9

Peptide Hormones and the Endocrine System

Hormones are mammalian metabolites that are produced by endocrine or ductless glands, are released directly into the blood, and are involved in eliciting responses by specific body organs and tissues. These biologically active metabolites either are steroidal or are derived from amino acids. The latter group of hormones consists primarily of peptides of various sizes, but a few nonpeptide metabolites (epinephrine, thyroxine) are known. Pertinent details about the nonsteroidal hormones and general aspects of endocrine products used as therapeutic agents will be discussed in this chapter. Specific details about the steroid hormones were presented previously (see page 173).

HISTORICAL DEVELOPMENT

Present therapeutic use of endocrine products is an outgrowth of the primitive practice of organotherapy. The use by Magnus in the 13th century of powdered hog testis to treat male impotence and of rabbit uterus to treat female sterility was a direct progenitor of the present therapy. The basic philosophy for the use of mammalian organs was ably expressed by Vicary in the 16th century when he said, "In what part of the body the faculty you would strengthen lies, take the same part of the body of another creature in whom the faculty is strong, as a medicine."

The origin and early development of endocrine therapy were empiric, but most of the present knowledge of endocrine function and therapy is the result of intensive investigations conducted over the past 35 years. Standardized powdered glands and glandular extracts initially provided more reproducible effects and better therapeutic control than did randomly selected glands, and isolated hormonal substances have offered additional advantages in most cases. Modern technology has permitted the ready synthesis of many hormones including a number of peptides, and the preparation of substances that mimic the actions of natural hormones (e.g., prednisone-cortisone).

Much of the obvious progress in the endocrine area is reflected in the nature of available products. However, a more precise comprehension of their physiologic functions and improved diagnostic procedures have contributed significantly to therapeutic advancement.

GENERAL PHYSIOLOGIC INVOLVEMENT AND THERAPEUTIC PHILOSOPHIES

Hormones function as chemical transmitters of selective stimuli between the var-

ious endocrine glands and specific body organs and tissues. Sufficient information is available to permit some generalizations about the modes by which hormones influence the metabolism of target cells and maintain homeostasis. The size and lipophilic character of steroids permit penetration of cell membranes, but many peptide hormones will not enter cells in the absence of a specialized transport system. The latter hormones, in many cases, bind to receptors on the surface of the cell and act in one of two ways: (1) to directly induce changes in membrane permeability for ions, glucose, amino acids, etc., or (2) to induce the production of a secondary messenger such as cyclic AMP, which transmits the signal of the hormone within the cell. Hormones that control membrane permeability, either directly or indirectly, include the estrogens, growth hormone, glucagon, glucocorticoids, insulin, testosterone, and vasopressin. Induction of enzyme formation and modification in the rate of enzymatic reactions are other known mechanisms of hormonal action.

Physiologic control of hormone formation or release to regulate hormone level is a vital aspect of maintaining metabolic homeostasis and integrity of body function. Two general regulatory mechanisms are currently recognized. There is a feedback mechanism that responds to change in concentration of some substance in the blood. The key substance may be a hormone or some other metabolite. For example, an increase in blood glucose in normal persons stimulates the release of insulin, and increased levels of triiodothyronine-thyroxine cause a decrease in thyrotropin secretion owing to an inhibition of the secretion of thyrotropin-releasing factor by the hypothalamus (see page 173). The second mechanism involves external stimuli and is mediated by the hypothalamus; the hypothalamus secretes releasing factors that act on the anterior pituitary to increase the release of specific tropic hormones.

Some manifestations of hormonally controlled processes are rather subtle, and their significance in the normal individual is usually recognized only by people trained in the health sciences. However, the widely recognized influence of the gonadal hormones on the development and function of the reproductive organs and sex characteristics illustrates the general type of fundamental involvement of hormones.

There is appreciable interaction among the functions of the various endocrine glands and a close correlation between the endocrine system and the central and autonomic nervous systems. Thus, a primary disturbance in an endocrine gland or therapy with a hormone may have far-reaching effects. Caution must be exercised in therapeutic management of such complex situations to avoid dangerous or irrational developments.

Disturbance in the function of an endocrine gland may take the form of excessive activity (hyperfunction) or diminished activity (hypofunction), to any degree. The most frequently encountered therapeutic situations involve the hypofunctioning gland. Replacement therapy merely uses endocrine preparations to supplement or totally replace abnormally low levels of endogenous hormone. Early diagnosis and treatment are essential in this type of therapy to avoid irreversible changes that can occur, such as cretinism, giantism, and other comparable conditions. Use of hormones for replacement purposes is usually long-term therapy, and, because these potent substances are normal body metabolites, serious side effects are usually minimal if caution is taken to balance the administered dosage with replacement needs. Insulin utilization provides a good example of this type of approach to a hypofunctioning endocrine system.

Hypofunctioning glands that retain some degree of activity can potentially be stimulated to approach normal activity by the use of drugs that are not hormones per se or by the inhibition of the normal cata-

bolic processes to conserve the limited supply of hormone. There are many variations to this type of therapeutic approach, but they all require metabolic capability in the glandular tissue and a thorough knowledge of biochemical detail. This general approach will probably become more useful in the future when greater knowledge is available.

Hormonal substances are not employed in treatment of hyperfunctioning endocrine glands. Antimetabolites may offer some potential for eliminating the deleterious effects of abnormally high levels of hormones. Alternate approaches to this kind of problem include surgical removal of part of the hyperfunctioning gland or selective destruction of some of the glandular tissues. Various radiation treatments, including the use of ^{131}I in certain thyroid conditions, represent the latter approach.

Sometimes, hormones have therapeutic utility for pharmacologic actions that are not directly related to normal endocrine functions. The use of glucocorticoids for anti-inflammatory and antirheumatic purposes falls into this category (see page 179). The potential danger of serious side effects is considerably greater when hormones are used for specific pharmacologic actions than when they are used for replacement therapy. Prolonged therapeutic use of a hormone, such as cortisone, may cause irreversible atrophy of the endocrine gland that normally produces the hormone or may induce other undesirable secondary responses. The safest use of a hormone as a therapeutic agent involves a short duration of therapy, e.g., the use of oxytocin to control postpartum hemorrhage. This short-term approach avoids problems associated with prolonged upset of the delicate balance among various endocrine systems.

COMMERCIAL PRODUCTION

Many drugs employed in medical practice and generally classified as endocrine products are by-products of the meat-packing industry. Thyroid, pancreas, adrenal, and pituitary glands of bovine and porcine origin are used for such purposes. The active principles obtained from such glands may vary in quantity and quality, depending on species, so that specific manufacturing processes are developed and used with particular species in mind.

Glands used in the manufacture of pharmaceutic products are collected in government-inspected packinghouses and must conform to the regulations of the Meat Inspection Department of the United States Department of Agriculture. Only glands from carcasses that are classified by federal inspectors as edible may be used.

Endocrine glands are technically fresh meat and must be processed in a manner that prevents deterioration. Glands are normally quick-frozen as soon as they are removed from the animal carcasses and maintained in a frozen state until processed. Processing varies with the nature of the ultimate endocrine product. The glands, in many cases, are subjected to extraction and fractionation treatments to yield purified hormones. However, the continued therapeutic acceptance of desiccated thyroid is a pragmatic reminder that satisfactory results sometimes can be achieved without costly isolation of the active principles. Frozen thyroid gland is simply dehydrated, defatted, powdered, standardized, and made into suitable dosage forms.

Chemical synthesis is a logical approach to ensuring the ready availability of adequate supplies of any natural metabolite of known structure and therapeutic utility. The feasibility of synthetic procedures is governed to a large degree by the complexity of the molecule and by the technical knowledge relating to the given type of compound. Hormones present no exception to these generalizations. Perhaps the only unusual feature that is applied commercially in the production of certain hormones involves the action of selected bio-

logic systems on foreign substrates. Various microorganisms metabolize certain steroids to give useful compounds. An indication of the significant contribution of microbial biotransformation of steroids is included in the steroid chapter (see page 176).

The Merrifield solid-phase synthesis of peptides is a technologic development of the 1960s of major significance for endocrine therapy. This technique involves basically attaching a carboxy-terminal amino acid to a resin column and synthesizing a polypeptide by passing a programmed sequence of reacting solutions through the column. There is no need to isolate each intermediate; the process can be automated, and commercially feasible synthesis has already been extended to peptides containing 24 to 32 amino acid residues (cosyntropin and calcitonin, respectively). A number of peptide hormones that were previously isolated from glandular materials are now prepared synthetically, and the prospects for new developments in the area of peptides with hormonal activity are among the best for any group of therapeutic agents.

