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Comparative estimation of anthocyanidin pigments in seven grape genotypes through UV–Vis spectroscopy

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ABSTRACT

Anthocyanidins, the sugar-free forms of anthocyanins, are key pigments responsible for red-to-blue coloration and antioxidant properties in fruits. Grape (*Vitis vinifera* L.) pomace, a major by-product of wine production, consists largely of skin, seeds, and pulp residues, with the peel being a rich source of anthocyanidin pigments. These compounds play an important role in determining fruit colour and antioxidant activity in fruits. Therefore, this study was undertaken to analyse the anthocyanidin composition of the peel of seven grape genotypes. The experiment was conducted at KITS, Coimbatore, using a randomised block design with seven treatments, each replicated thrice. Significant variation in anthocyanidin content was observed among the studied genotypes. Manjari Medika recorded the highest cyanidin-3-glucoside (366.17 mg/L), delphinidin-3-glucoside (364.5 mg/L), and peonidin-3-glucoside (622.24 mg/L), while H-516 exhibited maximum malvidin-3-glucoside (566.29 mg/L), pelargonidin-3-glucoside (459.69 mg/L), and petunidin-3-glucoside (325.96 mg/L). These findings indicate genotype-dependent variation in pigment composition. This study demonstrates the potential of grape peel as a natural source of anthocyanidin pigments for applications in food processing and nutraceutical industries.

Key words: *Vitis Vinifera*, antioxidant property, gut health, bioavailability, natural food ingredients, plant waste utilization.

INTRODUCTION

Grape (*Vitis vinifera* L.) was one of the most important cash fruit crops in the world, belonging to the *Vitis* genus and *Vitaceae* family. In FY2024, India cultivated about 179.62 thousand hectares (MoA & FW). According to DGCIS, the grape export volume for FY2025 is around 271,259.89 metric tons, or 353.54 USD million. One of the most widely consumed worldwide, grapes may be eaten both fresh (as table grapes) and processed (as wine, grape juice, molasses, and raisins) (Patil *et al*., 18). Grapes include many minerals, vitamins, antioxidants, and phenolic compounds that are essential for human health. These compounds' anti-ageing, anti-bacterial, and anti-inflammatory properties provide protection against cardiovascular diseases (Akram *et al*., 1).

Anthocyanins constitute an important subgroup of phenolic phytochemicals responsible for the red, purple, and blue pigmentation in many fruits, vegetables and flowers. Structurally, anthocyanins are glycosylated derivatives of anthocyanidins, in which one or more sugar moieties are commonly attached at the C-3 position of the C-ring and occasionally at other positions. The sugar-free forms are referred to as anthocyanidins (aglycones), whereas the corresponding glycosylated forms are known as anthocyanins (Khoo *et al.,* 12). Glycosylation enhances the stability, solubility, and biological activity of these compounds, contributing to their diverse physiological and health-promoting functions (Taiz and Zeiger, 21). The production of anthocyanins comes from the phenylalanine route, and anthocyanidin is produced when sugar molecules are removed. Anthocyanin is considered a flavonoid even though it has a positive charge at the oxygen atom of the C-ring of the basic flavonoid structure (Laleh *et al*., 16). They are classified into Malvidin, delphinidin, pelargonidin, petunidin, peonidin, and cyanidin based on the number and position of hydroxyl (–OH) and methoxy (–OCH_3) groups on the B-ring of the flavonoid skeleton (Khoo *et al*., 12). They are responsible for most of the red, purple, and blue flowers. In addition to their traditional use, red, blue, and purple fruits are frequently eaten for their healthful properties. Strong antioxidants are found in the anthocyanin pigments of berries, blackcurrants, and other red to blue-coloured fruits (Khoo *et al*., 12).

