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BMS-181885, a 5-$HT_{1B/1D}$ receptor ligand, in experimental models predictive of antimigraine activity and coronary side-effect potential

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Abstract

Many acutely acting antimigraine drugs have the ability to constrict porcine arteriovenous anastomoses as well as the human isolated coronary artery. These two experimental models seem to serve as indicators, respectively, for the therapeutic and coronary side-effect potential of the compounds. Using these two models, we have investigated the effects of BMS-181885 (3-[3-[4-(5-methoxy-4-pyrimidyl)-1-piperazinyl]propyl]-5-(1,2-dioxo-4-methyl-3-cyclobuten-3-yl)amino-1*H*-indole), a 5-$HT_{1B/1D}$ receptor ligand. In anaesthetised pigs, BMS-181885 (10, 30, 100 and 300 μg kg^{-1}) decreased the total carotid blood flow and conduction, exclusively at the expense of the arteriovenous anastomotic fraction as the capillary fraction did in fact increase. The highest dose (300 μg kg^{-1}) produced a reduction of $52 \pm 6\%$ from the baseline arteriovenous anastomotic flow. When carotid haemodynamic changes after a single 100 μg kg^{-1} dose of BMS-181885 or sumatriptan were studied at different time-points, BMS-188185 had a longer duration of action. Both BMS-181885 (pD_2: 7.9 ± 0.1; E_{max}: $9 \pm 3\%$ of the contraction to 100 mM K^+) and sumatriptan (pD_2: 6.3 ± 0.1; E_{max}: $28 \pm 8\%$ of the contraction to 100 mM K^+) contracted the human isolated coronary artery. The above results suggest that (i) the longer-lasting vasoconstrictor action of BMS-181885 on porcine carotid arteriovenous anastomoses may be related to its reported slow dissociation from 5-$HT_{1B/1D}$ receptor, and (ii) BMS-181885 should be able to abort migraine headaches in patients. It will be interesting to find out whether these properties are clinically important so that the drug exhibits less headache recurrence and coronary side-effects than sumatriptan.

Keywords: Antimigraine drug; Arteriovenous anastomosis; BMS-181885; Carotid artery; (Human); Coronary artery, human; Migraine; (Pig); Sumatriptan

1. Introduction

Sumatriptan is the first member of a completely new class of compounds (Humphrey et al., 1988; Saxena and Ferrari, 1989; Humphrey et al., 1990), which are now designated as 5-$HT_{1B/1D}$ receptor agonists. Sumatriptan is very effective in aborting migraine headaches (Ferrari, 1991; Ferrari and Saxena, 1993). The drug constricts large cranial and extracranial blood vessels (Saxena and Tfelt-Hansen, 1993), including bovine and human cerebral arteries (Hamel and Bouchard, 1991; Hamel et al., 1993), canine saphenous vein (Humphrey et al., 1988, 1990) and porcine carotid arteriovenous anastomoses (Den Boer et al., 1991a, 1992). The receptors mediating the vasoconstrictor effect of sumatriptan appear to be identical to recombinant 5-$HT_{1B/1D}$ receptors. Indeed, sumatriptan has a high affinity for these receptors (Peroutka and McCarthy, 1989; Beattie et al., 1994) and its vasoconstrictor response is antagonised by GR127935 (*N*-[methoxy-3-(4-methyl-1-piperazinyl)phenyl]-2′-methyl-4′-(5-methyl-1,2,4-oxadiazol-3-yl) [1,1-biphenyl]-4-carboxamide hydrochloride), a selective 5-$HT_{1B/1D}$ receptor antagonist (Clitherow et al., 1994; De Vries et al., 1996, 1997; Pauwels, 1996; Skingle et al., 1996). Although selective ligands at 5-HT_{1B} or 5-HT_{1D} receptors are not yet available, the sumatriptan-induced vasoconstriction is most likely mediated by the 5-HT_{1B} receptor, since mRNA for the 5-HT_{1B} but not 5-HT_{1D} receptor has been located in cranial blood vessels (Hamel and Bouchard, 1991; Hamel et al., 1993; Bouchelet et al., 1996).

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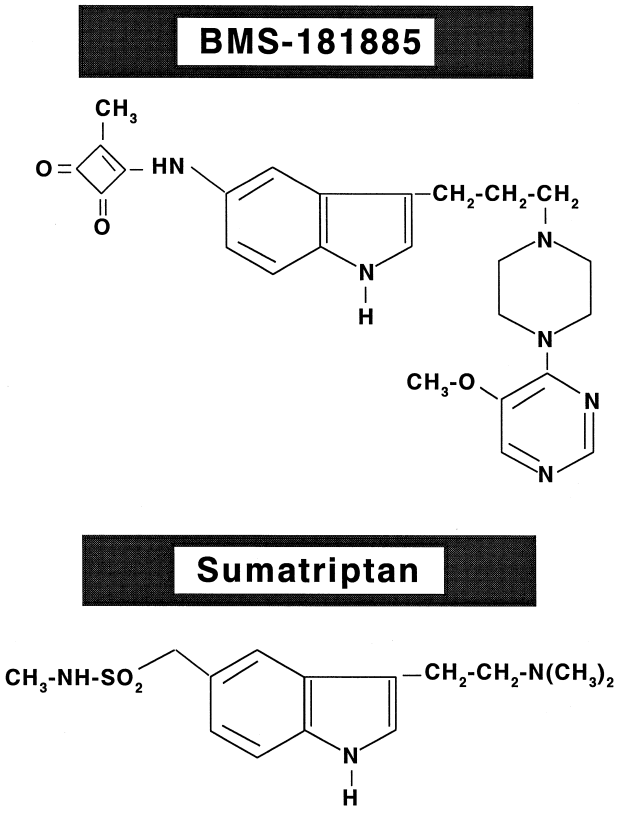

Fig. 1. Chemical structures of BMS-181885 (trihydrochloride salt; MW 569) and sumatriptan (succinate salt; MW 413).

