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Research Article

Advancing Nutritional Proficiency in Vegetable Crops: Introducing Pusa Purple Broccoli-1 as a Novel Crops

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Abstract

Pusa Purple Broccoli-1 (*Brassica oleracea* var. *italica*) is a valuable dietary source of anthocyanins and other bioactive compounds, offering significant potential for health-focused industries. However, anthocyanin bioavailability in humans remains limited and is influenced by factors such as food matrix and chemical structure. Despite these challenges, regular consumption of anthocyanin-rich foods like purple broccoli has shown promising benefits for cardiovascular health, cancer prevention, and cognitive function. This crop also serves as a natural food colorant, with anthocyanin concentrations ranging from 10 to 33 mg Cy3G per 100 g fresh weight. Experiments at various temperatures have demonstrated the effects of environmental conditions on anthocyanin accumulation, emphasizing the importance of optimizing anthocyanin accumulation conditions. Advances in genetic research and environmental management can enhance anthocyanin content, making *Pusa Purple Broccoli*-1 an important functional food. This review highlights the health benefits, bioavailability, and stability of anthocyanins in purple broccoli, promoting its use as a novel cole crop for improving human health and meeting the demand for nutrient-dense foods.

Keywords: Anthocyanin; Bioavailability; Cardiovascular Health; Functional Food; Pusa Purple Broccoli-1

Introduction

The increasing global demand for nutrient-dense foods has spurred the development of crops with enhanced nutritional profiles, including those rich in bioactive compounds such as anthocyanins. Among these, Pusa Purple Broccoli-1 (*Brassica oleracea* var. *italica*) has garnered significant attention due to its high anthocyanin content, positioning it as a valuable crop for both agricultural and health-focused industries. Anthocyanins, water-soluble pigments responsible for the red, purple, and blue hues in many fruits and vegetables, are well-known for their potent antioxidant properties and wide range of health benefits. The presence of these compounds in purple broccoli contributes to its ability to support human health by protecting against oxidative stress, inflammation, and various chronic diseases, such as cardiovascular disease and cancer.

Figure 1: Pusa Purple Broccoli-1.

Anthocyanins belong to the flavonoid family and play essential roles in plant physiology, including protection against environmental stressors such as ultraviolet (UV) radiation, cold temperatures, and pathogen attacks. In addition to their protective functions in plants, anthocyanins are increasingly recognized for their therapeutic potential in humans. They have been shown to improve cardiovascular health, reduce the risk of certain cancers, enhance cognitive function, and mitigate age-related neurodegenerative diseases. Their ability to scavenge free radicals and prevent oxidative damage is particularly relevant in combatting chronic diseases linked to oxidative stress, such as diabetes, neurodegeneration, and cardiovascular disorders [12,13].

Pusa Purple Broccoli-1, offers a rich source of anthocyanins and other polyphenolic compounds, making it an excellent candidate for inclusion in a health-promoting diet. The rising awareness of the link between diet and disease prevention has driven the exploration of anthocyanin-rich foods in reducing the risk of chronic illnesses. Anthocyanins have been shown to inhibit tumour growth, slow the progression of cancer cells, and improve endothelial function, which plays a crucial role in maintaining cardiovascular health [15].

Additionally, research has indicated that anthocyanin-rich vegetables, such as purple broccoli, may protect against oesophageal cancer and reduce age-related cognitive decline [1].

The bioavailability of anthocyanins, however, remains a challenge. Factors such as the food matrix, the glycosylation patterns of the anthocyanins, and environmental conditions during growth and post-harvest processing influence their absorption and effectiveness in the human body. Despite these limitations, the regular consumption of anthocyanin-rich foods, such as Pusa Purple Broccoli-1, may be necessary to harness the full benefits of these potent compounds. Bioavailability studies, while ongoing, suggest that the metabolites of anthocyanins may contribute significantly to their health-promoting effects, even when the parent compounds are poorly absorbed [11].

