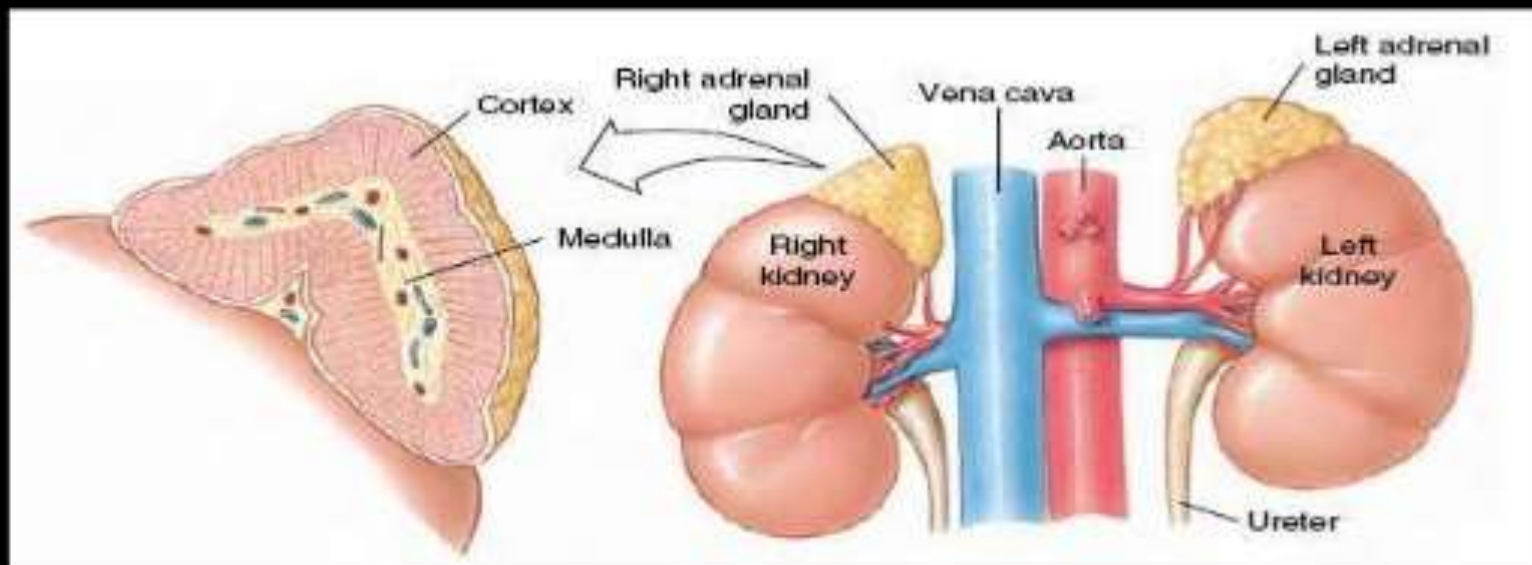


- **ADRENAL
FUNCTION TESTS**
- **Sangeeta Trimbake**

INTRODUCTION

- The two adrenal glands, each of which weighs about 4 grams, lie at the superior poles of the two kidneys.
- Each gland is composed two distinct parts: the inner medulla and the outer cortex.



Adrenal glands

- Each gland is divided into two parts:
 - **Cortex** – outer part of gland
 - Part of hypothalamus – pituitary – adrenal axis
 - Secrete a variety of steroid hormones
 - **Medulla** – inner part of gland, (20% of gland)
 - Part of sympathetic nervous system
 - Secrete catecholamines
 - Both parts are structurally and functionally different