The successful introduction of sumatriptan in antimigraine therapy has prompted the development of new 5-$HT_{1B/1D}$ receptor agonists with the aim of avoiding some of the drawbacks of sumatriptan, notably the coronary vasoconstriction and headache recurrence (Saxena and Ferrari, 1996). BMS-181885 (3-[3-[4-(5-methoxy-4-pyrimidyl)-1-piperazinyl]propyl]-5-(1,2-dioxo-4-methyl-3-cyclobuten-3-yl)amino-1*H*-indole; Fig. 1), a clinically effective migraine abortive agent (Unpublished data, Bristol-Myers Squibb files), displays high and selective affinities at 5-$HT_{1B/1D}$ receptors (Table 1). Interestingly, kinetic analyses revealed that BMS-181885 has a slow dissociation rate from 5-$HT_{1B/1D}$ receptors and, unlike sumatriptan and other such agents, it does not constrict isolated canine saphenous vein or guinea pig pressurised iliac artery, demonstrating competitive antagonism at some peripheral blood vessels (Yocca et al., 1997).

In the present investigation, we studied the effects of BMS-181885 in experimental models predictive of therapeutic activity in migraine i.e., constriction of carotid arteriovenous anastomoses in anaesthetised pigs (Saxena, 1990, 1995) and potential coronary side-effects, i.e., constriction of human isolated coronary artery (Connor et al., 1989; Chester et al., 1990; Bax et al., 1992; Bax and Saxena, 1993; MaassenVanDenBrink et al., 1998). The effects of BMS-181885 have been compared with those of sumatriptan to establish whether the drug has a longer duration of action and elicits less or no contraction of the human isolated coronary artery. Some of the results of this investigation have been reported at the 8th International Headache Congress (Yocca et al., 1997).

2. Materials and methods

2.1. Systemic and carotid haemodynamics in anaesthetised pigs

2.1.1. General

After an overnight fast, 19 domestic pigs (Yorkshire × Landrace; 10–15 kg) were anaesthetised with azaperone (160 mg, i.m.), midazolan hydrochloride (5 mg, i.m.) and metomidate (200 mg, i.v.), intubated and connected to a respirator (BEAR 2E, BeMeds, Baar, Switzerland) for intermittent positive pressure ventilation with a mixture of room air and oxygen. Respiratory rate, tidal volume and oxygen supply were adjusted to keep arterial blood gas values within physiological limits (pH: 7.35–7.48; pCO_2: 35–48 mm Hg; pO_2: 100–120 mm Hg). Anaesthesia was maintained with a continuous i.v. infusion of pentobarbitone sodium at 20 mg kg^{-1} h^{-1}. With this anaesthetic regimen, arteriovenous anastomotic blood flow is considerably higher than that in pigs in a conscious state or under thiopentone anaesthesia (Den Boer et al., 1993).

Catheters were placed in the inferior vena cava via the left femoral vein for the administration of drugs and in the aortic arch via the left femoral artery for the measurement of arterial blood pressure (Combitrans disposable pressure transducer; Braun, Melsungen, Germany) and the withdrawal of arterial blood for determining blood gases (ABL-510, Radiometer, Copenhagen, Denmark).

The common carotid arteries, external jugular veins and vagus nerves were identified. After ligation, both vagi and the accompanying cervical sympathetic nerves were cut and a catheter was placed in the right external jugular vein for the withdrawal of venous blood samples. The right common carotid artery was dissected free and a needle was inserted against the direction of blood flow for the admin-

Table 1
Binding affinities (pK_i values) of BMS-181885 and sumatriptan at 5-HT receptors

	5-HT_{1A} (Rat)	5-HT_{1B} (Rat)	5-HT_{1B} (Human)	5-HT_{1D} (Human)	5-HT_2 (Rat)	5-HT_3 (Rat)
BMS-181885	6.42	8.67	9.04	8.70	< 6.0	< 6.0
Sumatriptan	7.00	7.57	7.54	8.14	< 5.0	< 5.0

Data from the work of Yocca et al. (1997) and Bristol-Myers Squibb files, except for sumatriptan on rat receptors (Peroutka and McCarthy, 1989).

istration and uniform mixing of radioactive microspheres. Blood flow was measured in the right common carotid artery with a flow probe (internal diameter: 2.5 mm) connected to a sine-wave electromagnetic flow meter (Transflow 601-system, Skalar, Delft, The Netherlands). Heart rate was measured with a tachograph (CRW, Erasmus University, Rotterdam, the Netherlands) triggered by electrocardiogram signals. Arterial blood pressure, heart rate and carotid blood flow were continuously monitored on a polygraph (CRW, Erasmus University). During the experiment body temperature was kept at about 37°C and the animals were continuously infused with saline to compensate for fluid losses.

2.1.2. Distribution of carotid blood flow

The distribution of common carotid blood flow was determined with 15 ± 0.1 (S.D.) μm diameter microspheres labelled with either ^{141}Ce, ^{113}Sn, ^{95}Nb, ^{103}Ru or ^{46}Sc (NEN Dupont, Boston, USA). For each measurement a suspension of about 200 000 microspheres, labelled with one of the isotopes, was mixed and injected into the carotid artery. At the end of the experiment, the animal was killed and the heart, kidneys, lungs and the different cranial tissues were dissected out, weighed and put in vials. The radioactivity in these vials was counted for 5–10 min in a γ-scintillation counter (Packard, Minaxi autogamma 5000), using suitable windows for discriminating the different isotopes. All data were processed by a set of specially designed programs (Saxena et al., 1980), using a personal computer.

The fraction of carotid blood flow distributed to the different tissues was calculated by multiplying the ratio of tissue and total radioactivities by the total common carotid blood flow at the time of the injection of microspheres. Since little or no radioactivity was detected in the heart and kidneys, all microspheres trapped in lungs reached this tissue from the venous side after escaping via carotid arteriovenous anastomoses. Therefore, the amount of radioactivity in the lungs was used as an *index* of the arteriovenous anastomotic fraction of carotid blood flow (Saxena and Verdouw, 1982).

