Cultivar and Frozen Storage Effects on Muskmelon (*Cucumis melo*) Colour, Texture and Cell Wall Polysaccharide Composition

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Abstract: Changes in composition, drip loss and colour of Cantaloupe and Honey Dew melons were determined in fresh and frozen melons stored for 5 and 10 months at -23° C. Relationships between drip loss, CWP fractions and total CWP sugar composition were determined. During frozen storage, there were significant decreases (P < 0.05) in total CWP sugars in relation to increased storage time. The decrease in total sugars was more dramatic during the 0–5 month period than during the 5–10 month period of frozen storage. Drip loss was negatively correlated with total neutral sugar content as storage time increased. The CDTA fraction yield increased and all neutral sugars decreased significantly (P < 0.05) as storage time increased. Only the CDTA fraction yield was positively correlated with drip loss.

Key words: muskmelons, storage, texture, cell wall polysaccharides.

INTRODUCTION

Melons are commonly frozen to extend their limited seasonal availability but the quality of the frozen product may be significantly altered by changes in melon textural integrity. Freezing causes severe damage to cell membranes and is therefore responsible for a loss of turgor (Reid et al 1986). Loss of cell turgor may result in a leakage of cell contents (drip loss) and a change in the sensory crispness or juiciness of the tissue. The textural integrity of plant tissues is also influenced to a large degree by cell wall and middle lamella polysaccharide composition and structure (Van Buren 1979). However, it is less clear how cell wall composition contributes to a loss of firmness in plant tissues as a consequence of freezing and frozen storage.

Carr (1982) conducted a study on the effect of freezing rate and frozen storage time on strawberries and observed that the firmness of thawed strawberries was inversely proportional to frozen storage time. As storage time increased, there was a coincident decrease

in the water-soluble fraction of pectic polysaccharides as well as total cell wall uronide content. It was argued that the increase in drip loss during long-term frozen storage may be accounted for by the loss of water-soluble pectic materials from strawberry cell walls and middle lamella. Reid *et al* (1986) found that a loss in firmness of strawberries was accompanied by the release of pectin from the tissue.

Although there have been numerous studies on the effects of frozen storage on the firmness of thawed fruit, few have evaluated differences between related cultivars. Moreover, very little information exists on the effect of frozen storage on cell wall polysaccharide (CWP) composition in muskmelons. This study describes an investigation on the effect of cultivar and frozen storage time on the cell wall composition of muskmelons. Relationships between changes in CWP fractions or total sugars and drip loss are also reported.

MATERIALS AND METHODS

Two muskmelon (Cucumis melo) cultivars, Cantaloupe (Superstar) and Honey Dew (Volga), were harvested

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from the Hermiston Agriculture Research and Extension Center (Corvallis, Oregon, USA) at three stages of maturity—half-slip, green full-slip and yellow full-slip (Seelig 1973; Evensen 1983; Reed 1991)—and stored at 2°C prior to processing. Underripe, ripe and overripe melons of both cultivars were randomly divided into two replicate groups, processed separately into melon discs 1·2 cm in thickness and 2·2 cm in diameter, and then mixed thoroughly prior to packaging. Subsamples were taken for determination of composition, firmness and colour, and discs were stored at -40°C prior to CWP which took place within 6 days.

Determination of pH, titratable acidity, soluble solids, moisture, firmness, drip loss and colour were carried out as described in Simandjuntak *et al* (1996). In addition, procedures used for cell wall polysaccharide isolation and fractionation: analysis of neutral sugars, trifluoro-acetic acid insoluble fractions and uronic acid were the same as described earlier (Simandjuntak *et al* 1996) and therefore will not be repeated here.

RESULTS AND DISCUSSION

pH, titratable acidity and soluble solids

Significant (P < 0.01) reductions in pH and increases in titratable acidity (TA) were observed as frozen storage time increased from 0 to 5 and from 5 to 10 months for both Cantaloupe and Honey Dew melons (Table 1). Reddy (1986) reported that TA was significantly higher (P < 0.05) in melons after 6 and 12 weeks of frozen storage compared with the TA of fresh unstored melons. Boyle *et al* (1977) found that the total acids in raspberries stored at -7° C decreased dramatically within 2 weeks of storage.

