REPRODUCTIVE TECHNOLOGY

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Background

- Science and scientists
- Experimental animals
- Target animals

Tools

Applications

- Current
- Future

Ethics

Science

- Create new knowledge
  - Thinking
  - Experiments
- Apply knowledge
- Teach, publish information

Target Animals

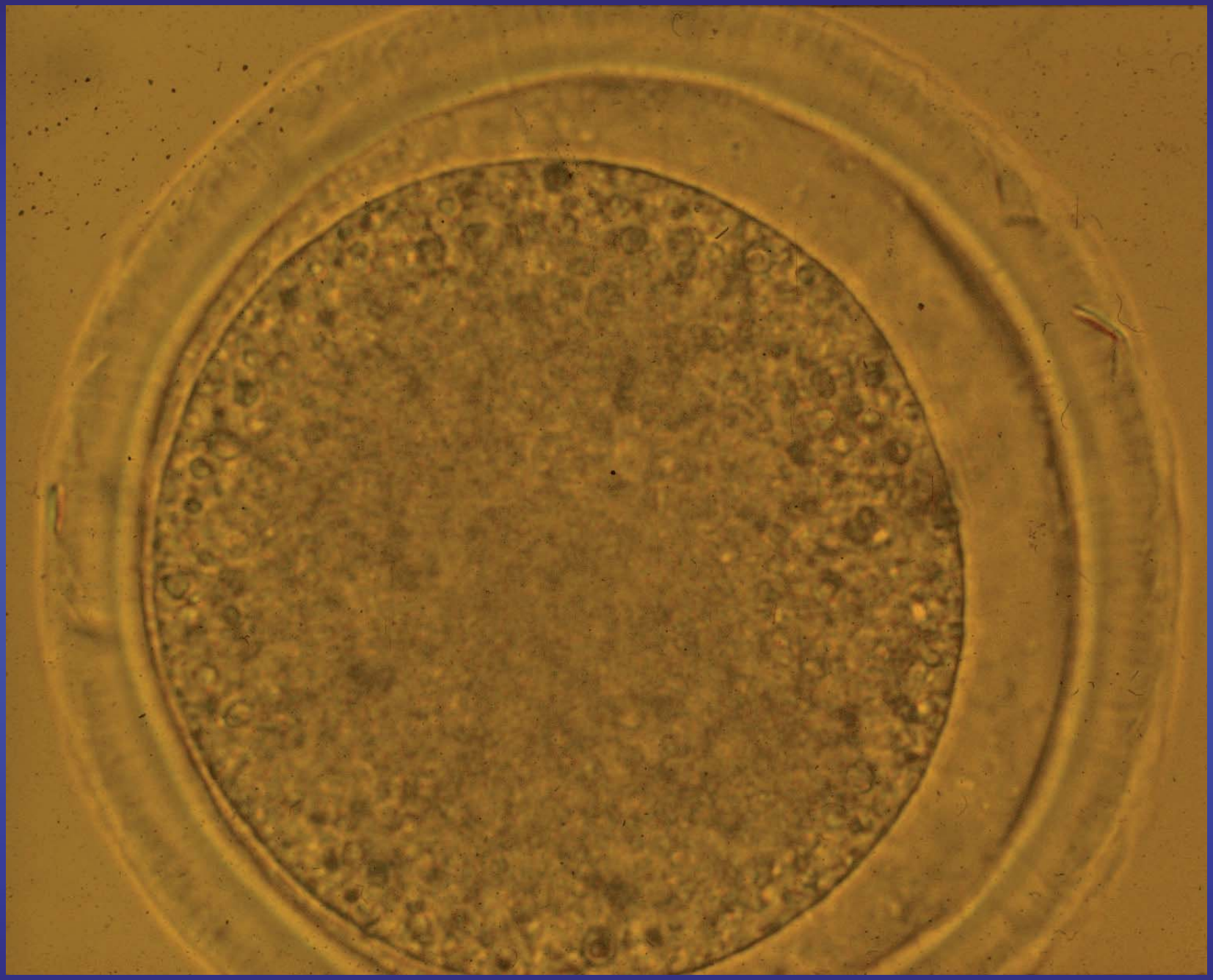
- Laboratory animals
- Farm animals
- Companion animals – dogs, cats, horses
- Wildlife and zoo animals
- People

Experimental Animals

- Laboratory animals
- Farm animals
- Target animals

Omne Vivum Ex Ovo

- Egg (oocyte)
- First week of embryonic development
- Sperm also essential



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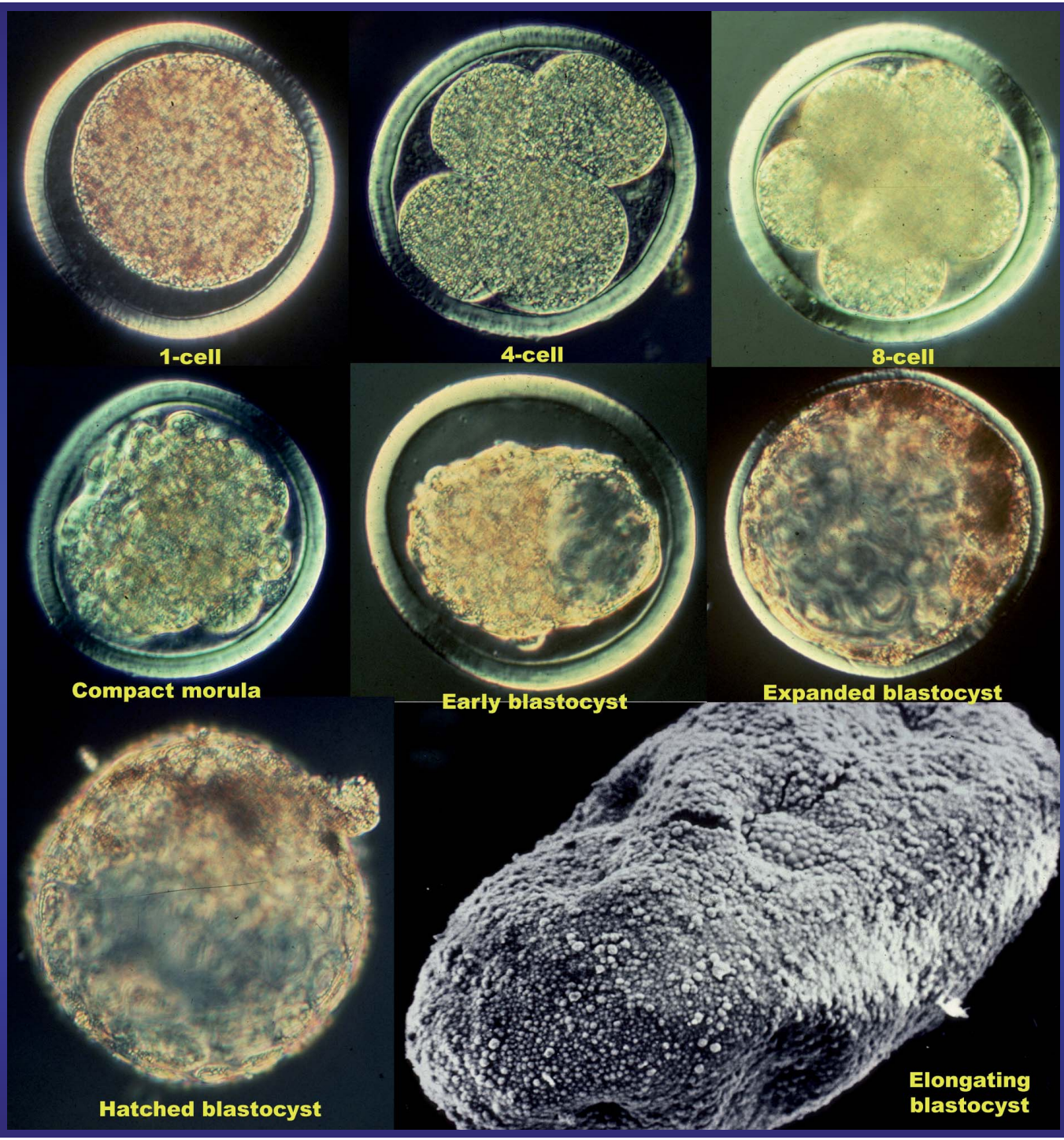

