stance P was different; we obtained it from Peptide Research Laboratories, Osaka, Japan whereas theirs was from Clinalfa, Calbiochem-Novabiochem (UK) Ltd, Nottingham, U.K.

Although L-NMMA did not inhibit substance P-induced forearm vasodilatation in our study, another group from our laboratory has demonstrated that L-NMMA inhibited substance P-induced coronary vasodilatation in humans but not forearm vasodilatation [3]: the same substance P was used for these two studies. Moreover, we have shown that L-arginine, a substrate of nitric oxide, augments acetylcholine-induced forearm vasodilatation but does not affect substance P-induced vasodilatation [4]. These findings support the notion that substance P-induced vasodilatation is not mediated by nitric oxide, at least in the forearm. Finally, there may be racial differences because Panza et al. [5] from the U.S.A. reported that substance P-induced vasodilatation was inhibited by L-NMMA. It may be necessary to perform experiments with the same protocol after exchanging sources of substance P.

REFERENCES


Contribution of nitric oxide to vasodilatation by substance P in human forearm vessels: author’s reply

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Clinical Science published two studies in the February 1997 issue [1, 2] that used the same drugs and similar techniques of intra-brachial infusion and bilateral forearm venous occlusion plethysmography but arrived at contrasting conclusions. We were unaware of the work of Shiramoto and colleagues before publication. We feel it was unhelpful that the researchers were not given the opportunity to draw out differences in the work, and confusing for the reader that these two articles were otherwise published without editorial guidance.

There is good evidence from animal studies [3, 4] and from clinical studies in the forearm [5–7] and coronary circulation [8] that substance P-induced vasodilatation is, at least in part, nitric oxide dependent.

Forearm administration of nitric oxide synthase inhibitors, such as L-NMMA, causes baseline vasoconstriction and reduces resting blood flow. This will alter resistance vessel tone and geometry as well as potentially influence the concentration of co-infused agents of interest [9]. Indeed, our group has shown previously [10] that baseline vasoconstriction potentiates the vasodilator response to vasodilators such as bradykinin. Thus, although substance P-induced vasodilatation had been shown previously to be inhibited by L-NMMA [5, 8], we wanted to confirm that this was not a non-specific effect of baseline vasoconstriction, and assess its nitric oxide dependence with reference to the standard agent, acetylcholine. We therefore compared acetylcholine- and substance P-mediated vasodilator responses in the presence of both L-NMMA and noradrenaline at doses which caused equivalent baseline vasoconstriction. Not only did we confirm earlier observations that L-NMMA inhibits substance P-mediated vasodilatation, but we also showed that acetylcholine and substance P vasodilator responses are augmented in the presence of baseline vasoconstriction with noradrenaline.

Our findings appear to be at odds with those of Shiramoto and colleagues. However, there are some difficulties between the approaches of the two studies. One striking feature of Shiramoto’s paper was the brevity of L-NMMA infusion. While both we and Shiramoto used locally active doses of L-NMMA, Shiramoto gave the infusion for 5 min only, whereas we co-infused L-NMMA throughout the study. The haemodynamic effects of L-NMMA, given either systemically [11] or locally [12] for 5 min, begin to wane after 10 min. A second important point is that, because baseline vasoconstriction per se augments responses to substance P, lack of augmentation in the presence of L-NMMA in Shiramoto’s study may be consistent with inhibition of the response. Without a constrictor control, no clear conclusion can be drawn. As Professor Wallace points out, one further clear difference relates to the potency of substance P-induced vasodilatation in the two papers. We used a different source of substance P from that used by Shiramoto and colleagues and sub- aliquoted and filtered it into vials for later use. The vials were all made in one batch and stored at −80°C before use. We have subsequently used substance P directly from the Clinalfa vials and have found much higher potency of substance P-induced vasodilatation [13]. This suggests that there may have been a loss of active peptide during the filtering and freezing process which may account for the difference in potency of the substance P between the two studies. Indeed, we have only demonstrated that L-NMMA inhibits responses to doses of sub-
stance P causing modest increases in blood flow. As we indicated in our paper [2]: '... caution should be applied to the extrapolation of our study findings to higher doses of substance P...'. Nevertheless, other workers have demonstrated inhibition with L-NMMA at doses of substance P causing greater (100–200%) increases in blood flow [5, 8].

We suggest that one further approach, which may help to resolve this issue, would be to use the ‘nitric oxide clamp’ developed by Stroes et al. [14]. Here, the baseline vasoconstriction produced by complete inhibition by L-NMMA of endogenous nitric oxide synthesis is ‘corrected’ by co-infusion of an exogenous nitric oxide donor such as sodium nitroprusside. This technique essentially eliminates basal and stimulated endogenous nitric oxide production and restores the resistance vessel tone and geometry to baseline conditions. If substance P is then administered while maintaining the ‘nitric oxide clamp’ the degree of vasodilatation compared with ‘unclamped’ conditions should define the contribution of nitric oxide. It is our suspicion that the contribution from nitric oxide to vasodilatation may decrease as the degree of vasodilatation increases, so that our results are not necessarily inconsistent with those of Shiramoto and colleagues.

REFERENCES


Editorial comment

Both of the original papers (Shiramoto et al. Clin Sci 1997; 92: 123–31; Newby et al. Clin Sci 1997; 92: 133–8) received full independent peer review and were evaluated by different editors of Clinical Science. No scientific journal has the policy of informing authors that other papers in the journal appear to disagree with their data. The peer review process can only establish whether a paper meets appropriate criteria in terms of validity of the data presented, and once a paper has been through a thorough reviewing process it is up to readers to decide on the merits of the data relative to those from any other paper.

Malcolm J. Jackson (Editor-in-Chief) on behalf of the Editorial Board