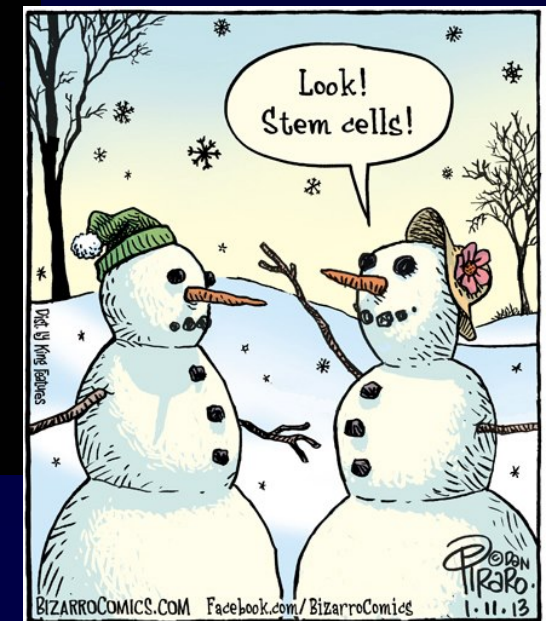
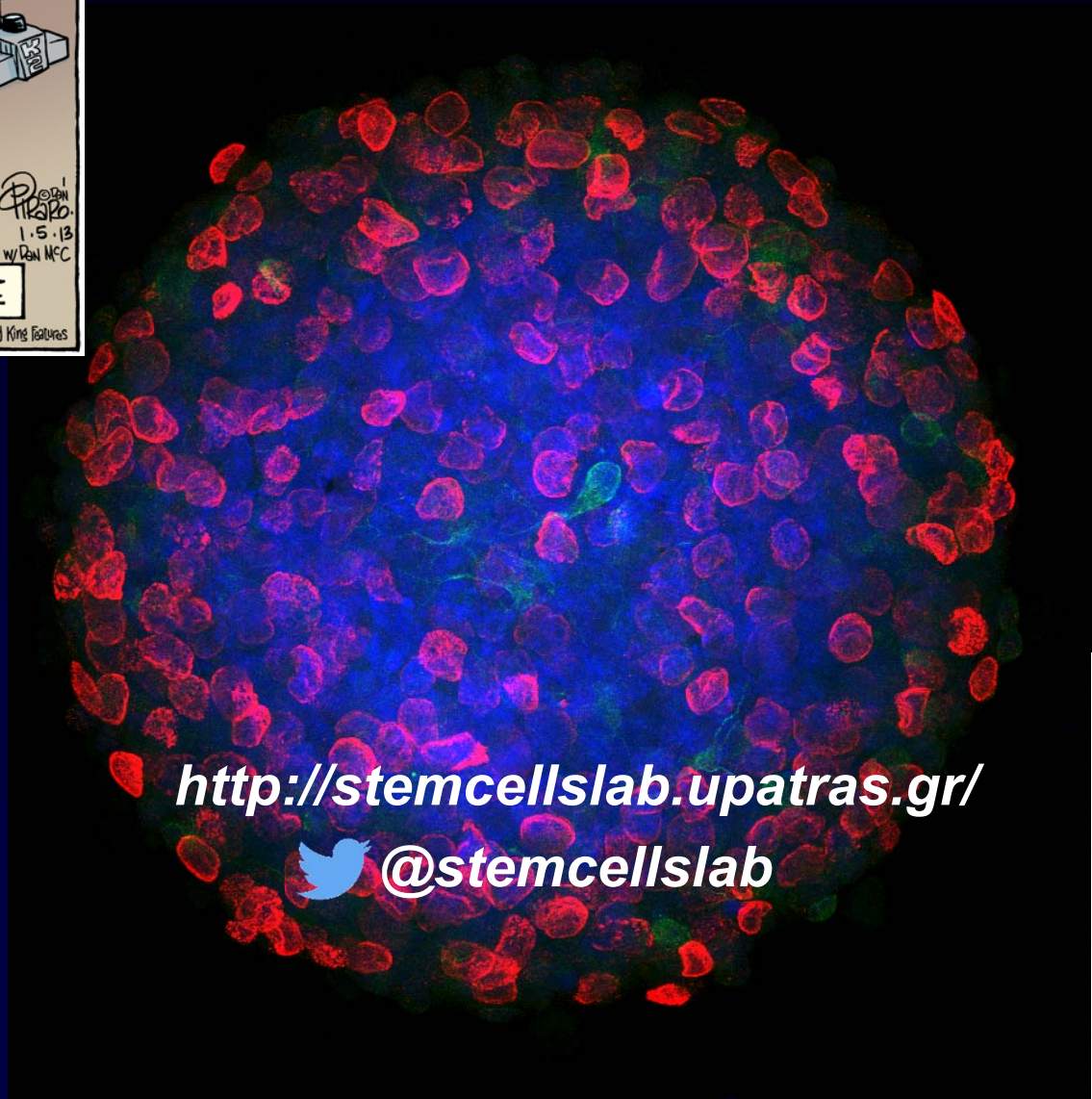
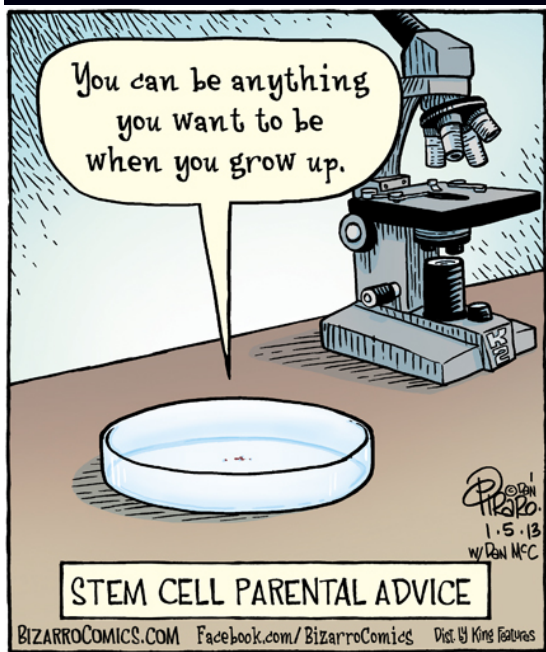




Βασικές ιδιότητες βλαστικών κυττάρων

- Αυτοανανέωση
- Διαφοροποίηση
 - Δυνατότητα δημιουργίας περισσότερων του ενός κυτταρικού τύπου

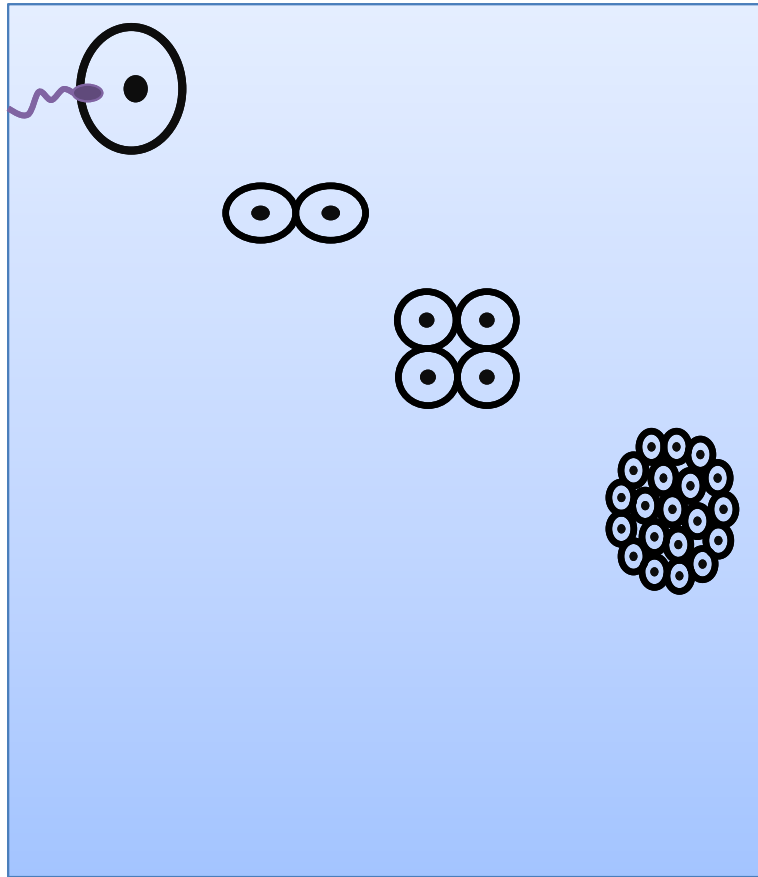
Φυσιολογικός ρόλος βλαστικών κυττάρων

- Δημιουργία των ιστών/οργάνων κατά την εμβρυογένεση
- Αντικατάσταση κυττάρων
- Απόκτηση νέων ιδιοτήτων
- Διατήρηση ομοιόστασης

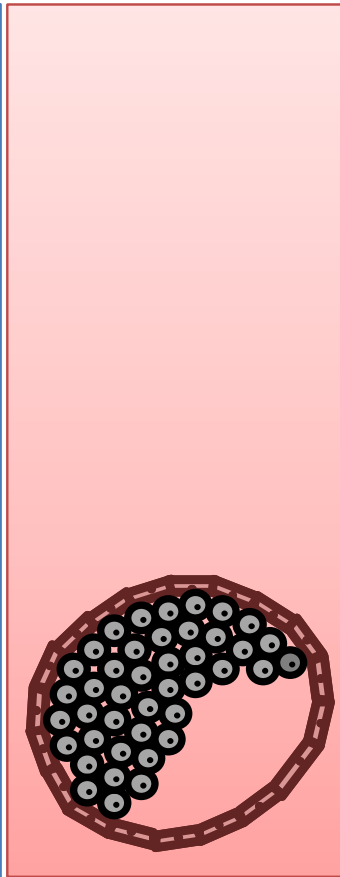
Βλαστικά κύτταρα

- Εμβρυϊκά βλαστικά κύτταρα
- Ενήλικα βλαστικά κύτταρα ή σωματικά βλαστικά κύτταρα

Πληθυσμοί βλαστικών κυττάρων

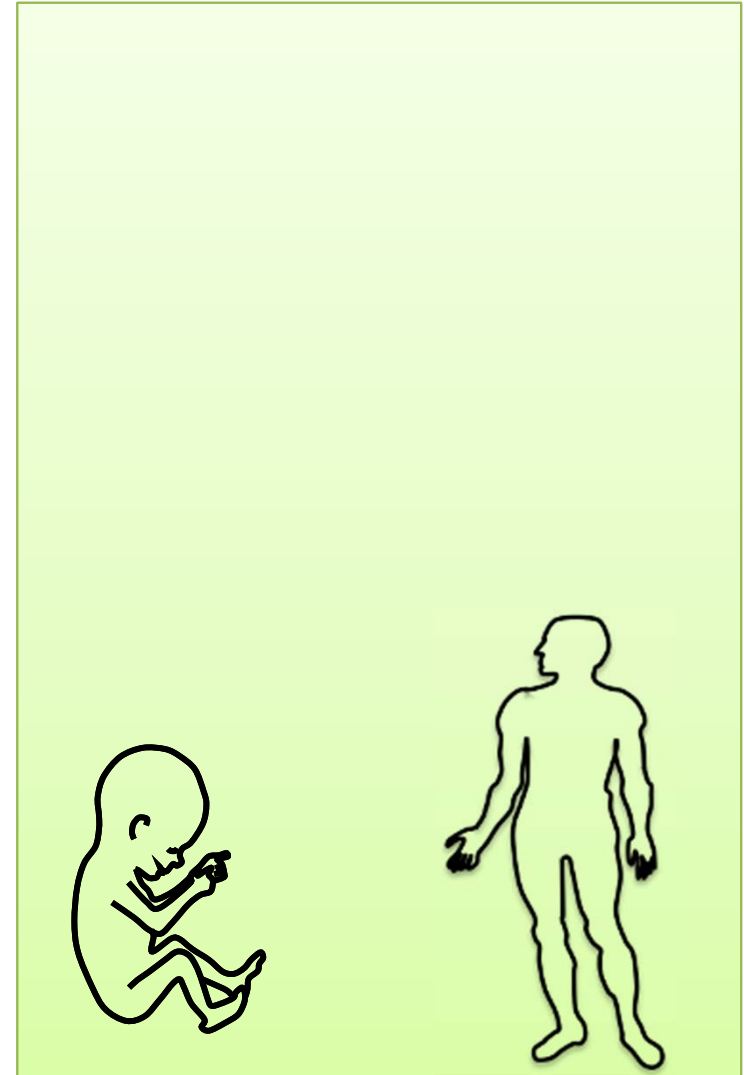


Ολοδύναμα
Εμβρυονικά βλαστικά κύτταρα



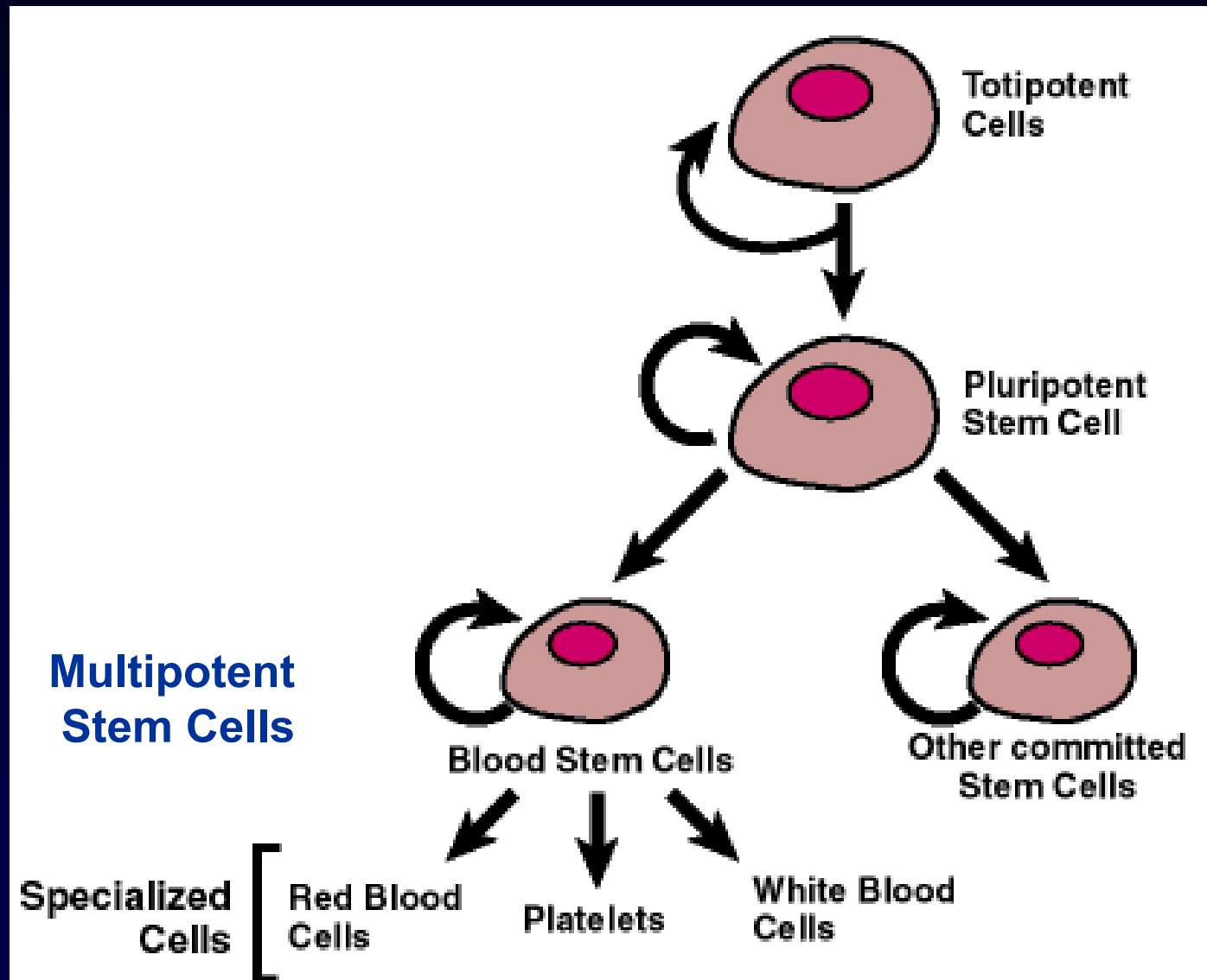
Πολυδύναμα
Εμβρυονικά
βλαστικά
κύτταρα

Εμβρυονικά βλαστικά κύτταρα
(ολοδύναμα, πολυδύναμα)



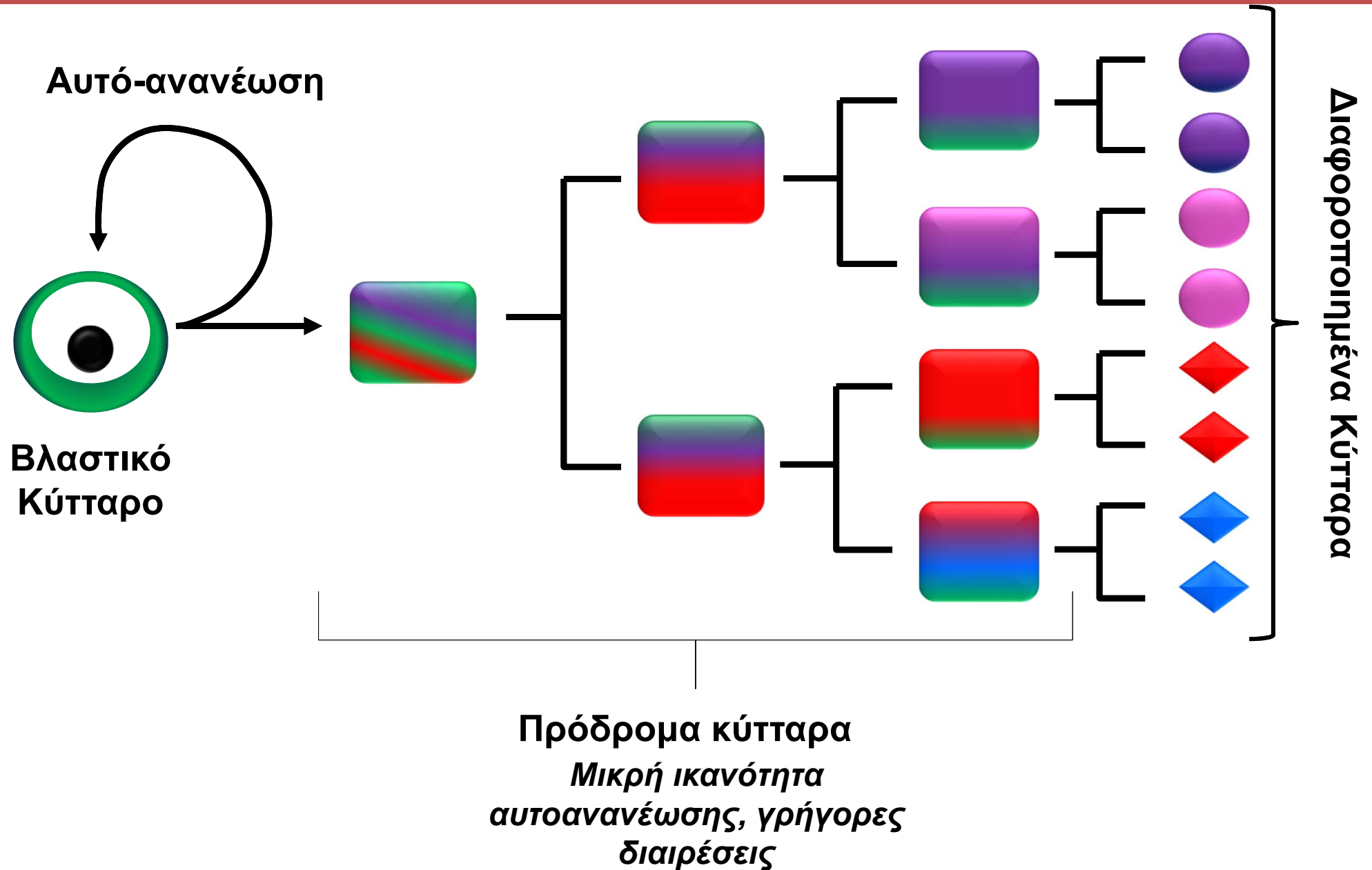
Σωματικά βλαστικά κύτταρα
Βλαστικά κύτταρα ιστών
Έμβρυο, νεογέννητο, ενήλικο άτομο

