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Vascular Adrenoceptors: An Update

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***Abstract*——The total and regional peripheral resistance and capacitance of the vascular system is regulated by the sympathetic nervous system, which influences the vasculature mainly through changes in the release of catecholamines from both the sympathetic nerve terminals and the adrenal medulla. The knowledge of the targets for noradrenaline and adrenaline, the main endogenous catecholamines mediating that**

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influence, has recently been greatly expanded. From two types of adrenoceptors (α and β), we have now nine subtypes (α_{1A}, α_{1B}, α_{1D}, $\alpha_{2A/D}$, α_{2B}, $\alpha_{2A/D}$, β_1, β_2, and β_3) and two other candidates (α_{1L} and β_4), which may be conformational states of α_{1A} and β_1-adrenoceptors, respectively. The vascular endothelium is now known to be more than a pure anatomical entity, which smoothly contacts the blood and forms a passive barrier against plasma lipids. Instead, the endothelium is an important organ possessing at least five different adrenoceptor subtypes ($\alpha_{2A/D}$, α_{2C}, β_1, β_2, and β_3), which either directly or through the release of nitric oxide actively participate in the regulation of the vascular tone. The availability of transgenic models has resulted in a stepwise progression toward the identification of the role of each adrenoceptor subtype in the regulation of blood pressure and fine-tuning of blood supply to the different organs: $\alpha_{2A/D}$-adrenoceptors are involved in the central control of blood pressure; α_1-(primarily) and α_{2B}-adrenoceptors (secondarily) contribute to the peripheral regulation of vascular tone; and $\alpha_{2A/D}$- and α_{2C}-adrenoceptors modulate transmitter release. The increased knowledge on the involvement of vascular adrenoceptors in many diseases like Raynaud's, scleroderma, several neurological degenerative diseases (familial amyloidotic polyneuropathy, Parkinson disease, multiple-system atrophy), some kinds of hypertension, etc., will contribute to new and better therapeutic approaches.

I. Introduction

"The nerves controlling the blood-vessels that supplied his face functioned so well that the skin, robbed of all its blood, went quite cold, the nose looked peaked, and the hollows beneath the young eyes were lead-couloured as any corpse's. And the Sympathicus *caused his heart, Hans Castorp's heart, to thump, in such a way that it was impossible to breathe except in gasps; and shivers ran over him, due to the functioning of the sebaceous glands, which, with the hair follicles, erected themselves".*—Thomas Mann, 1924

The operation of the sympathetic nervous system, especially of its cardiovascular branches, is nowhere in literature described better than in this passage from Thomas Mann's *Magic Mountain*, that great novel on pre-1914 Europe that the author places in a sanatorium at Davos in the Swiss mountains. Vasoconstriction, tachycardia, and contraction of the *musculi arrectorum pilorum* are Hans Castorp's autonomic responses when he first addresses his beloved Claudia Chaucat on Walpurgis-Night to borrow a pencil from her. This review probes the mechanisms that noradrenaline, the classical transmitter substance of the sympathetic vasoconstrictor fibers, uses to make blood vessels constrict; probes, in other words, the events that occurred in Hans Castorp when he borrowed the pencil.

Directly or indirectly, the blood vessels are the source of many and serious diseases that affect millions of people. In many respects, vascular physiology and pharmacology have changed dramatically over the last years. The discovery by Furchgott and Zawadzski in 1980 of endothelium-derived relaxing factor (EDRF[2]) revolutionized our knowledge and placed the endothelium in the center of the physiology and pathophysiology of the vascular tree; the cloning of many receptors brought about a true "Renaissance" in receptor pharmacology (Kenakin, 1997); and the possibility to "knock out" specific genes in experimental animals represents a new

[2] Abbreviations: EDRF, endothelium-derived relaxing factor; 5-MU, 5-methyl urapidil; WB-4101, 2-(2′,6′-dimethoxyphenoxyethyl)-aminomethyl-1,4-benzodioxane hydrochloride; BRL-37344, 1-(3-chlorophenyl)-2-[2-(4-(carboxymethoxy)phenyl)-1-methyl-ethylamino]-ethanol; CL-316243, disodium (*R,R*)-5-[2-[[2-(3-chlorophenyl)-2-hydroxyethyl] amino] propyl]-1,3-benzodioxole-2,2-dicarboxylate; SR-59230, 3-(2-ethylphenoxy)-1-[(1*S*)-1,2,3,4-tetrahydronaphtalen-1-yl-amino]-2*S*-2 propanol oxalate; L-748328, (*S*)-*N*-[4-[2-[[3-[3-(aminosulfonyl)phenoxy]-2-hydroxypropyl] amino] ethyl] phenyl] benzenesulfonamide; L-748337, (*S*)-*N*-[4 -[2-[[3-[3-(acetamidomethyl)-phenoxy]-2-hydroxypropyl] amino] ethyl] phenyl]benzenesulfonamide; cAMP, cyclic adenosine monophosphate; CGP-12177, (−)4-(3-*t*-butylamino-2-hydroxypropoxy)-benzimidazol-2-one; BMY-7378, (8-[2-[4-(2-methoxyphenyl)-1-piperazinyl]-ethyl]-8-azaspiro[4,5] decane-7,9-dionedihydrochloride); SHR, spontaneously hypertensive rat; WKY, Wistar-Kyoto rat; RS-17053, (*N*-[2-(2-cyclopropylmethoxyphenoxy)-ethyl]-5-chloro-α,α-dimethyl-1*H*-indole-3-ethanamine hydrochloride); U-46619, 9,11-dideoxy-11α,9α-epoxy-methano prostaglandin $F_{2\alpha}$; RWJ-38063, *N*-(2-{4-[2-(methylethoxy)phenyl] piperazinyl}ethyl)-2-(2-oxopiperidyl)acetamide; RWJ-69736, *N*-(3-{4-[2-(methylethoxy)phenyl]piperazinyl} propyl)-2-(2-oxopiperidyl)acetamide; RO-70−0004, 3-(3-{4-[fluoro-2-(2,2,2-trifluoroethoxy)-phenyl]-piperazin-1-yl}-propyl)-5-methyl-1*H*-pyrimidine-2,4-dione; RS-100329, 3-(3-{4 -[2,2,2-trifluoroethoxy)-phenyl]-piperazin-1-yl}-propyl)-5-methyl-1*H*-pyrimidine-2,4-dione; ZD-2079, ((*R*)-*N*-(2-[4-(carboxymethyl)phenoxy)] ethyl-*N*-(β-hydroxyphenethyl)ammonium chloride; LY-362884, 6-{4-[2-({2-hydroxy-3-[(2-oxo-2,3-dihydro-1*H*-benzimidazol-4-yl)oxy] propyl} amino)-2-methylpropyl]phenoxy}}nicotinamide; BRL-26830, (*R,R*)(±)-methyl-4-{2-[(2-hydroxy-2-phenyethyl)amino] propyl}benzoate; L-750355, (*S*)-*N*-[4-[2-[[3-[(2-amino-5-pyridinyl)oxy]-2-hydroxypropyl] amino]-ethyl]-phenyl]-4-isopropylbenzenesulfonamide; UK-14304, 5-bromo-6-(imidazoline-2-ylamino)quinoxaline; NO, nitric oxide; L-NAME, N^{ω}-nitro-L-arginine methyl ester; L-NMMA, N^{G}-monomethyl-L-arginine; cGMP, cyclic guanosine monophosphate; ICI-118551, ((±)-1-[2,3-(dihydro-7-methyl-1*H*-inden-4-yl)oxy]-3-[(1-methylethyl)amino]-2-butanol; CGP-20712, 2-hydroxy-5(2-(2-hydroxy-3−4((1-methyl-4-trifluoromethyl)1*H*-imidazole-2-yl)-phenoxy)propyl)amino)ethoxy)-bezamide monomethane sulfonate; ZM-215001, (*S*)-4-(2-hydroxy-3-phenoxypropylaminoethoxy)-*N*-(2-methoxyethyl)-phenoxyacetic acid; SR-58611, (*RS*)-[(25)-ethoxycarbonyl-methoxy-1,2,3,4-tetrahydronaphth-2-yl]-2-(chlorophenyl)-2 hydroethanamine hydrochloride; SR-59119, *N*-[(7-methoxy-1,2,3,4-tetrahydronaphtalen-(2*R*)-2-yl)-methyl]-(2*R*)-2-hydroxy-2(3-chlorophenyl)ethanamine hydrochloride; SR-59104, *N*-[(6-hydroxy-1,2,3,4-tetrahydronaphtalen-(2*R*)-2yl)-methyl]-(2*R*)-2-hydroxy-2-(3-chorophenyl)ethanamine hydro chloride; BRL-44408, 2-[(4,5-dihydro-1*H*-imidazol-2-yl)methyl]-2,3-dihydro-1-methyl-1*H*-isoindole; ACR-239, 2-[2,4-(2-methoxyphenyl)piperazin-1-yl] ethyl-4,4-dimethyl-1,3-(2*H*,4*H*)-isoquinolindione; MK-912, 1′,3′-dimethylspiro(1,3,4,5′,6′,7,12b)-octahydro-2*H*-benzo[*b*]furo[2,3-*a*]quinazoline)-2,4′-pyrimidin-2′one.

and important tool for a detailed study of the adrenoceptors, including those of the vascular system.

The present review aims at updating adrenoceptors in blood vessels, particularly on a functional point of view. Occasionally, some information is derived from nonvascular tissues; however, emphasis is placed on results obtained in blood vessels. Some reviews covering part of the present theme were published in the last few years (Insel, 1996; Strosberg, 1997; Summers et al., 1997; Docherty, 1998; Miller, 1998; Brodde and Michel, 1999; Bünemann et al., 1999; Freissmuth et al., 1999; Guimarães, 1999; Hein, 1999; Zhong and Minemann, 1999; Garcia-Sáinz et al., 2000; Gauthier et al., 2000; Hein, 2000; Kable et al., 2000).

II. Subclassification of Adrenoceptors

The adrenoceptors are the cell membrane sites through which noradrenaline and adrenaline act as important neurotransmitters and hormones in the periphery and in the central nervous system. The adrenoceptors are targets for many therapeutically important drugs, including those for some cardiovascular diseases, asthma, prostatic hypertrophy, nasal congestion, obesity, and pain.

The first step leading to the discovery of the adrenoceptors was made in the cardiovascular system—the observation by Dale (1905) that the pressor effect of adrenaline was reversed by ergotoxine into a depressor effect. An explanation for this phenomenon was not apparent until 43 years later! In 1948, Ahlquist noted two patterns in the relative ability of several sympathomimetic agonists to cause pharmacological responses in a series of organs and proposed the division of adrenoceptors into two types, α and β. This was subsequently confirmed by the identification of selective antagonists for these two sites: phentolamine and ergotamine for α-adrenoceptors; dichloroisoprenaline (Powell and Slater, 1958) and propranolol (Black et al., 1964) for β-adrenoceptors. Nineteen years later, it was shown that certain agonists and antagonists could distinguish β-adrenoceptor-mediated responses among tissues such as cardiac muscle and bronchial smooth muscle, implying the existence of subtypes of β-adrenoceptors (β_1 in cardiac muscle and β_2 in the bronchi) (Furchgott 1967, 1972; Lands et al., 1967a,b). Later on, the existence and differential tissue localization of α_1 and α_2 subtypes of α-adrenoceptors were discovered and characterized. The existence of subclasses of α-adrenoceptors has become evident from the results obtained by Starke and coworkers, who showed that pre- and postjunctional α-adrenoceptors differ with respect to the relative potencies of some agonists: low concentrations of clonidine and oxymetazoline selectively activate the prejunctional α-adrenoceptors, whereas phenylephrine and methoxamine selectively activate the postjunctional α-adrenoceptors (Starke, 1972; Starke et al., 1974, 1975b). Similarly, the relative potency of antagonists supported this differentiation: phenoxybenzamine was about 30 times more potent in blocking postjunctional than prejunctional α-adrenoceptors (Dubocovich and Langer, 1974) and yohimbine preferentially blocked prejunctional α-adrenoceptors (Starke et al., 1975a). Langer (1974) suggested that α-adrenoceptors mediating responses of effector organs should be referred to as α_1 and those mediating a reduction of the transmitter release during nerve stimulation as α_2. Later, it was found that α-adrenoceptors pharmacologically very similar to the prejunctional α_2-adrenoceptors are also found postjunctionally. Consequently, the nomenclature of α_1- and α_2-adrenoceptors, depending exclusively on the relative potencies of certain α-agonists and antagonists, was accepted (Berthelsen and Pettinger, 1977). In the late 1980s, the development of more selective drugs and the use of molecular cloning technology showed that there are more adrenoceptor subtypes than previously suspected. Nine different subtypes have now been cloned and pharmacologically characterized (Alexander and Peters, 1999).

A. α_1-Adrenoceptors

α_1-Adrenoceptors were first divided into two subtypes, α_{1A} and α_{1B}, based on the differential affinity of the receptors for 5-methyl urapidil (5-MU), WB-4101 (Morrow and Creese, 1986; Gross et al., 1988; Hanft and Gross, 1989; Boer et al., 1989) and the irreversible antagonist chloroethylclonidine (Han et al., 1987). α_{1A}-Adrenoceptors showed high affinity for 5-MU and WB-4101 and were insensitive to chloroethylclonidine, and α_{1B}-adrenoceptors were sensitive to CEC and had low affinity for 5-MU and WB-4101. At the present time, a consensus has been reached, such that the subdivision of α_1-adrenoceptors into three subtypes is generally accepted: α_{1A} (formerly α_{1c}; Schwinn et al., 1990), α_{1B} (Cotecchia et al., 1988), and α_{1D} (formerly $\alpha_{1a/d}$; Lomasney et al., 1991; Perez et al., 1991; Bylund et al., 1994; Ford et al., 1994). In humans, α_{1A}-, α_{1B}-, and α_{1D}-adrenoceptors are encoded by distinct genes located on chromosomes 8, 5, and 20, respectively (Hieble et al., 1995; Michel et al., 1995). Furthermore, human α_{1A}-adrenoceptor heterogeneity comes from the existence of multiple variants that differ in length and sequence of their C-terminal domains (Hirasawa et al., 1995). Additional truncated α_{1A}-adrenoceptor proteins have been reported (Chang et al., 1998). More importantly, no pharmacological or signaling differences were observed on expression of these different splice variants. According to Lattion et al. (1994), they may exhibit differential susceptibility to desensitization. A fourth α_1-adrenoceptor, the so-called α_{1L}-adrenoceptor, has been postulated (Holck et al., 1983; Flavahan and Vanhoutte, 1986a; Muramatsu et al., 1990), based exclusively on pharmacological criteria (e.g., relatively low affinity for prazosin and other antagonists such as RS-17053). This α_{1L}-adrenoceptor

seems to mediate constriction of human (Ford et al., 1996) and rabbit (Van der Graaf et al., 1997; Kava et al., 1998) lower urinary tract, guinea pig aorta (Muramatsu et al., 1990), and rat small mesenteric arteries (Stam et al., 1999). However, this hypothetical additional subtype resisted identification by biochemical and/or molecular techniques so far. Recent studies indicate that the α_{1L}-adrenoceptor may not be derived from a distinct gene, but represents a particular, energetically favorable, conformational state of the α_{1A}-adrenoceptor (Ford et al., 1998). Why these two pharmacological phenotypes occur requires further investigation (Ford et al., 1997, 1998).

It is well known that α_1-adrenoceptors are mainly coupled to $G_{q/11}$-protein to stimulate phospholipase C activity and that this enzyme promotes the hydrolysis of phosphatidylinositol bisphosphate producing inositol trisphosphate and diacylglycerol. These molecules act as second messengers mediating intracellular Ca^{2+} release from nonmitochondrial pools and activating protein kinase C, respectively (for reviews, see Hein and Kobilka, 1995; Zhong and Minneman, 1999; García-Sáinz et al., 2000). The three cloned α_1-adrenoceptor subtypes have different efficiencies in activating phospholipase C. According to Theroux et al. (1996), the ranking order of coupling efficiency (increase in inositol triphosphate formation and intracellular Ca^{2+}) after agonist occupation of recombinant α_1-adrenoceptors expressed in human embryonic kidney 293 cells was: $\alpha_{1A} > \alpha_{1B} > \alpha_{1D}$. All three α_1-adrenoceptor subtypes can couple to phospholipase C through protein $G\alpha_{q/11}$, only α_{1A}- and α_{1B}-subtypes couple to protein $G\alpha_{14}$, and only the α_{1B}-subtype couples to protein $G\alpha_{16}$ (Wu et al., 1992). Other studies support that native α_{1B}-adrenoceptors (but not α_{1A}- or α_{1D}-adrenoceptors) can also couple to protein $G\alpha_o$ in rat aorta (Gurdal et al., 1997) suggesting a functional role for this coupling. Other signaling pathways have also been shown to be activated by α_1-adrenoceptors: Ca^{2+} influx, arachidonic acid release, phospholipase D activation, and activation of mitogen-activated protein kinase (for a review, see Zhong and Minneman, 1999). Currently, no close relationship can be established between specific subtypes and signaling mechanisms.

α_1-Adrenoceptor subtypes are differentially regulated. Although the maximal down-regulation after a prolonged exposure to phenylephrine was similar for α_{1A}- and α_{1B}-adrenoceptors, the threshold concentration of phenylephrine for significant reduction was 100-fold higher for α_{1A}- than for α_{1B}-adrenoceptors. In contrast, phenylephrine up-regulated α_{1D}-adrenoceptors in a time- and concentration-dependent manner (Yang et al., 1999).

B. α_2-Adrenoceptors

It is now clear that there are three subtypes of α_2-adrenoceptors: $\alpha_{2A/D}$, α_{2B}, and α_{2C}. This subdivision, although primarily based on radioligand binding data, was preceded by results obtained in functional studies and confirmed by molecular cloning. For the α_{2B}- and α_{2C}-adrenoceptors, the pharmacological characteristics are consistent across mammalian species; however, the α_{2A}-adrenoceptor cloned from human and porcine tissue differs slightly in its amino acid composition from the homologous receptor cloned from the rat, mouse, or guinea pig in having a serine residue rather than a cysteine, at the position corresponding to Cys^{201}. To the three different genes, four pharmacological subtypes correspond since the Ser^{201} receptor possesses pharmacological properties different from the Cys^{201} receptor, and the two have been distinguished as α_{2A} (e.g., humans) and as α_{2D} (e.g., rodents) (Bylund et al., 1992; Starke et al., 1995; Trendelenburg et al., 1996; Paiva et al., 1997; Guimarães et al., 1998). These two orthologs will be simply referred to as $\alpha_{2A/D}$, unless some distinction between them has to be made. In humans, the genes coding for α_{2A}-, α_{2B}-, and α_{2C}-adrenoceptors are localized in chromosomes 10, 2, and 4, respectively (Regan et al., 1988; Lomasney et al., 1990; Weinshank et al., 1990).

Pharmacologically it is well known that the different α-adrenoceptor antagonists possess different potency/affinity for the different α_2-adrenoceptor subtypes: prazosin for example, has relatively high affinity for α_{2B}- and α_{2C}-adrenoceptors and very low affinity for α_{2A}- and α_{2D}-adrenoceptors (Latifpour et al., 1982; Nahorski et al., 1985; Bylund et al., 1988); yohimbine and rauwolscine are more potent than phentolamine and idazoxan on α_{2A}-adrenoceptors, whereas reversed relative potencies are observed for α_{2D}-adrenoceptors (Starke, 1981; Ennis, 1985; Lattimer and Rhodes, 1985; Alabaster et al., 1986; Limberger et al., 1989). The comparison of the functional potency of several antagonists with their affinity to all subtypes, as determined either in radioligand assays in native tissues possessing only one subtype or in cells transfected with recombinant α_2-adrenoceptors, shows full agreement. So, this functional approach has been extensively used to characterize α_2-autoreceptor subtypes in the different tissues (Hieble et al., 1996). Systematic studies recently undertaken to characterize prejunctional α_2-adrenoceptor subtypes in different species confirmed that receptors with α_{2A} properties occur in some species and receptors with α_{2D} properties occur in others (Bylund et al., 1994; Starke et al., 1995; Trendelenburg et al., 1996; Paiva et al., 1997; Guimarães et al., 1998) (Table 1).

However, some rare discrepancies to this postulate have been reported: in the rat vena cava (Molderings and Göthert, 1993) and rat atria (Connaughton and Docherty, 1990), where the prejunctional receptors were classified as α_{2B}, and in the human kidney cortex (Trendelenburg et al., 1994) and human right atrium (Rump et al., 1995), where they appeared to belong to the α_{2C}-subtype. However, a reinvestigation of these unexpected subclassifications showed that the prejunctional receptors in rat vena cava and atria and in guinea pig urethra were α_{2D}, and those of human kidney were α_{2A}. Thus, in

TABLE 1
Distribution of α_2-adrenoceptor subtypes in blood vessels

Species and Vessel	Functional Subtype	Reference
Prejunctional		
Rat		
Femoral vein	α_{2D}	Paiva et al., 1999
Dog		
Mesenteric artery	α_{2A}	Daniel et al., 1995
Mesenteric vein	α_{2A}	Paiva et al., 1999
Saphenous vein	α_{2A},α_{2C}	Paiva et al., 1997
Human		
Gastric artery	α_{2A}	Guimarães et al., 1998
Ileocolic artery	α_{2A}	Guimarães et al., 1998
Saphenous vein	α_{2A}	Molderings and Göthert, 1995
Postjunctional		
Mouse		
Tail artery	α_{2D},α_{2C}	Chotani et al., 2000
Pressor response (anesthetized mouse)	α_{2B},α_{2D}	Link et al., 1996
Pressor response (pithed mouse)	α_{2B},α_{2D}	McCafferty et al., 1999
Rat		
Cremaster arterioles	α_{2D}	Leech and Faber, 1996
Cremaster venules	α_{2D}	Leech and Faber, 1996
Pressor response (pithed rat)	α_{2D},α_{2B}	Gavin and Docherty, unpublished data
Dog		
Saphenous vein	α_{2A}	Hicks et al., 1991; MacLennan et al., 1997
Pig		
Common digital artery	α_{2A}	Blayloch and Wilson, 1995
Palmar lateral vein	α_{2A}	Blayloch and Wilson, 1995
Human		
Choriocapillaris (retinal pigmented epithelium)	α_{2A}	Bylund and Chacko, 1999
Saphenous vein	α_{2C}	Gavin et al., 1997

* Because α_{2A} and α_{2D}-adrenoceptors are orthologous, this table also shows the species distribution of each ortholog.

contrast to previous suggestions, all these receptors conform to the rule that α_2-autoreceptors belong, at least predominantly, to the genetic $\alpha_{2A/D}$-subtype (Trendelenburg et al., 1997).

Although the vast majority of tissues express more than one subtype, there are rare tissues expressing only one subtype: α_{2A} in human platelets (Bylund et al., 1988), α_{2B} in the rat neonatal lung (Bylund et al., 1988), α_{2C} in opossum cells (Murphy and Bylund, 1988), and α_{2D} in the rat submaxillary gland (Michel et al., 1989).

α_2-Adrenoceptors are predominantly coupled to the inhibitory heterotrimeric GTP-binding protein inhibiting the activity of adenylyl cyclase (Cotecchia et al., 1990; Wise et al., 1997), inhibiting the opening of voltage-gated Ca^{2+} channels (Cotecchia et al., 1990) and activating K^+ channels (Surprenant et al., 1992). The α_2-adrenoceptors may also couple to other intracellular pathways involving Na^+/H^+ exchange and the activation of phospholipase A_2, C, and D (Limbird, 1988; Cotecchia et al., 1990; MacNulty et al., 1992; Kukkonen et al., 1998). In neurons, α_2-adrenoceptors inhibit N-, P-, and Q-type voltage-gated Ca^{2+} channels (Waterman, 1997; Delmas et al., 1999; Jeong and Ikeda, 2000).

Like the α_1-adrenoceptors, the three α_2-adrenoceptor subtypes are regulated differentially. Human α_{2C}-adrenoceptors do not appear to down-regulate following exposure to agonists (Eason and Liggett, 1992; Kurose and Lefkowitz, 1994); $\alpha_{2A/D}$- and α_{2B}-adrenoceptors down-regulate apparently due to an increase in the rate of receptor disappearance (Heck and Bylund, 1998).

C. β-Adrenoceptors

Three distinct β-adrenoceptor subtypes have been cloned so far: β_1, β_2, and β_3 (Bylund et al., 1994). These subtypes are encoded by three different genes located on human chromosomes 10 (β_1), 5 (β_2), and 8 (β_3). The human β_3-adrenoceptor has 49 and 51% overall homology at the amino acid level with human β_2- and β_1-adrenoceptors, respectively (Emorine et al., 1989; Granneman and Lahners, 1994). Other species homologs of the human β_3-adrenoceptor have also been cloned (for a review, see Strosberg, 1997). β_1 and β_2-Adrenoceptors are well known pharmacologically since the classical papers by Lands et al. (1967a,b). They mediate cardiovascular responses to noradrenaline released from sympathomimetic nerve terminals and to circulating adrenaline. They are stimulated or blocked by many compounds that are used to treat important and common diseases, such as hypertension, cardiac arrhythmias, and ischemic heart disease.

The existence of a third β-adrenoceptor subtype (β_3-adrenoceptor), which was previously shown to mediate lipolysis in rat adipocytes (Harms et al., 1974; Arch et al., 1984; Wilson et al., 1984; Bojanic et al., 1985; Emorine et al., 1989), was also found in blood vessels where it mediates vasodilation (Cohen et al., 1984; Molenaar et al., 1988; Rohrer et al., 1999). β_3-Adrenoceptors are not blocked by propranolol, and other conventional β-adrenoceptor antagonists are activated by β_3-adrenoceptor selective agonists like BRL 37344 and CL 316243 (for reviews, see Manara et al., 1995; Strosberg, 1997; Sum-

mers et al., 1997; Fischer et al., 1998) and are blocked by β_3-adrenoceptor antagonists like SR-59230, which has been described as β_3-adrenoceptor selective in rat brown adipocytes (Nisoli et al., 1996), rat colonic motility assays (Manara et al., 1996), and human colonic circular smooth muscle relaxation activity assays (De Ponti et al., 1996). More recently, Candelore et al. (1999) did not confirm the selectivity of SR-59230 for human β_3-adrenoceptors, but described two compounds, namely L-748328 and L-748337 that display greater than 90-fold selectivity for human β_3- versus β_1-adrenoceptors, and 20- and 45-fold selectivity versus human β_2-adrenoceptors, respectively. The pharmacology of β_3-adrenoceptors is clearly distinct from that of β_1- and β_2-adrenoceptors; however, one has to bear in mind that there are differences between rodents, where β_3-adrenoceptors were studied initially, and humans, and this contributes to some confusion in the subclassification of β-adrenoceptors (Wilson et al., 1996; Arch, 1998). Furthermore, there are also differences depending on the methodological approach used. For example, the potency of catecholamines at the human β_3-adrenoceptor was found to be 1 to 2 orders of magnitude higher when determined in an intact cell cAMP accumulation assay than in a membrane-based adenylyl cyclase activation assay (Wilson et al., 1996).

On the basis of many pharmacological and molecular studies, the existence of a fourth β-adrenoceptor subtype was postulated (for reviews, see Arch and Kaumann, 1993; Barnes, 1995; Strosberg and Pietri-Rouxel, 1996; Kaumann, 1997; Strosberg, 1997; Summers et al., 1997; Galitzky et al., 1998; Strosberg et al., 1998; Brodde and Michel, 1999). These receptors would include the receptor in rat soleus muscle, which mediates glucose uptake (Roberts et al., 1993) and the receptor in human and rat heart, which mediates positive chronotropism and inotropism (Kaumann and Molenaar, 1996, 1997; Kaumann et al., 1998; Oostendorp and Kaumann, 2000) (putative β_4-adrenoceptor). A receptor cloned from turkey (β_t-adrenoceptor) has no mammalian counterpart (Chen et al., 1994). In mouse brown adipose tissue (±)-CGP-12177, a partial agonist at β_3-adrenoceptors, which is also antagonist at β_1- and β_2-adrenoceptors, evoked a full metabolic response that was of a similar magnitude in wild-type and β_3-adrenoceptor knockout mice; however, the metabolic response to CL-316243 was abolished (Preitner et al., 1998). This unexpected result supports the view that a new β-adrenoceptor, distinct from β_1-, β_2-, and β_3-adrenoceptor and referred to as putative β_4-adrenoceptor, is present in brown adipose tissue and can mediate a maximal lipolytic stimulation (Preitner et al., 1998). A similar occurrence was reported for the heart. In β_3-adrenoceptor knockout mice, CGP-12177A increased the force and rate of atrial contractions, and these effects were not antagonized by propranolol, but were antagonized by bupranolol (Kaumann et al., 1998). Furthermore, the binding of (−)-[^{3}H]CGP-12177A was similar in ventricular membranes from hearts of wild-type and β_3-adrenoceptor knockout mice; this provides evidence that the cardiac putative β_4-adrenoceptor is distinct from the β_3-adrenoceptor (Kaumann et al., 1998). More recently, evidence was obtained that this putative fourth β-adrenoceptor subtype is a particular state of β_1-adrenoceptor (see *Section III.C.1.c.*).

All β-adrenoceptor subtypes signal by coupling to the stimulatory G-protein $G_{\alpha s}$ leading to activation of adenylyl cyclase and accumulation of the second messenger cAMP (Dixon et al., 1986; Frielle et al., 1987; Emorine et al., 1989). However, some recent studies indicate that, under certain circumstances, β-adrenoceptors, and particularly the β_3-adrenoceptor, can couple to G_i as well as to G_s (Asano et al., 1984; Chaudry et al., 1994; Xiao et al., 1995; Gauthier et al., 1996).

Intracellular events following β-adrenoceptor activation are also linked to ion transport. It is well known, for example, that protein kinase A activated by cAMP phosphorylates L-type Ca^{2+} channels, facilitating Ca^{2+} entry, and producing the positive inotropic effect in atria and ventricles, increased heart rate in the sino-auricular node, and accelerated the conduction in the atrio-ventricular node. In addition to mechanisms that indirectly lead to alterations in ion transport, β-adrenoceptor activation is more directly linked to ion channels: β-adrenoceptor stimulation is able to activate L-type Ca^{2+} channels via $G_{\alpha s}$ (Brown, 1990); in airway smooth muscle, β-adrenoceptor activation opens Ca^{2+}-dependent K^+ channels and charybdotoxin—a specific inhibitor of the high conductance Ca^{2+}-activated K^+ channel—antagonizes the relaxant effects of β-adrenoceptor agonists (Miura et al., 1992; Jones et al., 1993).

Multiple mechanisms control the signaling and density of G-protein-coupled receptors. The termination of G-protein-coupled receptor signals involves binding of proteins to the receptor. This process is initiated by serine-threonine phosphorylation of agonist-occupied receptors, both by members of the G-protein-coupled receptor kinase family and by second-messenger-activated protein kinases such as protein kinase A and protein kinase C. Receptor phosphorylation by G-protein-coupled receptor kinase is followed by binding of proteins termed arrestins, which bind to the phosphorylated receptor and sterically inhibit further G-protein activation (Luttrell et al., 1999). Desensitized receptor-arrestin complexes undergo arrestin-dependent targeting for sequestration through clathrin-coated pits (Goodman et al., 1996; Luttrell et al., 1999). Sequestrated receptors are ultimately either dephosphorylated and recycled to the cell surface or targeted for degradation (Luttrell et al., 1999).

In addition, many other G-protein-coupled receptors are sequestrated from the cell membrane and become inaccessible to their ligands. Both receptor/G-protein uncoupling and receptor sequestration may involve the participation of arrestins or other proteins. A model for

receptor regulation has been developed on the basis of data from studies of the β-adrenoceptors. However, according to recent reports, other G-protein-coupled receptors, like muscarinic receptors in the cardiovascular system, may be regulated by mechanisms other than those that regulate the β-adrenoceptors (for a review, see Bünemann et al., 1999).

III. Postjunctional Adrenoceptors in Vascular Smooth Muscle

Because vascular smooth muscles possess both α- and β-adrenoceptors, the net response to agonists that like adrenaline stimulate both types of receptors depends on the relative importance of each population. For example, while in the dog saphenous vein, in vitro adrenaline causes contraction, which is enhanced by β-adrenoceptor blockade (Guimarães, 1975); in the rabbit facial vein, adrenaline causes relaxation, which is enhanced by α-adrenoceptor blockade (Pegram et al., 1976). On the other hand, the contractile response of the saphenous vein to adrenaline is converted into a relaxation when an α-adrenoceptor antagonist is present (Guimarães and Paiva, 1981a), and the relaxation caused by adrenaline in the rabbit facial vein is converted into a contraction when a β-adrenoceptor antagonist is present (Pegram et al., 1976). Thus, while in the dog saphenous vein, the α-adrenoceptor-mediated influence dominates, in the rabbit facial vein the dominating influence is exerted by β-adrenoceptor.

In the vast majority of vascular tissues, α-adrenoceptor-mediated effects predominate, such that to demonstrate in vitro β-adrenoceptor-mediated responses using adrenaline as agonist, both α-adrenoceptor blockade and active tone of the tissue must be present. When a pure or almost pure β-adrenoceptor agonist like isoprenaline is used, the only requirement to obtain β-adrenoceptor-mediated responses is the presence of tone. The threshold for α-adrenoceptor-mediated effects in large arteries and veins is between 1 and 10 nM noradrenaline (Guimarães, 1975; Bevan, 1977). The levels of noradrenaline and adrenaline in human arterial plasma at rest are about 2 and 0.5 nM, respectively (Engleman and Portnoy, 1970; DeQuattro and Chan, 1972). In the dog, the level of noradrenaline is similar. Thus, at rest, most vessels are scarcely influenced by circulating catecholamines. However, in the rat mesentery, precapillary sphincters have a threshold response to adrenaline and noradrenaline of 0.1 to 1 nM (Altura, 1971), and rat plasma adrenaline and noradrenaline levels average 2.5 and 3 nM, respectively (Donoso and Barontini, 1986). Although in vivo sensitivity cannot be directly related to plasma catecholamine levels, these data suggest that precapillary sphincters may be affected by circulating catecholamines even under resting conditions, in contrast to other vessels. In humans, during exercise, plasma noradrenaline and adrenaline may reach levels 30 times higher than those at rest, which may have a profound effect on vessels.

A. α_1-Adrenoceptors

It is important to underline that many of the advances made in the last years in the field of receptors in general and on vascular adrenoceptors in particular were due to the possibility to generate knockout mice. However, one should not forget that the lack of a given receptor from conception may be compensated by adequate adjustments, whereas its functional elimination by an antagonist is not acutely compensated (Rohrer and Kobilka, 1998). This is something one must bear in mind when results obtained in wild-type mice are compared with results obtained in knockout mice. It is dangerous to assume that knockout animals differ from the wild-type by no more than the absence of one receptor subtype.

