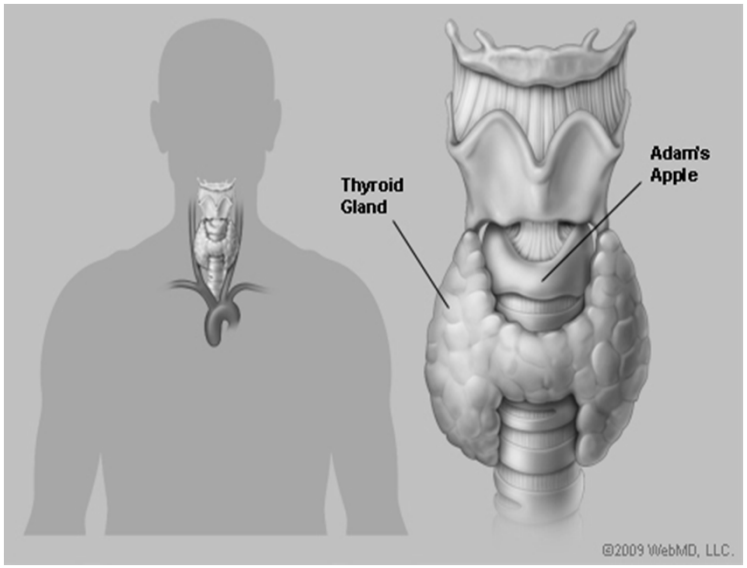

Thyroid function tests

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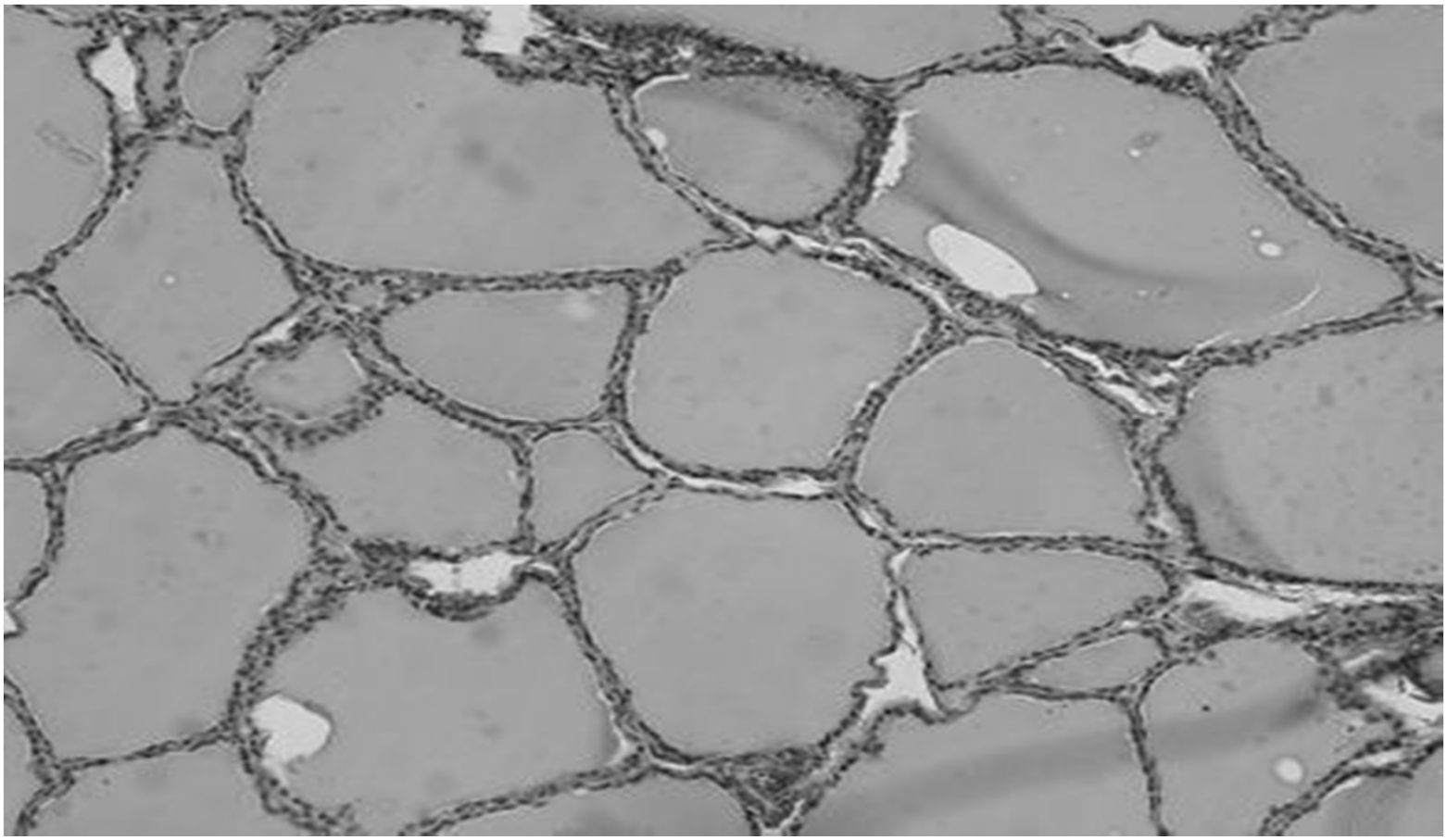
Chemical Pathology

