Introduction to the Endocrine System

**OBJECTIVES**

1. List the main endocrine glands of the body.
2. List the chemical nature of the major hormones.
3. Describe how the chemical nature influences hormone synthesis, storage, secretion, transport, clearance, mechanism of action, and appropriate route of exogenous hormone administration.
4. Explain the significance of hormone binding to plasma proteins.
5. Describe the major signal transduction pathways, and their mechanism for termination, for different classes of hormones and provide a specific example of each.

**Endocrine glands** secrete chemical messengers, called hormones (Box 1.1), into the extracellular fluid in a highly regulated manner. Secreted hormones gain access to the circulation, often via fenestrated capillaries, and regulate target organs throughout the body. The endocrine system is composed of the pituitary gland, the thyroid gland, parathyroid glands, and adrenal glands (Fig. 1.1). The endocrine system also includes the ovary and testis, which carry out a gametogenic function that is absolutely dependent on their endogenous endocrine function. In addition to dedicated endocrine glands, endocrine cells reside as a minor component (in terms of mass) in other organs, either as groups of cells (the islets of Langerhans in the pancreas) or as individual cells spread throughout several glands, including the gastrointestinal (GI) tract, kidney, heart, adipose tissue, and liver. In addition, there are several types of hypothalamic neuroendocrine neurons that produce hormones. The placenta serves as a transitory exchange organ, but also functions as an important endocrine structure of pregnancy.

The endocrine system also encompasses a range of specific enzymes, either cell-associated or circulating, that perform the function of peripheral conversion of hormonal precursors (see Box 1.1). For example, angiotensinogen from the liver is converted in the circulation to angiotensin I by the renal-derived enzyme renin, followed by conversion to the active hormone angiotensin II by the transmembrane ectoenzyme angiotensin I–converting enzyme (ACE) that is enriched in the endothelia of the lungs (see Chapter 7). Another example of peripheral conversion of a precursor to an active hormone involves the two sequential hydroxylations of vitamin D in hepatocytes and renal tubular cells.

Numerous extracellular messengers, including prostaglandins, growth factors, neurotransmitters, and cytokines, also regulate cellular function. However, these messengers act predominantly within the context of a microenvironment in an autocrine or paracrine manner, and thus are discussed only to a limited extent where needed.

To function, hormones must bind to specific receptors expressed by specific target cell types within target organs. Hormones are also referred to as ligands, in the context of ligand receptor binding, and as agonists, in that their binding to the receptor is transduced into a cellular response. Receptor antagonists typically bind to a receptor and lock it in an inactive state, unable to induce a cellular response. Drugs that bind to and alter the activity of steroid hormone receptors are referred to as selective receptor modulators. For example, Tamoxifen is a mixed estrogen receptor agonist/antagonist, and thus is referred to as a “selective estrogen receptor modulator” or SERM. Loss or inactivation of a receptor leads to hormonal resistance. Constitutive activation of a receptor leads to unregulated, hormone-independent activation of cellular processes.

The widespread delivery of hormones in the blood makes the endocrine system ideal for the functional coordination of multiple organs and cell types in the following contexts:

1. Allowing normal development and growth of the organism
2. Maintaining internal homeostasis
## BOX 1.1 A List of Most Hormones and Their Sites of Production

### Hormones Synthesized and Secreted by Dedicated Endocrine Glands

#### Pituitary Gland
- Growth hormone (GH)
- Prolactin
- Adrenocorticotropic hormone (ACTH)
- Thyroid-stimulating hormone (TSH)
- Follicle-stimulating hormone (FSH)
- Luteinizing hormone (LH)

#### Thyroid Gland
- Tetraiodothyronine (T\(_4\); thyroxine)
- Triiodothyronine (T\(_3\))
- Calcitonin

#### Parathyroid Glands
- Parathyroid hormone (PTH)

#### Islets of Langerhans (Endocrine Pancreas)
- Insulin
- Glucagon
- Somatostatin

#### Adrenal Gland
- Epinephrine
- Norepinephrine
- Cortisol
- Aldosterone
- Dehydroepiandrosterone sulfate (DHEAS)

### Hormones Synthesized by Gonads

#### Ovaries
- Estradiol-17β
- Progesterone
- Inhibin

#### Testes
- Testosterone
- Antimüllerian hormone (AMH)
- Inhibin

### Hormones Synthesized in Organs with a Primary Function Other Than Endocrine

#### Brain (Hypothalamus)
- Antidiuretic hormone (ADH; vasopressin)
- Oxytocin
- Corticotropin-releasing hormone (CRH)
- Thyrotropin-releasing hormone
- Gonadotropin-releasing hormone (GnRH)
- Growth hormone–releasing hormone (GHRH)
- Somatostatin

#### Brain (Pineal Gland)
- Melatonin

#### Heart
- Atrial natriuretic peptide (ANP)

#### Kidney
- Erythropoietin

#### Adipose Tissue
- Leptin
- Adiponectin

#### Stomach
- Gastrin
- Somatostatin
- Ghrelin

#### Intestines
- Secretin
- Cholecystokinin
- Glucagon-like peptide-1 (GLP-1)
- Glucagon-like peptide-2 (GLP-2)
- Glucose-dependent insulinotropic peptide (GIP; gastrin inhibitory peptide)
- Motilin

#### Liver
- Insulin-like growth factor-I (IGF-I)

#### Hormones Produced to a Significant Degree by Peripheral Conversion

#### Lungs
- Angiotensin II

#### Kidney
- 1α,25-dihydroxyvitamin D

#### Adipose, Mammary Glands, Other Organs
- Estradiol-17β

#### Liver, Other Organs
- Testosterone

#### Genital Skin, Prostate, Sebaceous Gland, Other Organs
- 5-Dihydrotestosterone (DHT)

#### Many Organs
- T \(_3\)
3. Regulating the onset of reproductive maturity at puberty and the function of the reproductive system in the adult

In the adult, endocrine organs produce and secrete their hormones in response to feedback control systems that are tuned to set-points, or set ranges, of the levels of circulating hormones. These set-points are genetically determined but may be altered by age, circadian rhythms (24-hour cycles or diurnal rhythms), seasonal cycles, the environment, stress, inflammation, and other influences.

Major forms of endocrine disease are caused by lack of hormone (e.g., hypothyroidism), excess of hormone (e.g., hyperparathyroidism) or dysfunction of receptor (hormonal resistance). It is important to appreciate that hormones often stimulate both the differentiated function and growth of target tissues and organs. This underlies the role of hormones in driving neoplastic transformation and cancer progression (i.e., the existence of hormonally responsive cancers). The pathogenesis of these and other forms of endocrine disease are discussed in the subsequent chapters.

The material in this chapter covers generalizations common to all hormones or to specific groups of hormones. The chemical nature of the hormones and their mechanisms of action are discussed. This presentation provides the generalized information necessary to categorize the hormones and to make predictions about the most likely characteristics of a given hormone. Some of the exceptions to these generalizations are discussed later.

### CHEMICAL NATURE OF HORMONES

Hormones are classified biochemically as proteins/peptides, catecholamines, steroid hormones, and iodothyronines.

The chemical nature of a hormone determines the following:

1. How it is synthesized, stored, and released in a regulated manner
2. How it is carried in the blood
3. Its biologic half-life ($t_{1/2}$) and mode of clearance
4. Its cellular mechanism of action

#### Proteins/Peptides

The protein and peptide hormones can be grouped into structurally related molecules that are encoded by gene families (Box 1.2). Protein/peptide hormones gain their specificity from their primary amino acid sequence, which
confers specific higher-order structures, and from post-translational modifications, such as glycosylation.

Protein/peptide hormones are synthesized on the polyribosome as larger preprohormones. The nascent peptides have at their N terminus a group of 15 to 30 amino acids called the signal peptide, which directs the growing polypeptide through the endoplasmic reticular membrane into the cisternae. The signal peptide is enzymatically removed, and the protein is then transported from the cisternae to the Golgi apparatus, where it is packaged into a membrane-bound secretory vesicle that buds off into the cytoplasm. Posttranslational modification occurs in the endoplasmic reticulum, Golgi apparatus, and secretory vesicle.