1. In Vitro. In most mammalian species, contraction of vascular smooth muscle is predominantly mediated via α_1-adrenoceptors. Although the existence of both α_1- and α_2-adrenoceptors has been shown by functional studies in vivo, it has been difficult to demonstrate functional postjunctional α_2-adrenoceptors in most arteries in vitro (De Mey and Vanhoutte, 1981; McGrath, 1982; Timmermans and van Zwieten, 1982; Polónia et al., 1985; Guimarães, 1986; Aboud et al., 1993; Burt et al., 1995, 1998). In isolated canine aorta and canine femoral, mesenteric, jejunal, renal, and splenic arteries, contractile responses were exclusively α_1-adrenoceptor-mediated (Polónia et al., 1985; Shi et al., 1989; Daniel et al., 1999). In the arteries of other mammalian species, α_1-adrenoceptors also predominate: in rat aorta (Han et al., 1990; Aboud et al., 1993); in rat carotid, mesenteric, renal, and tail arteries (Han et al., 1990; Villalobos-Molina and Ibarra, 1996); and in human arteries (Flavahan et al., 1987a).

In veins, particularly in cutaneous veins, at the postjunctional level, α_1- and α_2-adrenoceptors both contribute to vasoconstriction (Flavahan and Vanhoutte, 1986a; Guimarães et al., 1987). In dog and human saphenous veins, α_2-adrenoceptors are the predominant receptors mediating contraction (Müller-Schweinitzer, 1984; Guimarães and Nunes, 1990; Docherty, 1998).

The question of which α_1-adrenoceptor subtype is involved in vasoconstrictive responses to sympathomimetic agonists is not easy to answer. Vascular smooth muscle tissues express mixtures of α_1-adrenoceptor subtypes (Miller et al., 1996) and in most cases responses to α_1-adrenoceptor agonists are probably due to activation of more than one subtype (Van der Graaf et al., 1996a; Zhong and Minneman, 1999). mRNA for the α_{1A}-adrenoceptor is expressed at very high levels in peripheral arteries, around 90% of the total α_1-adrenoceptors message pool (Guarino et al., 1996), but in most cases, there is lack of correlation between protein expression of one adrenoceptor subtype and the function this receptor mediates (Hrometz et al., 1999; Ohmi et al., 1999). The rat

is a good case to exemplify this pharmacological problem (Aboud et al., 1993; Kong et al., 1994; Saussy et al., 1996; Piascik et al., 1997; Stam et al., 1999). Despite the fact that the mRNA for all three cloned α_1-adrenoceptor subtypes has been found in the rat mesenteric artery, as well as the aorta and pulmonary artery (Xu et al., 1997), the contraction in response to phenylephrine in these three vessels is primarily α_{1D}-adrenoceptor-mediated, α_{1B}-adrenoceptor being secondarily involved (Hussain and Marshall, 2000). A similar lack of correlation was demonstrated in a study involving several arteries of the rat: although in terms of level of mRNA expression for α_1-adrenoceptor subtypes, the ranking order was α_{1A}- > α_{1B}- > α_{1D}, only α_{1B}-adrenoceptors played a functional role in mesenteric resistance artery, whereas α_{1D}-adrenoceptors were implicated in mediating the contraction of the aorta and femoral, iliac, and superior mesenteric arteries (Piascik et al., 1997). Similarly, in the rabbit, all α_1-adrenoceptor subtypes coexist in the aorta and in the mesenteric, renal, and iliac arteries. However, although the renal and iliac arteries contract predominantly via the activation of α_{1D}-adrenoceptors in response to noradrenaline and secondarily via activation of α_{1A}- and α_{1B}-adrenoceptors, the aorta contracts via the activation of α_{1A}- and α_{1B}-adrenoceptors (Satoh et al., 1998, 1999). According to functional results, it seems that in the rat the α_{1A}- and α_{1D}-adrenoceptor subtypes regulate the larger vessels, whereas the α_{1B}-adrenoceptors control the small resistance vessels (Leech and Faber, 1996; Piascik et al., 1997; Gisbert et al., 2000). In the dog mesenteric artery, α_1-adrenoceptors are predominantly of the α_{1A}-subtype (Daniel et al., 1999).

Table 2 summarizes the α_1-adrenoceptors subtypes primarily responsible for the contractile responses of the main arteries from species mostly currently used in research: α_{1A}- and α_{1D}-subtypes are those mainly involved in the contractions evoked by α_1-adrenoceptor agonists.

α_1-Adrenoceptors are also involved in the regulation of vascular smooth muscle growth. Findings by some authors suggest that prolonged stimulation of chloroethylclonidine-sensitive, possibly α_{1B}-adrenoceptors, induce hypertrophy of arterial smooth muscle cells, whereas stimulation of α_{1A}-adrenoceptors attenuates this growth response (Chen et al., 1995; Siwik and Brown, 1996).

2. In Vivo. There is also longstanding evidence that multiple α_1-adrenoceptor subtypes are involved in the regulation of peripheral vascular function in vivo (McGrath, 1982; Minneman, 1988; Bylund et al., 1995b). However, the individual contribution of each of the α_1-adrenoceptor subtypes has not been established. Of the three known α_1-adrenoceptor subtypes, α_{1A}- and α_{1D}-adrenoceptors have most often been implicated in the regulation of vascular smooth muscle tone (see Table 2). There are discrepancies between results obtained in vitro and in vivo involving α_1-adrenoceptors. Although in vitro studies in rats had indicated a predominant role of the α_{1D}-adrenoceptor in the vascular contractions caused by α_1-adrenoceptor agonists (Piascik et al., 1995; Hussain and Marshall, 2000), surprisingly experiments in α_{1B}-knockout mice show that the maximal contractile response of aortic rings to phenylephrine was reduced by 40% and the mean arterial blood pressure response to phenylephrine was decreased by 45%, showing that the α_{1B}-adrenoceptor is important for blood pressure and the contractile response of the aorta evoked by α_1-adrenoceptor agonists (Cavalli et al., 1997). In the pithed rat, the systemic blood pressure is tonically regulated by the interaction of peripheral sympathetic nerves with vascular α_{1A}-adrenoceptors (Vargas et al., 1994), although vascular α_{1D}-adrenoceptors have a role in the pressor response to phenylephrine (Zhou and Vargas, 1996). Also, in the pithed rat, it was shown that the selective α_{1D}-adrenoceptor antagonist BMY-7378 not only antagonized the pressor effect of phenylephrine, but also was more potent in young prehypertensive spontaneously hypertensive rats (SHRs) than in young WKY rats. The presence of α_{1D}-adrenoceptors in the resistance vasculature of prehypertensive and hypertensive rats may indicate that they are involved in the development/maintenance of hypertension (Villalobos-Molina et al., 1999). Thus, it may be concluded that, in rats in vivo, the pressor response to phenylephrine is mediated by vascular α_{1A}- and α_{1D}-adrenoceptors (Vargas et al., 1994; Guarino et al., 1996; Zhou and Vargas, 1996).

In human vasculature, as in that of other mammals, α_1-adrenoceptors play a crucial role in the regulation of vascular tone. In healthy volunteers, Schäfers et al. (1997, 1999) showed that, whereas 2 mg of doxazosin (a selective α_1-adrenoceptor antagonist) nearly completely antagonized the blood pressure increasing effect of i.v. administered noradrenaline (10 to 160 ng/kg · min), 15 mg of yohimbine (a selective α_2-adrenoceptor antagonist) only slightly attenuated noradrenaline effect. With regard to this finding, one should bear in mind that the administration of exogenous noradrenaline does not necessarily result in identical concentrations in the biophase of the postjunctional α_1- and α_2-adrenoceptors; there may develop a certain ratio biophase α_1/biophase α_2. Moreover, this ratio may be different for noradrenaline released from sympathetic nerves (see Distribution of vascular adrenoceptors). The available information regarding α_1-adrenoceptor subtypes mediating vasoconstriction in humans is still very scarce.

In conclusion to the role played by each α_1-adrenoceptor subtype in the maintenance of vascular tone and in vascular responses to α_1-adrenoceptor ligands, one can say that there is a lack of correlation between two sets of results disturbing their interpretation. First, the lack of correlation between protein expression of a given adrenoceptor and the functional role this adrenoceptor plays; second, the lack of correlation between the results obtained in vitro (Table 2) and in vivo. Despite that, according to the vast majority of the authors, it seems that

TABLE 2
Distribution of α_1-adrenoceptor subtypes in blood vessels

Species and Vessel	Receptor Subtype		Reference
	mRNA or Protein	Functional	
Mouse			
AC01 cells[a]		α_{1D}	Ohmi et al., 1999
Rat			
Aorta	α_{1A}, α_{1B}, α_{1D}*	α_{1D}**	Hrometz et al., 1999*; Kenny et al., 1995**; Piascik et al., 1995**; Testa et al., 1995**;
		α_{1A}, α_{1B}, α_{1D}***	Van der Graaf et al., 1996a***
Carotid artery		α_{1D}	Villalobos-Molina and Ibarra, 1996
Mesenteric artery	α_{1A}, α_{1B}, α_{1D}*	α_{1D}, α_{1A}, α_{1B}(?)**	Hrometz et al., 1999*; Villalobos-Molina and Ibarra, 1996**; Lachnitt et al., 1997**; Piascik et al., 1997**
Tail artery	α_{1A}, α_{1B}, α_{1D}*	α_{1A}**	Hrometz et al., 1999*; Lachnit et al., 1997**; Piascik et al., 1997**
Renal artery	$\alpha_{1B} > \alpha_{1D} > \alpha_{1A}$*	α_{1A}, α_{1D}**	Hrometz et al., 1999*; Han et al., 1990**; Piascik et al., 1997**
Iliac artery	α_{1A}, α_{1B}, α_{1D}*	α_{1D}**	Hrometz et al., 1999*; Piascik et al., 1997**
Femoral artery	$\alpha_{1B} > \alpha_{1A} > \alpha_{1D}$*	α_{1D}**	Hrometz et al., 1999*; Piascik et al., 1997**; Hrometz et al., 1999**
Pulmonary artery		α_{1D}	Hussain and Marshall, 1997
Mesenteric resistance arteries	α_{1A}, α_{1B}, α_{1D}*	α_{1B}**	Hrometz et al., 1999*; Van der Graaf et al., 1996b**; Piascik et al., 1997**
Skeletal muscle arteries		α_{1D} (α_{2D})	Leech and Faber, 1996
Skeletal muscle veins		α_{1B}, α_{1D}	Leech and Faber, 1996
Vena cava		α_{1B}	Sayet et al., 1993
Guinea pig			
Aorta		α_{1L}	Yamamoto and Koike 1999
Nasal mucosa vasculature		α_{1A} (α_{1L}) (?)	Tanimitsu et al., 2000
Rabbit			
Thoracic aorta		α_{1A}, α_{1D}, α_{1B}, α_{1L}	Takayanagi et al., 1991; Fagura et al., 1997; Muramatsu et al., 1998
Carotid artery		α_{1L}	Muramatsu et al., 1990
Abdominal aorta		α_{1A}, α_{1B}	Satoh et al., 1999
Iliac artery		α_{1D}, α_{1A}, α_{1B}	Satoh et al., 1999
Mesenteric artery		α_{1L}, α_{1D}	Van der Graaf et al., 1997; Satoh et al., 1999
Renal artery		α_{1D}, α_{1A}, α_{1B}	Satoh et al., 1999
Ear artery		α_{1A}, α_{1D} (?)	Fagura et al., 1997
Cutaneous resistance arteries		α_{1B}, α_{1L}	Smith et al., 1997
Dog			
Aorta		α_{1B}	Low et al., 1998
Lingual artery		α_{1A}	Skrbic and Chiba, 1992
Mesenteric artery		α_{1A}	Daniel et al., 1999
Subcutaneous resistance arteries		α_{1A}/α_{1L}	Argyle and McGrath, 2000
Saphenous vein		α_{1A}	Hicks et al., 1991
Pig			
Coronary artery		α_{1A}	Yan et al., 1998
Human			
Aorta	α_{1D}		Rudner et al., 1999
Carotid artery	α_{1A}		Rudner et al., 1999
Femoral artery	$\alpha_{1A} > \alpha_{1D}$		Rudner et al., 1999
Iliac artery	α_{1A}		Rudner et al., 1999
Mammary artery	$\alpha_{1A} > \alpha_{1B} > \alpha_{1D}$		Rudner et al., 1999
Celiac artery	$\alpha_{1A} > \alpha_{1B}$		Rudner et al., 1999
Hepatic artery	α_{1A}		Rudner et al., 1999
Mesenteric artery	$\alpha_{1A} \gg \alpha_{1B}$		Rudner et al., 1999
Splenic artery	$\alpha_{1A} > \alpha_{1B}$		Rudner et al., 1999
Omental artery	$\alpha_{1A} > \alpha_{1B}$, α_{1D}		Rudner et al., 1999
Renal artery	$\alpha_{1A} > \alpha_{1B}$		Rudner et al., 1999
Pulmonary artery	$\alpha_{1A} > \alpha_{1B}$, α_{1D}		Rudner et al., 1999
Right coronary artery	α_{1A}		Rudner et al., 1999
Left coronary artery	α_{1A}		Rudner et al., 1999
Circumflex artery	α_{1A}		Rudner et al., 1999
Lingual artery		α_{1A}, α_{1B}	Skrbic and Chiba, 1992
Iliac vein	α_{1A}, α_{1B}, α_{1D}		Rudner et al., 1999
Vena cava	$\alpha_{1A} \gg \alpha_{1D}$		Rudner et al., 1999
Saphenous vein	α_{1A}, α_{1B}		Rudner et al., 1999
Omental vein	α_{1A}		Rudner et al., 1999
Renal vein	None		Rudner et al., 1999
Pulmonary vein	$\alpha_{1A} > \alpha_{1B} > \alpha_{1D}$		Rudner et al., 1999
Prostate vessels		α_{1A} (α_{1L}) (?)	Marshall et al., 1995

[a] A novel vascular smooth muscle cell line cloned from p53 knockout mice (Ohmi et al., 1999).

in the rat α_{1A}-adrenoceptors have a prominent role in the regulation of blood pressure, although α_{1B}- and α_{1D}-adrenoceptors are also functionally present and participate in the responses to exogenous agonists (Piascik et al., 1990; Schwietert et al., 1992; Vargas et al., 1994; Guarino et al., 1996; Zhou and Vargas, 1996).

3. α_1-Adrenoceptor Antagonists in the Symptomatic Treatment of Prostatic Hypertrophy. Clinical interest in this target comes from the fact that selective α_{1A}-adrenoceptor antagonists may have significant therapeutic advantages over nonsubtype selective α_1-adrenoceptor antagonists in the treatment of benign prostatic hypertrophy. Which is the basis for the hypothetical differential effect of α_{1A}-adrenoceptor antagonists at vascular tissue and prostate? Are α_1-adrenoceptors of vascular- and prostatic smooth muscle different? Several studies have shown that the α_{1A}-adrenoceptor subtype accounts for the majority of α_1-adrenoceptor mRNAs and expressed protein in human prostatic smooth muscle and mediates contraction in this tissue (Price et al., 1993; Faure et al., 1994; Lepor et al., 1995; Michel et al., 1996; Schwinn and Kwatra, 1998). However, recent experiments carried out in rat mesenteric arteries (a tissue the α_1-adrenoceptors of which, like those of the prostate, have low affinity for prazosin and RS-17053) (Ford et al., 1996), showed that the affinity of prazosin and RS-17053 was not altered by changing the experimental conditions (lowering temperature, inducing tone via KCl or U-46619—a derivative of prostaglandin $F_{2\alpha}$), calling again our attention to the problem of the putative α_{1L}-adrenoceptors (Yousif et al., 1998; Stam et al., 1999). On the other hand, which is the α_1-adrenoceptor subtype that mediates contractile vascular responses in humans? The few reports on α_1-adrenoceptors in resistance arteries failed to show that a particular α_1-adrenoceptor subtype is of primary importance in the sympathetic control of these vessels. Probably, as animal studies have suggested, each vessel possesses mixtures of α_1-adrenoceptor subtypes, and responses to α_1-adrenoceptor agonists are due to stimulation of more than one subtype (Michel et al., 1998b; Ruffolo and Hieble, 1999; Zhong and Minneman, 1999; Argyle and McGrath, 2000). In a very recent study, it was shown that the receptor subtype mediating the constriction of canine resistance vessels is an α_{1A}-/α_{1L} adrenoceptor (Argyle and McGrath, 2000), which is the same that has been proposed as mediating the adrenergic responses in prostate (McGrath et al., 1996). Thus, the relative selectivity of α_{1A}-adrenoceptor antagonists, if there is any, may not depend on differences between subtypes, but rather on differences between local functional expressions of the receptors. In single human prostatic smooth muscle cells, MacKenzie et al. (2000) showed that the affinity of a prazosin analog for native human α_{1A}-adrenoceptors was higher than for human cloned α_{1A}-adrenoceptors expressed in cell cultures. This suggests that a tissue-specific affinity state of the same receptor genotype exists, and this could be a potential differentiator of drug action (MacKenzie et al., 2000).

Halotano et al. (1994) reported a slightly lower potency for 5-MU and WB-4101 in the human iliac artery, compared with the human urethra suggesting a therapeutic benefit in prostatic symptoms without causing the vascular side effects associated with α_1-adrenoceptor blockade. However, the degree of selectivity of the different compounds until now available to treat benign prostatic hypertrophy (doxazosin, alfusosin, terazosin) is not enough to eliminate cardiovascular side effects, such as dizziness, orthostatic hypotension, asthenia, and occasionally syncope (Michel et al., 1998a; Chapple and Chess-Williams, 1999; Pulito et al., 2000). The moderately selective α_{1A}-adrenoceptor antagonist tamsulosin has been introduced for this purpose (Foglar et al., 1995). When directly comparing equieffective dosis of terazosin (a selective α_1-adrenoceptor antagonist) with tamsulosin in patients with prostatic hyperplasia, Lee and Lee (1997) observed that tamsulosin caused significantly fewer side effects; however, Schäfers et al. (1998) less enthusiastically concluded that further experimental and clinical work was required to unequivocally demonstrate this advantage of selective α_{1A}-adrenoceptor antagonists. Very recently, the selectivity of tamsulosin, doxazosin, and alfuzosin was determined by comparing their effects on the human prostate and human mesenteric arteries in vitro. It was observed that tamsulosin exhibited a 10-fold selectivity for the prostate over the artery, a degree of selectivity that was compatible with its claimed clinical benefit (Davis et al., 2000). A possible explanation for the clinical advantages of tamsulosin was given by Hein et al. (2001), who showed that the α_1-antagonist with the least vascular effects in humans in vivo also was the drug with the least inverse agonism in vitro (tamsulosin).

Some new aryl piperazine compounds were recently synthesized, which in binding experiments to recombinant human α_1-adrenoceptors showed high α_{1A}-adrenoceptor subtype selectivity (Pulito et al., 2000). Furthermore, some of them were more potent in inhibiting noradrenaline-evoked contraction of rat prostate tissue than those of rat aorta tissue: RWJ-38063 and RWJ-69736 were 319- and 100-fold more potent in their effects on prostate tissue than aorta tissue. In anesthetized dogs, both compounds suppressed the intraurethral pressure response to phenylephrine to a greater extent than the mean arterial pressure response (Pulito et al., 2000). Other new compounds like RO-70−0004 and RS-100329 (Williams et al., 1999) and some aryldihydropyrimidinones (Barrow et al., 2000) also show an approximately 100-fold selectivity for α_{1A}- versus α_{1B}- and α_{1D}-adrenoceptor subtypes.

α_1-Adrenoceptor agonists have been used clinically in the treatment of stress incontinence, acting to increase urethral tone by contracting urethra smooth muscle. Efforts are also being made to identify agents of this kind, selective enough to act on the urethra without causing increases in blood pressure (Ruffolo and Hieble, 1999).

B. α_2-Adrenoceptors

1. In Vitro. At the postjunctional level, α_2-adrenoceptors were not found in vitro in the vast majority of the arterial vessels (Table 1). No constrictor activity of α_2-adrenoceptor agonists is present in large arteries; when it appears, it is generally restricted to small arteries/arterioles (Docherty and Starke, 1981; Polónia et al., 1985; Aboud et al., 1993; Leech and Faber, 1996; Daniel et al., 1999). Using rabbit polyclonal antibodies for the α_2-adrenoceptor subtypes, it was observed that $\alpha_{2A/D}$- and α_{2C}-adrenoceptors are present in the smooth muscle of mouse tail arteries, the expression of α_{2C}-adrenoceptors being smaller in distal arteries than in proximal arteries (Chotani et al., 2000). In contrast to the difficulty in demonstrating postjunctional α_2-adrenoceptors in arteries in vitro, they are consistently found in many isolated veins of different species (De Mey and Vanhoutte, 1981; Constantine et al., 1982; Shoji et al., 1983; Guimarães et al., 1987). This is why the characterization of α_2-adrenoceptor subtypes involved in vascular responses to sympathomimetic agonists is being made in veins (or in vivo). The $\alpha_{2A/D}$-subtype is the predominant one in almost all the veins until now studied: $\alpha_{2A/D}$ in dog saphenous vein (Hicks et al., 1991; MacLennan et al., 1997); $\alpha_{2A/D}$ in rabbit skeletal muscle venules (although predominantly α_{1D}) (Leech and Faber, 1996); and $\alpha_{2A/D}$ (most probably) in the porcine palmar lateral vein (Blaylock and Wilson, 1995). In good agreement with the premise that the α_{2A}- and α_{2D}-adrenoceptors represent species orthologs (Bylund et al., 1995a)—α_{2A} occurring in humans, dogs, pigs, and rabbits and α_{2D} occurring in rats, mice, and cows—it was observed that postjunctional α_2-adrenoceptors of the canine mesenteric vein are predominantly α_{2A}, whereas those of the rat femoral vein are predominantly α_{2D} (Paiva et al., 1999). In human saphenous vein, correlation of α_2-adrenoceptors antagonist potency with binding affinity suggests the contribution of the α_{2C}-subtype (Gavin et al., 1997).

2. In Vivo. α_2-Adrenoceptors are essential components of the neural complex system regulating cardiovascular function (Ruffolo et al., 1991) (see *Section IV.*). When clonidine-like α_2-adrenoceptor agonists are intra-arterially administered to wild-type mice, they cause an initial brief pressor effect that is gradually reversed to hypotension at the same time as the animal experiences a severe bradycardia (Link et al., 1996; MacMillan et al., 1996). This is a typical cardiovascular response to intravenous administration of an α_2-adrenoceptor agonist also in humans and other species (Hoefke and Kobinger, 1966; Kallio et al., 1989; Bloor et al., 1992). Three main factors are now known to participate in this biphasic response: activation of α_2-adrenoceptors on vascular smooth muscle cells that is responsible for the initial and transient hypertensive phase; activation of α_2-adrenoceptors in the brainstem leading to a reduction in sympathetic tone with a resultant decrease in blood pressure and heart rate (this hypotensive effect has been the rationale for the use of clonidine in the treatment of hypertension); and a third factor, namely the stimulation of prejunctional α_2-adrenoceptors located on sympathetic terminals innervating the vascular smooth muscle cells, an effect that augments the hypotensive effect due to stimulation of central α_2-adrenoceptors (Ruffolo et al., 1993; Urban et al., 1995). Bearing in mind that all the three subtypes of α_2-adrenoceptors ($\alpha_{2A/D}$, α_{2B}, and α_{2C}) are present in the vascular tree (Hicks et al., 1991; Gavin et al., 1997; MacLennan et al., 1997; Paiva et al., 1999), which is the involvement of each subtype, if any, in these responses?

The lack of subtype-selective ligands and the cross-reactivity of α_2-adrenoceptor agonists with imidazoline receptors made it impossible, until the recent development of knockout animals, to assess the involvement of the individual α_2-adrenoceptor subtypes not only in the hypotensive response to α_2-adrenoceptor agonists, but also in the general physio-pharmacology of the cardiovascular system.

3. Blood Pressure Regulation in α_2-Adrenoceptor-Deficient Mice. Five mouse strains with genetic alterations of α_2-adrenoceptor expression have been generated; they offer new pathways to further identify the role of each subtype (Hein, 1999; Kable et al., 2000): deletion of $\alpha_{2A/D}$- ($\alpha_{2A/D}$-knockout), α_{2B}- (α_{2B}-knockout), or α_{2C}-gene (α_{2C}-knockout) (Link et al., 1995, 1996; Altman et al., 1999). More recently, mouse strains lacking $\alpha_{2A/D}$-, α_{2B}-, or α_{2C}-subtypes were crossed to generate double knockout mice. However, the only viable animals were those lacking both $\alpha_{2A/D}$- and α_{2C}-adrenoceptors ($\alpha_{2A/DC}$-knockout) (Hein et al., 1999). Mice have been also developed with a point mutation of the $\alpha_{2A/D}$-gene ($\alpha_{2A/D}$-D79N). The D79N mutation causes a replacement of the aspartate with an asparagine residue at position 79, in the second transmembrane domain of the $\alpha_{2A/D}$-adrenoceptor. In cultured cell lines, the $\alpha_{2A/D}$-D79N mutant receptor failed to activate K^+ currents, but exhibited normal inhibition of voltage-gated calcium channels and cAMP production (Surprenant et al., 1992). It was expected that the expression of this mutation in the intact animal would provide insight into the signal transduction mechanisms mediating the effect of α_{2A}-adrenoceptor stimulation. However, $\alpha_{2A/D}$-D79N mice showed about an 80% reduction in α_2-adrenoceptor binding, as determined by radioligand studies in the brain (MacMillan et al., 1996). Thus, the $\alpha_{2A/D}$-D79N receptor expressed in vivo showed different characteristics, compared with its expression in vitro, thus behaving as a functional knockout.

In these D79N $\alpha_{2A/D}$-adrenoceptor mice, the hypotensive response to intra-arterial infusion of α_2-adrenoceptor agonists was almost absent, while the initial hypertensive response remained unchanged. This alteration in the cardiovascular response demonstrates that $\alpha_{2A/D}$-

adrenoceptors mediates the brainstem hypotensive response not only to endogenous catecholamines, but also to imidazoline-based α_2-adrenoceptor agonists (MacMillan et al., 1996). Accordingly, any hypothetical important role of the so-called imidazoline receptors as mediators of this effect can be ruled out.

As an alternative approach, knockout mice deficient in $\alpha_{2A/D}$-, α_{2B}-, or α_{2C}-adrenoceptors were developed (Link et al., 1996). All these different strains of knockout animals are viable, fertile, and develop normally (Hein et al., 1998).

In α_{2B}-knockout mice, the initial pressor response to α_2-adrenoceptor agonists was abolished and the hypotensive effect occurred immediately and was significantly greater than that observed for control animals showing that the initial hypertensive phase was due to activation of α_{2B}-adrenoceptors on vascular smooth muscle and that the vasoconstrictory α_{2B}-adrenoceptors in the peripheral vasculature counteract the therapeutic hypotensive action of α_2-adrenoceptor agonists (Link et al., 1996; Hein et al., 1998). Thus, if $\alpha_{2A/D}$-adrenoceptor agonists are developed selective enough to avoid α_{2B}-adrenoceptor stimulation, their antihypertensive effect should be enhanced. The bradycardia evoked by α_2-adrenoceptor agonists in α_{2B}-knockout mice was not changed.

Mice with a deletion of the α_{2C}-adrenoceptor gene showed no differences from wild-type mice in their hypertensive, hypotensive, and bradycardic responses to α_2-adrenoceptor agonists (Link et al., 1996; Hein et al., 1998). Results obtained in $\alpha_{2A/D}$-knockout mice confirmed and extended those obtained with $\alpha_{2A/D}$-D79N mice, demonstrating that most of the classical effects ascribed to α_2-adrenoceptor agonists are mediated by $\alpha_{2A/D}$-adrenoceptors. Both $\alpha_{2A/D}$-D79N and $\alpha_{2A/D}$-knockout mice failed to become hypotensive in response to exogenous α_2-adrenoceptor agonists (MacMillan et al., 1996; Altman et al., 1999). However, whereas the heart rate of $\alpha_{2A/D}$-D79N mice was not significantly different from their controls—indicating that prejunctional regulation of catecholamine release was preserved—$\alpha_{2A/D}$-knockout mice had: 1) basal resting heart rates more than 180 beats/min greater than their control littermates ($\alpha_{2A/D}$-knockout 581 $\pm$ 21/min versus wild-type 395 $\pm$ 21/min) (Hein et al., 1998); 2) a significant depletion of tissue noradrenaline stores, which can be ascribed to an enhanced release of noradrenaline from sympathetic nerves; and 3) higher plasma noradrenaline levels, as indicated by a 25% reduction in the density of β-adrenoceptors (Altman et al., 1999). All these changes can be explained by a higher basal level of sympathetic tone resulting from the loss of $\alpha_{2A/D}$-adrenoceptor-mediated inhibition of the vasomotor center.

It is somewhat surprising that resting blood pressure was unaffected in $\alpha_{2A/D}$-knockout mice. Several factors can be advanced to explain the lack of an hypertensive response to the disappearance of $\alpha_{2A/D}$-adrenoceptors: first of all, the main vasoconstrictory influence exerted by the sympathetic nervous system is mediated through α_1-adrenoceptors (Rohrer et al., 1998a); and some other changes like the redistribution of blood to individual vascular beds (which differ with respect to their α_2-adrenoceptor population) (MacMillan et al., 1996) and some compensatory adjustments from the renin-angiotensin and nitric oxide systems may also contribute to keep the resting blood pressure at roughly its normal value (Cavalli et al., 1997; Altman et al., 1999). It is also difficult to interpret the role played by tachycardia; it will depend on the amount of venous blood delivered to the heart. Interestingly, a study in which the effect of antisense to $\alpha_{2A/D}$-adrenoceptors was checked in the rat, the antisense sequence when given intrathecally, caused an increase of the systolic blood pressure (Nunes, 1995), indicating that in this animal species central α_2-adrenoceptors regulating blood pressure belong to the $\alpha_{2A/D}$-subtype. Moreover, this increase in blood pressure suggests that the centrally mediated hypotensive effect of α_2-agonists may be more important in the rat than in the mouse.

C. β-Adrenoceptors

1. In Vitro

a. β_1- and β_2-Adrenoceptors. According to Lands and coworkers (1967a,b), the β-adrenoceptors in the peripheral vessels were classified as β_2. Later studies using more selective agonists and antagonists showed that relaxation of vascular smooth muscle cells resulted from activation of either β_1- or β_2-adrenoceptor subtypes, and that the involvement of each subtype depended on the vascular bed and the species (O'Donnell and Wanstall, 1984; Guimarães et al., 1993; Shen et al., 1994, 1996; Begonha et al., 1995). β_2-Adrenoceptors represent the predominant subtype in most vascular smooth muscles, although β_1-adrenoceptors may contribute to vasodilation (for a review, see Osswald and Guimarães, 1983). In a few vessels, β_1-adrenoceptors appear to predominate, e.g., coronary arteries (O'Donnell and Wanstall, 1985; Begonha et al., 1995) and cerebral arteries (Edvinsson and Owman, 1974). In contrast to the heart where a maximal increase in force of contraction is obtained by stimulation of β_1-adrenoceptors only and activation of β_2-adrenoceptors causes no more than a submaximal effect (Kaumann et al., 1989; Motomura et al., 1990), in the vessels (at least in the veins), the maximum relaxation evoked by β_2-adrenoceptors is larger than that evoked by β_1-adrenoceptor stimulation (Guimarães and Paiva, 1981c) (Fig. 1). The maximal β-adrenoceptor-mediated relaxation varies from vascular bed to vascular bed (see *Section VI.C.*) and crucially depends on the level of the tone of the tissue (Guimarães, 1975; Begonha et al., 1995) (Table 3). The maximal β-adrenoceptor-mediated relaxation of the canine veins vary markedly from vein to vein (Table 4). After precontraction with methox-

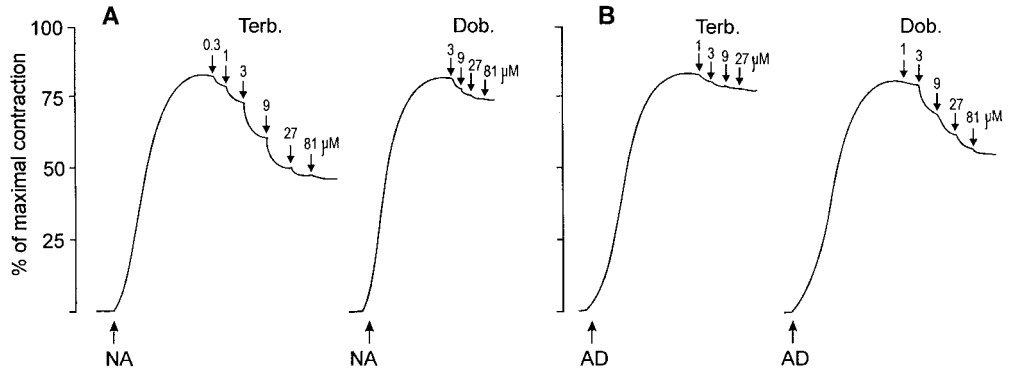

FIG. 1. Dog saphenous vein strips. After contractions of about the same magnitude (75% of the maximum) caused by noradrenaline (300 nM) (NA) or adrenaline (510 nM) (AD), concentration-response curves (relaxation) to terbutaline (Terb) or dobutamine (Dob) were determined. Because the concentration of noradrenaline used to cause the precontraction also occupies β_1-adrenoceptors, the relaxation to the selective β_1-adrenoceptor agonist dobutamine was much smaller than that to terbutaline. Conversely, because the concentration of adrenaline used to precontract the tissue also occupies β_2-adrenoceptors, relaxation to the selective β_2-adrenoceptor agonist terbutaline was much smaller than that to dobutamine. Furthermore, the maximum relaxation to terbutaline was greater than that to dobutamine (Guimarães and Paiva, 1981c).

TABLE 3
Maximal relaxant effect of isoprenaline in different canine arteries

Artery	At 95% of Maximal Contraction	At 80% of Maximal Contraction	At 65% of Maximal Contraction	At 50% of Maximal Contraction	At 35% of Maximal Contraction
Coronary	110	105	105	115	110
Pulmonary	0	16	17	31	50
Splenic	0	0	16	76	95
Mesenteric	0	3	70	82	90

Relaxant effects of isoprenaline were determined on rings precontracted with phenylephrine. The effect of isoprenaline was tested at five different levels of precontraction. In the coronary artery, the relaxation was larger than 100% since the tone reached a level lower than the baseline (adapted from Begonha et al., 1995).

