Journal of Environmental Biology
p-ISSN: 0254-8704 • e-ISSN: 2394-0379 • CODEN: JEBIDP

JEB®

Original Research Paper
Volume 47 Issue 2 March 2026 pp. 254-262
DOI: http://doi.org/10.22438/jeb/47/2/MRN-5709

J. Environ. Biol., 2026

Journal website : www.jeb.co.in ★ E-mail : editor@jeb.co.in

Optimization of extraction and stability characterization of rose petal natural dye: A standardized approach

S. Keerthana[1], B. Singh[1]*, R. Jain[1], M.K. Singh[1], A.K. Tiwari[1], A. Joshi[2] and R. Bansal[3]

[1]*Division of Floriculture and Landscaping, ICAR-Indian Agricultural Research Institute, New Delhi-110 012, India*
[2]*Division of Food Science and Post Harvest Technology, ICAR-Indian Agricultural Research Institute, New Delhi-110 012, India*
[3]*Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110 012, India*

Received: 05 March 2025 **Revised:** 15 May 2025 **Accepted:** 15 October 2025

***Corresponding Author Email:** bflori17feb@gmail.com ***ORCiD:** https://orcid.org/0009-0003-5633-2696

Abstract

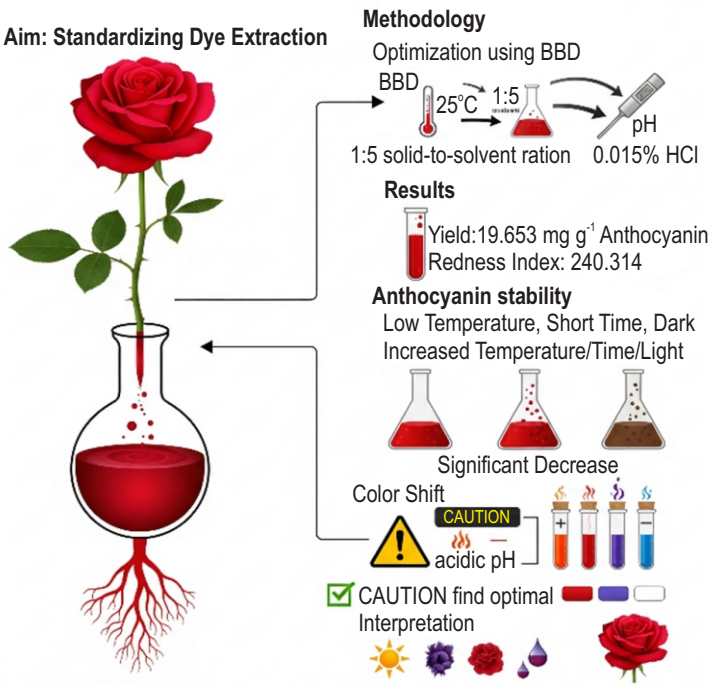
Aim: Standardization of natural dye extraction process from the petals of rose flower for maximum yield of natural dye.

Methodology: Optimization of natural dye extraction process was conducted using Box Behnken Design of Response Surface Methodology followed by stability studies to study the effect of different factors involved in the degradation of extracted anthocyanin.

Results: Optimal anthocyanin extraction from rose petals, yielded 19.653 mg g^{-1} with a high redness index of 240.314, was achieved at 25°C using a 1:5 solid-to-solvent ratio and 0.015% HCl. Anthocyanin stability decreased exponentially with increased temperature and time, with natural light exposure causing the most significant degradation. The dyes color shifted from bright red in acidic condition to blue in alkaline conditions.

Interpretation: The study successfully identified optimal conditions for extracting vibrant red anthocyanin dye from roses. However, the sensitivity of dye to heat, light, and pH variations indicate a need for careful handling and storage to maintain its colour and stability.

Key words: Anthocyanin, Natural dye, Optimization, Rose flower petals, Stability



How to cite: Keerthana, S., B. Singh, R. Jain, M.K. Singh, A.K. Tiwari, A. Joshi and R. Bansal: Optimization of extraction and stability characterization of rose petal natural dye: A standardized approach. *J. Environ. Biol.*, **47,** 254-262 (2026).

Introduction

Roses are a valuable natural resource, appreciated both for their aesthetic and practical uses. They serve as popular cut flowers, religious offerings, and garden decorations. The red hues, derived from anthocyanins, also provide antibacterial benefits (Saati *et al.,* 2018). Far from being solely decorative, rose blossoms hold considerable industrial value. Products like rose water, rose oil, rose concrete, dried petals, dried buds, and rose absolute are extracted from roses and are essential components in the global cosmetic, perfume, food, and pharmaceutical sectors (Mallick *et al.,* 2024). Economically significant, rose petals yield valuable compounds like vitamin C, used in natural dyes, biopolymers, and products such as Pankhuri (dried petals), Gul-e-roghan (rose essence), and Gulkand (rose petal jam) (Najem *et al.,* 2011). As a major global commodity, with key producers like India, Bulgaria, Iran and Turkey, roses face economic hurdles due to short lifespan of cut flowers, limiting farmer profits. Increasing shelf life or finding alternative uses for less-than-perfect blooms is vital for economic optimization (Leghari *et al.,* 2016).

