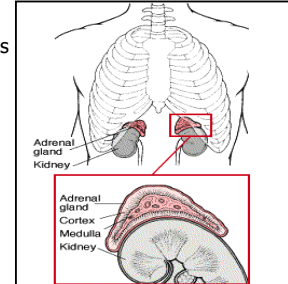
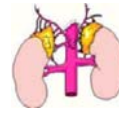


Adrenal gland - glucocorticoids

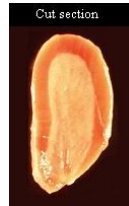
Adrenal gland - anatomy

- Located above kidneys
- A.k.a. suprarenal glands



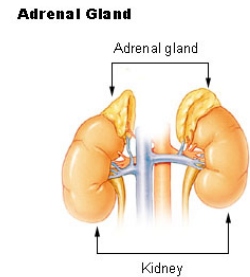
Adrenal gland - anatomy

- Two Regions
 - Medulla (inner)
 - Cortex (outer)
- Medulla & cortex are functionally different glands of different embryological origins:
 - Medulla derives from ectoderm (neural crest)
 - Cortex derives from mesoderm
- Is actually two separate organs in amphibians



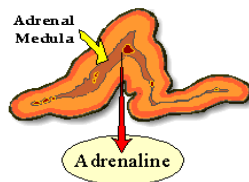
Blood supply

- The adrenals are well supplied with arterial blood from the aorta, renal, and phrenic arteries
- They have the highest rate of blood flow in the body

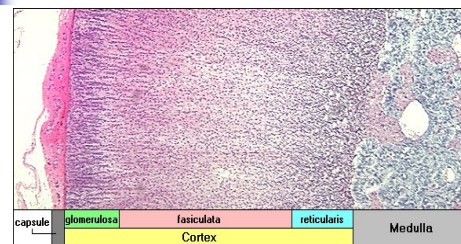


The medulla

- The medulla is responsible for production of adrenaline, which our body requires to cope with stressful situations

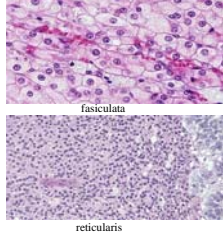


Three cortical zones



Histology of the adrenal cortex

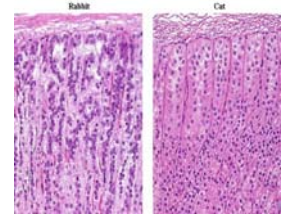
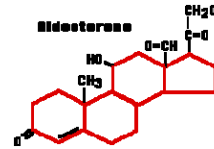
- Three concentric zones comprise 80-90%
 - Zona glomerulosa
 - Zona fasciculata
 - Zona reticularis



- Boundaries between the zones are indistinct, but the cellular patterns in each zone are clear

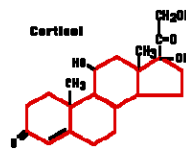
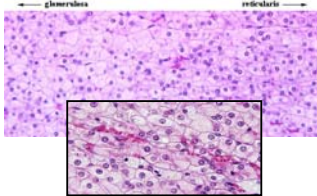
Zona glomerulosa (outer zone)

- Responsible for producing mineralcorticoids (aldosterone)



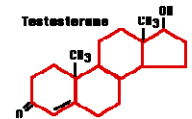
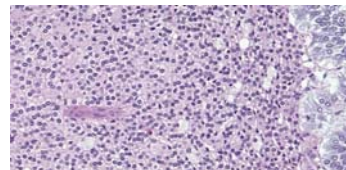
Zona fasciculata (middle zone)

- Responsible for producing glucocorticoids (cortisol)

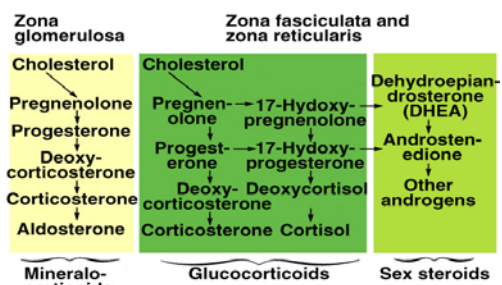


Zona reticularis (inner zone)

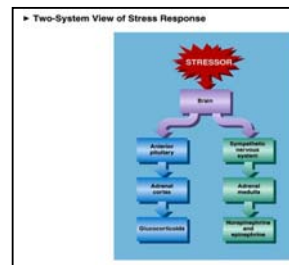
- Responsible for producing sex steroids (dehydroepiandrosterone (DHEA) : androgens)