TABLE 4
Maximal relaxant effect of isoprenaline in different canine veins

Vein	Maximal Relaxation in % of Previous Contraction
External jugular	86
Cephalic	91
Azygos	93
Pulmonary	88
Inferior vena cava	
High segment	91
Low segment	13
Portal	25
Mesenteric	17
Splenic	85
Renal	89
Femoral	82
Lateral saphenous	91

Vein strips were precontracted with methoxamine (5 μM) (Furuta et al., 1986).

amine, isoprenaline antagonized more than 80% of the precontraction in cephalic, external jugular, azygos, renal, femoral, saphenous, pulmonary, splenic, and the superior part of the inferior vena cava; whereas in the portal, mesenteric, and inferior segments of vena cava, the maximal relaxation antagonized less than 25% of the previous contraction (Furuta et al., 1986).

b. β_3-Adrenoceptors. The participation of a third β-adrenoceptor subtype in β-adrenoceptor-mediated vasodilatation was suggested by results obtained in several studies. Pindolol, a nonselective β-adrenoceptor antagonist with significant agonist activity, caused relaxation of canine isolated perfused mesenteric vessels (Clark and Bertholet, 1983) and rat aorta precontracted with KCl (Doggrell, 1990). In both instances, the vasorelaxant effect of pindolol was not significantly antagonized by propranolol, thus suggesting the presence of a β-adrenoceptor subtype different from the conventional β_1- and β_2-adrenoceptors, and the effect of isoprenaline was ascribed not only to activation of β_2- and β_1-adrenoceptors, but also to that of an additional adrenoceptor. Similar propranolol-resistant components to isoprenaline-induced relaxations have been observed in rat carotid artery (Oriowo, 1994; MacDonald et al., 1999), rat mesenteric artery (Sooch and Marshall, 1995), rat aorta (Gray and Marshall, 1992; Oriowo, 1995; Sooch and Marshall, 1997), rat pulmonary artery (Sooch and Marshall, 1996; Dumas et al., 1998), and canine pulmonary artery (Tamaoki et al., 1998; Tagaya et al., 1999). The involvement of β_3-adrenoceptors in isoprenaline-induced relaxation of vascular smooth muscle was demonstrated by the use of preferential β_3-adrenoceptor agonists and antagonists. In the rat carotid artery, the selective β_3-adrenoceptor agonist BRL-37344 and the selective β_2-adrenoceptor agonist, salbutamol, were not antagonized by propranolol (100 nM), and pretreatment of the artery segments with BRL-37344 did not desensitize the tissue to the relaxant effect of isoprenaline and salbutamol; it is noteworthy to point out that the pD_2 for salbutamol was 5.0, a value that is not consistent with the activation of β_2-adrenoceptors (Oriowo, 1994). In the same tissue, MacDonald et al. (1999) confirmed the presence of β_3-adrenoceptor by the relaxant effects of two selective β_3-adrenoceptor agonists, BRL-37344 and ZD-2079. A β_3-adrenoceptor-mediated vasorelaxation was

also observed in the canine pulmonary artery, an effect that was exerted through a cAMP-dependent pathway (Tagaya et al., 1999).

The presence of β_3-adrenoceptors has been reported also in veins. In the rat portal vein, activation of β_3-adrenoceptors stimulates L-type Ca^{2+} channels through a $G\alpha_s$-induced stimulation of the cyclic AMP/protein kinase A pathway and the subsequent phosphorylation of the channels (Viard et al., 2000).

c. Putative β_4-Adrenoceptors. Although there is now convincing evidence supporting the functional presence of β_3-adrenoceptors in vascular tissue, various observations cannot be fully explained by the existence of β_1-, β_2-, and β_3-adrenoceptors only. For example, the propranolol-resistant component of the effect of isoprenaline in rat aorta is not antagonized by either the selective β_3-adrenoceptor antagonist SR-59230 (Brawley et al., 2000a) or cyanopindolol, a β_3-adrenoceptor antagonist in the guinea pig ileum (Blue et al., 1989; Oriowo, 1994; MacDonald et al., 1999). Similarly, in rat aorta, the selective β_3-adrenoceptor agonist CGP-12177 was resistant to the blockade by SR-59230 (Brawley et al., 2000a). Also in agreement with these data, cyanopindolol did not inhibit the relaxant effect of isoprenaline in rat aorta, although it did antagonize the effect of isoprenaline at β_3-adrenoceptors in the distal colon and fundic strips (Oriowo, 1994). Furthermore, the order of potency for unconventional partial adrenoceptor agonists in rat aorta was contrary to that obtained at β_3-adrenoceptors in other tissues (Oriowo, 1994; Brawley et al., 2000a). According to these data, the propranolol-resistant component of the effect of isoprenaline was suggested to be mediated by the putative β_4-adrenoceptor. However, the selective β_3-adrenoceptor agonist BRL-37344, which does not activate this putative β_4-adrenoceptor (Malinowska and Schlicker, 1996; Galitzky et al., 1998), caused concentration-dependent relaxation in the rat aorta (Brawley et al., 2000a). Also in disagreement with the β_4-adrenoceptor hypothesis was that cyanopindolol showed higher potency than CGP-12177 in rat aorta, whereas it had been shown to have lower potency at the putative β_4-adrenoceptor (Malinowska and Schlicker, 1996). The identity of the receptor mediating the β_3-adrenoceptor–independent effects of CGP-12177 was clarified in a recent study in which it was shown that activation of adenylyl cyclase by CGP-12177 in β_3-adrenoceptor-knockout mice is mediated by β_1-adrenoceptors (Konkar et al., 2000a). The same authors showed that activation of β_1-adrenoceptors by CGP-12177 or LY-362884 (a second aryloxypropanolamine) is significantly more resistant to inhibition by β-adrenoceptor antagonists, compared with activation by catecholamines and suggests that catecholamines and aryloxypropanolamines interact with two distinct active conformational states of the β_1-adrenoceptor (Konkar et al., 2000b): one state that is responsive to catecholamines and is inhibited with high affinity by CGP-12177 and LY-362884, and a novel state that is activated by aryloxypropanolamines but is resistant to inhibition by classical β-adrenoceptor antagonists (Konkar et al., 2000b). Results leading to a similar conclusion were obtained in a rat model of cardiac failure: desensitization to isoprenaline and CGP-12177 after myocardial infarct and resensitization (after pertussis toxin treatment) occurs in parallel, suggesting that the β_1- and the putative β_4-adrenoceptor use the same pathway. Furthermore, antagonist affinity studies confirmed that drugs acting at β_1-adrenoceptors also interact with putative β_4-adrenoceptors with approximately 100 times lower affinity, suggesting that CGP-12177 causes its cardiac effects by interacting with a low affinity state of the β_1-adrenoceptor (Kompa and Summers, 2000). Recently Kaumann et al. (2001) showed that, in double β_1-/β_2-adrenoceptor knockout mice CGP-12177 did not at all affect force of contraction or heart rate, indicating an obligatory role of β_1-adrenoceptor for effects evoked by stimulation of the putative β_4-adrenoceptor. Accordingly, it is quite likely that there exist no fourth β-adrenoceptor, but the effects of CGP-12177 are due to an atypical interaction of this compound with the β_1-adrenoceptor.

2. In Vivo. β-Adrenoceptor-mediated vasodilation is thought to play an important physiological role in the regulation of vascular tone. Stimulation of peripheral β-adrenoceptors leads to relaxation of the vascular smooth muscle, thereby controlling the peripheral vascular resistance and consequently the distribution of blood to the different organs. During exercise, for example, activation of β-adrenoceptors contributes to the increased blood flow to skeletal muscle.

Although much has been learned about the role of the individual β-adrenoceptor subtype using classical pharmacological approaches in vitro, experiments in awake and unrestrained animals are of crucial importance to determine the real influence of factors coming into play, since reflex pathways can be obscured in anesthetized animals and are absent in the in vitro preparations. Experiments carried out in vivo in wild-type animals of several species had already indicated that the activation of β_2- and β_1-adrenoceptors led to relaxation of vascular smooth muscle of both arteries and veins (Taira et al., 1977; Vatner et al., 1985). However, it is now widely accepted that β-adrenoceptors other than the classical β_1- and β_2-adrenoceptors are also involved in the relaxation of the vasculature.

β_3-Adrenoceptors were recently shown to play a role in regulating peripheral vasodilatation, although this role was highly species-dependent (Shen et al., 1996) (Fig. 2).

In conscious dogs, the selective β_3-adrenoceptor agonist BRL-37344 caused a long-lasting vasodilation even in the presence of propranolol, whereas isoprenaline was totally ineffective when given in a dose that was equieffective prior to propranolol. This vasodilation occurred

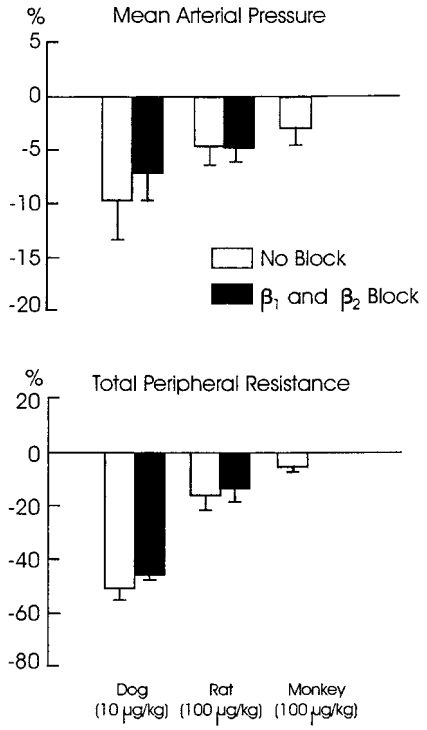

FIG. 2. The effects of CL-316243 on mean arterial pressure and total peripheral resistance are compared in the absence (open bars) and presence (closed bars) of β_1-/β_2-adrenoceptor blockade. Graded decrease in responsiveness to CL-316234 can be seen from the dog to the rat and to the monkey. β_1-/β_2-Adrenoceptor blockade did not affect significantly either parameter. Adapted from Shen et al. (1996).

primarily in the skin and fat, and persisted in the presence of a complete blockade of all known neural or hormonal pathways, indicating that probably it was due to activation of β_3-adrenoceptors (Tavernier et al., 1992; Berlan et al., 1994; Shen et al., 1994). Similarly, BRL-26830, another selective β_3-adrenoceptor agonist caused a marked increase in blood flow to brown adipose tissue in the anesthetized rat (Takahashi et al., 1992). Although these results support the view that the effect of BRL-26830 is β_3-adrenoceptor-mediated, they do not provide an unequivocal demonstration. The increase in blood flow may well be secondary to an augmented metabolic process (Shen and Claus, 1993), since BRL-37344 causes marked increases in the plasma levels of free fatty acids and insulin. In the rat, the selective β_3-adrenoceptor agonist CL-316243 induced a marked increase in both islet blood flow and plasma insulin level, and these increases were abolished by bupranolol, a β_1,β_2,β_3-adrenoceptor antagonist but not by nadolol—a β_1,β_2-adrenoceptor antagonist, indicating that β_3-adrenoceptors caused a vasodilation of microvessels in the islets of Langerhans (Atef et al., 1996).

Experiments carried out in rats, dogs, and monkeys showed a graded decrease in responsiveness to CL-316243 from the dog to the rat to the monkey, and suggest that β_3-adrenoceptor agonists do not evoke cardiovascular effects in primates (Shen et al., 1996). Furthermore, it was also shown that while BRL-37344 was selective enough to discriminate between β_3-adrenoceptors and the other two β-adrenoceptor subtypes (β_1 and β_2) in the dog (until now no data has been reported on the existence of putative β_4-adrenoceptors in the dog), only CL-316243 was able to provide a similar discrimination in the rat (Shen et al., 1994, 1996). This species-dependent selectivity of β_3-adrenoceptor agonists (Shen et al., 1996) is one possible explanation for the lack of effect of β_3-adrenoceptor agonists in primates. Another obvious explanation is that there are few or no β_3-adrenoceptors in primates. The presence of functional β_3-adrenoceptors in primates either in vessels or in the adipocytes is still a debatable question. Although some authors did not find evidence supporting this hypothesis (Zaagsma and Nahorski, 1990; Langin et al., 1991; Rosenbaum et al., 1993), many others found evidence supporting their existence in humans (Arner, 1995; Clément et al., 1995; Lönnqvist et al., 1995; Lowell and Flier, 1995; Walston et al., 1995) (see *Section IX.*).

Most recently, the compound L-750355 was identified as a potent and selective β_3-adrenoceptor agonist in cloned human and rhesus monkey β_3-adrenoceptors expressed in Chinese hamster ovary cells (Forrest et al., 2000). Furthermore, it was shown that L-750355 stimulates lypolisis in isolated human and rhesus adipocytes in vitro and causes tachycardia (propranolol-sensitive) and hyperglycerolemia (propranolol-resistant) in anesthetized rhesus monkeys (Forrest et al., 2000).

In summary, stimulation of β_3-adrenoceptors by the selective β_3-adrenoceptor agonists BRL-37344 and CL-316243 in mice, rats, and dogs causes long-lasting reductions in both blood pressure and total peripheral resistance, indicating that β_3-adrenoceptors are present in the vasculature (Tavernier et al., 1992; Shen et al., 1994, 1996; Rohrer et al., 1999). In contrast, in conscious monkeys and baboons, neither drug caused significant cardiovascular effects (Shen et al., 1996). Furthermore, CL-326243 appears to be a more selective β_3-adrenoceptor agonist than BRL-37344.

The recent progress in the development of knockout mice made it possible to selectively disrupt the gene for each of the β-adrenoceptor subtypes (β_1, β_2, and β_3) (Susulic et al., 1995; Rohrer et al., 1996; Revelli et al., 1997; Chruscinski et al., 1999); or for both β_1- and β_2-adrenoceptors (Rohrer et al., 1999). Animals lacking β_1- or β_2- or β_3-adrenoceptors had normal prenatal development, appeared grossly normal, were fertile, and showed normal resting cardiovascular parameters (Chruscinski et al., 1999). One should bear in mind, as already pointed out, that the lack of a given receptor from the conception may lead to compensatory changes.

In β_1-adrenoceptor-knockout mice, basal cardiovascular indices were unchanged and the capacity to respond to stresses like exercise was normal (Rohrer et al., 1996, 1998a), although the high variability in blood pressure in conscious unrestrained mice may have obscured a slight trend toward lower values in β_1-adrenoceptor-

knockout animals (Desai et al., 1997; Rohrer et al., 1998b). The hypotensive response to isoprenaline was not significantly different from that in wild-type mice; although the percent increase in heart rate after isoprenaline was significantly smaller in β_1-adrenoceptor-knockout than in wild-type animals (Rohrer et al., 1998b), and the nonselective β-adrenoceptor antagonist propranolol caused a modest pressor response in both wild-type and β_1-adrenoceptor-knockout mice (Rohrer et al., 1998b). In β_2-adrenoceptor-knockout mice, the resting cardiovascular parameters (heart rate and blood pressure) appeared completely unaltered. The major effects of β_2-adrenoceptor gene deletion were observed only during exercise. Apparently, β_2-adrenoceptor-knockout mice tolerate the workload better than wild-type controls. However, they were hypertensive during exercise, suggesting an imbalance between the vasoconstrictive and vasorelaxant effects of endogenous catecholamines (Chruscinski et al., 1999). In β_2-adrenoceptor-knockout mice, the hypotensive response to the nonselective β-adrenoceptor agonist isoprenaline is significantly attenuated, confirming that β_2-adrenoceptors play an important role in vascular relaxation and indicating that part of the hypotensive response to isoprenaline depends on the activation of other β-adrenoceptor subtypes.

In general terms, in double knockout mice lacking β_1- and β_2-adrenoceptors, changes in basal physiological cardiovascular functions are virtually nonexistent. However, functional deficits in vascular reactivity are revealed when β-adrenoceptors are stimulated by β-adrenoceptor agonists or exercise. In double β_1- and β_2-adrenoceptor-knockout mice, the hypotensive response to the selective β_3-adrenoceptor agonist CL-316243 is markedly enhanced (Fig. 3) (Rohrer et al., 1999). This enhancement may be ascribed to a vascular β_3-adrenoceptor up-regulation, since it was demonstrated that β_1-adrenoceptors are up-regulated in the adipose tissue of β_3-adrenoceptor-knockout mice (Susulic et al., 1995). Conversely, in the ileum, β_3-adrenoceptor-knockout mice, β_1-adrenoceptors functionally compensate for the lack of β_3-adrenoceptors (Hutchinson et al., 2001). Deficiencies in β-adrenoceptor signaling in double β_1- and β_2-adrenoceptor-knockout mice can be compensated for by increases in the density and/or signaling efficiency of other β-adrenoceptor subtypes (Rohrer et al., 1999).

The involvement of β_3-adrenoceptors in vascular relaxation is more and more widely accepted. According to Liggett et al. (1993), β_3-adrenoceptors, unlike β_1- and β_2-adrenoceptors, lack regulatory phosphorylation sites for G-protein receptor kinases, a characteristic that increases the resistance to agonist-evoked desensitization. Thus, under conditions of a persistent overstimulation of the sympathetic nervous system, β_1- and β_2-adrenoceptors are desensitized and β_3-adrenoceptors may represent a functional alternative (Rohrer et al., 1999). The fact that double knockout β_1- and β_2-adrenoceptor animals are supersensitive to β_3-adrenoceptor agonists is consistent with this tentative explanation.

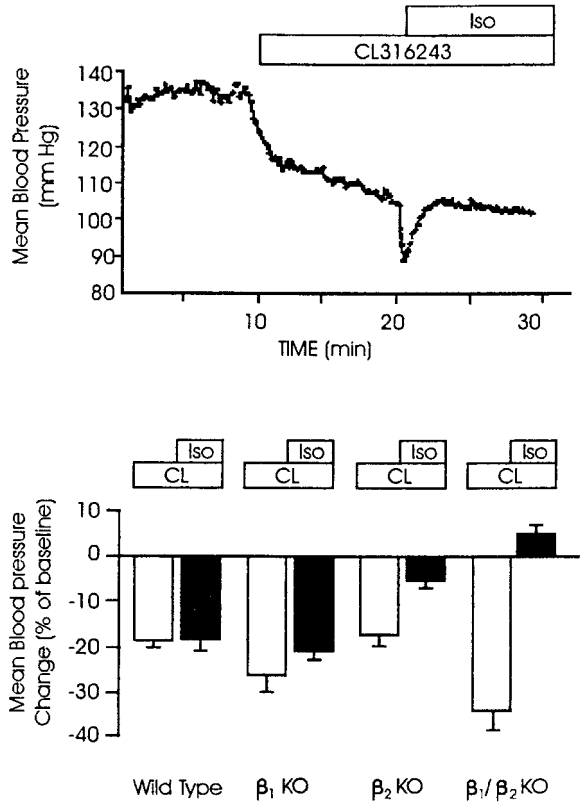

FIG. 3. Hemodynamic responsiveness to the β_3-adrenoceptor agonist CL-316243 (CL; open bars) and the nonselective β-adrenoceptor agonist isoprenaline (Iso; closed bars). Upper panel shows a blood pressure tracing of wild-type mouse given a single bolus injection of CL-316243 (100 μg/kg). Ten minutes later, isoprenaline (3 μg/kg i.a.) was administered. Bar graph in the lower panel shows the effects caused by the same drugs under identical conditions in β_1-, β_2-, and β_1-/β_2-knockout (KO) animals. Adapted from Rohrer et al. (1999).

Bearing in mind all these data, particularly those from knockout animals, it seems that β_1-adrenoceptors are those that predominantly regulate cardiac contractility and rate of heart, β_2-adrenoceptors are those that predominantly mediate the vasodilation evoked by sympathomimetic agonists, and β_3-adrenoceptors those that predominantly control lipolysis in adipose tissue. This is an oversimplified conclusion, which, however, may be valid for the majority of species.

IV. Prejunctional Adrenoceptors

A. α_2-Adrenoceptors

At the prejunctional level, α_2-adrenoceptors have been found in vitro in every vascular tissue (arteries and veins) until now studied where they mediate a negative modulation of the release of noradrenaline (Starke, 1987; Langer, 1997). Experiments in isolated organs support the view that the $\alpha_{2A/D}$-subtype is the principal prejunctional α_2-adrenoceptor; some studies indicate that, in certain tissues, including the heart and some vessels, another α_2-subtype might also be involved in the

regulation of the release of noradrenaline (Oriowo et al., 1991; Limberger et al., 1992; Guimarães et al., 1997; Trendelenburg et al., 1997; Docherty, 1998; Ho et al., 1998). Results obtained in knockout mice clearly confirmed this hypothesis. In the vas deferens, for example, it was observed that maximal inhibition by α_2-adrenoceptor agonists of the electrically evoked contractions was reduced to 50% in mice lacking $\alpha_{2A/D}$, whereas the effect of these agonists in α_{2C}-adrenoceptor-deficient mice was unchanged. Furthermore, in mice lacking both the $\alpha_{2A/D}$- and the α_{2C}-adrenoceptors, the prejunctional effect of α_2-adrenoceptor agonists was abolished, indicating that the residual 50% response to α_2-adrenoceptor agonists in $\alpha_{2A/D}$-knockout animals was due to prejunctional α_{2C}-adrenoceptors (Hein et al., 1998). Also in the heart atria and in the brain cortex, deletion of $\alpha_{2A/D}$-adrenoceptors reduced, but did not abolish, the inhibitory effect of the α_2-selective agonist UK-14304, indicating that a second α_2-autoreceptor operates in both sympathetic and central adrenergic neurons. However, the loss in E_{max} of α_2-adrenoceptor agonists was smaller in the heart than in the brain, supporting the view that, in the peripheral tissues, prejunctional non-$\alpha_{2A/D}$-adrenoceptors are functionally more important (Hein et al., 1999; Trendelenburg et al., 1999). Again, in $\alpha_{2A/DC}$-knockout mice, the concentration-dependent inhibition of noradrenaline release caused by α_2-adrenoceptor agonists in the atria was abolished, indicating that the α_{2C}-adrenoceptor is the second type involved in regulating noradrenaline release (Hein et al., 1999). Thus, the hypothesis that more than one α_2-adrenoceptor subtype might be present at the prejunctional level in the same tissue was confirmed in knockout animals.

According to Link et al. (1992), noradrenaline has a higher affinity for α_{2C}- than for $\alpha_{2A/D}$-adrenoceptors. In mouse atria, Hein et al. (1999) confirmed that noradrenaline was more potent on α_{2C}- than on $\alpha_{2A/D}$-adrenoceptors (EC_{50} = 16 nM, 20 nM, and 156 nM in wild-type, $\alpha_{2A/D}$-knockout, and α_{2C}-knockout animals, respectively) and showed that α_{2C}-adrenoceptors inhibit transmitter release at low levels of sympathetic tone and that $\alpha_{2A/D}$-adrenoceptors are required to control release at higher levels of sympathetic tone.

Unfortunately there are no studies in isolated vascular tissues from knockout animals. However, the calculation of the correlation between the pK_i values of several antagonists at some canine and human vessels and their pK_d values at prototypical α_{2A}, α_{2B}, α_{2C}, and α_{2D} radioligand binding sites (Altman et al., 1999) shows that, as in other peripheral tissues, the main modulatory role is played by $\alpha_{2A/D}$-adrenoceptors and suggests that more than one α_2-adrenoceptor subtype participate in the feedback inhibition of transmitter release (Table 5). This calculation still suggests that α_{2B} or α_{2C} or both may be involved in this modulatory influence. Additionally, it can be concluded that the existence of a second type of prejunctional receptor in vascular tissues of $\alpha_{2A/D}$-knockout mice does not result from a compensatory mechanism (Altman et al., 1999), since these results were obtained in wild-type animals.

There is no doubt that in vessels the control mechanism of noradrenaline release is highly active at physiological frequencies of electrical stimulation. Evidence supporting the possibility of a tonic physiological role of a negative feedback control of noradrenaline release appeared before the discovery of prejunctional α-adrenoceptors. The first report pointing in this direction was that by Malik and Muscholl (1969), who showed that noradrenaline, in doses that did not alter basal resistance, slightly reduced the response of the perfused mesenteric artery to sympathetic nerve stimulation. This was confirmed by the observation, in the rabbit pulmonary artery, that the inhibitory influence of a fixed concentration of an exogenous agonist was not as effective at high as at low frequencies of stimulation (Starke and Endo, 1975). Indeed, at the higher frequencies, the negative influence of α-adrenoceptor agonists is less because the prejunctional α-adrenoceptors are already more intensely activated by endogenous noradrenaline (Vizi et al., 1973; McCulloch et al., 1975).

Numerous pharmacological findings obtained by many authors in several species, including humans, support the view that a negative feedback mechanism mediated by prejunctional α_2-adrenoceptor operates under physiological conditions of noradrenergic neurotransmission (for reviews, see Starke, 1977, 1987; Langer, 1981, 1997). However, this hypothesis was contested by Kalsner (1982) and Kalsner and Westfall (1990). The latest observations in knockout animals clearly show that prejunctional α_2-adrenoceptors are really autoreceptors that accomplish a physiological function.

TABLE 5

	$\alpha_{2A/D}$	α_{2B}	α_{2C}
Dog saphenous vein	r = 0.90*	r = 0.78	r = 0.85*
	Slope = 0.95*	Slope = 0.41	Slope = 0.89*
Human gastric artery	r = 0.94*	r = 0.66	r = 0.71
	Slope = 1.28*	Slope = 0.61*	Slope = 0.88
Human ileocolic artery	r = 0.96*	r = 0.68	r = 0.57
	Slope = 1.39*	Slope = 0.77	Slope = 0.88

Correlation between $pEC_{30\%}$ values of antagonists at prejunctional α_2-autoreceptors of the canine saphenous vein (Paiva et al., 1997), and human gastric and iliocolic arteries (Guimarães et al., 1998b) and pK_i values at binding sites (HT29 cells, in A; neonatal rat lung, in B; OK cells, in C; and rat submaxillary gland, in D).
* $p < 0.05$.

B. β-Adrenoceptors

The release of noradrenaline from sympathetic varicosities is modulated by a large number of prejunctional auto- and heteroreceptors. Facilitatory β-adrenoceptors have been shown in various sympathetically innervated tissues of many species both in vitro (Adler-Graschinsky and Langer, 1975; Majewski, 1983; Misu and Kubo, 1986; Encabo et al., 1996) and in vivo (Boudreau et al., 1993; Tarizzo et al., 1994; Vila and Badia, 1995). Those receptors, which have been demonstrated to be of the β_2-subtype (Dahlöf et al., 1978; Guimarães et al., 1978; Göthert and Hentrich, 1985; Coopes et al., 1993), may be activated by circulating adrenaline (Stjärne and Brundin, 1975) and/or by adrenaline taken up by and released from sympathetic varicosities as a cotransmitter (Majewski et al., 1981a,b; Misu et al., 1989; Tarizzo and Dahlöf, 1989). Very recently, the existence of prejunctional β_2-adrenoceptors located on nerve terminals that release nitric oxide (NO: NOergic nerve terminals) of the porcine basilar arteries was proposed by Lee et al. (2000). According to these authors, noradrenaline released from sympathetic nerves upon application of nicotine, acts on prejunctional β_2-adrenoceptors of NOergic nerve terminals to release NO, resulting in vasodilatation. In humans, there is also evidence for facilitatory β-adrenoceptors on noradrenergic nerve endings, both in vitro (Stjärne and Brundin, 1975; Stevens et al., 1982) and in vivo (Brown and Macquin, 1981; Vincent et al., 1982; Blankestijn et al., 1988). However, whereas prejunctional α_2-adrenoceptors (of the $\alpha_{2A/D}$- and α_{2C}-subtype) play an important physiological role in the feedback inhibition of neurotransmitter release (Starke, 1987; MacMillan et al., 1996; Altman et al., 1999; Trendelenburg et al., 1999), the physiological role of the facilitatory prejunctional β-adrenoceptors remains controversial (Abrahamsen and Nedergaard, 1989, 1990, 1991; Floras, 1992; Apparsundaram and Eikenburg, 1995; Coopes et al., 1995). A hypothetical pathological implication of the facilitatory prejunctional β_2-adrenoceptors originated when Guimarães et al. (1978) first showed that, in canine saphenous vein, propranolol was able to reduce the electrically evoked tritium overflow after preloading of the venous tissue with ^{3}H-adrenaline, but not when it had been preloaded with ^{3}H-noradrenaline. Confirmatory results were obtained in different in vitro vascular peparations (Majewski et al., 1981a; Misu et al., 1989; Rump et al., 1992) and also in vivo studies: in the anesthetized rabbit (Majewski et al., 1982); in the anesthetized dog (Boudreau et al., 1993); in the pithed rat (Vila and Badia, 1995); and in the demedulated rat (Coopes et al., 1993, 1995). On the basis of these results, the hypothesis was put forward that high levels of adrenaline within the synaptic cleft could either directly stimulate prejunctional β_2-adrenoceptors or be taken up by sympathetic nerves, and then be coreleased and activate prejunctional β_2-adrenoceptors. In both cases, stimulation of this positive feedback loop would lead to an increased release of noradrenaline, which might represent an early step in the development of hypertension (Majewski et al., 1981b; Misu et al., 1990; for a review, see Floras, 1992). Depletion of plasma adrenaline by surgical adrenal demedullation attenuates the development of hypertension in 4-week-old SHRs (Borkowski and Quinn, 1985), and this effect is antagonized by depot implants of adrenaline. Moreover, the prohypertensive effect of adrenaline in demedullated SHRs was abolished by concomitant treatment with β_2-adrenoceptor antagonists (Borkowski and Quinn, 1985). The development of hypertension was attenuated only in SHRs demedullated at 6 weeks of age or younger, indicating that a period of critical sensitivity of prejunctional β_2-adrenoceptors to the facilitatory effect of catecholamines may exist (Borkowski, 1991). In humans, episodes of sympathoadrenal activation repeatedly causing increases in plasma adrenaline concentration might, by direct (as a hormone), indirect (as a cotransmitter), or both ways, initiate or facilitate the development of primary hypertension (Brown and Macquin, 1981; Blankestijn et al., 1988). Although attractive and based on a large amount of experimental work, this theory has not received conclusive support so far. Floras et al. (1988) reported that 30 min after a local infusion of adrenaline into the forearm of volunteers, there was a facilitation of neurogenic vasoconstriction that was due to a delayed facilitation of noradrenaline release caused by the previous infusion of adrenaline. Stein et al. (1997), under identical conditions, did not observe any delayed facilitatory effect on noradrenaline spillover in the forearm of normotensive or borderline hypertensive subjects. However, these authors observed an increase in systemic noradrenaline spillover 30 min after the infusion of adrenaline, suggesting that there is a delayed facilitatory response to adrenaline in specific organs, the identification of which would be of importance in elucidating the role of this mechanism in the pathogenesis of hypertension. In a comparative study involving patients with longstanding essential hypertension and normotensive control subjects, Chang et al. (1994) evaluated the kinetics of noradrenaline by measuring in the forearm the appearance rate of noradrenaline in plasma and the spillover of noradrenaline into plasma before and after the infusion of adrenaline and found no differences between the two groups. Similarly, in 40 healthy volunteers, loading of sympathetic nerve terminals of the human forearm with adrenaline did not augment subsequent neurogenic vasoconstriction or noradrenaline release in response to sympathetic stimulation (Goldstein et al., 1999). Using a different methodological approach, Thompson et al. (1998) also obtained negative results: isometric handgrip contraction evoked similar responses in total and cardiac noradrenaline spillovers, and in muscle sympathetic activity before and after an infusion of adrenaline. The influence of muscle contrac-

tion and blood flow on noradrenaline and adrenaline spillover was also studied in the in situ canine gracilis muscle, and spillover of both amines was observed (Lavoie et al., 2000). Since adrenaline is not synthesized locally, adrenaline spillover means that it was taken up from the circulation, stored in the vesicles, and then re-released with noradrenaline (Lavoie et al., 2000).

Although convincing evidence for the positive feedback loop hypothesis is still lacking, it may be inadvisable to regard it as disproven. As suggested by Folkow (1982), the development of hypertension by this mechanism may be restricted to a specific subset of individuals genetically predisposed to high blood pressure. As pointed out by Floras (1992), there are no prospective evaluations of the predictive value of augmented plasma adrenaline concentrations in childhood and adolescence as an indicator for later hypertension development; it is possible that, as shown for SHRs (Borkowski 1991), a period of critical sensitivity of prejunctional β_2-adrenoceptors to catecholamines exists during which the pathological development starts. If this were true for humans and if it were known into which age this critical period fell, an adequate treatment might avoid the progress toward an established hypertension.

V. Endothelial Adrenoceptors

A. α_2-Adrenoceptors

It is now widely accepted that vascular endothelium plays an important role in the function of the cardiovascular system (Moncada et al., 1991a; Vaz-da-Silva et al., 1996; Busse et al., 1998). Functional evidence suggesting that α_2-adrenoceptors play a role in the physiology of the vasculature was first reported by Cocks and Angus (1983), who showed that, in isolated coronary, renal, and mesenteric arteries, noradrenaline and clonidine caused relaxation that was inhibited by selective α_2-adrenoceptor antagonists and was eliminated by removal of the endothelium. Similar results were obtained in several isolated arteries (Egleme et al., 1984; Angus et al., 1986) and veins (Miller and Vanhoutte, 1985). Soon after the discovery of the EDRF (Furchgott and Zawadzki, 1980)—later on identified with NO—(Ignarro et al., 1987; Palmer et al., 1987), it was demonstrated that activation of α_2-adrenoceptors on endothelial cells stimulates the release of NO, an action that would tend to attenuate vasoconstriction produced by activation of postjunctional vascular α_1-adrenoceptors (Angus et al., 1986; Vanhoutte and Miller, 1989; Richard et al., 1990). Furthermore, it was suggested that endothelial α_2-adrenoceptors mediate release of EDRF in coronary microvessels (Angus et al., 1986). Thus, it appears that α_2-adrenoceptor agonists do indeed have the capability of modulating vascular responsiveness via stimulation of the release of NO in both large arteries and microcirculation. Furthermore, it was reported that noradrenaline-induced release of nitric oxide is enhanced in mineralocorticoid hypertension (Bockman et al., 1992), indicating that endothelial α_2-adrenoceptors may play an important role in the regulation of vascular tone not only in physiological, but also in pathological conditions.

Which α_2-adrenoceptor subtype is responsible for this modulatory influence? The first study aiming at characterizing the α_2-adrenoceptor subtypes present on vascular endothelium was carried out in pig coronary arteries and showed that the endothelium of this vessel possesses both $\alpha_{2A/D}$- and α_{2C}-adrenoceptors, the latter predominating (77% of α_{2C} versus 23% of $\alpha_{2A/D}$). However, despite the prominent presence of α_{2C}-adrenoceptors, the $\alpha_{2A/D}$-adrenoceptor subtype is the one mediating endothelium-dependent relaxation (Bockman et al., 1993). Interestingly, it was shown that in the rat mesenteric artery the α_2-adrenoceptor that is coupled to endothelium-dependent NO-mediated relaxation belongs to the $\alpha_{2A/D}$-subtype appearing in its α_{2D}-version (Bockman et al., 1996). Also in the endothelium of different species, the $\alpha_{2A/D}$-adrenoceptors serve the same function (Bylund et al., 1995a). Contrary to what was expected, cAMP is not involved in the signal transduction pathway for $\alpha_{2A/D}$-adrenoceptor-mediated NO formation (Bockman et al., 1996).