Approximately three-fourths of the world's cultivated grapes are utilized for wine production. The by-products generated during the winemaking process, including skins, pulp, seeds, and stems, collectively constitute grape pomace, which is often

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regarded as an agro-industrial waste. However, grape pomace is a rich source of bioactive compounds, particularly phenolics, flavonoids, anthocyanins, proanthocyanidins, and stilbenes, which are predominantly concentrated in the grape skin. These phytochemicals possess strong antioxidant properties and have attracted considerable interest as natural food colorants and functional ingredients (Serea *et al.,* 19). Therefore, the present study aimed to characterize and evaluate the anthocyanidin composition of the peel tissues of seven wine grape genotypes.

MATERIALS AND METHODS

This experiment included seven different grape genotypes which were collected in and around the regions of Then Palani and Odaipatti in the Theni district of Tamil Nadu, as well as from the Grape Research Station (GRS), Theni. Freshly harvested grape samples were collected, and the peels were removed from the berries for further analysis. The varieties included Madhu Angoor, Manjari Medika, Krishna Seedless, Jyothi, Red Globe, Muscat, and H-516. From each variety, three bunches were collected. From each bunch, berries were randomly selected, and the peels were carefully separated from the selected berries as per the standard procedure. The study was conducted at Karunya Institute of Technology and Sciences. The experimental data were collected as per the standard procedure and analysed using a Randomized Block Design (RBD) with three replications. For the analysis of grape peel samples, a microprocessor UV-Vis double-beam spectrophotometer (Model: KLUV-2100) was used.

Determination of anthocyanidin content in grape peel was done using the pH-differential method (Giusti and Wrolstad, 8). The materials used were 0.025 M of potassium chloride buffer, pH 1.0; 0.4 M of sodium acetate buffer, pH 4.5; and 1 g of the sample and solvent (5 ml) based on the compounds listed in Table 1. From that extract, take 0.2 ml and make it up to 3 ml using the potassium chloride and sodium acetate buffer (if the dilution factor (DF) = 15, then take 0.2 ml of extract and make it 3 ml). Make up two different dilution buffers separately in each test tube. Then, perform UV-Vis spectrophotometry and allow it to calibrate for 30 mins. Blanking is done with a UV-Vis spectrophotometer using distilled water at the wavelength listed in Table 2. Then, measure the absorbance of each dilution buffer at $A_{vis\text{-}max}$ and at A_{700} nm against the blank (1). After that, calculate the monomeric anthocyanidin pigment concentration in the grape peel using the below-mentioned formula (2).

$$\text{Absorbance (A)} = (A_{vis\text{-}max} - A_{700})\ pH_{1.0} - (A_{vis\text{-}max} - A_{700})\ pH_{4.5} \ldots\ldots\ (1)$$

$$\text{Monomeric anthocyanin pigment (mg/litre)} = (A \times MW \times DF \times 1000)/\ (\varepsilon \times 1) \ldots\ldots\ (2)$$

A – absorbance from equation (1).

MW- Molecular Weight

DF- Dilution Factor

ε - Molar absorptivity.

Analysis of Variance (ANOVA) was performed on the collected data for different anthocyanidin compounds, which were analysed using a Randomised Complete Block Design with seven treatments that were replicated thrice. The OPSTAT software suite and Microsoft Excel were used to analyse the collected data. The significance level was at $P \leq 0.01$.

Table 1: Solvents used for the analysis of different anthocyanidin compounds.

S. No.	Anthocyanidin	Solvent	References
1.	Cyd-3-glu, Dpd-3-glu	1% HCl in methanol	(Siegelman and Hendricks, 20; Asen *et al*., 2)
2.	Mvd-3-glu, Pg-3-glu	0.1% HCl in methanol	(Koeppen and Basson, 13; Giusti *et al*., 7)
3.	Pnd-3-glu, Ptd-3-glu	5% of 1N HCl in methanol	(Heredia *et al*., 9)

Table 2: Wavelength, molecular weight and molar absorptivity of different anthocyanidin compounds.

