ABSTRACT

Title: Role of EYA1 in neuroblastoma cell proliferation

Background:
Neuroblastoma is a common pediatric cancer which is responsible for about 15% of childhood cancer-related deaths. It arises during early stages of development from neural crest cells of the sympathetic nervous system. Recent work has shown high risk neuroblastoma (40-50% five-year survival rate) to be associated with overexpression and amplification (>10 copies) of the MYCN gene, which belongs to the MYC family of transcription factors. The EYA (eyes absent) family of genes (originally identified in the fruit fly, Drosophila) includes transcriptional co-activators which control cell growth and survival, thus playing an important role in the development of organs. The aberrant activity of the human homologs of EYA (EYA1-4) has been linked to a variety of cancers. In particular, high levels of nuclear EYA1 have been associated with high-risk neuroblastoma and with high levels of nuclear MYCN. EYA1 is also overexpressed in breast cancer, and medulloblastoma, while its phosphatase activity is essential for breast cancer cell proliferation. As such, a better understanding of the role of EYA1 and its phosphatase activity in neuroblastoma is important for developing more targeted treatment methods.

Objective:
Examine the role of EYA1 in neuroblastoma cell proliferation with EYA1 knockdown neuroblastoma cell lines (genetic inhibition of EYA1). Study the sensitivity of various MYCN amplified and MYCN non-amplified cell lines to two drugs (benzarone, and MLS60) which are known to inhibit EYA phosphatase activity (pharmacological inhibition).

Results:
Reduced cell proliferation was observed in two EYA1 knockdown neuroblastoma cell lines (KELLY, with amplified MYCN; and SK-N-AS, with non-amplified MYCN). Six neuroblastoma cell lines (KELLY, SK-N-BE(2)C, and LAN-5, with amplified MYCN; and SK-N-AS, SH-SY5Y, and SH-EP1, with non-amplified MYCN) were all sensitive to both drugs.

Conclusion:
The notable reduction in cell proliferation in the EYA1 knockdown neuroblastoma cell lines can be followed up with proliferation experiments involving more cell lines, as well as cell cycle experiments to determine whether inhibition of EYA1 leads to reduced cell proliferation or to increased cell death (both of which are consistent with our current observations). Further rescue experiments will continue to examine this possibility. While all the tested neuroblastoma cell lines are sensitive to both drugs, further biochemical inhibition experiments with purified proteins of the EYA family will shed light onto the specificity of these drugs. Ongoing experiments will also determine the drug sensitivity of non-neuroblastoma cell lines. Pharmacological inhibition of EYA1 may eventually lead to therapeutic methods to target neuroblastoma.