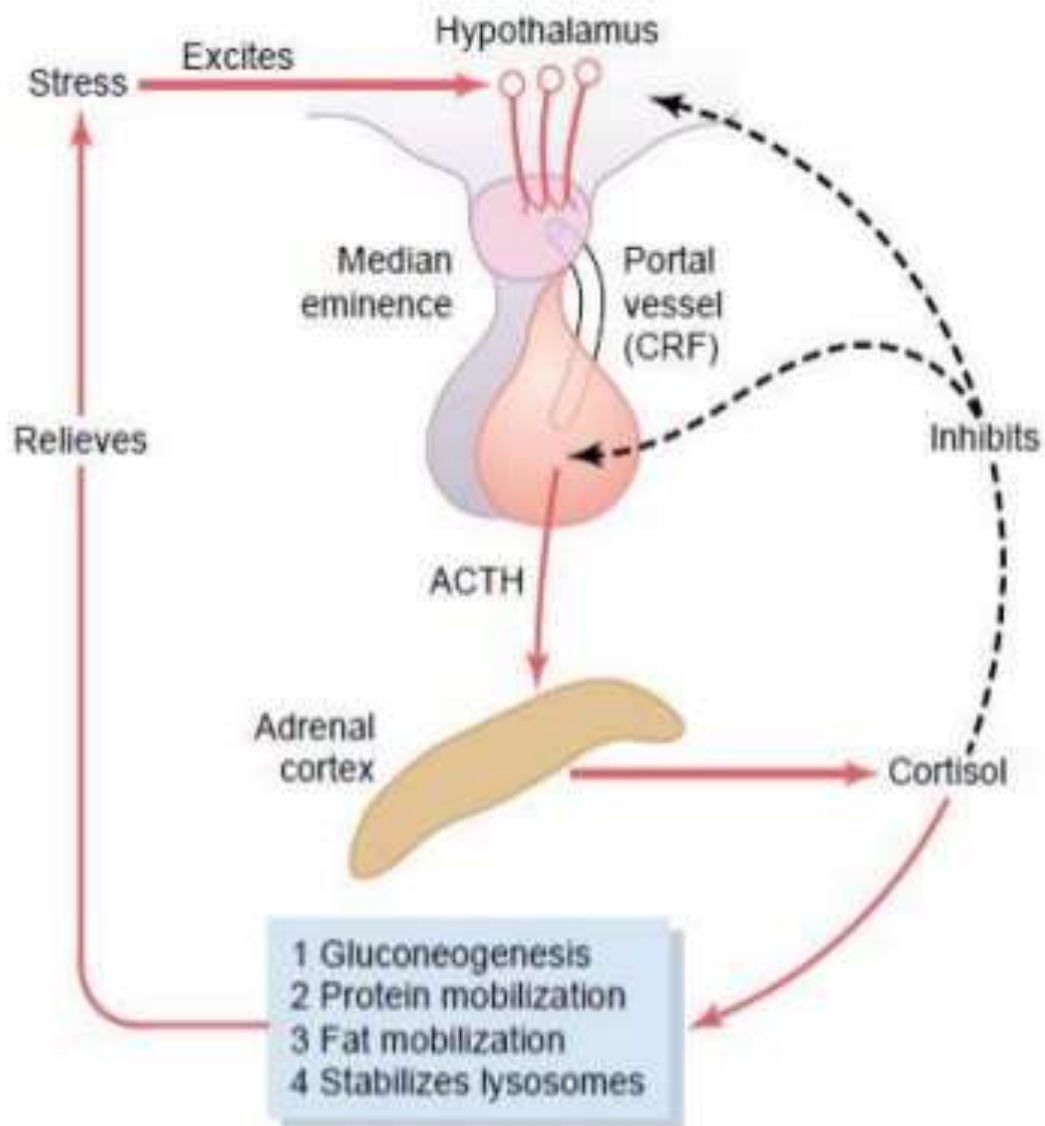
Adrenal cortex

- The large cortical cells are arranged into three layers or zones :
 - The **zona glomerulosa**,
 - The thin outermost layer
 - Constitute about 15% of cortex
 - The **zona fasciculata**,
 - The middle and largest portion
 - Constitute about 75% of cortex.
 - The **zona reticularis**,
 - The innermost zone.

ADRENAL CORTEX:

- Secretes corticosteroids- mineralo-corticosteroids, gluco-corticosteroids and androgenic hormones.
- *Mineralo-corticosteroids*- affect electrolytes of extracellular fluids: sodium and potassium.
- *Gluco-corticosteroids*- increase the blood glucose level.
- *Androgenic hormones*- exhibit the same effects as the male sex hormones.

- The cortisol has two basic anti-inflammatory effects:
 - (1) It can block the early stages of the inflammation process before inflammation even begins.
 - (2) If inflammation has already begun, it causes rapid resolution of the inflammation and increased rapidity of healing.
 - *Cortisol reduces lymphocyte production and hence, suppresses immunity.*
 - Cortisol Blocks the Inflammatory Response to Allergic Reactions.
-



ADRENAL INSUFFICIENCY: CAUSES



20.44 Causes of adrenocortical insufficiency

Secondary (\downarrow ACTH)

- Withdrawal of suppressive glucocorticoid therapy
- Hypothalamic or pituitary disease

Primary (\uparrow ACTH)

Addison's disease

Common causes

- Autoimmune
 - Sporadic
 - Polyglandular syndromes (p. 794)
- Tuberculosis
- HIV/AIDS
- Metastatic carcinoma
- Bilateral adrenalectomy