S. No.	Anthocyanidin	Wavelength (nm)	Molecular weight (MW)	Molar absorptivity (ε)	References
1.	Cyanidin-3-glucoside	530	449.2	34300	(Siegelman and Hendricks, 20)
2.	Delphinidin-3-glucoside	543	465.2	29000	(Asen *et al*., 2)
3.	Malvidin-3-glucoside	538	493.2	29500	(Koeppen and Basson, 13)
4.	Pelargonidin-3-glucoside	508	433.2	17330	(Giusti *et al*., 7)
5.	Peonidin-3-glucoside	536	463.2	11300	(Heredia *et al*., 9)
6.	Petunidin-3-glucoside	546	479.2	12900	(Heredia *et al*., 9)

RESULTS AND DISCUSSION

Various anthocyanidin compounds present in grape peel were analysed in the seven grape genotypes. The results revealed a significant difference in the anthocyanidin compounds among the genotypes, which are classified based on the number and position of hydroxyl (–OH) and methoxy ($–OCH_3$) groups on the B-ring of the flavonoid skeleton. Based on Table 3, Manjari Medika had the highest cyanidin-3-glucoside (366.17 mg/L), followed by H-516 (255.96 mg/L) and Jyothi (52.84 mg/L). At 18.66 mg/L, Red Globe has the lowest compound of cyanidin-3-glucoside. This value indicates a high amount of reddish-purple pigment present in Manjari Medika and lower in the Red Globe variety of grapes (Khoo *et al*., 12). These compounds can cure gingival fibroblasts and oral epithelial cells, which counteracts nicotine's harmful effects (Desjardins *et al*., 5). It could reduce the generation of ROS in the mitochondrion, enhance the viability of β-cells, and prevent their death. Additionally, it increased the levels of insulin protein and the transcripts of the insulin-like growth factor II gene in the rat insulinoma cell line INS-1 (Bartel *et al*., 3). After 15 weeks of cyanidin-3-glucoside treatment, diastolic dysfunction and cardiac hypertrophy are prevented (Mattioli *et al.,* 17).

Delphinidin-3-glucoside is another important anthocyanidin, with the highest concentration in Manjari Medika (364.5 mg/L), followed by H-516 (234.84 mg/L), Muscat (43.07 mg/L), and Jyothi (43.07 mg/L). This compound is found less in Red Globe (15.87 mg/L). This is responsible for the blue-reddish or purple pigmentation which is rich in Manjari Medika and lower in Red Globe (Khoo *et al*., 12). It is effective against several molecular subtypes of established cancer cell lines (Iqbal *et al*., 11). Delphinidin showed anticancer efficacy in treating ovarian cancer with a poor prognosis. It is noticeable that delphinidin-3-O-glucoside has the greatest anticancer efficacy among the Delphinidin derivatives due to its rapid absorption and appearance in blood plasma within 15 minutes of oral dosing (Iqbal *et al*., 11).

The highest amount of malvidin-3-glucoside is found in H-516 (566.29 mg/L) and followed by Manjari Medika (509.87 mg/L) and Jyothi (125.6 mg/L). Malvidin-3-glucoside is lower in Red Globe. Purple and blue pigment is lower in Red Globe compared to other varieties and higher in H-516 (Khoo *et al*., 12). Malvidin-3-glucoside exhibited significant antioxidant properties in endothelial cells and functioned as a nutritional compound to avert oxidative stress-related disorders and protect cells from oxidative degradation (Huang et al., 10).

H-516 (459.69 mg/L) has the highest amount of pelargonidin-3-glucoside, followed by Manjari Medika (413.2 mg/L) and Jyothi (92.98 mg/L) (Table 4). The lesser amount of this compound is present in Madhu Angoor (23.62 mg/L). This compound expresses red- and orange-coloured pigments in the peel, which is higher in H-516 and lower in Madhu Angoor (Khoo *et al*., 12). Pelargonidin-3-glucoside possesses a variety of neuroprotective properties, including direct control of oxidative stress, suppression of inflammatory responses, and support for neural regeneration, all of which are interrelated. It greatly reduced neuropathic pain and enhanced motor function (Kooshki *et al*., 14).

