Published OnlineFirst August 25, 2009; DOI: 10.1158/0008-5472.CAN-09-1795

Meeting Report

2009 Biospecimen Research Network Symposium: Advancing Cancer Research through Biospecimen Science

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Abstract

This report details the proceedings of the 2009 Biospecimen Research Network (BRN) Symposium that took place on March 16 to 18, 2009, the second in a series of annual symposia sponsored by the National Cancer Institute Office of Biorepositories and Biospecimen Research. The BRN Symposium is a public forum addressing the relevance of biospecimen quality to progress in cancer research and the systematic investigation needed to understand how different methods of collection, processing, and storage of human biospecimens affect subsequent molecular research results. More than 300 participants from industry, academia, and government attended the symposium, which featured both formal presentations and a day of workshops aimed at addressing several key issues in biospecimen science. An additional 100 individuals participated via a live webcast (archived at http://brnsymposium.com). The BRN Symposium is part of a larger program designed as a networked, multidisciplinary research approach to increase the knowledge base for biospecimen science. Biospecimens are generally understood to represent an accurate representation of a patient's disease biology, but can instead reflect a combination of disease biology and the biospecimen's response to a wide range of biological stresses. The molecular signatures of disease can thus be confounded by the signatures of biospecimen biological stress, with the potential to affect clinical and research outcomes through incorrect diagnosis of disease, improper use of a given therapy, and irreproducible research results that can lead to misinterpretation of artifacts as biomarkers. Biospecimen research represents the kind of bricks-and-mortar research that provides a solid scientific foundation for future advances that will directly help patients. [Cancer Res 2009;69(17):6770–2]

Introduction

Significant efforts in translational research aim to reverse the rising tide of cancer morbidity and mortality by using clinical and molecular data from individual patients to develop and validate targeted therapies, treat patients with greater specificity, reduce adverse events, and determine disease predisposition to allow early detection and prevention of cancer. At the center of this personalized medicine approach are the human biospecimens that provide the critical link between the information encoded in the molecular signatures of an individual's specific cancer and clinical action that is based on that information. However, progress requires that the biospecimens used in translational research and personalized medicine must be of the highest quality. At present, variability pervades the collection, processing, storage, and annotation of the majority of human biospecimens available for research. Such heterogeneous practices generate biospecimens of unknown molecular integrity and contribute to irreproducible research results, impeding the development of more effective therapeutics and diagnostics.

To speed progress in cancer research, the National Cancer Institute (NCI) is addressing the issue of biospecimen quality through its Office of Biorepositories and Biospecimen Research (OBBR). To help the United States move toward standardized procedures for biospecimen collection, processing, and storage, OBBR issued the *NCI Best Practices for Biospecimen Resources* in 2007, encouraging the use of appropriate standardized protocols and quality assurance/quality control procedures. To generate the data needed for evidence-based biospecimen practices, OBBR established the Biospecimen Research Network (BRN) to coordinate and support systematic investigation into how collection, processing, and storage of human biospecimens affect subsequent molecular analysis.

To accelerate the development of appropriate biospecimen-related technologies and methodologies, and to provide input to the BRN as it continues to shape its support of biospecimen research, the NCI held the 2nd Annual BRN Symposium: Advancing Cancer Research Through Biospecimen Science on March 16 to 18, 2009, in Bethesda, Maryland. More than 300 participants from industry, academia, and government attended the symposium, which featured both formal presentations and a day of workshops aimed at addressing several key issues in biospecimen science. An additional 100 individuals participated via a live webcast (archived at http://brnsymposium.com).[5] This Meeting Report summarizes recent scientific findings germane to the development of sound, reproducible, evidence-based methods for collecting, processing, storing, and annotating biospecimens from cancer patients.

Attacking Preanalytic Variability

A major disappointment in oncology today is the continued difficulty in identifying reproducible molecular signatures that would serve as diagnostics for the early detection of cancer or as predictive/prognostic markers of efficacy for specific therapies in specific patients. Elizabeth Hammond (University of Utah School of

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doi:10.1158/0008-5472.CAN-09-1795

[5] Office of Biorepositories and Biospecimen Research, National Cancer Institute. 2nd Annual Biospecimen Research Network Symposium [cited 2009 May 8]. Available from: http://brnsymposium.com/meeting/brnsymposium/2009/webcast-registration.asp.

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Medicine, Salt Lake City, UT) underscored this issue by noting that although there are some 660 commercial tests available for measuring germ-line mutations and gene alterations, there are only 30 or so tests available that could be effectively used to predict patient response to a specific therapy. This disparity arises in part from the lack of standardized procedures for collecting, processing, storing, and annotating biospecimens from cancer patients. Consequently, biomarker-directed research based on these biospecimens is built on a shifting experimental foundation, making it difficult to compare results from different experiments, let alone different laboratories. David Agus (Cedars-Sinai Medical Center, Los Angeles, CA) noted that the poor state of biospecimen-related procedures has a profoundly negative effect on patient care by making it more difficult for cancer drug developers to fairly and accurately assess whether new therapies actually would be effective for particular groups of patients.

