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The effect of captopril on the reflex control heart rate: possible mechanisms

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1 Angiotensin converting enzyme inhibitors reduce blood pressure without reflex tachycardia, possibly as a result of enhanced hypothesis that this results from the removal of the parasympathetic activity. We examined the vagolytic action of angiotensin II or alternatively by acetylcholinesterase inhibition.

2 Both captopril and [Sar^{1}ala^8] angiotensin II, (saralasin), caused modest falls in blood pressure, without increasing heart rate in normotensive subjects.

3 Captopril and saralasin significantly attenuated the vagally mediated heart rate slowing after facial immersion in water. There was a close correlation between the effects produced by captopril and saralasin on the diving reflex.

4 Infusion of subpressor doses of angiotensin II, reversed the hypotensive effect of captopril and returned the bradycardia after facial immersion to placebo level.

5 *In vitro* neither captopril nor enalapril or lisinopril affected bovine erythrocyte acetycholinesterase activity.

6 The parasympathetic effect of angiotensin converting enzyme inhibitors appear to reflect a direct consequence of the removal of angiotensin II.

Keywords angiotensin converting enzyme inhibitors heart rate angiotensin II acetylcholinesterase parasympathetic effect

Introduction

Absence of a reflex tachycardia accompanying blood pressure reduction appears to be a property common to angiotensin converting enzyme inhibitors. This is unusual following acute dosing with a vasodilator drug and may reflect a consequence of the removal of angiotensin II on autonomic reflex mechanisms. Angiotensin II has been shown to have a presynaptic facilitatory effect on sympathetic neurotransmission *in vitro* (Starke, 1977; Zimmerman, 1981). However, converting enzyme inhibitors have no clinical or biochemically identifiable effects on sympathetic tone or sympathetically mediated reflexes in normal (Millar *et al.*, 1982) or hypertensive man (Niarchos *et al.*, 1982; Zanella *et al.*, 1981). Enhanced parasympathetic tone has been reported following converting enzyme inhibition with captopril in hypertensives (Sturani *et al.*, 1982) and in normotensives (Campbell *et al.*, 1985) and with the nonsulphydryl converting enzyme inhibitors enalapril and lisinopril (Ajayi *et al.*, 1985). This may contribute to the lack of reflex tachycardia. However, the possible mechanisms by which converting enzyme inhibitors may increase vagal tone in man are unclear. Angiotensin II has been shown to have opposing effects on heart rate. A baroreflex mediated bradycardia through its hypertensive action (Lumbers *et al.*, 1979) is mitigated by a central action which inhibits vagal discharge

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(Scroop & Lowe, 1968). Parasympathetic activation after captopril could be caused by the removal of the vagolytic action of angiotensin II at central (Scroop & Lowe, 1968; Lumbers *et al.*, 1979; Lee *et al.*, 1980) or peripheral (Potter, 1980) neuro-effector sites. Alternatively, converting enzyme inhibitors (being peptidase inhibitors) may cause cholinesterase inhibition. Acetylcholinesterases bear structural similarities to, and may act as peptidases in the brain (Chubb *et al.*, 1980). It has also been reported that captopril and edrophonium, a cholinesterase inhibitor, modified vagal reflexes in a similar manner (Campbell *et al.*, 1985).

Our study examines these alternative hypotheses in man and *in vitro*.

Methods

The studies were undertaken in two parts:
1. A clinical pharmacological assessment of the cardiovascular and autonomic effects of (a) converting enzyme inhibition with oral captopril 50 mg, (b) angiotensin II antagonism with [Sar^{1}ala^8] angiotensin II (saralasin) intravenously, (c) replacement of angiotensin II after either captopril or placebo by intravenous infusion.
2. An *in vitro* assessment of the effect of edrophonium and angiotensin converting enzyme inhibitors; captopril, lisinopril (MK521) and enalaprilat (MK422), the active metabolite of enalapril, on bovine erythrocyte acetylcholinesterase activity.

Subjects

Eight normotensive sodium replete males aged 24–35 years, and weighing between 58–77 kg took part in the study, after informed written consent and relevant physical and laboratory examinations. All subjects were judged healthy and the study was approved by the local Ethics Committee.

Captopril/angiotensin II study

This was a double-blind, placebo-controlled study, to observe the autonomic effects of captopril and placebo, and the effects of replacement of angiotensin II after captopril. Subjects received either captopril (50 mg) or identical placebo 1 week apart in a crossover fashion.

