APTAMERS AND ODORANT BINDING PROTEINS – INNOVATIVE RECEPTORS FOR ELECTRONIC SMALL LIGAND SENSING

Projektträger:
Danube Private University

Wissenschaftliche Leitung:
Wolfgang Knoll

Weitere beteiligte Einrichtungen:
Universität Ulm

Handlungsfeld(er)
Umwelt, Klima und Ressourcen

Wissenschaftsdisziplin(en)
1040 - Chemie (70 %)
1030 - Physik, Astronomie (10 %)
1060 - Biologie (20 %)

Förderinstrument: Projekte Grundlagenforschung

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Fördersumme: € 300.000,00

Kurzzusammenfassung:
Die quantitative Bestimmung kleiner Analyten in klinisch relevanten Flüssigkeiten oder in Luft ist zur Zeit sowohl aus wissenschaftlicher Sicht, als auch für praktische Anwendungen eine anspruchsvolle Herausforderung. Die wesentlichsten Elemente liegen dabei in (i) der Empfindlichkeit des Transducers, (ii) der Selektivität, die zur Unterscheidung zwischen konkurrierenden Analyten erforderlich ist, und (iii) der Unterdrückung unspezifischer Bindungsvorgänge. Bei elektronischen Sensorikkonzepten zeichnen sich Graphen-basierte Feldeffekttransistoren (gFETs) durch ihre geringe Größe, ihre hervorragenden elektrischen Eigenschaften und ihre hohe Empfindlichkeit gegenüber oberflächennahen Ladungen und elektrischen Feldern aus. Die Spezifität für einen bestimmten Zielanalyten kann in gFETs durch die Anbindung von zielspezifischen Rezeptoren an der Oberfläche integriert werden; die sehr kleine Debye-Länge in (physiologischen) Pufferlösungen schränkt die Signalstärke jedoch erheblich ein. Ein Schwerpunkt der vorgeschlagenen Arbeiten ist die Feststellung des Einflusses der verwendeten Oberflächenchemie, die für die Immobilisierung dieser Rezeptoren auf der Transduceroberfläche verwendet wird, und wie diese Kopplung die endgültigen Messsignale beeinflusst. Ein weiterer Ansatz befasst sich mit Möglichkeiten zur Beeinflussung der Debye-Beschränkungen durch die Verwendung „Polymerbrushes“ oder Hydrogelen auf der Sensoroberfläche. Für die Ausweitung dieser Studien auf die Überwachung von THC direkt in der Luft schlagen wir vor, die natürliche Schleimhaut durch eine ultradünne Hydrogelschicht auf dem Transistor zu imitieren, die die empfindlichen bioorganischen Komponenten vor Luft schützt, indem sie sie in einem vollständig hydratisierten Zustand hält, ohne zu verhindern, dass die flüchtigen organischen Verbindungen Zugang zu den Rezeptoren auf der Sensoroberfläche erhalten, indem sie durch die Hydrogelschicht diffundieren.

Schlüsselbegriffe:
Sensorik, (Atem)gas Analytik, RGO-Feldeffekttransistor, Rezeptorsystems

ERGEBNISSE

PEER-REVIEWED PUBLICATION

Clickable" graphene nanoribbons for biosensor interfaces
We report on the synthesis of "clickable" graphene nanoribbons (GNRs) and their application as a versatile interface for electrochemical biosensors. GNRs are successfully deposited on gold-coated working electrodes and serve as a platform for the covalent anchoring of a bioreceptor (i.e., a DNA aptamer), enabling selective and sensitive detection of Interleukin 6 (IL6). Moreover, when applied as the intermediate linker on reduced graphene oxide (rGO)-based field-effect transistors (FETs), the GNRs provide improved robustness compared to conventional aromatic bi-functional linker molecules. GNRs enable an orthogonal and covalent attachment of a recognition unit with a considerably higher probe density than previously established methods. Interestingly, we demonstrate that GNRs introduce photoluminescence (PL) when applied to rGO-based FETs, paving the way toward the simultaneous optical and electronic probing of the attached biointerface.
https://doi.org/10.1039/D3NH00590A
https://pubs.rsc.org/en/content/articlelanding/2024/nh/d3nh00590a

Dynamic studies of antibody-antigen interactions with an electrolyte-gated organic transistor
Affinity-based biosensors employing surface-bound biomolecules for analyte detection are important tools in clinical diagnostics and drug development. In this context, electrolyte-gated organic transistors (EGOTs) are emerging as ultrasensitive label-free biosensors. In this study, we present an EGOT sensor integrated within a microfluidic system. The sensor utilizes the cytomegalovirus (CMV) phosphoprotein 65 as a biorecognition element to detect the pathological biomarker human anti-cytomegalovirus antibody in solution. The biorecognition element is grafted onto the gate electrode by exploiting the polyhistidine-tag technology. Real-time monitoring of the EGOT response, coupled with a two-compartment kinetic model analysis, enables the determination of analyte concentration, binding kinetics, and thermodynamics of the interaction. The analysis of the relevant kinetic parameters of the binding process yields a reliable value for the thermodynamic equilibrium constant and suggests that the measured deviations from the Langmuir binding model arise from the co-existence of binding sites with different affinities toward the antibodies.
https://doi.org/10.1016/j.xcrp.2024.101918
https://www.sciencedirect.com/science/article/pii/S2666386424001668

