Overview on the ADONIS Project:

Accurate Diagnosis of prostate cancer using Optoacoustic detection of biologically functionalized gold Nanoparticles - A new Integrated Biosensor System

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Introduction

Prostate cancer is the most common male-specific cancer observed in the European Union and is the second leading cause of cancer death in men in our industrialized countries. The choice of treatment and its efficiency is largely dependent on the stage and on the degree of advancement of the cancer when it is diagnosed. Screening procedures like digital rectal examination (DRE) and free prostate specific antigen (PSA) level testing are well established but lack accuracy, yielding only 80% of prostate cancers diagnosed in an early state. By providing a more accurate and precise tool for diagnosing prostate cancer in its early stages, the percentage of curable cancer patients would increase radically.

Principle of the Adonis Project

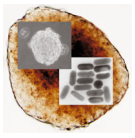
The **ADONIS Project** intends to prove the concept of using optoacoustic imaging in combination with biologically functionalised nanoparticles as an integrated biosensor based system for the production of specific and sensitive data for accurate diagnosis of prostate cancer.

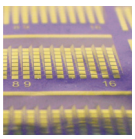
The achievement of this objective requires excellent know-how in several fields:

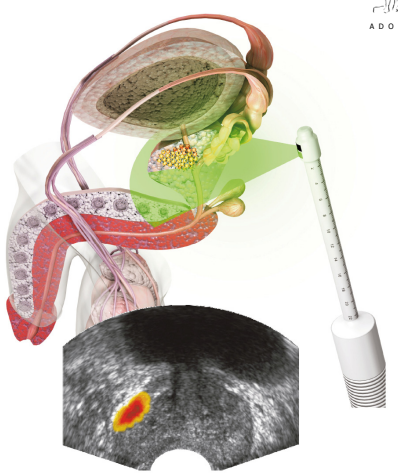
Laser Technology
A specialised laser excitation method for generating sound from gold nanoparticles allows the penetration of tissue segments of several centimeters thickness by infrared laser light.

Bio-Functionalization of Nano-sized Gold particles
for specifically and selectively binding pathological tissue as well as providing targeted delivery of these particles to the region of interest in the body.

Ultrasound Technologies and Image Reconstruction
capable of detecting the location and quantity of nano sized particles using the sound signals they generate following laser light excitation. Data processing and presentation of images for accurate visualisation of pathological regions.







Development and Perspectives

To allow the detection optimization of the biosensor, a 3D cell culture technique (spheroid culture) was developed to be closest to the *in vivo* tumor aspect. LNCaP, a human prostate carcinoma cell line which is reported to express PSMA, is the cellular model used for this work. This culture is performed in the Rotating Cell Culture System (Synthecon, Inc.) and produces spheroids or aggregates with sizes from 1mm up to 1cm. Observation of these entire spheroids under phase contrast microscope (*Fig.1*) and histological slices (*Fig.2*) show that these aggregates are composed of a tissue layer surrounding a necrotic core.

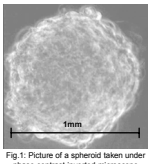


Fig.1: Picture of a spheroid taken under phase contrast inverted microscope (one week old).

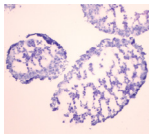


Fig.2: Hematoxylin/Eosin staining of small spheroids of ~1mm sizes.

The development of the biosensor is firstly performed to target the Prostate Specific Membrane Antigen (PSMA), a transmembrane protein considered as a suitable biomarker by its increased expression with prostate cancers progression and metastasis (70% in benign epithelium and 80% in invasive cancer cells). Detection of PSMA on LNCaP cell surface by immunostainning was performed on spheroid slices and on monolayer culture (*Fig.3 and 4*).

In prospect, following the validation of the bio-functionalization of the produced nano-sized gold particles, the detection of the completed biosensor will be tested on large spheroids and finally tested on *in vivo* model.

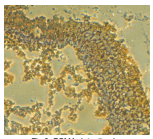


Fig.3: PSMA detection by ImmunoHistoChemistry using peroxidase on spheroid slice [spheroid of ~5mm size]. Spatial organization of PSMA-positive cells in the outermost cell layers and in the necrotic core were observed.

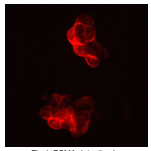


Fig.4: PSMA detection by ImmunoCytoChemistry using Qdots (Molecular Probes™) on monolayer culture (40X). Membrane distribution of PSMA is clearly visualized.

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