Intracellular Accumulations:

- Under some circumstances, cells may *accumulate abnormal amounts of various Substances that* may be ***harmless*** or cause varied **degrees of *cell injury***.

The *locations of these substances* either in the ***cytoplasm****, within* ***organelles****, or in the* ***nucleus****.*

- These substances may be *synthesized by the affected cells or may be produced elsewhere.*

Types of these accumalated substances:

1. ***Normal cellular constituent***: accumulated in excess, such as water, lipids, proteins, and carbohydrates.
2. ***Abnormal substance***, either exogenous, such as a mineral or products of infectious agents, or endogenous, such as a product of abnormal synthesis or metabolism.
3. *Pigment*s.

Accumulation of these substances is occurred by three general pathways,

1-Accumulation of normal substances, either due to increase their production or due to failure to remove these substances by metabolism, like **Fatty change** in the liver.
2-A normal or abnormal endogenous substance accumulates because of genetic or acquired defects in its metabolism, like **storage diseases** or **alph-1 antitrypsin deficiency**. (mutations cause defective folding and transport of this protein).
3-An abnormal exogenous substance is accumulated within the cells without defects that are seen in previous two points. Like accumulation of **carbon or silica.**

Fatty changes (steatosis):Abnormal accumulations of triglycerides within the **hepatocytes** (the main site because the liver play central role in fat metabolism) , can also involve heart, skeletal muscles, & kidney.

The etiology of steatosis alcoholism, starvation: increases FA mobilization from peripheral store., Protein malnutrition: decrease synthesis of apoprotein., Obesity, Diabetes mellitus , Hypoxia, Liver toxin like CCL4, Drugs like estrogen, steroid, tetracyclinetoxins.
When the fatty change is mild, it may have no effect on cellular function. More severe fatty change may impair cellular function.
Recently proved, that **severe form** of fatty change may lead to **liver cirrhosis** & **hepatocellular carcinoma.**

Mechanisms of Fatty changes (steatosis):

Excess accumulation of triglycerides within the liver may result from defects in any one of the events in the sequence from fatty acid entry to lipoprotein exit from hepotocytes.

Morphology of Fatty Changes:

- In any site, Fat accumulation appears as ***clear vacuoles*** within parenchymal cells.
- Accumulation of glycogen & water also produce clear vacuoles.
- special techniques are needed to distinguish these three types of clear vacuoles:
- The identification of clear vacuoles of lipids require preparation of **frozen tissue sections** (avoid formalin), and then the sections are stained with **Sudan IV** or **Oil red-O.**
- The identification of **glycogen** clear vacuoles requires staining with **periodic acid-Schiff** (PAS) reaction .

3. Clear vacuoles of **water are negative** staining for Sudan IV or Oil red- O, & (PAS).

- ***Gross*** In the liver, mild fatty change …….not affect the gross appearance.
- In progressive fatty changes …… the organ enlarges & more yellowish,
- In extreme instances……. Bright yellow, soft, greasy liver.

Mic.: Early fatty change, there are minute vacuoles in the cytoplasm & around the nucleus of fat cells. As the process progresses, the vacuoles coalesce, creating cleared spaces that displace the nucleus to the periphery of the fat cells.

Protein Accumulation:

- Protein accumulation is less common than lipid accumulation.
- Intracellular accumulations of proteins usually appear as rounded, eosinophilic droplets, vacuoles, or aggregates in the cytoplasm.
- **Examples of protein accumulation:**
- In the kidney, there is accumulation of albumin in the cytoplasm of tubular cells of proximal tubules, which occur in diseases associated with increased protein filtration through the glomeruli (like **nephrotic syndrome**) & increase reabsorption of albumin by the tubular cells, accumulated protein appear as pink, hyaline cytoplasmic droplets, this is reversible process.
- There is marked accumulation of newly synthesized **immunoglobulins** that may occur in the RER of some **plasma cells**, resulting in rounded, eosinophilic **Russell bodies**.
- In **alcoholic liver diseases**, there is accumulation of intracellular proteins (keratin intermediate filaments) **(Mallory body)**, which appear as an **eosinophilic inclusion** in the liver cells.
- **Neurofibrillary tangle** which is aggregation of proteins that are present in the brain of Alzheimer disease.

