Thyroid Function Test
Learning objectives

✓ Normal functioning of thyroid gland including production and regulation of thyroid hormones.
✓ Various parameters used to assess thyroid function.
✓ Thyroid function tests in various thyroid dysfunctions.
✓ Analytical and radiological methodologies to assess thyroid functions.
A brief review of Thyroid gland

• Butterfly shaped gland located in front of neck.

• Weight: 15 – 20 gm

• Thyroid follicle is the secretory unit

• Two-in-one gland:
  - Thyroid follicular cells secrete thyroid hormone
  - Parafollicular (C) cells secrete calcitonin
THYROID SYNTHESIS AND TRANSPORT
Thyroid Follicular Epithelial Cell

Iodine Active Transport → \( I^- \) → Oxidation and organification of iodine by performed by Thyroid Peroxidase → \( I^- \) → \( I_2 \)

T4/T3 release into circulation

Processed Thyroglobulin Lysosomal Degradation and release of T4, T3, MIT, and DIT

Thyroglobulin

Coupling of MIT and DIT by Thyroid Peroxidase

Thyroglobulin

Processed Thyroglobulin Endocytosis
Products of Thyroid gland

• Mono-iodotyrosine (MIT) and Di-iodotyrosine (DIT)

• Thyroid hormones
  – Thyroxine (T4) (3,5,3',5'-L-tetraiodothyronine)
  – Tri-iodothyronine (T3) (3,5,3'-L-triiodothyronine)

• Reverse T₃ (rT₃) (3,3’5’-L-triiodotyrosine)
Peripheral metabolism of thyroid hormones

- Only T₃ binds intranuclear thyroid hormone receptors (TRs)

- De-iodinases remove iodine moiety from T₄
  - 3 types D1, D2 and D3
  - 40% T₄ → T₃ by D1 and D2
  - 45% T₄ → rT₃ by D1 and D3
Biological functions of thyroid hormone

1. Growth and development of fetus and child

2. Calorigenic effect or **thermogenesis** is the major effect of thyroid hormone.
   – It is mediated by uncoupling of oxidative phosphorylation.
   – Increases $O_2$ consumption within tissue
   – Enhances mitochondrial metabolism
   – Increases sensitivity to catecholamines
3. **Earliest effect of T4 is stimulation of RNA synthesis** and consequent increase in protein synthesis. Higher concentration of T3 causes **protein catabolism and negative nitrogen balance**.

4. Basal Metabolic Rate (BMR) is increased (Thyroxine increases cellular metabolism)

5. **Gluconeogenesis and carbohydrate oxidation** are increased.

6. **Fatty acid metabolism is increased.** Cholesterol degradation is increased and hence cholesterol level in blood is decreased, which is another hallmark of hyperthyroidism.
Regulation of thyroid hormone production

Thyroid hormones in circulation

- Both $T_3$ and $T_4$ are highly plasma protein bound.
- Unbound form is biologically active.
Binding Proteins

• Main function ➔ maintainance of a large pool of hormone that can be mobilised when needed.

• Plasma proteins that bind thyroid hormones are
  – Albumin – more CAPACITY
  – Prealbumin – Transthyretin (thyroxine binding prealbumin)
  – Globulin – thyroxine binding globulin (TBG) – more AFFINITY
• T3 & T4 are LIPOPHILLIC; their free forms in plasma are in equilibrium with a larger pool of protein bound thyroid hormones

• Free thyroid hormones are added to the circulating pool by the thyroid

• These free hormones are the physiologically active forms that send feedback to inhibit pitutary secretion of TSH
Alteration in Thyroid hormone binding proteins

