18 | The Renin–Angiotensin–Aldosterone System and Other Vasoactive Substances

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DRUG LIST

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Benzapril	212	Losartan	213
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THE RENIN–ANGIOTENSIN SYSTEM

The renin–angiotensin system is important for the regulation of vascular smooth muscle tone, fluid and electrolyte balance, and the growth of cardiac and vascular smooth muscle. A normally functioning renin–angiotensin system contributes to the routine control of arterial blood pressure. A variety of basic and clinical investigations have resulted in a broader understanding of the role of the renin–angiotensin system in the cardiovascular pathophysiology of hypertension, congestive heart failure, and more recently, atherosclerosis. Whether or not abnormal activity of the renin–angiotensin system contributes to the primary etiology of these diseases, *pharmacological inhibition of the renin–angiotensin system has proved to be a valuable therapeutic strategy in the treatment of hypertension and congestive heart failure.*

The classical renin–angiotensin system comprises a series of biochemical steps (Fig. 18.1) leading to the production of a family of structurally related peptides (e.g., angiotensin II, angiotensin III, and other smaller peptides with bioactivity). Sites for pharmacological intervention in this system include the enzymatic steps catalyzed by renin, angiotensin-converting enzyme (ACE), and angiotensin receptors that mediate a particular physiological response.

Renin

Renin is an enzyme that is synthesized and stored in the renal juxtaglomerular apparatus and that catalyzes the formation of a decapeptide, *angiotensin I,* from a plasma protein substrate. Renin has a narrow substrate specificity that is limited to a single peptide bond in angiotensinogen, a precursor of angiotensin I. Renin is considered to control the rate-limiting step in the ultimate production of angiotensin II. Control of renin secretion by the juxtaglomerular apparatus is important in determining the plasma renin concentration.
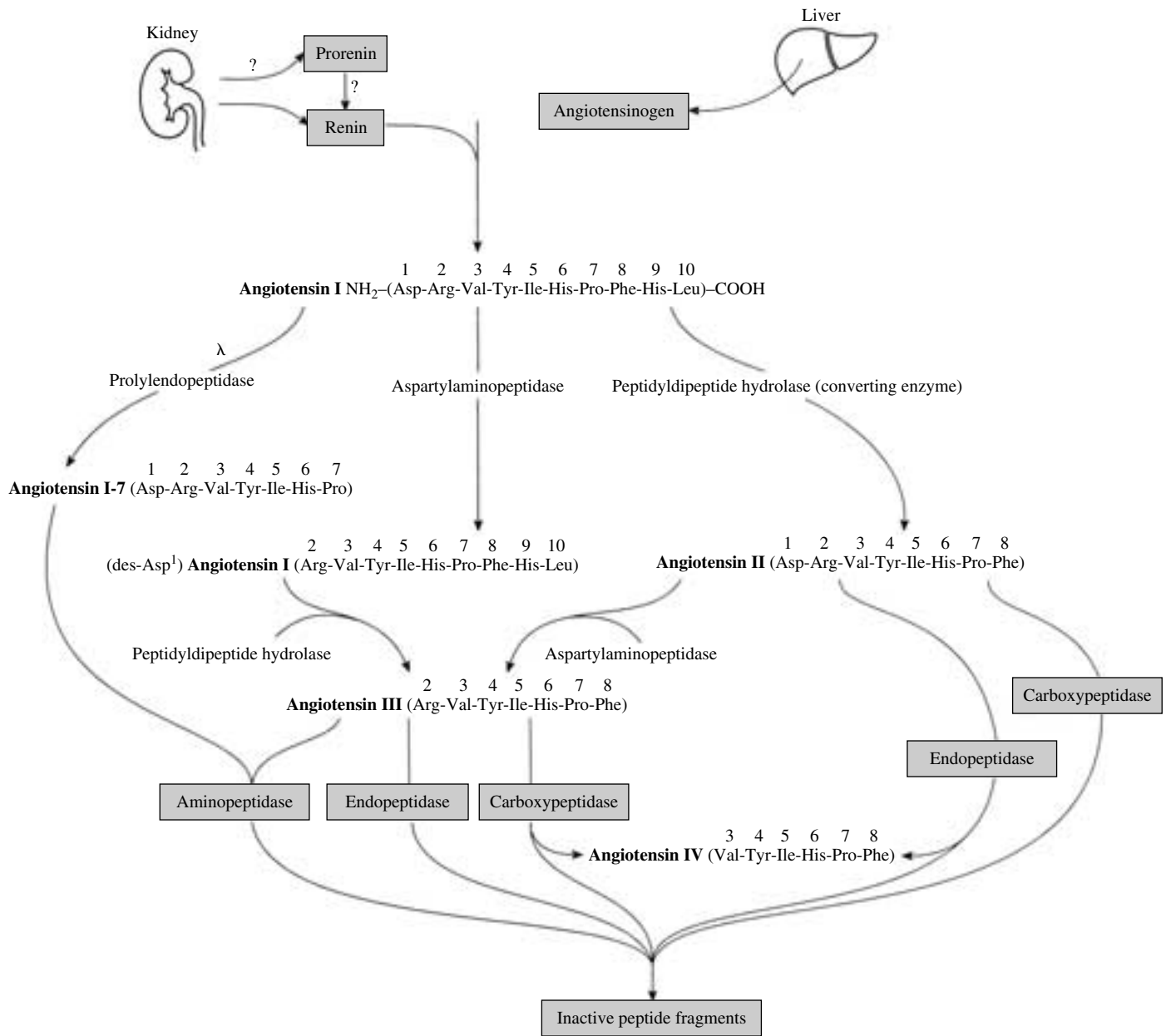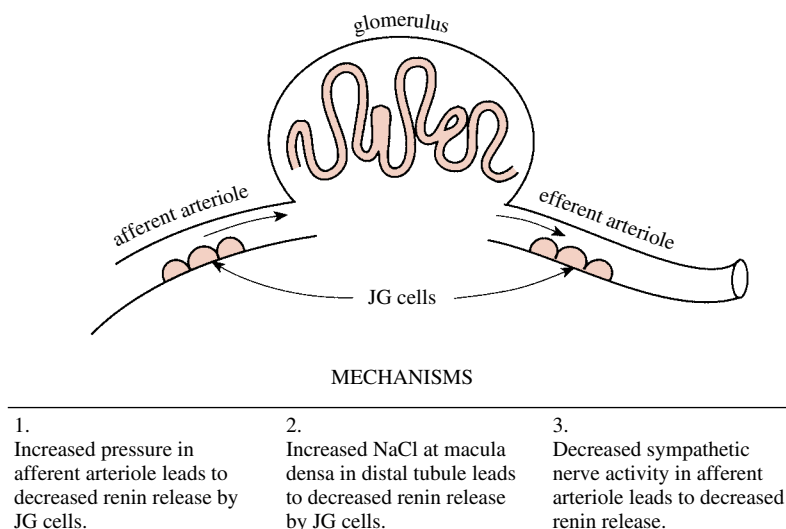
FIGURE 18.1
Synthesis and structures of the angiotensins.

