HHS Public Access

Author manuscript

*J Nucl Med*. Author manuscript; available in PMC 2016 March 01.

Published in final edited form as:

*J Nucl Med*. 2016 January ; 57(1): 144–150. doi:10.2967/jnumed.115.158915.

# Successful Translation of Fluorescence Navigation During Oncologic Surgery: A Consensus Report

**Eben L Rosenthal**[1], **Jason M Warram**[1], **Esther de Boer**[1,2], **James P Basilion**[3], **Merrill A Biel**[4], **Matthew Bogyo**[5,6], **Michael Bouvet**[7], **Brian E Brigman**[8], **Yolonda L Colson**[9], **Steven R DeMeester**[10], **Geoffrey C Gurtner**[11], **Takeaki Ishizawa**[12], **Paula M Jacobs**[13], **Stijn Keereweer**[14], **Joseph C Liao**[15], **Quyen T Nguyen**[16], **Jim Olson**[17,18], **Keith D Paulsen**[19], **Dwaine Rieves**[20], **Baran D Sumer**[21], **Michael F Tweedle**[22], **Alexander L Vahrmeijer**[23], **Jamey P Weichert**[24], **Brian C Wilson**[25,26], **Michael R Zenn**[27], **Kurt R Zinn**[28], and **Gooitzen M van Dam**[29]

[1]Division of Otolaryngology, department of Surgery, University of Alabama at Birmingham, Birmingham, AL, USA [2]Department of Surgery, University Medical Center Groningen, University of Groningen, The Netherlands [3]Case Center for Imaging Research, Department of Radiology Case Western Reserve University, Cleveland, OH, USA [4]Department of Otolaryngology-Head and Neck Surgery, University of Minnesota, Minneapolis, MN, USA [5]Department of Pathology, Stanford School of Medicine, Stanford, CA, USA [6]Department of Cancer Biology Program, Stanford School of Medicine, Stanford, CA, USA [7]Department of Surgery, University of California San Diego, La Jolla, CA, USA [8]Division of Orthopaedic Surgery, Department of Surgery, Duke University Medical Center, Durham, NC, USA [9]Division of Thoracic Surgery, Brigham and Women's Hospital, Boston, MA, USA [10]Department of Surgery, Keck School of Medicine of the University of Southern California, Los Angeles, CA, USA [11]Hagey Laboratory for Pediatric Regenerative Medicine, Department of Surgery, Division of Plastic and Reconstructive Surgery, Stanford University School of Medicine, Stanford, CA, USA [12]Artificial Organ and Transplantation Surgery Division, Department of Surgery, Graduate School of Medicine, University of Tokyo, Tokyo, Japan [13]Cancer Imaging Program, DCTD, NCI, Bethesda, MD, USA [14]Department of Otorhinolaryngology Head and Neck Surgery, Erasmus Medical Center, Rotterdam, The Netherlands [15]Department of Urology and Bio-X Program, Stanford University School of Medicine, Stanford, CA, USA [16]Division of Head and Neck Surgery, University of California San Diego, San Diego, CA, USA [17]Clinical Research Division, Fred Hutchinson Cancer Research Center, Seattle, WA, USA [18]Seattle Children's Hospital & Regional Medical Center, Seattle, WA, USA [19]Department of Diagnostic Radiology, Geisel School of Medicine, Dartmouth College, Hanover, NH, USA [20]Washington, DC, USA [21]Department of Otolaryngology, Simmons Comprehensive Cancer Center, University of Texas Southwestern Medical Center, Dallas, TX, USA [22]Department of Radiology, The Wright Center for Innovation in Biomedical Imaging, The Ohio State University, Columbus, OH, USA [23]Department of Surgery, Leiden, University Medical Center, Leiden, The Netherlands [24]Department of Radiology, University of Wisconsin, Madison, WI, USA [25]Princess Margaret Cancer Centre, UHN, Toronto, Ontario, Canada, USA [26]Medical

Communicating Author: Eben L Rosenthal, MD, Division of Otolaryngology, Boshell Building 563, Birmingham, AL 35233, Tel: (205) 934-9766, Fax: (205) 934-3993, erosenthal@uabmc.edu.