Subjects arrived fasted at the Clinical Investigation Unit at 08.30 h after avoiding coffee, tea, alcohol and cigarettes from 22.00 h on the night before the study day. After 10 min of supine rest, an intravenous cannula (Venflon) was inserted into an arm vein for the infusion of angiotensin II.

From precordial ECG leads, on line computer analysis of beat to beat (R-R interval) changes were possible (Kelman, personal communication). Blood pressure was recorded by means of a semiautomatic sphygmomanometer (Sentron, Bard Biomedical). After baseline measurements, subjects took captopril or placebo tablets with 200 ml water. Blood pressure and heart rate were recorded before and at 30, 60, 90, 120, 150 and 180 min after dosing.

Autononic reflexes Tests of autonomic reflex function were undertaken in duplicate at the following times:
30 min after dosing (pre-infusion).
30 min after starting angiotensin II infusion (during infusion).
30 min after stopping angiotensin II infusion (post-infusion).

Diving test Subjects sat for 5 min with faces 3–4 cm above a basin of water at 18–20°C. They immersed their faces into the water, without a deep inspiration. They remained immersed for as long as tolerated, but for at most 30 s. The ECG was continuously recorded. The parameter of interest was the difference between the pre-diving heart rate (heart period) and the longest heart period during facial immersion (minimum heart rate).

Valsalva's manoeuvre After basal recordings of blood pressure and heart rate, volunteers exhaled to hold a column of mercury in a modified sphygmomanometer at 50 mm Hg for 15 s. A second blood pressure recording was taken to coincide with the blood pressure overshoot which occurs 5–7 s after termination of the procedure. The ECG was continuously recorded during the strain, and for 30 s after relief of strain. The blood pressure overshoot represents the difference between pre- and post-Valsalva blood pressure. The Valsalva ratio is the ratio of the longest heart period after strain (phase IV) to the shortest during strain (phase II).

Cold pressor test Subjects immersed one arm in melting ice for 2 min. Blood pressure and heart rate were measured in the contralateral arm before, and at 1 min intervals. The results are expressed as the maximum change in blood pressure and heart rate induced by the test.

Standing to lying test This was adapted from the protocol described by Bellavere & Ewing (1982).

Subjects stood erect and still, for 5 min, and then lay down. The electrocardiogram was continuously recorded. The R–R intervals (beat to beat), in the first 25 beats of changing posture were recorded. The immediate heart rate response is under vagal control. The parameter of interest was the ratio of the mean erect heart period (average of last 10 R-R intervals in the standing position) to the minimum heart period during the test (standing to lying ratio). This is a reflection of vagal activity and is abnormal in patients with cardiac parasympathetic damage (Rodrigues & Ewing, 1983).

Intravenous infusions of angiotensin II A subpressor dose of angiotensin II (2.5 ng kg^{-1} min^{-1}) was infused into each subject for 60 min on both placebo and captopril days. This dose did not raise the diastolic blood pressure by more than 10 mm Hg. Blood pressure and heart rate were monitored every minute during the infusion. Autonomic reflexes were tested before and 30 min after starting the infusion (see Figure 1). The autonomic reflexes were repeated 30 min after stopping the infusion.

Saralasin infusion study

The same eight subjects were studied at least 1 week after completion of the protocol described above. On arrival, they lay supine for 30 min, and supine and erect blood pressures and heart rates were recorded. Autonomic tests were carried out prior to saralasin infusion. Saralasin was infused at 1 μg kg^{-1} min^{-1} for 20 min, thereafter autonomic tests were repeated while the infusion continued. The infusion rate was increased to 10 μg kg^{-1} min^{-1} for 10 min, and autonomic reflex testing repeated. The two dose levels administered were used to delineate a hypotensive dose from agonist dose see (Figure 2).

In vitro *assessment of the effect of converting enzyme inhibitors on bovine erythrocyte acetylcholinesterase activity*

The effects of edrophonium a cholinesterase inhibitor and the angiotensin converting enzyme inhibitors, captopril, enalapril and lisinopril were investigated *in vitro*. Acetylcholinesterase activity was determined by a colorimetric assay. The assay is dependent upon the measurement of the complex formed between thiocholine, generated by the action of cholinesterase on the substrate acetylthiocholine, and dithiobis (nitrobenzoate). Bovine erythrocyte acetylcholinesterase (E.C. 3117) was used as the standard enzyme preparation and the velocity of the

