Monitoring leafy vegetables through packaging films with hyperspectral images

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Abstract

Fresh-cut or minimally processed fruit and vegetables have been physically modified from its original form (by peeling, trimming, washing and cutting) to obtain a 100% edible product that is subsequently packaged (usually under modified atmosphere packaging –MAP) and kept in refrigerated storage. In fresh-cut products, physiological activity and microbiological spoilage, determine their deterioration and shelf-life. The major preservation techniques applied to delay spoilage are chilling storage and MAP, combined with chemical treatments (antimicrobial solutions antibrowning, acidulants, antioxidants, etc.). The industry looks for safer alternatives. Consequently, the sector is asking for innovative, fast, cheap and objective techniques to evaluate the overall quality and safety of fresh-cut products in order to obtain decision tools for implementing new packaging materials and procedures. In recent years, hyperspectral imaging technique has been regarded as a tool for analyses conducted for quality evaluation of food products in research, control and industries. The hyperspectral imaging system allows integrating spectroscopic and imaging techniques to enable direct identification of different components or quality characteristics and their spatial distribution in the tested sample.

The objective of this work is to develop hyperspectral image processing methods for the supervision through plastic films of changes related to quality deterioration in packed ready-to-use leafy vegetables during shelf life.

The evolutions of ready-to-use spinach and watercress samples covered with three different common transparent plastic films were studied. Samples were stored at 4 ºC during the monitoring period (until 21 days). More than 60 hyperspectral images (from 400 to 1000 nm) per species were analyzed using *ad hoc* routines and commercial toolboxes of MatLab®. Besides common spectral treatments for removing additive and multiplicative effects, additional correction, previously to any other correction, was performed in the images of leaves in order to avoid the modification in their spectra due to the presence of the plastic transparent film. Findings from this study suggest that the developed images analysis system is able to deal with the effects caused in the images by the presence of plastic films in the supervision of shelf-life in leafy vegetables, in which different stages of quality has been identified.

Key words: artificial vision, multivariate analysis, watercress, spinach, ready to eat.

1. Introduction

Fresh-cut or minimally processed fruit and vegetables have been physically modified from its original form (by peeling, trimming, washing and cutting) to obtain a 100% edible product that is subsequently packaged and kept in refrigerated storage. In fresh-cut products, physiological activity and microbiological spoilage, determine their deterioration and shelf-life. Many factors determine if either microbiological or physiological spoilage mechanisms will dominate. However, there is an interaction between these mechanisms.

The major preservation techniques applied to delay spoilage are chilling storage and modified atmosphere packaging (MAP), combined with chemical treatments (antimicrobial solutions antibrowning, acidulants, antioxidants, etc.). Chlorine is a common efficient sanitation agent but there is the risk of undesirable by-products upon reaction with organic matter and this may lead to new regulatory restrictions in the future. Moreover, its efficacy is poor for some products. Consequently the minimal processing industry wants safer alternatives. Several antimicrobial washing solutions, O_3, UV–C radiation, intense light pulses, innovative MAP under superatmospheric O_2 and novel gases, alone or in combination, are presently considered promising treatments. However, change from use of conventional to innovative sanitizers requires knowledge of the benefits and restrictions as well as a practical outlook. To provide adequate answers to the minimal fresh processing industry needs, eco-innovative sanitation techniques must be addressed for each specific commodity in order to avoid unintended results. These techniques must satisfy the consumers and maintain a balance between sensory and quality.

Frequently, the quality decay of fresh-cut products during storage is assessed through respiration rate, composition atmosphere within package, microbiological quality (viable cell concentration of total viable bacterial count, lactic acid bacteria, yeasts and molds), dry matter, pH, colour parameters and sensory evaluations (Conte et al. 2009; Artés-Hernández et al. 2010). However, the most of methods applied are time consuming and expensive. Consequently, the sector is asking for innovative, fast, cheap and objective techniques to evaluate the overall quality and safety (or some of the specific quality parameters) of fresh-cut products in order to obtain decision tools for implementing new packaging procedures.

