A Randomized, Controlled, Double-Blind Crossover Study on the Effects of 2-L Infusions of 0.9% Saline and Plasma-Lyte® 148 on Renal Blood Flow Velocity and Renal Cortical Tissue Perfusion in Healthy Volunteers

Abeed H. Chowdhury, BSc, MRCS, Eleanor F. Cox, PhD,† Susan T. Francis, PhD,† and Dileep N. Lobo, DM, FRCS, FACS**

Objective: We compared the effects of intravenous infusions of 0.9% saline ([Cl^-] 154 mmol/L) and Plasma-Lyte 148 ([Cl^-] 98 mmol/L, Baxter Healthcare) on renal blood flow velocity and perfusion in humans using magnetic resonance imaging (MRI).

Background: Animal experiments suggest that hyperchloremia resulting from 0.9% saline infusion may affect renal hemodynamics adversely, a phenomenon not studied in humans.

Methods: Twelve healthy adult male subjects received 2-L intravenous infusions over 1 hour of 0.9% saline or Plasma-Lyte 148 in a randomized, double-blind manner. Crossover studies were performed 7 to 10 days apart. MRI scanning proceeded for 90 minutes after commencement of infusion to measure renal artery blood flow velocity and renal cortical perfusion. Blood was sampled and weight recorded hourly for 4 hours.

Results: Sustained hyperchloremia was seen with saline but not with Plasma-Lyte 148 ($P < 0.0001$), and fall in strong ion difference was greater with the former ($P = 0.025$). Blood volume changes were identical ($P = 0.867$), but there was greater expansion of the extravascular fluid volume after saline ($P = 0.029$). There was a significant reduction in mean renal artery flow velocity ($P = 0.045$) and renal cortical tissue perfusion ($P = 0.008$) from baseline after saline, but not after Plasma-Lyte 148. There was no difference in concentrations of urinary neutrophil gelatinase–associated lipocalin after the 2 infusions ($P = 0.917$).

Conclusions: This is the first human study to demonstrate that intravenous infusion of 0.9% saline results in reductions in renal blood flow velocity and renal cortical tissue perfusion. This has implications for intravenous fluid therapy in perioperative and critically ill patients. NCT01087853

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With annual sales of 10 million and 200 million liters in the UK and USA, respectively (data from Baxter Healthcare), 0.9% saline is probably the most commonly used intravenous crystalloid.[1] Large volume infusions of saline can cause hyperchloremic acidosis,[2–5] fluid overload,[6] interstitial edema,[3,7] and adverse postoperative outcomes.[8,9] These problems are less pronounced with the use of balanced crystalloids, which have an electrolyte composition closer to that of plasma.[2,4,10,11]

Hyperchloremia may also affect renal function adversely. Animal studies have suggested that it is the critical determinant for changes in renal blood flow, mediated primarily by effects on afferent and intrarenal arterial vessels.[12–15] In canine experiments, intrarenal infusion of chloride-containing solutions, such as 0.9% saline or NH_4Cl, led to reductions in renal blood flow and glomerular filtration rate (GFR).[12] Other animal experiments have shown that K^+-induced reduction in renal vessel diameter was both dependent on and responsive to increasing extracellular chloride.[14,16] Moreover, at pathologically elevated concentrations, chloride led to severe renal vasoconstriction.[14]

Although human studies have shown that renal excretion of an acute saline load is sluggish when compared with balanced crystalloids,[2,7] and it is largely dependent on a slow and sustained suppression of the renin-angiotensin-aldosterone axis,[3] the effects of saline excess on renal hemodynamics have hitherto not been studied in humans.

In this study in healthy adult human volunteers, we used a validated experimental model[2,3,7] to study physiological responses to an intravenous infusion of 2 L of 0.9% saline and Plasma-Lyte 148 (both Baxter Healthcare, Thetford, UK) over 1 hour. The aims of this study were to compare the potential of the infusions to produce hyperchloremic acidosis; changes in blood and extravascular fluid volume; and changes in renal volume, renal artery blood flow velocity, and renal cortical tissue perfusion using validated magnetic resonance imaging (MRI) techniques.[17–19]

SUBJECTS AND METHODS

We performed this randomized, double-blind, cross-over study at a university teaching hospital and recruited 12 healthy adult male volunteers with a body weight of 65 to 85 kg after obtaining informed written consent. Those with acute illness in the preceding 6 weeks, on regular medication, with a history of substance abuse or having factors precluding MRI were excluded. The UK National Research Ethics Service and Medicines and Healthcare Products Regulatory

From the *Division of Gastrointestinal Surgery, Nottingham Digestive Diseases Centre National Institute for Health Research Biomedical Research Unit, Nottingham University Hospitals, Queen's Medical Centre; and †Sir Peter Mansfield Magnetic Resonance Centre, School of Physics and Astronomy, University of Nottingham, Nottingham, UK.

