

**ANTHOCYANINS: BEYOND HUMAN NUTRITION
PRACTICAL APPLICATIONS OF ANTHOCYANINS**

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Abstract

Anthocyanins belong to a parent class of molecules called flavonoids under the subgroup of secondary metabolites. They are glycosylated polyphenolic compounds with a range of colours varying from orange, red and purple to blue in flowers, seeds, fruits and vegetative tissues. Anthocyanins play an important role in facilitating plant reproduction as they attract pollinators and seed dispersers by imparting bright colours etc. Anthocyanins produce even more variety of stable colours due to copigmentation. Copigmentation is a solution phenomenon in which pigments and other noncolored organic components form molecular associations or complexes. It generally results in an enhancement in the absorbance and in some cases, a shift in the wavelength of the maximum absorbance of the pigment. Colouration due to copigmentation of anthocyanins with other noncolored organic components have been realized in increasing colour of wines and grapes. This phenomenon can be utilized in many other uses of anthocyanins. In addition to their colourful characteristics, anthocyanins protect plants from several biotic and abiotic stresses, which may provide them a better adaptation to climate change. Anthocyanins have many health benefits. Due to anti-oxidant and anti-inflammatory properties, anthocyanin can prevent many deadly diseases such as cardio-vascular disease (CVD), cancer, diabetes, obesity etc. A brief description of the versatile uses of anthocyanins are presented in this article.

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Anthocyanins belong to a parent class of molecules called flavonoids under the subgroup of secondary metabolites. They are glycosylated polyphenolic compounds with a range of colours varying from orange, red and purple to blue in flowers, seeds, fruits and vegetative tissues etc. (Tanaka and Ohmiya, 2008). Anthocyanins synthesized *via* the phenyl propanoid pathway. They are water- soluble vacuolar pigments, odourless, nearly flavourless and moderately astringent. Biochemically, anthocyanins are mostly 3- glycosides or acylated form of anthocyanidins.

Anthocyanins play an important role in facilitating plant reproduction as they attract pollinators and seed dispersers by imparting bright colours (Harborne and Williams, 2000; Hoballah *et al.*, 2007). Anthocyanins produces even more variety of stable colours due to copigmentation. Copigmentation is a solution phenomenon in which pigments and other noncolored organic components form molecular associations or complexes. It generally results in an enhancement in the absorbance and in some cases, a shift in the wavelength of the maximum absorbance of the pigment. Colouration due to copigmentation of anthocyanins with other noncolored organic components have been realized in increasing colour of wines and grapes (Boulton, 2001). This phenomenon can be utilized in many other uses of anthocyanins. In addition to their colourful characteristics, anthocyanins protect plants from several biotic and abiotic stresses (Chalker-Scott, 1999; Ahmed *et al.*, 2014), which may provide them a better adaptation to climate change.

Anthocyanins have many health benefits. Due to anti-oxidant and anti-inflammatory properties, anthocyanin can prevent many deadly diseases such as cardiovascular disease (CVD), cancer, diabetes, obesity etc. It also improves vision power and having anti-microbial effect. Anthocyanin inhibited platelet aggregation (Rechner *et al.*, 2005) and improved lipid profile and platelet function in healthy volunteers (Alvarez-Suarez *et al.*, 2014). Improved visual function in patients with normal tension glaucoma (Shim *et al.*, 2015). It also prevented impairment of photoreceptor cell function during retinal inflammation (Miyake *et al.*, 2012). Anthocyanin prevented retinal degeneration induced by N-methyl-N-nitrosourea (Paik *et al.*, 2012). It enhances antioxidant capacity, and prevented insulin resistance in human subjects with type 2 diabetes (Li *et al.*, 2015). Estimated daily intake of anthocyanin is 12.5 mg/d in the United States of America. Apart from various health benefits, there is the other aspect of anthocyanins use which makes them more important. This is related to industrial uses of anthocyanins. A brief description of the versatile uses of anthocyanins are presented below:

As an Intelligent Colour Indicator Packaging

Smart packaging provides an opportunity to judge the status of packed commodity from the package itself. Recently a type of smart packaging, which can show different colour in different pH according to the state of the food and quality or freshness of food is strongly increasing. The pH-responsive colour-changing function of anthocyanins is useful for making colour indicator smart packaging films and have been widely used for the production of the colour indicator films for various types of food packaging applications. Anthocyanins show different structure and colour in different pH. It exhibit higher stability under acidic condition due to the formation of flavylium cation. Generally, anthocyanins are reddish in acidic condition, pink in neutral condition and blue in basic condition. At low pH

anthocyanins exhibited carbinol, pseudobase and quinoidal structure and quinoidal structure at pH 7-8 and then transform into chalcone structure.

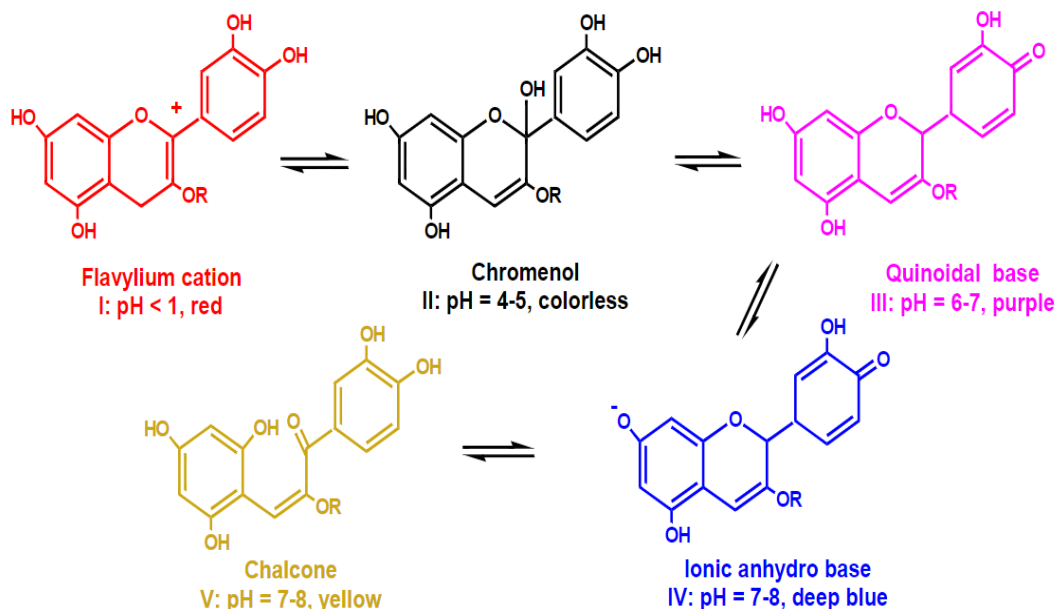


Fig.1: Structure of anthocyanin at different pH

Recent studies shows eco-friendly biodegradable polymer-based colour indicator films incorporated with anthocyanins. Biopolymers are mainly used rather than synthetic plastics as solid supports of anthocyanins because biopolymers are biodegradable, biocompatible, nontoxic and environmentally friendly (Kumar *et al.*, 2020). The colour indicator films could be very beneficial for the evaluation of the safety and quality of the packaged food at the spot. Besides, this type of natural pigment added packaging films also exhibit potent antioxidant activity, which can be useful to prevent colour change or lipid oxidation of packaged food products. The intelligent pH-responsive packaging films have a bright future in the food packaging industry to ensure food product quality and safety (Roy *et al.*, 2020).

