Acute Leukemia Translational Research Initiative Planning Workshop Report

January 26, 2016
8:30 am – 4:30 pm
Location: OICR | West Tower Boardroom 5-20/21

Attendees

Invited

Melissa Anders	University Health Network (UHN)
Cheryl Arrowsmith	Structural Genomics Consortium (SGC)
Harry Atkins	Ottawa Hospital Research Institute
Gary Bader	University of Toronto
Dalia Barsyte	SGC, University of Toronto
Mick Bhatia	McMaster University
Steven Chan	University Health Network
Jayne Danska	The Hospital for Sick Children (SickKids)
John Dick	University Health Network
Cynthia Guidos	SickKids
Vikas Gupta	Princess Margaret Cancer Centre
Hans Hitzler	SickKids
Michael Hoffman	University Health Network
Keith Humphries	Terry Fox Lab, BC Cancer Agency
Norman Iscove	Princess Margaret Cancer Centre
Natasha Kekre	The Ottawa Hospital
Brian Leber	McMaster University, Juravinski Cancer Centre
Mathieu Lupien	Princess Margaret Cancer Centre
Mark Minden	UHN
Meaghan O'Reilly	Sunnybrook Research Institute
Chris Paige	UHN
Rob Rottapel	Princess Margaret Cancer Centre, UHN
Mitchell Sabloff	The Ottawa Hospital
Len Salmena	University of Toronto
Aaron Schimmer	Princess Margaret Cancer Centre
Liran Shlush	UHN
Paul Spagunolo	University of Waterloo
Jean Wang	UHN
Jim Whitlock	SickKids
Karen Yee	Princess Margaret Cancer Centre

OICR & FACIT

Rima Al-awar	Director, Drug Discovery Program
Philip Awadalla	Principal Investigator, Ontario Health Study Program
John Bartlett	Director, Transformative Pathology Program
Robert Campos	Head, Research Operations
Jeff Courtney	Chief Commercial Officer, FACIT
Craig Earle	Director, Health Services Research Program

Tom Hudson	President and Scientific Director, OICR
David O'Neill	VP, Business Development, FACIT
Rebecca Tamarchak	Director, Strategic Planning and Outreach

Guest

Dawn Richards	Medical Writer

Please note that this is a summary of the workshop prepared by the organizers. For more details please contact the TRI workshop leaders:

- John Dick: jdick@uhnresearch.ca
- Aaron Schimmer: aaron.schimmer@utoronto.ca
- Mitchell Sabloff: msabloff@toh.ca

1. Workshop goals and deliverables/outcomes

The OICR Cancer Stem Cell (CSC) Program began in 2007 and was reviewed 3 years ago. Its focus has been on identification of genetic determinants of tumour heterogeneity, genetic diversity, the tumour microenvironment, epigenetic pathways and development. The concept of stemness is viewed as an important tumour feature and how it influences outcomes is key to the Program. While the Program started with a focus on leukemia and brain cancer, it has progressed and built expertise in other systems, breaking down silos to take stem cell thinking into account when investigating cancer biology. The Program has also been able to significantly leverage OICR's initial investment into additional grant support, clinical trials, and commercialization opportunities.

The current CSC Program will be ending March 31, 2017, and instead, two of its longstanding primary foci of leukemia and brain cancer will be developed as their own Translational Research Initiative (TRI) proposals. This represents both an important evolution in the CSC Program and highlights the competitiveness of the cancer stem cell hypothesis in the areas of leukemia and brain cancer. The focus of the workshop was to consider if measuring or targeting stemness properties affect leukemia patient outcomes. Both basic scientists and clinical leaders were invited to the workshop to discuss the potential to organize a pan-Ontario acute myeloid leukemia (AML) network. Subsequent to the workshop, TRI workshop planning leaders will work with specific workshop participants to develop a Letter of Intent (LOI) around Acute Leukemia.

