**U.S. Department of Health and Human Services**
**Public Health Service**
**National Institutes of Health**
**National Cancer Institute**

8th Virtual Meeting
Frederick National Laboratory Advisory Committee

# Summary of Meeting
**October 18, 2021**

**National Cancer Institute**
**National Institutes of Health**
**Bethesda, Maryland**

**National Cancer Institute**
**8th Virtual Meeting of the Frederick National Laboratory Advisory Committee**

**18 October 2021**

**Summary of Meeting**

The Frederick National Laboratory Advisory Committee (FNLAC) convened for its 8th Virtual Meeting on 18 October 2021. The meeting was open to the public on 18 October 2021, from 1:00 p.m. to 4:00 p.m. EDT. The FNLAC Chairperson, Dr. Candace. S. Johnson, President and CEO, M&T Bank Presidential Chair in Leadership, Roswell Park Comprehensive Cancer Center, presided.

**FNLAC Members**
Dr. Candace. S. Johnson (Chair)
Dr. Andrea H. Bild*
Dr. Catherine M. Bollard
Dr. John H. Bushweller*
Dr. Timothy A. Chan
Dr. Lisa M. Coussens
Dr. Channing J. Der* (absent)
Dr. Raymond N. DuBois
Dr. Scott W. Hiebert
Dr. Allison Hubel
Dr. Dineo Khabele*
Dr. Nilsa C. Ramirez Milan
Dr. Denise J. Montell
Dr. Patrick Nana-Sinkam
Dr. Lincoln D. Stein
Dr. Linda F. van Dyk

*Pending Appointment

***NCI Senior Leadership***
Dr. Stephen J. Chanock (absent)
Dr. James H. Doroshow
Dr. Paulette S. Gray
Dr. Anthony Kerlavage (absent)
Dr. Kristin Komschlies
Dr. Douglas R. Lowy
Dr. Tom Misteli (absent)
Ms. Donna Siegle
Dr. Dinah S. Singer

**Executive Secretary**
Dr. Wlodek Lopaczynski

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## I. OPENING REMARKS—DR. CANDACE S. JOHNSON

Dr. Candace. S. Johnson, Chair, called to order the 8th Virtual Meeting of the Frederick National Laboratory Advisory Committee (FNLAC) and welcomed the Committee members, National Cancer Institute (NCI) staff, and guests. Dr. Johnson reminded members of the conflict-of-interest guidelines and confidentiality requirements. Members of the public were welcomed and invited to submit to Dr. Wlodek Lopaczynski, Executive Secretary, in writing and within 10 days, any comments regarding items discussed during the meeting.

**Motion.** A motion to approve the minutes of the 28 June 2021 FNLAC meeting was approved unanimously.

Dr. Johnson called Committee members' attention to the future meeting dates listed on the agenda. She also noted that the next FNLAC meeting will be held on 24 February 2022 and will be virtual and that the 10–11 July 2022 and 12–13 October 2022 meetings currently are planned to be held in person.

**Motion.** A motion to confirm the 2022 and 2023 FNLAC meeting dates was approved unanimously.

## II. NCI DIRECTOR'S REPORT—DR. NORMAN E. SHARPLESS

Dr. Norman E. Sharpless, Director, NCI, also welcomed the FNLAC members and attendees to the meeting. He provided updates on the NCI activities, NCI budget, activities related to coronavirus disease 2019 (COVID-19), NCI programs and initiatives, and progress in cancer research.

**New And Retiring FNLAC Members.** Dr. Sharpless welcomed Dr. Johnson, FNLAC member since May 2021, as the new Chair. He acknowledged the new FNLAC members: Dr. Andrea H. Bild, Professor, Division of Molecular Pharmacology, Department of Medical Oncology and Therapeutics, City of Hope Comprehensive Cancer Center; Dr. John H. Bushweller, Professor, Department of Molecular Physiology and Biological Physics, School of Medicine, University of Virginia; Dr. Channing J. Der, Sarah Graham Kenan Distinguished Professor, Department of Pharmacology, Lineberger Comprehensive Cancer Center, University of North Carolina at Chapel Hill; Dr. Allison Hubel, Professor, Department of Mechanical Engineering, University of Minnesota; Dr. Dineo Khabele, Professor and Chair, Department of Obstetrics and Gynecology, School of Medicine, Washington University in St. Louis; and Dr. Linda F. van Dyk, Professor and Vice-Chair, Department of Immunology and Microbiology, University of Colorado Anschutz Medical Campus.

On behalf of the NCI, Dr. Sharpless recognized the contributions made by Dr. Raymond N. DuBois, Dean, College of Medicine, Professor, Departments of Biochemistry and Medicine, Medical University of South Carolina, whose term of office has expired. He expressed appreciation for Dr. DuBois' service and dedication over the course of his term on the Committee.

**National Cancer Act of 1971 (NCA) 50th Anniversary.** Dr. Sharpless remarked that the NCI is continuing to commemorate the 50th anniversary of the NCA of 1971 (NCA-50), an Act that increased funding for and made cancer a topic discussed publicly. The NCA of 1971 also enabled many principal authorities for the NCI, including establishing the Frederick National Laboratory for Cancer Research (FNLCR). The NCI has been joined by other groups across the cancer community, including the NCI-Designated Cancer Centers (Cancer Centers) and the American Association for Cancer Research (AACR) in this national media campaign, to reflect on five decades of progress. Dr. Sharpless commented that the NCA-50 is not just about the NCI; it is about everyone who has an investment in cancer research, from

academia to industry to nonprofits and advocacy to philanthropy. Even with the major advances, 600,000 people in the United States die from cancer annually, and it remains a major cause of morbidity for patients with the disease. The opportunity exists for additional and significant progress in cancer research.

