How to improve my ART outcome
Quality Control / Quality Assurance (QA/QC)
Infertility is a disease of the reproductive system characterized by inability to achieve pregnancy after 12 or more months of regular unprotected sexual intercourse.

In this competitive era, IVF clinics maintaining high success rates can only flourish and one thing, which is central to this idea, is quality. Clinics, which can maintain quality in all aspects of patient care, laboratory and clinical practices will set an example for others to follow. It not only detects flaws in the system but also inspires us towards continuous improvement in results and patient satisfaction.

Keeping this in mind we have designed panel of Three Pan India workshops to create awareness about the quality management in infertility.

I am sure you would enjoy the meetings and reading the manual.

Dr M Gouri Devi
President - IFS

According to the World Health Organization, infertility is a disease of the reproductive organs and is defined, as the inability of sexually active couples taking no contraceptives to achieve pregnancy within 1 year.

The treatment of infertility involves many variables many of which are beyond our control. One aspect in our practice, which can help us control these variables, is the practice of total quality management. It not only consists of quality control and assurance but also involves taking appropriate corrective actions and adding value to the existing practice.

I sincerely hope that these programs on “How to improve ART practice” will be of immense use to ART practitioners.

I am sure you would enjoy these programs, which we have put up with great hardwork and sincerity. I am grateful to Origio India for their support in organizing these events.

Prof (Dr) Pankaj Talwar
Secretary General - IFS
I feel honored to be handed over the responsibility of conducting series of 3 meetings in Kolkata, Patna and Ahmedabad on “How to Improve my ART outcome (QA/QC)”.

I am extremely delighted to present before you this handbook on Quality Control and Assurance in Assisted Reproduction Technology.

“Quality is a Lousy Idea – if it is Only an Idea”

Quality management should be an integral part of any ART program’s DNA and good practices should be imbibed at spinal level. After achieving the said level of practice, one never feels burdened and quality becomes a way of life. The book is a compilation of series of lectures by renowned experts in the field where we focus on the art of ovarian stimulation and nitty gritty of techniques such as ovum retrieval, embryo transfer and vitrification.

This will be followed by session on proper documentation and audits, the information we derive from these and how to troubleshoot if reports are not satisfactory. We also look at practical aspects such as what patients go through while undergoing an IVF cycle and how we can improve ART dropout rate. In the end, we touch upon what we dread that is how to anticipate and be prepared for dealing with unexpected events in the IVF lab.

I am confident this book will be handy for anyone looking to improve clinical and lab practices.

I would like to thank President and Secretary, IFS for conceiving, organizing and overseeing the whole program. I would like to thank the contributors for taking time off and sending such comprehensive presentations.

I would also like to thank the Origio team for their support in organising the meetings and without which this would have been difficult to achieve. In the end, my sincere thanks to the local chapter secretaries for the extraordinary effort which they have put in.

Dr Sarabpreet Singh
National Coordinator
IFS & Origio QA/QC Initiative

- MD (AIIMS)
- Senior Consultant & Chief Embryologist at the Artemis Health Institute since 2009
- IFS and ESHRE-certified Senior Clinical Embryologist
- Convenor, IFS SIG (Clinical Embryology)
- Executive member of Academy of Clinical Embryologists, India
- Area of Interest: Cryobiology and Male Infertility
• Director, Gouri Hospitals Ltd.
• Director, Ridge IVF Group. (Runs a chain of IVF centres)
• President, Indian fertility society
• Ex-Secretary General, Indian Fertility Society
• Executive, AOGD governing council
• Member, Executive Board, NARCHI, DGES, FPSI
• Ex Vice President, NARCHI
• Chairperson, Advocacy & Ethics Committee, IFS.
• State Quality Assurance Committee (SQAC) Govt of NCT of Delhi.
• Member: MTP advisory committee, Govt Of NCT of Delhi
• Member Advisory committee on ethical practices in the field of obstetrics, Govt of NCT, Delhi
• Recipient of Kanak Goel Award 1995-1996 from IMA.
• Chairman’s Appreciation Award by IMA AMS – 2002
• Dr. APJ Abdul Kalam Excellence Award – 2017
• Economic Times Award one of the Most Inspiring Gynecologists of India

She is a keen academician, has organized many conferences, has been a speaker in many national and international conferences. Has many publications to her credit

• Sec IFS.
• Secretary Fertility preservation society of India.
• Editorial board of multiple Infertility journals.
• Member Advisory committee ICMR
• Member Infertility committee FOGSI
• Editor Nexus / Artext – E bulletin of IFS
• Awarded Vishisht seva medal by the President of India for working in field of infertility
• Associate Editor FSR
• Set up four centres for Armed forces .
• Experience of 10,000 and ET cycles .
• Member International society of fertility preservation.
• Trained Human Embryonic Stem Cell Derivation – Israel
• Trained in ovarian cortex freezing (fertility preservation) - Paris
• Trained in PGD – Germany, Spain
• Trained in QA/QC-Spain
• Edited 6 books

Dr M Gouri Devi
M.D

Col Pankaj Talwar, VSM
Professor and HOD
ART Centre, Army Hospital, New Delhi
Venue and Dates

AHMEDABAD
30th Sept, 2018

PATNA
23rd Sep, 2018

KOLKATA
2nd Sept, 2018

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Dr. Anita Singh
Organising Chairperson

Dr Suparna Bannerjee
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<th>Contributed by</th>
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<td>Individualized ovarian stimulation for IVF: finding a balance between cost, safety and outcome</td>
<td>Dr Jayesh Amin</td>
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<td>Is 100% outcome achievable in Vitrification?</td>
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<td>Dr Sarabpreet Singh Dr Pranay Ghosh</td>
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<td>Panel discussion: What lies between an average and successful IVF program</td>
<td>Dr Piya Ray Dr Saroj Agarwal</td>
</tr>
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# Programme for the day

<table>
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<th>Time</th>
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<tr>
<td>08:30 - 09:00</td>
<td>Registration</td>
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<tr>
<td>09:00 - 09:30</td>
<td>Welcome address</td>
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<tr>
<td>09:30 - 09:50</td>
<td>Individualized ovarian stimulation for IVF: finding a balance between cost, safety and outcome</td>
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<td>09:50 - 10:10</td>
<td>Errorless OPU and ET: Is it possible?</td>
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<td>10:10 - 10:30</td>
<td>Is 100% outcome achievable in Vitrification?</td>
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<td>10:30 - 11:00</td>
<td>Tea &amp; discussion</td>
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<td>11:00 - 11:20</td>
<td>KPIs in clinical practice: what is ideal?</td>
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<td>11:20 - 11:40</td>
<td>Why patients don't turn up for repeat IVF?</td>
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<td>11:40 - 12:00</td>
<td>Expecting the unexpected in IVF lab?</td>
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<td>Tea &amp; discussion</td>
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<td>Panel discussion: What lies between an average and successful IVF program</td>
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<td>13:30 - 13:40</td>
<td>Vote of Thanks</td>
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<td>Lunch</td>
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<td>S. No.</td>
<td>Topic</td>
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<td>Expecting the unexpected in IVF lab?</td>
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<td>7</td>
<td>What lies between an average and successful IVF program</td>
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1. Individualized ovarian stimulation for IVF: finding a balance between cost, safety and outcome
INDIVIDUALIZED OVARIAN STIMULATION FOR IVF FINDING A BALANCE BETWEEN COST, SAFETY & OUTCOME

Why individualise?

- Obtain good number & quality egg to produce good euploid embryos
- Avoid OHSS
- Avoid cycle cancellation
- Maximise our success / transfer

Which two groups of patients can be divided?

- GOOD PROGONOSIS
- LOW PRONOGOSIS

HOW CAN WE DEFINE?
AMH predicts ovarian response

<table>
<thead>
<tr>
<th>Value of AMH (ng/mL)</th>
<th>Ovarian response</th>
</tr>
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<tbody>
<tr>
<td>&lt;0.4</td>
<td>Extreme Poor Responder</td>
</tr>
<tr>
<td>0.4 – 1</td>
<td>Poor Responder</td>
</tr>
<tr>
<td>1 – 2</td>
<td>Poor Responder</td>
</tr>
<tr>
<td>2 – 3.5</td>
<td>Normal Responder</td>
</tr>
<tr>
<td>3.5 – 6</td>
<td>Hyper Responder</td>
</tr>
<tr>
<td>&gt;6</td>
<td>Hyper Responder</td>
</tr>
</tbody>
</table>

La Marca A. Hum Reprod 2007 ; 22:766-71

HOW MANY OOCYTES ARE NEEDED FOR ATLEAST ONE EUPLOID EMBRYO?

Mean number of oocytes needed and age

- Age <29: 
  - >6 COCs
  - >5 MII oocyte
  - >4 fertilized oocytes
  - >2 blastocyst
  - Euploidy rate 80%
  - 1 euploid blastocyst

- Age 29-40: 
  - >11 COCs
  - >9 MII oocyte
  - >7 fertilized oocytes
  - >3 blastocyst
  - Euploidy rate 80%
  - 1 euploid blastocyst

- Age 42-49: 
  - >18 COCs
  - >16 MII oocyte
  - >13 fertilized oocytes
  - >5 blastocyst
  - Euploidy rate 20%
  - 1 euploid blastocyst
HOW MANY EGGS WERE IVF SUCCESS RATE IS MORE?

- 2011-Association between number of eggs & live birth in IVF treatment: analysis of 400135 cycles; Sunkara, et.al concluded 15 eggs can maximise LBR
- 2013 Optimum number of oocytes in IVF treatment: analysis of 2455 cycles; Ji J, et.al concluded oocytes number should be 6-15 for achieving a live birth

- Women undergoing COS for their first IVF/ICSI cycle and planned SET should be informed that, although the number of oocytes retrieved does not affect LBR in the fresh cycle, the higher the oocyte yield the higher the probability to achieve a live birth after utilization of all cryopreserved embryos.
What are the key concerns before starting the stimulations?

- WHICH PROTOCOL?
- WHICH GONADOTROPHINS?
- WHAT IS THE STARTING DOSE?
- IS ONE SIZE FIT FOR ALL?
- IS BOUTIQUE APPROACH (individualize) WILL HELP AND WHEN?

