

ONION BULB COMPOSITION AND ONION BULB FIRMNESS

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ABSTRACT

Chemical analysis of the cell walls of onion bulbs was performed to determine whether the proportions of different components were related to the firmness of the bulb. Cellulose and hemicellulose contents were not correlated with firmness.

Firm onions had high levels of non-uronide polysaccharides present in cell-wall extracts made with cold water, cold ammonium oxalate, and hot water. Uronides ("pectins") in the cold water and oxalate fractions did not differ in soft or firm onions. Levels of uronides were elevated in the hot-water extracts from firm onions.

Theoretical analysis indicates that bulb firmness should be related to the tensile strength of cell-walls. Since no difference was found in cellulose and hemicellulose concentrations in cell walls of firm or soft onions, it is postulated that adhesion of cell-wall fibrils to one another and the strength of the middle-lamella, rather than the strength of the fibril, are the factors limiting firmness in onions.

Additional Key Words: Allium, bulbs, firmness, hardness, elasticity, polysaccharides, tensile.

INTRODUCTION

Firm onions are desirable because they ship and store better than soft onions. On the other hand, complaints that New Zealand onions are too "hard" or too "tough" have come from some export markets. It is possible to select for onion firmness empirically, but we have not known what biochemical or structural parameters affect this property. Better information about the factors determining firmness could indicate whether other breeding objectives (such as dry matter concentrations) are likely to modify firmness.

In an empirical study of the relationship between onion bulb firmness and water potential and its components, it was found that firmness was affected by both the concentration of soluble solids and by water potential (Mann and Grant, in preparation). Unexpectedly, turgor pressure had relatively less influence on firmness.

A theoretical analysis of vegetable firmness (Pitt and Davis, 1984; Pitt, 1982) suggested that liquid-filled tissues convert external pressure (stress) into a bulge (strain). Resistance to compression consists, therefore, of an initial stage in which any elastic air spaces in the onion are collapsed, followed by a stage in which additional compression requires movement of cells relative to one another and/or rupture of the cell wall. This movement is opposed by the tensile strength of the cell walls and by the shear strength of the middle lamellae.

The influence of initial turgor pressure on bulk firmness is complex. On the one hand, high turgor can enhance stiffness; on the other hand, it can predispose cell walls to rupture when compression occurs. Pressure from external forces rapidly increases the initial turgor pressures. The speed with which external force is applied is important, since slow motions can force liquid out of cells without causing rupture of the walls. This 'finite-element' model of Pitt and Davis differs from the usual 'coiled spring/shock absorber model' that is applicable to the behaviour of

materials like cheese, marshmallows, and rubber (Bourne, 1967; Morrow and Mohsenin, 1966).

Crushing of onions is an inelastic phenomenon, described by Ang *et al.*, (1960) as a mode of "continuous failure." Microscopic examination of crushed onions (J. Mann, unpublished data) has revealed both rupture of the cell wall and failure of the middle lamella in damaged tissue.

If firmness is determined by the tensile strength of cell walls and by the strength of the middle-lamella, it is not clear why the intracellular concentration of soluble solids should contribute to firmness, unless the physiological accumulation of soluble solids is somehow related to the strength of the cell wall.

The present paper presents some preliminary studies on the carbohydrates of onion cell-walls, and values for the measured firmness of these bulbs. The results suggest that cellulose and hemicellulose and pectic substances do not contribute to bulb firmness, but that non-uronide polymers which are not fructans seem to be positively correlated with firmness.

MATERIALS AND METHODS

Firmness was measured by pressing a 1 cm diameter rod onto the "equator" of the bulb which was mounted on a plaster-of-Paris support in an Instron testing apparatus. A maximum force of 5 kg was applied. A chart recorder monitored both force applied (stress) and the distance the probe penetrated (strain). The linear portions of the force/distance curves were used to determine the apparent bulk modulus of elasticity.

Turgor pressure was calculated as the difference between osmotic pressure (freezing point) and water potential (Shardakov equilibrium test). Table 1 contains

TABLE 1: Summary of firmness (bulk elastic modulus, as MPa), % soluble solids by refractometry, and turgor pressure (MPa) for selected onion cultivars. Data summarised from Mann and Grant (in preparation).

| Cultivar | Origin | Firmness | % Solids | Turgor |
|-----------------------|--------|----------|----------|--------|
| Early Longkeeper | N.Z. | 4.07 | 12.8 | 0.47 |
| Pukekohe Longkeeper | N.Z. | 3.94 | 10.9 | 0.49 |
| Porters Early Globe | N.Z. | 2.13 | 9.8 | 0.30 |
| Southport White Globe | U.S.A. | 3.28 | 16.5 | 0.38 |
| IPB7233 | U.S.A. | 2.35 | 6.1 | 0.07 |
| IPB2172 | U.S.A. | 2.00 | 7.1 | 0.37 |
| Kaizuka Wase | Japan | 1.92 | 8.5 | 0.40 |
| Wankei | Japan | 1.65 | 10.2 | ND |
| Hikari | Japan | 1.44 | 7.6 | ND |

