I. INTRODUCTION
   a. The work hormone comes from the Greek *hormaein*, which means “to excite”, or “to stir up”. The classical definition of a hormone is a chemical substance that is released into the blood stream in small amounts and is delivered by the circulatory system to its target cells where it elicits a typical response. However, it is now recognized that there are a whole spectrum of hormones or hormone-like substances that are involved in cell-to-cell communication that are not secreted directly into the blood stream. They reach target cells by diffusion through interstitial fluids.

   b. The classical endocrine glands are
      i. Adrenal gland
      ii. Islets of Langerhans
      iii. Gonads
      iv. Pituitary gland
      v. Parathyroid glands
      vi. Placenta
      vii. Thyroid gland

   c. The classic hormones can be divided into three categories
      i. Amino acids and their derivatives – epinephrine and other catecholamines, dopamine. While thyroid hormones are considered amine hormones, they have little physiologic similarity to the other hormones in this class aside from similar precursors. They are most similar to the steroid hormones and will be discussed below.
         1. Four hormones are known – epinephrine, norepinephrine, dopamine and serotonin. They are synthesized from tyrosine (first 3) and tryptophan (serotonin).
         2. These hormones do not have a hierarchic “feedback system”. Their release is determined by a physiologic “end effect” of the hormones e.g. blood pressure. They are stored in secretory granules called chromaffin granules. They do not have binding proteins, a reflection of their relatively high water solubility.
         3. Amine hormones act via cell surface receptors which are G-protein linked to cAMP, in general.
      ii. Steroids – i.e. cortisol, testosterone, estradiol
         1. All steroid hormones share a common precursor – cholesterol. Their synthesis is regulated by a series of enzymatic pathways, which are present only in the adrenal gland or the gonads.
2. Steroid hormones are not stored in secretory granules. Their synthesis is closely linked temporally to their secretion. They usually have specialized binding proteins, which carry these relatively water-insoluble hormones to their target organs.

3. Steroid hormones enter cells by simple diffusion and bind to receptors located in the cytosol and/or nucleus which translocate to the nucleus and interact with specific DNA sequences called steroid response element (SRE’s) stimulating transcription of the appropriate gene.

4. Thyroid hormones, which do not share structural homology with steroid hormones, interact with their receptors, which are similar to steroid receptors, in a mechanism very similar to steroid hormones. The one difference is that the thyroid hormone receptors are located in the nucleus. Once the thyroid hormone binds to the receptor, this complex interacts with the thyroid response elements of thyroid-responsive genes thereby regulating gene transcription.

iii. Peptides and proteins i.e. insulin, growth hormone, oxytocin, angiotensin, parathyroid hormone

1. Peptide hormones are produced by transcription of mRNA in the nuclei of specialized endocrine cells. They move to the cytosol where they associate with ribosomes of the rough endoplasmic reticulum and move to the Golgi and eventually enter and are stored in secretory granules. The contents of the secretory granules are discharged by regulated pathways initiated by triggering external stimuli. A second constitutive pathway of secretion may also occur.
more directly from the Golgi which is less responsive to secretory stimuli.

2. Peptide hormones bind to cell surface receptors, which activate a variety of signal transduction systems - ie. G-protein/cAMP, G-protein/phospholipase C, G-protein/phospholipase A₂, cGMP, tyrosine kinase, and tyrosine kinase-associated systems as well as ion channels.

II. GENERAL CHARACTERISTICS OF HORMONE SYSTEMS - Necessary components of any endocrine regulatory system
   a. Ability to detect an actual or threatened homeostatic imbalance
   b. Coupling mechanism to activate the secretory apparatus
   c. Secretory apparatus
   d. Hormone
   e. End-organ capable of responding to the hormone
   f. Detection system which can recognize that the hormonal effect has occurred and that the hormone signal can be terminated
   g. Mechanism for removing the hormone from the target cells and blood
   h. Synthetic apparatus to replenish the hormone in the secretory cell

III. HORMONES IN THE BLOOD
   a. Hormones are secreted into the extracellular fluid and can enter the bloodstream by passive diffusion down steep concentration gradients or by an active energy-requiring process. The hormones distribute rapidly throughout the extracellular fluid and are not preferentially directed toward the target cells. Diffusion through capillary pores largely accounts for the delivery of hormones to the target cells.
   b. Hormone secretion can be episodic, pulsatile, or follow a daily or circadian rhythm. This may make it necessary to make multiple serial measurements of a hormone before a diagnosis of an altered hormonal state can be made.
   c. Endocrine-related diseases occur when the hormone concentration in the blood is inappropriate for the existing physiologic situation. They are not necessarily related to the absolute amounts of hormone in the blood.
   d. Most amine, peptide and protein hormones are readily soluble in plasma and no special mechanisms are required for their transport. There may be exceptions – i.e. growth hormone that has its own binding protein.
   e. In contrast, about 90% of the steroid or thyroid hormones are carried or bound to plasma protein. Only about 1-10% of the hormone is free in solution. Only the free hormone can interact with a receptor. This interaction or binding is a dynamic equilibrium.

IV. GENERAL ACTIONS OF HORMONES
   a. The cell rather than the hormone determines the specific response elicited in a given cell. Different cell types may respond to the same stimulus in different ways. Example: Vasopressin affects both renal tubule collecting ducts and vascular smooth muscle. In the kidney, it increases the permeability of the collecting ducts to water; it causes vasoconstriction in vascular smooth muscle.
b. Only cells that have the specific receptor for a particular hormone will be able to respond to that hormone. Specificity for the hormone action therefore resides at the level of the receptors.

c. Some hormones have a more or less restricted distribution. Example: Some hypothalamic hormones are carried by small blood vessels directly to the anterior pituitary.

d. Hormones can be involved in chemical signalling via classic endocrine pathways or paracrine or autocrine pathways.
   i. Endocrine pathways involve the secretion of a hormone by a secretory cell, which then travels a large distance in the systemic circulation to its target gland.
   ii. Autocrine pathways regulate the secretory cell itself by the hormone it secreted.
   iii. Paracrine pathways involve regulation of nearby cells by hormones without entering the systemic circulation.

V. FEEDBACK REGULATION

a. Endocrine cells, in addition to having the ability to synthesize and secrete their hormones, also have the ability to sense the biological consequences of secretion of that hormone.

b. First order negative feedback is the simplest type and forms the basis for more complex modes of regulation.

c. More commonly, second- and third-order feedback loops are present. These systems can provide better fine-tuning of the control system and provide some redundancy with a degree of “fail-safe” mechanisms if one or more components of the system fail.

Example of theoretical 2nd and 3rd order control of feedback loops. Question marks represent proposed feedback to the CNS by one or more of the hormones secreted in the cascade – ie. Hypothalamic releasing hormones, trophic hormones from the pituitary as well as the hormone secreted by the target gland.
VI. HORMONE DEGRADATION
   a. Biological effects of hormones do not generally “use-up” the hormone, so mechanisms must exist to degrade them once they have conveyed their information.
   b. This degradation may occur by hydrolysis by degradative enzymes, oxidation, reduction, aromatization, deiodination, conjugation with glucuronide, methylation and other methods. Depending on the hormone, degradation or clearance of the hormone may occur in a variety of organs most commonly the liver, second most commonly the kidney but also the lung, muscles, blood as well as the target organ itself. Often, recognizable end products appear in the urine or blood and can be measured as an index of hormone production.
   c. The half-lives of different hormones vary considerably, ranging from 5 minutes to several hours.

VII. MODULATION OF THE HORMONAL MESSAGE - The hormonal message is subject to modifications that may occur anywhere from its initial synthesis to its final arrival at its target site. Subsequently, the expression of the message at this site (i.e., its action) may also be modified. The modulations or alterations of the hormonal message or its final action may be physiologic or pathologic. They include
   a. Gene mutations which may effect the synthesis of the hormone, its processing or its receptor.
   b. Chromosomal deletions e.g. Turner’s syndrome which is the result of partial or complete X-chromosomal deletion.
   c. Alternative gene processing can result in two different hormones. For example, two distinct TSH receptors can arise from one gene; & the synthesis of calcitonin and the significantly different calcitonin-gene-related peptide (CGRP), which has different biologic activities, from the same gene.
   d. Posttranslational modification such as occurs in post-translational glycosylation of pituitary glycoprotein hormones e.g. TSH, LH and FSH.
   e. Alterations of binding proteins e.g. increases in thyroid binding globulin seen in pregnancy result in increased levels of total (but not free) thyroid hormone.
   f. Endogenous hormone antagonists.
   g. Antibodies to hormones or their receptors e.g. classic example is Grave’s disease, which is characterized by activating autoantibodies to the TSH receptor resulting in hyperthyroidism.
   h. Receptor alterations or postreceptor changes, which alter signal transduction (biochemical events beyond receptor binding). Some of the major pathophysiologic alterations in type 2 diabetes fall into this category.
   i. Hormone clearance (see above under degradation)
VIII. MEASUREMENT OF HORMONES AND THEIR ACTIVITY

a. Radioimmunoassay (RIA) - The discovery and development of the radioimmunoassay for insulin resulted in the Nobel Prize in Medicine being awarded in 1977 to Dr. Rosalind Yallow for work done by her and Dr. Solomon Berson (who died before the award was bestowed) in the 1950’s. The development of the RIA technique helped advance endocrinology as a separate subspecialty of medicine. This competitive binding assay technique allowed for measurement of very small concentrations of a hormone.

b. Bioassays – one of the earliest methods for measuring hormone activity. In these assays, the sample to be studied interacts with a biological system with a measurable biologic effect. An example of this type of assay are mouse calvarial cell systems which are used to detect and quantify the stimulation of bone resorption ability by biologic fluids.

c. Dynamic testing – These tests are either stimulation or suppression tests and are used clinically to investigate syndromes of hormone excess (suppression tests) or hormone deficiency (stimulation test). An example here is the dexamethasone suppression screening test for Cushing’s syndrome – a syndrome of cortisol excess. In this test, dexamethasone, a synthetic glucocorticoid which is not measured in the cortisol assay, is given to patients to suppress the pituitary axis (feedback inhibition) and cortisol is measured the next morning. If endogenous cortisol cannot be suppressed, the patient may have Cushing’s syndrome.