Most spectroscopic applications implement NIR and/or VIS ranges, and they register small portions of the studied tissues (in coincidence with the size of the fibber-optic window). Depending on the uniformity of the attributes in the products, different areas have to be sampled; in case to be searching for external damage, vision has to be used in any case. Multispectral (few wavelengths) and hyper-spectral (complete spectra) vision combine spectral and spatial information, acquiring spectra for every pixel in the image. Ongoing research in this area is bound to produce a new analytical methodology in agro-food processes, as it is now in the pharmaceutical sector (Gowen et al., 2007). Recently, research on multispectral and hyperspectral imaging techniques were carried out in the inspection of fresh-cut products (Lunadei et al., 2012; Diezma et al., 2011; Siripatrawan et al., 2011). However, no known procedures have been implemented using the imaging of the samples through plastic films, which would allow monitoring the same samples along the storage period. The objective of this work is to develop hyperspectral image processing methods for the supervision through plastic films of changes related to quality deterioration in packed ready-to-use leafy vegetables during shelf life.

2. Material and Methods

The evolutions of ready-to-use spinach samples covered with three different common transparent plastic films were studied: commercial PPLUS 160 (P1) and 190 (P2) and bi-oriented polypropylene 30 µm thicknesses (P3). Each leaf was individually kept inside a Petri dish with a piece of grey plastic used for reference, and covered by the plastic film under evaluation (Fig.1). Samples were stored at 4 ºC during the monitoring period.

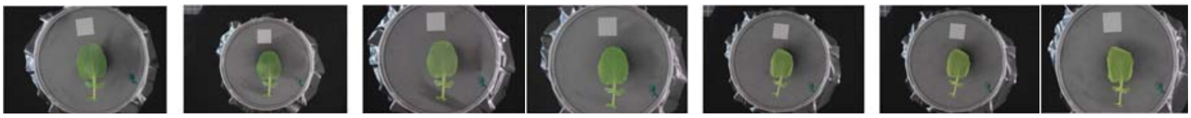

FIGURE 1: RGB images of a leaf of watercress along the monitoring period; the grey reference inside de plate can be identified.

Three groups of 5 spinach leaves were randomly selected from bags acquired in a local market. Each group was assigned to one of the three selected plastic. Hyperspectral images were acquired 7 times along 21 days (5 leaves x 3 plastics x 7 dates). Regarding watercress, only bi-oriented polypropylene was used taking into account the previous results obtained on spinaches. Nine leaves were monitored along 18 days (9 leaves x 7 dates).

Hyperspectral vision system consisted on a CCD camera with a spectrograph Headwall Photonics HyperspecTM VNIR working in the range between 400 and 1000. The spectral binning was configured to obtain 189 wavelengths (spectral resolution 3.17 nm). The acquisition and the storage of the images were made through specific software (Headwall HyperespecTM). The illumination was provided by two halogens lamps with regulated and variable intensity. Each individual leaf was placed on a platform that moved under the camera (Micos-MOCO motor). The sample was scanned line by line according to the movement of the push-broom system, acquiring the whole surface of the beam of the leaves.

Besides common spectral treatments for removing additive and multiplicative effects, additional correction, previously to any other correction, was performed in the images of leaves in order to avoid the modification in their spectra due to the presence of the plastic transparent film. Possible modifications of the transmittance of the plastic film with time were monitored and removed considering the spectra of the grey reference at each date and correcting with the spectra of the grey reference at first day.

Calibration sets were composed of the average spectra of the supervised leaves (n=105 for spinaches and n=63 for watercress). Smoothing and second derivative was computed on spectra in order to remove any additive effect. Additionally, the multiplicative effect was corrected by standard normal variate (SNV) procedure.

Principal component analyses were computed on the calibration sets. The resulting loadings were used for the projection of the corresponding corrected hyperspectral images. Artificial images of scores were obtained from such projections. These artificial images of scores were studied by means of Analysis of Variance to check if significant differences along the storage periods appear and to compare the behaviour of plastic films in the preservation of the quality and freshness. A*d hoc* routines and commercial toolboxes of MatLab® were used in the analyses.

