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**Research Article**

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# Influence of Steady-State Alterations in Acid-Base Equilibrium on the Fate of Administered Bicarbonate in the Dog

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A B S T R A C T Previous workers have shown that metabolic acidosis increases the apparent space through which administered bicarbonate is distributed. This finding has been ascribed to the accompanying acidemia and to the consequent availability of a large quantity of hydrogen ion that accumulates on nonbicarbonate tissue buffers during the development of acidosis. To test this hypothesis, bicarbonate space was measured in dogs with a broad range of steady-state plasma [$HCO_3^-$] in association with alkalemia as well as with acidemia. Appropriate combinations of pH and plasma [$HCO_3^-$] were achieved by pretreating the animals to produce graded degrees of each of the four cardinal, chronic acid-base disorders. Metabolic acidosis ($n = 15$) was produced by prolonged HCl-feeding; metabolic alkalosis ($n = 17$) by diuretics and a chloride-free diet; and respiratory acidosis ($n = 9$) and alkalosis ($n = 8$) by means of an environmental chamber. Animals with normal acid-base status ($n = 4$) were also studied. Sodium bicarbonate (5 mmol/kg) was infused over 10 min to the unanesthetized animals; observations were carried out over 90 min. The results obtained from animals with metabolic acid-base disturbances demonstrated an inverse relationship between bicarbonate space and initial plasma pH, con-

firming the previous findings of others. By contrast, the results obtained in animals with respiratory acid-base disturbances demonstrated a direct relationship between bicarbonate space and initial plasma pH. The pooled data revealed that bicarbonate space is, in fact, quite independent of the initial pH but is highly correlated with the initial level of extracellular [$HCO_3^-$]; dogs with low extracellular [$HCO_3^-$] ($\simeq 10$ meq/liter) whether acidemic or alkalemic, have a bicarbonate space that is 25% larger than normal and some 50% larger than in dogs with high extracellular [$HCO_3^-$] ($\simeq 50$ meq/liter). We conclude from these results that the increased bicarbonate space in metabolic acidosis (and respiratory alkalosis) does not reflect the availability of more hydrogen ions for release during bicarbonate administration, but merely evidences the wider range of titration ($\Delta$pH) of nonbicarbonate buffers that occurs during alkali loading whenever plasma [$HCO_3^-$] is low.

## INTRODUCTION

The fate of administered bicarbonate is conventionally described in terms of the apparent space of distribution for this ion. Because a sizeable portion of administered bicarbonate is dissipated by body buffers, one can envision the apparent space of distribution as having anatomical and nonanatomical subdivisions; the anatomical portion corresponds to the extracellular volume in which bicarbonate is freely dissolved, and the nonanatomical, to a purely theoretical "space" large enough to accomodate (at the prevailing extracellular bicarbonate concentration) all of the administered bicarbonate unaccounted for by extracellular stores. Numerous observations have established that the anatomic and nonanatomic portions of the apparent bicarbonate space are approximately equal in size in

normal individuals, i.e., that the apparent bicarbonate space is ~40–50% of body weight as compared with an extracellular fluid volume of ~20% of body weight (1–6).

Bicarbonate is often administered to patients with metabolic acidosis and such use has prompted investigators to examine the effect of acidosis on the apparent space of distribution for this ion. Although earlier workers had concluded that metabolic acidosis has a negligible effect on the internal distribution of administered bicarbonate (4, 7), more recent studies demonstrated a significant enlargement of the bicarbonate space under these circumstances (5, 6). This latter finding was ascribed to the associated acidemia and to the consequent availability of a large quantity of hydrogen ion on nonbicarbonate (largely intracellular) buffers that accumulated during the development of acidosis. An alternative possibility, which seemed equally plausible to us, is that a low level of plasma bicarbonate concentration, per se, mandates an enlargement of the bicarbonate space by enlarging the range of pH over which nonbicarbonate buffers will be titrated by a given quantity of administered base.

This study was designed to distinguish between these alternatives. The apparent bicarbonate space was examined in animals with widely different levels of plasma pH but similar values for plasma bicarbonate concentration. The results indicate that the enlargement of bicarbonate space in animals with metabolic acidosis is a consequence not of acidemia but of the low plasma bicarbonate concentration.

## METHODS

53 acute bicarbonate infusion studies were carried out on 36 female mongrel dogs ranging in weight between 10.2 and 18.2 kg. The animals were fed 30 g/kg per d of a synthetic diet until the day of infusion. The diet contained <1.0 meq sodium/100 g, <0.1 meq potassium/100 g, and <0.5 meq chloride/100 g (8). The daily diet was supplemented with 2.5 meq/kg body wt of potassium as neutral phosphate and 2.5 mmol/kg body wt of sodium chloride, except as noted below. The diet was homogenized with twice its weight of distilled water before feeding. Animals that did not eat spontaneously were tube fed; animals that vomited were excluded from further study. Blood samples were obtained by percutaneous arterial puncture; rectal temperature was measured at the time of blood sampling.

Five groups of animals were prepared before the acute infusion with the intent of achieving a broad range of steady-state plasma bicarbonate concentrations in association with acidemia as well as with alkalemia. Accordingly, animals with each of the four cardinal acid-base disturbances and animals with normal acid-base status were studied.[1]

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[1] In some instances, a given animal received two bicarbonate infusions, one at each of two levels of severity of the same acid-base disturbance.

### Group 1. Chronic metabolic acidosis

Metabolic acidosis (nine dogs, 15 studies) of graded severity was induced by adding hydrochloric acid or L-lysine-monohydrochloride (Sigma Chemical Co., St. Louis, MO) to the daily diet. For a given study, the same daily dose of acid was used throughout but, to achieve a wide range of steady-state bicarbonate decrements for the group as a whole, the daily dose of acid ranged between 2 and 8 mmol/kg body wt. The development of a chronic steady state was insured by feeding the acid for at least 7 d (9).

### Group 2. Chronic respiratory alkalosis

Sustained hyperventilation (seven dogs, eight studies) was produced by exposing the animals to an hypoxic atmosphere within a large environmental chamber (10). The ambient oxygen concentration within the chamber was lowered from 21 to 9% over a period of 2–3 d. The chamber atmosphere was maintained at a level of 9% oxygen for at least 7 d, a period known to be adequate for the development of a chronic steady state (11).

### Group 3. Chronic respiratory acidosis

Respiratory acidosis (seven dogs, nine studies) was produced by exposing the animals to carbon dioxide within a large environmental chamber (10). The animals were maintained at a given level of inspired carbon dioxide (8 and/or 11%) for at least 7 d, a period known to be adequate for the development of a chronic steady state (8). A normal atmospheric oxygen concentration of 21% was maintained in all studies.

### Group 4. Chronic metabolic alkalosis

Metabolic alkalosis (13 dogs, 17 studies) of graded severity was produced by administering ethacrynic acid, 50 mg orally, for 1–5 d. To obviate correction of the diuretic-induced alkalosis, sodium neutral-phosphate was substituted for the dietary sodium chloride supplement used in the other groups. 5 d were allowed to lapse after the last dose of ethacrynic acid before evidence for a chronic steady state of acid-base equilibrium was sought.

### Group 5. Normal acid-base equilibrium

Four dogs were used in the four studies performed. Four of the animals used in group 4 were also studied under normal conditions several days before the induction of metabolic alkalosis.

### Acute experimental protocol

Each animal received an acute sodium bicarbonate infusion following the establishment of a chronic steady state of acid-base equilibrium. In each instance, a chronic steady state was judged to have been present when neither $PaCO_2$ nor plasma bicarbonate concentration varied by > ±5% from the mean value for at least three consecutive days before the acute infusion.

On the day of the acute experiment, each animal was weighed and allowed to lie comfortably on a table; the diet was witheld. An indwelling bladder catheter was placed

under topical anesthesia (xylocaine jelly, Astra Company, Worcester, MA). Body weight was corrected for residual urine volume. 10–15 ml of an aqueous solution of sodium radiosulfate (^{35}S, New England Nuclear, Boston, MA) was injected into a peripheral vein; the dose of ^{35}S was calculated to deliver $\sim$0.25 $\mu$Ci/kg body wt. Arterial blood samples were obtained at 30, 60, 90, and 120 min after injection for measurement of plasma sulfate radioactivity. The 90- and 120-min samples were also utilized for base-line measurements of electrolyte and acid-base composition.

Animals were accepted for study only if (a) the mean of the two control values for plasma bicarbonate concentration differed by no >2 meq/liter from the value obtained on the previous day and (b) the mean of the two control observations for $PaCO_2$ differed by no >3 mmHg or 10% (whichever was greater) from the value obtained on the previous day.

Sodium bicarbonate, 5 mmol/kg body wt, was administered intravenously as a 1-N solution over a 10-min period. Arterial blood samples (10 ml) were drawn anaerobically at 30, 60, and 90 min from the midpoint of the infusion. Urine was collected anaerobically in four consecutive periods, one before bicarbonate infusion and three at 30-min intervals following bicarbonate infusion; complete bladder emptying was achieved by manual compression over the suprapubic area.

