P-508: A pilot study to identify Single Nucleotide Variants (SNVs) as predictors of oocyte/embryo quality in fertile women undergoing Preimplantation Genetic Testing for Monogenic Disorders (PGT-M)

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BACKGROUND

Genetic profiling of prospective parents has been one of the approaches for identifying biomarkers of IVF success. Specific genotypes have been associated with IVF failure, gamete/embryo quality etc., with most studies focusing on infertile populations. Recent studies identified SNVs (rs1801133, rs1801131 in *MTHFR*, and rs2305957 on chromosome 4 linked to *INTU, SLC25A31, HSPA4L, PLK4, MFSD8, LARP1B* and *PGRMC2*) associated with embryo quality or chromosomal status in infertile women[1, 2]. Additionally, preimplantation development may be influenced by pathways involved in follicle development, meiosis, mitosis and DNA repair, for which many other genes have been implicated in human and animal studies.

STUDY QUESTION

Are SNVs in 26 selected genes potentially involved in preimplantation development, associated with oocyte and embryo quality in fertile women undergoing PGT-M?

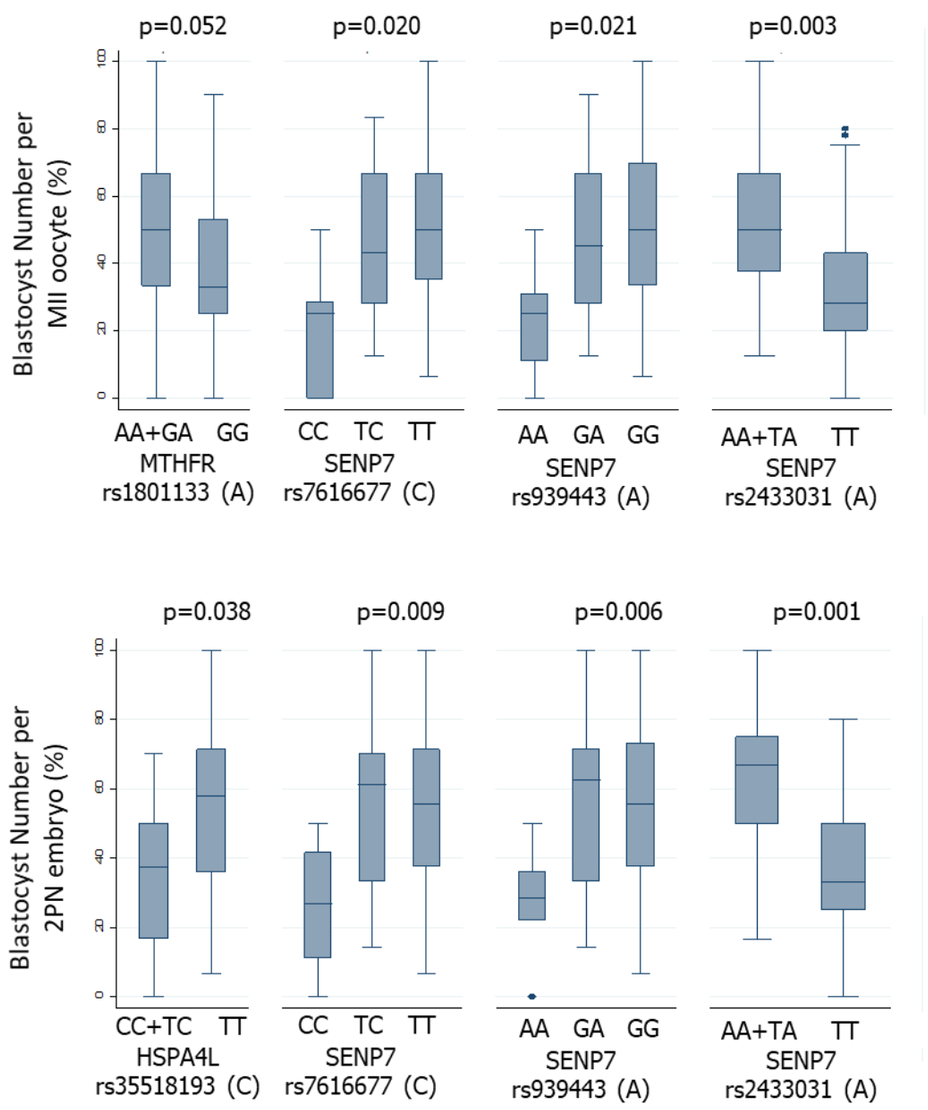
PARTICIPANTS/MATERIALS, SETTING, METHODS

