

# ***Collagen Tissue***

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**Collagen and elastin - fibrous proteins.**

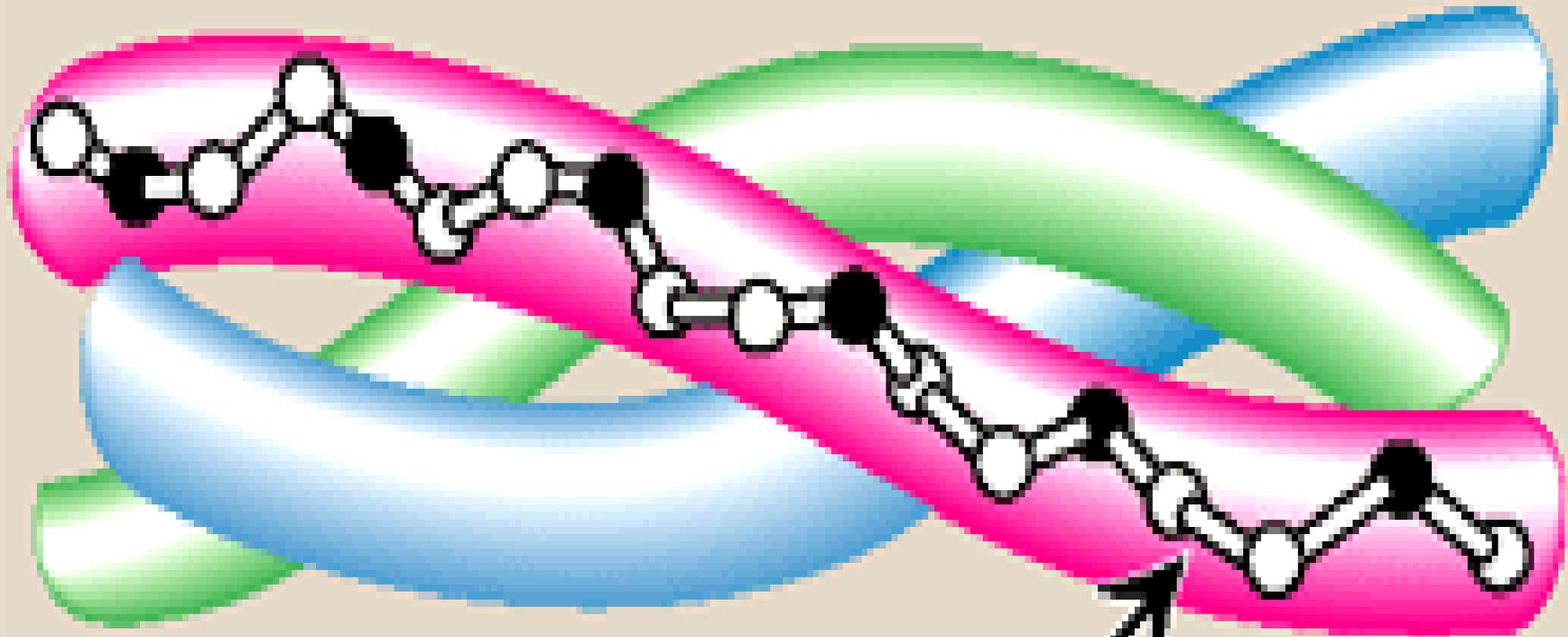
**Approximately 1,000 amino acids long.**

**Serve structural functions in the body.**

**Components of skin, connective tissue, blood vessel walls, and sclera and cornea of the eye.**

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- **Collagen is the most abundant protein in the human body.**
- **A long, rigid structure**
- **Three polypeptides (as “ $\alpha$  chains”) are wound around one another in a rope-like triple helix .**
  
- **In other tissues, collagen may be bundled in tight, parallel fibers that provide great strength, as in tendons.**
  
- **Collagen of bone occurs as fibers arranged at an angle to each other so as to resist mechanical shear from any direction.**
  
- **Collagen may be as a gel that gives support to the structure, as in the extracellular matrix. E.g. vitreous humor of the eye.**
  
- **In the cornea of the eye, collagen is arrange as to transmit light with a minimum of scattering.**



**Collagen  $\alpha$  chain**

## **Types of collagen**

**More than twenty collagen types.**

**The three polypeptide  $\alpha$  chains are held together by hydrogen bonds between the chains.**

**Variations in the A.A. sequence of the  $\alpha$  chains result in structural components that are about the same size but with slightly different properties.**

**These  $\alpha$  chains are combined to form the various types of collagen found in the tissues.**

**For example, the most common collagen, type I, contains two chains called  $\alpha_1$  and one chain called  $\alpha_2$  ( $\alpha_1_2\alpha_2$ ), whereas type II collagen contains three  $\alpha_1$  chains ( $\alpha_1_3$ ).**

- 1. Fibril forming collagens.**
- 2. Network forming collagens.**
- 3. Fibril associate collagens.**

<b>TYPE</b>	<b>TISSUE DISTRIBUTION</b>
	<b>Fibril-forming</b>
<b>I</b>	<b>Skin, bone, tendon, blood vessels, cornea</b>
<b>II</b>	<b>Cartilage, intervertebral disk, vitreous body</b>
<b>III</b>	<b>Blood vessels, fetal skin</b>
	<b>Network-forming</b>
<b>IV</b>	<b>Basement membrane</b>
<b>VII</b>	<b>Beneath stratified squamous epithelia</b>
	<b>Fibril-associated</b>
<b>IX</b>	<b>Cartilage</b>
<b>XII</b>	<b>Tendon, ligaments, some other tissues</b>

The collagens can be organized into three groups, based on their location and functions. Basement membranes are thin, sheet-like structures that provide mechanical support for adjacent cells, and function as a semipermeable filtration barrier to macromolecules in organs such as the kidney and the lung.

