What next for preimplantation genetic screening (PGS)? A position statement from the ESHRE PGD Consortium steering committee†

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Since 2004, there have been 11 randomized controlled trials (RCTs) mainly for advanced maternal age (AMA), which have shown no benefit of performing preimplantation genetic screening (PGS). Ten of the RCTs have been performed at the cleavage stage and one at the blastocyst stage. It is probable that the high levels of chromosomal mosaicism at cleavage stages, which may result in the tested cell not being representative of the embryo, and the inability to examine all of the chromosomes using fluorescence in situ hybridization, have contributed to the lack of positive outcome from the RCTs. We suggest that future RCTs should examine alternative biopsy timing (polar body and/or trophectoderm biopsy), and should apply technologies that allow more comprehensive testing to include all chromosomes (microarray-based testing) to determine if PGS shows an improvement in delivery rate. Currently there is no evidence that routine PGS is beneficial for patients with AMA and conclusive data (RCTs) on repeated miscarriage, implantation failure and severe male factor are missing. To evaluate benefits of PGS, an ESHRE trial has recently been started on patients with AMA using polar body biopsy and array-comparative genomic hybridization, which should bring more information on this patient group in the near future.

Key words: PGS / ESHRE PGD Consortium / randomised controlled trial

The main goal of aneuploidy screening of embryos derived from sub-fertile patients undergoing IVF is to increase their chance of a healthy pregnancy. Preimplantation genetic screening (PGS) has mainly involved the aspiration of a single cell followed by fluorescence in situ hybridization (FISH) using probes for a limited number of chromosomes to determine the ploidy status of the embryo. Subsequently, euploid embryos are selected for transfer and aneuploid embryos are discarded and 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 (RM) in patients with normal karyotypes (usually at least three previous miscarriages) and severe male factor (SMF) infertility.
(usually defined as abnormal semen parameters; Goossens et al., 2009). 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 articles on PGS using cleavage-stage
embryos (Gianaroli et al., 1997) and polar bodies (Munne et al.,
1995a,b; Verlinsky et al., 1995), there have been numerous
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 ESHRE PGD Consortium, from 116
cycles in the data collection from 1997 to 1998 to 3900 cycles in
2007 (Goossens et al., 2009).

Until recently, the majority of studies on PGS were non-randomized
studies with poor experimental design and inadequate control groups.
Only a few of these studies reported delivery rate as the end-point,
some involved small numbers of patients, some used 2-cell biopsies
and some used low numbers or random sets of FISH probes (Twisk
et al., 2006).

In 2007, Mastenbroek et al. published their RCT involving 200
patients per arm (control and treatment group) which showed a sig-
nificant lowering of the delivery rate in patients who had undergone
PGS. The article was highly criticized for its poor efficiency, low preg-
nancy rate in the control group, etc. (Cohen et al., 2007; Donoso
et al., 2007). Meanwhile, the American Society of Reproductive
Medicine (The practice committee of the Society of Assisted Repro-
ductive Technology and the American Society of Reproductive
Medicine, 2008), American College of Obstetricians and Gynaecolo-
gists (2008) and the British Fertility Society (Anderson and Pickering,
2008) have all issued statements that PGS should not be performed
for any indication. In 2007, the ESHRE PGD Consortium was
asked to write a position statement in reply to the Mastenbroek
paper, but at that time there was insufficient data to write such a state-
ment. Instead, a comment was written by some of the members of the
steering committee (Harper et al., 2008). We did not feel that we
could say that PGS should no longer be performed for AMA as
there were many criticisms of the three randomized controlled trials
(RCTs) published at that time (Staessens et al., 2004; Stevens et al.,
2004; Mastenbroek et al., 2007). Instead our conclusion was 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’.

