How to improve ART outcomes?

X.TURKISH-GERMAN GYNECOLOGY CONGRESS
ANTALYA, TURKEY/2014

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Ufuk University Faculty of Medicine
Gynecology and Obstetrics Department
For better outcome?

- Age (<37)
- First cycle
- High quality embryo at previous cycle
- Normal uterine cavity and absence of hydrosalphinx
- Absence of endometriosis
- Absence of endometrial factor
- Absence of abnormal gamet morphology
- High quality embryo for freezing
How to improve ART outcomes?
- Ovarian reserve evaluation
- Immunological screening
- A ‘must’ before ART?
- **Oocyte factor**
  - Age, smoking
  - Obesity, PCOS
  - Endometrosis
  - Ovarian surgery

- **Adjuvant therapies**
  - DHEA
  - Luteal Phase Support
  - GH

- **Outcome measurements**
  - Progesteron
  - Estradiol
- Embryo transfer

- Endometrial factor

- timing?
- tecnique?

- USG
- Scratch
OVARIAN RESERVE EVALUATION
Ovarian reserve: AMH, D3 FSH, E2, inhibin B

AMH < 1.96 ng/mL $\rightarrow$ (<4 oocytes) $\rightarrow$ 300 IU/day

AMH > 4.2 ng/mL $\rightarrow$ 150 IU $\rightarrow$ 100-125 IU/day
High OHSS risk

Low FSH dose (125 IU/gün)

GnRH ant. protocol

Agonist trigger

Gonadotropin dosage should be decided according to age

AMH < 0.01 ng/mL
Poor reserve
FSH < 15 IU/L
High dose gonadotropin

AMH 1.96 - 4.2 ng/mL

AMH > 4.2 ng/mL
High OHSS risk
Low FSH dose (125 IU/gün)
GnRH ant. protocol
Agonist trigger
Antral Follicle Count

- AFC <4 → cycle cancellation 37x
- 3D and 2D USG same but AFC is less effective for predicting ART outcome.
- AMH and AFC have similar efficacy for predicting pregnancy outcome.

Gibreel A, Hum Fertil 2009
Ovarian Reserve Tests (ORT)

- AMH, is better than D3 FSH, estradiol and inhibin B for predicting ART outcome.

- For predicting high response AMH >FSH, AFC
- For predicting poor response AMH=AFC>FSH

Seifer DB, Fertil Steril, 2002
Hazout A, Fertil Steril, 2004
Muttukrishna S, BJOG, 2004
Penarrubia J, Hum Reprod, 2005
Tremellen KP, Obstet Gynecol, 2005

Nardo LG, Fertil Steril, 2009
IMMUNOLOGICAL SCREENING
<table>
<thead>
<tr>
<th>Autoantibodies</th>
<th>Frequency in infertile women</th>
<th>Correlation with Infertility</th>
<th>Other</th>
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<tbody>
<tr>
<td>Antiphospholipid</td>
<td></td>
<td>-</td>
<td>Habituel abortion</td>
</tr>
<tr>
<td>Antithyroid</td>
<td>Minimally</td>
<td>-</td>
<td>Tiroiditis, abortion</td>
</tr>
<tr>
<td>Antigliadin</td>
<td>Minimally</td>
<td>-</td>
<td>Celiac disease</td>
</tr>
<tr>
<td>Antisperm</td>
<td>No effect</td>
<td>-</td>
<td>Fertilisation failure</td>
</tr>
<tr>
<td>Antinuclear</td>
<td>Minimally</td>
<td>-</td>
<td>Otoimmune disease</td>
</tr>
<tr>
<td>Antiovarian</td>
<td>Minimally</td>
<td>-</td>
<td>Otoimmune disease</td>
</tr>
</tbody>
</table>
Immunologic screening Before ART

- APA screening is not necessary before ART!  
  \[\text{ASRM}\]

- APA(+) is not associated with poor outcome.  
  \[\text{Buckingham KL, J Reprod Immunol 2009}\]

- Anticoagulant therapy does not change outcome of APA(+) patients.  
  \[\text{ASRM, 2008}\]

- Routine thyroid autoantibody screening is not suggested.

- If habituel abortion history is positive; autoantibody evaluation may be valuable
Immunological screening
Before ART

Which patients should be screened?

- History of venous thromboembolism
- Presence of high risk trombophilia(+) at 1. degree relatives
- Venous thromboembolism (age of <50 (+))
Tyroid Functions? Antibodies? Before ART

**Thyroid Function Tests:**
- Anovulatuar and idiopathic infertile patients 5-6%
- Tubal and male factor (+) 2%

TSH, sT4 must be evaluated before ART
BEFORE ART:

H/S?
L/S?
Myomectomy?
Polypectomy?
HSG?
Is hysteroscopy routine before ART?

Not suggested !
• No signs
• For uterine cavity evaluation US/HSG/H/S
• Age <35, male factor (+), family history (-)

Hysteroscopy suggested !

1. Age >35, abnormal uterine bleeding, abnormal clinical symptoms
2. 2 IVF failure
3. Sign + (Intrauterine polyp, submucosal leiomyoma, uterine septum, Ashermans syndrome…).
Is laparoscopy routine before ART?

If the etiology of the infertility is clear it is unnecessary

Laparoscopy is suggested;

1. Presence of pelvic inflammatory disease
2. Presence of hydrosalpinx, endometriosis or endometrioma
Myomectomy before ART

- Intramural & >4 cm  distorting cavity/degenerated:
  Myomectomy
- Intramural & >5cm not distorting cavity:
  Myomectomy
- Subserosal & >5cm:
  Myomectomy
- Submucosal & >1cm Myoma / Polyp:
  Myomectomy

IM Leiomyoma: does not change pregnancy rates

SM leiomyoma: increases pregnancy rates but same abortion rates

Pritts et al, Fertil Steril, 2009
Fibroids and infertility: an updated systematic review of the evidence

Elizabeth A. Pritts, M.D., a William H. Parker, M.D., b and David L. Olive, M.D.a

a Wisconsin Fertility Institute, Middleton, Wisconsin; and b Department of Obstetrics and Gynecology, University of California, Los Angeles, California

### Table 6

Effect of myomectomy on fertility: submucosal fibroids.

<table>
<thead>
<tr>
<th>Outcome</th>
<th>Number of studies/substudies</th>
<th>Relative risk</th>
<th>95% confidence interval</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>A. Controls: fibroids in situ (no myomectomy)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clinical pregnancy rate</td>
<td>2</td>
<td>2.034</td>
<td>1.081–3.826</td>
<td><em>P</em> = .028</td>
</tr>
<tr>
<td>Implantation rate</td>
<td>0</td>
<td>—</td>
<td>—</td>
<td></td>
</tr>
<tr>
<td>Ongoing pregnancy/live birth rate</td>
<td>1</td>
<td>2.654</td>
<td>0.920–7.658</td>
<td>Not significant</td>
</tr>
<tr>
<td>Spontaneous abortion rate</td>
<td>1</td>
<td>0.771</td>
<td>0.359–1.658</td>
<td>Not significant</td>
</tr>
<tr>
<td>Preterm delivery rate</td>
<td>0</td>
<td>—</td>
<td>—</td>
<td></td>
</tr>
<tr>
<td><strong>B. Controls: infertile women with no fibroids</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clinical pregnancy rate</td>
<td>2</td>
<td>1.545</td>
<td>0.998–2.391</td>
<td>Not significant</td>
</tr>
<tr>
<td>Implantation rate</td>
<td>2</td>
<td>1.116</td>
<td>0.906–1.373</td>
<td>Not significant</td>
</tr>
<tr>
<td>Ongoing pregnancy/live birth rate</td>
<td>3</td>
<td>1.128</td>
<td>0.959–1.326</td>
<td>Not significant</td>
</tr>
<tr>
<td>Spontaneous abortion rate</td>
<td>2</td>
<td>1.241</td>
<td>0.475–3.242</td>
<td>Not significant</td>
</tr>
<tr>
<td>Preterm delivery rate</td>
<td>0</td>
<td>—</td>
<td>—</td>
<td></td>
</tr>
</tbody>
</table>

Myomectomy before ART

- Postoperative synechiae with monopolar cautery \(\%35-45\), bipolar cautery \(\%7.5\)

- Second look H/S is suggested after 6-8 weeks after procedure (\(\%10\) synechiae +)

_Touboul C. Fertil Steril 2009_

_Capmas M, Curr Opinion, 2013_
Polypectomy before ART

- Polyp:
  - Abnormal implantation
  - Irregular bleeding
  - Inhibition of sperm transport
  - Inflammatory process
  - Glycodelin secretion

→ Excision (H/S)

- RCT: H/S polypectomy improves pregnancy outcomes when compared with biopsy. (%63 vs %28)

Perez-Medina T et al. Hum Reprod 2005
Hydrosalpinx before ART?