In recent years, scientific research has focused on understanding the biosynthesis and regulation of anthocyanins, particularly in crops like broccoli. The anthocyanin biosynthetic pathway is highly conserved across plant species and involves a series of enzymatic reactions starting with phenylalanine. Key enzymes such as phenylalanine ammonia-lyase (PAL), chalcone synthase (CHS), flavonoid 3'-hydroxylase (F3'H), and dihydroflavonol reductase (DFR) play critical roles in anthocyanin production [10]. Regulatory genes, including transcription factors from the MYB, bHLH, and WD40 families, orchestrate the expression of these structural genes, controlling anthocyanin accumulation in plants [10]. In broccoli, the BoMYB2 gene has been identified as a major regulator of anthocyanin synthesis, influencing the intensity of purple pigmentation in the florets [14]. Environmental factors also play a significant role in modulating anthocyanin accumulation. Conditions such as UV exposure, light intensity, and temperature can enhance or inhibit anthocyanin biosynthesis, making climate management an important consideration for maximizing anthocyanin content in crops like Pusa Purple Broccoli-1 [5]. Genetic studies and the development of molecular marker-assisted selection (MAS) have further advanced breeding programs aimed at improving anthocyanin content and stability in broccoli. Technologies like Kompetitive Allele-Specific PCR (KASP) offer cost-effective genotyping solutions for accelerating the development of anthocyanin-rich varieties [10].

In conclusion, Pusa Purple Broccoli-1, with its high anthocyanin content, holds great potential as a functional food for improving human health. Its antioxidant properties, along with its ability to protect against chronic diseases, make it a valuable addition to health-conscious diets. Further research into the bioavailability, stability, and health effects of anthocyanins will continue to enhance the understanding and utilization of purple broccoli in both food systems and therapeutic applications. Additionally, advancements in genetic research and environmental management will enable the optimization of anthocyanin-rich crops, ensuring their viability and efficacy in meeting the growing demand for nutrient-dense foods.

Effect of temperature gradients on anthocyanin accumulation in Pusa purple broccoli-1

The experiment was conducted during 2022-23 at the Indian Agricultural Research Institute, New Delhi. The use of different transplanting times to check different stages of broccoli at different temperature conditions. Time of transplanting had three levels: T_1 (October 10th), T_2 (November 10th), and T_3 (December 10th); (Table 1), significantly affecting environmental conditions during growth. Each combination was randomly assigned to individual plots.

Extraction of total anthocyanin

Chemicals and reagents

- Potassium chloride (KCl): 0.03 M solution, adjusted to pH 1.0 using concentrated hydrochloric acid (HCl).
- Sodium acetate (CH_3COONa): 0.4 M solution, adjusted to pH 4.5 using acetic acid.
- Distilled water: Used as a blank for spectrophotometric measurements.
- Anthocyanin standards: Cyanidin-3-glucoside (Cy-3-glc) equivalents were used for calculations.

Sample preparation

Neat extracts of the test samples were used for anthocyanin quantification.

pH differential method

Anthocyanin content was determined using the pH differential method described by Hosseinian., *et al*., [6]. Two buffer solutions were prepared for the assay

- Potassium chloride buffer (pH 1.0): Prepared by dissolving potassium chloride in distilled water to a concentration of 0.03 M, and adjusting the pH to 1.0 with hydrochloric acid.
- Sodium acetate buffer (pH 4.5): Prepared by dissolving sodium acetate in distilled water to a concentration of 0.4 M, and adjusting the pH to 4.5 with acetic acid.

pH Adjustment and Measurement

The pH of both buffer solutions was confirmed using a calibrated pH meter before analysis.

Sample Analysis

For each sample, 0.5 ml of the extract was added to 1.5 ml of each buffer solution. The mixtures were thoroughly mixed, and the pH was rechecked to ensure the correct pH levels were maintained. Absorbance measurements were taken at 520 nm using a spectrophotometer, with distilled water serving as the blank.

