



*Research article*

## **Phenolics, anthocyanins, and antioxidant capacity in the tassels of purple waxy corn: Effects of temperature and time during storage**

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**Abstract:** Corn tassel is an agricultural waste product that contains valuable phytochemicals and antioxidants with various potential uses. Proper post-harvest management is vital to maintain the bioactive compounds and favorable properties for processing. This study aimed to evaluate the responses of phenolics, anthocyanins, and antioxidant capacity of purple waxy corn tassels to different storage conditions and durations. Storage conditions (controlled vs. ambient) that varied in temperature and duration (ranging from 6 to 48 hours) significantly altered most of the observed parameters. Phenolics were more resistant to increased temperature and prolonged storage than anthocyanins. Determining the optimal storage duration was slightly complicated as the ideal duration for each observed parameter varied. The tassels can be stored at cold temperatures for up to 48, 6, and 24 hours to obtain the highest levels of phenolics, anthocyanins, and antioxidant activity, respectively. The correlation coefficients between phenolics and antioxidant activity were significant in both fresh and dried tassels. Optimizing the storage conditions to retain phenolics can also help maintain high levels of antioxidant capacity in corn tassels. Controlled storage conditions were the best way to retain tassel weight, phenolics, anthocyanins, and antioxidant capacity in the purple tassels of waxy corn. The most prolonged acceptable storage durations varied depending on the traits. The optimum light and oxygen exposures during storage and the best drying methods are still uncertain; therefore, further research is necessary to establish good handling practices for corn tassels.

**Keywords:** bioactive compound; by-products utilization; corn tassel; cold storage; post-harvest management; *Zea mays* L.

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## 1. Introduction

Food processing creates millions of tons of byproducts (peels, spent grains, cobs, brans, etc.) annually, leaving a substantial financial burden on food processors and causing adverse ecological impacts. Nevertheless, several studies have proven that these byproducts have high phytoconstituents, indicating that extracting them from agro-byproducts is paramount. Moreover, there has been a recognition for recycling and revalorizing agro-industry wastes or byproducts as a multidisciplinary working area within the circular bioeconomy [1]. Besides, the wastes and byproducts from the agro-industry are fantastic sources of raw materials with growing social and economic effects that could aid in addressing the increased need for natural products as ingredients in functional food and nutraceutical companies [2,3].

Corn plants have male and female flowers. The male flower, called the tassel, produces pollen for pollination. Female plants are detasseled before shedding the pollens to prevent self-pollination and ensure the genetic purity of hybrid seeds [4]. This results in many fresh tassels because female plants occupy most seed production areas due to the 3:1 female to male ratio [5]. Those tassels are considered waste because they are not edible. However, tassels contain nutrients and natural compounds with bioactive properties and are useful for making cosmetics, functional foods, and natural pharmaceuticals [6–9]. *In vitro* studies have shown that phenolics extracted from corn tassels can reduce gastric cancer cell proliferation and exhibit antimicrobial properties against food-borne pathogens, human colorectal, and lung cancer cells [10,11]. Arctigenin, a major compound extracted from corn tassels, has several beneficial health properties, including being an amylase enzyme inhibitor, antidiabetic, anti-inflammatory, antidepressant, antiviral, anticancer, and anti-rheumatic agent [12].

Enhancing the quality of tassel-based products is crucial to commercializing phytochemicals derived from tassels. However, growing environments can hinder progress. The interaction between the cultivar and the growing environment is complex and can alter the production of phytochemicals from tassels. Cultivar selection under optimum growing environments may obtain high-quality bioactive phytochemicals from tassels [13]. Post-harvest management is another crucial factor that can impact the quantity and quality of the compounds. Tassels typically have a high relative water content to maintain pollen viability and enzyme activity. Any changes in the temperature and relative humidity due to environmental factors can alter the enzymatic processes in the pollen, leading to substantial changes in the properties of tassel phytochemicals [14].

Purple tassels are a rich source of anthocyanins and antioxidants [13,15]. It is essential to investigate post-harvest management practices such as tassel water content, storage conditions, and storage durations to prevent the loss of these bioactive phytochemicals during processing. However, more research is needed on post-harvest management practices affecting these compounds in corn tassels. Therefore, this study aimed to (i) assess the phenotypic variations of purple tassels on total phenolic content, total anthocyanin content, and antioxidant activities under two storage conditions and four storage durations, and (ii) determine the best treatment combinations to achieve optimal production of each given parameter. The information obtained in this study will help corn growers and processors optimize their post-harvest practices to obtain high-quality tassel-based bioactive phytochemicals.