- **Hydrosalpinx**: Decreased implantation rates.

- Decreases IVF success rates up to 50%:
  - Mecanical or toxic effect
  - ‘Wash-out’ effect to the embryo
Hydrosalpinx therapy

1. Reconstructive surgery
2. Medical therapy (doxycycline)
3. Salpingectomy
4. Proximal Tubal Occlusion
   - Clips
   - Electrocautery
   - Hysteroscopy: **Essure**, Adiana, Ovabloc
5. Salpingostomy
6. Aspiration

*Johnson N et al. Cochrane Database Syst Rev 2010*
Hydrosalpinx Therapy

- Laparoscopic salpingectomy is suggested.
- Laparoscopy eases OPU and lessens possible complications like abscess, torsion.

*Johnson N et al. Cochrane Database Syst Rev 2010*
Pregnancy rates after in-vitro fertilization in cases of tubal infertility with and without hydrosalpinx: a meta-analysis of published comparative studies

### Table II. Pregnancy rates

<table>
<thead>
<tr>
<th>First author and year of publication</th>
<th>Hydrosalpinx group No. (%)</th>
<th>Group without hydrosalpinx No. (%)</th>
<th>Odds ratio (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Andersen(^a) (1994)</td>
<td>9/91 (9.8)</td>
<td>224/744 (30.1)</td>
<td>0.25 (0.13–0.52)(^b)</td>
</tr>
<tr>
<td>Strandell (1994)</td>
<td>14/121 (11.57)</td>
<td>89/367 (24.25)</td>
<td>0.41 (0.22–0.75)(^b)</td>
</tr>
<tr>
<td>Sims (1993)</td>
<td>43/234 (18.37)</td>
<td>341/1287 (26.49)</td>
<td>0.62 (0.44–0.89)(^b)</td>
</tr>
<tr>
<td>Blazar(^a) (1995)</td>
<td>39/161 (24.22)</td>
<td>116/385 (30.13)</td>
<td>0.74 (0.44–1.13)</td>
</tr>
<tr>
<td>Van Dromme (1995)</td>
<td>7/69 (10.14)</td>
<td>14/61 (22.95)</td>
<td>0.38 (0.14–1.01)</td>
</tr>
<tr>
<td>Sharara (1996)</td>
<td>27/103 (26.21)</td>
<td>30/89 (33.70)</td>
<td>0.70 (0.38–1.30)</td>
</tr>
<tr>
<td>Akman (1996)</td>
<td>1/14 (7.1)</td>
<td>24/98 (24.5)</td>
<td>0.24 (0.03–1.91)</td>
</tr>
<tr>
<td>Murray (1996)</td>
<td>8/45 (17.77)</td>
<td>57/141 (40.42)</td>
<td>0.32 (0.14–0.73)(^b)</td>
</tr>
<tr>
<td>Katz (1996)</td>
<td>16/95 (16.84)</td>
<td>467/1268 (36.82)</td>
<td>0.35 (0.20–0.60)(^b)</td>
</tr>
<tr>
<td>Fleming (1996)</td>
<td>18/77 (23.37)</td>
<td>63/212 (29.71)</td>
<td>0.72 (0.39–1.32)</td>
</tr>
<tr>
<td>Wainer(^a) (1997)</td>
<td>49/267 (18.35)</td>
<td>199/867 (22.95)</td>
<td>0.75 (0.53–1.07)</td>
</tr>
<tr>
<td>Barma(^a) (1997)</td>
<td>42/106 (39.62)</td>
<td>502/1150 (43.65)</td>
<td>0.85 (0.56–1.27)</td>
</tr>
<tr>
<td>Ng(^a) (1997)</td>
<td>9/41 (21.95)</td>
<td>11/92 (11.96)</td>
<td>2.07 (0.78–5.47)</td>
</tr>
<tr>
<td>De Witt(^a) (1997)</td>
<td>41/224 (18.3)</td>
<td>66/326 (20.25)</td>
<td>0.88 (0.57–1.36)</td>
</tr>
<tr>
<td>Total</td>
<td>323/1642 (19.67(^%))</td>
<td>2203/7061 (31.2%)</td>
<td>0.64 (0.56–0.74)(^b)</td>
</tr>
</tbody>
</table>

\(^a\)Excluding biochemical pregnancies.
\(^b\)Odds ratio significantly different from 1 (\(P < 0.05\)).
\(\chi^2\)-test for heterogeneity (with 13 df) = 29.2 (\(P < 0.05\)).
CI = confidence interval.
OOCYTE FACTOR

Age
Smoking
Obesity
PCOS
Endometriosis
Ovarian surgery
Age

Pregnancy %

Cycle number
Analysis of 2,386 consecutive cycles of in vitro fertilization or intracytoplasmic sperm injection using autologous oocytes in women aged 40 years and above

Gamal Serour, M.D., a, b Ragaa Mansour, Ph.D., b Ahmed Serour, M.D., a, b Mona Aboulghar, M.D., b Yahia Amin, M.D., b Omnia Kamal, B.S., b Hesham Al-Inany, M.D., b and Mohamed Aboulghar, M.D. b

a Al Azhar University, and b Egyptian IVF and ET Center, Cairo, Egypt

<table>
<thead>
<tr>
<th>Outcome</th>
<th>40</th>
<th>41</th>
<th>42</th>
<th>43</th>
<th>44</th>
<th>≥45</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of initiated cycles</td>
<td>742</td>
<td>595</td>
<td>429</td>
<td>251</td>
<td>150</td>
<td>219</td>
<td>2386</td>
</tr>
<tr>
<td>Number of pickup cycles</td>
<td>673</td>
<td>536</td>
<td>379</td>
<td>206</td>
<td>111</td>
<td>99</td>
<td>2004</td>
</tr>
<tr>
<td>Cancellation rate</td>
<td>9.3%</td>
<td>10%</td>
<td>12%</td>
<td>18%</td>
<td>26%</td>
<td>55%</td>
<td>16%</td>
</tr>
<tr>
<td>No. of embryo transfer cycles</td>
<td>601</td>
<td>480</td>
<td>337</td>
<td>178</td>
<td>101</td>
<td>86</td>
<td>1783</td>
</tr>
<tr>
<td>Positive β-hCG</td>
<td>190</td>
<td>125</td>
<td>61</td>
<td>32</td>
<td>12</td>
<td>5</td>
<td>425</td>
</tr>
<tr>
<td>No. of clinical pregnancies</td>
<td>148</td>
<td>92</td>
<td>55</td>
<td>16</td>
<td>4</td>
<td>3</td>
<td>318</td>
</tr>
<tr>
<td>Clinical pregnancy rate per pickup</td>
<td>22.4%</td>
<td>17.2%</td>
<td>14%</td>
<td>7.8%</td>
<td>3.6%</td>
<td>3</td>
<td>17.9</td>
</tr>
<tr>
<td>Miscarriage rate</td>
<td>39%</td>
<td>44.4%</td>
<td>51.3%</td>
<td>64.3%</td>
<td>75%</td>
<td>67%</td>
<td>44.8%</td>
</tr>
<tr>
<td>No. of deliveries</td>
<td>72</td>
<td>40</td>
<td>20</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>139</td>
</tr>
<tr>
<td>Live birth per initiated cycle</td>
<td>10%</td>
<td>7%</td>
<td>5%</td>
<td>2%</td>
<td>0.7%</td>
<td>0.5%</td>
<td>6.7%</td>
</tr>
<tr>
<td>Live birth rate per oocyte pickup</td>
<td>11%</td>
<td>7.5%</td>
<td>5.3%</td>
<td>2.4%</td>
<td>0.9%</td>
<td>0.5%</td>
<td>8%</td>
</tr>
<tr>
<td>Live birth per embryo transfer</td>
<td>12%</td>
<td>8.5%</td>
<td>5.9%</td>
<td>2.8%</td>
<td>1%</td>
<td>1.1%</td>
<td>8.8%</td>
</tr>
</tbody>
</table>

