Attachment No. 4

Summary of Professional Accomplishments

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Summary of Professional Accomplishments

1. Name

Paweł Niedziałkowski

2. Diplomas, degrees conferred in specific areas of science or arts, including the name of the institution which conferred the degree, year of degree conferment, title of the PhD dissertation.

- Ph.D., (Doctor of Philosophy) 29/06/2010
  Title: Synthesis of amino acid and crown ethers derivatives containing redox-active and chromophore centers.
  Supervisor: Prof. Tadeusz Ossowski, D.Sc.

- M.Sc., (Master of Science) 31/05/2004
  Title: Synthesis of amino acid derivatives containing coordination and redox-active centers
  Supervisor: Prof. Tadeusz Ossowski, D.Sc.

3. Information on employment in research institutes or faculties/departments or school of arts.

- **Assistant professor** (from 01/10/2010 - currently) - University of Gdańsk, Faculty of Chemistry, Department of Analytical Chemistry, until 31/08/2012, from 01/09/2012 to 21/01/2014 Department of Organic Chemistry, and from 22/01/2014 - currently, Department of Analytical Chemistry,
- **Research Assistant** (from 06/04/2009 to 30/09/2010) - University of Gdańsk, Faculty of Chemistry, Department of Analytical Chemistry

4. Description of the achievements, set out in art. 219 para 1 point 2 of the Act.

Title of Scientific Achievement:

Modification and surface studies of electrode materials for analytical and bioanalytical purposes

The described scientific achievement is a series of 12 publications solving significant issues related to the research of the properties of electrode materials and development of techniques of their modification to obtain new components exhibiting different parameters in relation to the initial electrodes.

The main goal, apart from investigating the properties of unmodified electrode materials, was to obtain new highly sensitive materials as a basis for the detection of specific analytes at very low concentration levels. The papers described in scientific achievement are presented below.

1) H.1. R. Bogdanowicz✉, M. Sawczak, **P. Niedziałkowski**, P. Zięba, B. Finke, J. Ryl, J. Karczewski, T. Ossowski, Novel Functionalization of Boron-Doped Diamond by Microwave Pulsed-Plasma Polymerized Allylamine Film, *Journal of Physical Chemistry C*. 118 (2014) 8014–8025. [1]IF$_{2014}$ **4,772**, [2]MNiSW$_{2021}$ **140**.

2) H.2. R. Bogdanowicz✉, M. Sawczak, **P. Niedziałkowski**, P. Zięba, B. Finke, J. Ryl, T. Ossowski, Direct amination of boron-doped diamond by plasma polymerized allylamine film, *Physica Status Solidi (a)*. 211 (2014) 2319–2327. IF$_{2014}$ **1,616**, MNiSW$_{2021}$ **70**.

3) H.3. **P. Niedziałkowski**, T. Ossowski✉, P. Zięba, A. Cirocka, P. Rochowski, S. J. Pogorzelski, J. Ryl, M. Sobaszek, R. Bogdanowicz✉, Poly-l-lysine-modified boron-doped diamond electrodes for the amperometric detection of nucleic acid bases, *Journal of Electroanalytical Chemistry*. 756 (2015) 84–93. IF$_{2015}$ **2,822**, MNiSW$_{2021}$ **70**.

4) H.4. **P. Niedziałkowski**, R. Bogdanowicz✉, P. Zięba, J. Wysocka, J. Ryl, M. Sobaszek, T. Ossowski, Melamine-modified Boron-doped Diamond towards Enhanced Detection of Adenine, Guanine and Caffeine, *Electroanalysis*. 28 (2016) 211–221. IF$_{2016}$ **2,851**, MNiSW$_{2021}$ **70**.

5) H.5. K. Siuzdak, M. Ficek, M. Sobaszek, J. Ryl, M. Gnyba, **P. Niedziałkowski**, N. Malinowska, J. Karczewski, R. Bogdanowicz✉, Boron-Enhanced Growth of Micron-Scale Carbon-Based Nanowalls: A Route toward High Rates of Electrochemical Biosensing, *ACS Applied Materials & Interfaces*. 9 (2017) 12982–12992. IF$_{2017}$ **8,097**, MNiSW$_{2021}$ **200**.

6) H.6. D. Nidzworski, K. Siuzdak, **P. Niedziałkowski**, R. Bogdanowicz✉, M. Sobaszek, J. Ryl, P. Weiher, M. Sawczak, E. Wnuk, W. A. Goddard, A. Jaramillo-Botero✉, T. Ossowski, A rapid-response ultrasensitive biosensor for influenza virus detection using antibody modified boron-doped diamond, *Scientific Reports*. 7 (2017) 15707. IF$_{2017}$ **4,122**, MNiSW$_{2021}$ **140**.

7) H.7. K. Siuzdak*, **P. Niedziałkowski***, M. Sobaszek, T. Łęga, M. Sawczak, E. Czaczyk, K. Dziąbowska, T. Ossowski, D. Nidzworski*, R. Bogdanowicz✉, Biomolecular influenza virus detection based on the electrochemical impedance spectroscopy using the nanocrystalline boron-doped diamond electrodes with covalently bound antibodies, *Sensors and Actuators B: Chemical*. 280 (2019) 263–271. IF$_{2019}$ **7,100**, MNiSW$_{2021}$ **140**.

8) H.8. **P. Niedziałkowski**✉, Z. Cebula, N. Malinowska, W. Białobrzeska, M. Sobaszek, M. Ficek, R. Bogdanowicz, J. S. Anand, T. Ossowski, Comparison of the paracetamol electrochemical determination using boron-doped diamond electrode and boron-doped carbon nanowalls, *Biosensors and Bioelectronics*. 126 (2019) 308–314. IF$_{2019}$ **10,257**, MNiSW$_{2021}$ **200**.

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[1]IF - is given based on the Journal Citation Reports (JCR) database of Clarivate Analytics according to the year of publication. In the case of the paper [H.12.], IF values were given from the year 2020

[2]The points awarded by the MNiSW comes from the MEiN list from 1 December, 2021 published at www.wykazy.net.pl according to the lists published at www.gov.pl/web/edukacja-i-nauka

*The authors equally contributed to the paper [H.7.]

9) H.9. R. Bogdanowicz, **P. Niedziałkowski**, M. Sobaszek, D. Burnat, W. Białobrzeska, Z. Cebula, P. Sezemsky, M. Koba, V. Stranak, T. Ossowski, M. Śmietana✉, Optical Detection of Ketoprofen by Its Electropolymerization on an Indium Tin Oxide-Coated Optical Fiber Probe, *Sensors*. 18 (2018) 1361. IF_{2018} **3,031**, $MNiSW_{2021}$ **100**.

10) H.10. M. Janik✉, **P. Niedziałkowski**✉, K. Lechowicz, M. Koba, P. Sezemsky, V. Stranak, T. Ossowski, M. Śmietana, Electrochemically directed biofunctionalization of a lossy-mode resonance optical fiber sensor, *Optics Express*. 28 (2020) 15934–15942. IF_{2020} **3,894**, $MNiSW_{2021}$ **140**.

11) H.11. **P. Niedziałkowski**, P. Ślepski, J. Wysocka, J. Chamier-Ciemińska, L. Burczyk, M. Sobaszek, A. Wcisło, T. Ossowski, R. Bogdanowicz, J. Ryl✉, Multisine impedimetric probing of biocatalytic reactions for label-free detection of DEFB1 gene: How to verify that your dog is not human?, *Sensors and Actuators B: Chemical*. 323 (2020) 128664. IF_{2020} **7,460**, $MNiSW_{2021}$ **140**.

12) H.12. **P. Niedziałkowski**✉, M. Bojko, J. Ryl, A. Wcisło, M. Spodzieja, K. Magiera-Mularz, K. Guzik, G. Dubin, T. A. Holak, T. Ossowski, S. Rodziewicz-Motowidło, Ultrasensitive electrochemical determination of the cancer biomarker protein sPD-L1 based on a BMS-8-modified gold electrode, *Bioelectrochemistry*. 139 (2021) 107742. IF_{2020} **5,373**, $MNiSW_{2021}$ **100**.

4.1 Introduction

The current trend observed in chemical analytics is the increasing use of electroanalytical methods for the detection of chemicals. The advantage of using the above methods over traditional measurement techniques is their simplicity, relatively low cost, and reduced analysis time. The use of new electrode materials or chemically modified conductive materials allows the detection of analyzed compounds at very low concentration levels. The chemical modification of the electrodes allows the detection of biological compounds, which identification has been impossible so far.

At the moment, it is extremely challenging to research new electrode materials that permit simultaneous monitoring of analyte presence using two or more measurement techniques. Obtaining conductive materials that allow simultaneous measurements using electroanalytical and spectroscopic methods offers the opportunity not only to detect substances at low concentrations but also to use such a material as a potential opto-electrochemical sensor. This assumption can be achieved if a modified optical fiber is used as an electrode material.

The main research objective of the presented scientific achievement was the development of procedures for the modification of electrode materials in order to obtain new ones capable of detecting the investigated analytes and to study the physicochemical properties of the modified surface.

In my research, I have focused on developing techniques for the modification of boron-doped diamond (BDD) electrodes which, depending on the degree of boron doping and the content of the non-diamond carbon phase, exhibit semiconducting properties or metallic conductivity.

The (BDD) electrodes are excellent materials, characterized by a wide operating range (potential window) and high stability in aqueous solutions [1]. These types of electrodes are ideal for electroanalytical measurements due to their very low background current. At the same time, the modification of their surface allows the detection of analyzed compounds at unattainable detection levels.

The modification of the electrode surface (BDD) with an organic film formed on the surface significantly increases its sensitivity compared to the initial electrode, as I have shown in [H.3., H.4., H.6., H.7., H.11.]. In addition, I also decided to use diamond electrodes (BDDs) and (B:CNWs) as materials for the detection of selected analytes, without prior modification [H.8., H.5.]. I have also focused on developing and optimising the electrode surface modification (BDD) process to obtain a new electrode material exhibiting different electrochemical and optical properties to the initial electrode [H.1., H.2.]. In order to carry out simultaneous detection by optical and electrochemical techniques, I have also modified the surface of optical fiber covered with indium tin oxide (ITO) layer [H.9., H.10.]. This work, in addition to other publications, is the result of a consortium grant "Conductive photonic structures for multi-parametric biochemical diagnostics" Sonata Bis 4, in cooperation with Professor at the Warsaw University of Technology, Mateusz Śmietana, D.Sc., who was the principal investigator, while I was the principal investigator on the side of the University of Gdańsk.

In addition to the above-presented types of substrates that I used for modification, I also developed a method to functionalize the surface of a gold electrode, proving the ability to functionalize many electrode materials. In collaboration with scientists from the University of Gdansk and Jagiellonian University, who are conducting research on the interactions of proteins and 1-[[3-bromo-4-[(2-methyl[1,10-biphenyl]-3-yl)methoxy]phenyl]methyl]-2-piperidine-carboxylic acid (BMS-8). I developed an efficient method for its attachment to the gold electrode surface, obtaining an ultrasensitive surface allowing detection of the programmed cell death protein 1 (PD-L1) ligand [2], as described in [H.12.]. Based on a detailed analysis of the impedance spectra presented in this paper, the previously unattainable ability to distinguish between the above-mentioned ligand (PD-L1) and programmed death protein 1 (PD-L1) was demonstrated.

In summary, the main objective of my post-doctoral research was to develop procedures for the surface modification of electrodes consisting various materials and to investigate the reaction mechanisms of unmodified electrodes in order to use them for sensory measurements. An additional goal was to develop measurement procedures using electrochemical tools that allow simultaneous use of measurements with optical techniques for the detection of selected analytes. During the realization of the research objective, due to the interdisciplinary character of the conducted investigations, it was necessary to cooperate with highly specialized research groups from several scientific centers. In the discussed papers, I have taken a leading role in the planning and implementation of the surface functionalization of the investigated materials, as well as the verification of their effectiveness and the identification of detection mechanisms.

4.2 Properties and modification of boron doped diamond (BDD) electrodes

Due to their unique properties, the (BDD) electrodes are used to analyze metal ions, organic compounds and biomolecules. These electrodes have a wide potential window both in aqueous and non-aqueous solutions and are insensitive to the presence of dissolved oxygen in the electrolyte. Particularly, the (BDD) electrodes are used in sensory analyses due to their very low and stable background current, about 10 times lower than in the case of glassy carbon (GC) electrodes, and very high resistance to deactivation associated with the accumulation of the products of electroanalysis [3,4]. Boron is most commonly used for doping diamond electrodes, due to the fact that it has a low activation energy of 0.37 eV [5]. The doping of boron leads to a p-type semiconductor, and the conductivity of the electrodes depends on the amount of boron that has been used. Highly doped electrodes can exhibit semimetallic conductivity [6]. On the other hand, the n-type semiconductors, are obtained by doping with phosphorus, nitrogen or sulphur [7].

The modification of the electrodes (BDD) is determined by the outer surface of the electrode, which can be hydrogenated or oxidized. The hydrogenated surfaces are obtained during the chemical vapour deposition (CVD) process [8], while oxidized surfaces can be obtained by anodic polarization, by oxygen plasma, ozonation or in the presence of a strong oxidant [9].

Hydrogenated electrodes (BDD) allow the detection of substances which are difficult to detect on other electrodes, such as serotonin [10], glucose [11], L-Cysteine [12] or DNA [13]. In contrast, oxidized (BDD) electrodes have found application in the oxidation of substances containing a positive charge in their structure. The oxidation of dopamine [14], uric acid [15] and glutathione [16] are examples of the use of oxidized (BDD) electrodes. The oxidized electrodes are also used for the determination of oxygen, pH [17] and salinity of seawater [18].

