Responses to low dose intravenous perindoprilat infusion in salt deplete/salt replete normotensive volunteers

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- 1 Intravenous ACE inhibitor therapy appears to have a role in the treatment of acute heart failure and early after myocardial infarction. Practical experience with intravenous administration with activation of renin is limited. We report responses to perindoprilat (Pt, 0.67 mg) or placebo (P) infused over 4 h in normotensive male volunteers ($n = 12$, 19–28 years, 53–77 kg) with double-blind, placebo controlled salt depletion (SD) or salt repletion (SR) as a model of the activated renin system.
- 2 Salt depletion caused no significant fall in serum sodium (P, 139.4 ± 2.4 ; Pt, 138.3 ± 1.9) compared with salt replete preparation (P, 139.9 ± 1.2 ; Pt, 139.7 ± 0.9) but elevation of plasma renin activity 2–3-fold. Pretreatment baseline systolic blood pressure following salt depletion (P, $121 \pm 9.3/71 \pm 7.9$; Pt, $121.5 \pm 9.6/69 \pm 8.1$) was higher than following salt replete preparation (P, $114 \pm 9.5/61 \pm 7.2$; Pt, $116.9 \pm 6.9/67 \pm 7.2$).
- 3 Baseline corrected supine SBP fell significantly and to a similar extent following active treatment regardless of activation of the renin system (SD, $-14.6 \pm 9.5/-9.4 \pm 6.4$; SR, $-12 \pm 14/-10.1 \pm 6.6$) compared with placebo (SD, $-6.1 \pm 6/-3.7 \pm 5.6$; SR, $-4.7 \pm 10/-1.3 \pm 6.5$). Heart rate was unchanged. Blood pressure response peaked at 4 h coincident with the end of infusion, peak drug concentration (SD, $15.8 \pm 2.5 \text{ ng ml}^{-1}$; SR, $16 \pm 2.8 \text{ ng ml}^{-1}$) and mean maximal plasma ACE inhibition (SD, $2.7 \pm 0.9 \text{ EU l}^{-1}$ SR, $2.7 \pm 0.7 \text{ EU l}^{-1}$; compared with P; SD, $25.5 \pm 6 \text{ EU l}^{-1}$; SR, $24.7 \pm 5.6 \text{ EU l}^{-1}$). BP recovered rapidly following discontinuation of infusion.
- 4 Controlled onset of ACE inhibition is feasible following low dose constant rate infusion of perindoprilat. Modest and predictable falls in blood pressure occur. This may have clinical application in heart failure where blood pressure control is important. The blood pressure response to low doses of ACEI, as used to initiate therapy in heart failure, may not depend solely on AngII withdrawal as activation of the RAS appears not to affect the response.

Keywords perindoprilat intravenous ACE inhibition blood pressure
renin angiotensin system

Introduction

The blood pressure response to ACE inhibitor treatment has been a matter of clinical concern due to isolated falls in blood pressure, in patients with RAS activation such as heart failure or diuretic treated hypertension [1, 2]. Interpretation origins of these episodes vary but most would suggest that there is a direct relationship [3, 4] to the acute withdrawal of

angiotensin II (AngII) controlling arterial [5] and venous [6] tone according to individual regional sensitivity [7].

More recent clinical studies with ACE inhibitors focus on the widening role of these drugs affecting cardiac and vascular remodelling following recent myocardial infarction [8, 9]. They are being used

earlier after myocardial infarction and their use is advocated not only in heart failure but more recently in asymptomatic left ventricular dysfunction. The benefits of this treatment have recently been confirmed [10–12] but the restrictive entry criteria of the initial studies has raised concern about the risk/benefit ratio in vulnerable or unstable patients [13]. Excessive blood pressure falls have been suggested to be markers of potential poor drug tolerance and may exclude individual patients from benefit.

Although the effective oral doses of these drugs can be derived from trial results, the relationship to mediators of response (e.g. reductions in AngII or potentiating kinins or prostanoids) is unclear. Low doses are appropriate for the initiation of therapy. There is no clearly defined rationale for initial dose selection [14] and there is some evidence to differentiate agents within the class at least at low doses of oral prodrugs [15].

Intravenous ACE inhibitor therapy is relevant to the treatment of acute heart failure [16] and possibly myocardial infarction [11]. Controlled practical experience with intravenous administration in states where there is activation of renin angiotensin systems (RAS) is limited. Given the emphasis on cautious introduction of these agents where there is activation of the renin angiotensin system [17] we have chosen to study a regimen of low dose infusion of the intravenous diacid ACE inhibitor perindoprilat on a background of dietary and diuretic based salt depletion. In normal subjects this procedure is safe [18–20] and is relevant to the response in diuretic treated patients with heart failure.