Aneuploidy* (Munne et al, 2003)
Effects of cigarette smoking upon clinical outcomes of assisted reproduction: a meta-analysis

A.L. Waylen¹, ², M. Metwally³, G.L. Jones³, A.J. Wilkinson⁴, and W.L. Ledger⁵

<table>
<thead>
<tr>
<th>Study</th>
<th>Smokers n/N</th>
<th>Non-smokers n/N</th>
<th>OR (random) 95% CI</th>
<th>Weight %</th>
<th>OR (random) 95% CI</th>
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</thead>
<tbody>
<tr>
<td>Elekrogen 1981</td>
<td>1/20</td>
<td>4/21</td>
<td>1.20</td>
<td>100.00</td>
<td>0.56 [0.43, 0.73]</td>
</tr>
<tr>
<td>Ohe 2001</td>
<td>2/40</td>
<td>20/90</td>
<td>2.51</td>
<td>9.40</td>
<td></td>
</tr>
<tr>
<td>Torni 2004</td>
<td>4/17</td>
<td>9/43</td>
<td>2.98</td>
<td>9.02</td>
<td></td>
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<tr>
<td>Tropp 1986</td>
<td>3/68</td>
<td>12/76</td>
<td>3.01</td>
<td>12.46</td>
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<tr>
<td>Agipsi 1984</td>
<td>4/38</td>
<td>20/62</td>
<td>3.37</td>
<td>11.25</td>
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</tr>
<tr>
<td>Ohe 2000</td>
<td>5/38</td>
<td>17/38</td>
<td>3.60</td>
<td>9.19</td>
<td></td>
</tr>
<tr>
<td>Gutterson 1996</td>
<td>5/50</td>
<td>10/50</td>
<td>4.04</td>
<td>10.70</td>
<td></td>
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<tr>
<td>Shaver 1984</td>
<td>8/29</td>
<td>31/73</td>
<td>4.78</td>
<td>9.44</td>
<td></td>
</tr>
<tr>
<td>Van Voorhis 1996</td>
<td>8/39</td>
<td>141/361</td>
<td>5.64</td>
<td>4.10</td>
<td></td>
</tr>
<tr>
<td>Ehlherr 1998</td>
<td>11/65</td>
<td>23/108</td>
<td>5.97</td>
<td>0.75</td>
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</tr>
<tr>
<td>Harrison 1990</td>
<td>8/103</td>
<td>119/542</td>
<td>6.37</td>
<td>0.28</td>
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<td>Stenzl 1998</td>
<td>13/103</td>
<td>18/68</td>
<td>6.47</td>
<td>1.02</td>
<td></td>
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<tr>
<td>Hughes 1994</td>
<td>13/185</td>
<td>29/182</td>
<td>6.74</td>
<td>0.87</td>
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<tr>
<td>Wright 1996</td>
<td>18/36</td>
<td>132/396</td>
<td>6.89</td>
<td>1.92</td>
<td></td>
</tr>
<tr>
<td>Soares 2007</td>
<td>35/44</td>
<td>351/680</td>
<td>7.38</td>
<td>0.48</td>
<td></td>
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<tr>
<td>Patterson 1981</td>
<td>19/124</td>
<td>50/206</td>
<td>8.02</td>
<td>0.67</td>
<td></td>
</tr>
<tr>
<td>Feichtinger 1997</td>
<td>40/142</td>
<td>126/399</td>
<td>9.82</td>
<td>0.85</td>
<td></td>
</tr>
<tr>
<td>Wegele 1995</td>
<td>49/200</td>
<td>194/634</td>
<td>10.50</td>
<td>0.74</td>
<td></td>
</tr>
<tr>
<td><strong>Total (95% CI)</strong></td>
<td><strong>1204</strong></td>
<td><strong>3959</strong></td>
<td><strong>100.00</strong></td>
<td><strong>0.56</strong></td>
<td><strong>[0.43, 0.73]</strong></td>
</tr>
</tbody>
</table>

Figure 2: Odds ratio of clinical pregnancy rate per cycle.
Obesity

- Weight ↑ Fertility ↓
- High gonadotropin and cost
- Diet, exercise, stress management, habituel education
- 5-7 kg weight loss, decreases insulin resistance, induces spontaneous ovulation and pregnancy.

Clark AM, Hum Peprod, 1998
PCOS-COH

COH PROTOCOLS

- Minimal-mild stimulation
- GnRH agonist
- GnRH antagonist

MODIFIED PROTOCOLS

- GnRH agonist trigger
- Metformin
- IVM
- Embryo freezing
‘PCOS’: Metformin

- PCOS; *Insulin resistance*
- RCT
  - Metformin started 16 weeks before cycle
  - Dose and duration heterogenous (2x500mg, 3x850mg)

- *Metformin > placebo*
- *Increased live birth rates*

*Tso LO, Cochrane Database Syst Rev 2009*
Metformin and OHSS

- Metformin $\downarrow$ hCG day testosteron, free androgen
- hCG day E2 and VEGF OHSS $\downarrow$
- Long GnRH agonist protocol OHSS $\downarrow$

*TSO LO, Cochrane Database Syst Rev 2009*
OHSS Prediction

- AMH (sens. %90.5, spes. %81.3)
- AMH cut-off 3.36 ng/mL

Lee et al, 2008

Figure 2: Selection of cut-off value for basal serum AMH to predict OHSS by ROC curve analysis. The selected value was 3.36 ng/mL, with a sensitivity of 90.5% (95% CI 69.6–98.5) and a specificity of 81.3% (95% CI 75.8–86.0).
OHSS Prevention

- PCOS and OHSS history
- Mild stimulation
- Antagonist protocols
- Coasting
- Analog trigger
- IV albumin-OPU
- Cryopreservation
- In vitro maturation
- Low dose hCG
- Metformin and dopamine

Aboulghar, Rep Biomed Online, 2009
OHSS Prevention

Follicular phase
- AMH
- Antral follicle count
- Age
- History

Day of ovulation triggering
- More than 18 follicles +/- E2 > 5000 pg/ml
- 5000-3500 IU uHCG or 250 mcg rec-HCG
- GnRH Agonist Triggering (0.2 mg Triptorelin, 0.6 mg Buscopan, 1 mg Leuprolide)

Luteal phase
- Signs of Early OHSS
  - Freeze all embryos
  - 2PN or Day 2/3
- Proceed to Day 5, evaluate patient
- Supplement luteal phase
  - 1500 IU uHCG on OPU
  - Oral or Luteal LH 300 IU/2nd day
  - Im. Progesterone + E2
- Freeze all embryos
  - 2PN or Day 2/3
- Freeze half embryos on day 2/3 and culture the rest to Day 5

Papanikolaou EG, Reproductive Biology & Endocrinology, 2011
Endometriosis and ART

Ultra-long protocol

- GnRH agonist 3-6 months or OCs 6-8 weeks before ART

Surrey ES et al. Fertil Steril 2002
De Ziegler et al. Fertil Steril 2010

Ongoing pregnancy rates and MII oocyte and number of embryos
Endometriosis: surgery before ART?

