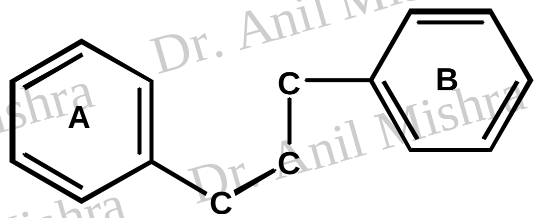
- **Anthocyanins are Natural Plant Pigments**
  - Widely distributed among flowers, fruits, and vegetables, anthocyanins belong to a group of plant compounds called flavonoids.



  - They are glycosides and their aglycons
  - Sugar free pigments are known as **Anthocyanidins**
  - These are water soluble pigments
  - Occur in Cell Sap
  - Are responsible for the large variety of colours in plants
  - Color of flowers depends upon the presence of co-pigments such as **flavones**, **flavonols etc.**
  - Recently, there has been interest in anthocyanins, not only for their colour properties, but due to their activity as antioxidants.

- **Structure**
  - Anthocyanidin is an extended conjugation made up of the aglycone of the glycoside anthocyanins. Next to chlorophyll, anthocyanins are the most important group of plant pigments visible to the human eye.

GLYCOSIDE = AGLYCONE + GLYCONE

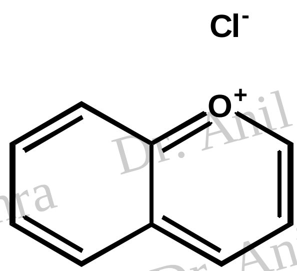
↓ ↓ ↓

ANTHOCYANIN ***ANTHOCYANIDIN*** ***SUGAR***

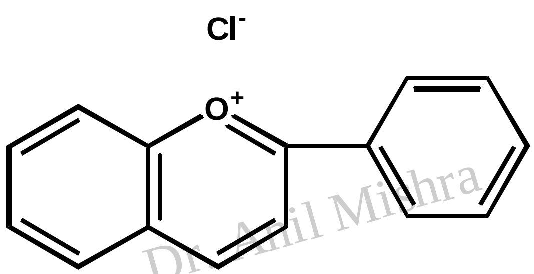
  - The different compounds contain the same carbon skeleton but differ in the nature of substituent groups
  - The sugars that are present include glucose, galactose, rhamnose, and arabinose.
  - Sugars are present most commonly at the C-3 position, while a second site for glycosylation is the C-5 position and, more rarely, the C-7 position.
  - The sugars provide additional sites for modification as they may be acylated with acids such as *p*-coumaric, caffeic, ferulic, sinapic, acetic, malonic or *p*-hydroxybenzoic acid.
  - Because of the diversity of glycosylation and acylation, there are at least 300 naturally occurring anthocyanins.

General Nature of Anthocyanins

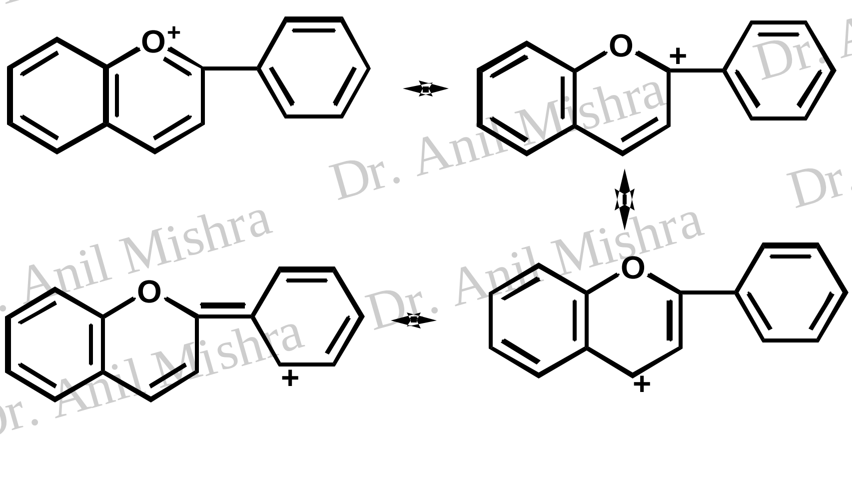
- Fundamental nucleus is **benzopyrylium chloride**



- Parent compound is **2-phenylbenzopyrylium chloride** or **Flavylium chloride**



This can be represented by a number of resonating structures

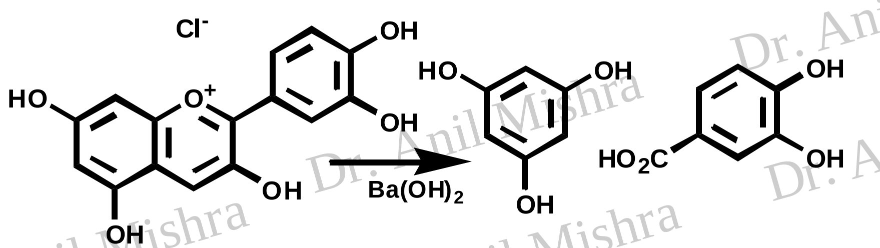


Anthocyanin

↓ HCl Hydrolysis

Anthocyanidin

- **Number of Hydroxyl and Methoxyl groups determined by analytical methods**
- **Nature of Sugar Moiety identified by usual methods of sugar chemistry**
- **Structure confirmed by the following reaction**



Cyanadin chloride **Phloroglucinol** **Protocatechuic acid**

Position of Methoxyl groups indicated by getting methylated products

There are more than 200 different types of anthocyanins that fall into six major groups

Trivial Name	Chemical Name *IUPAC name*	Occurrence
Pelargonidin	**3,4',5,7-Tetrahydroxyflavylium chloride** ***3,5,7-Trihydroxy-2-(4-hydroxy-phenyl)-chromenylium; chloride***	**Orange-red to scarlet flowers** e.g. orange red dahlia
Cyanidin	**3,3',4',5,7-Pentahydroxyflavylium chloride** ***2-(3,4-Dihydroxy-phenyl)-3,5,7-trihydroxy-chromenylium; chloride***	**Crimson to bluish flowers** e.g. red roses
Delphinidin	**3,3',4',5,5',,7-Hexaahydroxyflavylium chloride** ***3,5,7-Trihydroxy-2-(3,4,5-trihydroxy-phenyl)-chromenylium***	**Violet to blue flowers** e.g. Delphinium
Peonidin	**3,4',5,7-Tetrahydroxy-3'-methoxy-flavylium chloride** ***3,5,7-Trihydroxy-2-(4-hydroxy-3-methoxy-phenyl)-chromenylium; chloride***	**Flowers less blue than cyanidin group** e.g. red peony
Malvidin (Syringidin)	**3,4',5,7-Tetrahydroxy-3',5'-dimethoxy-flavylium chloride** ***3,5,7-Trihydroxy-2-(4-hydroxy-3,5-dimethoxy-phenyl)-chromenylium; chloride***	**Flowers less blue than Delphinidin group** *e.g. primula viscosa*
Hirsutidin	**3,4',5,-Tetrahydroxy-3',5',7-trimethoxy-flavylium chloride** ***3,4,5-Trihydroxy-2-(4-hydroxy-3,5-dimethoxy-phenyl)-7-methoxy-chromenylium; chloride***	**Present in *primula hirsuta***

Various groups of flavanoids give rise to characteristic color reactions, and so it is possible to assign a flavanoid to its class