# **1. Fibril-forming collagens**

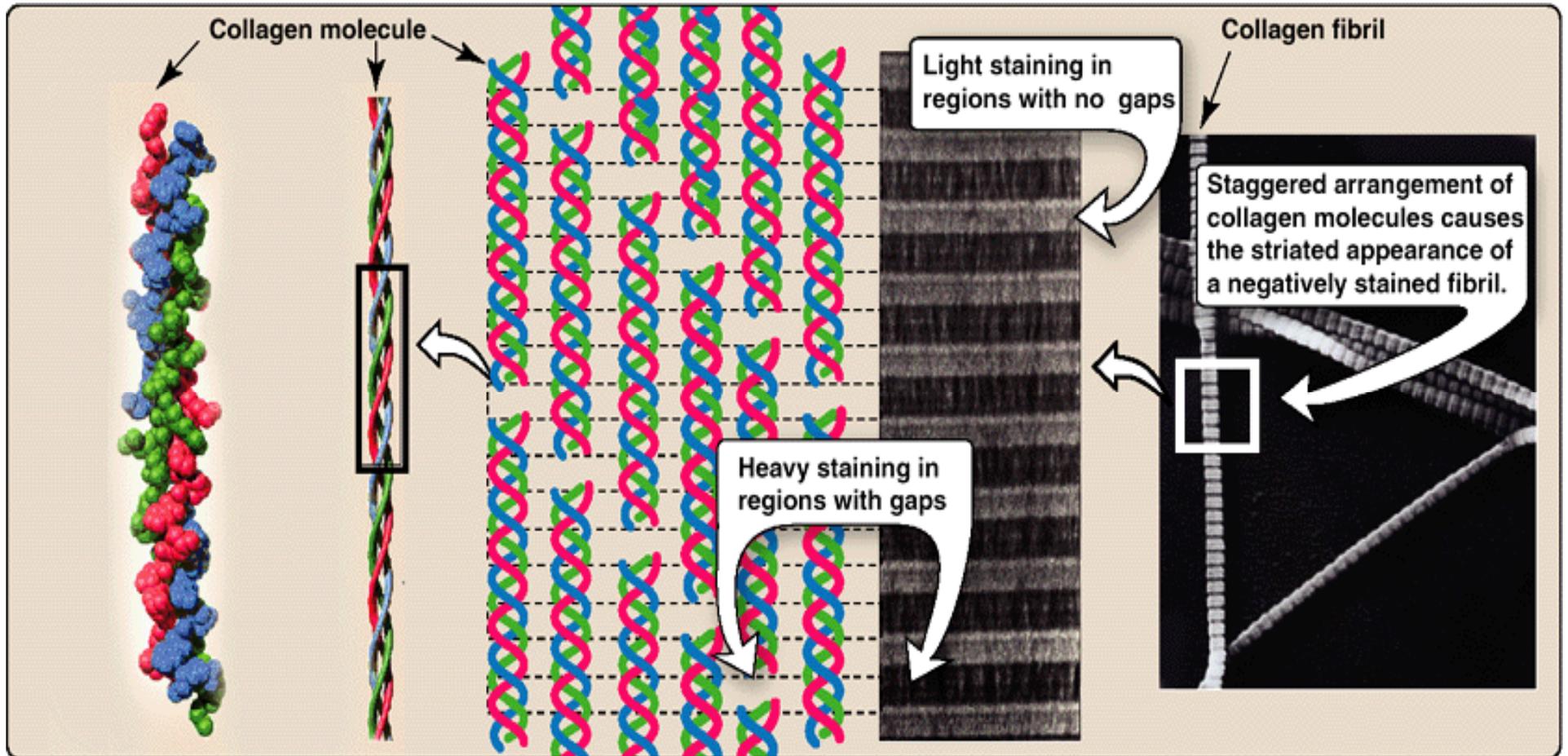
**Types I, II, and III are the fibrillar collagens.**

**In the electron microscope, these linear polymers of fibrils have characteristic banding patterns, reflecting the regular staggered packing of the individual collagen molecules in the fibril .**

