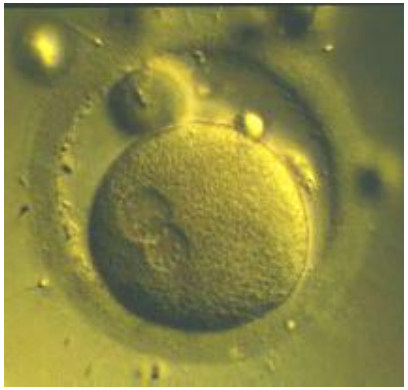
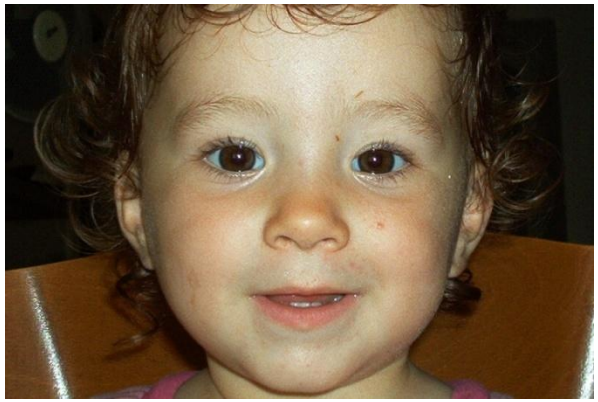
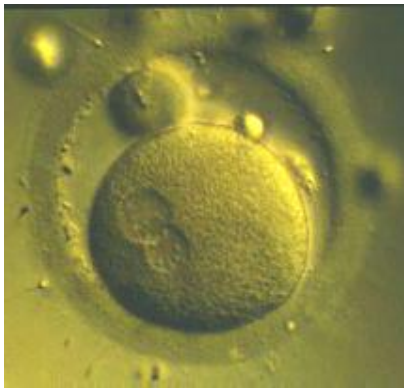
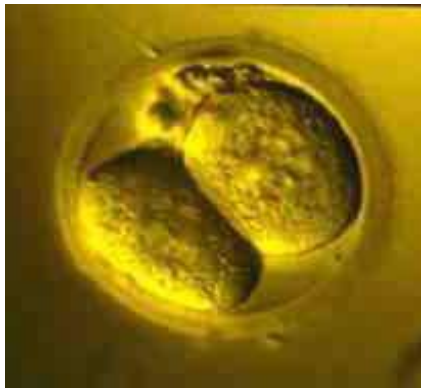
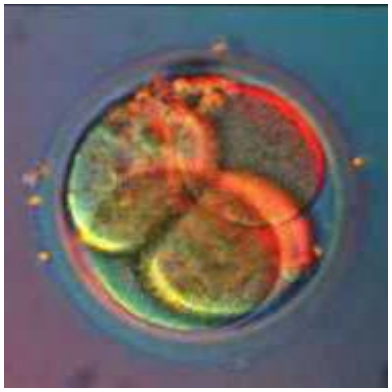
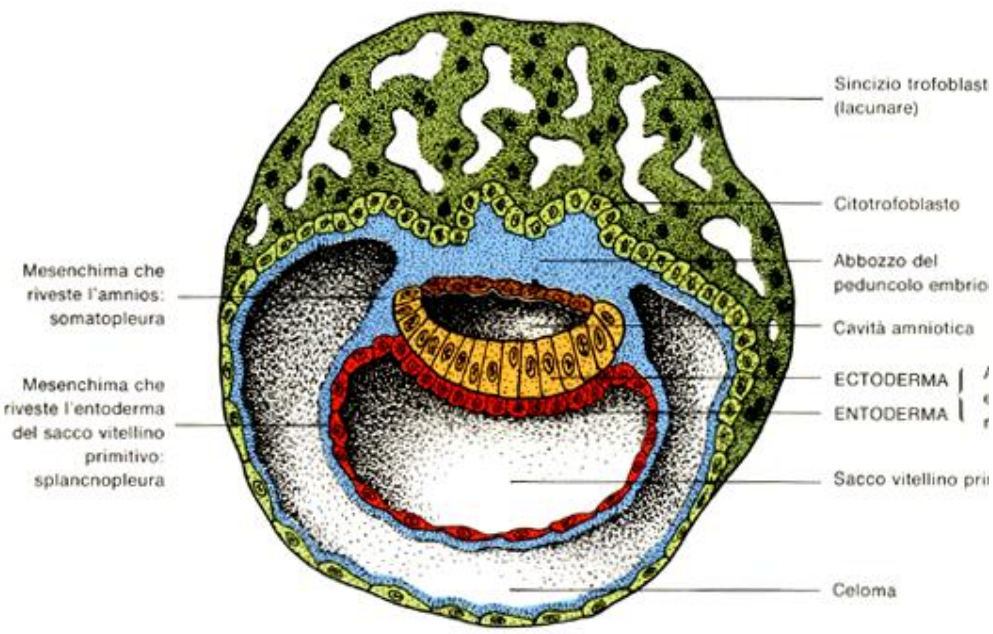
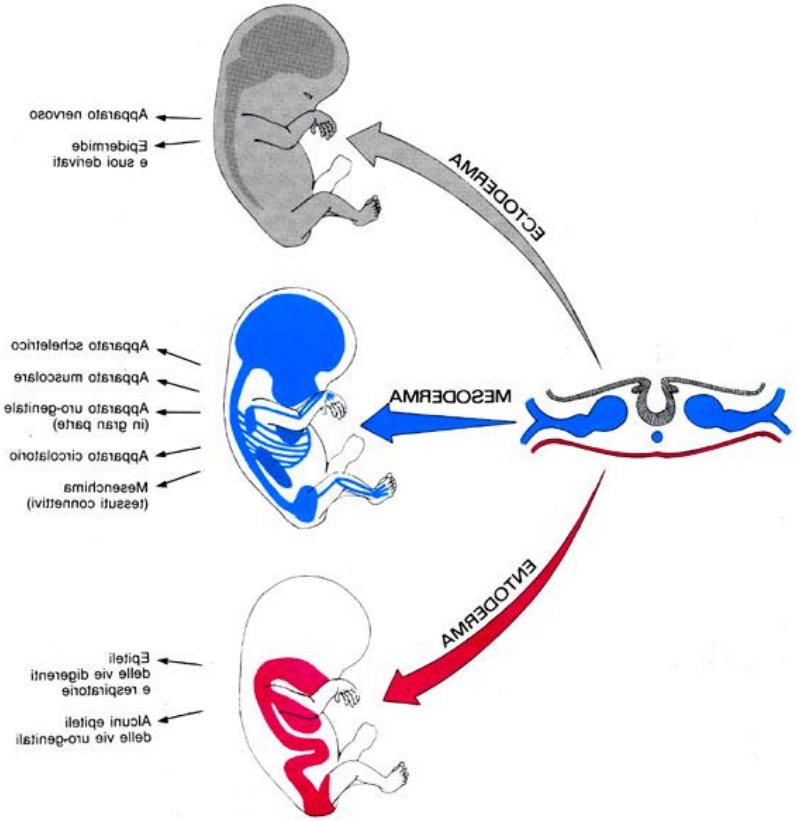




Durante lo sviluppo embrionale, si differenziano piu' di **200 tipi istologici** diversi per dare origine a miriadi di cellule specializzate.

Circa **30-40.000 geni** sono coinvolti nello specificare l'organismo umano



Sincizio trofoblast
(lacunare)
Citotrofoblasto
Abbozzo del
peduncolo embrio
Cavità amniotica
ECTODERMA
ENTODERMA
Sacco vitellino pri
Celoma
Mesenchima che
riveste l'amnios:
somatopleura
Mesenchima che
riveste l'entoderma
del sacco vitellino
primitivo:
splancnopleura


Apparato nervoso
Epidermide
e suoi derivati
ECTODERMA
Apparato scheletrico
Apparato muscolare
Apparato uro-genitale
(in gran parte)
Apparato circolatorio
Mesenchima
(tessuti connettivi)
MESODERMA
Epiteli
delle vie digerenti
e respiratorie
Alcuni epiteli
delle vie uro-genitali
ENTODERMA

WHAT ARE THE DIFFERENT KINDS OF STEM CELLS

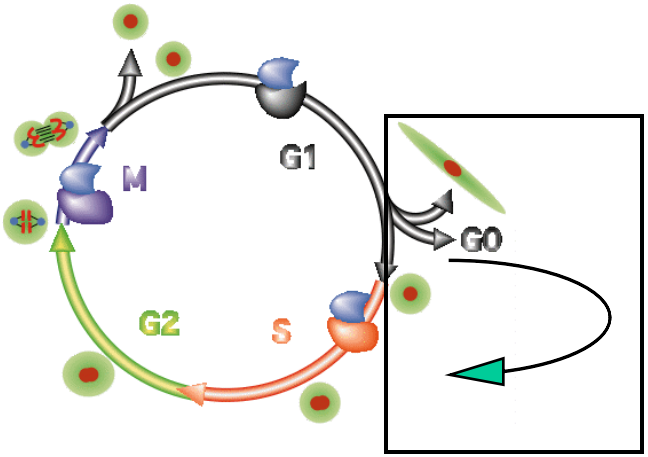
Unipotent (progenitor) stem cells are those whose terminally differentiated progeny consist of a single cell type only

Multipotent stem cells are those which can give rise to several terminally differentiated cell types constituting a specific tissue or organ.

Pluripotent stem cells are able to give rise to all different cell types *in vitro*. They cannot on their own form an embryo.

