



# ASSISTED REPRODUCTIVE TECHNOLOGIES (ART)

Dr. Herve Lucas, MD, PhD,  
Biologist, Andrologist

Dr. Taher Elbarbary, MD  
Gynecologist-Obstetrician

Geneva Foundation for Medical Education and research

# Definitions of Assisted Reproductive Technologies ?

## Techniques involving oocyte retrieval and ejaculated sperm:

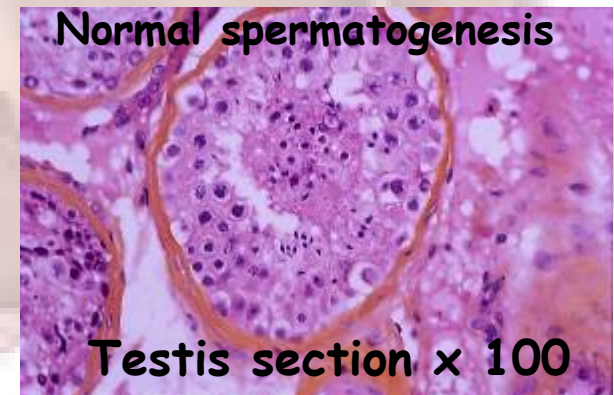
- Gamete Intra-Fallopian transfer (GIFT), Peritoneal Oocyte Sperm Transfer (POST)
- Zygote Intra-fallopian Transfer (ZIFT).
- In Vitro Fertilization (IVF)
- Tubal Embryo Transfer (TET)

## Techniques Including sperm retrieval:

- Testicular Sperm Aspiration (TESA)
- Per-cutaneous Epididymal Sperm Aspiration (PESA)
- Microsurgical Epididymal sperm Aspiration (MESA)
- Non-Scalpel Vasal Sperm Aspiration (NSVSA)

## For injection:

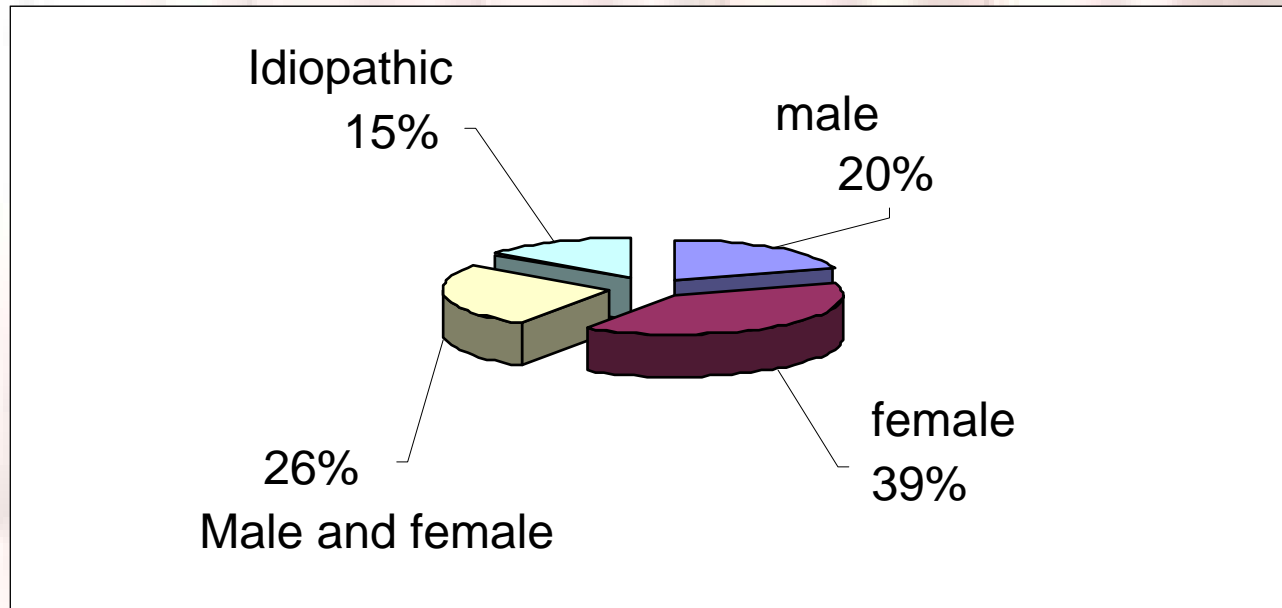
- IntraCyttoplasmic Sperm Injection (ICSI)



Don't forget artificial Insemination...easy, low cost...!

# Who are the candidates of ART ?

**15 % of couples wanting to conceive in Europe**



**Indications for ART *including artificial inseminations...***

# Steps of ART

Monitored hyperovulation

## OOCYTE RETRIEVAL

Oocyte vitrification

Oocyte culture

Extracorporeal insemination

### Sperm retrieval:

- Homologous sperm:
  - By ejaculation
  - By surgical retrieval (MESA-TESE-STW)
  - Sperm bank
- Heterologous sperm:
  - Sperm bank

(MESA-TESE)

Cryopreservation of zygotes due to OHSS risk

Fertilization control

- 30-50 thousands motile spermatozoa in culture with each oocyte (IVF)
- microassisted (ICSI)

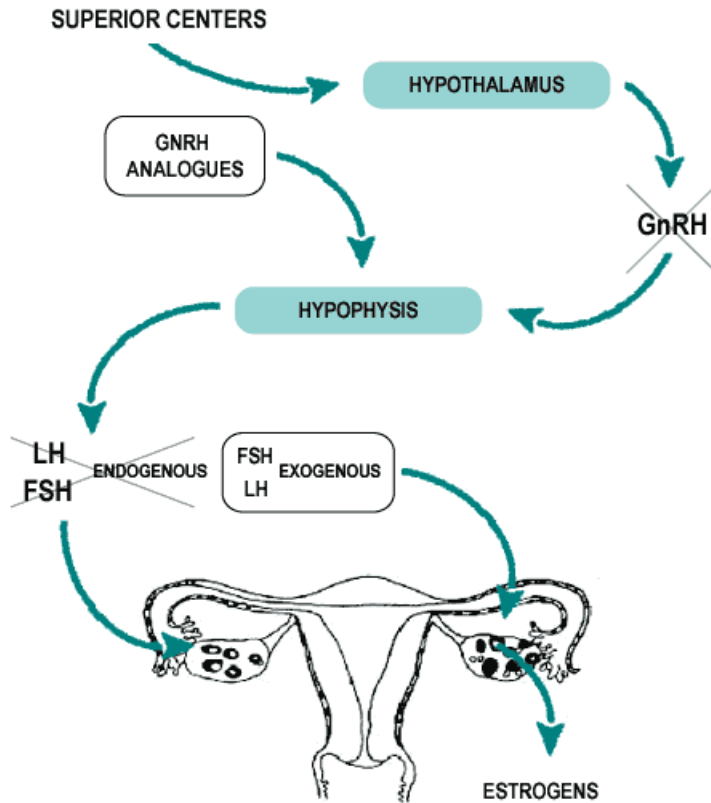
Embryo cryopreservation

Embryo culture

(AZH - biopsy)