ADRENAL GLANDS

The **adrenals** (suprarenals) in humans are a pair of small glands; one is situated over the superior medial aspect of each kidney. Each average gland measures 5 × 25 × 50 mm; together, the adrenals weigh 4 to 18 g.

The adrenals were first described by Eustachius in the 16th century and were long supposed to function in the inhibition of fetal urination and in the prevention of renal stones in the adult. Knowledge of adrenal function began with Addison in 1849 and is still far from complete.

Each adrenal consists embryologically, histologically, and functionally of 2 distinct glandular entities that are grossly combined into one organ. Cells of the adrenal cortex secrete steroid hormones, which were discussed previously (see page 177).

The adrenal medulla is composed of cells that migrated out from the embryonic neural crest and are analogous to the peripheral sympathetic neurons of the autonomic nervous system. The adrenal medulla secretes epinephrine and norepinephrine (normally in a ratio approximately 17:3) and functions as a sympathetic postganglionic structure.

The adrenal medulla is not essential for life, and no diseases of deficiency are known. Therapeutic use of these hormones is based on the pharmacology of sympathomimetic amines and not on the principle of replacement. Epinephrine elicits its vasoconstrictor and vasopressor responses, acting in general as a sympathomimetic agent of rapid onset but brief duration of action. It is administered by intravenous or intramyocardial injection in cardiac arrest. Bronchodilation, resulting from epinephrine's *beta*-receptor adrenergic activity, is particularly useful in alleviating acute asthmatic attacks. The labile catechol function precludes oral administration of epinephrine, which must be administered by subcutaneous or intramuscular injection or absorbed through a mucous membrane.

The catecholamine hormones are metabolically inactivated in several ways. A major pathway involves catechol *O*-methylation, but oxidative deamination with monoamine oxidase is especially significant owing to the therapeutic use of MAO inhibitors.

Biosynthesis of Epinephrine. From the biosynthetic viewpoint, epinephrine may be considered an alkaloidal amine of the phenylpropanoid type. Its derivation from tyrosine has been demonstrated experimentally. Tyrosine is oxidized to dihydroxyphenylalanine (dopa), which is decarboxylated and oxidized in the side chain. The norepinephrine thus produced is converted to epinephrine by transfer of a methyl group from active methionine (Fig.

9–1). The rate-limiting step appears to be the conversion of tyrosine to dopa.

Epinephrine is (−)-3,4-dihydroxy-α-[(methylamino)methyl] benzyl alcohol. It may be isolated as a hormone from adrenal medulla or may be prepared synthetically. Dextrorotatory epinephrine is almost completely inactive, and optically inactive mixtures have approximately half the activity of natural epinephrine. The specific rotation of epinephrine is not less than −50° and not more than −53.5°. Epinephrine occurs as a white to nearly white, microcrystalline, odorless powder that gradually darkens when exposed to light and air.

PRESCRIPTION PRODUCTS. Adrenalin®, Epifrin®, Epinal®, Eppy/N®, Epitrate®, Glaucon®, and Sus-Phrine®.

PROPRIETARY PRODUCTS. Adrenalin®, AsthmaNefrin®, Bronitin Mist®, Bronkaid Mist®, Medihaler-Epi®, Primatene Mist®, and Vaponefrin®.

Epinephrine is incorporated into a variety of pharmaceutic formulations for therapeutic utilization. The hormone is frequently solubilized in aqueous preparations using hydrochloric acid or tartaric acid (bitartrate), and a water-soluble borate complex is sometimes used in ophthalmology. Acidity favors the stability of this labile compound, but no epinephrine-containing solution should be used if a brown color or a precipitate has formed. Formulations for therapeutic use of this hormone include a 1:1000 aqueous solution for topical purposes, a 1:100 aqueous solution for inhalation, metered-dose aerosol products, sterile aqueous solutions (1:1,000, 1:10,000, and 1:100,000) for parenteral administration, a sterile 1:200 suspension in oil for prolonged systemic action, and several ophthalmic solutions (1:50 to 1:400) for use in open-angle glaucoma and for other ophthalmic purposes.

Levarterenol or (−)-norepinephrine is (−)-α-(aminomethyl)3,4-dihydroxybenzyl alcohol. It is usually used as the bitartrate salt, which is a white or faintly gray, crystalline powder. It slowly darkens when exposed to air and light. Solutions of this hormone should not be used if they are brown in color or contain a precipitate. Levarterenol is a sympathetic stimulant closely related in chemical structure and pharmacologic action to epinephrine. Its chief difference in clinical utility lies in its predominantly *alpha*-receptor adrenergic activity. It is a strong peripheral vasoconstrictor and is especially useful in restoration of blood pressure in acute hypotensive situations.

USUAL DOSE. Levarterenol bitartrate is usually administered by intravenous infusion. Usually, the equivalent of 4 mg of levarterenol is placed in 1000 ml of 5% dextrose injection and infused at a rate adjusted to maintain blood pressure.

PRESCRIPTION PRODUCT. Levophed®.

Dopamine or 3,4-dihydroxyphenylethylamine is a biosynthetic precursor of norepinephrine and epinephrine, and this endogenous catecholamine has sympathomimetic properties. It acts directly to stimulate *alpha* and *beta-1* receptors and indi-

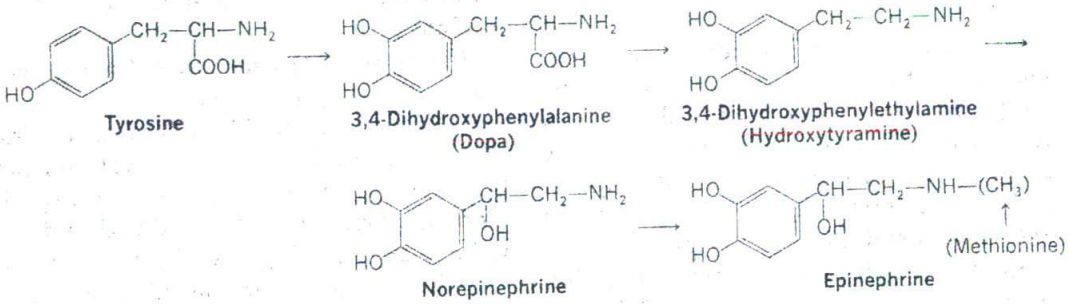


Fig. 9–1. Biosynthesis of epinephrine.

rectly by causing the release of stored norepinephrine. Response of the adrenergic receptors to dopamine is dose-dependent, higher concentrations being required to elicit *alpha*-receptor effects.

Dopamine is used to treat cardiac decompensation and patients with acute hypotension. It is administered by intravenous infusion. Initial infusion rates are 0.5 to 5 μg per kg of body weight per minute; most patients can be maintained on a dose of 20 μg per kg of body weight per minute or less.

PRESCRIPTION PRODUCTS. Dopastat®, Intropin®.

THYROID GLAND

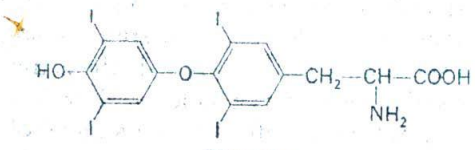
The **thyroid gland** in man consists of 2 lobes that are lateral and inferior to the anterior aspect of the larynx and are connected across the larynx by an isthmus to produce a U-shaped structure averaging 30 g in weight. Galen vaguely described the thyroid in the 2nd century, but its identity as a ductless gland was first described by Holler in 1776. Interestingly, Roger of Palermo used sponges and seaweed, high in iodine content, in the treatment of goiter (thyroid enlargement) in the 12th century.

Thyroid gland mobilizes dietary iodine, converting it to an organic compound that can accelerate metabolic processes. It is necessary to the development and function of all body cells. The actual molecular form of the thyroid hormone or hormones, exclusive of calcitonin which has a hypocalcemic action that is distinct from the physiologic responses usually associated with this gland, remains unclear. The iodine-containing levorotatory amino acids, thyroxine and triiodothyronine, occur in the gland and are physiologically active upon oral administration. These metabolites also occur in the gland bound with globulin (thyroglobulins), and the exogenously administered amino acids could conceivably bind with serum protein to form physiologically active molecules that are responsible for the ultimate hormonal action. The thyroid gland can store thyroglobulins and other iodometabolites. The release of these thyroid hormones appears to be controlled by thyrotropin, a hormone of the anterior pituitary.

