1. From the Editors

The CICR editorial board has seen some changes since the turn of the New Year. Dr Billy Day has since 2012 served as co-editor and on behalf of the editorial board I would like to thank him for his commitment to help steer the CICR newsletter, contribution to developing it and not least for his great sense of humor! Great news is that Dr Day will still be on the scene and share his experience and insight in the capacity of 'Past Editor'. Also, we are grateful to Drs. Zoe Cournia, Biomedical Research Foundation, Academy of Athens (2012-2016), Jinhui Zhang, Texas Tech University (2012-2016) and Christopher Van, University of Massachusetts Medical School (2014-2016) for their hard work in bringing the chemistry news to you. New members of the editorial board are Drs. Gunnar Boysen, University of Arkansas for Medical Sciences (2016-2018) and Terry W. Moore, University of Illinois at Chicago (2016-2019). Finally, but not least, we welcome Dr George S. Sheppard, AbbVie Inc, as Editor-Elect (2016).

Other news in the CICR community relates to the steering committee. Drs. Vinod Patel (Sanofi) and Melissa M. Vasbinder (Ribon Therapeutics) stood for election of CICR Chairperson-Elect 2016-2017 with the latter winning the election. Here on editorial board we thank both Drs. Patel and Vasbinder for standing and look forward to working closely with Dr Vasbinder in the near future. Dr Vasbinder has many years of experience of leading oncology chemistry groups in industry. Specifically, she has worked across all phases of drug discovery from target identification through candidate selection on targets such as Chk-1, IAP, JAK, and B-Raf.

For more information on the full editorial team, CICR newsletter, steering committee or other CICR-related matters please click on the link below:
http://www.aacr.org/MEMBERSHIP/PAGES/SCIENTIFIC%20WORKING%20GROUPS/CICR-NEWSLETTER___A5B924.ASPX#.VsR24qFFCQc

Cancer molecular diagnostics has experienced a revolution in the post-genomic era. Deeper understanding of the molecular events involved in cancer initiation, progression, response to therapy, and metastasis are continually evolving. These technologies combined with clinical characteristics of cancer, have also helped us realize the molecular heterogeneity within different cancer types. High-throughput genomic, transcriptomic, epigenetic, proteomic, and metabolomic technologies are allowing systematic studies of biological samples to identify novel molecular indicators of disease. These technologies can be applied to a variety of types of biological samples including blood, urine, tissue, and stool. In tissue, molecular diagnostics are routinely used to determine course of cancer treatment, for example, overexpression of estrogen and progesterone receptors and *HER2* in breast cancer to determine use of hormone therapy and Herceptin, respectively. Developing a new diagnostic test shares the same challenges as developing a new drug.

In the area of cancer detection, exciting new molecular diagnostic tests are emerging, for example, the stool-based test for colon cancer, which detects 7 DNA mutation biomarkers, 2 DNA methylation biomarkers, a Hemoglobin biomarker, and Beta actin. New technologies are providing insight into potential future generations of biomarkers for molecular diagnostics. For example, the use of desorption electrospray ionization mass spectrometry to examine levels of lipids to identify surgical margins in brain cancer. Another exciting prospect is how the molecular targets identified by high-throughput technologies can be combined with new developments in engineering resulting in exquisite sensitivity, speed, and ease of access to testing. Overall, the outlook for molecular diagnostics is promising in exciting areas such as improving early cancer detection, distinguishing aggressive from indolent cancer, determining treatment type, prognosticating response to therapy, and identifying cancer recurrence.

In this issue we are profiling Dr. Livia S. Eberlin, a newly appointed Assistant Professor at the Chemistry Department of The University of Texas at Austin. Dr Eberlin's group is interested in using ambient ionization mass spectrometry in creative ways to address critical problems in cancer research, which amongst other things include tools discovery to be implemented in molecular diagnostics.

Finally, we would like to bring to the attention of the CICR community the next AACR Annual Meeting, which will take place in New Orleans (16-20th April). For details of the CICR town hall meeting or CICR-sponsored sessions at the AACR meeting.

Editorial co-author: Dr Sharon Pitteri

2. Selected Research Highlights

Proteomic Maps of Breast Cancer Subtypes To date, most 'omic studies of breast cancer that have attempted to distinguish molecular differences among subtypes have focused on DNA and RNA analyses. In recent years, developments in technologies have enabled deep proteomic profiling of clinical samples to be performed. Tyanova et al. describe such a Super-SILAC study of 40 FFPE breast tumors including estrogen receptor positive (luminal), HER2 positive, and triple negative subtypes. More than 10,000 proteins were quantified across the tumors and functional differences between breast cancer subtypes were observed including energy metabolism, cell growth, mRNA translation, and cell-cell communication. A panel of 19 proteins was found to differ between breast cancer subtypes. Of these, only three were correlated with gene copy number variations and eleven with mRNA levels. These results suggest a predictive signature and discrimination pathways for breast cancer subtypes that may have clinical use.

