BIOCHEMISTRY

Substrates

	FED (2hr)	FASTING (12-18hr)	STARVATION (1-3d)
Liver (thiophorase xx)	Glc	FA	aa / fa
Muscle	Glc	FA	KB
Adipose	Glc	FA	fa
Brain	Glc	Glc (FA xBBB)	KB
RBC	Glc	Glc	Glc
Heart	FA	FA	KB

'BHM'

Upto 18hrs: Glycogenolysis
18-48hrs: Gluconeogenesis
2-5d: FA oxidn / KB synthesis & breakdown

Cellular transport

Primary active transport :

- Na^+-K^+ ATP pump
- Proton pump

Na^+ OUT 3 — K^+ IN 2

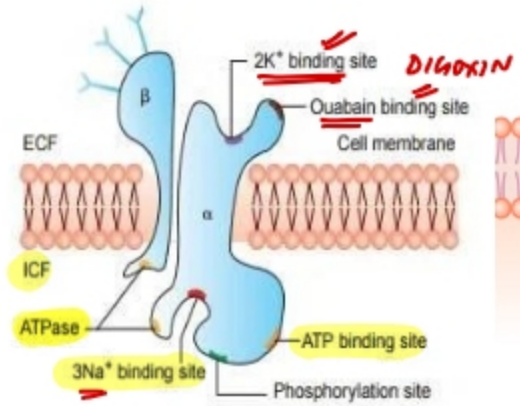
Secondary active transport :

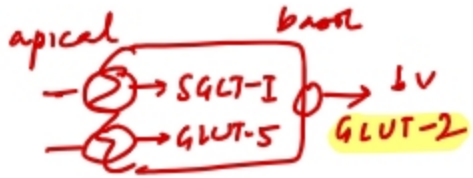
Energy indirect

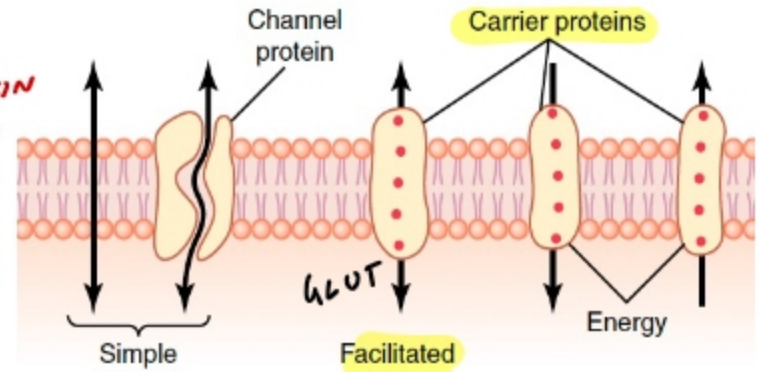
- SGLT
- Na I symport

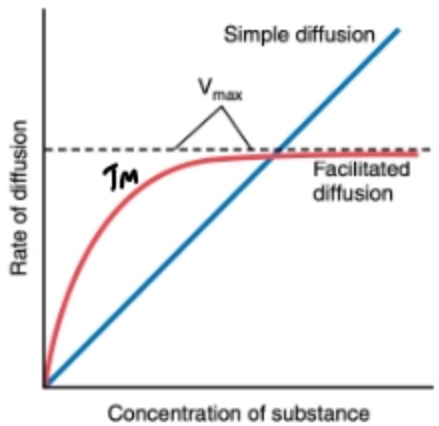
SGLT-1: Intestine >> β-PST

SGLT-2: β-PCT







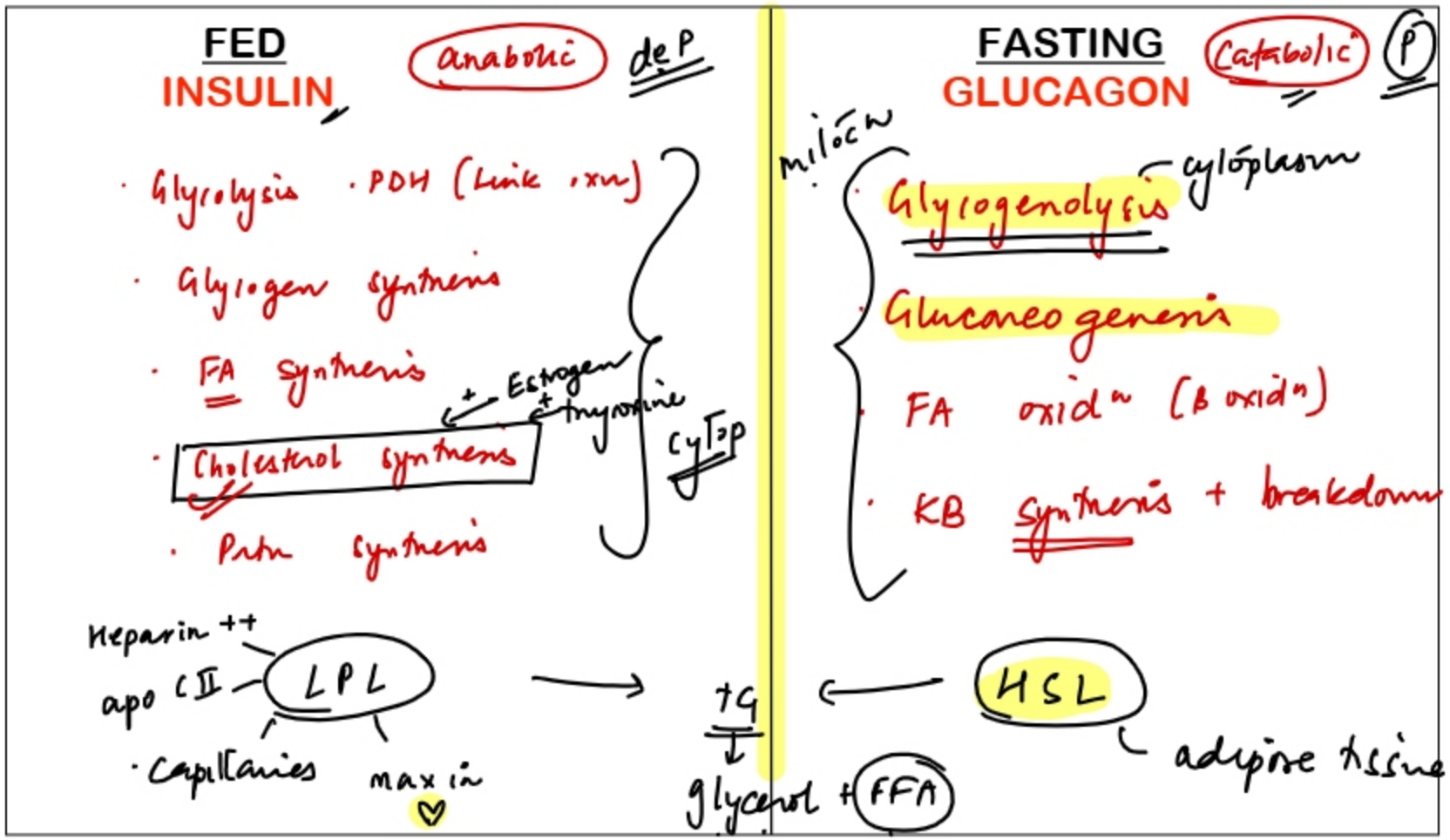


Warburg effect - aerobic glycolysis

18FDG - PET

Transporter	Location	Function
GLUT-1	Brain, Kidney, Placenta, Erythrocytes, Cancer	Basal uptake of glucose, High affinity
GLUT-2	Liver, Pancreatic cell, Small intestine	Fed state
GLUT-3	Brain, Kidney, Placenta	Glucose uptake, Maximum affinity for glucose
GLUT-4	Heart, Skeletal muscle, Adipose tissue	Insulin-stimulated glucose uptake
GLUT-5	Small intestine, Sperms	Fructose

Basics Of Metabolism



BOTH- C + M

- **H** Heme synthesis
- **U** Urea cycle
- **G** Gluconeogenesis
- **P** Pyrimidine synthesis → CUT

PEROXISOME

- β-oxidation of VLCFA (>22C) - 0 ATP
- α-oxidation of Branched-Chain FA (phytanic acid)
- Synthesis of Bile Acids, Plasmalogens
- Catabolism of Amino Acids & Ethanol
- Generate H202

SER = Microsome

- Cholesterol synthesis ⊖ Fibrates
- Bile Acid synthesis (7a-OH)
- G-6-PO4ase

Proteosome

Degrade ubiquitinated proteins — Bortezomib
Lewy body dementia — MM: ⊖ apoptosis

HSL Inhibited by:
Insulin, PGE1, Niacin, Leptin

Diabetics

↑ Catabolic

(insulin ↓ / resistance)

↑ HSL

↓ LPL

- ↑ chylomicron (TG)
- ↑ VLDL

Alcoholics

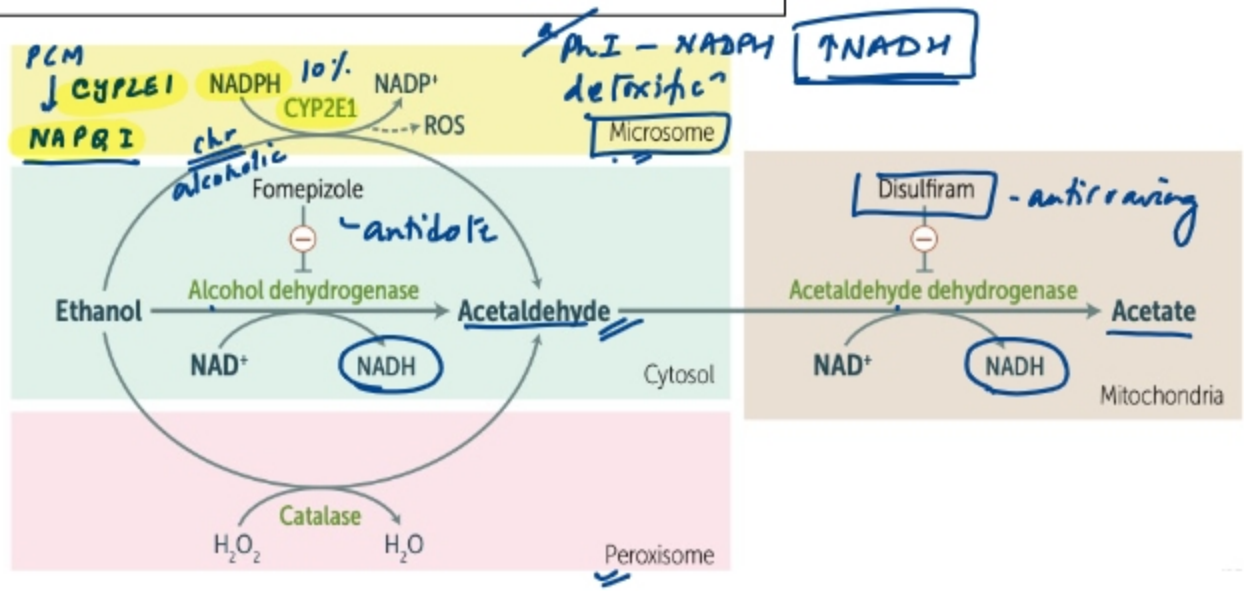
↑ NADH/NAD+ → Gluconeogenesis (−)
→ Krebs Cycle (−)

↑ Pyruvate → Lactic acidosis

→ fasting Hypoglycemia

Acetyl CoA → ketoacidosis

→ FA → Fatty Liver

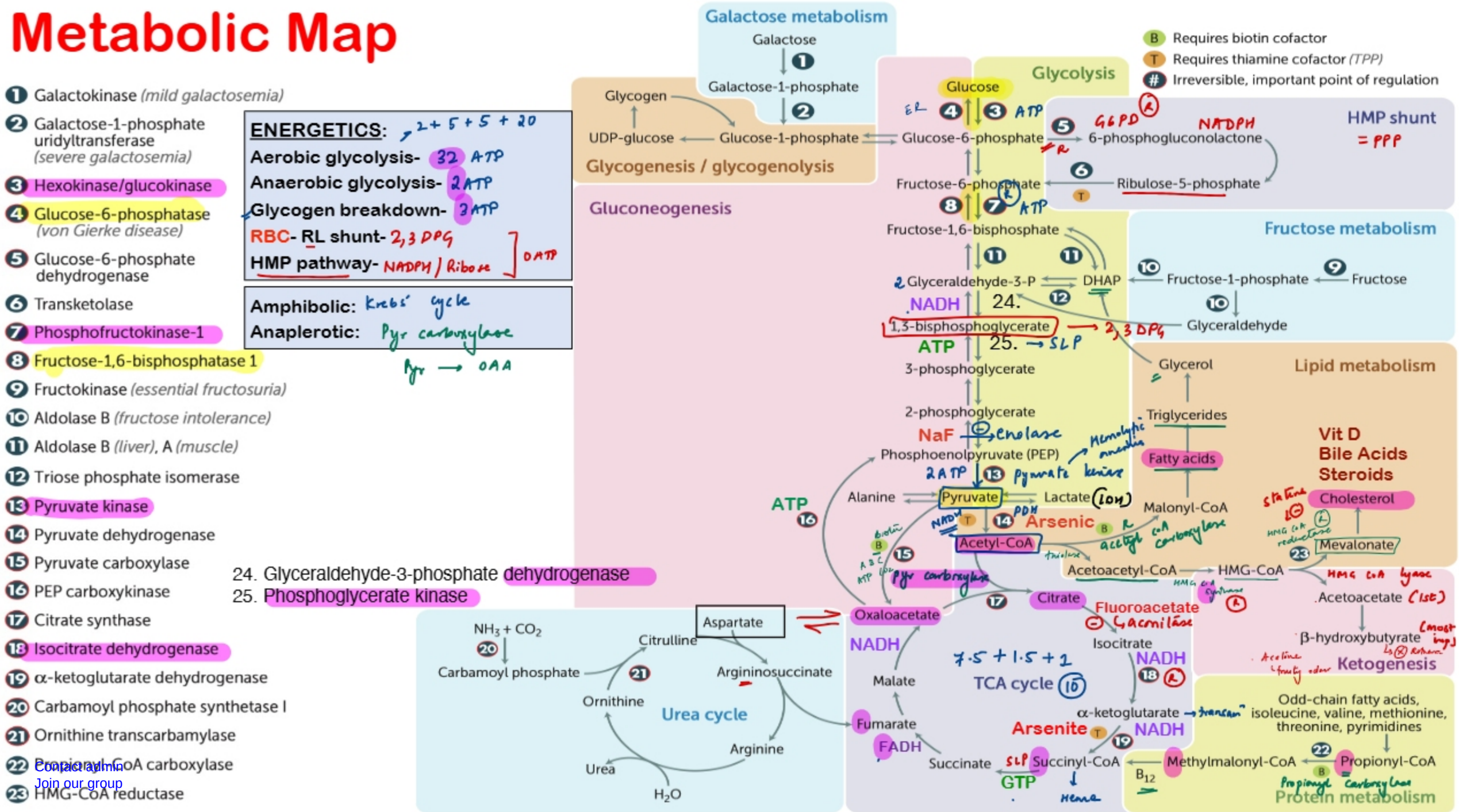


PhI – NADPH detoxific

↑NADH

Disulfiram - antidrinking

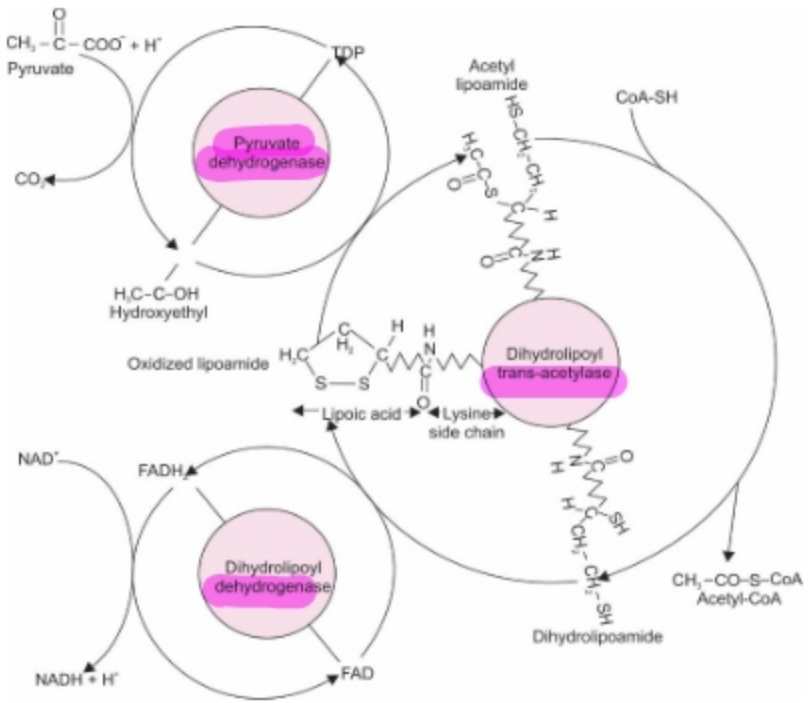
Metabolic Map



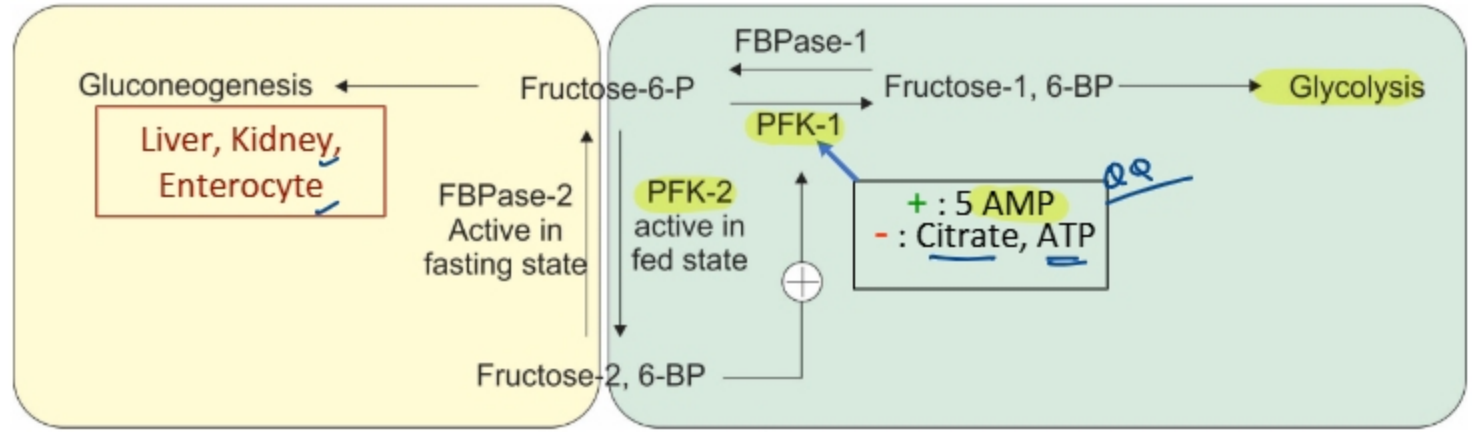
Facts!