The one of the ways to modify the (BDD) electrodes are to place thin oxide layers on the electrode surface, for instance titanium oxide (TiO_2) [19], zinc oxide (ZnO) [20] or cobalt oxide (CoO) [21]. The electrode surface can also be doped with metal nanoparticles: gold [22], silver [23], platinum [24] or metal oxides: ruthenium (RuO_2) [25] and iridium (IrO_2) [26]. There are also photochemical methods for the modification of (BDD) electrodes using suitable alkenes terminated with amine [27], carboxyl [28], hydroxyl [29] and thiol [30] groups. The above technique can also be used to anchor perfluorinated alkyl chains [31], ferrocene derivatives [32], or derivatives containing phosphate groups [33] on the electrode surface.

The oxidized (BDD) electrodes are most often functionalized by silanization reactions with suitable substrates allowing the formation of new functional groups such as alkyl, hydroxyl, amine or carboxyl groups on the surface [34].

4.3 Properties and modification of oxide (ITO) and gold (Au) electrodes.

The transparent electrode materials are used for electronic devices such as touch screens and in solar cells or light-emitting diodes [35]. Indium doped tin oxide (ITO), which is an n-type semiconductor, is one of the most widely used transparent materials in the visible range [36]. The electrodes containing a thin film (ITO) on their surface are mainly obtained by chemical vapor deposition from the gas phase (CVD) [37]. The above layers can also be obtained by magnetron sputtering [38] or physical vapor deposition (PVD) by laser deposition [39].

The (ITO) layers are also formed by liquid phase deposition by electrochemical deposition techniques, co-precipitation of appropriate hydroxides, or sol-gel method [40]. During the deposition of (ITO) thin films, it is extremely important to determine the appropriate parameters that affect the optical, electrical, and mechanical properties, which results in their practical application [41].

The (ITO) electrodes are often used in electroanalysis due to their transparency, electrical stability, and low background current. In order to improve electrochemical efficiency, modification of electrodes (ITO) with different materials such as metal nanoparticles, quantum dots, graphene, graphite, carbon nanotubes, and metal oxides is carried out: TiO_2, CuO, NiO, WO_3 or ZnO [42].

There are several techniques to modify (ITO) electrodes by coating of the surface by modifier and drying, electrodeposition, and by immersion and evaporation in vacuum [42]. However, the most popular is the chemical method, where substances capable of forming ester bonds with -OH groups on the electrode surface are used.

Gold electrodes, due to their possibility of modification have found an extensive application in electroanalysis. In recent years, porous electrodes with an extended surface area, used in catalysis and the construction of new biosensors, have become of great interest [43]. The surfaces of gold electrodes can be modified by using alkanethiols and their derivatives. Alkanethiols on the surface of electrodes show the ability to form self-assembled monolayers (SAMs), which has a practical application in creating new types of sensors and biosensors. A great advantage of alkanethiols is the stability of the formed monolayers and the possibility of their further modification by chemical methods.

4.4 Discussion of the results: modification of (BDD)electrodes - their characteristics and properties

In the papers [H.1.] and [H.2.] of the described scientific achievement, I decided to develop procedures for the modification of (BDD) electrodes after prior modification of the electrode surface with allylamine using a microwave plasma reactor. The surface modification was performed in two steps. The first modification step was carried out to create a polymeric polyallylamine layer on the surface. It was performed in Greifswald (Germany) at the Leibniz Institute for Plasma Research and Technology (INP). In a second step, the obtained electrodes were further functionalized with the following groups of chemical compounds.

The first group of compounds used for functionalization was the fluorescent molecules Rhodamine 110 and L-tryptophan derivative (Fmoc-Trp(Boc)-OH. The selection of these molecules provided an opportunity to verify the effectiveness of the electrode functionalization process and characterize the newly obtained materials by electrochemical methods and by performing fluorescence measurements. The second group of compounds was 9,10-anthraquinone derivatives, which I had worked on after my Ph.D. The first used for electrode surface modification was the lysine derivative N^{α}-tert-butoxycarbonyl-L-N^{ε}-(9,10-dioxo-9,10-dihydro-anthracen-1-yl)-lysine (Boc-Lys(AQ)-OH), which I obtained by modifying a synthesis procedure reported in the literature previously [44]. By increasing the duration of the reaction and changing the ratio of the reactants used, I increased the reaction efficiency from 64.8% to over 90%. The second compound used for modification was a new, not described in the literature compound based on 9,10-anthraquinone, being a lysine derivative obtained as a poly-L-lysine dendrimer containing in its structure 4 redox-active molecules of 9,10-anthraquinone. This molecule was obtained on a solid support using Fmoc/tBu synthesis and purified by the HPLC technique, while the compound's identity was determined by mass spectra. I also used solid support synthesis to obtain other 9,10-anthraquinone analogs, described in [B.3.].

By the modification of the (BDD) electrode, I confirmed that polymerization by allylamine leads to the formation of a reactive film. As a result of covalent attachment reactions of selected molecules, I proved that the (BDD) electrode can be further modified. This was confirmed by electrochemical methods (cyclic voltammetry, CV), fluorescence methods, and the contact angle measurements of the electrodes before and after the modification process.

In the case of functionalization of the electrode surface with (Boc-Lys(AQ)-OH) and tryptophan derivative, I used the reaction for their attachment with N,N-diisopropylcarbodiimide (DIC) in the presence of 1-hydroxybenzotriazole (HOBt). During the attachment of Rhodamine 110 and the dendrimer derivative the reaction was carried out in the presence of N,N-diisopropylcarbodiimide (DIC) with catalytic amounts of 4-(dimethylamino)pyridine (DMAP). In both cases, the reactions were carried out in a mixture of methylene chloride (DCM) and dimethylformamide (DMF) in a volume ratio of 1:1. A scheme of the procedure used to modify the (BDD) electrodes is shown in Figure 1. In contrast, in Figure 2 I have placed the structures of the compounds that were deposited on the electrode as a result of the performed reactions.

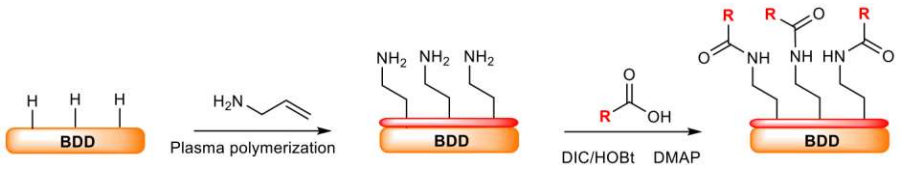

Figure 1. The scheme of the (BDD) electrode modification procedure [H.1., H.2.].

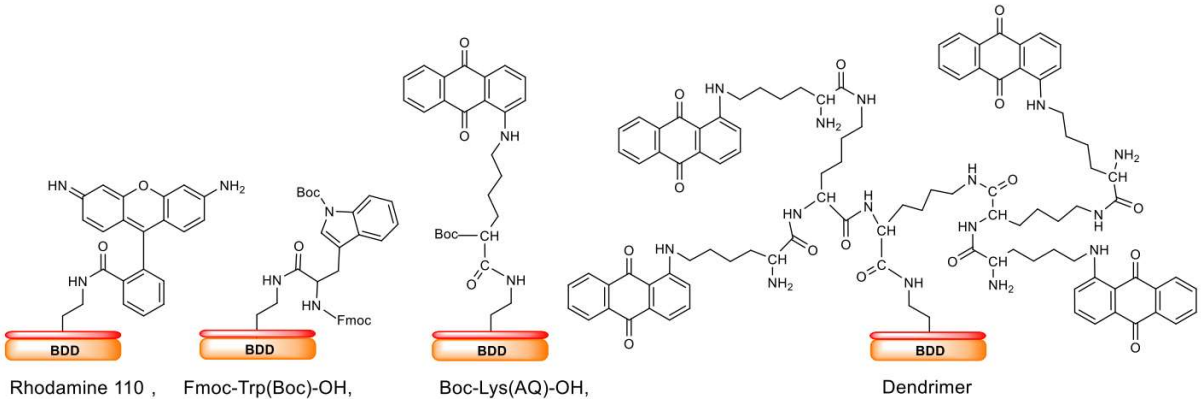

Figure 2. The chemical structures of compounds obtained on the (BDD) electrode by two-step surface modification [H.1., H.2.].

The electrochemical results for solutions containing the following redox standards: $[Fe(CN)_6]^{3-/4-}$, $Fe^{2+/3+}$ or quinone/hydroquinone presented in [H.1.] confirm that the deposited polyallylamine forms an insulating layer, blocking the charge transfer process. This is probably related to the interaction of amine groups present on the electrode surface. This process is independent of the measured pH in the range 5 to 9. An extremely important conclusion from the electrochemical measurements presented in [H.1., H.2.] is that the attachment of the investigated compounds to the surface of (BDD) electrode modified with polyallylamine, regardless of their type, significantly enhances the electron transfer through the double layer of the electrode.

The main conclusion of the [H.1.] work is that the (BDD) electrodes were modified with Fmoc-Trp(Boc)-OH, Boc-Lys(AQ)-OH, and a synthetic dendrimer based on L-Lysine, also enables the registration of fluorescent signals. However, the (BDD) electrode after modification with Rhodamine 110 shows the fluorescence as observed to the pure unmodified (BDD) electrode at low detection levels.

In the work [H.1.] I have proved that the surface functionalization process of a (BDD) electrode can be validated by both cyclic voltammetry and fluorescence techniques.

The functionalization of (BDD) surface electrodes with redox-active derivatives of 9,10-anthraquinone is an interesting matter due to the properties of the above molecules. Therefore, in a separate work I decided to compare the properties of electrodes modified with one molecule of Boc-Lys(AQ)-OH or with a dendrimer based on L-Lysine containing four moieties of 9,10-anthraquinone in the structure. The results presented in [H.2.] are a continuation of the work initiated in [H.1.] and constitute a separate group of investigations.

The analysis of the fluorescence measurements presented in [H.2.] shows that an increased amount of 9,10-anthraquinone molecules present in the modifying structure causes a decrease in fluorescence intensity. The fluorescence signal for the electrode modified with the

Boc-Lys(AQ)-OH derivative is higher than the signal recorded for the dendrimer derivative containing four 9,10-anthraquinone moieties in its structure.

In the paper [H.2.] I have proved a significant effect of the modification with L-lysine and 9,10-anthraquinone derivatives on the charge transfer kinetics, in comparison to the unmodified (BDD) electrode, and also that the charge transfer process is more efficient for modified electrodes regardless of the used derivative as a modifier.

In particular, the differences between modified and unmodified electrodes are observed in an electrolyte solution containing a positively charged redox probe $Fe^{2+/3+}$. Probably it is associated with the interactions of 9,10-anthraquinone with positively charged ions. The measurable current magnitudes in the solution containing $Fe^{2+/3+}$ are almost the same regardless of the modifier. The conclusion is that the modification of the (BDD) surface electrode by a redox-active molecule enhances charge transfer, regardless of the number of redox-active molecules in the modifying molecule.

In summary, in work [H.2.] I have proven that the electrode modified with 9,10-anthraquinone molecule not only has different properties from the unmodified one but also it was confirmed that the signals from 9,10-anthraquinone are not observed during electrochemical measurements.

In the next step of my research, I planned a modification of the (BDD) electrodes to obtain a highly sensitive surface in the presence of nucleic bases: adenine and guanine, as it was presented in the paper [H.3.]

Adenine (A) and guanine (G) belong to purine bases, whereas thymine (T) and uracil (U) belong to the pyrimidine bases. These compounds are the main components of nucleic acids, performing a key role in the most important biological processes occurring in every human cell. Since 1989, the research related to the use of electrochemical methods in the analysis of nucleic acids [45], which allow to determine inexpensively and quickly the DNA damage or to detect a specific fragment of the DNA chain using a specially designed electrode surface, has been extremely developed. The majority of electrochemical biosensors designed to monitor oxidative DNA damage are based on the detection of purines, mainly: adenine and guanine [46]. The above idea became my basic research objective, realized and described in the paper [H.3.].

In order to obtain a sensitive electrode for the presence of purines: (A) and (G) in the studied solution, I have functionalized the (BDD) electrode surface using poly-L-lysine. In paper [H.3.] I described and compared the efficiency of the electrochemical modification method and electrode modification "by immersion".

During the realization of the above goal, the electrochemical oxidation process of (A) and (G) was presented only on glassy carbon electrodes considered only differential pulse voltammetry (DPV) [47,48]. The detection of (A) and (G) on unmodified (BDD) electrodes were described only in one paper [49]. Therefore, I decided to modify the surface (BDD) electrode with poly-L-lysine in order to increase the biocompatibility of the investigated analytes, through the presence of both positively and negatively charged functional groups present within one molecule on the electrode. Such functionalization was expected to increase the detection ability of purine bases on the investigated electrode.

In [H.3.], for the first time, I have demonstrated the usage of poly-L-lysine as a (BDD) electrode surface modifying agent for the detection of purine bases. I have modified the (BDD)

electrodes using two different functionalization techniques, as shown in Figure 3a. In the first method described as modification by "immersion", I used a dilute solution of poly-L-lysine in water where the electrodes were placed for 24 hours.