**Type I collagen fibers are found in supporting elements of high tensile strength e.g.tendon and cornea.**

**Type II collagen molecules are restricted to cartilaginous structures.**

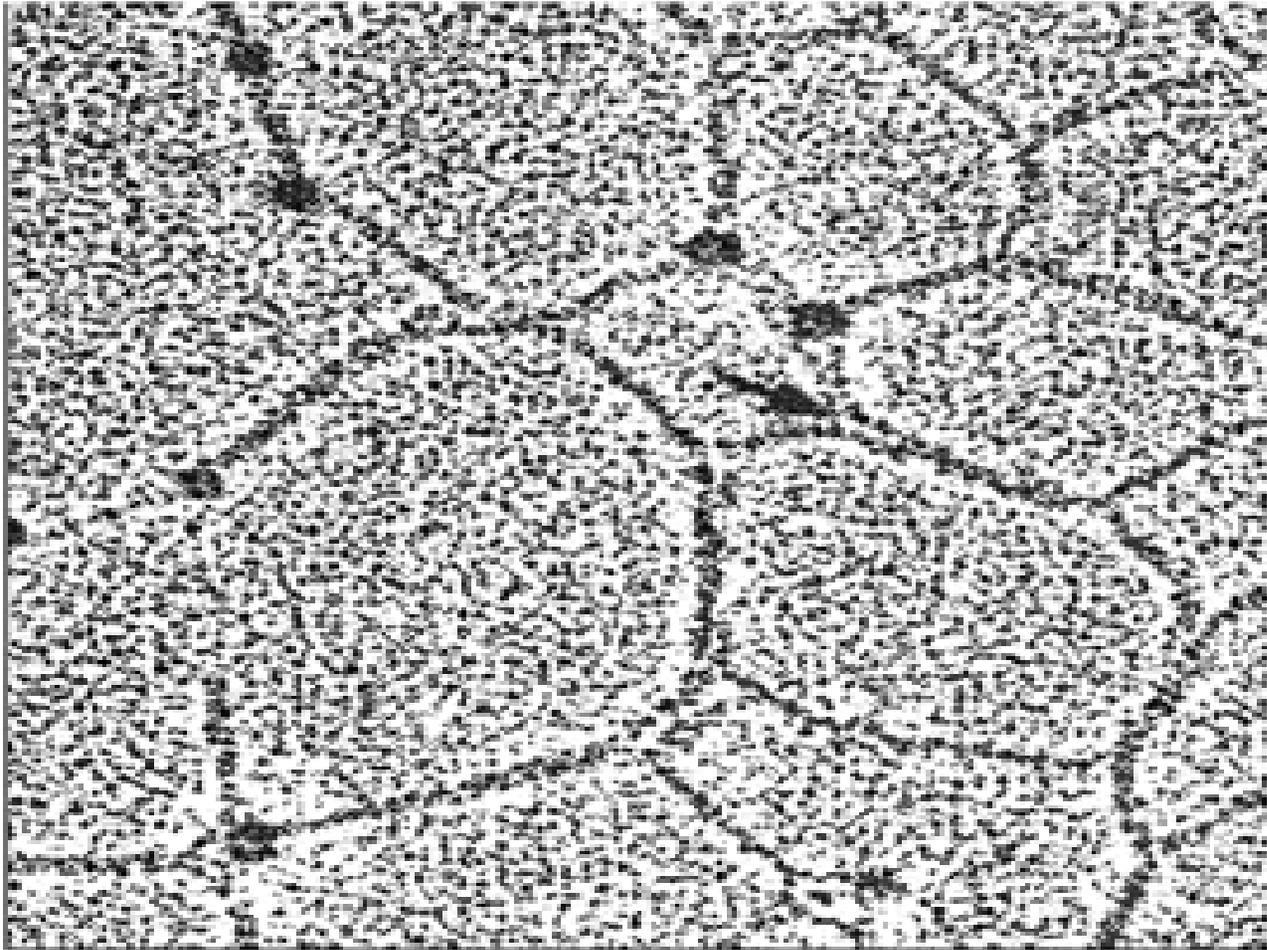
**Type III collagen are found in more distensible tissues e.g. blood vessels.**



## **2. Network-forming collagens**

Form a three-dimensional mesh.

For example, type IV that constitutes a major part of basement membranes



### **3.Fibril-associated collagens**

- **It bind to the surface of collagen fibrils.**
- **linking these fibrils to one another and to other components in the extracellular matrix.**

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# **Structure of collagen**

- 1. Amino acid sequence***
- 2. Triple-helical structure***
- 3. Hydroxyproline and Hydroxylysine***
- 4. Glycosylation***

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# ***1. Amino acid sequence***

Collagen is rich in Proline and Glycine.

Glycine, the smallest amino acid, is found in every third position of the polypeptide chain.

It fits into the restricted spaces where the three chains of the helix come together.

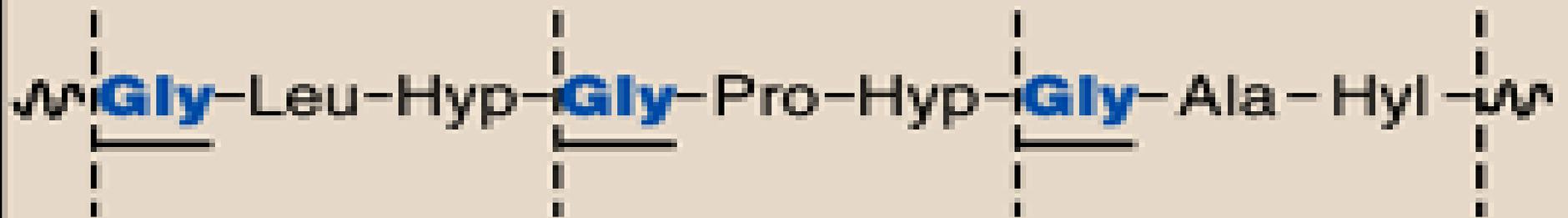
**-Gly-X-Y-** where X is frequently proline

Y is often hydroxyproline or hydroxylysine.

Thus, most of the  $\alpha$  chain can be regarded as a polytripeptide whose sequence can be represented as  $(\text{-Gly-Pro-Hyp-})_{333}$ .

Proline facilitates the formation of the helical conformation of each  $\alpha$  chain because its ring structure causes “kinks” in the peptide chain.

