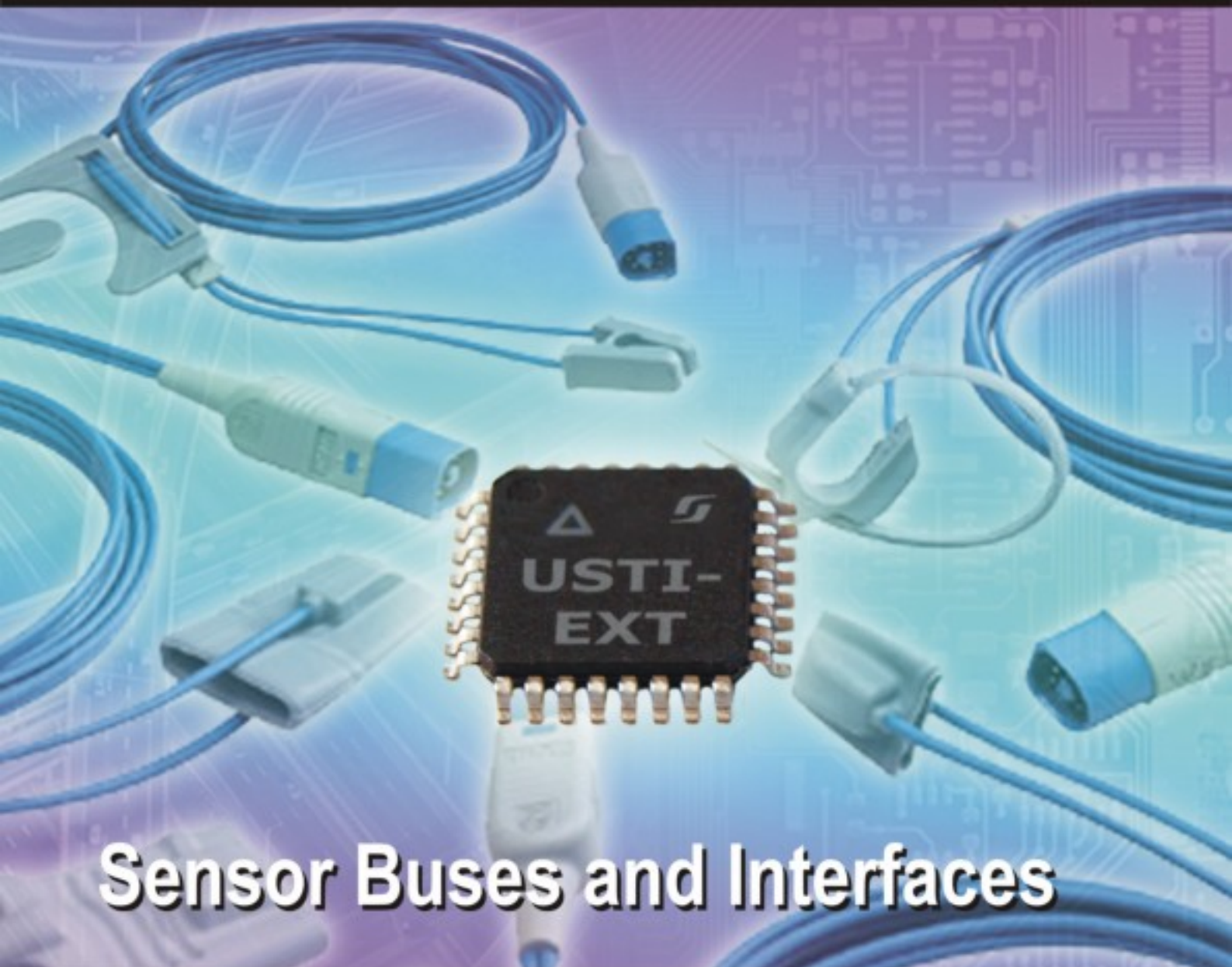


ISSN 1726-5479

SENSORS & TRANSDUCERS

vol. 140
5 / 12



Sensor Buses and Interfaces

International Frequency Sensor Association Publishing



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May 2012

www.sensorsportal.com

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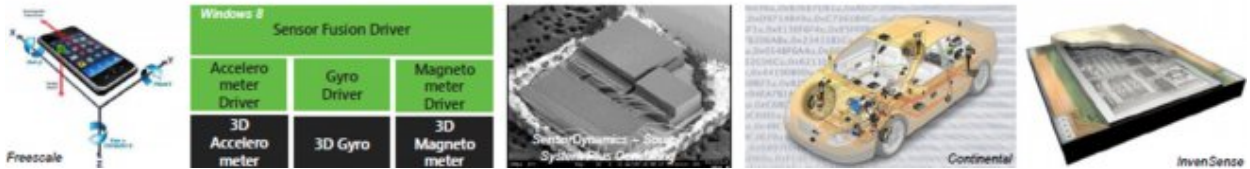
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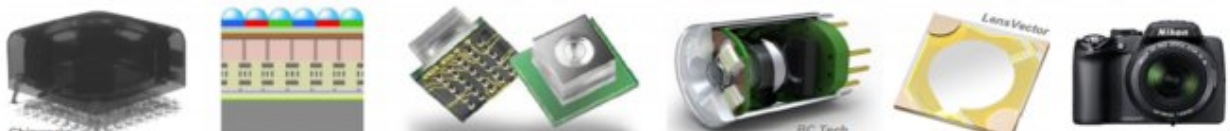
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Applications of Electronic Nose Based on MOX and QMB Sensors

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Received: 27 March 2012 /Accepted: 23 May 2012 /Published: 31 May 2012

Abstract: The electronic nose emerged in the mid 80's due that the food industry needed quicker instruments to be applied in food control for the characterization of smell, in order to complement the work done by human panels. But scientific researchers noticed that electronic nose could also be used in other fields, such as environmental contamination, health, defence, automotive industry, control of hazardous substances, etc. The aim of this research article is to explain how electronic nose work and to report the latest researches obtained with electronic nose in the Centre of Investigation of Solids (CINSO). *Copyright* © 2012 IFSA.

Keywords: Electronic nose, MOX sensors, QMB sensors, extra-virgin olive oil, garlic, tomato, volatile compounds.

1. Introduction

Of the five senses, smell has always been the most difficult to define. Understanding how it works has been the goal of researchers for many years. The smell depends on many chemicals that give a character and unique qualities [1].

The ability to measure and identify the optimal development of odour and taste characteristics is a constant crucial point in the development of many products [2]. The electronic nose can be defined as an instrument with sensors or chemicals columns which has a program containing a chemometric recognition model, which can distinguish and compare individual or complex odour in a sample.

