Scientific report

regarding the project implementation in the period January 2015 - December 2015

In the period January-December **2015** were carried out activities included in the project implementation plan, activities needed to achieve the overall objective of the project, an electronic system based on chemical sensors and biosensors for analysis of biogenic amines. Next will present the work performed, objectives achieved and disseminating activities carried out this year.

Testing of sensors and biosensors

1. Selection of sensors and biosensors arrays for applications on real samples

For the analysis of real samples were selected sensors and biosensors with appropriate properties for the aim of this research and those that are able to detect biogenic amines with adequate sensitivity.

The first category of sensors that were included in sensors array of electronic system are sensors based on polypyrrole doped with different anions. Doping anions and electrosynthesis conditions are presented in Table 1.

| Doping agent | Concentration $(mol \times L^{-1})$ | Electrochemical technique | Conditions |
|--------------|-------------------------------------|---------------------------|------------|
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Table 1. The conditions of electropolymerization used in the development of polypyrrole sensors

The second category of sensors included in the sensors array are *screen-printed electrodes* based on carbon nanomaterials, employed as received or modified in the laboratory with phthalocyanines. Screen-printed carbon electrodes (4 mm in diameter) purchased from Dropsens (www.dropsens.com, models 110 CNT, 110 GPH, 110 CNF) were used. These screen-printed electrodes are designed for the development of sensors and biosensors with an enhanced electrochemical active area. Carbon nanomaterials (Carbon Nanotubes-CNT, Carbon Nanofibres-CNF and Graphene-GPH) have excellent mechanical, electrical, and thermal properties thus become excellent modifier to the carbon matrix for enhancing the sensitivity and selectivity.

From the category of biosensors were included a variety of biosensors in the array; the most important ones are summarized below.

Carbon paste based biosensor for serotonin. Carbon paste electrodes were prepared by mixing carbon nanopowder (<50 nm particle size (TEM), \geq 99% trace metals basis, Sigma-Aldrich) and the cobalt(II) phthalocyanine (15%, w/w). Nujol was used as the binder of the multi-component composite mixture. Paste was packed into the body of a 1mL PVC (polyvinylchloride) syringe and compacted. A metallic copper wire was used as an electrical contact. The tyrosinase (Ty) was immobilized on the above CoPc-CPE (CPE modified with CoPc) by drop-and-dry technique followed by cross-linking with glutaraldehyde.

Glassy carbon electrode based biosensor for catecholamines. The glassy carbon electrode (GCE) surface was polished with alumina paste, washed with ultrapure water before and rinsed in methanol. The electrode active part was an 4 mm diameter disk. The other parts of carbon electrode were covered with isolating epoxy resin. After cleaning process, the GCE surface was coated with 10 μ L of the SWCNTS suspension (1.0 mg×mL⁻¹ in methanol). The solvent was evaporated in air at room temperature. The

enzyme, Ty, was immobilized on the above GCE modified with SWCNTs (SWCNT-GCE) by drop-and-dry technique followed by cross-linking.

Polypyrrole doped with anions used in the case of sensors were used for immobilization of DAO by crosslinking. Different types of **biosensors based on polypyrrole and aminooxidases** (monoamine oxidase A, monoamine oxidase B, and diamine oxidase, respectively) were also included in the array. They have different sensitivities and selectivities towards biogenic amines. For example the biosensor based on diamino oxidase (DAO) from Porcine kidney, E.C. 1.4.3.6), cross-linked by glutaraldehyde on electrosynthesized polypyrrole films is high sensitive for histamine detection.

A biosensor for the determination of dopamine and epinephrine by using **tyrosinase** (from mooshroom, E.C. 1.14.18.1), cross-linked by glutaraldehyde on **graphene modified screen-printed carbon electrodes** was developed and included in the array.

For the determination of catecholamines a biosensor based on *tyrosinase*, cross-linked by glutaraldehyde on *amide functionalized single-walled carbon nanotubes* modified screen-printed carbon electrodes was included in the array.

Ty-SWCNT-COOH/SPE biosensor for tyramine. 5.0 mg×mL⁻¹ of tyrosinase solution was prepared with 0.01 M phosphate buffer solution at pH 7.0. 50 μ L of 0.01 M phosphate buffer (pH 7.0) containing 5 mg×mL⁻¹ of Ty was drop coated onto 12.56 mm² area of SWCNT-COOH (*carboxyl functionalized single-walled carbon nanotubes*) thick film, and dried at 4°C for 10 min. Then the electrode was treated with glutaraldehyde vapor to immobilize Ty onto SWCNT-COOH/SPE surface resulting in a Ty-SWCNT-COOH/SPE biosensor. After that, the biosensor was fully washed with ultrapure water to remove all chemicals physically adsorbed.