Petunidin-3-glucoside is highly present in H-516 (325.96 mg/L), followed by Muscat (92.98 mg/L) and Jyothi (88.03 mg/L). The lowest amount of this compound is present in Krishna Seedless (3.39 mg/L). This compound is known for dark red or purple pigmentation in grapes, which is higher in H-516 and lower in Krishna seedless (Khoo *et al*., 12). It is a strong antioxidant that protects cells from damage caused by free radicals. It also has many properties

Table 3: Estimation of different anthocyanidin content present in peel of seven grape genotypes.

Anthocyanin (mg/L)	Cyanidin- 3- glucoside	Delphinidin- 3- glucoside	Malvidin- 3- glucoside
Krishna Seedless	37.32 ± 0.28	37.53 ± 0.43	63.33 ± 0.63
Madhu Angoor	21.41± 0.21	23.08 ± 0.28	25.54 ± 0.34
Manjari Medika	366.17 ± 3.67	364.5 ± 1.70	509.87 ± 2.07
Muscat	45.96 ± 0.26	43.07 ± 0.37	51.09 ± 0.39
Red Globe	18.66 ± 0.36	15.87 ± 0.47	24.48 ± 0.38
Jyothi	52.84 ± 0.34	43.07 ± 0.37	125.6 ± 1.60
H-516	255.96 ± 2.56	234.84 ± 1.44	566.29 ± 2.19
CD (0.01)	2.555	1.05	1.498
SE (d)	1.16	0.476	0.68

Table 4: Estimation of different anthocyanidin content present in peel of seven grape genotypes.

Anthocyanin (mg/L)	Pelargonidin- 3- glucoside	Petunidin- 3- glucoside	Peonidin- 3- glucoside
Krishna Seedless	47.61 ± 0.41	3.39 ± 0.09	47.95 ± 0.45
Madhu Angoor	23.62 ± 0.32	36.21 ± 0.31	54.1 ± 0.50
Manjari Medika	413.2 ± 1.80	44.29 ± 0.39	622.24 ± 2.24
Muscat	41.24 ± 0.34	96.95 ± 0.85	147.56 ± 1.36
Red Globe	24.74 ± 0.34	23.96 ± 0.36	56.56 ± 0.56
Jyothi	92.98 ± 0.88	88.03 ± 0.83	127.89 ± 1.29
H-516	459.69 ± 1.89	325.96 ± 1.46	463.6 ± 1.80
CD (0.01)	1.273	0.836	1.269
SE (d)	0.578	0.38	0.576

that may help it fight cancer, such as being an antioxidant, anti-inflammatory, antiproliferative, and apoptosis-inducing agent on cancer cells (Kowalczyk *et al*., 15).

Manjari Medika (622.24 mg/L) has the highest amount of peonidin-3-glucoside, followed by H-516 (463.6 mg/L) and Muscat (Table 4). A lesser amount of this compound is found in Madhu Angoor (54.1 mg/L). This compound represents the purplish-pink and red colour, which are higher in Manjari Medika and lower in Madhu Angoor (Khoo *et al*., 12). peonidin 3-glucoside against cell growth and induced apoptosis of human breast cancer (Chen *et al*., 4).

The results from these experiments suggest that a major amount of the pigment is higher in Manjari Medika and H-516. According to earlier reports on the relative bioavailability of red wine anthocyanidin, the glycosides of peonidin had the highest relative bioavailability, followed by those of cyanidin, malvidin, delphinidin, and petunidin (Frank *et al*., 6). Because of their limited bioavailability, anthocyanins are not as well absorbed into the bloodstream and are excreted in urine and faeces at high rates, which decreases their potential to scavenge free radicals (Khoo *et al*., 12). Berries' skin's anthocyanin concentration has decreased over the past several decades due to environmental factors, including temperature in wine-producing regions worldwide. Sometimes trunk girdling, leaf removal, cluster thinning and ABA application can also affect berry skin colouration (Teshigawara *et al*., 22).

