, the flushing (and pressure-creating) action of carbon dioxide facilitates the recovery of petroleum products in oil and gas wells. [4] Sometimes the gas is used to immobilize animals in humane killing operations at commercial slaughterhouses and is also occasionally used in cloud-seeding operations. [11]

NIOSH estimates that approximately two million workers are potentially exposed to carbon dioxide in the United States. Table XIV-2 [12] is a list of occupations with potential exposure to carbon dioxide.

Historical Reports

Reed and Comley [6] attributed the earliest recognition of carbon dioxide gas as a separate substance to Van Helmont (1577-1644). Van Helmont was reported to have identified the gas as a combustion product of charcoal and recognized it to be the same gas as that produced during natural fermentation. Cutting [13] reported that in the early 1800's Hickman used carbon dioxide gas as an anesthetic for animal surgery. Hickman completed a series of experiments in which animals were rendered unconscious after inhaling small quantities of the gas. [14] Surgical procedures were performed which, according to Hickman, were not painful to the animals while under the influence of the gas. [14] A century after Hickman's first experiments with carbon dioxide, Leake et al [15] repeated

these tests and confirmed the anesthetic properties of the gas. This experimentation led to the use of carbon dioxide as a shock inducer for the treatment of psychiatric patients. The authors [15] also reported that another investigator, Meduna, had begun similar studies using carbon dioxide as therapy for treating psychiatric disorders.

Foregger [16] attributed the precise identification of "fixed air" (carbon dioxide) to Joseph Black, who published a dissertation entitled "Experiments on Magnesia Alba, Quicklime and other Alcaline Substances" in 1756. Black observed that carbon dioxide was different from common air and that, although it remained "fixed" in air, it had specific properties of its own. He also noted that limewater could be used to determine the presence of "fixed air" in solution. In addition, Black reported that carbon dioxide extinguished flames and had an affinity for lime. He also observed that the convulsions and subsequent death of exposed animals were toxic effects of carbon dioxide. Black noted that animals recovered upon timely removal from exposure to the gas and that the gas "arises from the lungs of animals." [16]

In 1838, newspaper accounts [17,18] of the death of a church night watchman from the effects of a gas emitted by a coal stove led to a series of discussions between Bird and Snow. [17,18] These discussions, which were printed as part of the report of death, finally led to agreement that it was probably "carbonic acid gas" (carbon dioxide) which did, in fact, produce the ill effects. Foregger [19] reported Snow's observations in experiments on animals and on himself that carbon dioxide gas was toxic when inhaled. Snow then concluded that a decrease in oxygen content in the atmosphere resulting from an increased carbon dioxide concentration was

dangerous to life and that the two conditions had synergistic effects. He showed that inhalation of carbon dioxide gas displaced oxygen thereby resulting in the deaths of white mice exposed to it. Snow also concluded that carbon dioxide had direct effects on the body distinct from those due to lack of oxygen.

In an 1892 report entitled "The Physiological Effects of Air Vitiating by Respiration," Haldane and Smith [20] commented on the results of extensive experiments by themselves and others on the toxicity of carbon dioxide gas. The experiments were conducted in an enclosed chamber and the carbon dioxide was merely the subject's own exhaled breath rebreathed. As a control, the same procedure was used, except that a pan of soda-lime absorbed the carbon dioxide in the chamber. The authors concluded that the dangers of air vitiating by respiration were due to excesses of carbon dioxide gas and not to oxygen deficiencies; that an excess of the gas caused hyperpnea* at 3-4%; that 10% carbon dioxide produced extreme respiratory distress; and that an excess of the gas induced frontal headaches.

Physiologic Considerations

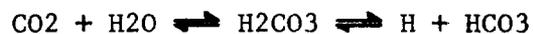
Carbon dioxide has many important functions in maintaining normal body activities. It is a key factor in the control of respiration and cerebral circulation. [21] It acts peripherally, both as a vasodilator and as a vasoconstrictor, and is a powerful cerebral vasodilator. [22] At high concentrations, it exerts a stimulating effect on the CNS (central nervous system), while excessive levels exert depressant effects. [22]

Since it is a gas, its concentration is often considered in terms of

partial pressure* or tension relative to the total atmospheric pressure of 760 mmHg. Thus, the normal partial pressure of carbon dioxide (pCO₂) in air is 0.03% of 760 mmHg, or 0.2 mmHg.

During its production in the body, carbon dioxide diffuses from tissue cells into the surrounding capillaries and is carried by the blood in chemical combination with hemoglobin; in physical solution as dissolved carbon dioxide, carbonic acid, or bicarbonate ions; and as minor amounts of other carbamino compounds (carbon dioxide in combination with plasma proteins). [3] Carbon dioxide is produced in the body and its partial pressure under normal conditions in pulmonary capillary blood (46 mmHg) is greater than that in alveolar air (40 mmHg). [3] The gas is exchanged freely through the alveolar membrane and is thus released from the lungs by diffusion because of the concentration gradient existing between the blood and the air in the alveoli*. Because of the free exchange through the alveolar membrane, any increased carbon dioxide tension in the alveolar air resulting from increased pCO₂ in the inspired air will immediately increase the carbon dioxide tension of the blood.

Dissolved carbon dioxide in the blood undergoes hydration according to the following reaction [3]:



The enzyme carbonic anhydrase*, found in the erythrocytes* and in the renal tubular epithelium, catalyzes the initial stage of this reaction. [23] The presence of this enzyme allows the reaction to proceed extremely rapidly. As a result, there is an accumulation of bicarbonate anions in the erythrocytes. Equilibrium is maintained by the diffusion of bicarbonate

anions into the plasma. Since cations (eg, hydrogen ions) cannot readily diffuse through the erythrocyte membrane, electrical neutrality within the red blood cell is maintained by the diffusion of chloride anions into the cell from the plasma (chloride shift). [3] The hydrogen ions remaining in the red blood cells combine with hemoglobin, while small amounts of unreacted carbon dioxide in the plasma combine with plasma proteins. [3] The hydration of carbon dioxide also occurs in plasma although, since it is not catalyzed by carbonic anhydrase, it proceeds very slowly. [3,23] In terms of the body's acid-base balance, the Henderson-Hasselbalch equation [3] shows that pH depends on the ratio of bicarbonate ions to carbon dioxide in the following way (see Appendix V):

$$\text{pH} = \text{pK} + \log \frac{[\text{HCO}_3]}{[\text{CO}_2]}$$

This relationship has been demonstrated experimentally by Van Ypersele de Strihou et al [24] who studied data from 420 patients with chronic hypercapnia*. Through a combination of the above reactions, an increase in the partial pressure of carbon dioxide (pCO₂) in the blood is reflected by increases in both the hydrogen and the bicarbonate ions which result in respiratory acidosis if the capacity of the blood buffering systems is exceeded. This rapidly triggers respiratory elimination of carbon dioxide through an increased ventilation rate. While pulmonary compensation for respiratory acidosis is rapid, renal regulation of the acid-base balance requires an extended period of time and initially involves a retention of bicarbonate. With prolonged carbon dioxide exposure, there is increased bicarbonate excretion. Renal excretion of ammonium ions and titratable acid provides for removal of hydrogen ions in exchange for reabsorption of

sodium ions which then form sodium bicarbonate in plasma. [23] Therefore, the respiratory mechanisms act to stabilize the carbon dioxide concentration while renal mechanisms act in stabilizing the bicarbonate ion concentration.

The extremely easy diffusion of carbon dioxide through tissue membranes accounts for the rapidity with which the gas produces respiratory and CNS effects. [25] Its solubility in tissue fluids is approximately 20 times greater than that of oxygen. [25] As arterial carbon dioxide tension increases, various chemoreceptors in the body are affected. The central and peripheral chemoreceptors are sensitive to changes in carbon dioxide tension and are freely accessible to carbon dioxide because of its rapid diffusibility. These chemoreceptors include the peripheral cardiovascular ones in the aortic arch and in the carotid body and the central medullary chemoreceptors. The medullary respiratory chemoreceptors are extremely sensitive to changes in both the hydrogen ion and the carbon dioxide concentrations in the cerebrospinal fluid (CSF). [26] Because of this and the extreme permeability of the blood-brain and blood-CSF barriers to carbon dioxide, small changes in blood carbon dioxide concentration are readily identified by the central chemoreceptors. [26] Therefore, respiratory and CNS responses to changes in carbon dioxide concentrations are immediate and unlike responses to any other weak acid. This sensitivity and homeostasis are central concepts throughout the discussion of the effects of carbon dioxide in this document.

At very high levels, carbon dioxide induces depressed respiration and the CNS effects are narcotic. [27] This is evidenced by studies [22,28] which showed that loss of consciousness occurs at high concentrations and

is indicative of the direct effects of carbon dioxide, rather than of a homeostatic mechanism in action.

Acclimatization, or development of tolerance, as a result of chronic exposure at elevated carbon dioxide levels has been demonstrated. In these situations, the body develops a tolerance, or an apparently reduced sensitivity, to the respiratory and cardiovascular changes which occur. The information provided in the following sections confirms the fact that this is accomplished, in part, through the activation of compensatory mechanisms to offset the resultant imbalance and restore the body to homeostasis. In general, it appears that, the greater the physiologic imbalance, the greater the compensation necessary to facilitate an adequate adjustment.

Results of the following studies are also summarized in Tables III-1 through III-7 at the end of this chapter.

Effects on Humans

(a) Clinical Effects on the Central Nervous System

Friedlander and Hill [28] exposed 37 young adult psychiatric patients to 30% carbon dioxide in oxygen. These patients had received no previous treatment of this type. During administration of the gas mixture by mask for 50-52 seconds, the average patient "lost consciousness" within 24-28 seconds and regained it after 110 seconds.

Lambertsen [22] described the neurologic effects of the abrupt administration of high concentrations of carbon dioxide. Neurologic signs, including eye flickering, psychomotor excitation, and myoclonic twitches appeared after approximately 1.5 minutes of exposure at 10% of the gas. At

15%, the same signs were recorded, as were increased muscle tone, perspiration, flushing, restlessness, dilated pupils, leg flexion, and torsion spasms. At concentrations of 20 and 30% carbon dioxide, the same signs were observed in addition to tonic and tonic-clonic seizures. Unconsciousness and convulsions occurred in less than 2 minutes at 30% carbon dioxide in oxygen. All of these signs were evident at all concentrations within 3 minutes after exposure to the gas began.

Studies of the level of carbon dioxide and the time necessary to produce unconsciousness were also performed by Spealman. The results were not published separately but were presented in the 1953 edition of Aviation Toxicology. [29] The experiments consisted of exposing three subjects, ages and sex unspecified, to carbon dioxide-air mixtures consisting of 17.0% carbon dioxide and 17.3% oxygen and of 27.9% carbon dioxide and 15.0% oxygen. The subjects, who were administered the gas by mask, were requested to breathe the mixtures to the point of unconsciousness. The mixture containing 17% carbon dioxide was tolerated for an average of 37 seconds (range, 20-52 seconds), while the mixture containing 27.9% carbon dioxide could be tolerated for only 25 seconds (range, 16-35 seconds). All subjects experienced throat irritation, increased respiration, dimness of vision, and dizziness. After the exposure, they were unable to answer questions or to support themselves for periods of from 17 to 52 seconds. The author reported the presence of muscle spasms in two of the subjects. These symptoms were thought to be indicative of the start of convulsions.

Other studies on the toxicity of acute carbon dioxide exposures have identified many overt clinical symptoms. Upon exposure at 7.6% of the gas in oxygen for an average of 7.4 minutes, dyspnea*, dizziness, headache,

sweating, and "fullness in head" were reported. [27] These same symptoms in addition to restlessness and faintness were reported during 3.8 minutes of exposure at 10.4% of the gas. [27] Loss of consciousness was reported in 1 of 42 subjects at 7.6% and in 3 of 31 at 10.4% carbon dioxide. The majority of the headaches reported began or were accentuated after the exposure. In a study on the effects of carbon dioxide on the CNS, Schaefer [30] reported headache, dizziness, restlessness, and/or dyspnea in subjects at 7.5% carbon dioxide for 15 minutes. The signs and symptoms reported by these 42 subjects are presented in Table XIV-3. Both of these studies [27,30] concerned short-duration exposures (3.8, 7.4, and 15 minutes) and yielded data on acute responses. Sechzer et al [31] reported severe symptoms of carbon dioxide inhalation, such as headache, sweating, and auditory and visual hallucinations which appeared in most subjects only when concentrations in excess of 7% were administered. Vomiting was present in some subjects and nearly all lost consciousness at levels above 80 mmHg (10.5%) carbon dioxide.

While the preceding signs and symptoms were observed during acute exposures to carbon dioxide, the following neurologic effects have been reported during chronic exposures. Clinical symptoms of chronic hypercapnia in patients suffering from pulmonary disease included headache, somnolence, mental confusion, lassitude, irritability, and unconsciousness. [32] Other signs and symptoms associated with this syndrome were hyporeflexia, occasional convulsions, flaccid paralysis, tremors, engorged retinal veins, papilledema, and death. [32] Glatte et al [33] reported that a 5-day continuous exposure in a chamber at an ambient concentration of 3% carbon dioxide resulted in mild, frontal headaches in four of seven

subjects during the first 2 days of exposure. Intermittent exposure of one subject at a gradually increasing concentration of from 0.03% to 3% carbon dioxide in air, reaching the maximum after 12-15 hours, resulted in symptoms of emotional disturbance. [34] Since only one person was studied in this experiment, [34] the role of emotional variability unrelated to carbon dioxide must be considered in evaluating these results.

(b) Effects on Respiratory Function

Dripps and Comroe [27] reported the respiratory effects of carbon dioxide on 44 healthy males, aged 21-26 years, who were exposed to carbon dioxide at concentrations of 7.6 or 10.4%. The gas was supplied through a face mask as the subject sat on a chair or remained stationary on a bicycle. The average respiratory minute volume* observed at 7.6% carbon dioxide was 51.5 liters/minute, and at 10.4% of the gas it was 76.3 liters/minute. For comparison, minute volume data from several investigators were graphed. The graph showed that respiration rises from a normal resting level in air (approximately 0.03% carbon dioxide) of 7 liters/minute to averages of approximately 8 liters/minute at 1% carbon dioxide and 9, 11, 14, and 26 liters/minute at 2, 3, 4, and 5% carbon dioxide, respectively. These elevated respiratory rates rapidly decreased after cessation of increased carbon dioxide inhalation, returning to normal 3 minutes after gas administration was discontinued.

In a study reported by Faucett and Newman, [35] 23 healthy males were exposed at a constant concentration of 1.5% carbon dioxide in air (20.5% oxygen) for 42 days in a submarine which served as the experimental chamber. This experiment was known as "Operation Hideout" and has yielded data on which were based many subsequent publications. The exposure was

preceded and followed by 9-day control periods at atmospheric air to assess baseline data. Laboratory testing was conducted for 35 days preceding and 35 days following the control periods. Preliminary analyses of the results showed an 8% increase in alveolar carbon dioxide tension after 2 days of exposure at 1.5% carbon dioxide. The carbon dioxide tension remained elevated throughout the 9-day postexposure control period. Also observed was an 8% increase in ventilatory rate, which eventually returned to normal levels within 3 days after exposure. During the first 14 days of exposure, increased oxygen consumption was noted; the level returned to normal during the latter part of the experiment. Respiratory excretion of carbon dioxide dropped during the initial phase of exposure and subsequently increased for 2 weeks. This was followed by another drop in the final 2 weeks of exposure and a return to normal when subjects again breathed normal air. Ventilatory sensitivity to a challenge with 5% carbon dioxide was tested (by measuring rate and depth of respiration) at the following intervals: before the experimental period, at the end of the 6-week test period, and again 3 weeks after the exposure. Ventilatory response to a challenge of 5% carbon dioxide was significantly decreased at the end of the sixth week and did not return to pretest levels even after a 3-week recovery period. This decreased respiratory response indicated possible adaptation or tolerance to the gas on continued exposure.

