The influence of naloxone on the circulatory effects of captopril

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1 The fall in blood pressure produced by captopril is not accompanied by a compensatory tachycardia. Angiotensin converting enzyme may participate in the metabolism of endogenous opioids and these substances can lower blood pressure without reflex tachycardia. It is therefore possible that the hypotensive effect of captopril involves an action on endogenous opioid metabolism.

2 The influence of naloxone, an opiate receptor antagonist, on the cardiovascular effects of captopril has been studied in nine normal volunteers.

3 Mean blood pressure following a 70° head up tilt 1.5 h after captopril was significantly lower than following placebo, but the blood pressure following the combination of captopril and naloxone differed from neither the placebo nor the captopril values. Blood pressures (mm Hg) were: placebo 97.7 ± 5.9 ; captopril 83.3 ± 14.2 ; captopril + naloxone 89.6 ± 11.8 .

4 Heart rate (beats/min) following a 70° head up tilt at 1.5 h was the same on all three treatments: placebo 83.4 ± 21.1 ; captopril 83.3 ± 16.5 ; captopril + naloxone 83.6 ± 11.8 .

5 Maximum systolic blood pressure achieved during isometric exercise following captopril + naloxone was intermediate between, and did not differ significantly from, pressures obtained on the other two treatment days.

6 We conclude that co-administration of naloxone may attenuate the effect of captopril on blood pressure and that endogenous opioids could be involved in the actions of this drug.

Keywords naloxone captopril blood pressure

Introduction

The rationale for the early investigation and subsequent clinical use of captopril was its action as a competitive inhibitor of the enzyme which converts angiotensin I to angiotensin II. While this action may be sufficient explanation for the antihypertensive efficacy of captopril in some subjects (particularly patients with renovascular hypertension, those with high plasma renin

activity and those who are salt depleted) it does not provide an explanation for some other effects of the drug. For example, in normal subjects and in patients with hypertension the fall in blood pressure produced by captopril is not accompanied by an increase in heart rate (Cody *et al.*, 1979; Millar *et al.*, 1981; Shepherd *et al.*, 1982). Several explanations have been offered including a selective effect on venous tone (Cody *et al.*, 1979), or a sympathoinhibitory action via removal of facilitatory presynaptic angiotensin II effects (Hatton *et al.*, 1982). Another possible mechanism is that reduced

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formation of angiotensin II will increase vagal tone by release of the parasympathetic nervous system from inhibition by angiotensin II (Sturani *et al.*, 1982). In the present study we have investigated a further possibility, namely that captopril reduces the metabolic breakdown of endogenous opioids. Opioid receptor agonists have been shown to lower blood pressure without producing a compensatory tachycardia both in animals and man (Petty & Reid, 1982; Rubin *et al.*, 1983). Captopril inhibits carboxypeptidase, one of the enzymes responsible for the inactivation of enkephalins *in vitro* (Arregui *et al.*, 1979) and, more specifically, has also been shown to reduce the metabolism and increase the analgesic potency of metenkephalin (Stine *et al.*, 1979).

If captopril were to act partially through an effect on endogenous opioids then one would expect that an opiate receptor antagonist might unmask a tachycardia in response to the hypotensive action of captopril. We have tested this hypothesis by co-administering naloxone and captopril.

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Methods

The study protocol was approved by the Research and Ethical Committee of the Glasgow Northern District and all subjects gave written consent. The study involved nine males aged between 19 and 36 years. All were in good health and were non smokers. None was taking any concurrent prescription or non prescription drugs. Each subject was studied on three occasions, separated by at least a week, when they received naloxone plus captopril, naloxone vehicle plus captopril or naloxone vehicle plus captopril placebo. The order of drug administration was determined by randomised Latin Square design and the study was double blind. Naloxone (Sterling Winthrop) was given in the dose of 0.2 mg/kg over 30 min into an antecubital vein. Captopril (Squibb UK Ltd) was given in a dose of 50 mg orally. The captopril (or placebo) was given 1.5 h following the beginning of the naloxone or inactive vehicle infusion. This timing was determined by our previous observation that naloxone has its maximum effect on blood pressure fall during sleep 3–4 h following administration (Rubin *et al.*, 1981) and also significantly increases baroreflex sensitivity during sodium nitroprusside infusion at this same time (Rubin *et al.*, 1983). Since captopril