Class	**Aqueous NaOH**	**Conc. H_2SO_4**	**Mg-HCl**
Anthocyanins	Blue to violet	Yellowish-orange	Red(fades to pink)
Flavones	Yellow	Yellow to orange	Yellow to red
Flavanols	Yellow to orange	Yellow to orange	Red to magenta
Flavanones	Yelow to orange (cold) Red to purple (hot)	Orange to crimson	Red, magenta, violet, blue
Isoflavones	Yellow	Yellow	Yellow
Leucoanthocyanins	Yellow	Crimson	pink

Position of Sugar Moiety

Anthocyanin

↓ **Methylation**

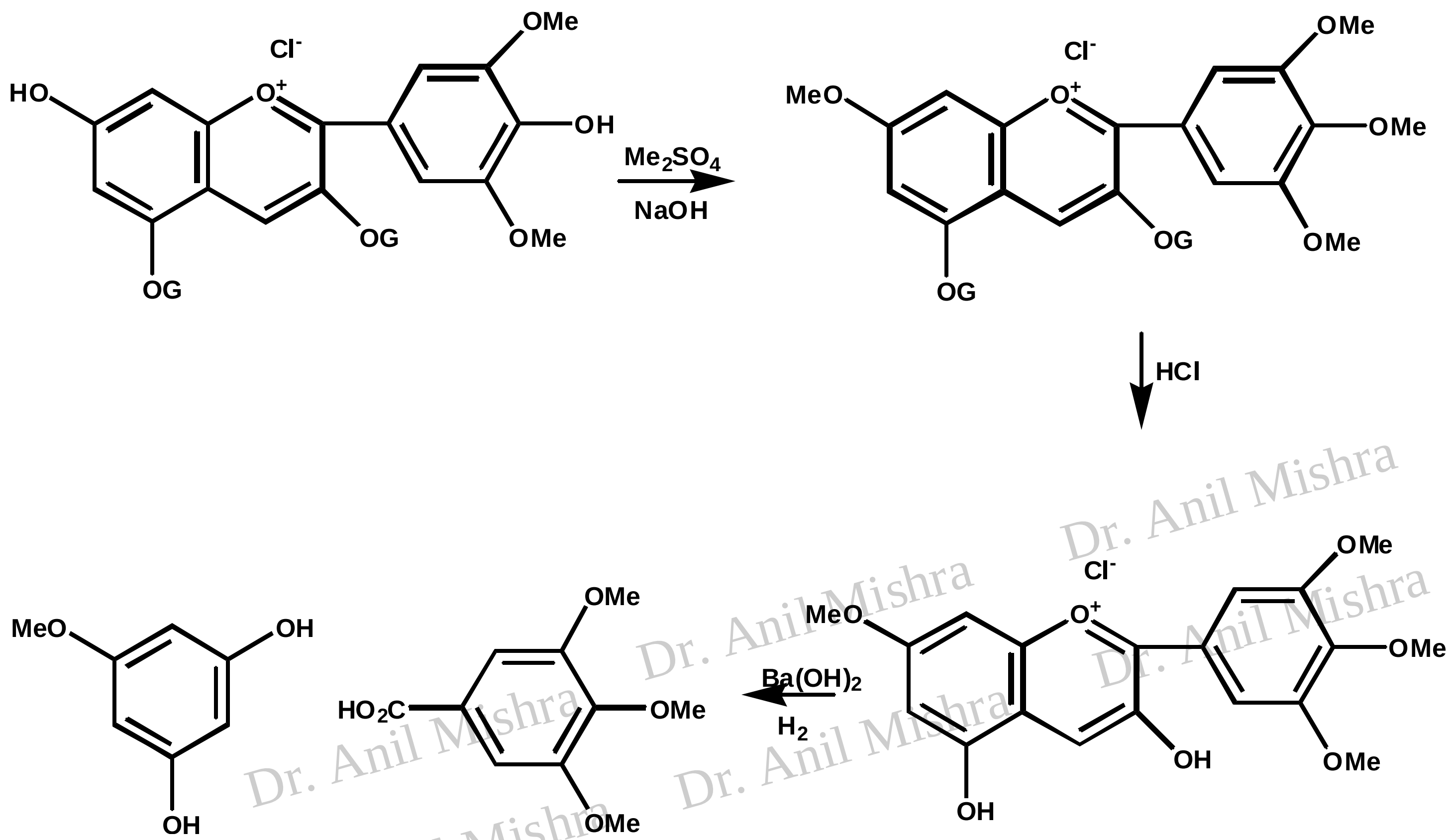
Methylated Anthocyanin

↓ **HCl Hydrolysis**

Methylated Anthocyanidin

↓ **$Ba(OH)_2$**

Position of free hydroxyl indicated the point of attachment of sugar

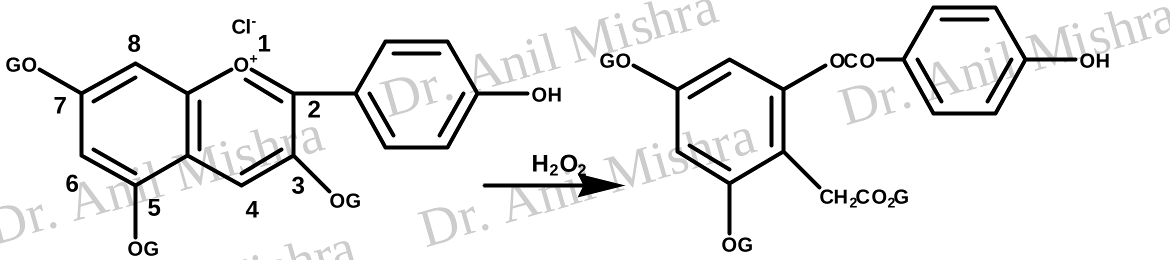


- The reactions involve
    - Methylation of Anthocyanins
    - Hydrolysis with HCl removes the sugar moiety
    - Reaction with $Ba(OH)_2$ in atmosphere of hydrogen gives two products.

POSITION OF FREE HYDROXYL GROUP INDICATES THE POINT OF ATTACHMENT OF SUGAR.

The problem now arises is that which of the hydroxyl group in monomethylphloroglucinol was attached to the sugar?

Position of sugar can be confirmed by the following reaction



A

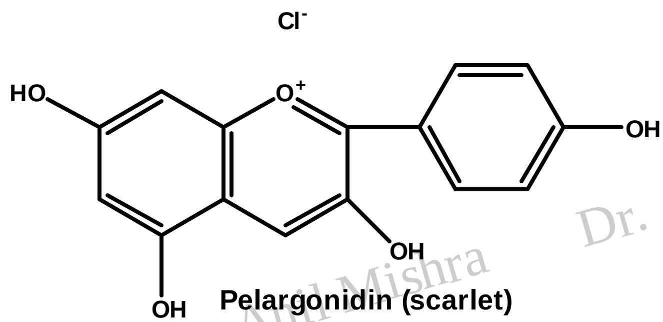
B

- **Reaction of hydrogen peroxide on anthocyanin A gives B**
  - **If A has a sugar residue at position 3, this sugar residue in B will be readily hydrolyzed by dilute ammonia.**
  - **If the sugar residue is at position 5 or 7, this can only be removed by heating with dilute HCl.**

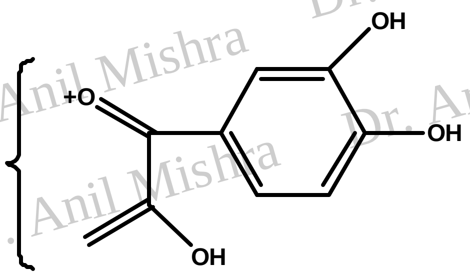
Position 3 can be distinguished from 5 and 7 but the latter two cannot be distinguished from each other.