There is no consensus!
- Unilateral vs bilateral
- Size, age, previous surgery
- Surgical technique
- >4 cm endometrioma → SURGERY

Similar success rates after surgical management

Garcia-Velasco et al. Fertil Steril 2004

Recurrent IVF failure → Pregnancy after surgery %72

Littman E et al. Fertil Steril 2005
ADJUVANT THERAPIES

DHEA ?
Luteal Phase Support ?
GH ?
DHEA increases FSH activity at granulosa cells during preantral and antral period by binding androgen receptors.

Gleicher, Reproductive Biology and Endocrinology, 2011
**DHEA**

- Meta-analysis/ 3 RCT
- 4-10 weeks 75mg/day DHEA

DHEA and control group;

Spontaneous Microdose flare up FSH 300–450 + hMG 150

Long GnRH protocol rFSH450+rLH150

Microdose agonist flare FSH 300-450 + hMG 150

Narkwichean A, Rep Bio & Endocrinol, 2013
200 cycle; DHEA no significant advantage!

Narkwichean A, Rep Bio & Endocrinol, 2013
Luteal Phase Support

- **Synthetic progesterone** is (oral dydrogesterone) more beneficial than micronised progesterone.

- **Estrogene and hCG** do not effect outcomes.

- Route of progesterone administration **does not** change the results.

*Van der Linden M, Cochrane, 2012*
GnRHa for luteal phase support, positively effects the outcomes.

Van der Linden M, Cochrane, 2012
Growth hormone

- Insulin like growth factor-1
- FSH effect
- Oocyte maturation

- Embryo quality and implantation rates

Bachelot A, Endocrinology, 2002

Mendoza C, Human Reprod, 2002
Mendoza C, Human Reprod, 1999
GH

- Meta-analysis;

For Poor responders;

GH addition increases clinical pregnancy and live birth rates!

Kolibianakis, Hum Rep, 2009
Growth Hormone (GH) ?

- *Growth hormone*;
- GH- Poor responder ; positive effects

*Kyrou D, Fertil Steril 2009*

- GH- Normo responder ; not effective!

*Duffy JMN et al. Cochrane Rev*
Not effective within antagonist protocol!

Eftekhar M, Arch Gynecol Obstet, 2013
CYCLE SUCCESS

Progesterone
Estradiol
Progesterone levels

- Meta-analysis/ 6 RCT
- **Late follicular phase** P levels are low?
- Age of <39, IVF cycle (n=1866)
- **rFSH +GnRH ant** protocol
- P : **1.5ng/ml**

### Progesterone levels

<table>
<thead>
<tr>
<th>On day hCG P &gt;1.5ng/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>157/1866 (%8.4)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>POOR RESPONSE (1-5 oosit)</th>
<th>%4.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>HİGH RESPONSE (&gt;18 oosit)</td>
<td>%19</td>
</tr>
</tbody>
</table>

Progesterone levels

Frequency distribution of serum P levels on the day of hCG administration for women with low ovarian response (<6 oocytes), normal ovarian response (6–18 oocytes), and high ovarian response (>18 oocytes).

Ongoing pregnancy rate per embryo transfer and associated 95% confidence interval by number of oocytes retrieved and serum P level on the day of hCG.

POOR and NORMO responder patients
High P levels are associated with low pregnancy rates

HIGH responder patients High P levels are not associated with pregnancy rates.

Estradiol after hCG

n= 1712 IVF cycle

- Retrospective /age ;21-45 /
- GnRH agonist protocol
- GnRH antagonist protocol or ‘microdose flare’

- Estradiol levels ;
  - On hCG day (early in the morning)
  - Post-hCG (after 10-12 h)

Kondapalli , Hum Rep, 2012
Estradiol after hCG

E2 levels;

- **Group A**: 1065 >10%
- **Group B**: 525 plato
- **Group C**: 122 >10%

Kondapalli, Hum Rep, 2012
# Estradiol after hCG

## Table 1: Patient characteristics in three groups stratified by serum estradiol response to hCG administration.

|                         | Group A (>10% rise), n = 1065 | Group B (±10% plateau), n = 525 | Group C (>10% fall), n = 122 | P-value  \\
|-------------------------|-------------------------------|---------------------------------|-------------------------------|---------
| Diminished ovarian reserve | 16.9                         | 23.4                            | 25.4                          | 0.002<sup>a</sup>,<sup>b</sup> |
| Polycystic ovarian syndrome | 13.9                         | 5.3                             | 7.4                           | <0.001<sup>a</sup>,<sup>b</sup> |

<table>
<thead>
<tr>
<th>Ovarian stimulation protocol (%)</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>GnRH agonist</td>
<td>81.0</td>
<td>71.2</td>
<td>75.4</td>
</tr>
<tr>
<td>GnRH antagonist</td>
<td>3.8</td>
<td>5.1</td>
<td>5.7</td>
</tr>
<tr>
<td>Microdose GnRHα Flare</td>
<td>15.2</td>
<td>23.6</td>
<td>18.8</td>
</tr>
</tbody>
</table>
Estradiol after hCG

- >%10 decrease
  - Clinical pregnancy & live birth %40-50

- Plato (± %10)
  - Clinical pregnancy & live birth >25%.

Kondapalli, Hum Rep, 2012
POOR RESPONSE

Protocol shift
Mild stimulation
Antagonist protocol
Microdose protocol
Oocyte pooling?
IVF? ICSI?
Poor Responder

- Suggested gonadotropine dose 300 IU/day
- >300 IU doses are not effective

Why?
FSH rec. gen polymorphism?
Ser/Ser allele variants are gonadotropin insensitive

Centre for Clinical Effectiveness
Cochrane Database 2000

Cai J, Fertil Steril 2007
rFSH or hMG have same outcomes for IVF/ICSI cycles.

Al-Inany H, Gynecol Endocrinol 2005

There is insufficient data for addition of LH to ART outcomes.

Poor Responder

- PRINT trial (n=111) poor responder (RCT)
  - (1) the GnRH agonist long protocol
  - (2) the GnRH agonist short protocol
  - (3) the GnRH antagonist protocol

Sunkara SK Fertil Steril 2014
GnRH agonist long protocol

- **Day 1 of cycle**
  - Period
  - Day 1 of stimulation
  - Start FSH injections
- **Day 21**
  - Period
  - Day 1 of stimulation
- **Day 9/10 of stimulation**
  - Scan to assess follicular growth
- **HCG administered 36 hours prior to egg retrieval**
- **EMBRYO TRANSFER**
- **EGG RETRIEVAL** (average day 13-15)
- **Pregnancy test (2 weeks after egg collection)**
  - Progesterone for luteal support
GnRH agonist short protocol

- **Day 1 of cycle**
  - Start FSH injections on day 3/4 of cycle (Day 1 of stimulation)

- **Period**
  - Start GnRH agonist (sniffing or injections on day 2/3 of cycle)

- **Day 9/10 of stimulation**
  - Scan to assess follicular growth

- **Stop GnRH agonist** (sniffing or injections)
  - HCG administered 36 hours prior to egg retrieval

