Dear Colleagues,

At Cleveland Clinic, we believe the “Science is the Care.” I am proud to say that we are home to one of the most robust cancer research enterprises in the United States, encompassing basic, clinical and translational research. The studies conducted in our labs and clinics and highlighted here in Cancer Consult represent the future of diagnostic testing, technological advances in treatments and improved therapeutics.

One of the greatest unmet needs in cancer diagnostics are tools that accurately predict whether a lesion is cancerous or benign. Investigators at Cleveland Clinic and several other leading breast imaging centers are participating in an observational study, the PIONEER-01 Pivotal Study, evaluating a new imaging technology that may better help us make this difficult, critical judgment.

Cleveland Clinic’s Rose Ella Burkhardt Brain Tumor and Neuro-Oncology Center is an international leader in diagnosing and treating primary and metastatic brain tumors. Our physicians studied the use of properly tuned, very-low-intensity, intermediate-frequency electric fields — otherwise known as tumor treating fields (TTFIELDS) — and were among the first in the United States to prescribe TTFIELDS to treat glioblastoma patients. This research led to the marketing approval of the NovoTTF-100™ System in the United States for use in patients with recurrent glioblastoma.

Genomics is continuing to revolutionize risk assessment, tumor classification and treatment. Radiation oncologist Mohamed Abazeed, MD, PhD, recently has joined Cleveland Clinic and has established a lab focused on identifying the genetic abnormalities that make tumors resistant to treatment and developing a physiologically relevant model to test gene/drug treatment protocols.

As physicians and researchers, we are acutely aware of the decrease in federal funding for cancer research. We are pleased to announce that Keith McCrae, MD, is a recipient of an American Society of Hematology (ASH) Bridge Grant, a one-year, $100,000 award to provide crucial interim support to his research on endothelial cell function, including the roles of endothelium in thrombosis and cancer.

Also included in this issue is a selection of the abstracts presented at the 2014 Annual Society for Clinical Oncology (ASCO) Annual Meeting. It is only through continued research that we can accelerate landmark advances in the prevention, treatment and cure of cancer. I hope that you find the information in these pages useful in your practice. Please do not hesitate to contact me with any questions, concerns or suggestions at 216.444.6922 or bolwellb@ccf.org.

Sincerely,

Brian J. Bolwell, MD, FACP
Chairman, Taussig Cancer Institute

On the cover: Large-scale genomic data is synthesized and then parsed using novel computational algorithms to identify genes involved in resistance to therapies in current use.
A large gap remains in understanding which of the many cancer mutations is actionable — meaning susceptible to drug therapy — and which is simply a passenger or silent mutation.

Mohamed Abazeed, MD, PhD, is working to answer that question. Dr. Abazeed joined Cleveland Clinic in late 2013 from the Harvard Radiation Oncology Program.

As a radiation oncologist with training in radiation science, genomics and molecular biology, Dr. Abazeed designs physical and computational platforms to help discern which mutations are significant and which may not be. His work involves identifying the genetic abnormalities that make tumors resistant to treatment and developing a physiologically relevant model to test gene/drug treatment protocols.

Dr. Abazeed’s research approach initially used more than 1,000 genetically diverse, patient-derived cell lines from a variety of solid tumors. Grown in culture, these cell lines were comprehensively genetically profiled and tested to see how they respond to radiation or particular drugs. “We have developed unique computational platforms based on matching algorithms that allow us to couple the resistance of these tumors with particular genetic alterations,” says Dr. Abazeed.

**Xenografts as Patient Avatars**

Clinical tissue samples form the basis of the next step in the research process. Dr. Abazeed’s team receives a section of solid tumor within hours of a cancer patient’s surgery. That tissue, which will be fully sequenced to determine its genomic profile, is injected into a mouse to create a xenograft. Better known as patient-derived tumor xenografts, these models preserve the original characteristics of a patient’s cancer and mimic the disease more effectively than cancer cell line-derived models. This allows Dr. Abazeed’s team — which includes cancer biologist Craig Peacock, PhD, whose expertise is in the development of the patient-derived mice — to

(continued on page 4)
evaluate therapies in a more individualized and targeted way, potentially leading to significantly improved clinical trial design.

“That is the concept of a patient avatar,” Dr. Abazeed explains. “We can apply the information taken from the cell line data in terms of what the genetic predictors of response to therapy may be, and we test them using this more physiological system.” In short, the cell line information gives some indication of which therapy may be most effective, and the xenograft provides a specific individual test bed.

Investigators previously employed this approach, but its use has been limited due to the required expertise and access to fresh tissue.

“It doesn’t influence a patient’s treatment directly yet, but you can see how the information gathered from these studies significantly accelerates both clinical studies and potentially therapeutic options for patients,” says Dr. Abazeed. His team injected its first mouse with a patient tumor sample at the Taussig Cancer Institute in March 2014.

Dr. Abazeed’s laboratory is equipped with next-generation genetic sequencers, coupled with industry-modeled, high-throughput profiling capabilities, enabling the investigators to rapidly generate and test novel hypotheses. In addition to Dr. Peacock, who arrived in 2014 from Johns Hopkins University, Dr. Abazeed’s team includes postdoctoral research fellow Brian Yard, PhD.

Dr. Abazeed will first study the cell line/xenograft approach in lung carcinomas. If it proves to be a physiologically relevant system, the team will expand to other solid tumor types.
World-Class Genetics Training

After completing his radiation oncology residency training at Boston’s Brigham and Women’s Hospital, Dr. Abazeed was awarded a two-year postdoctoral appointment at the Broad Institute of MIT and Harvard. The Broad Institute is a premier academic, genomics-focused research organization performing leading-edge cancer genetics research.

While at the Broad, Dr. Abazeed and colleagues discovered that alterations in the \textit{NFE2L2} gene are present in 34 percent of lung squamous cell cancers, which makes these cancers resistant to radiation and chemotherapy. Working with Novartis, the team developed a drug, BKM-120, targeted against that mutation.