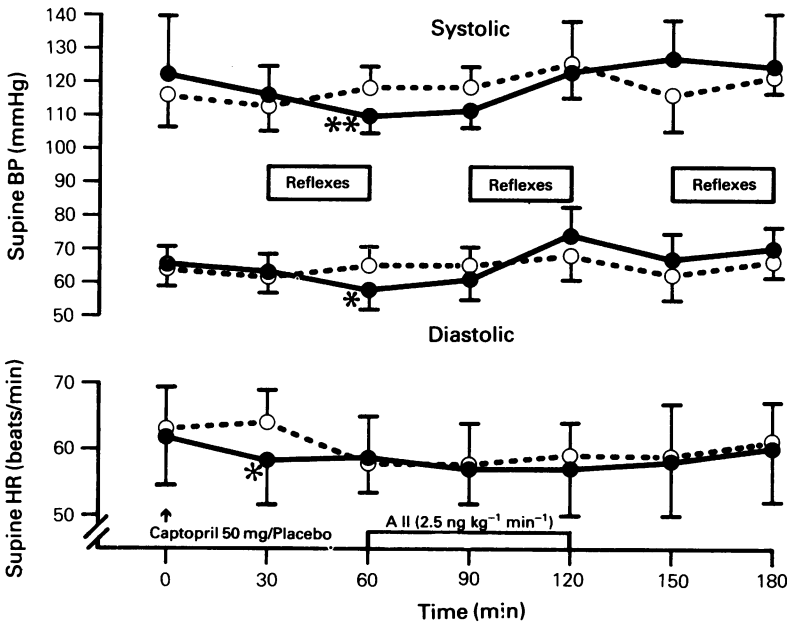

Figure 1 Supine blood pressure and heart rate after placebo (o---o), or captopril (•——•) before, during and after the infusion of angiotensin II 2.5 ng kg^{-1} min^{-1}, $n = 8$. mean ± s.d. $^{*}P < 0.05$, $^{**}P < 0.02$.

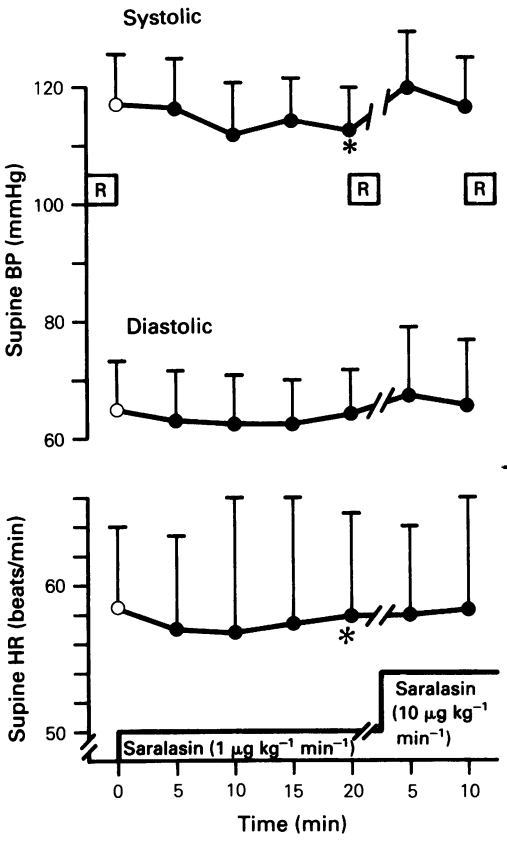

Figure 2 Supine blood pressure and heart rate before (○) and during infusions of saralasin (●——●), $n = 8$, mean ± s.d. $^*P < 0.02$ compared with pre-infusion values. R- reflexes.

enzyme reaction was measured as a change in optical density at 4605 nm over a 1 min period at substrate concentrations ranging from 0.01 to 0.05 nmol/l. Captopril, enalapril and lisinopril were added to give final concentrations of 0.3, 3 and 30 nmol/l, whilst edrophonium was added to give concentrations of 0.6, 3 and 6 nmol/l.

Statistical analyses

The results obtained from the cardiovascular and autonomic measurements are expressed as mean ± s.d. The results from the captopril/angiotensin II study were subjected to repeated measures analysis of variance (ANOVA) followed by *t*-tests for paired and unpaired data as appropriate. Results obtained from the saralasin protocol were analysed by Friedman two-way analysis of variance (ANOVA). The null hypothesis was rejected at $P < 0.05$).

The correlation between captopril and saralasin effects on the diving reflex was examined by linear regression analysis.

Results

General

All subjects tolerated infusions of saralasin and angiotensin without untoward effects. One subject experienced mild postural dizziness during saralasin infusion.

