



Doctoral Dissertation Defense Announcement

“ Obesity in Angiogenesis during Adipose Tissue Remodeling and Regenerative Lung Growth.”



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Cell and Developmental Biology
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Committee in Charge:

Akiko Mammoto, MD. PhD (Mentor)

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Allison Ebert, PhD

Kenichiro Taniguchi, PhD

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Date: Wednesday, August 21, 2024

Time: 10:00 AM (CST)

Defense Location: Bolger Auditorium

Zoom: Contact thunyenyiwa@mcw.edu for zoom link

Graduate Studies:

Foundations of Biomedical Sciences I-IV

Techniques in molecular and cell biology

Professional Development I-II

Writing a Scientific Paper

Writing Individual Fellowship

Reading and Research

Statistics for Basic Sciences

Organ Systems Physiology

Advanced Cell Biology

Developmental and Stem Cell Biology

Research Ethics Discussion Series

Ethics & Integrity in Science

Journal Club

Current Concepts of Cardiovascular Biology

Doctoral Dissertation

Dissertation

“Obesity in Angiogenesis during Adipose Tissue Remodeling and Regenerative Lung Growth.”

Obesity is regarded as the world epidemic affecting more than 1 billion people, with one in every three adults being overweight in the US alone. Obesity is a condition that is mostly caused by high calorie intake compared to metabolized resulting in adipose tissue dysfunction, excess lipid deposition on other organs and homeostatic imbalance. This condition also causes systemic inflammation and delayed healing. Obesity is a disease risk factor to various diseases, especially, metabolic diseases, cardiovascular diseases, and some cancers. Obesity causes endothelial cell (EC) dysfunction due to the proinflammatory state supported by this condition. In this thesis we explore: the effects of obesity on (1) human subcutaneous adipose ECs angiogenic activities and (2) mouse lungs during compensatory regenerative growth.

Adipose, a versatile tissue used for storage of excess nutrients, can expand through adipogenesis and angiogenesis during feast state and release stored energy during starvation, undergoing constant remodeling. However, this remodeling is disrupted under obese conditions. Transcription factor TWIST1 controls angiogenesis and vascular function. We hypothesize that obesity affects the expression of TWIST1 which has negative downstream effects to angiogenic genes, thereby inhibiting angiogenesis in adipose tissue. Here we isolated human adipose ECs of lean vs. obese individuals from human subcutaneous adipose tissues. This study found that the levels of TWIST1 and SLIT2 which controls angiogenesis are lower in endothelial cells (ECs) isolated from obese (BMI >30) compared to those from lean (BMI <30) individuals. We examined the effects of TWIST1 and SLIT2 by altering their gene expressions in both conditions and studying their effects on angiogenic activities and vascular formation using gel implantation system.

Overexpression of TWIST1 restores SLIT2 expression in obese adipose ECs. Angiogenic activities such as EC migration and DNA synthesis are inhibited in obese human subcutaneous adipose ECs compared to lean adipose ECs, while TWIST1 overexpression restores the effects. Obese adipose ECs inhibit blood vessel formation in the gel. Overexpression of TWIST1 in obese ECs restores blood vessel formation in the gel, while SLIT2 knockdown inhibits the effects. These findings suggest that obesity impairs adipose tissue angiogenesis through TWIST1-SLIT2 signaling.

The effects of obesity on lung vascular and alveolar morphogenesis during lung regeneration remain unknown. In this thesis we investigate the effects of obesity on lung regeneration focusing on the contribution of adiponectin- an adipokine, to angiogenesis and lung growth. We utilize unilateral pneumonectomy (PNX) on leptin deficient (*Lep^{ob/ob}*) mice and adiponectin knockout mice and angiogenic activities on human lung ECs to understand the

effects. This study demonstrated that regenerative lung growth and expression of vascular endothelial growth factor VEGF, its receptor VEGFR2 and transcription factor TWIST1 are induced by PNX, while the effects are inhibited in *Lep^{ob/ob}* obese mice and adiponectin knockout mice. The levels of adiponectin, increase in mouse lungs after unilateral PNX, while the effects are attenuated in *Lep^{ob/ob}* obese mice. Adiponectin agonist, AdipoRon stimulates post-PNX lung growth and vascular and alveolar regeneration in *Lep^{ob/ob}* obese mice. Adiponectin stimulates angiogenic activities in lean and obese human lung endothelial cells (ECs), which is inhibited by Twist1 knockdown. These findings suggest that obesity impairs lung vascular and alveolar regeneration.