- **Anthocyanins with a free hydroxyl at position 3 are rapidly oxidized by ferric chloride, the anthocyanins are rapidly decolorized in this reaction.**

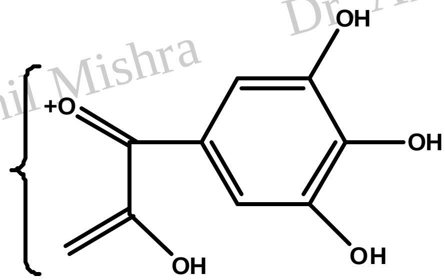
- **Nature of the sugar linkage (a or b) is confirmed by the enzyme maltase (a linkage) and emulsin (b linkage)**



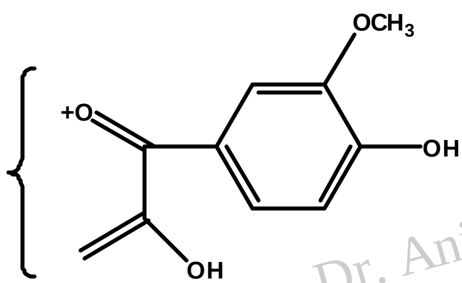
Pelargonidin (scarlet)



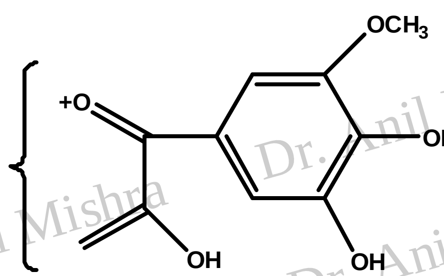
Cyanidin(crimson)



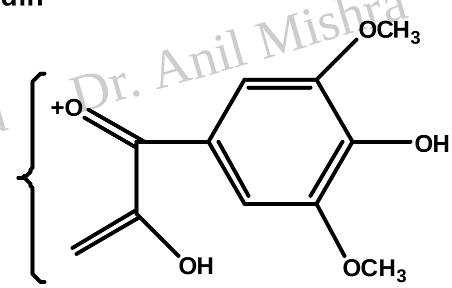
Delphinidin(mauve)