## Analytical methods

Methods used for determining sodium, potassium, and chloride have been reported previously (12). Total $CO_2$ was measured by autoanalyzer (Technicon Instruments Corp., Tarrytown, NY) and the results were confirmed daily by the manometric technique of Peters and Van Slyke according to a protocol previously described (13). pH was measured anaerobically at 39°C by glass electrode (13). Bicarbonate concentration and $PaCO_2$ were calculated from the Henderson-Hasselbalch equation. pH, pK', and the solubility coefficient of $CO_2$ were corrected for temperature; pK' was also corrected for pH (14–16). Total plasma protein concentration was determined by refractometry (17). [35]S beta-radioactivity was measured in a liquid scintillation solution (ACS, Amersham Corp., Arlington Heights, IL) using a Tri-Carb scintillation spectrometer (model 3002, Packard Instrument Co., Downers Grove, IL).

## Calculations and definitions

The concentration of plasma water (in kilograms per liter) was calculated from the plasma total protein concentration (in grams per deciliter) by the following equation: $[H_2O]_p = 0.986 - 0.00745\ [Pr]_p$ (18). The extracellular concentrations of anions and cations were calculated from their concentrations in plasma water using a correction factor of 0.95 for the Donnan effect. Initial total erythrocyte volume was calculated from the hematocrit and an assumed plasma volume of 0.045 liter/kg body wt. Subsequent values for total erythrocyte volume were corrected for the quantity of blood removed during the experiment.

Initial extracellular fluid (iECF) volume was estimated from the radiosulfate space (19). The counts per liter of plasma water (corrected for Donnan effect) were divided by injected counts for each of the four blood samples obtained before bicarbonate infusion. Radiosulfate space was calculated from the zero-time intercept of the least squares line drawn through the logarithms of these quotients (19).

Changes in extracellular fluid (ECF)[2] volume ($\Delta$ECF) following bicarbonate infusion were estimated using chloride space arithmetic, as follows:

$$\Delta ECF = eECF - iECF,$$

$$eECF = (iECF \times i[Cl^-]_e + b_{Cl})/e[Cl^-]_e,$$

where eECF = experimental ECF volume, iECF = initial ECF volume (radiosulfate space), $i[Cl^-]_e$ = initial extracellular chloride concentration, $b_{Cl}$ = chloride balance, and $e[Cl^-]_e$ = experimental extracellular chloride concentration (18). Because no chloride was administered, the term $b_{Cl}$ represents urinary chloride losses over the respective time interval. Chloride space arithmetic assumes that internal shifts of chloride do not occur.

## Correcting $[HCO_3^-]_p$ and pH for the effects of acute changes in $PaCO_2$

Bicarbonate loading tends to elevate $PaCO_2$ for two reasons: (a) carbon dioxide is generated as some of the administered bicarbonate is titrated and (b) ventilation is dampened by the alkalinizing effects of the administered base. Although the resulting change in $PaCO_2$ is small (Results), to whatever extent hypercapnia does accompany bicarbonate loading, some portion of the measured increment in $[HCO_3^-]_p$ will reflect the titration of nonbicarbonate buffers by carbonic acid (21). This unwanted influence on $[HCO_3^-]_p$ could, in theory, have been eliminated by controlling ventilation, but anesthesia would have been required to do so. To avoid anesthesia, we chose instead to calculate the small effect of the observed change in $PaCO_2$ and to correct the measured value for $[HCO_3^-]_p$ accordingly. Estimates of the influence of acute increments in $PaCO_2$ on $[HCO_3^-]_p$ were derived from experiments in which acute hypercapnia was produced in unanesthetized dogs with chronic steady states of acid-base equilibrium identical to those studied here (22, 23). A value of 0.18 meq/liter per mmHg increment in $PaCO_2$ was used for normal animals, animals with metabolic acidosis, and animals with metabolic alkalosis; a value of 0.26 meq/liter per mmHg increment in $PaCO_2$ was used for animals with respiratory alkalosis.[3] In no instance did correcting $[HCO_3^-]_p$ for the effects of $PaCO_2$ require a downward adjustment by more than 1.3 meq/liter.

For purposes of data analysis, it was necessary to have values for blood pH following bicarbonate infusion that were uninfluenced either by changes in $PaCO_2$ per se or by that portion of the $\Delta[HCO_3^-]_p$ related to changes in $PaCO_2$. Accordingly, postinfusion values for pH ($pH^c$) were calculated (using the Henderson-Hasselbalch equation) from the respective control value for $PaCO_2$ and the corrected value for plasma bicarbonate ($[HCO_3^-]_p^c$), as defined above.

## Fate of retained bicarbonate

The amount of bicarbonate retained ("retained bicarbonate") at any time following the acute infusion was calculated

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as the difference between the amount administered and the accumulated change in urinary bicarbonate excretion. Having an independent measure of the extracellular space as well as measurements of protein concentration and erythrocyte mass permitted an explicit accounting of much of the retained bicarbonate. Two major divisions and several subdivisions of retained bicarbonate were defined as follows:

*Bicarbonate freely dissolved in the ECF.* (a) Bicarbonate retained within the original volume of extracellular fluid (iECF) was calculated as the product of iECF volume and the $\Delta[HCO_3^-]_e^c$.

(b) Bicarbonate retained within the increment in extracellular volume produced by the acute infusion was calculated as the product of $\Delta$ECF volume and the prevailing $[HCO_3^-]_e^c$.

*Bicarbonate not freely dissolved in the ECF.* (a) Bicarbonate titrated by protons released from plasma proteins was estimated as 0.11 times the product of $\Delta$ plasma $pH^c$, plasma total protein concentration and plasma volume (assumed to be constant at 4.5% body wt). The factor, 0.11, is the buffer value of plasma proteins expressed as millimoles per gram per pH unit (24, 25).

(b) Bicarbonate retained in erythrocytes was calculated as 0.5 times the product of $\Delta[HCO_3^-]_e^c$ and total erythrocyte volume. The factor 0.5 was taken as the ratio of bicarbonate concentration in erythrocytes to that in plasma (3, 25).

(c) Bicarbonate titrated by protons released from hemoglobin was estimated as 60 times the product of $\Delta$ plasma $pH^c$ and total erythrocyte volume. The factor, 60, is the product of the molar buffer value of hemoglobin (which is taken to be 3) and millimoles of hemoglobin per liter erythrocytes (which is assumed to be 20) (26).

(d) Bicarbonate entering the "intracellular compartment" was estimated as the difference between total retained bicarbonate and the sum of the bicarbonate accounted for explicitly by the above calculations. Consequently, this subdivision of retained bicarbonate corresponds to the aggregate contribution of all non-extracellular repositories including skeletal muscle, visceral organs, other soft tissues, and bone (3, 27).

The theoretical fluid volume through which retained bicarbonate appeared to distribute ("apparent bicarbonate space") was calculated by dividing retained bicarbonate by the change in extracellular bicarbonate concentration ($\Delta[HCO_3^-]_e^c$) and was expressed as a percentage of body weight. This conventional definition of bicarbonate space was modified only to the extent that $\Delta[HCO_3^-]_e$ was corrected for the simultaneous $\Delta PaCO_2$, as discussed above.

## Statistical analysis

Statistical analyses were carried out by means of analysis of variance for paired or unpaired groups of data, as appropriate. Regression functions were calculated according to conventional techniques. The terms "significant" or "significantly different" will be used, unless otherwise specified, to describe differences which have a P value of <0.01.

## RESULTS

*General remarks.* The term "metabolic studies" is used in connection with pooled data from animals with metabolic acidosis, metabolic alkalosis, and normal acid-base status; the term "respiratory studies" is used in connection with pooled data from animals with re-

spiratory acidosis, respiratory alkalosis, and normal acid-base status. In some instances, data from all five groups are pooled for analysis.

Throughout the acute experimental protocol, all animals remained quiet, alert, and easy to manage. Administration of radiosulfate and sodium bicarbonate produced no apparent ill effects.

*ECF volume and composition.* As can be seen in Table I, mean ECF volume during control ranged from 23.0 to 26.4% of body wt among the five experimental groups. In no group did control ECF volume differ significantly from that of any other; however, by design, the groups with chronic acid-base disturbances featured graded degrees of severity. For this reason, we combined in turn the data from normals with that from each of the other groups and examined the least squares relationship between initial ECF volume and initial extracellular bicarbonate concentration. This analysis revealed a significant inverse relationship for the metabolic alkalosis group ($y = -0.18x + 29.82, r = -0.447, P < 0.05$), in keeping with the progressively more severe volume depletion anticipated in this group as a function of the severity of the diuretic-induced hyperbicarbonatemia. Analysis also revealed a significant direct relationship for the metabolic acidosis group ($y = 0.17x + 22.79, r = 0.469, P < 0.05$), in accordance with the progressively greater cationic losses characteristic of mineral acid-induced hypobicarbonatemia of graded severity (9). Under the conditions of these studies therefore, the mean iECF volume expected for animals with $i[HCO_3^-]_e$ of 10, 21, or 50 meq/liter of metabolic origin was 24.5, 26.2, or 20.8% of body wt, respectively. No significant relationship between iECF volume and $i[HCO_3^-]_e$ was detected for either the respiratory acidosis or the respiratory alkalosis group.