The original gene transcript is called either a prehormone or a preprohormone (Fig. 1.2). Removing the signal peptide produces either a hormone or a prohormone. A prohormone is a polypeptide that requires further cleavage before the mature hormone is produced. Often this final cleavage occurs while the prohormone is within the Golgi apparatus or the secretory vesicle. Sometimes prohormones contain the sequence of multiple hormones. For example, the protein, proopiomelanocortin (POMC), contains the amino acid sequences of adrenocorticotropic hormone (ACTH) and α-melanocyte-stimulating hormone (αMSH). However, the pituitary corticotrope produces ACTH only, whereas keratinocytes and specific hypothalamic neurons produce αMSH, but not ACTH. The ability of cells to process the same prohormone into different peptides is due to cell type expression of prohormone convertases, resulting in cell-specific processing of the prohormone.

Protein/peptide hormones are stored in the gland as membrane-bound secretory vesicles and are released by exocytosis through the regulated secretory pathway. This means that hormones are not continually secreted, but rather that they are secreted in response to a stimulus, through a mechanism of stimulus-secretion coupling. Regulated exocytosis is induced by an elevation of intracellular Ca²⁺ along with activation of other components (e.g., small G proteins), which interact with vesicular and cell membrane components. This ultimately leads to the fusion of the secretory vesicular membrane with the cell membrane and exocytosis of the vesicular contents.

Protein/peptide hormones are soluble in aqueous solvents and, with the notable exceptions of the insulin-like growth factors (IGFs) and growth hormone (GH), circulate in the blood predominantly in an unbound form; therefore they tend to have short biologic half-lives ($t_{1/2}$). Protein hormones are removed from the circulation by receptor-mediated endocytosis and lysosomal turnover of hormone receptor complexes (see later). Many protein hormones are small enough to appear in the urine in a
physiologically active form. For example, follicle-stimulating hormone (FSH) and luteinizing hormone (LH) are present in urine. Pregnancy tests using human urine are based on the presence of the placental LH-like hormone, human chorionic gonadotropin (hCG).

Proteins/peptides are readily digested if administered orally. Hence, they must be administered by injection or, in the case of small peptides, through a mucous membrane (sublingually or intranasally). Because proteins/peptides do not cross cell membranes readily, they signal through transmembrane receptors.

**Catecholamines**

Catecholamines are synthesized by the adrenal medulla and neurons and include norepinephrine, epinephrine, and dopamine (Fig. 1.3; Box 1.3). The primary hormonal product of the adrenal medulla is epinephrine, and to a lesser extent, norepinephrine. Epinephrine is produced by enzymatic modifications of the amino acid tyrosine. Epinephrine and other catecholamines are ultimately stored in secretory vesicles that are part of the regulated secretory pathway. Epinephrine is hydrophilic and circulates either unbound or loosely bound to albumin. Epinephrine and norepinephrine are similar to protein/peptide hormones in that they signal through membrane receptors, called adrenergic receptors. Catecholamines have short biologic half-lives (a few minutes) and are inactivated by intracellular enzymes. Inactivated forms diffuse out of cells and are excreted in the urine.

**Steroid Hormones**

Steroid hormones are made by the adrenal cortex, ovaries, testes, and placenta (Box 1.4). Steroid hormones from these glands fall into five categories: progestins, mineralcorticoids, glucocorticoids, androgens, and estrogens (Table 1.1). Progestins and the corticoids are 21-carbon steroids, whereas androgens are 19-carbon steroids and estrogens are 18-carbon steroids. Steroid hormones also include the active metabolite of vitamin D, which is a secosteroid (see Chapter 4).

Steroid hormones are synthesized by a series of enzymatic modifications of cholesterol (Fig. 1.4). The enzymatic modifications of cholesterol are of three general types: hydroxylations, dehydrogenations/hydrogenations, and breakage of carbon-carbon bonds. The purpose of these modifications is to produce a cholesterol derivative that is sufficiently unique to be recognized by a specific receptor. Thus progestins bind to the progesterone receptor (PR), mineralocorticoids bind to the mineralocorticoid receptor (MR), glucocorticoids bind to the glucocorticoid receptors, and estrogens bind to nuclear hormone receptors. The regulation of the synthesis of steroid hormones is complex. They are regulated at the level of transcription by the binding of specific regulatory proteins to DNA. They are also regulated at the level of secretion by the actions of other hormones and neurotransmitters.
receptor (GR), androgens bind to the androgen receptor (AR), estrogens bind to the estrogen receptor (ER), and the active vitamin D metabolite binds to the vitamin D receptor (VDR).

The complexity of steroid hormone action is increased by the expression of multiple forms of each receptor. Additionally, there is some degree of nonspecificity between steroid hormones and the receptors they bind to. For example, glucocorticoids bind to the MR with high affinity, and progestins, glucocorticoids, and androgens can all interact with the PR, GR, and AR to some degree. An appreciation of this "cross-talk" is important to the physician who is prescribing synthetic steroids. For example, medroxyprogesterone acetate (a synthetic progesterone given for hormone replacement therapy in postmenopausal women) binds well to the AR as well as the PR. As discussed subsequently, steroid hormones are lipophilic and pass through cell membranes easily. Accordingly, classic steroid hormone receptors are localized intracellularly and act by regulating gene expression. More recently, membrane and juxtamembrane receptors have been discovered that mediate rapid, nongenomic actions of steroid hormones.

Steroidogenic cell types are defined as cells that can convert cholesterol to pregnenolone, which is the first reaction common to all steroidogenic pathways. Steroidogenic cells have some capacity for cholesterol synthesis but often obtain cholesterol from circulating cholesterol-rich lipoproteins (low-density lipoproteins and high-density lipoproteins; see Chapter 3). Pregnenolone is then further modified by six or fewer enzymatic reactions. Because of their hydrophobic nature, steroid hormones and precursors can leave the steroidogenic cell easily and so are not stored. Thus steroidogenesis is regulated at the level of uptake, storage, and mobilization of cholesterol and at the level of steroidogenic enzyme gene expression and activity. Steroids are not regulated at the level of secretion of the preformed hormone. A clinical implication of this mode of secretion is that high levels of steroid hormone precursors are easily released into the blood when a downstream steroidogenic enzyme within a given pathway is inactive or absent (Fig. 1.5). In comparing the ultrastructure of a protein hormone–producing cell to that of a steroidogenic cell, protein hormone–producing cells store the product in secretory granules and have extensive rough endoplasmic reticulum. In contrast, steroidogenic cells store the precursor (cholesterol esters) in the form of lipid droplets, but do not store the product. Steroidogenic enzymes are localized to smooth endoplasmic reticulum membrane and within mitochondria, and these two organelles are numerous in steroidogenic cells.

An important feature of steroidogenesis is that steroid hormones often undergo further modifications (apart from those involved in deactivation and excretion) after their release from the original steroidogenic cell. This is referred to as peripheral conversion. For example, estrogen synthesis by the ovary and placenta requires at least two cell types to complete the pathway of cholesterol to estrogen (see Chapters 10 and 11). This means that one cell secretes a precursor, and a second cell converts the precursor to estrogen. There is also considerable peripheral conversion of active steroid hormones. For example, the testis secretes sparingly little estrogen. However, adipose, muscle, and other tissues express the enzyme for converting testosterone (a potent androgen) to estradiol-17β. Peripheral conversion of steroids plays an important role in several endocrine disorders (e.g., see Fig. 1.5).