A Phase IIb clinical trial to evaluate BKM-120 is underway at the Dana-Farber Cancer Institute. Dr. Abazeed and colleagues filed a patent on their mutation discovery and the coupling of that mutation to the drug. The clinical trial will help determine whether patients with the mutation respond better to the medication than do those lacking the mutation.

At Taussig Cancer Institute, Dr. Abazeed’s first research project with the mouse xenografts will involve the study of BKM-120, which is also known to counteract the \textit{KEAP1} mutation, found in 23 percent of patients with lung adenocarcinomas. “Before moving on to a Phase II study, we want to test this drug/gene combination in a more physiological system,” he explains, “and we believe these primary xenograft models are an ideal link between the bench and the bedside.”

Going forward, Dr. Abazeed will focus primarily on identifying treatments for the low-frequency genetic mutations found in cancers. Because it would be nearly impossible to find enough patients with low-frequency mutations to conduct a statistically meaningful clinical trial, Dr. Abazeed hopes to initiate a new type of clinical study at the Taussig Cancer Institute.

“We would love to implement a clinical trial design that compares traditional chemotherapy to targeted therapies in cancers with multiple low-frequency alterations,” he says.
Currently, many women with a suspicious breast lesion require a biopsy. Yet, of the 1.7 million core needle or surgical breast biopsies performed each year in the United States, more than 70 percent yield benign findings.

Investigators at Cleveland Clinic and several other leading breast imaging centers are participating in an observational study evaluating a new imaging technology that may better predict which breast masses are cancerous and which are benign.

The Imagio® breast imaging system is based on the technology of opto-acoustic imaging (OAI).

“It is a combination of optical imaging, which is light, and sound imaging, which is ultrasound,” says Stephen Grobmyer, MD, Cleveland Clinic’s principal investigator for the clinical trial, Director of Surgical Oncology and Director of the Breast Center. “That combination gives a unique way to look at tissue.”

Instead of radiation, the technology sends pulses of safe near-infrared light that penetrate breast tissue. The light creates sound waves within the tissue that are detected by an ultrasound transducer.

Measuring Hemoglobin Oxygenation

The Imagio OAI system currently under study at Cleveland Clinic and other sites uses two specific light wavelengths. At one of these wavelengths, the strongest absorber of light is oxygenated hemoglobin. At the other, deoxygenated hemoglobin absorbs the light most strongly.

“When you use those two different wavelengths, you can create a map of oxygenated blood and deoxygenated blood within tissue,” Dr. Grobmyer explains. “This technology is providing us with functional imaging based on the amount of oxygenated/deoxygenated blood within a lesion and provides us with information about metabolism and oxygen exchange without using any contrast agent at all.”

Cancerous tissue tends to have a higher metabolic rate and extract oxygen from blood more readily than does normal resting tissue. The combination of hemoglobin concentration and relative oxygenation is intended to aid differentiation between malignant and nonmalignant lesions. “We are hoping that by looking at the pattern of oxygenated/deoxygenated blood, along with the image of the lesion, we can better predict which masses are more likely to be cancerous and non-cancerous,” he says.

Details of Study

The PIONEER-01 Pivotal Study of the Imagio system is a multicenter investigation that aims to enroll 2,000 patients. Cleveland Clinic intends to enroll 165 patients in this study — patients with breast lesions classified via ultrasound as Breast Imaging Reporting and Data System (BI-RADS) 3 (recommended to have short-term follow-up) and BI-RADS 4 and 5 (requiring biopsy).

Women participating in the trial undergo imaging with the Imagio system before anything further is done. Their imaging information is sent for evaluation by radiologists who are not associated with
the patient’s clinical care. If a study participant has a biopsy, her Imagio results are then compared with the pathology results to determine correspondence. Patients who do not undergo a biopsy are followed for a year, after which they receive a follow-up OAI evaluation. The presumption is that if a lesion is stable at a year, it is unlikely to be cancerous.

OAI is not intended to be a first-line screening tool at this time. “It has to be a focused study where you find a lesion on an ultrasound or mammogram and then home in on it with this imaging device and just look at this area,” says Dr. Grobmyer. “The goal is to reduce the number of benign biopsies being done. It is a high bar because you can’t afford to miss a cancer. That is the challenge with any technology like this.”

But preliminary statistical studies indicate that OAI may have the potential to define a subset of patients who may not need biopsy. The PIONEER-01 trial is the first prospective clinical study of the technology on a large group of women. Says Dr. Grobmyer, “If this works out, instead of going to biopsy, in many cases we could apply this technology and inform the patient right then and there that there is no chance of cancer, and she can go home with no need for further work-up.”
Physicians and investigators from Cleveland Clinic’s Taussig Cancer Institute made significant contributions to the American Society of Clinical Oncology (ASCO) 2014 Annual Meeting in Chicago, describing their research in multiple presentations. Here is an account of one of those presentations and abstracts of two others. (Cleveland Clinic authors in the abstracts are listed in bold.)

Study: Novel Approach to Breast Cancer Treatment May Help Prevent Premature Ovarian Failure, Boost Fertility and Survival

For women who go through chemotherapy to treat breast cancer, premature ovarian failure (POF) is a common consequence.

Unfortunately, POF can lead to issues such as amenorrhea, menopausal symptoms, sexual dysfunction and infertility. However, recent findings from a Phase III clinical trial may offer patients new hope for avoiding POF — and perhaps for reducing the risk of cancer recurrence.

**Improved Fertility and Mortality**

In the past, researchers have investigated preventing ovarian cycling during chemotherapy with hormone-suppressing drugs, such as goserelin. The goal: to preserve ovarian function.

But past studies have been inconclusive, says Halle Moore, MD, a Cleveland Clinic oncologist and Chair of the Taussig Cancer Institute Survivorship Committee. Many studies used the return of menstrual bleeding as a surrogate for protecting ovarian function. That is a potentially misleading endpoint because menstruation does not always indicate fertility and may not predict longer-term ovarian function. In addition, few data are available on pregnancy outcomes using this approach.