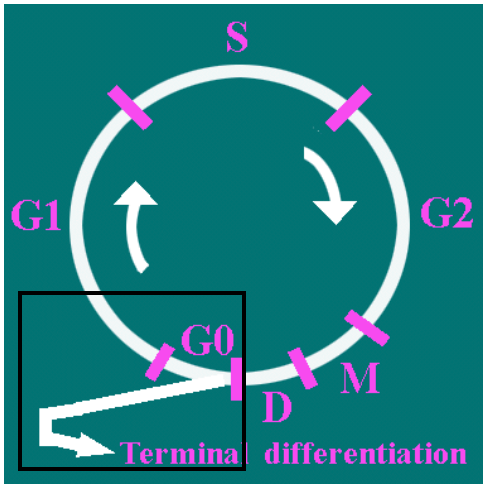
Cellule **ciclanti**

- Cellule ematopoietiche
- Spermatogoni



Cellule **transitoriamente quiescenti**

- osteoblasti (osso adulto)
- muscolari satelliti
- altre cellule staminali



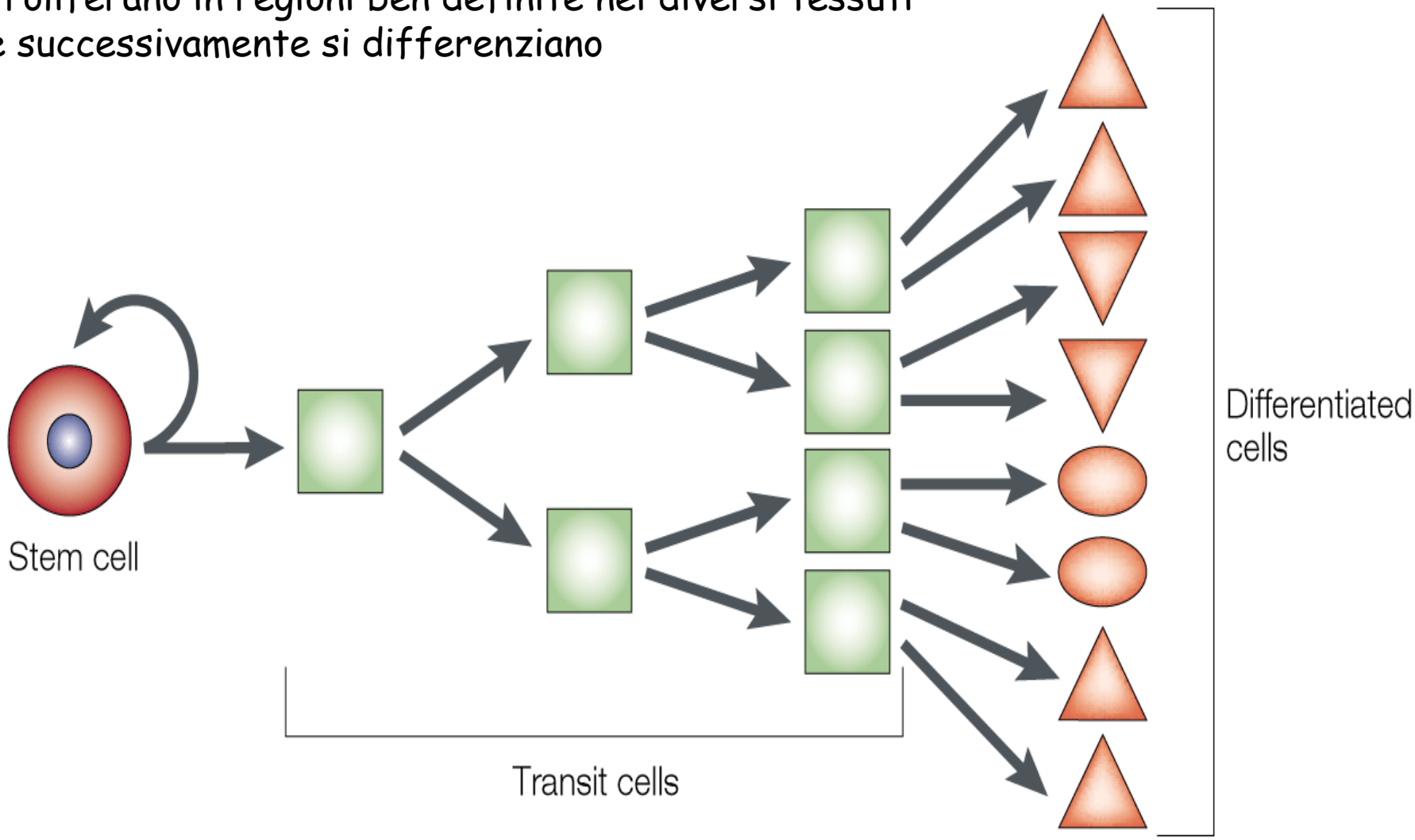
Cellule **irreversibilmente quiescent**

- cellule del Sertoli
- fibre muscolari
- granulociti
- neuroni

Nei tessuti soggetti a rinnovamento, sono presenti delle cellule che hanno mantenuto la capacita' proliferativa, a basso livello di differenziamento:

le cellule staminali

Proliferano in regioni ben definite nei diversi tessuti e successivamente si differenziano



In tessuti a rapido rinnovamento (es. ematopoietico, epiteli) l'attivita' proliferativa e' molto intensa

In tessuti piu' stabili (osso, muscolo) la cellula staminale si trova in una fase di quiescenza da cui fuoriesce a seguito di segnali proliferativi (legati a fenomeni di riparazione o rigenerazione post-traumatica).

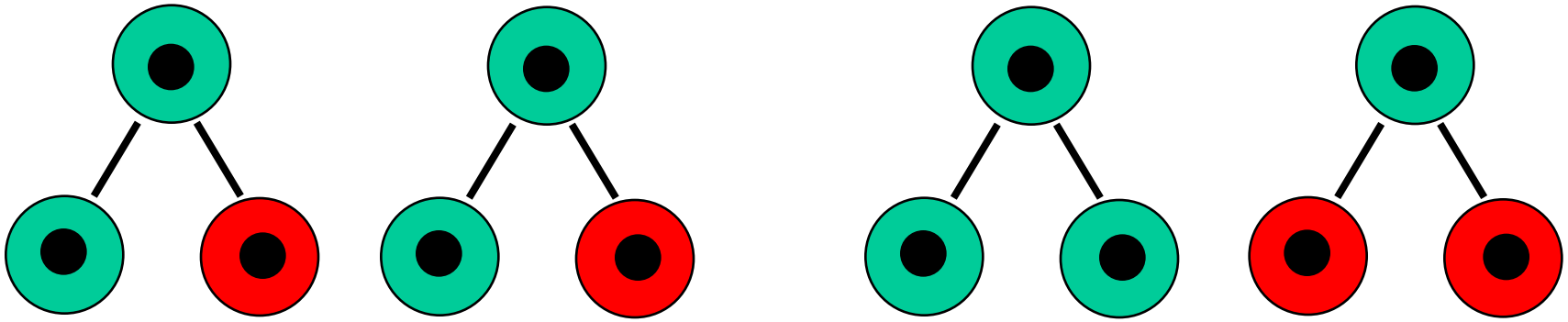
Replicazione di automantenimento della popolazione staminale

Replicazione differenziativa da' origine a cellule della popolazione differenziata

Negli invertrebrati: mitosi bivalente
Nei vertebrati: mitosi simmetrica

Mitosi asimmetrica o bivalente

Mitosi simmetrica



Due meccanismi per mantenere costante il numero di cellule staminali:

A) mitosi asimmetrica, una cellula si differenzia, l'altra rimane staminale,
B) mitosi simmetrica, alcune si differenziano, altre mantengono le caratteristiche della cellula madre, si dividono senza differenziarsi

Le funzioni della cellula staminale di:

- **Sopravvivenza per lunghi periodi nella fase quiescente**
- **Automantenimento**
- **Origine progenie differenziata**

Segnali derivanti dal microambiente che circonda la cellula staminale determinano quale strada intraprendere (automantenimento o differenziamento).

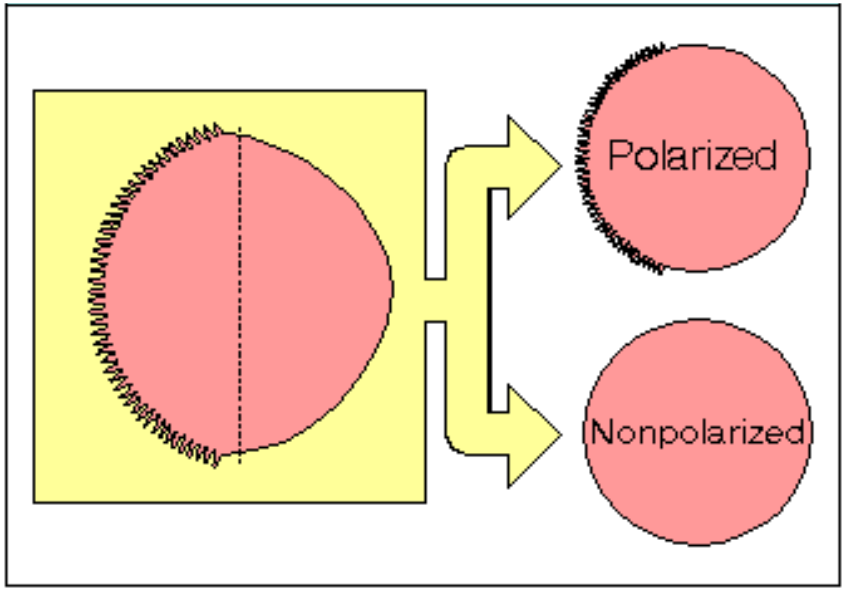
- Produzione di **fattori mitogeni** nell'area di mortalita' cellulare intorno alla staminale (es FGF, EGF)

- Rimozione di **fattori antimitogeni** prodotti dalle cellule sane circostanti (es. TGFß (inibisce staminale epitelio cripte intestinali e germinali primordiali)

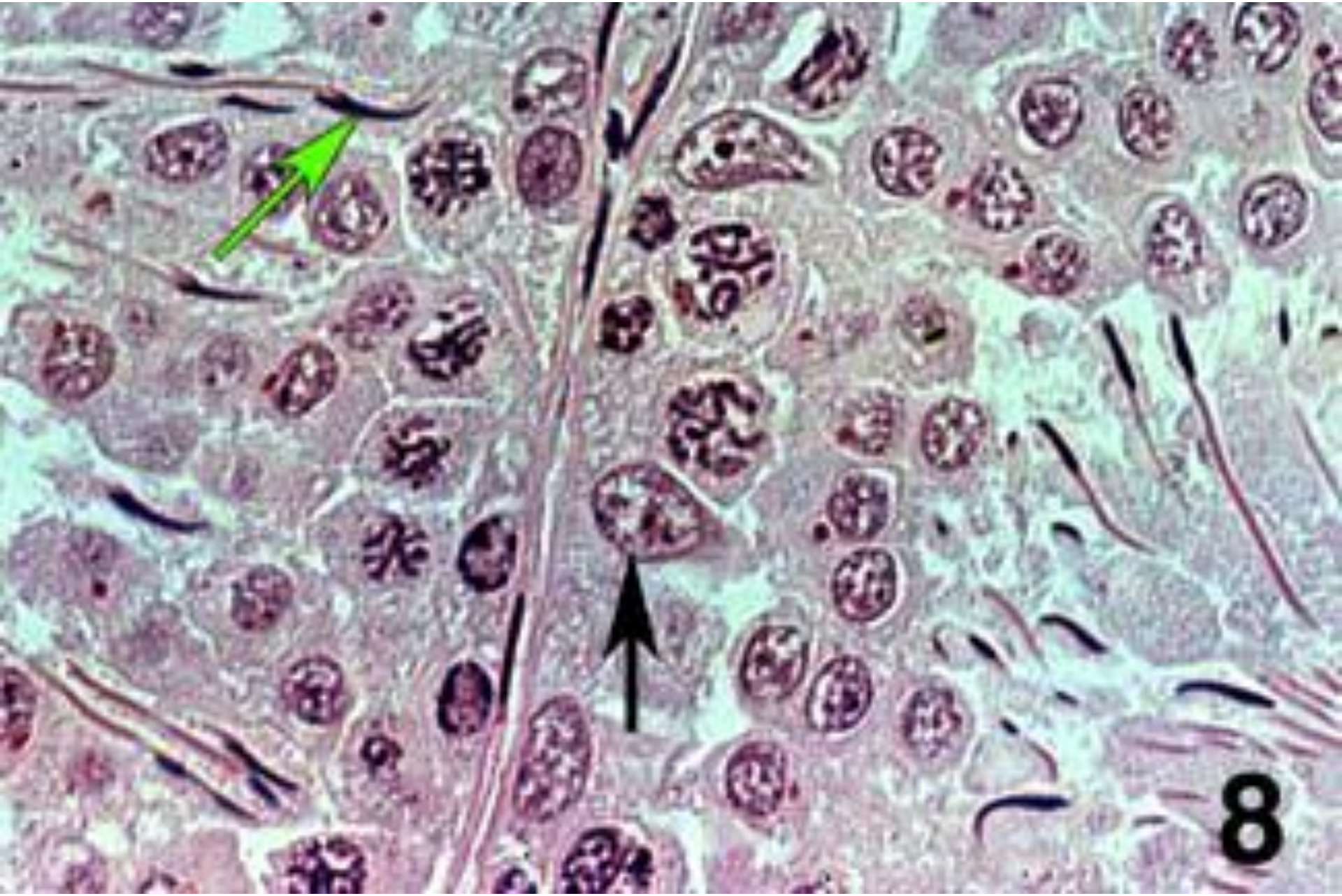
Meccanismi intracellulari autonomi

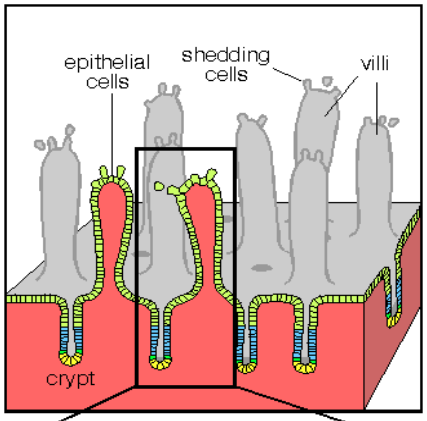
- Es. **distribuzione asimmetrica di recettori** sulla membrana della staminale

