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Impact of chitosan coatings on shelf-life, nutrient elements and biochemical qualities of country beans (*Phaseolus lunatus* L.) at postharvest storage

Sushmi Saha[1], Md. Zakir Hossen[1]*, Supti Mallick[1], Md. Shohidul Alam[1], and Quazi Forhad Quadir[1]

[1]Laboratory of Plant Nutrition and Environmental Chemistry, Department of Agricultural Chemistry, Faculty of Agriculture, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh.

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***Corresponding author:**
Laboratory of Plant Nutrition and Environmental Chemistry, Department of Agricultural Chemistry, Faculty of Agriculture, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh.
Email: zakirhm_ac@bau.edu.bd

ABSTRACT

Purpose: In Bangladesh, postharvest damage to various vegetables is common because of a lack of appropriate technologies. Country beans (*Phaseolus lunatus* L.), one of Bangladesh's main winter vegetables, are cultivated throughout the country, which provides numerous health advantages. A research experiment was performed to measure the impact of chitosan covering on weight loss, shelf-life, and some nutritional characteristics of country beans at postharvest storage. **Research Method:** The experiment was set up in a completely randomized design (CRD) with three replications and four treatments at room temperature (≈23-25ºC), and the treatments were: T0 (control), T1 (coating with 0.10% solution), T2 (coating with 0.20% solution) and T3 (coating with 0.30% solution). **Findings:** The use of 0.20% chitosan prevented weight reduction by 1.59% as compared to the control and extended the shelf-life up to 23.3% in country beans at 10 and 12 days after postharvest storage (DAPS), respectively. Chlorophyll-a, chlorophyll-b and total chlorophyll contents varied from 0.75-1.59, 1.36-2.86 and 2.11-4.45 mg g^{-1} tissue at 5 DAPS and 0.61-1.26, 1.10-2.27 and 1.70-3.53 mg g^{-1} tissue at 10 DAPS, respectively. Chitosan treatment T2 significantly enhanced calcium (0.77%) and phosphorus (0.51%) contents in the country beans during postharvest storage at 5 DAPS. Additionally, treatment T3 significantly increased total phenolics (3.06 mg $100g^{-1}$ tissue) in the country beans during postharvest storage at the same DAPS. **Research limitations:** The study could not measure some traits (i.e., anti-radical activity, the activity of antioxidant enzymes, etc.) due to a lack of laboratory facilities. **Originality/Value:** This experiment revealed that country beans covered with 0.20% chitosan solutions could be utilized to enhance several nutritional properties, check weight loss, and prolong the shelf-life.

INTRODUCTION

Among the major winter vegetables of Bangladesh, country beans (*Phaseolus lunatus* L.) are cultivated throughout the country. Dietary intake of beans is associated with numerous health advantages. It is an excellent nitrogen source and several vital micronutrients, including iron, zinc, magnesium, and potassium (Messina, 2014). Campos-Veja et al. (2010) stated that "beans are rich in protein, carbohydrate, dietary fiber, and are a good source of antioxidants, as well as vitamins and minerals." Furthermore, a diet that combines animal proteins with vegetable proteins is important to get the complete pool of amino acids (Melo et al., 2012). The acreage and annual production of beans were 45029 acres and 110116 metric tons in the 2013-14 fiscal year, which were 56941 acres and 170067 metric tons in the 2021-22 fiscal year (BBS, 2021; 2023) indicating an increasing trend for both areas and production of beans in Bangladesh. A similar observation was also reported by Hasan et al. (2014), and they stated that bean production is profitable, and farmers are interested in cultivating more beans in the Mymensingh and Comilla districts of the country.

Vegetables are usually purchased as a fresh agricultural product for human consumption but every year, a large volume of various vegetables decomposed in our country because of a lack of effective postharvest management solutions. Postharvest losses of vegetables include damage due to spillage and degradation during handling, storage, and transportation after harvesting to distribution. Additionally, other factors including damage from insects and mites, diseases brought on by non-infectious pathogens, and pathological rots all contribute to the postharvest loss of vegetables (Katiyar et al., 2015). However, the use of suitable chemicals throughout the pre and postharvest stages may occasionally increase the product's shelf-life and make it available over an extended period of time (Yahaya & Mardiyya, 2019; Duan et al., 2019).

