Use of mobile phone camera to quantify calcium in fruits

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RESUMEN:
Los cultivos frutales suponen el eje de la economía de muchos países, es por ello que la mejora de sus capacidades es muy requerida a la hora de incrementar la producción para mejorar el rendimiento económico. Sin embargo, el uso indebido de productos para acelerar la producción o para evitar algún tipo de carencia nutricional puede dar lugar a otros problemas de carencias que pueden afectar a la calidad final del producto y también a nivel medioambiental. Es por ello, que una pronta detección de las carencias que sufren los cultivos es de máxima importancia para mejorar la calidad y el rendimiento de las producciones sin la necesidad de gastar cantidad innecesarias de productos que además contaminan el medioambiente. Es por ello, que el trabajo que se presenta es una alternativa bastante adecuada para solucionar estos problemas. Este trabajo consiste en el desarrollo de un método quimiométrico capaz de detectar carencias nutricionales (en especial de Calcio) en cultivos frutales (concretamente en cerezos) mediante el uso de las fotografías realizadas con dispositivos móviles. Con este método, se permite cuantificar el contenido de Calcio con el suficiente tiempo de antelación para corregirlo con el producto necesario.

Palabras clave: Carencias nutricionales, calcio en frutas, cerezos, quimiometría, fotografías móviles, calidad del fruto

ABSTRACT:
Fruits crops are usually an important economy factor in many countries; therefore, the improve of this field is important to increase the production and the economy yield. However, the unnecessary employment of correctors to increase the production or correct some nutritional problems could lead to another nutritional problems affecting the final quality of the product and also could damage the environment. Consequently, a fast and early detection of the nutritional crop problems is really important to improve the quality and the production yield without the necessity of waste huge amounts of money in products which could led to environment problems. That is why the presented work in an interesting alternative to solve these problems. This work involves the development of a chemometric tool able to detect nutritional crop problems (calcium problems in cherry trees in this case) using smarphones photografies. With this methodology, it is possible to quantify the calcium in the cherry peel with enough time to correct the nutritional problem with the necessary product.

Keywords: Nutritional crop problems, calcium in fruits, cherry trees, chemometrics, mobile phone photografy, fruit quality

INTRODUCTION

Agriculture has become much more than a way to produce food for the non-stop growing worldwide population. Plants have become important also for produce energy and it is our more important weapon to deal with the global warming issue. However, it should be important to recognize which plants should be used in the different fields (alimentation, energy, others). Therefore, plants or fruits initially produced for feed human population after analysis could be employed for other uses due to problems in the nutrient or pesticide profile. Also, it is important to do a correct early selection of plants and/or fruits that are not adequate for feed humans and animals, also reducing the large amount that every year is wasted (around 1/3 of the total produced). This is not only a problem for producers, which see their incomes reduced, also for environment for the extra employment of nutrients, pesticides and other products. In addition, the waste of food is a disrespect for the undeveloped countries that are suffering famine and see how the developed countries waste tones of fruits and plants every month.

To avoid this problem, an early detection of possible diseases or nutrient deficiencies is necessary nowadays. However, the traditional methods usually employed for these analysis are expensive, laborious, and needs samples to be recollected. In addition, this samples are removed from the field and could not be employed for sell it afterwards leading to reduce the amount of product recollected wasting plants or fruits that could not be fated to other uses. Otherwise, the recollected samples are a representative aliquot and the results are extrapolated to all the products where the sampling was made. This methodology, could led to important errors due to the variability of the fruits and plants and also due to sampling issues. On the other side, the analysis in routine labs is not easy and sometimes the amount of samples is elevated and this could lead to errors in the analysis that could be determinant to allow the use of these fruits or plants as human food. Other problem associated to the current analytical methods is the necessity of transport of samples from the field to the laboratory, since these analyses are made *ex situ*. For these reasons, the development of new strategies to detect in a simply, economic and rapid way, the possible problems with fruits or plants should be studied and developed. But also, a way to have the capability to detect *in situ* these problems could be an interesting approach to reduce the employment of large amount of nutrients or pesticides which could led to environmental problems.