The Molecular Taxonomy of Primary Prostate Cancer The Cancer Genome Atlas (TCGA) is focused on increasing the understanding of the molecular basis

of cancer through genome analysis. Prostate cancer is heterogeneous in its molecular alterations and variations in clinical course. In this recent manuscript, a TCGA study of 333 primary prostate tumors is described. 74% of tumors fell into one of seven subtypes defined by specific gene fusions or mutations. Notably, epigenetic analysis showed substantial heterogeneity within a phenotype and the androgen receptor transcription levels also varied widely by subtype. A substantial portion of tumors had a potentially actionable molecular defect. This study provides a better understanding of some of this heterogeneity and some potentially actionable targets.

Patient-Level DNA Damage and Repair Pathway Profiles and Prognosis after Prostatectomy for High-Risk Prostate Cancer Current clinical measures to predict prostate cancer outcome include prostate specific antigen levels, Gleason score, and tumor stage. However, additional measures are needed to better predict which high-risk prostate cancer patients will develop metastatic progression after primary treatment. Evans et al. hypothesize that a method to measure DNA damage and repair would provide prognostic utility for high-risk prostate cancer patients. Gene expression data from FFPE prostatectomy samples from 1090 patients with high-risk prostate cancer was analyzed to profile nine DNA damage and repair pathways. Further DNA damage and repair pathway gene mutation in cohorts from the literature were also analyzed. DNA damage and repair pathways were discovered and found to be weakly correlated with clinical variables. DNA damage and repair pathway genes were found to be rarely mutated. A DNA damage and repair signature was found to be significantly associated with overall survival and lack of recurrence and metastasis, with stronger performance in younger patients. These findings may be useful in stratifying high-risk prostate cancer patients.

RNA-Seq of Tumor-Educated Platelets Enables Blood-Based Pan-Cancer, Multiclass, and Molecular Pathway Cancer Diagnostics Best et al. report the use of mRNA sequencing of tumor-educated blood platelets (TEPs), which have an altered RNA profile due to involvement in tumor growth signaling, for improved cancer diagnosis. This technique is a blood-based biopsy that is able to distinguish primary versus metastatic tumors as well as the location of the primary tumor with 96% and 71% accuracy, respectively. Further, tumors that were MET or *HER2*-positive or had mutant forms of *KRAS*, *EGFR*, or *PIK3CA* were accurately identified. These results represent a major accomplishment in the field, as this is the first liquid biopsy diagnostic that can both detect localized disease and be employed for multiclass cancer diagnosis.

Oriented Immobilization of Fab Fragments by Site-Specific Biotinylation at the Conserved Nucleotide Binding Site for Enhanced Antigen Detection Technological advancements in molecular diagnostic applications come along with the reduced size of the devices that necessitate the oriented immobilization of antibodies and antibody fragments. Researchers from the University of Notre Dame developed a UV photo-cross-linking functionalization method named UV-

NBS. Using this method, they site-specifically functionalize anti-PSA antibody fab fragments with an IBA-Biotin linker at the Nucleotide Binding Site (NBS) without unfavorably impacting its antigen binding activity. Then, they immobilize the site-specifically biotinylated anti-PSA fab fragments on a streptavidin coated detection surface. Their results indicate that the UV-NBS method provides not only the highest level of the immobilized fab fragment on the surface, but also, because the antigen binding activity is preserved, antigen detection sensitivity is significantly enhanced compared to other typical immobilization methods: physical adsorption, NHS-Biotin labeling, and ε-NH_3^+ crosslinking. The limit of detection for PSA utilizing the UV-NBS method was also notably lower than other methods, with an LOD of 0.4 pM PSA.

An Effective Immuno-PET Imaging Method to Monitor CD8-Dependent Responses to Immunotherapy Many immune therapies cause systemic changes in immune cell number and localization, but there are currently no good methods for monitoring whole-body immune responses to immunotherapy. Positron emission tomography (PET) has the potential to measure changes in immune cell levels both intratumorally and systemically. Tavaré et al., describe an anti-CD8 antibody labeled with the positron-emitting radionuclide ^{89}Zr. This PET tracer was used to track T-cells in three different mouse models of immunotherapy (antigen-specific adoptive T-cell transfer, anti-checkpoint blockade therapy, and agonistic antibody therapy). This approach could be used as a companion diagnostic to noninvasively profile responses to cancer immunotherapy.