***[Note: The presence of proline dictates that the helical conformation of the  $\alpha$  chain cannot be an  $\alpha$  helix.]***



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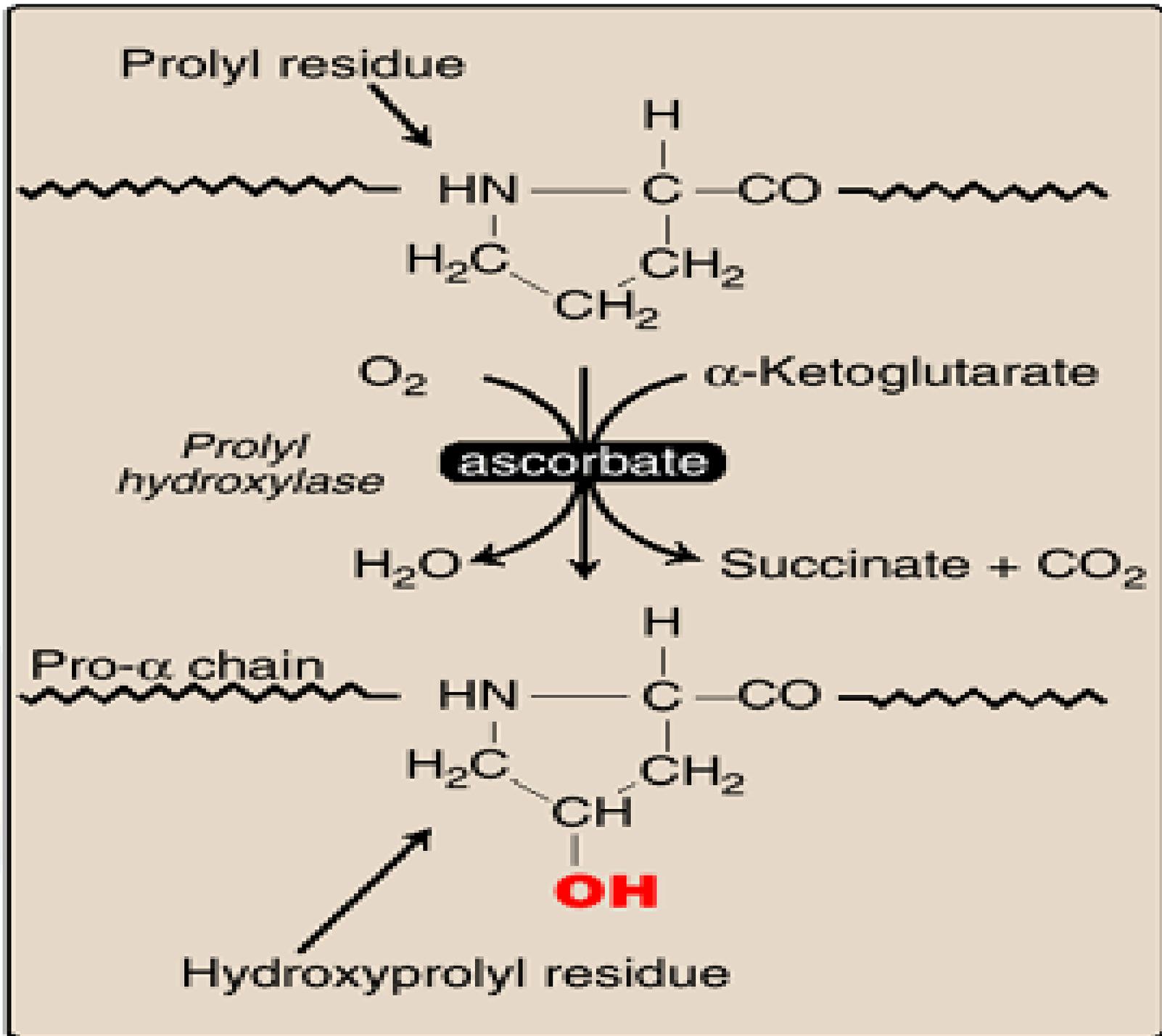
## ***2. Triple-helical structure***

- **$\alpha$  chains are folded into compact structures.**
- **Collagen has an elongated, triple-helical structure.**
- **Places many of its amino acid side chains on the surface of the triple-helical molecule.**
- **This allows bond formation between the exposed R-groups of neighboring collagen monomers, resulting in their aggregation into long fibers.**

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### ***3. Hydroxyproline and Hydroxylysine***