2.1.3. Experimental protocols and calculations

The experiments were started after a stabilization period of about 1 h. At baseline, heart rate, mean arterial blood pressure, carotid blood flow and its distribution as well as arterial and jugular venous blood gases were measured. Thereafter, two series of experiments were performed. In the first series of experiments, the animals ($n = 7$) received cumulative i.v. doses of BMS-181885 (10, 30, 100 and 300 μg kg^{-1}) every 20 min, a time interval which was arbitrarily selected. Exactly 15 min after each dose of the compounds, the haemodynamic variables were assessed again. In the second series of experiments, the animals received a single i.v. dose (100 μg kg^{-1}) of either BMS-181885 ($n = 6$) or sumatriptan ($n = 6$). The haemodynamic variables were assessed again 30, 60, 90 and 120 min after the drug administration. The changes from the baseline values caused by BMS-181885 (10, 30, 100 or 300 μg kg^{-1}) or by a single dose of 100 μg kg^{-1} of

Table 2
Changes in heart rate (HR), mean arterial blood pressure (MABP) and the difference in arterial and jugular venous oxygen saturations (A–V O_2 dif) after cumulative doses of BMS-181885 ($n = 7$) and at different time-points after 100 μg kg^{-1} of BMS-181885 or sumatriptan ($n = 6$ each) in anaesthetised pigs

	BMS-181885 (μg kg^{-1})				
	Baseline	10	30	100	300
HR (beats min^{-1})	113 ± 4	111 ± 4	109 ± 4[a]	108 ± 4[a]	106 ± 4[a]
MABP (mmHg)	95 ± 4	97 ± 5	97 ± 4	94 ± 5	88 ± 6[a]
A–V O_2 dif (%)	14.6 ± 3.2	19.1 ± 4.2[a]	22.2 ± 4.0[a]	24.2 ± 3.8[a]	25.5 ± 3.8[a]
	Time (min) after 100 μg kg^{-1}				
	Baseline	30	60	90	120
HR (beats min^{-1})					
BMS-188185	111 ± 4	100 ± 4[a]	97 ± 3[a]	95 ± 4[a]	93 ± 4[a]
Sumatriptan	108 ± 2	102 ± 3[a]	99 ± 3[a]	97 ± 3[a]	95 ± 2[a]
MABP (mmHg)					
BMS-181885	83 ± 1	85 ± 4	84 ± 5	84 ± 6	83 ± 7
Sumatriptan	86 ± 3	86 ± 4	83 ± 5	83 ± 5	83 ± 4
A–V O_2 dif (%)					
BMS-181885	13.6 ± 3.7	23.8 ± 5.4[a]	23.1 ± 6.5[a]	21.4 ± 5.6[a]	23.6 ± 7.5[a]
Sumatriptan	17.7 ± 4.5	20.9 ± 6.3	19.1 ± 5.9	18.6 ± 5.9	18.2 ± 5.4

All values are presented as means ± S.E.M.
[a]Significant difference ($P < 0.05$) from the corresponding baseline value.
[b]Significant difference ($P < 0.05$) from the change elicited by sumatriptan at the corresponding time-point.

BMS-181885 or sumatriptan at different time-points were calculated.

2.2. Human isolated coronary artery

2.2.1. Tissue preparation

The hearts, provided by the Rotterdam Heart Valve Bank (Bio Implant Services/Eurotransplant Foundation), were stored at 0–4°C in a sterile organ protecting solution (UW, EuroCollins, or HTK-Brettschneider) immediately following circulatory arrest (Ploeg et al., 1992). They were obtained from six heart beating organ donors (four males, two females; age: 11–44 years; mean age 33 ± 5 years), who died of non-cardiac disorders (three cerebrovascular accidents, one trauma, two hypoxia) less than 24 h before the tissue was taken to the laboratory.

After arrival in the laboratory, the right epicardial coronary artery was removed and placed in a cold, oxygenated Krebs bicarbonate solution of the following composition: 118 mM sodium chloride, 4.7 mM potassium chloride, 2.5 mM calcium chloride, 1.2 mM magnesium sulphate, 1.2 mM potassium dihydrogenphosphate, 25 mM sodium bicarbonate and 8.3 mM glucose; pH 7.4. As described previously (Bax et al., 1993), the vessel was cut into rings of approximately 4 mm of length and suspended on stainless steel hooks in 15 ml organ baths containing the Krebs bicarbonate solution, aerated with 95% O_2 and 5% CO_2 and maintained at 37°C. Vessel segments containing macroscopically visible atherosclerotic lesions were not used. The segments were allowed to equilibrate for at least 30 min and washed every 15 min. Changes in tension were recorded using a Harvard isometric transducer. Preparations were stretched to a stable pretension of 20 mN. The tissue was exposed to 30 mM K^+ twice. Subsequently, the functional integrity of the endothelium was verified by observing relaxation to substance P (1 nM) after pre-contraction with prostaglandin $F_{2\alpha}$ (1 μM). After washout, the tissue was exposed to 100 mM K^+ to determine the maximal contractile response to K^+. The tissue was then allowed to equilibrate in the Krebs solution for a period of 30 min.

2.2.2. Experimental protocol and calculations

After equilibration, a cumulative concentration response curve to BMS-181885 or sumatriptan was obtained in logarithmic steps (0.1 nM–100 μM). Responses were

