

Metabolic Effects of Blood Transfusion

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DISTURBANCES in the chemical composition of blood which occur during its collection and storage in acid-citrate-dextrose (ACD) solution are well known.^{1,2} Blood pH on collection is 6.9 to 7.0 and falls during storage to 6.4 or 6.5 by the end of three weeks. The fall in pH on collection is deliberately induced by the citric acid of ACD solution †; the subsequent fall reflects the accumulation of lactic acid generated by the metabolic activity of cellular elements of the blood. Citrate is added (as citric acid and sodium citrate) in high concentration to ensure adequate binding of calcium and thus to prevent coagulation. Plasma inorganic phosphate rises as organic phosphates are broken down within the cellular elements of the blood. At the low temperatures of storage the red cells lose their ability to retain potassium, which gradually accumulates in the plasma. The initially high dextrose, included in ACD solution to provide a metabolic substrate for the red cells, falls during storage. A less well-known alteration is the marked elevation in ammonium ion which occurs on storage, to which attention has recently been directed.

The composition of ACD solutions A and B appears in table 1, and the chemical changes of storage are summarized in table 2. If ACD blood is transfused slowly, the metabolic im-

pact on the recipient is of little concern. The current, frequent practice of rapid, massive transfusion has required careful re-examination of the chemical effects of such transfusion and the possibility of resultant harm to the patient; some such examinations have been made, but many questions remain unanswered. For example, there is uncertainty as to whether metabolic acidosis observed during rapid transfusion is caused by the transfusion of blood of low pH, or by the circulatory shock for which the transfusion is given. Similar uncertainties or disagreements remain concerning the significance of, or even the existence of, transfusion-induced disturbances in calcium, potassium, phosphate, and ammonium.

Regardless of how well or how poorly the patient may tolerate the chemical insult of multiple transfusions, and whether or not we can identify such effects at all in the complex clinical situation, it would appear desirable for a blood transfusion to be as near "normal" as possible. In recognition of this self-evident fact, fresh heparinized blood is the usual choice for total body perfusion during cardiac bypass and for exchange transfusion in newborn infants. Unfortunately the poor viability of blood stored in heparin and the difficulty

TABLE 1. The Formulae of ACD Solutions A and B and the Volumes of Each Required for 100 ml. Volumes of Donor Blood

	Solution A	Solution B
Trisodium citrate ($\text{Na}_3\text{C}_6\text{H}_5\text{O}_7 \cdot 2\text{H}_2\text{O}$)	22.0 g.	13.2 g.
Citric acid ($\text{C}_6\text{H}_8\text{O}_7 \cdot \text{H}_2\text{O}$)	8.0 g.	4.8 g.
Dextrose ($\text{C}_6\text{H}_{12}\text{O}_6 \cdot \text{H}_2\text{O}$)	24.5 g.	14.7 g.
Water for injection (U.S.P.) to make	1,000 ml.	1,000 ml.
Volume per 100 ml. blood	15 ml.	25 ml.

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† Although markedly acid on collection at body, or at room temperature, blood pH returns close to normal at the temperature of storage, largely due to the markedly increased solubility of carbon dioxide. Thus, on collection in ACD solution pH is 6.9 or 7.0 measured at 37° C., but measured at 4° C., the temperature of storage, pH is found to be 7.3 or 7.4. Above pH 7.4 viability of red cells is severely jeopardized, and it is to prevent hypothermia-induced alkalosis during storage that an acid preservative is added.

TABLE 2. Characteristics of ACD-A Plasma During Storage at $4^{\circ} \pm 1^{\circ}\text{C}$.

	Unit Value	Days Stored				
		0	7	14	21	28
Dextrose	mg./100 ml.	350	300	245	210	190
Lactic acid	mg./100 ml.	20	70	120	140	150
Inorganic phosphate	mg./100 ml.	1.8	4.5	6.6	9.0	9.5
pH*		7.00	6.85	6.77	6.68	6.65
Hemoglobin	mg./100 ml.	0-10	25	50	100	150
Sodium	mEq./l.	150	148	145	142	140
Potassium	mEq./l.	3-4	12	24	32	40
Ammonia	$\mu\text{g.}/100\text{ ml.}$	50	260	470	680	—

* Determined with glass electrode.

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in control of hemostasis in the recipient render heparin unsuitable for routine preservation and transfusion. ACD remains the standard preparation and probably will for some time. Accordingly it is not surprising to find that much effort has been made to correct the "chemical lesion" of ACD blood prior to its administration. Addition of bicarbonate³ or THAM⁴ can correct the acid pH, and passage across selected ion exchange resins can remove citrate, phosphate, lactate, potassium and ammonium.⁵ But before the routine adoption of measures which might themselves be harmful, it is well to examine carefully the disturbances which they are supposed to prevent or correct.

