Model Answer

M.Sc. Zoology (First Semester) Examination-2013

Paper – LZT 103 (Endocrinology)

Section – A

1. (i) d (ii) b (iii) b (iv) c (v) c (vi) a
   (vii) c (viii) a (ix) d (x) b

Section – B

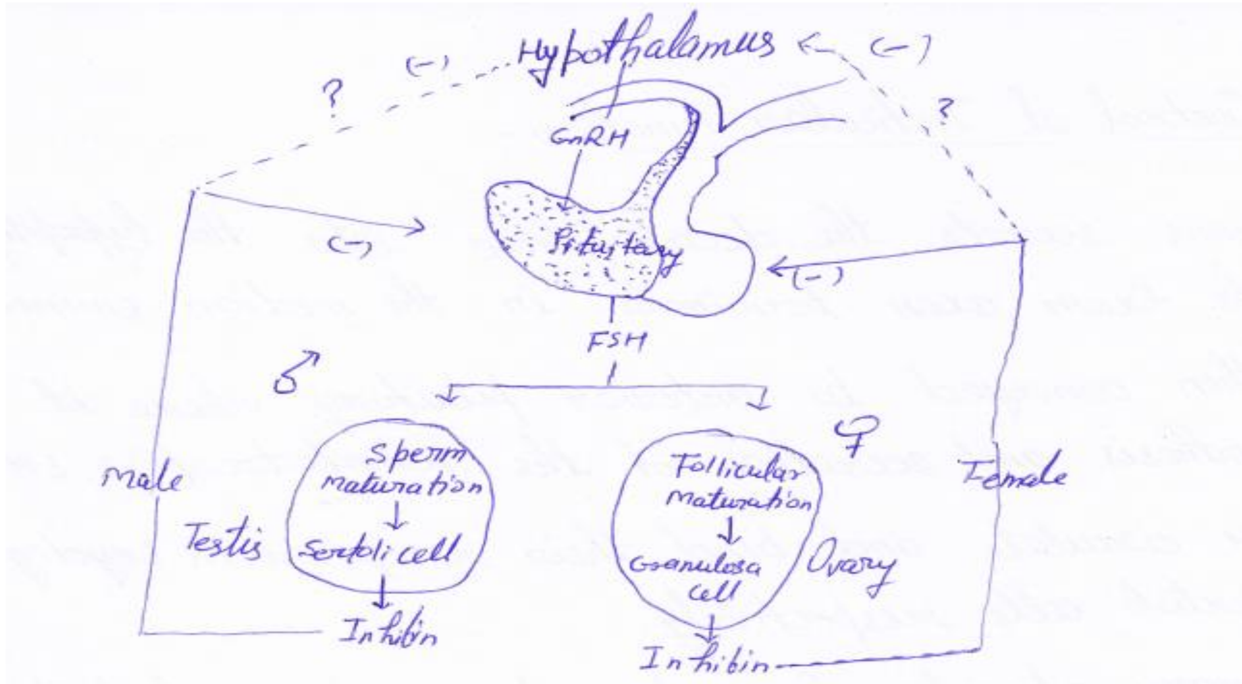
Q.2 Answer

Hormonal Control of Testicular Function

The endocrine regulation of testicular function, i.e., the production of sperm and of androgens is well investigated. The gonadotropins luteinizing hormone (LH) and follicle-stimulating hormone (FSH) are produced and secreted by the gonadotropic cells of the anterior pituitary. In males, they control steroidogenesis and gametogenesis in the testis. Pituitary gonadotropes are the central structure controlling gonadal function and in turn, are regulated by the hypothalamic gonadotropin- releasing hormone (GnRH). Since GnRH secretion is pulsatile, gonadotropin release also occurs in discrete peaks, more evident in the case of LH, due to its shorter half-life in circulation compared to FSH. LH and FSH are glycoprotein hormones secreted by the pituitary gland that control development, maturation and function of the gonad. Like the related thyroid stimulating hormone (TSH) and human chorionic gonadotropin (hCG), they consist of two polypeptide chains, α and β, bearing carbohydrate moieties. The primary functions of the testis, androgen production and gamete development, are regulated by the brain, e.g., hypothalamus and hypophysis via GnRH and gonadotropins. Importantly, the hypothalamohypophyseal circuit is subject to negative feedback regulation mediated by testicular factors. It is generally assumed that either testosterone and FSH alone are able to initiate, maintain and reinitiate spermatogenesis. In order to achieve quantitative effects on germ cell production and sperm numbers, at least under physiological conditions, both LH and FSH activities are needed.

Endocrine control of testicular function can be summarized in following points-

- GnRH neurons secrete the decapeptide into the hypophysial portal vessels from axon terminals in the median eminence.
- GnRH is then conveyed to anterior pituitary where it stimulate synthesis and secretion of the gonadotropins; LH&FSH.
- LH and FSH circulate and bind their receptors in Leydig cells and Sertoli cells respectively.
- LH is primary stimulus for steroidogenesis and testosterone secretion from the Leydig cells, and FSH supports the development and spermatogenic activities of Sertoli cells.
- The neurosecretion of GnRH occurs in an intermittent, or "pulsatile" pattern in both males and females.
- Changes in the frequency of GnRH pulses are believed to be ~~mediate~~ involved in the activation of the male reproductive axis during pubertal maturation, in transitions between reproductive states in seasonally breeding animals, and in other physiologically important changes in gonadotropin secretions.
- The sertoli cells produce a peptide hormone, inhibin, whose role is to exert feedback control of FSH.
- Inhibin itself is stimulated by FSH.
- Thus, a reciprocal relationship between FSH and inhibin secretion exists, where alterations in either hormone are self-limiting as they produce compensatory changes in the other.
- PRL may play a role in the control of testicular Leydig cell LH receptor number.