Steroid hormones are hydrophobic, and a significant fraction circulates in the blood bound to transport proteins (see later). These include albumin, but also the specific transport proteins, sex hormone–binding globulin (SHBG) and corticosteroid-binding globulin (CBG) (see later). Excretion of hormones typically involves inactivating modifications followed by glucuronide or sulfate conjugation in the liver. These modifications increase the water solubility of the steroid and decrease its affinity for transport proteins, allowing the inactivated steroid

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TABLE 1.1 Steroid Hormones

<table>
<thead>
<tr>
<th>Family</th>
<th>No. of Carbons</th>
<th>Specific Hormone</th>
<th>Primary Site of Synthesis</th>
<th>Primary Receptor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Progestin</td>
<td>21</td>
<td>Progesterone</td>
<td>Ovary placenta</td>
<td>Progesterone receptor (PR)</td>
</tr>
<tr>
<td>Glucocorticoid</td>
<td>21</td>
<td>Cortisol, Corticosterone</td>
<td>Adrenal cortex</td>
<td>Glucocorticoid receptor (GR)</td>
</tr>
<tr>
<td>Mineralocorticoid</td>
<td>21</td>
<td>Aldosterone, 11-Deoxycorticosterone</td>
<td>Adrenal cortex</td>
<td>Mineralocorticoid receptor (MR)</td>
</tr>
<tr>
<td>Androgen</td>
<td>19</td>
<td>Testosterone, Dihydrotestosterone</td>
<td>Testis</td>
<td>Androgen receptor (AR)</td>
</tr>
<tr>
<td>Estrogen</td>
<td>18</td>
<td>Estradiol-17β, Estriol</td>
<td>Ovary placenta</td>
<td>Estrogen receptor (ER)</td>
</tr>
</tbody>
</table>

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CHAPTER 1 Introduction to the Endocrine System

hormone to be excreted by the kidney. Steroid compounds are absorbed fairly readily in the GI tract and therefore often may be administered orally.

**Thyroid Hormones**

Thyroid hormones are classified as iodothyronines (Fig. 1.6) that are made by the coupling of iodinated tyrosine residues through an ether linkage (Box 1.5; also see Chapter 6). Their specificity is determined by the thyronine structure, but also by exactly where the thyronine is iodinated. Normally, the predominant iodothyronine released by the thyroid is $T_4$ (3,5,3',5'-tetraiodothyronine, also called thyroxine), which acts as a circulating precursor of the active form, $T_3$ (3,5,3'-triiodothyronine). Thus peripheral conversion through specific 5'-deiodination plays an important role in thyroid function (see Chapter 6). Thyroid hormones cross cell membranes by both diffusion and transport systems. They are stored extracellularly in the thyroid as an integral part of the glycoprotein molecule thyroglobulin (see Chapter 6). Thyroid hormones are sparingly soluble in blood and are transported in blood bound to thyroid hormone-binding globulin (TBG). $T_4$ and $T_3$ have long half-lives of 7 days and 24 hours, respectively. Thyroid hormones are similar to steroid hormones in that the thyroid hormone receptor (TR) is intracellular and acts as a transcription factor. In fact, the TR belongs to the same gene family that includes steroid hormone receptors and VDRs. Thyroid hormones can be administered orally and sufficient hormone is absorbed intact to make this an effective mode of therapy.

**TRANSPORT OF HORMONES IN THE CIRCULATION**

A significant amount of steroid and thyroid hormones is transported in the blood bound to plasma proteins that are produced in a regulated manner by the liver. Protein and polypeptide hormones are generally transported free in the blood. There exists an equilibrium among the concentrations of bound hormone, free hormone, and plasma transport protein.

The free hormone is the biologically active form for target organ action, feedback control, and clearance by uptake and metabolism. Consequently, in evaluating hormonal status, one must sometimes determine free hormone levels rather than total hormone levels alone. This is particularly important because hormone transport proteins themselves are regulated by altered endocrine and disease states.

Protein binding serves several purposes. It prolongs the circulating $t_{1/2}$ of the hormone. The bound hormone represents a “reservoir” of hormone and as such can serve to buffer acute changes in hormone secretion. In addition, steroid and thyroid hormones are lipophilic and hydrophobic. Binding to transport proteins prevents these hormones from simply partitioning into the cells near their site of secretion and allows them to be transported throughout the circulation.

**CELLULAR RESPONSES TO HORMONES**

Hormones regulate essentially every major aspect of cellular function in every organ system. Hormones control the growth and proliferation of cells. Hormones regulate the differentiation of cells through genetic and epigenetic
changes and their ability to survive or undergo programmed cell death. Hormones influence cellular metabolism, ionic composition, and transmembrane potential. Hormones orchestrate several complex cytoskeletal-associated events, including cell shape, migration, division, exocytosis, recycling/endocytosis, and cell-cell and cell-matrix adhesion.

*Fig. 1.5* Example of the effect of an enzyme defect on steroid hormone precursors in blood.

Hormones regulate the expression and function of cytosolic, membrane, and secreted proteins, and a specific hormone may determine the level of its own receptor, or the receptors for other hormones.

Although hormones can exert coordinated, pleiotropic control on multiple aspects of cell function, any given hormone does not regulate every function in every cell type. Rather, a single hormone controls a subset of cellular functions in only the cell types that express receptors for that hormone (i.e., the target cells). Thus selective receptor expression determines which cells will respond to a
given hormone. Moreover, the differentiated epigenetic state of a specific cell will determine how it will respond to a hormone. Thus the specificity of hormonal responses resides in the structure of the hormone itself, the receptor for the hormone, and the cell type in which the receptor is expressed. Serum hormone concentrations are extremely low (picomolar to nanomolar range). Therefore a receptor must have a high affinity, as well as specificity, for its cognate hormone.

Hormone receptors fall into two general classes: transmembrane receptors and intracellular receptors that belong to the nuclear hormone receptor family.

Transmembrane Receptors

Most hormones are proteins, peptides, or catecholamines that cannot pass through the cell membrane. Thus these hormones must interact with transmembrane protein receptors. Transmembrane receptors are proteins that contain three domains (proceeding from outside to inside the cell): (1) an extracellular domain that harbors a high-affinity binding site for a specific hormone; (2) one or more hydrophobic, transmembrane domains that span the cell membrane; and (3) a cytosolic domain that is linked to signaling proteins.

Hormone binding to a transmembrane receptor induces a conformational shift in all three domains of the receptor protein. This hormone receptor binding–induced conformational change is referred to as a signal. The signal is transduced into the activation of one or more intracellular signaling molecules. Signaling molecules then act on effector proteins, which, in turn, modify specific cellular functions. The combination of hormone receptor binding (signal), activation of signaling molecules (transduction), and the regulation of one or more effector proteins is referred to as a signal transduction pathway (also called simply a signaling pathway), and the final integrated outcome is referred to as the cellular response.

Signaling pathways linked to transmembrane receptors are usually characterized by the following:

A. **Receptor binding** followed by a conformational shift that extends to the cytosolic domain. The conformational shift may result in one or more of the following:
   1. Activation of a guanine exchange function of a receptor.
   2. Homodimerization and/or heterodimerization of receptors to other receptors or coreceptors within the membrane.
   3. Recruitment and activation of signaling proteins by the cytosolic domain.

B. **Multiple, hierarchical steps** in which downstream effector proteins are dependent on and driven by upstream receptors and signaling molecules and effector proteins. This means that loss or inactivation of one or more components within the pathway leads to hormonal resistance, whereas constitutive activation or overexpression of components can provoke a cellular response in a hormone-independent, unregulated manner.

C. **Amplification** of the initial hormone receptor binding–induced signal, usually by inclusion of an enzymatic step within a signaling pathway. Amplification can be so great that maximal response to a hormone is achieved upon hormone binding to a fraction of available receptors.

D. **Activation of multiple divergent or convergent pathways** from one hormone receptor–binding event. For example, binding of insulin to its receptor activates three separate signaling pathways.

E. **Antagonism by constitutive and regulated negative feedback reactions.** This means that a signal is dampened or terminated by opposing pathways. Gain of function of opposing pathways can result in hormonal resistance. Signaling pathways use several common modes of informational transfer (i.e., intracellular messengers and signaling events). These include the following:

1. **Conformational shifts.** Many signaling components are proteins and have the ability to toggle between two (or more) conformational states that alter their activity, stability, or intracellular location. As discussed previously, signaling begins with hormone receptor binding that induces a conformational change in the receptor (Fig. 1.7). The other modes of informational transfer discussed later either regulate or are regulated by conformational shifts in transmembrane receptors and in downstream signaling proteins.