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Reprints: Dileep N Lobo, DM, FRCS, FACS, Division of Gastrointestinal Surgery, Nottingham University Hospitals, Queen's Medical Centre, Nottingham NG7 2UH, UK. E-mail: dileep.lobo@nottingham.ac.uk.

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Agency granted approvals. The protocol was registered at http://www.clinicaltrials.gov (NCT01087853).

Sample Size

Compared with balanced solutions, 2 L infusions of 0.9% saline can increase serum chloride concentrations by a mean (SD) of 4 (3.5) mmol/L,[1] a change anticipated to be adequate to show differences in renal hemodynamics. In order to detect an increase in serum chloride concentration from 104 mmmol/L to 108 mmmol/L after saline with an α error of 0.05 and a power of 90%, 10 subjects were required. Allowing for a 20% dropout rate, we recruited 12.

Randomization and Masking

Randomization was based on a computer-generated pseudo-random code using random permuted blocks of varying size, created by the Nottingham Clinical Trials Unit. There was equal probability to receive 1 of the 2 crystalloid infusions on the first study visit. Crossover studies using the alternate crystalloid were performed 7 to 10 days later. Access to the sequence was confined to the data manager and an independent pharmacist who masked the infusions with an opaque covering. The allocations were concealed from the investigators until completion of all interventions, data collection, and analyses.

Baseline Assessment

Participants reported at 0900 hours following a fast from midnight and after having abstained from alcohol, nicotine, and caffeine from 1800 hours. They did not eat or drink for the duration of the study. After voiding of the bladder, height and weight were measured to the nearest 0.01 m and 0.1 kg, respectively, using Salter 9000SV3R scales (Salter UK, Tonbridge, Kent, UK). Participants were allowed to stand to void urine and be weighed, but blood samples were taken after lying supine for at least 10 minutes.

A 16 G venous cannula (BD Venflon, Franklin Lakes, NJ) was inserted into each antecubital fossa (right for blood sampling, left for infusion). An initial 10 mL blood sample was drawn 10 minutes after cannula insertion for analysis of full blood count, serum electrolytes, urea, creatinine, albumin, and osmolality. A preinfusion urine sample was analyzed for osmolality and for concentrations of urea, sodium, potassium, and neutrophil gelatinase-associated lipocalin (NGAL). In addition, a 24-hour urine sample was collected before each study for calculation of creatinine clearance.

Interventions

Two 1-L bags of 0.9% saline or Plasma-Lyte 148 (Table 1) were infused over 1 hour in random order on separate occasions 7 to 10 days apart with subjects supine, using 2 MRI-compatible infusion pumps (MEDRAD Continuum MR Infusion System, MEDRAD Inc, Warrendale, PA) and starting at time 0. Body weight measurements and blood samples were repeated at 60, 90, 120, 180, and 240 minutes after the start of the infusion.

Participants were permitted to pass urine as needed and, in all cases, at the end of the study. The time of each micturition was noted, and urine volume was measured. Urine pooled over the 4-hour period was analyzed for osmolality and concentrations of urea, creatinine, sodium, potassium, and urinary NGAL.

TABLE 1. Characteristics of the 2 Crystalloid Infusions

	0.9% Saline	Plasma-Lyte 148
Sodium (mmol/L)	154	140
Chloride (mmol/L)	154	98
Potassium (mmol/L)	—	5
Magnesium (mmol/L)	—	1.5
Acetate (mmol/L)	—	27
Gluconate (mmol/L)	—	23
Strong ion difference (mmol/L)	0	50
Sodium supplied as	NaCl 9 g/L	NaCl 5.26 g/L
pH	5.4	7.4
Theoretical osmolarity (mOsm/L)	308	294.5
Total solute weight (g)/L	9	14.6
$[Na^+]$:$[Cl^-]$ ratio	1:1	1.43:1

Hematologic and Biochemical Analyses

Hematologic and biochemical parameters were measured by methods we have used previously, with interassay imprecision expressed as coefficients of variance of 0.6% to 4%.[2,3,7] Urinary NGAL was measured by enzyme-linked immunosorbent assay (BioPorto Diagnostics, Gentofte, Denmark). Interassay variation expressed as a coefficient of variance (range) was 3.4% (2.5%–8.4%).

