Differentiation of Neuroblastoma Cells: Can MicroRNAs be Used as a Therapeutic? A Systematic Review

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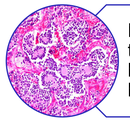
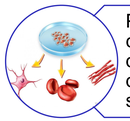
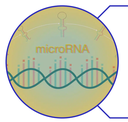
Introduction



Neuroblastoma (NB) is a pediatric solid tumor with remarkable genetic heterogeneity and poor outcomes in high-risk disease



Poorly differentiated neuroblastoma cells have a worse prognosis than differentiated cells, therefore, differentiation induction is a treatment strategy in neuroblastoma



MicroRNAs (miRs) are small RNAs that control post-transcriptional gene regulation

aim

Study Aim: To determine which miRs promote NB cell differentiation and through what mechanisms

Design/Sample

A systematic literature search of Ovid MEDLINE and Embase (Elsevier) was performed using search terms focused on the concepts of microRNA, neuroblastoma, and cell differentiation.

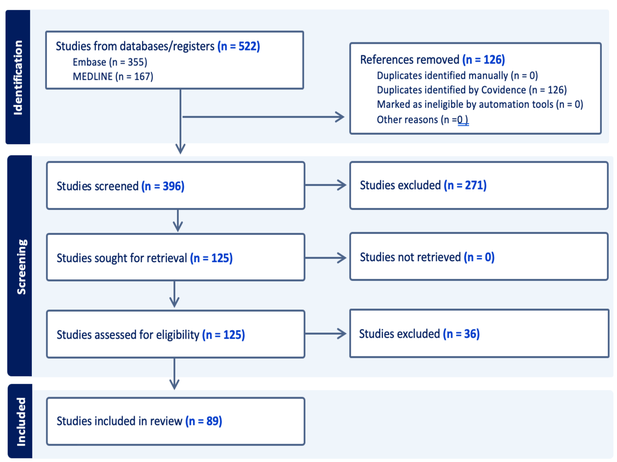


Figure 1: PRISMA flow chart for this review

Results

Table 1: Common miRs impacting NB maturation & differentiation

MicroRNAs	Molecular Targets
miR-10a/b	NCOR2
miR-18-20	estrogen receptor alpha, RUNX1, RUNX3, TrkA and p75
miR-23-27	Brn-3b, Notch1
miR-34	BCL2, MYCN, LYAR, CTCF, E2F3, N-myc, C-myc, Survivin, Cyclin D3, Vimentin, Notch1
miR-124	ELF4, RARG, STAT3, CDK4, EZH2, N-myc, MYB, PPARG, TCF3, TCF12, CDC42, PTBP1, PTBP2, MAP2, PLEC, ITGB1
miR-125b	PHOX2B, BAX and BCL-2
miR-128	MYB, PPARG, TCF3, TCF12, CDC42, NTRK3, BCL2
miR-129a	Notch1
miR-130a-3p	MeCP2
miR-137	MYB, PPARG, TCF3, TCF12, CDC42
miR-142-3p	Stau1
miR-143-3p	NOTCH-1 and NOTCH-2
miR-221	PTEN
miR-335	HAND1 and JAG1
miR-376a	NFKB, NFAT
miR-494	HO-1, HOXD9, HOXD10 and PHOX2B
miR-495	HOXD9, HOXD10 and PHOX2B
miR-506-3p	PLAGL2, MYCN, CDK4, STAT3
miR-543	HOXD9, HOXD10 and PHOX2B
miR-939	NFKB, NFAT
miR-2110	TSKU protein
miR-3613-3p	MCPIP1 (ZC3H12A) gene

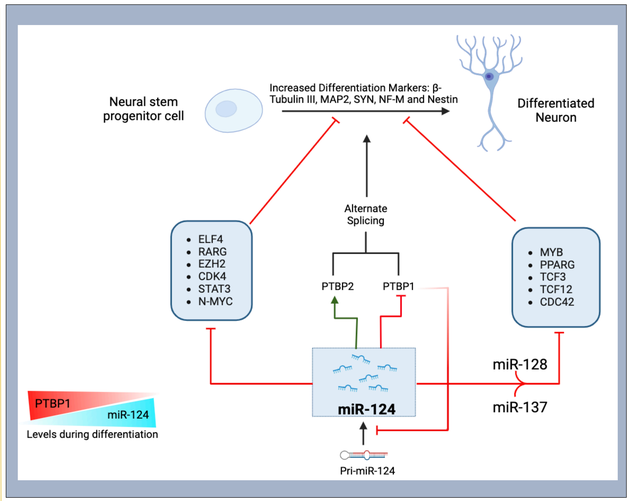


Figure 2: In vivo and in vitro miR-124 expression early in development is low due to the inhibition by PTBP1, but as the PTBP1 levels decline, mature miR-124 is expressed that inhibits PTBP1 and induces PTBP2 to result in a particular alternative splicing to causes differentiation. MiR-124 then targets many molecular targets alone and in combination with other miRs to induce differentiation and limit the proliferation of NB cells.