Three generally accepted mechanisms are involved in the regulation of renin secretion (Fig. 18.2). The first depends on renal afferent arterioles that act as stretch receptors or baroreceptors. Increased intravascular pressure and increased volume in the afferent arteriole inhibits the release of renin. The second mechanism is the result of changes in the amount of filtered sodium that reaches the macula densa of the distal tubule. Plasma renin activity correlates inversely with dietary sodium intake. The third renin secretory control mechanism is neurogenic and involves the dense sympathetic innervation of the juxtaglomerular cells in the afferent arteriole; renin release is increased following activation of α_1-adrenoceptors by the neurotransmitter norepinephrine.

Angiotensin II, the primary end product of the renin–angiotensin system, acts on the juxtaglomerular cells to inhibit the release of renin; this process is therefore a negative feedback mechanism. The half-life of renin in the circulation is 10 to 30 minutes, with inactivation occurring primarily in the liver. Small amounts of renin are eliminated by the kidneys. Pure human renin

MECHANISMS

1.	2.	3.
Increased pressure in afferent arteriole leads to decreased renin release by JG cells.	Increased NaCl at macula densa in distal tubule leads to decreased renin release by JG cells.	Decreased sympathetic nerve activity in afferent arteriole leads to decreased renin release.

FIGURE 18.2

Mechanisms of regulation of renin secretion from the kidney. JG, juxtaglomerular.

has been used to develop specific inhibitors of the enzyme. Low-molecular-weight orally effective renin inhibitors are under development.

Angiotensinogen

Human plasma contains a glycoprotein called *angiotensinogen,* which serves as the only known substrate for renin. Angiotensinogen must undergo proteolysis before active portions of the protein are sufficiently unmasked to exert biological effects. Angiotensinogen is synthesized in many organs, including the liver, brain, kidney, and fat. Its gene transcription and plasma concentrations increase following treatment with adrenocorticotropic hormone (ACTH), glucocorticoids, thyroid hormone, and estrogens, as well as during pregnancy and inflammation and after nephrectomy. Angiotensinogen also has been found in large quantities in cerebrospinal and amniotic fluid. Mutations in the angiotensinogen gene have been reported to be linked to human hypertension.

Angiotensin-Converting Enzyme: A Peptidyl Dipeptide Hydrolase

Metabolism of angiotensinogen by renin produces the decapeptide *angiotensin I.* This relatively inactive peptide is acted on by a dipeptidase-converting enzyme to produce the very active octapeptide *angiotensin II.* In addition to converting enzyme, angiotensin I can be acted on by prolyl endopeptidase, an enzyme that removes the first amino acid to form angiotensin 1-7, a peptide primarily active in the brain. ACE has been

identified in vascular endothelial cells, epithelial cells of the proximal tubule and small intestine, male germinal cells, and the central nervous system. The lung vascular endothelium contains the highest concentration of ACE, and therefore, the lung serves as the major organ for the production of circulating angiotensin II. Although ACE was originally thought to be specific for the conversion of angiotensin I to II, it is now known to be a rather nonspecific peptidyl dipeptide hydrolase that can cleave dipeptides from the carboxy terminus of a number of endogenous peptides (e.g., substance P, bradykinin). Peptides with penultimate prolyl residues are not cleaved by converting enzyme; this accounts for the biological stability of angiotensin II. Inhibition of converting enzyme results in an elevated pool of angiotensin I. A mutation deletion in the ACE gene has been linked to a higher risk factor for hypertension, left ventricular hypertrophy, and myocardial infarction.

The Angiotensins

The amino acid composition of the peptides and enzymes involved in the synthesis and metabolism of the angiotensins is shown in Figure 18.1. Angiotensin I is believed to have little direct biological activity and must be converted to angiotensin II or angiotensin 1-7 before characteristic responses of the renin–angiotensin system are manifested. Angiotensin I and II are metabolized at their animo terminus by *aspartyl aminopeptidase,* an enzyme in plasma and numerous tissues. Angiotensin II is rapidly metabolized by aspartyl aminopeptidases, endopeptidases, and carboxypeptidases, while angiotensin III is hydrolyzed by aminopeptidases, endopeptidases,

and carboxypeptidases (Fig. 18.1). The biological activity of angiotensin III ranges from one-fourth to equipotent with angiotensin II, depending on the response being monitored. The smallest biologically active peptide in this system is angiotensin IV, which exerts unique actions in the central nervous system and periphery that are distinct from those of angiotensin II.

ANGIOTENSIN RECEPTORS

In 1988, a series of reports described the ability of imidazole acetic acid derivatives to act as antagonists at the angiotensin receptor. During the course of characterization of these compounds, it became apparent that certain tissues contained different subtypes of angiotensin receptors. Angiotensin receptors have been classified into two subtypes, AT1 and AT2. Each receptor subtype has been cloned and sequenced, with only 32% homology in the protein sequences for the two receptors. The AT1 receptor uses G proteins as signal transducers and is coupled through traditional second-messenger systems that involve phospholipase C and calcium mobilization, inhibition of adenylyl cyclase, stimulation of mitogen-activated protein kinases and the JAK/STAT pathway, and activation of Jun-kinase. In contrast, the signaling cascades of the AT2 receptor involve the activation of phosphorylases, which inhibit phosphorylation steps of certain types of cell growth.