2. Background

- OICR Strategic Plan 2016-2021: overview

Tom Hudson presented OICR's 5-year strategic plan (2016-2021), including the Institute's mission and goals. Important highlights include the aims to: advance Ontario's best cancer research to improve cancer care and treatment; perform cutting-edge translational cancer research, enhancing Ontario's global leadership in cancer research; partner with the Ontario cancer community; and drive adoption and/or commercialization of cancer innovations in Ontario. Emphasis was placed on OICR's community outreach efforts, intended to forge collaborations and to move the most promising ideas to the clinic for impact.

- Translational Research Initiatives

The Institute's concept of Translational Research Initiatives (TRIs) was discussed, which require expertise, funding and innovative approaches to move findings and technologies to the clinic. Workshop participants were encouraged to consider leveraging networks supported by OICR (e.g., Global Alliance for Genomics & Health, Ontario Tumour Bank, Canadian Cancer Clinical Trials Network) as well as OICR's Technology Programs that can play a role in providing expertise and access to technologies to the Ontario research community, and in supporting OICR's strategic initiatives.

TRIs represent large scale, multi-disciplinary collaborations between laboratory and clinical scientists, to advance Ontario assets and improve cancer patient outcomes. They should focus on a clinical need that builds on assets or innovations in an area of leadership for Ontario. TRIs will include 2-5 projects, with at least one mandatory clinical trial that must begin within the first 2 years of a TRI. A budget of up to $10 M over 4 years may be requested for a TRI, with the clinical trial budget comprising at least $2 M over the 4 years (if less than $2 M, the total budget will be reduced accordingly). Additional supplemental funding from external sources should be sought at the start of the TRI and throughout the course of the TRI.

TRI workshops are intended to support development of LOIs for TRIs. TRI's will be led by two co-leaders (preferably not from the same institution), one scientific and one clinical, while the TRI manager will provide coordination and administrative capabilities. The eventual TRI leaders are not necessarily the same individuals as the TRI planning workshop committee.

The timing for TRI development was described as follows:

- Declaration of interest – by April 15, 2016
- LOI submission – May 2, 2016 (Note: The LOI will include an overview statement (1 page), research plan summary (4 pages), description of the team (3 pages), and high-level budget)
- LOI selection – July 15, 2016 (Note: Externally peer-reviewed. Only TRI LOI submissions rated medium or high will be invited for a TRI Funding Request submission)
- TRI Funding Request submission – October 31, 2016
- TRI Funding Request international review panel – February 2017
- Funding begins – April 2017.

Workshops are intended to build consensus around the TRI priorities, discuss potential projects, identify collaborations among Ontario scientists, consider how to best leverage OICR Technology Programs, and identify potential sources of co-funding. A workshop report will be generated to inform the community about the workshop, allowing those who were unable to attend the workshop an opportunity to provide input or become a participant in an application, and to facilitate LOI applications. Although more than one LOI per theme may emerge and will be accepted for review, the group was reminded that this will be a competitive process with no guaranteed funding outcomes. There was a reminder that all guidelines about the TRI process are online (https://events.oicr.on.ca/tri-workshop-leukemia-guidelines) and if there are any questions about the process, to contact OICR's Scientific Secretariat. Participants were specifically reminded that TRI funding is not eligible for clinical trial overhead since the Canadian Cancer Clinical Trials Network funds infrastructure related to clinical trials already.

3. Presentation Summary

There were several presentations focused on progress in OICR AL projects, including clonal diversity and stemness, biomarkers and signatures, potential therapeutic targets and approaches as well as a series of speakers on" hot topics" in acute leukemia related to novel therapeutic strategies and mechanisms of stemness. Clinical leaders presented various clinical leukemia programs and their resources across Ontario highlighting the opportunity to harness Ontario's single payer system and its ability to look at clinical data, and perform high quality research in this environment. A series of technology platforms were also presented that may provide potential collaborations for AL researchers, to specifically address some of the barriers that were discussed throughout the workshop. These technologies included the following areas: pathway and network analysis, Structural Genomics Consortium, population genomics, DNA methylation, epigenomics and related computational biology. Finally there was a group discussion around what the biggest clinical needs are and ways to address them that would dramatically advance the field (Please see agenda for details on speakers).

