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BASIC PHARMACOLOGY

Sertraline Causes Strong Coronary Vasodilation: Possible Relevance for Cardioprotection by Selective Serotonin Reuptake Inhibitors

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Summary. Objective: **Although Selective Serotonin Reuptake Inhibitors (SSRIs) are important antidepressant drugs, knowledge of their vaso active effects is limited. Vaso active effects of the SSRI sertraline were studied in rings of rat aorta, human Internal Mammary Arteries (IMAs) and in Langendorff perfused rat hearts.**

Methods: **The effects of sertraline (0.1 to 300 μmol $\cdot$ L^{-1}) on precontracted rat aortic and IMA rings were evaluated in organ bath chambers. Precontraction was elicited by serotonin (5-HT; 10 μmol $\cdot$ L^{-1}), phenylephrine (PE; 10 μmol $\cdot$ L^{-1}) and potassium chloride (KCl; 50 mmol $\cdot$ L^{-1}). In addition, the effects of sertraline on PE induced contraction curves were established by subjecting vascular rings to increasing doses of PE (1 nmol $\cdot$ L^{-1} to 10 μmol $\cdot$ L^{-1}) in the presence of sertraline or vehicle. Finally, the effects of sertraline on *ex vivo* coronary flow in rat hearts were examined using a retrograde Langendorff perfusion model.**

Results: **Sertraline elicited dose-dependent relaxation, independent of the substance used for precontraction ($p < 0.025$). Sertraline showed a rightward shift of dose-response curves to PE ($p < 0.01$). Vasodilatory effects of SSRIs were endothelium independent. In perfused rat hearts, sertraline (0.3 to 10 μmol $\cdot$ L^{-1}) showed a concentration-dependent increase in coronary flow that returned to baseline levels after wash-out of the antidepressant ($p = 0.005$).**

Conclusions: **One of the SSRIs, sertraline, showed marked vasodilatory effects in rat aorta and human IMAs. Sertraline elicited vasodilatation in coronary arteries during perfusion of rat hearts. These hemodynamic effects may explain the observed beneficial effects in myocardial ischemia and infarction.**

Key Words. **5-HT (5-hydroxytryptamine, serotonin), pharmacology, smooth muscle relaxation, vasodilation, sertraline, SSRI**

Introduction

Coronary artery disease (CAD) and depression represent the top contributors to the burden of diseases in the year 2010 [1]. Moreover, both diseases often coexist within the same person [2]. Therefore, increasing attention is paid to antidepressant interventions in patients with CAD.

Since their introduction in 1987, the use of Selective Serotonin Reuptake Inhibitors (SSRIs) has increased dramatically [3]. Accordingly, it can be expected that SSRIs will be prescribed increasingly to CAD patients, including patients in the aftermath of coronary artery bypass surgery (CABG) [4]. However, our knowledge of the cardiovascular effects of SSRIs is mainly based on animal studies in a variety of vascular beds [5–8]. In addition, SSRIs revealed conflicting results with both vasodilatory [5,9] and vasoconstrictory responses [7]. Unfortunately, vaso active effects of drugs on different arteries are not always comparable. The net result is dependent on the species, route of administration, experimental conditions, presence of functional endothelium, etcetera.

Based on these considerations, we investigated the vaso active effects of sertraline, one of the most frequently prescribed SSRIs, on *human* Internal Mammary Arteries (IMAs), which are employed as bypass grafts in coronary surgery. Moreover, we set out to

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evaluate, for the first time, the effects of sertraline on *coronary* flow in isolated perfused rat hearts.

Material and Methods

The effects of sertraline were tested on three different types of vasculature:

- Rat aorta
- Human IMAs
- Rat coronary artery bed

Rat aorta and human IMAs were tested *in vitro* using ring preparations of these blood vessels mounted in organ baths, and rat coronary artery bed was tested *ex vivo* during Langendorff experiments. All procedures were reviewed and approval was obtained from the Animal Research Committee (rat studies) and the Institutional Review Board (IMA-study) of the University Hospital Groningen.