A Summary scheme of the roles of FSH and inhibin in the control of testicular (and ovarian) function

Q. 3 Answer

Physiological Roles of Androgens -

- FSH and testosterone are required for the initiation of spermatogenesis during sexual maturation.
- FSH interaction with Sertoli cells plasmalemma receptors results in cAMP production and synthesis of an Androgen binding protein (ABP).
- Presumably, cAMP activates a protein kinase that leads to the genomic production of a mRNA coding for ABP.
- Testicular receptors for LH are specifically localized to the Leydig cells.
- In response to LH, cAMP is produced which then causes Leydig cells testosterone production.
- The dramatic developmental and behavioural changes in the male at puberty result from enhanced testosterone secretion by testis.

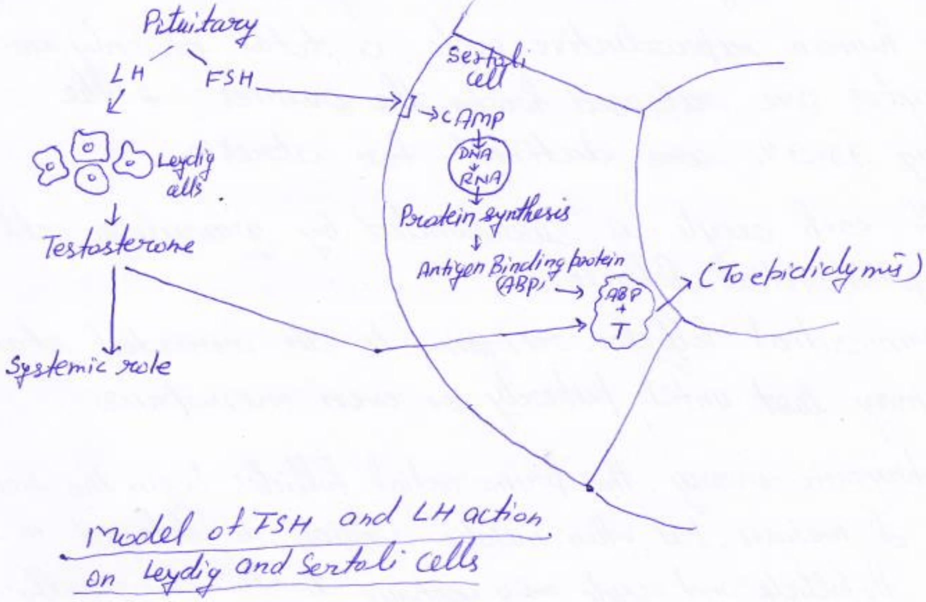
- The accessory reproductive glands are dependent on testosterone to enable them to contribute secretory products to the semen.

- sebaceous gland activity is stimulated by androgens most likely derived from testosterone.

- Acne often present in male is due to increased activity of these glands.

- In most vertebrates, skin, hair, and feather colouration differ between the sexes. These pigmentary changes are usually androgen dependent.

- Growth and shedding of antlers is under the control of androgens.

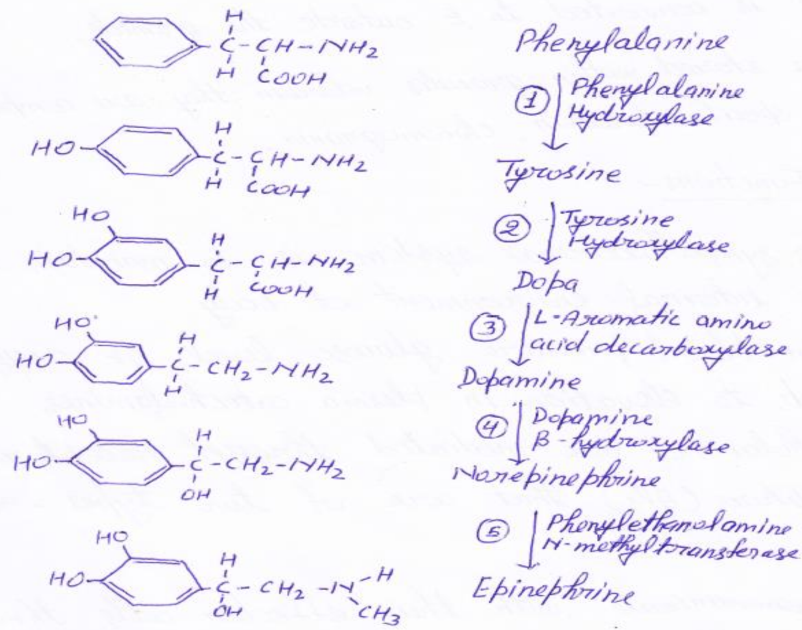


Model of FSH and LH action on Leydig and Sertoli cells

- Androgens exert anabolic actions in muscle, cartilage and other tissue
- There is an age-associated decrease in serum testosterone level.
- Testosterone is clearly necessary to maintain libido