Administration of sodium bicarbonate resulted in expansion of ECF volume in all experimental groups; the degree of expansion was not significantly different among the groups and ranged from 60 to 129 ml at 30 min. Thereafter, a progressive return of ECF volume towards control was observed; in each group, the degree of expansion at 90 min was significantly less than that at 30 min. Control hematocrit was significantly elevated in the group with chronic respiratory alkalosis, in keeping with the hypoxia-induced hyperventilation imposed to produce this acid-base disturbance. Hematocrit values, as well as plasma total protein concentration were significantly lower in all groups at 30 min after bicarbonate infusion; subsequently, they tended to return towards control values.

Control plasma bicarbonate concentrations ($[HCO_3^-]_p$), by design, covered a wide spectrum ranging from 8.5 to 16.5 meq/liter in animals with chronic metabolic acidosis, from 11.9 to 15.2 meq/liter in an-

## TABLE I
Changes in ECF Volume and Composition following Administration of Sodium Bicarbonate to Dogs with Various States of Chronic Acid-Base Equilibrium

| | Normal (n = 4) | | | | Chronic metabolic acidosis (n = 15) | | | | Chronic metabolic alkalosis (n = 17) | | | | Chronic respiratory alkalosis (n = 8) | | | | Chronic respiratory acidosis (n = 9) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min |
| ECF volume (% body wt) | 26.4 ±1.1 | | | | 25.0 ±0.3 | | | | 23.0 ±0.8 | | | | 24.0 ±1.1 | | | | 24.2 ±1.4 | | | |
| ΔECF volume (ml) | | 86 ±33 | 37 ±22 | 42 ±32 | | 129 ±19 | 91 ±15 | 53 ±10 | | 115 ±17 | 39 ±11 | 23 ±10 | | 63 ±12 | 15 ±23 | 1 ±21 | | 60 ±8 | 37 ±10 | −5 ±16 |
| [Protein]$_p$ (g/100 ml) | 6.3 ±0.2 | 5.7 ±0.2 | 5.8 ±0.2 | 5.9 ±0.2 | 6.6 ±0.2 | 5.9 ±0.2 | 6.0 ±0.2 | 6.1 ±0.2 | 7.2 ±0.2 | 6.5 ±0.2 | 6.8 ±0.2 | 6.8 ±0.2 | 7.3 ±0.2 | 6.5 ±0.2 | 6.6 ±0.2 | 6.6 ±0.2 | 6.6 ±0.2 | 6.0 ±0.2 | 6.2 ±0.2 | 6.3 ±0.2 |
| Hct (%) | 39.7 ±1.7 | 34.9 ±0.9 | 35.3 ±1.4 | 35.9 ±1.6 | 42.2 ±1.4 | 37.2 ±1.5 | 37.8 ±1.4 | 38.6 ±1.4 | 44.1 ±0.9 | 40.1 ±0.9 | 41.5 ±1.0 | 41.3 ±1.0 | 57.2 ±2.1 | 53.4 ±1.9 | 54.1 ±2.1 | 54.1 ±1.9 | 43.4 ±1.4 | 39.2 ±1.6 | 41.2 ±1.8 | 41.7 ±1.6 |
| Weight (kg) | 12.9 ±0.8 | | | | 14.6 ±0.4 | | | | 13.5 ±0.6 | | | | 12.5 ±0.5 | | | | 11.9 ±0.4 | | | |

Values represent means±1 SE.

imals with chronic respiratory alkalosis, from 28.2 to 36.4 meq/liter in animals with chronic respiratory acidosis, from 23.0 to 42.3 meq/liter in animals with chronic metabolic alkalosis. Control plasma bicarbonate concentrations in animals with normal acid-base equilibrium ranged from 18.9 to 20.4 meq/liter.

As shown in Table II, extracellular bicarbonate concentration ($[HCO_3^-]_e^c$) increased significantly in all groups after bicarbonate infusion. The change in $[HCO_3^-]_e^c$, however, was not significantly different among groups at any point of observation; mean values for $\Delta[HCO_3^-]_e^c$ varied from +9.1 to +10.5 meq/liter at 30 min. Progressively smaller $\Delta[HCO_3^-]_e^c$ values were observed over the ensuing hour in all groups. Bicarbonate infusion resulted in a small but significant increment in $PaCO_2$ in all but the respiratory acidosis group. $PaCO_2$ did not change significantly in any group over the final hour of observation.

Plasma $pH^c$, as did $[HCO_3^-]_e^c$, increased significantly in all experimental groups following bicarbonate infusion.[4] Values were maximally elevated at 30 min and returned progressively towards control thereafter. As can be seen in Fig. 1, a significant inverse relationship was found between $[HCO_3^-]_e$ and $\Delta$ plasma $pH^c$ at each point of observation in both metabolic and respiratory studies. When the 30-min data from all 53 studies were pooled, the hyperbolic function shown in the figure provided an excellent fit, as assessed by covariance analysis. At 60 and 90 min, relationships with very similar slopes but lower intercepts were found (Fig. 1, right hand panel).

As shown in Table II, extracellular sodium concentration in each group was significantly higher than control at 30 min following sodium bicarbonate administration and remained elevated to the same degree in subsequent periods. Extracellular potassium concentration, by contrast, fell significantly in all groups by 30 min but, as with sodium concentration, remained unchanged throughout the remainder of the study. Extracellular chloride concentration was significantly lower in all groups at 30 min after bicarbonate infusion and, in general, tended to return towards control over the ensuing hour.

Plasma unmeasured anion concentration, defined as

the sum of plasma sodium and potassium concentrations minus the sum of plasma chloride and bicarbonate concentrations, was not significantly altered by bicarbonate infusion in any of the experimental groups. Furthermore, the small reduction in plasma protein concentration coupled with the rise in plasma pH that were observed after bicarbonate infusion left the anionic equivalency of plasma proteins virtually unchanged (28, 29). This observation, taken together with the stability of the unmeasured anion concentration, serves to exclude a sizeable accumulation of organic acids following infusion of sodium bicarbonate.

*Bicarbonate excretion.* As can be seen in Table III, bicarbonate excretion was virtually nil during control in all experimental groups and increased to a variable extent following bicarbonate infusion. Animals with metabolic acidosis excreted the least amount of administered bicarbonate, whereas animals with respiratory acidosis excreted the most; cumulative bicarbonate excretion at 90 min averaged 0.5% of the administered load in the metabolic acidosis group and 43% of the administered load in the respiratory acidosis group. The corresponding values for the metabolic alkalosis and respiratory alkalosis groups were 35 and 20%, respectively.

*Fate of retained bicarbonate.* Table IV depicts the mean values for retained bicarbonate in the five experimental groups at each point of observation. With the exception of the metabolic acidosis group, in which bicarbonate excretion was negligible, the amount of bicarbonate retained declined progressively as a function of time but, in keeping with the differences in alkali excretion rates noted above, did so more rapidly the higher the initial level of extracellular bicarbonate concentration.

Using measurements of ECF volume, total plasma protein concentration and hematocrit, it was possible to obtain a somewhat more explicit accounting of the internal distribution of retained bicarbonate than that provided by the theoretic "apparent bicarbonate space." As indicated in the Methods section, retained bicarbonate can be thought of as existing in two major divisions: (*a*) that freely dissolved in the ECF, including both the initial and the expanded portions thereof and (*b*) that not freely dissolved in the ECF, encompassing the bicarbonate titrated by plasma proteins, dissolved in erythrocyte water, titrated by hemoglobin, and removed by the "intracellular" compartment (i.e., soft tissue and bone buffers). The distribution of retained bicarbonate among these various subdivisions is depicted in Table IV for each group at each point of observation.

Marked differences were observed among the animals with respect to the internal distribution of retained bicarbonate. A consistent and striking feature,

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[4] Values for pH during experimental periods were calculated from the corrected plasma $[HCO_3^-]$ and the preinfusion $PaCO_2$ (Methods). This calculated correction permitted an assessment of the effects of bicarbonate infusion, per se (i.e., without the complicating influence of unavoidable changes in $PaCO_2$). In each case, calculated pH values were slightly higher than the corresponding measured values, reflecting the fact that the observed $\Delta PaCO_2$ was proportionately greater than the required correction in plasma $\Delta[HCO_3^-]$. The differences between measured and calculated pH values ranged between 0.03 and 0.06 pH units.

