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# A comparison of the contractile effects of 5-hydroxytryptamine, sumatriptan and MK-462 on human coronary artery *in vitro*

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1 MK-462 (*N*,*N*-dimethyl-2-[5-(1,2,4-triazol-1-ylmethyl)-1H-indol-3-yl]ethylamine) is a novel selective 5-$HT_{1D}$-receptor agonist which in clinical trials has been shown to be an effective antimigraine agent. As angiographic studies have shown that sumatriptan (an established 5-$HT_{1D}$-receptor agonist) can cause coronary artery vasoconstriction in patients, we compared the effects of MK-462 with those of 5-HT and those of sumatriptan, on isolated segments of human coronary artery *in vitro*.

2 Coronary arteries were obtained from explanted hearts from patients ($n$ = 22, 2 females, 20 males, aged 21-60 years) undergoing cardiac transplantation. Endothelium-denuded ring segments of coronary artery, 2mm long were mounted in organ-baths for isometric tension recording. For each arterial ring segment, a cumulative concentration-effect curve to either 5-HT, sumatriptan or MK-462 was determined. After maximal response to each agonist had been obtained, ketanserin (a 5-$HT_2$ receptor antagonist) 0.6 μM was added to the tissue bath, followed by methiotepin (0.6 μM) and the reduction in tension produced by the addition of each antagonist was determined.

3 Out of 22 coronary arteries studied, only 10 showed any response (contraction) to 5-HT. Not all arteries which responded to 5-HT contracted in response to both sumatriptan and MK-462 (one ring from each artery being exposed to a single agonist in each case). Both sumatriptan and MK-462 ($E_{max}$ values of 57.6% ($n$ = 6) and 32.5% ($n$ = 8) with respect to 45 mM KCl, respectively) were significantly less efficacious than 5-HT ($n$ = 10) in contracting human coronary artery ($P< 0.03$ and $P < 0.001$ respectively) and furthermore MK-462 was significantly less effective than sumatriptan ($P < 0.04$). These $E_{max}$ values were similar to the $E_{max}$ values which were obtained from four vessels which responded to all three agonists. Ketanserin partially reduced the response to each of the agonists, and the further addition of methiotepin removed the remainder of the response indicating the involvement of 5-$HT_{1D}$-receptors and possibly 5-$HT_2$-receptors.

**Keywords** human coronary artery coronary vasoconstriction 5-hydroxytryptamine

## Introduction

Previous studies with 5-hydroxytryptamine (5-HT) have suggested that this endogenous neurotransmitter and local hormone may be involved in the pathophysiology of coronary artery vasospasm [1, 2, 3, 4]. In human isolated coronary artery, 5-$HT_{1D}$ receptors can mediate vasodilatation in pre-contracted vessels, which may or may not be endothelium-dependent [5, 6], whilst both 5-$HT_{1D}$- and 5-$HT_2$-receptors appear to be able to mediate coronary vasoconstriction [2, 7]. Thus, 5-HT can give rise either to coronary vasodilatation or to coronary vasoconstriction, depending on the balance between these opposing

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effects. In coronary angiographic studies, 5-HT increased coronary artery diameter and blood flow in normal subjects, but decreased these parameters in patients with coronary artery disease [3, 4]. In contrast to the *in vitro* studies, however, these *in vivo* studies showed that the decrease in coronary artery diameter was antagonised by ketanserin, leading to the suggestion that 5-$HT_2$ receptors were involved in mediating vasoconstriction in response to 5-HT [8].

Sumatriptan is a drug used for the treatment of acute migraine, and is reported to be a selective agonist at the 5-$HT_{1D}$ receptor [9, 10]. As with 5-HT, intravenous administration of sumatriptan has been shown to produce coronary artery constriction in patients undergoing coronary angiography [11] and in this study, coronary vasoconstriction in response to this drug occurred without associated symptoms or electrocardiographic changes. Recently, however, there have been a few reports of cardiac ischaemia [12, 13], and one report of myocardial infarction [14] with sumatriptan. Although coronary side-effects are rare, such cases are of concern in the use of drugs of this class in patients with migraine and concomitant silent ischaemic heart disease.