B. β-Adrenoceptors

It is now widely accepted that β-adrenoceptors exist on endothelial cells and contribute to the regulation of vasomotor tone. The role these receptors play, the mechanisms by which this role is played and the β-adrenoceptor subtypes that are involved are still debatable questions. Some of the first studies on the existence of endothelial β-adrenoceptors and some others carried out later on did not find evidence supporting their existence. Removal of the endothelium or inhibitors of NO synthase were found to have no influence on isoprenaline-evoked relaxations in rat aorta (Konishi and Su, 1983), canine coronary arteries (Cohen et al., 1983, 1984; White et al., 1986), rat carotid artery (Oriowo, 1994), or human internal mammary artery (Molenaar et al., 1988). In contrast, many other authors reported that removal of endothelium reduces the relaxations caused by β-adrenoceptor agonists in several isolated vessels from different species, including humans (Grace et al., 1988; Kamata et al., 1989; Dainty et al., 1990; Gray and Marshall, 1992; Delpy et al., 1996; Toyoshima et al., 1998; Ferro et al., 1999; Trochu et al., 1999; Brawley et al., 2000a,b; Vanhoutte, 2000). Surprisingly, in the same preparation (the thoracic aorta), different authors found opposite results. This discrepancy may be ascribed to one or more of the following factors: 1) the agent used to precontract the vessel was either noradrenaline (which also activates β-adrenoceptors) or phenylephrine (which lacks affinity for β-adrenoceptors) (Guimarães, 1975); 2) the level of the precontraction at which the β-adrenoceptor-mediated relaxation might not be the same and the magnitude of the relaxant effect is critically dependent

on the extent of the pre-existing tone (Guimarães, 1975); and 3) the β_3-adrenoceptor agonist used may be more or less active on β_1- and β_2-adrenoceptors.

According to Eckly et al. (1994), the reduction in response to isoprenaline after pretreatment with L-NAME or endothelial removal can be explained by the fact that the precontraction of the vessel is greater than in control tissues due to the disappearance of NO production under resting conditions. This enhancement of the precontraction would counteract the relaxation (Guimarães, 1975). However, it has been shown that endothelial removal does not consistently increase the preconstriction caused by noradrenaline or phenylephrine (Delpy et al., 1996; Brawley et al., 2000b). Many other kinds of evidence also support the view that β-adrenoceptors are present in endothelial cells and mediate relaxing responses in which NO is involved. First of all, the presence of β-adrenoceptors was confirmed by radioligand binding studies in cultured bovine aortic endothelial cells (Steinberg et al., 1984), by autoradiography in human cardiac endocardium (Buxton et al., 1987), in the endothelium of internal mammary artery and saphenous vein (Molenaar et al., 1988), and by biochemical data obtained in cultured human umbilical vein endothelial cells (Ferro et al., 1999). Furthermore, in vivo studies in cat hindlimb (Gardiner et al., 1991), canine coronary artery (Parent et al., 1993), and newborn pig pial arteries (Rebich et al., 1995) support a role of vascular endothelium in β-adrenoceptor-mediated relaxation. In humans, it was also found that forearm blood flow increases by infusion of either isoprenaline or salbutamol into the brachial artery, and coinfusion of the nitric oxide synthase inhibitor L-NMMA blocks this response to either drug (Dawes et al., 1997). Additionally, it had been shown that relaxant responses of the rat aorta to isoprenaline are inhibited by methylene blue and hemoglobin (Grace et al., 1988), indicating that the endothelium-dependent NO/cGMP system may be activated by stimulation of β-adrenoceptors (Grace et al., 1988; Gray and Marshall, 1992; Iranami et al., 1996).

The second point concerns the role played by endothelial β-adrenoceptors and the mechanism through which they induce their effects. Pretreatment with L-NAME and endothelium removal exert a similar inhibitory influence on isoprenaline-evoked relaxation, and the combination of the two procedures has no additional effect, compared with either treatment alone (Gray and Marshall, 1992; Ferro et al., 1999). On the basis of this evidence, these authors concluded that β-adrenoceptor-mediated vasorelaxation is totally endothelium-dependent: isoprenaline-evoked relaxation is due to the elevation of cyclic AMP caused by β_2-adrenoceptor stimulation, and this elevation activates the L-arginine/NO system and gives rise to vasorelaxation (via cGMP formation) (Gray and Marshall, 1992; Ferro et al., 1999). However, other authors found little or no effect of endothelium removal on isoprenaline-evoked relaxations (Konishi and Su, 1983; Moncada et al., 1991b; Eckly et al., 1994; Satake et al., 1996). The hypothesis that incomplete removal of endothelium might account for some remaining relaxation to isoprenaline (Gray and Marshall, 1992) can be discarded at least in some cases in which part of the relaxation to isoprenaline remained, although removal of endothelium had abolished acetylcholine-evoked relaxation (Brawley et al., 2000b). In these cases, treatment with L-NAME of the endothelium-free preparations caused no further effect on the isoprenaline-evoked relaxations (Brawley et al., 2000b). Thus, it appears that the relaxant effect of isoprenaline involves two components: one endothelium-dependent and another endothelium-independent (Brawley et al., 2000b). The endothelium-dependent component is triggered by β-adrenoceptor activation and leads to the promotion of NO production/release (Gray and Marshall, 1992; Ferro et al., 1999; Brawley et al., 2000b), or to some kind of enhancement of smooth muscle β-adrenoceptor-mediated relaxation by basal release of NO (Grace et al., 1988; Delpy et al., 1996).

A third point refers to the endothelial β-adrenoceptor subtype(s) involved in isoprenaline-evoked relaxation of the vascular smooth muscle. It is now recognized that β-adrenoceptors located in the endothelium play an important role in the relaxant response to isoprenaline, since the nonselective β_1-and β_2-adrenoceptor antagonist propranolol antagonized this relaxant effect (Oriowo, 1995; Sooch and Marshall, 1996; Brawley et al., 2000a,b). However, recent studies carried out in humans—either in umbilical veins in vitro (Ferro et al., 1999) or in the forearm in vivo (Dawes et al., 1997)—showed that vasorelaxation to isoprenaline was abolished by the selective β_2-adrenoceptor antagonist ICI-118551 and remained unchanged in the presence of the β_1-adrenoceptor antagonist CGP-20712, indicating that as in the vascular smooth muscle cells (Lands et al., 1967a,b), the endothelial β-adrenoceptors are totally or at least predominantly of the β_2-subtype (Dawes et al., 1997; Ferro et al., 1999). Furthermore, it was observed that, after L-NAME treatment or removal of endothelium, relaxant responses to isoprenaline were still unaffected by propranolol, suggesting that they were mediated by β_3- and/or the low-affinity state of β_1-adrenoceptors, formerly proposed as putative β_4-adrenoceptors (Brawley et al., 1998).

Additionally, it was shown that relaxation of rat thoracic aorta was also caused by selective β_3-adrenoceptor agonists like CGP-12177 (Mohell and Dicker, 1989), cyanopindolol (Engel et al., 1981), ZD-2079 (Grant et al., 1994), ZM-215001 (Tesfamariam and Allen, 1994), and SR-58611 (Trochu et al., 1999), further supporting the presence of β_3-adrenoceptors (Brawley et al., 2000a,b).

Endothelial removal or pretreatment with L-NAME significantly reduced the relaxation caused by isoprenaline or SR-58611 (Trochu et al., 1999), but had less effect on the relaxation caused by another selective β_3-

adrenoceptor agonist BRL-37344 (MacDonald et al., 1999) or CGP-12177 (Brawley et al., 2000a,b). Furthermore, sodium nitroprusside enhanced isoprenaline effects as previously reported (Maurice and Haslam, 1990), but had little or no effect on the response to CGP-12177 (Brawley et al., 2000a,b) or on the relaxation to isoprenaline in the presence of propranolol. After L-NAME had reduced responses to both isoprenaline and CGP-12177, sodium nitroprusside restored them, but the contribution of NO to the atypical β-adrenoceptor-mediated response was less than to β_1- and β_2-adrenoceptor-mediated relaxation (Brawley et al., 2000a,b). Both of these procedures indicate that exogenously applied NO interacts with the β_1- and β_2-adrenoceptor signaling pathway to a greater extent than with the non-β_1-/β_2-adrenoceptor pathway. However, the non-β_1-/β_2-adrenoceptor-mediated component of the response to isoprenaline appears to be partially endothelium-dependent, since L-NAME or endothelium removal attenuated isoprenaline relaxation in the presence of propranolol (Shafiei and Mahmoudian, 1999; Trochu et al., 1999; Brawley et al., 2000b).

All of these findings show that the endothelium/NO pathway modulates β_1- and β_2-adrenoceptor-mediated responses in rat aorta to a greater extent than non-β_1-/β_2-adrenoceptor-mediated responses (Brawley et al., 1998; MacDonald et al., 1999) and indicate that non-β_1-/β_2-adrenoceptors are present in the endothelium of some mammalian arteries.

Which non-β_1-/β_2-adrenoceptor? This is still a difficult question to answer. First of all, in the rat pulmonary vessels, several β_3-adrenoceptor agonists (SR-58611, SR-59119, and SR-59104) caused relaxant effects. However, only the effect of SR-59104 was antagonized by the selective β_3-adrenoceptor antagonist SR-59230 (Dumas et al., 1998). In the rat thoracic aorta, β_3-adrenoceptors are mainly located on endothelial cells, and act in conjunction with β_1- and β_2-adrenoceptors to mediate relaxation through activation of an NO synthase pathway and subsequent increase in cyclic GMP levels (Trochu et al., 1999).

VI. Distribution of Vascular Adrenoceptors

A. Localization in Relation to Sympathetic Nerve Terminals

In vascular tissue, α- and β-adrenoceptors are not situated close to each other (Guimarães et al., 1981a,b; Guimarães, 1982), such that when there is a change in the concentration of circulating adrenaline coming from the blood or of noradrenaline coming from the sympathetic nerve terminals, it does not affect α- and β-adrenoceptors equally; there are two different biophases for sympathomimetic agonists: one for α-adrenoceptors around the nerve terminals, where the concentration of the agonist available for α-effect is mainly governed by uptake into these terminals; and one for β-adrenoceptors in the neighborhood of catechol-*O*-methyl transferase whose activity is the main factor determining the concentration of the agonist available for the β-effect (for a review, see Guimarães, 1982; Guimarães et al., 1982). This will contribute to the fact that the different vessels have different sensitivities to sympathomimetic amines, such that some of them may be under the control of circulating catecholamines, whereas others are not.

In 1980, Yamaguchi and Kopin showed that, in the pithed rat, the pressor response to sympathetic nerve stimulation is the result of activation of α_1-adrenoceptors, whereas the pressor effects of the exogenous catecholamines are medited by α_2-adrenoceptors. This conclusion was consistent with the suggestion that postjunctional vascular α_2-adrenoceptors might be located at extrajunctional sites (Langer et al., 1981). This differential location of α_1- and α_2-adrenoceptors in relation to the nerve terminals was confirmed in conscious rabbits, where it was shown that pretreatment with 6-hydroxydopamine augmented the pressor response to α_1-adrenoceptor agonists without changing the responses to α_2-adrenoceptor agonists (Hamilton and Reid, 1981). All these results are consistent with the hypothesis that α_1-adrenoceptors are located in the vicinity of sympathetic nerve terminals, strategically situated to be activated by noradrenaline coming out from the nerves, whereas α_2-adrenoceptors are situated extrajunctionally and may be activated preferentially by circulating catecholamines, particularly adrenaline. However, in the isolated rabbit portal vein, the selective α_1-adrenoceptor antagonist prazosin failed to antagonize the contractions evoked by electrical stimulation of the vessel, whereas the selective α_2-adrenoceptor antagonist rauwolscine did antagonize these contractions (Docherty and Starke, 1981). In agreement with this report, it was shown that, in the canine saphenous vein, the inhibition of neuronal uptake by cocaine enhanced the contractile response to noradrenaline more in the presence of prazosin (α_2-adrenoceptor-mediated response) than in that of yohimbine (α_1-adrenoceptor-mediated response) (10- versus 6-fold; Guimarães et al., 1983). Furthermore, yohimbine was more potent than prazosin in antagonizing the effect of noradrenaline released from nerve terminals either by electrical stimulation or tyramine (Guimarães et al., 1983; Cooke et al., 1984; Flavahan et al., 1984; Pereira et al., 1991). More recently, it was reported that, in the canine saphenous vein in vitro, the contractile response evoked by electrical stimulation is mediated by three receptors: α_1- and α_2-adrenoceptors and P2X-receptors; the α_1-adrenoceptor and the P2X-receptor-mediated contractions develop immediately after starting the stimulation and reach the maximum very quickly, whereas the α_2-adrenoceptor-mediated contraction develops slowly, although reaching a maximum of similar magnitude. The purinergic component was smaller than the other two. Cocaine, which did not change the purinergic response, enhanced

both adrenoceptor-mediated components but enhanced more markedly the α_2-adrenoceptor-mediated- than the α_1-adrenoceptor-mediated component (Hiraoka et al., 2000).

In surgically denervated canine saphenous veins, Flavahan et al. (1987b) showed that α_2-adrenoceptor-mediated responses to noradrenaline are augmented, whereas α_1-adrenoceptor-mediated responses are not. All these results indicate that, in contrast to the arteries, in these veins α_2-adrenoceptors are situated closer to the sympathetic nerve terminals than α_1-adrenoceptors.

A similar differential location was also encountered among β-adrenoceptors. β_1-Adrenoceptors that are very responsive to noradrenaline are "innervated" and mediate responses to sympathetic nerve activity, whereas β_2-adrenoceptors that are insensitive to noradrenaline are functionally "noninnervated" and function as hormone receptors for adrenaline from the adrenal medulla (Russel and Moran, 1980; Bryan et al., 1981). In electrically driven rabbit papillary muscles, it was shown that both α_1- and β-adrenoceptors are located near or within the synaptic clefts of the sympathetic nerve endings (Dybvik et al., 1999). However, in the severely failing human heart, whereas α_1-adrenoceptors are apparently located close to the nerve endings, the down-regulated β-adrenoceptors are situated outside the range of the neuronal influence, a fact that may have functional implications (Skomedal et al., 1998). As far as the veins are concerned, it was shown that, in normal strips of canine saphenous vein precontracted by prostaglandin $F_{2\alpha}$ in the presence of phentolamine, there was no relaxant response to either electrical stimulation or tyramine. However, in strips preloaded with adrenaline and precontracted by prostaglandin $F_{2\alpha}$ in the presence of phentolamine, electrical stimulation or tyramine caused frequency- or dose-dependent relaxations up to a maximum of 53.6% and 49%, respectively, of the steady-state precontraction (Guimarães and Paiva, 1981b). These results seem to indicate that, in this venous tissue, β_2-adrenoceptors are located relatively close to the nerve terminals, whereas β_1-adrenoceptors are not innervated (or are not abundant enough), since adrenaline coming from the nerves can efficiently cause relaxation whereas noradrenaline does not.

Apparently, the adrenoceptors under sympathetic control vary from vascular bed to vascular bed, and the vascular tone results from the simultaneous activation of receptors that are differentially influenced in the different vascular areas by the transmitters coming out from the sympathetic nerve endings. The receptors, the activation of which more importantly contribute to the basal vascular tone, are those that are "innervated": the α_1-adrenoceptors in the arterial vessels, the α_1- and β_1-adrenoceptors in the heart, and α_2- and β_2-adrenoceptors in the veins.

B. Distribution Upstream and Downstream

In this section, we deal with those smooth muscle receptors that are functionally involved in the responses to activation of adrenoceptors.

Noradrenaline contracts the vascular smooth muscle of most major arteries by activating postjunctional α_1-adrenoceptors (see *Section III.A.1.*). However, in several blood vessels, α_2-adrenoceptors also contribute to the vasoconstriction caused by noradrenaline, particularly in cutaneous arteries and veins (Polónia et al., 1985; Flavahan and Vanhoutte, 1986a). Most interesting is that the contribution of α_1- and α_2-adrenoceptors to the vasoconstriction caused by noradrenaline changes along the length of a single vessel (Bevan et al., 1980). In the arteries of the limbs, the participation of α_2-adrenoceptors for the α-adrenoceptor-mediated vasoconstriction caused by noradrenaline increases from the proximal to the distal parts of these vessels. A comparison between human proximal (dorsalis pedis and arcuate arteries of the foot and superficial palmar arch of the hand) and distal arteries (digital arteries of the foot and hand) showed an increased prominence of α_2-adrenoceptors on distal, compared with proximal, arteries (Flavahan et al., 1987a). The reduction in α_2-adrenoceptor responsiveness from distal to proximal arteries continues in more proximal blood vessels such that α_2-adrenoceptor-mediated responses are not present in larger arteries of the limbs (Thom et al., 1985). In the mouse tail artery, the contractile response resulting from α_2-adrenoceptor activation is also greater at the distal than at the proximal level, whereas the opposite pattern was observed for α_1-adrenoceptors (Chotani et al., 2000).

This pattern of increasing α_2-adrenoceptor responsiveness from proximal to distal arteries is apparently not observed in the cerebral circulation. Bevan et al. (1987) showed that, in the cerebral arteries, the α-adrenoceptor responsiveness becomes progressively less important with successive branching of arteries. Small branches seem to have no functional α-adrenoceptors (Bevan et al., 1987). This may explain the observation that, in mice lacking the $\alpha_{2A/D}$-adrenoceptors, the pressor response to intra-arterial injection of α_2-adrenoceptor agonists (UK-14304 or dexmedetomidine) was blunted when the injection was given into the femoral artery, but not when the injection was given into the carotid artery (MacMillan et al., 1996).

In contrast to what was observed in arteries, it was shown that on isolated strips of canine saphenous and cephalic veins the maximum contractile effect of α_2-adrenoceptor activation was markedly smaller at the distal than at the proximal level; however, there was no change in the potencies of the selective α_2-adrenoceptor agonists UK-14304 or BHT 920 along the length of the vessels (Guimarães and Nunes, 1990), indicating that the density of α_2-adrenoceptors is higher at the proximal than at the distal level. This was confirmed in perfusion

experiments with distal segments of canine saphenous vein where it was observed that they responded very poorly or did not respond at all to α_2-adrenoceptor agonists applied to either the intima or the adventitia (Nunes et al., 1991). Since α_2-adrenoceptors are very sensitive to changes in temperature and are abundant in cutaneous blood vessels (of the limbs in humans and dogs and of the tail in the rat) (see *Section VIII.*), they appear to be important for thermoregulation, by constricting on cooling and dilating when exposed to a warmer environment. It is not surprising that the distribution of α_2-adrenoceptors in cerebral and cutaneous arteries is different, since the functional role of arteries in these two beds is not comparable.

Large coronary vessels possess both α- and β-adrenoceptors, whereas small vessels of the coronary circulation possess only β-adrenoceptors (Bohr, 1967).

β-Adrenoceptors are also not uniformly distributed along the length of a single vessel. The thoracic aorta of the rabbit shows considerable β-adrenoceptor-mediated activity, whereas in the abdominal aorta β-adrenoceptor-mediated activity is practically nonexistent (Bevan et al., 1980). In the dog saphenous vein, the maximal relaxation to isoprenaline was much larger in the distal than in the proximal vein, whereas the effectiveness of forskolin did not vary, irrespective of the tone and the segment of vein used (Guimarães et al., 1993). Furthermore, the effectiveness of dobutamine increased from the proximal to the distal part, whereas that of the selective β_2-adrenoceptor agonist terbutaline decreased. This indicates that the effectiveness of β-adrenoceptor activation and the contribution of β_1-adrenoceptors to the relaxation increase from the proximal to the distal part of the canine saphenous vein (Guimarães et al., 1993).

It is not easy to propose an explanation for this differential distribution of β-adrenoceptors, since the functional consequences of their activation by any of the endogenous ligands (adrenaline and noradrenaline) is always masked by the predominant effect resulting from simultaneous activation of α-adrenoceptors.

C. Distribution in Particular Vascular Beds

There is also a regional variation in the distribution of vascular adrenoceptors. For many years, it has been accepted, at least in humans, that splanchnic and skeletal muscle vascular beds dilate to adrenaline because β-adrenoceptors predominate in their vessels, whereas adrenaline consistently reduces renal and skin blood flow, because in renal and skin vessels α-adrenoceptors are predominant (for reviews, see Innes and Nickerson, 1970; Hoffman and Lefkowitz, 1995).

The cerebral and coronary arteries are of particular importance in the whole of the vascular system, because of the vital functions of the organs they supply. The cerebral circulation of many species has an abundant and dense sympathetic innervation. However, the response of the cerebral vasculature to sympathetic nerve activity is comparatively small (Bevan et al., 1980; Toda, 1983). In humans, the influence of sympathetic innervation on the tone of cerebral vasculature is weak and reflects not only a low density of innervation, but also a reduced number of α-adrenoceptors (Bevan et al., 1998a). Furthermore, the sympathetic neurogenic control of cerebral arteries decreases with decreasing diameter of the vessel, such that the human pial arteries pratically do not contract in response to nerve stimulation (VanRipper and Bevan, 1991; Bevan et al., 1998a). Whereas the maximum vasoconstriction to noradrenaline in the middle meningeal artery reaches 34% of the maximum to KCl, in the pial artery it reaches only about 10% of the maximum. The cerebral arteries of the rat and pig do not contain functional α-adrenoceptors (Bevan et al., 1987). There is also little evidence for a significant β-adrenoceptor population in cerebral arteries (Bevan et al., 1998a). However, this relative lack of postjunctional adrenoceptors does not necessarily mean a lack of influence of the sympathetic nerves on the cerebral circulation. Some influence may be exerted through a cross-talk between sympathetic nerves and other neuronal systems. The nicotine-induced relaxation in the porcine basilar artery appears to result from the activation of nicotinic receptors on the presynaptic adrenergic nerve terminals; this activation causes release of noradrenaline that activates β_1-adrenoceptors located on NOergic nerves and promotes the release of NO (Toda et al., 1995; Zhang et al., 1998; Lee et al., 2000). Another indirect effect mediated by adenoceptors is that observed in segments of rabbit middle cerebral arteries, where the activation of endothelial α_2-adrenoceptor causes a reduction in endothelin-1 production and promotes vascular relaxation (Thorin et al., 1997).

In the coronary circulation, the relative amount of α- and β-adrenoceptors and the relative functional role they play also does not fit into the general pattern of the vascular beds. In the pig, the small coronary vessels exhibit little or no α-adrenoceptor-mediated activity, and the large coronary artery contains α_1-adrenoceptors, mainly of the α_{1A}-subtype, but the functional importance of their vasoconstrictive effect is unclear (Yan et al., 1998). Also in the coronary artery of the dog, the functional role of α-adrenoceptors varies between undetectable and of little expression (Begonha et al., 1995). In vessels with spontaneous tone, isoprenaline causes concentration-dependent relaxations, whereas noradrenaline and adrenaline cause either contraction (of small magnitude) or relaxation. However, after the tone had been elevated by phenylephrine, both adrenaline and noradrenaline cause concentration-dependent relaxations with a maximum effect that sometimes did not fully antagonize the previous tone (Ross, 1976; Guimarães et al., 1993; Begonha et al., 1995). This is not true for isoprenaline, which, at any level of tone, causes a relaxation that totally antagonizes the previous contrac-

tion (Table 3). In contrast to the mesenteric, splenic, and pulmonary arteries, where β_2-adrenoceptors predominate, in the coronary arteries β_1-adrenoceptors are largely predominant if not exclusive (Begonha et al., 1995). In fact, whereas in the systemic arteries adrenaline was much more potent than noradrenaline as an agonist and the selective β_2-adrenoceptor antagonists ICI-118551 was much more potent than atenolol at antagonizing the responses to isoprenaline; in the coronary arteries, noradrenaline was more potent than adrenaline; and ICI-118551 and atenolol were equipotent as antagonists of isoprenaline (Begonha et al., 1995). Also in the dog, it was shown that the magnitude of β-adrenoceptor-mediated responses of epicardial coronary arteries is inversely related to the size of the vessel (Krauss et al., 1992). The difference was independent of α-adrenoceptors, endothelium, and second messenger processing, suggesting a mechanism based on β-adrenoceptor density (Krauss et al., 1992). As far as the splanchnic vascular bed is concerned, experiments carried out on isolated mesenteric and splenic arteries showed, at least in the dog, that α-adrenoceptor-mediated effects always predominate over β-adrenoceptor-mediated responses when adrenaline is used as agonist (Guimarães and Paiva, 1981a; Begonha et al., 1995). It is possible that β-adrenoceptors are associated only with arterioles and precapillary sphincters, which regulate the peripheral resistance observed in vivo and that are not available for studies in vitro. However, in experiments in which the hindlimb of the dog was perfused, adrenaline, despite reaching arterioles and precapillary sphincters, caused concentration-dependent increases of the perfusion pressure, showing that α-adrenoceptor-mediated vasopressor effect predominates also in this vascular bed (Teixeira, 1977). An interesting peculiarity was shown in the acral regions of the cutaneous circulation, where the vascular tone is primarily controlled by humoral mechanisms mediated at postjunctional α_2-adrenoceptors; α_1-adrenoceptors that mediate neuronally evoked constriction in the cutaneous vasculature contribute little to the sympathetic regulation of this bed (Willette et al., 1991).

VII. Influence of Maturation and Aging

Maturation and aging are associated with many alterations in vascular adrenergic mechanisms. From birth to adulthood (maturation) and from adulthood to old age (aging or senescence), important changes occur in animal models as in humans at the receptor level, neurotransmitter process, and catecholamine inactivation.

In general terms, one can accept that maturation is associated with an increase, whereas aging is associated with a reduction in the adrenergic influence on the physiological processes.

A. On α-Adrenoceptors

It is well documented that the responsiveness of vascular smooth muscle to α_1-adrenoceptor activation is present at birth (Guimarães et al., 1994) and that it changes with age, although in the majority of functional studies no important alterations in responses to noradrenaline had been demonstrated either during maturation or aging (for a review, see Docherty, 1990). In the dog mesenteric artery and rat aorta, small reductions in the responsiveness to sympathomimetic amines were reported during maturation (McAdams and Waterfall, 1986; Toda and Shimizu, 1987), whereas a decrease in α-adrenoceptor-mediated functions with aging was observed in the rat tail artery (Fouda and Atkinson, 1986) and in the rat aorta (Hyland et al., 1987; Wanstall and O'Donnell, 1989). According to Satoh et al. (1995), the potency of noradrenaline in the rat aorta increased with age from 3 to 10 weeks, but decreased from 10 to 40 weeks. In the pig coronary arteries, the endothelium-dependent relaxation to noradrenaline via the α_2-adrenoceptors decreases with aging (Murohara et al., 1991).

It has been suggested that the age-related changes in α_1-adrenoceptor-mediated vasoconstrictor responses in isolated blood vessels might result from changes in the expression of the α_1-adrenoceptor subtypes; accordingly, functional, radioligand binding, and molecular biology studies using rat aortic tissue have shown that with age the expression of the α_{1A} subtype is increased, that of the α_{1B} subtype is decreased, and that of the α_{1D}-subtype does not change (Gurdal et al., 1995a,b). However, in the pithed rat, it was shown that the selective α_{1D}-adrenoceptor antagonist BMY-7378 displaced the dose-response curve to phenylephrine in young prehypertensive SHRs, but had no effect in young WKY rats; whereas in adult WKY rats, BMY-7378 caused a greater shift in the concentration-response curve to phenylephrine than in younger animals (Villalobos-Molina et al., 1999). The presence of α_{1D}-adrenoceptors in the resistance vasculature of the prehypertensive and hypertensive rats may indicate that α_{1D}-adrenoceptors are involved in vascular hyperreactivity (Villalobos-Molina et al., 1999). This apparent contradiction may well be due to the fact that the aorta and the resistance vessels are functionally totally different. The results obtained by Xu et al. (1997) confirm that aging changes heterogeneously the expression of α_1-adrenoceptor subtypes. These authors determined the changes in mRNA levels of α_1-adrenoceptor subtypes during maturation and aging in aortae and in renal, pulmonary, and mesenteric arteries isolated from 3-, 12-, and 24-month-old rats. They observed that, in the aorta, α_{1A}-, α_{1B}-, and α_{1D}-adrenoceptors declined with aging, whereas in the renal artery there was a decrease in mRNA for the α_{1B}-adrenoceptor in aged rats. However, in mesenteric and pulmonary arteries, there were no changes in mRNA levels for any of the subtypes. The results obtained on the aggregatory

responses of human platelets in radioligand binding studies also show no important differences with maturation and aging in the affinity of ligands for the binding site (Buckley et al., 1986: Davis and Silski, 1987). Vascular contractile responsiveness seems to increase with aging, and this supersensitivity may be related to the pronounced increase in the maximal pressor effect of α_1-adrenoceptor stimulation observed in the adult pithed rat (Ibarra et al., 1997). Because α_{1D}-adrenoceptors represent the predominant subtype that mediates contraction in the aorta, carotid, and mesenteric arteries of SHRs (Villalobos-Molina and Ibarra, 1996), it may be that α_{1D}-adrenoceptors play some role in the pathogenesis/maintenance of hypertension (Ibarra et al., 1998).

The α_2-adrenoceptor-mediated negative modulation of noradrenaline release is fully developed at birth (Guimarães et al., 1991, 1994). However, while at the postjunctional level phenylephrine is equipotent in adults and neonates, indicating that postjunctional α_1-adrenoceptors do not change during maturation, UK-14304 is about 4 times more potent at inhibiting noradrenaline release evoked by electrical stimulation in adults than in neonates (Guimarães et al., 1991). A similar difference in potency of UK-14304 at inhibiting noradrenaline release in adults and neonates was also observed in mouse atria (A.U. Trendelenburg and K. Starke, personal communication). One possible explanation for this difference is that the fractional release of noradrenaline is much higher in neonates than in adults (Guimarães et al., 1991; Moura et al., 1993). Hence, the concentration of noradrenaline in the biophase during electrical stimulation is higher in neonates than in adults; consequently, the inhibitory effect of any given concentration of UK-14304 is smaller, and its IC_{50} is higher in neonates than in adults (Starke, 1972; Fuder et al., 1983). Based on the temporal and regional pattern of α_2-adrenoceptor mRNA expression in rat brain, it has been suggested that the perinatal increase in receptor density may serve specific roles in development, including neuronal migration, maturation of neurons, and mediation of sensory functions (Winzer-Serhan and Leslie, 1997; Winzer-Serhan et al., 1997a,b). According to Happe et al. (1999), α_2-adrenoceptors are functionally coupled to G-protein throughout postnatal development and, therefore, are able to mediate signal transduction upon stimulation by noradrenaline and adrenaline. In the neonatal rat lung, there is a pure and dense population of α_{2B}-adrenoceptors, which is assumed to be the only one existing in this tissue. However, the number of these receptors falls to undetectable levels in adults (Latifpour and Bylund, 1983). In mice, functionally important prejunctional α_2-adrenoceptors exist in atria and vas deferens already at the age of 1 day, which are mainly $\alpha_{2A/D}$. With maturation, the $\alpha_{2A/D}$-adrenoceptors increase their functional influence. However, the development of prejunctional α_{2C}-adrenoceptors is much more impressive. They are almost absent at birth, become influencial after birth, and reach maximum activity in adult life. In atria from adult $\alpha_{2A/D}$-adrenoceptor knockout mice, there is as much autoinhibition as in adult wild-type atria (A. U. Trendelenburg and K. Starke, personal communication).

B. On β-Adrenoceptors

In the canine saphenous vein, pre- and postjunctional β-adrenoceptor-mediated effects are lacking at birth. However, the responses to forskolin, a direct-acting stimulant that bypasses the need for β-adrenoceptors on their linkage to stimulatory G-protein subunits, are already present at birth; this shows that the lack of responses to isoprenaline is linked to either a lack or some kind of immaturity of the receptors or G-protein (Guimarães et al., 1994). Furthermore, it was shown that β_2-adrenoceptor-mediated effects and the increase in the adrenaline content of the adrenal gland have a parallel time course (Paiva et al., 1994). Thus, both the prejunctional and the postjunctional β_2-adrenoceptor-mediated effects increase with increasing age (until adulthood), as does the adrenaline content of the adrenal gland, such that at 2 weeks the β_2-adrenoceptor-mediated maximum effect is about 50% of that of the adult; and at 1 month, it is fully developed (Paiva et al., 1994). The relationship between the content of adrenaline of the adrenal medulla and the development of β_2-adrenoceptor-mediated responses was analyzed also in the rat, a species in which β_2-adrenoceptor-mediated responses develop earlier than in the dog, such that at birth these responses are already fully expressed. Interestingly, whereas the adrenaline content of the canine adrenal medulla at birth is about 3% that of the adult, in the rat it is about 50%. This suggests a link between adrenaline and the maturation of β_2-adrenoceptor-mediated effects, indicating that either adrenaline triggers the expression of β_2-adrenoceptor-mediated effects or that the expression of adrenaline formation and β_2-adrenoceptor-mediated effects are simultaneously evoked by the same event (Moura et al., 1997). Similarly, in the mouse, a species in which adrenaline represents about 60% to 70% of total catecholamines in the adrenal medulla of 1-day-old animals, prejunctional β_2-adrenoceptors fully operate at that age (A.U. Trendelenburg and K. Starke, personal communication). This hypothesis is in good agreement with the report that in early neonatal life isoprenaline, instead of producing desensitization of responses, enhances expression or efficiency of β-adrenoceptor signaling (Giannuzzi et al., 1995). Recently, it was demonstrated that agonist treatment in the neonate causes an enhancement of coupling rather than an uncoupling of receptors from G-proteins (Zeiders et al., 1999), and the reversal from enhancement of coupling to uncoupling occurs (in cardiac cells) between postnatal days 11 and 14 (Zeiders et al., 2000).

β-Adrenoceptor-mediated relaxation was compared in the pulmonary vein of the fetal (145 $\pm$ 2 days of gesta-

tion) and newborn lamb. Isoprenaline caused greater relaxation in newborn than in fetal lambs. Also, in humans, the sympathetic nerves play a more important role in the regulation of cerebrovascular tone in the infant than in the adult (Bevan et al., 1998b). Biochemical studies showed that isoprenaline and forskolin evoked a greater increase in cAMP content and in adenyl cyclase activity of pulmonary veins in the newborn than in the fetal lamb. These results show that β-adrenoceptor-mediated relaxation of the pulmonary veins increases with maturation (Gao et al., 1998). However, according to Conlon et al. (1995), there is no change in myocardial ventricle β-adrenoceptor G-protein coupling capacity or adenylate activation with aging beyond maturity. These authors showed that aging between 6 and 26 months in male Wistar rats is not accompanied by changes in myocardial β-adrenoceptor signal transduction and capacity for formation of the high-affinity β-adrenoceptor G-protein coupled complex with the agonist. It was also found that an age-related impairment of myocardial β-adrenoceptor up-regulation occurs with aging (Conlon et al., 1995).