IX. MEASUREMENT OF HORMONE RESPONSIVENESS

a. Maximum responsiveness - The effect obtained at saturating the receptor

b. Sensitivity – The hormone concentration required for half-maximal response - index of sensitivity of target cell

c. Threshold concentration - Hormone concentration required to elicit a measurable response

“IT is summarized in the competing reactions shown (to the left). The concentration of the unknown unlabeled antigen is obtained by comparing its inhibitory effect on the binding of radioactively labeled antigen to specific antibody with the inhibitory effect of known standards”

Dr. Rosalyn Yallow, Nobel Lecture December 8, 1977
Hormone Responsiveness

X. OUTLINE FOR ORGANIZING HORMONE INFORMATION
a. Be familiar with
   i. Essential features of feedback regulation
   ii. Essentials of competitive binding assays
b. For each hormone, be familiar with
   i. Its cell of origin
   ii. Its chemical nature, including
       1. Distinctive features of its chemical composition
       2. Biosynthesis
       3. Whether it circulates free or bound to plasma proteins
       4. How it is degraded and removed from the body
   iii. Its principle physiologic actions
       1. At the whole body level
       2. At the tissue level
       3. At the cellular level
       4. At the molecular level
       5. Consequences of inadequate or excess secretion
   iv. What signals or perturbations in the internal or external environment evoke or suppress its secretion
       1. How the signals are transmitted
       2. How that secretion is controlled
       3. What factors modulate the secretory process
       4. How rapidly the hormone acts
       5. How long it acts
       6. What factors modulate is actions
Chapter 2 The Hypothalamus and the Anterior Pituitary Gland
Mary Zoe Baker, M.D.

Reading: Costanzo, p.387-400.

I. INTRODUCTION - The hypothalamus and the pituitary gland work closely together to control and coordinate many of the endocrine systems. The pituitary gland consists of an anterior lobe and a posterior lobe. This next section will deal exclusively with the anterior pituitary gland

II. ANTERIOR PITUITARY HORMONES
   a. The anterior pituitary gland (aka – anterior lobe of the pituitary, adenohypophysis) consists of histologically distinct types of cells closely associated with blood sinusoids that drain into the venous circulation,
   b. There are 6 well-known anterior pituitary hormones produced by separate kinds if cells. All are small proteins. They are:
      i. Adrenocorticotrophic hormone (ACTH) secreted by corticotrophs. (30%)
      ii. Thyroid stimulating hormone (TSH) secreted by thyrotrophs (5%)
      iii. Growth hormone (GH) secreted by somatotrophs (20%)
      iv. Prolactin (PRL) secreted by lactotrophs (15%)
      v. Follicle-stimulating hormone (FSH) secreted by gonadotrophs (15%)
      vi. Luteinizing hormone (LH) secreted by gonadotrophs (15%)
   c. These hormones can be grouped into “families” according to their structural and functional homology
      i. TSH, FSH and LH family are all glycoproteins with sugar moieties covalently linked to asparagine residues in their polypeptide chains. Each hormone consists of two subunits, α & β, which are not covalently linked. The α subunits are identical; the β subunits are unique and give each hormone its biologic specificity.
      ii. ACTH family is all derived from the pro-opiomelanocortin (POMC) gene. The family includes ACTH, γ− & β-lipotropin, β-endorphin and MSH. Only ACTH has established physiologic actions in humans

III. HYPOTHALAMIC RELEASING HORMONES
   a. The secretory activity of these hormones is regulated by specific releasing factors, hormones synthesized by neuroendocrine cells in the hypothalamus as well as neurotransmitters from the CNS.
      i. Corticotropin-releasing hormone (CRH or CRF) => ACTH
      ii. Thyrotropin-releasing hormone (TRH or TRF) => TSH
      iii. Gonadotropin-releasing hormone (GnRH) => FSH and LH
      iv. Growth hormone-releasing hormone (GHRH) => GH
      v. Growth hormone-inhibiting hormone (GHRIH or somatostatin (preferred)) => inhibits GH
   b. The releasing hormones are secreted from hypothalamus in response to neural inputs from other areas of the central nervous system. These inputs are generated by changes in either the external or internal environment of the body
c. Depending on the nature of the event and the signal generated, the secretion of a particular releasing hormone may be either stimulated or inhibited which, in turn, affects the rate of secretion of the associated anterior pituitary hormone.
d. Four of the anterior pituitary hormones have effects on the morphology and secretory activity of other endocrine glands. They are called trophic hormones because they turn on or nourish other endocrine cells.
   i. ACTH maintains the size of certain cells in the adrenal cortex and stimulates release of several steroid hormones.
   ii. TSH maintains the size of the thyroid follicles and stimulates the release of thyroid hormones.
   iii. FSH and LH both act on the ovaries and testes
e. The two remaining anterior pituitary hormones are not generally thought of as trophic hormones.
   i. GH directly stimulates the growth of the body during childhood and affects many cell types.
   ii. Prolactin is essential for the synthesis of milk during lactation.

IV. ADENOCORTICOTROPHIC HORMONE (ACTH)
a. ACTH is the physiologic regulator of the synthesis and secretion of glucocorticoids by the zona fasiculata and zona reticularis of the adrenal cortex.
b. ACTH stimulation the synthesis of these steroid hormones and promotes the expression of the genes for various enzymes involved in steroidogenesis. It also maintains the size and functional integrity of the cells of the zona fasiculata and zona reticularis.

c. Corticotropin-releasing hormone (CRH or CRF) is synthesized in cells located in the paraventricular nuclei of the hypothalamus. Secretory granules containing CRH are stored in the axon terminal and released when an appropriate stimulus is received. CRH is carried to the anterior pituitary by the hypophyseal portal circulation.

d. CRH binds to membrane receptors that are coupled to adenylate cyclase by stimulatory G proteins (G_s). Adenylate cyclase is stimulated and cAMP rises in the cell. cAMP activates protein kinase A (PKA) which then phosphorylates the P-proteins involved in stimulating ACTH secretion and the expression of the pro-opiomelanocortin (POMC) gene. The POMC gene and is associated mRNA is involved in maintaining the capacity of the cell to synthesize the precursor for ACTH.

e. The increase in glucocorticoid concentration in the blood resulting from the action of ACTH on the adrenal cortex inhibits the secretion of ACTH and CRH (negative feedback).

f. Stress (physical or emotional) increases the secretion of ACTH regardless of the level of glucocorticoids present. Enhanced neural activity associated with the stress increases the secretion of CRH and thereby increases ACTH secretion. If the stress persists, glucocorticoid levels remain high and, in effect, the set point of the hypothalamic-pituitary-adrenal axis is changed.

g. This interactive relationship is called the hypothalamic-pituitary-adrenal axis and negative feedback is essential for normal operation of this system.

h. Glucocorticoid deficiency and certain stresses also increase the concentration of antidiuretic hormone (ADH or AVP) in the portal blood. ADH also has the capacity to stimulate the secretion of ACTH thus amplifying the effect of CRH.
i. ACTH is also implicated in the normal sleep-wake cycle of humans (the circadian rhythm). The hypothalamic-pituitary-adrenal axis functions in a pulsatile manner that results in a number of bursts of activity over a 24-hour period. This appears to be due to rhythmic activity in the central nervous system itself which then affects CFH secretion.

Normally, there is nocturnal peak (generally just before waking) of ACTH that is driven by a burst of CRH secretion. This is followed by an increase in plasma cortisol (see left). The timing of this cyclic activity can be shifted by alterations in the normal sleep-wake cycle (e.g., in individuals who chronically work night shifts. This diurnal pattern is not present in comatose patients, in blind individuals or if an individual is constantly exposed to either light or dark. This peak may also be blunted in Cushing’s syndrome.

j. Glucocorticoid Deficiency States – Three clinical presentations
   i. Removal of the adrenal glands or damage caused by disease (e.g., Addison’s disease or TB – not often seen now), stimulates the release of large amounts of ACTH, which can result in hyperpigmentation because ACTH in high concentrations stimulates melanocyte activity.
   ii. Individual with inherited glucocorticoid deficiency due to lack of the ability to produce certain necessary enzymes have high blood levels of ACTH due to the lack of negative feedback on ACTH secretion. The result is hypertrophy of the adrenal glands and this collection of genetic disease are collectively called congenital adrenal hyperplasia – CAH
   iii. Adrenal cortex atrophy occurs in individuals treated chronically with large doses of glucocorticoids because ACTH secretion is inhibited and its trophic influence on the adrenal cortex is lost. This atrophy can be reversed but can take up to a year, depending upon the dose, duration or potency of the glucocorticoid.