1-cell
4-cell
8-cell
Compact morula
Early blastocyst
Expanded blastocyst
Hatched blastocyst
Elongating blastocyst

TOOLS

Half of Nobel prizes in physiology or medicine concern new tools

New Tools

- New/greatly improved tools appear constantly
- About every 7 years, truly novel tools occur which revolutionize science and applications

Revolutionary Tools

- Recombinant DNA
- Cryopreservation of embryos
- Transgenic technology
- Somatic cell nuclear transplantation
- Polymerase chain reaction
- Fertilization by sperm injection
- Stem cell biology

Embryos

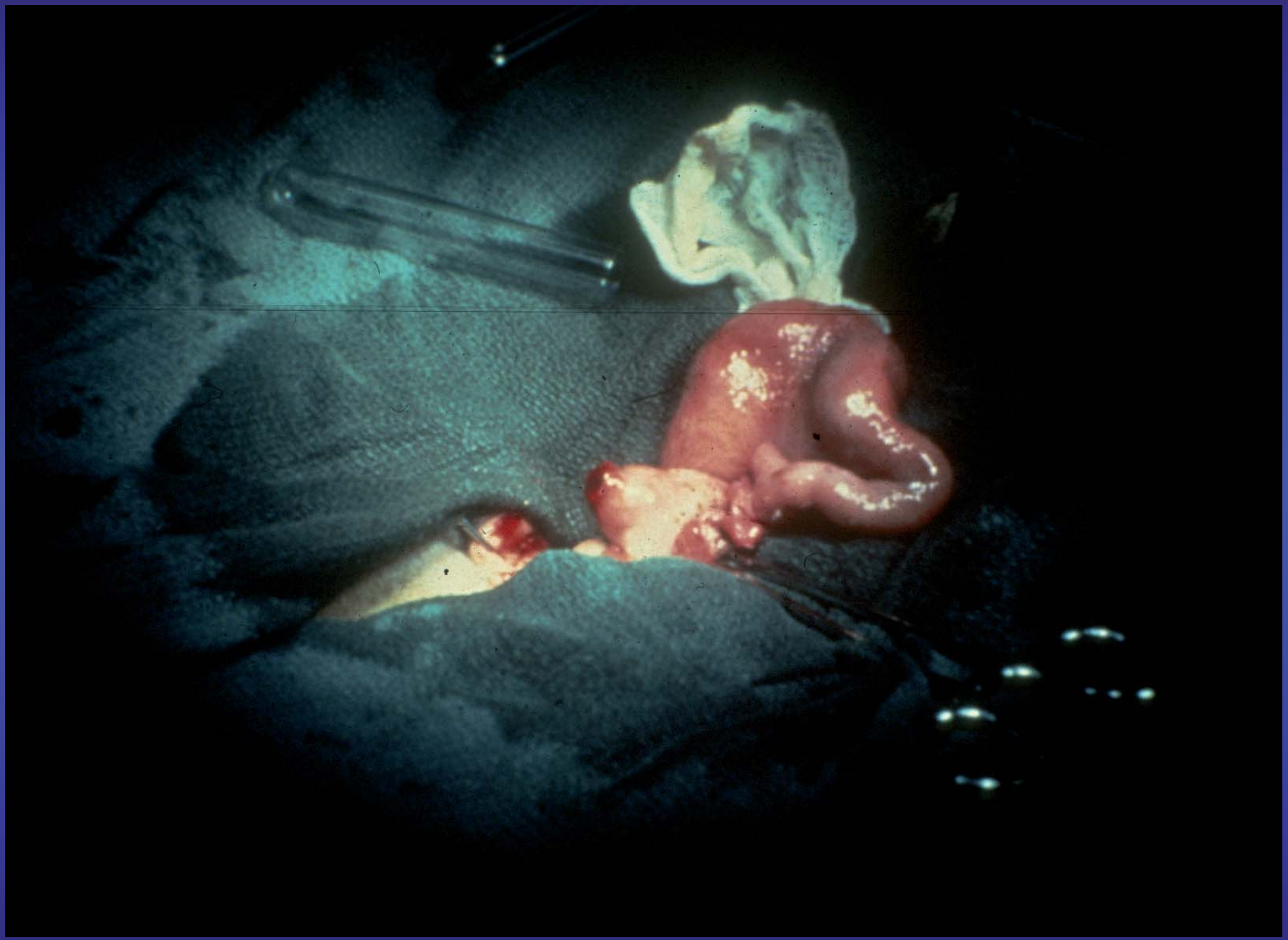
- Create by IVF or recovery from the reproductive tract
- Culture for days
- Freeze and store at -196°C for decades
- Determine sex
- Determine genetic make-up

Embryos

- Separate cells to make identical twins, triplets, quadruplets
- Mix 2 embryos to make chimera
- Add, delete, or correct genes
- Transfer to reproductive tract to make an animal/person

