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Original Research Paper

Evaluation of pre-treatments for shelf life extension of minimally processed fenugreek (*Trigonella foenum graecum* L.)

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ABSTRACT

Potential of different pretreatments for shelf life enhancement and nutritional quality maintenance in minimally processed fenugreek (*Trigonella foenum-graecum*) leaves (MPFL) at 8°C storage were evaluated. Pretreatment agents such as calcium propionate, calcium chloride, kinetin, gibberellic acid, 1-methyl cyclo propene, neutral electrolysed water and sodium hypochlorite were used. The results have proven that a five minutes dip in kinetin solution (50 mg L^{-1}) was the best among all tested pretreatments; and it resulted in achieving eight days shelf life compared to four days in control samples. Besides this, better maintenance of total antioxidants (1735 mg ascorbic acid equivalent antioxidant capacity/kg^{-1}) and proteins (44 g/kg^{-1}), at levels close to that of fresh leaves, could be achieved. Calcium propionate (1.5 g L^{-1}) was identified as the second best treatment. It was also shown that minimally processed fenugreek leaves (MPFL) is a rich source of vitamins B_1, B_2, E and K. The B vitamins were susceptible to deterioration during storage, while their loss could be minimized with kinetin treatment. Sanitizers like chlorine (100 mg L^{-1} available chlorine) and neutral electrolysed water (100 and 200 mg L^{-1}) have reduced the shelf life of MPFL indicating their phytotoxicity to FL. The results of the study open up the possibility of using kinetin as an effective pretreatment tool for MPFL in minimal processing industry.

Keywords: Calcium chloride, fenugreek leaves, kinetin, minimal processing, pretreatment, sanitizer

INTRODUCTION

Fenugreek (*Trigonella foenum-graecum* L.) is an annual herb cultivated for its use as a seed spice as well as leafy green vegetable. The plant is cultivated globally, with India being the major producer. Fenugreek seeds and leaves are used as a spice and herb in various cuisines, particularly in Indian, Middle Eastern, and North African dishes. The seeds have a slightly bitter taste and a nutty flavor, while the leaves have a slightly sweet, celery-like taste. Fenugreek leaves, popularly known as '*methi*' in Indian cuisine, is used in curries, stews, and soups for their distinctive flavor. Fresh leaves are incorporated into dough to make traditional Indian breads like '*methi paratha*' and '*methi thepla*'. For leafy green purpose, the plants are uprooted before flowering and sold in loose bundles in local markets (Ahmad et al., 2016, Salam et al., 2023). Similar to other leafy greens, fenugreek leaves (FL) also have very limited shelf life. Minimal processing and packaging using a suitable protocol is an opportunity to add value to FL and to improve shelf life, while retaining its fresh-like nutritional and sensory qualities. Minimally processing refers to the techniques to transform plant-derived foods into convenience foods in the form of ready-to-eat (RTE) or ready-to-cook (RTC) products with an extended shelf life with retention of fresh-like qualities.

Minimally processed leafy greens are susceptible to spoilage due to yellowing, rotting, wilting, loss of flavour, nutritional quality loss etc. These quality deteriorations are summative effects of physiological and microbiological causes in the prepackaged leafy greens. Wound induced responses of the leaf tissues such as increased respiration rate, ethylene accumulation, microbial proliferation due to nutrient leakage, chlorophyll degradation etc. results in the visible manifestation of quality deterioration. (Artes & Allende, 2005). Hence, depending on the inherent nature of the tissues and the intended types of use in cuisines, minimal processing methods vary among the crops.

Pretreatment standardization is an important step for protocol optimization of minimally processed vegetables. Successful pretreatments reported in leafy greens include antimicrobial treatments, texture improving agents, growth hormones, ethylene inhibitors, phytoalexins etc. Along with maintenance of consumer appeal, these pretreatments contribute to the quality retention such as vitamin C, antioxidants and other phytochemicals (Hassan et al., 2012; Ranjitha et al., 2017; Silveira Alexandre et al., 2022). This study was aimed at evaluating the effect of different such pretreatment strategies to find out an optimum pretreatment to enhance shelf life and to understand the associated quality changes in minimally processed fenugreek leaves (MPFL) during storage at commercial chiller temperatures (8^0C) maintained in supermarkets.