The data obtained from electronic nose is qualitative, due that the aim of the instrument is to analyze and recognize the traces of complex odour by evaluating together the volatile components of the sample analyzed imitating the human olfactory system.

1.1. System Electronic Nose

The human olfactory system consists of a set of olfactory cells that interact with aromatic molecules. The signals are sent to the olfactory bulb and from there to the brain, which processes all the information [2, 3]

On the other hand, with electronic nose the system is similar as the human olfactory, the volatile molecule interacts with a sensor system or a column system, where each sensor or column equivalent to a human olfactory cell and the data provided are processed by a computer system in order to obtain a response equivalent to the odour fingerprint of the sample analyzed.

The use of electronic nose based on different sensor technologies has been suggested for the rapid detection of quality-related volatile compounds for various food products. Gas sensors that are commonly used in electronic noses are non-selective toward individual compounds but show sensitivity toward certain classes of compounds. This property induces their potential for monitoring quality, associating it with varying levels of different classes of produced volatile compounds.

The electronic nose consists of three basic components: the sampling system, the detection system (sensors or columns) and the system of data processing.

1.2. The Sampling System

The techniques used to obtain volatile compounds from the sample and to transport it to the sensor system or columns are by headspace techniques. These techniques are based on generating a vapor phase (headspace) in equilibrium with the sample, solid or liquid, at a given temperature.