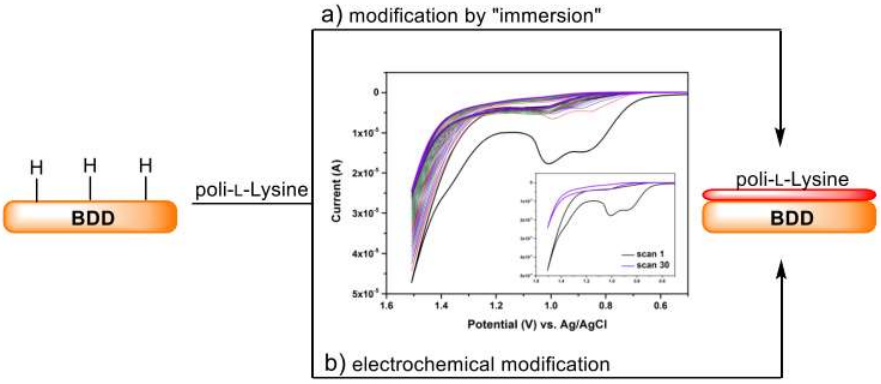

Figure 3. The scheme of electrode modification (BDD) (a) with poly-L-lysine solution by immersion (b) by the electropolymerization as described in [H.3.].

The second electrode modification (BDD), which I have proposed, was an electrochemical method of poly-L-Lysine deposition. In order to obtain a stable poly-L-lysine layer, I applied the (CV) method by performing 30 scans in the potential range of 0.5 V to 1.5 V vs. Ag/AgCl, in phosphate buffer solution (PBS) pH=7.4 containing poly-L-Lysine. Figure 3b shows the cyclic voltammetry obtained during (BDD) electrode modification. As a result of the conducted investigations, the decrease of oxidation currents up to a constant level was observed. This indicates that a stable coating has been obtained.

In the paper [H.3.] a comparative analysis of the obtained electrodes with unmodified (BDD) electrodes was presented using the following techniques: scanning electron microscopy (SEM), contact angle measurement, and X-ray photoelectron spectroscopy (XPS). Obtained results confirm that both methods cause a stable modification of electrodes. While, the comparative studies carried out applying the (CV) technique, performed in 0.5 M Na_2SO_4 solution containing different redox probes: negatively charged $[Fe(CN)_6]^{3-/4-}$, positively charged Fe^{2+}/Fe^{3+} and neutral quinone/hydroquinone system (Q/H_2Q) reveal the differences between the applied methods of electrode modification.

Poly-L-Lysine modification of both "by immersion" and electrochemically modified electrodes exhibits a wider potential window than the unmodified electrode, whereby the widest range is observed for an electrode modified by "immersion". Investigations performed in a solution containing a negatively charged redox probe $[Fe(CN)_6]^{3-/4-}$, usually applied as a sensitive probe for electrode surface modification [50], reveal that the significant changes were observed for the modified electrodes. Differences in voltammetry were observed for both modifications of the bare (BDD) electrode. However, the highest currents and the lowest peak-to-peak separation (ΔE) were obtained for the electrochemically modified (BDD) electrode. The calculated values of the electron transfer rate constant k^0 compared to bare (BDD) electrode are also different. Additionally, the investigations carried out in an electrolyte containing Fe^{2+}/Fe^{3+} as a redox probe also showed different electrochemical behavior to the bare (BDD) electrode. The electrochemical study in quinone/hydroquinone (Q/H_2Q) solution also

confirmed that the novel modified (BDD) electrodes catalyze the H_2Q oxidation reaction more efficiently than the bare (BDD) electrode.

The (DPV) experiments confirmed that the (BDD) electrodes modified by poly-L-Lysine are more sensitive for the detection of nucleic bases compared to the bare electrode and the electrochemically modified electrode is more sensitive in detecting adenine (A) and guanine (G). The lowest detected concentration of adenine (A) and guanine (G) using an electrode modified by "immersion" was 3 μM and 4 μM, respectively. The detection level was about two times higher in case of electrochemically modified (BDD) electrode.

While I was continuing my work on the detection of nucleic bases, I decided to develop and optimize a method for efficient functionalization of (BDD) electrode capable of detecting not only adenine (A) and guanine (G), but also caffeine (K) which belongs to purines. Therefore, as an electrode modifier, I used melamine (2,4,6-triamino-1,3,5-triazine), the presence of which on the electrode surface as polymelamine causes an increase in conductivity. It is caused by the delocalization of π electrons within the molecule located on the electrode surface [51]. Motivated by the above idea, in the work [H.4.], for the first time, I have presented the procedure of (BDD) electrode modification by melamine for the detection of adenine (A), guanine (G), and caffeine (K). For this purpose, I have adapted a modification procedure that has been previously used to functionalize other carbon-based electrodes: (GC) and multiwalled carbon nanotubes (MWCNTs) [52-54].

I have presented a method for the (BDD) electrode modification by melamine, conducting an electropolymerization process using (CV) technique. I achieved the intended effect as a result of successive 20 scans performed in the potential range from 0 V to + 1.6 V in 1 M H_2SO_4 solution containing 1 mM melamine, which was confirmed by (SEM) and (XPS) studies. The structure of the polymer formed on the electrode surface (BDD) and the modification process is shown in Figure 4. The voltammetry obtained during modification of the (BDD) electrode significantly differs from the voltammetry on the previously mentioned carbon electrodes.

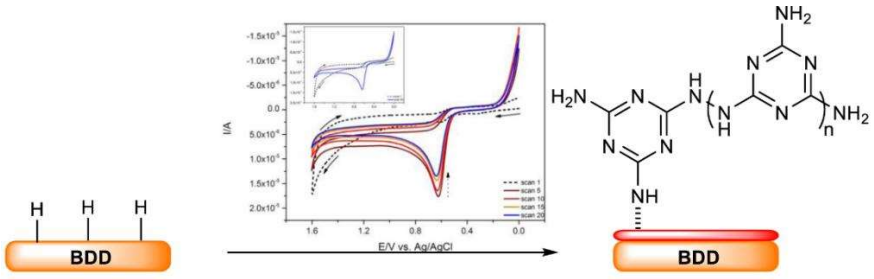

Figure 4. The scheme of BDD electrode modification with melamine by electropolymerization process [H.4.].

The mechanism of polymelamine formation on the surface involves the formation of HN-NH type bonds, between amino groups from two different melamine moieties.

The electrochemical measurements reveal that the modification of the electrode surface results in shortening electrode potentials range, especially in the negative potential range. Additionally, the changes of electron transfer kinetics in the electrolyte containing $[Fe(CN)_6]^{3-/4-}$ as a redox probe for the electrodes before and after modification is also observed.

The detection of adenine (A), guanine (G), caffeine (K) as well as single- and double-stranded DNA from calf thymus carried out on modified (BDD) electrodes using the (DPV) technique clearly confirms much higher detectability of the above analytes in comparison to bare (BDD) electrodes. The melamine modified (BDD) electrode allowed detection of adenine (A) at a concentration of 0.2 µM and caffeine (K) at a concentration of 0.4 µM, and it was 16 times more sensitive than the unmodified electrode. On the other hand, the obtained electrode was the most sensitive to guanine. The lowest detected guanine concentration on the unmodified electrode (BDD) was 98 µM, while on the melamine-modified electrode, the concentration was 0.023 nM. Comparison of guanine detection on both modified and unmodified (BDD) electrodes is shown in Figures 5a and 5b, respectively.

Analysis of real samples in the form of single- and double-stranded DNA also confirmed the higher sensitivity of the modified electrodes. Figure 5c shows a comparison of deoxyadenosine and deoxyguanosine oxidation obtained on bare a (BDD) electrode and a polymelamine-modified electrode. The additional signal observed in both voltammograms probably comes from the oxidation of other nucleosides [46,55].

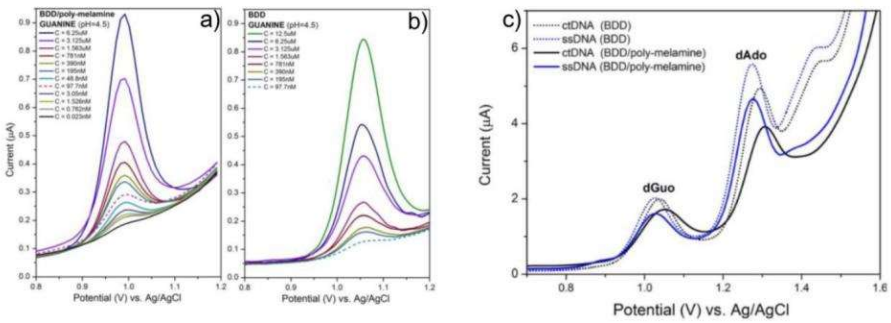

Figure 5. Comparison of guanine detection on (a) the modified (b) unmodified electrode (BDD) and (c) comparison of single double-stranded DNA detection on the above electrodes [H.4.].

In the next steps of my research, it was necessary to study the charge transfer kinetics and sensing properties of the boron-doped nanowalls (B:CNWs), a newly developed electrode material. Analyzing the charge transfer kinetics of (B:CNWs) electrodes, I found that these electrodes exhibited an extremely developed surface therefore I have decided to analyze nucleic bases and double-stranded DNA directly on these electrodes without modification.

In the paper [H.5.] I have presented the performed studies of electrodes (B:CNWs) for their use in the detection of adenine (A) and guanine (G). In the supplementary information to the above paper, I have included the results of analyzes of the application of nanowalls-type electrodes for the simultaneous detection of adenine (A), guanine (G), thymine (T), and cytosine (C), and separately only thymine (T) and cytosine (C).

During the simultaneous detection of adenine (A) and guanine (G), as shown in Figure 6a and 6c, I noticed thatvery high current signals are observed. These are noticed regardless of the applied technique and are much higher compared to the response of other electrodes for the normalized geometric electrode area. The discussed property results in high sensitivity of the investigated (B:CNWs) electrodes. The calculated limit of detection (LOD) values for were 1.36 µM and 1.60 µM for guanine and adenine, respectively.

The comparison of the sensitivity of adenine and guanine detection on a (B:CNWs) electrode with a boron-doped diamond electrode (BDD) [49], as well as on an unmodified (GC) electrode [48,56] or (GC) modified with oxidized graphene [57], boron-doped carbon nanotubes (B-CNTs) [58] and mesoporous carbon fibers [59], I have shown that the (B:CNWs) electrodes don't require modification and are ideal for the detection of adenine and guanine.

I have also noticed that for the investigated (B:CNWs) electrodes, irrespective of the used method, two ranges of linearity are observed, within the high and low concentrations of the analyte, as shown in Figures 6b and 6d. This is most likely due to the adsorption of adenine and guanine on the electrode surface.

The results of simultaneous analysis of all nucleic bases adenine (A), guanine (G), cytosine (C) and thymine (T) showed that applying the electrode (B:CNWs) it is possible to detect all nucleic bases simultaneously. The separate analysis of cytosine (C) and thymine (T) allows detection of these compounds at concentrations of 0.125 mM and 62 µM, respectively. During the analysis of double-stranded DNA, I have observed three simultaneous signals coming from guanine (G), adenine (A), and cytosine (C), which indicates the high sensitivity of the investigated electrodes.

In summary, in [H.5.] I have shown that boron-doped nanowalls (B:CNWs) electrodes are very suitable for detecting nucleic bases and DNA.

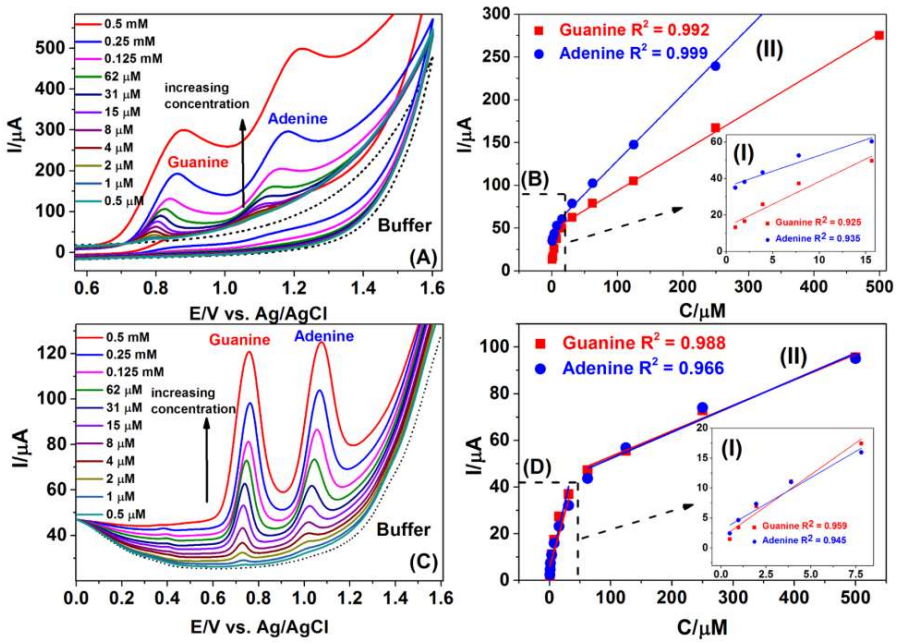

Figure 6. Voltammograms of guanine and adenine detection obtained by (a) (CV) (c) (DPV), and (b), (d) relationship of peak height to the concentration of investigated compounds [H.5.].