Derived Values

Creatinine clearance was calculated from a standard formula.[20] Blood volume at time 0 was estimated according to the method described by Nadler et al.[21]

$$BV_0 = 0.03219 \times BW_0 + 0.3669 \times Ht^3 + 0.6041,$$

where BV_0 is blood volume in liters at time 0, BW_0 is body weight in kilogram at time 0, and Ht is the height in meter. Calculations for changes in blood volume and extravascular fluid volume were based on changes in hematocrit and body weight and were made using the formula we have described previously.[3]

$$\Delta Hct_t(\%) = \frac{Hct_0 - Hct_t}{Hct_0} \times 100,$$

where ΔHct_t (%) is the percentage reduction in hematocrit at time t, Hct_0 is the hematocrit at time 0, and Hct_t is the hematocrit at time t.

$$\Delta BV_t(\%) = BV_0 \left\lfloor \frac{\Delta Hct_t}{Hct_t} \right\rfloor \times 100,$$

where ΔBV_t(%) is the percentage change in blood volume at time t, and

$$BV_t(L) = \frac{BV_0 \times [100 + \Delta BV_t(\%)]}{100}$$

where BV_t (L) is the blood volume in liters at time t.

$$\Delta BV_t(L) = BV_t - BV_0,$$

where ΔBV_t (L) is the change in blood volume in liters at time t from baseline (time 0).

$$\Delta TBW_t(L) = BW_t - BW_0,$$

where ΔTBW_t (L) is the change in total body water in liters at time t, BW_t is the body weight in kilograms at time t and BW_0 is the body weight in kilograms at time 0.

$$\Delta EFV_t(L) = \Delta TBW_t - \Delta BV_t,$$

where ΔEFV_t (L) is the change in extravascular fluid volume in liters at time t, ΔTBW_t is the change in total body water in liters at time t from baseline (time 0), and ΔBV_t is the change in blood volume in liters at time t from baseline (time 0).

The apparent strong ion difference (SID_a) was calculated as described by Stewart,[22]

$$SID_a \text{ (mmol/L)} = [Na^+] + [K^+] - [Cl^-].$$

MRI Protocol

Images were obtained using a 1.5T Philips Achieva MR scanner (Philips Healthcare Systems, Best, The Netherlands). Subjects were scanned supine using a body transmit and 4-element sensitivity encoding (SENSE) torso coil. A series of multislice balanced-Turbo-Field-Echo images were initially acquired in 3 orthogonal planes to locate the kidney and vessels of interest to aid slice positioning. The MRI protocol consisted of a series of noninvasive MRI measurements to assess renal cortical tissue perfusion and renal artery blood flow velocity.

A base equilibrium scan was also acquired for quantification of renal cortical tissue perfusion. At the start of the infusion, data were collected from arterial spin labeling (ASL) and phase contrast angiography (PCA)-MRI to determine renal blood flow velocity[18] and then scans were repeated at specific time points over the course of the 60-minute infusion. Each ASL measurement took approximately 5 minutes, and each PCA was collected in a breath-hold. After the infusion, an additional 2 sets of PCA-MRI measurements were collected such that measurements were made up to 90 minutes. During the infusions, subjects were scanned at 7-minute intervals to assess the time-course of the response.

ASL Protocol

Respiratory-triggered ASL data (288 × 300 mm field of view, 3 × 3 mm^2 voxel, 8-mm slice thickness, 5 slices) were collected with a True-FISP readout [echo time/repetition time 2.1/4.1 ms, SENSE 2, flip angle 60°, low-high acquisition, and half-Fourier acquisition].[19]

Phase Contrast-MRI Protocol

Phase contrast (PC)-MRI on the right renal artery was performed using a single slice turbo-field-echo technique with the imaging slice placed perpendicular to the vessel, 15 phases were collected across the cardiac cycle. Imaging parameters were: repetition time/echo time 6.9/3.7 ms, flip angle 25°, number of excitations 2, reconstructed resolution 1.17 × 1.17 × 6 mm^3, turbo-field-echo factor depended on the subjects' heart rate, velocity encoding 100 cm/s for renal artery. Each PC measurement was acquired during a single 20 to 25 seconds' breath hold.

Data Analysis

The PCA data were analyzed using the Philips Q-flow software (Philips Medical Systems). A region of interest was drawn over the vessel of interest, and the mean velocity (cm/s) over the cardiac cycle, across the vessel, calculated. In a separate scan session, the within-session reproducibility of measured mean renal artery flow velocity was assessed and the coefficient of variance was found to be 2.7%.

For the ASL data, ASL tag and control images were motion corrected to the base image using FMRIB Software Library and difference images (label-control) calculated.[19] Individual difference images were then averaged to create a single perfusion-weighted difference map. A perfusion (*f*) map in units of mL/100 g/min was then formed using a kinetic model[23] using the individuals' perfusion-weighted difference image, base equilibrium image, and known tissue T_1.[24] Mean renal cortical perfusion values were then estimated by segmenting the kidney into tissue type and averaging across both kidneys. As with renal artery flow velocity, within-session coefficient of variance of measured renal cortical perfusion was assessed and found to be 3.3%.