The headspace is the amount of gases that are introduced into the sensor or column. The gases are volatile compounds obtained from the sample during its thermal equilibrium.

1.3. The Detection System

In the market there are three types of noses that differ mainly in the detection system: gas sensors, mass spectrometry and gas chromatography.

- Sensors gases: gas sensors exhibit the property of modifying any of its electrical properties of the odour when the compounds interact with its surface. A battery of sensor response is obtained which is the sum of the interactions made by all the volatile molecules and the resulting diagram can be interpreted as a fingerprint of the odour of the sample analyzed.

There are different types of sensors that can be classified according to material used: semiconductors doped tin oxide, conducting polymers and piezoelectric crystals (quartz microbalance).

- Mass spectrometry: sensors coupled to a mass spectrometer as detection system. The spectrum obtained is equivalent to the response of a multisensor with many sensors as ions formed.
- Gas Chromatography: The spectrum obtained is equivalent to the response of a multisensor.

1.4. Data Analysis

The amount of information obtained from the detection system is very large. To handle this large data set it is necessary to use statistical tools to visualize natural groupings of samples.

The first step is to process the sensor signals in order to transform the data into a suitable form for subsequent treatments. Next, the processed data can be analyzed using various statistical techniques: principal component analysis (PCA), cluster analysis (CA), SIMCA (Soft Independent Modelling Class Analogy), KNN (K-Nearest Neighbours) and principal component regression (PCR) [4].

2. Materials and Methods

Since 1998, the CINSO (Center for Research in Solid-CITEDEF CINSO /UNIDEF-MINCEF-CONICET), has worked with a prototype called electronic nose MOSES II. It was designed by the University Tübingen (Germany) GmbH and Lennartz Elektronische.

2.1. Electronic Nose

An electronic nose MOSES II (Modular Sensor System) was used to discriminate odours. MOSES II contains two modules of gas sensors, one of them composed of eight quartz microbalance sensors (QMB). This type of sensors consists of vibrating quartz crystals covered with polymeric selective coatings, on which gases are adsorbed. The initial vibration frequency (ν_0) of crystals decreases according to the mass increase because of the gases adsorption and the difference between ν_0 and the final frequency (ν_f) results proportional to the adsorbed gas concentration.

The other module of MOX sensors (SnO_2) is composed by eight pure and doped semiconductive SnO_2 sensors. Doping with different elements increases SnO_2 selectivity for different gases. The SnO_2 surface conductivity changes as the semiconductor adsorbs oxidising or reductive gases [16]. The adopted configuration results very flexible for general purposes and convenient for a wide range of applications.

These types of sensors allow us to make the fingerprint of the odour through the signals obtained from the set of sensors. Briefly, and as examples, some of the themes developed in the CINSO with the electronic nose will be mentioned.

2.2. Samples Analyzed with an Electronic Nose

2.2.1. Effect of Light on Odour Profile, Volatile Compounds and Vitamins of Extra-virgin Olive Oil

Extra-virgin olive oil varieties samples were properly obtained from fresh, mature fruits of good quality provided by an Oil mill from Chacras de Coria (Mendoza, Argentina)

Varieties of Argentine extra virgin olive oil were exposed to light and darkness during 24 weeks. Odour changes were monitored using an electronic nose. Samples from both varieties were placed in clear glass bottles of 500 mL, containing a headspace of 3% and hermetically closed.

Samples were located in illuminating equipment at a distance of 80 cm from the lights tube (D65 Osram-temperature: 4227 8C and luminance of 1580 lux). Samples were exposed to light and

darkness during 8 h, 5 day's a week during 120 days. Analysis was carried at intervals of 2 weeks until 24 weeks. Procedure was repeated thrice and triplicate samples were taken each time.

2.2.2. Effect of Frying on the Odor Profile of Olive Oil Extra Virgin, Volatile Compounds and Vitamins

Changes in the odour profile of extra virgin olive oil varieties (Arbequina (ARB) and Arauco (ARA)) were monitored during frying process with an electronic nose.