Peonidin

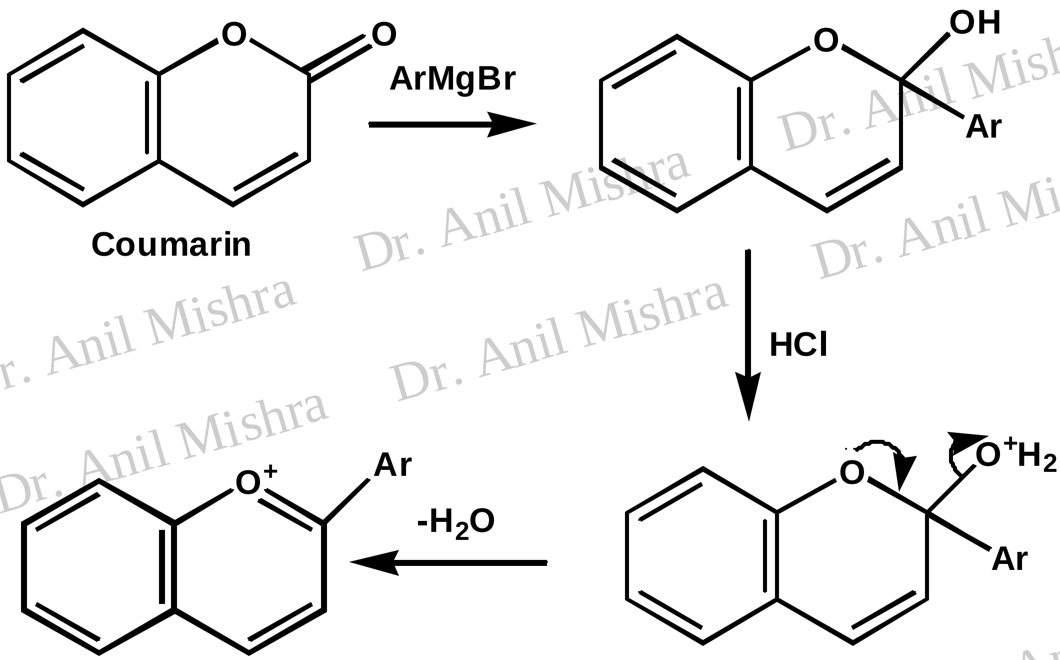


Hirsutidin

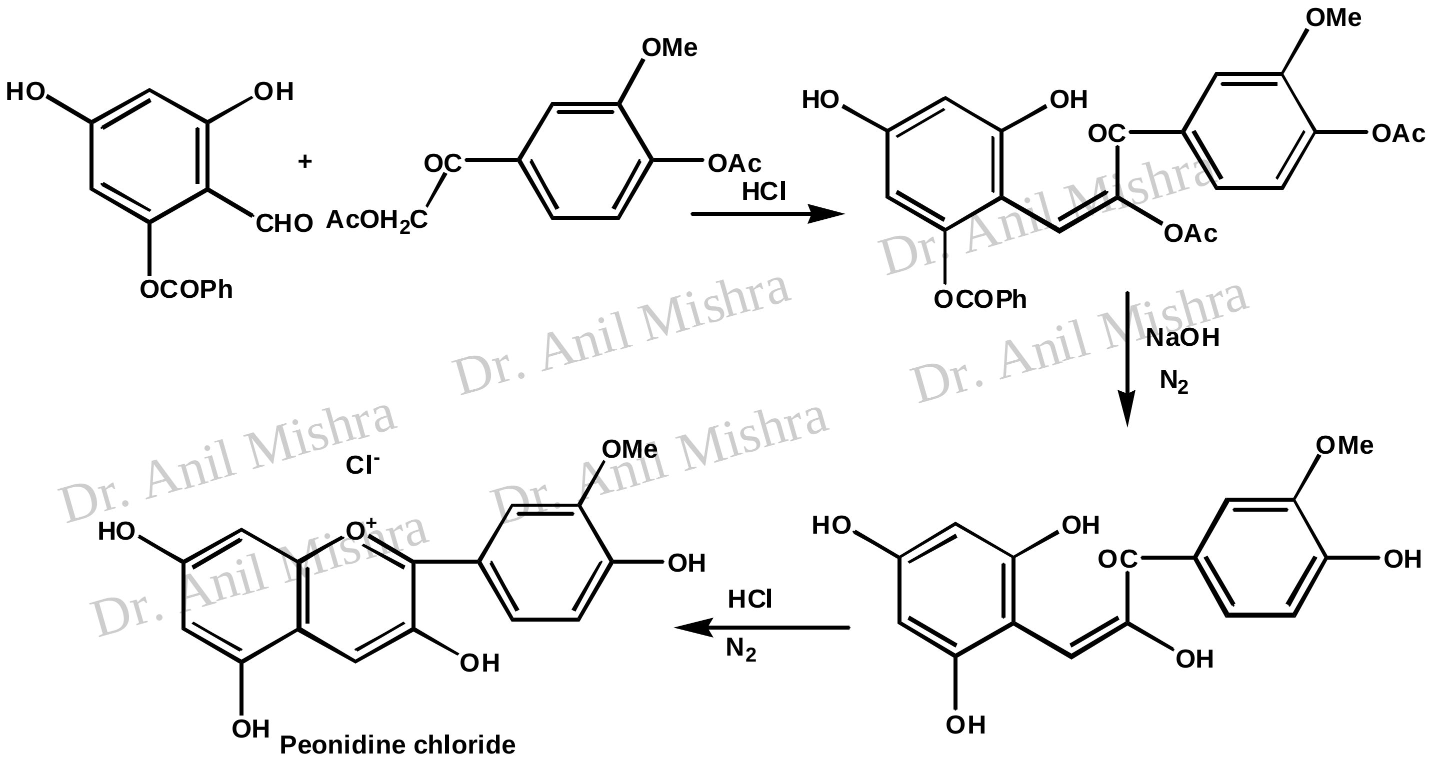


Malvidin

Willstätter's Synthesis

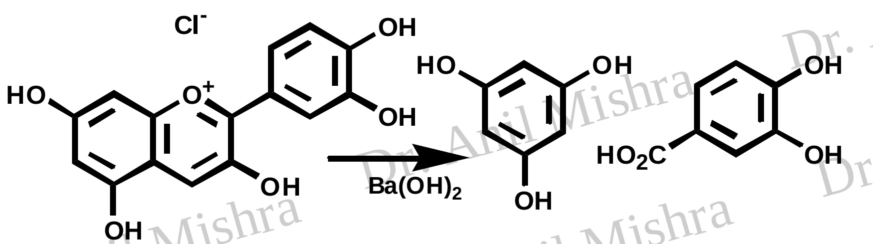


Robinson's Method



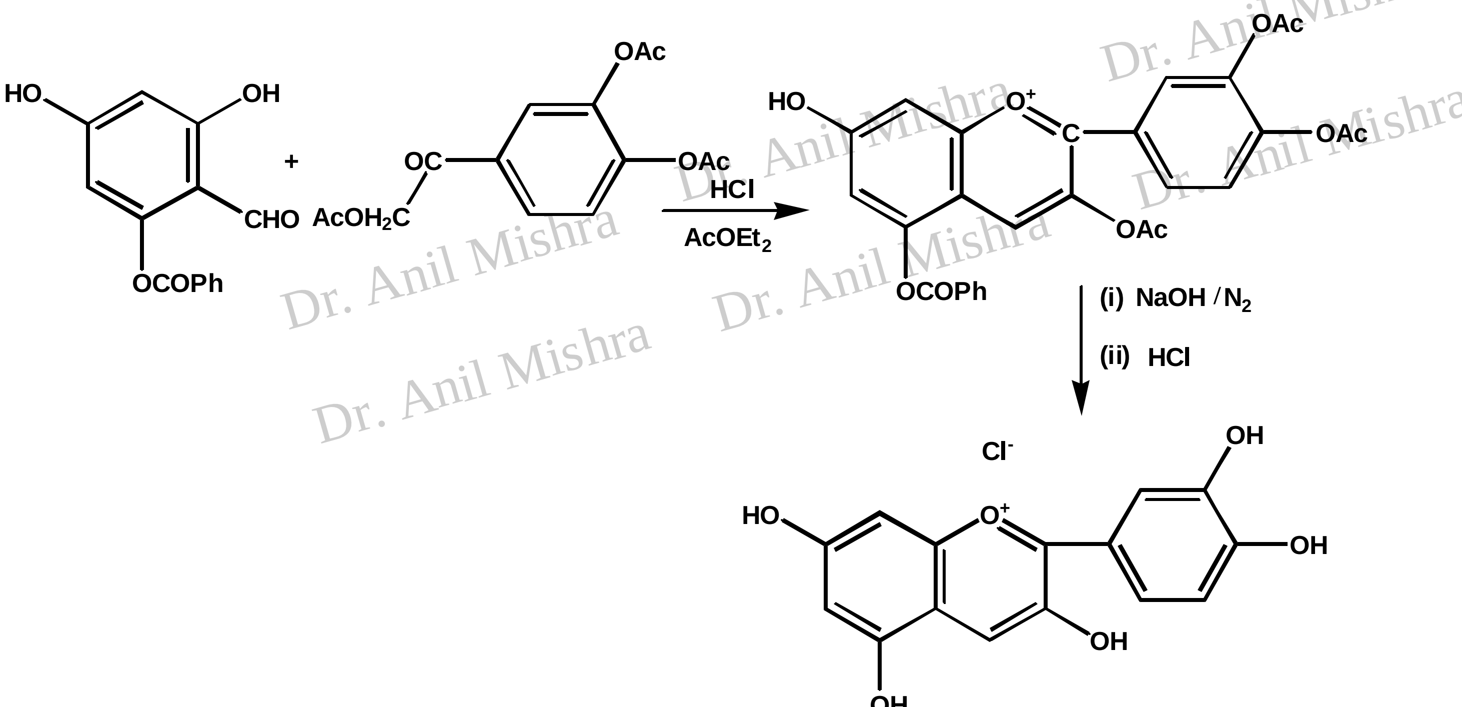
Structure of Cyanidin Chloride

- Hydrolysis of Cyanin chloride gives cyanidin chloride and two molecules of D-glucose
  - It contains two D-Glucose moieties
- Forms a penta-acetate
  - Contains five hydroxyl groups and no methoxyl groups
- KOH fusion degrades the molecule into phloroglucinol and protocatechuic acid



Cyanadin chloride Phloroglucinol Protocatechuic acid

- Structure confirmed by Robinson's synthesis



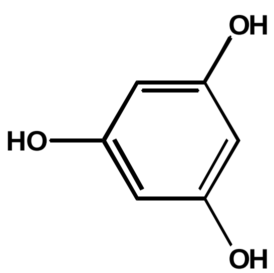
Cyanidin Chloride

Hydrolysis of Anthocyanins

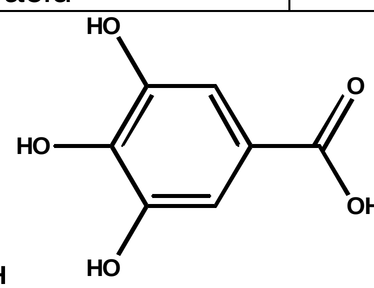
Anthocyanin	**Anthocyanidin**	**No. of Sugars** **Name of Sugar**	**Other product** **No. of molecules** **Name**
Cyanin Chloride	**Cyanidin Chloride**	2 D-Glucose	
Pelargonin Chloride	**Pelargonidin Chloride**	2 D-Glucose	
Delphinin Chloride	**Delphinidin Chloride**	2 D Glucose	2 *p*-hydroxybenzoic acid
Peonin Chloride	**Peonidin Chloride**	2 D Glucose	
Malvin Chloride	**Malvidin Chloride**	2 D Glucose	
Hirsutin Chloride	**Hirsutidin Chloride**	2 D Glucose	

Degradation

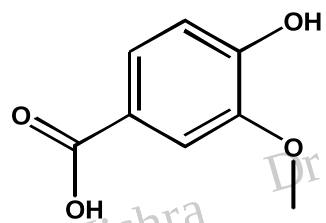
Anthocyanidin	Reagent	Products	No of Methoxyl groups
Cyanidin Chloride	KOH	➢ Phloroglucinol ➢ Protocatechuic acid	0
Pelargonidin Chloride	KOH	➢ Phloroglucinol ➢ *P*-hydroxybenzoic acid	0
Delphinidin Chloride	KOH	➢ Phloroglucinol ➢ Gallic acid	0
Peonidin Chloride	KOH	➢ Phloroglucinol ➢ 4-hydroxy-3-methoxybenzoic acid	1
Malvidin Chloride	Boiling $Ba(OH)_2$	➢ Phloroglucinol ➢ Syringic acid	2
Hirsutidin Chloride	Boiling $Ba(OH)_2/H_2$	➢ Monomethyl phloroglucinol ➢ Syringic acid	3