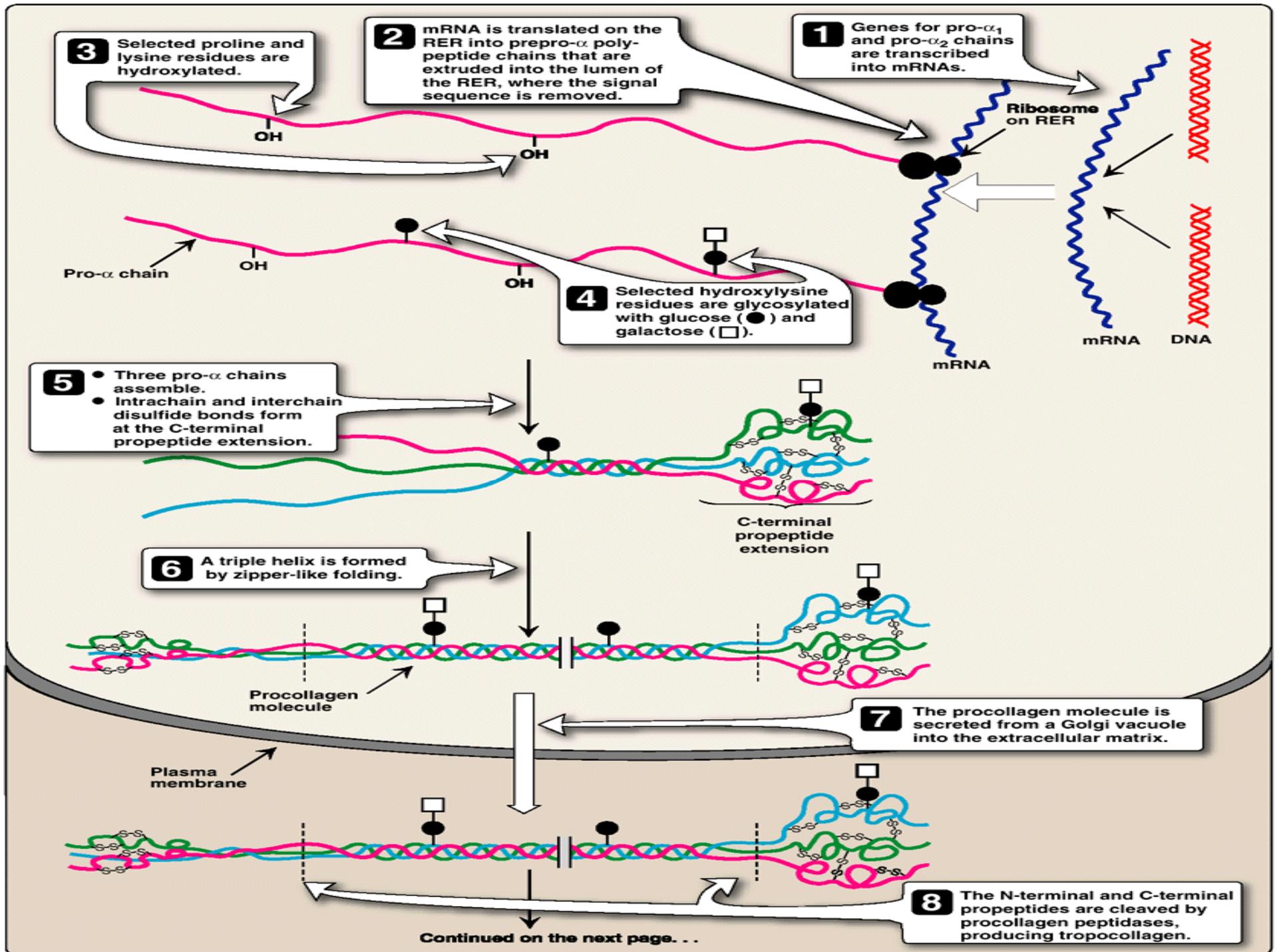
- Hydroxylation of some of the proline and lysine residues after their incorporation into polypeptide chains.
- The hydroxylation is an example of post-translational modification.
- *Hydroxyproline is important in stabilizing the triple-helical structure of collagen because it maximizes interchain hydrogen bond formation.*
- Reactions require the reducing agent vitamin C , without which the hydroxylating enzymes, prolyl hydroxylase and lysyl hydroxylase, are unable to function.
- *In the case of ascorbic acid deficiency , collagen fibers cannot be cross-linked, greatly decreasing the tensile strength of the assembled fiber. The resulting deficiency disease is known as SCURVY.*
- Patients with ascorbic acid deficiency also often show bruises on the limbs as a result of subcutaneous extravasation of blood (capillary fragility).



## 4. Glycosylation

- The hydroxyl group of the hydroxylysine residues of collagen may be enzymatically glycosylated.
- Most commonly, glucose and galactose are attached to the polypeptide chain prior to triple-helix formation.

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# ***Biosynthesis of collagen***

The polypeptide precursors of the collagen molecule are formed in fibroblasts / osteoblasts of bone / chondroblasts and secreted into the extracellular matrix.

After enzymic modification, the mature collagen monomers aggregate and become cross-linked to form collagen fibrils.

1. Formation of pro- $\alpha$  chains
2. Hydroxylation
3. Glycosylation
4. Assembly and secretion
5. Extracellular cleavage of procollagen molecules
6. Formation of collagen fibrils
7. Cross-link formation

# 1. Formation of Pro- $\alpha$ chains

- Pre-pro- $\alpha$  chains contain a special amino acid sequence at their N-terminal ends.
- This sequence acts as a signal for that chain's destination to leave the cell.
- The signal sequence facilitates the binding of ribosomes to the rough endoplasmic reticulum (RER), and directs the passage of the prepro- $\alpha$  chain into the lumen of the RER.
- That signal sequence is rapidly cleaved in the endoplasmic reticulum to yield a precursor of collagen called a pro- $\alpha$  chain.

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## 2. Hydroxylation

- Proline and lysine residues found in the Y-position of the -Gly-X-Y- sequence can be hydroxylated to form hydroxyproline and hydroxylysine residues.
- These hydroxylation reactions require molecular oxygen and the reducing agent vitamin C , without which the hydroxylating enzymes, prolyl hydroxylase and lysyl hydroxylase, are unable to function.
- In the case of ascorbic acid deficiency , collagen fibers cannot be cross-linked, greatly decreasing the tensile strength of the assembled fiber. The resulting deficiency disease is known as SCURVY.
- Patients with ascorbic acid deficiency also often show bruises on the limbs as a result of subcutaneous extravasation of blood (capillary fragility).

The legs of a 46-year-old man with scurvy



### **3. Glycosylation**

**Some hydroxylysine residues are modified by glycosylation with glucose or glucosyl-galactose .**

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## 4. Assembly and secretion

- The formation of procollagen begins with formation of interchain disulfide bonds between the C-terminal extensions of the pro- $\alpha$  chains.
- This brings the three  $\alpha$  chains into an alignment favorable for helix formation.
- The procollagen molecules are translocated to the Golgi apparatus, where they are packaged in secretory vesicles.
- The vesicles fuse with the cell membrane, causing the release of procollagen molecules into the extracellular space.