The distribution of the AT1 and AT2 receptor subtypes is species and tissue specific. *The major biological functions of angiotensin II (cardiovascular regulation) are mediated through the ATI receptor.* In contrast, despite the increased presence of AT2 receptors in fetal tissues, a lack of AT2 receptors appears to be compatible with life. Current evidence suggests that in general, stimulation of the AT2 receptor appears to oppose those physiological actions of angiotensin II that are mediated through the AT1 receptor.

Angiotensin IV, the smallest bioactive peptide product of the renin–angiotensin system, interacts with a unique receptor termed the *angiotensin IV receptor;* this receptor exhibits minimal affinity for angiotensin II or angiotensin III.

PHARMACOLOGICAL ACTIONS

While the following discussion addresses the pharmacology of angiotensin II that is mediated through the AT1 receptor, most of these responses also follow administration of angiotensin III. Generally, angiotensin III is less potent than angiotensin II. Angiotensin 1-7 is considered to be biologically active and has been demonstrated to exert effects that are similar to, opposite of, or totally distinct from those of angiotensin II.

Vascular Smooth Muscle Contraction

The intravenous injection of angiotensin II results in a sharp rise in systolic and diastolic pressures. The response is consistently reproducible when small doses of angiotensin II are injected; however, larger amounts of the peptide produce *tachyphylaxis* (loss of response on repeated administration). The mechanism underlying tachyphylaxis to angiotensin II is unknown, but it may involve receptor internalization and/or desensitization. Subcutaneous and intramuscular injections are much less potent and have a longer duration of action than do comparable doses given intravenously. Infusions that cause an immediate pressor response tend to result in tachyphylaxis over several hours. On a molar basis, angiotensin II is about 40 times as potent as norepinephrine. The pressor response to angiotensin II is caused by its direct receptor-mediated effect on vascular smooth muscle. The peptide stimulates the formation of the second messenger inositol 1,4,5-triphosphate, which results in a release of intracellular Ca^{++} and ultimately smooth muscle contraction.

Heart Rate and Contractility

The administration of angiotensin II to an animal with intact baroreceptor reflexes results in reflex bradycardia in response to the marked vasoconstriction. When baroreceptor reflexes are depressed (barbiturate anesthesia) or if vagal tone is inhibited (atropine or vagotomy), angiotensin directly induces cardiac acceleration.

Angiotensin II stimulates the influx of Ca^{++} into cardiac muscle cells and can exert a direct inotropic effect at cardiac muscle. In addition, angiotensin II can stimulate the sympathoadrenal system and thereby increase myocardial contractility. In contrast to its effects on vascular smooth muscle, the ability of angiotensin to increase the contractile force of the heart is far less potent. Therefore, in spite of the positive chronotropic and inotropic effects produced by angiotensin II, cardiac output is rarely increased. In fact, angiotensin II may decrease cardiac output through reflex bradycardia induced by the rise in peripheral resistance that it causes. In contrast, centrally administered angiotensin II increases both blood pressure and cardiac output.

Vascular Permeability

Angiotensin II can cause a net fluid accumulation in tissues and has been shown to increase the permeability of the endothelium in large arteries and to widen the interendothelial spaces in the aorta and in coronary, mesenteric, and peripheral arteries. This response to angiotensin II probably reflects the effect of elevated pressure on the endothelial permeability barrier. The peptide also stimulates the release of the vasodilator prostacyclin from arterial endothelial cells.

Growth

Angiotensin II alters the growth of vascular smooth muscle, cardiac myocytes, and cardiac fibroblasts through mechanisms related to increased cell proliferation (hyperplasia) and protein deposition (hypertrophy). These actions of angiotensin II on cell growth involve interactions with other growth factors and are relevant to the pathophysiology of both hypertension and congestive heart failure.

Central Nervous System

Administration of angiotensin II into the vertebral circulation increases peripheral blood pressure. This hypertensive action, mediated by the central nervous system, is primarily the result of an increase in central efferent sympathetic activity going to the periphery. The area postrema of the caudal medulla appears to be the structure responsible for the central cardiovascular actions of angiotensin II.

Angiotensin II produces changes in body hydration and thirst by a direct action in the central nervous system. The administration of angiotensin II into the septal, anterior hypothalamic, and medial preoptic areas stimulates drinking behavior in several species. Part of the volume response also may be caused by the antinatriuretic and antidiuretic effects of angiotensin II.

Angiotensin II, administered into the central nervous system, increases the release of luteinizing hormone, adrenocortical hormone, thyroid-releasing hormone, β-endorphin, vasopressin, and oxytocin from the anterior pituitary. In contrast, centrally administered angiotensin II inhibits the release of anterior pituitary growth hormone and prolactin.

Sympathetic Nervous System

Angiotensin II, acting at presynaptic receptors on noradrenergic nerve terminals, potentiates the release of norepinephrine during low-frequency sympathetic nerve stimulation. Aside from its action on the nerve terminals of postganglionic sympathetic neurons, angiotensin II can directly stimulate sympathetic neurons in the central nervous system, in peripheral autonomic ganglia, and at the adrenal medulla.

Adrenal Cortex and Aldosterone Secretion

Angiotensin II stimulates aldosterone synthesis and secretion from the glomerulosa cells of the adrenal cortex. The aldosterone secretion induced by angiotensin II in humans is not accompanied by an increase in glucocorticoid plasma levels. Chronic administration of angiotensin II will maintain elevated aldosterone secretion for several days to weeks unless hypokalemia ensues.

ANTAGONISTS OF THE RENIN–ANGIOTENSIN SYSTEM

A summary of the agents that inhibit the renin–angiotensin system and their sites of action is provided in Figure 18.3.

