

Soft fruit anthocyanins and their impact on oxidative stress in model systems

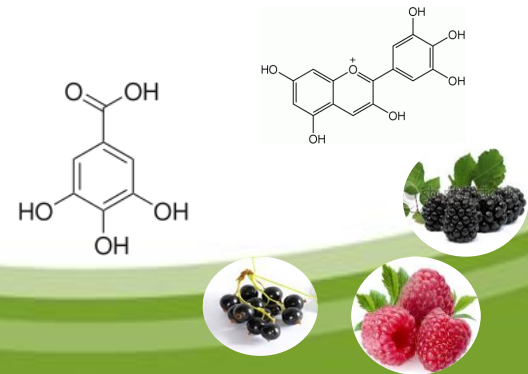
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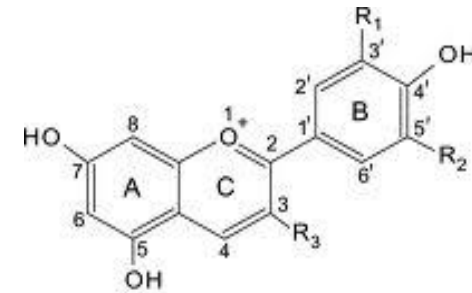
The James
Hutton
Institute

 Future-proofing berryfruit
CLIMAFRUIT



Anthocyanins...

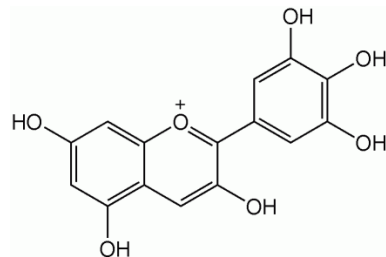
- Belong to the group of polyphenols and family of flavonoids
- Found in number of foods
- Pigments that gives fruits and vegetables their rich coloring
- Health-promoting benefits



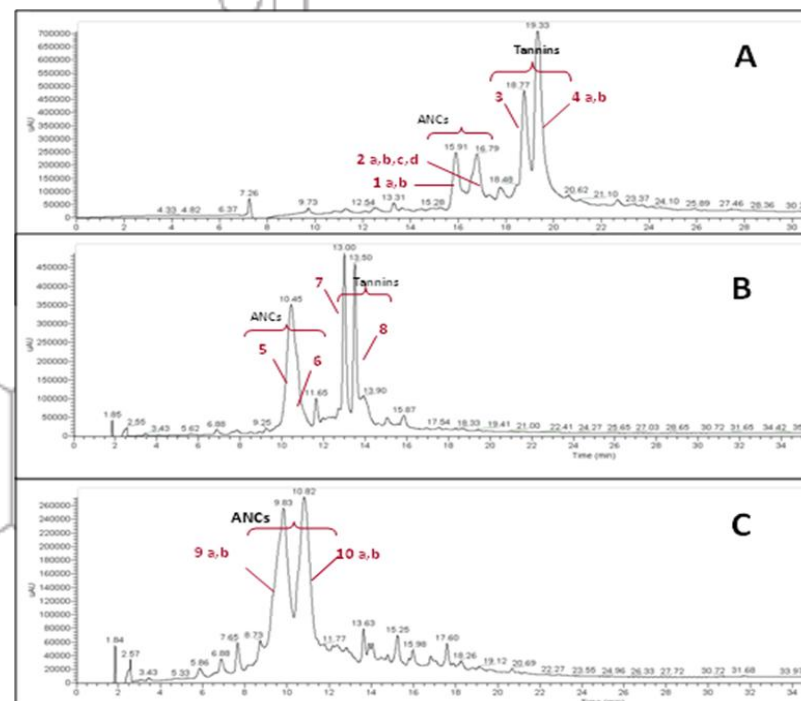
Aglycone	R ₁	R ₂	R ₃
Delphinidin	OH	OH	OH
Cyanidin	OH	H	OH
Petunidin	OCH ₃	OH	OH
Peonidin	OCH ₃	H	OH
Malvidin	OCH ₃	OCH ₃	OH
Pelargonidin	H	H	OH



Delphinidin...