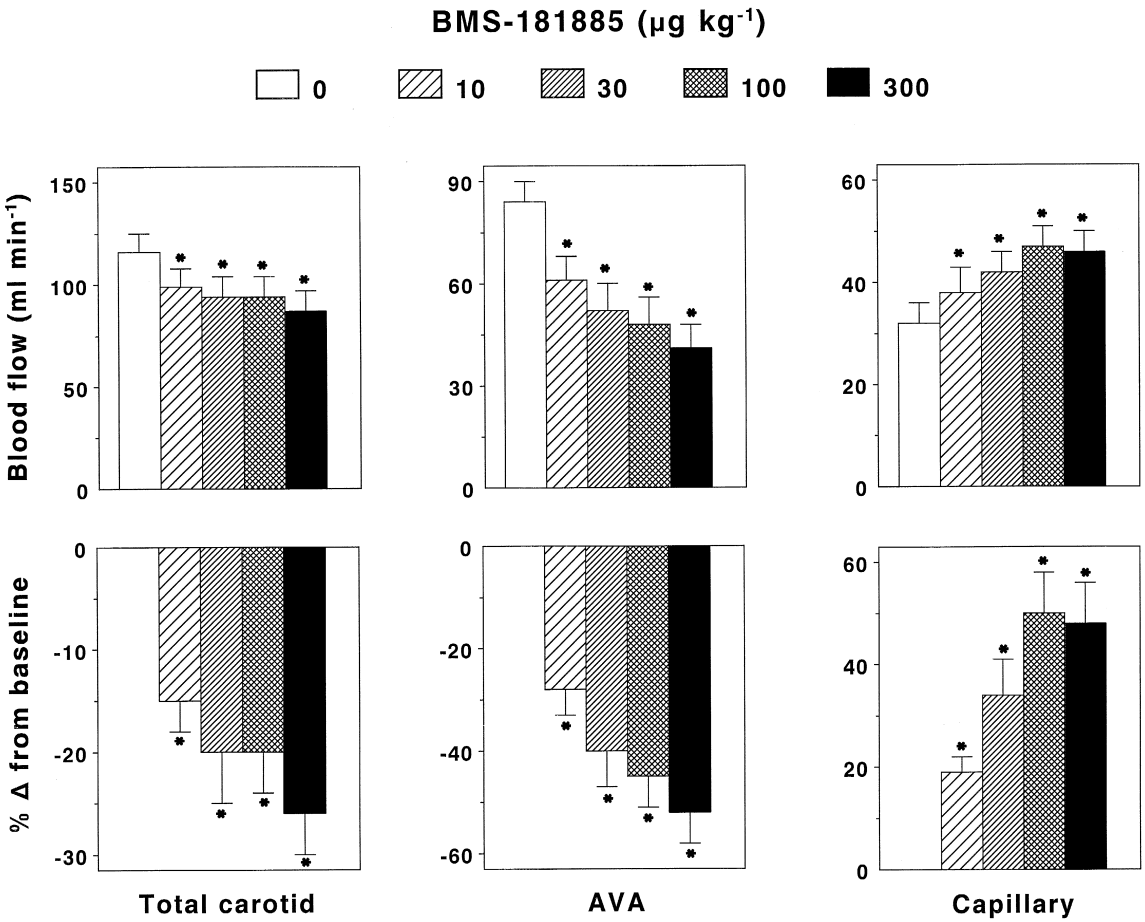

Fig. 2. Effect of BMS-181885 on the total carotid blood flow and its arteriovenous anastomotic (AVA) and nutrient (capillary) fractions in anaesthetised pigs. Upper panels, absolute values; Lower panels, percent change from baseline values. All values are presented as means ± S.E.M. $* P < 0.05$ vs. baseline.

expressed as a percentage of K^+ (100 mM)-induced contractions or in mN. All curves were obtained in a paired, parallel experimental set-up, i.e., both agonists (sumatriptan and BMS-188185) were tested on separate segments from the six coronary arteries. In four cases we used one segment each for the two compounds, but in two hearts one segment was used for BMS-188185 and two segments were used for sumatriptan. In the latter case, the averaged data per artery was used in further analysis.

Curves covering the full sigmoidal range were analyzed by means of a computerised curve fitting technique (De Lean et al., 1978) to obtain pD_2 values ($-\log EC_{50}$, i.e., the negative logarithm of the molar concentration of an agonist needed to reach half of its maximal response, E_{max}), which were averaged for the two agonists.

2.3. Data presentation and statistical comparisons

All data have been expressed as means ± S.E.M. In the haemodynamic study, the significance of the changes (from baseline values) induced by the different doses (10, 30, 100 or 300 $\mu g\ kg^{-1}$) of BMS-181885 and at different time-points (30, 60, 90 or 120 min) after 100 $\mu g\ kg^{-1}$ of BMS-181885 or sumatriptan was evaluated with Duncan's new multiple range test, once an analysis of variance (randomised block design) had revealed that the samples represented different populations (Saxena, 1985). In the isolated coronary artery studies, the significance of difference between the E_{max} and pD_2 values of sumatriptan and BMS-181885 was calculated by Student's paired *t*-test (SPSS/PC + statistical package). Statistical significance was accepted at $P < 0.05$ (two-tailed).

2.4. Compounds

Apart from the anaesthetics, azaperone, metomidate (both from Janssen Pharmaceutica, Beerse, Belgium), midazolan hydrochloride (Hoffmann La Roche, Mijdrecht, the Netherlands) and pentobarbitone sodium (Apharmo, Arnhem, the Netherlands), the compounds used in this study were: prostaglandin $F_{2\alpha}$ Tris salt and substance P acetate (both purchased from Sigma Chemical, St. Louis, USA); sumatriptan succinate (gift: Dr. H. Connor, Glaxo-Welcome Research Laboratories, Stevenage, Herts, UK; MW 413), BMS-181885 (3-[3-[4-(5-methoxy-4-pyrimidyl)-1-piperazinyl]propyl]-5-(1,2-dioxo-4-methyl-3-cyclobuten-3-yl)amino-1*H*-indole tri-hydrochloride; Bristol-Myers Squibb, Wallingford, CT, USA; MW 569) and heparin sodium (Leo Pharmaceutical Products, Weesp, the Netherlands) to prevent clotting of the catheters. For in vitro experiments, all compounds (substance P, prostaglandin $F_{2\alpha}$, sumatriptan and BMS-181885) were dis-