Blood pressure and heart rate

(a) *Effect of captopril with and without angiotenin II* Captopril significantly reduced supine systolic and diastolic blood pressure, 60 min after administration prior to angiotensin II infusion. Supine blood pressure fell significantly after captopril from 123± 8/65 ± 6 mm Hg to 111 ± 8/58 ± 6 mm Hg, ($P < 0.05$) compared with 118 ± 11/65 ± 6 mm Hg to 119 ± 7/65 ± 6 mm Hg after placebo. The profile of blood pressure and heart rate is shown (Figure 1). Infusion of 2.5 ng kg^{-1} min^{-1} of angiotensin II, abolished the hypotensive effect of captopril, but did not increase blood pressure on the placebo day. Supine heart rate was significantly reduced 30 min after dosing with captopril (placebo 63.5 ± 5.5 beats/min and captopril 58.0 ± 6.5 beats/min) ($P < 0.05$). Captopril did not affect erect blood pressure or heart rate.

(b) *Effect of saralasin infusions* These results are shown in Figure 2. Infusion of saralasin at 1 μg kg^{-1} min^{-1} caused a modest but significant fall in supine blood pressure compared to pre infusion values. Systolic blood pressure fell from 118.1 ± 8.0 mm Hg to 113.6 ± 9.1 mm Hg at the end of infusion ($P < 0.02$). Diastolic blood pressure fell insignificantly from 65.4 ± 8.2 mm Hg to 61.9 ± 6.8 mm Hg. Infusion at 10 μg kg^{-1} min^{-1} did not alter blood pressure.

Supine blood pressure and heart rate profile during saralasin infusion is shown in Figure 2. Supine heart rate fell from 59.2 ± 5.6 to 56.7 ± 8.2 beats/min at the end of the infusion ($P < 0.02$). Saralasin at both dose levels reduced the erect heart rate. Control heart rate after 5 min standing was 83.6 ± 8 beats/min. This was reduced to 78.5 ± 9.3 beats/min after saralasin 1 μg kg^{-1} min^{-1} ($P < 0.01$) and 77.3 ± 8.9 beats/min ($P < 0.01$) after saralasin 10 μg kg^{-1} min^{-1}.

Autonomic reflexes

These results are shown in Tables 1 and 2.

Table 1 The influence of angiotensin II (2.5 ng kg^{-1} min^{-1}) on autonomic responses to captopril, $n = 8$ (mean ± s.d.)

Test	*Pre-angiotensin II infusion*		*Angiotensin II infusion*		*Post-angiotensin II*	
	Placebo	*Captopril*	*Placebo*	*Captopril*	*Placebo*	*Captopril*
Diving test						
△ Heart rate (beats/min)	−30.4 ± 15.4	−23.6* ± 11.8	−24.7 ± 11.3	−22.8 ± 13.7	−25.6 ± 15.9	−19.8* ± 12.9
Valsalva manoeuvre						
SBP (mm Hg) post-Valsalva	119.6 ± 9.1	129.1 ± 15.0	124.1 ±19.8	124.0 ± 13.6	128.4 ± 16.3	132.5 ± 13.9
Valsalva ratio	1.88 ± 0.44	1.87 ± 0.52	2.19*† ± 0.41	1.90 ± 0.62	2.07 ± 0.74	1.99 ± 0.61
Cold pressor test						
△ SBP (mm Hg)	12.1 ± 9.3	6.0 ± 6.5	5.4 ± 8.9	13.4 ± 8.5	10.2 ± 8.8	12.3 ± 6.6
△ DBP (mm Hg)	8.9 ± 8.4	11.1 ± 4.4	15.1 ± 16.0	6.3 ± 10.3	7.9 ± 8.2	11.0 ± 10.1
△ Heart rate (beats/min)	9.6 ± 5.4	7.2 ± 5.9	1.8† ± 5.0	6.6 ± 3.2	6.0 ± 4.2	9.9 ± 6.1
Standing to lying test						
Standing to lying ratio	1.18 ± 0.14	1.25* ± 0.022	1.19 ± 0.063	1.20 ± 0.084	1.16 ± 0.076	1.19 ± 0.075

SBP and DBP = systolic and diastolic blood pressures respectively.
* $P < 0.05$ by ANOVA compared with placebo or captopril.
† Compared with pre-infusion.

Table 2 The effect of saralasin on autonomic reflexes, $n = 8$ (mean + s.d.)