2. **Covalent phosphorylation of proteins and lipids** (Fig. 1.8). Enzymes that phosphorylate proteins or lipids are called kinases, whereas those that catalyze dephosphorylation are called phosphatases. Protein kinases and phosphatases can be classified as either tyrosine-specific kinases and phosphatases or serine/threonine-specific kinases and phosphatases. There are also mixed function kinases and phosphatases that recognize all three residues. An important lipid kinase is phosphatidylinositol-3-kinase (PI3K; see later). The phosphorylated state of a signaling component can alter the following:
   a. **Activity.** Phosphorylation can activate or deactivate a substrate, and proteins often have multiple sites of phosphorylation that induce quantitative and/or qualitative changes in the protein’s activity.
   b. **Stability.** For example, phosphorylation of proteins can induce their subsequent ubiquitination and proteasomal degradation.
   c. **Subcellular location.** For example, the phosphorylation of some nuclear transcription factors induces their translocation to and retention in the cytoplasm.
d. Recruitment and clustering of other signaling proteins. For example, phosphorylation of the cytosolic domain of a transmembrane receptor often induces the recruitment of signaling proteins to the receptor where they are phosphorylated. Recruitment happens because the recruited protein harbors a domain that specifically recognizes and binds to the phosphorylated residue. Another important example of recruitment by phosphorylation is the recruitment of the protein kinase Akt/PKB to the cell membrane, where it is phosphorylated and activated by the protein kinase, PDK1. In this case Akt/PKB and PDK1 are recruited to the cell membrane by the phosphorylated membrane lipid, phosphatidylinositol 3,4,5-triphosphate (PIP₃).

3. Covalent acetylation/deacetylation of proteins. Acetylation (as well as phosphorylation) of histones and other chromatin proteins imparts epigenetic regulation by altering chromatin structure and accessibility in a regulated and, in some cases, heritable manner. Many extranuclear proteins are also regulated by their degree of acetylation. Acetyl transferases drive acetylation, whereas deacetylases drive deacetylation. A major deacetylase family is comprised of the seven sirtuins (SIRTs).

4. Noncovalent guanosine nucleotide triphosphate (GTP) binding to GTP-binding proteins (G proteins). G proteins represent a large family of molecular switches, which are latent and inactive when bound to GDP, and active when bound to GTP (Fig. 1.9). G proteins are activated by guanine nucleotide exchange factors (GEFs), which promote the dissociation of GDP and binding of GTP. G proteins have intrinsic GTPase activity. GTP is normally hydrolyzed to GDP within seconds by the G protein, thereby terminating the transducing activity of the G protein. Another G-protein termination mechanism (which represents a target for drug development to treat certain endocrine diseases) is the family of proteins called regulators of G-protein signaling (RGS proteins), which bind to active G proteins and increase their intrinsic GTPase activity.
5. **Noncovalent binding of cyclic nucleotide monophosphates to their specific effector proteins** (Fig. 1.10). Cyclic adenosine monophosphate (cAMP) is generated from adenosine triphosphate (ATP) by adenylyl cyclase, which is primarily a membrane protein. Adenylyl cyclase is activated and inhibited by the G proteins, Gs-α and Gi-α, respectively (see later). There are three general intracellular effectors of cyclic AMP (cAMP):

- **a. cAMP binds to the regulatory subunit of protein kinase A (PKA)**; also called cAMP-dependent protein kinase. Inactive PKA is a heterotetramer composed of two catalytic subunits and two regulatory subunits. cAMP binding causes the regulatory subunits to dissociate from the catalytic subunits, thereby generating two molecules of active catalytic PKA subunits (PKA\(_\varepsilon\)). PKA\(_\varepsilon\) phosphorylates numerous proteins on serine and threonine residues.
Substrates of PKA include numerous cytosolic proteins as well as transcription factors, most notably cAMP-responsive element–binding protein (CREB protein).

b. A second effector of cAMP is Epac (exchange protein activated by cAMP), which has two isoforms. Epac proteins act as GEFs (see earlier) for small G proteins (called Raps). Raps in turn control a wide array of cell functions, including formation of cell-cell junctional complexes and cell-matrix adhesion, Ca²⁺ release from intracellular stores (especially in cardiac muscle), and in the augmentation of glucose-dependent insulin secretion by glucagon-like peptide-1 in pancreatic islet β cells (see Chapter 3).

c. cAMP (and cyclic guanosine monophosphate [cGMP], discussed later) also binds directly to and regulates ion channels. These are of two types: cyclic nucleotide gated (CNG) channels and hyperpolarization-activated cyclic nucleotide modulated (HCN) channels. For example, norepinephrine, which acts through a Gs-coupled receptor, increases heart rate in part through increasing a depolarizing inward K⁺ and Na⁺ current via an HCN at the sinoatrial node.

cGMP is produced from GTP by guanylyl cyclase, which exists in both transmembrane and soluble forms (Fig. 1.11). The transmembrane form of guanylyl cyclase is a hormone receptor, natriuretic peptide receptor (NPR-A and NPR-B), for the natriuretic peptides (atrial = ANP; brain = BNP; C-type = CNP). The soluble form of guanylyl cyclase is activated by another messenger, nitric oxide (NO). Nitric oxide is produced from molecular oxygen and arginine by the enzyme nitric oxide synthase (NOS). In vascular endothelial cells, endothelial NOS (eNOS) activity is the target of vasodilatory neuronal signals (e.g., acetylcholine) and certain hormones (estrogen). NO then diffuses into vascular smooth muscle and activates soluble guanylyl cyclase to produce cGMP. cGMP activates protein kinase G (PKG), which phosphorylates and regulates numerous proteins. In vascular smooth muscle, this leads to relaxation and vasodilation. As discussed earlier, cGMP also regulates ion channels. cAMP and cGMP are degraded to AMP and GMP, respectively, by phosphodiesterases (see Figs. 1.10 and 1.11), thereby terminating their signaling function. Phosphodiesterases represent a large family of proteins and display cell-specific expression. cAMP phosphodiesterases are inhibited by caffeine and other methylxanthines. cGMP is degraded by cGMP phosphodiesterases, of which one isoform is inhibited by sildenafil.

![Fig. 1.11 Membrane-bound and soluble guanylyl cyclases. R and C, Regulatory and catalytic subunits, respectively, of protein kinase G (PKG). eNOS, endothelial nitric oxide synthase; NO, nitric oxide; sGC, soluble guanylyl cyclase.](image-url)
(Viagra). In some contexts, cAMP and cGMP can modulate each other (a phenomenon called cross-talk) through the regulation of phosphodiesterases. For example, oocyte arrest is maintained by high levels of cAMP. The LH surge decreases cGMP in surrounding follicle cells by decreasing the local production of a natriuretic peptide. This results in lowered oocyte cyclic GMP. Because cGMP inhibits the oocyte cAMP-specific phosphodiesterase, lowered cGMP leads to decreased cAMP, thereby allowing the oocyte to complete the first meiotic division (see Chapter 10).

6. Generation of lipid informational molecules, which act as intracellular messengers. These include diacylglycerol (DAG) and inositol 1,4,5-triphosphate (IP$_3$), which are cleaved from phosphatidylinositol 4,5-bisphosphate (PIP$_2$) by membrane-bound phospholipase C (PLC). DAG activates certain isoforms of protein kinase C (Fig. 1.12). IP$_3$ binds to the IP$_3$ receptor, which is a large complex forming a Ca$^{2+}$ channel, on the endoplasmic reticulum membrane, and promotes Ca$^{2+}$ efflux (see later) from the endoplasmic reticulum into the cytoplasm. Some isoforms of DAG-activated PKC are also Ca$^{2+}$ dependent, so the actions of IP$_3$ converge on and reinforce those of DAG. The DAG signal is terminated by lipases, whereas IP$_3$ is rapidly inactivated by dephosphorylation.