On the other side, technology is improving really fast, many gadgets are getting smaller, with improved characteristics and more portable. A good example of this is the smartphone industry, which is always improving as the years go by and also, nowadays the smartphones that we have in our pockets are even more powerful than the computer that was used to send the first man to the moon. With such a powerful device in the pockets of almost two billion humans on earth, it is interesting to use these devices to improves our life. Although the main function of our smartphones is the communication with the improvement of the processors, graphical and ram characteristics and also with the adding of high resolution photography cameras, our mobile phones are ready for much more than the simply communications. In the last years, many researchers are using smartphones for different applications as bad-smell sensing (Ninh *et al.*), chemical analyzer (García *et al*.), protein detector in proteins (Lai *et al.*), fluorescence analyzers of ochratoxin A (Bueno *et al.*), pathogenic H5N1 viruses (Yeol *et al*.) and other viruses (Priye *et al.*) or to detect blood glucose (Devhadasan *et al*.). However, in the field of fruits and vegetables not too much work was done employing smartphones beyond an application to determine the fruit ripeness in apples (Das *et al*.).

As said before, since the analysis methods to determine nutrient deficiency in plants are made by expensive and slow techniques the development of new fast, cheap and easy techniques to analyze crops are necessary. In this sense, the employment of the new smartphones technologies is perfect for this field. In this work, the first approach to develop a new methodology to quantify calcium, a typical nutrient deficiency problem, in cherries by the employment of smartphone camera is presented. With this aim, the fruits were photographed with an android mobile phone and the corresponding photographs were treated in OriginLab® software to convert to black and white and to obtain the resultant numerical matrix, where some lines in different points were selected and transported to the IBM SPSS® software to make a multiple linear regresion analysis.

The method is able to quantify the calcium content in the peel of cherries in new samples by interpolating the adequate variables in the model. This new methodology led to *in situ*, cheap, easy and fast determination of nutrients in plants. In addition, the developed method allows the detection of problem in specific trees or plants, which could be translated in save cost expenses in correctors inasmuch as only specific plants/trees should be treated.

MATERIAL AND METHODS

Chemicals and samples

Calcium carbonate and lanthanum (III) chloride were purchased from Sigma-Aldrich (Milwaukee, WI, USA). Hydrochloric acid was bought from VWR chemicals (Radnor, PA, USA). Deionized water was prepared in the Barnstead deionizer (Sybron, Boston, MA, USA). Prime Giant and Irnet cherries samples were kindly donated by Agrostock S.A. (Fraga, Huesca, Spain).

Instrumentation and software

A Gallur (Manises, Valencia, Spain) muffle was employed for sample calcination. Perkin Elmer Atomic Absorption Spectrometer 1100 (Perkin Elmer, Waltham, MA, USA), a microcomputer-controlled atomic absorption/emission spectrometer with an integrated CRT screen was employed for calcium determination. Crucibles were purchased from VWR chemicals. A One Plus 3T android 7.1 (Nougat) smartphone with a 16 MP was used for obtain the fruits photos. OriginLab® 9.01 (OriginLab, Northampton, MA, USA) and ACDSee software (ACDSee systems, Victoria, Canada) was used for photo treatment and IBM statistical package software v24 (IBM, Armonk, NY, USA) was employed for chemometric analysis.

Calcium quantification by atomic absorption spectroscopy procedure

Peel from cherry was removed by employing a knife and was divided in about 0.5 g for humidity analysis and around 1.0 g for calcium determination. Humidity was determined by placing the samples in an oven at 70 ºC until constant weight. For calcium determination, the peel was placed in a crucible and calcined at 500 ºC for 2 h. After calcination, 5 mL of hydrochloric acid 0.25 M was added to the samples and transported to volumetric flasks by filtering through a 0.45 µm nylon filter. The adequate diluted samples were measured in the atomic absorption instrument and interpolated in a calibration curve between 1 and 20 mg L^{-1} Ca^{2+} solutions prepared from $CaCO_3$.

