VILNIUS UNIVERSITY

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COMPUTATIONAL MODELLING OF BIOSENSORS OF COMPLEX GEOMETRY

Summary of doctoral dissertation
Physical sciences, informatics (09 P)

Vilnius, 2011

Doctoral dissertation was prepared in 2006–2010 at Vilnius University.

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The dissertation will be defended at the public meeting of the Council of Scientific Field of Informatics in the Vilnius University Digital Science and Computing Center on the 30$^{\text{th}}$ of June, 2011, at 2 p.m.

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The summary of the doctoral dissertation was distributed on the ___ of May, 2011. The doctoral dissertation is available at the library of Vilnius University.

Doctaral dissertation preparation was funded by the European Social Fund under Measure VP1-3.1-ŠMM-07-K "Support to Research of Scientists and Other Researchers (Global Grant)", Project "Developing computational techniques, algorithms and tools for efficient simulation and optimization of biosensors of complex geometry".

VILNIAUS UNIVERSITETAS

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KOMPIUTERINIS SUDĖTINĖS GEOMETRIJOS BIOJUTIKLIŲ MODELIAVIMAS

Daktaro disertacijos santrauka
Fiziniai mokslai, informatika (09 P)

Vilnius, 2011

Disertacija rengta 2006–2010 metais Vilniaus universitete.

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Disertacija bus ginama viešame Informatikos mokslo krypties tarybos posėdyje 2011 m. birželio mėn. 30 d. 14 val. Vilniaus universiteto Skaitmeninių tyrimų ir skaičiavimo centre.

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Disertacijos santrauka išsiuntinėta 2011 m. gegužės mėn. ___ d.
Disertaciją galima peržiūrėti Vilniaus universiteto bibliotekoje.

Disertacija parengta įgyvendinant projektą „Kompiuterinių metodų, algoritmų ir įrankių efektyviam sudėtingos geometrijos biojutiklių modeliavimui ir optimizavimui sukūrimas“, finansuojamą iš ES Socialinio fondo pagal VP1-3.1-ŠMM-07-K priemonę „Parama mokslininkų ir kitų tyrėjų mokslinei veiklai (Visuotinė dotacija)“ lėšų.

Introduction

Field of the research and relevance of the problem

Biosensors are analytical devices mainly used to detect analytes and measure their concentrations. Main parts composing a biosensor are a biologically active element, usually an enzyme, that recognizes specific analyte and a transducer, that converts biological recognition event into an electrical signal. This electrical signal is then amplified, processed and presented to the user of the biosensor [1–3].

Amperometric biosensors are relatively cheap, highly sensitive and reliable devices, widely used in various fields, especially in clinical diagnostics, drug detection, food analysis and environment monitoring [4–7]. According to a research made by "Global Industry Analysts Inc.", global market for biosensors was $8.2 billion in 2009, and an average annual growth rate of 6.3% is expected in the future [8].

A lot of experiments are needed when developing and calibrating new biosensors. Enzymes are usually expensive materials, and therefore number of experiments, performed during development of new biosensors should be minimized. Mathematical modelling is widely used for optimizing and analyzing operation of biosensors, thus helping to reduce number of physical experiments required during the development of new biosensors [9, 10]. The processes governing operation of a biosensor are usually described by nonlinear partial differential equations [11, 12].

Analytical solutions for the equations, describing behavior of a biosensor, are known for some particular cases only. Numerical methods are usually employed, when investigating a behavior of biosensors with a complex structure in wide ranges of parameter values. In this research the method of finite differences and the method of alternating directions are employed [13].

Biosensors are developed using transducers operating on different principles [14]. Among the most widely used are biosensors, based on optical [15] and amperometric [16] transducers. An operation of optical biosensors is based on light emission or absorption. An operation of amperometric biosensors is based on electrochemical reaction. Electrons emitted in the electrochemical reaction are collected by an electrode forming a current, considered as an output of the biosensor.

Various biologically active materials are employed when developing biosensors [17]. These materials are used to recognize particular substances, the biosensor is targeted for. Multistage reactions involving enzymes are often used in biosensors. Kinetics of simplest enzymatic reactions is described by Michaelis-Menten equations, while more complex equations are employed, when the operation of the biosensor is based on more complex reactions.

Practical biosensors have complex structure. They are developed using materials with different properties, usually tied together forming multilayered structure. Porous materials, perforated and selective membranes as well as carbon nanotubes [18] are widely used when developing new biosensors. Carbon nanotubes are widely used in the industry, because of its unique properties. Recently, carbon nanotubes started to be used for development of high sensitivity biosensors [3, 14, 19–24].

The simplest amperometric biosensor is composed of an electrode and an enzyme, tied on its surface. In order to simulate operation of such biosensor it is enough to consider a segment representing its section in one point of its surface. In the model of such biosensor the reaction-diffusion equations describe processes in all the segment equally. When modelling more complex biosensors, the two-dimensional space is often needed to define the model, and the model is usually composed of several

areas, where the processes are modelled by different equations.

Usually, when modelling biosensors, for each biosensor of unique structure unique mathematical model is build. The mathematical model is then approximated using numerical methods and implemented as a program, solving system of equations of the approximated model [11]. The construction of mathematical and numerical models is a task, requiring a time and a thoughtfulness. Automated construction of such models as well as programs, implementing them, can simplify these tasks and make modelling of biosensors simpler and more resistant to mistakes.

Object of the research

The object of this research is mathematical and computational models describing operation of biosensors made of several parts with different properties. The dissertation covers models, formulated in one and two-dimensional spaces by partial differential equations with non-linear terms, and solved numerically, using the method of finite differences. The numerical models were implemented by a computer program.

Objectives and tasks of the research

The objective of this research is to automate construction of computational models for biosensors, that can be modelled using partial differential equations, in one or two-dimensional space. Then investigate properties of particular biosensors by performing computer aided modelling. The following tasks were solved to achieve the objective.

1. Propose an approach of modelling enzyme-loaded carbon nanotube electrode, used in development of biosensors and define a model for a biosensor with carbon nanotube electrode and perforated membrane.
2. Investigate approaches of modelling a perforated membrane, used when developing biosensors, and conditions for applying them.
3. Generalize the structure of models, formulated in one-dimensional or two-dimensional space, for biosensors constructed of several parts with different properties.
4. Propose a language allowing to define models of biosensors in domain specific terms, and develop algorithms as well as a computer software simulating operation of biosensors using models formulated in one or two-dimensional space.
5. By applying the developed software, investigate an adequateness of the proposed models and an influence of structural and geometrical properties on behavior of the modelled biosensors.

Research methods

The models considered in the dissertation were formulated using nonlinear partial differential equations. The method of finite differences and the method of alternating directions were applied when approximating the equation systems of the models. The computer aided simulation was used for investigation of properties of the considered biosensors.

Development of computer software, model construction automation and simulation of biosensors were based on analysis of published models by categorizing structural parts of the models and the ways of connecting them.

Results and scientific innovation of the work

1. The mathematical model for the biosensor with a carbon nanotube electrode was developed. Adequateness of the model was investigated using computer aided simulations.

2. The conditions at which the one-dimensional mathematical model can be used instead of two-dimensional one for accurate prediction of the biosensor response were defined. The procedure for determining averaged diffusion coefficient was proposed.

3. Elements, used to build models of biosensors with a complex structure were defined.

4. The biosensor description language was proposed and the computer software, simulating an operation of biosensors in the one-dimensional space and a rectangular domain of the two-dimensional space, was developed.

5. The impact of structural and geometrical properties on a response of the biosensor with a carbon nanotube electrode was investigated, performing computer experiments using the developed software.

Practical application of the results

The model for the biosensor with a carbon nanotube electrode allows one to investigate properties of the biosensor and processes taking place inside it. The proposed way of modelling the carbon nanotube electrode can be used to model other biosensors, incorporating carbon nanotube electrodes.

Conditions were defined for using one-dimensional models to simulate biosensors with perforated membranes precisely. In a lot of cases, application of one-dimensional models allows to perform simulation of such biosensors more effectively, than using two-dimensional models.

The developed software can be used for a computer aided investigation of a biosensor behavior and its properties. By using this software, models can be formulated more easily for biosensors with a complex structure. This allows to simplify an investigation of the structure impact on the operation of a biosensor. The software can also be effectively used when formulating new biosensor models, enabling easier validation the model.

Properties of biosensors, determined perforing computer aided experiments, can be used to develop more effective biosensors.

The dissertation results have been used in an implementation the following projects: "Development of bioelectrocatalysis for synthesis and analysis (BIOSA)" funded by a grant (No. PBT-04/2010) from the Research Council of Lithuania and "Developing computational techniques, algorithms and tools for efficient simulation and optimization of biosensors of complex geometry" funded by the European Social Fund under Measure VP1-3.1-ŠMM-07-K "Support to Research of Scientists and Other Researchers (Global Grant)".

Defended statements

1. Biosensors with carbon nanotube electrodes can be modelled at a macroscopic level, treating the nanotube region as a homogeneous one.

2. One-dimensional models can be used for precise simulation of biosensors with an outer perforated membrane, but not in all the cases.