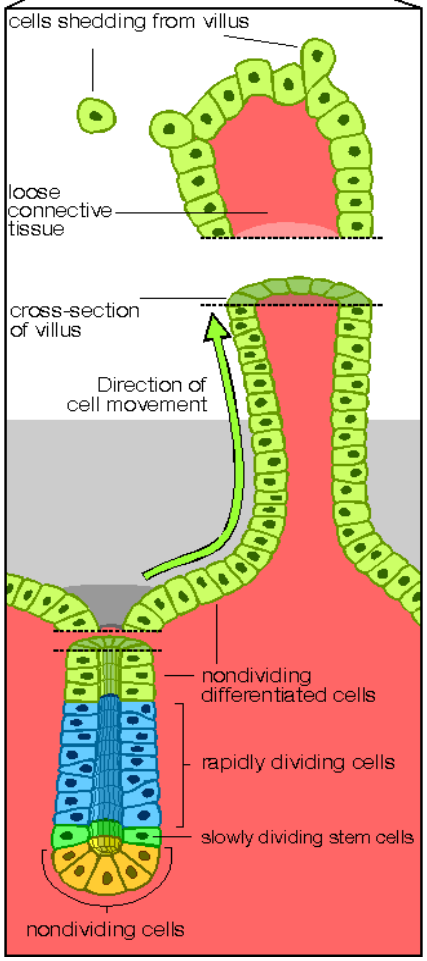
- Attivazione dell'enzima **telomerasi**

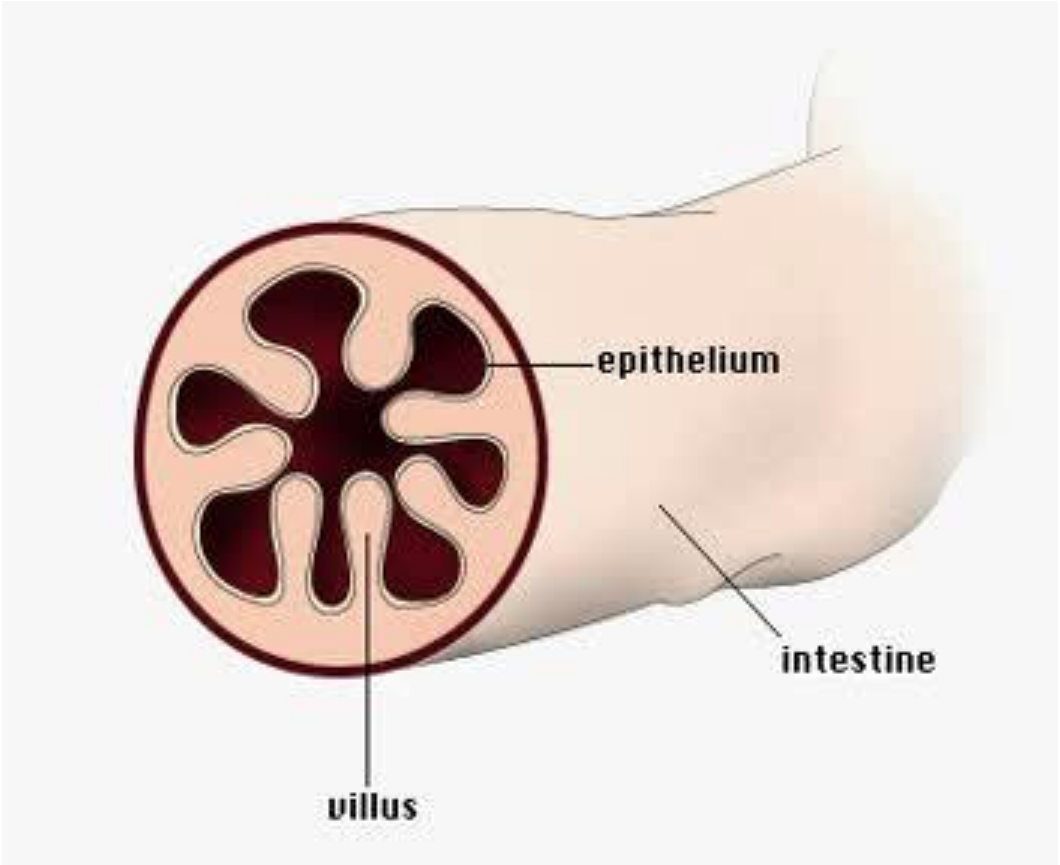


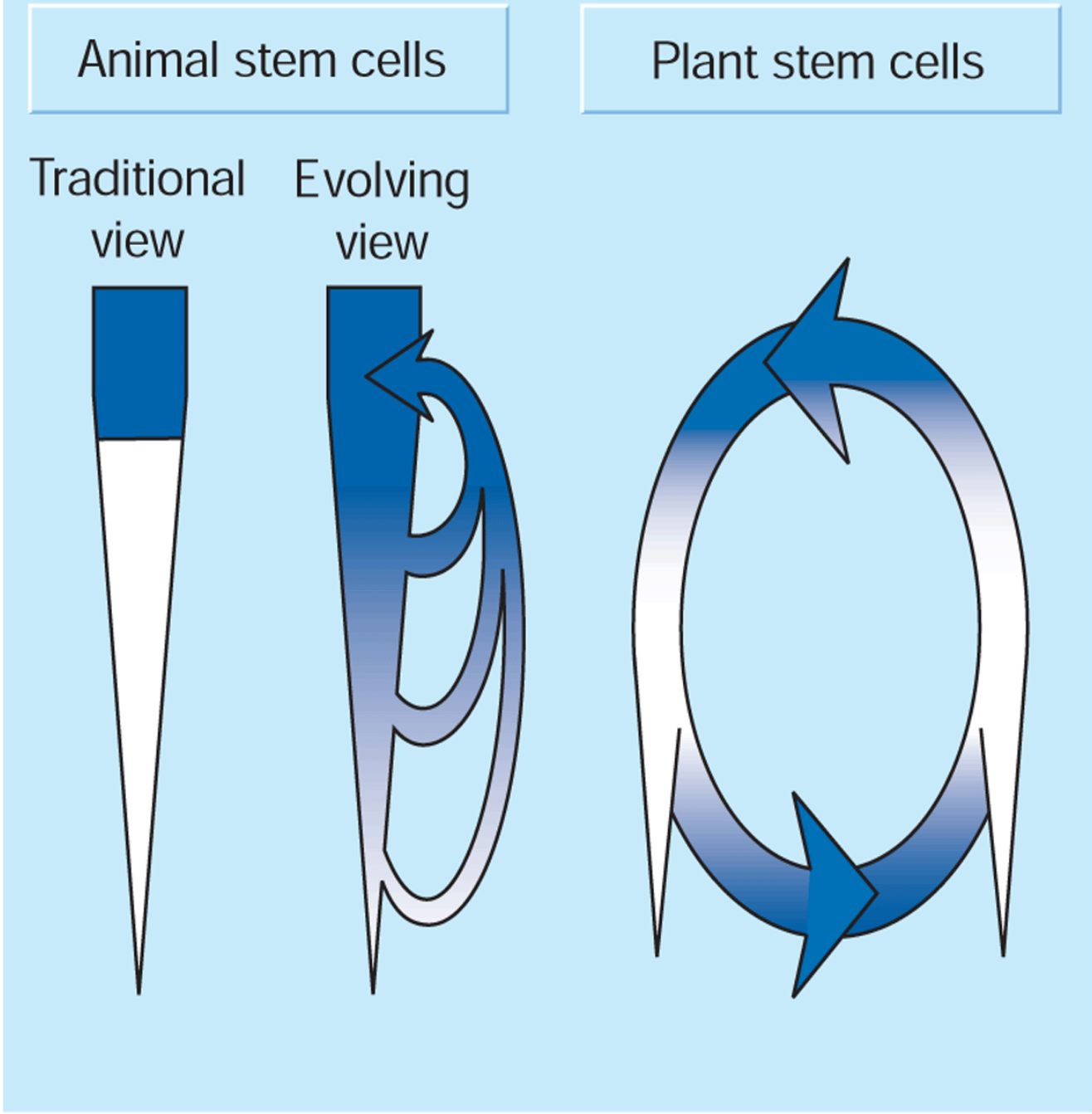
Le cellule staminali hanno una **localizzazione definita** nell'ambito del tessuto


8


epithelial cells
shedding cells
villi
crypt


cells shedding from villus
loose connective tissue
cross-section of villus
Direction of cell movement
nondividing differentiated cells
rapidly dividing cells
slowly dividing stem cells
nondividing cells