Glycogen Accumulation: Excess accumulation of glycogen can be seen in the followings:

In poorly controlled **Diabetes Mellitus**, glycogen will accumulate in renal tubular epithelium, cardiac muscles, & beta cells of Islet cells of pancreas.

- In **glycogen storage diseases**, there is enzymatic defect that result in accumulation of glycogen in various cells of body.

Pathologic Calcification

Pathologic calcification is the abnormal tissue deposition of calcium salts, together with smaller amounts of iron, magnesium, and other mineral salts

There are two forms of pathologic calcification.

1. **DYSTROPHIC CALCIFICATION**: The cause is NOT hypercalcemia; typically, the serum calcium concentration is normal

- occurs locally in **dying tissue (areas of necrosis**, whether they are of coagulative, caseous, or liquefactive type, and in foci of enzymatic necrosis of fat,
- or previously **Damaged tissue**, such as:
- areas of old trauma, **in the atheromas of advanced atherosclerosis**, **in aging or damaged heart valves , tuberculous lymph node**.

Gross: fine, white granules, often felt as gritty deposits

Mic: By H&E the calcium salts have a **basophilic, amorphous granular, sometimes clumped appearance.** (Intracellular, extracellular, or in both locations)

2. **Metastatic calcification**:

- Deposition of calcium salts in otherwise **normal tissues** .
- it almost always **results from hypercalcemia** secondary to some disturbance in calcium metabolism.

There are four principal causes of

1. hypercalcemia: Excess calcium intake, such as in the milk-alkali syndrome (nephrocalcinosis and renal stones caused by milk and antacid self-therapy)

2. hyperparathyroidism : due to primary hyperthyroidism or produced by other tumors

3-destruction of bone tissue, occurring with primary tumors of bone marrow (e.g., multiple myeloma)

4- vitamin D–related disorders, including vitamin D intoxication, sarcoidosis(MQ activate a vit D precursor).

4. renal failure, which causes retention of phosphate, leading to secondary hyperparathyroidism

- Metastatic calcification may occur widely throughout the body but principally affects the interstitial tissues of the gastric mucosa, kidneys, lungs, systemic arteries, and pulmonary veins.

Pigments: They are colored substances, either **Exogenous** or **endogenous .**

Exogenous Pigments (coming from outside)**.**

- **Anthracosis (Carbon accumulation):** accumulation of carbon particles within the alveolar macrophages & transported to the lymph nodes of the tracheobronchial tree (appear as black discoloration).
- **Tattooing:** is a form of localized, exogenous pigmentation of the skin. The pigments inoculated are phagocytosed by dermal macrophages.

Endogenous Pigments (synthesized within the body itself)**.**

Lipofuscin:

- composed of polymers of lipids and phospholipids complexed with protein,
- Lipofuscin is **NOT** injurious to the cell or its functions.
- Its important sign of **free radical injury** and **lipid peroxidation.**
- **MIC.** it appears as a yellow-brown, finely granular intracytoplasmic, often perinuclear pigment.
- Typically seen in the **liver** and **heart** of aging patients or patients with **severe malnutrition, atrophy**, **aging process** and **cancer cachexia**.
- Grossly lipofuscin in the heart is called **brown atrophy**.

Melanin:

It is an endogenous, non-hemoglobin-derived, brown-black pigment formed when the enzyme tyrosinase catalyzes the oxidation of tyrosine to dihydroxyphenylalanine in melanocytes in various organs. Excess of melanin can be seen in malignant melanoma.

Hemosiderin:

- is a hemoglobin-derived, golden yellow-to brown, granular or crystalline pigment(iron containing pigment)
- Hemosidrin is representing aggregation of ferritin.
- Iron can be identified in the tissue by **Prussian blue reaction**.
- Excesses of iron cause hemosiderin to accumulate within cells, either as a localized process or as a systemic iron overload.

Local excesses of iron: The best example of localized hemosiderosis is the common bruise, (color changes in the bruises are due to deposition of hemosidrin in the macrophages)

systemic overload of iron:

hemosiderin is deposited in many organs and tissues, a condition called **hemosiderosis:**

(1) increased absorption of dietary iron,(2) impaired use of iron,

(3 hemolytic anemias, (4) transfusions because the transfused red cells constitute an exogenous load of iron.

