

**Non-destructive optical characterisation of fruits
with time-resolved reflectance spectroscopy**

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Abstract

Increasing attention is being paid to the possible development of non-invasive tests for the assessment of the quality of fruits. We propose a novel non-destructive method for the measurement of the internal optical properties of fruits and vegetables by means of time-resolved reflectance spectroscopy in the visible and NIR range.

A fully automated instrumentation for time-resolved reflectance measurements was developed. It is based on mode-locked laser sources and electronics for time-correlated single photon counting, and provides a time-resolution of 120-160 ps. The system was used to probe the optical properties of several species and varieties of fruits and vegetables in the red and NIR range (650-1000 nm).

In most fruits, the absorption line shape is dominated by the absorption peak of water, centred around 970 nm. Generally, the absorption spectra also show the spectral features typical of chlorophyll, with maximum at 675 nm. In particular, for what concerns apples, variations in peak intensity are observed depending on the variety, the degree of ripeness as well as the position on the apple.

For all the species and varieties considered, the transport scattering coefficient decreases progressively upon increasing the wavelength.

Keywords: Fruit, optical properties, photon migration, time-resolved reflectance

1. INTRODUCTION

The assessment of the internal quality of fruits is getting a more and more important task for the market. However, to this purpose only destructive techniques are presently used, based on chemical-physical tests (to evaluate sugar content and total acidity) and on mechanical tests (to estimate firmness). This necessarily implies that just few samples can be tested and only average information on the whole batch is obtained. On the contrary, non-invasive methods for quality assessment could be applied to each item, even repeatedly over time if necessary to monitor ripening and storage conditions, with evident commercial advantages. However, non-destructive methods presently applied, like colorimetry, usually provide information only on the external aspect and the superficial properties, like skin colour and superficial defects.

In the last years, time-resolved reflectance spectroscopy has attracted the interest of the biomedical community, as it allows the non-invasive optical characterisation of turbid media like biological tissues and most fruits as well.^{1,2} A short light pulse is injected into the medium to be analysed and during its propagation scattering and absorption events cause attenuation, delay, and broadening. Thus the time-distribution of the diffusely reflected light brings information on the optical properties of the traversed medium, and best-fit with a proper theoretical model leads to the simultaneous evaluation of both absorption and transport scattering coefficients.

The technique is sensitive mainly to the bulk, not to the superficial properties of the medium, and consequently it could provide interesting information on the internal quality of fruits. The aim of our study thus was the non-destructive assessment of the internal optical properties of fruits, using time-resolved reflectance spectroscopy from 610 to 1000 nm, as a first step toward the non-destructive evaluation of the internal quality of fruits.

2. MATERIALS AND METHODS

A synchronously-pumped mode-locked dye (DCM) laser was used as the excitation source from 650 to 695 nm, while an actively mode-locked Titanium Sapphire laser provided light in the wavelength range of 700 to 1000 nm.

A couple of 1 mm plastic-glass fibres (PCS1000W, Quartz et Silice, France) delivered light into the tissue and collected the reflected photons. For the measurements reported in the

following, the power density at the distal end of the illumination fibre never exceeded 10 mW. A home-built holder allowed us to position the fibres at a relative distance of 2 cm, parallel to each other, normal to and in contact with the sample surface.

A double microchannel plate photomultiplier (R1564U with extended red or S1 photochatode, Hamamatsu, Japan) and an electronic chain for time-correlated single-photon counting were used for detection. A scanning monochromator removed the room light and eventual fluorescence. A small fraction of the incident beam was coupled to a 1 mm fibre (PCS1000W, Quartz et Silice, France) and fed directly to the photomultiplier to account for any eventual time drift of the instrumentation and changes in the system transfer function due to wavelength tuning.

Overall, the system transfer function was <120 ps and 160 ps FWHM in the red and near IR, respectively.

The instrumentation is fully automated and the analysis and display of the measured spectra was performed in real time. A PC controlled the laser tuning and power, the monochromator scanning, and the optimisation of the system transfer function, by adjusting the laser cavity length. The overall measurement time (for data acquisition and system adjustment) was less than 10 s/wavelength.

The transport scattering and absorption spectra were constructed by plotting, as a function of wavelength, the values of the transport scattering coefficient μ'_s and absorption coefficient μ_a as obtained from fitting the experimental data with a standard solution of the diffusion approximation to the transport equation for a semi-infinite homogeneous medium.³ The theoretical curve was convoluted with the system transfer function and normalised to the area of the experimental curve. The fitting range included all points with a number of counts higher than 10% of the peak value on the rising edge of the curve and 1% on the tail. The best fit was reached with a Levenberg-Marquardt algorithm⁴ by varying both μ'_s and μ_a in order to minimise the reduced χ^2 .