MK-462 (*N*,*N*-dimethyl-2-[5-(1,2,4-triazol-1-ylmethyl)-1H-indol-3-yl]ethylamine) is a novel selective 5-$HT_{1D}$-receptor agonist [15] and has been shown in placebo controlled clinical trials to be an effective antimigraine agent [16]. In order to ascertain the effects of MK-462 on coronary vasomotor tone, we examined the effects of MK-462 on isolated segments of human coronary artery *in vitro*. Because clinical and organ bath evidence has shown variability in 5-HT responsiveness [17], we used 5-HT itself to ascertain which arteries were 5-HT responsive and compared 5-HT evoked contractions with responses evoked by MK-462. We also used sumatriptan as a positive control for 5-$HT_{1D}$-receptor induced contractions. For each artery responding to either 5-HT, MK-462 or sumatriptan we determined whether the contractions were reversed by methiotepin or ketanserin.

The study was approved by the University of Cambridge and Addenbrooke's Hospital Ethics Committee.

## Methods

### *Patients*

Coronary artery tissue was obtained from 22 patients undergoing orthotopic cardiac transplantation. Anaesthesia was with nitrous oxide/oxygen and trichloroethylene by inhalation, with intravenous papaveretum and midazolam, and vecuronium as muscle relaxant. Upon institution of cardiopulmonary bypass, heparin was administered, with pancuronium as muscle relaxant and trichloroethylene for maintenance of anaesthesia. Other drugs administered to all patients pre- and perioperatively were prednisolone, azathioprine, cyclosporin, anti-thymocyte globulin, cefotaxime and flucloxacillin. Other drugs being taken preoperatively by different patients were: diuretics (thiazides, loop diuretics and potassium-sparing diuretics), angiotensin converting enzyme inhibitors, warfarin, heparin, aspirin, nitrates, calcium channel antagonists, histamine receptor antagonists, dopamine, digoxin, simvastatin, oral hypoglycaemics (sulphonylureas and biguanides), opiates, paracetamol, non-steroidal anti-inflammatory agents, disopyramide, amiodarone, flosequinan, omeprazole, allopurinol and benzodiazepines. All patients had end-stage (New York Heart Association class IV) heart failure, secondary to ischaemic heart disease ($n$ = 8), dilated cardiomyopathy ($n$ = 9), Eisenmenger's syndrome ($n$ = 2), rheumatic heart disease ($n$ = 1), mitral valve disease of undetermined aetiology ($n$ = 1) or transposition of the great arteries ($n$ = 1).

### *Preparation of coronary artery rings*

Coronary artery was immediately dissected from the explanted heart and collected in oxygenated modified Krebs solution of the following composition (mM): $Na^+$ 125, $K^+$ 5, $Ca^{2+}$ 2.25, $Mg^{2+}$ 0.5, $Cl^-$ 98.5, $SO_4^{2-}$ 0.5, $HCO_3^-$ 32, $HPO_4^{2-}$ 1, and EDTA 0.04. The artery was cleaned of surrounding fat and connective tissue and the endothelium removed by passing a metal rod gently through the lumen. It was then cut into 2 mm-long rings (multiple segments being obtained from a single artery) and stored overnight at 4° C prior to use. The efficiency of endothelium-removal was tested for by a lack of a relaxant response to ACh (1–20 μM; data not shown).

Ring segments were placed over two L-shaped steel wire hooks, the lower hook being clamped on a tissue stand, the upper hook being attached to a Swema SG4-45 strain-gauge transducer, and mounted in a 50 ml organ-bath maintained at 37° C and containing modified Krebs solution as above supplemented with (mM): $Na^+$ 15, fumarate 5, pyruvate 5, L-glutamate 5, and glucose 10. This was bubbled constantly with 95% $O_2$/5% $CO_2$. Rings were tensioned to 20 mN and allowed to equilibrate for 30 minutes; following this, the contractile response to 45 mM KCl was determined. Repeated contractions were elicited to 45 mM KCl (with washouts in between to restore baseline tension) until stable reproducible contractions were obtained.

### *Concentration-effect curves to 5-HT, sumatriptan and MK-462*

Following washout of the KCl, once baseline tension had been re-established, cumulative concentration-effect curves were determined to 5-HT, sumatriptan and MK-462 (concentration range 2 nM–60 μM, half-log unit increments). Only one concentration-effect curve was determined for each individual ring segment, in order to avoid possible interaction effects of exposure of tissues to multiple agonists. Following the highest dose of each agonist, 0.6 μM ketanserin was added to the bath, and the response (i.e. reduction in tension) determined. Following this, 0.6 μM

methiotepin (in the continuing presence of ketanserin) was added, and the response once again determined. All contractions were expressed as a percentage of the force developed in response to 45 mM KCl for each ring segment.