## 2. Materials and methods

### 2.1. Plant materials, experimental design, and sample preparation

Two separate experiments were conducted to avoid potential bias due to the different water contents of tassels. The first experiment used fresh tassels, while the second used dry tassels. Both experiments were performed using a purple waxy corn hybrid named “KND” grown under field conditions during the dry season of 2020/2021 at the Agronomy Field Crop Research Station, Khon Kaen University, Khon Kaen, Thailand (N 16° 28' E 102° 49', 200 masl). The genotype KND was chosen because of its ability to express intense purple tassels and exhibit high levels of total anthocyanins, phenolics, and antioxidant activities [16].

The experiment was conducted using a  $2 \times 4$  factorial in a completely randomized design (CRD) with six replications. Factor A consisted of two storage conditions: controlled condition (SC1) at 4 °C and 45% relative humidity (RH), and ambient condition (SC2) at 25 °C and 30% RH. Factor B included four storage durations: 6, 12, 24, and 48 h, which were labeled as SD6, SD12, SD24, and SD48, respectively.

For the experiment, 15 uniform tassels were collected on the first day of the pollen-shed stage, following the method previously reported by Duangpapeng et al. [15]. The tassels were cut into small pieces, stored according to the treatment combinations, and then dipped in liquid nitrogen to stop the enzymatic activity. The samples were then stored at -20 °C until further extraction for the first experiment. The same method was applied for the second experiment, but those samples were freeze-dried and stored at -20 °C until further extraction.

### 2.2. Sample extraction and measurement on total phenolic content

For sample extraction, 10 grams of tassel was mixed with 100 mL of 95% ethanol in a 500 mL amber glass reagent bottle. The mixture was incubated at room temperature in the dark for three days. Subsequently, the sample was filtered through a vacuum filter and stored at -20 °C. The extracted samples were then used for measuring phenolic and antioxidant levels.

The total phenolic content of the sample was determined using the Folin-Ciocalteu method with slight modifications [17]. Briefly, 10  $\mu$ L of the extract was mixed with 120  $\mu$ L of Folin-Ciocalteu and stored at room temperature for 5 min. Then, 120  $\mu$ L of  $\text{Na}_2\text{CO}_3$  solution with a concentration of 60  $\text{g L}^{-1}$  was added, and the solution was mixed and incubated in the dark for 90 min. The absorbance was measured at 725 nm using a microplate reader (EnSight Multimode plate reader, Waltham, MA, USA). A standard curve was created using Gallic acid solution (2-40  $\mu\text{g mL}^{-1}$ ). The result was expressed in micrograms of gallic acid equivalent per gram of sample ( $\mu\text{g GAE g}^{-1}$ ).

### 2.3. Sample extraction and measurement on total anthocyanin content

The pH differential method described by Lee et al. [18] was followed to extract and measure the total anthocyanin content. First, we added 1 g of the sample to a solution of 5 mL 0.1% hydrochloric acid in methanol (v/v) and sonicated it for 10 min. Then, we centrifuged the sample at 7,000 rpm for 20 min at 4 °C. Finally, we filtered the supernatant through 0.22  $\mu\text{m}$  membrane filters and stored it at -20 °C for future use.

The sample was diluted and divided into two parts. One part, which was 0.6 mL, was mixed with

2.4 mL of either pH 1.0 (0.025 M KCl) or pH 4.5 (0.4 M CH<sub>3</sub>COONa) buffer. The mixture was then mixed and stored at room temperature for 30 min. The absorbance at 520 and 700 nm was observed using a microplate reader. The result was calculated using the following equation:

$$\text{Total anthocyanin content (mg L}^{-1}\text{)} = (A \times \text{MW} \times \text{DF} \times 1000) / (\epsilon \times l) \quad (1)$$

where A is the absorbance of the diluted sample, calculated from  $A = (A_{520} - A_{700})_{\text{pH 1.0}} - (A_{520} - A_{700})_{\text{pH 4.5}}$ , MW is the molecular weight of cyanidin-3-glucoside (449.2 g mol<sup>-1</sup>), DF is the dilution factor, 1000 is a conversion unit from molar to ppm, the molar absorptivity ( $\epsilon$ ) is 26,900 M<sup>-1</sup> cm<sup>-1</sup>, and l is inner size fluorescence cuvette (1 × 1 cm). The result was expressed in microgram cyanidin-3-glucoside equivalent per gram of sample ( $\mu\text{g C3G g}^{-1}$ ).

