

The Thesis of the PhD dissertation

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**APPLICATION OF ELECTRONIC TONGUE AND NEAR INFRARED
SPECTROSCOPY TO DETECT ADULTERATION OF SOME FOODS WITH HIGH
ECONOMIC VALUE**

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1. INTRODUCTION

Food quality is an important aspect of the food industry that has gained increased attention with the continuous development of living standards and the relative change of dietary structure. There is a rising and persistent increase in demand for nutritious and safe food owing to the recent surge in fraudulent activities in the food industry, especially for foods with high economical and global value such as wine, protein powders and meat. Chemical procedures (conventional methods) used for monitoring food quality include the Kjeldahl method (for crude protein), Soxhlet Extraction Method (for crude fat), pH meter (acidity/alkalinity), colorimeter (for color) (Pugliese and Sirtori, 2012). Some of these methods however, are tedious, destructive, time consuming and in some cases expensive and often do not give a complete overview of the food being investigated.

Challenges such as these have led to the exploration of rapid alternatives such as near-infrared spectroscopy (NIRS), which can provide information through vibrational bonding in the form of overtones and combination bands in the wavelength range 750-2500 nm. In spite of the low intensity, the band shape often defines a single compound or group of compounds, making this technique suitable for qualitative and quantitative purposes for food authentication. In addition, the emergence of handheld devices capable of remote analysis could also be explored for practical quality control purposes. Another rapid alternative for food quality control, is the electronic tongue (e-tongue). The International Union of Pure and Applied Chemistry (IUPAC) defines the electronic tongue as “a multisensory system consisting of a number of selective sensors and uses advanced mathematical procedures for signal processing based on pattern recognition and/or multivariate data analysis” (Vlasov et al., 2005). In comparison with the human tongue, e-tongue has improvements in the sensitivity, selectivity, and multiplexing capacity of modern biosensors (Perumal and Hashim, 2014). It is capable of providing rapid, real-time, accurate and reliable data about various samples under study and has gained fame in the pharmaceutical, cosmetics, environmental control, engineering (petroleum), agriculture, food beverage industries.

Both e-tongue and NIRS require chemometrics and multivariate data analysis for results interpretation. Multivariate data analysis in food authenticity are simply, mathematical measures that can be employed to provide real-time and reliable statistical information about a production variation or deviation from quality, otherwise known as fingerprinting.

2. OBJECTIVES

The aim of this thesis was to develop rapid techniques for the determination and prediction of foods with economical and global value using near infrared spectroscopy (NIRS) and electronic tongue (e-tongue). There were primarily two main objectives.

The first main objective was to apply a benchtop spectrophotometer and handheld spectrophotometer to develop models that could discriminate, classify and predict:

1. Different classes of Tokaji wine and also, to classify inferior Tokaji wines that were adulterated with grape must concentrate and sucrose at different sugar concentrations.
2. Whey, beef and pea protein powders that were adulterated with four nitrogen-based adulterants: urea, glycine, taurine and melamine at very low concentrations.
3. Whey, beef and pea protein powders that were adulterated with four nitrogen-based adulterants by emulating practical situations through scanning through a low-density polyethylene (LDPE) plastic bag using a handheld spectrophotometer.

The second main objective was to apply a potentiometric e-tongue to:

1. Develop models that could discriminate, classify and predict different classes of Tokaji wine and also, to classify inferior Tokaji wines that were adulterated with grape must concentrate and sucrose at different sugar concentrations.
2. Determine the optimal dilution level of meat extract for e-tongue evaluation in the meat industry and also to develop three standardized sample preparation methods for meat analysis with the e-tongue, using red meat and poultry adulteration as case studies.

3. MATERIALS AND METHODS

3.1. Determination of Tokaji Aszu and Tokaji Forditas wine adulteration

Grape must concentrate (G.M.C) and four Tokaji wines of different quality grade (in increasing order of quality): Tokaji Forditas II, Tokaji Forditas I, Tokaji Aszu II and Tokaji Aszu I, were obtained from experts at the winemaking Tokaji region of Hungary. The wines were produced according to the standard requirements of Tokaji wine production as described: maceration of first class botrytized berries with base wine (fermented grape juice) to get Tokaji Aszu I wine and maceration of second class botrytized berries with base wine to get Tokaji Aszu II wine. Tokaji Forditas I and Tokaji Forditas II wines were prepared by reusing the botrytized berries from Tokaji Aszu I and Aszu II wine production for a second maceration with base wine, which is the normal practice in making Tokaji Forditas wines. The wines had a maceration period of 48 hours and resulted in diverse sugar concentrations that were determined by HPLC at the Department of Oenology, Szent Istvan University (now, Hungarian University of Agriculture and Life Sciences).

Tokaji Forditas I and Forditas II wine wines were manipulated with G.M.C of 775.3 g/L sugar concentration to mimic the sugar concentrations of the authentic Tokaji wines. The adulteration was done in steps, resulting in four different adulteration levels (C1 -C4) each for Forditas I wine and Forditas II wine adulteration. The sugar concentration of the adulterated wines covered the range of sugar concentration generally existing in the authentic Tokaji wines. The purpose of this artificial adulteration was to monitor the possibility of producing wines of similar quality to the authentic Tokaji wines by sugar manipulation. Also included was a complex wine referred to as “Base_sugar” in this study. This wine contained sucrose during its refermentation and was not macerated with botrytized berries, but was refermented after the addition of sugar. It was meant to represent complex forms of adulteration. Three repeats of each sample were prepared using a 50% dilution factor as this was reported to be the recommended dilution level for wine analysis with the electronic tongue (Soós *et al.*, 2015).

3.2. Determination of optimal dilution and optimal extraction for meat analysis with the e-tongue

Fresh poultry (turkey and chicken breast) and red meat (beef and pork thigh) were purchased from reputable supermarkets (SPAR) in Budapest, Hungary and transported to the laboratory for processing and analysis. One kilogram of each meat type was minced in the laboratory with a locally made mincer before commencing the experiments.

3.2.1. Determination of optimal dilution

Minced turkey and chicken were artificially adulterated to four different concentration levels: 100%, 97%, 95% and 90% w/w of turkey/chicken to have a total of 20 g per sample (meat mixture concentration). Each meat mixture was extracted by transferring the sample into a 200 mL volumetric flask and filled up to volume with distilled water then homogenized by vigorously shaking in the flask for 3 minutes. It was then filtered using a wire mesh filter (1mm pore size) to obtain the stock filtrate. This method was referred to as the “Raw meat extraction method”. From the stock filtrate of each of the prepared meat mixtures, 0.5%, 1% and 2% w/v dilutions were prepared by pipetting 5 mL, 10 mL and 20 mL respectively, into separate 100 mL volumetric flasks and homogenized by shaking the flask for 3 minutes before filling up to volume with distilled. transferring into 100 mL glass beakers for e-tongue analysis.