A biosensor based on a *carbon screen-printed electrode modified with Prussian Blue and diamine oxidase* was included in the array.

The biosensor **based on Langmuir – Blodgett film of tyrosinase, arachidic acid and dysprosium bisphthalocyanine** for the electrochemical detection of tyramine and dopamine was taken into account for the array.

In conclusion, in this task were selected the sensors and biosensors optimal for detection and/or quantification of biogenic amines. The novel sensors and biosensors are based on different electrode designs, different materials for immobilization, electron mediators and enzymes. Multi(bio)sensor system developed is able to detect all categories of biogenic amines.

2. Selecting methods for data processing

The objective of this activity is to establish the methods of data analysis in order to discrimination and classification of samples analyzed and establishing correlations between different types of measurements.

For processing the data obtained with sensors and biosensors developed in this project they were optimized and used several methods of multivariate data analysis: principal component analysis (PCA), partial least squares - discriminant analysis (PLS-DA), analysis of variance (ANOVA), Soft independent modelling of class analogies (SIMCA), t test, multiple regression by partial least squares method for a parameter or several parameters (PLS1 and PLS2).

Sensors and biosensors provide complex voltammograms (variety of peaks at different potentials and with different currents). The intrinsic complexity and cross-selectivity of the signals generated by the array of sensors and biosensors are advantages because the data set contains large amount of information about the sample. But, the fact that the whole data set contains meaningful information can difficult the data processing. In consequence, a pre-treatment step to reduce of the number of variables (without loss of information) is required.

For the analysis of the data obtained with sensors or biosensors developed in this project it is required a preprocessing step. Cyclic voltammograms (CV) and the voltammograms obtained by square wave voltammetry (SWV) are differently analyzed.

Using this method, SWV curve is multiplied by a number of 10 kernel functions, and further integrated with respect to potential. Ten parameters for each SWV are obtained. An example of applying the kernel method to data obtained by square wave voltammetry is presented in Figure 1.

Figure 1. Applying of the kernel method for the voltamograms registered by SWV

Cyclic voltammograms were mathematically pre-processed and used as data source for multivariate data analysis. Using kernel method, the cyclic voltammogram curve (i vs. E) is divided in anodic and cathodic part. Then, the anodic curve is multiplied by a number of 10 smooth, bell-shaped windowing functions, and integrated with respect to potential. By this pre-processing technique the information throughout the global response is reduced to 10 representative parameters per each curve. An example of applying the kernel method to data obtained by cyclic voltammetry is presented in Figure 2.

Figure 2. Aplicarea metodei kernel pentru voltamogramele inregistrate prin CV

Once the voltammograms have been pre-processed and the number of variables reduced, such variables are used as the input for multivariate data analysis using for instance Principal Component Analysis or Partial Least Squares Discriminant Analysis as discrimination and classification methods. Other methods were also employed as described below.

In the following paragraphs are discussed some of the results obtained in the research work carried out and applying different methods of data analysis.

Array of sensors based on Ppy. Discrimination of biogenic amines

The responses obtained when using SPE modified with polypyrrole showed a high degree of complexity, since transient responses observed in the voltammograms are related to the electrode material and to the nature and concentration of the amine molecules present in the solutions (and to the interactions electrode-solution).

This makes possible to use the sensors in an array configuration. The pattern of responses generated by the array is a fingerprint of the sample studied. This pattern can be related with certain features or characteristics of the samples by means of chemometrics.

In order to evaluate the discrimination capabilities of the array of voltammetric sensors, Principal Component Analysis was conducted using the information obtained from the array formed by Ppy modified SPE sensors. Figure 3 shows the PCA results as a three-dimensional scores plot of principal components that allow obtaining well-defined and separated clusters.

Figure 3. PCA score plot of the cyclic voltammograms of the amine solutions with Ppy –based sensor

array

PCA has been validated by full cross validation method and an optimal number of 5 principal components have been used. The first three principal components explain the 97% of the information (PC1= 57%; PC2=24%; PC3=16%).