SA13 2014

The Thyroid

- Consists of follicles lined by a layer of epithelial cells
- Colloid which contains thyroglobulin and some thyroalbumin is enclosed by the epithelial cells
- The parafollicular cells (C cells) in the thyroid secrete calcitonin
- Mainly thyroxine (T4) and some triiodothyronine (T3) is secretes by the thyroid

Normal thyroid histology



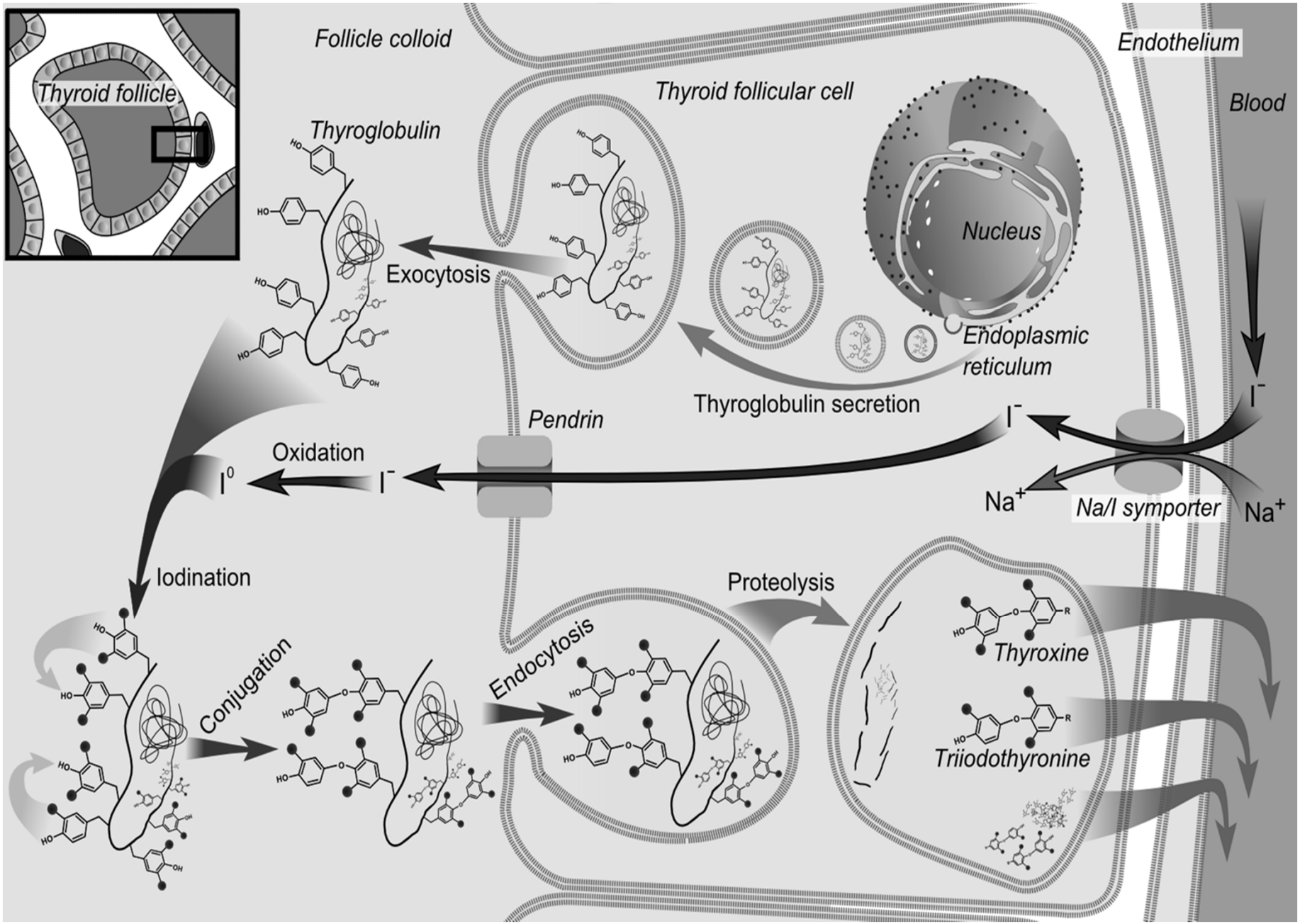
- www.histology-world.com

Actions of the thyroid

- The metabolic rate of most tissues are increased by thyroid hormones
  - ↑ mitochondrial metabolism
  - ↑ membrane Na-K- ATPase activity (15-40% ↑ in resting energy expenditure)
- Positive Inotropic (cardiac contractility) and chronotropic (heart rate) effects
- Promote differentiation and growth (NB in fetal, neonatal and sexual development)
- Stimulate protein synthesis, carbohydrate (CHO) and lipid metabolism
- Gluconeogenesis (formation of glucose from non-CHO substrates) and insulin degradation is accelerated

Synthesis of Thyroid Hormones

1. Iodide is actively transported into the follicular cells
2. Thyroid peroxidase (TPO) oxidise iodide to iodine
3. In the thyroglobulin thyrosine are iodated to monoiodotyrosine (MIT) and diiodotyrosine (DIT)
4. 2 DIT condense to form T4 and 1 DIT condense with 1 MIT to form T3, these thyroid hormones are synthesized at the follicular cell / colloid interface and stored in the colloid (TPO enzyme controls the reactions)
5. Resorption of thyroglobulin by the follicular cells into lysosomes, through pinocytosis, are stimulated by TSH.
6. T4 and T3 are released into the circulation by proteolytic degradation of the colloid.