epithelium
intestine
villus


Animal stem cells
Plant stem cells
Traditional view
Evolving view

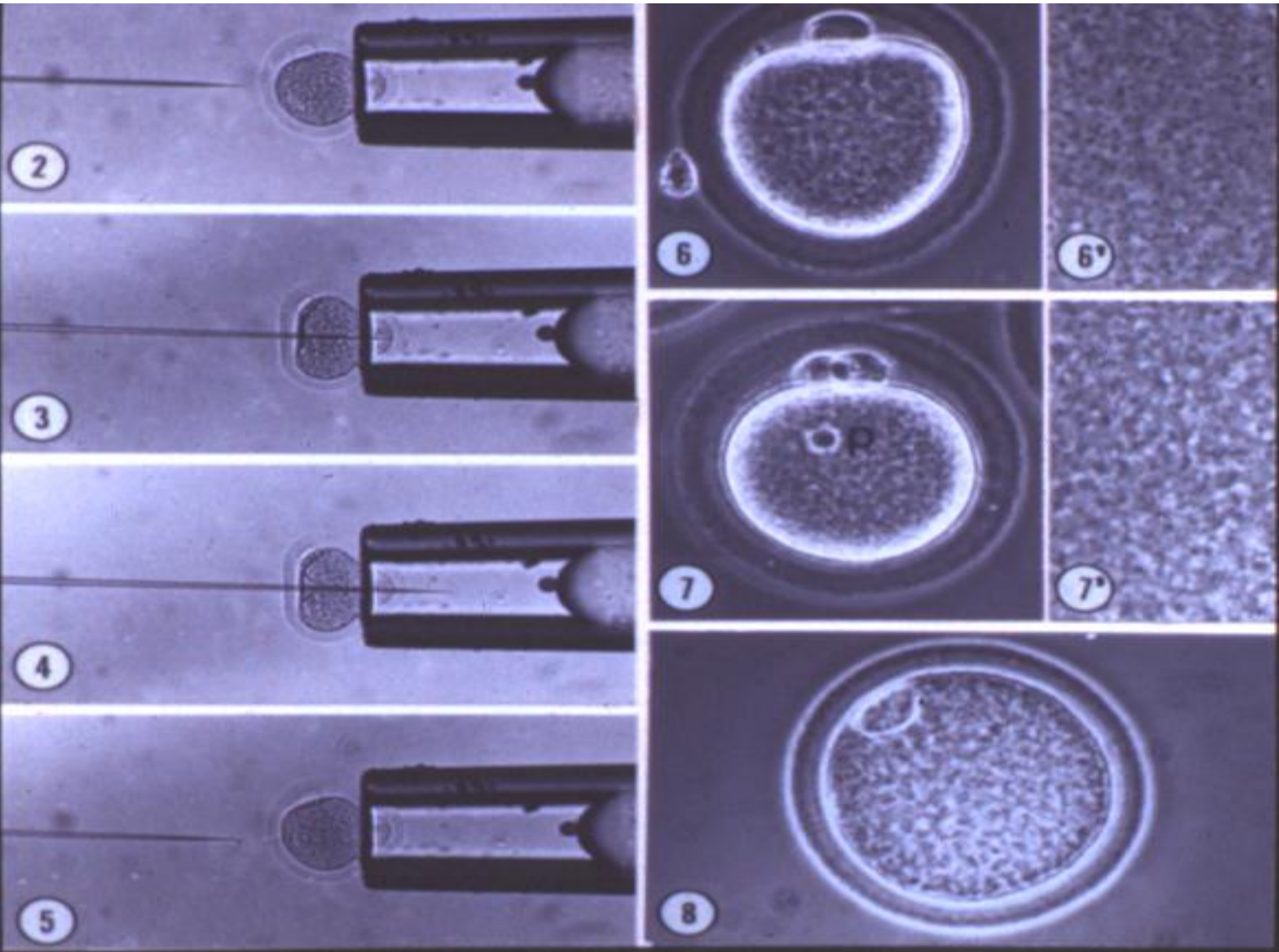
Hans Spemann

(1869 - 1941)

La differenziazione cellulare e' un processo terminale o il genoma di una cellula adulta puo' essere riprogrammato ?

1938 *Cloning is envisioned*

Hans Spemann proposed what he called a **"fantastical experiment."** He suggested taking the nucleus from a cell of a late-stage embryo, juvenile or adult and transplanting it into an egg. In other words, **cloning**.


2
3
4
5
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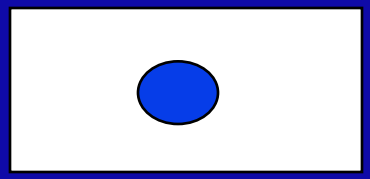
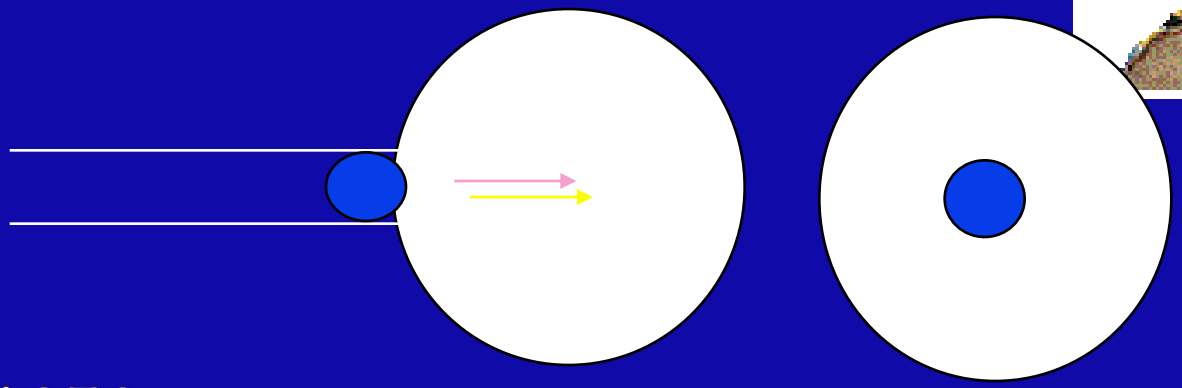
6'
7
P
7'
8

First cloning experiment with frogs

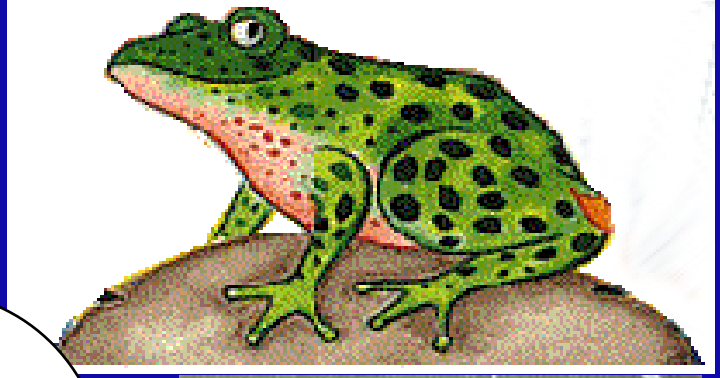


somatic cell



enucleated egg

embryo

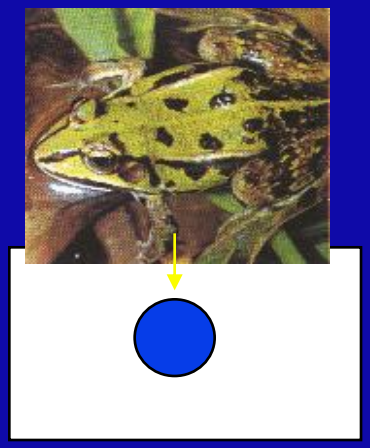


1952

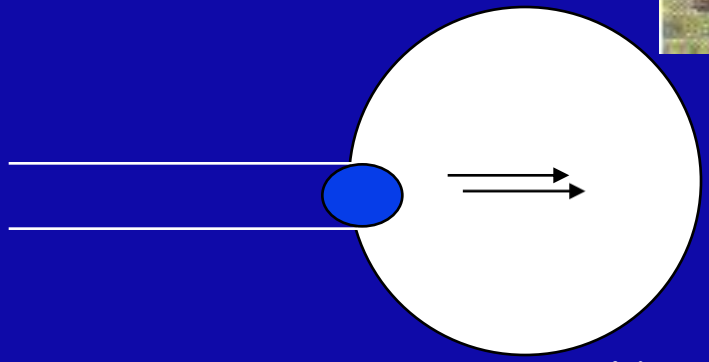
Robert Briggs and T.J. King used a pipette to suck the nucleus from the cell of an advanced frog embryo and added it to a frog egg. It did not developed to a tadpole.

1970

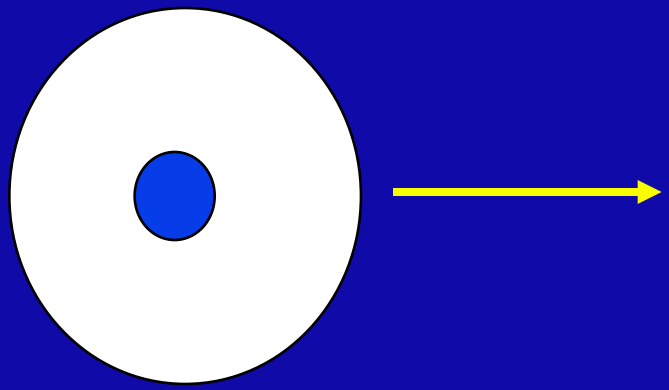
John Gurdon tried the same procedure



somatic cell



enucleated egg



embryo

2%

He showed that transplanted nuclei reverted to an embryonic state.

1981

Cloning of mice is claimed

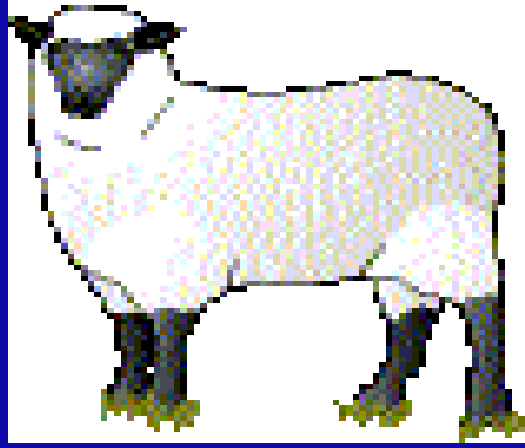
Karl Illmensee and Peter Hoppe reported that they produced normal mice from mouse embryo cells.

Nobody was able to repeat these experiments

Cell 23, 9-18 (1981)

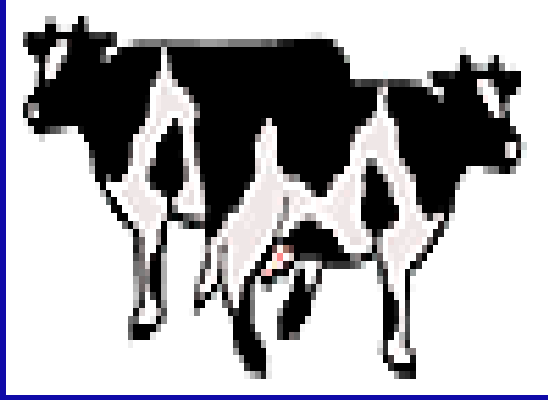
1984

First embryo cloning with sheep is reported



Steen Willadsen reported that he cloned a live lamb from immature sheep embryo cells. Others later replicate his experiment using a variety of animals, including cattle, pigs, goats, rabbits and rhesus monkeys.

