THE CURRENT ROLE OF INERT GASES IN THE SEARCH FOR ANESTHESIA MECHANISMS[1]

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> "If the inert gases can be considered as true anesthetics, the action of these spherically symmetrical atoms without any permanent dipoles furnishes the most conclusive demonstration that anesthesia need not depend on the effects of any specific structural grouping."
>
> (Butler—1950)

Twelve years have passed since the above statement appeared in Thomas C. Butler's comprehensive review, "Theories of General Anesthesia" (5). During this time, any doubts as to the ability of the "inert" gas xenon to cause anesthesia have been resolved. In 1951 Cullen and Gross (14) demonstrated conclusively that the chemically inert gas xenon is an anesthetic agent for man at atmospheric pressure. In consideration of the work that has been done in the field of inert gas anesthesia in the past decade, it seems appropriate to review the subject at this time.

The purpose of this review may be considered fourfold: 1) to summarize the work which has been done in establishing the biological role of xenon and other rare gases, 2) to familiarize the reader with the chemistry of the rare gases, 3) to review several of the newer theories of anesthesia, and 4) to consider the conditions which the chemistry of xenon imposes on any postulated mechanism of anesthesia. In their discussions the reviewers intend to confine their remarks primarily to the molecular level. In no way do they wish to imply, by such a limited approach, that the progress in neuroanatomy and neurophysiology is of any lesser importance. Only by eventual combination of knowledge on all levels will a more complete understanding of the phenomenon of anesthesia be attained.

The use of the term "inert gas" demands some explanation and definition. The phrase is used by the present authors in a metabolic sense and includes those gases which generally are considered to exert their biological effects without undergoing any change in their own chemical structures or modifying the

[1] The preparation of this review was supported in part by grants 2B-5270 and B-3625 from the National Institutes of Health.

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primary chemical structure of other substances—in chemical terms, those gases which do not form covalent or hydrogen bonds with tissue constituents under biological conditions. The term, therefore, is used to include such gases as nitrous oxide, ethylene, and cyclopropane as well as nitrogen, helium, neon, argon, krypton, and xenon. Although several of these gases are not anesthetic at less than one atmosphere pressure, all except helium and neon have been shown to produce "narcotic" effects at elevated pressures. The "sleep-of-the-deep," a result of increased nitrogen pressures, as discussed by Behnke (1b), is perhaps the most familiar example. The term "rare gases" will be used to designate only the members of the last group of the periodic table of elements.

A formal definition of the terms "narcosis" and "anesthesia" poses a more difficult problem. The present authors agree with Butler's statement (5) that "... vagueness and multiplicity of meanings are so firmly established in the usage of 'narcosis' and 'narcotic' in English as to render futile any insistence on rigid definitions, even for technical purposes." It would seem that such insistence belongs to the sphere of semantics. In this review, the terms are used interchangeably to denote a general depressant action on a biological system, and, in most cases, the term used was that preferred by the author whose paper was being reviewed.

I. THE BIOLOGY OF THE RARE GASES

Inasmuch as this review may be considered an extension of a portion of Butler's earlier review (5) it will not be necessary to recount the observations made by Lawrence *et al.* (31) and Tobias *et al.* (71) which call attention to the possibility that xenon may have narcotic effects. In the year following the appearance of Butler's review, Cullen and Gross (14) submitted mice, rats, and rabbits to atmospheres of approximately 80% krypton and 20% oxygen and observed no unequivocal evidence of narcosis. After 15 minutes of exposure, rabbits displayed some loss of head reflex, slight diminution in response to painful stimuli, some slowing of respiration, and a tendency to remain in induced, unnatural postures. Krypton–oxygen mixtures (80%:20%) administered to human subjects did not produce noticeable effects, but a 50% mixture of xenon and oxygen brought about an increase in pain threshold in man as measured with a Hardy-Wolff-Goodell apparatus. When the xenon concentration was increased to 70% in oxygen, two persons reported an incipient loss of consciousness after inhalation of the mixture for three minutes.

The first use of xenon as the only anesthetic agent (with 0.2 mg atropine premedication) in a surgical procedure was described by Cullen and Gross. Although evidences such as eyeball movement, active intercostal muscles, and the character of the respiratory pattern indicated the patient was only lightly anesthetized, no evidence of reaction to pain was observed, and full recovery from the effects of the anesthetic was achieved within 5 minutes.

Since this time, xenon-oxygen mixtures have been used for a number of surgical procedures. However, the current high cost of xenon precludes its use as a general anesthetic. The present cost remains at approximately $30 per liter. Extensive

clinical pathological studies associated with xenon anesthesia were reported in 1953 (48). In addition to urine analyses, the studies included hematologic and chemical analyses of blood. Recordings of blood pressure, respiratory rates, and electrocardiograms were also made. All of these data indicated changes in only a few of the entities examined, none of which is unique to xenon anesthesia. The relative elevation in the segmented cells of the peripheral blood during xenon anesthesia is consistent with that reported for ether, nitrous oxide, cyclopropane, ethylene, and chloroform anesthesia. Likewise, the decrease in serum potassium, which has been noted with ether and cyclopropane anesthesia, was observed also with xenon and probably is due to a decrease in general metabolic activity rather than to a specific effect of xenon. It is evident from these studies and others performed by Pittinger and his colleagues that there was a minimal disturbance of biochemical and physiological processes brought about by the xenon.

Morris *et al.* (38) paid particular attention to the effects of xenon anesthesia on electroencephalographic and blood gas measurements in human patients upon whom a variety of surgical procedures was performed. In all cases, a satisfactory degree of anesthesia was provided with xenon within a few moments. Excitement during induction was absent, and the patient recovered from the anesthesia within two or three minutes following cessation of xenon administration. Blood oxygen and carbon dioxide values were within normal ranges for all patients. The slower frequency of the dominant EEG activity associated with clinical anesthesia produced by xenon was appreciably faster than the slow activity described as being characteristic of cyclopropane and ether anesthesia. Only three levels of electroencephalographic activity were observed with xenon, rather than six or seven. These differences may have been due to variations in depths of anesthesia achieved with the different substances.

Since only a light plane of surgical anesthesia is attained with the administration of 80% xenon in oxygen at atmospheric pressure, the question arose whether the electroencephalographic patterns of profound xenon anesthesia would show a closer resemblance to the patterns obtained with other anesthetics. A series of characteristic encephalographic patterns which correlated with the depths of anesthesia and the concentration of the agents in arterial blood has been reported for ether, nitrous oxide and cyclopropane anesthesia (11, 19, 55, 62, 63). Within each series, the transition from one pattern to another was accompanied by a change in the dominant rhythm and in some cases by a significant change in voltage. During the stages of very deep clinical anesthesia electroencephalographic activity was minimal or completely suppressed. Pittinger *et al.* (51) undertook to determine whether the electroencephalographic patterns obtained with xenon at partial pressures sufficiently elevated to produce deep surgical anesthesia would show similar changes. As a result of these studies it was clearly established that inhalation of xenon under increased partial pressure led to profound anesthesia in monkeys. The anesthesia was accompanied by an early onset of apnea and a difference in the order of disappearance of reflexes with increasing depth of anesthesia as compared to the effects of ether in man.

The partial pressure of oxygen was maintained at approximately 200 mm of mercury and no signs of hypoxemia were observed, even though oxygen comprised only 9% of the inspired gas mixture.