Statistical Analysis

Grouped data (0.9% saline vs Plasma-Lyte 148) are represented as mean (SEM [standard error of mean]). The significance of differences between the 2 groups was tested using the repeated-measures analysis of variances with Bonferroni's correction for multiple comparisons and the Student paired *t* test, with differences being considered significant at $P < 0.05$. GraphPad Prism v 5.0d for Macintosh statistical software package (GraphPad Software Inc, La Jolla, CA) was used.

TABLE 2. Baseline Parameters Before Infusion

	Before Saline	Before Plasma-Lyte 148	*P*
Weight (kg)	70.7 (2.1)	70.7 (2.1)	0.959
Height (m)	1.80 (0.02)	1.80 (0.02)	1.000
Body mass index (kg/m^2)	21.8 (0.5)	21.8 (0.5)	0.948
Hemoglobin (g/dL)	14.8 (0.2)	14.8 (0.2)	0.758
Hematocrit	0.444 (0.005)	0.442 (0.005)	0.707
Chloride (mmol/L)	103 (0.3)	104 (0.4)	0.206
Strong ion difference (mmol/L)	42.7 (0.4)	42.4 (0.8)	0.621
Bicarbonate (mmol/L)	30.2 (0.5)	29.1 (0.6)	0.214
Serum albumin (g/L)	43.5 (0.4)	43.9 (0.7)	0.435
Serum osmolality (mOsm/kg)	296 (1.0)	297 (1.4)	0.383
Serum creatinine (μmol/L)	87 (2.6)	85 (3.0)	0.062
Creatinine clearance (mL/min)	118.9 (5.4)	121.5 (5.5)	0.074
Renal artery blood flow velocity (cm/s)	34.8 (1.5)	33.4 (1.8)	0.180
Renal cortical tissue perfusion (mL/100 g/min)	333 (16)	324 (16)	0.615
Renal volume (mL)	300 (12)	310 (14)	0.243
Calculated blood volume (L)	5.0 (0.1)	5.0 (0.1)	0.959

n = 12, all values mean (SEM). Differences were not significant for all parameters (Student paired *t* test).

RESULTS

Thirteen male subjects were approached and 12, with a mean (SEM) age of 22.7 (0.7) years, were recruited. Baseline parameters before each infusion were similar (Table 2). All participants completed both arms of the study and were included in the analysis. None reported adverse effects.

Changes in Weight, Blood Volume, and Extravascular Fluid Volume

Weight changes were proportional to the volume of crystalloid infused and urine excreted (Fig. 1 and Table 3). At the end of the study (240 minutes), the increase in weight was 1.2 kg following 0.9% saline compared to 0.84 kg following Plasma-Lyte 148 ($P = 0.022$). Both infusions produced similar changes in hematocrit, hemoglobin, and serum albumin concentration, reflecting equivalent plasma dilution (Fig. 1). Initial plasma expansions of 29% and 26% of the infused volume were observed for Plasma-Lyte 148 and 0.9% saline, respectively. At the end of 4 hours, 14% of Plasma-Lyte 148 and 12% of 0.9% saline infusions remained within the intravascular compartment.

An expansion of the calculated extravascular fluid compartment was greater after 0.9% saline (Fig. 1). At its peak, the change in extravascular fluid volume was 1484 mL following 0.9% saline infusion compared with 1155 mL following Plasma-Lyte 148 ($P = 0.031$). This surplus of extravascular fluid associated with 0.9%