<table>
<thead>
<tr>
<th>Increase in TBG concentration or affinity</th>
<th>Decrease in TBG concentration or affinity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genetic causes</td>
<td>Genetic causes</td>
</tr>
<tr>
<td>Non thyroidal illness such as HIV infections, hepatitis an estrogen producing tumors</td>
<td>Non thyroidal illness such as surgical stress, chronic liver disease, nephrotic syndrome</td>
</tr>
<tr>
<td>Pregnancy, new born</td>
<td>Drugs such as anabolic steroids, large doses of glucocorticoids</td>
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<td>Drugs such as OCPs, Estrogens, Tamoxifen</td>
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</table>
Clinical disorders of Thyroid gland

- Based on history, clinical examination and laboratory results; patients can be classified into:
  - Euthyroid
  - Hypothyroid
  - Hyperthyroid

<table>
<thead>
<tr>
<th>Hypothyroidism</th>
<th>Hyperthyroidism</th>
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</thead>
<tbody>
<tr>
<td>Mental dullness</td>
<td>Nervousness</td>
</tr>
<tr>
<td>Increased sleep, lethargy</td>
<td>Sleeplessness</td>
</tr>
<tr>
<td>Hoarseness of voice</td>
<td>Excessive sweating</td>
</tr>
<tr>
<td>Cold intolerance</td>
<td>Heat intolerance</td>
</tr>
<tr>
<td>Weight gain</td>
<td>Weight loss</td>
</tr>
<tr>
<td>Constipation</td>
<td>Diarrhoea</td>
</tr>
<tr>
<td>Signs: Bradycardia, dry skin</td>
<td>Signs: tachycardia, systolic murmurs</td>
</tr>
</tbody>
</table>

Signs:
- Hypothyroidism: Bradycardia, dry skin
- Hyperthyroidism: Tachycardia, systolic murmurs
# Etiology of Primary and Secondary Hypo & Hyperthyroidism

<table>
<thead>
<tr>
<th>Hypothyroidism</th>
<th>Hyperthyroidism</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ENDOGENOUS</strong></td>
<td><strong>ENDOGENOUS</strong></td>
</tr>
<tr>
<td>Autoimmune thyroid diseases:</td>
<td></td>
</tr>
<tr>
<td>- Hashimoto thyroiditis</td>
<td></td>
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<tr>
<td>- Atrophic thyroiditis</td>
<td></td>
</tr>
<tr>
<td>- Post partum thyroiditis</td>
<td></td>
</tr>
<tr>
<td>Autoimmune thyroid diseases:</td>
<td></td>
</tr>
<tr>
<td>- Graves</td>
<td></td>
</tr>
<tr>
<td>- Hashitoxicosis</td>
<td></td>
</tr>
<tr>
<td>- Post partum thyroiditis</td>
<td></td>
</tr>
<tr>
<td>Na(^+) / Iodide pump dysfunction</td>
<td>Toxic multinodular goiter: Toxic adenoma, familial</td>
</tr>
<tr>
<td>Thyroperoxidase enzyme deficiency, Developmental disorders etc.</td>
<td>HCG secreting tumors.</td>
</tr>
<tr>
<td><strong>EXOGENOUS</strong></td>
<td><strong>EXOGENOUS</strong></td>
</tr>
<tr>
<td>Iodine excess or deficiency, dietary goiterogens</td>
<td>Thyroid destruction by viral or bacterial thyroiditis</td>
</tr>
<tr>
<td>Drugs such as lithium, thionamides</td>
<td>Iodine induced hyperthyroidism</td>
</tr>
</tbody>
</table>
Thyroid auto antibodies

• Main auto antibodies are:
  • Thyroid peroxidase autoantibodies (TPOAs)
  • Thyroglobulin autoantibodies (TGAs)
  • Thyroid microsomal autoantibodies (TMAs)
  • Thyroid receptor autoantibodies (TRAs)
Detection of thyroid antibodies

<table>
<thead>
<tr>
<th>Antibody Name</th>
<th>Disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thyroglobulin autoantibodies (TGAs)</td>
<td>Thyroid cancer</td>
</tr>
<tr>
<td>Thyroid peroxidase autoantibodies (TPOAs)</td>
<td>Hashimoto's Thyroiditis (auto immune)</td>
</tr>
<tr>
<td>Thyroid stimulating immunoglobins</td>
<td>Graves disease</td>
</tr>
</tbody>
</table>
Indications for Thyroid function tests :-