Calculation and Analysis of Total Anthocyanin Content

The absorbance difference between the two buffer solutions was used to calculate the anthocyanin concentration, expressed as cyanidin-3-glucoside equivalents (Cy-3-glc), using the following equation:

$$Total\ Anthocyanin\ Content\ (mg/100g) = \frac{A \times MW \times DF \times 1000}{\varepsilon \times l}$$

Where

- A is the absorbance difference A_{520} (pH 1.0 - pH 4.5),
- MW is the molecular weight of cyanidin-3-glucoside (449.2 g/mol),
- DF is the dilution factor,
- ε is the molar extinction coefficient for Cy-3-glc (26,900 L/mol·cm),
- l is the path length of the cuvette (1 cm).

The anthocyanin content was expressed in μg per 0.5 ml of extract.

Treatments	Total anthocyanin mg/100g (Fresh weight basis)				Total anthocyanin mg/100g (Dry weight basis)			
	R1	R2	R3	Mean	R1	R2	R3	Mean
T_1	32.15	28.14	27.9	29.39[a]	224.7	212	205.5	214.19[a]
T_2	16.49	20.75	18.5	18.57[b]	131.5	143.3	134	136.13[b]
T_3	10.38	12.42	13.2	12.0[c]	76.94	81.77	83.18	80.63[c]

Table a

This study highlights the crucial role of temperature in anthocyanin accumulation in Pusa Purple Broccoli-1, especially during curd development. The T1 transplanting (10th October) experienced ideal temperature ranges (15-20°C during the vegetative phase, 10-15°C for curd formation, and 20-23°C for curd maturation), resulting in the highest anthocyanin content. In contrast, T2, with temperatures rising to 23-25°C between 5-15 February during curd maturation, showed lower anthocyanin levels, as the elevated temperatures disrupted anthocyanin synthesis. T3, exposed to temperatures exceeding 25°C during maturation, exhibited the lowest anthocyanin accumulation due to heat-induced pigment degradation.

These findings emphasize the importance of maintaining moderate temperatures to optimize anthocyanin production, as excessive heat negatively impacts both fresh and dry-weight pigment concentrations. The research offers valuable insights for improving crop quality by managing transplanting dates and environmental conditions, particularly in regions with varying climates.

Bioavailability of anthocyanin

Anthocyanins, the pigments responsible for the rich purple hue in crops like Pusa Purple Broccoli-1, have garnered attention for their potential health benefits. However, their bioavailability, and the extent to which they are absorbed and utilized by the body remains a complex challenge. For anthocyanins to be effective, they must be ingested, absorbed, distributed, and metabolized, but these processes are influenced by factors such as the food matrix, including the broccoli itself, and the chemical structure of the anthocyanins present.

Research has shown that anthocyanin bioavailability is generally low, often below 1% [2,11]. In Pusa Purple Broccoli-1, the anthocyanins are likely non-acylated forms, which are more easily absorbed than their acylated counterparts [17]. Despite poor absorption, the health benefits of anthocyanins may be attributed to their metabolites or other polyphenolic compounds found in this broccoli variety, contributing to antioxidant activity [11]. Given the rapid absorption and elimination of anthocyanins, regular consumption of Pusa Purple Broccoli-1 may be necessary to achieve sustained health effects, highlighting the importance of incorporating this variety into a balanced diet to maximize its potential benefits.

Stability of Anthocyanin

The utilization of anthocyanins as natural colorants in food systems, such as those found in Pusa Purple Broccoli-1, is often hindered by their susceptibility to degradation during processing and storage. Factors like pH, temperature, light exposure, and interactions with other compounds significantly affect the stability of these pigments. The chemical structure of anthocyanins in Pusa Purple Broccoli-1, which likely includes non-acylated forms, plays a crucial role in determining their vulnerability to such conditions.

The stability of anthocyanins highlights strategies to mitigate degradation. For example, acylated anthocyanins, such as those in radishes, have been shown to exhibit greater thermal stability, particularly under acidic conditions [8]. This suggests that non-acylated anthocyanins in Pusa Purple Broccoli-1 might be more prone to degradation in heat or neutral pH environments. Additionally, studies on moisture control, such as those by Garzon and Wrolstad [4], emphasize the importance of reducing water activity levels to preserve the colour and integrity of anthocyanins during storage.

