

NEW DEBATE

What next for preimplantation genetic screening?

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Preimplantation genetic diagnosis for aneuploidy screening (preimplantation genetic screening—PGS) has been used to detect chromosomally normal embryos from subfertile patients. The main indications are advanced maternal age (AMA), repeated implantation failure, repeated miscarriages and severe male factor infertility. Many non-randomized PGS studies have been published and report an increase in implantation rate, and/or a decrease in miscarriage rate. Recently, two randomized controlled trials have been conducted on patients with AMA as the only indication. Neither study showed a benefit in performing PGS using live birth rate as the measure of success. The debate on the usefulness of PGS is ongoing; the only effective way to resolve the debate is to perform more well-designed and well-executed randomized clinical trials.

Keywords: preimplantation genetic screening; randomized controlled trials; miscarriage; live birth rate

The main goal of aneuploidy screening of embryos *in vitro* derived from subfertile patients undergoing IVF is to increase their chance of a healthy pregnancy. Such preimplantation genetic screening (PGS) involves the aspiration of a single cell followed by fluorescence *in situ* hybridization (FISH) using probes for a limited number of chromosomes on the nucleus of this cell to determine the ploidy status of the embryo. Subsequently, euploid embryos are selected for transfer and aneuploid embryos are discarded, analysed to provide confirmatory diagnosis or used for research.

The main indications suggested for PGS are advanced maternal age (AMA; usually defined as maternal age over 37 or 38 years), repeated implantation failure (RIF; usually defined as three or more failed embryo transfer procedures involving high-quality embryos), repeated miscarriage in patients with normal karyotypes (RM; usually at least three previous miscarriages) and severe male factor infertility (SMF; usually defined as abnormal semen parameters)

(Harper *et al.*, 2006). In addition, PGS has been used for a variety of 'other' indications including a previous sporadic genetically abnormal pregnancy, poor embryo quality, previous radiotherapy and single embryo transfer (ESHRE PGD Consortium, unpublished data).

Since the publication of the first papers on PGS on cleavage stage embryos (Gianaroli *et al.*, 1997) and polar bodies (Munné *et al.*, 1995; Verlinsky *et al.*, 1995), there have been several hundred publications on this topic and PGS has been established in many IVF centres worldwide. There has been a steady increase in the number of PGS cycles reported to the Consortium, from 116 cycles in the data collection from 1997–1998 to 2087 cycles in 2004 (Harper *et al.*, 2007). The preliminary analysis of data from 2005 follows this trend with 2316 PGS cycles (ESHRE PGD consortium, unpublished data).

There have been a number of non-randomized comparative studies of IVF/ICSI with or without PGS but most report only on implantation rate and clinical pregnancy rate per

embryo transfer procedure whereas the main outcome measure in subfertility studies is suggested to be live birth rate per cycle and per patient (Barlow, 2003). For AMA, Obasaju *et al.* (2001) found no increase in implantation rate or clinical pregnancy rate. Munné *et al.* (1999) originally found no increase in implantation rate, but an increase in ongoing pregnancy and live birth rate per embryo transfer procedure. In a later study, the same group found a significant increase in implantation rate but did not report the live birth rate (Munné *et al.*, 2003). Gianaroli *et al.* (1999) reported an increase in implantation rate and clinical pregnancy rate per embryo transfer procedure. Recently, other studies have shown a significant reduction in spontaneous abortions (Munné *et al.*, 2006; Colls *et al.*, 2007). For RIF patients, Gianaroli *et al.* (1999) showed that PGS offered no improvement in the implantation or clinical pregnancy rate per embryo transfer procedure. For RM patients, Munné *et al.* (2005) showed a significant reduction in spontaneous abortions.

The most obvious criticisms of non-randomized studies are their poor experimental design and inadequate control groups. Few of these studies report delivery rate as the end point, some involve small numbers of patients, some use two-cell biopsies and some use low numbers of probes.

Three randomized controlled trials have been performed for AMA (Staessen *et al.*, 2004; Stevens *et al.*, 2004; Mastenbroek *et al.*, 2007). The first two showed no significant difference in ongoing and live birth rates between the control and PGS group. The latest study showed that the PGS group had a significant decrease in the chance of achieving an ongoing pregnancy and live birth (Mastenbroek *et al.*, 2007). A randomized controlled trial evaluating the effectiveness of PGS in subfertile women under the age of 35 undergoing IVF treatment with single embryo transfer (aimed at helping choose the 'best' embryo for single embryo transfer) showed no benefit of PGS (Staessen *et al.*, 2007, abstract presented at the ESHRE meeting, Lyon). No randomized controlled trials have been reported for other PGS indications.

There are also criticisms for each of the three randomized trials for AMA. One study reported on only 39 patients (Stevens *et al.*, 2004). Staessen *et al.* (2004) biopsied two cells, as opposed to one as is usual in PGS, which may have an adverse affect on embryo viability (Cohen *et al.*, 2007). The most recent and comprehensive study by Mastenbroek *et al.* (2007) showed a high percentage of embryos without a diagnosis (20%), and did not include probes for chromosomes 15 and 22 (trisomies of which are prevalent among spontaneous abortions). In addition, the pregnancy rate in the PGS group was 6% when only undiagnosed embryos were transferred and this was significantly lower compared to the non-PGS group suggesting a detrimental effect of the embryo biopsy procedure itself.

The debate on the benefit of PGS is ongoing (Twisk *et al.*, 2006; Cohen *et al.*, 2007). The disadvantages of PGS are that it is labour intensive, invasive to the embryo and expensive. Mosaicism is common in cleavage stage embryos and analysis of only one cell means that some embryos, that may have developed normally, would not be considered for transfer if the single cell tested gives an abnormal result. The advantages

of PGS are that aneuploid embryos which have little or no viability are not transferred or frozen. In this way, high-risk patients potentially avoid miscarriage and a viable abnormal pregnancy. Knowledge of a high frequency of aneuploidy in their embryos may further help some patients achieve closure or choose other options, such as donor gametes, to achieve pregnancy. Finally fewer trisomic conceptions appear to result (Munné *et al.*, 2005), and fewer embryos are replaced, potentially reducing multiple conception risk.

The rapid increase in the application of PGS has raised questions about its efficacy for routine use. The authors of this paper believe that the most effective way to resolve the debate about the usefulness of PGS is to perform well-designed and well-executed randomized clinical trials. The trials should be designed to assess the anticipated benefits (a substantial improvement in live birth rates and reduction in miscarriage rates), in relation to the risk and cost for the couple, while addressing biopsy safety, test accuracy and diagnostic efficiency, in order to identify appropriate indications for PGS. ESHRE is investigating the possibility of setting up a multi-centre randomized controlled trial. The results of this trial should help to clarify whether PGS has any value for IVF patients, and if so, to specify those subsets of patients for whom PGS is efficacious.

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