RACING TO DAY 4: EARLY BLASTOCYST FORMATION IS LINKED TO SUPERIOR EMBRYO QUALITY AND HIGHER EUPLOIDY RATES

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Background: Limited research is available specifically detailing the proportion of human embryos that develop into expanded blastocysts by Day 4 of development. However, studies on accelerated embryo development have shown that a small percentage of embryos can reach the blastocyst stage this early, typically representing an outlier group in standard IVF embryo culture system procedures. Most embryos tend to reach the expanded blastocyst stage by Day 5 or 6, which is considered the optimal window for development. While these early Day 4 blastocysts do exist, their occurrence is not common, and research into their viability and associated euploidy rates is still ongoing.
Objective: This study aims to determine the proportion of human embryos that develop into expanded blastocysts by Day 4 and assess their development quality and euploidy rates compared to those reaching the blastocyst stage on Days 5 -7.
Material and Methods: This retrospective analysis from November 2023 – September 2024 included data from 2,839 autologous expanded blastocyst embryos cultured in Global Total media (Cooper Surgical) and incubated in an Embryoscope Plus time lapse incubator (Vitrolife). Expanded blastocyst development timings (hours post insemination [HPI]) were stratified into 5 groups: (90–109 hours (late Day 4 [D4+]), 110–119 hours (early day 5 [D5-]), 120–129 hours (late day 5 [D5+], 130–139 hours (early day 6 [D6-]), and 140–169 hours (late day 6 and day 7 [D6+/7]). Comparisons of blastocyst development timing rates, euploidy rates and the quality of inner cell mass (ICM) and trophectoderm (TE) were statistically analyzed using a Chi-squared test and adjusted using a Bonferroni correction.
Results: Distribution rates of expanded blastocyst formation were 9%, 39%, 29%, 17% and 6%, at D4+, D5-, D5+, D6- and D6+/7, respectively ($p<0.001$). The data in Table 1 illustrates the distribution of expanded blastocyst formation based on patient age with younger patients showing a higher likelihood of earlier expanded blastocyst formation ($p<0.001$) and age a shift toward slower expanded blastocyst formation as maternal age increases. Euploidy rates of expanded blastocyst formation were 62%, 52%, 39%, 28% and 21% at D4+, D5-, D5+, D6- and D6+/7, respectively ($p<0.001$). Figures 1 and 2 illustrate that faster developing embryos also showed better morphokinetic qualities, with a higher proportion of grade A ICM and TE cells ($p<0.001$).

Table 1. Distribution rates of expanded blastocyst formation based on age

SART Age Groups	Timing					Total count
	D4+	D5-	D5+	D6-	D6+/7	
<35	13% (130)	43% (446)	25% (260)	15% (153)	4% (37)	1026
35-37	8% (61)	39% (283)	30% (217)	16% (119)	6% (42)	722
38-40	5% (33)	34% (217)	32% (204)	21% (135)	8% (51)	640
41-42	8% (20)	35% (92)	36% (95)	17% (44)	3% (9)	260
>43	6% (11)	33% (63)	30% (58)	19% (37)	11% (22)	191

Figure 1. Distribution of ICM Grade based on Expanded Blastocyst Formation Timing