Acid-Base Balance

The collection of blood in either of the standard ACD solutions is associated with an initial pH of 6.9 to 7.0 which falls during a three week period of storage to 6.4 or 6.5 as dextrose is glycolized to lactic acid.^{1,2} Much attention has been directed to the apparent likelihood that such an acid preparation might be harmful.^{7,8,9} The danger of the hydrogen ion "load" has probably been overstated, however.

Each 500 ml. unit of ACD blood contains approximately 0.6 g. citric acid, equivalent to 8.6 mM hydrogen ion; lactic acid may rise to above 100 mg./100 ml. contributing another 5-6 mM hydrogen ion in each unit of blood.

Thus, to a unit of three week old blood approximately 14 mM hydrogen ion has been added as citric or lactic acid. If this amount of citric and lactic acid were infused directly into the patient and were to remain unmetabolized and in the extracellular fluid, a rapid depletion of blood and body buffers might be expected. But, of course, this is not the case. Virtually all of the added hydrogen ion has been buffered prior to transfusion by plasma bicarbonate and by hemoglobin and other protein within the donor red cells. At worst the patient receives blood in which the buffers have already been sharply reduced with a resultant dilution of the patient's extracellular bicarbonate. Assuming a constant alveolar ventilation and arterial Pco_2 , this fall in bicarbonate would, of course, result in a fall in pH.

The initial effect of transfusion is a fall in bicarbonate, but as the citrate and lactate of transfused blood are rapidly redistributed and metabolized new bicarbonate is formed which is immediately available to buffer new hydrogen ion; and the ultimate effect of multiple transfusion on acid-base balance is a metabolic alkalosis. More than half of the citric acid in ACD solution is added as the sodium salt, and thus as citrate is metabolized to bicarbonate, a considerable excess of sodium and of bicarbonate ions remains.^{10,11}

An important question remains to be answered. Is there a brief but severe acidosis?

It would appear inevitable that an initial increase in hydrogen ion (*i.e.*, fall in pH) must occur when acid blood is given very rapidly. This effect has been clearly shown by Foote, Trede, and Maloney,¹² who reported an initial, transient fall in pH to 7.25 during cardiopulmonary bypass using ACD blood. The remarkable thing about this observation is that the acidosis was so mild and so brief.

A few simple calculations help to explain the probable events. Assume that acid-base balance is initially normal in the patient, including a normal bicarbonate of 25 mM/liter. Assume that citric and lactic acids have combined with all, or nearly all of the bicarbonate of the donor blood, and that the donor blood bicarbonate (corrected to P_{CO_2} 40 and fully oxygenated) is approximately 5 mM/liter. Assume that the patient's blood volume is 5,000 ml., and that 5,000 ml. of ACD blood (with heparin and calcium added) is used to "prime" the oxygenator. And assume that on beginning bypass there is instantaneous complete mixing. The resultant bicarbonate concentration in the patient and oxygenator should then be 15 mM/L., or the average of the patient's and the donor blood. A fall in bicarbonate of 50 per cent, or nearly 50 per cent, will result in a fall in pH to 7.1, assuming P_{CO_2} remains unchanged (*i.e.*, if bicarbonate falls 50 per cent, the ratio of bicarbonate to carbonic acid falls from the normal 20:1 of pH 7.4 to 10:1 of pH 7.1). One must now consider in addition that bicarbonate will distribute itself very rapidly throughout the extracellular space. Thus, if one assumes a further twofold increase in the volume of distribution, bicarbonate should be close to 20 mM/liter, after initial redistribution. A fall in bicarbonate of this magnitude would result in a fall in pH to 7.25 or 7.3 (again assuming constant P_{CO_2}) and this is precisely what Foote and his colleagues found.

The above example, although somewhat simplified, accurately reflects the mechanism of dilutional acidosis which may occur during the rapid transfusion of ACD blood. The effect is one of transient dilution or depression of bicarbonate concentration. An important corollary is that the transfusion does not add free hydrogen ion. The changes in pH which occur are most appropriately con-

sidered as secondary to changes in bicarbonate and to changes in the ratio of bicarbonate to P_{CO_2} . The importance of P_{CO_2} , as well as of bicarbonate, should be noted. Howland, and Schweizer,¹³ have reported marked elevations in P_{CO_2} in stored ACD blood, and it is apparent that the low pH of such blood is at least partly secondary to a high P_{CO_2} . Attention should also be called to the need for increase in ventilation to excrete the additional free carbon dioxide contained in donor blood, and resulting from the metabolism of citrate and lactate; but even at a rate of transfusion of 500 ml. of blood every 5 minutes the increase in CO_2 to be excreted by the lungs would be less than 10 per cent.