This necessitates exploring alternative value-added applications for roses, particularly for less-than-perfect blooms that would otherwise go to waste. One such promising avenue is the extraction of natural dyes. A part from beauty and fragrance, rose petals have different applications and are a source of natural red-to-yellow dye (Salman *et al.,* 2023). Natural dyes offer a gentler, more aesthetically pleasing colour palette, characterized by soft, lustrous, and subtle hues, creating a calming effect. Unlike synthetic dyes, natural colourants are non-toxic, environmental friendly, pose no risk of cancer or poisoning and eliminate disposal concerns. Their extraction and application typically involve fewer chemical reactions, making them a simpler and potentially safer option than synthetic alternatives (Gulrajani, 2001). Roses are easily cultivated, making them a sustainable alternative to synthetic dyes, and their variety allows for a diverse colour range. Furthermore, rose dyes exhibit excellent colourfastness, ensuring longer-lasting, fade-resistant colours. Their anthocyanin pigments offer a range of red, pink, and purple hues (Kumari *et al.,* 2022). In anthocyanin research, solvent extraction frequently serves as a preliminary step, yielding a crude extract for subsequent analysis. To achieve maximum anthocyanin yield and purity, researchers meticulously refine extraction variables, including solvent selection, pH levels, temperature, and extraction duration. This optimization process is essential for reliable and reproducible results. In view of the above, this study aimed to standardize the natural dye extraction process from rose flowers to achieve maximum dye yield.

Materials and Methods

The petals of rose flower used in the natural dye extraction process were procured from the Ghazipur market, New Delhi, and the extraction itself was conducted at the Division of Floriculture and Landscaping at ICAR-IARI, New Delhi. Rose petal anthocyanin, a natural dye, was isolated through a 90-min hot-air oven solvent extraction. This process utilized a specific ratio of rose petals to a solvent (water and Hcl). To optimize the anthocyanin extraction process, researchers used a statistical tool called Response Surface Methodology (RSM), a widely accepted statistical technique for optimizing the extraction of bioactive compounds from plant sources (Bezerra *et al.,* 2008). The yield of natural dye was determined using anthocyanin content and redness index.

Estimation of total anthocyanin content: Total Anthocyanin Content (mg g^{-1}) was determined by pH differential method (Rapisarda *et al.,* 2000) and expressed as cyanidin-3-glucoside (Cy-3-glc) equivalents, as per the following formula:

$$TAC\left(\frac{mg}{g}\right) = \frac{A \times MW \times DF \times 1000}{\varepsilon \times L \times M}$$

where, *A* is the is the absorbance value in visible region, equalling $(A_{\lambda max}\text{-}A_{700})_{pH1.0}$ - $(A_{\lambda max}\text{-}A_{700})_{pH4.5}$, which corresponds to sodium acetate-acetic acid buffer solution when pH = 4.5 and potassium chloride-hydrochloric acid buffer solution when pH = 1.0 to dilute the anthocyanin extracts; *M*W is the molecular weight (g mol^{-1}) of Cy-3-glc (449.2 g mol^{-1} in the analysis); DF is the dilution factor; ε is the molar extinction coefficient (26,900 for Cy-3-glc); L is the path length of the cuvette (1 cm in the analysis) and M is weight (g) or volume (ml) of the sample taken.

Redness Index: Redness index measurements were determined using a method adapted from Mathieu *et al.* (1998). To quantify this index, RGB values of samples were captured under dark conditions using a high-resolution camera positioned consistently six inches above the samples. This approach, following Basavaraja *et al.* (2022), eliminated the influence of external light. The attained images were then analyzed to extract RGB values for individual pixels, which were subsequently used to calculate the redness index based on the RGB colour cube model (http://www.rapidtables.com/web/color/colortester .htm). Each colour coordinate may vary from 0 to 255.

$$Redness\ Index\ (RI) = \frac{R^2}{(B \times G^3)}$$

Optimization of anthocyanin extraction protocol: The natural dye extraction process was optimized using a Box-Behnken Design (BBD) combined with Response Surface Methodology (RSM). Three factors—acid concentration (0%, 0.1%, 0.2%), temperature (25°C, 45°C, 65°C), and solid-to-solvent ratio (1:5, 1:10, 1:15)—were tested at three levels, coded as -1, 0, and +1. A total of 17 experiments, including five canter point replicates, were conducted in randomized order to minimize uncontrolled factor effects. A quadratic model was developed to analyze the individual and interactive effects of variables on the responses: total anthocyanin content and redness index. Numerical optimization was performed using Design-Expert® software (Stat-Ease 360), with constraints applied to ensure practical ranges. Stability studies were conducted on the optimized conditions to assess dye stability. Stability testing of anthocyanin was conducted to assess its degradation under varying conditions of temperature, pH, and light exposure. Based on the optimized solutions derived from Response Surface Methodology (RSM), two specific extraction conditions were

selected for the study: 0% HCl, 250°C extraction temperature, and a 1:5 solid-to-solvent ratio, and 0.014-0.15% HCl, 250°C extraction temperature, and a 1:5 solid-to-solvent ratio. Anthocyanin extract was exposed to varying temperatures (10°C, 25°C, 45°C, 70°C, and 100°C) for up to 3 hrs, with samples collected at 1hr interval (5-min intervals at 100°C for 15 min). The pH effect was studied by exposing 1.0 ml of extract to buffer solutions (pH 1–7) at room temperature, with immediate analysis of total anthocyanin content. Light exposure was tested by storing 20 ml of extract in transparent and amber-capped tubes under continuous light, continuous dark, and natural conditions for 1 month, with samples collected every 2 days for the first 14 days and weekly, thereafter. Total anthocyanin content was measured to assess stability under these conditions.