Innovative approaches like encapsulation have also been explored to improve anthocyanin stability in food systems. For instance, Idham., *et al*. [7]. demonstrated that encapsulating anthocyanins with polysaccharides improved their resistance to degradation. This technique could be applied to Pusa Purple Broccoli-1 anthocyanins to enhance their viability in processed foods. However, the presence of certain compounds, like ascorbic acid, can negatively impact anthocyanin stability, as observed in acerola extracts, underscoring the need to consider ingredient interactions when incorporating purple broccoli into food products. The degradation kinetics of anthocyanins in different matrices, as reported by Kirca., *et al*. [9], also highlight the importance of designing tailored strategies to maintain the vibrant colour and bioactivity of Pusa Purple Broccoli-1 anthocyanins in various applications.

Pusa purple broccoli 1 in human health solutions

Purple Broccoli is known for its large, fleshy flower heads, which can appear in shades of purple colour arranged in a tree-like structure on branches stemming from a thick, edible stalk. Often referred to as the "crown of jewel nutrition," broccoli is not only appreciated for its delicate taste but also for its rich nutritional profile. It contains 2.8% digestible protein, 2.6g fibre, 6.0g carbohydrates, 0.3g fat, 567 IU vitamin A, 81.2 mg vitamin C, 92.8 μg vitamin K, 30 mg sodium, 42.8 mg calcium, and 60.1 mg phosphorus per 100g of the edible portion.

Research by Nichenametla., *et al*. [12]. emphasizes anthocyanins' potential in preventing tumour growth, slowing cancer cell progression, and maintaining cardiovascular health by improving blood vessel function and endothelial integrity, potentially reducing the risk of atherosclerosis. Sulforaphane, found in broccoli, is known to inhibit tumour growth and lower cancer risk, while indole-3-carbinol helps combat breast and lung cancers. Broccoli, along with other cruciferous vegetables, contains sulphur-based phytochemicals like glucosinolates and S-methyl cysteine sulfoxide, both of which are linked to cancer prevention. Glucosinolate-derived compounds, such as isothiocyanates and indoles, are particularly effective in reducing cancer risks.

Moreover, broccoli contains important phytochemicals like beta-carotene, indoles, and isothiocyanates. It offers significantly more vitamin A compared to cauliflower and cabbage, providing 130 times and 22 times more, respectively. Often regarded as one of the most nutritious vegetables, broccoli can be enjoyed in various forms, such as a side dish, in casseroles, soups, stir-fries, or raw in salads. It bears a resemblance to cauliflower but forms a head of green, purple, or white buds with thick, fleshy flower stalks. Both the heads and stalks are consumed either cooked or raw.

Studies also suggest that anthocyanin-rich vegetables, such as Pusa Purple Broccoli, may alleviate oesophageal cancer and mitigate age-related neurological decline. In addition, anthocyanins contribute to heart health, improve vision, support cognitive function, aid in ulcer healing, and alleviate conditions related to capillary fragility [1,16]. These health-promoting effects have spurred significant research into the applications of anthocyanins, including their extraction, purification, and stability in food systems [3]. Understanding the biosynthesis and regulation of anthocyanins is essential for harnessing their full therapeutic potential. Genes that regulate anthocyanin accumulation, such as R2R3-MYB, bHLH, and WD40 transcription factors, play a key role in controlling their synthesis [10,17]. Environmental factors, including light and temperature, also influence anthocyanin production in crops like broccoli [5]. Genetic studies have identified important loci, such as BoMYB2, that regulate anthocyanin biosynthesis in broccoli [14].

Conclusion

Pusa Purple Broccoli-1, rich in anthocyanins and bioactive compounds, holds significant potential as a functional food for promoting human health. Despite limited anthocyanin bioavailability due to factors such as food matrix and chemical structure, regular consumption of this anthocyanin-rich crop offers promising benefits for cardiovascular health, cancer prevention, and cognitive function. Studies demonstrate that environmental conditions, particularly temperature, critically influence anthocyanin accumulation. Advances in genetic research and environmental management strategies could further optimize anthocyanin levels, reinforcing Pusa Purple Broccoli-1's role as a natural food colorant and a nutrient-dense crop for addressing global health and dietary needs.

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Conflict of Interest

The authors declare no conflicts of interest.

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