Deacetylated chitosan is partially dissolved in water, which can easily be dissolved in a weak acid solution. Therefore, its application in postharvest storage of fruits and vegetables is harmless, making it a potential preservative for coating various agricultural commodities (Hosseinnejad & Jafari, 2016; Romanazzi et al., 2017; Duan et al., 2019). According to Liang et al. (2017), it is the only alkaline natural polysaccharide with biologically interchangeable and biodegradable properties. Green beans have a high rate of respiration and heat emission, which affect metabolic processes, speed up decomposition, and reduce shelf-life after harvest (ElSayed et al., 2017; Liu et al., 2022). Thus, a partially permeable chitosan layer all over the green bean regulated the interior temperature, lowered transpiration losses, and maintained constant levels of enzyme (peroxidase and polyphenol oxidase) activities, which led to normal free radicals' production, enhanced the biochemical properties and storage of the green beans (ElSayed et al., 2017; Liu et al., 2022).

Furthermore, chitosan has antifungal properties against a variety of fungi. Its application showed a significant increase in different enzymes, viz. β-1,3-glucanase, chitosanase, and chitinase, which play a role in preventing fungal growth (Zhang & Quantick, 1998). Meanwhile, chitosan′s broad-spectrum antimicrobial activities have been extensively documented (Liu et al., 2022; Sun et al., 2018; Hosseinnejad & Jafari, 2016; Alvarez et al., 2013) and *in vivo* investigations have shown that it can check or suspend the postharvest deterioration of different agricultural commodities (Sultana et al., 2019; Zakir et al., 2022). Laboratory trials showed that covering tomato fruits and carrots with chitosan solution (0.2% and 0.3%, respectively) extended their shelf-life in postharvest storage by reducing the decomposition of fruits and weight loss (Sultana et al., 2019; Zakir et al., 2022).

Due to a lack of adequate postharvest technologies, nutritional quality and consumers' acceptability of different vegetables are reduced remarkably in our country. Thus, vegetable producers/ farmers are deprived both of product loss and financial benefit, which has a lot of harmful effects on the country′s economy (Hossain et al., 2017). It has been well-documented that postharvest chitosan application improves the quality of uncooked green beans (Donsi et al., 2015; Severino et al., 2014). However, to our knowledge, the impact of chitosan treatment on the shelf-life and quality of beans has not been investigated in the context of our country. On the other hand, among the phytochemical substances, chlorophylls, polyphenols, and flavonoids are likely to contribute to health benefits (Grosso et al., 2013; Isabelle et al., 2010). Therefore, the goal of this study was to determine a suitable dose of chitosan for postharvest application in order to extend the shelf-life of country beans, as well as to explore its influence on some nutrients and biochemical features, particularly the contents of chlorophylls and total phenolics.

MATERIALS AND METHODS

Collection and sorting of country beans

Three (3.0) kg of country beans were collected from the grower′s/ producer′s field of the Sadar Upazila of Mymensingh district at full maturity level during January 2020. After that, all bean samples were transported to the Laboratory of Plant Nutrition and Environmental Chemistry, Deptartment of Agricultural Chemistry, BAU, Mymensingh. Then sorting/ screening of country beans was done manually based on their shape, size, and colour. Decomposed and pest-infested beans were discarded at this stage. Finally, almost similar shapes, sizes, and coloured beans were chosen for the laboratory trial.

Treatments and their preparation

Chitosan was obtained from Research-Lab Fine Chem Industries, Maharashtra, India (CAS No. 9012-76-4; Deacetylation >80%) and used in the present study to prepare coating solutions. There were 4 (four) treatments, namely- T0 (no coating solution), T1 (coating with 0.10% solution), T2 (coating with 0.20% solution) and T3 (coating with 0.30% solution). To prepare 0.10, 0.20, and 0.30% treatment solutions, exactly 1.0, 2.0, and 3.0 g chitosan was dissolved in 25 mL of glacial acetic acid (conc.) and then transferred into a 1.0 L volumetric flask containing about 500 mL distilled water. Before making the final volume, the pH of the solution was adjusted to 5.0 with 0.1 M NaOH, following the method adopted by Sultana et al. (2019).

Postharvest application of chitosan

A total of 16 beans were selected for the application of different chitosan treatments. These bean samples were immersed for a half minute in each treatment except T0 (control). In the case of control, beans were likewise immersed in water (distilled) with a pH of 5.0 adjusted with diluted glacial acetic acid or NaOH. Then, all samples were left to air dry beneath a ceiling fan for an hour at a temperature of 25°C. There were three replications and four treatment combinations; thus, a total of 12 (4×3) clusters of beans were handled in this laboratory trial. After applying chitosan solutions, all samples were placed in zip-lock bags following the techniques outlined by Sultana et al. (2019). Finally, all treated samples were stored in a dark place at normal (ambient) temperature (≈23-25°C).

