Preharvest Factors and Fresh-cut Quality of Leafy Vegetables

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Quality characteristics of leafy greens
1. Fresh appearance
2. No discoloration,
3. No decay,
4. Crisp texture,
5. Good flavour (aroma and taste)

Main causes of quality loss
1. Browning
2. Off-flavours
3. Texture loss

Pre-harvest factors that affect quality of leafy greens
1. Genotypes
2. Environmental conditions
3. Agricultural practices
4. Maturity at harvest
5. Harvest time

1. Genotype
Criteria for the selection
- Yield (high plant density, low core length, solid midrib),
- Visual appearance, flavor,
- Resistant to internal defects (tipburn and pinkrib),
- Health promoting constituents

1. Genotype: Selection based on health promoting constituents
Individual and total polyphenol content of baby leaves

*Graph showing polyphenol content of baby leaves for different genotypes.*
1. **Genotype**

**Criteria for the selection**
- Yield (high plant density, low core elongation, solid midrib),
- Visual appearance, flavor,
- Resistant to internal defects (tipburn and pinkrib),
- Health promoting constituents
- Selection of cultivars with lower response for cutting

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**Harvest dates:**
- December
- January
- April

**Tissues studied:**
- Photosynthetic + midrib
- Midrib

**Results:**
- Cultivar 1 vs cv 4: higher susceptibility to browning, lower vitamin C, higher phenolic content and higher enzymatic activities of browning related enzymes (PPO and POD)

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**Differences between two Romaine cultivars**

<table>
<thead>
<tr>
<th>Quality attributes</th>
<th>1</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vitamin C</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>Phenolic content</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>PAL</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>POD</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>PPO</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>% PPO activation</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>Respiration rate</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>Browning in air</td>
<td>✔</td>
<td>-</td>
</tr>
<tr>
<td>Browning in MAP</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Off-flavours in MAP</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

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**Biomarkers of enzymatic browning (AGL2013-48529-R)**
Plant metabolites including phenolic compounds and other primary and secondary metabolites evaluated using a metabolomic approach

- **Agilent QTOF 6550**
- **Agilent QQQ 6460**

**UPLC-MS-QTOF (Quadrupole + Time of Flight):** Non-targeted metabolomics. Exact molecular weight and chemical formula. High sensitivity. Authentic standards for identification

**UPLC-MS-QQQ (Triple Quadrupole):** Targeted metabolomics. High sensitivity. Authentic standards for quantitation
1. Genotype: Selection based on low off-flavor potential
Biomarkers of enzymatic browning (AGL2013-48529-R)

The untargeted metabolomic study was able to differentiate three groups of metabolites that were able to explain differences in the browning process.

### Phospholipids, Fatty acids and Phenylpropanoids

<table>
<thead>
<tr>
<th>Compound</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>PS(18:2(9Z,12Z)/0:0) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>PI(18:3(6Z,9Z,12Z)/0:0) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>PS(20:5(5Z,8Z,11Z,14Z,17Z)/0:0) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>LysoPE(0:0/20:3(8Z,11Z,14Z)) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>LysoPE(0:0/18:0) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>LysoPE(15:0/0:0) Phospholipids</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

### Volatile Compounds

- **Greenforest Honeydew melon**: Sweet melon, Fruity, Good, Skunky
- **Sea, Rotten fish**: Leather, Fishy, Decaying fish
- **Mushroom**: Intense, Sweaty, Rotten, Foul
- **Raw almond**: Intense, Sweaty, Rotten, Foul
- **Bitter, Rotten fish**: Intense, Sweaty, Rotten, Foul

### Analysis of volatiles by GC-MS Solid Phase Micro Extraction (SPME)

Combination of gas chromatography-coupled offactometry techniques (GC-O) and gas chromatography–mass spectrometry (GC-MS).
1. Genotype selection

Conclusions

- Selection of cultivars is critical because of the strong influence in the “freshness” of the fresh-cut product

Advantages

- Consistency of the raw material
- Consistency of the quality of fresh-cut product
- Consistency of the stability of the shelf life

Breeding program for lettuce

1. Genotype selection

Conclusions

1. Genotype selection

Advantages

- Consistency of the stability of the shelf life
- Consistency of the raw material
- Consistency of the quality of fresh-cut product

