

DONALD G. VIDT, MD AND ALAN W. BAKST, RPH, PHARM.D, EDITORS

# Adrenergic receptors: structure and function

ROBERT M. GRAHAM, MD

■ Adrenergic receptors are cell surface glycoproteins that recognize and selectively bind the catecholamines, norepinephrine and epinephrine, which are released from sympathetic nerve endings and the adrenal medulla.<sup>1-3</sup> By transducing the external catecholamine stimulus into an intracellular signal, these receptors mediate the actions of the sympathetic nervous system, including a variety of responses such as arteriolar smooth muscle contraction and cardiac contraction, which are critically involved in cardiac function and blood pressure homeostasis.<sup>1,4</sup> Activation or blockade of these receptors is, thus, a major therapeutic approach for the management of a number of cardiovascular disorders, including hypertension, angina pectoris, and cardiac arrhythmias.<sup>4,5</sup> Additionally, alterations in these receptors or in their coupled intracellular effectors may contribute to or underlie the pathogenesis of cardiovascular diseases such as cardiac hypertrophy, congestive heart failure, and hypertension. Over the past few years, major insights have been gained into the physiology and pharmacology of these receptors, particularly in relationship to the molecular mechanisms involved in their ability to mediate "transmembrane signalling." These insights, which will be reviewed here, have resulted from the isolation and characterization of adrenergic receptors,<sup>3</sup> from the determination of their deduced primary and secondary structures, and from structure-function studies using classical biochemistry and pharmacology as well as recombinant DNA techniques.<sup>6,7</sup>

□ INDEX TERMS: RECEPTORS, ADRENERGIC □ CLEVE CLIN J MED 1990; 57:481-491

**A**DRENERGIC RECEPTORS are broadly classified as  $\alpha_1$ ,  $\alpha_2$ ,  $\beta_1$ , and  $\beta_2$  based on their different pharmacological specificities and physiologic effects,<sup>1</sup> although there is now considerable evidence that additional subtypes exist that activate distinct cellular signals. Initially, two types

of "adrenotropic" receptors,  $\alpha$  and  $\beta$ , were described, based on the finding that responses to adrenergic stimuli could be divided into two major categories.<sup>8</sup> Although each receptor subtype mediated both excitatory and inhibitory responses, and these varied with the effector organ or system, it was their ability to respond to a series of sympathomimetic amines with a distinct order of potency that allowed them to be uniquely distinguished.

Using similar techniques based on differences in effector-organ specificity for adrenergic agonists,  $\beta$ -adrenergic receptors were subsequently subclassified into two major groups,  $\beta_1$  and  $\beta_2$ .<sup>9,10</sup> This subdivision was confirmed with the development of selective  $\beta_1$ - and  $\beta_2$ -antagonists.<sup>11</sup> Evidence from radioligand binding studies have, more re-

From the Department of Heart and Hypertension Research, Research Institute, The Cleveland Clinic Foundation. This article was produced by Consultants in Medical Education, Inc., Manhasset, New York, under an educational grant from Schering-Plough International.

Address reprint requests to R.M.G., Department of Heart and Hypertension Research, The Cleveland Clinic Foundation, One Clinic Center, 9500 Euclid Avenue, Cleveland, Ohio 44195.

cently, additionally indicated that, although one or the other subtype predominates in most tissues, both can coexist in the same tissue and on the same cell.<sup>12</sup> Nevertheless, the distinction between  $\beta$ -adrenergic receptor subtypes is subtle; their binding affinities for even the most discriminant drugs differ by only 10- to 100-fold; both stimulate the formation of cAMP, and both use the same coupling protein to initiate signaling.<sup>7</sup> It is of interest, therefore, that it has now been demonstrated that both  $\beta$ -receptor subtypes are the products of distinct genes located on different chromosomes, and that these genes encode unique, although highly homologous, proteins.<sup>7</sup>

A subclassification of  $\alpha$ -receptors was not proposed until some 25 years after that for  $\beta$ -adrenergic receptors. At first, this was based on anatomical considerations to differentiate postjunctional  $\alpha$ -receptors ( $\alpha_1$ ) that mediate responses, such as vascular smooth muscle contraction, from prejunctional release-modulating receptors ( $\alpha_2$ ) that inhibit stimulus-induced norepinephrine release from sympathetic nerve terminals.<sup>13</sup> With the development and characterization of subtype selective  $\alpha$ -adrenergic agonists and antagonists, a more broadly applicable and functional classification of  $\alpha$  receptors was proposed<sup>14</sup> and has been confirmed by (1) the findings of radioligand binding studies and biochemical receptor characterization experiments<sup>1</sup>; (2) by in vitro and in vivo physiologic studies<sup>1</sup>; and (3) more recently, by the isolation of distinct genes for each receptor subtype.<sup>15,16</sup>

#### STRUCTURE AND FUNCTION OF ADRENERGIC RECEPTORS

Seven adrenergic receptors (Table 1) have been cloned and their amino acid sequences deduced from the nucleotide composition of their genes. They are all members of a superfamily of receptors<sup>6,7</sup> that includes, at this time, over 30 distinct but related proteins (Figure 1). Like other members of this gene family, all adrenergic receptors are composed of single polypeptide chains, ranging from 402 to 515 amino acids in length. Each receptor contains seven

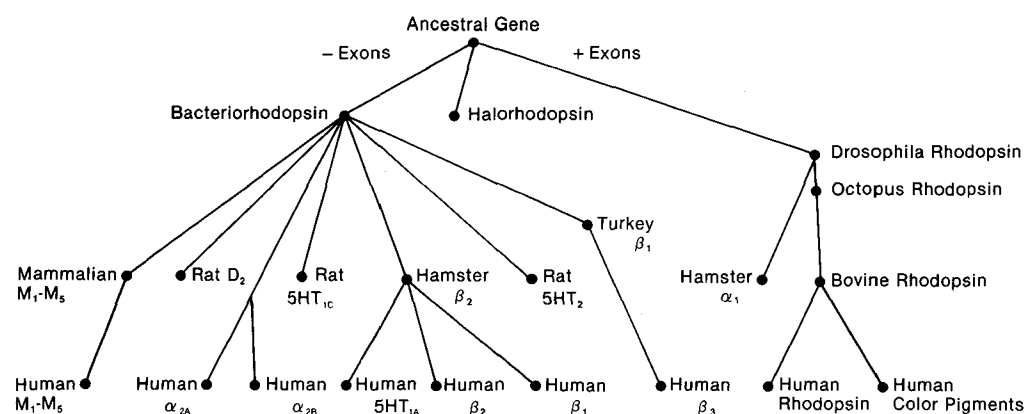


FIGURE 1. Hypothetical evolutionary tree linking the genes for adrenergic receptors with those of structurally related receptors.

TABLE 1  
CHARACTERISTICS OF CLONED ADRENERGIC RECEPTORS

Subtype	Amino acids	Mr* (kDa)	Species	Tissue
$\alpha_1$	515	80	Hamster	Smooth muscle
$\alpha_{2a}$	450	64	Human	Platelet
$\alpha_{2b}$	461	67	Human	Kidney
$\beta_1$	477	62	Human	Placenta
$\beta_1$	483	53	Turkey	Erythrocyte
$\beta_2$	418/413	64	Hamster/ human	Lung/placenta
$\beta_3$	402	65	Human	Placenta

\*Mr, apparent molecular weight as determined by sodium dodecyl sulfate polyacrylamide gel electrophoresis. In most instances, glycosylation of the receptor proteins results in their migrating as species of higher molecular weights than those calculated from their amino acid composition.

stretches of 20 to 28 hydrophobic amino acids that likely represent membrane spanning regions. In this regard, their structure is highly homologous to that of rhodopsin,<sup>7</sup> which mediates visual transduction (Figure 2).