- Hemosidrin is found at first in the mononuclear phagocytes of the liver, bone marrow, spleen, & lymph nodes.
- With progressive accumulation of hemosidrin, parenchymal cells throughout the body (mainly liver, pancreas, heart & endocrine organs) become bronzed in color.
- At the stage of hemosdiderosis, iron pigment does not damage the parenchymal cells or impair the function of the organs.
- While at the stage of **hemochromatosis** (more extensive accumulation of iron), associated with tissue injury , scarring and organ dysfunction . e.g. liver fibrosis, heart failure, & diabetes mellitus.

- Bronze color of skin with diabetes mellitus is called **Bronze Diabetes Mellitus**.

Amyloidosis

• It is a disorder characterized by the **extracellular deposits** of abnormal proteinaceous material (**misfolded proteins**) that aggregate to form insoluble fibrils (which are soluble in their normal folded configuration).

• **Pathogenesis:** results from abnormal folding of proteins, which become insoluble, aggregate, and deposit as fibrils in extracellular tissues

• ***physical nature:*** *By EM :*amyloid is composed mainly of continuous, nonbranching fibrils, Insoluble , linear, rigid measuring 7.5 to 10 nanometers in width.

- **Chemical nature of Amyloidosis:**
- Most common forms of amyloid proteins are:
- **AL** (amyloid light chain), derived from plasma cell & contains **immunoglobulin light chain**.
- **AA** amyloid fibril, associated with **chronic inflammatory diseases**, not related to immunoglobulins, & synthesized in the **liver**.
- **Aβ** is found in the **cerebral** lesion of **Alzheimer disease**.
- **Transthyretin (TTR)** is a normal serum protein that binds and transports thyroxine. Mutant forms of TTR are deposited in a group of genetically determined disorders referred to as familial amyloid polyneuropathies. Normal TTR is also deposited in the heart of aged individuals (senile systemic amyloidosis).
- • **β2 microglobulin**, found in patients on long-term hemodialysis.

Classification of Amyloidosis:

- **Localized Amyloidosis**. which include
- **Senile Amyloidosis**. Seen in the Alzheimer disease.
- **Endocrine amyloid**. Localized deposits of amyloid in certain endocrine tumors (medullary carcinoma of the thyroid gland .
- **Isolated atrial Amyloidosis**.
- **Systemic (Generalized) Amyloidosis** , which include
- **Primary Amyloidosis (immunocyte associated Amyloidosis)**.

This is the most common form of Amyloidosis.
The best example in this type is Amyloidosis associated with **Multiple myeloma** (malignant neoplasm of plasma cells).
Major amyloid protein in this type is **AL** protein.

- **Secondary Amyloidosis(** Reactive Systemic Amyloidosis).
- Major amyloid protein in this type is AA.

1-It is associated with many **Chronic Inflammatory Conditions** like,TB, bronchiectasis, & chronic osteomyelitis are the common causes.
2-associated with **autoimmune diseases** e.g. rheumatoid arthritis.
3- associated with **tumor**s like :Renal cell carcinoma, Hodgkin lymphoma .

Morphology of Amyloidosis:

- **There are no distinctive patterns of organ distribution of amyloid deposits,** e.g. in **secondary Amyloidosis** liver, spleen, lymph nodes, adrenal & thyroid are commonly affected, while **in primary Amyloidosis** is commonly affect the heart, GIT, respiratory tract, peripheral nerves & skin.
- **Gross** : Accumulation of larger amounts of amyloid will result in enlargement of organs, & waxy firm consistency.
- **MIC**. the deposition of amyloid is always begins between the cells, often closely adjacent to basement membranes & with more deposits will result in destruction of the cells.
- The histologic diagnosis of amyloid is based almost entirely on its staining characteristics, which include
- **Congo red:** under light microscope show pink or red color of amyloid deposits, while under polarized microscope, it appears as apple green.
- Immunohistochemistry staining: can demonstrate the AA, AL, & transthyretin of amyloid fibrils.
- Electron microscope: reveals amorphous non oriented thin fibrils.
- **Kidney:**

Amyloidosis of the kidney is the most common & most serious involvement in the disease.

Gross. Either unchanged in the size or abnormally become enlarged, pale, gray, firm; & in advanced cases, the kidney may be reduced in their size.

Mic. Amyloid principally deposited in the glomeruli & peritubular tissue.

- **Liver:**

Gross. Massive enlargement of liver (up to 9000 grams), pale, gray, & waxy in consistency.

Mic. Amyloids first appear in the space of Disse, and then extend to involve the hepatocytes & sinusoids