This thesis revealed potential candidate genes that contribute to the dysregulation of adipose tissue remodeling and lung regeneration under obese conditions. The modulation of these key genes could be potential therapeutic measure to improve adipose remodeling and lung restoration and healing in obese patients.

Tendai Hunyenyiwa
Curriculum Vitae
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Education

- 2019-2024 (expected) Ph.D. Cell and Developmental Biology
Medical College of Wisconsin, WI
- 2017-2019 MS Biology
Andrews University, MI
- 2012-2016 Bachelor of Science Biology
Southwestern Adventist University, TX

Research Experience

- 2019-present **Research Assistant, Medical College of Wisconsin, WI**
Effects of obesity on angiogenesis using human subcutaneous adipose endothelial cells
-Gene manipulation and nucleic acid isolation in endothelial cells
-Migration and Edu assay
-Immunocytochemistry on endothelial cells
Effects of obesity on lung regeneration
-Biomolecule extraction (DNA, RNA, protein, exosomes) from tissues and cells
-Immunohistochemistry on lung tissues
-Next-Generation Sequencing: library preparation and data analysis
- 2017-2019 **Research Assistant, Andrews University, MI**
Effects of novel dihydropyridines on glioblastoma
-Made hybrid dihydropyridines following the Hantzsch reaction
-Used NMR /IR for identification and purity check
-Performed a cell viability test and established LD50
-Maintained and used Glioblastoma cell-line U87
-Performed invasion assay, scratch assay

Fellowship

- 2023- AHA Research Supplement Fellowship to Promote Diversity in Science
(23DIVSUP1068738)

Awards and Honors

- 2023 NAVBO Travel Award
2023 MCW Graduate Student Association Travel Award
2022 ASIP 2022 Trainee Scholar Award
2022 MCW Graduate Student Association Travel Award
2021 NAVBO Vascular Biology 2021 Outstanding Talk Award
2021 MCW Graduate School of Biomedical Sciences Minority Scholarship
2021 ASIP 2021 Trainee Scholar Award

- 2018-2019 Graduate Student Grant in Aid of Research and Proposals Robson Newbold School Grad (Andrews University)
- 2013-2016 Silver Award: Scholarship award presented for maintaining a GPA above 3.5 (Southwestern Adventist University)

Peer Reviewed Publications

- Kyi, P., Hendee, K., **Hunyenyiwa, T.**, Matus, K., Mammoto, T., & Mammoto, A. (2022). Endothelial senescence mediates hypoxia-induced vascular remodeling by modulating PDGFB expression. *Frontiers in medicine*, 9, 908639. <https://doi.org/10.3389/fmed.2022.908639>
- Mammoto, T., **Hunyenyiwa, T.**, Kyi, P., Hendee, K., Matus, K., Rao, S., Lee, S. H., Tabima, D. M., Chesler, N. C., & Mammoto, A. (2022). Hydrostatic Pressure Controls Angiogenesis Through Endothelial YAP1 During Lung Regeneration. *Frontiers in bioengineering and biotechnology*, 10, 823642. <https://doi.org/10.3389/fbioe.2022.823642>
- Hunyenyiwa, T.**, Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Obesity Inhibits Angiogenesis Through TWIST1-SLIT2 Signaling. *Frontiers in cell and developmental biology*, 9, 693410. <https://doi.org/10.3389/fcell.2021.693410>
- Hendee, K., **Hunyenyiwa, T.**, Matus, K., Toledo, M., Mammoto, A., & Mammoto, T. (2021). Twist1 signaling in age-dependent decline in angiogenesis and lung regeneration. *Aging*, 13(6), 7781–7799. <https://doi.org/10.18632/aging.202875>

Manuscripts Submitted

- Hunyenyiwa, T.**, Kyi, P., Johnstone, D., Mammoto, T., & Mammoto, A., (2024). Adiponectin mediates inhibition of angiogenesis and regenerative lung growth in *Lep^{ob/ob}* mice.