The amino terminus of each receptor is located extracellularly and, in most instances, contains several sites at which the proteins are glycosylated. The carboxyl terminus is located intracellularly and contains sites at which the proteins are post-translationally modified by phosphorylation or by thioesterification with palmitic acid. The transmembrane spanning regions are linked by three intracellular and three extracellular loops of variable length, with the third intracellular loop most likely interacting directly with the receptor coupled G-protein. The first and second extracellular loops contain cysteine residues that are highly conserved in almost all

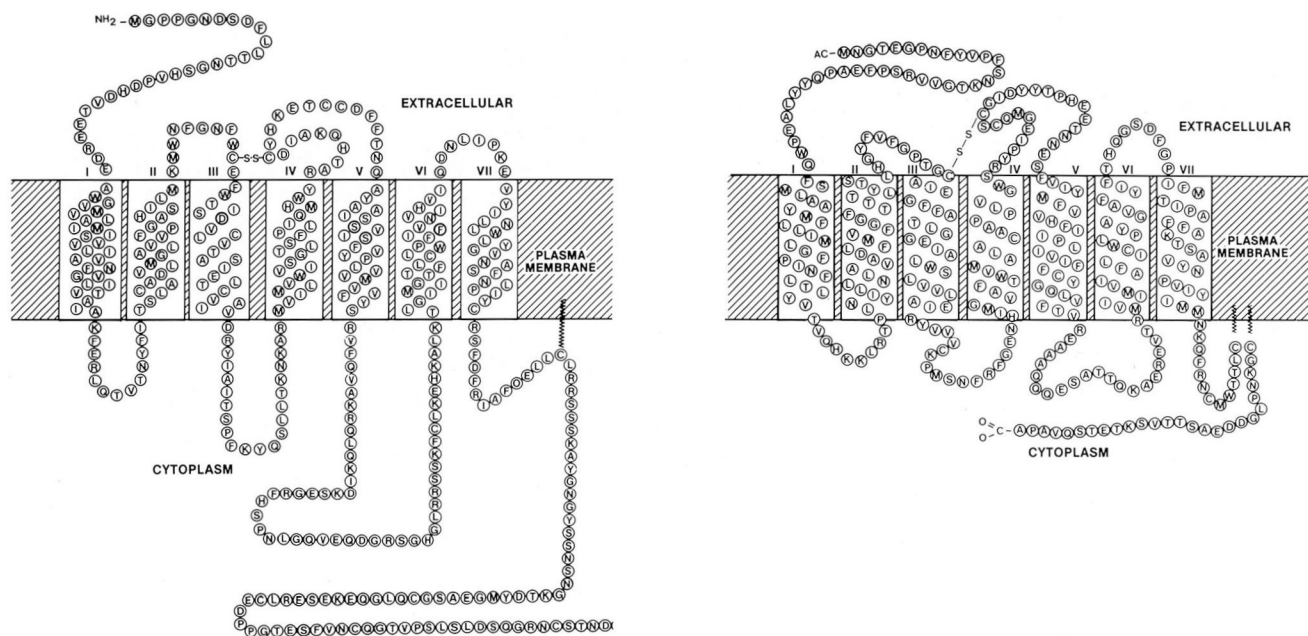


FIGURE 2. Models for the transmembrane topology of the hamster  $\beta_2$ -adrenergic receptor (left panel) and the bovine rhodopsin receptor (right panel). The similarity in the structures of these polytopic proteins is apparent. See text for a detailed description of the receptors' structures. Adapted from references 6 and 34.

receptors. In the  $\beta$ -adrenergic receptor and in rhodopsin, these cysteine residues are essential for the correct folding of the proteins and their expression on the plasma membrane, as well as for the maturational processing of their glycan moieties.<sup>31,32</sup> With rhodopsin, this has been demonstrated to be due to the formation of a disulfide bond between these cysteine residues, which is essential for protein folding.<sup>33</sup>

Interestingly, the ligand-binding domain of  $\beta$ -adrenergic receptors involves residues within the putative  $\alpha$ -helical transmembrane regions of each protein, rather than residues in extracellularly located hydrophilic regions.<sup>6</sup> This finding is of note in relationship to the known orientation of the ligand, 11-*cis* retinal, which sits in the rhodopsin binding pocket and is buried within the membrane bilayer.<sup>32</sup> Retinal binds to rhodopsin by forming a Schiff base with a lysine (Lys<sup>296</sup>) in the seventh transmembrane segment of rhodopsin. Additionally, a glutamic acid (Glu<sup>113</sup>) in the third transmembrane segment serves as the counterion for the retinylidene Schiff base.<sup>34</sup> It is of interest, therefore, that structure-function studies of the  $\beta_2$ -adrenergic receptor indicate that adrenergic agonists most likely interact with the receptor by forming an ion pair between the protonated amine of the ligand and the carboxylate side

chain of an aspartic acid (Asp<sup>113</sup>).<sup>6</sup> This residue, like Glu<sup>113</sup> of rhodopsin, is also located in the third transmembrane segment. In addition, two serine residues in the fifth transmembrane segment, and a phenylalanine in the sixth transmembrane segment, probably participate in ligand binding by forming hydrogen bonds with the hydroxyl groups and a hydrophobic bond with the aromatic ring of adrenergic ligands, respectively.<sup>6</sup>

As mentioned above, the third intracellular loop of adrenergic receptors may interact directly with the receptor-coupled G-protein. This interaction probably involves charged amino acids in both proteins that participate in the formation of putative amphiphilic helices. The amphiphilic regions in the third intracellular loop may become exposed as a result of the change in receptor conformation that occurs with agonist binding.<sup>6</sup>

Finally, there is evidence that phosphorylation of amino acids in the third intracellular loop and, in the carboxy terminal tail of  $\beta$ -adrenergic receptors, plays a role in desensitizing the receptor to subsequent agonist stimulation.<sup>35,36</sup>

#### ADRENERGIC RECEPTOR RESPONSES

Adrenergic receptors are widely distributed (Tables 2 and 3) in a variety of tissues and organs where they me-

**TABLE 2**  
TARGET TISSUES AND RESPONSES OF  $\alpha$ -ADRENERGIC RECEPTORS

Receptor subtype	Tissue	Response
$\alpha_1$ -adrenergic	Smooth muscle: vascular, uterus, trigone, pilomotor, ureter, sphincters (gastrointestinal and bladder), eye (iris), radial, vas deferens	Contraction
	Smooth muscle (gastrointestinal)	Relaxation
	Liver*	Glycogenolysis, gluconeogenesis, ureogenesis
	Myocardium	Increased force of contraction
	Central nervous system	Increased locomotor activity, neurotransmission
	Salivary glands	Secretion ( $K^+$ , $H_2O$ )
	Kidney (proximal tubule)	Glucogenesis
$\alpha_2$ -adrenergic	Adipose tissue	Glycogenolysis
	Sympathetic nerve terminal	Inhibition of norepinephrine release
	Vascular smooth muscle	Contraction
	Platelets	Aggregation, granule release
	Central nervous system	Sedation, inhibition of sympathetic outflow, neurotransmission
	Adipose tissue	Inhibition of lipolysis
	Eye	Decreased intraocular pressure
	Endothelium	Release of vasodilator substance
	Jejunum	Inhibition of secretion
	Kidney	Inhibition of renin release
	Pancreatic islet cells	Inhibition of insulin release
	Cholinergic neurons and cell bodies of noradrenergic neurons	Inhibition of firing
	Melanocytes	Inhibition of MSH-induced granule dispersion

\*Applies mainly to the rat; in humans,  $\beta_2$ -adrenergic responses predominate.

diate responses critical to cardiovascular, respiratory, metabolic and other homeostatic systems.<sup>1,4,5</sup> Receptor-regulated control of these processes occurs either directly at the tissue or organ level or indirectly through modulation of sympathetic neurotransmission at the level of the central nervous system, ganglia or pre-junctional nerve endings.