General approaches to COS in IVF
How to improve my ART outcome Quality Control / Quality Assurance (QA/QC)

Indian Fertility Society & Origio India Initiative

Same live birth rate

Gonadotrophin-releasing hormone antagonists for assisted reproductive technology

WHICH GONADOTROPHINS?
For the comparison HMG or HP-HMG versus FSH-P there was also no difference in the evidence on live birth rate (OR 1.36, 95% CI 0.58 to 3.18, 3 trials, 138 women, I² = 0%, low-quality evidence).

This suggests that for a woman with a live birth rate of 18% with HMG or HP-HMG, the chance of live birth following uFSH is between 9% and 37%.

Good Prognosis

- NORMAL RESPONDER
- HYPER RESPONDER
What is the starting dose
One size fit for all?

CONCLUSIONS: When using a starting dose of 225 IU rhFSH combined with the multiple dose of 0.25 mg cetrorelix from stimulation day 6, significantly more oocytes were obtained than with a starting dose of 150 IU rhFSH.
Live birth or ongoing pregnancy

- **200 versus 100 IU** (OR 0.88, 95% CI 0.57 to 1.36; N = 522; 2 studies; I² = 0%). This suggests that if the chance of live birth or ongoing pregnancy with 100 IU is 20%, then the chance with 200 IU would be 13% to 26%.

- **225/200 versus 150 IU** (OR 1.03, 95% CI 0.57 to 1.86; N = 277; 1 study). This suggests that if the chance of live birth or ongoing pregnancy with 150 IU is 19%, then the chance with 200/225 IU would be 12% to 31%.

- **300 IU versus 225 IU** (OR 0.65, 95% CI 0.32 to 1.32; N = 135, 1 study). This suggests that if the chance of live birth with 225 IU is 40%, then the chance with 300 IU would be 17% to 47%.

- In the third comparison, the confidence interval remains wide, and it is not clear whether there is any effect from 300 IU versus 225 IU.

Cancellation due to low ovarian response to stimulation in comparison A (100 versus 200 IU/day recFSH) was observed to be more frequent in the 100 IU/day recFSH dose group [OR 5.02 (calculated pooled estimates 16.4 and 3.8%, respectively); 95% CI 2.19–11.51; P = 0.0001].

There was no difference in cancellation rate for low response in comparison B (150 versus 200–250 IU/day recFSH) [OR 1.10 (calculated pooled estimates 4.4 and 4.0%, respectively); 95% CI 0.59–2.05; P = 0.76].

Nice recommendations

- Use an individualised starting dose of gonadotropin based on
  - Age
  - BMI
  - Ovarian reserve
  - Presence of PCO

- Never use gonadotropin dose >450IU
Good responder

WHAT ABOUT HYPER RESPONDER?

OHSS FREE CLINIC – A RELAITY
Predicted hyper responder

- We randomized 255 women to a daily FSH dose of 100 IU and 266 women to a daily FSH dose of 150 IU.
- The cumulative live birth rate was 66.3% (169/255) in the reduced versus 69.5% (185/266) in the standard group (relative risk (RR) 0.95 [95%CI, 0.85–1.07], P = 0.423).
- The occurrence of any grade of OHSS was lower after a lower FSH dose (5.2% versus 11.8%, RR 0.44 [95%CI, 0.28–0.71], P = 0.001), but the occurrence of severe OHSS did not differ (1.3% versus 1.1%, RR 1.25 [95%CI, 0.38–4.07], P = 0.728).
- As dose reduction was not less expensive (€4.622 versus €4.714, delta costs/woman €92 [95%CI, −479–325]), there was no dominant strategy in the economic analysis.

TAILOR APPROACH: IS IT POSSIBLE?

AMH/AFC for choosing protocol?

- High Responders (≥50 IU)
  - Safe & Effective
- Normal Responders (150–225 IU)
  - Safe & Effective
- Poor Responders (≤100 IU)
  - Aggressive Strategy
  - Short stimulation
  - Moderate cancellation

Not benefit from higher FSH dose
Reduced treatment burden
Concept of individualized ovarian control on age, basal FSH, AMH & BMI

<table>
<thead>
<tr>
<th>AMH Level</th>
<th>Starting Dose FSH</th>
<th>Approx equivalent in µg</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;5 pmol/l</td>
<td>210 IU</td>
<td>10-15 µg</td>
</tr>
<tr>
<td>5-14.9 pmol/l</td>
<td>180 IU</td>
<td>8-11 µg</td>
</tr>
<tr>
<td>&gt;15-29.9 pmol/l</td>
<td>150 IU</td>
<td>6-9 µg</td>
</tr>
<tr>
<td>&gt;30-44.9 pmol/l</td>
<td>120 IU</td>
<td>4-7 µg</td>
</tr>
<tr>
<td>&gt;45 pmol/l</td>
<td>90 IU</td>
<td>2-5 µg</td>
</tr>
<tr>
<td>Not Available</td>
<td>120-180 IU</td>
<td>6-11 µg</td>
</tr>
</tbody>
</table>

LOW PROGNOSIS GROUP
Suboptimal
(4-9 oocytes)
means less than optimal
(10-15 oocytes)

So what is
“hypo-response”? 

Hypo-response to rFSH

- Hypo-responders are women with normal ovarian reserve who can achieve 'adequate'
  number of oocytes retrieved and oestrogen production

BUT...
There is an increase in the cumulative rFSH dose
(i.e. >3000 IU) and in the stimulation length

Hormonal, functional and genetic biomarkers in controlled ovarian stimulation: tools for matching patients and protocols

- Suboptimal responders
- Normal responders
- Hypo-responders
How to improve my ART outcome

Quality Control / Quality Assurance (QA/QC)

Indian Fertility Society & Origio India Initiative

Concept of individualized ovarian control on age, basal FSH, AMH & BMI

Basal FSH levels and ampoules of FSH used in COS for patients with variants of the FSH receptor. Basal FSH levels (left panel) and ampoules of FSH used in COS (right panel) for homozygote wild-type (Asn/Asn), heterozygote (Asn/Ser680) and homozygote (Ser680/Ser680) carriers of the Ser680 variant of the FSH receptor. ASN = asparagine; CPRs = clinical pregnancy rates; COS = controlled ovarian stimulation; FSH = follicle-stimulating hormone; r-FSH = recombinant follicle-stimulating hormone; Ser680 = Serine680. Adapted from Perez Mayorga et al. 2000

Patient genetic profiles: interpretation of physiology and biomarker levels

<table>
<thead>
<tr>
<th>Genetic profile</th>
<th>Interpretation of genetic profile</th>
</tr>
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<tbody>
<tr>
<td>Low AMH levels; low AFC</td>
<td>Suggests that even high doses of FSH would be ineffective and that LH would not improve results. Patient would benefit from counseling to better understand her limited chances of success.</td>
</tr>
<tr>
<td>FSH receptor variant (eg. Ser680); good AMH levels; good AFC</td>
<td>Suggests a good prognosis, but it also predicts a genetic hyporesponsivity to FSH that should be considered when formulating COS treatment.</td>
</tr>
<tr>
<td>r-LH variant in β subunit of LH receptor</td>
<td>Suggest that a patient might benefit from LH supplementation during COS.</td>
</tr>
</tbody>
</table>

AMH = anti-Müllerian hormone; AFC = antral follicle count

Four Groups of Patient with Lower Prognosis

**GROUP 1**
Young patients (<38 years) with adequate ovarian reserve parameters (AFC >8; AMH >1.2 ng/ml) and with an unexplained poor or suboptimal ovarian response leading to failure

**GROUP 2**
Older patients (35 years) with adequate ovarian reserve parameters (AFC >8; AMH >1.2 ng/ml) and with an unexplained poor or suboptimal ovarian response leading to failure

**GROUP 3**
Young patients (<38 years) with poor ovarian reserve pre-stimulation parameters (AFC <8; AMH <1.2 ng/ml)

**GROUP 4**
Older patients (35 years) with poor ovarian reserve pre-stimulation parameters (AFC <8; AMH <1.2 ng/ml)
Four group of patient with low prognosis

... But how many eggs we need?

GROUP 3
Young patients (<35 years) with poor ovarian reserve pre-stimulation parameters (AFC<5; AMH<1.2 ng/ml)

GROUP 4
Older patients (>35 years) with poor ovarian reserve pre-stimulation parameters (AFC<9; AMH<1.2 ng/ml)

Four group of patient with low prognosis

... So we need many eggs from women with low reserve
How can we do it if no Gn can compensate?

GROUP 3
Young patients (<35 years) with poor ovarian reserve pre-stimulation parameters (AFC<5; AMH<1.2 ng/ml)

GROUP 4
Older patients (>35 years) with poor ovarian reserve pre-stimulation parameters (AFC<9; AMH<1.2 ng/ml)

Characterization of Ovarian Follicular Wave Dynamics in Women

Documentation of major and minor follicular waves during the menstrual cycle challenges the traditional theory that a single cohort of antral follicles grows only during the follicular phase of the menstrual cycle.

A new model for ovarian follicular development during the human menstrual cycle

Sixty-eight percent of women exhibited two waves of follicle development during the luteal and 32% exhibited three waves. Waves were characterized by an increase and subsequent decrease in the number of follicles 5 mm occurring in association with the growth of 2 follicles to 6 mm.
Duostim in low prognostic patient

In low prognosis patients (group 3-4 Poseidon), duostim can maximize the number of oocytes per menstrual cycle increasing the chance of obtaining the embryo that can give a live birth and it could be applied in all patients with few "fertile time" left available.
Individualized versus standard FSH dosing in women starting IVF/ICSI: an RCT. Part 1: The predicted poor responder


Summary:
In women with a predicted poor ovarian response (AFC<11) undergoing IVF/ICSI, an increased FSH dose (225/450 IU/day) does not improve cumulative live birth rates as compared to a standard dose (150 IU/day)

How to reduce the cost?