3. When considering geometrical and structural properties of the biosensors, models, formulated in a rectangular domain of the two-dimensional space, can be generalized and elements composing such models can be defined.

4. The operation of the biosensor with a carbon nanotube electrode is influenced noticeably by the structural properties of the carbon nanotubes.

1. Computer aided modelling of biosensors

When developing mathematical models, biosensors are usually treated as reaction-diffusion systems [11, 12]. Models are usually formulated on the macroscopic level, considering concentrations of the substances, mass transport by diffusion and reaction rates. Such models are described by partial differential equations. These equations are usually nonlinear, therefore analytical solutions are known for special cases only. For simulation of biosensors in wide ranges of its parameter values, numerical methods are used [13].

A lot of calculations are needed to get precise results, when simulations are performed using numerical methods. Moreover, large amount of computer aided experiments has to be carried out when investigating an impact of parameters on a response of the biosensor. To make the computer aided investigations of the biosensors more effective, the models are usually simplified. The models are commonly formulated in one or two-dimensional spaces, and numerical approximations are done in such way, that the systems of equations could be solved effectively [11, 12]. In development of mathematical models for biosensors, homogenization process is widely used [25], allowing to tread periodic mediums as homogeneous ones. The process allows to build simpler models, in some cases even number of space dimensions for domain of the model can be reduced.

There are no widely used tools for simulating action of biosensors. Processes, considered in models of biologic cells are treated similarly to those, taking place in the biosensors and are modelled as reaction-diffusion systems. There are a lot of computer software solutions for modelling processes taking place in biologic cells. Even if processes considered in models of biologic cells are similar to those in biosensors, application of cell modelling software for modelling biosensors is limited, because most of them are based on stochastic methods, that are not effective when modelling biosensors. Some of those solutions, based on partial differential equations, can be used to simulate biosensors [26]. Nevertheless, the use of such software is complicated when modelling biosensors, because they are quite generalized and don't take into account optimizations, that are possible when considering simulation of biosensors only.

Several languages were developed to describe models of biological systems. Today, most widely used are SBML and CellML [27]. These languages allows one to describe structure of a system as well as network of reactions, taking place in it. Nevertheless, the use of these languages for defining models of biosensors is limited, mostly because the structures used for describing geometrical properties of a system are absent.

2. Mathematical models for biosensors with CNT electrode and perforated membrane

2.1. Biosensor with carbon nanotube electrode

A structure of the biosensor with a carbon nanotube electrode and an outer perforated membrane [28] is schematically shown in figure 1.

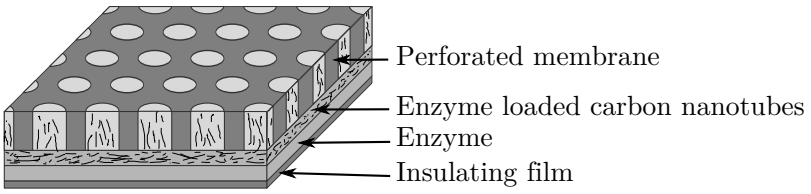

Figure 1. Principal structure of the biosensor with a carbon nanotube electrode and an outer perforated membrane. Figure is not to scale.

The considered biosensor is composed of four layers with different properties and sizes. The base for other mediums is insulating film, shown as a bottom layer in figure 1. When developing the biosensor, carbon nanotubes are tied to a perforated membrane and loaded by an enzyme. Part of the enzyme is left on the surface of the carbon nanotube layer. This part of the enzyme is shown in the figure as the layer between the insulating film and carbon nanotubes. Carbon nanotubes act as an electrode in this biosensor. Due to the technology, used to develop the biosensor, some of the carbon nanotubes are sinked into the holes of the perforated membrane. Nevertheless, the enzyme does not fill these holes.

Two stage enzymatic reaction takes place in the layer of the enzyme. Catalyzed by the enzyme (E), this reaction converts a substrate (S) to a product (P) in presence of a mediator (M),

$$\mathrm{E_{ox}} + \mathrm{S} \xrightarrow{k_1} \mathrm{E_{red}} + \mathrm{P}, \tag{1}$$

$$\mathrm{E_{red}} + \mathrm{M_{ox}} \xrightarrow{k_2} \mathrm{E_{ox}} + \mathrm{M_{red}}, \tag{2}$$

where $\mathrm{E_{ox}}$ and $\mathrm{E_{red}}$ stand for an enzyme respectively in the oxidized and reduced forms and $\mathrm{M_{ox}}$ and $\mathrm{M_{red}}$ represent the mediator in the oxidized and reduced forms respectively. In the layer of the carbon nanotubes the reduced mediator $\mathrm{M_{red}}$ is re-oxidized in the electrochemical reaction

$$\mathrm{M_{red}} \xrightarrow{k_3} \mathrm{M_{ox}} + n_e\,\mathrm{e}^- . \tag{3}$$

Electrons emitted in this reaction form a current, that is considered as an output of the biosensor. Electrochemical reactions are usually treated as very fast ones, in modelling of amperometric biosensors [1, 2].

When formulating models for biosensors with perforated membranes it is usual to consider one unit cell, covering one hole of the membrane and its environment, and formulate the model in the r–z plane of the cylindrical coordinate system. A profile of the unit cell of the biosensor with carbon nanotube electrode is shown in figure 2.

In figure 2, r_1 stands for a radius of the hole of the membrane, and r_2 stands for a radius of the entire unit cell. Areas Ω_1 and Ω_2 correspond to the enzyme layer and the carbon nanotube electrode, Ω_3 stands for a hole of the perforated membrane,

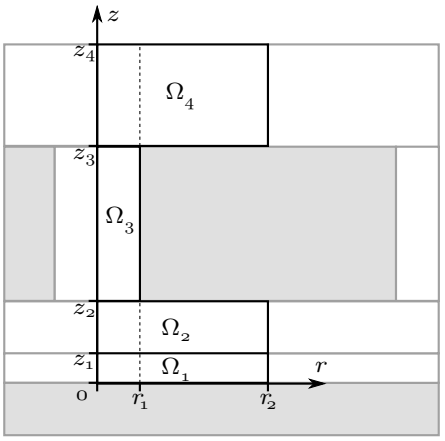

Figure 2. Profile of the unit cell of the biosensor. The figure is not to scale.

and Ω_4 — for a Nernst diffusion layer. Let the open regions Ω_i, $i = 1,2,3,4$, shown in figure 2 are defined as follows:

$$\begin{aligned} \Omega_1 &\equiv (0, r_2) \times (0, z_1), \quad \Omega_2 \equiv (0, r_2) \times (z_1, z_2), \\ \Omega_3 &\equiv (0, r_1) \times (z_2, z_3), \quad \Omega_4 \equiv (0, r_2) \times (z_3, z_4). \end{aligned} \tag{4}$$

Carbon nanotubes are aligned uniformly in the biosensor. Alignment of the nanotubes influence the diffusion of substances along the nanotubes and across them differently [29, 30]. This property is modelled by using different diffusion coefficients for different directions,

$$\Lambda(U) \equiv D_{U,r} \frac{1}{r} \frac{\partial}{\partial r} \left(r \frac{\partial U}{\partial r} \right) + D_{U,z} \frac{\partial^2 U}{\partial z^2}, \tag{5}$$

where $\Lambda(U)$ is an operator describing diffusion of the substance U, U is a concentration of the substance, $D_{U,r}$ and $D_{U,z}$ are diffusion coefficients along the axes r and z correspondingly.

The enzyme in the biosensor is immobilised and therefore is not affected by the diffusion. In the enzyme-loaded regions Ω_1 and Ω_2, the dynamics of the enzyme concentration is affected only by the enzymatic reactions (1) and (2). The dynamics of the enzyme concentration is described by the following reaction equations:

$$\begin{aligned} \frac{\partial E_{ox,i}}{\partial t} &= -k_1 E_{ox,i} S_i + k_2 M_{ox,i} E_{red,i}, \\ \frac{\partial E_{red,i}}{\partial t} &= k_1 E_{ox,i} S_i - k_2 M_{ox,i} E_{red,i}, \quad (r, z) \in \Omega_i,\ i = 1,2,\ t > 0, \end{aligned} \tag{6}$$

were t stands for time, $U_i = U_i(r, z, t)$, $U \in \{E_{ox}, E_{red}, S, M_{ox}\}$ is the concentration of the corresponding species in the closed area $\overline{\Omega}_i$, coefficients k_1 and k_2 are the rates of the reactions (1) and (2), respectively.