- **EGG RETRIEVAL** (average day 13-15)
  - Pregnancy test (2 weeks after egg collection)
    - Progesterone for luteal support

- **EMBRYO TRANSFER** (3 or 5 days following egg retrieval)
GnRH antagonist protocol

1. **Start FSH injections on day 2/3 of cycle (Day 1 of stimulation)**
   - Day 6 scan and further scans to identify lead follicle of 14mm

2. **Day 9/10 of stimulation**
   - Scan to assess follicular growth

3. **HCG administered 36 hours prior to egg retrieval**

4. **EGG RETRIEVAL**
   - Average day 13-15

5. **EMBRYO TRANSFER**
   - 3 or 5 days following egg retrieval

6. **Pregnancy test**
   - 2 weeks after egg collection

7. **Progesterone for luteal support**

8. **GnRH antagonist injections**
   - Started when lead follicle identified and continued to day 9/10

9. **Period**

   - Day 1 of cycle

   - Sunkara SK Fertil Steril 2014
Poor Responder

- **Oocyte number**
  Long agonist = Antagonist > Short agonist

- **Gonadotropine dose**
  Long agonist > Short agonist & Antagonist

- **Ongoing pregnancy**
  Short and Long agonist - %8.1 Antagonist - %16.2

Long agonist and antagonist protocols are more efficient

Sunkara SK Fertil Steril 2014
<table>
<thead>
<tr>
<th></th>
<th>P value</th>
<th>GnRH agonist/ antagonist</th>
<th>Microdose flare-up</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of used gonadotropin ampoules</td>
<td>0.591</td>
<td>44.12 ± 8.20</td>
<td>45.20 ± 6.93</td>
</tr>
<tr>
<td>Duration of stimulation (Days)</td>
<td>0.610</td>
<td>11.60 ± 1.32</td>
<td>11.42 ± 1.61</td>
</tr>
<tr>
<td>No. of retrieved oocytes</td>
<td>0.802</td>
<td>4.61 ± 3.53</td>
<td>4.42 ± 3.63</td>
</tr>
<tr>
<td>No. of transferred embryos</td>
<td>0.954</td>
<td>2.44 ± 2.10</td>
<td>2.31 ± 2.41</td>
</tr>
<tr>
<td>Fertilization rate (%) (Per cycle)</td>
<td>0.458</td>
<td>62 ± 27</td>
<td>58 ± 30</td>
</tr>
<tr>
<td>Clinical pregnancy rate (%) (Per cycle)</td>
<td>0.389</td>
<td>13.3%</td>
<td>10%</td>
</tr>
</tbody>
</table>

**Results:** There were no significant differences between the groups in the number of used gonadotropin ampoules (p=0.591), duration of stimulation (p=0.610), number of retrieved oocytes (p=0.802), fertilization rate (p=0.456), and the number of transferred embryos (p=0.954). The clinical pregnancy rates were statistically similar in group I (10%) compared with group II (13.3%, p=0.389).
Minimal Stimulation/poor responder

Minimal stimulation protocol at The Muasher Center for Fertility and IVF.

- hCG 10,000 IU
- GnRH-Antag. 0.25 mg/day
- Clomiphene Citrate 100 mg/day
- Gonadotropins 150 IU/day

Cycle Day: 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18

Retrieval  Transfer

Minimal Stimulation/poor responder

Minimal stimulation versus full stimulation in low responders at the Muasher Center for Fertility and IVF, 2009–2010.

<table>
<thead>
<tr>
<th>Stimulation protocol</th>
<th>Minimal</th>
<th>Full</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>13</td>
<td>42</td>
<td>NS</td>
</tr>
<tr>
<td>Age (y)</td>
<td>38.7 ± 3.7</td>
<td>38.9 ± 2.9</td>
<td>NS</td>
</tr>
<tr>
<td>Day-3 FSH (mIU/mL)</td>
<td>12.1 ± 2.7</td>
<td>10.1 ± 3.7</td>
<td>NS</td>
</tr>
<tr>
<td>E₂ at hCG (pg/mL)</td>
<td>808 ± 353</td>
<td>1,082 ± 561</td>
<td>&lt;.05</td>
</tr>
<tr>
<td>Vials of gonadotropins</td>
<td>9.7 ± 3.3</td>
<td>49.8 ± 7.4</td>
<td>&lt;.01</td>
</tr>
<tr>
<td>Days of monitoring</td>
<td>3</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Mature oocytes</td>
<td>2.4 ± 1.6</td>
<td>3.8 ± 2.3</td>
<td>&lt;.05</td>
</tr>
<tr>
<td>Embryos transferred</td>
<td>2.0 ± 1.1</td>
<td>2.1 ± 1.2</td>
<td>NS</td>
</tr>
<tr>
<td>Clinical pregnancy/ cycle</td>
<td>38% (5/13)</td>
<td>36% (15/42)</td>
<td>NS</td>
</tr>
<tr>
<td>Clinical pregnancy/ transfer</td>
<td>42% (5/12)</td>
<td>47% (15/32)</td>
<td>NS</td>
</tr>
</tbody>
</table>

Note: E₂ = estradiol; FSH = follicle-stimulating hormone; hCG = human chorionic gonadotropin.

* Four patients canceled before retrieval. Six patients had retrieval without transfer.

Zarek, Mild/mixed stimulation for IVF. Ferti Steril 2011
‘Oocyte Pooling’

- N=724 poor responder
- <5 oocytes
- D3 FSH >11 IU/ml
- AFC <6
- AMH < 0.7ng/ml

**Group I:** ‘low response, accumulation of oocytes and vitrification’ n=242  
**Group II:** low response, fresh oocytes n=482

‘Oocyte Pooling’

- OCs 15-21 day
- Flexible antagonist protocol

- Fertilisation rates same
- Cycle cancellation 4x in Low Response-fresh group

Cumulative pregnancy rates are higher with oocyte pooling

IVF or ICSI?

• **ICSI indications:**
  Male factor
  Conventional IVF failure,
  Surgically achieved spermatozoa
  Before PGD

*Borini A, Reprod Biomed Online, 2009; Griffiths TA, Hum reprod, 2000*
Intracytoplasmic sperm injection (ICSI) for non-male factor infertility: a committee opinion

The Practice Committees of the American Society for Reproductive Medicine and Society for Assisted Reproductive Technology
American Society for Reproductive Medicine, Birmingham, Alabama

CONCLUSIONS

- There are no data to support the routine use of ICSI for non-male factor infertility.
- ICSI may be beneficial for patients using PGT, IVM, or cryopreserved oocytes.
- The safety and cost of ICSI in the setting of non-male factor infertility must be considered.
EMBRYO QUALITY

PGD

CGH

Time-lapse embryoscopy

Metabolomics
<table>
<thead>
<tr>
<th>Day 0</th>
<th>Day 1</th>
<th>Day 2</th>
<th>Day 3</th>
<th>Day 4</th>
<th>Day 5-6</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Embryo Development" /></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Morphology Assessment:
- PS, ZP, PB cytoplasm and spindle grading
- PN grading

### Cleavage Rate, Assessment of Fragmentation

### ICM Size

### PGD / PGS:
- Polar body biopsy
- Blastomere biopsy
- TE biopsy

### Transcriptomics:
- mRNAs in follicular cells

### Analysis of Metabolism:
- Puryvate and glucose uptake
- Oxygen consumption
- Amino acid turnover
- HLA-G levels
- Leptin levels
- Metabolome
- Secromome and metabolome

### Time-lapse Imaging:
- Cytoplasmic flows
- Rate and synchrony of the first cleavage divisions
Embryo Quality

Uptake
- Glucose
- Pyruvate
- Amino Acids
- Oxygen

Production
- Lactate
- Ammonium
- Amino Acids
- sHLA-G
- HOXA 10 Regulator
- PAF
Embryo Selection