Glucokinase- Low affinity
- Liver, β-cells → High Km
- Fed state, Insulin-activated

Hexokinase: High affinity
- All tissues → Low Km
- Inhibited by Glu-6-P



Co-factors for PDH : vit B1, B2, B3, B5, Lipoic acid ↑ (−) Ac



Transamination: vit B6

SGPT / ALT: pyruvate ⇌ alanine (glutamate → α ketoglutarate)

SGOT / AST: oxaloacetate ⇌ aspartate (glu → α ketoglu)

Gluconeogenesis Substrates:
1) Alanine (Cahill)
2) Lactate (Cori)
3) Glycerol (TG)
4) Propionyl CoA (Odd-chain FA)

NOT ACETYL CoA QQ (activator)

Rossman fold: NADP Binding Domain

HMP pathway sites:
- Lens / RBC — NADPH
- Detoxification of xenobiotics in Phase 1 QQ — NADPH
- Adipose
- Adrenal cortex / Gonads
- Liver

(Adipose, Adrenal cortex / Gonads → fa, cholesterol synthesis)

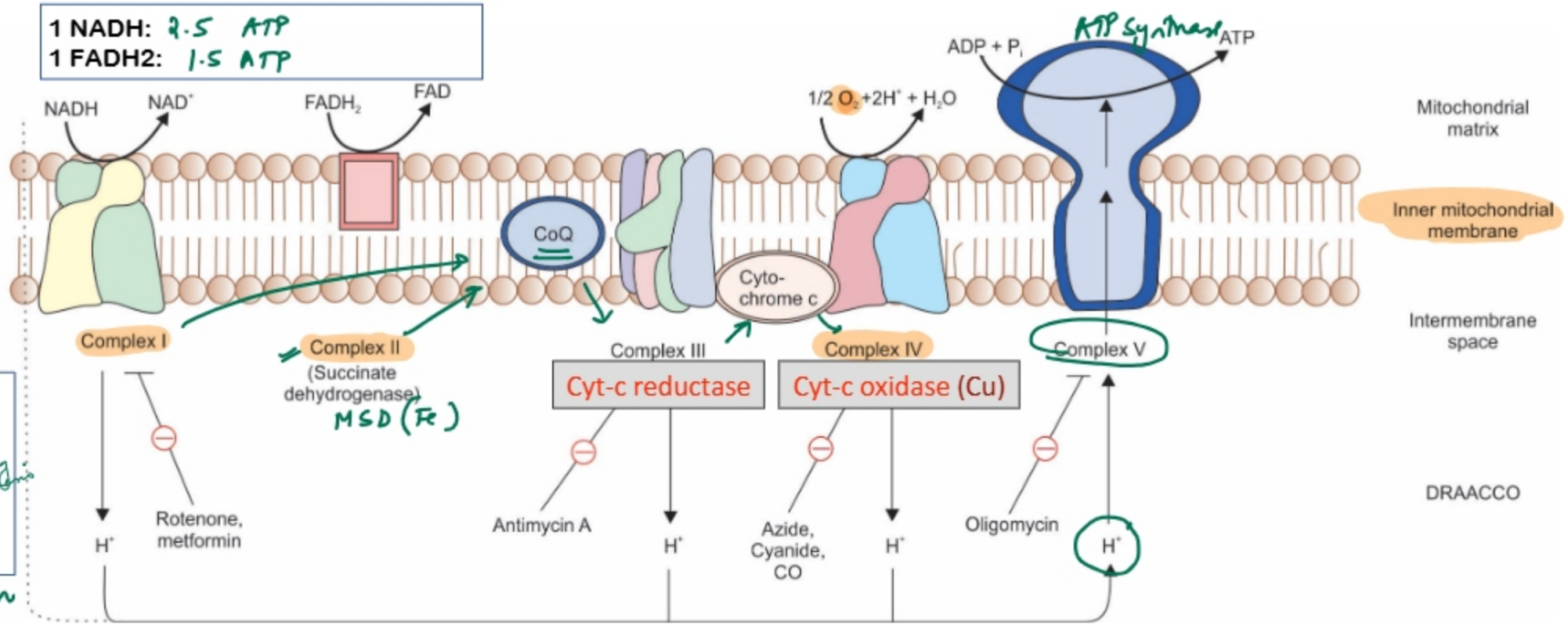
Sources of NADPH:
- G6PD HMP
- Malic enzyme: Pyr → Malate
- Isocitrate DH cytoplasmic

Carnitine shuttle FA oxid"

Citrate shuttle FA synthesis

Malate shuttle QQ oxaloacetate / NADH glycolysis

ETC



Uncouplers:
- Dinitrophenol
- Thyroxine
- Bilirubin
- Brown fat
- Aspirin overdose

Thermogenesis

Complex I :
- Metformin
- Rotenone
- Phenobarbitone
- Piericidin A

Complex II :
- Malonate
- Carboxin
- TTFA

Complex III :
- Antimycin (3)
- Dimercaperol / BAL (3)

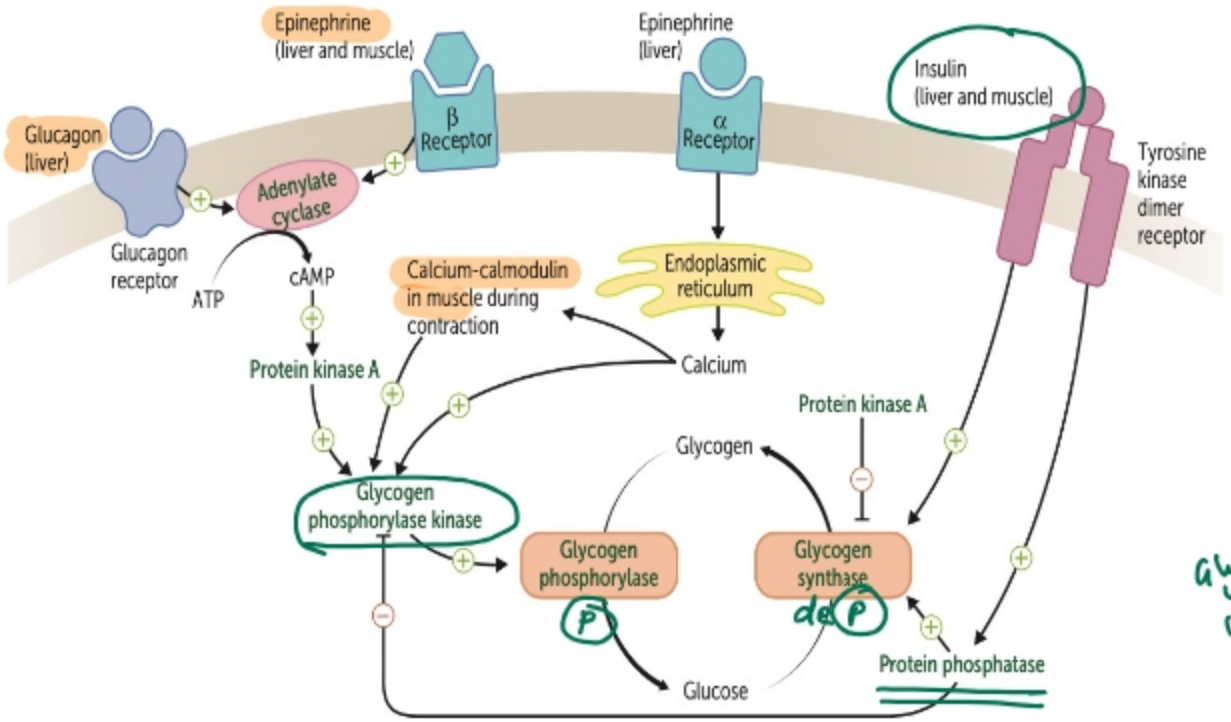
Complex IV :
- Azide
- Cyanide
- CO
- H_2S

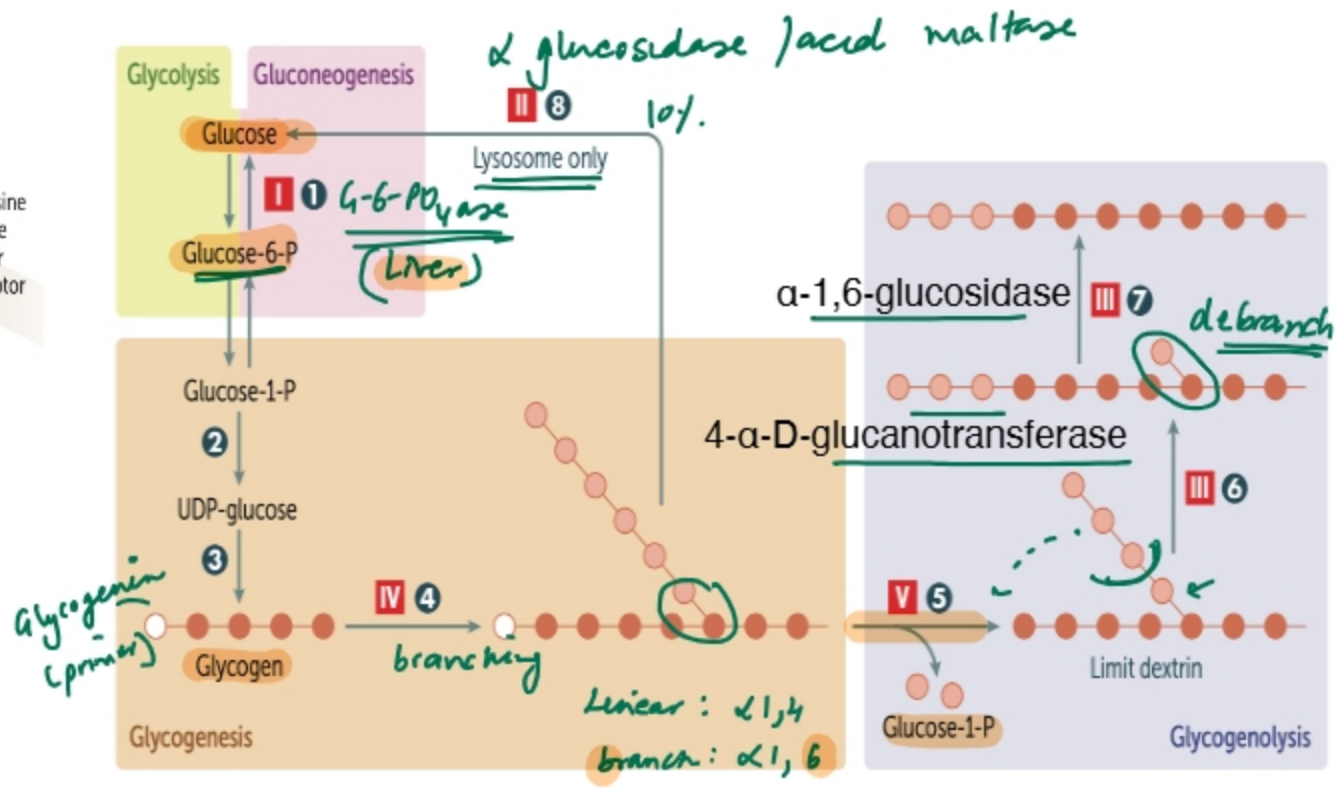
Complex V :
- Oligomycin

ATP transporter :
- Atractyloside

Phosphocreatine (PCr): Creatine + Phosphate (from ATP) = "Storage" ATP Mobilized for exercise

Glycogen Metabolism





N : 50% glc ↑ in 10min

Glucagon Challenge test –ve

Von Gierke's — absent

Cori's — some ↑

Fasting hypoglycemia

0
I
III
VI

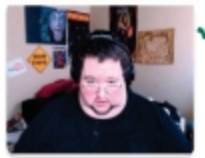
Exercise intolerance

0
III
V
VII

Neither

II
IV

Glycogen Storage Disorders



Glucose ← Glucose - 6- Phosphate

Von-Gierke ds. < glycogenolysis, gluconeogenesis

Glucose - 6- Phosphatase (ER)

Absent in Muscle

De-branching enzyme → Cori's ds.

Glucose - 1- Phosphate

UDP-Glucose Pyrophosphatase

UDP-Glucose

Glycogen Phosphorylase

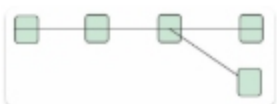
Liver, Muscle

Glycogen

Glycogen synthase

Branching enzyme → Anderson's ds.

Branching of Glycogen



- Hypoglycemia
- Hyperlipidemia
- Hyperuricemia
- Fatty liver
- Liver adenoma

McArdle's ds. V
muscle exercise intolerance
↓glc ↓lactate
rhabdomyolysis
Mburia
2nd wind phenomenon

Her's ds VI
liver
fasting hypoglycemia
(x severe)

PFK-1 VII
Tauri's ds.
hemolytic anemia
exercise intolerance

Acid Maltase
(Lysosomal Alpha- glucosidase)
♡ muscle
Pompe's ds. II
Hypotonia / HOCM
vERT

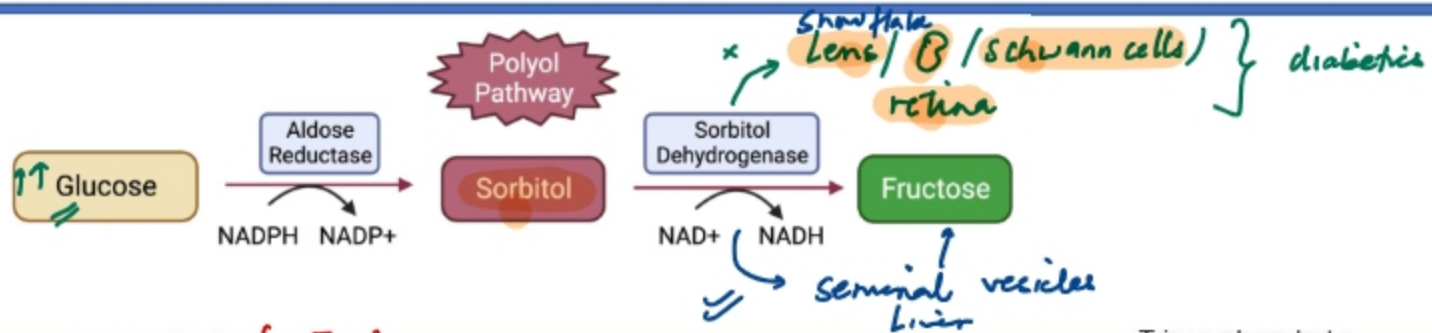
GSD = 0
absent glycogen OR

IV Anderson branching
A B C D
toxic to liver
↓ cirrhosis
amylopectin

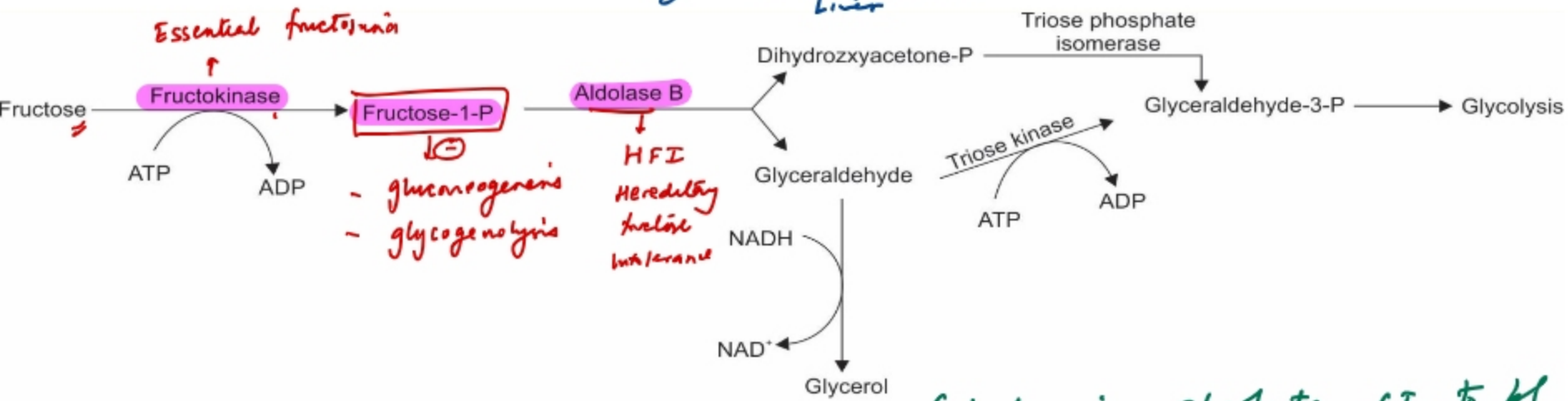
III Cori / Forbes debranching
↓
Hypoglycemia
Dextrins

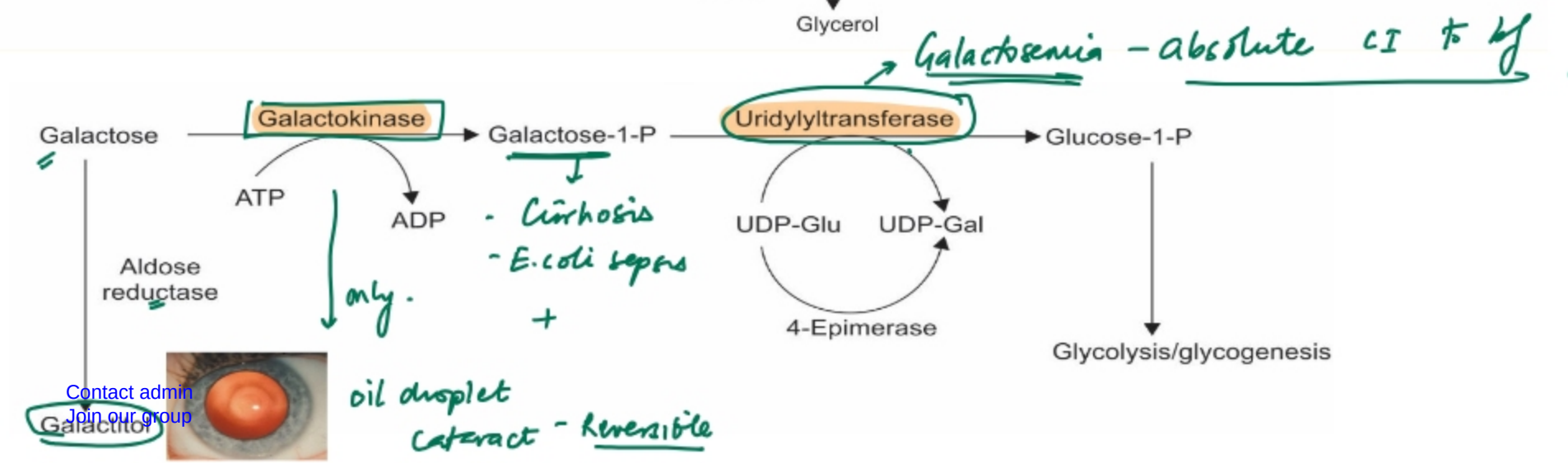
- Coenzyme Glycogen Phosphorylase: B6
- Fanconi-Bickel syndrome: GLUT - 2
- ER G-6-P transporter: GSD 1b

Fructose & Galactose Metabolism

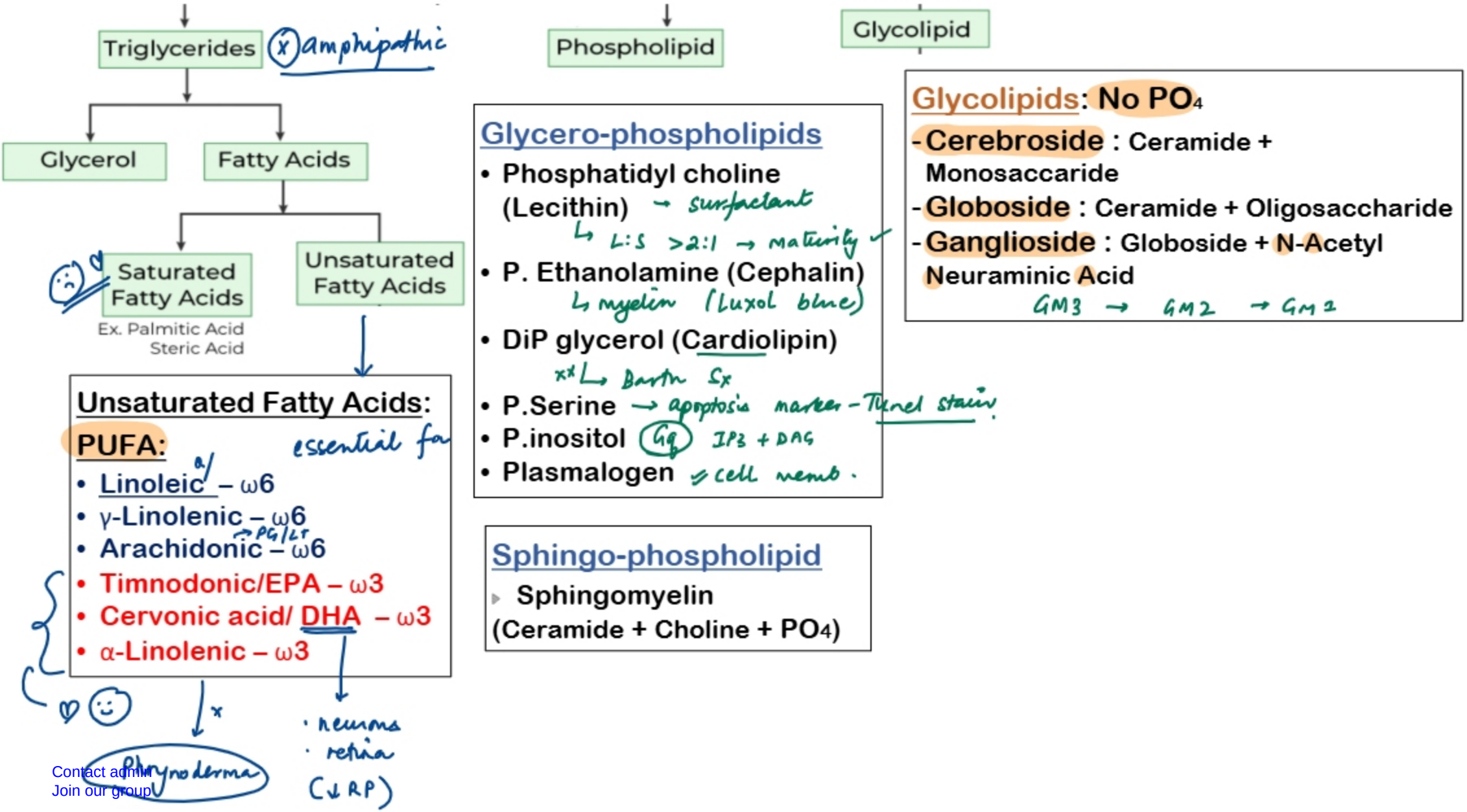


Trehalose	α 1,1	glc - glc
Maltose	α 1,4	
Isomaltose	α 1,6	
Lactose		glc + galactose
Sucrose		glc + fructose
Lactulose		gal + fructose





Types of Lipids



Lysosomal Storage Disorders

Fabry, Gaucher: No CRS, No MR

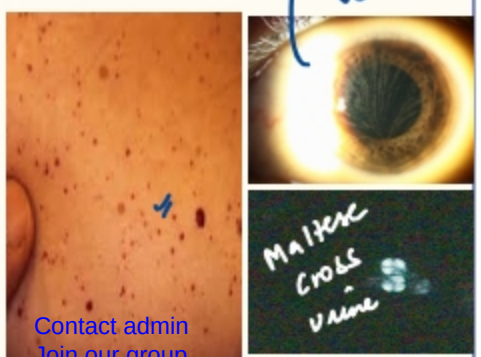
My Fabrite activity is Ceramics. We made A GalaXy

Fabry's → α-galactosidase A — XLR

Ceramide

- neuropathy
- Hypohydrosis
- angiokeratomas

vertex k / corneal verticillata

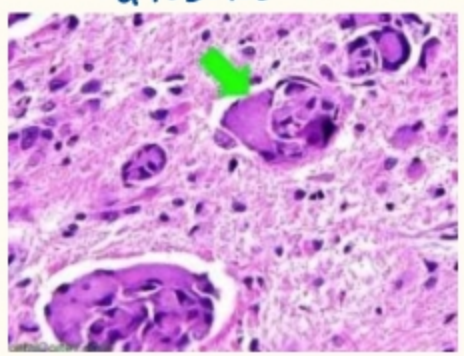


Crab glob is out of this galaxy!