#### 2.4. Measurement on antioxidant capacity

To determine the antioxidant properties, we conducted two tests: the ferric-reducing antioxidant power (FRAP) assay and the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity [19]. For the FRAP assay, a fresh reagent was prepared by mixing 300 mM acetate buffer pH 3.6, 10 mM TPTZ 2,4,6-tris(2-pyridyl)-S-triazine solution in 40 mM HCl, and 20 mM FeCl<sub>3</sub> at a ratio of 10:1:1 (v/v/v). The extracted sample of 10  $\mu\text{L}$  was mixed with 190  $\mu\text{L}$  of the FRAP reagent and incubated in the dark for 30 min. The absorbance was measured at 593 nm using a microplate reader. Gallic acid (0.1–3.0  $\mu\text{g mL}^{-1}$ ) was used as a positive control, and the standard curve was calibrated using FeSO<sub>4</sub> (0.55–13.67  $\mu\text{g mL}^{-1}$ ). The result of the FRAP assay was expressed in an equivalent molar of FeSO<sub>4</sub> (M FeSO<sub>4</sub>).

To prepare the DPPH solution, we dissolved the DPPH reagent in methanol to obtain a final concentration of 200  $\mu\text{M}$ . A 96-well microplate was used for mixing 10–70  $\mu\text{L}$  of the sample with the DPPH solution at a 1:1 ratio. The mixture was then incubated under dark conditions at room temperature for 30 min. After that, the mixture was read at a wavelength of 514 nm using a microplate reader. For the positive control, gallic acid was used at concentrations ranging from 0.1 to 5.0  $\mu\text{g mL}^{-1}$ . The results were expressed as the inhibitory concentration 50 (IC<sub>50</sub>) of DPPH scavenging activity ( $\mu\text{g mL}^{-1}$ ).

#### 2.5. Statistical analysis

The data was statistically analyzed for each parameter using a two-way analysis of variance (ANOVA) following a 2 × 4 factorial experiment in a completely randomized design (CRD) to test the significant difference among treatments [20]. The mean comparison was performed using the least significant difference (LSD) at  $p \leq 0.05$ . Pearson linear correlation analysis was conducted to estimate the relationships among the measured traits. The statistical analyses were performed using Statistix10 software (version 10.0, Analytical Software, Tallahassee, FL, USA).

