"اللَّهُمَّ لا سَهْلَ إلاَّ ما جَعَلْتَهُ سَهْلاً، وأنْتَ تَجْعَلُ الحَزْنَ إذَا شِئْتَ سَهْلاً "

Cushing syndrome

Biochemistry Team 437

Color index:
Doctors slides
Doctor's notes
Extra information
Highlights

Revised By
Hadeel Awartani Abdullah AlOmar

Endocrine block

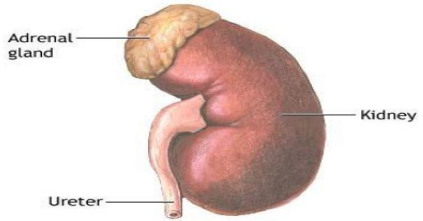
Objectives:

- To identify physiological and biochemical characteristics of Cortisol.
- To understand the diagnostic algorithm for Cushing's Syndrome.
- To understand the interpretation of laboratory and radiological investigations for diagnosis of Cushing's Syndrome.

Adrenal Gland

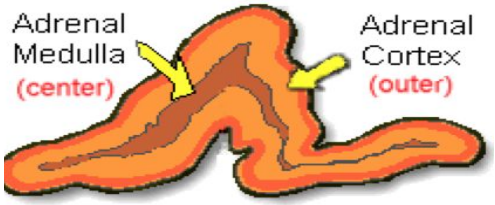
Anatomy

The adrenal gland is situated on the anterosuperior aspect of the kidney



Histology

The adrenal gland consists of two distinct tissues of different embryological origin, the outer cortex and inner medulla.



Zona **G**lomerulosa	Zona **F**asciculata	Zona **R**eticularis
The outermost zone → aldosterone (the principal mineralocorticoid).	The deeper layer of the cortex → glucocorticoids – mainly cortisol (95%)	The deeper layer of the cortex → sex hormones

