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Extraction of Anthocyanin from Purple Cabbage and Its Use in Cosmetics

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Abstract

This study investigates the extraction, purification, stability, and applications of anthocyanins, which are natural pigments known for their strong antioxidant properties. Anthocyanins are widely used in the food, cosmetic, and pharmaceutical industries due to their health benefits and vibrant coloration. The research examines different extraction and purification methods, including solvent extraction, ultrasound-assisted extraction, and chromatography, aiming to enhance both yield and stability. Additionally, it analyzes key environmental factors such as pH, temperature, and light exposure that impact anthocyanin stability. The findings highlight anthocyanins' potential in disease prevention, particularly their cardiovascular and anti-cancer properties. Their role as natural, safer alternatives to synthetic colorants in food packaging and cosmetics is also emphasized. However, challenges related to their stability and large-scale industrial application persist. Future research should focus on optimizing processing techniques, improving encapsulation methods, and expanding their commercial viability. The study contributes to the development of sustainable, health-focused products across multiple industries.

Key Words: Anthocyanins, extraction, purification, stability, applications, antioxidants.

1. Introduction

The term "anthocyanin" comes from the Greek words "flower" and "blue." Natural pigments that belong to the flavonoid family are called anthocyanins. The most significant pigments found in vascular plants are anthocyanins. They are safe and simple to incorporate into aqueous media, which makes them appealing for usage as naturally occurring water-soluble colorants. These pigments are what give some plants' flowers and fruits their glossy orange, pink, red, violet, and blue hues (Özen & Eksi, 2017,

2456-1878; Cisowska & Hendrich, 2011, 1-158; Teresa et al., 2013, 1804-1816; Landi et al., 2015, 0098-8472). The aglycon forms of anthocyanins, sometimes referred to as anthocyanidins, are made up of hydroxyl and methoxy groups arranged in different locations and have as their structural base the flavylium ion or 2-phenylbenzopyrilium. The number and configuration of hydroxyl and methoxy moieties have allowed for the identification of over 635 anthocyanins. The six most widely known anthocyanidins in

plants are malvidin, cyanidin, peonidin, delphinidin, petunidin, and pelargonidin (Fig. 1) (Wrolstad et al., 2005, 423-428; Yan et al., 2023, 59).

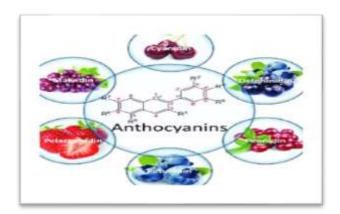


Fig. 1: Types of Anthocyanins and Their Natural Sources

Plants can be red, magenta, or blue due to the presence of anthocyanins, which are naturally occurring water-soluble pigments of flavonoids, in the vacuoles of their flowers, fruits, stems, and leaves. The stability and safety of natural pigments are higher than those of manufactured ones. Among these, anthocyanins are among the greatest natural alternatives to synthetic pigments and have been utilized extensively because to their safe and beneficial biological properties (Kaur et al., 2021, 1–14; Xue et al., 2024, 2815). These pigments also give some plants their vivid orange, pink red, violet, and blue colors in their fruits and flowers. The flavylium cation is frequently used to represent anthocyanins in a mildly acidic aqueous solution (pH 2). (Fig. 2) Through acid-base, water addition-elimination, and isomerization events, anthocyanins achieve equilibrium and change into a range of colorful and colorless forms at the pH levels found in food, plants, and the digestive system (between pH = 2 and pH = 8) (Le *et al.*, 2019, 468; (Verma et al., 2023, 1366-1373). Anthocyanins give flowers, fruits, and vegetables their vibrant appearance and aid in attracting animals, which promotes pollination and seed distribution. revealed that anthocyanins may also play a role in shielding plants from UV-induced harm. Additionally, they protect DNA and the photosynthetic system from strong radiation fluxes and act as antioxidants. Other potential roles of anthocyanins, such resistance to drought or protection from cold stress, are linked to actions specific to certain plant classes (Rana et al., 2023, 280-286; Janeiro & Brett, 2007, 1779–1786). However, the potential medical benefits of plant anthoacyanins have been extensively researched. For example, preventing cardiovascular diseases (CVDs) and having anti-inflammatory, anti-cancer, antimicrobial, and antidiabetic properties. Additionally, given their influence on the food sector, the usage of ACNs

as natural colorants, preservatives, and functional additives is examined. Additionally, it is said that they are used in the cosmetic industry because of their anti-aging and skinprotective qualities, which makes them desirable for skincare formulas (Cardoso et al., 2015, 702-709; Teng et al., 2020, 339-346).

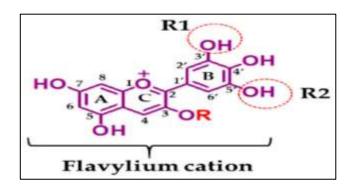


Fig. 2: Flavylium Cation Structure of Anthocyanins.