Rare causes

- Lymphoma
- Intra-adrenal haemorrhage (Waterhouse–Friedrichsen syndrome following meningococcal septicaemia)
- Amyloidosis
- Haemochromatosis

Corticosteroid biosynthetic enzyme defects

- Congenital adrenal hyperplasias
- Drugs
 - Metyrapone, ketoconazole, etomidate

Disorders of adrenal cortex


- Patient with adrenal disorders can present with features related to:
 - **HYPOFUNCTION OF THE GLAND**
 - **HYPERFUNCTION OF THE GLAND**

Adrenal Hypofunction

- Adrenal insufficiency leads to a reduction in the output of adrenal hormones
 - glucocorticoids and/or mineralocorticoids
- Two types of adrenal insufficiency
 - **Primary insufficiency**
 - inability of the adrenal glands to produce enough steroid hormones
 - **Secondary insufficiency**
 - inadequate pituitary or hypothalamic stimulation of the adrenal glands

Adrenal Hypofunction

- **Causes**

- **Glucocorticoid treatment**
 - **Autoimmune adrenalitis**
 - **Tuberculosis**
 - Adrenalectomy
 - Secondary tumor deposits
 - Amyloidosis
 - Haemochromatosis
 - Histoplasmosis, tuberculosis, CMV, AIDS
 - adrenal haemorrhage
- 
- Common

Adrenal Hypofunction

- **Causes**

- Metabolic failure in hormone production
 - Congenital adrenal hyperplasia e.g. 21-hydroxylase deficiency, 3- β -hydroxysteroid dehydrogenase deficiency
 - Enzyme inhibition e.g. ketoconazole
 - Accelerated hepatic metabolism of cortisol e.g. phenytoin, barbiturates, rifampicin

Adrenal Hypofunction

Addison's disease Primary hypoaldosteronism

Addison's disease: pathogenesis

- **Progressive destruction of entire adrenal cortex ,**
This is usually autoimmune based.
- Most likely the result of cytotoxic T lymphocytes,
although 50% of patients have circulating adrenal
antibodies.

Addison's disease: Clinical features

Common	Less common
Tiredness, generalized weakness, lethargy	Hypoglycemia
Anorexia, nausea, vomiting	Depression
Hyponatremia Hyperkalemia ,Hypercalcemia Convulsions	
Dizziness and postural hypotension	
Pigmentation	
Loss of body hair (woman)	

ADRENAL CRISIS

- **Causes :**
 - Precipitated by stress
 - infection, trauma or surgery in patients with incipient adrenal failure/treated with glucocorticoids if dosage is not increase
 - Adrenal haemorrhage
 - due to cx of anticoagulant treatment
 - Meningococcal septicaemia

INVESTIGATIONS (HORMONAL)

- **Plasma cortisol concentration**
 - <50nmol/L at 0900H → effectively diagnostic
 - >550nmol/L excludes the Dx
- **ACTH stimulation test / Synacthen test**
- Measurement of **plasma ACTH**
- Metyrapone test
- CRH stimulation test
- Plasma **renin** and **aldosterone** levels

Plasma cortisol

- The initial definitive test
- Cortisol level in blood show diurnal variation
- The levels high in morning (8.00 am)than in evening.
- Usual morning 8.00 am levels range between 8-26 ug/dl
- Loss of diurnalrythm is an indication of adrenal cortex function.

Plasma cortisol

- Levels are higher in hyperadrenocorticism and lower in hypoadrenocorticism.

PLASMA ACTH MEASUREMENT

- To differentiate between primary and secondary adrenal failure
 - Primary insufficiency - ACTH increased
 - Secondary insufficiency - ACTH decreased

Stimulation test for Hypofunction

INVESTIGATIONS (HORMONAL)

- **ACTH stimulation test / Synacthen test**

SHORT TEST	LONG TEST
<ul style="list-style-type: none">➤ Take blood sample at 0900H for measurement of cortisol	<ul style="list-style-type: none">➤ Day 1 : inject 1 mg depot ACTH IM im
<ul style="list-style-type: none">➤ Inject 250µg ACTH im or iv	<ul style="list-style-type: none">➤ Days 2 and 3 : repeat
<ul style="list-style-type: none">➤ Take further blood sample after 30 and 60 min for cortisol measurement	<ul style="list-style-type: none">➤ Day 4 : perform short ACTH test

Stimulation test for Hypofunction

- Rapid ACTH stimulation tests
- 25 units or 250 μg of ACTH administration intravenously or intramuscularly is coupled with basal and post ACTH administration cortisol measurement at 30 and 60 minutes in plasma.
- There should be a minimum rise of more than 7 $\mu\text{g}/\text{dl}$ over the basal level or plasma level should be more than 18 $\mu\text{g}/\text{dl}$

Stimulation test for Hypofunction

- Rapid ACTH stimulation tests
- Lower results show hypofunction.
- Test may not show the rise in severely ill patients.