**Annual Plan and Budget Proposal for Fiscal Year 2023.** Dr. Sharpless noted that the NCA of 1971 established the NCI Bypass Budget (i.e., Professional Judgment Budget) process, which is a budget sent directly to Congress. He announced that the NCI released its *Annual Plan and Budget Proposal for Fiscal Year 2023* in August 2021. This year, the NCI Professional Judgment Budget for fiscal year (FY) 2023 proposes $7.8 billion (B) to increase R01 paylines to the 13th percentile, allowing a greater number of meritorious applications to be funded, including those for new and junior scientists. The NCI previously proposed a "5 in '25" plan to increase funding for the Research Project Grant (RPG) pool to reach a 15th percentile payline for R01 grants for established investigators by FY 2025. Robust and sustained investments are needed to achieve that level of commitment to investigator-initiated research, requiring strong support from Congress. Steady rates of increase to the NCI budget have enabled raising paylines from the 8th percentile to the 11th percentile in FY 2020. Dr. Sharpless remarked that achieving the payline goals will be a gradual process, given the high out-year costs of the RPG pool and the significant growth in R01 applications the NCI has experienced in recent years. This proposed budget would allow the NCI to keep pace with the continued need to invest in programs and priorities outside of the RPG pool.

Dr. Sharpless highlighted statistics on the national cost of cancer care in the United States. He reflected on the comments of philanthropist Mary Woodard Lasker, who played a critical role in passing the NCA. The costs of cancer care in 2020 ($208.9 B) and the modeled economic impact of lost productivity due to cancer mortality ($147.6 B) are estimated to be in the hundreds of billions of dollars. Even so, the model reported by Bradley *et al.* in 2008 does not illustrate the financial toxicity associated with cancer placed on the individual patient. This numerical analysis also does not account for the tragedy and devastation of cancer that is familiar to all. Because the cost of cancer in societal and financial terms is immense, the NCI is proposing an FY 2023 appropriation of $7.8 B. Dr. Sharpless remarked on how the federal investment in cancer research is an effective use of these funds that is leading to better understanding of cancer and noted that the *Annual Plan* highlights scientific priorities and emerging opportunities. Copies of the full report and the executive summary can be accessed from the NCI website.

**NCI Budget and Appropriations.** Dr. Sharpless reminded the FNLAC members that the NCI FY 2021 regular appropriations include $195 M for the Cancer Moonshot℠ and $50 M for the Childhood Cancer Data Initiative (CCDI). In FY 2021, the NCI was allotted $306 M for serology research, separate from the regular appropriations. The FY 2022 President's budget released in May 2021 proposed $6.73 B for the NCI, a 2.73 percent increase over the FY 2021 enacted budget. The House Appropriations Subcommittee on Labor, Health and Human Services, Education, and Related Agencies (Labor-HHS) passed its bill out of committee in June 2021 and included $6.99 B for the NCI ($430 M increase). The Senate Appropriations Subcommittee on Labor-HHS has not released its bill. The federal government is operating under a continuing resolution that funds the government through 3 December 2021.

**FNLCR Awareness Campaign.** Dr. Sharpless provided an update on the NCI's plans for enhancing awareness of the FNLCR. The goals are to increase usage of the FNLCR resources and to educate a variety of audiences, including extramural researchers, members of Congress, cancer research advocates, and current and potential partners. An internal working group composed of representatives from NCI-Frederick, Office of Communications and Public Liaison, and the FNLCR communications staff completed initial audience research. Interviews with 9 FNLCR stakeholders, including extramural research program directors and FNLCR staff knowledgeable of programs and resources, and 17 FNLCR and NCI leaders have been conducted. Results are currently being analyzed, and a survey of extramural researchers will be upcoming.

**NCI Programs and Initiatives.** Dr. Sharpless updated the FNLAC members on activities related to the NCI programs and initiatives. The Cancer Moonshot℠ has provided unprecedented opportunities to the cancer research community. Congress has expressed interest in and is envisioning a Cancer Moonshot℠ 2.0. On 9 September 2021, 5 years after the NCAB Blue Ribbon Panel (BRP) delivered its report to the NCAB and the NCI, Dr. Sharpless and the NCI hosted a virtual BRP Report Anniversary Seminar. He was joined by the BRP co-chairs, Drs. Tyler Jacks, Elizabeth M. Jaffee, and Dinah S. Singer, as well as BRP member Dr. María Elena Martínez, to discuss Cancer Moonshot℠ progress. The NCI is planning projects for beyond the end of the 7-year funding period in FY 2023 and is exploring ways to transition those efforts into existing programs.

The CCDI, in the second year of its 10-year funding plan, is progressing at a steady pace. This Initiative seeks to create an infrastructure and a community to aggregate and use data from every child with cancer in the United States. Aligning with the September observance of Childhood Cancer Awareness Month, the NCI planned several activities. On 13 September 2021, the CCDI leadership hosted an overview and progress webinar. Dr. Lynn Penberthy, Associate Director, Surveillance Research Program, Division of Cancer Control and Population Sciences (DCCPS), provided an update on one CCDI component, the National Childhood Cancer Registry (NCCR), an effort to link registry data with other data sources to learn about outcomes and treatment for children with cancer throughout the nation. At the 1 September 2021 NCAB meeting, Dr. Jack F. Shern, Lasker Clinical Research Scholar, Pediatric Oncology Branch (POB), presented on integrating genomics into the pediatric oncology clinic. The NCI blog, *Cancer Currents: An NCI Cancer Research Blog,* recently featured a post by the CCDI leadership about seeing a promising future in the progress against childhood cancer.