- Implantation failure → aneuploidy

- ‘Comprehensive chromosome screening’ (CCS) techniques
  - single nucleotide polymorphism (SNP) array
  - quick polymerase chain reaction analysis,
  - comparative genomic hybridization (CGH) array
## Genetic Tests - Embryo

### Table 1: Overview of most common techniques used in genetic testing of embryos.

<table>
<thead>
<tr>
<th></th>
<th>Amplification-based PCR (multiplex PCR and PGH)</th>
<th>Fluorescent in situ hybridization (FISH)</th>
<th>Array CGH</th>
<th>SNP array</th>
<th>Quantitative SNP array analysis and karyomapping</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single-gene defects</td>
<td>x</td>
<td></td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>HLA typing</td>
<td>x</td>
<td></td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Chromosome screening</td>
<td>x</td>
<td>x (5 – 12 chr)</td>
<td>x (24 chr)</td>
<td>x (24 chr)</td>
<td>x (24 chr)</td>
</tr>
<tr>
<td>Aneuploidy</td>
<td>x</td>
<td>x (5 – 12 chr)</td>
<td>x (24 chr)</td>
<td>x (24 chr)</td>
<td>x (24 chr)</td>
</tr>
<tr>
<td>Duplication/deletions</td>
<td>x</td>
<td></td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Reciprocal/Robertsonian imbalance</td>
<td>x</td>
<td></td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
</tbody>
</table>

*Hens K, Hum Rerod Update, 2013*
Embryo Selection

- Blastocyst transfer:
  - First clivage time,
  - Time between 2. stage to 3. stage ,
  - Time between ICSI to 5 cells

Meseguer MH, Human Reprod 2011;
Hashimoto S, Fertil Steril 2012;
Wong CC, Nat Biotechnol 2010;
Embryo Selection
PGD vs PGS

- Preimplantation Genetic Diagnosis (PGD)
- Preimplantation Genetic Screening (PGS)

- PGD;
  For patients who may have a genetic defect
- PGS;
  For evaluation embryos in terms of chromosomes

Harper, Segupta, 2012
Kahraman, 2011
Embryo Selection
PGD

- Implantation rate 5X,
- Pregnancy rate 2x

- Translocation (+);
  Abortion rate 6x
  Pregnancy rate 7x

Verlinsky Y, Preimplantation Genetic Diagnosis, 2006
Gianaroli L, Reprod BioMed Online, 2004
Embryo Selection
CGH

- CGH array
  - polar body
  - Blastocyst trophoectoderm cells

- Less mozaisism
- Less harm to the embryo

Santos MA, Hum Reprod 2010
Forman EJ, Semin Reprod Med 2012
Embryo Selection
CGH

- CGH increases pregnancy rates.

- Prospective RCT – Ongoing pregnancy rate (%69 vs %42) (P=0.009)

Forman EJ, Hum Reprod 2012
Yang Z, Mol Cytogenet 2012

Yang Z, Mol Cytogenet 2012
Genetic Tests- ‘Embryoscope’

- **Morphological properties**
  - Non-invasive
  - Analysis of ‘snapshot’ images

- *Time-lapse* images of embryonic development.

- Higher implantation rates with embryos selected by embryoscope
Metabolomics

- Metabolomic characteristics are different in implanted and non-implanted groups.


- Embryo viability indexes, Raman and NIR are high due to spectroscopic analysis.

Noninvasive metabolomic profiling as an adjunct to morphology for noninvasive embryo assessment in women undergoing single embryo transfer

Emre Seli, M.D., a Carlijn G. Vergouw, M.Sc., b Hiroshi Morita, B.Agr., c Lucy Botros, M.Sc., d Pieter Roos, Ph.D., d Cornelius B. Lambalk, M.D., Ph.D., b Naoki Yamashita, M.D., c Osamu Kato, M.D., c and Denny Sakkas, Ph.D. a,d

Yellow: viability index < 0.3
Red: Viability index > 0.3
Blue: implantation rates for different morphological grades

Implantation rates of day 3 embryos comparing morphological grades and a viability index of less than or greater than 0.3.
Transcriptomics

Transcriptomic analysis → Genetic expression analysis

- Cellular development
- Biological problems
- Embryonal viability
- Harmful peripheral factors
- Morphological anomalies
Genetic Tests

- For embryonal viability → Cumulus cell gen express.
- Time-lapse molecular analysis.
- Sperm count < 5 million → CGH %72 anomaly
- Sperm count > 5 million → CGH %53 anomaly
Genetic Tests

- PIF \[\rightarrow\] Preimplantation genetic factor
- From endometrial stromal cells
- At first trimester from extravillous cytotrophoblasts
- To determine placental complications
- To define embryo development
Genetic Tests

- CCS  ➔ Comprehensive Chromosomal Screening
- qpcR based CCS analysis
- %97.6 - %98.6 positive
- qpcR is more accurate than a CGH and can define genomic imprinting defects
How to diagnose sperm dysfunction?
- Calcium activation
- Catsper activation
- Proteomics (Sperm fertility array)
- Swim-up gradient
- IMSI, ICSI, PICS
- Birefrigence
- PLA2, ABCA, CD84, CMKLR1
- MAC’S (Magnetic cell sorting)
Genetic Tests

- ERA (Endometrial Receptivity Array)

- This molecular test allows to diagnose endometrium is receptive or not by analysing the expression 238 genes related to endometrial receptivity.

- Endometrial biopsy must be performed at P+5 (hormone replacement therapy cycle) or at LH+7 (natural cycle)
Genetic Tests

- **ERA (Endometrial Receptivity Array)**
- Biopsy of the uterine fundus with a Pipelle catheter or similar. About 30 mg or 3 mm tissue is enough.
- If the result of the first ERA test is non-receptive, this means that the window of implantation may be displaced; it is necessary to validate this displacement with a second ERA test.
- This second analysis will lead to the day in which the endometrium shows a receptive status, and therefore the thawing of eggs or embryos and their transfer must be scheduled to coincide with the day in which the receptive result has been obtained.
- In less than 1% of patients, the first ERA test is non-receptive without a recommendation for a new implantation window, which does not suggest a therapeutic solution.
EMBRYO TRANSFER

Time?

Technique?
Live birth rates after transfer of equal number of blastocysts or cleavage-stage embryos in IVF. A systematic review and meta-analysis

Evangelos G. Papanikolaou¹, Efstratios M. Kolibianakis, Herman Tournaye, Christos A. Venetis, Human Fatemi, Basil Tarlatzis and Paul Devroey