Receptors of the  $\beta_1$ -subtype play a major role in regulating cardiac contractility and heart rate. Although receptor-mediated cAMP generation via  $G_s$  has been considered to be the major signalling mechanism for these responses, as discussed above, there is recent evidence to suggest that, as importantly,  $G_s$  may be directly involved

by its ability to modulate plasma membrane ion channels.<sup>29</sup>  $\beta_2$ -Adrenergic receptors are also involved in circulatory control by their ability to mediate vascular smooth muscle dilatation. Smooth muscle relaxation is also of major importance in the control of airway resistance as well as in gastrointestinal activity. In skeletal muscle,  $\beta_2$ -adrenergic receptors mediate  $K^+$  uptake and, in this tissue, as well as the liver and pancreas, they play an important role in regulating intermediary metabolism by mediating glycogenolysis, gluconeogenesis, and insulin secretion.

$\alpha$ -Adrenergic receptors are also intimately involved in cardiovascular function as well as in modulating sympathetic activity at the central and peripheral nervous system level.  $\alpha_1$ -Adrenergic receptors increase peripheral resistance by mediating vascular smooth muscle contraction, and also cause smooth muscle contraction in a variety of other organs, such as the bladder, uterus, ureter, and eye.  $\alpha_2$ -Adrenergic receptors modulate peripheral resistance both directly by mediating the release of vasodilator substances from endothelium and by inhibiting neurotransmitter release from prejunction sympathetic nerve terminals.

#### PHARMACOLOGY OF ADRENERGIC AGONISTS AND ANTAGONISTS

Adrenergic receptors of all subtypes are activated by the endogenous catecholamines, norepinephrine and epinephrine. These compounds are  $\beta$ -phenylethylamines, which are the prototype of sympathomimetic amines, consisting of a benzene ring and an ethylamine side chain.<sup>4</sup> They possess one point of asymmetry at the  $\beta$ -carbon atom, and their pharmacologic activity generally resides in the R(-)-enantiomer, with the S(+)-enantiomer being considerably (10- to 500-fold) less potent. Compounds that have full agonist activity generally have hydroxyl groups at both the 3 and 4 positions of the benzene ring.

The other major requirement for sympathomimetic activity is the separation of the benzene ring from the amino group of the ethylamine side chain by two carbon atoms. At physiologic pH, this amino group is protonated and, together with the benzylic hydroxyl group and the two phenolic hydroxyls, is a major determinant of receptor binding. When one or both of the phenolic hydroxyls is absent, sympathomimetic activity is reduced. Hydroxyl groups at the 3 and 5 positions confer  $\beta_2$ -receptor selectivity on compounds with large amino substituents. An increase in the size of the alkyl substituent on the amino group increases  $\beta$ - $\alpha$ -receptor activity, although a notable exception is phenylephrine,

which has an N-methyl substituent, but is almost a pure  $\alpha_1$ -agonist.

In addition to  $\beta$ -phenylethylamines, there are a variety of noncatecholamines that possess sympathomimetic activity. However, responses to these compounds are in part determined by their ability to release norepinephrine from sympathetic nerve terminals. Responses to these agents are thus mostly due to  $\alpha$ - and  $\beta_1$ -receptor-mediated effects since norepinephrine is a weak  $\beta_2$ -agonist.

A variety of compounds are available that block either  $\alpha$ - or  $\beta$ -adrenergic receptors or both. The prototype of  $\beta$ -adrenergic antagonists is dichloroisoproterenol (DCI) in which the phenolic hydroxyls of the  $\beta$ -agonist, isoproterenol, have been substituted with chlorides.<sup>11</sup> However, it is not used clinically as it still retains prominent partial agonist activity at  $\beta$ -adrenergic receptors. Propranolol [1-(isopropylamino)-3-(1-naphthoxy)-2-propanol] was the first  $\beta$ -adrenergic antagonist to become widely used clinically and has superseded DCI as the prototype  $\beta$ -blocker.<sup>37</sup> It is a highly potent, nonselective  $\beta$ -adrenergic blocking agent that is devoid of any agonistic activity (ie, intrinsic sympathomimetic activity, or ISA). A variety of structurally related nonselective and subtype selective  $\beta$ -blockers have subsequently been developed, and many are useful therapeutic agents.<sup>5</sup> More recently, additional compounds have been developed that either block both  $\beta$ - and  $\alpha$ -adrenergic receptors or possess both  $\beta$ -blocking and direct vasodilator activity.

$\alpha$ -Adrenergic blocking drugs<sup>1</sup> are structurally diverse and include compounds of the imidazoline class that are both nonselective (eg, phentolamine, tolazoline) and  $\alpha_2$ -selective (eg, idazoxan) antagonists; yohimbans and their carboxamide analogs (eg, yohimbine and rauwolfscine) that are  $\alpha_2$ -selective antagonists; and a variety of compounds that display  $\alpha_1$ -selective blocking activity, including quinazolines (eg, prazosin), benzodioxanes (eg, WB 4101), phenylethylamines (eg, HEAT) and indoles (eg, indoramin). Additionally, there are compounds that irreversibly inactivate  $\alpha$ -receptors, such as haloalkylamines (eg, phenoxybenzamine) and compounds that block  $\beta$ - as well as  $\alpha$ -adrenergic receptors, such as ethanolamines (eg, labetalol).

The availability of this wide variety of agents has greatly aided in the identification and characterization of adrenergic receptor subtypes and their effector responses, as well as in their isolation and molecular characterization. Adrenergic agonists and antagonists are also widely used in clinical medicine for treating a plethora of diverse disorders ranging from cardiac

TABLE 3  
TARGET TISSUES AND RESPONSES OF  $\beta$ -ADRENERGIC RECEPTORS

Receptor subtype	Tissue	Response
$\beta_1$ -adrenergic	Myocardium	Increase in heart rate Increase in contractility and conduction velocity Increase in automaticity and conduction velocity Increase in automaticity and conduction velocity
	S-A node	
	Atria	
	A-V node	
	His-Purkinje system	Increase in automaticity and conduction velocity Increase in contractility, conduction, velocity, automaticity, and rate of idioventricular pacemakers
	Ventricles	
	Kidney	Renin secretion
	Adipose tissue	Lipolysis
Posterior pituitary	Antidiuretic hormone secretion	
Sympathetic nerve terminal	Increased neurotransmitter release	
$\beta_2$ -adrenergic	Smooth muscle: vascular, uterus, gastrointestinal (stomach, intestine, gall bladder and bile ducts), bladder (detrusor), lung (tracheal and bronchial)	Relaxation
	Skeletal muscle	Increased contractility, glycogenolysis, K <sup>+</sup> uptake
	Liver	Glycogenolysis and gluconeogenesis
	Pancreas	Insulin secretion
	Splenic capsule	Relaxation
	Salivary glands	Amylase secretion
	Lung: bronchial glands	Increased secretion
	$\beta_3$ -adrenergic	Ileum (guinea pig)
Fat cells (rat)		Lipolysis

arrhythmias and hypertension to asthma, glaucoma, and premature labor (vide infra).  $\beta$ -Adrenergic receptors of the  $\beta_1$ -subtype recognize adrenergic agonists with the following relative order of potency: isoproterenol > epinephrine = norepinephrine > > phenylephrine, and are inactivated more effectively with  $\beta_1$ -selective blockers, such as betaxolol, than with  $\beta_2$ -selective blockers, such as ICI 118551.

