Early cleavage of human embryos to the two-cell stage: A simple, effective indicator of implantation and pregnancy in intracytoplasmic sperm injection.
Early cleavage of human embryos to the two-cell stage: A simple, effective indicator of implantation and pregnancy in intra cytoplasmic sperm injection.
APPENDIX – 2

INFORMED CONSENT

“Early cleavage of human embryos to the two-cell stage: a simple, effective indicator of implantation and pregnancy in IVF / Intracytoplasmic sperm injection”

Name of the principle investigator: Mrs.G.Manjula

Description of the study:

Purpose of the study

This research is studied

1. To find out the implantation rate when early cleavage embryos are used for transfer compared to late cleavage embryos
2. To find out the pregnancy rate when early cleavage embryos are used for transfer compared to late cleavage embryos

Voluntary participation

Inclusion criteria

1. All patients enrolled for IVF/ICSI during this study period will be included in the study.
2. All categories of female partners will be included in the study.
3. All categories (idiopathic, male factor, azoospermic) of male partners will be included in the study.
4. Patient having only early cleavage embryos will be included in the study.
5. Patient having only late cleavage embryos will be included in the study.
Exclusion criteria

1. Patient having both early & late cleavage embryos will be excluded in the study.

The study will be conducted in accordance with the ethical principle and after obtaining approval from the Institutional Ethical committee clearance.

The following datas & observations will be collected during the study

1. Age of the partners
2. Duration of infertility
3. Semen analysis of male partner
4. protocol for stimulation
5. Doses of gonadotropins used
6. No of follicles
7. No of oocytes retrieved
8. No of Matured Oocytes (M II)
9. Method of choice (IVF/ICSI)
10. No of oocytes inseminated or injected
11. No of oocytes fertilized
12. Examination early cleavage at 27 hours after insemination /injection of oocytes
13. No of embryos transferred
14. No of embryos cryopreserved
15. hCG value after 14 days of transfer
16. No of sac seen
17. Implantation rate
18. Pregnancy rate

**Duration of participation**

We expect the duration between September 2010 to September 2012. This study will be conducted in the Fertility Unit, Sri Ramachandra Medical Centre for Assisted reproductive Technology, (SMART) Dept of Obstetrics & Gynaecology, Sri Ramachandra Medical College and Research Institute, Sri Ramachandra University, Porur, Chennai-116. The study protocol will follow ICMR Guidelines ART.

**No of participants**

The minimum sample required in each group is 51.

Patients will be divided into two groups to evaluate the pregnancy outcome.

Early-cleavage group - Patient having only early cleavage embryos for transfer.

Late cleavage group - Patient having only late cleavage embryos for transfer

However if more patients enrolled for IVF/ICSI with in the study period we will include in our study.
Appendix

Specific procedure:

- The patients who are attending infertility clinic at SRMC will be investigated and those patients who require IVF/ICSI will be included in the study.

- Two stimulation protocols will be used in this study; the GnRH agonist protocol or the GnRH antagonist protocol.

- In both groups, 10,000 IU hCG will be administered when at least two follicles reached 18 mm in diameter, and oocyte will be recovered transvaginally with ultrasound guidance 34-36 h after hCG administration.

- Motile sperm will be isolated and each oocyte will be inseminated with motile spermatozoa. For the ICSI procedure, washed spermatozoa will be injected into the oocyte using standard techniques. Embryos will be cultured in a CO2 incubator at 37°C.

- Normal fertilization will be confirmed by the presence of two pronuclei and two polar bodies 16–20 h (day1) after insemination for IVF or ICSI. On the same day, early cleavage examination will be performed at 27 hours after insemination. Embryos displaying two cells at inspection will be designated as 'early cleavage'. The embryos that had not yet cleaved to the 2-cell stage will be designated as 'late cleavage'.

- Two or three embryos will be transferred on Day2. The embryos that are not transferred are cry preserved.
• We will note the endometrial thickness for all patients.

• The luteal phase will be supported by vaginal supplementation or injection of 200 mg micronized natural progesterone three times a day.