The Tools of Reproductive Technology

Superovulation



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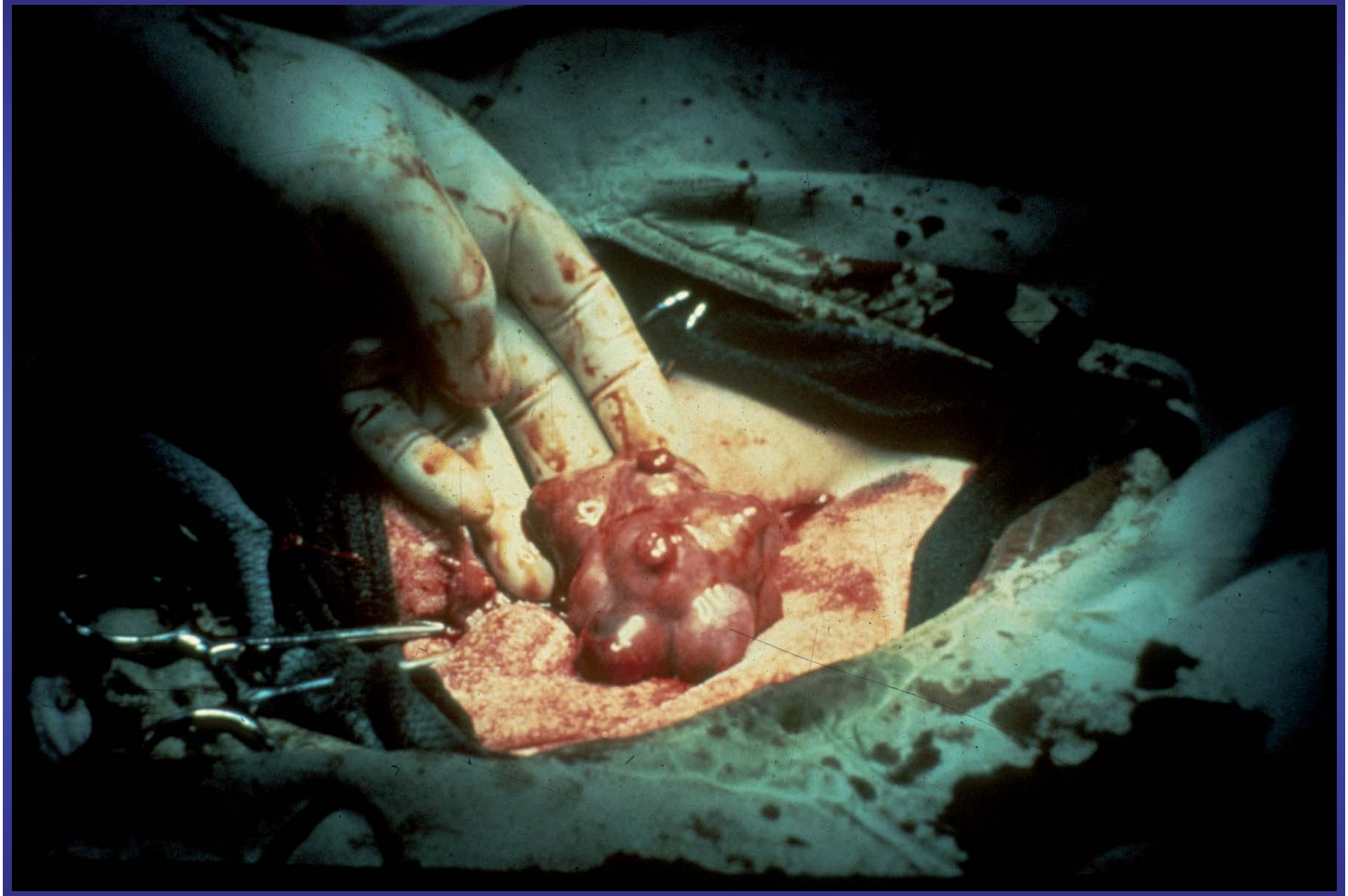

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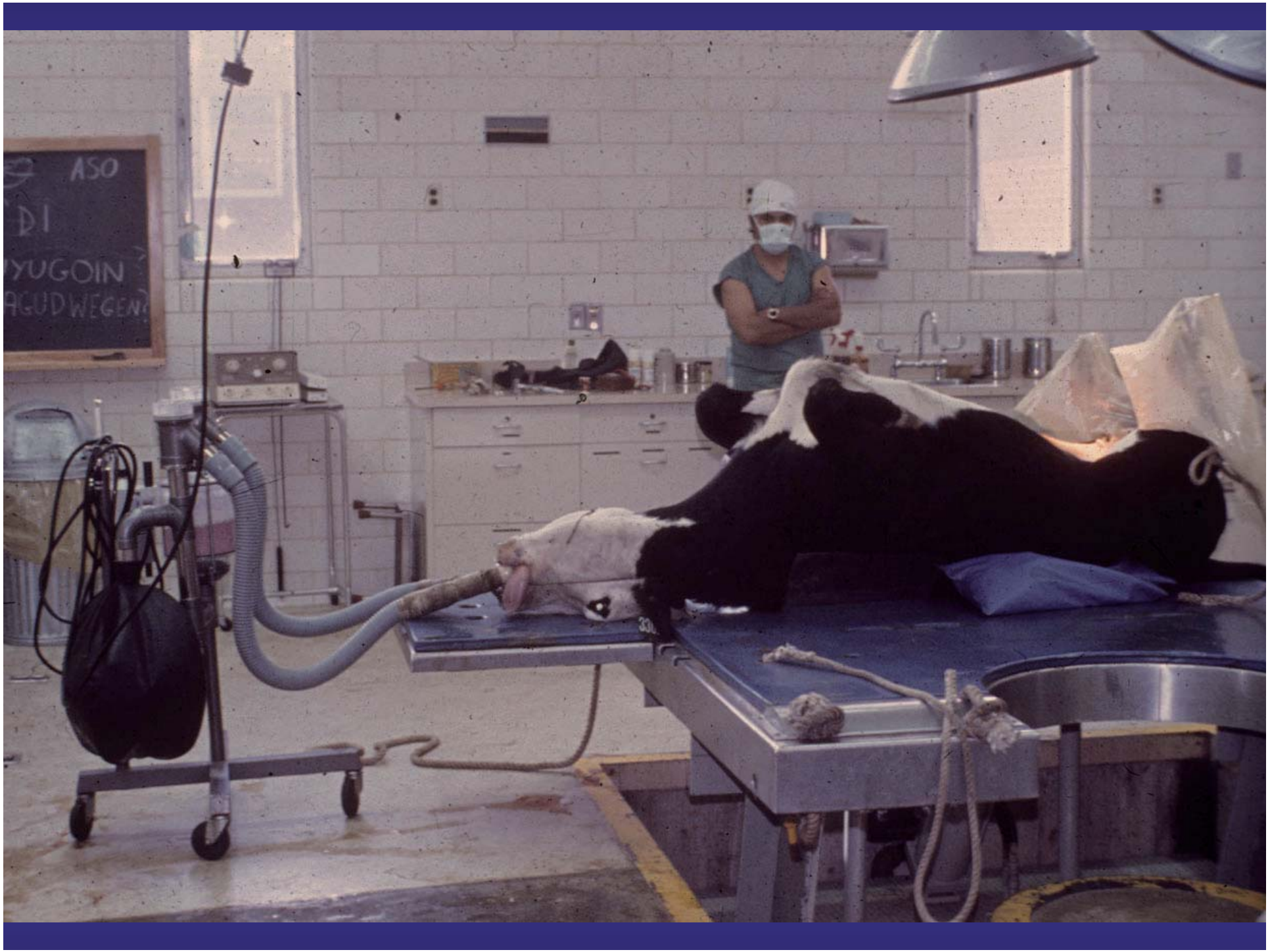
3- to 10-fold increase in egg production