This β-adrenoceptor-mediated relaxing capacity, which increases during the first weeks of life, then declines as the age increases. The loss of vasodilator response to isoprenaline in the rat aorta has been reported at different ages ranging from 3 to 22 months (for a review, see Docherty, 1990). There is not only a decrease in the maximum relaxation to isoprenaline with aging, which has been reported for the rabbit aorta, rat pulmonary artery, rat mesenteric artery, human saphenous vein, canine mesenteric artery, but also an increase in the EC_{50} of isoprenaline: in the aorta of 5- and 20-week-old rats preconstricted with phenylephrine, the pD_2 values for isoprenaline were 7.97 and 6.57, respectively (Borkowski et al., 1992), indicating a marked reduction in the potency of this β-adrenoceptor agonist. According to Dohi et al. (1995), with increasing age, maximum β-adrenoceptor-mediated relaxation decreases in most arteries, but not in veins. Also SHRs exhibit an age-related loss in vasodilator β-adrenoceptor responsiveness. However, the maximum relaxation to sodium nitrite or to sodium nitroprusside is not reduced (O'Donnell and Wanstall, 1986; Küng and Lüscher, 1995).

Because most studies show no change with age in the number of β_1- or β_2-adrenoceptor-binding sites of the human lymphocytes and rat heart and because cAMP production in response to forskolin and dibutyril cyclic adenosine monophosphate is also reduced by aging in the rat myocardium and human lymphocytes, it seems likely that the change is not at the receptor level but in the coupling to the adenylate cyclase via G-proteins. In healthy volunteers of different ages, isoprenaline-induced increases in heart rate were significantly greater in young than in old ones (Brodde et al., 1998). However, β-adrenoceptor numbers and subtype distribution were unchanged as determined in patients undergoing open heart surgery. The decrease in β-adrenoceptor-mediated efficiency is due to a reduced activity of the catalytic unit of the adenylyl cyclase (Brodde and Pönicke, 1998).

Prolonged or repeated exposure to β-agonists in adults results in a compensatory desensitization that reduces responsiveness (for a review, see Summers et al., 1997). In older animals, the predominant effect is heterologous desensitization mediated at the level of the G-protein. During development, however, responses in most systems increase with age and with the maturation of neuronal inputs (Giannuzzi et al., 1995). Instead of producing desensitization of responses, agonist exposure promotes receptor signaling by enhancing expression and/or catalytic efficiency of adenylyl cyclase. These developmental differences are likely to be important in the maintenance of tissue responsiveness during the period in which innervation develops (Guimarães et al., 1994; Giannuzzi et al., 1995; Moura et al., 1997).

Regarding the age-related involvement of the endothelium in β-adrenoceptor-mediated responses, it has been shown that aging reduced endothelium-dependent relaxations to acetylcholine and isoprenaline in aortas from both normotensive and SHR rats (Arribas et al., 1994; Küng and Lüscher, 1995; Satake et al., 1995; van der Zypp et al., 2000). However, the responses to the NO-donor nitroprusside sodium, which directly activates the soluble guanylyl cyclase and formation of cGMP, were very similar in adult and old rats of either strain. This indicates that the impairment of the response to acetylcholine and isoprenaline is due to functional changes of the endothelium, rather than the vascular smooth muscle (Küng and Lüscher, 1995; van der Zypp et al., 2000). Furthermore, it was observed that, in endothelium-intact aortas, the nitric oxide synthase inhibitor *L*-NMMA attenuated the isoprenaline-induced relaxation to a similar extent in both age groups, suggesting that although NO was involved in the response to isoprenaline, it cannot have been responsible for the age-related difference (van der Zypp et al., 2000). It was also observed that the age-dependent reduction in isoprenaline-mediated relaxation in aorta was greater in K^+ than in phenylephrine-constricted aortas (Borkowski et al., 1992; Chapman et al., 1999), supporting the view that the signaling pathway involved in isoprenaline-induced relaxation switches toward an increased role of K^+ channels in older rats. Thus, the signaling pathways involved in β-adrenoceptor-mediated responses are multifactorial. They include a NO-dependent pathway, that does not depend on age; an endothelium-independent pathway involving cAMP, which appears to decline with age; and a third factor apparently endothelium-dependent that involves tetraethylammonium-sensitive K^+ channels and increases with age (Satake et al., 1996, 1997; van der Zypp et al., 2000).

VIII. Influence of Temperature on Vascular Adrenoceptor-Mediated Responses

It is common knowledge that cold makes the skin pale and heat makes the skin red. In intact organisms, exposure to cold causes cutaneous veins to constrict, whereas deeper veins dilate thus transferring venous blood from the superficial to the deep circulation to reduce heat loss (Vanhoutte, 1980). In isolated veins of the dog contracted with exogenous noradrenaline or sympathetic nerve stimulation, cooling enhances (saphenous) or reduces (femoral) the contractile responses (Vanhoutte and Lorenz, 1970). Furthermore, it was reported that cooling enhances the contractile response to the selective α_2-adrenoceptor agonist UK-14304 (Flavahan and Vanhoutte, 1986b; Nunes and Guimarães, 1993), does not change that to the selective α_1-adrenoceptor agonist phenylephrine (Flavahan and Vanhoutte, 1986b; Nunes and Guimarães, 1993), and markedly reduces the response to chloroethylclonidine (Nunes and Guimarães, 1993). Similarly, cooling enhanced the α_2-adrenoceptor-mediated contractile effect evoked by electrical stimulation on the human saphenous vein (Harker et al., 1994). Flavahan and Vanhoutte (1986b) explained the different sensitivity of cutaneous and deep blood vessels to cooling on the basis of a different α_1-adrenoceptor reserve: in the saphenous vein, an enhanced α_2-adrenoceptor-mediated effect is added to a nonreduced α_1-adrenoceptor-mediated response (because in this vein there is a large α_1-adrenoceptor reserve that buffers the α_1-adrenoceptor-mediated response from the inhibitory influence of cooling); in the femoral vein, the α_2-adrenoceptor-mediated effect is so inefficient that its enhancement does not compensate for the markedly reduced α_1-adrenoceptor-mediated responses (which is depressed because there is no α_1-adrenoceptor reserve). The same authors had previously shown in the saphenous vein that under normal conditions, cooling to 24°C did not affect the responses to phenylephrine, whereas it did reduce markedly this response after partial irreversible blockade of α_1-adrenoceptors with phenoxybenzamine (Flavahan and Vanhoutte, 1986b). However, the enhancement of the α_2-adrenoceptor-mediated responses remained to be explained.

In the deer digital arteries, a different reactivity was found in winter and summer: in the cold winter, they were either insensitive or had a reduced sensitivity to the vasodilator action of histamine, compared with arteries collected in summer (Callingham et al., 1998; Milton et al., 1999).

Very recently, it was shown that, in the mouse tail artery, at 37°C, vasoconstriction to the α_2-adrenoceptor agonist UK-14304 was antagonized by the selective $\alpha_{2A/D}$-adrenoceptor antagonist BRL-44408, but was not antagonized by the α_{2B}- and α_{2C}-adrenoceptor antagonist ACR-239 or the preferential α_{2C}-adrenoceptor antagonist MK-912. However, at 28°C, the enhanced vasoconstrictor response to UK-14304 was inhibited by low concentrations of the preferential α_{2C}-adrenoceptor antagonist MK-912, whereas ACR-239 was ineffective and the selective $\alpha_{2A/D}$-adrenoceptor antagonist BRL-44408 showed an inhibitory effect that was not different from that observed at 37°C. These results indicate that, at 28°C, α_{2C}-adrenoceptors contribute to α_2-adrenoceptor-mediated vasoconstriction and probably are responsible for the supersensitivity to α_2-adrenoceptor agonists caused by cold (Chotani et al., 2000). Interestingly, this is not a phenomenon exclusively occurring with vascular smooth muscle of superficial vessels, since hypothermia enhanced α_2-adrenoceptor-mediated responses in rat vas deferens in such a way that the lack of any response to UK-14304 at 37°C was converted to evident contractions at 20°C (Gonçalves et al., 1989).

IX. Vascular Adrenoceptors in Some Diseases

Vascular adrenoceptors may be affected in many diseases, sometimes as a consequence of alterations suffered by the vessels and sometimes by participating themselves in the genesis of diseases or by being their primary cause.

For many diseases, there are no animal models. Even in well studied animal models, such as the SHRs, relevance to human "essential hypertension" is unknown. As a consequence, one often has to rely on observations with patients. Additionally, in vivo experiments in animals (e.g., measurements of blood pressure) often involve multifactorial systems, the analysis of which is far more complex than in the in vitro experiments. For instance, the determination of maximum responses of the blood pressure to pressor agents is often impossible; hence, it is difficult or impossible to provide a full and satisfactory description of an "enhanced pressor response" observed in this or that disease: is this phenomenon due to a parallel shift of the dose-response curve to the left, to an increase of the maximum response, or to both? Is this phenomenon generated by a change in the structure of the blood vessels by a change in the mechanisms that inactivate the test agonist, by a change in G-proteins or second messenger mechanisms, or in change of the adrenoceptors? Given these unavoidable limitations of attempts to delineate the role played by adrenoceptors in various diseases, it is not surprising that hard facts are rare. However, such studies have provided valuable hints.

In 1929, Lewis postulated that Raynaud's disease resulted from a "local fault" of the blood vessel wall. This fault could be an anomalous regulation of α_2-adrenoceptors. It appears now clear that α_2-adrenoceptors play a role in the development of Raynaud's disease (Freedman et al., 1995; Chotani et al., 2000). Cold augments constriction to α_2-adrenoceptor activation without affecting the responses to α_1-adrenoceptor stimulation or to any other vasoconstrictor agent (Flavahan and Vanhoutte,

1986a; Chotani et al., 2000). Nonselective α_2-adrenoceptor antagonists abolish cold-induced vasospastic crises in patients with primary Raynaud's disease (Freedman et al., 1995). However, according to many authors, there is no abnormal reactivity of α_2-adrenoceptors in subjects with primary Raynaud's disease (Lindblad et al., 1989; Coffman and Cohen, 1990; Freedman et al., 1993). Therefore, the local fault in Raynaud's crisis may represent a consequence of a cold-induced functional expression of α_{2C}-adrenoceptors that appear to be silent at normal temperature (Chotani et al., 2000). Thus, inhibition of α_{2C}-adrenoceptors may provide a highly selective therapeutic measure for this disease.

Vasospasm and ischemic organ injury are functional changes that play an important role in the pathogenesis of scleroderma (Kahaleh, 1990). These functional changes were attributed to a failure of vascular endothelium in releasing nitric oxide (Freedman et al., 1999). However, in arterioles isolated from uninvolved skin of patients with scleroderma, the constrictor responses to the selective α_2-adrenoceptor agonist UK-14304 were increased, whereas those to KCl or the selective α_1-adrenoceptor agonist phenylephrine were similar to controls. This selective increase in the reactivity of α_2-adrenoceptors was not altered by removing the endothelium, indicating that the enhanced constrictor effect was not due to changes in endothelial dilator activity, but to an enhancement of the α_2-adrenoceptor-mediated responses of the vascular smooth muscle cells (Flavahan et al., 2000).

In several neurological degenerative and genetic disorders, there are also important changes in α-adrenoceptor-mediated responses. In the majority of these situations, the pathological process involves primarily the sympathetic postganglionic neurones, leading to a progressive denervation of some organs, including the blood vessels. The most prominent cardiovascular symptom in all these conditions is orthostatic hypotension, which is a common complaint due to a sympathetic neurocirculatory failure and sometimes forces the patients to be bedridden, even if they are still able to work. In the familial amyloidotic polyneuropathy, an autosomal dominant disorder with an estimated prevalence of about 1/1000 in the population of the most affected areas in the northwest of Portugal (Andrade, 1952; Carvalho et al., 1997), there is a progressive degeneration of the sympathetic nerves leading to complete denervation. Concomitantly, there is a marked supersensitivity to the vasoconstrictor action of noradrenaline (Falcão-de-Freitas, 1996; Carvalho et al., 1997), probably due to an up-regulation of α-adrenoceptors. It is not yet known whether all α-adrenoceptor subtypes are equally involved or some particular subtype(s) is predominantly implicated. A similar enhancement of α-adrenoceptor-mediated responses occurs in patients with congenital dopamine-hydroxylase deficiency (Man in't Veld et al., 1987). In this disorder, there is no conversion of dopamine to noradrenaline causing a lack of the transmitter at postganglionic sympathetic neurones (Man in't Veld et al., 1987; Rea et al., 1990). Recently, it was shown that many patients with Parkinson's disease have evidence of peripheral sympathetic denervation causing a deficient release of noradrenaline in the heart and blood vessels with a consequent α-adrenoceptor up-regulation (Magalhaes et al., 1995; Netten et al., 1995; Goldstein et al., 2000). In diabetic polyneuropathy, there is also an autonomic dysfunction leading to a progressive sympathetic denervation and to a more or less marked increase in α-adrenoceptor-mediated responses (for review, see Watkins, 1998). There are other degenerative neurological diseases, like the multiple system atrophy (Shy Drager syndrome), in which the main pathological changes leading to the neurocirculatory failure with severe orthostatic hypotension occur in the central nervous system without marked alterations of the peripheral sympathetic nerves (Zoukos et al., 1999; Goldstein et al., 2000). Very recently, it was shown that overexpression of the α_{1B}-adrenoceptor causes apoptotic neurodegeneration with a corresponding multiple system atrophy (Zuscik et al., 2000). The resulting symptoms (impaired hindlimb function and seizures) could be rescued with the α_1-adrenoceptor antagonist terazosin, indicating that α_1-adrenoceptors participated directly in the pathology. These findings suggest a link between α_{1B}-adrenoceptor function and the etiology of Shy-Drager syndrome (Zuscik et al., 2000).

Sometimes, β-adrenoceptors are also involved in these neurological degenerative diseases. Both in multiple sclerosis and multiple system atrophy, there is sympathetic neurocirculatory failure with supersensitivity to α-adrenoceptor agonists. However, whereas in multiple sclerosis there is also an up-regulation of the β-adrenoceptors expressed on peripheral blood mononuclear cells, in multiple system atrophy there is not (Zoukos et al., 1999).

According to many authors, α-adrenoceptors may be involved in the pathogenesis/maintenance of some kinds of hypertension. Not only sensitivity to salt is a common trait in patients with essential hypertension, but there is also experimental evidence suggesting that salt loading causes hypertension via a mechanism involving α_2-adrenoceptors. A recent comparison of the effect of subtotal nephrectomy and salt loading in α_{2B}-adrenoceptor knockout mice, in α_{2C}-adrenoceptor knockout mice, and in wild-type mice showed that only the α_{2B}-adrenoceptor knockout mice have no significant increase in blood pressure (Makaritsis et al., 1999). Both the wild-type and α_{2C}-adrenoceptor knockout mice had significant blood pressure increases, indicating that α_{2B}-adrenoceptors are relevant for the development of this kind of hypertension. On the other hand, some data draw attention to the possible role played by changes in α_1-adrenoceptors in the development/maintenance of hypertensive states. In the resistance vasculature of young prehyper-

tensive and hypertensive rats, a high density of α_{1D}-adrenoceptors was found (Stassen et al., 1997; Ibarra et al., 1998; Xu et al., 1998). Furthermore, in endothelium-denuded tail artery and aorta, the maximum contractile response to phenylephrine and chloroethylclonidine was higher in SHR than in WKY rats (Villalobos-Molina et al., 1999; Ibarra et al., 2000), suggesting that α_{1D}-adrenoceptors may be involved in vascular supersensitivity leading (or in some way being linked) to the hypertensive state. It is also possible that changes at postadrenoceptor level, such as altered levels of G-proteins may be related to the higher reactivity to agonists in hypertension (Li et al., 1994; Kanagy and Webb, 1996). These results indicate that future studies with α_{1D}-adrenoceptor knockout animals may be helpful to explain the role (if any) of these alterations in genesis/maintenance of hypertension.

Trp64Arg mutation of β_3-adrenoceptor has been suggested to confer susceptibility to essential hypertension (Morris et al., 1994); this thesis was contested by Fujisawa et al. (1997) and confirmed by Tonolo et al. (1999). These authors concluded that the Trp64Arg polymorphism of the β_3-adrenoceptor gene is associated more often with high blood pressure than with normal blood pressure.

A naturally occurring variation found in about 8% of Europeans and North Americans actually restores in humans the arginine residue present in animals (Strosberg, 1997). This variation was found to be associated with 1) an increased capacity of obese French patients to gain weight (Clément et al., 1995); 2) an early onset of noninsulin-dependent diabetes mellitus in obese Pima Indians (Walston et al., 1995); and 3) an early onset of noninsulin-dependent diabetes mellitus and clinical features of the insulin resistance syndrome in Finns (Widén et al., 1995). Although these alterations are related to β_3-adrenoceptors in adipocytes, they may be particularly important since it is believed that in terms of the risks of cardiovascular disorders, visceral obesity is the most dangerous form of regional fat accumulation, the form of obesity that is more directly linked to β_3-adrenoceptor activity (Arner, 1995).

X. Conclusions

New possibilities are now offered by molecular biology (knockout animals, genetically altered receptors, measurements of mRNA, etc.) that will help in clarifying receptor function. However, data obtained in experiments carried out in knockout animals must be carefully interpreted, keeping in mind the multiple ways to compensate for the lack of this (or these) adrenoceptor subtype(s).

Regarding the subclassification of adrenoceptors, two controversial points are now on the way to being solved: the existence of a fourth α_1-adrenoceptor subtype (the α_{1L}-adrenoceptor) and a fourth β-adrenoceptor (the β_4-adenoceptor). It is now accepted that these hypothetical subtypes correspond to low-affinity states of the α_{1A}- and the β_1-adrenoceptors, respectively.

The pharmacology of human α_1-adrenoceptors often differs from that of the corresponding α_1-adrenoceptor subtypes of experimental animals. Then, the identification of α_1-adrenoceptor subtypes present in human vasculature may be useful for the discovery of new selective compounds effective in the treatment of prostatic hypertrophy, pulmonary hypertension, and coronary insufficiency.

A fascinating point is that, in the vast majority of the organs, the adrenoceptors expressed there do not correspond to the functional roles they play. The potential role of some adrenoceptor subtypes apparently unimportant under normal conditions, should be kept in mind and carefully taken into consideration. It has been described that some sodium channels (II and III α-isoforms), which are functionally important during the earlier stages of life, loose their important roles in adult life and reappear functionally active under some pathological conditions (Aronica et al., 2001). This fact should be linked to the interesting fact that expression and function of a given adrenoceptor subtype changes their role during a lifetime. For example the α_{2B}-adrenoceptor, which is densely represented during intrauterine development, disapears after birth, whereas the α_{1D}-adrenoceptors that have no important role under physiological conditions may become important in some hypertensive states. This dynamic balance can also be exemplified by the fact that α_{2C}-adrenoceptors, which at 37°C are nonfunctional, become functionally predominant at lower temperatures.

Prejunctional $\alpha_{2A/D}$-adrenoceptors are now well established to be primarily responsible for the regulation of the release of noradrenaline under physiological conditions. However, also, α_{2C}-adrenoceptors play a minor role in this regulatory mechanism.

The existence of α- and β-adrenoceptors in the endothelium and the importance of the endothelial system in the physiology and pathophysiology of the vascular system has to be considered; however, it is intriguing that the activation of α- and β-adrenoceptors lead to the same effect: an increase in NO formation/release.

From a therapeutic standpoint (and as far as β-adrenoceptors are concerned), there are many instances where β-adrenoceptor-subtype selective stimulation (asthma, atrioventricular block, obesity) or block (hypertension, coronary insufficiency) is desired. Therefore, a still more detailed knowledge of subtype-specific functions is necessary as drugs, which are more selective, are required.

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REFERENCES

Aboud R, Shafi M and Docherty JR (1993) Investigation of the subtypes of α_1-adrenoceptor mediating contraction of rat aorta, vas deferens and spleen. *Br J Pharmacol* **109:**80–87.

Abrahamsen J and Nedergaard OA (1989) Presynaptic action of adrenaline on adrenoceptors modulating stimulation-evoked ^{3}H-noradrenaline release from rabbit isolated aorta. *Naunyn-Schmiedeberg's Arch Pharmacol* **339:**281–287.

Abrahamsen J and Nedergaard OA (1991) Lack of presynaptic modulation by isoprenaline of ^{3}H-noradrenaline release from rabbit isolated ear artery. *Naunyn-Schmiedeberg's Arch Pharmacol* **343:**161–165.

Adler-Graschinsky E and Langer SZ (1975) Possible role of a β-adrenoceptor in the regulation of noradrenaline release by nerve stimulation through a positive feedback mechanism. *Br J Pharmacol* **53:**43–50.

Ahlquist RF (1948) A study of the adrenotropic receptors. *Am J Physiol* **153:**586–600.

Alabaster VA, Keir RF and Peters CJ (1986) Comparison of the potency of α_2-adrenoceptor antagonists *in vitro*: evidence for heterogeneity of α_2-adrenoceptors. *Br J Pharmacol* **88:**607–615.

Alexander SPH and Peters JA (1999) *Receptor and Ion Channel Nomenclature Supplement*, 10th ed, Trends in Pharmacological Sciences, Cambridge, UK.

Altman JD, Trendelenburg AU, MacMillan L, Bernstein D, Limbird L, Starke K, Kobilka BK and Hein L (1999) Abnormal regulation of the sympathetic nervous system in α_{2A}-adrenergic receptor knockout mice. *Mol Pharmacol* **56:**154–161.

Altura BM (1971) Chemical and humoral regulation of blood flow through the precapillary sphincter. *Microvasc Res* **3:**361–384.

Andrade C (1952) A peculiar form of peripheral neuropathy: familial atypical generalized amyloidosis with special involvement of peripheral nerves. *Brain* **75:**408–427.

Angus JA, Cocks TM and Satoh K (1986) The α-adrenoceptors on endothelial cells. *Fed Proc* **45:**2355–2359.

Apparsundaram S and Eikenburg DC (1995) Role of prejunctional *beta*-adrenoceptors in rat cardiac sympathetic neurotransmission. *J Pharmacol Exp Ther* **272:**519–526.

Arch S Jr (1998) Pharmacology of β_3- and putative "β_4"-adrenoceptors. *Pharmacol Toxicol* **83:**40–41.

Arch S Jr, Ainsworth AT, Cawthorne MA, Piercy V, Sennit MV, Thody VE, Wilson C and Wilson S (1984) Atypical β-adrenoceptor on brown adipocytes as target for anti-obesity drugs. *Nature (Lond)* **309:**163–165.

Arch S Jr and Kaumann AJ (1993) Beta 3 and atypical beta-adrenoceptors. *Med Res Rev* **13:**663–729.

Argyle SA and McGrath JC (2000) An α_{1A}-α_{1L}-adrenoceptor mediates contraction of canine subcutaneous resistance arteries. *J Pharmacol Exp Ther* **295:**627–633.

Arner P (1995) The β_3-adrenergic receptor—a cause and a cure of obesity? *N Engl J Med* **333:**382–383.

Aronica E, Yankaya D, vanVliet E, Lopes da Silva F and Gorter J (2001) Induction of neonatal channel II and III *alpha*-isoform mRNAs in neurons and microglia after *stautus epilepticus* in the rat. *Eur J Neurosci* **13:**1261–1266.

Arribas S, Marin J, Ponte A, Balfagon G and Salaices M (1994) Norepinephrine-induced relaxations in aorta mediated by endothelial *beta*-adrenoceptors. Impairment by aging and hypertension. *J Pharmacol Exp Ther* **270:**520–527.

Asano T, Katada T, Gilman AG and Ross EM (1984) Activation of the inhibitory protein of adenylate cyclase, Gi, by beta-adrenergic receptors in reconstituted phospholipid vesicles. *J Biol Chem* **259:**9351–9354.

Atef N, Lafontan M, Doublé A, Hélary C, Ktorza A and Pénicaud L (1996) A specific β_3-adrenoceptor agonist induces increased pancreatic islet blood flow and insulin secretion in rats. *Eur J Pharmacol* **298:**287–292.

Barnes PJ (1995) Beta-adrenergic receptors and their regulation. *Am J Respir Crit Care Med* **152:**838–860.

Barrow JC, Nantermet PG, Selnick HG, Glass KL, Rittle KE, Gilbert KF, Steele TG, Homnick CF, Freidinger RM, Ransom RW, Kling P, Reiss D, Broten TP, Schorn TW, Chang RS, O'Malley SS, Olah TV, Ellis JD, Barrish A, Kassahun K, Leppert P, Nagarathnam D and Forray C (2000) In vitro and in vivo evaluation of dihydropyrimidinone C-5 amides as potent and selective alpha (1A) receptor antagonists for the treatment of benign prostatic hyperplasia. *J Med Chem* **43:**2703–2718.

Begonha R, Moura D and Guimarães S (1995) Vascular β-adrenoceptor-mediated relaxation and the tone of the tissue in canine arteries. *J Pharm Pharmacol* **47:**510–513.

Berlan M, Galitzky J, Bousquet-Melou A, Lafontan M and Montrastruc JL (1994) *Beta*-3 adrenoceptor-mediated increase in cutaneous blood flow in the dog. *J Pharmacol Exp Ther* **268:**1444–1451.

Berthelsen S and Pettinger WA (1977) A functional basis for classification of α-adrenergic receptors. *Life Sci* **21:**595–606.

Bevan JA (1977) Some functional consequences of variation in adrenergic synaptic cleft width and in nerve density and distribution. *Fed Proc* **36:**2439–2443.

Bevan JA, Bevan RD and Duckles SP (1980) Adrenergic regulation of vascular smooth muscle, in *Handbook of Physiology—Cardiovascular System* (Bohr DF, Somlyo AP and Sparks HV eds) pp 515–566, Williams & Wilkins Company, Baltimore.

Bevan RD, Dodge J, Nichols P, Penar PL, Walters CL, Wellman T and Bevan JA (1998a) Weakness of sympathetic neural control of human pial compared with superficial temporal arteries reflects low innervation density and poor sympathetic responsiveness. *Stroke* **29:**212–221.

Bevan RD, Dodge J, Nichols P, Poseno T, Vijayakumaran E, Wellman T and Bevan JA (1998b) Responsiveness of human infant cerebral arteries to sympathetic nerve stimulation and vasoactive agents. *Pediatr Res* **44:**730–739.

Bevan JA, Duckworth J, Laher I, Oriowo MA, McPherson GA and Bevan RD (1987) Sympathetic control of cerebral arteries: specialization in receptor type, reserve, affinity and distribution. *FASEB J* **1:**193–198.

Black JW, Crowther AF, Shanks RG, Smith LH and Dornhorst AC (1964) A new adrenergic beta-receptor antagonist. *Lancet* **I:**1080–1081.

Blankestijn P, Tulen J, Boomsma F, Van Eck HJR, Mulder P, Man in't Veld AJ, Van Den Meiracker AH, Moleman P, Derkx FHM, Lamberts SJ and Schlekamp M (1988) Support for adrenaline-hypertension hypothesis: 18 hour pressor effect after 6 hours adrenaline infusion. *Lancet* **ii:**1386–1389.

Blaylock NA and Wilson VG (1995) Pharmacological characterization of noradrenaline-induced contractions of the porcine isolated palmer lateral vein and palmar common digital artery. *Br J Pharmacol* **114:**694–702.

Bloor BC, Frankland M, Alper G, Raybould D, Weitz J and Shurtliff M (1992) Hemodynamic and sedative effects of dexmedetomidine in dog. *J Pharmacol Exp Ther* **263:**690–697.

Blue DR, Bond RA, Adham N, Delmondo R, Michel A, Eglen RM, Whiting RL and Clarke DE (1989) Interaction of dihydroalprenolol and cyanopindolol with atypical β-adrenoceptors in guinea-pig ileum. *Br J Pharmacol* **96:**242P.

Bockman CS, Gonzalez-Cabrera I and Abel PW (1996) Alpha-2 adrenoceptor subtype causing nitric oxide-mediated vascular relaxation in rats. *J Pharmacol Exp Ther* **278:**1235–1243.

Bockman CS, Jeffries WB and Abel PW (1993) Binding and functional characterization of *alpha*-2 adrenergic receptor subtypes on pig vascular endothelium. *J Pharmacol Exp Ther* **267:**1126–1133.

Bockman CS, Jeffries WB, Pettinger WA and Abel PW (1992) Enhanced release of endothelium-derived relaxing factor in mineralocorticoid hypertension. *Hypertension* **20:**304–313.

Boer R, Grassegger A, Scudt C and Glossmann H (1989) (+)-Niguldipine binds with very high affinity to Ca^{2+} channels and to a subtype of α_1-adrenoceptors. *Eur J Pharmacol* **172:**131–145.

Bohr DF (1967) Adrenergic receptors in coronary arteries. *Ann NY Acad Sci* **139:**799–807.

Bojanic D, Jansen JD, Nahorski SR and Zaagsma J (1985) Atypical characterization of the β-adrenoceptor mediating cAMP generation lipolysis in the rat adipocyte. *Br J Pharmacol* **84:**131–137.

Borkowski KR (1991) Effect of adrenal demedullation and adrenaline on hypertension development and vascular reactivity in young spontaneously hypertensive rats. *J Auton Pharmacol* **11:**1–14.

Borkowski KR, Gross R and Schneider H (1992) Vascular β-adrenoceptor-mediated responses in hypertension and ageing rats. *J Auton Pharmacol* **12:**389–401.

Borkowski KR and Quinn P (1985) Adrenaline and the development of spontaneous hypertension in rats. *J Auton Pharmacol* **5:**89–100.

Boudreau G, Péronnet F, De Champlain J and Nadeau R (1993) Presynaptic effects of epinephrine on norepinephrine release from cardiac sympathetic nerves in dogs. *Am J Physiol* **265:**H205–H211.

Brawley L, MacDonald S and Shaw AM (1998) Role of endothelium in classical and atypical β-adrenoceptor-mediated vasorelaxation in rat isolated aorta. *Br J Pharmacol* **122:**395.

Brawley L, Shaw AM and MacDonald A (2000a) β_1-β_2- and atypical β-adrenoceptor-mediated relaxation in rat isolated aorta. *Br J Pharmacol* **129:**637–644.

Brawley L, Shaw AM and MacDonald A (2000b) Role of endothelium/nitric oxide in atrypical β-adrenoceptor-mediated relaxation in rat isolated aorta. *Eur J Pharmacol* **398:**285–296.

Brodde O-E, Konschak U, Becker K, Rüter F, Poller U, Jakubetz J and Radke J (1998) Cardiac muscarinic receptors decrease with age. *J Clin Invest* **101:**471–478.

Brodde O-E and Michel M (1999) Adrenergic and muscarinic receptors in the human heart. *Pharmacol Rev* **51:**651–689.

Brodde O-E and Pönicke K (1998) Age-dependent changes of human cardiac β-adrenoceptors. *Pharmacol Toxicol* **83:**35–36.

Brown AM (1990) Regulation of heartbeat by G protein-coupled ion channels. *Am J Physiol* **259:**H1621–H1628.

Brown MJ and Macquin I (1981) Is adrenaline the cause of essential hypertension? *Lancet* **ii:**1079–1081.

Bryan LJ, Cole JJ, O'Donnell SR and Wanstall JC (1981) A study designed to explore the hypothesis that *beta*-1 adrenoceptors are "innervated" receptors and *beta*-2 adrenocepors are "hormonal" receptors. *J Pharmacol Exp Ther* **216:**395–400.

Buckley C, Curtin D, Walsh T and O'Malley K (1986) Ageing and platelet α_2-adrenoceptors. *Br J Clin Pharmacol* **21:**721–722.

Bünemann M, Lee KB, Pals-Rylaarsdam R, Roseberry AG and Hosey MM (1999) Desensitization of G-protein-coupled receptors in the cardiovascular system. *Ann Rev Physiol* **61:**169–192.

Burt RP, Chapple CR and Marshall I (1995) Evidence for a functional α_{1A}-(α_{1c}) adrenoceptor mediating contraction of the rat epididymal vas deferens and an α_{1B}-adrenoceptor mediating contraction of the rat spleen. *Br J Pharmacol* **115:**467–475.

Burt RP, Chapple CR and Marshall I (1998) α_{1A}-Adrenoceptor mediated contraction of rat prostatic vas deferens involves ryanodine Ca^{2+} stores and Ca^{2+} influx stimulated by diacylglycerol and PKC. *Br J Pharmacol* **123:**317–325.

Busse R, Fisslthaler B and Fleming I (1998) Activation of the endothelial nitric oxide synthase by fluid shear stress. *Pharmacol Toxicol* **83:**47–48.

Buxton BF, Jones CR, Molenaar P and Summers RJ (1987) Characterization and autoradiographic localization of β-adrenoceptor subtypes in human cardiac tissues. *Br J Pharmacol* **92:**299–310.

Bylund DB, Blaxall HS, Iversen LJ, Caron MG, Lefkowitz RJ and Lomasney JW (1992) Pharmacological characteristics of α_2-adrenergic receptors: comparison of pharmacologically defined subtypes with subtypes identified by molecular cloning. *Mol Pharmacol* **42:**1–5.

Bylund DB and Chacko DM (1999) Characterization of α_2 adrenergic receptor subtype in human ocular tissue homogenates. *Invest Ophthalmol Vis Sci* **40:**2299–2308.

Bylund DB, Eikenberg DC, Hieble JP, Langer SZ, Lefkowitz JR, Minneman KP, Molinoff PB, Ruffolo RR and Trendelenburg U (1994) IV International Union of Pharmacology Nomenclature of Adrenoceptors. *Pharmacol Rev* **46:**121–136.