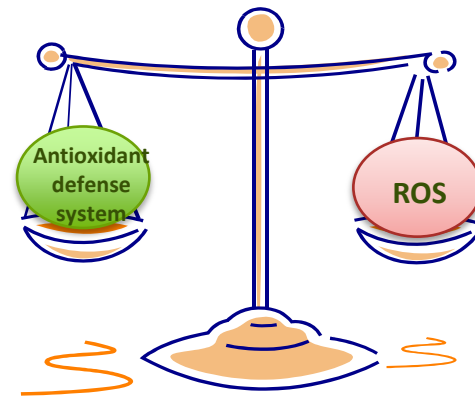
- Belongs to the family of flavonoids and subgroup anthocyanins
- Can be found in the berries (blackcurrants) and grapes (red wine) and is responsible for giving a blue/purple tinge
- Delphinidin is pH-sensitive and changes colour to red/purple in acidic solution and to blue in alkali solution
- Antioxidant potential...?



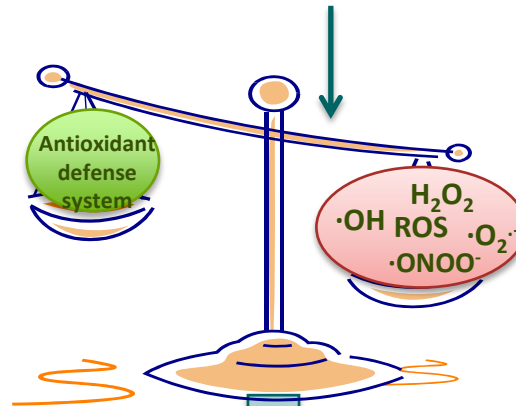
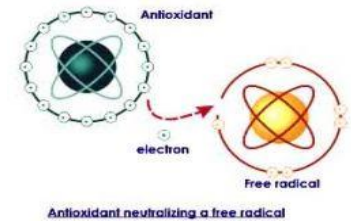
PDA profile of phenolic compounds in raspberry (A), blackberry (B) and blackcurrant (C) extracts

- Cyanidin-3-*O*-glucoside (2b,5,10a)
- Cyanidin-3-*O*-rutinoside (2d,6,10b)
- Delphinidin-3-*O*-glucoside (9a)
- Delphinidin-3-*O*-rutinoside (9b)

Oxidative stress cause of cell aging and death



Normal, balanced stage



Source of Reactive Oxygen Species

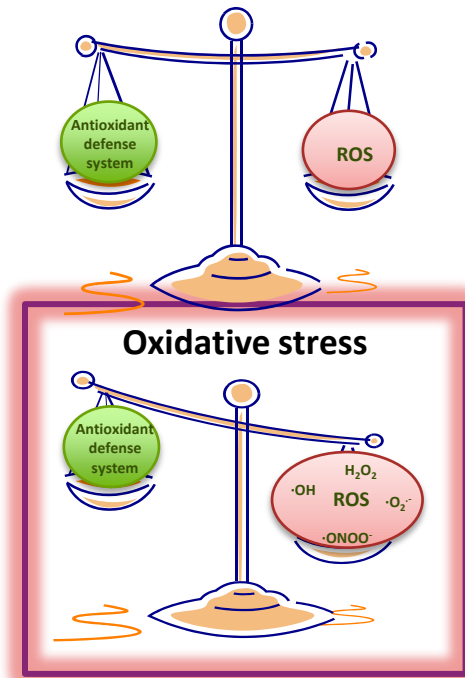
- Air pollution
- UV rays
- Bio products from food and chemicals metabolism
- Cellular respiration

- 
- | | |
|----------------------|----------------------|
| • Biomembrane damage | • Protein damage |
| • DNA damage | • Lipid peroxidation |