In a later, more detailed analysis of the "Operation Hideout" study, [35] Schaefer et al [36] confirmed and expanded on the experimental conclusions. They reported an increase in respiratory minute volume during days 24-42 of the exposure to 1.5% carbon dioxide from 2.85 to 3.81 minute volumes/sq m. Minute volume fell on resumption of normal air breathing and

subsequently increased slightly. During the entire exposure and for the first 9 days postexposure, tidal volume* was increased. The maximum was reached near the end of the exposure period at which time tidal volume was in excess of 800 ml. The increased respiratory rate seen initially remained elevated for about 35 days but dropped to below control levels near the end of the exposure, remaining low during 4 weeks of recovery on air. The alveolar carbon dioxide tension was significantly elevated throughout the 42-day exposure and remained above control levels throughout the 9-day recovery period. This elevated alveolar carbon dioxide tension indicated bodily retention of the gas. The respiratory exchange ratio (the ratio of carbon dioxide output to oxygen uptake) was seen to follow the pattern of respiratory excretion of carbon dioxide, decreasing during the first 3 weeks of exposure, then rising during the second phase to reach greater-than-control levels after the return to normal air. Based on these and the earlier observations, Schaefer et al [36] divided the 42-day exposure period into two parts. The first phase was characterized by a somewhat lower although still significantly increased carbon dioxide excretion and respiratory exchange ratio. The second phase was characterized by increased alveolar carbon dioxide tension, increased respiratory minute volume, and increased excretion of carbon dioxide. This phase continued through the 9-day recovery period. The first phase was indicative of uncompensated acidosis* which lasted during days 1-23, while compensated acidosis, the second phase, was observed on days 24-42. Alterations in blood pH also paralleled these respiratory changes and will be discussed in the section dealing with effects on acid-base balance.

Schaefer et al [36] also reported a significant increase in anatomic dead space*. They theorized that a direct effect on smooth muscle, which dilated the airway, caused this increase. The buffering capacity of bicarbonate in the arterial blood led to limited increases of carbon dioxide tensions in arterial and mixed venous blood; this action limited the respiratory response to carbon dioxide. Gude and Schaefer [37] also reported this phenomenon in a later study in which they also observed 60 and 61% increases in alveolar dead space after 20 days of exposure to 0.8 and 0.9% carbon dioxide, respectively.

Schaefer [38] reported that previous extended exposure to carbon dioxide apparently resulted in the development of tolerance to subsequent short-term exposures. His experiment involved more than 60 healthy subjects who were divided into high- and low-sensitivity groups according to their initial respiratory responses after 10- to 15-minute exposures to 5.4 and 7.5% carbon dioxide. The author neither specified how the gas was administered, nor did he indicate the sex of the subjects. The high-ventilatory-response group was composed primarily of laboratory personnel. The low-ventilatory-response group was composed partly of young naval personnel and instructors at an escape training tank. The tank instructors' daily duties involved long periods of breath-holding under water, which resulted in an accumulation of carbon dioxide in body tissues. These men were slow breathers and had lower respiratory rates and higher tidal volumes (1.018 liters) than the high-response group (0.760 liters). Both groups were exposed to carbon dioxide at concentrations of 1.5, 3.3, 5.4, and 7.5% for 15 minutes at each concentration. The low-ventilatory-response group consistently showed a decreased response at all levels of

increased carbon dioxide. At 1.5% carbon dioxide, this group had a higher tidal volume and lower respiratory rate than did the high-response group, while at 3.3% of the gas, the groups showed very similar responses. At 5.4 and 7.5% carbon dioxide, the high-ventilatory-response group showed a markedly increased response over the low group in both tidal volume and respiratory rate. The low-ventilatory-response group had an initial alveolar carbon dioxide tension 2 mmHg higher than the high-response group prior to the experiment. Possibly, the low-response subjects had developed a tolerance to carbon dioxide as a result of their duties and the natural selection apparent in their chosen field of diving. This tolerance could also possibly be attributed to a repeated exposure for brief periods to elevated tissue carbon dioxide as a consequence of breath holding. This conclusion is supported by a decreased ventilatory response to a 5% carbon dioxide challenge observed in diving instructors during performance of their duties and an increased response to this challenge after a 3-month absence from their assigned duties. The results suggested that the effects of carbon dioxide must always be viewed in terms of the subject's normal respiratory pattern.

Tashkin and Simmons [39] studied the effects of carbon dioxide on airway conductance. Nine normal (aged 18-36 years) and eight asthmatic (aged 19-34 years) subjects were exposed to 2.5, 5, 7.5, and 10% carbon dioxide in air. Recorded data included airway resistance, respiratory frequency, specific airway conductance, thoracic gas volume (the volume of gas in the entire thorax, whether in communication with the airways or not), and end-tidal carbon dioxide tension. Normal subjects showed decreases in specific airway conductance during inhalation of 5, 7.5, or

10% carbon dioxide. Results at the 2.5% level of carbon dioxide were not significantly different from those observed during air exposure. Since no such changes were observed during 2 hours of breathing room air, spontaneous airway narrowing over the time of the experiment was excluded as an explanation for this effect. Further, the investigators taught the subjects to voluntarily adjust their rate and depth of respiration to simulate conditions of inhalation of 7.5 and 10% carbon dioxide by coaching them until the desired rate (in synchronization with a metronome) and depth (as monitored by an oscilloscopic monitor) were reached and maintained. Under these simulation conditions, the airway conductance did not vary; thus, the mechanical effect of increased ventilation was excluded as an explanation for the conductance decrease. The use of specific cholinergic- and adrenergic-blocking drugs yielded results which similarly indicated that hypercapnia caused airway constriction. The responses to 7.5 and 10% carbon dioxide after atropine administration were the same as those produced by hypercapnia alone. Taskin and Simmons [39] concluded that the constriction had not been mediated by a vagal or local parasympathetic reflex. In contrast, at 5% carbon dioxide, atropine had a protective effect against increased airway resistance, indicating a cholinergic, reflex, constrictor effect at the lower carbon dioxide levels. The authors regarded the results indicative of a direct effect on airway constriction of hypercapnia induced by 7.5 and 10% carbon dioxide in the inspired air. The asthmatic subjects' responses were similar to those of normal subjects in degree of increased ventilation. However, at 5-10% carbon dioxide, airway constriction was not evident. Further, cholinergic blockade did not affect constriction. In contrast, beta-adrenergic blockage produced by

propranolol pretreatment caused significant airway constriction in asthmatics during inhalation of 5-10% carbon dioxide. The authors concluded that this response demonstrated protection in asthmatic subjects against constriction by sympathetic dilatation which the beta-adrenergic block appeared to unmask. Additionally, considering the results of propranolol studies, the authors suggested that higher concentrations of carbon dioxide (7.5 and 10%) had stimulated sympathetic bronchodilation sufficiently to offset partially the direct airway-constrictor effect of the gas.

The respiratory response to carbon dioxide in terms of the total mechanical work performed by the respiratory muscles was characterized by Brodovsky et al. [40] The effects of changes in arterial carbon dioxide tension were studied in 12 normal subjects and 10 emphysematous patients. The investigators observed ventilation and measured oxygen consumption and pCO₂. The total mechanical work of breathing was calculated from the relationship of the oxygen cost of increased ventilation to the efficiency of the respiratory muscles. Measurements were made at various levels of ventilatory response, which were created in the laboratory by interposing dead space between a spirometer and the subject, thereby increasing alveolar carbon dioxide levels. The authors concluded that incremental changes in total mechanical work were more sensitive measures of the respiratory response to carbon dioxide than were increments of ventilation or oxygen consumption. They also reported that the response of the emphysematous subjects was lower than that of normal subjects in terms of total mechanical work done. Since the emphysematous patients also had a lower normal ventilatory response to carbon dioxide, this work tends to

confirm previously reported findings [35,38] regarding respiratory adaptation, since the chronic pulmonary insufficiency of emphysematous patients creates a chronic hypercapnic situation.

A subsequent study by Schaefer et al [41] reported the effects of intermittent exposure to carbon dioxide on respiration. One healthy man was exposed to carbon dioxide at an increasing concentration from 0.03 to 3% carbon dioxide for 15 hours daily for 6 days. Resting respiratory minute volume, oxygen consumption, carbon dioxide excretion, alveolar carbon dioxide, and oxygen tensions were measured twice daily. The authors [41] demonstrated that, after only 3 days, the daily 9-hour return to normal air was not sufficient to eliminate the gas which had accumulated during the 15-hour test period. The authors noted an increased ventilatory response to a subsequent challenge with 5% carbon dioxide. Also noted was the doubling of respiratory minute volume at the end of 15 hours. After the subject spent 9 hours in room air, all values had returned to normal except alveolar carbon dioxide tension, which rose on days 4 and 5 to a peak higher than that reached during the 15-hour test periods. This was evidence that carbon dioxide was retained in body tissues and was not entirely released even during the 9-hour rest period. These responses to intermittent carbon dioxide are the opposite of those Faucett and Newman [35] and Schaefer et al [36] found after chronic exposure to the gas. However, this report was based on one experiment with only one subject which seriously limits its usefulness.

Respiratory acclimatization to 3% carbon dioxide was demonstrated by Chapin et al [42] in an experiment with two healthy men. The men were exposed continuously to the gas in 21% oxygen for 78 hours in an airtight

environmental chamber. Ventilation volume, respiratory frequency, and samples of alveolar gas* were measured. Alveolar gas pCO₂ for both resting and breath holding reached a maximum between 8 and 13 hours of exposure. Twenty hours after the exposure, the values had returned to their original levels. The criteria used by the authors as being indicative of reduced ventilatory sensitivity were maintenance of higher-than-normal alveolar carbon dioxide levels during air breathing, carbon dioxide breathing, or breath holding, coupled with a lowered ventilatory response to carbon dioxide. In a subsequent portion of the experiment, the subjects were exposed to challenges of 5.7 and 5.9% carbon dioxide both before and after acclimatization. The unacclimatized (preexposure) respiratory response to 5.7% of the gas was 43.3 liters/minute and the acclimatized (immediate postexposure) respiratory response to 5.9% was 32.7 liters/minute. In contrast, the unacclimatized response to 3% carbon dioxide, measured early during the exposure, was 15.05 liters/minute, and the acclimatized response, later during the exposure, was measured at 12.9 liters/minute. This, according to the authors, indicated a damping of ventilatory response due to acclimatization after a 78-hour exposure to 3% carbon dioxide.

Further evidence of respiratory acclimatization has been provided by Kuznetsov and Kalinichenko. [43] In a study involving seven healthy men, aged 20-25 years, adaptation to low carbon dioxide levels was reported. The subjects were exposed at carbon dioxide concentrations of 7.1-14.2 mmHg (0.9-1.9%) for 30 days in a pressure chamber. At normal pressure and exposure at 7.5-7.9 mmHg (0.99-1.04%), alveolar carbon dioxide tension increased from 37.9 to 42.0 mmHg, while pulmonary ventilation generally increased by 0.5-1 liter/minute. In some of the subjects, pulmonary

ventilation did decrease. Although it is not clear at which carbon dioxide level this occurred, at a higher carbon dioxide level, 14.7-15.8 mmHg (1.9-2.08%), ventilation generally increased 1-2.5 liters/minute, while alveolar carbon dioxide tension rose to 46 mmHg. The alveolar carbon dioxide tension rose steadily throughout the experiment, while ventilation reached a maximum early and retained that level throughout the experiment.

(c) Cardiovascular Effects

Effects of high carbon dioxide concentrations on the electrocardiogram (ECG) were studied by MacDonald and Simonson. [44] The subjects were 17 males, aged 25-48 years. All were psychiatric patients hospitalized at the time of the experiment, and all had normal cardiac functions and no evidence of hypertension prior to the study. A mixture of 30% carbon dioxide and 70% oxygen was administered by mask for an average of 38 seconds. Narcosis was evident approximately 20-30 seconds after administration of the gas mixture was begun. In 16 of the 27 episodes involving 12 of the 17 patients, some abnormality of auricular or nodal activity was reported. Among the changes noted were extrasystoles (premature contractions), premature auricular and nodal beats, auricular tachycardia, and supraventricular tachycardia. These, the authors noted, were not alarming but might suggest that serious supraventricular or ventricular disturbances could occur, especially if any preexisting cardiac problems were present. Friedlander and Hill [28] also have reported extrasystoles in 5 of 37 patients exposed to 30% carbon dioxide and 70% oxygen. However, it is not possible to conclude definitely that carbon dioxide alone was responsible for the observed cardiac abnormalities since the effect of the high concentration of oxygen used in these experiments

remains unclear.

Okajima and Simonson [45] reported on the effects of 6% carbon dioxide, administered through face masks for 6-8 minutes, on the ECG's of 148 healthy men. The study subjects were divided into groups of older (mean age 60.9 years) and younger (mean age 23.3 years) men. The authors observed a significant (19.08%) decrease in the amplitude of the QRS complex of the ECG in the older men as compared to the change of 11.63% in the younger men. Okajima and Simonson [45] concluded that the more pronounced change seen in the older men was probably due to a decreased tolerance to carbon dioxide which they suggested was an age-related effect. The authors did not state whether the observed changes were indicative of further serious complications.

Determinations of the various circulatory responses to carbon dioxide were made by Dripps and Comroe [27] in 1947. Forty-four healthy men were exposed to 7.6 and 10.4% carbon dioxide through an aviation-type face mask for respective averages of 7.4 and 3.8 minutes. General increases in systolic and diastolic blood pressures were observed. The average increases in systolic blood pressure were 30.8 mmHg at 7.6% carbon dioxide and 33.4 mmHg at 10.4%. The increases in diastolic pressure were 22.2 mmHg at 7.6% carbon dioxide and 25.0 mmHg at 10.4%. When the subjects returned to normal air, the diastolic blood pressure decreased immediately to below the pretest level, while the systolic pressure fell only slightly from its elevated level. At 7.6% carbon dioxide, the average pulse rate increased approximately 17 beats/minute, and, at 10.4% carbon dioxide, an increase of approximately 16 beats/minute was noted.

The effects of various levels of carbon dioxide concentration on cerebral blood flow were reported by Kety and Schmidt. [46] Twelve healthy men were exposed to 5 or 7% carbon dioxide in 21% oxygen with 74 or 72% nitrogen for 15-30 minutes. Blood oxygen, carbon dioxide, and pH values, as well as mean arterial blood pressures, and systolic and diastolic blood pressures were determined. The authors calculated cerebral oxygen consumption and cerebrovascular resistance from these data. The studies at 5 and 7% carbon dioxide showed an average increase of 75% (53-93 cc/100 g/min) in cerebral blood flow as a result of exposure. Carbon dioxide content and hydrogen ion concentration in arterial blood also rose, although cerebral oxygen consumption did not. The mean cerebrovascular resistance was reduced by about one-third. Arterial blood pressure rose as expected; however, cardiac output was not changed significantly. The headaches reported in previous studies [27,30,31,33] probably can be explained in part by the cerebral blood flow increases these authors observed.

The previously cited report by Sechzer et al [31] also noted a small incidence of cardiac arrhythmias in subjects exposed to between 7 and 14% carbon dioxide. Eight of 12 subjects exhibited some type of rhythm irregularity when exposed to the gas such that the exhaled carbon dioxide tension was in excess of 34 mmHg (4.4%). These irregularities were minor except for nodal or ventricular extrasystoles noted in a few subjects. All subjects were at rest during the experiment. Another report of cardiac abnormalities appeared in the article by Sinclair et al [47] discussed in subsection (g), Effects on Acid-Base and Electrolyte Balance. Twelve men were exposed at 3.9 or 2.7% carbon dioxide for 5, 11, or 30 days. Nine

incidents of cardiac abnormalities were reported; however, five of these occurred in one subject. All incidents were associated with exercise routines, some performed in normal room air. Ectopic foci* were the predominant abnormality. There was no correlation between increased carbon dioxide levels or workload and the incidence of such occurrences.

(d) Behavioral Effects

Schaefer, [30] in a study designed to investigate the effects of carbon dioxide and electrolyte shifts on the CNS, performed experiments on 42 healthy subjects divided into high- and low-ventilatory-response groups. The author did not specify the method of administration of the gas or the ages or sex of the subjects. Fifteen-minute exposures at carbon dioxide concentrations of 1.5, 3.3, 5.4, and 7.5% were described. The flicker-fusion and alpha-blocking times of the subjects were two of the responses measured. Schaefer observed that the low-ventilatory-response group showed no appreciable symptoms except at the 7.5% level. Measurement of flicker-fusion threshold showed a decrease after exposure to carbon dioxide at 3.3% or greater. Simultaneously, the latent time of alpha blocking after light stimulus increased. These results led the author to conclude that depression of the CNS was occurring. Because the low-ventilatory-response group also showed some effects indicative of autonomic nervous system involvement including changes in blood sugar, pulse rate, and muscle potential, the author speculated on the possibility that adaptation to increasing carbon dioxide levels had occurred in this group.

