Clinical Pregnancy Following Pre-Implantation Genetic Diagnosis for Cystic Fibrosis

Abstract:
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Pre-implantation genetic diagnosis (PGD) is an established alternative to prenatal testing for couples at risk of transmitting genetic disorders such as cystic fibrosis (CF). PGD screens pre-implantation embryos, allowing the safe transfer of those identified as unaffected. Awareness of CF carrier status in Ireland is increasing following the introduction of neonatal screening in 2011. PGD is the most acceptable reproductive strategy for many at risk Irish couples but until now the treatment necessitated travelling abroad. In 2012, the Irish Medicines Board licenced two Irish fertility clinics to carry out embryo biopsy for PGD. This is the first reported clinical pregnancy following PGD carried out in Ireland.

Introduction
Cystic Fibrosis (CF) is an autosomal recessive condition caused by mutations of the cystic fibrosis transmembrane conductance regulator (CFTR) gene. Ireland has the highest incidence of CF in the world and 1 in 19 of the population is a carrier. The birth of an index case has always identified couples at risk (where both parents are carriers) but awareness of CF risk is now rising due to the introduction of neonatal CF screening in July 2011 (identifying both disease and carrier status). Couples at risk must make difficult reproductive choices. Pre-implantation genetic diagnosis (PGD) may be the most acceptable strategy for many Irish couples but until now it has been necessary to travel abroad for the treatment.

Case Report
A couple was referred to Cork Fertility Centre (CFC) for assessment with regard to PGD. The male partner, aged 30 years, was affected by CF (homozygous for F508 mutation); the female partner was a known CF carrier (G551D mutation). Both partners underwent routine fertility assessment. semen analysis showed azoospermia. Normal levels of FSH, AMH and a normal antral follicle count indicated satisfactory ovarian reserve. Testicular sperm extraction (TESE) was carried out at CFC confirming the presence of motile sperms which were cryopreserved. Blood samples from both partners were sent to Reprogenetics (Oxford, U.K.) for preliminary genetic analysis. Polymerase chain reaction (PCR) was used to amplify a fragment of DNA containing the CFTR gene. The presence or absence of the mutation was then determined using mini-sequencing. Additionally, polymorphisms (highly variable pieces of DNA situated in close proximity to the CFTR gene), were amplified and analysed. The risk of a mis-diagnosis from this technique is negligible. However current technology allows for an error rate of 1 to 2%.

The female partner underwent a routine IVF cycle. Thirty eggs were collected and inseminated by intracytoplasmic sperm injection (ICSI) using the cryopreserved sperms. Fifteen embryos were biopsied on day 3 following egg collection; a single blastomere was removed from each and sent to Reprogenetics for genetic analysis while the embryos remained in culture. Five days following egg collection the genetic analysis results identified 5 unaffected embryos. A fresh embryo transfer was not carried out because of concerns about the possibility of ovarian hyperstimulation syndrome and the embryos were cryopreserved using a closed vitrification system. A frozen embryo transfer cycle was carried out 8 weeks after the fresh cycle, with a single embryo transferred. Ultrasonography confirmed a viable singleton intrauterine pregnancy.

Discussion
PGD was first described in the medical literature in 1990. The European Society of Human Reproduction and Embryology (ESHRE) PGD Consoripium 2007 reported that 5,878cycles of PGD had been performed in 57 European centres, resulting in 1,206 live births. In the Irish case, the first reported clinical pregnancy following PGD carried out in Ireland, is an important milestone. Its outcome depended on the reliability of several technical advances- ICSI/TESE, blastocyst culture, embryo biopsy and embryo vitrification. Although cryopreservation, in this case, had not been intended at the outset it will be a necessary part of all PGD cases in the future as biopsy moves from day 3 to day 5 (the day 5 culture, embryo biopsy and embryo vitrification. Although cryopreservation, in this case, had not been intended at the outset it will be a necessary part of all PGD cases in the future as biopsy moves from day 3 to day 5 (the day 5 embryo is more robust and several cells can be removed, increasing the certainty of genetic diagnosis). The challenge involved here was greater because only 50% of embryos were likely to be unaffected by CF (compared to 75% when both partners are carriers for the condition). Increasing awareness of genetic risk is inevitable and where Irish couples are burdened with difficult reproductive choices, the option of PGD in Ireland is a welcome development.

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References