Sampling of the blood vessels

Male Wistar rats ($n = 26$; 322–584 g; Harlan, Zeist, The Netherlands) were anaesthetized with isoflurane in 2% oxygen. The thoracic descending aorta was removed and placed in a Krebs-buffer solution of the following composition ($mMol \cdot L^{-1}$): NaCl (120.4), KCl (5.9), $CaCl_2$ (2.5), $MgCl_2$ (1.2), NaH_2PO_4 (1.2), glucose (11.5), and $NaHCO_3$(25.0), aerated with 95% O_2 and 5% CO_2.

Human arterial segments were harvested of IMAs that were discarded from patients undergoing elective Coronary Artery Bypass Graft surgery ($n = 22$ in total, mean age 63 years, 73% male). The majority of the patients used frequently prescribed cardiac medication (e.g., statins, β-blockers, ACE-inhibitors, aspirin), but none of the patients received antidepressant medication. IMA vessels were stored after surgery in Krebs-buffer at 4°C until preparation (time to preparation was maximal 16 h).

Preparation of blood vessels

Methods have been described previously [10,11]. Briefly, the blood vessels were cleaned of adhering tissue and cut into rings of 2 mm in length. The number of rings taken from each vessel varied from 5 to 16 (rat aorta) and from 3 to 11 (human IMA). In experiments in which the role of the endothelium was studied, 50% of the rings were subjected to removal of intimal surfaces by inserting a steel wire into the lumen and gently rolling the rings. Rings were mounted in an organ bath containing the above-mentioned buffer at 37°C, connected to an isotonic displacement transducer, given a preload of 1.4 g, and allowed to equilibrate for at least 60 min. Rings were then stimulated with 10 $\mu mol \cdot L^{-1}$ of the α_1-adrenoceptor agonist phenylephrine (PE) (2–3 times with intermediate washing) to test vessel viability and for determination of a control response for reference of contractile responses; rings not appropriately responding to repeated challenge with 10 $\mu mol \cdot L^{-1}$ PE (i.e., contraction < 100 μm) were excluded from the experiments. Functional integrity of the rings with intact endothelium was confirmed routinely by the presence of relaxation induced by 10 $\mu mol \cdot L^{-1}$ metacholine (ME) in PE-precontracted rings.

Experiments in arterial rings

After washing and restabilization, the contractile and dilatory effect of sertraline was studied at baseline and after precontraction, respectively. To this end, increasing concentrations of sertraline (0.1 to 300 $\mu mol \cdot L^{-1}$) were administered cumulatively either to unstimulated rings at baseline or rings stimulated with one of three different vasoconstrictor agents (i.e. either 10 $\mu mol \cdot L^{-1}$ PE, 50 $mmol \cdot L^{-1}$ KCl, or 10 $\mu mol \cdot L^{-1}$ 5-HT). The effect of sertraline was always studied in comparison to the effect of vehicle (dimethyl sulfide oxide, DMSO), similarly given to parallel rings under the same conditions.

In an additional series of experiments, we also tested the effect of pre-incubation (90 min) with different concentrations of sertraline (0.1–10 $\mu mol \cdot L^{-1}$) or vehicle (DMSO) on contractions induced by increasing concentrations of PE (1 $nmol \cdot L^{-1}$ to 10 $\mu mol \cdot L^{-1}$). To eliminate any confounding effects of vaso active substances released from the endothelium, we only used endothelium-denuded rings for the latter experiments.

Isolated perfused rat heart

Male Wistar rats ($n = 12$, Harlan, Zeist, The Netherlands) were anaesthetized with isoflurane in 2% oxygen and heparinized (500 $IU \cdot kg^{-1}$ administered via the tail vein). The hearts were rapidly excised and immersed in ice-cold 0.9% NaCl. Retrograde perfusion of the aorta, essentially by the Langendorff method was achieved immediately using a modified Tyrode solution equilibrated with 95% O_2 and 5% CO_2, as described previously [12]. This buffer was filtered through a 1.2 μm pore-size filter before reaching the heart. Perfusion pressure was maintained at 60 mmHg and temperature (measured at the aortic cannula tip) was kept between 38.0 and 38.5°C. The hearts beat spontaneously throughout the experiment. Coronary flow (volume of perfusion fluid per unit of time) was measured by a microprocessor, which controlled the perfusion pressure by adjusting the peristaltic perfusion pump.