Weybrew [34] investigated the effects of carbon dioxide on psychologic function. In a 30-cu m recompression chamber, a 24-year-old man was exposed to carbon dioxide at a concentration which increased from

0.03 to 3% over a 15-hour period. The experiment continued for 12 days and consisted of two 3-day control periods at the beginning and end of a 6-day test period. Tests of psychologic efficiency included: single-digit addition, letter cancellation, and response analysis testing (RATER). In addition, the subject was offered a list of adjectives from which to choose those indicative of his mood and interests. Results of the various tests did not show any remarkable changes caused by the carbon dioxide exposure. The author concluded from the adjective-choice test that the experiment had produced some emotional changes in the subject. Since these tests were conducted in a chamber, Weybrew believed that the confinement may have influenced the experimental data and that no validation of results could be produced.

A report confirming the absence of psychomotor impairment on inhalation of 4% carbon dioxide in six healthy volunteer airmen was published by Storm and Giannetta. [48] The subjects were exposed continuously to 30 torr (4%) carbon dioxide for 2 weeks which were preceded and followed by 2-week baseline and recovery periods in an environmental chamber. Subjective psychomotor tests included the SAM complex coordinator and the repetitive psychometric measures (RPM). The subjects were trained in both procedures before the experiment. The SAM complex coordinator was used only during the baseline and recovery periods; the RPM was administered daily during the test period. The 2-week lack of work with the complex coordinator resulted in the loss of skill by all subjects. Since this was seen in the experimental group and in the six control subjects, the decrement was attributed to lack of practice rather than to the effects of the exposure. No decrement in performance with the RPM was

noticed in either group. The test series measured complex tracking performance, eye-hand coordination, and problem-solving ability, all of which were unaffected by 2 weeks of exposure to 4% carbon dioxide.

The effects of carbon dioxide on vision were reported by Weitzman et al [49] in 1969. In this study, one healthy 24-year-old man was exposed at a gradually increased concentration of from 0.03 to 3.0% carbon dioxide for 15 hours on each of 6 days; the 6-day exposure period was preceded and followed by 3-day control periods in room air. The maximum concentration of 3% was reached within 12-15 hours. The study was performed in a large pressure chamber with regulated temperature, ventilation, humidity, and carbon dioxide concentration. The tests of visual function included visual acuity, color and night vision sensitivities, amplitude of accommodation, and monitoring acuity. All were standard optical tests. The data showed no evidence of changes in overall visual acuity. The authors reported that color threshold sensitivity was poorer during the test period although no measure of this diminished capacity was cited. For comparison, Weitzman et al included conclusions in this report of a similar experiment at 1.5% carbon dioxide. They reported that no decrements in sensory function were found during exposure to 1.5% carbon dioxide. The only diminished sensitivity observed occurred on exposure to 3% of the gas; and, at this level, only color sensitivity was affected.

A 1949 report by Schaefer [50] concerned the effects of chronic exposure to 3% carbon dioxide on the psyche and excitatory processes of the peripheral nervous system. The experiment was conducted in an environmental chamber with the carbon dioxide level adjusted to 3% for a total of 8 days. The results indicated a radical change in the subjects'

behavior caused by the exposure. On the first day, the effect was stimulating, even to the extent that the subjects undertook solving complex mathematical problems and studying scientific texts. The author stated that they exhibited a degree of euphoria on this day. By the second day of the exposure, the subjects were exhausted, confused, inefficient, and inattentive; their behavior was erratic. By the third day, the confusion had subsided, although the author stated that the subjects still had not returned to normal, nor did they return to preexposure behavioral patterns throughout the remainder of the 8-day exposure. The subjects reported troubled sleep with frequent dreams and nightmares the first two nights. These results were evidence of an acute, excitatory phase followed by a chronic, depressive phase of effects of the exposure as identified by the author. The same phases were evident in analyses of consciousness, sensitivity, attentiveness, manual skills tests, and a measurement of nerve sensitivity or chronaxie (the shortest duration of an effective electrical stimulus having a strength equal to twice the minimal strength needed for excitation). Decreases in chronaxie are indicative of effects on nerve-muscle impulse transmission. In general, the excitatory effects were seen during the first day of exposure to 3% carbon dioxide. During continuous exposure at this concentration of the gas, a suppressive effect was observed which was accompanied by significant performance decrements in the subjects. However, although not referred to by the author, any behavioral effects resulting from prolonged confinement may also play a significant role in this study and cannot be divorced from the behavioral effects, if any, of carbon dioxide inhalation.

The study by Glatte et al [33] cited elsewhere also included psychomotor tests during a 5-day exposure to 3% carbon dioxide. The tests used in this study were completed three times daily. These tests included arithmetic, vigilance, hand steadiness, memory, problem solving, and auditory monitoring. A statistical analysis of variance indicated no significant trends, decrements, or changes from the control measurements. The authors concluded that, by all tests, no performance changes were detected.

An experiment reported by Bullard and Crise [51] concerned the effects of carbon dioxide on the shivering response. Six healthy men (mean age, 24 years) were exposed to 2.5-6% carbon dioxide from a Douglas bag via a Douglas respiratory valve for 75-minute periods while sitting in a room with an ambient temperature of 5 C. During the exposure period, recordings of electromyographs, ECG's, mean body temperature, and shivering were made. The shivering response was determined from observations, by each subject's perception of shivering, and by measuring the mechanical motions of each subject. Results indicated that, although shivering was totally suppressed on inhalation of 6% carbon dioxide in one-half of the subjects, slight shivering persisted in the others. After 30 minutes of 6% carbon dioxide, there was a breakthrough and shivering resumed in the subjects. The breakthrough shivering was more severe than at control levels. During the periods of inhalation of carbon dioxide, the subjects indicated that they felt warmer. These reactions are of importance because they relate to a worker's responses to cold stress during inhalation of carbon dioxide.

Evidence has been presented demonstrating that concentrations of up to 3% do not affect problem-solving ability, sequential reaction, and eye-

hand coordination. Two of the reports centered on the results of a single study involving only one subject, thereby limiting interpretation of the results. [34,49] Since both authors reported the effects on the same subject and each was concerned with a different aspect of the study, individual variations in response must be considered in any critical evaluation.

(e) Effects on Renal Dynamics

Investigators have also examined the relationship of renal circulation and electrolyte metabolism to inhalation of increased concentrations of carbon dioxide. Yonezawa [52] reported on experiments with 19 subjects, 13 of whom had renal disease while 6 were normal controls. Neither the method of administration of the gas nor the sex of the study subjects was given. Measurements of renal vascular resistance, venous pressure, plasma flow, blood flow, glomerular filtration rate, and filtration fraction were made before and during a 30-minute exposure to 5% carbon dioxide. Also measured were plasma sodium, potassium, chloride, and bicarbonate concentrations. Increases in carbon dioxide tensions in arterial and venous blood were accompanied by a corresponding decrease in pH. No changes were noted in sodium, potassium, or chloride concentrations, while bicarbonate concentration increased. Subjects with advanced renal disease showed decreases in arterial-venous differences of potassium and sodium, while bicarbonate arterial-venous differences increased. The normal subjects showed significant increases in renal plasma and blood flow. Those subjects with renal disease showed only slight changes. The same pattern was seen in the measurements of glomerular filtration. Decreased renal vascular resistance was seen in all

subjects, and increased renal venous pressure was observed in both groups. The author speculated that respiratory acidosis caused the vascular dilatation evidenced by increased renal blood flow and decreased renal vascular resistance.

Schaefer et al [53] investigated the acid-base and electrolyte responses at intermittently increased carbon dioxide concentrations. They reported doubling of urine excretion on the fourth and fifth days of 15 hours of exposure to carbon dioxide at a maximum concentration of 3%. The concentration attained this maximum only after increasing steadily for 12-15 hours each day. The increase in urine volume was accompanied by increases in organic acids, titratable acidity, ammonia, and hydrogen ion excretion. This was the same, single-subject experiment reported elsewhere. [34,49,41] The authors [53] also reported that the accumulated carbon dioxide was eliminated by renal mechanisms, as indicated by the increases in titratable acidity and excreted organic acids. They correlated this excretion with the increase in hydrogen ion excretion.

(f) Neuroendocrine Effects

The effects of carbon dioxide on neuroendocrine function were reported by Sechzer et al. [31] These investigators exposed 12 healthy men (aged 18-37 years) to 7-14% carbon dioxide for 10-20 minutes. The carbon dioxide in oxygen mixture was administered through a rubber mouthpiece; the nose was closed with a noseclip. The concentration of the test mixture of carbon dioxide and oxygen was determined from the end-expired carbon dioxide tension. When the carbon dioxide tension had reached the desired concentration, it was maintained for 10-20 minutes and then terminated. The total exposure period averaged 22 minutes (range 8-35 minutes).

Measurements were made of respiratory minute volume, arterial pressure, heart rate, and plasma concentrations of epinephrine, norepinephrine, and 17-OH-corticosteroids. The results indicated significant increases in plasma catecholamines and steroids for all levels of end-expired carbon dioxide greater than 50 mmHg (6%). The concentrations of epinephrine, norepinephrine, and 17-OH-corticosteroids were each statistically correlated with the increase in carbon dioxide. These increases were stated by the authors to be evidence of increased sympathoadrenal activity during exposure. The epinephrine and norepinephrine could have been responsible for some of the increased cardiac activity noted by these and other investigators. In addition to these findings, the authors also reported clinical symptoms experienced by their subjects as reported in subsection (a).

In a study already discussed, Schaefer [30,38] also investigated effects on neuroendocrine function. In this experiment, 42 healthy subjects were exposed to carbon dioxide at concentrations of 1.5, 3.3, 5.4, and 7.5% for 15 minutes each. Data on stimulation of the sympathetic nervous system were assessed; determinations included oxygen consumption, pulse rate, blood sugar, and eosinophil* count. Results indicated a decrease in the number of eosinophils and increases in blood sugar, oxygen consumption, and muscle potential above 3.3% carbon dioxide. Pulse rate was significantly increased at a 7.5% concentration of the gas. At 5.4% carbon dioxide, the greatest decrease in eosinophils was observed. These responses were less evident in the low-ventilatory-response group than in the high-ventilatory-response group, suggesting that the tolerance developed by these persons exists even with respect to autonomic response.

(g) Effects on Acid-Base and Electrolyte Balance

Sinclair et al [47] reported a 2-year series of related experiments designed to determine the rate and degree of acclimatization of men at rest at increased carbon dioxide levels. The investigators studied three groups of four healthy subjects each. The age and sex of the subjects were not specified. All experiments were performed in a chamber with an artificially created atmosphere and were preceded and followed by control periods of 2-5 days' duration. In the first two groups, exposure to 30 mmHg (3.9%) carbon dioxide was studied during 5- and 11-day periods, respectively. The third group was exposed to 21 mmHg (2.7%) carbon dioxide for 30 days. In all experiments, measurements of ventilation, acid-base indices of arterial blood, and lumbar CSF were made. Test data from the 5-day exposure revealed that the arterial pH reached its lowest level after 20 hours of exposure, at which time it was depressed by 0.025 units. This initial decrease was followed by a slow increase until the values reached normal levels again on the fifth day of exposure. The pH of the CSF reached its lowest level after 8 hours and nearly returned to normal by the end of the exposure. Parallel increases in arterial and CSF bicarbonate were seen and reached a peak on the fifth day of exposure. The CSF pH remained depressed after 2 days of the control period and paralleled a slightly elevated CSF carbon dioxide tension. Although the 11- and 30-day studies were conducted at different carbon dioxide concentrations, the subjects in both groups showed peaks in arterial carbon dioxide after 16 and 20 hours of exposure and after 16 and 20 hours of recovery.

The subjects generally tolerated test exposures well, apart from mild headaches and an awareness of increased ventilation. [47] The maximum

compensation for decreases in arterial pH required 5 days, while the reduction in ventilatory response occurred during the first 24 hours of exposure. The authors and technicians reported awareness of their own increased ventilation on entering the 30-mmHg (3.9%) environments for 2-8 hours daily; however, they experienced little difficulty in performing their tasks. Their awareness of increased ventilation subsided 1-2 hours after the exposure.

In 1961, Schaefer [54] addressed the question of what effects chronic exposure to carbon dioxide had on the the body's acid-base balance. He cited the experiment originally performed by Faucett and Newman, [35] in which 23 healthy subjects were confined in a submarine and exposed to carbon dioxide at a concentration of 1.5% for 42 days. The exposure period was scheduled between two 9-day control periods. In this report, Schaefer again based his observations on the major intervals of uncompensated and compensated respiratory acidosis. The 42-day exposure was divided into an initial 23-day uncompensated acidosis followed at days 24-42 by compensated acidosis. The graphic presentation of results indicated that the pH of the venous blood never dropped below 7.25 during uncompensated acidosis from an initial value of approximately 7.37 during uncompensated acidosis. The author concluded that the results indicated a combination of respiratory acidosis and metabolic alkalosis*. Schaefer identified renal compensation as the cause of alkalosis from the recorded pH and bicarbonate data. Relating the changes in basic physiologic parameters to observed pH changes is essential for understanding the significance of the pH difference. Plasma calcium was identified as a pH-dependent variable, while plasma inorganic phosphorus, increased tidal volume, and decreased respiratory

rate were identified as independent of pH. Subsequent experimental results indicated that the use of buffers to ameliorate conditions caused by the acidosis were not adequate since several of these physiologic adaptations were pH independent.

In 1963, in another report of data from the same study, [35] Schaefer [55] described additional pH findings. He reported that the concentrations of chloride in red blood cells and plasma were also identified as pH independent, while plasma calcium levels showed an initial decrease with a subsequent rise mirroring the compensation of acidosis. In yet another report of the same study, Schaefer et al [56] provided more data and a clearer basis for the previously cited conclusions. They indicated that the inorganic phosphorus concentration also indicated a mirroring of the pH changes occurring in the body. Additional corroboration of the pH dependence of plasma calcium was presented.

Schaefer et al, [57] in 1964, reported further findings on the acid-base balance and electrolyte changes caused by exposure to carbon dioxide. The 42-day experiment with 23 healthy subjects was again the basis of these findings. Acid-base studies were completed on 20 of the 23 subjects. The experimental procedures have been described earlier. [35,36,54] The acid-base and electrolyte studies identified the initial acidotic response of the body and the subsequent mechanisms required to achieve compensation. Venous blood samples were drawn from 10 of the subjects for determination of a total electrolyte profile in blood; similar profiles were completed on urine and feces samples. Also measured were electrolyte concentration and bicarbonate levels in plasma and red blood cells. Corrections for body temperature and electrolyte intake were made. The measurements indicated

the various metabolic patterns which occurred during the phases of uncompensated and compensated respiratory acidosis. The phase of uncompensated acidosis (days 1-23) was characterized by a decrease of 0.06 units in extracellular pH, decreases in urine pH, bicarbonate excretion, and pulmonary carbon dioxide excretion. The pulmonary carbon dioxide excretion data were taken from a previously cited report [33] of the same study. The days of compensated acidosis (days 24-42) were characterized by a return of blood pH to normal levels, a rise in urinary pH, and increases in bicarbonate and pulmonary carbon dioxide excretion. The plasma electrolytes did not appear to follow these phases, as red blood cell sodium increased and cell potassium decreased throughout the exposure period and during the 9 days of recovery. Pulmonary carbon dioxide excretion peaked on the first day of recovery, while urinary excretion of carbon dioxide followed on the second. Another peak in pulmonary excretion on the ninth day was considered indicative of the equilibration of the slow bone-calcium store of carbon dioxide. The authors, [57] therefore, identified the bones as having the slowest carbon dioxide release rate. While the pH changes set the pattern for uncompensated and compensated acidosis, the carbon dioxide tension remained elevated (5 mmHg) throughout the experimental period and during the 9-day recovery period.