MATERIALS AND METHODS

Leaves along with petiole were removed from freshly uprooted fenugreek plants (*Trigonella foenum-graecum* L.), then washed in potable water and excess water was drained off. These leaves were then subjected to different pretreatments based on the earlier reports on improvement of shelf life on various vegetables. Antimicrobial compounds are known to improve both shelf life and safety due to their obvious microbicidal action, while hormones and ethylene inhibitors can act as anti-senescent factors retaining the vitality of plant cells. Calcium salts also known to prolong the freshness of various fresh cut produce (Artes et al., 2005, Hassan et al., 2012; Ranjitha et al., 2017). The following chemical agents were used in the experiment to standardize a suitable pre-treatment for MPFL. This included antimicrobial agents (T_1, T_9, T_{10}), plant hormones (T_2, T_3, T_4, T_5), ethylene inhibitor (T_6, T_7, T_8), and calcium salts (T_{11}, T_{12}). The MPFL samples dipped in potable water served as control (T_{13}).

Pretreatments were carried out by five minutes dip of the washed leaves in the above freshly prepared solutions followed by surface drying. About 100 g leaves were packaged in 50 µm thick LDPE bags with pinholes. The pinholes were made to ensure ventilation. The samples were stored in walk in cold room maintained at 8°C. Each Twelve replications were maintained for each treatment initially, and three replications each were removed on 4th, 6th and 8th day for sampling and biochemical analysis.

Treatment	Pretreatment agent
T_1	Sodium hypochlorite (100 mg L^{-1} available chlorine)
T_2	Gibberellic acid (50 mg L^{-1})
T_3	Gibberellic acid (100 mg L^{-1})
T_4	Kinetin (25 mg L^{-1})
T_5	Kinetin (50 mg L^{-1})
T_6	1- Methyl cyclo propene (1-MCP) (100 µg L^{-1})
T_7	1- MCP (250 µg L^{-1})
T_8	1- MCP (500 µg L^{-1})
T_9	Neutral electrolyzed water (NEW: 100 mg L^{-1} available chlorine)
T_{10}	Neutral electrolyzed water (NEW: 200 mg L^{-1} available chlorine)
T_{11}	Calcium chloride (1.5 g L^{-1})
T_{12}	Calcium propionate (1.5 g L^{-1})
T_{13}	Water (control)

Quality evaluation of packaged MPFL

Sensory evaluation of MPFL was conducted on a 5 point scale based on greenness, fresh leaf odor, texture and overall acceptability (5: excellent, 4: very good, 3: good, 2: average and 1: poor. A score <3 was considered unmarketable). Color values of MPFL were recorded using hand held portable colorimeter (Model: Konica Minolta, CR-10 color reader) at periodic intervals .Physiological loss of weight (PLW) of MPFL were recorded by gravimetric method.

Total chlorophyll and carotenoids were estimated spectrophotometrically by standard AOAC method (AOAC, 1995a; AOAC, 1995b). Antioxidant parameters *viz*., ferric reducing anti-oxidant potential (mg AEAC kg^{-1}), total phenols (mg gallic acid equivalent kg^{-1}), were also measured spectrophotometrically (Benzie & Strain, 1996; Singleton & Rossi, 1965). Total proteins were estimated by Lowry's method (Lowry et al., 1951). Vitamin profiling was done using UPLC-MS/MS method (Santos et al., 2012); and the elution was monitored using a TQDMS/MS (Waters, USA) system.

Statistical analysis

Data obtained were statistically analyzed for single factor analysis of variance (ANOVA) using complete randomized design (CRD) in MS Excel 2007 software, least significant difference values at 99% confidence level was calculated using the standard formula

RESULTS AND DISCUSSION

Considering the requirement of fresh-like characteristics in minimally processed vegetables, sensory evaluation was taken as the benchmark for the shelf life evaluation of MPFL.