For thermal oxidation, frying pans were filled with 600 mL of extra virgin olive oil (EVOO) and heated at 180 °C. Oil samples were obtained at the beginning of the experiment and then, four times at intervals of 15 min (t_{15}) during 60 min (t_{60}). Frying procedure was repeated thrice and triplicate samples were taken each time.

2.2.3. Electronic Nose Study of Powdered Garlic

Garlic cloves of eight different cultivars (Fuego, Sureño, Perla, Castaño, Gostoso, Nieve, Norteño and Unión (being the original Spanish names given at INTA preserved) cultivated at INTA-La Consulta, Mendoza (Argentina) were peeled, weighted, cut into slices and dehydrated by two different methods: I) Oven-drying and II) lyophilization.

In the first case, samples were dried in oven at 50 °C for 24 h. The second process consisted in lyophilization in a Virtis-Freeze mobile (12 L). Freezing in liquid nitrogen was performed in order to produce an immediate cooling and to avoid the enzymatic hydrolysis of precursor compounds to preserve the flavour. Study of the odour profile of garlic was applied to discriminate garlic specimen dried by lyophilization and oven-dried and humidified specimen (before dried by both techniques) for each cultivar.

Samples remained in the lyophilizing equipment for 24 h and then, they were placed in sealed vials to preserve them from humidity until electronic nose measurements were performed.

2.2.4. Tomato Quality during Short-Term Storage Assessed by Colour and Electronic Nose

Wild-type tomato plants cv. Money Maker and tomato plants overexpressing and silencing *Asr1* gene under the control of promoters 35S and B33 were grown under controlled conditions in a greenhouse (200 $\mu\text{mol PARs-1 m}^{-2}$, 60% RH, 23 °C).

Fruits were harvested manually from plants grown in the National Institute of Agropecuary Technology, during the summer at the ripening stage 5 (light red) (USDA colour chart, 1975). Fruits of uniform shape and size and free from fungal infection were selected. After harvest, fruits were washed with a solution of hypochlorite (150 ppm de Cl_2 as hypochlorite of sodium), air-dried at atmospheric temperature, and individually labelled and weighed. Samples were kept at 19 ± 0.5 °C and 85% RH and analyzed weekly (7 days) for three weeks (21 days).

An assay based on an electronic olfactory system was set to evaluate tomato fruits by sensing the aromatic volatiles during post harvest storage of 21 days at 19 ± 0.5 °C in darkness. Olfactory system measurements were coupled with colour values. Odour profile and senescence parameters were carried out at 7-day intervals.

3. Results and Discussion

3.1. Effect of Light on Odour Profile, Volatile Compounds and Vitamins of Extra-virgin Olive Oil

Principal Component Analysis applied to analyze data obtained from the responses of the eight sensors of SnO₂ doped (MOX) samples for Arauco extra virgin olive oil exposed to light and darkness showed a principal component (PC₁) which explained 84.4 % of the total variance. The CP₁ was correlated positively with SnO₂ sensors (data not shown). On the other hand, Arbequina extra virgin olive oil samples subjected to light and darkness, explained 95.6 % of the total variance and was also correlated positively SnO₂ with sensors (data not shown).

Fig. 1 shows the average of the eight SnO₂ doped sensors (MOX) as a function of Principal Components (PC₁) in relation to exposure (light and darkness) as a function of time (weeks) for Arauco and Arbequina extra virgin olive oil.

