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Topic category: Clinical science
Topics: Embryology (incl. IVF/ICSI, gamete and embryo selection, culture, cryopreservation, vitrification, developmental biology)
Abstract title: Does growth retardation in human blastocysts decrease implantation potential after embryo transfer through an increase of the abnormal spindles?

Authors: Hashimoto, S.; Amo, A.; Ito, K.; Nakaoka, Y.; Morimoto, Y.
IVF Namba Clinic

Study question: To assess the potential of growth-retarded embryos, the implantation potential and the spindle shape of vitrified–warmed blastocysts were assessed among normally developing and growth-retarded blastocysts.
Summary answer: The incidence of abnormal spindle morphology increased and the implantation competence decreased following vitrification in growth-retarded embryos compared with normally developing embryos, but there were no significant differences in the chromosomal abnormalities of abortuses and the incidences of peromelus of babies between 2 groups.
What is known already: There are conflicting data on whether the human embryo growth rate affects the outcome of vitrified–warmed blastocyst transfer. Various types of spindle abnormality occur in human cleavage- and blastocyst-stage embryos. A recent systematic review and meta-analysis concluded that growth-retarded embryos that develop to the blastocyst stage by day 6 have the same implantation potential as their day 5 counterparts if the morphology of day 6 blastocyst is similar to that of day 5 blastocyst.
Study design, size, duration: This was a retrospective cohort study including 878 single vitrified–warmed blastocyst transfers between January 2010 and July 2012, and an experimental study using 108 vitrified–warmed blastocysts donated to research. The local IRB of IVF Namba clinic approved this study. Data were compared using the Mann–Whitney nonparametric *U*-test.
Participants/materials, setting, methods: In a clinical study, we compared the

implantation rates of vitrified–warmed embryos that developed to the blastocyst stage on day 5 after insemination with those that required culture to day 6. In an experimental study, vitrified blastocysts were immunostained with an anti-α-tubulin antibody, an anti-γ-tubulin antibody and DAPI.

Main results and the role of chance: In the clinical study, the implantation rate of growth-retarded embryos (47%, *n:* 270) was significantly lower (P<0.05) than that of normally developing embryos (57%, *n:* 608). However, there were no differences in the chromosomal abnormalities of abortuses and the incidences of peromelus of babies between 2 groups. In the experimental study, a total of 533 spindles were analyzed in both day 5 and day 6 blastocysts. Confocal image analysis was accomplished by capturing a z-series stack of 0.5-μm-thick optical sections encompassing the entire blastocyst. Only spindles with fusiform poles and with chromosomes aligned at the equator were classified as normal. The incidence of abnormal spindles in growth-retarded embryos (47%, *n:* 274) was significantly higher (*P*<0.01) than in normally developing embryos (30%, *n:* 259).

Limitations, reasons for caution: Further studies are required to clarify the link between an increase in abnormal spindle formation and a decrease in embryonic implantation potential.

Wider implications of the findings: This study provided new insights on the implications of an increase in abnormal spindle formation in growth-retarded human blastocysts.

Study funding/Competing interest(s): Part of this work was supported by a grant from the Japan Society for the Promotion of Science (JPS-RFTF 23580397 to S.H.). No other competing interests are declared.

Key words: blastocyst formation, growth retardation, embryo transfer, pregnancy rate, spindle