The mass transport by diffusion of the substrate takes place in all the regions: the enzyme layer (Ω_1), the enzyme-loaded carbon nanotube layer (Ω_2), the holes of the perforated membrane (Ω_3) and the Nernst diffusion layer (Ω_4). In both layers containing the enzyme, the substrate also participates in the reaction (1). The dynamics of the substrate concentration S_i in the region Ω_i is described as follows

$(i = 1, 2, 3, 4)$:

$$\frac{\partial S_i}{\partial t} = \begin{cases} \Lambda(S_i) - k_1 E_{ox,i} S_i, & \text{for } i = 1,2, \\ \Lambda(S_i), & \text{for } i = 3,4. \end{cases}, \quad (r, z) \in \Omega_i,\ t > 0. \tag{7}$$

The mass transport by diffusion of the oxidised mediator M_{ox} takes place inside entire biosensor as well as in the outer diffusion layer. In both layers containing the enzyme, the mediator is involved in the enzymatic reaction (2). In the carbon nanotube regions, the oxidised mediator is regenerated by the electrochemical reaction (3). Consequently, the dynamics of the M_{ox} in the region Ω_i is described by the following equations $(t > 0)$:

$$\frac{\partial M_{ox,i}}{\partial t} = \begin{cases} \Lambda(M_{ox,i}) - k_2 M_{ox,i} E_{red,i}\,, & \text{for } i = 1, \\ \Lambda(M_{ox,i}) - k_2 M_{ox,i} E_{red,i} + k_3 M_{red,i}\,, & \text{for } i = 2, \\ \Lambda(M_{ox,i}), & \text{for } i = 3, 4, \end{cases} . \tag{8}$$

The reduced mediator (M_{red}) is generated in the areas filled with the enzyme, in reaction (2). The reduced mediator is generated but is not consumed in the layer of the enzyme, therefore it accumulates and is transported by diffusion. The M_{red} is re-oxidized in the electrochemical reaction (3), in the layer of the carbon nanotubes. The dynamics of M_{red} is described by the following equations $(t > 0)$:

$$\frac{\partial M_{red,i}}{\partial t} = \begin{cases} \Lambda(M_{red,i}) + k_2 M_{ox,i} E_{red,i}\,, & \text{for } i = 1, \\ \Lambda(M_{red,i}) + k_2 M_{ox,i} E_{red,i} - k_3 M_{red,i}\,, & \text{for } i = 2, \\ \Lambda(M_{red,i}), & \text{for } i = 3, 4, \end{cases} , \tag{9}$$

where $M_{red,i} = M_{red,i}(r, z, t)$ is a concentration of the reduced mediator in the area Ω_i, i = 1,2,3,4.

During the modelling experiments the buffer solution is well stirred. This leads to constant concentrations of the species above the Nernst diffusion layer [31],

$$S_4(r, z_4, t) = S_0,\ \ M_{ox,4}(r, z_4, t) = M_0,\ \ M_{red,4}(r, z_4, t) = 0,\ \ t > 0,\, r \in [0, r_2], \tag{10}$$

where S_0 and M_0 are the concentrations of the substrate and the mediator in the buffer solution. The non-leakage boundary conditions are used to represent the symmetry of the unit cell of the biosensor. These conditions are also applied to the surface of the perforated membrane and the insulating film. On the boundary between adjacent regions having different diffusivities, the matching conditions are applied.

The biosensor operation starts $(t = 0)$ when the substrate S and the mediator M_{ox} are infused into the buffer solution. At this time the mediator and the substrate is absent in the biosensor, and only appears on the external boundary of the Nernst diffusion layer,

$$S_4|_{\Gamma_4} = S_0, \quad M_{ox,4}|_{\Gamma_4} = M_0. \tag{11}$$

Initially, the whole enzyme is in the oxidised form and is assumed to be homoge-

neously distributed in the enzyme layer ($t = 0$),

$$E_{ox,1} = E_0,\ (r,z) \in \Omega_1, \quad E_{red,i} = 0,\ (r,z) \in \Omega_i,\ i = 1, 2, \tag{12}$$

where E_0 stands for the initial concentration of the enzyme. It was assumed that the carbon nanotube mesh homogeneously saturates with the enzyme at a concentration possibly different from that in the enzyme layer,

$$E_{ox,2} = \eta E_0,\ (r,z) \in \Omega_2,\ t = 0, \tag{13}$$

where η is the ratio the enzyme initial concentration in the enzyme layer Ω_1 to that in the carbon nanotube layer Ω_2, practically, $0 \leq \eta < 1$ [28].

The response of the biosensor is generated due to the electrochemical reaction (3). The electrons released in this reaction form a current that is amplified and presented to the end-user. The electrochemical reaction (3) takes place in the layer of the enzyme-loaded carbon nanotubes and is present in all the volume of the layer. The current density generated in the layer of carbon nanotubes at a steady state is defined as

$$J = \lim_{t\to\infty} j(t), \quad j(t) = \frac{2n_e F k_3}{r_2^2} \int_{z_1}^{z_2} \int_0^{r_2} M_{red,2} r\, dr\, dz\,, \tag{14}$$

where n_e is a number of electrons released in the mediator re-oxidation, F is the Faraday constant, and $j(t)$ is a current density, generated by the biosensor at time t.

For the simplicity, the species S, M_{ox} and M_{red} are assumed to have the same diffusion properties, i.e. the diffusion coefficients of all these species are assumed to be the same for a certain medium. The enzyme and the Nernst layers are assumed to be homogeneous,

$$\begin{aligned} D_{U_1,r} &= D_{U_1,z} = D_e, \quad U_1 \in \{S_1, M_{ox,1}, M_{red,1}\}, \\ D_{U_4,r} &= D_{U_4,z} = D_n, \quad U_4 \in \{S_4, M_{ox,4}, M_{red,4}\}, \end{aligned} \tag{15}$$

where D_n stands for the diffusion coefficient of the substrate and the mediator in the buffer solution as well as in the Nernst diffusion layer, and D_e is the corresponding diffusion coefficient in the enzyme.

Due to a non-homogeneous distribution of the carbon nanotubes, the effective diffusion coefficients for the regions Ω_2 and Ω_3 can be obtained by applying the homogenization process [25, 30]. The enzyme-loaded carbon nanotube layer (Ω_2) can be considered as a three compartment domain: the enzyme, the carbon nanotubes and the buffer solution. The holes of the perforated membrane (Ω_3) consist of two compartments: the carbon nanotubes and the buffer solution. The effective diffusion coefficients for these two mediums can be expressed in terms of the volume fraction, the tortuosity and the diffusion coefficients for the consisting compartments [25, 30, 32]. Assuming small diffusion coefficient of the species in the carbon nanotubes as well as low volume fraction of the mesh of carbon nanotubes [33], the effective diffusion coefficients can be expressed as

$$\begin{aligned} D_{U_2,\zeta} &= \theta_{2,\zeta}(\eta D_e + (1-\eta) D_n), \quad U_2 \in \{S_2, M_{ox,2}\}, \\ D_{U_3,\zeta} &= \theta_{3,\zeta} D_n, \quad U_3 \in \{S_3, M_{ox,3}\}, \quad \zeta \in \{r, z\}, \end{aligned} \tag{16}$$

where $\theta_{i,r}$, $\theta_{i,z}$ are the tortuosities of the medium in r and z directions, respectively, $i = 2, 3$. Values of the tortuosities $\theta_{i,r}$ and $\theta_{i,z}$ depend on the structural anisotropy

of the carbon nanotubes, particularly on the nanotubes orientation [30].

When modelling amperometric biosensors, electrochemical reactions are usually assumed to have very high reaction rate coefficient and consuming all available reagents immediately. In the case of the biosensor with carbon nanotube electrode, assuming the electrochemical reaction (3) to be very fast, the model can be simplified. As the reaction is treated as very fast one, its rate fully depends on the concentration of M_{red}, which is generated as a product of the reaction (2). Consequently, in the region Ω_2 the rate of the electrochemical reaction (3) is equal to the rate of the enzymatic reaction (2). This allows to simplify the proposed model by replacing (8) with the following equation, when $t > 0$:

$$\frac{\partial M_{ox,i}}{\partial t} = \begin{cases} \Lambda(M_{ox,i}) - k_2 M_{ox,i} E_{red,i}\,, & \text{for } i = 1, \\ \Lambda(M_{ox,i}), & \text{for } i = 2,3,4, \end{cases}, \quad (r,z) \in \Omega_i. \tag{17}$$

and (9) by

$$\frac{\partial M_{red,1}}{\partial t} = \Lambda(M_{red,1}) + k_2 M_{ox,1} E_{red,1}, \quad (r,z) \in \Omega_1,\, t > 0. \tag{18}$$