1994



First cloning of more advanced embryo cell

Neal First cloned calves from embryos that have grown to at least 120 cells

1996

Groundwork laid for cloning of adult sheep

Ian Wilmut and Keith Campbell repeated First's experiment with sheep, but put embryo cells into a resting state before transferring their nuclei to sheep eggs. The eggs developed into normal embryos and then into lambs

Nature 380, 64-66 (1996)

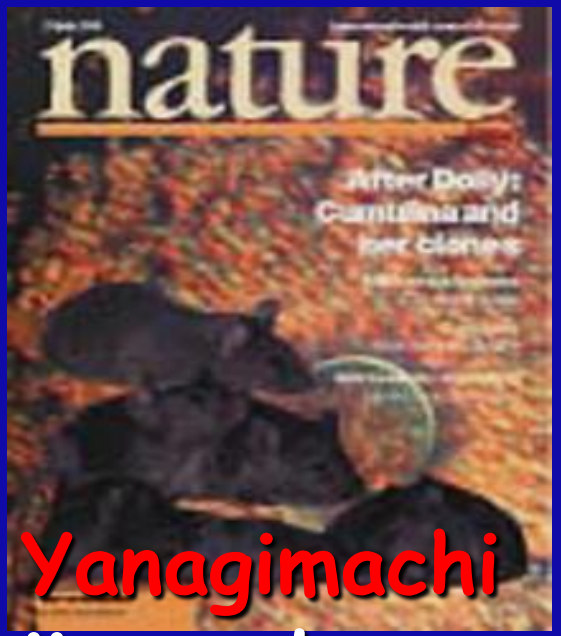
February 1997

Adult sheep is cloned

Ian Wilmut and Keith Campbell reported that they had cloned a 6-year-old adult sheep from an udder cell

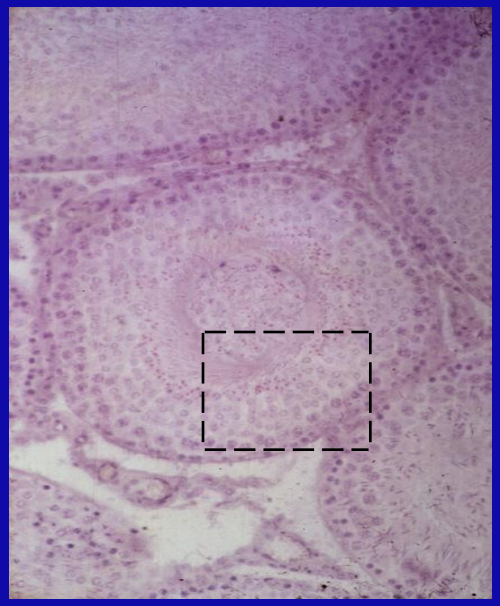
Cloning mice

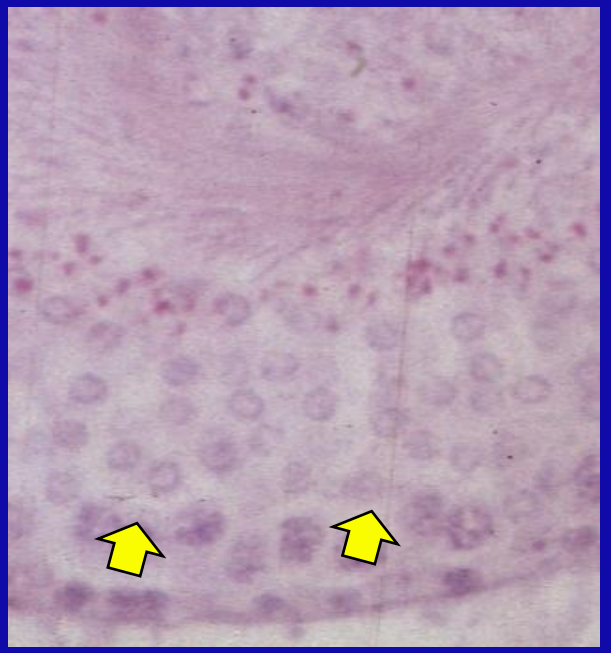
July 1998



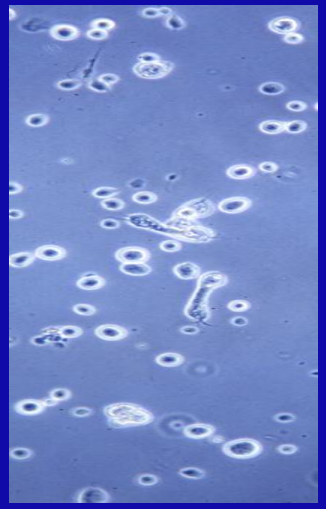
Nature 394, 369-374 (1998)

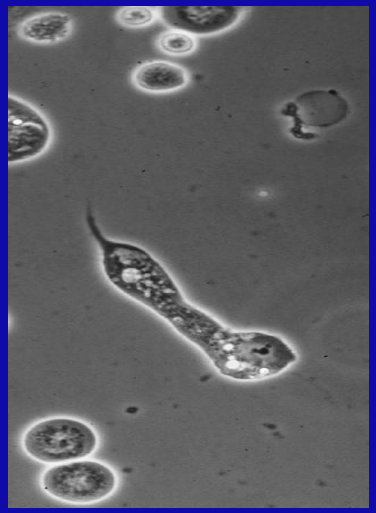
Nature press release: **Ryuzo Yanagimachi** of the Unversity of Hawaii and an international team from the **US**, **Japan**, **Italy** and the **United Kingdom** report the birth of no fewer than 22 healthy and fertile mice which have been cloned from nuclei of adult ovarian cumulus cells