Environmental factors such pH, temperature, light, antioxidants, and metal ions can particularly affect anthocyanins, changing their chemical structure and resulting in color changes as well as a reduction in their biological activity and utilization (Le et al., 2019, 468; Nile et al., 2015, 60-68).

2. Methods of Research and the tools used

2.1. Extraction of Anthocyanins from Red Cabbage

Materials and Reagents

Red cabbage – 50 g

Ethanol (96%) – diluted to 70% with distilled water Citric acid (Lemon salt) – used to adjust Ph Distilled water

Extraction Procedure

Preparation of the Extraction Solvent: Ethanol (96%) was diluted with distilled water to achieve a final concentration of 70%. The volume ratio was calculated using the dilution formula:

Where: $C_1 V_1 = C_2 V_2$

- $C_1 = 96\%$ (initial ethanol concentration)
- V_1 = volume of 96% ethanol (to be determined)
- C_2 = (desired ethanol concentration)
- V_2 = final total volume of solution

Assuming a total final volume of 100 mL, the required ethanol volume is: $V_1 = (70 \text{ x } 100) / 96$

Thus, 72.9 mL of ethanol (96%) was mixed with 27.1 mL of distilled water to obtain 100 mL of 70% ethanol.

Extraction Process:

- o The **50** g of finely chopped red cabbage was submerged in the prepared 70% ethanol solution.
- Citric acid was added to adjust the pH to approximately
 3-4, enhancing anthocyanin stability and solubility.
- The mixture was allowed to soak for 24 hours at room temperature in a sealed container, ensuring complete extraction.
- After the extraction period, the solution was filtered to remove solid residues.
- The ethanol was evaporated using mild heat to concentrate the anthocyanins.
- After evaporation, 13.4 mL of anthocyanin-rich extract was obtained.

2.2. Qualitative Identification of Anthocyanins

To confirm the presence of anthocyanins, a simple pH-based test was conducted:

- A small portion of the extracted solution was divided into test samples.
- The samples were exposed to different pH conditions using acidic (lemon juice) and alkaline (baking soda) solutions.
- A visible color change was observed: red-pink in acidic conditions and green-blue in alkaline conditions, confirming the presence of anthocyanins.

2.3. Formulation of Anthocyanin-Infused Lip Balm

Materials and Ingredients

Shea butter -16 mLBeeswax -10 mLCoconut oil -8 mLLanolin -4 mLAnthocyanin extract -1.2 mLBerry flavoring -0.8 mL

Preparation Procedure

- Melting Phase: Shea butter, beeswax, coconut oil, and lanolin were combined and melted together in a double boiler at 60–70°C to ensure homogeneity.
- 2. **Incorporation of Anthocyanins:** Once the mixture was fully melted, the anthocyanin extract was carefully added. Lanolin was crucial for stabilizing the anthocyanins in the oil-based formula, as it has amphiphilic properties that aid in dispersing water-soluble compounds into lipophilic bases.
- 3. **Flavor Addition:** Berry flavoring was added to enhance the sensory appeal of the lip balm.
- Molding and Solidification: The liquid mixture was poured into 8 small lip balm molds (each approximately 5 mL in capacity). The molds were left at room temperature until the lip balm solidified.

Observations and Final Product Evaluation

- The lip balm had a **smooth, uniform texture** with a slight pinkish hue due to the anthocyanin infusion.
- Stability tests showed no phase separation, indicating proper emulsification of the anthocyanins within the formulation.

The product exhibited **a pleasant berry aroma** and provided a moisturizing effect upon application.

3. Discussion

3.1. Chemical structure and types

Even though anthocyanins' chemical structure plays a significant role in determining their potential function, because of their unique chemical makeup, anthocyanins are characterized by an electron deficit and are highly reactive to ROS (reactive oxygen species), also known as free radicals; as a result, they are regarded as powerful natural antioxidants (Janeiro & Brett, 2007, 1779–1786; Pervaiz et al., 2017, 2).

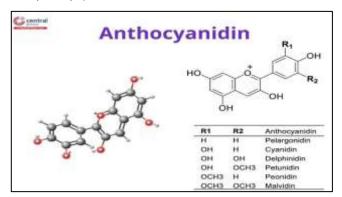


Fig. 3: Chemical structure of Anthocyanidin

In nature, anthocyanins are mostly found as heterosides. A flavonoid skeleton with a C6-C3-C6 configuration, also referred to as a flavilium ion or 2-phenylbenzopyrilium cation, is the structural basis of the aglycon form of anthocyanins, also known as anthocyanidin. This skeleton is made up of two aromatic rings, A and B, joined by a caring with two double bonds, which gives the C-ring a positive charge and displays hydroxyl and methoxyl groups in different positions (Câmara et al., 2022, 5133; Enaru et al., 2021, 1967). Both synthetic and naturally occurring anthocyanins range from yellow to purple, depending on the complexation in the B-ring, the limiting pH, the phase of anthocyanin accumulation, especially in organic molecules, and Nature contains large amounts of the six primary anthocyanidin types: malvidin (Mv), pelargonidin (Pg),