BeMiller (1986) stated that pectins achieve maximum stability at about pH 4·0 and that below or above this level de-esterification and depolymerisation occur concurrently. Solubilisation and de-esterification of poly-

uronides may have occurred during prolonged storage of melons, which were approximately pH 6 when fresh, or during homogenisation prior to pH measurement. Both reactions could lead to an increased number of free carboxyl groups (pectic and pectinic acids) and free hydrogen ions. Increases in TA may be attributed to dehydration and condensation of water on packaging materials during prolonged frozen storage, however one would expect SS levels to appear higher also, and this was not the case.

Soluble solids

Neither melon cultivar showed significant changes in soluble solids (SS) levels during frozen storage (Table 1). We believe this to be the first study reported on the effect of frozen storage on the SS contents of Cantaloupe and Honey Dew melons. However, in a related investigation of the effects of refrigerated storage on SS content in muskmelons, Cohen and Hicks (1986) evaluated the effects of storage at 5, 12·5 and 20°C. The authors found that there was no effect of storage temperature on SS content after 2, 5 or 9 days at any temperature; therefore, it is reasonable that SS content did not change during periods of frozen at -23°C in our study.

Drip loss

Drip loss increased significantly as storage time increased in both Cantaloupe and Honey Dew melons (Table 1). An increase in drip loss indicates greater loss of liquid cellular components and may have been caused by either mechanical or enzyme-catalysed disruption of cell walls and membranes during frozen storage. Ice crystal growth may occur during frozen storage and crystals may impart mechanical damage by physically rupturing cell walls.

Drip loss is typically ascribed to three factors, eg high internal pressure in the product, formation of ice crys-

TABLE 1
Composition, firmness and colour of melon discs after frozen storage times

Cultivar	Month	pΗ	TA^a (%)	SS (° $Brix$)	Drip loss (%) ^b	Firmness (g)	Moisture (%) ^b	Colour				
								L	а	b	Hue angle (°)	
Cantaloupe	0	6.48	0.05	9.2	12.9	189	91.4	44.4	12.3	22.7	28.5	
	5	6.26	0.07	9.32	15.2	_		54.6	12.0	21.6	29.1	
	10	6.09	0.09	9.5	17.2	_	_	55.0	9.9	19.5	26.9	
Honey Dew	0	6.16	0.08	11.0	11.2	1200	89.2	54.9	-7.4	20.8	-19.7	
	5	5.80	0.09	10.7	12.1			64.3	-7.1	20.8	-18.9	
	10	5.76	0.14	10.6	14.9			64.9	-6.7	19.8	-18.6	

^a TA is % w/w anhydrous citric acid.

^b % is w/w.

tals in the product and the irreversibility of water removal from cells (Jul 1984). However, Jul (1984) claims that none of these factors completely explain the phenomenon of drip loss. Boyle et al (1977) state that although the effect of freezing rate on textural properties has been intensely studied, there is still disagreement on the desirability of slow vs fast freezing. As storage time increased, increased disruption may have led to greater drip loss. Reddy (1986) reported an increase in the drip loss of frozen muskmelons as storage time increased.

Colour

Hunter L (lightness), a (redness to greenness) and b (yellowness to blueness) values were evaluated and hue angles were calculated. L values increased as storage time increased in both melon cultivars (Table 1). The a values for Cantaloupe decreased, while those for Honey Dew increased with storage time. The b values for Cantaloupe melons decreased continuously as frozen storage time increased and Honey Dew melons decreased only between 5 and 10 months of storage.