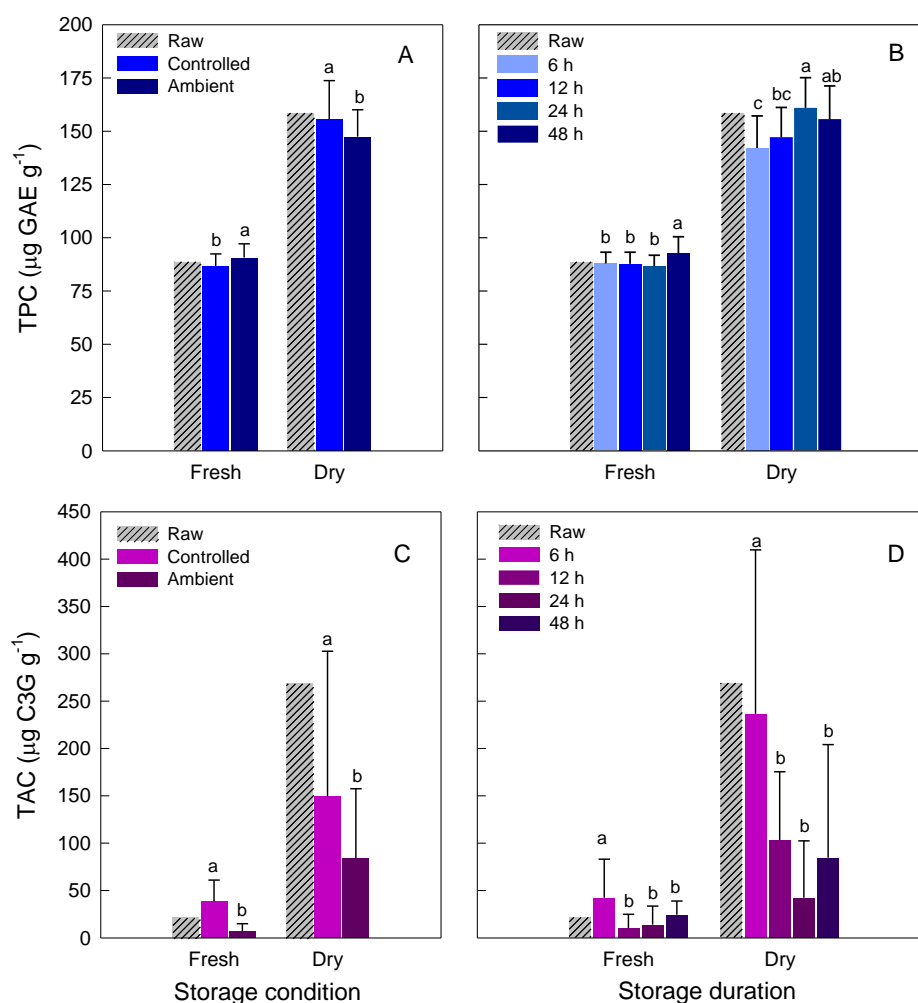
### 3. Results and discussion

#### 3.1. Variations of total phenolic content in corn tassels

Converting agricultural by-products into high-end healthcare products is a great way to improve agricultural productivity. Plant phenolics are secondary metabolites that are synthesized in plant tissues. Phenolics play pivotal roles in biological functions and are regarded as the largest bioactive

phytochemicals found in corn tassels [12]. However, multiple factors, including cultivars, growing environments, and extraction solvents, impact the synthesis of bioactive compounds such as phenols, anthocyanins, and antioxidants [21–23]. Our current investigations focused on the effects of storage conditions and duration, which may alter the bioactive phytochemicals of corn tassels.

Our study found that dry tassels had a higher total phenolic content (TPC) than fresh tassels (Figure 1A, 1B). The storage conditions had a significant effect ( $p \leq 0.05$ ) on TPC for both fresh and dry tassels (data not shown). Fresh tassels stored in ambient conditions (SC2) had slightly higher TPC means than those stored in controlled conditions (SC1). In contrast, dry tassels showed higher TPC when stored in SC1 than in SC2 (Figure 1A). Based on these results, we recommend that users store fresh tassels in SC2, while dry tassels should be kept in SC1 for optimal TPC retention.



**Figure 1.** Effects of storage conditions and storage duration on total phenolic content (A, B) and total anthocyanin content (C, D) extracted from fresh and dry tassels.

The duration of storage had a significant impact on TPC in both fresh and dry tassels (data not shown). When fresh tassels were assayed, TPC had no significant alteration until 24 h of storage (SD24) (Figure 1B). Prolonging the storage time up to 48 h (SD48) increased the TPC means significantly, implying that SD48 was the best treatment to obtain the highest TPC of fresh tassels. In contrast, the

TPC means of dry tassels showed an early increase when the storage time was extended from 6 to 48 h. Treatments SD24 and SD48 resulted in significantly higher TPC means than treatments SD6 and SD12, implying that extended storage of dry tassels up to 24 h offered the highest TPC, yet it remained high at 48 h. We also noted that the TPC means of treatment SD48 seemed slightly higher than the raw treatment (SD0) in fresh tassels and slightly lower than the SD0 in dry tassels, indicating that the TPC losses during extended storage could be negligible.

The interaction between storage condition and storage period (SC × SD) on the TPC of fresh and dry tassels was significant ( $p \leq 0.05$ ) (Table 1). Based on all the treatment combinations, fresh tassels stored at ambient temperature for 48 and 24 h had the highest TPC means, 98.1 and 90.3  $\mu\text{g GAE g}^{-1}$ , respectively. In contrast, dry tassels stored under controlled conditions for 24 and 48 h showed the highest TPC means, 171.0 and 166.1  $\mu\text{g GAE g}^{-1}$ , respectively.

The amount of water in corn tassel cells during storage can affect the variation of TPC. Corn tassels, especially in pollen grain, contain high water levels during the flowering stage [24]. Once harvested, tassels can quickly lose water, and the rate of water loss can vary depending on storage conditions. This water loss triggers stress for the tassels, leading to changes in physiological properties and enzyme activities and increased cellular leakage during storage [25]. Ultimately, this can result in broader phenotypic variations of tassel TPC.

In our present study, temperature and time during storage were the two main factors affecting the TPC of tassel (Figure 1A, B). Previous studies reported that these factors could substantially impact the total phenolics of various crops and plant species, such as yerba mansa [26], cornelian cherry [27], coffees [28], eggplant, strawberry, grape, bilberry, red raspberry, and plum [29]. For instance, in dried apricots, the TPC remained stable when stored under low temperatures and decreased when stored under thermal conditions [30]. Likewise, the TPC of litchi peels, obtained as a by-product in litchi processing, showed different reduction rates when stored at two contrasting temperatures. Samples stored at 4 °C had a lower percent reduction (20.2%) than those stored at 27 °C (37.8%) and could also be preserved for more extended periods of up to seven days [31].