petunidin (Pt), delphinidin (Dp), cyaniding (Cy), and peonidin (Pn) (Celli et al., 2017, 20; Salimi et al., 2021, 246). Fruits and vegetables that contain anthocyanidins are connected to one or more glycosidic units. Additionally, sugars can be acylated with various organic acids and connected as mono, di, or triglycosides. Position 3 of the aglycon is always where the glycosidic units are connected to the anthocy-anidin by either α or β linkage (Liu et al., 2018, 52; Landi, 2015, 170-177). Positions 5 and 7 are associated with extra sugars in the anthocyanin molecule, and less often, positions 30 and 50. The most prevalent sugars that create anthocyanins are glucose and galactose in hexoses and rhamnose, arabinose, and xylose in pentoses. Common acylating agents include ferulic and sinapic acids, as well as cinnamic acids, often p-cinnamic or caffeic acid. Aliphatic acids include oxalic, succinic, malic, malonic, and acetic acids (Bahreini et al., 2024, e31795; Bonerz et al., 2007, 355–364). An enormous range of anthocyanins can be found in nature.the amount of hydroxylated groups, the kind and quantity of attached sugars to their structure, the aliphatic or aromatic carboxylates bonded to the sugar in the molecule, and the location of these bonds are the primary distinctions between them. Their biological potential is directly influenced by the quantity of hydroxyl groups, the degree of acylation and glycosylation, the catechol residue on the B ring, and the oxonium ion on the C ring (Fig. 3) (Verma et al., 2023, 1366-1373; Castañeda-Ovando et al., 2009, 859-871). The six more prevalent anthocyanidins found in fruits and vegetables are as follows: Cy 50%, Dp 12%, Pg 12%, Pn 12%, Pt 7%, and Mv 7%. The glycoside derivatives that are more prevalent in nature are 3monosides, 3-biosides, 3,5-, and 3,7-diglucosides; the presence of the 3-glucoside derivatives is 2.5 times more common than that of the 3,5-diglucosides; and the most prevalent anthocyanin is Cy-3-glucoside (Silva et al., 2007, 374–382; Constantin & Istrati, 2022, 1084).

3.2. Sources

ACN-rich fruit include red or violet grapes, cranberries blackberries, raspberries, blueberries, strawberries, and cherries. These substances are also found in considerable amounts in vegetables like red cabbage, eggplant, and red onions, particularly those that have a reddish or purplish color (Fig. 4) (Khoo *et al.*, 2017, 779; Mohammadalinejhad & Kurek, 2021, 3936).

Flowers like roses and hibiscus, as well as legumes like black soybeans, are noteworthy for their detectable ACN concentration. ACN levels are known to be higher in herbal and spice ingredients such as black elderberry, bilberry, and blackcurrant. Furthermore, the amount of ACNs is

influenced by some varieties of maize, such purple corn, and cereals like black rice (Castañeda-Ovando *et al.*, 2009, 859–871; Kalschne *et al.*, 2019, 30-35).

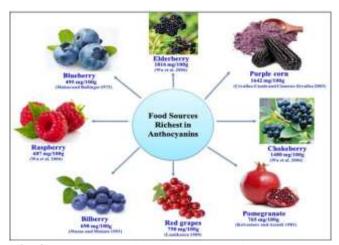


Fig. 4: Food sources Richest in Anthocyanins

The natural acidity of berries, which include glycosides of cyanidin, is the reason for the diversity in anthocyanin (ACN) content among varieties. The stability and composition of ACNs are impacted by the acidic environment, resulting in a variety of profiles among berries. Interestingly, cyanidin-derived glycosides are more frequently discovered in berries than malvidin, peonidin, and petunidin. This could be because some fruit varieties have fewer biosynthetic routes or are more susceptible to degradation (Nile et al., 2015, 60-68; Guo et al., 2022, 7573–7591). This variance emphasizes how intricately genetic predispositions, metabolic processes, environmental factors interact to shape the ACN composition of berries. The primary ACN found in mulberry fruit is C3G, which is followed by cyanidin-3-rutinoside (C3R). Five ACNs, including C3RG, C3RGa, C3G, C3Ga, and C7G, were found in mulberry fruit using high-speed counter-current chromatography. Pomegranates (Punica granatum) are known for having a high amount of ACN, and its antioxidant qualities are attributed to delphinidin-3,5diglucoside, cyanidin-3,5-diglucoside, and pelargonidin-3,5-diglucoside (Enaru et al., 2021, 1967; Awika et al., 2004, 293–301).