Q. 4 Answer

(a)



Pathway of Catecholamine biosynthesis

- Pathway of catecholamine biosynthesis within CNS, sympathetic postganglionic neurons and adrenal chromaffin tissue appear to be identical.
- Conversion of tyrosine to E involves four steps:
  i) hydroxylation at the three position of the phenolic ring;
  ii) side-chain decarboxylation;
  iii) side-chain hydroxylation; and
  iv) N-methylation

- Dopamine is an important neurotransmitter within CNS.
- In dopaminergic neurons, DA is the final step in catecholamine biosynthesis.
- In noradrenergic cells DA is then converted to NE within the chromaffin granules.
- The NE produced is converted to E outside the granule.
- Catecholamines are stored within granules wherein they are complexed with ATP and a specific protein, chromogranin.

(b)

Biological actions of Adrenocorticoids -

Total loss of adrenocortico secretion usually causes death. Without mineralocorticoids, K concentration of ECF rises markedly. The Na^+ and Cl^- conc. decreases and total extracellular fluid volume and blood volume also becomes greatly reduced. Because glucocorticoids are absent water, carbohydrate and protein metabolism become abnormal and thus death follows.

I - Physiological effects of Glucocorticoids -

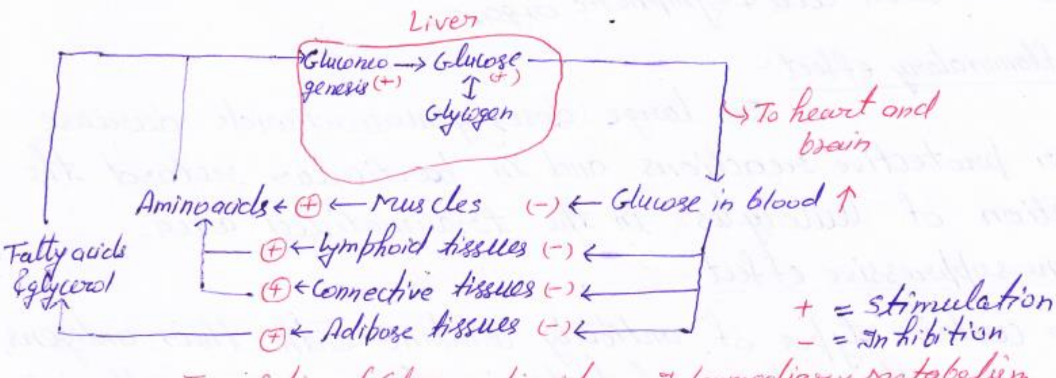


Fig. Action of Glucocorticoids on Intermediary Metabolism

- In peripheral tissues glucocorticoids are catabolic.
- Protein synthesis is depressed. In adipose tissues glucocorticoids increase lipolysis.
- In liver, gluconeogenesis, glycogen deposition and urea production are increased.
- Glucocorticoids thus exert an antinsulin action and make diabetes worse. However, brain and heart are spared and extra glucose supply is ensured to these vital organs.

II - Permissive action of glucocorticoids -

- Small amount of glucocorticoid must be present for a number of metabolic reactions to occur.
- Permissive actions include the requirement of glucocorticoides to be present for glucagon and catecholamines to exert their calorigenic effects.

III - Other effects of Glucocorticoids.

1) Anti-inflammatory effect
2) Immunosuppressive effect
3) Resistance to stress
4) Exocrine secretary effect – Increase secretion of Hcl & pepsinogen
5) Effects on bone
6) Effects on blood vessel
7) Effects on water metabolism
8) Effects on blood cells & lymphatic organs –

Q. 5 Answer

Signal transduction is the process whereby information from outside the cell is conveyed into the cell. This often involves messenger systems. One such system involves a first messenger, such as a hormone, which binds to a cell surface receptor. The binding stimulates production of a second messenger inside the cell. Several molecules have been implicated as second messengers. A widely use one is cyclic AMP (cAMP). cAMP-dependent signal transduction mechanisms involve three separate proteins:

1. A hormone receptor;
2. Adenylate cyclase; and
3. A G protein

G proteins, also known as guanosine nucleotide-binding proteins, are a family of proteins involved in transmitting signals from a variety of different stimuli outside a cell into the inside of the cell. G proteins function as molecular switches. Their activity is regulated by factors that control their ability to bind to and hydrolyze guanosine triphosphate (GTP) to guanosine diphosphate (GDP). When they bind GTP, they are 'on', and, when they bind GDP, they are 'off'.