has its maximum effect on blood pressure 1.5 h following administration, we concentrated data collection immediately before and 1.5 h following administration of captopril or placebo. An additional set of observations was made 6 h after captopril administration at which time reversal of its effects was anticipated. Blood pressure was measured both supine and following a 5 min 70° head up tilt using a semi automated sphygmomanometer (Bosomat 2, Bosch & Sohn, West Germany) before and at 1.5 and 6 h following administration of captopril or placebo. Heart rate was recorded concurrently from precordial ECG leads. Further measurements of blood pressure and heart rate were taken (after recovery from tilt) during forearm isometric exercise as described below. Blood samples were taken via an indwelling venous cannula for analysis of plasma noradrenaline concentration and plasma renin and angiotensin converting enzyme activity.

Noradrenaline was analysed by a single isotope radioenzymatic assay (Henry *et al.*, 1975) and plasma renin activity was measured by the method of Skinner (1967). Angiotensin converting enzyme (ACE) activity was measured by the method of Cushman & Cheung (1970) as modified by Chiknas (1979). The angiotensin converting enzyme activity assay was performed 'on line' within 2–4 h of sample collection in order to prevent the reversal of inhibition of converting enzyme by captopril which is known to occur on sample storage (Roulston & MacGregor, 1980). Results of ACE activity are expressed as units per ml where one unit is the quantity of enzyme generating 1 nmol hippuric acid per minute under assay conditions.

Isometric exercise

Maximum voluntary forearm contraction (MVC) was determined before the study. Forearm exercise consisted of 3 min handgrip at 30% of MVC. Readings of blood pressure were taken using the contralateral arm at 0, 1, 2 and 3 min, and heart rate was recorded continuously.

Data analysis

Data are presented as mean $\pm$ s.d. Blood pressure is expressed as mean values, calculated as diastolic + 1/3 pulse pressure. Blood pressure and heart rate data in the supine position and following tilt were analysed by Latin Square analysis and all other data were compared by Friedman two way analysis of variance.

Results

Three subjects felt light headed during tilt at the 1.5 h recording following captopril alone but no subjects experienced any symptoms either on the placebo day or when captopril was given with naloxone. In the supine position there was no difference in blood pressure between the three treatment groups at 1.5 h (Table 1a). The change in blood pressure following a 5 min 70° head up tilt is shown in Table 1b. Captopril alone significantly lowered blood pressure but the combination of captopril and naloxone resulted in a

Table 1a Supine mean blood pressure (mm Hg) following placebo captopril alone or captopril plus naloxone

Time (h)	Placebo	Captopril	Captopril + naloxone
0	90.2 ± 7.5	90.4 ± 8.1	84.8 ± 5.8
1.5	87.7 ± 7.8	83.2 ± 7.0	85.9 ± 7.7
6	86.8 ± 5.6	84.2 ± 4.5	80.6 ± 5.8

The 95% confidence interval for difference between treatment means at the 1.5 h time was 4.9.

b Mean blood pressure (mm Hg) following a 5 min 70° head up tilt in subjects receiving placebo, captopril alone or captopril plus naloxone

Time (h)	Placebo	Captopril	Captopril + naloxone
0	93.8 ± 7.1	94.8 ± 5.7	93.2 ± 5.2
1.5	97.7 ± 5.9	83.3 ± 14.2	89.6 ± 5.8
6	95.2 ± 6.2	89.1 ± 7.5	89.8 ± 7.3

The 95% confidence interval for difference between treatment means at the 1.5 h time was 12.6.

smaller fall in blood pressure which was intermediate between captopril alone and placebo and was not significantly different from either placebo or from captopril alone. The heart rate in response to a 5 min 70° head up tilt is shown in Table 2. There was no difference in heart rate between the three treatment groups at any time point. The effect of treatment on the systolic blood pressure response to handgrip 1.5 h after captopril is shown in Table 3. The maximum systolic blood pressure achieved during isometric exercise was reduced by captopril while the pressure achieved following captopril + naloxone was intermediate between, and not significantly different from, that achieved on the other two treatment days. Heart rates were similar in all three groups.