Data recorded at postharvest storage

Data on the shelf-life of country beans were recorded at 8, 10, and 12 days after postharvest storage (DAPS), while the weight loss data of beans were measured at 2, 4, 6, 8, and 10 DAPS. On the other hand, for chemical analyses, four (4) country beans from each replication were randomly selected at 0 (fresh), 5, and 10 DAPS. However, the shelf-life of country beans was measured by static testing, where bean samples were left on a dark shelf to rot and were then visually counted until 40-50% of a bean was rotten at the respective storage time (days) (Zakir et al., 2022).

$$Shelf-life\ (\%) = \frac{No.of\ beans\ decay\ at\ storage \times 100}{Total\ no.of\ beans\ in\ storage} \qquad (1)$$

Measurement of biochemical quality of country beans

Two (2) country-bean samples from each replication were collected at 0 (fresh), 5, and 10 DAPS to determine total phenol and chlorophyll contents. Total phenol estimation in the country beans was carried out with the Folin-Ciocalteau reagent as outlined by Sadasivam and Manickam (1996). The concentration of phenols in the country beans was calculated against the catechol standard curve and expressed as mg phenols/100 gm material. Chlorophyll is extracted in 80% acetone, and the absorption reading was taken at 663 nm and 645 nm using a spectrophotometer as described by Sadasivam and Manickam (1996). The contents of different types of chlorophylls (per gram tissue) present in the aqueous extract of country beans were measured with the equations mentioned below:

$$mg\ chlorophyll - a\ = 12.7\ (A_{663}) - 2.69\ (A_{645}) \times \frac{V}{1000 \times W} \dots \dots \dots (2)$$

$$mg\ chlorophyll - b\ = 22.9\ (A_{645}) - 4.68\ (A_{663}) \times \frac{V}{1000 \times W} \dots \dots \dots \dots \dots (3)$$

$$mg\ total\ chlorophyll\ = 20.2\ (A_{645}) + 8.02\ (A_{663}) \times \frac{V}{1000 \times W} \dots \dots \dots \dots (4)$$

In the above equations, A stands for absorbance at a specific wavelength; v means the final volume of chlorophyll extract in 80% acetone, and w refers to the fresh weight of the sample used to prepare the extract.

Measurement of mineral elements of country beans

To measure the contents of different mineral elements (Ca, Mg, Na, K, P, and S), collected country bean samples were chopped first and then oven-dried at a temperature of approximately 50°C following the techniques as outlined by Singh et al. (1999). The samples were then pulverized in a grinding ball mill and utilized to make an extract applying a combination of HNO_3 and $HClO_4$ at a 2:1 ratio (Singh et al., 1999). Among the mineral elements, calcium and magnesium in the aqueous extract were assessed titrimetrically. On the other hand, phosphorus and sulfur contents in the same extract were measured by spectrophotometry, while the amount of sodium and potassium were estimated by flame photometry (Singh et al., 1999).

Statistical analysis

The data were expressed as the average of three replications and standard deviations using the statistical software Minitab 17 (Minitab Ltd., UK) following analysis of variance (ANOVA) and a general linear model. The least significant difference (LSD) was performed to separate treatment means at a specific time of data collection. Pearson correlation investigation was executed to assess the correlations among the nutritional qualities (total phenol, chlorophyll, and mineral elements) of country beans.

RESULTS AND DISCUSSION

Weight loss of country beans

The impact of different treatments (chitosan coverings) on the reduction of weight loss of country beans at two-days intervals of postharvest storage is shown in Table 1, and in all cases, results were statistically insignificant. Reduction of weight of bean samples ranged from 4.73-5.99, 8.20-9.29, 10.89-12.19, 13.31-14.75, and 16.04-17.63% at 2, 4, 6, 8, and 10 DAPS, respectively. The study results revealed that the degree of weight reduction was greater in the T0 (no coating solution) treatment at all DAPS. While postharvest chitosan coating treatment T2 (0.20%) considerably decreased weight loss in most cases of data recording. However, statistically, there were insignificant variations in weight loss of country beans in between the treatments T1 and T2 during postharvest storage. Compared to the control (T0), treatments T1, T2, and T3 can preserve weight loss by 1.31, 1.59, and 0.43%, respectively, at 10 DAPS.