The only way to reduce preanalytic variability is to implement standard operating procedures (SOP) developed with input from all stakeholders in the biospecimen arena. That is the approach that Utah's Intermountain Healthcare took to address the unacceptable variability in estrogen receptor assay results seen across its 27 hospitals performing breast tumor resection. After determining that the variable results stemmed from a lack of shared biospecimen SOPs, Hammond and her colleagues developed standardized protocols and then worked with hospital leaders to garner support for and eventually adoption of these SOPs by surgeons and pathologists. Consequently, variability in estrogen receptor assay results across the Intermountain Healthcare system has been reduced significantly, increasing the accuracy and reproducibility of this assay for patient care. Such fundamental improvements can be difficult to implement but have direct benefit for cancer patients.

Hartmut Juhl (Indivumed GmbH, Hamburg, Germany) described the extensive SOPs that his company developed for biospecimen handling, noting that more than half of the company's employees are involved in carrying out these protocols through contractual agreements with hospitals, surgical clinics, and oncology units in the United States and Germany. Using this labor-intensive process, the company has built a biobank containing well-annotated biospecimens from 10,000 cancer patients and is adding samples from new patients at a rate of 2,000 per year. The company uses these biospecimens to improve its understanding of how preanalytic handling of biospecimens affects the reproducibility of analytic results and to search for new prognostic biomarkers.

Juhl and others presented data describing the effect of specific preanalytic variables on biospecimen quality. One of the biggest effects noted by these speakers arises from differences in the time between tissue harvesting and stabilization, either by rapid cooling or with chemical fixatives. A common theme of these presentations was that it is critical to reduce to a minimum the amount of time that harvested tissues spend under ischemic conditions before stabilization. Many speakers called for the development of molecular markers for biospecimen integrity that could be used to identify those biospecimens unsuitable for more than histopathology-based diagnostic procedures. David Rimm (Yale University School of Medicine, New Haven, CT) reported on a BRN-sponsored research project to develop a tissue immunologic competence model for formalin-fixed, paraffin-embedded (FFPE) tissue. This research will identify multiple proteins whose expression is compromised as a function of postsurgical ischemic time and hypoxia. Katy Williams (University of California at San Francisco, San Francisco, CA), also sponsored by the BRN, is establishing reference ranges for candidate markers of protein damage in serum and plasma samples, including indicators of proteolysis, oxidation, and aggregation. These data will be used to create a panel of reference markers that can be easily assayed in serum or plasma as acute sentinels of serum and plasma degradation.

Gennady Bratslavsky (NCI, Bethesda, MD) showed that surgical techniques have an effect on gene expression in renal carcinoma biospecimens. During laparoscopic surgery, the tumor sits at body temperature for a considerable period of time once it has been disconnected from the local blood supply. Thus the tissue is subjected to extended ischemia times at body temperature, a situation that differs greatly from when biospecimens are harvested using open surgical techniques. Dr. Bratslavsky's data suggested that it will be essential to account for ischemia-sensitive genes when evaluating renal biomarkers for prognostication and therapeutics. In another presentation, Douglas Clark (Johns Hopkins University School of Medicine, Baltimore, MD) found that fine-needle aspirations produce variable results that depend both on the surgeon performing this type of biopsy and on where in the tumor the needle is inserted. Nonetheless, fine-needle aspirations minimize ischemia and produce biospecimens comprising live cells that can be quick frozen, and may provide more accurate assessments of biochemical pathway function and tumor drug sensitivity than is possible using standard FFPE tissue slices.

However, as Guido Hennig (Siemens Healthcare Diagnostic Products, Cologne, Germany) showed, FFPE biospecimens can yield valuable prognostic information. Because of the widespread availability of FFPE biospecimens, Siemens has taken the approach of developing prognostic gene profile markers for breast cancer using fresh-frozen tissue and then using that data to create a prognostic algorithm that would be applied to FFPE biospecimens. The company plans to conduct independent retrospective and prospective randomized clinical trials to see if the algorithm translates from fresh-frozen tissue to FFPE tissue.

Experimental Design in Biospecimen Science

Several speakers noted that poor experimental design and improper use of statistical techniques can be problematic in biospecimen science and biomarker research. Terry Speed (University of California at Berkeley, Berkeley, CA) spoke to the importance of randomization, replication, and local control of variability when designing experiments to look for clinically relevant molecular signatures. He noted in particular the value of factorial experimental designs that are both statistically powerful and efficient in their use of valuable tissue samples, and he proposed a two-phase experimental design that would be sufficiently powered to explore the effect of changing multiple preanalytic variables, including patient, acquisition, processing, and storage variables.