The study concluded that anthocyanidin in the peel of seven grape varieties showed that Manjari Medika and H-516 had a higher amount of pigmentation that was comparable to the other varieties. Variation in anthocyanidin content among grape genotypes may be attributed to genetic differences regulating flavonoid biosynthesis, as well as environmental factors such as temperature and light exposure during berry development. These compounds found in the peel of grape berries help us quench ROS, thereby increasing the antioxidant properties of grapes. This pigmentation also can be used as an alternative to the artificial food colours and they have different nutraceutical and pharmacological benefits. This compound has angiogenic activity, which prevents the formation of new blood vessels that supply oxygen to cancer cells. The evaluation of other coloured varieties of grapes can be done to know their pigmentation, and furthermore, analysis can be done to study the pharmacological, nutraceutical and health benefits. The compound also has a greater scope in the food industry, where it mainly attracts customers.

AUTHOR'S CONTRIBUTION

Conceptualization of research (G); Designing of the experiments (UBC); Contribution of experimental materials (AO); Execution of experiments and data collection (TD); Analysis of data and interpretation (VTV); Preparation of manuscript and literature review (G,M); Supervision and critical review (BA,GRS).

DECLARATION

The authors affirm that they do not have any potential conflicts of interest.

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REFERENCES

1. Akram, M. T., Qadri, R., Khan, M. A., Hafiz, I. A., Nisar, N., Khan, M. M., Feroze, M. A. and Hussain, K. 2021. Morpho-phenological characterization of grape (*Vitis vinifera* L.) germplasm grown in northern zones of Punjab, Pakistan. *Pak. J. Agric. Sci.*, **58**(4): 1223-36.

2. Asen, S., Stuart, N. W. and Siegelman, H. W. 1959. Effect of various concentrations of nitrogen, phosphorus and potassium on sepal color of *Hydrangea macrophylla*. *Proc. Am. Soc. Hortic. Sci.*, **73**: 495–502.

3. Bartel, I., Koszarska, M., Strzałkowska, N., Tzvetkov, N. T., Wang, D., Horbańczuk, J. O., Wierzbicka, A., Atanasov, A. G. and Jóźwik, A. 2023. Cyanidin-3-O-glucoside as a nutrigenomic factor in type 2 diabetes and its prominent impact on health. *Int. J. Mol. Sci.*, **24**(11): 9765.

4. Chen, P. N., Chu, S. C., Chiou, H. L., Chiang, C. L., Yang, S. F. and Hsieh, Y. S. 2005. Cyanidin-3-glucoside and peonidin-3-glucoside inhibit tumor cell growth and induce apoptosis *in vitro* and suppress tumor growth *in vivo*. *Nutr. Cancer*, **53**(2): 232–43.

5. Desjardins, J., Tanabe, S., Bergeron, C., Gafner, S. and Grenier, D. 2012. Anthocyanin-rich black currant extract and cyanidin-3-O-glucoside have cytoprotective and anti-inflammatory properties. *J. Med. Food*, **15**(12): 1045–50.

6. Frank, T., Netzel, M., Strass, G., Bitsch, R. and Bitsch, I. 2003. Bioavailability of anthocyanidin-3-glucosides following consumption of red wine and red grape juice. *Can. J. Physiol. Pharmacol.*, **81**(5): 423–35.

7. Giusti, M. M., Rodriguez-Saona, L. E. and Wrolstad, R. E. 1999. Spectral characteristics, molar absorptivity and color of pelargonidin derivatives. *J. Agric. Food Chem.*, **47**: 4631–37.

