

EMBRYONIC CELL FREE DNA ANALYSIS IN SPENT CULTURE MEDIA: TEST RELIABILITY AND CLINICAL APPLICATIONS

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SESSION CONTENT

The high incidence of aneuploid embryos in IVF (ranging from 20% to 100%) is an important biologic burden. Preimplantation genetic testing for aneuploidy (PGTA) is, at present, the most reliable method to assess the chromosomal status of preimplantation embryos. Currently, DNA isolated and amplified from trophoctoderm (TE) biopsies and analyzed by means of next-generation sequencing (NGS) is the state of the art for this technique.

Despite the value of PGT-A, there are two main unsolved problems causing controversies in the scientific community. First, embryo biopsy requires specialized equipment and expertly trained operators to maintain protocols to protect embryo viability. However, possible harm to the embryo is always a concern of both doctors and patients.

Even similar ongoing implantation rates having been reported in a study comparing transfer of nonmanipulated blastocysts and blastocysts with TE biopsy, the putative variability associated with the different expertise of operators that were not properly trained should not be dismissed. Second, mosaicism, a biologic event occurring at any stage of embryo development, can have an impact on the accuracy of diagnosis based on the analysis of only 5–10 cells from a blastocyst with >100 cells, introducing uncertainty.

There is increasing evidence for a “true” noninvasive approach consisting of the analysis of cell-free DNA (cfDNA) released by the embryo into the spent blastocyst medium (SBM) during the late stages of preimplantation development. SBM, in which the embryo is cultured, is routinely discarded at the time of transfer or freezing. Several publications have compared the chromosomal results of the criterion standard PGT-A from TE biopsies and those of the cfDNA in the SBM to establish concordance rates. All reports have obtained high cfDNA amplification rates, ranging from 80% to 100%. However, the concordance rates have been variable, with 3.5% reported in a proof of concept study and 30.4%, 65%, and 85.7% reported in later studies.

Since March 2018, Igenomix has been leading a Worldwide Multicentric Study (*NCT0352093*). The aim of this study is to estimate the concordance rates between TE biopsies and SBM with an optimized protocol incorporating technical improvements at two levels: the culture conditions in the IVF laboratory and the NGS protocol applied to the analysis of the SBM. Importantly, embryos were not subjected to any intervention during in vitro culture, such as previous assisted hatching, vitrification, or blastocentesis.

In addition, to test the functional relevance of embryonic cfDNA testing, we retrospectively compared the clinical outcome of euploid single embryo transfer (SETs) with concordant or discordant results in TE biopsy versus SBM, considering that the selection of embryos for transfer was always performed according to the results obtained by TE biopsies and that the SBM was subsequently analyzed.

OBJECTIVES

According to the exposed above, the general learning objectives of this session is to elucidate to the audience what is non-invasive preimplantation genetic test, what is already described on scientific literature and what are still under research.

The specific objective is to present how Igenomix has been approaching this innovative technology, what we have learned after two articles published and one RCT study on going and how could this new diagnostic strategy changes the IVF centers routine either their clinical perspective of the preimplantation genetic chromosome screening and embryo selection.

Multicenter concordance study (8 centers involved, n=3,245 embryos)

Hypothesis: High concordance between TE and SBM ($\geq 80\%$), with better clinical outcomes in euploid TE-SBM.