Results and Discussion

The combined and individual impacts of independent variables—acid concentration, extraction temperature, and solid-to-solvent ratio—on the responses, including total anthocyanin content and redness index, were evaluated using Response Surface Methodology (RSM). The outcomes obtained under various experimental conditions are presented in Table 1. The findings are discussed as follows:

The effect of acid concentration, extraction temperature and solid to solvent ratio on total anthocyanin content is illustrated in Fig. 1 and 2. Fig. 1 shows that with increase in the temperature from 25 to 65°C, there was a significant quadratic effect on the total anthocyanin content with maximum yield at approximately 25°C. Similarly, a rise in acid concentration from 0 to 0.2% resulted in the increase of total anthocyanins up to some extent followed by a slight decrease. Fig. 2 reflected that with increase in solid to solvent ratio, the total anthocyanin content of extract significantly increased in a linear manner. Overall, the quadratic model was significant (F value 4.63) with a non-significant lack of fit. The regression equation (I) for total anthocyanin content is:

$$\text{Total anthocyanin content} = 3.06 - 1.09\,X_1 - 3.22\,X_2 - 3.30\,X_3 + 10.34\,X_2^2 \quad \ldots\ldots (I)$$

The effect of acid concentration, extraction temperature and solid to solvent ratio on redness index is shown in Fig. 3 and 4. The perusal of data showed that with increase in the temperature from 25 to 65°C, no significant change was reported in the redness index. However, an increase in acid concentration from 0 to 0.2% resulted in the linear increase of redness index of the extract. Fig. 4 reflected that with increase in solid to solvent ratio, the redness index of extract was significantly increased in a linear manner. The temperature of extraction did not exhibit any

Table 1: Response of 17 runs of Box Behnken Design with total anthocyanin content, redness index and colour expression

Coded level				Actual level				
X_1	X_2	X_3	Concentration of acid (X1) %	Temperature (X_2) °C	Solid: Solvent ratio (X_3)	Total anthocyanin content (TAC)	Redness index (RI)	Colour expression
0	0	0	0.1	45	01:10	7.86	97.65	
1	1	0	0.2	65	01:10	2.67	687.68	
1	0	1	0.2	45	01:15	0.80	8.67	
-1	0	1	0.0	45	01:15	0.21	0.00035	
0	0	0	0.1	45	01:10	1.65	751.17	
-1	1	0	0.0	65	01:10	14.71	0.0006	
0	0	0	0.1	45	01:10	0.05	361.00	
0	0	0	0.1	45	01:10	1.87	819.20	
1	0	-1	0.2	45	01:05	2.05	1682.00	
0	-1	-1	0.1	25	01:05	25.04	744.20	
-1	-1	0	0.0	25	01:10	15.06	0.00030	
0	1	-1	0.1	65	01:05	15.06	732.19	
1	-1	0	0.2	25	01:10	13.70	10.61	
0	1	1	0.1	65	01:15	7.16	75.00	
-1	0	-1	0.0	45	01:05	0.96	0.006	
0	-1	1	0.1	25	01:15	8.58	0.21	
0	0	0	0.1	45	01:10	3.43	737.88	

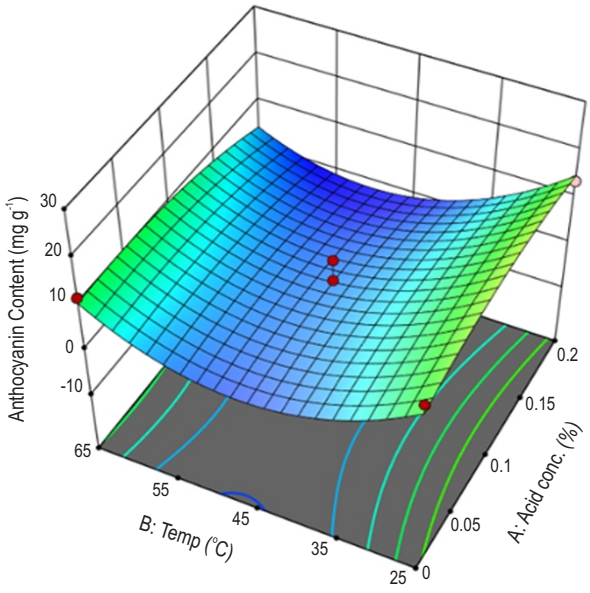


Fig. 1: Interactive effect of acid concentration (A) and temperature (B) on total anthocyanin content of extracted natural dye.

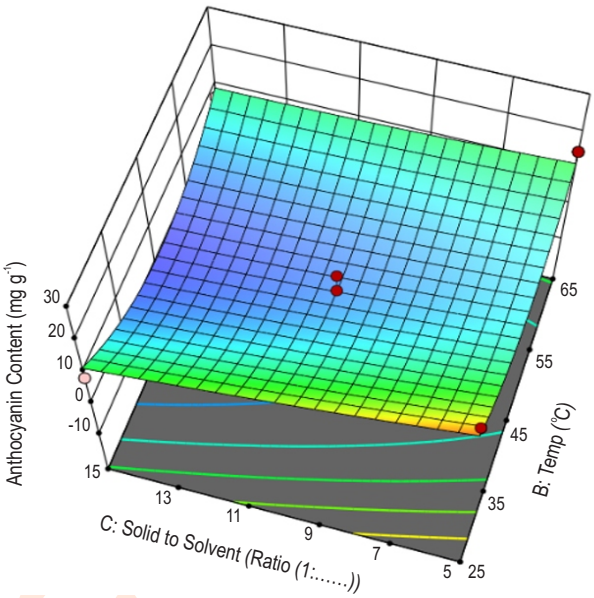


Fig. 2: Interactive effect of temperature (B) and solid: solvent (C) on total anthocyanin content of extracted natural dye.