- www.wikipedia.org/wiki/File:Thyroid_hormone.png

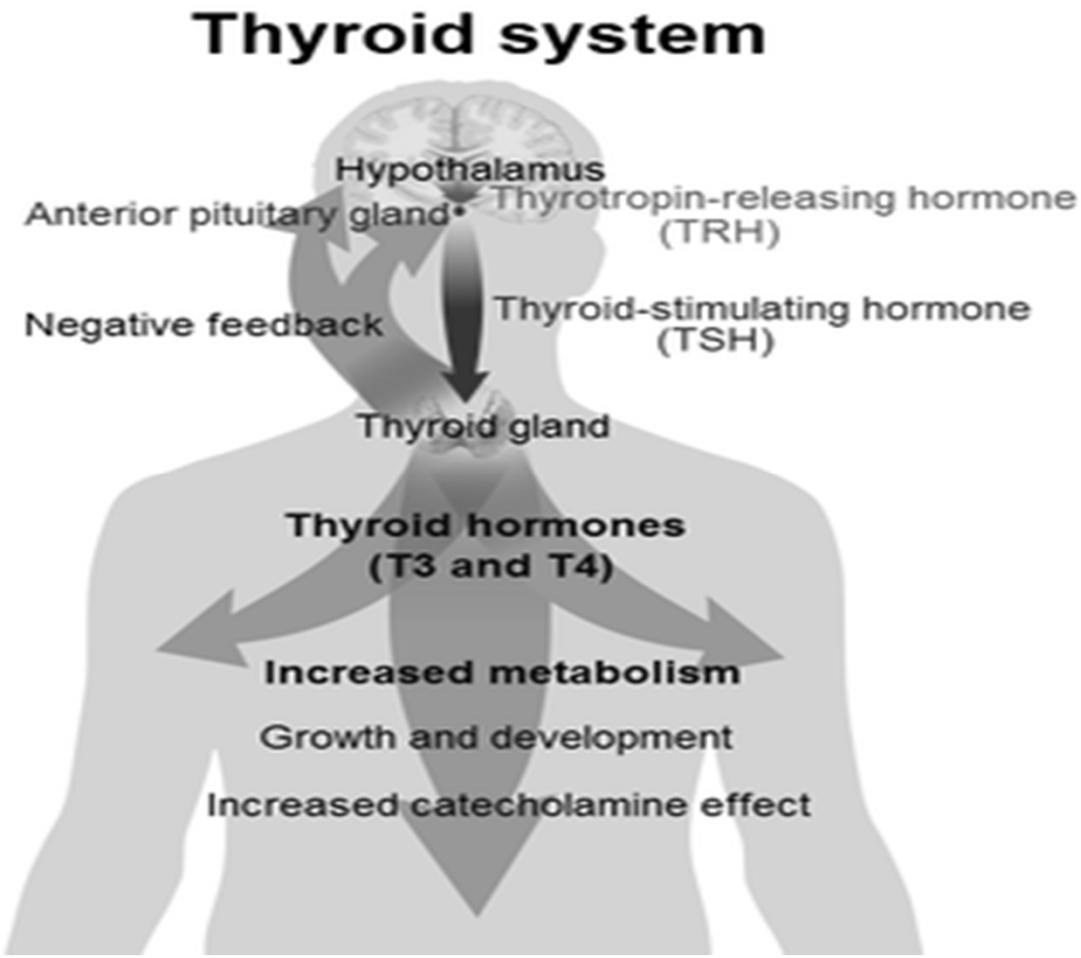
Transport and Metabolism of Thyroid hormones

- 40% T4 secreted by the thyroid gland is converted to T3 by deiodinase in peripheral tissue (liver and kidney)
- 45% of secreted T4 is deiodenated to rT3 (biologically inactive reverse triiodothyronine)
- T4 is 99.97% and T3 is 99.7% bound to carrier proteins:
  - Thyroxine-binding globulin (TBG)
  - Thryxonine-binding prealbumin (TBPA) and
  - Albumin
- Half-life of T4 is 5-7 days and T3 is 1-2 days
- The free fraction is the biologically active form and the large protein bound fraction acts as a reservoir
- T4 and T3 are conjugated by the liver and excreted into the bile as sulphates and glucouronides

Control of Thyroid Hormone Synthesis and Secretion

- TRH (Thyrotrophin-releasing hormone) is released from the hypothalamus, which stimulates the release of TSH (thyroid-stimulating hormone)
- TSH controls the release of thyroid hormones. TSH bind to specific receptors on the follicular cells, which stimulates the release of thyroid hormones within one hour (IGF1-like factor and platelet-derived growth factor are also release when TSH binds to these receptors)
- ↑ in thyroid hormones ⊣ TRH receptors on the pituitary cells, thus ↓ TSH secretion (negative feedback)