As Renewable Hair Dyes from Black Currant Fruit Waste

Hair-dying industry is now becoming more popular and highly economic industry. But, some of these dyes may be harmful to humans and environment. Recently, Rose *et al.*, (2018), developed a natural, non-toxic hair dye from black-currant skins which is usually discarded with juice. The team used these pigments in a dye paste and applied it to bleached human hair, producing vivid colour and they could produce reds and violets by modifying the formulation. So, they concluded that anthocyanin based blackcurrant dyes

are comparable to conventional colourant and used to produce a variety of colour and shades and sustain colour after 12 shampoos.

As Renewable Textile Dyes from Blackcurrant Fruit Waste

Silk and wool fabrics were dyed using the purified black currant extract containing anthocyanins dye. Anthocyanin dyes are sustainable, bio-degradable, skin friendly and have many more health benefits. They generally showed pink colour without addition of metals. Dyeing in combination with aluminum sulphate provided deeper colours- with range of purples when applied pH 2.2 to 7, and blues at pH 9 and 11., attributable to formation of stabilized quinonoidal base AL^{3+} . Dyeing in combination with iron acetate produced brown and green shades due to the formation of anthocyanin- Fe^{3+} complex. After washing, the majority of silk and wool samples were rated grade 5, exhibiting no difference between washed and unwashed sample. Thus the best results were obtained when wool and silk were dyed with the complex of anthocyanin dyes and metals.

As Food Colourant

Xue and coworker (2019) reported that when red-fleshed apple anthocyanins were co-pigmented with caffeic acid and the copigmented complexes were encapsulated by gum arabic and maltodextrin using spray and freeze drying. Then stability of anthocyanin was increased used in food and pharmaceutical industries as value-added natural food pigments (Tidder *et al.*, 2018).

As Visual Markers

Anthocyanin fluoresce, enabling a tool for plant cell research to allow live cell imaging without a requirement for other fluorophores. Anthocyanin production may be engineered into genetically-modified materials to enable their identification visually (Kovinich *et al.*, 2012). An mutant allele of the transcription factor gene MYB10 from apple induces anthocyanin production throughout the plant. This gene were transformed into strawberry, apple plants to determine whether it could be used as a visible selectable marker for plant transformation as an alternative to chemically selectable markers. After transformation, red coloured calli, red shoots and red well growing plants were scored indicating the potential of anthocyanin marker (Kortstee *et al.*, 2011).

As Photosensitizer

A dye-sensitized solar cell (DSSC) is a device used for the conversion of visible light power into electricity. These cells are under research works due to their appealing features such as low production costs. The production cost and energy conversion efficiency of DSSCs is strongly influenced by the types of dyes used to harvest photons. Photosensitizer is a very important component for this solar cell and anthocyanin used as photosensitizer in it. Anthocyanin absorb more amount of light and show better photo-voltaic performances and convert light energy to electrical energy. Thus mixture of anthocyanin pigments with different absorption characteristics would give a synergistic effect to increase light absorption in different regions as different colours promote light harvesting in different wavelengths and have particular quantity injection of electrons

leading to increase in photocurrent and photo voltage. Thus, anthocyanins have the potential to be used as environmental friendly and economic photosensitizer.

It is very clear from the above discussion that anthocyanins have wide range of usage, which should utilize at their highest level.