Krabbe's → β-galactosidase

- optic atrophy
- thalamic ↑density

Globoid bodies



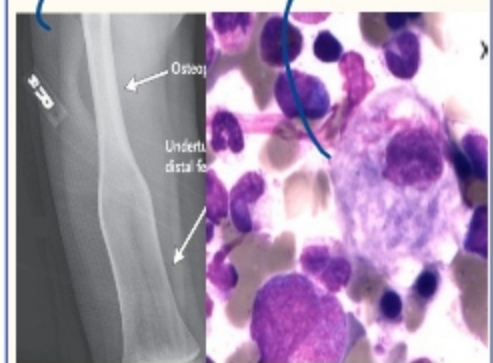
Oh my Gauch! He broke the bones- We need to glu it! (mc)

Gaucher's / β-glucosidase / glucocerebrosidase

- osteopenia / AVN
- HSM + Pancytopenia

Erlenmeyer flask

crumpled tissue paper



A Gang of six small Jews eating onions

GM2 gangliosides

Tay Sachs

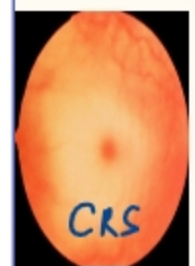
Hexosaminidase A

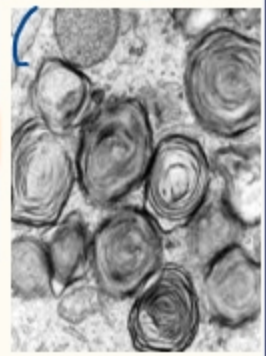
(A+B: Sandhoff)

(X) HSM

- ↑acusis
- ↑startle reflex

onion skin bodies





Pick Zebra's big nose with your sphinger with foam

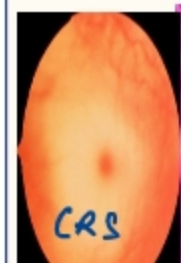
Niemann Pick

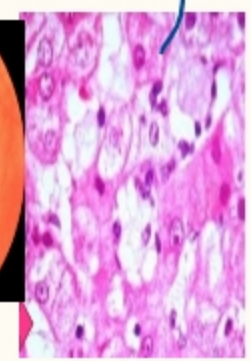
≤ HSM

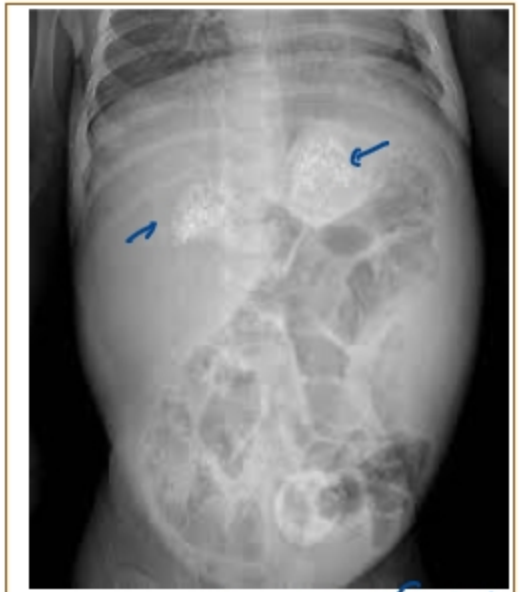
Sphingomyelinase

foamy mphge

Zebra bodies



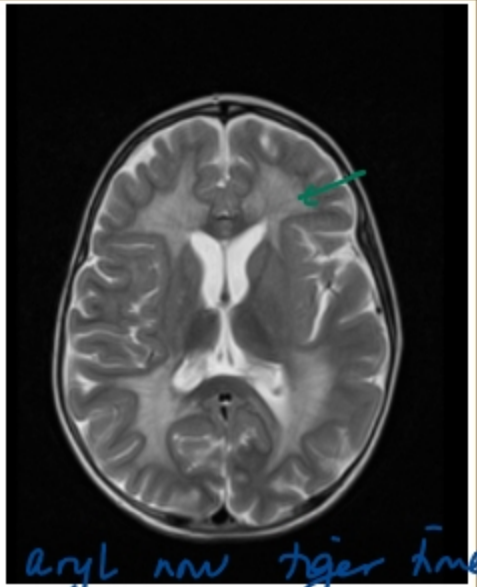




adrenal Ca^{2+} (L/L)

Wolman D

acid lipase xx

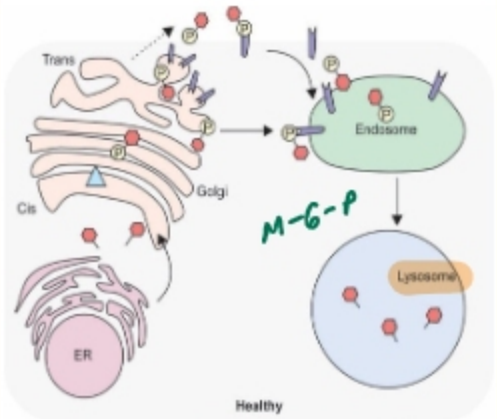


aryl nw tiger line

Aryl sulfatase x

TIGROID WM

MLD D/D: PMD



N-Acetyl Glucosamine phosphotransferase

I-cell D

Ceramidase deficiency
Mimics RA Farber's D

Leukodystrophy N-aspartoacylase xx
NAA peak on MRS CANAVAN's

Macrocephaly-CATS
Canavan's
Alexander's
Tay Sachs'
Sandhoff

MPS: Heparan sulfate + Dermatan sulfate

I du wanna Hunt!

No Corneal clouding	HUNTER	Iduronate -2-suflatase	XLR
Corneal clouding	Hurler's	α-L-iduronidase (Partial defect- Sheie)	AR

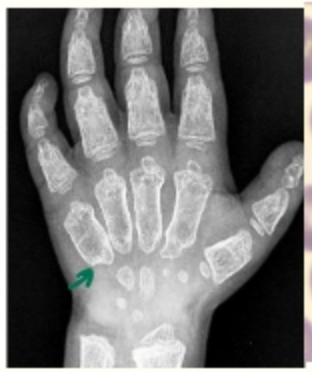
L, (x) MR

MC MPS: Sanfillipo D

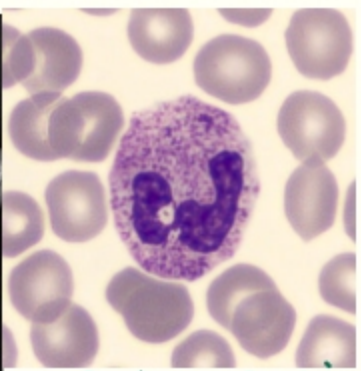
Hyaluronidase defect: Natowicz D

MPS – MR ✓

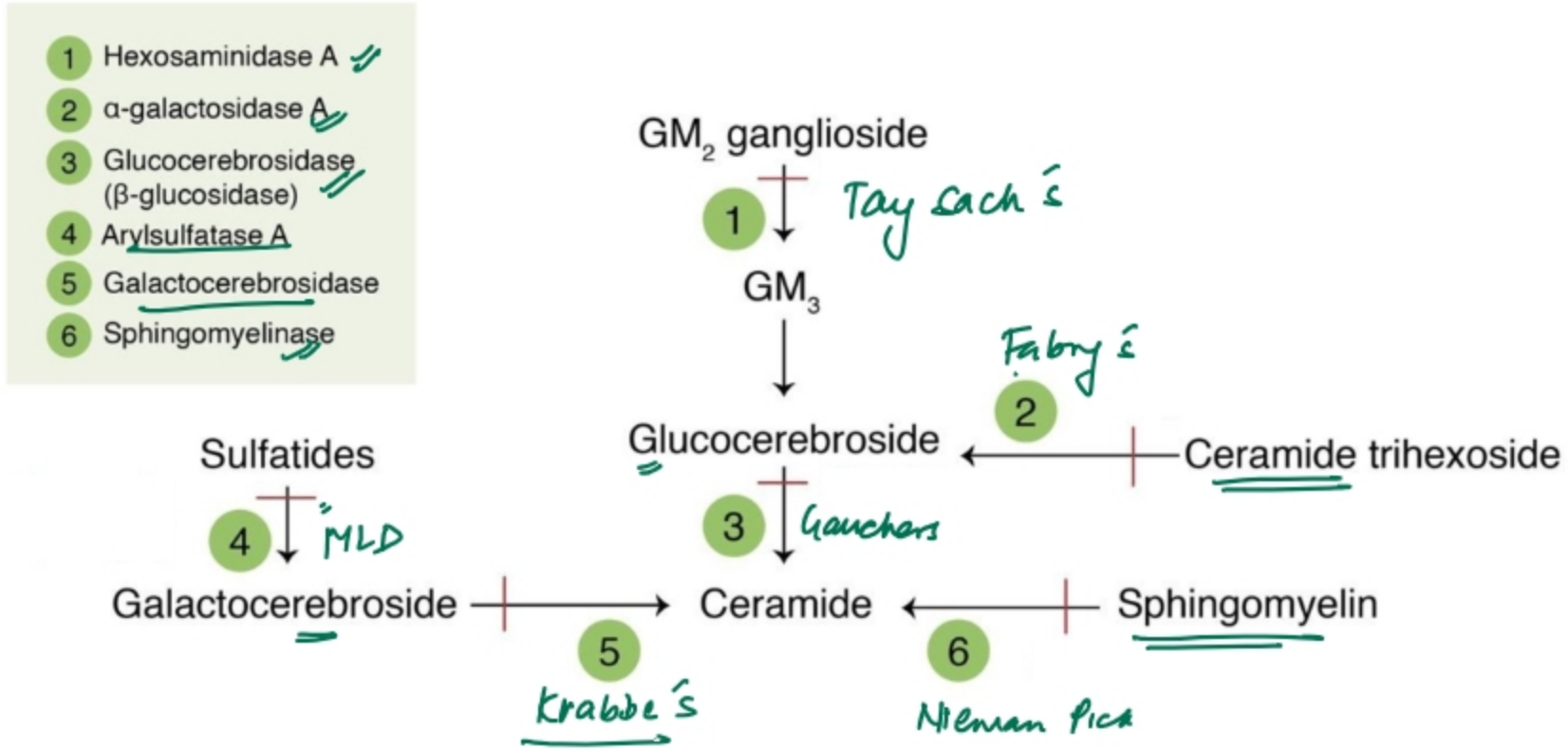
Coarse facies



bullet mc

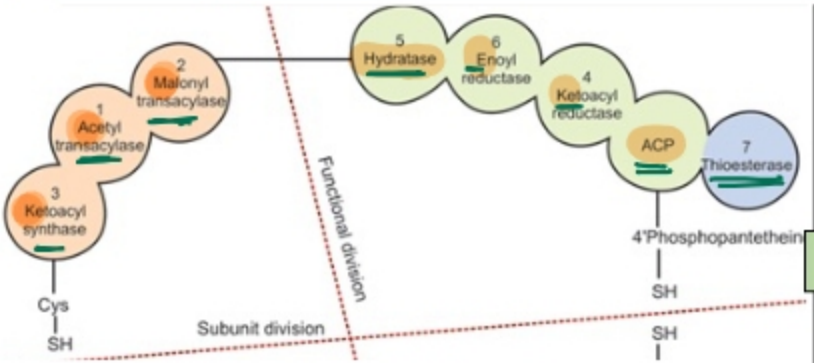


Reilley bodies



Fatty Acid Metabolism

No ATP in Alpha oxidation & Omega oxidation



Synthesis

Fatty acid synthesis
(palmitate, a 16C FA)

FA Synthase Complex → ← NADPH

Malonyl-CoA

Insulin (+) → Acetyl-CoA carboxylase ← Glucagon (−)

ATP

CO_2 *(biotin)*

Acetyl-CoA

ATP citrate lyase

Cytoplasm

Mitochondrial membranes

Citrate shuttle

Mitochondrial matrix

Citrate

Liver/adipose/
lactating mammary glands

DeH: FADH 1.5
Hydratase
DeH: NADH 2.5
Thiolase
ATP: 28 + 80 − 2 = 106 ATP

Degradation

All except Brain, RBC

Fatty acid + CoA

ATP → AMP

Fatty acyl-CoA synthetase

Fatty acyl-CoA

RLE

Carnitine palmitoyl transferase I

Carnitine shuttle

Fatty acyl-CoA

16C Palmitate (7)

β-oxidation (acyl-CoA dehydrogenases) (4)

Acetyl-CoA (8)

Fatty Acid Oxidation Defects

PEROXISOMES :

MCAD deficiency

Non-ketotic hypoglycemia
Dicarboxylic aciduria
(ω-oxidn)

Refsum D

Retinitis Pigmentosa
Icthyosis
Phytanic acid ↑

Lomitapine

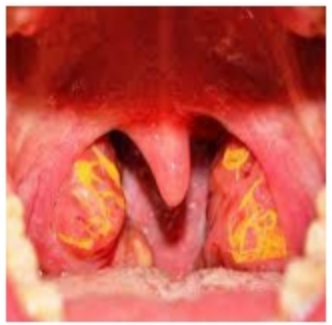
Abetalipoproteinemia

· MTTP (−)

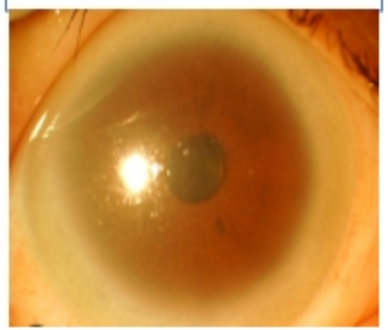
B-48 chylomicrons

HDL Normal
LDL / VLDL Absent

HDL ↓



HDL ↓
VLDL ↑↑



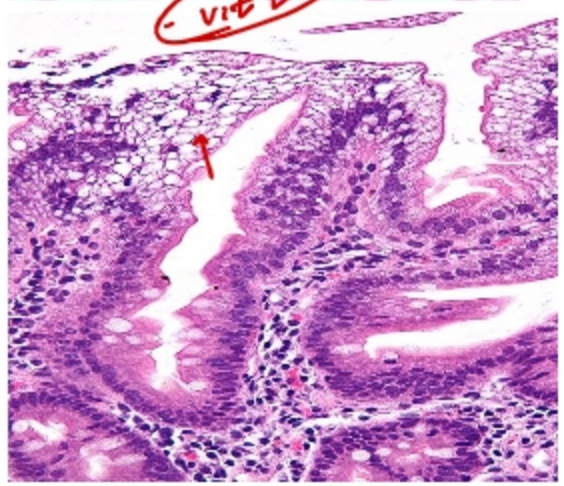
Jamaican vomiting fruit sickness

Unripe Ackee fruit
Hypoglycin

Zellweger's D

VLCFA ↑
Phytanic acid ↑
Absent Peroxisomes

· Vit E



Tangier's
ABCA1 gene.

Fish-eye D
Partial
LCAT

Complete LCAT
↳ Norum's D

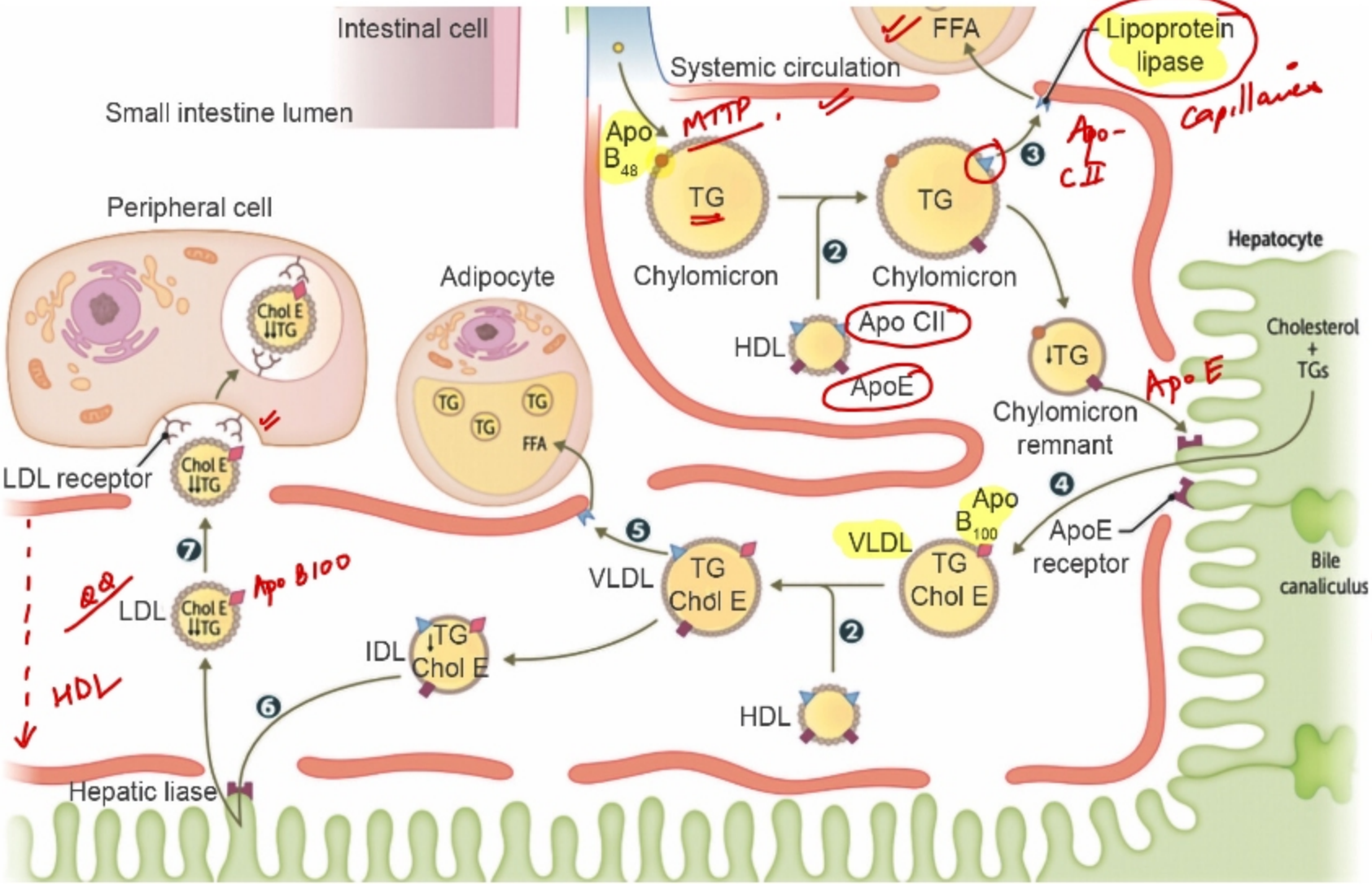
Carnitine def

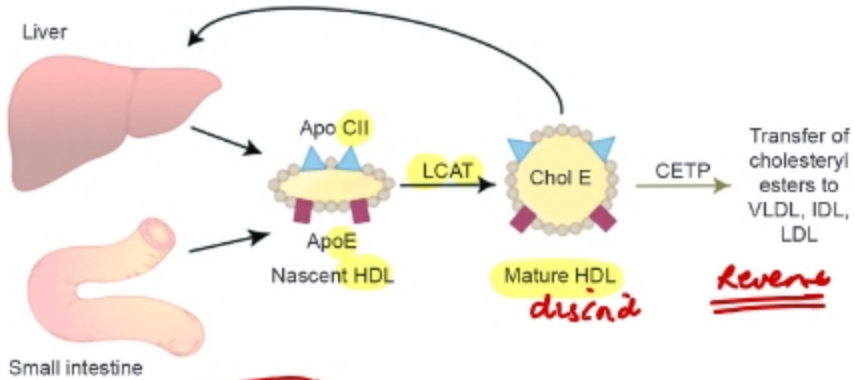
LCFA/ TAG accumulation in muscle
Skeletal myopathy, rhabdomyolysis, DCM

X-linked adrenoleukoD

VLCFA ↑
White matter abN
Adrenal insufficiency

Cholesterol Metabolism





Apo A1: LCAT activator

Apo B-48 - chylomicrons

Apo - CII - LPL +

Apo - E - Chyl remnants

Apo - B100 - LDL receptor.