Sensory qualities and instrumental colour values

The sensory parameters of stored MPFL were notably influenced by the pretreatments applied (Table 1). Among all parameters for sensory evaluation, the score for the trait 'overall acceptability' is of most importance, as it is a direct indication of the marketability. The overall acceptability score of the control samples were less than 4 within four days of storage. This indicated that the control samples lost their marketability premium within four storage days. Pretreatment with Kinetin at a concentration level of 50 mgL^{-1} (T_5) retained the 'very good' overall acceptability score even after 8 days of storage. Calcium propionate (T12) pretreatment also performed well, ranking highest till six days of storage; but with a sudden drop in acceptability score on 8th day.

The sensory quality deterioration in samples from most of the treatments was mainly in appearance due to colour degradation and texture loss in various samples. This was also supported with data on instrumental evaluation of colour. Hunter's L*, a*, b* values, and total color difference ("E) (Table 2). Samples from T_5 and T_{12} demonstrated statistically superior a* values (indicative of greenness) compared to other treatments. Sodium hypochlorite (T_1) followed by control (T_{13}) showed the highest b* value (indicative of yellowness) at the end of the storage period. The greatest total color difference was observed in T_1 followed by T_5, while T_{12} followed by T_5 showed the least, corroborating the sensory observations regarding colour. It is also clear that sensory and colour qualities deteriorated within four days of storage when MPFL were not given any pretreatments (control samples). Pretreatment with plant hormones, *viz*., kinetin and GA_3 application also resulted in a dose dependent response on MPFL quality during storage. The sensory properties of GA_3 100 mgL^{-1} could be maintained till 6 days.

Table 1 : Effect of pretreatments on the sensory qualities of MPFL during storage at 8°C

Pre-treatment	4 days				6 days				8 days			
	Greenness	Fresh odour	Texture	Overall acceptability	Greenness	Fresh odour	Texture	Overall acceptability	Greenness	Fresh odour	Texture	Overall acceptability
T_1: Chlorine 100 mgL^{-1}	3.1	4.0	3.5	3.5	3.0	2.9	2.9	3.2	2.9	2.5	2.2	2.2
T_2: GA_3 50 mgL^{-1}	3.2	3.5	3.3	3.2	3.5	4.0	2.9	3.5	3.5	3.1	3.0	3.0
T_3: GA_3 100 mgL^{-1}	3.5	4.2	3.2	4.2	4.2	4.0	3.2	4.2	3.4	3.1	3.4	3.2
T_4: Kinetin 25 mgL^{-1}	4.2	4.3	4.1	4.0	4.1	4.1	3.1	3.5	3.5	3.5	3.1	3.5
T_5: Kinetin 50 mgL^{-1}	4.5	4.4	4.2	4.5	5.0	4.4	4.2	4.5	4.1	4.2	4.2	4.1
T_6: 1-MCP 100 µLL^{-1}	3.2	3.0	2.9	3.0	3.0	2.0	2.0	2.0	3.2	1.9	2.4	2.5
T_7: 1-MCP 250 µLL^{-1}	3.0	3.0	2.9	3.1	3.1	3.1	2.2	3.1	3.4	2.1	1.5	2.1
T_8: 1-MCP 500 µLL^{-1}	3.5	3.0	3.2	3.5	3.5	2.0	2.3	2.5	3.1	2.2	1.5	1.2
T_9: NEW 100 mgL^{-1}	3.0	3.5	3.1	3	3.0	3.5	2.1	3.1	1.9	1.8	1.5	1.5
T_{10}: NEW 200 mgL^{-1}	3.1	3.0	2.5	3.0	3.0	3.3	2.5	3.0	1.9	1.2	1.5	2.2
T_{11}: Calcium chloride 15 g/L	4.0	3.0	3.2	3.5	4.0	3.1	2.0	3.5	3.2	3.0	2	2.3
T_{12}: Calcium propionate 15 g/L	5.0	4.1	4.1	5.0	5.0	4.2	4.1	5.0	4.5	4.4	3.5	3.5
T_{13}: Control	3.5	3.2	4.1	3.0	4.0	3.1	3.5	3.5	3.0	3.2	2.0	3.1
SEm±	0.03	0.05	0.01	0.03	0.02	0.05	0.09	0.04	0.04	0.08	0.08	0.04
F test	NS	NS	0.5	NS	0.5	1.0	0.5	0.5	0.8	0.7	0.3	0.8