Bylund DB, Hass NA, Cerutis DR and Blaxall HS (1995a) Characterization of alpha-2 adrenergic receptor subtypes. *Pharmacol Commun* **6:**87–90.
Bylund DB, Ray-Prenger C and Murphy TJ (1988) α_{2A} and α_{2B} Adrenergic receptor subtypes: antagonist binding in tissues and cell lines containing only one receptor subtype. *J Pharmacol Exp Ther* **245:**600–607.
Bylund DB, Regan JW, Faber JE, Hieble JP, Triggle CR and Ruffolo RR Jr (1995b) Vascular α-adrenoceptors: from the gene to the human. *Can J Physiol Pharmacol* **73:**533–543.
Callingham BA, Carr G and Milton AS (1998) The responses of the digital artery of the follow deer, *Dama dama*, in a mild winter. *J Physiol* **511P:**2P.
Candelore MR, Deng L, Tota L, Guan X-M, Amend A, Liu Y, Newbold R, Cascieri MA and Weber AE (1999) Potent and selective human β_3-adrenergic receptor antagonists. *J Pharmacol Exp Ther* **290:**649–655.
Carvalho MJ, Meiracker AH, Boomsma F, Man in't Veld AJ, Freitas J, Costa O and Falcão-de-Freitas A (1997) Improved orthostatic tolerance in familiar amyloidotic polyneuropathy with unnatural noradrenaline precursor L-*threo*-3,4-dihydroxyphenylserine. *J Auton Nerv Syst* **62:**63–71.
Cavalli A, Lattion A-L, Hummler E, Ninniger M, Pedrazzini T, Aubert J-F, Michel MC, Yang MN, Lembo G, Vecchione C, Mostardini M, Schmidt A, Beermann F and Cotecchia S (1997) Decreased blood pressure response in mice deficient of the α_{1b}-adrenergic receptor. *Proc Natl Acad Sci USA* **94:**11589–11594.
Chang PC, Kriek E and van Brummelen P (1994) Sympathetic activity and presynaptic adrenoceptor function in patients with longstanding essential hypertension. *J Hypertension* **12:**179–190.
Chang PJ, Chang TK, Yamanishi SS, Salazar FHR, Kosaka AH, Khare R, Bhakta S, Jasper JR, Shieh IS, Lesnick JD, Ford APDW, Daniels DV, Eglen RM, Clarke DE, Bach C and Chan HW (1998) Molecular cloning, genomic characterization and expression of novel human α_{1A}-adrenoceptor isoforms. *FEBS Lett* **422:**279–283.
Chapman J, Schutzer W, Watts VJ and Mader SL (1999) Impaired cholera toxin relaxation with age in rat aorta. *J Gerontol* **54:**B154–B159.
Chapple CR and Chess-Williams R (1999) *Curr Opin CPNS Invest Drugs* **1:**231–238.
Chaudry A, MacKenzie RG, Georgic LM and Granneman JG (1994) Differential interaction of beta (1)- and beta (3)-adrenergic receptors with G (i) in rat adipocytes. *Cellular Signalling* **6:**457–465.
Chen L, Xin X, Eckhart AD, Yang N and Faber JE (1995) Regulation of vascular smooth muscle growth by alpha 1-adrenoceptor subtypes in vitro and in situ. *J Biol Chem* **270:**30980–30988.
Chen XH, Harden TK and Nicholas RA (1994) Molecular cloning and characterization of a novel beta-adrenergic receptor. *J Biol Chem* **269:**24810–24819.
Chotani MA, Flavahan S, Mitra S, Daunt D and Flavahan NA (2000) Silent α_{2C}-adrenergic receptors enable cold-induced vasoconstriction in cutaneous arteries. *Am J Physiol* **278:**H1075–H1083.
Chruscinski AJ, Rohrer DK, Schauble E, Desai KH, Bernstein D and Kobilka BK (1999) Targeted disruption of the β_2 adrenergic receptor gene. *J Biol Chem* **274**: 16694–16700.
Clark BJ and Bertholet A (1983) Effects of pindolol on vascular smooth muscle. *Gen Pharmacol* **14:**117–119.
Clément K, Vaisse C, Manning B, Basdevant A, Guy-Grand B, Ruiz J, Silver KD, Shuldiner A, Froguel P and Strosberg AD (1995) Genetic variation in the β_3-adrenergic receptor and an increased capacity to gain weight in patients with morbid obesity. *N Engl J Med* **333:**352–354.
Cocks TM and Angus JA (1983) Endothelium-dependent relaxation of coronary arteries by noradrenaline and serotonin. *Nature (Lond)* **305:**627–630.
Coffman J and Cohen RA (1990) Alpha 2-adrenergic and 5-HT_2 receptor hypersensitivity in Raynaud's phenomenon. *J Vasc Med Biol* **2:**100–106.
Cohen RA, Shepherd JT and Vanhoutte PM (1983) 5-Hydroxytryptamine can mediate endothelium-dependent relaxation of coronary arteries. *Am J Physiol* **245:** 1077–1080.
Cohen RA, Shepherd JT and Vanhoutte PM (1984) Neurogenic cholinergic prejunctional inhibition of sympathetic beta adrenergic relaxation in the canine coronary artery. *J Pharmacol Exp Ther* **229:**417–421.
Conlon D, Johnston A, O'Malley K, Turner P and Kilfeather S (1995) Effect of ageing and propranolol administration on myocardial β-adrenoceptor receptor function in mature rats. *Eur J Pharmacol* **289:**283–290.
Connaughton S and Docherty JR (1990) Functional evidence for heterogeneity of peripheral prejunctional α_2-adrenoceptors. *Br J Pharmacol* **101:**285–290.
Constantine JW, Lebel W and Archer R (1982) Functional postsynaptic α_2- but not α_1-adrenoceptors in dog saphenous vein exposed to phenoxybenzamine. *Eur J Pharmacol* **85:**325–329.
Cooke JP, Shepherd JT and Vanhoutte PM (1984) The effect of warming on adrenergic neurotransmission in canine cutaneous vein. *Circ Res* **54:**547–553.
Coopes RP, Smit J, Benthem L, Van der Leest J and Zaasgma J (1995) Co-released adrenaline markedly facilitates noradrenaline overflow through prejunctional β_2-adrenoceptors during swimming exercise. *Eur J Pharmacol* **274:**33–40.
Coopes RP, Smit J, Khali NN, Brouwer F and Zaasgma J (1993) Strong activation of vascular prejunctional β_2-adrenoceptors in freely moving rats by adrenaline released as a co-transmitter. *Eur J Pharmacol* **243:**273–279.
Cotecchia S, Kobilka BK, Daniel KW, Nolan RD, Lapetina EY, Caron MG, Lefkowitz RJ and Regan JW (1990) Multiple second messenger pathways of α-adrenergic receptor subtypes expressed in eukaryotic cells. *J Biol Chem* **265:**63–69.
Cotecchia S, Schwinn DA, Randall RR, Lefkowitz RJ, Caron MG and Kobilka BK (1988) Molecular cloning and expression of the cDNA for hamster α_1-adrenergic receptor. *Proc Natl Acad Sci USA* **85:**7159–7163.
Dahlöf C, Kahan T and Ablad B (1978) Increased noradrenaline release in rat portal vein during sympathetic nerve stimulation due to activation of presynaptic β-adrenoceptors by noradrenaline and adrenaline. *Eur J Pharmacol* **50:**75–78.
Dainty IA, McGrath JC, Spedding M and Templeton Agb (1990) The influence of the initial stretch and the agonist-induced tone on the effect of basal and stimulated release of EDRF. *Br J Pharmacol* **100:**767–773.
Dale HH (1905) On some physiological actions of ergot. *J Physiol* **34:**163–206.
Daniel EE, Brown RD, Wang YF, Low AM, Lu-Chao H and Kwan CY (1999) α-Adrenoceptors in canine mesenteric artery are predominantly 1A-subtype: pharmacological and immunochemical evidence. *J Pharmacol Exp Ther* **291:**671–679.
Daniel EE, Gaspar V, Berezin I and Kwan CY (1995) Characterization of α_2-adrenoceptors and other adrenoceptors in membranes isolated from dog mesenteric nerve axons. *J Pharmacol Exp Ther* **275:**978–986.
Davis BJ, Noble A, Chapple CR and Chess-Williams R (2000) Human prostate-artery selectivity of tamsulosin, doxazosin and alfuzosin. *Br J Pharmacol* **129:**52P.
Davis PD and Silski C (1987) Ageing and the α_2-adrenergic system of the platelet. *Clin Sci* **73:**507–513.
Dawes M, Chowienczyk PJ and Ritter JM (1997) Effects of inhibition of the L-arginine/nitric oxide pathway on vasodilation caused by β-adrenergic agonists in human forearm. *Circulation* **95:**2293–2297.
Delmas P, Abogadie FC, Milligan G, Buckley NJ and Brown DA (1999) $\beta\gamma$ dimmers derived from G_o and G_i proteins contribute to different components of adrenergic inhibition of Ca^{2+} channels in rat sympathetic neurons. *J Physiol* **518:**23–36.
Delpy E, Coste H, Le Monnier De and Gouville A-C (1996) Effects of cyclic GMP elevation on isoprenaline-induced increase in cyclic AMP and relaxation in rat aortic smooth muscle: role of phosphodiesterase 3. *Br J Pharmacol* **119:**471–478.
De Mey J and Vanhoutte PM (1981) Uneven distribution of postjunctional alpha$_1$- and alpha$_2$-like adrenoceptors in canine arterial and venous smooth muscle. *Circ Res* **48:**875–884.
De Ponti F, Braziano G, Croci T, Arcidiaco M, Crema F and Manara L (1996) Functional evidence of atypical β_3-adrenoceptors in the human colon using the β_3-adrenoceptor antagonist SR 59,230. *Br J Pharmacol* **117:**1374–1376.
DeQuattro VL and Chan S (1972) Raised plasma catecholamines in some patients with primary hypertension. *Lancet* **1:**806–809.
Desai KH, Sato R, Schauble E, Barsh GS, Kobilka BK and Bernstein D (1997) Cardiovascular indexes in the mouse at rest and with exercise: new tools to study models of cardiac disease. *Am J Physiol* **272**:H1053–H1061.
Dixon RA, Kobilka BK, Strader DJ, Benovic JL, Dohlman HG, Frielle T, Bolanowski MA, Bennett CD, Rands E, Diehl RE, Mumford RA, Slater EE, Sigal IS, Caron MG, Lefkowitz RJ and Strader CD (1986) Cloning of the gene and cDNA for mammalian beta-adrenergic receptor and homology with rhodopsin. *Nature (Lond)* **321:** 75–79.
Docherty JR (1990) Cardiovascular responses in ageing: a review. *Pharmacol Rev* **42:**103–125.
Docherty JR (1998) Subtypes of functional α_1- and α_2-adrenoceptors. *Eur J Pharmacol* **361:**1–15.
Docherty JR and Starke K (1981) Postsynaptic α-adrenoceptor subtypes mediating nerve-evoked contractions in rabbit blood vessels *in vitro*. *Br J Pharmacol* **74:** 803P.
Doggrell SA (1990) Relaxant and beta 2-adrenoceptor blocking activities of (±)-, (+)- and (−)-pindolol on the rat isolated aorta. *J Pharm Pharmacol* **42:**444–446.
Dohi Y, Kojima M, Sato K and Luscher TF (1995) Age-related changes in vascular smooth muscle and endothelium. *Drugs Aging* **7:**278–291.
Donoso AO and Barontini M (1986) Increase in plasma catecholamines by intraventricular injection of histamine. *Naunyn-Schmiedeberg's Arch Pharmacol* **334:**188–192.
Dubocovich ML and Langer SZ (1974) Negative feedback regulation of noradrenaline release by nerve stimulation in the perfused cat's spleen: differences in potency of phenoxybenzamine in blocking the pre- and post-synaptic receptors. *J Physiol* **237:**505–519.
Dumas M, Dumas J-P, Bardou M, Rochette L, Advenier C and Giudicelli J-F (1998) Influence of β-adrenoceptor agonists on the pulmonar circulation. Effects of a β_3-adrenoceptor antagonist, SR 59230A. *Eur J Pharmacol* **348:**223–228.
Dybvik T, Osnes J-B and Skomedal T (1999) Similar localization of α_1 and β-adrenoceptors in rabbit heart in relation to sympathetic nerve endings. *Eur J Pharmacol* **381:**135–140.
Eason MG and Liggett SB (1992) Subtype selective desensitization of alpha-2-adrenergic receptors. Different mechanisms control short and long-term agonist-promoted desensitization of alpha 2C10, alpha 2C4, and alpha 2C2. *J Biol Chem* **267:**25473–25479.
Eckly AE, Stoclet J-C and Lugnier C (1994) Isoprenaline induces endothelium-independent relaxation and accumulation of cyclic nucleotides in the rat aorta. *Eur J Pharmacol* **271:**237–240.
Edvinsson L and Owman C (1974) Pharmacological characterization of adrenergic alpha and beta receptors mediating the vasomotor responses of cerebral arteries in vitro. *Circ Res* **35:**835–849.
Egleme C, Godfraind T and Miller RC (1984) Enhanced responsiveness of isolated rat aorta to clonidine after removal of the endothelial cells. *Br J Pharmacol* **81**:16–18.
Emorine LJ, Marullo S, Briend-Sutren MM, Patey G, Tate K, Delavier-Klutchko C and Strosberg AD (1989) Molecular characterization of the human β_3-adrenergic receptor. *Science (Wash DC)* **245:**1118–1121.
Encabo A, Ferrer M, Salaices M, Manso R, Marín J and Balfagon G (1996) Effect of clenbuterol on the modulation of noradrenaline release in the rat tail artery. *J Auton Pharmacol* **16:**243–256.
Engel G, Hoyer D, Bertold R and Wagner H (1981) (±) (^{125}Iodo)cyanopindolol, a new ligand for β-adrenoceptors: identification and quantification of subclasses of β-adrenoceptors in guinea pig. *Naunyn-Schmiedeberg's Arch Pharmacol* **317:**277–285.
Engleman K and Portnoy B (1970) A sensitive double-isotope derivative assay for norepinephrine and epinephrine. *Circ Res* **26:**53–57.
Ennis C (1985) Comparison of the α_2-adrenoceptors which modulate noradrenaline release in rabbits and rat occipital cortex. *Br J Pharmacol* **85:**318P.
Fagura MS, Lydford SJ and Dougall IG (1997) Pharmacological classification of alpha 1-adrenoceptors mediating contractions of rabbit isolated ear artery: comparison with rat isolated thoracic aorta. *Br J Pharmacol* **120:**247–258.
Falcão-de-Freitas A (1996) Treatment of orthostatic hypotension with L-threo-DOPS in familial amyloidotic polyneuropathy. *Amyloid: Int J Exp Clin Invest* **3:**212–213.
Faure C, Pimoule C, Vallancien G, Langer SZ and Graham D (1994) Identification of α_1-adrenoceptor subtypes present in the human prostate. *Life Sci* **54:**1595–1605.
Ferro A, Queen LR, Priest RM, Xu B, Ritter JM, Poston L and Ward JPT (1999)

Activation of nitric oxide synthase by β_2-adrenoceptors in human umbilical vein endothelium in vitro. *Br J Pharmacol* **126:**1872–1880.
Fisher MH, Amend AM, Bach TJ, Barker JM, Brady EJ, Candelore MR, Carrol D, Cascieri MA, Chiu S-HL, Deng L, Forrest MJ, Hegarty-Friscino B, Guan X-M, Hom GJ, Hutchins JE, Kelly LJ, Mathvink RJ, Metzger JM, Miller RR, OK HO, Parmee ER, Saperstein R, Strader CD, Stearns RA, Thompson GM, Tota L, Vicario PP, Weber AE, Woods JW, Wyvratt MJ, Zafian PT and MacIntyre DE (1998) A selective human β_3-adrenergic receptor agonist increases metabolic rate in Rhesus monkeys. *J Clin Invest* **101:**2387–2393.
Flavahan NA, Cooke JP Shepherd JT and Vanhoutte PM (1987a) Human postjunctional alpha-1 and alpha-2 adrenocepors: differential distribution in arteries of the limbs. *J Pharmacol Exp Ther* **241:**361–365.
Flavahan NA, Flavahan S, Liu Q, Wu S, Tidmore W, Wiener CM, Spence RJ and Wigley FM (2000) Increased α_2-adrenergic constriction of isolated arterioles in diffuse scleroderma. *Arthritis Rheum* **43:**1886–1890.
Flavahan NA, Miller VM, Aarhus LL and Vanhoutte PM (1987b) Denervation augments alpha-2 but not alpha-1 adrenergic responses in canine saphenous veins. *J Pharmacol Exp Ther* **240:**589–593.
Flavahan NA, Rimele TJ, Cooke JP and Vanhoutte PM (1984) Characterization of *alpha*-1 and *alpha*-2 adrenoceptors activated by exogenous or nerve-released norepinephrine in the canine saphenous vein. *J Pharmacol Exp Ther* **230:**699–705.
Flavahan N and Vanhoutte PM (1986a) α_1-Adrenoceptor subclassification in vascular smooth muscle. *Trends Pharmacol Sci* **7:**347–349.
Flavahan N and Vanhoutte PM (1986b) Effect of cooling on *alpha*-1 and *alpha*-2 adrenergic responses in canine saphenous and femoral veins. *J Pharmacol Exp Ther* **238:**139–147.
Floras JS (1992) Epinephrine and the genesis of hypertension. *Hypertension* **19:**1–18.
Floras JS, Aylward PE, Victor RG, Mark AL and Abboud FM (1988) Epinephrine facilitates neurogenic vasoconstriction in humans. *J Clin Invest* **81:**1265–1274.
Foglar R, Shibata K, Hone K, Hirasawa A and Tsujimoto G (1995) Use of recombinant alpha 1-adrenoceptors to characterize subtype selectivity of drugs for the treatment of prostatic hypertrophy. *Eur J Pharmacol* **288:**201–207.
Folkow B (1982) Physiological aspects of primary hypertension. *Physiol Rev* **62:**347–504.
Ford APDW, Arredondo NF, Blue DR, Bonhaus DW, Jasper J, Kava MS, Lesnick J, Pfister JR, Shieh IA, Vimont RL, Williams JT, McNeal JE, Stamey TA and Clarke DE (1996) RS-17053 (*N*-(2-(cyclopropylmethoxy) ethyl)-5-chloro-α, α-dimethyl-1*H*-indole-3-ethanamine hydrochloride), a selective α_{1A}-adrenoceptor antagonist, displays low affinity for functional α_1-adrenoceptors in human prostate: implications for adrenoceptor classification. *Mol Pharmacol* **49:**209–215.
Ford APDW, Chang DJ, Clarke DE, Daniels DV, Eglen RM, Gever JR, Jasper JR, Kava MS, Lachnit WG, Lesnick JD, Melov TD, Stepan GT and Williams TJ (1998) *Alpha*-1A- versus *alpha*-1L-adrenoceptors: a pharmacological comparison. *Pharmacol Toxicol* **83:**12–15.
Ford APDW, Daniels DV, Chang DJ, Gever DJ, Jasper JR, Lesnick JD and Clarke DE (1997) Pharmacological pleiotropism of the human recombinant α_{1A}-adrenoceptor: implication for α_1-adrenoceptor classification. *Br J Pharmacol* **121:**1127–1135.
Ford APDW, Williams TJ, Blue DR and Clarke DE (1994) α_1-Adrenoceptor classification: sharpening Occam's razor. *Trends Pharmacol Sci* **15:**167–170.
Forrest MJ, Hom G, Bach T, Candelore MR, Cascieri MA, Strader C, Tota L, Fisher MH, Szumiloski J, Ok HO, Weber AE, Wyvratt M, Vicario P, Marko O, Deng L, Cioffe C, Hegarty-Friscino B and MacIntyre E (2000) L-750355, a human β_3-adrenoceptor agonist; in vitro pharmacology and profile of activity in vivo in the rhesus monkey. *Eur J Pharmacol* **407:**175–181.
Fouda A-K and Atkinson J (1986) Sensitivity to noradrenaline and electrical stimulation decreases with age in the rat tail artery. *Naunyn-Schmiedeberg's Arch Pharmacol* **344:**37–39.
Freedman RR, Baer RP and Mayes MD (1995) Blockade of vasospastic attacks by alpha2-adrenergic but not alpha1-adrenergic antagonist in idiopathic Raynaud's disease. *Circulation* **92:**1448–1451.
Freedman RR, Girgis R and Mayes MD (1999) Endothelial and adrenergic dysfunction in Raynaud's phenomenon and scleroderma. *J Rheumatol* **26:**2386–2388.
Freedman RR, Moten M, Migaly P and Mayes M (1993) Cold-induced potentiation of alpha 2-adrenergic vasoconstriction in primary Raynaud's disease. *Arthritis Rheum* **36:**685–690.
Freissmuth M, Waldhoer M, Bofill-Cardona E and Nanoff C (1999) G protein antagonists. *Trends Pharmacol Sci* **20:**237–245.
Frielle T, Collins S, Daniel KW, Caron MG, Lefkowitz RJ and Kobilka BK (1987) Cloning of the cDNA for the human beta 1-adrenergic receptor. *Proc Natl Acad Sci USA* **84:**7920–7924.
Fuder H, Muscholl E and Spemann R (1983) The determination of presynaptic pA_2 of yohimbine and phentolamine on the perfused rat heart under conditions of negligible autoinhibition. *Br J Pharmacol* **79:**109–119.
Fujisawa T, Ikegami H, Yamato E, Hamada Y, Kamide K, Rakugi H, Higaki J, Murakami H, Shimamoto K and Ogihara T (1997) Trp64Arg mutation of beta3 adrenergic receptor in essential hypertension: insulin resistance and the adrenergic system. *Am J Hypertens* **10:**101–105.
Furchgott RF (1967) The pharmacological differentiation of adrenergic receptors. *Ann N Y Acad Sci* **139:**553–570.
Furchgott RF (1972) The classification of adrenoceptors (adrenergic receptors). An evaluation from the standpoint of receptor theory, in *Catecholamines*, *Handbook of Experimental Pharmacology XXXIII* (Blaschko H and Muscholl E eds) pp 283–335, Springer, Berlin.
Furchgott RF and Zawadzki JV (1980) The obligatory role of endothelial cells in the relaxation of arterial smooth muscle by acetylcholine. *Nature (Lond)* **288:**373–376.
Furuta T, Ishikawa N and Shigei T (1986) Regional variation in β-adrenoceptor-mediated relaxation of canine veins. *Blood Vessels* **23:**236–245.
Galitzky J, Langin D, Montrastruc J-L, Lafontan M and Berlan M (1998) On the presence of a putative fourth β-adrenoceptor in human adipose tissue. *Trends Pharmacol Sci* **19:**164–165.
Gao Y, Tolsa JF, Botello M and Raj JU (1998) Developmental change in isoproterenol-mediated relaxation of pulmonary veins of fetal and newborn lambs. *J Appl Physiol* **84:**1535–1539.
García-Sáinz JA, Vázquez-Prado J and Medina LC (2000) α_1-Adrenoceptors function and phosphorylation. *Eur J Pharmacol* **389:**1–12.
Gardiner SM, Kemp PA and Bannet T (1991) Effects of N^G-nitro-L-arginine methyl ester on vasodilator responses to acetylcholine, 5′-N-ethylcarboxamidoadenosine or salbutamol in conscious rats. *Br J Pharmacol* **103:**1725–1732.
Gauthier C, Langin D and Balligand J-L (2000) β_3-Adrenoceptors in the cardiovascular system. *Trends Pharmacol Sci* **21:**426–431.
Gauthier C, Tavernier G, Charpentier F, Langin D and Lemarec H (1996) Functional beta (3)-adrenoceptor in the human heart. *J Clin Invest* **98:**556–562.
Gavin KT, Colgan M-P, Moore D, Shanik G and Docherty JR (1997) α_{2C}-Adrenoceptors mediate contractile responses to noradrenaline in human saphenous vein. *Naunyn-Schmiedeberg's Arch Pharmacol* **355:**406–411.
Giannuzzi CE, Seidler FJ and Slotkin TA (1995) Beta-adrenoceptor control of cardiac adenylyl cyclase during development: agonist pretreatment in the neonate uniquely causes sensitization, not desensitization. *Brain Res* **694:**271–278.
Gisbert R, Noguera MA, Ivorra MD and D'Ocon P (2000) Functional evidence of a constitutively active population of α_{1D}-adrenoceptors in rat aorta. *J Pharmacol Exp Ther* **295:**810–817.
Goldstein DS, Golczynska A, Stuhlmuller J, Holmes J, Rea RF, Grossman E and Lenders J (1999) A test of the "epinephrine hypothesis" in humans. *Hypertension* **33:**36–43.
Goldstein DS, Holmes C, Sheng-Ting L, Bruce S, Metman LV and Cannon RO (2000) Cardiac sympathetic denervation in Parkinson disease. *Ann Intern Med* **133:**338–347.
Gonçalves J, Proença J, Albuquerque AA and Paiva MQ (1989) Hypothermia induces supersensitivity of mouse vas deferens to adrenergic agonists: evidence for postjunctional $alpha_2$-adrenoceptors. *J Pharm Pharmacol* **41:**52–54.
Goodman OB, Krupnick JG, Santini F, Gurevich VV, Penn RB, Gagnon AW, Keen JH and Benovic JL (1996) β-Arrestin acts as a clathrin adaptor in endocytosis of the β-adrenergic receptor. *Nature (Lond)* **383:**447–450.
Göthert M and Hentrich F (1985) Identification of presynaptic β_2-adrenoceptors on the sympathetic nerve fibres of the human pulmonary artery. *Br J Pharmacol* **85:**933–941.
Grace GC, MacDonald PS and Dusting GJ (1988) Cyclic nucleotide interactions involved in endothelium-dependent dilatation in rat aortic rings. *Eur J Pharmacol* **148:**17–24.
Granneman JG and Lahners KN (1994) Analysis of human and rodent β_3-adrenergic receptor messenger ribonucleic-acids. *Endocrinology* **135:**1025–1031.
Grant TL, Mayers RM, Quayle SP, Briscoe MG, Howe R, Rao BS, Rao J, Rao G and Holloway BR (1994) Zeneca ZD2079 is a novel β_3-adrenoceptor agonist. *Br J Pharmacol* **112:**213P.
Gray DW and Marshall I (1992) Novel signal transduction pathway mediating endothelium-dependent β-adrenoceptor vasorelaxation in rat thoracic aorta. *Br J Pharmacol* **107:**684–690.
Gross G, Hanft G and Rugevics C (1988) 5-Methyl-urapidil discriminates between subtypes of the α_1-adrenoceptor. *Eur J Pharmacol* **151:**333–335.
Guarino RD, Perez DM and Piascik MT (1996) The α_1-adrenenergic receptors. *Cell Signal* **8:**323–333.
Guimarães S (1975) Further study of the adrenoceptors of the saphenous vein of the dog: influence of factors which interfere with the concentrations of agonists at the receptor level. *Eur J Pharmacol* **34:**9–19.
Guimarães S (1982) Two adrenergic biophases in blood vessels. *Trends Pharmacol Sci* **3:**159–161.
Guimarães S (1986) Postsynaptic α-adrenoceptors in blood vessels: discrepancies between results obtained in vivo and in vitro, in *New Aspects of the Role of Adrenoceptors in the Cardiovascular System* (Grobecker H, Philippu A, Starke K eds) pp 129–138, Springer-Verlag, Berlin.
Guimarães S (1999) Important recent advances in cardiovascular adrenergic mechanisms. *Ann Rev Biol Sci* **1:**31–58.
Guimarães S, Brandão F and Paiva MQ (1978) A study of the adrenoceptor-mediated feedback mechanisms by using adrenaline as a false transmitter. *Naunyn-Schmiedeberg's Arch Pharmacol* **305:**185–188.
Guimarães S, Figueiredo IV, Caramona MM, Moura D and Paiva MQ (1998) Prejunctional α_{2A}-autoreceptors in the human gastric and ileocolic arteries. *Naunyn-Schmiedeberg's Arch Pharmacol* **358:**207–211.
Guimarães S, Mota A and Begonha R (1993) The effectiveness of β-adrenoceptor stimulation and the contribution of β-adrenoceptors increase from the proximal to the distal part of the canine saphenous vein. *Naunyn-Schmiedeberg's Arch Pharmacol* **347:**596–600.
Guimarães S, Moura D, Paiva MQ and Vaz-da-Silva MJ (1994) Lack of postjunctional *beta* adrenoceptor-mediated effects in the canine saphenous vein at birth. *J Pharmacol Exp Ther* **268:**990–995.
Guimarães S and Nunes JP (1990) The effectiveness of α_2-adrenoceptor activation increases from the distal to the proximal part of the veins of the canine limbs. *Br J Pharmacol* **101:**387–393.
Guimarães S and Paiva MQ (1981a) Two distinct adrenoceptor-biophases in the vasculature: one for α- and the other for β-agonists. *Naunyn-Schmiedeberg's Arch Pharmacol* **316:**195–199.
Guimarães S and Paiva MQ (1981b) Two different biophases for adrenaline. *Naunyn-Schmiedeberg's Arch Pharmacol* **316:**200–204.
Guimarães S and Paiva MQ (1981c) Are β-agonists able to occupy β-adrenoceptors without causing effect? A study on the saphenous vein of the dog. Proceedings of the 4th Meeting on Adrenergic Mechanisms, pp 37, Porto, Portugal.
Guimarães S, Paiva MQ and Moura D (1982) Evidence for the existence of distinct biophases for α- and β-adrenoceptors in the vascular tissue. *J Cardiovasc Pharnacol* **4:**S58–S62.