Test	*Control*	*Saralasin (1 μg kg^{-1} min^{-1})*	*Saralasin (10 μg kg^{-1} min^{-1})*
Diving test			
△ Heart rate (beats/min)	−29.7 ± 14.2	−18.6 ± 9.4*	−19.8 ± 11.7**
Valsalva manoeuvre			
SBP (mm Hg) post-Valsalva	118.4 ± 16.0	123.9 ± 12.0	
Valsalva ratio	1.84 ± 0.61	1.83 ± 0.44	1.79 ± 0.47
Cold pressor test			
△ SBP (mm Hg)	13.3 ± 4.8	15.5 ± 12.0	7.7 ± 8.6
△ DBP (mm Hg)	8.1 ± 6.7	9.2 ± 6.4	5.2 ± 4.8
△ Heart rate (beats/min)	7.7 ± 4.6	4.1 ± 5.8	3.0 ± 7.3
Standing to lying test			
Standing to lying ratio	1.14 ± 0.050	1.20 ± 0.075	1.17 ± 0.113

SBP and DBP: Systolic and diastolic blood pressure
*$P < 0.05$**, $P < 0.01$ (ANOVA).

Diving reflex The results of diving test are shown in Table 1. Following placebo, all subjects responded to apnoeic facial immersion in water by a fall in heart rate (prolongation of heart period) (Figure 3). The effect of captopril, on the diving induced fall in heart rate was significantly different from that of placebo ($P = 0.004$, ANOVA).

Pre-infusion, captopril significantly attenuated the diving-induced bradycardia ($P < 0.05$). (The 95% confidence interval for the difference between captopril and placebo was from 0.813 to 12.86). The difference between captopril and placebo was abolished by infusion of angiotensin II, (95% confidence interval −1.4 to 6.89). After stopping angiotensin II, the heart rate change during diving on captopril was significantly different from placebo (95% confidence interval 0.345 to 11.25).

Influence of saralasin on diving reflex The effects of captopril, and saralasin infusion 1 μg and 10 μg kg^{-1} min^{-1} on the fall in heart rate during facial immersion are shown in Figure 3. Saralasin like captopril, significantly attenuated the diving induced heart rate fall. (Table 2).

The placebo corrected effects of captopril and saralasin on the vagally mediated diving

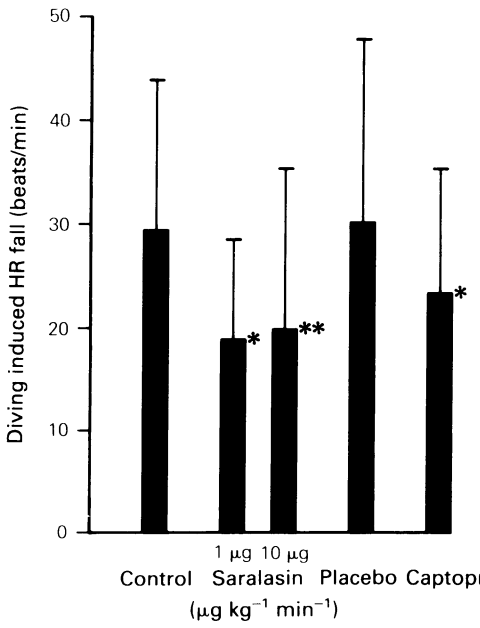

Figure 3 The diving reflex: The effect of captopril and saralasin on the bradycardia during apnoeic facial immersion in water, $n = 8$, mean ± s.d. *$P < 0.05$, **$P < 0.01$ (ANOVA).

bradycardia were closely correlated, saralasin at 1 μg kg^{-1} min^{-1} ($r = 0.7445$, $P < 0.033$) and at 10 μg kg^{-1} min^{-1} ($r = 0.7717$, $P < 0.026$) (Figure 4).

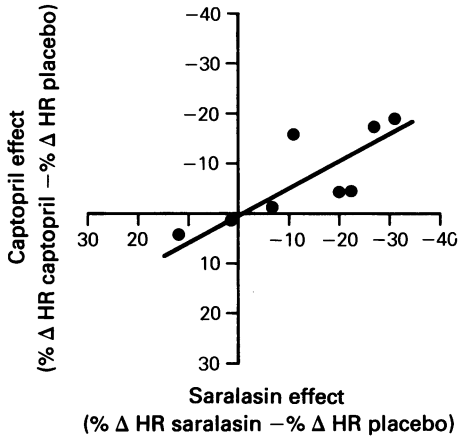

Figure 4 The correlation between the placebo corrected effects of captopril and saralasin (1 μg kg^{-1} min^{-1}) on the bradycardia during apnoeic facial immersion. $y = 0.462x - 0.488$, $r = 0.74$, $P < 0.03$, $n = 8$.