Regarding obtained data on weight loss of country bean samples, the study found the treatment T2 (i.e., coating with 0.2% chitosan solution) is the best (4.73%, 8.21%, 11.26%, 13.53% and 16.04% at 2, 4, 6, 8 and 10 DAPS, respectively) at normal temperature (≈23-25°C) conditions. Similar observations (0.30% and 0.20% chitosan solution) were also reported by Zakir et al. (2022) and Sultana et al. (2019) in the case of carrot and tomato, respectively stored at room temperature. Furthermore, according to Jianglian and Shaoying (2013), in fresh-cut lotus root, the use of a mixture coating containing chitosan (1%) and phytic acid (1%) reduced weight loss and malondialdehyde content, delayed discolouration (brown), inhibited the functions of enzymes (peroxidase, and polyphenol oxidase), and kept vitamin C contents at a level that was reasonably high. Zhang et al. (2018) stated that the application of gallic acid–chitosan derivatives to preserve cherry tomatoes exhibits great antioxidant ability in scavenging DPPH, hydroxyl, and superoxide anion radicals, which led to longer fruit ripening, decreased weight loss, high hardness, and little change in epidermal colour. Chitosan coatings have the potential to create a partially permeable layer surrounding the green bean that maintains the inner temperature and decreases transpiration losses (ElSayed et al., 2019), resulting in reduced weight loss. Similarly, El-Mogy et al. (2020) reported that fresh fruits and vegetables spend some weight after harvest as a result of respiration and transpiration. Therefore, the country bean coated with chitosan may be able to minimize transpiration and evaporation by producing a tiny, waterproof layer on the outside of the fruits and vegetables (Duan et al., 2019), which might account for the reduced weight loss. In addition, according to Zhang et al. (2011), chitosan treatment reduced the rate of transpiration and preserved water in the vegetables, which resulted in reducing weight loss of green beans. The present study summarized that postharvest application of this polysaccharide (chitosan) covering could be applied to prevent weight loss of country beans.

Table 1. The effect of chitosan coating treatments on weight loss of country beans at different days after postharvest storage (DAPS) at normal temperature (≈23-25ºC).

Treatments	Weight loss (%)					
	Fresh (0 DAPS)	2 DAPS	4 DAPS	6 DAPS	8 DAPS	10 DAPS
T_0	0.0	5.99	9.29	12.19	14.75	17.63
T_1	0.0	5.03	8.20	10.89	13.31	16.32
T_2	0.0	4.73	8.21	11.26	13.53	16.04
T_3	0.0	5.29	8.68	11.40	14.02	17.20
LSD (0.05)	0.0	1.56	1.57	1.59	1.99	1.65
SE (±)	0.0	0.48	0.48	0.49	0.61	0.51
Level of significance	NS	NS	NS	NS	NS	NS
CV%	0.0	15.72	9.71	7.38	7.48	5.21

NS = Non-significant

Storage longevity (shelf-life) of country beans

The pictorial views on the impact of different doses of chitosan coatings on country beans at fresh, 2, and 5 DAPS are shown in Figure 1. However, the storage longevity (shelf-life) of country beans was measured by static testing, where bean samples were left on a dark shelf to decay and were then visually counted until 40-50% of a bean was rotten at the respective storage time (days) and the results were expressed in %. There were significant differences observed in shelf-lives of country beans at 10 ($p<0.01$) and 12 ($p<0.05$) DAPS. The impact of chitosan application on shelf-lives of country beans varied from 93.3-100.0, 50.0-73.3, and 43.3-66.7% at 8, 10, and 12 DAPS, respectively (Table 2). Up to 10 DAPS, the obtained findings demonstrated no significant change in the country beans shelf-life between the treatments T2 and T3. However, treatment T2 had the highest shelf-life at 12 DAPS. When contrasted with the control (T0), the chitosan application at 0.10, 0.20, and 0.30% can extend the shelf-life of beans by up to 3.4, 23.3, and 13.3%, respectively, at 12 DAPS. Sultana et al. (2019) also reported almost similar observations in the case of tomato fruits. According to Liu et al. (2007), gray mould (induced by *Botrytis cinerea*) and blue mould (induced by *Penicillium expansum*) in tomato fruits preserved at room and refrigeration temperature have been greatly reduced by the application of 0.5 and 1% chitosan solution. Furthermore, Romanazzi et al. (2017) stated that both preharvest and postharvest application of chitosan had shown promising effects in disease control. They also stated that chitosan has a dual mechanism of action on pathogens and plants. According to their findings, chitosan inhibits the development of rotten fungi and foodborne microbial growth and persuades resistance reactions in the host plant. ElSayed et al. (2019) stated that low respiration rates during the storage of vegetables and fruits could reduce sugar losses, impacting the shelf-life and storage quality of vegetables. In addition, they also stated that the fresh green bean's active oxygen-scavenging systems were improved by the decrease in enzymes (peroxidase and polyphenol oxidase) activities. Therefore, before storage, green beans treated with chitosan may reduce the production of reactive oxygen species (ROSs) and/or promote the growth of alternative mechanisms for quenching the produced ROSs in the stored beans. Similarly, Hong et al. (2012) supported the idea that applying chitosan coating solution on guava fruits increased antioxidant capability, which is helpful in suspending the ripening process of fruits. Thus, the study results suggested that chitosan covering could be utilized to improve the shelf-life of country beans during postharvest storage, which could be owing to chitosan's ability to suppress postharvest damages of beans due to various pathogens/microbes.

