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EFFECTS OF POSTHARVEST TREATMENTS ON STORAGE QUALITY OF LIME (CITRUS LATIFOLIA TANAKA) FRUIT

A thesis presented in partial fulfilment of the requirements for the degree of Doctor of Philosophy in Food Technology at Massey University, New Zealand

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Limes (*Citrus latifolia* Tanaka) are an attractive fruit crop but generally suffer a loss in value as their colour changes from green to yellow. Various approaches were taken to slow degreening including low temperature storage, use of controlled atmosphere (CA) environments, and treatment of fruit with physiologically active agents such as gibberellic acid (GA$_3$). However, the cold storage life of lime fruit can also be restricted by a number of factors including chilling injury (CI) and rots. Various pretreatments such as the use of fungicide (thiabendazole, TBZ) and hot water dipping (HWD) and several postharvest regimes based on temperature conditioning (step down technique) and intermittent warming (IW) regimes were further investigated to protect the fruit against rots and CI during cold storage. The objective of this study was to determine what storage conditions and pretreatments would permit long term storage of NZ limes with minimal loss of quality.

CA storage (10% O$_2$ with 0 or 3% CO$_2$) was compared to regular air storage (RA) and IW (varying durations) treatments across a range of temperatures. Although some CA storage regimes could assist in delaying degreening, none of the treatments provided protection against CI. CA storage at 3% CO$_2$ delayed yellowing and gave better fruit quality than the low CO$_2$ treatment. High CO$_2$ CA treatments at 5 or 7°C decreased the rate of colour change compared to other constant temperature treatments but did not protect against CI. CI limited storage of fruit under all conditions at constant low temperatures.

Including fungicide (TBZ) in the dip water reduced the incidence of rots and had a secondary effect on protection against CI of lime fruit. However, fungicide use may sometimes exacerbate stresses such as heat injury on lime peel. Hot water dipping has been shown previously to hold potential as a storage pretreatment, but this technique may give risk of damage on produce if it is dipped at too high a temperature. Some HWD treatments did delay degreening, but there was no major effect on CI. HWD at > 47°C for ≥ 4 min caused heat injury to NZ limes. All HWD treatments showed severe CI (>15%) after 10 weeks of cold storage; and HWD fruit stored under RA at 13°C did not...
show any CI but showed some pitting (≤10%) and degreened rapidly. Overall no suitable HWD treatment for limes was identified in this trial.

This project identified the critical periods and temperature conditions for successful IW of limes. The IW conditions successfully delayed losses in quality of lime fruit provided the first warming period was applied within the first 20 days of storage. At least 2-cycle IW was required to maintain lime quality during long term storage. Some benefits were found after just one cycle of IW treatment but there were not enough to extend storage life.

IW storage benefited fruit quality and provided the highest overall fruit quality of all postharvest treatments tested. The degreening of lime during cold storage at 5°C could be delayed by IW treatments in which the fruit were stored at 5°C for 12, 16 or 20 days then moved to 15°C for 2 days. Both 2- and 6-cycle IW treatments proved satisfactory for maintaining colour on the green and yellow side of lime for 12 weeks of storage. IW treatments in which fruit were warmed within 20 day of cold storage did not show significant CI symptoms after 12 weeks of storage, and the 2-cycle IW treatment showed only a low percentage of CI fruit at this time. A 2-cycle IW treatment was almost as effective as 6 cycles, and a step down treatment also showed some promising results, indicating that it may be possible to further optimize the time and duration of variable temperature storage regimes to meet both quality requirements and the constraints of temperature management in commercial coolstores. The application of these regimes to other citrus species may also be beneficial. There are a number of physiological explanations that may account for the effectiveness of IW including positive effects on heat shock protein (HSP) and cell membranes. Nutritional factors such as vitamin C and flavonoid compositions were also investigated and fruit that did not show visible CI were found to retain at-harvest levels of these factors. Practical ways of implementing IW are discussed.

In order to understand the effectiveness of IW on degreening, I used a logistic model to describe degreening of lime peel. This modelling approach demonstrated that IW did not change the mechanism of lime degreening based on the similarity between the hue values predicted by the model and the actual hue values measured during lime storage. The activation energy (Ea) for degreening based on either hue angle (H°) or colour score (CS) during air storage was estimated to be ~53 and ~86 KJ.mol⁻¹, respectively. Relationship
between colour (H° and CS) and chlorophyll content, relationship between reflectance spectra (%), chlorophyll content and H° of lime fruit stored under different conditions are presented and discussed. This data allowed deduction to be made about the changes in individual pigments that are driving colour change during “good” and “bad” storage.
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**Abbreviations**

- a*: CIE Lab ‘a’ value measured by a colorimeter
- ABA: abscisic acid
- ACC: 1-aminocyclopropane carboxylic acid
- ANOVA: analysis of variance
- APX: ascorbate peroxidase
- b*: CIE Lab ‘b’ value measured by a colorimeter
- °C: degrees Celsius
- CAT: catalase
- cm: centimetre
- C₂H₄: ethylene
- C₂₀H₃₉: phytol
- C*: chroma
- CA: controlled atmosphere
- Cₐ: chlorophyll a
- Cₐ: chlorophyll b
- Cₓ+c: carotenoids
- CF: compression firmness
- d: day
- PFR: Plant and Food Research
- CI: chilling injury
- CO₂: carbon dioxide
- CS: colour score
- DNA: deoxyribonucleic acid
- DSM: diosmin
- e: exponential
- Eₐ: activation energy
- EB: extraction buffer
- Eq.: equation
- ERC: eriocitrin
- °F: degrees Fahrenheit
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FC fluorescent compound
FW fresh weight
GA$_3$ gibberellic acid
GAs gibberellins
GC gas chromatography
GR glutathione reductase
hr hour
H° hue angle
H°$_C$ H°$_{CR200}$ (hue angle measured by chromameter CR200)
H°$_S$ H°$_{Spectrophotometer}$ (hue angle measured by spectrophotometer CM-2600d)
$H-$ maximum hue
$H+$ minimum hue
H1 Harvest 1
H2 Harvest 2
H3 Harvest 3
H4 Harvest 4
H5 Harvest 5
HI heat injury
HPLC high performance liquid chromatography
HSP hesperidin
HWD hot water dipping
HWRB hot water rinsing and brushing
IRF isorhoifolin
IW intermittent warming
J Joule
K degrees Kelvin
$k$ reaction rate constant
$k_0$ preexponential Arrhenius constant (time$^{-1}$)
kD kiloDaltons
kg kilogram
kJ kiloJoule
KOAc potassium acetate
$L^*$ Lightness measured by a colorimeter
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<tr>
<th>Symbol</th>
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<tr>
<td>LCMS</td>
<td>liquid chromatography mass spectrometry</td>
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<td>LSD</td>
<td>least significant difference</td>
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R680  reflectance at 680 nm
R520  reflectance at 520 nm
R480  reflectance at 480 nm
RA    regular air
RH    relative humidity
RNA   ribonucleic acid
ROS   reactive oxygen species
RP    rusty pigment
RTN   rutin
rpm   revolutions per minute
s     second
SAM   S-adenosylmethionine
SAM dec.  S-adenosylmethionine decarboxylase
SCE   spectral component excluded
SCI   spectral component included
SE    standard error
SOD   superoxide dismutase
Spd   spermidine
Spm   spermine
SSC   soluble solids content or Brix°
t    time
\( t_0 \) reference time at day 0
\( t_{\text{ref}} \) reference time (d)
T     temperature (Kelvin)
TA    titratable acidity
TBZ   thiabendazole
TC    temperature conditioning
UV    ultra violet light