

# **ADRENAL FAILURE (INSUFFICIENCY) – DIAGNOSTIC TESTS**

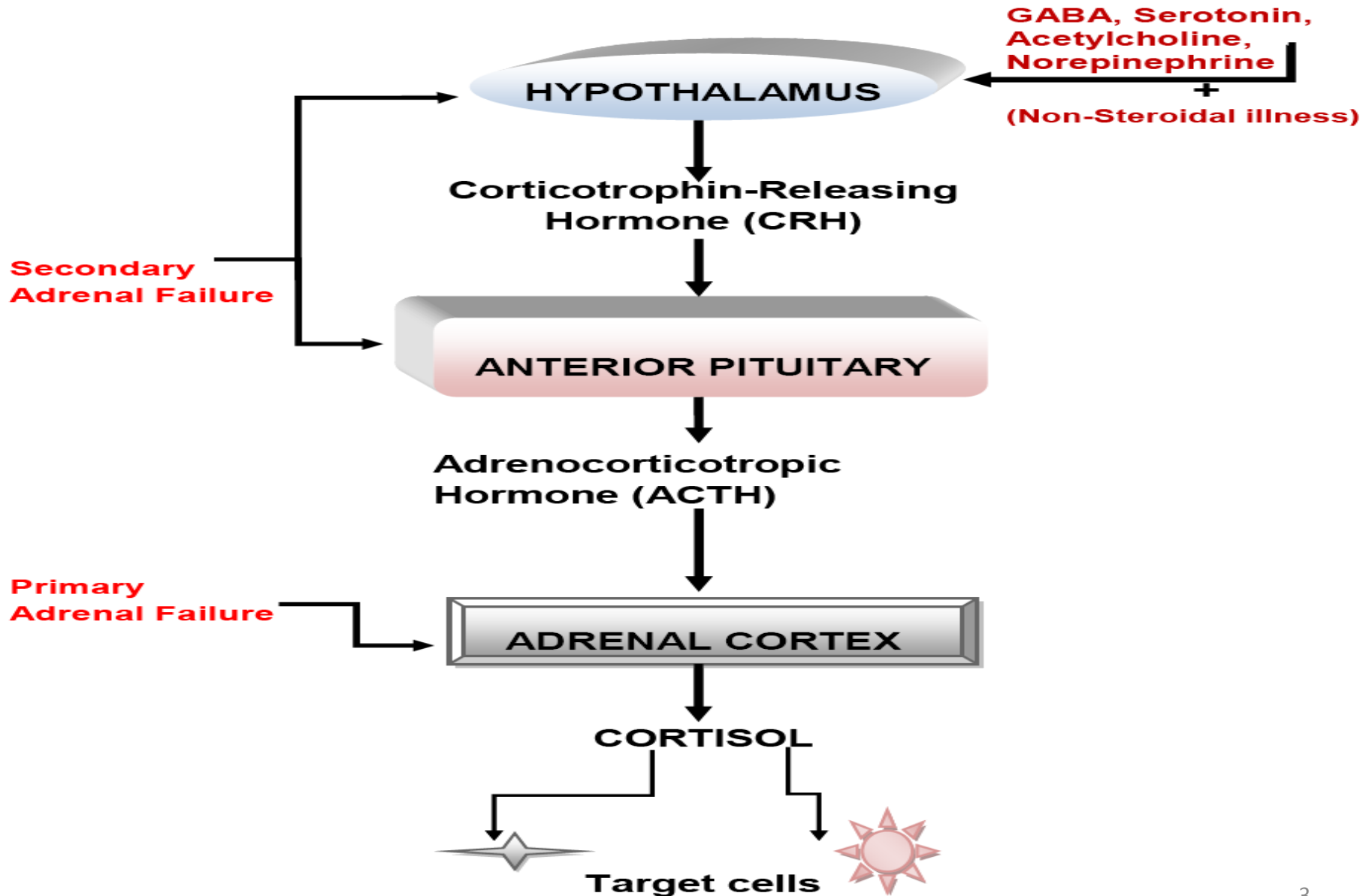
**UNIVERSITY OF PAPUA NEW GUINEA  
SCHOOL OF MEDICINE AND HEALTH SCIENCES  
DISCIPLINE OF BIOCHEMISTRY AND MOLECULAR BIOLOGY  
BMLS III & BDS IV**