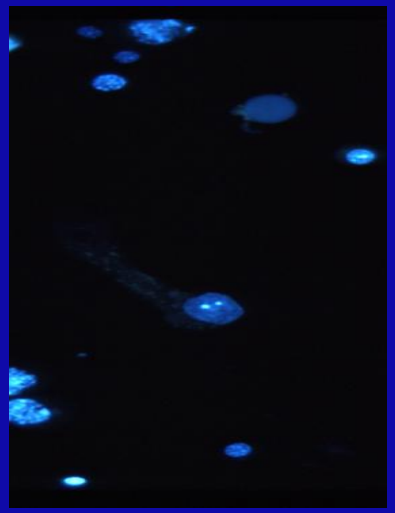




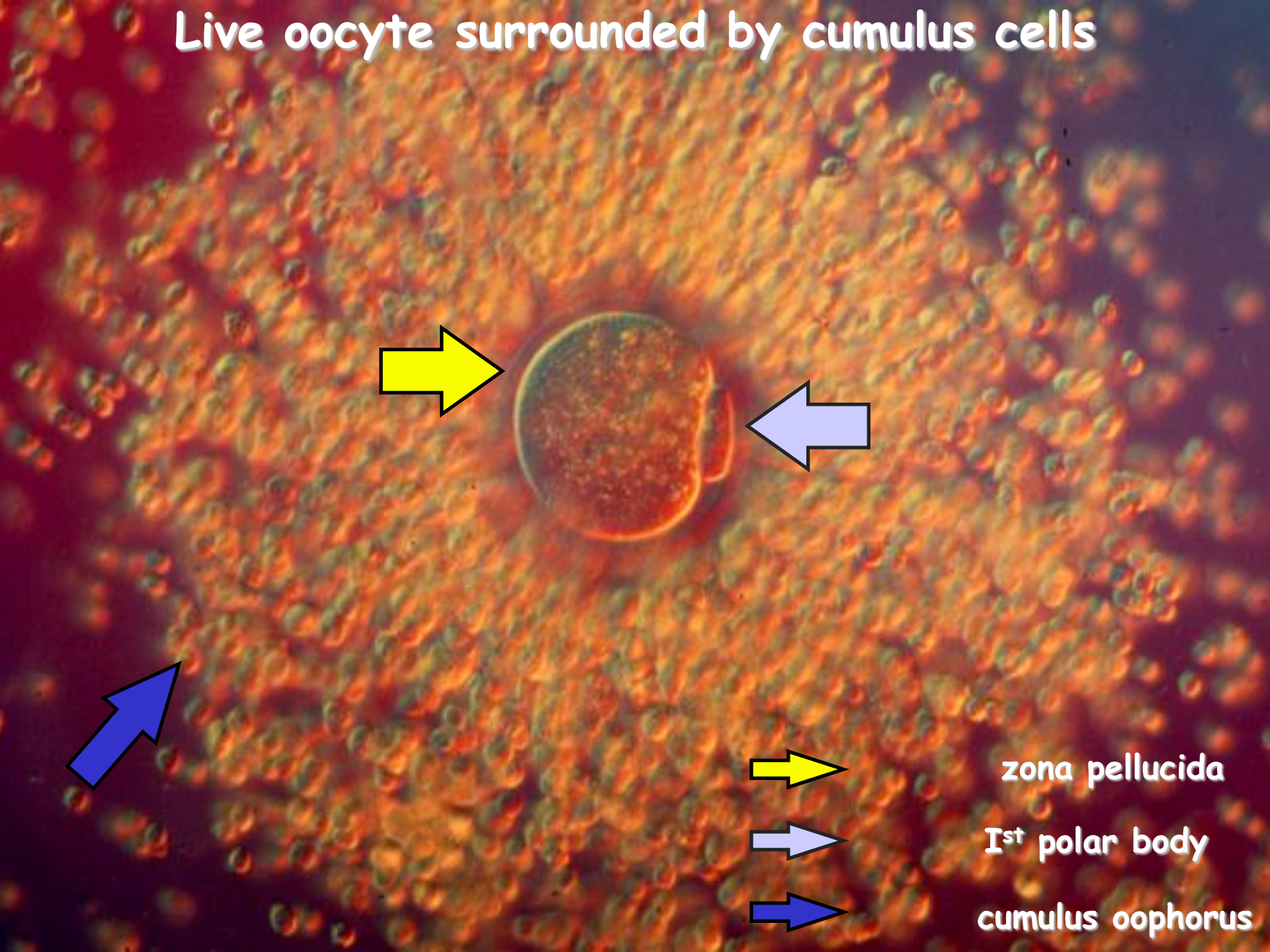
Sertoli cells



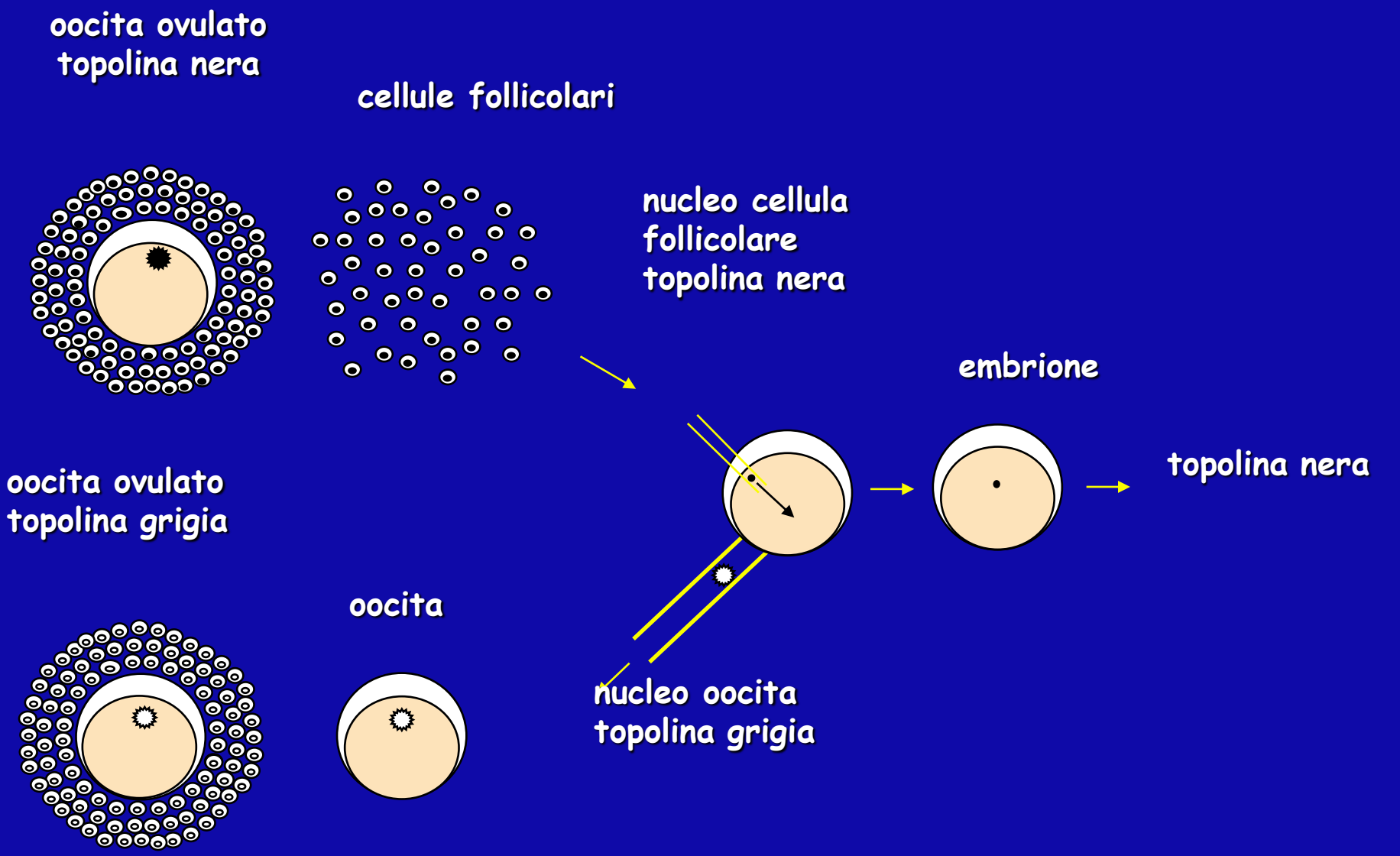


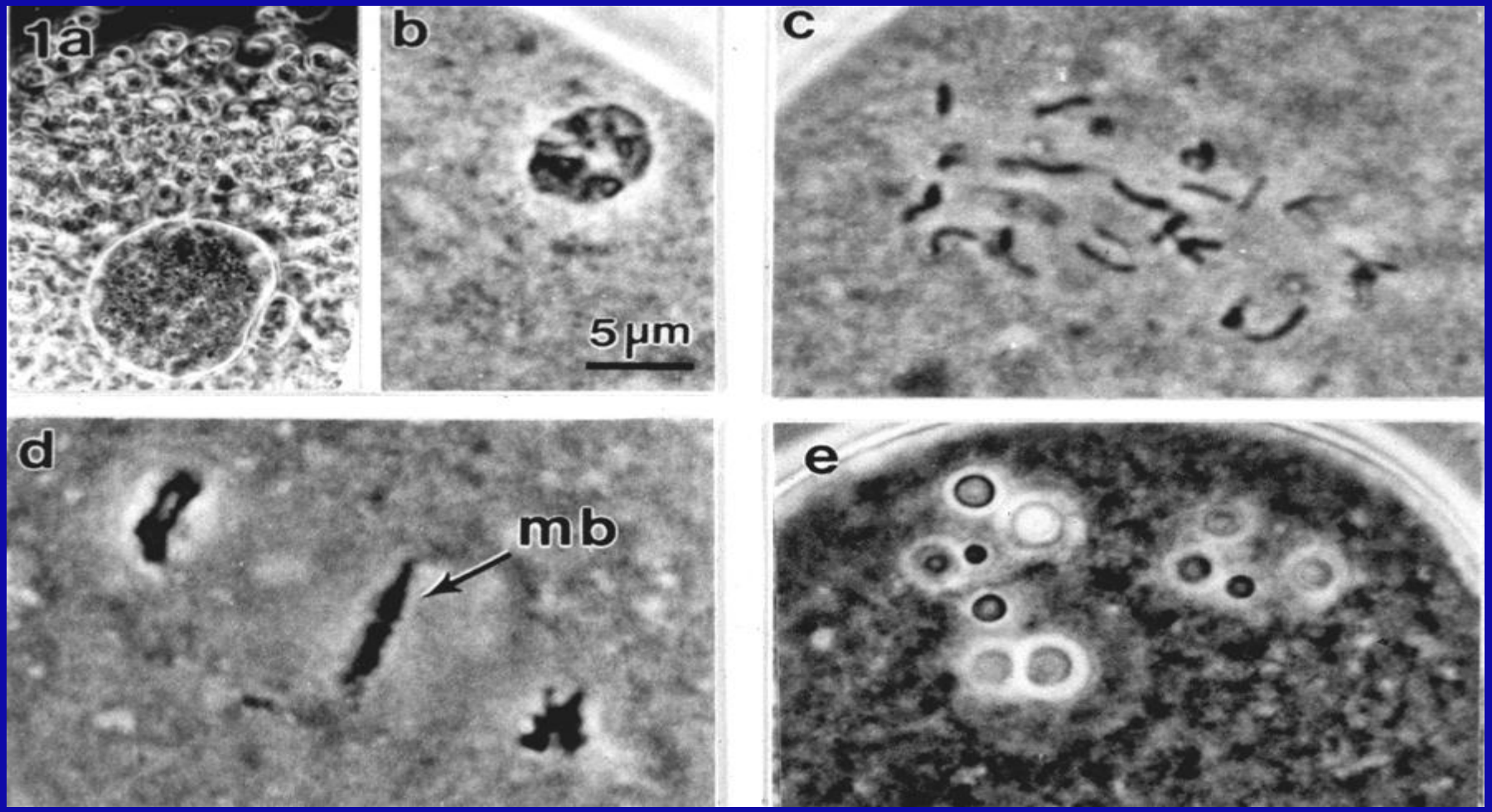


Live oocyte surrounded by cumulus cells

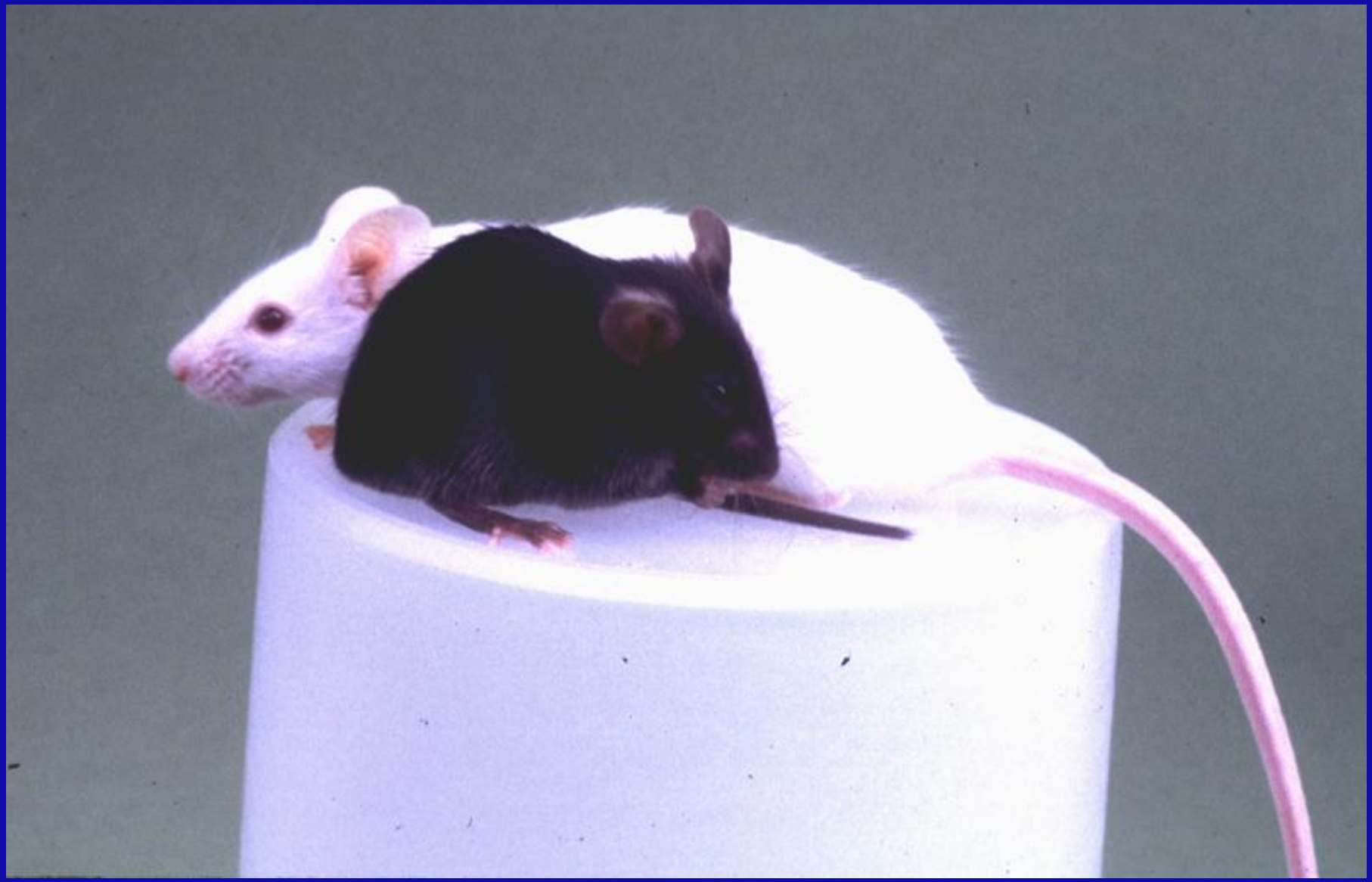


Trasferimento nucleare





In vitro development of enucleated oocytes following injection of **cumulus cell nuclei**



Cumulina and her foster mother

Il genoma di una cellula adulta di mammifero puo' essere riprogrammato all'interno del citoplasma di un oocita


IMMIGRATION
EUROPE UNDER SIEGE
HUMAN CLONING IS CLOSER THAN YOU THINK
For couples who can't have a child—
or who have lost one—the unthinkable
may soon be possible. Here are the perils


Scienze +
di della scienza
L. 15.000
MATERIA E ANTIMATERIA • AMMASSI DI GALASSI
LE SCIENZE
edizione italiana di SCIENTIFIC AMERICAN
L'EVOLUZIONE DELLA STATURA
FARMACI e CLONAZIONE
quale FUTURO?

Raffaello Cortina Editore

Gina Kolata

Cloni

Da Dolly all'uomo?

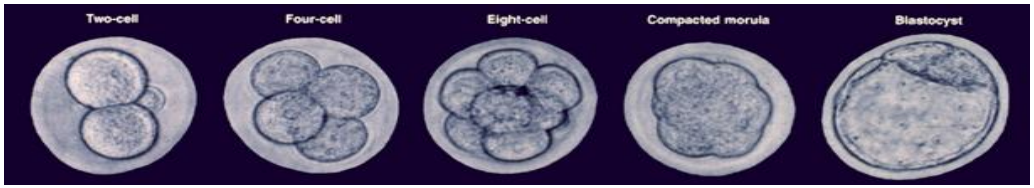
Scienza e Idee

Collana diretta da Giulio Giorello

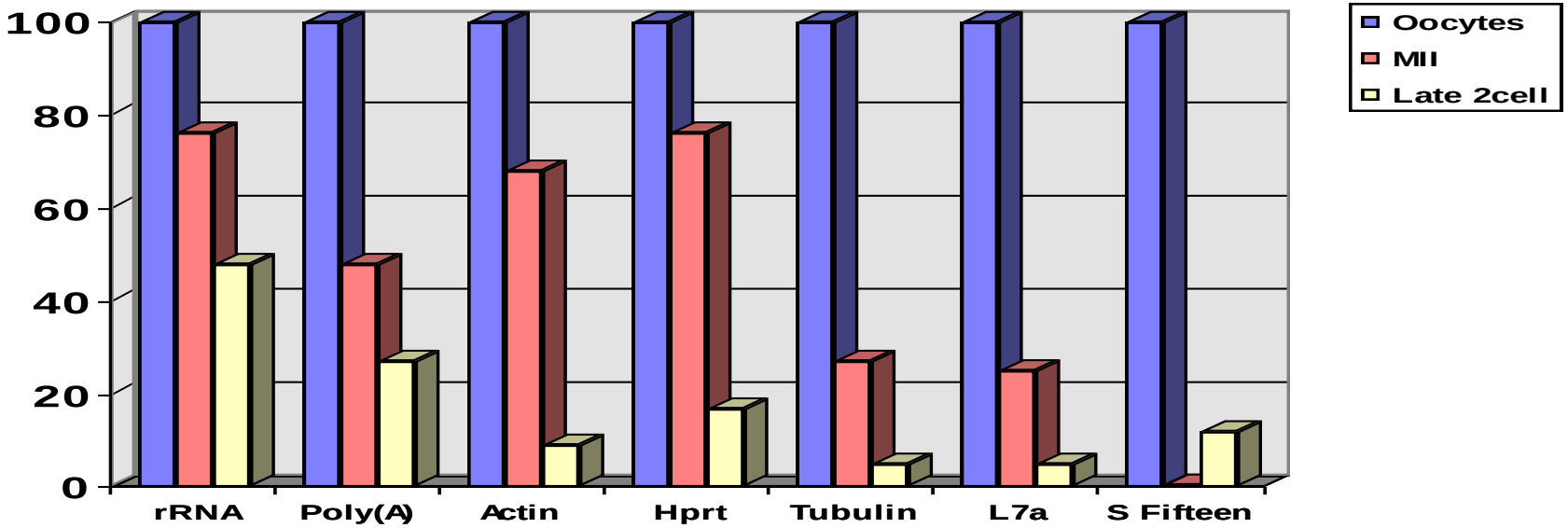
a) Timing of the process of genetic reprogramming

b) Timing of gene expression

c) Identify those ooplasmic determinants capable of erasing the genetic information of the somatic genome and able to guide the next stages of genomic reprogramming and development