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## **5. Extracellular cleavage of Procollagen molecules**

**After their release, the procollagen molecules are cleaved by N- and C-procollagen peptidases, which remove the terminal propeptides, releasing triple-helical tropocollagen molecules.**

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## 6. Formation of Collagen fibrils

- **Individual tropocollagen molecules spontaneously associate to form collagen fibrils.**
- **They form**
  - » **An ordered**
  - » **Overlapping**
  - » **parallel array**
  - » **with adjacent collagen molecules arranged in a staggered pattern**
  - » **each overlapping its neighbor by a length approximately three-quarters of a molecule.**

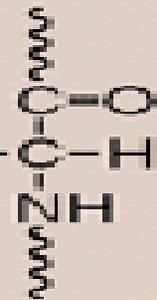
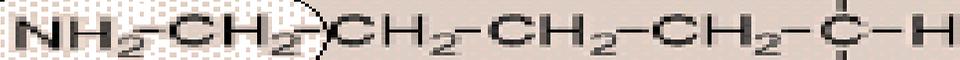
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## 7. Cross-link formation

- The fibrillar array of collagen molecules serves as a substrate for lysyl oxidase ( $\text{Cu}^{2+}$ -containing extracellular enzyme) oxidatively deaminates some of the lysyl and hydroxylysyl residues in collagen.
- That result (allysine and hydroxyallysine) can condense with lysyl or hydroxylysyl residues in neighboring collagen molecules to form covalent cross-links
- And, thus, mature collagen fibers.
- Cross-linking is essential for the tensile strength & the proper functioning of connective tissue.

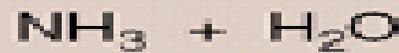
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Lysine residue

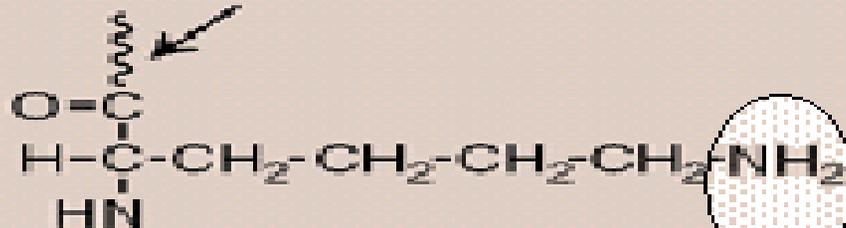


$\text{O}_2$

*Lysyl oxidase*

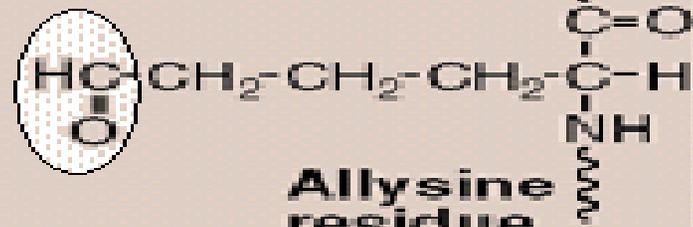


Collagen chain



Lysine residue

Collagen chain



Allysine residue



# **Degradation of collagen**

- Normal collagens are highly stable molecules.
- Half-lives as long as several years.
- However, connective tissue is dynamic and is constantly being remodeled, often in response to growth or injury of the tissue.
- Breakdown of collagen fibrils is dependent on the proteolytic action of collagenases.
- For type I collagen, the cleavage site is specific, generating three-quarter and one-quarter length fragments. These fragments are further degraded by other matrix proteinases.

## **Collagen diseases**

- Defects in any one steps of collagen fiber synthesis can result in a genetic disease.
- Provide tissues with the low tensile strength.

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# Ehlers-Danlos syndrome (EDS)

- **Generalized connective tissue disorders.**
- **EDS can result from a deficiency of**
  - **lysyl hydroxylase deficiency or**
  - **procollagen peptidase deficiency or**
  - **from mutations in the A.A. sequences of collagen types I, III, or V.**
- **Mutated chain either degraded or accumulated to high levels in intracellular compartments.**
- **Because collagen type III is an important component of the arteries, potentially lethal vascular problems occur.**
- **Type III is a minor component in the skin so patients with EDS also show fragile, stretchy skin and loose joints.**

# Stretchy skin of Ehlers-Danlos syndrome



# Osteogenesis imperfecta

- This disease, known as [Brittle Bone Syndrome](#), is also inherited disorders distinguished by bones that easily bend and fracture .
- Retarded wound healing
- Rotated and twisted spine leading to a “humped-back” appearance.
- Type I O.I. is called [Osteogenesis Imperfecta Tarda](#).
- Due to decreased production of  $\alpha 1$  and  $\alpha 2$  chains.
- Presents in early infancy with fractures due minor trauma.
- May be suspected if prenatal ultrasound detects bowing or fractures of long bones.