Hypothalamic Pituitary Thyroid axis



- www.wikipedia.org/wiki/File:Thyroid_hormone.png

Factors affecting thyroid hormone concentration

1. Age
2. Pregnancy
3. Intra-individual variation
4. Non-thyroidal illness (NTI) / sick euthyroid syndrome
5. Drugs

Age

- Birth:
  - TSH and T4 ↑ rapidly, returns to normal after 3 days
  - Thus tests for hypothyroidism should be done between 3-5 days after birth
- Childhood:
  - TSH, FT4 are within adult ranges, FT3 ↑ range than in adulthood

Pregnancy

- 2-3 times ↑ in TBG due to ↑ oestrogens
- Thus TT4 and TT3 ↑ by 30-100%
- Due to ↑ HCG in early pregnancy:
  - FT4 and FT3 ↑ slightly and TSH ↓ slightly
- Later on when HCG decreases again, FT3 and FT4 ↓ slightly

Intra-individual variation

- Very little fluctuation within an individual

Non-thyroidal illness

- Definition: changes in serum thyroid hormones without any evidence of thyroid disease when the patients have any other diseases
- Systemic illness or fasting leads to ↓ peripheral conversion of T4 to T3 (probable due to glucocorticoids):
  - ↓ FT3, ↑ rT3 and ↓ TSH but FT4 remains normal
- When patients recover TSH shows a transient rise
- Thus best to leave thyroid testing until the patient has recovered.

Drugs that influence thyroid function and their mechanism

↓ TSH secrection	Dopamine, Glucocortocoids, Octreotide
↓ T3 and T4 secretion	Lithium, Iodide, Amiodarone, Aminoglutethimide
↑ T3 and T4 secretion	Iodide, Amiodarone
Altered T4 metabolism	Phenobarbitone, Rifampacin, Phenytoin, Carbamazepine
↓ T4 conversion to T3	Propothiouracil, Amiodarone, B-antagonists, Glucocorticoids
T4 absorption alteration	Ferrous sulphate, Cholestyramine, Aluminium hydroxide, Sucralfate, Ca carbonate, PPI, Raloxifene
Autoimmune disease or thyroiditis	Interferon-alpha, Sunitinib

Amiodarone

- Amiodarone an anti-arrhythmic drug, containing large amounts of iodine
- It inhibit T4 conversion to T3 leading to ↑T4 and ↓T3
- It causes a transient ↑ in TSH
- It inhibit iodine uptake by thyroid cells, causing hypo/hyperthyroidism
- Amiodarone induced hyperthyroidism are seen more frequently in iodine-deficient areas and hypothyroidism in iodine rich areas.

Lithium

- Antidepressant
- Inhibits synthesis and secretion of T4 and T3
- Could cause:
  - Hypothyroidism
  - Goitre
  - Subclinical hypothyroidism
  - Hyperthyroidism due to thyroiditis
- Needs to be monitored 6 monthly

Interferon-alpha

- Used in the treatment of Hepatitis C and malignancies
- Cause thyroid dysfunction in 8% of patients
- Could cause
  - Autoimmune hypothyroidism (subclinical)
  - Destructive thyroiditis
  - Grave's hyperthyroidism

Sunitinib

- Tyrosine kinase inhibitor used for oncology treatment
- Could cause thyroid dysfunction in 60-70% of patients especially hypothyroidism

Drugs that interfere with the measurement of FT4

- Heparin
- Furosemide
- Salicylate
- Carbamazepine

Laboratory assessment of thyroid function

1. Serum TSH
2. Total T4 and T3
3. Free T4 and T3
4. TRH test
5. Thyroglobulin
6. Antibodies (AB) to thyroidal antigens
   - AB to thyroid peroxidase (TPO)/ thyroid microsomal Abs
   - AB to thyroglobulin
   - AB to TSH receptor / thyroid-stimulating immunoglobulins (TSI)

TSH

- Measured by immunometric assay, which are classified as first, second and third generation assays with lower detection limit of 1.0 mU/L, 0.1 mU/L and 0.01 mU/L (the latter being the best)
- A normal TSH [ ], implies a normal thyroid function (Apart from pituitary dysfunction)
- ↑ TSH and ↓ FT4 → Hypothyroidism
- ↑ TSH and N FT4 → Subclinical hypothyroidism
- ↓ TSH, must be able to distinguish between NTI and hyperthyroid state (Third generation is able to)
- ↓ TSH and ↑ FT4 → Hyperthyroidism
- NB TSH is slow to respond to thyroid treatment, thus not reliable to assess adequate therapy < 6 weeks

Total T4 and total T3

- Now generally replaced by free T4 and T3 due to all conditions that could affect the abnormal thyroid binding globulin (TBG) concentration

- ↑ TBG
  - Genetic
  - Pregnancy
  - Oestrogens (OC, oestrogens)
  - Newborns

↓ TBG

-Genetic

-Androgens, anabolic steroids, Glucocorticoids

-Protein-losing states (Nephrotic syndrome)