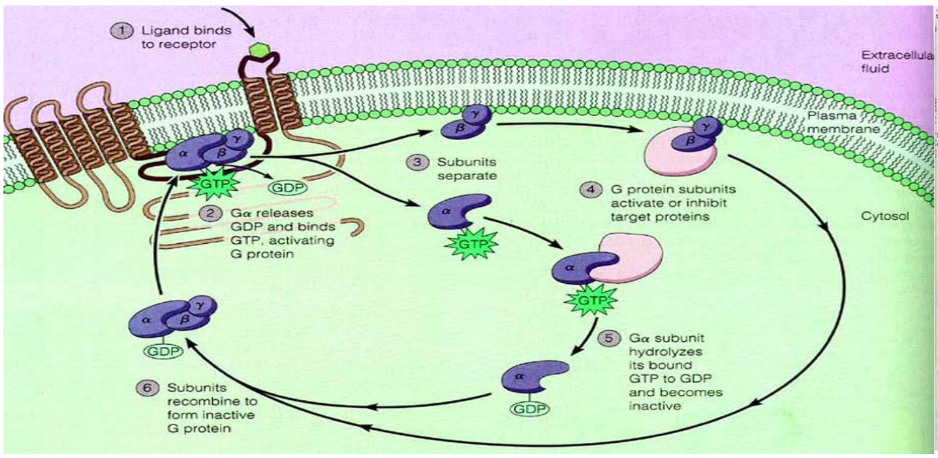
G protein is made up of alpha (α), beta (β) and gamma (γ) subunits. Beta and gamma subunits (regulatory subunit) can form a stable dimeric complex referred to as the beta-gamma complex. Alpha subunit is catalytic subunit.

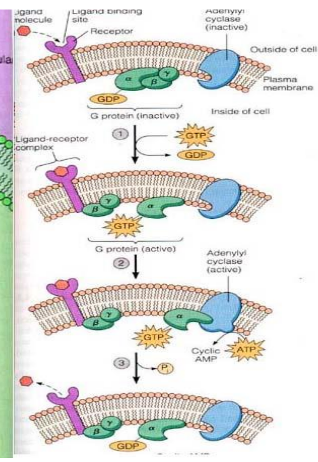
G proteins located within the cell are activated by G protein-coupled receptors (GPCRs) that span the cell membrane. Signaling molecules bind to a domain of the GPCR located outside the cell. An intracellular GPCR domain in turn activates a G protein. The G protein activates a cascade of further signaling events that finally results in a change in cell function. G protein-coupled receptor and G proteins working together transmit signals from many hormones, neurotransmitters, and other signaling factors. G proteins regulate metabolic enzymes, ion channels, transporter, and other parts of the cell machinery, controlling transcription, motility, contractility, and secretion, which in turn regulate diverse systemic functions such as embryonic development, learning and memory, and homeostasis.

In summary, the signal transduction pathway involves the following steps:

1. Binding of extracellular hormone or agonist to a receptor causes a conformational change in the receptor that stimulates it to interact with a nearby molecule of Gs.

2. This in turn stimulates an exchange of bound GDP for GTP--that is, the dissociation of GDP from Gs, to be replaced by GTP. A class of protein factors called guanine nucleotide exchange factors (GEF) assists in the exchange of GDP and GTP.
3. Gs is thereby converted to a protein that activates adenylate cyclase, producing cyclic AMP from ATP.
4. This results in activation of cAMP-dependent protein kinase (protein kinase A), with consequent phosphorylation of target proteins, such as phosphorylase b kinase in cells that activate glycogen phosphorolysis.
5. Phosphorylation of target enzymes results in stimulation or inhibition of metabolic reactions.
Continued activation of Gs depends on the presence of bound GTP.
The Gi protein functions similarly, but it responds to extracellular signals whose response is the inhibition of adenylate cyclase. Here the binding of GTP provokes an inhibitory interaction of Gi with adenylate cyclase, which decreases the synthesis of cAMP.

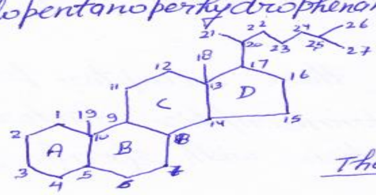




Q. 6 Answer

Steroid Hormones

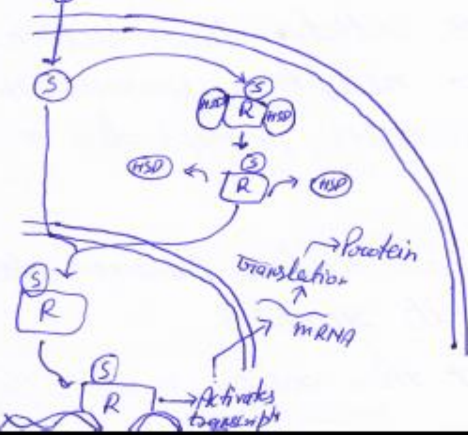
- steroid refers to a variety of lipoidal compounds having a basic structure of four carbon rings known as cyclopentanoperhydrophenanthrene ring or steroid nucleus.



The steroid nucleus

- steroid and thyroid hormones initiates their actions by combining with intracellular receptors, present in the nucleus and, in some cases, perhaps also in cytoplasm.
- Small fraction of free steroid can diffuse through plasmalemma and nuclear membrane to bind its receptor.
- They enter all cells but manifest their action in cells that have receptors.