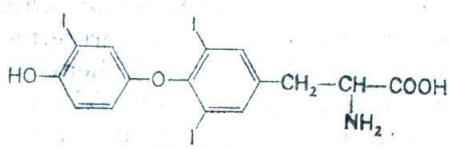
Manifestations of hypothyroidism may be caused by an iodine deficiency and a resulting lack of precursor moieties for the hormonal substances (simple hyperplastic goiter, which is characterized by a compensatory enlargement of the gland), by a deficiency of thyrotropic factors, or by other metabolic irregularities. The first two causes may be corrected by adding iodine to the diet or administering thyrotropin, respectively. Naturally, replacement therapy with thyroid hormones can be used for a deficiency of any origin.

A hypothyroid condition results in some degree of cretinism in infants and of myxedema in adults. Cretinism is characterized by retarded and abnormal growth; arrested sexual development; mental deficiency; thickened, dry skin; thickened tongue; coarsened features; and a fall in the metabolic rate. The features of myxedema include general lethargy; retarded mental processes; increased body fat; susceptibility to cold and fatigue; cardiac dilatation; dry, thickened skin; and coarsened features with a thickened, protruding tongue.

Thyroid hyperactivity results in thyrotoxicosis characterized by increased heart rate, blood pressure, nervous excitability, and metabolic rate; muscular weakness



Thyroxine



Triiodothyronine

with tremor; loss of body weight and fat; and an increased tolerance to cold but intolerance to heat. When accompanied by protrusion of the eyeballs (exophthalmos), the condition is known as exophthalmic goiter (Graves' or Basedow's disease). The course of the disease is marked by occasional crises or "storms," which may result in abrupt death. These symptoms of thyroid hyperactivity may result from overdose of thyroid preparations; thus, the rationality of using thyroid preparations in obesity "cures" is questionable. Treatment of organic hyperthyroidism is principally surgical, aided by radioactive iodine and by antimetabolites such as propylthiouracil. Improved manipulation of the latter aids may even supplant surgery in selected cases.

Preparations of thyroid hormones are useful for replacement therapy in cretinism or myxedema. Early diagnosis and treatment are essential in cretinism to avoid irreversible body and mental retardation. These preparations are also employed to prevent myxedema in cases when the thyroid gland must be surgically removed. Preparations from the thyroid gland are sometimes used as pharmacologic agents for their influence on various metabolic processes, but the success of such therapy is difficult to predict.

The responses elicited by exogenous thyroxine and triiodothyronine differ quantitatively. Triiodothyronine exhibits a more rapid onset and a shorter duration of action. This difference appears to be related to the extent of ionization of the 4'-phenolic group in the respective substances at physiologic pH. Thyroxine is approximately 90% ionized and is predominantly protein-bound in the blood. The reversible binding to plasma proteins is apparently electrostatic, and the strength of the bond varies among the different plasma proteins. Drugs such as salicylates and phenytoin disrupt the weaker bonds (potential drug-drug interaction) to increase the physiologic impact of thyroid hormones.

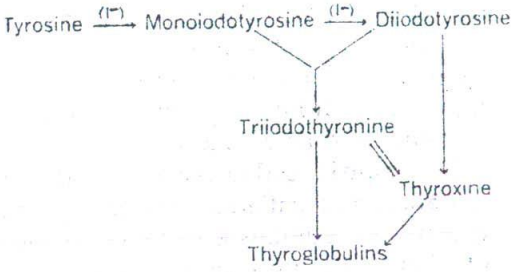


Fig. 9–2. Biosynthesis of thyroid hormones.

Relative binding and biopharmaceutic properties of thyroxine and triiodothyronine are also illustrated by their normal serum levels. The normal human serum levels of triiodothyronine range from 60 to 160 ng per ml, but the concentration of thyroxine is approximately 60 times higher.

Biosynthesis of Thyroid Hormone. The reactions leading to thyroxine formation in the thyroid gland are still incompletely understood. The first step in the biosynthesis is a peroxidation of iodide to "active iodine," which then reacts with tyrosine to first form 3-monoiodotyrosine and subsequently 3,5-diiodotyrosine. Two molecules of the latter compound react to form thyroxine. Alternatively, a molecule of the diiodotyrosine may react with a molecule of monoiodotyrosine to form triiodothyronine (liothyronine), which is then iodinated to yield thyroxine (Fig. 9–2). Some evidence shows that thyrotropin exerts an influence on the condensation of 2 molecules of the iodoamino acids to yield either thyroxine or triiodothyronine, and some degree of reversibility has been noted for the biosynthetic conversion of triiodothyronine to thyroxine. Triiodothyronine is the most active of the known thyroid metabolites, and approximately one third of the thyroxine excreted by the normal thyroid gland undergoes metabolic deiodination to triiodothyronine in peripheral tissues. Such considerations suggest that triiodothyronine is truly the active molecular form responsible for thyroid activity, but the significance of its metabolic relationship with thyroxine relative to

biologic control processes needs further clarification.

Deiodination in various body tissues is the major catabolic pathway for thyroxine and triiodothyronine. However, other metabolic pathways are known, including deamination and oxidation in the kidney to form acetic acid analogs and conjugation in the liver followed by biliary excretion. Some differences have been detected in the latter pathway. The β-glucuronide is the major conjugation product of thyroxine, but the sulfate ester is more common for triiodothyronine. None of the catabolic pathways is especially rapid, and manipulation of these processes offers no significant potential for achievement of therapeutic objectives.

Thyroid is the cleaned, dried, and powdered thyroid gland previously deprived of connective tissue and fat. It is obtained from domesticated animals that are used for food by humans. Thyroid contains not less than 0.095% and not more than 0.125% of total thyroxine and triiodothyronine, and the ratio of thyroxine to triiodothyronine is not less than 5. It is free from iodine in inorganic or any form of combination other than that peculiar to the thyroid gland.

USE AND DOSE. Thyroid is effective in oral therapy. The usual dose is 15 to 180 mg daily. The effect of a single dose of thyroid orally, or of thyroxine orally or intravenously, is not manifest for some 24 to 48 hours; it reaches a maximum in 8 to 10 days and decreases slowly over a period of several weeks. Hence, accumulation may occur, and dosage schedules must be adjusted individually to the needs of the patient.

PRESCRIPTION PRODUCTS. S-P-T®, Thyrar®, and Thyro-Teric®.

Thyroglobulin is obtained by fractionation of porcine thyroid gland. It contains thyroxine and triiodothyronine in a ratio of not less than 2.8. The source is restricted to *Sus scrofa* Linné var. *domesticus* Gray (Fam. Suidae) because the hog accumulates especially high levels of the thyroid hormones and has a higher proportion of triiodothyronine.

USE AND DOSE. Thyroglobulin is used in essentially the same manner as thyroid. The usual daily dose is 16 to 200 mg.

PRESCRIPTION PRODUCT. Proloid®.

Sodium levothyroxine is the sodium salt of the levo isomer of thyroxine, an active physiologic principle obtained from the thyroid gland of domesticated animals used for food by humans. It can also be prepared synthetically.

USES AND DOSE. Sodium levothyroxine is classed as a thyroid hormone. It is used for replacement therapy of reduced or completely absent thyroid function (manifested as myxedema, cretinism, and mild forms of hypothyroidism). Consistent potency, prolonged duration of action, and easily monitored plasma levels prompt many authorities to consider levothyroxine the agent of choice for thyroid replacement therapy. It is also used as adjunct therapy to suppress thyrotropin release in adenoma goiter and thyrotropin-dependent thyroid gland carcinoma. Its use to treat such vague symptoms as dry skin, fatigue, habitual abortion, obesity, and sterility in the absence of confirmed hypothyroidism is inappropriate and may be dangerous. The usual dose is 25 to 300 μg once a day.

PRESCRIPTION PRODUCTS. Levothroid®, Synthroid®, Synthrox®, and Syroxine®.

Sodium liothyronine is the sodium salt of the levorotatory isomer of 3,3',5-triiodothyronine. This physiologically active compound is a naturally occurring thyroid hormone, but quantities needed for commercial purposes are provided by chemical synthesis.

USE AND DOSE. Sodium liothyronine is used for the same purposes as sodium levothyroxine. Liothyronine, compared to other thyroid agents, has better gastrointestinal absorption, a more rapid onset of action, and a shorter duration of action. The usual dose is the equivalent of 5 to 100 μg of liothyronine, once a day.

PRESCRIPTION PRODUCTS. Cytomel®, Cytronine®.

Liotrix is a 4:1 mixture of synthetic sodium levothyroxine and sodium liothyronine. The effects of this mixture are claimed to resemble closely those of endogenous thyroid secretion and to give laboratory protein-bound iodine test results that are more consistent with clinical response than are the results obtained with other preparations. However, recognition of the conversion of thyroxine to triiodothyronine in peripheral tissues raises questions about any therapeutic advantage of this mixture over its individual components.

PRESCRIPTION PRODUCTS. Euthyroid®, Thyrolar®.