The boundary conditions should be modified accordingly. On the boundary, where the enzyme-loaded carbon nanotube layer (Ω_2) touches the entirely enzyme-loaded layer (Ω_1), the concentration of M_{ox} is influenced by the reaction (3). The mediator re-oxidation reaction (3) is considered to be so fast, that whole diffusive M_{red}, touching the border between enzyme loaded regions, is immediately re-oxidised,

$$M_{red,1}|_{\Gamma_1} = 0, \tag{19}$$

$$D_{M_{ox,2},z} \left.\frac{\partial M_{ox,2}}{\partial z}\right|_{\Gamma_1} = D_{M_{ox,1},z} \left.\frac{\partial M_{ox,1}}{\partial z}\right|_{\Gamma_1} + D_{M_{red,1},z} \left.\frac{\partial M_{red,1}}{\partial z}\right|_{\Gamma_1}, \tag{20}$$

$$M_{ox,1}|_{\Gamma_1} = M_{ox,2}|_{\Gamma_1}\,.$$

The response of the biosensor is generated because of the electrochemical reaction (3). As the reaction is assumed to be very fast, its rate fully depends on the concentration of M_{red}, which is generated as a product of the reaction (2). Consequently, in the region Ω_2 the rate of the electrochemical reaction (3) is equal to the rate of the enzymatic reaction (2). M_{red} is also generated in the enzyme layer (Ω_1). Differently from the region Ω_2, here the mediator is not re-oxidised. The M_{red} produced in Ω_1 is consumed on the boundary with the layer of carbon nanotubes. The total current density generated by the biosensor, assuming reaction (3) to be very fast, is

$$j(t) = \frac{2n_e F}{r_2^2} \int_0^{r_2} \left(k_2 \int_{z_1}^{z_2} E_{red,2} M_{ox,2} dz - D_{M_{red,1},z} \left.\frac{\partial M_{red,1}}{\partial z}\right|_{\Gamma_1} \right) r\, dr\,. \tag{21}$$

2.2. One-dimensional model for biosensor with a perforated membrane

Two-dimensional models of biosensors require much more computations than corresponding one-dimensional models. Though the one-dimensional models are more effective in computations, they describe structure of a biosensor not as precise, as two-dimensional models do. Particular amperometric biosensor was considered to

investigate conditions, where one-dimensional models can be used instead of two-dimensional models to simulate biosensors with perforated membranes. The considered biosensor is composed of several layers. The electrode is covered by a selective membrane, on which the enzyme is immobilized. The biosensor is then covered with a perforated membrane. When defining a mathematical model for the biosensor, it was assumed, that the enzyme can partially fill the holes of the perforated membrane. The structure of the considered biosensor is shown in figure 3, on the left side.

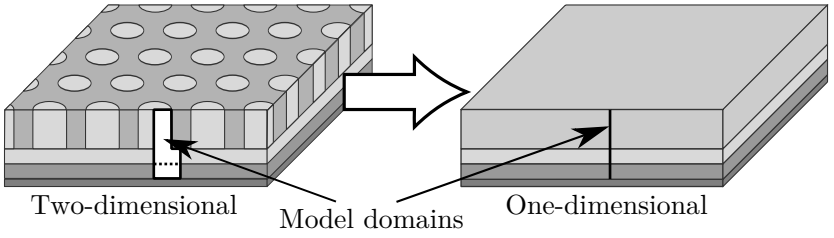

Figure 3. Principal structure of the biosensor with a perforated membrane (on the left), and the structure of the same biosensor (on the right), considering the perforated membrane as a homogeneous medium. The figure is not to scale.

Mathematical models formulated in one and two-dimensional spaces are known [34, 35] for the biosensor shown in figure 3. An enzymatic reaction, appearing in the layer of the enzyme of this biosensor, is described by the Michaelis-Menten kinetics and is schematically shown as

$$\mathrm{E} + \mathrm{S} \rightleftarrows \mathrm{E}\,\mathrm{S} \rightarrow \mathrm{E} + \mathrm{P}, \tag{22}$$

where S is a substrate and P is a product of the reaction, E stands for the enzyme and E S is an enzyme-substrate complex. The product of the reaction is participating in the electrochemical reaction on the surface of the electrode, emiting electrons forming an output current of the biosensor.

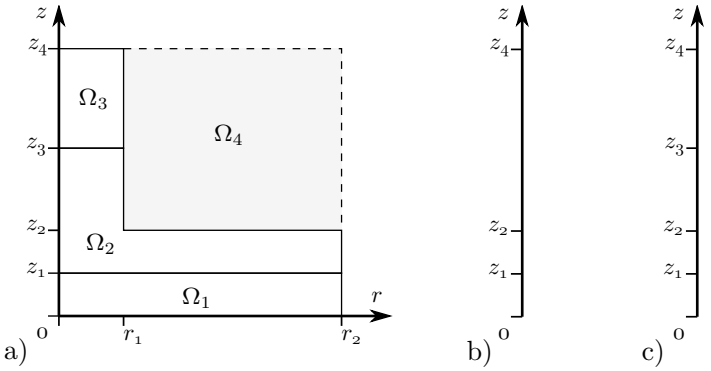

Figure 4. A profile of the unit cell of the biosensor in the two-dimensional model (a), and a structure of the corresponding one-dimensional models (b, c). One-dimensional model can be formulated considering parts of the perforated membrane with and without the enzyme as separate layers (c) or treating them as one homogeneous layer (b).

The model of the biosensor is formulated in the r-z plane of the cylindrical coordinate system, when modelling the biosensor in the two-dimensional space. As shown in figure 4, the model is composed of three areas: Ω_1 stands for the selective membrane, Ω_2 — the enzyme layer, partially filled holes of the perforated membrane, and Ω_3 — part of the hole, filled with the buffer solution, where the enzyme is ab-

sent. Areas Ω_2 and Ω_3 model processes taking place in the perforated membrane. In the enzyme region Ω_2, the enzymatic reaction and the diffusion of the substrate and the product take place. The dynamics of the concentrations is described by the reaction-diffusion equations. In the region Ω_3, the mass transport of both species by diffusion takes place. Governing equations, modelling processes in regions Ω_2 and Ω_3 are the following:

$$\begin{aligned} &\frac{\partial S_2}{\partial t} = D_2\Delta_{2\mathrm{d}}S_2 - R(S_2), \quad \frac{\partial P_2}{\partial t} = D_2\Delta_{2\mathrm{d}}P_2 + R(S_2), \quad (r,z)\in\Omega_2, \\ &\frac{\partial S_3}{\partial t} = D_3\Delta_{2\mathrm{d}}S_3, \qquad\qquad \frac{\partial P_3}{\partial t} = D_3\Delta_{2\mathrm{d}}P_3, \qquad\qquad (r,z)\in\Omega_3,\ t>0, \end{aligned} \tag{23}$$

where t stands for time, $S_i = S_i(r,z,t)$ and $P_i = P_i(r,z,t)$ are the substrate and product concentrations in closed region $\overline{\Omega}_i$, $i = 2,3$, D_2 and D_3 are diffusion coefficients of the substrate and the product in the corresponding medium, $\Delta_{2\mathrm{d}}$ is the Laplace operator for the r-z plane of the cylindrical coordinate system.

Assuming the perforated membrane to be the periodic medium, the homogenization process can be applied to it [25]. According to this approach, the perforated membrane is replaced by a homogeneous medium with the properties similar to the properties of the perforated membrane. This makes description of the biosensor operation in the one-dimensional space possible [34, 36]. Since the perforated membrane is a non-homogeneous media, a homogenization process have to be applied to it [25, 34]. The dynamics of the concentrations of the substrate and product in the homogenized perforated membrane is described as follows ($t>0$):

$$\frac{\partial S_3^*}{\partial t} = D_3^*\Delta_{1\mathrm{d}}S_3^* - \gamma R(S_3^*), \quad \frac{\partial P_3^*}{\partial t} = D_3^*\Delta_{1\mathrm{d}}P_3^* + \gamma R(S_3^*), \quad z\in(z_2,z_4), \tag{24}$$

where $S_3^* = S_3^*(z,t)$ and $P_3^* = P_3^*(z,t)$ are the concentrations of the substrate and the product in the layer $[z_2,z_4]$, D_3^* is the effective diffusion coefficient of the substrate and product in the homogenized perforated membrane, γ is the correction coefficient for the rate of the enzymatic reaction.

According to the volume averaging approach [30], the correction coefficient γ for the reaction rate can be calculated as the volume fraction of the enzyme in the entire perforated membrane,

$$\gamma = \alpha\beta, \qquad \alpha = \frac{\pi r_1^2}{\pi r_2^2} = \frac{r_1^2}{r_2^2}, \quad \beta = \frac{z_3 - z_2}{z_4 - z_2}, \tag{25}$$

where α stands for a perforation level, and β is a level of filling the holes with the enzyme.

One of the most general restrictions for the effective diffusivity D_3^* can be expressed as follows:

$$0 \le D_3^* \le \max(D_2, D_3)\,. \tag{26}$$

The more precise evaluation of the diffusivity D_3^* should take into consideration the geometry of the membrane perforation. The volume averaging approach can be also applied to evaluate the effective diffusivity D_3^* [25, 30].

In the case when the material is a two-phase composite, the effective diffusion coefficient d^* is considered as a function of the constituent diffusion coefficients (d_1

and d_2) and the volume fraction (υ) [37, 38],

$$\frac{d_1 d_2}{\upsilon d_2 + (1-\upsilon) d_1} \leq d^* \leq \upsilon d_1 + (1-\upsilon) d_2 \,. \tag{27}$$

where d_i is the diffusion coefficient of the species in a phase i, $i = 1, 2$, and υ is the volume fraction of the species in the phase 1. Accordingly, the volume fraction of the species 2 equals $(1 - \upsilon)$. The effective diffusion coefficient d^* in a two-phase composite can be also evaluated by it's upper bound given in (27) and the tortuosity factor θ ($0 \leq \theta \leq 1$) [39, 40],

$$d^* = \theta(\upsilon d_1 + (1-\upsilon) d_2) \,. \tag{28}$$

Very similar approach to the effective diffusion coefficient was applied in modelling of glucose diffusion through an isolated pancreatic islet of Langerhans [32].

