

Summary of 14th Annual MMIG Meeting (Merkel cell carcinoma Multi-Center Interest Group)

Friday March 1, 2019 (summary version date May 20, 2019)

American Academy of Dermatology Annual Meeting, Washington, DC

Prepared by Song Park, Paul Nghiem, Tomoko Akaike and Coley Doolittle-Amieva

Announcements:

- 1) 1st International Symposium on Merkel Cell Carcinoma – October 21-22, 2019, Moffitt Cancer Center, Tampa, FL
This conference is a first-of-its kind gathering of thought leaders and active investigators in MCC. The goals are to share new unpublished scientific developments, promote scientific discourse, and establish new collaborations that can improve our understanding of MCC biology, pathogenesis, and therapy. Detailed program and information can be found at <https://moffitt.org/for-healthcare-providers/continuing-education/provider-conferences/international-symposium-on-merkel-cell-carcinoma/> . If you are interested in attending this meeting or have specific questions, please contact Dr. Tsai (Kenneth.Tsai@moffitt.org). **Abstract submission deadline is May 31, 2019.** Some trainee fellowships will be made available for excellent abstract submissions.
- 2) Video of Plenary Lecture, “Less toxic, more effective: A win-win for Merkel cell carcinoma patients”, by Paul Nghiem at the 2019 AAD Annual Meeting is available at <https://merkelcell.org/news-and-publications/2019/american-academy-of-dermatology-2019-plenary-session/>
- 3) If you are interested in presenting at next year’s MMIG meeting in Denver, CO, on Friday March 20, 2020 (5 – 7 pm), please send Paul an email (pnghiem@uw.edu) with a proposed topic that is relevant to MCC patient care or translational research.

Speakers/Topics (detailed in following pages):

1. **Which biomarker approach best predicts response to PD-(L)1 blockade: PD-L1 IHC, tumor mutation burden, gene expression profiling, or multiplex IHC/IF?**
Steve Lu (Johns Hopkins)
2. **T-cells and survival in Merkel cell carcinoma: Quantity and quality matter**
Michael Tetzlaff (MD Anderson)
3. **How the paranuclear cytokeratin dot may be blocking apoptosis in MCC**
Isaac Brownell & Natasha Hill (NIH)
4. **The Merkel virus antibody test: Update on outcomes/performance and a practical guide for clinicians**
Coley Doolittle-Amieva & Song Park (University of Washington)
5. **SCOUT (Skin Cancer OUTcomes) Consortium: Connecting physician experts in skin cancer with clinical trials**
Vishal Patel (George Washington University)
6. **Project Data Sphere & collecting MCC patient outcomes data: Getting a noble goal to actually work**
David Miller (MGH)

1. Which biomarker approach best predicts response to PD-(L)1 blockade: PD-L1 IHC, tumor mutation burden, gene expression profiling, or multiplex IHC/IF?

Steve Lu (Johns Hopkins)

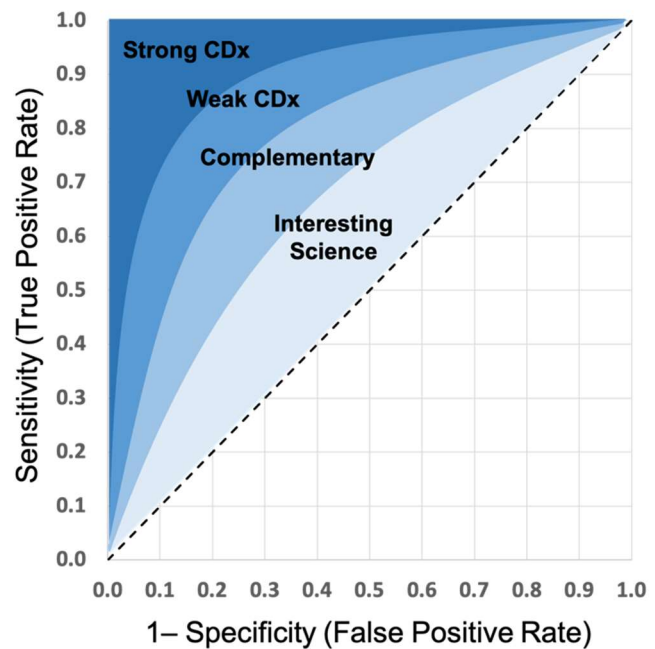
Steve Lu is an MD/PhD student who performed a meta-analysis to assess which biomarker approach best predicts response to anti-PD-(L)1 therapy across a variety of cancers. Although different biomarker assay modalities have been proposed such as PD-L1 immunohistochemistry (IHC), tumor mutational burden (TMB), gene expression profiling (GEP), and quantitative and/or spatial assessment of multiple proteins by multiplex IHC/immunofluorescence (mIHC/IF), the relative sensitivity and specificity of these approaches have yet to be systematically established. Lu reviewed 45 relevant studies by system review (PRISMA) and performed a meta-analysis to determine each approach's ability to discriminate between responders and non-responders to therapy by mapping sensitivity and specificity data for each study and calculating relative area under the curves (AUCs) as a metric of each biomarker's ability. Among these modalities, mIHC/IF has the highest area under curve (AUC).