Dr. Moore is first author of a new study designed to bridge this gap in the literature. She presented the results of the Prevention of Early Menopause Study (POEMS) at the 2014 American Society of Clinical Oncology Annual Meeting.

POEMS was designed to determine whether adding goserelin — a luteinizing hormone-releasing hormone (LHRH) analog that suppresses the production of estrogen — to cyclophosphamide-based chemotherapy would reduce POF in stage I-IIIA breast cancer, as compared to chemotherapy alone. The study focused specifically on estrogen receptor-negative/progesterone receptor-negative breast cancer.

Outcomes of interest included ovarian failure at two years (defined by amenorrhea for the prior 6 months and elevated follicle-stimulating hormone [FSH]), pregnancy rates and survival rate. Among the promising study findings:

Of the 135 premenopausal women evaluable for the primary study endpoint, ovarian failure was observed in 22 percent of those in the control group but in only 8 percent of women in the goserelin group (odds ratio [OR] = 0.30, p = 0.03).

When the definition of POF was loosened to include either amenorrhea or elevated FSH — but not both — POF was observed in 45 percent of women in the control group but in only 20 percent in the treatment group (OR = 0.29, p = 0.006).

Among the entire evaluable study population of 218 patients, rates of pregnancy also favored the
Disease-free survival (p = 0.04) and overall survival (p = 0.05) were higher among women receiving goserelin.

“The POEMS findings represent the first demonstration of improved fertility prospects when ovarian suppression with goserelin was administered with chemotherapy,” Dr. Moore says. “They provide reassurance as to the safety of this approach.”

Transforming Treatments

Currently, there is no standardized method to prevent chemotherapy-induced POF. However, the POEMS findings could change the way oncologists treat patients with early-stage breast cancer. The possible loss of fertility often influences the treatment decisions young women make; many decline therapies associated with POF. But this new approach may help ease that concern and increase the number of tools at oncologists’ disposal.

“The option of ovarian suppression with an LHRH agonist can now be offered to prevent unwanted ovarian failure, which may be particularly important for young women interested in maintaining future fertility,” Dr. Moore says. “Ovarian failure can result in bone density loss, menopausal symptoms and infertility, but the latter is probably the greatest concern for young women who are still interested in having a family. Clearly, all of these are very important survivorship issues.”

Even though this study focused on specific types of cancer, the findings may prove to be applicable elsewhere, too. More research is needed, but Dr. Moore notes that such treatment may be useful for women with hormone receptor-positive breast cancer, as well as other cancers treated with similar chemotherapy, such as non-Hodgkin’s lymphoma.

SELECT: A Multicenter Phase II Trial of Adjuvant Erlotinib in Resected Early-stage EGFR Mutation-positive NSCLC

NA Pennell, JW Neal, JE Chaft, CG Azzoli, PA Janne, R Govindan, TI Evans, DB Costa, RP Greenener Rosovsky, HA Wakelee, RS Heist, AT Shaw, JS Temel, MA Shapiro, A Muzikansky, M Lanuti, TJ Lynch, MG Kris, LV Sequist

Background: Epidermal growth factor receptor (EGFR) mutant non-small cell lung cancer (NSCLC) is exquisitely sensitive to EGFR tyrosine kinase inhibitors (TKIs). Retrospective data suggest adjuvant TKIs may improve outcomes. This trial is the first to prospectively test the efficacy of adjuvant erlotinib in EGFR-mutant NSCLC.

Methods: Eligible patients (PTs) had resected stage IA-IIIA NSCLC harboring a TKI-sensitizing EGFR mutation. PTs were treated with erlotinib 150 mg/day for 2 years after completion of standard adjuvant chemotherapy and/or radiotherapy. With a sample size of 100 PTs the study was powered to demonstrate a primary endpoint of 2-year disease-free survival (DFS) > 85%, compared with a historical control of 76% in resected early-stage EGFR-mutant NSCLC.

Results: 100 PTs were enrolled at seven sites between 1/08 and 5/12; 45% stage I; 27% stage II; 28% stage IIIA. 89 PTs have reached 2-year follow-up. Toxicities were typical of erlotinib, with no G4/G5 events and 1 G2 pulmonary fibrosis. 40% of PTs required dose reduction to 100 mg/day and 16% required two dose reductions to 50 mg/day. 69% of PTs completed at least 22 months of erlotinib. With median follow-up of 3 years, the 2-year DFS is 90% (97% stage I, 73% stage 2, 92% stage 3). Median DFS and overall survival have not been reached. 24 PTs have recurred; only two during erlotinib treatment and 22 after stopping erlotinib, with a median time to recurrence of 12 months after stopping erlotinib. 63% (n = 15) of PTs with recurrence underwent repeat biopsy, and only 1 T799M was detected. 71% (n = 17) of recurrent PTs were re-treated with erlotinib, with 10 PTs remaining on erlotinib, treatment range 2 to 42 months. 8 PTs have died.

Conclusions: PTs with EGFR mutation-positive NSCLC treated with adjuvant erlotinib have an improved 2-year DFS compared to historical genotype-matched controls. Recurrences are rare on erlotinib and most occur in the 12 months after discontinuation, suggesting longer duration of adjuvant treatment may be beneficial. Recurrent cases after adjuvant erlotinib remain generally sensitive to EGFR TKIs.

Outcome of Male Patients and Black Patients Enrolled in S0221, an Intergroup Chemotherapy Study

GT Budd, WE Barlow, HC Moore, TJ Hobday, JA Stewart, C Isaacs, M Salim, JK Cho, KS Albain, K Rinn, HK Chew, GV Burton, TD Moore, GS Salakovic, BA McGregor, LE Flaherty, RB Livingston, DL Lew, J Gralow, GN Hortobagyi

Background: Assessment of the outcome of minority populations within clinical trials may give insight into important tumor and host factors relevant to those populations.

