Workshop on imaging science development for cancer prevention and preemption

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Abstract. The concept of intraepithelial neoplasm (IEN) as a near-obligate precursor of cancers has generated opportunities to examine drug or device intervention strategies that may reverse or retard the sometimes lengthy process of carcinogenesis [92,93,144,189]. Chemopreventive agents with high therapeutic indices, well-monitored for efficacy and safety, are greatly needed, as is development of less invasive or minimally disruptive visualization and assessment methods to safely screen nominally healthy but at-risk patients, often for extended periods of time and at repeated intervals. Imaging devices, alone or in combination with anticancer drugs, may also provide novel interventions to treat or prevent precancer.

1. Introduction

Current advances in molecular imaging [40,91,176,196] and biomarker discovery for various cancers [124] enable earlier detection and diagnosis of recognizable
precancerous lesions (e.g., papillomas, nodules, etc.), possibly even differentiating fields of transformed cells from their normal tissue surroundings (Fig. 1). Indeed, increasing numbers of available imaging modalities and visualization tools (Fig. 2) have generated real questions regarding which instruments and image-driven interventions produce the most significant and cost-effective patient outcomes for the widest variety of cancer modalities.

Adopting specific imaging technologies as standards for oncologists, radiologists, and nuclear medicine specialists requires evidence-based demonstrations of sensitivity/specificity and predictive value with clinical precision for each instrument design. Significant competition is expected among imaging devices and biomarker assessment methodologies as each developer negotiates the hurdles and challenges along various engineering, clinical, and regulatory routes to the marketplace.

A workshop convened by the National Cancer Institute (NCI) Cancer Imaging Program in Gaithersburg, MD, provided an overview of these convergent issues and processes and facilitated discussion on the state of imaging science development for cancer prevention and preemption. Presentations by more than 50 researchers and translation scientists focused on instruments and techniques for evaluation of early cancer and image-guided intervention (IGI) in clinical settings. Keynote speakers presented introductions to issues surrounding the application of imaging techniques, including dis-
ease incidence and prevalence, pathology and mechanism of lesion development, epidemiology, risk factors and models, clinical presentation of disease, standard of care, ongoing and new clinical protocols, screening and early detection procedures, treatment interventions (drugs, surgery, IGI), biomarkers, and currently used imaging modalities. The keynote talks were followed by presentations on imaging research and roundtable discussions with experts from industry, academia, and government who considered priorities for further research and development efforts needed to qualify investigational devices or methods for clinical use.

Progress in endoscopic or other optical assessment methods and complementary approaches to biopsy and ablation of suspicious lesions were highlighted in several sessions on hollow organ or surface epithelial neoplasias (e.g., Barrett’s esophagus, superficial bladder cancer). Advances in instrumentation and development of biomarkers for precancer detection in solid organs (e.g., breast, prostate) were necessarily focused on remote or indirect imaging modalities such as computed tomography (CT), magnetic resonance imaging (MRI), positron emission tomography (PET), and ultrasound or thermal spectroscopy.

2. Summary of presentations

2.1. Barrett’s esophagus

Dr. Carlo Maley, Wistar Institute, described the evolution of Barrett’s esophagus or esophageal intestinal metaplasia and its association with chronic gastroesophageal reflux disease (GERD) [121]. Barrett’s esophagus has a risk of progression to esophageal adenocarcinoma ranging from 0.5–1%/year [21]; its incidence is increasing faster than any other cancer in the Western world, perhaps related to the epidemic of obesity and attendant GERD seen in many developed countries. Current intervention standards include proton pump inhibitor therapy for reflux and esophagectomy if frank carcinoma is identified. Aberrant clones within the pre-malignant Barrett’s mucosa may cover complex concave shapes on the surface of the distal esophagus. Diagnosis and surveillance is based on extensive random biopsy sampling methods, typically performed every 2 cm down the Barrett’s segment under direct endoscopic visualization. Careful prospective analysis of biopsy material has found a correlation between risk of progression to cancer and extent of aneuploidy and loss of heterozygosity (LOH) of various tumor suppres-
sor genes, describing a risk pathway predictive of patient outcomes [120,163]. Genetic heterogeneity within Barrett’s mucosa can be ranked in time and space from an initial lesion showing p16 LOH with a relatively low-risk ratio progressing to p53 LOH (which has the highest risk ratio for development of carcinoma), then aneuploidy. Better methods to image proliferative or mutant clones would permit less onerous biopsy sampling, a better understanding of how clones expand, as well as the potential to intervene before higher-risk lesions progress.

Dr. Prateek Sharma, University of Kansas, has applied just such a series of innovations to Barrett’s esophagus assessment, using in vivo chromoendoscopic tissue staining, magnification and high-definition endoscopy, and narrow-band imaging (NBI) filters, to better define regions of dysplasia. Conventional white-light inspection of Barrett’s segments can be enhanced by applying Lugol’s iodine, indigo carmine, or methylene blue spray to the esophageal surface to distinguish complex and often subtle patterns of intestinal metaplasia at its junction with normal squamous surface epithelium. Videendoscopes equipped with a charge-coupled device (CCD) chip use high pixel densities to produce high image resolution. Furthermore, with a movable higher magnification lens (35–115X power), endoscopists can evaluate microscopic mucosal features within the regions of Barrett’s metaplasia, such as raised villiform or tubular patterns, well correlated with risk of progression [41,179,180]. Higher-intensity blue-light illumination with special NBI spectral filters enables imaging of both superficial epithelial and deeper tissue features such as capillary vascular patterns within regions of interest. NBI can be used without tissue dye augmentation with the detection of blood-vessel hemoglobin as the primary chromophore; it has high sensitivity and specificity in distinguishing high-grade dysplasia (HGD) in Barrett’s disease [87,177]. Novel imaging techniques can potentially help identify neoplastic areas in Barrett’s endoscopy; a banding suction device then can be used to resect suspicious areas of dysplasia under direct narrow-band visualization.

Real-time optical biopsy of Barrett’s lesions would avoid the trauma and delay associated with current conventional histopathologic analysis. Dr. Adam Wax, Duke University, described several proof-of-concept approaches to this problem, which involve diagnosing disease while evaluating intact living tissue at the cellular level. Low-coherence interferometry (LCI) combined with oblique angle detection of scattered light (a/LCI) detects precancerous changes based upon epithelial nuclear size and texture at various tissue depths without sectioning, fixation, or applying exogenous contrast agents. Signal discrimination parameters for a/LCI have been worked out for various nuclear characteristics of both cancer and precancer cell lines in vitro [220] as well as in an in vivo animal carcinogenesis model [221], where reversal of precancerous epithelial changes with an apoptosis-inducing chemopreventive agent impacted a/LCI-detected nuclear features. Ex vivo evaluation of resected Barrett’s specimens by a/LCI scanning of up to 20 × 10 cm areas of mucosa showed a marked correlation between tissue nuclear atypia classified as diseased or normal by a/LCI, and tissue dysplasia grades assigned by histopathology. Measurements of large tissue surface areas that once took 30 minutes to perform can now be performed in 40 milliseconds due to the rapidity and sensitivity of current a/LCI fiber optic probes with a laser scanning spectrometer and improved processing power [159]. A portable instrument system with these new design features has the potential to become a clinically relevant measurement and diagnostic tool.

Dr. Gregory Ginsburg, University of Pennsylvania, reviewed current clinical standards for observational management of Barrett’s esophagus and the variety of endoluminal therapies now available as alternatives to surgery. Individuals with long-segment disease (1–7% of Barrett’s population) have greater risk for progression to cancer compared to those with short-segment disease (6–17%) [57,75]. If HGD is detected during surveillance biopsy, 30% of patients typically show an intramucosal cancer if esophagectomy is performed, and 16–60% develop cancer from 5–7 years after diagnosis and without further management [24,164,173]. However, 40–80% of Barrett’s patients with HGD don’t progress, with ~50% remaining stable and ~25% regressing to lower-grade disease. Because of this wide variability in outcomes and current lack of precision in risk stratification, endoscopic ablation or mucosal resection therapies may be preferable to esophagectomy. Before considering superficial ablative therapies, it is advisable to evaluate the esophagus by methods such as endoscopic ultrasound in order to rule out disruptions in the mucosal wall or submucosal adenopathy that might indicate more invasive disease [175]. ablative endoluminal therapies include thermal and cryotherapy approaches and cytotoxic approaches such as photodynamic therapy with photofrin/aminolevulinic acid (ALA). Thermal ablation instruments include direct-contact electrocautery probes or circumferential radiofrequency (RF) balloon electrodes (e.g., HALO [360].
from BARRx, Inc.), and non-contact systems such as laser or argon plasma beam coagulators. These instruments are relatively safe and unlikely to cause perforations, but carry the risk of leaving residual “buried Barrett’s” under regenerating mucosa [136]. PDT with photofrin or ALA taken up preferentially by dividing cells provides a potentially more specific method of delayed necroinflammatory ablation. A pivotal trial that resulted in FDA approval showed the PDT technique to be highly efficacious in eliminating circumferential long-segment Barrett’s disease with multi-focal HGD, thereby reducing progression to cancer during 2–5 years follow-up [146]. Complications such as postoperative stricture were not markedly different than those seen for other thermal ablative methods; however, some skin photosensitization by the dyes, and areas of PDT “skip” requiring multiple treatment sessions, were noted. All ablative methods have the disadvantage of destroying mucosa that might provide valuable histopathologic diagnostic information. Because of this, focal or short-segment Barrett’s is often better managed with instruments that permit endoluminal mucosal resection (EMR) [3], such as the suction/snare device described by Dr. Sharma. Dr. Ginsburg’s unpublished research indicates that multimodal endoluminal therapeutic approaches combining techniques such as PDT with EMR may be optimal in complex cases.

2.2. Bladder cancer

The bladder is another hollow organ prone to develop superficial tumors within a readily accessible mucosa. Dr. Edward Messing, University of Rochester, provided an overview of suspected disease etiology (smoking, chemical/industrial exposures) and some demographic risk factors (\( > 2:1 \) male/female ratio, median age of 70 at presentation). Currently, no well-accepted urine biomarker exists for the earliest forms of bladder carcinogenesis, usually detected through patient self-reporting of hematuria or irritative voiding [133,139].