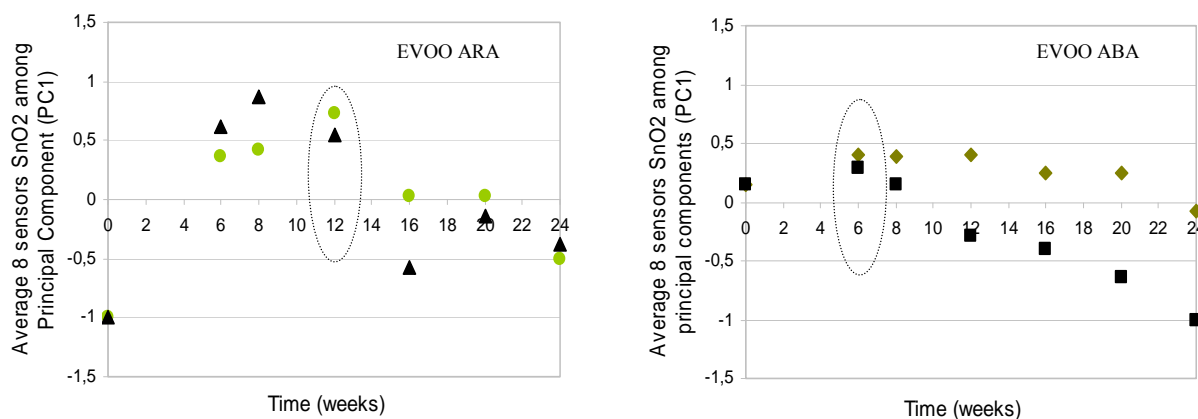


Fig. 1. Average of eight sensors of SnO₂ on CP₁ in olive oil extra virgin (EVOO) of: (a) varietal Arauco (ARA) exposed to light (□ ●) and darkness (▲) as a function of time (weeks) and (b) varietal Arbequina (ARB) exposed to light (◆) and darkness (■) as a function of time (weeks).

Arauco extra virgin olive oil at initial time (t_0) with light and darkness exposure showed a negative value on the PC₁, indicating a low response of the sensors. On the other hand, olive oil exposed to light after 12 weeks showed an increase in response of the sensors. Showing a defined separation between light and darkness exposure.

On the other hand, the data obtained from the responses of SnO₂ doped sensors for Arbequina Olive oil, showed a slightly positive value on the PC₁ at initial time (t_0) for samples exposed to light and darkness. Arbequina varieties showed a notable change in the odour from week 6, compared to Arauco (after 12 weeks of exposure) [5].

Analysis of Variance (ANOVA) results of volatile compounds (3-methyl butanal; *n*-pentanal; *n*-hexanal; *n*-heptanal and *n*-nonanal) in both varieties did not show significant differences between light and darkness exposure. Tocopherols (alpha and gamma) showed an important degradation for both varieties after light and darkness exposure, the most important decrease for both varieties was that corresponding to light exposure.

3.2. Effect of Frying on the Odour Profile of Olive Oil Extra Virgin, Volatile Compounds and Vitamins

Analysis applied to data obtained by electronic nose shows the grouping of extra virgin olive oil (EVOO) of Arauco (ARA) and Arbequina (ARB) as a function of different frying times.

One PC₁ was found, accounting 96.6 % of the total variation. PC₁ showed a positive correlation between MOX (doped SnO₂) sensors (S1: □, S2: □, S3: □, S4: □, S5: □, S6: □, S7: □, S8: □) and EVOO (ARA) for all frying times: 15 min (▲), 30 min (▲), 45 min (▲), 60 min (▲). Conversely, EVOO (ARB) showed a negative correlation with doped SnO₂ sensors for all frying times (15 min (●), 30 min (●), 45 min (●) and 60 min (●)).

Fresh samples for EVOO ARA at initial time (▲) and ARB at initial time (●), were located on negative PC₁. EVOO (ARA) corresponding to frying time 60 min (▲) showed the highest positive scores representing the highest sensors response (S1: □, S2: □, S3: □, S4: □, S5: □, S6: □, S7: □, S8: □).

The analysis of variance showed an increase in the production of volatile compounds, in particular: 3 - methyl butanal, *n*-pentanal, *n*-hexanal, *n*-heptanal and *n*-nonanal for both varieties [6, 7].