<table>
<thead>
<tr>
<th>Study or sub-category</th>
<th>Blastocyst n/N</th>
<th>Cleavage n/N</th>
<th>OR (fixed) 95% CI</th>
<th>Weight %</th>
<th>OR (fixed) 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>01 Truly randomized</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coskun et al. (2000)</td>
<td>35/100</td>
<td>33/101</td>
<td></td>
<td>11.43</td>
<td>1.11 [0.62, 1.99]</td>
</tr>
<tr>
<td>Rienzi et al. (2002)</td>
<td>29/50</td>
<td>27/48</td>
<td></td>
<td>6.20</td>
<td>1.07 [0.48, 2.39]</td>
</tr>
<tr>
<td>Van der Auwera et al. (2002)</td>
<td>29/70</td>
<td>20/66</td>
<td></td>
<td>6.46</td>
<td>1.63 [0.80, 3.30]</td>
</tr>
<tr>
<td>Bungum et al. (2003)</td>
<td>32/61</td>
<td>36/57</td>
<td></td>
<td>9.47</td>
<td>0.64 [0.31, 1.34]</td>
</tr>
<tr>
<td>Heiress and et al. (2004)</td>
<td>22/64</td>
<td>25/90</td>
<td></td>
<td>7.81</td>
<td>1.15 [0.57, 2.32]</td>
</tr>
<tr>
<td>Kolibianakis et al. (2004)</td>
<td>76/226</td>
<td>75/234</td>
<td></td>
<td>26.19</td>
<td>1.07 [0.73, 1.59]</td>
</tr>
<tr>
<td>Papanikolaou et al. (2006)</td>
<td>42/80</td>
<td>27/84</td>
<td></td>
<td>6.70</td>
<td>1.63 [1.02, 2.61]</td>
</tr>
<tr>
<td>Papanikolaou et al. (2006)</td>
<td>58/175</td>
<td>41/176</td>
<td></td>
<td>14.63</td>
<td>1.63 [1.02, 2.61]</td>
</tr>
<tr>
<td>Main analysis (95% CI)</td>
<td>323/826</td>
<td>284/846</td>
<td></td>
<td>88.89</td>
<td>1.27 [1.03, 1.55]</td>
</tr>
<tr>
<td>Test for heterogeneity: $\chi^2 = 9.53$, df = 7 ($P = 0.22$), $I^2 = 26.5%$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Test for overall effect: $Z = 2.28$ ($P = 0.02$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| 02 Pseudo-randomized  |                |              |                   |          |                   |
| Sensitivity analysis (95% CI) | 375/954   | 315/945     |                   | 100.00   | 1.29 [1.07, 1.56]  |
| Test for heterogeneity: $\chi^2 = 9.85$, df = 8 ($P = 0.28$), $I^2 = 18.8\%$ |
| Test for overall effect: $Z = 2.64$ ($P = 0.008$) |

![Figure 4](image) Clinical pregnancy rate per randomized couple

Time for ET?

- **Blastocyst Transfer**
  - Uterine and embryonic syncronisation
  - Uterine micro-environment hyperstimulation
  - Uterine contraction
  - Embryo selection
  - Implantation rates

Gardner DK, Fertil Steril, 1996;
Barnes FL, Theriogenology, 2000;
Fanchin R, Hum Reprod, Hum Reprod, 2001
Embryo Transfer: Technique

- ET: 85% implantation failure
- 30% transfer technique

* Edwards RG. Hum Reprod 1995; 10: 60-6
** Li et al. J Assist Reprod Genet 2005; 22: 3-8
The relative importance of factors important for successful embryo transfer.\textsuperscript{a}

<table>
<thead>
<tr>
<th>Priority</th>
<th>Mean score\textsuperscript{b}</th>
</tr>
</thead>
<tbody>
<tr>
<td>Removal of hydrosalpinges</td>
<td>6.8</td>
</tr>
<tr>
<td>Absence of blood or mucus</td>
<td>6.6</td>
</tr>
<tr>
<td>Type of catheter</td>
<td>6.1</td>
</tr>
<tr>
<td>Not touching fundus</td>
<td>5.8</td>
</tr>
<tr>
<td>Avoiding tenaculum</td>
<td>5.7</td>
</tr>
<tr>
<td>Removal of all mucus</td>
<td>5.2</td>
</tr>
<tr>
<td>Ultrasonography of cavity before procedure</td>
<td>4.3</td>
</tr>
<tr>
<td>Leaving catheter in place for 1 minute</td>
<td>4.2</td>
</tr>
<tr>
<td>30 minutes of bed rest</td>
<td>3.8</td>
</tr>
<tr>
<td>Trial transfer</td>
<td>3.1</td>
</tr>
<tr>
<td>Ultrasonographic monitoring</td>
<td>2.6</td>
</tr>
<tr>
<td>Antiprostaglandins to prevent uterine contractions</td>
<td>1.9</td>
</tr>
</tbody>
</table>

\textsuperscript{a} Data from reference 11.

\textsuperscript{b} The possible score for each factor was on a scale of 1 to 10.

Optimizing the technique of embryo transfer

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Division of Reproductive Endocrinology and Infertility, University of Iowa Hospitals and Clinics, Iowa City, Iowa

SUMMARY
Evidence-based Guidelines

1. Effort should be made to avoid “difficult” transfers.
2. Ultrasound guidance will result in easier transfers with improved outcomes.
3. Soft catheters should be used when feasible.

Recommendations Based on Expert Opinion

1. Trial transfers allow better preparation for difficult transfers.
2. Cervical mucus should be removed to potentially decrease bacterial contamination and mucus plugging of the catheter.
3. Embryos should be deposited in the midportion of the uterus.
4. Negative pressure should be minimized during withdrawal of the catheter.
5. The procedure should be done in a minimum amount of time.
### Transfer Method

#### Table 1

Characteristics of patients, complications, and clinical outcome after ET.

<table>
<thead>
<tr>
<th></th>
<th>Full bladder (n = 67)</th>
<th>Empty bladder (n = 64)</th>
<th>Clinical touch (n = 40)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age</td>
<td>34.9 ± 4.6</td>
<td>35.7 ± 4.9</td>
<td>35.9 ± 5.9</td>
<td>NS</td>
</tr>
<tr>
<td>BMI</td>
<td>24.5 ± 4.9</td>
<td>23.2 ± 3.8</td>
<td>23.7 ± 4.2</td>
<td>NS</td>
</tr>
<tr>
<td>Baseline FSH</td>
<td>6.2 ± 2.3</td>
<td>5.7 ± 2.2</td>
<td>6.1 ± 3.7</td>
<td>NS</td>
</tr>
<tr>
<td>No. oocytes retrieved/patient</td>
<td>10.7 ± 4.2</td>
<td>12.7 ± 5.7</td>
<td>10.9 ± 6.5</td>
<td>NS</td>
</tr>
<tr>
<td>No. embryos transferred</td>
<td>2.6 ± 0.8</td>
<td>2.4 ± 0.7</td>
<td>2.4 ± 1</td>
<td>NS</td>
</tr>
<tr>
<td>Use of obturator (%)</td>
<td>13.4</td>
<td>32.8</td>
<td>32.5</td>
<td>&lt; .02</td>
</tr>
<tr>
<td>Use of tenaculum (%)</td>
<td>8.9</td>
<td>26.5</td>
<td>25</td>
<td>&lt; .002</td>
</tr>
<tr>
<td>Use of hysteroscope (%)</td>
<td>1.5</td>
<td>14</td>
<td>15</td>
<td>&lt; .002</td>
</tr>
<tr>
<td>Blood in the catheter (%)</td>
<td>5.5</td>
<td>7.8</td>
<td>14.3</td>
<td>NS</td>
</tr>
<tr>
<td>Retained embryos (%)</td>
<td>0</td>
<td>3.1</td>
<td>5</td>
<td>NS</td>
</tr>
<tr>
<td>Implantation rate (%)</td>
<td>16.1</td>
<td>15.4</td>
<td>14.3</td>
<td>NS</td>
</tr>
<tr>
<td>Clinical pregnancy rate (%)</td>
<td>39</td>
<td>38.7</td>
<td>35.7</td>
<td>NS</td>
</tr>
<tr>
<td>Ectopic pregnancies (%)</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Abortion rate (%)</td>
<td>14.3</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note: NS = not significant.