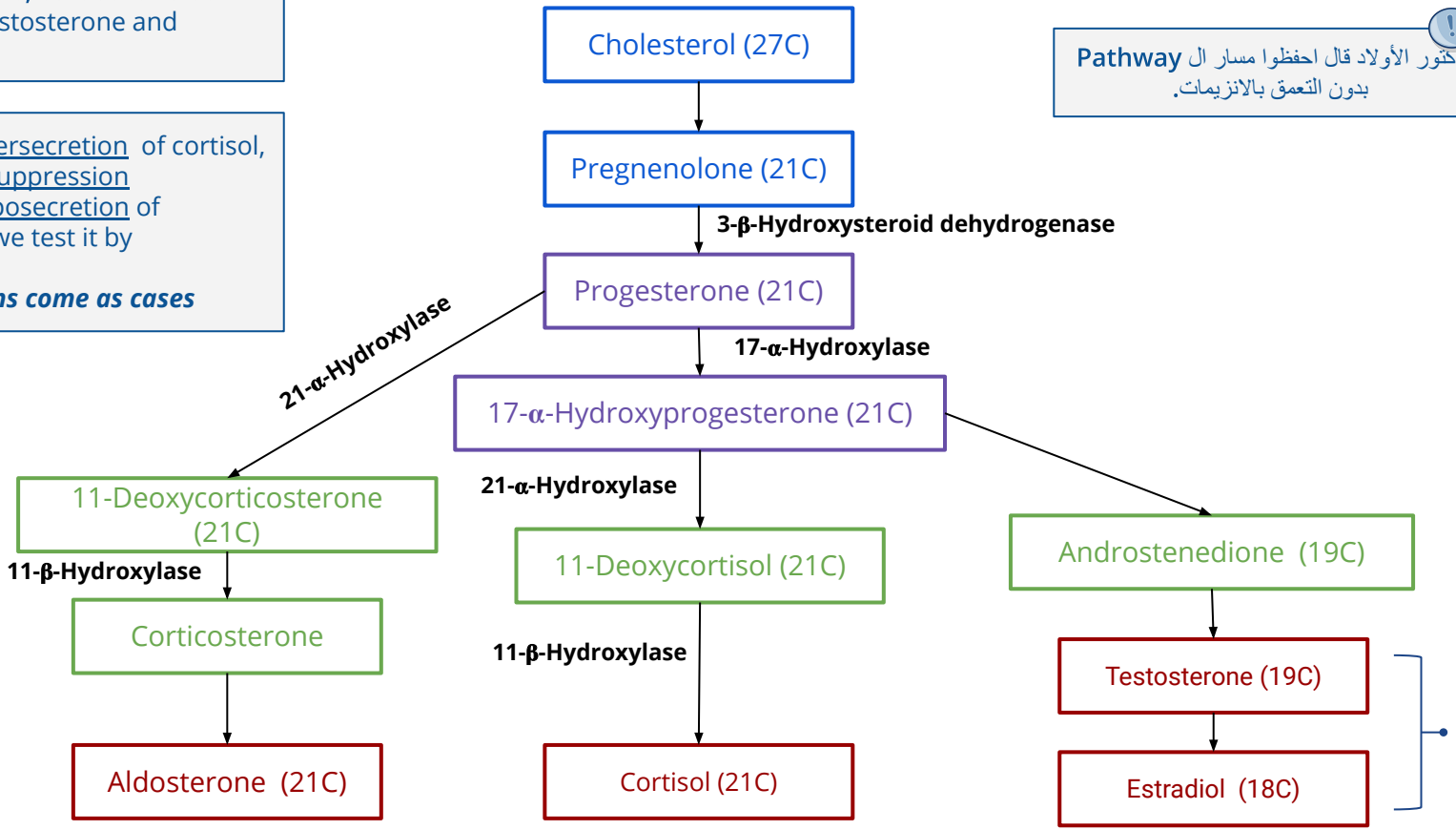
Steroid Hormone Synthesis

All you need to know from this slide is that **cholesterol is the precursor for aldosterone, cortisol and sex hormones** (testosterone and estradiol)

General guide

Cushing: Hypersecretion of cortisol, we test it by suppression
Addisons: hyposecretion of aldosterone, we test it by stimulation
Most questions come as cases

دكتور الأولاد قال احفظوا مسار ال Pathway بدون التعمق بالانزيمات.



IMPORTANT!

Hypothalamic-Pituitary-Adrenal (HPA) Axis

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- The hypothalamus secretes **corticotropin-releasing hormone** (CRH) which stimulates the anterior pituitary gland to synthesis and release **ACTH**.
- ACTH acts on the zona fasciculata cells → release of **glucocorticoids** (Cortisol).

- When we have any stimulus like stress (emotional or surgery) there will be stimulation of hypothalamus to secrete CRH to stimulate anterior pituitary to secrete ACTH, now ACTH will stimulate the adrenal cortex 2nd layer (zona fasciculata) to secrete the cortisol. That is why we need to keep this axis intact.
- Negative feedback happens when I have plenty of cortisol so it will suppress the pituitary and the hypothalamus secretions.

Glucocorticoid Functions

- It is a catabolic enzyme
- Main metabolic function is hyperglycemia
- Recall: Gluconeogenesis also happened in kidney

Glucocorticoids have widespread metabolic effects on carbohydrate, fat and protein metabolism.

Upon binding to its target, cortisol enhances metabolism in several ways:

In the liver[1]:

- Cortisol is an insulin antagonist[2] and has a weak mineralocorticoid action:
  - ↑↑ Gluconeogenesis → production of glucose from newly-released amino acids and lipids
  - ↑↑ Amino acid uptake and degradation[3]
  - ↑↑ Ketogenesis.

In the adipose tissue:

- Cortisol → ↑↑ Lipolysis through breakdown of fat.

In the muscles[4]:

- Cortisol →↑↑ proteolysis and amino acid release.

