Endocrinology examination

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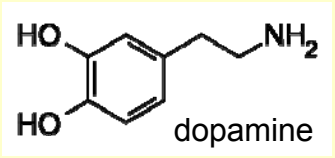
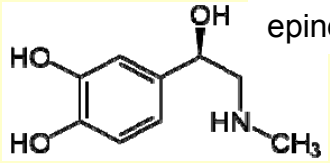
Hormone biochemistry

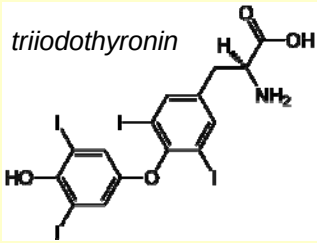
- **Amino acid derivatives**
  - catecholamins (dopamine,epinephrine), thyroid hormones



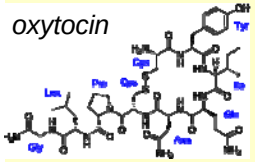
dopamine



epinephrine

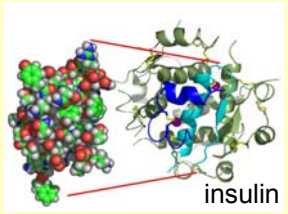


- **Small peptides**

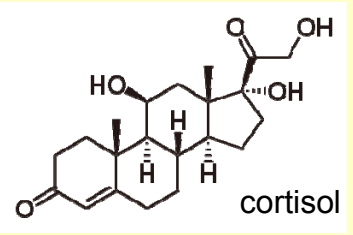


oxytocin, vasopressin (ADH), somatostatine, gonadotropin-releasing hormone (GnRH), thyrotropin-releasing hormone (TRH)

- **Proteins**
  - insulin, LH, PTH

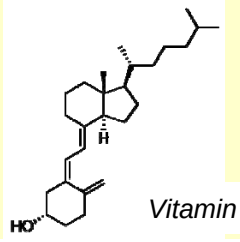

insulin

- **Steroid hormones** (synthesized from cholesterol-based precursors)
  - cortisol, estrogen



cortisol

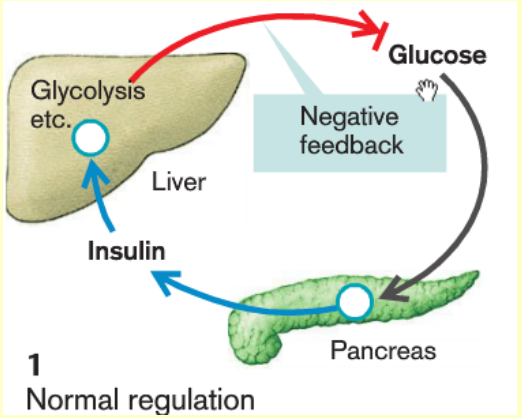
- **Vitamine derivatives**
  - Retinoids (Vitamin A), vitamin D



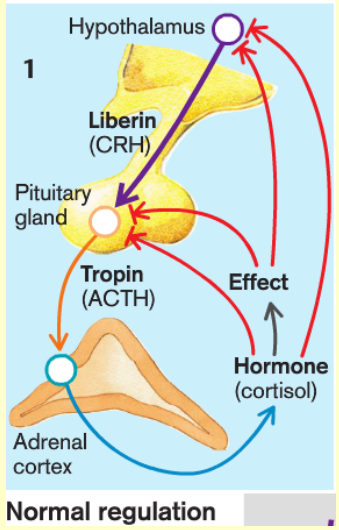
Vitamin D

Hormones are parts of regulatory circuits

- Pituitary independent (simple circuits)



- Hypothalamus – pituitary regulated hormones
  - Hypothalamus (liberins/releasing hormones)
  - Pituitary (Tropins)



Color atlas of pathophysiology (2010), Silbernagel, Lang

Causes of hormone excess or hormone receptor hyperfunction

- Hormone is secreted by tumor cells
- The receptor for hormone is constitutively activated
- A non-physiological stimulator of the receptor (e.g. antibody) or **receptor crosstalk**
- The post-receptor signal transduction pathway contain an abnormal protein that signals continued receptor occupancy
- Hormone is ingested accidentally, deliberately or therapeutically

Hormone receptor receptor cross-talk

Hormones and Receptor Families

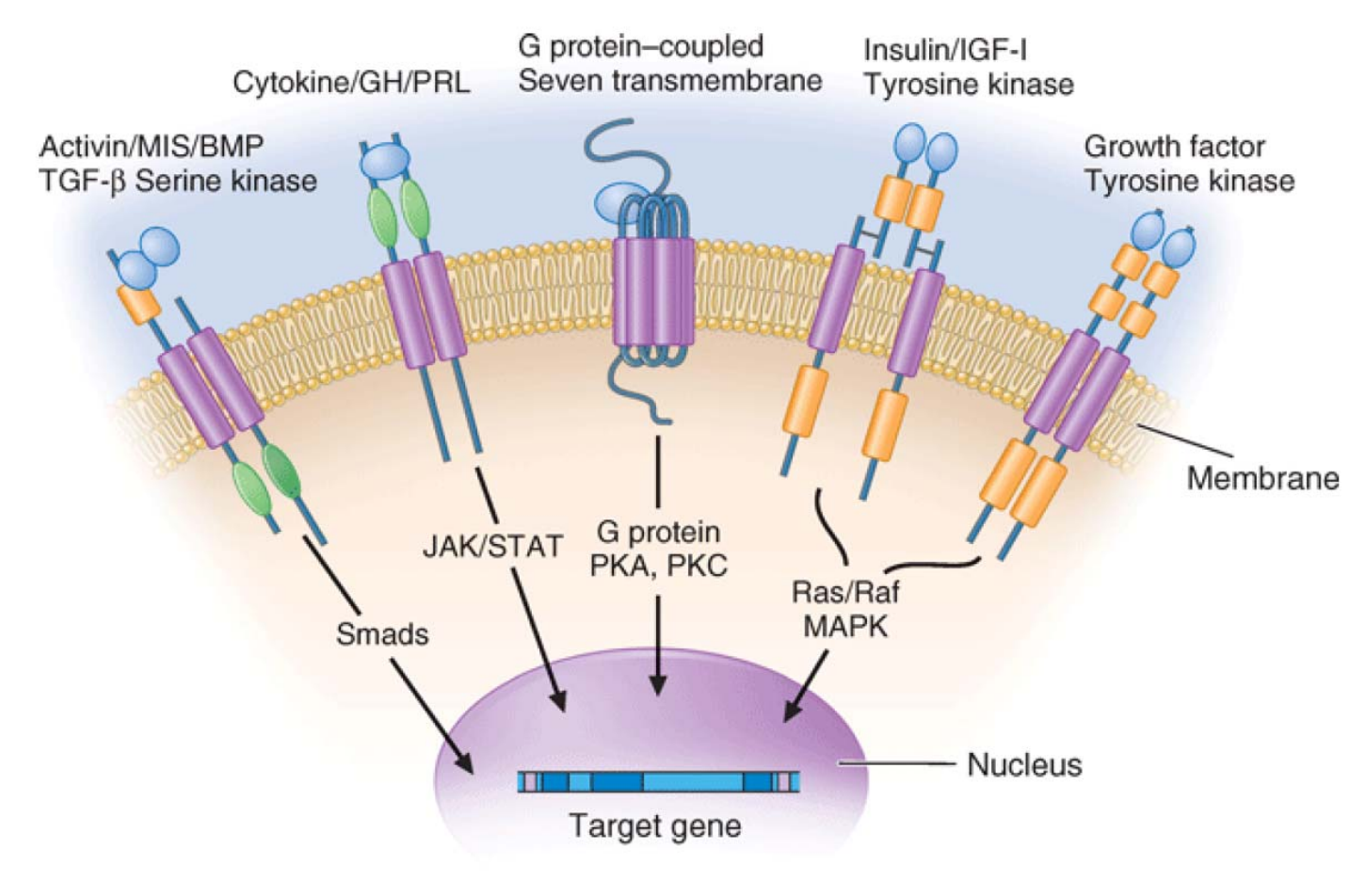
Membrane Receptor Families and Signaling Pathways