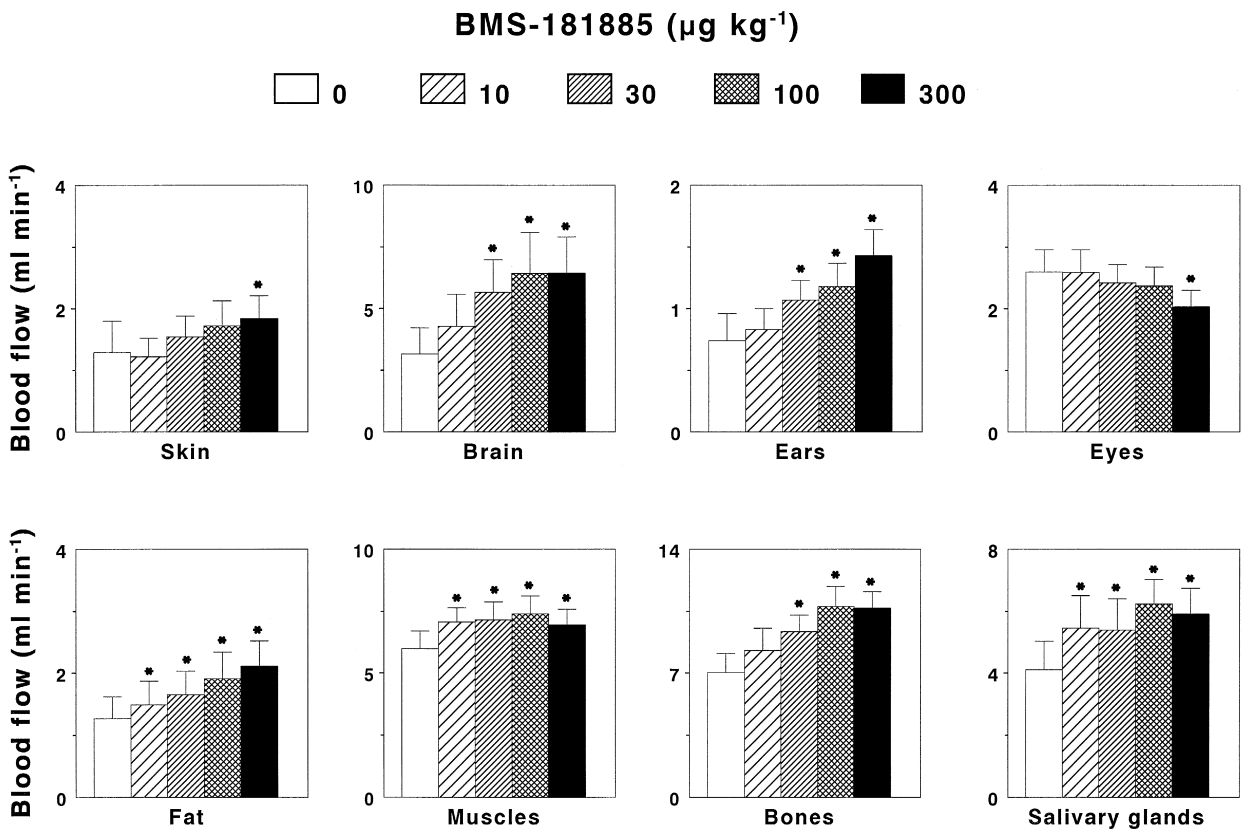

Fig. 3. Effect of BMS-181885 on the distribution of carotid blood flow to different cranial tissues in anaesthetised pigs. All values are presented as means ± S.E.M. * $P < 0.05$ vs. baseline.

solved in distilled water, while for in vivo experiments BMS-181885 was dissolved in physiological saline and all doses refer to the salt.

2.5. Ethical approval

The protocols for the two parts of the investigation were approved by the joint Ethical Committees of the Erasmus University Rotterdam and the University Hospital Rotterdam 'Dijkzigt', dealing with the use of animals and humans in scientific experiments.

3. Results

3.1. Systemic and carotid haemodynamics in anaesthetised pigs

3.1.1. Systemic haemodynamics

As shown in Table 2, heart rate decreased moderately, but significantly, after the three highest doses (30, 100 and 300 μg kg^{-1}, i.v.) of BMS-181885. Mean arterial blood pressure did not change with the first three doses, but decreased moderately with the highest dose. Similarly, in experiments where the effects of a single 100 μg kg^{-1} (i.v.) dose of BMS-181885 or sumatriptan was followed in time, heart rate also decreased moderately with both drugs. Heart rate remained lower throughout the observation period of 2 h. Neither drugs affected arterial blood pressure (Table 2).

3.1.2. Arterio-jugular venous oxygen saturation difference

BMS-181885 elicited an increase in the arterio-jugular venous oxygen saturation difference right from the first dose onwards and this effect was evident even at 120 min after the single 100 μg kg^{-1} dose of BMS-188185. No significant change was observed after the single 100 μg kg^{-1} dose of sumatriptan (Table 2).

3.2. Carotid haemodynamics

The effects of increasing doses of BMS-181885 on carotid haemodynamics in anaesthetised pigs are shown in Fig. 2. The total carotid blood flow and its arteriovenous