Valsalva's manoeuvre The blood pressure overshoot following the Valsalva manoeuvre was unaffected by captopril and angiotensin II infusion. Infusion of angiotensin II, was, however, associated with increased Valsalva ratio on placebo (time/treatment interaction $P = 0.011$). There was no alteration in blood pressure response or Valsalva ratio during infusion of saralasin.

Cold pressor test The actions of captopril or placebo on heart rate response to the cold pressor test are shown (Table 1). Captopril had no effect on the pressor or chronotropic response to cold stimulus. Infusion of angiotensin II 2.5 ng kg^{-1} min^{-1} significantly attenuated the heart rate rise on placebo, but not on captopril. Saralasin infusion did not significantly alter responses to the cold pressor test in the numbers studied.

Standing to lying test The effects of captopril and saralasin on the immediate heart rate response to lying down are shown in Tables 1 and 2. There was a transient cardio-acceleration (increase in heart rate) maximal at the third or fourth beat, followed by a progressive increase in R-R interval (fall in heart rate) by the 25th beat. Pre-infusion, captopril significantly increased the standing to lying ratio ($P < 0.05$). Saralasin had no significant effect on this parameter.

Bovine erythrocyte acetylcholinesterase activity

Line weaver-Burke (double reciprocal plots) of acetylcholinesterase enzyme kinetics on edrophonium, captopril, enalapril and lisinopril are shown in Figure 5. Edrophonium inhibited bovine erythrocyte acetylcholinesterase activity E.C 3117). Neither enalapril nor lisinopril or captopril affected cholinesterase activity.

Discussion

We investigated the putative mechanism(s) by which converting enzyme inhibitors may increase cardiac vagal activity. We hypothesized that this may result from an angiotensin II dependent action, through removal of angiotensin mediated central or peripheral vagal inhibition or a non-angiotensin II mediated effect such as cholinesterase inhibition. We found no evidence for the latter mechanism in a system in which the effect of edrophonium, a competitive cholinesterase inhibitor, was readily demonstrable.

Our results provide a comparative cardiovascular and autonomic profile of converting enzyme inhibition with captopril and specific antagonism of angiotensin II by saralasin in the same subjects. Both agents caused modest but significant falls in supine blood pressure in this group of sodium replete normotensives which is consistent with previous reports from our group (Shepherd *et al.*, 1982) and others. We did not observe a significant pressor response to saralasin at the doses used, indicating little agonist effect.

Neither captopril nor saralasin impaired the pressor or chronotropic response to the sympathetically mediated cold pressor test or the Valsalva manoeuvre. This finding is in accord with earlier findings in man (Millar *et al.*, 1982; Sturani *et al.*, 1982; Campbell *et al.*, 1985; Ibsen *et al.*, 1983; Ajayi *et al.*, 1985) and indicates that it is unlikely that converting enzyme inhibition exerts a substantial effect on sympathetic tone in man (Niarchos *et al.*, 1982). Moreover, infusion of angiotensin II did not facilitate the sympathetically mediated blood pressure overshoot following Valsalva or the blood pressure changes during the cold pressor test. Angiotensin II infusion, however, increased the Valsalva ratio and attenuated the heart rate increment during cold stimulus. While this may be related to baroreflex mediated bradycardia, it is not clear why the changes were observed after

