Recent Advances in Knowledge About Beta-Adrenergic Receptors: Application to Clinical Cardiology

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The properties of beta-adrenergic receptors in the cardiovascular system have been studied in the past by two experimental approaches, which can be termed pharmacologic and biochemical. In the pharmacologic approach, the nature of a drug interaction with receptors is deduced from alterations in the physiologic properties of the tissue caused by administration of various concentrations of the drug. Many important concepts about beta-adrenergic receptors have come from such indirect pharmacologic studies. The biochemical approach directly assesses the interaction of drugs with beta-adrenergic receptors by studying the binding of radiolabeled antagonists and agonists with the receptor. This relatively new approach has provided a large amount of new information regarding the intrinsic properties of beta-adrenergic receptors and modification of these properties by physiologic stresses, administration of drugs and disease states. The biochemical approach has also been applied recently to the study of beta-adrenergic receptors in human beings. In the future, substantial clinically relevant new information regarding the nature of beta-adrenergic receptors in physiologic and pathologic conditions should result from application of a combination of the biochemical and physiologic approaches to studies in human beings.

The properties of receptors for hormones, neurotransmitters and drugs have come to be analyzed by two distinct experimental approaches, which generally can be termed pharmacologic and biochemical (1). The pharmacologic approach utilizes intact functioning organs or tissues which, under specified experimental conditions, are exposed to a series of drugs for the generation of concentration-response data. The response measured is a change in a physiologic characteristic of the organ or tissue under study. The data are then analyzed and interpreted utilizing mathematical models of drug-receptor interactions (1). Since the original definition of beta-adrenergic receptors developed by Ahlquist (2) in 1948 with the pharmacologic approach, much important information regarding beta-adrenergic receptors has been gleaned from studies with functioning tissues. For example, the different chemical domains on catecholamine molecules which determine the affinity of beta-adrenergic receptors for these compounds as well as the intrinsic pharmacologic activity of the amines were distinguished by the pharmacologic approach (3). The classification of beta-adrenergic receptors into two subtypes, designated beta_1 and beta_2, was originally accomplished by detailed concentration-response studies with intact tissues (4).

Although many such important concepts about beta-adrenergic receptors have been deduced from pharmacologic studies, the development and application of the biochemical approach to the study of beta-adrenergic receptors has initiated a new era of discovery concerning the intrinsic properties of these receptors. The technology for the biochemical approach was first developed and validated only in 1974, but the discoveries based on this approach already have been voluminous. These recent discoveries dealt not only with the fundamental biochemistry and molecular pharmacology of receptors but also with clinically relevant findings in animal models of disease or drug therapy and in human beings. Thus, in a relatively short time the biochemical approach to the study of adrenergic receptors has been translated into application to clinical studies. This brief review focuses on new clinically relevant findings of particular interest to cardiologists which have resulted from the application of the biochemical approach, sometimes coupled with the pharmacologic approach, to the study of beta-ad-
rnergic receptors. More generalized discussions of recent discoveries regarding beta-adrenergic receptors can be found in numerous review articles (5–14).

**The biochemical approach.** This approach to the study of beta-adrenergic receptors is similar in principle to that of a variety of other hormone and neurotransmitter receptor assays and to many of the principles of radioimmunooassay (6,7,14,15). In all of these cases the interaction of a protein (receptor or antibody) with a ligand (drug or antigen) is examined. To allow detection and quantification of the interaction, the ligand is radiolabeled—thus the use of the terms radiolabeled ligand binding assays and radioimmunoassays in describing these approaches. In the case of receptor assays, a suitable organ or tissue source of receptors is obtained, generally the organ is homogenized, portions of the homogenate are then incubated with radiolabeled ligand, unbound radiolabeled ligand is separated from that which has interacted with receptors and the amount bound to receptors is quantified by counting radioactivity (14,15).

