Dissertation Kristina Althoff - Abstract

Neuroblastoma is a common solid tumor of infancy and accounts for more than 15% of pediatric cancer deaths. Amplification of *MYCN* oncogene defines the most aggressive subtype of neuroblastoma with poor prognosis. In the present study, a new Cre-conditional *MYCN*-driven transgenic mouse model was generated to analyse tumorigenesis *in vivo*. When targeting *MYCN* expression to the neural crest in transgenic LSL-*MYCN*;*Dbh-i*Cre mice, palpable tumors developed, which arose predominantly from both adrenals, but also from superior cervical ganglia and *ganglion coeliacum*. LSL-*MYCN*;*Dbh-i*Cre-induced tumors consisted of small round blue cells and possessed neurosecretory vesicles. Expression of human *MYCN* and specific neuroblastoma marker genes was high in tumors. Furthermore, murine neuroblastomas recapitulated genomic aberrations and *MYCN* mRNA as well as microRNA (miRNA) signatures described for human neuroblastoma. MiRNAs are small noncoding RNA molecules that bind target mRNAs resulting in translational repression or degradation of the mRNA. In cancer, they can act as tumor suppressors or oncogenes. In human primary neuroblastomas, low expression of miR-542-p and miR-137 was highly correlated with unfavourable prognosis and miR-542-3p was upregulated in murine LSL-*MYCN*;*Dbh-i*Cre tumors. Exogenous expression of these miRNAs in established neuroblastoma cell lines reduced viability and proliferation, increased apoptosis and induced cell cycle arrest. Furthermore, direct interaction between the predicted targets *survivin* (miR-542-3p) and *KDM1A* (miR-137) was validated by luciferase reporter assays and regulation was demonstrated on mRNA as well as protein level. Ectopic expression of miR-542-3p or miR-137 phenocopied the knockdown of respective targets. In addition, we were able to validate a direct interaction between miRNAs and their targets by performing rescue experiments. Treatment of mice bearing neuroblastoma xenografts with miR-542-3p-loaded nanoparticles resulted in downregulation of survivin and induced reduced tumor cell proliferation as well as increased apoptosis.

In conclusion, the novel LSL-*MYCN*;*Dbh-i*Cre neuroblastoma mouse model contribute to a better understanding of *MYCN*-driven neuroblastoma and to identification of relevant genes and miRNAs. Combination with other transgenic models, regulating potential relevant genes, could provide new insights into neuroblastoma tumorigenesis. Restoring function of tumor suppressive miRNAs, such as miR-542-3p or miR-137, could represent a feasible approach to reduce neuroblastoma aggressiveness.