*Statistics*

Concentration-effect curves were fitted to data for each individual ring segment by least squares analysis of regression using the equation:

$$E = E_{max}/(1+(EC_{50}/\text{agonist concentration})^{nH})$$

where $E_{max}$ is the maximum contraction evoked by each agonist (relative to 45 mM KCl = 100%), $EC_{50}$ is the agonist concentration which evokes half-maximal response (relative to the maximum response produced by that particular agonist) and nH is the Hill coefficient; this was performed using an iterative procedure with Grafit software (Erithacus). The slope coefficients from each concentration-effect curve (using untransformed data) were analysed using one-way analysis of variance with repeated measures on agonist treatment. *A priori* post-analysis of variance comparisons (with a correction for multiple comparisons) were made between each pair of agonists (5-HT *vs* sumatriptan, 5-HT *vs* MK-462 and sumatriptan *vs* MK-462). $E_{max}$ values for each agonist group were compared using unpaired Student's *t*-test.

*Materials*

MK-462 was the benzoate salt (*N*,*N*-dimethyl-2-[5-(1,2,4-triazol-1-ylmethyl)-1H-indol-3-yl]ethylamine)) and sumatriptan was the succinate salt. Both agonists were synthesized by Merck Sharpe & Dohme Research Laboratories. Ketanserin tartrate and (±)-methiotepin maleate were from RBI, St Albans, UK. 5-HT and all other chemicals were from Sigma Chemical Company Ltd, Poole, UK.

## Results

Coronary arteries were studied from 22 patients undergoing cardiac transplantation. Ring segments ($n$ = 78) from all 22 arteries exhibited contractile responses to 45 mM KCl (5.6 ± 0.6 mN). Ring segments from additional arteries which did not respond to KCl were not utilised further.

Of the 22 patients, segments obtained from 10 of the arteries contracted to 5-HT (up to the maximum concentration tested i.e. 60 μM, '5-HT responders'), whilst segments from the remaining 12 arteries showed no response. Some segments showed considerable spontaneous phasic contractile activity, whereas others did not. In the former, agonist responses were characterised by an increase in height of the phasic tension wave. In the latter, agonist responses were characterised by a rise in baseline tension ('tonic' contraction). This phenomenon has been previously described [18]. Examples of the contractile responses are shown in Figure 1.

Of the 10 arteries which responded to 5-HT ('5-HT responders'), individual segments from three arteries were tested for responses to MK-462 alone and all of these three arteries were found to contract to MK-462. The remaining seven ('5-HT responders') arteries were tested for responses both to MK-462 and to sumatriptan (i.e. two segments from each artery were used, one segment being exposed to MK-462 and one segment to sumatriptan); of these, four contracted in response to both drugs, two contracted to sumatriptan but not to MK-462, and one contracted to MK-462 but not to sumatriptan. Mean concentration-effect curves to each agonist are shown in Figure 2. One-way analysis of variance for agonist treatments made on the slope-coefficients for the concentration-effect curves (obtained from tissues which were exposed to the three agonists, see Table 1) showed a significant effect of agonist treatment ($F$ = 7.67, d.f. = 2,12, $P$ = 0.007) and individual comparisons showed that the curve for MK-462, but not for sumatriptan, was significantly different from that for 5-HT (MK-462 *vs* 5-HT; $F$ = 38.27, d.f. = 1,6, $P$ = 0.0008; sumatriptan