Ολοδύναμα και Πολυδύναμα Βλαστικά κύτταρα



Διαφοροποίηση βλαστικών κυττάρων μία πολυσταδιακή διαδικασία

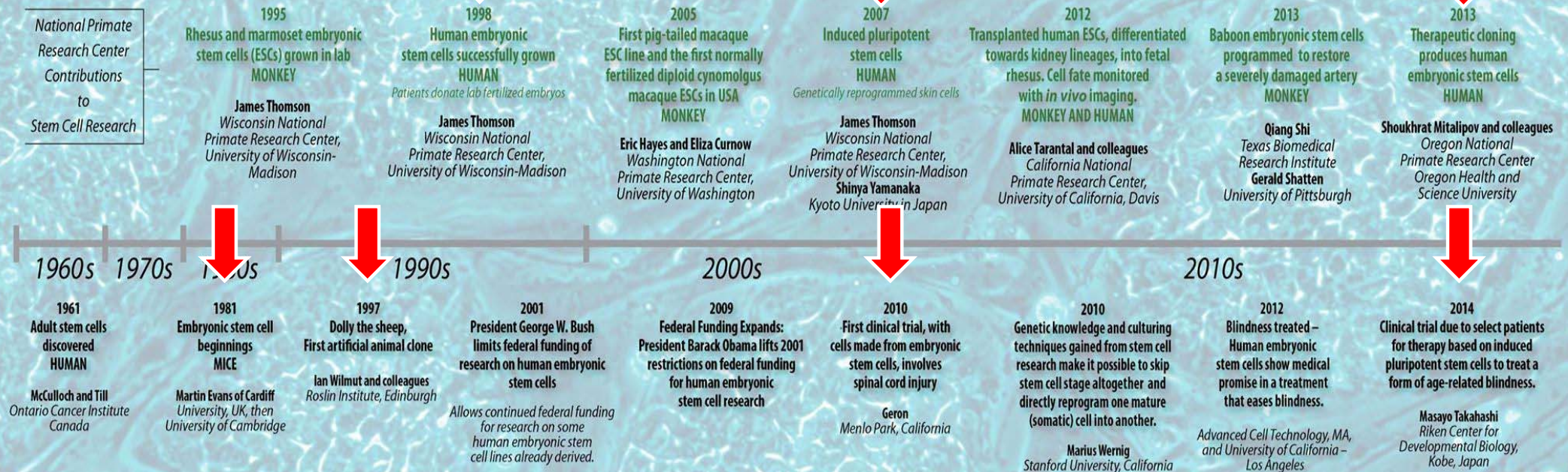
Κυτταρική γενεαλογία σε έναν αναγεννώμενο ιστό



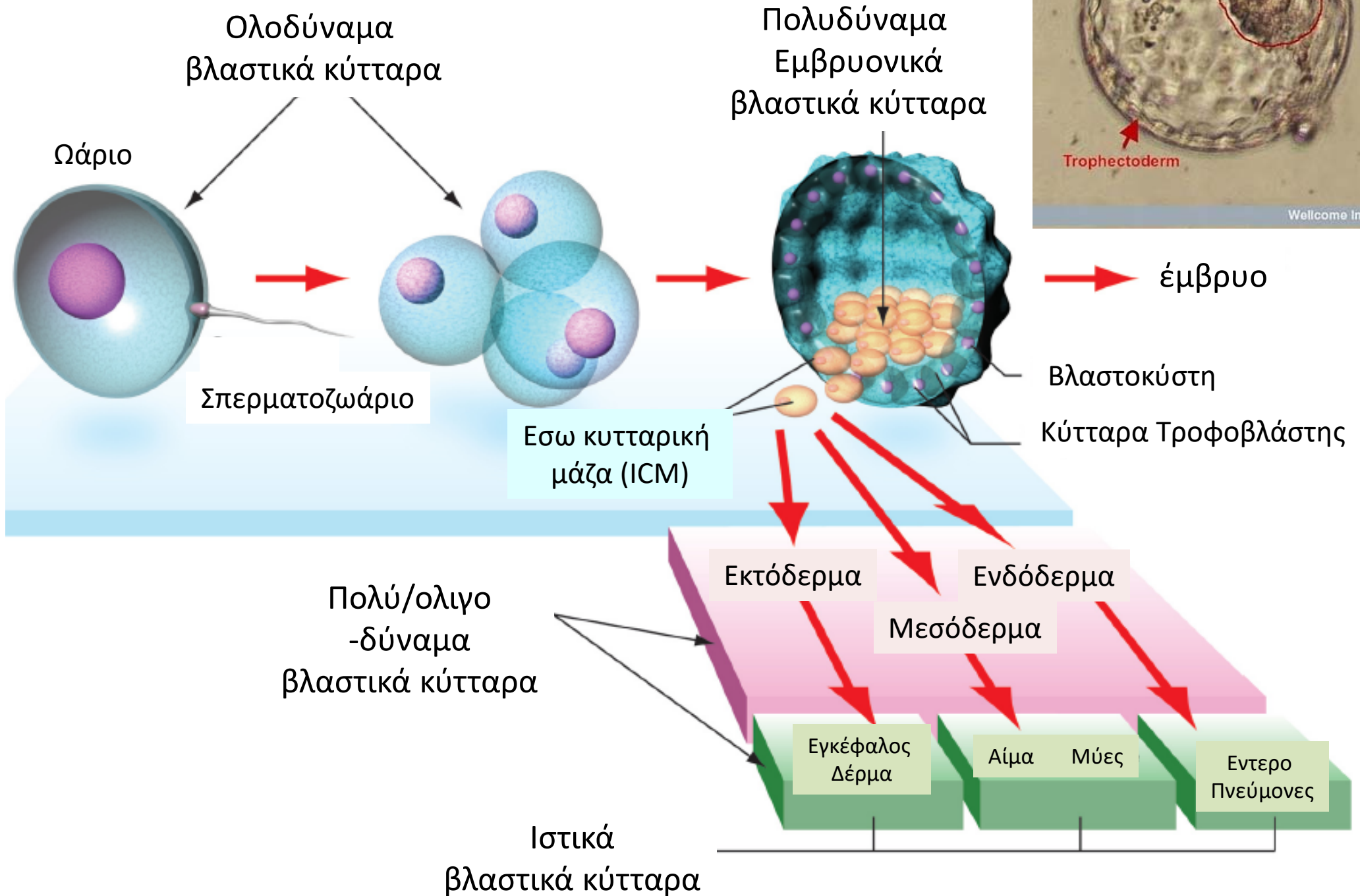
Σημαντικές ημερομηνίες για τη μελέτη των βλαστικών κυττάρων

NATIONAL PRIMATE RESEARCH CENTERS *On the forefront of stem cell research*

■ Since 1961, researchers have been investigating the possibilities of what stem cells can contribute to advancing human and animal health.



Embryonic stem cells (ES cells)



Πολυδύναμα εμβρυικά βλαστικά κύτταρα (ES)

**The Nobel Prize in Physiology or Medicine for 2007
jointly to
Mario R. Capecchi, Martin J. Evans and Oliver Smithies
for their discoveries of
"principles for introducing specific gene modifications in mice by
the use of embryonic stem cells"**



Mario R. Capecchi



Sir Martin J. Evans



Oliver Smithies

- *Modification of genes by homologous recombination*
- *Embryonic stem cells – vehicles to the mouse germ line*
- *Birth of the knockout mouse – the beginning of a new era in genetics*
- *Gene targeting is used to study health and disease*

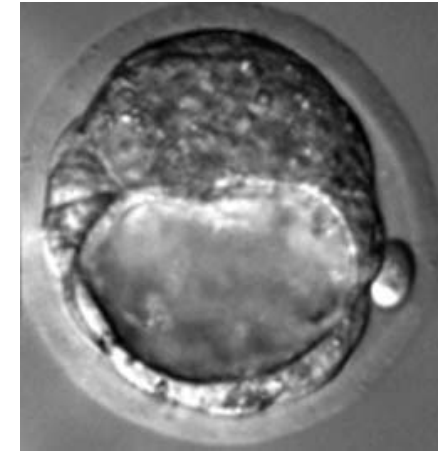
1981-Πρώτη in vitro καλλιέργεια βλαστικών κυττάρων από ποντίκια

- Ο Martin Evans είναι ο πρώτος που ταυτοποιεί τα εμβρυικά βλαστικά κύτταρα σε ποντίκια.
- Επιτυγχάνει να αναπτύξει κυτταρικές σειρές από το ενδόδερμα εμβρύων στο στάδιο της βλαστοκύστης.

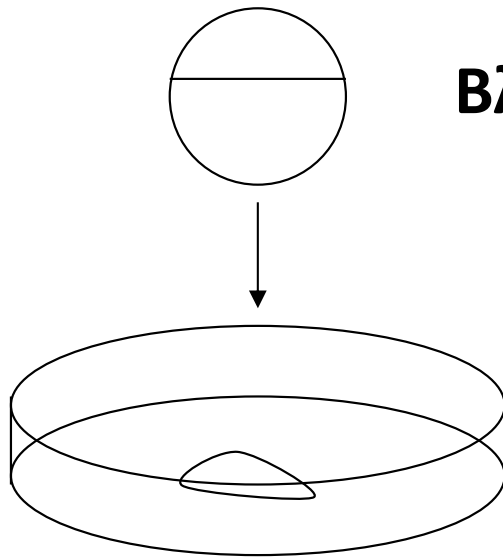


Martin Evans

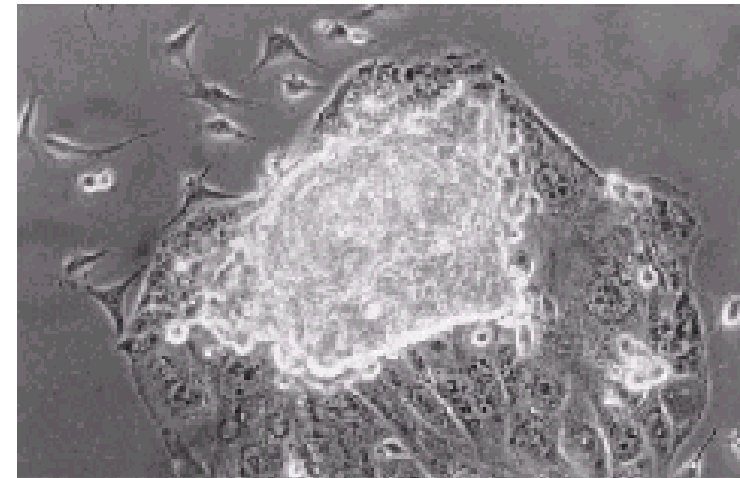
Δημιουργία ES cells



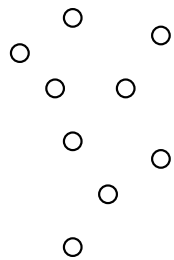
Βλαστοκύστη



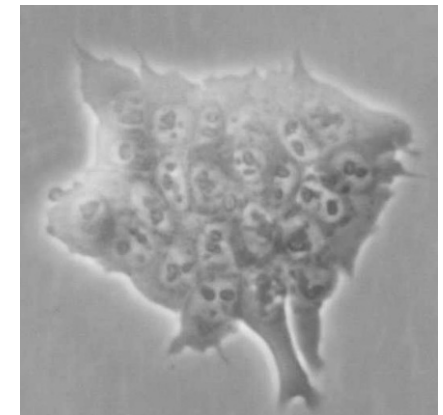
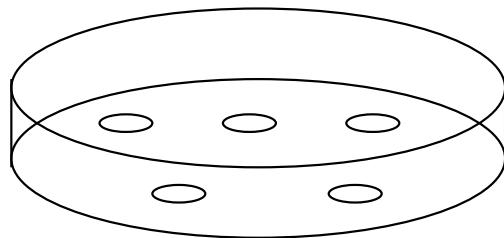
**Προσκόλλησ
η και
ανάπτυξη**

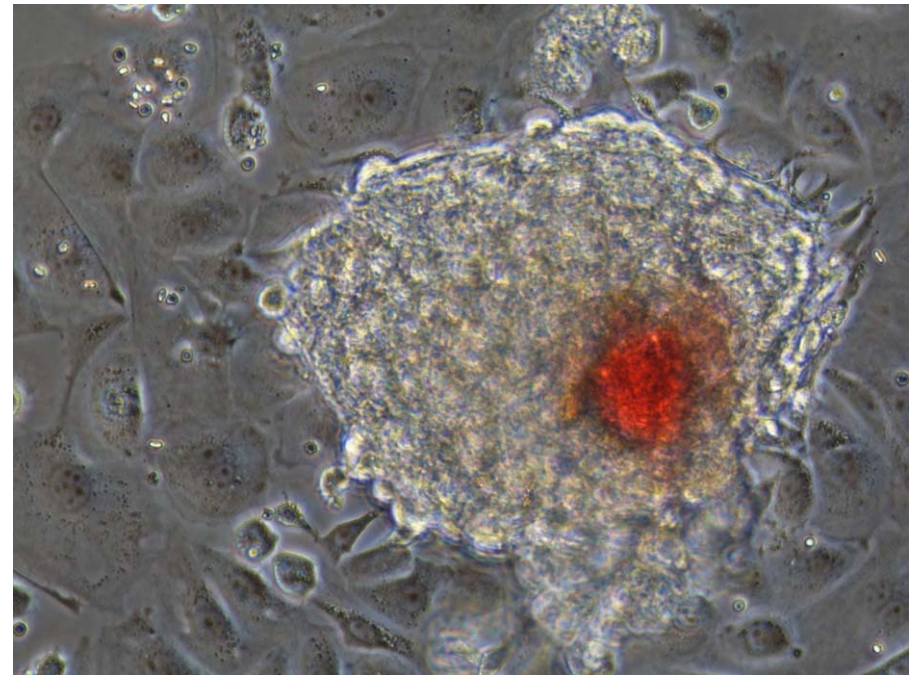
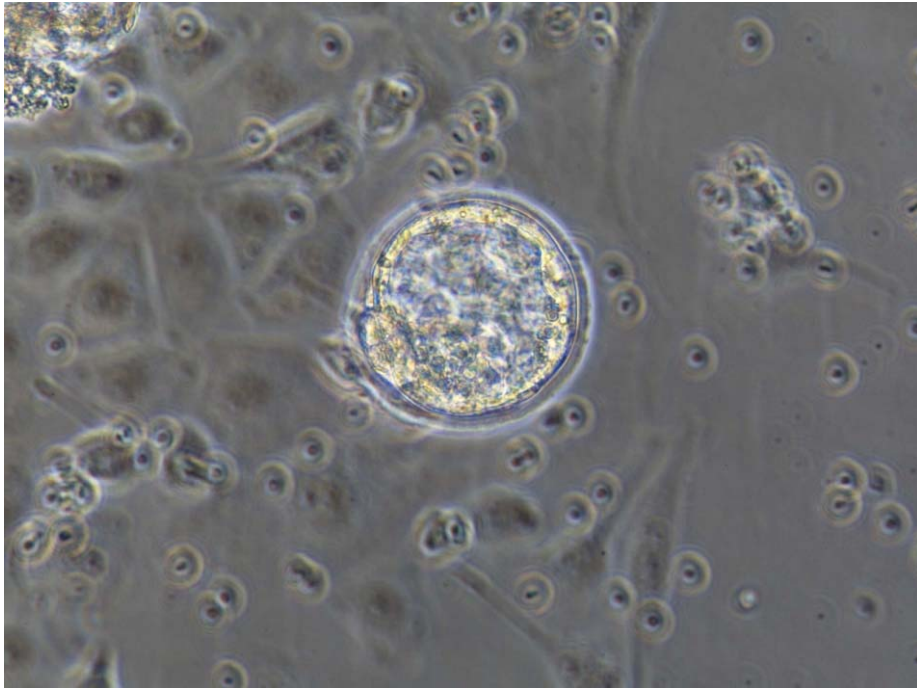
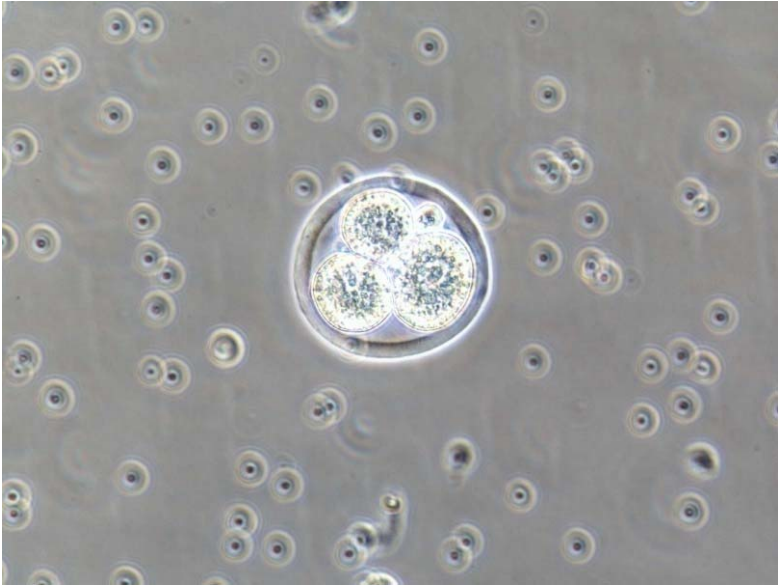