IN IVF CYCLE THE MAJORITY OF COST IS OF HORMONES

- So for cost reduction we need to decrease total dose of hormone required and also the duration of stimulation
- For that we need to compare the cumulative live birth rate in
  - Natural cycle
  - Min to mild stimulation
  - Conventional stimulation
Multiple Natural Cycles of IVF Required for Reasonable Success Rate


4 cycles of natural cycle IVF = 1 cycle conventional IVF

Outcomes of Natural Cycle IVF

- High cancellation rate (premature LH surge, failed retrieval, etc)
- Success rates remain consistently low
- Only multiple cycles for the same couples provide similar success to conventional IVF
- Per cycle costs 20 – 30% of conventional (although IVF retrieval, laboratory and embryo transfer costs remain the same)

Comparison of Pregnancy rates for poor responders using IVF with mild ovarian stimulation versus conventional IVF: a guideline

American Society for Reproductive Medicine, Birmingham, Alabama


CONCLUSION: Mild ovarian-stimulation protocols with IVF generally aim to use less medication compared with conventional IVF. In patients expected to be poor responders with IVF (based on poor response to a prior IVF cycle, age ≥40 years, and /or Bologna criteria), pregnancy rates tend to be low regardless of protocol. There is fair to good evidence that clinical pregnancy rates are not substantially different using mild-stimulation protocols compared with conventional IVF in poor-responder populations. Based on one study, mild stimulation with CC was cost-effective compared to conventional IVF with high-dose gonadotropins.
Conclusions

- Flexible Antagonists for all is the right future.
- Adding LH in hyper responder and normal responder is controversial.
- Adding LH is beneficial in poor responder.
- Mild stimulation (150 IU to 200 IU rFSH) in compared to conventional stimulation gives the same cumulative live birth rates, with patient friendly approach and cost effective.
- Mild Simulation is the last option in poor responder.
- Duo stimulation is the new approach to treat poor responder.
- Pretreatment estradiol valerate is always beneficial.
- Query Growth hormone is debatable.

SAFETY:
1: LUPERIDE TRIGGERING
2: OHSS FREE CLINIC

COST EFFECTIVE:
1: HMG IS EQUALLY EFFECTIVE (DEBEAT STILL CONTINUES FOR ADDING LH)
2: 150 IU VS ANY HIGHER DOSE EQUAL EFFECTIVE

OUTCOME:
CUMULATIVE LIVE BIRTH REMAINS SAME WITH ANY PROTOCOL OR ANY DOSE (150 IU IS THE LOWEST THRESHOLD DOSE)

THANK YOU
2. Errorless OPU and ET: Is it possible?
To Err Is Human

**Human error** means that something has been done that was “not intended by the actor; not desired by a set of rules or an external observer; or that led the task or system outside its acceptable limits”.

 Applies to the practice of ART

TECHNICAL ERRORS: can compound the problem

---

To err is human even in IVF

**To ensure smooth and errorless operations:**

- Reduce potential source of error (HAZARDS)
- Reduce possibility that something unpleasant happens (RISKS)
- A safety culture right from training should be part of program’s DNA
- Reinforcement about how to avoid possible sources of error is the key

---

Errors in oocyte retrieval – where we can go wrong

- Treatment
- Operational
- Technical
- laboratory
AIM

To get maximum yield of oocyte with minimum complications

Factors affecting oocyte yield

- Magnitude of ovarian stimulation
- Type of anesthesia (local, sedation or general)
- Type of aspiration needle (wide or narrow bore or single or double channel)
- Aspiration alone or aspiration with follicular flushing
- Experience and skill of the surgeon.

Ovarian stimulation

Choosing individualized starting dose of Gn acc to
- Age
- BMI
- Ovarian reserve markers (AFC, AMH)
- Previous IVF response
• Effective conscious sedation and analgesia for pain relief
• Any method can be used
• Simultaneous use of sedation combined with analgesia (opiates), further enhanced by paracervical block or acupuncture techniques:
• Better pain relief than occurred with one modality alone.
• Evidence insufficient to show conclusively whether any of the interventions influenced pregnancy rates.
• All techniques: a high degree of patient satisfaction.
• Women's preferences and resource availability for choice of pain relief merit consideration in practice.

Errors in oocyte retrieval

Treatment errors
Stimulation protocol
• Quantity and quality of oocytes
Trigger
• Time
• Dose (mixing it properly)
• Type of trigger

Errors in oocyte retrieval

Trigger
• Not given properly
• Time and dose of trigger (Ensure patient has taken on time)
• Donors make most mistakes
• HCG (only the saline is given and powder not mixed)
Errors in oocyte retrieval

Operational
- Haemorrhage
- Bowel injury
- Bladder injury
- Infectious complications (rupture of endometrioma)
- Anesthetic

Errors in oocyte retrieval

Technical errors
- Identify the patient (time out)
- Pressure of suction pump (high pressure leads to empty follicles)
- Temperature (mobile nest, media, work station)
- Minimal duration of exposure to blood after retrieval (Rinsing, incubation)
- Duration of exposure to light (oocytes)

Ovarian stimulation

- Type of aspiration needle
- Suction pressure
- Follicular flushing or no flushing
### Type of aspiration needle

- Any size between 15-18 G has been used in different studies
- Smaller size needles cause less postoperative pain with similar yield of oocytes.

### Suction pressure

- 100-120mmHg
- IVM – aspiration pressure should be further decreased
- High suction pressure:
  - Oocytes become denuded of cumulus cells
  - Negative impact of increasing aspiration pressures is greater in larger-gauge needles

### Follicular Flushing

**In DOR patients – to maximise the oocyte yield**

**Benefits:**
May increase the oocyte yield thus increasing pregnancy rate

**Disadvantages:**
- Time consuming
- More media use
- Risk of infection
- Remove some follicular cells that could potentially serve an important endocrine luteal support function.
How to improve my ART outcome

Quality Control / Quality Assurance (QA/QC)

Indian Fertility Society & Origio India Initiative

Errors in oocyte retrieval

- follicular flushing probably: little or no effect on live birth rates compared with aspiration alone.
- Data suggest little or no difference between follicular flushing and aspiration alone with respect to oocyte yield, total embryo number, or number of cryopreserved embryos.

OCR: a safe procedure but is not without risks.

- Most important identifiable risk factors for occurrence of complications are:
  - High number of oocytes retrieved
  - Long duration of the procedure
  - Mean time per oocyte retrieved
  - Inexperience of the surgeon
  - Younger patients with a lesser BMI
  - H/0 prior abdominal or pelvic surgery or PID

Conclusion(s): Oocyte retrieval can be considered a safe procedure but is not without risks. The most important, identifiable, risk factors for the occurrence of complications are: [1] high number of oocytes retrieved, [2] a long duration of the procedure and mean time per oocyte retrieved, [3] inexperience of the surgeon, [4] younger patients with a lesser BMI, and [5] history of prior abdominal or pelvic surgery or pelvic inflammatory disease.

Ovarian stimulation

Appraisal of clinical complications after 23,827 oocyte retrievals in a large assisted reproductive technology program

Conclusions: Oocyte retrieval can be considered a safe procedure but is not without risks. The most important, identifiable, risk factors for the occurrence of complications are: [1] high number of oocytes retrieved, [2] a long duration of the procedure and mean time per oocyte retrieved, [3] inexperience of the surgeon, [4] younger patients with a lesser BMI, and [5] history of prior abdominal or pelvic surgery or pelvic inflammatory disease.
**Oocyte retrieval** is a particularly sensitive procedure and special attention should be given to temperature and pH as well as efficient and quick handling

- An identity check before the oocyte retrieval is mandatory.
- The time between oocyte retrieval and culture of washed oocytes should be minimal. Prolonged oocyte exposure to follicular fluid is not recommended

Appropriate equipment must be in place to maintain oocytes close to 37°C. Flushing medium, collection tubes and dishes for identifying oocytes should be pre-warmed. Follicular aspirates should be checked for the presence of oocytes using a stereomicroscope and heated stage, usually at 8-60x magnification. Exposure of oocytes to light should be minimised
Timing of retrieval, number of collected oocytes and the operator should be documented.
Embryo transfer

- One of the most critical steps in IVF is embryo transfer
- The technique has a great impact on the IVF results
- In a survey of 80 IVF practitioners, standardization of ET technique was considered the most important factor influencing the success rate

Embryo Transfer: Goal

To deliver the embryo(s) atraumatically to a location within the endometrial cavity that maximizes the chance of implantation

Standardization of Technique

Evaluation of uterine cavity
- Dummy ET
- Ultrasonographic evaluation

Avoiding the initiation of uterine contractions
- Avoid touching the uterine fundus
- Soft catheters
- Gentle manipulation
- Uterine relaxing substances
**Standardization of Technique**

- Removal of cervical mucus
- Ensure that the ET Catheter passed the internal cervical Os
- Prevention of embryo expulsion

**Potential negative factors associated with ET**

- Uterine contractions
- Failure to pass the Internal Cervical Os
- Cervical mucus

**Important Points**

- Mock ET should have been performed before the cycle and properly documented so that there is no problem while performing it
- Identify patient (staff should be aware of the time and identity of the patient)
- Identify the embryos
Technical errors in ET

- If the laboratory is some distance from the embryo transfer room, arrangements should be made to maintain temperature and pH whilst transporting embryos
- A double identity check of the patient, the patient file and the culture dishes is mandatory immediately before the transfer

ASRM practice guideline (2015)

The following interventions are supported by the literature for improving pregnancy rates:
- Abdominal ultrasound guidance for embryo transfer
- Removal of cervical mucus
- Use of soft embryo transfer catheters
- Placement of embryo transfer tip in the upper or middle (central) area of the uterine cavity, about 1-1.5cm proximal to the fundus
- Immediate ambulation once the embryo transfer procedure is completed

Disclosure: 2011 ASRM ethics committee report

- Ethical obligation
- Errors that affect the number of quality of embryos should be disclosed
- Obligatory to disclose errors where gametes or embryos are switched
- Promote culture of truth-telling
- Write procedures for disclosure
- Rigorous procedures for proper ID and prevention of loss
Disclosure

Difficult

- Hard to admit mistake has been made
- Reputation
- Legal fears
- Blame focused on individual, not system

Procedure for Identification

- Witnesses for every step
- Wristband checks for all egg retrievals and transfers
- Time out prior to egg retrieval
- At time of transfer, TV monitor displays pt. name and DOB on bottom of dish to pt., nurses and physician

Conclusions

- The practice of reproductive medicine can involve medical errors in which gametes and embryos are lost, degraded, or misdirected, as well as near misses in which errors are averted before producing any clinical impact.
- This should include written labeling as well as verbal identification at the initiation of embryo transfer.
Conclusions

- The best ethical practice is for programs to have in place rigorous procedures to prevent errors.
- To prepare for the possibility that errors may occur despite these procedures, programs should foster an environment of truth telling that will allow prompt identification and disclosure of errors to patients.
- It is recommended that clinics have written policies and procedures that outline how to reduce and disclose medical errors.