The separated clusters indicate that the five solutions could be clearly discriminated from each other. In addition, the positions of the clusters are related to the electrochemical properties of the tested solutions. It has to be noticed that the cluster corresponding to the ammonia, appears in the left side of the diagram, far apart from the rest of the amines. Aliphatic amines appear in the right side of the diagram. A clear discrimination between primary amine (CAD), secondary amine (DMA) and tertiary amine (TMA) is observed, also. The heterocyclic amine, HIS has a particular electrochemical behavior that permits to discriminate it from aliphatic amines and ammonia.

Fish freshness monitoring

Fish freshness has been monitored trough the global assessment of spoilage products (including biogenic amines) using a multisensor array based in Ppy. For this purpose, fishes were eviscerated, washed and stored at 4°C during 10 days in a closed box. Every day, muscle samples were prepared and measured with Ppy based sensors.

A characteristic pattern of the deterioration of fish stored in ice can be divided into four phases: i) fish is fresh and has a sweet, seaweed and delicate taste (highly fresh); ii) there is a loss of the characteristic odor and taste. The flesh becomes neutral but has no off-flavors (fresh); iii) there is sign of spoilage and a range of volatile, unpleasant-smelling substances are produced; iv) fish is spoiled and has a putrid odor.

Principal Component Analysis was used to analyze the degradation process measured with the array of sensors. Figure 4 shows the PCA score plot obtained using the electrochemical signals registered every day using Ppy-based sensors.

Figure 4. PCA score plot of fish freshness monitoring with Ppy –based sensor array

The PCA score plot of the three first principal components accounts for 79% of variance. Clearly discriminated clusters can be observed. The first cluster, that appears in the left side of the figure corresponds to samples analyzed days 1 and 2 and correspond to a highly fresh product. Samples analyzed in days 3 and 4 did not show any off odor and could be classified as fresh product. The clusters appear in the central part of the figure. Samples collected days 5 and 6 showed off odors (degraded product). The last clusters that appears on the right side of the figure corresponds to samples collected on days 7 to 10 (spoiled fish).

PLS-DA was used to classify the day of fish degradation from the sensor array response.

A supervised method, the Partial Least Squares-Discriminant Analysis (PLS-DA) was used to evaluate the classification capability of the system.

As presented in Table 2, the fully cross-validated PLS-DA model (using an optimal number of 6 latent variables), revealed a clear identification of the fish degradation phases. Table 2 collects the quantitative data derived from the PLS-DA regression model.

| Group | Correl | ation | Root mean square error | | | |
|-------|------------------------|--------|------------------------|------------|--|--|
| | coeffi | cients | | | | |
| | Calibration Validation | | Calibration | Validation | | |
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| Table 2. Results of the calibr | ation and validation of PLS-DA |
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As observed, both the calibration and the validation values involved a good-quality model performance are achieved (large correlations between sensors and categorized variables, and low root mean square errors of calibration and validation). These results indicate that this methodology is able to real time monitor the fish freshness during storage.

Sensors based on carbonaceous materials for detection of biogenic amines.

SPEs modified with three carbonaceous materials: CNT, GPH and CNF were used for the analysis of different biogenic amines in the solution. The PCA score plot is presented in the Figure 5.

Figure 5. PCA score plot of CV obtained with SPEs in amine solutions The sensors array is able to discriminate among solutions of biogenic amines.

Application in detection of histamine

Different types of fish samples were analyzed with the developed biosensor. The purpose of this diversity of samples was to evaluate if the biosensor was able to quantify the amount of histamine in different fish species with good reliability.

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Use of ANOVA in data analysis

Electrochemical data were tested for statistical significance using one-way ANOVA routines running under Excel. The factor was fish species. Values of p < 0.05 were considered statistically significant. In Table 4 are presented the ANOVA significance results. ANOVA was based on honestly significant difference test (Tukey test). This test is based on pairwise comparison among means as presented in equation:

en It of nd

 $M_i - M_j$ = difference between pair means

MSE = mean square error

n_h = the harmonized mean

As presented in Table 4 the population means are significantly different. The samples could be classified in groups in agreement with type of fishes. ANOVA showed significant differences among these groups based on the results obtained with biosensors.

| e 4. ANOVA significance rest | | | | | |
|------------------------------|--------------|--|--|--|--|
| Contrast | Significance | | | | |
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* p < 0.05 (significant).

** p < 0.01 (highly significant).

*** p < 0.0001 (extremely significant).

Basic statistics

Ty/PO4-Ppy/Pt biosensor was applied to the determination of tyramine in salted sauerkraut samples. Total tyramine content of the sample, expressed in tyramine equivalent units, was analyzed using the standard addition method.