Table 2. The effect of chitosan coating treatments on shelf-life of country beans at different days after postharvest storage (DAPS) at normal temperature (≈23-25ºC). Different letters indicating statistical significance at P-values ≤ 0.01 & ≤ 0.05.

Treatments	Shelf-life (%)			
	Fresh (0 DAPS)	8 DAPS	10 DAPS	12 DAPS
T_0	100.00	96.70	50.00 b	43.30 b
T_1	100.00	93.30	63.30 a	46.70 b
T_2	100.00	96.70	73.30 a	66.70 a
T_3	100.00	100.00	70.00 a	56.70 ab
LSD (0.05)	-	6.28	11.06	15.51
SE (±)	-	1.92	3.39	4.76
Level of significance	-	NS	**	*
CV%	-	3.45	9.16	15.44

** = Significant at 1% level of probability, * = Significant at 5% level of probability, NS = Not significant

Fig. 1. Pictorial views of country bean samples coated with different treatments at 2 and 5 days after post-harvest storage (DAPS) along with fresh samples.

Biochemical properties of country beans at the storage

Total phenolic content

Polyphenols are the most abundant bioactive components in different common bean cultivars. The main phenolic components in common beans are- phenolic acids, flavonoids, and pro-anthocyanidins (Yang et al., 2018). The effect of chitosan coatings on the estimated total phenolic contents in the country beans at different storage times is shown in Table 3. Its content varied from 2.55-3.06 and 3.20-4.18 mg/ 100g sample (fresh wt.) at 5 and 10 DAPS, respectively. The study results revealed that at storage, phenolic contents in the country beans reduced significantly ($p<0.01$), and chitosan coating treatments have no effect on preserving the total phenolic contents, which might be due to the chemical degradation of polyphenol and the production of the phenolic-protein complex (Xu & Chang, 2008). Furthermore, in the beginning, chitosan coating treatments lead to lower exposure to stressful situations; thus, natural defense substances, like polyphenols, were produced lower (Winter & Davis, 2006). The phenolic makeup and antioxidant properties of common bean varieties and their processed products have also been investigated extensively worldwide (Yang et al., 2018; Silva et al., 2018; Mastura et al., 2017; Aquino-Bolanos et al., 2016), and similar results were reported by them. Coelho et al. (2007) reported that long-time storage could change the colour, texture, total phenolic content, and total pro-anthocyanidins contents of beans. Martin-Cabrejas et al. (1997) pointed out that long-term storage could lower total phenolic content while increasing the total pro-anthocyanidins contents of beans. However, interestingly it is evident from Table 3 that the total phenolic content increases at 10 DAPS for all treatments, which might be due to the lower efficacy of chitosan film with higher storage time (Sultana et al., 2019; Zakir et al., 2022). Moreover, due to this reason, increases microbial stressful situations during longer storage time, which accelerates the internal production of natural defense substances, like polyphenols (Winter & Davis, 2006). However, because of their antioxidant capabilities and capacity to neutralize free radicals, phenolic compounds serve a significant physiological function in boosting plants' stress tolerance and shielding plant tissue from the damaging results of oxidative stress (Sharma et al., 2019). But the behavior of polyphenols to chitosan application depends on the class of compounds investigated as well as the biostimulant's concentration (Sánchez-Hernández et al., 2022). Similarly, Deng et al. (2018) stated that with higher enzyme (polyphenol oxidase) activity oxidation of phenolics may be accelerated. Thus, at 10 DAPS the content of total phenol decreases with increasing doses of chitosan.