3. Results

The application of principal component analysis (PCA), a non supervised multivariate analysis, on hyperspectral images, has allowed the visualization of the evolution of the two studied vegetables trough the plastic covers.

3.1. Spinaches

Figure 2 presents the results of the PCA applied on the calibration set of spinaches (n=105); the loadings (left) and the scores (right) of first principal component, PC1, that is the most sensitive to the evolution of leaves with time.

Figure 3 shows the images of scores for three spinach leaves, one by each plastic film; a relationship between the values of scores and the time of storage can be observed, in spite of the high variability that appears within the leaves. ANOVAs were computed on the images of scores for comparing the effect of the plastic films on the preservation of the freshness. All scores of the images of the leaves were grouped for each date (seven dates). Attending to the values of the F (Fisher), could be supposed that leaves covered by P3 presented higher evolution; however the differences neither in box and whisker plot of the scores nor in the F between plastics are higher enough to be considered concluding (Fig.4)

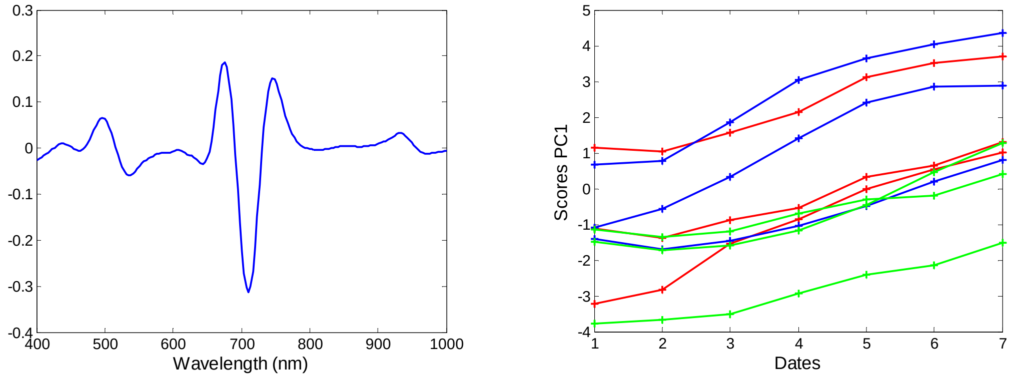

FIGURE 2: Loadings (left) and scores (right) of PC1 on calibration set of spinaches, the colour of lines corresponds to the three plastic films: red P1, blue P2 and green P3.

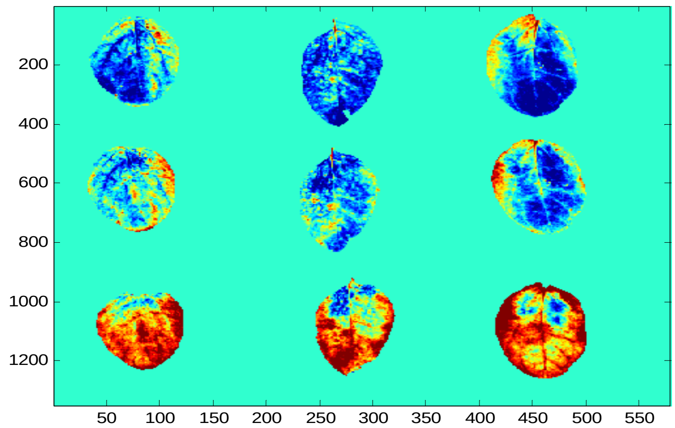

FIGURE 3: Virtual images of scores of PC1 on spinaches. Each column corresponds to a leaf covered with a type of plastic film: P1 (first column), P2 (second column) and P3 (third column). Each row corresponds to a date: first day of storage, intermediate day and last day.