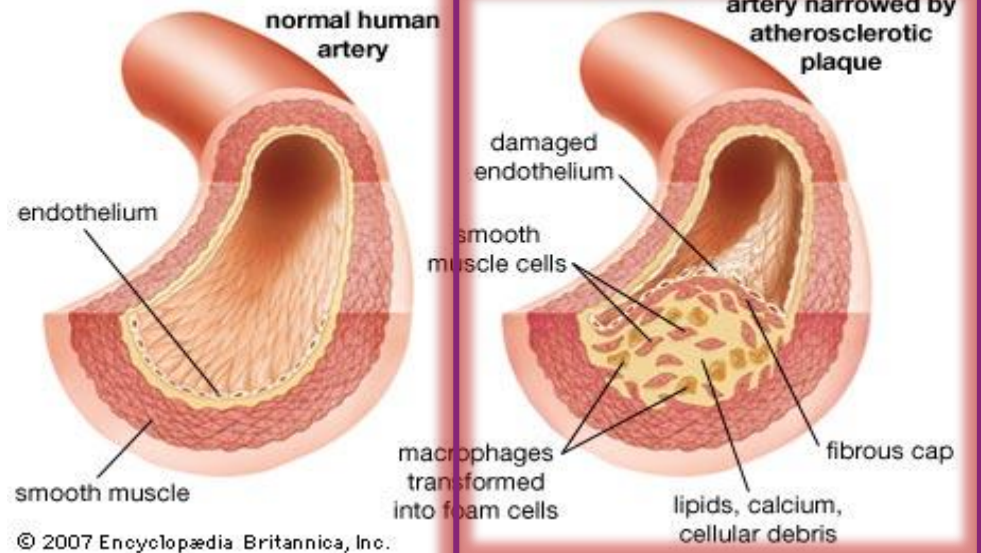


Cancer, CVD, Diabetes type I II, Neurodegenerative diseases

Free radicals cause cardiovascular diseases development



Atherosclerosis



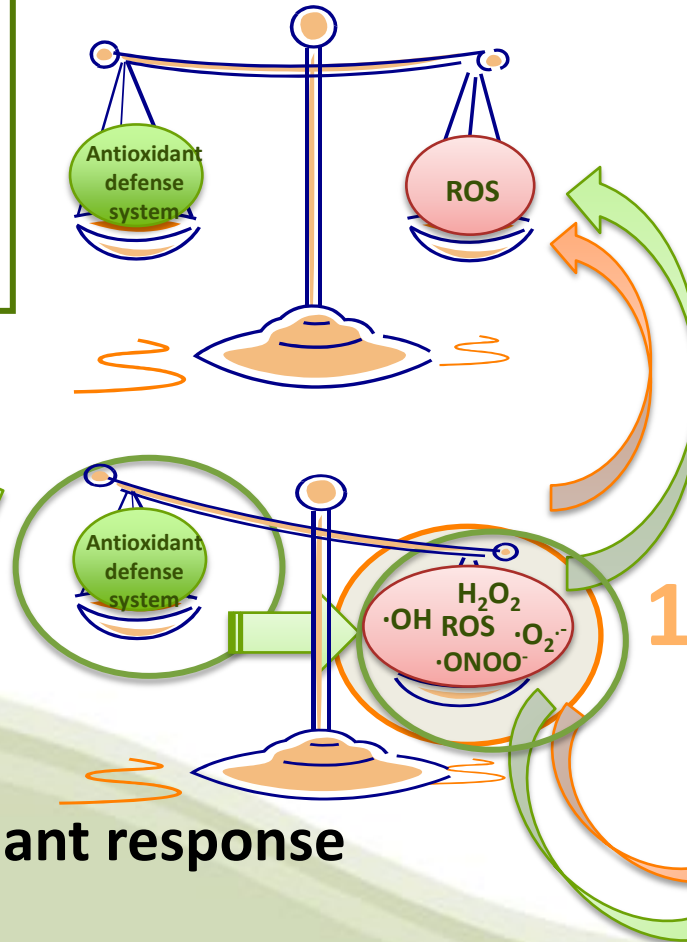
- Protein damage and lipid peroxidation leads to loss of the membrane integrity, cell damage and death
- Biomembrane damage leads to the cell death
- DNA damage leads to mutations

**Stroke, Atherosclerosis,
High Blood Pressure, Heart
attack**

Possible role of polyphenols...