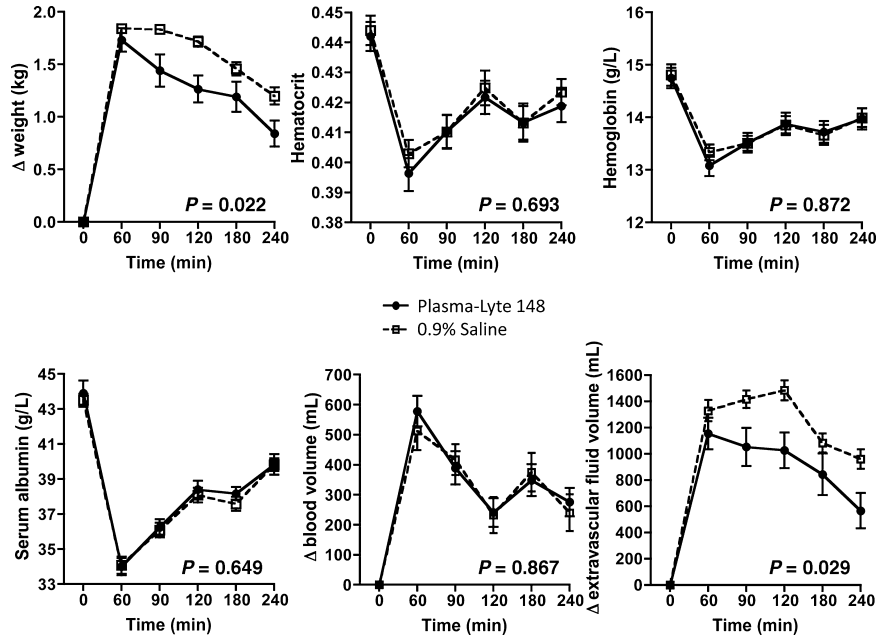

FIGURE 1. Changes in body weight, hematocrit, hemoglobin, serum albumin, blood volume, and extravascular fluid volume after infusion of 2 L of 0.9% saline and Plasma-Lyte 148 over 1 hour. All values are mean (SEM). The *P* values are for the test of 0.9% saline versus Plasma-Lyte 148 using the analysis of variances and a repeated measures model.

TABLE 3. Urinary Changes

	0.9% Saline	Plasma-Lyte 148	*P*
Time to first micturition after start of infusion (min)	142 (16)	90 (12)	0.006
Postinfusion urinary volume (mL)	533 (57)	833 (87)	0.002
Preinfusion urinary osmolality (mOsm/kg)	859 (82)	836 (68)	0.629
Postinfusion urinary osmolality (mOsm/kg)	549 (39)	418 (37)	0.004
Total postinfusion urinary sodium (mmol)	128 (13)	104 (12)	0.025
Total postinfusion urinary potassium (mmol)	60 (8)	49 (6)	0.244

n = 12, all values mean (SEM). Statistical significance was calculated using the Student paired *t* test.

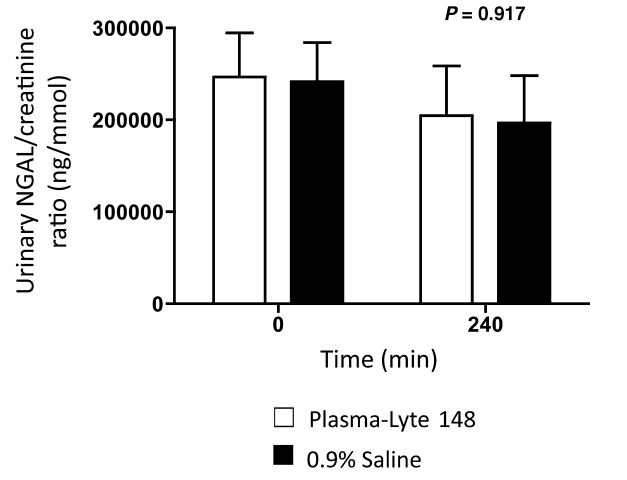

FIGURE 2. Changes in urinary NGAL/urinary creatinine ratio after infusion of 2 L of 0.9% saline and Plasma-Lyte 148 over 1 h. All values are mean (SEM). The *P* value is for the test of 0.9% saline versus Plasma-Lyte 148 calculated using the Student paired *t* test.

saline persisted at all time points and at the end of the study equated to a difference of 394 mL ($P = 0.043$).

Urinary Volumes, Biochemistry, and Urinary NGAL

Urinary responses are shown in Table 3. Urinary concentration of NGAL expressed as a ratio of urinary creatinine (to correct for differences in volume) was not increased following either infusion, and differences between the infusions were not significant (Fig. 2).

Changes in Serum Biochemistry and Apparent Strong Ion Difference

After infusion of 0.9% saline, serum chloride concentrations peaked at 109 mmol/L and remained above the upper limit of the physiological range (105 mmol/L) for the duration of the study (Fig. 3). Hyperchloremia was not seen after Plasma-Lyte 148. Analogous changes were observed with the apparent SID_a, the fall in which was significantly lower after 0.9% saline than Plasma-Lyte 148 (Fig. 3), indicating an acidemia after saline. Changes in serum osmolality and concentrations of sodium and potassium were similar after both infusions (Fig. 3).