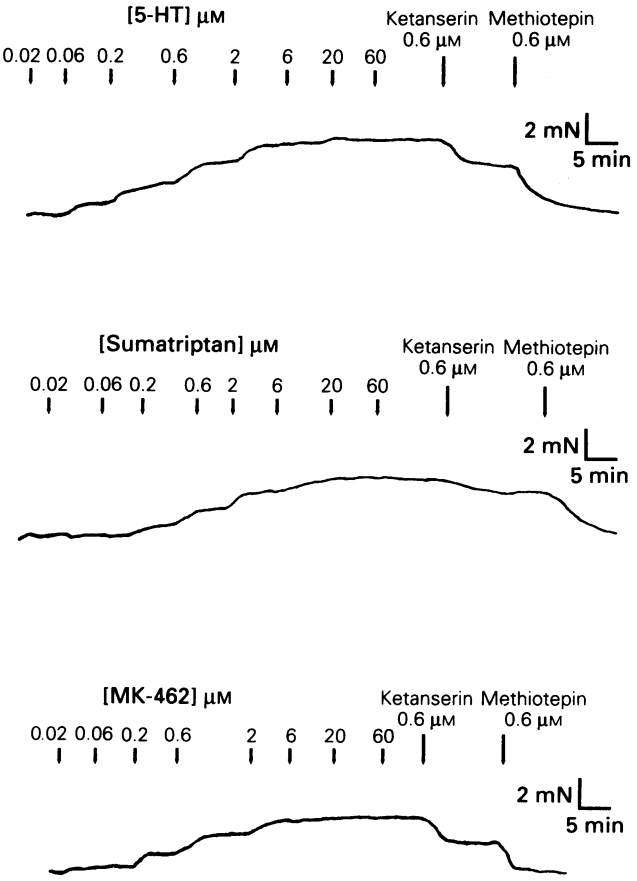


**Figure 1** Chart records of the contractile response of ring segments of human isolated coronary artery evoked by cumulative addition of a) 5-HT, b) sumatriptan and c) MK-462. Examples are taken from patients 14, 10 and 3 respectively (see Table 1). The effect of ketanserin and methiothepin is also shown.

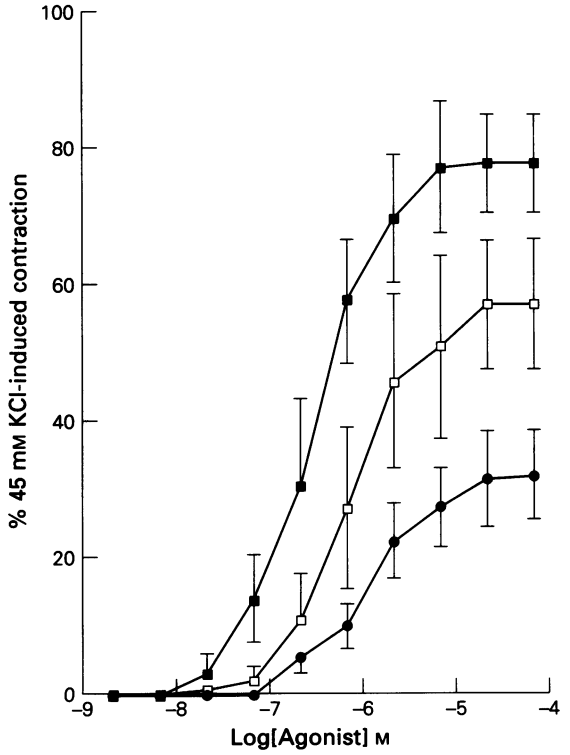


**Figure 2** Ring segments of human coronary arteries were set up in organ baths, as described in Methods, and concentration-effect curves were determined to 5-HT (closed squares; $n = 10$), sumatriptan (open squares; $n = 6$) and MK-462 (closed circles; $n = 8$). Ring segments which did not respond to the agonists are not included in this figure. Values plotted are mean ± s.e. mean, expressed as a percentage of the contractile force induced by 45 mM KCl.

*vs* 5-HT: $F = 2.16$, d.f. = 1,6, $P = 0.386$) and that the curve for MK-462 was not significantly different from that for sumatriptan ($F = 3.61$, d.f. = 1,6, $P < 0.106$). Comparisons were also made using Student's *t*-test between the $pEC_{50}$ values and the $E_{max}$ values for the three agonists in tissues which showed a contractile response. The $pEC_{50}$ values were not significantly different for the three agonists (Table 1); however, there were significant differences in the $E_{max}$ values for the three agonists. The rank order of efficacy was 5-HT > sumatriptan > MK-462 ($n$ = 10, 6 and 8 respectively see Table 1). Comparison of $E_{max}$ values obtained from these segments showed that the $E_{max}$ values for both sumatriptan and MK-462 were significantly less than that for 5-HT ($P < 0.001$ and $P < 0.03$ respectively), and $E_{max}$ for MK-462 was significantly less than that for sumatriptan ($P < 0.04$). The maximum response evoked by 5-HT was 78.0% (95% CI = 63.9-92.1%) of the response elicited by 45mM KCl.

The maximal contractions elicited to each of the three agonists were reduced upon exposure to 0.6 μM ketanserin. Upon further addition of 0.6 μM methiotepin, the contractile responses were further reduced or completely abolished (Figure 3).