Confirmatory diagnosis of transitional cell carcinoma (TCC) is then made by cystoscopic examination of the bladder. TCC is staged through pathologic assessment of papillary tumors recovered by transurethral resection (TURBT) or biopsy of more extensive areas of TCC \textit{in situ} (TIS). Single low-grade superficial (Ta, T1) TCC, with a low rate of progression to muscle invasion (5–10%), but a high rate of recurrence, is typically managed by surveillance cystoscopy every three months with TURBT as needed. Higher-grade lesions (stage Ta, T1, TIS) have a 20–70% progression rate requiring intravesical therapy with antimitotics or immunomodulatory agents such as \textit{Bacillus Calmette-Guerin} (BCG) as an adjunct or alternative to TURBT. Muscle-invasive disease is usually treated by cystectomy. Using intraoperative near-infrared imaging to trace infusions of fluorescent dyes such as indocyanin green that are taken up by the cavernous nerves in the pelvic plexus may be able to spare the complication of nerve damage-induced impotence often associated with this procedure.

Application of optical fluorescence to improve detection and outcome of TCC was presented by Dr. H. Barton Grossman, MD Anderson Cancer Center. Although current standard white-light cystoscopy is highly reliable in detecting bladder cancer, certain TIS lesions are visually difficult to detect and residual tumors may often remain after TURBT. Intravesical infusion of dyes such as ALA that make TCC more visible when illuminated by blue light have the potential to improve the diagnostic sensitivity of cystoscopy. Several clinical trials comparing ALA fluorescence to white-light cystoscopy found a reduced risk of recurrence or progression when TURBT was performed with ALA [9, 48]. Recent development of a hexyl ester of ALA, HAL (Hexvix®), has made adoption of fluorescence microscopy even more practical. HAL instillation reduces the amount of time required for dye uptake by a bladder tumor to one hour. In several clinical studies [85,171], HAL increased TCC detection sensitivity to 96% compared to 77% for white-light cystoscopy. False-positive regions of normal mucosa, noted with HAL as well as ALA, were produced by non-specific fluorescence seen with areas of post-BCG inflammation. Increasing the interval after intravesical therapy should permit resolution of this inflammation and increase specificity of the fluorescence technique.

Bladder cancer that has advanced to invade muscle requires radical cystectomy (described above) as well as extensive dissection of the surrounding pelvis, including removal of draining lymph nodes. Successful identification and resection of nodes has a direct survival benefit for these patients [73]. Standard imaging modalities like CT and MRI rely primarily on node size to detect metastasis, despite the considerable overlap in size between benign and malignant nodes. Attempting to distinguish between involved and uninvolved nodes using MRI contrast enhancement agents such as gadolinium, and metabolic imaging methods such as \textit{\( ^{18}\)fluoro-2-deoxyglucose} positron emission tomography (FDG-PET), are usually confounded by urinary concentration of tracer in
the adjacent bladder. Dr. Mukesh Harisinghani, Massachusetts General Hospital, reviewed his experience with a novel MRI nanoparticle contrast agent to delineate and stage nodal involvement in bladder cancer. These ultrasmall (21 nm) dextran-coated supermagnetic iron oxide particles (ferumoxtran-Combidex) show contrast uptake and decreased signal intensity on T2-weighted MRI images as a result of their uptake by normal lymph node macrophages (i.e., “black is benign”). In contrast, areas of lymph node infiltrated with tumor cells show space-filling regions without nanoparticle uptake and retain their high-signal-intensity MRI density. An initial clinical trial of 48 patients scheduled for cystectomy [35] recovered 172 lymph nodes after ferumoxtran infusion imaging; this MRI contrast-enhancement method yielded sensitivity and specificity of 95% and 95% when compared to routine histopathologic diagnosis, resulting in a negative predictive value for patient outcome of 100% and a positive predictive value of 90%. Whether this contrast agent clears rapidly enough from uninvolved nodes to permit accurate follow-up of response monitoring is still questionable, as is whether its initial uptake is confounded by lymph node fibrosis, produced by chemotherapeutic agents and radiotherapy, which may be more routinely observed for cancers in other organs. At present, the agent has not been approved by FDA.

Dr. Irving Bigio, Boston University, reviewed the concept of noninvasive optical tissue biopsy and described various spectroscopic methods applicable to bladder cancer; most common are UV-induced fluorescence of exogenous agents and autofluorescence spectroscopy, along with Raman and infrared reflectance techniques. Dr. Bigio and his colleagues are currently working on elastic scattering spectroscopic (ESS) approaches which, when performed using appropriate fiberoptic instruments, are point-sensitive to morphologic tissue architectural variations at cellular and subcellular levels. Although not directly applicable to construction of tissue images, ESS can provide a backscatter signal spectrum that differentiates between tissue histomorphometry characteristics of benign, cancer-adjacent, and malignant bladder mucosa after appropriate computational image analysis [137]. Complex spectra generated by ESS relate to the spatial variations of the indices of refraction displayed by all cellular components, including water, proteins, lipids, and DNA, when comprising higher-order structures such as membrane bilayers and cytoplasm that include cell organelles and nuclei. Morphological changes in these cellular structures during carcinogenesis, including shifts in nucleus-to-cytoplasm ratio, nuclear shape factors, chromatin distribution, and organelle and inclusion arrangement are characteristic of all pathologic processes and generate variations in photon-scattering spectra that, collected in a sufficient database, enable identification of disease margins and can improve accuracy when selecting regions of transformed bladder mucosa for resection or ablation.

2.3. Breast cancer

Dr. Carol Fabian, University of Kansas, summarized the current state of the art for risk assessment and breast cancer early detection. Screening mammography has now become routine for women in the US; breast density pattern analysis using breast imaging, reporting, and data systems (BI-RADS) can add additional discriminatory power to the score derived from the common Gail risk assessment tool [53,205] used to advise women regarding their risk of cancer development. Components of the Gail risk formula (age, reproductive history, familial risk, atypical biopsy results, etc.) that result in a calculation of >1.67% estimate of cancer development within five years cause an assessment of high risk. Similarly, >75% breast density (BI-RADS 4) indicates a greater than five-fold increase in five-year risk relative to those with the lowest BI-RADS score. Additional cytomorphologic monitoring may be indicated for these high-risk individuals, including nipple aspirate fluid and ductal lavage sampling, or random fine-needle aspiration (FNA) [43,178,225]. New molecular or tissue histomorphometric biomarker methods (e.g., immunocytochemical staining for proliferative antigens or hormone receptors, automated quantitative nuclear morphometry) are being examined to reduce the interpretive variance seen with standard cytology index scoring systems (Masood score). Risk assessment formula, breast density measurements, and cytomorphicologic screening procedures have all been incorporated into cancer prevention trial designs to provide high-risk cohort selection and monitor effects of intervention with cancer-preventive drugs. For example, tamoxifen substantially reduced mammographic density at least in younger women, most likely reflecting epithelial stroma content of breast tissue and impact of the antiestrogen on glandular and ductal epithelial structures at risk. More recently, newer selective estrogen receptor modulators (SERMs) such as arzoxifene, and estrogen synthesis inhibitors such as aromatase inhibitors (letrozole, etc.), are under evaluation as cancer preventive agents using breast density, cytomorphicologic,
and other molecular biomarker measurement methods (e.g., insulin-like growth factor (IGF)-1/IGF-BP3 ratio) to assess safety and efficacy [42,97]. However, standard CT methods to determine breast density are not optimal for risk assessment and treatment monitoring in every setting. Younger, very high-risk women (e.g., those with BRCA1/2 mutations), who opt for prophylactic surgery, would benefit from a modality that does not use radiation, which may in itself increase the risk of transformation events in the breast. MRI and ultrasound approaches appear fruitful. Older and post-menopausal women with low breast density due to adipose tissue replacing stroma may still be at considerable risk for cancer development; imaging modalities such as thermal or transillumination (TLM) spectroscopy [181] may better predict vascular changes associated with incipient tumor formation.

Dr. Katherine Ferrara, University of California at Davis, described the role of ultrasound in breast tumor imaging, the prospects for enhancing its sensitivity and specificity by addition of contrast agents, and its potential in IGI. Specifically, the addition of “microbubble” contrast agents to current standard ultrasound techniques used in differential diagnosis of suspicious lesions and in guiding biopsy needle placement could improve tumor resolution and capacity to identify tumor vascular volume and flow [47]. These gas-filled (lipid-encapsulated) ultrasound-detectable microbubbles show enhanced regions of viable tumor with high integrated blood flow and an increased extent of perfusion that is well correlated with CT-based estimates and confirmed with histology. Decreased tumor flow parameters document microbubble-enhanced ultrasound imaging response to treatment with antiangiogenic agents. The promise of this innovative technology was illustrated by a series of experiments providing proof-of-concept for molecular imaging and drug delivery applications. Ligand-specific targeting of liposome-encapsulated fluorocarbon gas microbubbles provided a two- to three-fold increase in tumor vascular localization identifiable by ultrasound when compared to control microbubble treatments. When ultrasound is applied, labeled microbubbles are concentrated up to 30-fold in targeted vessels in vitro and up to six-fold in vivo. Deflection of the ligand-bearing bubble away from the vascular flow onto the vessel wall by the ultrasound force pulse is thought to be the mechanism behind this concentration phenomenon. By further increasing the ultrasound force, drug- or gene-loaded nanoparticles or multiplayer liposome shells could undergo enhanced extravasation into the tumor parenchyma rather than passive infiltration/diffusion from the tumor vascular bed. This insonification technique is thought to deliver more drug, and, therefore, potentiate cell death within the region of ultrasound treatment. In a preclinical model, rats with bilateral subcutaneous tumor implants infused with paclitaxel-containing nanoparticles showed decreased tumor growth rate on their insonated side. Development of new, high-power, multi-modality ultrasound transducer probe arrays to permit simultaneous or sequential imaging, radiation force, or fragmentative operation modes, will be important in bringing this new technology to the clinic.

Dr. Sergio Fantini, Tufts University, discussed near-infrared spectral imaging for breast cancer. Optical imaging has been used to identify and display the vascular network of the breast, usually by exploiting the absorption spectrum of hemoglobin (690 nm) that provides spatial separation from other normal breast components such as lipids and water. This technology makes increased vessel density at a tumor site apparent. Images can be enhanced by increasing density of two-dimensional arrays of illumination sources and detectors, or by using a phased-array intensity approach to achieve increased depth discrimination. In addition, multiple wavelength information from this system can potentially assess mammary tumor oxygenation [63] by examining hemoglobin oxygen saturation spectra at 750–856 nm. Tumor hypoxia increases in more aggressive tumors that may be largely dependent on anaerobic metabolism for growth. As a result of modeling the optical spectral signal at various levels of hemoglobin oxygenation, the next-generation instrument will image oximetry data largely independent of tumor size, shape, and depth within the breast.