TABLE 2: Carbohydrate fractions of onion cell walls expressed as a percentage of 80% ethanol-insoluble material, dry weight basis.

| Cultivar Name | Cold-water | | Cold-oxalate | | Hot-water | | Hemi-cellulose | Cellulose |
|---------------------------|------------|-------|--------------|-------|-----------|-------|----------------|-----------|
| | Neut. | Uron. | Neut. | Uron. | Neut. | Uron. | | |
| Early Longkeeper | 8.13 | 3.67 | 1.61 | 5.52 | 13.58 | 18.65 | 19.82 | 29.02 |
| Pukekohe Longkeeper | 5.83 | 3.84 | 1.53 | 6.24 | 15.86 | 20.59 | 17.80 | 28.32 |
| Porters Early Globe | 5.48 | 5.43 | 1.42 | 6.64 | 12.29 | 17.54 | 20.49 | 30.70 |
| IPB 7233 | 4.66 | 4.92 | 1.33 | 5.70 | 12.09 | 17.33 | 20.77 | 33.17 |
| IPB 2172 | 4.63 | 5.86 | 1.11 | 5.83 | 11.30 | 16.15 | 21.56 | 33.55 |
| F-Ratio | 4.29* | 2.12 | 1.76 | 0.35 | 2.25 | 2.32 | 1.73 | 4.82* |
| Correlation with firmness | 0.79 | -0.17 | 0.65 | -0.09 | 0.65 | 0.63 | -0.13 | -0.26 |

representative data; further details will be given in another paper (Mann and Grant, in preparation). For the work reported herein, firmness was determined individually on three bulbs of each of four cultivars, and on four bulbs of a fifth cultivar. These bulbs were also used for chemical analysis.

Carbohydrate determinations were performed as follows: Bulbs were weighed, halved with a sharp knife, and after removal of the basal plate, dissected into six individual scales and the remaining inner core. The sections were weighed, freeze-dried, and dried in vacuo over phosphorus pentoxide. They were then reweighed for calculation of dry matter (DM), ground, and stored frozen in sealed bottles until analysis. The analysis was made on a sample of 250 mg taken from equal quantities of scales 3 and 4.

Carbohydrate fractions were sequentially extracted from the powdered samples as follows: "soluble sugars" (80% ethanol, 10 cm³, 20°C, 1 h, 3x); water-soluble polysaccharide (water, 10 cm³, 0°C, 1 hr, 2x); ionically-bound pectin (0.5% ammonium oxalate, 10 cm³, 20°C, 0.5 h, 2x); covalently bound pectin (water, 10 cm³, 100°C, 1 h, 2x); hemicellulose (1 M sulfuric acid, 10 cm³, 100°C, 1x, with one 10 cm³ water wash combined with the hydrolysate). The residue was washed with methanol, dried at 40°C, and subjected to Saeman hydrolysis of cellulose (72% sulfuric acid, 0.5 cm³, 20°C, 3 h, then diluted to 10 cm³ with water and heated 100°C for 1.5 h). All extractions were carried out in 10 cm³ screw-cap polypropylene tubes. This extraction sequence differs from traditional methods

since the extraction with ammonium oxalate preceded the extraction with hot water.

An aliquot of each fraction (80% ethanol through hot-water-soluble) was made 1 M with respect to sulfuric acid and hydrolysed for 2 h at 100°C. Neutral sugars were measured by the PAHBAH method of Lever (1972). Uronides were determined by reaction with m-hydroxydiphenyl (Blumenkrantz and Asboe-Hansen, 1973) and fructose by the thiobarbituric acid method (Blakeney and Mouton, 1980).

The relationship between firmness and chemical composition was evaluated by regression analysis of data for all bulbs, regardless of cultivar (Table 2 and 3). The correlation coefficients were corrected for degrees of freedom; they may vary somewhat from coefficients that might be calculated using the means of each cultivar.

RESULTS

Table 1 shows examples of firmness and turgor pressure of a range of onion cultivars, confirming the obvious fact that New Zealand Pukekohe Longkeepers are much harder than typical Japanese cultivars. Differences in firmness cannot be accounted for by differences in turgor pressure. Comparison of the average values for firmness from the large-scale experiments is summarised in Table 1, with the means of individual bulbs given in Tables 2 and 3, shows that varietal firmness remains reasonably consistent from year to year.