Conserving glucose:

- By inhibiting uptake into muscle and fat cells.

[1] The most important effect
[2] That is why cushing → hyperglycemia
[3] That's why we will have high level of Urea
[4] Effect on muscle → catabolic, that is why patients with cushing have thin limbs

Insulin resistance → hyperglycemia

Regulation of ACTH and Cortisol Secretion

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1 Negative feedback control:

ACTH release from the anterior pituitary is stimulated by hypothalamic secretion of corticotrophin releasing hormone (CRH).

- CRH → ↑ ACTH → ↑[Cortisol]
- ↑[Cortisol] or synthetic steroid suppress CRH & ACTH secretion[1]

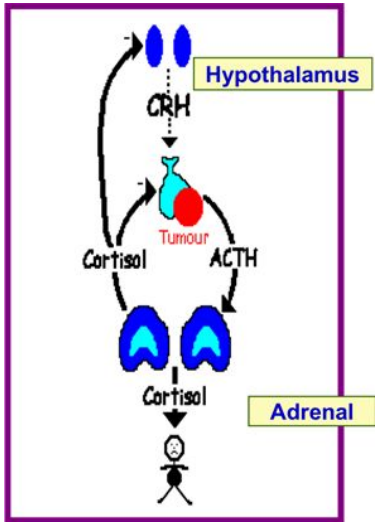
2 Stress:

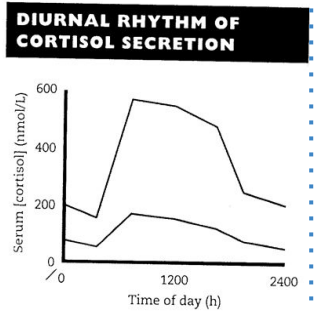
(e.g. major surgery, emotional stress) Stress → ↑↑ CRH & ACTH → ↑↑ Cortisol

Usually before we draw blood for cortisol test we let the patient rest for a while to make sure the results aren't altered by emotional stress.

3 The diurnal rhythm of serum cortisol:

- Highest Cortisol level in the morning (8 - 9 AM).
- Lowest Cortisol level in the late afternoon and evening (8 - 9 PM).





The diurnal rhythm of cortisol secretion; the area between the curves represents values that lie within the reference range.

- Diurnal rhythm is one of the tools that we can use to rule in or rule out cushing
- Cortisol should be double in the morning than it is in the evening (half of the morning value at night)
- Loss of diurnal rhythm is one of the Cushing syndrome investigations "high all the time, night and day"

[1] If I give exogenous (synthetic) cortisol "dexamethasone" , the cortisol will be low because of negative feedback.

IMPORTANT!

Plasma Cortisol-binding Globulin (CBG)

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- In the circulation, glucocorticoids are mainly protein-bound (about **90%**)[4], chiefly to **CBG** (transcortin).[1]

Increases	Decreases
in pregnancy and with estrogen treatment (e.g. oral contraceptives)[2].	in hypoproteinemic[3] states (e.g. nephrotic syndrome).

- The biologically active fraction of cortisol in plasma is the free (unbound) component.

[1] **We test the total cortisol** in the serum (bound and unbound)
[2] Gives us false high cortisol on a test
[3] Hyperproteinuria
[4] cortisol is steroid hormone so it is hydrophobic that is why it need carrier

Cortisol and ACTH Measurements

Serum (Cortisol) and plasma (ACTH):

- Samples must be collected (without venous stasis)[3] between 8 a.m. and 9 a.m. and between 10 p.m and 12 p.m. because of the diurnal rhythm.
- Temporary ↑↑ in these hormones may be observed as a response to emotional stress[1].

Urinary Cortisol excretion:

- Cortisol is removed from plasma by the liver metabolically inactive compounds excreted in urine mainly as conjugated metabolites (e.g. glucuronides).
- A small amount of cortisol is excreted unchanged in the urine (UFC)[2].
- In normal individuals: Urinary free cortisol (UFC) is < 250 nmol/24h.

