Uncovering stem cell signaling pathways disrupted in Tønne- Kalscheuer Syndrome

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Protein ubiquitylation controls virtually all aspects of eukaryotic biology. A key outstanding question in the field is the role of the ubiquitin system in disease. This is particularly true for neurodevelopmental disorders with intellectual disability (ND/ID) which are a heterogenous group of diseases driving lifelong deficits in cognition and behaviour of affected individuals and have no cure. Tonne-Kalscheuer syndrome (TOKAS) is one such ND/ID which is caused by missense variants in the RLIM gene which encodes the E3 ubiquitin ligase RNF12. However, the underlying mechanisms involved in TOKAS pathogenesis are poorly understood.

Using a pluripotent stem cell model, we find that TOKAS pathogenic variants disrupt RNF12 E3 ubiquitin ligase activity resulting in a failure to efficiently degrade RNF12 substrates, causing abnormal neural differentiation that likely underpins TOKAS aetiology. Furthermore, our research identifies a novel signaling network involving SRPK induced RNF12 phosphorylation to control RNF12 E3 ligase activity and transcription of neural genes in pluripotent cells. However, the relevant substrate(s) and molecular targets involved in TOKAS remain to be identified and are key to fully understand disease pathogenesis and shed light on mechanisms governing TOKAS.

Via a combination of proteomic, biochemical, gene editing, and cell biology approaches, we have identified small nucleolar ribonucleoprotein (snoRNP) components Dyskerin, NOP56 and NOP58 as candidate substrates of RNF12. These proteins mediate pre-rRNA modifications which drive correct pre-rRNA maturation, and ribosomal function, and is required for correct neural differentiation. Importantly, ubiquitylation of snoRNP components remains poorly studied and could represent a key aspect of snoRNP regulation which could be disrupted in TOKAS. Based on this, we hypothesize that RNF12 regulates snoRNP component function and this mechanism is disrupted in TOKAS.

Understanding of RNF12 ubiquitylation networks to snoRNPs in stem cells will be key step to gain molecular insight in TOKAS pathogenesis and understand the role of ubiquitylation in ND/ID. This research will make progress towards mechanistic characterisation of this disease which will open novel opportunities for therapeutic intervention.