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The influence of age on renal and extrarenal effects of frusemide

F. ANDREASEN[1], U. HANSEN[2], S. E. HUSTED[2], C. E. MOGENSEN[3] & E. B. PEDERSEN[4]
[1]Division of Clinical Pharmacology, Institute of Pharmacology, University of Aarhus, DK-8000 Aarhus C, [2]Medical Department I, Aarhus Amtssygehus, DK-8000 Aarhus C, [3]Medical Department M, Aarhus Kommunehospital, DK-8000 Aarhus C, [4]Medical Department C, Aarhus Kommunehospital, DK-8000 Aarhus C, Denmark

1 The effect of frusemide 80 mg i.v. was compared during 24 h in 10 young and eight elderly healthy male volunteers following a 24 h control period in the ward.

2 During the 30 min following the injection the increments in excretion of urine, sodium, potassium and frusemide were significantly smaller in the elderly.

3 The 24 h increase in sodium excretion was significantly larger in the elderly.

4 The endogenous 24 h creatinine clearance was reduced by 12% ($P < 0.01$) in both age groups.

5 The frusemide induced changes in the 8 h serum concentration curves for albumin differed significantly between the two groups (analysis of variance, $P < 0.01$). The drug induced increase in albumin concentration became significant in the young 5 min after the injection. In the elderly it took more than 15 min before the increase in serum albumin reached significance. The average maximal increase in albumin concentration was 14.3% in the young and 9.7% in the elderly ($P < 0.05$).

6 No difference was seen between the two age groups in the significant frusemide induced increases in the 24 h albumin excretion but in the elderly a significantly larger decrease in the 24 h excretion of β_2-microglobulin was observed ($P < 0.05$).

7 No significant age difference was observed in the initial significant increases in diastolic blood pressure observed in both age groups or between the later changes in systolic blood pressure which was significantly reduced in the young only.

8 The slower haemoconcentration response in the elderly seemed associated with the slower secretion rate of frusemide to the tubular lumen. We found no evidence of an age related difference in tubular cell response to frusemide. It is emphasized that a maximal initial frusemide response in the elderly, in contrast to what was found in the young, probably was not achieved by the 80 mg i.v.

Keywords frusemide age haemodynamics protein excretion sodium potassium

Introduction

Although the site of action of frusemide is on the tubular cells of the kidney the aim of the intravenous administration of the drug usually is the achievement of an immediate effect on the cardiovascular system.

Patients receiving frusemide intravenously frequently are elderly (Andreasen & Mikkelsen, 1977; Perez *et al.*, 1980) but most studies on the human pharmacology of the drug have been performed in young, healthy volunteers (Cutler & Blair, 1979; Benet, 1979). Therefore it is difficult for the attending physician to distinguish between possible disease-related and anticipated age-related deviations in the clinical pharmaco-

logy of the drug. Our recent demonstration of age related changes in the pharmacokinetics of frusemide (Andreasen *et al.*, 1983) suggests that also the pharmacodynamics of the drug could be influenced by advancing age.

The purpose of the present investigation was to study the influence of age on renal and extrarenal effects of intravenous frusemide. In particular we wanted to know whether advancing age influenced the possibility of achieving an immediate effect on the cardiovascular system.

Methods

A mutual protocol for the entire study secured that the young and elderly subjects were treated in exactly the same way. Because the group of young volunteers was so much easier to find the results from that group were compiled at first. A study of the individual variation in response inside that group was published (Andreasen *et al.*, 1982) and that publication included a detailed description of the protocol. Therefore only a brief account is given here.

Subjects Eight male volunteers in the age group 60–70 years were compared with 10 male volunteers in the age group 20–35 years. All gave consent to the study after being carefully informed of the procedures involved. None had any history or other evidence of disease from the heart, lungs, liver or kidneys and none received permanent drug therapy. The average body weight of the young was 71 kg with a range from 57 to 83 kg and for the elderly it was 76 kg (64–83 kg). The average of sixteen measurements of the systolic blood pressure (BP) during the 24 h control period showed a mean of 123 mm Hg in the young (112–136) and 126 in the elderly (108–161). The corresponding figures for the diastolic BP were 78 mm Hg for the young (66–89) and 80 mm Hg for the elderly (66–95).