On 17 September 2021, the NCI formalized a new collaboration with the Health Resources and Services Administration and U.S. Food and Drug Administration (FDA) on the Cancer Diagnostic Devices (CD2) Interagency Task Force, focusing on accelerating diagnostic devices for near-patient use. The CD2 Interagency Task Force will organize scientific and programmatic coordination, discuss areas of regulatory challenges of bringing these devices to market and ways to overcome them, and emphasize challenges specific to rural and medically underserved communities.

**NCI Response to COVID-19.** Dr. Sharpless reported that in terms of serology support, the NCI and National Institute of Allergy and Infectious Diseases (NIAID) launched the Serological Sciences Network for COVID-19 (SeroNet), a nationwide collaboration of 25 institutions to increase serological testing and understand the immune response to SARS-CoV-2 infection and vaccination. The FNLCR has continued to play a major role in the NCI response to the COVID-19 pandemic. SeroNet has been operational since October 2020 and has been productive, with data reported in 90 publications, some in high-impact journals.

The NCI observed a dramatic decrease in cancer screening as the COVID-19 pandemic evolved. In fact, COVID-19 caused a disruption in care on all levels (e.g., screening, diagnosis, treatment, research) and has exacerbated health disparities. EPIC Health Research Network data showed that colon cancer screening rates decreased dramatically in early 2020, persisted for several months, then partially recovered. The NCI Population-based Research to Optimize the Screening Process (commonly called PROSPR) Consortium showed similar trends. It is estimated that more than 10 million screening events were missed in the United States in 2020 and early 2021. With diagnoses occurring in later stages, evidence of up-staging in national data is likely. Dr. Sharpless remarked on a collective and national call to minimize any long-term effect of the COVID-19 pandemic on patients with cancer, which is not solely an NCI problem. He highlighted the efforts of other cancer organizations, such as the AACR and American Society of Clinical Oncology (ASCO), to provide new guidelines for cancer care during this time of national crisis.

In addition, cancer care and cancer research were severely disrupted by the COVID-19 pandemic. Dr. Sharpless commended the ingenuity and fortitude of American cancer researchers required to continue to produce stellar science during the COVID-19 era. Although basic science proceeded well, the accrual for screening trials across the NCI National Clinical Trials Network (NCTN) significantly decreased. Overall, weekly accruals recovered in the NCTN by the end of FY 2020, but not evenly across trials. Industry and other NCI networks have not seen a robust return in enrollments, leading to longer trials and delaying results for patients. The NCI remains concerned about the effects of COVID-19 on cancer research.

**Cancer Research Updates.** Dr. Sharpless updated the FNLAC members on three research initiatives. The Dual Anti-CTLA-4 and Anti-PD-1 blockade in Rare Tumors (DART) trial successfully used a basket clinical trial approach to a Phase II trial with a common infrastructure that is useful in several settings. Results of this study, led by the Southwest Oncology Group (SWOG) Cancer Research Network and sponsored by the Division of Cancer Treatment and Diagnosis (DCTD), were reported in the 1 July 2021 issue of the *Journal for Immunotherapy of Cancer* and indicated that using ipilimumab and nivolumab to treat angiosarcoma is promising. Tumors in four patients partially or completely responded to treatment, and two other patients had long-term, but stable, disease for more than 6 months. These data align with the results of the "Count Me In" initiative's angiosarcoma project, led by the Broad Institute of Massachusetts Institute of Technology and Harvard University, and are a significant advancement for patients with this rare cancer. DART also will be investigating other rare subtypes of cancers of the ovary, small intestine, lung, sinuses, pancreas, and breast. This use of a basket trial model is similar to the NCI-Molecular Analysis for Therapy Choice (NCI-MATCH) rare tumors studies, which have shown compelling efficacy in certain rare-disease populations. Although small-scale unrandomized Phase II trials can present regulatory challenges, cancer patients have benefitted from the FDA Oncology Center of Excellence's vision to approve drugs based on this trial design using expedited reviews and an accelerated process.

The NCI POB investigators, in collaboration with researchers from Washington University in St. Louis, reported findings in the 31 August 2021 issue of *PLOS Medicine* of using liquid biopsy in neurofibromatosis type 1 (NF1) to distinguish between benign plexiform neurofibroma (PN) tumors and malignant peripheral nerve sheath tumors (MPNSTs). NF1 tumors almost always are diagnosed in childhood, and approximately 15 percent transform into lethal MPNSTs. MPNST is a challenging clinical syndrome requiring biopsies and serial imaging and remains a clinical issue. In this study, the researchers developed a blood test that could reduce the need for full-body scan imaging or biopsies for diagnosing these patients. This test could offer a sensitive and inexpensive approach to cancer monitoring in patients with NF1 tumors. Dr. Sharpless highlighted that POB investigator Dr. Brigitte Wideman previously had reported her research using selumetinib to treat patients with NF1 tumors.

Recent findings from the Sherlock-Lung study were reported by DCEG researchers in the 6 September 2021 issue of *Nature Genetics*. The study aims to perform genomic and evolutionary classification of lung cancers in never smokers (LCINS) to infer processes leading to LCINS tumor formation. Early results from the first 232 general population cases were mostly of patients of European origin. Plans are to expand the study to analyzing LCINS in Asia and to people experiencing specific environmental and industrial exposures, such an indoor air pollution and coal-fired plants. The study has identified several LCINS subtypes, but no molecular signature, as exists for smokers, has emerged.

**Cancer Health Disparities Research and Workforce Diversity.** Dr. Sharpless highlighted that the NCI has worked over the last decade to increase clinical trials accruement among minority populations and has been successful. The accrual of Hispanic and Black/African American patients in NCI clinical trials has significantly increased over the past 20 years, primarily because of the efforts of the NCI Community Oncology Research Program (NCORP) and its Minority/Underserved Community

Sites, but could be further improved. The NCI issued requests for applications (RFAs) for the new Connecting Underrepresented Populations to Clinical Trials (CUSP2CT) program. The purpose of CUSP2CT is to implement and evaluate multilevel and culturally tailored outreach and education interventions, with the primary goal to increase referral of racial/ethnic minority populations to NCI-supported clinical trials. Applications for the Data, Evaluation, and Coordinating Center RFA and the Resource-Related Research Projects RFA will be due 19 November 2021.