7. Noncovalent Ca$^{2+}$ binding (see Fig. 1.12). Cytosolic levels of Ca$^{2+}$ are maintained at very low levels (i.e., 10$^{-7}$ to 10$^{-8}$ M), by either active transport of Ca$^{2+}$ out of the cell, or into intracellular compartments (e.g., endoplasmic reticulum). As discussed earlier, IP$_3$ binding to the IP$_3$ receptor increases the flow of Ca$^{2+}$ into the cytoplasm from the endoplasmic reticulum. Ca$^{2+}$ can also enter the cytoplasm through the regulated opening of Ca$^{2+}$ channels in the cell membrane. This leads to an increase in Ca$^{2+}$ binding directly to numerous specific effector proteins, which leads to a change in their activities. Additionally, Ca$^{2+}$ regulates several effector proteins indirectly, through binding to the signaling protein, calmodulin. Several of the Ca$^{2+}$/calmodulin targets are enzymes, which amplify the initial signal of increased cytosolic Ca$^{2+}$. The Ca$^{2+}$-dependent signal is terminated by the lowering of cytosolic Ca$^{2+}$ by cell membrane and endoplasmic reticular Ca$^{2+}$ ATPases (i.e., Ca$^{2+}$ pumps).

Transmembrane Receptors Using G Proteins

The largest family of hormone receptors is the G-protein-coupled receptor (GPCR) family. These receptors span the cell membrane seven times and are referred to as 7-helix transmembrane receptors. The G proteins that directly interact with GPCRs are termed heterotrimeric G proteins and are composed of an $\alpha$ subunit (Go), and a $\beta/\gamma$ subunit dimer (G$\beta/\gamma$). The Go subunit binds GTP and functions as the primary G-protein signal transducer. GPCRs are, in fact, ligand-activated GEFs (see earlier). This means that on hormone binding, the conformation of the receptor shifts to the active state. Once active, the GPCR induces the exchange of GDP for GTP, thereby activating G$\alpha$. One hormone-bound receptor activates 100 or more G proteins. GTP-bound Go then dissociates from G$\beta/\gamma$ and binds to and activates one or more effector proteins (Fig. 1.13).

How do G proteins link specific hormone receptor-binding events with specific downstream effector proteins? There are at least 16 Go proteins that show specificity with respect to cell-type expression, GPCR binding, and effector protein activation. A rather ubiquitous G$\alpha$ protein is called G$s$-$\alpha$, which stimulates the membrane enzyme, adenylyl cyclase, and increases the levels of another messenger,
cAMP (see earlier). Some GPCRs couple to $\text{Gi-}\alpha$, which inhibits adenyl cyclase. A third major hormonal signaling pathway is through $\text{Gq-}\alpha$, which activates phospholipase C (PLC). As discussed previously, PLC generates two lipid messengers, DAG and IP$_3$, from PIP$_2$. Defects in G-protein structure and expression are linked to endocrine diseases such as pseudohypoparathyroidism (loss of Gs activity) or pituitary tumors (loss of intrinsic GTPase activity in Gs, thereby extending its time in the active state).

GPCR-dependent signaling pathways regulate a broad range of cellular responses. For example, the pancreatic hormone, glucagon, regulates numerous aspects of hepatic metabolism (see Chapter 3). The glucagon receptor is linked to the Gs-cAMP-PKA pathway, which diverges to regulate enzyme activity at both posttranslational and transcriptional levels. PKA phosphorylates and thereby activates phosphorylase kinase. Phosphorylase kinase phosphorylates and activates glycogen phosphorylase, which catalyzes the release of glucose molecules from glycogen. Catalytic subunits of PKA also enter the nucleus, where they phosphorylate and activate the transcription factor, CREB protein. Phospho-CREB then increases the transcriptional rate of genes encoding specific enzymes (e.g., phosphoenolpyruvate carboxykinase).

In summary, signaling from one GPCR can regulate a number of targets in different cellular compartments with different kinetics (Fig. 1.14).

As mentioned, G-protein signaling is terminated by intrinsic GTPase activity, converting GTP to GDP. This returns the G protein to an inactive state (bound to GDP). Another termination mechanism involves desensitization and endocytosis of the GPCR (Fig. 1.15). Hormone binding to the receptor increases the ability of GPCR kinases (GRKs) to phosphorylate the intracellular domain of GPCRs. This phosphorylation recruits proteins called $\beta$-arrestins. GRK-induced phosphorylation and $\beta$-arrestin binding inactivate the receptor, and $\beta$-arrestin couples the receptor to clathrin-mediated endocytotic machinery. Some GPCRs are dephosphorylated and rapidly recycled back to the cell membrane (without hormone), whereas others are degraded in lysosomes. GRK/$\beta$-arrestin-dependent inactivation and endocytosis is an important mechanism for hormonal desensitization of a cell after exposure to excessive hormone. Hormone receptor endocytosis (also called receptor-mediated endocytosis) is also an important mechanism for clearing protein and peptide hormones from the blood.

Receptor Tyrosine Kinases

Receptor tyrosine kinases (RTKs) can be classified into two groups: the first acting as receptors for several growth factors (e.g., epidermal growth factor, platelet-derived growth factor), and the second group for insulin and IGFs. The former group of RTKs comprises transmembrane glycoproteins with an intracellular domain containing intrinsic tyrosine kinase activity. Growth factor binding induces dimerization of the RTK within the cell membrane, followed by transphosphorylation of tyrosine residues, generating phosphotyrosine (pY). The phosphotyrosines
function to recruit proteins. One recruited protein is phospholipase C, which is then activated by phosphorylation and generates the messengers DAG and IP$_3$ from PIP$_2$ (see earlier). A second critically important protein that is recruited to pY residues is the adapter protein, Grb2, which is complexed with a GEF named SOS. Recruitment of SOS to the membrane allows it to activate a small, membrane-bound monomeric G protein called Ras. Ras then binds to its effector protein, Raf. Raf is a serine-specific kinase that phosphorylates and activates the dual-function kinase, MEK. MEK then phosphorylates and activates a mitogen-activated protein kinase (MAP kinase, also called ERK). Activated MAP kinases then enter the nucleus and phosphorylate and activate several transcription factors. This signaling pathway is referred to as the MAP kinase cascade, and it transduces and amplifies a growth factor–RTK signal into a cellular response involving a change in the expression of genes encoding proteins involved in proliferation and survival.

The insulin receptor (IR) differs from growth factor RTKs in several respects. First, the latent IR is already dimerized by Cys-Cys bonds, and insulin binding induces a conformational change that leads to transphosphorylation of the cytoplasmic domains (Fig. 1.16). A major recruited protein to pY residues is the insulin receptor substrate (IRS), which is then phosphorylated on tyrosine residues by the IR. The pY residues on IRS recruit the Grb2-2/SOS complex, thereby activating growth responses to insulin through the MAP kinase pathway (see Fig. 1.16). The pY residues on the IRS also recruit the lipid kinase, PI3K, activating and concentrating the kinase near its substrate, PIP$_2$, in the cell membrane. As discussed earlier, this ultimately leads to activation of Akt/PKB, which is required for the metabolic responses to insulin (Fig. 1.17). The IR also activates a pathway involving the small G protein, TC-10 (see Fig. 1.17). The small G-protein-dependent pathway and the Akt/PKB pathway are both required for the actions of insulin on glucose uptake (see Chapter 3).

RTKs are downregulated by ligand-induced endocytosis. Additionally, the signaling pathways from RTKs, including IR and IRS, are inhibited by serine/threonine phosphorylation, tyrosine dephosphorylation, and the suppressor of cytokine signaling proteins (see next section).

Receptors Associated with Cytoplasmic Tyrosine Kinases
Another class of membrane receptor falls into the cytokine receptor family and includes receptors for GH, prolactin, erythropoietin, and leptin. These receptors, which
**Fig. 1.16** Signaling from the insulin receptor (a receptor tyrosine kinase) through the MAPK pathway. *pY*, Phosphorylated tyrosine residue in protein.