Final Thoughts

Errors are inevitable!

- We can ignore them or
- Use them to improve
- Acknowledge them
- Understand why they happened - get to the source of the problem
- Opportunities to improve processes
3. Is 100% outcome achievable in Vitrification?
Introduction

Melting point, TM,
glass transition temperature, TG,
homogeneous nucleation temperature, TH,
devitrification temperature, TD,

The fundamental issue in all vitrification methods is to achieve and maintain conditions within the cells that guarantee a glass like state and prevent the formation of ice crystals throughout the cooling, as well as during the warming process.

- An optimal balance between:
  - The speed of cooling, warming (time and temperature), and
  - The optimal cell dehydration and penetration of CP when the cells are exposed to concentrated hypertonic solutions

Overview

- Step 1: Selection of embryos (top and good)
- Step 2: Exposure of embryos to the CPsol.
- Step 3: Loading on the carrier device and plunging it in liquid nitrogen (LN₂).
- Step 4: Storage in LN₂ containers.
- Step 5: The warming process.
- Step 6: Rehydration and removal of the intracellular cryoprotectant (CP).
- Step 7: Selection of warmed embryos before embryo transfer.

Step 1: Selection of Embryos

- Transferable: Top, Good and Fair
- Freezable: Top and Good
How to set-up Vitrification Cooling Media

- Follow manufacturer’s instructions
- Innovation is ok but experimentation is not
- should be warmed to RT for atleast 30-60 min

Written instructions explained and pasted

Labeling
Step 2: Exposure of Embryos

- CP toxicity decreases with temperature:
  - Work on RT, ice
- CP toxicity is time dependent
  - Don't experiment with time
- CP toxicity is concentration dependent:
  - Two-step strategy – preincubate in equilibrium solution (5-15 min): 20 – 50% of the VS concentration
  - Short exposure to VS – approx 1 min

Well vs Drops

ES: 5-15 min
Stage of embryos

Step 3: Loading on the carrier device and plunging it in Liquid Nitrogen
Step 3: Loading on the carrier device and plunging it in liquid nitrogen (LN₂)

Sinking vs floating devices

Aspirating Embryos with Minimal Volume

Minimum Drop Size

Probability of fracturing = CR × μ × V

e probability of vitrification is = CR × μ × 1/V
Proper Placement and Documentation

- Traceability and witnessing
- Document everything – expire time of E5 and V5
- Avoid multiple patients - separate
- Samples should remain under liquid nitrogen to avoid desiccation

Avoiding Vapor

- The vapor coat: prevents a truly high rate of cooling
- Eliminating vapor is important
- Filled to the brim – 0.5 cm
- Cryocane,
- Cap
- LN2 Slush
- Pre-cooled metal surfaces

Capping the Device
Seki and Mazur, 2012 demonstrated that the warming rate is dominant over the cooling rate: closed devices

Warming rates are extremely important:
- Nucleation of unfrozen freezable water at low temperatures, which may lead to the freezing of this water upon warming, with attendant injury
- keep the samples for 1–3 s in air to avoid fracture damage caused by gas bubbles occurring in the too rapidly immersed samples
Things to Avoid

- **Safety:** cryopreserving multiple patients simultaneously
- Never place culture dishes or dilutions from different patients on the microscope stage simultaneously.
- During each step in the process one should verify a patient’s name and repeat it aloud.

**Centre well vs 4 well dishes for warming**

- Spillage
- Media Volume
- Embryos under vision during warming
- Push rather than aspirate if attached
- Embryos float to the top
- Change microscope settings
- Gentle

**Indian Fertility Society & Origio India Initiative**
Osmotic Injury

- CONCENTRATION OF VITRIFICATION MEDIA MORE THAN WARMING MEDIA WHICH BEING HYPOTONIC RUSHES IN
- TEMPERATURE OF VITRIFICATION MEDIA – not as recommended

Take Home Message

- Freeze inferior embryos first especially at a new set up
- Use device which sinks than floats
- Universal warming media - SF is nothing more than another way of performing vitrification.
- Freeze 2-3 embryos per device (no selective warming)
- Day 2 vs Day 3 vs Day 5 freezing – multifactorial besides operator’s preference
- Blastocyst – hatching n post warming incubation

THANK YOU
4. KPIs in clinical practice: what is ideal?
Key Performance Indicator (KPI)

- Performance measurement to identify deviations from the optimum or from established limits, and then being able to act when such deviation exceed certain limit.
- Plays an important role in avoiding adverse consequences and maintaining optimal performance.

Why KPIs?

- Evaluating the introduction of a technique or process
- Establishing minimum standards for proficiency
- Monitoring ongoing performance
- Benchmarking
- Quality improvement.

KPIS: How do they work?

- Comparing the result with standards
- Standardized methods of measurement
- Specificity of data to be collected and periodicity of measurement
KPIs: Key Features

- Significant
- Reliable
- Routine

LABORATORY KPIs

CLINICAL KPIs

Laboratory KPIs

- Employed for internal quality control in IVF/ICSI programs, using indicators such as oocyte fertilization, cleavage embryo rates, percentage of top quality embryos, etc.
- Can be plotted and compared with established limits for the mean and standard deviation values, so that deviations can be easily recognized as warnings or action points

Advantages

- The KPIs score strategy application could result in an immediate evaluation of the patient's clinical and laboratory performance in the ART cycle
- In addition, internal quality control benchmarks could be evaluated
A6. Evaluation and quality assurance (KPIs)

A6.1. The laboratory should establish quality indicators for systematically monitoring and evaluating the laboratory's performance evaluated on a regular basis previous year's results as benchmarks.

A6.2. Intervals between analysis should be based on the laboratory case-load.

A6.3. Laboratories should calculate their own alert conditions for all parameters.

A6.4. Deviation from these benchmarks should warrant investigation in line with the clinic's non-conformance policy.
• ESHRE revised guidelines for good practice in IVF laboratories (2015)
• KPIs repeatedly mentioned but not covered in detail

AIM:
• To establish KPIs for use in monitoring “fresh” IVF and ICSI cycles
• To achieve an international consensus regarding a minimum list of IVF laboratory Indicators and KPIs
• Specific definitions for these Indicators
• Recommended values for each KPI
Types of Indicators

- **RIs (reference indicators):** related to the oocytes coming into the laboratory - indirect indicators of the response to ovarian stimulation
- **PIs (performance indicators):** were those for which data should be documented and stored
- **KPIs** were those related to the ART laboratory

Data collection for indicators - monthly basis

Reference population for Indicator Values

- Female patients <40 years old;
- Own fresh oocytes;
- Ejaculated spermatozoa (fresh or frozen);
- No PGD/PGS (PGT)
- Routine IVF and ICSI

RI - Indirect Indicator of Ovarian Stimulation

- Good stimulation: yields a well-expanded cumulus oophorus complex (COC) with higher number of MII oocytes
- Poor stimulation: results in abnormal COC morphology and fertilization and increased rate of aneuploidy
- Oocytes retrieved should be 80–95% of follicles measured in stimulated cycles
- Proportion of MII oocytes at the time of ICSI should be 75–90%

Deviations from the expected range needs investigation of any changes in stimulation protocol
Recommendations for ART modalities

- No KPI for sperm recovery rate because this is heavily dependent on the processing method
- Recommendations for IUI or IVF treatment: post-wash sample showing at least 90% progressive motility
- There was no cut-off value recommendation for ICSI treatment, beyond the spermatozoa ideally being alive

**Giant oocytes and oocytes with smooth endoplasmic reticulum cluster should not be injected**

**Total ICSI failed fertilization rate is not a PI, but should be reported and investigated**

---

**Indicators**

<table>
<thead>
<tr>
<th>RI</th>
<th>Calculation</th>
<th>Benchmark value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proportion of oocytes recovered (cumulated cycles)</td>
<td>No. oocytes retrieved / No. oocytes injected × 100</td>
<td>&gt;95</td>
</tr>
<tr>
<td>Proportion of MII oocytes at ICSI</td>
<td>No. MII oocytes / No. total oocytes injected × 100</td>
<td>&gt;75</td>
</tr>
</tbody>
</table>

*PI: performance indicator; COC: cumulus-oocyte complex.*
- The Alpha benchmarks -
Oocyte cryopreservation

<table>
<thead>
<tr>
<th>Slow Freezing</th>
<th>Vitrification</th>
</tr>
</thead>
<tbody>
<tr>
<td>75%</td>
<td>85-95%</td>
</tr>
</tbody>
</table>

No more than 10% (absolute) lower than that of a comparable population of fresh oocytes

The same as for a comparable population of fresh embryos

No more than 10-30% (relative) lower than that of a comparable population of fresh embryos

---

- The Alpha benchmarks -
Cleavage stage embryo cryopreservation

<table>
<thead>
<tr>
<th>Slow Freezing</th>
<th>Vitrification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Survival (Intact)</td>
<td>55%</td>
</tr>
<tr>
<td>Survival (&gt;50%)</td>
<td>85%</td>
</tr>
<tr>
<td>Implantation</td>
<td>Same as that of a comparable population of fresh embryos</td>
</tr>
</tbody>
</table>

---

- The Alpha benchmarks -
Blastocyst stage embryo cryopreservation

<table>
<thead>
<tr>
<th>Slow Freezing</th>
<th>Vitrification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Survival</td>
<td>85%</td>
</tr>
<tr>
<td>Implantation</td>
<td>Same as that of a comparable population of fresh embryos</td>
</tr>
</tbody>
</table>
Clinical KPIs

- Age
- BMI
- AMH
- Stimulation protocol
- Number of oocytes obtained

Maximum KPI: 25

- Age <= 36
- AMH >= 2
- Metaphase II oocytes >= 7
- Fertilisation rate >= 65%
- Number of top quality embryos >= 2
- Clinical pregnancy probability is 70%

Maximum KPI: 5

- Age >= 40
- AMH < 1
- Metaphase II oocytes <= 3
- Fertilisation rate < 50%
- Top quality embryos - none
- Clinical pregnancy probability is 3%
Key performance indicators score (KPIs-score) based on clinical and laboratorial parameters can establish benchmarks for internal quality control in an ART program.