Consensus equivalents for comparison of the various thyroid preparations, based on 65 mg (1 gr) of thyroid are thyroglobulin, 65 mg; sodium levothyroxine, 100 μg; sodium liothyronine, 25 μg; and liotrix formulations containing 50 or 60 μg of levothyroxine and 12.5 or 15 μg of liothyronine.

Sodium dextrothyroxine is the salt of the synthetically prepared dextrorotatory isomer of thyroxine. This substance is effective in a high dose (up to 8 mg daily) for the treatment of hypothyroidism, but its occasional use for such purposes is restricted to patients with cardiac disease who cannot tolerate other thyroid medications.

Dextrothyroxine also reduces serum cholesterol and low-density lipoproteins, and it has been classed as a hypocholesterolemic agent. Its greatest therapeutic utility is in this area, but patients must be monitored carefully for ischemic myocardial changes and other adverse reactions. The hypocholesterolemic dosage regimen starts with 1 mg daily for a month; the dosage is increased in 1-mg increments at intervals no shorter than 1 month until a satisfactory lowering of β-lipoprotein has been achieved or until a maximum daily dose of 8 mg is reached.

PRESCRIPTION PRODUCT. Choloxin®.

PITUITARY

The human **pituitary gland or hypophysis** is situated in a small cavity in the sphenoid bone at the base of the skull and is attached to the base of the brain by a short stalk. It weighs about 0.5 g. Galen considered it a strainer for spinal fluid, and Vesalius later thought it was the source of mucus, lubricating the nasopharynx. Pituitary is from the Latin *pituita*, meaning slime or mucus.

The pituitary body is in reality 2 glands by origin and function:

1. The anterior lobe is ectodermal in origin—derived from an outpouching from the primitive pharynx.
2. The posterior lobe is neural in origin—derived from an outpouching of the base of the brain.

Posterior Pituitary

Extracts of **posterior pituitary** lobe exhibit the following effects in experimental animals and in humans:

1. A pressor effect, owing to arteriolar and capillary vasoconstriction;
2. Direct stimulation of smooth muscle, seen in the intact animal or in preparations of isolated muscles;
3. An antidiuretic action, effected by increasing the tubular and collecting duct resorption of water in the kidney.

The diversity of these effects complicated attempts at therapeutic utilization of unfractionated extracts of the posterior pituitary (Pituitrin S®), and the use of such preparations has been largely superseded by the use of oxytocin (ocytocin) or vasopressin, the 2 hormonal substances that have been isolated from this gland. However, these 2 hormones do not appear to provide a clear-cut separation of the physiologic properties associated with the secretions of the gland. The 2 hormones are closely related octapeptides (Figure 9–3) with 5 of the amino acid residues in a cyclic structure involving a labile cystine moiety. These similar cyclopeptides are difficult to separate quantitatively, and part of the lack of definitive pharmacologic responses

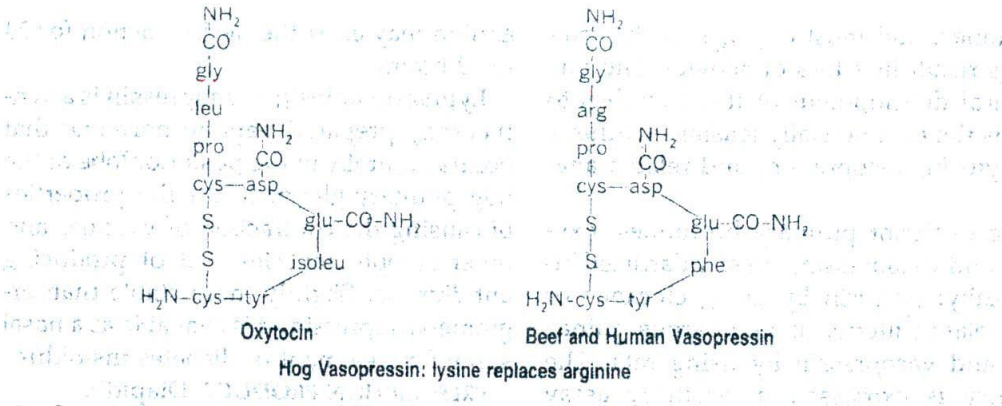


Fig. 9–3. Structures of oxytocin and vasopressin.

using the hormones isolated from nature has been correctly attributed to impurities. This problem is eliminated when the hormones are prepared synthetically, but their actions, especially those of vasopressin, still show a degree of overlap.

Oxytocin (α-hypophamine) is the uterine-stimulating fraction, and it is relatively free from action on other smooth muscle. It is especially active on the pregnant uterus, which has been sensitized by estrogens. Oxytocin appears to increase the permeability of uterine cell membranes to sodium ion with an effective augmentation of the contracting myofibrils. Some physiologic involvement by oxytocin in the normal onset of labor is suspected, but further scientific clarification is needed.

Vasopressin (β-hypophamine) is the antidiuretic principle; the pressor effects of this hormone are observed only when large quantities are administered. Vasopressin regulates the threshold for resorption of water by the epithelium of the renal tubules. The hormone is released into the blood when osmoreceptors in the hypothalamic nuclei detect an increased extracellular electrolyte concentration in the serum or a decreased blood volume. The resulting fluid conservation contributes to maintenance of homeostasis.

No clinical conditions have yet been associated with hyperfunction of posterior pituitary. A deficiency state is seen only in the condition of diabetes insipidus, which follows a deficiency of the antidiuretic principle. The increased urine output when drinking alcoholic beverages is caused physiologically, in part, by a temporary deficiency state because alcohol inhibits the release of vasopressin.

Diabetes insipidus (literally an outpouring of tasteless urine) is characterized by a failure of renal resorption of water—there is a tremendous diuresis and associated tremendous thirst and water intake. This condition must not be confused with the diabetes mellitus of insulin deficiency.

The biologic half-lives of oxytocin and vasopressin are short, a factor that can be considered an advantage or a disadvantage, depending on therapeutic objectives. Vasopressin is employed in replacement therapy for the management of diabetes insipidus, and one of the concerns in formulation is a need for an increase in the duration of action. Oxytocin is used as a pharmacologic agent for its oxytocic properties, and thus the desired duration of therapy is short.

The relatively small size of these polypeptides provided a focusing point for initial studies on chemical synthesis and structure-activity comparisons that undoubtedly have implications for all physiologically active polypeptides. No prepared analogs, with one possible exception, have been superior in therapeutic properties to the 2 naturally occurring

hormones, and most changes in the molecules result in a loss of activity. The one practical development of these studies to date is the commercially feasible synthesis of oxytocin, vasopressin, and related analogs.

The posterior pituitary hormones, oxytocin and vasopressin, are standardized biologically; oxytocin by using chickens or the isolated uterus of nulliparous guinea pigs and vasopressin by using rats. The potency is expressed in arbitrary assay units based on a Posterior Pituitary Reference Standard.

Vasopressin injection is a sterile solution in water for injection of the water-soluble, pressor principle prepared by synthesis or obtained from the posterior lobe of the pituitary. Either 8-L-arginine-vasopressin (beef vasopressin) or 8-L-lysine-vasopressin (hog vasopressin) meets monographic requirements.

Vasopressin injection is standardized so that 1 ml possesses a pressor activity equivalent to 20 USP posterior pituitary units. It is used to control neurohypophyseal diabetes insipidus but is ineffective in the nephrogenic form of the disease. It is also used as a peristaltic stimulant in postoperative ileus and to control acute hemorrhage in the gastrointestinal tract and esophagus. The usual dose, intramuscularly or subcutaneously, is 5 to 10 units, 2 or 3 times a day.

PRESCRIPTION PRODUCT. Pitressin®.

The dosage of vasopressin must be adjusted to the needs of the individual patient, but convenient therapy is handicapped to a degree by the relatively rapid inactivation of the hormone in the body. The need to administer 5 to 10 USP units of vasopressin (vasopressin injection), 2 or 3 times daily, is common. Formulation of vasopressin tannate in peanut oil (Pitressin® Tannate) was developed to give a gradual release of the hormone and a longer duration of action; an intramuscular injection of 2.5 to 5 USP units of this preparation may exert the desired action for 24 to 72 hours.

Lypressin or lysine-vasopressin is a synthetically prepared peptide hormone that occurs naturally in the posterior lobe of the hog pituitary gland. It has the properties of causing the contraction of vascular and other smooth muscles and of producing antidiuresis. Slightly more stable than arginine-vasopressin, it is available as a nasal spray for treatment of diabetes insipidus.

PRESCRIPTION PRODUCT. Diapid®.