4. Workshop Deliverables

Here the workshop leaders have attempted to summarize the results of the workshop organized around OICR's expected deliverables to a) identify the clinical needs of acute leukemia patients and b) to identify projects that may address these needs. In addition, we highlight opportunities and begin to organize ourselves into projects that may be suitable for the LOI.

Clinical Needs of Acute Leukemia Patients

There were two 45-minute discussions during the workshop revolving around the projected future state of clinical care of AL, and research projects that would help drive us towards these ends. Several needs were discussed including the following:

- Physicians require tools to precisely monitor disease burden at the time of remission in a sensitive way that would allow them to react prior to overt relapse. Related to the monitoring of minimal residual disease would be the availability of therapeutic interventions such as small molecules or immune modifiers, including transplant. In addition, we need to have therapeutic options that target leukemic stem cells.
- We have identified genetic signatures based on stemness that are both highly predictive of initial response to therapy (LSC17 score) and prognostic for future recurrence (LSC3 score). We need to determine how to use these signatures in the clinic to identify patients at high risk for relapse or not responding to upfront therapy.
- AML is a heterogeneous disease with mutations spread over many clones. It would be useful to have a tool that will take into account a more comprehensive analysis of mutations that will enable description of distinct subpopulations and their response to therapy and during MRD. Being able to monitor clones from diagnosis through treatment, understanding the specific mutations that define those clones, and assessing which clinical trials for which patients from this standpoint would also be important. Beyond somatic mutations, the group also recognized that, epigenetics, drivers of regulation, etc. also influence the heterogeneity of AML and response to therapy.
- The need to monitor predictors of relapse, through the development of biomarkers of relapse and to help develop trials when residual disease is present. A strategy that would include treating before the disease is fully obvious may be a consideration.

- In younger patients, relapse is the biggest unmet clinical need. In addition, novel therapies for patients with primary refractory leukemia are important.
- It was recognized, that many elderly patients are too frail for standard induction chemotherapy and novel, effective, and less toxic therapies are required. To this end, therapies targeting LSCs may be effective (See above).
- The group considered patients who develop aplasia after induction chemotherapy for AML. Do these patients have normal hematopoietic cells to come back? Are their stem cells depleted?
- The pre-leukemia concept was identified as a potentially transformative approach for leukemia prevention, if you could determine which people need treatment before disease onset.

Barriers to Overcoming Clinical Needs

- The current drug development paradigm fails to account for patient heterogeneity and clonal heterogeneity and for that reason, clinical trials on non-selected patient populations often fail.
- Patient-derived xenografts allow identification of variants indicative of unique AML populations; however, using xenografts to select chemotherapeutic options for a patient could be too slow in a clinical setting given the urgency of treating AML. As a companion study to a clinical trial, it would be possible to apply a xenograft-based study to capture information on the biology of cells in parallel to a clinical trial - the need for biomarkers are clear.
- Lack of tools to propagate primary cells in culture; however, genetics are much more straightforward to obtain. Infrastructure for the sequencing throughput required for clonality experiments are lacking across the province

Potential projects to address the specific clinical needs

As a result of the presentations and discussions at the workshop, several project ideas are under development for the LOI submission.

Clinical Network Project

There was discussion of developing a clinical trial network to speed accrual to trials and increase the availability of primary samples for study. To facilitate such a network, it would be important to collect clinical and other information since no one site represented by the streamline clinical strategies so patients are treated in a standardized manner with common protocols, common lab data and clinical data. There is also a leveraging possibility with large multinational co-operative groups in adult and pediatric AML and ALL that bank samples and do trials, offering to perform analysis, and once sophisticated multivariate analysis to test observations is demonstrated, they could test their findings in a prospective clinical trial. Based on the input from the workshop, we are considering proposing a pan-Ontario clinical network as a part of the TRI that would include the largest leukemia programs in the province, which together sees more than 300 new AL cases per year (Princess Margaret: PI=Schimmer; Ottawa: PI=Sabloff; and McMaster: PI=Leber; SickKids: Hitzler). Through this network, we would develop common protocols for laboratory investigation and treatment of AL and run clinical trials associated with the AL TRI. These will enhance accrual to existing or coming LSC-based trials. More specifically this infrastructure will provide support for ethics, biostatistics, coordination of patient recruitment, clinical trial management and trial monitoring through existing and new resources at the Princess Margaret. Moreover, Ontarian investigators would have access to a centralized infrastructure for clinical trials in AL using a secure web interface that will be developed as part of this project.