Clinical KPIs (C–KPIs) – age, AMH and number of oocytes collected
Laboratory KPIs (L–KPIs) – fertilization rate, embryo quality

This paper analyzed if a KPIs-score strategy with clinical and laboratorial parameters could be used to establish benchmarks for internal quality control in ART cycles.
Other Factors

- When C-KPI & L-KPIs are maximum but still gestation rate is low then
  - Embryo transfer technique
  - Endometrial receptivity problem
  - Implantation window
  - Luteal phase supplementation

Clinic and Laboratory Interaction

Good communication between laboratory and clinic regarding cycle planning and cycle review, ensuring appropriate procedures are ordered, and proper identification of the patient/patient's specimens are crucial for good outcomes in IVF programs

Take Home Message

- The KPIs system was used to detect early warning signals in gamete/embryo cultures
- Each clinic should establish its own key performance indicators and benchmarks
- Develop a systematic, transparent, and consistent approach to data collection and analysis and calculation of KPIs
THANK YOU

Further Reading


5. Why patients don’t turn up for repeat IVF?
Introduction

- There has been significant developments in the field of ART in the last four decades since the birth of the first IVF baby in 1978
- Though the treatment has become simple and effective
- In spite of this, majority of the patients do not come back for a repeat treatment cycle after the failure of the initial attempt
- It is largely due to the fact that ART still remains an expensive, time-consuming and physically & emotionally draining treatment with an uncertain outcome

Why patients don’t turn up for repeat IVF

- TREATMENT ASSOCIATED
  - Psychological burden
  - Physical stress
  - Financial burden
  - Ovarian hyper-stimulation syndrome (OHSS)
  - Ectopic pregnancy

- PATIENT FACTORS
  - UNREALISTIC EXPECTATIONS: efficacy of ART
  - Elderly patients feel they are past the reproductive age
  - Unable to accept failure
  - Higher drop out in elderly patients

Clinic Related

- Inadequate counseling
- Inadequate confidence / faith in the treatment / doctor
- Unfriendly staff
- Inadequate preparation for a long treatment duration requiring
- multiple cycles of ART
- Lower success rates
- High miscarriage rates
Patients Perceptions

- Mahlstedt et al. found that 82% of the patients did not view the decision to enter the IVF program to be a difficult one and that upon entry to the program, 56% of patients indicated they would repeat the IVF treatment if the initial procedure was not successful.
- Interestingly, after the first IVF cycle, only 37% of couples planned to repeat the treatment.
- Factors they found that influenced patients’ plans not to repeat IVF included emotional strain, medical barriers, and disruption of activities.
- They feel they have passed the reproductive age.

Factors influencing patients’ decision not to repeat IVF

Women who did not pursue a second in vitro fertilization cycle after a failed cycle were surveyed. The major reason for not pursuing a second cycle was financial.

Key words: finances, in vitro fertilization, psychological stress.

Table III. Responses to Open-Ended Questions

<table>
<thead>
<tr>
<th>Questions</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Can you think of any significant positives about IVF experience?</td>
<td></td>
</tr>
<tr>
<td>Professionally competent</td>
<td>2</td>
</tr>
<tr>
<td>Caring supportive staff</td>
<td>2</td>
</tr>
<tr>
<td>Staff helpful</td>
<td>1</td>
</tr>
<tr>
<td>Staff absent</td>
<td>1</td>
</tr>
</tbody>
</table>
Physical, Mental and Emotional Stressors in Females

Many patients describe IVF as an emotional roller-coaster

- Female under goes physical pain of taking injections
- Multiple sittings of tVS
- The stress of undergoing ovum pickup
- being informed that very few ova have been retrieved
- Expectation crashes when told not good/very few embryos have developed
- Finally awaiting result and being told that Bhcg is less than 10 on day 15 after embryo transfer

Financial drain in IVF

- Since IVF is a complex process and needs a series of procedures thus cost rises
- Cost also increases with donor’s sperm/eggs, sperm retrieval process, ICSI, assisted hatching, embryo freezing cost etc.
- High cost of hormones, disposables & media
- Limited success rate: many require multiple attempts of fresh and frozen cycles
- Many patients under loan, sell land or jewelry to afford single cycle of IVF
PHYSICALLY TIRING
- Multiple visits
- Synthetic hormones, and they make patient feel intense PMS, mood swings, hot flashes
- Temporary weight gain/bloating are common, as are headaches, breast tenderness and nausea
- Some women feel skin irritation at the injection site

TIME CONSUMING
- Fertility treatment takes time, and may disrupt couples work schedule and routine at home. And the uncertainty of the outcome just adds more stress
- Working class women affected more

Misinformation and lack of transparency
- Mostly pts are informed the maximal CPR/LBR, where as it may be different in their individual case due to added pelvic pathology/hormonal milieu
- Pts don’t want to believe the lower end of success rate so their expectations are always on higher side, so more depression after failure

Unrealistic expectations: Guaranteed baby/
Male baby
- Many centers advertise/show unrealistic success rates for alluring the couple
- Since a large amount of money is involved they expect 100% result and expect miracles from doctors
- Indians have a great desire for male baby so many times center allure by false promises which later they don’t fulfill or deny
Stressed out patients looking for instant results

The prevalence of stress among women was 80%.

- Univariate analysis revealed that predictors of stress were years of marital life
- Duration and type of infertility
- History of gynecological surgery,
- Cycles of ovulation induction and intra-uterine inseminations,
- Present and past psychiatric morbidity, coping difficulties, gynecological diagnosis, and severity of premenstrual dysphoric disorder

Counselling

- Counseling: type of therapy to provide emotional and psychological support for a person who is undergoing certain difficulties, or experiences in life
- Well trained counselor is a must in IVF setting
- The following situations are usually addressed during IVF counseling:
  - Implications of treatment
  - Failed IVF treatment
  - Succeeding attempt after unsuccessful procedure
  - Successful IVF treatment and successful birth
  - Miscarriages
  - Handling mixed reactions from other sources
  - Alternative options

Inadequate Counseling

- Counseling has an important role
- COUNSELING not only informs the couple of procedure, side effects and complications but also addresses their questions and apprehensions
- Inadequate time & importance to counseling increases patients stress manifolds
- Individualized counseling after failures has to be emotionally supportive as well as informative about cause of failure and chances of success in future attempts
How to prevent ART drop out

- Better success rates
  - proper case selection
- Patient-friendly treatment
- Cost-effective treatment:
  - dropout rate is lower with antagonist cycle and mild ovarian stimulation, in contrast to the traditional agonist cycle with standard stimulation dose
- Continuity of care delivered by the same team consisting of the doctor, the nurse and the counselor

Misinformation and Lack of Transparency

- Availability of 24 X 7 communication and counselling as and when necessary give the couples necessary emotional support and encourages them to come for repeated attempts of treatment
- Discounts on subsequent cycle
- Lower professional fee
- Help from the government and charitable organisations
- Cryopreservation of good quality surplus embryos

THANK YOU
6. Expecting the unexpected in IVF lab?
FUNCTIONING OF AN IVF LAB

You can’t improve what you cannot measure!

The Vienna consensus: report of an expert meeting on the development of art laboratory performance indicators††

ESHRE Special Interest Group of Embryology† and Alpha Scientists in Reproductive Medicine‡
Failure to retrieve gametes

(SEMEN SAMPLE)
- FROZEN SAMPLE
- COUNSELING
- ICSI
- OOCYTE FREEZING

(OOCYTES)
- No oocytes are UF
- Semen sample

What to do?
- Flush follicles
- Check aspiration pump before frozen/thawed oocyte transfer
- Check reproductive hormones: progesterone, estradiol

FROZEN OOCYTES IN CASE OF DONOR OPU
You can't improve what you cannot measure!
Osmolarity

- The optimal osmolarity for pre-implantation embryos is 260 mOsm/L.

<table>
<thead>
<tr>
<th>Company</th>
<th>Medium</th>
<th>Slurrya</th>
<th>Osmolarity range (mean) mOsm/kg</th>
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<tbody>
<tr>
<td>LeicaTec® IVF/IVF®</td>
<td>AlcLys</td>
<td>5%</td>
<td>208-270 (2020)</td>
</tr>
<tr>
<td>Immucyte</td>
<td>Clearance 5%</td>
<td>8%</td>
<td>208-292 (2020)</td>
</tr>
<tr>
<td>Origio® (MediLab)</td>
<td>Universal IVF</td>
<td>Clearance 5%</td>
<td>277-295 (2020)</td>
</tr>
<tr>
<td>DM14®</td>
<td>Clearance 5%</td>
<td>8%</td>
<td>272-298 (2020)</td>
</tr>
<tr>
<td>Embryo3®</td>
<td>Clearance 5%</td>
<td>8%</td>
<td>272-298 (2020)</td>
</tr>
<tr>
<td>BlastAssist®</td>
<td>Clearance 5%</td>
<td>8%</td>
<td>272-298 (2020)</td>
</tr>
<tr>
<td>Vitrolabo®</td>
<td>G-1™ (G2 series)</td>
<td>Clearance 5%</td>
<td>256-266 (2011)</td>
</tr>
<tr>
<td>G-2™ with HESA (G2 series)</td>
<td>Clearance 5%</td>
<td>249-257 (2011)</td>
<td></td>
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<tr>
<td>G-3™ (G2 series)</td>
<td>Clearance 5%</td>
<td>8%</td>
<td>255-303 (2009)</td>
</tr>
<tr>
<td>G-4™ with HESA (G2 series)</td>
<td>Clearance 5%</td>
<td>248-258 (2009)</td>
<td></td>
</tr>
</tbody>
</table>

Source: Methods Mol Biol. 922, 125-140. doi: 10.1007/978-1-59745-975-6_5

Factors affecting osmolarity during drop preparation:

- Volume
- Temperature
- Airflow
- Oil covering

- G2HTE R2
- Drop volume
- LAV OF
- Quiet stirring
- WASH DROPS recommended

Changes in osmolarity:

- Embryos change their volume
- Cystadone damage
- Gene expression
- Gene imprinting
A precise control over pH is essential:

- Embryonic development (Luan et al., 2000)
- Cell differentiation and growth (Van den Ouweland & De Waele, 2000)
- Cell division and proliferation (Eckardt, 1996)
- Protein and DNA synthesis (Eckardt, 2001)
- Membrane transport (Evans, 1993)
- Secretion, calcium level modulation and cytoskeletal dynamics (Deregibus, 2001)

Currently 7.35 ± 0.05 in an environment of 5-6% CO2 (Siddique, 2004; Osiris, 2004)

The pH is a logarithmic scale — 0.2 unit decrease = 50% of IVF more
pH variations < 0.2 affect:

- Gene expression
- Cell number of the blastocyst
- Implantation

pH
How to improve my ART outcome: Quality Control / Quality Assurance (QA/QC)

<table>
<thead>
<tr>
<th>Cell Stage</th>
<th>pH (Philips et al. 2006)</th>
<th>pH (Dale et al. 2008)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Germinal vesicle (intact)</td>
<td>7.04 ± 0.07</td>
<td>7.3 ± 0.3</td>
</tr>
<tr>
<td>Metaphase I oocyte</td>
<td>7.03 ± 0.04</td>
<td>N.A.</td>
</tr>
<tr>
<td>Metaphase II oocyte</td>
<td>6.98 ± 0.02</td>
<td>7.4 ± 0.1</td>
</tr>
<tr>
<td>Metaphase II oocyte (aged)</td>
<td>N.A.</td>
<td>7.5 ± 0.2</td>
</tr>
<tr>
<td>3 Pronuclei oocyte</td>
<td>N.A.</td>
<td>7.4 ± 0.1</td>
</tr>
<tr>
<td>2-8-cell embryo</td>
<td>7.12 ± 0.01</td>
<td>N.A.</td>
</tr>
</tbody>
</table>

**Irvine**  | **Sage**  | **Vitrolife** | **Life Global** | **Origio** | **Cook** |

**Time log for pH of 50 uL and 500 uL medium under oil:**

**Time log for pH of 50 uL under pre-equilibrated oil:**

- pH: sub-optimal cut 3 min outside the incubator
- No activity should take longer than 3 min
- A stop watch is an embryologist’s best friend
TEMPERATURE

CONTROL:
- During follicular aspiration;
- During transport of embryos;
- During "bag search" and handling at OOL;
- During IVF.

Impact of different culture temperature on human embryo development:

<table>
<thead>
<tr>
<th>Temperature</th>
<th>Blastocyst rate %</th>
<th>Embryo grade</th>
<th>Implantation rate %</th>
</tr>
</thead>
<tbody>
<tr>
<td>37°C</td>
<td>65.7</td>
<td>A (65.7)</td>
<td>60.5</td>
</tr>
<tr>
<td>31.9°C (1.5 min)</td>
<td>60.5</td>
<td>B (60.5)</td>
<td>55.1</td>
</tr>
<tr>
<td>27.1°C (5 min)</td>
<td>55.1</td>
<td>C (55.1)</td>
<td>50.1</td>
</tr>
<tr>
<td>37°C (3 min)</td>
<td>50.1</td>
<td>D (50.1)</td>
<td>45.1</td>
</tr>
<tr>
<td>37°C (1 min)</td>
<td>45.1</td>
<td>E (45.1)</td>
<td>40.1</td>
</tr>
<tr>
<td>37°C (20 min)</td>
<td>40.1</td>
<td>F (40.1)</td>
<td>35.1</td>
</tr>
</tbody>
</table>

*Note: The study was conducted under controlled laboratory conditions, allowing for consistent and controlled results. The data reflects the average performance of embryos exposed to different temperatures.*

Spindle dynamics in relation to changes in temperature:

Failed fertilization
Aborted fertilization
Aneuploidy
Low embryo development
Fragmentation
Gene expression
Implantation

Spindle disassembly is temperature- and time-dependent.
Rigorous thermal control during intracytoplasmic sperm injection stabilizes the meiotic spindle and improves fertilization and pregnancy rates

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**Table 1:** Impact of Temperature Control on IVF Outcomes

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Sperm 1</th>
<th>Sperm 2</th>
<th>Sperm 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>35.1 ± 3.4</td>
<td>35.1 ± 4.6</td>
<td>36.1 ± 3.4</td>
</tr>
<tr>
<td>Temperature control</td>
<td>46.1 ± 1.4</td>
<td>36.1 ± 2.8</td>
<td>46.1 ± 1.6</td>
</tr>
</tbody>
</table>

**Figure 1:** Influence of temperature on sperm motility and acrosome reaction. (A) Sperm motility as a function of incubation time (minutes) at various temperatures (0°C vs. 37°C). (B) Percentage acrosome reaction (AR) as a function of incubation time (minutes) at various temperatures (0°C vs. 37°C).
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**IMPORTANCE OF AIR QUALITY IN IVF**

Evidence from Human Studies

<table>
<thead>
<tr>
<th>Study</th>
<th>Population Description</th>
<th>Method Details</th>
<th>Outcome/Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Huang et al., 2006</td>
<td>IVF cycles</td>
<td>Timing culture in incubators with and without VOC exposure.</td>
<td>Higher IVF cycle success rates were observed in incubators with VOC exposure.</td>
</tr>
<tr>
<td>Ouyang et al., 2009</td>
<td>IVF cycles</td>
<td>Timing culture in incubators with and without VOC exposure.</td>
<td>Higher IVF cycle success rates were observed in incubators with VOC exposure.</td>
</tr>
<tr>
<td>Nozawa et al., 2008</td>
<td>IVF cycles</td>
<td>Reduction in miscarriage rates observed in cycles performed in incubators with VOC exposure.</td>
<td></td>
</tr>
<tr>
<td>Xie et al., 2003</td>
<td>IVF transfer</td>
<td>Reduction in air pollutants associated with an increase in the number of high-quality embryos.</td>
<td></td>
</tr>
</tbody>
</table>

**GAS COMPOSITION**

- Medical grade CO₂
  - Specification: Medical carbon dioxide specification is.
  - Carbon dioxide purity: 99.5 % ± 0.5 %

The medical carbon dioxide cylinder specification complies with the current European Pharmacopeia monograph (EP 3).

**Effects of reduced oxygen concentration in a predominantly male-biased transfer program**

<table>
<thead>
<tr>
<th>Oxygen Level</th>
<th>Clinical Pregnancy</th>
<th>Implantation</th>
<th>Live Birth</th>
</tr>
</thead>
<tbody>
<tr>
<td>21% O₂ (%)</td>
<td>961/15 (64.4%)</td>
<td>905/15 (60.5%)</td>
<td>65/115 (57.8%)</td>
</tr>
<tr>
<td>5% O₂ (%)</td>
<td>641/15 (46.4%)</td>
<td>122/29 (41.4%)</td>
<td>48/115 (42.0%)</td>
</tr>
</tbody>
</table>

Source:([1](#))

- Reduced embryo development
- Increased fragmentation
- Zona hardening
  - Inhibited fertilization
  - Reduced implantation
  - Increased fragmentation

**REDUCE O₂**

- Include antioxidants in media
- Reduce oxygen tension
- Use reduced oxygen pressure (high EOS generator)

**5% O₂**

- Enhanced growth
- Improved development
- Increased implantation success

**20% O₂**

- No development
- Reduced implantation
- Decreased success
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Microbiological Monitoring

- **Settle plates**
  - Mold and blood-agar Petri dishes
  - Surface sampling from table, floor, air, countertops
  - Five-finger glove printing
  - Incubators, workstations, air, floor, wall

### WORK AREA
- Is the cell culture hood properly set up?
- Is the cell culture hood in an area free from drafts and through traffic?
- Is the work surface uncluttered, and does it contain only items required for your experiment?
- Did you wipe the work surface with appropriate disinfectant before work?
- Are you regularly cleaning and sanitizing your incubators, refrigerators, thawers, and other laboratory equipment?

### PERSONAL HYGIENE
- Did you wash your hands?
- Are you wearing personal protective equipment?
- Do you wear rings and jewelry?
- If you have long hair, is it tied in a cloth cap?
- Are you using a pipette to work with liquids?

### REAGENT AND MEDIA
- Have you sterilized any reagents, media, and solutions you have prepared in the laboratory using the appropriate procedures?
- Did you wipe the outside of the bottles, flasks, and plates with disinfectant before placing them on your work surface?
- Are all your bottles, flasks, and other containers capped when not in use?
- Are all your plates stored in sterile re-sealable bags?
- Do you use media after the expiration date?

### HANDLING
- Are you working slowly and deliberately, mindful of aseptic technique?
- Did you wipe the outside of the bottles, flasks, and plates with disinfectant before placing them on your work surface?
- Are the caps or covers face down on the work area?
- Are you using sterile glass pipettes or sterile disposable plastic pipettes to manipulate all liquids?
- Are you using a sterile pipette tip only once to avoid cross contamination?
- Are you careful not to touch the pipette tip to anything non-sterile, including the outside of the bottle threads?
- Did you mop-up any spillage immediately, and wiped the area with disinfectant?
THANK YOU
7. What lies between an average and successful IVF program
How to improve my ART outcome

Quality Control / Quality Assurance (QA/QC)

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CLINICAL SCENARIO 1

A successful IVF clinic started a new lab in a new facility and it reported low results for the initial months ... what could go wrong?