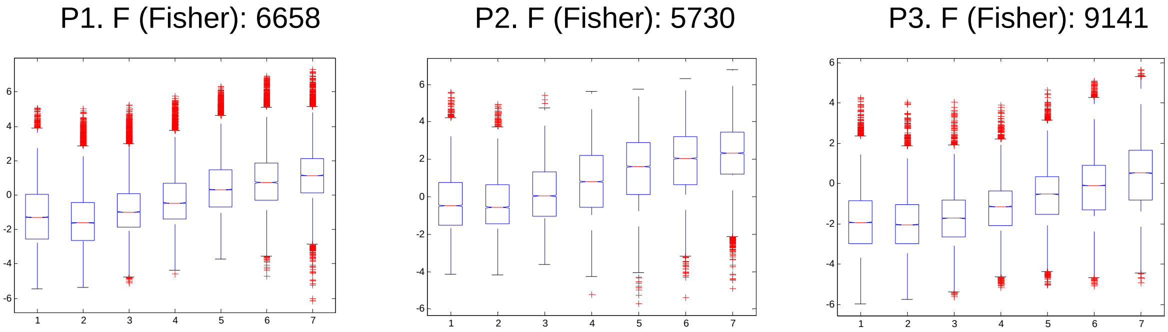

FIGURE 4: Evolution of PC1 scores of the leaves along the seven tested day. Left to right plastic films P1, P2 and P3.. X-axis: dates. Y-axis score values: boxes correspond to the average values plus/minus the standard deviation; whiskers to 1.96 times standard deviation. Average values are indicated by horizontal red lines. Outliers with red points.

3.2. Watercress

Because of plastic films P3 showed on spinaches the worst ability for preservation of freshness, on watercress only bi-oriented polypropylene (P3) was used trying to accentuate their deterioration rate Figure 5 (left) shows the loadings corresponding to the second principal component obtained from the calibration set of watercress (n=63). On Figure 5 (rigth), the evolution of the averages of scores of each leaf of the calibration set can be observed. Although there is an increase in scores over time for all the leaves, there is also a great variability in the average values along the whole period of measurement. Mostly of the average values evolve parallel; the average value of a leaf at starting date could influence the value at the end of the storage period.

On Figure 6 the virtual images of scores of PC2are represented. It can be checked that he evolution rate is different in each leave; notice that the initial stage of the watercress in the image is similar, however the stage reached at the end of the storage period is clearly different. The variability within the leaves is lower in the initial stage of evolution compared to the leaves at intermediate and final stages (Fig. 6).

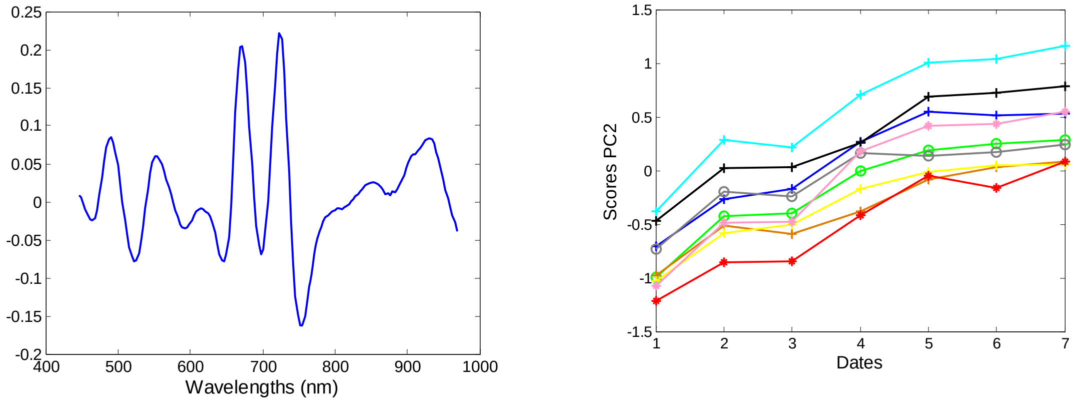

FIGURE 5: Loadings (left) and averages of PC2 scores of each leaf (right) on calibration set of watercress, each line corresponds to one leaf along the storage period.

