Commentary: when embryos hit the brakes

If we have gleaned any one general principle from the time-lapse literature with regard to embryo selection, it is this: successful outcome from the transfer of a blastocyst depends somewhat upon it reaching that stage but even more so upon how and when it was reached. The simple visualization of an emerging fluid-filled space in a morula belies the underlying workings of a myriad of ordered molecular interactions. Without a doubt, blastulation is a complex process, with the resulting blastocyst representing the successful derivation of divergent cell lineages, a culmination of an ordered sequence of events that begins with the "start signal," compaction, in mammalian embryos. Reviewed elegantly by Sozen et al. (1), this process begins as junctional complexes form at apicolateral and lateral sites on blastomeres, followed by polarization within the outer cells. From mouse studies, it is clear that cell polarity proteins (Par 1, Par 3, aPKC, Jam 1, Ezrin) along with transcription factors, such as Cdx2, play critical mechanistic roles in this process. In fact, mouse knockout studies reveal a complex genomic interplay, indicating that both maternal and zygotic Cdx2 expression are required for blastocyst formation from morulae (2). Similar processes are present in human embryogenesis as well, but blastocyst formation from the initiation of compaction is an inefficient process, at least in contemporary culture systems, and one that either slows or even fails on a somewhat regular basis. For example, Iwata et al. (3) used time-lapse cinematography to observe thawed human embryos, cryopreserved at early stages, and saw that compaction was initiated at the eight-cell stage in 86.1% of embryos. Of these, only approximately half developed into good-quality blastocysts. If compaction began earlier, as it did in 13.9% of embryos, the results were even worse, with only 18.8% forming good-quality blastocysts.

Determining why this happens is not easy, even with tools for the dynamic study of early embryogenesis, such as time-lapse microscopy. As Kirkegaard et al. have shown (4), variation in the temporal events of development is complicated, in that it is most likely caused by a combination of factors, not a single one, and is highly dependent upon patient factors; thus, embryo origin is a major confounder in the timing of preimplantation embryogenesis. As an example, they note that the morphokinetic parameters of time to initial blastocoel formation and time to full blastocoel formation are both affected by FSH dose and by the number of prior IVF attempts. Patient age was also seen to influence blastulation: time to initial blastocoel formation occurred significantly later with older age. Just how extrinsic factors impinge upon intrinsic mechanisms governing blastocoel formation remains a mystery.

Perhaps with time our understanding of the requirements for the successful determination and differentiation of trophectoderm and inner cell mass lineages will allow for intervention should they falter—after all, why not "precision embryology"? As it is now, however, we are faced with decisions regarding what to do with slowly developing embryos, ones that often are all that a patient has available after IVF. What to do—discard, fresh transfer, culture further, culture further and transfer, culture further then vitrify? This is the dilemma addressed in the current issue of the journal by Haas et al. (5), who examined the developmental fate of delayed morulae, ones forming on day 5 vs. cavitating morulae formed on day 5. How significant an event is cavitation to further development and viability if embryogenesis is delayed? As the authors demonstrate in this retrospective analysis of a large number of delayed embryos, it depends and is not necessarily all that intuitive. Stopping short with a delayed embryo on day 5 when another 24 or even 48 hours of culture time is available for the embryo to continue development is not the likely strategy of many assisted reproductive technology programs. However, Haas et al. (5) compared the pregnancy rates obtained from day-5 fresh transfer between delayed morulae and cavitating delayed morulae, seeing no statistical difference. Yet when each of these was cultured further until day 6 for potential vitrification, the delayed morulae produced significantly fewer blastocysts, only one in five, and approximately half the number compared with those cavitating on day 5. The resulting blastocysts seemed to be of equivalent quality when vitrified and transferred at a future date because the pregnancy rates were similar between the two groups. The authors quite reasonably conclude that fresh transfer of delayed morulae is the prudent alternative to continued culture and vitrification, a valuable piece of information derived from good numbers.

Is there a larger lesson to be gained? Likely so, because it is nearly a universal strategy in embryo culture to give a subpar embryo all the time that is available to become what we think is acceptable. That pregnancies are obtained from the transfer of embryos cryopreserved on day 7 of culture attests to it being a seemingly reasonable approach. Perhaps it is not so in all cases, however, and the timing of cavitation may indeed be one marker that portends embryonic fate upon continued culture vs. stopping short with fresh transfer. This places embryologists in a similar professional situation as musicians, trapeze artists, and demolition experts, whereby decisions regarding timing are everything. Embryos that gently apply the brakes may be very different entities from those that do so abruptly, and our laboratory and clinical decisions should be informed by them rather than assuming that additional time in culture yields additional quality.

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https://doi.org/10.1016/j.fertnstert.2018.09.017

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