In the next step of my research, I carried out work related to the application of diamond electrodes for electroanalytical purposes. In papers [H.6.] and [H.7.], I have presented the results of optimization of the modification method for polycrystalline (BDD) electrodes [H.6.] and nanocrystalline boron-doped (B:NCD) electrodes [H.7.]. The main objective of the above works was the ability to attach polyclonal antibodies (anti-M1) to the surface of electrodes to detect the M1 protein, which was obtained from two types of H1N1 and H3N2 influenza virus.

The modification of the electrode surface was developed to detection of the M1 protein at low concentration levels.

In the first step of the described (BDD) electrode modification method, I applied an electrochemical surface functionalization with a diazonium salt, using 4-aminobenzoic acid and sodium nitrite as substrates. Using 4-aminobenzoic acid allowed me to obtain reactive carboxyl groups on the electrode surface. As a consequence of the electro-reduction reaction of the diazonium salt, the aryl radicals which are generated react with the electrode surface, permanently modifying it [60], as shown in Figure 7. This reaction enables further functionalization of the (BDD) electrode surface.

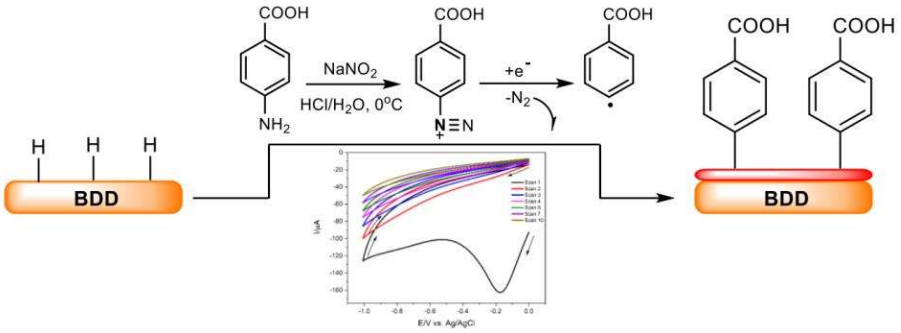

Figure 7. Mechanism of (BDD) electrode modification using diazonium salt of 4-aminobenzoic acid. Cyclic voltammetry obtained via (BDD) electrode modification [H.6., H.7.].

The next step of (BDD) electrode surface modification involved attaching a polyclonal anti-M1 antibody (aM1) to the electrode surface, which resulted in the preparation of an electrode capable of detecting influenza virus M1 protein. An antibody attachment reaction was carried out using a mixture consisting of 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC) as an activator of the carboxyl group and (N-Hydroxysuccinimide) (NHS) to obtain the active ester [61]. This reaction allowed to attach the anti-M1 antibody to the electrode surface. The scheme of the performed reaction is shown below in Figure 8.

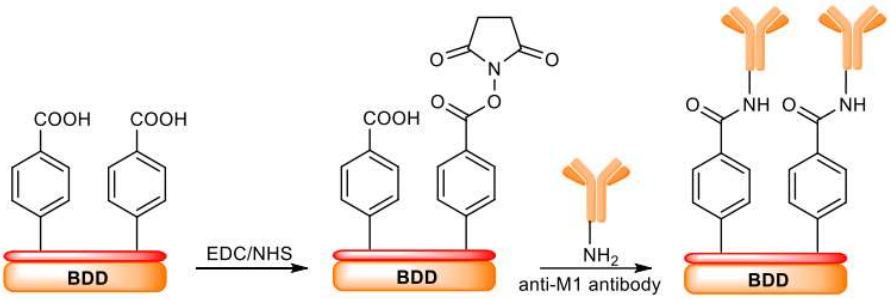

Figure 8. The scheme of (BDD) electrode modification with aM1 polyclonal antibody [H.6., H.7.].

As a consequence of attachment of polyclonal aM1 antibody to the electrode, efficient detection of influenza M1 virus protein on different electrodes was described in papers [H.6.] and [H.7.]: in paper [H.6.] polycrystalline boron-doped diamond (BDD) electrodes were used, while in paper [H.7.] nanocrystalline boron-doped diamond (B:NCD) electrodes were applied.

The developed method of attaching aM1 antibody to the electrode surface enabled detection of influenza virus M1 protein from different influenza A virus subtypes. The resulting electrode surface was used to study the detection of influenza virus after blocking the empty sites with bovine serum albumin (BSA), preventing non-specific protein interactions due to physisorption on the electrode substrate. A scheme of the electrode modification is shown in Figure 9, the structure of the influenza A virus M1 protein is based on the crystallographic structure presented in [62].

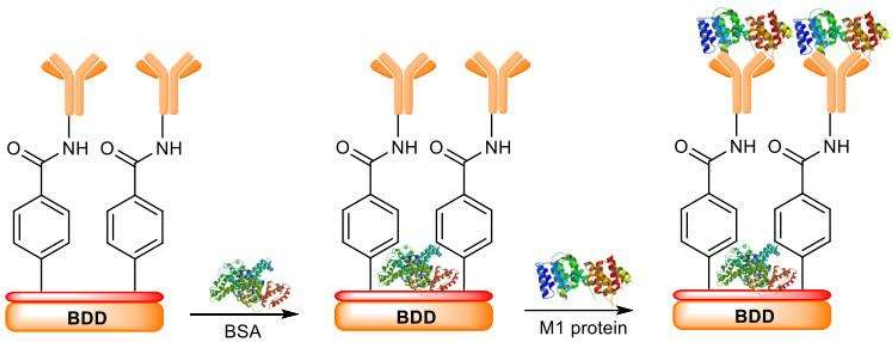

Figure 9. The detection scheme of the M1 protein on modified [H.6., H.7.].

In the paper [H.6.], the electrochemical impedance spectroscopy (EIS) method was used to prove that the obtained electrodes have a limit of detection (LOD) of about 1×10^{-15} g/ml (1 fg/ml) in the examined solution. The reaction time of the modified electrodes was also checked, and it was found to be very short, at only 5 minutes. The experiments were also carried out after incubating the electrodes in artificial saliva containing M1 virus protein, obtained from the H1N1 virus, in the presence of yeast cells (*Candida Albicans*) and bacteria (*Streptococcus Aureus*). The obtained measurement results confirmed the extreme specificity of the antibodies used for modification and, consequently, the universality of the proposed method for (BDD) electrode functionalization. Due to the fact that the influence of the investigated pathogens is negligible, the obtained electrodes offer a potential opportunity for commercialization.

In the paper [H.7.] boron-doped nanowalls (B:NCDs) electrodes were used to compare the effect of substrate on the specificity of the interaction of a polyclonal anti-M1 antibody (aM1), for the detection of influenza virus M1 protein. Conducting an analogous modification of the (B:NCD) electrodes, it was shown that the results obtained for the M1 virus protein determination were at a very low level of detection. The calculated (LOD) was equal to 5×10^{-14} g/ml (50 fg/ml), which corresponds to the detection of several units of influenza virus. The modified electrodes (B:NCD) not only have unusual sensitivity but, similar to the electrodes (BDD), their use as a sensor allows for the detection of influenza M1 virus protein within 5 minutes. These electrodes also exhibit great stability because even after 3 weeks of storage they retain approximately 85% of their original sensitivity.

In summary, the main achievement of the conducted research was the development and optimization of an efficient method to modify diamond (BDD) and (B:NCD) electrodes to attach an anti-M1 polyclonal antibody to the electrode surface for the detection of influenza A virus M1 protein. The applied method proved to be very effective, and the performed measurements allowed to obtain phenomenal detection limits of the M1 protein, several orders

of magnitude lower than previously proposed methods for detecting the influenza virus protein. In addition, the proposed electrode modification method has been adapted to attach other biomolecules, therefore papers [H.6.] and [H.7.] are highly cited by other researchers. According to the Scopus database, paper [H.6.] has been cited 61 times and the paper [H.7.] 30 times (as of 24.01.2022).

Based on the experience gained as described in papers [H.6. and H.7.], I have developed an efficient surface modification method of (BDD) electrode for rapid recognition of genetic material. The effectiveness of the proposed method was verified for the detection of the DEFB1 gene and was presented in the paper [H.11.]. The detection of the DEFB1 gene, which encodes the beta-defensin 1 protein in humans, is an extremely important issue. This gene is found only in (*Homo sapiens*) and its identification allows differentiation of genetic material from human and animal origin. Achievement of an aptasensor capable of differentiating the human DEFB1 gene in a very short time may be crucial during the initial analysis of genetic material samples of unknown origin in order to eliminate and pre-differentiate samples of animal origin, which may have remarkable applications not only in medicine but also in forensic analysis.

In the paper [H.11.], I presented a modification method of (BDD) electrode with an oligonucleotide of the sequence: 5'- CCC AGT TCC TGA AAT CCT GA-3'. This modification, together with the usage of the dynamic electrochemical impedance spectroscopy (DEIS) technique, enabled a successful study to identify genetic material extracted from saliva of human origin and to distinguish it unambiguously from animal material.

My main contribution to the paper [H.11.] was the development of the (BBD) electrode functionalization, its verification, and the subsequently performed electrochemical measurements determining the efficiency of the obtained aptasensor. In the first step, based on the procedure of electrode modification described in papers [H.6. and H.7.] I used diazonium salt of 4-aminobenzoic acid to obtain reactive carboxyl groups on the electrode surface by electro-reduction of diazonium salt of 4-aminobenzoic acid. In a second step using a mixture (EDC/NHS), I attached a modified oligonucleotide H_2N-(CH2)6-5'- CCC AGT TCC TGA AAT CCT GA-3, complementary to the DEFB1 gene, to the electrode surface with amide bond formation. The modification of the electrode surface with an oligonucleotide allowed the analysis of genetic material previously collected and isolated from humans and animals. Directly before electrochemical measurements, a denaturation process was performed to obtain a single strand of DNA. A scheme of the electrode modification, as well as the idea of the denaturation of the biological test samples used for the experiments before the measurements, is shown in Figures 10 a and 10 b.

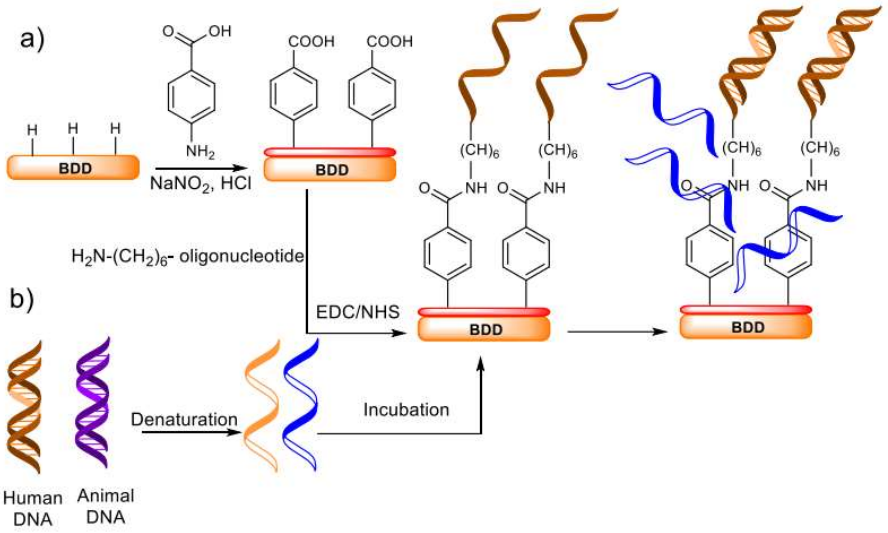

Figure 10. A scheme of (a) (BDD) electrode modification with 4-aminobenzoic acid and oligonucleotide, (b) DNA denaturation and incubation with functionalized electrode (BDD) [H.11.].

The obtained results by dynamic impedance spectroscopy technique in potentiodynamic electrode polarization (pDEIS) mode, in the potential range of - 1 V to +1 V vs Ag/AgCl allowed us to monitor the hybridization process of ssDNA of the DEFB1 gene to the complementary strand present on the electrode surface.

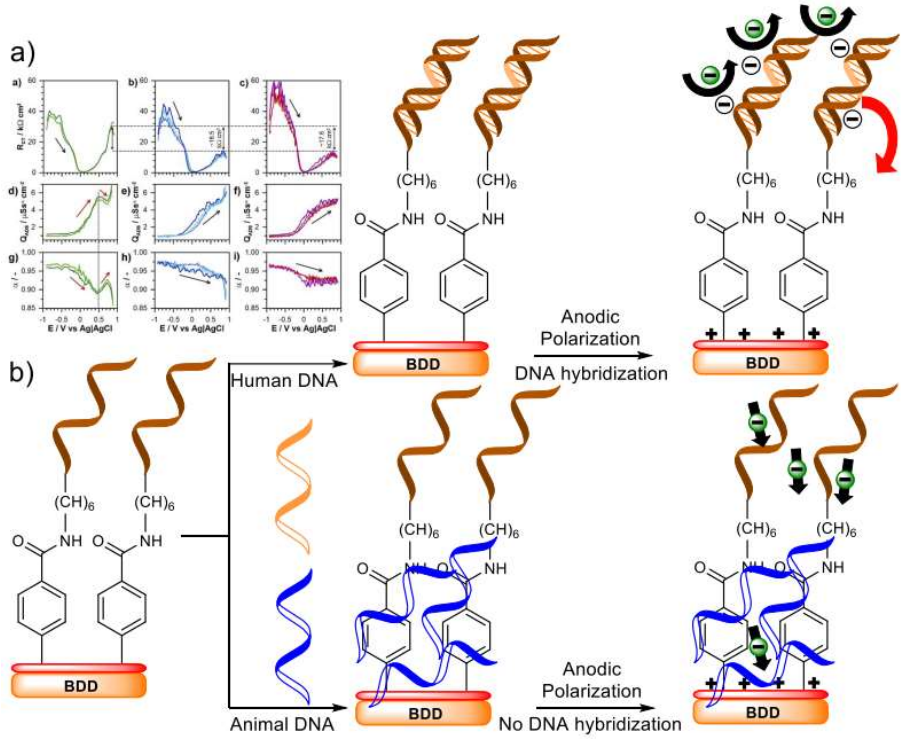

Figure 11. a) Results of capacitance changes observed during measurements b) Scheme of DNA denaturation and DNA hybridization process on (BDD) electrode surface [H.11.].