In handling high-moisture raw plant materials, it is essential to consider temperature and extended time during storage. The study found that temperature had an obvious effect on the TPC of fresh and dry tassels, with ambient and cold temperatures having different impacts. Furthermore, TPC varied notably depending on the duration of storage (6, 12, 24, and 48 h). Surprisingly, satisfactory TPC levels were retained after two days of storage, as shown by the similar TPC means between the SD48 and SD0 treatments. This result indicates that phenolic production using fresh and dry tassels can be stored for up to 48 h, and phenolics are tolerant to degradation. However, to validate this conclusion, further investigations should consider other factors, such as light and oxygen exposures during storage and drying methods, that may impact the quantity and quality of tassel phenolics.

### 3.2. Variations of total anthocyanin content in corn tassels

Anthocyanins are natural pigments that give plants a red, blue, or purple color. They belong to the group of phenolics and are found in plant tissues such as flowers, fruits, and tubers [32]. Purple tassel anthocyanins accumulate in the vacuole tissues of the glume and anthers [33]. This study found that dry tassels had a significantly higher total anthocyanin content (TAC) than fresh tassels, regardless of the storage conditions or duration (Figure 1C, 1D). However, the TAC varied greatly during storage when different storage conditions and durations were used. The SC1 treatment provided significantly

higher TAC than the SC2 treatment in fresh and dry tassels, indicating that cold storage was more effective in retaining the accumulation of tassel anthocyanins. The TAC of fresh and dry tassels decreased significantly after 6 h of storage, suggesting that total anthocyanins were vulnerable to degradation due to raised temperatures and prolonged durations. Furthermore, tassel storage for 6 h had a detrimental effect on TAC. Among all treatment combinations tested, tassels stored under controlled conditions for 6 h showed the highest TAC means, representing 71.2 and 359.0  $\mu\text{g C3G g}^{-1}$  in fresh and dry samples, respectively (Table 1).

Several factors can contribute to the presence of anthocyanins in plant tissues. Among them, light is the most influential factor, as it plays a fundamental role in gene activation that affects the biosynthesis of anthocyanins in maize plant tissues and cells [33,34]. Temperature is the second factor that affects anthocyanin synthesis in plants. For example, in *Arabidopsis thaliana*, cold temperature significantly promotes anthocyanin accumulation, while high temperature degrades anthocyanin synthesis [35]. Our present study found that although TAC was susceptible to reductions due to environmental factors, cold storage at 4 °C resulted in better TAC retention than ambient storage. This finding is consistent with previous reports on other horticultural species. For instance, in litchi pericarps, TAC was reduced by 41.3% during 7 days under cold conditions, whereas a high TAC reduction of 73.0% was observed during 3 days at room temperature [31]. The study also observed that the TAC after 3 days of cold storage remained three times higher than that under room temperature.

Research has shown that storing berry juice in a cold environment can help increase the shelf-life of anthocyanins. After 49 weeks of storage at room temperature, 11–15% of the original anthocyanin concentration remained [36]. However, anthocyanins were lost by 95–99% in grape juice during long-term storage for 280 days at temperatures of 25 and 35 °C. On the other hand, storage at 5 °C resulted in a loss of 50–80% of anthocyanins, which was found to vary based on grape variety [37]. Cold storage can help to prevent the rapid degradation of anthocyanins and maintain the quality of grape juice. When it comes to blueberry powder, its TAC declined with increasing temperature. Ambient storage at 25 °C was more suitable than high-thermal storage (45–80 °C) for retaining the optimum TAC in blueberry powder [38]. In contrast, the total anthocyanins in sorghum were not affected by various storage temperatures ranging from 4 to 40 °C during storage for 180 days [39].

Apart from storage conditions and duration, processing is another factor contributing to the degradation of bioactive phytochemicals found in purple corn. A study found that steam cooking is a better method to maintain the concentration of anthocyanin and phenolic compounds and the antioxidant capacity in purple waxy corn compared to boiling [40]. Another study showed that purple corn milk from steam corn kernels had less degraded anthocyanin and phenolic compounds and a higher DPPH radical scavenging value than milk made with directly boiled kernels. However, both steamed and boiled kernels exposed to longer thermal processing times had lower compound concentrations than raw purple corn milk [41]. Furthermore, longer thermal processing times caused the purple color to fade or become colorless [42] and decreased the antioxidant capacity of purple corn flour [43]. By comparing our findings with previous reports, we learned that anthocyanins in fresh agricultural materials containing water might be sensitive to temperature and storage time. However, those factors did not matter in dry grain products. Therefore, it is crucial to identify appropriate storage temperatures for each crop to maintain the quantity and quality of favorable bioactive compounds during post-harvest management. Our current findings recommend storing corn tassels under cold conditions for no longer than 6 hours.