Because it manifests as a reddish-purple pigment that resembles magenta, cyanidin is primarily found in berries and other red vegetables in nature. Pelargonidin is a red pigment that is freely present but gives fruits their red color and flowers their orange tint (Liu et al., 2018, 52; Silva et al., 2017, 3072–3083). Delphinidin gives flowers their blue hue by appearing as a blue, reddish, or purple pigment in the plant. Petunidin, a dark red or purple water-soluble methylated anthocyanin, is frequently found in purple flowers and blackcurrants (Celli et al., 2017, 20; Anggraini

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et al., 2020, 5893-5900). The magenta pigment known as peonidin is another methylated anthocyanin that is widely present in red wines, berries, and grapes. A purple pigment, malvidin is an O-methylated anthocyanin that gives some flowers their blue hue. It is the primary pigment in red wines. Delphinidin, cyanidin, and pelargonidin are therefore frequently found in fruits, but peonidin and petunidin are the most common anthocyanins in flowers (Mozos et al., 2021, 811; Gonçalves et al., 2022, 2272).

3.3. Factors

3.3.1. PH

An indication of pH is anthocyanin. Red flavylium cations, which are extremely stable, are the predominate form of anthocyanins at low pH values (≤3). The distinctive brilliant red hue seen in acidic conditions is provided by this type (**He** *et al.*, 2016, 132-145). These pigments change into colorless pseudobases when the pH reaches moderate values (4–5). Anthocyanins exhibit a violet or purple hue at neutral pH. They convert to blue quinoidal bases at higher pH values (8–9), which causes noticeable color changes and decreased stability. Under normal circumstances, these structural changes may be reversible, but exposure to high temperatures may cause them to become irreversible (Fossen *et al.*, 1998, 435-440).

3.3.2. Temperature

Another element that contributes to the destabilization of the anthocyanin molecular structure is temperature; when the temperature rises, the degree of anthocyanin degradation increases. We hypothesize that the hydrolysis of the 3-glycoside structure, which provides protection for unstable anthocyanin, may be the cause of the rapid degradation of anthocyanin at higher temperatures (Laleh et al., 2006, 90-92; Bakhshayeshi et al., 2006, 428-433). Heat processing is frequently used to preserve and extend the shelf life of food while also ensuring food safety. The application of high temperatures for particular processing times might result in noticeable changes like color, anthocyanin levels, and antioxidant capacity, depending on the intended shelf life and the food's functional properties (Oancea, 2021, 1337).

3.3.3. Light

Another element that influences anthocyanin stability is light. Light has two effects: first, it speeds up their decomposition; second, it is essential for their synthesis in living tissues, encouraging the buildup of pigments (Verma et al., 2023, 1366-1373; Khayyat et al., 2020, 955-968). Interestingly, the greatest loss of anthocyanins occurs when exposed to fluorescent light. These substances give many

plants, including berries and grapes, their vivid hues because anthocyanins have an excellent ability to absorb visible light (**Junior** *et al.*, **2023**, **565**).

3.3.4. Co-pigmentation

The chemical relationships between pigments and other (often colorless) organic molecules in the solution—often referred to as cofactors—cause the phenomena of copigmentation. It stabilizes the anthocyanins' colorful structural forms and intensifies their hue (Sari, 2016, 422-430).

When combined with anthocyanins, the co-pigments stabilize the anthocyanin pigments by either a bathochromic shift in the absorption spectra (UV–Vis region) or a hyperchromic action, depending on the acidity of the solutions. Through their delocalized π -electron systems, the co-pigments may attach to the structural forms of anthocyanins that lack electrons (**Amongne** *et al.*, **2020**, **23**). Numerous substances are being studied as co-pigments, including as flavonoids, alkaloids, amino acids, organic acids, nucleotides, polysaccharides, metallic ions, and anthocyanins themselves (self-association). According to researchers, depending on the type of co-pigments, the co-pigmentation reactions needed to intensify and change the color of anthocyanin pigments may take place through intra-or intermolecular interactions (**Bimpilas** *et al.*, **2016**, **39-46**).

3.3.5. Metallic interaction

Metal co-pigmentation is a technique used in the food industry. Co-pigmentation with positively charged alkaline earth metals or with poor metals (+2, +3) produces most powerful color effects. Only molecules generated from cyanidin, delphinidin, and petunidin among anthocyanins may chelate metals because of the free hydroxyl groups in the B ring. The most prevalent metals that can combine with anthocyanins are potassium (K), tin (Sn), magnesium (Mg), copper (Cu), and iron (Fe) (**Pramananda** *et al.*, 2021, 012104).

3.3.6. Self - Association

Anthocyanins have the ability to bond with one another; this interaction is known as self-association. Because a greater concentration of anthocyanins is required to develop and detect this phenomenon, the resultant complex does not exhibit a degree of connection as strong as co-pigmentation. In the case of neutral species, some self-association contacts are stronger than others, but they can be destabilized because of the rejection between negatively charged anionic bases and positively charged flavylium cations (Gençdag et al., 2022, 100238).