Renin Inhibitors

The acid protease inhibitor *pepstatin* and some analogues of angiotensinogen can competitively inhibit the formation of angiotensin I by human renin. Highly specific renin inhibitors may prove beneficial as antihypertensive agents or in the treatment of congestive heart failure. Despite extensive efforts to develop renin inhibitors, most compounds capable of inhibiting renin are large peptidelike molecules that lack adequate physical chemical properties to permit oral absorption.
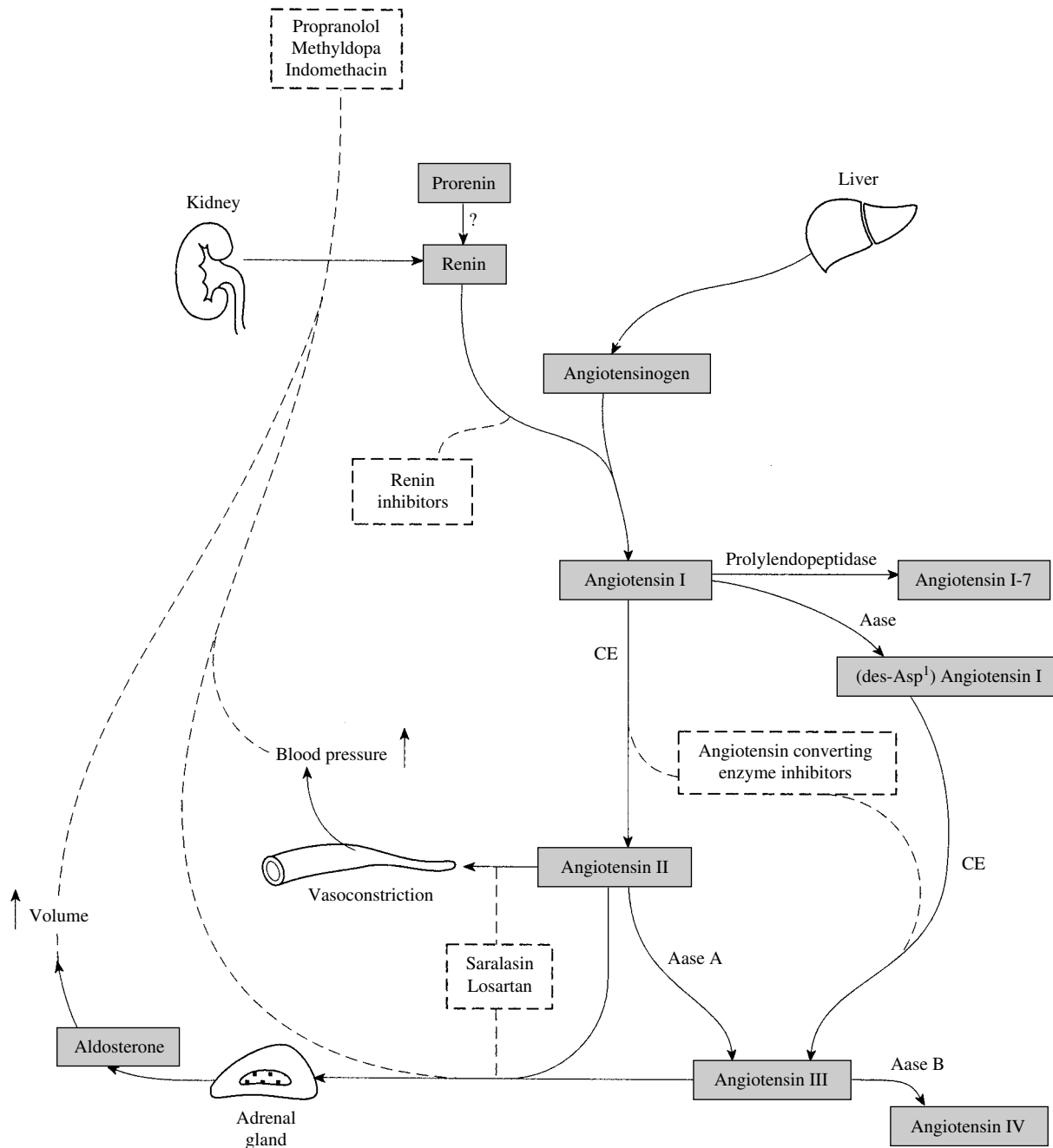
Angiotensin-Converting Enzyme Inhibitors

Many of the orally active ACE inhibitors are prodrugs. These include perindopril, quinapril, benazepril, ramipril, enalapril, trandolapril, and fosinopril.

Captopril

Captopril (*Capoten*) is an orally effective ACE inhibitor with a sulfhydryl moiety that is used in binding to the active site of the enzyme. Captopril blocks the blood pressure responses caused by the administration of angiotensin I and decreases plasma and tissue levels of angiotensin II.

Pharmacological Actions
Treatment with captopril reduces blood pressure in patients with renovascular disease and in patients with essential hypertension. The decrease in arterial pressure is related to a reduction in total peripheral resistance. Most studies demonstrate a good correlation between the hypotensive effect of inhibitors and the degree of blockade of the renin–angiotensin system. Many of the pharmacological effects of captopril are attributable to the inhibition of angiotensin II synthesis. However, ACE is a relatively nonselective enzyme that also catabolizes a family of kinins to inactive products (Fig. 18.4). Bradykinin, one of the major kinins, acts as a vasodilator through mechanisms related to the production of nitric oxide and prostacyclin by the vascular endothelium. Thus, administration of the ACE inhibitor captopril not only inhibits angiotensin II production but also prevents the breakdown of bradykinin. Increases in bradykinin concentrations after administration of ACE inhibitors contribute to the therapeutic efficacy of these compounds in the treatment of hypertension and congestive heart failure. However, alterations in bradykinin

FIGURE 18.3
Agents that inhibit the renin–angiotensin–aldosterone system and points at which they act. CE, converting enzyme; Aase, aspartyl aminopeptidase.

concentrations are also thought to contribute to cough and angioedema sometimes seen after ACE inhibition.

The hypotensive response to captopril is accompanied by a fall in plasma aldosterone and angiotensin II levels and an increase in plasma renin activity. Serum potassium levels are not affected unless potassium supplements or potassium-sparing diuretics are used concomitantly; this can result in severe hyperkalemia.

There is no baroreflex-associated increase in heart rate, cardiac output, or myocardial contractility in response to the decrease in pressure, presumably because captopril decreases the sensitivity of the baroreceptor reflex.