These observations suggest that degradation of β -carotene (yellow to orange pigment) in Cantaloupe melons and degradation of chlorophyll (green pigment) in Honey Dew melons may have occurred as frozen storage time increased and degradation was more pronounced during the later (5–10 month) period of frozen storage. Watt (1977) reported that water-soluble nutrients such as ascorbic acid are susceptible to loss during frozen storage. Both β -carotene and chlorophyll are

sensitive to changes in acidity, and either acid hydrolysis or increased activity of enzymes able to degrade these pigments may have resulted in their degradation (Boyle *et al* 1977).

Cell wall polysaccharide yields

CWP yields in Cantaloupes decreased slightly (9.6%) after 5 months of storage, but no difference in yield was found between the 5 and 10 month storage times. CWP yield of Honey Dew melons were not affected by frozen storage time (Table 2). There have been no other reported studies of changes in CWP yield during frozen storage with which the results of this study may be compared. Simandjuntak *et al* (1996) reported higher CWP yields as well as α -cellulose contents in fresh Honey Dew melons compared with Cantaloupe melons. It may be that, in general the cell wall materials of Honey Dew melons are more stable than those of Cantaloupe melons.

Van Buren (1979) stated that cellulose has the function of providing rigidity and resistance to tearing. Therefore, the lower relatively cellulose content observed in Cantaloupe melons may result in greater losses in Cantaloupe CWP yields, compared with Honey Dew, especially during the initial stages (0–5 months) of frozen storage. Carr (1982) examined the effect of frozen storage time on cell wall uronide content of strawberries and reported a decrease in the total uronides as frozen storage progressed, but alcoholinsoluble yields were not reported.

TABLE 2
Yields of CWP and CWP fractions of Cantaloupe and Honey Dew at different storage times

	0 month (mean)	SD	5 months (mean)	SD	10 months (mean)	SD
Cantaloupe						
CWP (mg g ⁻¹ FW melon discs) ^a	4.92	0.06	4.45	0.03	4.42	0.04
CWP fraction (mg g ⁻¹ CWP) ^b						
CDTA	215.00	4.13	240.00	5.20	305.00	6.70
Na ₂ CO ₃	113.00	2.26	129.00	3.80	120.00	3.70
GTC	79.50	2.49	68.70	2.10	61.00	4.90
KOH	131.00	2.88	121.00	5.40	111.00	2.80
Residue	499.00	13.40	477.00	10.30	449.00	13.40
Honey Dew						
CWP (mg g ⁻¹ FW melon discs) ^a	5.20	0.06	5.19	0.10	4.96	0.05
CWP fraction (mg g ⁻¹ CWP) ^b						
CDTA	225.00	7.69	255.00	8.31	315.00	3.33
Na ₂ CO ₃	82.50	2.74	129.00	2.99	78.70	0.63
GTC	136.00	3.61	77.40	1.97	73.50	1.12
KOH	78.50	2.50	63.80	2.27	64.70	1.72
Residue	516.00	17.30	511.00	13.90	510.00	14.10

^a The average of two isolations.

^b The average of two fractionations (duplicate) from combination of the two.

Cell wall polysacchartide fraction yields

The CDTA fraction yield increased significantly (P < 0.01) with increasing frozen storage time in both Cantaloupe and Honey Dew melons (Table 2). For both cultivars, the Na₂CO₃ fraction increased during the period from 0 to 5 months of frozen storage (13% in Cantaloupe and 56% in Honey Dew), then decreased during the 5–10 month period of frozen storage (7% in Cantaloupe and 39% in Honey Dew). These observations suggest that pectin modification occurred during the prolonged frozen storage of melon discs.

Pectin modification may occur as a result of the action of a number of enzymes, in particular pectinesterase and polygalacturonase (Haard and Salunkhe 1975). Polygalacturonase hydrolyses the glycosidic bonds between adjacent polygalacturonic acid residues and results in formation of shorter chain polymers and subsequent tissue softening. Pectinesterase hydrolyses methyl esters from galacturonic acid residues and produces pectic and pectinic acids, in addition to freeing carboxyl residues. Enzyme activity is slowed at temperatures below -18° C (Boyle *et al* 1977), however during long-term storage enzyme activity may be sufficient to result in qualitative changes.