The NCI and the National Institute on Minority Health Disparities (NIMHD) are assisting the NIH in administering the Faculty Institutional Recruitment for Sustainable Transformation (FIRST) Initiative, which is a Common Fund initiative. FIRST will establish a national network of large cooperative agreements (U54s) to academic institutions committed to the development of minority faculty. The initial awards have been issued to six institutions, and the FIRST Coordinating Evaluation Center (managed by the NIMHD) has been established. FIRST seeks to create cultures of inclusive excellence at NIH-funded institutions by implementing a set of well-integrated evidence-based strategies and evaluating their impact based on specified metrics on culture inclusion and diversity.

**Update on Global Cancer Research.** Dr. Sharpless announced that the Center for Global Health (CGH) is celebrating its 10th anniversary. The NCI established the CGH in 2011, which led to the development and promotion of resources that advance global cancer research and collaboration, particularly in low- and middle-income countries (LMICs). Dr. Sharpless and CGH Director Dr. Satish Gopal published a commentary, "Cancer as a Global Health Priority", in the 6 August 2021 issue of *The Journal of the American Medical Association,* calling attention to the burden of cancer in LMICs.

In June 2021, President Joseph Biden and Prime Minister Boris Johnson agreed to convene the first U.S.–U.K. Bilateral Cancer Summit. The NCI staff has been working with colleagues at Cancer Research United Kingdom and other stakeholders in both governments to organize this summit. A planning meeting is scheduled for November 2021, and the summit is projected to be held in the spring of 2022.

**Leadership Appointments.** Dr. Sharpless announced that Dr. Robert T. Croyle, Director, DCCPS, who has had a celebrated career in federal service, will be retiring in early 2022. Dr. Katrina Goddard has been selected as the new DCCPS Director.

**In the discussion, the following points were made:**

- Dr. Sharpless commented on the retirement of Dr. Francis S. Collins, NIH Director. He remarked that Dr. Collins has been tremendously effective in informing Congress of NIH activities and has a strong relationship with Congress (with broad bipartisan support). Dr. Sharpless continued that the NIH Director has to be someone in whom Congress believes and who Congress is confident will be a good steward of federal dollars. The White House will nominate a candidate, who will then be scheduled for Senate confirmation. An Acting NIH Director could be appointed.

- LCINS, like other cancers, is likely heterogenous and consists of diverse mutational signatures because of other environmental agents (e.g., indoor air pollution, diesel exhaust). Cohorts of known exposure on an international level, such as the Sherlock-Lung study, are providing new insights. Although the causes of cancer without a definitive molecular signature are rare, many patients' cancer will have no identifiable signature. The field relies on epidemiologic studies and research on carcinogens, but those are not the best approaches. Determining causality and focusing attention on countries with low and high lung cancer incidences and accompanying tumor burden will be critical.

## III. ACTING ASSOCIATE DIRECTOR: FREDERICK UPDATE—DR. KRISTIN L. KOMSCHLIES

Dr. Kristin L. Komschlies, Acting Associate Director, NCI at Frederick, and former Deputy Director, Office of Scientific Operations (OSO), provided an update on the NCI at Frederick and OSO activities. Dr. Komschlies, who began at FNLCR as a postdoctoral fellow, has had a career in research as a principal investigator, later moving into administrative positions. As the primary contracting officer representative for the NIH Federally Funded Research and Development Center (FFRDC) contract for more than 5 years, Dr. Komschlies has a clear understanding of the capabilities of an FFRDC and the tools and resources it provides to the NCI and the NIH communities. The FNLCR is one of 42 FFRDCs sponsored by a government agency to support all aspects of basic and applied research as well as research and development (R&D).

FNLAC members were reminded of the three main visions the NCI has for the FNLCR. The first is to function as a nucleus for large-scale projects not easily performed elsewhere or through other mechanisms. The second is to serve as a hub for technology development. The third is to serve as a resource to support the intramural and extramural components of the NCI and the NIH, particularly NIAID. The FNLCR, a unique and collaborative R&D resource, is equipped as an FFRDC to tackle difficult, urgent, and intractable problems. An FFRDC is well-suited for this role because of the federal acquisition regulations, special authorities, and opportunities for subcontracting, all allowing the FNLCR to rapidly pivot resources to enable a national response. One recent example of such a response was to the COVID-19 pandemic by rapidly (within 24 to 48 hours) converting parts of the FNLCR human papillomavirus (HPV) Serology Laboratory to SARS-CoV-2 serology. Another example is establishing and operating a chimeric antigen receptor (CAR) T-cell facility within the FNLCR Biopharmaceutical Development Program that now is supporting multicenter CAR T-cell clinical trials.

The main focus of the FNLCR is to maximize the capabilities of an FFRDC in terms of its unique acquisition and response capabilities, flexibility, rapid response, and increased efficiency. Dr. Komschlies emphasized that the FNLCR requirements for providing support are to offer what cannot be achieved as effectively by other NCI components or through other government mechanisms (e.g., cooperative agreements) and to not compete with the commercial sector. The FFRDC/FNLCR is a dynamic environment that supports difficult research challenges with complex and multifaceted components, streamlines operations beyond just the procedural aspects, stretches the boundaries as an FFRDC, and thinks creatively about contracting and conducting science. The NCI at Frederick looks forward to input from the FNLAC on ways of enhancing the effectiveness of the FNLCR.

