Electronic Tongue applied to beef quality: a first approach

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Abstract — **This study investigate if the Electronic Tongue can be a useful and inexpensive tool for the qualitative analysis of fresh meat. Analyses were conducted mainly on beef broth to assess the ability of the instrument to separate significantly the samples according one or more classification variables. As an example protocol was applied to 29 finishing bulls (final LW=565±89.6 and age=468±73.3 days) randomly divided into three groups: T1 (n=8), T2 (n=8) and C (n=12). The T1 and T2 groups were respectively treated with two different illegal hormonal substances. The third group (C) served as control. On samples of M. *Longissimus thoracis* analyses were conducted by an ET with 8 ISE sensors sensitive at: $Ca^{2+}Mg^{2+}$, Cl^{-}, CO_3^{2-}, Cu^{2+}, NH_4^{+}, NO_2^{-}, Pb^{2-}, Zn^{2+}. The implemented protocol needs 50g of minced cooked meat, cooled for 18h (0°C ÷ +4°C), heated to the analysis' temperature (25°C) and measured three times for 180s with intervals of 5s between recordings, using a Faraday cage. The Canonical discriminant analysis shows a significant separation among control and treated groups. The first canonical variable explains the 70% of between-class separation and divides T1 from T2 and C groups due to the Ca^{2+}-Mg^{2+} and Zn^{2+} sensors. The second canonical variable divides T1 and T2 groups from the Control group due to the pH and Cl^{-} sensor. The Electronic Tongue seems to be a useful tool to identify meat subject to illegal treatments.**

Keywords - **Beef, Meat Quality, Electronic Tongue.**

I. INTRODUCTION

The characteristics of food are usually evaluated by sensory panel or by instrumental analytical techniques [1] that often require laborious sample preparation with time delays, high costs and the need for qualified personnel [2].

In addition, maintenance costs and consumption of reagents may discourage their use in small industries and/or laboratories [3].

To solve these problems in recent years, several alternatives have been evaluated with the development of sensor systems for the discrimination of complex matrices such as the Electronic Tongue [4, 5, 6, 7]. This approach offers the possibility of real-time monitoring matrices of interest, even by remote control, without pre-treatment of complex sample and without removing interference [8, 9]. These systems mimic the behaviour of the human senses and rely on the use of sensor systems with low selectivity (responding to a number of substances in the sample) and high cross-sensitivity, combined with a chemometric means for a multidimensional processing of information [3]. All the responses of the ET sensors create a "map" of non-specific signals that constitute the "product profile" also called "food fingerprint". These systems have advantages such as low cost, ease of use, response speed, versatility, no sample pre-treatment and objectivity of analysis, besides being useful support to chemical-physical and sensory analysis. Also, the cost of such instruments can be further amortized when repeated and continuous controls are required [10]. The objective of the ET is not to measure the individual components, but to obtain, in general, images more similar to "human aspects" and its attributes (taste, ripeness, quality, etc.), which are common complex systems [11, 12].

The ET can be applied to qualitative analysis (recognition, classification and identification), quantitative analysis (determination of the concentration of multiple components) or both types simultaneously, depending on the membership of the sensor system and the method of data processing used [9, 13].

Often in some fields is more important to have the tools for rapid qualitative analysis of arrays, rather than complex quantitative analysis. For example, in the fields of food industry where food is prepared, marketed and/or stored there is the need for tools for quick and easy evaluation of the quality [2].

The purpose of this study is the application of ET for the recognition of meat, with the ultimate goal of creating a Electronic Tongue protocol (ETp) to be incorporated in our manual of procedures for assessing overall quality of the meat [14, 15, 16], in order to make on the same single sample analysis better correlated with each other. This protocol could be a useful tool to define the meat quality profile efficiently and could help meat industry to provide appealing products and consumers to get reliable information. To achieve the goal we have worked on different aspects of the ET analysis in order to optimize the reliability of processes according the practical and economic aspects.

