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Influence of different concentrations of nitric oxide on fruit quality of sweet pepper and mango under mixed loading conditions

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Abstract

In this study, mango fruits (*Mangifera indica* L.) were stored together with sweet peppers to simulate mixed load shipping conditions. Sheets of Nitric oxide with different concentrations (40, 60 and 80 ml/l) were placed in mango packages. Sets with different treatments of treated and or untreated (control) mango fruits were placed together with sweet peppers, then each of treatments was kept separated in cold-storage rooms at 10 °C + 90% RH, for 35 days. Samples from mangos and sweet peppers were examined at 7 days' intervals for physical and chemical quality parameters. For both mango fruits and sweet peppers nitric oxide at 60 ml/l treatment showed a significant reduction of weight loss and decay percentages, and maintained general appearance, fruit firmness, total soluble solids (TSS), ascorbic acid content, and total sugars. Total chlorophyll also was steadily maintained. Hence nitric oxide at 60 ml/l significantly proved to be a potential treatment to delay ripening and keeping better overall quality attributes of both mango and sweet peppers fruits stored together as compared to other treatments and control under cold storage conditions.

Keywords: general appearance; mango; mixed loading; nitric oxide; sweet pepper

Introduction

Sweet pepper (*Capsicum annuum*, L.) is one of the most common vegetables in the world and is well known for its nutritional values It contains high content of vitamin c, which is a crucial nutrient in the human diet. During post-harvest, capsicums are susceptible to chilling injury if stored below 7 °C for long periods, Nevertheless, storage period for pepper about 28 days at 7.5 °C and 14 days when stored at 5 °C also highly sensitive to diseases (Cantwell, 2013). Since sweet pepper is a non-climacteric fruits, its senescence is mainly accelerated by water loss during respiration. González-Gordo *et al*. (2019) stated that the exogenous application of Nitric Oxide (NO) started to be used in research, mainly for extending the post-harvest stage of both climacteric and non-climacteric fruits. They also mentioned that fruits fumigation with NO could extend their

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post-harvest life and maintain their qualities. In addition, they reported that results of NO application on sweet peppers indicated that NO governs fruit ripening and exerts an anti-maturation effect.

Mango (*Mangifera indica* L.) has a pleasant aroma and unique flavor and contains several of nutrients. It gradually developed into the second most important grown and consumed tropical fruit in the world (Ke *et al*., 2022). The global production of mango is approximately 54.83 million tons per planted area exceeding 55 million hectares in 2020 according to the data from the FAO. Consumption form of mango has gradually transformed from local sales to export sales modes. However, over extended periods of cold chain transportation, deterioration and decay in the quality of mango caused by ripening during storage and transportation often occur, resulting in high losses. Mango also is a climacteric fruit during the ripening with increase in respiration intensity accompanied by the rapid synthesis of ethylene (Ding and Zhang, 2021).

The homeostasis of nitric oxide in fruits and vegetables is maintained through regulation of degradation and its synthesis. Nitric oxide can be synthesized via the reductive pathway and oxidative pathway (Dean and Harper, 1988; Yamasaki and Sakihama, 2000). Nitrite reduction is the major source of nitric oxide in fruits, which occurs by both enzymatic and non-enzymatic mechanisms (Salgado *et al*., 2013). Nitrate reductase in the cytosol, nitrite reductase in the plasma membrane, nitrate reductase in plastids, and xanthine oxidoreductase are involved in reductive nitric oxide synthesis (Rockel *et al*., 2002; He and He, 2020). Many researches oxidative pathways of nitric oxide synthesis have been studied.

Nitric oxide plays an important role in quality changes of postharvest vegetables and fruits by delaying senescence or ripening, controlling postharvest diseases, inhibiting browning, and alleviating chilling injury.

These effects are related to the ability of nitric oxide to increase antioxidant enzyme activity inhibit exogenous ethylene synthesis, inhibit exogenous ethylene synthesis, increase the accumulation of antimicrobial substances, activate antimicrobial enzymes, and maintain membrane integrity and a high energy level. However, it is still unknown how nitric oxide affects these enzymes' activities and the involved modifications. In addition, nitric oxide synthase and its action mechanism in plants needs to be further studied. Importantly, the effectiveness of nitric oxide treatment on preserving the quality depends on the species of fruits and vegetables, the concentration, and form (e.g., liquid or gaseous) of nitric oxide applied (Chaki *et al*., 2015; Rodriguez-Ruiza *et al*., 2019; Gonzalez-Gordo *et al*., 2019; Shi *et al*., 2019; Li *et al*., 2022; Liu *et al*., 2023).

The present study aims to study the effect of using environmentally safe different concentrations of Nitric oxide sheets on the storage duration, and quality parameters of sweet pepper mixed load with mango fruits and its effect on fruits quality parameters of sweet pepper and mango during Cold storage conditions.

Materials and Methods

In the current study, sweet pepper (*Capsicum annuum* L. cv. 'Casiano F1 hybrid') and mango (*Mangifera indica* L. cv. 'Keitt') fruits were obtained from a private farm located at Wadi Natrun, Elbehira Governorate, Egypt. during the two successive seasons of 2020 and 2021. Fruits were harvested at the commercial maturity stage and transported immediately to the laboratory.

Fruits of mango and sweet peppers were then sorted and selected for uniformity of size, weight, and absence of visible pathological infections or mechanical damage. The selected fruits were packed separately in carton boxes. Different concentrations of nitric oxide were applied at (40, 60 and 80 ml/l) as sheets and were placed on mango cartons, mango fruits without treatments served as control, then each treatment was triplicated. Sets of Fruits packages contain sweet peppers and mango treated and or untreated were then stored for 35 days at 10±1 °C with a relative humidity (RH) of 90%. Fruits were periodically assessed for physical and chemical quality attributes at day 0 and at intervals of 7 days throughout all the storage period.