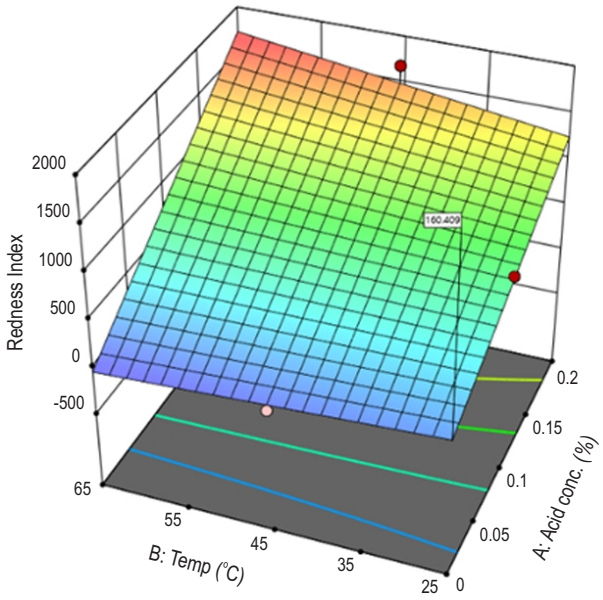


Fig. 3: Interactive effect of acid concentration (A) and temperature (B) on redness index of extracted natural dye.

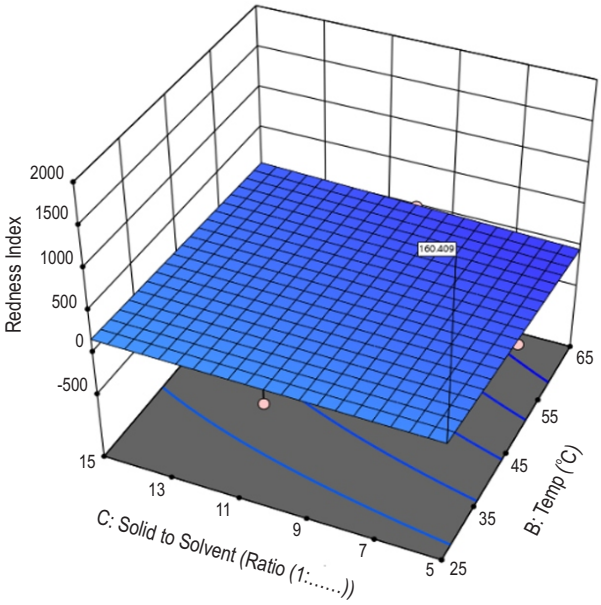


Fig. 4: Interactive effect of temperature (B) and solid: solvent (C) on total anthocyanin content of extracted natural dye.

effect on redness index. Overall, the linear model was significant (F value 5.20) with a non-significant lack of fit. A higher redness index could be perfectly correlated with the intense reddish tinge in the colour expression or *vice versa* in the particular row in Table (1) resulted from its respective combinations of process variables. The regression equation (II) for redness index is:

$$\text{Redness Index} = 394.56 + 298.62\ X_1 - 384.31X_3 - 418.33\ X_1X_3 \ldots\ldots\ldots\ldots\ldots\ldots (II)$$

Both models exhibited adequate precision values greater than 4.0, indicating their reliability for navigating the design space. The optimal conditions for better total anthocyanin content

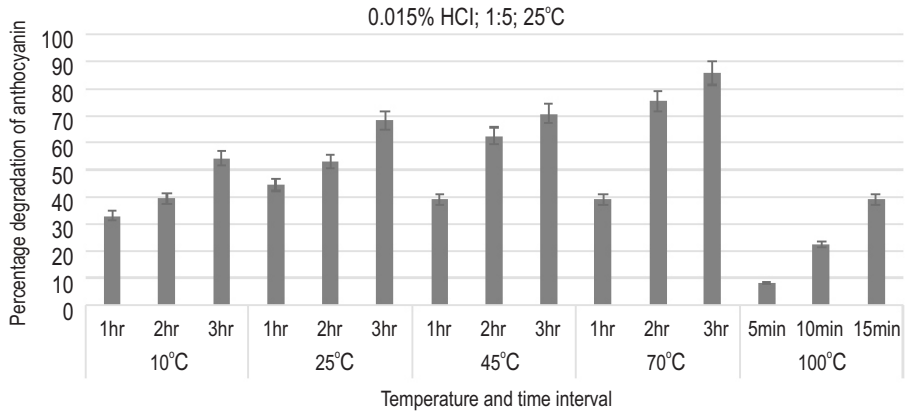


Fig. 5: Effect of temperature on stability of anthocyanin with acidic extract.

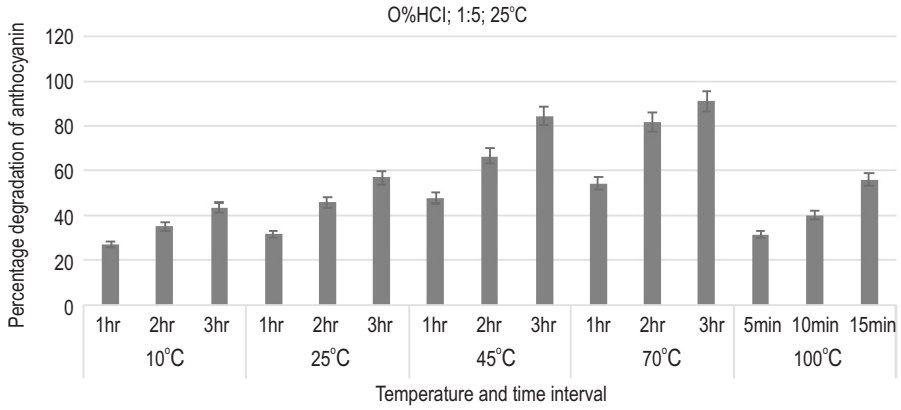


Fig. 6: Effect of temperature on stability of anthocyanins with aqueous extract.

