Evaluating the Prognostic Significance of Circulating mRNAs in Children with Refractory or Relapsed Neuroblastoma (RR-NBL); a BEACON-Neuroblastoma Biomarker Study

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Introduction/Background: Despite therapeutic advances, high-risk neuroblastoma remains clinically challenging, with a 5-year survival below 50%. For relapsed/refractory (RR) cases, overall survival (OS) drops to <20%. Previously, we showed that detecting the adrenergic (ADR) neuroblastoma mRNAs, PHOX2B (paired-like homeobox 2B) and TH (tyrosine hydroxylase), in blood predicts prognosis (1). Given that neuroblastoma comprises both ADR and more mesenchymal-like (MES) cells, which influence progression and relapse (2,3), we analysed the expression of ADR and a panel of MES mRNAs in RR-NBL patients from the BEACON-Neuroblastoma trial (NCT02308527).

Aims: Early risk identification could improve outcomes by guiding treatment modifications. Using ADR markers we predict outcomes in 80% of high-risk patients using simple blood samples. We hypothesis that by including MES-marker expression prognostic information for the remaining 20 % of patient could be obtained.

Methods: RNA was extracted from 2ml blood samples collected in PAXgene™ tubes at trial entry (n=74) and at cycles 2 (n=42), 4 (n=31), 6 (n=24), and end of treatment (n=25) (4). Panels of ADR and MES mRNAs were quantified by RT-qPCR using TaqMan Low Density Arrays (TLDA). Euclidean clustering generated heat maps identifying ADR and MES gene signatures.

Results: Baseline detection of TH, PHOX2B, or both correlated with reduced progression-free survival (PFS) and OS, with combined detection showing stronger associations (PFS HR 2.68; OS HR 2.84). Their presence post-cycle 2 and 6 correlated with progression and poor OS. Clustering of the TLDA data revealed an ADR signature at baseline, which intensified at treatment end, supporting its role in relapse. MES mRNA profiling further stratified patients, offering potential new biomarkers of risk.

Conclusion/Implications: Blood-based detection of TH and PHOX2B at trial entry and treatment end identifies RR-NBL patients at highest risk. MES mRNAs could enhance risk stratification. This simple test could help prioritise children for alternative therapies.

References:

1. Viprey VF, Gregory WM, Corrias MV, Tchirkov A, Swerts K, Vicha A, Dallorso S, Brock P, Luksch R, Valteau-Couanet D, Papadakis V, Laureys G, Pearson AD, Ladenstein R, Burchill SA. Neuroblastoma mRNAs predict outcome in children with stage 4 neuroblastoma: a European HR-NBL1/SIOPEN study. J Clin Oncol. 2014 Apr 1;32(10):1074-83. doi: 10.1200/JCO.2013.53.3604. Epub 2014 Mar 3. PMID: 24590653.

2. van Groningen T, Koster J, Valentijn LJ, Zwijnenburg DA, Akogul N, Hasselt NE, Broekmans M, Haneveld F, Nowakowska NE, Bras J, van Noesel CJM, Jongejan A, van Kampen AH, Koster L, Baas F, van Dijk-Kerkhoven L, Huizer-Smit M, Lecca MC, Chan A, Lakeman A, Molenaar P, Volckmann R, Westerhout EM, Hamdi M, van Sluis PG, Ebus ME, Molenaar JJ, Tytgat GA, Westerman BA, van Nes J, Versteeg R. Neuroblastoma is composed of two super-enhancer-associated differentiation states. Nat Genet. 2017 Aug;49(8):1261-1266. doi: 10.1038/ng.3899. Epub 2017 Jun 26. PMID: 28650485.

3. Thirant C, Peltier A, Durand S, Kramdi A, Louis-Brennetot C, Pierre-Eugène C, Gautier M, Costa A, Grelier A, Zaïdi S, Gruel N, Jimenez I, Lapouble E, Pierron G, Sitbon D, Brisse HJ, Gauthier A, Fréneaux P, Grossetête S, Baudrin LG, Raynal V, Baulande S, Bellini A, Bhalshankar J, Carcaboso AM, Geoerger B, Rohrer H, Surdez D, Boeva V, Schleiermacher G, Delattre O, Janoueix-Lerosey I. Reversible transitions between noradrenergic and mesenchymal tumor identities define cell plasticity in neuroblastoma. Nat Commun. 2023 May 4;14(1):2575. doi: 10.1038/s41467-023-38239-5. PMID: 37142597; PMCID: PMC10160107.

4. Viprey VF, Corrias MV, Kagedal B, Oltra S, Swerts K, Vicha A, Ladenstein R, Burchill SA. Standardisation of operating procedures for the detection of minimal disease by QRT-PCR in children with neuroblastoma: quality assurance on behalf of SIOPEN-R-NET. Eur J Cancer. 2007 Jan;43(2):341-50. doi: 10.1016/j.ejca.2006.08.007. Epub 2006 Oct 4. PMID: 17023157.