The beginning of cardiopulmonary bypass represents the ultimate in "ultra" massive transfusion, and as such deserves special consideration. Furthermore, it provides the opportunity to observe the effects of large volumes of blood given electively as we have seen above, and to separate these effects from the hypoxic metabolic effects of shock. Severe metabolic acidosis has been frequently reported to occur during and following cardiopulmonary bypass, but it is generally recognized to be caused by inadequate tissue perfusion at low pump flow rates¹⁴ and not related to the composition of the transfused blood (which, for this special purpose, is often heparinized, fresh, and of essentially normal chemical composition).

Exchange transfusion in newborn infants offers another opportunity to observe the effects of very large transfusion under elective, or semi-elective conditions. Severe acidosis may occur and has been attributed to transfused ACD blood.^{15, 16} However, it is by no means clear what the state of the circulation is in these sick infants, who during the exchange are allowed to undergo proportionately large and rapid fluctuations in blood volume. There is reason to suspect the acidosis to be caused by shock rather than transfusion.

Whether metabolic acidosis is caused by transfusion, or, as seems more likely, by the shock for which transfusion is given, it would seem desirable to prevent or to correct it. Nahas and his associates have recommended the addition of tromethamine (THAM) to ACD blood prior to its administration to re-

thetic agents^{31,32} has recently been reported, and the possible increased hazards of citrate accumulation secondary to decreased metabolism during hypothermia,³³ or in the presence of liver disease have also been suggested.²³

It is argued that citrate-induced hypocalcemia is prevented by the rapid mobilization of new calcium from bone. The evidence for this opinion is the known ability of the experimental animal to replace calcium lost when his blood is passed repeatedly over an ion exchange resin,^{34,35} and the slight increase in total calcium which may occur during the infusion of ACD solution²⁶ or sodium citrate.³⁶ There is little doubt that bone calcium is mobilized in large quantities in response to an elevated serum citrate. Weidner and Clowes reported elevations in serum total calcium from a control of 10 mg./100 ml. to 15 mg./100 ml. during the infusion of citrate in nephrectomized dogs.³⁷ But during their parallel studies in dogs with intact kidneys, massive urinary losses of calcium and citrate occurred, and serum total calcium fell markedly. The observation of citrate induced calciuria and hypocalcemia has been amply confirmed.^{38,39,40}

Evidence for or against calcium mobilization during transfusion in man is fragmentary. However, in a large series of patients, this author has previously failed to observe any evidence of increase in serum total calcium accompanying increases in citrate.²³ Failure to observe elevations in calcium during clinical transfusion may be attributable to renal losses or may be secondary to elevations in phosphate. Serum inorganic phosphate becomes markedly elevated during the storage of blood^{1,2} and may be markedly elevated in the patient during transfusion.⁴¹ The hypocalcemic effects of an increased serum inorganic phosphate are well known^{42,43,44} and could be expected to offset any calcium mobilizing effect of an elevated citrate.

It has also been suggested that the low blood pH which may occur in the transfused patient may prevent or minimize the effects of hypocalcemia. Although a fall in pH within the physiologic range is associated with a modest shift from protein bound to ionized calcium,^{45,46} any effect of changes in pH which might be observed clinically on the binding

of calcium by citrate is negligible,⁴⁷ and it certainly cannot be assumed that a fall in pH will lessen any effects of citrate. To the contrary, acidosis will most certainly add to the problem, since renal excretion of citrate is known to be markedly depressed by a fall in pH.^{48,49}

If the toxic effects of elevated serum citrate are caused by the binding of calcium, they should be prevented by the administration of an appropriate calcium salt. In the animal and in man, calcium completely reverses the hemodynamic effects of experimentally elevated serum citrate.^{24,50} The elevation of serum citrate is rate dependent, and the uptake, distribution and metabolism of citrate should be accurately predictable. It is difficult, however, to predict the degree of citrate rise or ionized calcium fall during the rapidly changing circumstances of clinical transfusion, and accordingly, there is at present no reliable way to calculate an appropriate dose of calcium.