In the range of measured values of the optical coefficients, with our set-up and the theoretical model we use, the accuracy in the absolute estimate of both μ'_s and μ_a is usually

better than 10%. However, the error made in the assessment of line shapes is definitely smaller (<2%)^{5,6}

Time-resolved reflectance measurements were carried out every 5 nm from 650 to 1000 nm on apples of three different varieties (Golden Delicious, Granny Smith, and Starking Delicious), Hayward kiwis, peaches and nectarines with yellow flesh, Cantaloupe melons, and Daniela tomatoes. On each sample, in general measurements were performed in two positions, usually corresponding to the most and the least coloured side, respectively.

3. RESULTS AND DISCUSSION

The absorption and transport scattering properties of fruits in the red and NIR regions of the spectrum were estimated performing measurements on more than 50 items per variety.

Tests carried out previously on intact fruits and after the skin removal had already proved that the presence of the skin does not significantly influence the measurement. Moreover, it was confirmed experimentally on apples (data not shown) that time-resolved reflectance spectroscopy effectively evaluates the bulk optical properties of fruits. Actually, the probed volume extends up to few centimetres inward. Thus data reported in the following refer to the internal optical properties of fruits.

For what concerns the absorption spectra, two main features are usually observed, with different relative weight depending on the species considered. A typical example of absorption spectrum, obtained from a Starking apple, is shown in Figure 1. A first peak is detected in the red, centred around 675 nm, and is attributed to chlorophyll-B, that shows a matching spectral feature. The spectrum is dominated by an IR absorption maximum, peaking around 970 nm and due to the water content. Minor spectral features, again attributed to water, are usually observed around 740 and 840 nm. Different apple varieties present very similar line shapes, with only minor differences in the relative weight of the two major peaks. As summarised in Table 1, even peaches and nectarines with yellow flesh, melons, and tomatoes are characterised by very similar absorption properties. Kiwis represent a major exception to this behaviour as chlorophyll absorbs at 675 nm significantly more than water at 970 nm. All species show comparable water absorption at 970 nm, with only melons often characterised by somewhat higher values.

In the case of apples, different varieties of the same species were considered to check whether their optical properties are significantly different. Similar water contents are suggested by the μ_s values measured at 970 nm (Table 1), with the Starking apples possibly containing slightly more water than the other varieties. On the other hand, differences among varieties are detected in the absolute value of the absorption coefficient at the chlorophyll peak. As reported in Table 1, the Granny apples are characterised by the highest absorption (*i.e.* are the richest in chlorophyll), while the Golden are definitely the least absorbing variety considered. The marked spread of the values at 675 nm for fruits of the same variety is probably related, at least in part, to the different levels of ripening at the time of the measurement, and consequently to the different chlorophyll content of the measured samples. However, it should be taken into account that the absorption reduces with post-harvest ripening, in agreement with the progressive decrease in the chlorophyll content. Actually, repeated measurements performed in subsequent days on the same samples confirmed that the chlorophyll absorption progressively reduces over time.

Major variations in the red absorption are observed also as a function of the measurement position on the apple: the more coloured side, better exposed to the sun light is characterised by a lower absorption at 675 nm.

For what concerns the scattering properties, no particular spectral details are detected. As expected, the transport scattering coefficient decreases slightly upon increasing the wavelength. A typical spectral behaviour is reported in Figure 2 for a Starking apple.

As already noted for the absorption, differences but also some overlap among fruit varieties and species are detected also in the scattering coefficient. As examples of the measured values, the typical range of the transport scattering coefficient at 750 nm is reported in Table 2 for the different species. Not only distinct apple varieties, but most of the fruit species considered show similar scattering properties, with μ_s' usually varying between 10 and 20 cm⁻¹. Only tomatoes and kiwis differ significantly for their low scattering (4-10 cm⁻¹).

4. CONCLUSIONS

We have proved that time-resolved reflectance spectroscopy can assess non-destructively the internal optical properties of fruits. Work is presently in progress to identify possible

correlations between the optical coefficients and the chemical-physical and mechanical parameters commonly used to evaluate the internal quality of fruits. In particular, the chlorophyll absorption at 675 nm is related to the degree of ripening in apples, while measurements performed on kiwis showed the existence of a correlation between the degree of firmness and the value of the scattering coefficient.