By contrast, at  $\beta_2$ -receptors the relative order of potency of agonists is isoproterenol > epinephrine > norepinephrine > > phenylephrine; additionally, the antagonist ICI 118551 is more potent than betaxolol.

$\beta_3$ -Adrenergic receptors are activated more potently by norepinephrine than by epinephrine and display the following relative order of potency for agonists: isoproterenol > BRL 37344 > noradrenaline > epine-

phrine. Interestingly, however, most classical  $\beta$ -adrenergic antagonists are ineffective as  $\beta_3$  blocking agents, with only the  $\beta_1$ -selective blocker, CGP20712A, and the  $\beta_2$ -selective blocker, ICI 118551, displaying any degree of blocking activity.

$\alpha$ -Adrenergic receptors of both the  $\alpha_1$ - and  $\alpha_2$ -subtypes are activated with reasonably similar potency by both epinephrine and norepinephrine.<sup>1</sup> However, they can be readily identified by their relative potency for a series of agonists (eg, amidephrine, methoxamine, and phenylephrine for  $\alpha_1$ -receptors and BHT 933, UK 14,304 and p-aminoclonidine for  $\alpha_2$ -receptors) and antagonists (eg, prazosin, WB 4101, and HEAT for  $\alpha_1$ -receptors and idazoxan, rauwolscine and yohimbine for  $\alpha_2$ -receptors). Moreover, recent evidence suggests that the  $\alpha_{1A}$  and  $\alpha_{1B}$ -subtypes can be discriminated by the ability of methoxamine to more potently activate the  $\alpha_{1A}$  than  $\alpha_{1B}$  subtype, and by the ability of phentolamine and WB4101 to more readily block  $\alpha_{1A}$  than  $\alpha_{1B}$  receptors. Receptors of the  $\alpha_{2A}$  and  $\alpha_{2B}$ -subtypes recognize rauwolscine with similar affinity. However, those of the  $\alpha_{2A}$ -subtype have high affinity for the agonist, oxy-metazoline, and low affinity for the antagonist, prazosin, whereas  $\alpha_{2B}$  receptors recognize these ligands with the opposite order of affinities.<sup>20,23</sup>

---

#### ADRENERGIC AGENTS: THERAPEUTIC APPLICATIONS

---

Compounds that activate or block adrenergic receptors are widely used as therapeutic agents.  $\beta$ -Agonists, particularly nonselective agents such as isoproterenol and epinephrine, are particularly useful as cardiac stimulants in the treatment of hypotensive disorders and bradyarrhythmias, and in the treatment of various allergic disorders.<sup>4,5</sup> Compounds that selectively activate  $\beta_1$ -adrenergic receptors, such as dopamine and dobutamine, are also used as cardiac stimulants in treating certain forms of congestive heart failure, whereas  $\beta_2$ -selective agonists are potent bronchodilators for the treatment of asthma, and are useful in arresting premature labor since they relax uterine smooth muscle.  $\alpha$ -Adrenergic agonists are also of use in the treatment of hypotensive disorders because of their ability to contract vascular smooth muscle and increase peripheral resistance. This effect also underlies their use in controlling hemorrhage, as nasal decongestants, and to retard the absorption of local anesthetics. Compounds that selectively activate  $\alpha_2$ -receptors, such as clonidine and guanabenz, are effective antihypertensive agents, which lower arterial pressure by inhibiting sympathetic outflow. Finally, these agents are widely used as mydriatics and to stimulate contraction of the uterus postpartum.

A number of  $\beta$ -adrenergic antagonists are available as therapeutic agents for the treatment of angina pectoris, cardiac arrhythmias, hypertension, migraine, thyrotoxicosis, anxiety states, tremors, and glaucoma, and to limit myocardial infarct size.<sup>4,5</sup> Differences between the various agents are related not only to their variable selectivity for  $\beta_1$ - or  $\beta_2$ -receptors, but also to their non- $\beta$ -blocking properties, such as intrinsic sympathomimetic activity, membrane stabilizing effects, lipophilicity, metabolism, and duration of action. All agents available for clinical use, however, competitively antagonize the  $\beta$ -receptor-mediated effects of sympathomimetic amines due to a direct interaction with the receptor that is independent of any effects on adrenergic nerves or on the basic response mechanism of effector cells.

Although the major action of these agents is to block  $\beta$ -receptors, some (eg, pindolol, oxprenolol, acebutolol, practolol, and alprenolol) produce weak agonist responses. This partial agonist activity or ISA may be of advantage in patients at risk from  $\beta$ -blockade—for example, asthmatics and subjects with congestive heart failure—because the ISA may limit the degree of  $\beta$ -receptor antagonism. Thus, pindolol has been reported to be “bronchoprotective” in many patients who develop bronchospasm with propranolol.

Agents that possess ISA or are  $\beta_1$ -selective may also be desirable for use in patients with peripheral vascular disease, such as intermittent claudication or Raynaud's phenomenon. In these patients, blockade of vasodilatory  $\beta_2$ -receptors may further reduce blood flow, which is already compromised in the extremities. Other side effects of  $\beta$ -blockers include a number of metabolic and central nervous system effects. The metabolic effects are mainly due to their  $\beta$ -blocking activity and include hypoglycemia in insulin-dependent diabetics, a masking of the symptoms of hypoglycemia and, in certain circumstances, hyperkalemia. These effects are due to blockade of  $\beta$ -receptor-mediated insulin release, hepatic and skeletal muscle glycogenolysis and gluconeogenesis, skeletal muscle  $K^+$  uptake and increases in cardiac rate and force of contraction.  $\beta$ -Blockers, particularly nonselective agents, may also impair lipoprotein metabolism leading to increases in serum triglycerides and to decreases in high-density lipoprotein cholesterol levels.

As mentioned above, compounds are available for clinical use that block both  $\beta$ - and  $\alpha$ -adrenergic receptors. The prototype of these compounds is the ethanolamine, labetalol, which exhibits both selective  $\alpha_1$ - and nonselective  $\beta$ -adrenergic blocking activity and is useful as an antihypertensive agent.<sup>38,39</sup> However, its  $\alpha$ -blocking potency is only one-tenth that of phen-

tolamine, and it is approximately one-third as potent as propranolol in blocking  $\beta$ -receptors. Moreover, it has two asymmetrical centers and is supplied as a mixture of four isomers. This likely underlies its multiple actions, including, in addition to  $\alpha_1$ - and  $\beta$ -blockade,  $\beta_2$ -selective intrinsic sympathomimetic activity, direct vasodilatation (independent of its  $\alpha_1$ -blocking properties and/or  $\beta_2$  agonist properties), and blockade of the neuronal uptake of norepinephrine. This latter effect may be undesirable since it may limit the antihypertensive efficacy of labetalol by increasing the concentration of norepinephrine available to complete with its  $\alpha$ - and  $\beta$ -blocking action.