Desmopressin or 1-deamino-8-D-arginine-vasopressin is a synthetic analog of arginine-vasopressin. It possesses very little of the undesirable pressor effect of the parent hormone and has a relatively long duration of action. Desmopressin is used for the same purposes as vasopressin and is available as an injection and a nasal solution. The usual parenteral dosage for diabetes insipidus is 2 to 4 μg daily in 2 divided doses.

PRESCRIPTION PRODUCTS. DDAVP® and Stimate®.

Oxytocin is a synthetically prepared peptide hormone that occurs naturally in the posterior lobe of the pituitary gland. Glandular material is no longer used as a commercial source of this hormone. The oxytocic principle is available as an injection for induction of labor for medical indications (use for elective induction is inappropriate) and for control of postpartum hemorrhage. A nasal spray is available to promote milk ejection on the infrequent occasions when this is a problem in breast feeding.

The dosage for induction of labor is determined by uterine response; the intravenous infusion is started at a rate of 0.001 to 0.002 units per minute. The dose is increased in similar increments until a contraction pattern that resembles normal labor has been established. The dosage for control of postpartum hemorrhage is 10 units intramuscularly after delivery of the placenta.

PRESCRIPTION PRODUCTS. Pitocin®, Syntocinon®.

Anterior Pituitary

The anterior lobe of the pituitary exerts a profound influence on the growth and development of the body and on its sex characteristics through its stimulating actions on the other endocrine glands. The **anterior pituitary** has been referred to as the "master gland," the "conductor of the endocrine symphony." Primary disturbances in anterior pituitary function may result in widespread endocrine involvement and generalized secondary disturbances. Therapy in such complex situations is far from simple and is associated with unusual potential for undesirable side effects. Effective therapeutic utilization of the hormones of the anterior pituitary is still in an early stage of development.

Hormones with adrenocorticotropic, gonadotropic, and thyrotropic activities are used for therapeutic or diagnostic purposes, as is growth hormone (somatropin). Another hormone recognized as originating in the anterior pituitary is prolactin. All of these hormones are glycoproteins or proteins, and they tend to be produced and accumulated in small quantities. Available evidence indicates that, with the exception of adrenocorticotropin, their molecular weights are in the 20,000 to 40,000 range, considerable interrelationships exist among subunits of the various hormonal substances, and labile disulfide bonds occur in these hormones. These factors contribute to the difficulty in securing concentrated, purified extracts with only a single hormonal activity. Technical barriers to the feasible synthesis of such molecules have further limited the medicinal utilization of the anterior pituitary hormones. They appear to have short biologic half-lives (less than 30 minutes), but preliminary evidence suggests that the length of half-life may vary with the pathologic status.

The gonadotropic hormones present several interesting features. At least 2 gonadotropic hormones are excreted by the anterior pituitary; these hormones elicit distinct responses in females and males, and glycoproteins with similar activities are produced by the chorionic villi of the placenta. The follicle-stimulating hormone (FSH or follitropin) is necessary for maturation of the ovarian follicles in females and for maturation of the seminiferous tubules of the testes in males. Luteinizing hormone (LH or lutropin) or interstitial cell-stimulating hormone is essential for causing ovulation and for the development and maintenance of the corpus luteum in the ovary. In the male, LH is apparently active in the development of the Leydig cells of the testes.

Feedback, hypothalamic, and perhaps other mechanisms stimulate the secretion of the tropic hormones of the anterior pituitary. Hypothalamic releasing factors have received considerable attention recently, and a tripeptide and a decapeptide from the hypothalamus have been shown to cause the release of thyrotropin and gonadotropins, respectively. The latter factor appears to lack specificity and to initiate the release of both FSH and LH. The potential for administration of releasing factors of small molecular size is obvious, but problems with variable response preclude therapeutic application at the current stage of knowledge. The first implication of a releasing factor to achieve therapeutic utility may involve inhibition. Prolactin facilitates development of breast cancer, and the inhibition of prolactin release holds clinical promise. Gonadotropin and thyrotropin releasing factors are used for diagnostic purposes.

Adrenocorticotropin, ACTH, or corticotropin is a straight-chain polypeptide containing 39 amino acid residues. A portion of the peptide molecule with 20 amino acid residues has the full biologic activity of this hormone. Corticotropin injection is a sterile preparation of the peptide hormone that is derived from the anterior lobe

of the pituitary of mammals used for food by humans and that exerts a tropic influence on the adrenal cortex. Its tropic effects involve primarily glucocorticoids. Corticotropin may be used in collagen disease, particularly in rheumatoid arthritis and acute rheumatic fever, when the adrenal cortex is functional; it avoids the glucocorticoid-associated problem of atrophy of the adrenal cortex. The usual dose, intramuscularly, is 20 USP units, 4 times a day. It can be administered by intravenous infusion if a rapid response is desired; however, this situation is uncommon because the most frequent objection to the use of corticotropin injection is the short duration of action.

PRESCRIPTION PRODUCT. Acthar®.

Repository corticotropin injection is corticotropin in a solution of partially hydrolyzed gelatin (Cortigel®, Cortrophin® Gel, Cotropic® Gel, H P Acthar® Gel). This formulation has a prolonged therapeutic effect; its usual intramuscular dose is 40 USP units, once daily. A similar, prolonged duration of action is obtained with a suspension prepared by adsorbing corticotropin on zinc hydroxide (Cortrophin® Zinc).

Cosyntropin (Cortrosyn®) is a synthetically prepared peptide subunit of corticotropin and is used as a diagnostic aid in suspected adrenal insufficiency. It contains 24 amino acid residues and presents a lesser risk of allergenic reactions than the natural hormone.

Chorionic gonadotropin (HCG or choriogonadotropin) is a gonad-stimulating polypeptide hormone obtained from the urine of pregnant women. This anterior-pituitarylike substance resembles LH in its response and is used as replacement therapy to stimulate descent of the testes in cryptorchidism and to stimulate the development of interstitial cells of the testes in delayed adolescence and hypogonadotropic eunuchoidism. The major indication for withdrawal of therapy or reduction of dosage is sexual precociousness. Other degenerative side effects may be noted in females; hence, chorionic gonadotropin is infrequently used in females although it can be used effectively in conjunction with menotropins to treat infertility and to maintain the functional integrity of the corpus luteum in some cases of habitual abortion. HCG has been used in weight-loss clinics, but its effectiveness in adjunct therapy for the treatment of obesity has not been demonstrated.

Chorionic gonadotropin is a relatively labile glycoprotein that contains about 12% of galactose. It must be administered parenterally and is formulated in a dry mixture with suitable diluents and buffers to give greater shelf life. The usual dose, intramuscularly, is 500 to 5000 USP units, 3 times a week.

PRESCRIPTION PRODUCTS. Android-HCG®, A.P.L. Secules®, Chorex®, Corgonject-5®; Follutein®, Glukor®, Gonic®, Libigen®, Pregnyl®, Profasi HP®.

Menotropins or urogonadotropin is a purified preparation of gonadotropins obtained from the urine of postmenopausal women. The urine gonadotropin levels are high in such women because atrophic ovaries cannot respond to tropic stimulation, thus precluding feedback suppression. The preparation contains approximately equal amounts of FSH and LH. It is used to enhance fertility in anovulatory women with functional ovaries and to stimulate spermatogenesis in hypogonadotropic males.

The usual dosage regimen for females involves intramuscular injection of 75 units each of FSH and LH daily for 9 to 12 days to stimulate follicular growth and maturation. One day after the last menotropins injection, 10,000 units of HCG is administered to simulate a preovulatory LH surge and to induce ovulation.

The usual therapeutic regimen for males involves 4 to 6 months of pretreatment with HCG (5000 units 3 times a week). Menotropin (75 units each of FSH and LH) is administered intramuscularly 3 times a week, and concurrent administration of HCG is continued at the rate of 2000 units

twice weekly. The menotropin therapy should continue for at least 4 months.

PRESCRIPTION PRODUCT. Pergonal®.

Gonadorelin is a synthetic decapeptide that is identical to the gonadotropin-releasing factor of the hypothalamus. It is used diagnostically in suspected gonadotropin deficiency to evaluate the functional capacity and response of the gonadotropes of the anterior pituitary. The test in females should be conducted in the early follicular phase of the menstrual cycle. The usual test dose is 100 μg intravenously or subcutaneously.

PRESCRIPTION PRODUCT. Factrel®.

Somatropin, somatotropin, or growth hormone influences a number of essential growth processes. It stimulates linear growth of bones during development, and its anabolic effects include an increased intracellular transport of amino acids and a net body retention of nitrogen, phosphorus, and potassium. Hypofunction of the growth-stimulating activity in children results in the pituitary dwarf; in adults, such deficiency often produces an increased delicacy of structure, referred to as acromicria.