-Severe NTI

-Salicylates and phenytoin

Free T4 and free T3

- ↑ FT3 and FT4 in hyperthyroidism
- In T3 toxicosis, only FT3 is ↑
- ↓ FT4 in hypothyroidism (FT3 could be normal)
- Interferences in these assays:
  - Rheumatoid factor
  - Complement
  - Antibodies that bind to T4 and T3
  - Antibodies to animal immunoglobulins

TRH test

- TSH response is measured after IV TRH
- In hypothyroidism: the response is exaggerated
- In hyperthyroidism: the response is suppressed
- A normal TRH response excludes thyroid dysfunction
- But due to the sensitive TSH assays available this test is not done routinely

Thyroglobulin

- Used to follow up patients with thyroid carcinoma
- ↑ Thyroglobulin post treatment, suggests recurrence of tumour
- Thyroglobulin antibodies can cause interference, should ideally be measured with thyroglobulin

Antibodies to thyroidal antigens

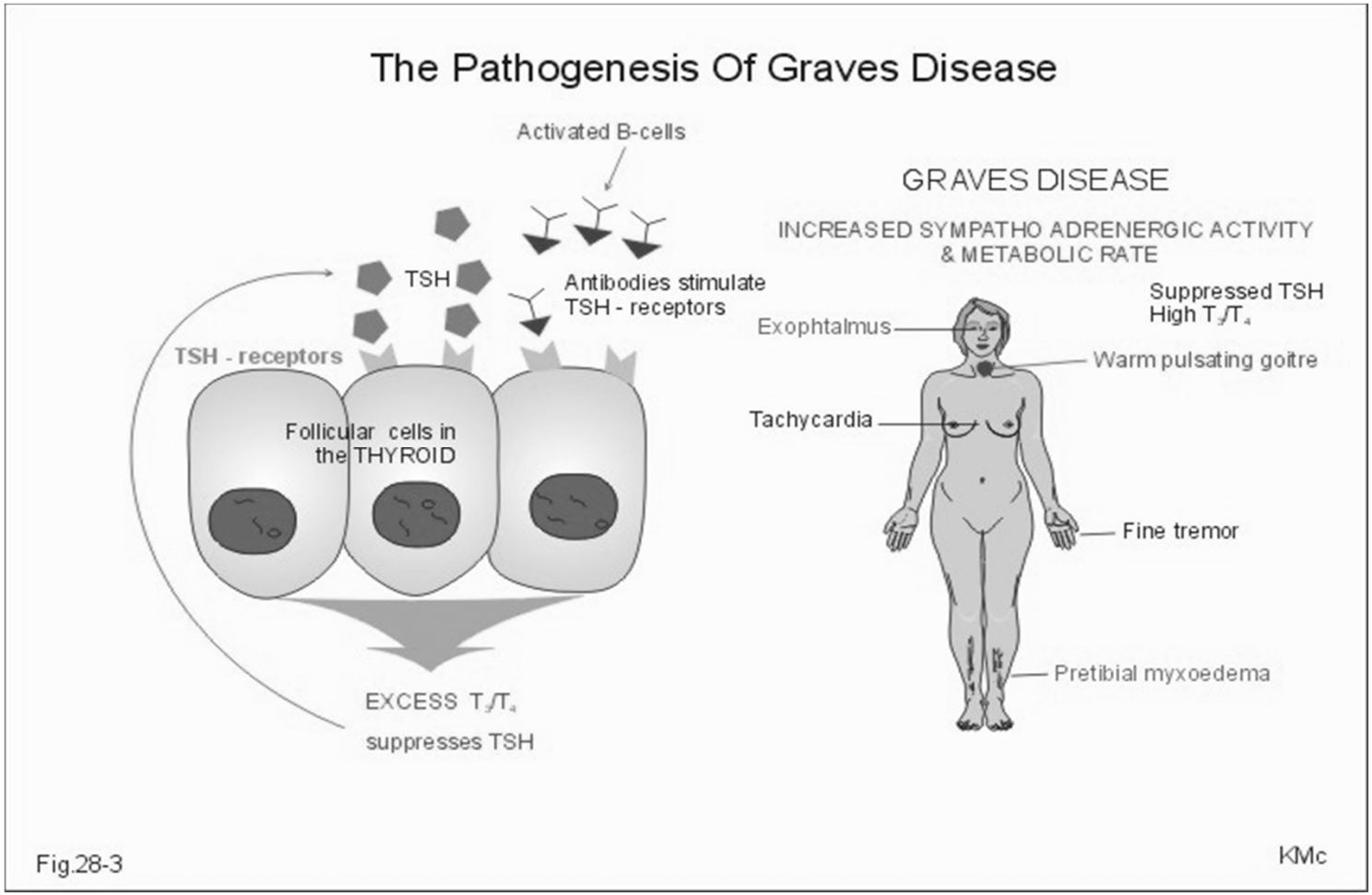
- TPO AB (Thyroid peroxidase AB)
  - Thought to be pathogenic in destructive autoimmune thyroid disease
  - Present in 95% of patients with Autoimmune hypothyroidism (Hashimoto's disease)
  - Present in 70-80% of patients with Grave's disease
  - Also present in apparently health individuals

AB to thyroglobulin

- Also seen in patients with autoimmune disease but less frequently
- Normally done in conjunction with thyroglobulin to exclude interference

Antibodies (AB) to TSH receptor/ thyroid-stimulating immunoglobulin (TSI)