Hormonal products of adrenal cortex



Cortisol:stress hormone response



Steroidogenesis

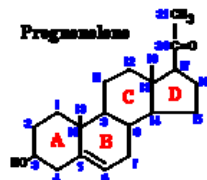
- Steroid hormones are produced from cholesterol
 - Cholesterol comes from LDL in blood
- Synthesis requires many oxidative enzymes located in mitochondria and endoplasmic reticulum

Steroidogenesis

- The hypothalamic-pituitary axis controls adrenal steroid production through ACTH release
 - ACTH binds to its receptor and activates adenylyl cyclase and production of cAMP
 - cAMP activates protein kinases which converts cholesterol esters into free cholesterol
 - Cholesterol is transported to mitochondria by StAR (steroidogenic acute regulatory protein)
 - rate-limiting step in steroid hormone production is the transport of free cholesterol from the cytoplasm into mitochondria

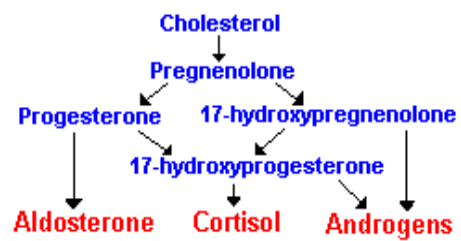
Steroidogenesis

- Within mitochondria, cholesterol is converted to pregnenolone by an enzyme in the inner membrane called CYP11A1



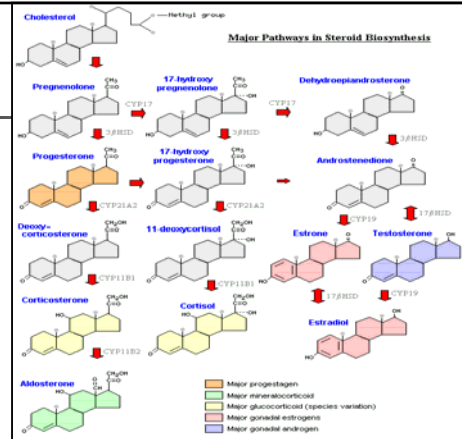
Steroidogenesis

- Pregnenolone is removed from the mitochondria and modified to form the 3 major types of adrenal hormones



Steroidogenesis

- Newly synthesized steroid hormones are rapidly secreted from the cell
- Increases in secretion reflect accelerated rates of synthesis

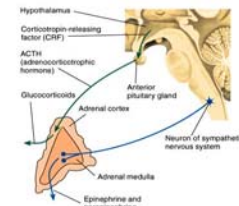


Cortisol secretion

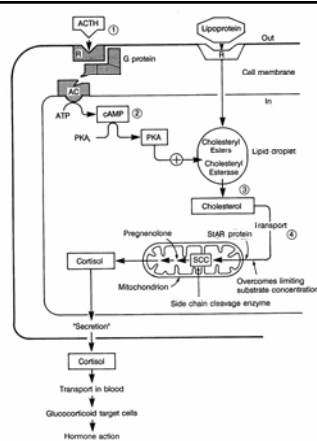
- Control
 - Hypothalamus secretes CRH → adenohypophysis secretes ACTH → adrenals secrete cortisol
- Rate
 - 8-25 mg/day
 - plasma [] = 40-180 ng/mL
 - vary with time of day & menses

Control of cortisol secretion

Control of the Secretion of Glucocorticoids by the Adrenal Cortex and of Catecholamines by the Adrenal Medulla



Control of cortisol secretion

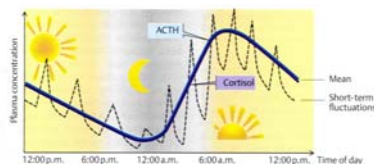


Patterns of cortisol secretion

- episodic & variable
- episodes follow ACTH secretion by 15-30 min
- burst in early am (an hour after awoken)
- rest of day brief bursts (7-15 episodes)
- increased release with coffee consumption
- increased during sleep stage when protein is being recycled
- increases with increased exercise time & intensity
- ↑ cortisol ↓ testosterone

Natural episodic secretion rhythms

- After ACTH has been produced, cortisol will be evident 15 to 30 minutes later
- There are usually 7-15 episodes per day
- There is a major burst in the early morning before awakening