Quality assessments of fruits by physical characteristics

Weight loss percentage:

Samples of each treatment were weighed at weekly intervals until the end of experiment. Weight loss (%) was calculated as follows: Weight loss% = [(Initial weight - weight of fruits at sampling date)/Initial weight of fruits] x 100.

Sensorial overall quality attributes

Sensory evaluation was performed during storage at 7-day intervals. Panel members were requested to assess fresh quality measurement as follows:

1. Visual quality (general appearance): based on overall visual appearance, it was evaluated following a 9-point rating scale where 9, excellent; 7, very good; 5, good (limit of consumer acceptability); 3, fair (limit of usability), and 1, extremely poor (Gorny *et al*., 2002; *Medina et al*., 2012).
2. Decay: estimated visually using scores, as described by Kader *et al*. (1973) on 5-1 scale, with reference points of: 5, severe; 4, moderately severe; 3, moderate; 2, slight; 1, none. The score attribution depends on morphological effects such as color change, microorganism effects, smell and decay percentage on fruits.

Firmness:

Fruits from each replicate were taken at weekly intervals and changes in fruit firmness was measured in kg/cm^2 by digital force Gauge model FGV 50 A, Shimpo Instrument Co, Japan, with total capacity of 20 kg/cm^2 and resolution of 0.01 kg/cm^2 using cone pointed head

Quality assessments of fruits by chemical characteristics

Total soluble solids percentage (TSS)

TSS percentage was determined as a composite juice sample by digital refractometer of model Abbe Leica according to (A.O.A.C., 2000) and expressed as a percentage.

Ascorbic acid content

Total ascorbic acid was determined using the dye 2, 6-dichloro-phenol indophenols method (A.O.A.C., 2000).

Total sugars (TS)

TS (mg /100g FW) was determined according to Nelson (1974) - Somogyi (1952) Method, were determined colormetrically using spectrophotometer model 6305 UV/visible range with 520 nm wavelength (Sadasivam and Manickam, 2004).

Total chlorophyll (mg/ 100 g)

Total chlorophyll was determined as described in (Shehata *et al*., 2018). In brief, 0.5 g of fresh sample were homogenized with 5 mL dimethyl formamide and kept in the dark in the refrigerator for 48 h. The absorbance was then measured at 470, 647 and 663 nm with a spectrophotometer (model UV-2401 PC, International Equipment Trading LTD. (IET), Milano, Italia).

Statistical analysis

Data of the two seasons were arranged and statistically analysed using Mstatic. The comparison among means of the different treatments was determined by using Duncan's test; the data were tabulated and statistically analysed according to a factorial complete randomized design (Snedecor and Cochran, 1982).

Results

Weight loss

Data in Tables 1a and 1b declare that weight loss percentage of sweet peppers and mango fruits was significantly affected by different concentrations of nitric oxide compared to untreated fruits during storage period.

Data also reveal that, weight loss percentage of sweet pepper and mango often significantly increased as the storage period increased, this was regardless to the treatment.

Mango fruits exposed to nitric oxide 60 ml/l and storage with sweet pepper fruits gave the lowest significant values of weight loss percentage meanwhile the highest value of weight loss percent was recorded in untreated for sweet pepper and mango fruits these results were true in both studied seasons. these results were in harmony with those obtained by, Zaharah and Singh (2011), as they mentioned that nitric oxide reduces the loss of weight of mangos during storage period. The reduction of weight loss rate may be attributed to reduction in respiration rates during storage period; the obtained results of fruit weight loss are in agreement with (Hu *et al*., 2014, Li *et al*., 2022; Liu *et al*., 2023).

With respect to the effect of the interaction between concentrations of nitric oxide and storage period on the loss in weight, data show that there was an increase in weight loss in sweet peppers and mango fruits in all treatment up to the end of storage period although mean values resulted always significantly lower than the control fruits. However, lowest weight loss percentages were obtained in mango fruits exposed to nitric oxide at 60 ml/l stored with sweet peppers at the end of storage period compared to control and treatments in both studied seasons.

Table 1a. Effect of nitric oxide treatments on weight loss%, decay and general appearance (score) of sweet peppers during cold storage