V. THYROID STIMULATING HORMONE (TSH)
   a. The follicular cells of the thyroid gland produce and secrete L-thyroxine – (T4) and triiodothyronine (T3). These thyroid hormones act on many cells to change the expression of certain genes and thereby change the capacity of the target cells to produce particular proteins.
   b. TSH is the physiologic regulator of T4 and T3 synthesis and secretion. It also promotes nucleic acid and protein synthesis in the thyroid follicular cells and this maintains the size and integrity of those cells.
c. TRH is the main physiologic stimulator of TSH synthesis and secretion and is produced by neurons in various areas of the hypothalamus. These neurons secrete TRH at a relatively constant rate and therefore the concentration of TRH perfusing the follicular cells does not change greatly.

d. TRH binds to membrane receptors that are coupled to phospholipase C (PLC) by G proteins ($G_\text{p}$). PLC hydrolyzes phosphatidylinositol 4,5 (PIP$_2$) in the plasma membrane, generating inositol triphosphate (IP$_3$) and diacylglycerol (DAG). IP$_3$ mobilizes intracellularly bound Ca$^{2+}$. The rise in Ca$^{2+}$ stimulates TSH secretion proteins (P-proteins) involved in stimulating TSH secretion and the expression of the genes for the $\alpha$ & $\beta$ subunits of TSH.

e. Although TRH has a tonic stimulatory effect on TSH secretion, the main factor that regulates TSH secretion is the concentration of thyroid hormones in the blood.

f. Increased levels of free T$_3$ in the blood downregulate the TRH receptors on the anterior pituitary thyrotrophs. This system is called the hypothalamic-pituitary-thyroid axis.

g. Thyroid hormone affects many physiologic functions and organ systems
  i. **Basal metabolic rate** – thyroid hormones increase oxygen consumption in all tissues *except brain, gonads and spleen* by increasing the synthesis and activity of Na$^+$-K$^+$-ATPase.
  ii. **Metabolism** – thyroid hormones increase glucose absorption from gastrointestinal system – increase both protein synthesis and degradation but the net effect is catabolic (degradation) – results in decreased muscle mass.
  iii. **Cardiovascular and respiratory systems** – thyroid hormones increase both cardiac output (increased heart rate and stroke volume) by inducing the synthesis of $\beta_1$-adrenergic receptors which increase heart rate and contractility. This results in an increase in ventilation as a result of increased O$_2$ consumption.
  iv. **Growth** – thyroid hormones act synergistically with growth hormone and somatomedins to promote bone formation.
v. **Central nervous system** – in the perinatal period, thyroid hormone is essential for normal maturation of the CNS – in adults, hypothyroidism causes a number of abnormalities including listlessness, slow movement, impaired memory and decreased mental capacity.

vi. **Autonomic nervous system** – thyroid hormones interact with the sympathetic nervous system – some of the effects of the thyroid hormones are similar to those of catecholamines acting through β adrenergic receptors. β adrenergic blocking agents (e.g. propanolol) can be used to treat symptoms related to hyperthyroidism.

h. TSH secretion does not appear to be affected in any consistent way by emotional of physical stress.

i. Thyroid hormone abnormalities are the most common endocrine pathologies.

   i. **Hyperthyroidism** – Grave’s disease is the most common hyperthyroid abnormality. It is an autoimmune disorder characterized by increased circulating levels of thyroid-stimulating immunoglobulins. Diagnosis of hyperthyroidism is based upon the clinical symptoms of weight loss accompanied by increased food intake, excessive heat production and sweating, rapid heart rate, and tremor, nervousness, and weakness coupled with the documentation of increased levels of thyroid hormones. Treatment is by radioactive ablation of the thyroid gland using I\(^{131}\), administration of propothiouracil or methimizole (both drugs which inhibit the synthesis of thyroid hormones) or surgical removal of the thyroid.

   ii. **Hypothyroidism** – The most common form is Hashimoto’s thyroiditis – autoimmune destruction of the thyroid gland. Surgical resection of the thyroid gland – sometime called surgical hypothyroidism – is also seen, although not as often as a couple of generations ago when most goiters (enlarged thyroid glands regardless of functional status) were removed surgically. The diagnosis is based on clinical symptoms of increased weight despite decreased food intake (decreased metabolic rate), decreased heat production with cold intolerance, and decreased heart rate. Treatment usually consists of administration of thyroid hormone – most commonly T\(_4\).

VI. **GROWTH HORMONE**

a. Growth hormone is essential for the normal rate of body growth during childhood and adolescence. It is secreted throughout life and remains physiologically important even after growth has stopped. It affects many aspects of carbohydrate, lipid and protein metabolism.

b. GH is produced in the somatotrophs of the anterior pituitary. GHRH release from the hypothalamus promotes expression of the GH, which ultimately results in the production of GH. Expression of the GH gene is
also stimulated by thyroid hormones and therefore individuals who are thyroid hormone deficient are also GH deficient.
c. Secretion of GH is regulated by two opposing hypothalamic-releasing hormones, growth hormone-releasing hormone (GHRH) and somatostatin (SRIF). The rate of GH secretion is determined by the net effect or balance of these two factors.
d. GHRH is synthesized in the cell bodies of neurosecretory neurons in the arcuate and ventromedial nuclei of the hypothalamus. Somatostatin production occurs mainly in the anterior periventricular region.
e. GHRH binds to membrane receptors, which are coupled to adenylate cyclase (cAMP) by stimulating G proteins (G_s). Adenylate cyclase is stimulated and cAMP rises in the cell. cAMP activates protein kinase A (PKA) that then phosphorylates proteins (P-proteins) involved in stimulating GH secretion and the expression of the gene for GH.
f. Somatostatin binds to membrane receptors, which are coupled to adenylate cyclase by inhibitory G proteins (G_i). This inhibits the ability of GHRH to stimulate adenylate cyclase and therefore blocks its action on GH secretion.
g. GH also stimulates the production of insulin-like growth factor (IGF-1 or somatomedin-C). This is a mitogenic agent that mediates the growth-promoting action of GH.
h. The interactive relationship involving GHRH, somatostatin, GH, and IGF-1 comprises the hypothalamic-pituitary-axis.
i. The feedback system responsible for regulating the concentration of GH in the blood is shown below

j. GH is secreted in periodic bursts, which produce large but short-lived peaks in GH concentration in the blood. These bursts ultimately result from neural activity generated in higher levels of the central nervous system, which affects the secretory activity of GHRH and somatostatin in the hypothalamus.
k. The main physiologic action of GH is on body growth. A growing child has bursts of GH secretion during both sleep and wake periods. As aging
occurs, the episodes of GH become less frequent during the awake period. In adulthood, GH bursts only occur in response to deep sleep.

1. Several abnormal developmental states occur as a result of GH deficiencies or excessive secretion. Example: GH deficiency causes a decrease in the rate of body growth and can result in pituitary dwarfism. The actual deficiency may be the result of defects in the mechanisms that control GH secretion or in the ability of the somatotrophs to produce GH. Example: Excessive GH in childhood results in gigantism. Excessive GH in adulthood does not cause gigantism because the growth plates of the long bones have already closed, but the bones of the face, ands, feet can become thicker and the liver may hypertrophy. This is known as acromegaly.

m. GH also affects certain aspects of fat and carbohydrate metabolism. It also functions as a counter-regulatory hormone that limits the action of insulin.

n. The following summarizes the target organs and activities of the anterior pituitary hormones:

<table>
<thead>
<tr>
<th>Pituitary Hormone</th>
<th>Major target organ(s)</th>
<th>Major Physiologic Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>GROWTH HORMONE</td>
<td>Liver, adipose tissue</td>
<td>Promotes growth (indirectly), control of protein, lipid and carbohydrate metabolism</td>
</tr>
<tr>
<td>THYROID STIMULATING HORMONE</td>
<td>Thyroid gland</td>
<td>Stimulates secretion of thyroid hormones</td>
</tr>
<tr>
<td>ACTH</td>
<td>Adrenal gland (cortex)</td>
<td>Stimulates secretion of glucocorticoids</td>
</tr>
<tr>
<td>PROLACTIN</td>
<td>Mammary gland</td>
<td>Milk production</td>
</tr>
<tr>
<td>Luteinizing Hormone</td>
<td>Ovary and testis</td>
<td>Control of reproductive function</td>
</tr>
<tr>
<td>Follicle Stimulating Hormone</td>
<td>Ovary and testis</td>
<td>Control of reproductive function</td>
</tr>
</tbody>
</table>
I. Introduction
   a. The thyroid hormones thyroxine (T₄) and triiodothyronine (T₃) play an important role in the regulation of body development and in governing the rate at which metabolic processes take place in individual cells. The regulatory functions of these two hormones are carried out by altering gene expression.
   b. T₃ and T₄ are produced in the follicular cells of the thyroid gland from a thyroglobulin precursor. Thyroglobulin itself is also produced in the follicular cells and stored in the lumen of these cells until needed. This represents a large reservoir available for later processing.
   c. Thyroid hormones are essential for normal brain development. They also regulate basal metabolic rate (BMR), energy metabolism, and protein synthesis.
   d. Thyroid hormones have major actions on all adult tissues except adult brain, testes and spleen.
   e. Thyroid hormone production and release is regulated by the thyrotropin-stimulating hormone (TSH) and, in turn, TSH is regulated by thyrotropin-releasing hormone (TRH).