Methods: S0221 is a phase III trial of various dose schedules of doxorubicin, cyclophosphamide and paclitaxel. S0221 allowed enrollment of male patients (PTs) and was sufficiently large to enroll significant numbers of minority populations. Here, we report the outcome of male PTs and PTs of black race.

Results: Among 3,236 total PTs entered on S0221, 23 (0.7%) were male and 378 (12%) were of self-reported black race. Non-significant differences in male/female patients were seen in HER2 positivity (9%/19%), hormone receptor (HR) positivity (100%/66%), lymph node negativity (17%/26%), and treatment completion rate (70%/74%), but men were more often ≥ 60 years old (48%/21%, p = 0.006). Males had significantly worse disease-free survival (DFS), with 5-year DFS 49%/82%; HR = 3.16, Log-Rank p = 0.0003. Male gender remained an adverse factor for DFS in a Cox model adjusted for treatment, HER2 status, HR status, age, and nodal status (HR = 3.53, p < 0.001). Black/non-black patients did not significantly differ in HER2 positivity (22%/18%). Black patients were less likely to be HR-positive (51%/69%, p < 0.001), to have > 4 nodes involved (29%/36%, p = 0.02), to be age > 60 years (15%/22%, p = 0.008) and to complete treatment (63%/75%, p < 0.001). Black patients had worse DFS, with 5-year DFS of 75%/82%; HR = 1.55, Log-Rank p = 0.0001. Black race remained an adverse factor for DFS in a Cox model adjusted for treatment, HER2 status, HR status, and nodal status (HR = 1.48, p = 0.001) after analysis was limited to those completing therapy, adjusted for treatment (HR = 1.48, p = 0.01).

Conclusions: Male patients enrolled in S0221 had a markedly worse DFS, a finding which persisted after adjustment for tumor characteristics. Black patients in S0221 treated with third generation regimens had a worse DFS than non-black patients. These results are similar to those for post-menopausal HR+ black patients treated with a second-generation regimen (5FU/doxorubicin/cyclophosphamide) in S8814, suggesting a generalized phenomenon which is not improved with contemporary regimens.
All cells constitutively release submicron vesicles, termed “extracellular vesicles” (EV). EV with diameters greater than approximately 400 nm are generally referred to as “microparticles,” while EV less than 100 nm in diameter are usually termed “exosomes.” Most microparticles are derived by budding from the plasma membrane, while exosomes are derived from multivesicular bodies within endosomes.

EV are derived from all cell types, including platelets, endothelial cells, leukocytes and, in patients with cancer, malignant cells.

Due to their relatively large size, microparticles may be detected by flow cytometry and stained with specific antibodies to determine their cell of origin. While estimates of total circulating microparticle numbers vary, most studies estimate concentrations of approximately $1 - 3 \times 10^6$/mL of plasma.

Exosomes’ small size renders them below the limit of detection by flow cytometry, though they may be detected using biophysical approaches. Estimates of total plasma EV concentrations derived using such measurements are in the range of $10^8 - 10^9$/mL.

Since EV release from cells is enhanced by cellular activation or damage, elevated levels of circulating EV provide a biomarker for such processes.

**EVs and Antiphospholipid Antibody Syndrome**

Our laboratory has long-standing interest in endothelial cell function, including the roles of endothelium in thrombosis and cancer. A particular area of interest is antiphospholipid antibody syndrome (APS), a disorder characterized by thrombosis and recurrent pregnancy loss in patients with antiphospholipid antibodies (aPL).

Antiphospholipid antibodies are a broad family of antibodies that includes lupus anticoagulants, anticardiolipin and anti-β2-glycoprotein I (β2GPI) antibodies, all of which may be detected in the clinical laboratory. Despite the confusing nomenclature, most pathologic aPL are actually directed against β2GPI, a phospholipid-binding protein that is abundant in plasma.

When activated or otherwise damaged, endothelium becomes dysfunctional, losing its normal anticoagulant ability and expressing procoagulant properties. Our previous work has demonstrated that aPL activate endothelial cells through a multi-receptor pathway leading to activation of the protein complex NF-κB. The transcriptional activity of NF-κB results in decreased expression of anticoagulant, anti-inflammatory genes, and increased expression of procoagulant and proinflammatory genes. Our studies suggest that vascular activation is an important component of APS, as we observed significant elevations of circulating endothelial cell, platelet and tissue factor-expressing microparticles in 47 patients with APS compared with 150 healthy controls.

We have also explored the mechanisms of microparticle release by endothelial cells in response to aPL. We hypothesized that microparticle budding from cells would depend on rearrangements
Keith McCrae, MD, Awarded ASH Bridge Grant

Cleveland Clinic hematologist-oncologist Keith McCrae, MD, is the recipient of a 2014 American Society of Hematology (ASH) Bridge Grant.

The one-year, $100,000 awards are meant to provide critical interim support during a time of severe cutbacks in federal research funding. ASH gives approximately 30 of the Bridge Grants annually to member investigators whose hematology research proposals, despite earning high scores, were not funded by the National Institutes of Health (NIH) due to deep budget reductions. The NIH’s current budget is more than $700 million below what the agency received prior to mandatory 2013 sequestration spending cuts.

The ASH awards help researchers gather additional data to strengthen the resubmission of their NIH grant applications. Of the 29 scientists who received ASH Bridge Grants in 2012 and 2013, the society reports that nearly one-third went on to obtain NIH funding.

Dr. McCrae is the Director of Taussig Cancer Institute’s Benign Hematology Program, and is a staff member of the Department of Hematologic Oncology and Blood Disorders and the Department of Cellular and Molecular Medicine.

His research involves endothelial cell function, including the roles of endothelium in thrombosis and cancer. Dr. McCrae’s particular focus is antiphospholipid antibody syndrome, a disorder characterized by thrombosis and recurrent pregnancy loss in patients with antiphospholipid antibodies.