After an equilibration period of 10 min, baseline measurements were made, and subsequently concentration-response relations were determined with increasing concentrations of either sertraline (0,3 to 10 $\mu mol \cdot L^{-1}$) or vehicle (DMSO). Each concentration was added to the perfusion fluid and mean changes in coronary flow were measured within 5 min. Baseline coronary flow as well as sertraline- and

vehicle-induced changes in flow were corrected for heart weight ($ml \cdot min^{-1} \cdot g^{-1}$).

Compounds

Sertraline hydrochloride was a gift from Pfizer Inc (Groton, USA). Pfizer Inc was not financially involved, or concerned with either the study design, data analysis or writing process; acetyl-β-methylcholine chloride was purchased from Aldrich-Chemie (Steinheim, Germany); 5-hydroxytryptamine (5-HT) and l-phenylephrine hydrochloride (PE) from Sigma (St Louis MO, USA), and compounds for the buffer solutions from Merck (Darmstad, Germany). Sertraline hydrochloride was dissolved in DMSO (dimethyl sulfide oxide). The DMSO concentration for the highest concentration sertraline stock solution was 20%, yielding a maximal final concentration DMSO of 0.2% in the organ bath. All other compounds were dissolved and diluted in water or saline, where appropriate, and prepared daily from stock solutions.

Calculations and statistical analysis

Responses of individual rings were expressed as a percentage of precontraction. Comparisons between the complete concentration-response curves were made by repeated measures analysis of variance with a correction for multisample aspherity as recommended by Ludbrook [13] (General Linear Model procedure). Comparisons between responses to specific concentrations were made using Student t test. For both statistical procedures, SSPS 11.1 for Windows was used. Unless stated otherwise, all data are expressed as mean ± standard error of the mean (SEM). Differences were considered significant at a level of $p < 0.05$. Figures were plotted by SigmaPlot 8.0 for Windows.

Results

Prior to vascular experiments, the viability test with 10 $\mu mol \cdot L^{-1}$ PE caused appropriate contraction in 222 out of 249 rat aortic rings (89%) and 104 out of 148 human IMA rings (70%). As expected, ME was unable to induce vasodilation in the endothelium denuded rings: mean change (% of precontraction) of endothelium intact rings versus endothelium denuded rings was -69 ± 5.3 versus 0.2 ± 1.3 for rat aortic rings ($p < 0.001$) and -54 ± 9.5 versus -7 ± 5.2 for human IMA rings ($p < 0.001$).

Effects of sertraline in rat aortic and human IMA rings

Administration of sertraline to unstimulated ring preparations at baseline did not result in contractile responses (data not shown), but did cause significant dilation in rings previously stimulated with a vasoconstrictor agent. The level of precontraction between arterial rings subsequently subjected to sertraline versus those subsequently subjected to vehicle (i.e. control rings) did not statistically differ (neither after PE or KCl, nor after 5-HT; all tests: $p > 0.05$). In all cases, sertraline induced a dose-dependent relaxation of precontracted vascular rings compared to vehicle ($p < 0.05$ for all tests; Figs. 1 and 2 for rat aortic rings and human IMA rings, respectively). In almost all of the comparisons between sertraline and vehicle, sertraline started to significantly dilate the aorta at concentrations higher than 10 $\mu mol \cdot L^{-1}$, resulting in full-reversal of precontraction at higher concentrations. Moreover, there were no significant differences between rubbed and unrubbed rings (all tests $p > 0.25$), indicating that presence of a functional endothelium was not a prerequisite for sertraline-induced dilation. The property of sertraline to antagonize constriction in human and rat artery was confirmed in the reversed experiment, in which preincubation of ring preparations with sertraline significantly suppressed contractile responses elicited with PE (Figs. 3a and b).

Effect of sertraline on coronary flow in the isolated rat heart

After the stabilization period of 10 minutes, baseline coronary flow was comparable in hearts assigned to receive vehicle and those assigned to receive sertraline (respectively 11.5 and 12.4 $ml \cdot min^{-1} \cdot g^{-1}$; $p = 0.6$). Sertraline, but not vehicle, significantly increased coronary flow in a dose-dependent fashion ($p = 0.005$; Fig. 4). After a 10 min wash-out period at the end of the experiment, the coronary flow fully returned to baseline levels, pointing to a reversible sertraline-induced coronary vasodilation.