Oral Presentations (external)

- Hunyenyiwa, T.**, Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2024). Obesity in Angiogenesis and Regenerative Lung Growth. Presented at NAVBO InFocus - Vascular Metabolism.
- Hunyenyiwa, T.**, Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2023). Obesity in Angiogenesis and Regenerative Lung Growth. Presented at Vascular Biology 2023.
- Hunyenyiwa, T.**, Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). Obesity in lung vascular and alveolar regeneration. Presented at Pathology for Investigators, Students, and Academicians (PISA) 2022.
- Hunyenyiwa, T.**, Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Obesity inhibits angiogenesis through TWIST1-SLIT2 signaling. Presented at the Adipose Biology Seminar Series.
- Hunyenyiwa, T.**, Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Obesity inhibits angiogenesis through TWIST1-SLIT2 signaling. Presented at Vascular Biology 2021.
- Hunyenyiwa, T.**, Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). Obesity Inhibits Angiogenesis through TWIST1-SLIT2 Signaling.

Presented at Pathology for Investigators, Students, and Academicians (PISA) 2021.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). ROBO/SLIT in Obesity-dependent Changes in Angiogenesis and Lung Regeneration. Presented at Experimental Biology 2021.

Hunyenyiwa, T., Smith, D., & Murray, D. (2018). A-2 The Effects of Novel Dihydropyridine Derivatives as Anti-invasive Agents Against Glioblastoma. Presented at the Michigan Academy of Science, Arts, & Letters.

Oral Presentations (MCW)

Hunyenyiwa, T., Hendee, K., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). The effects of obesity on adipose tissue remodeling. Presented at the Cell Biology, Neurobiology & Anatomy Seminar Series.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). Obesity in angiogenesis and lung regeneration. Presented at the Developmental Biology Club.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). Obesity in angiogenesis and lung regeneration. Presented at the CVC ATVB Signature Program Meeting.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). Obesity in angiogenesis and lung regeneration. Presented at the Developmental Biology Club.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (Year). Obesity in angiogenesis and lung regeneration. Presented at the CVC ATVB Signature Program Meeting.

Poster Presentations (external)

Hunyenyiwa, T., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2023). Obesity in Angiogenesis and Regenerative Lung Growth. Presented at Cell Biology 2023.

Hunyenyiwa, T., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). Effects of obesity on angiogenesis and regenerative lung growth. Presented at the 22nd International Vascular Biology Meeting.

Hunyenyiwa, T., Hendee, K., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). Obesity in angiogenesis and lung regeneration. Presented at Experimental Biology 2022.

Hunyenyiwa, T., Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Obesity inhibits angiogenesis through TWIST1-SLIT2 signaling. Presented at Vascular Biology 2021.

Hunyenyiwa, T., Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). TWIST1-SLIT2 signaling in angiogenesis in obese adipose tissue. Presented at Vasculata 2021.

Poster Presentations (MCW)

Hunyenyiwa, T., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2023). Effects of obesity in angiogenesis and regenerative lung growth. Presented at the CVC Research Retreat.

Hunyenyiwa, T., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). Effects of obesity in angiogenesis and regenerative lung growth. Presented at the 32nd Annual Graduate School Research Poster Session.

Hunyenyiwa, T., Kyi, P., Matus, K., Mammoto, T., & Mammoto, A. (2022). Effects of obesity in angiogenesis and regenerative lung growth. Presented at the CVC Research Retreat.

Hunyenyiwa, T., Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Obesity inhibits angiogenesis through TWIST1-SLIT2 signaling. Presented at the 31st Annual Graduate School Research Poster Session.

Hunyenyiwa, T., Hendee, K., Matus, K., Kyi, P., Mammoto, T., & Mammoto, A. (2021). Inhibition of angiogenesis in obese adipose tissue through TWIST1-SLIT2 signaling. Presented at the CVC Research Retreat.

Hunyenyiwa, T., Hendee, K., Matus, K., Mammoto, T., & Mammoto, A. (2021). Obesity in vascular and alveolar morphogenesis after pneumonectomy. Presented at the 30th Annual Graduate School Research Poster Session.

Positions and Employment

2017-2019	Head Biology Teaching Assistant, Andrews University, Berrien Springs MI
2016-2017	Science Teacher, Ketchum Adventist Academy, Ketchum, OK

Professional memberships

2023 -	American Association for Cell Biology; Trainee Member
2022 -	American Heart Association; Trainee Member
2020 -	North American Vascular Biology Organization (NAVBO); Trainee Member
2020 -	American Society for Investigative Pathology (ASIP); Trainee Member

Leadership Positions

2023	Organizer, Vascular Biology 2023 Pre-conference for Trainees
2021-	IDP International Student Committee, Medical College of Wisconsin
2021	Session Co-Chair (Moderator), PISA 2021
2015-2016	Student Senate, Southwestern Adventist University
2014-2015	Biology Club President, Southwestern Adventist University