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Pre-harvest factors that affect quality of leafy greens

1. Genotypes
2. Environmental conditions
3. Agricultural practices
4. Maturity at harvest
5. Harvest time

Convenience Catalogue

Other factors such as environmental and cultural practices influence shelf life

Storage conditions influence shelf life. Experimental design: Active MAP (0.5% O₂ + 8-10% CO₂) + 24 h in air

Results must be confirmed in different seasons and years

Seed companies must provide the selected cultivars through several years (confidential agreement)

Growers must use recommended cultural practices for cultivation
2. Environmental conditions: Temperature, RH, light and rainfall

- Influence cultivation in different production areas and growing cycles

Fresh-cut Romaine lettuce cultivars stored for 11 d at 7ºC in Active MAP + 24h 7ºC after opening the bags

- Influence differences in the specifications of raw material
  - Maturity indices: size, weight, compactness

Fresh weight vs harvest week of baby leaf red lettuces

- Influence diseases and disorders of lettuces

Appearance, location and evolution of spots and alterations observed in lettuce leaves (Blancard et al., 2005)

- Influence in the content of phenolic acids and flavonoids

Phenolic compounds vs harvest week of baby leaf red lettuces

Respiration rate of fresh-cut iceberg lettuce harvested in different months stored for 9 d at 7ºC and 0.5% O2

Radiation and temperature showed positive correlations with the content of phenolic acids and flavonoids, but not all of the parameters.
Greenhouse vs field production

Soilless system in greenhouse: lack of lettuce heart and bolting in the case of butterhead and the lack of color in the case of red-leaved genotypes, as well as the presence of tipburn in all genotypes.

Environmental conditions: Temperature and light intensity

• Protected culture system: increases yield, allows off-season production, controls abiotic factors and facilitates pest management.
• Cultivation in open field: more resistant crops because of the adverse weather conditions and long growing cycle.

Pre-harvest factors that affect quality of leafy greens

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Agricultural practices: Fertilizers

Organic soil amendments

Agricultural practices: Irrigation

Crop: Romaine and iceberg lettuce
• Total cultivated area: 1080 m²
• Number of treatments: 5
• Number of harvest: 6

Yield (g m⁻²)

% Water Content

Phenolic content (mg 100 g⁻¹)

Glucosinolates (mg 100 g⁻¹)
3. **Agricultural practices: Irrigation**

Visual quality of fresh-cut iceberg lettuce stored during 13 days in MAP (3 d at 4 °C and 7 °C for the rest of the storage) as affected by different irrigation regimes.

**Low irrigation doses are recommended for quality and economic reasons.**

3. **Agricultural practices: Irrigation**

Cut edge browning (left) and PPO (right) of fresh-cut iceberg lettuce stored in air 7 °C as affected by different irrigation regimes.

3. **Agricultural practices: Soil and soilless systems**

Content of phenolic compounds in different lettuce types.

3. **Agricultural practices: Nutrient solutions**

Macronutrients (NO₃⁻ and macrocations (K⁺, Ca²⁺ and NH₄⁺))

Genotype and season strong influence on quality and shelf life of fresh-cut product.

3. **Agricultural practices: Salinity**

Clarkson et al. (2003) observed that lettuces exposed to an stress increased the plasticity which conferred higher acceptability and shelf life to the product.

**Elasticity** (reversible extension) and **plasticity** (irreversible extension)

Lettuces cultivated in soilless under different salinity concentrations (mM NaCl).

3. **Agricultural practices: Salinity**

Analysis of the internal anatomy of the tissue

![Image captured by the optical microscope (A) and the corresponding cellular vision in the inner part of the area studied (B).]

Higher procesability if there is a combination between small cells with a high solute concentration and strong cellular walls (Clarkson et al. 2003; Zhang et al., 2007; Wagstaff et al., 2010).
3. Agricultural practices: Salinity

Analysis of the internal anatomy of the tissue

Lettuces cultivated in soilless under different salinity concentrations (mM NaCl)

Ue, upper epidermis; le, lower epidermis; pp, palisade parenchyma; sp, spongy parenchyma; ie, intercellular spaces

3. Agricultural practices: Drought stress

Content of phenolic compounds with mild and severe deficit irrigation (DI)

3. Agricultural practices

Conclusions

- Optimization and control of agricultural practices are of paramount importance, particularly IRRIGATION, to ensure the quality leafy greens.
- The combination of adequate production systems and suitable cultivars is considered essential to ensure the shelf life of leafy greens.