1. Evaluation of thyroid gland in case of clinical suspicion of thyroid disorder
   - Hyperthyroid
   - Hypothyroid
     - Primary (TSH High)
     - Secondary or tertiary (TSH low)

2. Follow up

3. Screening of congenital hypothyroidism.

??? Screening of asymptomatic individuals
TSH

Elevated

Normal

No further testing

Decreased

FT₄

Elevated

Normal

FT₄

Elevated

Decreased

Subclinical hypothyroidism

T₃ toxicosis

Primary hyperthyroidism

Central hyperthyroidism

If Euthyroid: Thyroid hormone resistance

If hyperthyroid: TSH dependent hyperthyroidism
### Routine TFT Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
<th>Normal Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>TSH</strong></td>
<td>Thyroid stimulating hormone</td>
<td>0.5 – 5 mIU/L</td>
</tr>
<tr>
<td><strong>T₄</strong></td>
<td>Serum Total thyroxine</td>
<td>65 – 150 nmol/L</td>
</tr>
<tr>
<td><strong>T₃</strong></td>
<td>Serum total triiodothyronine</td>
<td>1.8 – 3 nmol / L</td>
</tr>
<tr>
<td><strong>fT₄</strong></td>
<td>Free T₄</td>
<td>10 – 23 pmol / L</td>
</tr>
<tr>
<td><strong>fT₃</strong></td>
<td>Free T₃</td>
<td>4 – 7.4 pmol / L</td>
</tr>
</tbody>
</table>

**Thyroid Autoantibodies**

- TPOAs (Thyroid Peroxidase Antibodies)
- TGAs (Thyroglobulin Antibodies)
- TMAs (Thyroid Microsomal Antigen)
TSH

- Best initial test for screening
- Hypothyroidism elevated TSH ( > 5 mIU/L)
- Hyperthyroidism low TSH ( < 0.5 mIU/L)
- All modern TSH methods based on ELISA
• Specimen collection and storage
  – Serum or plasma is used
  – Stable for 5 days at 2-8 °C and 1 month when frozen
• Secretion is circadian, peak between 2 am and 4 am and nadir between 5 pm to 6 pm
• TSH surges immediately after birth (25 –160 mIU/L) and stabilizes in first few weeks.
• Decrease in first trimester due to HCG stimulation
Measurement of Total thyroxine (T₄)

- Principal hormone secreted by thyroid gland
- Highly protein bound (＞99.9 %)
- Total T₄ gives very limited clinical information
- If normal serum binding capacity: total T₄ is inversely proportional to TSH and proportional to free T₄
Measurement of Total triidothyronine ($T_3$)

- Principal active thyroid hormone.
- Only 99% is bound but binding is weak.
- Useful in diagnosis and monitoring of $T_3$ thyrotoxicosis.
Other Thyroid parameters

- Thyroglobulin (Tg)
- Thyroid binding globulin (TBG)
- Reverse T₃ (rT₃)
- Tg mRNA in serum
- Thyroid autoantibodies
Cholesterol

• **Hypothyroidism**: cholesterol level $\uparrow$ (cholestrol carrying lipoprotein degradation decreased
• But not diagnostic as raised in other conditions like $\rightarrow$ DM, HTN, obstructive jaundice
• However this level effective in monitoring the effectiveness of therapy
Radioactive iodine uptake

- Administration of radioactive iodine / technitium pertechnetate allows visualisation of thyroid tissue in neck and throughout body.
- It helps to reveal whether uptake is low or high. Esp in following diseases:
  - Hemithyroid (toxic hyperactive nodule)
  - Cold nodule (nodule fails to take tracer)
  - Ectopic thyroid tissue
**TRH** (Thyrotropin Releasing Hormone) **Response Test**

- Determining basal levels and levels 15 – 30 mins after an IV bolus of TRH

- TRH administration will stimulate the production of TSH

- If the Hypothalamo – Pitutary – Thyroid Axis is normal; $T_3, T_4$ secretions will be increased:
• An abnormal response is seen in:
  
• **Hyperthyroidism**: the negative feedback effect of high T4 overpowers the stimulant effect of TRH. Here thyroid hormones are elevated.