Dr. Komschlies commented that the current re-competition of the FNLCR operations and technical support contract provides another opportunity to fine-tune this FFRDC. She described examples of projects currently meeting the NCI's vision for this Center. The FNLCR is coordinating national mission programs. The RAS Initiative, a hub-and-spoke model of research and collaboration, is investigating RAS biology using four novel approaches with various collaborators. The FNLCR has a key role in the NCI COVID-19 response and is supporting NIAID COVID-19 clinical trials, the SeroNet Coordinating Center, and the NCI-FDA SARS-CoV-2 Serology Validation Program. In terms of technology development, the National Cryo-Electron Microscopy (Cryo-EM) Facility (NCEF) launched in 2017 and is exclusively available to the extramural community at no cost to the user. The Nanotechnology Characterization Laboratory (NCL) established in 2004 has enabled nanoparticle development, physical characterizations, and *in vitro* and *in vivo* studies. Regarding support to intramural and extramural communities, the FNLCR has a role in other NCI programs and initiatives, such as imaging informatics, cancer imaging archives, Cancer Research Data Commons, Pediatric MATCH, Patient-Derived Models Repository (PDMR), NCI Experimental Therapeutics (NExT) program, Chemical Biology Consortium, National Clinical Laboratory Network, and Data Coordination Center for

the Genomic Data Commons. In addition, the FNLCR provides support for clinical trials, including Therapeutically Applicable Research to Generate Effective Treatments (TARGET). Dr. Komschlies noted that further details on the NCEF and NCL will be provided later in the meeting.

## IV. UPDATE: NATIONAL CRYO-EM FACILITY—DR. DWIGHT NISSLEY

Dr. Dwight Nissley, Director, Cancer Research and Technology Program (CRTP), FNLCR, provided an update on the NCEF, a major activity of the Advanced Technology Research Facility on the FNLCR campus. He noted the organizational structure of the FNLCR in the context of the NCEF: Dr. Ethan Dmitrovsky is Laboratory Director and President, Leidos Biomedical Research, Inc. (Leidos Biomed); Dr. Leonard P. Freedman is Chief Science Officer; and Dr. Nissley is CRTP Directorate Head, one of six FNLCR directorate heads reporting to Dr. Freedman. The CRTP provides technology support to the NCI and the NIH, has outward facing laboratories that are extramural enabling (e.g., NCL and Antibody Characterization Laboratory), and supports efforts related to national missions, of which the NCEF is one. The NCEF launched in 2017 with a data collection component in response to the need of extramural cancer research investigators to collect cryo-EM data for their projects. Dr. Thomas Edwards, Senior Microscopist, serves as the technical lead for the NCEF and is supported by two other microscopists, a scientific project manager, and an information technology team. The second component, Advanced Cryo-EM Technology (ACT), was established in 2019 to explore new platforms, methods, and technology development for the cryo-EM field. Dr. Jana Ognjenovic, Senior Scientist, is ACT group lead.

Dr. Sriram Subramaniam, Former National Cryo-EM Consultant and Founding Director (currently at the University of British Columbia), previously identified three user communities that would benefit from the facility, which Dr. Nissley described. Group 1 represents researchers with experience in cryo-EM technology, who are the key drivers of growth of cryo-EM in the United States. Members of this group have some access to local screening microscopes, but inadequate access to high-end instrumentation. To accommodate this group, the NCEF provides, at no cost to the user, access to two Titan™ Krios 300 kiloelectron volt (keV) microscopes that are state-of-the-art. Users with cancer-related projects can gain access to imaging slots through the NCEF access portal and are provided reports containing all images, imaging parameters, and statistics to evaluate their samples. In addition, the ACT group has two 200 keV microscopes, the JEOL Inc. CRYO ARM™ 200 and the Thermo Fisher Glacios, to evaluate cutting-edge and lower-cost technologies for high-resolution structure determination. The NCEF has supported more than 700 data collections over the past 4 years for 130 extramural investigators from 50 different U.S. academic institutions, resulting in 51 publications, 19 of which were published in 2021.

Group 2 represents structural biologists in adjacent disciplines (e.g., X-ray or nuclear magnetic resonance spectroscopy). Members of this group understand the value in using cryo-EM and possess expertise in protein biochemistry but need training in cryo-EM specimen preparation, data collection, and processing. Recognizing that grid preparation and screening is a major bottleneck for Group 2 investigators, the NCEF leadership proposes a grid preparation and screening service to provide access to the latest generation of grid-freezing technology machines (e.g., Chameleon®, VitroJet™). Samples will be sent to the NCEF and frozen by the facility staff, and grids will be screened on an NCEF microscope.

Group 3 represents biologists with interest in important biomedical problems who are interested in adding cryo-EM methods to their toolkit but need training and collaboration in all aspects of the workflow (i.e., from protein purification to the final interpretation of the structures). To accommodate this group on a 3- to 5-year time frame, the NCEF leadership plans a 1-week training course to commence in the summer of 2022. Lecturers external to the NCI and FNLCR will be invited to gain expert insights into state-of-the-art workflows and processes. Attendees will have the opportunity to bring their own samples; work with different alternative grid preparation platforms; learn how to collect, store, and transfer data for processing; and build models to generate three-dimensional (3D) structures for their proteins of interest.

The three areas of focus of ACT are new hardware and software platforms, grid preparation, and challenging biochemistry (e.g., cancer targets), with the goal of mapping specific biological projects (e.g., RAS Initiative, NF1 and NCI-defined high value complexes) to the current and future National Cryo-EM Program (NCEP) portfolio. The ACT group has installed and validated new microscope/camera combinations, collected cryo-EM data and determined the structure of beta-galactosidase at 1.8 angstrom (Å) resolution using the CRYO ARM 200 microscope and at 2.0 Å on a Thermo Fisher Glacios. These data sets have been deposited in the Electron Microscopy Public Image Archive (commonly called EMPIAR). The ACT group also has shown the structure for beta-galactosidase at a higher resolution of 1.61 Å resolution using the 200 keV microscope, with new structural features detected.