Dr. Mitchell Schnall, University of Pennsylvania, reviewed use of MRI to detect and direct appropriate therapy in breast cancer, and compared its sensitivity to that of ultrasound and mammography. As confirmed by biopsy in several single and multicenter studies, MRI, especially when used with contrast enhancement, is superior to other imaging methods in detecting multicentric and multifocal disease [13,101,108,218]. Instances in which other modalities out-performed MRI included 12% of cancers that did not show contrast enhancement, typically DCIS lesions or invasive cancers identified through calcification. Lack of MRI enhancement still had a 94% negative predictive value for the presence of invasive cancer [172]. MRI can improve identification of candidates for breast-conserving therapy by eliminating those with multifocal lesions beyond the primary tumor field [78] and providing guidance on un-
dertaking adjunctive chemohormonal or radiation therapy. Another application of MRI is in guiding treatment planning and targeting focal breast tumor ablation therapy. Focused ultrasound (FUS) provides a non-invasive thermal ablation effect within breast tissue by using sonication to raise tissue temperatures, creating a circumscribed area of tumor necrosis while sparing external and adjacent normal tissues. Real-time MRI can monitor tissue-heating progress and follow-up MRI can estimate the extent and success of FUS-induced necrosis. In an initial clinical trial, results predicted by MRI visualization were highly correlated with biopsy findings from the ablation site [58,94].

2.4. Prostate cancer

Dr. Eric Klein, The Cleveland Clinic, provided an overview of current incidence and prevalence data on prostate cancer, shared some current concepts regarding risk factors and etiology of the disease, and detailed available or pending findings from recent large screening and intervention trials with the potential to influence standard of care and development of imaging technology.

In the Western world, one in seven men older than 70 will develop prostate cancer. Though prostate-specific antigen (PSA) screening means most men will now be diagnosed with early disease (85% not palpable on digital rectal exam (DRE), fewer grade 8–10 Gleason score tumors, etc.), significant debate still exists regarding which tumors are biologically significant and clinically relevant. Infection and/or inflammation are hypothesized to be the basis for prostate carcinogenesis, whether driven by an infectious agent [209] or by dietary or environmental exposure to substances that cause oxidative stress and chronic inflammation [140]. A number of prostate cancer susceptibility genes control or regulate pathways associated with cellular defense; intervention with protective agents such as non-steroidal antiinflammatory (NSAIDs) drugs or free-radical scavenging drugs hold promise for primary prevention or as adjuncts to other therapies. Managing more advanced, extraglandular disease, or biochemical failure after radical prostatectomy or external beam radiation, remains problematic; challenges remain in imaging disease stage and progression associated with these distinct classes of cases [99,187]. A wealth of fundamental data soon to be available from large screening trials (e.g., European Organization of Research and Treatment of Cancer (EORTC), or Prostate, Lung, Colorectal, and Ovarian Cancer (PLCO)), and implications for prevention science from recent findings of hormone antagonist studies such as the Prostate Cancer Prevention Trial (PCPT) [204] and toremifene [157,190], should stimulate new ways of thinking about treatment and diagnosis in urology and imaging communities.

Dr. Angelo De Marzo, Johns Hopkins University, detailed pathological considerations in establishing prostate cancer diagnoses and the need for precision imaging procedures in this process. More than a million trans-rectal ultrasound (TRUS)-guided biopsies are performed in the US every year, typically as a result of DRE and/or PSA elevation. Although sophisticated prostate volumetric mapping tools can more accurately direct placement of multiple-needle core biopsies into transitional, peripheral, and central prostate zones (thought to have differential risks of tumor development), this blind sampling often misses known cancer lesions. Progression to adenocarcinoma is now believed to originate in precursor lesions such as proliferative inflammatory atrophy (PIA), proceeding through high-grade prostatic intraepithelial neoplasia (HGPIN) to higher and higher Gleason grade cancers [34,140]. Evidence for this progression model has been bolstered by morphological mapping studies, immunohistochemical studies, and molecular biology studies [34]. All stages of carcinogenesis may be seen in a single core biopsy or a series of cores. A diagnosis of cancer (~10–30% of all biopsies) then requires determination of features such as tumor type, Gleason score for each positive core, number of positive cores, and percent of core involvement before treatment strategies (hormonal therapy, prostatectomy, radiation) can be recommended. Imaging modalities that could identify highly suspicious areas within the prostate would be very helpful for directing the location in which the biopsy cores are taken. Additional quantitative tissue histomorphometry and nuclear morphology derived from image analysis would improve the overall assessment of tumor presence/absence, extent, and Gleason grade [214]. Better accuracy in determination of disease status and severity would allow for more informed choices regarding treatment or watchful waiting and thus could reduce unnecessary procedures and treatments caused by over-diagnosis.

Among imaging methods, dynamic contrast-enhanced (DCE) MRI with 3 Tesla scanners has potential for improving diagnosis and prognostic accuracy in prostate cancer. Dr. Iclal Ocak of the NCI Molecular Imaging Program discussed the use of MRI techniques in evaluating disease, noting that the hetero-
geneity of disease ranging from very indolent to highly aggressive presents a quandary for therapy selection that may be addressed with improved imaging methods. Addition of the endorectal coil to the 3 Tesla scanner imaging protocol increases the signal available, reduces noise from other anatomical sites, and most patients tolerate it well. These new generation 3 Tesla MR scanners, used with dynamic contrast material injection have increased sensitivity and specificity of the T2-weighted image with improved signal to noise ratios [32]. Prostate and other tumor vasculature often has an altered morphology and increased permeability relative to surrounding normal tissue parenchyma [128]. As a result, tumor area shows early wash-in and early wash-out of contrast agent due to increased permeability and leakiness of pathologic tumor vessels. DCE-MRI with the current SENSE (sensitivity encoding) protocol [210] correlated with biopsy or prostatectomy data holds promise for developing a tool to distinguish tumor foci by detection of early enhancement and early clearance of contrast agent from the extravesicular extracellular tumor microenvironment. Magnetic resonance spectroscopy (MRS) has also shown great potential in the localization, staging, and assessment of aggressiveness of prostate cancer, which is characterized by reduced levels of citrate and polyamines, and increased level of choline in proton MRSI (magnetic resonance spectroscopy imaging). The differences in concentration of metabolites have been attributed to increased phospholipid turnover associated with increased tumor cell proliferation and increased cellularity within the tumor foci [31]. The higher field scanners also have advantages for MRSI in prostate cancer detection due to higher spectral/spatial resolution and decreased temporal resolution compared with lower field scanners. The increase in spectral resolution at 3 Tesla using an endorectal coil may offer better separation of choline, polyamine, and creatine peaks that overlap considerably at lower Tesla. Thus, MRSI has the potential to significantly improve metabolic assessment of prostate cancer. MRI with an endorectal coil at 3 Tesla remains to be the most promising tool for stratification of patients toward “watchful waiting,” standard therapy, or more aggressive treatment.

Because ultrasound and MRS approaches sense fundamentally different properties within prostate tissue (mechanical and chemical), combining the two techniques could be synergistic for improving classification of prostate cancer risk. Dr. Jeffery Ketterling discussed collaboration between groups at Riverside Research Institute in New York City and Virginia Mason Medical Center in Seattle. Using a clinical ultrasound data acquisition system, they are building a database of images to be correlated with clinical case data and histologic “truth” from biopsy and resection specimen archives. Serving as a teaching set, their neural-network computer system incorporates data from 617 biopsies (83% negative and 17% positive) derived from 64 patients (64% benign diagnosis and 36% cancer), along with relevant clinical data (age, PSA, etc.) and baseline B-mode ultrasound scans. Subtle differences in tissue histology microarchitecture displayed as unique scattering properties on ultrasound are believed to be correlated with and predictive of biopsy outcome. Incorporation of these factors in the current teaching set has enhanced discrimination between cancerous and noncancerous tissue with a receiver operating characteristic (ROC) curve area of 0.84 compared to 0.64 obtained by ultrasound assessment alone. Similar ROC curve areas have been obtained for tumor area classification with citrate/choline MRSI [86]. Current efforts attempt to register ultrasound and MRS data into three-dimensional data sets and correlate combined tissue-type profiles with prostatectomy histology.

Dr. Clare Tempany, Harvard Medical School, spoke on evolution of MRI-guided diagnosis and therapy from brachytherapy seed placement developed in the late 1990s [33] to current state-of-the-art techniques. Real-time intraoperative image fusion and open-interventional magnet designs (e.g., GE Healthcare) with MRI-compatible needles and anesthesia now guide robot-assisted prostate biopsy that is more accurate and reliable than TRUS procedures. This system allows for biopsies with MR guidance from multiparametric images such as H1 Proton spectroscopy or DCE-MRI. As in breast cancer ablative therapy, FUS is also seen as a non-invasive treatment alternative to prostate cancer radiotherapy and surgical treatments. MR-guided FUS approaches to prostate cancer follow many of the same design principles learned from earlier studies on ultrasound surgery for uterine leiomyomas [203], where beam paths need to be calculated to avoid bowel and bone, and staged rounds of sonication are used to obtain optimal heating using MR thermometry to guide and directly monitor in real-time the effective thermal ablation. Research is underway to develop an MR-guided FUS device for prostate cancer ablation. Trials have been performed in Europe with over 460 men with low-risk localized prostate cancer to be enrolled in the Ablatherm multicenter trial, which will build on preceding successful early-phase investigations [55,119,207] as evidence needed for regulato-
ry approval for the Ablatherm instrument system and procedure. Dr. Tempany concluded her presentation by describing the advanced multi-modal image-guided operating room with 3T MRI, PET/CT, and procedure room (AMIGO GEHC) soon to be installed at Brigham and Women’s Hospital.

2.5. Ovarian and pancreatic cancer

Drs. Michael Birrer, NCI Center for Cancer Research, and Margaret Tempero, University of California at San Francisco, reviewed cancers of the ovary and pancreas, some of the most deadly and difficult to detect. The occult nature and late-stage presentation of these cancers, and their lack of response to most current forms of intervention, are in part due to a dearth of reliable systemic biomarkers and good imaging alternatives for early detection. In ovarian cancer, five-year survival for stage I disease is 90%, yet 70% of diagnoses occur at later stages of disease where survival drops precipitously. Although certain high-risk groups of women (those with family history or BRCA 1/2 mutations) have been identified, vague symptoms and no identifiable ovarian cancer precursor lesion confront public health practitioners and oncologists with the daunting prospect of mass population screening for a disease with low prevalence. The best available imaging approach to date, transvaginal ultrasound/sonography (TVS), still lacks sensitivity and specificity, though it can be somewhat improved by adding CA125 serum marker to the screen [182]. Among 22,000 postmenopausal women randomized to observation or annual CA125 testing followed by TVS screening in 77,000 women is pending as part of the NCI, PLCO study.