- G Protein–Coupled Seven-Transmembrane (GPCR)
- Receptor Tyrosine Kinase
- Cytokine Receptor–Linked Kinase
- Serine Kinase

Nuclear receptor family

- type 1 (steroid) receptors
  - androgen, estrogen, mineralocorticoid, glucocorticoid, progesterone
- type 2 receptors
  - thyroid hormone, vitamin D, retinoic acid, or lipid derivatives
- Orphan receptors

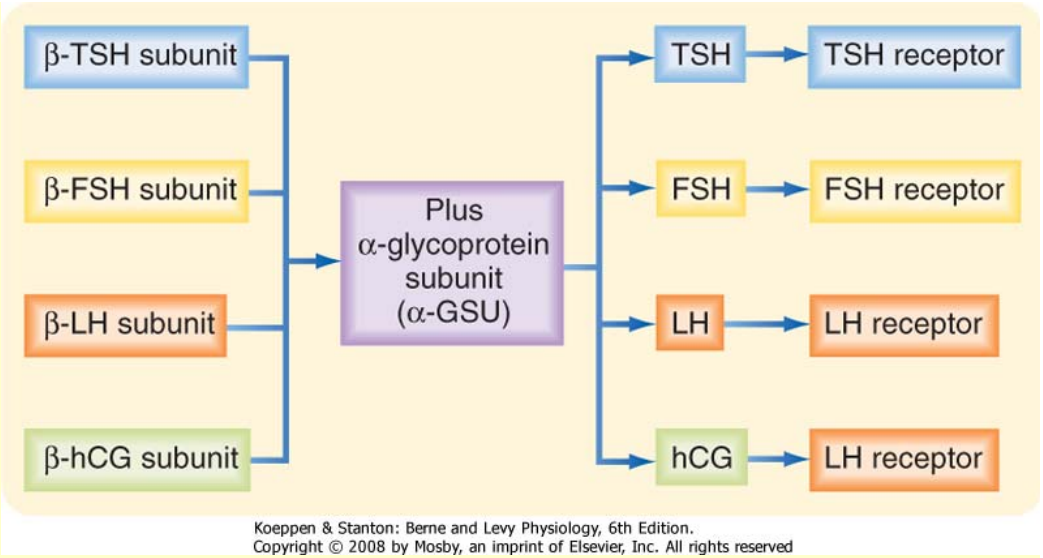
Membrane Receptor Families and Signaling Pathways



Membrane receptor signaling. MAPK, mitogen-activated protein kinase; PKA, -C, protein kinase A, C;

Source: Longo DL, Fauci AS, Kasper DL, Hauser SL, Jameson JL, Loscalzo J: *Harrison's Principles of Internal Medicine, 18th Edition*: www.accessmedicine.com

G Protein–Coupled Seven-Transmembrane (GPCR)



- Receptors for glycoprotein hormone family consisting of thyroid-stimulating hormone (TSH), follicle-stimulating hormone (FSH), LH, and human chorionic gonadotropin (hCG),
- Evolved for each of the heterodimer (common α subunit) hormones
  - Minimal overlap of hormone binding
    - e.g. TSH binds with high specificity to the TSH receptor but interacts minimally with the LH or the FSH receptor
  - Hormone cross-reactivity with other receptors
    - e.g. Very high levels of hCG during pregnancy stimulate the TSH receptor and increase thyroid hormone levels, resulting in a compensatory decrease in TSH

G Protein–Coupled Seven-Transmembrane (GPCR)

- Receptor cross-talk
  - PTH and parathyroid hormone–related peptide (PTHrP)
  - PTH is produced by the parathyroid glands
  - PTHrP is expressed at high levels during development and by a variety of tumors
- Both hormones bind to a single PTH receptor that is expressed in bone and kidney.
- Excessive production of either hormone
  - Hypercalcemia and hypophosphatemia
- Diferential diagnosis of hypercalcemia:
  - Hyperparathyroidism
  - Malignancy producing PTHrP

Receptor tyrosin kinase

- Insulin/IGF family
  - Insulin and insulin-like growth factor I (IGF-I) and IGF-II
  - structural similarities that are most apparent when precursor forms of the proteins are compared.
  - Moderate cross-talk among the members of the family
  - High concentrations of an IGF-II precursor produced by certain tumors (e.g., sarcomas) can cause hypoglycemia, partly because of binding to insulin and IGF-I receptors
  - High concentrations of insulin also bind to the IGF-I receptor, perhaps accounting for some of the clinical manifestations seen in severe insulin resistance.

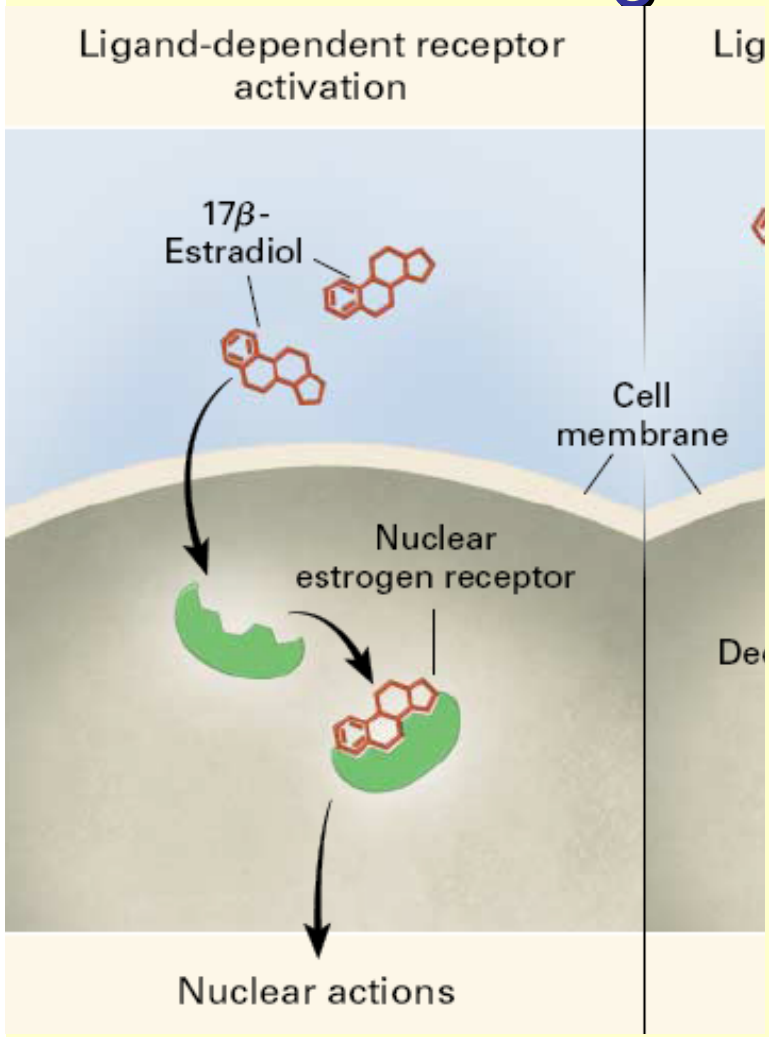
Nuclear receptor family

- Nearly 100 members
- The hormone-binding domains are variable and higly specific