To verify that the radiolabeled ligand is bound to a receptor, certain criteria must be fulfilled (5–15). First, drugs should compete with the radiolabeled ligand for interaction with the receptor having the same rank order of potency they possess in intact tissue pharmacologic studies. Second, the biologically active stereoisomer should compete with the radiolabeled ligand for binding more potently than the inactive stereoisomer. Third, it should be possible to saturate all specific binding sites. Finally, only those organs or tissues that pharmacologic studies have shown to possess the receptors of interest should bind the radiolabeled ligand specifically. If these criteria of pharmacologic specificity, stereospecificity, saturability and appropriate localization of binding and certain other criteria are met, it can be assumed with reasonable confidence that the radiolabeled ligand is bound specifically to the receptor.

**Application to cardiac tissues.** Because the cardiovascular system is regulated by the sympathetic nervous system, many of the radiolabeled ligand binding studies of beta-adrenergic receptors have been performed in cardiac tissues. In fact, one of the first radiolabeled ligand binding studies of beta-adrenergic receptors was done in homogenates of canine heart (16). Subsequently, many studies of beta-adrenergic receptors derived from mammalian sources have been done on the heart (17–20). Although the results of many of these experiments in animals have been relevant to clinical medicine, it is desirable to test some of the conclusions from animal studies directly in human beings. To perform studies on beta-adrenergic receptors in human subjects, a readily available tissue source of receptors is required. The membranes of circulating leukocytes (polymorphonuclear leukocytes and lymphocytes) of human beings and certain animals contain beta-adrenergic receptors that can be studied directly with radiolabeled ligand binding assays (21). Accumulating evidence suggests that certain properties of the receptors in circulating leukocytes, such as density per unit surface area of membrane, reflect the properties of receptors in cardiovascular tissues. For example, administration of propranolol to rats caused an increase in density of beta-adrenergic receptors not only in lymphocytes but also in the ventricles and lungs of the same animals from which the lymphocytes were taken (22). In clinical studies, manipulations that altered the density of beta-adrenergic receptors in circulating leukocytes changed the heart rate response either to physiologic stress or to administration of catecholamines in a manner consistent with the conclusion that beta-adrenergic receptors in the heart change in a qualitatively similar fashion as those on leukocyte membranes. For example, in subjects who had infusion of isoproterenol for 30 to 60 minutes, beta-adrenergic receptor density in leukocytes was increased and the elevation in heart rate in response to a "pulse" infusion of isoproterenol was augmented (23). Subjects treated with propranolol had more leukocyte beta-adrenergic receptors and significantly greater increases in heart rate and heart rate-blood pressure product on standing (24). Persons whose diets were changed from a low to a high sodium content showed an elevation in leukocyte beta-adrenergic receptor density as well as an increase in sensitivity to the positive chronotropic effects of isoproterenol (25). Thus, it appears to be possible to examine the effects of diseases and drugs on the beta-adrenergic receptors of the cardiovascular system in human beings by studying the receptors on the membranes of circulating leukocytes. However, the validity of this approach will have to be verified carefully in each clinical setting in which it is utilized.

**Clinically Relevant Information From Radiolabeled Ligand Binding Studies of Beta-Adrenergic Receptors**

**Verification of Physiologic Principles**

**Beta, and beta, receptors.** The pharmacologic subclassification of beta-adrenergic receptors into the subtypes beta, and beta, has facilitated the development of subtype-specific agonists and antagonists. Thus, clinicians now have the option of using beta, selective antagonists to treat angina pectoris, certain cardiac arrhythmias, hypertension and other cardiovascular diseases with reduced concern about blocking beta,-receptors in bronchioles and peripheral arterioles. Similarly, bronchoconstriction can be treated with beta,-selective agonists with less concern about producing tachycardia from stimulation of beta,-receptors in the heart. Thus, the subclassification of beta-receptors is not only of pharmacologic interest, but also has important applications in the clinical management of patients. Although this subclassification was originally proposed on the basis of indirect pharmacologic studies (4), the true nature of subtypes of
beta-receptors was not revealed until the advent of radio-labeled ligand binding studies. This biochemical approach has confirmed directly the existence of two subtypes of beta-adrenergic receptors (20,26-28) and more recently suggested that the molecular properties of beta-1 and beta-2 receptors probably differ (29). Thus, the two subtypes of beta-adrenergic receptors appear to be distinct molecular entities (29). Biochemical studies, therefore, have verified at the molecular level the concept of beta-adrenergic receptor subtypes that was originally deduced only from indirect pharmacologic studies.