When modelling holes of the perforated membrane by right cylinders, the tortuosity of the holes equals approximately to unity, $\theta \approx 1$. Assuming zero diffusivity of both species in the insulator region Ω_4 and the unity tortuosity of holes, the formula (28) was applied to the entire perforated membrane to calculate the effective diffusion coefficient D_3^*,

$$D_3^* = \alpha \left(\beta D_2 + (1-\beta) D_3\right) . \tag{29}$$

Although, the volume averaging approach is widely used, there are several known cases, where the approach provides incorrect results. The case of an impermeable aggregate is among them [41]. In such case, more precise modelling of a reaction-diffusion system requires additional parameters [42]. On the other hand, the two-dimensional model taking into consideration the geometry of the membrane perforation requires no correction coefficients.

3. Generalized modelling of biosensors with complex geometry

In order to automate the development of computational models for biosensors, a computer software, that is able to simulate biosensors modelled in the one or two-dimensional space, was created. As an input, the software is accepting descriptions of biosensors formulated in domain specific terms. The numerical and computational models are created automatically according to the description supplied by a user of the software. The biosensor simulation software is able to simulate biosensors of complex structure, composed of several parts with different properties. It was developed using C++ programming language [43]. The software is open-sourced and publicly available at *http://github.com/kape1395/biosensor.solver-2D*. The C++ language was chosen mostly because of its high performance in floating-point computations.

Task for the biosensor simulation software is formulated by defining structural, geometrical and chemical properties of the considered biosensor. Biosensor description language is XML based [44] and was created to cover all these aspects of a model. Furthermore, the language allows the user to express the properties of the biosensor by relatively simple constructions. The XML was chosen because of its wide acceptance in the industry and tools, available for processing it. Formal grammar of the language was defined using "XML Schema". Inheritance semantics, provided by the "XML Schema", was used to implement extension points of the language.

The biosensor description language was developed as a domain specific language.

Use of domain specific terms as well as specialization on biosensors allowed to create fairly simple language. The language allows to formulate a model by defining a coordinate system and a domain of the model, substances considered in the model, reactions between the substances, homogeneous mediums, processes taking place on the boundaries of the mediums and a transducer. Homogeneous mediums and boundaries between them are defined in terms of reactions and diffusion of the substances.

In order to make the system more maintainable, the biosensor simulation software was developed composing it of several modules with defined interfaces and responsibilities. The cornerstone modules, composing the system are: the core, implementing the data model of the system and all the control functions, the solvers, implementing actual simulation algorithms, and the user interface, providing a user with an access to functions of the system.

The core of the system was implemented as a library defining interfaces and extension points as well as providing main algorithms. It is independent of the user interface as well as particular methods of biosensor simulation. This module implements parser of the biosensor description language. The parser converts the model supplied by a user to an internal data model that is shared between all the modules of the system. Use of the internal data model helps to isolate all the modules of the system from changes in the language. Besides the parser, eventing and simulation control mechanisms as well as input/output functions are implemented in the core of the system.

The user interface of the system was implemented as two non-interactive command line tools. One of them performs simulation of biosensors, while other is used to extract results of previously performed simulations in variety of projections. All the arguments for the tools are provided in the command line, output of the system is written to a filesystem and the standard output streams. This interface allows the user to employ various scripting languages for automated execution of computer experiments. Series of experiments are usually needed to be performed when investigating properties of biosensors.

The solver module implements particular methods for simulating operation of biosensors. Two concrete solvers are currently implemented in this module. They are both based on the method of finite differences and use semi-implicit scheme for approximation of the model equations. One of the solvers implements simulation of biosensors in the one-dimensional space while the other — in the two-dimensional space, in the Cartesian or the r-z plane of the cylindrical coordinate system. The one-dimensional solver gives no new functionality for the system, but allows simulations to be performed much faster for the models, that can be formulated in the one-dimensional space.

In order to make the solver general enough to simulate biosensors of different structure, it was developed in such way, that a solver for a particular model would be constructed from pre-defined primitives. Structure of the solver as well as the primitives are shown in figure 5.

As shown in figure 5, particular solver is assembled of three types of primitives. The `AreaSubSolver` primitive is responsible for simulation of diffusion-reaction processes in a rectangular area, representing part of homogeneous medium. The `BoundSubSolver` is used to connect `AreaSubSolver`s together as well as to simulate all processes on boundaries of the model domain. The `CornerSubSolver` is responsible for simulation of processes in angles of rectangular domains.

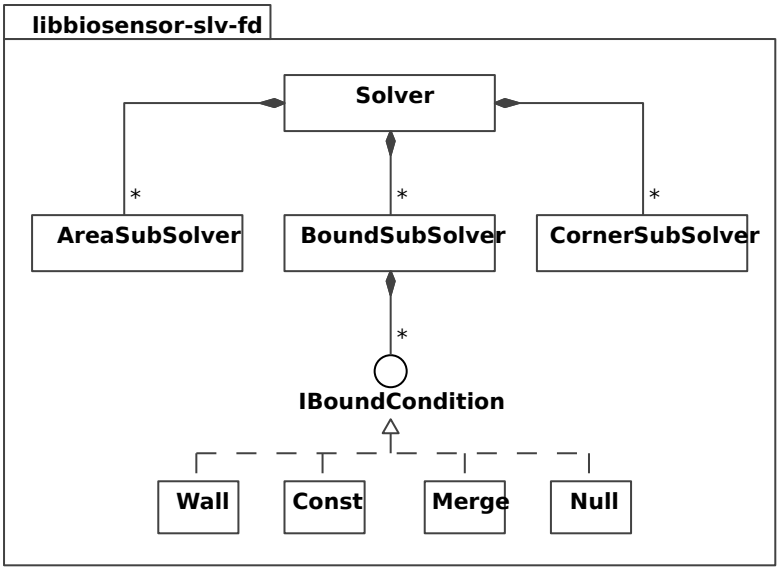

Figure 5. Primitives used to dynamically assemble particular solver.

4. Investigation of biosensor properties

Computer aided experiments were performed to investigate an adequateness of the models, presented in section 2, and properties of the modelled biosensors. The experiments were performed using the software, described in the section 3.

4.1. Validation of the model for the biosensor with carbon nanotube electrode

The following parameter values were used as a basic configuration, when investigating an adequateness of the model for the biosensor with the carbon nanotube electrode and properties of this biosensor [28, 45]:

$$\begin{aligned}
&r_1 = 2\times10^{-7}\mathrm{m}, \quad r_2 = 8\times10^{-7}\mathrm{m}, \quad E_0 = 4.55\times10^{-2}\mathrm{mol\,m^{-3}}, \quad n_e = 2,\\
&d_1 = 10^{-7}\mathrm{m}, \quad d_2 = 4\times10^{-7}\mathrm{m}, \quad d_3 = 10^{-5}\mathrm{m}, \quad d_4 = 1.5\times10^{-4}\mathrm{m},\\
&D_e = 3\times10^{-10}\mathrm{m^2\,s^{-1}}, \quad D_n = 2D_e = 6\times10^{-10}\mathrm{m^2 s^{-1}},\\
&k_1 = 6.9\times10^{2}\mathrm{m^3 mol^{-1} s^{-1}}, \quad k_2 = 6.9\times10^{4}\mathrm{m^3 mol^{-1} s^{-1}},\\
&\eta = 0.5, \quad \theta_{2,r} = \theta_{3,r} = \theta_r = 0.125, \quad \theta_{2,z} = \theta_{3,z} = \theta_z = 0.25,
\end{aligned} \tag{30}$$

where $d_1 = z_1$ and $d_i = z_i - z_{i-1}$, $i = 2, 3, 4$ are corresponding thicknesses of the layers composing the biosensor.

An adequateness of the proposed mathematical model for the biosensor with the carbon nanotube electrode was investigated by comparing results of the computer simulations with the results obtained by performing physical experiments [28, 46]. Results of the investigation are shown in figure 6.

As shown in figure 6, the results of the computer simulations are similar to those, obtained by performing physical experiments. The relative error of the simulations was not greater than 10%, when the biosensor was approaching to a steady state. Results of corresponding physical experiments can vary up to 10%, when repeating them several times [28], so the error of the simulations can be considered acceptable.