Guimarães S, Paiva MQ and Moura D (1987) Alpha$_2$-adrenoceptor-mediated responses to so-called selective alpha$_1$-adrenoceptor agonists after partial blockade of alpha$_1$-adrenoceptors. *Naunyn-Schmiedeberg's Arch Pharmacol* **335:**397–402.
Guimarães S, Paiva MQ, Moura D and Vaz-da-Silva MJ (1991) Presynaptic α-autoadrenoceptors on peripheral noradrenergic neurones of newborn rabbits and dogs, in *Advances in the Biosciences Presynaptic Receptors and Neuronal Transporters* (Langer SZ, Galzin AM and Costentin J eds) vol 82, pp 11–14, Pergamon Press, Oxford.
Guimarães S, Paiva MQ and Pereira O (1997) α-Adrenoceptor-mediated prejunctional effects of chloroethylclonidine in the canine saphenous vein. *J Pharmacol Exp Ther* **282:**1326–1330.
Guimarães S, Paiva MQ and Polónia JJ (1983) Alpha$_1$- and alpha$_2$-adrenoceptors of the dog saphenous vein and their relation to the sympathetic nerve terminals. *Prog Neuropsychopharmacol Biol Psych* 155.
Gurdal H, Cai G and Johnson MD (1995a) α_1-Adrenoceptor responsiveness in aging aorta. *Eur J Pharmacol* **274:**117–123.
Gurdal H, Seasholtz TM, Wang HY, Brown RD, Johnson MD and Friedman E (1997) Role of G alpha q or G alpha o proteins in alpha 1-adrenoceptor subtype-mediated responses in Fischer 344 rat aorta. *Mol Pharmacol* **52:**1064–1070.
Gurdal H, Tilakaratne N, Brown RD, Fonseca M, Friedman E and Johnson MD (1995b) The expression of α_1-adrenoceptor subtypes changes with age in the rat aorta. *J Pharmacol Exp Ther* **275:**1656–1662.
Halotano A, Takahashi H, Tamaki M, Komeyama T, Koizumi T and Takeda M (1994) Pharmacological evidence of distinct α_1-adrenoceptor subtypes mediating the contraction of human prostatic urethra and peripheral artery. *Br J Pharmacol* **113:** 723–728.
Hamilton CA and Reid JL (1981) The effect of 6-hydroxydopamine on rabbit peripheral alpha adrenoceptors *in vivo* and *in vitro*. *Br J Pharmacol* **72:**119P.
Han C, Abel PW and Minneman KP (1987) Heterogeneity of α_1-adrenergic receptors revealed by chloroethylclonidine. *Mol Pharmacol* **32:**505–510.
Han C, Li J and Minneman KP (1990) Subtypes of α_1-adrenoceptors in rat blood vessels. *Eur J Pharmacol* **190:**97–104.
Hanft G and Gross G (1989) Subclassification of α_1-adrenoceptor recognition sites by urapidil derivatives and other selective antagonists. *Br J Pharmacol* **97:**691–700.
Happe HK, Bylund DB and Murrin LC (1999) Alpha$_2$ adrenergic receptor functional coupling to G proteins in rat brain during postnatal development. *J Pharmacol Exp Ther* **288:**1134–1142.
Harker CT, Bowman CJ, Taylor LM and Porter JM (1994) Cooling augments human saphenous vein reactivity to electrical stimulation. *J Cardiovasc Pharmacol* **23:** 453–457.
Harms HH, Zaagsma J and Van der Wal B (1974) β-Adrenoceptor studies. III. On the β-adrenoceptors in rat adipose tissue. *Eur J Pharmacol* **25:**87–91.
Heck DA and Bylund DB (1998) Differential down-regulation of alpha-2 adrenergic receptor subtypes. *Life Sci* **62:**1467–1472.
Hein L (1999) Clinical and molecular pharmacology of α_2-adrenergic receptors. Novel insight from transgenic animal models, in *Clinical Pharmacology. Prediction of Drug Actions from In Vitro Approaches. Fact or Fiction?* (Kuhlmann J, Roden DM eds) pp 25–29**,** W Zuckschwerdt Verlag, Munich.
Hein L (2001) Transgenic models of α_2-adrenergic receptor subtype function. *Rev Physiol Biochem Pharmacol* **142:**161–185.
Hein L, Altman JD and Kobilka BK (1999) Two functionally distinct α_2-adrenergic receptors regulate sympathetic neurotransmission. *Nature (Lond)* **402:**181–184.
Hein P, Goepel M, Cotecchia S and Michel MC (2001) A quantitative analysis of antagonism and inverse agonism at wild-type and constitutively active hamster α_{1B}-adrenoceptors. *Naunyn-Schmiedeberg's Arch Pharmacol* **363:**34–39.
Hein L and Kobilka BK (1995) Adrenergic receptor signal transduction and regulation. *Neuropharmacology* **34:**357–366.
Hein L, Spindler M, Altman JD and Kobilka BK (1998) Alpha-2-adrenoceptor subtypes: distribution and function. *Pharmacol Toxicol* **83:**26–28.
Hicks PE, Barras M, Herman G, Maudit P, Armstrong JM and Rossignol B (1991) α-Adrenoceptor subtypes in dog saphenous vein that mediate contraction and inositol phosphate formation. *Br J Pharmacol* **102:**151–161.
Hieble JP, Bylund DB, Clarke DE, Eikenburg DC, Langer SZ, Lefkowitz RJ, Minneman KP and Ruffolo BB (1995) International Union of Pharmacology. X. Recommendation for nomenclature of α_1-adrenoceptors: consensus update. *Pharmacol Rev* **47:**267–270.
Hieble JP, Ruffolo RR Jr and Starke K (1996) Identification, characterization and subclassification of α_2-adrenoceptors: an overview, in *α_2-Adrenergic Receptors: Structure, Function and Therapeutic Implications* (Lanier SM, Limbird LE eds) pp 1–18, Harwood, Amsterdam.
Hiraoka Y, Taniguchi T, Oshita M and Muramatsu I (2000) Pharmacological analysis of neurogenic, sympathetic mediated through alpha-1-, alpha-2-adrenergic and purinergic receptors in the dog saphenous vein. *Pharmacology* **60:**188–194.
Hirasawa A, Shibata K, Horie K, Takei Y, Obika K, Tanaka T, Muramoto N, Takagaki K, Yano J and Tsujimoto G (1995) Cloning, functional expression and tissue distribution of human α_{1C}-adrenoceptor splice variants. *FEBS Lett* **363:** 256–260.
Ho SL, Honner V and Docherty JR (1998) Investigation of the subtypes of α_2-adrenoceptor mediating prejunctional inhibition in rat atrium and cerebral cortex. *Naunyn-Schmiedeberg's Arch Pharmacol* **357:**634–639.
Hoefke W and Kobinger W (1966) Pharmakologische wirkungen des 2-(2,6-dichlorphenylamino)-2-imidazolin-hydrochloride, einer neuen, antihypertensiven substanz. *Arzneim-Forsch* **16:**1038–1050.
Hoffman BB and Lefkowitz RJ (1995) Catecholamines, sympathomimetic drugs and adrenergic receptor antagonists, in *Goodman & Gilman: The Pharmacological Basis of Therapeutics* (Hardman JG, Limbird LE, Molinoff PB, Ruddon RW and Gilman AG eds) 9th ed, pp 199–248, McGraw-Hill, New York.
Holck MI, Jones CHM and Haeusler G (1983) Differential interactions of clonidine and methoxamine with the postsynaptic α-adrenoceptor of rabbit main pulmonary artery. *J Cardiovasc Pharmacol* **5:**240-248.
Hrometz SL, Edelman SE, McCune DF, Olges JR, Hadley RW, Perez DM and Piascik MT (1999) Expression of multiple α_1-adrenoceptors on vascular smooth muscle; correlation with the regulation of contraction. *J Pharmacol Exp Ther* **290:**452–463.
Hussain MB and Marshall I (1997) Characterization of α_1-adrenoceptor subtypes mediating contractions to phenylephrine in rat thoracic aorta, mesenteric artery and pulmonary artery. *Br J Pharmacol* **122:**849–858.
Hussain MB and Marshall I (2000) α_1-Adrenoceptor subtypes mediating contractions of the rat mesenteric arteries. *Eur J Pharmacol* **395:**69–76.
Hutchinson DS, Evans BA and Summers RJ (2001) β_1-Adrenoceptors compensate for β_3-adrenoceptors in ileum from β_3-adrenoceptor knock-out mice. *Eur J Pharmacol* **132:**433–442.
Hyland L, Warnock P and Docherty JR (1987) Age-related alterations in α_1- and β-adrenoceptor-mediated responsiveness of rat aorta. *Naunyn-Schmiedeberg's Arch Pharmacol* **335:**50–53.
Ibarra M, López-Guerrero JJ and Villalobos-Molina R (1998) Further evidence for the predominance of α_{1D}-adrenoceptors in arteries of normotensive and spontaneously hypertensive rats. *Pharmacol Rev Comm* **10:**135–142.
Ibarra M, Pardo JP, López-Guerrero JJ and Villalobos-Molina R (2000) Differential response to chloroethylclonidine in blood vessels of normotensive and spontaneously hypertensive rats: role of α_{1D}- and α_{1A}-adrenoceptors in contraction. *Br J Pharmacol* **129:**653–660.
Ibarra M, Terrón JA, López-Guerrero JJ and Villalobos-Molina R (1997) Evidence for an age-dependent functional expression of α_{1D}-adrenoceptors in the rat vasculature. *Eur J Pharmacol* **322:**221–224.
Ignarro LJ, Buga GM, Wood KS, Byrns RE and Chaudhuri G (1987) Endothelium-derived relaxing factor (EDRF) produced and releases from artery and vein is nitric oxide. *Proc Natl Acad Sci USA* **84:**9265–9269.
Innes IR and Nickerson M (1970) Drugs acting on postganglionic adrenergic nerve endings and structures innervated by them (sympathomimetic drugs), in *Goodman & Gilman: The Pharmacological Basis of Therapeutics* (Goodman LS and Gilman A eds) 4th ed, pp 478–523; The MacMillan Co, London.
Insel PA (1996) Adrenergic receptors–evolving concepts and clinical implications. *N Engl J Med* **334:**580–585.
Iranami H, Hatano Y, Tsukiyama Y, Maeda H and Mizumoto K (1996) A beta-adrenoceptor agonist evokes a nitric oxide-cGMP relaxation mechanism modulated by adenylyl cyclase in rat aorta. *Anesthesiology* **85:**1129–1138.
Jeong SW and Ikeda SR (2000) Effect of G protein heterotrimer composition on coupling of neurotransmitter receptors to N-type Ca^{2+} channel modulation in sympathetic neurons. *Proc Natl Acad Sci USA* **97:**907–912.
Jones TR, Charette L, Garcia ML and Kaczorowski GJ (1993) Interaction of iberiotoxin with beta-adrenoceptor agonists and sodium nitroprusside on guinea pig trachea. *J Appl Physiol* **74:**1879–1884.
Kable JW, Murrin LC and Bylund DB (2000) In vivo gene modification elucidates subtype-specific functions of α_2-adrenergic receptors. *J Pharmacol Exp Ther* **293:** 1–7.
Kahaleh MB (1990) Vascular disease in scleroderma: endothelial T lymphocyte-fibroblast interactions. *Rheum Dis Clin North Am* **16:**53–73.
Kallio A, Scheinin M, Koulu M, Ponkilainen R, Ruskoaho H, Viinamäki O and Scheinin H (1989) Effects of dexmedetomidine, a selective α_2-adrenoceptor agonist, on hemodynamic control mechanisms. *Clin Pharmacol Ther* **46:**33–42.
Kalsner S (1982) The presynaptic receptor controversy. *Trends Pharmacol Sci* **3:**11–16.
Kalsner S and Westfall TC (1990) Presynaptic receptors and the question of autoregulation of transmitter release. *Ann NY Acad Sci* **604:**23–29.
Kamata K, Miyata N and Kasuya Y (1989) Involvement of endothelial cells in relaxation and contraction responses of the aorta to isoproterenol in naïve and streptozotocin-induced diabetic rats. *J Pharmacol Exp Ther* **249:**890–894.
Kanagy NL and Webb RC (1996) Increased responsiveness and decreased expression of proteins in deoxycorticosterone hypertension. *Hypertension* **27:**740–745.
Kaumann AJ (1997) Four β-adrenoceptor subtypes in mammalian heart. *Trends Pharmacol Sci* **18:**70–76.
Kaumann AJ, Engelhardt S, Hein L, Molenaar P and Lohse M (2001) Abolition of (−)-CGP 121777-evoked cardiostimulation in double β_1/β_2-adrenoceptor knockout mice. Obligatory role of β_1-adrenoceptors for putative β_4-adrenoceptor pharmacology. *Naunyn-Schmiedeberg's Arch Pharmacol* **363:**87–93.
Kaumann AJ, Hall JA, Murray KJ, Wells FC and Brown MJ (1989) A comparison of the effects of adrenaline and noradrenaline on human heart: the role of β_1-and β_2-adrenoceptors in the stimulation of adenylate cyclase and contractile force. *Eur Heart J* **10:**29–37.
Kaumann AJ and Molenaar P (1996) Differences between the third cardiac β-adrenoceptor and the colonic β_3-adrenoceptor in the rat. *Br J Pharmacol* **118:**2085–2098.
Kaumann AJ and Molenaar P (1997) Modulation of human cardiac function through 4β-adrenoceptor populations. *Naunyn-Schmiedeberg's Arch Pharmacol* **355:**667–681.
Kaumann AJ, Preitner F, Sarsero D, Molenaar P, Revelli J-P and Giacobino JP (1998) (−)-CGP 12177 causes cardiostimulation and binds to cardiac putative β_4-adrenoceptors in both wild-type and β_3-adrenoceptor knockout mice. *Mol Pharmacol* **53:**670–675.
Kava MS, Blue DR, Vimont RL, Clarke DE and Ford APDW (1998) α_{1L}-Adrenoceptor mediation for lower urinary tract tissues of man. *Br J Pharmacol* **123:**1359–1366.
Kenakin T (1997) Preface, in *Pharmacologic Analysis of Drug-Receptor Interaction* (Kenakin T ed) 3rd ed, pp xiii, Lippincott-Raven, Philadelphia.
Kenny BA, Chalmers DH, Philpott PC and Naylor AM (1995) Characterization of an α_{1D}-adrenoceptor mediating the contractile response of rat aorta to noradrenaline. *Br J Pharmacol* **115:**981–986.
Kompa AR and Summers RJ (2000) Desensitization and resensitization of β_1- and putative β_4-adrenoceptor mediated response occur in parallel in a rat model of cardiac failure. *Mol Pharmacol* **57:**252–258.
Kong JQ, Naylor DA and Fleming WW (1994) Functional distribution and role of

α_1-adrenoceptor subtypes in the mesenteric vasculature of the rat. *J Pharmacol Exp Ther* **268:**1153–1159.
Konishi M and Su C (1983) Role of endothelium in dilator response of spontaneously hypertensive rat arteries. *Hypertension* **5:**881–886.
Konkar AA, Zhai Y and Granneman JG (2000a) β_1-Adrenergic receptors mediate β_3-adrenergic-independent effects of Cgp 12177 in brown tissue. *Mol Pharmacol* **57:**252–258.
Konkar AA, Zhu Z and Granneman JG (2000b) Aryloxypropanolamine and catecholamine ligand interactions with the β_1-adrenergic receptor: evidence for interaction with distinct conformations of β_1-adrenergic receptors. *J Pharmacol Exp Ther* **294:**923–932.
Krauss SL, Dodge JT and Bevan JA (1992) Magnitude of β-adrenoceptor-mediated responses of dog epicardial arteries: inverse relation to diameter. *Am J Physiol* **263:**H1422–H1429.
Kukkonen JP, Renvaktar A, Shariatmadari R and Akerman Keo (1998) Ligand- and subtype-selective coupling of human alpha-2-adrenoceptors to Ca^{++} elevation in Chinese hamster ovary cells. *J Pharmacol Exp Ther* **287:**667–671.
Küng CF and Lüscher TF (1995) Different mechanisms of endothelial dysfunction with aging and hypertension in rat aorta. *Hypertension* **25:**194–200.
Kurose H and Lefkowitz RJ (1994) Differential desensitization and phosphorylation of three cloned and transfected α_2-adrenergic receptor subtypes. *J Biol Chem* **269:**10093–10099.
Lachnitt WG, Tran AM, Clarke DE and Ford APDW (1997) Pharmacological characterization of an α_{1A}-adrenoceptor mediating contractile responses to noradrenaline in isolated caudal artery of rat. *Br J Pharmacol* **120:**819–826.
Lands AM, Arnold A, McAuliff JP, Luduena FP and Brown TG Jr (1967a) Differentiation of receptor systems activated by sympathomimetic amines. *Nature (Lond)* **214:**597–598.
Lands AM, Luduena FP and Buzzo HJ (1967b) Differentiation of receptors responsive to isoproterenol. *Life Sci* **6:**2241–2249.
Langer SZ (1974) Presynaptic regulation of catecholamine release. *Biochem Pharmacol* **23:**1793–1800.
Langer SZ (1981) Presynaptic regulation of the release of catecholamines. *Pharmacol Rev* **32:**337–362.
Langer SZ (1997) 25 years since the discovery of presynaptic receptors: present knowledge and future perspectives. *Trends Pharmacol Sci* **18:**95–99.
Langer SZ, Massingham R and Shepperson NB (1981) Differential sensitivity to prazosin blockade of endogenously released and exogenously administered noradrenaline: possible relationship to the synaptic location of α_1- and the extrasynaptic location of α_2-adrenoceptors in dog vascular smooth muscle. *Br J Pharmacol* **72:**123P.
Langin D, Portillo MP, Saulnier-Blache J-S and Lafontan M (1991) Coexistence of three β-adrenoceptor subtypes in white fat cells of various mammalian species. *Eur J Pharmacol* **199:**291–301.
Latifpour J and Bylund DB (1983) Characterization of adrenergic receptor binding in rat lung: physiological regulation. *J Pharmacol Exper Ther* **224:**186–192.
Latifpour J, Jones SB and Bylund DB (1982) Characterization of (^{3}H) yohimbine binding to putative α_2-adrenergic receptors in neonatal rat lung. *J Pharmacol Exp Ther* **223:**606–611.
Lattimer N and Rhodes KF (1985) A difference in the affinity of some selective α_2-adrenoceptor antagonists when compared on isolated vasa deferentia of rat and rabbit. *Naunyn-Schmiedeberg's Arch Pharmacol* **329:**278–281.
Lattion A-L, Diviani D and Cotecchia S (1994) Truncation of the receptor carboxyl terminus impairs agonist-dependent phosphorylation and desensitization of the α_{1B}-adrenergic receptor. *J Biol Chem* **269:**22887–22893.
Lavoie JL, Trudeau F and Béliveau L (2000) Effect of blood flow and muscle contraction on noradrenaline spillover in the canine gracilis muscle. *Can J Physiol Pharmacol* **78:**75–80.
Lee E and Lee C (1997) Clinical comparison of selective and non-selective alpha1A-adrenoceptor antagonists in benign prostatic hyperplasia: studies on tamsulosin in a fixed dose and terazosin in increasing doses. *Br J Urol* **80:**606–611.
Lee TJ, Zhang W and Sarwinski S (2000) Presynaptic beta (2)-adrenoceptors mediate nicotine-induced Noergic neurogenic dilation in porcine basilar arteries. *Am J Physiol* **279:**H808–H816.
Leech CJ and Faber JE (1996) Different α-adrenoceptor subtypes mediate constriction of arterioles and venules. *Am J Physiol* **270:**H710–H722.
Lepor H, Tang R, Kobayashi S, Shapiro E, Forray C, Wetzel JM and Gluchowski C (1995) Localization of the α_{1A}-adrenoceptors in the human prostate. *J Urol* **154:** 2096–2099.
Lewis T (1929) Experiments relating to the peripheral mechanisms involved in spasmodic arrest of the circulation in fingers: a variety of Raynaud's disease. *Heart* **15:**7–101.
Li P, Zou AP, Al-Kayed NS, Rusch NJ and Harder DR (1994) Guanine nucleotide-binding proteins in aortic smooth muscle from hypertensive rats. *Hypertension* **23:**914–918.
Liggett SB, Freedman NJ, Schwinn DA and Lefkowitz RJ (1993) Structural basis for receptor subtype-specific regulation revealed by a chimeric β_3-/β_2-adrenergic receptor. *Proc Natl Acad Sci USA* **90:**3665–3669.
Limberger N, Mayer A, Zier G, Valenta B, Starke K and Singer EA (1989) Estimation of pA_2 values at presynaptic α_2-adrenoceptors in rabbit and rat brain cortex in the absence of autoinhibition. *Naunyn-Schmiedeberg's Arch Pharmacol* **340:**639–647.
Limberger N, Trendelenburg AU and Starke K (1992) Pharmacological characterization of presynaptic α_2-autoreceptors in rat submaxillary gland and heart atrium. *Br J Pharmacol* **107:**246–255.
Limbird LE (1988) Receptors linked to inhibition of adenylate cyclase: additional signaling mechanisms. *FASEB J* **2:**2686–2695.
Lindblad LE, Ekenvall L, Etzell BM and Bevegard S (1989) Adrenoceptors in Raynaud's disease. *J Cardiovasc Pharmacol* **14:**881–888.
Link RE, Daunt D, Barsh G, Chruscinski A and Kobilka B (1992) Cloning of two mouse genes encoding α_2-adrenergic receptor subtypes and identification of a single amino acid in the mouse α_2-C10 homolog responsible for an interspecies variation in antagonist binding. *Mol Pharmacol* **42:**16–27.
Link RE, Desai K, Hein L, Stevens ME, Chruscinski A, Bernstein D, Barsh GS and Kobilka B (1996) Cardiovascular regulation in mice lacking α_2-adrenergic receptor subtypes b and c. *Science (Wash DC)* **273:**803–805.
Link RE, Stevens ME, Kulatunga M, Scheinin M, Barsh G and Kobilka B (1995) Targeted inactivation of the gene encoding the mouse α_{2C}-adrenoceptor homolog. *Mol Pharmacol* **48:**48–55.
Lomasney JW, Cotecchia S, Lorenz W, Leung W-Y, Schwinn DA, Yang-Feng TL, Brownstein M, Lefkowitz RJ and Caron M (1991) Molecular cloning and expression of the cDNA for the α_{1A}-adrenergic receptor. *J Biol Chem* **266:**6365–6369.
Lomasney JW, Lorenz W, Allen LF, King K, Regan JW, Yang-Feng TL, Caron MG and Lefkowitz RJ (1990) Expansion of the α_2-adrenergic receptor family: cloning and expression of a human α_2-adrenergic receptor subtype, the gene for which is located on chromosome 2. *Proc Natl Acad Sci USA* **87:**5094–5098.
Lönnqvist F, Thöme A, Nilsell K, Hoffstedt J and Arner P (1995) A pathogenic role of visceral fat of β_3-adrenoceptors in obesity. *J Clin Invest* **95:**1109–1116.
Low AM, Lu-Chao H, Wang YF, Brown RD, Kwan CY and Daniel EE (1998) Pharmacological and immunocytochemical characterization of subtypes of alpha-1 adrenoceptors in dog aorta. *J Pharmacol Exp Ther* **285:**894–901.
Lowell BB and Flier JS (1995) The potential significance of β_3-adrenergic receptor. *J Clin Invest* **95:**923.
Luttrell LM, Ferguson SSG, Daaka Y, Miller WE, Maudsley S, Della Rocca GJ, Tin F-T, Kawakatsu H, Owada K, Luttrell DK, Caron MG and Lefkowitz RJ (1999) β-Arrestin-dependent formation of β_2-adrenergic receptor-Src protein kinase complexes. *Science (Wash DC)* **283:**655–661.
MacDonald A, McLean M, MacAuly L and Shaw AM (1999) Effects of propranolol and L-NAME on beta-adrenoceptor-mediated relaxation in rat carotid artery. *J Auton Pharmacol* **19:**145–149.
MacKenzie JF, Daly CJ, Pediani JD and McGrath JC (2000) Quantitative imaging in live human cells reveals intracellular α_1-adrenoceptor ligand binding sites. *J Pharmacol Exp Ther* **294:**434–443.
MacLennan SJ, Luong LA, Jasper JR, To ZP and Egler RM (1997) Characterization of α_2-adrenoceptors mediating contraction of dog saphenous vein: identity with the human α_{2A} subtype. *Br J Pharmacol* **121:**1721–1729.
MacMillan LB, Hein L, Smith MS, Piascik ML and Limbird LE (1996) Central hypotensive effects of the α_{2a}-adrenergic receptor subtype. *Science (Wash DC)* **273:**801–803.
MacNulty EE, McClue SJ, Carr IC, Jess T, Wakelam MJO and Milligam G (1992) α_2-C10 adrenergic receptors expressed in rat-1 fibroblast can regulate both adenylylcyclase and phospholipase D-mediated hydrolysis of phosphatidylcholine by interacting with pertussis toxin-sensitive guanine nucleotide-binding proteins. *J Biol Chem* **267:**2149–2156.
Magalhaes M, Wenning GK, Daniel SE and Quinn NP (1995) Autonomic dysfunction in pathologically confirmed multiple-system atrophy and idiopathic Parkinson's disease—a retrospective comparison. *Acta Neurol Scand* **91:**98–102.
Majewski H (1983) Modulation of noradrenaline release through activation of presynaptic β-adrenoceptors. *J Auton Pharmacol* **3:**47–60.
Majewski H, Hedler L and Starke K (1982) The noradrenaline release rate in the anaesthetized rabbit: facilitation by adrenaline. *Naunyn-Schmiedeberg's Arch Pharmacol* **321:**20–27.
Majewski H, Rand MJ and Tung L-H (1981a) Activation of prejunctional β-adrenoceptors in rat atria by adrenaline applied exogenously or released as a co-transmitter. *Br J Pharmacol* **73:**669–679.
Majewski H, Tung L-H and Rand MJ (1981b) Hypertension through adrenaline activation of prejunctional β-adrenoceptors. *Clin Exp Pharmacol Physiol* **8:**463–468.
Makaritsis KP, Handy DE, Johns C, Kobilka B, Gravas I and Gravas H (1999) Role of alpha 2B-adrenergic receptors in the development of salt-induced hypertension. *Hypertension* **33:**14–17.
Malik KU and Muscholl E (1969) Effect of sympathomimetic amines on the response of the perfused mesenteric artery preparation to adrenergic nerve stimulation. *Arzneim-Forsch* **19:**1574–1579.
Malinowska B and Schlicker E (1996) Mediation of the positive chronotropic effect of CGP 12177 and cyanopindolol in the pithed rat by atypical β-adrenoceptors, different from β_3-adrenoceptors. *Br J Pharmacol* **117:**943–949.
Man in't Veld AJ, Boomsma F, Moleman P and Schalekamp MADH (1987) Congenital dopamine-betahydroxylase deficiency, a novel orthostatic syndrome. *Lancet* **i:**183–188.
Manara L, Croci T and Landi M (1995) Beta$_3$-adrenoceptors and intestinal motility. *Fundam Clin Pharmacol* **9:**332–342.
Manara L, Badone D, Baroni M, Boccardi G, Cecchi R, Tiziano C, Guidice A, Guzzi U, Landi M and Le Fur G (1996) Functional identification of rat atypical β-adrenoceptor by the first β_3-selective antagonists, aryloxypropanolaminotetratine. *Br J Pharmacol* **117:**1374–1376.
Maurice DH and Haslam RJ (1990) Nitroprusside enhances isoprenaline-induced increases in cAMP in rat aortic smooth muscle. *Eur J Pharmacol* **191:**471–475.
McAdams RP and Waterfall JF (1986) The effect of age on the sensitivity of pre- and postsynaptic alpha-adrenoceptors to agonists and antagonists in the rat. *Naunyn-Schmiedeberg's Arch Pharmacol* **334:**430–435.
McCafferty GP, Nadelsky DP and Hieble JP (1999) Characterization of postjunctional alpha-adrenoceptors in the pithed mouse. *Gen Pharmacol* **33:**99–105.
McCulloch MW, Bevan JA and Su C (1975) Effects of phenoxybenzamine and norepinephrine on transmitter release in the pulmonary artery of the rabbit. *Blood Vessels* **12:**122–133.
McGrath JC (1982) Evidence for more than one type of postjunctional α-adrenoceptor. *Biochem Pharmacol* **31:**467–484.
McGrath JC, Lepor H and Wyllie MG (1996) Report of a unique meeting between the alpha-blocker subcommittee and the pharmacological industry. *Urology* **48:**665–667.
Michel AD, Loury D and Whiting RL (1989) Differences between the α_2-adrenoceptor

in rat submaxillary gland and the α_{2A}- and α_{2B}-adrenoceptor subtypes. *Br J Pharmacol* **98:**890–897.
Michel MC, Grübbel B, Taguchi K, Verfürth F, Otto T and Kröpfl D (1996) Drugs for treatment of benign prostatic hyperplasia: affinity comparison at cloned α_1-adrenoceptor subtypes and in human prostate. *J Auton Pharmacol* **16:**21–28.
Michel MC, Kenny B and Schwinn DA (1995) Classification of the α_1-adrenoceptor subtypes. *Naunyn-Schmiedeberg's Arch Pharmacol* **352:**1–10.
Michel MC, Mehlburger L, Bressel H-U, Schumacher H, Schäfers RF and Goepel M (1998a) Tamsulosin treatment of 19,395 patients with lower urinary tract symptoms: does co-morbidity alter tolerability? *J Urol* **160:**784–791.
Michel MC, Taguchi K, Schäfers RF, Williams TJ, Clarke DE and Ford APDW (1998b) α_1-Adrenoceptor subtypes in the human cardiovascular and urogenital systems. *Adv Pharmacol* **42:**394–398.
Miller RJ (1998) Presynaptic receptors. *Ann Rev Pharmacol Toxicol* **38:**201–227.
Miller JW, Hu ZW, Okazaki M, Fujinaga M and Hoffman BB (1996) Expression of alpha 1 adrenergic receptor subtype mRNAs in the rat cardiovascular system with aging. *Mech Ageing Dev* **87:**75–89.
Miller VM and Vanhoutte PM (1985) Endothelial α_2-adrenoceptors in canine pulmonary and systemic blood vessels. *Eur J Pharmacol* **118:**123–129.
Milton AS, Carr GA, Luby CD, Scarlett JA, White R and Callingham BA (1999) Changes in reactivity of the digital artery of the fallow deer, *Dama dama*, in summer and winter. *J Therm Biol* **24:**465–470.
Minneman KP (1988) α_1-Adrenergic receptor subtypes, inositol phosphates and sources of cell calcium. *Pharmacol. Rev* **40:**87–119.
Misu Y and Kubo T (1986) Presynaptic β-adrenoceptors. *Med Res Rev* **6:**197–225.
Misu Y, Kuwahara M, Amano H and Kubo T (1989) Evidence for tonic activation of prejunctional β-adrenoceptors in guinea-pig pulmonary arteries by adrenaline derived from the adrenal medulla. *Br J Pharmacol* **98:**45–50.
Misu Y, Kuwahara M, Goshima Y and Kubo T (1990) Stress-induced increases in adrenaline further activate tonically presynaptic β-adrenoceptors in renal arteries from prehypertensive rats of spontaneously hypertensive strain. *Arch Int Pharmacodyn* **305:**100–110.
Miura M, Belvisi MG, Stritton CD, Yacoub MH and Barnes PJ (1992) Role of potassium channels in bronchodilator responses in human airways. *Am Rev Respir Dis* **146:**132–136.
Mohell N and Dicker A (1989) The β-adrenergic radioligand (^{3}H)-CGP-12177, generally classified as an antagonist, is a thermogenic agonist in brown tissue. *Biochem J* **261:**401–405.
Molderings GJ and Göthert M (1993) Pharmacological characterization of the inhibitory α_2-autoreceptors on the sympathetic nerves of human, rat and rabbit blood vessels. *Fundam Clin Pharmacol* **7:**372.
Molenaar P, Malta E, Jones CR, Buxton BR and Summers RJ (1988) Autoradiographic localization and function of β-adrenoceptors on the human internal mammary artery and saphenous vein. *Br J Pharmacol* **95:**225–233.
Moncada S, Palmer RMJ and Higgs EA (1991a) Nitric oxide: physiology, pathophysiology, and pharmacology. *Pharmacol Rev* **43:**109–142.
Moncada S, Rees DD, Schulz R and Palmer RMJ (1991b) Development and mechanism of a specific supersensitivity to nitrovasodilators after inhibition of vascular nitric oxide synthesis in vivo. *Proc Natl Acad Sci USA* **88:**2166–2170.
Morris AD, Petrie JR and Connel JMC (1994) Insulin and hypertension. *J Hypertens* **12:**633–642.
Morrow AL and Creese I (1986) Characterization of α_1-adrenergic receptor subtypes in rat brain: a reevaluation of ^{3}H-WB 4101 and ^{3}H-prazosin binding. *Mol Pharmacol* **29:**321–333.
Motomura S, Zerkowski H-R, Daul A and Brodde O-E (1990) On the physiologic role of beta-2 adrenoceptors in the human heart: in vitro and in vivo studies. *Am Heart J* **119:**608–619.
Moura D, Paiva MQ, Vaz-da-Silva MJ and Guimarães S (1997) Ontogenesis of beta-adrenoceptors and the adrenaline content of the adrenals. *Pharmacol Toxicol* **81:**14.
Moura D, Vaz-da-Silva MJ, Azevedo I, Brandão F and Guimarães S (1993) Release and disposition of ^{3}H-noradrenaline in saphenous vein of neonate and adult dogs. *Naunyn-Schmiedeberg's Arch Pharmacol* **347:**186–191.
Müller-Schweinitzer E (1984) Alpha-adrenoceptors, 5-hydroxytryptamine receptors and the action of dihydroergotamine in human venous preparations obtained during saphenectomy procedures for varicose veins. *Naunyn-Schmiedeberg's Arch Pharmacol* **327:**299–303.
Muramatsu I, Murata S, Isaka M, Piao HL, Zhu J, Suzuki F, Miyamoto S, Oshita M, Watwnabe Y and Taniguchi T (1998) Alpha 1-adrenoceptor subtypes and two receptor systems in vascular tissues. *Life Sci* **62:**1461–1465.
Muramatsu I, Ohmura T, Kigoshi S, Hashimoto S and Oshita M (1990) Pharmacological subclassification of α_1-adrenoceptors in vascular smooth muscle. *Br J Pharmacol* **99:**197–201.
Murohara T, Yasue H, Ohgushi M, Sakaino N and Jougasaki M (1991) Age related attenuation of the endothelium dependent relaxation to noradrenaline in isolated pig coronary arteries. *Cardiovasc Res* **25:**1002–1009.
Murphy TJ and Bylund DB (1988) Characterization of alpha-2 adrenergic receptors in OK cell, an opossum kidney cell line. *J Pharmacol Exp Ther* **244:**571–578.
Nahorski SR, Barnett DB and Cheung Y-D (1985) α-Adrenoceptor-effector coupling: affinity states or heterogeneity of the α-adrenoceptor? *Clin Sci* **68:**39s–42s.
Nedergaard OA and Abrahamsen J (1990) Modulation of noradrenaline release by activation of presynaptic β-adrenoceptors in the cardiovascular system. *Ann NY Acad Sci* **604:**528–554.
Netten PM, de Vos K, Horstink MW and Hoefnagels WH (1995) Autonomic dysfunction in Parkinson's disease, tested with a computerized method using a Finapres device. *Clin Auton Res* **5:**85–89.
Nisoli E, Tonello C, Landi M and Carruba MO (1996) Functional studies on the first selective β_3-adrenergic receptor antagonist SR 59,230A. *Mol Pharmacol* **49:**7–14.
Nunes JP (1995) Central α_2-adrenoceptors and blood pressure regulation in the rat. *Eur J Pharmacol* **278:**183–185.
Nunes JP and Guimarães S (1993) Chloroethylclonidine irreversibly activates postjunctional alpha$_2$-adrenoceptors in the dog saphenous vein. *Naunyn-Schmiedeberg's Arch Pharmacol* **348:**264–268.
Nunes JP, Moura D, Guimarães S and De la Lande IS (1991) Postsynaptic α-adrenoceptors in the perfused canine saphenous vein in vitro. *Pharmacol Res* **23:**409–414.
O'Donnell SR and Wanstall JC (1984) *Beta*-1 and *beta*-2 adrenoceptor-mediated responses in preparations of pulmonary artery and aorta from young and aged rats. *J Pharmacol Exp Ther* **228:**733–738
O'Donnell SR and Wanstall JC (1985) Responses to the β_2-selective agonist procaterol of vascular and atrial preparations with different functional β-adrenoceptor populations. *Br J Pharmacol* **84**:227–235.
O'Donnell SR and Wanstall JC (1986) Thyroxine treatment of aged or young rats demonstrates that vascular responses mediated by β-adrenoceptors subtypes can be differentially regulated. *Br J Pharmacol* **88:**41–49.
Ohmi K, Shinoura H, Nakayama Y, Goda N and Tsujimoto G (1999) Characterization of alpha 1-adrenoceptors expressed in a novel vascular smooth muscle cell line cloned from p53 knockout mice, P53LMAC01 (AC01) cells. *Br J Pharmacol* **127:**756–762.
Oostendorp J and Kaumann AJ (2000) Pertussis toxin supresses carbachol-evoked cardiodepression but does not modify cardiostimulation in mouse left atria: no evidence for β_2- and β_3-adrenergic functions. *Naunyn-Schmiedeberg's Arch Pharmacol* **361:**134–145.
Oriowo MA (1994) Atypical beta-adrenoceptors in the rat isolated common carotid artery. *Br J Pharmacol* **113:**699–702.
Oriowo MA (1995) Different atypical beta-adrenoceptors mediate isoprenaline-induced relaxation in vascular and non-vascular smooth muscles. *Life Sci* **56:**PL269–PL275.
Oriowo MA, Hieble JP and Ruffolo RR Jr (1991) Evidence for heterogeneity of prejunctional alpha-2-adrenoceptors. *Pharmacology* **43:**1–13.
Osswald W and Guimarães S (1983) Adrenergic mechanisms in blood vessels: morphological and pharmacological aspects. *Rev Physiol Biochem Pharmacol* **96:**53–122.
Paiva MQ, Mota A, Moura D and Guimarães S (1997) Prejunctional α_{2A}-autoreceptors in the canine saphenous vein. *Naunyn-Schmiedeberg's Arch Pharmacol* **356:**368–373.
Paiva MQ, Morato M, Moura D and Guimarães S (1999) A comparative study of postsynaptic α_2-adrenoceptors of the dog mesenteric and rat femoral veins. *Naunyn-Schmiedeberg's Arch Pharmacol* **360:**165–170.
Paiva MQ, Moura D, Vaz-da-Silva MJ and Guimarães S (1994) Postnatal development of vascular β-adrenoceptor-mediated responses and the increase in the adrenaline content of the adrenal gland have a parallel course. *Naunyn-Schmiedeberg's Arch Pharmacol* **350:**28–33.
Palmer RMJ, Ferrige AG and Moncada S (1987) Nitric oxide release accounts for the biological activity of endothelium-derived relaxing factor. *Nature (Lond)* **327:**524–525.
Parent R, Al-Obaidi M and Lavallée M (1993) Nitric oxide formation contributes to β-adrenergic dilation of resistance coronary vessels in conscious dogs. *Circ Res* **73:**241–251.
Pegram BL, Bevan RD and Bevan JA (1976) Facial vein of the rabbit: neurogenic vasodilation mediated by β-adrenergic receptors. *Circ Res* **39:**854–860.
Pereira O, Moura D, Nunes P, Vaz-da-Silva MJ and Guimarães S (1991) Involvement of α_1- and α_2-adrenoceptors in the responses of proximal and distal segments of the canine saphenous vein to exogenous and endogenous noradrenaline. *Naunyn-Schmiedeberg's Arch Pharmacol* **343:**616–622.
Perez DM, Piascik MT and Graham RM (1991) Solution-phase library screening for the identification of rare clones: isolation of an α_{1D}-adrenergic receptor cDNA. *Mol Pharmacol* **40:**876–883.
Piascik MT, Guarino RD, Smith MS, Soltis EE, Saussy DL and Perez DM (1995) The specific contribution of the novel alpha-1D adrenoceptor to the contraction of vascular smooth muscle. *J Pharmacol Exp Ther* **275:**1583–1589.
Piascik MT, Hrometz SL, Edelman SE, Guarino RD, Hadley RW and Brown RD (1997) Immunocytochemical localization of the *alpha*-1B adrenergic receptor and the contribution of this and the other subtypes to vascular smooth muscle contraction: analysis with selective ligands and antisense oligonucleotides. *J Pharmacol Exp Ther* **283:**854–868.
Piascik MT, Kusiak JW and Barron KW (1990) α_1-Adrenoceptor subtypes and the regulation of peripheral hemodynamics in the conscious rat. *Eur J Pharmacol* **186:**273–278.
Polónia J, Paiva MQ and Guimarães S (1985) Pharmacological characterization of postsynaptic α-adrenoceptor subtypes in five different dog arteries in vitro. *J Pharm Pharmacol* **37:**205–208.
Powell CE and Slater IH (1958) Blocking of inhibitory adrenergic receptors by a dichloro analog of isoproterenol. *J Pharmacol Exp Ther* **122:**480–488.
Preitner F, Muzzin P, Revelli J-P, Seydoux J, Galitzky J, Berlan M, Lafontan M and Giacobino J-P (1998) Metabolic response to various β-adrenoceptor agonists in β_3-adrenoceptor knockout mice: evidence for a new β-adrenergic receptor in brown adipose tissue. *Br J Pharmacol* **124:**1684–1688.
Price DT, Schwinn DA, Lomasney JW, Allen LF, Caron MG and Lefkowitz RJ (1993) Identification, quantification and localization of mRNA for three distinct alpha$_1$ adrenergic receptor subtypes. *J Urol* **150:**546–551.
Pulito VL, Li X, Varga SS, Mulcahy LS, Clark KS, Halbert SA, Reitz AB, Murray WV and Jolliffe LK (2000) An investigation of the uroselective properties of four novel α_{1a}-adrenergic receptor subtype-selective antagonists. *J Pharmacol Exp Ther* **294:**224–229.
Rea RF, Biaggioni I, Robertson RM, Haile V and Robertson D (1990) Reflex control of sympathetic nerve activity in dopamine-β-hydroxylase deficiency. *Hypertension* **156:**107–112.
Rebich S, Devine JO and Armstead WM (1995) Role of nitric oxide and cAMP in β-adrenoceptor-induced pial artery vasodilation. *Am J Physiol* **268:**H1071–H1076.
Regan JW, Kobilka TS, Yang-Feng TL, Caron MG, Lefkowitz JR and Kobilka BK

