Molecular fluorescence imaging:

a new promising tool in surgical oncology

George Themelis, Ph.D⁴, Athanasios SarantopoulosB.Sc.¹, Niels J. Harlaar, M.D¹²³, Gooitzen M. van Dam M.D/Ph.D²³, VasilisNtziachristos M.Sc./Ph.D¹

1) Chair for Biological Imaging & Institute for Medical and Biological Imaging, TechnischeUniversitätMünchen and Helmholtz ZentrumMünchen, Munich, Germany

2) Department of Surgery, and 3) BioOptical Imaging Center Groningen, University Medical Center Groningen, Groningen, The Netherlands

Corresponding author: George Themelis, Institute for Biological and Medical Imaging (IBMI), Helmholtz ZentrumMünchen, IngolstaedterLandstrasse 1, 85764 Neuherberg, Germany, telephone: +49-89-3187-1243, e-mail: george.themelis@helmholtz-muenchen.de

Abstract

Objective: To improve the surgical procedure and outcome by means of real-time molecular imaging feedback of tumor spread and margin delineation using targeted near-infrared fluorescent probes with specificity to tumor biomarkers.

Summary background data: Surgical excision of cancer is often confronted with difficulties in the identification of cancer spread and the accurate delineation of tumor margins. Currently, the assessment of tumor borders is afforded by post-operative pathology or, and less reliable, intraoperative frozen sectioning. Fluorescence imaging is a natural modality for intra-operative use, since it relates directly to the surgeon’s vision and offers highly attractive characteristics such as high-resolution, sensitivity and portability. Via the use of targeted probes it also becomes highly tumor specific and can lead to significant improvements in surgical procedures and outcome.
**Methods:** Mice bearing xenograft human breast tumors were injected with an $\alpha_v\beta_3$-integrin receptor targeted fluorescent probe and in-vivo visualized using a novel real-time multi-spectral fluorescence imaging system. Confirmatory ex-vivo imaging, bioluminescence imaging and histopathology were used to validate the in-vivo findings.

**Results:** Bioluminescence images were all in good correspondence with the fluorescence images in respect to anatomical localization. Fluorescence imaging detected all tumors and successfully guided total tumor excision by effectively detecting small tumor residuals, which occasionally were missed by the surgeon. Tumor tissue exhibited Target to Background Ratio (TBR) of ~4.0 which was significantly higher compared to white-light (WL) images representing the visual contrast. Histopathology confirmed the capability of the method to identify tumor negative margins with high specificity and better prediction rate compared to visual inspection.

**Conclusion:** Real-time multispectral fluorescence imaging using tumor specific molecular probes is a promising modality for tumor excision by offering real time feedback to the surgeon in the operating theatre.