Changes in Renal Artery Blood Flow Velocity, Renal Volume, and Renal Cortical Tissue Perfusion Determined by MRI

The 2 infusions produced significantly different responses in renal artery blood flow velocity. After an initial rise lasting 14 minutes, renal artery blood flow velocity returned toward but not below

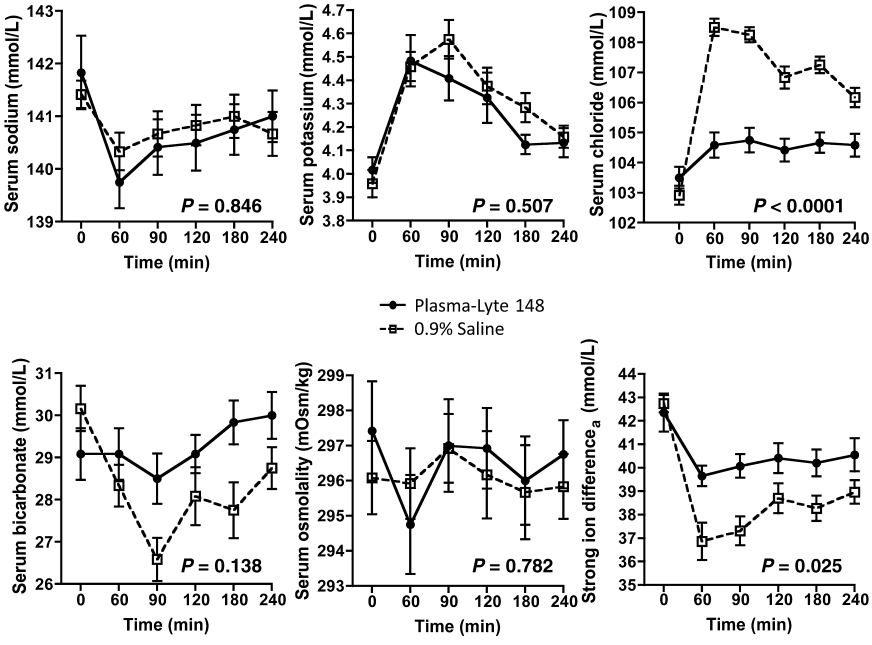

FIGURE 3. Changes in serum sodium, potassium, chloride, bicarbonate, osmolality, and apparent SID after infusion of 2 L of 0.9% saline and Plasma-Lyte 148 over 1 hour. All values are mean (SEM). The *P* values are for the test of 0.9% saline versus Plasma-Lyte 148 using the analysis of variances and a repeated measures model.

baseline velocities following Plasma-Lyte 148 infusion and remained stable for the remainder of the 90-minute MR scanning period (Fig. 4). In contrast, after 7 minutes of 0.9% saline infusion, there was a progressive decline in renal artery blood flow velocity, with a maximum reduction in velocity compared to baseline of 13% observed after 42 minutes. At the end of the MR scanning period of 90 minutes, 0.9% saline infusion had produced a decrease in mean renal artery blood flow velocity of 3.0 cm/s, representing a 9% reduction in velocity compared to baseline (Fig. 4). In this study, the mean (SD) baseline renal artery area was 0.23 (0.07) cm^2. Change in cross-sectional area of the renal artery could not be detected in response to the infusion because this was below the spatial resolution of the PCA measurement (the minimum detectable change is 0.05 cm^2, which is equivalent to a 22% change), thus renal artery blood flow velocity (cm/s) rather than renal artery flow (mL/min) measures are reported.

Both infusions increased renal volumes, with a peak increase of 4% following 0.9% saline and 3% following Plasma-Lyte 148 (Fig. 4). After an initial rise in renal cortical tissue perfusion following Plasma-Lyte 148 infusion, values returned toward but not below baseline (Fig. 4). However, after infusion of 0.9% saline, there was a progressive decline in renal cortical tissue perfusion with a maximum reduction observed at 28 minutes of 39.3 mL/100 g/min, equating to an 11.7% reduction in cortical tissue perfusion from baseline.

DISCUSSION

This is the first study in humans to show that the intravenous infusion of 2 L of 0.9% saline over 60 minutes results in reductions in renal blood flow velocity and renal cortical tissue perfusion, changes not observed after infusion of a balanced crystalloid. These findings are consistent with observations from animal experiments, which have shown unfavorable effects of elevated extracellular chloride concentration on vascular resistance,[25,26] GFR,[12,26] and renin activity.[27,28] The degree of congruence of changes in serum biochemistry and blood and extravascular fluid volumes with those seen in our previous experiments[2,3,7] provide further validation of reproducibility of the model.