Procedure The subjects stayed in the hospital from 08.00 h until 09.00 2 days later. The first 24 h period (09.00–09.00 h) was a control period. A standard hospital diet was given (sodium content 150–200 mmol), soft drinks as desired but no alcoholic beverages were allowed.

Between 09.00 h and 09.02 h on the second morning 80 mg frusemide was injected i.v. From 09.07 h on both mornings blood was collected at short intervals at first and longer intervals later on. Urine was collected in fractions of 30 min periods at first and longer intervals after 1 h.

The BP was measured by standard sphygmomanometer technique by the same investigator throughout for each subject and phase V of the Korotkoff sounds was used.

Analytical methods

The concentration of frusemide in blood and urine was determined by h.p.l.c. (Andreasen *et al.*, 1981). Standard laboratory methods were used for measurement of sodium, potassium, creatinine and for albumin in serum. Albumin and β_2-microglobulin (Phadebas®, β_2-microtest) in urine were determined by radioimmunoassay (Evrin *et al.*, 1971; Miles *et al.*, 1970). During the study the radioimmunoassay of arginine vasopressin (AVP) in plasma (Robertson *et al.*, 1973) became available here. Therefore initial changes in the serum concentration of that hormone were followed in the elderly only.

Calculations The filtered load of Na^+ and K^+ is determined by the product of the glomerular filtration rate (GFR), the serum concentration of the ion in question [ion] and the time period studied. The concentration of creatinine was determined in each sample of serum and urine and by using creatinine clearance (CL_{cr}) as a measure of GFR (Bennett & Porter, 1971) we estimated the filtered load of sodium as

$$F_L = CL_{cr} \times [Na^+] \times \text{time period} \quad (1)$$

The reabsorbed load (R_L) of sodium could then be estimated if the excreted load (E_L) was known as

$$R_L = F_L - E_L \quad (2)$$

The statistical significance of differences was assessed by the Wilcoxon test for paired comparisons or the Mann–Whitney test. Correlations were studied by linear regression analysis. *P*-values below 0.05 were considered significant. When differences observed at multiple sampling times were to be assessed we used analysis of variance for repeated measurements design.

Results

Figure 1 illustrates that the very fast diuretic response in the young was maximal during the first half hour after the drug administration. In the elderly the average maximal response was reached during the second half hour. The 24 h increase in diuresis caused by frusemide was 1154 ml (75% more than the volume produced during the control period) in the elderly and its was 942 ml (48% more) in the young (NS). In Figure 2 the individual increases in diuresis during the initial 30 min are related to the excretion rate of

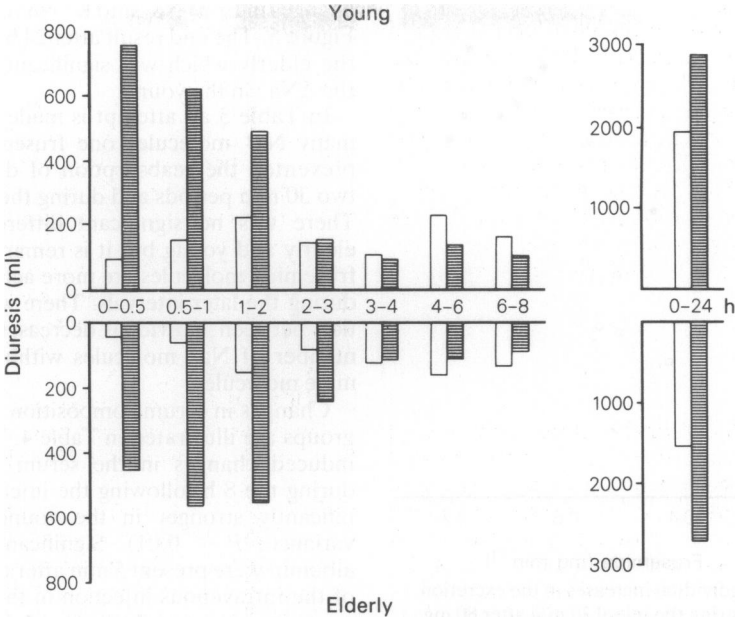

Figure 1 Average diuretic responses to 80 mg frusemide injected intravenously at 0 h to 10 young (above) and eight elderly (below) healthy male volunteers (▤). The diuresis during the control period is also shown (□).