The obtained results were analyzed using a suitable equivalent circuit to identify the characteristics of changes in electrical parameters during the measurement. Based on these results, it was clearly indicated that, in particular, the obtained values of charge transfer resistance (Rct) for human DNA are almost twice higher (under conditions of positively charged electrode surface) than in the case of any other samples of animal origin (household pets and domestic fowl). The above-mentioned results are attributed to the differential interactions of the negatively charged $Fe(CN)_6^{3-/4-}$ redox couples with the positively charged electrode surface containing double or single-stranded DNA. Consequently, this allowed us to follow the DNA hybridization process on the electrode surface, as shown in Figure 11 b. The applied measurement methodology also allowed us to follow the changes in orientation of DNA double helix formation relative to the positively charged electrode surface, shown by the recorded capacitance changes (Figure 11 a). The optimal potential where the most significant changes in impedance spectra were observed was 0.85 V.

In conclusion, the main advantage of the (BDD) electrode modification procedure which I had proposed was the ability to detect the DEFB1 gene, [H.11.] in a short time, much shorter in comparison to traditional DNA hybridization analyses at very low concentration levels of 1 ng/ml. In paper [H.11.], I also proved that the process of DNA hybridization on the modified electrodes is not observable in the case when the examined sample was the material collected from animals due to the lack of DNA double helix formation on the electrode surface.

In the paper [H.5.] I had presented the usage of boron-doped nanowalls (B:CNWs) electrodes for the detection of nucleic bases and DNA. The obtained results confirmed the remarkable sensitivity of the electrodes towards the determined analytes. Continuing my research on (B:CNWs) and (BDD) electrodes, I decided to detect paracetamol on unmodified surfaces. The results of paracetamol analysis by (CV) and (DPV) techniques were presented in the paper [H.8.]. The choice of studied analyte was dictated by the needs to find a fast and sensitive method for the determination of paracetamol, due to the extremely high increase in the consumption of the mentioned drug and its occurrence in the environment.

Based on the results obtained by the (CV) technique at different pH, I established a different mechanism of oxidation of paracetamol on the two electrodes. The oxidation process at the electrode (BDD) is a two-electron involving one proton, while at the (B:CNWs) electrode, the mechanism is a two-electron including two protons. Additionally, based on the analyzed voltammograms obtained for different scan rates, I proved that the mechanism of paracetamol oxidation at the (B:CNWs) electrodes is a diffusion-controlled process while the oxidation process at the (BDD) electrode has a different nature. Taking into account the high difference in the cathodic and anodic current peaks, it was suggested that the oxidation of paracetamol on the (BDD) surface occurs more efficiently than its reduction or the oxidation products inhibit the electrode surface. The obtained voltammograms are shown in Figures 12 a and 12 b.

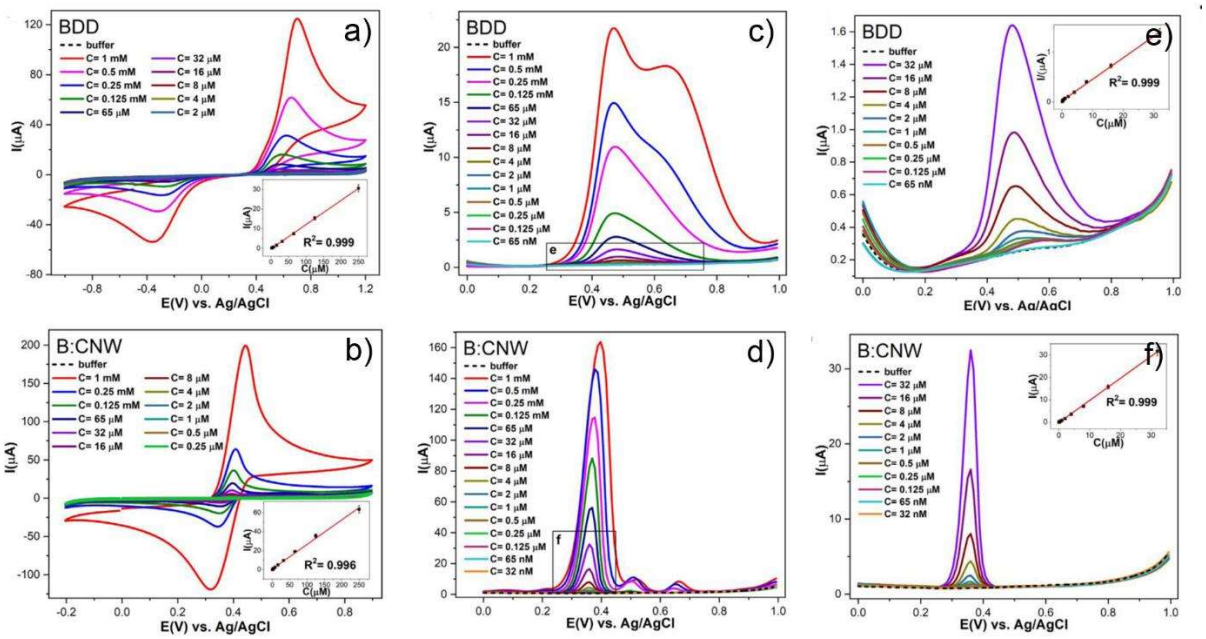

Figure 12. Voltammograms obtained (a), (b) from (CV) measurements, (c) (d) from (DPV) measurements, (e), (f) magnifications of (DPV) obtained on (BDD) and (B:CNWs) electrodes [H.8.].

The (CV) results show a linear relationship between the increase in the concentration of the analyte and the peak height at the cathode and anode, as well as a shift in potential values for the reduction and oxidation peaks at both electrodes. For the (B:CNWs) electrode, a higher level of electrode process reversibility is observed in comparison to the (BDD) electrode for the same concentrations of paracetamol. The lowest measurable concentration was 2 μM and 0.25 μM for the (BDD) and (B:CNWs) electrodes, respectively, which similarly to the previously obtained results indicates the superiority of the (B:CNWs) electrode over the (BDD) electrode. There is also a difference in the oxidation of paracetamol observed based on the results obtained by the (DPV) technique. The additional peaks are obtained on both electrodes as a consequence of the electropolymerization process by the oxidation products, during the analysis in the range of high concentrations. In the case of the (BDD) electrode, an additional signal appears that interferes with the analyzed peak. In contrast, in the case of the (B:CNWs) electrode, two additional peaks are observed, but at different potential values, which consequently does not affect the detection sensitivity (Figure 13 b and 13 c), and magnifications at (Figure 13 e and f). The calculated limits of detection (LOD) are 0.430 μM for the electrode (BDD) and 0.281 μM (B:CNWs), respectively. The obtained paracetamol detection limits for the described electrodes are much lower than those reported in the literature. I have shown that (BDD) and (B:CNWs) do not require modification and are ideal for electrochemical detection of paracetamol.

It was also proved that the (B:CNWs) electrode was perfect for analyzing paracetamol at very low concentrations, which was carried out in artificial urine. The concentration range in which the (B:CNWs) electrode gave analytical signals was from 65 nM to 1 μM, while the calculated limit of detection (LOD) was 75 nM.

In summary, in the paper [H.8.] I have presented a comparative analysis of (B:CNWs) and (BDD) electrodes for the detection of paracetamol. The obtained results established the oxidation mechanisms of paracetamol on the investigated electrodes and determined the detection limits (LOD). The result obtained allowed us to determine the mechanisms of paracetamol oxidation on the electrodes studied and the limits of detection (LOD).

Based on the studies presented in [H.8.] and [H.5.], the superiority of the (B:CNWs) electrodes over the (BDD) electrodes in sensory measurements of nucleic bases, DNA, and paracetamol is clearly established.

4.4 Discussion of the results: Modifications of oxide (ITO) and gold (Au) electrodes - their characteristics and properties

Continuing my drug detection research, in my next paper, I decided to analyze the popularly used ketoprofen. Therefore, the main objective of the [H.9.] work was to investigate the mechanism of its oxidation on the surface of an optical fiber sensor coated with a thin film (ITO) and to verify the deposition efficiency of ketoprofen on an electrode (GC) and an electrode made of commercial (ITO).

Using an optical fiber sensor coated with a thin film (ITO) enables simultaneous optical and electrochemical detection in real-time. The possibility of developing the electrochemical investigations and complementing them with other methods allows for obtaining new sensory materials. Following the above idea, I obtained the consortium grant "Conductive photonic structures for multiparametric biochemical diagnostics" Sonata Bis 4, no. 2014/14/E/ST7/00104, I was the Principal Investigator on the University of Gdansk side.

In the paper [H.9.], I have presented for the first time the detection of ketoprofen by modifying the ITO layer on the surface of the optical fiber by its oxidation. The most common method for ketoprofen analysis is HPLC chromatography coupled to a mass spectrometer [63], UV [64] or fluorescence detection [65]. There are also a few literature reports considering the application of electrochemical techniques using a mercury drop electrode [66] or an ion-selective electrode [67]. There was only one work on ketoprofen oxidation at (BDD) electrodes when this study was conducted [68], where the authors were focused on the decomposition of the ketoprofen rather than its analysis. The paper [H.9.] was the first on the electroanalysis of ketoprofen.

In the above work, I developed a procedure for the electrochemical deposition of ketoprofen on an (ITO)-coated fiber optic sensor and I modified the (GC) and (ITO) electrodes with ketoprofen.

I performed ketoprofen's oxidation by the (CV) technique at different concentration ranges from 1 μM to 1 mM, on an ITO coated fiber optic sensor, a scheme of which is shown in Figure 13 a. The resulting voltammograms of the fiber optic sensor modification with a ketoprofen concentration of 2 mM are shown in Figure 13 b. The electrochemical characterization of the electrode, confirming the changes in the properties of the modified sensor, is also shown in Figure 13 b. As a result of the performed modification process of the fiber optic sensor, by oxidizing the carboxyl group of ketoprofen on the electrode surface, the deposition of the resulting product occurred, which was also simultaneously monitored by optical measurements. In Figure 13 c, I have presented a plot of the wavelength dependence of

the transmission (T) recorded for two boundary concentrations of ketoprofen during the 1 µM and 1 mM polymerization process, which illustrates the dependence of the recorded magnitudes of change during the oxidation of ketoprofen. The most significant changes are observed at a wavelength around λ_R = 650 nm.

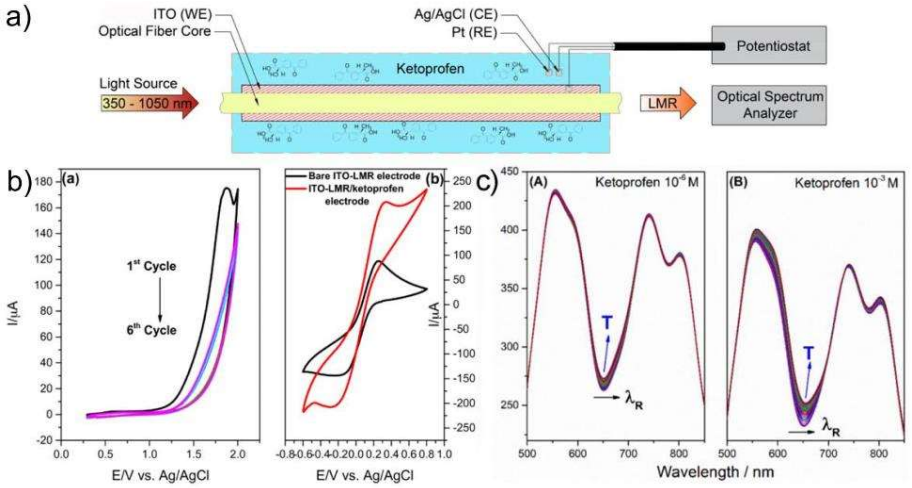

Figure 13. a) Schematic representation of the fiber optic sensor used in the experiments and the idea of the experiments conducted, b) the sensor modification process and cyclic voltammograms characterizing the fiber optic sensor before and after the modification process in 0.5 Na_2SO_4 solution containing 5 mM $[Fe(CN)_6]^{3-/4-}$, c) changes in the optical response of the studied sensor recorded during electropolymerization of ketoprofen on ITO surface for two boundary concentrations [H.9.].

In the above work, I have described and characterized for the first time the oxidation of ketoprofen on electrode materials other than the fiber optic sensor. The (GC) electrode and the (ITO) coated glass electrode was chosen as reference materials, which covered with a layer of ketoprofen show different electrochemical properties compared to the unmodified bare electrode. The obtained results confirm the effectiveness of the described method of modifying the electrode surface, which is illustrated in Figures 14 a and 14 b. In conclusion, in paper [H.9.], I have presented a method to oxidize ketoprofen to coat the surface of a fiber optic sensor, thus enabling its detection.