Methods

Twelve normotensive male volunteers (19–28 years; 53–77 kg) free of concomitant illness or drug therapy gave written and informed consent for their participation in the study. Normal health was confirmed by clinical history, physical examination, biochemical and haematological screen, 24 h urinary sodium excretion (100–250 mmol) and 12 lead electrocardiogram. The protocol was approved by the local Research and Ethical Review Committee.

Controlled manipulation of salt status was implemented using a variation of our previously described protocol [19, 20] based on dietary and oral diuretic treatment for 3 days prior to each of 4 study days conducted 14 days apart. During all phases volunteers were provided with a complete diet which contained a total of 40 mmol sodium per day (approx 60–80 mmol potassium). Dietary instruction by a qualified dietitian, and an accompanying diet sheet, defined allowed or disallowed supplemental foods as required. Free fluid intake was encouraged but alcohol prohibited during the preparatory phases or the study day (24 h). Subjects continued on the dietary regimen during the study day and until the return visit 24 h following drug administration.

During the 3 day pretreatment schedule where salt depletion was indicated by randomisation the subjects

received frusemide 40 mg (Antigen Pharmaceuticals), twice daily at 09.00 h and 12.00 h with placebo Slow Sodium (ten tablets daily taken two three times daily and four at night). Where salt repletion was indicated subjects received placebo frusemide twice daily and active salt repletion as ten Slow Sodium tablets daily (100 mm day⁻¹). The last dose of diuretic was therefore administered 20 h before drug administration. Following instructions on technique, 24 h urinary collections were performed at the screening visit, over the 24 h preceding each of the study days (day 3) and over the 24 h of each study day (day 4).

Volunteers attended the Research Unit after an overnight fast (10 h) on four separate occasions, 14 days apart, to receive perindoprilat (0.67 mg in 20 ml sterile saline infused at 5 ml h⁻¹ over 4 h) or placebo after salt depletion or salt repletion. Treatments were administered in a randomised, double-blind, cross-over design according to a pre-prepared schedule administered independently of the investigators by the Department of Pharmacy of the hospital.

Procedure

On attendance (07.00 h) subjects rested supine for at least 1 h following placement of two heparinised venous cannulae, one in each antecubital fossae for the purposes of blood sampling and infusion. Basal blood samples and blood pressure readings were collected supine prior to measurement of erect blood pressure and heart rate and bladder voiding. Blood pressure and heart rate were determined in triplicate using a semiautomatic device (Datascope Acutorr 3A, Paramus, New Jersey) at frequent intervals during the study day. Blood samples were drawn at intervals for the determination of serum drug concentration, serum electrolytes, plasma ACE activity and plasma renin activity. All subjects had been supine for at least 50 min prior to blood sampling for renin activity. Subjects received 200 ml of fluid at the start of infusion (0 h), a further 200 ml was given with a light breakfast (2 h), 400 ml with lunch (4 h) and 200 ml with an evening snack (9 h). All meals/food were given after the appropriate sampling was complete.

Analyses

Serum and urinary electrolytes were determined using a routine autoanalyser by the hospital biochemistry laboratory. Serum concentrations of perindoprilat and angiotensin converting enzyme activity were determined employing an enzyme substrate assay based on the technique of Tocco *et al.* [21]. Plasma renin activity was determined using the generation rate of AngI in the presence of phenylmethyl sulphonyl fluoride and EDTA as inhibitors [22].

Blood pressure and heart rate data were compared using repeated measures analysis of variance with Bonferroni correction adjusted for treatment comparisons between groups. Electrolyte data were compared using Friedman one way analysis of variance. Data are illustrated as mean $\pm$ 1 s.d.

Results

Effects of manipulation of renin angiotensin system through salt status

The salt depletion/repletion regimen was well-tolerated, no spontaneous symptoms were reported and all subjects completed the protocol without difficulty. The effects of pre-study preparation on serum and urinary electrolyte status is illustrated in Table 1.

There were no statistically significant changes in serum electrolytes before dosing on the morning of study comparing salt replete or salt deplete preparation. Sodium was unaltered but there was a trend towards lower potassium and chloride with higher bicarbonate, urea and creatinine comparing the salt replete preparation with the salt deplete state. As expected supine plasma renin was elevated 3–4-fold before treatment with placebo (7.2 ± 4.7 ng AI ml⁻¹ h⁻¹) or perindoprilat (12.0 ± 4.6 ng AI ml⁻¹ h⁻¹) by salt depletion compared with salt replete (placebo, 2.1 ± 1.3 ng AI ml⁻¹ h⁻¹; perindoprilat, 2.7 ± 2.4 ng ml⁻¹ h⁻¹).