Embryo Recovery and Transfer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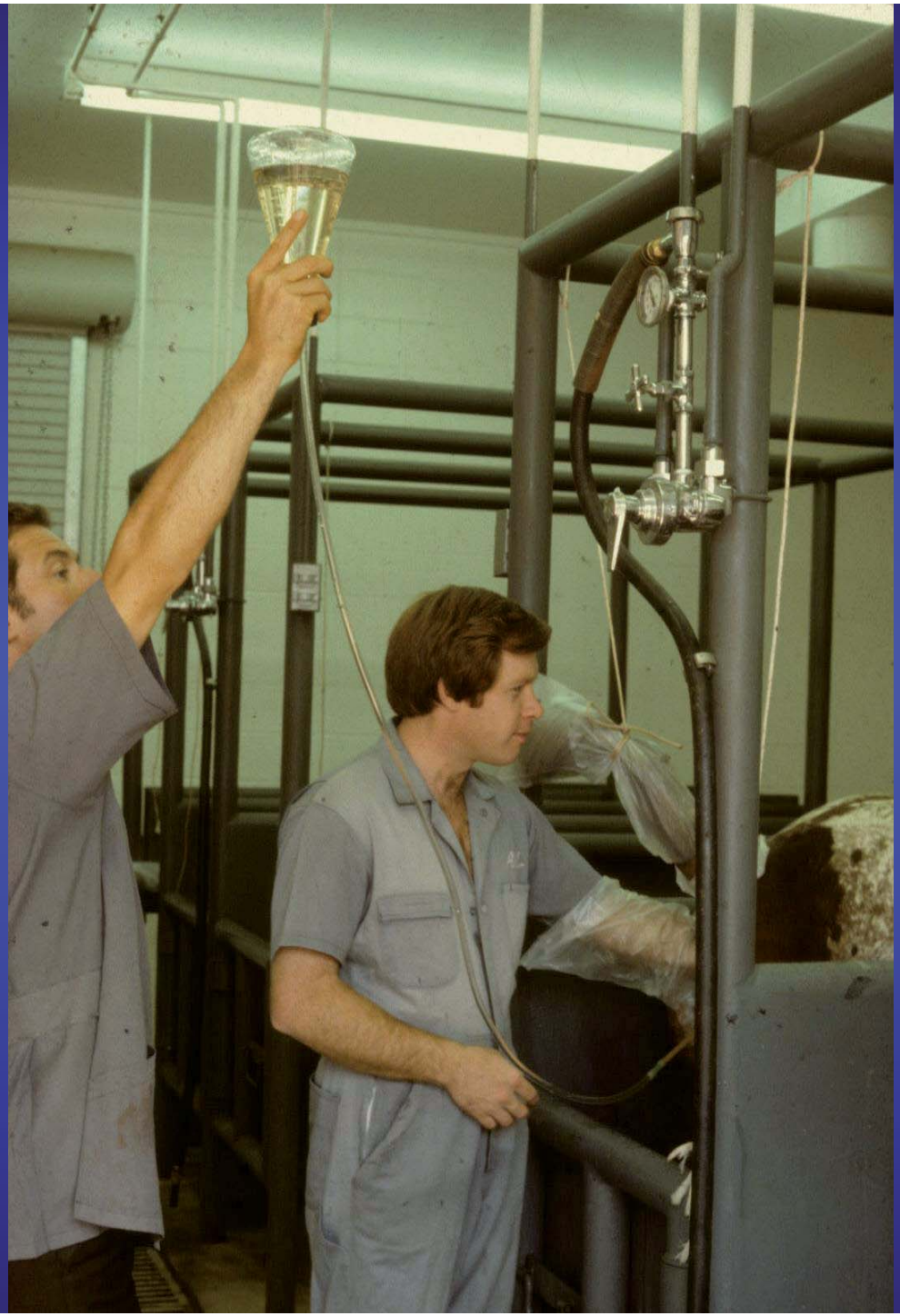