“My career is critically dependent on extramural funding during the current era when clinical revenues and institutional support mechanisms are also under pressure,” Dr. McCrae says. “The ASH Bridge Grant provides me with support to keep my research program alive while federal grant revisions are in progress.”
in the actomyosin cytoskeleton, which controls endothelial shape and membrane properties. We observed that upon exposure to aPL and their autoantigen, β2GPI, assembly of the actomyosin cytoskeleton was dramatically stimulated. Cytoskeletal assembly was dependent on phosphorylation of the myosin regulatory light chain (RLC) and was required for enhanced release of microparticles. Inhibitors of RLC phosphorylation blocked microparticle release from cultured endothelial cells exposed to aPL.

**Conclusions and Research Directions**

Taken together, these findings allow us to draw several conclusions. First, the elevation of circulating microparticles in patients with aPL suggests ongoing vascular activation, even in the absence of active thrombosis. Second, the release of microparticles from endothelial cells is critically dependent on assembly of the actin cytoskeleton, and the microparticles' release thereby may be blocked by Rho kinase inhibitors, which have been suggested as potential therapeutics for patients with cancer. Third, the increased expression of tissue factor by microparticles from APS patients suggests that they may contribute directly to thrombosis development.

EV also contain proteins, messenger RNA (mRNA) and small RNA species, particularly microRNA (miRNA), that may be freely transferred from the cell of origin to other cells. The content of EV varies dynamically according to the cell of origin and the nature of the stimulus that enhances their release.

To better understand the role of EV in disease, we have expanded our studies to include a detailed analysis of the biochemical and molecular composition of EV isolated from patients, including their content of specific proteins, mRNA and miRNA. Though our initial studies focused on APS, we have broadened our focus to include EV from patients with glioblastoma multiforme as well as pancreatic and other gastrointestinal cancers.

Proteomic and genomic analyses of tumor-derived circulating EV from patients with cancer may provide an opportunity to noninvasively obtain a “molecular snapshot” of the tumor in real time, providing information useful in designing therapies, determining prognosis, and monitoring molecular responses to standard and experimental agents.

**Confronting the Complexities of Steroid Receptors in Prostate Cancer**

The mechanisms that tumors use to shield themselves from chemotherapy and hormonal therapy often are targets for the development of new treatment strategies. Finding ways to dismantle the armor of drug resistance can restore cancer cells' vulnerability and extend patients’ lives.

However, as has become apparent from studies of advanced prostate cancer, tumors can adopt a drug-resistance mechanism that is essential for the life of their human host. This entanglement tactic makes it “particularly challenging” to devise treatment approaches aimed at deactivating the tumor’s drug resistance while not compromising the patient, Cleveland Clinic oncologist and researcher Nima Sharifi, MD, writes in a recent analysis and commentary in *The New England Journal of Medicine* (NEJM).

The key to disentanglement is a deeper understanding of how these conjoined drug-resistance/life-essential mechanisms work to combat newer hormonal therapies, said Dr. Sharifi, who holds the Kendrick Family Endowed Chair for Prostate Cancer Research in Lerner Research Institute’s Department of Cancer Biology. He is an associate staff member of the Taussig Cancer Institute’s Department of Solid Tumor Oncology, and of the Glickman Urological & Kidney Institute’s Department of Urology.

Prostate cancer provides an illustration of the complexities of drug resistance, and of the important role of steroid receptors.

**Development of Castration-resistant Tumors**

Standard treatment for metastatic prostate cancer is androgen deprivation therapy via medical or surgical castration. However, tumors eventually become castration-resistant. This resistance stems from tumors’ increased expression of the androgen receptor, and from the acquired ability to manufacture their own supply of 5α-dihydrotestosterone (DHT), the most potent androgen, from precursor steroids. Dr. Sharifi’s
own recent research identified the first mutation responsible for increasing DHT synthesis.

Patients treated with the androgen-synthesis inhibitor abiraterone and the powerful androgen-receptor antagonist enzalutamide have shown increased survival. But even this new generation of hormonal therapies targeting the androgen receptor has fleeting success. Within days of exposure to enzalutamide, according to research by Vivek Arora, MD, PhD, Charles Sawyers, MD, and colleagues at Memorial Sloan-Kettering Cancer Center, a subset of prostate cancer cells is triggered to begin glucocorticoid receptor expression. This rapidly progresses to massive receptor upregulation and the development of enzalutamide resistance.

In essence, Dr. Sharifi writes in NEJM, when the next-generation hormonal therapies double down on the androgen receptor, the tumor apparently responds by producing an alternative steroid receptor that takes on at least part of the androgen receptor’s job.

Rethinking Interventions

While the androgen receptor is not essential for life, the glucocorticoid receptor is. That poses challenges for testing pharmacologic interventions involving the glucocorticoid receptor, and also requires a rethinking of current clinical practice, which relies on glucocorticoids as an important prostate cancer treatment.

If tumor expression of the glucocorticoid receptor spurs clinical resistance to enzalutamide, might there be other impacts? Dr. Sharifi asks in the NEJM article. “Is it possible that glucocorticoid administration has direct effects on the tumor that may be detrimental to the patient in other clinical settings?”

Future Investigative Approaches

Answers to these questions will depend in part on the collection of tumor tissue from patients who have castration-resistant prostate cancer, Dr. Sharifi says. This will permit researchers to identify and dissect the molecular signaling mechanisms that drive resistance, including the possibility that this might involve the glucocorticoid receptor.