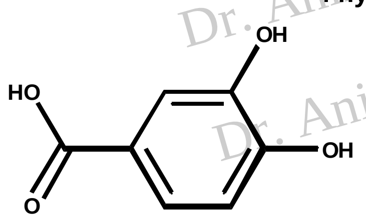
phloroglucinol



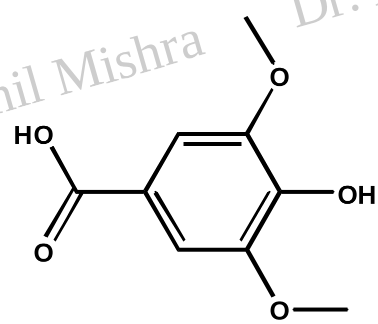
gallic acid



4-hydroxy-3-methoxybenzoic acid



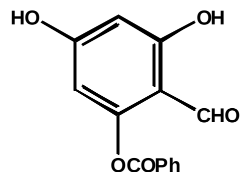
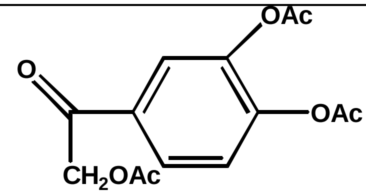
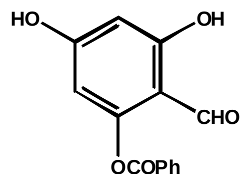
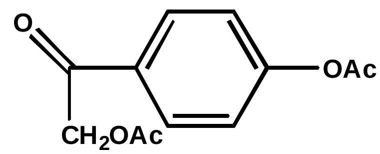
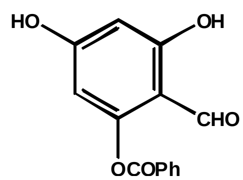
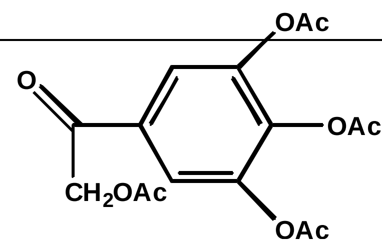
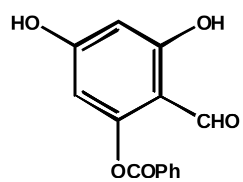
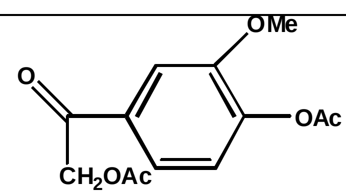
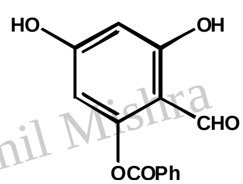
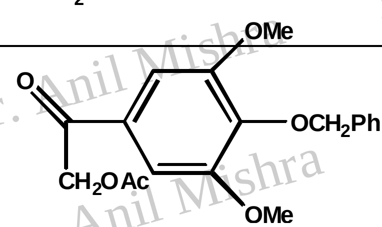
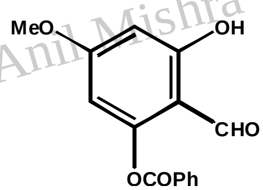
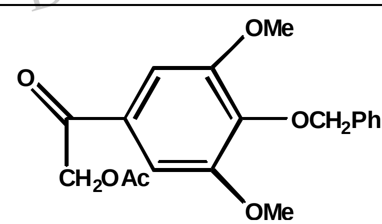
Protocatechuic acid



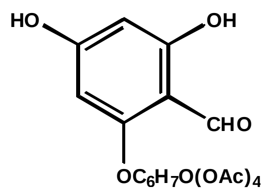
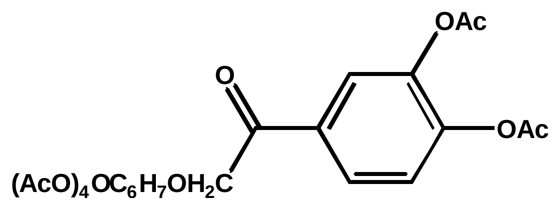
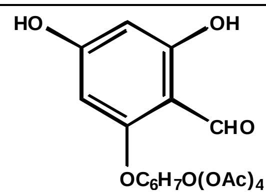
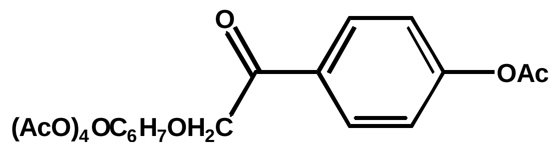
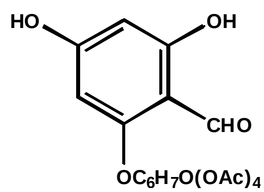
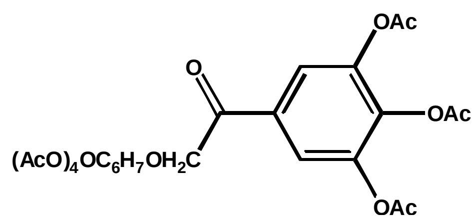
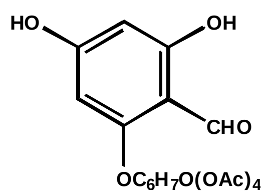
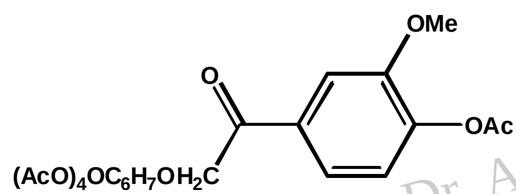
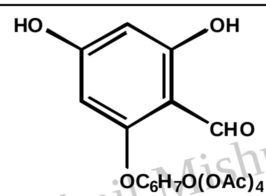
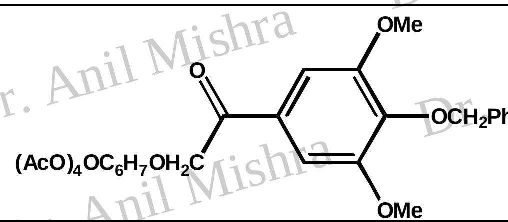
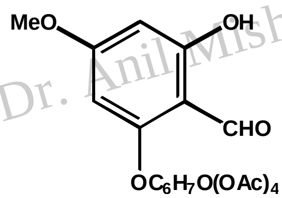
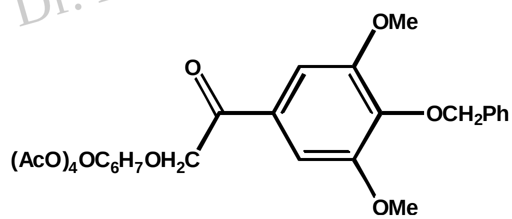
syringic acid