Rescuing Genetics

- Animals infected with viruses
- Circumvent infertility
- Endangered species

SEXING SPERM

HO33342 binds to DNA

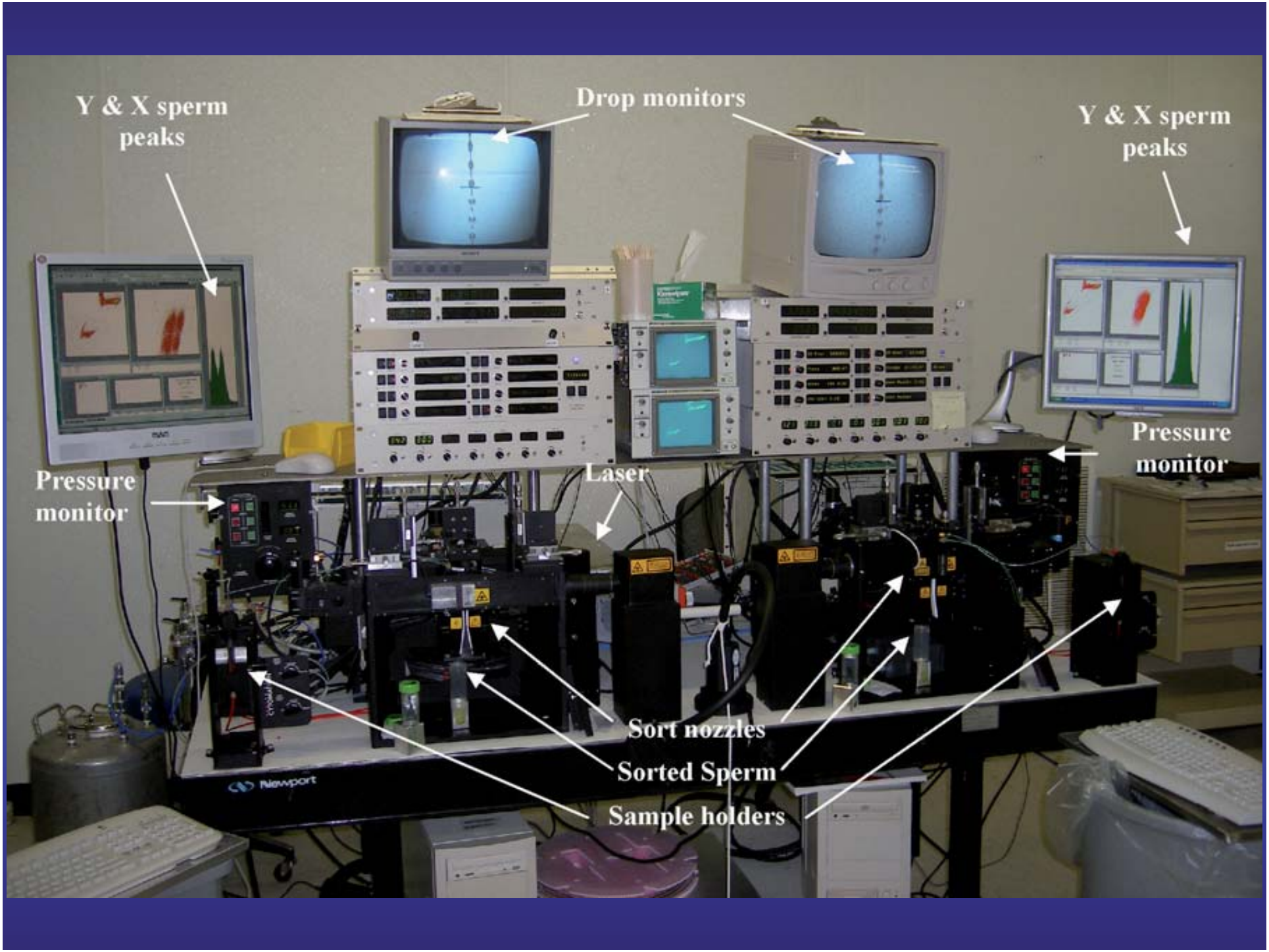
X- sperm have more DNA

Aim laser light at sperm

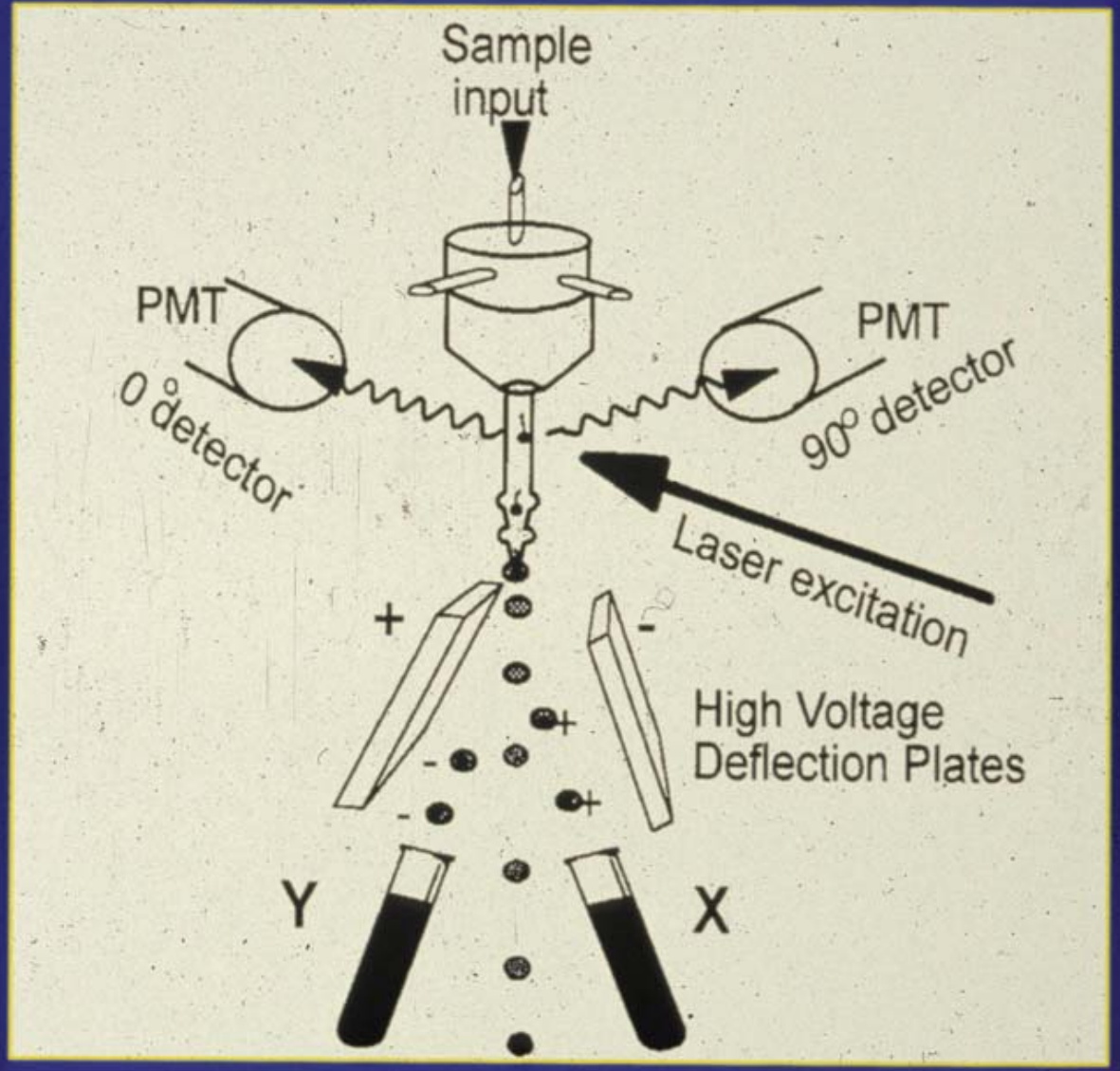
HO33342 fluoresces

Measure fluorescence

Computer analysis


Y & X sperm peaks
Drop monitors
Y & X sperm peaks
Pressure monitor
Laser
Pressure monitor
Sort nozzles
Sorted Sperm
Sample holders
Newport