There are now 11 RCTs published on PGS, 10 using cleavage-stage
biopsy (Staessens et al., 2004, 2008; Stevens et al., 2004; Mastenbroek
et al., 2007; Blockeel, 2008; Hardarson et al., 2008; Mersereau et al.,
2008; Debrock et al., 2009; Meyer et al., 2009; Schoolcraft et al.,
2009) and one using blastocyst biopsy (Jansen et al., 2008). All have
used FISH testing of a limited number of chromosomes and none
have shown an improvement in delivery rates, with some showing a
significant decrease in delivery rates after PGS. Most of the RCTs
have been for patients with AMA (Staessens et al., 2004; Stevens
et al., 2004; Mastenbroek et al., 2007; Hardarson et al., 2008;
Debrook et al., 2009; Schoolcraft et al., 2009).

The lack of positive outcome from these RCTs can be explained by
the likelihood that the tested blastomere is not representative for
the whole embryo (Vanneste et al., 2009a). Indeed, high levels of
chromosomal mosaicism have been observed in blastomeres from
cleavage-stage embryos evaluated by FISH for a limited number of
chromosomes in infertile women (Harper et al., 1995; Munne et al.,
1995a, b) or by array technology for all chromosomes in fertile
women (Vanneste et al., 2009b). Therefore, future work in this area
should explore different timing for biopsy (polar body and trophecto-
derm biopsy) and the use of new technology that allows for more
comprehensive screening of chromosomes (array-based technology).

Already, clinics are applying array-comparative genomic hybridization
(a-CGH) at the cleavage stage for PGS (Hellani et al., 2008). Before
these procedures are used routinely, the array platforms need to be
validated (Le Caïnec et al., 2006; Fiegler et al., 2007, Mammas et al.,
submitted) and RCTs are needed to prove that use of this procedure
will result in a significant increase in delivery rates (Harper and Harton,
submitted).

We also said in our comment (Harper et al., 2008) that ESHRE was
investigating setting up a multicentre RCT for PGS. This study has
been set up and is currently undergoing initial trials to assess the tech-
nology (Geraedts et al., 2009). The trial will be performed on patients
with AMA using polar body biopsy and a-CGH. To fully explore the
PGS question, a similar trial will need to be conducted on blastocyst
biopsy.

As for PGS for other indications, such as RIF (Blockeel et al., 2008),
RM and SMF, there is a lack of data. Like the use of PGS for AMA, it
would stand to reason that a different stage of biopsy and array-based
technology would be needed to assess these indications. Centres that
have already begun RCTs using ‘older’ methods (cleavage-stage biopsy
with FISH testing for a limited set of chromosomes) are encouraged to
finish the trial and report the results to add to the literature. Different
subgroups of each indication and different age ranges of the patients
chosen for the RCTs and culture methods could affect the results of
the RCTs (Beyer et al., 2009). We strongly recommend that clinics
interested in these indications should perform RCTs to validate the
use of PGS for these patients using delivery rate as the standard
outcome measure.

Conclusion

The widespread use of PGS without evidence of its ability to improve
delivery rates has been a problem in the field of IVF. We must learn
from this experience and ensure that techniques are brought into
our treatment programmes only when there is scientific data to
support their use. It is hoped that other centres will undertake rigor-
ous RCTs to validate the use of PGS so that in future only proven
techniques are applied in clinical practice.

There is now ample evidence that PGS for AMA using cleavage
stage biopsy and FISH testing of a limited number of chromosomes
is not a valid procedure and should be replaced by more appropriate
approaches. Until results of RCTs using a different biopsy stage and
arrays can demonstrate a significant increase in delivery rates, there
is no evidence that routine PGS is beneficial for patients with AMA.
Currently there is a lack of scientific data on the use of PGS applied
for RM, implantation failure and SMF and so RCTs are also required
for these indications. Multicentre studies such as the one now
launched by ESHRE should be encouraged to obtain information on
best approaches and eventually establish valid techniques for PGS in
routine practice in the benefit of patients.


References


Cohen J, Wells D, Munne S. Removal of 2 cells from cleavage stage embryos is likely to reduce the efficacy of chromosomal tests that are used to enhance implantation rates. Fertil Steril 2007;87:496–503.