• To assess treatment outcome, pregnancies will be reported as positive when beta hCG serum levels are >10 mIU/ml, 10 days post transfer and increased to four times that value on day 14 post transfer. Serum hCG will be measured 14 days after oocyte retrieval. An increase of serum hCG to levels above 50 IU/l indicated pregnancy. Pregnancy confirmed by sac seen in ultrasound 4-5 weeks of post oocyte retrieval.

POSSIBLE RISKS TO THE PARTICIPANT

This early cleavage examination of embryos does not cause any risk to the embryos and the participants.

POSSIBLE BENEFITS TO THE PARTICIPANTS

By examining the early cleavage, we can predict the embryo quality, capacity to implant, ability to produce the blast cyst etc., and predict the success rate.

We expect the early cleavage embryo implant at a rate 3 fold higher than the late cleavage embryo and the pregnancy rate will be double the rate in early cleavage embryo transfer than the late cleavage embryo transfer.

COST & PAYMENT TO THE PARTICIPANT

No cost for this research and no remunerations will be provided for the participants.
CONFIDENTIALITY

The information obtained in this study is strictly confidential. Your name will not be used in publications or conference presentations.

PARTICIPANT'S RIGHT TO WITHDRAW FROM THE STUDY

You have the right to refuse to participate in this study, the right to withdraw from the study and the right to have your data destroyed at any point during or after the study, without penalty.

VOLUNTARY CONSENT BY THE PARTICIPANTS

PARTICIPATION IN THIS STUDY IS COMPLETELY VOLUNTARY, AND YOUR CONSENT IS REQUIRED BEFORE YOU CAN PARTICIPATE IN THIS STUDY.

I have read this consent form (or it has been read to me) and I fully understand the contents of this document and voluntarily consent to participate in this study. All of my questions concerning this study have been answered. If I have any questions in the future about this study, they will be answered by the investigators listed below. I understand this consent ends at the conclusion of this study.
Early cleavage of human embryos to the two-cell stage: A simple, effective indicator of implantation and pregnancy in intra cytoplasmic sperm injection.

Appendix

CONTACT ADDRESS WITH PHONE NUMBER

Principle Investigator: From SRMC
Mrs. G. MANJULA.
Embryologist,
Sri Ramachandra Medical Centre for Assisted reproductive technology (SMART)
Department of Reproductive Medicine,
Sri Ramachandra University,
Ph: 044 -24765520, 24768403 ext: 626, 8984.
Mobile: 9840627333, 9043916442, 9840627444

By signing this form, I agree to participate in this study. A copy of this form has been given to me.

Date :

Husband’s signature:
Husband’s thumb impression
Wife’s signature:
Wife’s thumb impression
Witness name:
Witness signature:
CERTIFICATION OF INFORMED CONSENT

I certify that I have explained the nature & purpose of this study to the above named individual, and I have discussed the potential benefits of this study participation. The questions the individual had about this study have been answered & we will always be available to address future questions.

Date: Signature of person obtaining consent:

Name of Principle Investigator:

Signature of Principle Investigator:
Early cleavage of human embryos to the two-cell stage: A simple, effective indicator of implantation and pregnancy in intra cytoplasmic sperm injection.
Appendix

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**PROFORMA**

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### OOCYTE COLLECTION

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<th>Surgeon</th>
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<th>Time Start</th>
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<th>Duration</th>
<th>HCG-inj</th>
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<td>IVF/ICSI</td>
<td>Eggs Retrieved</td>
<td>M2</td>
<td>GV</td>
<td>Denu Time</td>
<td>Cervix</td>
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### PROCESSING DETAILS

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### SEMEN ANALYSIS

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<th>Date</th>
<th>Motility</th>
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<th>Morphology</th>
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<th>No of cells, Grade</th>
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### EMBRYO TRANSFER

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<th>Date</th>
<th>Donor Spemrs</th>
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<th>Ast Embryologist</th>
<th>Time eval</th>
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### TESA/TESE

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<th>Motility %</th>
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### APPENDIX PROFORMA

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| Day 2 | Day 3 | Date | Date | Blood Grp | FSH | LH | E2 | AMH | | |
|-------|-------|------|------|-----------|-----|----|----|-----| | |

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<th>Lut. Support</th>
<th>No. of cells</th>
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