Increasing pressures of xenon resulted in increasing depths of anesthesia and fairly consistent changes in the electroencephalographic activity. The major occurrences were a decrease in the dominant frequency and a decrease in the overall frequency range. A very slow background frequency would often be present, although a relatively faster frequency might be dominant. Monorhythmicity was either absent or occurred only at elevated pressures. Even at the highest pressures of xenon (3 atmospheres), burst suppression and total suppression were not observed, although the animals were relaxed and areflexic. The absence of any clear cut "levels" of electroencephalographic activity which paralleled "levels" of anesthesia, and the fact that in monkeys and in human beings the apparent clinical depth of anesthesia exceeded the electroencephalographic estimate of depth of anesthesia based on scales derived from ether or cyclopropane, might be taken to indicate that when electroencephalograms are used as estimates of the depth of anesthesia, the scales must be calibrated for each individual anesthetic agent.

The history of xenon as an anesthetic is unique in that much of the earliest work was carried out on man, the only species thus far investigated which can be anesthetized with the agent at atmospheric pressure. Deep anesthesia can be achieved in other mammals, but only with elevated pressures (2 to 3 atmospheres) of xenon. In addition to the work reported by Pittinger *et al.* (51) concerning anesthesia in monkeys subjected to high partial pressures of xenon, Domino *et al.* (16) reported anesthesia in dogs. During the induction and recovery periods in dogs exposed to 80% xenon–20% oxygen mixtures at elevated pressures, these workers were able to record from the olfactory bulb and amygdala hypersynchronous electrical waves, similar to those obtained in ether anesthesia. Neocortical recordings showed increased voltage and *delta* wave activity during deep xenon anesthesia. A marked second stage of excitement was observed, which could be diminished by increasing the partial pressure of xenon.

Xenon anesthesia thus seems like that produced by most inhalation anesthetics. The reasons for the differences in sensitivities to xenon and other anesthetics displayed by various animal species are not clear. Whether the answer depends on differences in the organization of the central nervous system, or merely reflects possible differences in transport and solubility of the gases must be investigated.

The greater solubility of xenon in water and in lipid as compared to nitrogen and oxygen led Pittinger *et al.* (52) to use xenon instead of air in encephalography in the hope that the rare gas would be eliminated much more rapidly than air and therefore lead to less postencephalographic distress. The quality of the roentgenographic results was similar to that obtained with air. There seemed to be no greater or lesser degree of distress during the encephalographic procedures with xenon than with air, although much of the xenon was eliminated

during the first 2 postencephalographic hours. Residual gas, probably a mixture of xenon, oxygen and nitrogen, was present after 24 hours. The absence of any clinical evidence of sedation or respiratory depression was noted by these authors.

Thus it seems established that xenon can produce anesthesia. Because of its chemical unreactiveness, which will be discussed in the following section, it is extremely unlikely that xenon can be involved in any of the usual types of metabolic processes. The gas therefore is an ideal tool to be used in the study of mechanisms of anesthesia produced by those agents generally considered metabolically inert, such as cyclopropane, nitrous oxide, and ethylene. Usual methods of analysis dependent upon compound formation are not feasible for xenon because of its aforementioned chemical unreactiveness. Hence, Featherstone *et al.* (20) employed radioactive xenon to measure the distribution of the gas in canine tissues after 20 minutes of xenon administration. The time period chosen represents twice the time of administration of xenon required in human patients before surgical procedures are begun and, as later shown by Pittinger *et al.* (49) is sufficiently long to permit saturation of most regions of the brain.

Twenty minutes of xenon administration to the dog produced definite narcotic effects although anesthesia was not achieved at atmospheric pressure. The investigation revealed no differences in the xenon content of tissue taken from five different areas of the brain. Kidney contained 90% as much xenon as the brain; liver and spleen contained 58% as much; striated muscle and perirenal fat contained 28% of the brain levels. The adrenal gland, however, was found to take up one and one-half to two times more xenon than any other tissue examined. Chenoweth (7) has observed a similar high adrenal concentration of anesthetic in the case of methoxyfluorane and several other anesthetic agents.

In 1954 Pittinger *et al.* (49) showed that 20 minutes of xenon inhalation produced an equilibrium state with respect to the concentration of gas taken up by the tissues, in an experiment in which they investigated the rate of uptake of radioactive xenon by various anatomical and functional areas of dog brain. Concomitant analyses of gas concentrations in the adrenal gland and several other tissues also were reported. The data obtained by this method after 2-minute and after 6-minute periods of rebreathing of a 80%:20% xenon–oxygen mixture from a closed system showed that there was a significantly slower uptake of xenon by the cerebral parietal cortex than by the thalamus, hypothalamus, caudate nucleus, or medulla oblongata. It is of interest to note that the xenon contents of the adrenals at the 2- and 6-minute periods were comparable to those of the brain tissues other than the cortex, but thereafter exceeded them, and that the rate of xenon-uptake by the kidney paralleled that of the cerebral cortex during the initial period but then dropped off. It appears from these studies that most areas of the brain are practically saturated with xenon within less than six minutes of inhalation of the gas at constant tension. This general conclusion was substantiated by the more recent work of Landau *et al.* (30b) and Kety (30a), who used a different inert diffusible substance and did not find a slower uptake by the cerebral cortex.

The unusual brevity of the induction and emergence periods of xenon an-

esthesia led Pittinger *et al.* (53) to study the kinetics of transfer of radioactive xenon between the blood and the brain of dogs during administration of the gas. The kinetic data were compared to those of transfer of radioactive chloroform-Cl^{38}. During the induction and emergence periods, biphasic curves were obtained for both anesthetics when the logarithm of the blood radioactivity was plotted against the time of inhalation. The rate constants which were calculated for the uptake and release of xenon and chloroform suggest that the brain functions as a 2-compartment system, represented presumably by the gray and white matter. The xenon and chloroform rate constants are similar and indicate that the transference and partitioning of these substances are essentially a flow-limited process not significantly influenced by any possible differences in permeability or diffusion rates *in vivo*. From the sustained high level of radioactivity in the recirculating blood, the authors calculated that approximately 25% of the chloroform of the pulmonary capillary blood crosses the blood-air interface in the lungs in each circulatory passage through the lungs, and that the same value for xenon is about 95%. The longer induction and emergence periods of anesthesia with chloroform as compared to xenon anesthesia seem to be due to the slower excretion of chloroform from the body.

In light of the work summarized thus far, it appears that the biological properties of xenon are not unlike those of nitrous oxide, ethylene, cyclopropane, and the other "inert" anesthetics. In addition, the chemical unreactivity of the rare gases makes the possibility of xenon's being metabolized extremely remote. These considerations did not rule out the possibility that xenon may interfere with normal tissue metabolism by some reversible physical mechanism. Quastel and Wheatly (61) had reported a depression of brain oxidation *in vitro* by "inert" nitrous oxide but this could not be substantiated by others (3, 4). Guinea pig brain respiration and oxidative phosphorylation *in vitro* in the presence of xenon and nitrous oxide were reported by Pittinger *et al.* (47) and Levy and Featherstone (33). These papers provide two instances in which satisfactory statistical methods were used in evaluating the influence of these gases on metabolism *in vitro*. In neither of these studies was there observed a difference in the respiration or oxidative phosphorylation of guinea pig brain tissue, using glucose or pyruvate as substrate. The authors concluded that the theories of anesthesia involving metabolic inhibition of glucose or pyruvate oxidation or the uncoupling of phosphorylation which are supported by data obtained only with barbiturates, are not applicable to the gaseous anesthetics, xenon or nitrous oxide, when the concentrations of these gases are those generally used in clinical anesthesia.

In contrast to these last findings, Cook and South (10, 67, 68) reported that xenon, as well as argon and helium, increased the rate of oxygen consumption and decreased the anaerobic glycolysis of slices of mouse brain, liver, or sarcoma *in vitro*. Hydrogen had little effect on oxygen consumption but decreased the glycolytic rate of liver and brain. These authors observed that there is a striking disparity between the physiological effects of these gases on intact organisms and their metabolic effects in *in vitro* systems, which cannot readily be explained by differences in the physical properties of these gases.