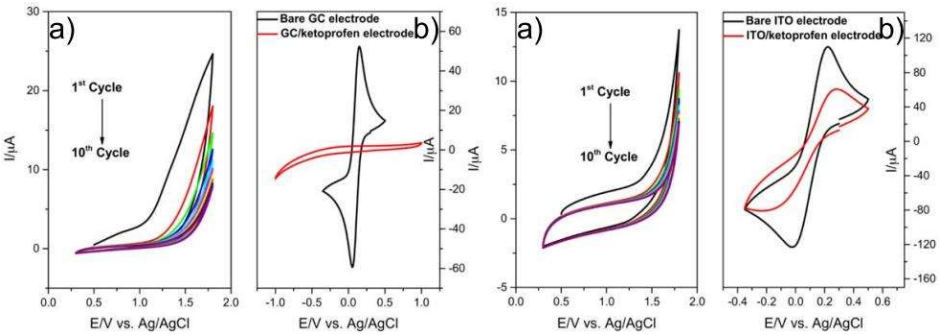

Figure 14. The cyclic voltammograms illustrating (a) the modification processes of the electrodes (b) electrochemical characterization of the electrodes in 0.5 M Na_2SO_4 solution containing 5 mM $[Fe(CN)_6]^{3-/4-}$ on the (GC) and (ITO) electrodes [H.9.].

Continuing the research work, which includes modifying optical fiber sensors covered with a transparent layer (ITO), in the work [H.10.] I used the above sensor for the detection of biomolecules. My role in the paper [H.10.] was to develop an electrochemical method for modifying a fiber optic sensor to detect avidin at various concentrations. The selection of avidin detection was performed due to the fact that this protein forms a complex with biotin, which is considered to be the strongest non-covalent bond found in nature [69]. Additionally, the avidin's affinity for biotin is used as a tool in many diagnostic tests, therefore it was extremely important to verify if the optical fiber coated by (ITO) could be effectively modified with biotin for avidin recognition.

The method I proposed for the electrochemical deposition of biotin hydrazide on the electrode surface to selectively bind avidin was based on a previously described procedure [70]. In the described method I have made a few changes optimizing the ranges of applied potentials and the number of repetitions carried out to achieve a stable layer on the surface of the fiber optic sensor. I utilized the biotin hydrazide electrodeposition method using 10 repetitions from 0 V to + 1.2 V vs Ag/AgCl, according to the mechanism shown in Figure 15 a. The main advantage of the presented electrodeposition process presented in Figure 15 b is the short time to obtain a stable coating. There are two postulated mechanisms to explain the occurring reaction. The first is related to the oxidation of the C=O group, while the more likely one involves the formation of a radical cation due to the oxidation of the hydrazide group [71]. The confirmation that the applied method to the modification of the fiber optic sensor is effective is indicated by the voltammetry and optical changes recorded simultaneously before and after the electrode functionalization process, as shown in Figures 15 c and 15 d.

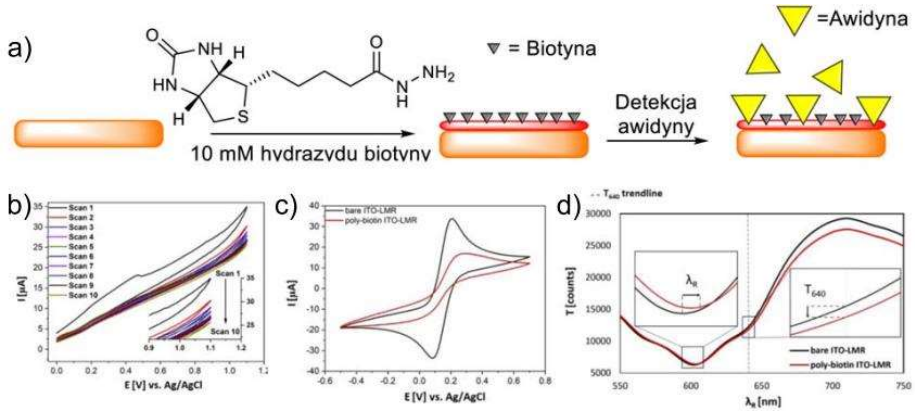

Figure 15. a) Scheme of electro-deposition on the surface of a fiber optic sensor, (b) Cyclic voltammograms obtained by electrochemical deposition of biotin hydrazide, (c) electrode characteristics before and after the electro-deposition process, (d) optical changes recorded before and after the electro-deposition process obtained in 0.1 M KCl solution containing 1 mM $Fe(CN)_6^{3-/4-}$ [H.10.].

Summarizing, as a result of the optical and electrochemical measurements described in [H.10.], I have demonstrated that an optical fiber sensor coated with (ITO), was modified with biotin and showed a response in the avidin concentration range from 0.01 to 0.1 mg/mL. I have presented a new electro-deposition method on the surface of a fiber optic sensor using biotin hydrazide.

In the further development of the electrode surface modification, I conducted a series of experiments using a different electrode material than I had used before. The selection of the used electrode material was related to the selection of the surface modification procedure as well as the substrates used in the modifications. Therefore, I decided to perform my research using gold electrodes to obtain electrodes capable of detecting the programmed death-ligand 1 (PD-L1) after their modification. I decided to verify whether the obtained electrodes would be capable of binding programmed cell death protein-1 (PD-1).

On the basis of a previous study by the co-authors of paper [H.12.], 1-[[3-bromo-4-[(2-methyl[1,10-biphenyl]-3-yl)methoxy]phenyl]methyl]-2-piperidinecarboxylic acid) (Figure 16 b) abbreviated as (BMS-8), interacts with the programmed death-ligand 1 (PD-L1) protein [72].

My role in the work [H.12.] was to develop an efficient method to attach the BMS-8 molecule to the electrode surface and to verify the effectiveness of the modification, leading to the design of a protein sensor (PD-L1).

Taking advantage of the BMS-8 molecule, I developed a method for its attachment to the surface of a gold electrode to detect the programmed death-ligand 1 (PD-L1) protein. An important result of my contribution was the successful differentiation of (PD-L1) from programmed cell death protein-1 (PD-1) - extremely important markers for many cancers. Selective identification of both compounds was possible based on capacitance characteristics analysis in technique (EIS) measurements. In the above work, I planned and carried out all electrochemical tests, their analysis, and interpretation.

The studies presented in paper [H.12.] allowed verification of the thesis that the programmed death-ligand 1 (PD-L1) protein and programmed cell death protein-1 (PD-1) would be detected using a gold electrode shown in Figure 16 and modified to anchor a molecule (BMS-8) on its surface. I modified the electrode in two stages. In the first stage, I modified the surface of the gold electrode with cysteamine, which gave the amino groups on the surface. In a further step, the above modification allowed attachment of (BMS-8) using EDC and NHS. The modification process is shown in Figure 16 c.

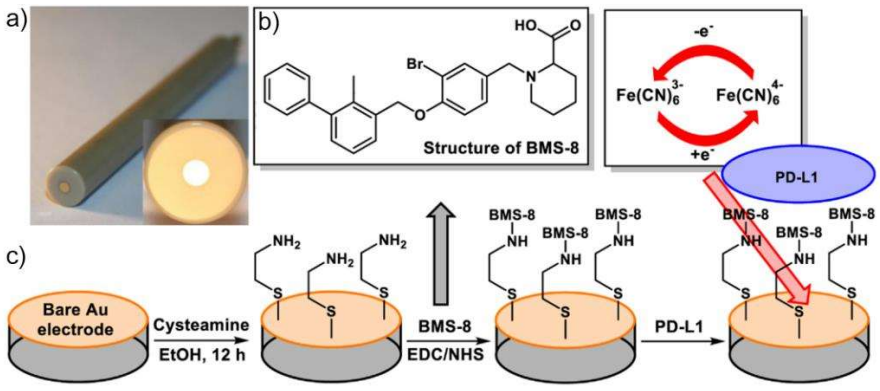

Figure 16. a) Picture of the gold electrode used for the study, b) formula of the BMS-8 moiety, c) scheme of the gold electrode modification [H.12.].

The efficiency of the modification process was confirmed by several methods, including (CV) and (EIS) as well as spectroscopy (XPS) and contact angle measurement [H.12.]. The resulting electrodes obtained by the above surface functionalization method were used to detect

the protein-ligand (PD-L1) and the electrode sensitivity was verified for the presence of the protein (PD-1). The analysis of my study shows that effective detection of (PD-L1) in the concentration range from 10^{-8} to even 10^{-18} M is possible using (EIS), and the calculated limit of detection (LOD) for (PD-L1) was 1.87×10^{-14} M, as shown in Figure 17 a. The results of (CV) analyses confirm the change in voltage-current characteristics at a concentration of (PD-L1) of 1×10^{-14} M, indicating the high sensitivity of the developed sensors. In order to check the selectivity of the obtained electrode for the presence of other proteins, a series of measurements were performed taking into account the detection of programmed death protein-1 (PD-1), BTLA, and CD160. The impedance measurement results clearly indicate that BTLA and CD160 proteins at a concentration of 1×10^{-8} M bind to the functionalized electrode surface with anchored (BMS-8) at a much lower degree than (PD-L1). Detailed measurement results are included in the supplementary information to the paper [H.12.].

The selectivity studies also revealed that although the BMS-8 molecule itself does not interact with the protein (PD-1) [73] the electrode exhibits a change in impedance characteristics over a wide range of protein concentrations, from 10^{-18} M to 10^{-8} M, as shown in Figure 17 b. In the paper [H.12.], it was hypothesized that the interaction (PD-L1) with the investigated electrode is probably specific to BMS-8, while the analogous interaction (PD-1) is a consequence of less well-defined surface interactions occurring on the functionalized electrode.

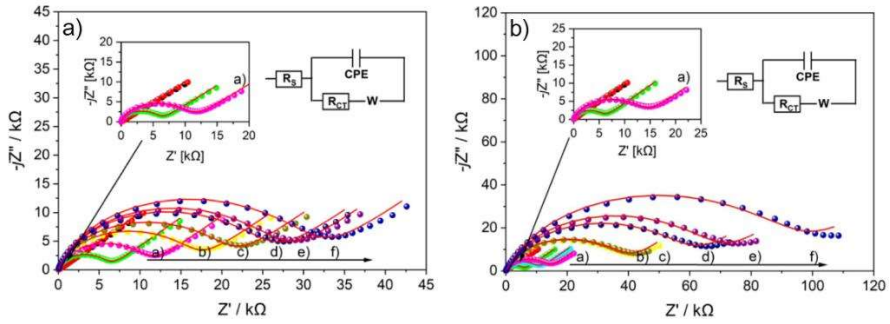

Figure 17. Impedance spectra obtained for electrodes modified with BMS-8 moiety in the absence and presence of (a) (PD-L1) protein and (b) (PD-1) protein, captured in 0.01 M PBS solution, pH 7.0, at different concentrations from (a) to (f): 10^{-18}, 10^{-16}, 10^{-14}, 10^{-12}, 10^{-10} and 10^{-8} M [H.12.].

Furthermore, in the paper [H.12.] I have shown that on the basis of the analysis of the obtained impedance spectra, it is possible to differentiate the impedance characteristic, specific for the programmed death-ligand 1 (PD-L1) protein and its receptor, programmed death-1 (PD-1), which is undoubtedly a significant achievement presented in the paper [H.12.].

I made the distinguish between (PD-L1) and (PD-1) based on the difference in the capacitance of the functionalization layer and the differences in the electrical inhomogeneity of the surface of the modified electrode, resulting from the so-called frequency dispersion of capacitance. The result of the impedance analysis is shown in Figure 18. The quasi-capacitance parameter increases with increasing analyte concentration for both, (PD-L1) and (PD-1). I also observed significant changes in the electrical homogeneity parameter due to the adsorption of both analyzed compounds. In the case of (PD-L1), the electrical homogeneity of the investigated electrodes is much higher than in the case of (PD-1) (higher value of the constant-

phase element exponent α). The phenomenon of the formation of more homogeneous surface distribution of the adsorption layer in the case of (PD-L1) could be attributed to the formation of a homodimer on the electrode surface, also confirmed by X-ray analysis [72]. For the analysis of electrical homogeneity during protein detection (PD-1), it can be concluded that the interaction is non-specific and may be random.

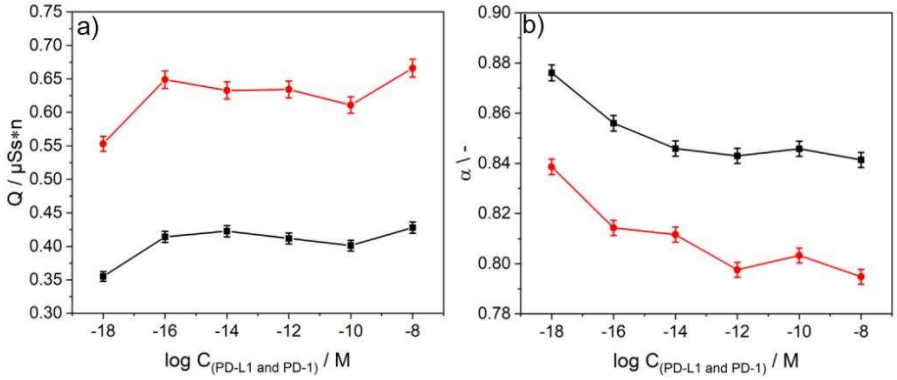

Figure 18. Relationship of (a) the calculated quasi-capacitance (Q) and (b) the heterogeneity coefficient with the concentration of proteins (PD-L1) (black line) and (PD-1) (red line) respectively expressed on a logarithmic scale [H.12.].