Biophysics, University of Toronto, Toronto, Ontario, Canada [27]Plastic and Reconstructive Surgery, Duke University Medical Center, Durham, NC, USA [28]Department of Radiology, University of Alabama at Birmingham, Birmingham, AL USA [29]Department of Surgery, Nuclear Medicine and Molecular Imaging and Intensive Care, University Medical Center Groningen, University of Groningen, The Netherlands, on behalf of the American Society of Image Guided Surgery (ASIGS)

## Abstract

Navigation with fluorescence guidance is emerging as a promising strategy to improve the efficacy of oncologic surgery in the last decade. The onus is on the surgical community to objectively assess the added value of this technique for routine use daily clinical practice, which will directly impact both the Food and Drug Administration (FDA) approval process and insurance reimbursement. In addition, it is critical to characterize the potential benefits over existing practices and to elucidate both the costs and safety risks. This report is the result a consensus meeting of the American Society of Image Guided Surgery (ASIGS) on February 6th, 2015 at Miami, Florida and reflects a consensus of the participant's opinions. Our objective is to critically evaluate the platform technology and its optical imaging agents and make recommendations for successful clinical trial development for clinical implementation of this highly promising approach in oncologic surgery.

## INTRODUCTION

While the field of surgery has recently experienced tremendous advances in optical technologies and robotics, one area that has remained constant is the dependence of the surgeon on visual and palpable cues that differentiate diseased versus healthy tissue, with its inherent limitations in sensitivity and specificity. Reliance on white light limits the visual contrast available to the operating surgeon to a narrow dynamic range in the colorimetric spectrum. Consequently, the ability to identify subclinical and deep seated disease states, during oncologic surgery is difficult, and the surgeon must rely on non-specific visual changes and manual palpation of subtle irregularities to guide successful excision without any reliable real-time feedback on its efficacy. The most common method of intraoperative margin control remains frozen section analysis, however this technique is time intensive and can sample only a small fraction of the wound bed, with even a degree of false-negatives. Given that the primary treatment modality for most solid tumors is radical surgery and since positive margins (defined as tumor cells present at the cut edge of the surgical specimen) are associated with increased local recurrence and indicate poor prognoses, real-time intraoperative distinction between tumor and normal tissue is urgently needed to improve surgical outcomes, and simultaneously prevent under- and overtreatment with its accompanying morbidity of vital structures.

Conventional anatomical imaging modalities, such as MRI, have been adopted for use in the operating room. Unfortunately, these are neither real-time nor tumor specific, costly and cannot be applied easily in the surgical field of view. Use of optical imaging for cancer-

specific navigation has been successfully introduced in glioma surgery with improvement in outcomes by using a fluorescent agent 5-ALA [1–2]. These findings demonstrate that optical imaging can be successfully applied to oncologic surgery. However, as this technique is approved already in Europe and is advancing towards routine use in the US, future clinical trials in the field of image-guided surgery will need to be designed in a way that rigorously evaluates the added benefit for patients while also assessing the cost effectiveness. Unlike the introduction of a new drug for the treatment of cancer, surgical trials evaluating fluorescence-guided resection present unique hurdles such as lack of standardization, difficulty in randomization, and variations in how surgeons currently determine normal vs. tumor interface during surgery. It is important to recognize that the lower rate of return compared to therapeutics which is anticipated from introduction of an imaging agent to the market place, it is critical for the surgical community to address these items early in the regulatory and approval process.

To accomplish these goals, the American Society of Image-Guided Surgery held a consensus meeting in February 2015 to discuss regulatory pathways, clinical trial design, and patient safety. Attendees included an international assembly of surgeons, scientists, and regulatory administrators who cooperatively addressed specific issues facing the translation of this technology. The objective of this meeting was to identify optimal routes for regulatory approval and successful trial outcomes. The general consensus of the meeting attendees concerning these topics is reported herein, which may serve as a standardized guidance for navigating the regulatory process and designing successful clinical trials in fluorescence-guided surgery for oncologic resection.

