INTRODUCTION

Principles of endocrinology and general mechanisms of hormone action

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The action of the endocrine system is required for: 1) the adequate neurological and somatic development and growth of the organism; 2) the ability of the organism to interact with the environment (stress); 3) the regulation of metabolic rate and many other functions that provide the homeostasis within each tissue of the organism; and 4) sex differentiation and reproduction of the organism.

Homeostasis is the property of a system in which a variable, such as the molecular concentration in a solution, is actively regulated to remain nearly constant. Examples of homeostasis include the regulation of body temperature, the pH of extracellular fluid, or the concentrations of ions (*e.g.*, sodium, potassium and calcium), as well as that of glucose in the blood, despite changes in the environment, diet, or level of physical activity. Each of these variables is controlled by a separate regulator or homeostatic mechanism involving hormones, which, together, maintain life. The concept was described by the French physiologist Claude Bernard in 1865 and the word homeostasis was coined by Walter Bradford Cannon in 1926.[1]

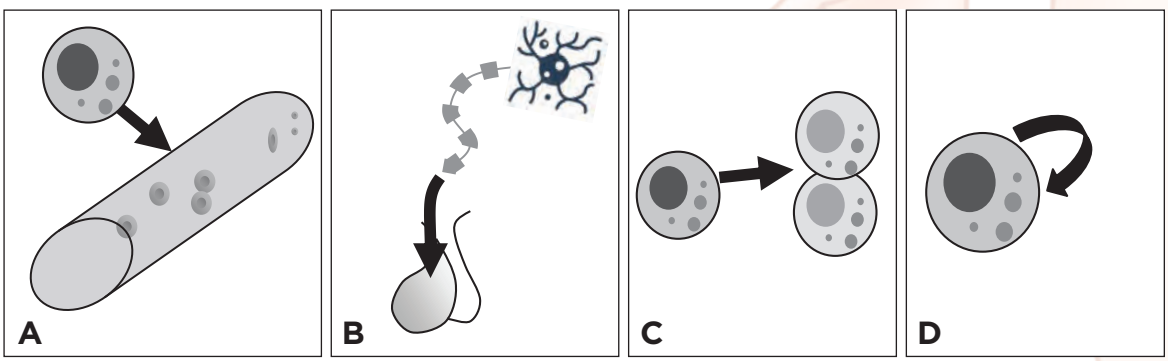
In the classic understanding, hormone (from the Greek verb ὁρμῶν, meaning "to set in motion," a word expressing the dynamic properties of hormones and the ability to elicit physiological responses at the target tissue level) is considered any member of a class of signaling molecules in multicellular organisms, that are transported

in the blood to distant organs to regulate physiological events or behavior. The substances that can be nowadays considered hormones are: proteins/peptides (*e.g.*, insulin or pituitary tropins, such as antidiuretic hormone, thyroid-stimulating hormone, or prolactin), steroids (*e.g.*, sex hormones, estrogens and androgens, or glucocorticoids and mineralocorticoids), vitamin (vitamin D and retinoids) or amino acid derivatives (*e.g.*, epinephrine, dopamine or thyroid hormone), cytokines (*e.g.*, leptin or adiponectin), growth and differentiation factors (*e.g.*, bone morphogenetic proteins, inhibins, anti-Müllerian hormone), eicosanoids (*e.g.*, prostaglandins and thromboxanes), as well as ions (*e.g.*, calcium) and gases (*e.g.*, nitrous oxide). Consequently, beyond the classic endocrine actions, hormones can be secreted by neurons (*e.g.*, neuroendocrine actions of hypothalamic neurons), or can regulate the activity of a close cell (*e.g.*, paracrine actions of growth factors secreted by oocytes or granulosa cells supporting the fundamental crosstalk required for ovarian folliculogenesis) or that of the same secretory cell (*e.g.*, autocrine feedback actions typically aimed to limit the activity of a cell type within a tissue)[2] (Figure 1.1.1).

Hormones exert their activity by binding to specific receptors in the target cell, resulting in a change of cell function. When a hormone binds the receptor, it results in the activation of a signal transduction pathway that can activate or inhibit gene transcription, resulting in increased or diminished expression of target proteins. Hormones can also act in rapid, non-genomic pathways that can be synergistic with genomic effects.