II. Regulation of follicular cells by TSH
   a. TSH interacts with a receptor on the basal membrane of the follicular cell. Through a series of steps, phosphorylation of key proteins accounts for the actions of TSH on these cells.
   b. Some actions of TSH on follicular cells occur rapidly and do not require protein synthesis. These include shifting the ratio of T₃ formation relative to T₄ formation, increasing the release of thyroid hormones into the circulation, and increased uptake of thyroglobulin.
   c. Other actions of TSH require protein synthesis. These include an increase in TSH receptors and an increase capacity for iodide uptake.
   d. The end result of TSH stimulation of the follicular cells is hypertrophy and hyperplasia.

III. Thyroid follicular cells
   a. An iodide transporter that requires oxidative phosphorylation and ATP is located on the basal membrane facing the circulation capillary side of the follicular cells. The iodine concentration within the follicular cells is much higher than the concentration of this ion in the blood. This provides a pool of iodine that can be used to maintain thyroid hormone production and secretion for approximately 2 months in the absence of any dietary iodine.
   b. When the thyroid gland is stimulated to secrete thyroid hormones, thyroglobulin is recovered from the lumen by pinocytosis and T₃ and T₄ are formed.
IV. Secretion of thyroid hormones
   a. When secreted, T<sub>3</sub> and T<sub>4</sub> become bound to several different plasma proteins. Only a very small amount of the T<sub>3</sub> and T<sub>4</sub> is in the free form, which is the form that interacts with the target cells. A dynamic equilibrium exists between the bound and free forms, which provides a buffer against wide swings in the concentration of T<sub>3</sub> and T<sub>4</sub> in the blood.
   b. In the bound form, these hormones are protected from metabolic inactivation and ultimate secretion in the urine and therefore have relatively long half-lives. The half-life of T<sub>4</sub> is about 7 days and that of T<sub>3</sub> about 1 day.

V. Control of secretion - The following diagram indicates the general scheme for the control if the secretion of thyroid hormones:

VI. Mechanism of thyroid hormone action - T<sub>3</sub> exerts its effects via a specific, high affinity nuclear receptor whose affinity is 10-fold greater than its affinity for T<sub>4</sub>. Concentrations of T<sub>4</sub> are higher but T<sub>3</sub> is more potent and more important biologically. The effects on mRNA production then alter mitochondrial function, which ultimately changes the expression of certain genes in the target cell. T<sub>3</sub> therefore influences growth and metabolism by changing the amounts of structural and enzymatic proteins in the cells.

VII. Thyroid hormone action during fetal life and post-natal period
   a. T<sub>3</sub> is essential for the development of the central nervous system in the fetus. Absence of this hormone is related to a reduction in cortical neurons, mental retardation, and reduced vascularization of the CNS.
b. Thyroid hormones promote the expression of the gene for growth hormone (GH) in the anterior pituitary. Thyroid deficiency during the post-natal period severely retards the growth of almost all organs.

VIII. Metabolic actions of thyroid hormone
a. Thyroid hormones increase oxygen consumption and heat production. This heat production is associated with cellular respiration and concomitant oxidation of dietary substrates.
b. Approximately 15-40% of the basal energy used by the cell is to maintain the Na\(^+\)/K\(^+\) electrochemical gradient. Thyroid hormone increases the concentration and activity of the Na\(^+\)-K\(^+\) ATPase and thus affects the Na\(^+\) membrane pump. The observed increase in oxygen consumption is related to an increase utilization of ATP by the sodium pump and an increase in Na\(^+\)-K\(^+\) ATPase.
c. Additional actions of T\(_3\) are to increase heart rate, cardiac output, and ventilation, and to decrease peripheral vascular resistance (indirect effect due to the consumption of O\(_2\).)
d. Thyroid hormones stimulate carbohydrate, fat and protein metabolism.
   i. These actions include stimulation of intestinal absorption of glucose, and glycogenolysis.
   ii. Hepatic gluconeogenesis is also increased under the influence of thyroid hormones.
   iii. Lipogenesis, lipolysis and oxidation of free fatty acids are stimulated by thyroid hormones. Lipolysis increases the availability of glycerol, a starting material required for gluconeogenesis. In situations of T\(_3\) excess, the balance is tipped towards lipolysis, which results in mobilization of fat stores and loss of those stores.
   iv. Thyroid hormones also increase protein synthesis and proteolysis. Proteolysis provides amino acids, which are needed for hepatic gluconeogenesis. In T\(_3\) excess, protein degradation exceeds synthesis so the net effect is a loss of muscle protein. So hyperthyroidism is a catabolic state.
e. In addition, thyroid hormones are permissive (needed for other hormones to work) – for the effects of cortisol, glucagons, and growth hormone.

IX. Pathologies related to thyroid hormone disorders
a. Hypothyroidism in adults is associated with a reduction in nerve conduction velocity, impaired mentation and gradual mental deterioration. Basal metabolic rate is reduced and body heat production is reduced. Skin vasoconstriction occurs to conserve body heat. Intermediary metabolism slows down.
b. Hyperthyroidism due to excessive production of thyroid hormones results in a generalized increase in metabolic activity and production of excessive body heat, which induces excessive sweating in an attempt to reduce body heat. Physical weakness and fatigue may be present along with enhanced nervousness and irritability.
Chapter 4 - The Adrenal Gland  
Mary Zoe Baker, M.D.

Reading: Costanzo, p. 408-420.

I. Introduction
   a. The adrenal glands are small endocrine glands located upon the upper pole of each kidney. The adrenal glands can be divided histologically and biochemically into two major subglands:
      i. The adrenal medulla is a modified ganglion synthesizing epinephrine and to a lesser extent, norepinephrine. The medulla is supplied by the splanchnic nerve.
      ii. The adrenal cortex comprises three layers that surround the medulla. It is a steroid-synthesizing region that produces:
          1. cortisol – produced in the zona fasciculata
          2. aldosterone – produced in the zona glomerulosa
          3. androgens, estrogens and progestins – zona reticularis
   b. Cortisol plays an essential role in adjusting metabolism of carbohydrates, lipids, and proteins in the liver, muscle, and adipose tissue during fasting, assuring an adequate supply of glucose and fatty acids for energy metabolism despite the lack of food intake.
   c. Humans cannot survive total adrenalectomy without replacement of two essential steroids produced by the gland: cortisol and aldosterone.
   d. Adrenal hormones
      i. Cortisol is synthesized from cholesterol, diffuses out of the adrenal gland and enters the blood where it is transported bound to corticosteroid-binding globulin, which is made in the liver,
ii. Aldosterone is synthesized from cholesterol with the last enzyme in the synthetic pathway – aldosterone synthase - present in the glomerulosa only. It has no specific binding protein and about 37% of aldosterone circulates free in the plasma. The renin-angiotensin system is the primary regulator of aldosterone secretion. ACTH does acutely raise aldosterone levels but is not important in long-term overall regulation of aldosterone synthesis/secretion. Aldosterone is important in regulating salt balance and extracellular volume.

iii. Sex steroids secreted by the adrenal gland are primarily androstenedione and dehydroepiandrosterone (DHEA – 60% of circulating DHEA has an adrenal source) and dehydroepiandrosterone sulphate (DHEA-S – 90% of circulating DHEA-S has an adrenal source). These compounds are androgens – albeit much weaker than testosterone. They can, however, be converted to testosterone peripherally.

II. The hypothalamic-pituitary-adrenal axis
a. The parvocellular neurons of the paraventricular nucleus (PVN - release of CRH and arginine vasopressin), the anterior pituitary (release of ACTH) and the adrenal fasciculata (release of cortisol) constitute the hypothalamic-pituitary-adrenocortical axis (HPA) axis.

b. The HPA functions at a basal level that is reflected by a circadian rhythm in circulating cortisol levels. The peak of cortisol is observed at about 8 AM and the nadir at about 8 PM.

c. The physiological regulator of cortisol biosynthesis is adrenocorticotropic hormone (ACTH). ACTH release and biosynthesis is regulated by corticotropin-releasing hormone (CRH).
d. The HPA is a major component of the so-called “fight or flight” response. It is responsible for maintenance of homeostasis in crisis situations or in response to physiologic perturbations. Activation of the HPA axis with subsequent elevations in cortisol aid in homeostatic maintenance following exposure to a “stressor”. Prolonged exposure to stressors can lead to an emergence of “stress related disease”.

e. The HPA axis works with the sympathetic nervous system in maintaining homeostasis following stress. Sympathetic (catecholaminergic) mechanisms provide short term, rapid changes in response to perturbations in homeostasis, while the HPA (cortisol) provides a longer-term mechanism to control homeostasis allowing continued resistance to homeostatic perturbations.