Chlorophyll content

The presence of chlorophyll pigments in the chloroplasts of plant tissues gives the green colour. It plays a crucial function in the metabolism of light energy and catalyzes the production of carbohydrates in collaboration with other pigments such as carotenoids. This study measured chlorophyll-a (chl-a), chlorophyll-b (chl-b), and total chlorophyll (total chl) contents in the country beans, and the impact of chitosan coatings on the estimated chlorophyll contents at 0 (fresh), 5 and 10 DAPS is presented in Table 3. Chlorophyll-a, chl-b and total chl contents varied from 0.75-1.59, 1.36-2.86 and 2.11-4.45 mg/g tissue at 5 DAPS and 0.61-1.26, 1.10-2.27 and 1.70-3.53 mg/g tissue at 10 DAPS, respectively. The maximum amounts of all types of chlorophylls were obtained by the application of T3 treatment (coating with 0.3% chitosan solution) at 5 DAPS. It is apparent from Table 3 that all types of chlorophyll contents in the country beans were minimum at control treatment, while increased considerably with the higher application doses of chitosan coatings at both 5 and 10 DAPS. There, the chitosan coating may reduce respiration rates, leading to slower rates of chlorophyll breakdown. This finding is consistent with that of ElSayed et al. (2019), who found that even after 7 days of storage, green beans coated with chitosan (1.5%) had the maximum concentration of total chlorophyll. The application of chitosan nanoparticles along with gibberellic acid was most effective in enhancing leaf area and levels of chlorophylls and carotenoids in beans (Pereira et al., 2017). Chitosan has been shown to boost chlorophyll content in stressful environmental conditions (Katiyar et al., 2015). On the contrary, Lai et al. (2007) stated that the loss of chlorophyll under stress conditions could be due to the oxidation of chloroplast lipids and proteins or the degradation of pigment-protein complexes that defend the photosynthetic machinery in plants. So, it can be summarized that the application of chitosan coatings can be used at postharvest storage to enhance chlorophyll contents in the country beans and the maximum benefit of chlorophylls can be achieved at 5 DAPS.

Table 3. The effect of chitosan coating treatments on biochemical constituents of country beans at different days after post-harvest storage (DAPS) at normal temperature (≈23-25ºC).

Treatment	Moisture (%)		Total phenol (mg $100g^{-1}$)		Chlorophyll-a (mg g^{-1})		Chlorophyll-b (mg g^{-1})		Total chlorophyll (mg g^{-1})	
	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS
T0	91.05	88.52	2.575b	4.184a	0.752c	0.607d	1.357c	1.094c	2.109c	1.701d
T1	91.77	89.35	2.548b	3.905a	1.189b	0.765c	2.145b	1.379b	3.334b	2.144c
T2	91.39	89.62	2.748b	4.081a	1.239b	1.259a	2.236b	2.269a	3.475b	3.528a
T3	91.18	89.07	3.059a	3.204b	1.588a	1.208b	2.863a	2.178a	4.450a	3.386b
$LSD_{0.05}$	0.92	0.93	0.27	0.22	0.13	0.04	0.19	0.12	0.31	0.14
SE (±)	0.280	0.290	0.082	0.068	0.040	0.013	0.059	0.036	0.096	0.043
Level of significance	NS	NS	**	**	**	**	**	**	**	**
CV%	0.53	0.05	5.02	3.45	5.76	2.25	4.73	3.63	4.95	2.77
Fresh bean (0 DAPS)	92.85 ± 1.02		6.242 ± 0.324		0.654± 0.021		1.199± 0.036		1.853± 0.063	

Different letters indicating statistical significance at P-value < 0.01. ** indicating at p < 0.01, and NS means non-significant.