## REGULATORY PATHWAYS AND OBTAINING AN IND

Early phase clinical trials will need to establish safety of the contrast agent as well as the accompanying imaging device. Imaging trials designed to evaluate safety typically administer small, diagnostic doses and therefore are less concerned with drug-induced toxicity. However, dose-independent toxicity, such as immunogenic reactions, represents a low incidence, but a significant risk and therefore can be difficult to detect in a limited enrollment, early phase trial. Conventional Phase I studies utilizing dose escalation to identify a maximally tolerated dose (MTD) are commonly applied to therapeutic drugs. This approach is not necessary for imaging agents. When diseased-specific contrast is the objective, reducing the uptake in normal tissues is just as critical as increasing the disease-specific uptake. Therefore, dosing should be scaled to determine optimal delineation of cancer compared to normal tissue, and the optimal contrast may not necessarily correlate with increasing dose. Early phase clinical trials should be designed to identify optimal dose and timing of imaging. In contrast to radionuclides, which emit high-energy photons with very little tissue attenuation, optical probes are subject to greater attenuation and therefore require greater doses to achieve suitable contrast. Considering this the consensus was that a dose escalation study is preferred over a microdosing scheme for early trials evaluating the safety and efficacy of an optical imaging agent.

## Exploratory IND (eIND) or Microdosing

In 2005, to ameliorate the significant demand of resources and time required for full Investigational New Drug (IND) applications, the Food and Drug Administration (FDA) implemented a subpharmaceutical single microdose (1/100 of standard clinical dose) or low dose (<30nmol) regulation under an exploratory IND (eIND or phase 0) [3–4]. The primary reason to obtain an eIND, as opposed going through the full IND process, is to accelerate first in human experience at a lower cost to obtain proof of concept data early in development. If the study is successful at the microdose, then additional toxicology will need to be performed for a full IND application. The eIND study can be initiated with less or very different preclinical support compared to phase I studies that seek dose-limiting endpoints. Trial design for eIND studies need to have limited human exposure and have no therapeutic or diagnostic endpoints. Guidance documents about obtaining an eIND can be found at (http://www.fda.gov/downloads/drugs/guidancecomplianceregulatoryinformation/guidances/ucm078933.pdf).

## IND-Enabling Toxicology

Submission of a full IND requires that the safety and efficacy can be demonstrated. The process of compiling an IND for a new imaging agent is well beyond the scope of this document, however there are several aspects to the development of a cancer specific fluorescent imaging agent worth noting. Nonclinical toxicology studies should be designed around pharmacology (mechanism of action and secondary effects), pharmacokinetics (PK parameters and differences in gender), and safety (effects on major organ systems). Doses in nonclinical studies should significantly exceed expected clinical doses and usually require at least 3 drug concentrations (or possibly a single high dose). Although generally a rodent and nonrodent species are selected for these studies, the test species must be pharmacologically responsive or have the appropriate antigen specificity to the proposed study drug. Agents with unique toxicities that may be less dose sensitive in humans, such as complex proteins (allergic response) or antigen targeting, may require use of non-human primates. The formulation of the study drug for toxicology does not need to meet the standard of clinical studies, however it should be nearly equivalent to ensure the results are transferable. The imaging guidance documents are found at (http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm092895.htm)

The FDA pre-IND consultation program provides a unique opportunity to test proposed pathways with the FDA review divisions where they may provide guidance on the data necessary to warrant IND submission. The pre-IND meeting is the ideal opportunity to obtain feedback on an optical imaging agent development plan that is specifically designed to maximize safety and limit expenditure of time and resources required for a successful IND application. A productive pre-IND meeting requires that the investigators have a comprehensive toxicology, manufacturing and clinical protocol plan in place that the FDA will answer specific questions about the suitability of the strategy, such as ‘are the proposed ECG monitoring time points sufficient?’ Traditionally the FDA will not provide input to open ended questions such as ‘What physiological monitoring should be performed during the toxicology experiments?’ Therefore, a fully prepared and mature development plan is

critical to provide the greatest benefit to the sponsoring party. It is suggested that the pre-IND meeting be conducted in person to allow for clear interaction around critical questions.

Fluorescent labeling of approved molecular agents, such as therapeutic antibodies, can be translated to the clinic with less toxicology studies compared to new agents [5] – also include L 1: Warram JM, de Boer E, Sorace AG, Chung TK, Kim H, Pleijhuis RG, van Dam GM, Rosenthal EL. Antibody-based imaging strategies for cancer. Cancer Metastasis Rev. 2014 Sep;33(2–3):809–22. doi: 10.1007/s10555-014-9505-5. PubMed PMID:24913898; PubMed Central PMCID: PMC4116453.. The purpose of the toxicology studies in this setting is to demonstrate that the fluorescently labeled agent has the same toxicity and pharmacokinetic profile as the unlabeled, approved agent. When fluorescently labeling the targeting agent, a low dye to protein ratio [6] favors similar clearance rates and limited change in the antigenicity of the molecule. Although the intellectual property issues surrounding these agents remains complex, successful clinical translation is more efficient and safe, which also makes these agents ideal from the perspective of the FDA. Once an IND has been successfully opened, additional patients on the same protocol or new protocols investigating a variety of cancer types can be added to the IND using the same drug product. The FDA must be notified of such changes and all such changes would need approval from an IRB.