Discussion

In the present study, we assessed the effects of the antidepressant sertraline on vascular tone in ring preparations of human IMA and rat aorta. Sertraline caused concentration-dependent dilation in preconstricted arteries in both species and exerts no contractile effects at baseline. Moreover, high dose sertraline was able to fully reverse the level of vascular constriction, independent of the type of vasoconstrictor used. Sertraline additionally showed the ability to antagonize PE-induced constriction in the reversed experiment. Together with the fact that sertraline also dose-dependently increased coronary flow in the isolated rat heart, we conclude that the SSRI sertraline may act as an endothelium independent vasodilator drug, not only in rat arteries (including in the coronary vascular bed), but also in human arteries.

To our knowledge, this is the first study on the vaso active effects of a SSRI on human IMAs and coronary flow in Langendorff experiments. The observed vasodilatory effects of sertraline are in line with studies investigating the vaso active effects of SSRIs in other

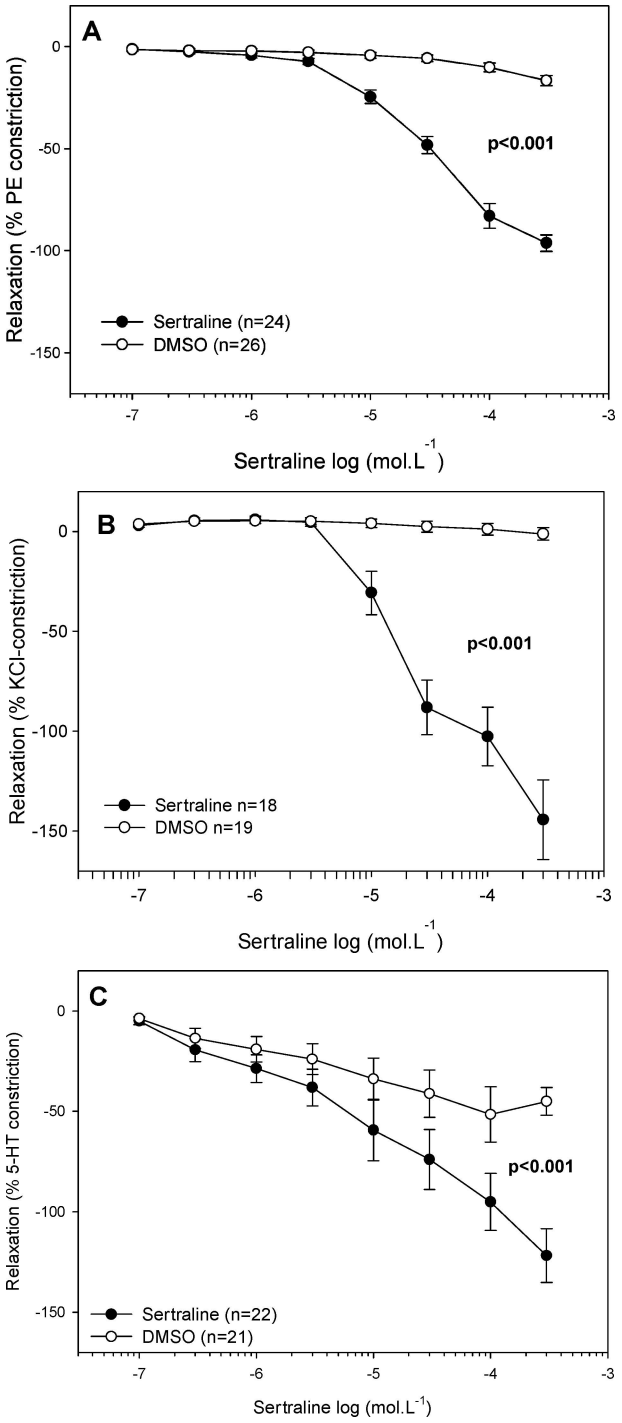

Fig. 1. *Sertaline induced relaxation in rat aortic rings. Rings were pre-constricted with either (a) 10 $\mu mol \cdot L^{-1}$ PE (b) 50 $mmol \cdot L^{-1}$ KCl or (c) 10 $\mu mol \cdot L^{-1}$ 5-HT before sertraline was administered in cumulative fashion. Comparisons were made to parallel control rings treated with vehicle. Relaxations are expressed as a percentage of constriction, and the data represent the mean ± SEM; lines represent the pooled results obtained with endothelium-denuded and—intact preparations (presence of the endothelium did not alter the responses, see also text).*