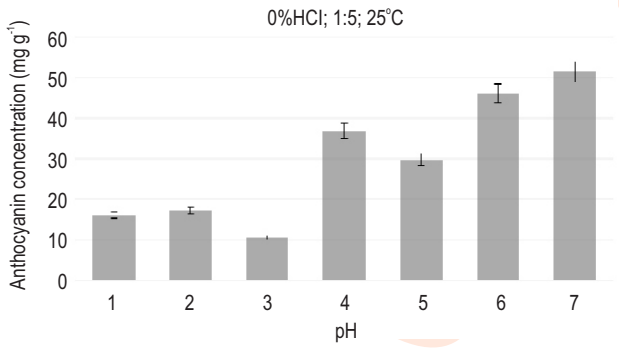


Fig. 7: Impact of pH on stability of anthocyanin with aqueous extract.

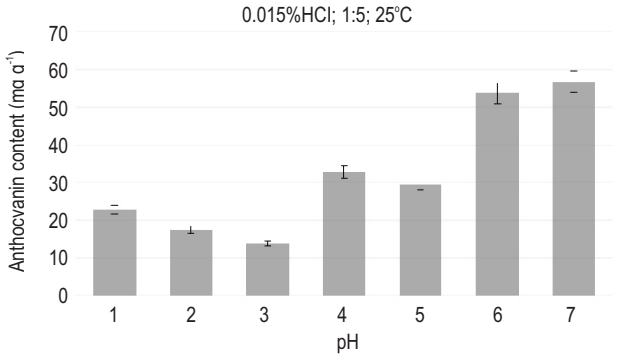


Fig. 8: Effect of pH on stability of anthocyanin with acidic extract.

(19.653 mg g^{-1}) and redness index (240.314) were achieved at an acid concentration of 0.014–0.015%, an extraction temperature of 25.185°C (approximately 25°C), and a solid-to-solvent ratio of 1:5. The desirability of optimization and prediction was 97.40% (0.974). Validation experiments under these conditions showed non-significant deviations from the model-predicted values. The extraction of anthocyanin is influenced by multiple factors, as supported by previous studies. Cacace and Mazza (2003) and Ryu and Koh (2018) highlighted that acid concentration significantly impacts anthocyanin extraction, with optimal levels varying by source. In this study, the increasing acid concentration initially enhanced both total anthocyanin content and redness

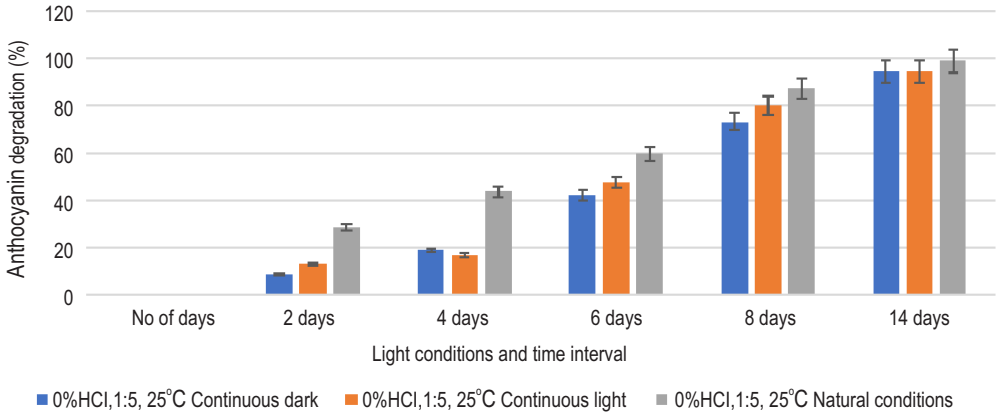


Fig. 9: Effect of light conditions on stability of anthocyanin with aqueous extract.

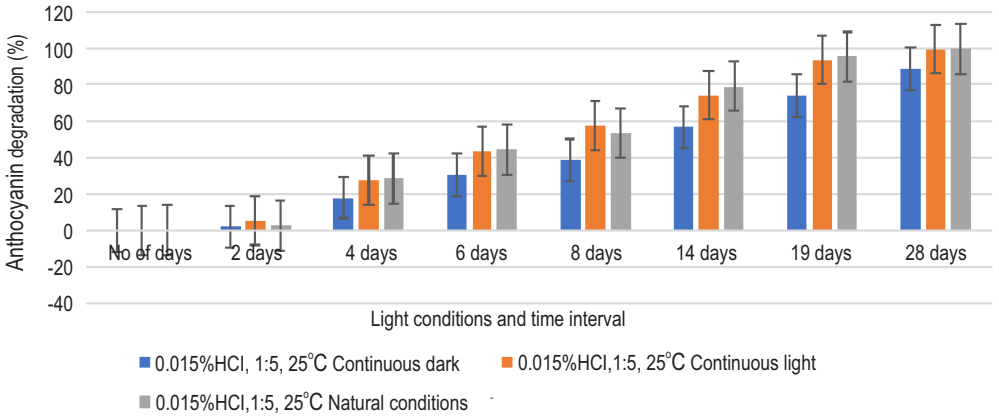


Fig. 10: Effect of light conditions on anthocyanin stability with acidic extract.

index, however, excessive concentrations showed a declining trend. Jeganathan *et al.* (2014) emphasized the importance of solid-to-solvent ratio, with higher ratios generally yielding more anthocyanins. This aligns with our findings, where increasing the solid-to-solvent ratio significantly improved both responses. Temperature exhibited a complex effect. A few studies, like those of Jeganathan *et al.* (2014) and Ku and Mun (2008) reported that moderate increase in temperature can make extraction more effective, others, like Cacace and Mazza (2003), noted that higher temperatures may degrade anthocyanins. In this study, temperature had a significant quadratic effect on total anthocyanin content, with the highest yield at 25°C, however, no significant effect was noted on the redness index. The P-values were used to identify significant terms in the regression models (Equations I and II). Model terms with P-values < 0.05 were considered significant, while those with P-values > 0.10 were deemed non-significant. This statistical approach ensured the reliability of models for predicting optimal extraction conditions.