After the adequateness of the model was validated, an impact of geometrical and

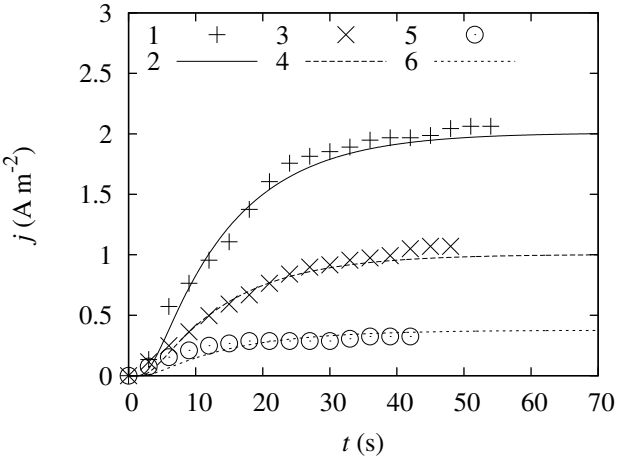

Figure 6. The response current density of the biosensor obtained by computer simulations (2, 4, 6) and performing physical experiments (1, 3, 5). The experiments were repeated with different concentrations of the substrate and the mediator: $M_0 = 0.2$, $S_0 = 9.9$ (1, 2), $M_0 = 0.05$, $S_0 = 4.98$ (3, 4), $M_0 = 0.005$, $S_0 = 1.99\,\mathrm{mol\,m^{-3}}$ (5, 6). Values of other parameters were defined in (30).

structural properties of the biosensor on its response was investigated. Investigation was done by performing computer aided experiments in a wide range of parameter values. One of the investigations was on the thickness of the carbon nanotube electrode.

4.2. Investigation of the properties of the biosensor with carbon nanotube electrode

An impact of the thickness of the carbon nanotube electrode on a response of the biosensor was investigated by performing computer aided experiments on a wide range of d_2 values. The steady state response current density J and the half-time of the steady state were measured when performing experiments [46]. Results of the investigation are shown in figure 7.

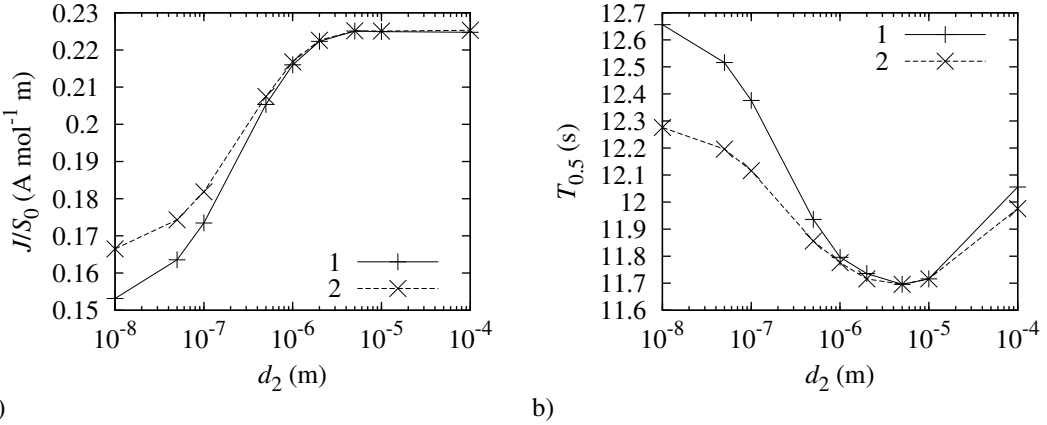

Figure 7. Dependency of the biosensor sensitivity (a) and the half-time of the steady state (b) on the thickness of the carbon nanotube electrode d_2. The results are shown for two substrate concentrations S_0: $5\,\mathrm{mol\,m^{-3}}$ (1) and $0.05\,\mathrm{mol\,m^{-3}}$ (2).

As shown in figure 7 a, a biosensor sensitivity J/S_0 is a non-linear monotonously increasing function of d_2. When the electrode is relatively thin ($d_2 < 5 \times 10^{-6}$ m), change of the electrode thickness has notable impact on the sensitivity of the biosensor and has almost no impact, when the layer is relatively thick.

The dependency of the half-time $T_{0.5}$ of the steady state on the thickness of the carbon nanotube electrode d_2 is shown in figure 7 b. This dependency is non-monotonous and has a minimum at $d_2 \approx 5 \times 10^{-6}$ m. This value matches approximately with the thickness of the electrode for which the increase of d_2 starts to have small impact on the response of the biosensor. Such behavior of the biosensor allows to select an optimal thickness for the carbon nanotube electrode and develop biosensors with better responsiveness.

4.3. Simulation of the biosensor with a perforated membrane using one-dimensional model

Numerical simulations were performed in order to determine conditions, when the one-dimensional model can be used instead of the two-dimensional model for a precise simulation of biosensors with a perforated membrane. Results of the one-dimensional model were compared to the corresponding results of the two-dimensional model, assuming latter as a precise ones. The one-dimensional model was evaluated by measuring its relative error

$$\eta_J(D, S_0) = \frac{|J - J^*|}{J}, \tag{31}$$

where D stands for the averaged diffusion coefficient D_3^* used in the experiments, S_0 stands for a substrate concentration in the buffer solution, J is a density of a steady state current, calculated using the two-dimensional model and J^* is the corresponding density, calculated using the one-dimensional model.

The relative error η_J depends on the value of the effective diffusion coefficient D_3^* used in the one-dimensional model and the catalytical as well as the geometrical parameters of the modelled biosensor. Values of all the parameters of the one-dimensional model excluding only D_3^* can be derived directly from the corresponding two-dimensional model. For a concrete substrate concentration S_0, the effective diffusion coefficient D_3^* can be expressed as a value minimizing the relative error η_J,

$$D_3^*(S_0) = \arg\min_D \eta_J(D, S_0), \quad 0 \le D \le \max(D_2, D_3), \tag{32}$$

where the upper value of D comes from (26). The minimization (32) can be achieved by changing D and solving the one-dimensional model of the biosensor action using different values D of D_3^*. In order to find the minimal value of D_3^* in the efficient way, the following procedure was introduced.

Let E^* be an ordered sequence of triplets $\langle D_{3,i}^*, J_i^*, T_{0.5,i}^* \rangle$, where J_i^* is the density of the simulated stationary current, $T_{0.5,i}^*$ is the half-time of the steady state, and $D_{3,i}^*$ denotes the averaged diffusion coefficient used in the simulation, i = 1,2,.... Each triplet in the sequence E^* couples the parameters characterizing a concrete simulation of the biosensor action by using one-dimensional model. The half-time $T_{0.5,i}^*$ of the steady state response stands for the dynamics of the biosensor action. The order in this sequence is preserved according to the following rule: $D_{3,i}^* \ge D_{3,i+1}^*, \forall i \ge 1$. The procedure of calculation of D_3^* is defined by the following steps:

1. Simulate the operation of a particular biosensor using the two-dimensional model. The steady state current density J and half-time $T_{0.5}$ of the steady state are results of this simulation to be used in the next steps. Go to step 2.

2. Perform a preliminary variation of the effective diffusion coefficient D_3^*. The biosensor responses are simulated by using the one-dimensional model changing values of $D_3^* \in [0, \max(D_2, D_3)]$. The simulation results are appended to the sequence E^*. Let M denote the number of elements in E^*. Go to step 3.

3. Construct a set of intervals $G = \{[D_{3,i+1}^*, D_{3,i}^*] : J_{i+1}^* \leq J \leq J_i^*$ or $J_{i+1}^* \geq J \geq J_i^*, i \geq 1\}$. If $G = \emptyset$ then go to step 4, otherwise go to step 6.

4. Find m $(1 \leq m \leq M)$ for which the difference $|I_m^* - I|$ is the minimal. If variation of the effective diffusion coefficient of the adjacent elements in the sequence E^* is small enough, i.e. $(D_{3,m-1}^* - D_{3,m+1}^*)/D_{3,m}^* < \epsilon$, then stop the procedure with $D_{3,m}^*$ as the output. Otherwise, go to step 5.

5. Simulate two more responses of the biosensor at $D_3^* = (D_{3,m-1}^* + D_{3,m}^*)/2$ and $D_3^* = (D_{3,m}^* + D_{3,m+1}^*)/2$, where m comes from step 4. Append the corresponding two triplets to the sequence E^* and go to step 3.

6. For each interval from the set G produced in step 3, apply the method of chords to find a number of values of D_3^* minimizing (32). Between them, find k-th for which the corresponding difference $|T_{0.5} - T_{0.5,k}^*|$ is minimal. The output of the procedure is $D_{3,k}^*$.

In step 2, the preliminary variation of the effective diffusion coefficient D_3^* can be done in a number of different ways. In this work, it was achieved by simulating the biosensor action by using the one-dimensional model at the values of D_3^* chosen as follows:

$$D_{3,j}^* = \begin{cases} \max(D_2, D_3) & j = 1; \\ \frac{D_{3,j-1}^*}{2} & j = 2, \ldots, N. \end{cases} \tag{33}$$

This sequence is constructed in the way to cover the entire domain of D_3^* $(0 < D_3^* \leq \max(D_2, D_3))$ and to find a smaller subdomain in which the value minimizing the error η_J exists. The result of each simulation is appended to the sequence E^*. The preliminary variation is performed until the stationary current density J_j^* starts to decrease and becomes smaller than J.