**VJT**

## What is the basic classification of Adrenal Failure?

- Basic classification of Adrenal failure (Adreno-Cortical Hypo-function) depends on location of lesion: (**Fig. 1**)
- **Primary Adrenal Failure** or Primary Adrenal Insufficiency (Addison's Disease) is due to:
  - **Failure of Adrenal Gland** due to destruction of Gland;
  - Cortisol and Aldosterone production may be affected,
- **Secondary Adrenal Failure** or Secondary Adrenal Insufficiency:
  - **Hypothalamic or Pituitary disease** leading to deficiency of ACTH (Corticotrophin) production;

**Fig. 1: HPA-Axis showing location of lesions**



## What are some causes of Primary Adrenal Failure?

- Several causes including:
- Infective (Tuberculosis, Meningococcal, HIV, etc)
- Autoimmune Adrenalitis,
- Metastasis (from Lung and Breast Carcinomas),
- Hemorrhage,
- Metabolic failure, insufficient hormone production, caused by:
  - Congenital Adrenal Hyperplasia (CAH),
  - Enzyme inhibitors, such as Metyrapone,
  - Cytotoxic agents (e.g. Etomidate),

## What are some signs and symptoms of Primary Adrenal Failure?

- Some **non-specific signs** and symptoms include:
- Weakness, Abdominal Pain, Nausea, Weight Loss, Shock, Hypotension, Acid-Base Disturbance, Lack of Libido, Loss of body hair in women, Psychiatric changes;
- **Some specific signs and symptoms include:**
  - **Hypoglycemia with Hyponatraemia,**
  - **Hyperkalemia,**
  - **Raised Serum Urea levels,**
  - **Hyper-pigmentation** (affecting Buccal mucosa),
  - Scars, Skin creases
- These conditions are life threatening and requires urgent investigation if suspected;

## What are some signs and symptoms of Secondary Adrenal Insufficiency?

- Some signs and symptoms of Secondary Adrenal Insufficiency may be identical to Primary Adrenal Insufficiency,
- **Hyper-pigmentation** does not occur in Secondary Adrenal Insufficiency (**WHY?**)
  - Because of Insufficient production of ACTH and other products of Proopiomelanocortin (POMC) metabolism {i.e., Melanocyte Stimulating Hormones (MSH)};

## What lab results are indicative of Primary Adrenal Insufficiency?

- Biochemical results indicative of Primary Adrenal Insufficiency include:
  - **Hyponatraemia,**
  - **Hyperkalemia,**
  - **Elevated Serum Urea,**
  - **Hyper-pigmentation;**

## What is the Biochemical basis for Hyponatraemia, Hyperkalemia, Elevated Serum Urea and Hyper-pigmentation in patients with Primary Adrenal Insufficiency?

- Lack of Aldosterone leads to pathological Sodium loss via Kidneys,
  - Resulting in contraction of Extracellular Fluid Volume (Hypovolaemia), causing Hypotension & **Pre-renal Uremia**,
  - Patient may develop **Sodium depletion** and **Potassium Retention** due to Aldosterone Deficiency,
- Hypovolaemia and Hypotension **stimulate AVP Secretion**, thus causing **Water Retention**,
- Absence of Cortisol impairs ability of Kidneys to excrete water, which leads to **Hyponatraemia**,



- Overall effect causes **reduction in Total Body Water**, (dehydration) reflected by **increase in Serum Urea**,
- Absent of **Cortisol** causes failure in **Negative Feedback control**, resulting in **excessive** secretion of **ACTH** from Anterior Pituitary,
- ACTH structure contains part of Amino Acid sequence of Melanocyte-stimulating hormone (**MSH**),
- **Excessive secretion ACTH** causes **darkening of skin and mucus membranes**, resulting from the action of MSH;