**Ultrasound guided embryo transfer**

- Clinical pregnancy rates increases (OR: 1.31, %95 CI 1.18-1.46)
- Ongoing pregnancy rates increases (OR: 1.38, %95 CI 1.16-1.64)
- No difference at live birth rates (OR: 1.14, %95CI 0.93-1.39)

*Brown J, Cochrane Database Syst Rev, 2010*

---

**Table IV. Meta-analysis of pregnancy rates in randomized trials. Ultrasound (US)-guided transfer versus clinical touch transfer**

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>US-guided transfer (%)</th>
<th>Clinical touch transfer (%)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Randomized trials</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coroleu et al.</td>
<td>2000</td>
<td>50 (91/182)</td>
<td>33.7 (61/180)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Tang et al.</td>
<td>2001</td>
<td>26.0 (115/441)</td>
<td>22.5 (81/359)</td>
<td>NS</td>
</tr>
<tr>
<td>Matorras et al. (this study)</td>
<td>2002</td>
<td>26.3 (67/255)</td>
<td>18.1 (47/260)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Quasi-randomized trials</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hurley et al.</td>
<td>1991</td>
<td>20.2 (19/94)</td>
<td>17.5 (43/246)</td>
<td>NS</td>
</tr>
<tr>
<td>Al-Shawaf et al.</td>
<td>1993</td>
<td>28.9 (44/152)</td>
<td>30.3 (27/89)</td>
<td>NS</td>
</tr>
<tr>
<td>Prapas et al.</td>
<td>1995</td>
<td>36.1 (22/61)</td>
<td>22.5 (16/71)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Kan et al.</td>
<td>1999</td>
<td>37.8 (37/98)</td>
<td>29.8 (28/97)</td>
<td>NS</td>
</tr>
<tr>
<td>Prapas et al.</td>
<td>2001</td>
<td>47.6 (206/433)</td>
<td>36.0 (229/636)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Global meta-analysis</td>
<td>2002</td>
<td>35.0 (601/1716)</td>
<td>27.5 (532/1938)</td>
<td>&lt; 0.0001^a</td>
</tr>
<tr>
<td>Meta-analysis including</td>
<td>2002</td>
<td>31.4 (273/870)</td>
<td>23.7 (189/799)</td>
<td>&lt; 0.001^b</td>
</tr>
</tbody>
</table>

^aχ² = 20; odds ratio (OR) = 1.4; 95% confidence interval (CI) = 1.23–1.64.

^bχ² = 12; OR = 1.5; 95% CI = 1.18–1.85.

NS = not significant.
The longer the ILDE 'interval loading-discharging embryos', the lower the pregnancy and implantation rates
Endometrial pattern

- Triple-line pattern
- No-triple line pattern
- Iso-echoic endometrium
- No-triple line pattern
- Homogeneous hyperechogenic pattern

Chen et al. Reproductive Biology and Endocrinology 2010, 8:30
Endometrial pattern

Table 1  Three-Dimensional sonographic and Power Doppler criteria for predicting endometrial receptivity

<table>
<thead>
<tr>
<th>3D Criteria</th>
<th>Findings</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Endometrial echogenicity</td>
<td>Hyper-echogenic</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Echogenic</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Triple line</td>
<td>2</td>
</tr>
<tr>
<td>Endometrial thickness</td>
<td>≤7 mm</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>&gt;7 mm</td>
<td>2</td>
</tr>
<tr>
<td>Endometrial volume</td>
<td>≤2.31 mm</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>&gt;2.31 mm</td>
<td>2</td>
</tr>
<tr>
<td>Sub-endometrial halo</td>
<td>Regular</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Disturbed</td>
<td>2</td>
</tr>
<tr>
<td>Endometrial flow</td>
<td>Yes</td>
<td>0</td>
</tr>
<tr>
<td>Sub-endometrial flow</td>
<td>No</td>
<td>2</td>
</tr>
<tr>
<td>RI</td>
<td>≥0.53</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>&lt;0.53</td>
<td>2</td>
</tr>
<tr>
<td>Vessel’s architecture</td>
<td>Simple</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Complex</td>
<td>2</td>
</tr>
</tbody>
</table>
Endometrial Scratch

- 6 RCT
  Clinical pregnancy
  (RR: 1.86, 95% CI 1.46–2.38]

- RIF (+)
  (2 RCT and 2 nonrandomised study)
  Clinical pregnancy
  (RR 2.32, 95% CI 1.72–3.13).

Nastri CO, Gynecol Endocrinol 2013
Increased live-birth and ongoing pregnancy rate

**Endometrial Scratch**

<table>
<thead>
<tr>
<th>Study</th>
<th>Treatment</th>
<th>Control</th>
<th>RR (fixed) 95% CI</th>
<th>Weight %</th>
<th>RR (fixed) 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Randomised controlled trial</td>
<td>Narvekar et al 2010</td>
<td>11/49</td>
<td>5/51</td>
<td>100.00</td>
<td>2.29 [0.86, 6.11]</td>
</tr>
<tr>
<td>Total (95% CI)</td>
<td>49</td>
<td>51</td>
<td></td>
<td>100.00</td>
<td>2.29 [0.86, 6.11]</td>
</tr>
<tr>
<td>Total events: 11 (Treatment), 5 (Control)</td>
<td>Test for heterogeneity: not applicable</td>
<td>Test for overall effect: Z = 1.65 (P = 0.10)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-randomised controlled studies</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Barash et al 2003</td>
<td>22/45</td>
<td>21/89</td>
<td></td>
<td>38.05</td>
<td>2.07 [1.28, 3.34]</td>
</tr>
<tr>
<td>Li et al 2004</td>
<td>17/35</td>
<td>4/36</td>
<td></td>
<td>10.64</td>
<td>4.37 [1.63, 11.70]</td>
</tr>
<tr>
<td>Raziel et al 2007</td>
<td>13/60</td>
<td>5/57</td>
<td></td>
<td>13.84</td>
<td>2.47 [0.94, 6.49]</td>
</tr>
<tr>
<td>Zhou et al 2008</td>
<td>25/60</td>
<td>14/51</td>
<td></td>
<td>37.45</td>
<td>1.82 [1.05, 3.14]</td>
</tr>
<tr>
<td>Total (95% CI)</td>
<td>200</td>
<td>243</td>
<td></td>
<td>100.00</td>
<td>2.28 [1.65, 3.14]</td>
</tr>
<tr>
<td>Total events: 77 (Treatment), 44 (Control)</td>
<td>Test for heterogeneity: Chisq = 2.51, df = 3 (P = 0.47), I2 = 0%</td>
<td>Test for overall effect: Z = 4.99 (P &lt; 0.00001)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*El-Toukhy T, RBM Online, 2012*
UNSUCCESSFUL ART CYCLES;

Embyo quality should be questioned

High Quality

Low Quality
Embryo quality: High

- H/S
- HSG (hydrosalpinx?)
- Endometrial thickness
- IMSI?  PGD?  CGH?  Embryoscope?
- Endometrial Receptivity Assay (ERA)
- Assisted Hatching
Embryo quality: Low

- Reassesment
- Shift to another protocol
- Look over laboratory
- Defragmentation
- Coculture
- Oocyte-Embryo pooling?
Thank you
ÜREME TIBBI DERNEĞİ JİNEKOLOJİK MİKROCERRAHİ KURSU - 4

24 MAYIS 2014 – 25 MAYIS 2014

Adres: Zeiss Eğitim Merkezi (Doğu Kent Bulvarı 450.cadde No:22 Birlik Mahallesi Çankaya Ankara )

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2- Prof. Dr. Erol Tavmergen
3- Prof. Dr. Serdar Dilbaz
4- Prof. Dr. Turan Çetin

Sekreter : Hatice DEDELİ (utd.sekreter@gmail.com / 0533 572 69 87 )
Ayrıca 07-08 Haziran 2014 tarihlerinde İstanbul Yeditepe Üniversitesi Kayışdağı Kampüsü’nde koordinatörlüğünü yine Prof. Dr. Recai Pabuçcu’nun yaptığı ve beşincisini gerçekleştireceğimiz kursumuza da Jinekolojik Mikrocerrahinin kurulucularından Sayın Prof. Dr. Victor Gomel katılacaklardır.