Transfer to uterus after 2-5 days

# PATIENT STIMULATION

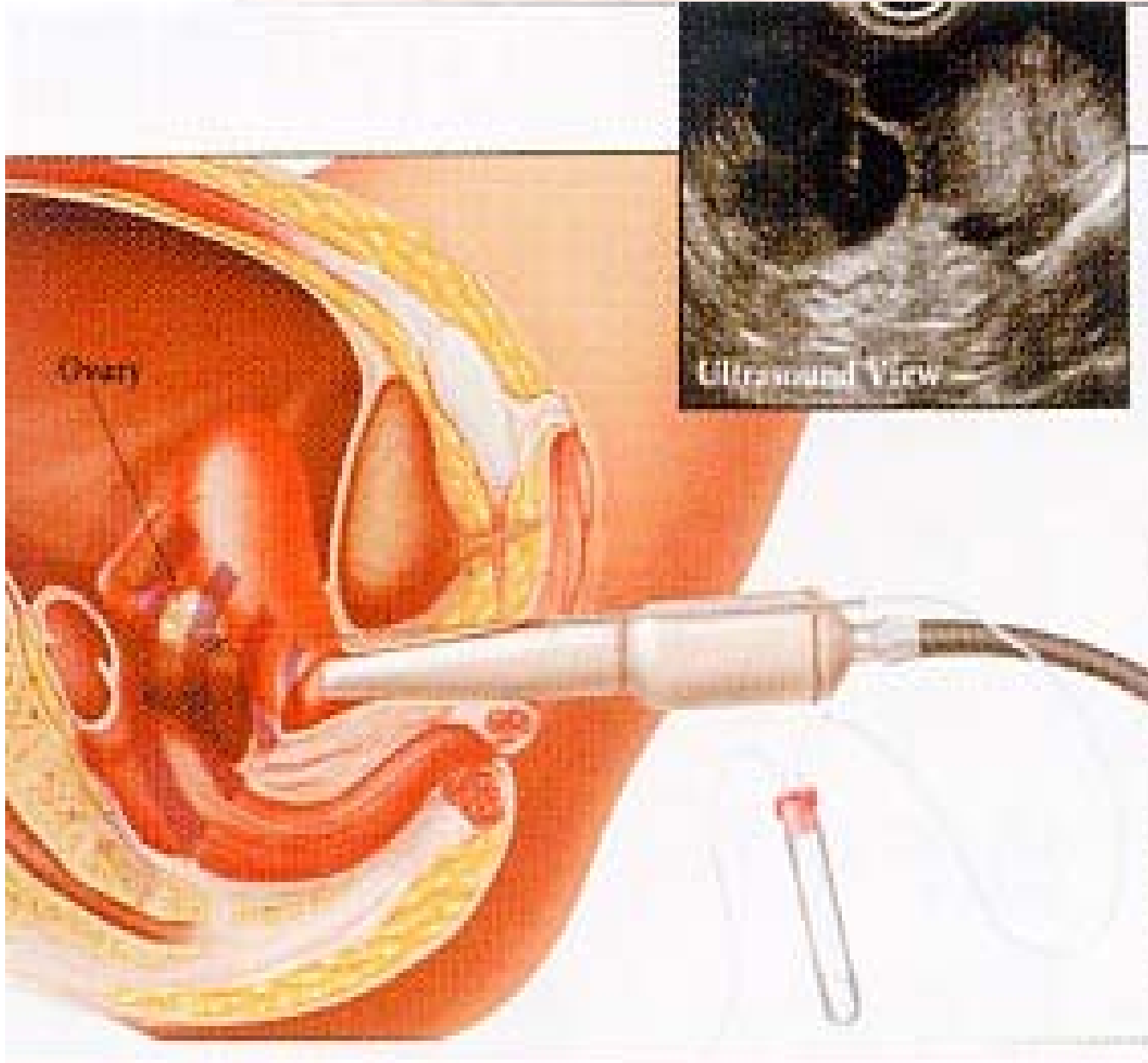


US image of multiple ovarian follicles



Estradiol levels in blood

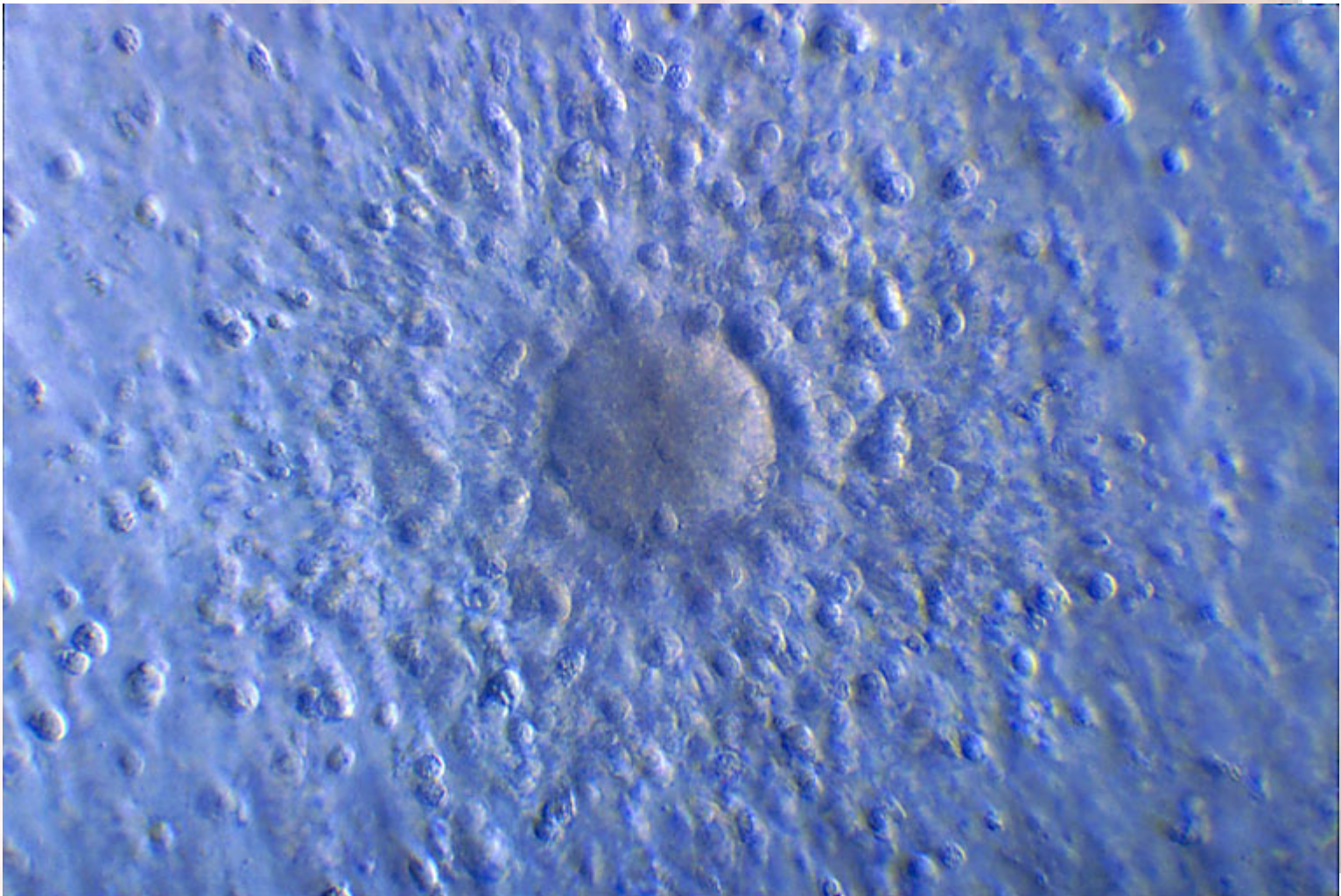
# Oocyte retrieval



# Syringes with cumulus-oocyte complex in follicular fluid



# Cumulus-oocyte complex





# Sperm preparation (various techniques for various utilisations, AI, IVF, ICSI, GIFT...)

