Statistics-enabled numerical blastocyst grading system: A proposal for debate and adoption

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Abstract

The authors have proposed a numerical statistics-enabled modified method of blastocysts grading that overcame the deficiency and paucity of a grading system that could be used for statistical analyses and calculations. The alphanumeric blastocyst grading system described previously in 1999 was modified by one of us to a statistics-enabled numerical blastocyst grading system in 2014. It allowed the statistical comparison of the quality of human blastocysts generated between different treatments, primarily for quality management and research. This mathematical modification of the alphanumeric grading system was used effectively since 2014 at the University of Malaya Fertility Centre, Kuala Lumpur, Malaysia, as a Standard Operating Procedure (SOP) in their routine IVF Laboratory practice. This method of grading is also useful in the selection of embryos for transfer and or cryopreservation in clinical application. With this numerical grading system, it is possible to statistically compare the qualities of human blastocysts generated between different treatments. This grading system could be very useful in assisted reproduction technology and reproductive biology as it is needed and will be very useful to statistically compare the quality of blastocysts generated between treatments for purposes of quality management and for reporting outcomes of research studies. The scores range from4 (excellent) to 1 (poor). It is versatile as it can be applied in the form of as split scores (eg: a split score 4/4/4 denote excellent blastocyst/excellent inner cell mass/excellent trophectoderm) or expressed as cumulative scores or as summarized scores explained in Chart 2, both for embryo selection as well as for statistical analyses.

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Background

Several methods of assessing blastocyst quality have been employed over the years (Gardner and Leese, 1999). At the present times, the most common method of blastocyst grading that appears to have stood the test of time is that based on morphology, and the most widely used method of grading based on morphology is that described by Gardner and Schoolcraft (1999). This method of grading is useful in the selection of embryos for transfer and or cryopreservation in clinical application. However the alphanumeric nature of this grading system does not lend itself to statistical calculations or analyses. A statistics-friendly grading system is needed to objectively compare groups of blastocysts, especially those that developed following different treatment protocols. In the early years of blastocyst culture a statistical method that could be used to compare the quality of blastocysts generated between different treatments was not available.

To overcome this deficiency, one of us (Ali, 2014) modified Gardner and Schoolcraft's

alphanumeric blastocyst grading system to a statistics-enabled numerical blastocyst grading system in 2014 which allowed the statistical comparison of the quality of human blastocysts between treatments, primarily for quality management and research. Ali's mathematical modification of Gardner and Schoolcraft's alphanumeric grading system was used effectively since 2014 at the University of Malaya Fertility Centre, Kuala Lumpur, Malaysia (Ali, 2014) as a Standard Operating Procedure (SOP) in their routine IVF Laboratory practice. With this numerical grading system, it is possible to statistically compare the qualities of human blastocysts generated between treatments.

Discussion

This numerical system is anticipated to further enhance selection of blastocysts with the high probability of implantation and pregnancy, and for cryopreservation. This grading method uses the same principle as the modified grading method for statistical analyses of cleavage stage embryos previously described by Ali and coworkers (Ali et al., 2000). The authors have considered the proposed numerical statistics-enabled modified method of blastocysts grading described by Ali (Ali, 2014) in detail and concluded that this numerical method of blastocyst grading overcame the deficiency and paucity of a grading system that could be used for statistical analyses and calculations. Such a grading system could be very useful in assisted reproduction technology and reproductive biology. The authors are not aware of any other statistical method available for differentiating the quality of human blastocysts generated between different treatments statistically. This statistics-enabled blastocyst grading method is needed and will be very useful to statistically compare the quality of blastocysts generated between treatments for purposes of quality management and for reporting outcomes of research studies.

We present here in chart form the salient features of the numerical blastocyst grading system with which statistical comparisons could be made. In the present method the blastocyst is graded 4 (excellent), 3 (Good), 2 (average) and 1 (poor) [similar to that of the method described for days 2, and 3 embryos [Ali et al., 2000]. It is versatile as it can be used as split scores which is more descriptive of the quality of the blastocyst. The split score describes the qualities or grades of the entire morphology of the blastocyst, its inner cell mass and trophectoderm individually, giving a clear picture of the state of the blastocyst (eg: a split score of 4/4/4 is indicative of an excellent blastocyst/with an excellent inner cell mass/ excellent trophectoderm). It can also be expressed as cumulative scores 12 (excellent) to 3 (poor) or as summarized scores explained the key (Chart 2), both for embryo selection as well as for statistical analyses. The summarized score of 4 (excellent) to 1 (poor) can further be converted to the reversed numerical score of 1 (excellent) to 4 (poor) or an alphatical score: A (excellent) to D (poor) for ease of comprehension by clinical staff.