3.2.2. Determination of optimal extraction method

Three different extraction methods were tested for meat analysis with e-tongue: “raw meat extraction method”, “frozen meat extraction method” and “cooked meat extraction method”. Raw meat extraction method: 20 g of meat mixture was extracted as described in the experiment for the determination of optimal dilution to obtain the stock filtrates. Frozen meat extraction method: 20g of meat mixture was stored by freezing at a temperature -18 °C. The Frozen samples were removed on the second day of storage, put into a water bath of 50 °C for 20 minutes for them to thaw then, prepared in a similar way as the raw meat extraction method to obtain the stock filtrate. Cooked meat extraction method: 20 g of meat mixture was put in a cooking pot containing 200 mL distilled water at room temperature, covered with the lid and boiled for five minutes. It was afterwards, filtered with a wire mesh filter (1 mm) to obtain the stock filtrate. Six defined concentrations (100%, 99%,97%, 95%, 90% and 80% w/w) of poultry and red meat mixtures were used to evaluate all the three described sample preparation methods. The efficiency of the three developed mixtures was determined by their ability to discriminate the different meat mixtures after e-tongue analysis with diluted extracts prepared from the three methods. Three repeats were prepared for each meat mixture concentration level resulting in a total of 18 samples for poultry and red meat adulterated mixture respectively, for each of the three sample extraction methods.

3.3. Determination of whey, beef and pea protein powder adulteration

Whey protein powder, beef protein powder, pea protein powder, taurine, and glycine were provided by SCITEC Ltd. (Dunakeszi, Hungary). Urea and melamine were acquired from Elemental SRL (Bihor, Romania) and Sigma-Aldrich Corporation (United States) respectively. All three protein powders were artificially adulterated using urea (U), glycine (G), taurine (T) and

melamine (M) as adulterants. The nitrogen content (N) of the adulterants were: urea (46.62% N), glycine (18.65% N), taurine (11.19% N) and melamine (66.60% N). To have equal particle size, protein powders and adulterants were sieved through a wire mesh sieve with pore size (0.6 mm) before adulteration. A combination pattern was developed to contain single adulterant mixtures (U, G, T, M), dual mixtures (GT, UG, GM, UT, TM, UM) and multiple mixtures (UGT, GTM, UGM, UTM, UGTM). This resulted in a total 15 different mixture combinations. All the mixture combinations were prepared to have total adulterant concentrations of 0.5, 1, 1.5, 2, 2.5, and 3 % w/w in whey, beef and pea protein powders. The exact amount of protein powder used in each mixture was calculated based on the nitrogen content of the individual adulterants using melamine as the base adulterant because it had the highest nitrogen content of 66.6%. Triplicates of each mixture were prepared with each weighing 3 g (total mass after adulteration) and rigorously homogenized by shaking for 3 minutes. In total, there were 273 samples per protein type. Barcode system was used for easy labelling and identification during scanning with the instruments.

3.4. Applied methods

3.4.1. Near infrared spectroscopy (NIRS)

The MetriNIR (MetriNIR Research, Development and Service Co., Budapest, Hungary) with a wavelength range of 750-1700 nm and a spectral stepping of 2 nm was used as the benchtop spectrophotometer. The NIR-S-G1 (InnoSpectra Co., Hsinchu, Taiwan) with a wavelength range of 900-1700 nm and a spectral stepping of 3-4 nm was used as the handheld spectrophotometer.

For determination of Tokaji wine adulteration, three consecutive transmittance spectra were collected for each repeat sample using benchtop and handheld spectrophotometers with an optical glass cuvette. For determination of protein powder adulteration, three consecutive diffuse reflectance spectra of each sample repeat were collected using three setups: benchtop spectrophotometer with optical glass cuvette, handheld spectrophotometer with optical glass cuvette, and handheld spectrophotometer with low density polyethylene (LDPE) zip lock bags.

Raw spectra inspection was performed to obtain the best wavelength selection range for the dataset from the Tokaj wine and protein powder experiments. Peaks and wavelengths that showed some correlations with the composition of the different analytes were also identified and discussed. Outlier detection was performed in principal component analysis (PCA) before applying preprocessing techniques. Different types of pretreatment and their different combinations were tested on the raw spectra for their ability to reduce baseline shifts, spectral noise, correct additive and/or multiplicative effects in spectral data and enhance spectral information.

3.4.2. Electronic tongue (e-tongue)

The Alpha Astree potentiometric e-tongue with Ag/AgCl reference electrode and Chemical modified Field Effect Transistor (CHEMFET) sensors from AlphaM.O.S (Toulouse, France) equipped with food grade sensors was used to discriminate the different wine and meat mixtures based on pattern recognition. Conditioning and calibration were performed according to the manufacturer's recommendation (Alpha Astree, 2010). E-tongue was used for the determination of Tokaji wine adulteration and the determination of optimal dilution and optimal extraction. During all e-tongue experiments, the tested sample volume was 100 mL, the sampling time was 120 seconds, the sampling frequency was 1 second, and the cleaning time with distilled water was 20 seconds. The last 10 seconds of the sensor signals, representing stabilized and optimal sensitivity of the different sensors were exported for statistical evaluations in R-project. Sensor signals were all pretreated with the additive correction relative to all samples (Kovacs *et al.*, 2020). In addition to the additive correction relative to all samples, a sensor optimization process was performed for the dataset from the determination of optimal extraction.

3.4.3. LDA multi-class classifications for determination of wine, meat and protein powder adulteration

Linear discriminant analysis (LDA) models were developed to classify the different sample mixtures in the experiment for the determination of Tokaji wine adulteration, determination of optimal dilution, determination of optimal extraction and the determination protein powder adulteration. The reliability of all the developed LDA models was tested by splitting the data into two groups: the training set and validation set. The training set consisted of two-third of the data which included spectra (or sensor signals for datasets from the e-tongue) from the first and second replicate samples. The validation set consisted of spectra (or sensor signals for datasets from the e-tongue) from the third replicate samples. Cross-validation was done three times for each instrumental setup (three-fold cross-validation). The statistical parameters used to evaluate the performance of the LDA models were the recognition accuracy (%) and prediction accuracy (%). Recognition accuracy (%), represents the accuracy of calibration, whereas prediction accuracy (%), represents the accuracy of cross-validation (%).