References

- An, J., Zhang, X., Bi, S., You, C., Wang, X., & Hao, Y. (2019). The ERF transcription factor MdERF38 promotes drought stress-induced anthocyanin biosynthesis in apple. *The Plant Journal*. doi:10.1111/tpj.14555
- Appelham, I., Wulff-Vester, A. K., Wendell, M., Hvoslef-Eide, A.-K., Russell, J., Oertel, A., ... Matros, A. (2018). *Colour bio-factories: Towards scale-up production of anthocyanins in plant cell cultures. Metabolic Engineering*, 48: 218–232.
- Bai, S., Tao, R., Yin, L., Ni, J., Yang, Q., Yan, X., ... Teng, Y. (2019). Two B-box proteins, Pp BBX 18 and Pp BBX 21, antagonistically regulate anthocyanin biosynthesis via competitive association with Pp HY 5 in the peel of pear fruit. *The Plant Journal*. doi:10.1111/tpj.14510
- Boulton, R. 2001. The copigmentation of anthocyanins and its role in the color of red wine: A critical review. *American Journal of Enology and Viticulture* . 52: 67-87.
- Chen, L., Tian, J., Wang, S., Song, T., Zhang, J., & Yao, Y. (2019). Application of melatonin promotes anthocyanin accumulation in crabapple leaves. *Plant Physiology and Biochemistry*.doi:10.1016/j.plaphy.2019.07.024.
- Das, P. K., Shin, D. H., Choi, S.-B., & Park, Y.-I. (2012). Sugar-hormone cross-talk in anthocyanin biosynthesis. *Molecules and Cells*,34(6): 501–507. doi:10.1007/s10059-012-0151-x.
- Deng, Q., Xia, H., Lin, L., Wang, J., Yuan, L., Li, K., ... Liang, D. (2019). SUNRED, a natural extract-based biostimulant, application stimulates anthocyanin production in the skins of grapes. *Scientific Reports*, 9(1).
- Farias-Cervantes, V. S., Chávez-Rodríguez, A., García-Salcedo, P. A., García-López, P. M., Casas-Solís, J., & Andrade-González, I. (2018). Antimicrobial effect and in vitro release of anthocyanins from berries and Roselle obtained via microencapsulation by spray drying. *Journal of Food Processing and Preservation*, e13713.
- Gaiotti, F., Pastore, C., Filippetti, I., Lovat, L., Belfiore, N., & Tomasi, D. (2018). Low night temperature at veraison enhances the accumulation of anthocyanins in Corvina grapes (*Vitis Vinifera L.*). *Scientific Reports*, 8(1).
- Gao-Takai, M., Katayama-Ikegami, A., Matsuda, K., Shindo, H., Uemae, S., & Oyaizu, M. (2019). A low temperature promotes anthocyanin biosynthesis but does not accelerate endogenous abscisic acid accumulation in red-skinned grapes. *Plant Science*.doi:10.1016/j.plantsci.2019.01.015.
- García-Pastor, M. E., Serrano, M., Guillén, F., Giménez, M. J., Martínez-Romero, D., Valero, D., & Zapata, P. J. (2019). Preharvest application of methyl jasmonate increases crop yield, fruit quality and bioactive compounds in pomegranate "Mollar de Elche" at harvest and during postharvest storage. *Journal of the Science of Food and Agriculture*.