## Pairing of Imaging Device and Agent

New imaging devices require FDA review and approval by the PreMarket Approval process (PMA, devices can be found at http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfPMA/pma.cfm). The 510(K) premarket submission mechanism allows an FDA review and approval based on the “substantial equivalence” of a new device, as compared with an already approved device granted by a PMA (date base= http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpmn/pmnsimplesearch.cfm). An imaging device and the medical imaging drug (optical contrast agent) may be paired for the FDA approval process and clinical marketing of this technology. The FDA uses the term “combination product” in reference to this pairing.

If a device can be used with more than one fluorophore, currently or prospectively, it does not have to be a combinational device. However, under circumstances where an imaging agent can only be imaged using a specific device, the agent and device are paired and classified as a combination product. To expand the range of devices and potential imaging agents that could be used interchangeably in the future, the general consensus from this meeting was that if a device can successfully image a fluorophore, then it can be applied to any targeting agent linked to that fluorophore. There is also a general consensus that if a device can successfully image a specific range of wavelengths, it can be used to image any fluorophore with excitation/emission spectrum that fell within that range. However, it is important to note that at the current time it is not certain if fluorescence imaging agents and devices will or will not require pairing for the FDA review and approval process. Currently, indocyanine green is paired with specific devices. Furthermore, it remains unclear how industry will view an open format device approach compared to a combination device strategy.

An FDA cleared device with established installation base can be used for clinical trials in order to expedite the approval process and potential translation of optical imaging agents. However, this may not be efficacious if the device is not optimized for the imaging spectrum of the adapted fluorophore. Furthermore, repurposed devices my not be ideal for the indication due to poor ergonomics, size limitations, and constrained integrated software. In general, optical imaging devices are considered low risk or non-significant risk and the safety concerns of the devices will be considered soley as a combination device in another section. The FDA has clear documentation on the process for IND-enabling toxicology studies and the IND application process (http://www.fda.gov/Drugs/DevelopmentApprovalProcess/ucm090361.htm).

## CLINICAL TRIAL DESIGN

### Early Phase Clinical Endpoints

The focus of early phase clinical trials should be on safety of the agent. Secondary end points would include identification of the appropriate drug dose and timing for surgical intervention. Preclinical work to define optimal dose/time or biodistribution will not translate unless the animal model is syngeneic. Optimal dose should be defined by toxicities along with the optimal tumor-to-background ratio (TBR) required to differentiate diseased from normal tissue. The general consensus is a TBR greater than 2 is thought to be clinically useful. The examination of tumor and background intensity may also be necessary during acquisitions at the lower end of detection. Therapeutic clinical trials are usually focused on identification of the dose limiting toxicities, however in optical imaging of cancer, the highest optimal tumor to background ratio is considered mutually important. In some cases, it is imagined that higher doses below the dose-limiting toxicity (DLT) may not translate necessarily to greater TBRs due to non-specific uptake in adjacent normal tissue. Ultimately, the optimal dose and timing will be target specific rather than fluorescence-specific. These parameters are highly dependent upon the PK of the agent and the physiology of the target. For example, targets in the stromal compartment can exhibit contrasting characteristics to targets that are membrane-bound. Additionally, the optimal dose and timing can depend on the formulation of the targeting agent; a full antibody PK is different than an activatable probe.

A viable option for determining optimal timing may involve radiolabeling of targeting agents followed by microdosing in patients. Single photon radionuclides such as In-111 or positron emitters such as Zr89, are already used in patients and can be easily added to targeting moieties followed by SPECT or PET imaging to determine whole body biodistribution and optimal timing. Off target accumulation of the agent would be useful in designing safety monitoring for clinical trials. However, this has less application when considering cleavable peptides. This would involve a separate IND for each imaging agent, however microdosing with radionuclides would permit the use of an eIND, which would require less resources and time to submit. Ultimately, the information gleaned from a phase 0 study using nuclear imaging will permit more efficient dosing schedule and clear safety parameters during a phase 1 study rather than focusing time and resources on logistical discoveries such as optimal time of surgery.