Several incremental improvements and alternatives to these standard methods for imaging adnexal masses in women have also been evaluated. Adding Doppler imaging to TVS has been explored for its ability to detect tumor-specific angiogenesis and increased blood flow. A meta-analysis of a large number of studies found that TVS morphology combined with Doppler imaging is significantly better at differentiating ovarian cancer from benign conditions (e.g., dermoid cyst, endometriosis, cystoma, pelvic inflammatory disease) than TVS alone [98]. Three-dimensional (3D) reconstruction and multiplanar volume-rendering displays of ultrasound data improve definition of adnexal masses and their relationship to internal anatomical structures. Data also support increasing specificity by confirmatory use of other imaging modalities such as CT or MRI after initial TVS/Doppler screening [102]. PET and SPECT scanning methods with metabolic (\(^{18}\)F-FDG), proliferative (\(^{18}\)F-fluorothymidine), or hypoxia (misonidazole) tracers have also been assessed for identifying ovarian cancer in both clinical and preclinical studies [69,112,131,194].

A similar lack of early symptoms, early invasion and metastasis, and chemoresistance characterize pancreatic cancer; more than 80% of patients are diagnosed with advanced or unresectable disease. With a cure rate of only 4–5%, and a median survival of only about three months for patients without treatment, pancreatic cancer was described by Dr. Tempero as the Mount Everest of cancer diagnosis and treatment problems. Some hints for screening strategies have come from epidemiologic associations shown for smoking, diabetes, and obesity, and genetic factors such as hereditary pancreatitis, BRCA2, HNPCC, and Li-Fraumeni syndromes. Due to significant advances in understanding the molecular pathogenesis of pancreatic cancer, a candidate pancreatic intraepithelial neoplastic (PanIN) precursor has been proposed [80]. Various oncogene activation or overexpression events (e.g., K-ras, epidermal growth factor receptor (EGFR)), tumor suppressor pathway mutations or derangements (p53, p16, SMAD), and other processes in the microenvironment affecting in-
vasion and metastasis (angiogenesis, desmoplasia, inflammation, etc.) in pancreatic cancer provide opportunities to apply these discoveries to improve early detection and treatment. Using appropriate risk stratification, it might be possible to deploy endoscopic ultrasound for early surveillance, followed by endoscopic retrograde cholangiopancreatography. As we identify more cases of early stage disease, we can build a biorepository of serum and normal DNA, pancreatic duct juice, and preinvasive and invasive tumor tissue for molecular and proteomic analysis. The uniquely hypoxic microenvironment of pancreatic cancer also suggests ways to exploit this phenomenon to develop imaging probes. New and better animal models of pancreatic cancer [2,74] provide test environments to evaluate these rationally designed diagnostic and treatment candidates.

2.6. Lung cancer

Dr. Stephen Lam, British Columbia Cancer Agency, introduced an examination of lung cancer by describing sequential histological stages seen during lung carcinogenesis, characterized by variations in nuclear size, shape, and staining properties of epithelial cells. Pathologically, lesions are graded as squamous metaplasia, mild dysplasia, moderate dysplasia, severe dysplasia, carcinoma in situ, and cancer; dysplasia level correlates with malignant potential. Numerous genetic changes occur in addition to histopathological changes. Studies have shown that 1.2–3.5% of moderate dysplasia, 8–50% of severe dysplasia, and 25–100% of carcinoma in situ will progress to invasive cancer within two years [18,20,79,83,104,135,170,215].

The epithelial surface of the lung is complex, consisting of a branching system of airways leading to gas exchange units with surface area the size of a tennis court. Lung cancer consists of several cell types that are preferentially located in different parts of the bronchial tree. No imaging modalities can visualize the whole surface of the lung and allow tissue sampling for pathological diagnosis and molecular profiling. Autofluorescence bronchoscopy has provided the first good method for detecting high-grade intraepithelial neoplasia (IEN) and obtaining biopsies in the central airways. However, biopsy may completely remove small lesions, giving the false impression that drug intervention has regressed IEN or that the majority of IEN lesions regress spontaneously.

Progress has been made in developing other optical imaging methods to characterize these lesions without removing them. Confocal microendoscopy, a high-speed scanning system with an ultra-sensitive detector and near-video frame rate, can be combined with autofluorescence to allow visual differentiation of the border between normal and abnormal epithelium [18]. Reflectance confocal microendoscopy allows cell-by-cell delineation [118]. Finally, large surfaces of epithelia can be scanned simultaneously using optical coherence tomography (OCT) [206] (see also under Skin Cancer).

Important challenges still exist in detecting and preventing neoplastic lung lesions, including identifying and managing peripheral adenocarcinoma. CT of the chest is a sensitive method to detect peripheral IEN lesions beyond the visual range of current fiberoptic bronchoscopes. Recently developed external navigational systems make it feasible to biopsy the peripheral airway under CT guidance, resulting in a better understanding of progression pathways of non-squamous lesions. Coupling microendoscopy with CT imaging may also be useful for following peripheral lesion progression non-invasively.

Twenty-five percent of cured lung cancer patients will get a second cancer in five years. Detection of early lesions does not necessarily discriminate which are premalignant. A possible solution would find nodules in at-risk patients, determine which are malignant versus benign using optical techniques, and ablate malignant nodules. The process would be repeated every few years.

Potential advances in lung cancer treatment include use of inhalational chemotherapy, possibly combined with imaging techniques to determine if treatment has reached target lesions.

Dr. Eric J. Seibel, University of Washington, described two new instruments under development that use current technologies for in vitro lung cancer screening and IGIs: optical projection tomography microscopy (OPTM) and scanning fiber endoscopy (SFE). OPTM generates 3D images with submicron and isometric resolution of cells and nuclei from cell suspensions injected into a rotating capillary tube. This multi-perspective image collection allows automated detection of nuclear abnormalities based on 3D tomographic reconstruction. The technology is similar to CT, using optics instead of x-rays. Ultimately, this instrument approach will be applied to image whole cells isolated from sputum samples and FNA.

SFE generates images in vivo using an ultra-thin and flexible catheter-like probe. Red, green, and blue (RGB) laser light (635, 532, and 442 nm) are combined into a single-mode optical fiber, the distal tip of which
is driven at mechanical resonance of 5 kHz. The fiber scanner generates 500-line images at 15 Hz by scanning the RGB laser light in a spiral pattern from the distal tip. The backscattered light is collected from the 80° scanned field by a ring of 12 plastic optical fibers that surround the fiber scanner. The current prototype probe is 1.6 mm in diameter. By using alternative light-collection methods, future prototypes will be as small as 1.0 mm in diameter.

A custom guidance system being tested in pigs introduces SFE through the working channel of a standard bronchoscope while being directed by a guide wire. The interactive 3D user interface has been developed to perform five functions: Automated segmentation and virtual rendering of airways from a chest CT scan; virtual bronchoscopic navigation of airway anatomy and path-planning for biopsy; real-time tracking and display of the endoscope tip using an electromagnetic sensor; cell sampling in the more peripheral lung under direct video imaging; and automated annotation of procedural events, including 3D path, optical diagnoses, and biopsy locations. SFE probe components are low in cost, allowing high-volume manufacturing and disposability.

Dr. Pierre Massion, Vanderbilt University, discussed approaches to solving one of the biggest problems in lung cancer research: Why do only 10–15% of smokers, and only 8% of patients with lung dysplasia, go on to develop lung cancer? Genomic and proteomic technologies may improve our understanding of this disease by allowing rapid and complete analysis of genome and proteome expressed in a given cell or tissue type. For cancer, differential profiling can determine if the genomic and/or proteomic profile of a set of cancers differs from a set of normal tissues or from one another. In addition, genome and proteome expression profiles have the potential to improve clinical management of lung cancer by improving classification or providing data to develop a diagnostic classifier or identify new molecular diagnostic and therapeutic targets [64].

Matrix-assisted laser desorption ionization mass spectrometry (MALDI-MS) creates molecular weight-annotated patterns key to tissue location, providing proteomic profiles that allow clear delineation of high-grade versus low-grade dysplasia, and preinvasive versus invasive neoplasia [160,191,226]. MALDI-MS profiling may help determine which preinvasive lesions are likely to develop into lung cancer. Potential clinical applications include biomarker discovery, tumor biology, and validation of targets found using other imaging technologies.

Areas of further development include increasing the number of proteins analyzed and pixel-to-pixel cycle time, integrating bioinformatic tools and image processing, and developing sample protocols targeted at specific tissues/diseases. Further bioinformatics development is also needed to improve rapid protein identification protocols, create a test pattern recognition algorithm to classify samples, and validate biomarker candidates prospectively in case-cohort studies.

Dr. Geoffrey McLennan, University of Iowa, reminded the audience that although biopsy is the gold standard for diagnosing lung cancer, information about a patient’s lung cancer risk obtained from biopsy is often incomplete. The paradox of lung cancer is that small peripheral lung cancers can be found by multidetector computed tomography (MDCT), but not diagnosed easily. The natural history of small peripheral lung cancers remains uncertain and therapeutic options are unsatisfactory. Most detected nodules suspicious for lung cancer represent noncancerous disease. Unlike other organs, the lung presents specific imaging problems because of motion associated with breathing and cardiac oscillation in the left lower lung. A further barrier to developing better imaging methods for lung nodules is the current lack of understanding regarding their pathological structure and function as related to 3D imaging [46]. Determining how much of a lung nodule is cancer and how it is organized appears critical for further development of more specific imaging and treatment strategies.

External magnetic guidance allows access to nearly 100% of peripheral nodules using a combination of technologies. Digital imaging allows rapid fine manipulation into a precise location within the lesion through magnetic guidance. Device placement can also be verified through magnetic tracking, with secondary fluoroscopic verification. Confocal or other microscopy can allow macro- or micro-optical sampling of the lesion. Finally, if the lesion is considered possibly malignant, ablation can be performed. Magnetic guidance can also stabilize a device for real-time microscopy. These methods have all been developed, and some have been tested in clinical research studies [68,174].

Dr. Kevin Cleary, Georgetown University, discussed the challenges of biopsying the lung, including pneumothorax, cumbersome Z-frame and remote center of motion (RCM), cardiac motion, and ribs in the path of needle insertion. Robotic devices are capable of tracking lesions and compensating for lung motion in real time with lower radiation doses to physicians and higher accuracy. In addition, robotic systems have the
A collaboration between the Computer Aided Interventions and Medical Robotics (CAIMR) group at Georgetown University, the Urology Robotics Laboratory at Johns Hopkins Bayview Medical Center, and the Department of Radiology at the University of Maryland Medical Center, is developing a robotic system to assist the physician in accurate CT fluoroscopy-guided needle biopsy of lung nodules. Completed first-phase testing focused on demonstrating the feasibility of using a joystick-controlled robotic “needle driver” to accurately hit simulated lesions in a phantom under CT fluoroscopy guidance. Initial results showed feasibility of motion tracking. However, current robotic technology is not capable of releasing the needle. The second phase involves developing an enhanced gripper and new end-effector that spins and releases the needle to minimize the force needed for needle placement. This phase is currently under development and will be tested first in phantom studies and then animal studies.