Captopril enhances cardiac output in patients with congestive heart failure by inducing a reduction in ventricular afterload and preload. Converting enzyme inhibitors have been shown to decrease the mass and wall
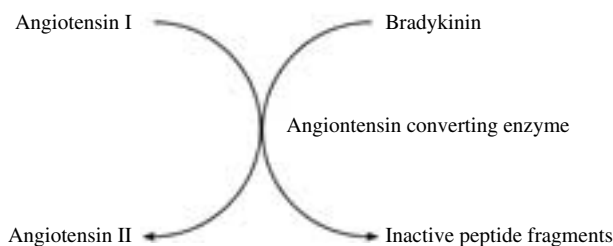
FIGURE 18.4
Interrelationship between the renin–angiotensin system and bradykinin.

thickness of the left ventricle in both normal and hypertrophied myocardium. ACE inhibitors lack metabolic side effects and do not alter serum lipids.

Pharmacokinetics

The onset of action following oral administration of captopril is about 15 minutes, with peak blood levels achieved in 30 to 60 minutes. Its apparent biological half-life is approximately 2 hours, with its antihypertensive effects observed for 6 to 10 hours. The kidneys appear to play a major role in the inactivation of captopril.

Clinical Uses

Captopril, as well as other ACE inhibitors, is indicated in the treatment of hypertension, congestive heart failure, left ventricular dysfunction after a myocardial infarction, and diabetic nephropathy. In the treatment of essential hypertension, captopril is considered first-choice therapy, either alone or in combination with a thiazide diuretic. Decreases in blood pressure are primarily attributed to decreased total peripheral resistance or afterload. An advantage of combining captopril therapy with a conventional thiazide diuretic is that the thiazide-induced hypokalemia is minimized in the presence of ACE inhibition, since there is a marked decrease in angiotensin II–induced aldosterone release.

If the patient is asymptomatic, captopril can be used as monotherapy in the treatment of congestive heart failure. The use of ACE inhibitors in the treatment of congestive heart failure is supported by results from large-scale clinical trials demonstrating a general reduction in the relative risk of death. In symptomatic patients captopril should be used in conjunction with a diuretic because of the weak natriuretic properties of ACE inhibitors. In combination, captopril will reduce afterload and preload and prevent diuretic-induced activation of the renin–angiotensin system. Finally, ACE inhibitors may slow the progression of congestive heart failure by limiting left ventricular hypertrophy.

In the treatment of diabetic nephropathy associated with type I insulin-dependent diabetes mellitus, captopril decreases the rate of progression of renal insufficiency and retards the worsening of renal function.

Adverse Actions

Approximately 10% of the patients treated with captopril report a dose-related maculopapular rash that often disappears when the dosage of captopril is reduced. Other common adverse effects are fever, a persistent dry cough (incidence as high as 39%), initial dose hypotension, and a loss of taste that may result in anorexia. These effects are reversed when drug therapy is discontinued. More serious toxicities include a 1% incidence of proteinuria and glomerulonephritis; less common are leukopenia and agranulocytosis. Since food reduces the bioavailability of captopril by 30 to 40%, administration of the drug an hour before meals is recommended. All converting enzyme inhibitors are contraindicated in patients with bilateral renal artery disease or with unilateral renal artery disease and one kidney. Use under these circumstances may result in renal failure or paradoxical malignant hypertension.

Prodrug Angiotensin-converting Enzyme Inhibitors

Most orally effective inhibitors of peptidyl dipeptide hydrolase are prodrug ester compounds that must be hydrolyzed in plasma to the active moiety before becoming effective. These drugs include benazepril (*Lotensin*), enalapril (*Vasotec*), fosinopril (*Monopril*), moexipril (Univasc), quinapril (*Accupril*), perindopril (*Aceon*), and ramipril (*Altace*). The ester group promotes absorption of the compound from the gastrointestinal tract. In contrast to captopril, the recommended dosing interval for these prodrug compounds is once to twice daily. These compounds are otherwise generally similar to captopril in their mechanism of action and indicated uses.

All prodrug ACE inhibitors are indicated for use as first-choice agents in the treatment of hypertension and congestive heart failure. In addition, results from clinical trials demonstrate that ramipril, a prodrug ACE inhibitor, can reduce the rate of death, myocardial infarction, and stroke in a broad range of high-risk patients who did not have heart failure. These results suggest that ACE inhibitors may be useful in the management of ischemia and atherosclerosis.

While essentially all ACE inhibitors have a similar mechanism of action and therefore exhibit similar efficacy in the treatment of hypertension and congestive heart failure, these drugs differ slightly in their pharmacokinetic profiles. Enalapril, lisinopril, and quinapril are excreted primarily by the kidney, with minimal liver metabolism, while the other prodrug compounds are metabolized by the liver and renally excreted. Thus, in patients with renal insufficiency, the half-life of renally excreted ACE inhibitors is prolonged. In addition, patients with impaired liver func-

tion may have a compromised ability to convert prodrug to the active drug moiety, so the efficacy of the compounds may be reduced. In addition, compounds dependent on liver metabolism for elimination may exhibit an increase in plasma half-life. An additional property that distinguishes among these prodrugs is their individual abilities to bind tightly to tissue ACE, as opposed to the circulating form of the enzyme. Of the prodrug inhibitors, quinapril and perindopril bind most tightly.

All ACE inhibitors are contraindicated during pregnancy. Their administration to pregnant women during the second and third trimesters of pregnancy has been associated with fetal and neonatal injury, including fetal death. A summary of the ACE inhibitors and their properties is provided in Table 18.1.

Angiotensin Receptor Antagonists

Angiotensin II can bind with high affinity to two distinct receptors, termed the angiotensin type 1 (AT1) and the angiotensin type 2 (AT2) receptor. These receptors belong to a superfamily of G protein–coupled receptors that contain seven transmembrane regions. The amino acid sequence of these receptors is highly conserved across species. The AT1 and AT2 receptors share only 34% homology, have distinct signal transduction pathways, and are not necessarily found on the same cell type or tissue. As previously discussed, most of the physiological effects of angiotensin II are mediated through effects at the AT1 receptor.