ACKNOWLEDGEMENTS

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Table 1. Typical ranges of absorption coefficient μ_a at 675 nm and 970 nm

| Species | μ_a (cm ⁻¹) at 675 nm | μ_a (cm ⁻¹) at 970 nm |
|--------------------------|---------------------------------------|---------------------------------------|
| Apple (Golden) | 0.016 – 0.114 | 0.354 – 0.386 |
| Apple (Granny) | 0.200 – 0.340 | 0.369 – 0.389 |
| Apple (Starking) | 0.097 – 0.233 | 0.373 – 0.406 |
| Peach (Yellow flesh) | 0.042 – 0.183 | 0.383 – 0.503 |
| Nectarine (Yellow flesh) | 0.032 – 0.178 | 0.397 – 0.500 |
| Kiwi (Hayward) | 0.506 – 1.165 | 0.409 – 0.452 |
| Melon (Cantaloupe) | 0.108 – 0.349 | 0.406 – 0.738 |
| Tomato (Daniela) | 0.063 – 0.106 | 0.558 – 0.598 |

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Table 2. Typical ranges of scattering coefficient μ'_s at 750 nm

| Species | μ'_s (cm ⁻¹) |
|--------------------------|------------------------------|
| Apple (Golden) | 12 – 16 |
| Apple (Granny) | 9 – 13 |
| Apple (Starking) | 15 – 21 |
| Peach (Yellow flesh) | 10 – 20 |
| Nectarine (Yellow flesh) | 12 – 17 |
| Kiwi (Hayward) | 4 – 9 |
| Melon (Cantaloupe) | 7 – 14 |
| Tomato (Daniela) | 4 – 6 |

FIGURE CAPTIONS**Figure 1** Absorption spectrum of a Starking apple.**Figure 2** Transport scattering spectrum of a Starking apple.

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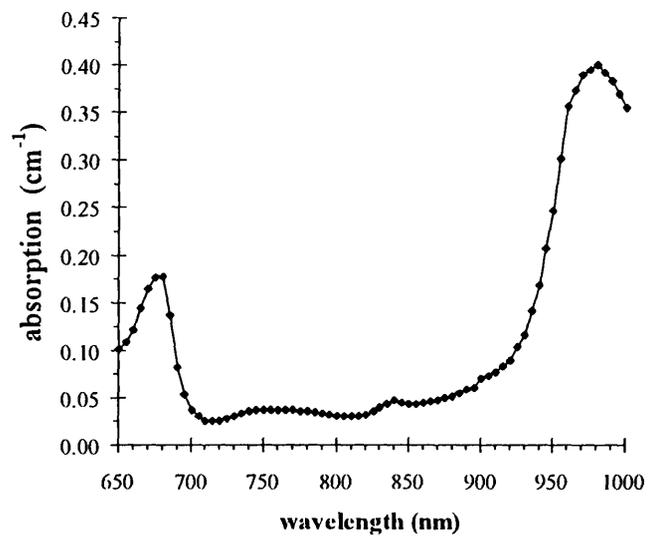


Figure 1. Absorption spectrum of a Starking apple.

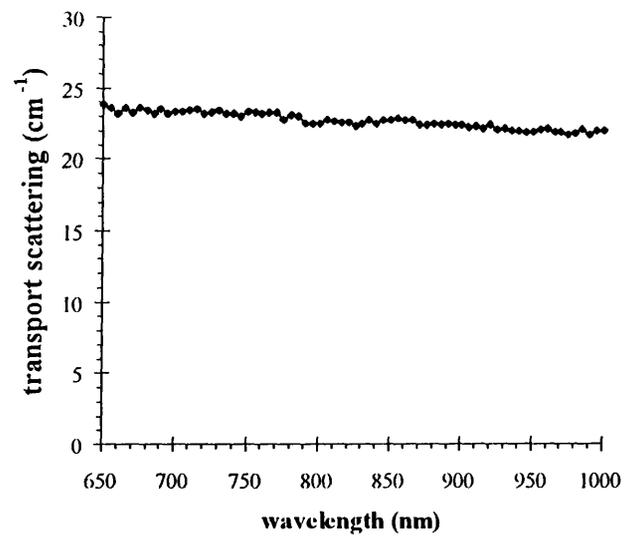


Figure 2. Transport scattering spectrum of a Starking apple.

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