Sound experimental design was critical to the success of a biomarker validation study presented by Scott Patterson (Amgen, Thousand Oaks, CA). This particular phase III trial examined whether KRAS was a predictive marker for response to panitumumab (Vectibix), an epidermal growth factor receptor (EGFR) inhibitor. KRAS is part of the EGFR signaling cascade, and prior studies suggested that the presence of mutant KRAS correlates with poor prognosis. Because of the forethought put into the phase III trial, there were sufficient numbers of high-quality, well-annotated biospecimens to test the hypothesis that

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KRAS could serve as a biomarker for progression-free survival. KRAS mutations were detected using a commercially available kit that met performance characteristics set by the Clinical and Laboratory Standards Institute, and the statistical analysis plan was finalized before unblinding the biospecimens' KRAS status. When the analysis was complete, it was clear that the hypothesis was correct and that the magnitude of the interaction between KRAS status and outcome was substantial.

NCI Programmatic Activity

NCI speakers reviewed several key activities designed to support biospecimen science. One such tool under development by the OBBR is the Biospecimen Research Database (BRD), a publicly available and searchable Web-based literature database containing published and peer-reviewed data pertinent to biospecimen science. The database is curated to highlight and summarize those results that provide further insights into the field of biospecimen science, and it is indexed based on the variables addressed and the biospecimens and technology platforms used. The goals of the BRD are to make existing and emerging biospecimen research data more accessible for users who are conscious of the potential confounding effects of biospecimen preanalytic variables on their research and to increase awareness of biospecimen effects on the results of molecular and histologic analyses.

To date, the BRD contains 155 research articles published from 1985 to 2008 in 75 peer-reviewed journals. Another 100 articles await secondary curation before being added to the database, and an additional 450 articles have been identified for inclusion in the BRD. The articles in the BRD represent 33 tissue types, 54 technology platforms, and 113 disease diagnoses. The BRD can be accessed at http://biospecimens.cancer.gov/brd.[6] Moving forward, the NCI will continue populating the database with pertinent and new research topics and conduct outreach activities to expand the user population. Plans are also being developed to conduct meta-analyses of curated articles to develop consensus recommendations and SOPs. Efforts are also under way to integrate the BRD with the cancer Biomedical Informatics Grid (caBIG) and to link with data from BRN-sponsored studies.

NCI staff also announced five new NIH Challenge Grants topics stemming from the American Recovery and Reinvestment Act of 2009. Proposals under these topics are currently under review. The five challenge areas and biospecimen-specific topics include the following:

- Bioethics: Unified informed consent document for biobanking and subsequent analysis of human biospecimens.
- Bioethics: Optimizing the timing of consent for biobanking to achieve ethical and research objectives.
- Biomarker discovery and validation: Biospecimen research to improve biomarker identification and validation.
- Clinical research: Biospecimen standardization for clinical assays.
- Genomics: Genomic changes introduced by biospecimen preanalytic variables.

More information on these challenge grants is available online.[7]

The meeting concluded with a discussion of the cancer Human Biobank (caHUB), envisioned as the first national biorepository in the United States. caHUB will be a unique, centralized public resource designed to ensure the adequate and continuous supply of high-quality human biospecimens to accelerate cancer research. caHUB will modernize the field of biobanking by acquiring and making available to the research community biospecimens that have been collected according to the highest technical and ethical standards, providing biospecimen reference samples that serve as benchmarks for specimen integrity and molecular type, conducting research that supports evidence-based biospecimen best practices, and creating opportunities for collaboration and information exchange across the research enterprise. In addition, caHUB will ensure the quality of its inventory by acquiring biospecimens that have been collected and processed according to evidence-based SOPs, annotated with comprehensive clinical, molecular, and collection data, and procured from patients who received high-quality care. The transformative nature of caHUB was recently recognized by Time Magazine as one of the top 10 "Ideas Changing the World Right Now" (March 23, 2009).

Conclusion

The BRN Symposium is part of a larger program designed as a networked, multidisciplinary research approach to increase the knowledge base about how different methods of biospecimen collection, processing, and storage alter the biological picture presented by a given biospecimen. Although generally understood to represent an accurate representation of a patient's disease biology, the biospecimen instead reflects a combination of disease biology and the biospecimen's response to a wide range of biological stresses. Thus, the molecular signatures of disease can be confounded by the signatures of biospecimen biological stress, with the potential to affect clinical and research outcomes through incorrect diagnosis of disease, improper use of a given therapy, and irreproducible research results that can lead to misinterpretation of artifacts as biomarkers. Understanding biospecimen stress-related effects is critical to making advances in translational research. Biospecimen research represents the kind of bricks-and-mortar research that provides a solid scientific foundation for future advances that will directly help patients.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Acknowledgments

Received 5/21/09; revised 6/24/09; accepted 6/24/09; published OnlineFirst 8/25/09.

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[6] Office of Biorepositories and Biospecimen Research, National Cancer Institute. The Biospecimen Research Database [cited 2009 May 8]. Available from: https://brd.nci.nih.gov.

[7] Office of Extramural Research, NIH, and the U.S. Department of Health and Human Services. NIH Challenge Grants in Health and Science Research [cited 2009 May 8]. Available from: http://grants.nih.gov/grants/funding/challenge_award/.

Cancer Research
The Journal of Cancer Research (1916–1930) | The American Journal of Cancer (1931–1940)

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Cancer Res 2009;69:6770-6772. Published OnlineFirst August 25, 2009.