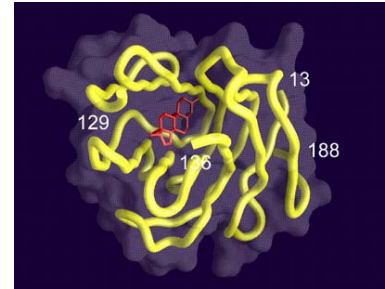
Steroid Hormones Transport

- Steroid Hormones are highly hydrophobic (slight ability to dissolve in water or plasma)
- To reach target tissue, they need assistance
 - An array of steroid-hormones-binding-proteins produced in the liver and released into the blood stream

Steroid Hormones Transport

- Steroid hormones when released from adrenal cortex into blood stream they bind to protein carriers
- Major protein carrier is albumin
- Other carriers are hormone-specific, such as sex hormone binding globulin (SHBG), or cortisol binding globulin (CBG)

Sex Hormone Binding Globulin (SHBG) bound to dihydrotestosterone



Steroid Hormones Transport

- Only unbound steroid hormones are biologically active (~2%)
- Binding globulins have an inhibitory effect on the steroid hormones
- To cross the target tissue membrane, the hormone must dissociate from its carrier protein
- Hormones can be in their inactive form when they cross the membrane (prohormone), requiring further target cell metabolism to activate them

Mechanism of action

- Enter target tissue via passive diffusion
 - **Because steroid hormones are lipids**
- Bind to specific glucocorticoid receptor localized in the cytoplasm
- When receptor is activated → disassociation from heat shock proteins & forms a receptor dimer →
- dimer crosses the nuclear membrane & binds to DNA @ specific sequences close to steroid-regulated genes →
- can ↓ ↑ gene expression & alters production of mRNA (this changes the cell's synthesis of certain proteins)

Nuclear Receptors

- Nuclear receptor superfamily, includes:
 - Steroid Hormone Receptors
 - Vitamins A&D & their dev. receptors
 - Metabolic Products Receptors
 - Xenobiotic Receptors
- Some receptors have affinity to different ligands:
 - Mineralocorticoid Receptor (MR) has equal affinity to Aldosterone and Cortisol
 - Androgen Receptor (AR) binds and responds to both Testosterone and Dihydrotestosterone

Nuclear Receptors

- Orphan Receptors:
 - Receptors that are recognized now but their activating ligands are yet to be known
- Variant Receptors:
 - Receptors that are a variant of known receptors but differ in splicing, making them unable to bind to ligands. However, they are thought to be biologically active by modulating the classical receptors

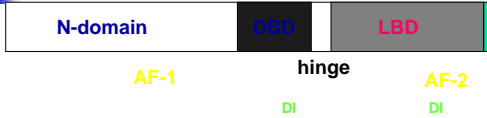
Steroid Hormone Receptors

- Receptors are proteins with molecular weight 50-100kd
- All of these receptors are localized in the nucleus via the Nuclear Localization Signal (NLS), except for glucocorticoid receptor (GR); which resides outside the nucleus when inactive
- Upon binding of a hormone, hormone-receptor complex becomes a transcription factor; i.e. stimulates / inhibits transcription of a particular gene

Glucocorticoid Receptors

- In the case of glucocorticoid receptor (GR), when unbound to a ligand, it is tethered in the cytoplasm to chaperone molecules, including heat shock proteins (hsp).
- When bound to ligand, GR undergoes conformational changes that dissociate it from the hsp and exposes its NLS, translocating it to the nucleus

Modular Structure of Steroid Receptors



AF-1/AF-2 transcription activation functions or "domains"
 DI: dimerization interfaces
 dependent(DBD) and independent
 DBD DNA binding domain
 LBD ligand binding domain

Modular Structure of Steroid Receptors

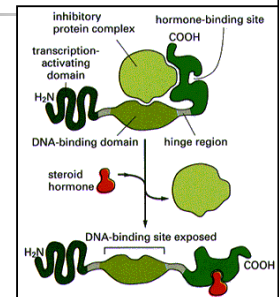
- the DNA-binding domain, shows the highest degree of homology among the receptors
- the ligand-binding domain, shows lower degree of homology. Greater homology is observed when the ligands are similar

Modular Structure of Steroid Receptors

- The activation domains, namely, those involved in affecting the transcription machinery, are named AF-1 and AF-2, respectively. (AF=activating function).
- other domains of the receptor: dimerization, HSP binding, and nuclear localization.