**Fig. 1.17** Signaling from the insulin receptor through the phosphatidylinositol-3-kinase (PI3K)/Akt/PKB pathway. *PIP_2*, Phosphatidylinositol 4,5-bisphosphate; *PIP_3*, Phosphatidylinositol 3,4,5 trisphosphate; *PKC*, protein kinase C; *pY*, phosphorylated tyrosine residue in protein; *R* and *C*, regulatory and catalytic subunits, respectively, of PI3K.

Cellular response
(Primarily mitogenic actions of insulin)

Cellular response
(primarily metabolic actions of insulin)
exist as dimers, do not have intrinsic protein kinase activity. Instead, the cytoplasmic domains are stably associated with members of the JAK kinase family (Fig. 1.18). Hormone binding induces a conformational change, bringing the two JAKs associated with the dimerized receptor closer together and causing their transphosphorylation and activation. JAKs then phosphorylate tyrosine residues on the cytoplasmic domains of the receptor. The pY residues recruit latent transcription factors called STAT (signal transducers and activators of transcription) proteins. STATs become phosphorylated by JAKs, which causes them to dissociate from the receptor, dimerize, and translocate into the nucleus, where they regulate gene expression.

A negative feedback loop has been identified for JAK/STAT signaling. STATs stimulate expression of one or more suppressors of cytokine signaling (SOCS) proteins. SOCS proteins compete with STATs for binding to the pY residues on cytokine receptors (Fig. 1.19). This terminates the signaling pathway at the step of STAT activation. Recent studies show that a SOCS protein is induced by insulin signaling. SOCS 3 protein plays a role in terminating the signal from the IR, but also in reducing insulin sensitivity in hyperinsulinemic patients.

**Receptor Serine/Threonine Kinase Receptors**

One group of transmembrane receptors are bound and activated by members of the transforming growth factor (TGF)-β family, which includes the hormones antimitlerian hormone and inhibin. Unbound receptors exist as dissociated heterodimers, called RI and RII (Fig. 1.20). Hormone binding to RII induces dimerization of RII with RI, and RII activates RI by phosphorylation. RI then activates latent transcription factors called Smads. Activated Smads heterodimerize with a Co-Smad, enter the nucleus, and regulate specific gene expression.

**Membrane Guanylyl Cyclase Receptors**

As discussed previously, the membrane-bound forms of guanylyl cyclase constitute a family of a receptors for natriuretic peptides (see Fig. 1.11). The hormonal role of atrial natriuretic peptide (ANP) will be discussed in Chapter 7.

**Signaling from Intracellular Receptors**

Steroid hormones, thyroid hormones, and 1,25-dihydroxyvitamin D act primarily through intracellular receptors. These receptors are structurally similar and are members of the nuclear hormone receptor superfamily that includes receptors for steroid hormones, thyroid hormone, lipid-soluble vitamins, peroxisome proliferator–activated receptors (PPARs), and other metabolic receptors (liver X receptor, farnesyl X receptor).

Nuclear hormone receptors act as transcriptional regulators. This means that the signal of hormone receptor binding is transduced ultimately into a change in the transcriptional rate of a subset of the genes that are expressed within a differentiated cell type. One receptor binds to a specific DNA sequence, called a hormone response element,
often close to the promoter of one gene, and influences the rate of transcription of that gene in a hormone-dependent manner (see later). However, multiple hormone receptor-binding events are collectively transduced into the regulation of several genes. Moreover, regulation by one hormone usually includes activation and repression of the transcription of many genes in a given cell type. Note that we have already discussed examples of signaling to transcription factors by transmembrane receptors. Table 1.2 summarizes the four general modes of hormonal regulation of gene transcription.

Nuclear hormone receptors have three major structural domains: an amino terminus domain (ATD), a middle DNA-binding domain (DBD), and a carboxyl terminus ligand-binding domain (LBD) (Fig. 1.21). The amino terminus domain contains a hormone-independent transcriptional activation domain. The DNA-binding domain contains two zinc finger motifs, which represent small loops organized by Zn\(^{2+}\) binding to four cysteine residues at the base of each loop. The two zinc fingers and neighboring amino acids confer the ability to recognize and bind to specific DNA sequences, which are called hormone-response elements (HREs). The carboxyl terminal ligand-binding domain contains several subdomains:

<table>
<thead>
<tr>
<th>Hormone Type</th>
<th>Steroid Hormones</th>
<th>Thyroid Hormones</th>
<th>Catecholamines, Peptides, Proteins</th>
<th>Catecholamines, Peptides, Proteins</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cell membrane</td>
<td>Passes through cell membrane</td>
<td>Passes through cell membrane, possibly use transporter</td>
<td>Binds to extracellular domain of transmembrane receptor</td>
<td>Binds to extracellular domain of transmembrane receptor</td>
</tr>
<tr>
<td>Cytoplasm</td>
<td>Binds to receptor, HRC translocates to nucleus</td>
<td>Moves through cytoplasm directly to nucleus to bind receptor</td>
<td>Ultimately activates cytoplasmic protein kinase, translocates to the nucleus</td>
<td>Activates a latent transcription factor in cytoplasm, TF translocates to the nucleus</td>
</tr>
<tr>
<td>Nucleus</td>
<td>HRC binds to response elements (often as dimer), recruits coregulatory proteins and alters gene expression</td>
<td>Hormone binds to receptor already bound to response elements, HRC induces exchange of coregulatory proteins, alters gene expression</td>
<td>Phosphorylates TF, which binds to DNA and recruits coregulatory proteins, alters gene expression</td>
<td>TF binds to DNA and recruits coregulatory proteins, alters gene expression</td>
</tr>
<tr>
<td>Examples</td>
<td>Cortisol</td>
<td>(T_3)</td>
<td>Glucagon</td>
<td>Growth hormone</td>
</tr>
</tbody>
</table>

HRC, Hormone-receptor complex; TF, transcription factor.
ATD (Amino Terminus Domain)
- Ligand-independent association with coregulatory proteins
- Ligand-independent phosphorylation sites

DBD (DNA Binding Domain)
- DNA binding via zinc finger domains
- Dimerization

LBD (Ligand Binding Domain)
- Ligand-binding
- Ligand-dependent association with coregulatory proteins
- Dimerization
- Nuclear translocation
- Association with chaperone proteins

Fig. 1.21 Domains of nuclear hormone receptor.

1. Site of hormone recognition and binding
2. Hormone-dependent transcriptional activation domain
3. Nuclear translocation signal
4. Binding domain for heat-shock proteins
5. Dimerization subdomain

There are numerous variations in the details of nuclear receptor mechanisms of action. Two generalized pathways by which nuclear hormone receptors increase gene transcription are the following (Fig. 1.22):

Pathway 1: Unactivated receptor is cytoplasmic or nuclear and binds DNA and recruits coactivator proteins on hormone binding. This mode is observed for the ER, PR, GR, MR, and AR (i.e., steroid hormone receptors). In the absence of hormone, some of these receptors are held in the cytoplasm through an interaction with chaperone proteins (so-called heat-shock proteins because their levels increase in response to elevated temperatures and other stresses). Chaperone proteins maintain the stability of the nuclear receptor in an inactive configuration. Hormone binding induces a conformational change in the receptor, causing its dissociation from heat-shock proteins. This exposes the nuclear localization signal and dimerization domains, so receptors dimerize and enter the nucleus. Once in the nucleus, these receptors bind to their respective HREs. The HREs for the PR, GR, MR, and AR are inverted repeats with the recognition sequence, AGAACANNNTGCT. Specificity is conferred by neighboring base sequences and possibly by receptor interaction with other transcriptional factors in the context of a specific gene promoter. The ER usually binds to an inverted repeat with the recognition sequence, AGGTCANNTGACCT. The specific HREs are also referred to as an estrogen-response element (ERE), progesterone-response element (PRE), glucocorticoid-response element (GRE), mineralocorticoid-response element (MRE), and androgen-response element (ARE). Once bound to their respective HREs, these receptors recruit other proteins, called coregulatory proteins, which are either coactivators or corepressors. Coactivators act to recruit other components of the transcriptional machinery and probably activate some of these components. Coactivators also possess intrinsic histone acetyltransferase (HAT) activity, which acetylates histones in the region of the promoter. Histone acetylation relaxes chromatin looping, making that region more accessible to transcriptional machinery. Although the mechanistic details are beyond the scope of this chapter, the student should appreciate that steroid receptors can also repress gene transcription through recruitment of corepressors that possess histone deacetylase (HDAC) activity and that transcriptional activation and repression pathways are induced concomitantly in the same cell. HDAC inhibitors are being studied in the context of treating some cancers because they restart the expression of silenced tumor suppressor genes.