## TABLE II

### Changes in Acid-Base Parameters and Extracellular Electrolytes following Administration of Sodium Bicarbonate to Dogs with Various States of Chronic Acid-Base Equilibrium*

| | Normal (n = 4) | | | | Chronic metabolic acidosis (n = 15) | | | | Chronic metabolic alkalosis (n = 17) | | | | Chronic respiratory alkalosis (n = 8) | | | | Chronic respiratory acidosis (n = 9) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min | Control | 30 min | 60 min | 90 min |
| $PaCO_2$ (mmHg) | 35 ±0.9 | 40 ±0.8 | 40 ±0.5 | 40 ±0.9 | 31 ±0.6 | 37 ±0.5 | 38 ±0.5 | 37 ±0.6 | 45 ±1.3 | 50 ±1.6 | 50 ±1.5 | 49 ±1.6 | 22 ±0.5 | 25 ±0.6 | 25 ±0.6 | 25 ±0.6 | 70 ±2.8 | 71 ±2.5 | 70 ±2.7 | 70 ±3.1 |
| $[HCO_3]$‡ (meq/liter) | 22.1 ±0.38 | 32.2 ±0.62 | 30.1 ±0.33 | 29.3 ±0.53 | 13.5 ±0.64 | 24.0 ±0.90 | 23.1 ±0.87 | 22.0 ±0.91 | 37.1 ±1.66 | 47.5 ±1.71 | 45.3 ±1.72 | 44.0 ±1.63 | 14.8 ±0.46 | 24.6 ±0.66 | 23.3 ±0.65 | 22.5 ±0.76 | 35.8 ±1.10 | 44.9 ±1.07 | 42.5 ±1.20 | 41.4 ±1.29 |
| pH§ | 7.37 ±0.01 | 7.52 ±0.01 | 7.49 ±0.01 | 7.48 ±0.01 | 7.20 ±0.02 | 7.43 ±0.01 | 7.41 ±0.01 | 7.39 ±0.01 | 7.47 ±0.01 | 7.57 ±0.01 | 7.55 ±0.01 | 7.54 ±0.01 | 7.40 ±0.01 | 7.60 ±0.01 | 7.57 ±0.01 | 7.55 ±0.01 | 7.28 ±0.01 | 7.37 ±0.01 | 7.35 ±0.01 | 7.34 ±0.01 |
| $[Na]_e$ (meq/liter) | 142 ±1.1 | 148 ±1.0 | 147 ±1.1 | 147 ±1.0 | 144 ±0.5 | 151 ±0.7 | 150 ±0.7 | 150 ±0.6 | 141 ±1.0 | 148 ±0.9 | 147 ±1.0 | 147 ±1.0 | 150 ±0.9 | 156 ±1.3 | 156 ±1.2 | 156 ±1.1 | 147 ±1.2 | 153 ±1.0 | 153 ±1.1 | 153 ±1.1 |
| $[K]_e$ (meq/liter) | 3.6 ±0.2 | 3.1 ±0.1 | 3.1 ±0.1 | 3.1 ±0.1 | 2.6 ±0.1 | 2.2 ±0.1 | 2.2 ±0.1 | 2.2 ±0.1 | 2.3 ±0.1 | 2.1 ±0.1 | 2.1 ±0.1 | 2.1 ±0.1 | 4.0 ±0.1 | 3.4 ±0.1 | 3.4 ±0.1 | 3.4 ±0.1 | 3.4 ±0.1 | 2.7 ±0.1 | 2.7 ±0.1 | 2.7 ±0.1 |
| $[Cl]_e$ (meq/liter) | 126 ±0.9 | 121 ±0.7 | 123 ±0.4 | 122 ±0.7 | 134 ±0.9 | 129 ±1.1 | 130 ±1.0 | 131 ±0.9 | 102 ±2.6 | 98 ±2.7 | 100 ±2.5 | 101 ±2.4 | 130 ±1.1 | 126 ±1.0 | 127 ±0.9 | 128 ±0.7 | 117 ±1.7 | 114 ±1.6 | 115 ±1.8 | 116 ±1.4 |
| Unmeasured anions‖ (meq/liter) | 12 ±0.6 | 12 ±1.1 | 12 ±1.0 | 13 ±1.3 | 13 ±0.6 | 14 ±0.7 | 14 ±0.7 | 14 ±0.4 | 18 ±0.7 | 18 ±1.0 | 18 ±0.8 | 18 ±0.8 | 23 ±1.1 | 24 ±1.3 | 23 ±1.4 | 23 ±1.1 | 13 ±0.7 | 13 ±0.7 | 14 ±0.9 | 13 ±1.1 |

* Values presented are the means ± 1 SE.
‡ All experimental values except those from the chronic respiratory acidosis group have been corrected for the observed increment in $PaCO_2$ following bicarbonate infusion (Methods).
§ All experimental values except those from the chronic respiratory acidosis group have been calculated from the corrected $[HCO_3]_p$ and the preinfusion $PaCO_2$ (Methods).
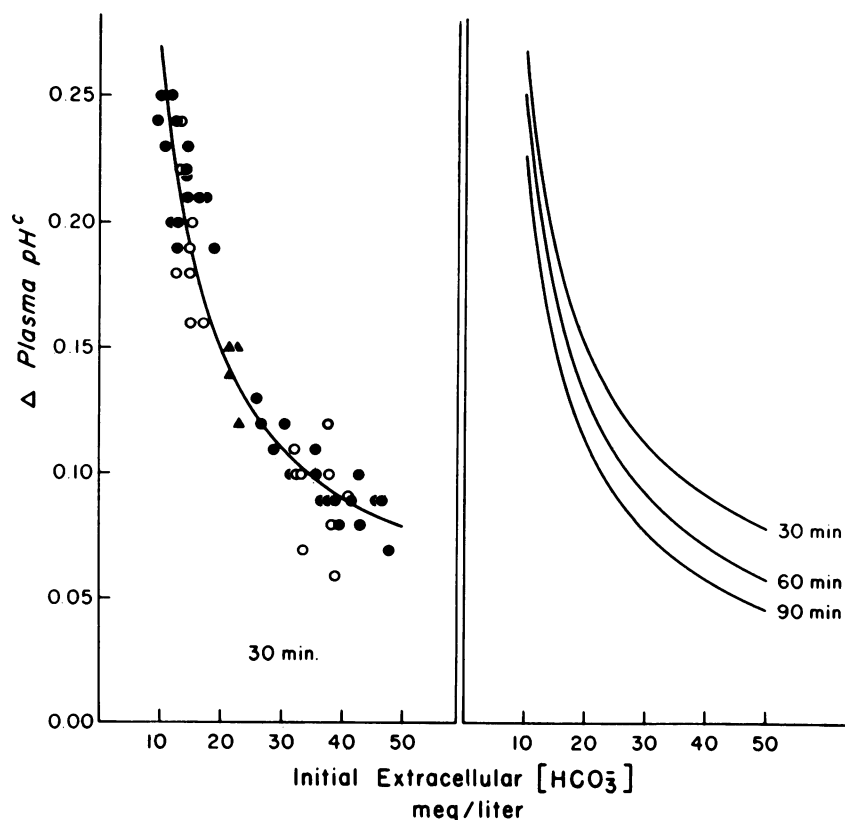‖ $(Na + K)_p - (HCO_3 + Cl)_p$.

Figure 1 Relationship between initial extracellular bicarbonate concentration and change in plasma pH following sodium bicarbonate infusion. Experimental values for plasma pH were calculated from the corrected plasma $[HCO_3^-]$ and the preinfusion $PaCO_2$ (Methods). The left-hand panel depicts the relationship at 30 min after infusion. Filled circles denote values obtained in dogs with metabolic acidosis and metabolic alkalosis. Open circles denote values obtained in dogs with respiratory acidosis and respiratory alkalosis. Filled triangles denote values obtained in dogs with normal acid-base status. The pooled data were best described by the hyperbolic function, $y = \dfrac{2.38}{x} + 0.032$ ($r = 0.957$). The right-hand panel compares the relationship at 30 min with those obtained at 60 min $\left( y = \dfrac{2.42}{x} + 0.01, r = 0.953 \right)$ and 90 min $\left( y = \dfrac{2.27}{x} + 0.001, r = 0.922 \right)$ after the infusion. The intercepts, but not the slopes, of these relationships are significantly different from each other. The slope of each relationship is significantly different from zero.

however, was the dynamic nature of the distribution process. Thus, throughout the period of observation, an operational steady state was never achieved with regard either to the apparent space of distribution of bicarbonate or to the compartmental distribution of the retained bicarbonate.

As shown in the left-hand panel of Fig. 2, pooled data from all studies yielded a significant inverse correlation between $i[HCO_3^-]_e$ and the apparent space of distribution for bicarbonate; on the average, animals with the lowest values for $i[HCO_3^-]_e$ (whether of respiratory or metabolic origin) had an ~50% larger space of distribution at 30 min than animals with the highest values. As shown in the right-hand portion of Fig. 2,

the apparent space of distribution increased continuously during the postinfusion period but the strong influence exerted by the $i[HCO_3^-]_e$ persisted throughout. Utilizing the relationships depicted in Fig. 2, one can calculate that animals with an $i[HCO_3^-]_e$ of 10 meq/liter, whether of respiratory or of metabolic origin, will exhibit on average a bicarbonate space of ~60% body wt at 30 min and 76% body wt at 90 min; at the opposite extreme, animals with an $i[HCO_3^-]_e$ of 50 meq/liter will exhibit an average bicarbonate space of ~41 and 53% body wt at 30 and 90 min, respectively.

