THE UNIVERSITY of ADELAIDE

MicroRNA MEDIATED GENE REGULATION IN CANCER

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This thesis is submitted to the University of Adelaide in the fulfilment of the requirements for the degree of

Doctor of Philosophy

August 2017

Table of contents

Overview

Cancer, as the second cause of death worldwide, requires further understanding of its mechanism to improve patient survival and quality of life. MicroRNAs are important regulators of translation and play major roles in normal cellular functions as well as cancer pathobiology. The broad aim of my study was to provide new insight into microRNA-mediated gene regulation in cancer. Although miRNAs account for the post-transcriptional regulation of more than 60% of human protein-coding genes, this work is mainly focused on two molecular pathways: the p53 signalling pathway and the androgen receptor signalling pathway, each of which have been shown to be strongly connected to tumorigenesis.

p53, a transcription factor that participates in multiple cellular functions, is considered the most important tumour suppressor and is mutated in ~50% of cancers. Previous evidence suggests that post-transcriptional deregulation of p53 by microRNAs contributes to tumorigenesis, tumour progression and therapeutic resistance. We found that the microRNA miR-766 was aberrantly expressed in breast cancer, and that over-expression of miR-766 caused accumulation of wild-type p53 protein in multiple cancer cell lines. Supporting its role in the p53 signalling pathway, miR-766 decreased cell proliferation and colony formation in several cancer cell lines, and cell cycle analyses revealed that miR-766 causes G2 arrest. At a mechanistic level, we demonstrate that miR-766 enhances p53 signalling by directly targeting MDM4, an oncogene and negative regulator of p53. Analysis of clinical genomic data from multiple cancer types supports the relevance of miR-766 in p53 signalling. Collectively, our study demonstrates that miR-766 can function as a novel tumour suppressor by enhancing p53 signalling.

Moreover, we have reported miR-9 as a novel miRNA that specifically down-regulates the expression of misssense p53 R248Q and R273H in multiple cell lines, while the wild-type p53 is upregulated and other p53 mutations are unaffected. We also identified a potential binding site within *TP53* ORF. A few potential mechanisms behind this unique observation are discussed. This part of my work provides novel evidence in the miRNA-mediated mutant p53 regulation and discusses the weakness of current miRNA target study.

Androgen receptor (AR) is a transcription factor that is the key driver of prostate cancer growth and progression. As such, AR and its downstream pathways are a critical target for prostate cancer treatment. MiRNAs participate in the regulation of these pathways by targeting AR itself or downstream genes. In our study, we identified miR-375 as a direct negative regulator of androgen receptor and its signalling pathways. Over-expression of miR-375 results in down-regulation of AR protein and mRNA levels and AR target genes *FKBP51* and *KLK3*, accompanied by growth inhibition of prostate cancer cells. Over-expression of AR rescued the effect of miR-375 over expression. Interestingly, AR binds the promoter region of the MIR375 gene and upregulates its expression. Thus, my work identifies a new feedback loop that balances the endogenous level of AR and miR-375 in prostate cells.

Overall, this work provides further understanding of how miRNAs regulate important gene pathways in different cancers.

Publications

Original research paper:

- Wang Q, Selth LA, and Callen DF. *MiR-766 induces p53 accumulation and G2/M arrest by directly targeting MDM4.* Oncotarget, 2017. 8(18): p. 29914-29924.
- Wang Q, Townley S, Paltoglou S, Callen DF, and Selth LA. *A novel miR-375-androgen receptor feedback loop regulates growth of prostate cancer cells.* Text in manuscript.

Review paper:

- Wang Q, Callen DF, and Selth LA. *The microRNA-p53 network: a game of balance.* Text in manuscript.

Presentation:

- Presentation: Wang Q, Callen DF. *Over-expression of miR-766 up-regulates p53 and represses cell proliferation by reducing MDM4.* Adelaide RNA Special Interest Group seminar, 31 October 2014, Adelaide, Australia
- Poster: Wang Q, Callen DF. *Over-expression of miR-766 up-regulates p53 and represses cell proliferation by reducing MDM4.* Florey International Postgraduate Research Conference, 25 September 2014, Adelaide, Australia.

- Poster: Wang Q, Callen DF. *Over-expression of miR-766 up-regulates p53 and represses cell proliferation by reducing MDM4.* 27th Lorne Cancer Conference 2015, 12-14 February 2015, Lorne, Australia.
- Poster: Wang Q, Selth LA, and Callen DF. *MiR-766 induces p53 accumulation and G2/M arrest by directly targeting MDM4.* Florey International Postgraduate Research Conference, 24 September 2015, Adelaide, Australia.