Pathway 2: Receptor is always in nucleus and exchanges corepressors with coactivators on hormone binding. This pathway is used by the thyroid hormone receptors (THRs), VDRs, PPARs, and retinoic acid receptors. For example, the THR is bound, usually as a heterodimer, with the retinoic acid X receptor (RXR). In the absence of thyroid hormone, the THR/RXR recruits corepressors. As stated earlier, corepressors recruit proteins with histone deacetylase (HDAC) activity. In contrast to histone acetylation, histone deacetylation allows tighter looping of chromatin, which makes promoters in that region less accessible to the transcriptional machinery. Thus THR/RXR heterodimers are bound to thyroid hormone response elements (TREs) in the absence of hormone and maintain the expression of neighboring genes at a “repressed” level. Thyroid hormone (and other ligands of this class) readily move into the nucleus and bind to their receptors. Thyroid hormone binding induces dissociation of corepressor proteins, thereby increasing gene expression to a basal level. The hormone receptor complex subsequently recruits coactivator proteins, which further increase transcriptional activity to the “stimulated” level.

Termination of steroid hormone receptor signaling is poorly understood but appears to involve phosphorylation, ubiquitination, and proteosomal degradation. Circulating steroid and thyroid hormones are cleared as described previously.

In summary, hormones signal to cells through membrane or intracellular receptors. Membrane receptors have rapid effects on cellular processes (e.g., enzyme activity, cytoskeletal arrangement) that are independent of new
Fig. 1.22 Two general mechanisms by which nuclear receptor and hormone complexes increase gene transcription. Coact, Coactivator proteins; corepress, corepressor proteins; GTFs, general transcription factors; HR, hormone receptor; HRE, hormone response element; RXR, retinoid X receptor.
protein synthesis. Membrane receptors can also rapidly regulate gene expression through either mobile kinases (e.g., PKA, MAPKs) or mobile transcription factors (e.g., STATs, Smads). Steroid hormones have slower, longer-term effects that involve chromatin remodeling and changes in gene expression. Increasing evidence points to rapid, nongenomic effects of steroid hormones as well, but these pathways are still being elucidated.

The presence of a functional receptor is an absolute requirement for hormone action, and loss of a receptor produces essentially the same symptoms as loss of hormone. In addition to the receptor, there are fairly complex pathways involving numerous intracellular messengers and effector proteins. Accordingly, endocrine diseases can arise from abnormal expression or activity of any of these signal transduction pathway components.

Overview of the Termination Signals

Most of what has been discussed in this chapter describes the stimulatory arm of signal transduction. As noted earlier, all signal transduction of hormonal signals must have termination mechanisms to avoid sustained and uncontrolled stimulation of target cells. Part of this stems from the cessation of the original stimulus for increasing a hormone’s level, and mechanisms to clear the hormone (i.e., removal of signal). However, there exists a wide array of intracellular mechanisms that terminate the signaling pathway within the target cells. Some of these are listed in Table 1.3. Note that overactivity of terminating mechanisms can lead to hormonal resistance.

### Table 1.3 Some Modes of Signal Transduction Termination

<table>
<thead>
<tr>
<th>Mechanism of Signal Transduction Termination</th>
<th>Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Receptor-mediated endocytosis linked to lysosomal degradation</td>
<td>Many transmembrane receptors</td>
</tr>
<tr>
<td>Phosphorylation/dephosphorylation of receptor or “downstream” components of signaling pathway</td>
<td>Serine phosphorylation of insulin receptor and insulin receptor substrate by other signaling pathways</td>
</tr>
<tr>
<td>Ubiquitination/proteasomal degradation</td>
<td>Steroid hormone receptors</td>
</tr>
<tr>
<td>Binding of an inhibitory regulatory factor</td>
<td>Regulatory subunit of PKA</td>
</tr>
<tr>
<td>Intrinsic terminating enzymatic activity</td>
<td>GTPase activity of G proteins</td>
</tr>
</tbody>
</table>

**SUMMARY**

1. The endocrine system is composed of:
   - Dedicated hormone-producing glands (pituitary, thyroid, parathyroid, and adrenal)
   - Hypothalamic neuroendocrine neurons
   - Scattered endocrine cells that exist as clusters of endocrine-only cells (islets of Langerhans) or as cells within organs that have a nonendocrine primary function (pancreas, GI tract, kidney)
   - Testes and ovaries, whose intrinsic endocrine function is absolutely necessary for gametogenesis
2. Endocrine signaling involves the secretion of a chemical messenger, called a hormone, that circulates in the blood and reaches an equilibrium with the extracellular fluid. Hormones alter many functions of their target cells, tissues, and organs through specific, high-affinity interactions with their receptors.
3. Protein/peptide hormones:
   - Are produced on ribosomes, become inserted into the cisternae of the endoplasmic reticulum, transit the Golgi apparatus, and finally are stored in membrane-bound secretory vesicles. The release of these vesicles represents a regulated mode of exocytosis. Each hormone is first made as a prehormone, containing a signal peptide that guides the elongating polypeptide into the cisternae of the endoplasmic reticulum.
   - Are frequently synthesized as preprohormones. After removal of the signal peptide, the prohormone is processed by prohormone convertases.
   - Typically do not cross cell membranes and act through transmembrane receptors (see later).
   - Mostly circulate as free hormones, and are excreted in the urine or cleared by receptor-mediated endocytosis and lysosomal degradation.
4. Catecholamine hormones:
   - Include the hormones, epinephrine (Epi) and norepinephrine (Norepi). Epi and Norepi are derivatives of tyrosine, which is enzymatically modified by several reactions. Ultimately, Epi and Norepi are stored in a secretory vesicle and are released through regulated exocytosis.
• Act through transmembrane GPCRs receptors called **adrenergic receptors**.

5. Steroid hormones:
   • Include cortisol (glucocorticoid), aldosterone (mineralocorticoid), testosterone, and dihydrotestosterone (androgens), estradiol (estrogen), progesterone (progesterone), and 1,25 dihydroxyvitamin D3 (secosteroid).
   • Are derivatives of cholesterol, which is modified by a series of cell-specific enzymatic reactions.
   • Are lipophilic and cross membranes readily. Thus steroid hormones cannot be stored in secretory vesicles. Steroid production is regulated at the level of synthesis. Several steroid hormones are produced to a significant extent by peripheral conversion of precursors.
   • Circulate bound to transport proteins. Steroid hormones are cleared by enzymatic modifications that increase their solubility in blood and decrease their affinity for transport proteins. Steroid hormones and their inactive metabolites are excreted in the urine.
   • Act through intracellular receptors, which are members of the nuclear hormone receptor family. Most steroid hormone receptors reside in the cytoplasm and are translocated to the nucleus after ligand (hormone) binding. Each steroid hormone regulates the expression of numerous genes in their target cells.