3.4.4. PLS regression models for determination of wine, meat and protein powder adulteration

Partial least squares regression (PLSR) was used to predict sugar concentrations of authentic wines in the experiment for the determination of Tokaji wine adulteration. It was also used to predict turkey and beef concentrations in both the experiment for the determination of

optimal dilution and the determination of optimum dilution level. PLSR was also used to predict urea, glycine, taurine and melamine concentration in whey, beef and pea protein powder mixtures. It was also used to predict concentrations of pure protein powder in the mixtures. The predictive significance of all the PLS regression models described was tested with leave-one-sample out cross-validations: all three repeats of a sample and their consecutive spectra was each time, left of the validation process. The statistical parameters used to evaluate the performance of the PLS regression models were the root mean square error of calibration (RMSEC) and the coefficient of determination (R^2C); in cross-validation (RMSECV, R^2CV). The accuracies for the independent predictions were reported as determination coefficient of prediction (R^2pred) and root mean square error of prediction (RMSEP). For all the models, the optimum number of latent variables was determined based on the minimum, RMSEC, RMSECV and RMSEP value to prevent over fitting of the models.

3.4.5. Limit of detection, limit of quantification and limit of quantification for determination of protein powder adulteration

For the protein powder mixtures, limit of detection minimum value (LODmin), limit of detection maximum value (LODmax), limit of quantification minimum value (LOQmin), limit of quantification maximum value (LOQmax), explained variance X (the actual dataset) and explained variance Y (the predicted dataset) were calculated through the partial least-squares (PLS) methods according to the International Union of Pure and Applied Chemistry (IUPAC) approach described by (Allegrini and Olivieri, 2014):

$$LODmin = 3.3 [SEN - var(x) + h0min SEN - var(x) + h0min var(ycal)]^{1/2}$$

$$LODmax = 3.3 [SEN - var(x) + h0max SEN - var(x) + h0max var(ycal)]^{1/2}$$

Where, SEN is the sensitivity (inverse of the length of the regression coefficient), var(x) is the variance of the instrument signals. h0min/max is the minimum/maximum distance between a hyperplane for the calibration set, representing the scores of the samples for which the analyte of interest is absent and the center of a normalized calibration score space. Var(ycal) is the variance in the calibration concentrations. Lower and upper limits of the LODmin/max interval (LODmin and LODmax) correspond to the calibration samples with the lowest and largest extrapolated leverages to zero analyte concentration (Lukacs *et al.*, 2018). LOQmin/max interval was obtained by multiplying the LODmin/max values with a factor value of three (Allegrini and Olivieri, 2014). LODmin/max and LOQmin/max values were used to further evaluate the performance of the models for detecting urea, glycine, taurine and melamine in protein powders

using all three setups: benchtop spectrophotometer with optical glass, handheld spectrophotometer with optical glass and handheld spectrophotometer with LDPE plastic bag.

4. RESULTS AND DISCUSSION

4.1. Determination of Tokaj wine adulteration with NIRS

Based on the location of the absorption peaks of the tested components and results of PCA, the wavelength ranges of 950–1650 nm was selected for the spectra optimization using benchtop. The spectra of handheld spectrophotometer were observed to be characterized by clipping between the wavelength range 1400-1500 nm, so the wavelength range 950-1400 nm was selected to eliminate the clipping before spectra optimization. Spectra from both benchtop and handheld spectrophotometer showed baseline offsets that was suspected to be due to the large distance from the unit's light sources to the reflective/diffusive backing of the transmittance cell. Handheld spectrophotometer had the highest baseline offset. The observed baseline offsets suggested that they may require some form of spectra correction. Savitzky-Golay smoothing (21 points) followed by standard normal variate (SNV) showed the highest classification accuracy for both the benchtop and handheld Spectrophotometers so this was applied before developing detailed LDA and PLSR models for the determination of Tokaji wine adulteration.

There were 100% cross-validation accuracies for all the different authentic wines using the benchtop spectrophotometer. Using the handheld spectrophotometer, all the authentic wines could be classified with 100% correct accuracy after cross-validation except the wine with the highest grade (Aszu I). Tokaji Aszu II and Forditas II could be classified with 80.18% and 89% cross-validation accuracy. Benchtop spectrophotometer could predict the sugar concentration of the authentic wines with R^2CV of 0.92 and RMSECV of 16.80 g/L. The handheld spectrophotometer could not predict the sugar content of the authentic wines with cross-validation although calibration models were possible. When PLSR models were developed for the benchtop spectrophotometer using a wavelength range of 950-1400 nm, lower accuracies were achieved with R^2CV 0.45 and RMSECV 46.59 g/L. This suggests that the first overtone wavelength range of water (1300-1600 nm) maybe vital for tracking Tokaji wine adulteration with grape must concentrate as already reported in other liquids (Muncan, 2019) through the novel scientific discipline known as “aquaphotomics” (R. Tsenkova *et al.*, 2018).

Using the benchtop spectrophotometer, there was average recognition of 97.01% and average prediction of 96.78% for the classification of authentic and Forditas I wine that was adulterated with grape must concentrate and sucrose to mimic the sugar concentration of the authentic wines. Using the handheld spectrophotometer, there was average recognition of 72.40% and average prediction of 68.22% for the classification of authentic and Forditas I wine that was adulterated with grape must concentrate and sucrose to mimic the sugar concentration of the

authentic wines. With cross validation, Aszu I, Aszu II, Forditas I and Forditas II could be classified 70.18%, 77.93%, 89% and 66.67% accuracy respectively.

Using the benchtop spectrophotometer, there was average recognition of 98.76% and average prediction of 98.78% for the classification of authentic and Forditas II wine that was adulterated with grape must concentrate and sucrose to mimic the sugar concentration of the authentic wines. Using the handheld spectrophotometer, there was average recognition of 81.87% and average prediction of 76.06% for the classification of authentic and Forditas II wine that was adulterated with grape must concentrate and sucrose to mimic the sugar concentration of the authentic wines. With cross validation, Aszu I, Aszu II, Forditas I and Forditas II could be classified 80.18%, 89%, 100% and 44.48% accuracy respectively.

The results from this study signal a potential for using near infrared spectroscopy to track Tokaji wine adulteration.