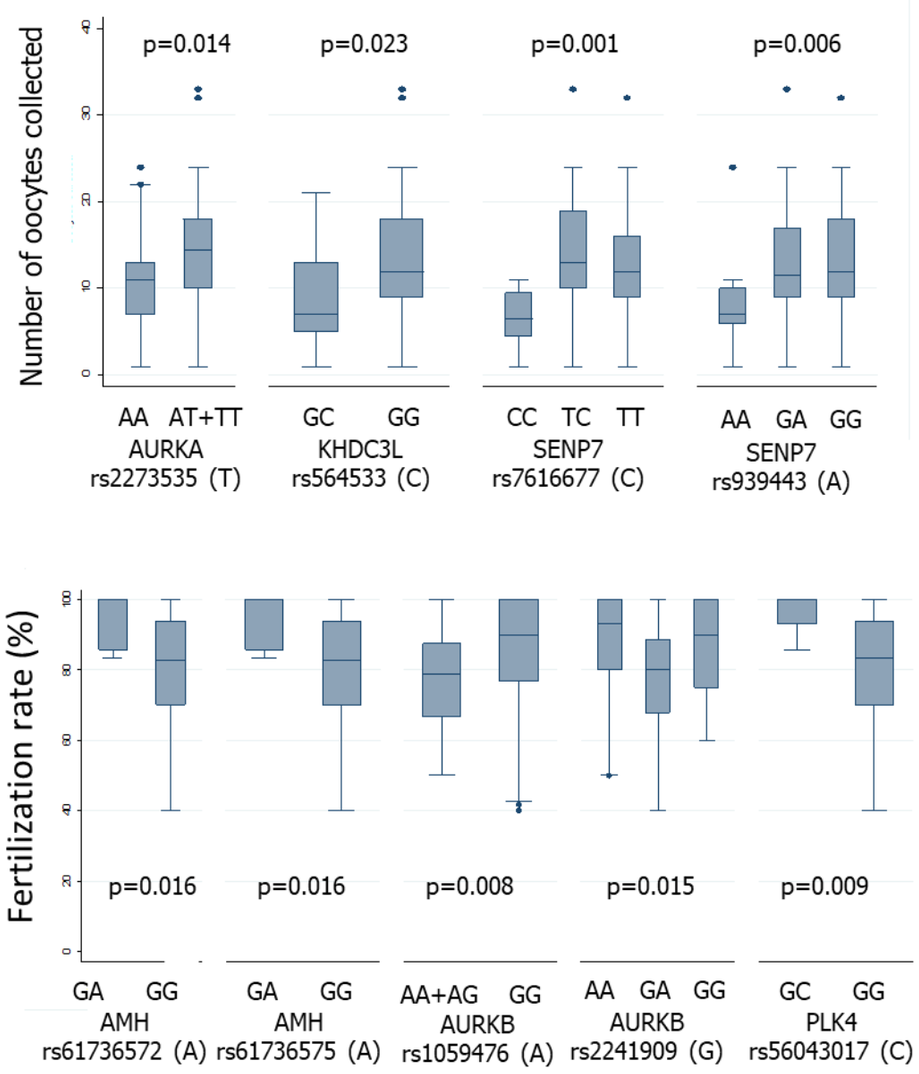
The study was performed at the University of Athens Laboratory of Medical Genetics, and focused on fertile couples undergoing PGT-M. NGS was performed for 85 women undergoing 107 PGT-M-cycles between 2013-2019. Maternal genotypes were analyzed in association with **number of oocytes collected**, **fertilization rate**, **percentage of blastocysts developed per MII oocyte** and **percentage of blastocysts per 2PN embryo** (outcome measures). Genomic DNA from fertile prospective mothers was genotyped using exonic NGS (Qiaseq™ Targeted Custom Panel, Miseq Reagent Nano kit v2, Illumina) for 18 genes (*AIRE-AMH-AURKA-AURKB-AURKC-FSHR-HSPA4L-HUWE1-INTU-KHDC3L-LARP1B-MFSD8-MTHFR-PGRMC2-PLK4-SENP7-SLC25A31-WBP1*) and 9 selected SNVs in a further 8 genes: rs175080(*MLH3*), rs1799963(*F2*), rs6025(*F5*), rs5918(*ITGB3*), rs5985(*F13A1*), rs1805087(*MTR*), rs1801394(*MTRR*), rs28756992(*MLH3*) and rs2305957(*HSPA4L*).
Qubit Fluorimeter (Invitrogen) and Bioanalyzer 2100 (Agilent) were employed for DNA and library quantitative and qualitative assessment. Data analysis was performed with Qiaseq DNA V3 panel analysis και VarAFT 2.13.

- The study (September 2017-2019) was approved by the University of Athens Bioethics Committee and National Authority of Assisted Reproduction
- Statistical analysis was performed by the Center for Clinical Epidemiology and Outcomes Research, using the statistical software STATA SE v.13

MAIN RESULTS

- A 20x coverage was achieved in all gene exons and SNVs investigated, identifying 121 variants.
- **Significant associations were revealed for 16 SNVs in *AMH, AURKA, AURKB, HSPA4L, KHDC3L, MTHFR, PLK4* and *SENP7*. These are illustrated in the graphs for each of the four studied parameters. Minor alleles for each SNV are indicated in brackets.**
- No significant associations were revealed for rs1801131 and rs2305957 in our study group.
- Of the 7 genes previously linked to rs2305957 on chromosome 4, only *HSPA4L* and *PLK4* revealed significant associations with the number of oocytes and oocyte fertilization rate in our study population [3, 4]
- Other studies have previously supported a role of *AURK*s, *SENP7*, *AMH* and *KHDC3L* in reproduction [5, 6, 7, 8]





LIMITATIONS:

The present data should be considered preliminary and taken with caution, setting the path for further investigation in a larger group of fertile women and prospective validation in other PGT-M cycles, to confirm our findings.
The main limitations of the study include its retrospective nature, whereby IVF practices have changed considerably over the period of the study (2013-2019).

WIDER IMPLICATIONS OF THE FINDINGS:

Infertility is a complex condition. Identifying prognostic factors in ART patients is confounded by multiple variables (parental age, stimulation and fertilization protocols, embryo transfer stage/conditions etc.). PGT-M facilitates investigation of a fertile ART population, minimizing many confounding variables, and potentially facilitating the identification of genomic biomarkers predictive of gamete/embryo quality.

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References:

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