Amperograms were recorded under the optimum conditions (applied potential –0.250 V, constant stirring of the sample, pH 7.0). A volume of 100 μ L of the extracted tyramine was placed into the electrochemical cell, containing 5 mL of phosphate buffer solution 0.01 M of pH 7.0. After that, successive additions of 100 μ L of a 10 μ M tyramine solution were carried out. Current vs. concentration regression parameters were evaluated using the XLstat software. The concentration of tyramine found was 15.56±0.42 μ M (n = 5, α = 0.05), with a RSD of 2.16%.

Therefore, concentration level found in sauerkraut sample was $264.52\pm7.14 \text{ mg kg}^{-1}$, value according to relative low content of biogenic amine in sauerkraut sample. To check the viability of this procedure in the determination of tyramine in salted sauerkraut samples, recovery studies were also performed. The total amount of tyramine in a spiked sample, 54.78 μ M, was determined by standard addition. Five replicates

were carried out. The concentration of tyramine found was $54.51 \pm 1.60 \mu$ M (n = 5, $_{2}$ = 0.05, RSD = 2.36%), with an average recovery of 99.32%.

Multivariate regresions

Changes in pH in the fish samples throughout the storage time are summarised in Table 5. Partial least squares (PLS1) regressions were performed to model the relationships between biosensor signals and the pH values of the samples. The initial pH of fish flesh was close to 7.49, which is a typical value in very fresh products. In the solution obtained by procedure beforementioned the pH is 6.81, which indicate that the KCl aqueous solution do not influence the pH of the fish flesh.

During the storage is registered a decrease in the pH as a consequence of the enzymatic and chemical processes occurred in the beginning the post-mortem stage. After 5 days the pH of the solution started to increase as consequence of the biogenic amine generation. An excellent correlation was obtained using a model with 5 latent variables and validated by full cross validation method. The regression coefficient was 0.973 in calibration and 0.961 in validation. Additionally, low values of RMSEC (0.042) and RMSEP (0.051) were accomplished.

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Table 5. pH evolution during fish storage at 4°C

Classification by SIMCA

Soft Independent Modelling of Class (SIMCA) has also been applied to classify the samples. This method calculates several PCA models for each class in a training set (full cross validated with an optimal number of principal components). In this case, the models were constructed using the healthy people (class 1) and seek people (class 2). Then, external unknown samples or replicates are compared to the class models and assigned to classes (seek or health) according to their likeness to the original training set. To display the results obtained with SIMCA, Coomans plots have been used, in which the unknown data is recognized or not by a model with a confidence interval of 95%.

As observed in the Coomans graphic presented in Figure 7 a 100% classification of clinical samples was achieved.

Figure 7. Coomans plot of clinical samples classification

Two type of database were constructed: one for quantification of specific analyte, and the second for classification of the samples in function of their characteristics.

Good correlations have been found between the signals obtained from the expert sensory system and the biogenic amine content by means of PLS regression models. Using SIMCA or PLS-DA models, correlations between (bio)sensor responses and heart diseases were established.

In conclusion, a great varieties of methods were developed and applied for data analysis with good results facilitating the establishment of data significance.

3. Analysis of biogenic amines in meat products, cheese and fermented beverages

For this purpose we have developed a multibiosensor system coupled with a multivariate data analysis for detection and/or quantification of biogenic amines. Analyses of samples were carried out by means of amperometric and/or voltammetry techniques.

Analysis of biogenic amines in meat products.

This work describes the development of an bioelectronic tongue for the quantification of biogenic amines in meat products. The second application is the monitoring of meat products quality in time, in accelerate degradation conditions. The bioelectronic tongue includes an array of four biosensors developed and optimized in the laboratory. The biosensors are based on carbon screen-printed electrodes modified with single-wall carbon nanotubes and enzymes. The enzymes were tyrosinase, diamine oxidase, peroxidase and monoamine oxidase. The measurements with biosensors were carried out by amperometry and cyclic voltammetry. Cyclic voltammograms show redox processes related to the electrochemical activity of the compounds from samples or formed in the reactions catalyzed by enzymes (i.e. biogenic amines, hydrogen peroxyde, hydroquinone derivatives). Data analysis were carried out by means of Principal Component Analysis, Partial Least Squares Discriminate Analysis, Partial Least Squares regression, and Analysis of Variance.