- POLYPHENOLS can act through co-action with cell membrane receptors
- Induction of the cell signaling pathways
- Modulation of the genes and proteins expression

2

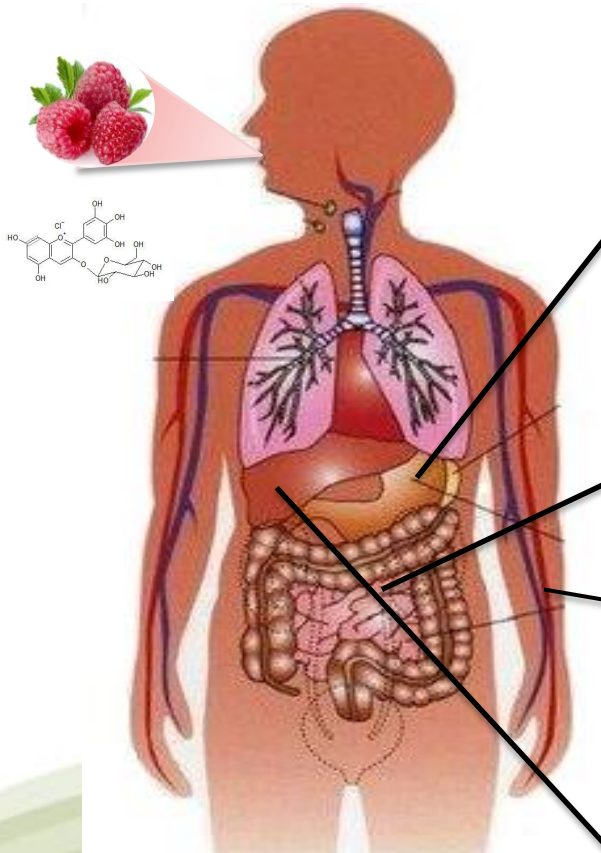


1. Direct antioxidant response

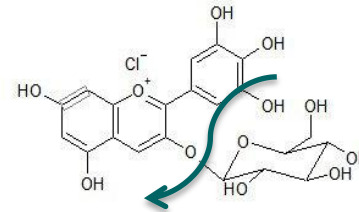
2. Indirect antioxidant response

O₂, H₂O, NO₂ etc.
Not harmful for
the cells anymore

Absorption and metabolism of delphinidin



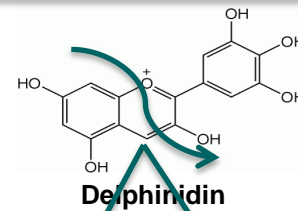
■ Stomach



Delphinidin-3-O-glucoside

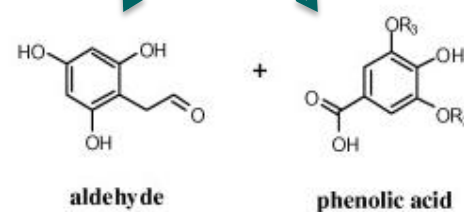
■ pH ~ 4