Growth hormones from various animal sources appear to be more species-specific in their activity than most other hormones. Human pituitary growth hormone contains 190 amino acid residues and 2 disulfide bonds. Although lack of a feasible natural source or a feasible synthetic procedure for this pituitary hormone precluded consideration of therapeutic use for many years, the discovery of a human chorionic analog appeared to circumvent these logistic barriers. The production of biosynthetic somatropin using modern technology is showing promise.

Serious safety questions have been raised recently about human-derived somatropin, and commercial supplies of the hormone were withdrawn from the market, at least temporarily, in mid-1985. A few patients who received this hormone between 1963 and 1980 died of apparent Creutzfeldt-Jakob disease. This rare degenerative neurologic disorder is caused by a "slow virus" that is resistant to conventional methods of sterilization, and transmission of the virus as a contaminant in somatropin injection is suspected. It is anticipated that human somatropin preparations will become available once again as soon as documentation is obtained that current purification procedures eliminate the contamination. Somatrem (Protropin®), a biosynthetic methionyl human growth hormone obtained by recombinant DNA technology, received accelerated FDA review and was approved for distribution in late 1985.

When somatropin is employed therapeutically, it is used to stimulate linear growth in patients with documented pituitary growth hormone deficiency. It is not indicated for use in patients with closed epiphyses. Many interactions, including influences of hormones of the adrenal cortex, gonads, parathyroid, and thyroid, contribute to growth. Therapy with somatropin must be monitored carefully. Use of this hormone is restricted to physicians experienced in the diagnosis and management of patients with pituitary growth hormone deficiency.

The usual dosage regimen of somatropin is 2 international units, intramuscularly, 3 times a week (a minimum of 48 hours between injections) for as long as growth continues. Treatment should be discontinued when the patient has reached a satisfactory adult height, when the epiphyses have fused, or when response to the therapy is no longer satisfactory. Five percent of the patients can be expected to develop neutralizing antibodies and to fail to respond to the hormone.

Thyrotropin is the thyrotropic principle of the anterior pituitary. A glycoprotein in the 28,000 to 30,000 molecular weight range, it is obtained from bovine glands and is purified to remove significant amounts of corticotropic, gonadotropic, and other hormones. Theoretically, it can be used in replacement therapy, but it is

used primarily as a diagnostic aid in evaluating thyroid function, including distinction between primary and secondary hypothyroidism, or as supportive therapy to facilitate the uptake of ^{131}I in treatment of toxic goiter or thyroid carcinoma. Thyrotropin is available as a lyophilized powder and is administered intramuscularly or subcutaneously, usually in daily doses of 10 international units.

PRESCRIPTION PRODUCT. Thytropar®.

Protirelin is the synthetic tripeptide, L-pyroglutamyl-L-histidyl-L-prolinamide. Identical to the thyrotropin-releasing factor from the hypothalamus, it is used diagnostically to assist in distinguishing secondary and tertiary hypothyroidism. The usual adult dose is 500 μg given intravenously.

PRESCRIPTION PRODUCTS. Relefact-TRH® and Thypinone®.

PANCREAS

The bulk of the **pancreas** is an exocrine gland that supplies digestive enzymes to the duodenum. Isolated groups of cells, the islets of Langerhans constituting about 3% of the gland, produce the hormonal substances. **Glucagon** is produced by the α-cells, and **insulin** is formed by the β-cells. Glucagon and insulin, both polypeptide hormones, exert counterbalancing actions on carbohydrate metabolism in the body.

Glucagon elicits a hyperglycemic response by increasing adenyl cyclase which, in turn, increases liver phosphorylase activity, a key factor in glycogenolysis. The hypoglycemic action of insulin appears to involve glucokinase, membrane transport, and perhaps other factors associated with normal metabolism of blood glucose. In the normal individual, these hormones function to maintain blood glucose within a physiologically tolerated balance by increasing or decreasing, respectively, the glucose level.

Pathologic conditions related to a deficiency in glucagon formation either do not occur, have an insufficient survival factor to create a medical problem, or are unrecognized. The typical case of a hypofunctioning pancreas gland results in insulin deficiency and the condition known as diabetes mellitus. This condition was described by Auretaeus in the first century A.D. as a siphoning of flesh into urine; it is characterized by a high blood-glucose level (hyperglycemia), excess glucose in the urine (glucosuria), and diuresis, resulting in dehydration and constipation. The primary impairment of glucose metabolism induces a number of secondary metabolic changes. Accumulation in the blood of such metabolites as β-hydroxybutyric acid, ketone bodies, and other breakdown products of fats is common. The untreated diabetic, as a result of such metabolic irregularities, suffers from severe acidosis, depression, coma, and ultimately death.

Treatment of diabetes mellitus with insulin is replacement therapy. Insulin prolongs life in the diabetic and permits a fuller and happier life, but its use does not cure or prevent the pathologic condition. Insulin is especially valuable in preventing the complications of diabetes that are frequently the cause of death: arteriosclerosis with hypertension, nephritis, superficial ulcers and infections, gangrene of the extremities, and gallstones.

Conditions of hyperinsulinism may result from overdosage of insulin, underfeeding, tumors of the pancreas, or certain pituitary or adrenal disturbances. Outstanding symptoms are fatigue, hunger, marked sweating, and convulsions.

In management of diabetes mellitus caused by insulin deficiency, an adequate diet is determined, and the amount and spacing of insulin dosage are established to keep the patient symptom-free and free from glucosuria. The plasma half-life of insulin is approximately 10 minutes; therefore, continuous delivery of the hormone is needed in replacement therapy. This is accomplished by subcutaneous adminis-

tration of a suspension that is formulated to provide slow release of the insulin. Insulin is a potent drug, and the therapeutic considerations are relatively complex and sophisticated. Dosage must be coordinated with dietary intake, and a regularized eating schedule is necessary to accommodate the programmed delivery of insulin and to avoid drug-induced hypoglycemia. The over-the-counter status of insulin, compared with other drugs, is inconsistent with the inherent complexity and risks of the therapy, but it is indicative of what can be achieved by concerted patient education programs.

One USP unit of insulin can cause the metabolism of approximately 1.5 g of glucose. Occasionally, insulin resistance or tolerance develops, and abnormally high doses of insulin are required to control diabetes. The resistance appears to involve antibody binding to restrict the biologic availability of much of the administered hormone; a high potency insulin (500 units per ml) is available by prescription for such cases. The objective of high doses in resistant cases is distinct from such uses in treating schizophrenic states. An insulin overdose in the latter situation is used to induce therapeutically convulsive (hypoglycemic) shock.

Glucagon can be used in diagnosis and in the treatment of hypoglycemia associated with improper management of diabetes mellitus or with pyschiatrically induced insulin shock. Intravenous administration of glucose can also be used to treat hypoglycemia, but glucagon is more convenient to use with unconscious or uncooperative nonhospitalized patients.

Glucagon is a straight-chained polypeptide containing 29 amino acid residues with a molecular weight of 3485. The amino acid sequence has been determined. The isoelectric point of the hormone is between 7.5 and 8.5. It is soluble in acids and bases, below 3.0 and above 9.5, respectively; these solutions are relatively stable. The alkali stability of glucagon can be used for the selective inactivation of insulin, which is a labile cystine-containing contaminant that is difficult to remove quantitatively in the isolation of this hormone.

Insulin is a polypeptide with a molecular weight of 5734. It contains 48 amino acid residues (including 3 cystine residues) that are arranged in 2 linear chains connected by disulfide linkages. Insulin tends to form dimeric and hexameric forms, a characteristic that resulted in an initial estimate of about 35,000 for the molecular weight of the hormone. This hormone has been studied more extensively than any other polypeptide hormone. One of the interesting observations resulting from these studie is the variation in amino acid residues 8 9, and 10, depending on the origin of th insulin (Fig. 9–4). The amino acid sequenc in these 3 positions has no effect on th normal physiologic properties of the poly peptide. Variations in this portion of th molecule cause some antigenic reactions and this is the basis for selecting all porcin insulin in cases of suspected insulin hy persensitivity or tolerance. However, mos of the early cases of hypersensitivity to i sulin preparations can be explained b other foreign proteins that were present i the formulations. The distinctive amin acid residue 30 of the larger peptide chai of human insulin is apparently without physiologic significance because an analog lacking amino acid residues 26 to 30 retains activity.