III. Feedback regulation of cortisol production
a. Under normal HPA regulation, cortisol provides negative feedback at the hypothalamic PVN, anterior pituitary corticotrops and hippocampus to inhibit CRH/AVP and ACTH release following exposure to a stressor.
b. Dampened or less sensitive negative feedback is also observed during aging with hippocampal aging, damage to the hippocampus (i.e. stroke) and in AIDS patient.
c. ADH or vasopressin (AVP) is also secreted by the PVN. ADH is a potent ACTH secretagogue and may play an important physiologic role in the regulation of ACTH secretion in stress states.

IV. Physiologic role of cortisol
a. Cortisol mediates its action by binding to and activating the glucocorticoid receptor or GR, which is primarily located in the cytosol in the unbound form. Once cortisol binds the GR, the cortisol-GR complex translocates to the nucleus. Once in the nucleus, the cortisol-GR associates with the glucocorticoid response elements (GREs) where gene expression can be either enhanced or diminished. The GRs are structurally similar to other receptors in the steroid family – mineralocorticoids, sex steroids, vitamin D and thyroid hormones. The activation of GR alters cellular functions by altering the expression of genes.
b. Cortisol also can bind to and activate the mineralocorticoid receptor (MR). Cortisol’s affinity for the MR is equal to that of aldosterone. MR and GR are both expressed in regions of the CNS. The MR is a functional receptor for cortisol in the CNS.
c. Cortisol is permissive in that the expression of a large number of genes is dependent upon the presence of cortisol. Cortisol itself may not activate regulated enzymes.
d. An enzyme induced by cortisol may need activation via phosphorylation.
e. Cortisol enhances and suppresses the expression of a large number of genes in response to changes in circulating levels of cortisol.
f. Cortisol is essential for the maintenance of normal CNS function.

V. Cortisol effects on metabolism
a. Cortisol is considered a catabolic steroid that has the following functions;
i. Facilitates the conversion of protein to glycogen
ii. Enhances the mobilization of muscle protein for gluconeogenesis by accelerating proteolysis while inhibiting protein synthesis. Elevated levels of cortisol (or synthetic glucocorticoids) can deplete the protein stores of the body (muscle, bone, connective tissue and skin)
iii. Antagonizes the actions of insulin on glucose uptake in muscle (while stimulating glucose uptake in CNS.)
iv. Increases the amount of circulating branch chain amino acids and enhances the conversion of alanine to glucose. Alanine levels are not increased since alanine is metabolized rapidly to glucose.
v. Mobilization of fat from the subcutaneous adipose tissue. The secreted fatty acids afford an alternative fuel to glucose, thereby increasing the availability of glucose. For reasons that are not understood, some of this fat is deposited centrally.
vi. Induces the enzymes involved in metabolism of amino acids, facilitating gluconeogenesis

b. Cortisol is essential for survival during fasting states because of proteolytic effects. During fasting, liver glycogen stores become depleted. If cortisol is absent during fasting, gluconeogenesis from protein is diminished and death from hypoglycemia can occurs.
c. During fasting, plasma levels of cortisol are only slightly to moderately elevated. This elevation of cortisol is sufficient because of the permissive effects of cortisol on key metabolic enzymes.

VI. Cortisol effects on cardiovascular and renal function – sustained elevations of cortisol can produced the following effects:
a. Increased vascular adrenergic receptors
b. Increased vascular sensitivity to sympathetic activity
c. Increased vascular sensitivity to angiotensin II
d. Increased angiotensinogen production by the liver

VII. Cortisol effects on immune/inflammatory response
a. Hypercortisolism also increases the production of erythropoietin which leads to polycythemia while hypocortisolism leads to anemia
b. With respect to inflammation, cortisol decreases arachadonic acid production, which leads to reductions in prostaglandins, leukotrienes and thromboxanes, which are important modulators of local blood flow and the inflammatory response. Cortisol also reduces the recruitment of leukocytes to sites of trauma or infection and decreases the phagocytic and bactericidal activity of neutrophils.
c. With respect to the immune response, cortisol is immunosuppressive because it decreases the number of circulating T-cells and suppresses T-cell recruitment to the site of antigen.
d. The overall general effect of glucocorticoids on immunity is on cellular mediated immunity. As therapeutics, glucocorticoids are used as immune suppressants in cases of autoimmune disorders, some malignancies and during organ transplantation.
VIII. Cortisol effects on bone
a. Excessive cortisol inhibits bone formation by
   i. Decreasing type I collagen synthesis
   ii. Suppressing differentiation of progenitor cells to active osteoblasts
   iii. Promotes apoptosis of osteoblasts
   iv. Decreases renal absorption of calcium
   v. Decreases intestinal uptake of calcium
b. Cortisol deficiency is associated with hypercalcemia, indicating that physiologic levels of cortisol are needed for maintenance of calcium homeostasis
Chapter 5 - Calcium and Phosphate Regulation
Mary Zoe Baker, M.D.

on the web at http://www.surgeongeneral.gov/library/bonehealth/content.html
   (optional)
3) An excellent website maintained by Dr. Susan Ott of the University of
   Washington http://courses.washington.edu/bonephys/ophome.html (optional)

I. Introduction
a. Calcium is a key element in the following important physiologic processes
   i. Nerve excitability
   ii. Neurotransmitter release from axon terminals
   iii. Excitation-contraction coupling in muscle cells
   iv. Serves as a second messenger in several intracellular signal
       transduction pathways
   v. Serves as a cofactor in some enzymatic reactions including some in
      the blood clotting cascade
   vi. Is the major constituent of bone
b. Phosphorus - most often in the form of phosphate – is also important in the
   following physiologic process
   i. Important component of intracellular pH buffering
   ii. Is a component of various metabolic intermediates – DNA, RNA,
      and phosphoproteins
   iii. Is a major component of bone

II. Bone Physiology
a. Bone Cells
   i. Osteoclasts
      1. Derived from the same precursor cells in the bone marrow
         that produce white blood cells (stem cells)
      2. Formed by fusion of precursor cells into large multinuclear
         cells, which fasten tightly to the bone and develop an area
         of intense activity in the area under the cell at the bone
         surface with a highly irregular border called a ruffled
         border. This ruffled border contains transport molecules
         that secrete hydrogen ions to the bone surface where the
         mineral is dissolved. Enzymes are secreted from the
         ruffled border that help break down the matrix.
      3. Excessive bone breakdown by osteoclasts is an important
         cause of bone fragility not only in osteoporosis, but also in
         other bone diseases such as hyperparathyroidism.
   ii. Osteoblasts
      1. Derived from precursor cells that can also be stimulated to
         become muscle, fat or cartilage
      2. Under the right conditions these cells differentiate to form
         new bone which produces the collagen that forms the bone
         matrix. Calcium- and phosphate-rich mineral is added to
the matrix to form the hard, yet resilient, tissue that is healthy bone. Osteoblasts lay down bone in orderly layers that add strength to the matrix. They also initiate the steps in bone remodeling. The first steps involved are as follows.

a. Macrophage colony stimulating factor (m-CSF) is secreted by the mature osteoblast. m-CSF occupies its receptor on the surface of the hematopoietic stem cell to increase numbers of progenitor cells which become available to form osteoclasts.

b. Osteoblasts also secrete the receptor activator of nuclear factor kappa B ligand (RANK ligand or RANKL) which occupies its receptor (RANK) on the osteoclast cell surface and stimulates full differentiation of osteoclasts. The RANK/RANKL interaction also increases osteoclast activity. Finally, osteoprotegerin (OPG) an exogenous (to the osteoclast) protein binds the RANKL and prevents it from interacting with its receptor. This blocks the formation and activation of osteoclasts.

c. PTH, 1, 25 dihydroxyvitamin D, IL-1 and other factors act on the osteoblast to increase RANKL and decrease OPG shifting towards osteoclast differentiation and thereby increased bone resorption. The balance between RANKL and OPG determines the rate of bone resorption. (See below)

iii. Osteocytes

1. Are osteoblasts buried in the matrix as it is being produced.
2. Are the most numerous cells in bone, extensively connected to each other and to the surface of osteoblasts by a network of small thin extensions. This network is critical
for the ability of bone to respond to mechanical forces and injury.

3. When the skeleton is subjected to impact there is fluid movement around the osteocytes and the extensions that transmit signals to the bone cells on the surface to alter their activity, either in terms of changes in bone resorption or formation.

