

Blastocyst versus early cleavage embryo transfer: A retrospective analysis of 4,165 transfers

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Summary

Purpose: To compare the clinical outcomes after the transfer of blastocysts versus early cleavage embryos in assisted reproduction technologies (ART).

Methods: A retrospective analysis of all the ovarian stimulation-in vitro fertilization-embryo transfer cycles performed at the Centre for Human Reproduction, Athens, Greece, between June 1997 and December 2001.

Results: The number of blastocysts transferred per ET was significantly lower compared to that of all early cleavage embryos. The implantation rate of blastocysts was significantly higher compared to that of all other modes of transfer. Clinical pregnancy rate after the transfer of blastocysts was significantly increased compared to that after transfer of any early cleavage embryo. The viable pregnancy rate after the transfer of blastocysts was significantly increased only compared to that after the transfer of day-2 embryos. There were no significant differences regarding the multiple gestation rates among the various modes of transfer.

Conclusion: The use of blastocysts in ART is beneficial when compared to that of day-2 embryos and at least comparable to that of day-3 embryos. Blastocyst culture and transfer remains a favourable and promising option in ART.

Key words: Assisted reproduction technologies; In vitro fertilization; Blastocyst.

Introduction

Having in mind the miraculous process of natural conception in humans where implantation occurs at the embryonic stage of a blastocyst, culture and replacement of day 2 or 3 embryos in assisted reproduction technology could be theoretically disadvantageous. Oviducts, the natural environment for two to eight cell embryos, provide a different nutritional and homeostatic environment compared to that of the uterus, whereas the ideal implantation signaling is achieved by the synchronization between the blastocyst and a properly receptive endometrium. As a consequence, the challenge of extended embryo culture and transfer at the blastocyst stage was attempted quite early in ART [1, 2] but the lack of proper culture media resulted in poor rates of embryo development to blastocysts. Co-culture of embryos with somatic cells somehow improved the outcome [3, 4] but the innovation came with the introduction of the sequential culture media [5], which follows the metabolic needs of the pre- and post-compaction embryos by mimicking the altering environment of the natural host of the culturing embryos.

Materials and Methods

Having adopted the option of blastocyst culture and transfer since 1997, we retrospectively analyzed the outcome of all the ovulation induction-embryo transfer cycles performed at the Centre for Human Reproduction, Athens, Greece, between June 1997 and December 2001. All oocytes and embryos were cul-

tured in sequential media from Vitrolife (IVF-20, G1.2 and G2.2, Scandinavian IVF Science AB, Göteborg, Sweden). Criteria for adopting the choice of embryo-transfer at the blastocyst stage were flexible including i) couples with two or more previous ART failures after the transfer of early cleavage embryos, ii) cycles with 15 or more oocytes per retrieval and iii) couple's request after discussion about the possible benefits as well as the risks of culture arrest. Statistical analysis was performed with the use of Pearson's chi-square test and the Student's t-test.

Results

Table 1 summarises the outcomes after the transfer of blastocysts and early cleavage embryos. During the above period of time, 15,490 embryos (9,372 of which resulted after ICSI) were transferred in 4,165 transfers (ET), including 3,642 blastocysts in 1,122 ET (3.22 ± 1.33 embryos per ET), 5,076 day-2 embryos in 1,402 ET (3.60 ± 1.39 embryos per ET), 1,129 day-2 embryos with assisted hatching in 285 ET (3.89 ± 1.61 embryos per ET), 3,353 day-3 embryos in 832 ET (4.07 ± 2.11 embryos per ET) and 2,290 day-3 embryos with assisted hatching in 524 ET (4.44 ± 1.65 embryos per ET). Scoring of early cleavage embryos revealed no overall significant differences concerning their quality. Regarding blastocyst culture outcome, from the initial 14,170 two-pronucleate embryos, 5,283 reached the blastocyst stage (blastulation rate 37.3%); of them 3,642 (68.94%) were transferred and 1,641 (31.06%) were frozen. There were no significant differences regarding the age of women receiving day-2, day-3 and blastocysts (mean \pm SD: 33.4 ± 4.1 , 32.8 ± 5.4 , 33.1 ± 4.4 , respectively) as

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Table 1. — Outcome after the transfer of blastocysts and early cleavage embryos.