## Discussion

Previous studies have suggested that the contractile response of human coronary artery to 5-HT is mediated through $5\text{-HT}_{2}$- and $5\text{-HT}_{1D}$-receptor activation [2, 7, 17]. This latter suggestion is consistent with observations made in the present study where the maximum response (mean data) to sumatriptan (a selective $5\text{-HT}_{1D}$-receptor agonist) was approximately

**Table 1** Potencies and efficacies of 5-HT, MK-462 and sumatriptan on 5-HT-responsive coronary arteries

| | *5-HT* | | *MK-462* | | *Sumatriptan* | |
|---|---|---|---|---|---|---|
| *Patient* | $pEC_{50}$ | $E_{max}$ | $pEC_{50}$ | $E_{max}$ | $pEC_{50}$ | $E_{max}$ |
| 3 | 6.33 | 101.0 | 5.93 | 17.0 | NT | NT |
| 6 | 6.42 | 94.7 | 5.80 | 70.3 | NT | NT |
| 7 | 6.19 | 52.4 | 5.80 | 20.8 | NT | NT |
| 10 | 6.34 | 98.1 | 5.44 | 20.3 | 5.80 | 62.9 |
| 11 | 7.30 | 101.0 | 6.20 | 13.3 | 6.19 | 87.8 |
| 12 | 7.30 | 62.3 | No response | | 6.64 | 74.7 |
| 14 | 6.10 | 63.5 | 6.11 | 37.7 | 6.22 | 40.2 |
| 16 | 6.77 | 100.0 | 6.54 | 44.1 | 6.77 | 56.3 |
| 20 | 5.78 | 43.7 | No response | | 5.94 | 23.7 |
| 22 | 6.10 | 63.5 | 6.15 | 36.4 | No response | |
| Grand mean | 6.46 | 78.0 | 6.00 | 32.5 | 6.26 | 57.6 |
| 95% CI | 6.15–6.62 | 63.9–92.1 | 5.77–6.23 | 19.4–45.6 | 5.95–6.57 | 39.0–76.2 |
| | ($n$ = 10) | | ($n$ = 8) | | ($n$ = 6) | |
| Mean | 6.63 | 90.7 | 6.0 | 28.9 | 6.25 | 61.8 |
| 95% CI for vessels responding to all agonists | 6.13–7.14 | 72.9–108.5 | 5.62–6.52 | 14.8–43.0 | 5.86–6.64 | 49.9–73.7 |
| | ($n$ = 4) | | ($n$ = 4) | | ($n$ = 4) | |

NT: not tested.
$E_{max}$: maximal contraction expressed as a percentage of contraction induced by 45 mM KCl.
$pEC_{50}$: negative logarithm of drug concentration required to produce half-maximal contraction.

70% of the maximum response evoked by 5-HT (although it should be noted that direct comparison of the agonists was not made on the same segments of artery due to possible desensitization effects). In the present study, the $E_{max}$ for sumatriptan relative to 5-HT was variable (see Table 1) probably reflecting differences in the contribution of 5-$HT_2$ receptor activation to the 5-HT-induced contractile response. Indeed, Kaumann *et al.* [17] showed considerable inter-patient variability in the contribution of 5-$HT_{1\ like}$- and 5-$HT_2$-receptor activation.

In our experiments, the contractile effects of 5-HT were found to be partially reversed by ketanserin and were further reduced, or even abolished, by further addition of methiotepin. It has previously been thought that ketanserin is a selective 5-$HT_2$-receptor antagonist, so that blockade by this drug reflects the involvement of 5-$HT_2$-receptors. However, it has recently been shown that ketanserin also has some affinity for the human 5-$HT_{1D}$-receptor [17]. This property could explain some or even all of the effects of ketanserin in reversing 5-HT-induced vasoconstriction in our experiments. This conclusion is supported by our finding that ketanserin also partially reversed the contractions evoked in response to sumatriptan and MK-462, drugs with little affinity for 5-$HT_2$-receptors [9, 15]. Ketanserin has been reported to have approximately 20-70 fold higher affinity for the 5-$HT_{1D\alpha}$-receptor subtype over the 5-$HT_{1D\beta}$-receptor subtype [17] and since for human cerebral blood vessels ketanserin (1μM) has no effect on 5-$HT_{1D}$-agonist-induced contractile responses, it has been suggested that these responses are mediated through the 5-$HT_{1D\beta}$-receptor subtype [17]. In our study, ketanserin (0.6 μM) reduced contractions to 5-HT, sumatriptan and MK-462 which implies the involvement of the 5-$HT_{1D\alpha}$-receptor subtype. However, we cannot eliminate the involvement of 5-$HT_2$-receptors (particularly in the case of 5-HT) and furthermore ketanserin has been shown to possess calcium channel blocking properties [19] which may also contribute to the reduction in contractions evoked by the three agonists [18, 20]. It should be noted, that Kaumann *et al.* [17] reported that in human coronary artery contractile responses to sumatriptan were insensitive to ketanserin (0.1 μM). It is possible that differences in the concentrations of ketanserin used and in experimental methodology may account for the differences in ketanserin-sensitivity observed in the study by Kaumann *et al.* [17] and the present results. Nevertheless, our results with 5-HT and sumatriptan confirm the occurrence of 5-$HT_{1D}$-receptor-mediated vasoconstriction in human coronary arteries, and do not exclude completely a 5-$HT_2$-receptor-mediated component.

