REPLY TO GLEICHER AND BARAD:

Noninvasive preimplantation genetic testing may provide the solution to the problem of embryo mosaicism

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We thank Drs. Gleicher and Barad for their interest in our recent paper, in which we report a higher reliability of noninvasive preimplantation genetic testing for aneuploidy (niPGT-A) in spent medium compared with conventional trophectoderm (TE) biopsy (1). However, we respectfully disagree with their comments.

The main points raised by Gleicher and Barad are that PGT-A, whether invasive or not, is not a useful test for ploidy because correction of chromosomal errors occurs downstream of blastocyst formation, and that this accounts for the high incidence of false positives associated with TE biopsy (2). We disagree that "self-correction" largely explains the high false-positive rate with the TE biopsy approach.

First, embryo aneuploidy arises from both meiotic and mitotic segregation errors. The age-related increased incidence of meiotic errors in human oocytes (3) is consistent with the maternal age-related increased incidence of spontaneous abortion (4). Indeed, it is generally accepted that meiotic aneuploidies are a primary cause of miscarriage and infertility (5). Meiotic aneuploidy is not "in flux" and is unlikely to be corrected by "self-correction."

Second, mitotic errors also occur and give rise to mosaic embryos (6). Such mitotic errors may undergo correction, presumably by apoptosis, as evidenced by our observation that cell-free DNA in spent culture media of euploid embryos sometimes exhibits abnormal DNA copy numbers (1). If such mitotic error correction is complete, then Gleicher and Barad's point regarding embryo ploidy "self-correction" might have validity. However, to our knowledge, the mitotic correction yield in human embryos is usually as low as 0 to 17.2% and is at most 40% (reviewed in ref. 7). Given the dominating meiotic error and the relatively low yield of correction from mitotic error, we argue that Gleicher and Barad (2) are unwarranted to dismiss diagnostic ploidy testing at the blastocyst stage as "largely irrelevant."

All who are using PGT-A with TE biopsy encounter false positives. In our study (1), we provide a solution to reduce the false-positive rate: by circumventing the mosaicism problem associated with TE biopsy.

Our approach assumes that cells in both the ICM and TE undergo apoptosis during preimplantation development (8, 9), resulting in DNA leakage into the culture medium. Gleicher and Barad (2) argue that the leaked DNA should primarily be TE-derived based on the higher number of cells in the TE compared with ICM. We counter this position with the observation that in chimeric mosaic mouse embryos aneuploid cells in the ICM were actively eliminated by apoptosis at a frequency 10-fold that of aneuploid cells in the TE (10). Therefore, we maintain that the most likely explanation for the false positives in PGT-A with TE biopsy is mosaicism, rather than self-correction, and that the false positives can be reduced by niPGT-A.

niPGT-A is a rather new approach (11) which requires further validation and is not intended to be a diagnostic test. It is a rule-in test as opposed to a rule-out test. Without touching the embryos, the test shows promise to identify euploid embryos for transfer with significantly reduced risks of false-positive and false-negative calls.

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