Clinical Trials

Clinicians among the participants felt that it is not only necessary to install a combination of chemotherapy required in the face of heterogeneity in the stem cell compartment, and to target LSCs, but also to treat the variety of lesions and to assess treatment in real time. To resolve these issues, we are considering initiating a multi-center pan-Ontario trial that will risk stratify patients based on current and novel genetic markers and will evaluate a series of LSC-targeted therapies. We could address some of the clinical questions that were raised: 1) Will introducing biomarkers (such as LSC3 and LSC17 scoring) better predict patient outcome? 2) Can we identify specific clones resistant to novel LSC-based therapies? 3) Can LSC-targeted therapies produce responses in high-risk patients?

Prevention of Relapse

Understanding the origins of relapse will help us to develop better therapies. We envision an approach that takes into account both the subclonal genetic diversity as well as the LSC properties. Studying stem cells to learn more about the clonal history of an AML tumour was discussed during the workshop. In-depth examination of an individual's blood cells including both leukemic blasts, pre-leukemic cells and non-leukemic "normal" blood stem and progenitor cells has allowed delineation of the origins of their AML, including clonal expansion and multi-lineage delineation. Furthermore, this has also permitted study into the origin of relapse and implications for treatment, and the contribution of genetic diversity to recurrence and therapeutic resistance. A specific example was described revealing the identification of a minor clone at diagnosis that became a major clone at relapse. Functional consequences are now being studied using xenografting methods. Understanding pre-leukemia will also provide an avenue into further understanding of relapse. Overall, progress in this area will require a focussed effort across the pan-Ontario network to enable the complex flow sorting, low cell input/single cell deep sequencing, and sophisticated informatics on sequential analysis of patients following their diagnosis. Such an infrastructure would greatly enhance our scientific understanding of disease progression as well as providing a valuable tool upon which clinical trials could be layered making our network a highly sought partner.

Biomarkers to address monitoring of disease

Previous investments from OICR have led to linking leukemic stem cell (LSC) properties to AML outcomes. Patient samples have been sorted based on phenotypic markers and a LSC gene signature was determined that can be used to stratify patients. The next step will be to move this forward to the clinic as a tool for physicians (Wang).

Sophisticated tools are being developed to monitor disease and to determine outcomes associated with drug response such as phosphoflow and mass cytometry (CyTOF) to discover biomarkers of leukemia drug responses, especially with the latter's ability to examine characteristics of single cells while routinely interrogating 30-40 different properties in parallel (Guidos). Perhaps the CyTOF technology could be a tractable option to mine signaling pathways, etc., to capture clonal information or drug response in real time in clinical trials. There is also the possibility of localization of LSCs and utilizing PET-CT for spatial imaging (Bhatia).

Therapeutics and therapeutic testing to address targeting LSCs

Overall the group considered that knowledge of normal stemness properties will be essential to understand the leukemic counterpart. Such information is essential to identify targets for therapy. For example, the genes of the homeobox cluster

(Iscove) represent central components of both HSC and LSC and numerous transcriptional and epigenetic/miRNA pathways have been identified in Ontario labs that control the stemness of cells. Several of these have potential therapeutic possibilities either to perturb self-renewal or to release LSC from their quiescence making them more sensitive to therapy. Several research areas were presented by Jayne Danksa (SYK and niche), Aaron Schimmer (ClpP), Mick Bhatia (niche), Steven Chan (mitochondrial pathways), Lenny Salmena (INPP4b), Paul Spagnuolo (fatty acid metabolism) where stemness vulnerabilities have been identified that could be potential therapeutic targets or lead to an increased knowledge of stemness. In general we envision a program that exploits the current expertise of the community that is focussing on the following four main therapeutic strategies targeting stem cell specific properties: mitochondria/metabolism, signalling, niche, transcription factor networks, and epigenetic/miRNA control.