Storage period (days)	Weight loss%																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
7	1.15	n*	0.86	p	0.85	p	0.94	o	0.95	E	1.34	m	1.03	o	0.93	p	1.18	n	1.12	E
14	1.95	j	1.44	l	1.23	m	1.60	k	1.56	D	1.95	j	1.45	l	1.35	m	1.63	k	1.59	D
21	2.85	e	2.35	i	1.95	j	2.53	g	2.42	C	2.87	e	2.35	h	2.05	i	2.54	g	2.45	C
28	3.44	b	2.73	f	2.45	h	2.94	d	2.89	B	3.54	b	2.85	e	2.71	f	3.15	b	3.06	B
35	4.03	a	3.22	c	2.95	d	3.44	b	3.41	A	4.23	a	3.44	c	3.24	d	3.63	b	3.63	A
Mean	2.68	A	2.12	C	1.89	D	2.29	B			2.78	A	2.22	C	2.06	D	2.43	B		
	Decay (score)																			
	First season										Second season									
0	1.00	d	1.00	d	1.00	d	1.00	d	1.00	D	1.00	d	1.00	d	1.00	d	1.00	d	1.00	D
7	1.00	d	1.00	d	1.00	d	1.00	d	1.00	D	1.00	d	1.00	d	1.00	d	1.00	d	1.00	D
14	1.33	cd	1.00	d	1.00	d	1.00	d	1.08	CD	1.67	c	1.00	d	1.00	d	1.00	d	1.17	D
21	1.67	bc	1.00	d	1.00	d	1.33	cd	1.25	C	2.00	bc	1.00	d	1.00	d	1.67	c	1.42	C
28	2.00	ab	1.33	cd	1.00	d	1.67	bc	1.50	B	2.33	b	1.67	c	1.00	d	1.67	c	1.67	B
35	2.33	a	1.67	bc	1.00	d	2.00	ab	1.75	A	3.00	a	2.00	bc	1.00	d	2.33	b	2.08	A
Mean	1.56	A	1.17	BC	1.00	C	1.56	A			1.83	A	1.28	B	1.00	C	1.44	B		
	General appearance (score)																			
	First season										Second season									
0	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A
7	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A
14	8.33	ab	9.00	a	9.00	a	9.00	a	8.83	A	7.67	bc	9.00	a	9.00	a	9.00	a	8.67	AB
21	7.67	bc	9.00	a	9.00	a	9.00	a	8.67	A	7.00	cd	9.00	a	9.00	a	9.00	a	8.50	B
28	7.00	cd	8.33	ab	9.00	a	7.67	bc	8.00	B	6.33	de	8.33	ab	9.00	a	7.00	cd	7.67	C
35	6.33	d	7.67	bc	8.33	ab	7.00	cd	7.33	C	5.67	e	7.00	cd	7.67	bc	6.33	de	6.67	D
Mean	7.89	C	8.67	AB	8.89	A	8.44	B			7.44	C	8.56	AB	8.78	A	8.22	B		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorder in each season, different capital letters refer to mean values)

Table 1b. Effect of nitric oxide treatments on weight loss%, decay and general appearance (score) of mango during cold storage

Storage period (days)	Weight loss%																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
7	2.45	n*	2.32	p	2.14	q	2.37	o	2.32	E	2.62	o	2.41	q	2.24	r	2.46	p	2.43	E
14	3.06	j	2.65	m	2.44	n	2.77	l	2.73	D	3.45	j	2.93	m	2.75	n	3.08	l	3.05	D
21	3.86	f	3.34	i	2.96	k	3.55	h	3.43	C	3.94	f	3.45	j	3.15	k	3.64	h	3.54	C
28	4.64	b	3.74	g	3.35	i	3.91	e	3.91	B	4.66	b	3.82	g	3.55	i	4.03	e	4.02	B
35	5.22	a	4.03	d	3.73	g	4.32	c	4.33	A	5.04	a	4.25	d	3.94	f	4.47	c	4.42	A
Mean	3.85	A	3.22	C	2.92	D	3.38	B			3.94	A	3.37	C	3.12	D	3.54	B		
	Decay score																			
	First season										Second season									
0	1.00	b	1.00	b	1.00	b	1.00	b	1.00	B	1.00	c	1.00	c	1.00	c	1.00	c	1.00	C
7	1.00	b	1.00	b	1.00	b	1.00	b	1.00	B	1.00	c	1.00	c	1.00	c	1.00	c	1.00	C
14	1.00	b	1.00	b	1.00	b	1.00	b	1.00	B	1.00	c	1.00	c	1.00	c	1.00	c	1.00	C
21	1.00	b	1.00	b	1.00	b	1.00	b	1.00	B	1.00	c	1.00	c	1.00	c	1.00	c	1.00	C
28	1.33	b	1.00	b	1.00	b	1.00	b	1.08	B	1.67	b	1.00	c	1.00	c	1.33	bc	1.25	B
35	2.00	a	1.33	b	1.00	b	1.33	b	1.42	A	2.33	a	1.33	bc	1.00	c	1.67	b	1.58	A
Mean	1.22	A	1.06	B	1.00	B	1.06	B			1.33	A	1.06	B	1.00	B	1.17	AB		
	General appearance score																			
	First season										Second season									
0	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A
7	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A
14	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A	9.00	a	9.00	a	9.00	a	9.00	a	9.00	A
21	7.67	bc	9.00	a	9.00	a	9.00	a	8.67	AB	7.00	cd	9.00	a	9.00	a	8.33	ab	8.33	B
28	7.00	cd	9.00	a	9.00	a	8.33	ab	8.33	B	6.33	de	8.33	ab	9.00	a	7.67	bc	7.83	C
35	6.33	d	8.33	ab	9.00	a	7.67	bc	7.83	C	5.67	e	7.00	cd	9.00	a	6.33	de	7.00	D
Mean	8.00	C	8.89	AB	9.00	A	8.67	B			7.67	C	8.56	B	9.00	A	8.22	B		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorder in each season, different capital letters refer to mean values)

Decay

Irrespective to the storage period, data in Tables 1a and 1b reveal clearly that, all examined nitric oxide concentrations recorded significantly lower decay score in comparison to control moreover, concentration of 60 ml/l on mango fruits was the most effective treatment for reducing decay incidence in both seasons on sweet peppers , while, all concentrations of nitric oxide was no significantly affected compared with untreated as control during storage period in reducing mango fruits decay. These results were in accordance with those recorded by (Chaki *et al*., 2015; Corpas, 2015; Rodriguez-Ruiza *et al*., 2019) on sweet peppers, and (Zaharah and Singh, 2011; Barman *et al*., 2014; Hu *et al*., 2014) on mangos fruit. As they reported that Nitric oxide has important effects of slowing maturation, delaying fruit ripening and also maintaining quality during storage period.

Data also clear that the decay of sweet pepper and mango fruits showed a progressive increment as the storage period increased. Li *et al*. (2022) and Liu *et al*. (2023) proposed that the increase in decay incidence during storage, might be attributed to the lowering of chemical and biological activity in fruits which in turn facilitates the infection of fruits by micro- organisms.