Stimulation test for Hypofunction

- Long ACTH stimulation test
- Lower dose of 1 $\mu\text{g}/\text{day}$ of ACTH is administered for several days to effectively stimulates adrenals than single dose 250 μg .
- This successfully assesses the adrenal insufficiency.

METYRAPONE TEST

- **Measures the ability of the pituitary gland to release ACTH** in response to decreased blood cortisol levels.
- Metyrapone inhibits cortisol production by blocking the conversion of 11-deoxycortisol to cortisol by 11-beta-hydroxylase

Metapyrone stimulation test

- 750 mg of Metapyrone is given mouth every 4 hours for 24 hours. Basal and post Metapyroneadministration plasma levels of 11- deoxycortisol , cortisol and ACTH are measured .
- In normal persons following results are seen
- Plasma 11- deoxycortisol should be $> 7\mu\text{g/dl}$ or 200 nmol/L .

Metapyrone stimulation test

- Plasma ACTH > 75 pgm /ml or 17 pmol/L.
- These response would indicate an normal pitutary .no response would indicate an ectopic ACTH or adrenal tumor because they are not under the pitutary adrenal feedback loop.

CRH STIMULATION TEST

- To differentiate between secondary adrenal insufficiency due to pituitary or hypothalamic dis.
- Results :
 - Pituitary disease – blunted or nil response
 - Hypothalamic lesions – positive response

Tests for mineralocorticoid activity.

PLASMA RENIN AND ALDOSTERONE

- Give an indication of mineralocorticoid activity.
- Adrenal insufficiency
 - Low aldosterone level with high renin

Tests for mineralocorticoid activity.

- Plasma aldosterone
- Levels of Plasma aldosterone are more in morning than evening .potassium intake and sodium restriction indiet also increases aldosterone levels.
- Morning blood levels in supine position are 6-22 $\mu\text{g}/\text{dl}$ in males and 5-33 $\mu\text{g}/\text{dl}$ in females.
- Levels increase in hyperaldosteronism and decrease in hypoaldosteronism.

Management

- **Hormone replacement**
- Life-long replacement therapy
 - Hydrocortisone and 9 α -fludrocortisone
- **Secondary adrenocortical insufficiency**
 - Hormone replacement
 - may also require more definitive treatment e.g. surgical removal of a pituitary tumour.

Disorders of adrenal cortex

ADRENAL HYPERFUNCTION

Primary aldosteronism

CONN'S SYNDROME

- Characterized by **autonomous** excessive production of **aldosterone** by **adrenal glands**
- Presents with HPT, hypokalaemic alkalosis and renal K⁺ wasting

Conn's Syndrome

- Causes:
 - Adrenal adenoma
 - Bilateral hypertrophy of zona glomerulosa cells
 - Adrenal carcinoma
 - Rare cause

Secondary aldosteronism

- Is increased adrenal production of aldosterone in response to non-pituitary, extra-adrenal stimuli
- Increase renin secretion
 - (hyperreninemic hyperaldosteronism)
- Commoner than primary aldosteronism

Conn's syndrome

- **Clinical features:**

- Hypertension : *aldosterone induced Na retention with increase in ECF volume*
- Muscle weakness: *Due to decrease K⁺*
- Muscle paralysis: *severe hypokalaemia*
- Latent tetany and paraesthesiae
- Polydipsia, polyuria and nocturia: *due to hypokalaemic nephropathy*

INVESTIGATION

- **Electrolyte & blood gasses:**
 - Hyponatraemia
 - Hypokalaemia
 - Alkalosis
 - Urinary potassium loss, *level > 30 mmol daily during hypokalaemia*

INVESTIGATION

- **Plasma aldosterone : renin activity ratio**

- Sensitive screening test
- No need to standardize posture

Ratio	Interpretation	Action
<800	Diagnosis excluded	Seek other cause
>1000,<2000	Diagnosis possible	Confirmatory test
>2000	Diagnosis very likely	Establish cause