4.2. Determination of Tokaj wine adulteration with e-tongue

There was 100% cross-validation accuracy for classifying all the different authentic wines using the e-tongue. There was average recognition accuracy of 99.54% and prediction accuracy of 98.17% when authentic wines and Forditas I adulterated wines were classified. With cross validation, all the authentic wines could be classified with 100% accuracy. There was average recognition of 100% and prediction of 93.17% when authentic wines and Forditas II adulterated wines were classified. There was 53.81% misclassification of authentic Forditas II wine as base sugarwine and 7.62% misclassification of base sugar wine as authentic Forditas II wine rate of 53.81% with cross-validation. Hirtherto this study, there was no report about using the e-tongue to detect adulteration of wines with grape must concentrate. The demonstrated classification capabilities of the e-tongue could be taken advantage of by the wine industries for reliable quality monitoring of botrytized wines. Additive correction relative to all samples improved the classification results of the e-tongue datasets and can be adopted for samples measurements performed with the same sample set and sequence. Sugar concentration of the wines could be predicted with R^2CV of 0.90 and $RMSECV$ of 17.67 g/L of wine using 7 latent variables.

4.3. Comparison of NIRS and e-tongue for determination of Tokaji wine adulteration

Comparatively, the e-tongue could classify Forditas I wines better than the benchtop spectrophotometer but the benchtop spectrophotometer could classify Forditas II wines better. This suggests that the benchtop spectrophotometer was more sensitive to higher sugar concentrations than the e-tongue as Forditas II adulterated wines contained more grape must concentrate because they were the lowest grade wines.

4.4. Determination of meat optimal dilution and optimal extraction for e-tongue analysis

There was average recognition and prediction accuracy of 100% respectively for the classification of adulterated meat samples using all three different dilutions. There was therefore a need to apply some other multivariate tools to ascertain the optimum dilution level. PLS regression was used for this. The different meat mixtures could be predicted with R^2CV in the range 0.65-0.95 and RMSECV generally, less than 2.14% w/v of turkey using all three dilutions. Dilution level 2 with 1% w/v was proven to be the optimum dilution level the highest R^2CV of 0.95 and the lowest RMSECV of 0.80% w/v among all the three tested dilution methods. It was used for subsequent experiments for the determination of optimal extraction method.

Using the raw meat extraction method to classify turkey/chicken mixtures there was average recognition accuracy of 81.28% and prediction accuracy of 58.35%. Using the raw meat extraction method to classify beef/pork mixtures, there was average recognition accuracy of 67.73% and prediction accuracy of 54.25%. Misclassification rates associated with the low concentrations of both turkey/chicken and beef/pork meat mixtures after raw meat extraction suggested that perhaps, the meat compounds extracted were not sufficient for detection and discrimination with the e-tongue. Extracting meat compounds with water may be a challenge especially with the presence of fat-soluble compounds, which is often processed with other methods such as the Soxhlet method, Bligh and Dyer method, Folch method, microwave solvent extraction etc (Hewavitharana *et al.*, 2020). This is particularly true as the samples with the highest concentrations: T080 and B080 always gave the best classification accuracies whereas, those with the lowest concentration T099 and B099 consistently gave the worst. Extraction of meat compounds often involve using denaturing or non-denaturing solutions, which can be expensive (Malva *et al.*, 2018). Better accuracies of 100% were however achieved with this extraction for determination of optimal dilution. This could be due to the differences in the range of the mixture concentrations.

Using the frozen meat extraction method to classify turkey/chicken mixtures, there was average recognition of 80.52% and prediction accuracy of 62.55%. Using the frozen meat extraction method to classify beef/pork mixtures, there was average recognition accuracy of 85.51% and prediction accuracy of 56.41%. Misclassifications observed with the frozen meat extraction method suggest that, the meat compounds extracted may also, not have been sufficient for detection and discrimination by the e-tongue sensors. The quality of frozen/thawed meat is affected by the amount of frozen and unfrozen water, freezing rate and the temperature and time of frozen storage (Daszkiewicz, Kubiak and Panfil, 2018). The characteristics of unfrozen water influences the rate and extent of physical, chemical, and biochemical processes in meat (Leygonie,

Britz and Hoffman, 2012) which, may have influenced drip and the concentrations of compounds necessary for the detection and discrimination with the e-tongue sensors.

Using the cooked meat extraction method to classify turkey/chicken mixtures, there was average recognition accuracy of 78.13% and prediction accuracy of 64.73%. Using the cooked meat extraction method to classify beef/pork mixtures, there was average recognition accuracy of 89.62% and prediction accuracy of 68.77%. Besides time consumption, elevated temperatures have been widely acknowledged to be effective in the extraction of bioactive compounds in diverse foods (Putnik *et al.*, 2018; Khan, Aslam and Makroo, 2019). Meat extraction by cooking was also reported to be effective in the extraction of compounds from chicken bone (Kumoro *et al.*, 2010) and also, for the detection beef adulteration with illegal hormonal substances (Goga, Ferraro and Barbera, 2011).

4.5. Comparison of three extraction methods for detecting turkey/chicken and beef/pork mixtures with e-tongue

Comparatively, all the three extraction methods yielded similar recognition accuracies but the cooked meat extraction yielded the best cross-validation accuracies for classifying turkey/chicken mixtures. Better accuracies were achieved for the red meat adulteration compared to the poultry adulteration. Additive correction relative to all samples also improved the classification results of all the datasets. The best PLS model for the prediction of chicken in turkey was achieved when the frozen meat extraction with distilled water method was used but was the meat extraction by cooking method for pork in beef. Generally, sensors HA, BB, ZZ, GA and JB provided the best accuracies using LDA simulations. It was sensors HA, BB, ZZ and GA for PLSR simulations.

4.6. Determination of protein powder adulteration with NIRS

Based on the location of the absorption peaks of the tested components and results of PCA, the wavelength range 950–1650 nm was selected and used for the spectra optimization. This was done for spectra from all three setups: benchtop spectrophotometer and optical glass, handheld spectrophotometer and optical glass and spectrophotometer and LDPE plastic. The chemical structures of the adulterants themselves could be related to some of these important absorption bands in the absorption plot to give a hint about adulterant presence, their mixture combinations or concentrations in the three protein powder mixtures.

The handheld spectrophotometer scanned through the optical glass had the highest base line shift in raw spectra analysis. The reason for this could be that, the optical glass surface could not fit properly to the window of the handheld spectrophotometer because of its structural design.

This may have resulted in a small air gap between the two surfaces that further influenced the optical path of the light that reaches the detector during analysis. Applying Savitzky-Golay smoothing (21 points) before MSC was the most effective pretreatment for predicting melamine in the protein powder mixtures using PLSR. This was deemed as the optimum pretreatment for PLSR analysis using benchtop spectrophotometer and optical glass. Applying the Savitzky-Golay smoothing (21 points) before SNV gave the highest classification accuracies for classifying all the different mixture combinations, pure whey, beef and pea protein powder. This was deemed as the optimum pretreatment for LDA analysis using benchtop spectrophotometer and optical glass. Applying Savitzky-Golay smoothing (21 points) before SNV was the most effective pretreatment for predicting melamine in the protein powder mixtures using PLSR, when the handheld spectrophotometer was used with both optical glass and LDPE.