The reasons for these discrepancies in the reported effects of xenon and other inert gases on tissue metabolism are not discernible at present. The possibility of species differences, as well as the inherent imperfections of the method used must be considered.

A comparison of the effect of 11 inert gases on intact organisms and isolated nerve tissue was reported in 1954 by Carpenter (6). The author shows a correlation between the pressures of gases necessary to protect 50% of a group of mice from electroshock and the solubility of the gases in olive oil. Although the pressures of gases necessary to abolish the action spike in stimulated rat sciatic nerve were higher than those required to protect against electroshock, the ratio of these two pressures was reasonably constant for those gases tested. The author concluded that depression of the central nervous system in the intact animal and the blocking effect on the isolated nerve were dependent upon the same mechanism.

Serious study of the effects of xenon and other rare gases on lower forms of life has begun just recently. In 1961, Sears and Gittleson (66) showed that elevated pressures of xenon could cause "narcosis" in asynaptic organisms. These workers reported that helium, argon, and nitrogen, at pressures up to 1000 pounds per square inch (66 atm) had little effect on the movement of paramecia, but that 250 pounds per square inch (16 atm) of xenon produced changes in cell structure and movement, and finally, narcosis.

In studying the effects of six inert gases on oxygen-dependent radiosensitivity, Ebert *et al.* (17) found that xenon was most potent in preventing radiation damage to growing bean sprouts, hydrogen and helium least potent, and nitrogen, argon, krypton and cyclopropane intermediate.

Schreiner *et al.* (64) measured the rate of oxygen-uptake and linear growth of *Neurospora crassa* when the mold was grown in the presence of helium, neon, nitrogen, argon, krypton, or xenon (each containing 5% oxygen). The results of these experiments revealed that the growth rate of this mold is related linearly to the square root of the molecular weight of the chemically unreactive gas present. The growth rate of the mold in 95% helium was approximately 90% greater than in 95% xenon. The respiration rate was observed to be approximately parallel to the growth rate, and the authors took this as evidence that the gaseous environment did not interfere with cytoplasmic metabolism.

The Schreiner and the Ebert groups both suggested that the data obtained in their respective experiments could have been a result of the displacement of oxygen normally present at critical intracellular sites.

Since the review by Butler in 1950 (5), the biology of the rare gases has been discussed in additional reviews by Pittinger and Cullen (50) and Pittinger and Keasling (54).

II. THE CHEMISTRY OF THE RARE GASES

The critical role which xenon has, and will continue to have, in the formulation of an acceptable theory of anesthesia demands some brief review of its chemistry at this time.

TABLE 1

Some physical constants of the rare gas elements

	Ref.	Helium	Neon	Argon	Krypton	Xenon
Polarizability, $\times 10^{24}$ cc	34	0.1*	0.39	1.63	2.46	4.00
Melting point, 1 atm, °C	18		−248.7	−189.3	−157.1	−111.8
Boiling point, 1 atm, °C	18	−268.9	−245.9	−185.89	−152.9	−108
Density, 0°C, 1 atm, g/l	18	0.1785	0.90035	1.7839	3.708	5.851
Heat of fusion, cal/mole	18	—	80	281	360	548
Heat of vaporization, cal/mole	18	22	440	1557	2310	3020
Critical temperature, °C	18	−267.90	−228.6	−122.5	−63.8	+16.6
Critical pressure, atm	18	2.26	26.86	47.996	54.1	58.218
Critical density, g/ml	18	0.069	0.484	0.531	0.78	1.55
Solubility in water, ml gas at STP/g H_2O, 20°C	18	0.0088	0.015	0.038	0.063	0.125

* Calculated value.

The gaseous element of atomic number 54 is aptly named xenon, derived from the Greek word meaning "stranger," in that it comprises only 0.000006% of the atmosphere. The average atomic weight of the 26 isotopes of xenon is 131.3. The subatomic configuration of this colorless, odorless, tasteless gas places it as the fifth member of the group of elements known as the "chemically inert" or "rare" gases. Members of this group are characterized by the completeness of their outer electron shell and the absence, therefore, of any great tendency either to gain or lose electrons. Thus, xenon is a mono-atomic gas and the slight extent to which its atoms interact is reflected in the closeness of its low melting and boiling points (−111.8 and −108°C, respectively) (29).

The rare gas atoms are composed of a dense positive nucleus surrounded by a more diffuse cloud of electrons, with the centers of positive and negative electric charge coinciding. The atoms are thus spherical and possess no permanent electric dipole. When an electric field is introduced, either that of a permanent nature or the field of another atom or molecule temporarily in the vicinity, the repulsion of like charges distorts the electric symmetry of the rare gas atom and an induced dipole is established in which one portion of the atom becomes relatively positive and the other portion relatively negative. The ease with which an induced dipole is established in an atom or molecule in an electric field of given strength is termed "polarizability." Within any group of elements, increasing atomic weight is accompanied by increased polarizability, due to the larger number of electrons and their greater distance from the positive nucleus. Properties which are dependent upon intermolecular interactions, as those listed in Table 1, show an increase in magnitude with increasing atomic weight because of the greater polarizability of the larger atoms. Atoms of the heavier rare gases, especially xenon, may become polarized in an electric field of sufficient strength and interact with surrounding atoms or molecules. An example of this type of interaction is provided by the studies of Munson and Hoselitz

in 1939 (40). During studies on the mobility of lithium ions through xenon in an electric field, these investigators discovered that, at room temperature and low field strength/pressure values, xenon atoms clustered around and migrated with the lithium ions, probably in the ratio of two gas atoms per ion.

Although the atoms of the rare gases have generally been thought to be incapable of forming covalent, ionic, or hydrogen bonds with other atoms,[3] the term "chemically inert" is somewhat misleading as several associations of xenon (and other rare gases) with other substances have been described in the literature. These complexes, in general, fall into two classes, the clathrates and the van der Waals molecules. This latter group includes HgXe (28), and CsXe (70), which thus far have been detectable only by spectroscopic means. Due to their extremely short half-lives and their minute concentrations, the van der Waals molecules are of no biological interest at this time.

Inasmuch as clathrates have not received much notice in the biological sciences, it seems profitable to present a brief description of their general nature before discussing the particular properties of the inert gas clathrates.

Clathrates are physical associations of two (or more) types of molecules in which there may be very little actual attraction of the molecules of one species for those of the second. The molecules are held in association by the formation of a cage-like structure by one of the components around an atom or molecule of another species. The cages are formed by covalent compounds which, because of their chemical configurations, cannot close-pack. These compounds tend, instead, to form lattice structures containing cavities in which other molecules of critical sizes may be entrapped. Formation of these cages is a step-wise phenomenon resulting from the intermolecular hydrogen-bonding of the lattice-forming molecules. Bonds of this nature are relatively weak and easily broken. Although the presence of an entrapped molecule in the interior of the cage stabilizes the clathrate structure through van der Waals interaction with the molecules forming the cage structure, solution of the crystals is sufficient to disrupt the lattice and release the trapped molecule.

Clathrates may arbitrarily be divided into two classes, 1) those in which the lattice is composed of organic molecules (*e.g.*, phenol, quinol) and 2) hydrates, in which water forms the supporting structure. The latter will be discussed in greater detail as they seem to be more pertinent to biological systems.