In keeping with previous experience of attenuation of the diuretic effect of frusemide, urinary volume was not significantly affected on the pre-study day (day 3), comparing salt deplete with salt replete. Salt deplete preparation was associated with a significantly lower urinary sodium excretion on the pre-study day despite active frusemide therapy. Twenty-four hour natriuresis exceeded dietary intake (40 mmol) at this point. Salt depletion was associated with a non-significant trend towards a rise in urinary potassium excretion but urea and creatinine excretion were unaffected.

Blood pressure responses to low dose perindoprilat or placebo

Baseline supine blood pressure before treatment was higher after salt depletion than after salt repletion (SD: before placebo, $121 \pm 9.3/71 \pm 7.9$; before perindoprilat, $121.5 \pm 9.6/69 \pm 8.1$ for salt depletion,

Figure 1 Changes in supine mean arterial pressure and heart rate (mean $\pm$ 1 s.d.) following perindoprilat (0.67 mg over 4 h, squares) or placebo (circles) after controlled salt depletion (open) or salt repletion (closed), $n = 12$.

vs SR: before placebo, $114 \pm 9.5/61 \pm 7.2$; before perindoprilat, $116.9 \pm 9.9/67 \pm 7.2$ for salt repletion).

The baseline corrected profiles of supine mean arterial pressure and heart rate in response to treatment are illustrated in Figure 1. A similar profile was evident with erect blood pressures. Supine blood pressure showed a similar mean maximal fall in mean arterial pressure after placebo, salt replete (-4 mm Hg, 9 h). Baseline corrected supine MAP fell significantly and to a similar extent following perindoprilat regardless of salt deplete (-11.2 mm Hg, 4 h) or salt replete preparation (-11.2 mm Hg, 4 h). There was no significant difference between the minimum mean arterial pressure following perindoprilat in the salt deplete state or salt replete state (difference between the supine mean MAP 3.8 mm Hg; 95% CI -3.1 , $+10.8$ mm Hg, $P = 0.26$). Erect blood pressure showed a similar fall in mean maximal mean arterial pressure after placebo either salt replete (-8 mm Hg, 7 h) or salt deplete (-8.4 mm Hg, 8 h). Baseline corrected erect MAP fell significantly and again to a similar extent following perindoprilat regardless of salt deplete (-17.5 mm Hg, 4 h) or salt replete preparation (-17.4 mm Hg, 4 h). Analysing the difference between the minimum mean arterial pressure following perindoprilat in the salt deplete or salt replete state there was no evidence of an effect of salt depletion on the response (difference between the erect mean MAP 4.7 mm Hg; 95% CI -4.5 , $+12.6$ mm Hg, $P = 0.28$).

Mean maximal falls in supine pressures after perindoprilat in the salt deplete state ($-14.6 \pm 9.5/-9.4$

Table 1 Effect of salt deplete or salt replete preparation on pre-treatment serum electrolytes (day 4) and 24 h pre-study (day 3) urinary volume and electrolyte composition independent of subsequent treatment with perindoprilat or placebo

	Salt replete	Salt deplete
<i>Serum (Pre-dose, day 4)</i>		
Sodium (mmol l ⁻¹)	139.9 ± 1.2	139.4 ± 2.4
Potassium (mmol l ⁻¹)	4.2 ± 0.3	3.9 ± 0.2
Chloride (mmol l ⁻¹)	104.4 ± 2.4	99.4 ± 5.5
Bicarbonate (mmol l ⁻¹)	25.7 ± 2.9	27.8 ± 2.8
Urea (mmol l ⁻¹)	4.7 ± 1.2	5.3 ± 1.1
Creatinine (μ mol l ⁻¹)	89.6 ± 13	96.0 ± 18
<i>Urine (24 h, day 3)</i>		
Volume (l)	1.52 ± 0.61	1.54 ± 0.9
Sodium (mmol 24 h ⁻¹)	165 ± 83	115 ± 85
Potassium (mmol 24 h ⁻¹)	67.4 ± 34	75.9 ± 35
Urea (mmol 24 h ⁻¹)	332 ± 142	335 ± 156
Creatinine (mmol 24 h ⁻¹)	12.3 ± 4.1	12.1 ± 4.9

± 6.4) or salt replete ($-12 \pm 14/-10.1 \pm 6.6$) were greater compared with placebo (salt deplete, $-6.1 \pm 6/-3.7 \pm 5.6$; salt replete, $-4.7 \pm 10/-1.3 \pm 6.5$). In all instances heart rate was not affected by perindoprilat compared with placebo. Blood pressure fall after active therapy in all instances coincided with the end of drug infusion at 4 h.