**Table 1.** Effects of storage conditions and storage duration on percent tassel weight loss, phytochemicals, and their antioxidant capacity in the tassels of purple waxy corn.

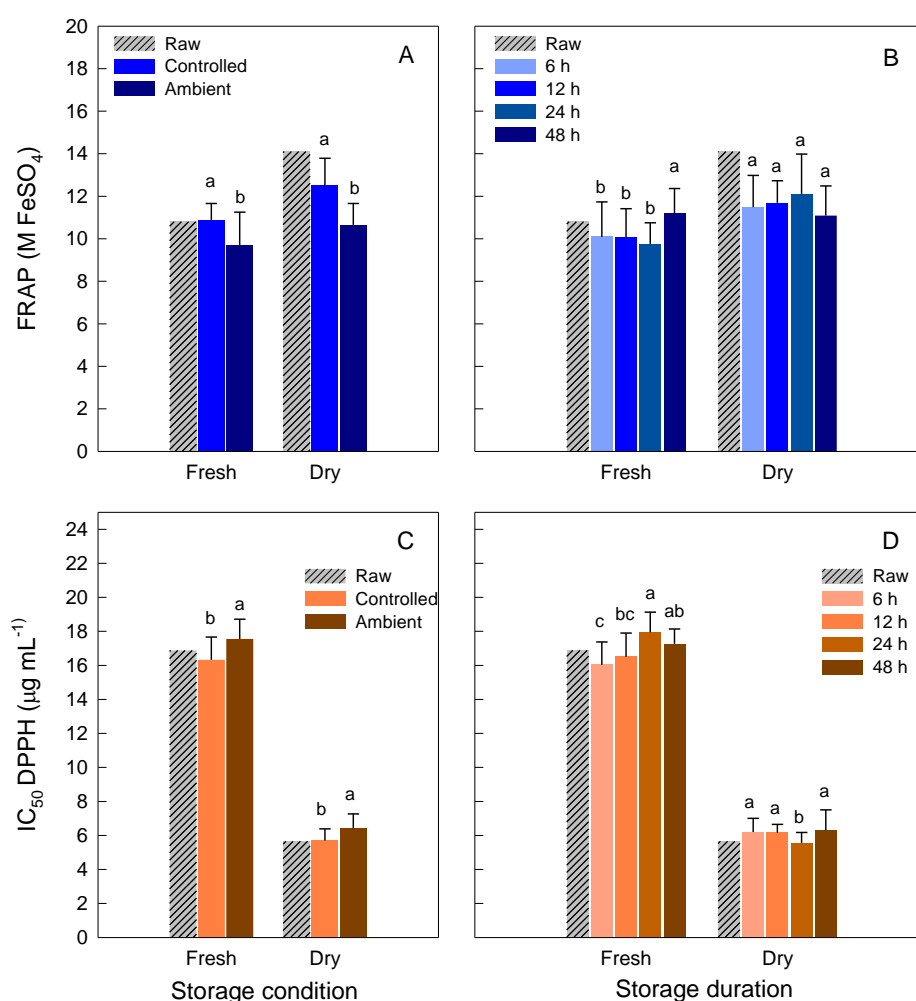
Condition	Duration (h)	Weight loss		Phytochemicals				Antioxidant capacity			
		(%)		TPC ( $\mu\text{g GAE g}^{-1}$ )		TAC ( $\mu\text{g C3G g}^{-1}$ )		FRAP (M FeSO <sub>4</sub> )		IC <sub>50</sub> DPPH ( $\mu\text{g mL}^{-1}$ )	
		Fresh	Dry	Fresh	Dry	Fresh	Dry	Fresh	Dry	Fresh	Dry
Controlled	6	2.8 ± 2.5	4.4 ± 1.7	88.6 ± 7.3	141.0 ± 5.9	71.2 ± 35.6	359.0 ± 108.7	11.5 ± 0.8	12.3 ± 1.0	15.5 ± 1.6	6.1 ± 0.5
	12	5.0 ± 1.8	6.7 ± 2.1	88.3 ± 3.4	144.0 ± 15.5	20.7 ± 17.4	154.3 ± 125.5	11.2 ± 0.4	12.1 ± 1.1	15.5 ± 0.6	6.3 ± 0.5
	24	10.6 ± 2.5	11.7 ± 4.1	82.9 ± 4.5	171.0 ± 12.9	27.8 ± 25.4	84.8 ± 165.3	10.5 ± 0.4	13.5 ± 1.5	17.1 ± 1.1	5.2 ± 0.6
	48	20.6 ± 6.5	21.7 ± 4.6	87.4 ± 6.0	166.1 ± 15.6	34.7 ± 17.8	ND	10.3 ± 0.8	12.2 ± 1.1	17.2 ± 0.9	5.3 ± 0.5
Ambient	6	9.4 ± 1.4	8.3 ± 1.8	87.3 ± 2.8	143.3 ± 21.4	13.7 ± 4.6	114.2 ± 114.8	8.7 ± 0.7	10.6 ± 1.4	16.6 ± 0.8	6.3 ± 1.1
	12	14.4 ± 2.7	12.8 ± 1.4	87.1 ± 7.4	150.5 ± 12.6	ND	52.2 ± 36.9	8.9 ± 0.7	11.2 ± 1.0	17.5 ± 1.2	6.0 ± 0.5
	24	19.4 ± 2.5	20.0 ± 3.0	90.3 ± 2.8	150.9 ± 5.6	ND	ND	9.0 ± 0.9	10.7 ± 0.9	18.8 ± 0.5	6.0 ± 0.4
	48	26.1 ± 2.5	28.3 ± 3.5	98.1 ± 5.2	145.3 ± 5.2	14.2 ± 17.7	169.2 ± 122.5	12.1 ± 0.8	10.0 ± 0.3	17.3 ± 1.0	7.4 ± 0.5
Mean		13.5	14.2	88.8	151.5	22.8	116.7	10.27	11.6	16.9	6.1
F-test		ns	ns	*	*	ns	**	**	ns	ns	**
LSD <sub>0.05</sub>		3.5	3.1	6.3	14.7	24.0	122.6	0.8	0.7	1.2	1.3