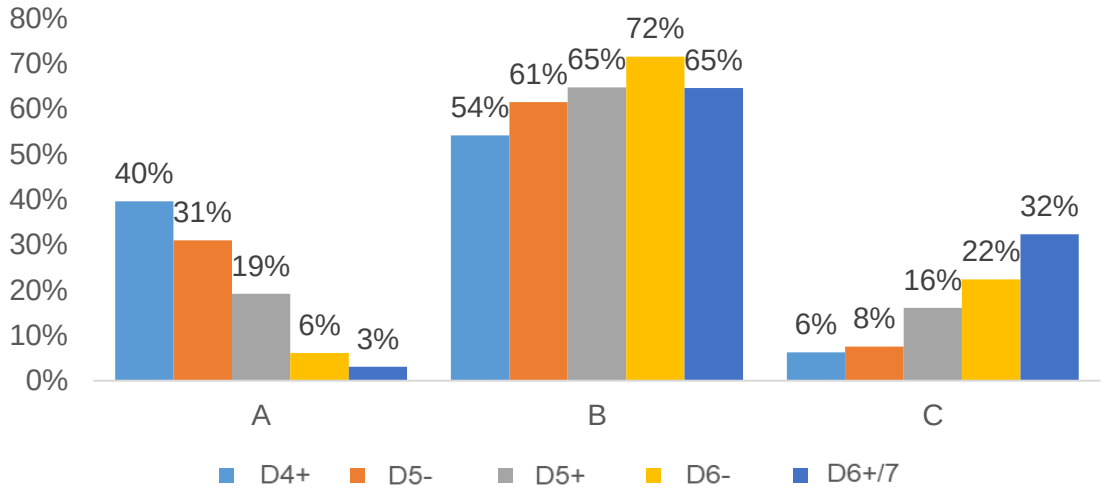
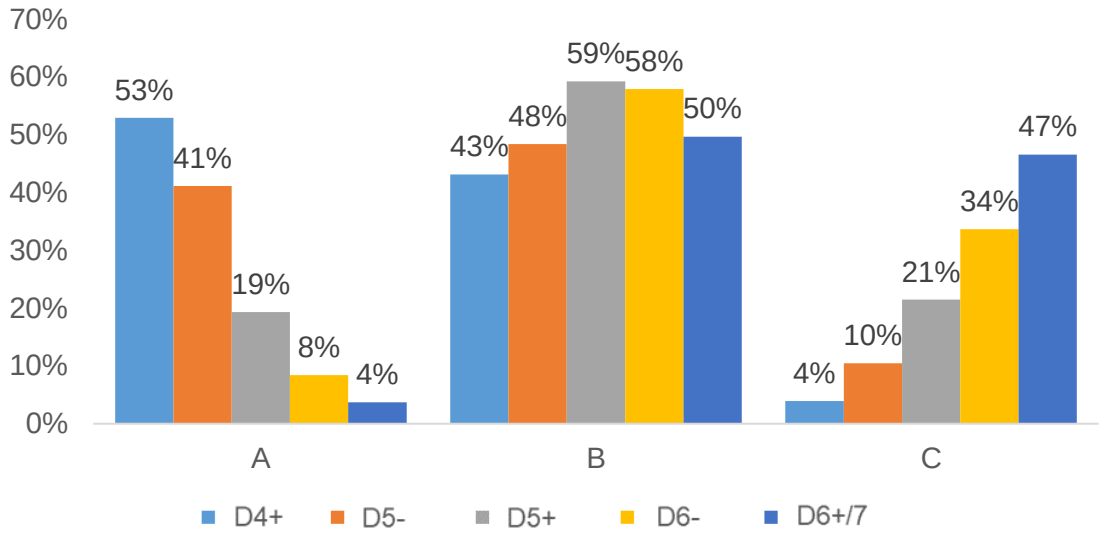


Figure 2. Distribution of Trophectoderm Grade based on Expanded Blastocyst Formation Timing



Conclusion: Earlier blastocyst formation is linked to better embryo quality based on ICM and TE morphology with a lower risk of chromosomal aneuploidies. The data also illustrates that younger patients show a higher likelihood of earlier expanded blastocyst formation and as maternal age increases a shift toward slower expanded blastocyst formation. These findings highlight the potential advantages of selecting day 4 embryos over later developing embryos. This can lead to an improved embryo selection criteria in IVF protocols which could lead to higher success rates.

Support: None

References: Cimadomo, Danilo et al. Day7 and low-quality blastocysts: opt in or opt out? A dilemma with important clinical implications. Fertility and Sterility, Volume 120, Issue 6, 1151 - 1159