Data indicate that, untreated fruits started to show decay symptoms after 14 days of storage for sweet peppers and 21 days for mango fruits and several decay symptoms at the end of storage period were observed, on the other hand mango fruits exposed to nitric oxide 60 ml/l and stored with sweet peppers showed nil visible decay symptoms till end of storage period (35 days) in both investigated seasons.

General appearance

As shown in Tables 1a and 1b general appearance of mango fruits exposed to nitric oxide either at 60 ml/l or 40 ml/l and stored with sweet peppers k, had the best appearance compared to other treatments in both seasons. The application of nitric oxide at different concentrations in the following order, 60 ml/l, 40 ml/l and

80 ml/l, had a high effect on post-harvest quality in terms of maintaining significantly better general appearance comparing to untreated fruits General appearance was maintained by using nitric oxide may have attributed to the effect of nitric oxide on the reduction of weight loss and rot rate of sweet peppers and mango fruits. Nitric oxide concentrations have beneficial effects on fruit physiology such as delaying ripening (Li *et al*., 2022; Liu *et al*., 2023).

General appearance of sweet pepper and mango fruits was affected with the prolongation of storage period in both seasons. This effect might be due to color change, shrivelling and decay, Similar results were reported by (Rodriguez-Ruiza *et al*., 2019) on sweet peppers fruits and on mangos fruit (Hu *et al*., 2014).

sweet pepper and mango fruits treated with nitric oxide at 60 ml/l did not exhibit any changes in their appearance up to 28 and 35 days of storage respectively. On the other hand, untreated sweet paper and mango fruits started to show decline of general appearance at day 21st of the storage and significantly recorded lower scores of appearances at the end of storage period (35 days). These results were true in both studied seasons.

Fruit firmness (kg/cm^2)

Results tabulated in Tables 2a and 2b show that firmness of sweet pepper and mango fruits was significantly affected by different concentrations of nitric oxide during storage period in both seasons. Data showed that various applied concentrations of nitric oxide had significantly higher fruit firmness as compared to untreated ones. However, the highest values of fruit firmness were recorded with mango fruits exposed to nitric oxide 60 ml/l stored together with sweet peppers, followed by fruits exposed to 40 ml/l, while lower values were found in untreated ones. Barman *et al*. (2014) and Hu *et al*. (2014) found that nitric oxide has increased mango fruits susceptibility to texture.

Data also show that there was a significant reduction in firmness of sweet pepper and mango fruits by the progresses of storage period in both seasons. This may due to the loss of cell wall integrity by increasing storage period as a result of breakdown of peptic substances, which led to an increase in soluble pectin and decrease in fruit firmness of pepper fruits (Ilić *et al*., 2012).

The interaction between concentrations and storage period on sweet pepper and mango fruits firmness showed that mango fruits exposed to nitric oxide 60 ml/l and storage with sweet pepper fruits maintained the fruit firmness for 35 days.

Total soluble solids (TSS) contents

Data in Tables 2a and 2b indicate that generally, all concentrations significantly maintained fruit T.S.S comparing to untreated ones. Moreover, mango fruits exposed to nitric oxide 60 ml/l and stored with sweet pepper fruits contained higher content of T.S.S. compared with other treatments in both seasons, followed by fruits exposed to 40 ml/l with significant differences between then, these results are in agreement with those obtained by Hu *et al*. (2014), Chaki *et al*. (2015), Corpas (2015), and Rodriguez-Ruiza *et al*. (2019).

Regarding to storage time data show that T.S.S content were decreased significantly and consistently with prolongation of storage period at cold storage conditions, in both seasons as illustrated by Li *et al*. (2022) and Liu *et al*. (2023).

The interaction between the two studied variables was significant in both seasons. All concentrations at different storage periods had higher T.S.S. content compared with untreated (control) fruits. While mango fruits exposed to nitric oxide 60 ml/l and stored with sweet pepper fruits recorded the highest T.S.S. content in both seasons.

Table 2a. Effect of nitric oxide treatments on firmness (kg/cm²), total soluble solids% and ascorbic acid content (mg/100 ml) of sweet peppers during cold storage