GTC fractions decreased in both melon cultivars during the first 5 months of frozen storage (13% for Cantaloupe and 43% for Honey Dew), and did not undergo further change during the 5-10 month storage period. The KOH fraction in Cantaloupe decreased only slightly throughout storage, while in Honey Dew melons it decreased by 18% during the 0-5 month period of frozen storage. It is possible that the low pH of PAW (pH = 1.30) used during the isolation of CWP of melons may affect the degradation of pectin and hemicellulose fractions, therefore GTC and KOH fraction yields may decrease and may not be recoverable during the period of CWP fractionation (Huber 1991). Unfortunately, there have been no other studies of frozen storage changes in CWP fractions to with which the results of this study may be compared.

Relationship of CWP fractions to drip loss

The relationships between CWP fraction yields and drip loss in Cantaloupe and Honey Dew melons as a function of frozen storage time are shown in Tables 1 and 2. Drip loss was positively correlated with CDTA fraction yields in both cultivars, showing an increase in relation to increased frozen storage time. After 0, 5 and 10 months of frozen storage in the Cantaloupe, the combined CDTA and Na₂CO₃ (pectin) fractions totalled 31.6%, 35.6% and 40.6% and the hemicellulose fractions (GTC and KOH) totalled 20.3%, 18.3% and 16.4% of the total CWP, respectively. Parallel measures for the Honey Dew, were 29.6%, 37.1% and 37.8% of

the total CWP for pectins and 20.7%, 13.6% and 13.3% of the total CWP for hemicelluloses.

The greatest percentage change in pectins was detected during the 0–5 month period of frozen storage, which may correspond to the period of maximum ice crystal growth during the initial period of storage. Ice crystal growth may have an effect on both pectin and hemicellulose integrity and may cause both cell separation and disruption of individual cell walls.

Neutral sugars in trifluoro-acetic acid (TFA-) soluble fractions

Neutral sugar composition of each CWP fraction are shown in Table 3. During frozen storage of ripe Cantaloupe and Honey Dew melons, there were significant decreases (P < 0.05) in the proportion of sugars with increased storage time. These declines appear to be more obvious in Honey Dew melons. The quantity of sugars in both melons decreased more dramatically during the 0-5 month period than during the 5-10 month period of frozen storage (Table 3). The mechanism by which CWP sugar composition decreased during frozen storage is not clear and further investigation would be instructive.

Sugars in TFA-Insoluble fractions

Sugar content in the TFA-insoluble fractions of both Cantaloupe and Honey Dew did not change significantly during frozen storage (Table 3). The only exception was the TFA-insoluble sugar content of Honey Dew melons, which decreased slightly (8.29%) during the period from 5 to 10 months of storage. For both melons, total sugars in the TFA-insoluble fractions were highest in the residue fraction, followed in decreasing order by the CDTA, Na2CO3, GTC and KOH fractions. In general, only a small proportion of the CWP was solubilised by TFA (10.4% to 14.6% of total CWP for Cantaloupe and 9.24% to 13.5% of total CWP for Honey Dew), whereas most of the polymers were only solubilised by concentrated (720 g kg⁻¹) H₂SO₄. The increase in CDTA fraction yields in both melons also occurred as frozen storage time increased (Table 2).

Use of TFA may result in incomplete hydrolysis of cell wall materials and therefore may result in an underestimation of sugars. Although TFA may solubilise polymers, results may be incomplete, unless reduction to monomers is achieved, and therefore interpretation should be limited. The authors recognise that use of a mhdp test would complement the data, but for the purpose of this study, results of the TFA hydrolysis were still able to provide an index to the effectiveness of the cell wall fractionation procedure. Results indicate changes in neutral sugars with storage and reflect solubilisation of different fractions.