3.3.7. Effect of the solvent

Antioxidants have been extracted from plant materials, including fruits, vegetables, and other foods, using a variety of solvent combinations in the literature. Water, ethanol, methanol, acetone, and their water mixes are the most commonly used solvents for phenolic chemical extraction (Boeing et al., 2014, 48). For the extraction of phenolics, moderately polar solvents (ethanol) are better than highly polar solvents (deionized water) or less polar solvents (ethyl acetate, acetone, etc.). Organic solvents (such as methanol, ethanol, or acetone) and water-based solvents (such as pH differential buffers) are the two primary solvents utilized for anthocyanin extraction. In an acidic environment, both extract anthocyanins (Taghavi et al., 2023, 1833).

3.3.8. Metal complexation

Complexes with divalent or trivalent cations can be formed by anthocyanins that have two or more vicinal hydroxyl functionalities (mostly cyanidin, delphinidin, and petunidin glycosides). This complex formation is linked to both anthocyanin stabilization and a significant bathochromic shift in the absorption spectrum. The most common metals present in anthocyanin complexes are tin, copper, iron, aluminum, magnesium, and potassium (Jackman *et al.*, 1987, 201-247). Color diversity is also influenced by certain metalloanthocyanin complexes; it has been observed that a number of metals complex with anthocyanins can change the color of pigments, particularly toward blue tones (Bahreini *et al.*, 2024, e31795).

3.3.9. Ascorbic Acid

As an antioxidant, ascorbic acid is important for human health. Additionally, because vitamin C levels show how food deteriorates during and after processing, it is essential for food processing and storage. According to certain studies, anthocyanins break down more quickly and lose color when ascorbic acid is present. Electrophilic substances such ascorbic acid, hydrogen peroxide, and bisulfites are thought to target the nucleophilic sites of anthocyanins (Enaru et al., 2021, 1967; Farr & Giusti, 2018, 744).

3.3.10. Oxygen

An important aspect influencing anthocyanins' strength is their unsaturated chemical structure, which makes them extremely sensitive to oxygen. The breakdown of anthocyanins is accelerated by oxygen exposure, which causes color loss or the development of brown compounds. In order to stop heat degradation, oxygen molecules must be eliminated. High temperatures and oxygen are especially detrimental to anthocyanin stability, as several investigations

have shown (**Kim** et al., 2022, 102065; Cavalcanti et al., 2011, 499-509).

3.4. Methods of extraction

3.4.1. Solvent extraction method (SEM)

The most popular technique for extracting anthocyanins is SEM. It can be divided into water extraction and organic solvent extraction, as well as cold and hot extraction, depending on the temperature at which it is extracted. According to studies, anthocyanins can be successfully collected in the following circumstances: Time spent extracting: 5 minutes to 4.2 hours. Temperature range: 34.7– 52.03°C al., (Xue et 2024, 2815). Methanol, ethanol, and acidified water are common solvent s used in anthocyanin extraction. However, there are toxicol ogical issues with the usage of ethanol and methanol. On th e other hand, subcritical water extraction is said to be a mor e ecologically friendly technique that uses acidified water a t high temperatures (110–

160°C) and pressures (40 bars). This method improves anth ocyanin stability and solubility (**Khoo** *et al.*, 2017, 779). Solubility, effectiveness, and environmental impact ar e all important considerations when choosing a solvent (**Ijo** d *et al.*, 2022, 17255). Notwithstanding SEM's simplicity and convenience of use, its practical applications are limited by a number of serious drawbacks, including long extraction durations, high solvent consumption, and low efficiency (**Tan** *et al.*, 2022, 100306).

3.4.2. Ultrasound assisted extraction (UAE)

An eco-friendly technique that improves the extraction of bioactive components while using fewer organic solvents is ultrasound-assisted extraction (UAE). UAE breaks down cell walls with sound waves, which helps release bioactive components and enhances the effectiveness and quality of extraction (Shen et al., 2023, 106646). The efficiency of the extraction procedure is greatly impacted by variables including temperature, pressure, and ultrasonic intensity (Dias et al., 2021, 105584). UAE uses frequencies between 20 and 2000 kHz for operation. This method is easy to use, economical, and appropriate for extraction operations of all sizes (Soumya et al., 2019, 280-285). In order to boost production and lessen its negative effects on the environment, UAE can be used in conjunction with methods including enzyme-assisted extraction, microwave heating, and green solvents (Fu et al., 2020, 104726).