Storage period (days)	Firmness (kg/cm²)																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
0	5.22	a*	5.22	a	5.22	a	5.22	a	5.22	A	5.01	a	5.01	a	5.01	a	5.01	a	5.01	A
7	4.91	c	5.01	bc	5.05	b	5.00	bc	4.99	B	4.62	e	4.76	c	4.81	b	4.72	d	4.73	B
14	4.45	f	4.67	de	4.76	d	4.60	e	4.62	C	4.17	i	4.41	g	4.57	f	4.40	g	4.39	C
21	3.84	i	4.27	g	4.42	f	4.17	g	4.18	D	3.64	m	4.05	j	4.21	h	3.92	k	3.96	D
28	3.22	m	3.84	i	4.03	h	3.71	j	3.70	E	3.03	r	3.63	m	3.86	l	3.43	o	3.49	E
35	3.39	l	3.54	k	3.72	j	3.34	l	3.50	F	2.55	s	3.33	p	3.54	n	3.17	q	3.15	F
Mean	4.17	D	4.42	B	4.54	A	4.34	C			3.84	D	4.20	B	4.33	A	4.11	C		
	Total soluble solids%																			
	First season										Second season									
0	7.28	a	7.28	a	7.28	a	7.28	a	7.28	A	7.04	a	7.04	a	7.04	a	7.04	a	7.04	A
7	7.11	c	7.13	c	7.17	b	7.11	c	7.13	B	6.65	d	6.71	c	6.83	d	6.70	c	6.72	B
14	6.87	f	6.93	e	7.01	d	6.87	f	6.92	C	6.24	i	6.47	f	6.60	e	6.33	h	6.41	C
21	6.54	i	6.63	h	6.74	g	6.62	h	6.63	D	6.03	m	6.21	j	6.37	g	6.13	k	6.18	D
28	6.11	n	6.33	k	6.43	j	6.25	l	6.28	E	5.64	p	5.83	n	6.08	l	5.75	o	5.82	E
35	5.81	q	6.03	o	6.15	m	5.91	p	5.98	F	5.24	s	5.54	q	5.76	o	5.40	r	5.49	F
Mean	6.62	D	6.72	B	6.80	A	6.67	C			6.14	D	6.30	B	6.45	A	6.23	C		
	Ascorbic acid (mg/100 ml)																			
	First season										Second season									
0	105.1	a	105.1	a	105.1	a	105.1	a	105.1	A	103.9	a	103.9	a	103.9	a	103.9	a	103.9	A
7	101.6	e	102.9	c	103.0	b	102.5	d	102.5	B	99.2	f	101.2	c	102.0	b	100.1	d	100.7	B
14	97.8	i	99.0	g	100.2	f	98.8	h	99.0	C	85.3	r	98.1	g	99.5	e	87.0	p	93.8	C
21	92.2	m	95.1	k	96.6	j	94.0	l	94.5	D	91.1	l	94.7	i	96.1	h	93.2	j	92.5	D
28	84.1	t	89.4	o	91.8	n	87.1	r	88.1	E	87.5	o	90.2	m	92.1	k	89.3	n	89.8	E
35	81.1	u	87.2	q	88.1	p	85.1	s	85.4	F	82.2	t	86.1	q	87.5	o	85.0	s	85.2	F
Mean	93.7	D	96.5	B	97.5	A	95.5	C			91.5	D	95.7	B	96.8	A	93.1	C		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at $P \leq 0.05$ (small letters refer to values recorder in each season, different capital letters refer to mean values)

Ascorbic acid content (mg /100 g FW)

Data in Tables 2a and 2b show that there were significant differences between the different concentrations of nitric oxide during storage period in both seasons. Data showed that fruits treated with various concentrations of nitric oxide resulted in significantly greater ascorbic acid content as compared with untreated fruits. However, the highest significant values of ascorbic acid content were obtained with mango fruits exposed to nitric oxide 60 ml/l and stored with sweet pepper. followed by mango fruits exposed to nitric oxide 40 ml/l and stored with sweet pepper., in the meantime, the lowest values were found in untreated fruits. These results were in agreement with those obtained by (Rockel *et al*., 2002; Zaharah and Singh, 2011; He and He, 2020).

Data also declare that there was a significant decrease in ascorbic acid content along with the increase of storage period for all treatments and this was true in both studied seasons.

Wills *et al*. (1981) attributed the reduction of vitamin C during storage to great metabolic activity during storage as it is respired.

Mango fruits exposed to nitric oxide 60 ml/l and stored with sweet peppers maintained the highest ascorbic acid content at 35 days of storage.

Table 2b. Effect of nitric oxide treatments on firmness (kg/cm²), total soluble solids% and ascorbic acid content (mg/100 ml) of mango fruits during cold storage

Storage period (days)	Firmness (kg/cm²)																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
0	10.93	a*	10.93	a	10.93	a	10.93	a	10.93	A	10.56	a	10.56	a	10.56	a	10.56	a	10.56	A
7	9.82	f	10.23	c	10.43	b	10.11	d	10.15	B	9.34	f	9.89	c	10.03	b	9.77	d	9.76	B
14	9.31	i	9.83	f	10.02	e	9.74	g	9.73	C	8.82	i	9.24	g	9.42	e	9.11	h	9.15	C
21	8.62	m	9.20	j	9.44	h	9.05	k	9.08	D	7.77	n	8.54	j	8.84	i	8.33	k	8.37	D
28	7.73	q	8.51	n	8.73	l	8.33	o	8.33	E	6.63	r	7.83	m	8.14	l	7.63	o	7.56	E
35	6.55	s	7.74	q	8.05	p	7.46	r	7.45	F	5.82	s	7.24	p	7.63	o	7.03	q	6.93	F
Mean	8.83	D	9.41	B	9.60	A	9.27	C			8.16	D	8.88	B	9.10	A	8.74	C		
	Total soluble solids%																			
	First season										Second season									
0	12.36	t	12.36	t	12.36	t	12.36	t	12.36	F	12.68	l	12.68	l	12.68	l	12.68	l	12.68	E
7	13.05	s	13.74	q	14.13	o	13.43	r	13.59	E	13.15	kl	13.92	ij	14.54	gh	13.64	jk	13.81	D
14	13.86	p	14.84	l	15.33	i	14.33	n	14.59	D	13.77	ij	14.72	gh	15.31	ef	14.24	hi	14.51	C
21	14.53	m	15.42	h	16.06	e	15.05	k	15.26	C	14.23	hi	14.93	efg	16.13	c	14.54	gh	14.96	B
28	15.14	j	16.04	e	16.95	b	15.64	g	15.94	B	14.85	fg	17.83	a	17.22	b	15.34	ef	16.31	A
35	15.91	f	16.84	c	17.85	a	16.34	d	16.74	A	15.45	de	16.32	c	18.31	a	15.93	cd	16.50	A
Mean	14.14	D	14.87	B	15.45	A	14.53	C			14.02	D	15.07	B	15.70	A	14.39	C		
	Ascorbic acid (mg/100 ml)																			
	First season										Second season									
0	48.52	a	48.52	a	48.52	a	48.52	a	48.52	A	45.33	a	45.33	a	45.33	a	45.33	a	45.33	A
7	46.73	e	47.51	c	47.95	b	47.35	d	47.38	B	41.75	e	42.53	c	43.05	b	42.14	d	42.37	B
14	42.63	j	44.33	g	45.03	f	44.02	h	44.00	C	36.63	k	40.23	g	41.05	f	39.61	h	39.38	C
21	37.92	o	41.22	k	42.72	i	40.83	l	40.67	D	32.14	p	36.84	j	37.93	i	36.03	l	35.74	D
28	33.14	s	38.04	n	39.13	m	37.72	p	37.01	E	28.23	s	32.93	n	34.12	m	32.35	o	31.90	E
35	28.22	u	33.97	r	35.83	q	33.04	t	32.76	F	24.73	u	28.74	r	30.23	q	28.04	t	27.93	F
Mean	39.53	D	42.26	B	43.20	A	41.91	C			34.80	D	37.77	B	38.62	A	37.25	C		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at $P \leq 0.05$ (small letters refer to values recorder in each season, different capital letters refer to mean values)