In 1967, Glatte et al [33] reported additional information on the effects of prolonged exposure to 3% carbon dioxide. Seven healthy men were exposed for 5 days in a space-cabin simulator. Five control days preceded the study, and 5 recovery days followed it. Full medical histories were taken, and blood and urine chemical determinations were made. Additionally, tests of liver and renal functions and hematologic studies

were performed. Using these baseline data, the investigators monitored changes developing during and after the experiment. The subjects were divided into two groups; the time of administration of the carbon dioxide was not revealed to the subjects. Diet and exercise were controlled by the study design. Psychomotor testing was repeated daily. In contrast to previously cited studies, [35,36] these authors [33] reported no significant physiologic differences between the test and control groups. Experimental data did show a mean decrease from 7.40 to 7.37 in blood pH after the initial 2 days of exposure. One subject experienced a decrease in blood pH from 7.40 to 7.35 during this period. Results also indicated increased arterial carbon dioxide tension of approximately 3-4 mmHg (0.39-0.53%) and increased serum bicarbonate with a mean increase of 1.5 mEq/liter. This was accompanied by a slight decrease over the test period in serum chloride, with a mean of approximately 3 mEq/liter. The authors reported no changes in renal ammonia and titratable acidity. The authors also reported no significant changes in serum electrolytes, blood sugar, BUN, serum creatinine, or liver function tests. They did report modest reductions in blood cell count which they attributed to the routine drawing of blood for test samples. Pulmonary function was normal. The authors concluded that, in general, the 3% carbon dioxide represented only a mild challenge, although four of the seven subjects reported frontal headaches during the first 2 days of exposure. The authors noted further that the study period was too short for an adequate assessment of changes in calcium metabolism. The results of respiratory studies indicated that the ratio of carbon dioxide production to oxygen consumed decreased from approximately 0.85 to 0.74. The ratio was reported to have returned to 0.85 upon return

to normal air. The investigators recommended further research to elaborate on this observed change. A study of such short duration generally would not be expected to yield sufficient data to define the parameters of physiologic response at slightly increased carbon dioxide levels.

Acid-base changes due to intermittent exposures to carbon dioxide were reported by Schaefer et al. [53] This experiment involved one healthy man who was exposed at an increasing carbon dioxide concentration of from 0.03 to 3% over a 15-hour daily period. He spent 9 hours/day in normal air, and the experiment continued for 6 days with 3 control days both before and after the study. The study was conducted in a large, pressure altitude chamber. Data were recorded on arterial and venous pH values, carbon dioxide and oxygen concentrations, magnesium, sodium, potassium, chloride, and calcium and phosphorus levels, as well as on blood lactate and pyruvate concentrations. Urine studies included volume; pH; calcium, magnesium, and phosphate concentrations; carbon dioxide, organic acids, ammonia, and titratable acidity levels. The results of this study indicated an immediate response to the carbon dioxide; urine volume nearly doubled, and excretion of organic acids, titratable acidity, and ammonia increased on the first day. These measurements declined on the following 2 days. On the fourth and fifth days, carbon dioxide excretion and acid load increased markedly during the air-breathing period. Urine volume rose again to a level greater than double that in the control period; this rise was accompanied by increases in organic acids, titratable acidity, ammonia, and hydrogen ion excretion, as reported in subsection (e), Effects on Renal Dynamics. Blood lactate and pyruvate levels were not affected by the exposure. By the second day of recovery, excretion of ammonia, titratable

acidity, and hydrogen ions decreased as elimination of bicarbonate greatly increased. Again, this is the same subject as previously documented, [34,41,49] and data from a single individual seriously limit the acceptability of these results.

The effects of a continuous exposure to 3% carbon dioxide were described by Schaefer. [58] In this study, healthy young men were exposed to the gas at a constant concentration of 3% for up to 8 days in an environmental chamber. Results indicated that after 3 days of exposure, a tolerance or adaptation of the respiratory center was evident. An increase in the respiratory threshold was observed both at rest and during exercise studies. Acid-base regulation was studied through measurement of blood bicarbonate, pH, serum and hemoglobin protein, alveolar carbon dioxide, and excretion of carbon dioxide and bicarbonate. Analyses of these studies showed that immediate responses to increased carbon dioxide were rises in dissolved carbon dioxide and bicarbonate in the blood. As the exposure period lengthened, renal acid excretion increased, alkali was retained, and bicarbonate content of the blood increased. This paralleled the compensated phase of respiratory acidosis. In the same report, the author cited results of a study in which a continuous exposure to carbon dioxide was interrupted 8 hours daily by breathing room air. In contrast to the continuous exposure, this intermittent study showed that the return to normal air was of insufficient duration to alter the developed patterns of compensation of the blood buffer system. The bicarbonate levels were unaffected by these interruptions. Data on carbon dioxide elimination indicated that the 8-hour return to normal air was not sufficient to rid the tissues of accumulated carbon dioxide, nor was it sufficient to make

alkali available for carbon dioxide elimination. This study demonstrated the length of time required for compensation to increased carbon dioxide levels and the time required for a full return to normal. Further, it showed that as much as 8 hours daily of breathing normal air is insufficient for the acid-base regulatory mechanisms to alter the readjusted homeostatic responses. This portion of the experiment, as described by the author, appeared to have been begun after several days of continuous exposure to 3% carbon dioxide. As a result, the observed inability of the buffer systems to return to normal may well have been merely the buffer systems' slow return to normal after chronic exposure, rather than the true effects of an intermittent exposure.

A report by Zharov et al [59] dealt with the psychologic, performance, and physiologic effects of continuous exposure to 1 and 2% carbon dioxide. The studies were performed to investigate problems of aviation medicine where intolerable accumulations of any substance are critical. In two experiments, two subjects each were confined to a 7-cu m pressure chamber for 30 days. The first pair was exposed to 1% carbon dioxide, the second to 2%. Evidence collected included total carbon dioxide content of the blood; pH and pCO₂ of blood; pCO₂ and pO₂ of alveolar air; urinary pH, ammonia, carbonates, titratable acidity; ECG; and arterial blood pressure. Results of the trials indicated that there was respiratory acidosis as evidenced by slight drops in blood pH, from 7.41 and 7.47 to 7.34 and 7.38, respectively, at 2% carbon dioxide. Increases in pCO₂ in blood and alveolar air were also recorded. Details of exercise regimes and the subjects' performances were not reported. The authors indicated that efficiency and functioning were measured by

electroencephalograph recordings (EEG's) and conditioned motor reflex tests. They also indicated that the subjects' efficiency in performing physical exercise remained high at 1%, but deteriorated after prolonged exposure to 2% carbon dioxide. This, the authors noted, was accompanied by psychologic tension. Further, the authors stated that all changes seen in the EEG's and conditioned motor reflexes as a result of carbon dioxide exposure were wholly reversible.

Messier et al [60] reported the effects on human calcium, magnesium, and phosphorus metabolism of continuous exposure in submarines to 0.8-1.2% carbon dioxide. A total of 31 men from 3 submarine patrols were studied. The first two patrols lasted 57 days, the third lasted only 3 weeks. During all patrols, the oxygen concentration was between 19 and 21%, and carbon monoxide was less than 25 ppm. Blood samples taken 9 months after the last patrol served as the controls. Mild respiratory acidosis was present as evidenced by decreased pH and increased pCO₂ and bicarbonate in the fourth week of the two long patrols. Additionally, decreased plasma chloride and increased red blood cell chloride confirmed this acidosis. By day 51, compensation was complete, and all of the measured indicators had returned to normal. Plasma electrolyte studies showed decreased potassium, immediately increased sodium, and decreased calcium. Plasma phosphorus levels were not affected, and plasma magnesium increased only on day 51. Red blood cell sodium and calcium increased, while potassium decreased. On day 51, red blood cell magnesium decreased. Renal regulation of acidosis was not apparent until after the third week since urinary pH was elevated throughout the patrol and urine calcium, phosphorus, and magnesium were decreased. Urine bicarbonate was generally elevated throughout the

exposures. Parathyroid regulation also was not apparent as parathyroid hormone and calcitonin levels did not change during the exposure. The decreased urinary calcium excretion did rise during the fourth and fifth weeks; however, it did not exceed control levels. It decreased again during the sixth-ninth weeks indicating a three-phased pattern of urinary calcium excretion. The decreased urinary calcium excretion was associated with decreased plasma calcium levels. The authors [60] concluded that this was evidence of hypocalcemia and hypocalciuria in contrast to hypercalcemia and hypercalciuria which are associated with exposure at higher levels of carbon dioxide. Also, the lowered blood bicarbonate levels during the first 3 weeks contrasted with increased blood bicarbonate levels found upon continuous exposure at 1.5% of the gas. [57] The authors suggested that the renal regulation of acid-base balance was responsible for increased blood bicarbonate at 1.5% carbon dioxide and was not active for the first 3 weeks at the lower concentrations (0.8-1.2%) which accounts for the decreased blood bicarbonate observed in this study. The authors [60] concluded that bone buffering replaced renal regulation during the first 3 weeks of continuous exposure at 0.8-1.2% carbon dioxide. By the fourth week, the bone store reached saturation and was dumped, raising pH, pCO₂, and blood bicarbonate. At this time, renal regulation became active in acid-base balance maintenance. The decreased urinary calcium excretion, blood bicarbonate concentration, and hypocalciuria of weeks 6-9 were associated with renewed storage of carbon dioxide in bones.

The most noticeable effect of increased carbon dioxide on the acid-base balance of the body is the acidosis caused by such exposure. As previously discussed, the body is equipped to compensate for challenges to

pH stability. Studies on humans have indicated that compensation for acidosis caused by continuous exposure to 1.5% carbon dioxide for 42 days will take approximately 23 days. The findings during the uncompensated period included a decreased physiologic pH, lowered plasma calcium and inorganic phosphorus, decreased pulmonary excretion of carbon dioxide, and decreased bicarbonate excretion. The compensated phase (days 24-42) was characterized by a return of the blood pH to normal, a rise in urinary pH, increases in pulmonary carbon dioxide and bicarbonate excretion, and increases in plasma calcium and inorganic phosphorus. It is also important that during a 5-day exposure to 3% carbon dioxide, the authors [33] stated that the changes were not indicative of a serious challenge. A pH drop accompanied by an increase in the serum bicarbonate level was observed within the first 2 days. Although some aspects of this study were hampered by its short duration, the data did indicate that the body apparently compensated more readily to the higher carbon dioxide level. An immediate bodily response to 3% carbon dioxide was also indicated in the study on intermittent exposure.

In contrast, continuous exposure to 0.8-1.2% carbon dioxide [60] indicated that renal compensation was not apparent until after the third week of exposure when carbon dioxide was dumped from bone stores thus increasing the concentration sufficiently to trigger compensatory mechanisms.

(h) Effects During Exercise

The effects of simultaneous exercise and hypercapnia were the object of experiments reported by Menn et al. [61,62] Eight healthy men (aged 18-21 years) were exposed to carbon dioxide at levels of 8, 15, 21, and 30

mmHg (1, 2, 2.8, and 3.9%) for 30 minutes while performing both steady-state and maximum exertion tests in an upright position on a bicycle ergometer. The subjects had undergone a 14-day training period involving daily increased workloads. The workload was reported in Watts (W), 1 W being equivalent to 0.01433 kcal/minute. The beginning load was 50 W (0.72 kcal/minute); the maximum attained was 250 W (3.58 kcal/minute). The study was performed in an environmental chamber with controlled carbon dioxide, temperature, and humidity. Respiratory rate, expired and inspired minute volume, rectal temperature, ECG, cardiac rate, blood pressure, arterial oxygen, and carbon dioxide tension were measured. All eight subjects completed all regimes at every carbon dioxide exposure level. The maximum exertion test was not performed at 3.9% carbon dioxide. No difficulty was reported at carbon dioxide levels below 21 mmHg (2.8%). At or beyond this level, the subjects reported respiratory symptoms during exercise performed at two-thirds maximum and at maximum levels. The two-thirds maximum level refers to that exercise level at which two-thirds of the maximum oxygen consumption was obtained during the pretest maximum exertion test. It was also equivalent to an average of 180 W (40 W is considered a moderate workload). Two subjects reported intercostal muscle pain from large ventilations at 2.8% of inspired carbon dioxide. Three others reported that respiratory difficulties had impaired their performance at the same carbon dioxide level. At the 3.9% carbon dioxide level, six subjects reported mild-to-moderate frontal headaches. The headaches generally occurred near the end of the exercise period and were not severe enough to interfere with the subjects' performances. Increased arterial carbon dioxide tension was linear and related to inspired carbon dioxide levels.

EKG changes in four subjects showed premature auricular and ventricular contractions, which according to the authors [61,62] were abnormalities also commonly associated with exercise in room atmosphere. Five of the nine incidents of ectopic foci occurred in only one of the subjects. Further, acid-base studies showed a greater reduction in blood pH values during exercise than at rest at all levels of carbon dioxide exposure. The authors concluded that exercise combined with hypercapnia does stress the respiratory system. They also stated that the premature ventricular contractions appeared to be related to neither the level of exercise nor the inspired carbon dioxide levels. Because subjects were able to complete all phases of the study regimen, apparently humans can perform strenuous work for short periods without obvious stress, even at high carbon dioxide levels.

In an exercise study, Craig et al [63] made use of inspiratory and expiratory resistances and carbon dioxide exposure to measure treadmill exhaustion limitations. Thirteen healthy men were tested on treadmill grades up to 22% with combinations of inspiratory resistances from 1.5 to 15.5 cm H₂O/liter/second, expiratory resistances of 2.0 and 3.9 cm H₂O/liter/second, and carbon dioxide levels of 3.1-3.9%. Both types of resistance and the carbon dioxide were regulated through a facepiece. Measurements included tidal volume, heart rate, and inspiratory flow, as well as expired carbon dioxide level. Exhaustion was the point at which the subject decided to end the routine. After 110 tests, results indicated that, at the minimal inspiratory and expiratory resistance levels, only concentrations in excess of 3% consistently resulted in reduced endurance. At maximum inspiratory and minimum expiratory resistance, the range of 3.1-

3.9% carbon dioxide levels did not appreciably change the subjects' endurance compared with that when the subjects breathed air. However, at levels above 4% carbon dioxide, the inhaled gas had a greater effect than the same resistance combination. While data from each carbon dioxide level were not enumerated, the grouped carbon dioxide exposure data at minimal resistance indicated that exhaustion was reached after 413 seconds at the combined minimal resistance levels. Time to exhaustion decreased from 567 seconds during air breathing to 413 seconds during exposure to carbon dioxide, while minute volume increased from 86 to 105 liters/minute, and tidal volume increased from 2.05 to 2.51 liters.

A study by Sinclair et al [64] focused on the physiologic response of the body during exercise to the inhalation of increased carbon dioxide concentrations. Four healthy men performed three levels of work (low, moderate, and heavy) in a chamber having a carbon dioxide concentration of 21 mmHg (approximately 2.8%). No equivalence in watts or oxygen consumption data were given by the authors, who used heart rate as a measure of workload. Measurements were done in air after 1 hour of exposure to the carbon dioxide (acute) and during chronic (15-20 days) exposure to the gas. The individual exercise regimes, performed twice daily, were 45 minutes long, separated by 5-hour rest periods. The exercises were performed in a supine position on a bicycle ergometer, with measurements recorded between the 12th and 15th minute of exercise. The authors found some changes in normal responses to exercise as a result of the stress-inducing increases in carbon dioxide exposures. Arterial pH was lowered proportionally with increasing carbon dioxide exposure and exercise stress. Another modified response was an increased retention of the carbon

dioxide; this, however, was anticipated because of the body's decreased ability to eliminate the excess gas as the concentration of carbon dioxide in the inspired air was increased. Exercise compounded the problem. The authors suggested that the cause was insufficient compensation for the diminished effectiveness in carbon dioxide elimination. A lesser degree of metabolic acidosis, also noted as workload increased during exposure to carbon dioxide, reflected a decreased production of organic acids according to the authors. They reported little difference between the acute and the chronic situations, although detailed data were not presented. They concluded that carbon dioxide at tensions up to 21 mmHg (2.8%) could be tolerated by normal subjects, both at rest and during strenuous, steady-state exercise. The authors [64] reported the absence of cardiac abnormalities and contrasted their results with those of Menn et al. [61,62]

An exercise regimen in the previously cited paper by Glatte et al [33] offered similar data on the ability of six healthy subjects to tolerate work during exposure to 3% carbon dioxide. The 5-day exposure, as well as the 5-day preexposure and postexposure control periods, included a steady, moderate exercise program involving two routines daily. Each routine was a 1-hour session on a bicycle ergometer with a 100-W load or 1.43 kcal/minute, the equivalent of a heavy workload. In comparison, 105-130 W would be similar to maximum exertion or extremely strenuous work not generally encountered in industry except for very short times. The two exercise routines were performed so that the first was on the first day, representing the acute period, and the last was on the last day, representing the chronic phase. Results indicated that the 1-hour exercise

session was tolerated well by the subjects. In control periods, exercise produced a marked increase in ventilation averaging from 12.5 to 40.6 liters/minute while, during exposure to carbon dioxide, ventilation increased from an average of 18.3 to 67 liters/minute. Pulse rate increased to a maximum of 152 beats/minute during the chronic exposure exercise period from a resting mean of 73 beats/minute. The maximum attained during exercise in the control period was 145 beats/minute from a resting mean of 69 beats/minute.