■ Small intestine



■ pH ~ 7 - 8

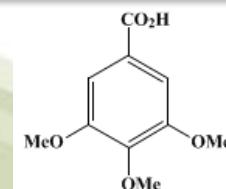
■ Blood



■ pH ~ 7.4

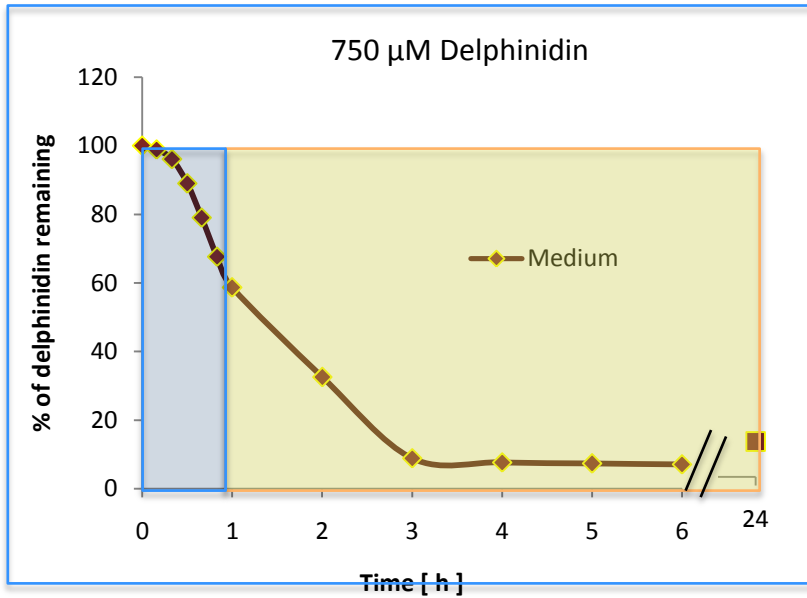
proglucinaldehyde + gallic acid

■ Liver



- Methylation
- Sulphation
- Glucuronidation

Delphinidin is very unstable compound



- Delphinidin degrades rapidly in cell culture medium (half life - less than 2h)

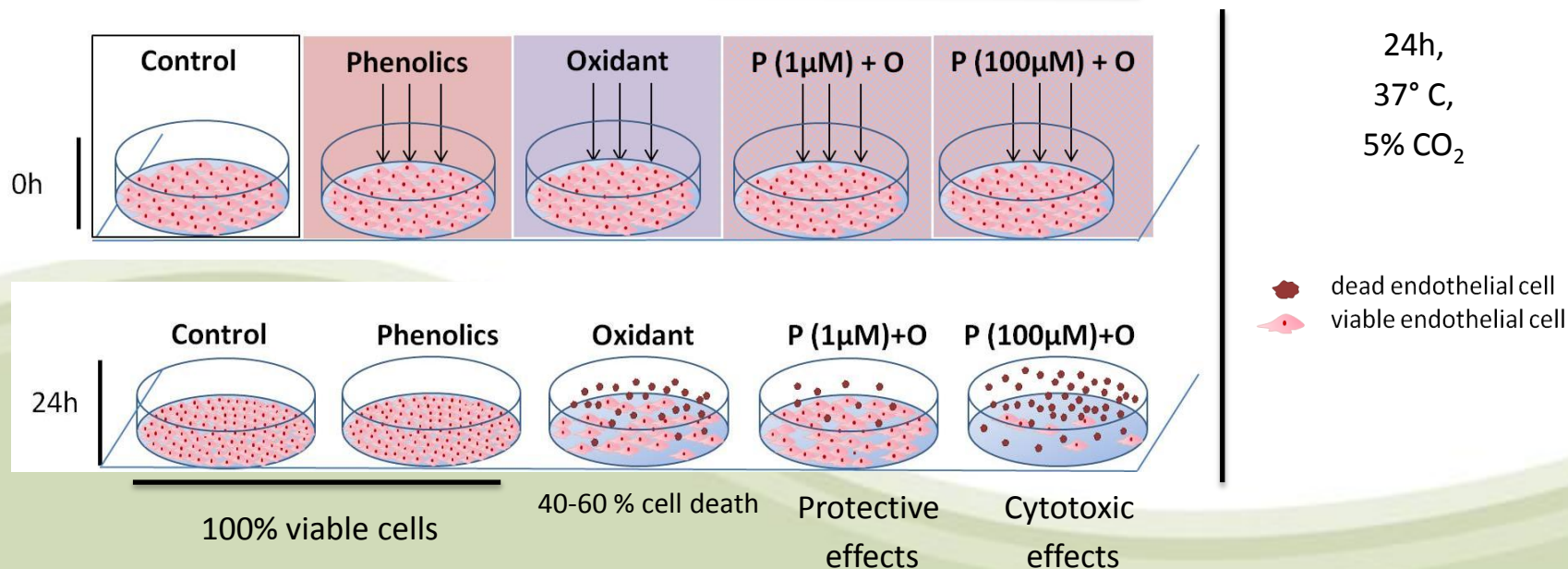
- Possible degradation products are: gallic acid and phloroglucinaldehyde

Media		pH	Delphinidin max absorbance [nm]
Cell culture medium (phenol red -free)		7.37	585