Villard (72) was the first to describe the formation of a hydrate of a rare gas, when in 1896 he reported the production of argon hydrate by cooling water to 0°C under 150 atmospheres argon. Although he was not aware of the nature of

[3] Since the writing of this manuscript, the production of several true chemical compounds of xenon (and of radon) has been reported. Bartlett (1a) described the reaction of xenon with platinum hexafluoride to form $Xe^{+}[PtF_6]^{-}$ and Chernick *et al.* (7a) demonstrated the production of several xenon fluorides (XeF, XeF_2, XeF_3, XeF_4) and of radon fluorides when the rare gases were heated to 400°C in the presence of an excess of fluorine and then rapidly cooled. Xenon tetrafluoride is a colorless crystalline compound with a vapor pressure of approximately 3 mm Hg at room temperature. Until it is shown to be otherwise, xenon must still be considered as incapable of forming any but van der Waals bonds under physiological conditions.

the compound formed, he reported its dissociation pressure at 0°C and 8°C as being 105 and 210 atmospheres, respectively. In 1902, deForcrand (23) reported the formation of hydrates of nitrogen, methane, carbon dioxide, and nitrous oxide (23) but did not demonstrate the production of hydrates of the rare gases until 1923, when he described the hydrate of krypton and predicted that the hydrate of xenon should be more stable than that of either argon or krypton (24). In 1925, sufficient quantities of pure xenon were made available to him so that he was able to produce its hydrate and confirm his earlier prediction (25). The physical properties of the hydrates of the rare gases are summarized in Table 2; it may be seen that the stability of the hydrate increases with the polarizability of the entrapped atom. However, as pointed out by Nikitin (41), when the series of gases is extended to include others besides the rare gases, no constant proportionality between polarizability and hydrate stability can be expected, although the general trend is still evident. Factors such as atomic radius or molecular size undoubtedly have some influence on hydrate stability.

The hydrates of helium and neon have not yet been reported but several attempts at their production are described in the literature. DeForcrand (25) reported the failure of these hydrates to crystallize at 0°C under pressures up to 260 atmospheres, and Nikitin (42) estimated that the dissociation pressure of neon hydrate at 0°C will be several thousand atmospheres. Nikitin was successful in crystallizing mixed hydrates of SO_2-Ne and SO_2-He, which contained 1.2% neon and less than 0.2% helium, respectively.

As is characteristic of clathrate compounds, the form of the hydrate crystal is determined by the configuration of the lattice water molecules, and thus many of the gaseous hydrates are isomorphic. The shape and polarity of water molecules prevents their close-packing and, by means of intermolecular hydrogen-bonding, the water molecules in the lattice of the gaseous hydrates form three cages of different sizes and shapes. The smallest of these cages is the dodecahedron, consisting of twelve regular pentagons. Intermediate is the tetrakaidecahedron, a fourteen-sided cage formed by twelve regular pentagons and two opposing hexagons. The largest cage is the hexakaidecahedron, with twelve pentagonal and four hexagonal sides. The hydrates of the smaller gases, such as xenon, are composed of dodeca- and tetrakaidecahedra. The tetrakaidecahedron can accommodate entrapped molecules with van der Waals diameters up to 6 Å, while the dodecahedron may be occupied only by molecules with diameters not over 5 Å. Xenon, the van der Waals diameter of which is approximately 4 Å, can therefore fit into both types of cages. The unit cell of xenon hydrate, as measured from one xenon atom to its nearest neighbor xenon atom, has an edge of about 12 Å and is composed of 46 water molecules, forming six tetrakaidecahedra and two dodecahedra (46).

The larger gases, such as chloroform, form hydrates composed of dodecahedra and hexakaidecahedra, in which only the hexakaidecahedra can accommodate the chloroform molecules. The dodecahedra may be filled with oxygen, nitrogen, or other small gas molecules which may be present, including many of the

TABLE 2

		Ref.
ARGON HYDRATE		
Composition	$A \cdot 5H_2O$	24
Dissociation pressure,		
0°C	98.5 atm	25
0°C	105 atm	72
8°C	210 atm	72
Dissociation temperature, 1 atm	−39°C	41
Calculated heat of formation	14.855 cal	24
KRYPTON HYDRATE		
Composition	$Kr \cdot 5H_2O$	24
	$Kr \cdot 5\frac{3}{4}H_2O$	1
Dissociation pressure 0°C	14.5 atm	24
Dissociation temperature, 1 atm	−25°C	41
Calculated heat of formation	14.712 cal	24
Critical temperature	12.5–13°C	24
KRYPTON DEUTEROHYDRATE		
Composition	$Kr \cdot 6D_2O$	27
Dissociation pressure, 8.3°C	34 atm	27
Calculated heat of formation	16.30 cal	27
XENON HYDRATE		
Composition	$Xe \cdot 6H_2O$ or $Xe \cdot 7H_2O$	25
	$Xe \cdot 5\frac{3}{4}H_2O$	45
Dissociation pressure,		
0°C	1.15 atm	25
8°C	2.987 atm	25
Dissociation temperature, 1 atm	−1.13°C	25
Calculated heat of formation	18.266 cal	25
Critical temperature	24°C	25
XENON DEUTEROHYDRATE		
Composition	$Xe \cdot 6D_2O$	27
Dissociation pressure, 8°C	3.25 atm	27
Calculated heat of formation	18.23 cal	27

smaller anesthetic agents. Excellent illustrations and more complete descriptions of the structures of the gaseous hydrates have been presented by Pauling (45).

Waller (73) reported in 1960 the formation of an interesting series of mixed hydrates which illustrate the role of both increasing polarizability and increasing molecular size in lending stability to the hydrate lattice. Double hydrates of some of the rare gases and several organic compounds were made; the dissociation temperatures at which the entrapped gas molecules were released at 1 atmosphere are listed in Table 3. For any of the three rare gases, the stability of the hydrate increases as the molecular size of the trapped organic molecule increases. Similarly, in any one group of organic hydrates, the stability of the double hydrate increases with increasing atomic weight and polarizability of the rare gas. The data illustrate an additional point when they are compared to the dissociation temperatures of the pure hydrates of argon, krypton, and

TABLE 3

*Temperature at which decomposition pressure is one atmosphere (°C)**

Inert Gas	Organic Liquid			
	Acetone	Methylene dichloride	Chloroform	Carbon tetrachloride
A	−8	−7	−4.8	−1.6
Kr	−5	+6.2	+9	+11.3
Xe	+3	+8.6	+10.9	+13.7

* (Reprinted with the permission of *Nature*, from Waller, J.: Nature, Lond. **186**: 429–431, 1960.)

xenon, as listed in Table 2. In each case there is a significant increase in the stability of the hydrate lattice brought about by the presence of the organic component. This would be in accord with Pauling's suggestion that anesthesia is caused by the changes in electrical properties of the central nervous system due to the formation of mixed hydrates of anesthetic agent and brain constituents, possibly the proteins (46).

Clathrate compounds of inert gases in which the lattice is formed by organic molecules have been studied by a number of investigators. Powell (59) described quinol clathrates of argon and of krypton in 1949, and of xenon in 1950 (60), all having the general formula $X \cdot 3C_6H_4(OH)_2$. Nikitin and Koval'skaya (43) reported the formation of phenol clathrates of the rare gases, radon and xenon, the latter compound having a dissociation temperature of 4°C at 1 atmosphere pressure. The crystal structure of the xenon clathrate, Xe·3PhOH, was studied by von Stackelberg *et al.* (69).

Cramer and Henglein (13) reported the formation of inclusion compounds of cyclodextrins and a number of gases, including krypton and xenon. These compounds are similar to the clathrates in that the gas is held in association with the carbohydrate within a cavity of the cyclodextrin molecule. The difference between inclusion compounds and clathrates is that the cavity of the cyclodextrins is formed by the cyclic union of 6, 7, or 8 glucose molecules by 1,4α-glycosidic linkages. Solution of the crystals releases the included gas molecule but, due to the greater strength of the covalent glycosidic bond, the lattice is not disrupted in solution. In view of the observations that cyclodextrins possess certain enzyme-like characteristics, Cramer (12) suggests that the formation of inclusion compounds may have a role in the biological world, possibly in some types of enzyme-substrate complexes.