- In Grave's disease autoantibodies of IgG class bind and stimulate TSH receptors on the thyroid follicular cells
- 60-90% of Grave's disease patients have these ABs
- These AB cross the placenta and cause hyperthyroidism in the fetus, thus important to measure in pregnant Grave's disease patients



- www.zuniv.net/physiology/book/chapter25.html

Interpretation of TFT

TSH	↓ FT4	Normal FT4	↑ FT4
↓	-NTI -Treatment of hyperthyroidism	-Subclinical hyperhtyroidism -Over-replacement of T4	-Hyperthyroidism -Over-replacement of T4
Normal	-NTI -Hypopituitarism	Euthyroid	-Erratic T4 replacement
↑	-Hypothyroidism -Inadequate T4 replacement	-Inadequate T4 replacement -Subclinical hypothyroidism	-Erratic T4 replacement -TSHoma -Thyroid hormone resistance

In summary

- Hypothyroidism:
  - ↓ TSH and ↑ FT4 and FT3
- Hyperthyroidism:
  - ↑ TSH and ↓ FT4 and normal FT3

In vivo tests of thyroid function

- Thyroid activity can be monitored by the uptake of iodine^{125} or technetium^{99}
  - ↑ uptake → hyperthyroidism
  - ↓ uptake → hypothyroidism
- Scintillation camera imaging after IV technetium^{99} is useful to identify nodules
  - Hot or active nodules
  - Cold or inactive nodules (possible malignancy)
  - Distinguish between uniformly uptake in Grave's disease and patchy uptake multinodular goitre