Calcium quantification by smartphone photography and chemometrics

First, a calibration curve or evaluation group of 10 cherries for each variety was prepared. For this purpose, atomic absorption measured samples were previously photographed with an android smartphone and related to the calcium content obtained from the conventional method. Next, the photographed cherries were cut and normalized to the same size and resolution with ACDSee software. This step is important in order to obtain normalized matrixes in terms of size. Then, the normalized photos were opened in the OriginLab® software and converted to black and white and transformed to a matrix which has the same lines and columns than the resolution (200x200). Afterwards, line 100 was selected for further studies, which was more or less in the center of the photo. The 200 values of the lines corresponding to the 10 cherries of each variety where inserted in the SPSS software and an extra column with the calcium content extracted from the atomic absorption measurements was added. Subsequently, a multiple linear regression was made. The obtained equation was used for further determination of new samples by following the steps indicated above.

RESULTS AND DISCUSSION

Preparation of the fruits photos

One of the important characteristics of this methodology is the treatment of the photo since go out from the smartphone until the numeric data matrix. For this purpose, it is important to normalized the photography since not all the fruits have the same size and the pixel positions will be displaced from one photo to another photo. For this reason, the normalization of the pixels contents to 200x200 helps to the data matrix and subsequently the different treatments to have a better characterization. Also, the employment of the black and white photos instead the original or other RGB spectra is due to the reduction of variables in the data matrix, since if we use the RGB spectra we will need at least three different matrixes, one for each color, this will complicate the extraction of the data. Also, if we use the direct color photo, it is true that we will obtain only one matrix, which represents the intensity of the color that is in each pixel and it will not be normalized. For these reasons, the normalization and the conversion to black and white should be employed before do anything else.

Discrimination of varieties

In order to select the best data for the analysis a previous study based in the capacity of the matrix data to difference between the two varieties (Prime Giant and Irnet) was performed by employing a linear discriminant analysis (LDA) by stepwise. For this purpose, five lines equally distributed in the matrix (lines 1, 50, 100, 150 and 200) which have 200 pixels or data points were selected to study the best line to differentiate between the two varieties. Therefore, Wilk's lambda was selected as the information that should be used to select the best line. As can be observed in Fig. 1A, the best Wilk's lambda (λ_w), the smaller one, was found for the line 150 since is the one that obtained the highest –Log λ_w. Also, this first discriminant function was obtained with only four variables from 200 introduced. It should be remarked that line 1 although was able to classify correctly all the cases, only one variable was needed; therefore, the λ_w was really high and in the case of the line 100, that line was not able to discriminate both varieties, that is the reason because a high λ_w (1) was attributed to this line. On the Fig. 1B, the graphic of the discriminant function obtained from line 150 was plotted with the 10 cases of each variety.

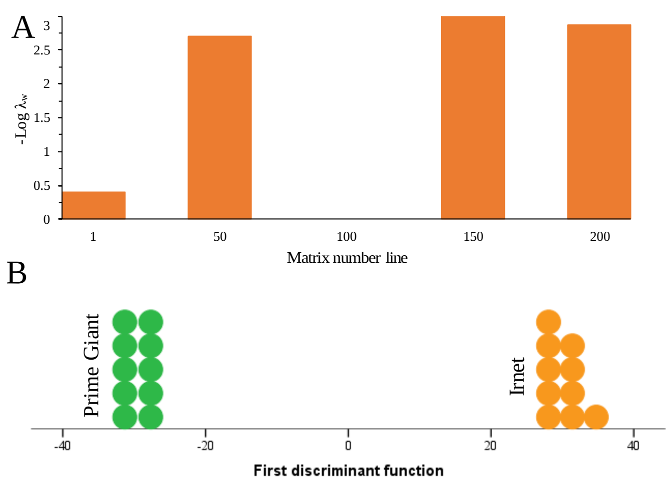

Fig. 1. –Log λ_w vs number of line (A) and representation of the first discriminant function for the matrix line 150 and the position of the 20 different samples in the function (B).