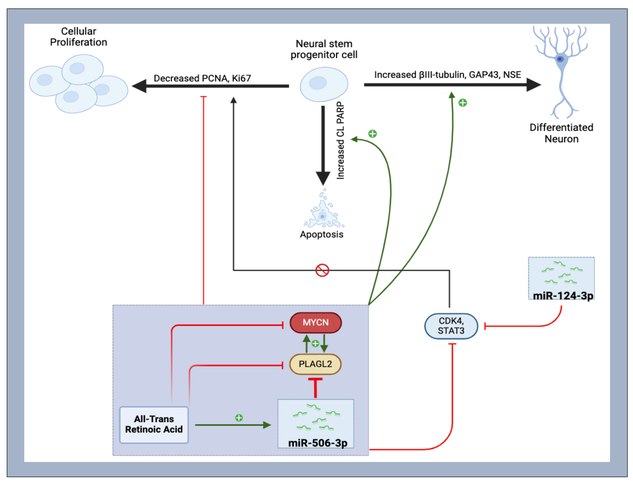


Figure 3: miR-506-3p is a very potent differentiation inducer of NB cells as it inhibits PLAGL2, the inducer of MYCN expression. This action alone limits cellular proliferation and also induces apoptosis and differentiation of neuroblasts. miR-506-3p also joins with miR-124-3p to inhibit CDK4 and STAT3 which limits the NB cell proliferation.

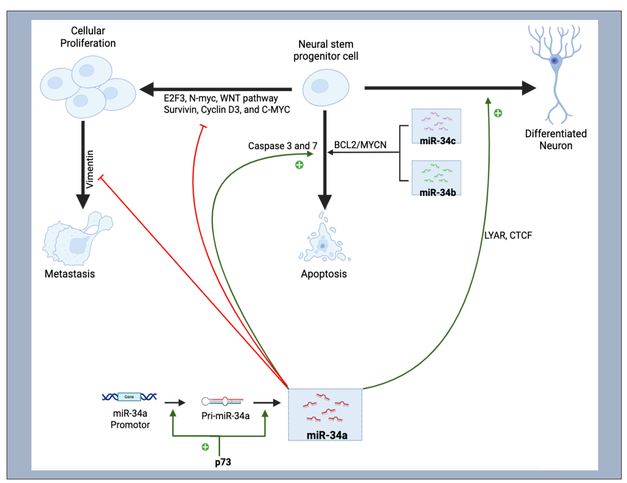


Figure 4: The miR-34 family is considered a tumor suppressor as an increase in p73 (part of the p53 family) increases it levels. Mir-34a upregulates caspase 3 and 7 to induce apoptosis, LYAR, and CTCF to induce differentiation and inhibition of many factors such as N-myc to limit proliferation. Mir-34b/c also show tumor-suppressing activity by modulating MCYN and BCL2.

Discussion

Key differentiation miR mechanisms of action:

- Expression of miR-124 can induce differentiation of neurons *in vitro* and *in vivo*
- MiR-34a has been implicated in neuronal differentiation, synaptogenesis, and cell proliferation.
- miR-506-3p is a potent inducer of differentiation and causes significant neurite outgrowth and increased differentiation factors

Clinical Correlation:

- miR-124-3p expression is significantly lower in high-risk stage 4 tumors, and low expression of miR-124-3p is associated with worse overall survival
- miR-34a is expressed at lower levels in unfavorable primary NB tumors and cell lines
- miR-506-3p, while normally expressed in differentiated neurons is completely absent from NB tissue samples

Conclusions/Next Steps

Multiple microRNAs have been shown to regulate differentiation of NB cells and warrant further investigation of therapeutic potential

- In vivo testing of differentiation-promoting miRs
- Combination miR therapy with chemotherapy
- Novel *in vivo* delivery methods for miR therapy

Acknowledgments

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