Mislocalization mutant *MESD* due to a biallelic missense variant causes osteogenesis imperfecta type XX by inducing cellular proteotoxicity

**Prajna Udupa**¹, Debasish Kumar Ghosh¹, Gandham SriLakshmi Bhavani¹, Hitesh Shah², Katta M. Girisha¹

¹Department of Medical Genetics, Kasturba Medical College, Manipal, Manipal Academy of Higher Education, Manipal, India.

²Department of Pediatrics, Kasturba Medical College, Manipal, Manipal Academy of Higher Education, Manipal, India.

**Background**: Bi-allelic variants in MESD, an endoplasmic reticulum (ER)-resident chaperone, can cause osteogenesis imperfecta (OI) type XX (MIM #618644). We report a missense variant c.697G>A in MESD in a girl diagnosed with OI type XX while deciphering the mechanisms of disease pathogenicity due to spatio-functional deregulation of mutant MESD.

**Objective**: Deciphering the molecular mechanisms of OI type XX.

**Methods**: Clinical diagnosis, genetic counselling, exome sequencing, Sanger sequencing, in vitro studies, biophysical studies, computational studies.

**Observations**: The proband had osteopenia, vertebral compression fractures, joint hypermobility, and skin laxity. Exome sequencing identified a rare and novel variant, NP_055969.1:p.Asp233Asn, in the ER retention motif (230REDL233>230RENL233) of MESD in the proband. We demonstrated that MESD is a direct chaperone of COL1A1 and COL1A2, and that mislocalization of MESDD233N leads to loss of its chaperone function, resulting in aggregation of COL1A1 and COL1A2. Loss of MESDD233N at ER also leads to unfolded protein response, resulting in increased expression of ER chaperones. Proband fibroblasts showed loss of LRP5/6 localization in the plasma membrane, resulting in transcriptional repression of bone-forming WNT-responsive target genes, such as BMP2, BMP4, and BMP7. Interestingly, aggregated COL1A1 and COL1A2 showed aggrephagy, although this was insufficient to reduce cellular proteotoxicity in proband cells.

**Discussion**: We show that MESD has an important chaperone role for LRP5 and COL1A1 and the lack of wildtype MESD in the ER leads to cytosolic aggregates that are mostly not secreted. We also noted that increased autophagy flux clears collagen aggregates by aggrephagy in the proband fibroblasts.

**Conclusion**: We show for the first time that osteogenesis imperfecta type XX is caused by aggregation of COL1A1. We show that loss of chaperone activity of MESD for COL1A1 leads to...
collagen aggregation. Overall, the imbalance of ER proteostasis due to mislocalization of MESD_D233N leads to downregulation of bone morphogenic protein expression and cellular proteotoxicity.