The chloride concentration in 0.9% saline is 1.5 times that of plasma, and the mechanism for the hyperchloremic acidosis is explained by the Stewart hypothesis,[22] whereby an increase in plasma chloride concentration decreases the SID_a and leads to acidosis. Balanced crystalloids, which have a chloride concentration much closer to that of plasma, do not give rise to this effect. Canine experiments on resuscitation from septic shock have shown that 0.9% saline accounted for more than one third of the acidosis observed, whereas lactate was responsible for only 10% of the acidosis.[5] The authors felt that a large proportion of the acid load could be attributed to differential shifts of Na^+ and Cl^- from the extravascular to the intravascular space.[5]

An unexpected finding in this study is that Plasma-Lyte 148 caused a decrease in SID_a. According to the Stewart hypothesis,[22] an increase in SID_a would be predicted if both the acetate and gluconate were completely metabolized. Although it is well known that acetate is rapidly metabolized, the fate of gluconate is not clear and it may be mostly eliminated unchanged in the urine, as it is in animals.[29] It is also curious that although Plasma-Lyte 148 contains only 98 mmol/L of chloride (less than the mean baseline value), the mean serum chloride concentrations increased marginally after infusion. This modest increase would explain the less than expected alkalinizing effect of the Plasma-Lyte 148, but it is not clear how this increase occurs, unless there were shifts of chloride from the intracellular space, exchanging for either acetate or gluconate.

In other canine experiments, intrarenal infusion of solutions containing chloride, such as 0.9% saline or 1.2 M NH_4Cl, led to reductions in total renal blood flow and GFR in both denervated and in situ kidneys.[12] This effect was absent for non–chloride-containing solutions such as dextrose or sodium bicarbonate.[12] Other experiments confirmed that reductions in renal arterial diameter are both dependent and responsive to increasing extracellular chloride with severe vasoconstriction observed at pathological concentrations.[14,16] Hyperchloremia may inhibit proximal tubular chloride reabsorption, increasing chloride delivery to the distal nephron, with subsequent negative feedback to afferent renal vessels to limit flow.[12] These chloride-sensitive responses have been confirmed in animal models[15,30] and lead to a reduction in renal artery blood flow, decrease in GFR, and suppression of renin secretion. We postulate that the decrease in renal artery flow velocity is in response to a probable decrease in

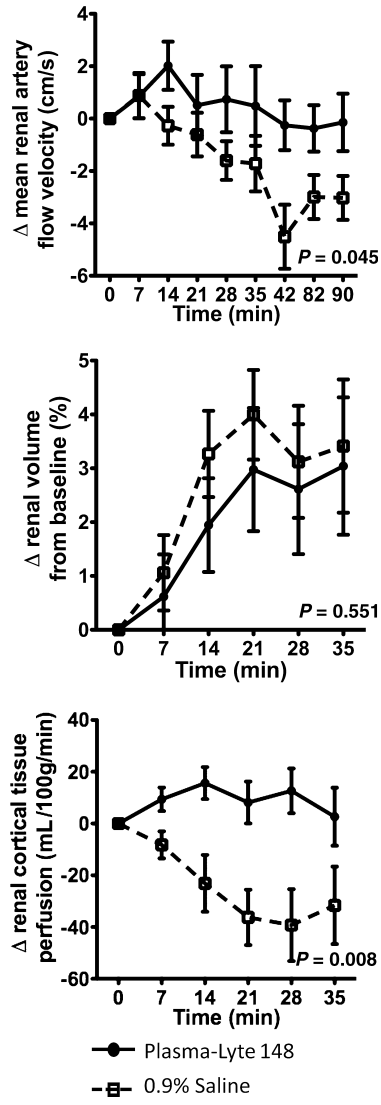

FIGURE 4. Changes in renal artery blood flow velocity, renal volume, and renal cortical tissue perfusion after infusion of 2 L of 0.9% saline and Plasma-Lyte 148 over 1 hour. All values are mean (SEM). The *P* values are for the test of 0.9% saline versus Plasma-Lyte 148 using the analysis of variances and a repeated measures model.

diameter of the renal artery and arterioles, and that this decrease in flow velocity led to a decrease in renal tissue perfusion. Poiseuille's law states that the laminar flow rate (Q) of an incompressible fluid along a tube is proportional to the fourth power of the radius (r) of the tube and is also dependant upon fluid viscosity (η), length of the tube (L), and the pressure difference between the ends (P). Flow rate (Q) (mL/min) is derived from the product of flow velocity (v) (cm/s) and the cross-sectional area of the vessel (πr^2 measured in cm^2), thus for a given pressure difference, flow velocity is proportional to the square of the radius. A decrease in flow velocity will accompany a decrease in vessel radius and lead to a greater decrease in flow.

More recently, the role of the macula densa in providing tubuloglomerular feedback to afferent vessels and also the signaling pathway leading to changes in GFR has been elucidated.[31,32] Elevated renal tubular chloride concentration results in the entry of chloride into the cells of the macula densa causing depolarization of the basolateral membrane via chloride channels.[33] Depolarization causes adenosine to be released from the macula densa, which in turn provides the signal for increased afferent arteriolar resistance and reduced GFR.[31,32]

Changes in circulating volume also influence renal blood flow and perfusion. However, in the present study, the calculated expansion of blood volume was identical. For this reason, we suggest the observed differences in renal blood flow velocity and renal cortical tissue perfusion are related to the differences in composition of the 2 solutions.