One of the most interesting of our findings was that not all vessels responded to 5-HT; out of 22 coronary arteries tested, only 10 responded. From the relatively small number of arteries used there was no apparent relation between responsiveness to 5-HT and disease state (including presence or absence of atheroma), drug therapy or patient age. The precise reason why some vessels responded and others did not is unknown. Lack of 5-HT responsiveness did not seem to be due to poor viability of the tissues, since repeated and reproducible contractions to 45 mM KCl could be elicited in these vessels. Furthermore, the magnitude of the KCl responses was not different between 5-HT responding and non-responding tissues.

Sumatriptan has been developed as a treatment for acute migraine and, whilst its precise mechanism of action in this disorder remains to be established, it probably involves stimulation of 5-$HT_{1D}$-receptors in intracranial blood vessels [21]. However, sumatriptan can also give rise to coronary artery vasoconstriction. Recent clinical trials have shown that MK-462 (10mg p.o.), a novel 5-$HT_{1D}$-receptor agonist, is as effective as sumatriptan (100mg p.o.) in relieving migraine headache [16]. We therefore compared the coronary

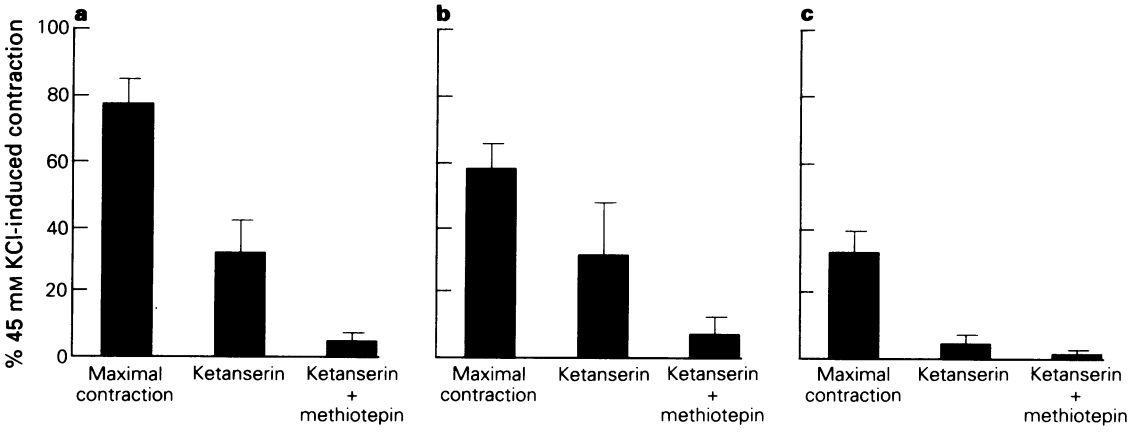


**Figure 3** Following maximal stimulation of coronary artery rings with agonist, 0.6 μM ketanserin and subsequently 0.6 μM methiotepin were added, and the effect on vascular tension observed, as outlined in **Methods**. Values plotted are mean ± s.e. mean ($n$ = 10, to include all 'responders' to 5-HT), expressed as a percentage of the contractile force induced by 45 mM KCl. Effects of sequential addition of ketanserin and methiotepin are shown on maximal responses to a) 5HT, b) sumatriptan and c) MK-462.