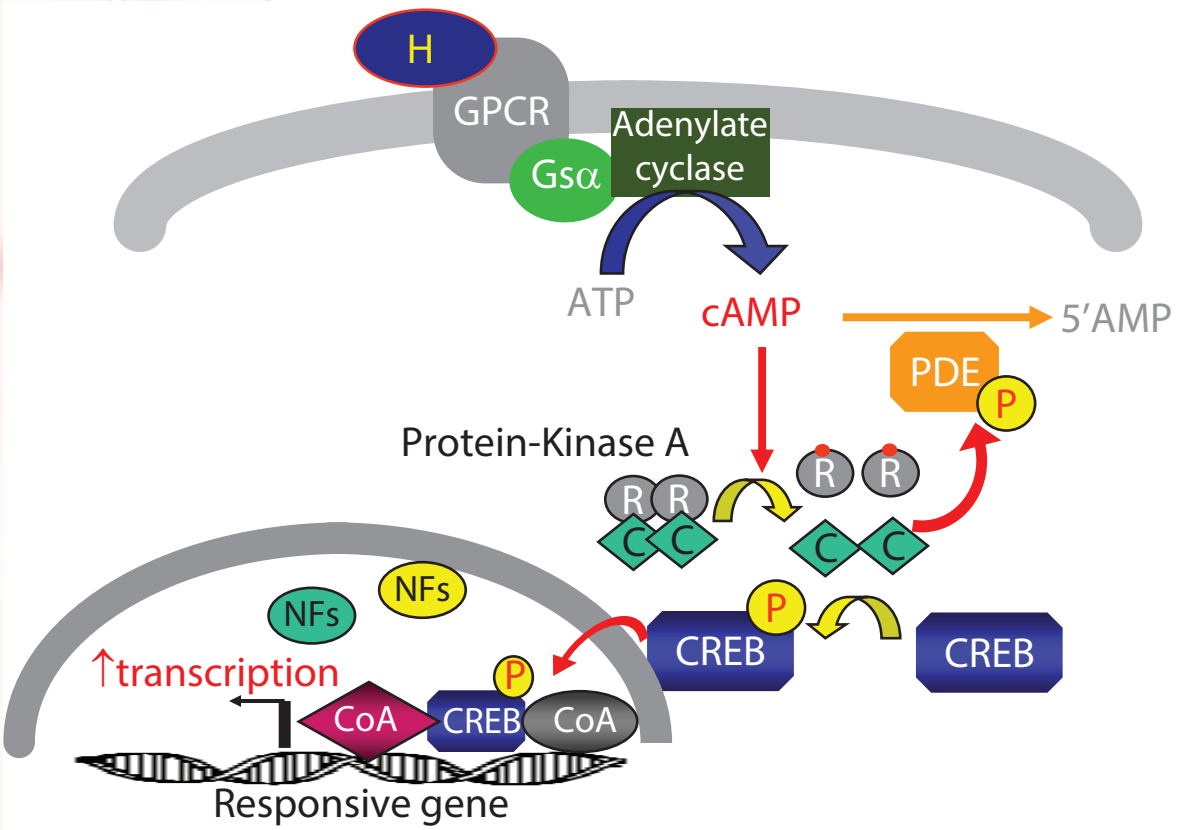
Water-soluble hormones (such as peptides and amines) generally act by binding receptors expressed on the surface of target cells (*e.g.*, G protein-coupled

Figure 1.1.1. Schematic illustration of the four different types of hormone action. A) The classic "endocrine" action is the one supported by hormones that are released in the blood stream and directed to act on distant tissues. B) Neurons secrete various substances that have a neuroendocrine action: typically, hypothalamic neurons regulate the pituitary activity by releasing various stimulatory or inhibitory factors in the portal system. C) Several cells secrete substances devoted to the regulation of the activity of proximal cells within the same organ/tissue: typically, growth and differentiation factors provide the crosstalk between oocytes and granulosa cells that is required for an adequate ovarian folliculogenesis (paracrine action). D) Several cells produce substances that can provide auto-regulatory feedback aimed to maintain cellular activities within certain physiological limits (autocrine action).