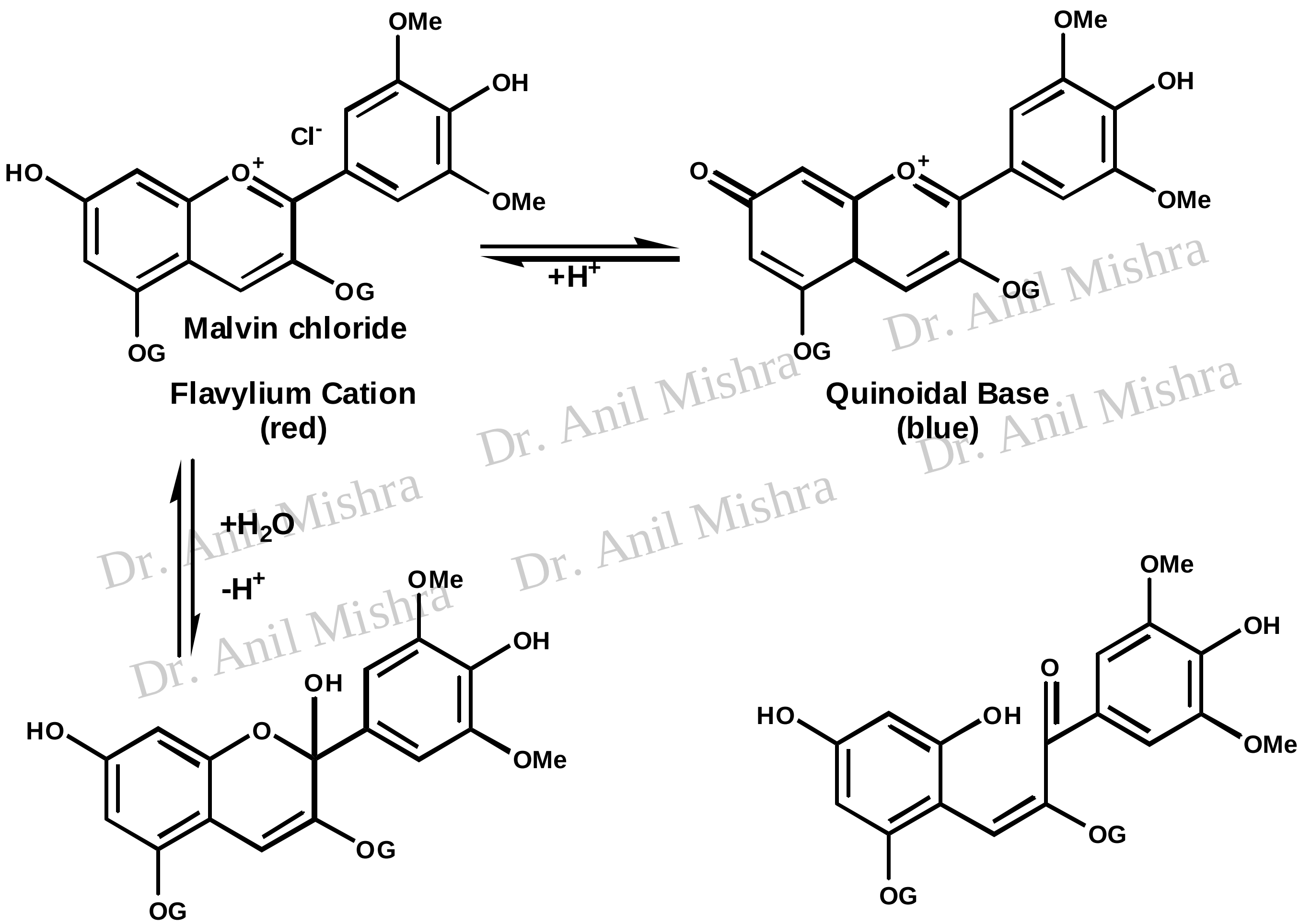
Anthocyanidin	Derivative	No of hydroxyl groups	No of Methoxyl groups
Cyanidin Chloride	Penta acetate	5	0
Pelargonidin Chloride	Tetra acetate	4	0
Delphinidin Chloride	Hexa-acetate	6	0
Peonidin Chloride	Tetra-acetate	4	1
Malvidin Chloride	Tetra-acetate	4	2
Hirsutidin Chloride	Tri-acetate	3	3

Robinson's synthesis of ANTHOCYANIDINS

Anthocyanidin	Phloroglucinol Derivative	Acetophenone Derivative
Cyanidin Chloride		
Pelargonidin Chloride		
Delphinidin Chloride		
Peonidin Chloride		
Malvidin Chloride		
Hirsutidin Chloride		

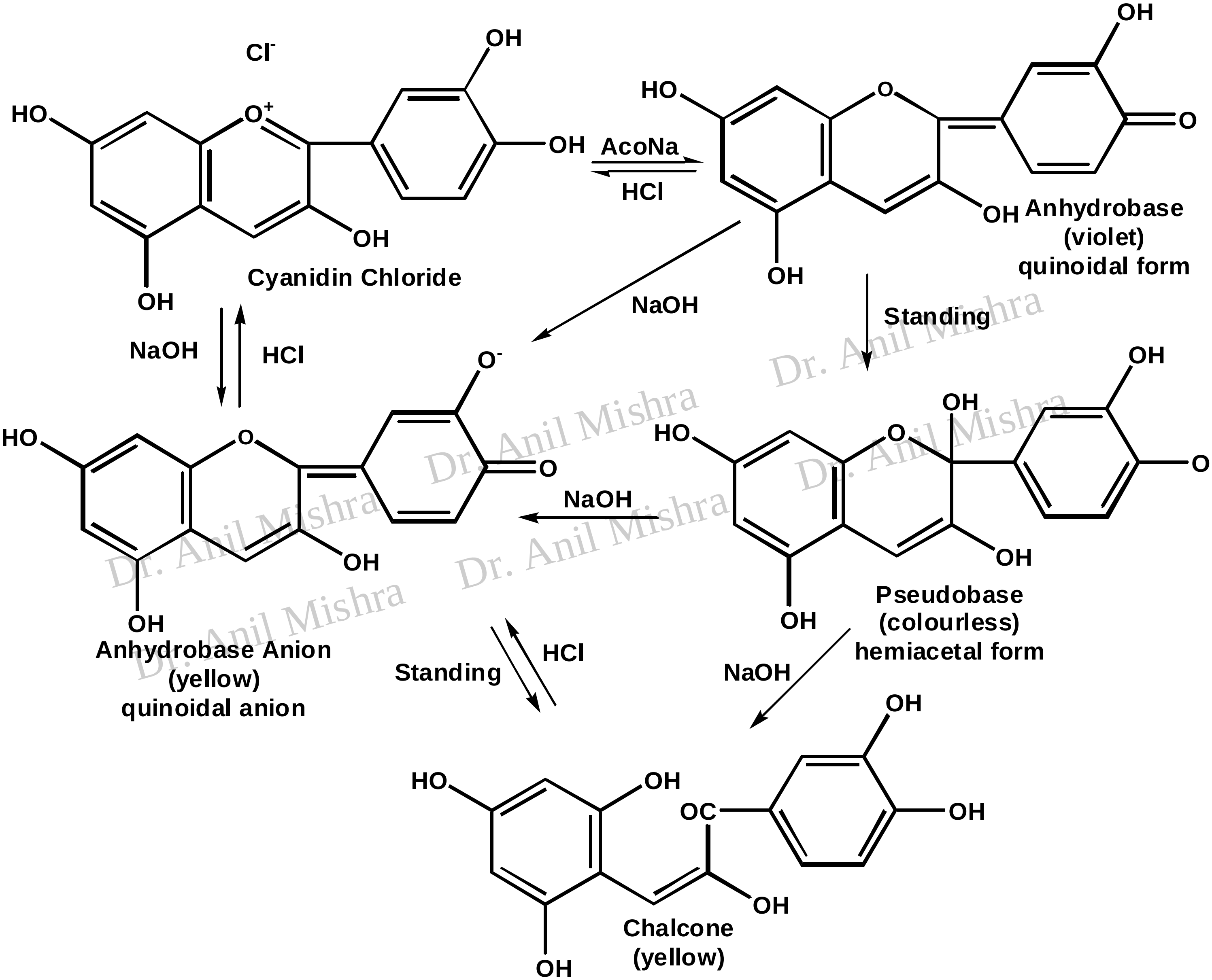
Robinson's synthesis of ANTHOCYANINS

Anthocyanin	Phloroglucinol Derivative	Acetophenone Derivative
Cyanin Chloride		
Pelargonin Chloride		
Delphinin Chloride		
Peonin Chloride		
Malvin Chloride		
Hirsutin Chloride		