frusemide. Significantly lower ($P < 0.01$) increases in diuresis and in frusemide excretion rate were seen for the elderly but the individual diuretic responses in the elderly were related to the corresponding frusemide excretion rates ($r = 0.7805$, $P < 0.05$). There was no such relationship for the young. Figure 3 shows similar relationships between the initial excretion rates of frusemide and sodium. For the elderly the linear regression coefficient was 0.7814 ($P < 0.05$). The position of the individual data points does not suggest that higher excretion rates of frusemide in young subjects would have been followed by higher excretion rates of urine or sodium.

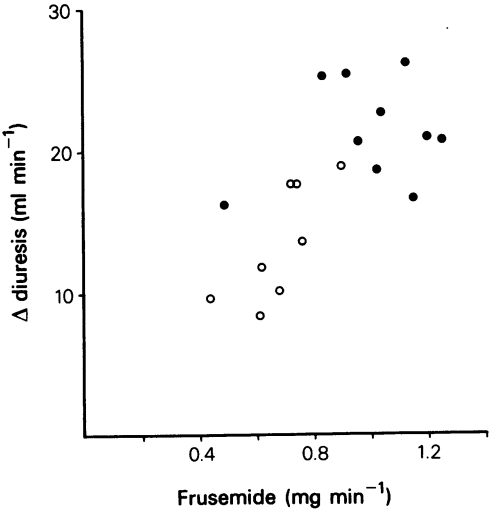

Figure 2 The individual increases in diuresis during the initial 30 min after 80 mg frusemide i.v. to 10 young (●) and eight elderly (○) healthy, male volunteers. The values are shown in relation to the excretion rates of frusemide.

There was no connection between free or total frusemide concentrations in serum and the diuretic or saluretic effect. The average serum concentration during the first half hour was 7.3 μg/ml in the young while it was 8.4 μg/ml in the elderly. The percentage of unbound drug was 1.3–1.4 in both age groups (Andreasen *et al.*, 1983).

Figure 4 illustrates the considerable variation found in the estimated frusemide induced changes in GFR (CL_{cr}) in the elderly as well as in the young subjects. For the elderly an apparent tendency to an increased GFR initially was followed by an apparent fall which became significant during the period 60–120 min. During the period from 30–60 min the frusemide induced depression in GFR was stronger in the young than in the elderly ($P < 0.01$). From 2 h after the injection of frusemide the GFR was approaching the value from the control day in both age groups. Table 1 shows that the endogenous 24 h

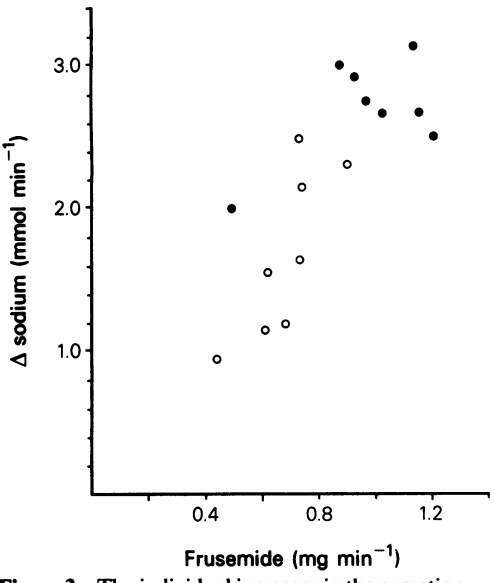

Figure 3 The individual increases in the excretion rate of sodium during the initial 30 min after 80 mg frusemide i.v. to eight young (●) and to eight elderly (○) healthy, male volunteers. The values are shown in relation to the excretion rates of frusemide.