In a previously cited publication, Schaefer [58] also described an exercise study in which healthy subjects were exposed to 3% carbon dioxide for up to 8 days. He found that, at the same inhaled carbon dioxide level, alveolar carbon dioxide tension was higher during work after adaptation than it was during rest. The depth of breathing was reported to have decreased and the rate of respiration to have increased while the subjects worked during chronic exposure. This results in increased oxygen intake and carbon dioxide excretion. Adaptation to the increased carbon dioxide level was apparent by decreased ventilation during work as well as during resting; however, the work levels never decreased to the extent that the resting levels did. The author offered no information on the subjects' ability to tolerate the work plus increased carbon dioxide atmosphere. All work was performed at 40 W (0.57 kcal/minute) which would be equivalent to a moderate work situation where frequent breaks are essential. No untoward effects were reported at this work level.

Clark et al [65] elucidated further the effects of a wide range of carbon dioxide exposures when combined with a full exercise regime. The regime consisted of walking or running on a treadmill on a 10% grade at

speeds of 1.8, 3.4, 4.8, and 6.0 mph for 6 minutes at each speed and without the treadmill stopping between speed adjustments. This 24-minute run of 1.6 miles was preceded and followed by resting control periods spent standing on the treadmill. The study was conducted in a 3,500-cu ft environmental chamber providing controlled temperature and carbon dioxide levels. The subjects were nine healthy, young Air Force members who had just completed basic training and who also had participated in a 12-week prestudy exercise conditioning program. During the experiment, the subjects exhaled into a valved mouthpiece connected to a spirometer for air analysis. They performed the exercise regime at the following inspired carbon dioxide levels: 10, 20, 30, and 40 mmHg (1.3, 2.6, 3.9, and 5.2%) and a control at room air. Even at the highest level of carbon dioxide (equivalent to about 5.2%) there was no change in oxygen consumption. Significant changes were seen in ventilatory response indices. Increases were seen in the volume of expired gas from a mean value of 10.13 ± 1.84 liters/minute at rest with no inspired carbon dioxide to a maximum of 169.06 ± 16.68 liters/minute at a maximum treadmill speed of 6.0 mph and at 40 mmHg of inspired gas. The measurements of acid-base balance confirmed the additive effects of respiratory acidosis caused by hypercapnia and of metabolic acidosis caused by exercise. The arterial pH, which dropped from about 7.41 at rest with no inspired carbon dioxide to 7.13 at a maximum treadmill speed and at 40 mmHg (5.2%) of inspired gas, was associated with a ninefold rise in arterial lactic acid. A twofold rise in blood lactic acid occurred between 4.8 and 6.0 mph during the experiment. The subjects experienced significant symptoms during the study, including the collapse of three subjects at maximum exercise and inspired pCO₂, mental confusion,

impaired vision (both central and peripheral), and severe headache (in the postexercise period). Severe headache, dyspnea, and impaired central and peripheral vision were also reported during a special, single-subject portion of the study conducted at a treadmill speed of 6.5 mph and at 5.2% carbon dioxide for 8 minutes. The authors also observed that initial exposure at the highest carbon dioxide concentrations (3.9% and 5.2%) caused the most severe symptoms. Since all subjects were able to run at 8.0 mph during air breathing and yet three collapsed at the highest pCO₂ while running at 6.0 mph, the authors surmised that changes in the intensity of either stressor would cause a reciprocal change in the subjects' ability to tolerate the other.

Luft et al [66] investigated the effects of exposure to 15 mmHg (1.9%) carbon dioxide combined with exercise. The study was divided into two parts. The first concerned exercise tolerance by 12 healthy men (mean age 26.5 years). The subjects exercised on a bicycle ergometer at 50 rpm with an initial brakeload of 300 kpm/minute (equivalent to 49 W) and subsequent brakeload increases of 75 kpm/minute (12.2 W) until the subjects could no longer maintain the pedaling rhythm. The initial resistance was maintained for 3 minutes with increases applied each minute thereafter. The exercise regimes were repeated in air and at an inspired carbon dioxide tension of 15 ± 2 mmHg (1.9%). Consequently, each subject was his own control. At each submaximal work load, the mean heart rate and systolic blood pressure were slightly higher during exposure to carbon dioxide; however, the increase was not significant. The heart rate was lower than control at the end point. The ventilatory measurements indicated that increased carbon dioxide resulted in significantly increased

ventilation at submaximal exercise levels. The increases were 40-50% above controls at all workloads except maximum, where the increase was only 2%. Other measurements included mean oxygen consumption and carbon dioxide elimination. The oxygen consumption was higher (though not significantly) at submaximal exercise and at increased carbon dioxide, and significantly lower (-13%) at maximum exercise with carbon dioxide. In contrast, carbon dioxide elimination was lower during carbon dioxide exposure than during the control period.

In the second part of the study, [66] 10 healthy men completed the same exercise routine and the carbon dioxide exposure level was maintained. In this series, however, respiratory measurements were made prior to exercise, during maximum exercising, and during recovery. These observations showed lower oxygen consumption during the last 2 minutes of exercise and during the first minute of recovery than during carbon dioxide exposure. The effects on acid-base balance were evident in decreased arterial pCO₂ during control exercising. This decrease lasted through the fourth minute of recovery. In contrast, during carbon dioxide exposure, the arterial pCO₂ rose during the final minute of exercise and dropped only slightly during the first minute of recovery. Bicarbonate which also fell during the control exercise, was correlated by the authors with the buildup of blood acid metabolites. Blood electrolyte levels generally increased, peaking the last minute of exercise. The increases in potassium and phosphorus were nearly 60%. The authors concluded that the carbon dioxide concentration of 1.9% was sufficient to affect an individual's exercise capacity by altering the physiologic elimination of carbon dioxide. The metabolic acidosis caused by maximum exercise was not reduced by

ventilation in the presence of 1.9% carbon dioxide which was already burdening the ventilatory process.

(i) Case Reports

An article [67] in Industrial Medicine and Surgery reported on characteristics of deaths in silos during each harvest season. The article, which appeared in 1953, remarked on the "unnecessary, inexcusable" deaths of farmers overcome by high carbon dioxide concentrations while stamping down silage. It stated that it was not unusual for the concentration of carbon dioxide to reach 38%. The gas is evolved as carbohydrates in the stored crops undergo atmospheric oxidation. The author reported that, although the carbon dioxide concentration was extremely high, the rapid deaths were caused by the displacement of oxygen, and therefore, death was due to asphyxiation. This article established that deaths of this nature had been reported since 1914.

Troisi [68] cited three deaths from carbon dioxide poisoning. Two youths, aged 18 and 12, collapsed after descending into a silo filled with green forage beans, barley, and oats to remove silage. A 61-year-old man who went to their rescue later succumbed. All three received artificial respiration; the youths were pronounced dead at the scene and the man was hospitalized. The man was reportedly "in extremis," the only vital signs being occasional cardiac contractions. Although his respiration was restored about an hour after administration of cardiac and respiratory stimulants, the patient died on the fifth day without having regained consciousness. The clinical picture was that of asphyxia, although observed toxic effects on the thermoregulatory center were reportedly due to carbon dioxide. The man had exhibited a progressive rise in temperature

which could not be attributed to any inflammatory condition. Although no measurements were made, the carbon dioxide concentration must have been high. In addition, other noxious gases such as nitrogen dioxide were probably present.

In an incident described by Williams, [69] two deaths were attributed to carbon dioxide poisoning aboard a ship carrying onions and crude brown sugar. Eight persons, including the ship's doctor, entered the hold in single order to assist the persons entering immediately before. Of the total, two died immediately; two had major symptoms; and four had only minor symptoms. The time each had spent in the hold varied from 5 to 15 minutes. The doctor and the first man to enter the hold had pulse rates of 100 and 156, respectively; they did not show signs of cyanosis but did show evidence of congested conjunctivae. The two who died were not autopsied. External examinations revealed no outward signs of injury, but both men had markedly congested conjunctivae and cyanosis of the buccal mucosa as well as of the toe and fingernail beds. The deaths were attributed to carbon dioxide poisoning. Although no estimation of the concentration of carbon dioxide in the hold was obtained, the authors surmised that it had reached 25-30%. The observed signs and symptoms, such as headache, giddiness, tinnitus, and loss of muscular power, were consistent with other descriptions of carbon dioxide poisoning. The authors offered a twofold explanation for the action of the gas: first, that of oxygen displacement, in which case carbon dioxide acts as a simple asphyxiant; secondly, as a respiratory stimulant and a narcotic.

Another account of deaths and poisonings occurring in ships' holds was reported by Dalgaard et al. [70] These authors described three deaths

and four cases of nonfatal poisonings arising as a result of working in ships' holds filled with fresh or putrefying trash fish or both. Each of the deaths was attributed to hypoxia* alone or to hypoxia compounded by carbon dioxide and alcohol. In one case, air samples were taken; the carbon dioxide concentrations were 20 and 22% at the middle and bottom of the hold, respectively. Signs and symptoms among the survivors included unconsciousness, cyanosis, sluggish reflexes, rattling respiration, and excessive motor unrest. In these cases, carbon dioxide was produced from the putrefaction of fish in sufficient quantity to have displaced oxygen, causing the hypoxia. However, at carbon dioxide concentrations as high as 20%, the direct toxic effects of this gas undoubtedly had some impact on the resultant signs and symptoms.

Nuttall [71,72] wrote a brief account of the toxic effects of carbon dioxide. The author identified two instances of near collapse of aircrew members on different flights from inhalation of carbon dioxide gas which had vaporized from dry ice during transport of frozen foods. The author further stated that one of the hazards of the gas was the rapidity of the onset of its effects and cautioned that the gas was more than a simple asphyxiant. He presented no environmental sampling data, although he did report that the aircraft were transporting 12,000 and 7,400 pounds of dry ice, respectively.

The cerebro-ocular effects of carbon dioxide poisoning were detailed in two reports [73,74] involving seven workers in deep artesian wells. Freedman and Sevel [73] reported that three of four men who entered a well died immediately; the fourth was found in a comatose state by a rescue party 10 minutes after he had entered. The authors gave no account of the

autopsy findings nor of the cited cause of death for the first three victims. The man found in a coma was hospitalized and remained comatose and completely unresponsive until his death 11 months later. An EEG recorded 6 months after hospital admission had shown arrhythmic activity of very low voltage, which was unchanged either when the patient's eyes were held closed or when he was challenged with painful stimuli. The autopsy report was limited to findings on the brain and eye. Major retinal abnormalities included the absence of ganglion cells, atrophy of the nuclear layer, and absence of endothelial cells in the retinal digest. The brain showed severe atrophy, especially in the posterior two-thirds of the hemispheres. The nonselective, widespread damage and the severity of the individual lesions of the brain contrasted with the expected findings from death caused by anoxia. For these reasons, the authors concluded that these lesions were due to the histotoxic effects of carbon dioxide. The effects on the eye were considered similar to those caused by anoxia. The death of this victim was reportedly due to asphyxiation by carbon dioxide.

The second incident, described by Sevel and Freedman, [74] involved three men, one of whom died immediately upon entering the well. The symptoms and clinical findings of the other two men were presented. Both patients experienced throbbing headaches, attacks of vertigo, poor memory, poor ability to concentrate, photophobia, difficulty sleeping, tinnitus, and diplopia (double vision). The patient exposed for the longest time also underwent a marked personality change and suffered from loss of eye movement as well as from visual field defects, enlargement of blind spots, and deficient dark adaptation. Both patients were diagnosed as suffering from carbon dioxide asphyxiation. Neither report gave any indication of

the level of carbon dioxide in the wells.

Duchrow [75] reported an incident of mass carbon dioxide poisoning in a potassium mine where a particularly large carbon dioxide gas bubble erupted. It occurred at a time when 90 miners were potentially exposed to the gas; 12 lost consciousness and 6 of these died as a result. The author neither estimated the concentration of the gas in the mine, nor described the signs and symptoms of the poisoned miners.

In 1973, a report [76] was published about 50 firefighters who were overcome by carbon dioxide while fighting a blaze. The report indicated that the combustion of plastics and acrylics produced high concentrations of carbon dioxide. No further details were given.

Epidemiologic Study

One epidemiologic study [77] with brewery workers has been reported. The exposed group was composed of 19 men (average age 38) who worked in the brewery cellars (fermentation rooms); 20 men (average age 50) in the control group worked in the bottling department and thus were presumed not exposed to carbon dioxide greater than ambient since, measurements of carbon dioxide levels were not made in this area. The effect of carbon dioxide exposure was monitored by determination of the blood standard bicarbonate level. Bicarbonate determinations for both groups were done before and after work on a Monday and after work on a Friday. There was no appreciable difference between the two groups in standard bicarbonate levels. The test and control groups both averaged 24.1 mEq/liter on Monday; after work on Friday, the bicarbonate levels were 22.8 mEq/liter and 22.2 mEq/liter, respectively. The authors gave neither an explanation

for the decrease in average bicarbonate levels nor data about the previous exposure histories of either group.

Ambient carbon dioxide concentrations in the brewery [77] were measured for all work shifts on an infrared spectrophotometer which ran continuously. A total of 3 shifts/day for 5 days were monitored. The collection hose was placed within the worker's breathing zone. The mean TWA concentration for all 15 shifts was 1.08% carbon dioxide with excursions up to 8% for 3 minutes, the highest TWA concentration during any shift was 1.95% and the lowest was 0.5% carbon dioxide. The authors concluded that there was no appreciable physiologic effect of chronic intermittent exposures at these levels. Since this is the only epidemiologic study found, these results cannot be effectively correlated with experimental studies which have reported other acid-base, electrolyte, cardiovascular, and behavioral changes.

Animal Toxicity

The following animal experiments have been selected to minimize duplicate reporting of results similar to those obtained from human studies. Included in this section are those studies which offer additional evidence as to the mechanism of action of carbon dioxide on the organism, or include chemical manipulations as indicators of mechanistic actions not possible to demonstrate in man.

(a) Effects on the Central Nervous System

An investigation into the effects of carbon dioxide on the cortical and subcortical areas of the brain in monkeys was reported by Schaefer.

[30] Eight monkeys (*Macaca mulatta*) were implanted with multilead electrodes in the cortical (cortex to hypothalamus) and subcortical (reticular formation) areas; two of the eight also had electrodes implanted on the surfaces of the anterior and posterior lobes. The monkeys were exposed to carbon dioxide at concentrations of from 0 to 30%, during which time electrical activity was recorded. A 10-minute exposure at 30% of the gas showed stimulation of the cortex. A more consistent finding was increased activity in the hypothalamus, which appeared in the first 5 minutes and was maintained for the entire 30-minute exposure. Upon exposure to air, this excitation disappeared rapidly; however, subcortical activity was still present. As the carbon dioxide concentration was increased, a depressant effect of carbon dioxide developed, as seen by increasing thresholds to electrical stimulation in the motor cortex. A dissociation of effects was noted since the posterior hypothalamus exhibited decreased thresholds indicative of increased excitability in this area.

The effects of carbon dioxide on CSF pressure were studied by Small et al. [78] They exposed five adult mongrel dogs to 5, 10, and 15% carbon dioxide in oxygen. During measurements of CSF pressure, they recorded central venous and arterial blood pressures. They found that CSF pressure rose with the increasing concentration of carbon dioxide, yet no relationship was observed between changes in the central venous and arterial blood pressures and the rise in CSF pressure. Further, the effects on cerebral vasculature were greater than the effects produced in cardiovascular and respiratory functions. The authors concluded that chronic hypercapnia might be accompanied by increased intracranial

pressure. The authors noted that normal pressures were regained immediately when carbon dioxide exposure was stopped.