Tumor exosome integrins determine organotropic metastasis Hoshino *et al.* reported the role of exosomes in metastatic organotropism. Using animal models, they showed that tumor-derived exosomes direct organ-specific metastasis by preparing a pre-metastatic niche for tumor cells. Proteomic approaches of exosomes from brain, liver, and lung cancers revealed 40 adhesion molecules, with integrins being the most abundant molecule. Subsequent investigation determined that the expression of integrins in exosomes is cell-type dependent. Specifically, integrin alpha 6 ($ITG\alpha_6$), $ITG\beta_4$, and $ITG\beta_1$ enriched in exosomes are associated with lung metastasis, whereas $ITG\beta_5$ found in exosomes promoted liver metastasis. RNA sequencing studies into the downstream targets of exosome interactions found that cell migration and pro-inflammatory genes, such as *S100* proteins, along with expression of Src, were upregulated in resident cells which interacted with exosomes. Importantly, analysis of plasma samples from patients showed that patients with lung metastasis had increased $ITG\beta_4$ levels, as compared to control, indicating that integrins can be used as a metastatic biomarker. Collectively, this study reveals a mechanism behind metastatic organotropism, thus proposing exosomal integrin-centered molecular signatures as diagnostic and therapeutic tools.

Single-Cell analysis reveals a stem-cell program in human metastatic breast cancer cells Lawson *et al.* showed that early stage mammary metastatic

cells displayed stem-like gene and protein expression profiles at the single-cell level. A microfluidic-based platform was used to multiplex gene and protein expression profiles to compare metastatic cells from tissues with low (in early-stage) versus high (in advanced-stage) metastatic burden. Interestingly, low-burden metastatic cells showed a distinct stem-like signature, which distinguishes them from the primary tumor cells, whereas high-burden metastatic cells were more similar to primary tumor cells. Additionally, single-cell analysis revealed that ~1.4% of primary tumor cells displaying stem-like signatures resided with low-burden metastatic cells. Indeed, such rare subpopulation of stem-like cells, possessing tumor-initiating potentials, was correlated with metastatic potential. Moreover, high-burden metabolic cells showed higher levels of cell-cycle promoting genes, indicating higher proliferative signatures than low-burden metastatic cells. Collectively, single-cell analysis of low-burden and high-burden metabolic cells advanced our understanding of the initiation and progression of tumor metastasis, providing new potential targets for metastatic disease.

Aptamers Selected to Postoperative Lung Adenocarcinoma Detect Circulating Tumor Cells in Human Blood Zamay *et al.* report the development of DNA-based aptamers that recognize protein biomarkers on circulating tumor cells (CTCs) with high affinity from clinical blood samples. Cell-SELEX technique allowed them to select anti-lung adenocarcinoma aptamers using postoperative lung cancer tissues. Additionally, they treated blood samples with hypotonic solutions to lyse erythrocytes and leukocytes, which help the selected aptamers bind to CTCs from the blood samples. Subsequent identification of aptamer-associated protein biomarkers for lung cancer revealed that various aptamer clones recognized various proteins as biomarkers of CTCs. Given the presented work flow would be applicable to specific cancer patients, such tumor-specific DNA aptamers selected from specific patient tumor tissues may possess great potential to advance noninvasive diagnostic strategies available in clinics.

Resensitization to Crizotinib by the Lorlatinib ALK Resistance Mutation L1198F Disease resistance is one of the biggest challenges in cancer care. Relapsing patients are usually treated with targeted therapies. Resistance to crizotinib is mediated by a variety of mechanisms, including mutations in the catalytic domain of ALK. Lorlatinib is a new-generation ALK inhibitor in early phase clinical testing. Shaw and co-authors report a case study of a 52-year-old woman with metastatic ALK-rearranged non-small-cell lung cancer (NSCLC) whose first line therapy included crizotinib, lorlatinib and others. When the cancer relapsed on lorlatinib, molecular assessment of the lorlatinib-resistant tumour specimen enabled the identification of a novel ALK aberration (ALK L1198F) that resulted in this resistance. While this mutation caused resistance to lorlatinib, it paradoxically enhanced sensitivity to crizotinib. Subsequent crizotinib therapy led to immediate response and improvement of overall condition.

Imaging, Biodistribution, and Dosimetry of Radionuclide-Labeled PD-L1 Antibody in an Immunocompetent Mouse Model of Breast Cancer PD-L1

(programmed cell death ligand 1) is a protein expressed on cells in the tumor microenvironment. Antibodies to PD-L1 that block binding of PD-L1 to the T-cell effector PD-1 (programmed cell death 1) are currently in clinical trials against a number of different cancers. Patients who overexpress PD-L1 typically respond better to anti-PD-L1 immunotherapy than those who do not, so that it would be beneficial to diagnose a patient's PD-L1 expression levels prior to treatment. Josefsson, et al. joined a PD-L1 antibody to a single photon emission computed tomography (SPECT) radionuclide, 111In, to develop a SPECT tracer that allows for imaging of breast tumor isografts in mice capable of producing T-cells. This work has the potential to be useful in companion diagnosis, but also in radioimmunotherapy, where an appropriate radionuclide can kill cells responsible for suppressing antitumor immunity.