**Elucidation of changes in responsiveness to sympathetic stimulation.** Changes in responsiveness of animals or human beings to sympathetic stimulation after treatment with certain drugs, after certain surgical procedures such as denervation or in association with certain diseases have been observed by pharmacologists and clinicians for many years. With the development of biochemical assays of beta-adrenergic receptors, some of the mechanisms for these changes are being elucidated (11). It is now established that beta-adrenergic receptors in plasma membranes of cells are dynamic entities. Physiologic stresses and pharmacologic or pathologic perturbations can result in alterations in the number of beta-adrenergic receptor binding sites, changes in the affinity of receptors for catecholamines or modulation of the "coupling" of the receptor to its proximate biochemical effector enzyme, adenylate cyclase (11). As a broad generalization, the density of beta-adrenergic receptors on target organs appears to be inversely related to the magnitude of stimulation of those receptors by agonists (10-13). Thus, when sympathetic nervous system activity (either neural activity or circulating catecholamines, or both) is high, the density of beta-adrenergic receptors in target organs will tend to be low. This decrease in receptor number has been termed down regulation. Conversely, in situations where sympathetic nervous system activity is low, beta-adrenergic receptor density will tend to be high. This is called up regulation. Recent clinical studies have shown that such changes in beta-adrenergic receptor density can occur even in response to normal physiologic alterations in sympathetic activity (25). Normal volunteers were fed low and high sodium diets either to stimulate or to suppress sympathetic nervous system activity, which was monitored by measurement of plasma and urinary catecholamine levels (25). When sodium intake was increased from 10 to 400 mEq daily, a change that reduced sympathetic nervous system activity, the density of beta-adrenergic receptors on leukocyte membranes increased by approximately 50% (25). That the changes in leukocyte receptor density reflected receptor changes in the cardiovascular system was suggested by the observation that the subjects' sensitivity to the positive chronotropic effects of isoproterenol was also increased (25).

An additional observation was that the density of beta-adrenergic receptors on leukocytes of these normal subjects was inversely correlated with circulating and 24 hour urinary catecholamine levels (25). The levels of catecholamines in the blood and urine presumably reflected sympathetic nerve activity. Thus, the higher the level of sympathetic nervous system activity (the greater the concentration of catecholamines in the receptor milieu), the more down regulation had occurred (that is, the lower the density of receptors). These results provide a plausible explanation for the substantial interindividual variation in density of receptors on leukocytes of normal subjects (25,30). They also provide one possible explanation for the differences in response of individual patients to certain drugs (see later).

**Changes with aging.** Resting heart rate and cardiac output tend to diminish with age (31,32). It is reasonable to hypothesize that one possible mechanism for this trend is a change in beta-adrenergic receptors in the heart. It has been observed that elderly persons have decreased responsiveness to infusions of isoproterenol (33) and a lesser number of beta-adrenergic receptors on lymphocytes (34). A possible mechanism for reduced numbers of beta-adrenergic receptors with senescence is diminished rate of receptor synthesis. In senescent rats, the rate of synthesis of beta-adrenergic receptors in the heart was lower than that in young rats (35). It is thus possible that some of the cardiac functional and electrophysiologic changes that occur with aging are related to changes in beta-adrenergic receptor density in the heart, although this subject needs to be investigated further.

These are examples of concepts originally derived from indirect pharmacologic studies that have been verified or illuminated by application of direct biochemical studies of beta-adrenergic receptors. It is likely that other physiologic concepts will be verified directly as more studies are done in the future.