- Binding of steroid to nuclear receptors results in receptor activation and its translocation to chromatin.
- The binding of steroid-receptor complex to chromatin (DNA and associated proteins) results in de-repression of specific gene and increased m RNA synthesis.
- Each ~~receptor~~ steroid hormone has its own specific intracellular receptor. Thus receptor specificity determines whether a particular cell type will respond to the steroid.
- Synthesis of m-RNA and the subsequent production of protein require a matter of hours or days to be fully manifested. In contrast, actions of most hormones on membrane receptors result in an immediate production of one or more second messengers.
- Steroid receptors belong to a superfamily of transacting "zinc finger" proteins.
- After binding of hormones, these receptor proteins (so-called "ligand-induced transcription factors") influence gene expression via association with specific DNA elements, known as HREs.



Non-genomic pathway -

- Numerous steroid actions are very rapid.
- Not all steroid-target interactions involve intracellular receptors.
- steroids might have immediate membrane-mediated effects in some target cells
- These membrane steroid receptors operate through second messenger systems to alter ion channels or produce other rapid effects in their target cells.

Q. 7 Answer

Anatomy of of Pituitary Gland -

- Composed of tissues that are derived from two diverse origins -

  Anterior lobe → Adenohypophysis.
  Posterior lobe → Neurohypophysis

- Adenohypophysis is glandular tissue
- Neurohypophysis consists of neuronal processes that originates from neurons in the hypothalamus
- Adenohypophysis is derived from oral ectoderm of the stomadeum known as Rathke's pouch.
- Neuronal component arises from the neural ectoderm of the floor of forebrain.
- Adenohypophysis can be subdivided into three regions
  1- Pars distalis
  2- Pars tuberalis
  3- Pars intermedia

- Two subregions can be identified in the neurohypophysis
1- the More interior median eminence
2- Pars nervosa

Pituitary gland (Hypophysis)
- Adenohypophysis
  - Pars distalis } .... Anterior lobe
  - Pars tuberalis }
  - Pars intermedia .... Intermediate lobe
- Neurohypophysis
  - Median eminence } ..... Posterior lobe
  - Pars nervosa }

Subdivisions of Adenohypophysis-

- consists of three subdivisions
  1- Pars tuberalis 2- Pars intermedia 3- Pars ~~nervosa~~ distalis.

1- Pars tuberalis -
- Consists of thin layer of cells ... of neurohyp...
- The portal vessel of the portal system pass near or through the pars tuberalis
- Characteristic of all tetrapod vertebrates.
- Presence of cells containing certain tropic hormones suggests that it is only an extension of pars distalis.
- Recent studies suggest that pars tuberalis is an important link between the pineal gland and the prolactin-secreting cells.

2- Pars Intermedia -

- Only one glandular cell type appears in the mammalian pars intermedia and is responsible for ~~melato~~ melanotropin (α-MSH) secretion.

- In mammals, α-MSH stimulates skin cells, known as melanocytes, to synthesize ~~melat~~ melanin.

3- Pars distalis.

- Five cellular types are present and are responsible for secretion of six pituitary tropic hormones
  - i- Corticotropin (ACTH)
  - ii- Thyrotropin (TSH)
  - iii- Growth harmone or somatotropin (GH)
  - iv- Prolactin (PRL)
  - v- Gonadotropin (GTH) → a. FSH, → b. LH
- In addition α-MSH, secreted by pars intermedia, is also included in tropic hormones.
- All of these hormones are protein or polypeptide.

Cellular types of the Adenohypophysis -

- The cells of pars distalis have been differentiated into
  - → somatotrophs
  - → lactotrophs (mammotrophs)
  - → corticotrophs
  - → thyrotrophs
  - → gonadotrophs
- The individual cells are also refered to as
  - → acidophils
  - → basophils
  - → cromophobes

Cell types	Harmone	Staining characteristic
Corticotroph	Corticotropin (ACTH)	Basophil
Thyrotroph	Thyrotropin (TSH)	Basophil
Gonadotroph.		
FSH-gonadotroph	Follitropin (FSH)	Basophil
LH-gonadotroph	Lutropin (LH)	Basophil
Lactotroph (mammotroph)	Prolactin (PRL)	Acidophil
Somatotroph	Growth harmone (GH)	Acidophil

Histochemical classification of pituitary pars distalis cells

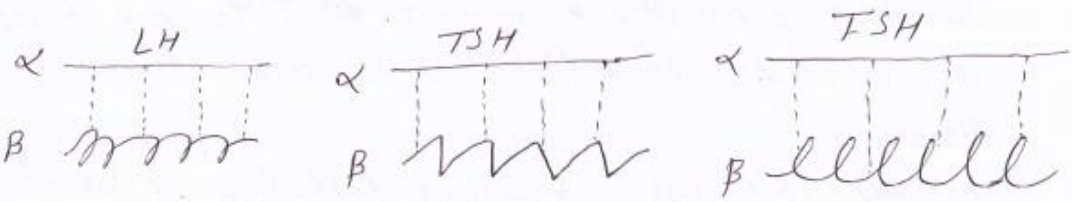
Subdivisions of Neurohypophysis -

Two regions
- → Median eminence
- → Pars nervosa

- Median eminence is more anterior portion
- Pars nervosa is posterior portion
- Both regions are composed of capillaries, pituicytes, and axonal tip of neurosecretory neurons originating in hypothalamus

Category I Tropic Hormones -

- All mammalian glycoprotein tropic hormones are composed of two peptide subunits. (Mol. wt. = 32 kDa)
- Biological half-lives for TSH and LH in mammals are about 60 minutes whereas that of FSH is about 3X longer.
- Each glycoprotein tropic hormones consists of two subunits, an α-subunits and a β-subunit.