Total sugars (mg /100 g FW)

It is worthy to note that total sugars content of sweet pepper and mango fruits was significantly affected by nitric oxide treatments at different concentrations Tables 3a and 3b. In the meantime, total sugars content showed a significant increase as the storage period increased recorded the highest significant values at the end of the storage period (35 days) this was regardless the treatments. this was observed in both investigated seasons irrespective of the applied treatments.

Data declared that untreated fruits had significantly higher total sugars than mango fruits exposed to nitric oxide with different concentrations and stored with sweet pepper fruits. Indeed, mango fruits exposed to nitric oxide at 60 ml/l and stored with sweet pepper showed significant less sugar content comparing to fruits exposed to nitric oxide at either 40 or 80 ml/l and stored with sweet pepper. These results are presumably due to the faster maturity in untreated mango fruits as compared with treated ones. These results are in agreement with (Chaki *et al*., 2015; Corpas, 2015; Rodriguez-Ruiza *et al*., 2019) on sweet peppers and (Zaharah and Singh, 2011; Barman *et al*., 2014; Hu *et al*., 2014) on mangos fruits.

The increase in total sugars during storage might owe much to the higher rate moisture loss through transpiration than the rate of dry matter loss through respiration. Also, the reduction in total sugars during storage might owe much to the rate of sugar loss through respiration than water loss through transpiration (Wills *et al*., 1981).

Mango fruits exposed to nitric oxide at 60 ml/l showed the lowest significant values of sugars content at all storage period comparing to other nitric oxide treatments and control. this was recorded in both studied seasons.

All concentrations at different storage periods had lowest total sugars content compared with untreated fruits (control, however it should be noted that when mango fruits exposed for 60 ml/l of nitric oxide and stored with sweet peppers, resulted in the lowest total sugars content in mango and sweet pepper in both seasons, comparing with other treatments and control. These results go in line with findings obtained by Trung *et al*. (2011), who showed that the total sugar content in the control fruit in apple was found to increase faster than in treated fruits.

Table 3a. Effect of nitric oxide treatments on total sugars (mg /100 g FW) and total chlorophyll (mg/ 100 g) of sweet peppers during cold storage

Storage period (days)	Total sugars (mg /100g FW)																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
0	4.56	r*	4.56	r	4.56	r	4.56	r	4.56	F	4.41	t	4.41	t	4.41	t	4.41	t	4.41	F
7	4.99	n	4.81	p	4.73	q	4.87	o	4.85	E	5.25	n	4.75	r	4.68	s	4.87	q	4.89	E
14	5.56	j	5.07	m	4.95	n	5.20	l	5.20	D	5.71	k	5.12	o	4.98	p	5.28	n	5.27	D
21	5.93	g	5.53	j	5.34	k	5.68	i	5.62	C	6.55	d	5.68	l	5.45	m	5.89	j	5.89	C
28	6.43	b	5.93	g	5.74	h	6.11	e	6.05	B	6.85	b	6.13	h	5.93	i	6.33	f	6.31	B
35	6.85	a	6.17	d	6.05	f	6.33	c	6.35	A	7.13	a	6.41	e	6.23	g	6.68	c	6.61	A
Mean	5.72	A	5.34	C	5.23	D	5.46	B			5.98	A	5.42	C	5.28	D	5.58	B		
	Total chlorophyll (mg/ 100 g)																			
	First season										Second season									
0	10.76	a	10.76	a	10.76	a	10.76	a	10.76	A	10.42	a	10.42	a	10.42	a	10.42	a	10.42	A
7	10.24	e	10.44	c	10.51	b	10.40	d	10.40	B	9.75	f	10.08	c	10.18	b	10.01	d	10.00	B
14	9.52	j	9.93	g	10.14	f	9.82	h	9.86	C	9.26	j	9.71	g	9.82	e	9.54	h	9.58	C
21	8.93	o	9.44	k	9.73	i	9.21	m	9.33	D	8.43	n	9.07	k	9.33	i	8.84	l	8.92	D
28	8.42	s	9.10	n	9.33	l	8.87	p	8.93	E	8.03	p	8.64	m	8.85	l	8.41	n	8.48	E
35	7.85	u	8.55	r	8.84	q	8.23	t	8.37	F	7.32	r	8.01	p	8.24	o	7.85	q	7.85	F
Mean	9.29	D	9.71	B	9.89	A	9.55	C			8.87	D	9.32	B	9.47	A	9.18	C		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorder in each season, different capital letters refer to mean values)

Table (3b). Effect of nitric oxide treatments on total sugars (mg /100g FW) and total chlorophyll (mg/ 100 g) of mango fruits during cold storage