Lipoproteins

Chylomicrons

- Max TG
- Exogeneous lipid
- Max size
- Min PL, cholesterol
- Min protein
- Min density

HDL

- Min TG
- Min size
- Max PL
- Max protein
- Max density

VLDL

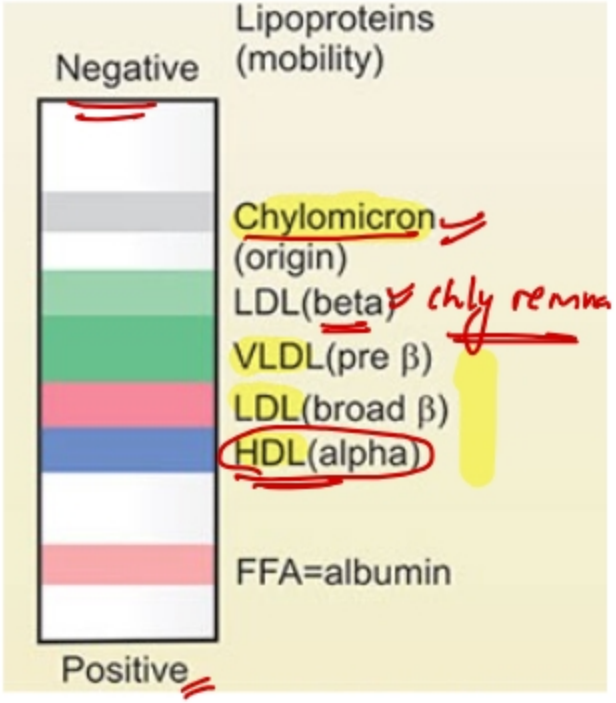
TG - Endogeneous

LDL

Max cholesterol

Friedwald F

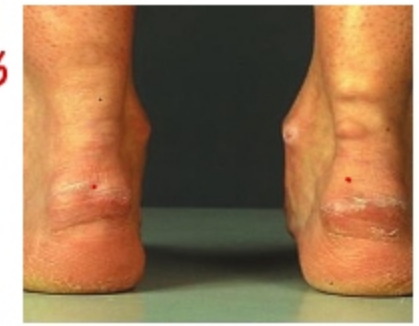
LDL = Total Cholesterol - HDL - (Triglycerides/5)

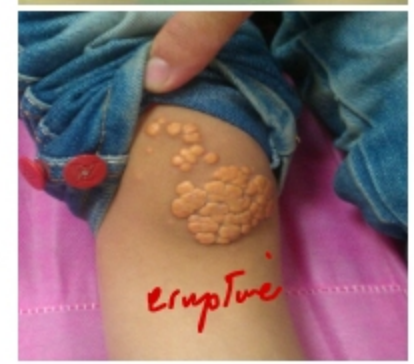


chly remna

Hyperlipoproteinemias-Frederickson classification

Type	Inherit	Pathogenesis	↑ Blood level	Clinical
I- Hyper-chylomicronemia	AR	Lipoprotein Lipase / ApoC-II deficiency	Chylomicrons, TG, Cholesterol	▪ Pancreatitis ▪ Eruptive/ Pruritic Xanthomas ▪ CREAMY supernatant
II- Familial hyper cholesterolemia	AD	Absent LDL receptors/ ApoB-100	IIa: LDL IIb: LDL + TG	▪ Accelerated atherosclerosis ▪ Tendon Xanthomas ▪ Corneal arcus, Xanthalsema (IIb) ▪ Tubo-eruptive xanthomas (IIb)
III- Dysbeta-lipoproteinemia	AR 3-E	Defective ApoE	Chylomicrons remnants, VLDL, LDL	▪ Premature atherosclerosis ▪ Tubero-Eruptive & Palmar Xanthoma ▪ Broad beta band
IV- Hyper-triglyceridemia	AD	↑↑ Hepatic VLDL	VLDL, TG Type 5: VLDL + chylomicrons	▪ Acute pancreatitis ▪ Insulin resistance ▪ Tubero-Eruptive Xanthoma





Amino Acids

Basic: HAL → Histidine / Arginine (most) / Lysine

Acidic: Glutamate / Aspartate

Imino acid: Proline

-OH containing: Serine / Threonine / tyrosine

Branched Chain: VIL - Valine / ISOL / Leucine → MSUD

Sulphur containing: Methionine / cysteine

Ketogenic only: Leucine / Lysine → PDH deficiency (XLD)

21st / 22nd: Co-translational modifn selenocysteine (UGA) / pyrollysine (UAG)

Aromatic / UV Light maximum 280nm: tryptophan

Universal Methyl donor: SAM - Methionine

Essential: PVT TIM HALL (Phe / valine, tryptoph / threonine, ISOL, Methion, Hist, argine, Lysine, Leucine)

Semi-essential/ Max buffering capacity: Histidine

Millon's Reaction, Folin-Ciocalteu's test: tyrosine (phenolic)

Hopkins-Cole reaction: tryptophan (indole ring)

Gyrate atrophy: ornithine ↑ (ornithine aminotransferase) → night blindness

Glycine + arginine + methionine: Creatine

Glycine + cysteine + glutamate: Glutathione

Lysine + methionine: Carnitine

-OH

- Bind phosphate
- Covalent modification
- O-glycosidic bonds

Glycine

- Serine
- Purines
- Heme
- Creatine
- Glutathione
- Glyoxalate -> glycine (aminotransferase) — 1° hyperoxaluria → Ca oxalate

Arginine

- NO = EDRF
- Creatine

Glutamate

B6 → GABA

Histidine

B6 → Histamine

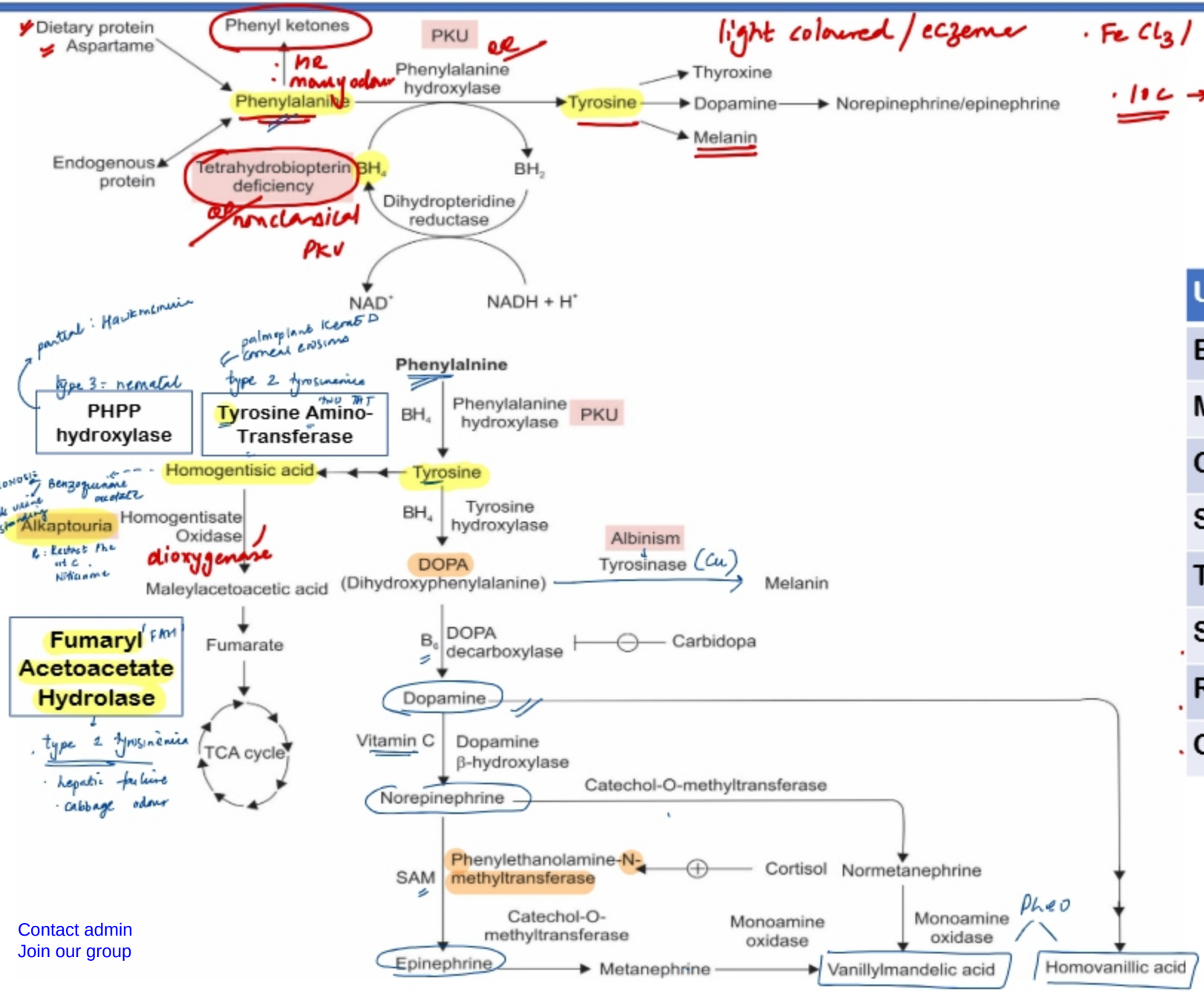
- Thyroxine
- Dopamine
- Catecholamines
- Melanine

TYROSINE

- Niacin
- Melatonin
- Serotonin

TRYPTOPHAN

Amino Acid Disorders

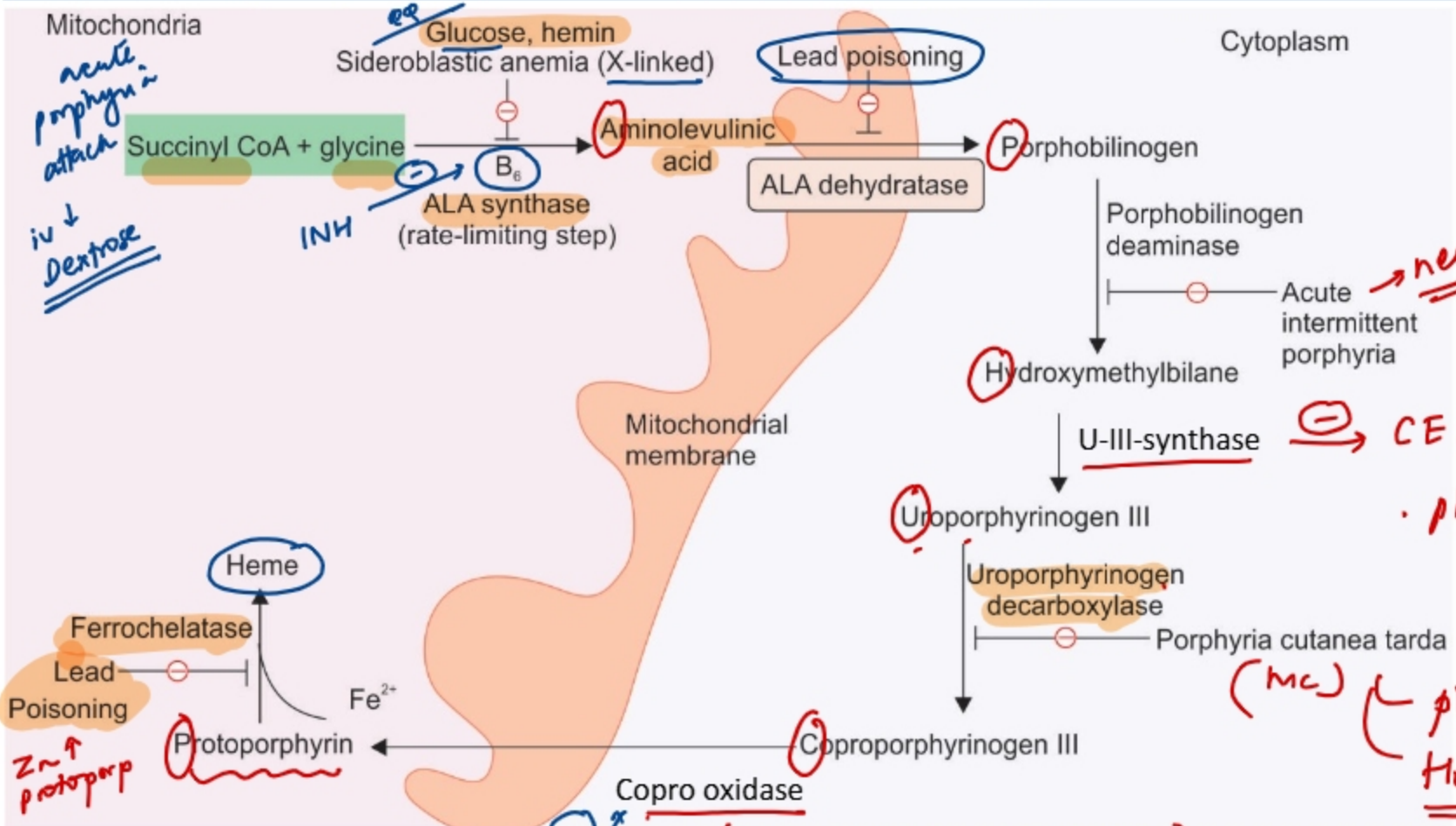


Garrod's tetrad:
- Cystinuria
- Alkaptonuria
- Albinism
- Pentosuria

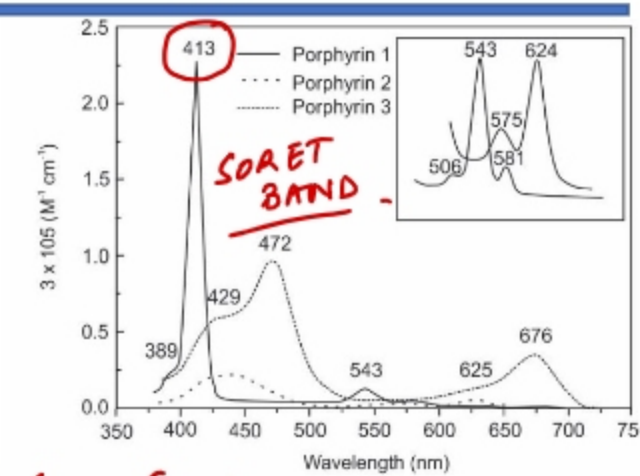
URINE ODOUR	CONDITION
Burnt sugar	MSUD
Mousy	PKU
Cabbage	type 1 tyrosinemia
Swimming Pool	Hawkinsinuria
Tom cat urine	Multiple carboxylase (B7)
Sweaty feet/ Cheesy	Isovaleric acidemia
Rotting Fish	Trimethyl amnuria
Oasthouse	Methionine malabsorp

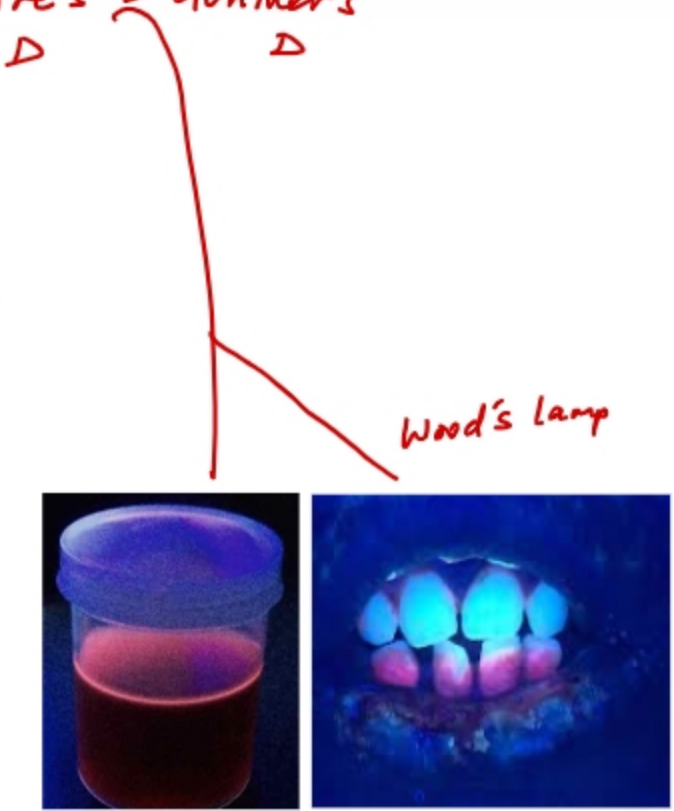
Heme Synthesis & Porphyrias

A → P → H → U → C → P
Pb AIP CEP PCT Her



acute porphyria attack
iv ↓ Dextrose
neurovisceral
CEP / vampire's = Gunther's
. photosn ↑↑
(MC) photosn ↑↑ Hep C
Hereditary Coproporphyria
Pb
APH - UCP

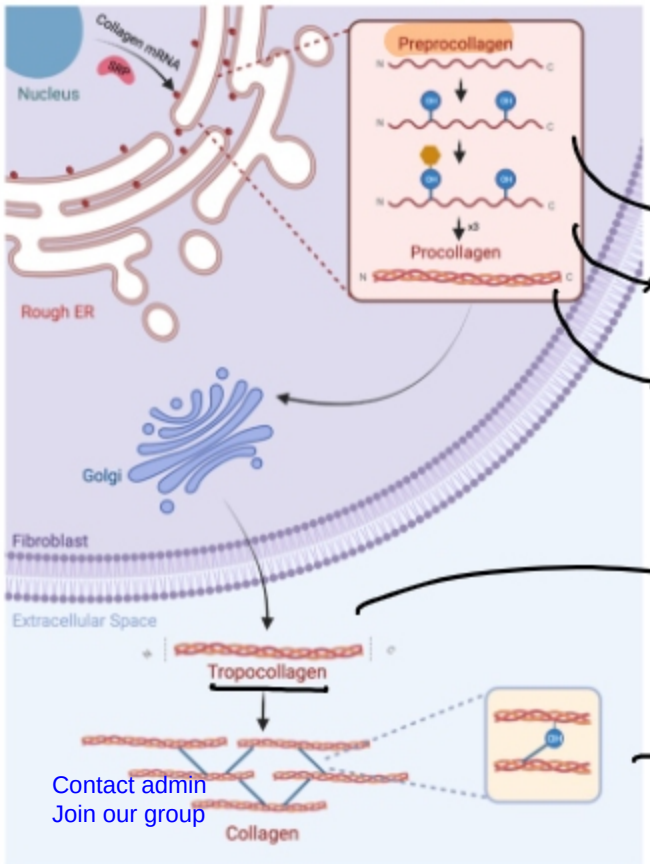




$$\text{heme} + \text{NADPH} + O_2 \xrightarrow{\text{heme oxygenase}} \text{biliverdin} + CO + Fe^{2+}$$

Collagen Synthesis & Disorders

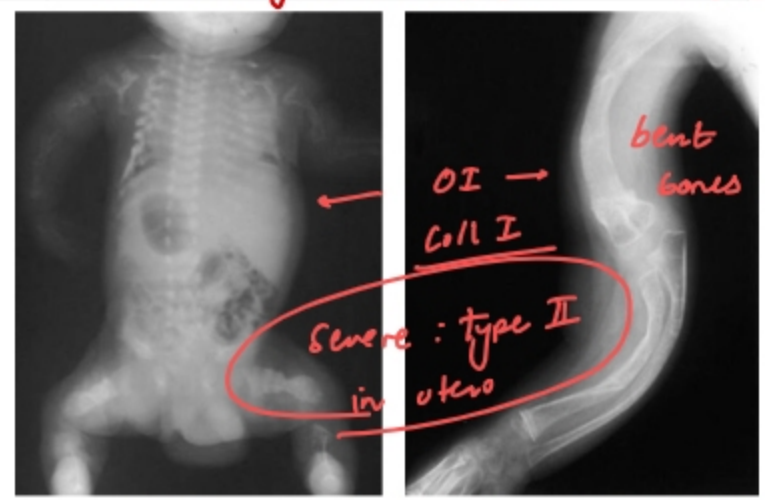
Type I(mc)	Bone, Tendon, Skin, Dentin, Cornea, Late wound repair, Fibrocartilage	Absent in: OI (AD)
Type II	Hyaline Cartilage, nucleus pulposus	
Type III	Reticulin, blood vessels, uterus, fetal tissue, early wound	Deficient in: EDS (vasc type) Classical type: type V coll
Type IV	Basement membrane (glomerulus), cochlea, lens	Defective in: ALPORT
Elastin	Skin, lungs, arteries,, epiglottis, ligamenta flava	Defective in: Marfan's (Ghent criteria)