Because $i[HCO_3^-]_e$ influenced the space of distribution of bicarbonate similarly in the metabolic and in the respiratory studies, it is axiomatic that control plasma

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TABLE III

Changes in Urinary Bicarbonate Excretion following Administration of Sodium Bicarbonate to Dogs
with Various States of Chronic Acid-Base Equilibrium

| | Normal (n = 4) | | | | Chronic metabolic acidosis (n = 15) | | | | Chronic metabolic alkalosis (n = 17) | | | | Chronic respiratory alkalosis (n = 8) | | | | Chronic respiratory acidosis (n = 9) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Con-trol | 30 min | 60 min | 90 min | Con-trol | 30 min | 60 min | 90 min | Con-trol | 30 min | 60 min | 90 min | Con-trol | 30 min | 60 min | 90 min | Con-trol | 30 min | 60 min | 90 min |
| Bicarbonate excretion ($\mu eq/min$) | 0 ±0.3 | 300 ±83.3 | 194 ±38.1 | 118 ±28.4 | 0 ±0 | 10 ±4.7 | 2 ±0.9 | 0 ±0.1 | 6 ±2.3 | 334 ±26.6 | 225 ±16.6 | 154 ±13.9 | 0 ±0 | 215 ±21.6 | 124 ±9.8 | 64 ±9.1 | 4 ±1.2 | 404 ±36.4 | 249 ±22.9 | 136 ±10.4 |
| Cumulative bicarbonate excretion, percentage of administered | | 16.4 ±3.3 | 25.5 ±4.4 | 31.0 ±5.4 | | 0.4 ±0.2 | 0.5 ±0.2 | 0.5 ±0.2 | | 17.8 ±1.1 | 28.0 ±1.5 | 35.0 ±1.9 | | 11.4 ±1.2 | 17.0 ±1.6 | 20.0 ±1.9 | | 23.8 ±1.4 | 36.4 ±2.2 | 43.3 ±2.3 |

Values represent the means±1 SE.

pH was not of overriding importance. Indeed, as indicated in Figs. 3 and 4, a significant negative correlation between the space of distribution of bicarbonate and the initial plasma pH was observed in the metabolic studies and a significant positive correlation was observed in the respiratory studies.

As is evident from the similarity of ECF measurements among the groups and the marked influence of $i[HCO_3^-]_e$ on the apparent space of distribution for bicarbonate, a much greater fraction of retained bicarbonate disappeared from the extracellular fluid in animals with low as compared with high initial extracellular bicarbonate concentrations; as shown in Fig. 5, when pooled data from all five groups were analyzed at each time interval, a significant inverse relationship was observed between $i[HCO_3^-]_e$ and the percentage of retained bicarbonate not found freely dissolved in the ECF. Based on the relationship depicted in Fig. 5, one can calculate that animals with an $i[HCO_3^-]_e$ of 10 meq/liter, whether of respiratory or of metabolic origin, will dissipate approximately two-thirds of retained bicarbonate outside of the ECF space by 30 min; at the opposite extreme, in animals with an $i[HCO_3^-]_e$ of 50 meq/liter, only approximately one-third of the retained bicarbonate will have disappeared from the ECF by 30 min. As is evident from Table IV, most of the bicarbonate not remaining freely dissolved in the ECF volume was removed by the "intracellular compartment." As was the case for the non-ECF bicarbonate as a whole (Fig. 5), "intracellular" buffering contributed more to the dissipation of retained bicarbonate in animals with hypobicarbonatemia (whether acidemic or alkalemic) than it did in those with hyperbicarbonatemia. In animals with an $i[HCO_3^-]_e$ of 10 meq/liter, whether of respiratory or metabolic origin, ~40% of the bicarbonate retained at 30 min would be expected to enter the "intracellular" compartment.

By contrast, only ~25% would be expected to do so in animals with an $i[HCO_3^-]_e$ of 50 meq/liter. Fig. 5 also illustrates that, as time elapses following bicarbonate infusion, a progressively larger fraction of retained bicarbonate disappeared from the ECF.

## DISCUSSION

This study demonstrates that the internal distribution of an acute alkali load is critically dependent on the initial level of plasma bicarbonate concentration, rather than on the prevailing pH. Hypobicarbonatemic animals, whether acidemic or alkalemic, exhibited a much larger apparent space of distribution for administered bicarbonate than did hyperbicarbonatemic animals. The second major finding, scarcely emphasized in previous studies of bicarbonate loading (2, 5, 6), is that the response to acute bicarbonate infusion is a highly dynamic one, characterized at all levels of initial plasma bicarbonate concentration by a continuous and rapid disappearance of retained base from the extracellular compartment. As a consequence, the apparent space of distribution for administered bicarbonate increases progressively for at least an hour or two after infusion and, hence, cannot meaningfully be assigned a single value, even at a given bicarbonate concentration.

Before elaborating on the major conclusions of this study, two important methodologic differences between this and previous investigations of acute bicarbonate loading deserve comment. The first involves the animal preparation used. Previous workers have utilized bilateral nephrectomy or ureteral ligation to obviate the loss of administered bicarbonate (2, 5, 6). We chose to avoid the anesthesia and surgical trauma involved in this approach in the belief that more meaningful data could be obtained from studying intact

# TABLE IV

## Distribution of Retained Bicarbonate following Administration of Sodium Bicarbonate to Dogs with Various States of Acid-Base Equilibrium

| | Normal (n = 4) | | | Chronic metabolic acidosis (n = 15) | | | Chronic metabolic alkalosis (n = 17) | | | Chronic respiratory alkalosis (n = 8) | | | Chronic respiratory acidosis (n = 9) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 30 min | 60 min | 90 min | 30 min | 60 min | 90 min | 30 min | 60 min | 90 min | 30 min | 60 min | 90 min | 30 min | 60 min | 90 min |
| Retained bicarbonate (meq/kg body wt) | 4.06 ±0.15 | 3.62 ±0.20 | 3.35 ±0.25 | 4.92 ±0.02 | 4.92 ±0.02 | 4.92 ±0.02 | 3.98 ±0.06 | 3.49 ±0.08 | 3.15 ±0.09 | 4.48 ±0.07 | 4.20 ±0.09 | 4.05 ±0.10 | 3.78 ±0.08 | 3.15 ±0.12 | 2.81 ±0.12 |
| **Bicarbonate freely dissolved in the ECF (meq/kg body wt)** | | | | | | | | | | | | | | | |
| In initial ECF volume | 2.36 ±0.14 | 1.83 ±0.10 | 1.63 ±0.18 | 2.34 ±0.10 | 2.10 ±0.09 | 1.85 ±0.10 | 2.13 ±0.07 | 1.66 ±0.07 | 1.35 ±0.07 | 2.03 ±0.10 | 1.68 ±0.08 | 1.46 ±0.11 | 2.17 ±0.13 | 1.60 ±0.16 | 1.32 ±0.15 |
| In ECF increment | 0.22 ±0.08 | 0.09 ±0.05 | 0.10 ±0.07 | 0.21 ±0.03 | 0.14 ±0.02 | 0.08 ±0.02 | 0.39 ±0.05 | 0.13 ±0.04 | 0.07 ±0.03 | 0.11 ±0.02 | 0.05 ±0.04 | 0.01 ±0.03 | 0.23 ±0.04 | 0.15 ±0.05 | -0.02 ±0.06 |
| Total | 2.57 ±0.17 | 1.92 ±0.11 | 1.72 ±0.19 | 2.55 ±0.13 | 2.24 ±0.10 | 1.93 ±0.11 | 2.52 ±0.08 | 1.78 ±0.10 | 1.42 ±0.08 | 2.14 ±0.11 | 1.72 ±0.12 | 1.47 ±0.13 | 2.40 ±0.16 | 1.75 ±0.20 | 1.31 ±0.19 |
| **Bicarbonate not freely dissolved in the ECF (meq/kg body wt)** | | | | | | | | | | | | | | | |
| Titrated by plasma proteins | 0.04 ±0.005 | 0.03 ±0.003 | 0.03 ±0.003 | 0.06 ±0.002 | 0.06 ±0.002 | 0.05 ±0.003 | 0.03 ±0.001 | 0.03 ±0.001 | 0.02 ±0.002 | 0.06 ±0.004 | 0.05 ±0.005 | 0.05 ±0.005 | 0.03 ±0.003 | 0.02 ±0.002 | 0.02 ±0.002 |
| Dissolved in erythrocyte water | 0.12 ±0.013 | 0.09 ±0.011 | 0.08 ±0.007 | 0.14 ±0.008 | 0.12 ±0.008 | 0.11 ±0.008 | 0.15 ±0.008 | 0.12 ±0.007 | 0.09 ±0.007 | 0.23 ±0.020 | 0.19 ±0.021 | 0.16 ±0.021 | 0.14 ±0.008 | 0.10 ±0.007 | 0.08 ±0.008 |
| Titrated by hemoglobin | 0.24 ±0.027 | 0.19 ±0.019 | 0.17 ±0.017 | 0.43 ±0.032 | 0.39 ±0.032 | 0.35 ±0.029 | 0.20 ±0.011 | 0.16 ±0.011 | 0.13 ±0.010 | 0.64 ±0.060 | 0.55 ±0.063 | 0.48 ±0.066 | 0.18 ±0.012 | 0.15 ±0.009 | 0.12 ±0.014 |
| Removed by "intracellular compartment" | 1.08 ±0.07 | 1.38 ±0.12 | 1.34 ±0.08 | 1.74 ±0.14 | 2.10 ±0.11 | 2.48 ±0.12 | 1.08 ±0.08 | 1.40 ±0.09 | 1.48 ±0.09 | 1.42 ±0.16 | 1.69 ±0.17 | 1.90 ±0.21 | 1.02 ±0.11 | 1.13 ±0.11 | 1.29 ±0.13 |
| Total | 1.49 ±0.04 | 1.70 ±0.11 | 1.62 ±0.07 | 2.37 ±0.13 | 2.68 ±0.11 | 2.99 ±0.11 | 1.46 ±0.08 | 1.71 ±0.09 | 1.72 ±0.09 | 2.35 ±0.11 | 2.48 ±0.11 | 2.58 ±0.14 | 1.38 ±0.10 | 1.40 ±0.10 | 1.51 ±0.12 |

