ZSWIM7 is associated with human female meiosis and familial primary ovarian insufficiency

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Introduction

- Primary ovarian insufficiency (POI) affects 1% of women and is associated with significant medical consequences[1].
- A genetic cause for POI can be found in up to 30% of women[2].
- Oogenesis is particularly critical for normal germ cell development and is dependent on meiosis.
- Pathogenic variants in several meiosis genes have already been associated with POI.

Aim

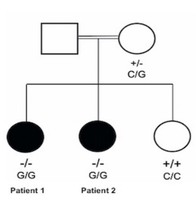
We aimed to identify the genetic mechanism underlying early-onset POI in two sisters from a consanguineous pedigree.

Methods

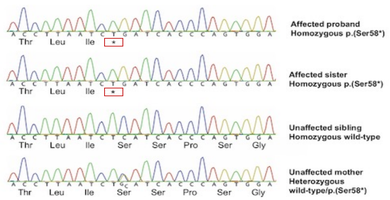
- Genome sequencing using an autosomal recessive model was performed in the two affected sisters and unaffected relatives.
- Quantitative reverse transcriptase PCR (qRT-PCR) was used to study the expression of *ZSWIM7* during fetal gonadal development.
- Four fetal ovary and testis samples (Human Developmental Biology Resource) were included at each of five developmental stages: Carnegie Stage (CS) 22/23 weeks post conception (wpc), 9wpc, 11wpc, 15-16wpc, and 19-20wpc. Four adult ovary and adult testis samples were also included.
- Further analysis of *ZSWIM7* expression in adult tissues was performed using GTEx data (v.8), the Human Protein Atlas (v20.1), and FANTOM5.
- The expression of *ZSWIM7* and associated DNA repair genes in fetal development was studied using bulk RNA sequencing of CS22/23, 9wpc, 11wpc, and 15-16wpc tissue (five per stage).

Results

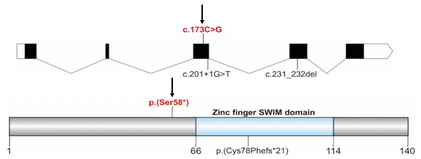
Only one homozygous variant co-segregating with the POI phenotype was found: a single nucleotide substitution in ***ZSWIM7***, NM_001042697.2: c.173C>G; resulting in predicted loss-of-function p.(Ser58*).



Affected kindred with the ZSWIM7 variant (NM_001042697.2:c.173C>G, p.(Ser58)). Solid symbols indicate affected family members. Genotype is indicated underneath tested family members.*

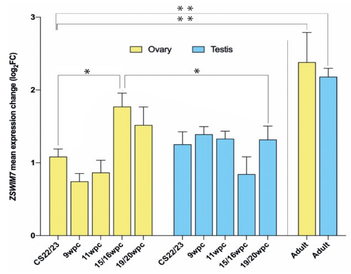


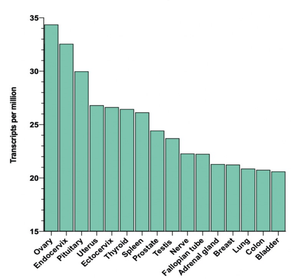
Sanger sequencing of the affected proband, an affected sister, and unaffected mother. NC_000017.11:g.15987294G>C; NM_001042698.2:c.173C>G; NP_001036163.1:p.(Ser58)*



Domains of the human ZSWIM7 protein. The p.(Ser58) stop-gain variant reported in this study is proximal (N-terminal) to the zinc finger SWIM domain (arrows). Previously reported ZSWIM7 variants (associated with male infertility) are also indicated.*

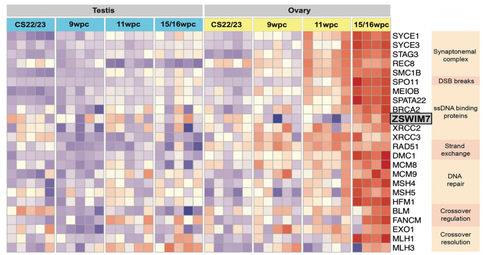
qRT-PCR demonstrated higher expression of *ZSWIM7* in the 15/16wpc ovary compared to testis and in the CS22/23 ovary compared to the 15/16wpc ovary, corresponding to peak meiosis in the fetal ovary. qRT-PCR analysis of adult ovary and testis showed relatively strong expression in the adult testis, where meiosis is actively occurring, but also in the adult ovary. This observation was supported following analysis of publicly available RNA expression datasets.





*Far left: qRT-PCR mean expression (log_2) of ZSWIM7 in various tissues compared to reference (GAPDH) and relative to the expression of ZSWIM7 in a Carnegie Stage (CS) 22 ovary sample. Four fetal ovary and fetal testis tissue samples were included at each of the following stages: CS22/CS23, 9wpc, 11wpc, 15/16wpc, and 19/20wpc. Bar heights indicate mean expression (*P < 0.05; **P <0.01). Left: ZSWIM7 expression across adult tissues from the GTEx database (v8). Data are expressed in transcripts per million.*

Bulk RNA sequencing of fetal tissue revealed that *ZSWIM7* showed similar temporal expression in the human fetal ovary to other homologous recombination genes known to be important for meiosis prophase I.

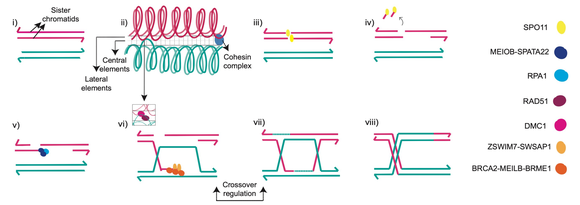


Heatmap representing differential gene expression of key meiotic genes during prophase I across four developmental timepoints (CS22/23, 9wpc, 11wpc, 15/16wpc).

Conclusions

- To our knowledge, this is the first time *ZSWIM7* has been associated with human POI.
- *ZSWIM7* Is a known DNA repair gene and has recently been associated with male infertility[3].
- *Zswim7(Sws1)/Swsap1* mutant mice reproduce the infertility phenotype, demonstrating marked meiotic abnormalities[4].
- These data provide evidence for a role for *ZSWIM7* in human female meiosis, implicate it in the pathogenesis of POI, and emphasize the importance of genes associated with homologous recombination and specifically meiosis prophase I in this condition.
- A broader mechanistic understanding of POI can be gained from considering meiotic genes as functional partners.

Genes associated with primary ovarian insufficiency (POI) and their relationship with the stages of meiosis I prophase.



References

1. European Society for Human Reproduction and Embryology (ESHRE) Guideline Group on POI, Webber L, Davies M, et al. ESHRE Guideline: management of women with premature ovarian insufficiency. *Hum. Reprod.* 2016;31(5):926-937.
2. Eskenazi S, Bachelot A, Hugon-Rodin J, et al. European Society for Human Reproduction and Embryology (ESHRE) Guideline Group on POI. Next generation sequencing should be proposed to every woman with "idiopathic" primary ovarian insufficiency. *J. Endocr. Soc* 2021;5(7):bvab032.
3. Li Y, Wu Y, Zhou J, et al. A recurrent ZSWIM7 mutation causes male infertility resulting from decreased meiotic recombination. *Hum. Reprod.* 2021.
4. Abreu CM, Prakash R, Romanienko PJ, Roig I, Keeney S, Jasin M. Shu complex SWS1-SWSAP1 promotes early steps in mouse meiotic recombination. *Nat. Commun.* 2018;9(1):3961.

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