Storage period (days)	Total sugars (mg /100 g FW)																			
	First season										Second season									
	Treat.																			
	Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean		Control		Nitric oxide 40 ml/l		Nitric oxide 60 ml/l		Nitric oxide 80 ml/l		Mean	
0	6.95	t*	6.95	t	6.95	t	6.95	t	6.95	F	7.63	s	7.63	s	7.63	s	7.63	s	7.63	F
7	9.05	o	7.83	r	7.34	s	8.13	q	8.09	E	10.13	n	9.13	q	8.72	r	9.54	p	9.38	E
14	10.93	i	9.24	n	8.53	p	9.73	l	9.61	D	11.54	j	10.43	m	9.63	o	10.94	k	10.63	D
21	12.35	e	10.34	k	9.62	m	10.93	i	10.81	C	13.75	d	11.52	j	10.73	l	12.05	h	12.01	C
28	13.96	b	11.03	h	10.83	j	12.15	f	11.99	B	14.63	b	12.45	g	11.63	i	13.14	f	12.96	B
35	15.25	a	12.43	d	11.64	g	13.34	c	13.16	A	15.95	a	13.23	e	12.43	g	13.94	c	13.89	A
Mean	11.42	A	9.64	C	9.15	D	10.20	B			12.27	A	10.73	C	10.13	D	11.21	B		
	Total chlorophyll (mg/ 100 g)																			
	First season										Second season									
0	54.12	a	54.12	a	54.12	a	54.12	a	54.12	A	50.38	a	50.38	a	50.38	a	50.38	a	50.38	A
7	48.71	e	50.73	c	51.31	b	50.11	d	50.22	B	41.83	e	45.14	c	46.23	b	44.61	d	44.45	B
14	41.64	k	45.31	g	47.52	f	44.63	h	44.77	C	35.21	j	40.33	g	41.62	f	39.12	h	39.07	C
21	37.07	o	42.02	j	43.12	i	41.51	l	40.93	D	30.53	n	34.92	k	36.14	i	33.85	l	33.86	D
28	32.36	q	37.82	n	39.22	m	37.05	o	36.61	E	25.12	t	30.22	o	31.32	m	29.07	p	28.93	E
35	27.32	t	32.25	r	34.18	p	31.71	s	31.36	F	22.04	u	26.12	r	27.21	q	25.33	s	25.18	F
Mean	40.20	D	43.71	B	44.91	A	43.19	C			34.18	D	37.85	B	38.82	A	37.06	C		

*Note: Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorder in each season, different capital letters refer to mean values)

Chlorophyll content

Data in Tables 3a and 3b revealed that chlorophyll contents were markedly decreased while the storage period increased this was regardless of the treatments. This decrement in chlorophyll could be attributed to the destruction by chlorophyllase activity and turn of chloroplasts to chromoplasts these results agreement with Shehata *et al*. (2018) on sweet peppers. As for the application of nitric oxide data illustrated that chlorophyll content was significantly influenced by all treatments since greater contents were kept as compared with control, moreover, higher levels of chlorophyll were observed with nitric oxide applied at 60 ml/l followed by 40 ml/l and 80 ml/l respectively showing significant differences

It should be noted that the lowest significant values of chlorophyll contents were observed with control at 35 days of cold storage in both mango and sweet peppers. Conversely the application of nitric oxide at 60 ml/l recorded significantly the highest content of chlorophyll at the same storage interval. These results were true in the two seasons and were in agreement with (Li *et al*., 2022; Liu *et al*., 2023).

Conclusions

The effect of the nitric oxide sheets applied on mango fruits with different concentrations (40, 60 and 80 ml/l), when stored together with sweet peppers was investigated in this study. Nitric oxide treatments markedly showed a positive effect on delaying ripening of both stored mango and sweet peppers compared to control. In particular, nitric oxide at 60 ml/l treatment significantly showed a reduction of weight loss and decay percentage, in the meantime, maintained general appearance, fruit firmness, total soluble solids (TSS), ascorbic acid, chlorophyll and total sugars contents. Nitric oxide at recommended concentration (60 ml/l), proved to be a favourable for extending storage life for both commodities in a mixed load conditions.

Authors' Contributions

All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

A.O.A.C. (2000). International Official methods of analysis of AOAC International. 17th edition. Gaithersburg, MD, USA, Association of Analytical Communities.

Barman K, Asrey R, Pal RK, Jha SK, Bhatia K (2014). Post-harvest nitric oxide treatment reduces chilling injury and enhances the shelf-life of mango (*Mangifera indica* L) fruit during low-temperature storage. The Journal of Horticultural Science and Biotechnology 89(3):253-260.

Cantwell M (2013). Bell pepper. Recommendations for maintaining postharvest quality. Postharvest Technology Research and Information Center, University of California, Davis, USA.

Chaki M, Morales PA, Ruiz C, Begara-Morales JC, Barroso JB, Corpas FJ, Palma JM (2015). Ripening of pepper (*Capsicum annuum*) fruit is characterized by an enhancement of protein tyrosine nitration. Annals of Botany 116:637-647. *https://doi.org/10.1093/aob/mcv016*

Corpas FJ (2015). What is the role of hydrogen peroxide in plant peroxisomes? Plant Biology (Stuttg) 17(6):1099-1103. *https://doi.org/10.1111/plb.12376*

Dean JV, Harper JE (1988). The conversion of nitrite to nitrogen oxide(s) by the constitutive NAD (P)H-nitrate reductase enzyme from soybean. Plant Physiology 88(2):389-395. *https://doi.org/10.1104/pp.88.2.389*

Ding W, Zhang Q (2021). Mango post-harvest treatment and storage preservation. China Food Safety 24:34-35. (In Chinese with English Abstract).