$\alpha$ -Adrenergic antagonists block the  $\alpha$ -receptor-mediated effects of sympathetic amines by a direct interaction with the receptor.<sup>5</sup>

However, in few is this property specific and many naturally occurring and synthetic agents also block 5-hydroxytryptamine and dopamine receptors as well as inhibit the reuptake of catecholamines. The most widely used agents are those that selectively block only  $\alpha_1$ -receptors and, thus, do not excessively increase catecholamine secretion by also antagonizing  $\alpha_2$ -receptors involved in the feedback inhibition of neurotransmitter release.<sup>39</sup>

Prazosin, a quinazoline, is the prototype  $\alpha_1$ -selective antagonist and is a useful agent for the treatment of hypertension. Its antihypertensive action is due to vasodilation resulting from competitive blockade of post-junctional  $\alpha_1$ -receptors. A further and potentially beneficial effect of this class of drugs is the ability to

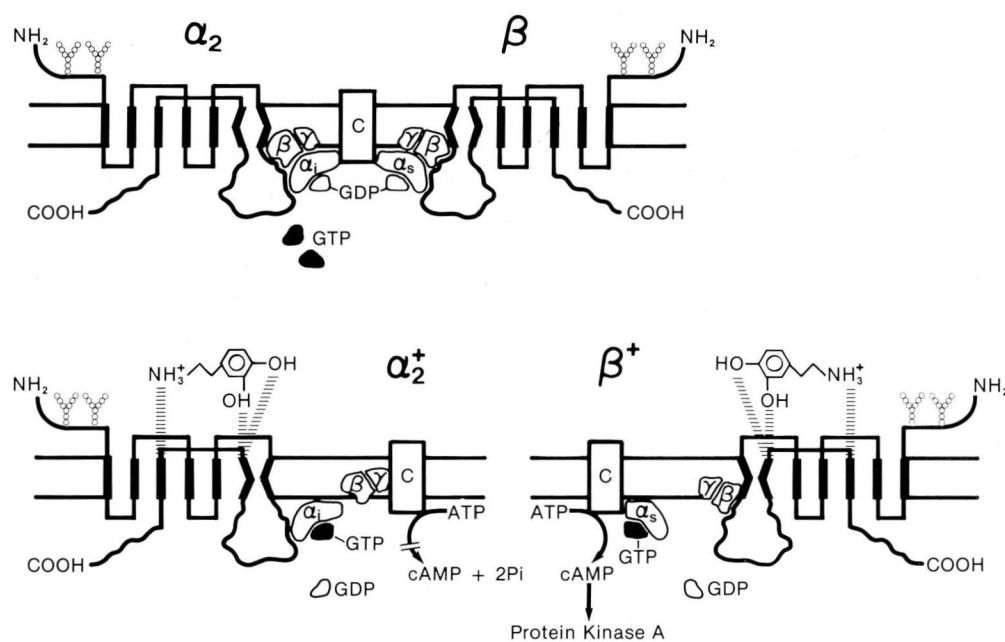


FIGURE 3. Signal transduction by  $\alpha_2$ - and  $\beta$ -adrenergic receptors. The receptors are represented as single polypeptides that span the plasma membrane (horizontal lines) seven times, with the amino terminus ( $NH_2$ ) located extracellularly and the carboxyl terminus ( $COOH$ ) located intracellularly. The third intracellular loop connecting the fifth and sixth transmembrane spanning regions is depicted as interacting with the receptor coupled G-proteins, each of which consists of three subunits ( $\alpha$ ,  $\beta$ , and  $\gamma$ ). The transmembrane spanning regions are most likely  $\alpha$ -helical in structure, and some of these regions may be kinked, as shown, because they contain prolyl residues. In the basal state (upper panel), GDP is bound to the  $\alpha$ -subunits of the G-proteins. Activation of the receptors ( $\alpha^+$ ,  $\beta^+$ ) by an agonist (lower panel) such as norepinephrine causes an alteration in the conformation of the receptor and its G-protein leading to dissociation of the GDP and association of GTP by the  $\alpha$ -subunits. As a result, the  $\alpha$ -subunits dissociate from the  $\beta\gamma$  subunits. With the  $\beta$ -adrenergic receptor, the resulting GTP-activated  $\alpha_s$  subunit then activates the membrane-bound enzyme adenylyl cyclase (C) leading to cAMP formation. The cAMP thus formed activates protein kinase A. With the  $\alpha_2$ -adrenergic receptor, the  $\beta\gamma$  subunits released with receptor activation prevents  $\alpha_{SGTP}$  stimulation of C and thus inhibits cAMP formation.

favorably alter lipoprotein metabolism, although the mechanism underlying this action remains unclear.<sup>40</sup>

Phenoxybenzamine is a moderately selective  $\alpha_1$ -blocking agent that lacks agonistic activity, but its specificity for  $\alpha$ -receptors is low since it blocks a variety of other receptors and also inhibits the neuronal uptake of norepinephrine. It is a haloalkylamine related to nitrogen mustards and irreversibly inactivates  $\alpha$ -receptors. Because of this property, it remains the antihypertensive drug of choice for the preoperative management of pheochromocytoma and for the prolonged management of cases not amenable to surgery, since in this condition hypertension is due to marked and excessive secretion of catecholamines from the tumor. Phenoxybenzamine and the weakly selective, competitive  $\alpha_1$ -antagonist, phen-

tolamine, are also used to control acute hypertensive episodes due to sympathomimetic overdosage or abuse, and to catecholamine excess associated with the use of monoamine oxidase inhibitors.

$\alpha$ -Adrenergic blockers are also of some use in the treatment of vasospastic disorders, such as Raynaud's phenomenon and acrocyanosis, for improving blood flow in shock states, and for reducing afterload in patients with congestive heart failure and pulmonary edema. Additionally,  $\alpha_1$ -selective agents may be of use in treating certain cardiac arrhythmias and in ameliorating urinary retention in patients with prostatic hypertrophy.

---

#### EVIDENCE FOR ADDITIONAL ADRENERGIC RECEPTOR SUBTYPES

---

There is considerable evidence for the existence of additional adrenergic receptor subtypes based on a variety of findings, including differences in the rank order of potency of adrenergic ligands for some responses, differences in the coupled effector pathways, and variations in the biophysical properties of the receptor proteins. With the  $\beta$ -adrenergic system, it has been postulated that an atypical receptor exists in guinea pig ileum, rat fat cells, and soleus muscle. In these tissues, the rank order of potency of agonists differs from that obtained at  $\beta_1$ - or  $\beta_2$ -adrenergic receptors, and novel compounds have been developed that selectively stimulate this atypical or  $\beta_3$ -adrenergic receptor.<sup>17</sup>

Using molecular biology techniques, the amino acid sequence of the  $\beta_3$ -adrenergic receptor has been deduced from the nucleotide sequence of the human gene.<sup>18</sup> Like other  $\beta$ -adrenergic receptors, the  $\beta_3$ -receptor promotes cAMP accumulation. However, its predominant role appears to be in the regulation of energy balance and metabolic rate by influencing thermogenesis.