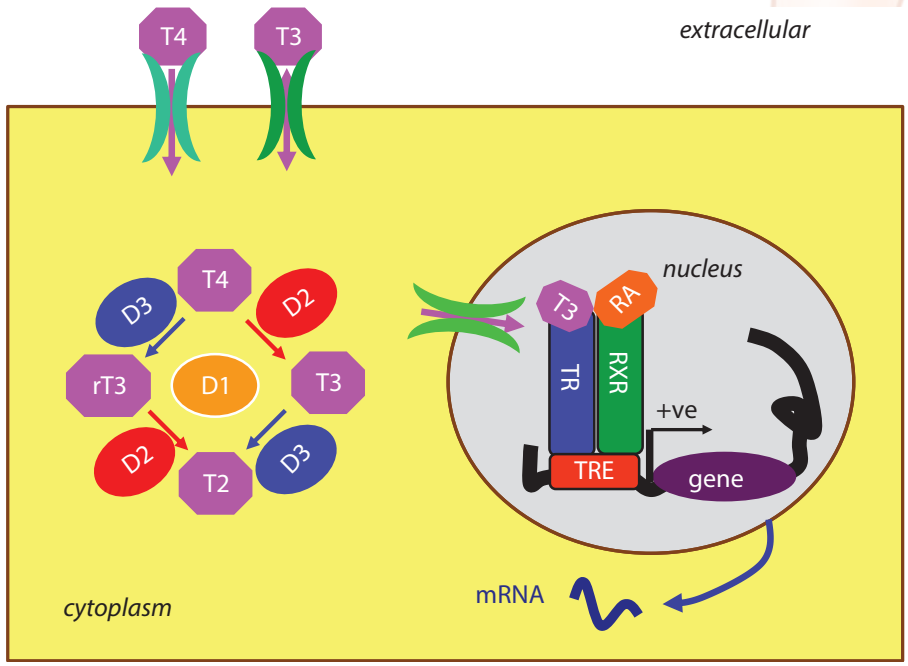
receptors, GPCRs, or tyrosine kinase receptors, TKRs) that transduce the hormonal signals inside the cells *via* second messengers (*e.g.*, cyclic AMP or Ca^{2+} or inositol-triphosphate, IP3, or protein phosphorylation cascade) (Figure 1.1.2). The lipid soluble hormones (such as steroids or thyroid hormone) typically circulate in blood bound to albumin or specific globulins of liver origin (*e.g.*, sex hormone binding globulin [SHBG]; cortisol binding protein or transcortin [CBG]; thyroid hormone binding globulin [TBG]) and their free fraction pass through the plasma membranes of target cells (both cytoplasmic and nuclear) to act within

Figure 1.1.2. Several water-soluble hormones interact with specific receptors on the cell membrane and elicit cellular responses through the activation of specific intracellular pathways. The scheme illustrates elements of the cAMP pathway that can be activated by hormone (H) binding to specific G-protein coupled receptors (GPCRs), characterized by seven transmembrane domains that can interact and activate specific G proteins. The recruitment of Gsα promotes the activation of the adenylate cyclase able to synthetize cyclic AMP (cAMP) from ATP. cAMP binds the regulatory (R) subunits of the protein-kinase A, and this event results in the release of the catalytic (C) subunits that phosphorylate protein targets like CREB. Phosphorylated CREB enters the nucleus where it can interact with several nuclear factors (NFs) and co-activators (CoA) and binds the regulatory regions of the cAMP-responsive genes eventually leading to the enhancement of their transcription. This positive signal can be switched off at several levels, including the activation by phospho-CREB of cAMP-phosphodiesterases (PDEs) that degrade the cAMP to 5′AMP.



their nuclei by binding nuclear receptors that act as ligand-dependent transcription factors (*e.g.*, glucocorticoid receptor, androgen receptor or thyroid hormone receptors). This transmembrane passage requires, at least for thyroid hormones, the action of specialized transporters, *e.g.*, MCT8 in the central nervous[2] (Figure 1.1.3).