Table 2 : Effect of pretreatments on the color characteristics of MPFL during storage at 8 °C

Treatment	4 days storage				8 days storage			
	L*	a*	b*	“E	L*	a*	b*	“E
Fresh leaves	46.11±1.01	-13.52±0.90	20.62±1.69		46.11±1.01	-13.52±0.90	20.62±1.69	
T_1: Chlorine 100 mgL^{-1}	42.15±1.74	-13.82±0.83	25.56±3.01	6.4±1.54	41.6±5.29	-12.88±0.87	28.36±7.32	9.80±6.28
T_2: GA_3 50 mgL^{-1}	41.21±1.69	-13.46±0.79	24.06±1.15	5.87±1.35	41.43±1.85	-12.55±0.98	24.56±1.76	6.67±0.92
T_3: GA_3 100 mgL^{-1}	41.43±1.23	-12.93±0.75	23.18±1.64	5.35±0.99	41.91±2.81	-12.81±1.27	23.11±3.70	6.64±1.16
T_4: Kinetin 25 mgL^{-1}	41.16±2.69	-13.54±0.69	21.81±2.08	5.07±1.89	41.93±2.23	-12.78±0.77	23.01±3.60	6.76±1.39
T_5: Kinetin 50 mgL^{-1}	45.49±2.88	-13.87±0.31	22.83±2.42	2.39±2.00	45.85±2.53	-12.25±1.14	21.43±3.22	5.02±2.27
T_6: 1-MCP 100 µLL^{-1}	41.64±0.84	-13.54±0.68	24.78±0.99	6.22±0.87	40.45±1.61	-12.68±1.03	21.86±3.23	6.77±0.25
T_7: 1-MCP 250 µLL^{-1}	42.2±1.88	-13.63±0.79	24.38±2.08	5.47±2.19	41.61±2.55	-12.66±0.49	22.71±3.44	6.87±2.31
T_8: 1-MCP 500 µLL^{-1}	41.33±0.56	-13.55±0.72	24.62±1.41	6.34±0.91	41.18±3.24	-11.90±0.44	21.77±2.85	6.38±1.65
T_9: NEW 100 mgL^{-1}	41.3±0.91	-13.68±0.50	24.56±1.26	6.33±0.83	41.49±2.32	-12.49±1.25	22.58±2.84	6.50±0.84
T_{10}: NEW 200 mgL^{-1}	42.01±2.37	-13.52±0.72	24.25±1.29	5.59±1.07	40.87±1.14	-12.41±1.19	22.76±2.56	6.36±0.93
T_{11}: Calcium chloride 15 g/L	41.86±2.32	-13.72±0.41	24.32±1.33	5.66±1.66	41.42±1.80	-12.67±1.11	22.52±2.76	6.37±1.87
T_{12}: Calcium propionate 15 g/L	45.15±2.03	-13.73±0.87	20.98±2.72	1.11±1.88	43.75±2.06	-13.38±0.55	22.61±3.77	4.69±2.52
T_{13}: Control	45.17±2.83	-13.65±0.69	24.01±2.32	5.62±2.21	41.6±5.29	-12.74±1.25	26.88±8.60	9.62±7.38
SEm±	0.04	0.04	0.03	0.08	0.02	0.02	0.004	0.19
CD at 1%	0.17	0.17	0.11	0.35	0.11	0.10	0.019	0.82

All values are expressed as mean of 10 replicated observations, ± standard deviation, * significant at 1% confidence level

Pigments and nutritional qualities

Total chlorophyll decreased in all treatments towards the end of storage, with highest retention in T_{12} (5321.0 mgkg^{-1}) followed by T_5 (4207.4 mgkg^{-1}) samples (Table 3). Pretreatment with neutral electrolyzed water resulted in drastic loss of total chlorophyll, as the content reduced to 474.40-628.87 mgkg^{-1} after storage. Kinetin was highly effective in retaining carotenoids level, while, neutral electrolyzed water pretreatment caused a highest loss of carotenoids.