Then Lu described additional predictive and prognostic value of combining multiple biomarkers. Although it is known that PD-(L)1 status on IHC can be a biomarker for response to anti-PD-(L)1 therapy, this status alone is insufficient to predict response to treatment. Lu introduced the idea that density of PD-1+ lymphocytes adjacent to PD-L1+ cells correlates with clinical response to anti-PD-1 therapy. That is, combining multiple biomarkers such as the density of PD-1+ lymphocytes, PD-L1+ cells and the proximity better predict response (mIHC alone shows characteristics similar to a weak companion diagnostic test).

Lastly, Lu talked about a different approach using the likelihood ratio which does not depend on prevalence. Even with this approach, mIHC/IF has benefits of both negative and positive likelihood ratio for response prediction. The data were also presented as a talk at SITC 2018, and a manuscript is in preparation.

- <http://blog.sitcancer.org/2018/11/sitc-2018-scientific-highlights-nov-9.html>

Meta-Analysis Reveals Correlations Between Response to Anti-PD-1/PD-L1 Therapy and Biomarker Assessment Methods



In order of improving performance of test: Interesting science → Complementary → Weak Companion diagnostic (CDx) → Strong CDx

2. T-cells and survival in Merkel cell carcinoma: Quantity and quality matter

Michael Tetzlaff (MD Anderson)

Dr. Tetzlaff talked about the importance of quantity and quality of T-cell infiltration in Merkel cell carcinoma.

Dr. Tetzlaff and his colleagues conducted research focusing on density and distribution of T cells. Using digital image analysis, the team quantified T cells in different locations (periphery, center or hot spot). His study showed higher density of CD8+ T-cells at the tumor periphery of MCC is a robust indicator of prognosis, associated with improved OS and DSS. This is particularly true for MCC patients who are sero-positive for MCPyV.

Dr. Tetzlaff's team then investigated the "quality" of T-cells in MCC tumors by sequencing the CDR3 beta region of the T cell receptor (TCR) and analyzed TCR clonality. To measure T-cell quality, Dr. Tetzlaff utilized Simpson's Dominance index, which is a good measurement that captures both diversity and richness of T-cell quality.

Favorable clinical features such as lower frequency of metastasis or lower stage were associated with higher Simpson's Dominance index. Furthermore, Simpson's Dominance index also correlated with longer time to first distant metastasis and longer disease-specific survival.

The combination of high Simpson's dominance score and high T cell density of tumor predicted longer disease-specific survival more accurately than either alone. These parameters can be integrated into a modified immunoscore for MCC based on T-cell quantity and quality. In summary, Simpson's Dominance Index which reflects T-cell quality is an additional informative biomarker in MCC, in addition to CD8 infiltration.

- Original publication from Dr. Tetzlaff
Feldmeyer L et al., Density, Distribution, and Composition of Immune Infiltrates Correlate with Survival in Merkel Cell Carcinoma, Clin Cancer Res. 2016 Nov 15;22(22):5553-5563.
<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5857157/>
- Al-Rohil RN et al., Intratumoral and peritumoral lymphovascular invasion detected by D2-40 immunohistochemistry correlates with metastasis in primary cutaneous Merkel cell carcinoma, Hum Pathol. 2018 Jul;77:98-107.

3. How the paranuclear cytokeratin dot may be blocking apoptosis in MCC

Isaac Brownell & Natasha T Hill (NIH)

A perinuclear dot-like pattern of cytokeratin20 (CK20) expression is a pathognomonic finding for MCC. However, some MCC tumors are negative for CK20 or have a diffuse cytoplasmic pattern. With the help of Dr. Paul Harms, Drs. Brownell and Hill assessed the expression pattern of cytokeratins in MCPyV-positive (VP) and MCPyV-negative (VN) MCC tumors. Overall 94% of MCC tumors had some cells with dot-like staining. All VP-MCC had areas of dot-like staining, whereas 7 of 53 (13%) of VN-MCC completely lacked dot-like staining.

In cells lines, only VP-MCC cell lines had CK20 dot aggregates, and the dots existed in two patterns: a paranuclear dot with perinuclear lattice or a paranuclear dot alone.