Nuclear receptor family exceptions

- **Glucocorticoid and mineralocorticoid receptors**
  - Mineralocorticoid receptor also binds glucocorticoids with high affinity
  - Enzyme (11 -hydroxysteroid dehydrogenase) in renal tubular cells inactivates glucocorticoids, allowing selective responses to mineralocorticoids
  - Very high glucocorticoid concentrations (e.g. Cushing's syndrome), the glucocorticoid degradation pathway becomes saturated, allowing excessive cortisol levels to exert mineralocorticoid effects (sodium retention, potassium wasting).
- **Estrogen receptor**
  - can bind an array of compounds, some of which have little apparent structural similarity to the high-affinity ligand estradiol.
  - susceptible to activation by "environmental estrogens" such as resveratrol, octylphenol, and many other aromatic hydrocarbons.
  - Clinically useful antagonists (e.g., tamoxifen) and selective estrogen response modulators (SERMs) such as raloxifene.

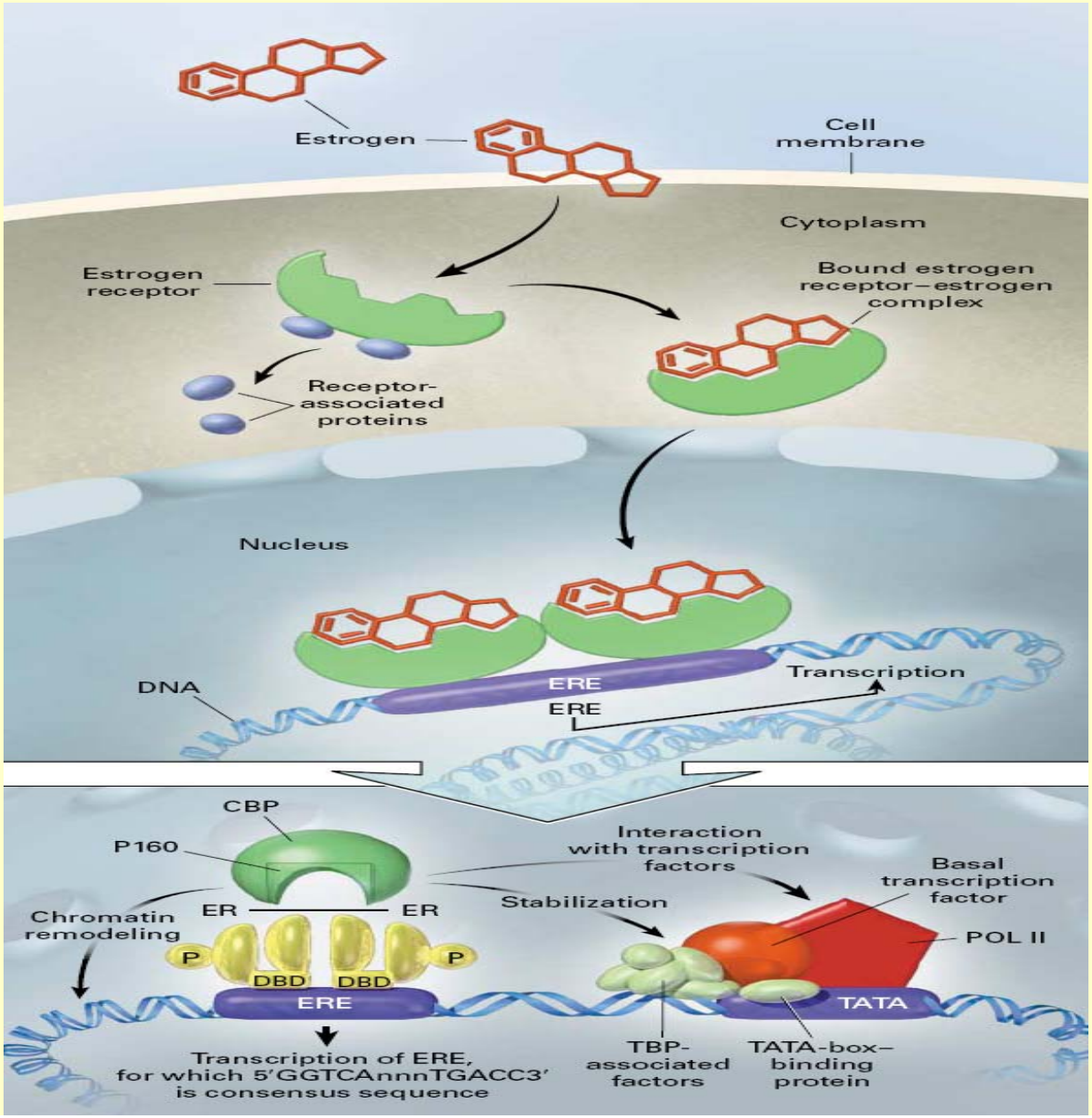
Ligand-Dependent and Ligand-Independent Estrogen-Receptor Activation