Regarding challenging cancer targets and leveraging existing biological projects, the ACT team is collaborating with the RAS Initiative team to determine a high-resolution structure of the NF1 protein to better understand the mutations related to NF1 and other diseases. After several rounds of optimizations, cryo-EM data collection and processing revealed two-dimensional classes and dimers. The first 3D map, at approximately 4 Å, allowed rigid body positioning of the NF1 protein functional domains, GAP-related domain and Sec 14 pleckstrin homology, and showed them within the dimer. Other examples of advancing cryo-EM through technology development include evaluations of the high-resolution structure of SARS-CoV-2 nonstructural protein 1 (Nsp1) bound to the 40 S subunit of human ribosome in a complex with the only FDA-approved drug that targets the human ribosome, omacetaxine mepesuccinate.

Dr. Nissley highlighted an outreach effort of the NCEP to establish a mechanism for NIH visiting researchers to advance their cancer-related projects to enable future cryo-EM work at their own institutions. This mechanism could provide opportunities for postdoctoral fellows who have limited structural biology expertise (e.g., Group 3) to work at both the FNLCR and their home institutions to advance their cancer projects. In closing, Dr. Nissley acknowledged the NCEF and ACT teams and noted that a search is in progress to fill the National Cryo-EM Consultant position. The FNLCR and the NCI are seeking a recognized thought leader in the cryo-EM field to provide advice on emerging technologies and, simultaneously, represent the needs of the extramural cancer research community.

**In the discussion, the following points were made:**

- Providing an infrastructure to assist cancer research investigators in generating multiprotein complexes is a viable path for the NCEP. The FNLCR Protein Expression Laboratory has state-of-the-art cloning and vector construction capabilities that can expand rapidly to address such a need.

- Structure-based drug design is another approach the NCI and the FNLCR could consider when expanding the NCEP portfolio.

## V. THE HUMAN P97 COMPLEX—DR. MINGLEI ZHAO

Dr. Minglei Zhao, Assistant Professor, Department of Biochemistry and Molecular Biology, University of Chicago, presented his data on studies of human p97 complex, a cancer target, collected using the NCEF. The p97 complex (also known as valosin-containing protein [VCP] or cell division control protein 48 [CDC48]) in *Saccharomyces cerevisiae*) is an abundant ATPase. This complex, which was first discovered in yeast genetic screens, is a member of the type-II ATPases Associated with various cellular Activity (AAA) family and is one of the most abundant ATPases in the cytoplasm. The mutations to the p97/VCP/CDC48 complex are associated with several neurodegenerative diseases, thus making it a viable cancer and antiviral drug target. Consisting of three domains (N, D1, D2) and 806 amino acids in its composition, the p97/VCP crystal structure was solved at 4.0 Å resolution in 2003 (Huyton *et al.*, 2003; DeLaBarre *et al.*, 2003) and several mutations have since been mapped.

Dr. Zhao has an interest is studying the p97/VCP complex because of its function and high abundance as a central hub of the cellular ubiquitin (Ub) system. The most common consequence of ubiquitination is degradation through the proteasome. Some of the other Ub-related processes include endoplasmic reticulum–associated, chromatin-associated, and mitochondria-associated degradation, ribosome quality control, and unfolded protein response. The p97/VCP complex also participates in various cellular processes (e.g., DNA repair, autophagy, cell cycle progression) through different adaptor proteins. Dr. Zhao hypothesized that inhibiting the p97/VCP complex (a known drug target) will further exacerbate the toxic effects to cancer cells and focused his research on the heterodimeric cofactor Ufd1/Npl4. The Zhao laboratory is addressing three key questions: How does p97 process ubiquitinated substrates through various cofactors? What are the physiological roles of disease mutations? What are the mechanisms of the various inhibitors?

Dr. Zhao described data (primarily the work of postdoctoral fellow Dr. Man Pan) targeting the p97 complex, the adaptor protein, and the polyubiquitinated substrate. The Zhao laboratory optimized a cell-based assay first developed by Blythe et al. (2017) and Bodnar et al. (2017) to measure the amount of *in vitro* polyubiquitinated substrate, which reflected the activity of p97 complex over time. Using a single-particle cryo-EM workflow, the results showed three structural classes of compounds at two conformations: closed state and open state. Superimposing the two structures showed a staircase conformation and the power stroke motion of the D1 and D2 rings, revealing the translocation mechanism of this ATPase. of this ATPase. Further structural evaluations illustrated that the inter-subunit sensing motif is critical for this staircase conformation.

Using NMS-873, a well-known selective allosteric non–ATP competitive inhibitor of p97 with cancer and antiviral activity, the Zhao laboratory was able to fully inhibit the activity of the p97 complex *in vitro*. The Zhao laboratory next solved the high-resolution cryo-EM structure of human p97 in a complex with NMS-873 at 2.4 Å. The well-resolved density map showed that NMS-873 binds a cryptic hydrophobic pocket in the D2 domain, thus defining the mechanism of action, inhibition of translocation via interaction with lysine 615. The cofactor protein was not well resolved in the p97 complex structure, but a slowing of the ATP hydrolysis provided some confirmation of its position and the engaged Ub moiety. In a working model, these data suggest that within the translocation mechanism of human p97, a substrate-loading stage exists in which the adaptor, Ufd1/Npl4, brings the polyubiquitinated tail inwards, inducing a conformational change in a seesaw motion. Previous studies have shown that the drug disulfiram, used to treat alcohol abuse, targets cancer cells through the p97 Npl4 adaptor. The Zhao laboratory found that a disulfiram derivative, diethyldithiocarbamate-copper, diffuses across the plasma membrane and hypothesizes that it likely targets the zinc finger motifs of Npl4. Dr. Zhao expressed appreciation to the NCEF staff for their support of this cryo-EM project.