The direct measurement of ionized calcium in the patient's blood would, of course, provide the most useful information on which to base therapy, but unfortunately no practical laboratory methods for this measure are currently available. Valuable indirect information can, however, be gotten from the Q-T interval of the electrocardiogram. Monitoring of the electrocardiogram is routine during exchange transfusion in infants^{51,52} and should be routine in all patients during massive transfusion.

If calcium is administered empirically during transfusion, it is likely that overdoses may be given, and it has been suggested that the routine use of calcium during transfusion may increase rather than decrease mortality.⁵³ It is difficult to interpret the clinical evidence presented in support of this thesis, but one may reasonably adopt a cautious attitude towards calcium replacement. But certain situations clearly call for calcium. In the use of citrated blood for open heart surgery, it is absolutely necessary to replace calcium prior to total body perfusion. Foote, Trede and Maloney⁵² have calculated that approximately 0.6 g. calcium chloride are necessary to offset the excess citrate of one 500 ml. unit of ACD blood, and this "formula" has been adopted by others.⁵⁴ Exchange transfusion with ACD

blood in the newborn infant may also be considered an absolute indication for calcium therapy, particularly since the reported rise in serum citrate^{55, 56} is sufficient to stop the heart in the experimental animal. It is also certainly reasonable to administer calcium in any situation where circulatory collapse accompanies multiple rapid transfusion, and in support of this at least a few well-documented case reports can be cited.^{57, 58, 59, 60} However, the increase in blood pressure which follows the administration of calcium⁶¹ must not be assumed to "prove" the prior existence of citric acid intoxication. Calcium is well known for its non-specific positive inotropic effects and is an effective antagonist to cardiac depression from many causes.

Another approach to the prevention of citrate intoxication has been suggested. Blood may be prepared for transfusion by passage across an ion exchange resin,⁶² thus removing calcium and preventing coagulation. Or, calcium and citrate of ACD blood may be removed by ion exchange prior to transfusion.^{6, 7} Such preparation still might seem to offer the danger of hypocalcemia, but here the mobilization of calcium probably is adequate to maintain calcium levels; and in any event, it should be possible to calculate precisely how much calcium to administer with each transfusion of calcium-free blood. The difficulty with citrated blood, on the other hand, rests with the large excess of citrate which binds the calcium of the recipient as well as the calcium of the blood to be transfused. But while perhaps resolving the citrate problem, the use of ion exchange resins introduces a new and more serious set of problems. Most of the platelets and many of the coagulation factors are removed by exchange resins⁶³ and the massive transfusion of blood prepared in this manner would almost certainly add to the well-recognized coagulation difficulties which may occur.

In an attempt to avoid the danger of ACD citrate, the use of other solutions has been proposed. Blood preserved in citrate phosphate solution contains 20 per cent less citrate, but this slight advantage hardly justifies the recommendation⁶⁴ for its use to lessen the danger of citric acid intoxication. Versene (EDTA) has also been proposed,⁶⁵ but would

probably increase the dangers of hypocalcemia. As with citrate it produces anti-coagulation by binding calcium, but unlike citrate it is not metabolized, and could be expected to lead to increased, rather than decreased, accumulated effects.

Potassium

During the storage of blood at low temperatures potassium is released from red cells into the plasma, and at the end of three weeks plasma potassium may be as high as 15–20 mEq./liter or higher.^{1, 2, 3} On rewarming, the red cell regains its ability to maintain its normal intracellular-extracellular gradient, and potassium is "pumped" back into the cell.⁶⁶ Presumably this occurs as blood is transfused into the patient, since serum potassium has been reported to remain within normal limits during multiple transfusion.^{25, 67, 68} However, there remains some concern that dangerous hyperkalemia may be caused by multiple transfusions.^{69, 70} In at least one clinical situation, exchange transfusion in newborn infants this concern is well founded, serum potassium elevations above 10 mEq./liter having been reported.^{71, 72} Here again, it is not possible to separate out the effects of the elevated plasma potassium of the transfused blood, since severe acidosis, which in itself may cause acute hyperkalemia,^{73, 74, 75} commonly occurs during exchange transfusion, as discussed above.

The hemodynamic effects of hyperkalemia have recently been explored in some detail by Smith and Corbascio.⁷⁶ The intravenous administration of potassium chloride to the lightly anesthetized dog resulted in surprisingly little hemodynamic depression prior to death, which occurred in ventricular fibrillation at serum concentrations of approximately 10 mEq./liter. Only when potassium chloride was administered to reserpinized dogs did the expected marked circulatory depression occur, and in these animals cardiac arrest occurred in asystole at serum potassium levels of 15 mEq./liter. Studies of the interaction of increased potassium and decreased calcium in the isolated guinea pig auricle have also been carried out in these laboratories.³⁰ Elevations in potassium were shown to potentiate the

myocardial depressant effects of hypocalcemia as anticipated.