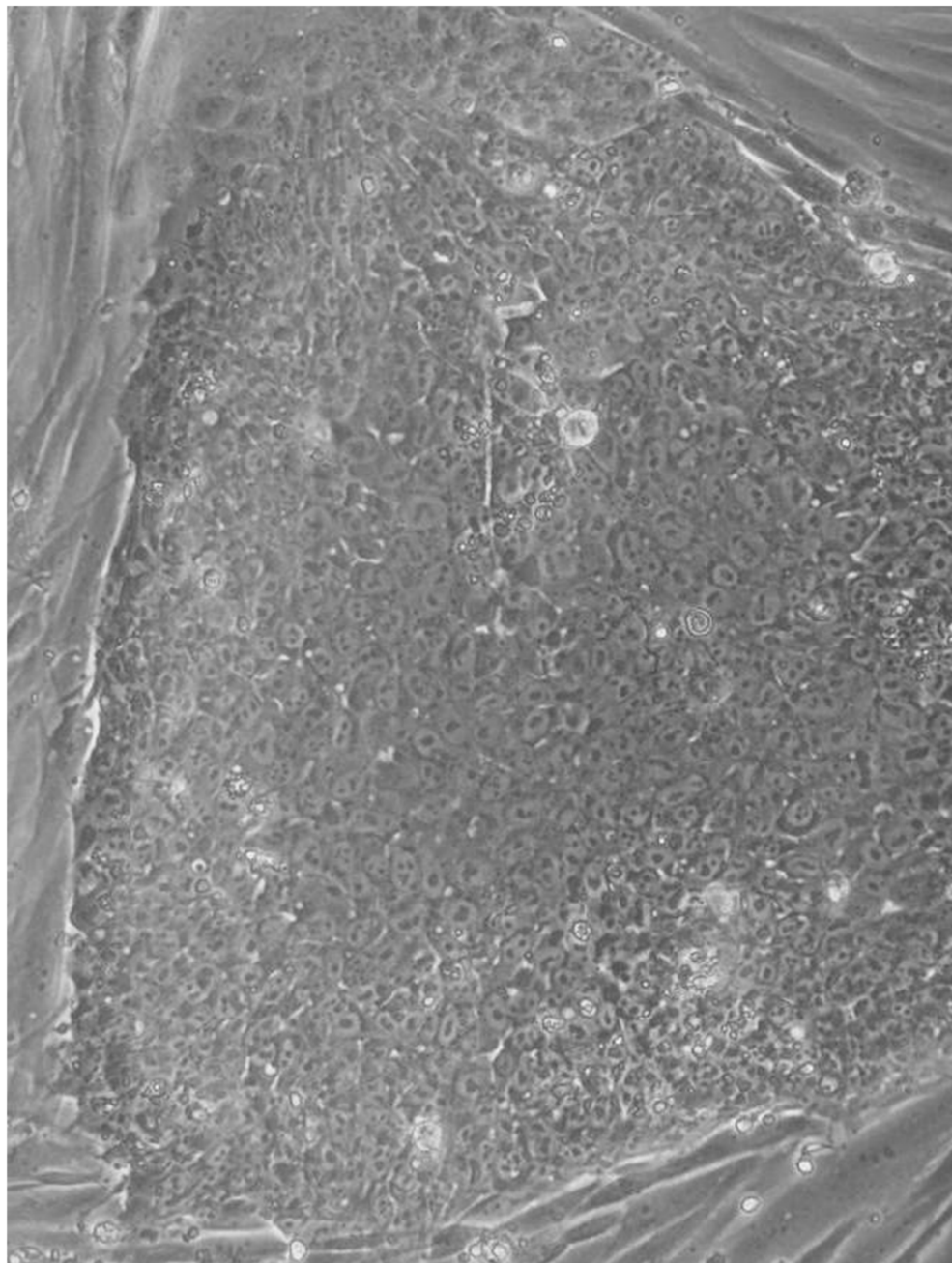
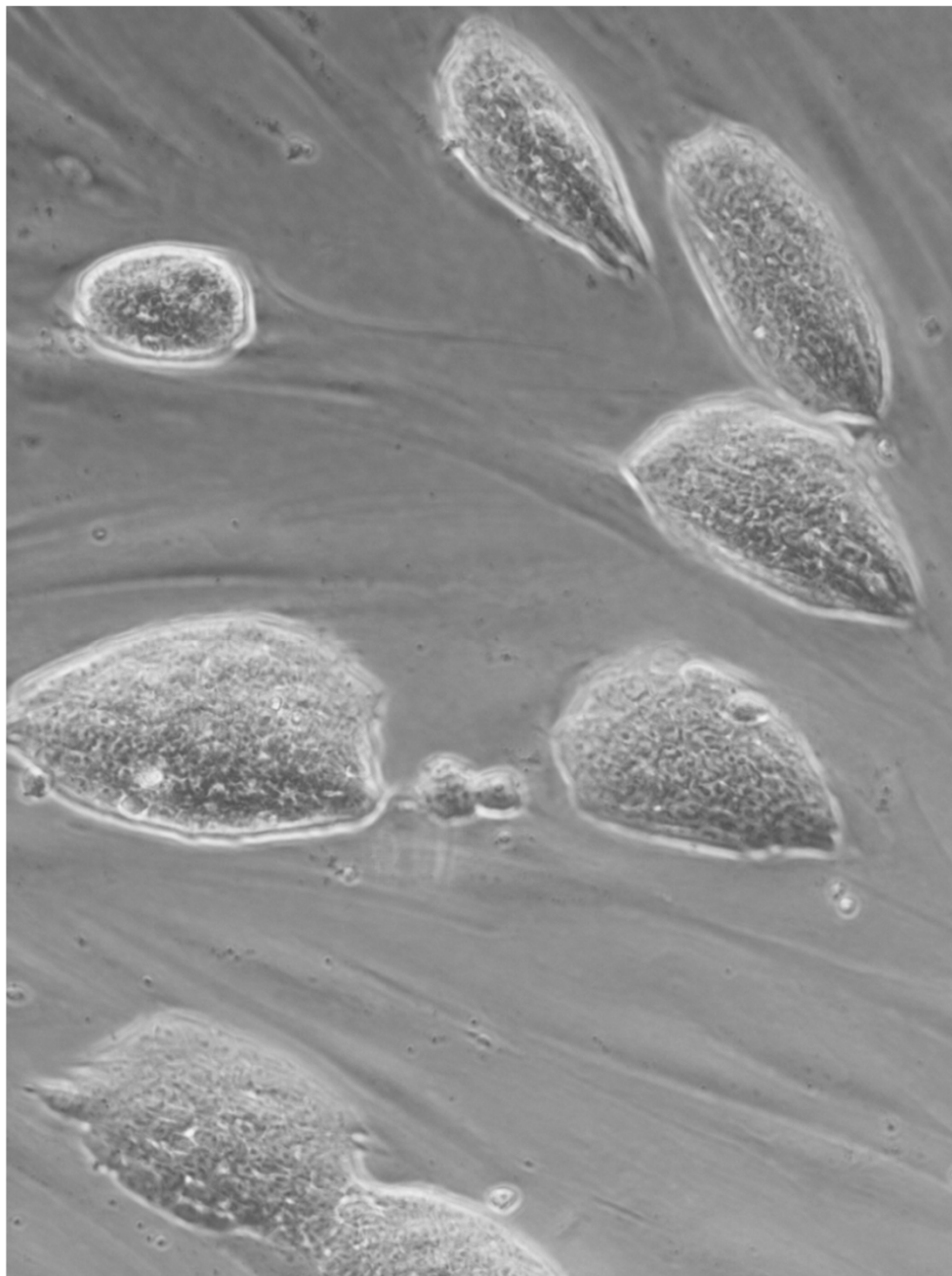
Πρώτη αποδιάταξη



**Πρωτογενής
αποικίες**

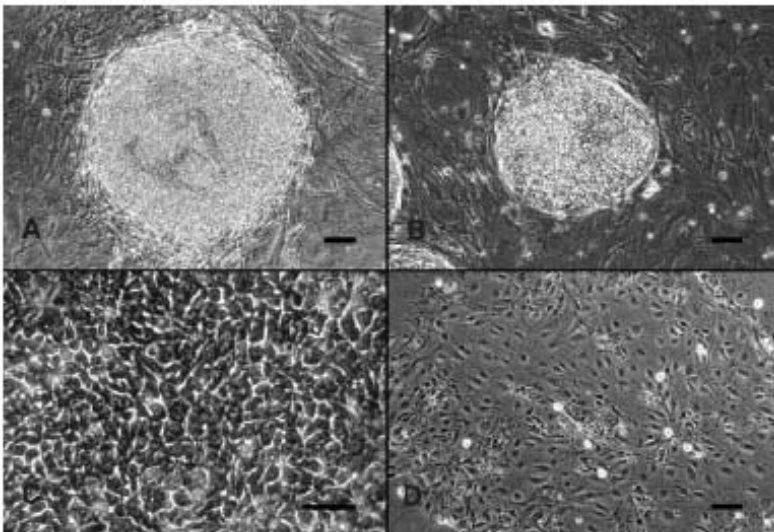






Απομόνωση Ανθρώπινων πολυδύναμων εμβρυικών βλαστικών (ES) κυττάρων

- 1998, James Thomson (University of Wisconsin-Madison)

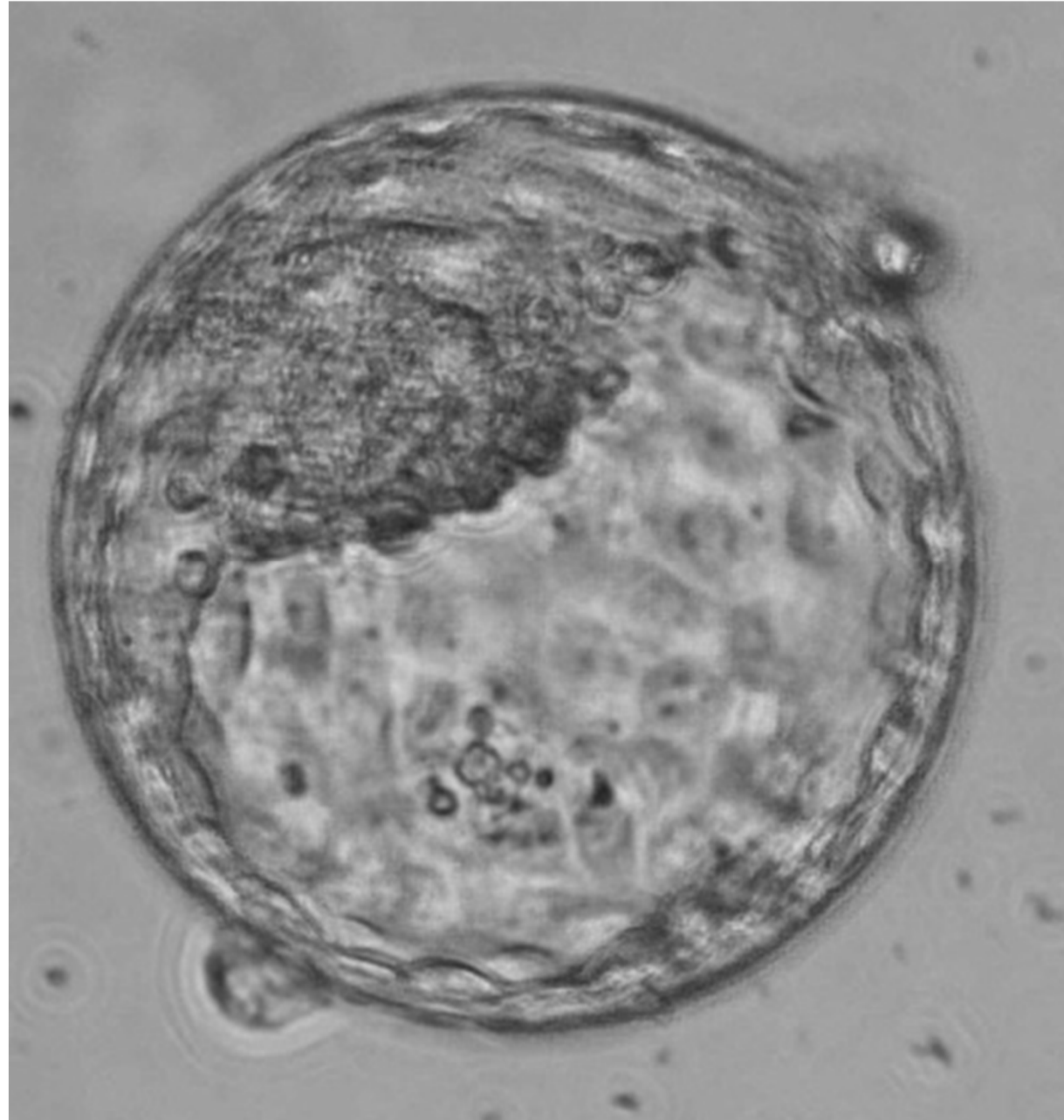


REPORTS Embryonic Stem Cell Lines Derived from Human Blastocysts

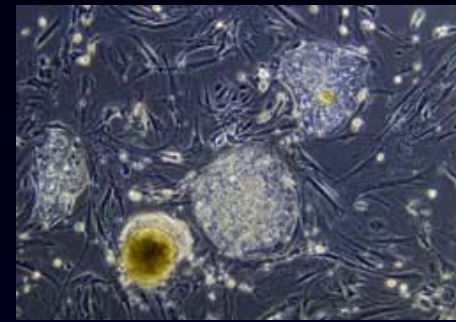
James A. Thomson,* Joseph Itskovitz-Eldor, Sander S. Shapiro,
Michelle A. Waknitz, Jennifer J. Swiergiel, Vivienne S. Marshall,
Jeffrey M. Jones

Human blastocyst-derived, pluripotent cell lines are described that have normal karyotypes, express high levels of telomerase activity, and express cell surface markers that characterize primate embryonic stem cells but do not characterize other early lineages. After undifferentiated proliferation in vitro for 4 to 5 months, these cells still maintained the developmental potential to form trophoblast and derivatives of all three embryonic germ layers, including gut epithelium (endoderm); cartilage, bone, smooth muscle, and striated muscle (mesoderm); and neural epithelium, embryonic ganglia, and stratified squamous epithelium (ectoderm). These cell lines should be useful in human developmental biology, drug discovery, and transplantation medicine.

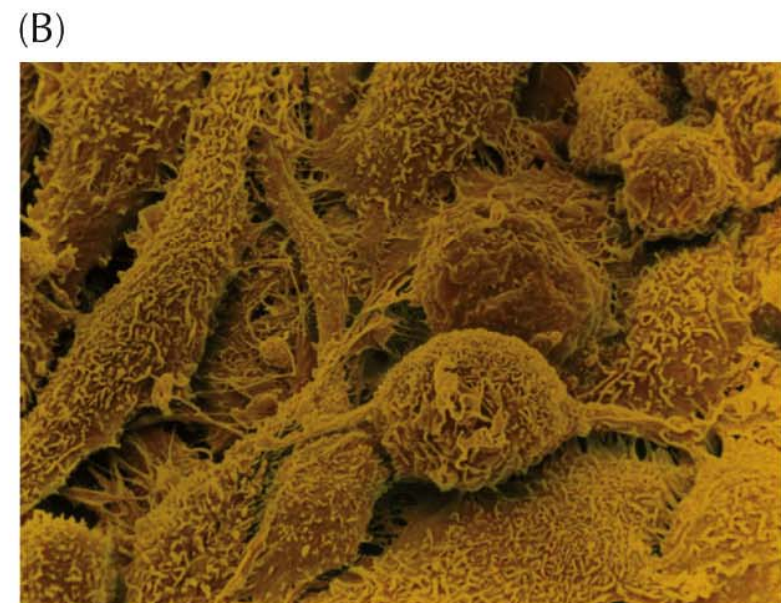
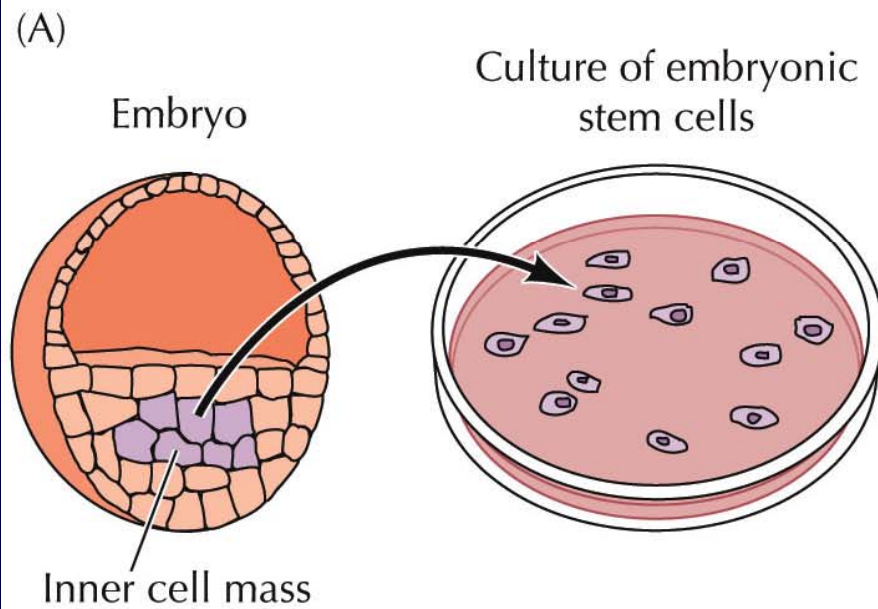
Ανθρώπινη βλαστοκύστη d6



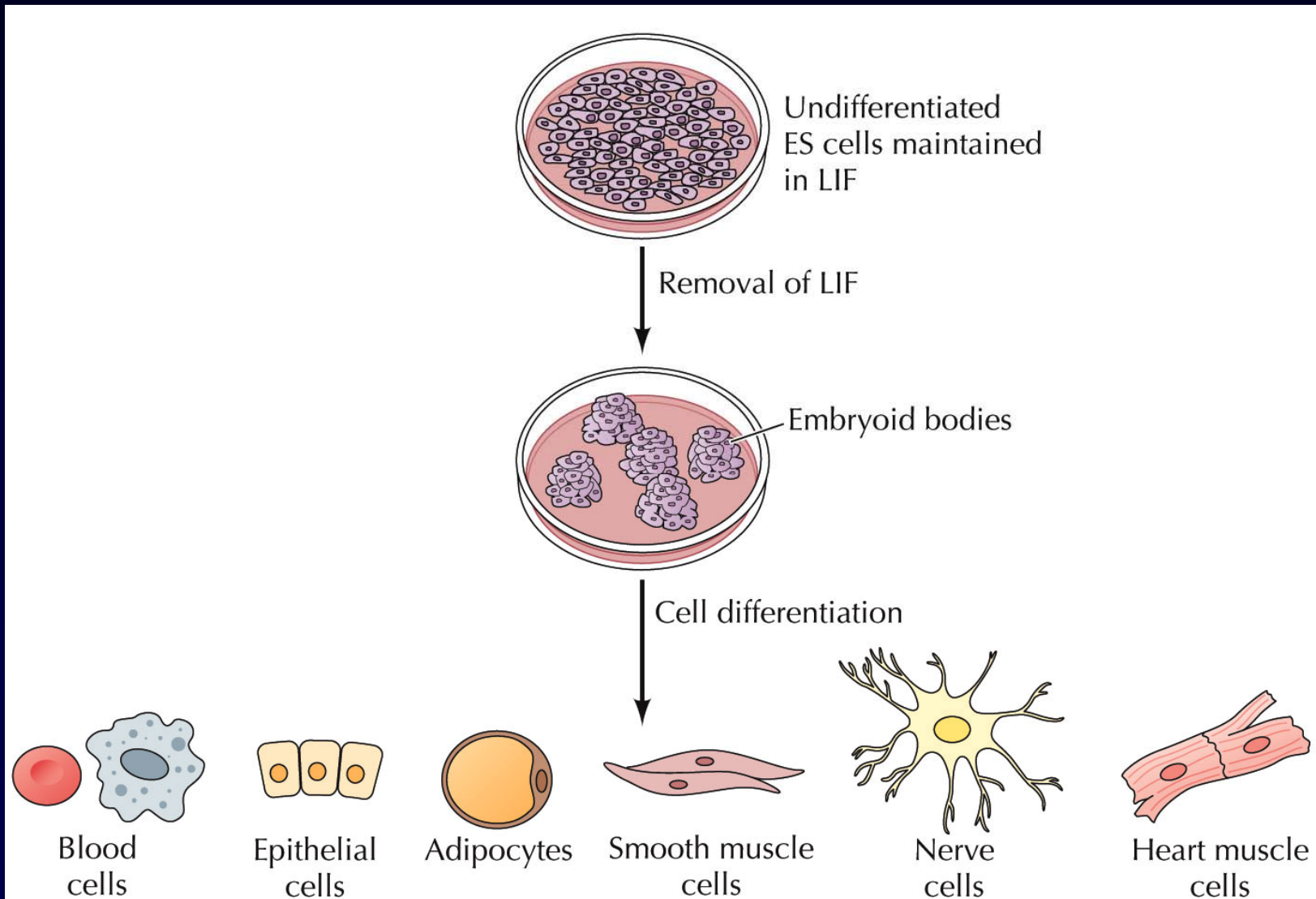
Καλλιέργεια ES



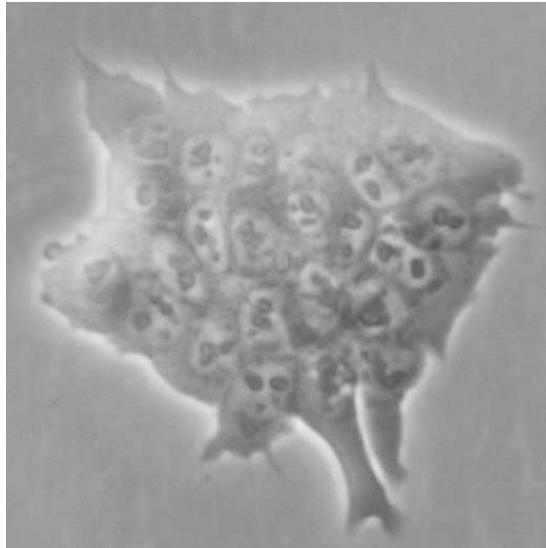
Courtesy of James Thomson,
U. Wisconsin-Madison