Dr. Robert Suh, University of California, Los Angeles, discussed the use of percutaneous transthoracic needle biopsy (TNB) for diagnosing pulmonary nodules. It allows previously inaccessible lesions to be sampled and tissue to be retrieved for histologic diagnosis [100]. Image guidance techniques, i.e., fluoroscopy, ultrasound, CT, and CT fluoroscopy, can be combined with TNB to improve accuracy of biopsy and treatment.

For example, only approximately 15% of patients diagnosed with lung cancer are surgical candidates. Though some patients have technically resectable disease, they cannot undergo surgical resection because of co-morbid cardiopulmonary disease. This latter patient population represents a suitable target for novel, minimally invasive “lung-sparing” therapies that could provide local control. Several minimally invasive image-guided thermal ablation techniques treat cancer by applying intense heat through a small probe inserted directly into the tumor.

Types of thermal energy include RF, microwave, laser, and cryoaBLation. RF ablation has been the best developed, secondary to the advent of bipolar, multi-electrode, and internal tip-cooling RF electrodes [109]. Pulmonary RF ablation allows safe thermal ablation of pulmonary metastases with a low complication rate and acceptable tumor control rate [216]. Percutaneous microwave therapy is also a safe new treatment for lung cancer, achieving a marked effect with minimum trauma [45]. Percutaneous cryoablation therapy for metastatic lung tumors is feasible and minimally invasive, with satisfactory local control [90].

Further areas of development for thermal ablation in lung cancer diagnosis and treatment include use of neo-adjuvant and adjuvant therapies (e.g., radiotherapy, brachytherapy, and chemotherapy), generating additional data through prospective and randomized trials designed to improve patient selection, refining image guidance and energy delivery, and providing post-thermal ablation follow-up.

Gene expression profiles of tumors may predict treatment response and outcome. A recent study was performed of molecular signatures in lung cancer biopsy specimens obtained from 23 patients undergoing CT-guided transthoracic biopsy or endobronchial brushing for undiagnosed nodules [17]. It was hypothesized that profiles derived from lung tumor biopsies would discriminate tumor-specific gene signatures and provide predictive information about outcome. Class prediction models for lung cancer histology and for cancer outcome produced a histology model that identified 99 genes differentially expressed by lung cancer subtypes and an outcome model that identified 42 genes associated with high risk for cancer death within 12 months. It was concluded that gene expression profiles of lung cancer biopsy specimens identify unique tumoral signatures that provide information about tissue morphology and prognosis. Specimens acquired by image-guided lung biopsy, used to identify biomarkers of clinical outcome, may assist in managing care of lung cancer patients. Prospective clinical trials are needed to determine efficacy and utility.

2.7. Colon cancer

Dr. Randall Burt, University of Utah, presented an overview of colon cancer etiology and current opportunities for its detection and treatment. About one in 17 persons in the US will get colon cancer in their lifetime. Risk factors include advanced age, ulcerative colitis, previous adenoma or cancer diagnosis, a high red meat/high fat diet, pelvic irradiation, alcohol or cigarette use, obesity, tall stature, cholecystectomy, and high sucrose consumption [26,71,114,156]. Colon cancer risk may be reduced 20–30% with a high vegetable/fruit diet, a high-fiber diet, high folate/methionine intake, high calcium intake, or hormone replacement, but prospective intervention studies with several of these agents have shown that vitamins C and E, selenium, β-carotene, and high-fiber, low-fat, optimal-nutrition diets provide no protection against
developing colon cancer [71]. However, calcium, aspirin, and cyclooxygenase (COX)-2 inhibitors provide significant protection; risk reduction of at least 60% is associated with physical activity or aspirin/NSAID use [6,15,71,123].

Most colon cancers arise from adenomatous polyps. Polyp-to-cancer evolution can take 10 to 20 years; therefore, most colon cancer screening is actually colon adenoma screening. The risk of an adenoma developing into cancer depends on polyp size, shape, and histology.

While the majority of colon cancers arise sporadically, up to 33% may be inherited, but with less than 5% associated with rare colon cancer syndromes [26,114]. Understanding the molecular pathogenesis of colon cancer began with discovering the adenomatous polyposis coli (APC) gene, a tumor suppressor gene that is mutated in familial adenomatous polyposis (FAP), a rare inherited syndrome. Somatic APC mutations are also found; >80% of all colon cancer may result from an initial APC gene mutation. Polyp-to-malignancy progression involves mutation of multiple additional genes, including K-ras, p53 and others [156]. Another rare syndrome, hereditary nonpolyposis colorectal cancer (HNPCC), arises from mutations of mismatch repair (MMR) genes. Fifteen per cent of sporadic colon cancers also exhibit somatic inactivation of MMR genes.

Symptoms of colon cancer range from rectal bleeding and abdominal pain to changes in bowel habits, weight loss, and decreased appetite. Symptoms are unusual in stage I or II cancer; but if detected in those stages, chances of survival are >80%. However, in stage III or IV, symptoms are common and the chances of survival are <30%. Overall five-year survival is now 64%, up from 50% in 1974.

Current screening guidelines for average-risk persons age 50 and older include annual fecal occult blood testing (FOBT), flexible sigmoidoscopy every five years, barium enema every five to 10 years, or colonoscopy every 10 years [162]. Individuals with a positive family history are recommended to begin screening at age 40 with any of the above tests. In the setting of a strong family history, defined as colon cancer in a first-degree relative at <50 years, or two affected first-degree relatives, screening should be done by colonoscopy, and begin at age 40 years or 10 years younger than the earliest diagnosis in the family. An extensive family history should be obtained if a strong family history is present in immediate relatives, and if indicated, genetic testing and special surveillance may be necessary.

Patient-acceptable, non-invasive screening for colon cancer is needed. Colonoscopy, though a great tool, requires more resources and trained gastroenterologists than will likely be available in the foreseeable future. Current advances in screening include immunohistochemical FOBT [228] and stool DNA testing [8]. Virtual colonoscopy is an exciting new technique which may become part of regular colon cancer screening in the future [138]. In this new procedure, CT images of the patient’s abdomen are used with a computer visualization system to virtually navigate within a constructed 3D model of the colon. This technology has the potential to be accurate, cost-effective, non-invasive, and comfortable for screening large segments of the population.

The standard of care for treatment for stages I–III colon cancer is surgery with adjuvant or neo-adjuvant chemo-radiation therapy for stages II and III rectal cancers, and adjuvant chemotherapy for stage III and selected cases of stage II colon cancer. The standard of care for stage IV colon cancer is chemotherapy with surgery in selected cases and adjuvant or neo-adjuvant chemo-radiation therapy if rectal. Fluorouracil/leucovorin with oxaliplatin or irinotecan (FOLFOX and FOLFIRI) is the most effective chemotherapy regimen for stage III and IV colon cancers [143,197]. The anti-EGFR and anti-vascular endothelial growth factor (VEGF) monoclonal antibodies cetuximab (Erbitux®) and bevacizumab (Avastin®), respectively, and oral fluoropyrimidines (e.g., capecitabine) are new treatment agents that appear to further improve colon cancer outcomes. In addition, agents under development include other anti-VEGF monoclonal antibodies, other anti-angiogenesis compounds, tyrosine kinase inhibitors (TKI), and anti-EGFR or anti-carcinoembryonic antigen vaccines [116].

One goal in studies of precancer and early cancer is to precisely isolate susceptibility, using currently available predictive biomarkers and imaging tools. At-risk patients would be identified through serum testing or imaging using a broad spectrum of genetic markers. However, specific markers not have been identified yet, and it is too early to tell which will be most important. Efforts in the field aim to develop beacon markers and biomarker panels. Patients with specific markers would then need aggressive screening using improved whole-body surveillance. Also needed are probes that would be useful for spatially locating lesions together with different types of validation methodologies. Recently, attention has been paid to biomarkers for prognosis
(methylation markers, CIMP, BRAF, MSI, BAX, TGF-$\beta_1$, SMAD, p53, and WAF1), diagnosis (Galectin-3 and NF1), and drug study (aberrant crypt foci) [19, 219].

Dr. Ruth Pfeiffer, NCI, described development of a colorectal cancer risk assessment tool [25,30,183–186]. A statistical model was developed to estimate the probability of first incidence of proximal, distal, and rectal cancer in white men and women ages 50 and older. Using logistic regression, relative risk was estimated separately for proximal, distal, and rectal cancer for a number of previously identified risk and protective factors (e.g., history of polyps, family history of colorectal cancer, smoking, body mass index, hormone replacement therapy, vegetable intake, vigorous exercise, regular aspirin/NSAID use, sigmoidoscopy in last 10 years). Data on white men and women ages $\geq 50$ was taken from two large population-based case-control colon cancer studies. Age-specific incidence rates from the Surveillance Epidemiology and End Results (SEER) study were used to compute baseline age-specific hazard rates. National mortality rates were used to estimate the hazard for competing causes of death. Relative risk estimates and risk factors differed among proximal, distal, and rectal cancer sites. Future plans include validation of the risk model in several large cohort studies, extension of the model to blacks and Hispanics using SEER baseline hazard rates, and distribution of a colorectal cancer risk assessment questionnaire for further testing and development [50].

Recently, researchers have been linking carcinogenesis and the Warburg effect, indicating that bioenergetics may lie at the heart of malignant transformation [54, 127]. It is now known that FDG uptake in colon, previously thought to be an incidental physiological finding, is associated with pathologic processes including malignancy in a majority of instances. Dr. John Hoffman, University of Utah, described several studies that confirmed that the presence of focal colonic FDG uptake on PET or PET/CT scan justifies a colonoscopy to detect premalignant or malignant lesions. The more metabolically active the polyp, the greater malignant potential exists.

A study of incidental colonic FDG uptake in 15 patients and its correlation with colonoscopic and histopathologic findings [201] concluded that nodular high FDG uptake (on a four-point scale) implies at least a 79% chance of abnormal histopathology. Another evaluation of (pre-)malignant colonic abnormalities in 39 patients for endoscopic validation of FDG-PET [37] found that, compared with colonoscopy, FDG-PET had sensitivity of 74%, specificity of 84%, and positive predictive value of 78%, though it failed to detect small polyps in four patients. Further, in a study of whole-body PET/CT tumor staging with integrated PET/CT colonography [213], PET/CT colonography proved accurate in local lymph node staging and staged nine out of 11 patients correctly. In addition, six additional extracolonic tumor sites were detected based on whole-body staging.