8. Giusti, M. M. and Wrolstad, R. E. 2001. Characterization and measurement of anthocyanins by UV-visible spectroscopy. *Curr. Protoc. Food Anal. Chem.*, **1**: F12.

9. Heredia, F. J., Francia-Aricha, E. M., Rivas-Gonzalo, J. C., Vicario, I. M. and Santos-Buelga, C. 1998. Chromatic characterization of anthocyanins from red grapes. I. pH effect. *Food Chem.*, **63**: 491–98.

10. Huang, W., Zhu, Y., Li, C., Sui, Z. and Min, W. 2016. Effect of blueberry anthocyanins malvidin and glycosides on the antioxidant properties in endothelial cells. *Oxid. Med. Cell. Longev.*, **2016**: 1591803.

11. Iqbal, S., Omara, T., Kahwa, I. and Khan, U. M. 2024. Anticancer potential of delphinidin and its derivatives: therapeutic and mechanistic insights. *Med. Chem. Res.*, **33**(10): 1769–86.

12. Khoo, H. E., Azlan, A., Tang, S. T. and Lim, S. M. 2017. Anthocyanidins and anthocyanins: coloured pigments as food, pharmaceutical ingredients, and the potential health benefits. *Food Nutr. Res.*, **61**(1): 1361779.

13. Koeppen, B. H. and Basson, D. S. 1966. The anthocyanin pigments of Barlinka grapes. *Phytochemistry*, **5**: 183–87.

14. Kooshki, L., Fakhri, S., Abbaszadeh, F., Kiani, A., Farzaei, M. H., Mohammadi-Noori, E. 2025. Pelargonidin improves functional recovery and attenuates neuropathic pain following spinal cord injury in rats: relevance to its neuroprotective, antioxidant, and anti-inflammatory effects. *Front. Pharmacol.*, **16**: 1547187.

15. Kowalczyk, T., Muskała, M., Merecz-Sadowska, A., Sikora, J., Picot, L. and Sitarek, P. 2024. Anti-inflammatory and anticancer effects of anthocyanins in *in vitro* and *in vivo* studies. *Antioxidants*, **13**(9): 1143.

16. Laleh, G., Frydoonfar, H., Heidary, R., Jameei, R. and Zare, S. 2006. The effect of light, temperature, pH and species on stability of anthocyanin pigments in four *Berberis* species. *Pak. J. Nutr.*, **5**(1): 90–92.

17. Mattioli, R., Francioso, A., Mosca, L. and Silva, P. 2020. Anthocyanins: a comprehensive review of their chemical properties and health effects on cardiovascular and neurodegenerative diseases. *Molecules*, **25**(17): 3809.

18. Patil, P. N., Al-Aamri, A. A., Al-Rubkhi, Z. A. and Al Abri, A. S. 2018. Evaluation of nutritional fact and study of various physico-chemical parameters of black, green and red grapes samples in the local fruit market. *Asian J Chem. Sci.* **8** (3):1-8.

19. Serea, D., Constantin, O. E., Horincar, G., Stănciuc, N., Aprodu, I., Bahrim, G. E. and Râpeanu. 2023. Optimization of extraction parameters of anthocyanin compounds and antioxidant properties from red grape (*Băbească neagră*) peels. *Inventions*, **8**(2): 59.

20. Siegelman, H. W. and Hendricks, S. B. 1958. Photocontrol of alcohol, aldehyde and anthocyanin production in apple skin. *Plant Physiol.*, **33**: 409–13.

21. Taiz, L. and Zeiger, E. 2010. *Plant Physiology*. 5th ed. Sunderland (MA): Sinauer Associates.

22. Teshigawara, Y., Sato, S., Asada, T., Nojiri, M., Suzuki, S. and Aoki, Y. 2024. Anthocyanin accumulation in grape berry skin promoted by endophytic *Microbacterium* sp. che218 isolated from wine grape shoot xylem. *Microorganisms*, **12**(9): 1906.

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