When investigating properties of biosensors it is important to evaluate the modelling error for a wide range of the substrate concentrations. An application of the two-dimensional model for calculation of the "true" biosensor response is an essential feature of the procedure used for determination of the effective diffusion coefficient D_3^*. The simulation of the biosensor response supposes a particular concentration of the substrate. If the substrate concentration effects the modelling error then it is important to determine the concentration to be used in the procedure when calculating a value of D_3^*. On the other hand, having a value of D_3^*, it is important to determine an interval of substrate concentrations for which the value of D_3^* can be applied for accurate prediction of the response. The relative error η_S was introduced to evaluate the error of the one-dimensional model at various substrate concentrations,

$$\eta_S(S_D, S_V) = \eta_J(D_3^*(S_D), S_V), \tag{34}$$

where D_3^* is the effective diffusion coefficient, S_D is the substrate concentration used in two-dimensional simulation when calculating the effective diffusion coefficient, S_V is the substrate concentration used in one-dimensional simulation. η_S can be called as a one-dimensional modelling error arose because of an application of D_3^* for the prediction of the biosensor response at the substrate concentration S_V.

The numerical simulations were performed at different geometries of the membrane perforation and levels of filling the holes with the enzyme. The following values of the model parameters were constant in all the numerical experiments:

$$\begin{aligned} &D_1 = 1\,\mu\mathrm{m}^2\mathrm{s}^{-1}, \quad D_2 = 300\,\mu\mathrm{m}^2\mathrm{s}^{-1}, \quad D_3 = 600\,\mu\mathrm{m}^2\mathrm{s}^{-1}, \\ &r_2 = 1\,\mu\mathrm{m}, \quad z_1 = 2\,\mu\mathrm{m}, \quad z_2 = z_1 + 2\,\mu\mathrm{m}, \quad z_4 = z_2 + 10\,\mu\mathrm{m}, \\ &K_M = 100\,\mu\mathrm{M}, \quad V_{max} = 10\,\mu\mathrm{Ms}^{-1}, \quad n_e = 2. \end{aligned} \tag{35}$$

In order to investigate the dependence of the relative error η_I on the level β of filing the holes of the perforated membrane with the enzyme, the biosensor response was simulated at the following three values of β: 0 ($z_3 = z_2$) when the holes were fully filled with the buffer solution (no enzyme in the holes), 0.5 ($z_3 = (z_2 + z_4)/2$) when the holes were half-filled with the enzyme, and 1 ($z_3 = z_4$) when holes were fully filled with the enzyme [47]. Calculated values of the relative error η_S are depicted in figure 8.

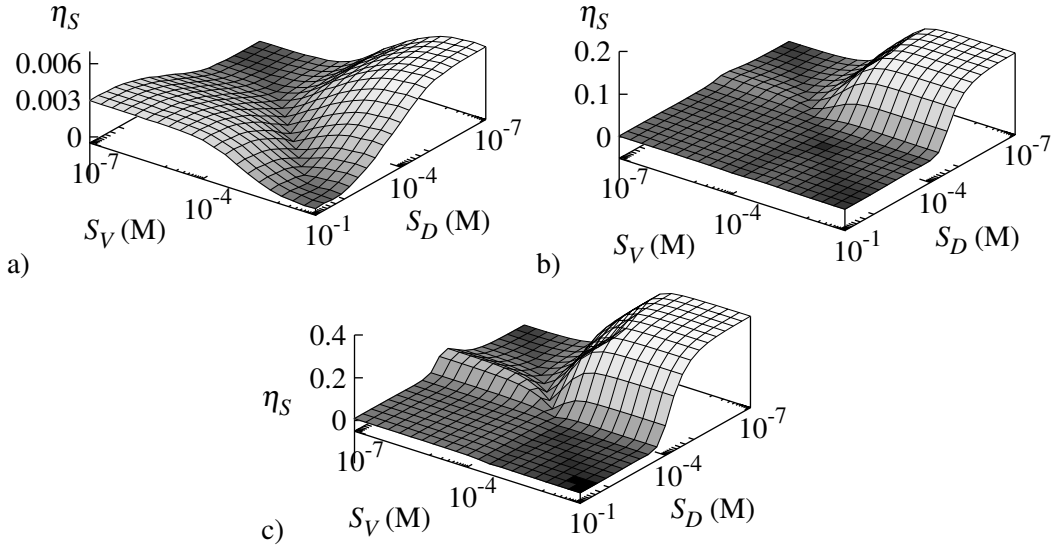

Figure 8. The relative error η_S at three levels (β) of the enzyme filling: 0 (a), 0.5 (b) and 1 (c); $\alpha = 0.01$, values of all other parameters are as defined in (35).

The smallest relative errors were achieved in the case when there was no enzyme in the holes (figure 8 a). In this case, η_S was less than 0.6 %. When the level of the enzyme raises, a preciseness of the one-dimensional model decreases. In the case when $\beta = 1$ the relative error of the one-dimensional model reaches 37% (figure 8 c). When the holes were half-filled with the enzyme, the modelling error was less than 15% (figure 8 b).

Figure 8 also shows the dependence of the error η_S on the substrate concentrations S_D and S_V. It can be seen from figure 8 that the relative error η_S is usually smaller when one-dimensional model is applied for the substrate concentration S_V smaller than that (S_D) used in two-dimensional simulation for evaluation of D_3^*. Con-

sequently, the substrate concentration used to find the effective diffusion coefficient should be chosen larger than concentrations for which the one-dimensional model will be applied.

Conclusions

1. The carbon nanotube electrode can be modelled at the macroscopic level and treated as a homogeneous medium, when modelling biosensors with such electrode. The electrochemical reaction can be modelled as a very fast reaction, taking place in all the volume of the electrode.

2. One-dimensional models can be used when modelling biosensors with an outer perforated membrane, but not in all the cases. More precise two-dimensional models should be used for simulation of biosensors when the perforation level of the membrane is very low or there is a lot of enzyme in the holes of the membrane.

3. The considered biosensor models, formulated in the one and two-dimensional spaces, can be generalized and the structural elements of the models can be defined. The construction of numerical and computational models for the biosensors can be automated, constructing them out of the structural primitives. The construction of mathematical models can be automated partially, by deriving some boundary conditions out of properties of related mediums.

4. The structural properties of the carbon nanotubes have noticeable impact on the output of the biosensor with the carbon nanotube electrode. The current generated by the biosensor has nonlinear dependence on a thickness of the enzyme layer and is proportional to the perforation level of the outer membrane.

Publications on the thesis topic

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About the author

Karolis Petrauskas was born in Vilnius, on 28th of October, 1981. In 2000 graduated from Mindaugas Secondary School, and in 2000–2006 obtained computer science bachelor and master degrees in the Vilnius University. In 2006–2010 carried out doctoral studies in the Vilnius University. Since 2001 is working in a telecommunications company, currently as an IT architect. Since 2006, Karolis Petrauskas is working in the Vilnius University as an assistant, where is supervising course works as well as bachelor and master thesis. Currently is conducting practical training sessions on object oriented programming in C++ and laboratory works of systems engineering.

Reziumė

Tyrimų sritis ir problemos aktualumas

Biojutikliai yra analitiniai įrenginiai, skirti medžiagoms aptikti bei jų koncentracijoms matuoti. Pagrindinės biojutiklio sudedamosios dalys yra biologiškai aktyvi medžiaga, paprastai fermentas, gebantis atpažinti konkrečią medžiagą ir keitiklis, transformuojantis biologinį atpažinimo faktą į elektrinį signalą. Šis elektrinis signalas yra sustiprinamas, apdorojamas ir pateikiamas biojutiklio vartotojui [1–3].

Biojutikliai yra taikomi įvairiose srityse, o ypač medicinoje, aplinkos užterštumo tyrimuose bei narkotinių medžiagų aptikimui. Pagal „Global Industry Analysts Inc.“ atliktą tyrimą, biojutiklių rinka 2009 metais sudarė apie 8.2 milijardo JAV dolerių ir yra tikimasi tolimesnio šios rinkos augimo, kasmet apie 6.3% [8].

Kuriant naują biojutiklį, daug darbo ir lėšų skiriama eksperimentiniams bandymams bei biojutiklių kalibravimui. Siekiant sumažinti fizinių eksperimentų skaičių yra pasitelkiamas matematinis biojutikliuose vykstančių procesų modeliavimas [9, 10]. Šie procesai yra aprašomi netiesinėmis diferencialinėmis lygtimis dalinėmis išvestinėmis [11, 12].

Biojutiklių veikimą aprašančių lygčių analiziniai sprendiniai yra žinomi tik atskiriems atvejams. Tiriant sudėtingos struktūros biojutiklius plačiame tiriamų medžiagų koncentracijų diapazone, matematinio modelio lygtims spręsti tenka naudoti skaitinius metodus. Šiame darbe yra taikomi baigtinių skirtumų bei kintamų krypčių metodai [13].

Realūs biojutikliai turi sudėtingą struktūrą. Jie būna sudaryti iš skirtingomis savybėmis pasižyminčių sluoksnių. Biojutiklių konstravimui yra naudojamos perforuotos ir selektyvios membranos, porėtos medžiagos ir t.t.. Viena tokių medžiagų yra anglies nanovamzdeliai [18]. Pastariuoju metu juos imta taikyti kuriant labai aukšto jautrumo biojutiklius [3, 14, 19–24].