In certain populations at risk, such as patients with FAP and attenuated FAP, the use of FDG-PET/CT and FDG-PET/CT virtual colonoscopy may allow for less invasive surveillance and assessment of overall polyp burden as well as detection of advanced lesions in the stomach and small bowel in addition to colon [227]. A study of FDG-PET’s ability to detect cancers in FAP patients and its impact on clinical management [211] found that it detected all cancers present; no patients with negative FDG-PET developed cancer. This suggests that positive FDG-PET in FAP patients should lead to further examinations to rule out cancer, and seems to justify a more conservative approach in patients with negative FDG-PET. Careful study and validation of this technique is still needed. Virtual colonoscopy is a less onerous potential screening tool than colonoscopy for surveillance of both routine and high-risk populations. Combining anatomic detail from virtual colonoscopy with metabolic information obtained through FDG may improve accuracy over either test alone. It is doubtful that FDG-PET/CT virtual colonoscopy would replace standard colonoscopy. However, it may prove useful as an adjunct to assist surveillance in these complicated patient groups.

Dr. Hemant Roy, Evanston-Northwestern Healthcare, maintained that screening the entire average-risk population (more than 70 million Americans over the age of 50) for colon cancer is impractical given the expense, lack of sufficient endoscopic capacity, and complication rates [103]. Therefore, limiting colonoscopy to those patients who harbor polyps/carcinoma is essential for efficacious and cost-effective colorectal cancer screening.

Many risk-stratification techniques currently exploit the field effect, which proposes that the genetic/environmental milieu leading to tumorigenesis in one area of the colon should be detectable in some form throughout the colon. The presence of a carcinogenic field effect or malignancy-associated changes can be inferred from finding distal focal neoplastic lesions (predictive of proximal lesions) and diffuse alterations
in histologically normal mucosa [14,29,67,155,199]. However, these current field effect markers lack sensitivity and, therefore, require expansion and additional development to improve diagnostic precision.

Light scattering, using the scattering angle and intensity of light at a particular wavelength to determine the size and structure of particles [96,169], is sensitive to particles 10–20 times smaller than regular microscopy. The new generation of light-scattering technology provides quantitative assessment of microarchitecture by four dimensional analyses including wavelength of light, scattering angle, azimuthal angle of scattering, and polarization of scattered light.

Two novel light-scattering technologies, four-dimensional elastic light-scattering fingerprinting (4D-ELF), and low-coherence enhanced backscattering spectroscopy (LEBS), allow quantitative assessment of the nanoscale architecture of cells, providing a practical marker for genetic/epigenetic changes of the field. Initial studies in two experimental animal models indicated that altered light-scattering signatures from historically normal mucosa preceded the earliest conventional markers of colon carcinogenesis [168].

Initial clinical studies have been conducted with support from NCI and the National Science Foundation. Current data collected from > 200 patients indicate that 4D-ELF analysis has > 90% accuracy in identifying risk of colon polyps/cancers [168]. Spectral markers are also being used to evaluate familial syndromes such as FAP and HNPCC. In sporadic lesion risk stratification, free-standing probes would be developed to use in conjunction with current screening techniques by physicians during annual physical exams; the probe would be inserted into the rectum similarly to taking a rectal temperature.

Dr. Thomas Wang, Stanford University, described the uses of in vivo molecular colon imaging, including understanding the dynamic behavior of mucosa, observing spatial relationships in situ, and guiding and monitoring response to therapy.

Candidate dysplasia-binding peptides have been identified as imaging probes using phage display [28,192]. Phage display provides a highly complex library of peptides that can be screened to identify ligands that preferentially bind to molecular markers of disease. Advantages of using small peptides as probes include low immunogenicity, short plasma half-life, high diversity, good mucosal penetration, and easy synthesis. These peptides can be conjugated to fluorescent molecules, permitting optical visualization of dysplasia in vivo by confocal microendoscopy.

Potential targets in the colon include cell junction, anti-apoptosis, cell growth, and cell proliferation markers. Clinical endpoints include detecting dysplasia, identifying tumor margins, assessing stage of transformation, and performing risk stratification. This will require validation of results with immunohistochemistry to confirm the identity of peptide binding biomarkers.

### 2.8. Head and neck cancers

Drs. Vali Papadimitrakopoulou and Walter Hittelman, University of Texas MD Anderson Cancer Center, discussed the detection of head and neck carcinogenesis. Dr. Papadimitrakopoulou described several limitations to current oral cancer screening methods that could be addressed by imaging, including over-reliance on clinical experience for visual recognition, mimicking of early neoplastic lesions by more common benign lesions, and reluctance of many patients to undergo biopsy. Vital dye staining has been used to increase sensitivity, but has low specificity and also requires expertise. Using brush biopsy for cytology delays diagnosis and has an unknown false negative rate. Optical technologies like fluorescence spectroscopy are promising ways to detect the changes in autofluorescence associated with carcinogenesis.

Identification of biomarkers that could serve as risk predictors has benefited from producing several candidates. Among the most significant are genetic instability in the form of chromosomal polysomy and aneuploidy, overexpression of cyclin D1 and EGFR, and allelic losses at 3p and 9p, and p53 alterations [76,81,110,147,166,167].

Current chemoprevention studies involve interventions targeted toward p53 abnormalities and inflammation using selective COX-2 inhibitors, natural products such as green tea components (epigallocatechin gallate (EGCG) and green tea catechins), and EGFR kinase inhibitors [132,222,223]. EGCG inhibits activation of HER-2/neu and downstream pathways in human head and neck and breast carcinoma cells and inhibits activation of EGFR and downstream Stat3 and cyclin D1 promoter activity and level. Green tea catechins inhibit angiogenesis of human umbilical vein endothelial cells by inhibiting VEGFR, inhibit chemotaxis and invasion, and induce apoptosis at high concentrations. EGCG inhibits DNA methyltransferase and reactivates methylation-silenced genes in cancer cell lines (e.g., p16, RAR/3, MGMT, hMLH1).

Retinoid chemoprevention trials have found that RAR/3 is up-regulated in oral premalignant lesions
(OPL) by retinoids, and up-regulation is correlated with response. High-dose retinoids reverse oral leukoplakia and prevent second primary head and neck tumors; however, high doses of retinoids are too toxic for patients to take for long periods of time [51,115]. There is a slow return to malignant phenotype when treatment ceases. Low-dose retinoids are superior to β-carotene for maintenance of leukoplakia remission.

Dr. Hittelman described the multistep process of field cancerization and tumorigenesis in the upper aerodigestive tract and opportunities for imaging this process at the molecular level by chromosomal in situ hybridization (CISH). Exposure of epithelium to carcinogenic insults and cofactors results in chronic tissue damage and wound repair. Early in the process, damaged tissue contains DNA adducts, mutations, and polycyosity. Further damage results in clonal formation and aneuploidy. Subsequently, the damaged tissue becomes premalignant, characterized by clonal outgrowth and clonal evolution. CISH techniques quantify levels of genetic changes in head and neck tumors and adjacent epithelium. Cells with abnormal chromosome copy numbers are detected in normal-appearing epithelium within the head and neck cancer field; the level of chromosome polycyosity increases with histological progression to cancer [76,110].

Genomic instability increases during tumorigenesis. Spatial genetic mapping studies indicate a process of random chromosome instability and multifocal clonal outgrowth throughout the epithelium at cancer risk. Chromosome instability is also detected in oral and laryngeal biopsies from leukoplakias [10,36,62,89,117,141,142,145,158,202]. Individuals exhibiting high levels of chromosome instability have a high rate of subsequent cancer development. A study in the bronchial epithelium of current and former smokers found that genetic instability decreases as a function of time after smoking cessation, but chromosome indices remain elevated [76]. These studies suggest that genetic instability is a driving force for head and neck tumorigenesis. Targeting processes that promote genetic instability may slow progression in head and neck cancer. Down-modulation of genetic instability may delay time to cancer onset. Assessing ongoing genetic instability and clonal outgrowth may therefore be useful to identify individuals at high cancer risk and monitor the impact of chemopreventive intervention.

New approaches are needed to detect and treat OPL, the precursor of oral cancer and indicator of increased risk. However, no reliable clinical or histologic features identify OPL, and its natural history is variable. Optical interrogation in vivo may provide improved understanding of the ways in which light interacts with the structural and molecular changes of oral cavity neoplasia. Dr. Calum MacAulay, British Columbia Cancer Research Center, described a simple hand-held optical diagnostic tool designed to serve this purpose.

The visually enhanced lesion scope (VELscope™) illuminates the mouth with blue light, which excites certain molecules in mucosal cells, causing them to absorb the light energy and re-emit it as visible fluorescence. Dysplastic cells emit dark green to black fluorescence, whereas normal tissue emits a pale green fluorescence. The device has been tested in 44 people; in all but one instance, normal and abnormal tissue could be distinguished correctly. The findings were confirmed by biopsy and standard pathology [105].

### 2.9. Cervical cancer

Drs. Diane Solomon and Jose Jeronimo, NCI, explained that cervical cancer is the second most common cause of cancer deaths in women worldwide. Most cervical cancers are squamous and arise in the transformation zone. Cervical squamous abnormalities are described in cytologic terms as low-grade and high-grade squamous intraepithelial lesions, and histologically as cervical intraepithelial neoplasia (CIN) grades 1, 2, and 3. Approximately 25% of cervical cancers in the US are adenocarcinomas arising in the endocervical canal, less accessible by screening modalities than squamous cancers.

Virtually all cervical cancer is caused by human papillomavirus (HPV). In particular, HPV 16, 18, 31, and 45 together account for approximately 80% of cervical cancers worldwide, with HPV 16 being the most likely to cause cancer. Evolution from HPV infection to precancer usually takes several years to a decade. Most recently, an understanding of the role of HPV in cervical carcinogenesis has been translated into clinical utility. Cervical cancer screening for the past 50 years has relied on cytologic evaluation of cells scraped from the surface of the cervix – the Papanicolaou test. Cell abnormalities can be identified and treated before cancer develops. Unfortunately, cytology is not perfectly sensitive or predictive. Cervical HPV testing has been utilized as a primary screening test in conjunction with cytology in women > 30 years old. The higher prevalence of HPV in younger women precludes the use of HPV testing in that age group.

The goals of imaging in the early detection of cervical cancer are to survey and document areas at risk and
to provide tools to accurately assess morphological and molecular biomarkers using inexpensive, portable, automated systems with high sensitivity, specificity, and significant positive and negative predictive values. Colposcopy and biopsy (previously the gold standard of diagnosis) detects only about two-thirds of prevalent CIN 2 and above, although taking more biopsies improves sensitivity.