Values represent the means ± 1 SE.
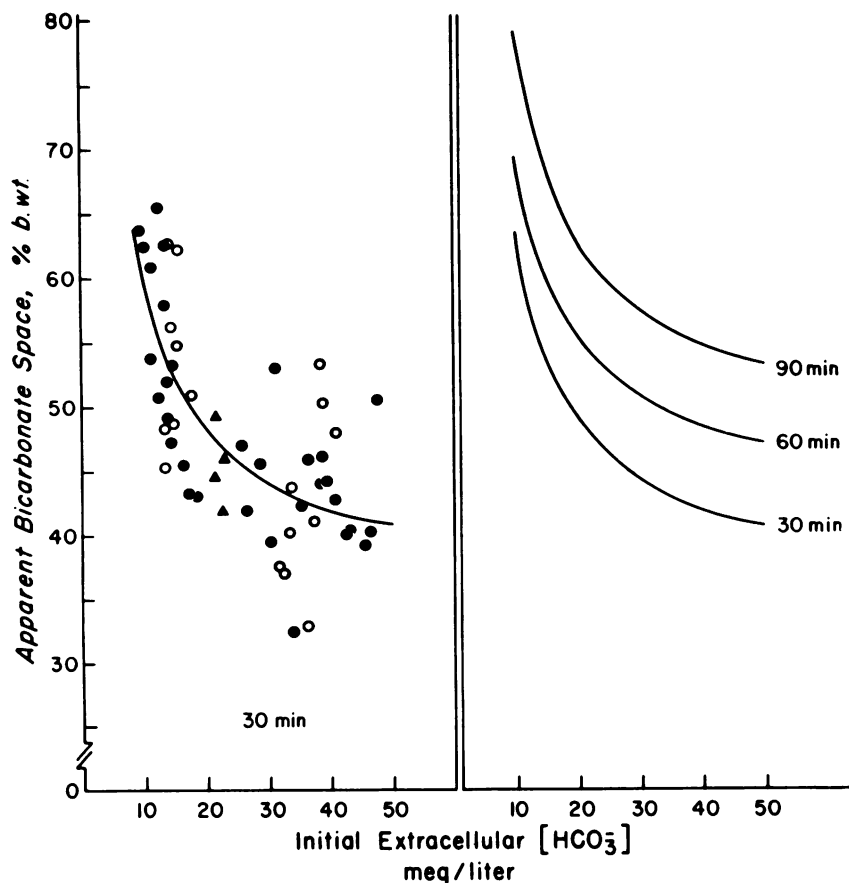
FIGURE 2 Relationship between initial extracellular bicarbonate concentration and apparent bicarbonate space following sodium bicarbonate infusion. The left-hand panel depicts the relationship at 30 min after infusion. Filled circles denote values obtained in dogs with metabolic acid-base disturbances, open circles, values in dogs with respiratory acid-base disturbances, and filled triangles, values in dogs with normal acid-base status. The pooled data are best described

by the hyperbolic function, $y = \dfrac{244.3}{x} + 35.9$ ($r = 0.726$). The right-hand panel compares the

relationship at 30 min with those obtained at 60 min $\left( y = \dfrac{243.5}{x} + 42.4, r = 0.570 \right)$ and 90 min

$\left( y = \dfrac{291.1}{x} + 47.3, r = 0.417 \right)$ after the infusion. The intercepts of these relationships are significantly different from each other. The slope of each relationship is significantly different from zero.

dogs in optimal physiologic condition. The advantages inherent in using an intact animal model in such studies are self-evident. One bit of tangible evidence of this advantage may be found in the apparent stability of plasma organic acid concentration in our animals as compared with the significant increase observed in previous studies (2, 5). The disadvantage to studying animals with intact renal function was that a variable portion of the infused base was excreted during the period of study; as a consequence, the actual amount of retained bicarbonate upon which our estimates of internal distribution are based differed significantly

among the various groups. Considering all five study groups and all three points of observation, the average amount of retained bicarbonate ranged between 2.8 and 4.9 meq/kg body wt (Table IV). Such differences by themselves, however, do not appear to influence the proportion of newly assimilated bicarbonate that undergoes a particular fate in normal animals; previous investigators have found a virtually identical pattern of internal distribution of alkali during the acute administration of as little as 2 to as much as 20 mmol/kg body wt of sodium bicarbonate in intact humans (3) and in animals with interrupted renal func-
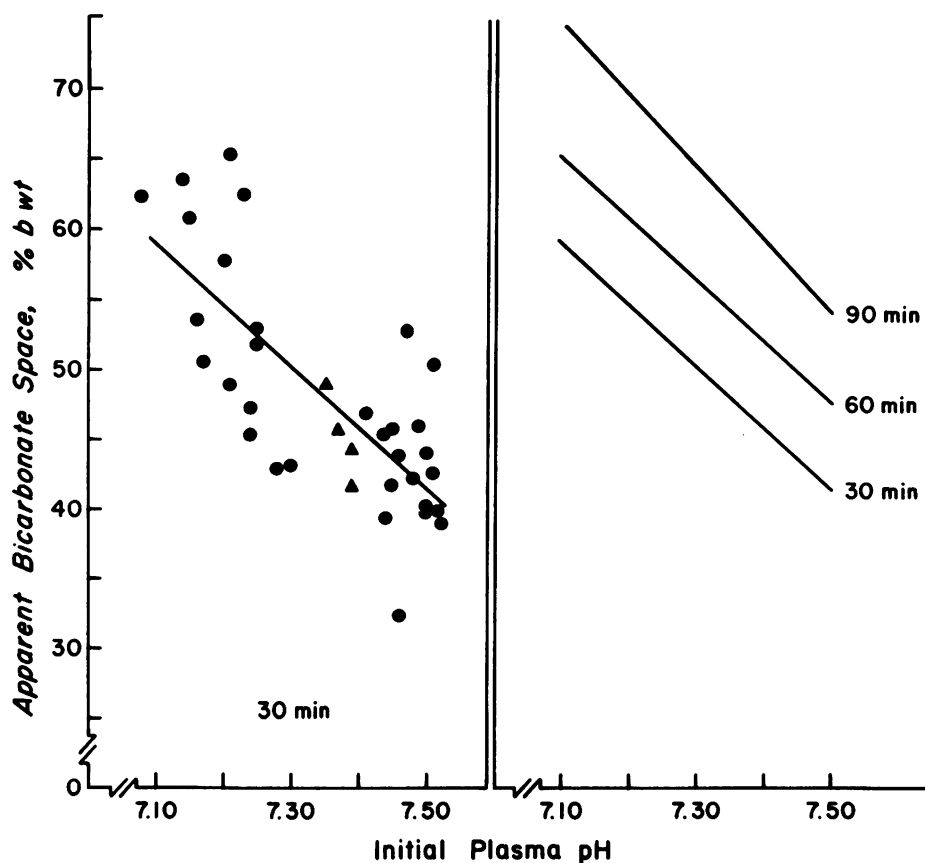
FIGURE 3  Relationship between initial plasma pH and apparent bicarbonate space following sodium bicarbonate infusion in animals with metabolic acidosis and metabolic alkalosis (filled circles) and in animals with normal acid-base status (filled triangles). The left-hand panel depicts the relationship at 30 min after infusion. The least squares regression line drawn through the data conforms to the equation $y = -43.2x + 365.9$ ($r = -0.748$). The right-hand panel compares the relationship at 30 min with those obtained at 60 min ($y = -43.1x + 370.7, r = -0.602$) and 90 min ($y = -50.0x + 429.2, r = -0.409$) after the infusion. The intercepts of these relationships are significantly different from each other. The slope of each relationship is significantly different from zero.

tion (2). In addition to excreting a variable fraction of the administered alkali load, our intact animals also had the potential for augmenting the amount of retained bicarbonate by continuing to excrete urinary net acid. On the one hand, this effect was almost certainly negligible in animals that manifested significant bicarbonaturia (i.e., all but the metabolic acidosis group) because net acid excretion was fully suppressed. On the other hand, to the extent that animals with preinfusion metabolic acidosis continued to excrete net acid following alkali infusion, the apparent space of distribution for administered bicarbonate would be even larger than that indicated by our calculations.

