NEWS AND VIEWS

CONTROVERSIES IN ASSISTED REPRODUCTION AND GENETICS

Making the Best Use of Human Embryos

The best use of human embryos must be that which maximizes a patient's chances of conceiving a normal singleton pregnancy and of giving birth to a healthy baby, with minimum risk to herself. Assisted conception treatment is no different from any other medical treatment, in that it should be tailored to the needs of individual patients, should do them no harm, and should maximize their chances of a successful outcome. In attempting to achieve this there are a number of issues that should be considered.

It must be remembered that the objective is not to achieve any pregnancy at any cost; therefore the question of the number of embryos that should be replaced should be the first consideration. There are couples for whom a high-order multiple pregnancy would be far more disastrous than no pregnancy at all. There is also the question of supernumerary embryos and maximization of their use by cryopreservation.

The number of embryos that can safely be transferred has been the subject of considerable debate. In some cases it is obvious—patients with a history of multiple pregnancy should clearly have the number of embryos replaced limited to one or two at the most. Replacement of two rather than three embryos has been shown to reduce the multiple pregnancy rate and improve perinatal outcome (1–3). However, in older patients, particularly those over the age of

40, there is a case for transferring three or even four embryos (4–6). The decline in conception rates with age is well documented (7,8). The decline in embryo quality (9) and implantation rate (10) and the increase in the proportion of chromosomally abnormal embryos (11) in relation to increasing age is well documented. However, it is possible that older patients who achieve pregnancy do so not because they had three embryos transferred, but because they were able to produce three good embryos to transfer, i.e., they had a sufficiently good response to stimulation and enough good-quality oocytes to fertilise and develop. Widra *et al.* (12) identified a subgroup of women over 40 who responded well to ovarian stimulation and produced good-quality embryos and achieved pregnancy rates similar to those of younger patients. Similarly, at our clinic, we have found that patients who have sufficient good-quality embryos remaining to cryopreserve have a higher pregnancy rate following fresh embryo transfer than those who have no embryos available to freeze. This suggests that pregnancy is related to the total number of good-quality eggs and embryos achieved, not just to the number replaced.

We have also found a significantly higher pregnancy rate in patients age 40+ who have three embryos replaced rather than two (Bourn Hall Clinic, unpublished data), but closer examination of the data shows that the pregnancy rate is significantly lower when there are only three embryos available to transfer. Very few two-embryo transfers were carried out electively in the over-40s, rather than because there were only two embryos to transfer. We would therefore need to carry out a randomized trial to

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establish true pregnancy rates with elective two- versus three-embryo transfers in the over-40s. Murdoch (13) suggests that this is not a significant problem, since the percentage of patients over the age of 39 is small and most of them do not respond well to stimulation and will, therefore, not have the option of a three-embryo transfer. This is not our experience. Approximately 10% of patients treated at our clinic are age 40 and above, i.e., 70–100 patients per year; the majority of them have three embryos to transfer, and for us it is therefore a significant dilemma. At the other end of the scale, those patients who have a history of multiple pregnancy, or who are young and have a high chance of conception, need to have their treatment planned in such a way that the risk of multiple pregnancy is eliminated as far as possible. The only practical way to achieve this is to replace only one embryo. Our problem with this, however, is our inability to identify those embryos that are most likely to implant. We still rely on our subjective assessment of embryo morphology as our major marker of developmental potential. One way of dealing with this is to extend the culture, to either a later cleavage or the blastocyst stage (14).