Activated by estrogen

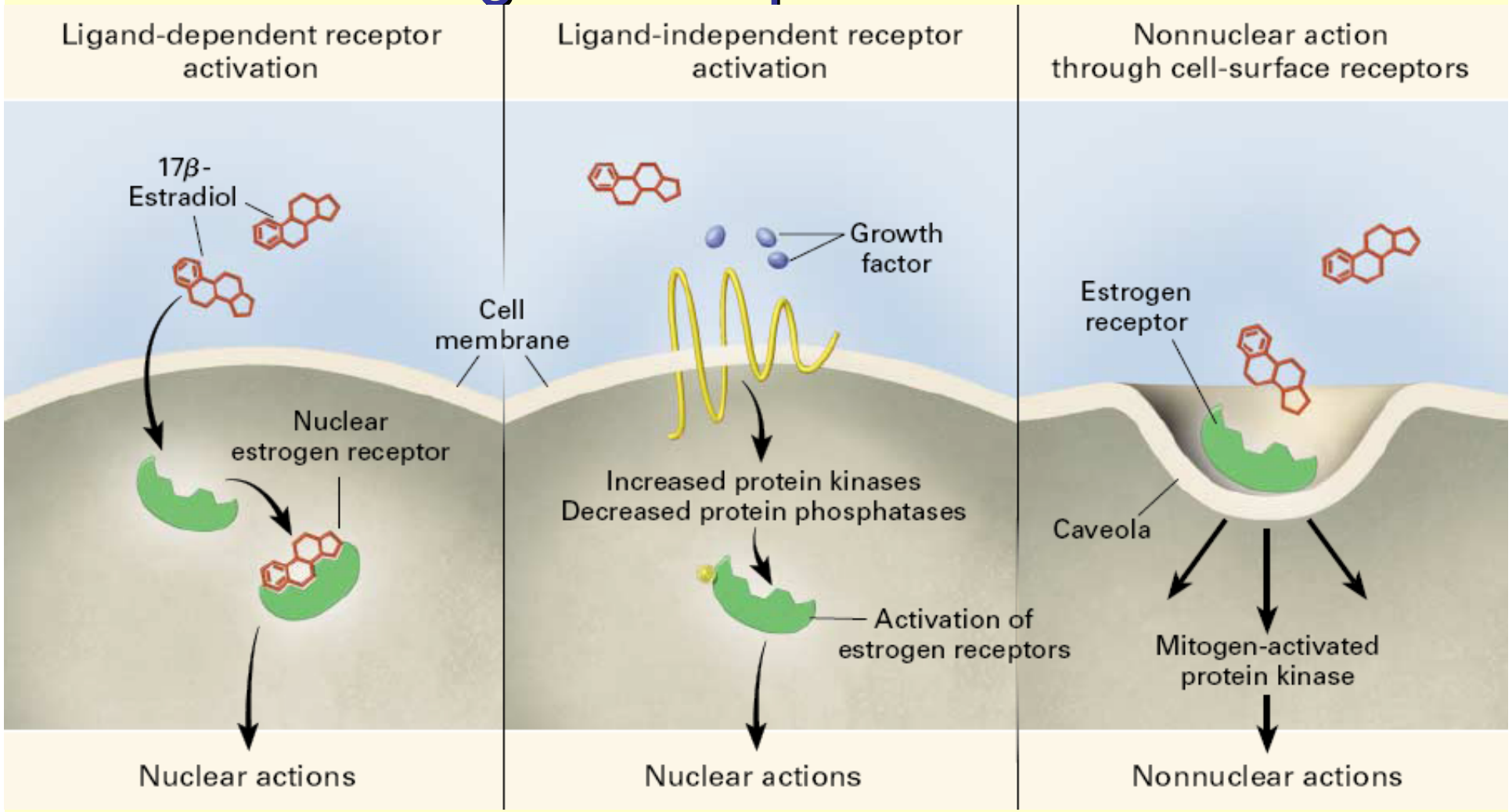
Classic Pathway

Classic Pathway of Estrogen Signal Transduction



1. estrogen molecule binds to an estrogen receptor
2. receptor dissociates from its cytoplasmic chaperones
3. hormone–receptor complex then moves to the nucleus
4. hormone–receptor complex binds to DNA and initiates transcription
5. estrogen receptors interact with several proteins - stabilization of the preinitiation complex, chromatin remodeling, and interaction with other transcription factors

Ligand-Dependent and Ligand-Independent Estrogen-Receptor Activation



Activated by estrogen
Classic Pathway

Activated independently of estrogen (phosphorylation of the receptor molecule)

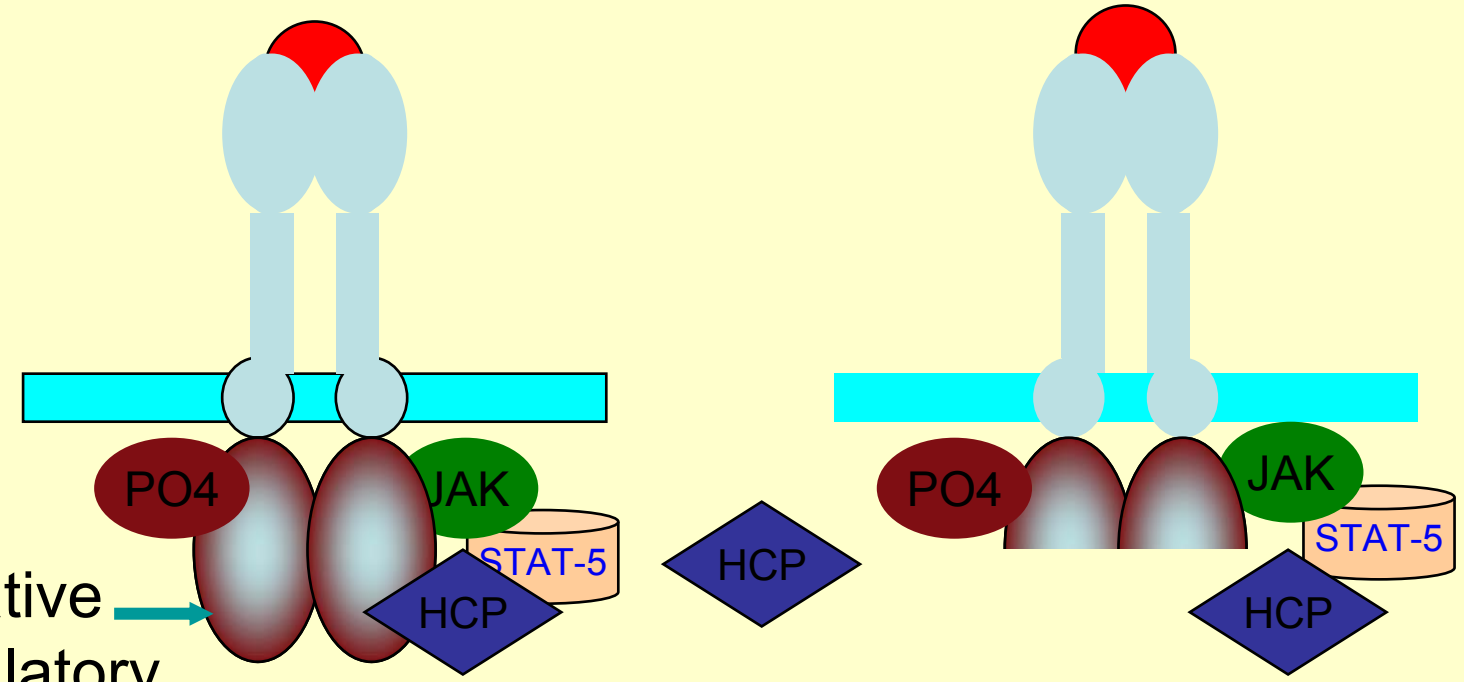
Nonnuclear estrogen-signaling pathway (in cell-membrane invaginations called caveolae)

Aberrant activation/inactivation of hormone receptor and signalling pathway

Polycythemia in Olympic game winner (5 starts 4 medals): Eero Mantyranta

Hb 23.1
Hct 68%

Erythropoietin Receptor



Negative Regulatory Domain

Truncation

The receptor is stuck in the “on” position

Causes of hormone deficiency

- A lack of the hormone
  - e.g. genetic deletion, damage to the endocrine gland, lack of a synthetic enzyme
- Production of a biologically inactive hormone
  - e.g. genetic mutation
- Hormone receptor or the down-stream signaling pathways is structurally abnormal and inactive
  - hormone resistance

Endocrinological examination

- Clinical symptoms
- Laboratory examinations
  - hormone concentration or production (e.g. 24 h collection)
  - measurement of regulated molecule (e.g. glucose)
  - tests: stimulatory or inhibitory
- Immaging methods

Clinical symptoms

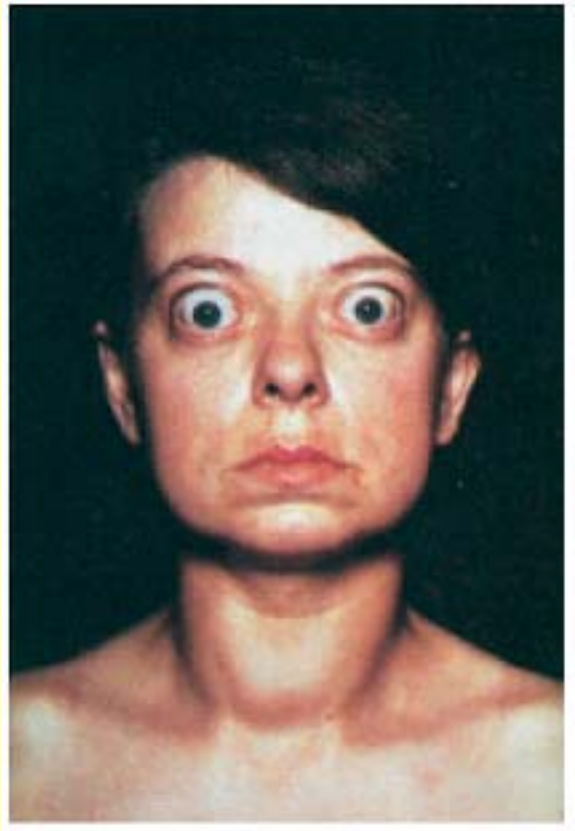
- Clinical manifestation of hormone excess or deficiency e.g. :
  - Acromegaly
  - Hyperthyroidsm
  - Cushing syndrome

