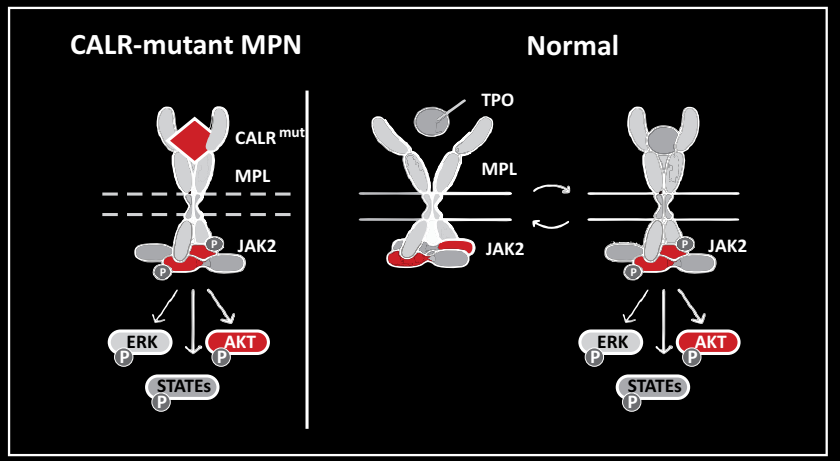
VARIANT SUMMARY						
Gene	Variant (cDNA Alteration)	Variant (Amino acid Alteration)	Variant Allele Frequency (VAF)	Variant Effect$	Variant Classification (AMP)#	Variant Classification (ACMG)!
CALR 2	c.1154_1155insTTGTC	p.Lys385AsnfsTer47	72.08	Frameshift Insertion	Tier2	Pathogenic

MOLECULAR RESULT INTERPRETATION AND SUBSEQUENT ROAD MAP:

The patient was tested negative for JAK2 mutation by RT-PCR but was found to be positive for CALR Type2, 5-bp TTGTC insertion mutation. No other clinically relevant mutation(s) was detected in the rest of the genes tested by NGS. The identified CALR mutation aids in the diagnostic workup for JAK2 negative myeloproliferative neoplasms (MPNs) and predicts the sensitivity towards FDA approved drug Ruxolitinib.

DISCUSSION:

Calreticulin (CALR) is a chaperone protein involved in many cellular processes in the cytoplasm and in the endoplasmic reticulum (ER). The two most frequent CALR mutations are a 52 bp deletion also called type 1, and a 5 bp insertion also called type 2. Mutant CALR proteins leads to the constitutive activation of the thrombopoietin (TPO) receptor, MPL, in myeloproliferative neoplasm (MPN) cells. In normal hematopoiesis (figure), the downstream activation of myeloproliferative leukemia protein (MPL) is regulated by the concentration of TPO to control hematopoiesis whereas in CALR-mutant cells (figure), mutant CALR constitutively activates the downstream molecules of MPL and induces oncogenic transformation in a MPL-dependent manner. Hence, the CALR mutations sensitize the patients for various JAK inhibitors, such as Ruxolitinib.



Model for the constitutive activation of the thrombopoietin (TPO) receptor, MPL, by mutant calreticulin (CALR) in myeloproliferative neoplasm (MPN) cells harboring the CALR mutation