Diagnosis

- **Perform saline infusion test (sodium loading)**
 - Method :
infusion of 1.25L of 0.9% saline over 2 hrs
 - Result:
plasma aldosterone remains >240 pmol/l confirm Conn's syndrome

Establish cause

- **Plasma Aldosterone level**
 - Method:
 - Morning blood sample (pt stayed recumbent since waking)
 - Second sample after 4 hrs stayed ambulant
- **Standing ↓ renal blood flow → stim renin sec
→ ↑ aldosterone level

Establish cause

- Imaging techniques
 - CT scan
 - MRI
 - Can differentiate adenoma from hyperplasia

CUSHING'S SYNDROME

Adrenal cortex hyperfunction

- Any condition resulting from overproduction of **primarily glucocorticoid (cortisol)**
- Mineralocorticoid and androgen may also be excessive

Pseudo-Cushing's syndrome

- Appear cushingoid and have some biochemical abnormalities of true Cushing's disease
- Causes
 - Severe depression
 - Alcoholism
 - Obesity
 - Polycystic ovarian syndrome

Etiology

- **Excessive cortisol (ACTH dependent)~75%**
 - Pituitary disease
 - Adenoma (90%)
 - Hyperplasia (10 %)
 - Ectopic ACTH syndrome
 - Malignancy - (bronchus, thymus, pancreas, ovary)
 - Ectopic CRH syndrome
 - Exogenous ACTH administration

Laboratory investigations

- There are two diagnostic steps in the investigation of patient suspected of having Cushing's syndrome
 - **Screening test**
 - for identification of Cushing's syndrome.
 - the demonstration of high plasma cortisol level
 - **Identification of cause**

1. Demonstration of increased cortisol

- Assessment of circadian rhythm in cortisol secretion
- 24-Hour urinary free cortisol excretion
- Overnight / low dose dexamethasone suppression test

Laboratory investigations

1. Assessment of circadian rhythm in cortisol secretion.

- Measure 8 am and 11 pm serum cortisol level
 - Normal : Serum value @ midnight is 50% less than value @ 8 am
 - Cushing's syndrome : rhythm is loss
 - Pseudo-Cushing : normal circadian.

Urinary free cortisol

- Urinary cortisol secretion is higher in day time (7.00 am to 7.00 pm) than night (7.00 pm to 7.00 am) hence a properly collected 24 hours urine sample is required.
- Urine cortisol levels range from 10-100 $\mu\text{g}/\text{day}$.
- Levels of urine cortisol increase in hyperadrenalism and decrease in hypoadrenalism.

Laboratory investigations

2. Measuring 24-hour urinary free cortisol

Level (umol/ 24 h)	Interpretation
< 300	Normal
300 - 700	Severe depression Stress
> 700	Diagnostic of Cushing's syndrome

Dexamethasone suppression test

- Screening test
- Plasma cortisol is measured after 1 mg of oral dexamethasone administration, the previous night before test.
- The next morning 8.00 am cortisol level of less than 5ug /dl is considered normal.
- Higher level is indicative of hyperfunction.

Laboratory investigations

3. Low dose Dexamethasone suppression test :

- 0.5 mg Dexametason (oral) given 6 hourly for 2 days
 - blood for plasma cortisol collected 6 hour after last dose
 - urine for UFC is collected before & on the 2nd day of Dexa
-
- Result:
 - UFC suppress by 50% (< 70nmol/24h) normal
 - plasma cortisol suppress < 140 nmol/L pseudo-Cushing
 - no suppression of UFC & Pl. cortisol Cushing's synd

Ufc – urinary free cortisol

2. Elucidation of the cause

- Plasma ACTH

- Normal < 50 ng/L

- Low – adrenal causes

- Elevated

- Slight – pituitary dependent Cushing's

- Gross – ectopic secretion of ACTH

Elucidation of the cause

- CRH Test

- Differentiate ectopic ACTH secretion and Cushing's disease.
- Cushing's disease – plasma ACTH increases 50% over baseline and cortisol increase by 20%
- Ectopic ACTH or adrenal tumour – no response

Elucidation of the cause

- **Imaging**

- CT scan of adrenal gland: TRO adrenal tumor
- MRI of pituitary gland: majority microadenoma (< 10mm). MRI reveal lesion in 50 - 60% of cases
- CT scan/MRI of thorax & abdomen: ectopic ACTH producing tumor

Treatment

- Depend of Cushing's syndrome depends on the etiology:
 - Adrenal adenoma
 - Adrenal Carcinoma – resection
 - Cushing's disease - transphenoidal hyposectomy
 - Drug (block cortisol synthesis) - metyrapone

Thank u