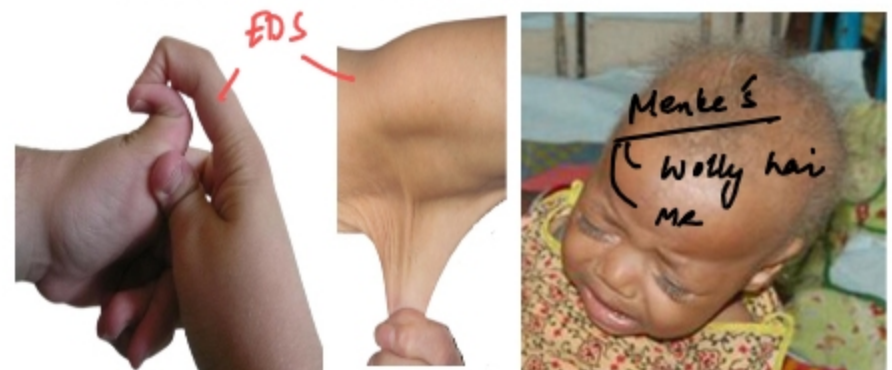


Collagen chain: Gly-X-Y (1/3)
Disrupter: Proline

Hydroxyline
Hydroproline

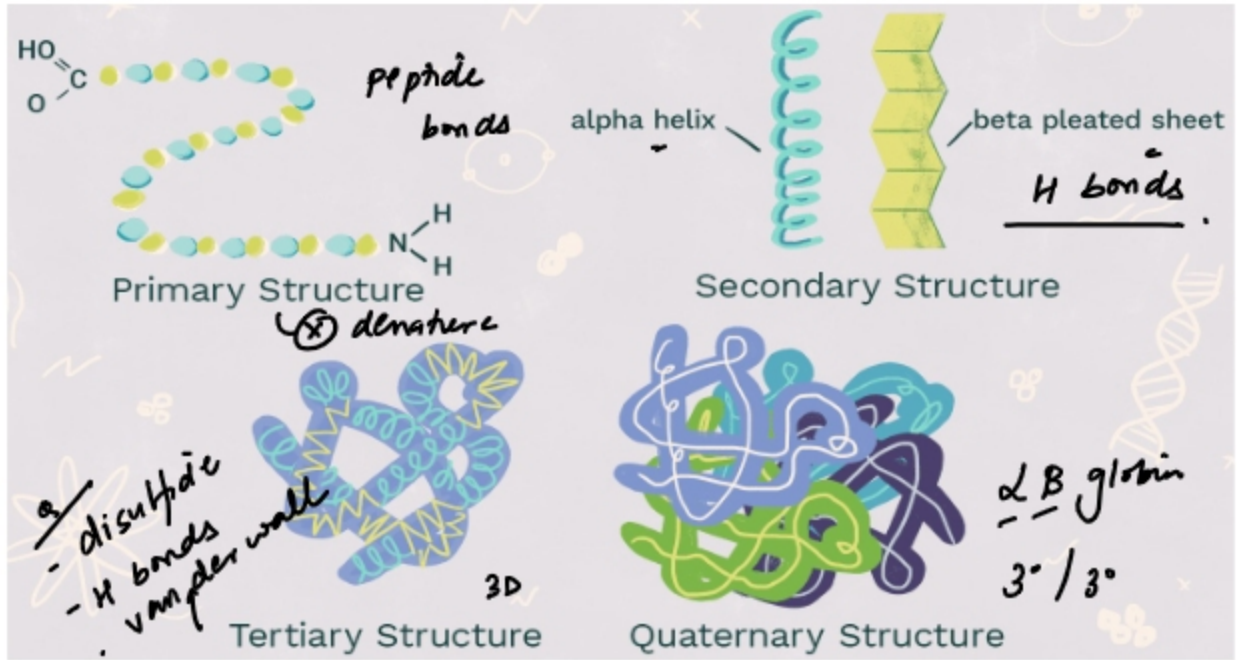
1. Hydroxylation ← vit C (scurvy)
2. Glycosylⁿ
3. Triple helix ← OI
4. cleavage N-C terminal ← EDS
5. cross-linking ← lysyl oxidase (Cu) ← Menkes kinky hair





Miscellaneous

Structure	Methods
Primary	Sanger's sequencing
	Edman's sequencing
	Reverse sequencing
Secondary	Optical rotator dispersion
	Ocular dichroism
Tertiary	X-ray crystallography < Cryo-electron
	UV spectroscopy, NMR spectroscopy



GAG : Hexosamine + Uronic acid (No lipid)

- Most abundant: Chondroitin SO4
- GAG with no protein linkage, No sulphate, Longest: Hyaluronic acid
- Cell migration during morphogenesis, Wound repair: "
- GAG with NO Uronic acid, Corneal transparency: keratan SO4
- Sclera, Atherogenic (LDL binding): Dermatan SO4
- LPL on endothelial surface, Plasma membrane receptor, GBM charge selectiveness: Heparan SO4

Miscellaneous

Metal	Metalloenzymes
Zn	Carbonic Anhydrase, Glutamate DH, LDH, ALA dehydratase, MMP, Zn finger DNA motifs: Steroids Deficiency (SLC39A4): Acrodermatitis enteropathica Hypogonadism ↓ wound healing
Cu	Tyrosinase, Cyt c oxidase, Cytoplasmic SOD, Ceruloplasmin, Lysyl oxidase → ATP7A → Menke's (Mn-mito) ATP7B - Wilson D
QQ Se	Glutathione peroxidase, Deiodinase, Thioredoxin reductase Deficiency: Keshan's (Q party) Excess: Alkali disease of livestock
Mo	Xanthine Oxidase, Sulfite Oxidase (Mo XO SO) XO XO XO
Chr	Potentiates action of insulin Q
Ni	Urease

All kinase/ phosphorylase/ carboxylase use Mg except pyruvate kinase- K

Catalyse the same reaction
Different structure, kinetics → Isoenzymes

- O- Oxidoreductase
- T- Transferase - kinase / glycogen PO_4 ase / "trans"
- H- Hydrolase amylase / lipase
- L- Lyase → decarboxylase / hydratase
- I- Isomerase
- L- Ligase - Carboxylase / Synthetase
- T- TRANSLOCASE → ATP Synthase

QQ
- HbA1C: ion-exchange chromatography ~120d
- Fructosamine: ~20d — glc control

- Highest thermic effect: Prtn
- RQ CO_2/O_2
- Carbohydrates: Q

	RQ	Caloric
Carbohydrates:	1	4 kcal/g
Fat:	0.7	9 kcal/g
Protein:	0.8	4 kcal/g
Alcohol	–	7 kcal/g

Miscellaneous

1

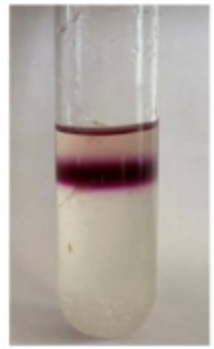
Benedict's
- reducing sugars in urine

NO REST - trehalose
Sucrose

2

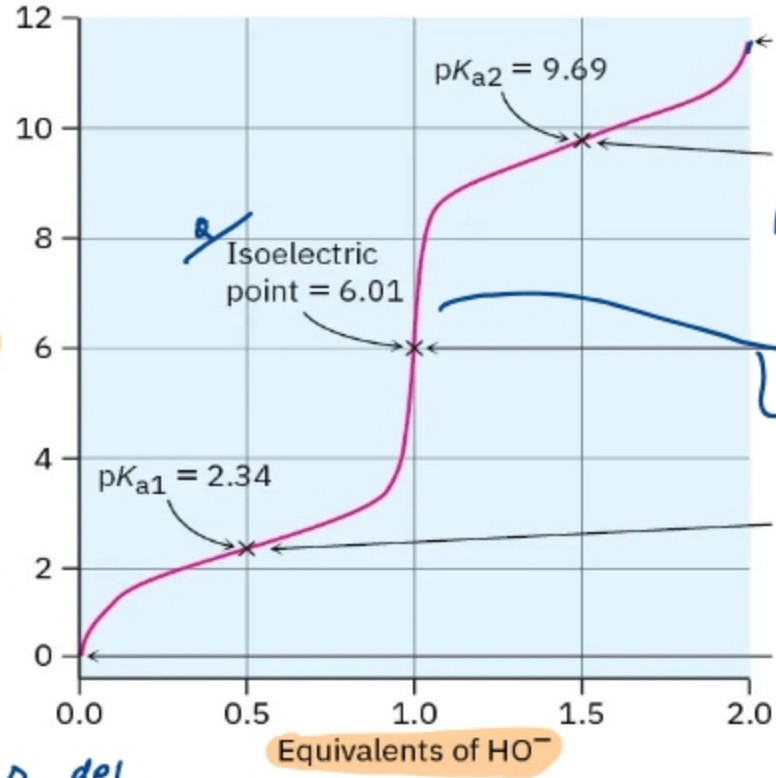
Δ test
Petrunia

3



Rothera's Nitroprusside test
- KB urine
(× β-DH butyrate)

+	−	+	: DKA
−	−	+	: Starv^n
−	−	−	: MCHAD def.



pk : max buffering achia

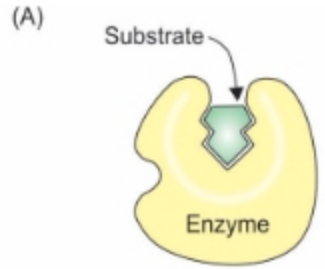
$NH_2 - C - COOH$ (C: 2 ionisable side chains)

net charge = 0

Max ppt of a prin

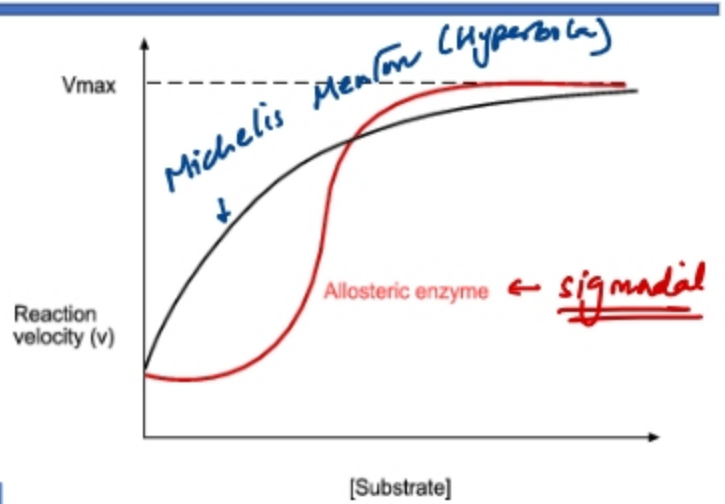
- Inhibition of glycolysis by O2 : Pasteur
- Lactic acidosis if glucose concentration increased in presence of O2 : Crabtree } Yeast
- Aerobic glycolysis: Warburg effect
- Direct positive Van Der Bergh's reaction: obstructive jaundice
- Arginine not lipotrophic (choline lecithin methionine Vit B12/ folate) = fatty liver
- Avoided in fish odour syndrome : CHOLINE
- Gums, Pectin, Mucilage (soluble): useful - DM

Enzyme Kinetics

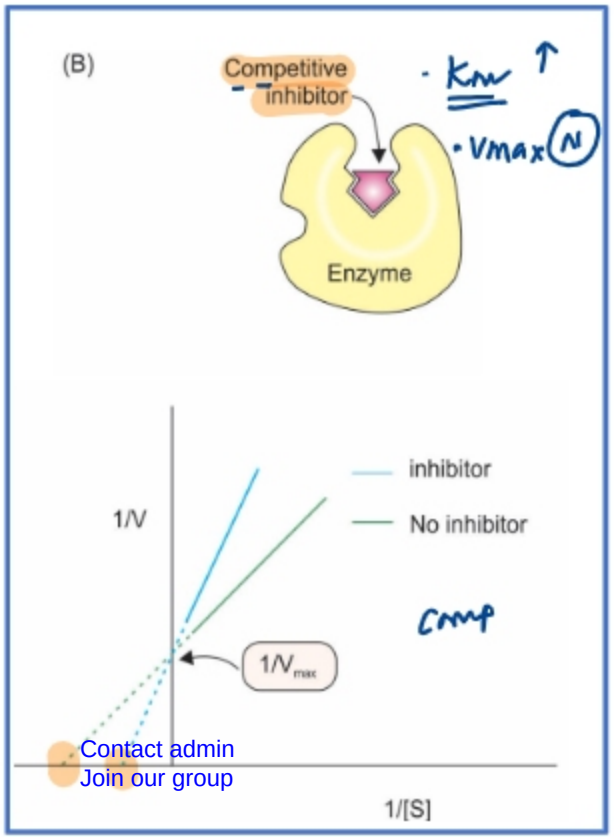


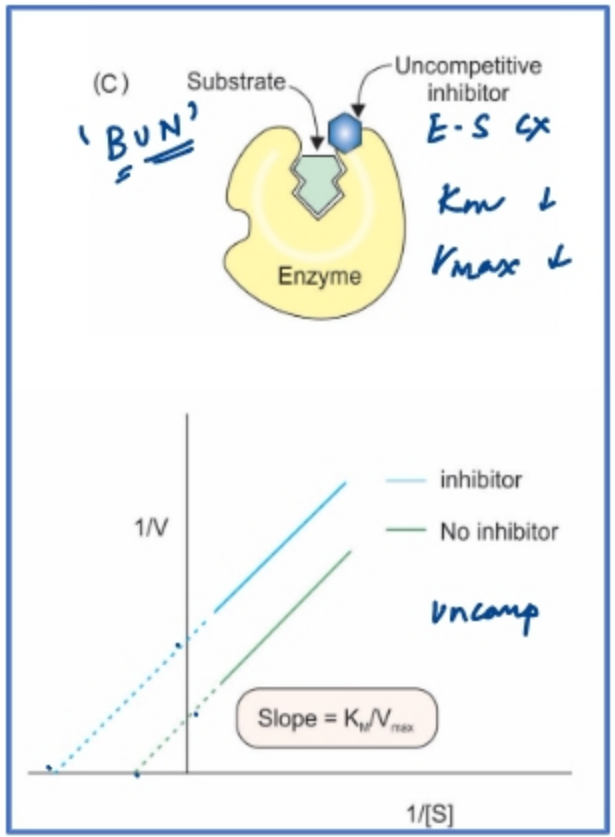
- Lock & Key: Emil Fischer
- Induced fit : Koshlands

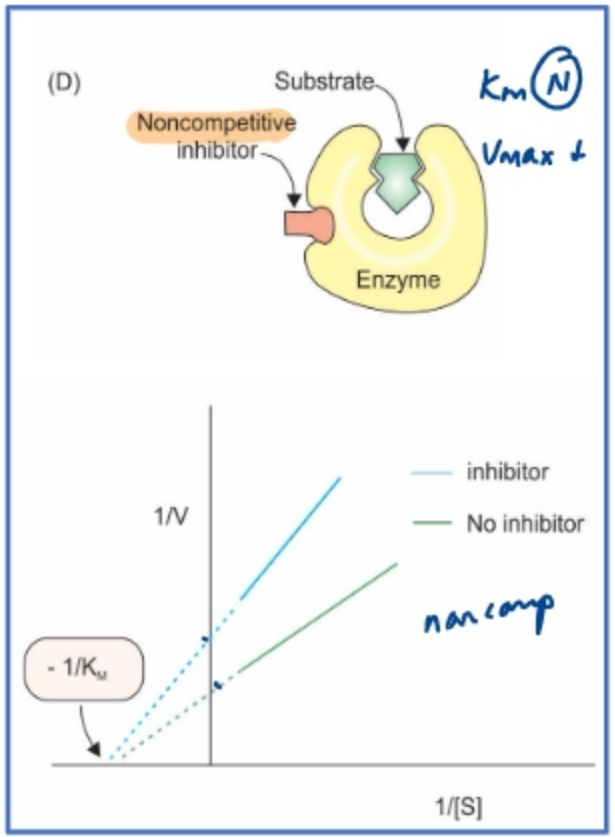
Kcat/Km: efficacy

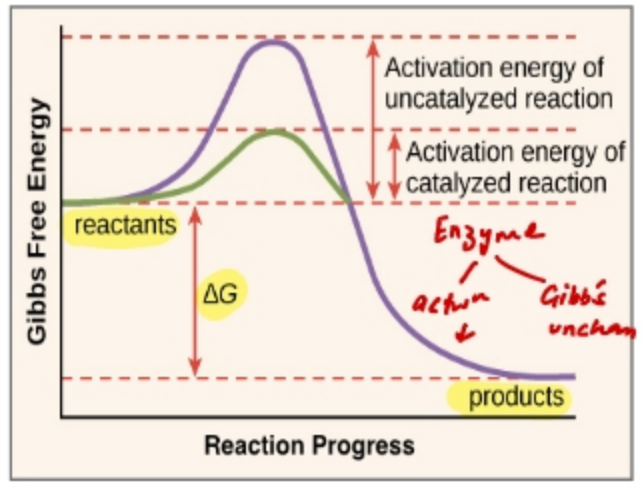


mixed : Km ↑ Vmax ↓









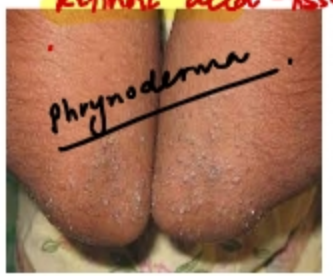
Vitamins & Deficiency Disorders

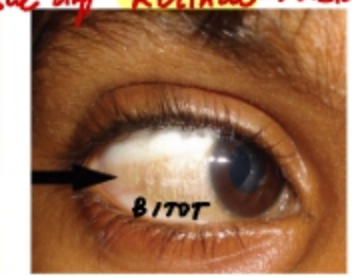
ADEK - fat B/C - water

vit A

Px: Golden rice (fortified)

Retinoic acid - tissue diff Retinal → rhodopsin → vision





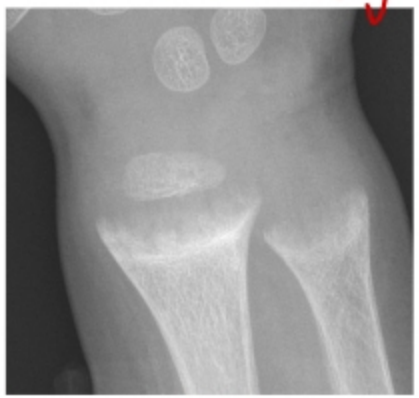
Stage	Features (6m-6yr)
XN	Night blindness – earliest clf → >1%
X1A	Conjunctival Xerosis – earliest sign
X1B	Bitot's spots – most sp sign → >0.5%
X2	Corneal xerosis
X3A	Corneal ulceration < 1/3 surface → >0.01%
X3B	Corneal > 1/3 surface
XS	Corneal scar → >0.05%
XF	Xerophthalmic fundus

vit A excess → teratogenic, pseudo tumor cerebri

Vit-A: Day 0,1,14
<1yr: 1 Lakh
>1yr: 2 Lakh

vit D

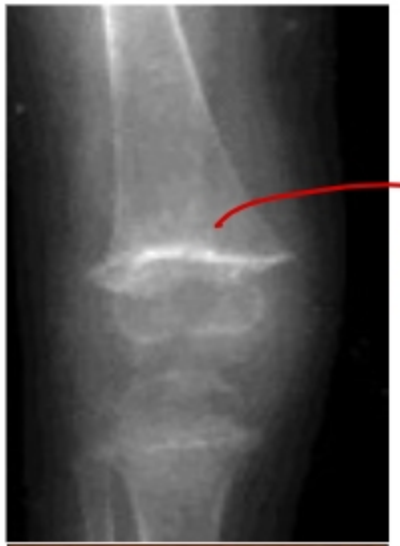
↑Ca / PO_4 ← intestine, bone, kidney



copping, splaying, fraying

vit C

Scurvy – collagen ↓



White line of Frenkel

perifollicular hge: corkscrew hair

bleeding gums

vit C ↑↑ → Increases oxalate → Ca oxalate ↑
Increases Fe

vit K

↓ glutamyl carboxylase ↓

↑ 2 | 7 | 9 | 10 (+)

Prth c/s (+)

(–) vit K epoxide reductase

Warfarin

vit K (↓): Hgic disease of newborn

vit K1 inj 1mg iv

vit E

- antioxidant
- Tocopherol

(↓) Hemolytic anemia (acanthocytes / spur cells)

Dorsal column

Spinocerebellar tracts

- mimics SACD (B12)
- Friedrich ataxia

vit B1 : Thiamine

Assay: RBC transketolase

- alcoholics
- polished rice

Be APT

Branch Chain Keto-DH:
MSUD - VIL ↑

Alpha Keto-Glutarate DH

PDH — oxidative decarboxy"

Transketolase

Beri-Beri
- Wet- HOCF (edema)
- Dry- neuropathy (sm)

G O A

Wernicke: Global Confusion, ophalmoplegia, ataxia
dorsomedial thalamus, MB
↓x

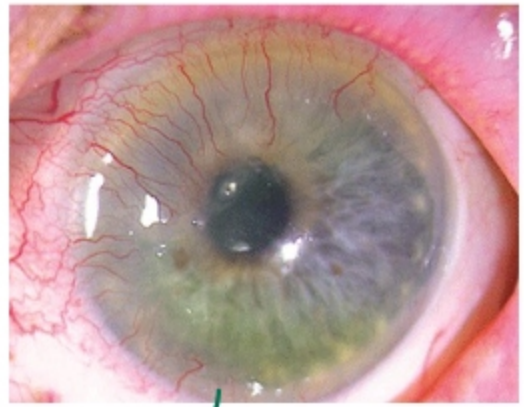
Korsakoff: AGA / RGA + confabulations

Lactic acidosis
alcoholic : B1 → 5% dextrose

vit B2 = Riboflavin

Assay: Glutathione reductase

FMN/FAD

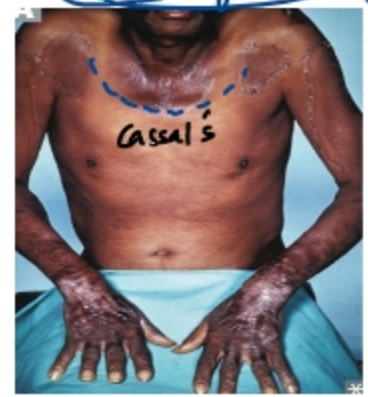


. Corneal neovascular"
. cheilitis
. magenta tongue

vit B3 = NIACIN

Assay: RBC NADH/NAD

Pellagra
Diarrhea
Dementia
Dermatitis
photo-sn



60 mg tryptophan — B2 / B6 → 1 mg of niacin (B3)