Model of activation of intracellular receptors

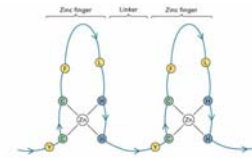
- In the absence of hormone, transcriptional activation is inhibited
- Hormone binding relieves inhibition
 - Release of inhibitory complex – hsp
- Exposing of DNA binding domain



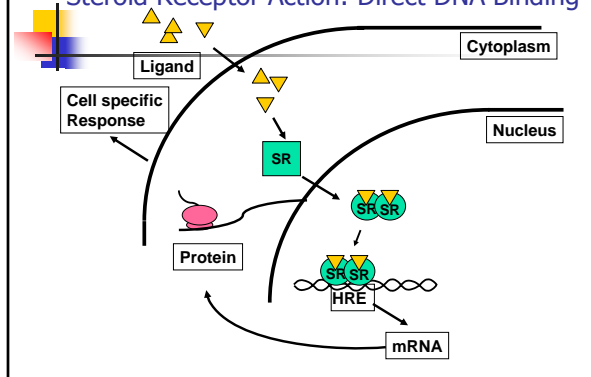
Ligand-Receptor DNA Transcription

- In the nucleus, the ligand-receptor complex recognizes and binds to specific locations on the DNA called Hormone Response Elements (HRE).
- Zinc fingers in the C-domain of the receptor mediates the HRE recognition and binding.
- Steroid Receptors act as homodimers when bound to DNA for regulation of gene transcription
- Other Nuclear Receptors act as heterodimers

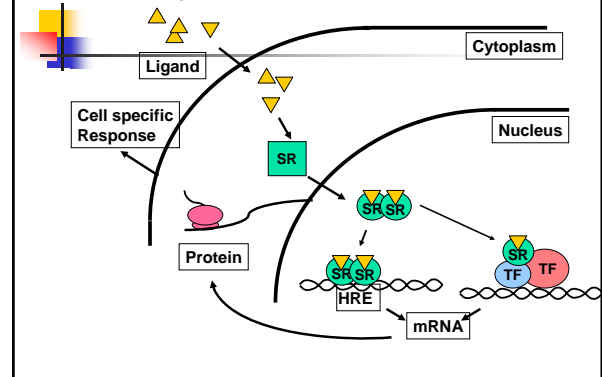
Zinc Fingers



Steroid Receptor Action: Direct DNA Binding



Steroid Receptor Action: Protein/Protein



Steroid Receptor Animation

- <http://www.maxanim.com/biochemistry/Steroid%20Hormone/Steroid%20Hormone.htm>

Glucocorticoids

- Main glucocorticoids in humans
 - Cortisol
 - Corticosterone
- Cortisol:corticosterone produced in humans in a ratio of 10:1
- Both are 21 carbon steroids & are 90-95% bound to plasma protein (transcortin)
- Under control primarily by ACTH

Corticosterone

- Biosynthesis
 - Cholesterol → ^{21β}hydroxylase → corticosterone → aldosterone
- Is not the primary glucocorticoid in humans & most mammals
 - Does serve this function some rodents
- Suppresses inflammatory reactions and affects the immune system
- Our focus will be on cortisol

Cortisol

- The primary glucocorticoid
- Essential for LIFE!!!
- The STRESS hormone
 - Need it when under prolonged stress
 - Mental & physical
 - Uses gluconeogenesis to breakdown muscle & fat into glucose which is used as brain energy
- Necessary to maintain critical processes in times of stress
- Controls inflammation reactions

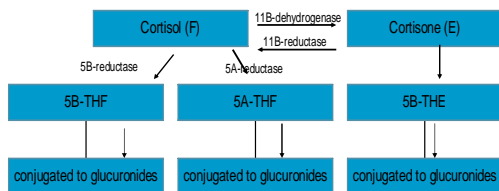
Cortisol metabolism

- Most bound to transcortin (α-2globulin) in circulation
 - transcortin synthesized in kidney and increased by estrogen
 - ↑ transcortin = ↑ bound cortisol
 - women with high levels or taking estrogen will have high total plasma cortisol levels but no ill effects of excess cortisol
- 15-20% bound to albumin (less tightly)
- 5% unbound
 - Free cortisol excreted into urine