## 1- Sperm retrieval



## 2- Sperm washing



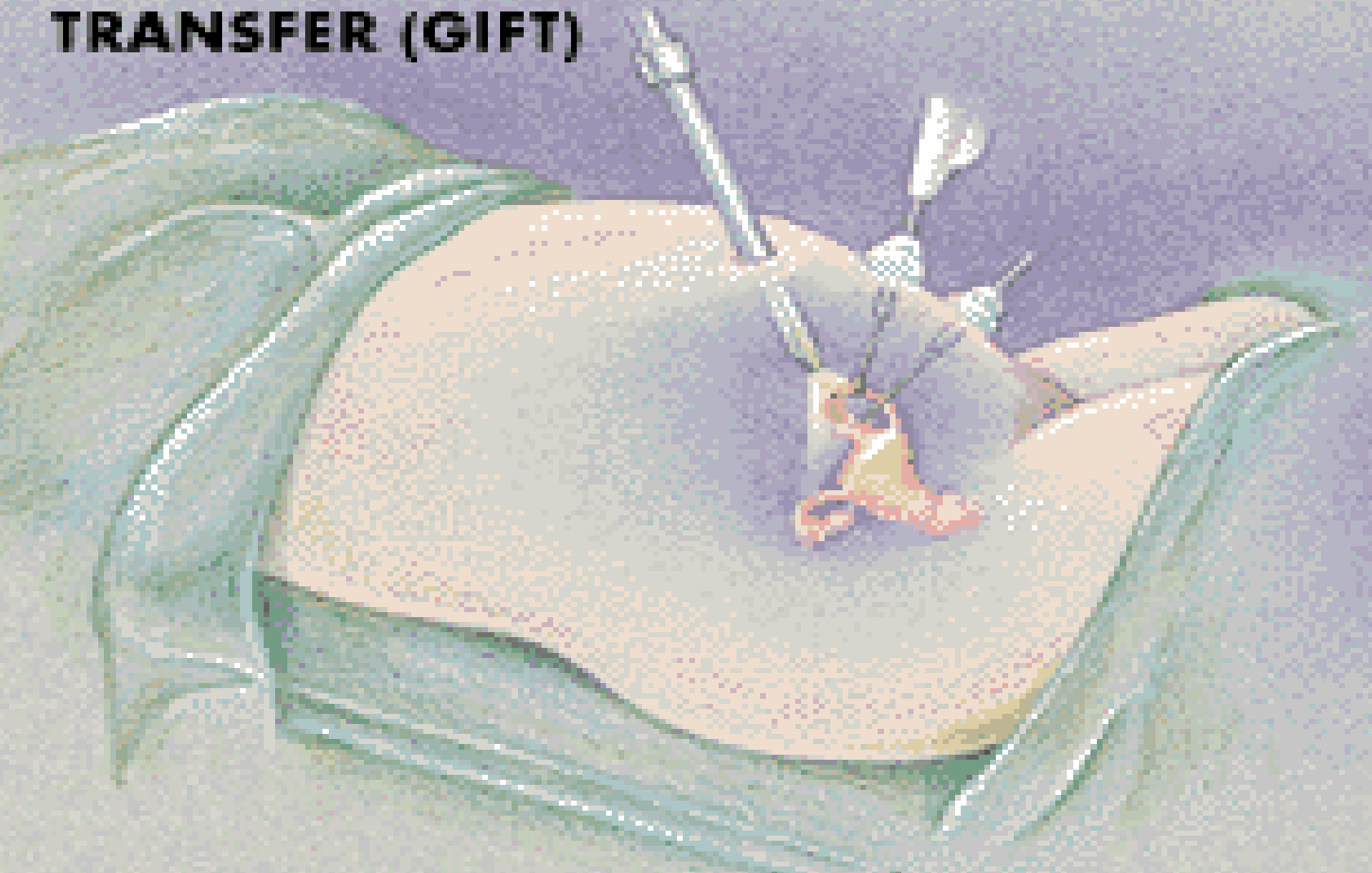
## Adaptation of the sperm preparation :

- Ejaculates vs freezing-thawed,
- Very poor sperms vs normospermia,
- Ejaculates vs testicular or epididymal,
- Sperm with antisperm-antibodies,
- Retro-ejaculated sperm....

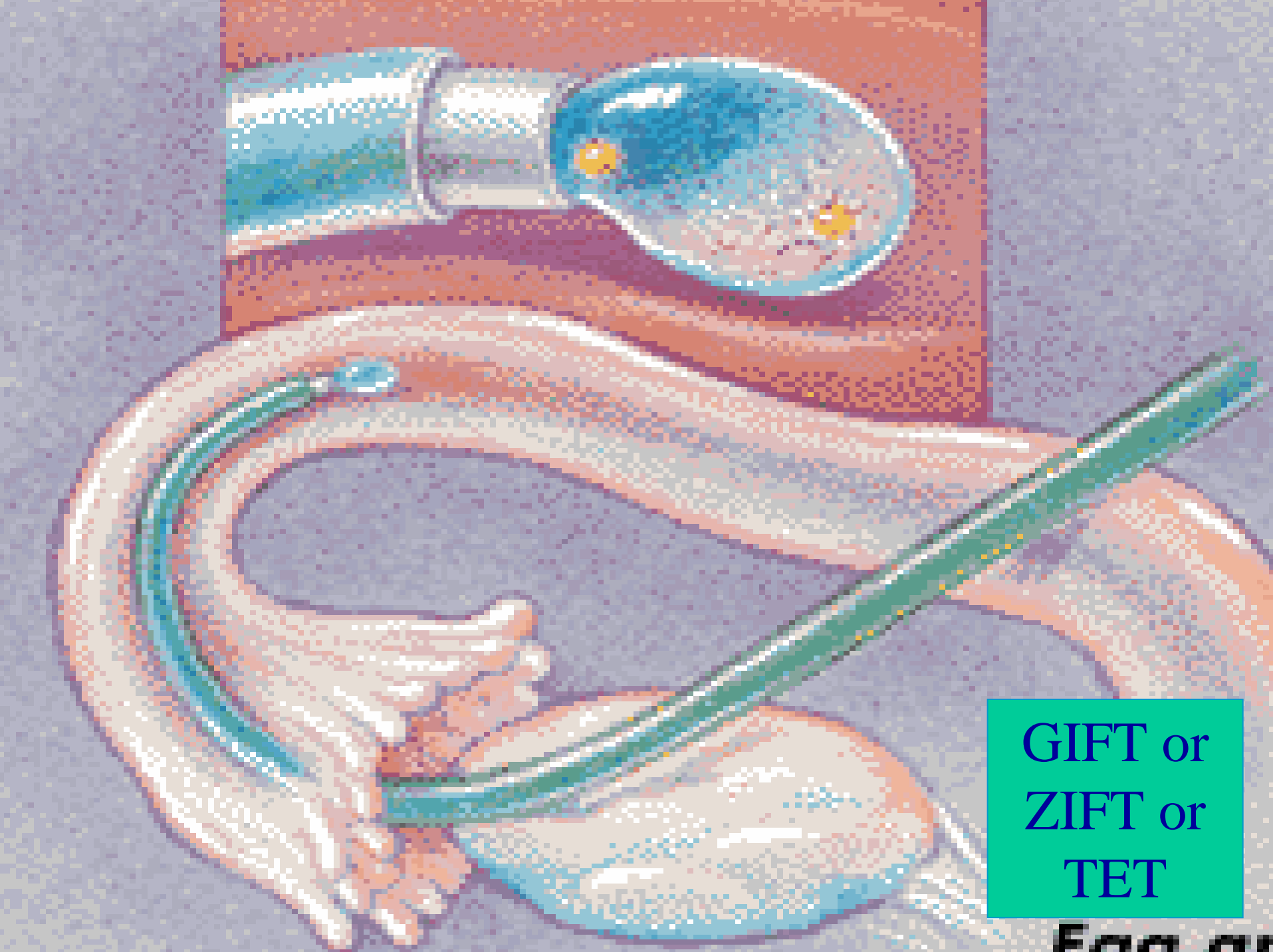
# Artificial inseminations (indication : >1M motile spz, tubes OK..)