(b) Effects on Respiratory Function

Niemoeller and Schaefer [79] studied the effects of chronic respiratory acidoses on the development of hyaline membranes and atelectases in male Hartley-strain guinea pigs and male albino Harvard Biological Laboratories colony rats exposed at different concentrations of carbon dioxide during prolonged continuous exposures. Hyaline membrane formation was associated with a loss of surfactant, which lessened surface tension* and led to decreased lung stability. The hyaline material is associated with respiratory distress diseases. The exposure time was divided into periods of uncompensated and compensated respiratory acidosis. The uncompensated period was seen to last 23-28 days with 1.5% carbon dioxide, 4-5 days with exposure at 3%, and only 2 days with 15% of the gas. Microscopic examination indicated that guinea pigs exposed to 3 and 15% carbon dioxide developed hyaline membranes, while those exposed at 1.5% did not. The greatest incidence was observed during uncompensated acidosis. The incidence of hyaline membranes was 100% in guinea pigs during uncompensated acidosis and declined to 0% during the compensated phase (days 14-93) in guinea pigs exposed to 15% carbon dioxide. Atelectasis was concomitantly observed in guinea pigs. However, it also developed upon exposure to 1.5% carbon dioxide during compensated and uncompensated respiratory acidosis. The authors noted that the hyaline membranes and atelectases were both located in the subpleural areas. In contrast, rats exposed to 1.5 and 3% carbon dioxide did not show evidence of hyaline membrane formation. Decreases in both occurrences were observed in guinea

pigs during compensated acidosis. Even when the blood carbon dioxide tension was twice the initial values during the compensated phase, the pulmonary changes declined to 0% from 100% during the uncompensated phase. The authors, therefore, concluded that the pulmonary changes were due to nonspecific effects of acidosis rather than to direct effects of carbon dioxide. They suggested that the absence of such pathologic findings in rats was indicative of species differences. Rats did exhibit edema on exposure to 3% carbon dioxide for 3 weeks, and the edema was extreme when the rats were exposed at 30 and 50% of the gas for 5 hours and 1 hour, respectively, but dissipated after 7 days of recovery in room air. The authors attributed these pulmonary abnormalities to stimulation of the adrenal glands and suggested that such effects of adrenal cortical activity were not uncommon.

In a followup study, Schaefer et al [80] exposed adult male guinea pigs of the Hartley strain to carbon dioxide in a plastic chamber. Data were gathered from electron microscope studies, surface tension measurements of lung tissue, and additional histochemical studies. These authors also identified four phases of pulmonary changes caused by 15% carbon dioxide. The initial phase (6 hours) was marked by uncompensated respiratory acidosis accompanied by pulmonary effusion and changes in the lamellar bodies of the granular pneumocytes. (The lamellar bodies are intracellular stores of surfactant, a material which lowers surface tension and tends to stabilize alveolar diameter.) This period is not associated with hyaline membrane formation. The second phase (6-24 hours) was associated with hyaline membrane formation. During the third phase (days 2-7), the surface tension returned to normal, the pulmonary edema

diminished, and hyaline membranes disappeared. The final phase was one of recovery, although the pCO₂ remained elevated. Cellular structure and function returned to normal. The authors concluded that the granular pneumocytes were the cells responsible for changes in the observed lung tissue surface tension.

Stinson and Mattsson [81] investigated the effects of carbon dioxide on cardiac rate and rhythm in six healthy rhesus monkeys, and reported findings pertinent to respiratory rate. They found that the respiratory rate increased twofold until a 10% carbon dioxide concentration was reached and thereafter decreased until the animals died. The authors concluded that depression of respiration overcomes stimulation at concentrations greater than 10% carbon dioxide. Similar plateauing at about 10% carbon dioxide has been shown in man. [27] They also reported that, in all cases, death ensued at concentrations greater than 60%. The authors also reported that survival at this concentration was possible if the concentration was attained at an elevation rate of 7.5-30%/hour (0.12-0.5%/minute) and the decrease was at a rate of 1-2%/minute.

The animal studies described in this section have indicated that carbon dioxide-induced respiratory acidosis may lead to hyaline membrane formation and atelectasis* in guinea pigs and can cause edema in rat lungs. However, hyaline membranes, which appeared only in guinea pigs, were attributed by the authors [79,80] to the absence of a specific enzyme system in the lungs of this species. The pulmonary edema* observed in rats was reversible upon return to room atmosphere.

(c) Cardiovascular Effects

The effects of carbon dioxide on cardiac rate and rhythm were investigated in rhesus monkeys by Stinson and Mattsson. [81] Six healthy

animals were placed in an environmental chamber for exposure at gradually increasing carbon dioxide concentrations at rates of 7.5, 15, or 30%/hour while oxygen was held constant at 20-21%. Electrocardiogram, respiratory rate, and body temperature were recorded. One animal was killed after initial exposure; the others were killed when, in response to elevated carbon dioxide concentration, respiration and electrical heart and brain activities ceased. These occurred during the fourth exposure to carbon dioxide. The authors reported no differences in effects caused by the rate of increase of carbon dioxide. The overall changes recorded included an increased heart rate at concentrations up to 10% of the gas and a subsequent decrease with higher concentrations until 35-40% was reached.

Stinson and Mattsson [82] reported comparable findings for three chimpanzees (*Pan troglodytes*) in a study of similar design and objectives. The same kinds of data were collected as in the previous study [81] and results of both were compared. Data from this study [82] demonstrated that chimpanzees, like rhesus monkeys, can survive carbon dioxide concentrations as high as 390 mmHg (51.3%). The authors suggested that humans could also survive such levels if the concentrations were raised and lowered gradually.

Stein et al [83] reported results in contrast to those previously described by other investigators. [30,35] In this experiment, 20 healthy male rhesus monkeys (*Macaca mulatta*) were selected and randomly divided into test and control groups of 10 monkeys each. The 10 test animals were exposed to 3% carbon dioxide in air for 93 days in a sealed chamber. A preparation period before the experiment allowed for baseline data gathering. Following the 93-day test, a recovery period was scheduled.

Dietary habits, weights, subjective estimates of respiratory rate, water intakes, complete blood chemistry determinations, and general activity were recorded for each monkey. Measurements and observations were made throughout the exposure period. The experiment revealed no statistically significant changes in any of the measured parameters. One monkey died of shigellosis on day 62. In addition to the routine analyses, five animals killed at random at the conclusion of the test were microscopically examined. The remaining four animals were killed 28, 35, 40, or 46 days after the test for similar studies. The autopsy reports were routinely negative. The authors concluded that there was no evidence of a biphasic reaction to carbon dioxide. No periods of uncompensated or compensated acidosis and no evidence of adrenal impairment were noted. The results of this study vary markedly from those of other animal studies.

Brown and Miller [84] studied the occurrence of ventricular fibrillation (an arrhythmia consisting of irregular and rapid ventricular excitation not followed by a propulsive ventricular contraction resulting in output of cardiac blood) after a fall in alveolar carbon dioxide. The investigators exposed 17 mongrel dogs, anesthetized with sodium thiopental, to a mixture of 30% carbon dioxide and 70% oxygen for 2 hours, at which time the concentration was raised to 40% carbon dioxide for 2 hours. The gas mixtures were administered from a spirometer through a tracheotomy tube. At the end of this exposure, 15 dogs were suddenly changed to breathing normal air and mechanically hyperventilated, and 2 dogs were slowly restored to breathing air. All dogs were ECG-monitored before introduction of the gas, at selected intervals during exposure to carbon dioxide, and during the transition to air. Blood pH, plasma carbon dioxide

concentration, and blood pressure also were determined during the experiment. All dogs suddenly exposed to air showed cardiac arrhythmias within 0.5-6 minutes; eleven of them developed ventricular fibrillation and died within 2.5-10 minutes after the sudden change. In contrast, the two dogs which were returned gradually to air breathing survived, and neither showed any signs of cardiac dysfunction. Average pH values fell from an initial 7.36 to 6.67 after 4 hours of carbon dioxide administration. The pH measurements were made on five dogs. The plasma carbon dioxide tension rose from 43.2 to 295 mmHg (5.7-38.8%). The authors also reported that the pH rise observed after a slow return to air breathing was exceedingly rapid: a pH of 7.3 was attained within 3-4 minutes. Although these results are obviously significant, the concentrations used were extremely high. A similar study in rats [85] revealed cardiac arrhythmias during exposure to 50 and 70% carbon dioxide mixed with 22% oxygen, with an increased incidence of arrhythmias noted upon rapid withdrawal.

(d) Neuroendocrine Effects

The effects of narcotic levels of carbon dioxide on adrenal cortical activity were reported by Schaefer et al. [86] Male rats of the Wistar-Hisaw strain and male guinea pigs of the Connaught strain were exposed to 30% carbon dioxide in air or a mixture of 30% carbon dioxide and 70% oxygen for 10 minutes or 1 hour. The animals were killed either immediately or 1 hour after exposure. Blood samples were drawn for determination of pH, whole blood carbon dioxide content, oxygen content, hematocrit, and blood counts including absolute eosinophils. Also determined were adrenal cholesterol levels, blood sugar, liver and muscle glycogen, blood lactic acid, plasma potassium and sodium, and adrenal weights. The guinea pigs

had a mortality rate of approximately 50% under both gas mixtures. Adrenal cholesterol decreased significantly after exposure to 30% carbon dioxide in air. The 10-minute exposure resulted in a drop from a normal level of 5.04 to 3.44 mg%. After 60 minutes of exposure, the level was 4.04 mg%. Absolute lymphocytes decreased slightly from a control count of 5,882 cells/cu mm to 4,122 and 5,507 cells/cu mm after 10 and 60 minutes, respectively. At the same time, the eosinophil counts increased from a control of 87 cells/cu mm to 206 and 257 cells/cu mm, after 10- and 60-minute exposures, respectively. A 1-hour exposure to 30% carbon dioxide in air resulted in increased adrenal weights which had not returned to normal after 1 hour of recovery. A 1-hour exposure to the mixture of 30% carbon dioxide and 70% oxygen resulted in changes in adrenal cholesterol and hematologic parameters; however, these changes were not as marked as those observed during exposure to carbon dioxide in air. The recovery period after both gas mixture exposures resulted in adrenal cortical stimulation, as evidenced by decreased adrenal cholesterol, lymphocytes, and eosinophils, although total leukocytes remained unchanged. The authors concluded from these results that the narcotic levels of carbon dioxide alone did not stimulate the adrenal cortex. When the gas exposure was complicated by hyperoxia (70% oxygen), increased adrenal cortical activity was observed. The authors also reported that changes noted in adrenal cholesterol and in lymphocytes reflected a stronger adrenal cortical stimulation after 10 minutes than after 60 minutes of carbon dioxide exposure.

In a study with four dogs exposed to a mixture of 30% carbon dioxide and 70% oxygen, Harrison and Seaton [87] demonstrated marked increases in

the secretions of the adrenal medulla. This gas mixture resulted in a pH drop from a preexposure level of 7.3 to 7.0 during mechanically controlled respiration and to 6.4 during spontaneous respiration. This change corresponded to increases in secretion of adrenalin and noradrenalin. Another group of four dogs exposed to the same gas mixture retained a pH level of 7.2 with administration of trihydroxyaminomethane (THAM) buffer. In this buffered state, secretion of adrenalin and noradrenalin remained normal. This study suggests a pH dependence on the sympathoadrenal activity and subsequent alterations induced by carbon dioxide.

Schaefer et al [88] reported on a series of studies in which six healthy guinea pigs were continuously exposed to carbon dioxide at a concentration of 15% for 7 days. The experiments were designed to identify the specific mechanism of the sympathoadrenal response to carbon dioxide. Before killing the animals, the investigators drew blood samples for measurements of blood pH and blood corticosteroids. At necropsy, the epinephrine content of the adrenals, nonesterified-free-fatty-acid levels, adrenal cholesterol concentration, and organ weights were determined. The experiments included 4 control days before administration of the carbon dioxide and 11 recovery days. Within 2 days after exposure to the gas, the test animals showed a 10% weight loss. During the remaining days of exposure, the animals gained weight and reached their initial body weights within 5-7 days. Animals killed after 1 day of exposure showed significantly increased adrenal weights. These remained high for 7 days. Spleens had decreased weights on the first day of exposure only, while the weights of the thymus and para-arterial nodes remained low throughout the exposure period. Studies of pH showed an initial drop during the first

hour of exposure with a consistent rise of 0.1 pH unit daily for the following 2 days. By the third day, the respiratory acidosis was nearly compensated. The first 3 days of exposure were accompanied by a dramatic rise in blood corticosteroids and fall in adrenal epinephrine concentrations. This 3-day period of uncompensated respiratory acidosis was accompanied by a rise in free fatty acids in the arterial blood. The phase was analogous to that reported in humans. [30,36] The adrenal cortical stimulation during this phase was also indicated by an observed lymphopenia and decreased adrenal cholesterol level in the animals.

In contrast, when guinea pigs were intermittently exposed to the same concentrations of carbon dioxide for 8 hours daily for 7 days, significantly different results were seen. [88] These animals experienced no compensation of the respiratory acidosis and no decline of sympathoadrenal responses. In addition to the blood pH studies repeated for all animals, tissue carbon dioxide and intracellular pH were measured in this group. The findings indicated that changes in intracellular pH values followed the carbon dioxide-induced extracellular pH fall after a few hours' lag. The intracellular pH rose to normal levels within 3 days of exposure; this rise corresponded with the identified extracellular pH pattern. From these observations, the authors concluded that the stress response in chronic hypercapnia depends on extracellular and related intracellular pH changes and is representative of a nonspecific pH-dependent effect. The interaction of adrenal cortical and adrenal medullary responses was demonstrated in this study as an increase in blood corticosteroids and a concomitant decrease in adrenal epinephrine. The epinephrine level was seen to increase after 6 hours. The authors

correlated this rise with the compensation of the respiratory acidosis phase. Further, they concluded that this was another demonstration of the pH dependence of the sympathoadrenal stimulation caused by chronic hypercapnia.

(e) Effects on Acid-Base and Electrolyte Balance and on Calcium Metabolism

Animal studies [89,90] have indicated acid-base changes similar to those already discussed in humans. Compensated and uncompensated acidoses are evident from the data reported in these papers and parallel these phases in human exposures. Data from these studies are presented in Table III-6 at the end of this chapter. Further data may be obtained from the references [89,90] as listed in Chapter VIII. The following study [91] offers information on calcium metabolism, an area not yet thoroughly investigated in humans.

In a 1961 study, Schaefer et al [91] reported on the calcium and phosphorus metabolic changes caused by prolonged exposure to 15% carbon dioxide. Male Hartley guinea pigs were exposed to the gas for periods of up to 73 days. Measurements were made of blood pH, hematocrit, plasma calcium, blood and urine inorganic phosphorus, plasma protein, and albumin-globulin ratio. Plasma calcium rose 0.4 mg% after 1 day while inorganic phosphorus declined by 0.2 mg%. By the second and third days, these changes became significant. Even after 20 days of exposure, the higher calcium and lower phosphorus levels were maintained. The authors suggested that these changes were indicative of parathyroid stimulation. This theory was substantiated by a corresponding increase in urinary phosphorus excretion. A statement included in the report indicated renal

calcification after 48 hours of exposure although no further data were offered. This effect was also reported to be associated with parathyroid stimulation.

(f) Effects on the Reproductive System

In a study by Mukherjee and Singh, [92] morphologic changes in spermatozoa and reduced fertility were found in mice exposed to approximately 35% carbon dioxide. Twenty male Swiss-strain mice, 10 controls and 10 test animals, were studied for changes in spermatozoa. The mice were repeatedly exposed to the gas for periods of 1 or 2 hours for a total of 6 hours in an environmental chamber. The exposures were divided so that single exposures were either for 1 or 2 hours. After 6 hours of exposure, the mice were killed and slides of the contents of the vas deferens were prepared. Measurements of spermatozoa after the males were exposed to carbon dioxide showed that sperm maturation had been affected. Measurements of sperm head area, head breadth, midpiece area, and midpiece breadth indicated statistically significant decreases. For the fertility studies, male and female mice were allotted to test and control groups of equal numbers (a maximum of five in each group). The experiment involved exposing males on day 1 for 4 hours and subsequently for 4.5 hours on each of the next 5 days. After the first exposure, the mice were paired with unexposed virgin females on each of the subsequent five nights. This experiment was repeated with new animals 11 times. Delayed effects of carbon dioxide exposure were studied in males from six of these trials. They were mated with new virgin females for 6 days, beginning 15 days after the end of the last carbon dioxide exposure. In the tests of fertility, the mean litter size ranged from 5.75 to 10.00 in the control group and

from 3.33 to 9.33 in the test group. The differences after the delayed fertility test were 6.75 to 8.00 in the control group and 5.00 to 9.25 in the test group. The number of males siring a litter varied from three to five in the control group and from one to four in the exposed group. Although the authors concluded that carbon dioxide exposure affected the conception rate of the mice, the variability in litter sizes of the test and control groups in the early fertility test, the minor differences in number of males siring litters, and the small group size preclude any reliable statistical evaluation of the results. Therefore, no definite correlation may be made between carbon dioxide exposure and effects on fertility of male mice. There was no significant difference in the test of delayed action of carbon dioxide.

