The University of Wisconsin - Madison

CMP

Cellular and Molecular Pathology

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Program of the Thesis Defense Seminar for the

Degree of Doctor of Philosophy

in Cellular and Molecular Pathology

"Epigenetic regulation of peripheral nerve myelination in development and injury"

Thursday, November 20th, 2014

2:00 p.m.

The John D. Wiley Conference Center

Waisman Center North Tower

Research conducted in the lab of

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Department of Comparative Biosciences

Epigenetic regulation of peripheral nerve myelination
in development and injury

Holly Hung
Under the supervision of
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Holly Hung's Thesis Abstract

Myelination is a vital aspect of central and peripheral nervous system development and function, and impaired myelin function is the causative factor in a number of important diseases, such as Multiple Sclerosis and Charcot-Marie-Tooth Disease (CMT). In the peripheral nervous system, Schwann cells produce and maintain the myelin sheath while retaining the developmental plasticity to dedifferentiate in response to injury, facilitate axon regeneration, and remyelinate after repair. While changes in gene expression for these process have been characterized, prior to this work no studies had examined epigenetic regulation of myelination by chromatin remodeling complexes or epigenetic changes to the chromatin landscape in nerve injury.

My graduate work aims to refine our knowledge of interactions between transcriptional and epigenetic regulatory networks that control peripheral nerve myelination. I investigated the role of the NuRD (Nucleosome Remodeling and Deacetylase) chromatin remodeling complex in peripheral nerve myelination and how found it regulates sites of enhancer activation by Egr2 (Early Growth Response 2), a critical transcription factor for Schwann cell myelination. I also analyzed histone marks which define regions of enhancer activation by ChIP-seq to discover and validate transcriptional regulators of Schwann cell myelination and repair.

Chromodomain helicase DNA-binding protein 4 (Chd4) is the catalytic core of the Nucleosome Remodeling and Deacetylase (NuRD) complex. It associates with Egr2 through interacting NAB (NGFI-A/Egr-binding) corepressors and is widely viewed to facilitate gene repression. This project published the first paper to study regulation of a chromatin remodeling complex in controlling peripheral nerve myelination and found, surprisingly an association of NuRD complex components with Egr2 and NAB on both activated and repressed genes in vivo. Together the results of this study underscored the necessity of Chd4 function to guide proper terminal differentiation of Schwann cells and implicate the NuRD chromatin remodeling complex as a requisite factor in timely and stable peripheral nerve myelination.

Further investigation into the molecular mechanisms of the NuRD complex in Schwann cell myelination showed Chd4 expression is required for proper formation of cis-regulatory binding sites that regulate expression of the myelin

gene, Peripheral myelin protein 22 (Pmp22). This study provides a novel epigenetic mechanism of Pmp22 regulation and suggests Chd4/NuRD is required for Egr2 binding and gene activation.

Finally, I examined myelinating and injury-induced demyelinating Schwann cells for dynamic regulation of histone mark H3K27ac, a chromatin mark which defines active enhancer regulatory sites. This study gives a genome-wide view of epigenetic changes to the chromatin landscape that respond to signaling pathways induced by nerve injury. Importantly, epigenetic annotation and sequence analysis of enhancer regions identified transcription factors previously associated with Schwann cell myelination (Sox10, Egr2) and injury (c-Jun) as well as the novel transcriptional regulators of each condition.

In total, these studies serve to further our knowledge of epigenetic and transcription factor regulation of gene expression networks that define different myelinated glial states in the peripheral nervous system.

Publications

1. **Hung, H.**, Sun, G., Sunduz, K., Svaren, J. "Dynamic Regulation of Schwann cell Enhancers after Peripheral Nerve Injury." (In submission J Biol Chem).

2. Ma, K.H., **Hung, H.,** Srinivasan, R., Xie, H., Orkin, S.H., and Svaren, J.P. "Regulation of peripheral nerve myelin maintenance by gene repression through Polycomb Repressive Complex 2 Polycomb Complex and Schwann cells." (In review, Journal of Neuroscience).

3. Lopez-Anido C., **Hung, H.,** Koenning, M., Srinivasan, R., Sun, G., Emery, B., Keles, S., and Svaren, J.P. "Comparative analysis of Sox10 binding patterns in Schwann cells and Oligodendrocytes" (In submission).

4. Sun, G., Srinivasan, R., Lopez-Anido, C., **Hung, H.,** Svaren, J. Sunduz, K**.** "In silico pooling of ChIP-seq control experiments." PLoS One 7;9(11):e109691 PMCID: 25380244

5. **Hung, H.**, Kohnken, R., Svaren, J. "The Nucleosome Remodeling and Deacetylase Chromatin Remodeling (NuRD) Complex is required for Peripheral Nerve Myelination." J Neurosci. 1; 32(5):1517-27, 2012. PMCID: PMC3292862

6. Mysliwiec, M.R., Carlson, C.D., Tietjen, j., **Hung, H.,** Ansari, A.Z., Lee, Y. "Jarid2 (Jumonji, AT rich interactive domain 2) regulates NOTCH1 expression via histone modification in the developing heart." J Biol Chem. 287, 1235-1241, 2012. PMCID: PMC3256911

Honor & Awards

T32 NIH Training Fellow Cellular and Molecular Pathology Training Grant, (2012 - 2013)
Richard and Jeannette Hoffman Wisconsin Distinguished Graduate Fellowship, (2011 - 2012)
Vilas Conference Presentation Grant, University of Wisconsin, Madison, (2010)

Presentations

Hung, H., Sun, G., Sunduz, K., and Svaren, J.
Epigenetic regulation active enhancers in Schwann cell myelination and demyelinating injury by Egr2 and Sox10. Midwest Chromatin and Epigenetics Meeting, Madison, WI, May 2014.

Hung, H. and Svaren, J.
CHD4/NURD Mediated Regulation of Myelin Maintenance in the Peripheral and Central Nervous Systems. American Society for Neuroscience Conference, Baltimore, MD, March 2012.

Hung, H., Srinivasan, R., Lopez-Anido, C., and Svaren, J
CHD4 is required for Efficient Myelination of the Peripheral Nervous System Chromatin and Transcriptional Mechanisms Group. University of Wisconsin, Madison, WI, March 2012.

Hung, H., Srinivasan, R., Lopez-Anido, C., and Svaren, J.
Schwann cell specific knockout of the NURD chromatin remodeling complex leads to Hypomyelination and Axon Degeneration. American Society for Neuroscience Conference, St. Louis, MO, March 2011.

Holly, H. and Svaren, J.
Chd4 is required for efficient myelination in the PNS
Molecular and Genetic Sciences Group (MGSG) Lecture Series. Madison, WI, March 2011.

Hung, H. and Svaren, J.
CHD4 is required for Efficient Myelination of the Peripheral Nervous System. Cold Spring Harbor Laboratory meeting on Glia in Health and Disease. Cold Spring Harbor, NY, July 2010.

Hung, H. and Svaren, J.
Inactivation of NURD Component CHD4 leads to Hypomyelination and Axon Degeneration in the Peripheral Nervous System. Waisman Center Research Symposium, Madison, WI, March 2010.

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