- The α-subunit is identical and β-subunit is specific.
- β-subunit is important for unique biological activity.

1) LH: Actions -

- Synthesis of androgens in both males and females is caused by LH action on testes and ovaries.
- LH acts through a G-protein-based, cAMP second messenger system.
- Gamete release (sperm release in males and ovulation in females) also is under control of of LH.
- LH causes formation of corpus luteum from ruptured ovarian follicles remaining after ovulation, and also stimulate corpus luteum to secrete progesterone.

2) FSH: Actions.

- Like LH, FSH binds to a membrane receptor and stimulates cAMP production as a second messenger
- FSH is primarily involved with gamete preparation: that is, ovarian follicle development in females and spermatogenesis in males.

- In females, FSH also stimulates the conversion of androgens into estrogens.

3) <u>TSH : Actions</u> -

- TSH also operates via a cAMP - dependent mechanism to increase synthesis of thyroid hormones, cause release of stored thyroid hormones, and secondarily increase iodide uptake by cells of the thyroid.
- Human may produce a variant TSHs, one of which is associated with pathological condition known as Graves' disease.
- Normal hTSH has a biological half-life of about 0.25 hours. The so called long-acting thyroid stimulator (LATS) in Grave's' disease has a biological half life of 7.5 hours.

<u>Category II Tropic Hormones</u> - (PRL & GH)

- PRL and GH are large, single-polypeptide hormones of similar structure
- Both GH and PRL have common effects on osmoregulation (renal function, intestinal fluid absorption), selective tissue growth (prostate gland, sebaceous gland), lactation, and other processes.
- Membrane receptors for GH and PRL are monomeric proteins that span the cell membrane only once.
- They do not activate adenylyl cyclase.

GH -
- GH ~~enhances~~ affects protein metabolism and electrolyte balance. These actions are mediated indirectly through somatomedins (insulin-like growth factors IGFs)
- ... leads to short stature in young

child, whereas overproduction of GH during early postnatal development leads to gigantism.

- In adult, excess GH secretion leads to acromegaly.
- Short stature may result from a pituitary failure of GH production or from a failure of the liver to respond to GH and synthesize somatomedins (Laron syndrome).
- The pathogenesis of acromegali has been explained by either a pituitary or a hypothalamic mechanism.
- In pituitary mechanism view overproduction of growth hormone may result from GH-secreting tumors of adenohypophysis.
- The hypothalamic hypothesis indicates the defect possibly from an overproduction of GHRH or an underproduction of somatostatin (GH release-inhibiting hormone).

PRL -

- Consists of single chain of 199 amino acids.
- It produces a variety of distinctive actions in animals, including effects associated with reproduction, growth, osmoregulation, and integument.
- Furthermore, PRL may produce synergistic actions with ovarian, testicular, thyroid, and adrenal hormones.
- The best-known action for PRL is the lactogenic effect on the mammary gland.
- PRL stimulates DNA synthesis, cellular proliferation, and the synthesis of milk proteins (casein and lactalbumin).
- In some sp. PRL may influence the synthesis of progesterone by corpus luteum. This is why earlier PRL was named as

Category III tropic Hormones -

- This category comprises several hormones derived from the same precursor, a prohormone known as proopiomelanocortin or POMC, and includes ACTH, α-MSH, LPH and β-endorphins.

- In corticotropes of pars distalis ⇒ POMC —cleaved→
  - → N-terminal 16K fragment = No biological role
  - → ACTH (39 a.a.)
  - → 91 a.a. LPH known as β-LPH (C-terminal) — may again be cleaved:
    - → β-endorphin (61-91 a.a. of β-LPH)
    - → γ-LPH (1-58 a.a. of β-LPH)

- In melanotropes of pars intermedia ⇒ ACTH
  - → α-MSH (residue 1-13 of ACTH)
  - → corticotropin-like peptide (CLIP) (residue 18-39 of ACTH)

1- Corticotropin (ACTH) -

- Corticotropin stimulates the adrenal cortex to secrete glucocorticoids (cortisol and/or corticosterone), hormones that alter protein and carbohydrate metabolism.
- ACTH consists of 39 amino acids in a single peptide chain
- Amino acids 1-23 of ACTH have full biological activity, 1-19 have 80% but fragment ~~1-13~~ 1-16 has very little ACTH activity

2- Melanotropin (α-MSH) -

- In mammals, the epidermal melanin-producing cell is the melanocyte that synthesizes melanin under the influence of α-MSH but extrudes it into the extracellular compartment

- Endorphins function as neuromodulators or neurotransmitters within the central nervous system through their morphine-like actions.
- In addition to their involvement with pain perception, endorphins influence release of neurotransmitters affecting tropic hormone release and can inhibit oxytocin release.