Dr. Rebecca Richards-Kortum, University of Texas, related development of dual field of view (FOV) systems which image the morphologic and molecular signatures of various neoplasias non-invasively in real time. These systems image reflected and fluorescent light at two spatial scales. Small FOV imaging systems have high resolution and maximize specificity. Confocal microscopy with fiber optics and miniature components allows visualization of morphology in vivo in real time [113]. Large FOV imaging systems use multispectral digital imaging with low (millimeter) resolution to image tissue morphology and maximize sensitivity (e.g., multispectral digital colposcope).

Imaging the molecular features of cancer requires molecular-specific contrast agents which can safely be used in vivo. To address this problem, optically active contrast agents have been developed to image the expression of three well-known molecular signatures of cervical neoplasia: overexpression of EGFR, matrix metalloproteinases (MMPs), and oncoproteins associated with HPV infection [188]. Contrast agents include dyes and nanoparticles. Overexpression of glucose transporters in tumor cells allows use of fluorescently labeled 2-NBD-deoxyglucose (2-NBDG) as a marker of cancer cells.

RNA is also being used as a target marker for HPV detection, as its expression can signal active HPV infection. RNA can also help differentiate between high-risk and low-risk HPV isotypes. It is imaged in living samples without washing using molecular beacon-based approaches, and can also be detected in fixed cells using in situ hybridization [39]. Both approaches can be easily adapted to the clinical setting. Detection using RNA has a high specificity.

Visualization of gold nanoparticle scattering can be used to screen for cervical cancer. This technique has the benefit of strong scattering, it causes no photobleaching, and is inert and biocompatible. The nanoparticle complex formed is simple, stable, and well-understood. In addition, use of metal nanoparticles provides up to five-fold more contrast. Procedure-induced toxicity is limited to localized heating with gold nanoparticles to temperatures well below the threshold of thermal damage.

Research involving integration of optical imaging systems and contrast agents with advances in functional genomics is on-going. Molecule-specific, optically active contrast agents and inexpensive, rugged, and portable imaging systems are being developed to monitor the 3D profile of targeted biomarkers for cervical, oral, and lung cancer. Four molecular signatures of neoplasia will be targeted: EGFR, MMP, telomerase, and integrins. The safety and efficacy of these agents and systems will be tested in animal models to provide support data for phase 1 and 2 clinical trials.

2.10. Skin cancer

Dr. Steve Stratton, University of Arizona, provided an overview of skin cancer prevention and its implications for imaging research. More than one million new skin cancer cases are diagnosed in the US annually. The estimated number of skin cancer deaths is 10,710 in 2006, 7,010 from melanoma and 2,800 from other skin cancers. One in five Americans will develop skin cancer. Incidence of melanoma has increased 690% from 1950 to 2001, with a 165% increase in mortality [5].

Skin cancer is usually preventable with early detection, and most skin cancer is curable when recognized and treated early. Basal cell carcinoma (BCC) and squamous cell carcinoma (SCC) have > 95% five-year survival rates. Behavioral risk factors for skin cancer, all related to excess ultraviolet radiation exposure, are modifiable. Biological risk factors include fair skin, individuals who tan poorly, burn easily, have a tendency to freckle, and have many moles (especially dysplastic moles).

BCC is by far the most common skin cancer, accounting for about 80% of nonmelanoma skin cancers (NMSC). It is locally destructive and rarely metastasizes. SCC accounts for about 20% of common NMSC. More than 10% of SCC will metastasize. About 60% arise from pre-existing actinic keratosis (AK). Most AK spontaneously regresses; however, 0.1–10% may convert to SCC [193]. Treatment of AK includes surgical therapy such as cryotherapy or curettage; topical drugs such as 5-FU, diclofenac, imiquimod, and ALA; and photodynamic therapy with ALA.

Melanoma is the deadliest form of skin cancer. One in five diagnosed will die from the disease. The role of ultraviolet radiation is less well-defined than in NMSC; however, more than five severe sunburns in adolescence doubles risk. Clinical melanoma visual criteria are asymmetry, uneven borders, multiple colors or shades, and a diameter greater than 6 mm (ABCD cri-
Dysplastic nevi (DN) may convert to melanoma, though there is controversy over whether they are considered precancerous lesions [65]. In case-control studies of risk markers for melanoma, 34% of patients with melanoma had DN. Relative risk estimates range from 1.0–16.7 for melanoma in the presence of DN.

Currently, drugs in Phase 2 clinical development for skin cancer chemoprevention include DFMO, perillyl alcohol (target: farnesyl transferase), and resiquimod (target: toll-like receptor). Phase 2 clinical trial endpoints include lesion regression, epidermal/nevus normalization by histopathology, protein biomarkers by immunohistochemistry, detection of DN by epiluminescence microscopy (dermoscopy), detection of AK and DN by nuclear karyometry, and detection of AK by OCT.

Skin cancer imaging can be used for early detection to avoid unnecessary resection, to assist diagnosis, and for therapy monitoring. Early detection of melanoma while it is thin is crucial. When < 1 mm thick, melanoma’s five-year survival rate is 95%; however, thickness > 4 mm decreases the five-year survival rate to 24%. A naked-eye exam is only 65–80% accurate in determining lesion thickness. In addition, the standard ABCD criteria may fail.

Dermoscopy (epiluminescence microscopy) uses a hand-held device for enhanced visualization of pigmented lesions, revealing atypical pigment networks and diffuse pigmentation. The diagnosis of atypical nevi is based on algorithms with specific pattern analysis and WHO clinical criteria. Dermoscopy improves diagnostic accuracy [125], and decreases the number of benign lesions misidentified and excised [27].

Nuclear karyometry is an optical imaging technique used for quantitative histopathology and cancer risk prediction [11]. Digitized images of abnormal nuclei in formalin-fixed, paraffin-embedded, H&E-stained tissue yield information about nuclear chromatin patterns of epithelial tumor cells [11]. Karyometric features and measurements of nuclear abnormality are computed, and discriminant function analyses are employed to define deviations from normal and to identify the subpopulations of nuclei exhibiting changes. When karyometry becomes more automated, it will be useful as a screening tool; currently, karyometric analysis can perform risk quantitation and be used as a surrogate biological endpoint in chemoprevention trials.

OCT, based on reflection delays of light directed into tissue, is similar to ultrasound imaging in that tissue structures are mapped based on delay times corresponding to the depths of measured structures; however, ultrasound imaging is based on echo delays of sound [52]. Using light instead of sound, OCT results in much higher resolution imaging. Since light travels so quickly, conventional techniques cannot be used to measure the reflection delay times needed to resolve such small (1 µm) structures. Short reflection delays are measured using interferometry. Light from a broadband source is split into reference and sample arms. Interference occurs when the optical path length to the reference arm mirror and a reflector in the sample are nearly matched. The interference signals contain information on the distance to the reflecting surfaces within the tissue, as well as velocity of any moving components within the tissue [38,208,217]. Single wavelength imaging is currently being used because it allows better imaging on the surface. However, a move towards multispectral imaging is being made.

ThorLabs, Inc. has created a compact OCT system that may be more comfortable for patients than investigator-built systems; however, the depth of imaging is reduced. The goals of OCT analysis in skin are to develop noninvasive imaging systems for characterization of sun damage and neoplasia, and to develop quantitative image metrics based on grayscale images that discriminate image cellular changes [12]. Improved instruments will permit therapeutic monitoring of topical skin cancer chemoprevention drugs during trials, and aid in developing a more objective index of therapeutic efficacy.

Currently, 80% of the 5.5 million skin biopsies performed per year in the US, at a cost of 2.15 billion dollars, are benign. There are 63,150 new cases of potentially fatal invasive melanomas and 40,780 new cases of melanoma in situ (lentigo maligna) each year [4,5,84,130,198]. Dr. Milind Rajadhyaksha, Memorial Sloan-Kettering Cancer Center, presented data showing that optical imaging can potentially eliminate excess costs and improve the accuracy of procedures associated with screening for melanoma.

As noted by Dr. Stratton, pigmented lesions as they progress from normal to malignant follow ABCD criteria, including disorder in the epidermis and dermis, disrupted cell borders, irregular and atypical shapes, pagetoid distribution of cells, elongated and thickened dendrites, and eccentric melanin patterns [22,23,56,106,148–152,161,200]. Confocal reflectance microscopy allows image-guided mapping of the developing lesions and has the potential to guide surgical excision of melanomas by detecting sub-clinical margins of melanoma in vivo.

Presurgical mapping of melanomas is important due to high rates of postsurgical local recurrence and unde-
tected subclinical margins. The current clinical standard uses presurgical biopsies to detect subsurface cancer-to-normal tissue margins, which can take 10–30 biopsies for large melanomas. Presurgical confocal mapping to determine subclinical, subsurface margins of melanomas \textit{in vivo}, and in particular lentigo maligna, may guide precise surgical excision, topical drug treatment, and monitoring.

Two confocal reflectance microscopes (VivaScope 1000® and VivaScope 3000®) are being studied to enable improved screening, diagnosis, presurgical planning, and intra-operative intervention in melanoma, with the goal of minimizing biopsy numbers and decreasing associated pain and expense. Results to date of one phase 1 study still in progress confirm that the confocal reflectance map correlates well to pathology in seven lentigo maligna and amelanotic melanoma patients. Clear margins were seen for surgical excisions from the scalp, cheek, and legs.

A phase 2 study is also being performed to determine the feasibility of using dermatoscopy-guided confocal imaging to find differences between melanomas and sun-damaged skin, and to detect margins to guide surgery. Thirty-six patients with four types of poorly defined melanomas were studied to determine the correlation of conventional dermoscopy \textit{versus} confocal imaging \textit{versus} histology. Image analysis was performed by independent dermatologists/pathologists. In another application, confocal mosaicing of BCC is also being studied to potentially guide Mohs surgery.

Due to the complexity of light scattering, a gap exists between development and application in medical optical imaging. Much work in spectral measurement is lacking. Large modeling equations are possible, but the question of normalization, which has to be referenced to localized sites, remains to be addressed. Simulations are only a starting point to look for solutions. A database is also required for spectral measurements for inter- and intra-person variations.

Dr. Atam Dhawan, New Jersey Institute of Technology, indicated that the current general clinical diagnosis rate of melanoma is < 70%. Imaging methods such as dermoscopy for imaging skin lesions to detect skin cancer use predominantly surface illumination and rely heavily on the pattern, texture, and color of the skin lesion. Although clinicians have found that dermoscopy methods like optical epiluminescence (ELM) can improve diagnostic accuracy by approximately 10–20%, room for significant improvement remains for \textit{in vivo} diagnosis of skin cancer [1,61,122]. The Nevoscope was developed as a novel, noninvasive optical instrument for \textit{in situ} imaging of skin lesions that combines ELM with optical transillumination (TLM), which is more sensitive in visualizing the superficial vascular network and increased blood volume associated with malignant melanoma and BCC.