Serotonin
Melatonin

2 x 3 = 6

- **Hartnup** (SLC6A- Tryptophan transporter) Obermeyer test
- **Carcinoid**
- **Maize** < Leucine (↓ +)
- **INH** B6 ⊖

vit B5 = Pantothenic

CoA

. Burning feet disease
. adrenal insufficiency

vit B6 = Pyridoxine

Assay: Xanthurenic acid ↑

vit B7 = BIOTIN

carboxylases

pyruvate → OAA

acetyl CoA → malonyl CoA

propionyl CoA → Methylmalonyl CoA

vit B9 = Folate

① Junk food, alcoholics, IAED

Assay: Histidine load test

vit B12 = Cobalamin

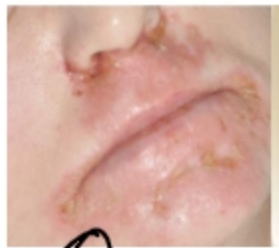
① - vegans

kuncle ? pigment

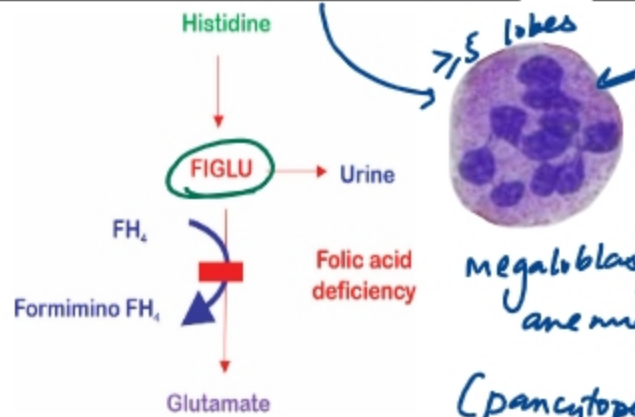
Assay: MMA ↑

Co-Factor:

- Decarboxylases
- ALA synthase
- Glycogen phosphorylase
- Transamination Histamine GABA



↓ → Dermatitis, Alopecia



megaloblastic anemia

(pancytopenia)

>/5 lobes

SACD:

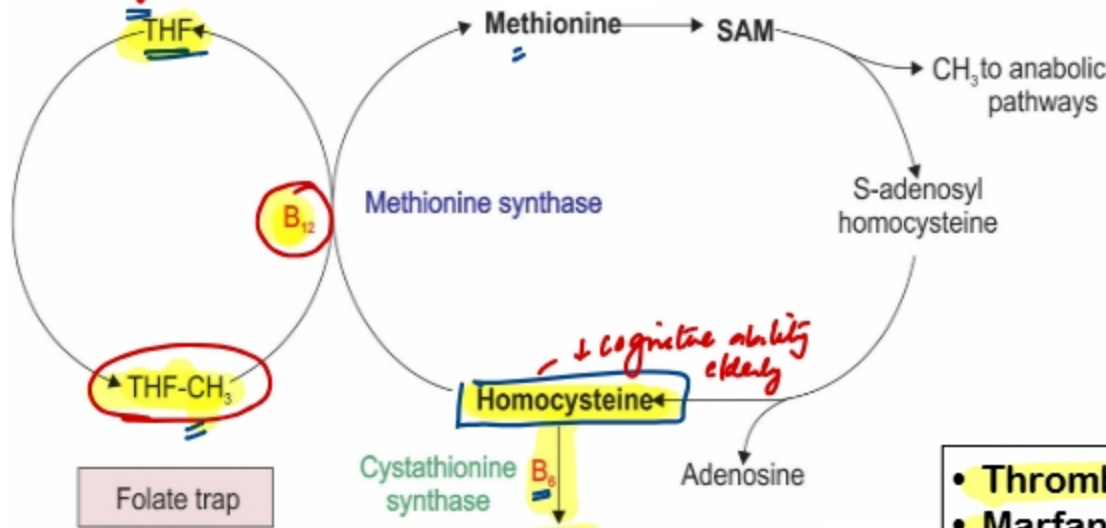
Dorsal columns + UMN

↓ proprioception, toe touch ↓ LC/S

+ neuropathy (LMN)

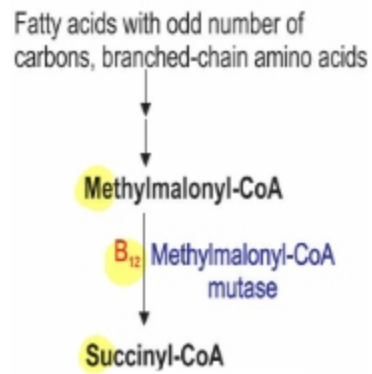
- Hypoglycemia
- Convulsions, Neuropathy
- Sideroblastic anemia

Rx of refractory sz: Pyridoxine



V → T

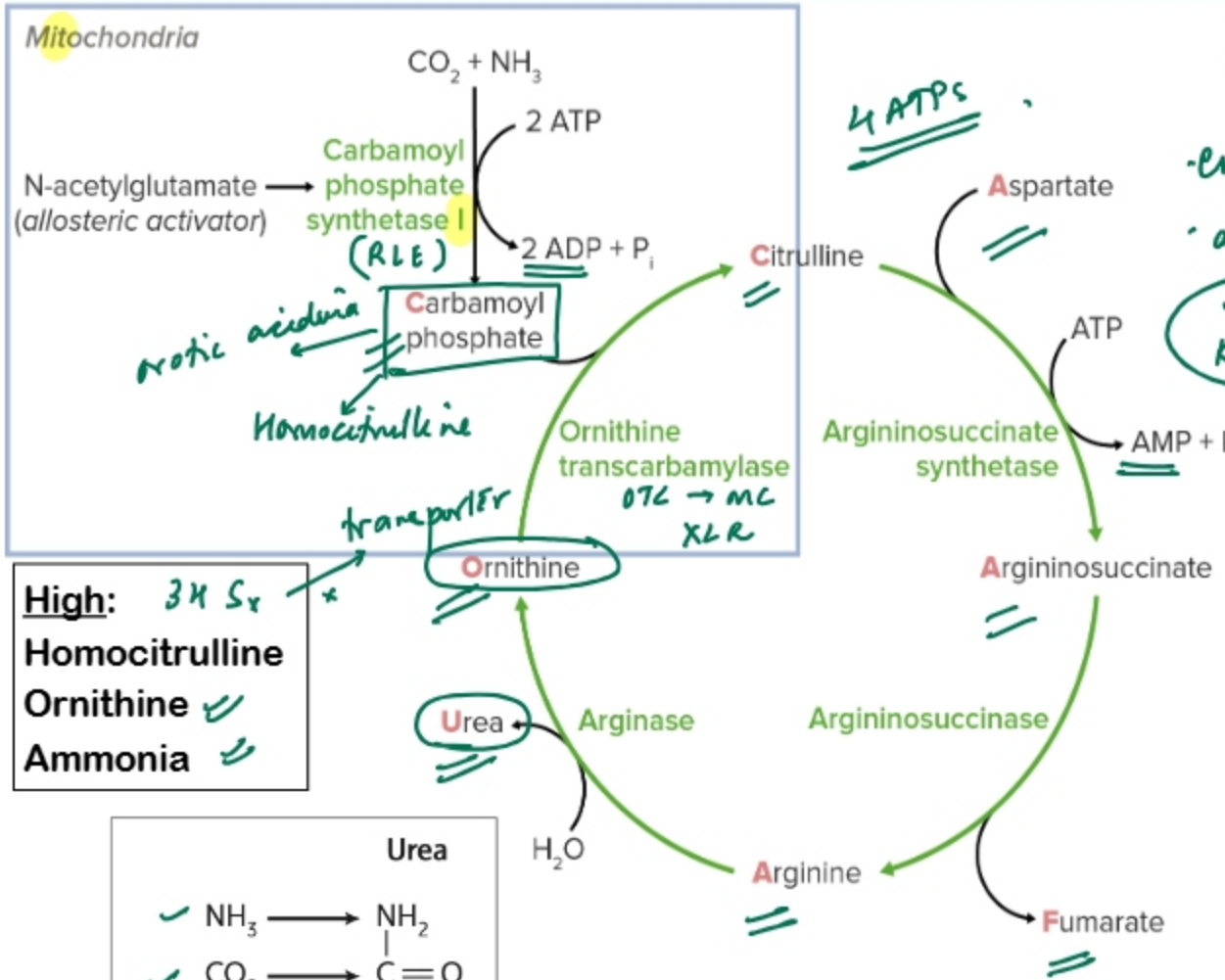
↓ cognitive ability elderly



- Thromboembolism, MI
- Marfanoid Habitus
- Ectopia lentis: inferonasal
- Cyanide nitroprusside test

vs Marfan - superotemp

Urea Cycle & Defects



4 ATPs

↑ NH_3
- Encephalopathy
- asterixis

↓ ATP
Krebs cycle

↑ creatin

resp alkalosis

CPS-1: Urea cycle- Mitochondria

CO_2 + NH3

CPS-2: Pyrimidine synthesis-Cytoplasm

CO_2 + Glutamine

Orotic Aciduria :

- ↑ NH_3 — OTC deficiency
- Megaloblastic anemia — UMP synthase

orotic aciduria

OTC → MC
XLR

High: 3 H Sx
- Homocitrulline
- Ornithine
- Ammonia

R: N-scavengers

Ordinarily, Careless Crappers Are Also Frivolous About Urination

- Sodium Benzoate a/ - Glycine → Hippuric acid
- Phenylacetate, Phenylbutyrate a/ - Glutamine

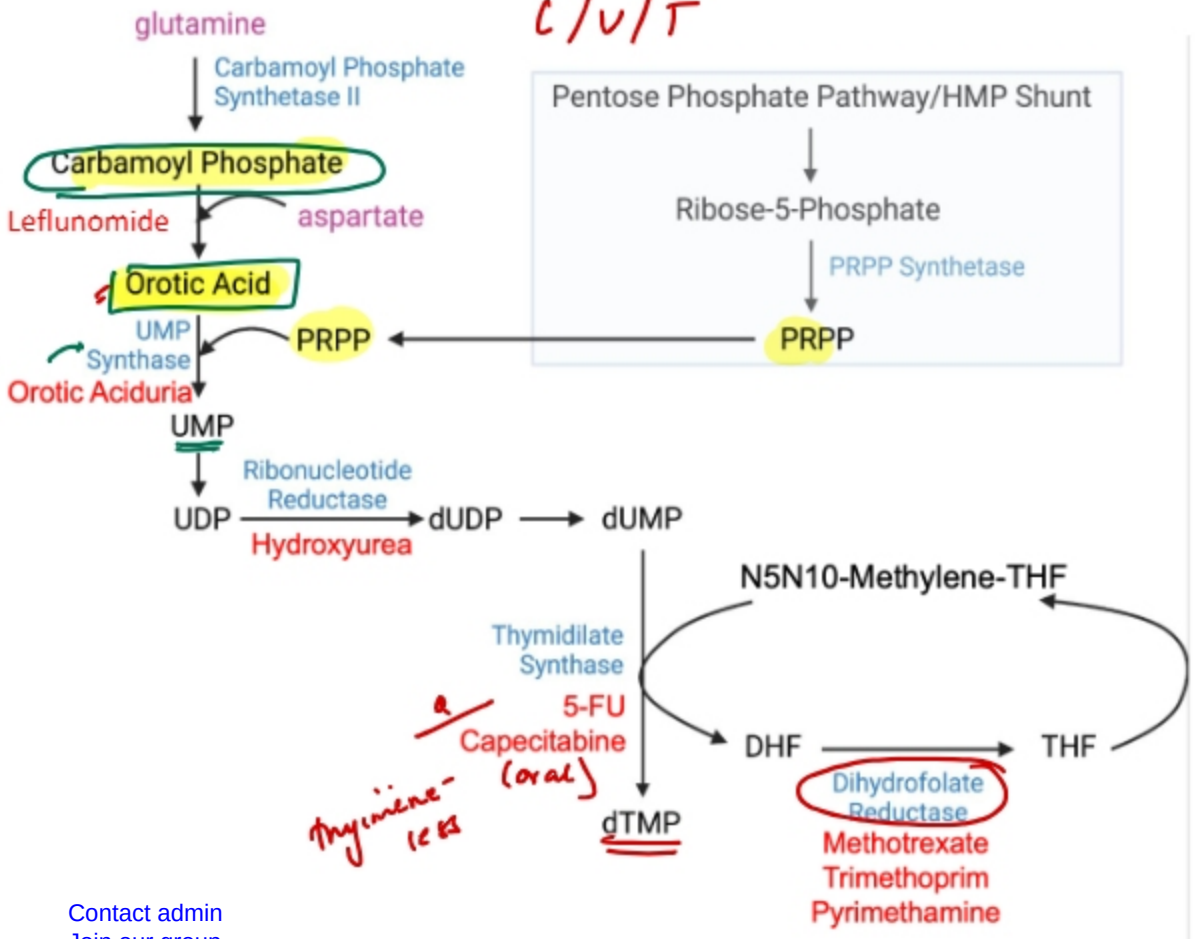
- Main ammonia transporter in body: Glutamine
- Main ammonia transporter in muscle: Alanine

Nucleic Acids: Purines & Pyrimidines

Glycine + Glutamine + Aspartate: Purine (A G)
Glutamine + Aspartate: Pyrimidine (C U T)

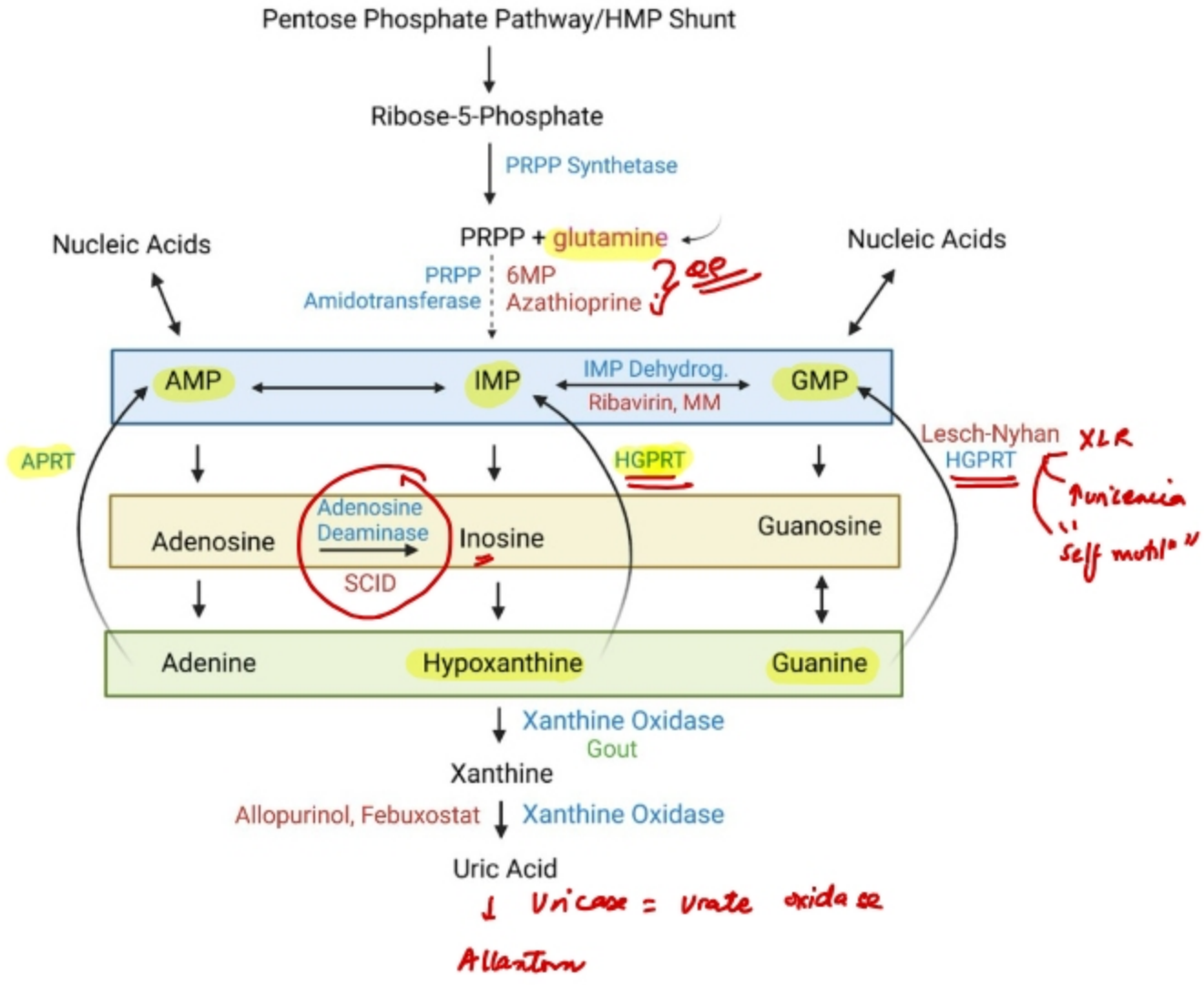
Deamination:	**Methylation:**
C → U	U → T

C / U / T

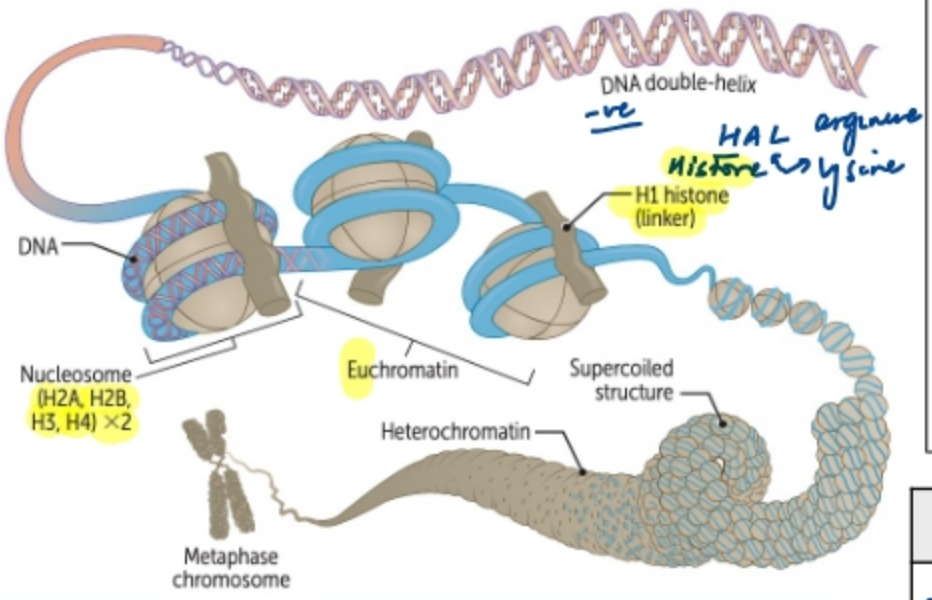


Hybridoma technique: Salvage pathway

Monoclonal Ab → Myeloma cell + B-cell — HAT medium



Basics of DNA

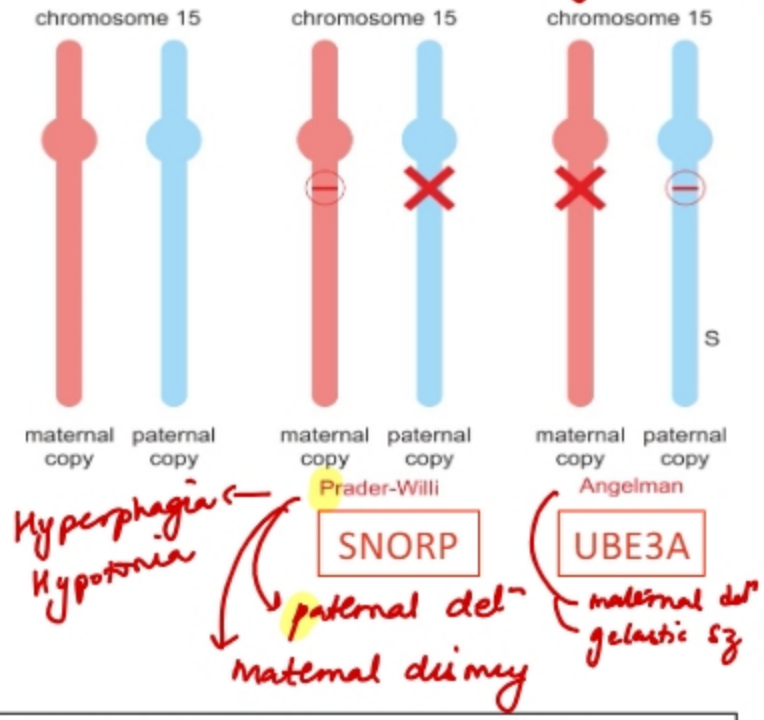


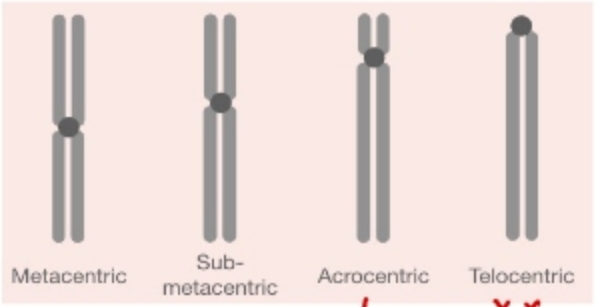
Epigenetics

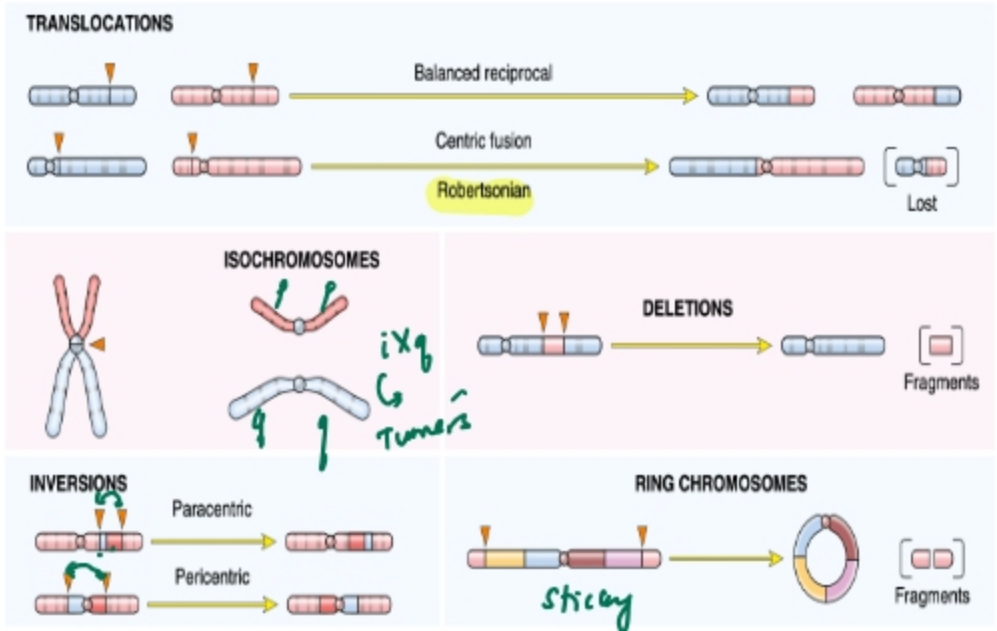
- Chemical changes in DNA
- Transmissible? ✓
- Reversible? ✓
- Sequence change? No
- * DNA methylation : mutes
- * Histone acetylation : activates
- * Histone deacetylation : deactivates