For the classification of pure and adulterated protein powders with based on their mixture combinations, there was average recognition accuracy of 74.01% and prediction accuracy of 74.09% using the benchtop spectrophotometer. Using the handheld spectrophotometer with optical glass yielded average recognition accuracies of 58.99% and prediction of 56.46%. Using the handheld spectrophotometer with LDPE plastic yielded average recognition accuracies of 62.17% and prediction of 54.48%.

For the classification of pure and adulterated protein powder mixtures at the lowest adulterant concentration of 0.5% w/w, using handheld spectrophotometer and optical glass yielded average recognition of 65.13% and average prediction of 53.49%. Pure whey, beef and pea protein powders could be classified with 66.89%, 100%, 89% accuracies respectively. Using the handheld spectrometer with handheld spectrophotometer with LDPE plastic bag yielded average recognition 83.79% and average prediction 56.19% achieved using handheld spectrophotometer with LDPE plastic bag. Pure whey, beef and pea protein powders could be classified with 66.89%, 66.89%, 89% accuracies respectively. Using the benchtop spectrophotometer, there was 99.47% average recognition and 98.75% average prediction for classifying all the protein powders at the lowest adulterant concentration of 0.5% w/w using benchtop spectrophotometer. There was no misclassification between pure protein powders and all mixture combinations with cross-validation. Benchtop spectrophotometer gave the best accuracies for classifying urea, glycine, taurine, melamine and their different mixture combinations at the lowest concentration of 0.5% w/w in whey, beef and pea protein powder. Using the handheld spectrophotometer with LPDE plastic bag gave better average accuracies than using the handheld spectrophotometer with optical glass. These models were developed to assume situations where producers may use very low concentrations because practically, tracking adulteration involves determining whether

adulteration has occurred or not and not necessary the percentage of its existence. Thus, 0.5% w/w being the lowest adulterant concentration tested in this study, it's detection regardless of the protein type would be more practical and be of much significance.

For classification of urea, glycine, taurine and melamine in protein powder using only mixtures with single adulterants, using the handheld spectrophotometer with optical glass yielded average 91.72% recognition and 90.43% prediction for classification of single adulterants in whey protein powder. Using the handheld spectrophotometer with LDPE plastic bag yielded an average recognition accuracy of 93.35% and 90.03% prediction were achieved when the LDPE plastic bag was used. For beef protein powder mixtures, there was an average classification accuracy of 95.66% recognition and 94.80% prediction accuracy when handheld spectrophotometer was used with optical glass. Average recognition and prediction accuracy were 95.82% and 91.99% respectively when handheld spectrophotometer and LDPE plastic bag was used. With or without cross-validation, pure beef protein powders could be classified with 100% correct accuracy when they were scanned through either optical glass or LDPE plastic bag. For pea protein powder mixtures, average 94.65% recognition and 93.71% prediction accuracy were achieved when handheld spectrophotometer was used with optical glass. Using handheld spectrophotometer and LDPE plastic bag, accuracies of 93.55% (recognition) and 94.02% (prediction) were achieved. The best accuracies for classifying urea, glycine, taurine and melamine in protein powder mixtures with single adulterants were achieved with the benchtop spectrophotometer for all the three protein powders. There was average 98.71% recognition and 96.28% prediction for whey protein powder, 100% recognition and 99.62% prediction for beef protein powder and 98.89% recognition and 98.88% prediction for pea. with cross-validation, pure beef and pea protein powder could be predicted with 100% correct accuracy but 11% of samples containing melamine were misclassified as pure whey protein powder.

4.7. PLSR prediction of urea, glycine, taurine and melamine concentrations in protein powder mixtures

All the models could predict the adulterants with R^2CV in the range 0.74-0.93 and RMSECV in the range 0.21- 1.57% w/w of adulterated protein powders. The models developed with the dataset from scanning through the LDPE plastic bag was generally weaker compared to those developed with the dataset from scanning through optical glass with the handheld spectrophotometer. Urea could be predicted with the highest accuracies for all three setups: benchtop spectrophotometer with optical glass, handheld spectrophotometer with optical glass and handheld spectrophotometer with LDPE plastic. RMSECV was generally lowest for the prediction of melamine in the protein powder mixtures. Glycine yielded the highest RMSECV. The best

model for predicting urea, glycine, taurine and melamine in protein powders regardless of their adulterant concentration of mixture combination, was as R^2CV of 0.93 and $RMSECV$ of 0.21% w/w using the benchtop spectrophotometer.

4.8. PLSR prediction of urea, glycine, taurine and melamine concentrations in protein powder mixtures from analysis with independent data

All the models could predict the adulterants with R^2_{pred} in the range 0.72-0.94 and $RMSEP$ in the range 0.18- 1.59% w/w of adulterated protein powders. Urea could be predicted with the highest accuracies for all three cases: with benchtop spectrophotometer, handheld spectrophotometer with optical glass and handheld spectrophotometer with LDPE plastic bag, the best was as R^2_{pred} of 0.94 and $RMSECV$ of 0.18% w/w using the benchtop spectrophotometer.

4.9. PLSR prediction of protein powder concentrations in protein powder mixtures from analysis with independent data

For prediction of protein powder concentration in all mixtures, using the handheld spectrophotometer with optical glass gave R^2CV 0.84, $RMSECV$ 1.38% w/w, R^2_{pred} 0.84 and $RMSEP$ 1.38% w/w. Using the handheld spectrophotometer with optical LDPE plastic gave R^2CV 0.86, $RMSECV$ 1.39% w/w, R^2_{pred} 0.84 and $RMSEP$ 1.47% w/w. Based on the R^2CV and $RMSECV$ values using the LDPE plastic bag gave the best results for predicting protein powder in the samples when the handheld spectrophotometer was used, compared to using the handheld spectrophotometer with optical glass. They both however, had similar R^2_{pred} and $RMSEP$ values. Using the benchtop spectrophotometer gave the best model for predicting protein powder concentrations in all the samples with R^2CV 0.86, $RMSECV$ 1.36% w/w, R^2_{pred} 0.87 and $RMSEP$ 1.30% w/w. For prediction of protein powder concentrations in protein powder mixtures that contained single mixtures of urea, glycine, taurine and melamine at concentrations of 0.5% w/w – 3% w/w, using the handheld spectrophotometer with optical glass gave R^2CV 0.84, $RMSECV$ 1.18% w/w, R^2_{pred} 0.88 and $RMSEP$ 1.58% w/w. Using the handheld spectrophotometer with optical LDPE plastic gave R^2CV 0.91, $RMSECV$ 1.36% w/w, R^2_{pred} 0.92 and $RMSEP$ 1.33% w/w. Based on the R^2CV , $RMSECV$, R^2_{pred} and $RMSEP$ values, using the handheld spectrophotometer with LDPE plastic bag gave the best results compared to using the handheld spectrophotometer with optical glass for predicting protein powder concentrations in protein powder mixtures that contained single mixtures of urea, glycine, taurine and melamine at concentrations of 0.5– 3% w/w. Using the benchtop spectrophotometer gave the best model for predicting protein powder concentrations in protein powder mixtures that contained single mixtures of urea, glycine, taurine and melamine at concentrations of 0.5% – 3% w/w. There was R^2CV 0.90, $RMSECV$ 1.41% w/w, R^2_{pred} 0.93 and $RMSEP$ 1.22% w/w.