The realization that more gas is soluble in whole blood, when the latter is equilibrated with a number of different inhalation anesthetics, than can be accounted for by the water and lipid content of blood, led Featherstone and his colleagues to investigate the possible associations of inert gases with proteins, the only other major blood constituent. A preliminary paper appeared in 1961 (21) which reported that the solubility of cyclopropane increased linearly with increasing concentrations of human serum albumin when the protein solutions

were saturated with the anesthetic at atmospheric pressure. Similar studies on the associations of solutions of crystalline bovine serum albumin and hemoglobin with cyclopropane, nitrous oxide, and other inert gases have since been carried out and reports of these are in preparation for publication. The suggestion that anesthetic agents may associate reversibly with proteins is not a new one but dates back as far as 1904 and 1905, when Moore and Roaf (36, 37) reported experiments which demonstrated that chloroform, ethyl ether, and several other anesthetic agents are more soluble in serum and hemoglobin solutions than in water or saline solution. The solubility of chloroform was also greater in tissue homogenates than in water or saline solution, and the increase in all three systems, *i.e.*, serum, hemoglobin, and tissue homogenates, could not be accounted for by the amount of lipid present. From these observations, and additional measurements which can be found in their papers, Moore and Roaf concluded that anesthetics are able to form unstable chemical "compounds" or physical aggregates with proteins. These anesthetic-protein complexes appeared to be stable only as long as the pressure of the anesthetic in solution was maintained.

An interesting theory was proposed by Östergren (44) in 1944 in which he suggested that colchicine mitosis, chromosome contraction, and narcosis are all caused by a single mechanism, that of interaction of a chemically unreactive compound with the lipophilic side-chains of proteins so as to cause a change in the shape of the protein molecule. Although Östergren's concept of narcosis is perhaps more encompassing than that which is generally held, his theory is an interesting one and in accord with the observations of the present authors that inert gases do interact with lipid-free proteins. Gavaudan *et al.* (15, 26, 58) enlarged upon Östergren's hypothesis by proposing that anesthetics "precipitate" the lipids dissolved within the lipophilic areas of proteins.

More recent notice of the associations of inert gases with protein include Kety's remark (30) in an article on the exchange of inert gases at the lungs and tissue, that increased solubility of gases in blood *might* be due to proteins. Lesser *et al.* (32) in 1952, while using cyclopropane to measure total body fat, mentioned that protein solutions adsorb a great deal of the anesthetic. Measurements were made of the solubility of cyclopropane in serum and hemoglobin solutions in order to correct their calculations for non-lipid adsorption of gas. In 1958 Possati and Faulconer (56) investigated the relationship of hemoglobin concentration and cyclopropane solubility in whole blood. Their data revealed a decrease in cyclopropane solubility as whole blood is diluted with serum, but no direct evidence of the association of the gas with hemoglobin was demonstrated by these authors.

No attempt has been made here to present a complete picture of all the known types of complexes or associations of the rare gases with other substances. The argon-boron trifluorides, for example, are known to exist but can not be expected to have any role in the production of anesthesia or other biological effects. An excellent two-volume treatise on the history, occurrences, and properties of the rare gases, edited by Cook (9) in 1961, is recommended to those

interested in further reading in this field. The associations mentioned, however, will emphasize the contention that the term "chemically inert," usually used to describe the behavior of the rare gases, is a misleading one.

III. THEORIES OF ANESTHESIA

In relating the effectiveness of different inhalation anesthetics to one another, it is difficult to avoid the use of the word "potency." Most often it has been used to compare the concentrations of various agents necessary to cause the loss of spontaneous movement of an organism when the agents are applied to the environment external to the organism. The major difficulty with this type of experiment is that the concentration of the agent in the external environment may in no way reflect the concentration of agent at the site of action *in vivo*. Present day ignorance of the locale of the site of action merely compounds this difficulty. A partial solution to this dilemma was offered by Ferguson (22) in 1939 when he pointed out that, since maintained anesthesia is an equilibrium condition, the problem may be considered from the thermodynamic viewpoint.

Since the thermodynamic activity[4] of a substance is equal in all phases of an equilibrium system, measurement of the activity of the anesthetic in the environment external to an anesthetized organism is an index of the activity of that agent at the site of action, although the location of that site of action is unknown. The use of the concept of thermodynamic activity has several advantages when applied to biological systems. Activity is a quantity which has been adjusted to correct for molecular deviations from ideality and, thus, is a more accurate measure of those molecules which are free to exert their biological effect. It is obvious, therefore, that a comparison of potencies of anesthetic agents is much more meaningful in terms of "activities" rather than "volumes per cent" or "moles per liter." Recalculation by Ferguson of the data from the older literature revealed several interesting points. He showed that isonarcotic vapor concentrations expressed as activity vary within a narrower range (0.01 to 0.07) than when expressed as volumes per cent (0.05 to 100%). It seemed, therefore, that a great many chemically unrelated compounds were nearly equally potent in producing anesthesia. Ferguson, however, placed more importance on the observation that as larger molecules in a homologous series are studied, greater and greater activity is found to be required to produce isonarcosis, until a member of the series is reached where the absolute solubility or vapor pressure of the substance is so low as to prevent sufficient quantities from being introduced into the environment. Ferguson suggested that this is the reason why higher members of homologous series are usually not anesthetics.

Brink and Posternak (2), in 1948, agreed with Ferguson that the measure of

[4] The thermodynamic activity is a measure of that portion of the molecules of a given substance which is not involved in nonspecific interactions with other molecules of the same substance or with solvent molecules, *i.e.*, those molecules which are free to exert a specific effect. In biology, thermodynamic activity is usually estimated from the ratio of the partial pressure of the substance at some biological endpoint divided by the vapor pressure of the pure substance at the same temperature (P_t/P_o).

an anesthetic *in vivo* is more meaningful in terms of thermodynamic activity, and pointed out that many anesthetics are soluble in olive oil to similar degrees, but that this correlation in no way supports or detracts from the hypothesis that anesthesia occurs *in vivo* in a lipid phase. Similarities in concentrations in *any* solvent merely reflect similarities in activity coefficients.

Brink and Posternak also agreed with Ferguson that two apparently conflicting trends seem to result when narcosis is considered in thermodynamic terms. For a great variety of anesthetic agents, equal degrees of narcosis do seem to occur at equal thermodynamic activities. However, the fact that for isonarcosis, increasing activity is needed within some homologous series must also be explained. The answer apparently lies in differences in the biological system being tested—a factor which had been totally ignored in earlier work. Posternak and Larrabee (57) were able to show that even within the same tissue of a single animal, the same anesthetics may act differently. In measuring the reversible suppression of transmission of nerve impulses in the stellate ganglion of the cat, these authors reported that those fibers which synapsed in the ganglion required fairly constant activities of various anesthetics to suppress transmission, while increasing thermodynamic activities were required to block axonal conduction in the non-synapsing fibers. The significance of these findings seems important in view of the scarcity in the literature of extensive studies of inhalation anesthetics on one test organism. In the formulation of any extensive theory of anesthesia it has been necessary to compile data from different species and from experiments performed under different conditions. It may well be that many of the deviations among anesthetics encountered in the theories of anesthesia thus far proposed arise from differences in the test systems employed, and are not inherent in the anesthetics themselves. At present, it may be concluded only that the test situation under consideration determines whether isonarcosis does or does not occur at equal thermodynamic activities.