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- **Type II OI is called Osteogenesis Imperfecta Congenita.**
- **It is more severe**
- **Patients die of pulmonary hypoplasia in utero or during the neonatal period.**
- **Most patients with severe OI have mutations in the gene for either the pro- $\alpha$ 1 or pro- $\alpha$ 2 chains of type I collagen.**
- **The most common mutations cause the replacement of glycine residues (in -Gly-X-Y-) by amino acids with bulky side chains.**
- **The resultant structurally abnormal pro- $\alpha$  chains prevent the formation of the required triple-helical conformation.**

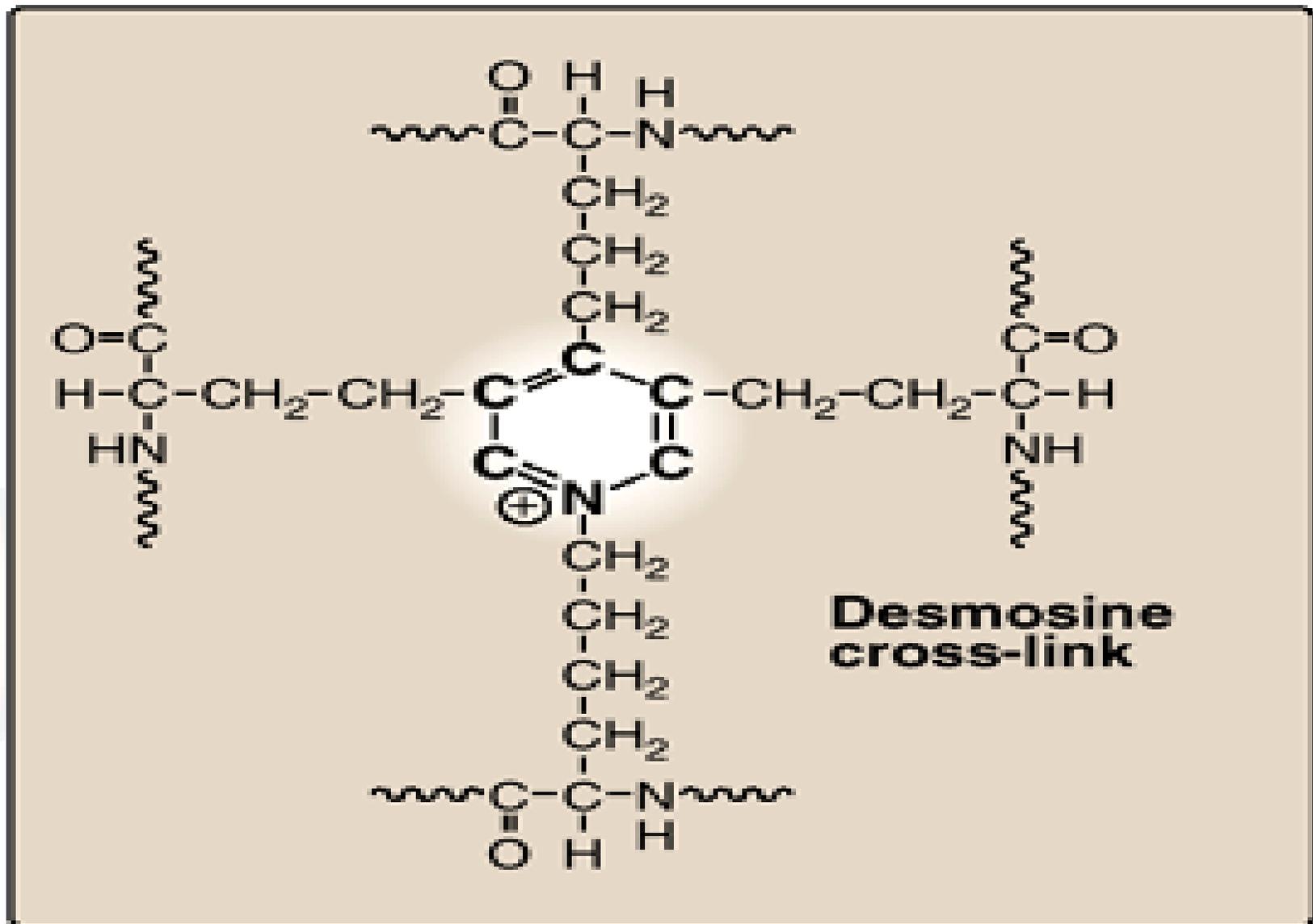
**Lethal form of osteogenesis imperfecta in which the fractures appear in utero, as revealed by this radiograph of a stillborn fetus.**



# Elastin

- In contrast to collagen, which forms fibers that are tough and have high tensile strength.
- A connective tissue protein with rubber-like properties.
- Elastic fibers composed of elastin and glycoprotein microfibrils are found in the lungs, the walls of large arteries, and elastic ligaments.
- They can be stretched to several times their normal length, but recoil to their original shape when the stretching force is relaxed.