Mechanism of Action and Pharmacological Actions

Losartan (*Cozaar*) was the first imidazole AT1 receptor antagonist developed and is a selective competitive antagonist to angiotensin II. Other AT1 receptor antagonists approved for the treatment of hypertension include valsartan (*Diovan*), irbesartan (*Avapro*), candesartan cilexetil (*Atacand*), telmisartan (*Micardis*), and eprosartan (*Teveten*). These antagonists share some pharmacological characteristics, including a high affinity for the AT1 receptor, little to no affinity for the AT2 receptor, high protein binding, and the ability to produce an almost insurmountable blockade of the AT1 receptor. Although all of the AT1 receptor antagonists are competitive blocking drugs, they only slowly dissociate from the receptor, and their effects cannot be easily overcome.

The administration of an AT1 receptor antagonist results in a decrease in total peripheral resistance (afterload) and cardiac venous return (preload). All of the physiological effects of angiotensin II, including stimulation of the release of aldosterone, are antagonized in the presence of an AT1 receptor antagonist. Reductions in blood pressure occur independently of the status of the renin–angiotensin system, making these drugs effective antihypertensives even in patients with normal to low activity of the renin–angiotensin system. Following the chronic administration of an AT1 receptor antagonist, plasma renin activity increases as a result of removal of the angiotensin II negative feedback.

Pharmacokinetic Profiles of AT1 Receptor Antagonists

While all AT1 receptor antagonists share the same mechanism of action, they differ in their pharmacokinetic profiles. Losartan is well absorbed following oral administration and undergoes significant first-pass liver metabolism to an active metabolite, EXP3174. This metabolite is a long-acting (6–8 hours) noncompetitive antagonist at the AT1 receptor that contributes to the pharmacological effects of losartan. Production of the long-acting metabolite accounts for the sustained antihypertensive properties of losartan following chronic therapy, which would otherwise eventually be overwhelmed by removal of the negative feedback system (inhibition of renin release) for angiotensin II production. Following oral administration, 6% of losartan is excreted unchanged in the urine.

Valsartan has a higher affinity for the AT1 receptor than losartan, does not have an active metabolite, and has a slightly longer duration of action than losartan. Irbesartan exhibits high bioavailability and high affinity for the AT1 receptor, does not have an active metabolite, and has a considerably longer duration of action than losartan. Candesartan cilexetil has an active

| TABLE 18.1 | Properties of Angiotensin-Converting Enzyme Inhibitors |

Inhibitor (trade name)	Contains Sulfhydryl Groups	Form	
		Prodrug	Nonprodrug
Captopril (*Capoten*)	X		X
Quinapril (*Accupril*)		X	
Ramipril (*Altace*)		X	
Fosinopril (*Monopril*)	X		X
Benzapril (*Lotensin*)		X	
Enalapril maleate (*Vasotec*)		X	
Lisinopril (*Prinivil, Zestril*)			X

metabolite with a long duration of action, is a prodrug, and exhibits an AT1 receptor affinity 80 times that of losartan. Telmisartan is the longest-acting AT1 receptor antagonist and has no active metabolites. In contrast, eprosartan has the shortest half-life of the AT1 receptor antagonists and has been suggested to exhibit selective blockade of some effects of angiotensin II more than others.

Clinical Uses of AT1 Receptor Antagonists

Angiotensin type 1 receptor antagonists are effective as monotherapy in the treatment of hypertension. While all of the AT1 receptor antagonists are effective in the treatment of hypertension, several comparative studies have suggested that longer-acting AT1 receptor antagonists, such as irbesartan, candesartan, and telmisartan, may be more effective than the shorter-acting antagonists at providing 24-hour control of blood pressure. In large-scale clinical trials, AT1 receptor antagonists did not exhibit a clear advantage over ACE inhibitors in reducing morbidity and mortality from congestive heart failure. Therefore, the use of AT1 receptor antagonists in the treatment of congestive heart failure is generally restricted to patients who do not tolerate ACE inhibitors.

Adverse Effects of AT1 Receptor Antagonists

All AT1 receptor antagonists have adverse effects that are not significantly different from those of a placebo, although first-dose hypotension may occur. Unlike ACE inhibitors, AT1 receptor antagonists do not produce a cough, suggesting that this side effect may be related to the buildup of bradykinin levels that occurs as a result of converting enzyme inhibition rather than to a reduction in angiotensin II levels. An additional difference between AT1 receptor antagonists and ACE inhibitors is that angiotensin II is capable of interacting at the AT2 receptor in patients treated with an AT1 receptor antagonist (but not following inhibition of ACE). The clinical significance of this difference is not understood.

Finally, additional enzymes have been identified that are capable of forming angiotensin II from angiotensin I, suggesting that inhibition of ACE may not be sufficient for the total elimination of angiotensin II. In contrast, AT1 receptor antagonists are capable of blocking the effects of angiotensin II regardless of its enzymatic route of formation. All AT1 receptor antagonists, like the ACE inhibitors, are contraindicated during pregnancy.

ALDOSTERONE

Aldosterone, produced by the adrenal cortex, acts at epithelial cells in the distal tubule of the nephron to increase the reabsorption of sodium and is therefore considered an important hormone in the regulation of electrolyte balance. Aldosterone exerts its effects at the nephron through mineralocorticoid receptors, which translocate to the nucleus upon aldosterone binding and exert genomic effects leading to increased sodium reabsorption. In addition to the epithelial effects of aldosterone at mineralocorticoid receptors, nonepithelial cells, including cardiac muscle and vascular smooth muscle cells and cells in the brain, can respond to aldosterone and result in left ventricular hypertrophy, cardiac and vascular fibrosis, and stimulation of sympathetic nervous system activity.