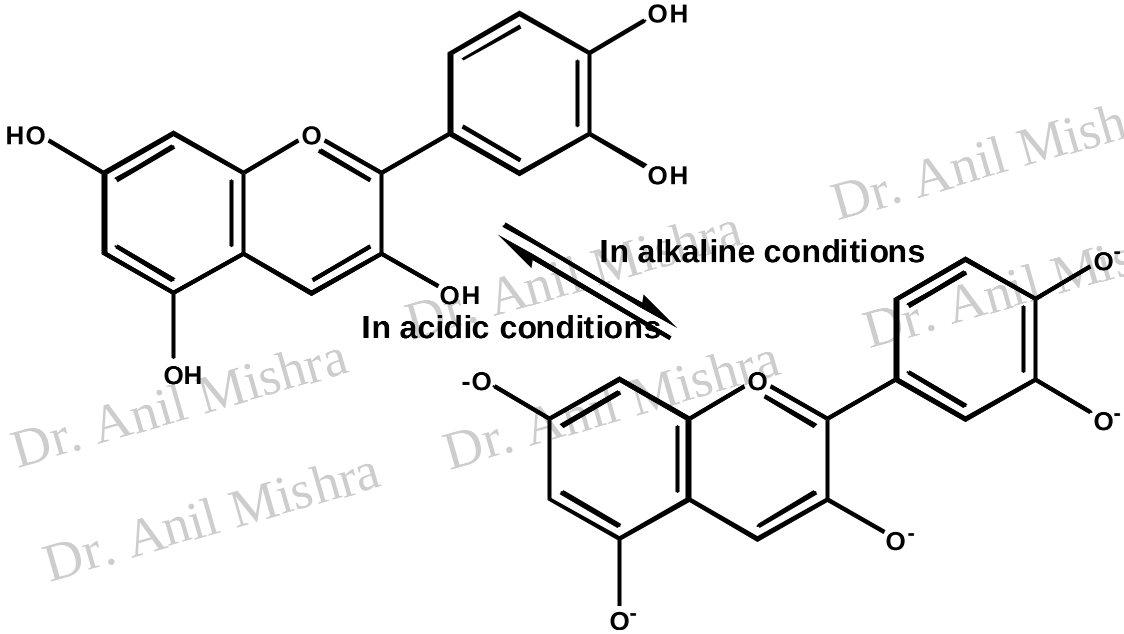


Carbinol pseudo base
(colourless)

Chalcone
(colourless)

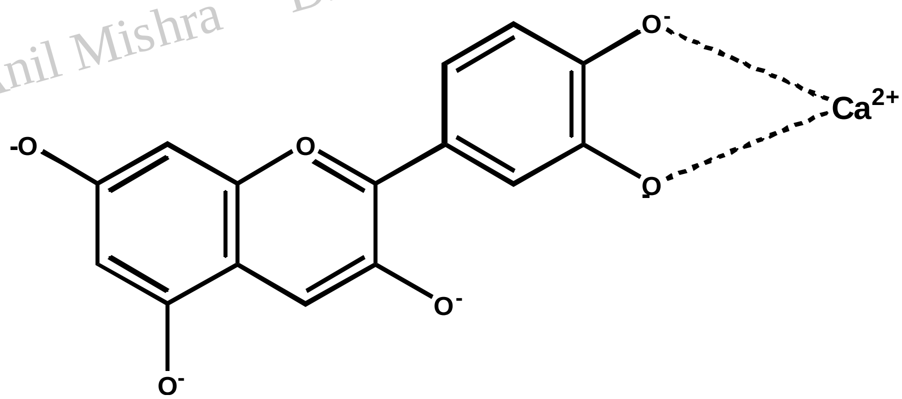

Cl-
O+
HO
OH
OH
OH
OH
Cyanidin Chloride
AcoNa
HCl
HO
O
O
OH
OH
Anhydrobase
(violet)
quinoidal form
NaOH
HCl
NaOH
Standing
O-
HO
O
O
OH
OH
Anhydrobase Anion
(yellow)
quinoidal anion
NaOH
OH
HO
O
OH
OH
OH
Pseudobase
(colourless)
hemiacetal form
Standing
HCl
NaOH
HO
OH
OC
OH
OH
OH
OH
Chalcone
(yellow)