Durability of natural dye was examined under diverse parameters, including different temperatures, pH levels and light conditions (continuous dark, continuous light, and natural conditions). Degradation of anthocyanin content was measured over time under different temperature and light conditions. Stability of anthocyanin was assessed across five temperatures (10°C, 25°C, 45°C, 70°C and 100°C) over specific time intervals. For acidic extract (0.015% HCl, 1:5 ratio, 25°C), degradation increased markedly with higher temperatures and longer exposure times as shown in Fig. 5. At 10°C, degradation was gradual, reaching 33.01% after 1 hr and 54.30% after 3 hr. In contrast, at 70°C, degradation was rapid, with 75.40% after 2 hr and 85.86% after 3 hr. At 100°C, significant degradation (39.10%) occurred within 15 min. A similar trend was observed for aqueous extract (0% HCl, 1:5 ratio, 25°C), with 27.03% degradation at 10°C after 1 hr and 90.87% at 70°C after 3 hr. At 100°C, degradation reached 56.05% within 15 min as shown in Fig. 6. Higher temperatures exponentially accelerated degradation, with

prolonged exposure intensifying the effect. For example, at 70°C, a substantial decline in total anthocyanin content (TAC) was observed within the first hour. These results underscore the sensitivity of anthocyanins to temperature, with the most pronounced degradation occurring under high temperatures and extended exposure. Anthocyanin degradation is strongly influenced by temperature and time, with elevated temperatures speeding up the process. Amperawati *et al.* (2019) noted that anthocyanin stability decreases with rising temperatures and longer heating durations, showing faster degradation rates at higher temperatures.

Similarly, Cemeroğlu *et al.* (1994) found that anthocyanin degradation in sour cherry juice followed a first-order reaction, with rates increasing significantly at higher temperatures, particularly at 80°C. Additionally, Suzery *et al.* (2020) confirmed that heating at 80°C led to a significant reduction in total anthocyanin content and antioxidant activity, emphasizing the adverse effects of prolonged high-temperature exposure on anthocyanin stability. This is further supported by a recent study by Kaya et al. (2024), which used an artificial neural network to predict anthocyanin degradation from Berberis Crategina DC., confirming that stability is critically dependent on a combination of low temperature, short exposure time, and an acidic pH. Our findings align with these studies, demonstrating that anthocyanin stability was maximum at lower temperatures (10°C, 25°C, and 45°C) compared to higher temperatures (70°C and 100°C). Degradation of anthocyanins was found to be exponential, increasing with both temperature and exposure time. These insights highlight the critical role of temperature control in preserving anthocyanin stability during storage and processing.

The stability of anthocyanin was assessed across a pH range of 1 to 7 using buffer solutions. For both aqueous (0% HCl, 1:5 ratio, 25°C) and acidic extracts (0.015% HCl, 1:5 ratio, 25°C), anthocyanin content varied significantly with pH (Fig. 7, 8). The maximum anthocyanin content was recorded at pH 7 (51.432 mg g^{-1} for aqueous and 56.729 mg g^{-1} for acidic extract), while the lowest values were observed at pH 3 (10.687 mg g^{-1} for aqueous and 13.879 mg g^{-1} for acidic extract). Anthocyanins exhibited greater stability in acidic conditions (pH 1–3), displaying vibrant red hues, while higher pH levels (pH 6–7) caused a color shift to blue and reduced stability. These findings indicate that anthocyanins are more stable and visually vibrant in acidic environments whereas alkaline conditions led to color changes and decreased stability. Anthocyanins, natural pigments found in fruits and vegetables, are highly pH-sensitive, altering their color based on environmental acidity (Verma *et al.,* 2023; Zuo *et al.* 2025). In acidic conditions, they remain stable and appear bright red in their flavylium form. However, in lesser acidic or alkaline environments, they become less stable, forming quinoidal bases and shifting to blue (Verma *et al.,* 2023).

The stability of anthocyanins is also influenced by molecular structure; larger sugar molecules enhance stability, while the presence of ascorbic acid reduces it (Levy *et al.,* 2019). Interestingly, their antioxidant activity increases in lesser acidic environments (Tina *et al*., 2020). Proteins like β-case in can protect anthocyanins from degradation, but their interactions are pH-dependent. In acidic conditions, electrostatic interactions dominate, while in less acidic environments, hydrogen bonding and van der Waals forces become more significant (Yang *et al*. 2023; Zhang *et al.* 2023). Understanding these pH-dependent interactions is crucial for preserving anthocyanin-rich products (Verma *et al.,* 2023; Zuo *et al.* 2025). Anthocyanins are generally more stable in acidic conditions, where they exhibit red hues (Wahyuningsih *et al.,* 2017; Ibrahim *et al.,* 2011). In alkaline environments, they transition to blue, although some anthocyanins, such as those from Musa acuminata bract, exhibit unique stability at specific pH ranges (Roobha *et al.,* 2011). Our results align with these findings, showing that anthocyanin content increased with pH but experienced a slight decrease at pH 5 . The color stability was greater at lower pH levels, with brighter red hues in acidic conditions and a shift to blue at higher pH levels. These insights emphasize the importance of pH control in maintaining the stability and visual appeal of anthocyanin-based products. The stability of anthocyanin was tested under continuous dark, continuous light, and natural light conditions over different time intervals. For aqueous extract (0% HCl, 1:5 ratio, 25°C), degradation reached nearly 30% after 5–6 days in both continuous light and dark conditions, but in natural light, 30% degradation occurred within 2–3 days. After 14 days, degradation exceeded 94% across all conditions, with natural light showing the highest degradation (99.13%). Anthocyanin was found to be more stable in dark conditions compared to continuous and natural light as shown in Fig. 9.