Protein content of MPFL slightly reduced during storage, although T_5 followed by T_{12} was highly effective in retention of total proteins compared to other pretreatments (Table 3). Total antioxidant activity as measured by Ferric reducing antioxidant potential (FRAP) showed significant difference among pretreatments after 8 days of storage at 8°C. Antioxidant activity of MPFL from T_5 pretreatment was 1735.2 mg AEAC kg^{-1}, a level on par with fresh MPFL (1720.2 mg AEAC kg $^{-1}$) and T_{12} (1859.5 mg

Table 3 : Effect of pretreatments on the pigment composition of MPFL after 8 days of storage at 8°C

Treatment	PLW (%)	Protein (g kg^{-1})	Chlorophyll (mg kg^{-1})	Carotenoids (mg kg^{-1})	FRAP (mg AEAC kg^{-1})	Phenols (mg GAE kg^{-1})
Fresh leaves	-	54.20 ± 3.96	17103.6±118.31	358.4±8.34	1720.2 ± 0.61	1130.9±9.84
T_1: Chlorine 100 mg L^{-1}	0.150	38.16± 1.13	2487.3±28.82	138.7±1.67	1412.8 ± 13.78	1988.2±7.00
T_2: GA_3 50 mg L^{-1}	0.130	39.04± 3.78	2248.5±24.34	241.5±1.93	1485.6±10.15	1771.2±0.00
T_3: GA_3 100 mg L^{-1}	0.130	46.24±1.13	3217.4±46.60	219.7±1.93	1569.2±20.45	1908.0±2.00
T_4: Kinetin 25 mg L^{-1}	0.130	44.44± 0.28	3396.0±31.85	349.8±3.78	1960.2±3.33	2001.2±1.83
T_5: Kinetin 50 mg L^{-1}	0.120	50.84±0.62	4207.4±11.92	290.8±1.41	1735.2±3.64	1614.4±3.17
T_6: 1-MCP 100 µ LL^{-1}	0.130	44.84±0.39	1270.2±28.02	191.9±2.14	2054.4±10.00	2193.4±0.00
T_7: 1-MCP 250 µ LL^{-1}	0.130	40.88±0.79	3316.3±100.99	200.4±3.63	1914.1±3.79	2035.4±8.67
T_8: 1-MCP 500 µ LL^{-1}	0.140	46.28±1.41	2457.5±24.61	256.7±0.77	2342.5±12.88	2201.7±9.84
T_9: NEW 100 mg L^{-1}	0.140	44.68±0.28	474.0±6.02	120.1±1.47	1859.5±14.85	2044.8±1.67
T_{10}: NEW 200 mg L^{-1}	0.140	43.08±0.55	628.7±0.00	158.7±0.07	2039.4±13.94	2093.2±3.84
T_{11}: Calcium chloride 15 g L^{-1}	0.130	43.76±1.01	1664.2±49.10	180.6±0.28	2511.8±10.15	2504.7±9.01
T_{12}: Calcium propionate 15 g L^{-1}	0.100	49.72±0.73	5320.9±47.98	191.2±0.61	1898.0±8.48	1798.4±16.84
T_{13}: Control	0.120	37.08±0.56	1384.5±3.95	180.9±2.75	1553.1±5.45	2057.8±16.18
F-test	NS	*	*	*	*	*
CD at 1%	0.0392	2.07	14.90	1.13	3.21	24.90

All values are expressed as mean of triplicates, ±standard deviation, *significant at 1% confidence level

AEAC kg^{-1}). But, highest antioxidant activity and phenolics levels were observed in T_{11} (1.5g L^{-1} $CaCl_2$). Phenolics level of stored MPFL treated with T_5 (1614.3 mg GAE kg^{-1}) was higher than that of freshly harvested leaves (1130.9 mg AEAC kg^{-1}).