Euchromatin	Heterochromatin
active, light, loose	inactive / dark / tight

Genomic Imprinting

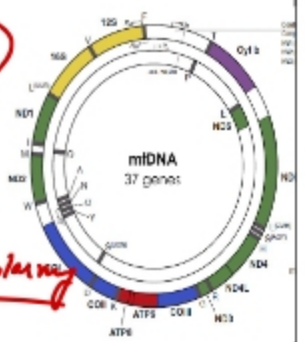




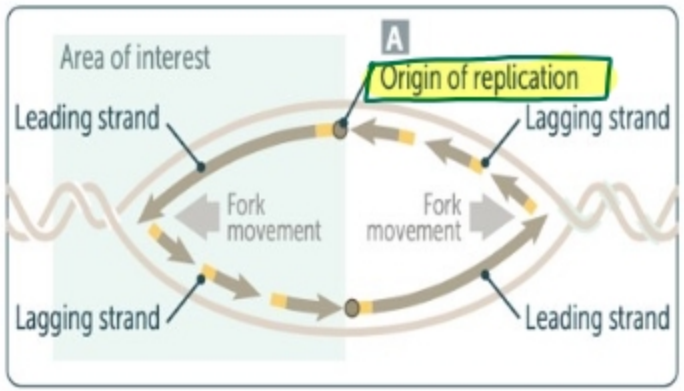


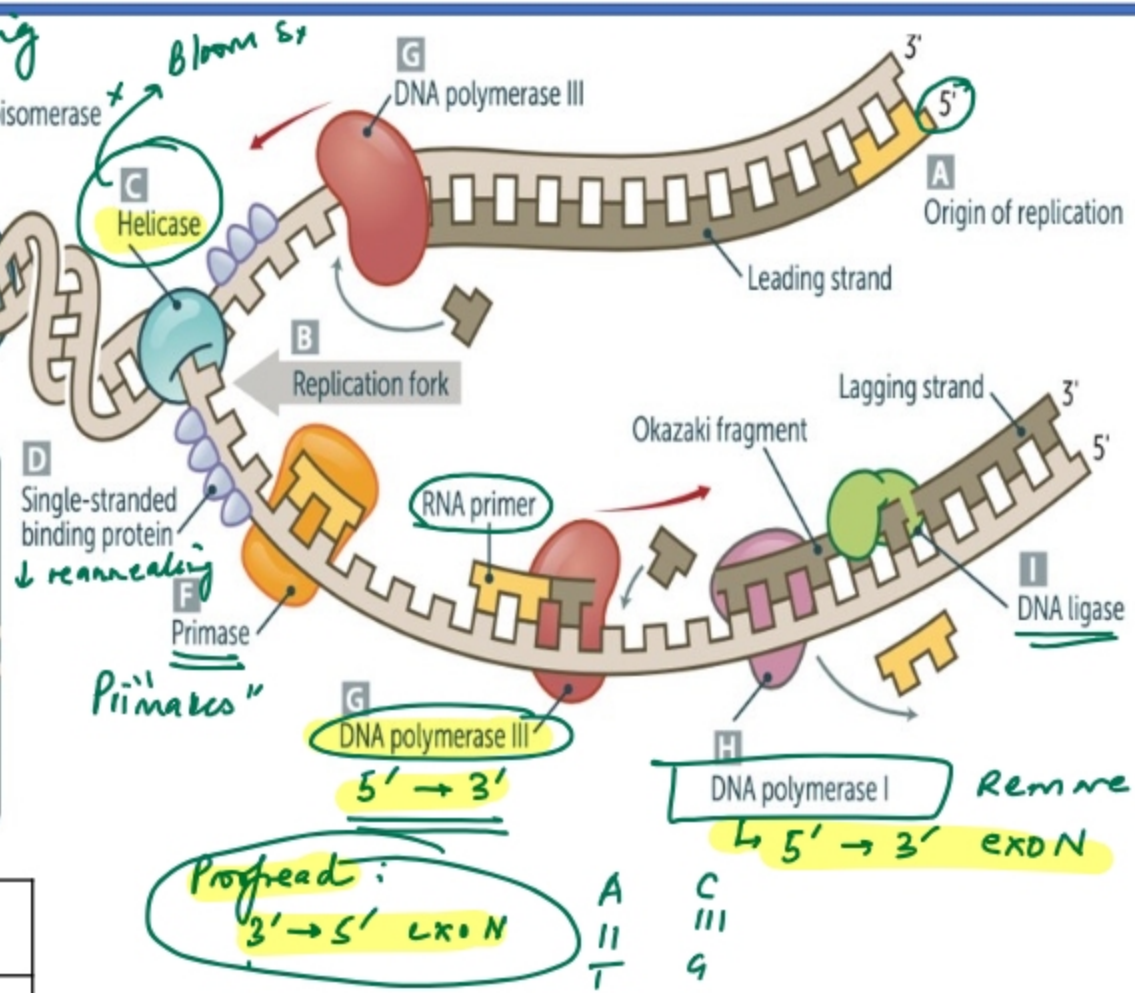
Mitochondrial DNA

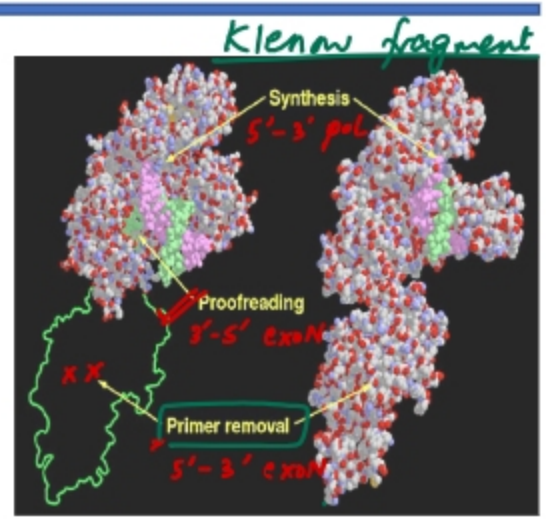
- 1 % of total DNA
- Inheritance: Maternal to all
- ETC proteins: 13 (20%)
- Copies: 2-10 copies – heteroplasmy
- DNA pol: γ
- Proofreading? No
- Histones? No



DNA Replication





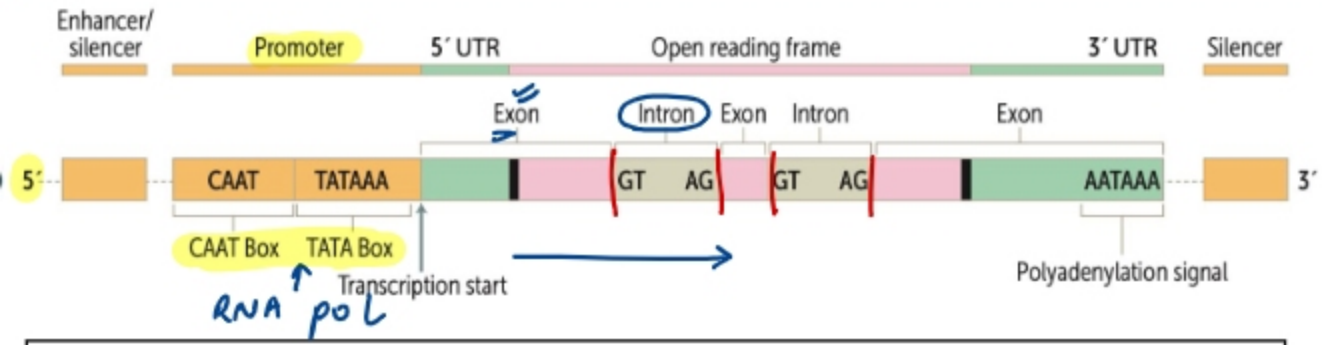


Pol α	Primase activity
Pol β	Repair process
Pol γ	Mitochondrial DNA synthesis
Pol δ	Lagging strand synthesis [Okazaki fragment]
Pol ε	Leading strand

Germ cells: Telomerase — Not Ribozyme
- TTAGGG
- RNA dep DNA polymerase
- Hayflick limit: Somatic cells 30-50

Ribozyme
- Peptidyl transferase
- snRNA
- Ribonuclease P

Transcription



Eukaryotes

- **RNA polymerase I: Makes rRNA**
- **RNA polymerase II: Makes mRNA, microRNA, snRNA**

Amanita phalloides — death cap mushroom

- **RNA polymerase III: Makes 5S rRNA, tRNA**
- **Prokaryotes:** RNA polymerase- *Rifampicin*

Q. The base sequence of the strand of DNA used as the template for transcription has the base sequence 5′ GATCTAC 3′. What is the base sequence of RNA product?

A. CUAGAUG

B. GTAGATC

C. GTAGATC

D. GUAGAUC

template : antisense 5′ GATCTAC 3′

coding = sense 3′ CTAGATG 5′

mRNA 3′ CUAGAUG 5′

hnRNA → mRNA

Post-transcriptional changes

- 7 methyl-Guanosine Cap at 5′ end (Not in tRNA / rRNA)
- Polyadenylation (200) at 3′ end (Not in histones)
- Splicing out of introns (by snRNA)

1 gene — 1 protein

exceptions

RNA editing

aa 3′→5′

Alternate RNA splicing

↓

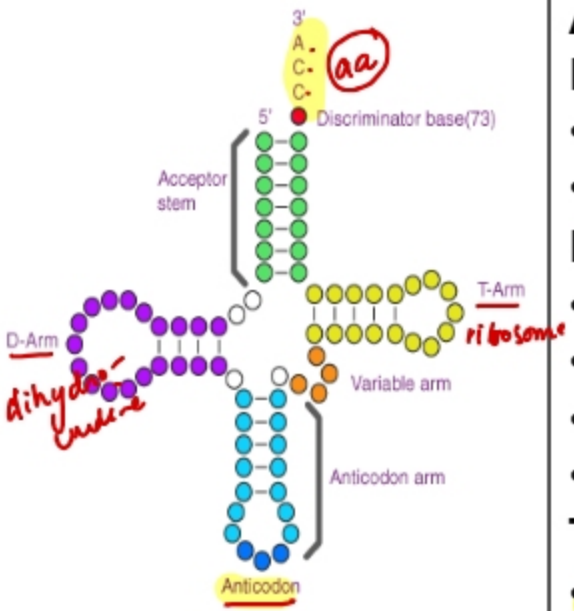
Isotype switching

IgM / IgD

↓

IgG / IgA / IgE

Translation



Amino-acyl tRNA synthetase-Proofreads and charges AA

Initiation

•Initiation factors bind to: **Shine–Dalgarno sequence** (P) / **Kozak sequence** (E)

•AUG start codon codes for **f-Met** (P) / **Met** (E)

Elongation

•**Aminoacyl-tRNA enters the A-site** using **2 GTP**

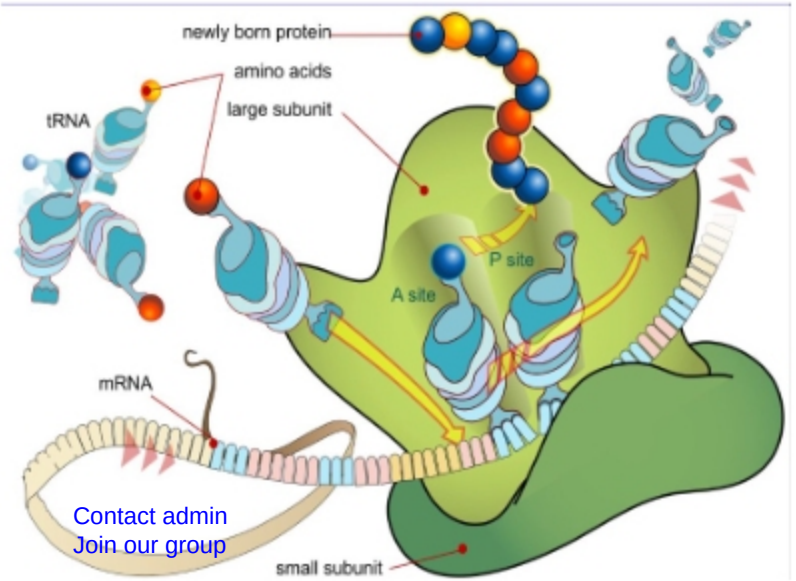
•**Peptidyl transferase (rRNA)** catalyzes the **peptide bond formation**

•**Ribosome translocates** along mRNA → polypeptide shifts from **A → P site**

•**Empty tRNA exits** from the **E site**

Termination

•**Stop codons** are recognized by **release factors (RFs)**



Genetic Code:

1. Unambigous
2. Degenerate / Redundant - Wobble
   EXCEPT: Methionine (AUG), Tryptophan (UGG)
3. Commaless, Non-overlapping
4. Universal (EXCEPT: Mitochondria)

STOP Codons : UAG, UGA, UAA

Supressor tRNA mutation:

MUTATIONS AND DNA REPAIR

Point mutations: UCA (serine)

- UCU (serine) (silent)
- CCA (proline) (missense)
- UAA (stop) (nonsense)

Transition: purine → purine / pyr → pyr

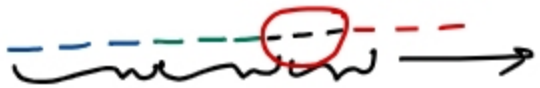
Tranversion: purine ⇌ pyr

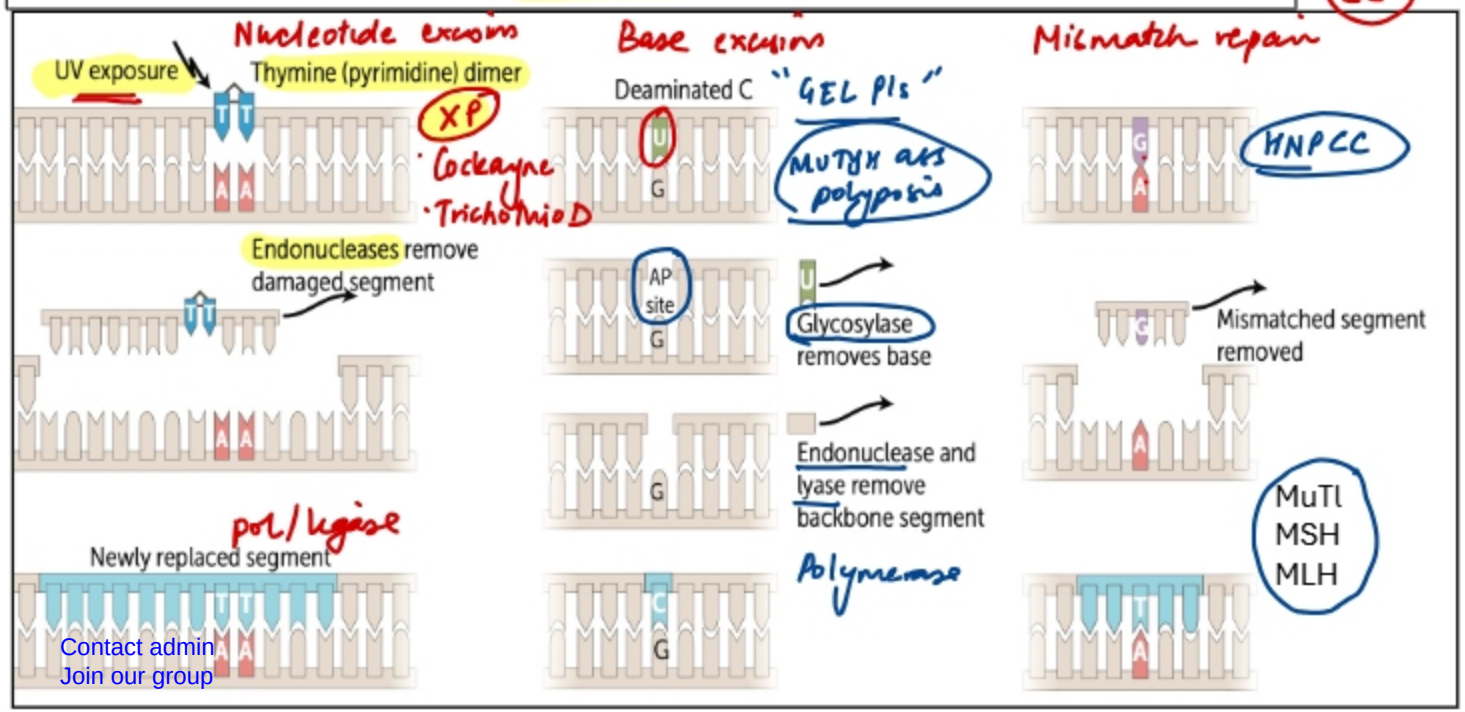
Frame shift: add^{n}/del^{n} – ⊗ in multiple of 3

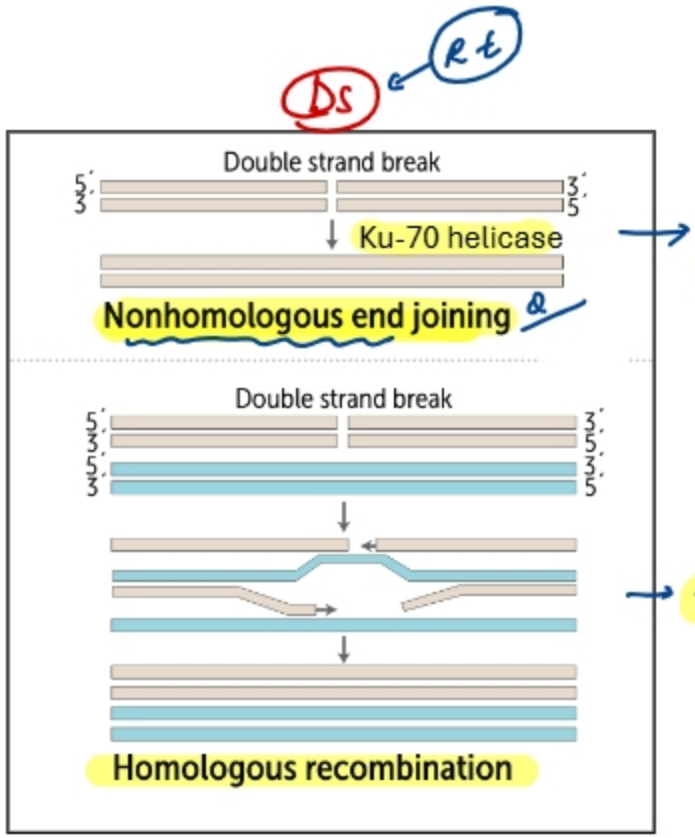
Mis-sense mutation: Sickle cell anemia - βC → Glu → Val