For acidic extract (0.015% HCl, 1:5 ratio, 25°C), 30% degradation was observed after 4–5 days in continuous light and 6 days in dark conditions, while natural light caused 30% degradation within 3–4 days. After 28 days, degradation reached 89.03% in dark conditions, 99.56% in continuous light, and 99.72% in natural light. Again, anthocyanin was more stable in dark conditions compared to light-exposed samples as shown in Fig. 10. Overall, anthocyanin degradation was fastest under natural light, followed by continuous light, with dark conditions providing maximum stability. These results underscore the sensitivity of anthocyanins to light exposure, particularly in natural settings.

Anthocyanins, natural pigments found in plants, are more stable in dark conditions than when exposed to light. Studies on the *Hibiscus sabdariffa, Tibouchina semidecandra,* mangosteen peel, and *Roselle calyces* extracts have consistently shown that these pigments degrade more rapidly under light exposure (Abdullah *et al.,* 2006; Chisté *et al.,* 2010; Pragalyaashree *et al.,* 2018). Our findings align with these studies, showing that anthocyanin degradation reached around 30% after 5–6 days in continuous light and dark conditions, but within 2–3 days under natural light. For acidic extract, 30% degradation occurred after 4–5 days in continuous light and 6 days in dark conditions, but within 3–4 days under natural light. Anthocyanin was consistently more stable in dark conditions compared to light-exposed samples.

This accelerated degradation under natural light is attributed to its broader spectrum of wavelengths, including ultraviolet radiation, which is particularly damaging to anthocyanins. This is further supported by a 2025 study by Zhou *et al.,* on grape anthocyanins, which provided detailed kinetic evidence that UV radiation significantly accelerates the degradation process. The findings underscore the critical role of UV as a key driver of instability. Additionally, natural conditions often involve higher oxygen exposure compared to controlled laboratory settings, and oxygen can interact with anthocyanins, promoting oxidation and degradation. These findings highlight the importance of minimizing light and oxygen exposure to preserve anthocyanin stability in natural dye applications.

This study effectively optimizes the extraction of red dye from roses, achieving better anthocyanin content and redness using specific parameters of acid concentration (0.014–0.015%), low temperature (25°C), and a 1:5 solid-to-solvent ratio. These findings offer practical utility by providing a clear framework for efficient natural dye production with enhanced color intensity. Furthermore, the research emphasizes crucial strategies for maintaining anthocyanin stability through controlled temperature, acidic pH, and light avoidance, ensuring the longevity and efficacy of natural dyes for various applications.

Acknowledgments

We acknowledge the support provided by the Head of the Division of Floriculture and Landscaping, ICAR-IARI, New Delhi for conducting this research experiment.

Authors' contribution: S. Keerthana: Execution of laboratory experiments and data collection; **B. Singh:** Conceptulization of research, Designing of experiments, Preparation of manuscript; **R. Jain:** Conceptulization of research, Designing of experiments; **M.K. Singh:** Contribution of experimental materials; **A.K. Tiwari:** Designing of experiments; **A. Joshi:** Designing experiments, Analysis of data and interpretation; **R. Bansal:** Contribution of experimental materials.

Funding: Indian Council of Agricultural Research, New Delhi.

Research content: The research content of the manuscript is original and has not been published elsewhere.

Ethical approval: The authors have followed and complied with the National/International Guidelines of relevant and appropriate authority for the use of plants in the experimental study.

Conflict of interest: The authors declare that there is no conflict of interest.

Data availability: The data is available with authors and will be made available as and when required.

Consent to publish: All authors agree to publish the paper in *Journal of Environmental Biology.*

References

Abdullah, Z., A. Ismail and N. Mohamed: Extraction and stability of anthocyanins from roselle (*Hibiscus sabdariffa* L.) calyces. *Food Chem.,* **100**, 1-7 (2006).

Amperawati, S.E., S. Suhartati and E. Widyastuti: Stability of anthocyanins extracted from red dragon fruit (*Hylocereus polyrhizus*) during pasteurization and storage. *JFST,* **56**, 3946-3954 (2019).

Basavaraja, T., A. Joshi, S. Sethi, B. Arora, B.S. Tomar, E. Varghese and A. Yadav: Extraction procedure of betalains pigments from hardy beetroot matrix and its stabilization. *J. Food Process. Preserv.,* **10**, 16844 (2022).

Bezerra, M.A., R.E. Santelli, E.P. Oliveira, L.S. Villar and L.A. Escaleira: Response surface methodology (RSM) as a tool for optimization in analytical chemistry. *Talanta,* **76**, 965-977 (2008).

Cacace, P. and G. Mazza: Anthocyanins: A review of their extraction, stability, and applications. *J. Food Sci.,* **68**, 1507-1518 (2003).

Cemeroğlu, D., I. Akata and A. Öztürk: Kinetic studies on the degradation of anthocyanins in sour cherry juice. *J. Food Sci.,* **59**, 881-884 (1994).

Chisté, M.C., M. García-Carrasco and F.J. Heredia: Stability of anthocyanins from *Hibiscus sabdariffa* L. *Food Res Int.,* **43**, 1325-1330 (2010).

Gulrajani, M.L.: Present status of natural dyes. *Ind. J. Fibr. Text. Res.,* **26**, 191-201 (2001).