Mineral element contents

The common bean is a widely consumed legume, which contains high levels of minerals and protein (Nchimbi-Msolla & Tryphone, 2010). The impact of chitosan application on several mineral elements in the country beans at different days after postharvest storage is shown in Table 4. The concentration of Ca, Mg, Na, K, P and S in the country beans ranged from 0.44-0.77%, 0.37-0.46%, 0.50-0.63%, 2.22-2.62%, 0.32-0.51%, and 0.14-0.17% (dry wt.) at 5 DAPS and 0.48-0.59%, 0.48-0.56%, 0.49-0.59%, 1.90-2.12%, 0.34-0.38%, and 0.12-0.15% (dry wt.) at 10 DAPS, respectively. Alternatively, the mean content of Ca, Mg, Na, K, P and S in fresh country bean samples was 0.372, 0.271, 0.330, 2.136, 0.330, and 0.141%, respectively. It can be seen from Table 4 that the treatment T2 (i.e., coating with 0.2% chitosan solution) produced the highest amounts of different elements (viz. Ca at 5 DAPS, Mg at 10 DAPS, Na at both DAPS, K at 10 DAPS, P at 5 DAPS, and S at both DAPS) in beans. According to Khazaei and Vandenberg (2020), fava beans contain 0.093-0.103% Ca, 0.123-0.148% Mg, 0.002-0.016% Na, 1.084-1.206% K, 0.434-0.606% P and 0.183-0.205% S. However, a few things needed to be more consistent in the mineral element content of the country bean that was used for analysis in the present study, which may be connected to sample size, maturity level, and number of seeds in the bean.

However, there were comparatively higher amounts of minerals except for Na and S in the country bean samples after postharvest storage, although the differences in most cases were insignificant (Table 4). Similar observation was also reported by Sultana et al. (2019) for postharvest storage of tomato. However, they could not provide any information as to why the mineral content of tomato fruits increased during storage. On the other hand, according to Youwei and Yinzhe (2013), the respiration rate, the amount of free radicals, and the level of disease resistance all dropped when the surface of the beans was coated with chitosan. As a result, the majority of nutrients are conserved to the maximum.

Table 4. The impact of chitosan treatments on concentration (± SD) of different mineral elements in the country beans when stored at normal temperature (≈23-25°C).

Treatment	Ca (%)		Mg (%)		Na (%)		K (%)		P (%)		S (%)	
	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS	5 DAPS	10 DAPS
T0		0.586	0.421	0.478	0.496	0.496	2.215	2.109	0.317b	0.341	0.147	0.141
T1	0.699a	0.476	0.408	0.517	0.605	0.491	2.521	2.124	0.447a	0.339	0.183	0.146
T2	0.773a	0.484	0.377	0.557	0.632	0.592	2.344	2.115	0.509a	0.342	0.163	0.137
T3	0.533b	0.488	0.461	0.421	0.589	0.495	2.262	1.995	0.455a	0.312	0.139	0.112
$LSD_{0.05}$	0.11	0.15	0.07	0.27	0.13	0.11	0.37	0.25	0.11	0.09	0.04	0.04
SE (±)	0.033	0.046	0.022	0.083	0.041	0.032	0.110	0.080	0.034	0.028	0.011	0.012
Level of significance	**	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS
CV%	9.45	15.61	9.30	29.14	12.14	10.82	8.38	6.37	13.57	14.46	12.21	15.31
Fresh bean (0 DAPS)	0.372 ± 0.026		0.271 ± 0.011		0.330 ± 0.058		2.136 ± 0.173		0.330 ± 0.035		0.141 ± 0.017	

Treatment T0= Control (no coating solution); T1= coating with 0.10% solution; T2= coating with 0.20% solution and T3= coating with 0.30% solution. ** indicating significant at $P < 0.01$, * indicating significant at $P < 0.05$, and NS means non-significant.

Correlations among the nutritional qualities of country beans

Pearson correlation coefficients among the nutritional qualities of country beans harvested at 5 and 10 DAPS are summarized in Tables 5 and 6, respectively. Total phenol contents of beans were positively correlated to Mg (r = 0.545) at 5 DAPS, while it showed highly significant positive relationships to Mg, K, and S (r = 0.791, 0.950, and 0.820, respectively) at 10 DAPS. But among the nutrient elements, Ca and P demonstrated highly significant negative relationships (r = -0.936 and -0.963, respectively) to the total phenol contents of beans. On the other hand, total phenol contents of beans were positively correlated to chlorophyll contents at 5 DAPS but negatively correlated to the same at 10 DAPS (Tables 5 and 6). Thus, it can be inferred that total phenol contents in beans negatively affected chlorophyll contents at a longer storage period. This might be due to the increase of total phenolic content at longer storage (Table 3) for all chitosan treatments, and lower efficacy of chitosan film with higher storage time. Thus, increases microbial stressful situations, which accelerates the internal production of natural defense substances, like polyphenols. The study also revealed highly significant positive correlations between each other of different types of chlorophyll contents (chlorophyll-a, chlorophyll-b, and total chlorophyll), which indicated that they were strongly associated with each other, and chitosan coating has the ability to boost chlorophyll content in stressful environmental conditions. Chitosan has also been identified by Katiyar et al. (2015) as a natural bioactive chemical to improve prospective physiological changes in crops. Furthermore, different types of chlorophylls also showed significant positive correlations to Na and P at both DAPS (Tables 5 and 6). Among the mineral elements, Na, K, P, and S were positively correlated with Ca (r = 0.900, 0.634, 0.838, and 0.834, respectively), and K and P showed the same relationships with Na (r = 0.548 and 0.979, respectively) at 5 DAPS (Table 5). Similarly, Mg demonstrated significant positive relationships with Na, K and S (r = 0.589, 0.921 and 0.976, respectively); Ca with P (r = 0.996), and K with S (r = 0.956) at 10 DAPS (Table 6). On the other hand, at 10 DAPS, Mg, K and S were negatively correlated with Ca (r = -0.935, -0.999, and -0.968, respectively), and Mg, K, and S exhibited the same correlations with P (r = -0.906, -0.999 and -0.943). However, such inverse relationships indicate that the content of these mineral elements is moved in the reverse direction, i.e., when the content of any one of these elements is increased, the other is decreased with the same magnitude and vice-versa.