# GAMETE INTRAFALLOPIAN TRANSFER (GIFT)



**Laparoscopy**

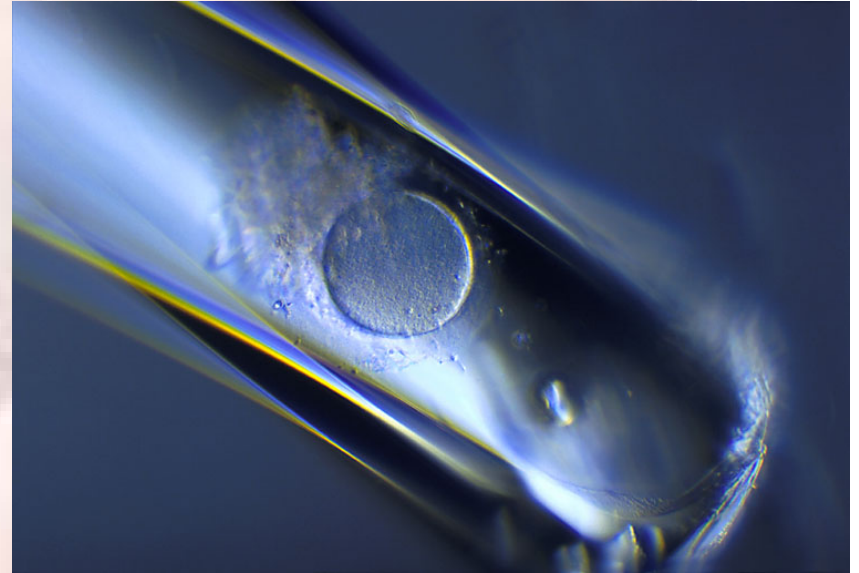


GIFT or  
ZIFT or  
TET

Egg on

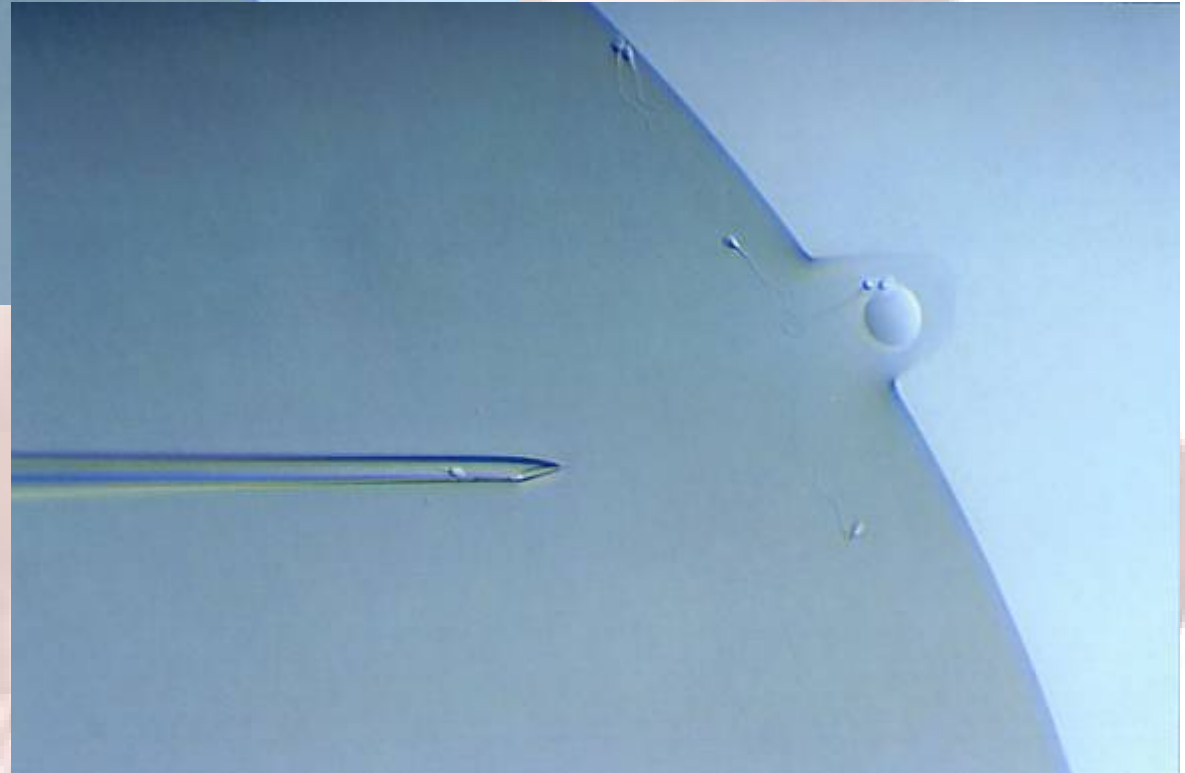
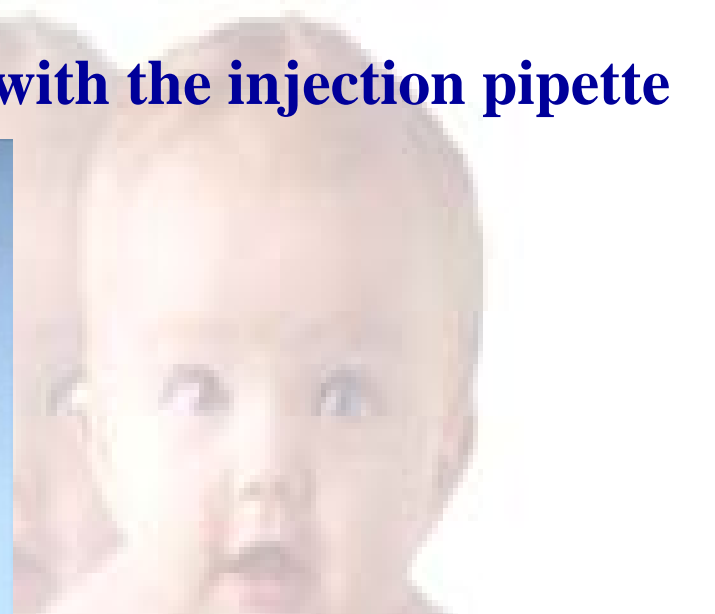
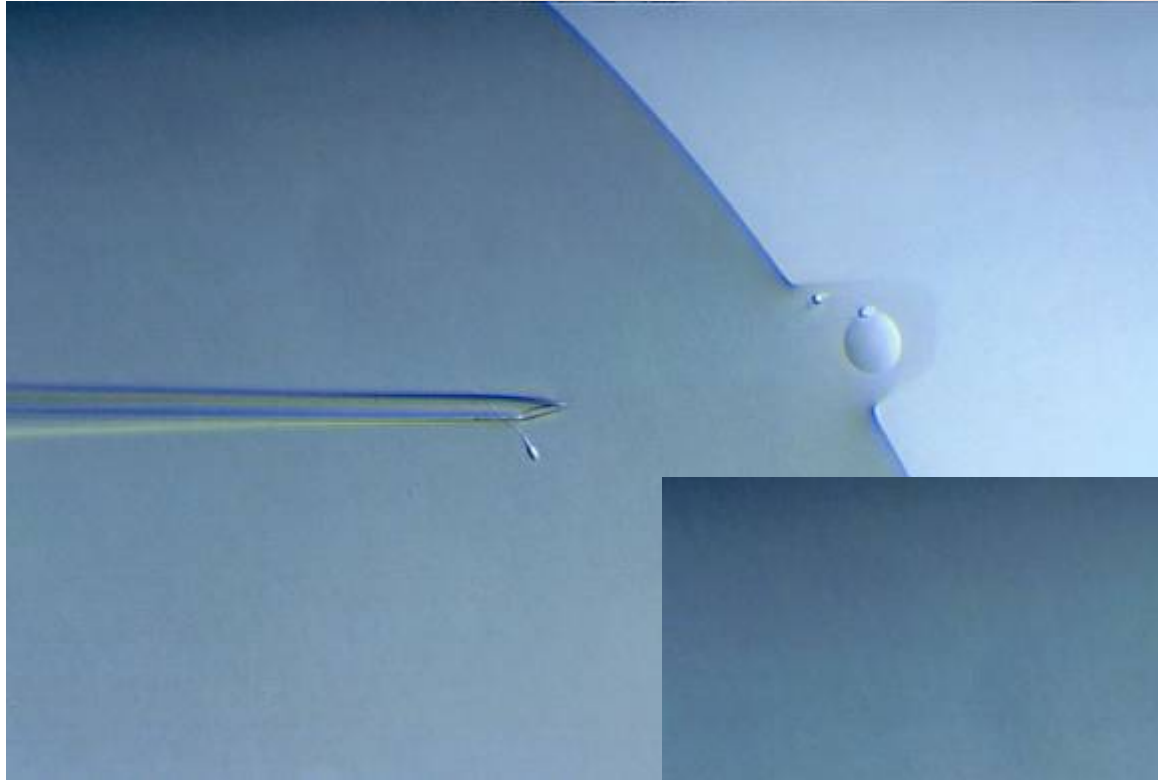
# ICSI