If these optical imaging agents are to be approved for disease assessment or diagnostic management, successful implementation of optical imaging to the routine management of cancer operations will require clear demonstration of patient benefit and clinical usefulness. There are both long-term and short-term endpoints that can serve to identify the potential success of optical imaging in oncologic surgery. The cost and follow-up of longer end-points are often difficult to implement in early stage clinical trials, but are also important to consider.

### Acceptable toxicity

Diagnostic imaging agents are often held to a very high standard for safety because they are commonly given to a greater number of patients at regular intervals for a range of disease types, including some with very benign outcomes. In chemotherapeutic trials with cyotoxic agents, an acceptable toxicity is usually a grade 1 or 2 reaction related to the study drug. Traditionally, anything over a grade 1 reaction is considered unacceptable for imaging agents. In light of this, unique considerations should be granted for an intra-operative imaging agent since the disease is life threatening and the patient is undergoing a major procedure usually under general anesthesia. The general consensus is that a limited number of grade 2 adverse events would be acceptable because these agents are only given to those patients with known tumors undergoing an invasive procedure rather than a broad population of patients undergoing purely diagnostic procedures. It should be noted that this applies primarily to tumors requiring aggressive surgical intervention that have a significantly high risk of mortality, compared to low risk procedures where there are limited risks associated with the intervention. Patients should be followed for four times the known half-life of the drug product to ensure that all possibly related safety events are captured.

### Patient Number

For early observational trials, the general consensus is that first in human trials should normally have less than thirty patients. Although certainly more patients could be included (consider up to 50), there should be stopping rules built into the protocol. For late phase trials, sufficiently powered patient numbers will depend upon which metric the field considers clinically relevant. The general consensus was that agencies would be concerned about consistency of side effects within a wide demographic. Therefore, phase III trials need to be multicenter site performed by cooperative groups so the sample size must be large enough for such analysis using standardized fluorescence readouts of the imaging devices in order to create generalizable data sets throughout the study population.

### Phase I Trials

Although early phase trials should focus on safety and dosing, future trials will be initiated based on data collected during these early examinations. Apart from safety data, the success of the imaging strategy (agent and device) can be evaluated in several ways. First, sensitivity and specificity of the intraoperative imaging strategy can be calculated relative to surgeon assessment and pathological assessment. Additional statistical analysis, as directed by the FDA guidance on medical imaging agents, may include likelihood ratios and receiver operator characteristic curves. Secondly, the TBR will provide information on the power of the strategy to provide sufficient contrast for disease delineation. Thirdly, the specificity of

the agent and device to accurately demarcate the disease border can be measured by correlating the fluorescence margin with the disease margin. In cases where a stable fluorescent probe agent is used, the agent will survive pathological processing and fluorescence microscopy can be used to correlate fluorescence with histological evidence of tumor on H&E stained sections. Furthermore, immunohistochemistry can be performed to map specific tumor antigens with fluorescence intensity to determine successful drug penetration and targeting.

With the complexities of surgical trials noted above and the existence of FDA Guidance documents defining clinical usefulness as the ability to distinguish normal from diseased tissue, the most critical endpoint is demonstrating whether the presence of fluorescence is specific for cancer. Correlating the fluorescence during surgical imaging presents a greater challenge due to the detailed mapping that is required to trace the fluorescence edge through the formalin fixation and histological processing. Considering the multiple indications optical imaging agents may ultimately be approved for, the common verification required for a cancer indication is demonstration that the imaging successfully delineates normal from abnormal tissue. The general consensus is that a common methodology can be introduced to accomplish this task and adopted by the field to standardize regulatory reporting.

Correlation of fluorescence with histological evidence of tumor is also confounded by the attenuation of tissue during imaging and mapping areas of fluorescence through the pathological process. Although detection of subclinical disease remains the primary objective of this imaging strategy, it should be recognized that it is very unlikely that this imaging technique will detect only a few hundred cells. Rather, the objective is to make an incremental improvement on the current limitation on the size of disease that can be reliably detected by intraoperative palpation and visual changes in the tissue. This is especially true for minimally invasive procedures where there is loss of tactile and optical feedback.