Reference

Ali J. Standard operating procedures for cIVF, embryo grading & embryo transfer. Reproductive Health Clinic, University of Malaya. Kuala Lumpur, Malaysia. September 03, 2014.

Ali J, Shahata MA, Al-Natsha SD. Formulation of a protein-free medium for human assisted reproduction. Hum Reprod. 2000;15(1):145-56

Al Helou, Ali J. Comparative development of human 1PN and 3PN day 5 blastocysts cultured in Global® and synthetic Synbios® media. Jeddah, KSA, 2025

de la Fuente R, King WA. Use of a chemically defined system for the direct comparison of inner cell mass and trophectoderm distribution in murine, porcine and bovine embryos. Zygote. 1997;5(4):309-320.

Gardner DK, Schoolcraft WB. In vitro culture of human blastocysts. In: Jansen R, Mortimer D, editors. London: Parthenon Publishing; 1999. p. 378–88

Godke RA. 1993. "The mouse blastocyst is most likely viable if there are at least 4 cells in its ICM". Personal communication.

Pons MC, Carrasco B, Rives N, Delgado A, Martínez-Moro A, Martínez-Granados L, Rodriguez I, Cairó O, Cuevas-Saiz I; SIG Embryology of ASEBIR. Predicting the likelihood of live birth: an objective and user-friendly blastocyst grading system. Reprod Biomed Online. 2023;47(3):103243

Chart 1: Statistics-enabled blastocyst grading system

Volume/ Expansion Grade/ Score	Salient Features of Blastocyst Morphological Development	Day 5 Blastocyst Quality Evaluation
1	Blastocoel cavity less than half the volume of the embryo	Poor
2	Blastocoel cavity equal to or more than half the volume of the embryo	Average
3	Full blastocyst, cavity completely filling the embryo, larger than its original volume	Good
4	Expanded blastocyst, cavity larger than the original volume of the embryo, thinning zona	Excellent
Scores	**Salient Features of Blastocyst development Beyond Day 5**	**Days 6/7**
4^{+}	Hatching expanded blastocyst, thinning zona, larger than the original volume of the embryo	Excellent
4^{+}	Completely hatched expanded blastocyst, thin zona, larger than original volume of embryo	Excellent
	$^{+}$The highest grade attainable is 4	
ICM Grade/ Score	**Quality of Inner Cell Mass [de la Fuente et al.,1997]**	
4	Large ICM, many cells, tightly packed ; rough visual estimate >25 cells	Excellent
3	Moderate size ICM, several cells, tightly packed; rough visual estimate >11-24 cells	Good
2	Small ICM, few cells, loosely packed; rough visual estimate >5-10 cells	Average
1	Very small or no ICM cells, 0 to 4 cells; rough visual estimate <4 cells** **(Godke, 1993)**	Poor
TE Grade/ score	**Quality of Trophectoderm (TE) [de la Fuente et al., 1997, Pons et al. 2023]**	
4	Many cells, tightly packed; cells in focal plane >20 cells	Excellent
3	Moderate number of cells, tightly packed, cells in focal plane >11-19 cells	Good
2	Several to few cells (less than 10 cell in visual focal plane) 5-10 cells	Average
1	Very few (less than 4 cells in visual focal plane) <4 cells	Poor

Explanation: The blastocyst is split scores or graded, eg: 4+4+4 (or 4/4/4); Excellent
or given cumulative scores, eg: (4+4+4) = 12; Cumulative Grade); Excellent
Or given summarized / calculated score/ grade, eg: 12/3 = Grade 4 ; Excellent

Chart 2: Key to Chart 1 for the statistics-enabled numerical blastocyst grading system

Statistics-enabled split score for blastocyst development + ICM +TE		Statistics-enabled cumulative whole blastocyst score		Summarized statistics-enabled score for entire blastocyst		Numerical and alphabetical grading for routine clinical application	
Split Grade	*Blastocyst Quality*	*Cumulative Grade*	*Blastocyst Quality*	*Calculated Grade*	*Blastocyst Quality*	*Numerical*	*Alphabetical*
4+4+4	Excellent	10-12	Excellent	4	Excellent	1	A
3+3+3	Good	7-9	Good	3	Good	2	B
2+2+2	Average	4-6	Average	2	Average	3	C
1+1+1	Poor	3	Poor	1	Poor	4	D