**Day (0): Decoronation of mature oocytes before injection (MII)**





# Day (0): Catch a single sperm with the injection pipette



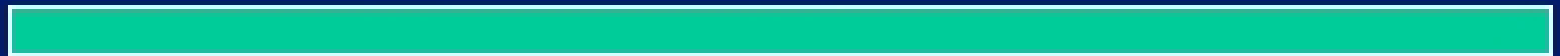
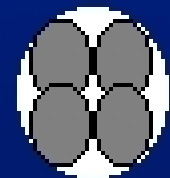
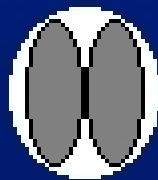
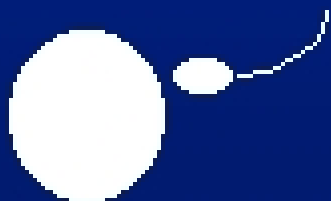
ICSI

# Intra-Cytoplasmic Sperm Injection Day (0)





# Fertilization control...and embryo development.

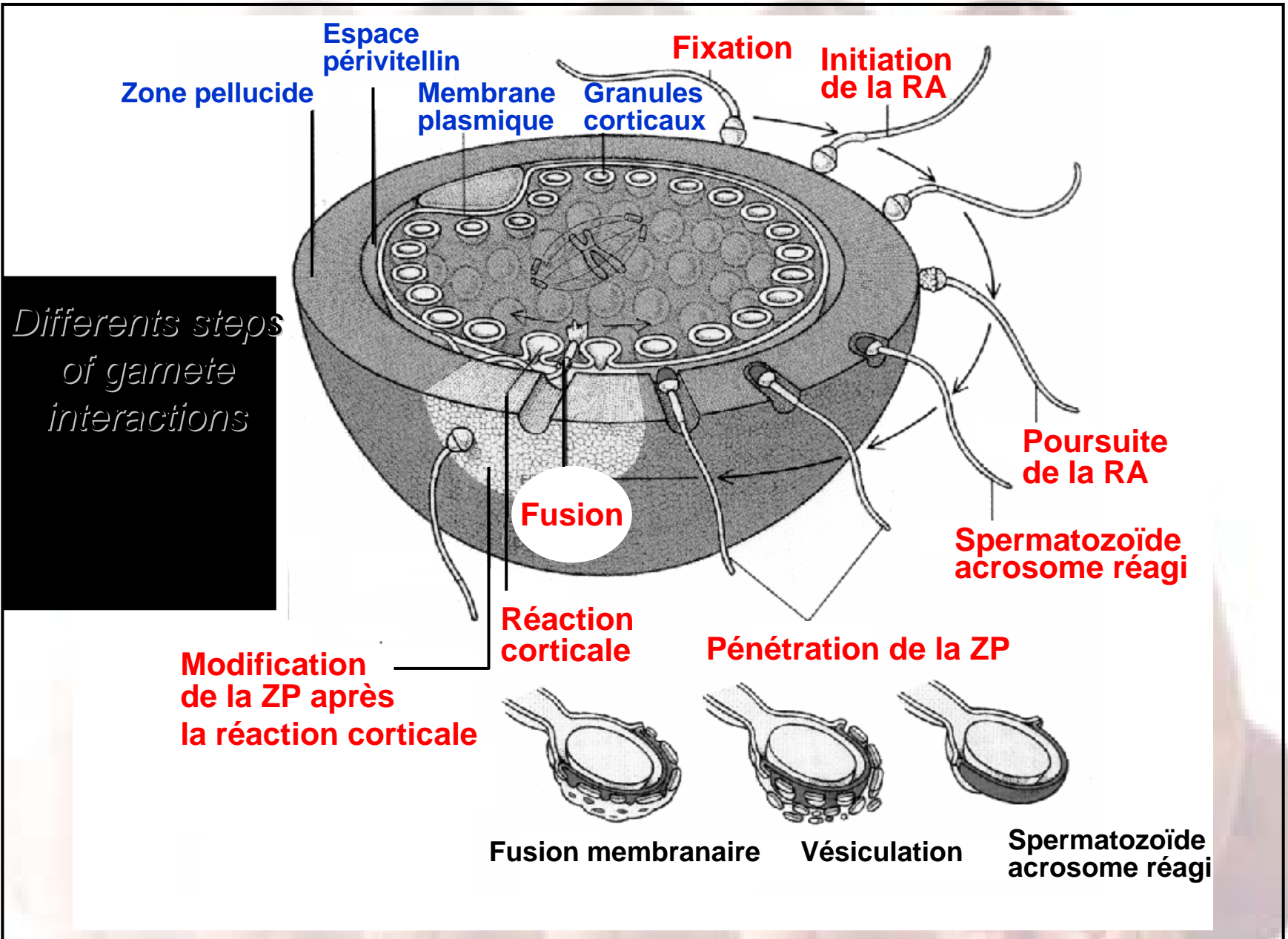


Insemination  
0 h

2 PN  
16-18 h

Early 2-cells  
25 h

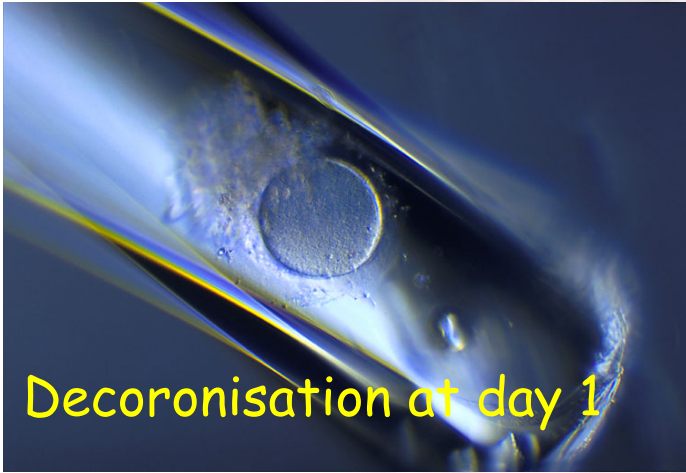
Transfer  
42-50 h



D 'après P. Wassarman

# Observation of zygotes

Day (1) 16-18 h **post-classical IVF**

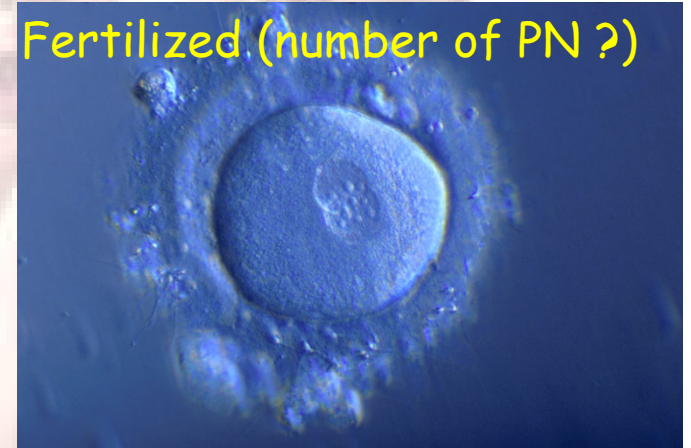


Decoronation at day 1



Living spermatozoa  
(motility) at 16-18h

Sperm-ZP binding



Fertilized (number of PN ?)