CL_{cr} was equally reduced in both age groups (12% ($P < 0.01$)). The frusemide induced depressions in R_L of sodium were significant in both age groups during the first 2 h (Table 2). The depressions were strongest in the young and the difference in response between the groups was significant during the maximal period of R_L depression from 30 to 60 min after the injection of the drug. The frusemide induced cumulative changes in E_L of Na^+ and K^+ over 24 h are seen in Figure 5. The end result after 24 h was a ΔNa^+ in the elderly which was significantly higher than the ΔNa^+ in the young.

In Table 3 an attempt is made to assess how many Na^+ molecules one frusemide molecule prevented the reabsorption of during the first two 30 min periods and during the second hour. There was no significant difference between elderly and young but it is remarkable that the frusemide molecules are more active ($P < 0.01$) during the later intervals. There was no connection between individual decreases in GFR and number of Na^+ molecules withheld per frusemide molecule.

Changes in serum composition in the two age groups are illustrated in Table 4. The frusemide induced changes in the serum albumin level during the 8 h following the injection were significantly stronger in the young (analysis of variance, $P < 0.01$). Significant increases in albumin were present 5 min after the completion of the intravenous injection in the young and a maximum was reached after 2 h. The elderly showed no significant increase at 15 min but after 1 and 2 h significant increases were seen. At all times the increases were significantly lower in the elderly than in the young. The average of the maximal percentage increases in serum albumin was 14.3 ± 4.0% in the young while it was 9.7 ± 4.1% in the elderly (young *vs* elderly, $P < 0.05$). The decreases in serum sodium were significant in both groups after 2 h whereas the potassium decreases which reached significance after 1 h in the young never reached significance in the elderly. The influence of frusemide on the

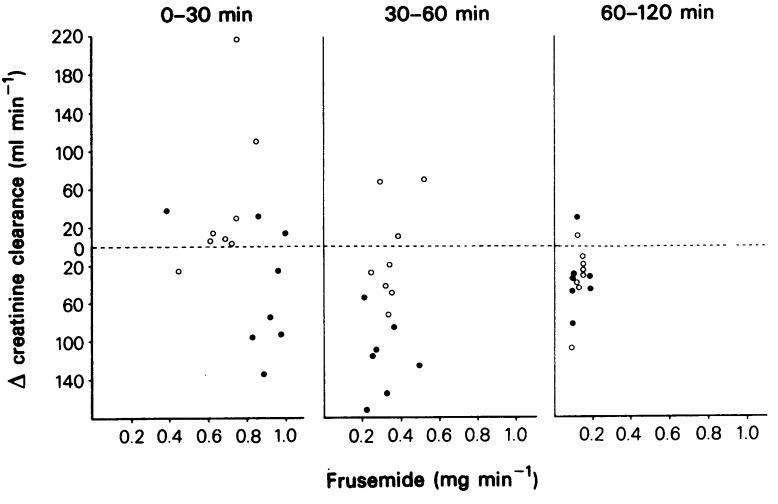

Figure 4 Individual changes in relation to the day of control, in CL_{cr} after i.v. injection of 80 mg frusemide from 0–2 min to eight young (●) and to eight elderly (○) healthy male volunteers. The values during 0–30, 30–60 and 60–120 min are shown in relation to the individual excretion rates of frusemide.

Table 1 The endogenous 24 h creatinine clearance (ml/min) determined during the 24 h control period and during the 24 h period following the intravenous administration of 80 mg frusemide i.v. over 2 min

	Young men (20–35 years)		*Elderly men (60–70 years)*	
Subject	*Control period*	*Drug period*	*Control period*	*Drug period*
1	118	103	93	76
2	144	123	84	75
3	151	127	104	80
4	138	119	103	89
5	132	112	99	80
6	—	—	81	84
7	124	122	99	85
8	—	—	98	92
9	129	136	—	—
10	142	106	—	—
Mean ± s.d.	125 ± 11	119 ± 11	95 ± 9	83 ± 6

average concentration of AVP in the groups of elderly subjects can be assessed in Table 5. No significant changes were found but there was a trend towards higher levels after frusemide. The mean of individual maximal increases rose from 2.4 to 3.4 pg/ml during the control period (NS) and from 3.4 to 8.1 pg/ml during the frusemide period ($P < 0.05$).