• **Hypopituitarism**: the pituitary could not respond to TRH. Plasma Thyroid levels → subnormal.

• **Primary Hypothyroidism**: exaggerated response, negative feedback effect of T4 reduced.
# Lab findings in Hyperthyroidism

<table>
<thead>
<tr>
<th>Condition</th>
<th>Plasma total T3 and T4</th>
<th>fT4</th>
<th>Plasma TSH</th>
<th>Response to TRH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grave’s Disease</td>
<td>Increase</td>
<td>High Increase</td>
<td>Decrease</td>
<td>Nil</td>
</tr>
<tr>
<td>Toxic Goiter</td>
<td>Increase</td>
<td>High Increase</td>
<td>Decrease</td>
<td>Nil</td>
</tr>
<tr>
<td>T3 Toxicosis</td>
<td>T3 Increase T4 Normal</td>
<td>Increase</td>
<td>Decrease</td>
<td>Sluggish</td>
</tr>
<tr>
<td>Excess intake of thyroxin</td>
<td>Increase</td>
<td>Mild Increase</td>
<td>Decrease</td>
<td>Sluggish</td>
</tr>
</tbody>
</table>
Lab Findings in Hypothyroidism

<table>
<thead>
<tr>
<th></th>
<th>(T_3) and (T_4) in blood</th>
<th>TSH in blood</th>
<th>Response to TRH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary Hypothyroidism</td>
<td>Decreased</td>
<td>Increased</td>
<td>Exaggerated Response</td>
</tr>
<tr>
<td>Secondary Hypothyroidism</td>
<td>Decreased</td>
<td>Decreased</td>
<td>No Response</td>
</tr>
<tr>
<td>Condition</td>
<td>Conc. Of Binding Protein</td>
<td>Total Plasma $T_3 T_4$</td>
<td>Free Plasma $T_3 T_4$</td>
</tr>
<tr>
<td>-----------------------------------------------</td>
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<td>------------------------</td>
</tr>
<tr>
<td>Hyperthyroidism</td>
<td>Normal</td>
<td>High</td>
<td>High</td>
</tr>
<tr>
<td>Hypothyroidism</td>
<td>Normal</td>
<td>Low</td>
<td>Low</td>
</tr>
<tr>
<td>Estrogen, Methadone, major tranquilizers</td>
<td>High</td>
<td>High</td>
<td>Normal</td>
</tr>
<tr>
<td>Glucocorticoids, androgens, danazol</td>
<td>Low</td>
<td>Low</td>
<td>Normal</td>
</tr>
</tbody>
</table>
Assignment

I. Describe synthesis and secretion of thyroxine.

II. Enumerate the thyroid function tests. Describe any one of them in detail.

III. Which parameter is seen in congenital hypothyroidism
   a) Increased TRH
   b) Decreased TRH
   c) Increased TSH
   d) Decreased TSH

IV. Which condition is associated with increased $T_3$
   a) Primary hyperthyroidism
   b) Primary hypothyroidism
   c) Primary hypoparathyroidism
   d) Primary hyperparathyroidism

V. Thyroxine formation requires which trace element
   a) Fluoride
   b) Calcium
   c) Iodide
   d) Phosphorus
Viva questions

1. What is the precursor of thyroxin?
2. What is the ratio of $T_3$ and $T_4$ in blood?
3. What is the function of TSH?
4. What is the function of thyroid hormones
5. Deficiency of thyroxine results in ?
6. What are the salient feature of hypothyroidism?
7. What is the cause of Graves Disease?