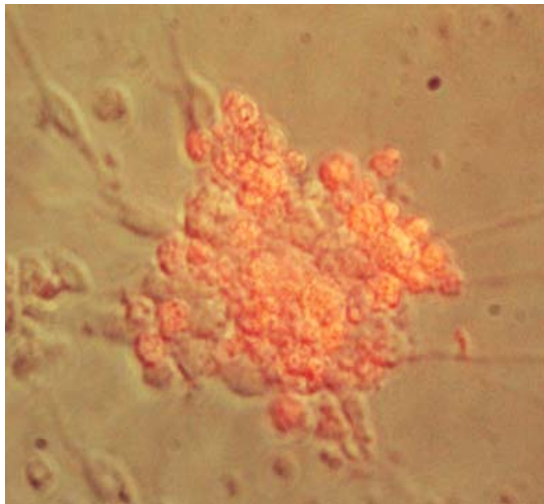
Καλλιέργεια και Διαφοροποίηση ES



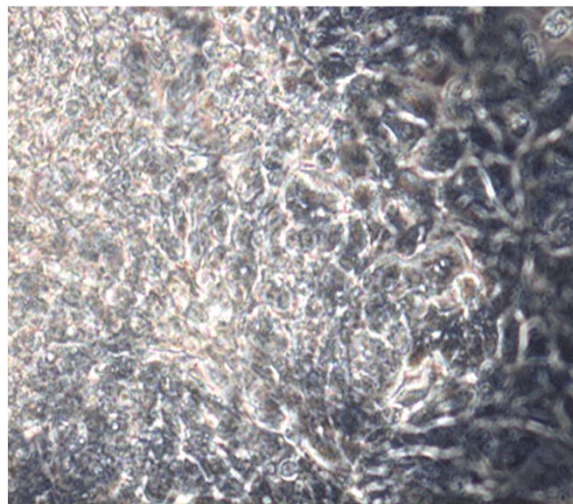
Δημιουργία διαφοροποιημένων κυττάρων από πολυδύναμα εμβρυικά βλαστικά κύτταρα (ES)



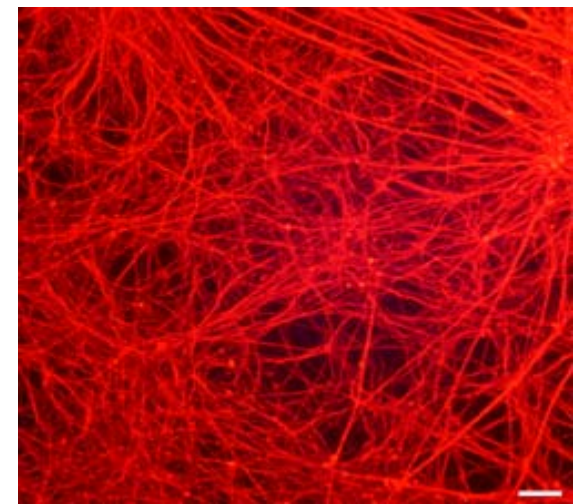
ES κύτταρα



Ενδόδερμα



Μεσόδερμα



Εκτόδερμα

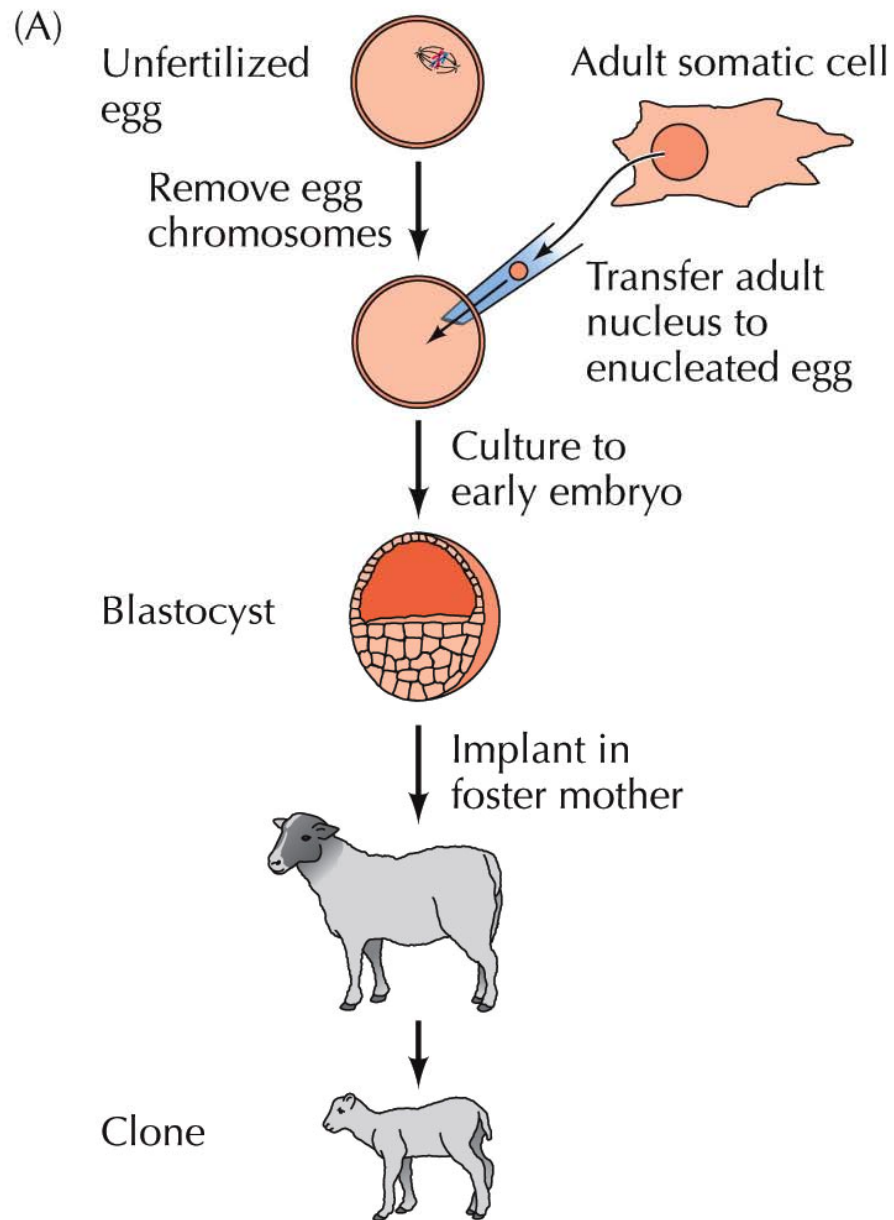
Θεραπευτική κλωνοποίηση

1997 Κλωνοποίηση του προβάτου Dolly
από τους Ian Wilmut and colleagues

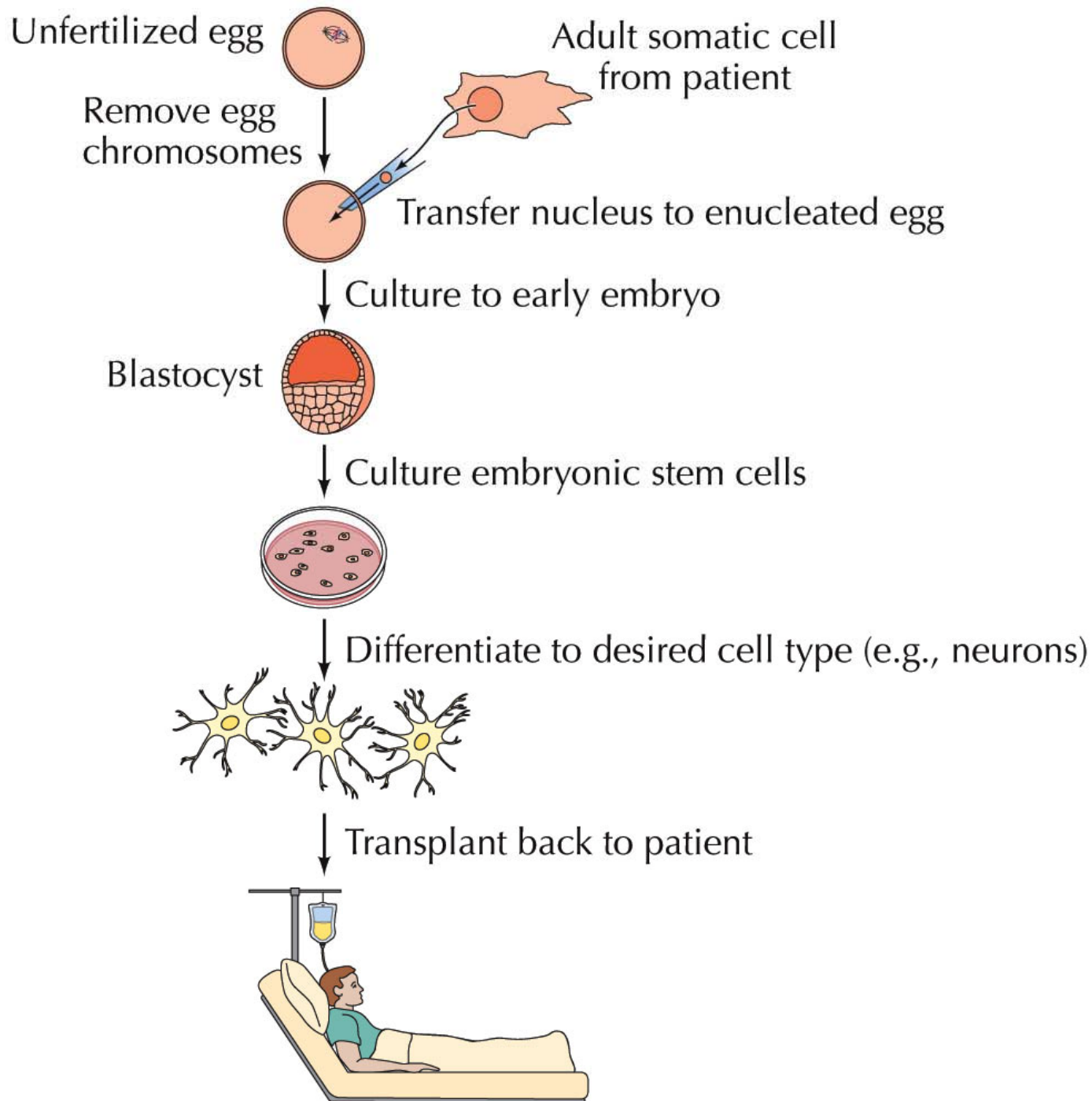
Η Dolly δημιουργήθηκε από μία
διαδικασία που καλείται **somatic cell
nuclear transfer**.

Δύσκολη και αναποτελεσματική μέθοδος για
θηλαστικά

Θεραπευτική κλωνοποίηση



Θεραπευτική κλωνοποίηση

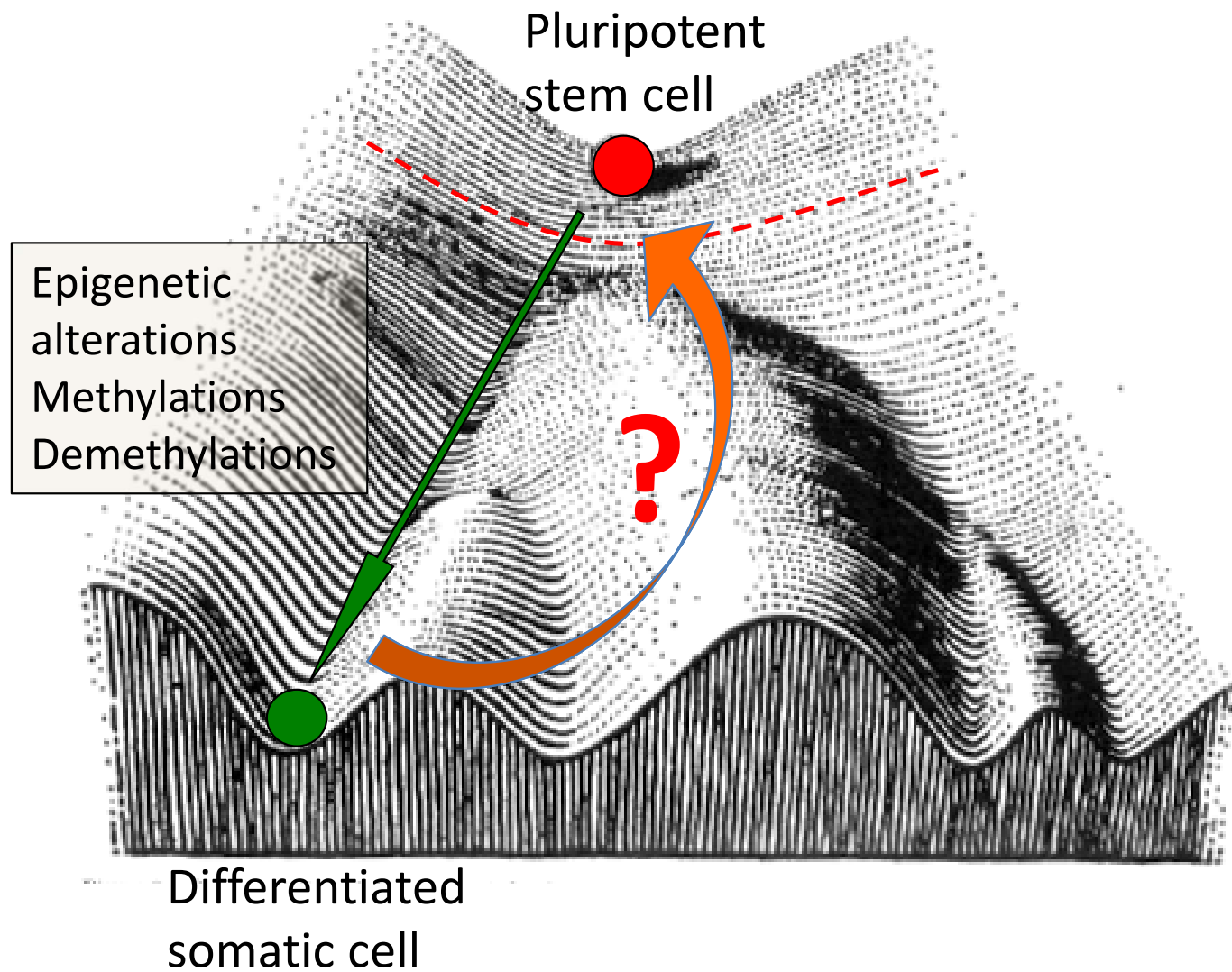


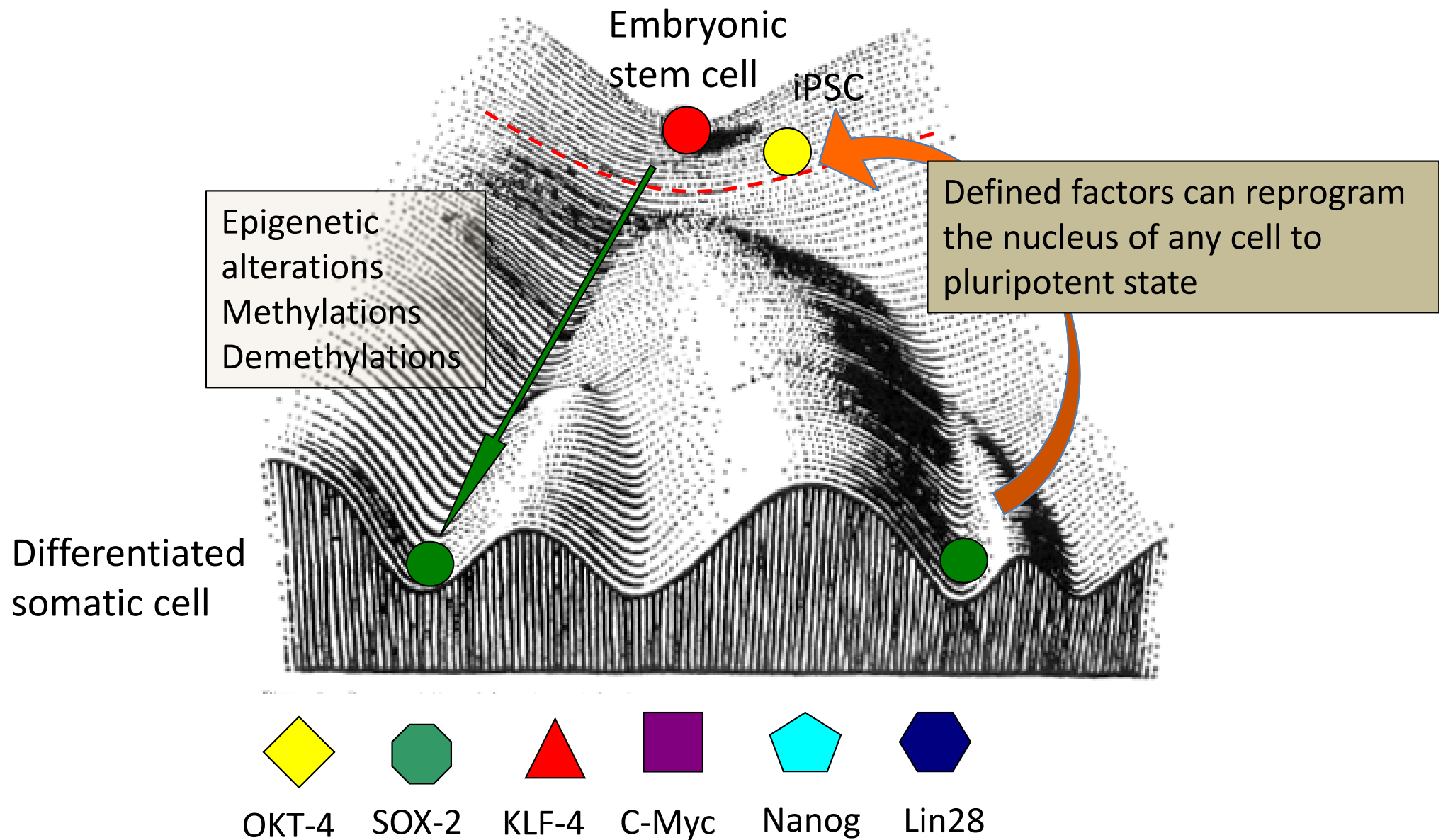
Θεραπευτική κλωνοποίηση

Προβλήματα:

- Χαμηλή αποτελεσματικότητα
- Ηθικά προβλήματα
 - αναπαραγωγική κλωνοποίηση
(**reproductive cloning**)

Κυτταρικός επαναπρογραμματισμός





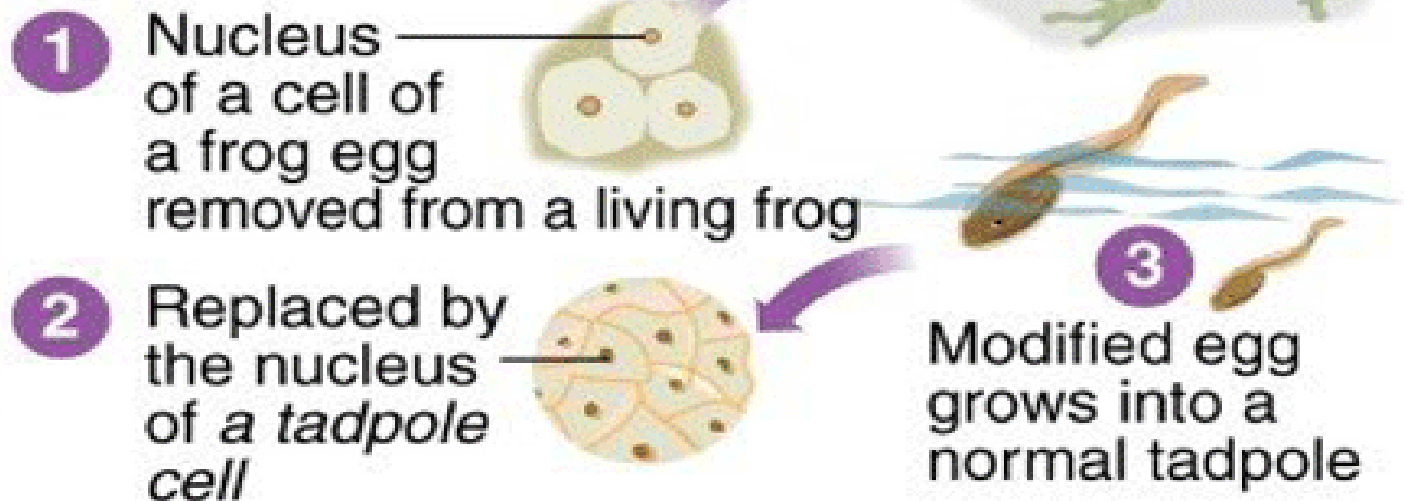
2012 Nobel in Medicine-Physiology

The Nobel Prize for Medicine

Shinya Yamanaka of Japan and John Gurdon of Britain honoured for the their work on cell programming



1962 **Gurdon** discovered that an adult frog cell could be **reprogrammed**



The first experiments

The Developmental Capacity of Nuclei taken from Intestinal Epithelium Cells of Feeding Tadpoles

by J. B. GURDON¹

From the Embryology Laboratory, Department of Zoology, Oxford

WITH ONE PLATE

INTRODUCTION

AN important problem in embryology is whether the differentiation of cells depends upon a stable restriction of the genetic information contained in their nuclei. The technique of nuclear transplantation has shown to what extent the nuclei of differentiating cells can promote the formation of different cell types (e.g. King & Briggs, 1956; Gurdon, 1960c). Yet no experiments have so far been published on the transplantation of nuclei from fully differentiated normal cells. This is partly because it is difficult to obtain meaningful results from such experiments. The small amount of cytoplasm in differentiated cells renders their nuclei susceptible to damage through exposure to the saline medium, and this makes it difficult to assess the significance of the abnormalities resulting from their transplantation. It is, however, very desirable to know the developmental capacity of such nuclei, since any nuclear changes which are necessarily involved in cellular differentiation must have already taken place in cells of this kind.