vasoconstrictor effects of MK-462 with those of sumatriptan and of 5-HT. To avoid possible desensitization effects each arterial segment was exposed to a single agonist concentration-effect curve. Our results suggest that MK-462, whilst equipotent to sumatriptan (in terms of $pEC_{50}$), is less efficacious than either sumatriptan or 5-HT in causing coronary vasoconstriction. The maximum contractile response evoked by 5-HT was similar in magnitude to that produced by 45 mM KCl, sumatriptan was less efficacious and MK-462 had relatively weak efficacy. The efficacy values obtained from all arterial segments which responded to either 5-HT, sumatriptan or MK-462 were similar to the values obtained from the four arteries which responded to all three agonists ($n$ = 4). The relative efficacies and $pEC_{50}$ values for 5-HT and sumatriptan obtained in the present study were similar to those previously reported [2, 7].

The reason for the difference between the efficacy of sumatriptan and MK-462 is not clear. *In vitro* functional assays using rabbit isolated saphenous vein have shown that MK-462, similarly to sumatriptan, is a full agonist with respect to 5-HT [15, 22, Hargreaves *et al.*, unpublished observations]. However, it is noteworthy that the coronary arteries in our experiments displayed some heterogeneity in their responses to sumatriptan and to MK-462. Two of the vessels which received all three agonists responded to 5-HT and to sumatriptan but not to MK-462, one artery responded to 5-HT and to MK-462 but not to sumatriptan, and four arteries responded to all three drugs. This may simply be due to differences in receptor density and/or coupling between different strips of the same vessel. Alternatively, it may be that sumatriptan and MK-462 have different affinities and/or efficacies at different subtypes of $5\text{-}HT_{1D}$-receptor, and that different coronary arteries express different subpopulations of $5\text{-}HT_{1D}$-receptor or may reflect actions at a so far unstudied member of the 5-HT receptor family. Such speculation should be clarified by molecular biological and radioligand binding studies, and their correlation with pharmacological responses, in such vessels.

Plasma level of sumatriptan achieved at normal therapeutic doses is 72.4 ng $ml^{-1}$ (range 54.9-108 ng $ml^{-1}$) following a 6mg subcutaneous injection [11], (corresponding to approximately 0.2 μM (range 0.15-0.3 μM)). These plasma levels are 2–3 fold lower than the $EC_{50}$ values obtained for sumatriptan in human isolated coronary artery (0.5 and 1.0 μM respectively, present study). However, in the angiographic study of MacIntyre *et al.* [11] higher concentrations of sumatriptan (i.e. 156 ng $ml^{-1}$ (range (94-223 ng $ml^{-1}$) corresponding to 0.43 μM (range 0.26-0.62 μM) were achieved more closely resembling the $EC_{50}$ values obtained on isolated coronary arteries. Furthermore, it has been recently reported that sumatriptan (6 mg s.c.) exerts vasopressor effects in the systemic and pulmonary arterial circulation [23]. It should be noted that the concentration of sumatriptan required to cause contraction of isolated coronary arterial segments is higher than the plasma level observed in the treatment of migraineurs.

In conclusion, we have found that human coronary arteries display heterogeneity in the presence or absence of contractile responses to 5-HT, which does not seem to be related to patient age, drug therapy or disease state. Where such responses occur, they are operated through $5\text{-}HT_{1D}$-receptors, and there may also be a $5\text{-}HT_2$-receptor component. Arteries which responded to 5-HT also tended to contract in response to sumatriptan and MK-462 and the results indicate that MK-462 could be less effective than sumatriptan in causing contraction of human coronary artery.

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## References

1 Ichikawa Y, Yokoyama M, Akita H, Fukuzaki H. Constriction of a large coronary artery contributes to serotonin-induced myocardial ischemia in the dog with pliable coronary stenosis. *J Am Coll Cardiol* 1989; **14**: 449–459.

2 Chester AH, Martin GR, Bodelsson M, *et al.* 5-Hydroxytryptamine receptor profile in healthy and diseased human epicardial coronary arteries. *Cardiovasc Res* 1990; **24**: 932–937.

3 Golino P, Piscioni F, Willerson JT, *et al.* Divergent effects of serotonin on coronary artery dimensions and blood flow in patients with coronary atherosclerosis and control patients. *N Engl J Med* 1991; **324**: 641–648.

4 McFadden EP, Clarke JG, Davies GJ, Kaski JC, Haider AW, Maseri A. Effect of intracoronary serotonin on coronary vessels in patients with stable angina and patients with variant angina. *N Engl J Med* 1991; **324**: 648–654.