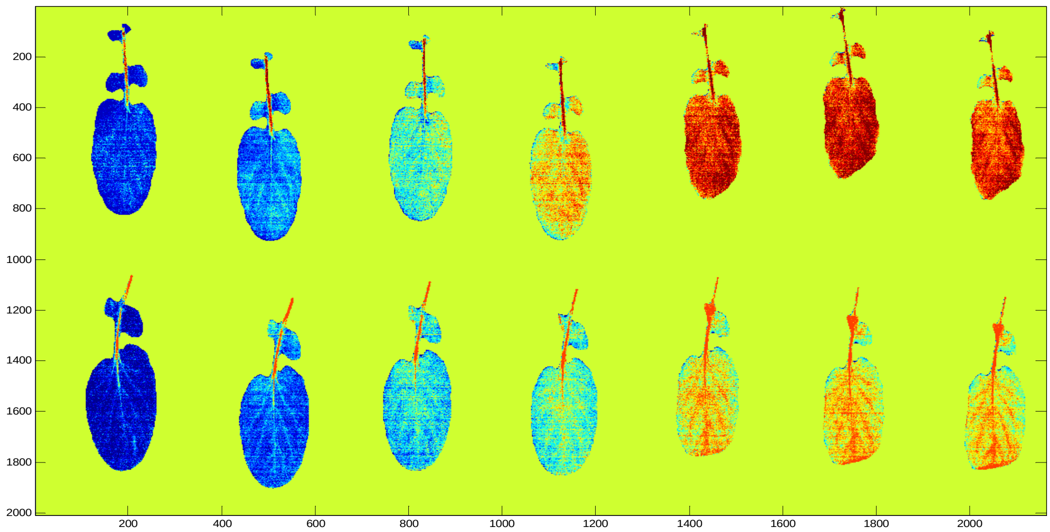

FIGURE 6: Virtual images of scores of PC2 on two leafs of watercress. Each row corresponds to one leaf supervised along 7 dates during the storage period.

ANOVAs were computed for each leaf individually; all scores of a leaf were grouped for each date and considered as a group for further ANOVA computation. The variability of F values denoted the differences in the evolution rate of the leaves (Fig. 7).

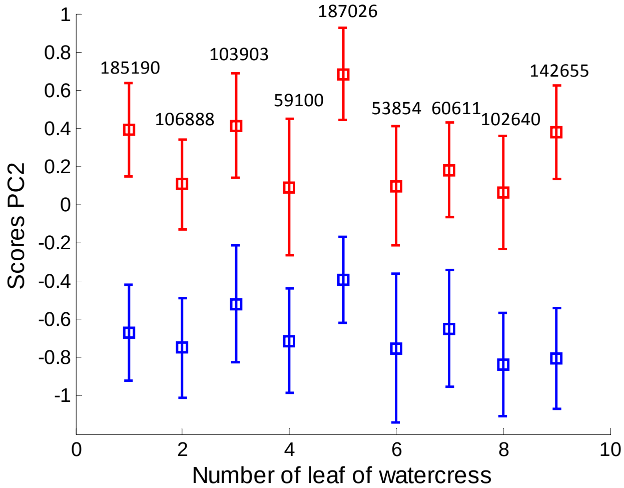

FIGURE 7: Mean and standard deviation of scores PC2 of each leaf on the first (blue) and the last (red) days. Values of F (Fisher) are presented for each leaf

4. Conclusions

The aim of this work was the development of a non-destructive analytical method for the supervision of shelf-life of ready-to-eat spinach and watercress leaves, using hyperspectral imaging. An image analysis procedure based on the consideration of a material reference has been developed to deal with the effects caused by the presence of plastic films covering or packaging leafy vegetables. Findings from this study suggest that this procedure could allow the supervision of shelf-life in packaged leafy vegetables discriminating between different storage periods. Moreover, the behaviour of the plastic films along the storage period has been evaluated for their ability to slow the loss of freshness.

Remarkable variability on initial freshness and evolution rate has been verified in leaves belonging to the same batch of packages, which indicates the advantages that could be gained if an image analysis system is applied to obtain an overall quality assessment of packaged products. The challenge for the next step of this research is the definition of multispectral indexes based on the loading obtained from PCA and on the pattern of the spectra, capable of following the evolution along storage and removing the effect of the plastic films.

Acknowledgments

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