- In the **serum we measure the total In the urine we measure the free** one only.
  [1] Because it can increase in CBG
  [2] The one we mesure, we do not measure the metabolites only UFC.
- Cortisol is hydrophobic So it is usually bound to glucuronide To become hydrophilic and more easily excreted in the urine (similar to bilirubin) This form of cortisol is inactive and not measured in labs
  [3] When we put the tourniquet to withdraw blood, if we left it for a long time it leads to protein stasis → increase CBG
  [4] We need to exclude these causes to diagnose cushing syndrome

Causes of Elevated Cortisol[4]

Increased cortisol secretion:

- Cushing's syndrome
- Exercise (severe)
- Stress, Anxiety, Depression
- Obesity
- Alcohol abuse
- Chronic renal failure

Increased CBG:

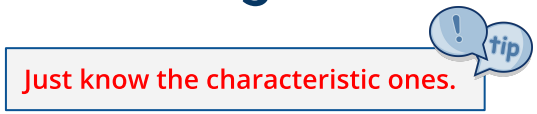
- Congenital
- Estrogen therapy
- Pregnancy

Psudeocushing is caused by:
pregnancy,estrogen replacement,renal failure,Alcohol abuse,Etc

Symptoms of Cushing Syndrome

- Weight gain, central obesity.
- Buffalo's hump.
- Moon face
- Excessive sweating
- Atrophy of the skin and mucous membranes
- Purple striae on the trunk and legs
- Proximal muscle weakness (hips, shoulders)
- Hirsutism
- The excess cortisol may also affect other endocrine systems→ ↓libido, amenorrhoea and infertility
- Patients frequently suffer various psychological disturbances ranging from euphoria to frank psychosis.

Signs



- Loss of diurnal rhythm of cortisol and ACTH.
- Hypertension (due to the aldosterone - like effects)
- Hyperglycemia or diabetes due to insulin resistance.
- Hypokalemic alkalosis
- ↑ Protein metabolism.
- Impaired immunity.