Unfertilized (1 polar body)

**Fertilization rate :**

1 PN zygote : problem of oocyte activation ? (spz or nucleus oocyte origin ?).

3 PN after ICSI : non extrusion of the second polar body... by lesion of meiotic spindle ?

3 PN after IVF : polyspermy = too many spz inseminated or bad quality oocyte because abnormal cortical granules)

This case: ICSI



**Abnormal fertilization**

(1 or 3 PN and 1 or 2 PB)

# Observation of zygote

Day (1) 16-18h post IVF or ICSI

**Normal fertilization**

(2PN and 2PB)



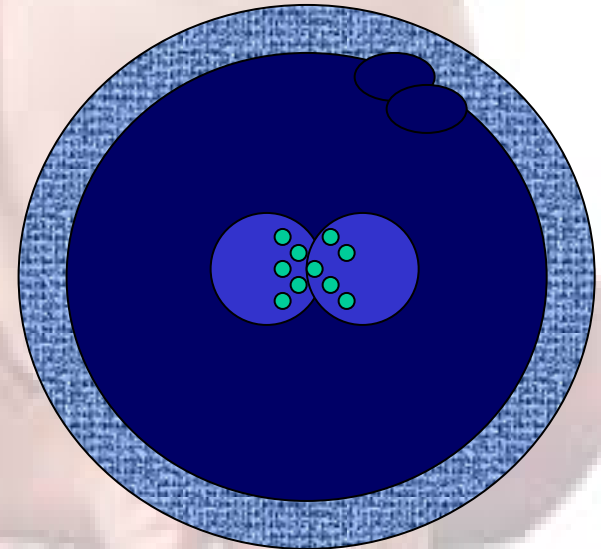
This case: IVF

Scoring of zygotes...  
Correlated with embryo quality (J2-J3)

•Day (1) (**PM**) two cell embryo : the early cleavage (25 h),

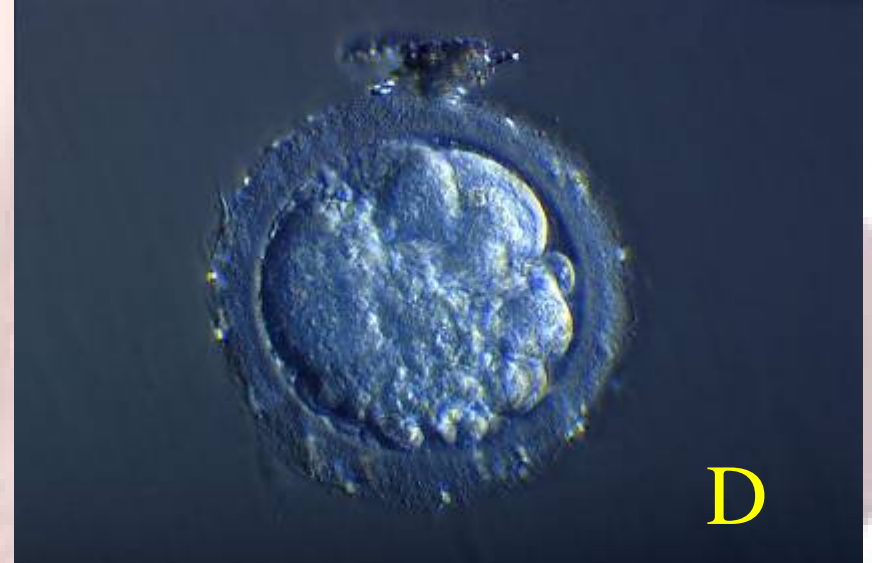


The same  
indication :  
good timing  
in embryo  
development.



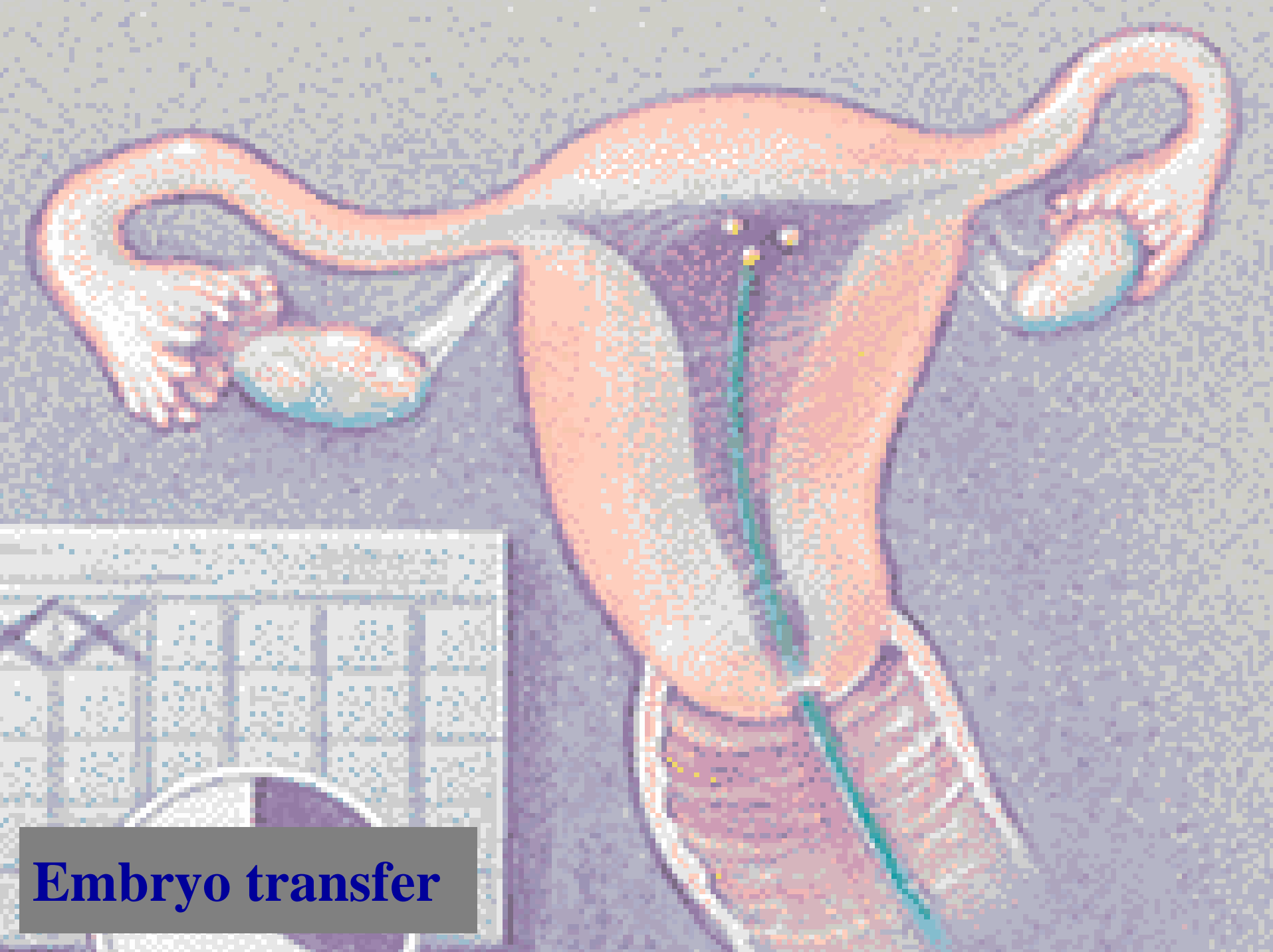
•Day (1) (**AM**) Zygote stage : best scoring zygote (16h),

# Day (2): 4-cell stage embryo quality.



## Day (4): Morula stage





**Embryo transfer**



# What are the risks of ART ?

Risk due to ovarian stimulation ?