The effects of carbon dioxide on the testes of rats were studied by VanDemark et al. [93] Forty mature male Wistar rats were exposed to 2.5, 5.0, or 10% carbon dioxide in 20% oxygen made up to 100% with nitrogen for 1, 2, 4, or 8 hours in an experimental chamber. The rats were killed immediately after exposure, and microscopic examination of the testes and seminal vesicles revealed that degenerative changes in the testes paralleled the concentration of carbon dioxide and duration of exposure. Eight-hour exposures did not produce changes significantly different from those effected by 4-hour exposures. Major histologic effects of the carbon dioxide exposure included tubular disturbances such as sloughing, as well as loss of luminal definition. The authors did not describe results after treatment with 2.5% carbon dioxide for 1 and 2 hours; however, exposure to the same concentration for 4 hours reportedly resulted in a typical intratubular disruption. Sloughing of tubular compartments and lack of

luminal definition were evident after 4 hours of exposure to 5% carbon dioxide. Four hours of exposure to 10% carbon dioxide also caused degenerative changes such as streaking and vacuolization. These changes appeared to be reversible, as testes microscopically examined 36 hours after exposure (concentration unstated) were normal. Further specific correlations of dose and response were not presented by the authors.

(g) Carcinogenic, Mutagenic, and Teratogenic Effects

The potential teratogenic effects of carbon dioxide were investigated by Haring. [94] The author exposed pregnant Sprague-Dawley rats to an atmosphere of 6% carbon dioxide containing 20% oxygen and 74% nitrogen in a plastic chamber in pairs for a single 24-hour period between days 5 and 21 of pregnancy. The number of chambers of rats exposed on each day of gestation varied from two to seven. Control animals were exposed to room atmosphere. Conditions of the pups at birth, method of delivery, uterine abnormalities, implantation sites, and body weight of the dams were recorded. The newborn pups were killed immediately after birth, and their chest organs were fixed for histologic examination. Seventy-one test dams produced 530 newborn, while 21 control dams produced 159 infant rats. The control dams yielded 80 (50.3%) males and 79 (49.7%) females; in the test group, 238 (44.9%) were males and 292 (55.1%) were females. The average litter sizes were 7.57 rats/litter in the control group and 7.47 in the treated group. Five (3.1%) of the 159 rat pups in the control group and 41 (7.7%) of the 531 test group pups were stillborn or died soon after birth. The test dams weighed more than the control rats. Cardiac malformations were reported in 106 (24.3%) of the test animals and in only 7 (6.8%) of the control group. Malformations were of five major types: (1) high

interventricular septal defects (7 cases); (2) low interventricular septal defects (8 cases); (3) riding aorta (in 24 cases, the aorta emerged from both ventricles and the pulmonary artery arose solely from the right ventricle); (4) partial transposition (24 cases); and (5) pulmonic or aortic stenosis with intact interventricular septum and myocardial hypertrophy (47 cases). Malformations of only the first type were found in 7 (6.8%) of 102 control young. The greatest incidence of malformations was in animals whose dams were exposed to carbon dioxide on the 10th day of gestation. While many aspects of the mechanism of action remain unsolved, the teratogenic effects of carbon dioxide identified by this author appear to be significant. Further, the author indicated that localized tissue overgrowth was present in the lungs, thymus, and heart and suggested that in some way, this accelerated growth may be related to the cardiac malformations described. In addition to these results, the incidence of skeletal malformations was 10.9% in the test series and 0.6% in the control group. The use of only one high concentration (6%) of carbon dioxide in this study precludes any evaluation of dose-effect relationships. Moreover, since this study involved exposure of the rat, an animal with a relatively short (21-day) gestation period, to a relatively high concentration of carbon dioxide, application of these results to higher species with longer gestation periods is not possible. This study [94] was listed in the Registry of Toxic Effects of Chemical Substances [95] as showing teratogenic effects.

A report by Grote [96] described possible teratogenic effects in rabbits exposed to high concentrations of carbon dioxide. Three mongrel rabbits were exposed to 10-13% carbon dioxide for 4-10 hours on 2 or 3

days between days 7 and 12 of pregnancy. Three control animals were kept under conditions similar to those of the test group, except in a normal atmosphere. Of the 67 pups from 11 litters yielded by dams that had been exposed to carbon dioxide, 16 had congenital malformations in the cervical, thoracic, or lumbar vertebral column. The one animal exposed only on the seventh and eighth days to 13% carbon dioxide for 10 hours had a litter in which no malformations were seen. The distribution of malformations observed in 16 pups (11 males and 5 females) showed a threefold greater susceptibility of males. The control group of 30 animals showed only one malformation (dysplasia of the third and fourth sternal segment). The author stated that perhaps 13% was not the tolerance limit in these studies and that future experiments at higher concentrations might offer additional information. The most common malformations observed in treated animals were hypoplasias of the entire vertebral structure or of individual vertebral parts. Malformations of the sternum and ribs occurred to a lesser extent and usually were associated with malformations of the vertebrae. The use of only three rabbits in the carbon dioxide portion of the study diminishes the value of these results. The predictive value of this study for humans is limited by the high concentrations (10-13%) at which the rabbits were exposed and the long exposure time relative to the short (30 days) gestation period. Exposures at these concentrations for this length of time would not be tolerated in humans. This study [96] was also listed in the Registry of Toxic Effects of Chemical Substances [95] as showing teratogenic effects.

A single article has been identified which ascribes significance to carbon dioxide snow (or carbonic acid) as a cancer-producing agent when

used as a chronic irritant. Mansens [97] used the snow solely as a cold-producing agent, administering it as a chronic irritant to the backs of mice that had had their hair removed with either sodium or barium sulfide. The strain, age, and sex of the mice were not specified. The snow was packed into a wooden rod, and the tip of the protruding snow was applied to the test animals. The animals were divided into three groups of 100, 30, and 30 mice; Group I was treated twice weekly; Group II was treated every other day; and Group III was treated daily. Each 1-second application resulted in momentary freezing of the skin site. Within 24 days, seven mice in Group I showed evidence of skin thickening at the treatment sites. By the following month, these areas were not evident, although new ones appeared in other mice. The investigator was unable to follow the progress of these mice further. In Group II mice, a local irritation reaction was evident after 7 days of treatment. After 3 weeks, the author described the foci as papilloma-like lesions. Some of these disappeared and some recurred at the same site. However, the numbers of such sites were not reported. The author did state that, in some cases, distinct carcinomas (type unspecified) developed. One history of carcinoma development was cited. It showed that in an animal whose carbon dioxide treatment was begun in mid-July, by August of the following year, a carcinoma was evident, and the mouse died in October of that year. No cause of death nor autopsy finding was stated. After 490 days of treatment, only one mouse was still alive from Group II. This mouse had a papilloma. Again, no causes of death nor autopsy reports were given. Of the 30 animals in Group III, 26 developed papillomas and 2 with carcinomas died. After 390 days, of six still alive in Group III, one had a carcinoma. The author concluded

that chronic application of a nonspecified cold irritant for at least 240 days resulted in carcinoma development in mice. The author described no other specific effects of carbon dioxide.

Correlation of Exposure and Effect

The most immediate and significant effects of acute exposure at a high carbon dioxide concentration are those on the CNS. Experiments with humans have shown that concentrations of 20-30% carbon dioxide in oxygen result in unconsciousness and convulsions within 1 minute of exposure. [22,28,44] Further, monkeys exposed to 30% carbon dioxide for 10 minutes have demonstrated excitation of the cortical and subcortical regions followed by depressant effects, as evidenced by a rise in brain excitation thresholds in these animals. [30] In humans, unconsciousness has occurred at a concentration of 17% after 37 seconds and after only 25 seconds of exposure at 27.9% carbon dioxide. [29] It has also been reported [22] that human subjects became unconscious at concentrations of 11-13% carbon dioxide after 8-23 minutes of exposure; neurologic symptoms including psychomotor excitation, myoclonic twitches, and eye flickering appeared after 1.5 minutes of exposure at 10-15% of the gas. [22] Other symptoms, such as headache, sweating, dizziness, mental clouding, faintness, and restlessness, were reported in humans after approximately 4 minutes of exposure to 10.4% carbon dioxide. [27] Headache, restlessness, and dizziness were also noted after 7-15 minutes of exposure at approximately 7.5% of the gas. [27,30] The recurrent complaint of headache during acute exposure may be caused, in part, by the observed increases in cerebral blood flow and in CSF pressure, which are also attributable to carbon

dioxide exposure. [46,78]

The effects of lower concentrations of carbon dioxide on respiratory function in humans have also been demonstrated to be immediate and significant. Graded increases in respiratory minute volume and ventilatory rate accompanying increased carbon dioxide in the inspired air have been reported. [27,35,36,41] One investigator found that the volume of expired air was more than 16 times the control volume on exposure at 5.2% of the gas during maximum exertion exercising. [65] Respiratory minute volume gradually increased from an average resting level of 7 liters/minute at 0.03% carbon dioxide to 8 liters/minute at 1%, 9 liters/minute at 2%, 11 liters/minute at 3%, and 26 liters/minute at 5% carbon dioxide. At 10.4% carbon dioxide, the highest concentration tested, the average respiratory minute volume was 76.8 liters/minute. Tidal volume also increased from a control level of 440 ml to 2,500 ml during exposure to 10.4% carbon dioxide. [27] Airway conductance has been shown to decrease upon exposure to 5-10% carbon dioxide, [39] and increases in anatomical dead space have been reported in response to 0.8 and 0.9% concentrations of the gas. [36,37] The dyspnea often experienced by study subjects [27,30,61,62] may be correlated with a combination of these respiratory effects and is reported to occur at respiratory minute volumes ranging from 62.7 to 86.8 liters/minute.

Cardiovascular changes during carbon dioxide exposure at rest have been reported. Inhalation of 6% carbon dioxide while at rest resulted in a decreased amplitude of the QRS complex of the ECG in 6-8 minutes. [45] Older subjects appeared more sensitive to this change. [45] The authors did not discuss whether these changes suggest that chronic cardiac

dysfunction could occur with continued exposure. In another experiment, [28] exposure to 30% carbon dioxide in 70% oxygen resulted in extrasystoles in a small percentage of patients. Similarly, MacDonald and Simonson [44] noted supraventricular arrhythmias and premature auricular and ventricular systoles on inhalation of 30% carbon dioxide and 70% oxygen. However, because high concentrations of both carbon dioxide and oxygen were used, it is difficult to evaluate these studies in terms of exposure to low concentrations of carbon dioxide in air. The cardiovascular effects of exposure to 1-3.9% carbon dioxide during exercise were observed by Menn et al [61,62] and consisted mainly of premature ventricular contractions. The authors stated that these changes are the most common ECG arrhythmias and are a frequent finding in normal men breathing air during exercise. Exercise performed during inhalation of 2.8% carbon dioxide [63,64] did not result in any cardiac changes. The majority of cardiac effects have been observed at high concentrations of both oxygen and carbon dioxide, while no cardiac changes or only minor alterations which cannot be definitely attributed to carbon dioxide exposure have followed inhalation at lower concentrations, even during exercise.

Investigations into the behavioral effects of exposure to increased carbon dioxide in humans have shown some increases in alpha-blocking latency indicative of CNS depression at 7.5% after 15 minutes of exposure. [30] During a 30-minute exposure to 6.1% carbon dioxide at 5 C, a significant suppression of shivering was evident. [51] Studies of intermittent exposure to 3% carbon dioxide (15 hours in carbon dioxide, 9 hours in room air) demonstrated no decrements in psychologic function in tests of single-digit addition and of letter cancellation. [34] No

decrements in the performance of subjective psychomotor tests were observed during chronic exposure to 4% carbon dioxide. [48] Minimally diminished color sensitivity after intermittent exposure to 3% carbon dioxide was reported as were optical aberrations after a similar intermittent exposure at 1.5% of the gas. [49] A single study involving chronic exposure to 3% carbon dioxide [50] indicated biphasic behavioral patterns. The first phase, lasting for only 1 day, was one of stimulation, as evidenced by increased mental awareness and activity; the second, beginning on day 2, was indicative of behavioral depression and mental exhaustion in the subjects. These mental alterations subsided by the third day, yet behavior never returned to normal throughout the 8-day exposure.

The most far-reaching actions of carbon dioxide in the body involved acid-base and electrolyte changes. Increased carbon dioxide levels resulted in respiratory acidosis in humans and in animals. [24,33,47,53,54,57,89,90] This acidosis developed more rapidly upon exposure to high concentrations and was compensated for by homeostatic mechanisms more quickly than after exposure at low concentrations. [54,58] In humans, blood pH values decreased after 2 days of a 5-day exposure to 3% carbon dioxide. [33] Other studies in humans have shown that continuous exposure to 1.5% carbon dioxide results in uncompensated respiratory acidosis for the first 23 days of a 42-day exposure. This acidosis was adequately compensated during days 24-42 by homeostatic mechanisms including blood buffers, ventilatory changes, and renal compensation. [54,57] The extent of this acidosis in response to a 1.5% continuous exposure, although measurable, was minor. The 0.12-unit drop in pH value was not correlated with any clinical symptoms. [54]

Neuroendocrine studies have documented responses to increased carbon dioxide that indicate increased adrenal activity. Increased outputs of norepinephrine, epinephrine, 17-OH-corticosteroids, and plasma catecholamines in response to inhalation of carbon dioxide at concentrations in excess of 6% have evidenced this sympathoadrenal response. [31] In animals, only 10 minutes of exposure to 30% carbon dioxide in air resulted in initial decreases in adrenal cholesterol and in lymphocytes, while the eosinophil count increased. [86] In humans, a 15-minute exposure to 3.3% carbon dioxide resulted in increased blood sugar, oxygen consumption, and muscle potential, while the number of eosinophils initially decreased. This was correlated with a decrease in pH and may have been indicative of the pH dependence of the mechanism. The eosinophil decrease was most pronounced at 5%, and the increase in pulse rate was significant at 7.5% carbon dioxide. It is probable that the observed increases in pulse rate, blood pressure, and eosinophils also were related in part to this adrenal response.

Acclimatization, or the development of tolerance to carbon dioxide, has been shown consistently. [35,38,42,57,58] Diminished respiratory response to a subsequent challenge of 5% carbon dioxide has been demonstrated after short or prolonged exposures at increased carbon dioxide concentrations of 7.5 and 1.5-3% carbon dioxide, respectively. [36,38,41] The increased ventilatory response produced by chronic carbon dioxide exposures returns to normal after 2-3 days of exposure at 1.5 and 3%. [35,58] This finding is generally associated with an increased ventilatory depth, [35,58] as well as with increased anatomical dead space [35,37] in the presence of increased alveolar carbon dioxide tension. [35,41,42] The

overall effect is an increased respiratory efficiency evidenced by improved oxygen intake and carbon dioxide excretion. Tolerance may be related to elevated carbon dioxide tissue levels as was especially apparent in groups of people who, in training, were subjected to periodic carbon dioxide retention through voluntary breath holding. [38] Adaptation appears to be related to symptoms experienced by subjects on exposure to 3% carbon dioxide. Headaches were experienced during chronic exposures and were present only for the first 2 days, [33] after which time adaptation seemed to overcome this symptom. Adaptation also appears to be characterized by cardiovascular acclimatization, evidenced by a leveling of pulse rate after an initial increase, with continued exposure to 3% carbon dioxide for 5 days. [33]

Intermittent exposures to carbon dioxide may result in effects unlike those resulting from chronic or from acute exposures. In studies in which only one subject was exposed to carbon dioxide for 15 hours and returned to room air for the following 9 hours, pulmonary and renal compensatory mechanisms were sufficient to return all systems to normal preexposure levels by the fourth day of exposure. [41,53] This is in contrast to the investigation in which attainment of compensation at a higher-than-normal pCO₂ level was demonstrated. [38] Intermittent exposure was characterized also by unexpectedly increased ventilatory response to challenges with higher concentrations of carbon dioxide, [41] which contrasts with the depressed ventilatory response evident after chronic exposures. [38] However, factors which must be considered in the interpretation of these findings include individual variation and other problems inherent in single-subject experiments. It is possible, however, that intermittent

exposures do result in a form of tolerance which is not as clearly demonstrated as that occurring during chronic exposure. No information which adequately supports this hypothesis has been found.