Effect of pH

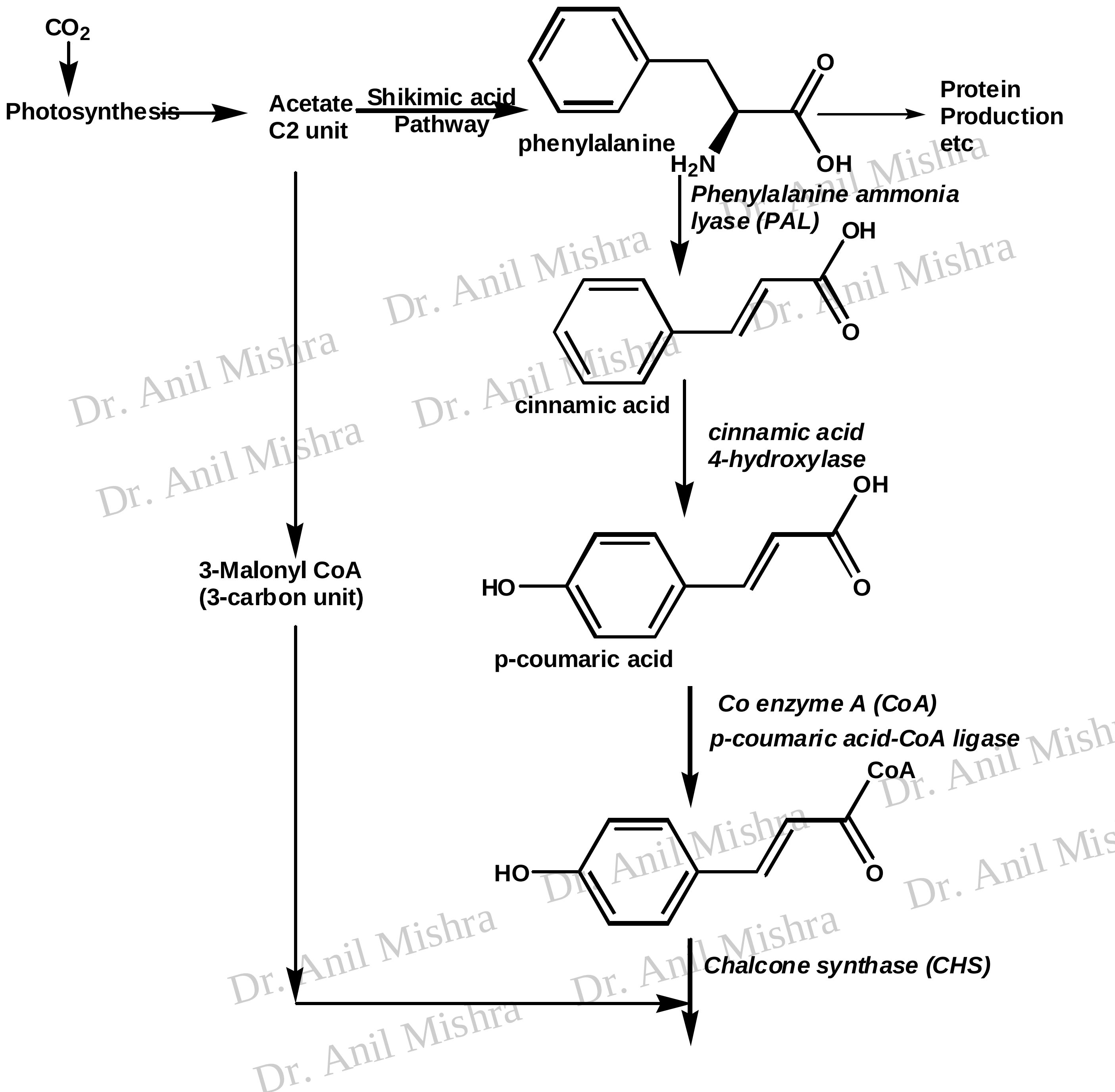

HO
O
OH
OH
OH
OH
In alkaline conditions
In acidic conditions
-O
O
O-
O-
O-
O-

CHELATES WITH METALS

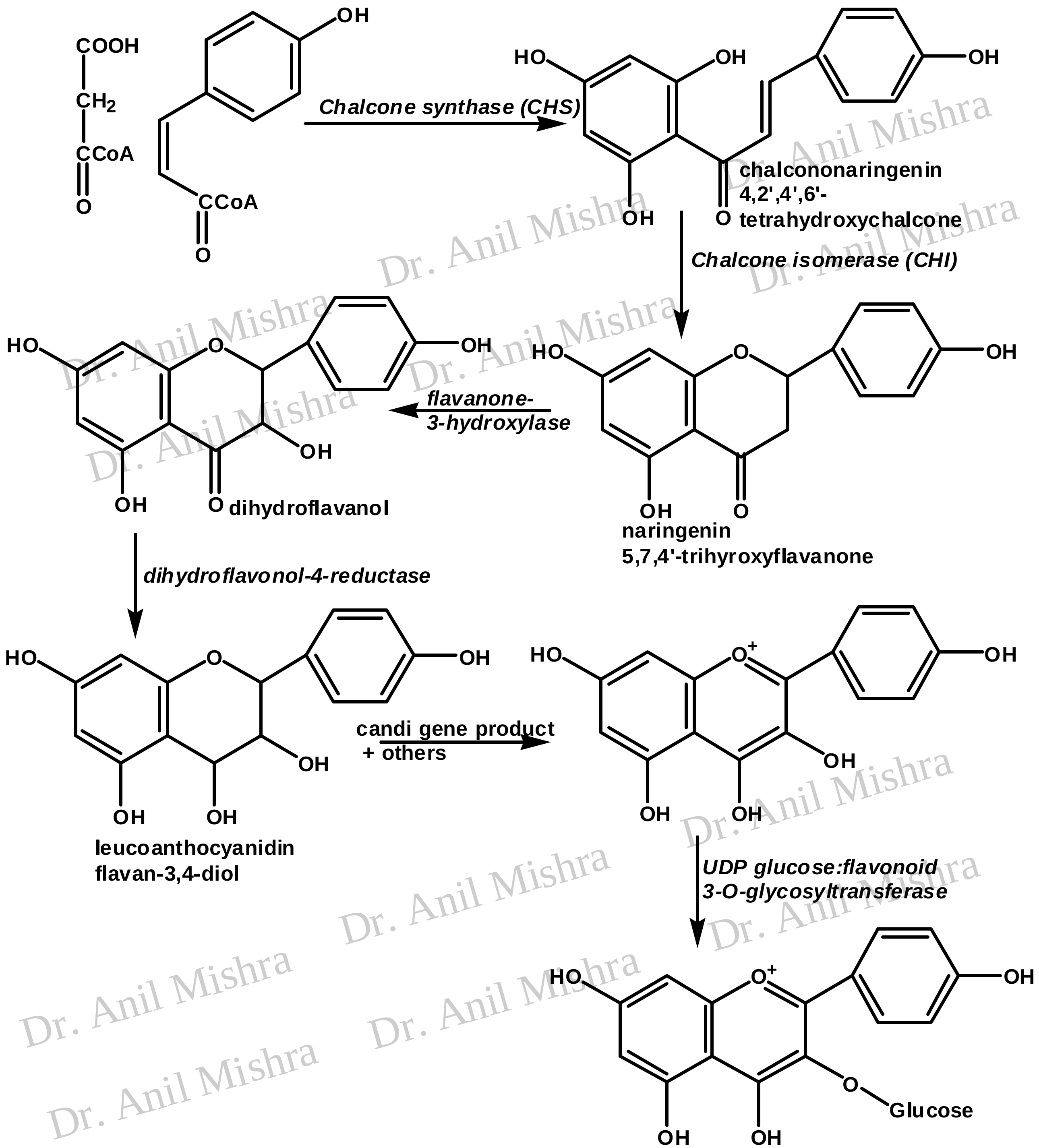
- Some metals, such as Fe^{+3} Al^{+3} and Ca^{+2} form deeply coloured **coordination complexes** with anthocyanins that have ortho-dihydroxy groups on the B-ring.
- Such metalo-anthocyanin complexes have been found to produce discolouration in some canned fruit products, including pears and peaches.



ANTHOCYANIN BIOSYNTHESIS 1



ANTHOCYANIN BIOSYNTHESIS 2



Anthocyanin Content Of Common Fruits

Fruit	Amount in mg/100g	Pelargonidin	Peonidin	Cyanidin	Delphinidin	Malvidin
Red Apple, Pear				X		
Bilberry	300-648	X	X	X	X	X
Blackberry	82-325, mostly 150			X		
Blueberry	25-495		X	X	X	X
Boysenberry	160			X		
Sweet Cherries	350-450		X	X		
Cranberry	50-80		X	X		
Hawthorne	400			X		
Loganberry	77			X		
Peach			X	X		
Plum				X		
Raspberry	213-428	X				
Strawberry	varies widely	X				

Anthocyanins apparently play a major role in several plant processes

- **Attracting insects for the purpose of pollination.** Advantage is made of the fact that the pigments absorb strongly in the UV (ultraviolet), visually attracting insects but with light wavelengths that are invisible to humans. These pigments play a major role in plant pollination - and in predation in carnivorous plants, attracting insects into the trap apparatus.

- Anthocyanin-related pigments serve as a **UV screen** and are produced in response to exposure of the plant to UV radiation, protecting the plant's DNA from damage by sunlight.

- Anthocyanins serve as **anti-feedents**, their disagreeable taste serving to deter predatory animals.

Therapeutic Uses

- **Anti-inflammatory**
- **Antioxidant**
- **Bleeding, excessive post-operative**
- **Blood Clots**
- **Blood Platelet Stickiness**
- **Blood Purification**
- **Bone and Joint Problems**
- **Breast Tenderness**
- **Breathing Disorders**
- **Bronchitis**
- **Cataracts**
- **Circulatory Disorders**
- **Diarrhea**
- **Enteritis**
- **Eyesight Disorders**
- **Fibrocystic Breast Disease**
- **Hemorrhoids**
- **Leg Vein Health**
- **Lymphatic System Disorders**
- **Microangiopathy**
- **Skin Problems**
- **Sore Throat**
- **Urinary Tract Problems**
- **Vascular Disorders**