Haemodynamic responses are illustrated in Figure 6 which shows an immediate and significant rise in diastolic BP in the young. In the elderly the average increase in diastolic blood pressure (DBP) occurred more slowly and none of the average increases shown on the figure reached significance. The individual average increases observed between either 20 or 30 min and 1 h were significant ($P < 0.01$). The systolic blood pressure (SBP) did not change significantly in the elderly whereas it fell significantly in the young after 2–3 h. In the young an initial significant fall in heart rate (HR) was followed by a gradual insignificant increase. In the elderly there was no initial fall and from 1 h HR was increased significantly. None of the graphs in Figure 6 reflected a significant difference between the age groups and none were connected with serum frusemide concentrations in young or elderly.

The renal excretion of albumin and of β_2-microglobulin were affected by i.v. frusemide. Table 6 shows that a significant increase in the 24 h excretion of albumin was observed in both age groups. The excretion of β_2-microglobulin was significantly decreased in the elderly. This decrease differed significantly ($P < 0.05$) from the change seen in the young.

Table 2 The influence of age on frusemide induced changes in the reabsorbed fraction of sodium. Day 1 was a control period. At 0 min 80 mg of frusemide was given intravenously on day 2

		0–30 min		*30–60 min*		*60–120 min*	
		Day 1	*Day 2*	*Day 1*	*Day 2*	*Day 1*	*Day 2*
R_L (per cent of filtered load absorbed)	Young $n = 8$	99.1 ± 0.3	80.9 ± 8.8	98.7 ± 0.3	42.2 ±14.6	98.4 ± 0.8	92.2 ± 2.5
	Elderly $n = 8$	98.9 ± 0.8	90.9 ± 3.9	98.6 ± 0.9	81.9 ± 8.2	98.7 ± 0.7	89.5 ± 6.6
Frusemide induced decreases in R_L (range)	Young	6.4–30.8		24–73		2.5–10.0	
	Elderly	3.6–13.6		4–29		3.1–15.0	
Significance of difference between age groups		NS		$P < 0.01$		NS	