The disulfide bonds that link the 2 peptide chains are major obstacles to the feasible synthesis of insulin, and the biosynthetic accomplishment of this feat has intrigued scientists. The recent elucidation of proinsulin has clarified significantly the biologic formation of the hormone. Proinsulin is a straight-chain polypeptide containing some 80 amino acid residues and all of the disulfide linkages inherent to insulin. The polypeptide sequences at the amino terminal and carboxylic acid terminal ends of the proinsulin molecule correspond to the 2 chains of insulin, and a

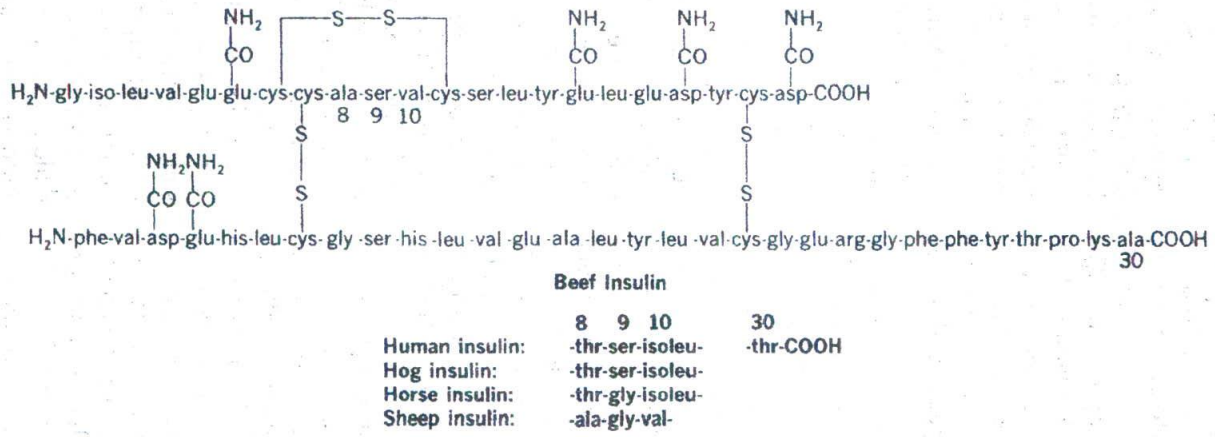


Fig. 9–4. Structures of insulin from several mammals.

proteolytic enzyme in the pancreas apparently cleaves peptide bonds to remove the connecting polypeptide sequence and to form the physiologically active, 2-chained insulin.

Insulin was crystallized in 1926 by the addition of traces of zinc, and crystals of zinc insulin formed the original reference standard of the USP. The isoelectric point of zinc insulin is 5.1 to 5.3. Thus, it is soluble at the alkaline pH of tissue fluids and is rapidly absorbed from subcutaneous injection sites. Insulin is digested by proteolytic enzymes (a common property of polypeptide hormones), and it is ineffective when given orally.

The production of both glucagon and insulin involves isolation of these substances from pancreas glands. The procedures are relatively complex, a situation that is common for the isolation and purification of most peptide molecules. At least 20 major steps are accomplished before a form of insulin suitable for human use is finally developed. Immediately following their removal from slaughtered animals, the raw beef and pork pancreases are frozen to prevent enzymatic destruction of the insulin in the gland. About 8000 lb of animal pancreases are needed to yield 1 lb of pure zinc insulin crystals.

The first step in actual production involves grinding of the frozen glands. Successive steps include extracting the still-frozen powder with acidic alcohol to obtain the insulin and to suppress enzyme activity, centrifuging the crude extract to separate the liquid from the glandular residue, clarifying the liquid extract containing the insulin and impurities, evaporating in vacuum to remove the alcohol, treating the concentrate to separate the fat, filtering to remove residual fat, adding salt water to precipitate the insulin, redissolving the precipitate, reprecipitating by isoelectric means, buffering to obtain a uniformly soluble product, washing, drying, and pooling the insulin obtained from other lots prepared in the same manner, and finally, determining the potency. Insulin prepared in this manner may be subjected to further purification by ion-exchange chromatography to yield a purified insulin product (not more than 10 parts per million proinsulin contaminant).

Commercial production of human insulin has been achieved using two independent approaches. Pork insulin is converted to human insulin by replacing enzymatically the terminal alanine amino acid residue with threonine. Recombinant DNA techniques have also been used for microbial synthesis. The genes (DNA) that direct the biosynthesis of the A and B polypeptide chains of human insulin are connected separately to DNA genetically coded for the enzyme β-galactosidase. This step puts the insulin genes under the control of a set of genes called the "lac operon"; this operon controls β-galactosidase synthesis and secretion. The connected genes are introduced separately into plasmids. The plasmids are then taken up by *Escherichia coli*, producing one strain that formed the A chain of insulin and another strain that formed the B chain. The addition of lactose to the bacterial cultures switches on the "lac operon" genes. This, in turn, switches on the gene for the production of either the A or the B chain of human insulin because these genes are now linked to the genes for the formation and secretion of β-galactosidase. The β-galactosidase is synthesized with the A and B chain of insulin attached, and these are recovered by chemically cleaving them from the enzyme. Consequently, the A chain of 21 amino acids and the B chain of 30 amino acids are purified separately, and the complete insulin molecule is chemically synthesized from the chains by forming 2 disulfide bridges.

The absolute therapeutic indication for human insulin may be rare. It produces a lower incidence of allergic reactions than many of the products from animal pancreas glands, and it may have special utility in the small proportion of diabetics allergic

to purified pork insulin. In addition, the bacterially produced hormone will relieve the American Diabetes Association's concern regarding a shortage of animal pancreas glands in the near future.

Glucagon for injection is a mixture of the hydrochloride with one or more suitable, dry diluents. When the aqueous injection is reconstituted, it has a pH between 2.5 and 3.0 and is usually formulated to contain 1 mg in each ml. The usual parenteral dose is 500 μg to 1 mg, repeated in 20 minutes, if necessary.

Insulin injection or insulin is a sterile, neutral solution of the active principle of the pancreas that affects the metabolism of glucose. Insulin injection contains 40, 100, or 500 USP insulin units in each ml. It is a prompt-acting preparation with a peak of action at 2 to 5 hours. This is the preparation of choice when glucose tolerance fluctuates rapidly; such situations may include the presence of a severe infection, shock, surgical trauma, or unstable diabetes. Insulin preparations, including insulin injection, must be labeled to indicate the nature (beef and pork, beef, pork, or human) of the insulin; when a product meets the standards for purified insulin, this must also be indicated on the label. The usual dose, for diabetic acidosis, intravenously, is 1 to 2 units per kg of body weight, repeated in 2 hours as necessary; for diabetes, 10 to 20 units, subcutaneously, 3 or 4 times a day according to the needs of the patient.

PROPRIETARY PRODUCTS. Beef and pork: Regular Iletin I®; purified beef: Beef Regular Iletin II®; purified pork: Actrapid®, Pork Regular Iletin II®, and Velosulin®; human: Humulin R®.

Preparations of insulin are marketed in multiple-dose ampules of varying unitage. The package color of commercial products varies with the unit value. There is increasing emphasis on use of 100-unit insulin formulations to reduce dosage errors. The need for 40-unit insulins is being studied, and the FDA may eventually decertify them.

Protamine zinc insulin suspension or protamine zinc insulin is a sterile suspension, in a phosphate buffer, of insulin modified by the addition of zinc chloride and protamine. The protamine is prepared from the sperm or from the mature testes of fish belonging to the genus *Oncorhynchus* Suckley, or *Salmo* Linné (Fam. Salmonidae). Protamine zinc insulin suspension provides 40 or 100 USP insulin units in each ml. It is a prolonged-acting insulin preparation. The usual dose, subcutaneously, is 7 to 20 units once a day.

PROPRIETARY PRODUCTS. Beef and pork: Protamine, Zinc and Iletin I®; purified beef or pork: Protamine, Zinc and Iletin II®.

Protamines are basic proteins; they combine with insulin to form protamine-insulin salts, stabilized by a trace of zinc. This complex has an isoelectric point of approximately 7.3; it is buffered to this point and dispensed in a smooth suspension. When injected subcutaneously, it is insoluble at the pH of tissue fluids and is therefore slowly absorbed to provide a prolonged action. Peak of action occurs at 14 to 20 hours, with some effect manifest over 36 hours.

Isophane insulin suspension, isophane insulin, or NPH insulin is a sterile suspension, in a phosphate buffer, of insulin made from zinc-insulin crystals modified by the addition of protamine in such a manner that the solid phase of the suspension consists of crystals composed of insulin, protamine, and zinc. It provides 40 or 100 USP insulin units in each ml. It is an intermediate-acting insulin preparation. The usual dose, subcutaneously, is 10 to 20 units, 1 or 2 times a day.

PROPRIETARY PRODUCTS. Beef and pork: NPH Iletin I®; purified beef: Beef NPH Iletin II®; purified pork: Insulatard NPH®, Pork NPH Iletin II®, and Protaphane NPH®; human: Humulin N®.