6. Thyroid hormones are:
   • **Iodinated derivatives of thyronine**. The term thyroid hormone typically refers to 3,5,3′,5′-tetraiodothyronine (T4, or thyroxine) and 3,5,3′-triiodothyronine (T3). T3 is an inactive precursor of T4, which is produced by 5′-deiodination of T4.
   • Synthesized and released by the thyroid epithelium (see Chapter 6 for more detail)
   • Circulate tightly bound to **transport proteins**
   • Lipophilic and cross cell membranes. T3 binds to one of several isoforms of thyroid hormone receptors (TRs), which form heterodimers with retinoid X receptor (RXR) and reside bound to their response elements in the nucleus in the absence of hormone. Hormone binding induces an exchange in the coregulatory proteins that interact with the TRs.

7. Protein, peptide, and catecholamine hormones signal through transmembrane receptors and use several common forms of informational transfer:
   • Conformational change
   • Binding by activated G proteins
   • Binding by Ca2+ or Ca2+-calmodulin. IP3 is a major lipid messenger that increases cytosolic Ca2+ levels through binding to the IP3 receptor.
   • Phosphorylation and dephosphorylation, using kinases and phosphatases, respectively. The phosphorylation state of a protein affects activity, stability, subcellular localization, and recruitment binding of other proteins. Note that phosphorylated lipids such as PIP2 also play a role in signaling.

8. Transmembrane receptor families:
   • **G-protein-coupled receptors** (GPCRs) act as guanine nucleotide exchange factors (GEFs) to activate the Ga subunit of the heterotrimeric α/β/γ G-protein complex. Depending on the type of Ga subunit that is activated, this will **increase cAMP levels**, **decrease cAMP levels**, or **increase protein kinase C activity and Ca2+ levels**. All catecholamine receptors (adrenergic receptors) are GPCRs. GPCRs are internalized by a receptor-mediated endocytosis that involves **GRK** and **β-arrestin**. Endocytosis results in the lysosomal clearance of the hormone. The receptor may be digested in the lysosome or may be recycled to the cell membrane.
   • The **insulin receptor** is a tyrosine kinase receptor that activates the Akt/PKB pathway, the G-protein TC10-related pathway, and the MAPK pathway. The insulin receptor uses the scaffolding protein **insulin receptor substrate (IRS; four isoforms)** as part of its signaling to these three pathways.
   • Some protein hormones (e.g., growth hormone, prolactin) bind to transmembrane receptors that belong to the **cytokine receptor family**. These are constitutively dimerized receptors that are bound by janus kinases (JAKs). Hormone binding interacts with both extracellular domains and induces JAK-JAK cross-phosphorylation, followed by recruitment and binding of **STAT proteins**. Phosphorylation of STATs activates them and induces their translocation to the nucleus, where they act as transcription factors.
   • Hormones that are related to transforming growth factor-β (TGF-β), such as antimüllerian hormone, signal through a coreceptor (receptor I and receptor II) complex that ultimately signals to the nucleus through activated Smad proteins.
   • **Atrial natriuretic peptide** (and related peptides) bind to a transmembrane receptor that contains a guanylyl cyclase domain within the cytosolic domain. These receptors signal by increasing cGMP, which activates protein kinase G (PKG) and cyclic nucleotide-gated channels. cGMP also regulates selective **phosphodiesterases**.

9. Intracellular Receptors
   • **Steroid hormones** bind to members of the **nuclear hormone transcription factor family**. Steroid
hormone receptors usually reside in the cytoplasm. Hormone binding induces **nuclear translocation**, **dimerization**, and **DNA binding**. Steroid hormone receptor complexes regulate many genes in a target cell.

**SELF-STUDY PROBLEMS**

1. How do protein hormones differ from steroid hormones in terms of their storage within an endocrine cell?
2. How does binding to serum transport proteins influence hormone metabolism and hormone action?
3. How would a large increase in the GTPase activity of Gs-α affect signaling through GPCRs linked to Gs-α?
4. What role does the IRS protein play in transducing insulin receptor signaling into a growth response? A metabolic response?
5. Name an example of a transmembrane receptor–associated transcription factor that translocates to the nucleus.
6. Explain the mechanism of receptor-mediated endocytosis of a hormone that binds to a GPCR.
7. What is the importance of the GEF activity of a GPCR to its ability to signal?
8. Explain how PLC generates two second messengers.

**KEY WORDS AND CONCEPTS**

- 7-Helix transmembrane receptors
- Adenyl cyclase
- Adrenal cortex
- Agonist
- Androgen
- Androgen receptor
- Androgen response element (ARE)
- Antagonist
- β-Arrestins
- Ca²⁺
- Ca²⁺ ATPases
- Ca²⁺ channels
- Calmodulin
- cAMP phosphodiesterase
- cAMP response element–binding protein (CREB)
- Catecholamine
- Cellular response
- cGMP phosphodiesterase
- Circadian (diurnal) rhythms
- Coactivator proteins
- Corepressors
- Corticosteroid-binding globulin
- Covalent phosphorylation of proteins and lipids
- Cyclic AMP
- Cyclic GMP
- Cyclic nucleotide monophosphates
- Cycloperhydrophenanthrene ring
- Cytokine receptor family
- Diacylglycerol (DAG)
- Docking protein
- Effector proteins
- Eicosanoids

- Endocrine gland
- Endocrine system
- Epinephrine
- Estrogen
- Estrogen receptor
- Estrogen response element (ERE)
- Exocrine gland
- Exocytosis
- G-protein exchange factor (GEF)
- Ga
- Gi-α
- Glucocorticoid
- Glucocorticoid receptor
- Glucocorticoid response element (GRE)
- Glucuronide conjugation
- GPCR kinase (GRK)
- G-protein-coupled receptor (GPCR)
- Gα
- Grb2/SOS
- Gs-α
- GTP-binding proteins (G proteins)
- Guanylyl cyclase
- Gβ/γ
- Heterotrimeric G proteins
- High-affinity receptor
- Histone acetyltransferase (HAT)
- Histone deacetylase (HDAC)
- Hormonal desensitization
- Hormonal resistance
- Hormone
- Hormone response elements (HREs)
- Inositol 1,4,5-triphosphate (IP₃)
Insulin receptor (IR)
Insulin receptor substrate (IRS)
Intracellular messengers
Intrinsic GTPase activity
Iodothyronine
JAK kinase family
Leukotrienes
Ligand
Ligand-activated GEF
Ligand-induced endocytosis
MEK
Mineralocorticoid
Mineralocorticoid receptor
Mineralocorticoid response element (MRE)
Mitogen-activated protein kinase (MAPK)
Mixed-function kinases and phosphatases
Nitric oxide (NO)
Norepinephrine
Nuclear receptor superfamily
Ovary
Peripheral conversion
Phosphatidylinositol 3,4,5-triphosphate (PIP$_3$)
Phosphatidylinositol-3-kinase (PI3K)
Phospholipase C
Phosphotyrosine (pY)
PKA catalytic subunit
PKA regulatory subunit
Placenta
Prehormone
Preprohormone
Progesterone receptor
Progesterone response element (PRE)
Progestin
Prohormone convertase
Prostacyclin
Prostaglandins
Protein kinase A (PKA)
Protein kinase B (PKB/Akt)
Protein kinase G (PKG)
Protein/peptide hormone
Raf
Ras
Receptor
Receptor serine/threonine kinases
Receptor tyrosine kinases (RTKs)
Regulated secretory pathway
Regulators of G-protein signaling (RGS proteins)
Second messenger hypothesis
Serine/threonine-specific kinases and phosphatases
Set-point
Sex hormone–binding globulin
Signal peptidase
Signal peptide
Signal recognition complex
Signal transduction pathway
Smads
STAT
Steroid hormone
Steroidogenic cells
Stimulus-secretion coupling
Sulfate conjugation
Suppressors of cytokine signaling (SOCS) proteins
Target cell
Target organ
Testis
Thromboxanes
Thyroid hormone receptor
Thyroid hormone–binding globulin
Thyroid hormone–response element (TRE)
Transforming growth factor (TGF)-β family
Transport proteins
Tyrosine kinases and phosphatases
Ultradian rhythms
Vitamin D
Vitamin D receptor
Vitamin D response element (VRE)