## Histology as the Gold Standard

To determine the sensitivity and specificity of the imaging agent, presence of disease must be confirmed using the current gold standard, which is histological analysis using H&E staining. However, the field of pathological evaluation of tumor specimens is subject to multiple inaccuracies including sampling error and loss of tissue orientation [7–9]. Correlating histological evidence of tumor with fluorescence is complicated by these limitations. This particularly applies to false positives where the presence of fluorescence within a large tissue mass may not be confirmed by histology because of a failure to fully sample the entire tumor, missing the very small region of tumor that was detected by fluorescence. It is not practical to serially section even a small tumor sample (e.g., 1 cm by 1 cm), which will need up to 2500 slides of 4 um and thus can be considered impractical and non-executable in daily routine. It may be necessary to use PCR-based assays to increase the sensitivity of the gold standard for microscopic disease.

## Thresholding

Fluorescence imaging, like PET imaging, can be thresholded along a continuum of intensity that must be standardized to an acceptable baseline. The general consensus is that in order for optical guided surgery to traverse federal regulation and advance to routine clinical use, there must be a widely adopted methodology for fluorescence assessment adopted within the field. For immediate identification of unknown samples in the operating room or pathology for a specific patient, the preferred methodology was to image the known cancer (tumor mass in situ) and known normal tissue to adjust the threshold to reveal diseased tissue apart from normal. This initializing of the threshold would be performed uniquely for each patient at the beginning of surgery. Appropriate thresholding would be performed based on the known samples, revealing the fluorescence intensity of the unknown tissue. This approach is considered optimal considering a fixed threshold is very difficult to establish due to differences in patients, tumors physiology, tissue properties, timing, molecular target expression, and clearance.

For additional standardization, relative quantification may be critical for objective assessment and reporting. Similar to standardized uptake value in assessment and reporting of PET imaging, absolute counts (fluorescence intensity) from unknown tissue and known normal tissue can be used to generate a ratio. Using this methodology, a ratiometric threshold for positive disease can be experimentally developed and integrated into the onboard device software to objectively identify disease tissue intraoperatively, in real-time. The general consensus is that this degree of standardized and objective assessment will be required by regulatory agencies in order to critically demonstrate the ability of the technique to assess disease.

## Advanced Phase Clinical Endpoints

Surgical resections are currently performed by obtaining a large margin of healthy tissue around the estimated tumor border in order to confirm negative margins. Consensus of the group was that the ability of this technology to differentiate normal from disease tissue should be the primary end point to identify clinical utility. However, clinical trials should also consider if this technique is superior to conventional white light assessment of the tumor margin. In highly invasive tumors (oral cavity squamous cell carcinoma or melanoma) the surgeon will add 1–2 centimeters to the palpable (or optical border) of the tumor. Delineation of the tumor extent in segmental or whole organ resections (lung, colon, or larynx) is less likely to find this technique valuable. Tumor biology will determine the definition of the margin as highly invasive or a pushing front.

In many cancer surgeries positive margins remain a challenge and are associated with poor outcomes. In the majority of cases, the outcome resulting from a single positive surgical margin is not mitigated successfully by subsequent surgery to clear the margin (i.e. re-excision of the positive margin) or adjuvant chemotherapy and/or radiation. The need for imaging to improve delineation of tumor and normal tissue is an obvious advantage to preventing incision through cancer, identifying suspicious or close margins, and guiding a consistent margin around the tumor. The value of such an imaging agent would be consistent with how the FDA views approval of such agents. According to the FDA Guidance

Documents for approval of imaging products Section 351(a) of the Public Health Service Act (42 U.S.C. 262(a)): “the ability to locate and outline normal structures or distinguish between normal and abnormal anatomy can speak for itself with respect to the clinical value of the information and will not require additional information substantiating clinical usefulness.”

In discussion amongst oncologic surgeons familiar with this technology, there were several concerns regarding the complexity of alternate clinical trial end points to show clinical benefit. Perhaps the most challenging aspect of clinical trial design in this setting is that the standard of care for excision remains surgeon assessment that is highly variable and subjective – ‘if it looks like cancer, cut it out.’ An imaging technology that is better than the accepted standard is likely to identify more positive margins compared to standard technique not less when implemented into routine procedures. It should be recognized that both functional outcomes and survival could be overshadowed in studies by post-operative adjuvant therapy, which is commonly performed in cases of positive margins. Furthermore, surgical outcomes are dependent on surgical technique where complete resection is balanced with functional or cosmetic outcome. Thus, survival depends on doing a radical resection balanced with functional outcome, however, oncologic outcomes without successful functional outcomes are meaningless.