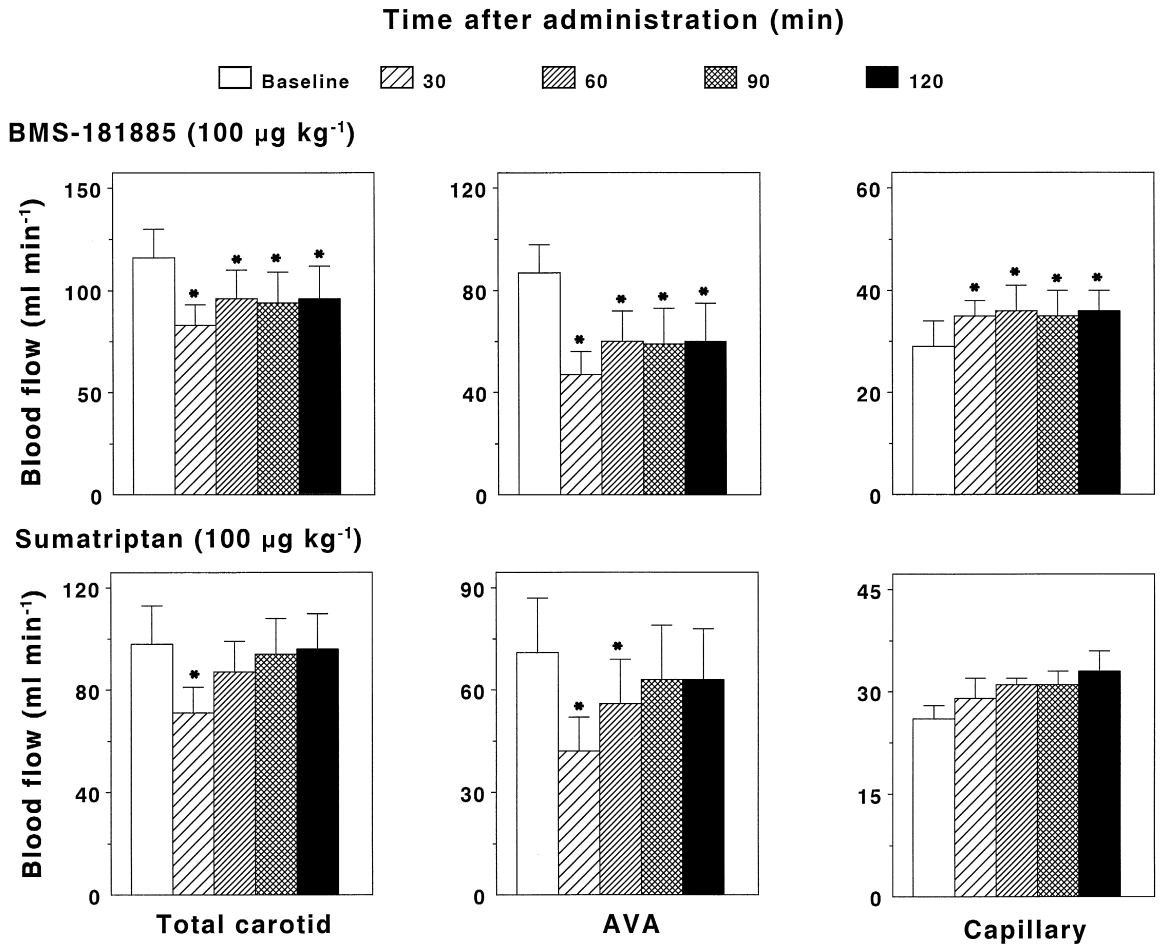

Fig. 4. Total, arteriovenous anastomotic (AVA) and nutrient (capillary) blood flow values in anaesthetised pigs measured before (baseline) and 30, 60, 90 or 120 min after i.v. administration of 100 μg kg^{-1} of BMS-181885 (upper panels) or sumatriptan (lower panels). All values are presented as means ± S.E.M. $* P < 0.05$ vs. baseline.

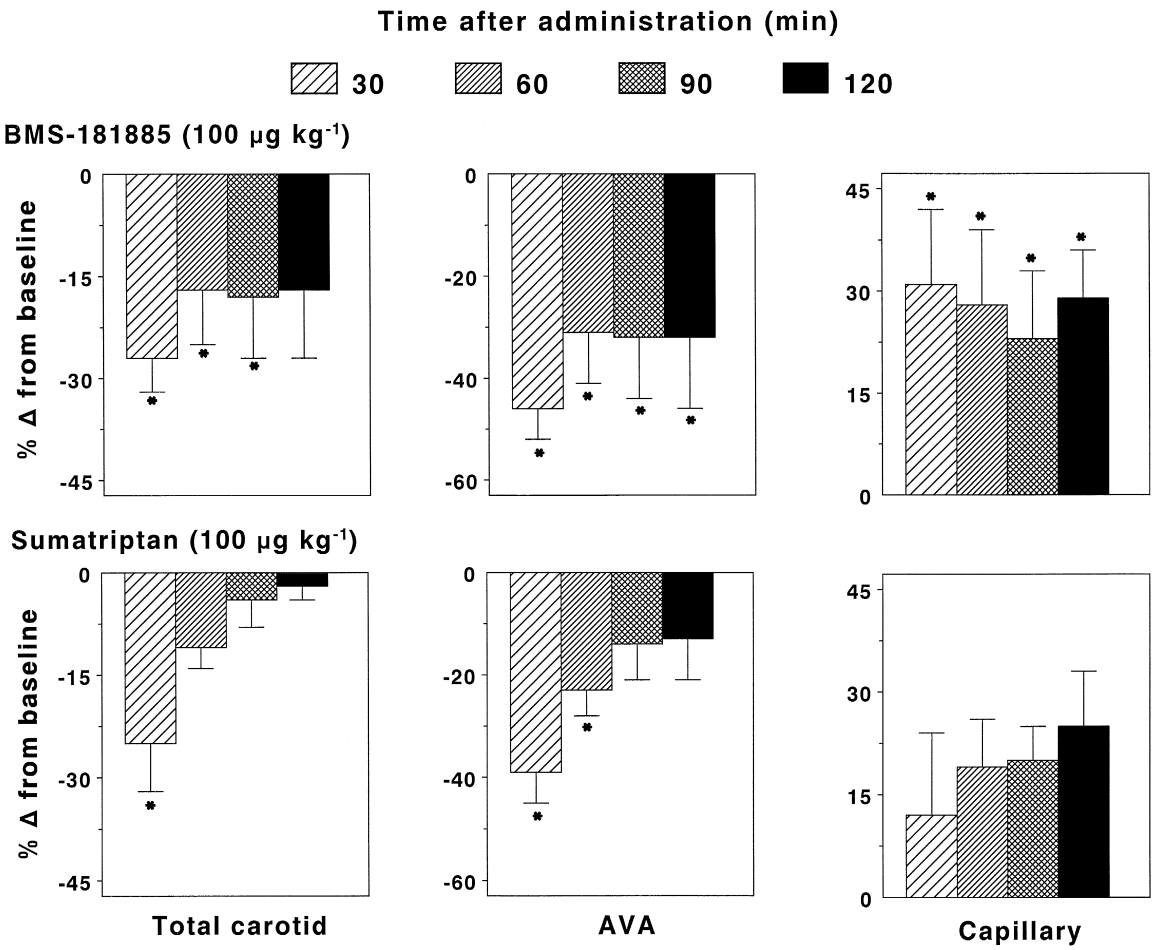

Fig. 5. Percent changes from baseline values of the total, arteriovenous anastomotic (AVA) and nutrient (capillary) blood flows in anaesthetised pigs at 30, 60, 90 or 120 min after i.v. administration of 100 μg kg^{-1} of BMS-181885 (upper panels) or sumatriptan (lower panels). All values are presented as means ± S.E.M. $*$ $P < 0.05$ vs. baseline.