Vasopresin and oxytocin are the major Neurohypophysial hormone -

- Two nonapeptides, oxytocin and arginine vasopressin (AVP) neurohormones are synthesized in the SON and the PVN.
- Most of these neurons project their axons to the pars nervosa although some neurons connect to the median eminence.
- Nonapeptides are stored into pars nervosa and can be released from neurosecretary neuronal endings directly into the general circulation.
- Oxytocin plays an instrumental role in stimulating milk release from the mammary gland through an action on contractile elements of breasts.
- Oxytocin also stimulates uterine contraction and therefore provides a major endocrine stimulus to the process of parturition.
- ~~Vasso~~ Vasopressin plays an essential role in water retention by its action on the collecting tubules of the kidney.

Q. 8 Answer

(i)

- Iodide is co-transported with Na^+ ($Na-I$ symporter) at basal membrane and transported across the apical membrane into the colloid by an apical porter called pendrin.
- More Tgb in lumen → Increases production of pendrin and ultimately iodine uptake from blood increases.
- Resulting concentration of iodide in thyroid exceeds plasma levels by 20 to 40 times.
- Patients with pendrin syndrome have defective gene and are unable to retain iodide in the thyroid gland.

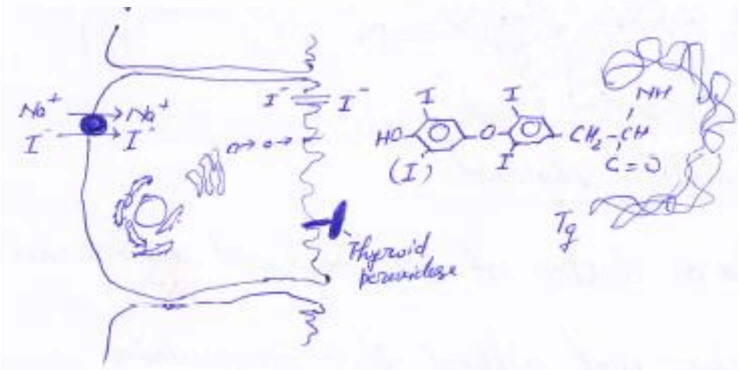
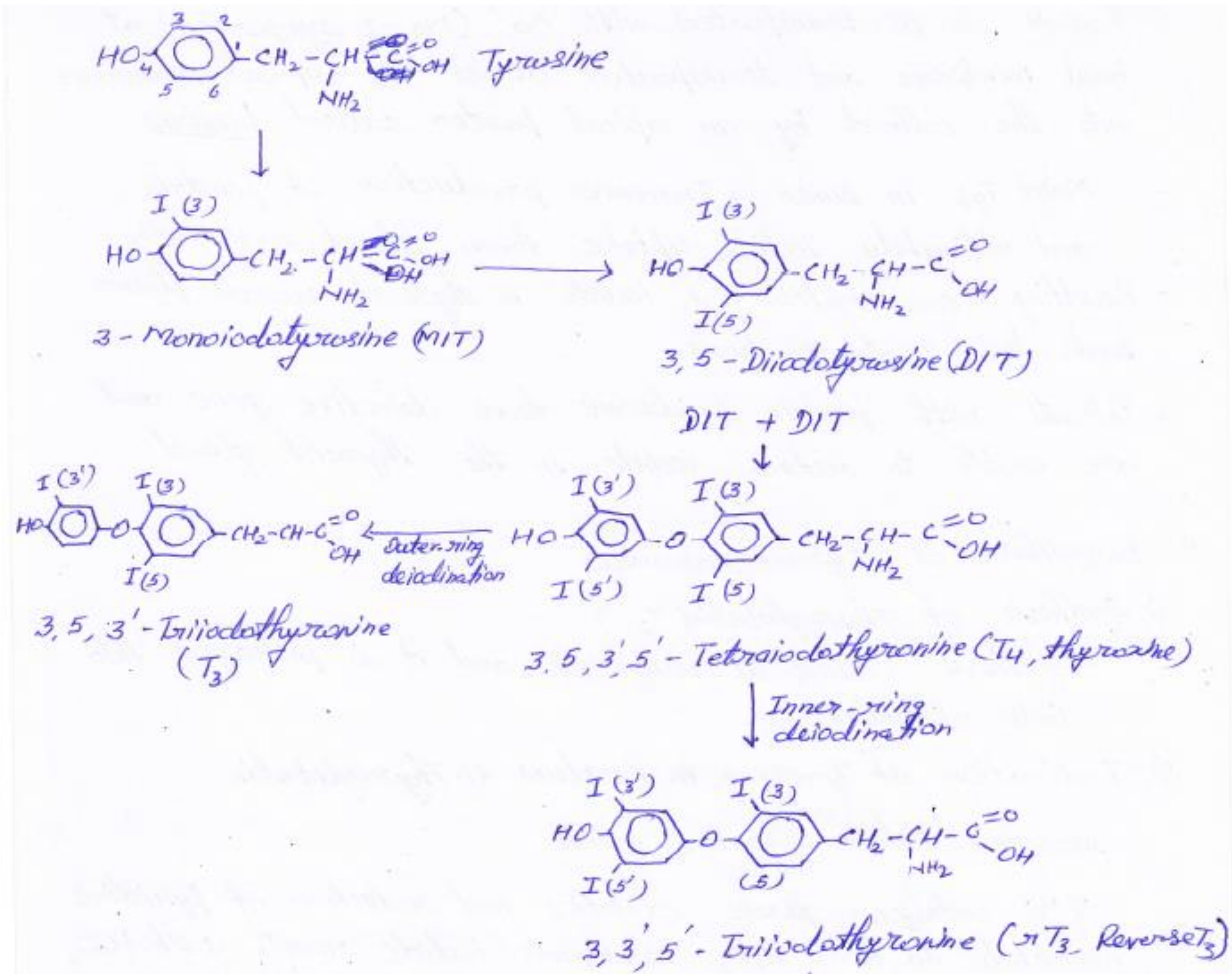