It was agreed that the use of primary leukemia xenografting as a means to evaluate drug efficacy across diverse sets of patients is one of Ontario's strengths and a unique resource to test drugs and/or modulate drug resistance. Clinical trial scale testing of primary leukemia response also enables the development of response biomarkers that could be used as a companion biomarker in human trials. In addition to xenografting, there is need to develop a faster means to evaluate drug response. The highly multiplexed CyTOF approach represents one means to capture the high dimensional data required at the single cell level to be able to make relevant predictions of response while still monitoring stemness properties. One potential project would be to extend the xenograft resource for testing therapeutics and make it available across Ontario, thereby increasing the collective chances of success. In addition, by systematically providing additional and improved clinical genomic information on samples in biobanks, more questions about biomarkers can be answered, which will be extremely beneficial to everyone involved.

5. Conclusions

There is a need to balance dreaming big and pragmatism. The challenge in AL is to develop more effective and less toxic therapy that improves the rate of remission and reduces the rate of relapse. Better monitoring of patients in remission to predict relapse is an important goal. All of these objectives require an improved understanding of the biology of AL cells and their related stem cells. OICR is enabling an opportunity to nucleate the strengths, to relieve choke points and deliver on a cohesive aim.

Acute Leukemia Translational Research Initiative Planning Workshop

January 26, 2016
8:30 a.m. – 4:30 p.m.
Light breakfast will be served at 8:00 a.m.
Location: OICR | West Tower Boardroom 5-20/21

TIME	AGENDA ITEM	PRESENTER
8:00 a.m.	**Arrivals and light breakfast**	
8:30 a.m. – 8:40 a.m.	**Opening remarks** • Workshop goals and deliverables/outcomes • Introduction of Workshop Planning Committee	*John Dick*
8:40 a.m. – 9:00 a.m.	**Background** • OICR strategic Plan 2017-2021: overview • Translational Research Initiative (TRI): overview, available funds, linkages to platforms and existing projects • Letters of Intent: Declaration, submission	*Tom Hudson*
9:00 a.m. – 10:00 a.m.	**Progress in OICR Acute Leukemia Projects (15 min each)** • Clonal diversity and Stemness a. John Dick - ALL b. Liran Shlush -AML • Biomarkers and Stemness a. LSC signature - Jean Wang b. CyTOF - Cynthia Guidos	*Session Leader: John Dick*
10:00 a.m. – 10:20 a.m.	**Break**	
10:20 a.m. – 11:40 p.m.	• Therapeutic Targets and Stemness a. SYK and RANKL - Jayne Danska b. Msi2 - Kristin Hope c. ClpP - Aaron Schimmer	*Session Leader: John Dick*
11:40 p.m. – 12:30 p.m.	**Lunch**	

12:30 p.m. – 1:30 p.m.	**Clinical Leukemia Programs (15 min each)** Suggested topics - Current diagnostic and clinical capabilities, companion studies, volumes, one interesting clinical/translational research project • Hans Hitzler - Sick Kids • Brian Leber - McMaster • Mitch Sabloff and Natasha Kekre - Ottawa • Karen Yee - Princess Margaret	*Session Leader: Mitch Sabloff*
1:30 p.m. – 2:45 p.m.	**Hot Topics in Acute Leukemia (15 min each)** • Mick Bhatia • Steven Chan • Norman Iscove • Lenny Salmena • Paul Spagunolo	*Session Leader: John Dick*
2:45 p.m. – 3:00 p.m.	**Break**	
3:00 p.m. – 3:50 p.m.	**Technologies - Lightning Round (5 min each)** • Cheryl Arrowsmith - SGC • Phillip Awadalla - genomics • Gary Bader - computational biology • Daniel DeCarvalho - DNA methylation • Michael Hoffman - computational biology • Mathieu Lupien - epigenomics • Mark Minden - biobank	*Session Leader: Gary Bader*
3:50 p.m. – 4:20 p.m.	**Group Discussion** What do you think is the biggest clinical need in acute leukemia? Describe an innovative or homerun proposal that would not incrementally but dramatically change the field?	*Session Leader: Aaron Schimmer*
4:20 p.m. – 4:30 p.m.	**Next steps/wrap up**	*John Dick*