**Alterations of Cardiovascular Beta-Adrenergic Receptors in Disease**

**Cardiovascular disease.** Myocardial ischemia. A few studies have examined changes in beta-adrenergic receptors in the heart or blood vessels in cardiovascular disease. In a canine model of experimental myocardial ischemia (36), the number of beta-adrenergic receptors was found to be increased in the ischemic myocardium in association with a marked reduction in tissue norepinephrine content. The affinity of the receptors for the radiolabeled antagonist was unchanged. The density of muscarinic receptors in the same region was also unchanged. In a subsequent study (37), it was found that the responsiveness of the ischemic region to isoproterenol infusion was enhanced. In response to the catecholamine, tissue cyclic adenosine monophosphate (cAMP) levels were elevated more and phosphorylase was activated to a greater extent in the ischemic region, which had increased beta-adrenergic receptor density, than in the normally perfused control region (37). The mechanism for
the increase in beta-adrenergic receptor density, whether related to a direct effect of ischemia or secondary to the functional sympathetic denervation, is as yet unknown. Also, the pathophysiologic significance of these findings remains to be established. However, these are interesting results that show a relatively rapid, selective change in beta-adrenergic receptor density in ischemic myocardium. One could speculate that these changes may participate in the generation of cardiac arrhythmias during myocardial ischemia.

Ventricular arrhythmias. In a cat model of myocardial ischemia and reperfusion, a variety of experiments suggested that ventricular arrhythmias that occurred during coronary reperfusion were related to enhanced alpha-adrenergic responsiveness (38). In this model, radiolabeled ligand binding studies demonstrated a twofold increase in density of alpha_2-adrenergic receptors early after occlusion of the coronary artery and during the first few minutes of reperfusion (39). In this model, beta-adrenergic receptor density did not change (39). It was hypothesized that the change in alpha_2-adrenergic receptor density might be related to the enhanced alpha-adrenergic receptor responsiveness that appeared to cause the lethal arrhythmias during reperfusion. The mechanism for the change in alpha_2-adrenergic receptor density and the reason for the difference between the results in this study and those in the previously mentioned canine study are unknown. These experiments in dogs and cats were the first attempts to study changes in beta-adrenergic receptors in myocardial ischemia. The ultimate significance of these results and their relevance to coronary artery disease in human beings remains to be established. However, in view of the large incidence of coronary artery disease and the wide use of beta-adrenergic receptor blocking agents, which may modify receptor density in these patients, this should be an important and fruitful area of future research.

Heart failure. The properties of beta-adrenergic receptors in the failing heart have been assessed in ventricles taken from human heart transplant recipients (40). Receptors in ventricular tissue of transplant donors were studied as controls. It was found that the density of beta-adrenergic receptors in failing left ventricles was approximately 50% less than that in donor control nonfailing hearts (40). This reduction in receptor density was accompanied by a diminution in isoproterenol-mediated stimulation of adenylate cyclase activity and muscle contraction (40). It seems possible that this reduction in receptor density is related to an increase in circulating and neurally released catecholamines that is known to occur in response to heart failure (down regulation). The reduced beta-adrenergic receptor density may account, at least partially, for the diminished contractile function reserve seen in some forms of chronic heart failure.

Hypertension. The properties of beta-adrenergic receptors in cardiac and vascular tissues have been examined in the spontaneously hypertensive rat. This animal model of hypertension is known to have abnormally high efferent sympathetic nerve activity that presumably contributes to the hypertension (41-43). The density of beta-adrenergic receptors in the heart and vascular tissues of these animals was reduced modestly compared with the density in normotensive control animals (44,45). It seems likely that these changes in density are related to the increased stimulation of receptors by catecholamines as a result of the augmented nerve activity (down regulation). Whether these changes (in arterioles, for example) contribute to the hypertension remains unknown.