Acromegaly

- Enlargement of the hands, feet, nose, lips and ears, and a general thickening of the skin
- Enlargement of internal organs (heart, kidneys, vocal cords: thick, deep voice and slowing of speech)
- Generalized expansion of the skull at the fontanelle
- Pronounced brow protrusion, often with ocular distension
- Pronounced lower jaw protrusion with attendant macroglossia and teeth gapping

Graves disease = Basedow's disease (Hyperthyroidsm)

- Mass in the neck
- Exofthalmus
- "orange-peel" skin

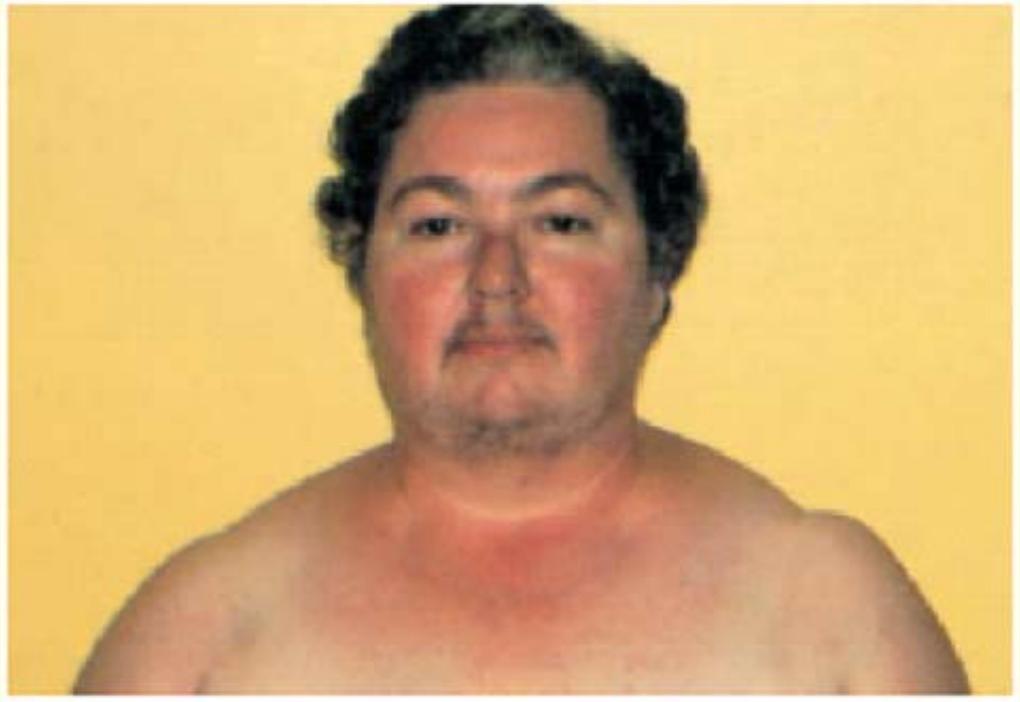


(Rubin E., Farber J.L. [1999]. *Pathology* [3rd ed., p. 1193]. Philadelphia: Lippincott-Raven)

Cushing syndrome

- Typical clinical signs:
  - central fat redistribution
  - Striae
  - proximal muscular weakness
- Common clinical signs (various diseases)
  - obesity
  - plethora (excess of RBC or bodily humours)
  - hypertension
  - glucose intolerance

Cushing syndrome: woman with pituitary adenoma



- Moon face
- Buffalo hump
- Increase facial hair
- Thinning of the scalp hair

(Rubin E., Farber J.L. [1999]. *Pathology* [3rd ed., p. 1193]. Philadelphia: Lippincott-Raven)

Laboratory examinations

- Measurement of hormones in blood (serum, plasma)
- Measurement of hormones or its metabolites in urine
- Measurement of hormones in saliva
- Function tests of the feedback loops

- RIA = Radioimmunoassay (developed by Rosalyn Yalow and Solomon Aaron Berson in the 1950s; Yalow received in 1977 Nobel Prize in Medicine for the development of the RIA for insulin)
- IRMA = immunoradiometric assay
- ELISA = Enzyme-linked immunoassay (sandwich-ELISA)

Measurement of hormones in blood

- Hormones with long half life
  - e.g. thyroxin, triiodthyronin
- Hormones with short half life
  - daily rhythm
    - e.g. cortisol 5 fold increase from midnight to dawn

ACTH and cortisol during the day

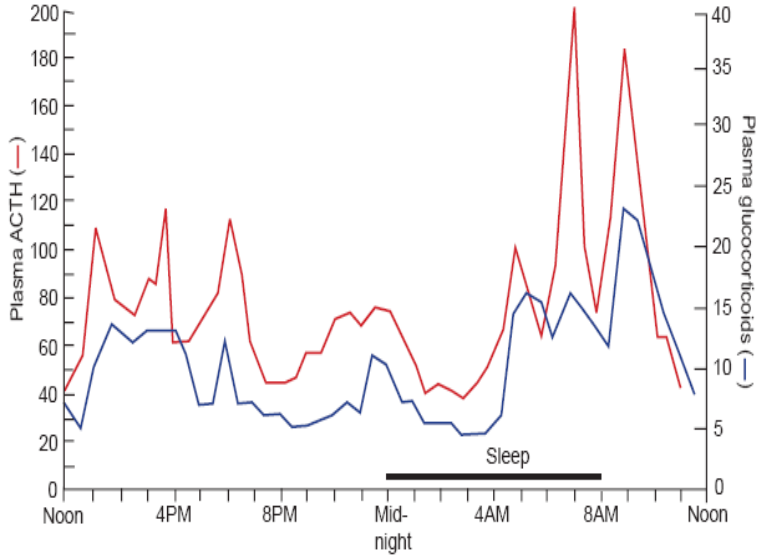
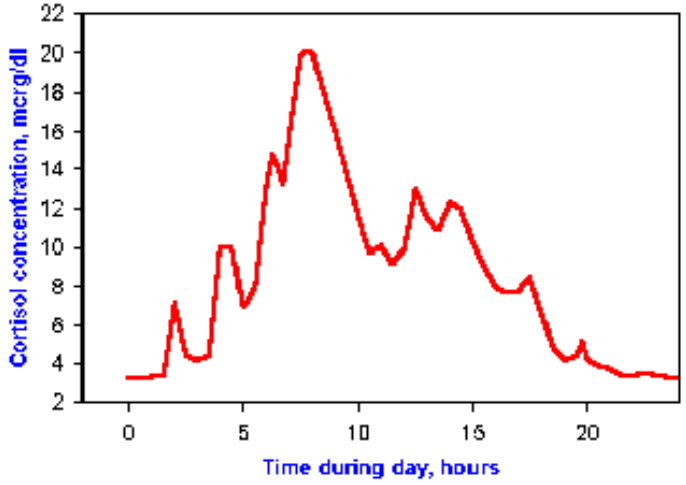