To summarize, in work [H.12.], I had presented for the first time an efficient method to modify a gold electrode with a BMS-8 moiety performed a detection study of programmed death-ligand 1 (PD-L1) protein, and performed an advanced analysis of impedance spectra to distinguish (PD-L1) from programmed cell death protein-1 (PD-1). Significantly, the capacitance characteristics for both analytes are significantly different irrespective of the concentrations analyzed, providing a clear distinction between co-occurring analytes. In addition to the physicochemical analysis of the obtained electrode surface, I performed an analysis of the effect of other BTLA and CD160 proteins on the sensitivity of the investigated electrodes.

4.6 Summary of scientific achievement

Based on the above scientific achievement, the most important conclusions concerning the modification of electrode surfaces and their use for sensory purposes are that:

- I have developed an efficient procedure for surface modification of (BDD) electrodes pretreated with allylamine with two groups of compounds: fluorescence-displaying and redox-active 9,10-anthraquinone derivatives [H.1. and H.2.].
- I have developed and compared the effectiveness of (BDD) electrode modification with poly-L-L-lysine by electrochemical and "by immersion" methods, efficiently detecting nucleic bases. The minimum detected concentration using the electrochemically modified (BDD) electrode was 6 µM and 8 µM for adenine (A) and guanine (G), respectively, whereas on the electrode was modified by "immersion" the detection level was two times lower [H.3.].
- I have shown that by my electrochemical modification of the (BDD) electrode with melamine, the detection of adenine, guanine and caffeine is possible at 0.2 µM, 0.023 nM and 0.4 µM levels. I have also demonstrated the modification of the electrode surface with melamine can be used for the detection of real single and double-stranded DNA samples [H.4.].
- I have defined for the first time the possibility of using (B:CNW) in electroanalysis, and I have demonstrated its superiority over the (BDD) electrode. I proved that the limit of detection (LOD) of guanine and adenine using the (B:CNW) electrode was 1.36 µM and 1.60 µM, respectively. I also proved that the (B:CNWs) electrode could be used for the detection of double-stranded DNA (ctDNA) [H.5.].
- Based on my contributions, the aptasensors and immunosensors have been developed with extraordinary low limits of detection (LOD). I have developed an efficient procedure for the surface modification of two types of electrodes: (BDD) and (B:NCD), allowing the attachment of polyclonal antibodies (anti-M1), resulting in the detection of M1 protein from two types of the H1N1 and H3N2 influenza virus. [H.6. and H.7.]. The modification which I have proposed allowed the detection of M1 protein at extremely low levels of detection (LOD) equal to (1 fg/ml) and (50 fg/ml) detected with (BDD) and (B:NCD) electrodes.

  I have also modified the electrode surface (BDD) with a specially selected oligonucleotide allowing detection of the DEFB1 gene at very low concentrations (1 ng/ml), which consequently allowed differentiation between human and animal genetic material [H.11.].
- I have also developed detection methods based on non-functionalized electrodes. I proved the superiority of electrodes (B:CNWs) over electrodes (BDD) by analyzing their ability to detect paracetamol, with the limit of detection (LOD) 0.430 µM for (BDD) electrode and 0.281 µM (B:CNWs), respectively [H.8.].
- I have performed an effective procedure for modifying an optical fiber coated with (ITO) performing oxidation of ketoprofen on the fiber surface, which enabled its tracking in concentration ranges from 1 µM to 1 mM [H.9.]. I also developed an

electrochemical method to modify a fiber optic sensor with a thin film (ITO) of biotin to detect avidin at concentrations from 0.01 to 0.1 mg/mL [H.10].

- I have developed a modification method of the gold electrode surface with a molecule (BMS-8) for the ultra-sensitive detection of the programmed death-ligand 1 (PD-L1) protein, with a detection limit (LOD) of 1.87×10^{-14} M. I have also obtained the ability to differentiate the analyzed (PD-L1) from its receptor, programmed death-1 (PD-1) [H.12.].

4.7 The directions for future research

In the near future, my planned work will focus on the search and electrochemical characterization of new electrode materials allowing the detection of single organic compounds and biomolecules such as bacteria, viruses, or proteins which are difficult to detect with traditional analytical techniques. To achieve this goal, I have already established the necessary cooperation with other research centers and obtained the necessary funding for this purpose.

I have also planned to continue research with the chemical modification of new electrode materials, since only in this way is it possible to achieve detection limits at very low concentrations. Furthermore, the work on novel electrode materials may allow obtaining selective materials for the analytes, which is undoubtedly a significant problem in electroanalysis. I intend to achieve this goal by using not only commercially available substrates as electrode materials but also to obtain my own electrode materials which have not yet been described in the literature. New materials that can be used for detection of analyte without prior functionalization are challenging. Modification with organic compounds by both chemical and electrochemical methods offers a great opportunity to obtain new sensors that can be used for the detection of many important substances.

I am currently conducting two grants in a consortium with the Gdańsk University of Technology (Opus 19 and Sonata Bis 10), the Principal Investigator of which is Jacek Ryl, D.Sc., Professor of the Gdańsk University of Technology. At the same time, I am the Principal Investigator of the consortium partner, University of Gdańsk.

The project Opus 19: " Electrochemical Au-Minecraft: a new approach to the construction of impedance biosensor systems", (No. 2020/37/B/ST7/03262, duration: 26/01/2021-25/01/2024), was based on the previous experiments described in work [H.11.] (DEFB1 gene detection) and [H.12.] (PD-L1 detection), where it was proven that impedance analysis allows obtaining detailed information about adsorbing molecules. Therefore, gold nanocubes (AuNCs) will be incorporated into sensory studies to enhance the molecular bond tracking effect. In this project, I am responsible for the synthesis and functionalization of the (AuNCs), as well as performing part of the electrochemical measurements.

Project Opus 19 involves obtaining (AuNCs) layers on the surface of electrodes and then their functionalization mainly with suitable oligonucleotides or antibodies to detect RNA polymerase (*Escherichia coli*). The obtained (AuNCs) will be also used to create a selective electrode surface for the detection of other biomolecules. The changes induced by the presence of the investigated analyte will be tracked by such techniques as (CV), (EIS), or (DEIS).

Within project Sonata Bis 10: "Technology for additive manufacturing of electroactive spatial structures from diamond-reinforced polylactide composites" (No. 2020/38/E/ST8/00409, duration: 25/03/2021-24/03/2024) the (BDD) nanomaterial will be implemented into 3D printed filaments in order to obtain new electrochemical sensors.

In this project, I am responsible for coordinating electrochemical studies, the synthesis of eutectic solvents (DES), and the electrochemical activation of electrodes based on poly-lactic acid (PLA) and diamond components. The Sonata Bis 10 project has planned to obtain new electrode materials based on (PLA) doped with other materials such as diamond, as well as carbon materials that can be obtained using 3D printing technology. Obtaining the new

filaments will provide relatively inexpensive electrode materials that will mainly serve for the detection of selected non-steroidal anti-inflammatory drugs, neurotransmitters, and antibiotics.

Regardless of the above-mentioned research projects, I also plan to develop electroanalytical methods for electrode functionalization as well as obtain new conductive substrates. Besides continuation of electrochemical research, I plan to improve my skills in the synthesis of bioactive organic compounds.

Currently, I am cooperating with the scientists of many research centers, I am also the supervisor of two master's theses, as well as I am cooperating with Ph.D. students in order to achieve the aforementioned research plans.

4.8. References

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5. Presentation of significant scientific or artistic activity carried out at more than one university, scientific or cultural institution, especially at foreign institutions

- **Before obtaining the doctoral degree (years: 2004-2010)**

My Ph.D. thesis was carried out in the Department of Analytical Chemistry at the University of Gdansk under the supervision of Prof. Tadeusz Ossowski D.Sc. I was mainly focused on developing new methods of synthesis, purification and analysis of organic compounds, trying to obtain mainly molecules that would simultaneously exhibit redox-active and chromophoric properties. In my work, I was mainly focused on the synthesis of crown ethers, amino acid derivatives, peptides, and other organic molecules. Additionally, a very considerable part of my work was focused on the synthesis of 9,10-anthraquinone derivatives by attaching this moiety to the above-mentioned molecules in order to obtain new organic compounds that could be used for molecular recognition of primarily the metal ions, as well as organic compounds, using spectroscopic and electrochemical methods in the monitoring the host-guest interactions. During the realization of my Ph.D. thesis, together with my thesis supervisor I mainly cooperated with Prof. Grzegorz Schroeder, D.Sc., at that time head of the Department of Supramolecular Chemistry, Faculty of Chemistry, Adam Mickiewicz University in Poznan. This collaboration has resulted in two papers in reputed scientific journals. Additionally, despite the above works, I have mainly concentrated on writing chapters of monographs, which were published in books edited by Prof. Grzegorz Schroeder, D.Sc., and Prof. V. I. Rybachenko, D.Sc., concerning supramolecular chemistry and molecular recognition.

- **After obtaining the doctoral degree (years: 2010-2021)**

In the first years after receiving my Ph.D., I continued my research on synthesizing 9,10-anthraquinone derivatives. My first post-doctoral work mainly involved synthesis, electrochemical property studies, and structural studies of the compounds which I obtained. The above publications were realized in cooperation with the team of Prof. Grzegorz Schroeder, D.Sc. The cooperation with Prof. Dr. Grzegorz Schroeder, is still actively continued, within the scope of research on a completely different class of compounds, notably on the study of the properties of Fe_3O_4 derivatives, as was demonstrated in papers [B.27. and B.33.], in which I am a corresponding author.

Then, I had designed a series of compounds containing in their structure 9,10-anthraquinone, modified in such a way that the obtained molecules, not described in the literature before, were characterized by high biological activity. To determine the antiproliferative activity of the obtained structures on various cancer cell lines, including drug-resistant ones, I have established active cooperation with Prof. Joanna Wietrzyk, D.Sc., from the Ludwik Hirszfeld Institute of Immunology and Experimental Therapy of the Polish Academy of Sciences. The cooperation resulted in one scientific paper [B.21.] and 8 national patents, of which the last one was published in 2020. The above collaboration resulted in the Ph.D. thesis of Elżbieta Wnuk, the topic of which was "Synthesis of new 9,10-anthraquinone

derivatives containing heterocyclic amine fragments and investigation of electrochemical, spectroscopic and biological properties of selected derivatives" defended on 12 July 2017, of which I was the associate supervisor.

At the same time, conducting research on the synthesis of organic compounds exhibiting biological activity I began cooperation with Robert Bogdanowicz, D.Sc., Professor of the Gdańsk University of Technology. As a result of this cooperation I have started to develop electrode modification methods, at first mainly (BDD) electrodes and, later on,other electrode materials. As a result of active cooperation, in addition to the works presented in the described scientific achievement, other works in the field of modification and use of electrode materials for sensory purposes have been produced, as well as works that were the result of research projects performed together. From 2015 to 2019, our collaboration was developed within the framework of the Harmonia 6 grant "Diamond-graphene sensory heterostructures" in which we were investigators.

My cooperation with Robert Bogdanowicz and his team is still actively continued to this day and concerns mainly research on physicochemical properties, modifications, and sensory studies of not only new electrode-based materials (BDD) but also modifications of materials based on diamonds and nanodiamonds including (B:CNWs).

At the same time, while I was researching the properties of modified diamond electrodes, I also established cooperation with Mirosław Sawczak, D.SC., assistant professor at the Robert Szewalski Institute of Fluid-Flow Machinery, Polish Academy of Sciences. As part of our recent cooperation, I have obtained a new material, of which the synthesis and characterization have been published in [B.41.].

Within the scientific cooperation, the results of which have been published mainly in the works [H.6. and H.7.], apart from the above-mentioned scientists from the Tricity universities, I established cooperation with Katarzyna Siuzdak, D.Sc., Professor at the Robert Szewalski Institute of Fluid-Flow Machinery of the Polish Academy of Sciences, and Dawid Nidzworski, D.Sc., representative of the SensDx company, which resulted in two national patents and one European patent, as described in detail in Appendix 5.

During my research on modification of electrode surfaces, I have started a scientific collaboration with Mateusz Śmietana, D.Sc., Professor at the Warsaw University of Technology, conducting innovative work on simultaneous detection of analytes on fiber optic sensors using optical and electrochemical techniques.

Working together with Mateusz Śmietana, D.Sc., in the years 2015 - 2019 as a result of the created scientific consortium, we conducted the Sonata Bis 4 grant, no. 2014/14/E/ST7/00104 "Conductive photonic structures for multiparametric biochemical diagnostics", in which I was the principal investigator in the side of the University of Gdańsk. In my research, I was mainly responsible for carrying out and developing electroanalytical methods on fiber optic sensors during sensory measurements. During the realization of the aforementioned grant, the majority of my research was performed in the laboratory of the Warsaw University of Technology, Faculty of Electronics and Information Technology, Institute of Microelectronics and Optoelectronics, in a series of more than 20 one- or several-day work visits, which resulted in several scientific papers. An extremely important fact is, that in the duration of the above grant, I carried out electrochemical investigations on the surfaces of optical fibers that had been modified with the ITO layer, which were obtained in the

laboratory of doc. dr. hab. RNDr. Vítězslav Straňák, Ph.D. at the Institute of Physics and Biophysics, Faculty of Science, University of South Bohemia, in České Budějovice, Czech Republic.