Significance of the changes in beta-adrenergic receptors. Beta-adrenergic receptors have now been studied in several cardiovascular diseases, and the receptors do seem to change in number in these pathologic states. Many more studies will have to be done, in both animal models and human beings, before the significance of these changes will be established. Questions that will have to be addressed include: 1) What is the general relevance of the findings from animal models? Are these results applicable to the clinical situation, or are they species-specific or even specific to the model used? 2) Are changes in receptors causally related to the cardiovascular diseases with which they are associated? Are they parallel events that contribute to the clinical manifestations of the disease? Or, are the changes merely parallel alterations without any pathophysiologic significance? 3) In conditions such as heart failure and hypertension which have multiple pathologic mechanisms and etiologies, are receptor changes similar or qualitatively different depending on the etiology or mechanism of the disease? 4) What will be the utility and validity of studying beta-adrenergic receptors in readily available blood elements (leukocytes) and, from these data, drawing conclusions about receptors in cardiac or vascular tissues?

Noncardiovascular disease. Hyperthyroidism. Some diseases that do not primarily occur in cardiac or vascular tissues are known to alter cardiac responsiveness to sympathetic nervous system stimulation. Perhaps one of the best known of such associations is that seen with altered thyroid state, particularly hyperthyroidism. Many of the clinical features of hyperthyroidism are suggestive of a hyperactive sympathetic nervous system. Certain of these features, such as tachycardia, can be readily treated with antisypathetic drugs such as reserpine or beta-adrenergic receptor blocking agents. However, many studies done over the years failed to demonstrate any clear evidence of excessive sympathetic nervous system activity; thus the role of the sympathetic nervous system in the clinical features of hyperthyroidism has remained controversial (46). Data from radiolabeled ligand binding assays have shed more light on this issue. Rats treated with exogenous thyroid hormone demonstrate the features of hyperthyroidism. Beta-adrenergic receptor density in the hearts taken from these animals is increased from 50 to 100% (47-49). Beta-adrenergic receptor density in membranes of cultured heart cells is also increased by
treatment with thyroid hormone (50). The biologic significance of these findings is supported by the observation that there is an augmented catecholamine-induced stimulation of cyclic AMP levels in the triiodothyronine-treated cells (50).

Clinical studies suggest that these observations are relevant to hyperthyroidism in human beings. Triiodothyronine was administered to normal volunteers for 1 week and leukocyte beta-adrenergic receptor density was measured (30). The number of receptors in leukocytes of treated subjects was approximately twofold greater than the number in placebo-treated control subjects. From these results, taken together with the animal thyrotoxicosis studies and the evidence that changes in leukocyte beta-adrenergic receptors reflect changes in the heart, it is reasonable to conclude that some of the cardiovascular manifestations of hyperthyroidism are due to an increase in the number of beta-adrenergic receptors in the heart.

Hypothyroidism. In contrast, patients with hypothyroidism appear to have reduced sympathetic nervous system activity. For example, a prominent clinical feature of hypothyroidism is bradycardia. The number of beta-adrenergic receptors in hearts from rats made hypothyroid with propylthiouracil was decreased modestly (49,51). The changes induced by hypothyroidism, however, appeared more complex and diffuse than the changes caused by hyperthyroidism. There was also a reduction in the number of alpha-adrenergic receptors and the activity of enzymes in sarcosome and sarcoplasmic reticulum was altered (49). Hypothyroidism also appeared to alter the interaction between the beta-adrenergic receptor and adenylate cyclase, suggesting decreased or impaired “coupling” of the receptor with the enzyme (51). This might lead to diminished cellular generation of cyclic AMP for a given magnitude of beta-receptor stimulation. Thus, in hypothyroidism there appears to be a reduction in the number of beta-adrenergic receptors and also diminished effectiveness in the translation of the effects of receptor stimulation to subsequent biochemical changes. These findings suggest a plausible mechanism for the bradycardia and diminished sympathetic responsiveness of the heart in patients with hypothyroidism.

In summary, patients with thyroid dysfunction manifest clinical signs suggestive of altered sympathetic nervous system activity or responsiveness to sympathetic stimulation. The changes in beta-adrenergic receptors in response to the altered thyroid state summarized here provide possible mechanisms for these clinical signs.