Several distinct  $\alpha_2$ -adrenergic receptor subtypes have been identified, and the primary structures of two of these subtypes have been determined.<sup>16,19</sup> One of these subtypes, the  $\alpha_{2A}$  receptor, is found on human platelets as well as on the human intestinal epithelial cell line (HT-29) and in porcine lung.<sup>20</sup> The  $\alpha_{2B}$ -subtype has been cloned from kidney, and it is also found in rat lung and the opossum kidney cell line (OK). These two subtypes can be distinguished on the basis of their ligand binding properties. Although these subtypes are highly homologous, differences do exist in their amino acid sequences, particularly in the region of the third intracellular loops that interact with coupled guanine nucleotide-binding proteins.<sup>16,19</sup> This suggests that these two subtypes utilize different effector systems for intracellular signalling. Indeed, the finding that the platelet

$\alpha_{2A}$ -receptor mediates  $\text{Na}^+\text{-H}^+$  antiporter activity,<sup>21</sup> whereas that in HT-29 cells does not,<sup>22</sup> indicates that additional heterogeneity may exist, even within the  $\alpha_{2A}$  subclassification. Apart from  $\alpha_{2A}$  and  $\alpha_{2B}$ -receptor-subtypes, a non-glycosylated form of an  $\alpha_{2B}$ -like receptor has recently been described in neonatal rat lung.<sup>23</sup> Finally, there is evidence for a binding site (the imidazole/guanidinium receptive site) that recognizes some  $\alpha_2$ -adrenergic ligands, but is clearly distinct from  $\alpha_2$ -adrenergic receptors.<sup>24</sup>

$\alpha_1$ -Adrenergic receptors are also most likely comprised of a group of several homologous but distinct proteins. Two subtypes, the  $\alpha_{1A}$  and  $\alpha_{1B}$ -receptors, have been identified on the basis of biochemical and pharmacologic studies,<sup>25,26</sup> and the hamster smooth muscle receptor, which is of the  $\alpha_{1B}$  subtype, has been cloned.<sup>15</sup> These subtypes differ with respect to their affinities for certain agonist and antagonist ligands; their ability to be irreversibly inactivated by some alkylating agents<sup>25</sup>; their sensitivity to photoaffinity labelling<sup>27</sup>; and in the "tightness" of their coupling to effector responses, as evidenced by the presence or absence of spare receptors.<sup>26</sup> In addition to  $\alpha_{1A}$ - and  $\alpha_{1B}$ -receptors, it is likely that other subtypes exist, since there are a variety of signal transduction pathways for these receptors (vide infra).

---

#### SIGNAL TRANSDUCTION MECHANISMS

---

All adrenergic receptors (Figures 3 and 4) are coupled to their effectors by one or other of a family of proteins that bind guanine nucleotides (G-proteins).<sup>1,6,7,25</sup> These G-proteins consist of three distinct subunits,  $\alpha$ ,  $\beta$ , and  $\gamma$ .<sup>28</sup> The  $\alpha$ -subunit is the most discriminant and, in most cases, is the effector subunit for receptor signalling. Receptor activation commences with adrenergic agonist recognition and binding. This leads to an alteration in the conformation of the receptor and its coupled G-protein, and induces  $\alpha$ -subunit exchange of guanosine triphosphate (GTP) for guanosine diphosphate (GDP). In the GTP-bound form, the  $\alpha$ -subunit of the heterotrimeric G-protein complex dissociates from the  $\beta\gamma$  subunits and either activates a coupled effector enzyme leading to the generation of a second messenger or directly modulates ion channels. These responses are terminated by an enzymic property of the G-protein  $\alpha$ -subunit. This GTPase activity hydrolyzes GTP to GDP, allowing reassociation of the  $\alpha$ -subunit (in the GDP-bound form) with the  $\beta\gamma$  subunits.

$\beta$ -Adrenergic receptors activate the membrane-bound enzyme, adenylate cyclase, via the G-protein,  $G_s$ , resulting in the hydrolysis of ATP and the formation of cAMP



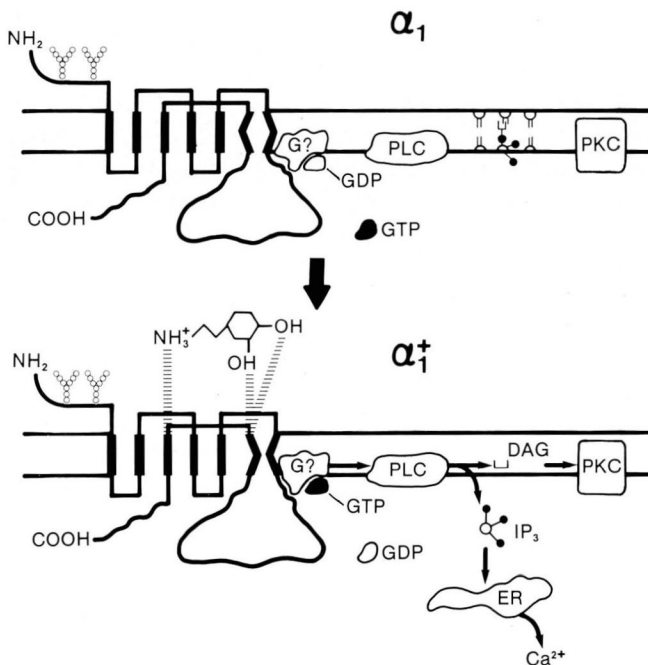


FIGURE 4. Signal transduction by  $\alpha_1$ -adrenergic receptors coupled to phospholipase C (PLC) activation. G?, the G-protein coupling  $\alpha_1$ -adrenergic receptor activation of PLC, the nature of which remains unclear; DAG, diacylglycerol; IP<sub>3</sub>, inositol-1,4,5-trisphosphate; PKC, protein kinase C; ER, endoplasmic reticulum.

(Figure 3). The resulting increase in intracellular cAMP is then available for activation of protein kinase A, leading to the phosphorylation of various effector proteins, including ion channels. Additionally, there is now compelling evidence that the GTP-bound form of the G<sub>s</sub>  $\alpha$ -subunit ( $\alpha_{sGTP}$ ), resulting from  $\beta$ -adrenergic receptor stimulation, may itself act as the second messenger and directly gate ion channels in a membrane-delimited process.<sup>29</sup> In myocardium, for example,  $\alpha_{sGTP}$  activates Ca<sup>2+</sup> channels, and it is this mechanism, rather than the generation of cAMP, that most likely underlies the ability of the cardiac sympathetic nerves, acting via  $\beta$ -adrenergic receptors, to regulate heart rate from beat to beat. Direct inhibition of sodium channels by  $\alpha_{sGTP}$  has also been demonstrated, particularly when the myocardial membrane is depolarized. In the setting of myocardial ischemia, which is associated with membrane depolarization and an increase in catecholamine release, such an effect may promote the development of cardiac arrhythmias by further inhibiting myocardial sodium currents.

$\alpha_2$ -Adrenergic receptor occupancy limits cAMP formation (Figure 3) by inhibiting adenylyl cyclase activity.<sup>21</sup> This process involves activation of G<sub>i</sub>, a G-protein with an  $\alpha$  subunit distinct from that of G<sub>s</sub>. Again, in this case, the  $\alpha$ -subunit is activated by the exchange of GTP for GDP, which promotes dissociation of the  $\beta\gamma$  subunits. Since G<sub>i</sub> is present in stoichiometric excess over G<sub>s</sub>, and since the  $\beta\gamma$  subunits of G<sub>s</sub> and G<sub>i</sub> are promiscuous, the  $\beta\gamma$  subunits released by  $\alpha_2$ -adrenergic receptor activation are available to complex with  $\alpha_s$  and, thus to prevent its activation of adenylyl cyclase. Thus, with  $\alpha_2$ -adrenergic receptor-mediated inhibition of adenylyl cyclase, it is the  $\beta\gamma$  subunits complex, rather than the  $\alpha$ -subunit, that mediates the receptor response. However, in this case, the  $\beta\gamma$  effector-complex acts in a negative manner to prevent adenylyl cyclase activation, rather than directly inhibiting the enzyme. It should, however, be noted that there is also evidence against the exclusive dependence of  $\alpha_2$ -receptor-mediated adenylyl cyclase inhibition on a  $\beta\gamma$  inhibitory action, and several other mechanisms have been proposed. These include a direct inhibition of adenylyl cyclase by activated  $\alpha_i$ ; competition by  $\alpha_i$  with  $\alpha_s$  for the active site on adenylyl cyclase, with  $\alpha_i$  having no (or less) cyclase activating activity, and direct inhibition of adenylyl cyclase by  $\beta\gamma$  complexes.