“The androgen receptor and glucocorticoid receptor are both steroid receptors,” Dr. Sharifi says. “Much is already known about how androgens and the androgen receptor are central drivers of tumor growth and enable resistance to various standard hormonal therapies. Two investigative approaches may help bring these findings closer to clinical practice. The first is to identify how clinical tumors modulate glucocorticoid receptor signaling through various standard therapies. The second is to ask whether modulating glucocorticoid receptor signaling alters tumor progression in clinical settings where this receptor is thought to be important.”
Multiparametric Magnetic Resonance Imaging Ultrasound (MP-MRI-US) Fusion Targeted Prostate Biopsy

Accurate localization/characterization of clinically important prostate cancer lesions among healthy men who are otherwise candidates for curative therapy is essential for successful active surveillance and focal therapy strategies.

Indeed, progression rates on active surveillance as high as 48 percent over short (<5 years) time intervals have been reported, and rates of advanced pathological features (Gleason score ≥ 4+3, extraprostatic extension, seminal vesicle invasion or lymph node metastasis) as high as 23 percent have been reported among men undergoing deferred radical prostatectomy.

The limitations of transrectal biopsy strategies to accurately characterize and localize prostate cancer are highlighted by the consistent 20 percent to 30 percent reclassification rate among low-risk men undergoing immediate repeat extended prostate biopsy (10 to 14 cores) on active surveillance protocols.

Likewise, among men undergoing repeat saturation biopsy (≥20 cores) after one and two prior negative biopsies, we reported a cancer detection rate of 33 percent and 19 percent, respectively. Among men with known low-risk prostate cancer undergoing three-dimensional transperineal mapping biopsy (3D-TPMB), a 20 percent negative biopsy rate has been reported despite 50- to 69-core sampling. Molecular tests such as Prolaris® and Oncotype Dx® may provide useful information regarding prostate cancer staging and risk, but their utility in patient selection for active surveillance and focal therapy (and subsequent monitoring) is unproven.

Imaging Advances Improve Diagnostic Accuracy

Advances in prostate imaging using multiparametric magnetic resonance imaging (MP-MRI) and the ability to apply this information to targeted biopsy strategies using transrectal ultrasound (termed MP-MRI-US fusion biopsy) have the potential to improve our ability to accurately localize and characterize individual prostate cancer lesions.

Advances in MRI such as improved anatomical resolution on T1 and T2 weighted images (T2WI) with the use of 3 Tesla magnets, and functional imaging sequences using diffusion-weighted imaging (DWI), dynamic contrast enhancement (DCE) and MR spectroscopic imaging (MRSI), allow better diagnostic accuracy for prostate cancer (Figure 1).

DCE allows for the visualization of blood perfusion via a bolus injection of gadolinium contrast during rapidly repeated scanning. Evidence of early and intense enhancement and washout in lesions is associated with angiogenesis seen in prostate cancers. DWI quantifies free water motion, which is restricted in lesions with increased cellularity, as is seen in prostate cancers. MRSI measures levels of choline (increased in cancer) relative to creatine and citrate peaks, and increases the specificity of low-signal intensity lesions seen on T2WI.

However, the incremental benefit of MRSI to DWI and DCE sequences appears to be limited. The use of MP-MRI for prostate cancer detection and characterization has been aided by a standardized grading system called PI-RADS that rates regions of interest on a scale of 1 to 5 based on the likelihood of clinically significant cancer. MP-MRI using the
PI-RADS scoring system has shown a sensitivity and specificity for prostate cancer detection of 67 percent and 92 percent, respectively, with 85 percent diagnostic accuracy.

The use of MP-MRI for targeted prostate biopsy has been limited by the time, expense and impracticality of performing biopsies under real-time MRI guidance in the MRI gantry. Likewise, the use of MP-MRI to perform targeted biopsies of suspected lesions using standard transrectal ultrasound (TRUS), or so-called cognitive recognition, has questionable accuracy.

**Imaging Fusion Results in Better Biopsy**

Technological developments now enable MP-MRI images to be “fused” with TRUS using specialized computer software to improve targeted biopsy of suspicious lesions. Various tracking systems have been described, including a 3-D US probe attached to a mechanical arm (Figure 2), and one that uses an electromagnetic (EM) field generator placed above the pelvis with a custom US probe embedded with a passive EM tracking sensor. The best approach has yet to be determined.

Using MP-MRI-US fusion targeted biopsy, cancer detection rates of 15 percent to 20 percent, 29 percent to 40 percent, and 50 percent to 71 percent have been reported among lesions classified as low, moderate, and high probability of cancer, respectively. Likewise, targeted biopsies are more likely to detect high-grade cancers (38 percent of which were missed by standard 12-core biopsy in one study). Targeted biopsy strategies also appear to be as accurate as 3D-TPMB for detecting clinically significant cancers. On average, 6 to 13 additional biopsies are taken per patient when a targeted biopsy strategy is added to a standard 12-core biopsy.

MP-MRI-US fusion targeted biopsy has the potential to significantly improve the localization and characterization of prostate cancer when added to a standard 12-core biopsy. It is anticipated that this strategy will enable better selection and monitoring of patients who choose active surveillance and focal therapy as a management option. This technology may also be useful in patients with suspicion of prostate cancer despite one or more negative biopsies. As of mid-2014, Cleveland Clinic will offer MP-MRI-US fusion targeted prostate biopsy to patients. We also plan to investigate its utility in active surveillance and focal therapy protocols.

*For references, please email the author.*
Despite advances in surgery, radiation therapy and chemotherapy, glioblastoma remains an incurable malignancy with a dismal median overall survival of 15 to 18 months. Preclinical studies have shown that when properly tuned, very-low-intensity, intermediate-frequency electric fields — tumor treating fields (TTFields) — can inhibit the growth of tumor cells. Cleveland Clinic’s Rose Ella Burkhardt Brain Tumor and Neuro-Oncology Center is taking a leading role in studying and using this emerging field of therapy.

The Essentials of Electric Fields

In the laboratory setting and clinical practice, electric fields show a wide range of effects on living tissues. At very low frequencies (< 1 kHz), electric fields stimulate excitable tissues through membrane depolarization. Well-known examples of such effects include nerve, muscle and heart stimulation by electric fields. In addition, low-frequency pulsed electric fields have been claimed to stimulate bone growth and accelerate fracture healing. However, as the frequency of the electric field increases above 1 kHz, the stimulatory effect diminishes.