Spironolactone (*Aldactone*), an antagonist of the aldosterone mineralocorticoid receptor, is used to treat primary aldosteronism, essential hypertension, and congestive heart failure (see Chapter 21). In the treatment of hypertension resulting from adrenal adenoma (primary aldosteronism) and in patients with essential hypertension, spironolactone lowers blood pressure primarily through blockade of epithelial mineralocorticoid receptors in the kidney, reductions in sodium and water reabsorption, and diuresis. The use of spironolactone in the treatment of essential hypertension is typically restricted to patients who do not respond appropriately to other agents and is often used in combination drug therapy. In large-scale clinical trials in patients with severe heart failure, administration of spironolactone markedly reduced morbidity and mortality without reducing blood pressure. Spironolactone is used to treat patients with moderate to severe heart failure who exhibit symptoms and ventricular dysfunction despite treatment with an ACE inhibitor or a diuretic.

Adverse effects of spironolactone therapy include hyperkalemia, gastrointestinal problems, gynecomastia (breast enlargement in males), and impotence. Gynecomastia and impotence arising from spironolactone treatment are results of significant blockade of the androgen and mineralocorticoid receptors. Novel selective mineralocorticoid receptor antagonists, such as eplerenone, are in clinical trials.

OTHER VASOACTIVE SUBSTANCES

Bradykinin

The kallikrein–kinin system is an enzymatic pathway giving rise to two predominant vasoactive peptides, kallidin and bradykinin. Kallikrein, the enzyme responsible for the formation of these peptides, exists in plasma and tissues. However, circulating levels of the end products, kallidin and bradykinin, are quite low because the kallikrein enzymes are present largely in inactive forms. In addition, the short half-life of these peptides (15 seconds) also contributes to low plasma levels. In general, the kinins produce relaxation of vascular smooth muscle and vasodilation. Bradykinin causes

vascular smooth muscle relaxation by stimulating the endothelium to release prostacyclin and nitric oxide. Blood flow to the brain, heart, viscera, skeletal muscle, and glands is increased. In nonvascular smooth muscle, bradykinin will produce a contractile response.

Other actions of kinins include activation of clotting factors simultaneously with the production of bradykinin. In the kidney, bradykinin production results in an increase in renal papillary blood flow, with a secondary inhibition of sodium reabsorption in the distal tubule. In the peripheral nervous system, bradykinin is important for the initiation of pain signals. It is also associated with the edema, erythema, and fever of inflammation.

Bradykinin exerts its physiological effects via two receptors, the B1 and B2 receptors, with most of its physiological effects being mediated by the B2 receptor. The precise function of the B1 receptor is unclear; however, some of the chronic inflammatory responses to bradykinin may be mediated through actions at this receptor.

Bradykinin antagonists of the B2 receptor are currently in development and may find utility in the treatment of pain associated with burns and such chronic inflammatory disorders as arthritis, asthma, and chronic pain.

Endothelin

Endothelins are a family of vasoactive peptides secreted by endothelial cells. The three major endothelin peptides are all composed of 21 amino acids. *Endothelins are the most potent vasoconstrictors known.* Contraction of vascular smooth muscle in response to endothelin is associated with an increase in intracellular calcium. Increases in endothelin levels have been reported in patients with vasospastic, hypoxic, and ischemic diseases. The two identified isoforms of endothelin receptors have differing affinity for the three endothelin peptides. Selective and nonselective endothelin receptor antagonists are in development for potential use in the treatment of hypertension and other disorders associated with increased vascular resistance.

Natriuretic Peptides

Natriuretic peptides are naturally occurring substances in the body that oppose the activity of the renin–angiotensin system. The natriuretic peptide family consists of atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), and C-type natriuretic peptide (CNP). All three natriuretic peptides are synthesized from cleavage of a larger precursor polypeptide. In the ventricles and brain, the synthesis of BNP predominates; ANP is synthesized by cardiac myocytes predominately in the atria; and CNP is synthesized in the brain, blood vessels, and kidney.

All three peptides exhibit similar biological activities; however, they differ in the potency of individual responses. The target organs of the natriuretic peptides include the kidneys, blood vessels, brain, and adrenal cortex. These peptides exhibit potent diuretic, natriuretic, and vasodilator effects. Natriuretic peptides promote endothelial permeability and the movement of water from the intravascular to the extravascular space. In the kidney, natriuretic peptides increase the glomerular filtration rate through vasodilation of the afferent arteriole and constriction of the efferent arteriole, inhibition of the reabsorption of sodium in the proximal and distal tubule, and inhibition of renin synthesis. In the brain, natriuretic peptides are involved in the regulation of central control of cardiovascular functions. These biological effects of natriuretic peptides come together to reduce venous return and total peripheral resistance, thereby improving cardiac performance and reducing blood pressure.

The release of ANP from the heart is regulated acutely by stretch of atrial myocytes and has been used as a marker for cardiovascular diseases, including congestive heart failure and hypertension. In addition, recent results demonstrate an increase in the circulating concentration of ANP following stroke and linkage of the ANP gene to patients who have strokes. Two types of atrial natriuretic receptors have been identified in target tissues, including guanylate cyclase–linked receptors (subdivided into types A and B) and a receptor thought to serve as a clearance mechanism for the removal of circulating ANP. Analogues that act as ANP agonists are being developed for use in hypertension and congestive heart failure.

In addition, a new class of drugs, termed vasopeptidase inhibitors, inhibit the enzymatic activity of ACE and neutral endopeptidase, the enzyme responsible for the breakdown of natriuretic peptides. The end result is a reduction in the synthesis of angiotensin II and an increase in the circulating level of natriuretic peptides such as ANP. Omapatrilat, a vasopeptidase inhibitor, is under study for the treatment of hypertension and congestive heart failure.

Nitric Oxide

Nitric oxide is a small, unstable free radical that acts as a biological messenger in many physiological responses. Because it can diffuse freely in all directions from its site of origin, regulation of the activity of nitric oxide is primarily through control of its synthesis. Formation of nitric oxide occurs through oxidation of the amino acid L-arginine, a reaction catalyzed by the enzyme nitric oxide synthase (NOS), to produce nitric oxide and L-citrulline. The forms of NOS differ in their cellular location and expression (constitutive expression versus inducible expression). Activation of synthesis of the inducible form

of NOS results in continued synthesis of nitric oxide for several hours. Inhibitors of NOS are analogues of arginine, including L-Nw nitroarginine (L- NNA) and L-Nw methylarginine (L-NMA), both of which decrease nitric oxide synthesis.