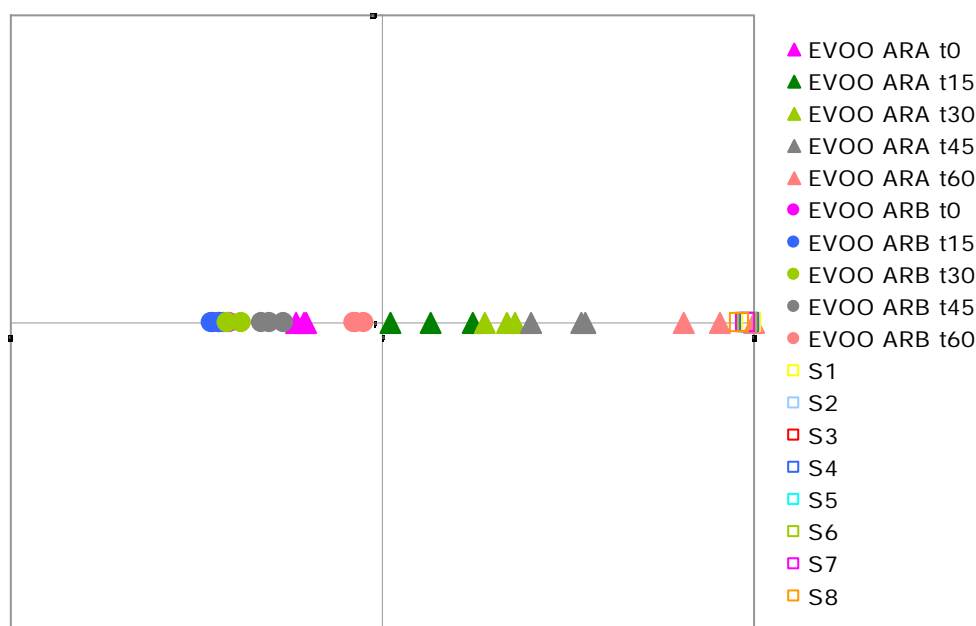


Fig. 2. Principal components analysis of electronic nose data corresponding to different frying time of extra-virgin olive oil (EVOO) for ARA (Arauco), at initial time (▲), 15 min (▲), 30 min (▲), 45 min (▲), 60 min (▲), and ARB (Arbequina), at initial time (●), 15 min (●), 30 min (●), 45 min (●) and 60 min (●) with different doping of SnO₂ (S1: □, S2: □, S3: □, S4: □, S5: □, S6: □, S7: □, S8: □).

3.3. Electronic Nose Study of Powdered Garlic.

3.3.1. Oven-dried and Lyophilized Samples

Applying the stepwise linear discriminant analysis to the principal components of the 16 signals (eight corresponding to MOX and eight to QMB sensors) properties, a 95 % successful classification was reached with cross validation. Values of the two first discriminant functions of each specimen,

accounting 95.7 % of the system variability (77.3 % for the first and 18.4 % for the second one). Two groups were defined according to samples oven-dried and lyophilized (data none shown).

3.3.2. Oven-dried and Lyophilized Samples with and Without Humidification

Including all the samples (dried and lyophilized, with and without humidification) a model could be built reaching a correct 87.3 % classification, by cross validation (69 success and 10 errors) applying the stepwise LDA on the principal components of the sensor curves properties (data of electronic nose). The three discriminant principal functions explained 95.6 %, 3.3 % and 0.5 % of the variance, respectively. Fig. 3 represents the bidimensional plot of the two first functions (summarizing 98.9 %). It enabled to observe that every non humidified specimen exhibited a similar behaviour, clustering in the inferior left zone of the plot. It is also observed that by humidification, the behaviour of oven-dried powders resulted different from that of lyophilized powders exhibiting a clearer separation among cultivars.