Cortisol metabolism

- Metabolized in liver by reductases & conjugated to glucuronides which are excreted via kidney
- T_{1/2} = 70-90 minutes
- Can be converted to cortisone
 - (an 11 keto analog)
 - 11β-dehydrogenase
 - Cortisol (F) → cortisone (E)
 - Inhibited by large consumption of licorice

Cortisol metabolism conversion of cortisol → cortisone



Intermediary metabolism

- Catabolic
 - Glucogenesis - primary function
 - facilitates breakdown of protein & connective tissue in muscles to glucose & glycogen
 - involves the ↑ breakdown of protein & decreased formation of new protein
 - Lipolysis
 - facilitates the breakdown of fat
 - redistributes body fat (mechanism unknown)

Carbohydrate metabolism

- Increased blood glucose levels via gluconeogenesis in the liver
 - Glucose synthesized from non-hexose substrates
 - amino acids & lipids
 - Enhances the enzymes involved in gluconeogenesis
- Decreases utilization of glucose by cells
 - DNA transcription in liver cells increases the enzymes responsible for converting amino acids to glucose & glycogen
 - Done via direct inhibition of glucose transport into cells
 - Result is an increased blood glucose level

Protein metabolism

- Decreases utilization of amino acids
 - Reduces protein formation
 - Occurs everywhere EXCEPT liver
- Extrahepatic protein stores reduced
 - amino acids not transported into muscle cells ↓ protein synthesis & ↑ amino acid blood levels
 - These high blood amino acid levels are transported more rapidly to hepatic cells → gluconeogenesis → glycogen formed → protein synthesis in liver

Fat metabolism

- Lipolysis
 - Mobilizes fatty acids & glycerol from adipose tissue →
 - ↑ their blood concentrations → makes more glycerol available for gluconeogenesis
 - Fat broken down & less formed
 - probably due to less glucose transported into fat cells
- Redistribution of body fat
 - ↑ formation of fat in trunk areas & face
 - ↓ fat (& muscle) from extremities
- Increased appetite

Functions - circulation

- Maintains body fluid volumes & vascular integrity
 - Water diuretic hormone & controls half the intestinal diuresis
 - Cortisol levels vary with water intake
 - ACTH inhibited by water deprivation
- Mineralcorticoid effect
 - Cortisol has mineralcorticoid effect
 - Not as potent as aldosterone (300-600x)
 - BUT cortisol levels are 200x aldosterone levels

Functions - circulation

- Vasoconstriction
 - BP regulation & cardiovascular function
 - Sensitizes arterioles to action of noradrenaline
- Decreased capillary permeability
- Maintains normal renal function

Functions - inflammation & immunity

- Anti-inflammatory & immunosuppressive
 - Recruits neutrophils
 - Inhibits prostaglandin & leukotriene synthesis
 - Inhibits expression of COX-II
 - ↑ transcription of genes coding for anti-inflammatory proteins & upregulation cytokine receptor expression
 - ↑ IL-10, lipocortin-1, neutral endopeptidase
 - ↓ phospholipase A2 & cyclo-oxygenase type 2 →
 - ↓ prostaglandins & leukotrienes
 - ↓ IL-1,3,4,5,8 ; TNF-α → ↓ inflammation due to cytokines

Functions - inflammation & immunity

- induction of endonucleases → apoptosis in eosinophils & lymphocytes
- Inhibits leukocyte & macrophage functions

Functions - CNS responses

- Negative feedback control on release of ACTH
- Modulates perception & emotion
 - Recall the term stress hormone

Functions - developmental

- Permissive regulation of fetal organ maturation
 - Intestinal enzyme maturation leads to various pulmonary functions
 - surfactant synthesis (phospholipid that maintains alveolar surface tension)
- Inhibition of linear growth in children due to direct effects on bone & connective tissue

Functions - hormone modification

- Inhibition on pituitary release of TSH & GnRH
 - Suppresses enzyme that converts T4 → T3
 - This decreases metabolic rate & may make it harder to lose body fat
 - In severe hyperthyroidism, use glucocorticoids to ↓ thyroid hormones &/or pituitary release of TSH
- Inhibition of GH by stimulating somatostatin
- May reduce IGF-1 expression
 - Recall, IGF-1 is an anabolic agent
 - responsible for most of the effects of GH
 - GH → IGF-1 in liver