Although inhibition of adenylyl cyclase appears to be the primary signalling mechanism for some  $\alpha_2$ -adrenergic receptor-mediated events, such as inhibition of lipolysis in fat cells, it does not account for many other effector-cell responses. In these instances, alterations in intracellular pH produced by enhanced Na<sup>+</sup>/H<sup>+</sup> exchange or in intracellular ion concentrations produced by Ca<sup>+</sup> or K<sup>+</sup> channel modulation, may be the primary signalling mechanisms associated with  $\alpha_2$ -adrenergic receptor activation.<sup>21</sup> For example, activation of Na<sup>+</sup>/H<sup>+</sup> exchange by  $\alpha_2$ -adrenergic receptor occupancy has been implicated as the most proximal receptor-coupled response in renal proximal tubule cells and platelets and in the NG-10815 cell line. Thus, the signalling pathway for agonist-induced dense granule secretion from platelets appears to involve initial activation of Na<sup>+</sup>/H<sup>+</sup> exchange and intracellular alkalinization. This, in turn, leads to the activation of the membrane-bound enzyme, phospholipase A<sub>2</sub>. The arachidonic acid liberated by this enzyme, after conversion via cyclooxygenase to endoperoxides or thromboxane A<sub>2</sub>, is then responsible for the activation of phospholipase C and the liberation of the putative mediators of platelet secretion, inositol phosphates and diacylglycerol. In neuronal preparations in which  $\alpha_2$ -adrenergic receptor activation inhibits neu-

rotransmitter release, the primary signalling mechanism appears to be an alteration in a  $\text{Ca}^{2+}$  conductance, a  $\text{K}^+$  conductance, or both. This results in hyperpolarization of the plasma membrane potential and subsequent inhibition of neuronal firing, and/or limits the availability of  $\text{Ca}^{2+}$  for electrosecretory coupling.

These responses also appear to involve a G-protein, although via a direct effect involving the activated GTP-bound  $\alpha$ -subunits, rather than by an indirect inhibitory action of dissociated  $\beta\gamma$  complexes. Interestingly, in cells of neuronal origin, the receptor-coupled inhibition of neurotransmitter release appears to involve  $\text{G}_o$ , a GTP-binding protein distinct from  $\text{G}_i$ .<sup>21</sup> Receptor coupling to either  $\text{G}_i$  or  $\text{G}_o$  can be perturbed by an endotoxin produced by *Bordetella pertussis* that catalyzes the covalent modification of these G-proteins by ADP ribosylation. Pertussis toxin treatment, thus, is a convenient and widely used tool for investigating the mechanisms of receptor signalling, and additionally allows G-proteins, such as  $\text{G}_i$  and  $\text{G}_o$  to be radiolabelled when [<sup>32</sup>P] NAD is included as a donor in the ADP-ribosylation reaction.

A G-protein-coupled pathway is also most likely involved in  $\alpha_1$ -adrenergic receptor signalling, although less is known about this complex, heterogeneous receptor system.<sup>1,25</sup> In most cases,  $\alpha_1$ -receptor occupancy leads to the activation of a phospholipase C (PLC), which hydrolyzes phosphatidylinositol-4, 5-bisphosphate ( $\text{PIP}_2$ ), a minor component of the plasma membrane phospholipid pool, to yield inositol 1, 4, 5-trisphosphate ( $\text{IP}_3$ ) and diacylglycerol (DAG) [Figure 4]). These second messengers promote intracellular calcium mobilization and protein kinase C activation, respectively. Receptor-mediated activation of PLC in most instances is a pertussis toxin-insensitive process. Nevertheless, a G-protein is most likely involved since the receptor response can be mimicked by direct activation with guanine nucleotides. In some tissues, such as vascular smooth muscle, receptor-activated calcium mobilization does not involve PLC activation and  $\text{PIP}_2$  hydrolysis; rather, receptor occupancy appears to regulate voltage-operated  $\text{Ca}^{2+}$  channels.<sup>26</sup> Whether a G-protein couples this receptor-mediated response is not clear. There is, however, considerable evidence to indicate that these

PLC-dependent and -independent responses are mediated by different subtypes of the  $\alpha_1$ -receptor, viz  $\alpha_{1B}$  and  $\alpha_{1A}$ , respectively.

A further complexity in  $\alpha_1$ -adrenergic receptor signalling is the finding that, in some cells, receptor occupancy can activate both PLC and phospholipase  $\text{A}_2$ , the former response being pertussis toxin-insensitive and the latter—leading to arachidonic acid formation—being pertussis toxin-sensitive.<sup>30</sup> Thus,  $\alpha_1$ -adrenergic signalling may involve at least two distinct G-protein intermediaries. Finally, there are also instances where  $\alpha_1$ -adrenergic receptor stimulation leads to cAMP generation, to inhibition of a transient outward  $\text{K}^+$  current, and to phospholipase D activation. The latter process results in the hydrolysis of phosphatidylcholine to phosphatidic acid and/or diacylglycerol. Whether these responses are mediated by yet different  $\alpha_1$ -adrenergic receptor subtypes remains unclear.

---

#### SUMMARY

Adrenergic receptors are a diverse but structurally related group of plasma membrane proteins that mediate the actions of the sympathetic nervous system. Much has been learned, of late, concerning the molecular mechanism by which they selectively recognize and bind catecholamines and by which they mediate transmembrane signalling. Although a wide array of therapeutically useful agents are available that activate or block these receptors, further insights into their structure, function and diversity, as well as the molecular mechanisms controlling their expression, are likely to be of critical importance for the further development of newer drugs that produce unique and more specific therapeutic effects. Additionally, a more detailed understanding of the receptor domains involved in ligand binding and in receptor-effector coupling should provide a more rational and less empiric basis for future drug design.

---

#### ACKNOWLEDGMENT

I am grateful to Ms. J. Holl and Ms. D. Strekal for editorial assistance. Studies from the author's laboratory were supported in part by NIH Grant NS19583.

---

#### REFERENCES

1. Graham RM, Lanier SM. Identification and characterization of  $\alpha$ -adrenergic receptors. [In] *The Heart and Cardiovascular System*, edited by H.A. Fozzard et al., Raven Press, NY, 1986, Chapter 50, pp 1059-1095.
2. Longabaugh, JP, Vatner DE, Homcy CJ. The  $\beta$ -adrenergic/adenylate cyclase system. [In] *The Heart and Cardiovascular System*, edited by H.A. Fozzard et al., Raven Press, NY, 1986, Chapter 51, pp 1097-1118.
3. Homcy CJ, Graham RM. Molecular characterization of adrenergic receptors. *Circ Res* 1985; 56:635-650.
4. Weiner N. [In] *The Pharmacologic Basis of therapeutics*, edited by Gilman et al., Macmillan, NY, 7th Edition, Chapters 8 and 9, 1985.