# Screening and Diagnostic Tests for Adrenocortical Insufficiency (Adrenal Failure)

## What basal lab tests are done on a patient suspected of having Adrenal Insufficiency?

- Before the patient is given any medication (Cortisol), blood must be collected for Basal measurements of:
  - Plasma Urea,
  - Electrolytes,
  - Glucose,
  - Serum Cortisol,
  - Plasma ACTH concentration;
- Patient intake of Sodium must be monitored to ensure adequate intake, whilst investigations proceed,

## Important points with respect to Cortisol and ACTH measurements

- Normal Serum [**Cortisol**] at **8a.m.**, or Normal 24-hour Urinary **Free Cortisol**, does not exclude **Primary Adrenocortical Insufficiency (Why?)**
  - Because patient may be able to maintain Normal Basal output of Cortisol but **is unable** to secrete adequate amounts of Cortisol in response to Stress;
- Serum [Cortisol] below **50nmol/L** at 8 a.m. is strong presumptive evidence for **Primary Adrenal Failure**;

- Diagnosis of Primary Adrenal Failure is unlikely, if Serum [Cortisol] is **550nmol/L** or more at 8 a.m. (in absence of Steroid Therapy),
- Diagnostic accuracy for Primary Adrenal Failure is greatly improved when **Serum [Cortisol] and [ACTH]** are measured at the same time; **Why?**
  - Because Low Serum [**Cortisol**] < **200nmol/L**, and Raised Serum [**ACTH**] > **200nmol/L** may be diagnostic of **Primary Adrenal Failure**,

## List some Biochemical tests used for Screening and Diagnosis of Adrenal Failure

- Biochemical tests for diagnosis of Adrenal failure:
  - Short Cosyntropin (Synacthen, Cortrosyn or Tetracosactrin) Test,
  - Depot (Long) Synacthen test,
  - Prolonged Cosyntropin Stimulation (Rose) test,
  - Corticotrophin Releasing Hormone (CRH) stimulation test,
  - Rapid Synacthen stimulation test,

## What is the procedure for Short Cosyntropin test?

- Short Cosyntropin (Synacthen, Cortrosyn or Tetracosactrin) test indicates ability of Adrenal Cortex to respond to ACTH,
  - ***Cosyntropin is Synthetic 1 – 24 analogue of ACTH,***
  - ***Trade name for Cosyntropin is Synacthen or Cortrosyn,***

## Procedure for Short Cosyntropin test includes the sequence:

- Measure Baseline Plasma [Cortisol], [Aldosterone]
- Patient is given **0.25mg** Synacthen as Intravenous bolus or as Intra-muscular Injection,
- Measure Plasma [Cortisol] & [Aldosterone] again after 30 minutes,
- Blood samples for Aldosterone can be held until results of Cortisol response are known,

## IMPORTANT TO NOTE

- Normally, Baseline Plasma [Cortisol] should be within Reference Range,
  - **280 – 720nmol/L at 08.00 am to 10.00 am**
  - Acceptable Baseline Plasma [Cortisol] > 225nmol/L
- There should be an increment of more than 200nmol/L Plasma [Cortisol] after Synacthen,
- Final Plasma [Cortisol] should be greater than 500nmol/L,



- Aldosterone response in Synacthen test may be blunted or absent in patients with Primary Adrenal Failure;
- In Secondary Adrenal Failure, Aldosterone response is normal (an increase of about twice the baseline value) in Synacthen test (**WHY?**),
  - Because the RAA-axis is not affected by decreased production of ACTH,

## How are the results of Short Cosyntropin test interpreted?

- **Criteria for interpretation of Short Cosyntropin test:**
- Three criteria should be met for normal response, these criteria are:
  - **Baseline Plasma [Cortisol] should be  $> 225\text{nmol/L}$**
  - **Final Plasma [Cortisol] should be  $> 500\text{nmol/L}$ ,**
  - **Increment in [Cortisol] should be at least  $200\text{nmol/L}$ ,**
- For a patient to be declared as normal all Three Criteria must be satisfied;