4.10. LODmin, LODmax, LOQmin and LOQmax for the determination of urea, glycine, taurine and melamine in protein powder

Handheld spectrophotometer with optical glass had LODmin in the range 0.18 -0.53% w/w. Only taurine had LODmin higher than the minimum adulterant concentration of 0.5% w/w used in this study. LODmax was in the range 0.61-3.10% w/w. Only taurine had LODmax higher than the maximum adulterant concentration of 3% w/w used in this study. Handheld spectrophotometer with LDPE plastic had LODmin in the range 0.16% -0.85% w/w. Only taurine and glycine had LODmin higher than the minimum adulterant concentration of 0.5% w/w used in this study. Benchtop spectrophotometer gave the lowest LODmin/max values for the determination of urea, glycine, taurine and melamine in the protein powder mixtures.

The findings from the determination of protein powder adulteration are particularly important because hitherto this studies, adulterant concentrations in literature ranged between 1-5% w/w and no scanning was done through plastic bag. In addition, classification of the complex adulterant mixture combinations reported in this study signals the potential of the spectrophotometers in detecting complex forms of protein powder adulteration. The findings in from this study provides grounds for detecting novel forms of adulteration in whey, beef and pea protein powder. Compared to traditional methodology such as the Dumas method where, a test run by takes from 5 to 10 min depending on sample weighing and combustion, a single scan with NIRS takes less than 1 min. In terms of expenses, the average cost of the Dumas method is about \$25 per sample, whereas the NIR test method can be <\$5.00 per sample) (Ingle *et al.*, 2016). The discriminatory and classification accuracies achieved with the benchtop and handheld spectrophotometers proves their potential for detecting urea, glycine, taurine and melamine concentrations as low as 0.5% w/w in protein powders and provides advantages from both time and cost perspective. The handheld spectrophotometer provides an extra advantage of expeditious onsite detection of adulterants.

5. CONCLUSION AND RECOMMENDATION

Authentic Tokaji Tokaji Forditas II, Forditas I Aszu II and Aszu I wines could be classified with 100% accuracy using the benchtop spectrophotometer at a wavelength range of 950-1650 nm. Handheld spectrophotometer could only classify authentic Tokaji Forditas I, Forditas II and Aszu II with 100% accuracy using the wavelength 950-1400 nm. The authentic wines were predicted using PLSR, with R^2CV of 0.92 and an RMSECV of 16.80 g/L of wine using the benchtop spectrophotometer at wavelength 950-1650 nm. Handheld spectrophotometer produced unsatisfactory results for predicting sugar concentrations of the wines. E-tongue could classify all authentic wines with 100% correct accuracy and predict them with R^2CV of 0.90 and an RMSECV of 17.67 g/L. Benchtop spectrophotometer could classify adulterated Forditas I and Forditas II wine mixtures with average cross-validation accuracies of 96.78% and 98.78% respectively. Handheld spectrophotometer could classify adulterated Forditas I and Forditas II wine mixtures with average cross-validation accuracies of 68.22% and 76.06% respectively. Adulterated Forditas I and Forditas II wine mixtures could be classified with average cross-validation accuracies of 98.17% and 93.10% using the e-tongue. The results in this study signal a potential for using electronic-tongue and near infrared spectroscopy to track Tokaji wine adulteration. For practically on industrial basis, further may be required with higher sample numbers for more robust models. The setup of the handheld spectrophotometer used in this study should also be carefully considered and adapted to better experimental procedures if it is to be used to track Tokaji wine adulteration.

Among the three tested dilution factors for the determination of optimum dilution for e-tongue analysis, 1% w/v dilution found to produce the best LDA results of 100% accuracy for classifying concentrations of turkey/chicken mixtures. With this dilution level, there was R^2CV of 0.95 and RMSECV of 0.80% w/v in PLSR. Using this dilution level for the three tested meat extraction methods, the cooked meat extraction method produced the best results for classifying turkey/chicken mixtures and beef/pork mixtures. There was average recognition of 78.13% and average prediction 64.72% for classifying turkey/chicken mixtures and average recognition of 89.62% and average prediction 68.77% for classifying beef/pork mixtures. Using this method, there was R^2CV of 0.72 and RMSECV of 3.83% w/v in PLSR. The determined optimal dilution and extraction method can be explored for rapid meat quality control checks with the electronic tongue; however, the study is recommended to be extended with alternative set of meat mixtures, wide range of mixture concentrations to ascertain the reliability of the methods for all meat types. This may also help understand why the cooked meat extraction method worked better for PLSR prediction of beef mixtures compared to the poultry mixtures.

Benchtop spectrophotometer gave the best accuracies for classifying single, dual, triple and quadruple mixtures of urea, glycine, taurine and melamine and that lowest concentration of 0.5% w/w. Using the handheld spectrophotometer with LPDE plastic bag gave better accuracies than using the handheld spectrophotometer with optical glass. Using the benchtop spectrophotometer gave the best model for predicting protein powder concentrations in all the samples with R^2CV 0.86, RMSECV 1.36% w/w, R^2pred 0.87 and RMSEP 1.30% w/w. Using the LDPE plastic bag gave better results for predicting protein powder in the samples compared to using the handheld spectrophotometer with optical glass. They both however, had similar R^2pred and RMSEP values. Using the benchtop spectrophotometer gave the best model for predicting protein powder concentrations in protein powder mixtures that contained only single adulterants of urea, glycine, taurine and melamine at concentrations of 0.5– 3% w/w. There was R^2CV 0.90, RMSECV 1.41% w/w, R^2pred 0.93 and RMSEP 1.22% w/w. Using the LDPE plastic bag gave the results compared to using the handheld spectrophotometer with optical glass for predicting protein powder concentrations in protein powder mixtures that contained single mixtures of urea, glycine, taurine and melamine at concentrations of 0.5– 3% w/w.