4. Failure of the osteoblasts to make a normal matrix occurs in a congenital disorder of the collagen molecule called osteogenesis imperfecta. At least 9 different phenotypes of osteogenesis imperfecta have been described.

b. Bone Remodeling – Involves the sequences of activation, resorption, reversal, and formation
   i. The activation step requires osteoblasts (either on bone surface or in marrow) acting on hematopoietic stem cells to differentiate into osteoclasts.
   ii. This begins the resorption step during which the multinuclear osteoclast attaches tightly to the bone surface and dissolves the calcified bone and matrix beneath it. This phase last a few weeks.
   iii. A brief reversal phase follows.
   iv. During the subsequent formation phase, new bone is laid down. During this process, osteoblasts are encased in bone, becoming osteocytes which are connected to each other and important in cell-to-cell communication. This phase may last several months. See figures below.
c. Types of Bone
   i. Cortical or compact bone
      1. 80% of total bone mass
      2. Long bones
      3. Very dense and provides much of the strength for weight bearing
      4. Low turnover - i.e. relatively metabolically inactive
   ii. Trabecular or Cancellous bone
      1. 20% of total bone mass
      2. Vertebral bodies, ends of the long bones
      3. Composed of a lattice of connecting bone trabeculae which can withstand compressive forces
      4. Very rapid turnover – i.e. relatively metabolically active

III. Body content and tissue distribution of calcium and phosphorous in a normal adult

<table>
<thead>
<tr>
<th>Total body content</th>
<th>Calcium</th>
<th>Phosphorous</th>
</tr>
</thead>
<tbody>
<tr>
<td>1300 g</td>
<td>600 gm</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Relative tissue distribution</th>
<th>Calcium</th>
<th>Phosphorous</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bones and teeth</td>
<td>99%</td>
<td>86%</td>
</tr>
<tr>
<td>Extracellular fluid</td>
<td>0.1%</td>
<td>0.08%</td>
</tr>
<tr>
<td>Intracellular fluid</td>
<td>1.0%</td>
<td>14%</td>
</tr>
</tbody>
</table>

IV. Inorganic constituents of bone

<table>
<thead>
<tr>
<th>Constituent</th>
<th>% of Total Body Content of Bone</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium</td>
<td>99</td>
</tr>
<tr>
<td>Phosphate</td>
<td>86</td>
</tr>
<tr>
<td>Carbonate</td>
<td>80</td>
</tr>
<tr>
<td>Magnesium</td>
<td>50</td>
</tr>
<tr>
<td>Sodium</td>
<td>35</td>
</tr>
<tr>
<td>Water</td>
<td>9</td>
</tr>
</tbody>
</table>
V. Blood forms
   Calcium:  
   - Ionized: 50%
   - Protein-bound: 40%
   - Inorganic salt: 10%

   Phosphorus:  
   At pH of 7.4  
   ~ 80% as HPO$_4^{2-}$
   ~ 20% as H$_2$PO$_4^-$

VI. Daily calcium balance in an adult.

VII. Parathyroid hormone
   a. Parathyroid hormone (PTH) is the major peptide regulator of calcium levels
   b. Some characteristics of this hormones are:
      i. Serum half-life is 6 minutes
      ii. Produced and secreted by the chief cells of the parathyroid gland
      iii. Biosynthesis and release is regulated directly at the level of the chief cells in response to circulating calcium levels.
      iv. Regulated indirectly by changes in circulating phosphate levels
      v. Is dependent on the presence of magnesium – hypomagnesia impairs PTH release.
      vi. Its expression is decreased by calcitriol in a negative feedback system.
      vii. Very small intracellular pool
   c. Regulation of PTH secretion
      i. The sensor on the parathyroid gland cell that detects changes in the ECF ionized calcium concentration is a calcium sensing receptor, a membrane protein that binds calcium extracellularly. It is coupled
to signaling pathways via G-proteins. These receptors are also found in the kidney.

ii. When ECF ionized calcium is high, calcium binds to this calcium sensing receptor and inhibits PTH secretion.

iii. When ECF ionized calcium is low, the receptor is not occupied by calcium and the inhibition of PTH secretion is removed.

d. Effects of PTH on calcium metabolism

i. Stimulates release of calcium from bone by stimulating bone resorption by osteoclasts.

ii. Stimulates calcium reabsorption in the kidney tubules.

iii. Indirectly promotes calcium absorption by the digestive tract, because it activates 1α hydroxylase in the kidney that produces 1,25-(OH)₂D₃ (the active form of vitamin D) of 25-hydroxy vitamin D.

VIII. Calcitonin

a. Calcitonin is produced by the parafollicular cells of the thyroid gland

b. Secretion of calcitonin is stimulated by an increase in plasma calcium and its net effect is to promote calcium deposition in bone by inhibition of osteoclast activity and formation. Stimulation of calcitonin secretion by gastrointestinal hormones provides an additional mechanism facilitating calcium uptake after ingestion of a meal.

c. Calcitonin has a weak effect on plasma calcium concentrations in humans. The effects of PTH are much more potent and override the effects of calcitonin. Calcitonin is important clinically in that it can be used clinically to treat hypercalcemia (although other, more potent agents are
used first) and osteoporosis (again, there are more potent treatments). Another clinical use is as a marker of medullary thyroid cancer, a malignancy of the C-cells.

IX. Vitamin D$_{3}$

a. Vitamin D$_{3}$ can be provided in the diet or directly in the skin as a result of ultraviolet light activation of synthesis from a precursor derived from cholesterol.

![Biosynthesis of vitamin D](image)

b. The active form of the hormone is 1,25 dihydroxycholecalciferol (1,25-(OH)$_{2}$D$_{3}$)

c. The major effects of 1,25 dihydroxycholecalciferol on calcium and phosphate metabolism are illustrated below. (1,25-(OH)$_{2}$D$_{3}$)
X. Overview of Calcium and Phosphate Metabolism

Note that calcitonin is not a significant regulator of calcium metabolism.

XI. Hypocalcemia and the integrated regulation of calcium and phosphate.
   a. With Hypocalcemia
      i. PTH release is stimulated
      ii. In the kidney
         1. Increased tubular reabsorption and decreased excretion of calcium and increased excretion of phosphate (1-15 minutes after PTH secretion)
         2. Stimulation of calcitriol synthesis and secretion
      iii. In bone
         1. Calcium and phosphate released from bone by activation of osteoclast activity and number
         2. Calcitriol acts on bone to synergize PTH (within hours) which prevents a recurrence of hypocalcemia in re-establishing calcium homeostasis
      iv. In the intestine, calcitriol increases calcium absorption.
   b. The end result of these actions is
      i. Increase in blood calcium levels resulting in a
      ii. Decreased stimulation of PTH secretion
      iii. Calcitriol negatively feeds back on the parathyroid glands to further suppress PTH secretion

XII. Hypercalcemia and the integrated regulation of calcium and phosphate.
    a. With Hypercalcemia
       i. PTH release is inhibited
       ii. In the kidney
1. Decreased tubular reabsorption and increased excretion of calcium and decreased excretion of phosphate (1-15 minutes after PTH secretion)

2. Inhibition of calcitriol synthesis and secretion

iii. In bone
   1. Osteoclast activity and number is decreased
   2. Calcitriol is decreased and mobilization of calcium from bone is decreased

iv. In the intestine, calcitriol decreases calcium absorption.

b. The end result of these actions is
   i. Decrease in blood calcium levels resulting in
   ii. Normalized PTH secretion

XIII. Phosphate Homeostasis

a. The kidney plays the dominant role in phosphorous homeostasis.

b. Phosphorous homeostasis is less tightly regulated than calcium.

c. PTH stimulates increase renal clearance of phosphate, increases release of phosphate from the bone, and increases absorption of phosphorous in the intestine.

d. Vitamin D stimulates phosphate reabsorption in the kidney and increases absorption in the intestine.

e. Hypophosphatemia stimulates $1,25(OH)_2D_3$ synthesis and suppresses PTH release.

\[
\text{Phosphorous from KIDNEY} \rightarrow \text{Low Serum Phosphorous} \rightarrow \text{1, 25 (OH)$_2$ D} \rightarrow \text{Ca & Phosphorous from BONE} \rightarrow \text{PTH} \rightarrow \text{Calcium from KIDNEY} \rightarrow \text{Increased serum Phosphorous} \rightarrow \text{No change in serum Calcium}
\]

Sequence of Events in Response to Hypophosphatemia

f. Hyperphosphatemia inhibits the synthesis of $1,25(OH)_2D_3$ and directly stimulates PTH release. PTH is also stimulated by the decrease in vitamin D which causes a decrease in ionized calcium. The increased PTH causes an increase in calcium absorption across the GI tract which then complexes with the excess phosphorous, further decreasing available calcium. This is a very common management problem in renal failure resulting in secondary hyperparathyroidism. In addition to the phosphate
problems, the failing kidney can not synthesize 1,25(OH)₂D₃ and low calcium levels further complicate the problem.

XIV. Hyperparathyroidism
   i. In primary hyperparathyroidism the primary abnormality is excess PTH secretion which typically is the result of a parathyroid adenoma or hyperplasia – most common
   ii. The symptoms below may be seen. Most patients in the US have asymptomatic hyperparathyroidism
      1. mental confusion
      2. depression
      3. headaches
      4. polyuria
      5. polydipsia
      6. corneal calcifications
      7. renal stones
      8. pancreatitis
      9. Bone loss
   iii. Lab and radiology findings include
      1. Hypercalcemia, mild hypophosphatemia, hypercalciuria
      2. Increased urinary cAMP and hydroxyproline
      3. Elevated serum alkaline phosphates
      4. Bone resorption on xray
   iv. Treatment varies depending upon the severity of the disease. Patients with severe disease and/or significant symptoms undergo surgical resection of the overactive parathyroid tissue. Less severe disease is managed medically by promoting diuresis to facilitate urinary calcium loss.