This fact enabled to infer that lyophilized powders (either dry or humidified) better retained their odour profile than oven-dried powders, maintaining the properties of each cultivar [8]

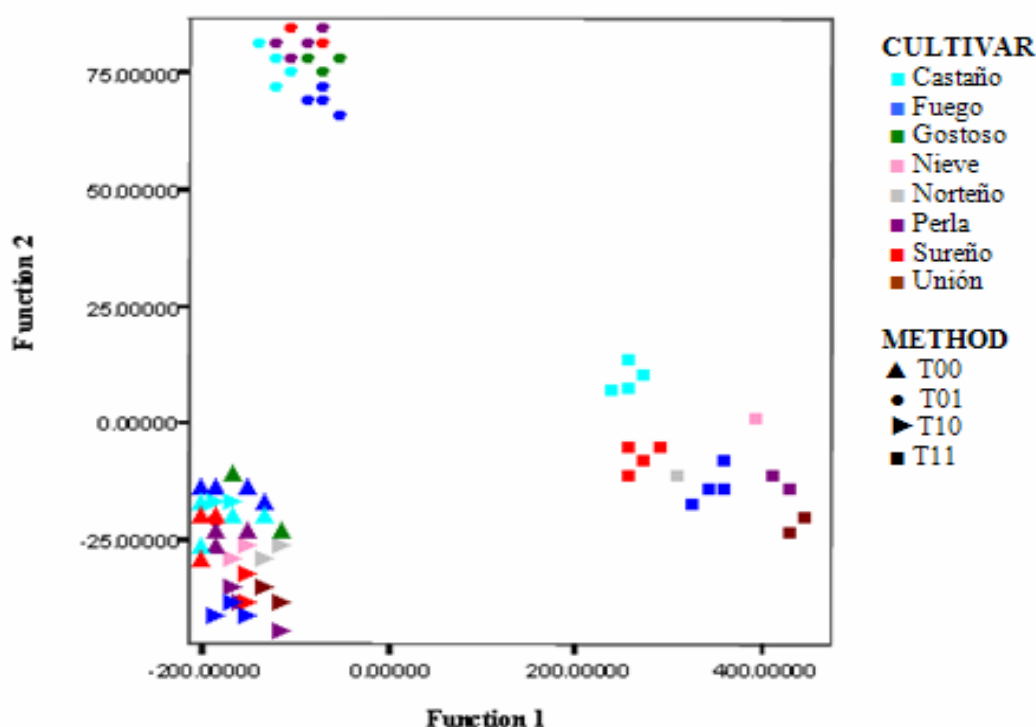


Fig. 3. Stepwise linear discriminant analysis applied on the principal components of the properties of the sensors curves of the electronic nose (using all the cases) for oven-dried-non humidified with distilled water (T00: ▲); oven-dried- humidified with distilled water (T01: ●); lyophilized-non humidified with distilled water (T10: ►) and lyophilized- humidified with distilled water (T11: ■) cultivars (Castaño: ■, Fuego: ■, Gostoso: ■, Nieve: ■, Norteño: ■, Perla: ■, Sureño: ■, Unión: ■).

3.4. Tomato Quality during Short-Term Storage Assessed by Colour and Electronic Nose

Discriminant function analysis applied to electronic nose data for MOX sensors showed three components, accounting for 99.2 % of the total variance. In the present assay, separation among groups according to storage time (0, 7, and 14 days) was observed for wildtype. Over expressed

(Money Maker) lines/plants of tomato showed difference between odour profile for day 0 and day 21, even though a no clear discrimination between 7 and 14 days was observed (Fig. 4).

Fruit lost weight almost linearly with shelf life ($P < 0.001$) presenting an averaged loss of 21% ($r^2 = 0.98$) for over expressed (Money Maker) lines/plants, 13 % ($r^2 = 0.97$) for silenced (Money Maker), and 14 % ($r^2 = 0.98$) for wild type during 21 days of storage.

Colour values L^* , a^* , and b^* data showed that colour properties changed during storage for all the lines considered. Correlations between odour profiles and colour parameter were obtained showing that the electronic nose is a useful technique for monitoring short-term storage of tomato [9].