For instance, in Table 6 are presented the performance characterists of a biosensor used in the quantification of biogenic amines in meat products.

| | DAO/CNT-SPE based biosensor | | | | | | |
|----------|--|--|--|--|--|--|--|
| Compound | LOD / μM I _{max} / μA K _M / μM | | | | | | |
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Table 6. Quantification of biogenic amines in meat products

It was found that bioelectronic tongue system is able to quantify the biogenic amine content in meat products (Italian ham, Sibiu salami, Banatean salami). Images of samples used in these studies are presented in the Figure 8.

Figure 8. Meat products under study

The values are in the range 380-450 mg×kg⁻¹, values below the maximum limit permitted in this type of food products. In the accelerate conditions of degradation, the increasing of biogenic amine amount is easy to follow by using the models based on bioelectronic tongue measurements.

Analysis of biogenic amines in beef meat extracts

In another study PPy based sensors were used for detection and quantification of biogenic amines in beef meat extracts. The performances of sensors towards amonia and putrescine are presented in the Table 7.

| Sensor | Compound LOD/ mM Recovery | | | | | | |
|---------|---------------------------|--|--|--|--|--|--|
| Ppy/FCN | | | | | | | |
| | | | | | | | |

Table 7. Performance characteristics of PPy based sensors

| Ppy/NP | | |
|------------------------------------|--|--|
| | | |
| Ppy/PWA | | |
| | | |
| Ppy/H ₂ SO ₄ | | |
| | | |
| Рру/Мо | | |
| | | |
| Ppy/AQS | | |
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Ppy/FCN sensor had the lowest limits of detection. The recovery is near to 100% for all sensors highlighting the viability of sensors to detect putrescine and ammonia in beef extract solution.

Analysis of biogenic amines in pickled and smoked fish samples

The tyramine level in different pickled and smoked fish samples was in the range of 16.7-61.8 mg×kg⁻¹ (Table 8). These levels of contamination are below the acceptable level of tyramine in foods.

Table 8. Quantification of biogenic amines in fish samples

| Food sample | Tyramine (mg×kg ⁻¹) 5 replicates | | | Average (mg×kg ⁻¹) | RSD (%) | 95% Confidence interval for mean | |
|-------------|---|--|--|-----------------------------------|------------|--|--|
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In other study the histamine content in carp fish samples was quantified during 5 days. In Table 9 are presented the results obtained.

Table 9. Histamine content in carp fish samples

| Number of days stored at 4°C | Res | Results obtained with biosensor (mg/kg) | | | | | RSD (%) |
|---------------------------------|-----|---|--|--|--|--|---------|
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The histamine content increasing during storage in the fridge at 4 Celsius degree. The histamine content after 4 days can have negative influences in the human health.

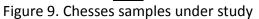
Analysis of biogenic amines in cheeses.

This paper describes the designing and optimization of an electrochemical biosensor for the determination of biogenic amines in blue cheese samples. The developed biosensor is based on a carbon screen-printed electrode, modified with Prussian Blue, which detect hydrogen peroxide produced by the reaction catalyzed by the diamine oxidase immobilized onto the surface of the electrode. Therefore, the biosensor detection mechanism is based on the electrochemical reduction of hydrogen peroxide.

The experimental conditions that influence the biosensitive properties of biosensors were optimized. In the optimal conditions of pH and potential applied, the biosensor performance characteristics were quantified. The measurements with the biosensor were carried out by amperometry, the current flowing through biosensor at -0.06 V was measured as a function of concentration of H₂O₂ in 100 mM phosphate buffer (pH 7.4).

The biosensor shows a low detection limit (0.045 μ M), and a linear range from 2x10⁻⁶ to 4x10⁻⁵ M. The biosensor fabrication is reproducible, relative standard deviation being 3.2%. Furthermore, the biosensor has good repeatability, and high affinity to biogenic amines typically found in blue cheeses.

Images of samples used in these studies are presented in the Figure 9.



Quantification of biogenic amines in cheese samples (Gorgonzola, Brie, Danish blue, and *Brânză de burduf*) was validated by standard addition method.

The results obtained are summarized in the table 10.

| Table 10. Results of biogenic amine | e quantification in cheese samples |
|-------------------------------------|------------------------------------|
|-------------------------------------|------------------------------------|

| Sample | Amount (mg× kg ⁻¹) | | | |
|--------|--------------------------------|--|--|--|
| | | | | |
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Analytical recovery of added BAs in the cheese samples are presented in the table 11.

| BAs (mg) added | BAs (mg) | Recovery (%) | |
|----------------|----------|--------------|--|
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Table 11. Analytical recovery of biogenic amines in cheese samples

The efficiency of biosensor for determinate BAs in cheese samples has been demonstrated.