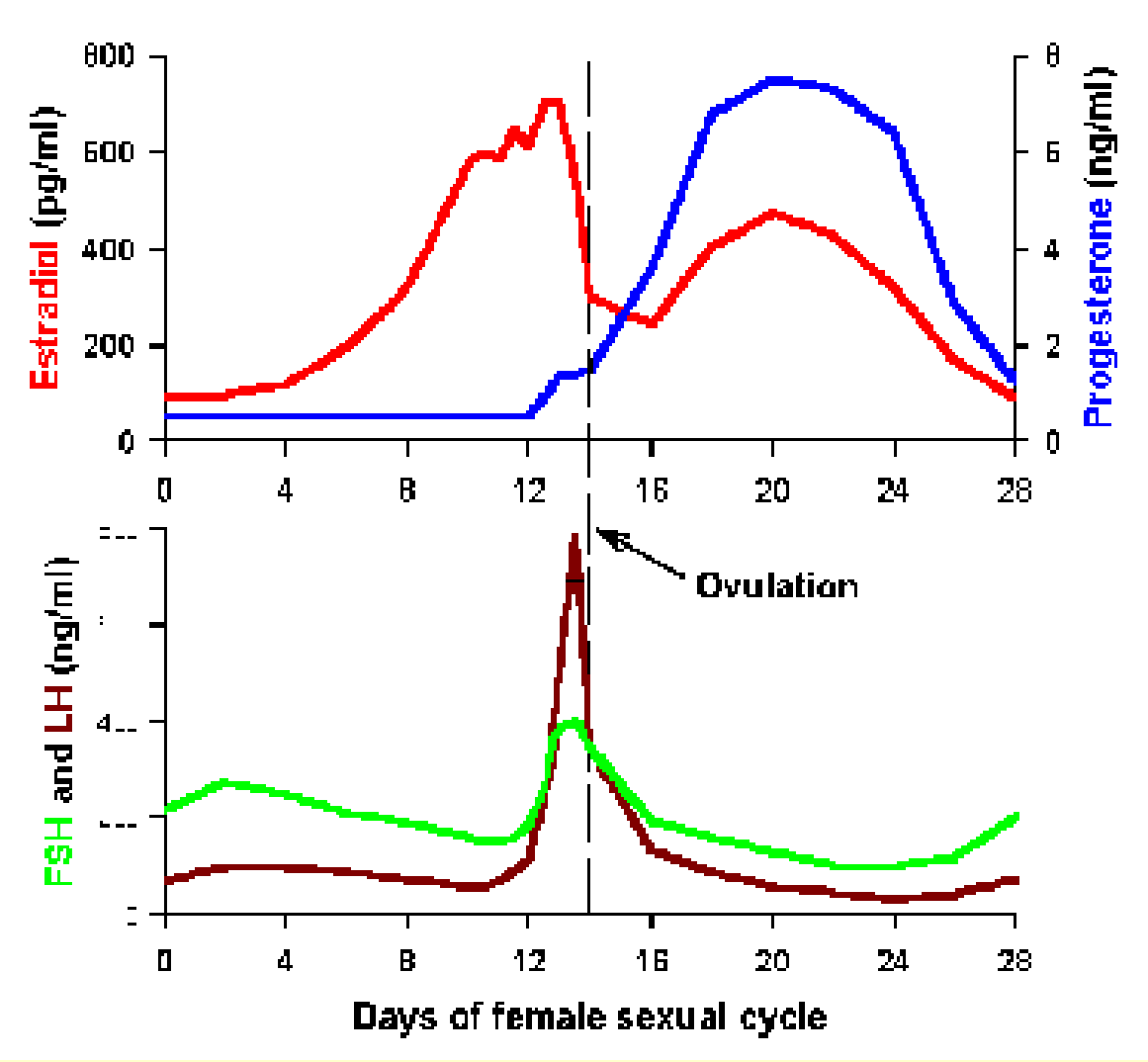
FIGURE 42-12 Pulsatile changes in the concentration of adrenocorticotropic hormone (ACTH) and glucocorticoids over a 24-hour period. The amplitude of the pulses of ACTH and glucocorticoids is lower in the evening hours and then increases greatly during the early morning hours. This is due to the diurnal oscillation of the hypothalamic-pituitary axis. (Modified from Krieger D.T. [1979]. Rhythms of CRF, ACTH and corticosteroids. In Krieger D.T. [Ed.], *Endocrine rhythms* [pp. 123–142]. New York: Raven)



Measurement of hormones in blood

- Hormones with long half life
  - e.g. thyroxin, triiodthyronin
- Hormones with short half life
  - daily rhythm
    - e.g. cortisol 5 fold increase from midnight to dawn
  - menstrual cycle

Hormone release during menstrual cycle



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  - e.g. thyroxin, triiodthyronin
- Hormones with short half life
  - daily rhythm
    - e.g. cortisol 5 fold increase from midnight to dawn
  - menstrual cycle
  - stress (catecholamins, cortizol)
  - pulse secretion
    - repeated measurement in short intervals, e.g. 20 min
  - proteolysis
    - ACTH, glucagon