A second major methodologic difference between our studies and previous reports is that we corrected our data for the influence of changes in $PaCO_2$ rather than attempting to obviate the mild hypercapnia that

naturally accompanies alkali infusion. To establish the increment in bicarbonate concentration produced solely by the direct effects of a given alkali load, some means must be used to eliminate the "contaminating" influence of any concomitant alteration in $PaCO_2$; this is true because bicarbonate concentration is unavoidably affected by acute changes in carbonic acid concentration through titration of nonbicarbonate buffers (21). Previous investigators have attempted to prevent changes in $PaCO_2$ through the use of mechanical ventilation, using either general anesthesia or muscle relaxation or both (5, 6). Although theoretically acceptable, this procedure for stabilizing $PaCO_2$ following bicarbonate infusion has not proven to be very reliable in practice; indeed, sizeable increments as well as decrements in $PaCO_2$ have been reported and no consideration has been given to the possible effects of these
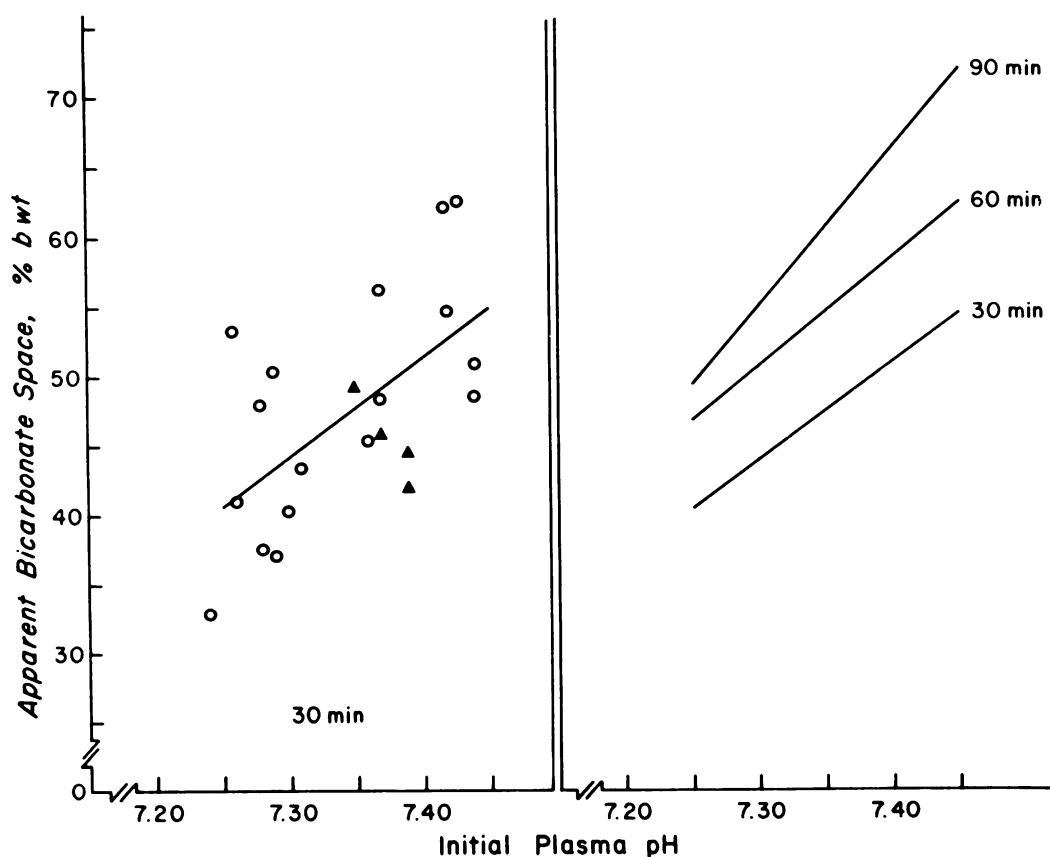
FIGURE 4 Relationship between initial plasma pH and apparent bicarbonate space following sodium bicarbonate infusion in animals with respiratory acidosis and respiratory alkalosis (open circles) and animals with normal acid-base status (filled triangles). The left-hand panel depicts the relationship at 30 min after infusion. The least squares regression line drawn through the data conforms to the equation $y = 71.8x - 480.0$ ($r = 0.608$). The right-hand panel compares the relationship at 30 min with those obtained at 60 min ($y = 79.5x - 529.7$, $r = 0.507$) and 90 min ($y = 113.8x - 775.7$, $r = 0.493$) after the infusion. The intercepts of these relationships are significantly different from each other and the slope of each relationship is significantly different from zero.

changes in $PaCO_2$ on the observed levels of plasma bicarbonate concentration (5, 6). In the current study, we allowed our animals to breathe spontaneously, thus avoiding the uncertain effects of anesthesia as well as the stresses associated with mechanical ventilation; in doing so, a small but significant increase in $PaCO_2$ of 3–6 mmHg was observed following bicarbonate infusion in all but the respiratory acidosis group (Table II). The effects of these changes in $PaCO_2$ on plasma bicarbonate concentration were then discounted during data analysis; to do this, we used correction factors obtained from acute carbon dioxide titration studies carried out over an identical $PaCO_2$ range in animals with experimental acid-base disturbances identical to those used here (22, 23). We believe this approach to eliminating the unwanted effects of increments in $PaCO_2$ is equally sound on theoretic grounds and

avoids the many pitfalls of controlled ventilation. At most, the required correction resulted in a downward adjustment in plasma bicarbonate concentration of 1.3 meq/liter. Although a truly accurate representation of the internal distribution of administered alkali must take proper cognizance of this small effect of secondary hypercapnia, it is noteworthy that an analysis based on the uncorrected data for extracellular bicarbonate yielded identical overall conclusions.

A major finding of this study is that the initial extracellular bicarbonate concentration exerts a striking influence on the apparent space of distribution for administered bicarbonate. What accounts for this effect? One theoretic possibility is that this influence might have reflected sizeable differences in the "anatomic" portion of the bicarbonate space, that is, in the actual volume of the ECF. This possibility is clearly

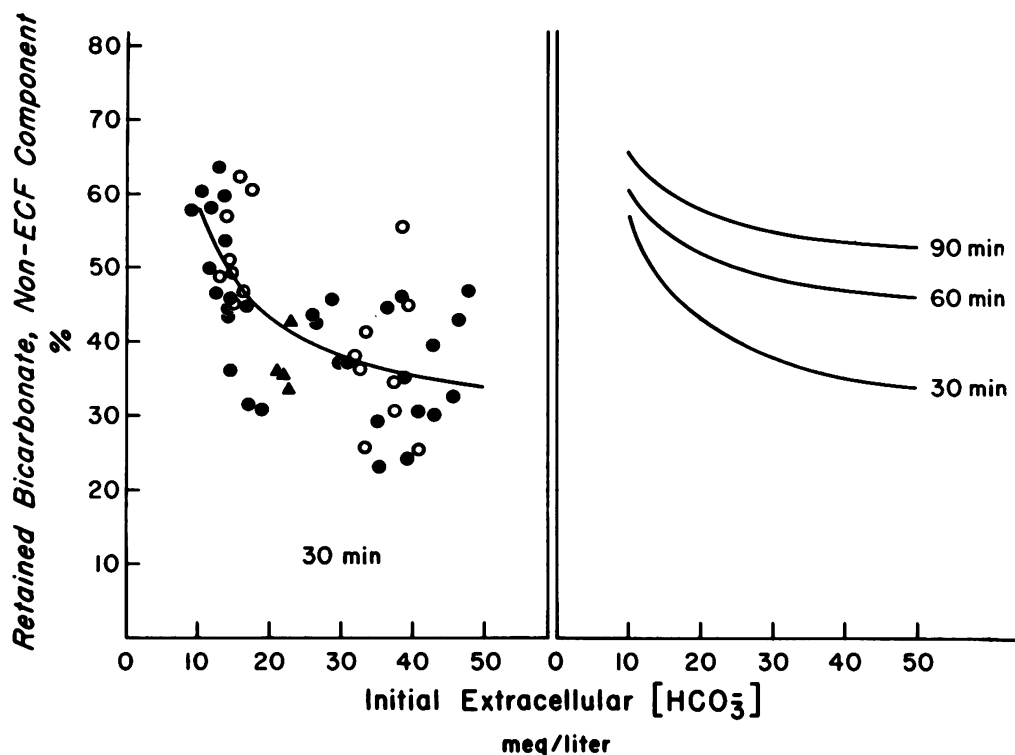*Bicarbonate Space in Chronic Acid-Base Disturbances* 879

FIGURE 5 Relationship between initial extracellular bicarbonate concentration and the fraction of retained bicarbonate that disappeared from the ECF following sodium bicarbonate infusion. The left-hand panel depicts the relationship at 30 min after infusion. Filled circles denote values obtained in dogs with metabolic acidosis and metabolic alkalosis. Open circles denote values obtained in dogs with respiratory acidosis and respiratory alkalosis. Filled triangles denote values obtained in dogs with normal acid-base status. The pooled data were best described by the hyperbolic function, $y = \dfrac{259.5}{x} + 28.1$ ($r = 0.660$). The right-hand panel compares the relationship at 30 min with those obtained at 60 min $\left( y = \dfrac{164.4}{x} + 42.3, r = 0.416 \right)$ and 90 min $\left( y = \dfrac{143.3}{x} + 49.4, r = 0.354 \right)$ after the infusion. The intercepts of these relationships are significantly different from each other. The slope of each relationship is significantly different from zero.

excluded by the present study. Radiosulfate measurements and chloride space arithmetic indicate that hypobicarbonatemic animals had neither a larger initial ECF volume nor a larger increment in extracellular volume following the acute infusion to account for their larger bicarbonate space. Maximal differences in extracellular volume between hypo- and hyperbicarbonatemic animals did not exceed 4% of body wt at any time during the experiments, whereas the concomitant differences in apparent space of distribution for bicarbonate were ~20% of body wt.