Our results suggest that even modest infusions of crystalloid lead to fluid redistribution within the extravascular space, highlighting the problem of interstitial edema, in particular with 0.9% saline. Excessive interstitial edema is known to disturb the physiology of a number of organ systems, particularly the heart, lungs, and gastrointestinal tract.[9] It also has implications for viscera surrounded by nonexpansible capsules as interstitial edema may increase renal intracapsular pressure, reduce microvascular blood flow, and impair renal function.[34] Stone et al[34] demonstrated improved function for kidneys subjected to capsulotomy following massive preoperative volume replacement with blood and crystalloid in hypovolemic surgical patients. We have observed a trend toward greater renal volumes following 0.9% saline infusion, supporting the idea of greater interstitial edema with fluids of high sodium and chloride content, and in keeping with the observed expansion of extravascular fluid volume. Moreover, the data suggest that an increase in renal volume and intracapsular pressure together with the effect of chloride on reducing renal afferent arterial blood flow velocity combine to produce the observed decline in renal cortical tissue perfusion. Increase in renal intracapsular pressure may decrease the pressure gradient across the renal arterioles and, in keeping with Poiseuille's law, may have an additional impact on decreasing flow velocity. Although there was no statistically significant difference for changes in renal volume between the 2 infusions, this observation does not necessarily invalidate this hypothesis. It is possible that tissue edema could give rise to elevated intracapsular pressure in the absence of a significant change in renal volume due to the presence of the renal capsule. Thus, one would not expect there to be significant changes in renal volume over the short term or with the volumes infused in this study. Moreover, the presence of the renal capsule could mean that even small increases in volume might result in significant increases in pressure within the kidney and reduce perfusion.

There has been some debate about the clinical consequences of hyperchloremic acidosis, with some authors challenging whether it has an impact on clinical outcome.[1,35] Our study provides further evidence that the effects of hyperchloremic acidosis are not benign. This has implications for the use of 0.9% saline in the clinical setting, especially for patients with critical illness, where disorders of acid-base balance are common. The effects on renal blood flow velocity and renal tissue perfusion may be of particular significance for patients with renal disease or for those patients undergoing renal transplantation. In a comparison of 0.9% saline and Ringers' lactate,[10] patients undergoing renal transplantation who received the former had greater degrees of acidosis and reduced creatinine clearance compared with patients receiving the latter. However, in the present study, we found no difference in urinary concentrations of NGAL after infusion with either crystalloid, indicating absence of overt tubular damage after either crystalloid at the rate and volumes infused. It is possible that 0.9% saline may have resulted in decreased renal cortical tissue perfusion and, thus, a decrease in GFR without injuring the tubules.

Some limitations of this study should be clarified. First, the data were derived from healthy subjects and rely upon the

consistency of the interface between vascular and extravascular spaces to calculate changes in blood volume. This interface may be subject to a number of influences in the clinical setting, especially in patients undergoing surgery or those with sepsis. However, it may be inferred that the metabolic response to injury may lead to the exaggeration of our results. In addition, for patients with existing renal impairment or acidosis, a significant reduction in renal blood flow and renal tissue perfusion would clearly be unfavorable. Second, the present study has confirmed that 0.9% saline infusions are associated with a hyperchloremic acidosis and accompanied by a decrease in renal artery flow velocity and renal cortical tissue perfusion. However, a direct causal effect between hyperchloremic acidosis and the latter has not been shown conclusively. Nevertheless, we have provided a novel and plausible hypothesis for the finding. Third, by infusing a 2-L bolus of crystalloid in 1 hour, it could be argued that our observations reflect fluid therapy in the clinical setting of resuscitation. To produce the effect of hyperchloremic acidosis in healthy subjects, an exaggerated response was required, thus necessitating the rate and volume of infusion. Lastly, our calculations of blood volume and extravascular fluid volume are not directly measured parameters but have been derived from changes in weight and hematocrit. This methodology, however, is validated and consistent,[2,3,7] as well as more suitable than isotope or labeled red cell dilution techniques, which cannot be used for serial measurements over a short time period.

In conclusion, we have shown that the hyperchloremic acidosis associated with a 2-L infusion of 0.9% saline has a detrimental effect on renal artery blood flow velocity and renal cortical tissue perfusion. Balanced crystalloids may, therefore, be safer than 0.9% saline in patients with existing renal disease and those at risk of developing renal dysfunction.

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