Angiotensin converting enzyme activity was reduced by captopril and plasma renin activity

Table 2 Heart rate (beats/min) following placebo, captopril alone and captopril plus naloxone

(a) Supine			
Time (h)	Placebo	Captopril	Captopril + naloxone
0	60.7 ± 12.5	61.0 ± 8.2	57.4 ± 11.0
1.5	57.9 ± 12.4	60.3 ± 9.2	58.7 ± 11.2
6	63.3 ± 13.3	64.6 ± 10.6	64.3 ± 11.1

(b) 5 min 70° head up tilt			
Time (h)	Placebo	Captopril	Captopril + naloxone
0	79.0 ± 14.8	80.6 ± 7.3	76.0 ± 9.4
1.5	83.4 ± 21.1	83.3 ± 16.5	83.6 ± 11.8
6	83.0 ± 17.7	86.1 ± 15.8	87.6 ± 16.6

was increased but there was no significant difference when captopril was given alone compared to the day when captopril was given together with naloxone (Table 4). The plasma noradrenaline concentration measured at the time of the fall in blood pressure produced by captopril was not significantly different either from the concentrations measured before captopril was given or from the corresponding values on the placebo day (Table 5).

Table 3 Systolic blood pressure (mm Hg) in response to sustained hand grip at 1.5 h

Time (min)	Placebo	Captopril	Captopril + naloxone
0	118 ± 8.2	107 ± 9.2	110 ± 8
1	125 ± 9.3	115 ± 12.2	118 ± 8.7
2	125 ± 8.8	115 ± 10.3	121 ± 9.2
3	128 ± 9	120 ± 9	119 ± 8.9

Discussion

There is now considerable evidence to suggest that endogenous opioids modulate blood pressure control at the level of the brain stem by reducing baroreflex sensitivity (Rubin *et al.*, 1981, 1983; Petty & Reid, 1982). The absence of compensatory tachycardia in response to a fall in blood pressure produced by captopril is consistent with decreased baroreflex activity. Since captopril has been shown to be capable of reducing the metabolism of enkephalins, we felt it possible that the absence of tachycardia seen following captopril could be explained by an action of this drug in preventing the metabolism of opioid peptides. This in turn could enhance their activity in brain stem areas which subserve cardiovascular homeostasis. This proposed mechanism would clearly depend on captopril or

Table 4 Angiotensin converting enzyme and plasma renin activity following captopril or captopril + naloxone

	Time (h)		
	0	1.5	6
ACE (C)	34.7 ± 15	10.9 ± 5	18.8 ± 8
ACE (C+N)	28.5 ± 10	9.1 ± 5	17.1 ± 5
PRA (C)	0.4 ± 0.2	1.3 ± 1	1.1 ± 0.8
PRA (C+N)	0.4 ± 0.2	2.3 ± 1.4	1.0 ± 0.7

C = captopril; N = naloxone

ACE activity is expressed as u ml^{-1} (see Methods) andPRA as $\text{ng ml}^{-1} \text{h}^{-1}$ of angiotensin I generated.Results are shown as mean ± s.d.; $n = 9$.

a metabolite crossing into the central nervous system.

If our hypothesis were correct, we expected naloxone to unmask a tachycardia in response to the fall in blood pressure produced by captopril. In addition, if an elevation of brain stem enkephalin concentration were involved in the actual lowering of blood pressure then we thought it possible that there would also be an attenuation of captopril's blood pressure lowering effect.

This study confirms previous observations that captopril lowers blood pressure without producing a reflex tachycardia (Millar *et al.*, 1981; Shepherd *et al.*, 1982). However, we found no difference in heart rate on the 3 study days, an observation which is not consistent with our initial hypothesis. The change in blood pressure following a 5 min 70° head up tilt was somewhat less easy to interpret. After 1.5 h there was a significant fall in blood pressure only on the day when captopril was given alone. When captopril and naloxone were given together the resulting fall in blood pressure was not significantly different from either placebo or captopril

treatments (Table 1). While appropriate to this study design, the Latin Square analysis which we used is a conservative statistical test which therefore could fail to suggest a real effect of naloxone in this instance.

Our findings with regard to systolic blood pressure response to isometric exercise were similar to the pattern of blood pressure seen during tilt. Again the combination of naloxone and captopril resulted in pressures which were intermediate between those of captopril alone and placebo. This suggests that the observed effects following naloxone could well be real rather than artefactual, but a clear inference is not possible.

In conclusion, we have not confirmed our initial hypothesis but data have been presented which suggest that endogenous opioids could still be involved in the haemodynamic actions of captopril.