II. MATERIALS AND METHODS

Analyses were conducted with the ET LiquiLAB (ECOSENSE, Italy), which uses a system of potentiometric electrochemical sensors. The instrument consists of a measuring device, a temperature sensor in stainless steel (-30 ÷ +120°C), eight ISE electrochemical sensors (Sensor System, Russia) and a standard commercial electrode to calomel 303/SCG/12 (Amel Electrochemistry, Italy). The measuring device has 8 high impedance electrode ISE inputs, an input for the reference electrode, an input for temperature sensor and a RS232 serial output for connection to the computer. The software, supplied with the instrument, is the LiquiLAB Light (ECOSENSE, Italy) which allows the execution of the analysis by setting the time length, the file name, the conversion of sensors' input, the data output expressed in mV and the opportunity to follow *on-line* (Labview) the trend of each sensor or as a whole and of the temperature. During the execution of the measure data is stored in a text file ".lqb" fully compatible with programs reading files ".txt".

For this experiment, were used 8 ISE electrodes sensitive to: Ca^{2+}-Mg^{2+}, Cl^{-}, CO_3^{2-}, Cu^{2+}, NH_4^{+}, NO_2^{-}, Pb^{2-}, Zn^{2+}. These electrodes provide different signals, for example the electrodes Ca^{2+}-Mg^{2+}, Cu^{2+}, NH_4^{+}, Pb^{2-} e Zn^{2+} with increasing concentration of ions provide an increasing signal (mV), while the electrodes Cl^{-}, CO_3^{2-} e NO_2^{-} behave opposite way.

During the test were used a pH meter PH 25 with an electrode for liquids 50 T (Crison, Spain), a Faraday cage, a bain-marie equipped with a mechanical agitator at 60 rpm.

As an example protocol was applied to 28 finishing bulls (final LW=565±89.6 and age=468±73.3 days) randomly divided into three groups: T1 (n=8), T2 (n=8) and C (n=12). The T1 and T2 groups were respectively treated with two different illegal hormonal substances. The third group (C) served as control. The implemented protocol needs 50g of homogenized meat (M. *Longissimus thoracis)* and 250 g of deionized water mixed in a cooking beaker, and inserted into a support with an agitator at 60 rpm. The sample was placed in a bain-marie at 85°C and, after reaching an internal temperature of 70 °C, cooked for 10 min by controlling the temperature with a thermocouple 10K Temp (Oakton, USA). The filtered broth was cooled for 18 h (0 °C ÷ +4 °C). The process of cooking and cooling was carried out to remove as much fat possible that could damage or make lose sensitivity to the electrodes, as well as to extract the greatest amount of analytes. After the cooling phase the broth was filtered again with a fine mesh filter added with two gauzes to remove any remaining meat and presence of fat. Finally the broth was heated to the temperature for analysis between 20 and 25°C (room temperature) through a bain-marie set at 25°C. Each sample was measured three times for 180s each with intervals of 5s between recordings, using a Faraday cage.

Data analysis was performed by SAS/ STAT in SAS 9.2 [17] using one-way analysis of variance (GLM) and treatment as independent variable. Results are expressed as LSmeans and MSE. To assess the reliability of the instrument were considered the coefficient of variation. Parameters were then submitted to a Canonical Discriminant Analysis, a dimensional reduction technique that performs both univariate and multivariate one-way analysis to derive canonical functions, i.e. linear combinations of the quantitative variables that summarize the variation among groups. Finally a Discriminant Analysis was applied to evaluate the model.

III. RESULTS AND DISCUSSIONS

To evaluate the effectiveness of the developed protocol and the ET instrument were analyzed over 150 samples, measured 3 times each. Here are reported the statistical results relating to an experimental design with 28 finishing bulls treated with illegal substances.

Table 1 shows the coefficients of variation (CV) of the parameters under consideration that vary between 3.1% to 16.7%, except for the sensors Cu^{2+} and Zn^{2+} showing the highest variability (30.4% and 21.7%) that can lead to more errors in samples discrimination. Compared with the CV of other parameters used in the analysis of meat quality are very interesting and this argues in favor of their reliability in the discrimination of samples. Palka and Daun [18] measuring fiber diameter found a CV ranging from 13% to 24% and for sarcomere length one of 3-8%. Bickerstaffe et al. [19] measuring shear force on beef by a MIRINZ tender meter, found a CV ranging from 11% to 45%. Denoyelle and Lebihan [20] for tenderness found a CV ranging from 15% to 20% and for compression from 15% to 24%. Moelich et al. [22] measuring cooked pork found a CV ranging from 13% to 14% for cooking loss, WB peak shear force from 11% to 13% and tenderness from 8% to 31%. Barbera and Tassone [15] measuring meat cooking shrinkage by MCS was found a CV ranging from 6% to 12%. Kim et al. [23] measuring WHC found a CV ranging from 1.5% to 10%, for tenderness from 5% to 15% and for juiciness from 12% to 20%.