Drs. Brownell and Hill characterized the paranuclear dot further using electron microscopy. The dot was in the polar region of the cell but not in any organelles or membrane compartment and was associated with the centrosome: microtubule organizing center. The dot colocalized with the centrosome in interphase cells while it disaggregated and reformed during mitosis. Further

experiments demonstrated the microtubules play a pivotal role in maintaining the dot, but kinesins have little effect. Drs. Brownell and Hill then discovered that the CK20 paranuclear aggregate is not ubiquitinated in VP-MCC, and inhibiting ubiquitination results in aggregation of cytokeratins in VN-MCC cells.

The team also found that Fas-Associated Death Domain (FADD) localized within the CK20 paranuclear aggregate which may facilitate evasion of apoptosis signaling at the membrane. In fact, MCC cells with paranuclear CK20 aggregates showed resistance to TNF- α induced killing signals. These findings suggest the paranuclear CK20 dot may play a role of blocking apoptosis in MCC.

4. The Merkel virus antibody test: Update on outcomes/performance and a practical guide for clinicians

Coley Doolittle-Amieva & Song Park (University of Washington)

Updated data on the Anti Merkel Panel (“AMERK”) serology test as well as its use in clinical practice were presented by the University of Washington team.

Since its development 10 years ago, the AMERK test has been validated and published in several papers. Based on data that support the clinical value of this test, it was included in the NCCN guidelines in 2018 as a suggested test for initial work-up of MCC as well as surveillance.

Updated analysis of the AMERK serology test was based on the data gathered over a 10-year period (2008-2018) including 774 patients (~2000 blood draws), of which 371 were seropositive. The most updated analysis was in 2019 and included 254 seropositive patients with ≥ 2 AMERK tests. 170 patients had stable or decreasing titers, and 168 of those patients did not have MCC recurrences detectable at that time (99% negative predictive value). 2 patients were outliers with no increase in their antibody titer at the time of recurrence, however in each case their antibody titer did increase on the very next antibody test after diagnosis of recurrence. 84 of the analyzed patients had increasing antibody titers and of those, 83 patients recurred. 76 recurrences were within 45 days of increase in antibody titer and 7 recurred later, with median time to recurrence being 292 days after the antibody titer increase. 1 patient has yet to recur (99% positive predictive value).

This test is clinically useful for both high and low risk patients as well as seropositive and seronegative patients. For seropositive patients it can detect early MCC recurrences and reduce the number of scans. It has prognostic indications for seronegative patients, who are 42% more likely to have a recurrence than seropositive patients and following them closely with scans is thus appropriate. We have been working hard to create an algorithm and guidelines for clinicians who will be ordering the AMERK serology test.

The UW team believes all newly diagnosed MCC patients should be tested at baseline. If seronegative, no further antibody testing is recommended and surveillance with imaging is appropriate. If seropositive, continued surveillance with serology is clinically useful. Our algorithm outlines different scenarios in seropositive patients including stable, increasing, and decreasing titers with recommendations for managing each.

For questions interpreting the AMERK serology test you can contact us at pnghiem@uw.edu or mccteam@uw.edu. For logistical questions please contact Krista LaChance at kcs27@uw.edu.

A Detailed algorithm to interpret AMERK results will be made available through merkelcell.org/sero

- Based on suggestions made at the MMIG meeting, we will begin a multi-institutional collaborative group to focus on issues surrounding this test. Some goals will be 1) troubleshooting tricky clinical issues that arise in using this test, 2) improving the clinical algorithm for how to use the test in clinical practice, 3) A cost-effectiveness analysis (sero-positive patients typically do not require any scans after their titer has fallen), 4) Publication of collaborative studies to optimize surveillance of MCC. We plan to have teleconferences every 4-6 weeks. If you are interested in participating in this group, please email pnghiem@uw.edu or kcs27@uw.edu for detailed information.

5. SCOUT (Skin Cancer OUTcomes) Consortium: Connecting physician experts in skin cancer with clinical trials

Vishal Patel (George Washington University)

Dr. Patel introduced SCOUT, a new consortium physician from multiple specialties who are expert in the study of non-melanoma skin cancer including squamous cell carcinoma and Merkel cell carcinoma. Dr. Chrysalynne Schmults (BWH) is a co-founder of SCOUT.

SCOUT was formed in Oct 2018 to facilitate and improve research in non-melanoma skin cancer care and provide members and industry with a professional network for collaboration. It provides to the pharmaceutical and biologic industry access to physician experts through a consortium comprised of expert members.

Physicians are invited as individuals (not as representatives of their employers) and may withdraw membership at any time. Members may be dermatologists, Mohs surgeons, medical oncologists, radiation oncologists, or specialists in surgical oncology, head and neck, or plastic surgery.

As SCOUT members, their names and professional titles are included in SCOUT materials. Members may (but are not required to) work on behalf of SCOUT to consult with companies in the biologic and pharmaceutical industry to develop and provide advice on clinical research study protocols for new drugs, devices and services. Of note, this does not preclude SCOUT members for providing consultation as individuals.