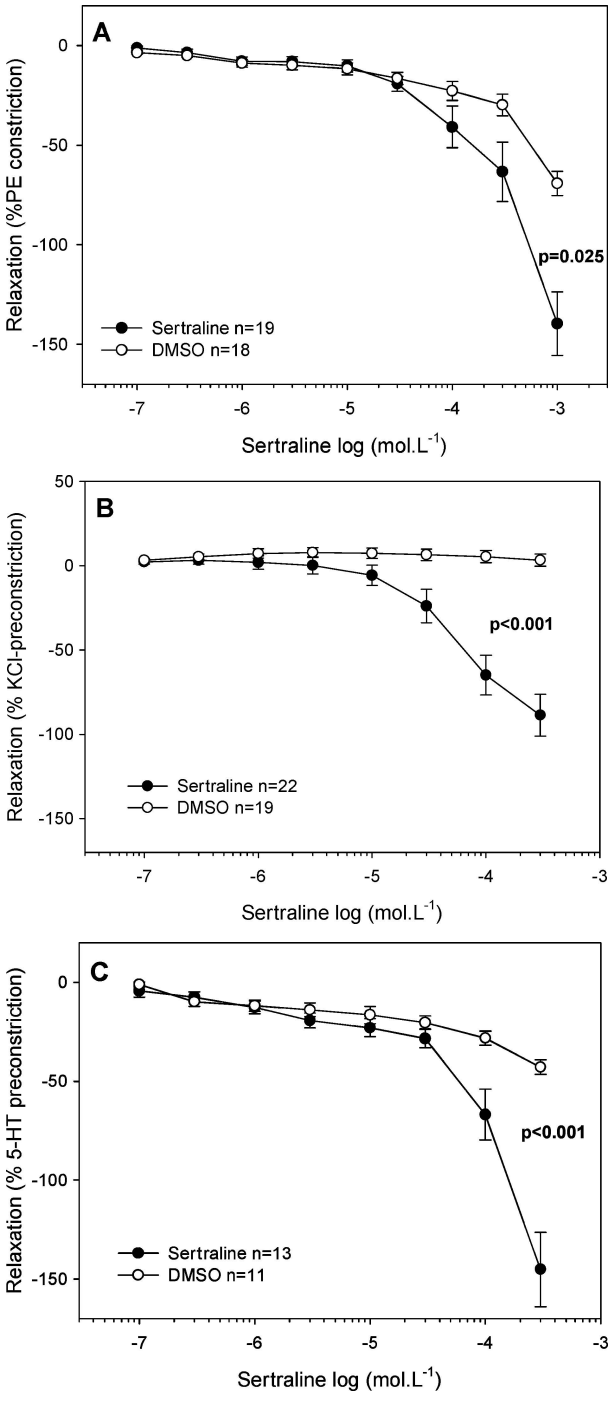

Fig. 2. *Sertaline induced relaxation in human IMA rings. Rings were pre-constricted with either (a) 10 $\mu mol \cdot L^{-1}$ PE (b) 50 $mmol \cdot L^{-1}$ KCl or (c) 10 $\mu mol \cdot L^{-1}$ 5-HT before sertraline was administered in cumulative fashion. Comparisons were made to parallel control rings treated with vehicle. Relaxations are expressed as a percentage of constriction, and the data represent the mean ± SEM; lines represent the pooled results obtained with endothelium-denuded and—intact preparations (presence of the endothelium did not alter the responses, see also text).*