Cortisol deficiency - etiology

- Primary Hypofunction
 - Site of impairment is in the adrenal gland
 - Primary hypofunction of adrenal glands used to be main reason for dysfunction (Addison's disease)
 - Caused by TB destruction of adrenal gland
 - Now either unknown cause OR autoimmune disease
- How it works:
 - adrenal gland destroyed → cortisol deficient → plasma levels ACTH high due to no inhibition via (-) feedback
 - additional exogenous ACTH can't stimulate the damaged gland

Cortisol deficiency

- Secondary Hypofunction
 - Loss of hypothalamic-pituitary function and deficiency of ACTH
 - Will usually have deficiencies of other glands regulated by hypothalamus-pituitary system
- How it works:
 - problem in hypothalamus or pituitary → plasma ACTH levels low → adrenals suppressed → cortisol levels low
 - adrenals do respond to exogenous ACTH

Primary vs. secondary hypofunction

Primary

- Site = adrenal
- ↑ ACTH secretion
- ↑ pigmentation
- Optic involvement rare
- ↓ weight
- No change in GH or gonadotropin
- Deficient in other adrenal hormones
- No response to exogenous ACTH

Secondary

- Site = hypothalamus-pituitary
- ↓ ACTH secretion
- ↓ pigmentation
- Optic involvement
- Weight changes vary
- ↓ in GH & gonadotropin
- Other adrenal hormones normal or slightly ↓
- Sluggish response to exogenous ACTH

Cortisol deficiency -

- Organs can not respond to everyday stress
 - Exercise
 - Salt overload
 - Hypoglycemia (caused by prolonged fasting or exercise)
- Any major stress will lead to death

Cortisol deficiency - intermediary metabolism

- In protein metabolism
 - decreased cortisol does not cause any increased protein synthesis
 - in skeletal & cardiac muscle, decreased cortisol leads to muscle fatigue & heart failure

Cortisol deficiency - intermediary metabolism

- in carbohydrate metabolism
 - extreme insulin sensitivity
 - ↑ susceptibility to hypoglycemia
 - harder to recover from hypoglycemia
- in fat metabolism
 - decreased cortisol does not cause any increased fat synthesis

Cortisol deficiency - circulation

- Hypotension
 - Abnormal vasodilatation → BP ↓
- Hyponatremia & hyperkalemia
 - Glomerular filtration falls → H₂O not excreted as fast
 - Inability to concentrate urine
 - Excessive urination → dehydration
 - Na-K balance via mineralocorticoid effects
 - Low levels of cortisol cause ↓ Na and ↑ K levels
 - BUT when BP ↓ → renin-angiotension mechanism activated → aldosterone stimulated NOT cortisol
- Hypovolemia due to decreased Na retention

Cortisol deficiency

- Inflammation & immunity
 - More likely to develop autoimmune disease
 - Role of cortisol in lysosome stability
 - Role of cortisol in phagocytosis
- CNS
 - Accentuates senses (taste, smell...)
 - Lethargy
 - Apathy
 - Lack of concentration
 - Link between jet lag & low cortisol

Cortisol deficiency

- Skin pigmentation
 - due to \uparrow ACTH (which affects melanin production)

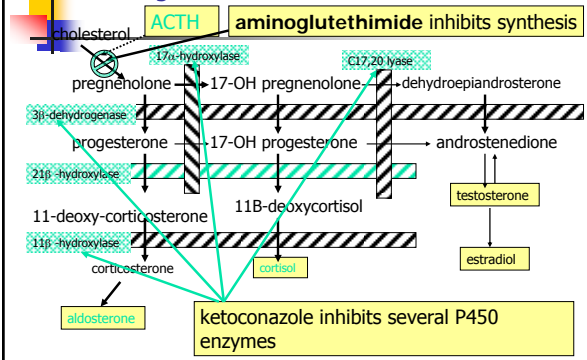
Cortisol deficiency- diseases

- Addison's Disease
 - affects 4 out of 100,000 people
 - can strike at any age
 - affects males and females equally
 - 30 % of people, the adrenal glands are destroyed by :
 - a cancer
 - amyloidosis
 - an infection such as tuberculosis
 - or another identifiable disease
 - in 70 %, the cause isn't known, suspect an autoimmune reaction