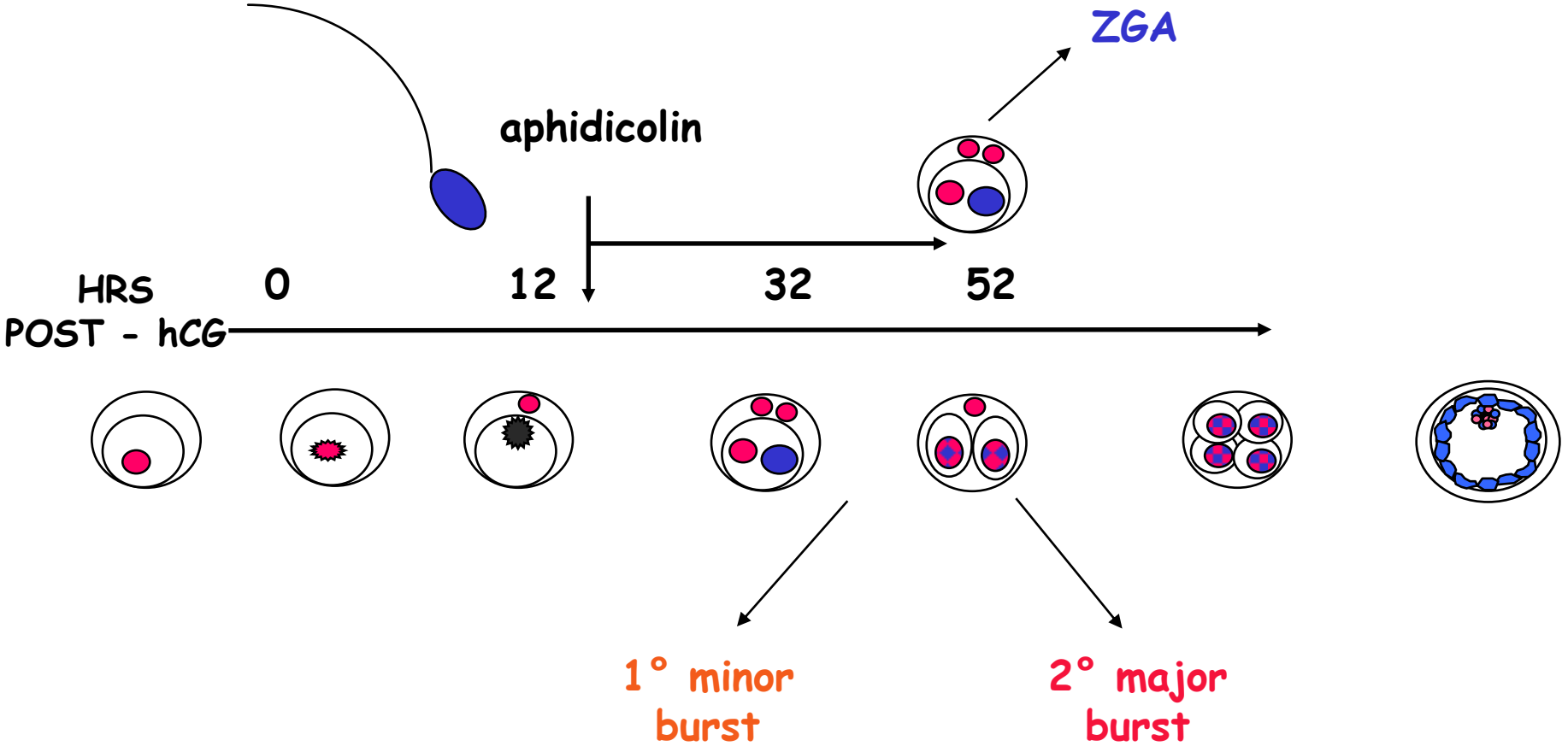
mRNA degradation



Successivamente alla ovulazione e alla fecondazione, gli RNA presenti nell'ooplasma vengono degradati. Ad un certo punto dello sviluppo preimpianto ha inizio l'espressione dei geni embrionali

ZYGOTIC GENOME ACTIVATION

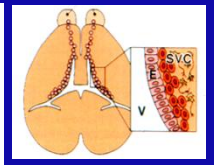
Nel topolino vengono espresse 40 nuove proteine nell'embrione a 2-cellule. I geni che codificano per queste proteine vengono espressi in due ondate (bursts)