There are other clinical trial endpoints that could be considered as secondary endpoints. These included retention of normal tissue, preservation of normal tissue function (i.e. nerves, ureters, lymphatics, vasculature), reduction of operative time and associated operating room costs, reduction in morbidities and complications related to prolonged general anesthesia, and change in rate of positive margins, reduction in the need for salvage surgery or adjuvant therapy when using the technique.

### Summary

The field of optical imaging for surgical guidance is rapidly expanding with the introduction of new agents and hardware that will transition into the market place over the next couple of years. Submission of an IND to investigate safety, molecular targeting and timing of surgery is the first step toward successful clinical implantation. As contrast-based optical imaging techniques are introduced into patients, a methodology for standardization of reporting is critical to achieve agent approval. The primary endpoint of initial clinical trials to define effectiveness should focus on the successful delineation of normal from abnormal tissues in order to identify clinical utility. Ongoing interactions with the FDA are necessary to determine the regulatory pathway for IND submission and eventually NDA approval.

## Acknowledgments

This document is the result of a consensus meeting held by the American Society of Image Guided Surgery (ASIGS) on February 6th, 2015 at Miami, Florida and reflects a synthesis of the participants opinions and concepts for the clinical translation and application of optical imaging in surgery.

## References


1. Stummer W, Novotny A, Stepp H, Goetz C, Bise K, Reulen HJ. Fluorescence-guided resection of glioblastoma multiforme by using 5-aminolevulinic acid-induced porphyrins: a prospective study in 52 consecutive patients. J Neurosurg. 2000; 93:1003–1013. [PubMed: 11117842]
2. Stummer W, Pichlmeier U, Meinel T, et al. Fluorescence-guided surgery with 5-aminolevulinic acid for resection of malignant glioma: a randomised controlled multicentre phase III trial. Lancet Oncol. 2006; 7:392–401. [PubMed: 16648043]
3. Bertino JS Jr, Greenberg HE, Reed MD. American College of Clinical P. American College of Clinical Pharmacology position statement on the use of microdosing in the drug development process. J Clin Pharmacol. 2007; 47:418–422. [PubMed: 17389550]
4. Sarapa N. Exploratory IND: a new regulatory strategy for early clinical drug development in the United States. Ernst Schering Res Found Workshop. 2007:151–163. [PubMed: 17117721]
5. Zinn KR, Korb M, Samuel S, et al. IND-Directed Safety and Biodistribution Study of Intravenously Injected Cetuximab-IRDye800 in Cynomolgus Macaques. Mol Imaging Biol. 2014
6. Cohen R, Stammes MA, de Roos IH, Stigter-van Walsum M, Visser GW, van Dongen GA. Inert coupling of IRDye800CW to monoclonal antibodies for clinical optical imaging of tumor targets. EJNMMI Res. 2011; 1:31. [PubMed: 22214225]
7. Basaran D, Salman MC, Boyraz G, et al. Accuracy of intraoperative frozen section in the evaluation of patients with adnexal mass: retrospective analysis of 748 cases with multivariate regression analysis. Pathol Oncol Res. 2015; 21:113–118. [PubMed: 24848925]
8. Chambers KJ, Kraft S, Emerick K. Evaluation of frozen section margins in high-risk cutaneous squamous cell carcinomas of the head and neck. Laryngoscope. 2015; 125:636–639. [PubMed: 25230253]
9. Novita G, Filassi JR, Ruiz CA, et al. Evaluation of frozen-section analysis of surgical margins in the treatment of breast cancer. Eur J Gynaecol Oncol. 2012; 33:498–501. [PubMed: 23185796]

**Limitations of Current Ablative Techniques**

- Excessive removal: Wide surgical margin of healthy normal appearing tissue to insure an adequate margin
- Time consuming: Frozen sections require significant delays in surgical practice and can be reversed on permanent section
- Conventional techniques no applicable to all tissues: Visual changes, palpation and frozen section analysis cannot be applied to all tissue types
- Healthy tissue must be resected for pathologist to determine negative margin: To confirm a negative margin by histological analysis, the tissue must be resected for analysis which can lead to poor cosmetic outcome, functional deficits or bleeding

**Considerations in Trial Design**

- Microdosing can be used to confirm target specificity but insufficient for intraoperative imaging
- It is currently unclear if the device and the drug product should be paired or general parameters for devices set for each drug product
- Dose and time ranging studies be performed in phase I clinical trial setting
- Acceptable toxicity for optical contrast agents for oncologic surgery should be between diagnostic and therapeutic agents.
- Grade 2 toxicity in 20% of the population is an acceptable threshold as a dose limiting toxicity.