The thermodynamic approach has, and will continue to have, an important role in the study of anesthesia. But thermodynamics, by its very nature, is not concerned with mechanisms of action; it is a study of the differences in energy states and is independent of the means by which these states are achieved. Thermodynamics alone cannot offer a mechanism of action of anesthesia but can serve as the basis of a theory, as seen in the paper published by Mullins (39) in 1954. Starting from the calculation of thermodynamic activity at narcosis, as suggested by Ferguson, Mullins proposed that the size of the various anesthetic molecules is a factor equal in importance to the number of molecules which reach the site of action. From a consideration of the studies of narcosis in the literature, Mullins inferred that the site of action is situated in a highly polar, non-aqueous phase of the cell which he termed the "membrane," and he suggested that narcosis occurred when a critical fraction of space within this "membrane" was occupied by inert molecules. "Narcosis by chemically inert molecules appears to take place when a constant fraction of the total volume of some non-aqueous phase in the cell is occupied by narcotic molecules. If the narcotic behaves ideally in this non-aqueous phase, the thermodynamic activity

of the narcotic multiplied by its molal volume is a constant, about 1 ml./mole." In terms of physiological mechanisms, Mullins postulated that occlusion of this critical portion of free space in a membrane might interfere with permeability to ions or molecules necessary to cell function.

While it can in no way be considered a "theory" of anesthesia, a paper by Wulf and Featherstone (74) in 1957 re-emphasized the probable importance of molecular volume in determining "anesthetic potency." The van der Waals constants, which are indices of the volume occupied by the atoms or molecules and of the attractive forces between them, were calculated and compared to anesthetic potency. A general trend was apparent in that increased potency seemed to correlate with increase in the magnitude of the van der Waals correction factors. An additional correlation was noted in that anesthetic potency seemed to be related to molar refraction, a measure of the polarizability of molecules.[5] This parallelism in correlations is not surprising in view of the interdependence of polarizability, molecular volume, and molecular attraction.

A similar correlation between anesthetic potency and molar refraction was noted by Pauling (46) in 1961 when he compared the dissociation pressures of the hydrates of anesthetic gases and the partial pressures of anesthetics necessary for narcosis, and showed that these were both related to the molar refraction. However, Pauling stated that the mechanism of anesthesia could not be the simple formation of hydrate crystals in the brain, as none of the hydrates of anesthetic gases thus far studied is stable at physiological temperature and pressure. Even at 0°C, the hydrate dissociation pressures for a number of agents are over 1 atmosphere, and at 37°C several atmospheres pressure would be necessary to stabilize nearly all the anesthetic hydrates. In order to account for the formation of hydrate microcrystals, Pauling suggested that some substance, or substances, in addition to the anesthetic molecules, are able to take part in hydrate formation and add stability to the hydrate structure. Because some smaller quaternary nitrogen compounds form hydrates with structures similar to those of the anesthetics but with much greater stability, Pauling proposed that the charged side-chains of proteins and solutes of the brain fluid are capable of stabilizing the hydrate microcrystals at temperatures not much lower than normal mammalian body temperature, perhaps as high as 25°C. The anesthesia which results from hypothermia and hibernation could then be explained as being caused by the formation of hydrate microcrystals of amino acids and charged side-chains of proteins in the synaptic regions of the brain.

Pauling argued that molecules of anesthetic, when present, would form mixed hydrates by occupying some of the chambers in the hydrate structure not already occupied by protein side-chains or other brain constituents. Mixed hydrates, as discussed previously, are more stable than those in which only one type of molecule is entrapped. The formation of such mixed hydrates would allow the microcrystals to exist at 10° to 15°C higher than in the absence of the gas. According to Pauling (46) the increase in ". . . the impedance offered by the network to the electric waves . . .", which would occur as a result of the formation of these microcrystals, would appear as anesthesia.

[5] Molar refraction = $4/3\ \pi\ N \times$ polarizability, where N is Avogadro's number.

The observation that many of the gases, the hydrates of which he was studying possessed anesthetic properties led Miller (35) also in 1961 to consider the possible role of hydrates in anesthesia. Miller showed that the ratio between the partial pressure of a given gas at anesthesia and the decomposition pressure of its hydrate at 0°C was relatively constant (0.1 to 0.7). By calculating the dissociation pressures which the hydrates would have at 37°C, Miller estimated that 34 times the pressure needed at 0°C is necessary for the existence of the hydrates of the smaller gases at body temperature, assuming that nothing in the brain fluid contributes to the stability of the hydrate; for the larger gases, approximately 1000 times the pressure would be necessary. Since several of the gases would at 37°C liquefy at much lower pressures than those calculated to be necessary to stabilize their hydrates, and because the pressures needed for anesthesia are even lower, Miller concluded that it would appear impossible that pure crystalline hydrates could form during anesthesia.

As Miller pointed out, hydrate formation is only one indication of the interaction of the inert gases with water in aqueous solution. Several thermodynamic properties of aqueous solutions of gas indicate that a portion of the gas molecules orient water around themselves, much as proteins do, so that the molecules are surrounded by a shell of water which is in a more highly ordered state than is the remainder of the water. This highly structured water is usually referred to as "iceberg" or "ice cover." In addition, liquid water is known to have some type of ice-like structure, which may or may not be like the structure of the hydrates, but with "free" water molecules occupying the center of the cages (45). Assuming that the cavities in the ice cover are the same as those of the hydrate structure, Miller derived equations which showed that the proportion of a hypothetical surface covered with structured water (ice-cover) in the different stages of anesthesia is proportional to the pressure of the anesthetic present. Past failures to show the production of crystalline hydrates of the larger anesthetics, such as diethyl ether, do not detract from Miller's arguments. The unusually high solubility of diethyl ether in water indicates that some interaction exists between the water and the organic molecules, and it seems likely that ether molecules are surrounded by "icebergs."

No extensive discussion of the manner in which the increase in structured water (ice cover) may produce anesthesia was presented. However, Miller suggested that an increase in structured water might have one or more of several effects: 1) to lower the electrical conductance of brain tissue, as Pauling proposed, 2) to "stiffen up" lipid or other membranes, or 3) to "plug up" membrane pores, reminiscent of Mullin's theory.

When comparing the two hydrate theories of anesthesia, that of Pauling and that of Miller, it might appear at first that they are in disagreement. Upon closer examination of each theory, any apparent differences resolve themselves into a problem of semantics. What Pauling calls "microcrystals" are extremely similar, if not identical, to the "ice cover" with which Miller is concerned. The sizes these aggregates attain, as conceived by each author, may be the only difference. The process of crystallization is a continuous one, beginning when two atoms or molecules condense and continuing until the aggregate becomes

too large to remain in solution. The visible solid which then precipitates is commonly called a "crystal." The distinction, however, is an arbitrary one. Under the magnification of an electron microscope, the limiting minimal size an aggregate must attain before it is designated as a "crystal" is greatly reduced. It appears, therefore, that Pauling and Miller have proposed similar mechanisms, perhaps at slightly different places on the crystallization-size continuum.

In order not to create the impression that all modern investigators in the field of anesthesia have abandoned the Meyer-Overton approach to the problem, it seems advisable to describe some of the more recent work dealing with lipid-solubility.

The ability of several inert gases to reverse oil-in-water emulsions was studied in 1957 by Sears and Fenn (65). One hundred seven, and 53 atmospheres of helium and nitrous oxide, respectively, did not reverse olive oil-in-water emulsions to water-in-oil emulsions; 100 atmospheres of nitrogen, 60 atmospheres of argon, and 0.01 atmosphere of carbon dioxide were sufficient to reverse emulsions in the majority of experiments attempted. Two properties required of a substance to reverse oil-in-water emulsions are higher solubility in oil than in water and the ability to lower the interfacial tension of the oil phase to a value lower than that of the water phase. The potency of argon, nitrogen, and helium in reversing oil emulsions correlated well with their lipid solubilities, but nitrous oxide seemed to require higher pressures than would be predicted from its oil solubility. The extremely low concentrations of carbon dioxide necessary to reverse emulsions were ascribed to its chemical reactivity. Sears and Fenn suggested that narcosis could be the result of the accumulation of sufficient inert gas in the lipids of a membrane so as to cause the lipid phase to be more continuous, and hence block the transmission of impulses across the synapse.

