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Ph.D. Thesis

**Functional analysis of the nuclear receptor NHR-25
during *C. elegans* development**



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České Budějovice, 2009

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Cover image: Movement series of a *C. elegans* male that carries *rol-6(su1006)* mutation.

Hajdušková, M., 2009: Functional analysis of the nuclear receptor NHR-25 during *C. elegans* development. Ph.D. Thesis, in English. - 117 p., Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic.

Annotation:

This thesis introduces a study of a conserved nuclear receptor NHR-25 and of its role during development of the nematode *Caenorhabditis elegans*. To address the function of NHR-25 in certain developmental processes, several approaches of genetic, cellular and molecular biology have been used. This work reveals an indispensable role of NHR-25 in asymmetric cell division and cell fate decision of the epidermal stem cells, seam cells. The results represent one of the first direct implications of any nuclear receptor being required for cell fate determination. It demonstrates a genetic *in vivo* interaction between NHR-25 and the Wnt/ β -catenin asymmetry pathway, which is essential for proper seam cell differentiation. Remarkably, NHR-25 is shown to modulate Wnt signaling in either synergistic or antagonistic manner, depending on cellular context. This finding provides a novel insight to the crosstalk between nuclear receptors and Wnt signaling. Moreover, it sheds light on our understanding how interacting signaling pathways control differentiation of multiple tissues in multicellular organisms.

Financial support:

This work was supported by grants 204/07/0948 and 204/09/H058 from the Czech Science Foundation, by the project 2B06129 from the Czech Ministry of Education, by the grant 524/03/H133 from the Grant Agency of the Czech Republic, and Z60220518 from the Institute of Parasitology. I was also supported by grant 50/2006/P-BF from the University of South Bohemia Grant Agency.

Declaration:

I hereby declare that I did all the work, presented in this thesis, by myself or in collaboration with the co-authors of the published article.

Further, I declare that in accordance with the Czech legal code § 47b law No. 111/1998 in its valid version, I consent to the publication of my Ph.D. thesis (in an edition made by removing marked parts archived by the Faculty of Science) in an electronic way in the public access to the STAG database run by the University of South Bohemia in České Budějovice on its web pages.

Author contributions

As the first author of the paper accepted by *Journal of Cell Science* (see chapter “Research article”), I designed many experiments and did the vast majority of the bench work. This included generation and phenotypic analyses of transgenic *C. elegans* strains for tissue-specific *nhr-25* RNAi, phenotypic analyses of the male tail morphology, and examination of T cell polarity defects by means of direct lineage analysis of the T cell progeny and by using dye-filling techniques in *nhr-25* mutant and RNAi worms. I discovered a synergistic genetic interaction of NHR-25 with the Wnt/ β -catenin asymmetry pathway and with the parallel RUNX signaling during the T cell differentiation. I evaluated the data and interpreted them in a complete manuscript draft that I wrote by myself, including preparation of the figures. Dr. M. Asahina, besides designing and supervising the project, contributed experiments showing ectopic expression of the Hox gene *mab-5* in *nhr-25* mutant male tails. Prof. M. Herman taught me how to perform the lineaging and dye-filling experiments in his lab. Dr. M. Jindra served as a scientific advisor and helped with writing of the paper. I also did most of the work described in "Unpublished preliminary results" except where stated otherwise directly in the thesis chapter.

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České Budějovice, June 16, 2009

Acknowledgments

I would like to thank my supervisor, Dr. Masako Asahina-Jindrová, for giving me the opportunity to work in her laboratory and for teaching me worm manipulation and genetic methods in *Caenorhabditis elegans*. I thank both of my supervisors, Drs. Masako Asahina-Jindrová and Marek Jindra, for scientific guidance during my studies and for the arrangement of my stay in the USA, which had been for me an immensely motivating research experience. I am very grateful for help during preparation of the manuscript and of this thesis, for useful discussion, and to Dr. Marek Jindra for tutelage on how to write and present scientific work and for advice on future scientific career. I thank Prof. Michael A. Herman (Kansas State University, Manhattan, USA) for the opportunity to work in his lab, for teaching me cell lineage analysis and for the nice time I have spent in Kansas. I appreciate the scientific and financial support of my research, provided by Prof. Julius Lukeš and Prof. Tomáš Scholz of the Institute of Parasitology.

I thank our lab assistant Aida Trojanová for service and all members of the Jindra and Asahina laboratories for a friendly atmosphere, psychological support and helpfulness, whenever I needed.

My special thanks go to my husband Ondra for inspiring suggestions and discussions, encouragement, never-ending patience, tolerance and love. Last but not least, I would like to thank my parents for their support during my studies and all my family and friends for remarkable comprehension.

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Abstract

Asymmetric cell divisions produce new cell types during animal development. Studies in *Caenorhabditis elegans* have identified major signal transduction pathways that determine the polarity of cell divisions. How these relatively few conserved pathways interact and what modulates them to ensure the diversity of multiple tissue types is an open question. The Wnt/ β -catenin asymmetry pathway governs polarity of the epidermal cell T in the *C. elegans* tail. Here, we show that the asymmetry of T cell division and morphogenesis of the male sensory rays require NHR-25, an evolutionarily conserved nuclear receptor. NHR-25 ensures the neural fate of the T cell descendants in cooperation with the Wnt/ β -catenin asymmetry pathway. Loss of NHR-25 enhances the impact of mutated nuclear effectors of this pathway, POP-1/TCF and SYS-1/ β -catenin, on the T cell polarity, while it suppresses the effect of the same mutations on asymmetric division of the somatic gonad precursor cells. Therefore, NHR-25 can either synergize with, or antagonize the Wnt/ β -catenin asymmetry pathway depending on the tissue context. Our findings define NHR-25 as a versatile modulator of Wnt/ β -catenin-dependent cell fate decisions.