4. Profile of a Young Scientist

	**Employment**	
	2016-	Assistant Professor at Department of Chemistry, University of Texas, Austin
	2012-2015	Postdoctoral Research at Stanford University (Adviser Prof Richard N. Zare)
	Education	
	2008-2012	Ph.D. in Chemistry, Purdue University
	2004-2007	B.S. in Chemistry, State University of Campinas, Brazil

Dr. Livia S. Eberlin is an assistant professor in the Chemistry Department of The University of Texas at Austin. She received her bachelor's degree in chemistry from the State University of Campinas, Brazil, in 2007, and her Ph.D. in analytical chemistry from Purdue University in 2012. In recognition of her doctoral thesis research in mass spectrometry, she and her doctorate adviser, Prof. R. Graham Cooks, received the Nobel Laureate Signature Award for Graduate Education in

Chemistry from the American Chemical Society, given to the best PhD thesis in Chemistry. Eberlin then moved to Stanford University for her postdoctoral research in the Department of Chemistry, supervised by Prof. Richard N. Zare. During her postdoctoral research, she received the L'Oréal for Women in Science Fellowship, a K99/R00 Pathway to Independence award from the NCI/NIH, and was named among the Forbes 30 under 30' list in Healthcare.

Dr. Eberlin is passionate about interdisciplinary research at the interface of chemistry and medicine. Her group focuses on applying novel mass spectrometry imaging technology to health related research. Dr. Eberlin's main goal is to develop innovative ambient mass spectrometry techniques in combination with statistical and molecular biology tools to investigate the molecular information from a variety of biological samples. In particular, her laboratory is interested in using ambient ionization mass spectrometry in creative ways to address critical problems in cancer research. In basic cancer research, the Eberlin laboratory studies tumor tissue behavior and chemically characterizes metabolic signatures of human cancers. Her laboratory also pursues improvements in mass spectrometry instrumentation in order to translate the tools developed in the laboratory to the clinic for *in situ*, real-time disease diagnosis. Her interdisciplinary research is carried out in collaboration with biologists, oncologists, surgeons, pathologists, statisticians and engineers to develop powerful approaches that can be used in real life scenarios such as in clinical practice.

5. Spotlight on World News

FDA Grants Breakthrough Therapy Designation for AstraZeneca's Durvalumab The FDA granted breakthrough therapy designation for AstraZeneca's durvalumab as a treatment for bladder cancer. The drug is a monoclonal antibody directed against PD-L1 is for the treatment of PD-L1 positive patients with inoperable or metastatic urothelial bladder cancer. The breakthrough therapy designation is meant to expedite the development of new drugs based upon encouraging clinical results.

US cancer institute to overhaul tumor cell lines After more than 25 years of heavy use by researchers around the world, the US National Cancer Institute (NCI) has decided to retire the NCI-60, its panel of 60 human cancer cell lines grown in culture. In late spring of this year, the institute will launch a rejuvenated repository of cancer models that are derived from fresh patient samples and tagged with details about their clinical past. The NCI effort reflects a wider trend focused on developing repositories of PDX models, which more closely reflect the clinical situation.

6. Career Forum

   https://cancercareers.org/Pages/default.aspx

   http://www.nature.com/naturejobs/science/jobs

   http://jobs.rsc.org/

   http://chemistryjobs.acs.org/

7. Conferences

   AACR Annual Meeting 2016
   April 16-20, 2016; New Orleans, Louisiana

   Proteomics: From New Technology to New Biology
   March 13-16, 2016. Boston, Massachusetts

   Novel Approaches for Cancer
   March 15-16, 2016. London, United Kingdom

   ASPET Annual Meeting at Experimental Biology
   April 2-6, 2016; San Diego, California

   Accelerating Anticancer Agent Development and Validation Workshop
   May 4-6, 2016; Bethesda, Maryland

   AACR Precision Medicine Series: Targeting the Vulnerabilities of Cancer
   May 16-19, 2016; Miami, Florida

   High Throughput Chemistry & Chemical Biology
   June 14-19, 2016; Colby-Sawyer College, New London, New Hampshire

   Chemical Biology in Drug Discovery
   June 16-17, 2016; Boston, Massachusetts

   Engineering and Physical Sciences in Oncology
   June 25-28, 2016; Boston, Massachusetts

   National Medicinal Chemistry Symposium
   June 26-29, 2016, Chicago, Illinois

   EMBL Chemical Biology Conference
   August 31-Sept 3, 2016; EMBL Heidelberg, Germany