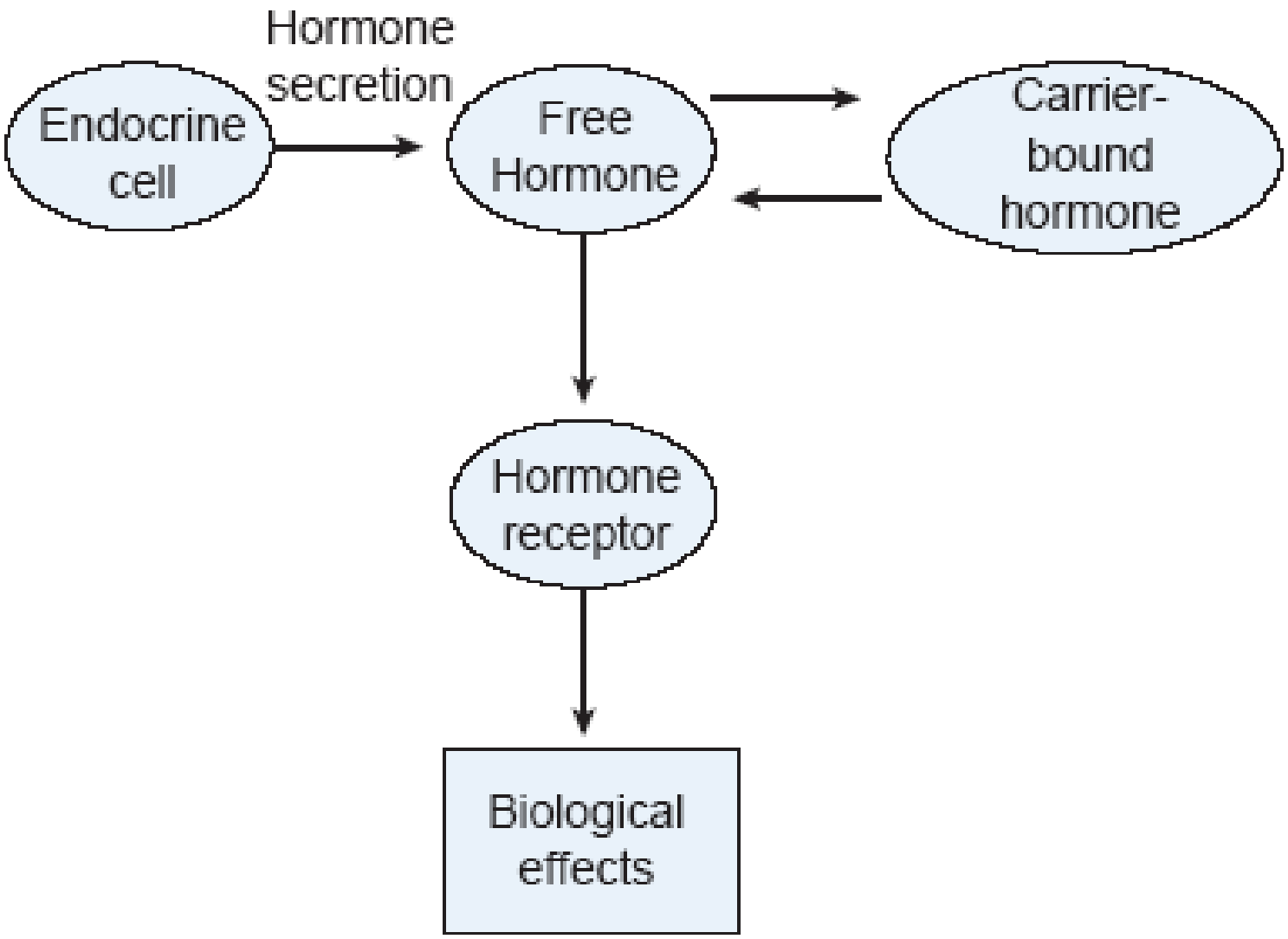


FIGURE 41-1 Relationship of free and carrier-bound hormone.

Measurement of hormone or hormone metabolites in urine

- Morning urine sample usually used for screening
  - Evaluation is based on creatinine concentration
- Assessment of hormone/metabolite production over 24 h period
- Depends on age, gender, and body weight
- E.g. 24 h urine cortisol = correlates with the amount of biologically available hormone, free cortisol, in blood

Measurement of hormones in saliva

- Nonprotein hormones
- Noninvasive – measurement of stress hormones (e.g. cortisol)

Function tests
Tests of the feedback loops

- Stimulation tests
  - are used in situations in which low hormone concentrations are expected = endocrine hypofunction
  - (adrenal insufficiency, pituitary hormone reserve, insulin test)
- Inhibition (suppression) tests
  - are used situations in which hormone concentrations are expected to be high = endocrine hyperfunction
  - (Cushing's sy)

Stimulation tests

- ACTH test
- Gonadoliberin stimulation test
- Insulin test

ACTH test

- stimulation with ACTH (8-9 am)
- measurement of cortisol
  - at least 2x increase is expected
- confirmation of primary adrenal insufficiency

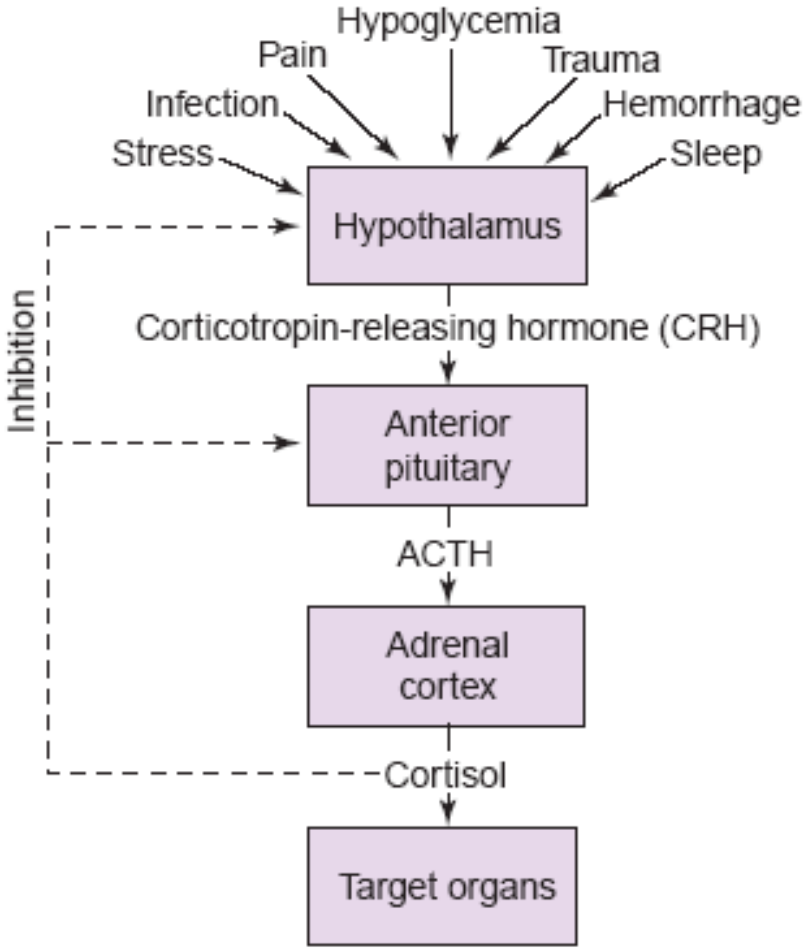


FIGURE 42-11 The hypothalamic-pituitary-adrenal (HPA) feedback system that regulates glucocorticoid (cortisol) levels. Cortisol release is regulated by ACTH. Stress exerts its effects on cortisol release through the HPA system and the corticotropin-releasing hormone (CRH), which controls the release of ACTH from the anterior pituitary gland. Increased cortisol levels incite a negative feedback inhibition of ACTH release.

Gonadoliberin stimulation test

- i.v. GnRH (LH-RH)
- measurement of LH, FSH and (testosterone – males) before, 25, and 45 minutes after GnRH administration
- Normal results:
  - LH ≥ 3x increase
  - FSH ≥ 2 x increase
  - Testosterone ≥ 50%
- indication: ovarial or testicular insufficiency