The experiments described below are some attempts to transplant nuclei from fully differentiated cells. Many of these nuclei gave abnormal results after transplantation, and several different kinds of experiments have been carried out to determine the cause and significance of these abnormalities.

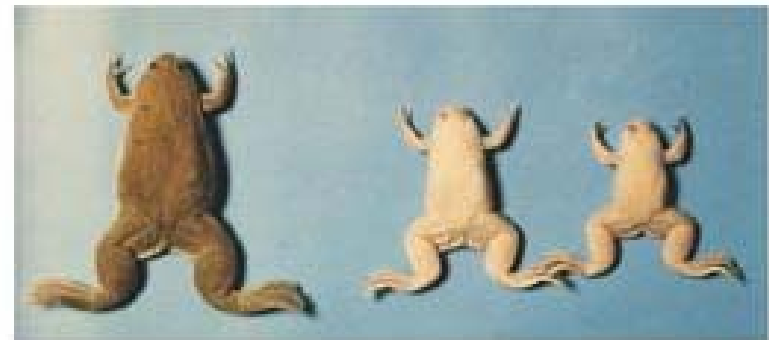
The donor cells used for these experiments were intestinal epithelium cells of feeding tadpoles. This is the final stage of differentiation of many of the endoderm cells whose nuclei have already been studied by means of nuclear transplantation experiments in *Xenopus*. The results to be described here may therefore be regarded as an extension of those previously obtained from differentiating endoderm cells (Gurdon, 1960c).

MATERIAL AND METHODS

The animals used for these experiments belong to the subspecies *Xenopus laevis laevis*. The transplantation technique has been carried out as described previously (Elsdale *et al.*, 1960), except that the donor tissue was exposed to

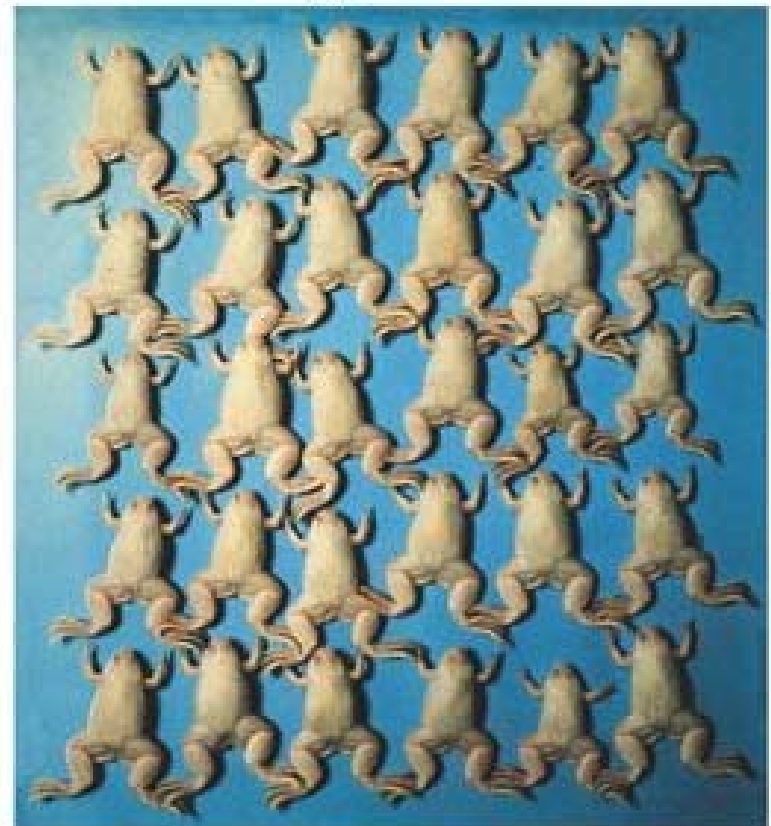
¹ Author's Address: Department of Zoology, Parks Road, Oxford, U.K.

[J. Embryol. exp. Morph., Vol. 10, Part 4, pp. 622-60 December 1962]



Wild-type donor
of enucleated eggs

Albino parents
of nucleus donor

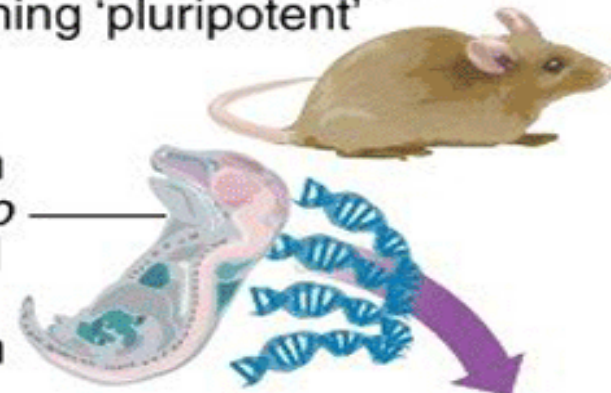


2012 Nobel in Medicine-Physiology

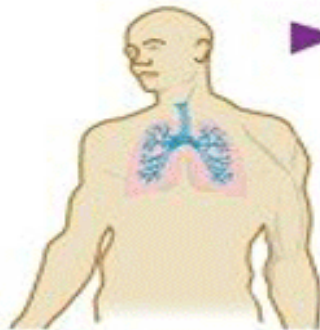


2006 Yamanaka discovered how mice cells could be **returned to their youthful state**, becoming 'pluripotent' stem cells

1 Four genes from *a mouse embryo* were transferred into cells taken from mouse skin



2 Reprogrammed, they became '**pluripotent**' stem cells which could be developed into all cell types of an adult mouse



► '**Pluripotent**' cells, including nerve, heart and liver cells, could now be generated from humans

► The discoveries have raised hopes that replacement tissue may in the future be grown to fight disease in humans

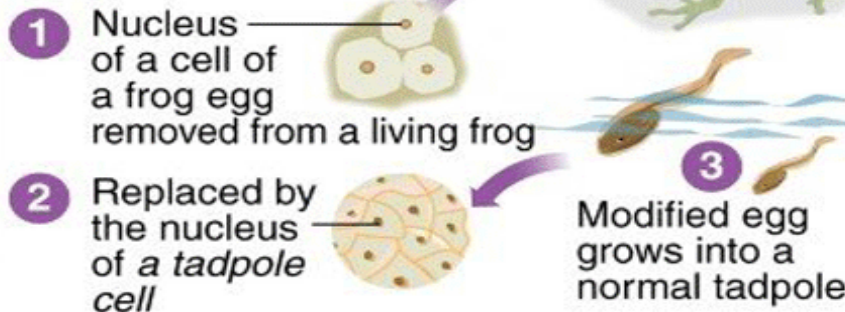
► The process avoids the need to use stem cells from early-stage embryos for research

The Nobel Prize for Medicine

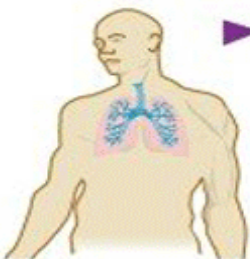
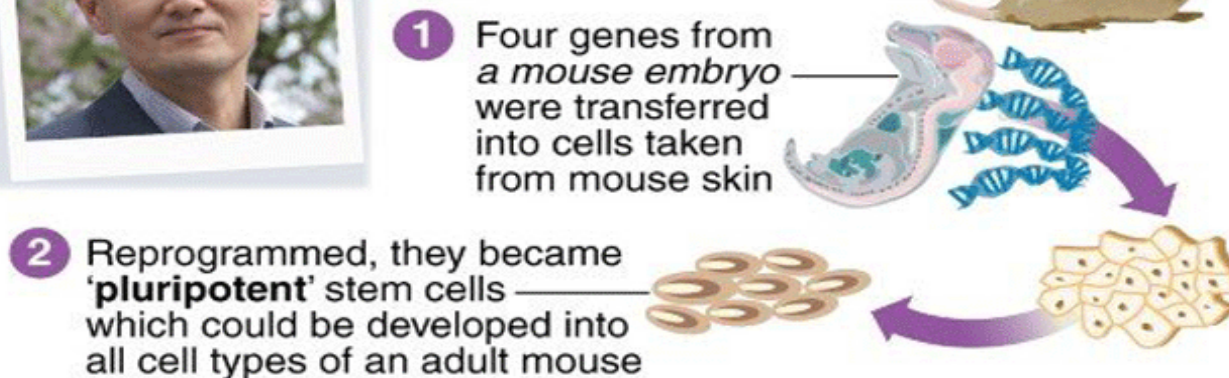
Shinya Yamanaka of Japan and John Gurdon of Britain honoured for their work on cell programming



1962 Gurdon discovered that an adult frog cell could be **reprogrammed**



2006 Yamanaka discovered how mice cells could be **returned to their youthful state**, becoming 'pluripotent' stem cells



► 'Pluripotent' cells, including nerve, heart and liver cells, could now be generated from humans

► The discoveries have raised hopes that replacement tissue may in the future be grown to fight disease in humans

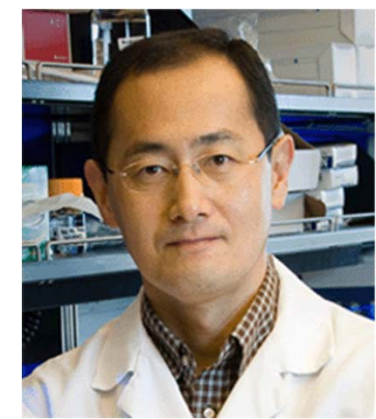
► The process avoids the need to use stem cells from early-stage embryos for research

2012-Βραβείο Nobel Ιατρικής- Φυσιολογίας

**Επαγόμενα πολυδύναμα βλαστικά
κύτταρα
(Induced pluripotent stem cells (iPS))**

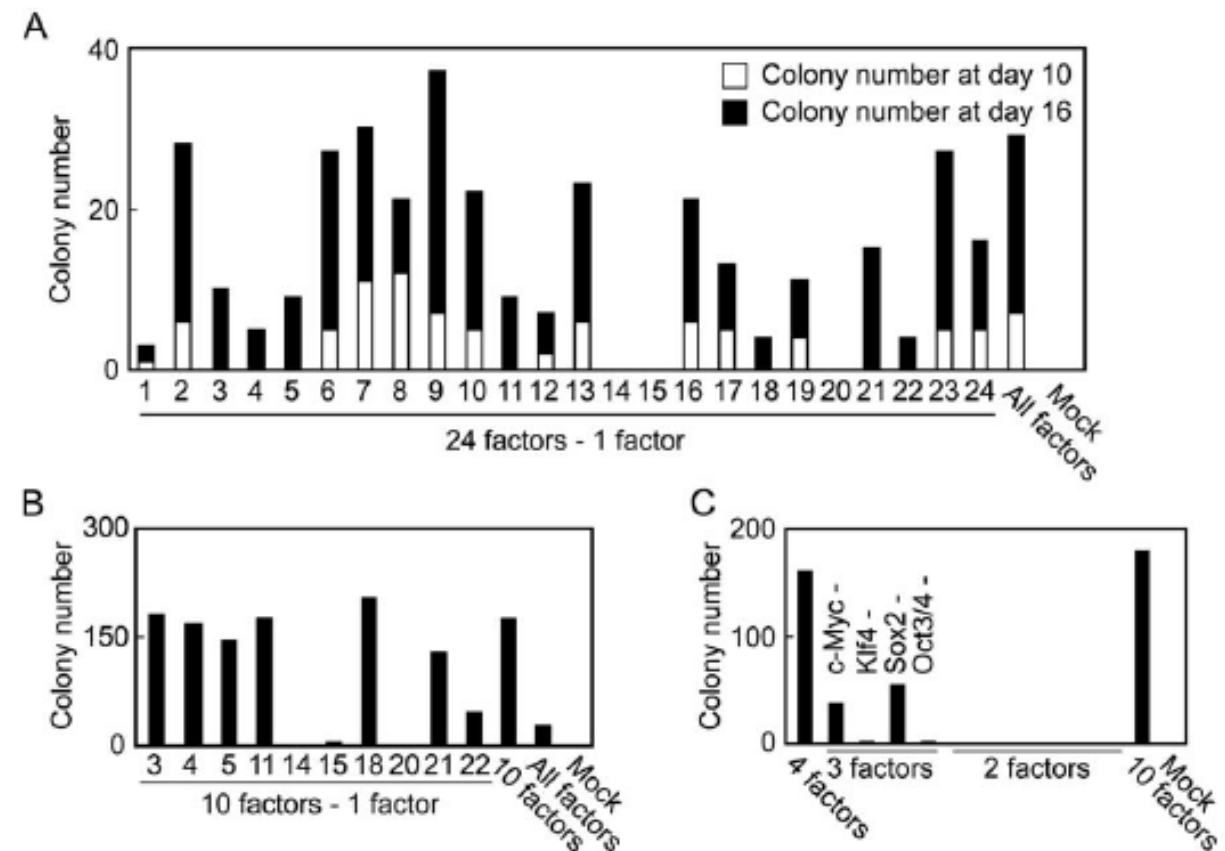
Induction of Pluripotent Stem Cells from Mouse Embryonic and Adult Fibroblast Cultures by Defined Factors

Cell



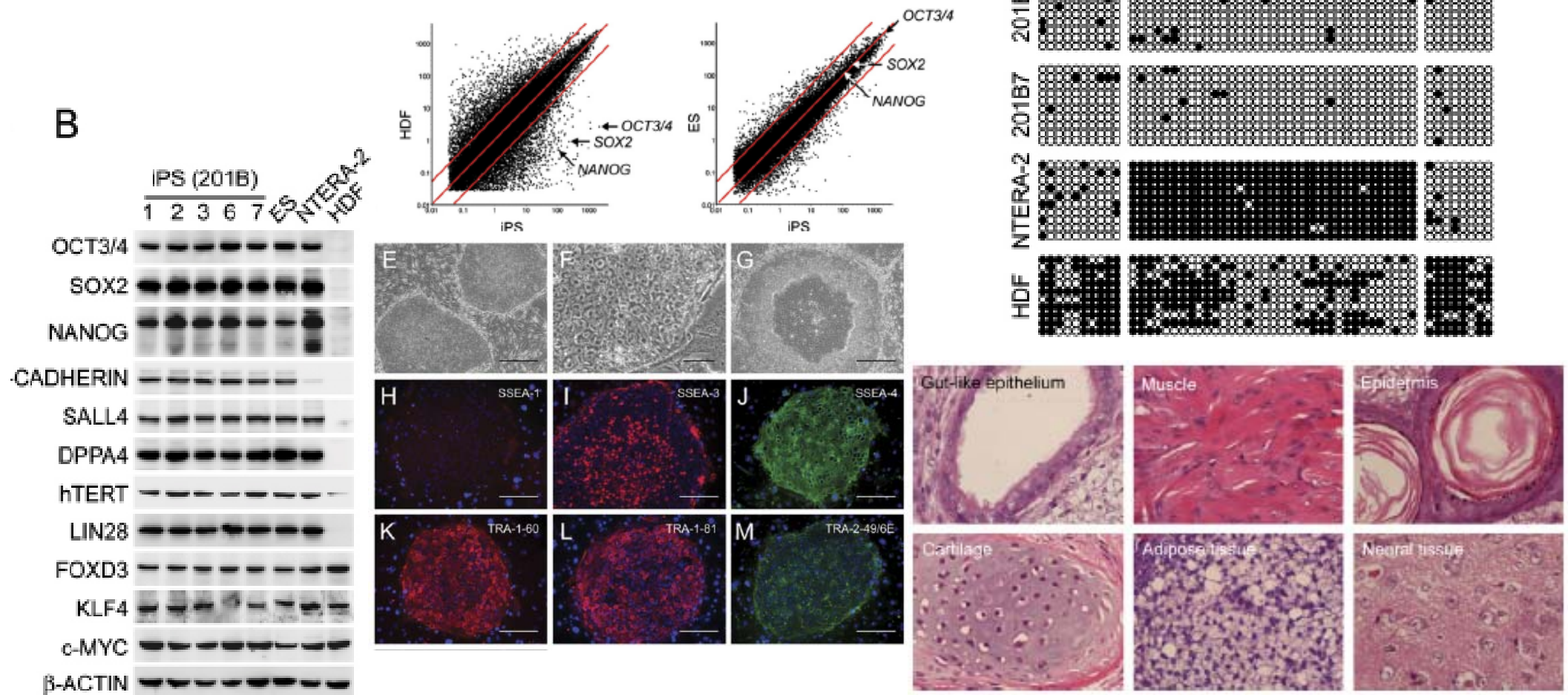
Kazutoshi Takahashi¹ and Shinya Yamanaka^{1,2,*}

We selected 24 genes as candidates for factors that induce pluripotency in somatic cells, based on our hypothesis that such factors also play pivotal roles in the maintenance of ES cell identity (see Table S1 in the Supplemental Data available with this article online). For