Table 5. Pearson correlation coefficients for nutritional qualities of country beans collected at 5 days after postharvest storage (n=12).

	Total phenol	Chl-a	Chl-b	T-Chl	Ca	Mg	Na	K	P
Chl-a	0.813**								
Chl-b	0.821**	0.998**							
T-Chl	0.818**	0.999**	1.000**						
Ca	-0.128 ns	0.417 ns	0.377 ns	0.392 ns					
Mg	0.545*	0.230 ns	0.286 ns	0.266 ns	-0.719**				
Na	0.273 ns	0.765**	0.740**	0.749**	0.900**	-0.369 ns			
K	-0.463 ns	0.097 ns	0.103 ns	0.101 ns	0.634*	-0.280 ns	0.548*		
P	0.430 ns	0.838**	0.810**	0.821**	0.838**	-0.323 ns	0.979**	0.365 ns	
S	-0.530 ns	-0.113 ns	-0.164 ns	-0.145 ns	0.834**	-0.978**	0.518 ns	0.458 ns	0.446 ns

Notes: Chl = Chlorophyll; T-Chl = Total chlorophyll; ns non-significant; * significant at $P < 0.05$ and ** significant at $P < 0.01$

Table 6. Pearson correlation coefficients for nutritional qualities of country beans collected at 10 days after postharvest storage (n=12).

	Total phenol	Chl-a	Chl-b	T-Chl	Ca	Mg	Na	K	P
Chl-a	-0.605*								
Chl-b	-0.701**	0.991**							
T-Chl	-0.703**	0.991**	1.000**						
Ca	-0.936**	0.458 ns	0.573*	0.571*					
Mg	0.791**	-0.116 ns	-0.247 ns	-0.244 ns	-0.935**				
Na	0.228 ns	0.635*	0.534*	0.532*	-0.319 ns	0.589*			
K	0.950**	-0.486 ns	-0.599*	-0.597*	-0.999**	0.921**	0.301 ns		
P	-0.963**	0.512 ns	0.623*	0.621*	0.996**	-0.906**	-0.284 ns	-0.999**	
S	0.820**	-0.286 ns	-0.408 ns	-0.403 ns	-0.968**	0.976**	0.402 ns	0.956**	-0.943**

Notes: Chl = Chlorophyll; T-Chl = Total chlorophyll; ns non-significant; * significant at $P < 0.05$ and ** significant at $P < 0.01$

CONCLUSION

The application of chitosan coating at different doses showed a remarkable positive effect to preserve weight loss and increase the shelf-life of country beans. Similarly, chitosan coatings also enhance different types of chlorophyll contents in the country beans during postharvest storage. Furthermore, the study found that chitosan coverings have the ability to enhance some mineral nutritional aspects. However, total phenolic contents in the country beans decreased considerably at storage, and they were not able to be protected by the chitosan application. Finally, this research found that chitosan covered with a 0.20 percent solution is suitable to maintain the nutritional values of country beans while also preventing weight loss and extending shelf-life. The effect of chitosan application on the contents of different mineral elements of fruits and vegetables is limited, and the cause of the observed phenomena is still unclear. Thus, a more thorough and critical quantitative investigation should be conducted, taking into account all factors to determine the actual status of mineral elements during postharvest storage of chitosan-treated fruits and vegetables, which will ultimately contribute to improved farming techniques and nutritional standards.

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