Disorders of the thyroid gland

1. Hyperthyroidism
   - Subclinical hyperthyroidism
   - Thyroid storm

2. Hypothyroidism
   - Subclinical hypothyroidism

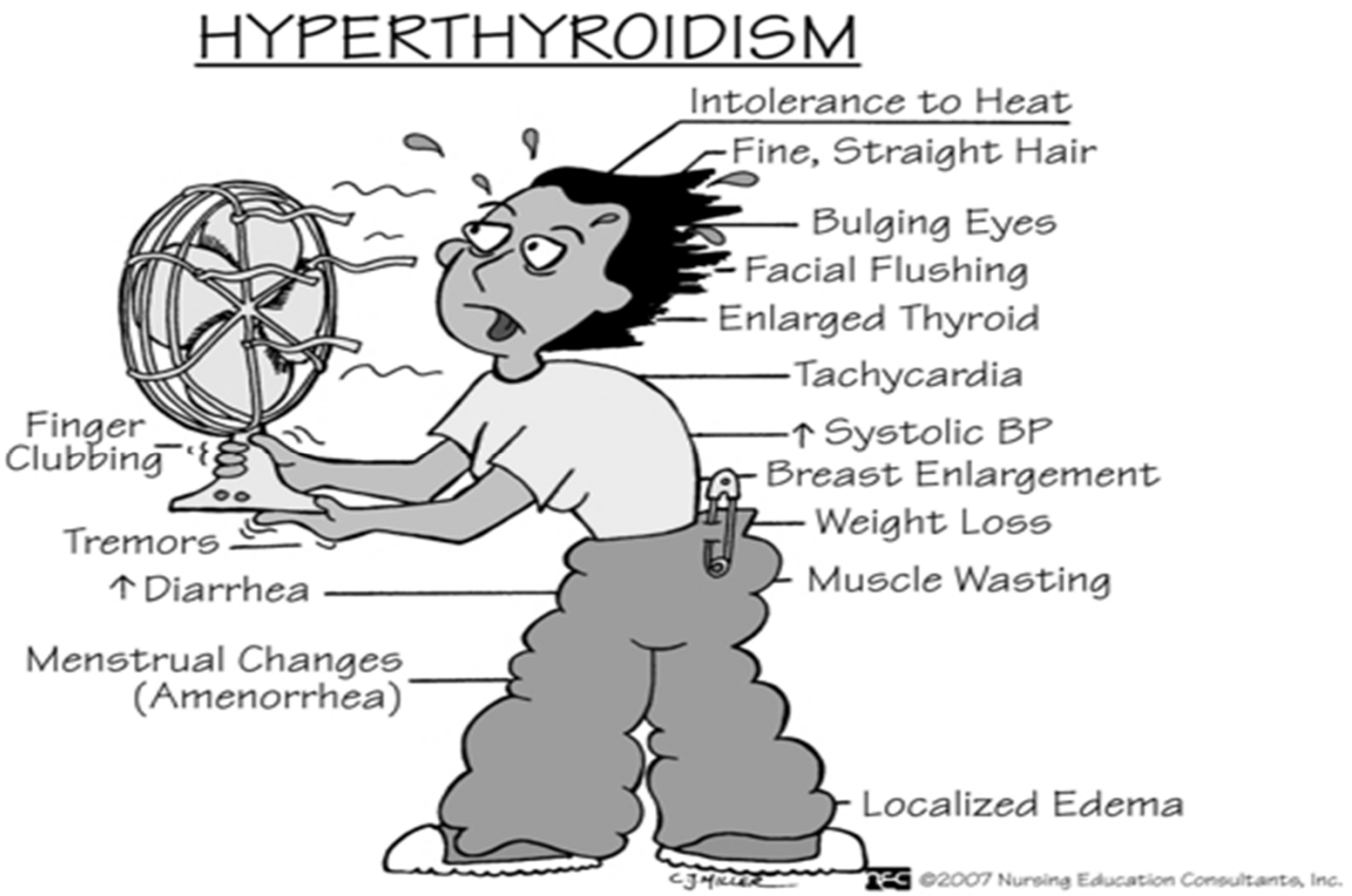
Hyperthyroidism

Causes

- Graves disease 85%
- Toxic multinodular goitre 6%
- Toxic solitary adenoma 3%
- Acute thyroiditis 3%
- Choriocarcinoma
- Hyperemesis gravidarum
- Drug induced thyrotoxicosis
- Postpartum thyroiditis
- Excessive ingestion of T4 and T3
- TSH-secreting adenoma

Clinical features of hyperthyroidism

- Symptoms
  - Weight loss
  - Fatigue
  - Menstrual irregularities
  - Increased sweating
  - Restlessness, agitation
  - Nervousness
  - Palpitations
  - Diarrhoea
- Signs
  - Tremor
  - Goitre
  - Tachycardia / AF (Atrial Fibrillation)
  - Muscle weakness
  - Proximal myopathy
  - Eye signs



- www.images.findthebest.com

Management

1. Anti-thyroid drugs: Carbimazole or Propylthiouracil
   - Takes up to 4-8 weeks to normalize FT4
   - B-blockers may relieve symptoms in this time because it inhibits the conversion of FT4 to FT3
   - 18 month treatment will result in remission in 30-40% of patients
   - Thus still NB to monitor therapy by TFT
2. Radioiodine
3. Partial thyroidectomy

Subclinical hyperthyroidism

- Definition: ↓ TSH with normal FT4 and FT3
- The subjects are more likely to develop overt hyperthyroidism (1-4% per year)
- Evidence that these patients have ↑ bone loss and ↑ risk for cardiovascular complications
- Treatment remains controversial

Thyroid storm

- Usually develops in untreated or incompletely treated thyrotoxicosis
- The crisis is precipitated by infection, surgery, trauma, DKA and parturution
- Clinical picture:
  - Severe hypermetabolism
  - Severe tachycardia, arrhythmia, heart failure
  - Disorientation, agitation, even coma
- Treatment NB: 20% mortality rate
  - B-blokkers, IV Na Iodide, Glucocorticoids and supportive measures