Besides being rich in phenolic antioxidants, fenugreek leaves were found to be very good source of vitamins such as E, K, B_1, B_4, B_5 and B_6. The loss of vitamins B_2, B_4 and B_5 could be reduced to the extent of 55-65% through kinetin treatment (T_5) over control. It is apparent that consumption of approximately 200 g of fenugreek leaves provide more than 20% of the recommended daily allowance of these vitamins. The levels of these vitamins are much higher in MPFL than some of the highly consumed nutrient rich important vegetables like raw spinach, raw broccoli, raw iceberg lettuce and raw lettuce (Food data Central, 2020).

Kinetin pretreatment (T_5) has imparted an appreciably high protective effect on all vitamins level maintenance; and could maintain vitamin E and vitamin K content on par with fresh leaves (Table 4). There was significant reduction in vitamin B levels during storage, though kinetin (T_5) pretreatment could maintain the levels in a better level than other treatments tested. Kinetin is a major naturally occurring cytokinin. Kinetin positively affects multiple aspects of plant growth by regulating cell division, organ formation and regeneration, senescence, apical dominance, vascular development, response to pathogens and nutrient mobility, chloroplast differentiation, and chlorophyll biosynthesis. The molecule is known to be naturally occurring in almost every organism, including human beings. In humans also, kinetin has got a role in healthy ageing, thus can be considered as a bioactive compound (Jan et al., 2007; Rattan, 2021). Gibberellins can stimulate rapid stem and root growth, induce mitotic division in leaves of some plants (Sun & Gubler, 2004), a mechanism which can be attributed to the effectiveness in extending the freshness of MPFL to a certain level compared to control.

Calcium propionate pretreatment was the second best pretreatment identified in this study, and showed far more superior effect on quality retention of MPFL than calcium chloride. Colour, sensory properties and chlorophyll were best maintained in calcium propionate treated MPFL till 6 days storage. However, by the end of 8 days storage, texture loss was visible as the tip of the leaves became soggy, thus reducing overall acceptability. Calcium ions strengthen plant cell wall by cross linking the bridges of middle lamella and calcium salts inhibit browning enzymes (Martin Diana et al., 2007; Li et al., 2024). The exact mechanism of texture loss initiated from leaf tip in MPFL is not explored in this study, but it may be assumed that propionic acid released from the salt is

Table 4 : Effect of pretreatments on the vitamin content of MPFL after 8 days of storage at 8°C

Treatment	Vitamin B_1 (mg kg^{-1})	Vitamin B_2 (mg kg^{-1})	Vitamin B_4 (mg kg^{-1})	Vitamin B_5 (mg kg^{-1})	Vitamin B_6 (mg kg^{-1})	Vitamin E (mg kg^{-1})	Vitamin K_1 (mg kg^{-1})	Vitamin K_2 (mg kg^{-1})
Fresh leaves	13.9±0.061	0.00233	7.5±0.029	3.5±0.001	3.6±0.001	1444.2±10.88	521.3±0.42	2.9±0.07
T_1: Chlorine 100 mgL^{-1}	4.4±0.002	0.00148	4.2±0.305	1.0±0.009	1.4±0.001	501.1±12.42	354.2±1.36	6.2±0.08
T_2: GA_3 50 mgL^{-1}	3.8±0.003	0.00108	2.3±0.045	0.16±0.01	0.8±0.063	870.2±0.51	272.5±0.01	2.7±0.08
T_3: GA_3 100 mgL^{-1}	3.6±0.001	0.00105	3.5±0.121	1.2±0.001	1.3±0.006	1178±0.40	259.3±0.39	4.8±0.13
T_4: Kinetin 25 mgL^{-1}	4.0±0.001	0.00117	3.2±0.015	1.3±0.001	1.4±0.004	913.9±2.21	337.7±0.12	5.9±0.22
T_5: Kinetin 50 mgL^{-1}	5.5±0.003	0.00290	6.7±0.088	3.6±0.005	1.7±0.002	1119.9±0.80	239.0±0.63	2.0±0.001
T_6: 1-MCP 100 μLL^{-1}	4.2±0.005	0.00102	3.2±0.001	0.21±0.001	1.3±0.001	1438.1±3.60	451.0±0.09	8.5±0.02
T_7: 1-MCP 250 μLL^{-1}	4.5±0.003	0.00143	3.1±0.054	1.3±0.001	1.3±0.005	808.4±7.96	206.4±0.47	7.3±0.04
T_8: 1-MCP 500 μLL^{-1}	4.5±0.004	0.00160	5.5±0.092	1.6±0.002	1.4±0.001	823.9±2.24	205.7±0.04	4.4±0.27
T_9: NEW 100 mgL^{-1}	4.3±0.010	0.00157	2.9±0.074	0.20±0.001	1.5±0.015	643.3±4.65	215.2±2.15	6.5±0.04
T_{10}: NEW 200 mgL^{-1}	4.5±0.005	0.00168	2.1±0.016	1.4±0.008	1.4±0.008	923.2±1.81	24.87±0.92	4.5±0.17
T_{11}: Calcium chloride 15 g/L	4.2±0.014	0.00104	3.8±0.002	1.3±0.001	1.3±0.001	923.2±1.81	290.1±1.36	4.5±0.001
T_{12}: Calcium propionate 15 g/L	4.8±0.005	0.00161	2.7±0.052	1.6±0.004	1.6±0.004	608.8±1.81	397.7±0.29	9.0±0.12
T_{13}: Control	3.8±0.022	0.00105	1.8±0.002	1.4±0.001	1.4±0.001	530.1±1.48	215.6±5.75	7.6±0.11
F-test	*	*	*	*	*	*	*	*
SEm±	0.012	0.00002	0.05	0.002	0.01	3.07	1.01	0.07
CD at 1%	0.48	0.00006	0.23	0.010	0.04	11.99	3.96	0.28