The interaction of inert gases with lipids was studied in another model system by Clements and Wilson (8). Monomolecular films of stearic acid, cholesterol, lecithin, and a lipoprotein extracted from beef lung, were spread on the surface of distilled water. Measurements of the surface tension were made before and after the introduction of anesthetic gases. In all cases tested, the surface tension was lower when the films were equilibrated with an anesthetic than when exposed to air, and it returned to the initial air value when the anesthetic was removed. At a partial pressure of nitrous oxide equal to that necessary for anesthesia, 0.16×10^{-10} mole of gas was adsorbed per square centimeter of film area (0.16 Gibbs). Calculating the partial pressures of other agents necessary to effect the same change in surface tension as produced by 0.16 Gibbs, Clements and Wilson showed that a good correlation exists between the potency of 11 anesthetic agents and their affinities for surface films; they interpreted this relationship as indicating that inert gases can significantly alter lipoprotein membranes *in vivo*.

IV. IMPLICATIONS OF THE CHEMISTRY OF XENON IN RELATION TO THE MECHANISM OF INERT GAS ANESTHESIA

The discovery of the anesthetic properties of xenon has stimulated a great deal of interest in the relation of chemical structure and anesthetic activity,

in that it demonstrated that the property of causing anesthesia cannot be assigned to any particular chemical group. Xenon, being an element incapable of forming ionic, hydrogen, or covalent bonds with other atoms under physiological conditions, restricts all postulated mechanisms of inert gas anesthesia to the "physical" level of molecular interactions. Of the four types of bonds important in biology, the covalent, the ionic, and the hydrogen bond are the strongest[6] and are generally called "chemical" in nature; the van der Waals bond is extremely weak and is usually considered as a "physical" attraction. This distinction between chemical and physical bonds is an arbitrary one and without real meaning. Under physiological conditions, a rare gas atom can interact with other atoms only through the production of a dipole induced within itself in the presence of an electric field.[7] Van der Waals forces are largely a result of the mutual production of dipoles in two atoms when they are sufficiently close to one another to cause the electrical field of each atom to influence the electrons and nucleus of the other.

Since the only possible interaction in which the rare gas molecules can participate *in vivo* is that of the induced dipole (of which the van der Waals attractive forces are a special type), the factors which influence these interactions must be important in the production of anesthesia. As discussed previously, the mole volume and the polarizability of the atoms are the major determinants influencing the strength of the van der Waals forces. Polarizability[8] and the effective mole volume,[9] as shown in Figure 1, are intimately related, and it is extremely difficult to evaluate the contribution of one without the other. If we assume that the inert gases, such as cyclopropane and nitrous oxide, produce anesthesia in the same manner as xenon, we are forced to conclude that mole volume and polarizability play a major role in determining whether any metabolically unreactive compound has anesthetic properties. If either of these two factors is compared separately to some measure of anesthetic potency, as shown

[6] Approximate relative bond strengths: covalent, 40–140 Kcal; ionic, 5 Kcal; hydrogen, 2–5 Kcal; van der Waals, 0.5 Kcal.

[7] See section II.

[8] Polarizability—calculated from the relationship:

$$\tfrac{4}{3}\pi N\alpha = \frac{n^2 - 1}{n^2 + 2} \cdot \frac{M}{d}$$

where α = electron polarizability; N = Avogadro's number; n = index of refraction; M = molecular weight; d = density.

[9] Effective mole volume is equal to the "b" factor in the van der Waals equation and is here either taken from reference 74 or calculated by the following relationship:

$$b = \frac{RT_c}{8P_c}$$

where R = gas constant and T_c and P_c = critical temperature and pressure, respectively. The effective volume is, therefore, the volume which 6.02×10^{23} molecules of a given substance appear to occupy. This value has also been called "excluded volume" or "covolume" by other authors.

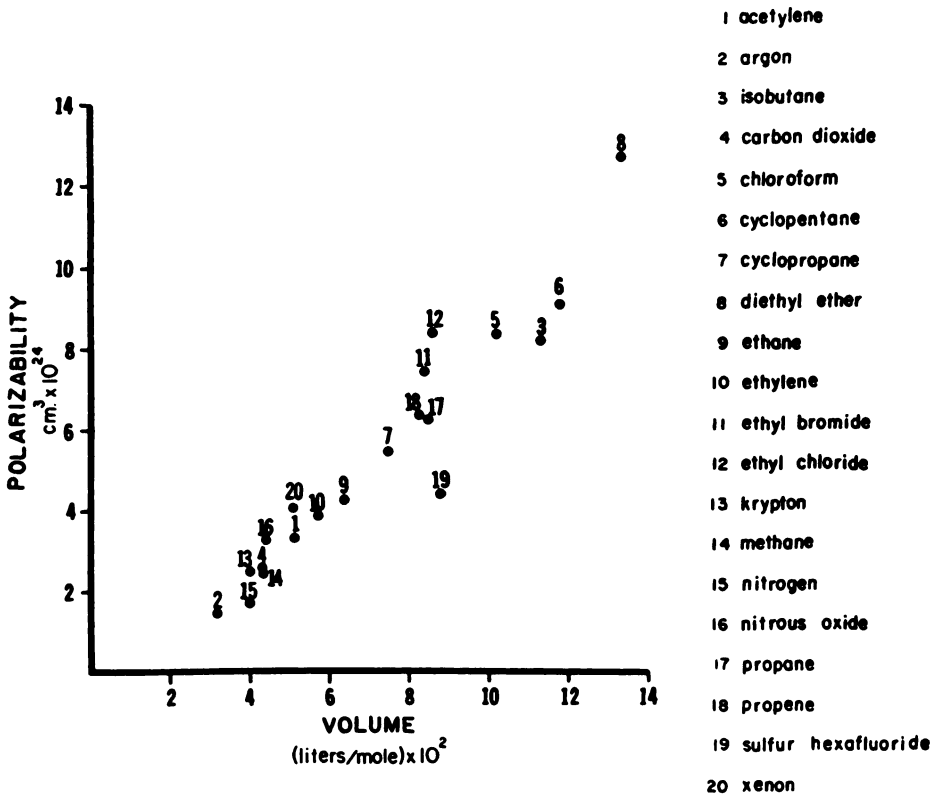

FIG. 1. The polarizabilities of 20 gaseous anesthetics plotted against their mole volumes.

in the top and middle of Figure 2, trends may be observed, but the closeness to linearity of the correlations leaves much to be desired. If both these factors are considered simultaneously, by plotting the ratio of polarizability to volume against the logarithm of the partial pressure (in mm mercury) necessary for anesthesia, as in the bottom of Figure 2, absolute linearity still does not result, but the deviations are reduced. Several compounds which would not be expected to act as the inert gases have been included for comparison. Carbon dioxide, because of its chemical reactivity, cannot be considered as an "inert gaseous anesthetic." The high solubility of diethyl ether suggests that this compound may form hydrogen bonds with water; accordingly, perhaps it should also not be included in the present definition of "inert gas." Of the remaining compounds which are anesthetics below a partial pressure of 760 mm mercury, the ratio of polarizability to volume falls within the range of 0.68 (ethylene) to 0.89 (ethyl bromide); those agents which require greater than 1 atmosphere pressure to produce anesthesia have smaller polarizability-to-volume ratios. Non-anesthetic gases, such as helium, neon, and hydrogen, could obviously not be plotted in Figure 2, but have extremely small polarizability-to-volume ratios (He, 0.09; Ne, 0.23; H_2, 0.29).