Investigations of Suspected Adrenocortical Hyperfunction

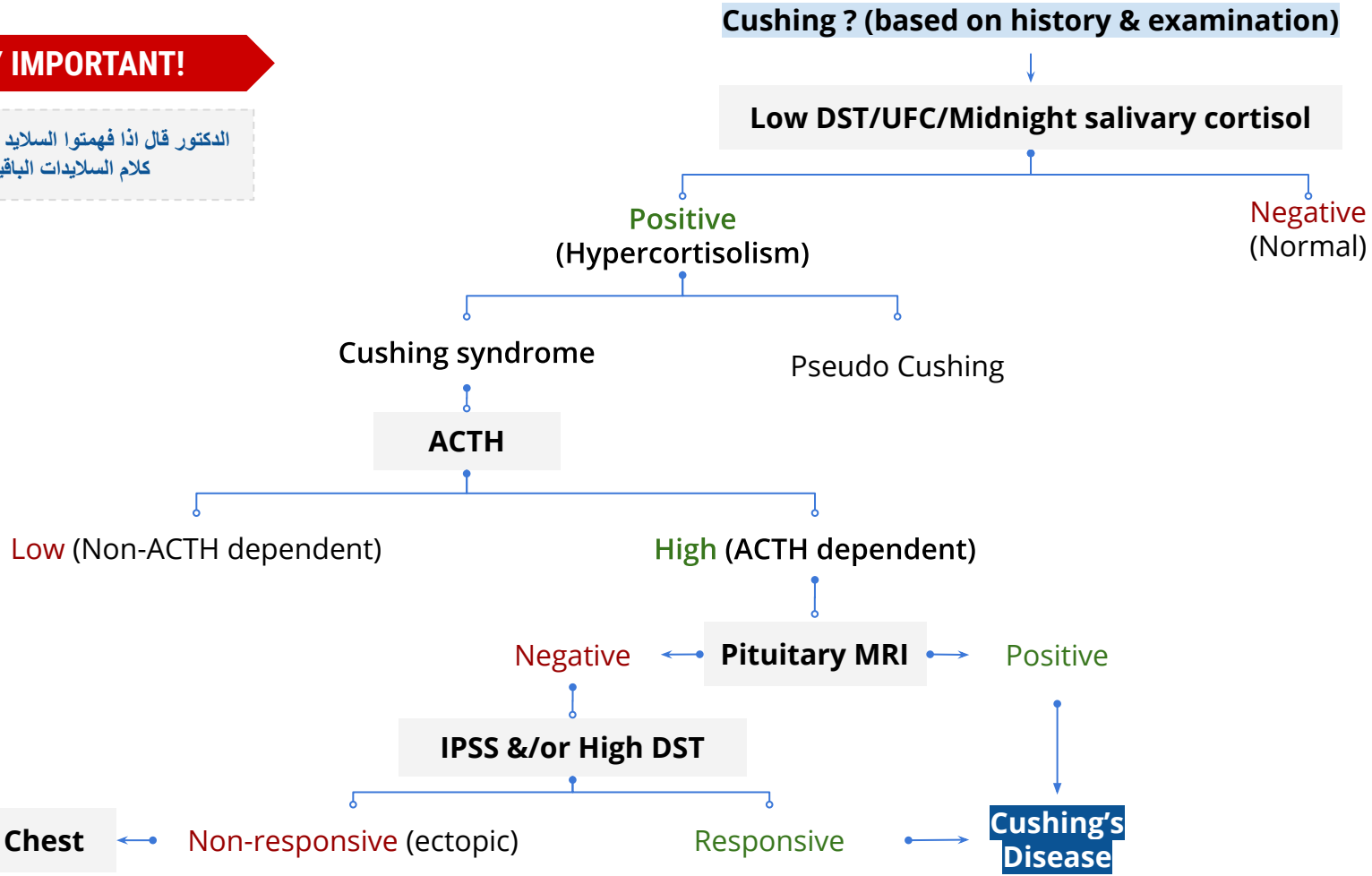
- **Screening and confirmatory tests*:** to assess the clinical diagnosis of adrenocortical hyperfunction.

- **Tests to determine the cause: to ascertain:** (Make certain)
  1) The site of the pathological lesion (adrenal cortex, pituitary or elsewhere?)
  2) The nature of the pathological lesion.

To see if there is hypercortisolism (most likely cushing cases) in the outpatient clinic.

VERY IMPORTANT!

الدكتور قال اذا فهمتوا السلايد هذا هو نفسه كلام السلايدات الباقية.



No need to know numbers

Screening Tests

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I suspect the patient to have hypercortisolism, I try to stimulate it.
- If the patient was normal, the cortisol will be suppressed
- If the patient has Cushing, it will be not be suppressed "high cortisol"> do more investigations

1) **Low-dose DST[1]:**
   - **Procedure:**
     1 mg dexamethasone (DXM) administered at 11-12 PM the night before attending the clinic. Serum cortisol is measured at 8-9 AM.
   - **Result:**
     Cortisol < 50 nmol/L (suppression)→ exclude hypercortisolemia (Cushing Syndrome)
   - **Precautions:**
     Drugs that induce hepatic microsomal enzymes (Phenobarbitone & phenytoin) → ↑ DXM metabolism and ↓ DXM blood level to achieve CRH suppression (false diagnosis of Cushing)[2]
2) **24 Hour urinary free cortisol:**
   - **Result: Cortisol < 250 nmol/day** →exclude Cushing Syndrome.
   - **Disadvantage:** incomplete collection of urine →a false-negative result.
3) **Midnight Salivary Cortisol.**
   Result: Cortisol < 100 ng/dL →exclude Cushing Syndrome.