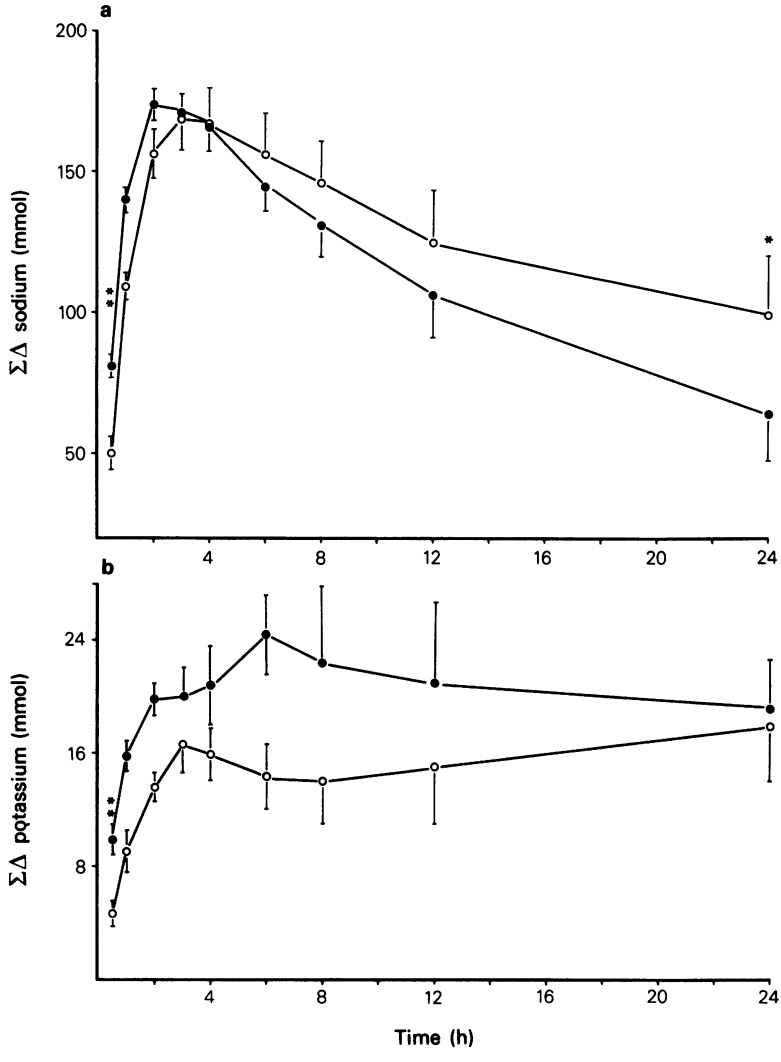

Figure 5 Average changes in the cumulative excretion of Na^+ (upper panel) and K^+ (lower panel) in the urine during 24 h after the injection of 80 mg frusemide to eight elderly (●) and eight young (○) healthy, male volunteers. The changes were calculated for each individual by comparison with a 24 h control period (Mean ± s.e. mean). Significance of differences between the two age groups are indicated, * $P < 0.05$, ** $P < 0.01$.

Discussion

A very early response to i.v. frusemide was a fall in the concentration of water in the circulating blood. Advancing age significantly delayed the onset of that response and the magnitude of the individual maximal responses was reduced in our elderly, healthy male volunteers. The reduction in circulating blood volume and the haemoconcentration were accompanied by compensatory changes in haemodynamics and in the 24 h excretion pattern of electrolytes.

Dose, concentration and effect

The intravenous administration of 80 mg frusemide over 2 min to elderly and young male volunteers did not produce the same inhibitory effect on the target process: The reabsorption of electrolytes over the tubular cell membrane. During the 30 min from the initiation of the drug injection significantly less sodium was reabsorbed in the young who also excreted significantly more potassium. That difference could not be explained by differences in the serum

Table 3 The ratio between frusemide induced increases in the urinary excretion rate of Na^+ and the excretion rate of frusemide (mol Na^+/mol frusemide) during the first 2 h after the administration of 80 mg frusemide to young and elderly healthy male volunteers (mean ± s.d.)

	0–30 min	*30–60 min*	*60–120 min*
Young $n = 8$	930 ± 230	2270 ± 670**	1800 ± 700*
Elderly $n = 8$	830 ± 200	1900 ± 470**	1870 ± 830*

Significance of difference within the group between initial and later collecting periods, * $P < 0.05$, ** $P < 0.01$.
No significant differences between young and elderly.

concentrations of frusemide which were significantly higher in the elderly. The variation in the individual responses in the elderly could in part ($r = 0.78$) be explained by differences in the rate of frusemide excretion in the urine. The course of the concentration effect curve in the elderly may indicate that stronger responses could have been achieved by higher doses or at least by higher frusemide excretion rates. A similar relationship was not present in the young who excreted frusemide significantly faster. The reason for this lack of connection in the young probably

Table 4 Average changes, in comparison with the preinjection values, in the serum concentration of albumin, sodium and potassium after the i.v. injection of 80 mg frusemide at 09.00 h (time 0) to young ($n = 10$) and elderly ($n = 8$) healthy, male volunteers (Day 2). For comparison the corresponding values from the preceding control day (Day 1) are shown.

			Time (min)						
		Prevalue (average of −10 and −5)	*7*	*15*	*60*	*120*	*240*	*360*	*480*
			Change in concentration						
Albumin (mmol l^{-1})									
Young	Day 1	709 ± 24	+13	0	− 1	+11	+11	+ 8	+10
	Day 2	711 ± 23	+21**	+31**	+86**	+99**	+70**	+58**	+60**
Elderly	Day 1	597 ± 40	− 6	− 1	+11	+15	− 1	+ 8	+26
	Day 2	730 ± 38	−12	+ 2	+41**	+48**	+15	+17	+20
Significance of differences in response between groups			$P < 0.01$	$P < 0.02$	$P < 0.01$	$P < 0.01$	$P < 0.01$	$P = 0.05$	$P = 0.05$
Sodium (mmol l^{-1})									
Young	Day 1	142 ± 1.3	0	− 1	− 1	− 1	0	− 1	− 1
	Day 2	142 ± 1.6	0	0	− 1	− 2*	− 1	− 2	− 2
Elderly	Day 1	142 ± 4.0	− 3	− 1	0	− 2	− 2	− 1	− 1
	Day 2	142 ± 1.5	0	0	0	− 2**	− 2*	− 3	− 3
Significance of differences in response between groups			NS	NS	NS	NS	NS	NS	NS
Potassium (mmol l^{-1})									
Young	Day 1	3.9 ± 0.12	0	0	+ 2	+ 1	− 1	0	0
	Day 2	4.0 ± 0.20	− 1	− 2	− 2*	− 2*	− 1	+ 1	− 1
Elderly	Day 1	4.1 ± 0.16	0	+ 1	− 1	− 1	− 1	0	− 1
	Day 2	4.1 ± 0.34	− 1	0	− 3	− 2	− 1	+ 1	− 1
Significance of differences in response between groups			NS	NS	NS	NS	NS	NS	NS