Members may attend the SCOUT Annual Meeting during ASCO and EADO to facilitate collaboration and knowledge in non-melanoma clinical care and research including facilitating investigator-initiated clinical trials. SCOUT revenue from consulting work or other funds (after covering administrative costs) will be distributed to members in the form of competitive research grants.

SCOUT is currently comprised of > 30 centers / sites and >40 members over the world, and continuously expanding its role. If you are interested in being a SCOUT member and attending the upcoming meeting in Chicago (During ASCO, May 31-June 3), please contact info@scoutconsortium.org or visit <https://www.scoutconsortium.org/>

6. Project Data Sphere & collecting MCC patient outcomes data: Getting a noble goal to actually work

David Miller (MGH)

Dr. Miller introduced a multi-center MCC patient registry in collaboration with Project Data Sphere (PDS) and updated progress on the project.

MCC is a rare cancer, and current clinical outcomes research on MCC are limited to large administrative data sets (e.g. NCDB), which often lack nuanced clinical information, or single-institution databases. The relatively small number of MCC patients dispersed in multiple institutions could potentially limit future development of drugs due to the difficulty of meeting significant “n” for approval.

The PDS MCC Registry is a multi-institutional collaborative effort that will prospectively follow and record outcome and events in MCC patients. It will adopt new methodologies, that will enable multiple investigators to examine real world outcome data in real time. Data from the Registry could be used to determine: (i) precise patient stratification into risk categories, (ii) identification of best practices, (iii) revelations about optimal sequence and combinations therapies, (iv) uncovering low incidence toxicities and (v) the generation of novel testable hypotheses. Importantly, the Registry offers a way forward in the yet-unsolved dilemma of drug development for rare tumors since the Registry’s design will allow for the creation of highly defined patient-level data that can be used as a robust comparator for single arm Phase I-II clinical trials.

The MCC Task Force comprises members from academic medical centers, the drug industry, the NIH and FDA. Project Data Sphere provides a secure, open-access data sharing platform designed to optimize research and yield rigorous and timely results. The Registry is utilizing REDCap as the electronic data collection system. The Task Force team is currently building consensus data fields, and will set up a detailed plan of data access model, etc. Test of a beta version and development of data quality plan and usage agreements are soon to be executed.

If you are interested in participating in this project, please contact Dr. David Miller at DMILLER4@mgh.harvard.edu

In attendance at the 2019 Washington, D.C. MMIG meeting

*= Joined online via Zoom

Afanasiev, Olga (Stanford University)
Akaike, Gensuke (University of Washington) *
Akaike, Tomoko (University of Washington)
Baker, Mairead (National Health Institute) *
Berg, Dan (University of Washington) *
Blenkiron, Cherie (University of Auckland) *
Blom, Astrid Ambroise Paré Hospital (AP-HP)
Brownell, Isaac (National Institute of Health)
Cook, Mac (University of Washington) *
Doolittle-Amieva, Coley (University of Washington)
Forero, Diaya (FUCS)
Frezza, Michael (EMD Serono)
Garman, Khalid *
Guitera, Pascale (Melanoma Institute Australia)
Hill, Natasha (National Institute of Health)
Horne, Michelle (Merck)
Kasturi, Vijay (EMD Serono)
Kudchadkar, Ragini (Emory University)
Kuhns, Jennifer (EMD Serono)
Lango, Miriam (Fox Chase Cancer Center) *
Lu, Steve (Johns Hopkins School of Medicine)
MacKenzie Ross, Alastair (Guy's and St. Thomas', London, UK) *
Marcazzan, Sabrina *
Miao, Lingling (National Institutes of Health)
Miller, David (BIDMC/MGH, Harvard)
Nagase, Kotaro (Japan)*
Nghiem, Paul (University of Washington)
Park, Song (University of Washington)
Patel, Vishal (George Washington University)
Perlis, Clifford (Abington Hospital Jefferson)
Silk, Anne (DFCI/BWH) *
Sober, Arthur (Massachusetts General Hospital)
Sunshine, Joel (Northwestern University Feinberg School of Medicine)
Takuria, Manisha (DFCI/BWH)
Tarabdkar, Erica (University of Washington)
Tetzlaff, Michael (MD Anderson)
Weiss, Jonathan (BIDMC/Harvard)

Goals of the Merkel cell carcinoma Multi-center Interest Group (MMIG)

- Promote communication and collaborative studies on MCC
- Enhance access to patient data and specimens
- Expand evidence-based care for MCC

The homepage for MMIG is available at:

<https://www.merkelcell.org/about-us/mmig/>

MMIG is funded in part by donations from Merkel cell carcinoma patients.

Please note that in many cases, these summaries reflect unpublished data and are provided to help MMIG members manage their patients and give an overview of what is being done at different centers for care and research.