Human exercise studies, which have confirmed the additive stress effects of increased carbon dioxide concentration and exercise, are important because of their applicability to occupational exposures. Exercise studies which simulate strenuous labor are more relevant than laboratory experiments in which subjects undergo test exposures at rest. At carbon dioxide levels below 2.8%, physically fit subjects tolerated strenuous-exertion-exercise programs (at 180 W, 2.58 kcal/minute) without detectable stress. [47,61,62,64] At or above this concentration, the subjects did begin to experience some benign ECG abnormalities while exercising even during air breathing, [47] as well as respiratory exhaustion and headaches. At 2.8 and 3.9% concentrations of carbon dioxide, intercostal-muscle pain, respiratory difficulty, and mild headaches were reported at workloads at or above 180 W, an extreme-stress situation. At 5.2% carbon dioxide, strenuous exercise (running at 6 mph on a treadmill at a 10% grade) resulted in mental confusion, impaired vision, and collapse (symptoms not reported at this concentration in the absence of exercise), thereby confirming the additive stress effects of carbon dioxide and exertion. [65] It has been suggested by Sinclair et al [64] that no appreciable differences in respiratory response were evident between acute and chronic exposures to the gas during exercise, although Schaefer has noted that the respiratory response to carbon dioxide was attenuated with exercise during chronic exposure. [58]

Carcinogenicity, Mutagenicity, and Effects on Reproduction

A single study [97] has suggested carcinogenic effects of dry ice. However, as previously stated, because the dry ice was used only for its cryogenic properties, the results were not sufficient justification for evaluation of the possible carcinogenicity of carbon dioxide.

No studies have been found in which the possibility of mutagenic or carcinogenic effects from inhalation of carbon dioxide was investigated.

Animal studies have suggested that carbon dioxide exposure may cause changes in the reproductive system. [92,94,96] Male mice briefly exposed to 35% carbon dioxide had structural alterations in sperm cells with possible subsequent effects on the conception rates of their mates. However, the variability of the data provided on litter size and the minor difference between numbers of treated and control males siring litters precluded a statistical analysis; therefore, no definite conclusion can be made on the effect of carbon dioxide on the fertility of male mice. [92] The relative value of this study as it pertains to human exposure is questionable because exposure to 35% carbon dioxide for more than a few minutes causes death. [44] Studies on rats [93] indicated that degenerative changes in testicular tissue occurred as a result of exposure to between 2.5 and 10% carbon dioxide for 4 or 8 hours. These changes were reversible, since testicular tissue appeared normal 36 hours after exposure.

Teratogenic effects produced by carbon dioxide in rats and rabbits also have been reported. These effects have included congenital spinal malformations in the young of pregnant rabbits exposed to 10-13% carbon dioxide for 4-10 hours/day for up to 3 days and cardiac abnormalities in rats exposed to 6% carbon dioxide for 24 hours. [94,96] Interpretation of

these results in terms of the human reproductive system is limited because only species with short gestation periods were tested and the concentrations of carbon dioxide used would not be well tolerated by humans. No reports have been found which attributed similar effects either to lower carbon dioxide concentrations or to exposure of other species.

Summary Tables of Exposure and Effects

The effects of acute, intermittent, and chronic exposures to carbon dioxide in humans which were presented in detail in Chapter III are summarized in Tables III-1, III-3, and III-5, respectively; those of exposure to carbon dioxide in animals are shown in Tables III-2, III-4, and III-6. The human exercise data are summarized in Table III-7. Throughout the tables, the symbol \uparrow means increased and \downarrow means decreased.

TABLE III-1

EFFECTS OF ACUTE EXPOSURES TO CARBON DIOXIDE ON HUMANS

Exposure Concentration and Duration	Exposure Method	Number Subjects*	Effects	Reference
30% 38 sec	Face mask	17 M	Narcosis, ECG abnormalities in 16 of 27 episodes experienced by 25- to 48-yr-old subjects	44
50 - 30% 52 sec	"	37	Unconsciousness in 24-28 sec; abnormal EEG's; cardiac irritability	28
30%, 20% 1 min	-	8	Unconsciousness and convulsions within 1 min	22
16 - 27.9% 35 sec, 17% 20 - 52 sec	Face mask	3	Throat irritation; ↑ respiration; dimness of vision; dizziness; unconsciousness	29
7 - 14% 10 - 20 min	Rubber mouthpiece	12 M	↑ plasma catecholamines and steroids, ↑ sympatho-adrenal activity; loss of consciousness above 10%; headache, sweating, etc, above 7%	31
15%, 10% 1.5 min	-	8	Neurologic signs: eye flickering, myoclonic twitches, dilated pupils, restlessness	22
10.4% 3.8 min, 7.6% 7.4 min	Face mask	44 M	↑ systolic and diastolic BP, ↑ pulse rate; ↑ respiratory minute volume; headache, dizziness, faintness, etc	27
7.5%, 5.4%, 3.3%, 1.5% 15 min at each	-	42	↓ total eosinophils; ↑ blood sugar, muscle potential, and O ₂ consumption indicative of ANS response; ↓ flicker fusion frequency, ↑ alpha blocking latency; 7.5% threshold for symptoms; depression of CNS activity	30
7.5%, 5.4%, 3.3%, 1.5% 15 min at each	-	60	↑ initial alveolar pCO ₂ , ↓ response to 5% CO ₂ challenge in low-ventilatory-response subjects; lesser effects in high-ventilatory-response subjects	38
7%, 5% 15 - 30 min	-	12 M	↑ (75%) cerebral blood flow; ↑ CO ₂ and H ⁺ in arterial blood	46
6% 6 - 8 min	Face mask	148 M	More decided ECG alterations in older group (mean age 60.9 yr) than in young group (mean age 23.3 yr)	45
5% 30 min	-	19	↓ vascular resistance, all subjects; ↑ renal blood flow, 6 normal subjects; constriction of renal vasculature, 13 renal disease subjects	52
2.5 - 6% 75 min	Douglas bag	6 M	Total suppression of shivering response in 3 of 6 healthy young (mean age 24) subjects in a cold (5 C) room; breakthrough shivering after 30 min	51

*Where reported, M = men

TABLE III-2

EFFECTS OF ACUTE EXPOSURES TO CARBON DIOXIDE ON ANIMALS

Exposure Concentration and Duration	Species	Number Animals*	Effects	Reference
5 - 70% 20 min	Rats	-	Arrhythmias in 10-55 sec after sudden withdrawal of CO ₂ above 40%	85
Up to 51%	Chimpanzees	3	Survival if concentration ↑ and ↓ slowly	82
40% 2 hr plus 30% 2 hr	Dogs	17	↑ plasma CO ₂ , ↓ pH; 15 deaths on sudden reexposure to air	84
35% 6 hr total	Mice	10 T, 10 C	↓ area and breadth of sperm head and midpiece; smaller litters by test males	92
30% 30 min	Rhesus monkeys	8	↑ cortical and hypothalamic activities; CO ₂ tolerance after exposure	30
30% 10 - 60 min	Rats, guinea pigs	-	↓ adrenal cholesterol after 10 min; ↓ lymphocytes; ↑ eosinophils; ↑ adrenal weights	86
30%	Dogs	8	Buffer-counteracted ↓ pH and ↑ adrenal medullary secretions	85
15%, 10%, 5%	"	5	↑ CSF pressure with ↑ CO ₂ concentration	78
7.5- 30%/hr	Rhesus monkeys	6	↑ heart rate; ↑ respiratory rate at up to 10% CO ₂ , then ↓ with ↑ CO ₂ until death	81
10 - 13% 4 - 10 hr/d x 2 or 3 d	Rabbits	3 T, 3 C	Exposure during gestation d 7-12: 2 does, 3 litters each; 1 doe, 2 litters; vertebral hypoplasias in 16 of 67 young	96
10%, 5.0%, 2.5% 1 hr, 2 hr, 4 hr, 8 hr	Rats	40	Reversible degenerative changes in testes	93
6% 24 hr	"	71 T, 21 C	Congenital cardiac and skeletal malformations possibly due to tissue overgrowth	94

*Where specified, T = test, C = control

TABLE III-3

EFFECTS OF INTERMITTENT EXPOSURES TO CARBON DIOXIDE ON A MAN

Exposure Concentration and Duration	Exposure Method	Number Subjects*	Effects	Reference
0 - 3% 15 hr/d x 6 d	Chamber	1	Impairment of scotopic and color sensitivities; no changes in visual acuity or depth of perception	49
0 - 3% 15 hr/d x 6 d	"	1	No alterations in vigilance, eye-hand coordination, sequential reaction, or problem-solving ability; emotional change apparent in subject during study	34
0 - 3% 15 hr/d x 6 d	"	1	↑ventilatory response to 5% CO ₂ challenge; ↑slope of CO ₂ tolerance curve	41
0 - 3% 15 hr/d x 6 d	"	1	↑pH and pCO ₂ in capillary blood; CO ₂ elimination through renal mechanism; blood lactate and pyruvate unaffected; urine volume doubled; ↑ organic acids and titratable acidity, d 1; ↑CO ₂ excretion and acid load, d 4-5	53

*A healthy 24-year-old man for all four studies [34,41,49,53]

TABLE III-4

EFFECTS OF INTERMITTENT EXPOSURES TO CARBON DIOXIDE ON ANIMALS

Exposure Concentration and Duration	Species	Number Animals*	Effects	Reference
15% 8 hr/d x 7 d	Guinea pigs	6	Initially ↓ pH, no acidotic compensation, no decline in sympathoadrenal response	88
Dry ice applications; up to 490 d	Mice	160	Cold-irritation-induced papillomas after minimum of 240 d of application	97

TABLE III-5

EFFECTS OF CHRONIC EXPOSURES TO CARBON DIOXIDE ON HUMANS

Exposure Concentration and Duration	Exposure Method	Number Subjects*	Effects	Reference
4% 2 wk	Environmental chamber	6 M	No psychomotor impairment; no decrement in complex-task performance by healthy young subjects	48
3.9% 5 d, 11 d; 2.7% 30 d	Chamber	12	↑ arterial and CSF bicarbonate; ↓ CSF pH; some cardiac abnormalities; headaches	47
3% 8 d	Environmental chamber	- M	Tolerance after 3 d; ↑ respiratory threshold; ↑ CO ₂ and HCO ₃ in blood of healthy subjects	58
3% 8 d	"	-	Excitatory, d 1: mental stimulation, euphoria; suppressive, d 2-8: exhaustion and confusion on d 2 abating but present throughout	50
3% 78 hr	"	2 M	On acclimatization, ↓ response to CO ₂ challenges	42
3% 5 d	Space cabin simulator	7	No changes in ammonia or titratable acidity; no changes in serum electrolytes, blood sugar, BUN, serum creatinine, or liver function; no significant changes in exercise or psychomotor studies	33
1.5% 42 d	Submarine	23 M**	Original "Operation Hideout" report: ↑ alveolar CO ₂ ; ↑ ventilatory rate; ↑ O ₂ consumption; initially ↑, then ↓ respiratory CO ₂ excretion	35
1.5% 42 d	"	23 M**	Inorganic phosphorus changes parallel to pH changes in other "Operation Hideout" reports, plasma calcium pH-dependent	56
1.5% 42 d	"	23 M**	Uncompensated phase (d 1-23): ↓ plasma pH, ↓ calcium, ↓ inorganic phosphorus, ↓ urine pH, ↓ bicarbonate excretion, ↓ pulmonary CO ₂ excretion; compensated phase (d 24-42): ↑ plasma calcium, ↑ pH, ↑ bicarbonate excretion, ↑ urinary pH	54, 55, 57
1.5% 42 d	"	23 M**	↑ minute volume, ↑ respiratory rate, ↑ anatomical dead space, ↑ tidal volume; ↓ CO ₂ excretion, uncompensated phase; ↑ O ₂ consumption, compensated phase	36
1.5% 42 d	"	23 M**	↑ alveolar CO ₂ ; ↑ ventilation; initially ↑ O ₂ consumption; initially ↑, then ↓ CO ₂ excretion; ↓ sensitivity to 5% CO ₂ challenge	28
0.8 - 1.2% 21 - 57 d	"	31 M	Compensated and uncompensated acidoses in long patrols; compensation by d 51; bone storage of CO ₂ 1st 4 wk, then excretion from bone with calcium	60

TABLE III-5 (CONTINUED)

EFFECTS OF CHRONIC EXPOSURES TO CARBON DIOXIDE ON HUMANS

Exposure Concentration and Duration	Exposure Method	Number Subjects*	Effects	Reference
1%, 2% 30 d	Pressure chamber	4 (2/level)	↓ blood pH; ↑ pCO ₂ of blood and alveolar air; ↓ ability to perform strenuous exercise after prolonged CO ₂ exposure	59
0.9 - 1.9% 30 d	"	7 M	↑ alveolar CO ₂ ; ↑ pulmonary ventilation	43
0.9%, 0.8% 20 d	Submarine	10	↑ physiologic dead space of 61 and 60%, respectively, during 29 routine patrols	37
10%, 7.5%, 5.0%, 2.5%	Spirometer	17	Subjects 9 normal, 8 asthmatic: evidence of ↑ airway constriction	39
***	***	420	All patients with chronic pulmonary insufficiency: plasma bicarbonate rise curvilinear to pCO ₂ ; blood pH rise linear to pCO ₂	24
***	Spirometer	22	Subjects 12 normal, 10 emphysematous: chronic pulmonary insufficiency similar to chronic hypercapnia; lowered respiratory sensitivity	40

*Where reported, M = men

**Participants in "Operation Hideout" reported by several authors [28,35,36,54-57]

***Data not applicable

TABLE III-6

EFFECTS OF CHRONIC EXPOSURES TO CARBON DIOXIDE ON ANIMALS

Exposure Concentration and Duration	Species	Number Animals*	Effects	Reference
20% 30 hr	Rats	-	Mortality 100%	89
15%, 3%, 1.5% Up to 93 d	Rats, guinea pigs	-	Hyaline membranes at 15 and 3% in guinea pigs, at 15% in rats; atelectases all levels in guinea pigs	79
15% 73 d	Guinea pigs	-	↑ plasma calcium; ↓ inorganic phosphorus; renal calcification after 48 hr of exposure	91
15% 11 d	Rats	-	↑ ammonia excretion; ↑ titratable acids; 40% mortality	89
15% 7 d	Guinea pigs	-	Uncompensated respiratory acidosis, 6 hr; hyaline membranes, 6-24 hr; membrane disappearance, d 2-7	80
15% 7 d	"	6	Initially ↓, then ↑ pH; acidotic compensation by d 3; ↑ blood corticosteroids, d 1-3; ↓ adrenal epinephrine	88
15%, 3.0%, 1.5%, 1.0% Up to 93 d	Rats, guinea pigs	279	Greater responses overall in guinea pigs; longer acidotic compensation time at lower exposures, serum enzyme activity pH-dependent	90
15% and 10% for alternate 2-d periods	Rats	-	↑ ammonia excretion; ↑ titratable acids	89
10% 14 d	"	-	↑ ammonia excretion; ↑ titratable acids; 5% mortality	89
3% 93 d	Rhesus monkeys	10 T, 10 C	No adrenal impairment; no biphasic acid-base reaction	83

*Where specified, T = test, C = control

TABLE III-7

EFFECTS ON HUMANS OF EXPOSURES TO CARBON DIOXIDE DURING EXERCISE

Exposure Concentration and Duration	Exposure Method	Number of Men	Exercise Conditions and Observations	Reference
5.2%, 3.9%, 2.6%, 1.3% 24 min at each %	Environmental chamber	9	Treadmill, 10% grade, subjects running at 1.8, 3.4, 4.8, and 6.0 mph; ↑ volume expired gas, ↑ acidosis from ↑ CO ₂ on exercise, ↓ blood pH	65
3.1- 3.9% Until subjects exhausted	Facepiece	13	Treadmill, up to 22% grade, subjects breathing with inspiratory and expiratory resistances; ↑ minute and tidal volumes, ↓ time to exhaustion	63
3.9%, 2.8%, 2.0%, 1.0% 30 min at each %	Environmental chamber	8	Steady-state and maximum-exertion exercises; respiratory symptoms at and above 2.8%, headaches at 3.9%	61, 62
2.8% 15 -20 d	Chamber	4	Exercises at 3 graduated levels; ↓ blood pH with ↑ CO ₂ on exercise, ↑ CO ₂ retention	64
1.9%	*	12	Ergometer-equipped bicycle at various brake loads; ↑ ventilation, ↑ O ₂ consumption, ↑ CO ₂ elimination	66
1.9%	*	10	Ergometer-equipped bicycle at various brake loads; ↑ blood pCO ₂ , ↑ potassium and phosphorus, metabolic acidosis	66

*Data not applicable