*Br. J. clin. Pharmac.* (1990), **29**, 774–776

# The effect of local converting enzyme inhibition on the dilator response to substance P in the human forearm

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It has been proposed that angiotensin converting enzyme (ACE) may play a part in the metabolism of substance P. Reduced metabolism following treatment with ACE inhibitors may cause accumulation of substance P to produce the adverse effect of cough. It has been shown in this study that, in contrast to angiotensin I and bradykinin, inhibition of local vascular ACE does not interfere with the vascular effects of substance P on forearm resistance vessels when this peptide is infused into the brachial artery of normal volunteers. These results suggest that endothelial ACE plays little part in the metabolism of intravascular substance P.

**Keywords** kininase II substance P forearm man

## Introduction

Angiotensin converting enzyme (ACE) is a glycoprotein that catalyses the release of carboxy-terminal dipeptides from a variety of substrates, including bradykinin and angiotensin I and has been shown to possess endopeptidase activity against substance P *in vitro* (Yokosawa *et al.*, 1985).

Treatment with drugs which inhibit ACE causes a dry, unproductive cough in up to 14% of patients (McEwan *et al.*, 1988; Webb *et al.*, 1986). The mechanism for this unusual adverse effect is not clear, but, as it is seen with all members of this class of drugs, it may be directly related to inhibition of ACE.

It has been variously proposed that the mechanism underlying the production of cough following therapy with ACE inhibitors may involve impaired metabolism of either bradykinin (Fuller & Choudry, 1987) or substance P (Morice *et al.*, 1987), both of which are known to produce cough. Bradykinin is rapidly inactivated by ACE and we have previously shown that local inhibition of vascular ACE potentiates the dilator action of bradykinin in forearm resistance vessels when infused into the brachial artery in man (Benjamin *et al.*, 1989). There is also evidence both *in vitro* (Yokosawa *et al.*, 1985) and *in vivo* (Casceiri *et al.*, 1983) in animals that the actions of substance P may be enhanced by inhibition of ACE.

The purpose of the present study was to investigate whether the dilator action of substance P in human forearm resistance vessels is influenced by local inhibition of vascular ACE.

## Methods

Six healthy male subjects, aged between 21 and 34 years, took part in the study, which was approved by St George's Hospital ethics committee. Subjects were studied in the supine position with room temperature (between 25 and 28° C) maintained within ± 1° C for each study.

Forearm blood flow was measured in both

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arms using venous occlusion plethysmography with temperature-compensated mercury-in-silastic strain gauges as previously described (Benjamin *et al.*, 1989). A 27 SWG steel needle was inserted into the left brachial artery under 1% lignocaine local anaesthesia.

Saline, then incremental doses of substance P (0.25, 0.5 and 1pmol $min^{-1}$) were infused into the left brachial artery, each for 10 min. After a further 10 min of saline infusion, enalaprilat (5μg $min^{-1}$), the active form of the converting enzyme inhibitor enalapril, was infused alone for 10 min and then co-infused with the same incremental doses of substance P, again each for 10 min. Forearm blood flow was measured during the final 3 min of each infusion period and the mean of the last five measurements was used for analysis. Percentage change in flow compared with saline was calculated as described by Benjamin *et al.* (1989). Statistical comparison was by the Wilcoxon signed rank test.

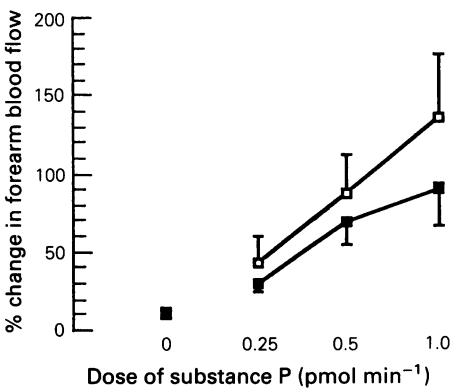


**Figure 1** Substance P was infused into the brachial artery alone (□) and then together with enalaprilat (5μg $min^{-1}$; ■). The percentage change in forearm blood flow was not significantly changed by co-infusion of enalaprilat.