- Gu, K.-D., Wang, C.-K., Hu, D.-G., & Hao, Y.-J. (2019). How do anthocyanins paint our horticultural products? *Scientia Horticulturae*, 249: 257–262. doi:10.1016/j.scienta.2019.01.034.
- Hu, B., Lai, B., Wang, D., Li, J., Chen, L., Qin, Y., ... Zhao, J. (2018). Three LcABFs are involved in the regulation of chlorophyll degradation and anthocyanin biosynthesis during fruit ripening in *Litchi chinensis*. *Plant and Cell Physiology*. doi:10.1093/pcp/pcy219.
- Jaakola, L. (2013). New insights into the regulation of anthocyanin biosynthesis in fruits. *Trends in Plant Science*, 18(9): 477–483.
- Jeong, S., Goto-Yamamoto, N., Kobayashi, S., & Esaka, M. (2004). Effects of plant hormones and shading on the accumulation of anthocyanins and the expression of anthocyanin biosynthetic genes in grape berry skins. *Plant Science*, 167(2): 247–252.
- Jezek, M., Zörb, C., Merkt, N., & Geilfus, C.-M. (2018). Anthocyanin Management in Fruits by Fertilization. *Journal of Agricultural and Food Chemistry*, 66(4): 753–764.
- Jia, H., Wang, S., Lin, H., Satio, T., Ampa, K., Todoroki, Y., & Kondo, S. (2017). Effects of abscisic acid agonist or antagonist applications on aroma volatiles and anthocyanin biosynthesis in grape berries. *The Journal of Horticultural Science and Biotechnology*, 93(4): 392–399.
- Khoo, H. E., Azlan, A., Tang, S. T., & Lim, S. M. (2017). Anthocyanidins and anthocyanins: colored pigments as food, pharmaceutical ingredients, and the potential health benefits. *Food & Nutrition Research*, 61(1): 1361779.
- Kovinich, N., Saleem, A., and Rintoul, T.L. (2012). Coloring genetically modified soyabean grains with anthocyanins by suppression of the proanthocyanidin genes ANR1 and ANR2. *Transgenic Research*, 21(4): 757-771.
- Landi, M., Tattini, M., & Gould, K. S. (2015). *Multiple functional roles of anthocyanins in plant-environment interactions*. *Environmental and Experimental Botany*, 119: 4–17.
- Li, D., Wang, P., Luo, Y., Zhao, M., & Chen, F. (2015). Health benefits of anthocyanins and molecular mechanisms: Update from recent decade. *Critical Reviews in Food Science and Nutrition*, 57(8): 1729–1741.
- Li, D., Zhang, X., Xu, Y., Li, L., Soleimani Aghdam, M., & Luo, Z. (2019). Effect of exogenous sucrose on anthocyanin synthesis in postharvest strawberry fruit. *Food Chemistry*.
- Li, G., Zhao, J., Qin, B., Yin, Y., An, W., Mu, Z., & Cao, Y. (2019). ABA mediates development-dependent anthocyanin biosynthesis and fruit coloration in *Lycium* plants. *BMC Plant Biology*, 19(1).
- Liu, T., Song, S., Yuan, Y., Wu, D., Chen, M., Sun, Q., ... Chen, K. (2015). Improved peach peel color development by fruit bagging. Enhanced expression of anthocyanin biosynthetic and regulatory genes using white non-woven polypropylene as replacement for yellow paper. *Scientia Horticulturae*, 184: 142–148.
- Man, Y.-P., Wang, Y.-C., Li, Z.-Z., Jiang, Z.-W., Yang, H.-L., Gong, J.-J., ... Wang, Z.-Y. (2014). High-temperature inhibition of biosynthesis and transportation of