3.4.3. Supercritical fluid extraction (SFE)

The supercritical fluid extraction (SFE) method consists of two main stages: extracting the target compound with a

supercritical fluid and then rapidly removing the fluid by adjusting temperature or pressure (Tena & Asuero, 2022, 286). SFE is recognized as an environmentally friendly and sustainable extraction technique. Supercritical carbon dioxide (CO2) is widely used as the primary solvent due to its non-toxic nature, cost-effectiveness, and ability to protect extracts from oxidation. To facilitate the extraction of polar compounds like anthocyanins, ethanol is often utilized as a co-solvent (Jiao & Pour, 2018, 237-244). SFE outperforms traditional extraction methods in terms of efficiency and reduced solvent usage. Studies have identified that the optimal conditions for extracting anthocyanins from Indian blackberries are 50°C and moderate pressure exceeding 100 bar. Adding 10% ethanol as a co-solvent significantly enhances the process's efficiency and selectivity (Wang et al., 2021, 101394). Innovations in SFE include hybrid techniques such as enzyme-assisted and ultrasound-assisted SFE, which boost efficiency, lower costs, and reduce extraction time. Another advancement is subcritical CO2 extraction, which combines supercritical CO2 with ethanol for improved results (Pazir et al., 2020, 14950). Despite the high initial costs of SFE, refining parameters like pressure, extraction duration, and co-solvent integration can make the process suitable for industrial applications. Further research should prioritize optimizing operational conditions and evaluating the economic viability of large-scale production (Woźniak et al., 2017, 322).

3.4.4. Microwave assisted extraction (MAE)

In the Microwave-Assisted Extraction (MAE) process, polar molecules in the food and solvent absorb electromagnetic energy in the microwave range, which results in ion migration and dipolar rotation. Plant cells are selectively affected by microwaves, which cause the water within the cell to evaporate and the cell wall to experience high pressure. The cell wall is ruptured and heated as a result of this process, which facilitates the solvent's penetration and the movement of materials from the cell to the solvent (Orcid et al., 2023, 2255). Anthocyanins from a variety of plants, including lavender and blueberries, have been extracted using MAE. According to studies, boosting microwave power elevates the temperature, which improves the extraction of anthocyanins but may also cause heat sensitive chemicals to degrade. Low-pressure circumstances or innovations such as nitrogen-protected and vacuum MAE have been used to improve the technique and lessen the adverse effects of heating (Putra et al., 2023, 88-98). Other methods have also been proposed, including Pressurized Microwave-Assisted Extraction (PMAE) and Solvent-Free Microwave Extraction (SFME). These techniques increase yields, decrease solvent use, and speed up extraction times.

Anthocyanin extraction also employs other pressure fluid extraction methods, such as Pressurized Liquid Extraction (PLE) and Supercritical Fluid Extraction (SFE) (Yiğit et al., 2021, 16120). Even though MAE is more efficient than traditional techniques at increasing yields and using less solvent, issues with anthocyanins' thermal stability and the requirement for extractor design optimization for large-scale applications still exist (Grigoras et al., 2012, 51-58).

3.5. Methods of purification3.5.1. Column chromatography method

One popular technique for isolating and purifying anthocyanins is column chromatography. It operates by taking advantage of variations in anthocyanin distribution coefficients between the solid and mobile phases. Common adsorbents include polyamide resins, Sephadex-100, and macroporous resins. Because of their high adsorption capacity, quick pace, affordability, and reusability, macroporous resins are very useful. Despite its efficacy, column chromatography's small-scale capacity limits its use in industrial settings (Shen *et al.*, 2023, 106646).

3.5.2. Membranes Separation method

Membrane separation is a physical procedure that separates and purifies target substances, such as anthocyanins, using selective permeable membranes. Membranes for microfiltration (MF), ultra-filtration (UF), and nano-filtration (NF) are frequently utilized. This technology's broad range of applications, low energy consumption, resilience to acids and alkalis, and environmental friendliness make it useful. Although membrane separation can improve the purity of anthocyanins, its widespread use for natural anthocyanins is limited by issues such as expensive costs and low purification efficiency (**Xue** *et al.*, **2024**, **2815**).

3.6. Applications of anthocyanin3.6.1. Applications in medical field



Fig. 5: Applications of anthocyanin in medical field

3.6.2. Antioxidant activity

Food items that are wrapped in various food films may be exposed to oxidation processes brought on by high temperatures and UV radiation, which can denaturize proteins and produce lipid peroxides. Since PVA-based bioplastics lack antioxidant activity on their own, functionalization with varying percentages of anthocyanins was helpful in giving them antioxidant activity (**Pantene** *et al.*, 2024, 9922).

Anthocyanin is an antioxidant. The antioxidant properties of anthocyanins can be explained by two distinct mechanisms: single-electron transfer (SET) and hydrogen atom donor (HAT). By removing a hydrogen atom from the antioxidant (AH+), the free radical R• transforms into a more stable product in the HAT process. The antioxidant (AH+) reduces the oxidized intermediates into the stable form in the SET process by giving the free radical an electron (**Tena** *et al.*, **2020**, **451**).

3.6.3. Anticancer activity

Anthocyanin inhibits tumors in a variety of ways, including by altering signal transduction pathways, stopping the cell cycle, and causing cancer cells to undergo autophagy or death. The generation of oxidative DNA adducts is decreased, Phase II detoxification enzymes are stimulated, and anthocyanins increase the cellular capacity to absorb oxygen radicals (Lakshmikanthan *et al.*, 2024, 101245; Pausescu *et al.*, 2022, 8103).