Ibrahim, H., A. Ismail and N. Mohamed: Extraction and stability of anthocyanins from roselle (*Hibiscus sabdariffa* L.) calyces. *Food Chem.,* **124**, 1133-1138 (2011).

Jeganathan, P., S. Manikandan and P. Sivakumar: Optimization of anthocyanin extraction from mulberry (*Morus alba* L.) fruits using response surface methodology. *Int. J. Food Sci. Technol.,* **49**, 1671-1678 (2014).

Kaya, H.P., T. Koç, L. Ekici and G. Toğa: Anthocyanin and bioactivity properties of berberis crategina DC. In buffer system and apple juice: impact of temperature, time, and pH; Prediction using artificial neural network. *Sigma J. Engineer. Natu. Sci.,* **42**, 438-449 (2024).

Ku, C.Y. and S.H. Mun: Optimization of anthocyanin extraction from purple-fleshed sweet potato using response surface methodology. *J. Food Eng.,* **85**, 561-567 (2008).

Kumari, P., D.V.S Raju, K.V. Prasad, S. Saha, S. Panwar, S. Paul, N. Banyal, A. Bains, P. Chawla, M. Fogarasi and S. Fogarasi: Characterization of anthocyanins and their antioxidant activities in Indian rose varieties (*Rosa× hybrida*) using HPLC. *Antioxid,* **11**, 2032 (2022).

Leghari, A.J., U.A. Laghari, A.H. Laghari and T.A. Bhutto: Cultivation of rose (*Rosa indica* L.). *J. Floricul. Landsca.,* **2**, 1-4 (2016).

Levy, R., Z. Okun and A. Shpigelman: The influence of chemical structure and the presence of ascorbic acid on anthocyanins stability and spectral properties in purified model systems. *Foods*, **8**, 207 (2019).

Mallick, S.R., J. Hassan, M.A. Hoque, H. Sultana, E. Kayesh, M. Ahmed, Y. Ozaki, A. Al-Hashimi and M.H. Siddiqui: Color, proximate composition, bioactive compounds and antinutrient profiling of rose. *Sci. Rep.,* **14**, 21690 (2024).

Mathieu, R., M. Pouget, B. Cervelle and R. Escadafal: Relations between satellite based radiometric indices simulated using laboratory reflectance data and typic soil colour of an arid environment. *Rem. Sens. Environ.,* **66**, 17–28 (1998).

Najem, W., M.E. Beyrouthy, L.H. Wakim, C. Neema and N. Ouaini: Essential oil composition of *Rosa damascena* Mill. from different localities in Lebanon. *Acta. Bot. Gallica,* **158**, 365-373 (2011).

Pragalyaashree, R., R. Kumar and R.P. Singh: Stability of anthocyanins from roselle (*Hibiscus sabdariffa* L.) calyces: Effect of pH, temperature and light. *Food Chem.,* **243**, 135-142 (2018).

Rapisarda, P., F. Fanella and E. Maccarone: Reliability of analytical methods for determining anthocyanins in blood orange juices. *J. Agric. Food Chem.,* **48**, 2249-2252 (2000).

Roobha, J.H., A. Ismail and N. Mohamed: Extraction and stability of anthocyanins from *Musa acuminata* bracts. *Food Chem.,* **126**, 1426-1431 (2011).

Ryu, D. and E. Koh: Optimization of anthocyanin extraction from purple sweet potato using response surface methodology. *Food Sci. Biotechnol.,* **27**, 1429-1435 (2018).

Saati, E.A., A.D. Pusparini, M. Wachid and S. Winarsih: The anthocyanin pigment extraction from red rose as antibacterial agent. *Malays. J. Fundame. Appl. Sci.*, **14**, 184-187(2018).

Salman, M., S. Adeel, N. Habib, F. Batool, M. Usama, F. Iqbal and A. Fatima: Extraction of anthocyanin from rose petals for coloration of biomordanted wool fabric. *Coatings,* **13**, 623 (2023).

Suzery, D., J.L. Silva and J.M. Rocha: Effect of pasteurization and storage on the stability of anthocyanins and phenolic compounds in blueberry juice. *Food Sci. Technol. Int.,* **26**, 347-353 (2020).

Tena, N., J. Martín and A.G. Asuero: State of the art of anthocyanins: Antioxidant activity, sources, bioavailability, and therapeutic effect in human health. *Antioxid*, **9**, 451(2020).

Verma, M., R.P. Singh and R. Kumar: Anthocyanins: A review on their stability, antioxidant activity, and applications in food and pharmaceutical industries. *Food Sci. Technol. Int.,* **29**, 489-506 (2023).

Wahyuningsih, A., S. Suhartati and E. Widyastuti: Stability of anthocyanins extracted from red dragon fruit (*Hylocereus polyrhizus*) during pasteurization and storage. *J. Food Sci. Technol.,* **56**, 3946-3954 (2017).

Yang, P., W. Wang, Z. Xu, L. Rao, L. Zhao, Y. Wang and X. Liao: New insights into the pH dependence of anthocyanin-protein interactions by a case study of cyanidin-3-O-glucoside and bovine serum albumin. *Food Hydrocoll.*, **140**, 108649 (2023).

Zhang, L., L. Yao, F. Zhao, A. Yu, Y. Zhou, Q. Wen, J. Wang, T. Zheng and P. Chen: Protein and peptide‐based nanotechnology for enhancing stability, bioactivity, and delivery of anthocyanins. *Adv. Healthc. Mater.,* **12**, 2300473 (2023).

Zuo, L., M. Ye, L. Chen, X. Xu, S. Huang, X. Li and W. Bai: Anthocyanin-rich berries and their bioactive compounds in ocular health: Mechanisms and therapeutic potential. *J. Agric. Food Chem.,* **73**, 23724-23741 (2025).