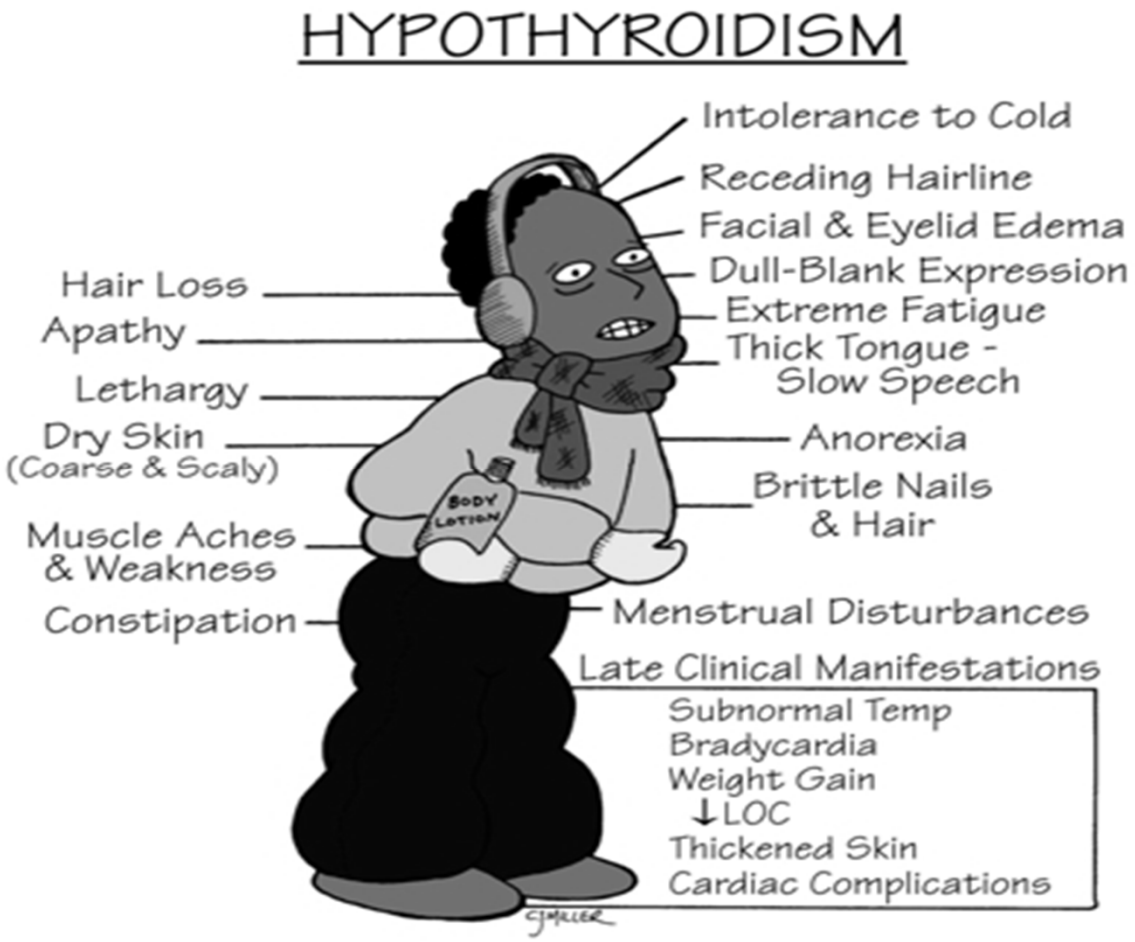
Hypothyroidism

- Causes:
  - **Primary**
    - **Chronic lymphocytic thyroiditis (Hashimoto's)**
    - **Atrophic hypothyroidism**
    - **Iatrogenic (Post surgery, radioiodine therapy)**
    - Subacute thyroiditis
    - Postpartum thyroiditis
    - Congenital
    - Dyshormonogenesis
    - Iodine deficiency
  - Drug induced
    - Lithium
    - Iodine
  - Secondary
    - Pituitary disease
    - Hypothalamic disease

- The most common cause of hypothyroidism is Hashimoto's, atrophic hypothyroidism and iatrogenic
- Secondary hypothyroidism:
  - ↓ TSH and ↓ FT4

Clinical features of hypothyroidism

- Symptoms
  - Tiredness and lethargy
  - Cold intolerance
  - Weight gain
  - Hoarseness of voice
  - Hair loss
  - Constipation
  - Depression
- Signs
  - Dry, coarse skin
  - Bradycardia
  - Slow relaxation of muscle
  - High cholesterol
  - Growth retardation
  - Carpal tunnel syndrome



- www.images.findthebest.com

Subclinical hypothyroidism

- Definition: ↑ TSH with normal FT4 and FT3
- Symptoms: non-specific; depression and tiredness; more common in patients over 60 years
- A significant proportion of these patients go on to develop overt hypothyroidism
- UK thyroid association recommends thyroxine replacement if TSH > 10 mU/L, especially with the presence of TPO antibodies
- Regular follow up of patients not on therapy is necessary (annually)

Management of patients with hypothyroidism

- Started on low dose thyroxine treatment, and increase as necessary
- The aim of the therapy is to keep TSH in the reference range (TSH takes weeks to normalize)
- Patients on long term thyroxine therapy should have their TFT done annually

Screening for Thyroid disease

- Congenital hypothyroidism
  - Prevalence: 1 in 3500 live births in the UK
  - If treated early (with thyroxine replacement) these babies develop normal, otherwise they develop mental retardation (cretinism)
  - Thus screening is important, which is normally done at 5-8 days of age by measuring the TSH concentration
- Screening for hypothyroidism in the elderly
  - Current practice is incidental screening due to costs
  - NB: NTI effects should be kept in mind

Goitre

- Definition: Enlargement of the thyroid gland
- Presentations:
  - Multi-nodular
  - Diffuse uniform enlargement
  - Single nodule
- Very common in iodine deficient areas
- May be associated with normal, hypo- or hyper thyroid function
- Diagnosis: Fine needle aspiration (FNA)

Tumours of the thyroid

- Malignant tumours of the thyroid
  - Follicular
  - Papillary
  - Medullary
  - Anaplastic carcinoma
- Medullary Ca originates from parafollicular cells and secretes calcitonin, thus calcitonin is measured as a tumour marker
- S-Thyroglobulin: is measured after total thyroidectomy in patients with thyroid carcinoma. Always remember that thyroglobulin AB may interfere in the measurement of thyroglobulin

Key points

1. T4 is the main hormone secreted by thyroid gland
2. Peripheral conversion of T4 is the main source of T3
3. TRH from the hypothalamus regulates the secretion of TSH by the pituitary gland, which regulates the secretion of T3 and T4. TRH and TSH is regulated by T3 and T4 through a negative feedback loop
4. Only 0.03% of T4 and 0.3% of T3 are free in the circulation and the free fractions are biologically active

5. Thyroid function test (TFT) normally consists of TSH and FT4. FT3 may be necessary if T3 toxicosis is suspected
6. In pregnancy and certain drugs could alter thyroid function, TFT should be interpreted with caution
7. Non-thyroidal illness or sick euthyroid syndrome are seen in patient who are ill and presents with abnormal TFT. The TFT need to be repeated when the patient has recovered
8. Hyperthyroidism presents with: ↑ FT4 and FT3 and ↓TSH, and is most commonly due to Grave's disease (autoimmune disease)

9. Subclinical hyperthyroidism:

↓TSH but FT4 and FT3 is normal.

Treatment still controversial

10. Hypothyroidism: ↓ FT4 and FT3 and ↑ TSH, and is most commonly due to Hashimoto's disease (autoimmune) and commonly seen in elderly women

11. Subclinical hypothyroidism:

↑ TSH but FT4 and FT3 is normal.

Thyroxine treatment is indicated is TSH > 10 mU/L

12. Incidental screening is indicated but not general population screening for thyroid disease.

Thank you

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