TABLE 3
Sugar composition of CWP fractions (mg g^{-1} CWP) at different storage times

Storage time (months)	CWP fraction			TFA	soluble s	sugars ^a		Total sugars in TFA soluble	TFA insoluble (Glu)	Uronic acid	Total sugars	
		Rham	Fuc	Ara	Xyl	Man	Gal	Glu	TTA Soluble	(0111)	acia	Sugars
Canteloupe												
0	CDTA	1.05	nd^b	3.67	0.60	nd	2.49	nd	7.81	14.60	222.00	245.00
	Na ₂ CO ₃	0.70	nd	3.12	0.58	nd	1.56	nd	6.98	7.95	107.00	122.00
	GTC	0.29	0.89	2.07	17.50	1.66	3.55	9.23	35.50	3.61	19.70	58.80
	KOH	nd	2.25	0.34	20.20	3.57	5.54	17.70	49.60	2.47	11.10	63.10
	Residue	1.04	0.71	2.79	2.93	2.86	6.55	18.10	35.00	320.00	82.80	438.00
	Total	3.09	3.84	12.00	41.80	8.09	19.70	45.10	135.00	349.00	443.00	926.00
5	CDTA	0.92	nd	3.00	1.29	nd	2.34	nd	7.56	15.50	234.00	257.00
	Na ₂ CO ₃	0.34	nd	1.81	0.61	nd	1.77	nd	4.83	9.67	62.90	77.40
	GTC	0.03	0.59	0.86	9.85	0.33	2.37	2.64	17.10	1.27	16.00	34.40
	KOH	nd	2.54	0.91	18.30	3.64	6.32	9.74	41.50	1.88	14.70	58.10
	Residue	0.22	0.46	2.73	2.01	2.68	6.23	11.40	25.70	314.00	81.10	421.00
	Total	1.50	3.60	9.30	32.10	6.65	19.00	23.80	96.60	343.00	409.00	848.00
10	CDTA	0.84	nd	2.84	0.04	nd	2.15	nd	6.15	1.00	287.00	295.00
	Na ₂ CO ₃	0.33	nd	1.74	0.55	nd	2.33	nd	4.95	9.71	44.80	59.40
	GTC	0.07	0.24	0.77	6.62	0.54	1.89	2.17	13.30	1.64	7.46	22.40
	KOH	nd	1.82	0.37	15.00	2.71	4.59	8.51	33.00	1.62	8.20	42.80
	Residue	0.29	0.57	2.56	3.22	2.49	7.19	11.40	27.70	312.00	58.50	398.00
	Total	1.53	2.63	8.28	25.40	5.74	18.10	22.10	85.10	326.00	406.00	817.00
0	CDTA	1.45	nd	5.14	0.95	nd	7.86	nd	15.40	28.50	191.00	235.00
	Na ₂ CO ₃	0.31	nd	1.33	2.00	nd	2.49	nd	4.32	5.01	60.70	70.00
	GTC	0.20	0.70	1.74	12.70	4.13	6.75	19.80	47.30	4.44	25.80	77.40
	KOH	nd	0.47	0.46	7.59	1.31	2.39	3.41	15.70	1.92	7.40	25.10
	Residue	trc	1.10	4.20	1.82	1.94	16.70	22.20	48.00	365.00	118.00	531.00
	Total	1.95	2.26	12.90	23.30	7.38	36.20	45.50	131.00	405.00	403.00	939.00
5	CDTA	0.39	nd	4.63	0.58	nd	6.34	nd	11.90	27.20	244.00	283.00
	Na ₂ CO ₃	0.20	nd	1.05	0.32	nd	2.98	nd	5.09	7.08	49.10	61.30
	GTC	nd	0.49	0.12	2.13	2.74	0.46	14.00	20.10	1.09	9.35	30.50
	KOH	nd	0.99	0.19	8.24	1.18	2.49	3.98	17.60	1.28	8.66	27.50
	Residue	0.11	0.25	2.64	3.15	1.82	11.60	16.70	36.30	374.00	71.70	482.00
	Total	0.70	1.72	8.63	14.40	5.74	23.80	34.70	91.00	410.00	382.00	884.00
10	CDTA	0.25	nd	3.61	0.47	nd	5.62	nd	9.94	29.10	192.00	231.00
	Na ₂ CO ₃	nd	nd	1.75	0.18	nd	2.20	nd	4.22	7.18	97.90	109.00
	GTC	nd	0.46	0.47	1.75	2.59	0.76	11.80	17.90	0.76	8.51	27.20
	KOH	nd	0.75	0.32	6.85	1.09	2.89	3.95	16.80	1.26	15.60	33.60
	Residue	0.11	0.38	1.51	3.86	1.04	9.23	12.60	28.80	338.00	72.60	438.00
	Total	0.35	1.58	7.66	13.10	4.72	20.70	28.40	77-60	376.00	387.00	840.00