Benchtop spectrophotometer yielded the lowest limit of detections (LOD's) and limit of quantifications (LOQ's) for quantifying urea, glycine, taurine and melamine in whey, beef and beef protein powder compared to when the handheld spectrophotometer was used. Comparatively, using the handheld spectrophotometer and optical glass yielded lower LODmin/max and LOQmin/max for some of the adulterants than when the handheld spectrophotometer was used with optical glass was used but samples scanned through the LDPE plastic bag, had a better repeatability when average LOD and LOQ's were evaluated. Urea and melamine had the lowest LOD and LOQ irrespective of the instrument or scanning method. Taurine always had the highest LOD and LOQ irrespective of the instrument or scanning method. For future studies regarding protein powder adulteration, it is recommended to examine practically worthy levels of protein powder adulteration so that specific models may be developed for them with well-defined target accuracies. Assessing the optical of the handheld spectrometer and experimental setup of any spectrophotometer that will be used is very much recommended before starting experiments to obtain reliable experimental datasets.

6. NEW SCIENTIFIC RESULTS

For purposes of these new scientific findings, benchtop spectrophotometer refers to the MetriNIR (MetriNIR, Research Development and Service Co., Budapest, Hungary) whereas, handheld spectrophotometer refers to the NIR-S-G1 (InnoSpectra Co., Hsinchu, Taiwan). E-tongue refers to the Alpha Astree potentiometric electronic tongue (AlphaM.O.S, Toulouse, France) equipped with seven sensors developed for food application (BB, HA, ZZ, GA CA, JE, JB), a reference electrode and a 16 position auto sampler.

1. Authentic Tokaji Forditas II, Forditas I, Aszu II and Aszu I wines were scanned in transfectance mode with benchtop spectrophotometer (950-1650 nm) and handheld spectrophotometer (950-1400 nm) using a glass cuvette with layer thickness 0.4 mm. E-tongue was also used to analyze the wines after 50% v/v aqua dilution. Classification models with linear discriminant analysis (LDA) and prediction models with partial least squares regression (PLSR) were developed using data from all the instruments.
 - **Benchtop spectrophotometer** could classify all the authentic wines with 100% accuracy. Sugar content of authentic wines was predicted with R^2CV of 0.92 and RMSECV of 16.80 g/L of wine.
 - **Handheld spectrophotometer** could classify the authentic wines with average cross-validation accuracy of 94.48% with Forditas I (11.04%) and Aszu II (11.04%) being misclassified as Aszu I. Model developed to predict sugar content of authentic wines with the current setup of handheld spectrophotometer was unsatisfactory.
 - **E-tongue could** classify all authentic wines with 100% correct accuracy and predict the sugar content of authentic wines with R^2CV of 0.90 and RMSECV of 17.67 g/L of wine.
2. Authentic Tokaji wines (Forditas II, Forditas I, Aszu II, Aszu I), adulterated Forditas I and Forditas II wines (add must concentrate to mimic authentic wine sugar concentration) and wine with added sucrose before refermentation were analyzed with spectrophotometers and e-tongue (after 50% v/v aqua dilution). The samples were scanned in transfectance mode with benchtop spectrophotometer (950-1650 nm) and handheld spectrophotometer (950-1400 nm) using a glass cuvette with layer thickness 0.4 mm. Classification models with linear discriminant analysis (LDA) and prediction models with partial least squares regression (PLSR) were developed using data from all the instruments.
 - **Benchtop spectrophotometer** could classify the different Forditas I and Forditas II wine mixtures with average cross-validation accuracies of 96.78% and 98.78%, respectively. There was no misclassification between authentic and adulterated wines.

- **Handheld spectrophotometer** could classify adulterated Forditas I and Forditas II wine mixtures with average cross-validation accuracy of 68.22% and 76.06%, respectively. There were misclassifications between authentic and adulterated wines in both Forditas I and Forditas II wine mixtures.
 - **E-tongue** could classify adulterated Forditas I and Forditas II wine mixtures with average cross-validation accuracies of 98.17% and 93.10%. There was no misclassification between authentic and adulterated wines in Forditas I wine mixtures. The wine containing sucrose before refermentation was the only adulterated wine misclassified (53.81%) as an authentic Tokaji wine (Forditas II wine).
3. Raw meat extracts from 100%, 97%, 95%, 90% w/w turkey/chicken mixtures were obtained using aqua dilution levels of 0.5%, 1% and 2% w/v and analyzed using e-tongue. Classification models with linear discriminant analysis (LDA) and prediction models with partial least squares regression (PLSR) were developed using data from e-tongue.
- **E-tongue** could classify turkey/chicken mixtures after 0.5%, 1% and 2% w/v dilution and predict turkey concentration with R^2CV 0.65, 0.95, 0.81 and RMSECV 2.14% w/w, 0.80% w/w 1.57% w/w for the respective dilution levels. Dilution level (1% w/v) was the optimum among the three tested dilution levels for e-tongue analysis.
4. Raw meat extracts from 100%, 99%, 97%, 95%, 90% and 80% w/w poultry (turkey/chicken) and 100%, 99%, 97%, 95%, 90% and 80% w/w red meat (beef/pork) were obtained using raw meat/frozen meat/cooked extraction method and diluted to 1% w/v. Classification models with linear discriminant analysis (LDA) and prediction models with partial least squares regression (PLSR) were developed using data from e-tongue.
- **E-tongue** could classify **poultry mixtures** with cross-validation of 58.35%, 62.55%, 64.72% for raw meat, frozen meat, cooked meat extraction methods respectively, and predict them with R^2CV 0.76, 0.81, 0.47 and RMSECV 3.34% w/w, 2.89% w/w and 4.93% w/w respectively.
 - **E-tongue** could classify **red meat mixtures** with cross-validation of 54.25%, 56.41%, 68.77% for raw meat, frozen meat, cooked meat extraction methods respectively, and predict them with R^2CV 0.76, 0.81, 0.47 and RMSECV of 3.34% w/w, 2.89% w/w and 4.93% w/w, respectively.