Better imaging and mapping sensitivity with multi-ring, multi-directional, multi-spectral TLM imaging is needed. In addition, validation of these techniques is needed.

\section*{2.11. Discussion session on development of imaging devices for esophagus and bladder precancer and cancer}

The discussion session after the presentations on esophagus and bladder touched on several issues common to imaging cancer and precancer in hollow organs and how current endoscopy/cytoscopy strategies might benefit from cross-disciplinary technology applications. Miniaturization approaches developed in endoscopy/bronchoscopy might be used to better visualize the ureter and upper tract cancers in urology. The standard four-quadrant cytoscopic exam might be improved by spectral or scattering imaging modalities developed for Barrett’s that would permit increased detection of the bladder surface, allowing inspection of suspicious regions for nuclear or structural abnormalities by magnification endoscopy. Most discussants believed that additional multicenter, controlled trials are needed to investigate whether the various ablation strategies employed for managing Barrett’s esophagus improve long-term outcomes compared to esophagectomy. Since thermal ablation itself induces an injury/repair process that might foster additional pathologies, adjunctive therapies with chemopreventive drugs such as NSAIDs should be evaluated in concert.

Continued investigation of heritable or molecular risk factors in bladder and esophageal cancer was of considerable interest. The well-known association between \textit{N}-acetyltransferase (NAT) genotypes, smoking, and bladder cancer risk [165] may have analogues in aerodigestive cancers such as the recently described risk variation seen with certain single nucleotide polymorphisms (SNPs) in the interleukin (IL)-1B and tumor necrosis factor (TNF) genes in gastric cancer [153]. New genotypic predictors of responsiveness to intravesical agents such as BCG have also been reported for bladder cancer [66,111], but none of these methods have been fully used for clinical trial design cohort selection or stratification.
Preclinical approaches to understanding the mechanism of novel anticancer agents such as histone deacetylase (HDAC) inhibitors could benefit from these new imaging techniques. Nuclear chromatin size and shape factor changes and their correlation with various spectral or optical scattering signals could be examined in animal models that employ DNA adduct-forming carcinogens, as could the impact of chromatin remodeling agents such as HDAC inhibitors. Molecular imaging of nuclear (e.g., apoptosis) and tissue architecture alterations in response to thermal coagulation and repair/regeneration might also provide critical information about the long-term safety of these interventions.

2.12. Discussion on development of devices for breast and prostate imaging

Several opinions on advantages and limitations of the described imaging technologies were voiced. Reduced radiation exposure compared to conventional mammography was seen as a distinct advantage for optical spectroscopic or MRI monitoring of the breast, particularly for high-risk younger women more likely to have dense breasts and requiring follow-up for longer periods of time. Although optical/spectral mammography is lower cost and less invasive than DCE-MRI, validation in a clinical setting will enable better understanding of its accuracy vis a vis tumor confounders (e.g., cysts, hematomas, water/lipid/oxygen signature variations during the menstrual cycle, etc.). Use of FUS for ablation or enhanced drug delivery in breast or prostate presented some unique challenges for gaining regulatory approval and clinical adoption. Concerns were voiced that high mechanical index FUS could produce the kinds of residual seromas or telangiectasias seen with other thermal therapy modalities. A scenario was raised in which heat shock protein induction might induce resistance to subsequent chemotherapy. All agreed that well-designed clinical studies could help assuage some of these concerns. FUS might have an advantage over lumpectomy in multifocal breast disease, where cosmesis is still desirable. This would not avoid the need to monitor local node status, but validation of axillary incision or percutaneous sentinel node biopsy in concert with FUS could be compared to mastectomy for multifocal disease. In prostate cancer, the biggest challenge now is not diagnosis, but how to differentiate men with indolent versus aggressive disease in order to avoid overtreating one population and undertreating the other. At present, the resolution available for imaging modalities described (e.g., ∼5 mm voxel size for MRS) is less than the size of typical tumor foci, but instruments and agents available to control rather than kill prostate cancer cells are improving rapidly.

2.13. Roundtable discussion on lung and colon cancer

The discussion of lung and colon cancer focused on the goals for both colon and lung cancer screening – to precisely isolate susceptibility, and to find a broad spectrum of serum or imaging based markers. Efforts in the field are already underway to develop beacon markers and cocktails. Patients with the specific markers would then be screened aggressively. Specific markers may not yet have been identified or stratified for their importance. Biospecimen repositories and image databases from large clinical trials will aid in this identification [70,134]. Once at-risk patients are identified, lesions will need to be spatially located. Also needed is improved whole body surveillance. Different types of validation methodologies need to be explored.

Lung lesions can be detected at an early stage, but it is still impossible to know whether the lesions will go on to be malignant. Since 25% of cured lung cancer patients will get a second cancer within five years, it is important to follow some early lesions in order to acquire information on lesion progression that may be lost with ablation. The process of monitoring some lesions in at-risk patients using some type of optical technique and ablating other lesions would have to be repeated every couple of years, much as AKs are followed and ablated for skin cancer control. Coupling microendoscopy with CT for visualizing peripheral lesions is an emerging technique. More precise, population-friendly treatment methods are needed. Image-guided intervention [59] and inhalational chemotherapy are potential treatment options for lung cancer.

For colon cancer screening, there is a need for patient-acceptable, noninvasive colon screening. While colonoscopy is a great tool, it is not the answer. Stool testing is an option; however, the fecal milieu and colonic preparative methods may negatively affect the ability to find markers. FOBT can be useful if done regularly. Fecal DNA testing is also an option [8]. PET scanning can be useful, since the more metabolically active the polyp, the greater the malignant potential [7]. A pill-cam device may permit evaluation of mucosal health during transit of the GI tract [60].
2.14. Discussion on head and neck, cervix, and skin cancer

The roundtable discussion on head and neck, cervix, and skin cancer focused on what needs to be done to close the gap between development and application of the imaging techniques discussed in the presentations. Several investigators received FDA 510k approval for devices presented after demonstrating safety in the development process. However, the gap between what is academically proven and what can be used clinically must be bridged. Optical devices require more work on spectral measurement, creation of large modeling equations, and databases for spectral measurements outlining inter- and intra-person variations. Simulations are important tools but can only be used as a starting point to help ask the right clinical questions.

Several aspects of cervical cancer screening were specifically discussed by the panel. Visualization using gold nanoparticle scattering raised concerns about light-induced toxicity. However, the localized heating stays well below the threshold of thermal damage. A poor correlation between HPV seroconversion as a biomarker and infection in the cervix was also noted. Discussion about the HPV status of male partners centered on the lack of an equivalent to cervical epithelium in the male reproductive tract, making it difficult to know when men are infected with HPV. As risk of penile cancer is lower for men than is risk of cervical cancer for women, studies of HPV infection in men associated with bladder cancer results are controversial. Interestingly, there is some association between HPV and head and neck cancer, mainly HPV 16 [77].

2.15. Translational opportunities for molecular imaging

Dr. Martin Pomper, Johns Hopkins University, provided a keynote overview on translational opportunities for molecular imaging across the field of oncology, illustrating additional advances in the areas of radiotracer development and more innovative cancer imaging applications in several organ systems presented at the Workshop. Modern pharmaceutical chemistry has applied molecular design and targeting approaches to drug development. Now the challenge is clinical testing of novel molecules which may have cytostatic rather than cytotoxic effects and which do not produce the frank tumor shrinkage assessable by conventional anatomic imaging approaches. Molecular imaging approaches address and solve these problems by producing tumor metabolic response maps, as well as providing new solutions to continuing problems of dose selection and pharmacokinetic/dynamic characterization that always confront early phase investigational drug optimization programs [91]. For instance, FDG-PET imaging has helped direct the development of EGFR tyrosine kinase inhibitors (TKI) such as erlotinib, which inhibit tumor glucose metabolism, a good reflection of clinical benefit. Development of a labeled probe of gefitinib, another EGFR TKI, may better define optimal pharmacokinetics and patient responders, much as labeled cytotoxics such as C11-paclitaxel did in the past. A prostate-specific membrane antigen (PSMA) monoclonal antibody labeled with 111-indium is the basis for developing a successful radiotracer (ProstaScint™) widely used in nuclear medicine to diagnose the presence of metastatic androgen-independent prostate disease and monitor its treatment [224]. Targeting this cell surface enzyme has also been the focus of therapeutic drug design efforts. Development of PSMA-specific inhibitors [230] has been accelerated by radiosynthesis of several analogues as PET tracers [154] to evaluate biodistribution of the molecule in animal models of prostate cancer [49]. Though PPAR-γ may be a more amenable target for development of cancer preventive agents, its labeled agonists may not have predictable kinetics when examined in vivo. For example, uptake of radiolabeled PPAR-γ ligand 111-C-GW7845 (rosiglitazone) was unaffected by cold chase with the unlabeled agent [126]. Similar preclinical testing of candidate imaging probes directed to early IEN biomarkers (e.g., prostate stem cell antigen, claudin) are also underway [49]. The pivotal role played by steroid hormone signaling in reproductive tract cancers suggested the value of developing probes to image their distribution and regulation in laboratory and clinical studies [95,129]. 18F-estradiol and 18F-progestins have been used to visualize breast and ovarian cancers; 18F-dihydrotestosterone has similar application in prostate cancer androgen-receptor studies [107]. A variety of novel reporter probe constructs have served as tools to dissect cancer-specific signal transduction cascades [88,229] and gene transcription (E2F, HGF monoclonal antibody), and to evaluate gene therapy and biological therapy strategies [16].

2.16. Overview of NCI and FDA programs relevant to imaging science development

Drs. Daniel Sullivan (Cancer Imaging Program), Jorge Gomez (Specialized Programs of Research Excellence), Sudhir Srivastava (Early Detection Research
Table 1

Internet resources for imaging science development

http://dtp.nci.nih.gov/docs/raid/raid_index.html
http://imaging.cancer.gov/programsandresources/specializedinitiatives/dcide
http://spores.nci.nih.gov/
http://edrn.nci.nih.gov/
http://deaninfo.nci.nih.gov/concepts/CA-02-016.htm
http://www.cancer.gov/trwg/
http://imaging.cancer.gov/programsandresources/specializedinitiatives/SAIRP
http://imaging.cancer.gov/programsandresources/specializedinitiatives/icmics
http://imaging.cancer.gov/
http://www.fda.gov/oc/combination/
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