5. Graham RM. The physiology and pharmacology of  $\alpha$ - and  $\beta$ -blockade. *Cardiovasc Med (Special Suppl. April)* 1981; 7-22.
6. Strader CD, Sigal IS, Dixon RA. Structural basis of  $\beta$ -adrenergic receptor function. *FASEB J* 1989; 3:1825-1832.
7. Dohlman HG, Caron MG, Lefkowitz RJ. A family of receptors coupled to guanine nucleotide regulating proteins. *Biochemistry* 1987; 26:2657-2664.
8. Ahlquist RP. A study of the adrenotropic receptors. *A J Physiol* 1948; 153:586-600.
9. Lands AM, Arnold A, McAuliff JP, Luduena FP, Brown TG. Differentiation of receptor systems activated by sympathomimetic amines. *Nature* 1967; 214:597-598.
10. Furchgott RF. The pharmacological differentiation of adrenergic receptors. *Ann NY Acad Sci* 1967; 139:553-570.
11. Slater IH, Powell CE. Blockade of adrenergic inhibitory receptor sites by 1-(3',4'-dichlorophenyl)-2-isopropyl-aminoethanol hydrochloride. *Fed Proc* 1957; 16:336.
12. Minneman KP, Pittman RN, Molinoff PB.  $\beta$  Adrenergic receptor subtypes: properties, distribution and regulation. *Ann Rev Neurosci* 1981; 4:419-461.
13. Langer SZ. Presynaptic regulation of catecholamine release. *Br J Pharmacol* 1974; 23:1793-1800.
14. Berthelsen S, Pettinger WA. A functional basis for classification of  $\alpha$ -adrenergic receptors. *Life Sci* 1977; 21:595-606.
15. Cotecchia S, Schwinn DA, Randall RR, Lefkowitz RJ, Caron ME, Kobilka BK. Molecular cloning and expression of the cDNA for the hamster  $\alpha 1$ -adrenergic receptor. *Proc Natl Acad Sci USA* 1988; 85:7159-7163.
16. Kobilka BK, Matsui H, Kobilka TS, et al. Cloning sequencing and expression of the gene coding for the human platelet  $\alpha 2$ -adrenergic receptor. *Science* 1987; 238:650-656.
17. Arch JRS, Ainsworth AT, Cawthorne MA, et al. Atypical  $\beta$ -adrenoceptor on brown adipocytes as target for anti-obesity drugs. *Nature* 1984; 309:163-170.
18. Emorine LJ, Marullo S, Briand-Sutren M-M, Patey G, Tate Delavier-Klutchko C, Strosberg D. Molecular characterization of the human  $\beta 3$ -adrenergic receptor. *Science* 1989; 245:1118-1121.
19. Regan JW, Kobilka TS, Yang-Feng TL, Caron MG, Lefkowitz RJ, Kobilka BK. Cloning and expression of a human kidney cDNA for an  $\alpha 2$ -adrenergic receptor subtype. *Proc Natl Acad Sci USA* 1988; 85:6301-6305.
20. Bylund JB.  $\alpha 2$ -Adrenergic receptors. A historical perspective. [In] *The  $\alpha 2$ -Adrenergic Receptor*, edited by Limbind LE. Humana Press, NJ, 1988, Chapter 1, pp 1-13, 1988.
21. Isom LL, Limbind LE. What happens next? A hypothesis linking the biochemical and electrophysiological sequelae of  $\alpha 2$ -adrenergic receptor occupancy with diverse receptor-mediated physiological effects. [In] *The  $\alpha 2$ -Adrenergic Receptor*, edited by LE Limbind. Humana Press, NJ, pp 323-363, 1988.
22. Cantiello HF, Lanier SM.  $\alpha$ -Adrenergic receptors and the NA<sup>+</sup>/H<sup>+</sup> exchanger in the intestinal epithelial cell line, HT-29. *J. Biol Chem* 1989; 264:16000-16007.
23. Lanier SM, Homcy CJ, Patenaude C, Graham RM. Identification of structurally distinct  $\alpha 2$ -adrenergic receptors. *J Biol Chem* 1988; 263:14491-14496.
24. Parini A, Coupry I, Graham RM, Uzielli I, Atlas D, Lanier SM. Characterization of an imidazoline/guanidinium receptive site distinct. *J Biol Chem* 1989; 264:11874-11878.
25. Minneman KP.  $\alpha 1$ -Adrenergic receptor subtypes, inositol phosphates, and source of cell Ca<sup>2+</sup>. *Pharmacol Rev* 1988; 40:87-119.
26. Tsujimoto G, Tsujimoto A, Suzuki E, Hashimoto K. Glycogen phosphorylase activation by two different  $\alpha 1$ -adrenergic receptor subtypes: methoxamine selectively stimulates a putative  $\alpha 1$ -adrenergic receptor subtype ( $\alpha 1a$ ) that couples with Ca<sup>2+</sup> influx. *Mol Pharmacol* 1989; 36:166-176.
27. Terman BI, Riek RP, Grodski A, Hess H-J, Graham RM. Identification and structural characterization of  $\alpha 1$ -adrenergic receptor subtypes. *Mol Pharmacol* 1990; 37:526-534.
28. Gilman AG. G proteins: Transducers of receptor-generated signals. *Ann Rev Biochem* 1987; 56:615-649.
29. Brown AM, Birnbaumer. Direct G protein gating of ion channels. *Am J Physiol* 1988; 254:H401-H410.
30. Birch RM, Luini A, Axelrod J. Phospholipase A2 and phospholipase C are activated by distinct GTP-binding proteins in response to  $\alpha 1$ -adrenergic stimulation in FRTL5 thyroid cells. *Proc Natl Acad Sci USA* 1986; 83:7201-7205.
31. Dixon RAF, Sigal IS, Candelore MR, et al. Structural features required for ligand binding to the  $\beta$ -adrenergic receptor. *Embo J* 1987; 6:3269-3275.
32. Kamik SS, Sakmar TP, Chen HB, Khorana HG. Cysteine residues 110 and 187 are essential for the formation of correct structure in bovine rhodopsin. *Proc Natl Acad Sci USA* 1988; 85:8459-8463.
33. Kamik SS, Khorana HG (Personal Communication).
34. Sakmar TP, Franke RR, Khorana HG. Glutamic acid-113 serves as the retinylidene Schiff base counterion in bovine rhodopsin. *Proc Natl Acad Sci USA* 1989; 86:8309-8313.
35. Bouvier MW, Hauschorff A, DeBasi A, et al. Removal of phosphorylation sites from the  $\beta$ -adrenergic receptor delays the onset of agonist-promoted desensitization. *Nature* 1988; 333:370-373.
36. Clark RB, Friedman JE, Dixon RAF, Strader CD. Identification of a specific site required for rapid heterologous desensitization of the  $\beta$ -adrenergic receptor by cAMP-dependent protein kinase. *Mol Pharmacol* 1989; 36:343-348.
37. Black JW, Crowther AF, Smith LH, Shanks RG, Dornhorst AC. A new adrenergic  $\beta$ -receptor antagonist. *Lancet* 1964; 1:1080-1084.
38. MacCarthy EP, Bloomfield SS. Labetalol: a review of its pharmacology, pharmacokinetics, clinical uses and adverse effects. *Pharmacotherapy* 1983; 3:193-217.
39. Graham RM. Selective  $\alpha 1$ -adrenergic antagonists: therapeutically relevant antihypertensive agents. *Am J Cardiol* 1984; 53:16A-20A.
40. Graham RM. Selective  $\alpha 1$ -adrenergic blockade, lipids and coronary heart disease risk: considerations in the treatment of mild hypertension. *Am J Med* 1989; 87:53S-56S.