**Considerations in Obtaining an IND**

- Submit eIND or IND
- Selection of species (agent dependent)
- Dose range
- Device selection

**Application of Optical imaging in operating room**

**In Situ Imaging**

- Goal to preserve normal tissue and completely excise tumor
- Enable the surgeon to accurately identify the tumor boarder
- Sample post resection wound bed
- Identify positive regional lymphatics

**Ex Vivo Imaging**

- Goal to improve sampling error
- Identify the tumor margins from primary specimen
- Evaluate margin samples obtained from the wound bed

**Table 1**

Ongoing clinical trials

| Biological agent | Trial phase | Identifier | Condition | Target | Device | PI | Sponsor |
|---|---|---|---|---|---|---|---|
| Bevacizumab - IRDye800CW | I | NCT01691391 | FAP* | VEGF | NIR fluorescence endoscopy | WB Nagengast | University Medical Centre Groningen |
| Bevacizumab - IRDye800CW | I | NCT01972373 | Rectal cancer | VEGF | NIR fluorescence endoscopy | WB Nagengast | University Medical Centre Groningen |
| Bevacizumab - IRDye800CW | I | NCT01508572 | Breast cancer | VEGF | | GM van Dam | University Medical Centre Groningen |
| Bevacizumab - IRDye800CW | I | NCT02129933 | Premalignant Esophageal lesions | VEGF | NIR fluorescence endoscopy | WB Nagengast | University Medical Centre Groningen |
| Cetuximab-IRDye800CW | I | NCT01987375 | Head and Neck cancer | EGFR | | EL Rosenthal | University of Alabama at Birmingham |
| MDX1201-A488 | Pilot | NCT02048150 | Prostate cancer | PMSA | | T Wilson | City of Hope Medical Center |
| ProstaFluor® | Observational | NCT01173146 | Prostate cancer | PMSA | | | Spectros Corporation |
| AVB-620 | | | Breast cancer | Proteases | | RY Tsien | UCSD |
| BLZ-100 | I | NCT02234297 | Glioma brain tumors | MMP-2 Annexin VII | Infrared imaging system | CG Patil | Blaze Bioscience Inc. |
| 5-ALA | I | NCT00870779 | High grade glioma, meningioma, pituitary adenoma or metastasis | | | DW Roberts K Paulsen | Dartmouth-Hitchcock Medical Center |
| 5-ALA | II | NCT01445691 | Brain tumors | | | C Hadjipanayis | Emory University |
| 5-ALA | I<br>II | NCT01128218 | Brain tumors | | | JW Cozzens | Southern Illinois University |
| 5-ALA | Observational | NCT01837225 | Breast cancer | | PRODIGI Imaging Device | WL Leong RS DaCosta | University Health Network, Toronto |
| 5-ALA | II | NCT00752323 | Brain tumors | | | A Sloan | Case Comprehensive Cancer Center |
| 5-ALA | I | NCT02191488 | Brain tumors | | | | Dartmouth-Hitchcock Medical Center |
| 5-ALA | III | NCT01502280 | Glioblastoma | | | N Sanai | St. Joseph's Hospital and Medical Center |
| 5-ALA | Observational | NCT02155452 | Glioblastoma | | | AV Moiyadi | Tata Memorial Hospital |

FAP=Familial Adenomatous Polyposis; ALA=aminolevulinic acid

## Table 2

| Exploratory IND | Traditional IND |
|---|---|
| • Only phase 0/I | • Drug development anticipated to advance to phase III |
| • No diagnostic/therapeutic endpoints | • Extensive preclinical data necessary |
| • More than one investigational drug considered | • May include diagnostic/therapeutic endpoints |
| • Rapid progress to humans for proof of concept | • Typically one investigational drug |