Sorting by charge



Johnson et al., J. Anim. Sci. 77, Suppl. 2J:213-220, 1999

SPERM SORTER

25,000 sperm/sec

80,000 drops/sec

180,000 measurements/detector/sec

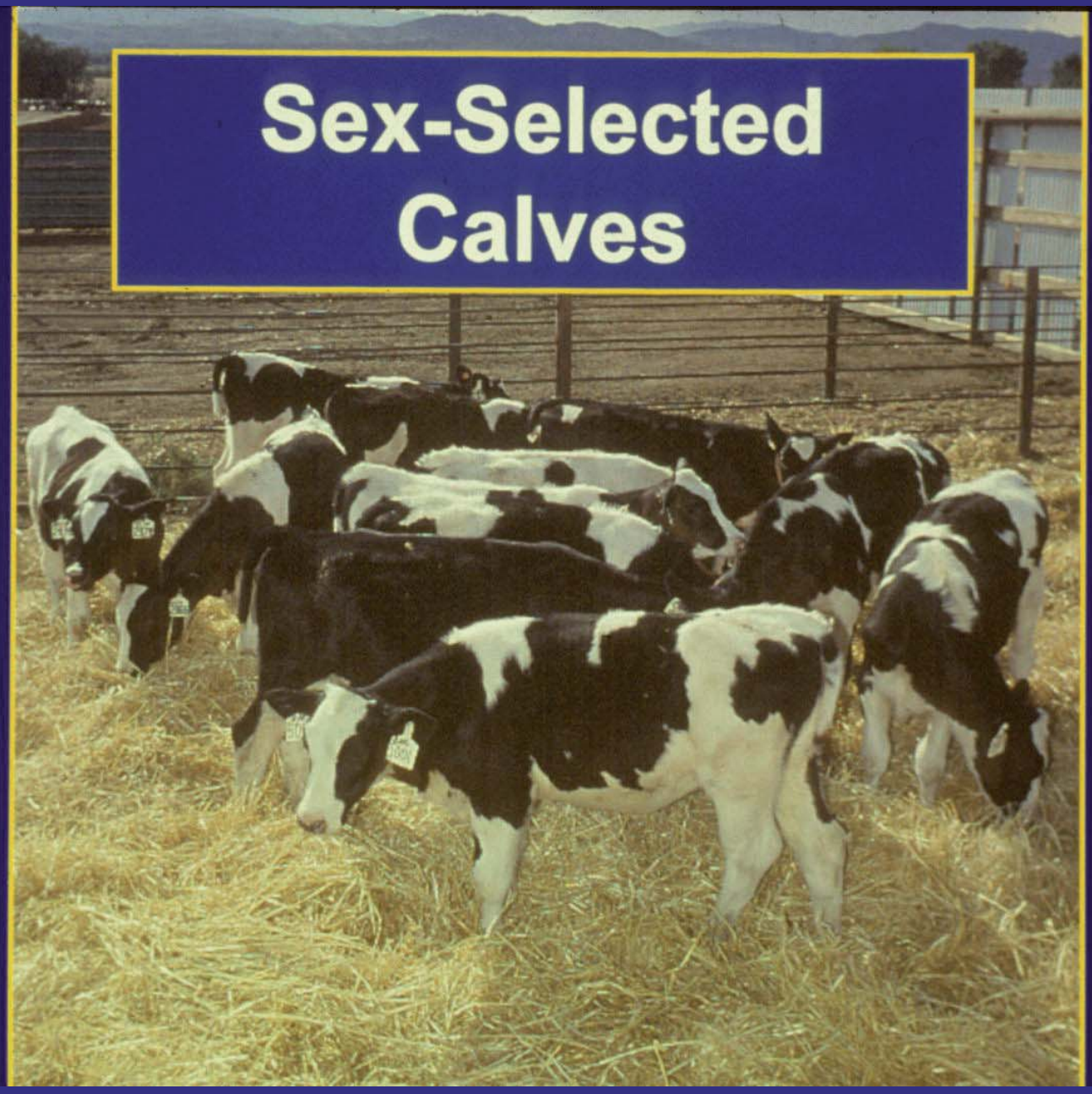
80 km/hour

Cost: >$500,000 for 2-nozzle version

PURITY

- Can exceed 95%
- Industry standard = 90%
- More pure = more expensive
- Similar accuracy X and Y

Sex is THE most important genetic trait


Sex-Selected
Calves

Cryopreservation

- Sperm
- Oocytes
- Embryos


CSU
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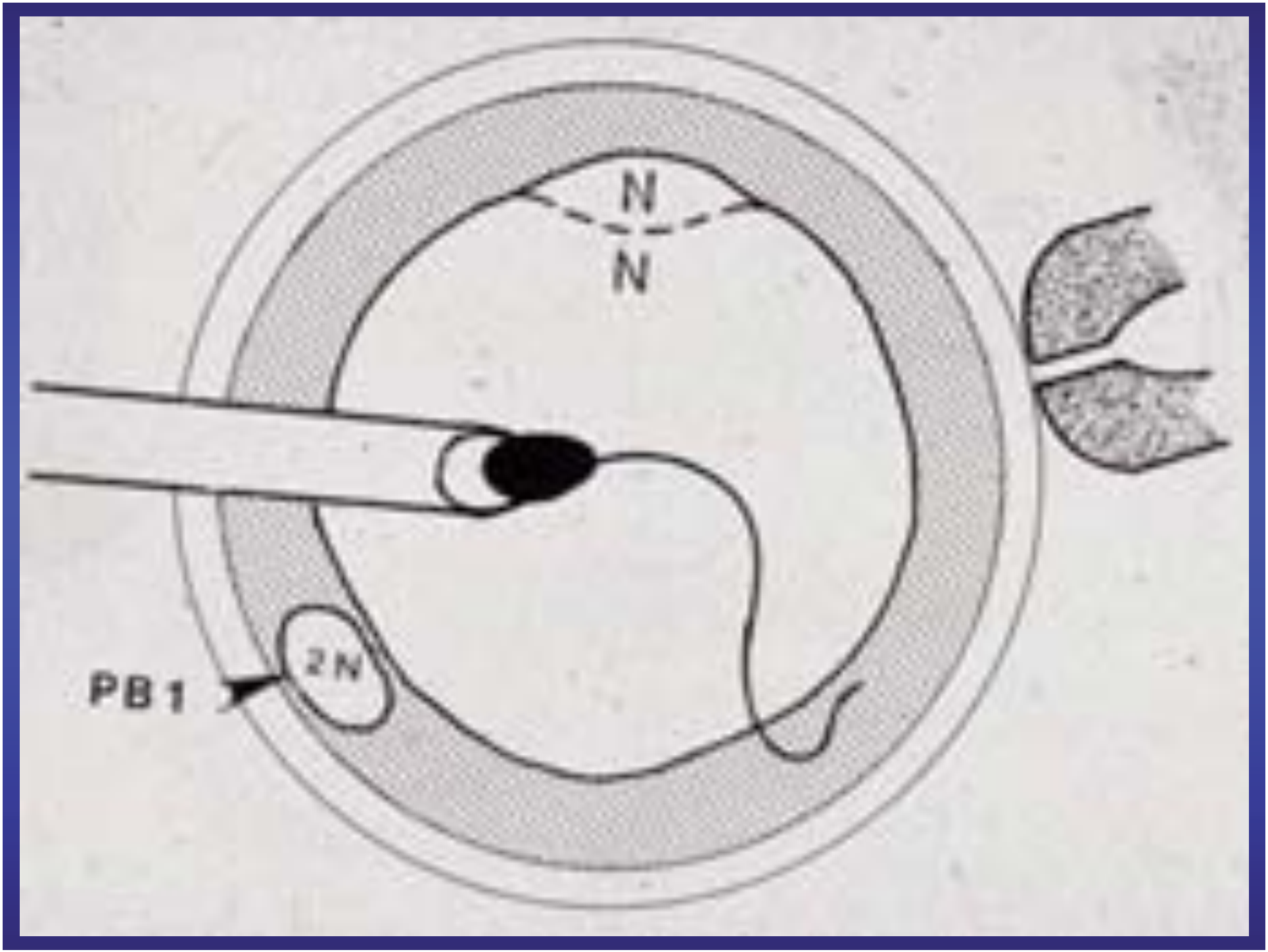
In Vitro Fertilization