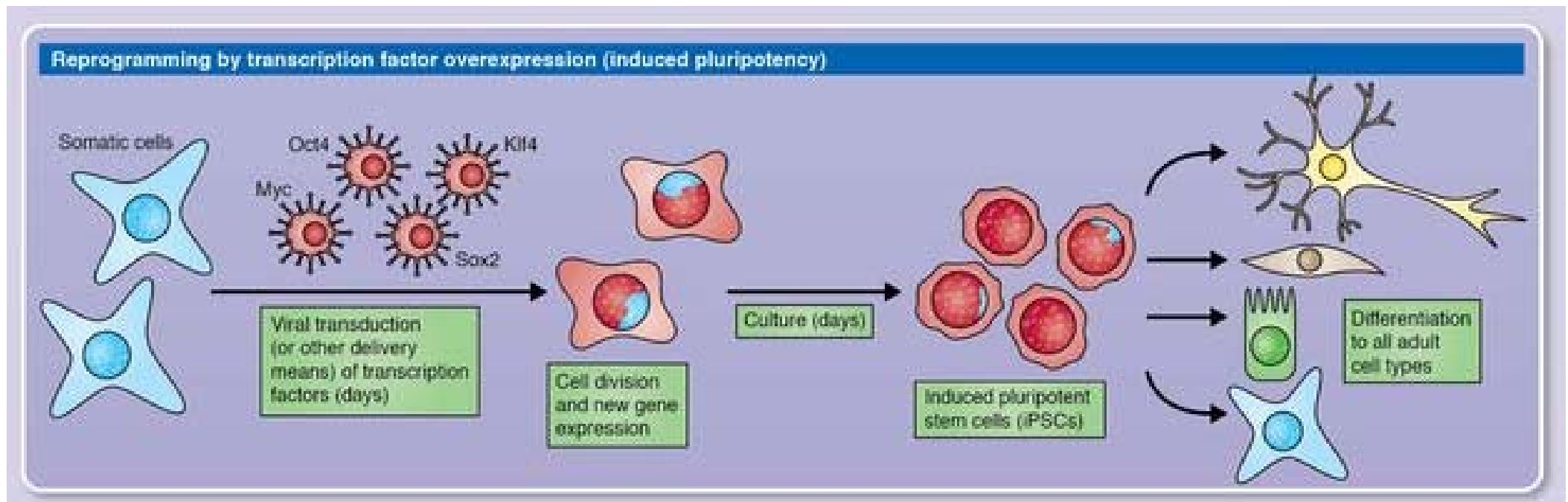
Induction of Pluripotent Stem Cells from Adult Human Fibroblasts by Defined Factors

Cell 131, 861–872, November 30, 2007



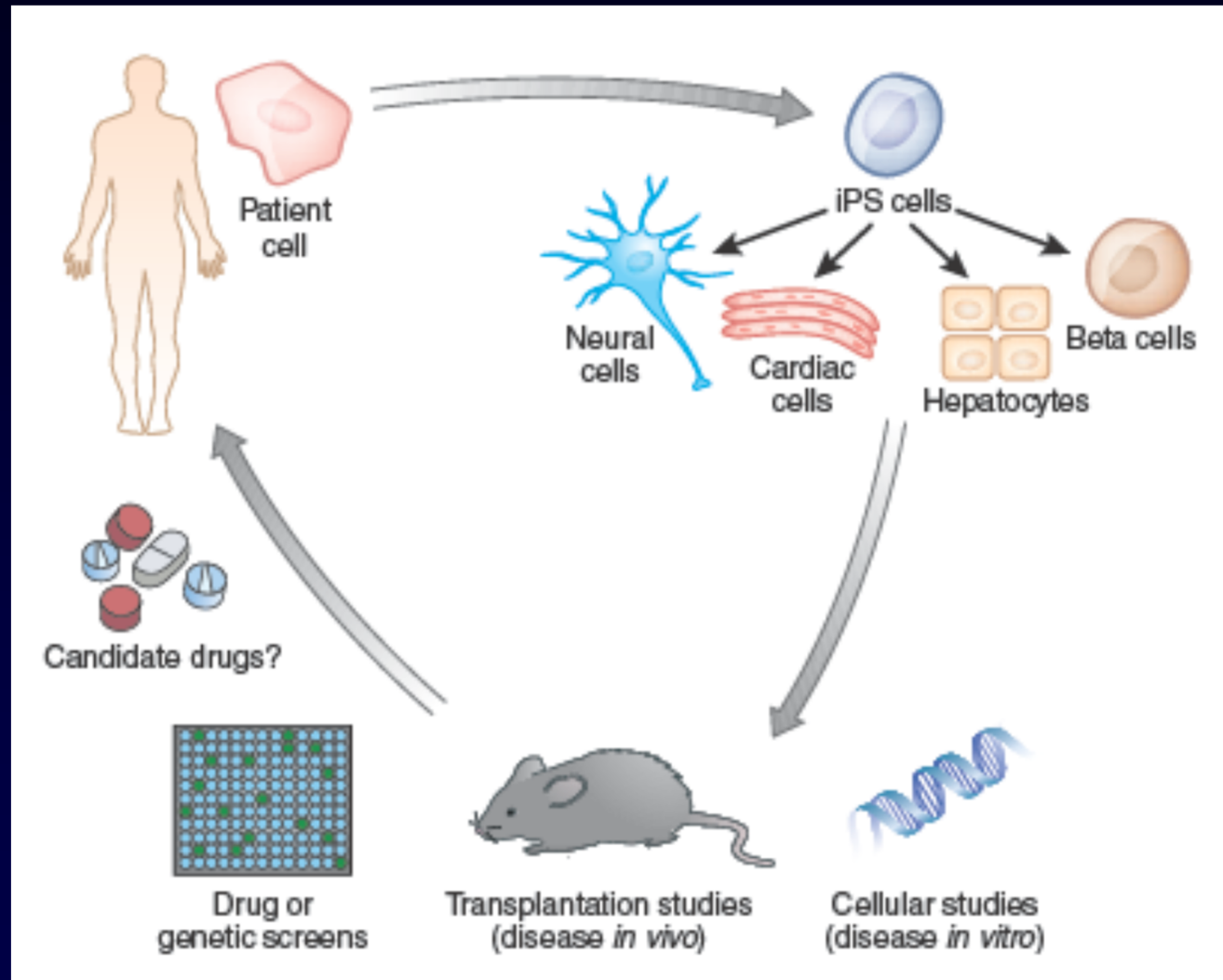
iPS cells are indistinguishable from embryo derived ES cells in morphology, pluripotency, gene expression and epigenetic profiles.

induced pluripotent stem cells (iPS)



Richard P. Halley-Stott et al. *Development* 2013;140:2468-2471

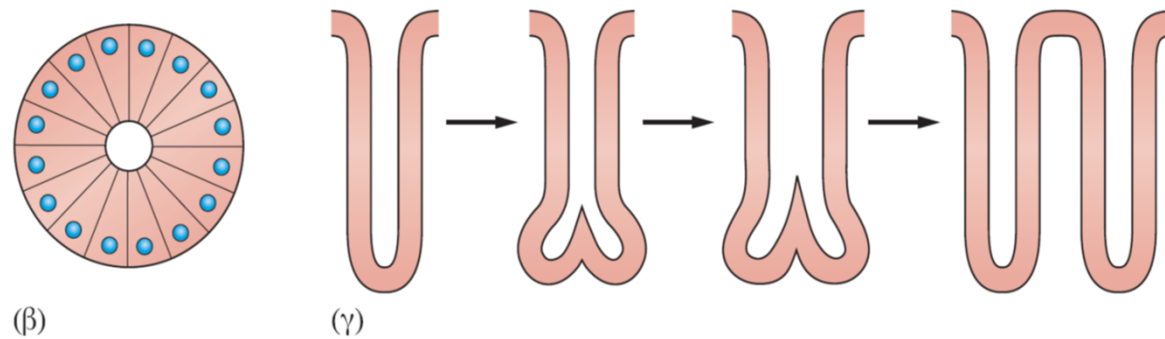
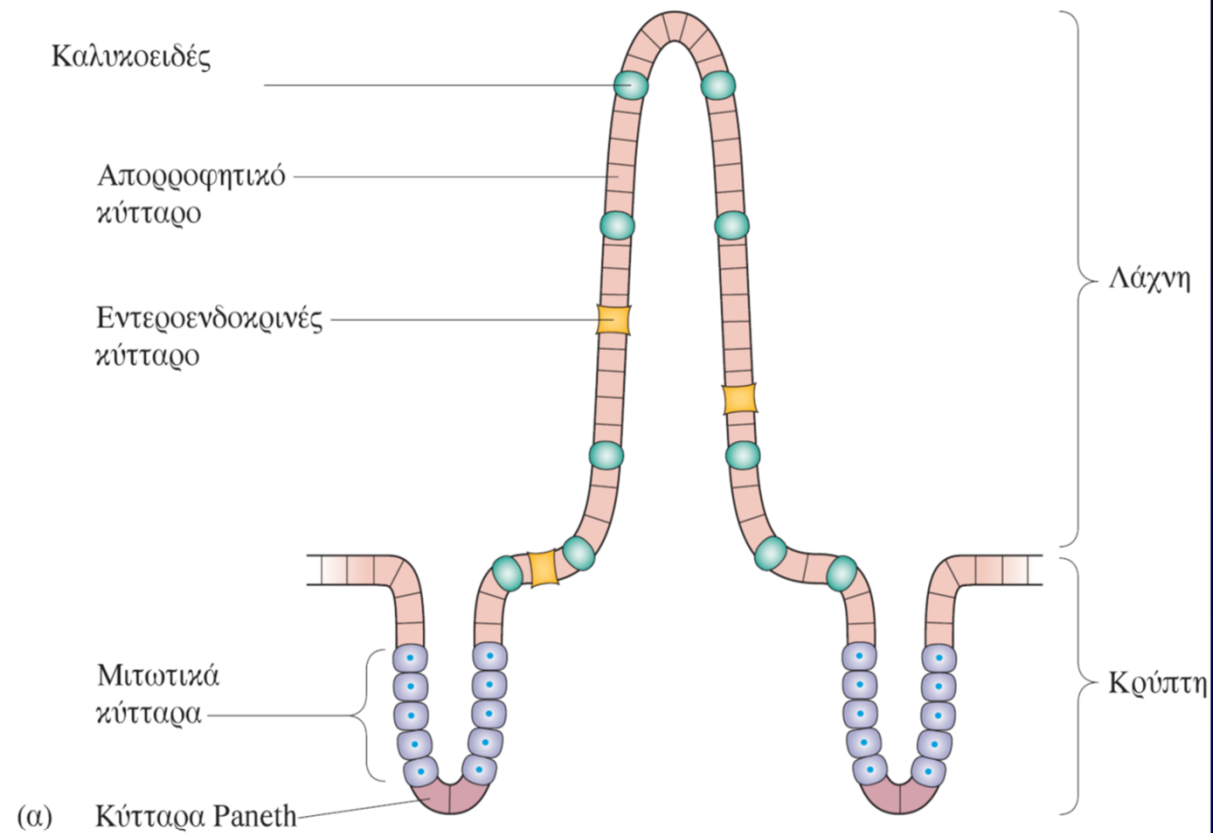
Εφαρμογές Επαγόμενων πολυδύναμων βλαστικών κυττάρων



Σωματικά βλαστικά κύτταρα

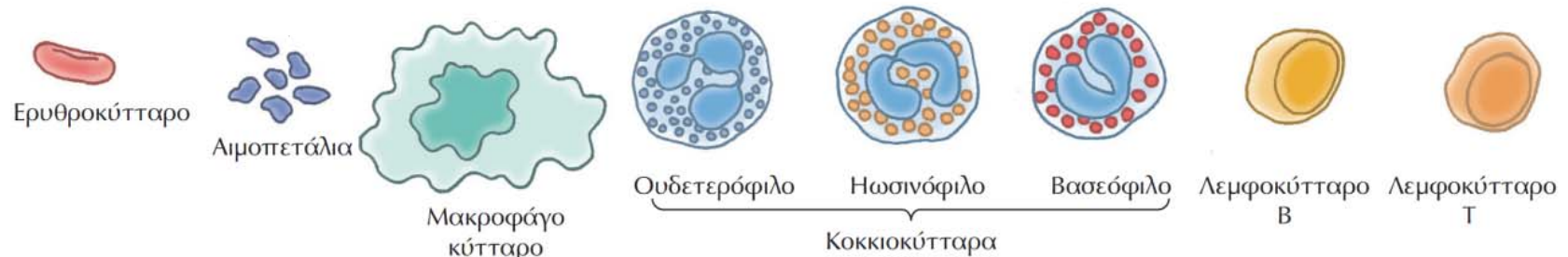
- Τα ενήλικα βλαστικά κύτταρα (ΕΒΚ) είναι μη διαφοροποιημένα κύτταρα που έχουν την ικανότητα αυτό-ανανέωσης ενώ διαφοροποιούνται σε περιορισμένο αριθμό κυτταρικών τύπων

Εντερικό επιθήλιο



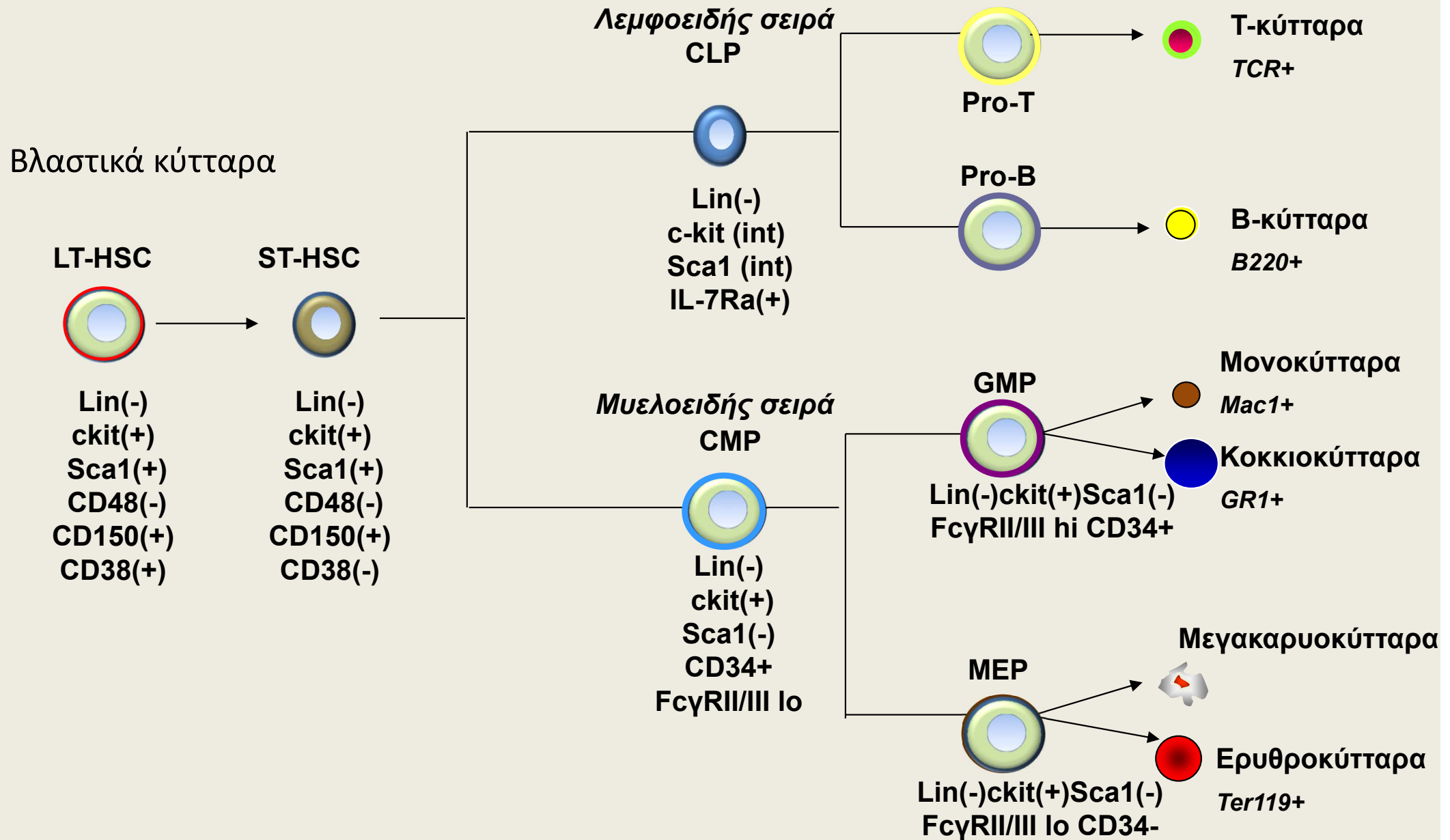
Βλαστικά κύτταρα του αιμοποιητικού συστήματος

Κύτταρα του αιμοποιητικού συστήματος



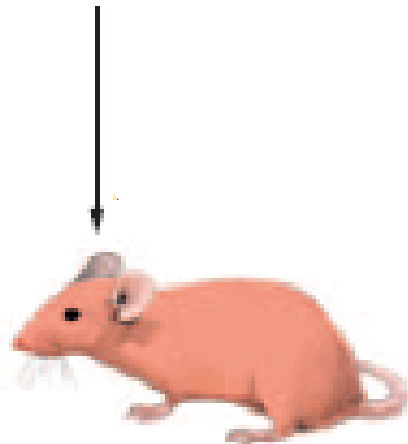
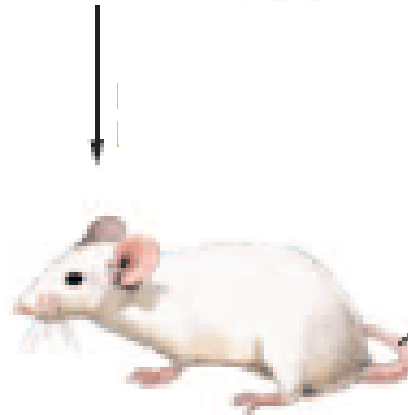
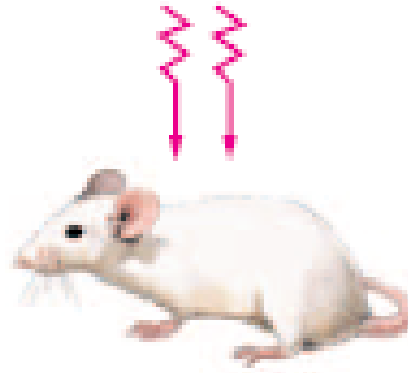
Αιμοποιητικά βλαστικά κύτταρα

Καθορισμός της μοριακής ταυτότητας



Πειράματα ανασύστασης του μυελού των οστών

Ακτινοβολία Χ σταματά την παραγωγή κυττάρων του αίματος και ο μυς θα καταλήξει εάν δεν του δοθεί θεραπεία



Ενεση κυττάρων του μυελού των οστών από έναν υγιή δότη



Ο μυς επιβιώνει καθώς τα αιμοποιητικά βλαστικά κύτταρα του δότη δημιουργούν νέα κύτταρα του αίματος

Η πρώτη προσπάθεια κυτταρικής θεραπείας

The NEW ENGLAND JOURNAL *of* MEDICINE

INTRAVENOUS INFUSION OF BONE MARROW IN PATIENTS RECEIVING RADIATION AND CHEMOTHERAPY*

E. DONNALL THOMAS, M.D.,† HARRY L. LOCHTE, JR., M.D.,‡ WAN CHING LU, PH.D.,§
AND JOSEPH W. FERREBEE, M.D.¶

COOPERSTOWN, NEW YORK, AND BOSTON, MASSACHUSETTS

Η αρχή ... παρουσία ενός πληθυσμού που μπορεί να αναγεννά τον ΜΟ

RADIATION RESEARCH 14, 213-222 (1961)

A Direct Measurement of the Radiation Sensitivity of Normal Mouse Bone Marrow Cells¹

J. E. TILL AND E. A. McCULLOCH

*Department of Medical Biophysics, University of Toronto, and the Divisions of
Biological Research and Physics of the Ontario Cancer Institute,
Toronto, Ontario*

Αλλογενής μεταμόσχευση κυττάρων του μυελού των οστών

THE LANCET

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IMMUNOLOGICAL RECONSTITUTION OF SEX-LINKED LYMPHOPENIC IMMUNOLOGICAL DEFICIENCY