^a Mean TFA soluble values from four injections. Rham, rhamnose; Fuc, fucose; Ara, arabinose; Xyl, xylose; Man, mannose; Gal, galactose; Glu, glucose.

Uronic acid in CWP fractions

Total CWP uronic acid decreased significantly (P < 0.01) from 0 to 5 months of frozen storage in both Cantaloupe or Honey Dew melons. Total uronic acid content did not change in either melon during the latter period of frozen storage.

Total uronic acid in the CDTA fraction increased as frozen storage time increased in Cantaloupes, however,

in Honey Dew melons there was an increase from 0 to 5 months, followed by a decrease from 5 to 10 months of storage. It was observed that as the amount of uronic acid increased in the CDTA fraction, the amount in the Na₂CO₃ fraction decreased. Conversely, when the amount of uronic acid decreased in the CDTA fraction, there was an increase in the Na₂CO₃ fraction. In addition, it was observed that uronic acid was present in all CWP fractions of both Cantaloupe and Honey Dew for

^b nd, not detected, ≤ 0.02 mg g⁻¹ CWP.

c tr, trace.

all storage periods. In both Cantaloupe and Honey Dew melons, uronic acid in the GTC fractions decreased significantly as frozen storage time increased. Whereas in Cantaloupe the KOH fraction of uronic acid increased from 0 to 5 months of frozen storage, in Honey Dew melons the KOH fraction uronic acid increased continuously from 0 to 10 months of frozen storage.

Relationship between drip loss and total sugars

As previously noted, an inverse relationship exists between drip loss and the firmness of melons as maturity advances (Simandjuntak et al 1996). Experiments conducted in this study were based upon the assumption that drip loss was indicative of firmness in both Cantaloupe and Honey Dew melons during periods of frozen storage. Table 1 demonstrates a consistent increase in drip loss in both Cantaloupe and Honey Dew melons as frozen storage time increased.

Although almost all of the CWP sugar contents decreased as storage time increased, the decrease was more pronounced during the period from 0 to 5 months than from 5 to 10 months of frozen storage. This may suggest that during the first 5 months ice crystal formation may result in more cell wall disruption than during the period from 5 to 10 months. A positive correlation was found between drip loss and frozen storage time, and negative correlations were calculated for all other CWP fraction sugars, with the exception that total galactose showed no correlation with drip loss.

CONCLUSIONS

During frozen storage of melon discs, changes occurred in the pH, TA, drip loss and colour of the two melon cultivars evaluated, but soluble solids did not change as storage time increased. The CDTA (pectin) fraction yields increased as storage time increased in both melon cultivars. The Na₂CO₃ (pectin) fraction increased from 0 to 5 months of frozen storage, whereas the GTC and KOH fractions decreased from 0 to 5 months and no further changes occurred as storage time increased. Neither the residue fraction or CWP yields were affected by frozen storage time in either melon cultivar.

Total rhamnose, arabinose, xylose, mannose, glucose and galactose contents decreased as frozen storage time increased suggested that pectins and hemicellulose fractions were modified and solubilised during frozen storage. Total sugar content in both types of melons decreased by a greater magnitude during the first 5 months of storage as compared to the period from 5 to 10 months. The observed changes may have been due to either mechanical or enzyme-catalysed changes in cell wall polymers.

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