Early attempts at culture to blastocyst gave poor results (15). However, more recent work has shown that the use of more appropriate media can give rise to blastocyst formation rates of 50% or above, with implantation rates in the range of 20 to 50% (16). Culture to blastocyst will, given current results, decrease the numbers of embryos available for cryopreservation. However, since the implantation rate per blastocyst is higher than for early cleavage-stage embryos, the overall pregnancy rates may be the same, but the risk of multiple pregnancy may be virtually eliminated, and the number of stimulation cycles and embryo transfers needed to achieve a pregnancy thereby reduced. This technique might also be applied to our older patients. Culture to the blastocyst stage would give some indication of the developmental competence, and thus we could avoid the need to replace three or more embryos in the hope that at least one of them could be normal. This would then reduce the risk of multiple pregnancy, however small, for these patients. There remains the possibility, however, that none of the cultured embryos will reach the blastocyst stage. A lower rate of blastocyst formation has been reported in women aged over 40, but the live birth rate per blastocyst is unaffected by age (17). It might be argued that if no embryos reach the blastocyst stage, then it is unlikely that any of them would have

implanted anyway. However, this assumes that the ability of embryos to develop in vivo is the same as their potential to develop in vitro. We cannot be certain that some embryos that fail to develop during in vitro culture would not have developed in vivo. There is also the problem that many patients prefer to have at least some embryos transferred rather than none, even though they realize their chance of success may be very small.

Culture to the blastocyst stage does appear to give the best chance of achieving a single pregnancy by replacing a single embryo. However, with the current strategies and technology, it is possible to maximize the chances of conception following a single in vitro fertilization (IVF) cycle by freezing excess embryos. Cumulative pregnancy rates in the range of 40–50% can be achieved if fresh embryo transfer is followed by two frozen embryo transfers (18,19). Horn *et al.* (19) have suggested that the ideal strategy is to cryopreserve some excess embryos at the pronucleate stage and some at cleavage stages, a strategy employed at our clinic. Then, if the pronucleate embryos fail to cleave after thawing, cleaving embryos may be available for transfer.

The variation in numbers of embryos transferred, the use of cryopreserved embryos, and patient factors such as age and cause of infertility complicate the presentation of results. To evaluate properly the outcome of a single IVF cycle, pregnancies from cryopreserved embryo transfers must also be taken into account (20). The major problem with this approach, as pointed out by the same authors, is that a variable amount of time must elapse before the complete outcome from any one cycle is known. Jones *et al.* (20) also suggest that we should add to the pregnancy rate only those frozen transfers that did not have a corresponding pregnancy from a fresh embryo transfer, so that we arrive at a figure for the percentage of cycles that give rise to at least one pregnancy. If, when adding in the frozen embryo pregnancies, we add them to the overall number of IVF cycles, then the true effect of frozen embryo transfers is minimized. At our unit they increase the overall clinical pregnancy rate from 16.8 to 22.2% per oocyte collection. However, if we take only those cycles in which embryos were frozen, the overall pregnancy rate is increased to 33.8%. Projecting the likely outcome if we assume that all patients who have frozen embryos will return for replacement, and they use all their embryos, the expected cumulative pregnancy rate per egg collection would be 46% (Bourn Hall Clinic, unpub-

lished data). These figures show that embryo cryopreservation does maximize the chance of conception from any one stimulation cycle and ensures that none of the excess embryos produced using current stimulation regimes are wasted.

If, instead of cryopreserving early embryos, we opted for extended culture and blastocyst transfer, we could still have embryos available for freezing at that stage, but the numbers would be lower, and fewer patients would have excess embryos. It might be argued, nevertheless, that the results should be assessed in the same way, and would be directly comparable. If blastocyst transfer in selected patients were to result in a lower clinical pregnancy rate per cycle, but avoid multiple pregnancies and achieve a higher percentage of babies born per embryo transfer, then it must be considered a successful technique, in the same way as early embryo cryopreservation is successful in achieving high cumulative pregnancy rates per cycle.

While it is important to monitor fertilization, implantation, and pregnancy rates from the quality assurance point of view, it must be remembered that success may be considered in other ways: to enable a couple to come to terms with their inability to produce normal embryos could also be considered a success, as could avoidance of a disastrous multiple pregnancy. Such events will never show up in our figures, however we adjust them.

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