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Χρήση ΟΠΑ για την ανάπλαση του ΜΟ

The NEW ENGLAND JOURNAL *of* MEDICINE

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Originally published: N Engl J Med. 1989 Oct 26;321(17):1174-8. pmid: 2571931

Hematopoietic reconstitution in a patient with Fanconi's anemia by means of umbilical-cord blood from an HLA-identical sibling

**Eliane Gluckman¹, Hal E. Broxmeyer², Arleen D. Auerbach³, Henry S. Friedman⁴, Gordon W. Douglas⁵,
Agnes Devergie¹, Helene Esperou¹, Dominique Thierry⁶, Gerard Socie¹, Pierre Lehn¹, Scott Cooper²,
Denis English², Joanne Kurtzberg⁴, Judith Bard⁷, and Edward A. Boyse⁷**

HSPCs (mouse LSK cells; Human CD34⁺ cells)

Self-renewal

Differentiation

Dormant LT-HSC

Mouse

CD48⁻CD150⁺

CD34⁻CD135⁻CD201⁺

Human

CD34⁺CD38⁻CD90⁺

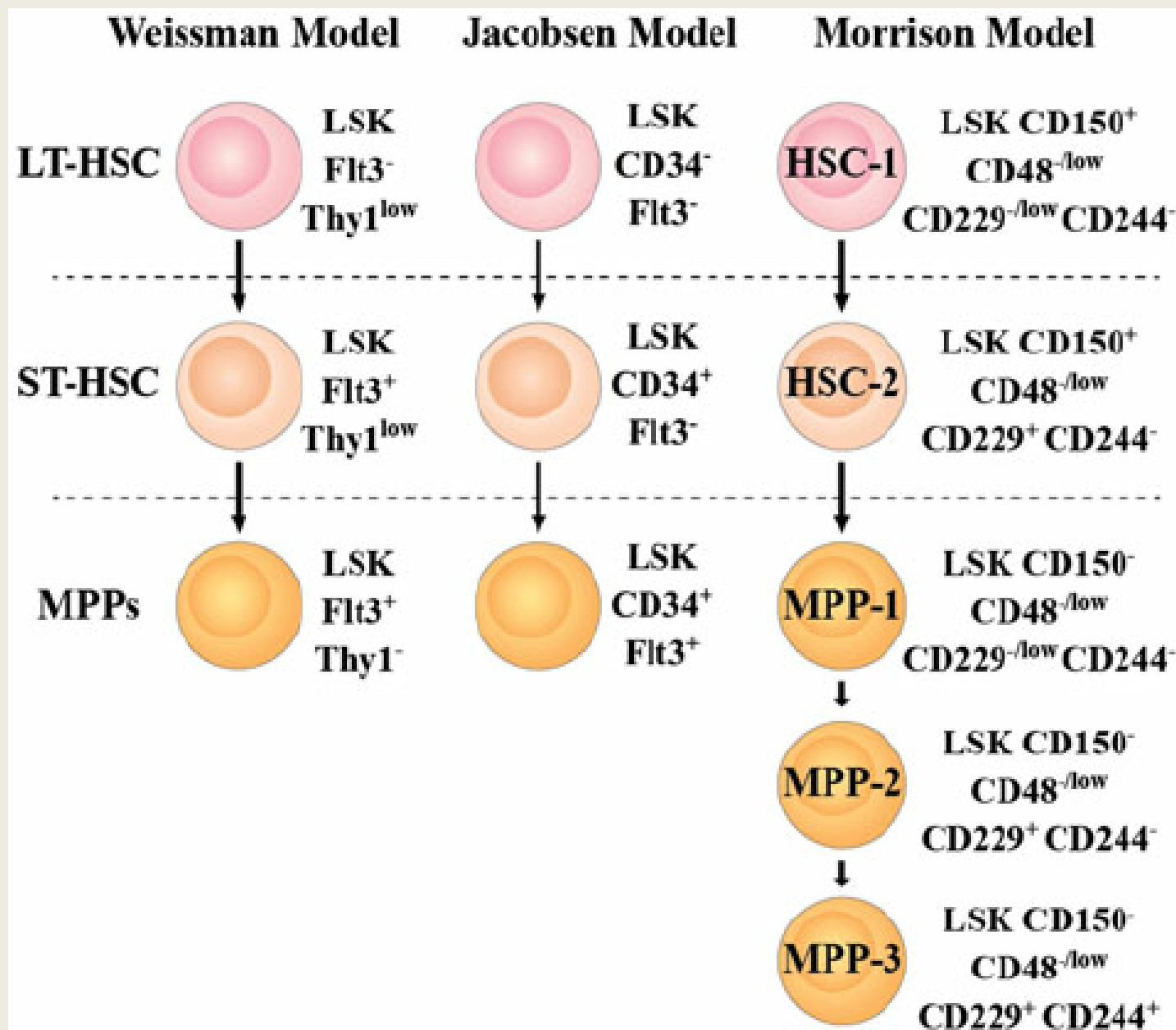
CD45RA⁻CD49f⁺GPI-80⁺



Active ST-HSCs



Differentiation prone MPPs

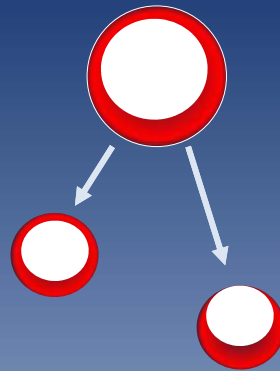


Μεσεγχυματικά βλαστικά κύτταρα «Πηγές»

- Πηγές MSCs
 - Μυελός των οστών
 - Λιπώδης ιστός
 - Βαρτόναιο γέλη του ομφαλίου λώρου
 - Πλακούντας
 - Νεογιλά δόντια-οδοντικός πολφός

Μεσεγχυματικά βλαστικά κύτταρα MSCs

Περικύτταρο



Μεσεγχυματικά Βλαστικά
κύτταρα



Μεσεγχυματικά βλαστικά κύτταρα

Αναγεννητική Δράση

- Παρακρινής δράση-Τροφική δράση
- Ρύθμιση της ανοσοαπάντησης-Αντιφλεγμονώδης δράση
- Διαφοροποίηση σε συγκεκριμένους τύπους κυττάρων

Βλαστικά κύτταρα του νευρικού συστήματος

...“Once the development was ended, the founts of growth and regeneration of the axons and dendrites dried up irrevocably. In the adult centers, the nerve paths are something fixed, ended, and immutable. Everything may die, nothing may be regenerated. It is for the science of the future to change, if possible, this harsh decree.” (1913)...

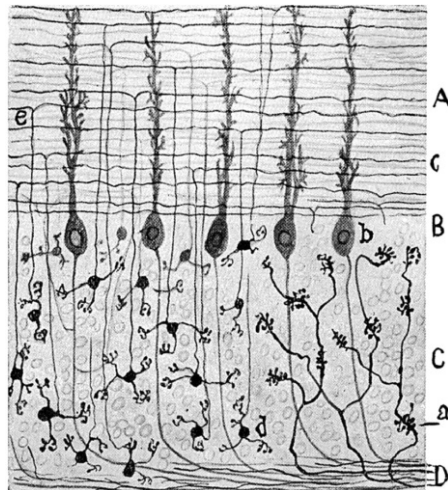
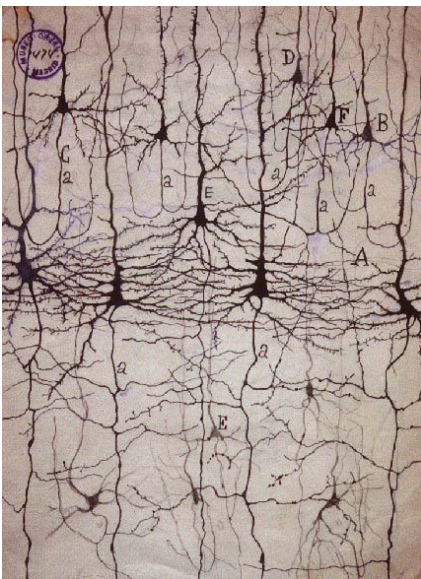
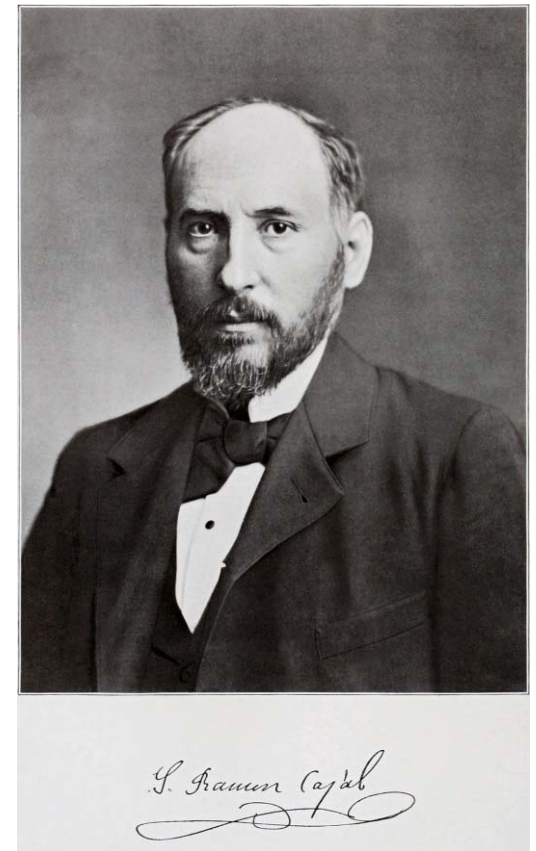


Figure drawn by Cajal (1954)

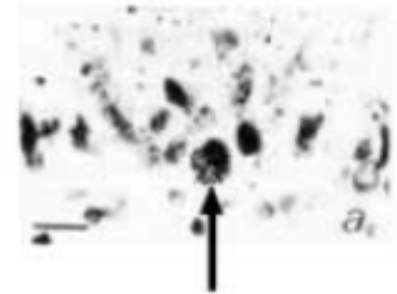
in the late 19th to early 20th century by the prominent histologist Ramon y Cajal:

The broken dogma: Neurogenesis in adult brain

'60s

J Altman

Radioactive thymidine



'70s

M Kaplan

**Radioactive thymidine
electron microscopy**



'80s

F Nottenbohm

**BrdU
neural markers
confocal microscopy**

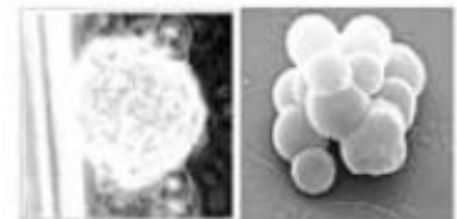


'90s

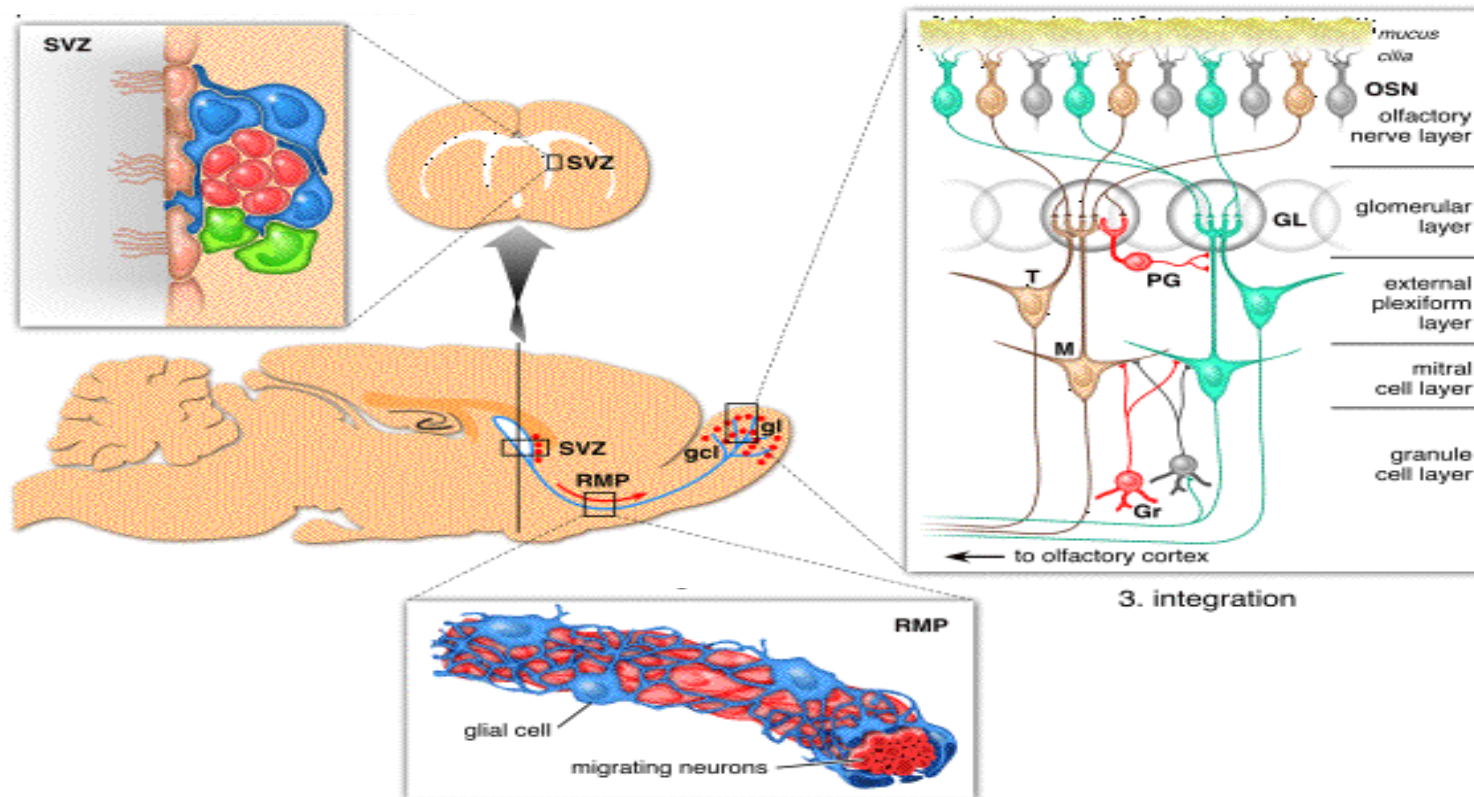
S Weiss

BA Reynolds

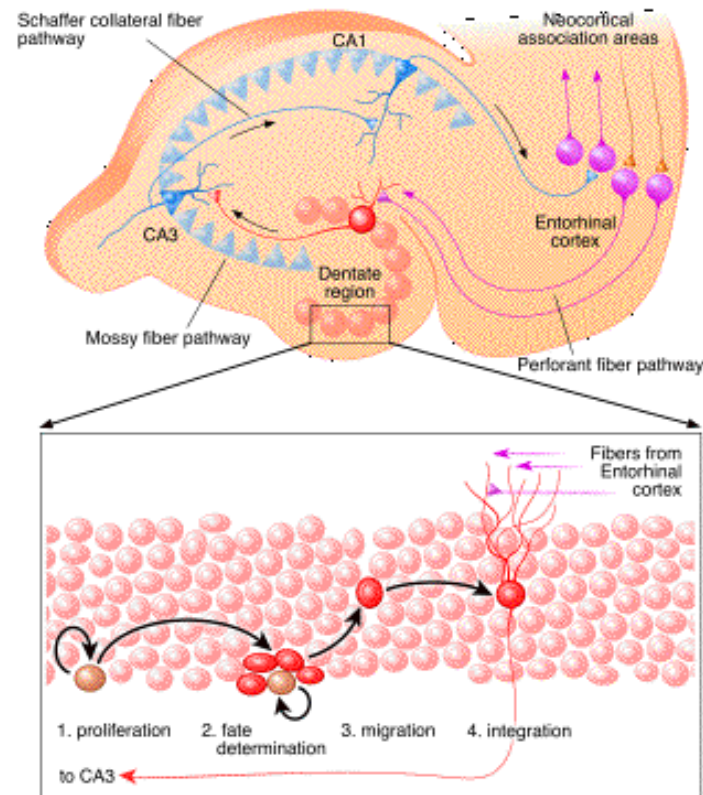
Neurospheres



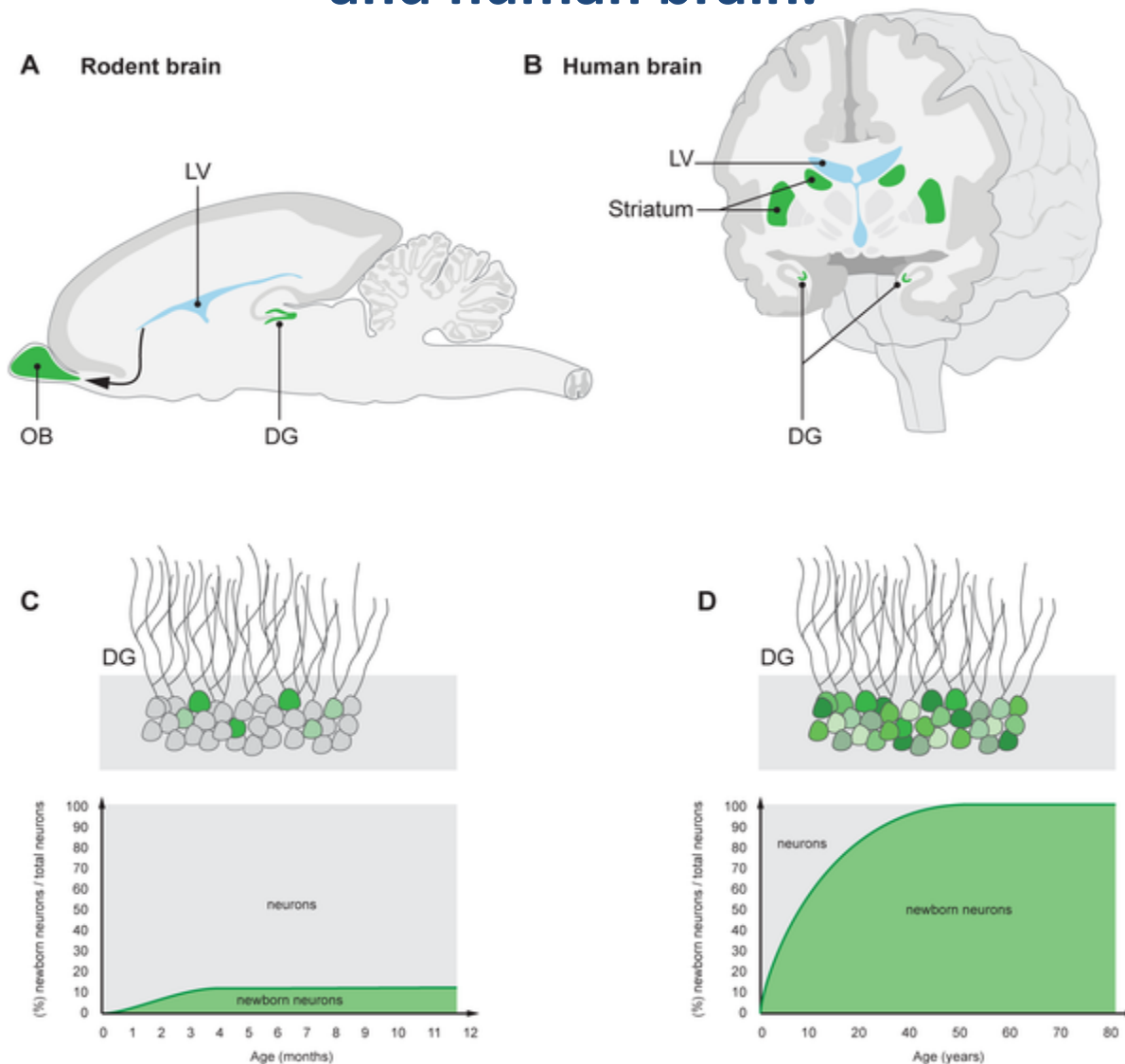
Τα βλαστικά κύτταρα εντοπίζονται στην υποκοιλιακή ζώνη μεταξύ ραβδωτού και πλάγιας κοιλίας



Βλαστικά/πρόδρομα νευρικά κύτταρα στην υποκοκκιώδη στοιβάδα της οδοντωτής έλικας του ιπποκάμπου



Schematic illustration of adult neurogenesis in the adult rodent and human brain.