Frameshift mutations: DMD, Tay-Sachs

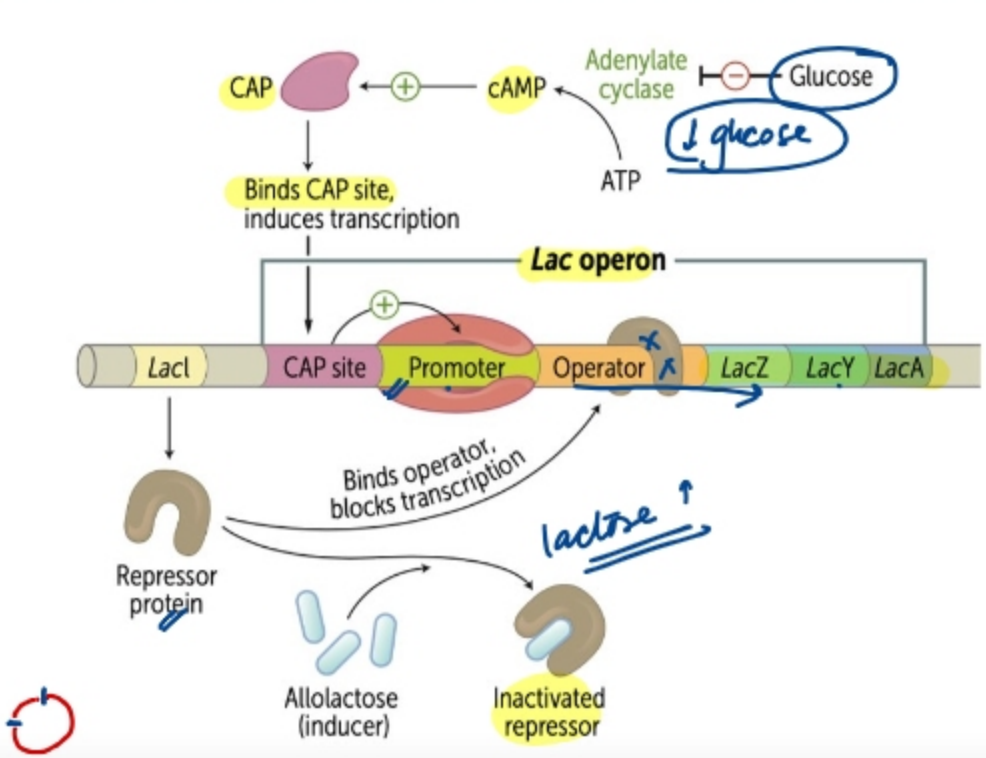
Splice site mutations: B-thalassemia, Marfan, Gaucher

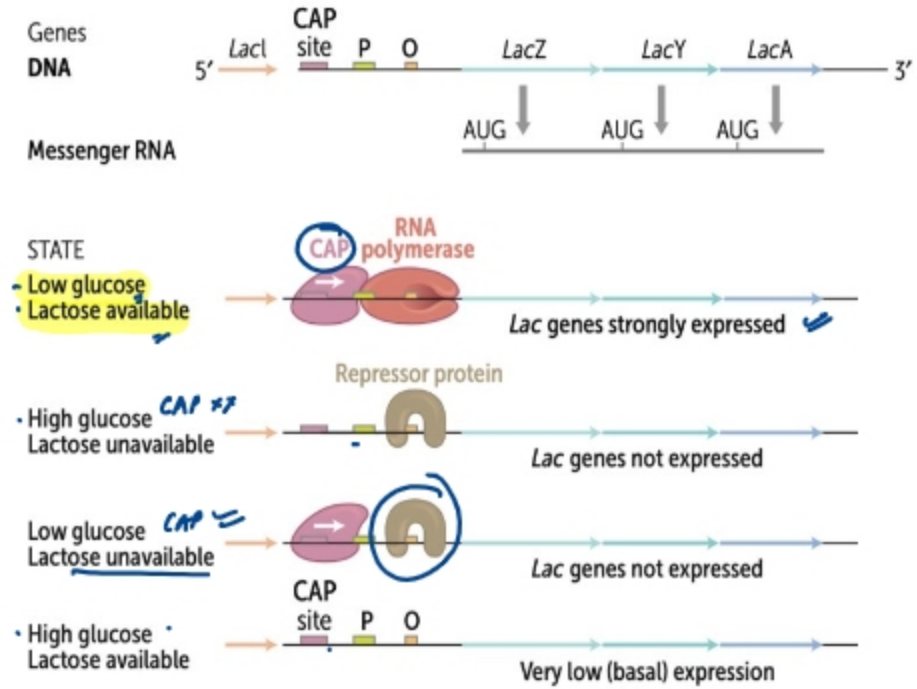


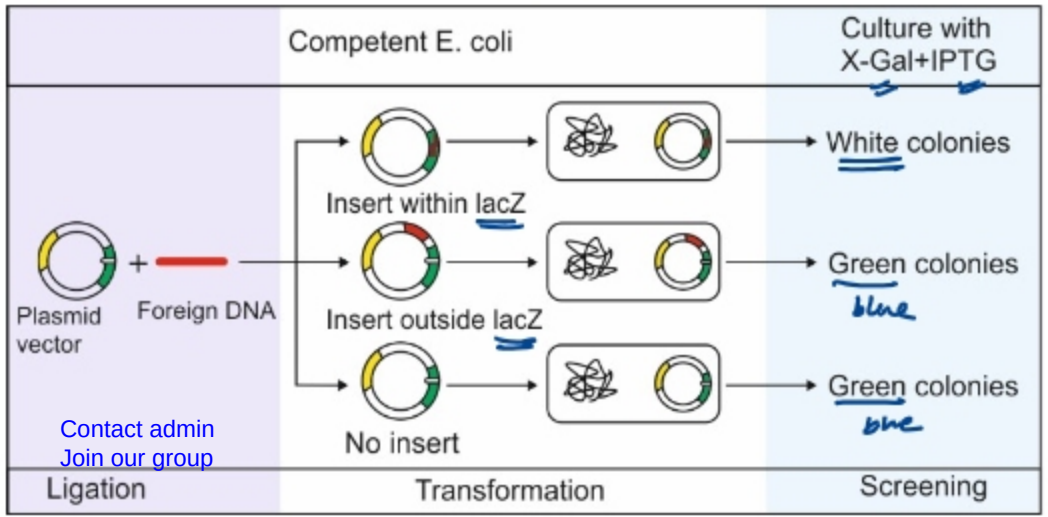




Lac Operon and Complementation



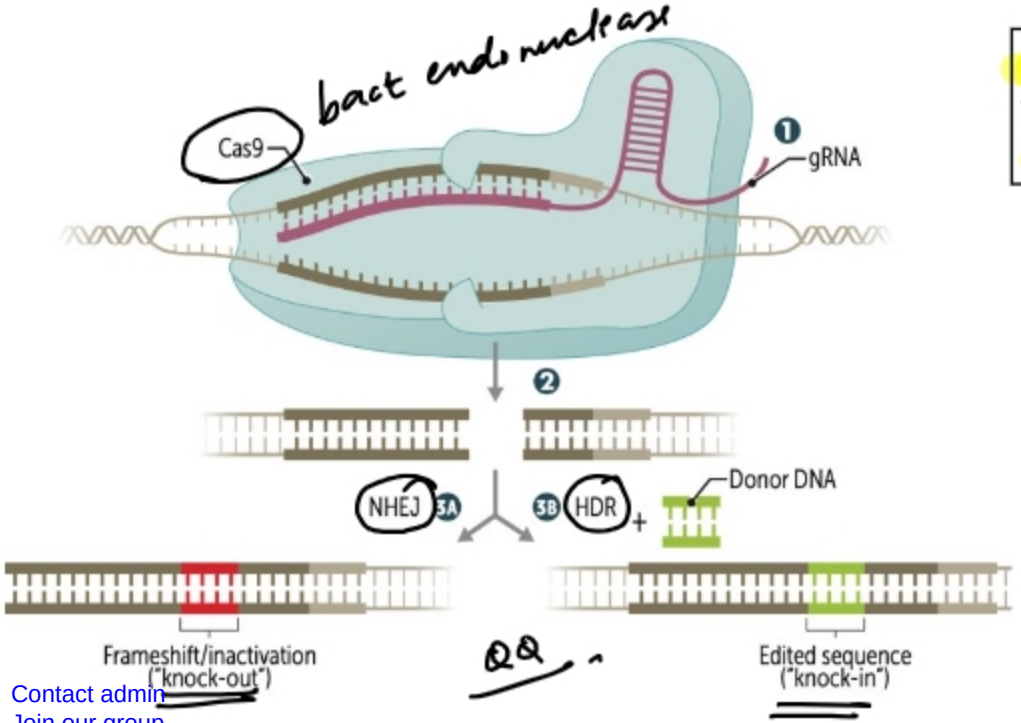




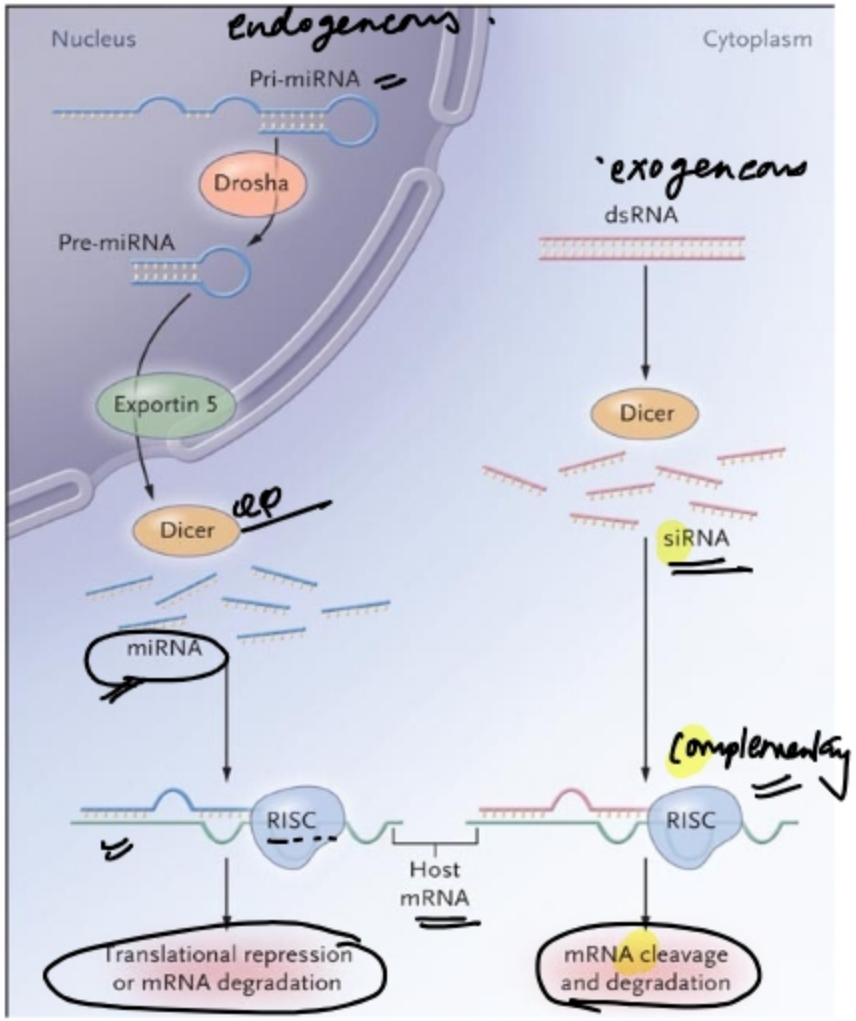
blue - white complementation.

GENETIC MODIFICATION

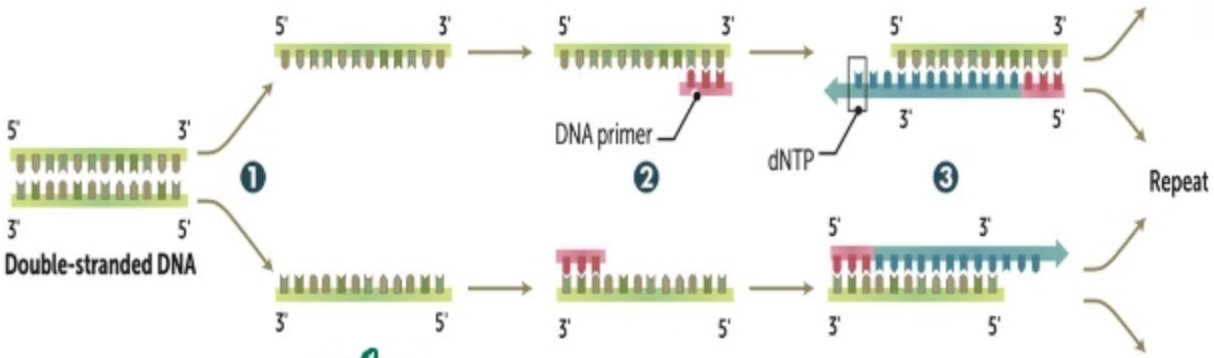
Gene Knock IN (insertion)
Gene Knock OUT (deletion)
Gene Knock DOWN = RNA Interference/ PTGS (post-transcriptional)
(si/mi RNA)
- 3'UTR
- 20-25 bp
- DICER-1: Pinealoblastoma / WT/ Pleuro-pulmonary blastoma/ Sertoli Leydig/ Multinodular Goitre



Charpentier used the CRISPR technique for gene editing: 2020 Nobel prize



PCR



1. Denaturation- 95°C
2. Annealing- 55°C
3. Elongation- 72° C

No of PCR cycles:

$= 2^n$

→ 26 hr
↓ 1/2
25 hr

DNA template, DNA primers, Heat stable DNA (Taq) polymerase, Mg, Deoxynucleotide triphosphates (dNTPs)

dideoxy NTPs → stop

Types of PCR:

- Real-time PCR-Quantitative-Ct value – cycle threshold. – viral load / Ph chromosome
- RT PCR (Tth polymerase) RNA → DNA – RNA viruses
- Digital droplet PCR ↓ level mutn / ct DNA = Liquid bx (circulating tumor DNA)
- Multiplex PCR : multiple primers – meningitis
- Nested PCR 2 rounds ∵ 2 sets of primers – ↑ sp

COVID 19 : RT-PCR → Real-time PCR

DNA Sequencing:-

-Sanger's

-3.2 billion bp/ 19.5k genes / Only 1.5 exons

-Exome sequencing – time / economical

-NGS: Whole Genome Sequencing (WGS)

↳ much faster / more sn

Chromosome walking

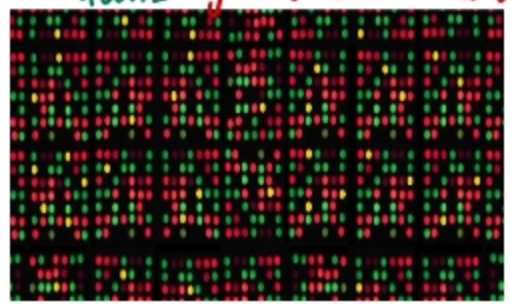
Gene-mapping to identify unknown DNA sequences that lie adjacent to a known sequence "walks" step-by-step along the chromosome.

MOLECULAR BIOLOGY TECHNIQUES

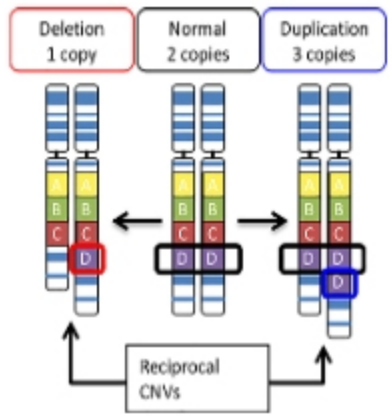
diseases / cancers / genetic linkage

DNA Microarray: Identify single nucleotide polymorphisms (SNP)
Small glass slide with short DNA probes attached
Sample DNA binds to complementary microarray DNA probes

GWAS - genome wide ass" study



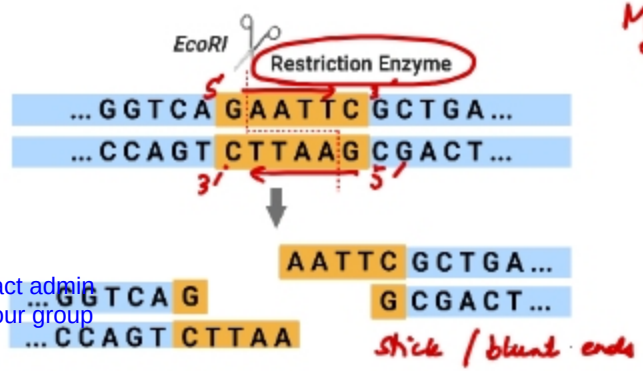
GWAS:
Microarrays on large population
Assess correlation between SNPs and complex disease



Multiplex Ligation-dependent Probe Amplification (MLPA)
CNV: Changes in the number of copies of a particular gene or genomic region: Deletion/ duplication/ amplification

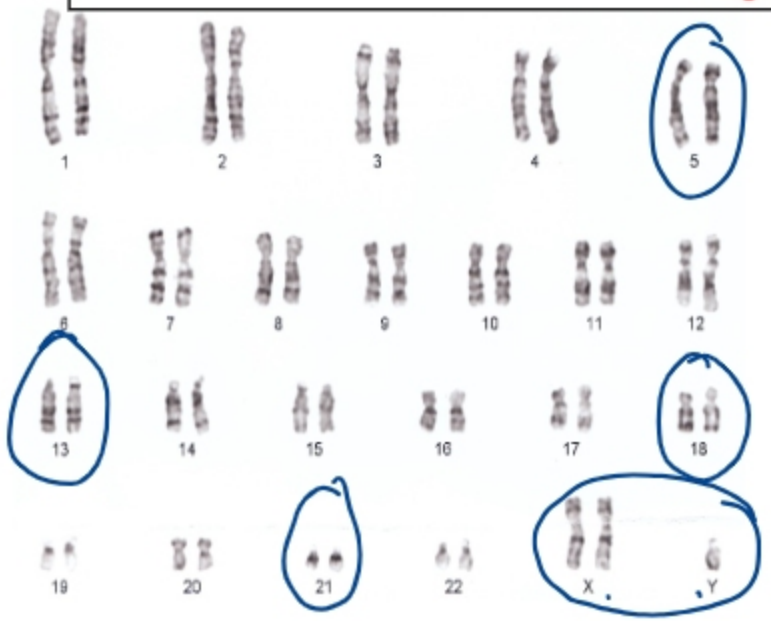
RFLP: Mutation affecting palindromic sites
Paternal disputes, Crime, Genome mapping

MADAM



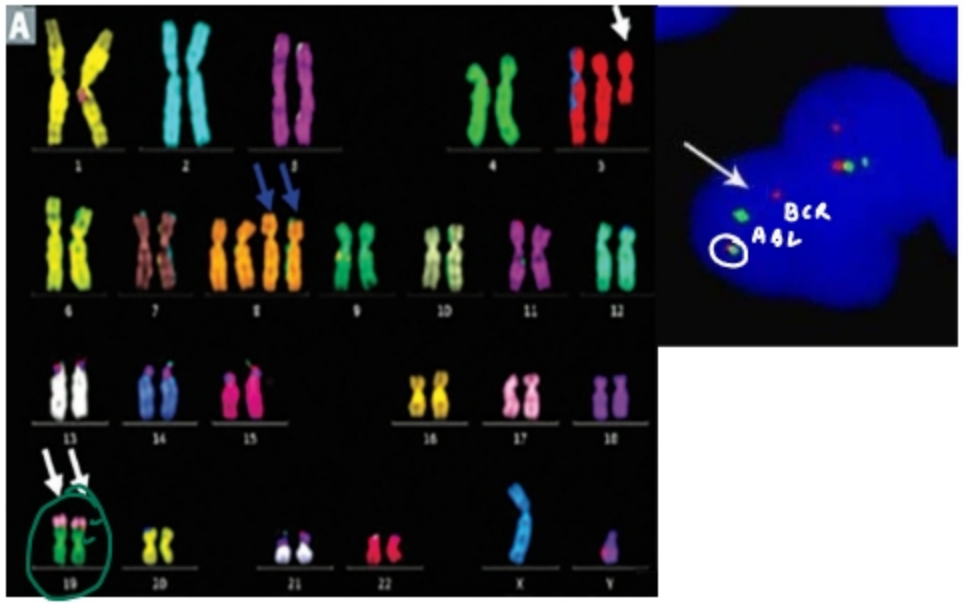
MOLECULAR BIOLOGY TECHNIQUES

Cytogenetics



Karyotype

- Best for ANEUPLOIDY
- Metaphase arrest — colchicine ↓MT (+)
- Fixative: Carnoy
- Dicentric staining: C-staining
- G/R/T/C/Q stain — Quinacrine (FISH)
  - Giemsa
  - Reverse
  - telomere
  - centromere



FISH

- Microdeletion
- Amplification
- Translocation

NOT point mutation

- RFLP
- sequencing

BLOTTING

"SNOW DROP"

- SOUTHERN- DNA
- NORTHEN- RNA
- WESTERN- protein
- SOUTHWESTERN/ DNA footprinting — DNA binding protein

A 12-year-old patient needs to get a molecular diagnosis of sickle cell anemia as three of his maternal cousins are affected. Arrange the steps in the correct sequence.

1.RT-PCR
2.Sample collection
3.FISH
4.RFLP
5.Cytogenetics
6.Conventional PCR
7.DNA extraction

Which of the following is used in diagnosing **aneuploidy**?

Multiple correct

1.FISH
2.Cytogenetics
3.Sanger sequencing
4.PCR