ND: not detected, TPC: total phenolic content, TAC: total anthocyanin content, FRAP: ferric-reducing antioxidant power, IC<sub>50</sub> DPPH: inhibitory concentration of DPPH scavenging activity by 50%, ns non-significant, \* and \*\* significant at  $p \leq 0.05$  and  $0.01$ , respectively, values are expressed as the mean ± SD from six replicates.



### 3.3. Variations of antioxidant potentials in corn tassels

Antioxidant activity in tassels was measured using the ferric-reducing antioxidant power (FRAP) assay. The results showed that tassels stored under controlled conditions had higher antioxidant activity than those stored under ambient temperature, both in fresh and dry tassels (Figure 2A). Moreover, fresh tassels stored for up to 48 h (SD48) had the highest antioxidant activity. In contrast, dry tassels showed no significant difference in antioxidant activity between short-term storage (SD0-SD24) and long-term storage (SD48) (Figure 2B). The interaction between storage condition and duration was significant only for fresh tassels (Table 1). Among all the treatments, tassels stored under ambient temperature for 48 h showed the highest antioxidant activity (12.1 M FeSO<sub>4</sub>), followed by those stored under controlled conditions for 6 and 12 h (11.5 and 11.2 M FeSO<sub>4</sub>, respectively).



**Figure 2.** Effects of storage conditions and storage duration on antioxidant capacity extracted from fresh and dry tassels determined by ferric-reducing antioxidant power (A, B) and DPPH radical scavenging activity (C, D).

The study found that cold storage was more effective than ambient storage in preserving the antioxidant potential of fresh and dry tassels. The IC<sub>50</sub> values, which represent the concentration of an

antioxidant-containing substance required to scavenge 50% of the initial DPPH radicals, were lower in tassels stored under controlled or cold conditions. It suggests that cold storage can better prevent the tassels from damage caused by DPPH radicals during storage. The study also found that the IC<sub>50</sub> values were the lowest at 12 and 24 h of storage in fresh and dry tassels, respectively. It means fresh and dry tassels can be preserved for up to 12 and 24 h, respectively, without losing their antioxidant capacity. In addition, the SC × SD interaction was significant only in dry tassels. Dry tassels stored under controlled conditions for 24 and 48 hours showed the highest potential of antioxidant activity with the lowest IC<sub>50</sub> DPPH values of 5.2 and 5.3 ug mL<sup>-1</sup>, respectively.

The study clearly shows that the FRAP and the DPPH assays effectively measure the antioxidant potential of corn tassels during storage. When stored under fluctuating ambient temperatures, tassels lose their antioxidant power, while cooler storage helps to preserve them. The duration of storage is also important, as antioxidant capacity tends to decrease soon after storage compared to fresh tassels. Our findings support previous studies on litchi and bitter gourd, showing that antioxidant activity decreases with prolonged storage, and higher temperatures speed up the process [31,44,45]. Therefore, the best way to preserve the antioxidant potential of corn tassels is to store them in a cool place for a short period.