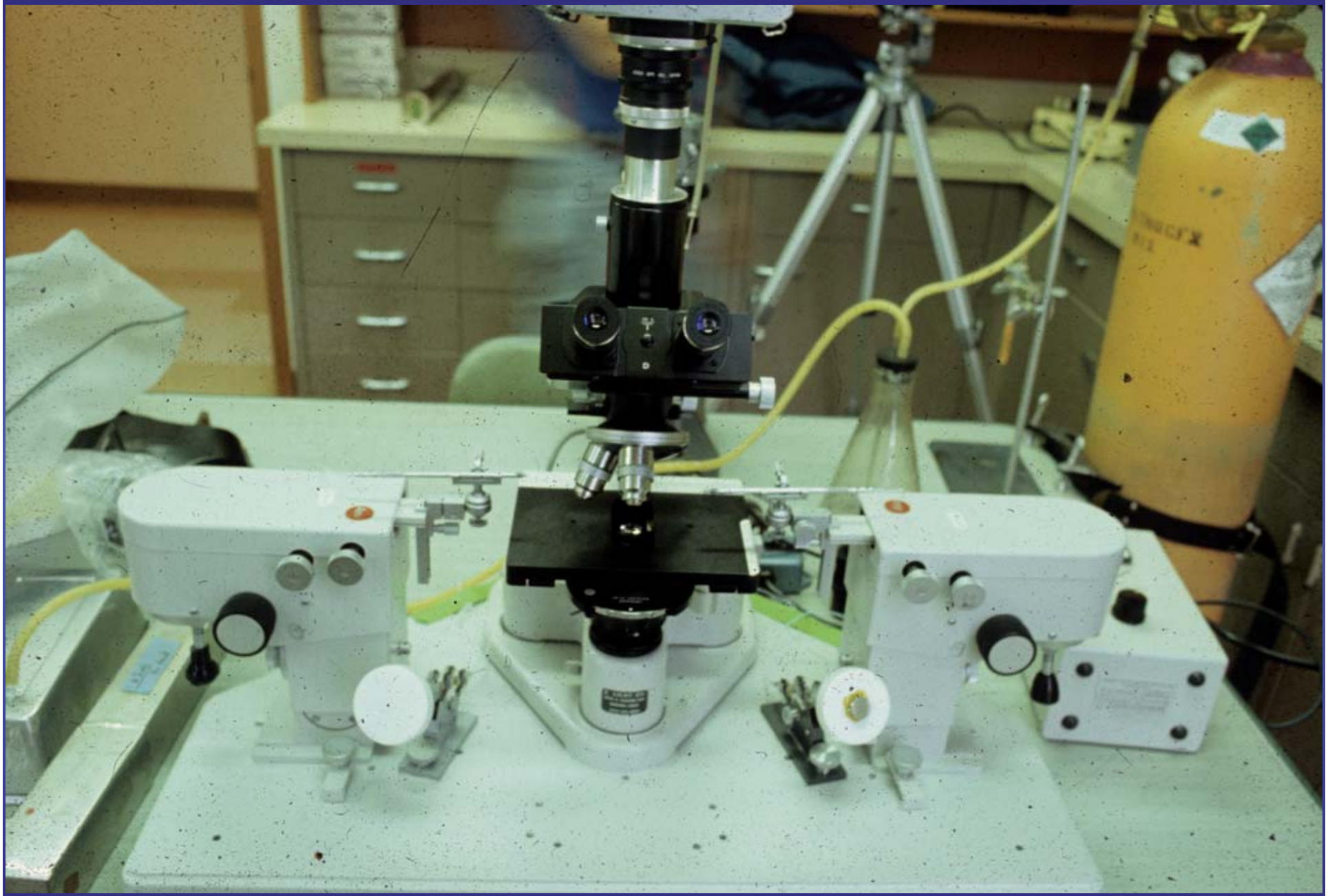
- 2010 Nobel prize
- Conventional
- Sperm injection