In our earlier work (Mann and Grant, in preparation) solute concentration measured refractometrically provided

TABLE 3: Concentration of cell wall components on a fresh weight basis. Firmness as MPa, DM as percentages, other values as mg per 10 g tissue.

| Cultivar Name | Firmness | % DM | Cold-water | | Cold-oxalate | | Hot-water | | Hemi-cellulose | Cellulose |
|---------------------------|----------|--------|------------|-------|--------------|-------|-----------|-------|----------------|-----------|
| | | | Neutr. | Uron. | Neutr. | Uron. | Neutr. | Uron. | | |
| Early Longkeeper | 3.57 | 13.1 | 9.4 | 4.3 | 1.9 | 6.4 | 15.6 | 21.4 | 22.5 | 33.2 |
| Pukekohe Longkeeper | 3.40 | 11.7 | 6.8 | 4.5 | 1.8 | 7.2 | 18.0 | 23.7 | 20.4 | 33.0 |
| Porters Early Globe | 1.87 | 8.8 | 4.3 | 5.9 | 1.5 | 7.2 | 13.4 | 19.0 | 22.3 | 33.4 |
| IPB 7233 | 2.54 | 10.2 | 4.6 | 4.5 | 1.2 | 5.2 | 11.4 | 16.2 | 19.1 | 30.6 |
| IPB 2172 | 1.81 | 9.3 | 4.3 | 5.5 | 1.0 | 5.5 | 10.6 | 15.2 | 20.1 | 31.4 |
| F-ratio | 17.67** | 7.98** | 4.48* | 0.77 | 2.77 | 0.28 | 4.15* | 3.20 | 1.96 | 0.21 |
| Correlation with firmness | NA | 0.57 | 0.60 | -0.30 | 0.55 | 0.00 | 0.59 | 0.58 | 0.00 | 0.03 |

a significant predictor of firmness, with a correlation coefficient of 0.58. A similar correlation of 0.57 based on dry matter percentage was found in this experiment (Table 3). These results confirm similar findings by Nieuwhof *et al.*, (1973).

One way to explain the correlation between soluble solids and firmness could be in terms of more or stronger cell walls in cultivar with high dry matter percentages. Walls were isolated as material insoluble in 80% ethanol. Three mixed fractions plus hemicellulose and cellulose were then extracted using cold water, ammonium oxalate, and hot water, respectively.

Initially, results were calculated as percentages of tissue dry weight (data not shown). It was realised, however, that most of this dry weight was composed of soluble fructans, hence this method of expressing results was meaningless. Results have therefore been presented as both percentages of the total cell walls (i.e. material insoluble in 80% ethanol) (Table 2) and as mg per 10 g fresh weight (Table 3). The latter values show wall components as a fraction of tissue volume, since the specific gravity of onions is close to unity.

Quite consistently, cell wall dry weight accounted for between 9 and 11% of the total dry weight. A cultivar with low total solids will therefore have less cell-wall material per gram fresh weight. This may explain the experimental correlation between solute concentration and firmness.

The cold-water soluble fraction represented about one percent of the total dry matter of onions. It is questionable whether materials soluble in cold water can contribute to cell wall structure. They may be artefacts arising from precipitation of sugars during the extraction with 80% ethanol. Firmer cultivars had significantly higher concentrations of cold-water soluble neutral polysaccharides, but interpretation of this is uncertain.

Neutral polysaccharides in both the oxalate-soluble and hot-water-soluble fractions were higher in firm bulbs, both as a percentage of cell walls or on a fresh weight basis.

As fructose was absent in these fractions, they are not fructans. Uronide (pectin) in the cold-water and cold-oxalate fractions was not correlated with firmness but they were positively correlated in hot-water extracts.

The concentration of hemicellulose and cellulose was not related to firmness. It had been anticipated that the

firmer cultivars, which have been postulated to have greater tensile strength in the cell wall, would have more of these two structural carbohydrates.

DISCUSSION

The purpose of this study was not to replace manual or instrumental methods of determining firmness with chemical analyses for nondestructive testing of firmness is more simple, and the chemical components that seem to be related to firmness do not show very high correlation coefficients. However these correlations and knowledge of the factors contributing to firmness can help the plant breeder develop softer or firmer onions as the market demands. Where onions are marketed for fresh consumption, high dry matter concentration could be a handicap if yields are thereby lowered. There could then be a tendency to select for strains which are high-yielding, but having low dry matter and the work reported shows that this may ultimately result in softer onions.

When the results were analysed on the basis of the composition of the cell wall, the structural components, cellulose and hemicellulose, were not correlated with bulb firmness. However, neutral sugar polymers extracted with cold ammonium oxalate and by hot water, were positively correlated with firmness. These neutral polysaccharides are likely to be a mixture of arabinogalactan polysaccharides plus side-chains from pectic polyuronide. Evidence for uronides (the traditional pectic substances) being involved with firmness was ambivalent, with only one fraction showing a positive correlation.

It is possible that the non-uronide carbohydrates that seem to contribute to firmness, are acting as adhesives bonding together the major fibrous components of the cell wall, or strengthening the nonfibrous middle lamella. In this experiment (and in a recent experiment to be reported later, Munro and Mann in preparation), oxalate treatment was needed to solubilise the strength-giving non-uronide material, which is therefore believed to be ionically bound to the rest of the wall matrix.

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