5 Berkenboom G, Unger P, Fang ZY, Degre S, Fontaine J. Comparison of responses to acetylcholine and serotonin on isolated canine and human coronary arteries. *Cardiovasc Res* 1989; **23**: 780–787.

6 Toda N, Okamura T. Comparison of the response to 5-carboxamidotryptamine and serotonin in isolated human, monkey and dog coronary arteries. *J Pharmacol Exp Ther* 1990; **253**: 676–682.

7 Connor HE, Feniuk W, Humphrey PPA. 5-Hydroxytryptamine contracts human coronary arteries predominantly via $5\text{-}HT_2$ receptor activation. *Eur J Pharmacol* 1989; **161**: 91–94.

8 Golino P, Piscione F, Benedict CR, *et al.* Local effect of serotonin released during coronary angioplasty. *N Engl J Med* 1994; **330**: 523–528.

9 Humphrey PPA, Feniuk W, Perren MJ, Connor HE, Oxford AW. The pharmacology of the novel $5\text{-}HT_1$-like receptor agonist, GR43175. *Cephalalgia* 1989; **9**: (Suppl. 9) 23–33.

10 Saxena PR, Ferrari MD. $5\text{-}HT_1$-like receptor agonists and the pathophysiology of migraine. *Trends Pharmacol Sci* 1989; **10**: 200–204.

11 MacIntyre PD, Bhargave B, Hogg KJ, Gemmill JD, Hillis WS. The effect of i.v. sumatriptan, a selective 5-HT-1-receptor agonist, on central haemodynamics and the coronary circulation. *Br J Clin Pharmacol* 1992; **34**: 541–546.

12 Willet F, Curzen N, Adams J, Armitage M. Coronary vasospasm induced by subcutaneous sumatriptan. *Br Med J* 1992; **304**: 1415.

13 Lloyd DK, Simmons V. Adverse reactions associated with sumatriptan. *Lancet*, 1993; **341**: 1092.

14 Ottervanger JP, Paalman JJA, Boxma GL, Stricker BHC. Transmural myocardial infarction with sumatriptan. *Lancet* 1993; **341**: 861–862.

15 Beer M, Middlemiss D, Stanton J, *et al.* An *in vitro* profile of the novel antimigraine 5-HT-$_{1D}$ receptor agonist MK-462. *Proceedings of Third IUPHAR meeting on serotonin* 1994; Abstract 98.

16 Ferrari M, Terwindt GM, Visser WH, Reines SA. A placebo-controlled dose-ranging study of MK-462 versus sumatriptan in migraine. In *New Advances in Headache Research: 4*, ed. Rose FC, London: Smith Gordon, 1994: 205.

17 Kaumann AJ, Frenken M, Posival H, Brown AM. Variable participation of 5-HT$_1$-like receptors and 5-HT$_2$ receptors in serotonin-induced contraction of human isolated coronary arteries. *Circulation* 1994; **90**: 1141–1153.

18 Cocks TM, Kemp BK, Pruneau D, Angus JA. Comparison of contractile responses to 5-hydroxytryptamine and sumatriptan in human isolated coronary artery: synergy with the thromboxane A$_2$-receptor agonist, U46619. *Br J Pharmacol* 1993; **110**: 360–368.

19 Razzaque Z, Longmore J, Hill RG. Differences in the effects of ketanserin on 5 HT-receptor mediated responses in rabbit saphenous vein and guinea-pig jugular vein. *Br J Pharmacol* 1994; **112**: 474P.

20 Sumner MJ, Feniuk W, McCormick JD, Humphrey PPA. Studies on the mechanism of 5-HT1 receptor-induced smooth muscle contraction in dog saphenous vein. *Br J Pharmacol* 1992; **105**: 603–608.

21 Humphrey PPA, Feniuk W. Mode of action of the antimigraine drug sumatriptan. *Trends Pharmacol Sci* 1991; **12**: 444–446.

22 Martin GR, MacLennan SJ. Analysis of 5-HT receptors in rabbit saphenous vein exemplifies the problems of using exclusion criteria for receptor classification. *Arch Pharmacol* 1990; **342**: 111–119.

23 Hood S, Birnie D, Docherty A, Pell A, Murdoch D, Hillis WS. The central haemodynamic effects of subcutaneous sumatriptan. *Br J Clin Pharmacol* 1995; **39**: 96P.

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