Acknowledgement

I would first like to express my sincere gratitude to my principal supervisor Prof. David Frederick Callen for his kind support and patient guidance throughout the past four years since the first day that I arrived in Australia in 2013. Prof. David Callen not only gave me this great opportunity to conduct my PhD at the University of Adelaide; he also generously provided me with a job in the lab as a research assistant even prior to starting my PhD candidature. During the years working under Prof. David Callen's supervision, his unsurpassed knowledge in cancer biology and research experience always broadened my horizon in the field and improved the quality and efficiency of my research. He also helped me improve my academic writing with extraordinary patience. I especially appreciated Prof. David Callen's open attitude towards unexpected small findings, which inspires me to remain curious while exploring the unknown field. Prof. David Callen encouraged me to embrace emerging new technologies in cancer genetics research, and supported me for the 2015 Winter School in Mathematical & Computational Biology at the University of Queensland, and the RStudio workshop at the University of Adelaide; both events opened a new direction in my research career.

I am also extremely grateful to Dr. Luke Selth for being my co-supervisor and supporting me through the second half of my PhD when my previous project was facing difficulty. Dr. Luke Selth is a very talented scientist with remarkable enthusiasm and immense knowledge in the non-coding RNA field. With the deep understanding of the molecular mechanisms, his insightful and enlightening comments helped me plan, scale and develop my project more efficiently. I enjoyed the discussions with Dr. Luke Selth that strengthened my precise logic in science, which will be essential for my future

research. Dr. Luke Selth introduced me to the field of bioinformatics, and helped me to develop data digging, meta-analysis and programming techniques. I also benefited from his skill in comprehensive data interpretation and precise scientific writing.

I am indebted to Dr. Paul Matthew Neilson, Dr. Kathleen Irene Pishas and Dr. Andrew Turner, who have worked in the Personalized Cancer Therapeutics Laboratory (PCTL) as postdoctoral fellows and have given me great advice and help during my study. Dr. Paul Neilson was co-supervising me when I first started working in the group, and I can never thank him enough for the inspiring and passionate discussions that we had in the beginning of my project. He greatly extended my understanding in non-coding RNA structure and function. Dr. Paul Neilson had the ability to influence people with his enthusiasm and faith when facing any challenges. Without his support, I would have had great difficulty in gaining confidence to begin independent research. Dr. Kathleen Pishas was my co-supervisor for a short period, and supported my work with thoughtful comments and suggestions. Dr. Andrew Turner joined PCTL in my third year of PhD, and managed the general lab issues as well as helped me with experiment design and material preparation. In particular, I also give my thanks to Dr. Andrew Turner for proofreading the paper and thesis via his editing business (according to the ASEP standards).

During my PhD candidature, I was lucky to work with a group of friendly and passionate young scientist. Many thanks goes to my good friends and colleagues Dr. Yu Feng, Dr. Alaknanda Adwal, Dr. Sheng Lei and Dr. Reshma Shakya, who are always will to help or discuss any issue that we are interested in. Their friendship fulfilled my

journey of PhD. Dr. Yu Feng and I both worded in the non-coding RNA area. Her solid knowledge in miRNA isoforms and RNA structure enhanced my confidence in the miR-9 research project. Dr. Alaknanda Adwal was warm-hearted and provided generous assistance in flow cytometry experiments.

In addition, I would like to thank Prof. Wayne Tilley for kindly giving me access to the Dame Roma Mitchell Cancer Research Laboratory (DRMCRL) and providing various resources for my work. While working in DRMCRL, I could never thank lab manager Adrienne Hanson enough for the lenti-virus training and her assistance in every aspect of work. My thanks also goes to the researchers and lab management staffs in DRMCRL. Dr. Rajdeep Das and Dr. Gerard Tarulli both helped me with reverse transwell invasion experiment and the confocal microscope training. Scott Townley and Steve Paltoglou both helped with the preparation of ChIP experiment. I would also like to extend my appreciation to research manager Suzanne Atkins for her important emotional support.

I believe it would not have been possible to complete my PhD without the Research Training Program Scholarship (formerly International Postgraduate Research Scholarship and Australian Post-graduate Award, which was generously granted in 2014 by the Australian government; this significantly eased financial pressures and allowed me to focus on my studies.

My heartfelt thanks goes to my parents. Although far away in a different country, they have always been unconditionally supporting all my decisions and goals with endless love, well wishes and encouragement. Finally, I would like to convey my deepest gratitude to my partner, Travis Roe, who has been my most trustworthy supporter, who has been the comfort in the hard times and the joy in the good days, and always endowed me with understanding and love.

Declaration

I, Qingqing Wang, certify that this work contains no material which has been accepted for the award of any other degree or diploma in my name, in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

In addition, I certify that no part of this work will, in the future, be used in a submission in my name, for any other degree or diploma in any university or other tertiary institution without the prior approval of the University of Adelaide and where applicable, any partner institution responsible for the joint-award of this degree.

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I acknowledge the support I have received for my research through the provision of an Australian Government Research Training Program Scholarship.

Qingqing Wang

27 August 2017