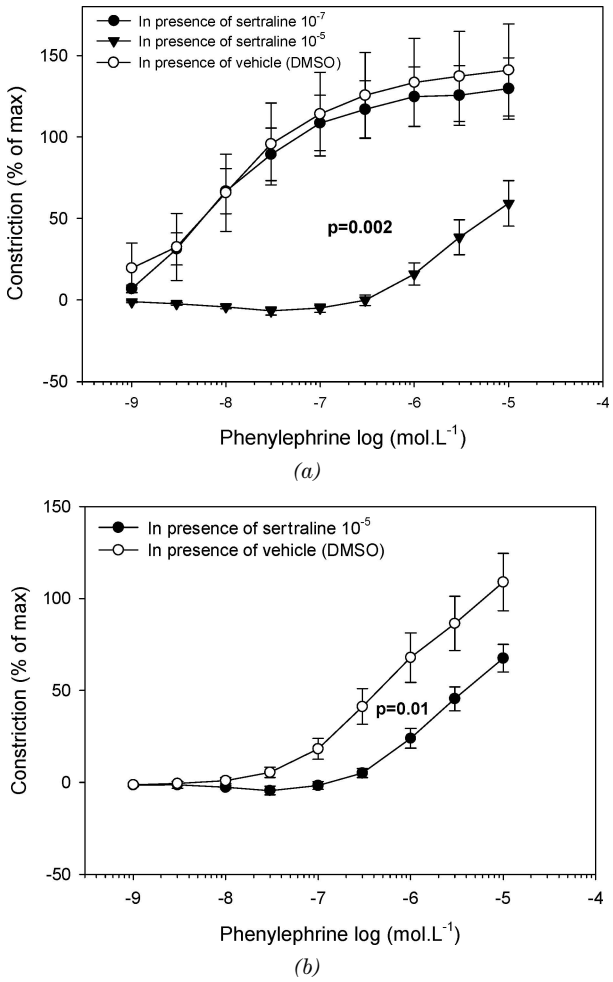

Fig. 3. *Phenylephrine (PE) induced constriction in (a) rat aortic rings and (b) human Internal Mammary Artery rings. Presence of sertraline (10 $\mu mol \cdot L^{-1}$) compared to vehicle (DMSO) significantly attenuated constrictory responses resulting in a rightward shift of the concentration-response curve ($p = 0.002$ for rat aortic rings and $p = 0.01$ for human Internal Mammary Artery rings). Constrictions are expressed as a percentage of maximum constriction assessed during viability tests with PE (10 $\mu mol \cdot L^{-1}$), and the data represent the mean ± SEM; lines represent the results obtained with endothelium-denuded preparations (see also text).*

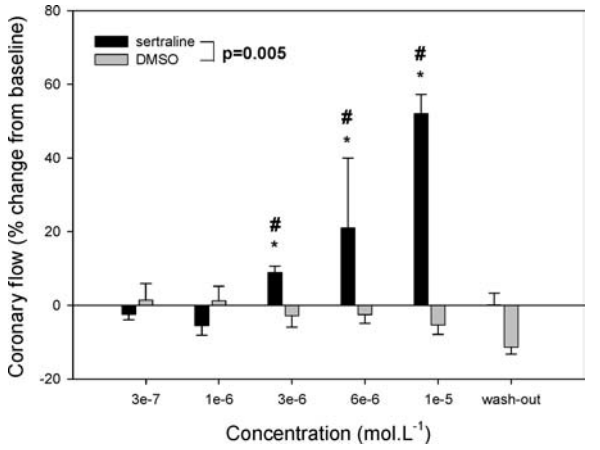

Fig. 4. *Change in coronary flow (corrected for heart weight (g) in Langendorff rat hearts during perfusion with vehicle (DMSO, $n = 5$) or sertraline ($n = 7$). Sertraline (0,3 to 10 $\mu mol \cdot L^{-1}$) elicited a dose dependent increase in coronary flow ($p = 0.005$). After wash-out of the compound, coronary flow returned to baseline levels. * $p < 0.05$ vs. baseline; # $p < 0.05$ vs vehicle.*

vessels. For example, from the data described by Cohen and Wiley [5] it can be deduced that fluoxetine had a vasodilatory effect in rat aorta after precontraction with norepinephrine and 5-HT. In 1983, Seabrook and Nolan [14] reported about a fluoxetine-induced rightward shift of a 5-HT dose-response curve in rat mesenteric arteries. A recent report of Ungvari et al. [8] describes the finding that fluoxetine elicited dilation of rat cerebral arteries. Sertraline produced concentration-dependent relaxation of precontracted mesenteric artery rings [15]. Although previous findings in rat aortic preparations were mixed [5,7,9], our data clearly provide evidence for the vasodilatory effects of sertraline. The fact that vasodilatory effects were also observed in studies with fluoxetine, suggest a possible class effect of SSRIs.