Įprastai, modeliuojant biojutiklius, kiekvienam struktūriškai naujam biojutikliui yra sudaromas matematinis modelis. Tuomet jis keičiamas skaitiniu ir įgyvendinama programinė įranga, leidžianti spręsti skaitinio modelio lygčių sistemą [11]. Matematinių bei skaitinių modelių kūrimas yra daug atidumo reikalaujantis darbas. Automatizuotas tokių modelių bei programų sudarymas leistų efektyviau vykdyti biojutiklių modeliavimą, šį darbą padaryti paprastesniu, atsparesniu klaidoms.

Tyrimo objektas

Šios disertacijos tyrimų objektas yra matematiniai ir kompiuteriniai biojutiklių modeliai, aprašantys biojutiklių, sudarytų iš kelių skirtingas savybes turinčių dalių,

veikimą. Disertacijoje nagrinėjami modeliai formuluojami vienmatėje bei dvimatėje erdvėse, aprašomi diferencialinėmis lygtimis dalinėmis išvestinėmis su netiesiniais nariais ir yra sprendžiami skaitiškai, naudojant baigtinių skirtumų metodą. Skaitiniai modeliai yra įgyvendinami kompiuterine programa.

Darbo tikslas ir uždaviniai

Šio darbo tikslas yra automatizuoti biojutiklių, kurių matematiniai modeliai aprašomi diferencialinėmis reakcijos-difuzijos tipo lygtimis vienmatėje arba dvimatėje erdvėje, kompiuterinių modelių sudarymą ir, taikant kompiuterinį modeliavimą, ištirti konkrečių biojutiklių savybes. Šiam tikslui pasiekti buvo sprendžiami sekantys uždaviniai.

1. Pasiūlyti fermentu prisotinto anglies nanovamzdelių elektrodo, taikomo konstruojant biojutiklius, modeliavimo būdą ir sudaryti biojutiklio su perforuota membrana ir anglies nanovamzdelių elektrodu modelį.

2. Ištirti perforuotos membranos, taikomos biojutikliuose, modeliavimo būdus ir jų taikymo aplinkybes.

3. Apibendrinti vienmatėje ir dvimatėje erdvėje formuluojamų modelių struktūrą biojutikliams, sudarytiems iš kelių skirtingas savybes turinčių dalių.

4. Pasiūlyti biojutiklių aprašo kalbą, leidžiančią formuluoti biojutiklių modelius dalykinės srities sąvokomis, bei sukurti algoritmus ir lanksčią programinę įrangą, leidžiančią vykdyti biojutiklių, kurių modeliai formuluojami vienmatėje arba dvimatėje erdvėje, veikimo modeliavimą.

5. Taikant sukurtą programinę įrangą, ištirti pasiūlytų modelių adekvatumą bei nagrinėjamų biojutiklių geometrinių ir struktūrinių savybių įtaką biojutiklio elgsenai.

Tyrimų metodika

Disertacijoje nagrinėjami biojutiklių modeliai buvo formuluojami netiesinėmis diferencialinėmis lygtimis dalinėmis išvestinėmis. Modelių lygčių sistemoms aproksimuoti buvo taikomi baigtinių skirtumų bei kintamų krypčių metodai. Disertacijoje pateikti biojutiklių tyrimai buvo atliekami taikant kompiuterinį modeliavimą.

Siekiant sukurti programinę įrangą, užtikrinančią automatizuotą biojutiklių matematinių modelių sudarymą bei jų sprendimą, buvo tiriami publikuoti biojutiklių modeliai, sisteminamos modelių struktūrinės dalys ir jų jungimo būdai.

Darbo rezultatai ir mokslinis naujumas

1. Sudarytas matematinis modelis biojutikliui su anglies nanovamzdelių elektrodu. Taikant kompiuterinį modeliavimą ištirtas modelio adekvatumas.

2. Nustatyti kriterijai, apibrėžiantys, kada biojutiklį su perforuota membrana galima modeliuoti vienmačiu modeliu. Pasiūlyta apibendrintojo difuzijos koeficiento radimo procedūra.

3. Susisteminti elementai, naudojami biojutiklių modelių formulavimui, pagrindinį dėmesį skiriant biojutiklio struktūrinėms savybėms modeliuoti.

4. Apibrėžta biojutiklių modelių aprašo kalba ir sukurta programinė įranga, leidžianti modeliuoti biojutiklių veikimą vienmačiais modeliais arba modeliais, formuluojamais stačiakampėje dvimatės erdvės srityje.

5. Taikant sukurtą biojutiklių modeliavimo programinę įrangą, ištirta biojutiklio su anglies nanovamzdelių elektrodu struktūrinių ir geometrinių savybių įtaka biojutiklio elgsenai.

Praktinė darbo rezultatų reikšmė

Disertacijoje pateiktas modelis biojutikliui su anglies nanovamzdelių eletrodu leidžia tirti biojutiklio savybes ir jame vykstančius procesus. Pasiūlytas nanovamzdelių elektrodo modeliavimo būdas gali būti taikomas ir kitų biojutiklių, kuriuose yra naudojamas toks elektrodas, modeliavimui.

Darbe nustatyti kriterijai, apibrėžiantys, kada galima naudoti vienmatį modelį biojutikliams su perforuota membrana. Daugeliu atvejų tai leidžia biojutiklių veikimo modeliavimą vykdyti daug efektyviau, nei tai galima atlikti naudojant dvimatėje erdvėje formuluojamus modelius.

Sukurtą programinę įrangą galima naudoti kompiuterizuotam biojutiklių savybių tyrimui. Naudojantis šia programine įranga galima lengviau formuluoti sudėtingos struktūros biojutiklių modelius bei vykdyti jų veikimo modeliavimą. Tai leidžia lengviau tirti struktūrinių biojutiklio pakeitimų įtaką jų veikimui. Ši programinė įranga taip pat gali būti efektyviai naudojama naujų biojutiklių modelių formulavimui, nes sudaro galimybes lengviau patikrinti modelio teisingumą.

Kompiuterinių eksperimentų pagalba nustatytos biojutiklių savybės gali būti panaudotos kuriant efektyvesnius biojutiklius.

Disertacijos rezultatai panaudoti įgyvendinant projektus „Bioelektrokatalizė sintezėje ir analizėje (BIOSA)“, kurį finansavo Lietuvos mokslo taryba (sutarties Nr. PBT-04/2010) ir „Kompiuterinių metodų, algoritmų ir įrankių efektyviam sudėtingos geometrijos biojutiklių modeliavimui ir optimizavimui sukūrimas“, finansuojamą iš ES Socialinio fondo pagal VP1-3.1-ŠMM-07-K priemonę „Parama mokslininkų ir kitų tyrėjų mokslinei veiklai (Visuotinė dotacija)“ lėšų.

Ginami teiginiai

1. Biojutiklius su anglies nanovamzdelių elektrodu galima modeliuoti makroskopiniame lygyje, nanovamzdelių sritį laikant homogeniška.

2. Modeliai, formuluojami vienmatėje erdvėje, gali būti taikomi tiksliam biojutiklių su išorine perforuota membrana modeliavimui, tačiau ne visais atvejais.

3. Biojutiklių modeliai, formuluojami stačiakampėje dvimatės erdvės srityje, nagrinėjant geometrines jų savybes, gali būti apibendrinti ir surasti primityvai, iš kurių tokie modeliai gali būti konstruojami.

4. Biojutiklių su anglies nanovamzdelių elektrodu veikimas yra žymiai įtakojamas struktūrinių anglies nanovamzdelių savybių.

Išvados

1. Modeliuojant biojutiklių veiksmą, anglies nanovamzdelių elektrodą galima laikyti homogeniška terpe ir analizuoti makroskopiniame lygyje. Elektrocheminę

reakciją šioje medžiagoje galima laikyti vykstančia visame tūryje ir esančią labai greita.

2. Biojutikliams su išorine perforuota membrana vienmatėje erdvėje formuluojami matematiniai modeliai gali būti taikomi, tačiau ne visada. Esant mažam perforacijos lygiui, ar skylutėse esant daug fermento, vienmačiai modeliai yra netikslūs. Tokiais atvejais turėtų būti naudojami detalesni, dvimatėje erdvėje formuluojami modeliai.

3. Nagrinėtus biojutiklių modelius, formuluojamus vienmatėje bei dvimatėje erdvėje galima apibendrinti ir išskirti primityvus, iš kurių šie modeliai sudaromi. Išskyrus tokius primityvus galima automatizuoti skaitinių ir kompiuterinių modelių sudarymą. Matematinių modelių formulavimas gali būti automatizuotas iš dalies, automatizuojant kai kurių kraštinių sąlygų parinkimą.

4. Biojutiklio su anglies nanovamzdelių elektrodu ir išorine perforuota membrana atsakas yra žymiai įtakojamas struktūrinių nanovamzdelių savybių. Šio biojutiklio generuojama pusiausvyroji srovė netiesiškai priklauso nuo fermento sluoksnio storio, tačiau yra proporcinga išorinės membranos perforacijos lygiui.

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