At very high frequencies (i.e., above many MHz), while the integration becomes even more effective, a completely different biological effect is observed: Tissue heating becomes dominant due to dielectric losses. This phenomenon is the basis for commonly used medical treatment modalities, including diathermy and radiofrequency tumor ablation, which can be applied through insulated electrodes.

Intermediate-frequency electric fields (i.e., tens of kHz to MHz) alternate too fast to cause nerve-muscle stimulation and involve only minute dielectric losses (heating). Such fields, of low to moderate intensities, are commonly considered to have no biological effect. However, a number of nonthermal effects, of minor biological consequence, have been reported even at low field intensities. These effects include microscopic particle alignment (i.e., the pearl chain effect) and cell rotation. With pulsed relatively strong electric fields (> 103 V/cm and 100-ms pulse length), reversible pore formation appears in the cell membrane, a phenomenon usually called electroporation.

Biological molecules are dipoles (composed of positive and negative charges), and dipole movement can occur with application of an external electric field. In situations where biological processes require precise spatial alignment, such as mitosis, externally applied electric fields can disrupt this process. This was the hypothesis that led to the initial work evaluating the ability of electric fields to disrupt mitosis in cancer cells.

Preclinical Rationale for TTFields Therapy

In laboratory studies, TTFields have demonstrated an inhibitory effect in all proliferating cancer cell types tested but had no effect on nonproliferating cells and normal tissues. Interestingly, different cell types showed specific intensity and frequency dependencies of TTField inhibition.

Two main processes occur at the cellular level during exposure to TTFields: arrest of proliferation

---

**Figure 1. Mechanism of action of tumor treating fields therapy.**

(A) Disruption of the formation of the mitotic spindle in metaphase. (B) Positive dielectrophoresis during anaphase.

and dividing cell destruction. The damage caused by TTFields to these replicating cells was dependent on the orientation of the division process in relation to the field vectors, indicating that this effect is nonthermal.

At the subcellular level it was found that TTFields disrupt the normal polymerization-depolymerization process of microtubules during mitosis (Figure 1). The described abnormal mitotic configurations seen after exposure to TTFields are similar to the morphological abnormalities seen in cells treated with agents that interfere directly or indirectly with microtubule polymerization (e.g., paclitaxel).

**TTFields in Action**

The NovoTTF-100A System (Novocure; Haifa, Israel) is a portable battery-operated device (Figure 2) that produces TTFields within the human body using surface electrodes (transducer arrays). The TTFields are delivered to the patient by surface transducer arrays that are electrically insulated so that resistively coupled electric currents are not delivered to the patient. The transducer arrays, which incorporate a layer of adhesive hydrogel and a layer of hypoallergenic medical tape, are placed on the patient’s shaved head. The arrays must be replaced every three to four days and the scalp reshaved to maintain optimal capacitative coupling between the arrays and the patient’s head.

All treatment parameters are preset by the manufacturer so there are no electrical output adjustments available to the patient. The patient must learn to apply and change transducer arrays, change and recharge depleted device batteries, and connect to an external electrical outlet.

**Key Clinical Trial**

A prospective, randomized, open-label trial with active parallel control was conducted to compare effectiveness and safety outcomes between patients with recurrent glioblastoma treated with NovoTTF-100A monotherapy (n = 120) and those treated with an effective best-standard-of-care chemotherapy (n = 117). NovoTTF-100A patients had comparable overall survival to patients receiving the best available chemotherapy in the U.S. today. Patients randomized to the device demonstrated fewer side effects and improved quality of life measures relative to the chemotherapy group. This led to U.S. marketing approval of the device in 2011 for use as monotherapy for patients with recurrent glioblastoma.

Cleveland Clinic participated in this study, and physicians in our Rose Ellaurkhardt Burkhardt Brain Tumor and Neuro-Oncology Center were among the first in the United States to prescribe the device for patients with recurrent glioblastoma.

**New Trials of Combined Use with Bevacizumab**

The Burkhardt Center is leading an ongoing multicenter study evaluating the efficacy of the NovoTTF-100A device in combination with bevacizumab, an FDA-approved therapy for patients with recurrent glioblastoma. Cleveland Clinic investigators are also leading a planned large international trial of the combination of NovoTTF-100A with bevacizumab for patients with recurrent glioblastoma that has failed to respond to bevacizumab alone. We look forward to the results from each of these investigations.

**References**


A new Cleveland Clinic study has found that men who undergo therapeutic radiation treatment for prostate cancer have no greater risk of developing myelodysplastic syndromes (MDS) than the general population.

Senior study author Mikkael A. Sekeres, MD, MS, of the Department of Hematologic Oncology and Blood Disorders in Cleveland Clinic’s Taussig Cancer Institute, says the findings came as a surprise.

Delivery of external beam radiation to the prostate inadvertently irradiates the surrounding pelvic bones, which are estimated to contain more than half the body’s reserve of active bone marrow mass.

“The medical community had long assumed that patients who received radiation therapy to treat prostate cancer were at a higher risk for developing MDS,” Dr. Sekeres says. “The reason for that is quite simply geography, since the prostate is right in the middle of the pelvis, and the pelvis is a major site of bone marrow production in the body.”

Study Details

The study, published recently in the *Journal of the National Cancer Institute*, was a joint effort between Taussig Cancer Institute and the Glickman Urological & Kidney Institute. The retrospective cohort analysis was based on data from 10,924 prostate cancer patients who were treated at Cleveland Clinic from 1986 to 2011 with either surgery, radiotherapy with external beam radiation (EBRT), or radiation therapy with brachytherapy.