Fig. Iodine uptake by thyroid follicle

TPO catalyzes glucose oxidation and reduction of pyridine nucleotides to form H_2O_2. Inorganic iodide reacts with H_2O_2 to form active iodide, which in turn is attached to tyrosine residues in Tgb.

One iodine attaches at position 3 → 3-monoiodotyrosine (MIT)
Second iodine attaches at position 5 → 3, 5-diiodotyrosine (DIT)
$DIT + DIT \longrightarrow T_4$

(ii) T_4 and T_3



The first step in the synthesis of thyroid hormones is iodine uptake. Iodide is taken up, converted to iodine, and then condensed onto tyrosine residues which reside along the polypeptide backbone of a protein molecule called thyroglobulin. This reaction results in either a mono-iodinated tyrosine (MIT) or di-iodinated tyrosine (DIT) being incorporated into thyroglobulin. This newly formed iodothyroglobulin forms one of the most important constituents of the colloid material, present in the follicle of the thyroid unit.

The other synthetic reaction is a coupling reaction, where iodotyrosine molecules are coupled together. If two di-iodotyrosine molecules couple together, the result is the formation of thyroxin (T4). If a di-iodotyrosine and a mono-iodotyrosine are coupled together, the result is the formation of tri-iodothyronine (T3).

From the perspective of the formation of thyroid hormone, the major coupling reaction is the di-iodotyrosine coupling to produce T4. Although T3 is more biologically active than T4, the major production of T3 actually occurs outside of the thyroid gland. The majority of T3 is produced by peripheral conversion from T4 in a deiodination reaction involving a specific enzyme which removes one iodine from the outer ring of T4.

The T3 and T4 released from the thyroid by proteolysis reach the bloodstream where they are bound to thyroid hormone binding proteins. The major thyroid hormone binding protein is thyroxin binding globulin (TBG) which accounts for about 75% of the bound hormone.

(iii)

CALCITONIN -

- Follicular cells of thyroid gland synthesize thyroid hormone.
- Another cells called (parafollicular) C-cells synthesize and secrete calcitonin.
- CT functions in the prevention of hypercalcemia.
- CT may protect against Ca^{2+} loss from the skeleton during such periods of Ca^{2+} stress as pregnancy, lactation, and prolonged Ca^{2+} deprivation.
- CT is involved in the regulation of feeding and appetite.
- CT directly stimulates vit. D metabolism and indirectly stimulates it by lowering plasma Ca^{2+} levels, resulting in the release of PTH, which activates ~~ret~~ renal vit D synthesis and secretion.

(iv)

- Parathormone is derived from the parathyroids, which in most mammals are embedded on the surface of the thyroid gland.
- In humans four parathyroid glands are present; they are located on the back side of the thyroid gland, one near each pole of the two lobes of the gland.

Human Thyroid with attached Parathyroids

- The parathyroids are composed of two cells types, the chief cells, which are the source of PTH, and the oxyphil cells, of unknown function.
- Removal of parathyroids results in a drop in plasma Ca^{2+} levels, which usually results in tetanic convulsions and death.

Synthesis -

Prepro parathyroid hormone (PreproPTH) (115 a.a.) —Cleavage→ ProPTH (90 a.a.) —6 residues are removed→ PTH (84 a.a.)

Parathormone Acts to Raise
Plasma Ca^{2+} Levels -

- Parathormone acts on a number of tissues, elevating plasma levels of Ca^{2+} and decreasing circulating concentrations of PO_4^{-3}

1- Bone Mineral metabolism
- In response to PTH, these cells secrete one or more cytokines that then stimulate osteoclastic activity leading to the resorption of bone. In this way, demineralization of bone results in elevated levels of Ca^{2+}.

2- Renal reabsorption of Calcium -
- A major physiological role of PTH is to increase renal tubular reabsorption of Ca^{2+}.

3- Intestinal Absorption of Ca^{++}.
- PTH enhances intestinal uptake of Ca^{2+} but this action may be mediated indirectly through its effects on vitamin D metabolism.

4- Control of vit-D synthesis -
- A major role of PTH in Ca^{2+} homeostasis is to stimulate the biosynthesis of 1,25- dihydroxyvitamin D_3 from vitamin D precursors by the kidney.

5- Renal excretion of phosphate