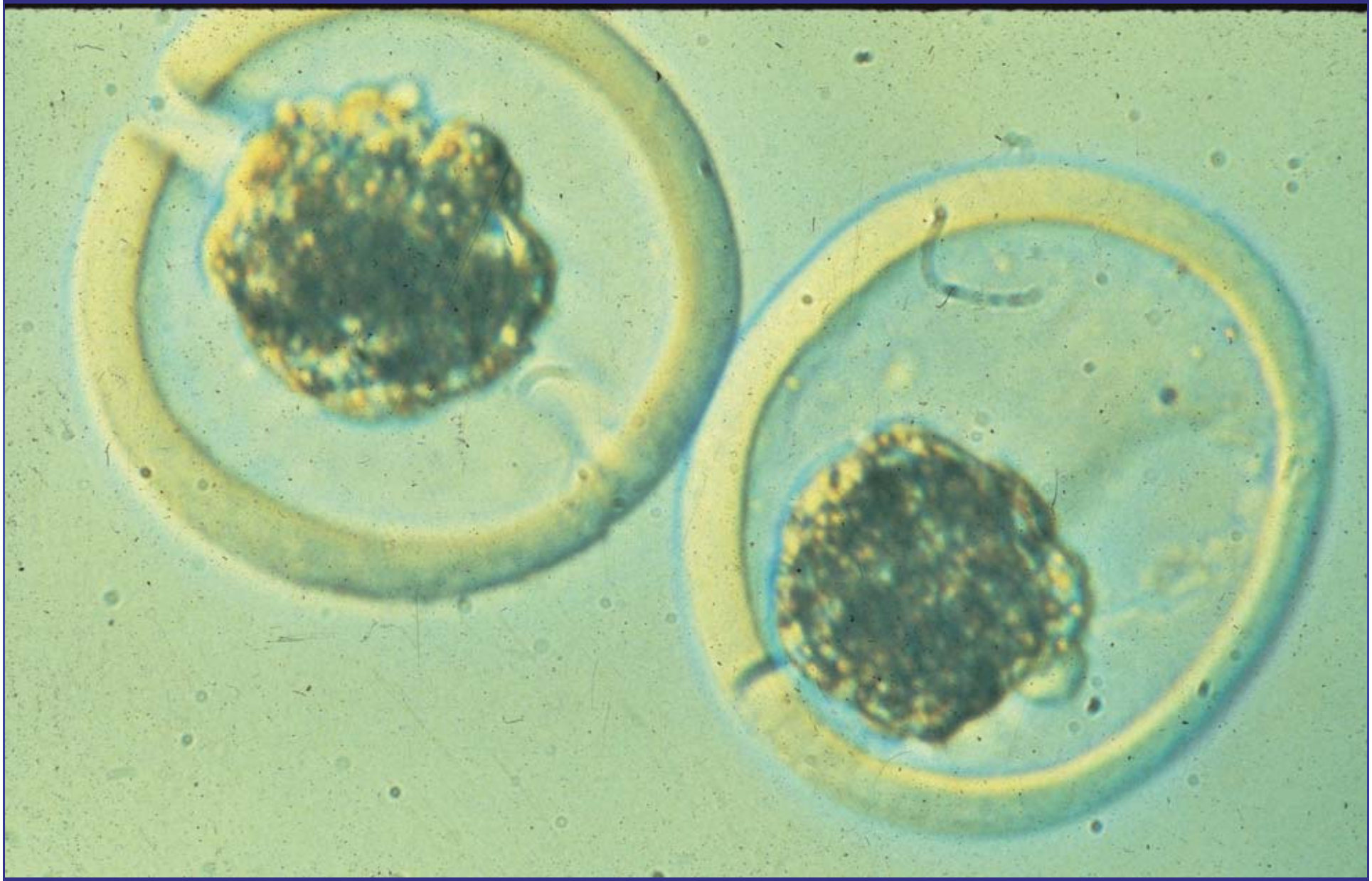

N
N
2N
PB1

Applications

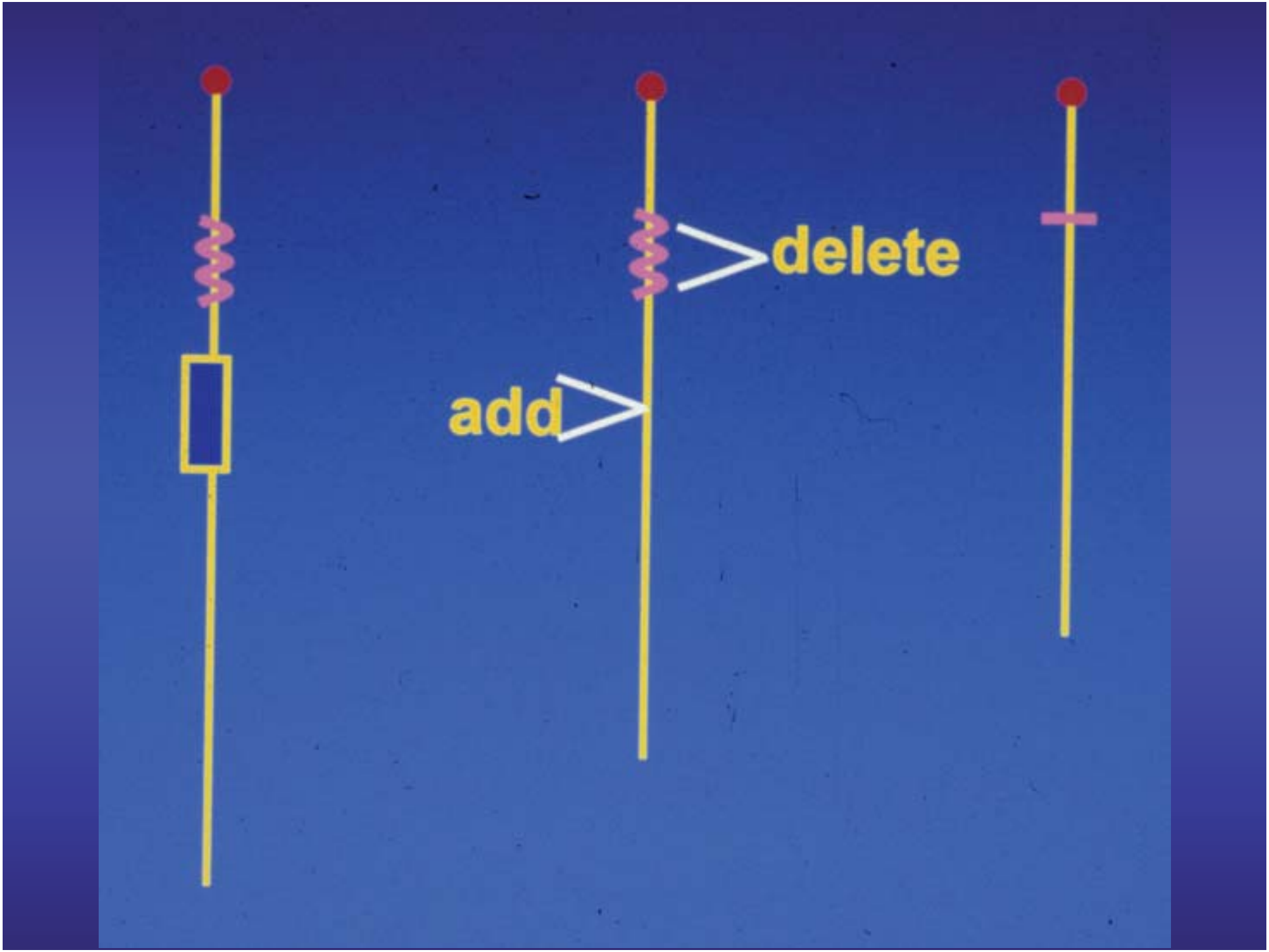
- Subfertile males
- Damaged sperm due to liquid nitrogen tank failure
- Freeze-dried sperm
  - Weeks at room temperature
  - Year at -20°C

Micromanipulation and Microsurgery






TRANSGENESIS
ADD GENES
DELETE GENES
CORRECT GENES


delete
add

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Transgenic Applications

- More efficient growth
- Polled (no horns)
- Resistance to disease
- Pharmaceuticals in milk

More Futuristic Ideas

- Transgenic bulls that only produce X (or Y) sperm
- Hibernation of beef cows


80C

Growth Genes on Y Chromosome

- Females remain smaller
- Extra growth expressed only after birth
- Sexed semen
  - Larger males
  - Smaller females

Offspring with 2 genetic fathers

Seidel 501
slide #119

Recreate Woolly Mammoth

- Frozen carcasses found in Siberia
- DNA best preserved in sperm
- Use oocytes from elephants
- Sperm are dead – use sperm injection
- 2X sperm = female
  1X + 1Y sperm = male
- Biological issues, e.g. imprinting
- Ethical issues

Scientific Ethics

- Fabrication of data
- Falsification of data
- Plagiarism
- Self correcting

Experimental Animals

- Treat humanely
  - Environment/facilities
  - Health and nutrition
- Minimize pain and stress
  - Trained personnel
  - Anesthetics and analgesics
- Good experimental design

Experiments

- Important or interesting?
- Appropriate use of resources?
- Unethical or dangerous?
- Doing nothing is an action
- Human applications?

Acknowledgements

- AVANTEA invitation
- Many mentors
- Scientific colleagues and students
- Technical and secretarial colleagues