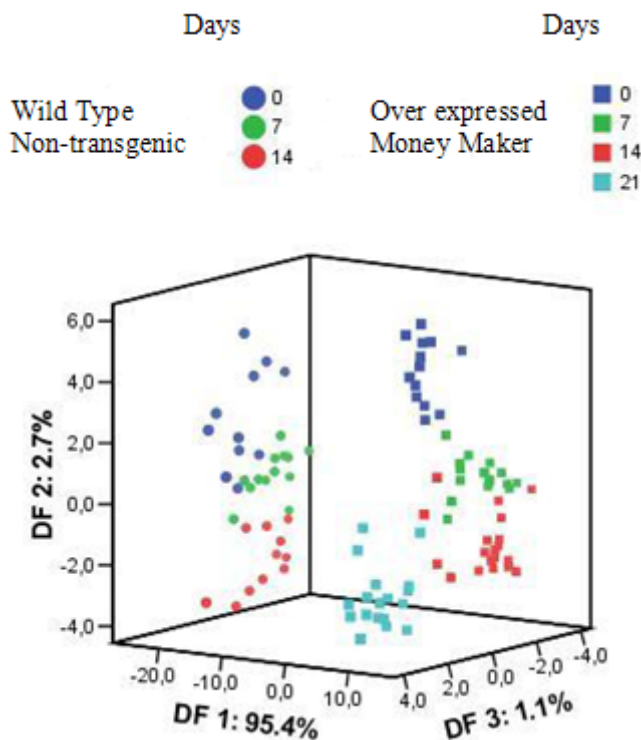


Fig. 4. Discriminant analysis of electronic nose for transgenic tomatoes (Money Maker over-expressed) and non-transgenic (WildType) as a function of storage (Money Maker over-expressed: ● day 0; ● day 7; ● day 14 and ■ day 21) and non-transgenic (WildType: ■ day 0; ■ day 7; ■ day 14).

4. Conclusion

Studies carried out with the electronic nose system based on the headspace were satisfactory and showed that this technique may have wide application, not only for the food industry but in other areas of interest. It is important to mention that they act quickly and objectively, allowing it to be a complementary tool for sensory analysis.

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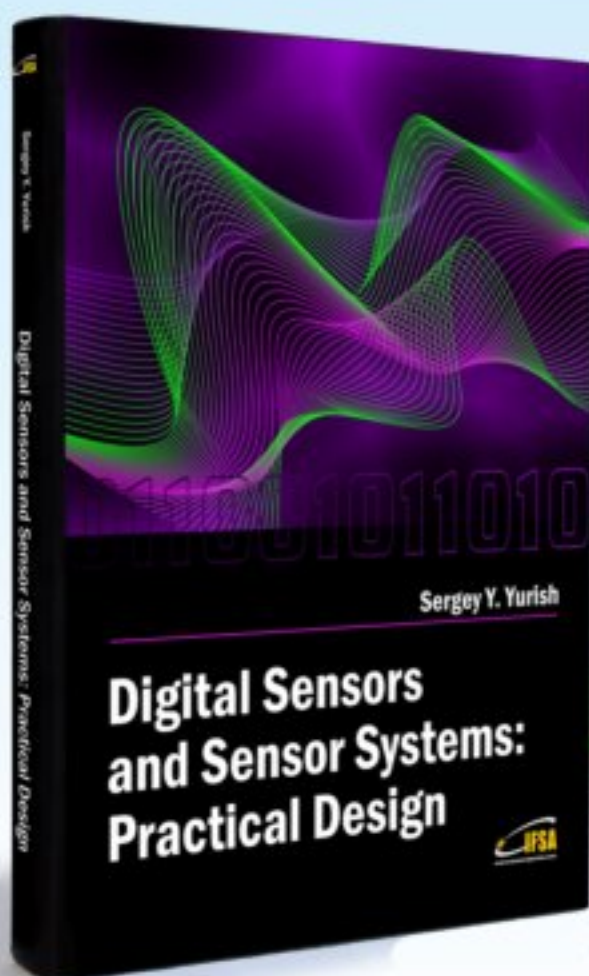
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