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EVALUATION OF DEVELOPMENTAL POTENTIAL IN EARLY EMBRYOS WITH MULTINUCLEAR BLASTOMERES USING CONFOCAL LIVE-CELL IMAGING

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OBJECTIVE: Multinuclear blastomeres (MNB) are often observed in human IVF embryos at the cleavage stage. The development of a confocal imaging system that includes an embryo culture system has made it possible to obtain time-lapse images of nuclear dynamics. In this study, we investigated the relationship between the presence of MNB in 2- and 4-cell stage embryos and their blastocyst formation using the confocal imaging system.
MATERIALS AND METHODS: Forty-six frozen-thawed pronuclear embryos intended for disposal were used after obtaining the informed consent of the patients and the approval of the Japan Society of Obstetrics and Gynecology research ethics committees. A mixture of mRNAs encoding enhanced green fluorescent protein coupled with α-tubulin and monomeric red fluorescent protein I fused with histone H2B was injected into the cytoplasm of the pronuclear embryos using a Piezo-drive manipulator. The pronuclear embryos were cultured in KSOM^{AA} medium under an atmosphere of 5% O_2, 5% CO_2 and 90% N_2. Time-lapse images were captured at 15-minute intervals until the day-5 stage using an all-in-one confocal imaging system.
RESULTS: Of the 41 cleavage stage embryos, MNB was observed in 32 embryos (78%) at the 2-cell stage and 27 (66%) at the 4-cell stage. At the 2-cell stage, the rate of embryos with MNB that developed to the blastocyst stage (34%, 11/32) was not significantly lower than that of embryos without MNB (67%, 6/9). However, in the 4-cell stage embryos, a significantly lower

blastocyst formation rate was observed for embryos with MNB (19%, 5/27), compared with the rate for embryos without MNB (86%, 12/14). The MNB observed in 9 of the 2-cell stage embryos had disappeared when the embryos were observed at the 4-cell stage.

CONCLUSION: The incidence of embryos with MNB was shown to be relatively high, particularly at the 2-cell stage. However, our data suggest that the presence of MNB at the 4-cell stage is closely associated with the embryo developmental potential.