The mechanism responsible for the vasodilatory responses of SSRIs has not been fully elucidated yet. Studies by Ungvari et al. [8] using fluoxetine suggest interference of this SSRI with L-type calcium channels to prevent Ca^{2+} entry. This is in line with our present finding that sertraline reduced contractions induced by high extracellular K^+, i.e. a condition which elicits smooth muscle contraction by promoting *extra*cellular Ca^{2+} influx. In addition, sertraline also reduced contraction after PE- and 5-HT, which also depend on (IP_3 mediated) calcium mobilisation/release from *intra*cellular sites. Thus, if inhibition of calcium-entry through L-type calcium channels would be the primary mechanism, sertraline would not be expected to attenuate these receptor-mediated contractions as observed. Therefore, it may be that the dilatory actions of sertraline resulted from interference with intracellular calcium signaling pathways in addition to blockade of calcium influx through L-type calcium channels, as was suggested after studies with fluoxetine [16]. Thus, in contrast to the alleged interference of sertraline with 5-HT metabolism to exert its antidepressive action [17], its vasodilatory effect seems not be mediated by the large family of 5-HT receptors [18], which in turn implies that its vasodilatory response may represent a pleimorphic effect of sertraline.

The results have to be considered in relation to study limitations. Compared to in vivo therapeutic plasma concentrations of sertraline (approximately 0.1 μmol $\cdot$ L^{-1}), vasodilation in this *in vitro* study was elicited by relatively high concentrations of sertraline (i.e. 1–30 μmol $\cdot$ L^{-1}). In general, it seems that drug concentrations in the organ bath need to be higher than *in vivo* to elicit an identical response. For instance, this holds true for the angiotensin receptor blocker losartan of which it has been repetitively shown that, in organ baths, the *in vitro* response of vessel rings occur at supraphysiologically concentrations [19]. It seems that *in vitro* findings obtained from studies in organ baths are well capable of predicting a general effect of a compound, but not the precise dose-response *in vivo*. Further *in vivo* research is needed to confirm a potential vaso active effect of sertraline at therapeutical levels.

Patients with CAD often suffer from depression. Unlike tricyclic antidepressants, SSRIs seem to be a safe and efficacious treatment for depression in these patients [20,21]. In fact, it was observed that SSRI-use is associated with reduced odds of myocardial infarction [22,23]. The antiplatelet and endothelium-protective properties of SSRIs might represent an attractive explanation for this observation [24]. Our study shows that, besides these antiplatelet and endothelium-protective properties, SSRI-mediated vasodilation and attenuation of vasoconstriction may be an additional explanation for presumed protective effects. Further research is warranted to explore the effects of sertraline and other SSRIs on vascular function. In particular, it needs to be investigated whether these laboratory findings can be translated to the clinical setting.