Phosphate

Marked elevations of inorganic phosphate occur during storage of blood,^{1,2} as mentioned earlier, and marked elevations of serum inorganic phosphate have been observed in the recipient of multiple transfusions.⁴¹ We have attempted to reproduce hyperphosphatemia by the rapid infusion of neutral sodium phosphate in the lightly anesthetized dog.³⁹ Infusions of phosphate several-fold greater than estimated ever to occur during transfusion in man have failed to elevate serum inorganic phosphate under these experimental conditions. It appears likely that elevations in phosphate occurring during clinical transfusion are at least partly dependent upon changes in acid-base balance, to which the serum inorganic phosphate concentration is acutely sensitive.^{73,77} Thus, the acidosis accompanying shock, in the treatment of which massive transfusion is usually given, can be expected to lead to a marked elevation in phosphate; and the transfusion itself very likely causes a further rise.

The possible etiologic role of marked elevations in serum inorganic phosphate in depression of serum total calcium, and possibly in the prevention of an increase in total calcium in response to citrate-induced fall in ionized calcium, has been alluded to above.

Miscellaneous

Lactate. The marked elevations in serum lactate which have been reported during massive transfusion are doubtless the result of many factors, shock, and perhaps anesthesia, as well as transfusion. Very large infusions of sodium lactate are well tolerated by the experimental dog,⁷⁸ and it can probably be assumed that any untoward effects of an elevated lactate are secondary to the effects of lactic acid on acid-base balance.

Ammonium. The increase in ammonium ion of banked blood is apparently well tolerated by normal man, but attention has recently been called to the possibility of harm in patients with liver disease.⁷⁹ Although this danger has not been documented clinically, it can certainly be added to the long list of in-

dications for the use of fresh blood in patients suffering from severe liver disorders. However, the danger of ammonium intoxication is probably not sufficient justification for treatment of ACD blood by its passage across an ion exchange resin, which has been recommended.^{80,81} Ammonium ion can certainly be removed by the resin, but, as mentioned above, with it are lost most of the platelets and many of the clotting factors,⁶³ substances of critical importance to patients suffering from liver disease.⁸²

Hypothermia. A final effect which should be mentioned is the fall in body temperature which may occur during the rapid transfusion of refrigerated blood.⁸³ Such inadvertent hypothermia can be expected to have its own direct, possibly deleterious effects, and to modify all metabolic responses. To consider these in detail is beyond the scope of the present review.

Summary and Conclusions

The many investigators who have concerned themselves with possible metabolic effects of blood transfusion have usually limited their attention to a single factor, to the exclusion of others. Examined individually none appear likely to be of great danger except under extreme conditions. But the clinical situation is never so simple, and many or all of the chemical alterations which have been discussed can be expected to occur simultaneously. During massive transfusion of citrated blood a rise in serum citrate will usually be accompanied by a fall in pH. Inorganic phosphate will rise. Total calcium may remain normal or near normal levels, but calculated ionized calcium falls. Serum potassium is usually within normal limits, but may rise markedly during exchange transfusion. If transfusion is administered during general anesthesia, these effects may be potentiated by the myocardial depressant effects of the anesthetic agent. If refrigerated blood is administered rapidly, the resultant hypothermia may further modify these effects. Any harmful metabolic effects of massive transfusion are almost certainly the result of the interaction of many of these simultaneous individual effects. To specify their combined effects with

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any precision is, of course, not possible. As we have seen, there is not even agreement on the occurrence and importance of the individual chemical disturbances.

Prevention of possible metabolic effects of transfusion can be achieved, at least under elective conditions, by the use of fresh heparinized blood, and this remains the preparation of choice for cardiopulmonary bypass and for exchange transfusion in newborn infants. However, when large amounts of blood are needed for emergency transfusion, we are limited to the use of blood as commonly collected and stored in ACD solution. And, in spite of the rather dramatic chemical alterations which occur in ACD blood and are described above, it remains by far the most serviceable preparation available. There is certainly none better in sight to replace it.

Efforts to correct the "chemical lesion" of collection and storage of blood in ACD solution should be viewed with some caution. The "corrected" blood may be more harmful than the unmodified blood. On the other hand, the possible metabolic effects of massive transfusion should be borne in mind, and diagnostic and therapeutic measures taken as appropriate.

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