**In the discussion, the following points were made:**

- Understanding how allosteric modulation of small molecules occurs using cryo-EM is one direction the field can consider in the future.

## VI. MOUSE MODELS FOR CANCER RESEARCH: AN NCI RESOURCE—DR. JOANNA M. WATSON

Dr. Joanna M. Watson, Chief, Tumor Metastasis Branch, Division of Cancer Biology (DCB), updated the FNLAC members on the NCI Mouse Repository for basic cancer research and translational oncology. Dr. Watson explained that the repository originated as a component of the Mouse Models of Human Cancers Consortium (MMHCC), which was active from 1998 to 2013 and involved researchers from across the United States and worldwide. Established to generate, validate, and disseminate mouse models of human cancers, the MMHCC served as a repository for related tools, databases, protocols, and

other resources. Housed at FNLCR and maintained after the MMHCC program concluded, the NCI Mouse Repository currently is overseen by the Laboratory Animal Science Program (LASP). It contains more than 160 strains unique to the repository that are available to researchers worldwide for the cost of shipping. Initially, mouse breeding pairs were available for order, but as of 2016, only cryopreserved germplasm is provided. Information on each strain (e.g., mutation type, organ site, genetic background, related publications) is available on the repository website. Most shipments are domestic, but one-third of orders are from internationally based researchers. The repository also includes a library of mouse embryonic stem cells (mESCs) expressing every known murine microRNA (miRNA). The mESCs conditionally express miRNAs under control of a reversible tetracycline-inducible system, and each cell line includes a fluorescent reporter gene to confirm expression. The cell lines can be purchased for a nominal processing fee, but purchase is restricted to investigators in the United States.

Dr. Watson highlighted recent publications and seminal findings involving the use of repository model strains. Dr. Tong Wu and his laboratory at Tulane University isolated tumor-initiating liver cells from mutagenized mice harboring a conditional mutation in the transforming growth factor β receptor (TGFβR) gene (strain 01XN5) to study the role of TGFβ signaling in hepatocellular carcinoma (HCC). Following splenic injection into a second set of mice, a subset of the transplanted cells was subjected to conditional inactivation of TGFβR. The data showed that cells with inactivated TGFβR readily formed HCC, whereas cells with the intact gene were less tumorigenic. These studies, which demonstrated a tumor suppressor role for TGFβ signaling, were published in the 16 July 2018 issue of Hepatology.

A study published in the 26 December 2018 issue of Cell Reports by Dr. Julian Downward at the Francis Crick Institute and colleagues investigated the role of phosphatidylinositol 3-kinase (PI3K) signaling in lung cancer in mouse strains containing a tetracycline-inducible epithelial growth factor receptor (EGFR) point mutation, L858R (strain 01XEA). The data showed that disruption of the PI3K RAS interaction led to reduced onset and increased regression of EGFR mutant–driven lung cancer. This mutation also is found in human lung cancers and is responsive to tyrosine kinase inhibitors, such as erlotinib and gefitinib.

Dr. Watson noted that colorectal cancer cells typically harbor mutations in four genes: adenomatous polyposis coli (APC), kirsten rat sarcoma virus (K-RAS), TGFβ, and tumor suppressor protein p53 (p53). Although cells with all four mutations, AKTP cells, are capable of metastasis, cells with only single or double mutations remain benign. Dr. Masanobu Oshima and colleagues at the Nano Life Science Institute, Kanazawa University, published in the 8 February 2021 issue of Nature Communications results of a study using mixed tumor cells carrying only APC and p53 mutations (AP cells) with AKTP cells. The study showed that the presence of malignant AKTP cells was sufficient to cause the AP cells to metastasize to the liver. The data revealed continued proliferation of AP cells upon depletion of AKTP cells, indicating an early role for malignant cells in driving metastasis. These studies used multiple mouse strains 01XN5, 01XM3, and 01XAB.

In a final study described by Dr. Watson, Dr. Daren R. Carpizo at the University of Rochester Medical Center and his colleagues used a variety of mouse strains from the repository to better understand the mechanisms of breast cancer vulnerabilities. Whereas 10 percent of triple-negative breast cancers harbor a germline breast cancer type 1 susceptibility protein (BRCA1) mutation, all BRCA1 mutant cells contain p53 mutations. This study used multiple mouse strains (01XA8, 01XM2, 01XAB) and investigated that role that p53 inactivation plays in triple-negative breast cancer and made use of zinc metallochaperones to restore wild-type activity in zinc-deficient mutant p53, resulting in inhibition of tumor growth in vivo. This study, demonstrating the therapeutic potential of metallochaperones, was published in the 15 April 2019 issue of NPJ Breast Cancer. Dr. Watson emphasized that none of the authors of these publications were affiliated with the MMHCC and noted that the number of publications citing cancer mouse models from the repository has increased steadily since 2000.

Dr. Watson described general outreach efforts performed by the NCI repository, including notices on the NCI DCB website and social media accounts as well as announcements at new grantee workshops and other meetings and seminars. The NCI repository also is advertised at targeted outreach events, including meetings of the Oncology Models Forum and International Society for Transgenic Technology meetings hosted by LASP.

**In the discussion, the following points were made:**

- The NCI Mouse Repository managers could consider joining the effort to standardize microbial flora in cancer models.

- The FNLCR could increase marketing efforts to both junior and senior investigators. For example, an email could be sent to NCI grantees on the research listserv to update investigators on the resources in the NCI Mouse Repository. This email could include a link to the repository website.

- Because model systems are evolving rapidly, it is important to acquire new strains. The NCI should encourage investigators to deposit their novel mouse strains into the repository.