Isophane insulin is insoluble at the pH of tissue fluids and therefore has a slow absorption rate. Maximum effect occurs at

8 to 12 hours. A preparation containing 70% isophane insulin and 30% insulin (Mixtard®) is available in a 100-unit formulation to give a rapid onset of activity and a 24-hour duration of action.

Insulin zinc suspension or lente insulin is a type of intermediate-acting insulin preparation. It consists of a mixture of crystalline and amorphous materials (approximately a 7:3 ratio) suspended in an acetate buffer. It provides 40 or 100 USP insulin units in each ml. The usual dose, subcutaneously, is 10 to 20 units, once a day.

PROPRIETARY PRODUCTS. Beef and pork: Lente Iletin I®; purified beef and pork: Lentard®; purified beef: Beef Lente Iletin II®; purified pork: Monotard® and Pork Lente Iletin II®.

The use of an acetate buffer provides a prolonged duration of action. Achievement of this objective without the addition of foreign proteins, such as protamine, circumvents occasional hypersensitivity problems associated with these additives. The ratio of crystalline and amorphous insulin is selected to give a convenient duration of action of approximately 24 hours.

Extended insulin zinc suspension or ultralente insulin is a sterile suspension, in an acetate buffer, of insulin modified by the addition of zinc chloride in such a manner that the solid phase of the suspension is crystalline. It provides 40 or 100 USP insulin units in each ml. It is a long-acting insulin preparation; the duration of action is determined by the particle size and persists for over 36 hours. The usual dose, subcutaneously, is 7 to 20 units, once a day.

PROPRIETARY PRODUCTS. Beef and pork: Ultralente Iletin I®; purified beef: Ultratard®.

Prompt insulin zinc suspension or semilente insulin is a sterile suspension, in an acetate buffer, of insulin modified by the addition of zinc chloride in such a manner that the solid phase of the suspension is amorphous. It provides 40 or 100 USP insulin units in each ml. It is a rapid-acting insulin preparation; however, its duration of action extends from 12 to 16 hours. The usual dose, subcutaneously, is 10 to 20 units, 1 or 2 times a day.

PROPRIETARY PRODUCTS. Beef and pork: Semilente Iletin I®; purified pork: Semitard®.

The chief difference between prompt zinc insulin suspension and extended zinc insulin suspension is the size of the particles that make up the solid phase of the suspensions. Prompt zinc insulin suspension has a shorter duration of action. This product is not intended to replace insulin injection in combating diabetic acidosis or in emergencies when immediate action is essential. In such cases, insulin injection should be administered.

PARATHYROID HORMONE AND CALCITONIN

The **parathyroid glands** in man are usually 4 in number, oval, 5 to 6 mm in length, and situated upon or imbedded in the dorsal surface of the thyroid gland. They develop and function independently of thyroid tissue. For a number of years, after their discovery by Sandstrom in 1880, the parathyroids were considered to be remnants of embryonic thyroid tissue.

The parathyroid glands exert a hormonal control over calcium metabolism. Acute deficiency results in tetany when the level of serum calcium falls from normal (10 to 11 mg/dl) to around 6 to 7 mg/dl. Fibrillary muscular twitching progresses to the convulsive state, culminating in death by tetanic spasm of the larynx and muscles of respiration.

Parathyroid hyperfunction produces a condition known as Recklinghausen's disease of bone (osteitis fibrosa cystica), characterized by bone pain, marked elevation of serum calcium with a fall in serum phosphate, and cystic rarefaction of bones with spontaneous fracture and deformity. The calcium removed from bone is excreted in the urine. A similar picture may result from overdose with extracts of parathyroid

gland. In either case, renal stones and calcification of soft tissues occur.

The **parathyroid hormone (parathyrin)** is a straight-chain polypeptide containing 83 amino acid residues and has a molecular weight of approximately 9500. A portion of the molecule that contains only 35 amino acid residues can elicit the significant physiologic activity of the hormone. The essential 35-amino-acid subunit of human parathyroid hormone differs in 5 or 6 of its amino acid residues from the animal parathyroid hormones that are available through the meat-packing industry. Immunologic recognition of this factor may contribute to the high incidence of tolerance noted in therapy. The hormone has a hypercalcemic action. Its principal effect involves bone resorption and calcium release, but it also promotes absorption of calcium from the gut and renal tubules.

Parathyroid hormone is inactivated in the intestinal tract, but it has been used parenterally in medicine for blood-calcium maintenance in cases of parathyroid tetany. Following injection, the blood calcium level rises in about 4 hours, reaching a maximum in about 16 hours and returning to the original level after 24 to 36 hours. Repeated or prolonged administration may establish a complete tolerance with abolition of therapeutic effect. To avoid this, dihydrotachysterol may frequently be substituted; this preparation may be given orally. Adequate intake of calcium, phosphate, and vitamin D must be assured. Some authorities question the justification for using parathyroid hormone for therapeutic purposes, and its future use may be primarily in the diagnostic area.

Parathyroid injection is a sterile solution in water for injection of the water-soluble principle of the parathyroid glands that has the property of increasing the calcium content of the blood. This preparation is biologically assayed and standardized so that 1 ml possesses a potency of not less than 100 USP parathyroid units, each unit representing 1/100 of the amount required to raise the calcium content of 100 ml of the blood serum of normal dogs 1 mg within 16 to 18 hours after administration.

Calcitonin is produced by the parafollicular or C cells of the thyroid gland, and recent clarification of its function has significantly increased our comprehension of calcium metabolism. This hormone exerts a hypocalcemic effect by suppressing bone resorption and by inhibiting tubular reabsorption of calcium in the kidneys. Thus, calcitonin and parathyroid hormone have counterbalancing actions similar to the situations observed with glucagon-insulin and other control mediators of biologic processes.

Calcitonin is a polypeptide containing 32 amino acid residues. The individual amino acid composition of calcitonin from different animal sources varies considerably. The key molecular features for biologic activity appear to include a prolinamide moiety at the carboxyl terminal end of the peptide and a cyclic subunit containing 6 amino acid residues, including the ring closing cystine, at the amino terminal end of the molecule. Reduction of the disulfide linkage causes a loss of activity.

Calcitonin can be used to treat Paget's disease (osteitis deformans) and postmenopausal osteoporosis and to control hypercalcemia secondary to other osteolytic conditions. The preparation used in therapy is a synthetic salmon calcitonin. Salmon calcitonin elicits 20-fold or more activity on a molar basis than human or porcine calcitonins; the apparent increased potency may be caused, in part, by a greater affinity for receptor sites and thus a slower rate of degradation and a longer duration of action. The usual maintenance dose for Paget's disease is 50 to 100 units subcutaneously per day or every other day. The usual dosage regimen in osteoporosis is 100 units subcutaneously or intramuscularly per day together with supplemental calcium and adequate vitamin D intake. Therapy to control secondary hypercalcemia usually starts with 4 units per kg of

body weight every 12 hours, and the dose may be increased, if necessary, up to 8 units per kg every 6 hours. Intramuscular injection and multiple sites are preferred when higher doses are required.

PRESCRIPTION PRODUCT. Calcimar®.

GASTROINTESTINAL HORMONES

The intestinal mucosa secretes such peptide hormones as cholecystokinin, gastrin, and secretin. These hormones facilitate digestion by stimulating the release by the gastrointestinal tract or the pancreas of various enzymes and other exocrine substances. There is no therapeutic indication for these hormones, but cholecystokinin (CCK®) and secretin (Secretin-Kabi® and Secretin-Boots®) from porcine duodenal mucosa are used in the diagnosis of pancreatic disorders. Cholecystokinin is also used as a diagnostic aid in cholecystography and cholangiography.

Cholecystokinin contains 33 amino acid residues in a linear chain, but the carboxylic acid-terminal octapeptide is fully active. Cholecystokinin stimulates the secretion of pancreatic digestive enzymes, the flow of bile, and the contraction of the gallbladder. Ceruletide (Tymtran®) and sincalide (Kinevac®) are synthetic deca- and octapeptides, respectively, which act similarly to cholecystokinin; they are used diagnostically.

Secretin normally increases the bicarbonate content and volume of secretion from the pancreas. It is a linear polypeptide containing 27 amino acid residues; the amino acid sequence has a noticeable similarity to that of glucagon.

Pentagastrin (Peptavlon®), a synthetic pentapeptide with effects similar to those of natural gastrin, is also used for diagnostic purposes. Pentagastrin increases gastrointestinal motility and stimulates the secretion of gastric acid, pepsin, and intrinsic factor; it is used to test gastric secretory function.

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