- anthocyanins results in the poor red coloration in red-fleshed *Actinidia chinensis*. *Physiologia Plantarum*, 153(4): 565–583.
- Moro, L., Hassimotto, N. M. A., & Purgatto, E. (2017). Postharvest Auxin and Methyl Jasmonate Effect on Anthocyanin Biosynthesis in Red Raspberry (*Rubus idaeus* L.). *Journal of Plant Growth Regulation*, 36(3): 773–782.
- Niu, S.-S., Xu, C.-J., Zhang, W.-S., Zhang, B., Li, X., Lin-Wang, K., ... Chen, K.-S. (2010). Coordinated regulation of anthocyanin biosynthesis in Chinese bayberry (*Myrica rubra*) fruit by a R2R3 MYB transcription factor. *Planta*, 231(4): 887–899.
- Rahim, M. A., Busatto, N., & Trainotti, L. (2014). Regulation of anthocyanin biosynthesis in peach fruits. *Planta*, 240(5): 913–929.
- Rose, P. M., Cantrill, V., Benohoud, M., Tidder, A., Rayner, C. M., & Blackburn, R. S. (2018). Application of Anthocyanins from Blackcurrant (*Ribes nigrum* L.) Fruit Waste as Renewable Hair Dyes. *Journal of Agricultural and Food Chemistry*, 66(26): 6790–6798.
- Routray, W., & Orsat, V. (2011). *Blueberries and Their Anthocyanins: Factors Affecting Biosynthesis and Properties. Comprehensive Reviews in Food Science and Food Safety*, 10(6): 303–320.
- Shi, L., Cao, S., Chen, W., & Yang, Z. (2014). Blue light induced anthocyanin accumulation and expression of associated genes in Chinese bayberry fruit. *Scientia Horticulturae*, 179: 98–102.
- Tai, D., Tian, J., Zhang, J., Song, T., & Yao, Y. (2014). A *Malus* Crabapple Chalcone Synthase Gene, *McCHS*, Regulates Red Petal Color and Flavonoid Biosynthesis. *PLoS ONE*, 9(10): e110570.
- Takos, A. M., Jaffe, F. W., Jacob, S. R., Bogs, J., Robinson, S. P., & Walker, A. R. (2006). Light-Induced Expression of a MYB Gene Regulates Anthocyanin Biosynthesis in Red Apples. *Plant physiology*, 142(3): 1216–1232.
- Wang, N., Zhang, Z., Jiang, S., Xu, H., Wang, Y., Feng, S., & Chen, X. (2016). Synergistic effects of light and temperature on anthocyanin biosynthesis in callus cultures of red-fleshed apple (*Malus sieversii* f. *niedzwetzkyana*). *Plant Cell, Tissue and Organ Culture (PCTOC)*, 127(1): 217–227.
- Wang, X.-F., An, J.-P., Liu, X., Su, L., You, C., & Hao, Y.-J. (2018). The nitrate-responsive protein MdBT2 regulates anthocyanin biosynthesis by interacting with the MdMYB1 transcription factor. *Plant Physiology*, pp.00244.2018. doi:10.1104/pp.18.00244.
- Wang, Y., Wang, N., Xu, H., Jiang, S., Fang, H., Su, M., ... Chen, X. (2018). Auxin regulates anthocyanin biosynthesis through the Aux/IAA–ARF signaling pathway in apple. *Horticulture Research*, 5(1).
- Xie, L., Su, H., Sun, C., Zheng, X., & Chen, W. (2018). Recent advances in understanding the anti-obesity activity of anthocyanins and their biosynthesis in microorganisms. *Trends in Food Science & Technology*, 72: 13–24.
- Xue, J., Su, F., Meng, Y., & Yurong, G. (2018). Enhanced Stability of Red-Fleshed Apple Anthocyanins by Copigmentation and Encapsulation. *Journal of the Science of Food and Agriculture*. doi:10.1002/jsfa.9555
- Yang, B., He, S., Liu, Y., Liu, B., Ju, Y., Kang, D., ... Fang, Y. (2020). Transcriptomics Integrated with Metabolomics Reveals the Effect of Regulated Deficit Irrigation on

- Anthocyanin Biosynthesis in Cabernet Sauvignon Grape Berries. *Food Chemistry*, 126170.
- Yang, J., Li, B., Shi, W., Gong, Z., Chen, L., & Hou, Z. (2018). Transcriptional activation of anthocyanin biosynthesis in developing fruit of blueberries (*Vaccinium corymbosum* L.) by preharvest and postharvest UV irradiation. *Journal of Agricultural and Food Chemistry*.
- Yousuf, B., Gul, K., Wani, A. A., & Singh, P. (2015). *Health Benefits of Anthocyanins and Their Encapsulation for Potential Use in Food Systems: A Review. Critical Reviews in Food Science and Nutrition*, 56(13), 2223–2230. doi:10.1080/10408398.2013.805316
- Zhang, Y., Butelli, E., & Martin, C. (2014). Engineering anthocyanin biosynthesis in plants. *Current Opinion in Plant Biology*, 19: 81–90.
- Zhang, Y., Jiang, L., Li, Y., Chen, Q., Ye, Y., Zhang, Y., ... Tang, H. (2018). Effect of Red and Blue Light on Anthocyanin Accumulation and Differential Gene Expression in Strawberry (*Fragaria × ananassa*). *Molecules*,23(4): 820.