References

1. Murray CJ, Lopez AD. Alternative projections of mortality and disability by cause 1990–2020: Global Burden of Disease Study. *Lancet* 1997;349:1498–1504.
2. Schleifer SJ, Macari-Hinson MM, Coyle DA, et al. The nature and course of depression following myocardial infarction. *Arch Intern Med* 1989;149:1785–1789.
3. Meijer WE, Heerdink ER, Leufkens HG, Herings RMC, Egberts ACG, Nolen WA. Incidence and determinants of long-term use of antidepressants. *Eur J Clin Pharmacol* 2004;60:57–61.
4. Connerney I, Shapiro PA, McLaughlin JS, Bagiella E, Sloan RP. Relation between depression after coronary artery bypass surgery and 12-month outcome: A prospective study. *Lancet* 2001;358:1766–1771.
5. Cohen ML, Wiley KS. Neuronal uptake inhibitors, nisoxetine and fluoxetine on rat vascular contractions. *Eur J Pharmacol* 1977;44:219–229.
6. Szabo C, Hardebo JE, Owman C. An amplifying effect of exogenous and neurally stored 5-hydroxytryptamine on the neurogenic contraction in rat tail artery. *Br J Pharmacol* 1991;102:401–407.
7. Gruetter CA, Lemke SM, Anestis DK, Szarek JL, Valentovic MA. Potentiation of 5-hydroxytryptamine-induced contraction in rat aorta by chlorpheniramine, citalopram and fluoxetine. *Eur J Pharmacol* 1992;217:109–118.
8. Ungvari Z, Pacher P, Kecskemeti V, Koller A. Fluoxetine dilates isolated small cerebral arteries of rats and attenuates constrictions to serotonin, norepinephrine, and a voltage-dependent Ca(2$^+$) channel opener. *Stroke* 1999;30:1949–1954.
9. Becker B, Morel N, Vanbellinghen AM, Lebrun P. Blockade of calcium entry in smooth muscle cells by the antidepressant imipramine. *Biochem Pharmacol* 2004;68:833–842.
10. Wagenaar LJ, Voors AA, Buikema H, et al. Functional antagonism of different angiotensin II type I receptor blockers in human arteries. *Cardiovasc Drugs Ther* 2002;16:311–316.
11. Voors AA, Oosterga M, Buikema H, et al. Dyslipidemia and endothelium-dependent relaxation in internal mammary arteries used for coronary bypass surgery. *Cardiovasc Res* 1997;34:568–574.
12. Buikema H, van Veldhuisen DJ, Hegeman H, van Gilst WH. Early pharmacologic intervention may prevent the deterioration in endothelial function after experimental myocardial infarction in rats: Effects of ibopamine and captopril. *J Card Fail* 1997;3:125–132.
13. Ludbrook J. Repeated measurements and multiple comparisons in cardiovascular research. *Cardiovasc Res* 1994;28:303–311.
14. Seabrook JM, Nolan PL. The vascular interaction of noradrenaline and 5-hydroxytryptamine. *Eur J Pharmacol* 1983;89:131–135.
15. Vila JM, Medina P, Segarra G, et al. Relaxant effects of antidepressants on human isolated mesenteric arteries. *Br J Clin Pharmacol* 1999;48:223–229.
16. Ungvari Z, Pacher P, Koller A. Serotonin reuptake inhibitor fluoxetine decreases arteriolar myogenic tone by reducing smooth muscle [Ca2$^+$]i. *J Cardiovasc Pharmacol* 2000;35:849–854.
17. Reimherr FW, Chouinard G, Cohn CK, et al. Antidepressant efficacy of sertraline: A double-blind, placebo- and amitriptyline-controlled, multicenter comparison study in outpatients with major depression. *J Clin Psychiatry* 1990;51:18–27 (suppl B).
18. Roth BL, Chuang DM. Multiple mechanisms of serotonergic signal transduction. *Life Sci* 1987;41:1051–1064.
19. Timmermans PB, Wong PC, Chiu AT, et al. Angiotensin II receptors and angiotensin II receptor antagonists. *Pharmacol Rev* 1993;45:205–251.
20. Glassman AH, O'Connor CM, Califf RM, et al. Sertraline treatment of major depression in patients with acute MI or unstable angina. *JAMA* 2002;288:701–709.
21. Strik JJ, Honig A, Lousberg R, et al. Efficacy and safety of fluoxetine in the treatment of patients with major depression after first myocardial infarction: Findings from a double-blind, placebo-controlled trial. *Psychosom Med* 2000;62:783–789.
22. Sauer WH, Berlin JA, Kimmel SE. Selective serotonin reuptake inhibitors and myocardial infarction. *Circulation* 2001;104:1894–1898.

23. Sauer WH, Berlin JA, Kimmel SE. Effect of antidepressants and their relative affinity for the serotonin transporter on the risk of myocardial infarction. *Circulation* 2003;108:32–36.
24. Serebruany VL, Glassman AH, Malinin AI, et al. Platelet/endothelial biomarkers in depressed patients treated with the selective serotonin reuptake inhibitor sertraline after acute coronary events: The Sertraline AntiDepressant Heart Attack Randomized Trial (SADHART) Platelet Substudy. *Circulation* 2003;108:939–944.