[1] Dexamethasone is a synthetic cortisol
[1] Normally. Increase in cortisol will lead to negative feedback on CRH & ACTH and a decrease in cortisol, which is what happens if the patient has a normal axis, and you give him 1 Mg of DST (low dose) . but if the patient has cushing, his cortisol is already very high. So 1 Mg wont do anything to suppress the axis.
[2] DXM metabolised fast so no negative feedback can occur and no suppression

Confirmatory Tests

[1] Dr.rana said it is important to know this information and also you should know what are the three screening tests.

- Positive results of **at least two screening tests** would confirm the clinical diagnosis[1] .
- Further investigations are required.

Tests Used to Determine the Cause of Cushing's Syndrome

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To differentiate ACTH-dependent from ACTH-independent:

- Plasma ACTH (Diurnal rhythm)

To distinguish between ACTH-dependent causes (Pituitary Vs Lung):

- High-dose DST.
- Inferior Petrosal Sinus Sampling[1] (IPSS, invasive procedure, we don't like to use normally)

Radiological tests:

MRI of pituitary and ultrasound or CT of adrenals.

Questions by Dr.Rana!

1- If i want to differentiate between ACTH-dependent from ACTH-independent what do I measure?

ACTH

2- If I want to know the location of the neoplasm what do I measure?

High-dose DST-Inferior Petrosal Sinus Sampling-MRI of pituitary and ultrasound or CT of adrenals

[1]It measure ACTH from the vein that drain the pituitary at the sametime they measure ACTH from the peripheral blood then compare it

Causes of adrenocortical hyperfunction (Cushing's syndrome)

ACTH - dependent:

1) Pituitary ACTH >90% (Cushing's disease).
2) Ectopic ACTH by neoplasms[1] <10%.

ACTH - independent:

1) Glucocorticoid therapy.
2) Adrenal tumor <20% (adenoma or carcinoma).

[1] Mainly found in the lung, the question might mention respiratory distress, weight loss to indicate cancer
[2] No ACTH dependent primary
[3] (ACTH secreting neoplasm in the LUNG) it is very high because it is uncontrollable.

Plasma ACTH

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It should be measured on blood specimens collected at 8-9 a.m. and 8-9 p.m.

Undetectable[2]:

Functional adrenal tumor → confirmed by abdominal CT scan to detect adrenal mass

↑↑ACTH:

Cushing disease (pituitary dependent)

↑↑↑↑ACTH:

Ectopic (non endocrine) that produce ACTH[3]
ACTH is sky high= three fold the normal range (its a hint)

High Dose DST

Also called prolong DST. it is used In patient confirmatory to know the location. It need to patient stay in the hospital

It is used to distinguish Cushing's disease from ectopic ACTH secretion.

- 2 mg DXM six-hourly for 48 hours to suppress cortisol secretion.
- Basal (pre-DXM) serum cortisol or 24-hour urine free cortisol is compared with the results at the end of the 48-hour period.
- Suppression is defined as a fall to less than 50 % of basal value.
- About 90 % of patients with Cushing's disease show suppression of cortisol output.
- In contrast, only 10% of patients with ectopic ACTH production (or with adrenal tumors) show suppression.

Why patients with cushing disease (pituitary) respond to high dose DST while other conditions does not? When patient have pituitary adenoma, pituitary hypertrophy(hyperplasia) so it will secrete a lot of ACTH, hence small dose of DXM will not cause negative feedback. But if I gave DXM continuously for 2 days it will start gradually to respond.

Other Blood Tests

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The following blood tests are commonly performed for patients suspected to have Cushing's syndrome:

- Full blood count
- Blood glucose
- Blood electrolytes and pH
- Renal function tests
- Liver function tests

Case Study

58 years old man was admitted with weight loss (think cancer) and respiratory distress (think lung tumor). He had increased pigmentation (indicate ACTH high) and BP was 140/80.