Physiological sites proposed for nitric oxide action include the immune system, where nitric oxide acts as a cytostatic agent, is tumoricidal, and can inhibit viral replication. In the cardiovascular system, nitric oxide is the biological mediator of vasodilator responses to agents such as acetylcholine and bradykinin, which act as receptors on endothelial cells to activate NOS and stimulate nitric oxide production. Diffusible nitric oxide then activates guanylate cyclase in vascular smooth muscle cells, leading to the production of cyclic guanosine monophosphate (GMP) and vasodilation. In the brain, stimulation of *N*-methyl-D-aspartate receptors on neurons leads to activation of the brain form of NOS and stimulates production of nitric oxide. The function of brain nitric oxide is thought to involve actions as a retrograde neurotransmitter whereby nitric oxide diffuses back to the presynaptic neuron to activate guanylate cyclase and increase cyclic GMP levels. Through these retrograde actions nitric oxide is thought to play a role in the neural circuitry involved in memory.

Even though nitric oxide is the physiological mediator of a variety of responses, excess nitric oxide is toxic to many cells as a result of its role in the production of peroxynitrite and resultant lipid oxidation. Inhibitors of the NOS enzyme are in clinical trials for the treatment of hypotension associated with septic shock. Administration of low concentrations of nitric oxide through respiratory ventilators has been implemented to treat persistent pulmonary hypertension of the newborn.

Study QUESTIONS

1. An accurate statement regarding the actions of both ACE inhibitors and AT1 receptor antagonists is that
   (A) Both classes of drugs increase bradykinin.
   (B) Angiotensin II can act at the AT2 receptor with both classes of drugs.
   (C) Both classes of drugs reduce total peripheral resistance.
   (D) Both classes of drugs decrease circulating angiotensin II levels.
   (E) Both classes of drugs are first-choice treatments for congestive heart failure.
2. Angiotensin II can
   (A) Increase the synthesis and release of aldosterone
   (B) Reduce the activity of the sympathetic nervous system
   (C) Be a potent positive inotropic at the heart
   (D) Relax vascular smooth muscle
   (E) Reduce the growth of cardiovascular cell types
3. The most potent vasoconstrictor known is
   (A) Bradykinin
   (B) Angiotensin II
   (C) Angiotensin IV
   (D) Natriuretic peptide
   (E) Endothelin
4. The mechanism of action of captopril is
   (A) Angiotensin receptor antagonist
   (B) ACE inhibitor
   (C) Aldosterone receptor antagonist
   (D) Bradykinin antagonist

5. L-Argenine serves as a precursor for
   (A) Bradykinin
   (B) L-Citrulline
   (C) Nitrous oxide
   (D) Atrial natriuretic peptide

ANSWERS
1. **C.** ACE inhibitors increase circulating bradykinin levels, while AT1 receptor antagonists have no effect on circulating bradykinin. The ability of converting enzyme inhibitors to increase bradykinin levels is thought to contribute to the benefits of this class of drugs in the treatment of hypertension and heart failure. With an ACE inhibitor, the actions of angiotensin II at both the AT1 and the AT2 receptor is decreased; however, with an AT1 receptor antagonist, angiotensin II can act at the AT2 receptor. Only ACE inhibitors decrease circulating angiotensin II levels; the level of angiotensin II may actually increase with an AT1 receptor antagonist because of removal of the endocrine feedback loop. ACE inhibitors have proven benefits in the treatment of congestive heart failure, while AT1 receptor antagonists are reserved for therapy of patients who have significant adverse effects from converting enzyme inhibition.
2. **A.** Angiotensin II has diverse physiological effects, including stimulating the synthesis and release of aldosterone from the adrenal cortex. This effect of angiotensin II results in fluid and water retention. The other answers are incorrect in that angiotensin II

stimulates the sympathetic nervous system, is a weak inotropic, contracts vascular smooth muscle, and increases the growth status of cardiovascular cell types.
3. **E.** Bradykinin and natriuretic peptide are vasodilators. Both angiotensin II and angiotensin IV are vasoconstrictors but not nearly as potent as any endothelin peptide.
4. **B.** Compounds that act as ACE inhibitors are particularly useful for the treatment of hypertension and congestive heart failure.
5. **C.** Nitric oxide is an important compound that acts as a biological messenger in many physiological responses. L-Citrulline is a product of the oxidation of L-argenine in the formation of nitric oxide. Bradykinin is formed from a precursor kininogen.

Atrial natriuretic peptide is synthesized by cleavage of a larger precursor polypeptide.

SUPPLEMENTAL READING

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CASE Study   Congestive Heart Failure with Complications

A. D. has refused to go for a yearly physical examination for more than 15 years despite a family history of cardiovascular disease. Over the past 6 months, A. D. has had to cut down and then eliminate his weekly soccer games because of extreme shortness of breath. Exasperated over his inability to exercise, A. D. goes to a primary care physician and is diagnosed with moderate to severe hypertension with a markedly enlarged heart. Unfortunately, A. D. has significant liver impairment from a previous bout of hepatitis. Based on knowledge of the renin–angiotensin system and the specifics concerning drugs targeted against this system, provide a rationale for use of two drugs to treat the cardiovascular disease in this patient.

ANSWER: This patient has congestive heart failure from long-standing hypertension. In addition, the patient has compromised liver function, limiting the choice of drug. Drugs that inhibit the renin–angiotensin system, specifically ACE inhibitors, are indicated in the treatment of both hypertension and congestive heart failure. Therefore, an ACE inhibitor is a rational pharmacological approach in this patient. However, the compromised liver function requires caution with any drug that requires liver metabolism for formation of the active drug moiety (i.e., prodrugs), or for drugs that are primarily eliminated by liver metabolism. With the exception of captopril and lisinopril, all of the available ACE inhibitors are prodrugs and require liver metabolism for elimination. Therefore, either captopril or lisinopril would be an appropriate ACE inhibitor in the treatment of this patient. AT1 receptor antagonists are indicated for the treatment of hypertension and when converting enzyme inhibitors are contraindicated in the therapy of congestive heart failure. Of this class of compounds, eprosartan, valsartan, and telmisartan do not require liver metabolism to produce an active compound.