Figure 1.1.3. Schematic representation of thyroid hormone action on positively regulated target genes. Both thyroxine (T4) and tri-iodothyronine (T3) can enter the target cells thanks to the activity of specific membrane transporters, like the monocarboxylate transporter 8 (MCT8). T3 is the active hormone and can directly enter the nucleus and induce the transcription of target genes. However, in condition of normal iodide intake with the diet T4 is the main thyroid product, and T4 is a pro-hormone that can either be activated or inactivated in target tissues upon specific tissue requirements. The intracellular deiodinases (D1-3) are enzymes devoted to the removal of specific iodine residues within T4. D2 is specifically designed to remove the iodine bound in position 5′ of the external aromatic ring of T4 and to activate thyroid hormone signal through the generation of T3. D3 is instead specifically designed to remove the iodine bound in position 5 of the internal aromatic ring of T4 and to reduce thyroid hormone signal by generating the inactive compound named reverse T3 (rT3). The D2 is expressed in several tissues, including the central nervous system, hypothalamus, pituitary, muscle, and brown adipose tissue. The D1 is mainly expressed in kidney and liver and can remove all iodide residues with sufficient affinity. The D3 can be expressed in several tissues and is increased in disease condition or starvation when the excitatory effect of T3 would be deleterious for the organism; these conditions are called non-thyroid illness or low-T3 syndromes. The active hormone T3 can bind with elevated affinity the ligand binding domain of nuclear T3-receptors (TRs), that act as ligand-dependent transcription factors by biding specific T3-responsive elements (TREs) in the regulatory regions of target genes. Typically, TRs hetero-dimerize on these TREs with the RXRs, the receptors for retinoic acid (RA) and this action amplifies the transcription of target genes.



In vertebrates, endocrine glands are specialized organs that secrete hormones into the blood stream. Endocrine glands are prevalently constituted by endocrine cells with variable embryological derivation:

- thyroid, parathyroids, pancreas islets and anterior pituitary derive from endoderm;
- adrenal medulla, neuroendocrine cells (*e.g.*, C cells in the thyroid or neuroendocrine cells of the gut), as well as posterior pituitary derive from neuroectoderm;
- adrenal cortex and gonads (testes and ovaries) have a mesoderm origin.

However, several other organs are known to have endocrine functions. Gut hormones were among the firstly discovered signaling substances that are linked to clinical syndromes. These hormones are secreted by enterochromaffin cells that constitute the diffuse hormonal system. In fact, the first discovered hormone was secretin, when Bayliss and Starling instilled acid into the denervated duodenum of a dog and observed the flow of pancreatic secretions and said this must be due to a hormone. The proliferation of these enterochromaffin cells is responsible for many of the neuroendocrine tumors. For instance, gastrin was identified as a potent gastric acid secretagogue in 1905, and Zollinger and Ellison in 1955 recognized it as the principal culprit for the Zollinger Ellison Syndrome due to gastric acid overproduction. More recently, Verner and Morrison discovered VIP in 1972 which identified the culprit behind the watery diarrhea, hypokalemia acidosis syndrome (WDHHA). Nearly all tissues are currently known to secrete bioactive substances with hormonal activity, *e.g.*, adipocytes secrete leptin, a signal of satiety to the hypothalamus; heart is secreting atrial natriuretic peptide and B-type natriuretic peptide (BNP) upon atrial or ventricular stretch, respectively, to promote diuresis; or kidneys secrete erythropoietin, the hormone promoting erythrocyte maturation.[3, 4]

Most of the hormones are completely active when released into the bloodstream (as is the case for prolactin or growth hormone), while others are prohormones that must be activated in specific cells through a series of activation steps that are commonly highly regulated. For instance:

- β cells of the pancreatic islets synthetize the inactive pro-insulin protein that is cleaved by specialized enzymes, called pro-convertases, in the secretory granules into the active hormone insulin and the inactive C-peptide;
- adrenocorticotropic hormone (ACTH), as well as melanocyte-stimulating hormones (α or γMSH) and endorphins, derive from the variable pro-convertase cleavage of the pro-hormone called proopiomelanocortin (POMC);
- thyroxine is the main inactive product of the thyroid gland and must be activated through the conversion to tri-iodothyronine by the tissue deiodinases (Figure 1.1.3);
- vitamin D and retinoic acid can be directly derived from diet but can also be generated and/or activated by endogenous synthetic pathways in different tissues.