Cortisol deficiency - diseases

- Other Causes
 - Ketoconazole & aminoglutethimide (antifungal drugs)
 - blocks synthesis
 - Congenital adrenal hyperplasia
 - synthesis enzyme defects
 - Use of exogenous corticosteroids
 - must taper off slowly
 - failure to do so results in adrenal not producing sufficient cortisol
 - can take weeks or months

Aminoglutethimide & ketoconazole



Congenital adrenal hyperplasia

- CAH is a genetic defect of the adrenal glands. A person with CAH will not be able to produce cortisol.
- In affected individuals, the disease begins early in gestation and leads to disease that is manifest at birth.
- Continued secretion of ACTH causes never ending stimulation of the adrenal cortex, leading to hyperplasia (an increase in the number of cells in that tissue).
- CAH is treated with hormone replacement, replacing one or both of the hormones missing

Cortisol excess: exogenous & endogenous

- Exogenous
 - Most cortisol excess is induced by steroid therapy (prednisone) to manage disease
 - asthma
 - rheumatoid arthritis
 - lupus
 - other inflammatory diseases
 - immunosuppression after transplantation
 - Side effects can be minimized by using the synthetic prednisone intermittently

Cortisol excess: exogenous & endogenous

- Endogenous
 - Due to excessive production of cortisol
 - ACTH- independent
 - Primary adrenal defect
 - ACTH-dependent
 - Overproduction of ACTH by pituitary
 - Overproduction of ACTH by ectopic ACTH-producing tumor
 - Both exogenous & endogenous hyperfunction show manifestations of Cushing's

Cortisol excess: intermediary metabolism

- in carbohydrate metabolism
 - ↑ blood glucose levels
 - ↓ sensitivity to insulin
- in protein metabolism
 - ↑ protein loss → muscle atrophy
 - thin skin
 - bone matrix & mass losses; bone formation ↓ → less Ca^{2+} absorbed & more excreted in urine → osteoporosis

Cortisol excess: intermediary metabolism

- in fat metabolism
 - redistribution of body fat:
 - ↑ trunk & face fat deposition & ↓ extremities fat deposition

Cortisol excess: circulation

- Hypertension
 - Due to Na retention & K excretion
- Hypervolemia
 - increased blood volume
- Hyponatremia
 - increased Na absorption
- Hypokalemia
 - increased K excretion

Cortisol excess: inflammation & immunity

- Decreases inflammation
- Increased infection susceptibility
 - Ab synthesis suppressed & normal immune responses to infecting pathogens suppressed
- Decrease in fibrous tissue formation due to decreased capillary permeability

Cortisol excess – effects on CNS

- Initially euphoria but replaced with depression
- Seizure threshold lowered

Cortisol excess – diseases and symptoms

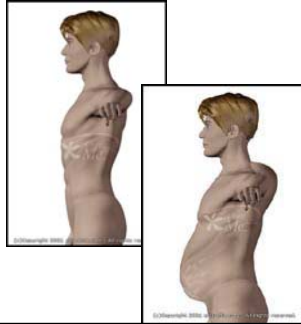
- | | |
|---|---|
| <ul style="list-style-type: none"> ■ Cushing's disease ■ Pituitary tumor ■ Small cell carcinoma (ectopic ACTH producing tumors) ■ Adrenal tumor <ul style="list-style-type: none"> ■ Benign adenoma | <p>Symptoms</p> <ul style="list-style-type: none"> ■ Moon face ■ ↑ fat in torso ■ Thin skin & bruise easily ■ Poor wound healing ■ ↑ BP ■ Osteoporosis ■ ↑ risk of diabetes & kidney stones ■ Slow growth in children |
|---|---|

Did you know?

- Cushing's syndrome, or hypercortisolism, is a disorder caused by prolonged exposure of the body's tissues to high levels of cortisol.
- Cushing's disease specifically means an excess of ACTH in the body
 - Affects adults aged 20 to 50
 - An estimated 10 to 15 of every million people are affected each year

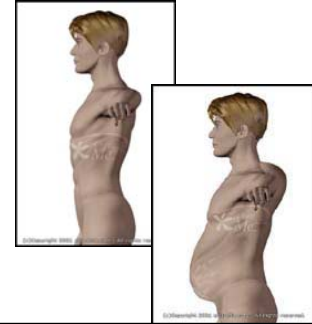
Cushing's syndrome symptoms

- Moon" face
- "Buffalo" hump on the back
- Protruding abdomen
- Thinning of arms and legs
- Emotional problems
- Menstrual periods decrease or stop
- Cuts heal slowly
- Impotence