3.6.4. Antidiabetic activity

A disorder linked to insulin resistance, type-2 diabetes, is less common when anthocyanin is present. Insulin insufficiently promotes the transfer of glucose in skeletal muscle and fat and insufficiently inhibits the generation of glucose in the liver, a condition known as insulin resistance. The mechanisms that hinder the pancreatic β -cell from secreting enough insulin to overcome insulin resistance (**Ghosh** *et al.*, 2007, 200-208).

3.6.5. Protection from Urinary tract diseases

The health benefits of anthocyanins, such as their ability to fight against urinary tract infections (UTIs), have been well investigated. UTIs are a prevalent medical issue that necessitates outpatient treatment. One major pathogen implicated in UTIs is Escherichia coli (Pausescu et al., 2022, 8103; Cerezo et al., 2020, 478).

3.6.6. Effects on cardiovascular diseases

CVD has an impact on anthocyanin. Anthocyanin has been demonstrated to have positive benefits in human trials that

evaluated risk factors for CVD following anthocyanincontaining treatments. Supplementing with anthocyanins enhanced the ability to efflux cholesterol (**Sapian** *et al.*, **2022**, **1344**; **Tena** *et al.*, **2020**, **451**).

3.6.7. Applications in Cosmetics

As customers' interest in natural materials and organic extracts grows due to worries about sustainability and health, the cosmetics sector has seen substantial development. Deodorants, hair colours, makeup, sunscreens, and skin and hair care products are just a few of the goods that include these substances (Câmara et al., 2022, 5133; Rose et al., 2018, 6790-6798). Anthocyanins (ACNs), which are natural pigments derived from plant-based sources such as blackcurrants, red grapes, and berries, are among the most promising ingredients in this field. They are used in a variety of products, such as sunscreens, hair colours, and anti-aging treatments (Câmara et al., 2022, 5133). Because ACNs are well-known for their capacity to prevent UV damage, lessen oxidative stress, and accelerate wound healing, they are a perfect fit for bio-based cosmetics. Nevertheless, these compounds have stability issues when exposed to heat and pH variations, which has led researchers to devise ways to improve their stability, such as mixing them with clay minerals or utilising them in novel emulsion systems (Table 1) (Câmara et al., 2022, 5133; Ștefănescu et al., 2023). According to recent studies, they have a significant role in improving skin health by blocking enzymes that damage the skin and slowing the growth of skin cancer cells without harming good cells. Because of these benefits, a lot of companies have begun adding ACNs to their goods in order to offer skincare and environmental protection solutions. These products demonstrate a move towards safer and more efficient treatments by providing both photoprotection and anti-aging effects (Table 1) [17] (Câmara et al., 2022, 5133).

3.6.8. Anthocyanins as anti-aging

The main pigments found in pomegranate arils are antioxidant substances called anthocyanins. Because oxidant exposure has a significant role in ageing and skin degeneration, anthocyanins may be able to protect the aged skin (Table 1) (**Ştefănescu** *et al.*, **2023**; **Abdellatif** *et al.*, **2019**, **2020**).

A Method for preparing a cream containing anthocyanin as an anti-aging agent

Approach Using acidified methanol, anthocyanins were extracted from fresh pomegranate arils and purified using Sephadex LH-20 gel-column chromatography. Additionally,

cold cream containing pomegranate anthocyanins was made using the fusion process. Compatibility, irritation, homogeneity, drug content, drug release, and stability tests were all performed on the prepared cream. Additionally, both human and abdominal rabbit permeation studies were conducted (Table 1) (Abdellatif *et al.*, 2019, 2020).

3.6.9. Anthocyanin as a hair dye

Because the anthocyanin profile from batch-to-batch extraction of the raw material was consistent, extracted anthocyanins from blackcurrant skin were tested as colours for human hair in the initial tests (Table 1) (Rose et al., 2018, 6790-6798). A dye base formulation was created using the extracted colourants in order to convert them into a practical hair dye system. Reflectance colour measurement was used to analyse the visible colour of the resulting hair dyeing, as the dye base technology did not permit direct examination by UV-vis or HPLC. Blackcurrant anthocyanin extract was discovered to be capable of producing vivid blue colouring on hair with high colour strength (Table 1) (Câmara et al., 2022, 5133).

3.6.10. Anthocyanin as a lipstick

A popular cosmetic item, lipstick has therapeutic, psychological, and social advantages. However, because certain formulations include high quantities of lead, it may be harmful to your health. Antioxidants may be able to mitigate the harmful effects of lead, according to research. Cultivated in Myanmar, Hylocereus polyrhizus (red dragon fruit) is a fruit high in anthocyanins and the antioxidant pigment betacyanin, which may help lessen oxidative stress brought on by lead. Because of this, it shows promise as a natural lipstick colourant ingredient (Table 1) (Lourith et al., 2023, 101005; Hoang et al., 2021, 106). Effective formulations provided a healthier substitute by exhibiting colour stability, antioxidant capacity, UV protection, melanin suppression, and skin penetration. With a wide range of colours and more than two years of shelf stability under accelerated testing, the utilisation of ACN sources with documented stability and health advantages proved successful (Table 1) (Lakshmikanthan et al., 2024, 101245; Oliveira et al., 2020, 7464).