	Blastocysts	Day-2 embryos with A/H ^a	Day-2 embryos with A/H ^a	Day-3 embryos	Day-3 embryos
No. of embryos	3642	5076	1129	3353	2290
No. of transfers	1122	1402	285	832	524
Embryos/transfer	3.22±1.33	3.60±1.39	3.89±1.61	4.07±2.11	4.44±1.65
Clinical pregnancies	420	348	67	271	158
Clinical pregnancy rate (%) ^b	37.43	24.82	23.50	32.57	30.15
Viable pregnancies	319	274	62	227	133
Viable pregnancy rate (%) ^b	28.43	19.54	21.75	27.28	25.38
Singleton pregnancies	217	196	50	151	98
Twin pregnancies	72	53	10	55	21
Higher order pregnancies	30	25	2	21	14
Singleton pregnancy rate (%) ^c	68.02	71.53	80.64	66.5	73.68
Twin pregnancy rate (%) ^c	22.57	19.34	16.12	24.22	15.78
High order pregnancy rate (%) ^c	9.40	9.12	3.22	9.25	10.52
Implantation rate (%)	12.38	7.42	6.73	9.66	7.94
Abortions	75	62	4	34	23
Abortion rate (%) ^b	6.68	4.42	1.40	4.08	4.38
Ectopic pregnancies	26	12	1	10	2
Ectopic pregnancy rate (%) ^b	2.31	0.85	0.35	1.20	0.38

^a Assisted Hatching; ^b per 100 transfers; ^c per 100 viable pregnancies.

well as regarding the cause of infertility. The number of blastocysts transferred per ET was significantly lower compared to that of all early cleavage embryos ($p < 0.0001$). Implantation rates (sacs per 100 embryos), clinical pregnancy rates (per 100 ET) and viable pregnancy rates (pregnancies beyond the 1st trimester per 100 ET) were: 12.38%, 37.43% and 28.43%, respectively in the blastocyst group, 7.42%, 24.82% and 19.54% in the day-2 group, 6.73%, 23.50% and 21.75% in the day-2 with assisted hatching group, 9.66%, 32.57% and 27.28% in the day-3 group and 7.94%, 30.15% and 25.38% in the day-3 with assisted hatching group. Statistics proved that the implantation rate of blastocysts was significantly higher compared to that of all other modes of transfer ($p < 0.0001$). Clinical pregnancy rate after transfer of blastocysts was significantly increased compared to that after transfer of any early cleavage embryo (blastocysts vs day-2 embryos ± assisted hatching: $p < 0.0001$, blastocysts vs day-3 embryos ± assisted hatching: $p < 0.05$). The viable pregnancy rate after transfer of blastocysts was significantly increased only compared to that after the transfer of day-2 embryos ($p < 0.0001$). The transfer of day-3 embryos resulted in the highest rate of multiple gestations (33.47 per % viable vs 31.61% in the blastocysts group) but statistics proved that there were no significant differences regarding the multiple gestation rates among the various modes of transfer for the particular number of embryos per transfer. Regarding abortions, the highest rate was observed after the transfer of blastocysts (6.68 per 100 transfers), being significantly increased compared to that after the transfer of day-2 and day-3

embryos ($p = 0.01$). The transfer of blastocysts resulted also in a significantly increased rate of ectopic pregnancies compared to that after the transfer of day-2 embryos with or without assisted hatching and day-3 embryos with assisted hatching.

Discussion

Reviewing the medical literature, the results from the use of blastocysts remain debatable. It seems that when traditional culture media is used, blastocyst culture and transfer does not improve implantation and viable pregnancy rates, whereas it is possibly linked to increased risks of transfer cancellation and decreased rates of cryopreservation [6-8]. On the other hand, the use of sequential media for the blastocyst culture has been related to increased implantation rates [9-12], decreased number of transferred embryos [9, 11, 12], decreased rate of multiple gestations [11, 12] and increased clinical pregnancy rates per transfer [13]. A critical review of our results, achieved with the use of sequential culture media, revealed that the selective transfer of blastocysts is definitely beneficial when compared to that of day-2 embryos and at least comparable to that of day-3 embryos (similar viable pregnancy rates and higher implantation and clinical pregnancy rates). Current trends towards reduction in the number of transferring embryos, running research on the quality criteria of early cleavage embryos along with the rapid development in the area of pre-implantation genetic diagnosis (application of which seems to be beneficial at the blastocyst stage), keep the blastocyst culture and transfer a favorable and promising option in ART.

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