#### 3.4. Tassel weight loss during storage and the associations among observed traits

The SC × SD interaction on tassel weight loss (TWL) was insignificant in fresh and dry tassels (Table 1). As a result, there were no crossover means of TWL, and the effects of each main factor, SC or SD, on TWL seemed linear. For instance, cold storage (SC1) consistently showed lower TWL means than ambient storage (SC2) for each storage duration level in fresh and dry tassels. Likewise, the means of TWL increased with prolonged storage duration from 6 to 48 h (SD6–SD48). It indicates that cold storage was more effective in minimizing tassel weight loss than ambient temperature, although the reductions were still inevitable with prolonged storage. The tassel weight loss may be attributed to evaporative water loss.

The correlation coefficients between TPC, FRAP, and IC<sub>50</sub> DPPH were stronger in dry tassels than in fresh ones (Table 2). The FRAP showed a positive and significant correlation with TPC and TAC ( $r = 0.43^{**}$  and  $0.38^*$ , respectively), whereas IC<sub>50</sub> DPPH exhibited a negative and significant correlation with TPC, TAC, and FRAP ( $r = -0.29^*$ ,  $-0.33^*$ , and  $-0.50^{**}$ , respectively). In dry tassels, TPC was negatively correlated with TAC ( $r = -0.30^*$ ) but positively correlated with FRAP ( $r = 0.60^{**}$ ). In addition, IC<sub>50</sub> DPPH was negatively correlated with TPC and FRAP ( $r = -0.77^{**}$  and  $-0.68^{**}$ , respectively).

**Table 2.** Correlation coefficients among total phenolic content (TPC), total anthocyanin content (TAC), and antioxidant capacity in purple corn tassels during storage.

Parameter	Fresh tassels			Dry tassels		
	TPC	TAC	FRAP	TPC	TAC	FRAP
TAC	-0.25ns			-0.30*		
FRAP	0.43**	0.38*		0.60**	0.13ns	
IC <sub>50</sub> DPPH	-0.29*	-0.33*	-0.50**	-0.77**	0.28ns	-0.68**

FRAP: ferric-reducing antioxidant power, IC<sub>50</sub> DPPH: inhibitory concentration of DPPH scavenging activity by 50%. ns non-significant, \* and \*\* significant at  $p \leq 0.05$  and  $0.01$ , respectively.

The results above indicate that phenolics correspond to antioxidation capacity for both assays. Tassels with high phenolic concentration possess high antioxidant activity, which agrees with previous reports. Principal component analysis (PCA) has shown a strong correlation between antioxidant activity and sub-groups of phenolic compounds, including protocatechuic acid and p-hydroxybenzoic acid contents [46]. PCA effectively classifies the yellow corn genotype based on antioxidant capacity [47] and identifies the relationship between phytochemicals and antioxidant activity in purple corn pericarp [48]. In another result, negative correlation coefficients between TAC and TPC imply that anthocyanins are not the major compound in the class of polyphenols in corn tassels. It is consistent with previous discussions [13]. In corn plants, anthocyanins regulated by the sun-red allele (*Pl* gene) are only synthesized in the vacuoles of the organs exposed to light [33,34]. In contrast, many other compounds possessing antioxidant potential can be accumulated in the pollen grains, such as volatile oil, lipids, and quercetin flavonoid pigments [8,25]. Further research is required on the responses of pollen grain or bee pollen compositions to storage practices to optimize the production of phytochemical compounds in corn tassels.

Dry corn tassels are a recommended raw material for phytochemical extraction due to their stability, practicality, and industrial convenience. They can substantially reduce electricity costs during post-harvest management and become profitable for growers and processors. However, there has yet to be a consensus on the optimal drying methods for obtaining high-quality and high-quantity bioactive phytochemicals from corn tassels. In previous studies, researchers have implemented various drying methods, such as air drying [6,9,10], oven drying [11,12], and freeze drying [13,15]. Further investigation and comparison of different drying methods are encouraged to determine the best practices for processors working on an industrial scale.

#### **4. Conclusions**

Temperature and time during storage significantly affected the phytochemicals and antioxidant capacity of purple corn tassels. Corn tassels stored under controlled conditions at 4 °C had higher levels of phenolics, anthocyanins, and antioxidant properties than those stored under ambient temperature. The optimal storage durations for retaining the highest levels of phenolics, anthocyanins, and antioxidant activity were trait-dependent, namely 48, 6, and 24 h, respectively. The remarkable relationship between phenolics and antioxidant activity was favorable, meaning users can retain high levels of both when handling corn tassels with appropriate practices.

#### **Use of AI tools declaration**

The authors declare they have not used Artificial Intelligence (AI) tools in the creation of this article.

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## Conflict of interest

All authors declare no conflict of interest.

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