Risk due to oocyte retrieval ?

Risk due to embryo transfer ? Multiple pregnancies

Risk due to ICSI ? Increased !!!

Double in comparison with naturally obtained pregnancy (male genetic analysis in oligozoospermia)

Risk due to Classical IVF ?

Increased in comparison with naturally obtained pregnancy (bias of observation of the children ? Techniques *in vitro* ?)

# What are the success rates of ART ?

## IVF

Deliveries per retrieval ..... 29.1%

## ICSI

In women < 35 years without male factor infertility..... 35.7%

In women > 35 years with male factor infertility..... 35.1%

In women < 40 years without male factor infertility..... 10.3%

In women > 40 years with male factor infertility..... 12.8%

## GIFT

In women < 35 years without male factor infertility..... 34.9%

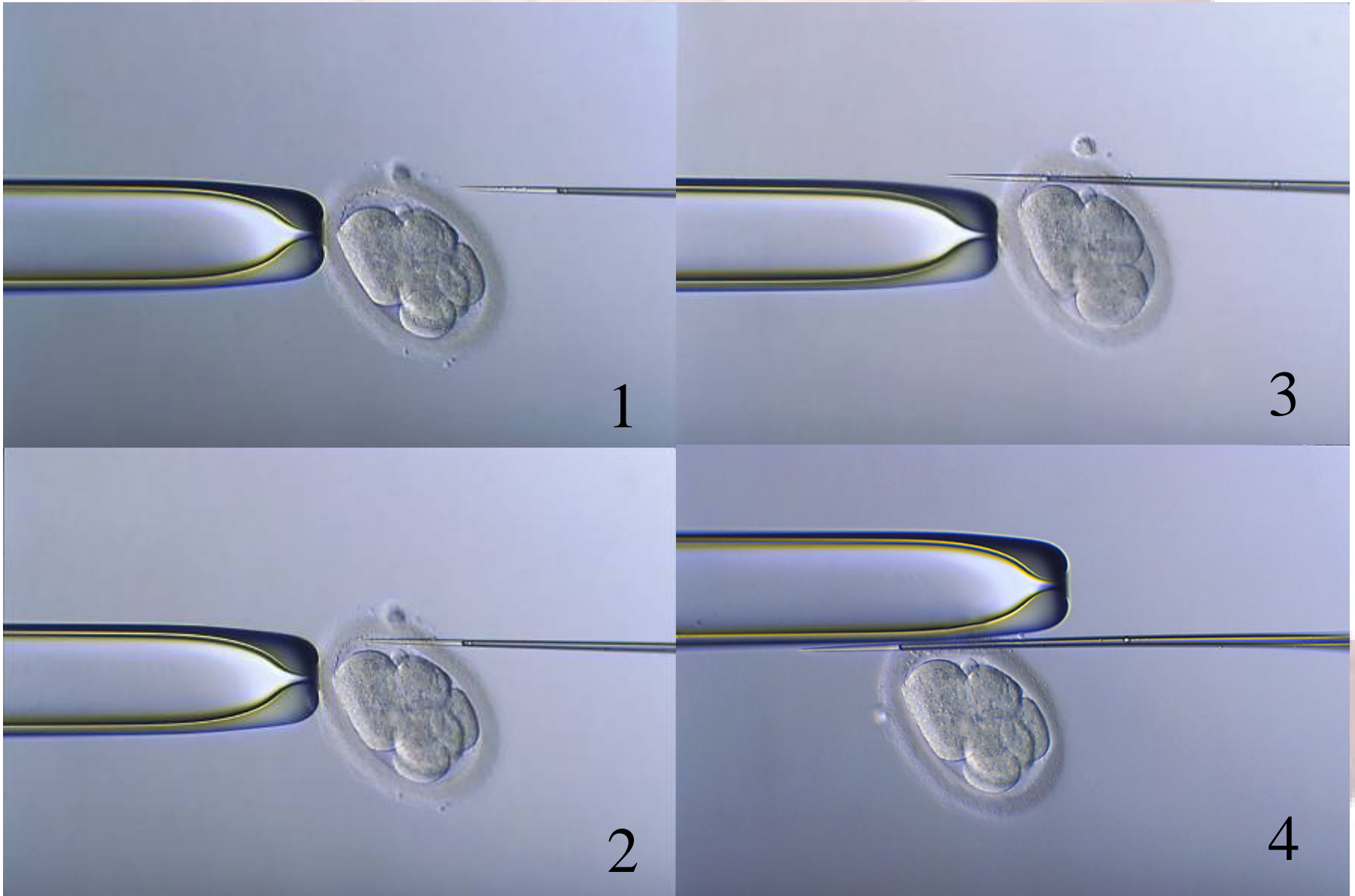
In women > 35 years with male factor infertility..... 22.2%

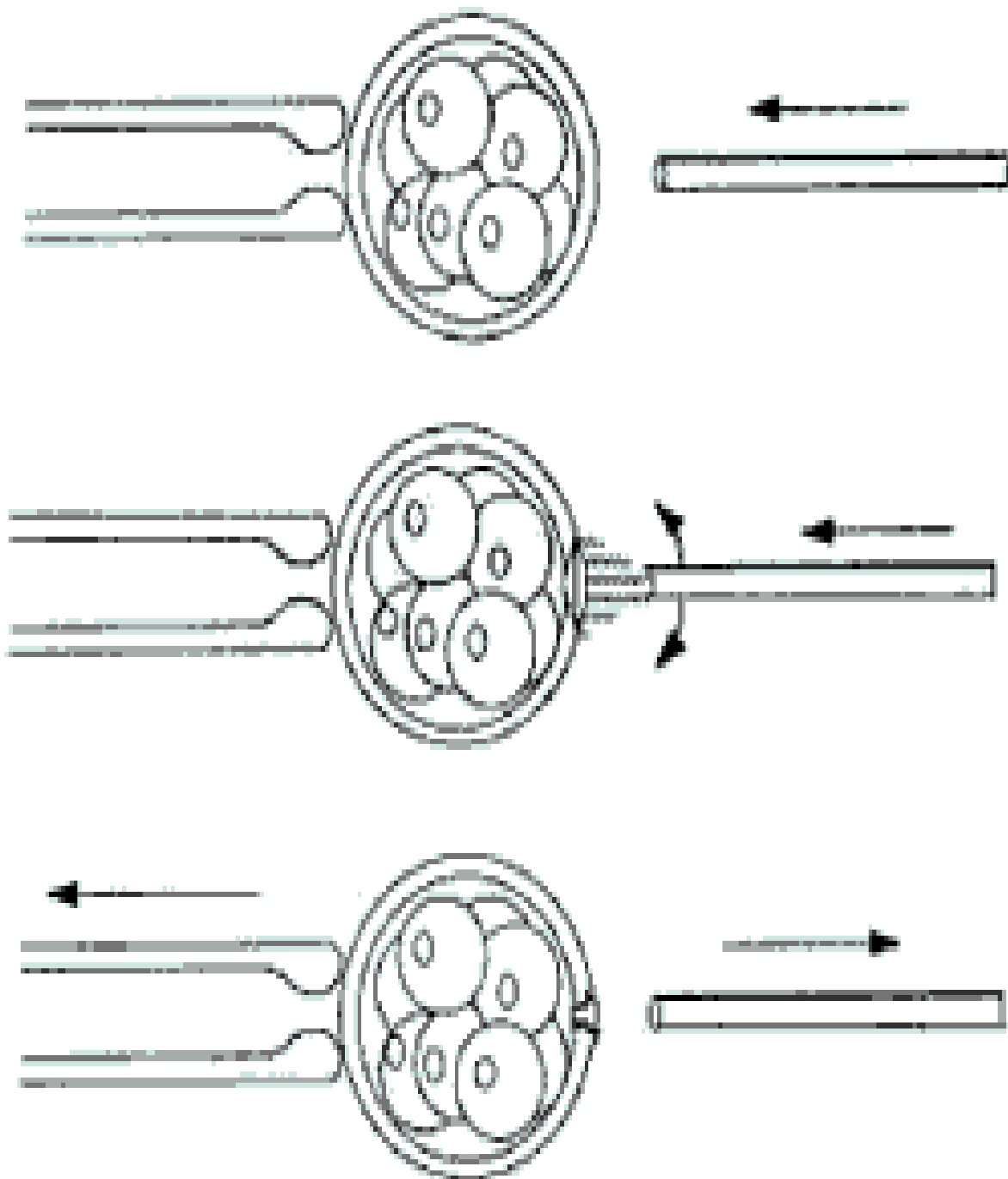
In women > 40 years without male factor infertility..... 09.1%