(1988) Cloning and expression of a human kidney cDNA for an α_2-adrenergic receptor subtype. *Proc Natl Acad Sci USA* **85:**6301–6305.
Revelli JP, Preitner F, Samec S, Muniesa P, Kuehne F, Boss O, Vassalli JD, Dulloo A, Seydoux J, Giacobino JP, Huarte J and Ody C (1997) Targeted gene disruption reveals a leptin-independent role for the mouse β_3-adrenoceptor in the regulation of body composition. *J Clin Invest* **100:**1098–1106.
Richard V, Tanner FC, Tschudi M and Luscher TF (1990) Different activation of L-arginine pathway by bradykinin, serotonin and clonidine in coronary arteries. *Am J Physiol* **259:**H1433–H1439.
Roberts SJ, Molennar S and Summers RJ (1993) Characterization of propranolol-resistant (−) (^{125}I)-cyanopindolol binding sites in rat soleus muscle. *Br J Pharmacol* **109:**344–352.
Rohrer DK, Bernstein D, Chruscinski A, Desai KH, Schauble E and Kobilka BK (1998a) The developmental and physiological consequences of disrupting genes encoding β_1 and β_2 adrenoceptors. *Adv Pharmacol* **42:**499–501.
Rohrer DK, Chruscinski A, Schauble EH, Bernstein D and Kobilka BK (1999) Cardiovascular and metabolic alterations in mice lacking both β1 and β2-adrenergic receptors. *J Biol Chem* **274:**16701–16708.
Rohrer DK, Desai KH, Jasper JR, Stevens ME, Regula DJ, Barsh GS, Bernstein D and Kobilka BK (1996) Targeted disruption of the mouse beta 1-adrenergic receptor gene: developmental and cardiovascular effects. *Proc Natl Acad Sci USA* **93:**7375–7380.
Rohrer DK and Kobilka BK (1998) Insights from in vivo modification of adrenergic receptor gene expression. *Ann Rev Pharmacol Toxicol* **38:**351–373.
Rohrer DK, Schauble EH, Desai KH, Kobilka BK and Bernstein D (1998b) Alterations in dynamic heart rate control in the β_1-adrenergic receptor knockout mouse. *Am J Physiol* **274:**H1184–H1193.
Rosenbaum M, Malbon CC, Hirsch J and Leibel RL (1993) Lack of β_3-adrenergic effect on lipolysis in human subcutaneous adipose tissue. *J Clin Endocrinol Metab* **77:**352–355.
Ross G (1976) Adrenergic responses of the coronary vessels. *Circ Res* **39:**461–465.
Rudner XL, Berkowitz DE, Booth JV, Funk BL, Cozart KL, D'Amico EB, El-Moalem H, Page SO, Richardson CD, Winters B, Marucci L and Schwinn DA (1999) Subtype specific regulation of human vascular α_1-adrenergic receptors by vessel bed and age. *Circulation* **100:**2336–23443.
Ruffolo RR Jr and Hieble JP (1999) Adrenoceptor pharmacology: urogenital applications. *Eur Urol* **36:**17–22.
Ruffolo RR, Nichols AJ, Stadel JM and Hieble JP (1991) Structure and function of α_2-adrenoceptors. *Pharmacol Rev* **43:**475–501.
Ruffolo RR, Nichols AJ, Stadel JM and Hieble JP (1993) Pharmacologic and therapeutic application of α_2-adrenoceptor subtypes. *Ann Rev Pharmacol Toxicol* **33:** 243–279.
Rump CL, Bohmann C, Schauble U, Schöllhorn J and Limberger N (1995) α_{2C}-Adrenoceptor-modulated release of noradrenaline in human right atrium. *Br J Pharmacol* **116:**2617–2624.
Rump CL, Schuster MJ and Schollmeyer P (1992) Activation of β_2-adrenoceptors by isoprenaline and adrenaline enhances noradrenaline release in cortical kidney slices of young spontaneously hypertensive rats. *Naunyn-Schmiedeberg's Arch Pharmacol* **345:**25–32.
Russel MP and Moran NC (1980) Evidence for lack of innervation of β_2-adrenoceptors in the blood vessels of the gracilis muscle of the dog. *Circ Res* **46:**344–352.
Satake N, Shibata M and Shibata S (1996) The inhibitory effects of iberiotoxin and 4-aminopyridine on the relaxation induced by β_1- and β_2-adrenoceptor activation in rat aortic rings. *Br J Pharmacol* **119:**505–510.
Sakate N, Shibata M and Shibata S (1997) Endothelium- and cytochrome P-450-dependent relaxation induced by isoprenaline in rat aortic rings. *Eur J Pharmacol* **319:**37–41.
Satake N, Zhou Q, Morikawa M, Inoue M and Shibata S (1995) Potentiating effect of nicorandil, an antianginal agent, on relaxation induced in isolated rat aorta: involvement of cGMP-inhibitable cAMP phosphodiesterase. *J Cardiovasc Pharmacol* **25:**489–494.
Satoh M, Enomoto K, Niwano H, Fujimura H, Toyama Y, Takayanagi I and Koike K (1998) Regional differences in α_1-adrenoceptor subtypes in rabbit arteries. *Eur J Pharmacol* **350:**67–73.
Satoh M, Enomoto K, Takayanagi I and Koike K (1999) Analysis of α_1-adrenoceptor subtypes in rabbit aorta and arteries: regional difference and co-existence. *Eur J Pharmacol* **374:**229–240.
Satoh M, Kanai S, Ashikawa N and Takayanagi I (1995) Age-related alteration of α_1-adrenoceptor mechanisms and inhibitory effects of isradipine. *Arch Int Pharmacodyn Ther* **330:**190–205.
Saussy DL, Goetz AS, Queen KL, King HK, Lutz MW and Rimele TJ (1996) Structure activity relationships of a series of buspirone analogs at α_1-adrenoceptors: further evidence that rat aorta α_1-adrenoceptors are of the α_{1D}-subtype. *J Pharmacol Exp Ther* **278:**136–144.
Sayet I, Neuilly G, Rakotoarisoa L, Mironneau J and Mironneau C (1993) Rat vena cava α_{1B}-adrenoceptors: characterization by (^{3}H)-prazosin binding and contraction experiments. *Eur J Pharmacol* **246:**275–281.
Schäfers RF, Goepel M and Michel MC (1998) Clinical relevance of α_1-adrenoceptor subtypes. *Pharmacol Toxicol* **83:**23–25.
Schäfers RF, Poller U, Pönicke K, Geissler M, Daul AE, Michel MC and Brodde O-E (1997) Influence of adrenoceptor and muscarinic receptor blockade on the cardiovascular effects of exogenous noradrenaline and of endogenous noradrenaline released by infused tyramine. *Naunyn-Schmiedeberg's Arch Pharmacol* **355:**239–249.
Schäfers RF, Nürnberger J, Herrmann B, Wenzel RR, Philipp T and Michel MC (1999) Adrenoceptors mediating the cardiovascular and metabolic effects of α-methylnoradrenaline in humans. *J Pharmacol Exp Ther* **289:**918–925.
Schwietert HR, Mathy MJ, Wilhelm D, Willfert B, Pfaffendorf M and Van Zwieten PA (1992) α_1-Adrenoceptor-mediated Ca^{2+}-entry from the extracellular fluid and Ca^2-release from intracellular stores: no role for $\alpha_{1A,\ B}$-adrenoceptor subtypes in the pithed rat. *J Auton Pharmacol* **12:**125–136.
Schwinn DA and Kwatra MM (1998) Expression and regulation of α_1-adrenergic receptors in human tissues. *Adv Pharmacol* **42:**390–394.
Schwinn DA, Lomasney JW, Lorenz W, Szklut PJ, Fremeau RT, Yang-Feng TL, Caron MG, Lefkowitz RJ and Cotecchia S (1990) Molecular cloning and expression of the cDNA for a novel α_1-adrenergic receptor subtype. *J Biol Chem* **265:**8183–8189.
Shafiei M and Mahmoudian M (1999) Atypical β-adrenoceptors of rat thoracic aorta. *Gen Pharmacol* **32**:557–562.
Shen Y-T, Cervoni P, Claus T and Vatner SF (1996) Differences in β_3-adrenergic receptor cardiovascular regulation in conscious primates, rats and dogs. *J Pharmacol Exp Ther* **278:**1435–1443.
Shen Y-T and Claus TH (1993) Potential mechanisms of β_3-adrenoceptor-induced peripheral vasodilation in conscious dogs. *Clin Res* **41:**348A.
Shen Y-T, Zhang H and Vatner SF (1994) Peripheral vascular effects of *beta*-3 adrenergic receptor stimulation in conscious dogs. *J Pharmacol Exp Ther* **268:** 466–473.
Shi AG, Kwan CY and Daniel EE (1989) Relation between density (maximum binding) and *alpha* adrenoceptor binding sites and contractile response in four canine vascular tissues. *J Pharmacol Exp Ther* **250:**1119–1124.
Shoji T, Tsuru H and Shigei T (1983) A regional difference in the distribution of postsynaptic alpha-adrenoceptor subtypes in canine veins. *Naunyn-Schmiedeberg's Arch Pharmacol* **324:**246–255.
Siwik DA and Brown RD (1996) Regulation of protein synthesis by alpha 1-adrenergic receptor subtypes in cultured rabbit aortic vascular smooth muscle cells. *J Cardiovasc Pharmacol* **27:**508–518.
Skomedal T, Dybvik T, Osnes JB and Aass H (1998) Location of myocardial α_1- and β-adrenoceptors in relation to sympathetic nerve endings in failing human myocardium. *Naunyn-Schmiedeberg's Arch Pharmacol* **358:**R633.
Skrbic R and Chiba S (1992) Pharmacological properties of alpha 1-adrenoceptor-mediated vasoconstrictions in dog and monkey lingual arteries: evidence for subtypes of alpha 1-adrenoceptors. *Heart Vessels* **7:**82–90.
Smith KM, MacMillan JB and McGrath JC (1997) Investigation of α_1-adrenoceptor subtypes mediating vasoconstriction in rabbit cutaneous resistance arteries. *Br J Pharmacol* **122:**825–832.
Sooch S and Marshall I (1995) An atypical β-adrenoceptor mediates relaxation of the rat isolated mesenteric artery. *Br J Pharmacol* **114:**22P.
Sooch S and Marshall I (1996) Evidence for atypical β-adrenoceptors in rat vasculature. *Br J Pharmacol* **117:**261P.
Sooch S and Marshall (1997) Atypical β-adrenoceptors in the rat vasculature. *Ann N Y Acad Sci USA* **812:**211–212.
Stam WR, Van der Graaf PH and Saxena PR (1999) Analysis of α_{1L}-adrenoceptor pharmacology in rat small mesenteric artery. *Br J Pharmacol* **127:**661–670.
Starke K (1972) Alpha sympathomimetic inhibition of adrenergic and cholinergic transmission in the rabbit heart. *Naunyn-Schmiedeberg's Arch Pharmacol.***274**: 18–45.
Starke K (1977) Regulation of noradrenaline release by presynaptic receptor systems. *Rev Physiol Biochem Pharmacol* **77:**1–124.
Starke K (1981) α-Adrenoceptor subclassification. *Rev Physiol Biochem Pharmacol* **88:**199–236.
Starke K (1987) Presynaptic α-autoreceptors. *Rev Physiol Biochem Pharmacol* **107:** 73–146.
Starke K, Borowski E and Endo T (1975a) Preferential blockade of presynaptic α-adrenoceptors by yohimbine. *Eur J Pharmacol* **34:**385–388.
Starke K and Endo T (1975) Presynaptic α-adrenoceptors. *Gen Pharmacol* **7:**307–312.
Starke K, Endo T and Taube HD (1975b) Relative pre- and postsynaptic potencies of α-adrenoceptor agonists in the rabbit pulmonary artery. *Naunyn-Schmiedeberg's Arch Pharmacol* **291:**55–78.
Starke K, Montel H, Gayk W and Merker R (1974) Comparison of the effect of clonidine on pre- and postsynaptic adrenoceptor in the rabbit pulmonary artery. *Naunyn-Schmiedeberg's Arch Pharmacol* **285:**133–150.
Starke K, Trendelenburg AU and Limberger N (1995) Presynaptic α_2-adrenoceptors: subtype determination. *Pharmacol Commun* **6:**99–108.
Stassen FRM, Willemsen MJJMF, Janssen GMJ and Demey JGR (1997) α_1-Adrenoceptor subtypes in rat aorta and mesenteric small arteries are preserved during left ventricular dysfunction post-myocardial infarction. *Cardiovasc Res* **33:**706–713.
Stein CM, Nelson R, Huai B, Wood M and Wood AJJ (1997) Norepinephrine release in the human forearm: effects of epinephrine. *Hypertension* **30:**1078–1084.
Steinberg SF, Jaffe EA and Bilezikian JP (1984) Endothelial cells contain beta adrenoceptors. *Naunyn-Schmiedeberg's Arch Pharmacol* **325:**310–313.
Stevens MJ, Rittinghausen RE, Medcalf RL and Moulds RFW (1982) Prejunctional β-adrenoceptors in human digital arteries. *Eur J Pharmacol* **83:**263–270.
Stjärne L and Brundin J (1975) Dual adrenoceptor-mediated control of noradrenaline secretion from human vasoconstrictor nerves: facilitation by β-receptors and inhibition by α-receptors. *Acta Physiol Scand* **94:**139–141.
Strosberg AD (1997) Structure and function of the β_3-adrenoceptor. *Ann Rev Pharmacol Toxicol* **37:**421–450.
Strosberg AD, Gerhardt CC, Gros J, Jockers R and Pietri-Rouxel F (1998) Reply on the putative existence of a fourth β-adrenoceptor: proof is still missing. *Trends Pharmacol Sci* **19:**165–166.
Strosberg AD and Pietri-Rouxel F (1996) Function and regulation of the three β-adrenoceptor. *Trends Pharmacol Sci* **17:**373–381.
Summers RJ, Kompa A and Roberts SJ (1997) β-Adrenoceptor subtypes and their desensitization mechanisms. *J Auton Pharmacol* **17:**331–343.
Surprenant A, Horstmann DA, Akbarali H and Limbird LE (1992) A point mutation of the α_2-adrenoceptor that blocks coupling to potassium but not calcium currents. *Science (Wash DC)* **257:**977–980.
Susulic VS, Frederich RC, Lawitts J, Tozzo E, Kahn BB, Harper M-E, Himms-Hagen

J, Flier JS and Lowell BB (1995) Targeted disruption of the β_3-adrenergic receptor gene. *J Biol Chem* **270:**29483–294922.
Tagaya E, Tamaoki J, Takemura H, Isono K and Nagai A (1999) Atypical adrenoceptor-mediated relaxation of canine pulmonary artery through a cyclic adenosine monophosphate-dependent pathway. *Lung* **177:**321–332.
Taira N, Yabuuchi Y and Yamashita S (1977) Profile of β-adrenoceptors in femoral, superior mesenteric and renal vascular beds of dogs. *Br J Pharmacol* **59:**577–583.
Takahashi H, Yoshida T, Nishimura N, Nakanishi T, Kondo M and Yoshimura M (1992) Beta3-adrenergic agonist, BRL-26830A, and alpha/beta blocker, arotinolol, markedly increase regional blood flow in the brown tissue in anesthetized rats. *Jap Circ J* **56:**936–942.
Takayanagi I, Harada M, Koike K and Satoh M (1991) Differences in α_1-adrenoceptor mechanisms for phenylephrine and tizanidine in rabbit thoracic aorta and common iliac artery. *Can J Physiol Pharmacol* **49:**1819–1824.
Tamaoki J, Tagaya E, Isono K and Nagai A (1998) Atypical adrenoceptor-mediated relaxation of canine pulmonary artery through a cAMP-dependent pathway. *Biochem Biophys Res Comm* **248:**722–727.
Tanimitsu N, Yajin K, Sasa M and Tsuru H (2000) α_1-Adrenoceptor subtypes and effect of α_{1A}-adrenoceptor agonist NS-49 on guinea pig nasal mucosa vasculature. *Eur J Pharmacol* **387:**73–78.
Tarizzo VI, Coopes RP, Dahlöf C and Zaasgma J (1994) Pre- and postganglionic stimulation-induced noradrenaline overflow is markedly facilitated by a prejunctional β_2-adrenoceptor-mediated control mechanism in the pithed rat. *Naunyn-Schmiedeberg's Arch Pharmacol* **349:**570–577.
Tarizzo V and Dahlöf C (1989) Adrenaline-induced enhancement of the blood pressure response to sympathetic nerve stimulation in adrenal demedullated pithed rat. *Naunyn-Schmiedeberg's Arch Pharmacol* **340:**144–150.
Tavernier G, Galitzky J, Bousquet-Melou A, Montrastruc JL and Berlan M (1992) The positive chronotropic effect induced by BRL 37344 and CGP 12177, two *beta*-3 adrenergic agonists, does not involve cardiac *beta* adrenoceptors but baroreflex mechanisms. *J Pharmacol Exp Ther* **263:**1083–1090.
Teixeira F (1977) The effect of drugs and denervation on removal and accumulation of adrenaline in the perfused hind-limb of the dog. *Arch Int Pharmacodyn Ther* **225:**221–231.
Tesfamariam B and Allen GT (1994) β_1- and β_2-adrenoceptor antagonist activities of ICI-215001, a putative β_3-adrenoceptor agonist. *Br J Pharmacol* **112:**55–58.
Testa R, Destefani C, Guarneri L, Poggesi E, Simonazzi I, Tadei C and Leonardi A (1995) The α_{1d}-adrenoceptor subtype is involved in the noradrenaline-induced contractions of rat aorta. *Life Sci* **57:**PL159–PL163.
Theroux TL, Esbenshade TA, Peavy RD and Minneman KP (1996) Coupling efficiencies of human alpha 1-adrenergic receptor subtypes: titration of receptor density and responsiveness with inducible and repressible expression vectors. *Mol Pharmacol* **50:**1376–1387.
Thom S, Calvete J, Hayes R, Martin G and Sever P (1985) Human vascular smooth muscle responses mediated by alpha$_2$ mechanisms *in vivo* and *in vitro*. *Clin Sci* **68:**147s–150s.
Thompson JM, Wallin BG, Lambert GW, Jennings GL and Esler MD (1998) Human muscle sympathetic activity and cardiac catecholamine spillover: no support for augmented sympathetic noradrenaline release by adrenaline co-transmission. *Clin Sci* **94:**383–393.
Thorin E, Shreeve SM, Thorin-Trescases N and Bevan JA (1997) Reversal of endothelin-1 release by stimulation of endothelial α_2-adrenoceptor contributes to cerebral vasorelaxation. *Hypertension* **30:**830–836.
Timmermans PBMWM and Van Zwieten PA (1982) α_2-Adrenoceptors: classification, localization, mechanisms and target for drugs. *J Med Chem* **25:**1389–1401.
Toda N (1983) Alpha adrenergic receptor subtypes in human, monkey and dog cerebral arteries. *J Pharmacol Exp Ther* **226:**861–868.
Toda N and Shimizu I (1987) Neuroeffector function in mesenteric arteries isolated from beagles of different ages. *J Pharmacol Exp Ther* **240:**223–227.
Toda N, Yoshida K and Okamura T (1995) Involvement of nitroxidergic and noradrenergic nerves in the relaxation of dog and monkey temporal veins. *J Cardiovasc Pharmacol* **25:**741–747.
Tonolo G, Melis MG, Secchi G, Atzeni MM, Angius MF, Carboni A, Ciccarese M, Malavasi A and Maioli M (1999) Association of Trp64Arg β_3-adrenergic-receptor gene polymorphism with essential hypertension in the Sardinian population. *J Hypertens* **17:**33–38.
Toyoshima H, Nasa Y, Hashizume Y, Koseki Y, Isayama Y, Kohsaka Y, Yamada T and Takeo S (1998) Modulation of cAMP-mediated vasorelaxation by endothelial nitric oxide and basal cGMP in vascular smooth muscle. *J Cardiovasc Pharmacol* **32:**543–551.
Trendelenburg AU, Hein L, Gaiser EG and Starke K (1999) Occurrence, pharmacology and function of presynaptic α_2-autoreceptors in $\alpha_{2A/D}$-adrenoceptor-deficient mice. *Naunyn-Schmiedeberg's Arch Pharmacol* **360:**540–551.
Trendelenburg AU, Limberger N and Starke K (1994) α_2-Adrenergic receptors of the α_{2C}-subtype mediate inhibition of norepinephrine release in human kidney cortex. *Mol Pharmacol* **45:**1168–1176.
Trendelenburg AU, Limberger N and Starke K (1996) The presynaptic alpha$_2$ autoreceptors in pig brain are alpha-2A. *J Pharmacol Exp Ther* **278:**462–467.
Trendelenburg AU, Sutej I, Wahl CA, Molderings GJ, Rump LC and Starke K (1997) A re-investigation of questionable subclassifications of presynaptic α_2-autoreceptors: rat vena cava, rat atria, human kidney and guinea-pig urethra. *Naunyn-Schmiedeberg's Arch Pharmacol* **356:**721–737.
Trochu J-N, Leblais V, Rautureau Y, Bévérelli F, Le Marec H, Berdeaux A and Gauthier C (1999) Beta 3-adrenoceptor stimulation induces vasorelaxation mediated essentially by endothelium-derived nitric oxide in rat thoracia aorta. *Br J Pharmacol* **128:**69–76.
Urban R, Szabo B and Starke K (1995) Involvement of α_2-adrenoceptors in the cardiovascular effects of moxonidine. *Eur J Pharmacol* **282:**19–28.
Van der Graaf PH, Deplanne V, Duquenne C and Angel I (1997) Analysis of α_1-adrenoceptors in rabbit lower urinary tract and mesenteric artery. *Eur J Pharmacol* **327:**25–32.
Van der Graaf PH, Shankley NP and Black JW (1996a) Analysis of the activity of α_1-adrenoceptor antagonists in rat aorta. *Br J Pharmacol* **118:**299–310.
Van der Graaf PH, Shankley NP and Black JW (1996b) Analysis of the effects of α_1-adrenoceptor antagonists on noradrenaline-mediated contraction of rat small mesenteric artery. *Br J Pharmacol* **118:**1308–1316.
Van der Zypp A, Kang K-B and Majewski H (2000) Age-related involvement of the endothelium in β-adrenoceptor-mediated relaxation of rat aorta. *Eur J Pharmacol* **397:**129–138.
Vanhoutte PM (1980) Physical factors and regulation of vascular smooth muscle function, in *Handbook of Physiology, Section 2: Circulation, volume II: Vascular Smooth Muscle* (Bohr DF, Somlyo AP, Sparks HV eds) pp 443–474, The American Physiological Society, Washington, DC.
Vanhoutte PM (2000) Are there functional endothelial adrenoceptors? *Pharmacol Toxicol* **87:**L3.
Vanhoutte PM and Lorenz RR (1970) Effect of temperature on the reactivity of saphenous, mesenteric, and femoral veins of the dog. *Am J Physiol* **218:**1746–1750.
Vanhoutte PM and Miller VM (1989) Alpha$_2$-adrenoceptors and endothelium-derived relaxing factor. *Am J Med* **87:**1S–5S.
VanRipper DA and Bevan JA (1991) Selective variation of agonist and neuronally mediated vasoconstriction with middle cerebral artery branch order. *J Pharmacol Exp Ther* **257:**879–886.
Vargas HM Zhou L and Gorman AJ (1994) Role of vascular alpha-1 adrenoceptor subtypes in the pressor response to sympathetic nerve stimulation in the pithed rat. *J Pharmacol Exp Ther* **271:**748–754.
Vatner SF, Knight DR and Hintze TH (1985) Norepinephrine-induced beta 1-adrenergic peripheral vasodilation in conscious dogs. *Am J Physiol* **249:**H49–H56.
Vaz-da-Silva MJ, Magina S, Domingues-Costa A, Moura D and Guimarães S (1996) The role of the endocardium in the facilitatory effect of bradykinin on electrically-induced release of noradrenaline in rat cardiac ventricle. *Br J Pharmacol* **118:**364–368.
Viard P, Macrez N, Coussin F, Morel J-L and Mironneau J (2000) Beta-3 adrenergic stimulation of L-type Ca^{2+} channels in rat portal vein myocites. *Br J Pharmacol* **129:**1497–1505.
Vila E and Badia A (1995) Chronic administration of oxprenolol and metoprolol attenuated sympathetic cardivascular responses in non-adrenalectomized pithed rats. *J Auton Pharmacol* **15:**393–402.
Villalobos-Molina R and Ibarra M (1996) Alpha 1-adrenoceptors mediating contraction in arteries of normotensive and spontaneously hypertensive rats are of the alpha 1D or alpha 1A subtypes. *Eur J Pharmacol* **298:**257–263.
Villalobos-Molina R, López-Guerrero JJ and Ibarra M (1999) Functional evidence of α_{1D}-adrenoceptor in the vasculature of young and adult spontaneously hypertensive rats. *Br J Pharmacol* **126:**1534–1536.
Vincent HH, Man in't Veld AJ, Boomsma F, Wenting GS and Schalekamp MADH (1982) Elevated plasma noradrenaline in response to β-adrenoceptor stimulation in man. *Br J Clin Pharmacol* **13:**717–721.
Vizi ES, Somogyi GT, Hadhazy P and Knoll J (1973) Effect of duration and frequency of stimulation on the presynaptic inhibition by α-adrenoceptor stimulation of the adrenergic transmission. *Naunyn-Schmiedeberg's Arch Pharmacol* **280:**79–91.
Walston J, Silver K, Bogardus C, Knowler WC, Celi FS, Austin S, Manning B, Strosberg AD, Stern MP, Raben N, Sorkin JD, Roth J and Shuldiner AR (1995) Time of onset of non-insulin-dependent diabetes mellitus and genetic variation in the β_3-adrenergic-receptor gene. *N Engl J Med* **333:**343–347.
Wanstall JC and O'Donnell SR (1989) Influence of age on calcium entry blocking drugs in rat aorta spasmogen-dependent. *Eur J Pharmacol* **159:**241–246.
Waterman SA (1997) Role of N-, P- and Q-type voltage-gated calcium channels in transmitter release from sympathetic neurones in the mouse isolated vas deferens. *Br J Pharmacol* **120:**393–398.
Watkins PJ (1998) The enigma of autonomic failure in diabetes. *J R Coll Physicians Lond* **32:**360–365.
Weinshank RL, Zgombick JM, Macchi M, Adham N, Lichtblau H, Branchek TA and Hartig PA (1990) Cloning, expression and pharmacological characterization of a human α_{2B}-adrenergic receptor. *Mol Pharmacol* **38:**681–688.
White RE, Jolly SR and Carrier GO (1986) Enhanced relaxation of canine coronary artery to isoproterenol and salbutamol after removal of endothelial cells. *Gen Pharmacol* **17:**497–499.
Widén E, Lehto M, Kanninen T, Walston J, Shuldiner AR and Groop LC (1995) Association of a polymorphism in the β_3-adrenergic-receptor gene with features of the insulin resistance syndrome in Finns. *N Engl J Med* **333:**348–351.
Willette RN, Hieble JP and Sauermelch CF (1991) The role of *alpha* adrenoceptor subtypes in sympathetic control of the acral-cutaneous microcirculation. *J Pharmacol Exp Ther* **256:**599–605.
Williams TJ, Blue DR, Daniels DV, Davis B, Elworthy T, Gever JR, Kava MS, Morgans D, Padilla F, Tassa S, Vimont RL, Chapple CR, Chess-Williams R, Eglen RM, Clarke DE and Ford APDW (1999) In vitro α_1-adrenoceptor pharmacology of RO 70–0004 and RS-100329, novel α_{1A}-adrenoceptor selective antagonists. *Br J Pharmacol* **127:**252–258.
Wilson C, Wilson S, Piercy V, Sennitt MV and Arch S Jr (1984) The rat lipolytic beta-adrenoceptor studies using novel beta-adrenoceptor agonists. *Eur J Pharmacol* **100:**309–319.
Wilson S, Chambers JK, Park JE, Ladurner A, Cronk DW, Chapman CG, Kallender H, Browne MJ, Murphy J and Young PW (1996) Agonist potency of the cloned human *beta*-3 adrenoceptor depends on receptor expression level and nature of assay. *J Pharmacol Exp Ther* **279:**214–221.
Winzer-Serhan UM and Leslie FM (1997) α_{2B}-Adrenoceptor mRNA expression during rat brain development. *Dev Brain Res* **100:**90–100.
Winzer-Serhan UH, Raymon HK, Broide RS, Chen Y and Leslie FM (1997a) Expression of α_2-adrenoceptors during rat brain development. I. α_{2A} Messenger RNA expression. *Neuroscience* **76:**241–260.
Winzer-Serhan UH, Raymon HK, Broide RS, Chen Y and Leslie FM (1997b) Expres-

sion of α_2-adrenoceptors during rat brain development. II. α_{2C} Messenger RNA expression and (^{3}H) rauwolscine binding. *Neuroscience* **76:**261–272.
Wise A, Watson-Koken MA, Rees S, Lee M and Milligan G (1997) Interactions of the α_{2A}-adrenoceptor with multiple Gi-family proteins: studies with pertussis toxin-resistant G-protein mutants. *Biochem J* **321:**721–728.
Wu D, Katz A, Lee CH and Simon MI (1992) Activation of phospholipase C by alpha 1-adrenergic receptors is mediated by the alpha subunits of Gq family. *J Biol Chem* **267:**25798–25802.
Xiao RP, Ji X and Lakatta EG (1995) Functional coupling of beta 2-adrenoceptor to a pertussis toxin-sensitive G protein in cardiac myocytes. *Mol Pharmacol* **47:**322–329.
Xu K, Lu Z, Wei H, Zhang Y and Han C (1998) Alteration of α_1-adrenoceptor subtypes in aortas of 12-month-old spontaneously hypertensive rats. *Eur J Pharmacol* **344:**31–36.
Xu KM, Tang F and Han C (1997) Alterations of mRNA levels of alpha$_1$-adrenoceptor subtypes with maturation and ageing in different rat blood vessels. *Clin Exp Pharmacol Physiol* **24:**415–417.
Yamaguchi I and Kopin IJ (1980) Differential inhibition of alpha-1 and alpha-2 adrenoceptor mediated pressor responses in pithed rats. *J Pharmacol Exp Ther* **214:**275–281.
Yamamoto Y and Koike K (1999) Alpha1-adrenoceptors in the guinea pig thoracic aorta. *J Smooth Muscle Res* **35:**181–192.
Yan M, Zhang Y, Du XJ and Han C (1998) Functional assessment of alpha 1-adrenoceptor subtypes in porcine coronary artery. *Clin Exp Pharmacol Physiol* **25:**682–685.
Yang M, Ruan J, Voller M, Schalken J and Michel MC (1999) Differential regulation of human α_1-adrenoceptor subtypes. *Naunyn-Schmiedeberg's Arch Pharmacol* **359:**439–446.
Yousif M, Kadavil EA and Oriowo MA (1998) Heterogeneity of alpha 1-adrenoceptor subtypes mediating noradrenaline-induced contraction of the rat superior mesenteric artery. *Pharmacology* **56:**196–206.
Zaagsma J and Nahorski SR (1990) Is the adipocyte β-adrenoceptor a prototype for the recently cloned atypical "β_3-adrenoceptor"? *Trends Pharmacol Sci* **11:**3–7.
Zeiders JL, Seidler FJ and Slotkin TA (1999) Agonist-induced sensitization of β-adrenoceptor signaling in neonatal rat heart: expression and catalytic activity of adenylyl cyclase. *J Pharmacol Exp Ther* **291:**503–510.
Zeiders JL, Seidler FJ and Slotkin TA (2000) Ontogeny of G-protein expression: control by beta-adrenoceptors. *Brain Res Dev Brain Res* **120:**125–134.
Zhang W, Edvinsson L and Lee TJ-F (1998) Mechanism of nicotine-induced relaxation in the porcine basilar artery. *J Pharmacol Exp Ther* **284:**790–797.
Zhong H and Minemann KP (1999) α_1-Adrenoceptor subtypes. *Eur J Pharmacol* **375:**261–276.
Zhou L and Vargas HM (1996) Vascular α_{1D}-adrenoceptors have a role in the pressor response to phenylephrine in the pithed rat. *Eur J Pharmacol* **305:**173–176.
Zoukos Y, Thomaides T, Mathias CJ and Cuzner ML (1999) High beta-adrenoceptor density on peripheral blood mononuclear cells in progressive multiple sclerosis: a manifestation of autonomic dysfunction. *Acta Neurol Scand* **99:**1706–1712.
Zuscik JM, Sands S, Ross AS, Waugh DJJ, Gaivin RJ, Morilak D and Perez DM (2000) Overexpression of the α_{1B}-adrenergic receptor causes apoptotic neurodegeneration: multiple system atrophy. *Nature Med* **6:**1388–1394.

Correction to "Vascular Adrenoceptors: An Update"

In the above article [Guimarães S and Moura D (2001) **53:**319–356], there are two errors in the text. In the Abstract, "nine subtypes (α_{1A}, α_{1B}, α_{1D}, $\alpha_{2A/D}$, α_{2B}, $\alpha_{2A/D}$, β_1, β_2, and β_3). . . " should be "nine subtypes (α_{1A}, α_{1B}, α_{1D}, $\alpha_{2A/D}$, α_{2B}, α_{2C}, β_1, β_2, and β_3). . . " and in Table 2, for the species *Rat, Femoral artery* under the heading *Functional*, α_{1D} should be α_{1L}.

The authors regret these errors and apologize for any confusion or inconvenience they may have caused.