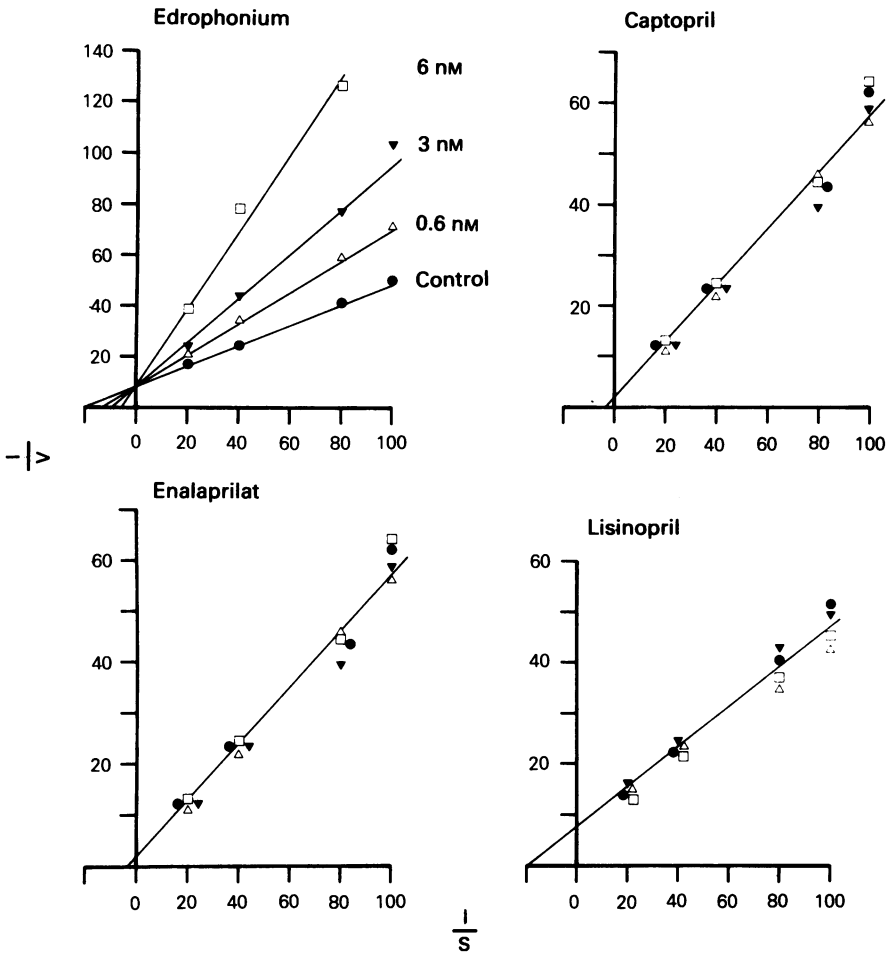

Figure 5 The influence of edrophonium, captopril, enalapril and lisinopril on bovine erythrocyte acetylcholinesterase activity: Lineweaver-Burke plots of the reciprocal of velocity ($\frac{1}{V}$) against the reciprocal of substrate concentrations ($\frac{1}{S}$). For captopril, enalapril and lisinopril (● = control; ▼ 30 nM, △ 3 nM, and □ 0.3 nM).

placebo but not after captopril. This finding contrasts with captopril which has been reported to decrease the Valsalva ratio and increase heart rate increment during cold stress (Campbell *et al.*, 1985). It is also noteworthy that subtle reflex changes to angiotensin II occur in the absence of blood pressure change (Bravo & Tarazi, 1978).

The bradycardia induced by apnoeic facial immersion in water is due to parasympathetic stimulation (Finley *et al.*, 1979) and is abolished by atropine. Paradoxically, however, the heart rate slowing is attenuated by edrophonium, a cholinesterase inhibitor and parasympathomimetic agent. A possible explanation of this is that after enhancement of initial vagal tone by pharmacological means, the scope for a further physiological increase in vagal tone is limited.

Both captopril and saralasin significantly attenuated this diving induced heart rate fall, in a similar manner to edrophonium. Further, there was a positive and significant correlation between the placebo corrected effects of captopril and saralasin on the vagally mediated bradycardia. The similarity between the autonomic effects of captopril and an angiotensin II antagonist indicate that the vagal effects of converting enzyme inhibitors are likely to be mediated by angiotensin II withdrawal. This view is supported by the finding of Bravo & Tarazi (1978) of enhanced parasympathetic activity following angiotensin II antagonist [Sar^{1}Thr^8] angiotensin II in the dog (Bravo & Tarazi, 1978) and of Wallace *et al.* (1976) in man.

Infusion of a subpressor dose of angiotensin II reversed the hypotensive action of captopril, in accord with the observation that the mechanism of action of ACE inhibitors is largely due to a withdrawal of vasoconstrictor role of angiotensin II. Infusion of angiotensin II appeared to abolish the difference between captopril and placebo on diving bradycardia. Consistent with this is the increased standing to lying ratio, a reflection of vagal activity (Rodrigues & Ewing, 1983) on captopril, before angiotensin II infusion.

Whether the vagal effects of angiotensin II withdrawal following converting enzyme inhibition occur at central sites (Lumbers *et al.*, 1979; Lee *et al.*, 1980) or at the periphery (Potter, 1982) remains to be determined. The vagal stimulating activity of the converting enzyme inhibitor captopril which was previously reported (Sturani *et al.*, 1982; Campbell *et al.*, 1985) was confirmed in this study.

In conclusion, there was a close similarity between the effects of captopril and angiotensin II antagonist, saralasin, on parasympathetic responsiveness. Angiotensin II reversed the effect of captopril on the diving reflex. These findings are consistent with our hypothesis that the increase in cardiac vagal activity after the ACE inhibitor, captopril, reflects a direct consequence of the removal of angiotensin II and not cholinesterase inhibition.