In women > 40 years with male factor infertility..... 11.1%

**Nb of transferred embryos, indications, AI before IVF ???**

# Mechanical assisted hatching

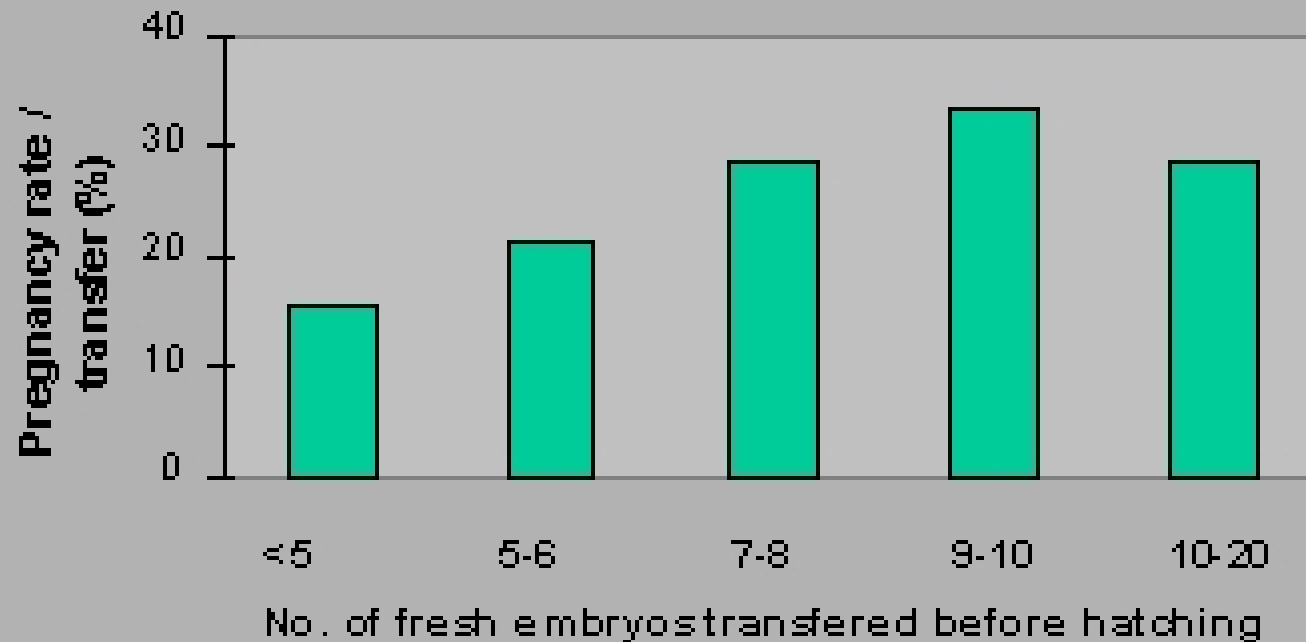




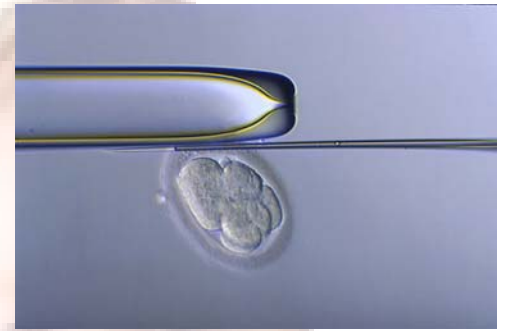
**Assisted  
hatching  
with  
acidic  
solution...  
now with  
LASER  
safer  
technique!**

# Correlation Between the Number of Embryos Transferred in the Previous Cycles and the Pregnancy Rate After the Assisted Hatching

## Pregnancy Rate / No. of Transferred Fresh Embryos in The Previous Cycles



# Our own experience (Paris + Geneva)



## Assisted Hatching indicated in :

- Failed Embryo Transfer (more than 3 ET of 2 good quality embryos).

and/or

- Thick Zona Pellucida ( $>15 \mu\text{m}$ )- Hard ZP

But ...



ESHRE analysis don't confirm the interest of Laser Assisted Hatching in any indication

Further study needed ?

# Pre-implantation genetic diagnosis (PGD)



Genetic analyze (x2 cells):

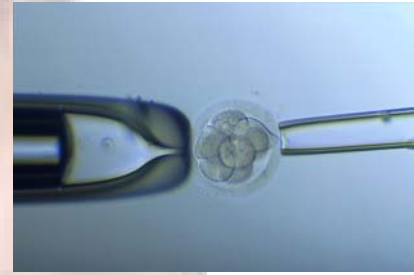


Number of Chr. : FISH

Gene alteration : PCR




# Pre-implantation Genetic Diagnosis (PGD)



- The benefits of PGD in **infertile** couples (2 Polar body, PCGD).
  - Research of aneuploidy in older women (decrease miscarriage)
- The benefits of PGD in **fertile/infertile** couple.
  - Sex embryo screening (Ethically discussed...)
- Who should have PGD ?
  - Fertile or infertile couples with known and genetically detectable disease.

**Very good genetic laboratory needed !**

# What is the future in ART :

- **Ovarian tissue cryopreservation and graft.**
  - **In Vitro Oocyte Maturation.**
  - **Reducing the number of embryo transferred (blastocyst culture development).**
  - **Pre-Implantation Genetic Diagnosis.**
  - **In Vitro maturation of male germinal cells.**
- 
- A group of approximately ten babies are sitting on a white surface, possibly a floor or a table. They are all looking towards the camera with various expressions. The babies are of different ethnicities and are dressed in simple clothing. The background is a plain, light-colored wall.

**What is the future in ART :**

**....To have primate model for evaluation of :**

**NEW BIOLOGICAL TECHNOLOGIES,  
NEW MALE/FEMALE TREATMENTS,**

**....Before Human using.**

