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Bruising Marks on Apples and Tissue Browning in Fruit Salads

Analysed by Time-resolved Fluorescence Spectroscopy

Bruising marks and brown discolouration often occur on the surfaces of fresh or sliced fruit. These displeasing visual traits cause consumer rejection. Through laser-induced fluorescence spectroscopy (LIFS) tissue browning can be detected before the symptoms become visible. Applications of LIFS are, for example, the development of new anti-oxidative fruit salad additives, which inhibit continuous quality losses. Preliminary experiments on data processing using derivative spectroscopy showed high discrimination potential.

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Keywords

Mechanical damage, tissue browning, fruit salad, time resolved fluorescence, non-destructive Quality losses of perishable produce appear due to mechanical stress during harvesting, transport and storage processes. Oxidative polymerisation of phenols, due to an increasing partial pressure of oxygen in the apoplast and the destruction of the vacuole, leads to fruit tissue browning. With the help of the fluorescence spectroscopy these bruising marks can be detected before being visible.

In the last years more fresh sliced salads (ready-to-eat salads) are quoted in the retail than ever. For these products, the consumer demands for more monitoring and controls with respect to quality and hygiene. Beside these requirements, the appearance of brown discolouration on the surface of the fresh or sliced fruit, is one of the decisive sale criteria. Suitable additives are used up until now to reduce the loss in aesthetic value and to avoid tissue decay. Consumers look critical at such additives based on acids as ascorbic and benzoic acid, which considerably affect the flavour of the product. Such diminutions can be avoided by optimising these supplements for fresh convenient products in terms of concentration and composition.

In the present work initial tests and results of the laser-induced fluorescence spectroscopy (LIFS) will be presented. The technique was non-destructively applied on sound, homogenised and on sliced fruits measuring time-resolved fluorescence spectra. Establishing a measuring routine for detecting tissue browning should contribute to a rapid screening method to detect bruising marks and to optimise suitable additives, in order to avoid tissue browning in fruit salads.

Experimental set-up

The fluorescence spectrometer (Laser Fluoroscope LF401 Lambda, I.O.M., Germany) used is equipped with a fibre-optic probe, which allows recording the spectra directly from the sample surface. A nitrogen laser emits short, intense laser pulses at 337 nm (*Fig. 1*). A coloured glass container was placed between the laser and the focusing lens to use the system as a dye laser and changing the excitation wavelength. In the case of installing the exchangeable glass container, the fluorescence radiation can be excited in the range from 380 nm to 620 nm. The fibre probe posses a length of 3 m with a kernel diameter of 600 µm. This exciting as well as receiving fibre is coupled to a Ycoupler with a length of 0.50 m. The tip of the fibre-optic probe is pasted into a highgrade steel tube shaped with an angle of 8° to avoid measuring diffuse reflectance. The acousto-optic tuneable filter (AOTF) makes it possible to vary the detection wavelength in 1 nm intervals. The signal is detected by a photo-multiplier tube (PMT) (350 nm to 820 nm) and subsequently recorded with a high time resolution (100 ps gate width).

Data processing and results of timeresolved fluorescence spectroscopy

The fruit is composed by several fluorescent compounds like phenols, which are responsible for the brown discolouration of fruit tissue due to their oxidative polymerisation. Every single fluorescent molecule is characterised by a specific absorption and emission wavelength and a typical lifetime [1]. The optimal time-gate position of the particular samples was evaluated by means of λ_{τ} -curve and fluorescence decay measurements. For the apple experiments, laser light was passed trough the dye laser to excite the probe at 337 nm and 488 nm using time gate positions of 6.5 ns and 4.5 ns, respectively. The banana slices were excited directly with the laser at 337 nm and measured at 7.5 ns.

The data set was processed step-by-step using mathematical methods [2] and computer based programmes (TABLE CURVE 2D, SPSS Science, USA) and evaluated by derivative spectroscopy. The fluorescence spectra were smoothed for avoiding to calculate on instrumental noise. The recorded spectrum is composed of several single spectra of every fluorescent compound included in the measured probe. Each spectrum shows typical fluorescence intensity maximums in spe-





the content of ascorbic acid (Merck, Germany). The values of integral calculus of the second derivative of the fluorescence spectra (70% smoothing) show, that using vitamin c of higher concentration led to lesser fluorescence intensity breakdown compared to the start measurement (*fig. 3*). Thus high concentrated ascorbic acid solution was the most suitable additive to avoid oxidative polymerisation of phenols in fruit tissue.

Outlook

The potential of the time-resolved fluorescence spectroscopy to non-destructively detect bruising marks and tissue browning of fruits was pointed out. The application of derivative spectroscopy applied to the fluorescence spectra is a suitable method for processing the LIFS data. Thus, a potential field of application for this new technology can be the optimisation of cutting processes or antioxidative supplements for fresh convenient products in terms of concentration and composition.

cific wavelength areas. Characteristic wavelength ranges can be detected using the extreme values of the second derivation of the spectra. These wide spectral fluorescence bands can be explained by the presence of single fluorescent fruit compounds.

During the storage, bruising marks appeared visibly on the apple fruits. The storage led to chlorophyll breakdown and fluorescence spectra displacement, due to physiological changes in the fruit tissue, visible on the red wavelength area (figure 2). Furthermore, when comparing fluorescence spectra of sound and bruised apples using LIFS, changes in compound contents appeared. These can be explained by the fact that bruising led to rapid chlorophyll breakdown resulting in a decrease in chlorophyll fluorescence intensity (peak at approx. 650 nm) in the red wavelength area and changes in the blue-green fluorescence caused by the reaction of phenols [3].

The surface browning of banana fruit slices appeared within 2 hours after cutting. After spraying the fruit surface with distilled water, natural lemon juice (460 mg vitamin c L⁻¹) and vitamin c solution with lower (460 mg vitamin c L⁻¹) and higher (330 g vitamin c L⁻¹) concentration led to characteristic responses during tissue decay. The vitamin c concentration of spray solutions was estimated using a rapid colour test-set, measuring

Literature

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Fig. 3: 2nd derivative (Savitzky-Golay, 70% smoothing) and integral min-max of fluorescence spectra, recorded at banana slices, treated with different additives