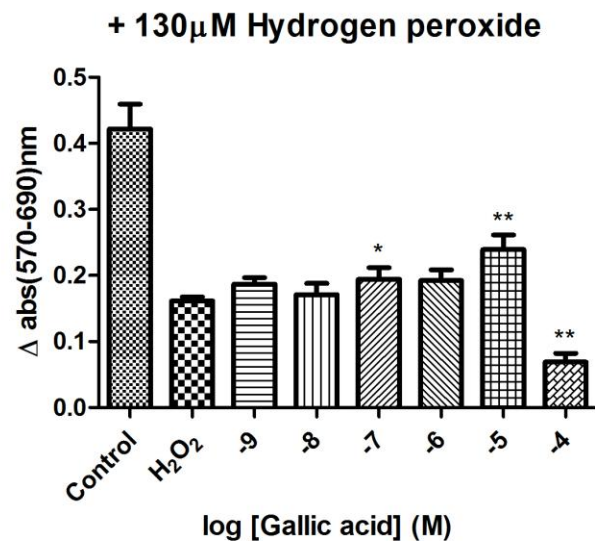
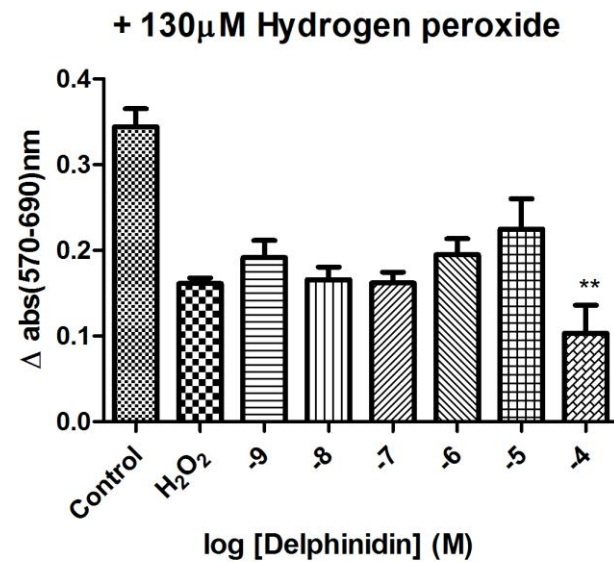
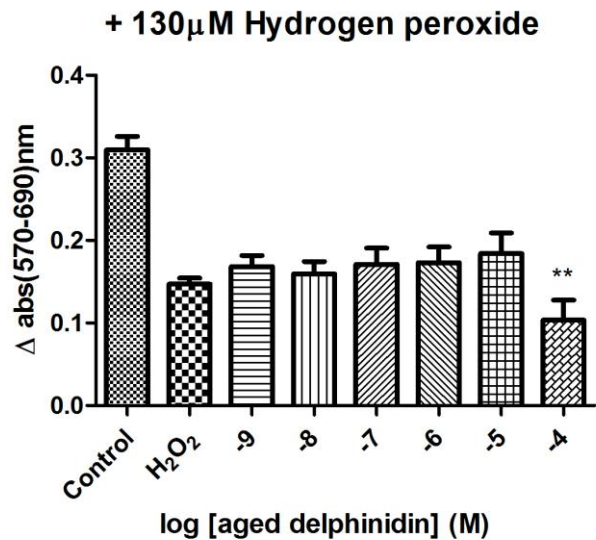
Testing protective effect of selected polyphenols against model of oxidative stress

HUVECs (human umbilical vein endothelial cells)	Oxidants	Concentrations	Phenolics	Concentrations
80-90% confluent cells used for treatment	H ₂ O ₂	130μM	Delphinidin	1nM, 10nM, 100nM, 1μM, 10μM, 100μM,
	Pyrogallol (O ₂ ⁻)	140μM	"Aged Delphinidin"	
Oxidants caused approx. 40-60% cell death			Gallic acid	
			Phloroglucinaldehyde	