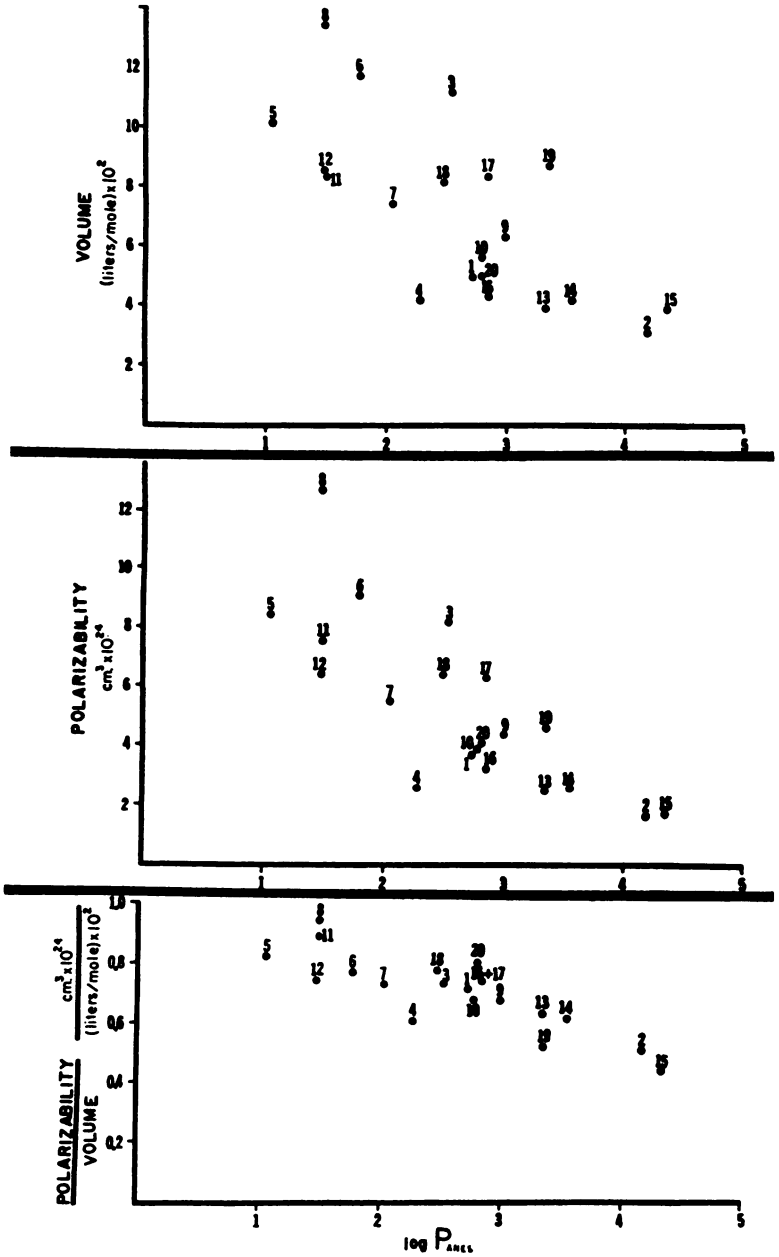

FIG. 2. (*top*) The effective mole volume of the same 20 anesthetics as shown in Figure 1 plotted against the logarithm of the pressure (in mm mercury) necessary for "anesthesia." (*middle*) The polarizability of the same 20 anesthetics as shown in Figure 1 plotted against the logarithm of the pressure (in mm mercury) necessary for "anesthesia." (*bottom*) The ratio of polarizability to volume of the same 20 anesthetics as shown in Figure 1 plotted against the logarithm of the pressure (in mm mercury) necessary for "anesthesia."

Two explanations for the minor deviations from strict linearity readily come to mind. First, no thorough study on one test organism of all the compounds listed in Figures 1 and 2 exists. Values used in clinical anesthesia in man have, of necessity, been compared to narcosis observed in mice. Recalling the work of Posternak and Larrabee (57), this may not be a valid comparison. Secondly, perhaps an additional, as yet unidentified, fundamental property, in addition to polarizability and effective mole volume, plays an important role in anesthesia.[10]

These two properties, polarizability and mole volume, have been considered at least indirectly in most theories of anesthesia. Many of the physical properties which have been correlated with anesthetic potency in the past are merely reflections of the intermolecular van der Waals attractions and hence, of the sizes and polarizabilities of the agents. The magnitude of lipid-solubility (Meyer-Overton theory) (5) is a measure of the van der Waals attraction between the gas and the lipid molecules. The extent of adsorption of an agent at a membrane or interface [Traube, Warburg, Lillie theories (5), and Clements and Wilson] is a measure of the van der Waals interactions between the molecules of the interface and those of the agent. The thermodynamic activity at narcosis (Ferguson, and Brink and Posternak theories), estimated from the ratio of the partial pressure at narcosis to vapor pressure of the pure substance, is related to polarizability, since the vapor pressure of the pure substance is dependent upon the attraction of the substance's molecules for one another. The degree to which an entrapped molecule stabilizes a clathrate structure (Pauling theory) is determined by the strength of its van der Waals attraction to the molecules forming the cage, and the molecular volume of the molecule determines whether it can fit inside and remain entrapped within the cage. It would seem to the present authors, therefore, that many investigators in the field of anesthesia have been rediscovering indirectly the same phenomenon, when correlations between any one physical property and anesthetic potency have been demonstrated.

At our present level of knowledge about factors which govern distribution *in vivo*, and the electrical properties of atoms which govern solutions of substances in polar and non-polar solvents, it would seem naïve to suggest that any *one* property determines the anesthetic potency of a given substance. Many processes must occur between the introduction of an anesthetic molecule into the lung alveoli and its arrival at the final site of action. For each of these processes to operate under optimal conditions, a different set of "chemical" and "physical" properties would be required of the anesthetic. The possession of any *one* property by a molecule may be a deterrent, an advantage, or immaterial to its ability to produce anesthesia in the intact organism. For example, the possession of a permanent dipole, such as nitrous oxide has, enhances the molecule's penetration into polar areas (it has a relatively high water solubility), decreases its entrance into lipid phases (it has a lower oil/water ratio than many

[10] Other data in the literature, such as lipid solubility, thermodynamic activity, *etc.*, would not seem very useful, because, as will be discussed later, polarizability, to a great extent, determines these factors.

anesthetics), and would seem immaterial in its role at the final site of action (many anesthetics, like xenon, have no permanent dipole).

As a final comment, it appears to the authors of this review that the correlation between the abilities of many substances to produce anesthesia and their solubilities in lipoid materials has diverted the attention of most investigators in the field of mechanisms of anesthesia for over 50 years. The recent re-emphasis of the suggestions that water, or proteins, or both, may have an important role in the mechanism of anesthesia provides an opportunity for a similar situation to develop. It is sincerely hoped that this does not occur. Any lengthy arguments about whether anesthesia occurs in lipid, water, or protein are, at this point, futile and meaningless. The modern concept of cellular membranes is based largely on monomolecular layers of cholesterol and phospholipids interspersed between areas of protein and water. If an anesthetic molecule occupies the area between a lipid monolayer and a hydrated protein molecule, is it "dissolved" in lipid or is it "interacting" with protein? At the level of monomolecular layers, such discussions become absurd. Nowhere in the body, with the possible exception of depot fat and glycogen storage granules, does any biological material exist in pure form. The biologist studies any cellular material in purified systems merely to evaluate its role in the more complex cellular system. Any effect he may discover in the test tube must always be related to the behavior of the intact cell, tissue, or organism, where the materials with which he is dealing exist, not as crystalline bovine serum albumin or olive oil, but as aqueous solutions of heterogeneous lipoproteins, nucleoproteins, and mucopolysaccharides—to mention merely the more obvious complexes existing in nature.

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