Gonzalez-Gordo S, Bautista R, Claros MG, Caas A, Palma JM, Corpas FJ (2019). Nitric oxide-dependent regulation of sweet pepper fruit ripening. Journal of Experimental Botany 70(17):4557-4570. *https://doi.org/10.1093/jxb/erz136*

Gorny JR, Hess-Pierce B, Cifuentes RA, Kader AA (2002). Quality changes in fresh-cut pear slices as affected by controlled atmospheres and chemical preservatives. Postharvest Biology Technology 24(3):271-278. *https://doi.org/10.1016/S0925-5214(01)00139-9*

He HY, He LF (2020). Crosstalk between melatonin and nitric oxide in plant development and stress responses. Physiologia Plantarum 170(2):218-226. *https://doi.org/10.1111/ppl.13143*

Hu M, Yang D, Huber DJ, Jiang Y, Li M, Gao Z, Zhang Z (2014). Reduction of postharvest anthracnose and enhancement of disease resistance in ripening mango fruit by nitric oxide treatment. Postharvest Biology and Technology 97:115-122. *https://doi.org/10.1016/j.postharvbio.2014.06.013*

Ilic ZS, Radmila T, Rados P, Sharon AT, Yaacov P, Elazar F. (2012). Effect of heat treatment and individual shrink packaging on quality and nutrition value of bell pepper stored at suboptimal temperature. International Journal of Food Science and Technology 47(1):83-90. *https://doi.org/10.1111/j.1365-2621.2011.02810.x*

Kader AA, Lipton WJ, Morris LL (1973). System for scoring quality of harvested lettuce. HortScience (8):408-409. *https://api.semanticscholar.org/CorpusID:89728598*

Ke Y, Dai T, Xiao M, Chen M, Liang R, Liu W, … Deng L (2022). Industry-scale microfluidizer system produced whole mango juice: Effect on the physical properties, microstructure and pectin properties. Innovative Food Science and Emerging Technologies 75(2):102887. *https://doi.org/10.1016/j.ifset.2021.102887*

Li C, Yu W, Liao W (2022). Role of nitric oxide in postharvest senescence of fruits. International Journal of Molecular Sciences 23(17):10046. *https://doi.org/10.3390/ijms231710046*

Liu Y, Chen T, Tao N, Yan T, Wang Q, Li Q (2023). Nitric oxide is essential to keep the postharvest quality of fruits and vegetables. Horticulturae 9(2):135. *https://doi.org/10.3390/horticulturae9020135*

Medina MS, Tudela JA, Jorge JA, Marin A, Allende A, Gil MI (2012). Short postharvest storage under low relative humidity improves quality and shelf life of minimally processed baby spinach (*Spinacia oleracea* L) Postharvest Biology and Technology 67:1-9. *https://doi.org/10.1016/j.postharvbio.2011.12.002*

Nelson N (1974). A photometric adaptation of the Somogyi methods for the determination of glucose. Journal of Biological Chemistry 153(2):375-380. *https://doi.org/10.1016/S0021-9258(18)71980-7*

Rockel P, Strube F, Rockel A, Wildt J, Kaiser WM (2002). Regulation of nitric oxide (NO) production by plant nitrate reductase *in vivo* and *in vitro*. Journal of Experimental Botany 53(366):103-110. *https://doi.org/10.1093/jexbot/53.366.103*

Rodriguez-Ruiza M, Mateos RM, Codesido V, Corpas FJ, Palma JM (2019). Characterization of the galactono-1, 4-lactone dehydrogenase from pepper fruits and its modulation in the ascorbate biosynthesis. Role of nitric oxide. Redox Biology 12:171-181. *https://doi.org/10.1016/j.redox.2017.02.009*

Sadasivam S, Manickam A (2004). Biochemical Methods. 2nd edition. New Age International (P) Ltd. Publishers, New Delhi.

Salgado I, Martinez MC, Oliveira HC, Frungillo L (2013). Nitric oxide signaling and homeostasis in plants: A focus on nitrate reductase and S-nitrosoglutathione reductase in stress-related responses. Brazilian Journal of Botany 36(2):89-98. *https://doi.org/10.1007/s40415-013-0013-6*

Shehata SA, Elmogy M, Mohamed HFY (2018). Postharvest quality and nutrient contents of long sweet pepper enhanced by supplementary potassium foliar application. International Journal of Vegetable Science 25(2):196-209. *https://doi.org/10.1080/19315260.2018.1523816*

Shi KK, Liu ZC, Wang JW, Zhu SH, Huang DD (2019). Nitric oxide modulates sugar metabolism and maintains the quality of red raspberry during storage. Scientia Horticulturae 256:108611. *https://doi.org/10.1016/j.scienta.2019.108611*

Snedecor CW, Cochran WG (1982). Statistical Methods. 7th Ed. The Iowa State Univ. Press. Ames. Iowa, USA, pp 325-330.

Somogyi M (1952). Noted on sugar determination. The Journal of Biological Chemistry 195(1):19-23. *https://doi.org/10.1016/S0021-9258(19)50870-5*

Trung TS, Phuong NTH, Stevens WF (2011). Protective effect of chitosan coating and polyethylene film wrapping on postharvest storage of sugar-apples. Asian Journal of Food and Agro-Industry 4(02):81-90.

Wills HHR, Lee THD, Graham WB, McGlasson WB, Hall EG (1981). An introduction in the physiology and handling of fruits and vegetables. New South Wales University Prees Limited Australia, pp 123-126.

Yamasaki H, Sakihama Y (2000). Simultaneous production of nitric oxide and peroxynitrite by plant nitrate reductase: In vitro evidence for the NR-dependent formation of active nitrogen species. FEBS Letters 468(1):89-92. *https://doi.org/10.1016/S0014-5793(00)01203-5*

Zaharah SS, Singh Z (2011). Mode of action of nitric oxide in inhibiting ethylene biosynthesis and fruit softening during ripening and cool storage of 'Kensington Pride' mango. Postharvest Biology and Technology 62(3):258-266. *https://doi.org/10.1016/j.postharvbio.2011.06.007*