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References

Ajayi, A. A., Campbell, B. C., Howie, C. A. & Reid, J. L. (1985). Acute and chronic effects of enalapril and lisinopril on reflex control of heart rate in normotensive man. *J. Hypertension* **3**, 47–53.

Bellavere, F. & Ewing, D. J. (1982). Autonomic control of the immediate heart rate response to lying down. *Clin. Sci.*, **62**, 57–64.

Bravo, E. L. & Tarazi, R. C. (1978). Haemodynamics of an angiotensin II antagonist in normal, unanesthetised dogs. *Circ. Res.*, **43**, Suppl. I, 27–31.

Campbell, B. C., Sturani, A. & Reid, J. L. (1985). Evidence of parasympathetic activity of the angiotensin converting enzyme inhibitor, captopril, in normotensive man. *Clin. Sci.*, **68**, 49–56.

Chubb, I., Hodgson, A. J. & White, G. H. (1980). Acetylcholine esterase EC3117 hydrolyses substance P. *Neuroscience,* S (12), 2065–2072.

Finley, J. P., Bonet, J. F. & Waxman, H. B. (1979). Autonomic pathways responsible for bradycardia on facial immersion. *J. appl. Physiol. Resp. Environ. Exercise Physiol.*, **47**, 1218–1222.

Ibsen, H., Egan, B., Osterziel, K., Vander, A. & Julius, S. (1983). Reflex haemodynamic adjustments and baroreflex sensitivity during converting enzyme inhibition with MK421 in normal humans. *Hypertension*, **5** (Suppl I): I-184–I–191.

Lee, W. B., Ismay, H. J. & Lumbers, E. R. (1980). Mechanisms by which angiotensin II affects the heart rate of conscious sheep. *Circ. Res.*, **47**, 286–292.

Lumbers, E. R., McCloskey, D. I. & Potter, E. K. (1979). Inhibition by angiotensin II of baroreceptor evoked activity in cardiac vagal efferent nerves in the dog. *J. Physiol.*, **294**, 69–80.

Millar, J. A., Derkx, F. M., McLean, Kathleen & Reid, J. L. (1982). Pharmacodynamics of converting enzyme inhibition: The cardiovascular endocrine and autonomic effects of MK421 and MK521. *Br. J. clin. Pharmac.*, **14**, 347–355.

Niarchos, A. P., Pickering, T. G., Morganti, A. & Laragh, J. H. (1982). Plasma catecholamines and cardiovascular responses during converting enzyme inhibition in normotensive and hypertensive man. *Clin. exp. Hyper.*, A4 (4 & 5), 761–789.

Potter, E. K. (1982). Angiotensin inhibits action of the vagus nerve at the heart. *Br. J. Pharmac.*, **75**, 9–11.

Rodrigues, E. A. & Ewing, D. J. (1983). Immediate heart rate response to lying down: simple test for cardiac parasympathetic damage in diabetics. *Br. med. J.*, **287**, 800.

Scroop, G. C. & Lowe, R. D. (1968). Central pressor effect of angiotensin mediated by the parasympathic nervous system. *Nature,* **220**, 1331–1332.

Shepherd, A. N., Campbell, B. C. & Reid, J. L. (1982). Effects of captopril on angiotensin converting enzyme inhibitors in normotensive sodium replete volunteers. *J. cardiovasc. Pharmac.*, **4**, 381–387.

Starke, K. (1977). Regulation of noradrenaline release by the pre-synaptic receptor, systems. *Rev. Physiol Biochem. Pharmac.*, **3** (Suppl. 1), 514–523.

Sturani, A., Chiarini, C., Degli Esposti, E., Santoro, A., Zuccala, A. & Zuchelli, P. (1982). Heart rate control in hypertensive patients treated by captopril. *Br. J. clin. Pharmac.*, **14**, 849–855.

Wallace, J. M., Kelm, H. J., Case, D. B., Lopez, J. & Laragh, J. H. (1976). Reductions of cardiac output with angiotensin blockade in hypertension. *Clin. Res.*, **24**, 244A.

Zanella, M. T., Bravo, E. L., Fouad, F. M. & Tarazi, R. C. (1981). Long term converting enzyme inhibition and sympathetic nerve function in hypertensive humans. *Hypertension,* **3** (Suppl II) II: 216–221.

Zimmerman, B. G. (1981). Adrenergic facilitation by angiotensin: does it serve a physiological function? *Clin. Sci.*, **60**, 343.

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