A Blautia producta specific gFET-based aptasensor for quantitative monitoring of microbiome quality
- H. Xing, Y. Zhang, R. Li, H.-M. Ruzicka, C. Hain, J. Andersson, A. Bozdogan, M. Henkel, U. Knippschild, R. Hasler, C. Kleber, W. Knoll, A.-K. Kissmann, F. Rosenau
Nanoscale Horiz., 2025, 10, 424-434.
https://doi.org/10.1039/D4NH00381D

Dual Electronic and Optical Monitoring of Biointerfaces by a Grating-Structured Coplanar-Gated Field-Effect Transistor
- R. Hasler, P. A. Livio, A. Bozdogan, S. Fossati, S. Hageneder, V. Montes-García, J. Movilli, T. Moazzenzade, L. Loohuis, C. Reiner-Rozman, A. Tamayo, C. Fiedler, M. Ibáñez, C. Kleber, J. Huskens, J. Dostalek, P. Samorì, W. Knoll
IEEE Sens. J, 2025, 25, 7, 10521-10529.
DOI: 10.1109/JSEN.2025.3533113
https://doi.org/10.1109/JSEN.2025.3533113

Specific gFET-Based Aptasensors for Monitoring of Microbiome Quality: Quantification of the Enteric Health-Relevant Bacterium Roseburia Intestinalis
- Y. Zhang, H. Xing, R. Li, J. Andersson, A. Bozdogan, R. Strassl, B. Draphoen, M. Lindén, M. Henkel, U. Knippschild, R. Hasler, C. Kleber, W. Knoll, A.-K. Kissmann, F. Rosenau
Adv. Healthc. Mater., 2025, 14, 2405827.
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Antimicrobial Activity of the Peptide C14R Against Ab Initio Growing and Preformed Biofilms of Candida albicans, Candida parapsilosis and Candidozyma auris

- J.-C. Walter, A.-K. Kissmann, D. Gruber, D. Alpízar-Pedraza, E. M. Martell-Huguet, N. Preising, A. Rodriguez-Alfonso, L. Ständker, C. Kleber, W. Knoll, S. Stenger, C. Firacative, F. Rosenau

Biomolecules, 2025, 15, 322.
https://doi.org/10.3390/biom15030322

The Power of Old Hats: Rediscovering Inosine-EpPCR to create starting libraries for whole Cell-SELEX

- G. Bolotnikov, A.-K. Kissmann, D. Gruber, A. Bellmann, R. Hasler, C. Kleber, W. Knoll, F. Rosenau

Biosensors, 2025, 15, 7, 448.
https://doi.org/10.3390/bios15070448

An Aptamer-Based gFET-Sensor for Specific Quantification of Gene Therapeutic Human Adenovirus HAdV-5

The combination of rGO-FETs (reduced Graphene Oxide Field-Effect Transistors) and DNA-oligonucleotide aptamers to sense analytes has been shown to be a promising technological approach, achieving high sensitivity and selectivity. With human adenovirus type 5 (HAdV-5) particles as the target, we here demonstrate the application of the aptamer/FET combination for detection of this medically and biotechnologically relevant viral vector. A focused anti-HAdV-5 aptamer library was evolved in a nine-round SELEX process, allowing for the specific fluorescent labeling of HAdV-5 and related subtypes. Moreover, this library was already sufficient to serve as the binding entity on a gFET sensor for sensitive quantification of the virus particles. Adenoviruses have been widely used as gene delivery vectors for gene therapy and genetic vaccination. The use of adenoviral vectors within the vaccination campaign against COVID-19 emphasized the need for robust biotechnological production processes, which additionally require sensitive product formation monitoring. We believe that these type of gFET-based aptasensors can serve as the technological monitoring basis in virus production processes in the near future.

https://doi.org/10.3390/bios15090605

FluMag-SELEX derived specific individual aptamers can fluorescently label the ageing immanent protein-crosslinker Glucosepane in diabetic mouse tissues

Nowadays, aptamers have transitioned into valuable antibody alternatives. We present the first anti-glucosepane aptamer targeting a key glycation product linked to aging and diabetes. Glu3, with high specificity and affinity, enabled the first-ever direct, fluorescence-based histological staining of glucosepane in murine tissue, distinguishing diabetic samples from wild-type samples without secondary reagents.

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