## Results

Substance P, when infused alone, caused a dose-dependent increase in blood flow in the infused forearm. Blood flow increased from 5.2 ± 0.7 ml 100 $ml^{-1}$ $min^{-1}$ during saline infusion, to 11.9 ± 1.2 ml 100 $ml^{-1}$ $min^{-1}$ at the highest dose of substance P ($P < 0.01$). Blood flow in the control forearm was not significantly altered, at 4.7 ± 0.6 and 5.2 ± 1.0 ml 100 $ml^{-1}$ $min^{-1}$ during saline and substance P respectively. Enalaprilat (5 μg $min^{-1}$) alone produced no significant change in blood flow in the infused forearm (from 6.4 ± 1.1 to 7.1 ± 1.4 ml 100 $ml^{-1}$ $min^{-1}$). The increase in blood flow in the infused forearm when substance P was co-infused with enalaprilat was not significantly different from that seen when substance P was infused alone; from 7.1 ± 1.4 to 12.1 ± 1.2 ml 100 $ml^{-1}$ $min^{-1}$. Blood flow in the control arm was not significantly altered; 5.3 ± 1.1 and 6.0 ± 1.3 ml 100 $ml^{-1}$ $min^{-1}$. The percentage change in forearm blood flow with substance P, with and without co-infusion of enalaprilat, are shown in Figure 1.

## Discussion

Angiotensin converting enzyme is an ubiquitous enzyme, found in plasma, and occurring as a membrane-bound ectoenzyme predominantly on vascular endothelial cells (Caldwell *et al.*, 1976; Johnston & Kohzuki, 1989). The capacity for substantial metabolism of both angiotensin I and bradykinin, initially demonstrated across the pulmonary circulation (Ferreira & Vane, 1967; Semple, 1977), has since been shown across the forearm circulation in man (Benjamin *et al.*, 1989). Hence this vascular bed provides a means by which to investigate the role of ACE in the metabolism of substance P in man.

The mechanisms by which substance P is metabolised in man are not entirely clear. The observation that intravascular administration of captopril enhances the sialogenic effects of substance P in the rat (Casceiri *et al.*, 1983) suggests that ACE may be involved in the metabolism of this peptide. The observations of Morice *et al.* (1987) that capsaicin-induced cough is potentiated by ACE inhibition in man, while cough stimulated by distilled water or citrate is unaffected, lends further support to this hypothesis, as capsaicin is known to cause release of substance P, and other tachykinins, from c-fibre nerve terminals.

The results of the present study show no effect of the ACE inhibitor enalaprilat on the vasodilation in response to local infusion of substance P. The slightly reduced dilator response to the second infusion of substance P in each study is consistent with the small degree of tachyphylaxis seen when this peptide is infused into the forearm (McEwan *et al.*, 1988). The dose of enalaprilat used in this study (5 μg $min^{-1}$) has been demonstrated previously to produce marked inhibition of endothelially located ACE within the forearm (Benjamin *et al.*, 1989), resulting in a three-fold increase in the dilator response to bradykinin, and a marked reduction in the response to angiotensin I, such that 100-fold greater doses are required to produce an

equivalent reduction in local blood flow. Hence, the absence of an effect of local ACE inhibition on the response to substance P in the present experiments shows that endothelially located ACE does not play an important role in the metabolism of this peptide in the forearm vascular bed. The failure of the forearm vascular ACE to metabolise substance P suggests that this enzyme has little tachykinin endopeptidase activity compared with its dipeptidyl carboxypeptidase activity. If this is also true of pulmonary ACE, it is less likely that ACE inhibition will directly result in elevated pulmonary substance P concentrations. It is, however, possible that increased concentrations of bradykinin resulting from ACE inhibition result in a secondary release of substance P from nerve terminals within the lung (Buck & Burks, 1986), and that this effect may be enhanced by capsaicin. It is also possible that in a small number of patients who develop cough with ACE inhibitor therapy the metabolism of substance P is more dependent on converting enzyme, and are therefore more sensitive to its inhibition.

N.B. was supported by Wellcome Research Training Fellowship.

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*(Received 5 January 1990, accepted 28 February 1990)*