3.6.11. Properties of blackberries, grapes, apples in skin care

In addition to being used in hair and fabric colours, blackberries also contain bioactive chemicals that have been shown to have anti-aging and skin-whitening properties. Apple ACNs have demonstrated promise in treating skin disorders such inflammation and acne and lowering sebum production (Table 1) (Oliveira et al., 2020, 7464).

In support of natural skinprotection, grape ACNs help prevent and lessen UV-induced skin damage. In addition, they support skin cell viability and whitening, prevent UVA damage, block enzymes that break down skin, and fight oxidative stress (Table 1) (**Petrov** *et al.*, 2024, 1209).

3.6.12. Anthocyanin as natural sun protection agent

Anthocyanins offer a number of biological qualities that including anti-inflammatory, help preserve skin, bacteriostatic, and antioxidant actions. Since anthocyanins have UV absorption bands in the 280-320 nm range they can physically block UV radiation directly (Table 1) (Câmara et al., 2022, 5133). In addition, anthocyanins can lessen melanoma by influencing aberrant melanogenesis, which is the process by which cells make melanin. Along with its strong antioxidant qualities, anthocyanin's antibacterial behaviour makes it more significant in a range of industries by preventing the colonisation of microorganisms including bacteria, fungus, and yeasts (Hoang et al., 2021, 106).

Table 1. Some plants containing anthocyanin and their cosmetics uses

Plant	Uses
Blackberries	The skin creams, skin repairing
Blueberries	sun protection factor , uv-A
	protection
	Prevent premature skin aging and
Chokeberries	wrinkling, moisture agents, UV
	filters
	Protect human
Strawberry	dermal fibroblasts against UV-A
	induced Damage
Mulberry	Skin care, anti- aging properties
	dehydration wrinkle reduction,
	Anti-aging, UV irradiation, or as a
	cream of beauty (e.g., body care,
	soap, face lifting creams), and later
Sour cherry	a prescribed product can be
	developed against eczema and
	various dermatitis
Sweet cherry	skin whitening agents
	collagen synthesis strengthens skin
	tissues, reduction in pigmentation
Red cabbage	loss, and improved growth and
	health activities as a face cream
Hibiscus	hair growth promoting activity, skin
	whitening, anti-aging

3.7. Applications in food and nutrition field 3.7.1. Food industry

Natural ingredients have a wide range of uses in the food industry, as demonstrated by the use of cranberries in beverages and dairy products and blackberries in desserts and beverages, such as fruit-flavored drinks, isotonic drinks, ready-to-drink iced tea, and vitamin waters (Lakshmikanthan *et al.*, 2024, 101245).

3.7.2. Food bio preservation

In the food industry, food safety and preservation are crucial concerns. In order to keep harmful and rotting bacteria out of food, preservatives are crucial. Anthocyanin has antibacterial activity. The microcapsules demonstrated antibacterial activity that anthocyanin microcapsules as a bio preservative to extend the shelf life of baked food products (**Da Silva** *et al.*, 2025, 110901).

3.7.3. Anthocyanins for active and intelligent food packaging

Food is shielded by food packaging from environmental contaminants and other factors like dust, shocks, scents, temperature, physical harm, light, microbes, and humidity. Applying natural pigments based on anthocyanins to food packaging materials can help to speed up commercial adoption by serving as a shelf-life indicator (**Singh** *et al.*, **2018**, **167-180**).

3.7.4. Anthocyanin as natural pigment

Anthocyanin pigments as food additives and colorants in order to increase customer acceptability of processed foods and beverages, natural colorants and additives are used. Among the pigments that are naturally produced from plants and have a pleasing hue are anthocyanins (**Khoo** *et al.*, **2017**, **799**; Cortez *et al.*, **2017**).

4. Conclusion

This research has provided an in-depth analysis of anthocyanins, their chemical structure, stability, extraction and purification methods, and their various applications across medical, cosmetic, and food industries. The results highlight anthocyanins' significant role as natural pigments and bioactive compounds with antioxidant, anti-inflammatory, and therapeutic properties. The study also emphasizes the challenges in maintaining anthocyanin stability due to environmental factors like pH, temperature, and light exposure. Based on the findings, it is recommended that future research focuses on enhancing anthocyanin stability using advanced extraction and encapsulation

techniques. Additionally, further studies should explore their potential as functional ingredients in pharmaceuticals, nutraceuticals, and sustainable food packaging. Policymakers and industry professionals can utilize these insights to promote the use of natural colorants, reducing reliance on synthetic additives. By integrating these results into practical applications, anthocyanins can contribute to safer, more sustainable solutions in medicine, cosmetics, and food production. Future studies should also consider interdisciplinary approaches to explore new applications and optimize their industrial use.

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