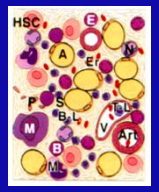


ADULTO

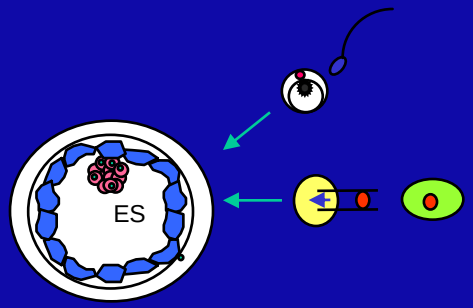
CORDONE OMBELICALE

EMBRIONE PREIMPIANTO

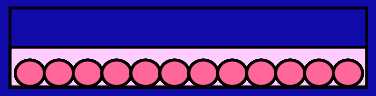




EPITELIALE INTESTINALE NEURALE EMATOPOIETICHE

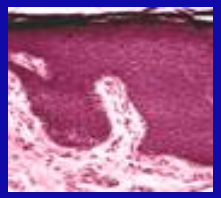


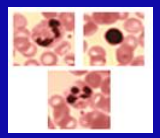
Isolamento

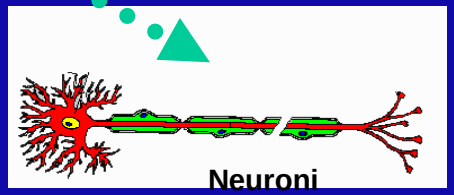


Coltura

Differenziamento





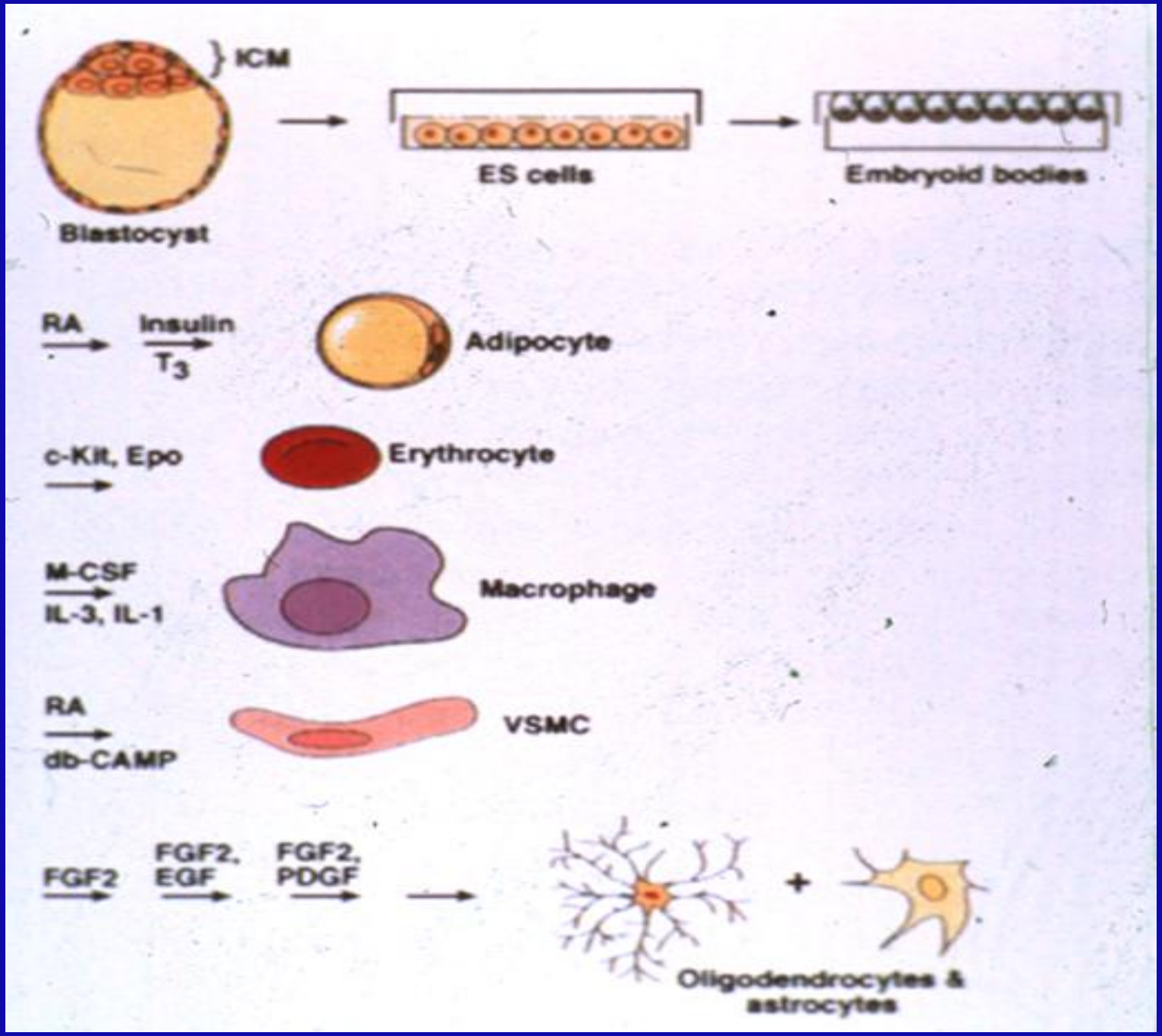


Epidermide

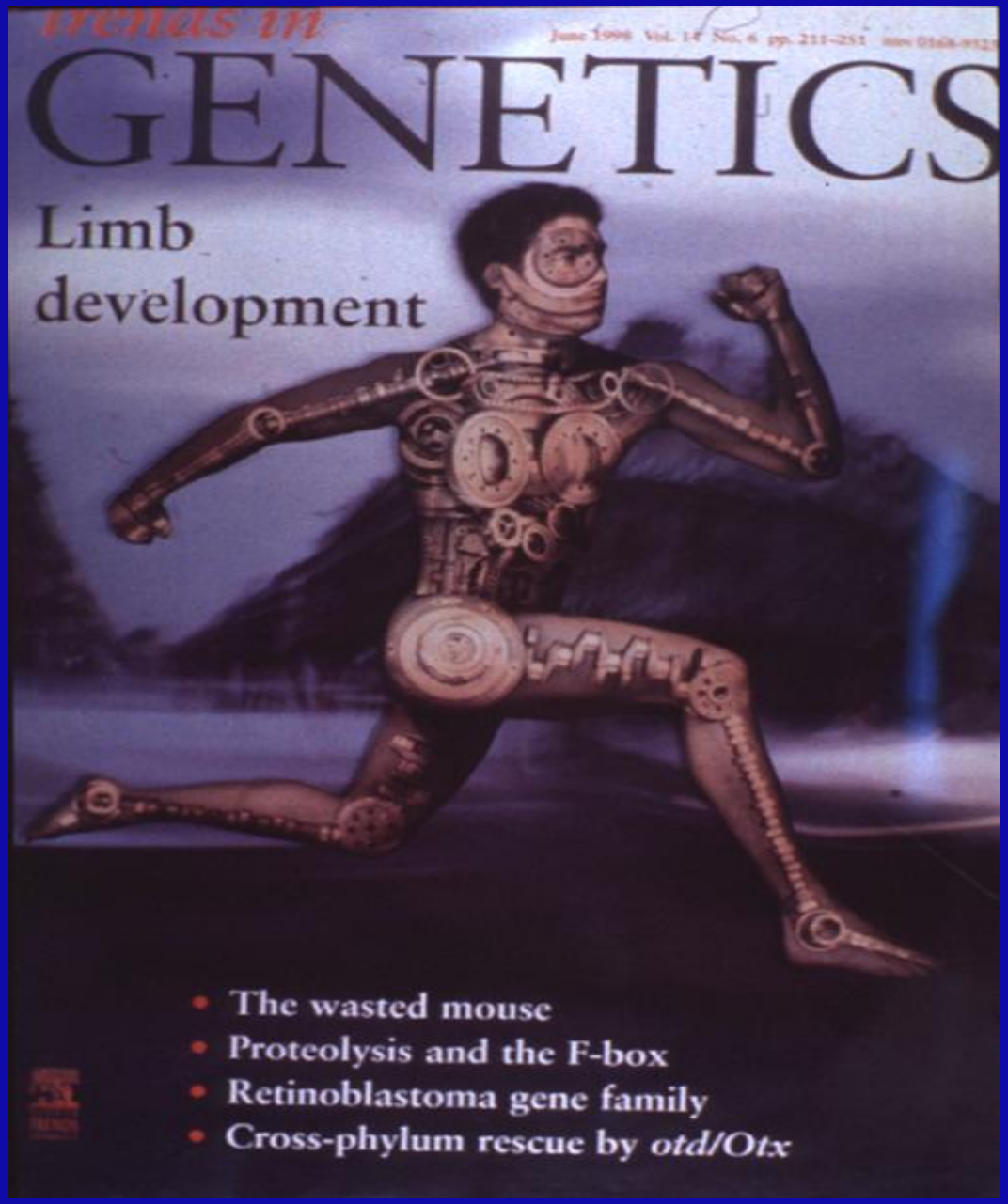
Cellule del sangue

Fibre muscolari

Terapia


ICM
Blastocyst
ES cells
Embryoid bodies
RA
Insulin
T_3
Adipocyte
c-Kit, Epo
Erythrocyte
M-CSF
IL-3, IL-1
Macrophage
RA
db-CAMP
VSMC
FGF2
FGF2, EGF
FGF2, PDGF
+
Oligodendrocytes & astrocytes


Science
17 December 1999
Vol. 286 No. 5448
Pages 2221–2416 $8
Breakthrough of the Year
Stem Cells Show Their Potential
AMERICAN ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE

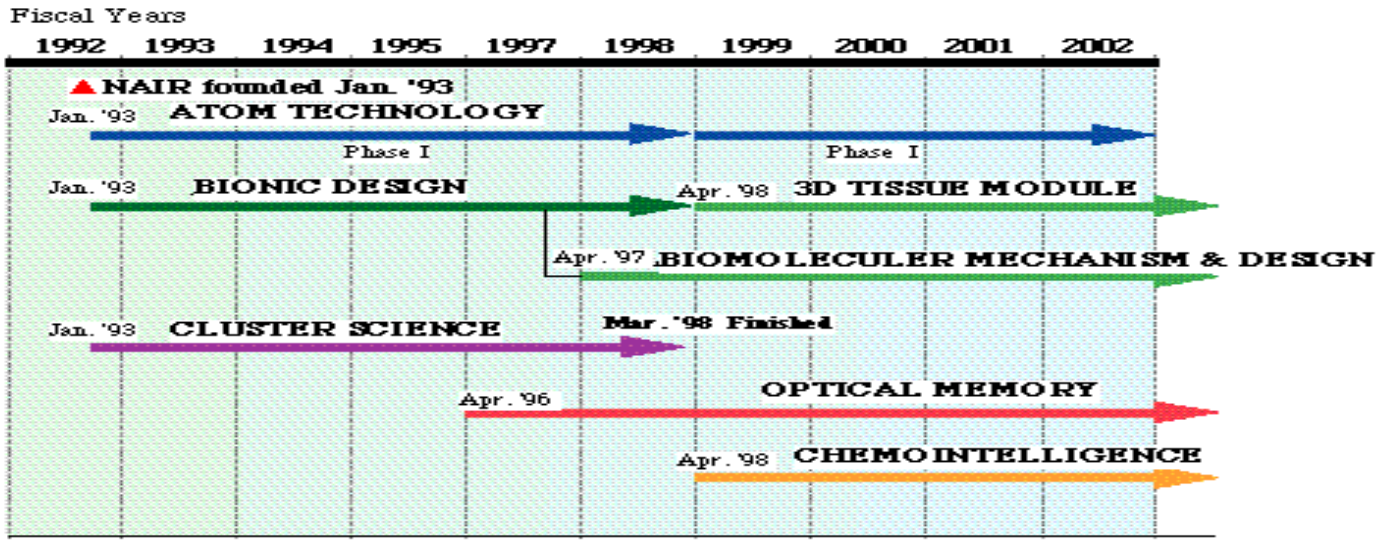

trends in
June 1998 Vol. 14 No. 6 pp. 211–251
GENETICS
Limb development
The wasted mouse
Proteolysis and the F-box
Retinoblastoma gene family
Cross-phylum rescue by otd/Otx

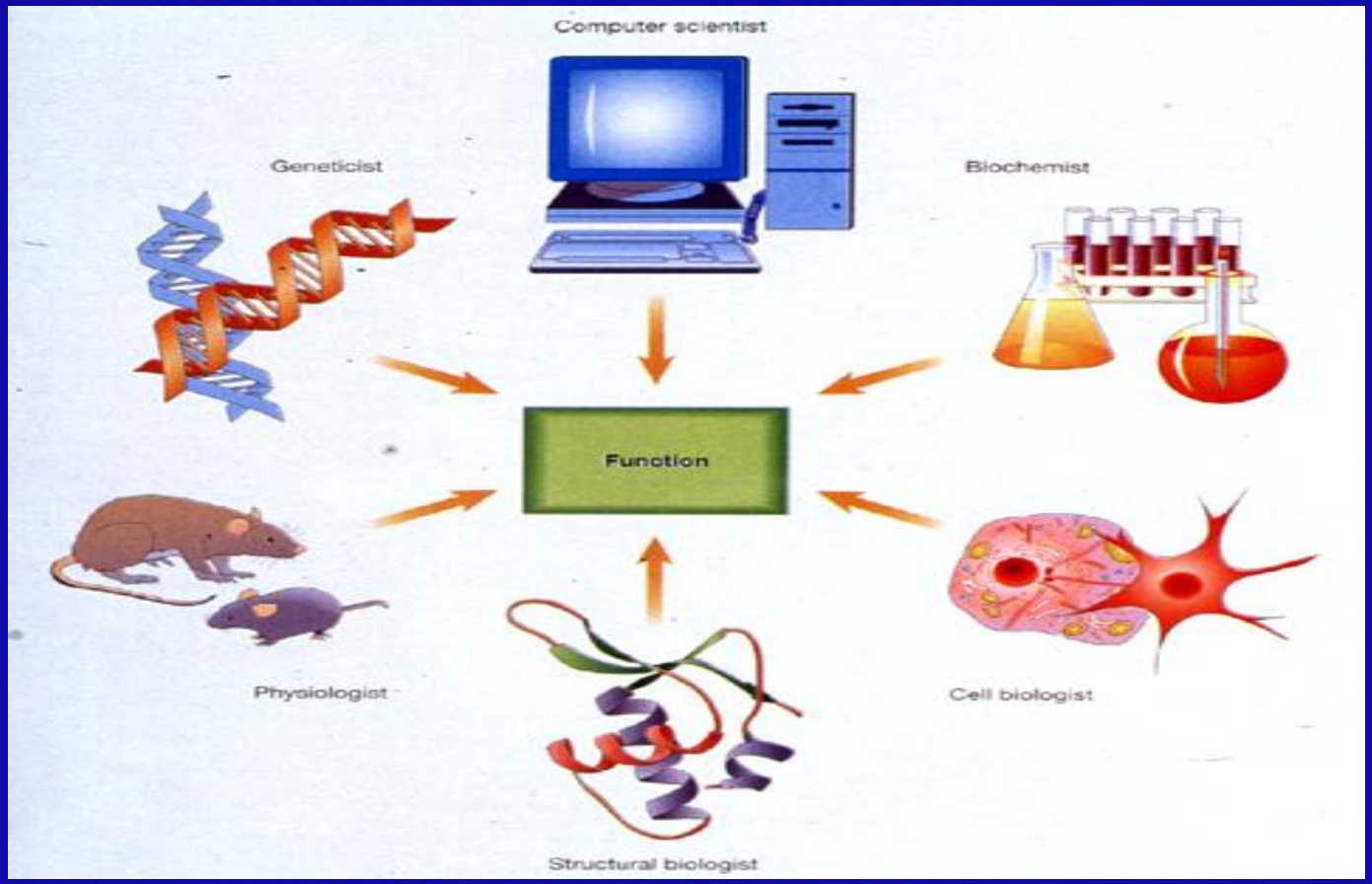
Diseases that could be helped by stem cell research.Number of persons affected in USA and in Italy

Condition	Number of persons affected (milion) USA	Italy
Cardiovascular diseases	58	9.5
Autoimmune diseases	30	
Diabetes	16	2.1
Osteoporosis	10	2.2
Cancer	8.2	0.5
Alzheimer's disease	4	0.5
Parkinson's disease	1.5	0.033
Burns (severe)	0.3	
Spinal cord injuries	0.25	
Birth defects	0.15 (per year)	0.016 (per year)
Total	128.4	

National Institute for Advanced Interdisciplinary Research (NAIR)

Research Projects




Computer scientist
Geneticist
Biochemist
Function
Physiologist
Cell biologist
Structural biologist