Analysis of biogenic amines in fermented beverages.

Wine and beer have been reported as a cause for headaches with patients susceptible to migraines. Histamine in alcoholic drinks could cause of allergic and allergic like adverse responses. Wine has been a more common source than beer. Principal biogenic amines that could be found in wines were detected and quantified in red wines and beers using biosensors developed in this project.

In the case of wines several red wine samples were analyzed. Samples were analyzed with the biosensors array by triplicate. Results obtained are included in Table 12.

| Biogenic amines (mg/L) | Merlot | Cabernet | Pinot Noir | Syrah | Feteasca Neagra | Nero D'Avola |
|------------------------|--------|----------|------------|-------|-----------------|--------------|
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Beer samples were analysed by means of biosensors array and the results obtained are included in Table 13. RSD of the biogenic amines detection was 3.4%.

| Biogenic amines (mg/L) | Noroc | Bergenbier | Timisoreana | Bucegi | Ciucas |
|------------------------|-------|------------|-------------|--------|--------|
| | | | | | |
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Other fermented food analyzed was the sauerkraut samples. Recovery (%), RSD and 95% confidence interval for mean for extracted tyramine from salted sauerkraut samples are presented in table 14.

Table 14. Biogenic amines in sauerkraut samples

The efficiency of biosensor for determinate tyra6mine in sauerkraut samples has been demonstrated.

Biosensors developed in this project have shown excellent performance characteristics and were successful employed for detection of biogenic amines in a great variety of samples.

In this year were carried out all the activities from the work plan and the results obtained were in agreement with the objectives of the research. The objectives of this year were completely accomplished.

Dissemination of results

Dissemination of research results was done by publishing 2 ISI papers, publication of 2 chapters in a monograph published in a prestigious publishing house, participating with 5 papers at international or national conferences, an article in the Bulletin of the Chemical Society of Romania and habilitation thesis in the field of chemistry.

Publication of ISI papers

1. I. M. Apetrei, C. Diaconu, C. Apetrei, C. Georgescu, Electrochemical biosensor based on carbon nanofibers and diamine oxidase for detection of norepinephrine, Romanian Biotechnological Letters 21(1) (2016), *it will be published in nr. 21, issue 1, 2016.*

2. I.M. Apetrei, C. Apetrei, Biosensing Application of Hybrid Thin Film Layers Based Biosensors, IEEE Sensors Journal 15 (2015), 6926 - 6932, http://dx.doi.org/10.1109/JSEN.2015.2473796

Publication of other papers

Constantin Apetrei. *Senzori voltametrici pe bază de polimeri organici electroconductori,* Buletinul Societatii de Chimie din Romania, nr. 1, 2015.

Habilitation thesis

Constantin Apetrei. *Development of novel sensors and biosensors with applications in food analysis,* Dunarea de Jos University of Galati, 2015.

Elaborating of chapters in international monograph

1. C. Apetrei, I. M. Apetrei, Chemical composition of corn oil, In Corn and Coconut Oil: Antioxidant Properties, Uses and Health Benefits, Editor C. Apetrei, ISBN: 978-1-63483-420-9, Nova Publishers, 2015, pp. 1-28.

https://www.novapublishers.com/catalog/product_info.php?products_id=55691&osCsid=a58c860a7ae1f4 d5f714272f3d819203

2. I. M. Apetrei, C. Apetrei, Quality analyses and authentication of coconut oil, In Corn and Coconut Oil: Antioxidant Properties, Uses and Health Benefits, Editor C. Apetrei, ISBN: 978-1-63483-420-9, Nova Publishers, 2015, pp. 131-158.

https://www.novapublishers.com/catalog/product_info.php?products_id=55691&osCsid=a58c860a7ae1f4 d5f714272f3d819203

Participating to international and national conferences and abstracts published in conferences books of abstracts

1. C. Apetrei, I. M. Apetrei, Development of voltammetric sensors based on screen-printing technology for detection of creatinine, **Euronanoforum 2015**, Riga, Latvia June 10-12, 2015, poster, http://euronanoforum2015.eu/wp-content/uploads/2015/03/Abstract_Apetrei.pdf

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> Project manager, Prof.dr. Constantin APETREI