proceedings

MDPI

Abstract

Design of a Multiplex Sensing Platform: AFM as a Nanolithographic Tool †

Silvia Maria Cristina Rotondi [1,*], Paolo Canepa [1], Maurizio Canepa [1,2] and Ornella Cavalleri [1]

1 Department of Physics, University of Genova, 16146 Genova, Italy; paolocanepa@unige.it (P.C.); canepa@fisica.unige.it (M.C.); cavalleri@fisica.unige.it (O.C.)
2 Istituto Nazionale di Fisica Nucleare Sezione di Genova, 16146 Genova, Italy
* Correspondence: silviamariacristina.rotondi@edu.unige.it
† Presented at the 4th International Electronic Conference on Biosensors, 20–22 May 2024; Available online: https://sciforum.net/event/IECB2024.

Keywords: DNA; hybridization; AFM; nanoshaving; nanografting; self assembled monolayers; quantitative imaging (QI); biosensor

Coupling spectroscopic ellipsometry (SE), quartz crystal microbalance with dissipation (QCM-D), X-ray photoemission spectroscopy (XPS), and atomic force microscopy (AFM), we developed a multi-technique approach to characterize the surface immobilization of probe DNA strands, as a tool for the design of a DNA-based biosensor for the detection of disease-related oligonucleotide strands [1–3]. The hybridization of complementary target sequences is monitored through in situ, non-destructive, and real-time analysis.

The multiplexing detection of different oligonucleotide sequences is of great interest for differential diagnosis. To this end, we exploit AFM in a nanolithography mode to obtain micrometric platforms of thiolated DNA. Grafting is performed by removing previously chemisorbed inert alkanethiol SAMs and replacing them with short thiolated DNA molecules. Changing grafting parameters, DNA patches with different molecular densities were obtained. The analysis of images acquired in low-perturbative quantitative imaging (QI) mode highlighted the coexistence of molecular domains of different heights and thus different densities, which were not formerly observed using contact AFM imaging. By exposing the DNA platforms to target DNA (down to the nM level), all patches increased in height, indicating a successful hybridization. Comparing the height of the patches before and after hybridization showed a higher relative height increase in the less dense patches, indicating them as most suitable for targeting oligonucleotide sequences [4]. This method allows the grafting of different thiolated DNA strands onto the same substrate. Different sequences, characterized by 10 mismatches, were employed. Upon exposing the platform to different targets, a selective hybridization of specific probe DNA patches was observed, demonstrating efficient multiplexing targeting.

check for updates

Citation: Rotondi, S.M.C.; Canepa, P.; Canepa, M.; Cavalleri, O. Design of a Multiplex Sensing Platform: AFM as a Nanolithographic Tool. *Proceedings* **2024**, *104*, 21. https://doi.org/10.3390/proceedings2024104021

Academic Editor: Michael Thompson

Published: 28 May 2024

Copyright: © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https://creativecommons.org/licenses/by/4.0/).

Author Contributions: Conceptualization, O.C. and M.C.; methodology, O.C.; formal analysis, S.M.C.R. and P.C.; investigation, S.M.C.R.; writing—original draft preparation, O.C. and S.M.C.R.; writing—review and editing, S.M.C.R., P.C., M.C. and O.C.; visualization, S.M.C.R. and P.C.; supervision, O.C.; funding acquisition, O.C. and M.C. All authors have read and agreed to the published version of the manuscript.

Funding: Financial support was received from the Università degli Studi di Genova and Italian Ministry of Education (grant RBAP11ETKA-005).

Institutional Review Board Statement: Not Applicable.

Informed Consent Statement: Not Applicable.

Data Availability Statement: The data presented in this study are available from the corresponding author upon request.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Pinto, G.; Parisse, P.; Solano, I.; Canepa, P.; Canepa, M.; Casalis, L.; Cavalleri, O. Functionalizing gold with single strand DNA: Novel insight into optical properties via combined spectroscopic ellipsometry and nanolithography measurements. *Soft Matter* **2019**, *15*, 2463–2468. [CrossRef] [PubMed]
2. Pinto, G.; Canepa, P.; Canale, C.; Canepa, M.; Cavalleri, O. Morphological and mechanical characterization of DNA SAMs combining nanolithography with AFM and optical methods. *Materials* **2020**, *13*, 2888. [CrossRef] [PubMed]
3. Pinto, G.; Dante, S.; Rotondi, S.M.; Canepa, P.; Cavalleri, O.; Canepa, M. Spectroscopic ellipsometry investigation of a sensing functional interface: DNA SAMs hybridization. *Adv. Mater. Interfaces* **2022**, *9*, 2200364. [CrossRef]
4. Rotondi, S.M.; Canepa, P.; Angeli, E.; Canepa, M.; Cavalleri, O. DNA Sensing Platforms: Novel Insights into Molecular Grafting Using Low Perturbative AFM Imaging. *Biosensors* **2023**, *23*, 4557. [CrossRef] [PubMed]

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.