Lab tests:		
Urea	8.6	(2.5-7 mmol/L)
Sodium	144	(135-145 mmol/L)
Potassium	2.0	(3.5-4.5 mmol/L)
Cortisol	1650	(150-550 nmol/L)
Post overnight DMX	1530	(<50nmol/L)

→ The screening test (low dose)

Diagnosis: Ectopic tumor (non endocrine) that produce ACTH

Further investigation revealed the following

DMX suppression test	Basal	after 48 h	after 48h
		0.5 mg qid	2.0 mg qid
Serum cortisol	1350 No suppression	1420	1100
	8 am	22.00 pm	
Plasma ACTH (ng/L)	220	180	

→ Ref. range: 7-51

Summary

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INVESTIGATIONS OF SUSPECTED ADRENOCORTICAL **HYPERFUNCTION :**			
Screening tests:	**1. Low-dose DST:**	**2. 24- hour urinary free cortisol:**	**3. Midnight Salivary Cortisol :**
	1 mg dexamethasone → Cortisol < 50 nmol/L (suppression)o exclude hypercortisolemia	Cortisol < 250 nmol/day exclude Cushing Syndrome.	Cortisol < 100 ng/dL o exclude Cushing Syndrome.
Confirmatory tests:	Positive results of at least two screening tests would confirm the clinical diagnosis.		
Tests to determine the cause :	**1. To differentiate ACTH-dependent from ACTH-independent :**	**2. To distinguish between ACTH-dependent causes (Pituitary Vs Lung):**	**3. Radiological tests:**
	Plasma ACTH : A. ACTH - dependent: 1.Pituitary ACTH >90% (Cushing's disease) (High ACTH). 2.Ectopic ACTH (very high ACTH) B. ACTH - independent: 1. Glucocorticoid therapy. 2. Adrenal tumor (undetectable ACTH)	A. High-dose DST (About 90 % of patients with Cushing's disease show suppression of cortisol output). B. Inferior Petrosal Sinus Sampling.	A. MRI of pituitary. B. ultrasound or CT of adrenals.

Take Home Messages

- Initial screening for Cushing by 24 h urine free cortisol, low-dose dexamethasone suppression test or midnight Salivary Cortisol.
- Confirmatory tests for Cushing by getting positive results of at least two of the screening tests.
- Tests to determine the cause of Cushing: Plasma ACTH, high-dose dexamethasone suppression test, Inferior Petrosal Sinus Sampling and radiological investigations.
- ACTH-dependent Cushing: due to pituitary causes (Cushing's disease) and due to ectopic production of ACTH.
- ACTH-independent Cushing: due to adrenal adenoma or carcinoma and due to steroid therapy (iatrogenic).

MCQs:

Q1: 29 year old male patient, came to the clinic with a history of increased facial hair over the past year, and purple striae on his abdomen. Lab results revealed cortisol level to be 780nmol/L (Normal is 150-550). Plasma ACTH was 1200 (normal is 7-51)
There was no hypoglycemia upon insulin induction, High dose DST showed no response & CRH test also showed no response.
Which of the following might be the correct diagnosis?

A. Secondary adrenal insufficiency
B. Pseudo-Cushing
C. Ectopic ACTH secreting tumor
D. Cushing's disease

Q2: patient came to you with hypercortisolism you suspected Cushing's ACTH levels were high a MRI of the pituitary gland is negative what other test do you need to do to confirm your diagnosis?

A. CT chest
B. IPPS/high DST
C. No need for more tests
D. Both A&B

Q3: Which one of the following is correct in a patient with an ectopic ACTH secreting tumor?

A. Low dose dexamethasone suppresses cortisol secretion.
B. The tumor ACTH secretion is more sensitive to Negative feedback.
C. High dose dexamethasone shows no suppression of cortisol.
D. MRI shows enlarged pituitary gland.

3-C
2- B
1- C

Girls team

- مجد البراك
- لجين عبدالله

Boys team

- انس القحطاني
- عبدالملك الشرهان
- سلطان الناصر
- عبدالله العنقري
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