anastomotic fraction were significantly decreased by all doses of BMS-181885. With the highest dose (300 μg kg^{-1}), the decrease in total carotid and arteriovenous anastomotic blood flow amounted to 26 ± 4% and 52 ± 6%, respectively. In contrast, the capillary fraction was significantly increased. This increase in capillary fraction

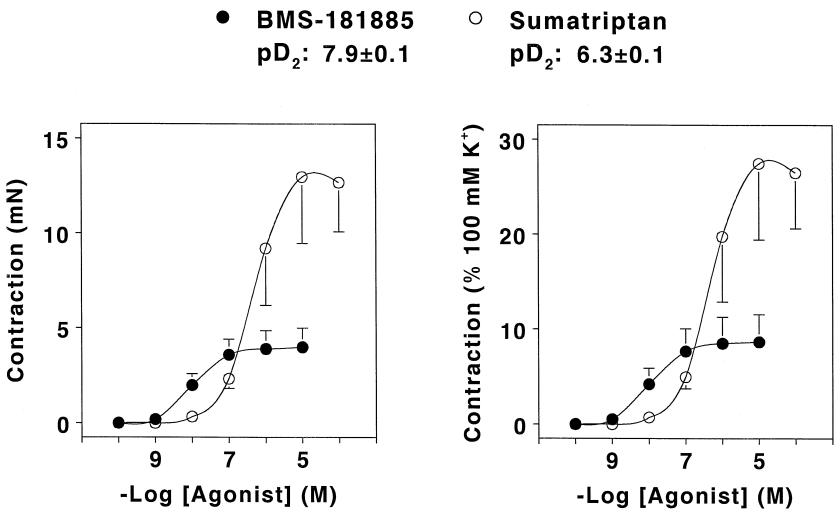

Fig. 6. Human isolated coronary artery. Contractile responses (mean ± S.E.M.; $n = 6$) to sumatriptan and BMS-181885, expressed both in mN (left panel) and as percentage of the response to 100 mM K$^+$ (right panel).

by BMS-181885 was noticed in several head tissues, including the skin, brain, ears, fat, muscles, bones and salivary gland (Fig. 3). The fraction distributed to the eye was slightly decreased after the highest dose.

The effects of a single 100 μg kg^{-1} dose of BMS-181885 and sumatriptan on carotid haemodynamics at different time-points are shown in Fig. 4 (absolute values) and Fig. 5 (% change from baseline). Both BMS-181885 and sumatriptan decreased carotid and arteriovenous anastomotic blood flows; with BMS-181885 capillary (tissue) blood flow also increased. The effects of BMS-181885 persisted for the 120-min observation period. In contrast, the effects of sumatriptan disappeared after 30–60 min.

3.3. Human isolated coronary artery

3.3.1. Effect of substance P and K^+

Coronary vessel segments pre-contracted with prostaglandin $F_{2\alpha}$ (1 μM) relaxed to substance P (1 nM) by $79 \pm 11\%$ (range 32–101%) of the contractile responses to prostaglandin $F_{2\alpha}$. This relaxant effect is a little more than in previous studies with substance P in the human isolated coronary artery obtained from patients undergoing cardiac transplantation (Bossaller et al., 1987). K^+ (100 mM) caused a mean contractile response of 47 ± 9 mN.

3.3.2. Effect of sumatriptan and BMS-181885

Both sumatriptan and BMS-181885 caused concentration-dependent coronary artery contractions (Fig. 6). The pD_2 of BMS-181885 (7.9 ± 0.1) was significantly higher than that of sumatriptan (6.3 ± 0.1) (paired *t*-test: $P < 0.001$). The E_{max} values for sumatriptan and BMS-181885 were, respectively, $28 \pm 8\%$ and $9 \pm 3\%$ of 100 mM K^+ ($P = 0.045$) or 14 ± 5 and 4 ± 1 mN ($P = 0.059$).

4. Discussion

4.1. Systemic haemodynamic changes

As reported earlier with sumatriptan (Feniuk et al., 1989; Den Boer et al., 1991a; Den Boer et al., 1992; De Vries et al., 1996) and avitriptan (Saxena et al., 1997), BMS-181885 caused a small decrease in heart rate. The mechanism of this bradycardiac effect is not clear, but it may be related to presynaptic inhibition of sympathetic neurons (Humphrey et al., 1988, 1990) or central 5-HT_{1A} receptor activation (Dreteler et al., 1989; Saxena and Villalón, 1990). However, bradycardia following the use of sumatriptan in patients is of little clinical relevance (Saxena and Tfelt-Hansen, 1993) and the same may be true for BMS-181885. Significantly, BMS-181885 did not produce any change in mean arterial blood pressure with the first three doses and a slight decrease was noted after the highest dose. Therefore, as is the case with sumatriptan (Humphrey et al., 1988, 1990; Den Boer et al., 1991a), the drug has a more selective vasoconstrictor action on cranial blood vessels than ergotamine, which may elicit a hypertensive response (Saxena and De Vlaam-Schluter, 1974; Den Boer et al., 1991b).

4.2. Carotid haemodynamic changes

In the past, we have reported that no significant changes in the carotid haemodynamics are observed during the experimental period after treatment with physiological saline (Saxena and Verdouw, 1982; Den Boer et al., 1991a). On the other hand, sumatriptan (Perren et al., 1989; Den Boer et al., 1991a; De Vries et al., 1996) and avitriptan (Saxena et al., 1997) clearly decrease porcine carotid arteriovenous anastomotic blood flow. In the present experiments, BMS-181885 elicited a reduction in the total carotid blood flow, which was exclusively due to the decrease in its arteriovenous anastomotic fraction; the capillary fraction distributed to several head tissues in fact increased. In conformity with the reduction of arteriovenous anastomotic blood flow, BMS-181885 significantly increased the arterio-jugular venous oxygen saturation difference.