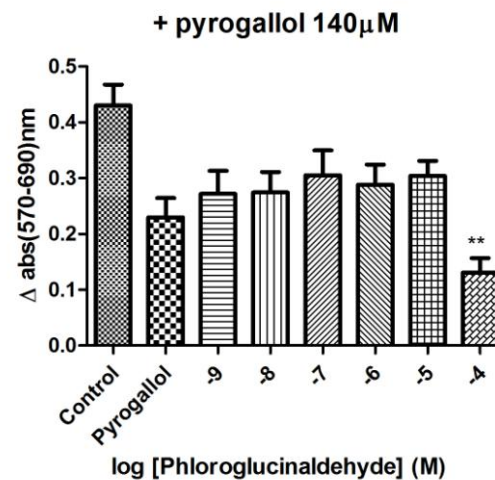
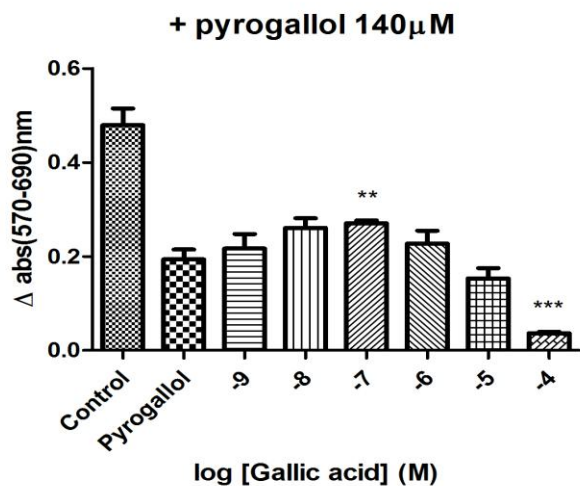
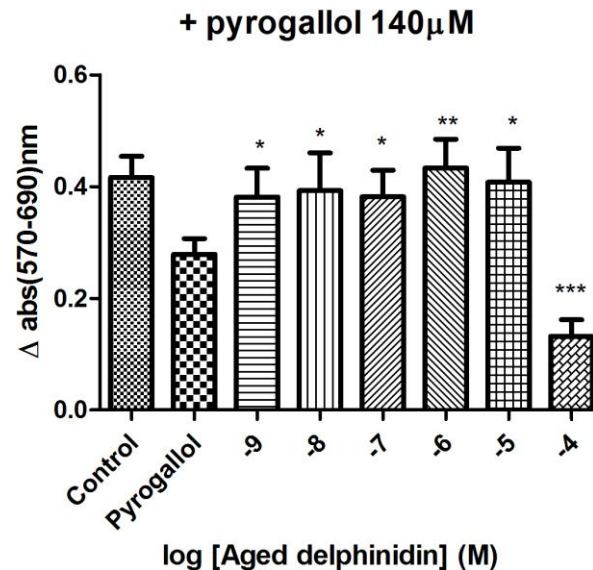
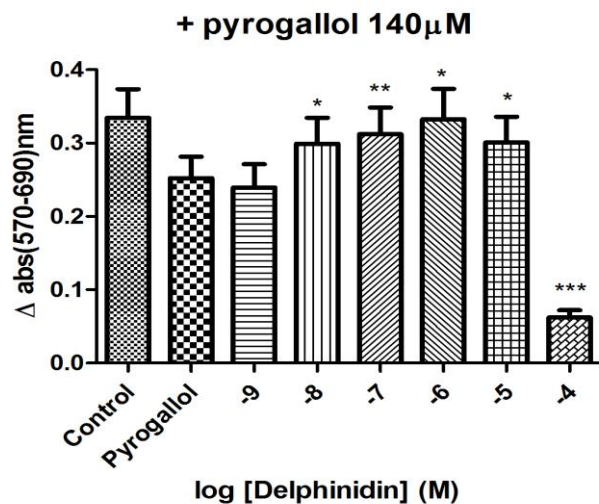
Co-treatment



Phenolics vs Hydrogen Peroxide



Phenolics vs Pyrogallol ($O_2^{\cdot-}$)



Conclusions

- Delphinidin might be spontaneously degraded to gallic acid and/or phloroglucinaldehyde
- Gallic acid rather than the parent compound itself might be responsible for protective effects
- Appropriate (low) concentrations important to mimic their mode of action *in vivo*
- Polyphenols have significant cytotoxic effects at concentrations at or above 10 μM
- Delphinidin and gallic acid both caused similar protective effects when present at concentrations at or below 1 μM

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