Conducting most of the work described in the present scientific achievement in order to characterize the modified electrodes, I have also initiated cooperation with Jacek Ryl, D.Sc., Professor of the Technical University of Gdansk, a specialist in the field of X-ray photoelectron spectroscopy (XPS) measurements, and measurements by dynamic electrochemical impedance spectroscopy (DEIS). As a result of our cooperation, apart from work [H.11.] concerning the modification and use of electrodes for sensing purposes, our cooperation is also based on other research areas, e.g., investigation of corrosion inhibitors [B.23. and B.25.] or investigation of core-shell materials [B.32. and B.33.].

During the realization of the above-mentioned OPUS 19 grant, an active cooperation between the group of Jacek Ryl, D.Sc., from the Institute of Nanotechnology and Materials Science of the Gdańsk University of Technology and the group headed by Prof. Grzegorz Węgrzyn, from the Faculty of Biology of the University of Gdańsk, and the above-mentioned Mirosław Sawczak, D.Sc., is in progress.

Whereas, during the realization of the Sonata Bis 10 grant, I cooperate with Krzysztof Formela Ph.D., an employee of the Gdańsk University of Technology in the Department of Polymer Technology, a specialist in the field of polymers, to obtain suitable polylactide-based (PLA) filaments.

I actively cooperate with many scientists from different universities and with members of their teams. I conduct cooperative research not only with scientists from my *Alma Mater* but also with scientists from other research centers. I have listed my main collaborators below and detailed the most important results of the collaboration.

- Paulina Kosikowska-Adamus, Ph.D., (Faculty of Chemistry, University of Gdańsk) - this cooperation resulted in patent application number P.439579, “A biosensor for identification and measurement of bacterial endotoxin LPS and a method for obtaining a biosensor for identification and measurement of bacterial endotoxin”.
- Paweł Rochowski, Ph.D., (Faculty of Mathematics, Physics and Computer Science) - physicochemical investigations of active compounds [B.31.].
- prof. Sylwia Rodziewicz-Motowidło, D.Sc., (Wydział Chemii, Uniwersytet Gdański) - investigations of protein detection [H.11.].
- Joanna Chamier-Cieminska, Ph.D., (Department of Forensic Medicine, Faculty of Medicine, Medical University of Gdańsk) - investigations on biological compounds [H.11.].
- Alicja Kuban-Jankowska, D.Sc., (Department of Medical Chemistry, Faculty of Medicine, Medical Univeristy of Gdańsk) - investigations of the activity of biological compounds [B.13., B.16.].
- Dmitry Tretiakow, MD-Ph.D., (Department of Otolaryngology Faculty of Medicine Medical Univeristy of Gdańsk) - human papillomavirus (HPV) protein detection assays.
- prof. Jacek Sein Anand, MD-D.Sc., (Division of Clinical Toxicology Faculty of Health Sciences with the Institute of Maritime and Tropical Medicine Medical University of Gdańsk) - analysis of chemical compounds [H.8., B.33.].

- prof. Krzysztof Łukaszuk, MD-D.Sc., (Division of Obstetric and Gynaecological Nursing Institute of Nursing and Midwifery Medical University of Gdańsk) - research on the detection of human chorionic gonadotropin (HCG).
- prof. Grzegorz Dubin, D.Sc., (Department of Biochemistry, Biophysics and Biotechnology, Jagiellonian University) - investigations on detection of (PD-1) and (PD-L1)) [H.11.].
- prof. Teodor Gotszalk, D.Sc., (Gotszalk (Department of Nanometrology, Faculty of Microsystem Electronics and Photonics, Wrocław University of Science and Technology) - investigations on characteristics of new materials [B.31.].

Awards and scholarships:

1. Award in the competition for the best Ph.D. thesis defended in 2010, organized by the Gdansk Branch of the Polish Chemical Society. (14/06/2011).
2. Third Degree Team Award of the Rector of the University of Gdansk in 2014 - for a series of publications on the characterization of intermolecular interactions in solution and at the interface of the surface of materials and organic compounds. (01/10/2014)
3. Prof. Gotfryd Kupryszewski Award for outstanding scientific achievements for young employees of the Faculty of Chemistry, University of Gdansk. (03/12/2014)
4. First Degree Team Award of the Rector of the University of Gdansk in 2020 - for a series of publications related to the characteristics of intermolecular interactions in solution and at phase boundaries. (01/10/2020)
5. Prof. Leszek Łankiewicz Award for the best master's thesis of interdisciplinary character defended at the Department of Chemistry, University of Gdansk, in 2017 - Natalia Malinowska, M.Sc. - winner, of which I was the supervisor.
6. Scientific scholarship for young doctoral students at the University of Gdańsk in 2012.

6. Presentation of teaching and organizational achievements as well as achievements in popularization of science or art

6.1. Teaching Achievements

- **Before obtaining the doctoral degree (years: 2004-2010)**

I started my didactic work with students as a student of Ph.D. studies at the Faculty of Chemistry, University of Gdansk. As part of my activities in the Department of Analytical Chemistry, I have mainly conducted analytical chemistry laboratories with the second-year chemistry students.

Since 2009, I have started to co-teach (in an equal number of hours) with Prof. Tadeusz Ossowski, a lecture on "Physicochemical methods of research in forensic science" for third-year students of chemistry. I have developed all laboratory activities in the physicochemical part for the needs of the above subject. I am currently conducting this subject with Prof. Tadeusz Ossowski, D.Sc., constantly developing the laboratory activities.

- **After obtaining the doctoral degree (years: 2010-2021)**

After obtaining my Ph.D., I started to teach original lecture called "Molecular Recognition", which I am still conducting. I also co-teach a lecture, "Research Methods in Supramolecular Chemistry", for chemistry students. Moreover, I give a lecture "Scientific methods of crime traces investigation by chemical methods" for the first-year full-time and extramural students of the Faculty of Law and Administration (WPiA) of the University of Gdańsk, specializing in criminology, for whom I also prepared lab experiments from the basis. Another lecture I give to full-time second-year students of criminology (WPiA) is a subject called "Counterfeiting" which I give to other lecturers from the Faculty of Chemistry, UG. Together with other members of the Department of Analytical Chemistry, I give a lecture called "Analytical aspects of intermolecular interactions" to third-year undergraduate chemistry students.

Besides the lectures mentioned above, I teach or have taught several laboratory courses in a wide range of subjects, as shown in the table below (Table 1). In Table 1 I have summarized both the lectures were given and the laboratory courses. The laboratory courses of "Advanced Chemistry Laboratory - Analytical Chemistry" for chemistry students and chemical business students have been mostly developed by me. I also teach "Laboratory of Advanced Chemistry - Analytical Chemistry" courses in English for international Erasmus exchange students.

Table 1. Detailed list of subjects I have taught after obtaining my PhD since 2009/2010.

Subject	Year, faculty, major	Academic Year	Hours per year
Analytical Chemistry, laboratory	2 year, Chemistry, Bachelor degree	from 2009/2010 to 2013/2014	from 60 to 120
Instrumental analysis, laboratory	1 year, Chemistry, 2nd level studies	from 2010/2011 to 2012/2013	from 45 to 60
Master's Seminar, seminars	2 year, Chemistry, 2nd level studies	from 2012/2013 to 2015/2016	from 15 to 30
Surfactants and biosurfactants, laboratory	3 year, Chemistry, Bachelor degree	from 2016/2017 to 2018/2019	from 10 to 15
Molecular Recognition, lecture	1 year, Chemistry, 2nd level studies	from 2010/2011 to currently	30
	2 year, Chemistry, 2nd level studies, part-time studies	from 2016/2017 to currently	18
Physicochemical methods in forensic science, lecture	3 year, Chemistry, Bachelor degree	from 2009/2010 to currently	15
Physicochemical methods in forensic science, laboratory	3 year, Chemistry, Bachelor degree	from 2013/2014 to currently	from 15 to 36
Research methods in supramolecular chemistry, lecture	2 year, Chemistry, 2nd level studies	from 2012/2013 to currently	7,5
	2 year, Chemistry, 2nd level studies	from 2017/2018 to currently	6
Laboratory of Advanced Chemistry - Analytical Chemistry, laboratory	1 year, Chemistry, 2nd level studies	from 2014/2015 to currently	from 45,5 to 104
	1 year, Chemical Business, 2nd level studies	from 2019/2020 to currently	from 6 to 18
	1 year, Chemistry, 2nd level studies, part-time studies	from 2016/2017 to 2017/2018	from 8 to 12
	Erasmus Students	from 2018/2019 to currently	from 8 to 14
Analytical Aspects of Intermolecular Interactions, lecture	3 year, Chemistry, Bachelor degree	from 2018/2019 to currently	from 4 to 5
Scientific methods of crime traces investigation by chemical methods, lecture	1 year, Criminology, 2nd level studies	from 2015/2016 to currently	15
	1 year, Criminology, 2nd level studies, part-time studies		5
Scientific methods of crime traces investigation by chemical methods, laboratory	1 year, Criminology, 2nd level studies		from 20 to 120
	1 year, Criminology, 2nd level studies, part-time studies		from 40 to 17
Counterfeiting, lecture	2 year, Criminology, 2nd level studies	from 2016/2017 to currently	3

In addition, I have completed the academic tutor school course and continue to improve my knowledge within didactics. I have completed, and I am attending the following courses:

1. I completed a 64-hour certification course of the School of Academic Tutors of Collegium Wratislaviense in 2020 as part of the Expert Training in Tutoring implemented from 06/11 to 13/12/2020.
2. Currently, I am participating in a 30-hour didactic course, "Developing didactic skills" organized by the Centre of Didactic Improvement and Tutoring of the University of Gdansk from 07/12/2021 to 28/01/2022

6.2. Scientific supervision - supervising: bachelor's and master's theses, assistant supervisor of doctoral theses

Supervision of:

a) Bachelor theses: 7
Kornelia Kozłowska (2021), Jakub Przesmycki (2020), Izabela Gryczka (2017), Adrian Koterwa (2017), Adam Walkowiak (2016), Agata Seroka (2016), Grzegorz Skowierzak (2015)

b) Master's theses: 16
Agata Fritza (2021), Izabela Wróblewska (2019), Adrian Koterwa (2019), Monika Hamera (2018), Adam Walkowiak (2018), Grzegorz Skowierzak (2017), Natalia Malinowska (2017), Joanna Sadzińska (2016), Dorota Klewicz (2016), Hanna Mielech (2015), Wioleta Białobrzeska (2015), Agnieszka Piątek (2014), Karolina Zajet (2014), Katarzyna Ciak (2013), Katarzyna Marczak (2013).

c) Assistant Supervisor: defended PhD thesis: 1, PhD thesis in progress: 1
Elżbieta Wnuk (2017), Zofia Cebula (currently).

6.3 Popularization of science

I am presenting my research results at scientific conferences and symposia, as mentioned in Appendix 5.

As a part of the popularization of science, I have actively participated in organizing open days of the Faculty of Chemistry, University of Gdansk, and I have given the following popular science lectures:

1. The Academic Summer School in the Center for Environmental Education in Starbienino. Invited speaker "Analytical methods in forensic science - selected issues" 12 – 18/09/2010 Starbieninio.
2. Open Day of the Faculty of Chemistry of University of Gdansk. Co-organizer with the Department of Analytical Chemistry, 1/04/2010, Gdansk.
3. Open Day of the Faculty of Chemistry of University of Gdansk. Invited speaker "Let's follow the bread crumbs - chemical analysis in revealing criminalistics traces", 19/03/2013, Gdansk.
4. Open Day of the Faculty of Chemistry of University of Gdansk. Invited speaker "Fragrant Chemistry - scents of beauty and death", 20/3/2017, Gdansk.
5. Open Day of the Faculty of Chemistry of University of Gdansk. Invited speaker "Selected toxic and poisonous substances as seen through the eyes of a chemist", 20/03/2018, Gdansk.
6. Open Day of the Faculty of Chemistry of University of Gdansk. Co-organizer with the Department of Analytical Chemistry, "Chemist detectives in action", 20/03/2019, Gdansk.
7. Open Day of the Faculty of Chemistry of University of Gdansk. Invited speaker "I'll find you! - chemistry in forensics", 10/03/2020, Gdansk.

I also participated in organizing and conducting workshops for young people as part of the Baltic Science Festival at the Faculty of Chemistry in the following editions:

1. X Baltic Science Festival at the Faculty of Chemistry, University of Gdansk. Co-organization of workshops with Dorota Zarzeczańska, Ph.D., and Anna Wcisło, Ph.D., entitled "Chemical Detectives", 23/05/2012, Gdansk.
2. XI Baltic Science Festival at the Faculty of Chemistry, University of Gdańsk. Co-organization of workshops with Dorota Zarzeczańska, Ph.D., and Iwona Dąbkowska Ph.D., entitled "Chemical Detectives", 24/05/2013, Gdansk.
3. XII Baltic Science Festival at the Faculty of Chemistry, University of Gdańsk. Co-organization of workshops with Dorota Zarzeczańska, Ph.D., entitled "Chemical Detectives", 23/05/2014, Gdansk.
4. XIII Baltic Science Festival at the Faculty of Chemistry, University of Gdańsk. Co-organization of workshops with Dorota Zarzeczańska, Ph.D., entitled "Chemical Detectives", 22/05/2015, Gdansk.
5. XIV Baltic Science Festival at the Faculty of Chemistry, University of Gdańsk. Baltic Science Festival at the Faculty of Chemistry, University of Gdańsk. Co-organization of workshops with Dorota Zarzeczańska, Ph.D., entitled "Detectives on the trail of environmental destroyers", 26/05/2017, Gdansk.

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(Applicant’s signature)