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Table 5 Plasma noradrenaline concentration following captopril, captopril + naloxone and placebo

	Time (h)		
	0	1.5	6.0
<i>Supine</i>			
C	2.0 ± 0.8	2.1 ± 1.1	2.1 ± 1.1
C+N	1.9 ± 0.8	1.8 ± 0.4	1.8 ± 0.7
P	1.9 ± 0.8	1.9 ± 0.7	2.0 ± 0.6
<i>Tilt</i>			
C	4.2 ± 1.9	5.8 ± 2.2	4.9 ± 1.8
C+N	3.9 ± 1.5	4.4 ± 1.9	4.7 ± 1.1
P	4.0 ± 1.5	4.5 ± 1.7	4.4 ± 2.0

C = captopril; N = naloxone; P = captopril placebo + naloxone placebo

Plasma noradrenaline concentration is expressed as nmol l^{-1} .Results are shown as mean ± s.d.; $n = 9$.

References

- Arnold, M. A., Carr, D. B., Togasaki, D. M., Pian, M. C. & Martin, J. B. (1982). Caffeine stimulates -endorphin release in blood but not in cerebrospinal fluid. *Life Sci.*, **31**, 1017-1024.
- Arregui, A., Lee, C. M., Emson, P. C. & Iversen, L. L. (1979). Separation of human brain angiotensin-converting enzyme from enkephalin-degrading activity. *Eur. J. Pharmac.*, **59**, 141-144.
- Chiknas, S. G. (1979). A liquid chromatograph-assisted assay for angiotensin converting enzyme (peptidyl dipeptidase) in serum. *Clin. Chem.*, **25**, 1259-1262.
- Cody, R. J., Tarazi, R. C. & Bravo, E. L. (1979). Haemodynamics of an orally-active converting enzyme inhibitor (SQ 14225) in hypertensive patients. *Clin. Sci. mol. Med.*, **55**, 453-459.
- Cushman, D. W. & Cheung, H. S. (1970). Spectrophotometric assay and properties of the angiotensin-converting enzyme of rabbit lung. *Biochem. Pharmac.*, **20**, 1637-1648.
- Hatton, R. & Clough, D. P. (1982). Captopril interferes with neurogenic vasoconstriction in the pithed rat by angiotensin-dependent mechanisms. *J. cardiovasc. Pharmac.*, **4**, 116-123.
- Henry, D. P., Starman, D. J., Johnson, D. G. & Williams, R. H. (1975). A sensitive radioenzymatic assay for norepinephrine in tissue and plasma. *Life Sci.*, **16**, 375-384.
- Millar, J. A., McGrath, B. P., Matthews, P. G. & Johnston, C. I. (1981). Acute effects of captopril on blood pressure and circulating hormone levels in salt replete and depleted normal subjects and essential hypertensive patients. *Clin. Sci.*, **61**, 75-83.
- Petty, M. A. & Reid, J. L. (1982). The effect of opiates on arterial baroreceptor reflex function in the rabbit. *Naunyn-Schmiedeberg's Arch. Pharmac.*, **319**, 206-211.
- Roulston, J. E. & MacGregor, G. A. & Bird, R. (1980). The measure of angiotensin converting enzyme in subjects receiving captopril. *New Engl. J. Med.*, **303**, 397.
- Rubin, P. C., Blaschke, T. F. & Guilleminault, C. (1981). Effect of angiotensin converting enzyme inhibitor, on blood pressure fall during sleep. *Circulation*, **63**, 117-121.
- Rubin, P. C., McLean, K. & Reid, J. L. (1983). Endogenous opioids and baroreflex control in humans. *Hypertension*, **5**, 535-538.
- Shepherd, A. N., Campbell, B. C. & Reid, J. L. (1982). Effects of captopril, on angiotensin converting enzyme inhibitor, in normotensive volunteers. *J. cardiovasc. Pharmac.*, **4**, 381-387.
- Skinner, J. L. (1967). Improved assay methods for renin 'concentration' and 'activity' in human plasma. Methods using selective denaturation of renin substrate. *Circ. Res.*, **20**, 392-402.
- Stine, S. M., Yang, H. Y. & Costa, E. (1979). Inhibition of in situ metabolism of ³H-Met⁵-enkephalin and potentiation of Met⁵-enkephalin analgesia by captopril. *Brain Res.*, **188**, 295-299.
- Sturani, A., Chiarini, C., Esposti, E. D., Santoro, A., Zuccala, A., Zucchelli, P. & Millar, J. A. (1982). Captopril (letter). *New Engl. J. Med.*, **307**, 59-60.

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