Drug concentrations, ACE inhibition and renin activities

The plasma profiles of measured perindoprilat and ACE activities are illustrated in Figure 2. Peak drug concentrations (SD, 15.8 ± 2.5 ng ml $^{-1}$; SR, 16 ± 2.8 ng ml $^{-1}$) and maximal plasma ACE inhibition following perindoprilat (SD, 2.7 ± 0.9 EU l $^{-1}$; SR, 2.7 ± 0.7 EU l $^{-1}$) coincided with the end of infusion. The onset of plasma ACE inhibition was rapid compared with the unchanged values following placebo (salt deplete, 25.5 ± 6 EU l $^{-1}$; salt replete, 24.7 ± 5.6 EU l $^{-1}$).

The profile of plasma renin activity is illustrated in Figure 3. Placebo treatment caused little change in PRA from baseline values. Active treatment produced a maximal rise in activity coincident with the end of infusion. Both the baseline values and extent of the reactive rise in PRA following perindoprilat in the salt deplete state were considerably greater than those observed following salt replete preparation.

Discussion

The use of salt depletion as a model of an activated RAS in previous reports has involved preparation of subjects in an open fashion, usually involving variable and often severe and/or unpalatable restriction of dietary salt intake [23–26]. Such low salt diets are

Figure 2 Profiles of drug concentration and plasma ACE activity (mean $\pm$ 1 s.d.) following perindoprilat (0.67 mg over 4 h, squares) or placebo (circles) after controlled salt depletion (open) or salt repletion (closed), $n = 12$.

Figure 3 Profile of plasma renin activity (mean $\pm$ 1 s.d.) following perindoprilat (0.67 mg over 4 h, squares) or placebo (circles) after controlled salt depletion (open) or salt repletion (closed), $n = 12$.

difficult for unselected volunteers, are impractical in outpatient circumstances, being liable to non-compliance. Where controlled studies manipulating salt status are required for repeated study the nature of the diet becomes more important to individual volunteers. We have chosen moderate sodium restriction (40 mmol day $^{-1}$) over 4 day study periods. In order to enhance activation of RAS, frusemide (40 mg twice daily) was used on each of 3 pre-study days. The pharmacodynamic response to frusemide has been extensively characterised [27, 28] and the twice daily regimen employed in our protocol [29] would generate an initial natriuresis, attenuated on repeated dosage [30–32]. To prevent overt fluid depletion a high fluid intake was specifically encouraged during the pre-study period and fluid intake on the study day carefully controlled. This is a simple and reproducible test bed to assess the response to drugs blocking the renin system in normal subjects. We have used salt depletion alone to define responses to renin inhibitors [18] or ester prodrug ACE inhibitors [33] and angiotensin AT1 receptor antagonists [19]. In the present study we have used double-blind activation of the RAS through salt depletion or salt replete normals for controlled single dose studies. Renin activation is comparable and relevant to that associated with chronic diuretic therapy or disease states [34].

There has been widespread concern over the so called 'first dose hypotensive response' to ACE inhibitors. The origin of the blood pressure fall is unclear [35]. Surprisingly given the widespread use of these drugs, studies investigating the determinants of individual response are not available. Using a model of RAS activation such as that described here it is possible to address the contribution of a particular drug, the dosage selected and the level of activation of the RAS on response. The baseline renin in this study is comparable with those seen in renovascular hypertension [36] or heart failure [37].

We have previously studied low dose constant rate infusion of intravenous diacid ACE inhibitors in

patients with stable chronic cardiac failure [38] and in normal subjects [39], and have examined pharmacokinetics for the potential to model the tissue distribution and response of these drugs in man [39–41]. Conventional wisdom has assumed that response to ACE inhibitor drugs results directly from AngII withdrawal [3].

In this study we saw no difference in the extent or pattern of blood pressure fall following ACE inhibition with or without activation of the RAS. It may be that during low dose initiation of therapy given that ACE is non-specific and known to have a higher affinity for alternative substrates such as kinins than AngI, that other aspects of ACE inhibitor pharmacol-

ogy other than AngII withdrawal are responsible for the reduction in blood pressure. Recent attention is concentrated on the role of bradykinin in mediating responses to ACEI. Controlled onset of ACE inhibition and predictable blood pressure responses is possible using low dose constant rate infusion.

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