Alterations of Cardiovascular Beta-Adrenergic Receptors With Drug Therapy

Effects of antisympathetic agents. It is well established that sympathetic denervation leads to enhanced tissue responsiveness to catecholamines, a phenomenon called denervation hypersensitivity. In denervation produced surgically or chemically (for example, with 6-hydroxydopamine), the hypersensitivity could be attributed to changes on at least two levels, prejunctionally at the sympathetic nerve terminal that normally removes catecholamines from the neuroeffector junctions and postjunctionally on the effector cell. Radiolabeled ligand binding studies have shown that with denervation the density of beta-adrenergic receptors on effector cells increases, presumably because in this condition the concentration of catecholamines in the receptor milieu is low (52). Treatment with drugs that antagonize sympathetic nervous system activity also results in increased receptor density. Administration of guanethidine, which both blocks release of norepinephrine from nerve terminals and depletes norepinephrine stores, resulted in increased beta-adrenergic receptor density in hearts of rats (53).

Beta-receptor blockade. Blockade of beta-adrenergic receptors also leads to increases in receptor density (up to 40 times) (22,54). These observations are particularly important clinically because of the prominent role of beta-adrenergic receptor blockade in the treatment of cardiovascular disease. Some patients who discontinue treatment with beta-receptor blocking drugs exhibit clinical findings suggestive of a hyperactive sympathetic nervous system (55–58). Patients with coronary artery disease may occasionally develop unstable angina or even myocardial infarction after sudden withdrawal of beta-adrenergic receptor blocking agents (55–58). Results from clinical studies suggest that this syndrome may be related to hyper-responsiveness of beta-adrenergic receptors to stimulation by catecholamines. It has been shown that human beings will exhibit hyper-responsiveness to infused catecholamines for up to 13 days after terminating beta-receptor blockade (59,60). A possible explanation for this is the previously described observation that beta-adrenergic receptors are increased in the hearts of animals treated with beta-adrenergic receptor blocking drugs (22,54). That similar increases may also occur in patients treated with beta-receptor blocking drugs is suggested by the observation that human subjects treated with propranolol exhibited increases in the density of beta-adrenergic receptors on leukocytes (24,25). After withdrawal of propranolol the subjects also showed augmented orthostatic changes in heart rate and heart rate-blood pressure product, suggesting that the changes in leukocyte receptors reflected changes in cardiac receptors (24).

Mechanism of propranolol withdrawal syndrome. Increases in beta-adrenergic receptors in the heart may thus be one mechanism to explain the relatively unusual entity referred to as the propranolol withdrawal syndrome (58). It is unknown why this phenomenon occurs only in a minority of patients treated with beta-receptor blocking drugs. One explanation may be differences in the level of sympathetic nervous system activity before initiation of beta-receptor blockade. In a given subject with high levels of sympathetic
activity, and thus high concentrations of circulating catecholamines, beta-adrenergic receptor density would be expected to be greatly reduced (25). If this subject is then treated with a beta-receptor blocking agent, which would release the down regulation, beta-receptor density might be expected to increase substantially (25). If beta-receptor blockade is then suddenly withdrawn this subject, who still has a high level of sympathetic nervous system activity and now a relatively high density of receptors, might be a prime candidate for developing the propranolol withdrawal syndrome. Although some of these thoughts are speculative, this would appear to be an important clinical issue, particularly with the increasing use of beta-receptor blocking agents in patients with angina pectoris and after myocardial infarction.