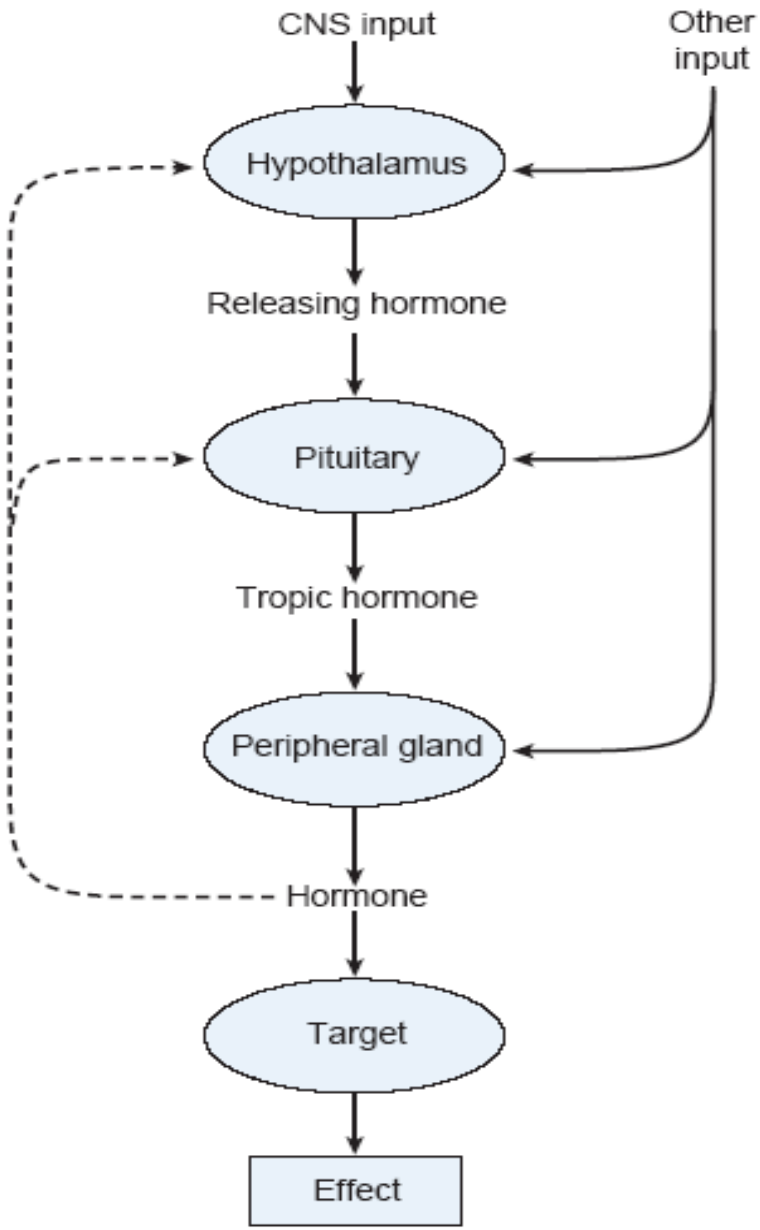


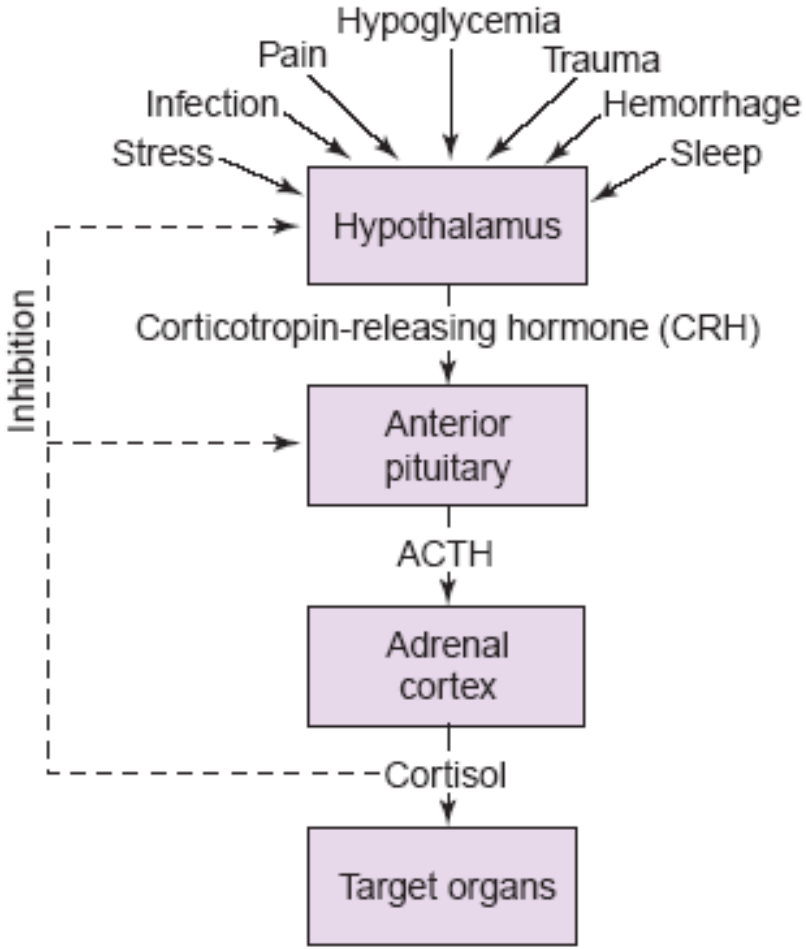
FIGURE 41-4 Hypothalamic–pituitary control of hormone levels. The *dashed line* represents feedback control.

Insulin test

- Insulin-induced hypoglycemia stimulates contraregulatory hormones (GH, ACTH – cortisole)
- i.v. aplication of insulin 0.3 U / kg
  - glycemia should decrease below 2.2 mmol/L
- physiologically GH > 400 pmol/l and cortisole > 550 nmol/L
- 10 % of healthy persons has false positive result of the test

Inhibition (suppression) tests

- Dexamethasone test
- Glucose test



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Dexamethasone test (short version)

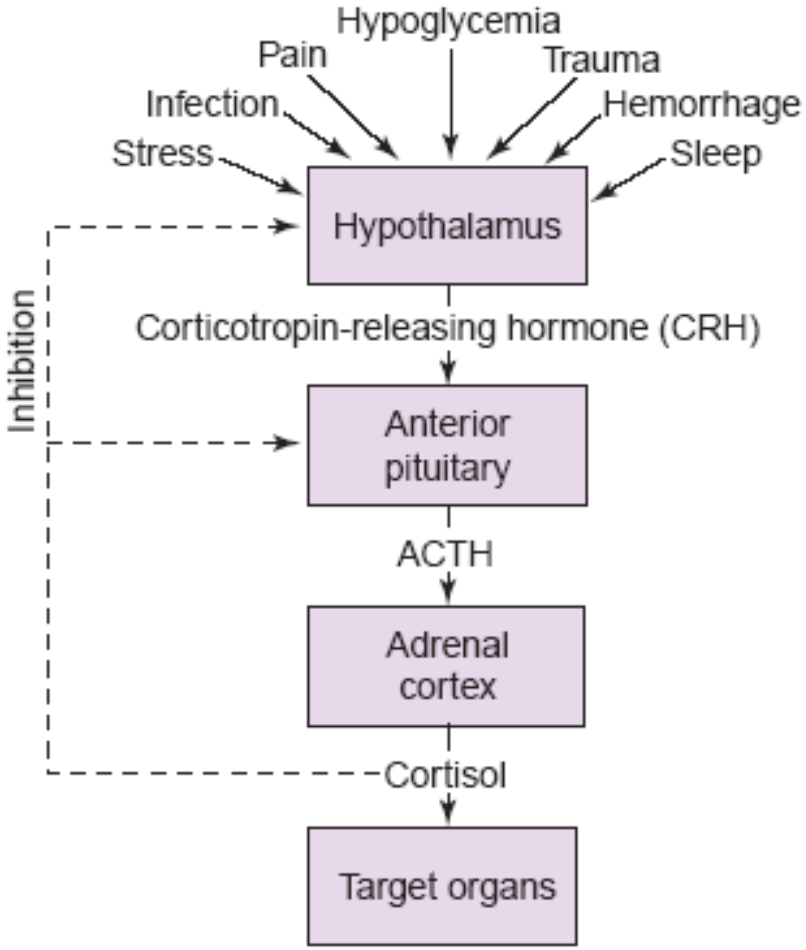
- p.o. Dexamethasone (2 mg) before midnight
  - Fasting blood/saliva sample is taken in the morning before and then after Dexamethasone administration
  - Expected result: suppression of cortisol concentration
- Confirmation of adrenal hyperfunction
  - Cushing's disease (pituitary adenoma) – suppression after more than 8 mg of Dexamethasone
  - Cushing sy - ectopic production of ACTH or autonomous production by tumor – no suppression
  - Adrenal gland adenoma - no suppression or by high dose

Glucose test

- Hyperglycemia suppresses the secretion of GH and ACTH
- Fasting + 100 g glucose p.o.
- GH measurement in 0, + 30, +60, +90 min.
- Physiologically GH should decrease below 1 μmol/L
- Pathological values in acromegaly

Factors to be taken in account in interpretation of hormone assays

- clinical symptoms and signs
- concentration of the variable regulated by the hormone
  - serum insulin concentration can only be interpreted simultaneously with glucose concentration
- concentration of other hormones in the feedback loop
  - correct interpretation of thyroid, adrenal or gonadal hormone concentrations requires the results of the appropriate pituitary hormone concentrations



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The end