The vasoconstrictor effect of BMS-181885 on porcine carotid arteriovenous anastomoses was qualitatively similar to that of sumatriptan (Den Boer et al., 1991a; De Vries et al., 1996). This effect of BMS-181885 appears to be less than that observed with sumatriptan as well as avitriptan in the same experimental model; the maximum decreases by 300 μg kg^{-1}, i.v. of BMS-181885 (Fig. 2), sumatriptan (De Vries et al., 1996) and avitriptan (Saxena et al., 1997) were 52 ± 6, 76 ± 4 and $72 \pm 4\%$, respectively. However, after similar reductions in the carotid blood flow following a single 100 μg kg^{-1}, i.v. dose at 30 min, the effect of BMS-181885 lasted longer than that of sumatriptan (see Figs. 4 and 5). Although differences in the plasma half-life values of sumatriptan and BMS-181885 in the pig can explain the longer duration of action of the latter drug, an equally likely factor may be the slow dissociation rate of BMS-181885 from 5-$HT_{1B/1D}$ receptors (Yocca et al., 1997).

In this study no attempt was made to analyze the mechanism of action involved in the constriction of porcine carotid arteriovenous anastomoses by BMS-181885. However, it is reasonable to assume that, as is the case with sumatriptan (Saxena et al., 1986; De Vries et al., 1996), 5-$HT_{1B/1D}$ receptors may mediate the responses to BMS-181885. Indeed, BMS-181885 has an even higher affinity than sumatriptan for human 5-$HT_{1B/1D}$ receptors (see Table 1). Furthermore, BMS-181885 constricts canine, bovine and human cephalic blood vessels with a greater potency than sumatriptan (Yocca et al., 1997).

4.3. Human isolated coronary artery

It is well known that sumatriptan constricts the human coronary artery, both in vivo (MacIntyre et al., 1993) and

in vitro (Connor et al., 1989; Bax et al., 1993; Chester et al., 1993; Kaumann et al., 1994; MaassenVanDenBrink et al., 1996, 1998). It is suggested that the sumatriptan-induced contractions of the human isolated coronary artery are mediated by the 5-HT_{1B} receptor (Kaumann et al., 1994). Our findings support this contention since BMS-181885 was about 30 times more potent than sumatriptan in contracting the human isolated coronary artery (pD_2: 7.9 and 6.3, respectively; Fig. 6) as well as in radiolabelled binding assay at the human 5-HT_{1B} receptor (pK_i: 9.05 and 7.54, respectively; Table 1).

As has been observed previously (Kaumann et al., 1994; MaassenVanDenBrink et al., 1996), the maximal response to sumatriptan on the human isolated coronary artery showed a considerable variability (see Fig. 6). Yet, the E_{max} of sumatriptan, when expressed as percentage of K^+ response, was significantly more than that of BMS-181885.

4.4. Clinical perspectives

A number of drugs effective in aborting migraine headaches, including the ergot alkaloids, ergotamine and dihydroergotamine (Johnston and Saxena, 1978; Schamhardt et al., 1979; Den Boer et al., 1991b; De Vries et al., 1998), sumatriptan (Perren et al., 1989; Den Boer et al., 1991a; De Vries et al., 1996), as well as second generation 5-$HT_{1B/1D}$ receptor agonists effective in migraine, e.g., zolmitriptan, rizatriptan and avitriptan (Saxena and Ferrari, 1996; Saxena et al., 1997), constrict carotid arteriovenous anastomoses, which may open up during the headache phase of migraine (Heyck, 1969; Saxena, 1990; Ferrari and Saxena, 1993; Saxena, 1995). Thus, it is not surprising that BMS-181885, which seems to be effective in preliminary clinical trials in aborting acute migraine attacks (Unpublished data, Bristol-Myers Squibb files), also constricts porcine arteriovenous anastomoses. The longer duration of action BMS-181885 as compared to sumatriptan in our experiments suggests that potentially BMS-181885 may have less headache recurrence rate than sumatriptan.

Interestingly, BMS-181885 displayed only a weak activity (1000-fold less potent than sumatriptan) in reducing electrically-evoked plasma extravasation in guinea pig dura-mater (Yocca et al., 1997). The clinical efficacy of BMS-181885 in migraine once again shows that the inhibition of neurogenic plasma extravasation may not be a good indicator of antimigraine activity. Also, it may be recalled that tachykinin NK_1 and endothelin $ET_{A/B}$ receptor antagonists, which potently inhibit plasma extravasation (Gupta et al., 1995; Shepheard et al., 1995; Brändli et al., 1996; Phebus et al., 1997), failed to show clinical efficacy in migraine patients (Diener, 1995; Goldstein, 1996; May et al., 1996). Moreover, the conformationally restricted sumatriptan analogue, CP-122,288 ((*R*)-*N*-methyl-[3-(1-methyl-2-pyrrolidinylmethyl)-1*H*-indol-5-yl]methane sulphonamide), which has similar affinities as sumatriptan at cloned human 5-HT_{1B} and 5-HT_{1D} receptors, but is 'super-potent' (10,000 × sumatriptan) in the rat neurogenic plasma extravasation model (Lee and Moskowitz, 1993; Gupta et al., 1995; Yu et al., 1997), was ineffective in acute migraine, when used at a dose comparable to its activity in this neurogenic inflammation model (Roon et al., 1997). Recently, it has been suggested that trigeminal ganglion stimulation in rats may have two components: dural plasma extravasation mediated by substance P, released from sensory C-fibres and dural vasodilatation mediated by calcitonin gene-related peptide, released from A δ-fibres (Shepheard et al., 1997). Shepheard et al. (1997) showed that while CP-122,288 is indeed 'super-potent' in inhibiting dural plasma extravasation, it was only equipotent with sumatriptan in inhibiting vasodilatation. Thus, it would appear that the inhibition of neurogenic vasodilatation rather than plasma extravasation is indicative of antimigraine activity. We do not know whether BMS-181885 attenuates neurogenic vasodilatation.

Lastly, the low efficacy of BMS-181885 in contracting the human isolated coronary artery is an attractive finding, which may suggest a low coronary side-effect potential. However, it will need a long clinical experience before it can be established that these in vitro findings are indeed advantageous in clinical setting. Interestingly, BMS-181885 fails to contract the dog saphenous vein or guinea-pig iliac artery, yet it causes potent contractions of cephalic vessels in human and other species (Yocca et al., 1997), exemplifying vascular tissue selectivity with this drug.

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