## VII. NANOTECHNOLOGY CHARACTERIZATION LABORATORY: SUPPORTING TRANSLATION OF CANCER NANOMEDICINES—DR. PIOTR GRODZINSKI

Dr. Piotr Grodzinski, Chief, Nanodelivery Systems and Devices Branch, Cancer Imaging Program, DCTD, provided an update on the NCL, a laboratory that has functioned as a partner program of the NCI Alliance for Nanotechnology in Cancer since its establishment in 2004. Other NCI Alliance Programs include such funding mechanisms as the Centers of Cancer Nanotechnology Excellence (CCNEs), Innovative Research in Cancer Nanotechnology (IRCN), Cancer Nanotechnology Training Centers, and Toward Translation of Nanotechnology Cancer Innovations (TTNCIs). Dr. Grodzinski explained that the CCNE Program was transitioned in 2020 to a couple of focused solicitations based on R01 grant mechanism. The Alliance has retained its productivity, with high-profile publications and about 130 startup companies pursuing more than 20 clinical trials.

The NCL was launched as an interagency collaboration between the NCI, FDA, and the National Institute of Standards and Technology (NIST). Funding for the program is provided entirely by the NCI. The main goal of the NCL is to facilitate the translation of nanotechnology medicine by providing a characterization of nanoparticle constructs. The NCL serves researchers in academia, industry, and government free of charge. A standardized assay cascade developed by the NCL includes physicochemical (e.g., size/size distribution, composition, purity, surface chemistry, stability), *in vitro* (e.g., sterility, hematology, immune cell function, toxicity), and *in vivo* (e.g., pharmacology, immunotoxicity, single- and repeated-dose toxicity) evaluations. Researchers in the field are encouraged to download and implement evaluation protocols, which are made available online.

Dr. Grodzinski described the two-phase application process. In the first phase, investigators submit a brief white paper summarizing the background information, novelty of the concept, compound synthesis and scale-up, and clinical impact. Applications are reviewed and accepted quarterly, with decisions announced within 45 days of the application deadline. After phase 1 approval, investigators submit an expanded written or oral proposal and respond to reviewer questions. This input is due within 3 months of receiving the invitation letter, and a final decision is remitted within 2 weeks. The overall acceptance rate is around 40 percent annually, with about 20 applications per cycle submitted mainly by industry and academic researchers. The NCL has characterized more than 450 nanomaterials in hundreds of collaborations with partners in industry, government, and academic institutions. A total of 72

characterization protocols have been developed. Although a variety of materials (e.g., metalloids, dendrimers, micelles, fullerenes, nucleic acid- and peptide-based compounds) have been studied, most submissions involve liposome- or polymer-based materials. The NCL began accepting COVID-19-related applications in December 2020 and has since accepted four projects. Some NCL-characterized nanomedicines have moved into and beyond clinical trials, with such compounds as Vyxeos® (CPX-351) and Hensify® (NBTXR3) recently gaining approval in the United States and the European Union.

Dr. Grodzinski summarized NCL efforts in education and knowledge dissemination. In 2020, the Assay Cascade protocols were downloaded from the NCL website 2,243 times. The majority of these protocols, each of which has its own digital object identifier (DOI), were physicochemical and immunotoxicity assays. Workshops for researchers held in December 2019 and May 2020 attracted nearly 200 participants, 60 percent of whom were principal investigators and 20 percent of whom were international investigators. A future bootcamp to provide hands-on nanotechnology experience to graduate students and postdoctoral researchers has been approved. The NCL organizes a series of technology development seminars, in addition to presenting at national and international research conferences. Approximately eight client reports and several manuscripts (e.g., methods development efforts, review articles) are published annually.

The NCL is involved in standards development; NCL collaborates with ASTM International and International Organization for Standardization (ISO) to standardize Assay Cascade protocols for nanoparticle characterization; three NCL protocols are recognized as the ASTM International standards, and many are in development. The NCL has supported the production of colloidal gold reference standards for nanoscale particles generated by NIST. The program also participates in interlaboratory studies comparing identical methods implemented across varying laboratories. To meet the demand of the NCL clientele and the increased submission of therapeutic nucleic acid nanoparticles, the NCL optimized an analytical technique to measure both total and free RNA concentrations utilizing a commercially available fluorescence assay kit. Recent NCL efforts have included studies of nanomedicine pharmacokinetics (i.e., measuring encapsulated and unencapsulated nanomedicines in circulation), as well as reformulation of promising drug candidates. Dr. Grodzinski provided an example of a tumor-targeted brefeldin prodrug, which is being reformulated to improve antitumor activity and reduce neurotoxicity. He also described the cancer Nanotechnology Laboratory (caNanoLab) Data Portal, a resource for curated data sharing that provides support for nanomaterial annotation and characterization. These efforts have been well received in the nanomaterials research community. In a recent survey conducted to probe public perception of the program, 75 respondents rated almost all aspects of the NCL as relevant or highly relevant to their research.

**In the discussion, the following points were made:**

- Few contract research organizations specialize in nanomedicine.
- Because methods developed within the NCL are published and accessible to the wider community, other institutions might be capable of conducting particular assays.
- The NCL is unique in housing all relevant assays in a central location.

## VIII. ADJOURNMENT—DR. CANDACE S. JOHNSON

Dr. Johnson thanked the Committee members and other participants for attending. Members were reminded to send potential agenda topics for future FNLAC meetings to Dr. Lopaczynski. There being no further business, the 8th Virtual Meeting of the FNLAC was adjourned at 4:00 p.m. EDT on Monday, 18 October 2021.

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| Date | Candace S. Johnson, Ph.D., Chair |
| Date | Wlodek Lopaczynski, M.D., Ph.D., Executive Secretary |