Whey, beef and pea protein powders were adulterated with urea, glycine, taurine and melamine at a total of 0.5%, 1%, 1.5%, 2%, 2.5% and 3% w/w adulteration using either single, dual, triple or quadruple mixture combinations (16 mixtures). The mixtures were scanned in diffuse reflectance mode using three setups: benchtop spectrophotometer with optical glass, handheld spectrophotometer with optical glass and handheld spectrophotometer with low density polyethylene (LDPE) plastic bag. Wavelength range for all three setups was 950-1650 nm.

5. *When only protein powder mixtures containing single adulterants were analyzed using linear discriminant analysis and predictions with independent data was performed using partial least squares regression:*

- **All three setups** could classify pure beef protein powder with 100% cross-validation accuracy.
- **Benchtop spectrophotometer with optical glass** could classify pure whey protein powder with 89% cross-validation accuracy and 11% misclassification as protein powder samples containing melamine. Protein powder concentration in the mixtures could be predicted with R^2_{CV} 0.86, RMSECV 1.36% w/w, R^2_{pred} 0.87 and RMSEP 1.30% w/w respectively.
- **Handheld spectrophotometer with optical glass** could classify pure whey and pure pea protein powder with 89% cross-validation accuracy each and 11% misclassification each as protein powder samples containing melamine. Protein powder concentration in the mixtures could be predicted with R^2_{CV} 0.84, RMSECV 1.38% w/w, R^2_{pred} 0.84 and RMSEP 1.38% w/w respectively.
- **Handheld spectrophotometer with LDPE plastic bag** could classify pure whey and pure pea protein powder with 89% cross-validation accuracy each and 11% misclassification each as protein powder samples containing glycine. Protein powder concentration in the mixtures could be predicted with R^2_{CV} 0.86, RMSECV 1.39% w/w, R^2_{pred} 0.84 and RMSEP 1.47% w/w respectively.

6. *When only samples containing the lowest adulterant concentration of 0.5% w/w were analyzed using linear discriminant analysis:*

- **Benchtop spectrophotometer with optical glass** could classify all the different mixture combinations with average cross-validation accuracy of 98.75%. There was no misclassification between pure protein powders and all mixture combinations.
- **Handheld spectrophotometer with optical glass** could classify all the different mixture combinations with average cross-validation accuracy of 53.49%. Pure whey, beef and pea protein powders could be classified with 66.89%, 100%, 89% accuracies respectively.

There was 33.11% and 11% misclassification of pure whey and pea protein powder respectively as adulterated protein powder samples.

- **Handheld spectrophotometer with LDPE plastic bag** could classify all the different mixture combinations with average cross-validation accuracy of 56.19%. Pure whey, beef and pea protein powders could be classified with 66.89%, 66.89%, 89% accuracies respectively and misclassifications of 11.04%, 22.08% and 11% respectively as adulterated protein powder samples. Pure whey and pea protein powders showed misclassifications (11.04%) amongst themselves.

7. *When all mixtures were analyzed using partial least squares regression with leave-one-sample-out cross-validation:*

- **Benchtop spectrophotometer with optical glass** could predict urea, glycine, taurine and melamine concentrations with R^2CV 0.93, 0.87, 0.93, 0.93 and RMSECV 0.21, 0.97, 0.94, 0.16% w/w respectively.
- **Handheld spectrophotometer with optical glass** could predict urea, glycine, taurine and melamine concentrations with R^2CV 0.89, 0.79, 0.86, 0.82 and RMSECV 0.27, 0.91, 1.30, 0.23% w/w respectively.
- **Handheld spectrophotometer with LDPE plastic bag** could predict urea, glycine, taurine and melamine concentrations with R^2CV 0.91, 0.74, 0.79, 0.75 and RMSECV 0.25, 1.09, 1.57, 0.29% w/w respectively.
- **All three setups** could predict urea with the highest accuracy.

8. *When limit of detection was calculated for urea, glycine, taurine and melamine in all the mixtures:*

- **Benchtop spectrophotometer with optical glass** produced the lowest average limit of detections (LOD's) 0.18%, 0.50%, 0.76% and 0.13% for urea, glycine, taurine and melamine respectively. Urea and melamine had average LOD's below the minimum tested adulterant concentration of 0.5% w/w.
- **Handheld spectrophotometer with optical glass** produced average limit of detections (LOD's) of 0.52%, 1.35, 1.81% and 0.40% for urea, glycine, taurine and melamine respectively. Melamine had average LOD below the minimum tested adulterant concentration of 0.5% w/w.
- **Handheld spectrophotometer with LDPE plastic bag** produced average limit of detections (LOD's) of 0.27%, 1.02%, 1.42% and 0.24% for urea, glycine, taurine and melamine respectively. Urea and melamine had LOD's below the minimum tested adulterant concentration of 0.5% w/w.

- **All three setups** produced LOD's in urea, taurine, glycine and melamine that were below the maximum tested adulterant concentration of 3% w/w. Taurine always had the highest LOD.

LIST OF PUBLICATIONS IN THE FIELD OF STUDIES

1. **John Lewis Zinia Zaukuu**, Zoltan Gillay, Zoltan Kovacs. Standardized Extraction Techniques for Meat Analysis with the Electronic Tongue: A Case Study of Poultry and Red Meat Adulteration. Volume 21, issue 2, page 481. <https://doi.org/10.3390/s21020481>. **Sensors (2021). Q1, IF: 3.240**
2. **John-Lewis Zinia Zaukuu**, Balkis Aouadi, Mátyás Lukács, Zsanett Bodor, Flóra Vitális, Biborka Gillay, Zoltan Gillay, László Friedrich, Zoltan Kovacs. Detecting Low Concentrations of Nitrogen-Based Adulterants in Whey Protein Powder Using Benchtop and Handheld NIR Spectrophotometers and the Feasibility of Scanning through Plastic Bag. Volume 25, issue 11, pages 2522. <https://doi.org/10.3390/molecules25112522> . **Molecules (2020). Q2, IF: 3.267**
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5. **John-Lewis Zinia Zaukuu**, János Soós, Zsanett Bodor, József Felföldi, Ildikó Magyar, Zoltan Kovacs. Authentication of Tokaji Wine (Hungaricum) with the Electronic Tongue and Near Infrared Spectroscopy. Volume 84, issue 12, page 3437-3444. <https://doi.org/10.1111/1750-3841.14956>. **Journal of Food Science (2019). Q2, IF: 2.478**
6. **John Lewis Zinia Zaukuu**, George Bazar, Zoltan Gillay, Zoltan Kovacs. Emerging trends of advanced sensor-based instruments for meat, poultry and fish quality—a review. Volume 60, issue 12, Pages 3443-3460. <https://doi.org/10.1080/10408398.2019.1691972>. **Critical Reviews in Food Science and Nutrition (2019). Q1, IF: 7.862**