4. Maturity at harvest

Influence of maturity stage on quality of fresh-cut Iceberg lettuce

Headspace gas composition of fresh-cut Iceberg lettuce at different maturity stages

Pre-harvest factors that affect quality of leafy greens

1. Genotypes
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4. Maturity at harvest

Ethanol (A) and acetaldehyde (B) of fresh-cut iceberg lettuce at different maturity stages

Days of storage

0 2 4 6 8 10 12 14

Acetaldehyde (µmol 100 g⁻¹fw)

Etanol (µL kg⁻¹fw)

Days of storage

0 2 4 6 8 10 12 14

Immature

Commercial

Over-mature

A

B

A

B

0 10 20 30 40 50 60 70 80 90 100

MS 2

MS 3

MS 4

Content of individual and total polyphenols

Density of planting (plants/m²)

300 30 7

Cultivation (Days)

66 95 130

Baby Multi Whole heads

Content of polyphenols (mg·100 g⁻¹ FW)

Green Leaf Red Leaf Lollo Rosso

Derivados de ácidos cafeicos

Flavonoides

Antocianos

Baby Multi Cabezas Baby Multi Whole heads

4. Maturity at harvest

Influence of maturity stage on quality of fresh-cut Romaine lettuce

Days of storage

-2 0 2 4 6 8 10 12 14 16

Unwashed Washed

Pre-harvest factors that affect quality of leafy greens

1. Genotypes
2. Environmental conditions
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4. Maturity at harvest
5. Harvest time
5. Harvest time: Time of the day for harvest

### January
- Growing cycle: 14 d
- Sun radiation: 7 h/day
- Tª: 10°C
- Rain: 0.1 mm

### April
- Growing cycle: 36 d
- Sun radiation: 9 h/day
- Tª: 13°C
- Rain: 1.1 mm

- **Dehydration of the leaves in the middle of the day but it recovers in the afternoon.**

#### Respiration rate of minimally processed baby spinach cultivated in spring, harvested at different times and stored for 9 d at 7°C

<table>
<thead>
<tr>
<th>Time</th>
<th>H1</th>
<th>H2</th>
<th>H3</th>
</tr>
</thead>
<tbody>
<tr>
<td>00h</td>
<td>8</td>
<td>12</td>
<td>20</td>
</tr>
<tr>
<td>12h</td>
<td>10</td>
<td>15</td>
<td>20</td>
</tr>
<tr>
<td>22h</td>
<td>15</td>
<td>18</td>
<td>20</td>
</tr>
</tbody>
</table>

- **In winter, the differences observed among harvest times were reduced after processing during storage, while in spring these differences remained.**
- Baby spinach could be harvested at any time of the day in winter and early morning in spring.
- Baby spinach harvested early in the morning had a higher water content, firm texture, lower respiration rate and better visual quality.

### Leaf water content and environmental Vapor Pressure Deficit (VPD)

- **5. Harvest time: Time of the day for harvest**

#### Winter
- Visual quality
- Weight loss
- Respiration rate

#### Spring
- Visual quality
- Weight loss
- Respiration rate

- **Preharvest Factors and Fresh-cut Quality of Leafy Vegetables**

#### Future steps

A greater understanding of the mechanisms, including the metabolites involved in browning, off-odor development and texture loss will lead to the identification of the key preharvest factors for each species or cultivar.
References: Preharvest Factors and Fresh-cut Quality of Leafy Vegetables

1: Genotypes

2: Environmental conditions

3: Agricultural practices

4: Maturity at harvest

5: Harvest time and postharvest handling

References: Preharvest Factors and Fresh-cut Quality of Leafy Vegetables

3: Agricultural practices

References: Preharvest Factors and Fresh-cut Quality of Leafy Vegetables

Thank you for your attention!