Thus, the influence of initial extracellular bicarbonate concentration on the apparent space of distribution of bicarbonate must have been a manifestation largely of variable bicarbonate removal from the extracellular compartment. Why did hypobicarbonatemic animals

transfer a larger fraction of retained bicarbonate out of the extracellular compartment than did normal or hyperbicarbonatemic animals? Previous studies, in which alterations in plasma bicarbonate concentration were induced solely by metabolic acid-base derangements, also detected an enlargement of bicarbonate space in the presence of hypobicarbonatemia (5, 6). The explanation advanced was that the associated acidemia was the critical determinant of the altered distribution of administered base; it was reasoned that animals with metabolic acidosis were poised to dissipate (through intracellular and extracellular buffering) an abnormally large fraction of administered bicarbonate because of the large burden of hydrogen ions sequestered on nonbicarbonate buffers as pH fell during induction of the basal acidosis (5, 6). It is clear

from the present study that this interpretation is not valid. Animals with comparably low levels of extracellular bicarbonate concentration but with alkalemia rather than acidemia (i.e., chronic respiratory alkalosis rather than metabolic acidosis) exhibited comparable enlargements of the apparent bicarbonate space. If anything, such animals would probably experience a reduction in the amount of hydrogen ion sequestered in the buffer pool during their response to prolonged hypocapnia.

The data from this study indicate that the critical factor accounting for the influence of initial plasma bicarbonate concentration on the internal distribution of alkali is the change in pH induced by acute alkali loading, rather than the prevailing pH at the onset of the infusion. In obeying the constraints of the Henderson-Hasselbalch equation, animals with hypobicarbonatemia for whatever reason will experience a larger change in pH in response to a given increment in plasma bicarbonate than will normal or hyperbicarbonatemic animals. In our experiments, the mean increment in plasma bicarbonate concentration pro-

duced by the infusion protocol was remarkably similar in all five groups (Table II). This similarity reflects the fact that animals with smaller bicarbonate spaces (i.e., those in the hyperbicarbonatemic groups) retained a smaller fraction of the administered base during the period of observation than did animals with larger bicarbonate spaces. In any event, the complex set of interacting factors that determined the observed increment in bicarbonate concentration culminated in a striking inverse correlation between the initial level of plasma bicarbonate and the change in pH (Fig. 1); on average, animals with low bicarbonate levels experienced a shift in pH at least 0.10 U greater than those with normal or elevated levels.

Given this inverse correlation and given the likelihood that changes in extracellular pH are accompanied by roughly proportional shifts in intracellular pH (30–32), it seems reasonable to hypothesize that the observed differences in the apparent space of distribution for administered base resulted from the widely different range of pH over which nonbicarbonate buffers were titrated in the various states of acid-base equi-
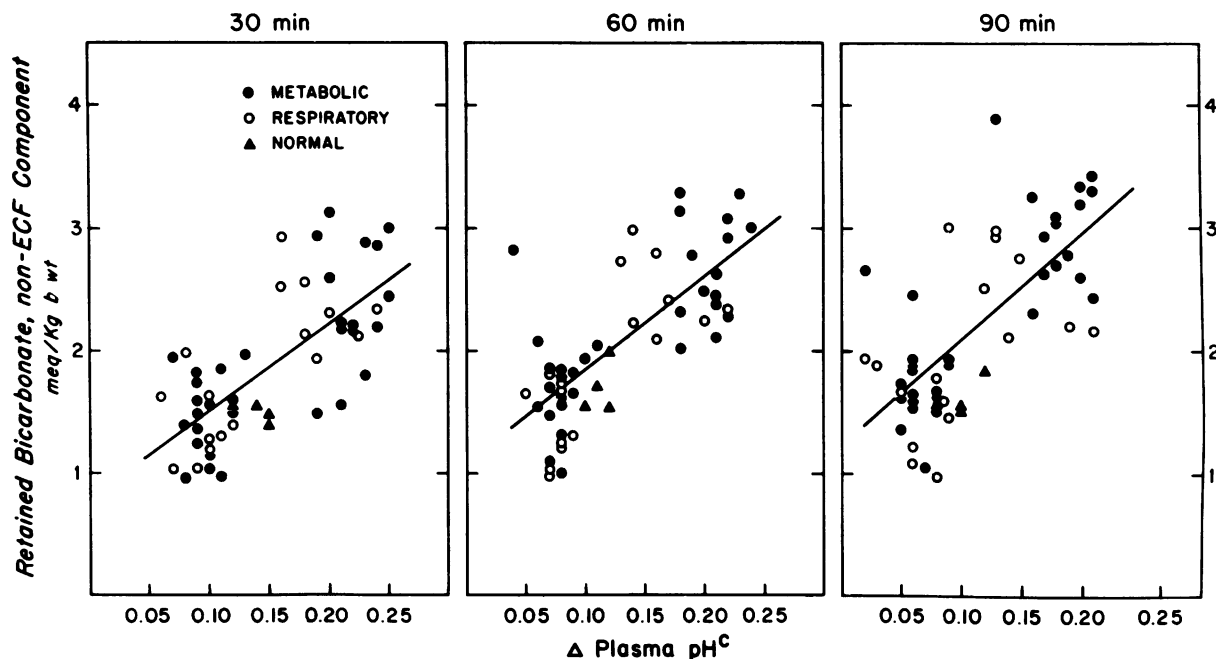


FIGURE 6 Relationship between change in plasma pH and amount of retained bicarbonate that disappeared from the ECF after sodium bicarbonate infusion. Experimental values for plasma pH were calculated from the corrected plasma [$HCO_3$] and the preinfusion $PaCO_2$ (Methods). The left-hand, middle and right-hand panels depict the relationship at 30, 60, and 90 min after infusion, respectively. Filled circles denote values obtained in dogs with metabolic acidosis and metabolic alkalosis. Open circles denote values obtained in dogs with respiratory acidosis and respiratory alkalosis. Filled triangles denote values obtained in dogs with normal acid-base status. The least squares regression lines drawn through the data conform to the equations, $y = 7.16x + 0.78$, $r = 0.719$ (30 min), $y = 7.61x + 1.07$, $r = 0.735$ (60 min) and $y = 8.57x + 1.21$, $r = 0.696$ (90 min). The intercepts of these relationships are significantly different from each other and the slope of each relationship is significantly different from zero.

librium studied.[5] If the aggregate buffer value of non-bicarbonate buffers is constant over the range of pH induced in the present study groups,[6] this hypothesis would predict a linear relationship between the observed change in plasma pH and the quantity of bicarbonate that disappeared from the extracellular compartment in each of the circumstances examined. As shown in Fig. 6, pooled data from all experiments analyzed in this fashion reveals just such a linear correlation for each of the three points of observation.

Fig. 6 also illustrates the second major finding of this study, namely the striking time dependence of the buffering response; over each 30-min interval following infusion, progressively more retained bicarbonate was dissipated by buffering for a given change in pH. The calculations upon which this conclusion is based cannot distinguish between ongoing production of endogenous acid and slowly equilibrating buffer stores (34, 35). However, if one assumes that endogenous acid production continued at rates typically observed for dogs ingesting the diets used in this study (8), <0.2 meq/kg body wt of administered alkali would have been consumed by this process over the entire 90-min period of study.

Slow equilibration of tissue buffers was, of course, manifestly present in animals with metabolic acidosis in which virtually none of the infused bicarbonate was excreted but in which plasma bicarbonate fell progressively from its peak value at 30 min (Table II). For each of the other groups, in which appreciable excretion of administered bicarbonate did occur, close analysis of Fig. 6 reveals that slow equilibration of tissue buffers contributed importantly to the continued dissipation of infused base and, hence, contributed to the postinfusion decline in plasma bicarbonate. This study sheds no light on the precise location(s) at which slow equilibration of nonbicarbonate buffers takes place. Bone is obviously an attractive candidate for such a sluggish response. Indeed previous investigators have demonstrated that bone buffers do begin to participate importantly in dissipating administered bicarbonate within the intervals examined here (27).

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[5] The erythrocythemia characteristic of animals with hypoxia-induced respiratory alkalosis did contribute in small part to the larger apparent space of distribution of bicarbonate. However, even if one discounts this effect, animals with chronic respiratory alkalosis would still feature a larger apparent bicarbonate space than did animals with chronic respiratory acidosis.

[6] The buffer value of hemoglobin and plasma proteins of normal subjects is stable over the physiologic range of pH (24). Moreover, the buffer value of plasma proteins is not significantly different in chronic metabolic acidosis as compared with chronic metabolic alkalosis (33). There are no data on the buffer value of tissue nonbicarbonate buffers during chronic alterations in acid-base equilibrium.

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