A total of 31 cases of therapy-related MDS were observed during the study period. In multivariable analyses, MDS rates were similar in patients who underwent surgery for prostate cancer compared with those who received some type of radiotherapy. The MDS rates observed in the study were found to be comparable to rates in population-based registries, including the Ohio Cancer Incidence Surveillance System (OCISS) and the Surveillance, Epidemiology, and End Results (SEER) database.

Advancing age is an independent risk factor for developing MDS, which the study analyses confirmed.

**Clinical Significance**

MDS is the most common bone marrow failure condition in the United States, with an age-adjusted incidence rate of 4.4 per 100,000 people. The majority of cases are spontaneous, but approximately 14 percent are considered therapy-related, occurring in cancer patients on average five to seven years after treatment with cytotoxic chemotherapy, radiation or both.

Exposure to ionizing radiation had been linked to subsequent development of MDS in several cancer cohorts. But until this study, the risk of MDS in prostate cancer patients treated with therapeutic radiation had remained unclear.

“What’s been problematic is that when an older man who has a history of receiving radiation for prostate cancer develops MDS, the radiation has been viewed as the cause for MDS — when it most likely was unrelated,” Dr. Sekeres explains. “Based on the results of our study, patients receiving radiation for prostate cancer can rest assured that they do not have excess risk for developing MDS within their lifetime.”

As patients were collected over a 25-year period, follow-up was limited. However, given the median patient age of 64 and average life expectancies, Dr. Sekeres says any influence that therapeutic radiation for prostate cancer might or might not have beyond the study’s follow-up period would likely be “clinically meaningless.”
Medical Innovation Summit to Focus on Cancer Technologies

Advances in cancer diagnosis and treatment and the advent of individualized, genome-based risk prediction and therapies will be the subject of Cleveland Clinic’s 2014 Medical Innovation Summit.

“Now, It’s Personal: Cancer Treatment and Personalized Medicine” is the theme of the three-day conference, to be held Oct. 27 to Oct. 29 at the Cleveland Convention Center, 300 Lakeside Avenue.

“There are many more cancer survivors today than ever before,” says Brian J. Bolwell, MD, FACP, Chairman of Cleveland Clinic’s Taussig Cancer Institute and of the event’s Steering Committee. “The timing is absolutely right to have an Innovation Summit about cancer medicine. It’s a unique opportunity to learn and to interact.”

The annual conference brings together physicians, researchers, healthcare executives, investors, political leaders, academics, and business, legal, regulatory and medical affairs officials to learn about and discuss leading-edge medical topics. A highlight is the announcement of the Top 10 Medical Innovations — the technologies that Cleveland Clinic physicians and researchers believe will have the greatest patient benefit, the most public interest and the highest likelihood of commercial success in the coming year.

To register, go to clevelandclinic.org/summit

Summit agenda topics include:

- Investing in Cancer: Why It’s Worth the Risk
- The Future of Genomics in Medicine
- The Business of Cancer Risks: You’re Well Now. Are You Going to Get Sick?
- Personalized Medicine Today: You Have Cancer. Now What?
- Cancer Around the World
- The Future of Cancer Imaging: The Next Generation Software and Hardware
- Policy’s Impact on Cancer Patients
- The Future of Breast Cancer
- Prevention, Diagnosis and Treatment of Prostate Cancer
- Targeted Therapies for Leukemia & Lymphomas
- The Next Generation of Cancer Drug Development: Tailored Treatments
- Radiation Therapy: The Role of Photons, Protons and Particles
The Cleveland Clinic Way

By Toby Cosgrove, MD,
CEO and President of Cleveland Clinic

Great things happen when a medical center puts patients first. Visit clevelandclinic.org/ClevelandClinicWay for details or to order a copy.

Resources for Physicians

Physician Directory
View all Cleveland Clinic staff online at clevelandclinic.org/staff.

Same-Day Appointments
Cleveland Clinic offers same-day appointments to help your patients get the care they need, right away. Have your patients call our same-day appointment line, 216.444.CARE (2273) or 800.223.CARE (2273).

Track Your Patients’ Care Online
Establish a secure online DrConnect account for real-time information about your patients’ treatment at Cleveland Clinic at clevelandclinic.org/drconnect.

Critical Care Transport Worldwide
To arrange for a critical care transfer, call 216.448.7000 or 866.547.1467. Visit clevelandclinic.org/criticalcaretransport to learn more.

CME Opportunities: Live and Online
Visit ccfcmoe.org to learn about the Cleveland Clinic Center for Continuing Education’s convenient, complimentary learning opportunities.

24/7 Referrals

Referring Physician Hotline
855.REFER.123 (855.733.3712)

Hospital Transfers
800.553.5056

On the Web at:
clevelandclinic.org/Refer123

Stay connected with us on …

Outcomes Data
View Outcomes books at clevelandclinic.org/outcomes.

Clinical Trials
We offer thousands of clinical trials for qualifying patients. Visit clevelandclinic.org/clinicalcancertrials.

Download Our New Physician Referral App!
Contacting us is now easier than ever.

With our free Physician Referral App, you can view all our specialists and get in touch immediately with one click of your iPhone®, iPad®, or Android™ phone or tablet.

Download today at the App Store or Google Play.

Cleveland Clinic is an integrated healthcare delivery system with local, national and international reach. At Cleveland Clinic, more than 3,000 physicians and scientists represent 120 medical specialties and subspecialties. We are a main campus, 18 family health centers, eight community hospitals, Cleveland Clinic Florida, the Cleveland Clinic Lou Ruvo Center for Brain Health in Las Vegas, Cleveland Clinic Canada, Sheikh Khalifa Medical City, and Cleveland Clinic Abu Dhabi.

In 2013, Cleveland Clinic was ranked one of America’s top hospitals in U.S. News & World Report’s annual “America’s Best Hospitals” survey. The survey ranks Cleveland Clinic among the nation’s top 10 hospitals in 14 specialty areas, and the top hospital in three of those areas.

The Cleveland Clinic
2500 South Point Drive
Cleveland, OH 44129
216.621.4000
www.clevelandclinic.org