- Normal response to Short Synacthen test excludes Primary Adrenocortical Insufficiency;
- Failure to meet any of the criteria indicates Adrenocortical Inadequacy;
- **Elevated Plasma [ACTH]** can be used to **confirm diagnosis** of Primary Adrenal Insufficiency in a patient with abnormal response to Short Synacthen test,

## What addition test should be carried out if the results from the Short Synacthen test are equivocal (unclear)?

- Patients with equivocal responses to Short Synacthen test may be re-tested after Stimulation of Adrenal Cortex with ***depot-Synacthen***,
- This longer acting material (1.0 mg) should be given IM daily for three days;
- On fourth day, Short Synacthen test should repeated,
  - If normal criteria for Short Synacthen test are fulfilled on the second testing, then Adrenal Insufficiency is not of Primary origin,
  - Such a result points towards Secondary Adrenocortical Insufficiency,

## What are some factors that can affect Short Synacthen test?

- Some factors that can affect Short Synacthen test to the point of invalidating the tests include:
  - Severe Emotional Stress,
  - Treatment with Glucocorticoids within 12-hours prior to Synacthen test,
  - Taking of Estrogen-containing Oral Contraceptives,

## What is the use and procedures for Prolonged Cosyntropin Stimulation (Rose) test?

- Prolonged Cosyntropin-Stimulation (Rose) test is used to differentiate Primary from Secondary Adrenal Failure;
- **Procedure for the test:**
  - Measure Baseline Plasma [Cortisol],
  - Measure 24-hr Urinary [17-Hydroxycorticosteroids] (17-OHCS),
  - Patient is given infusion of 0.25mg Cosyntropin for 48hrs, after the first 24hrs measurements are repeated;
  - Measure Plasma [Cortisol],
  - Measure 24-hr Urinary [17-OHCS],
  - At the end of infusion repeat the measurements,

## How are results of the Prolonged Cosyntropin Stimulation (Rose) test interpreted?

- In **Primary** Adrenal Insufficiency, no change is seen in Plasma [Cortisol] or 24-hr Urinary [17-OHCS] **WHY??**
  - Because Cortisol is not produced,
- In **Secondary** Adrenal Insufficiency, Incremental Increase occurs over the course of the Infusion **WHY??**
  - Because the problem is **not Primary**, Cosyntropin action stimulates Adrenal Cortex, which then produces Cortisol,
  - Results indicate that **Adrenal Cortex has undergone Atrophy** because of **Insufficient ACTH stimulation**; however, with **longer stimulation**, the Adrenal Cortex is capable of functioning,

# Diagnosis of Secondary Adrenocortical Insufficiency

## How can Secondary Adrenocortical Failure be diagnosed?

- **Low Plasma [Cortisol] and Low Plasma [ACTH]** indicate diagnosis of Adrenocortical Insufficiency **Secondary** to Hypothalamic or Pituitary disease,
- In such cases, whilst the Atrophied Adrenocortical cells may fail to respond in Short Synacthen test, the Adrenal Cortex can become responsive over a longer period of stimulation using **Depot (long) Synacthen test**,



## Procedure for the Depot (Long) Synacthen Test

- Measure Baseline Serum [Cortisol],
- Patient is administered Depot Synacthen (1.0 mg) Intramuscularly on three successive days,
- On each of the Three days Serum [Cortisol] must be measured between Five to Eight hours after administration of Depot Synacthen,
- (In each instance [ACTH] can also be measured)

## How are the results interpreted?

- In **Primary** Adrenocortical Insufficiency, Serum [Cortisol] will NOT INCREASE above 600nmol/L at 5 to 8 hours after the Third injection,
- In **Secondary** Adrenocortical Insufficiency, Stepwise increase in Serum [Cortisol] will be apparent after successive administration of the Depot Synacthen,

## IMPORTANT TO NOTE

- Poor responses to Prolonged Synacthen tests may occur in patients with Hypothyroidism (both Primary and Secondary),
- In patients with Hypothyroidism, Adrenal Function cannot be satisfactorily assessed until the Thyroid Deficiency has been corrected,
- Once a decision has been made as to whether Adrenal Insufficiency is Primary, or Secondary, the appropriate Imaging Technique should be used to rule out other treatable causes,