Significance of change, in comparison with prevalue, within the group
* $P < 0.05$, ** $P < 0.01$

Table 5 The influence of 80 mg frusemide i.v. on the average serum concentration of arginine vasopressin (AVP) (pg/ml) in elderly healthy male volunteers. At 09.00 h on day 2 frusemide was injected i.v. over 2 min.

	Time (h)			
	08.50	*09.07*	*09.15*	*10.00*
Day 1 ($n = 6$)	2.4 ± 1.2	2.8 ± 2.0	1.7 ± 0.8	3.1 ± 1.5
Day 2 ($n = 8$)	3.4 ± 1.9	3.1 ± 1.9	4.0 ± 3.6	5.7 ± 4.6

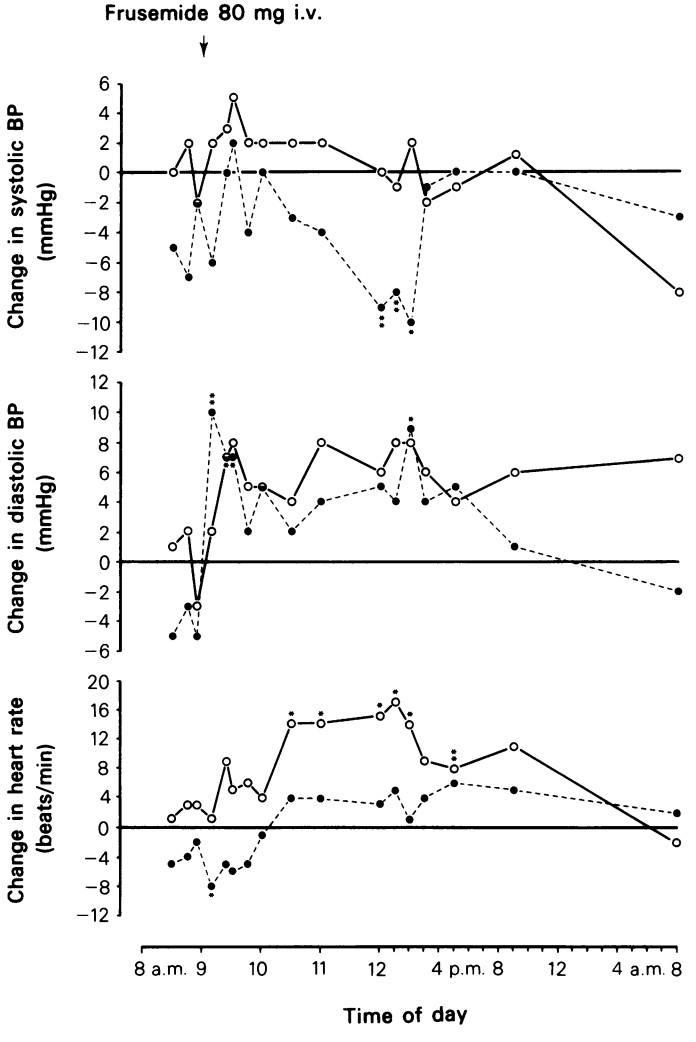

Figure 6 Average frusemide induced changes in haemodynamics in young (-●-) and elderly (-○-) healthy, male volunteers. The changes in blood pressure and heart rate over 24 h were calculated individually by comparison with the corresponding values during the 24 h control period. Significance of illustrated changes within the groups, * $P < 0.05$, ** $P < 0.01$. There were no significant differences between the responses in young and elderly.