All values are expressed as mean of triplicates, ± standard deviation, * significant at 1% confidence level the causative agent for textural damage. Low molecular weight carboxylic acid s such as propionic acids are phytotoxic due to their lipophilic nature and thus disturbing protonic load in biological membranes (Himanen et al., 2012).

Although, 1-methyl cyclo propene (1-MCP) at a higher dose (500 μLL^{-1}) had a superior effect compared to control, the treatment was inferior to pretreatments *viz*., kinetin (T_5) and calcium propionate (T_{12}). Effectiveness of 1-MCP as an antisenescence agent for enhancement of shelf life in minimally processed leafy greens such as pak choy, cilantro, broccoli, etc., is documented (Cefola et al., 2010; Al Ubeed et al., 2018). 1- MCP is an ethylene action inhibitor, and have positive role in extending the postharvest lofe of leafy greens and vegetables (Ghimire et al., 2024). Being a gaseous molecule, 1 -MCP is susceptible to loss through diffusion, and abundance of ethylene produced would have replaced the ethylene binding receptors of MPFL cells, eventually limiting its application as a shelf life enhancer.

Sanitizers *viz*., sodium hypochlorite and neutral electrolysed water (NEW), did not have a positive impact on quality. On the other hand, these agents caused faster deterioration of MPFL at the levels used in this study. This observation is contrary to several earlier published reports on improvement of shelf life and bioactive compounds due to use of NEW in fresh-cut leafy greens such as broccoli, mizuna baby leaves, kale etc. (Tomas-Callejas et al., 2011; Navarro-Rico et al., 2014; Ogunnigi et al., 2021). In general, the pretreatment agents used in this study which influenced the physiology of the plant tissue had much more implications on extending the shelf life of MPFL compared to that of antimicrobial agents.

CONCLUSION

This study evaluated various pretreatments to determine the most effective method for enhancing the shelf life and nutritional quality of minimally processed fresh leafy vegetables (MPFL). It identified that pretreatment with kinetin at 50 mg L^{-1} is safe and effective, followed by calcium propionate (2 g L^{-1}) was found to be the second most effective treatment. However, other pretreatments such as chlorine, neutral electrolyzed water, calcium chloride, gibberellic acid and 1-MCP, were not particularly effective for extending shelf life of MPFL. In addition to optimizing pretreatments for minimal processing, the study demonstrated that MPFL possesses strong antioxidant properties and are an excellent source of B vitamins. The potential application of kinetin dip treatment on a commercial scale to enhance the shelf life of MPFL is suggested through this study. The feasibility of reusing the pretreatment solution should also be evaluated from an economic perspective at the industry level.

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