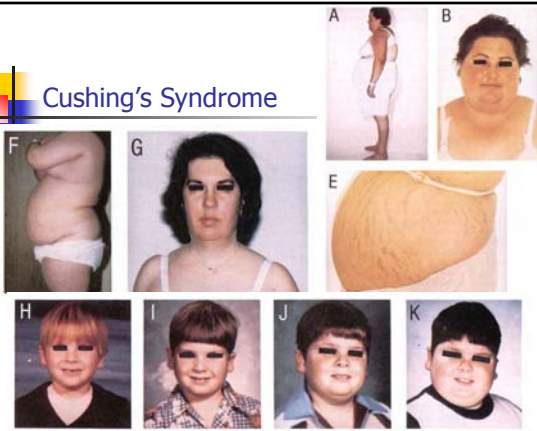


Cushing's syndrome symptoms

- Weakness
- Headache
- High blood pressure
- Acne
- Backache
- Excessive thirst
- Excessive urination
- Purple "stripes" on skin
- Easy bruising



Cushing's Syndrome



Did you know?

- Pituitary tumors are removed through the nose
 - This procedure is known as a transsphenoidal adenectomy

Ectopic ACTH Syndrome

- Benign or malignant tumors that arise outside the pituitary and produce ACTH
- Lung tumors cause over 50 percent of these cases.
 - The most common forms of ACTH-producing tumors are oat cell, or small cell lung cancer, which accounts for about 25 percent of all lung cancer cases
 - Other less common types of tumors that can produce ACTH are thymomas, pancreatic islet cell tumors, and medullary carcinomas of the thyroid

Glucocorticoid Effect Comparisons (potency at equivalent doses)

| | GLUCOCORTICOID EFFECT | MINERALOCORTICOID EFFECT |
|---|-----------------------|--------------------------|
| Glucocorticoids | | |
| Short-Acting (biologic $t_{1/2}$ = 8-12 h) | | |
| hydrocortisone | 1 | 1 |
| Intermediate-Acting (biologic $t_{1/2}$ = 18-36 h) | | |
| prednisone | 4 | 1 |
| triamcinolone | 5 | 0 |
| Long-Acting (biologic $t_{1/2}$ = 36-54 h) | | |
| dexamethasone | 30 | 0 |
| betamethasone | 25 | 0 |
| Mineralocorticoid | | |
| fludrocortisone | 10 | 250 |

Cortisol Suppression Tests

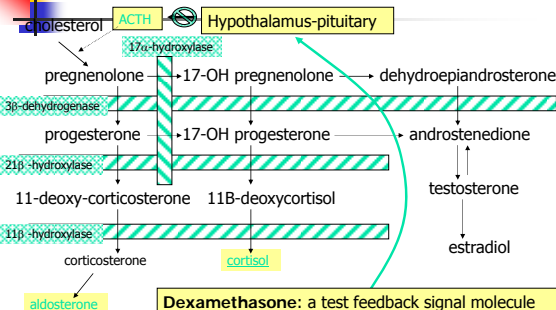
- Principle
 - Based on the ability of exogenous cortisol to exert (-) feedback on hypothalamus-pituitary release of ACTH
 - Can't measure with cortisol itself (exogenous would just replace endogenous)
 - Must use a more potent glucocorticoid derivative
 - usually dexamethasone

Diagnosis of H-P-A Axis status: H-P suppression test

- Dexamethasone:
 - used to evaluate the basis for elevated cortisol levels in individuals with suspected pituitary adenoma (Cushing's disease)
 - Normal response in Cushing's disease:
 - plasma ACTH and cortisol, urine 17-OH corticosteroid levels are reduced
 - Abnormal response in cortisol-producing adrenal tumor (low ACTH) or ectopic ACTH-producing tumors (high ACTH):
 - plasma ACTH and cortisol, urine 17-OH corticosteroid levels are unchanged

Dexamethasone:

Hypothalamus-Pituitary suppression test



Diagnosis of H-P-A Axis status: Adrenocortical function test

- Cosyntropin:
 - synthetic ACTH used as adrenal cortical stimulant
 - Normal response:
 - plasma cortisol levels are elevated
 - Abnormal response:
 - plasma cortisol level are unchanged