Table 6 The influence of age on frusemide induced changes in urinary excretion of albumin and of β_2-microglobulin (mean ± s.d.). Day 1 was the control day and day 2 was the 24 h period following the i.v. injection of 80 mg frusemide. The subjects were healthy male volunteers.

		Albumin		*β_2-microglobulin*	
		Day 1	*Day 2*	*Day 1*	*Day 2*
Urinary excretion (mg/24 h)	Young ($n = 9$)	8.1 ±4.2	12.8** ± 8.2	0.09 ±0.02	0.08* ±0.03
	Elderly ($n = 8$)	8.0 ±4.2	13.8** ±10.6	0.17 ±0.14	0.08* ±0.09
Range of frusemide induced changes in excretion (mg)	Young	−0.8 to +27		−0.03 to +0.04	
	Elderly	−1.9 to +18.3		−0.19 to −0.02	
Significance of differences in frusemide induced changes between young and elderly		NS		$P < 0.05$	

Significance of differences from day 1 to day 2 within the group.
* $P < 0.05$, ** $P < 0.01$

was that the maximal diuretic response was reached in some or all of the young subjects. A decrease in GFR, as it occurred here probably as a consequence of an excessively high diuresis, gave a lower number of electrolyte ions which frusemide could inhibit the reabsorption of, but simultaneously more time was available for the tubular secreted frusemide molecules to act in the preurine. For both age groups the observed effect of the decrease in GFR was that the reabsorption of more than twice as many sodium molecules was impeded by each frusemide molecule. The number of sodium molecules retained in the urine by each frusemide molecule did not differ between the two age groups and thus we found no indication of an age related difference in tubular response to frusemide.

Relationship between renal and extrarenal effects

All the extrarenal effects observed by the methods utilized here could be explained as secondary to inhibition of electrolyte reabsorption over the tubular cells. The larger volumes of water excreted initially by the young seemed to explain the significant age differences in the developing rate and magnitude of the haemoconcentration. No significant differences were observed between the age groups in the haemodynamic responses. Initially both age groups reacted with significant increases in DBP but during the next few hours the elderly reacted with a prolonged period with significant increases in HR while the conspicuous findings in the young were significant falls in SBP. Renin and angiotensin may have a role in this development and activation of the sympathetic nervous system also could be important. Plasma renin and plasma aldosterone levels are lower in elderly subjects (Pedersen, 1979). Intravenous frusemide activates the renin-aldosterone system (Hesse *et al.*, 1975) and a stronger stimulation of the secretion of these hormones in the young could explain the differences in the cation excretion pattern over the entire 24 h period in young and old subjects. The young conserved their sodium significantly better but apparently they lost a little more potassium than the elderly. In 10 healthy volunteers with a mean age of 31 years De Lima *et al.* (1981) found a significant increase of 1.5 pg/ml per hour after 20 mg frusemide i.v. The frusemide induced increases in the AVP concentration in plasma found in our elderly subjects by a four times higher dose were bigger. Dog experiments have indicated that vasopressin may be released *in vivo* in sufficient amounts to induce a vasoconstriction (Szczepanska-Sadowska, 1973; Cowley *et al.*, 1974). The present study does not permit any conclusions about a possible role of AVP in the observed haemodynamic responses, but an AVP induced rise in DBP remains a possibility.

Clinical implications The maximal achievable effect of intravenous frusemide apparently was reached when 80 mg were injected to young subjects with normal RPF (and normal tubular secretion capacity) and GFR. The results from our elderly male, healthy volunteers indicated that the maximal effect was not obtained by that dose in subjects with reduced tubular secretion capacity. This is of particular interest in elderly subjects with heart failure who may have filtra-

tion fractions increased from the normal 0.16–0.20 to 0.40–0.50. Considerably higher doses may be necessary therefore to achieve frusemide concentrations in the tubular fluid which in the individual patients are associated with the achievement of the strongest and most immediate effect on circulating blood volume.

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