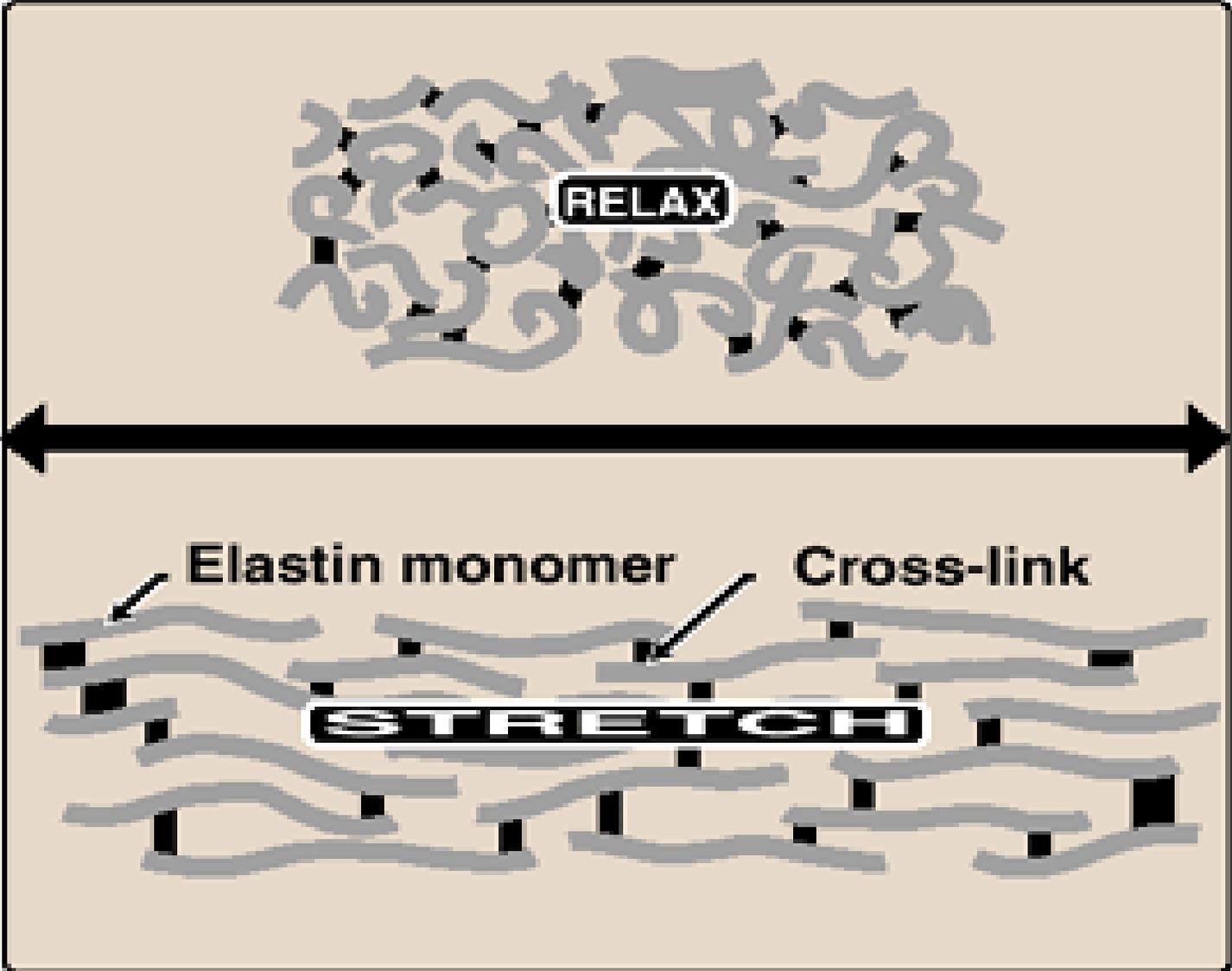
# Desmosine cross-link in elastin



# Structure of elastin

- Elastin is an insoluble protein polymer.
- A linear polypeptide composed of about 700 amino acids.
- Elastin is also rich in Proline and Lysine, but contains only a little hydroxyproline and no hydroxylysine.
- Some of the lysyl side chains of the tropoelastin polypeptides are oxidatively deaminated by , forming allysine residues.
- Three of the allysyl side chains plus one unaltered lysyl side chain from the same or neighboring polypeptides form a desmosine cross-link .
- This produces elastin—an extensively interconnected, rubbery network that can stretch and bend in any direction.
- Mutations in the fibrillin = Marfan syndrome — characterized by impaired structural integrity in the skeleton, the eye, and the cardiovascular system.

# Elastin fibers in relaxed and stretched conformations



## Role of alpha;1-antitrypsin in elastin degradation

- Blood and other body fluids contain a protein,  $\alpha$ 1-antitrypsin ( $\alpha$ 1-AT, AAT, currently also called  $\alpha$ 1-antiproteinase),
- that inhibits a number of proteolytic enzymes that hydrolyze and destroy proteins.
- $\alpha$ 1-AT has the important physiologic role of inhibiting a powerful protease that is released into the extracellular space and degrades elastin of alveolar walls, as well as other structural proteins in a variety of tissues.
- which may be important in the prevention of local tissue injury.

# Role of $\alpha$ 1-Antitrypsin in lungs

- In the normal lung, the alveoli are chronically exposed to low levels of neutrophil released from activated and degenerating neutrophils.
- This proteolytic activity can destroy the elastin in alveolar walls if unopposed by the inhibitory action of  $\alpha$ 1-AT.
- If neutrophil make proteolytic activity, emphysema results in lungs due to the destruction of the connective tissue of alveolar walls.
- $\alpha$ 1-AT prevents this proteolytic activity in alveoli.

## **Emphysema result due to $\alpha$ 1-Antitrypsin deficiency**

- **Purine base mutation GAG  $\rightarrow$  AAG, resulting in the substitution of lysine for glutamic acid at position 342 of  $\alpha$ 1-Antitrypsin is clinically the most widespread.**
- **Decreased secretion of  $\alpha$ 1-AT by the liver.**
- **A specific  $\alpha$ 1-AT methionine is required for the binding of the inhibitor to its target proteases.**
- **Smoking causes the oxidation and subsequent inactivation of that methionine residue so , the inhibitory action to neutralize elastase does not occur.**
- **Smokers with  $\alpha$ 1-AT deficiency, therefore, have a considerably elevated rate of lung destruction**
- **And a poorer survival rate than nonsmokers with the deficiency.**

$\alpha_1$ -Antitrypsin normally inhibits *elastase* released during phagocytosis by neutrophils present in alveoli of the lungs.

Neutrophil  
*elastase*

LUNG ALVEOLUS

Neutrophil

A deficiency of  $\alpha_1$ -antitrypsin permits neutrophil *elastase* to destroy lung.

$\alpha_1$ -Antitrypsin

Elastin

EXTRACELLULAR SPACE