XV. Hypoparathyroidism - This condition is seen most often in the patient who has undergone neck surgery
   a. Symptoms of hypocalcemia can be
      i. Convulsions
      ii. Obtundation
      iii. Tetany
      iv. Laryngospasm
      v. Cataracts
   b. Laboratory findings are
      i. Hypocalcemia
      ii. Low PTH levels
      iii. Low vitamin D levels
   c. Etiologies may include
      i. Radical neck surgery or as a complication of thyroid surgery
      ii. Hereditary diseases
      iii. Destruction by malignancies
   d. Treatment
      i. Calcitriol (not 25(OH) vitamin D – Why?)
      ii. Calcium
Chapter 6 - The Endocrine Pancreas  
Mary Zoe Baker, M.D.

Reading: Costanzo, p. 420-428.

I. Introduction
   a. The endocrine component of the pancreas resides in the islets of Langerhans. This account for about 1% of the total weight of the organ and it is estimated that there are approximately 250,000 islets in an adult pancreas. The exocrine pancreas is considered a part of the GI tract.
   b. The islets consist of four major cell types:
      i. \(\alpha\) cells (A cells) associated with glucagons
      ii. \(\beta\) cells (B cells) associated with insulin
      iii. \(\delta\) cells (D cells) associated with somatostatin (aka: GHRIH)
      iv. F cells associate with pancreatic polypeptide
   c. The hormones are released into the hepatic portal vein.

II. Role of the endocrine pancreas – The role of the hormones of the endocrine pancreas is to regulate energy metabolism. This is done by coordinating by the flow of endogenous glucose, free fatty acids, amino acids, and ketone bodies. These hormones also coordinate the storage of dietary nutrients. By coordinating the flow of energy stores the endocrine pancreas ensures that energy needs are met during basal and active states. The hormones involved in energy metabolism primarily act upon liver, muscle and adipose tissue.

III. Insulin
   a. Insulin is produced in the pancreatic islets in the \(\beta\) cells. A single polypeptide chain (prohormone) is acted upon by a specific endopeptidase to generate insulin. It is a two-chain polypeptide joined by disulfide bonds.
   b. The metabolic half-life of insulin is approximately 5-8 minutes. About half is degraded by hepatic insulinase. Insulin concentrations in the liver are higher than in the peripheral circulation.
   c. C-peptide is more stable than insulin (plasma half-life of hours) and measurement of C-peptide provides a reliable estimate of endogenous insulin production.
   d. Regulation of insulin synthesis and secretion
      i. The major stimulators of insulin synthesis and release are:
         1. Increases in plasma glucose
         2. Increases in plasma energy substrates (proteins, ketoacids, fatty acids, triglycerides)
         3. Increases in gastrointestinal peptides
      ii. Activity of the parasympathetic nervous systems (acetylcholine) also increases insulin levels.
      iii. The major inhibitors of insulin synthesis and release are:
         1. Decreases in plasma glucose
         2. Decreases in plasma energy substrates
         3. Somatostatin
iv. Activity of sympathetic nervous system (norepinephrine) also decreases insulin levels.

e. Regulation of insulin release and synthesis
   i. The β cell itself is highly responsive to changes in plasma glucose levels.
   ii. Several steps are involved in the regulation of insulin release and synthesis. The general scheme:
      1. A noninsulin-sensitive Glut-2 transporter located in the plasma membrane transports glucose into the β cell via facilitated diffusion. This process maintains intracellular levels of glucose equal to extracellular levels.
      2. Energy substrates are rapidly metabolized. ATP is generated which drives a Na⁺/K⁺ ATPase and intracellular K⁺ increases.
      3. K⁺ channels close, K⁺ efflux is inhibited and the β cell depolarizes which opens voltage-gated Ca⁺² channels.
      4. Ca⁺² influx stimulates release of insulin from internal stores (rapid) and activates calmodulin, which increases synthesis (slower release).
   iii. The total amount of insulin secreted after an oral glucose load is greater than an intravenous glucose load. This observation led to the discovery of incretins which augment β-cell response. Cholecystokinin and gastric inhibitory protein (GIP) are secreted by the GI tract in response to feeding and enhance insulin secretion. A third incretin, glucagon-like-peptide-1 (GLP-1) will be discussed below.
   iii. Sulfonylurea drugs stimulate insulin release by directly ‘closing’ the K⁺ channel and are therefore useful in treating type II non-insulin dependent diabetes. Diazoxide opens the K⁺ channel and is useful in managing hyperinsulinemia and the resultant hypoglycemia that can occur.
iv. During an infusion of glucose, there is a biphasic change in the plasma levels of insulin. There is an initial surge that represents the release of pre-existing stores of insulin (peaks at about 5 minutes) followed by a slower release of insulin that represents the synthesis of new insulin (peaks at about 1 hour).
   v. The response to oral ingestion of glucose is also biphasic. An initial peak occurs within about 5 minutes and a secondary peak occurs in about 25 minutes.

f. Insulin actions at the tissue and cellular level
   i. Insulin mediates its action via occupying a specific plasma membrane.
   ii. The insulin receptor (Ins-R) is a heterotetrameric protein consisting of two extracellular α subunits and two transmembrane β subunits. The binding of the ligand to the α subunit of IR stimulates the tyrosine kinase activity intrinsic to the β subunit of the receptor.
iii. The ability of the receptor to autophosphorylate and phosphorylate intracellular substrates and thereby trigger complex signaling cascades is essential for insulin’s mediation of the cellular responses to insulin. There are three major mechanisms by which this occurs.

1. SH2 (Src homology domain-2)-containing proteins can bind the phosphorylated tyrosine residues of the insulin receptor itself.
2. The Ins-R can phosphorylate and activate various cytoplasmic proteins at tyrosine residues.
3. The Ins-R can phosphorylate a family of proteins know as insulin-receptor substrates (IRS-1, -2, -3, and – 4)
4. The number of insulin receptors on the cell surface exceeds that needed for a maximal biologic response. Tissues can modulate their response to insulin by a variety of mechanisms. Down-regulation refers to the ability of the cell to decrease the number of insulin receptors at the cell surface. (done by decreasing receptor synthesis and accelerating degradation of the receptor). This situation is seen in patients with type 2 diabetes who have increased insulin levels.

iv. The effects of increasing plasma insulin are

1. In muscle
   a. Increased glucose uptake
   b. Net glycogen synthesis
   c. Net amino acid uptake
   d. Net protein synthesis

2. In adipocytes
   a. Increased glucose uptake and utilization
   b. Net triacylglycerol synthesis

3. In liver
   a. Increased glucose uptake
   b. Net glycogen synthesis
   c. Net triacylglycerol synthesis
   d. No ketone synthesis

<table>
<thead>
<tr>
<th>Increased Plasma Insulin</th>
<th>Increased Plasma Insulin</th>
<th>Increased Plasma Insulin</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Muscle</strong></td>
<td><strong>Adipocytes</strong></td>
<td><strong>Liver</strong></td>
</tr>
<tr>
<td>• Increased glucose uptake and utilization</td>
<td>• Increased glucose uptake and utilization</td>
<td>• Glucose uptake</td>
</tr>
<tr>
<td>• Net glycogen synthesis</td>
<td>• Net triacylglycerol synthesis</td>
<td>• Net glycogen synthesis</td>
</tr>
<tr>
<td>• Net amino acid uptake</td>
<td></td>
<td>• Net triacylglycerol synthesis</td>
</tr>
<tr>
<td>• Net protein synthesis</td>
<td></td>
<td>• No ketone synthesis</td>
</tr>
</tbody>
</table>
The effects of decreasing plasma insulin levels are:

1. **In muscle**
   a. Decreased glucose uptake and utilization
   b. Net glycogen catabolism
   c. Net amino acid release
   d. Fatty acid uptake and utilization

2. **In adipocytes**
   a. Decreased glucose uptake and utilization
   b. Net triacylglycerol catabolism and release of glycerol and fatty acids

3. **In liver**
   a. Glucose release due to net glycogen catabolism and gluconeogenesis
   b. Ketone synthesis and release.

<table>
<thead>
<tr>
<th>Decreased Plasma Insulin</th>
<th>Decreased Plasma Insulin</th>
<th>Decreased Plasma Insulin</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Muscle</strong></td>
<td><strong>Adipocytes</strong></td>
<td><strong>Liver</strong></td>
</tr>
<tr>
<td>• Decreased glucose</td>
<td>• Decreased glucose</td>
<td>• Glucose release due to</td>
</tr>
<tr>
<td>uptake and utilization</td>
<td>uptake and utilization</td>
<td>net glycogen catabolism</td>
</tr>
<tr>
<td>• Net glycogen catabolism</td>
<td>• Net triacylglycerol</td>
<td>and gluconeogenesis</td>
</tr>
<tr>
<td>• Net amino acid release</td>
<td>catabolism and release</td>
<td>• Ketone synthesis and</td>
</tr>
<tr>
<td>• Net fatty acid uptake</td>
<td>of glycerol and fatty</td>
<td>release</td>
</tr>
<tr>
<td>and utilization</td>
<td>acids</td>
<td></td>
</tr>
</tbody>
</table>

**IV. Glucagon**

a. Synthesis of glucagon takes place in the pancreatic α cell. A prohormone is synthesized which undergoes limited proteolysis generating glucagon.

b. The plasma half-life of glucagon is approximately 5-6 minutes. Around 80% is degraded by hepatic peptidases and effective glucagon concentrations in the liver are higher than in the peripheral circulation.

c. Glucagon acts at the liver through activation of a $G_{\alpha}$-coupled receptor activating c-AMP.

d. The major stimulators of glucagon synthesis and release are:
   i. Amino acids released after ingestion of a protein meal are the major secretagogue at the α cell.
   ii. Physiologic stimuli are decreases in plasma glucose, other plasma energy substrates ie. ketoacids, fatty acids, triglycerides, decreases in insulin levels.
   iii. Parasympathetic and sympathetic nervous system activity
   iv. Stress and exercise

e. The major inhibitors of glucagon synthesis and release are:
   i. Increases in plasma glucose
   ii. Increases in plasma energy substrates
   iii. Increases in insulin levels
f. The incretin glucagon-like-peptide-1 (GLP-1) is secreted in the intestinal L-cell by alternative processing of glucagons. GLP-1 is a weak glucagon analog but is a potent stimulator of insulin secretion. Currently, analogs of GLP-1 are in development as treatment for type 2 diabetes.

g. During insulin-induced hypoglycemia, low glucose overrides insulin inhibition of glucagon.