The univariate analysis (Table 1) shows Ca^{2+}-Mg^{2+}, NO_2^-, Zn^{2+} sensors and pH of broth are able to separate all three groups. The C group has the highest Ca^{2+}-Mg^{2+} signal, the intermediate NO_2^- signal and the lowest Zn^{2+} signal.

The Canonical Discriminant Analysis (Figure 1) shows a significant separation between control and treated groups. The first canonical variable accounts for the 70% of the total variability and divides T1 group from the others due to the Ca^{2+}-Mg^{2+} and Zn^{2+} sensors. The second canonical variable accounts for the remaining 30% and divides the two T1 and T2 groups from the Control group due to the pH and Cl^- sensor.

Finally a Discriminant analysis was applied to evaluate the model. In classification the accuracy was interesting with a 7% of total misclassification error. In cross-validation the accuracy was poor. The C group had 33% of misclassification error, T1 and T2 groups 50%. The ET approach to the qualitative analysis could contribute to classify meat treated by illegal substances. The effectiveness is variable depending on the used substance.

Table 1. LSMeans, MSE and Coefficient of Variation of the ET sensors (N=28)

LSMeans by sensors in the same row with different letters are significantly different (a, b: P<=.05; A, B : P<=.01)

		GROUPS				CV
Sensors		C	T1	T2	MSE	%
Ca^{2+}-Mg^{2+}	mV	99.78^{a}	95.96^{b}	95.57^{b}	17.672	4.64
Cl^-	mV	96.38	99.08	99.73	39.142	6.33
CO_3^{2-}	mV	171.69	171.29	161.72	649.255	14.78
Cu^{2+}	mV	-54.31	-51.55	-58.55	291.450	30.36
NH_4^+	mV	85.95	86.48	75.21	180.049	16.69
NO_2^-	mV	219.09^{A}	223.52^{A}	189.26^{B}	475.390	12.08
Pb^{2-}	mV	-356.8	-355.4	-361.8	123.66	3.08
Zn^{2+}	mV	-28.28^{a}	-34.14^{b}	-30.20^{ab}	40.712	21.71
Temp. broth	°C	24.4	25.0	23.9	2.31	6.21
pH broth		5.94^{a}	5.99^{ab}	6.30^{b}	0.143	6.54

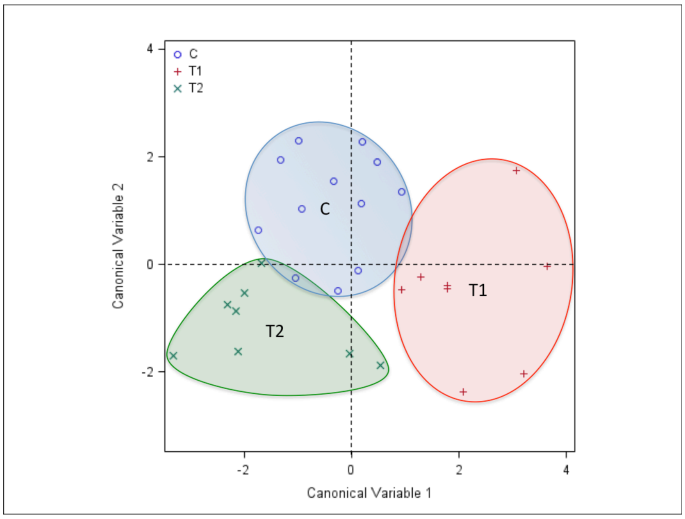

Figure 1. Canonical discriminant analysis of ET sensors

IV. CONCLUSIONS

The steps undertaken to create the ETp led to a standardized process for the extraction of analytes well integrate in the protocol for the global qualitative assessment of the meat developed at the Department of Scienze Zootecniche. The significant results show that the ET could be an interesting tool for assessing the meat. The future prospects for this tool is a low cost method with large and prolific perspective.

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