- Location
- Material used in construction? Flooring/adhesive
- Design flaw? Difficulty in maintenance!!
- Positive pressure ventilation?
- HEPA filters...inbuilt? CODA?
- Gas supplies same as parent?
- Personnel

These flaws can all contribute to greater circulating levels of volatile organic compounds (VOCs), and other contaminants inside an IVF lab. VOCs can be harmful to the growing embryos and have been found to severely reduce success rates.
Air Quality

- Air quality/VOCs likely impact embryo development
- Prudent to take preventative measures (Standard of care)
- HVAC VOC filtration
  - dedicated, proper intake placement, # exchanges, filters, etc
- Inline gas filters
- Appropriate design & building materials
- Burn-in & off-gassing before use

Burn-In & Validation

- Clean walls, ceilings, floors, cabinets/counters.
- Clean equipment (incubators)
  - Increasing temperature of the new area by 10-20°C or even higher temperatures
  - Increasing the ventilation rate which aid in the removal of VOC
  - Lab should be left closed during this time
  - AHUs and lighting switched on and left running

- Area purged with high ventilation repeatedly neutralises the offgassing by paint, adhesives, general building materials
- Run HVAC and incubators for ≥ ~2 weeks
- Validate
- Particle counts, VOCs
- Activated Charcoal in SS buckets
- Document the "commissioning" of the lab
INCUBATORS

- Purchase months in advance of their intended use
- Run decontamination program
- Microbial sampling for aerobic bacteria and fungi
- VOC levels determined
- Inline filters
- Activated charcoal granules

Flaws in AHU designing
**CLINICAL SCENARIO 2**

- A 26 yr old pt with regular cycles BMI 24
- Left salpingectomy done in 2014 for ectopic pregnancy
  HSG showed rt tubal cornual blockage
- Husband semen analysis normal
- TVS shows polycystic ovaries bilaterally
  - AMH 6
  - FSH 3.5
  - LH 10
  - E2 50
  - TSH 2.8
- Declined hysteroscopic tubal canulation and laparoscopy
- Decided to have IVF treatment

---

- OCP prescribed for 21 days.
- Antagonist protocol with 150 IU of Rec FSH prescribed for 10 days
- Cetrotix as antagonist
- 22 follicles developed E2 2618
- Triptorelin 0.2 mg given as trigger 35 hrs before OPU
- Follicle Sizes:
  - 17-18mm 4
  - 15-16mm 5
  - 12-14 mm 8
  - 8-10mm
- ET 9mm
- Egg retrieval from right ovary attempted
- 4 follicles aspirated: 2x15-16 and 2x13-14
- No eggs retrieved

---

- What will you do
  - Continue collection or stop
  - What will you ask
- 3 more follicles aspirated 1x17-18mm 1x15-16, 1x13-14.
- No eggs retrieved
- What will you do next
  - Abandon
  - Continue and aspirate all the follicles
  - All follicles aspirated and no eggs retrieved
• First step
  - Check date and time of trigger
  - Time interval between trigger and antagonist
  - Was the trigger supervised

• Second step
  - Consider flushing the follicles
  - Check suction pump/vacuum pressure/flow rate

• Third step
  - Stop collection
  - Give rescue HCG and collect 36 hrs later

How will you counsel this patient?
Patient wants to have IVF again
What is your action plan
  - Protocol
  - Dose of drug
  - Day of collection earlier or later
  - Trigger
Plan for next cycle
  - Antagonist cycle
  - Reduce dose of drug to 125
  - Step up if required
  - Dual trigger using GnRh agonist at 40 Hrs with HCG 1500 IU at 34 hrs
Empty follicle syndrome: Successful pregnancy following dual trigger
K. Gangula, Suchita Rathore, Ajay Garg, and Kumar Rao

CLINICAL SCENARIO 3

An IVF center suddenly has doubled their number of cases per month but reporting lower pregnancy rates… what could be the reason?

- The number of incubators were same, stability of incubators compromised, not withstanding the load of the cases couldn't maintain the co2 and temp properly
- The number of embryologist wasn't increased leading to stressful work conditions and over worked staff, resulting compromised total quality management in the lab
- We need to follow the staffing norm and think how many staff of which categories are required in the ART facility for target level of utilization?
- However, for an ART clinic a strong element of success is dependent on the availability of the right quantity of the right categories of staff
How to improve my ART outcome

Quality Control / Quality Assurance (QA/QC)

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McCulloh: Staff: 0.47 + Number of IVFs / 88.5
Boone & Higdon: 2.92 + number of procedures x 0.002

Table 2.3  Staffing norms for ART facilities in the United States

<table>
<thead>
<tr>
<th>Number of IVFs</th>
<th>McCulloh</th>
<th>Boone and Higdon</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.47</td>
<td>2.92</td>
</tr>
<tr>
<td>100</td>
<td>1.6</td>
<td>4.3</td>
</tr>
<tr>
<td>250</td>
<td>3.3</td>
<td>6.4</td>
</tr>
<tr>
<td>400</td>
<td>5.0</td>
<td>8.5</td>
</tr>
<tr>
<td>1000</td>
<td>11.7</td>
<td>16.9</td>
</tr>
</tbody>
</table>

CLINICAL SCENARIO 4

- 28 yr old patient operated for grade 4 endometriosis and endometrioma excision done in 2016
- In the OT note Bilateral spillage from tubes mentioned
- Patient has been trying to conceive for 1 yr
- TVS shows recurrent endometrioma 2cms on rt side and 3.5 cms on the left side. Hydrosalpinx not noted
  - AFC 8
  - AMH 1.2
  - FSH 8
  - LH 9.5
  - E2 65

- How would You like to proceed?
  - Natural cycle folliculometry and TI
  - Ovulation induction folliculometry
  - Stimulated IUI
  - IVF
• If IUI
  - what protocol and how many cycles
  - 2 cycles of IUI have failed pt now wants to go for IVF
• Would you operate on her before IVF
• Which protocol would you prefer

• Options of either stimulated IUI for 3-4 cycles or IVF
• Counsel women with endometrioma regarding the risks of reduced ovarian function after surgery and the possible loss of the ovary.
• The decision to proceed with surgery should be considered carefully if the woman has had previous ovarian surgery
• Use assisted reproductive technologies for infertility associated with endometriosis, esp. if tubal function is compromised or if there is male factor infertility, or other treatments have failed

FOGSI
• In stage I and II endometriosis, treatment with super ovulation and IUI improve fertility compared to expectant management
• Clinicians should take into consideration, age, duration of infertility, ovarian reserve and male factor (16). [Evidence level A]
• Previous ovarian surgery results in longer stimulation, higher FSH requirement, decreased oocyte number but no difference in fertilization, pregnancy outcome in subsequent ART cycles
• Surgical management of endometrioma does not significantly increase IVF pregnancy rate and ovarian response to stimulation compared to no surgery
• Check LH level on day 5-6 of stimulation
  Antagonist required only if LH levels not suppressed
  Or top up with agonist Lupride on daily basis (long protocol)
  Slow growth of follicles on day 8 with rec FSH may indicate need for Rec LH or HMG (Min dose 150 IU)

• COS using GnRh agonists or antagonists is effective in IVF patients with mild to moderate endometriosis and in those with endometrioma who did not undergo surgery [Evidence level A]
  Ultra-long protocol of GnRh agonists for a period of 3 – 6 months before ART improves the clinical pregnancy rates
  In women undergoing IVF, stage III and IV is associated with poor implantation and lower clinical pregnancy rate
  In infertile women with endometrioma smaller than 3 cm cystectomy prior to ART does not improve pregnancy rates
  In women with endometrioma larger than 3 cm, cystectomy is indicated prior to ART when it is associated with pain or inaccessibility of follicles

• Pt was given 2 cycles of GnRH agonist on 21/02/18 and 21/03/18 and then underwent antagonist cycle from 16/04/18 using 450 IU of Rec FSH
  - Is an antagonist necessary
  - What else could have been done
  - Would you like to change the stimulation protocol or continue if so why

  Rec FSH continued
  Slow growth of follicles noted on day 8
  11 mm on day 6 and 12 mm on day 8
  What can we do? Continue same or anything different?
On day 14, 8 follicles noted
- 2x17-18mm
- 2x 15-16
- 3x 14-15
- 1x12

Rec HCG given as trigger and egg Collection done 35 hrs later

- 7 brown eggs were collected
- 5 M 11
- 4 Fertilised
- Day 3
  - 2x Grade 2 8cell
  - 2x grade 3 6 cell
- To transfer or freeze
- Which day Day 3 or 5
How to improve my ART outcome Quality Control / Quality Assurance (QA/QC) Indian Fertility Society & Origio India Initiative

• Navid Esfandiari, D.V.M., Ph.D., H.C.L.D.
• Hasan Burjaq,

High serum FSH levels during the IVF-ET cycle were correlated with the formation of brown oocytes and affected embryo development, leading to a decrease in the pregnancy rate.

In contrast, no significant correlation was observed between brown oocytes formation and patients’ conditions like age, BMI, or duration and causes of infertility.

Therefore, limiting the FSH dose during controlled ovarian hyperstimulation is important for the production of high-quality oocytes.
CLINICAL SCENARIO 5


The embryologist moves to UK for higher education. The centre hires a fly by night embryologist who does cases in batches and carries his own disposables and media. The results suddenly fall and centre earns a bad name

Question 1. What probably went wrong at this laboratory? Do you think the disposables and media were the culprit?

Question 2. What is the importance of high quality IVF grade plastic?
Gametes and embryos are never exposed to a substance that will deleteriously impact their environment.

Exposure can occur during gamete acquisition, via culture in media, or via plastic ware with which the medium has come in contact, or via any airborne volatile or nonvolatile agent that may affect the culture material either directly or indirectly.

Great variability exists between different manufacturers of certain plastic ware as well as between different lots of the same item.

Culture of human gametes and embryos is performed in a plastic ware.

Purchase plastic ware products that have already tested non-toxic using a mouse embryo assay (MEQA).