Calcium quantification by multiple linear regression

Since the line 150 of the data matrix was selected as the best one for the study. A multiple linear regression (MLR) was made with this line. As can be observed in Fig. 2, the predicted value and the actual value fits really good for Prime Giant (Fig. 2A) and Irnet (Fig. 2B) cherries, respectively. Also, the method was able to predict the content of calcium in cherries peel without taking into account the variety of the cherry as shown in Fig. 2C. The method to obtain the MLR was able to obtain equations for determination of calcium peel contents with a low amount of variables as can be observed in equations 1, 2 and 3 for Prime Giant, Irnet cherries and both together, respectively:

$$[Ca] = 1.1\cdot10^{-14}–1.3\cdot10^{-4}\cdot P_{24}+5.9\cdot10^{-4}\cdot P_{159}+1.9\cdot10^{-4}\cdot P_{177}-2.3\cdot10^{-4}\cdot P_{199} \quad (1)$$

$$[Ca] = 7.1\cdot10^{-15}+7.4\cdot10^{-4}\cdot P_{109}+4.0\cdot10^{-4}\cdot P_{150}-6.3\cdot10^{-4}\cdot P_{184}-1.8\cdot10^{-4}\cdot P_{199} \quad (2)$$

$$[Ca] = -2.7\cdot10^{-14}+7.8\cdot10^{-4}\cdot P_{15}-1.4\cdot10^{-4}\cdot P_{25}-1.1\cdot10^{-4}\cdot P_{59}-9.4\cdot10^{-5}\cdot P_{159}+2.2\cdot10^{-4}\cdot P_{177}-5.2\cdot10^{-4}\cdot P_{182}-9.9\cdot10^{-5}\cdot P_{185}+ +8.9\cdot10^{-5}\cdot P_{199} \quad (3)$$

where [Ca] is in mg Ca^{2+} g^{-1} dry peel and P_n indicates the pixel position in the line. For example, P_{100} is the number that can be found in the position 100 of the employed line which corresponds to the pixel 100 in the normalized photo. In view of the results, equation 3 was used for further studies.

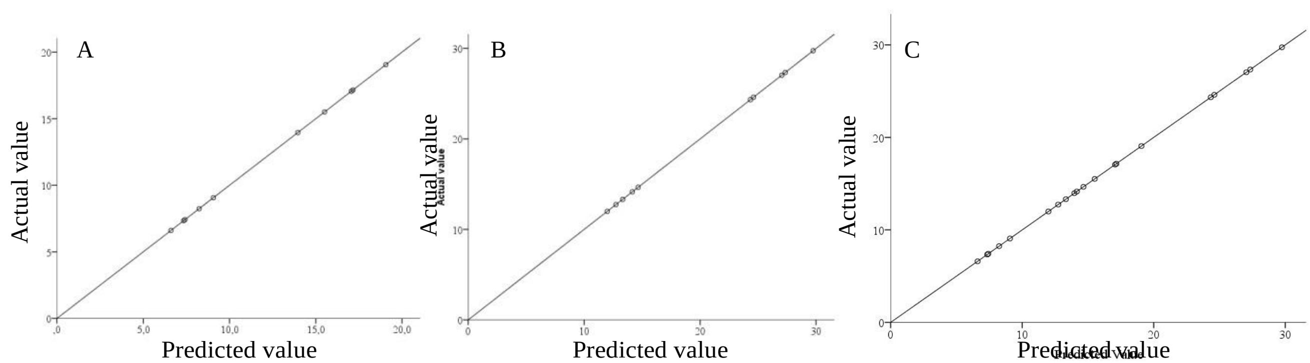

Fig. 2. Actual value plotted versus predicted value for equations 1 (A), 2 (B) and 3 (C).

Finally, the proposed method was employed with real samples not included in the calibration curve and compared with the ones obtained by the traditional atomic absorption (AA) spectroscopy methodology. As can be observed in Table I, both methodologies presented similar results which are in agreement with the quality of the presented methodology in this work.

Table I. Calcium peel contents founded for different samples in the two different methodologies

Variety	[Ca] (mg/g dry peel) by AA	[Ca] (mg/g dry peel) in this work
Prime Giant	**6.61**	**6.60**
	17.15	**17.16**
	17.06	**17.05**
	15.51	**15.51**
Irnet	**12.74**	**12.73**
	24.34	**24.33**
	29.74	**29.75**
	14.65	**14.65**

CONCLUSIONS

In this work, a new way to obtain important nutrient data from fruits was developed showing good results. The method, which employs the smartphone camera and a photo treatment, conversion to numeric matrix data and using LDA and MLR chemometric techniques is able to quantify the calcium content in cherries peels. As future perspectives, the method should be improved by adding new methodologies that helps to make easier the method, include new nutrient quantification as phosphorous or potassium and expand to other varieties of cherries and other fruits.

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