References

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The feedback mechanism

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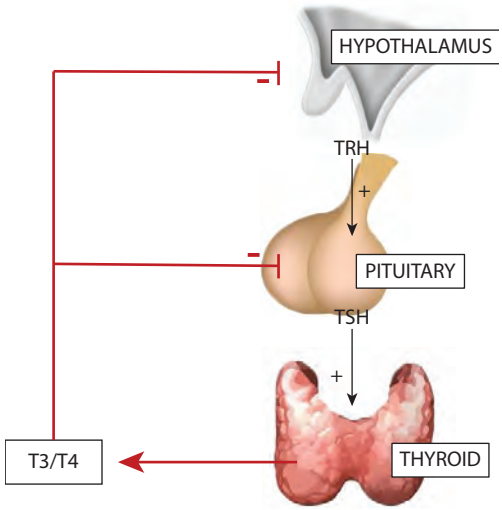
The action of the endocrine system is typically regulated by the negative feedback mechanism which is aimed to maintain the hormonal activities within the physiological range. Probably every hormone secretion has a particular set point that is controlled by downregulating the stimulatory pathways (when the set point is exceeded) and upregulating stimulatory pathways (when the hormone action falls below the desired set point). These regulatory loops are well illustrated by the major endocrine axes, constituted by hypothalamic nuclei, the anterior pituitary gland, and the peripheral glands (adrenals, gonads or thyroid). These axes contain both positive (*e.g.*, hypothalamic TRH stimulates thyrotropes to secrete thyroid-stimulating hormone [TSH], and TSH stimulates the thyroid gland to produce T4/T3) and negative components (*e.g.*, T4/T3 suppresses TRH and TSH). This negative component is the main actor in maintaining the activity of the endocrine axes within the physiological requirements, despite endogenous and environmental challenges (Figure 1.2.1).[1]

The feedback mechanism is regulating other endocrine systems that do not involve the hypothalamus and pituitary. As thyroid hormones suppress TSH, the elevation of blood calcium levels feeds back to inhibit PTH secretion from parathyroids, chloride acid inhibits gastrin secretion, while leptin elevation suppresses appetite and elevation of glucose stimulates insulin and inhibits glucagon secretion.[2]

The feedback mechanisms have several implications for the clinical practice. They provide useful insight into endocrine testing paradigms. Primary hypothyroidism is characterized by elevated TSH, an appropriate physiologic response to deficient thyroid hormone levels. Since TSH elevation above the reference range occurs already when thyroid hormone deficiency is mild, the TSH determination is considered the most accurate parameter to screen thyroid function. In addition, glucocorticoid

Figure 1.2.1. Schematic illustration of the hypothalamic-pituitary-thyroid axis. Hypothalamic TRH neurons release TRH into the pituitary portal system through which the neurohormone reaches pituitary thyrotropes to stimulate TSH subunit synthesis and TSH secretion. TSH reaches the thyroid through the circulation and stimulates thyroid hormone synthesis and secretion. In normal conditions, T4 is the main product that is peripherally converted into T3, the active hormone at the peripheral tissue level, by specific enzymes, the deiodinases. When T4 production exceeds the physiologic requirements, TRH and TSH are suppressed by the thyroid hormone negative feedback mechanism. In contrast, when T4 production falls below the pituitary set point, the negative feedback declines to augment pituitary TSH secretion and stimulation of the thyroid gland. This mechanism maintains T4/T3 levels within the physiologic requirements.



suppression of the CRH/ACTH axis (dexamethasone suppression test) is used to screen patients for hypercortisolism and the lack of circulating cortisol suppression below the expected threshold indicates Cushing Syndrome. In these cases, a suppressed ACTH indicates ACTH-independent Cushing, while normal/elevated ACTH levels indicate ACTH-dependent Cushing. Feedback mechanism has been exploited also for other clinical applications. The negative feedback of estrogens on GnRH pulsatility and gonadotropin release offers the fundamental basis for the efficacy of hormonal contraceptives. Combined hormonal contraceptives were in fact developed to prevent ovulation by suppressing the release of gonadotropins, because they maintain estrogen levels slightly over the feedback set point at hypothalamic-pituitary levels.[3]

References

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