Table 2.6 Contact materials and availability of prior mouse embryo testing

<table>
<thead>
<tr>
<th>Item</th>
<th>Available with prior mouse embryo assay?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Culture medium</td>
<td>Yes</td>
</tr>
<tr>
<td>Culture dishes</td>
<td>Yes</td>
</tr>
<tr>
<td>Embryo transfer catheters</td>
<td>Yes</td>
</tr>
<tr>
<td>Pipettes, serological</td>
<td>No, requires testing</td>
</tr>
<tr>
<td>Pipettes, micropipettes for moving embryos</td>
<td>Yes</td>
</tr>
<tr>
<td>Micropipets for ICSC, A7/F, and holding</td>
<td>Yes</td>
</tr>
<tr>
<td>Centrifuge tubes</td>
<td>No, requires testing</td>
</tr>
<tr>
<td>Culture tubes</td>
<td>No, requires testing</td>
</tr>
<tr>
<td>Pipette tips</td>
<td>Yes</td>
</tr>
<tr>
<td>Gas for maintenance of CO2</td>
<td>No, requires testing</td>
</tr>
<tr>
<td>Filters for gas or medium</td>
<td>No, requires testing</td>
</tr>
<tr>
<td>ICSI, intracytoplasmic sperm injection A7/F, assisted zona</td>
<td></td>
</tr>
</tbody>
</table>

PLASTICWARE AND VOCs

Table V. Compounds released from cell tissue culture grade petri dishes

<table>
<thead>
<tr>
<th>Material</th>
<th>&gt;50 mg/sample</th>
<th>≤50 mg/sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sulfuric acid</td>
<td>920.00</td>
<td>n-Butanol</td>
</tr>
<tr>
<td>Toluenes</td>
<td>180.00</td>
<td>3-Methylbutane</td>
</tr>
<tr>
<td>Acetone</td>
<td>150.00</td>
<td>Normal</td>
</tr>
<tr>
<td>2-Butanone</td>
<td>1,300</td>
<td>Butanol</td>
</tr>
<tr>
<td>Acetone dihydrate</td>
<td>100</td>
<td>3-Pentanone</td>
</tr>
<tr>
<td>n-Butane</td>
<td>100</td>
<td>n-Hexane</td>
</tr>
<tr>
<td>Benzaldehyde</td>
<td>100</td>
<td>Benzene isomer</td>
</tr>
<tr>
<td>Hexanal</td>
<td>70</td>
<td>n-Octane</td>
</tr>
<tr>
<td>Ethylbenzene</td>
<td>64.00</td>
<td>n-Nonane</td>
</tr>
<tr>
<td>2-Heptanone</td>
<td>38.00</td>
<td>Decanal</td>
</tr>
<tr>
<td>Cinnamaldehyde</td>
<td>10</td>
<td>Camphene</td>
</tr>
<tr>
<td>Propylbenzoate</td>
<td>10</td>
<td>Octanal</td>
</tr>
<tr>
<td>n- &amp; β-Xylenes</td>
<td>7.5</td>
<td>o-Xylene</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5.80</td>
</tr>
</tbody>
</table>
CLINICAL SCENARIO 6

- 28 yr old patient proceeding for IVF.
- Husband semen analysis
  - Count 4mill/ml
  - Motility
    - Rapid progressive 4%
    - Sluggish progressive 20%
    - Normal forms 4%
- Preparing for ICSI
- Husband working in UK coming home for 3 to 4 days
- Semen banking not done
- Not agreeable to have donor semen backup
- No erectile problems identified

- Pt stimulated with 225 IU of rec FSH using antagonist protocol
  - 12 follicles stimulated
  - ET 8mm
- On day of egg retrieval
  - Semen count 1mill/ml
  - Rapid progressive 0%
  - Sluggish progressive 4%
  - Normal forms nil%
- What is to be done?
- 2nd sample
- HOS test
- Sperm Mobil Media (pentoxiphylline / theophylline)
- Laser assisted immotile sperm selection (LAISS)
- TESE

Kovacic et al., (2006) reported the largest series (47 cycles) using testicular sperm, with a fertilization rate of 66% and a pregnancy rate of 38.3%.

Kahraman et al. (1997) reported the first pregnancy and delivery of a healthy child after ICSI treatment with immotile testicular spermatozoa in a patient with absolute asthenozoospermia.
Testicular sperm were recovered in all treatment cycles and fertilization occurred in six of seven cycles. Overall normal fertilization and transfer rates were 67% and 71%, respectively. One live birth was obtained after five ETs.

CLINICAL SCENARIO 7

An old centre with huge workload notice that their results are plummeting. An experienced embryologist advices them to hold audits to see where they are heading?

- What should be the frequency of audits?
- What parameters should be analysed?
- Internal / External audits

INTERNAL AUDITS

- Continuous Monitoring of Laboratory Performance in an Established IVF Unit
- Performance of the IVF laboratory can be monitored by auditing
  - Fertilization rates
  - Cleavage rates
  - Embryo quality
  - Pregnancy rate
  - Implantation rate, and
  - Multiple birth rates
- Frequency should be depending on the center's activity.
- However, when interpreting these statistics, consideration must be given to the activity
- of the unit and the types of patients treated, etc.
Furthermore, pregnancy rate alone should not be used as a QC measure to audit the performance of the laboratory. This is not least because it is affected by other variables, but also because a drop in pregnancy rate identifies a problem encountered at the time of treatment usually weeks before. A good QC program detects problems before they affect the pregnancy rate. Moreover, using pregnancy rate as a QC measure does not identify the source of the problem.

- Record Keeping and Documentation
- Fundamental to the success of an QA program is accurate documentation

---

Table 3.5: Kit of equipment parameters for QC and Frequency of QC

<table>
<thead>
<tr>
<th>Equipment</th>
<th>Frequency of QC</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Equipment</td>
<td>Daily</td>
<td>Maintained</td>
</tr>
<tr>
<td>Temperature</td>
<td>Daily</td>
<td>Room temperature</td>
</tr>
<tr>
<td>Light source</td>
<td>Daily</td>
<td>Radiation</td>
</tr>
<tr>
<td>Vacuum system</td>
<td>Daily</td>
<td>Vacuum pressure</td>
</tr>
<tr>
<td>Gas supply</td>
<td>Daily</td>
<td>Gas flow</td>
</tr>
<tr>
<td>Water supply</td>
<td>Daily</td>
<td>Water pressure</td>
</tr>
<tr>
<td>Ventilation</td>
<td>Daily</td>
<td>Air flow</td>
</tr>
<tr>
<td>QC equipment</td>
<td>Monthly</td>
<td>Temperature, pressure, flow</td>
</tr>
<tr>
<td>QC parameters</td>
<td>Quarterly</td>
<td>Temperature, pressure, flow</td>
</tr>
</tbody>
</table>

- DIALOGUE AND INTERACTION BETWEEN THE CLINICIAN AND THE EMBRYOLOGIST
- External audits
  - The assessment of internal controls, processes, and documentation is oriented more to a certification process than a standard audit.
  - This is all the more reason to ensure that the organization is fully prepared for the auditing experience, and that the compliance process works well and is successful.
CLINICAL SCENARIO 8

- 37 year old pt with BMI 36 recently married is seen in your clinic wanting to conceive quickly
  - AMH 0.6
  - FSH 12
  - LH 5
  - E2 86
  - TSH normal prolactin normal
  - AFC 4
- Husband's semen analysis
  - Count 8 mill
  - rapid progressive 40%
  - normal forms 4%
- Proceeding for IVF
- How would you prepare her to optimise response?

- Weight loss
- DHEA for 3 months
- For Husband
- Check for varicocele and any anatomical problems
- Lifestyle modification
- Antioxidants

- TVS Day 2 one Follicle of 11mm and rest of the three follicles 8-9mm
- How will you pretreat this pt before IVF treatment
- To deal with follicular asynchrony
- OCP
- Estradiol pretreatment in luteal phase
What protocol would you like to use
- Microflare
- Antagonist
- HMG or rec FSH
- Pt was stimulated using 300 IU HMG
- 4 follicles developed and 3 eggs retrieved on day 12
- Follicle size
  - 18mm
  - 16mm
  - 15mm
  - 14mm
- ET 6.5mm
- 3 eggs obtained
- IVF done
- 2 eggs being fertilised by multiple sperms (polyspermy)
- What are the implications
- How do we modify cycle for next attempt?

What are the implications of polyspermy?
How do we modify cycle for next attempt?
- Rec FSH or HMG
- Which dose
- Stimulation protocol
- Day of trigger
Stimulation

Consider dual stimulation for poor ovarian response patients to maximise egg collection.
Dose Of Gonadotrophins

- Current evidence supports a maximum daily dose of 300 IU of rFSH in the expected POR patient as higher doses do not increase neither the clinical pregnancy rate nor the live birth rate

Polyspermy

- Polyspermy occurs due to poor egg quality and possible postmaturity
- In older patients better to trigger at 16mm follicle in such cases

Day of Retrieval

- Follicles of POA/pOFI patients biologically as well as clinically, indeed, behaved very similarly to previously reported older women above age 43 years
- More mature (81.5 ± 4.5 vs. 55.8 ± 8.3%, P < 0.05) and fewer atretic oocytes (7.9 ± 3.0% vs. 28.2 ± 7.2%, P < 0.05) were obtained with ER t 16mm follicles than SR, while immature oocyte numbers were similar
- These observations suggest PL of follicles with SR

Stimulation

- If endometrial development poor consider freezing embryos. Inform about post thaw survival of embryos.
- If progesterone levels low on day of HCG consider fresh transfer if endometrial thickness optimal
CLINICAL SCENARIO 9

In a change of scenario, if you were an infertility patient, how would you select an IVF centre, what would be your criteria of a good clinic?

- It takes time and patience to select a successful IVF center.
- When you screen potential IVF sites, ask questions! Research the clinics in your area.
- Compare the various sites based on different qualities such as:
  - Are the physicians and embryologists qualified enough?
  - What specialties are they certified in?
  - Do the clinics use accredited labs?
  - What types of amenities and services are offered to patients?

IVF labs should maintain the best quality standards and should adopt standard operation protocols confirming to the practices followed globally to ensure the best of results and get their labs accredited.

- Get your IVF clinic registered with the National Registry for IVF clinics and ART banks managed by the Indian Council of Medical Research (ICMR)

- The auditing covers not just the Laboratory and Medical management areas but also patient safety, disaster management, Patient relations, grievance handling, counseling, administrative and HR protocols.

- The certification proves that the clinic was already 99% prepared and the audit help us improve and achieve 100% benchmark."
CONCLUSIONS

- There is no single “best” way to do anything in the IVF lab.
- Make your choices according to careful analysis of
  - Ability to control the operational characteristics (equipment)
  - Efficiency and simplicity of the procedural steps
  - Ability to minimize mistakes: reduce process and operator errors
  - Ability to optimize the requisite control variables
  - Ability to minimize the impact of adverse factors
- Define Indicators and select / set Benchmarks, then monitor your system processes and outcomes

Stimulation

THANK YOU
Indian Fertility Society & Origio India Initiative