V. Somatostatin
a. Secreted by the pancreatic δ cells of the pancreas as well as cells in the CNS (see pituitary chapter)
b. Suppresses growth hormone secretion
c. Also suppresses insulin, glucagons, gastrin, vasoactive intestinal peptide or VIP.
d. Biologic effects unclear
e. Major clinical utility has been the development of long-acting somatostatin analogs for the treatment of GH excess (acromegaly), insulin secretion in insulinomas and for use in the post-operative management of patients who have undergone pancreatic surgery to decrease the secretions of the pancreas.

VI. Target Tissues for Pancreatic Hormones
a. Insulin – liver, adipose tissue and muscle
b. Glucagon – Liver

VII. Insulin and carbohydrate metabolism
a. Insulin is the only major hormone that induces glucose uptake. It stimulates glucose transport in GLUT-4 (an insulin sensitive transporter) tissues and stimulates glucose utilization as an energy source. Both effects result in the decrease in plasma glucose following insulin release. A cartoon depicting the role of GLUT-4 in glucose transport is at the following website:
   http://arbl.cvmbs.colostate.edu/hbooks/pathphys/endocrine/pancreas/insulin_phys.html
b. Effects on the liver
   i. The liver stores glucose as glycogen, produces glucose via gluconeogenesis and it is the exclusive site of ketogenesis
   ii. Insulin increases hepatic glucose uptake, glucose oxidation, glycogen storage and the formation of cellular energy.
   iii. Insulin inhibits gluconeogenesis and glycogenolysis.
   iv. The indirect effects of insulin on liver carbohydrate metabolism include:
       1. Decreased free fatty acids delivery to liver (effects on adipose tissue)
       2. Decreased fatty acid β oxidation and generation of acetyl CoA
       3. The decrease in free fatty acids helps to reduce gluconeogenesis.
c. Effects on muscle
   i. The major sites of action are cardiac and skeletal muscle.
ii. Insulin stimulates glucose transport by increasing both the activity and number of GLUT-4 transporters.

iii. The following effect of insulin on muscle are important:
    1. Stimulates an increase in glucose oxidation (glycolysis)
    2. 20-50% of the glucose in muscle undergoes glycolysis/oxidation. This is a relatively slow effect that is observed several hours after insulin increases.
    3. 50-80% of the glucose is converted to glycogen via glycogen synthase. This is a rapid effect that is observed within minutes after insulin increases.
    4. Insulin stimulates the synthesis of protein in skeletal muscle.

d. Effects on adipose tissue
   i. Adipose tissue serves as the principal energy reserve in the body. Its total mass is about 10 kg (22 lbs.) in an average male. It is the principal source of free fatty acids. It releases some amino acids but not glucose.
   ii. Insulin stimulates glucose transport by increasing the number and activity of a GLUT-4 transporter. The rate of transport is the limiting step in glucose metabolism in this tissue.
   iii. Within the adipose tissue cell, glucose is stored as glycogen or metabolism to triglyceride.

VIII. Insulin and fat metabolism
   a. Effects on liver – insulin increases free fatty acids from glucose, decreases ketogenesis and favors cholesterol biosynthesis.
   b. Effects on muscle – Insulin suppresses lipoprotein lipase (action is inversely proportional to glucose uptake) and suppresses free fatty acid uptake (free fatty acids and glucose are competitive substrates).
   c. Effects on adipose tissue
      i. Glycerol liberated from lipolysis can serve as a substrate for gluconeogenesis.
      ii. Insulin plays a major role in energy storage in adipose tissue, essentially in the form of triglycerides, which represent 90% of the mass of adipose tissue. The overall net effect of insulin is an enhanced storage of fats (lipogenesis). Insulin inhibits mobilization and oxidation of fatty acids and rapidly decreases circulating triglycerides. In adipose tissue, insulin inhibits hormone sensitive triglyceride lipase activity.
      iii. There is thus a marked reduction in generation of ketoacids. The antilipolytic activity of insulin is the most important effects of insulin on adipocytes.

IX. Insulin and protein/amino acid metabolism
   a. Enhances amino uptake into liver and muscle
   b. Stimulates the incorporation of all amino acids into proteins in muscle
   c. Increases the number and translation al efficiency of ribosomes
d. Influences ion transport (Increases intracellular $K^+$; decreased intracellular $Na^+$)

X. Glucagon and carbohydrate metabolism
   a. Glucagon, epinephrine, GH, and cortisol are glucose liberating hormones and are counter-regulatory to insulin
   b. Effects on liver
      i. The liver is the major site of glucagon action on carbohydrate metabolism
      ii. Glucagon acts to increase blood glucose levels by stimulating both glycogenolysis and gluconeogenesis.

XI. Glucagon and fat metabolism
   a. Effects on liver
      i. Glucagon increases the $\beta$ oxidation of acetyl CoA and increases ketogenesis.
      ii. Glucagon increases free fatty acid formation, acetyl CoA carboxylase activity and decreases cholesterol synthesis
   b. Effects on adipose tissue
      i. The effects of glucagon on adipose tissue include increased activity and increased generation and release of free fatty acids (this is the source of free fatty acids and oxidation in the liver)
      ii. The central event in adipose tissue metabolism is the cycle of esterification of FA and triglyceride formation and FA/neutral fat lipolysis by a hormone sensitive lipase.
   c. Effects on protein/amino acid metabolism – glucagon enhances amino acid conversion to substrates for gluconeogenesis. Plasma amino acids are lowered as glucose increases.

XII. Central nervous effects of hypoglycemia
   a. The central nervous system requires glucose for energy and is sensitive to large decreases in glucose levels.
   b. The effects of hypoglycemia include restlessness, irritability, motor coordination, confusion, tingling sensations, headache, and sweating. Severe hypoglycemia results in loss of consciousness, coma and death.
   c. If there is a gradual decrease in glucose levels, the central nervous system can adapt to use ketone bodies but this adaptation requires several weeks to develop.

XIII. Clinical syndromes of islet cell dysfunction
   a. Insulin excess is the hallmark of an insulinoma which is a $\beta$ cell tumor of the pancreas. Associated signs and symptoms are
      i. Hypoglycemia as a result of hyperinsulinemia
      ii. Sympathetic activation by the hypoglycemia causing rapid heart rate, nervousness, sweating, hunger.
      iii. Weight gain (remember that insulin is an anabolic hormone)
      iv. Central nervous system dysfunction
   b. Type 1 diabetes which is the result of autoimmune $\beta$ cell destruction
      i. Symptoms
1. Polyuria due to the osmotic diuresis caused by the increased osmotic load of the glucose.
2. Thirst as a result of the polyuria
3. Volume depletion
4. Increased appetite
5. Weight loss
6. Frequent urination
7. Tissue damage as manifested by microvascular complications (retinopathy, neuropathy and nephropathy)
8. Increased risk for cardiovascular disease

ii. Metabolic derangements
1. Hyperosmolality of plasma
2. High plasma and urine glucose levels
3. Increased gluconeogenesis supported by uninhibited proteolysis and muscle catabolism
4. Increased urinary excretion of water and electrolytes
5. Negative nitrogen balance
6. Uninhibited lipolysis (decreased body fat)
7. Increased ketogenesis (decreased peripheral ketoacid utilization) leading to acidemia which can be profound and fatal – levels below pH of 6.8.
8. Increased carbonic acid as a result of neutralization of the “strong” ketoacids – carboxylic acids – by sodium bicarbonate.
9. Lowered pCO₂ as a result of acid-stimulation of hyperventilation as a compensatory mechanism.
10. Hypersecretion of glucagons

c. Type 2 diabetes is the result of systemic insulin resistance with a superimposed β cell secretory defect. It is often associated with obesity and characterized by:
   i. Loss of first phase insulin release by the β cell
   ii. Excessive hepatic glucose production by gluconeogenesis and glycogenolysis
   iii. Elevated plasma glucose, especially after carbohydrate ingestion.

d. Glucagon excess is the result of production and release of the α cell tumors. Associated signs and symptoms are
   i. Weight loss, distinctive skin rash, elevated plasma glucose and ketoacidosis
   ii. Reduced plasma amino acid
   iii. Increased urinary nitrogen