**Beta-adrenergic receptor stimulation.** If beta-adrenergic receptors can be regulated in response to ambient concentrations of catecholamines under physiologic conditions, then administration of exogenous beta-agonists would be expected to diminish the number of receptors on effector organs and tissues. It has been clearly established in model systems that exposure of cells to catecholamines leads to a reduction in the number of beta-adrenergic receptors in the membranes of the cells (down regulation) (61). Additionally, it has been shown that after catecholamine exposure, the ability of beta-agonists to “couple” the receptor to adenylate cyclase is diminished (61–63). In intact cells, this agonist-induced down regulation and desensitization is manifested as a diminished physiologic response to catecholamines (63). Clinical studies suggest that receptor number can change in response to administration of exogenous beta-agonists to human beings. Whether or not “coupling” is altered by the administration of catecholamines remains to be established. Infusion of isoproterenol in normal subjects produced a biphasic effect on leukocyte beta-adrenergic receptor number, an initial increase in density that lasted only about 1 hour followed by a later reduction in density (23). When patients with heart failure were treated with the beta-agonist pibuterol, a decrease in beta-adrenergic receptor density in lymphocytes was found (64). Treatment of subjects with the noncatecholamine beta₂-adrenergic receptor agonist terbutaline decreased beta₂-adrenergic receptor density in polymorphonuclear leukocytes (65). These observations suggest one possible mechanism for the phenomenon of tachyphylaxis. With sustained administration of catecholamines in a clinical setting, the decreased responsiveness over time that is sometimes observed may be partially related to a reduction in the number of beta-adrenergic receptors.

**Summary and Conclusions**

Although the technology for biochemical assessment of beta-adrenergic receptors (developed largely in nonmammalian model systems) was introduced only in 1974, it is now applied widely to clinically relevant studies in animals and human beings. The studies discussed are only a beginning and much remains to be done utilizing radiolabeled ligand binding assays in clinical studies of health and disease. For example, what are the effects of age, exercise, physical conditioning, diet and posture on cardiovascular beta-adrenergic receptors? The diseases that have begun to be studied need more detailed examination. In the studies cited, the main emphasis has been on measuring receptor number and affinity of the receptor for antagonists. It is possible that in addition to or instead of changes in number, there are changes in receptor “coupling” to adenylate cyclase so that for a given amount of receptor stimulation, adenylate cyclase will be activated to a greater or lesser extent in certain disease states. Such changes can be detected only by careful analysis of agonist interaction with the receptor and the ability of the receptor to form guanine nucleotide-dependent high affinity intermediate states (11).

It must be remembered that the beta-adrenergic receptor is only one of a series of components in the receptor-cyclase-protein kinase system that mediate the intracellular effects of catecholamines (66–71). In addition to the receptor, there are adenylate cyclase, cyclic AMP-dependent protein kinase and phosphatases which remove phosphate from proteins phosphorylated by protein kinase (66–68,70,72,73). Calcium ions, the intracellular handling of which is altered by beta-agonists, also participate in the mediation of intracellular effects of catecholamines (68,69). One or more of these other components in the system may be altered in cardiovascular disease or in response to drug therapy; until each of the components is examined, mechanistic conclusions about disease or drug effects must be reached with caution. Finally, diseases must be examined more specifically in terms of pathophysiologic mechanisms or etiologic causes of the diseases. For example, heart failure can be caused by multiple factors and the role and nature of changes in beta-adrenergic receptors may vary depending on the cause. The study of other cardiac diseases such as cardiomyopathies and cardiac arrhythmias should reveal interesting results. Thus, continued application of radiolabeled ligand binding assays to clinical cardiology should yield substantial, interesting and important new information. **The use of these assays will also be important in cardiovascular pharmacology, both for understanding better the actions of existing drugs and for new drug development.** The effects of noncatecholamines and nonbeta-adrenergic receptor blocking agents on beta-adrenergic receptors can be assessed expeditiously with radiolabeled ligand binding studies (74,75). New chemicals, thought to act on beta-adrenergic receptors, can be efficiently screened and characterized with in vitro binding assays. These assays can reveal whether a compound is an agonist or antagonist, the intrinsic sympathomimetic activity of agonists, the affinity of beta-adrenergic receptors for the compound and the se-
lectivity of the compound for beta_1- and beta_2-receptors (14). Radiolabeled ligand binding studies of alpha-adrenergic and muscarinic receptors make it possible to determine if a compound has any activity on these other receptors of the autonomic nervous system as well as on beta-adrenergic receptors (14,74).

Radiolabeled ligand binding assays of beta-adrenergic receptors thus represent a major advance in the science of sympathetic nervous system physiology and pharmacology. The application of these assays should continue to make major contributions toward progress in cardiovascular physiology, pathology and pharmacology.

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References