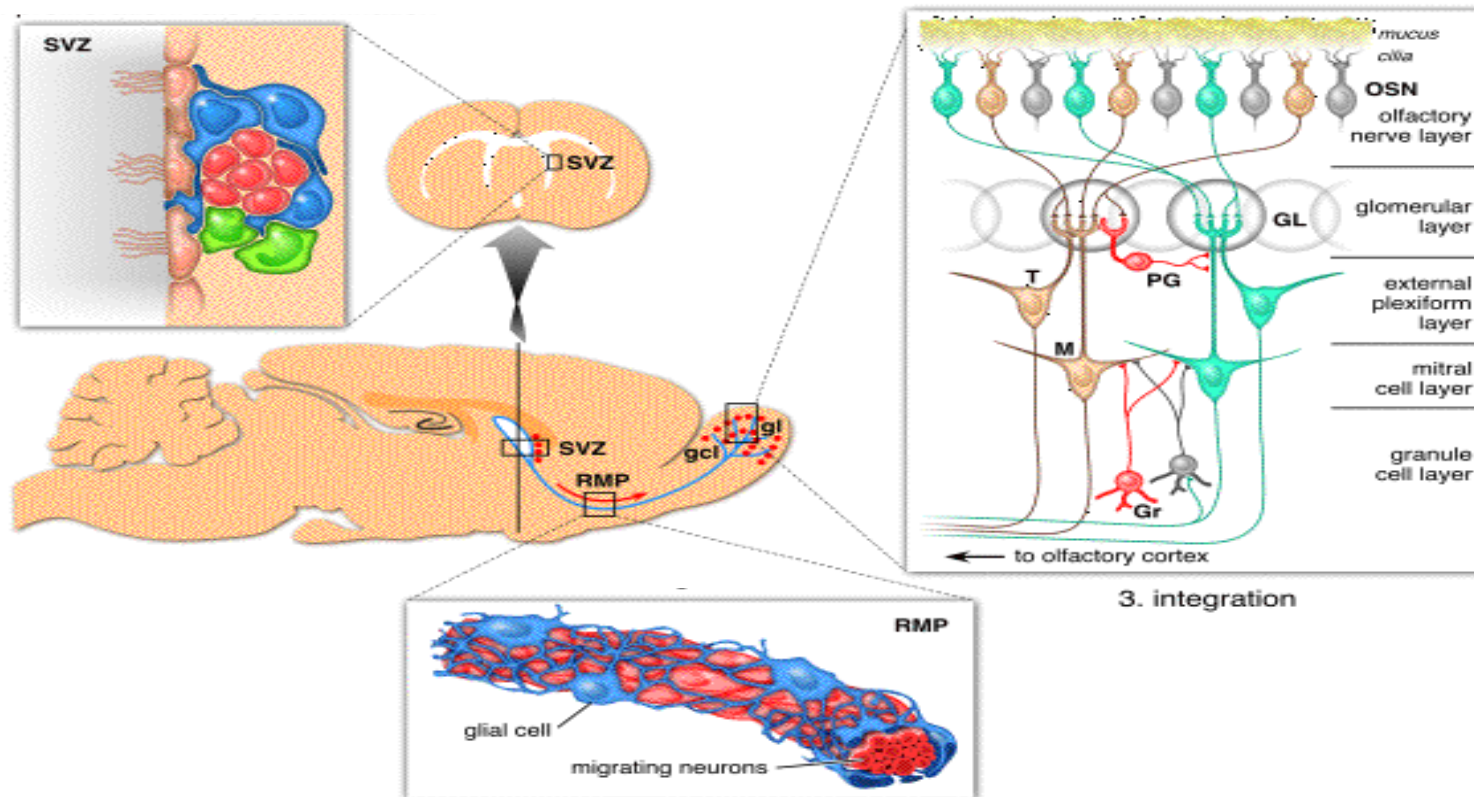


Ernst A, Frisén J (2015) Adult Neurogenesis in Humans- Common and Unique Traits in Mammals. PLoS Biol 13(1): e1002045.

doi:10.1371/journal.pbio.1002045

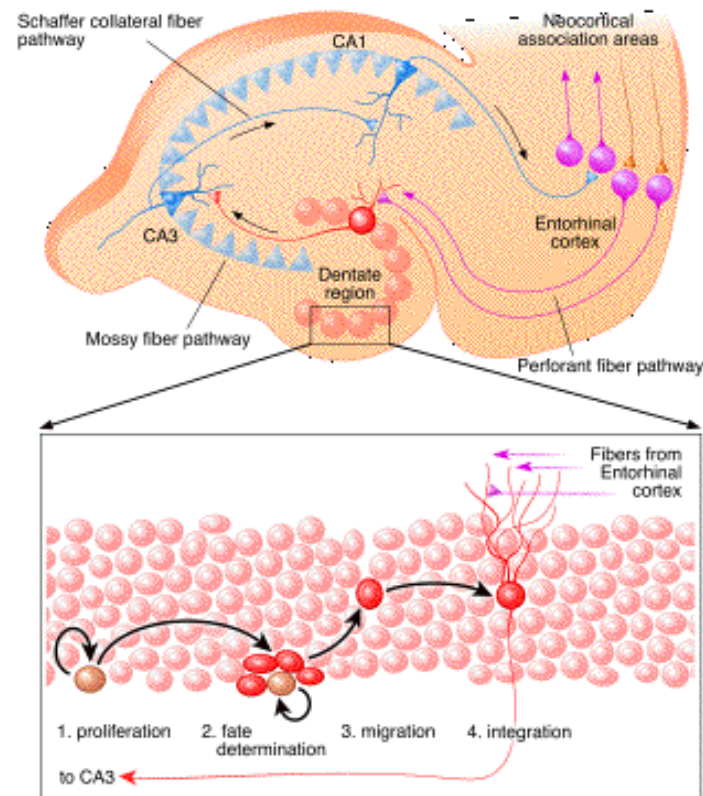
<http://127.0.0.1:8081/plosbiology/article?id=info:doi/10.1371/journal.pbio.1002045>

Τα βλαστικά κύτταρα εντοπίζονται στην υποκοιλιακή ζώνη μεταξύ ραβδωτού και πλάγιας



Αρχικά δίνουν παροδικά πολλαπλασιαζόμενα κύτταρα που μεταναστεύουν στην οσφρητική ταινία

Βλαστικά/πρόδρομα κύτταρα στην υποκοκκιώδη στοιβάδα της οδοντωτής έλικας του ιπποκάμπου

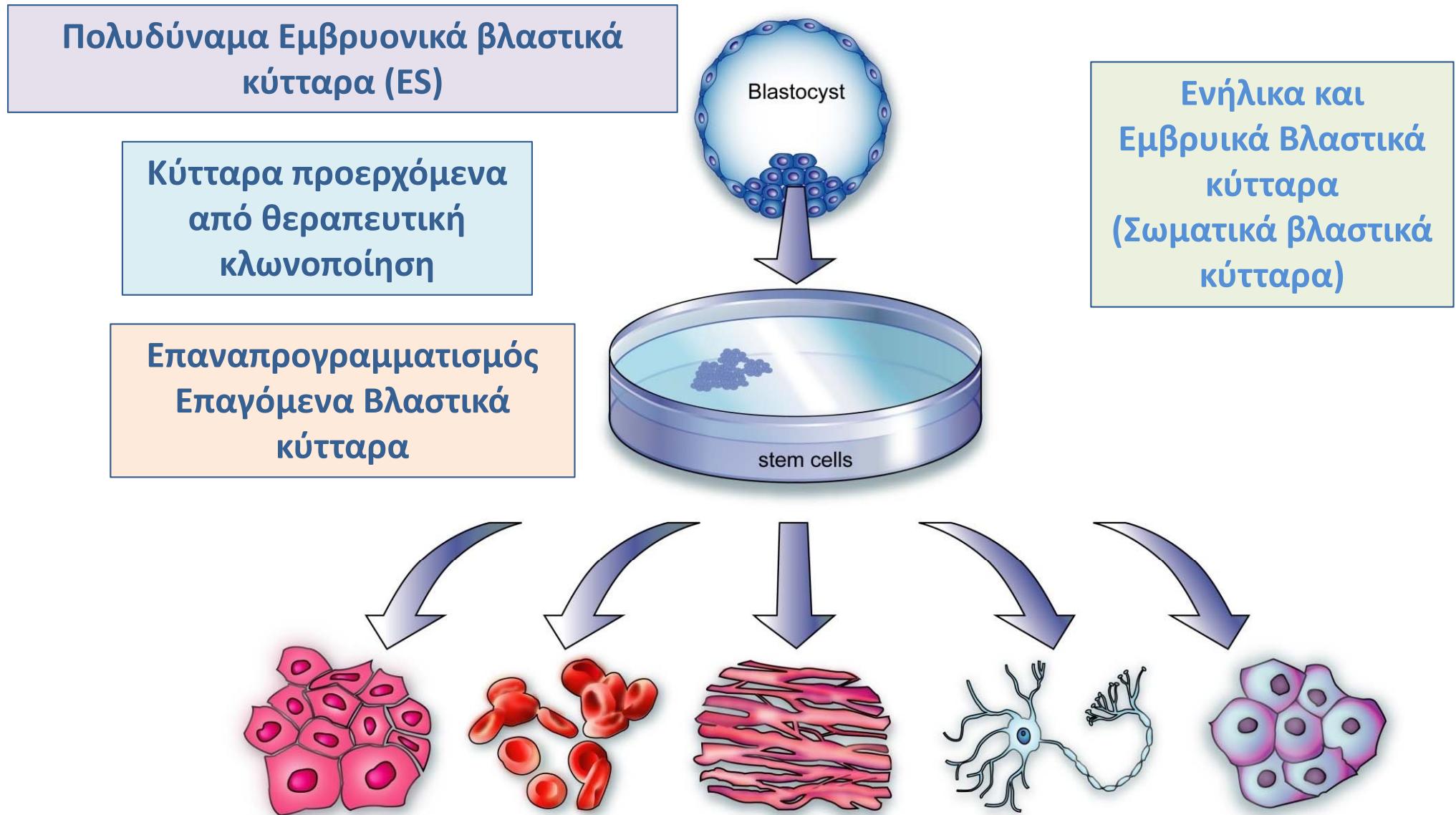


**Θεραπευτικές εφαρμογές βλαστικών
κυττάρων
Αναγεννητική Ιατρική**

Αναγεννητική Ιατρική-Κυτταρικές Θεραπείες

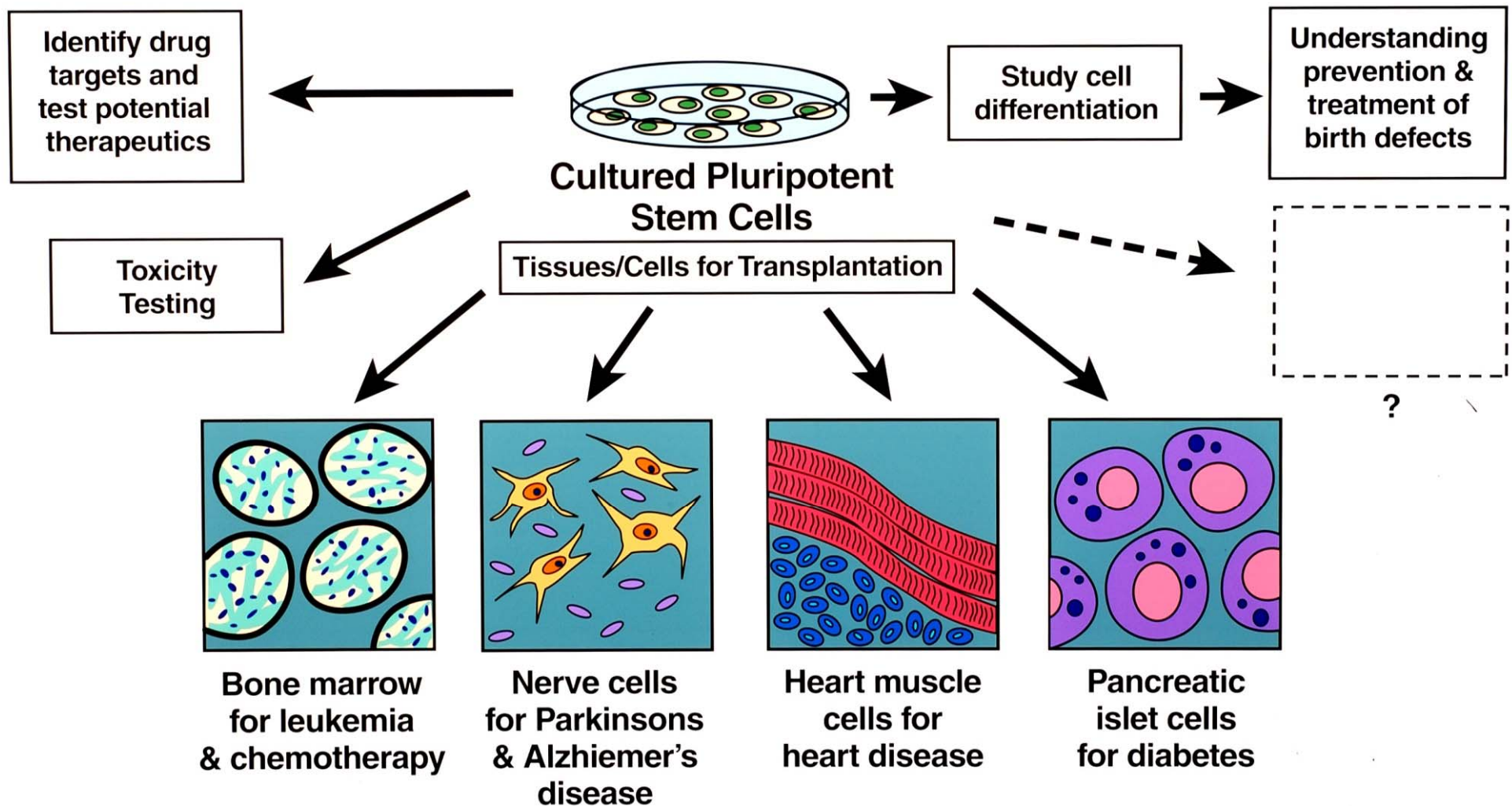
- Επιδιόρθωση/Αντικατάσταση κυττάρων, ιστών, οργάνων
 - Εμβιομηχανική, βιολικά, γονιδιακή θεραπεία
- Κινητοποίηση ενδογενών πληθυσμών βλαστικών κυττάρων

Διαφοροποίηση αρχέγονων/βλαστικών κυττάρων για θεραπευτικούς σκοπούς- -θεραπείες κυτταρικής υποκατάστασης



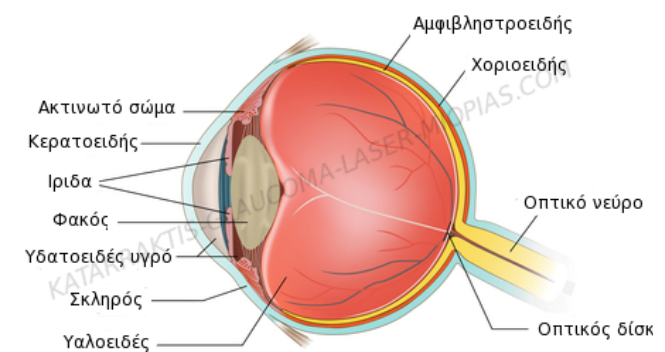
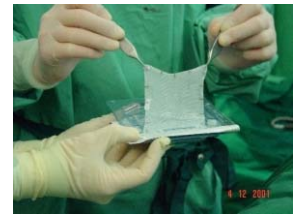
Εφαρμογές

The Promise of Stem Cell Research

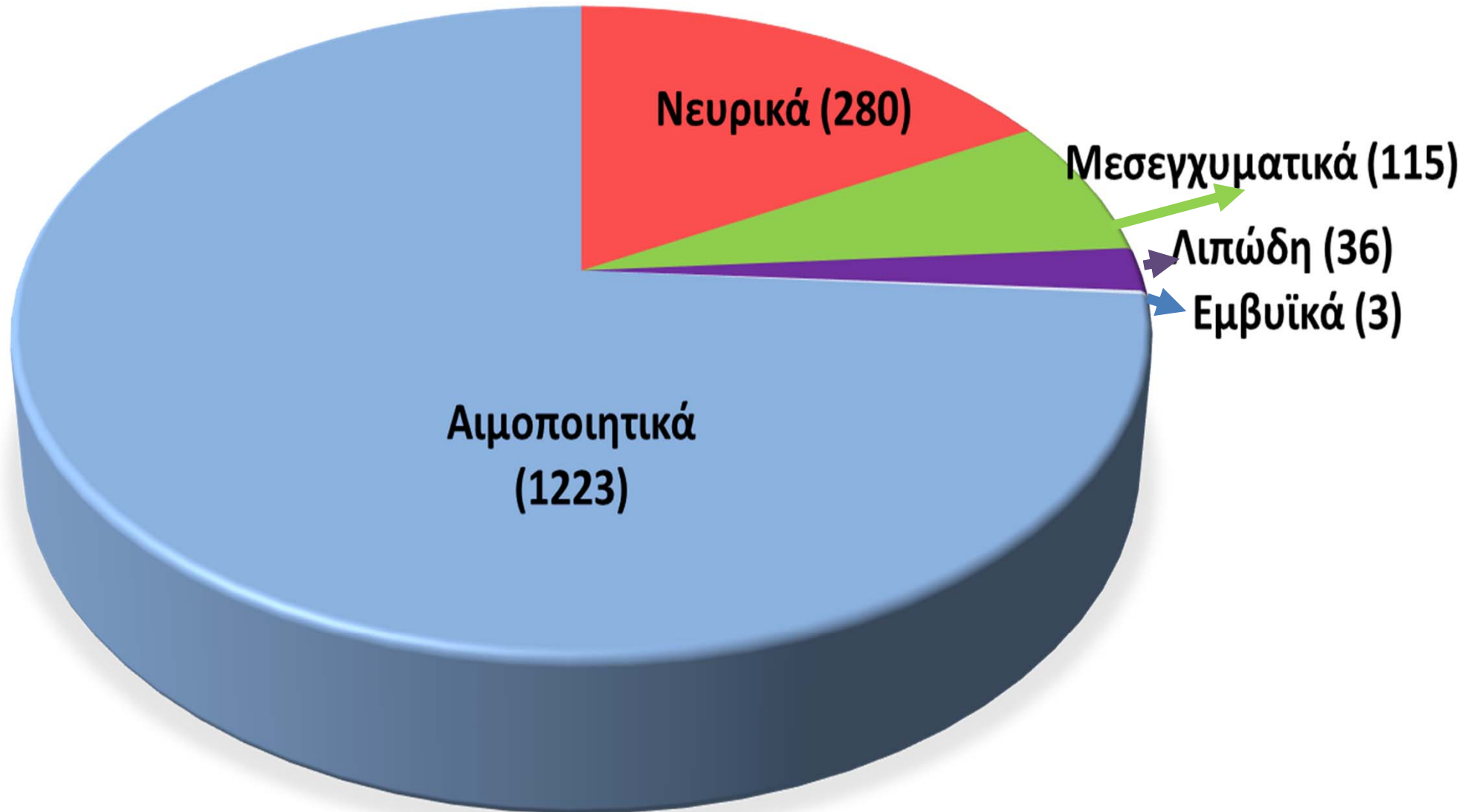


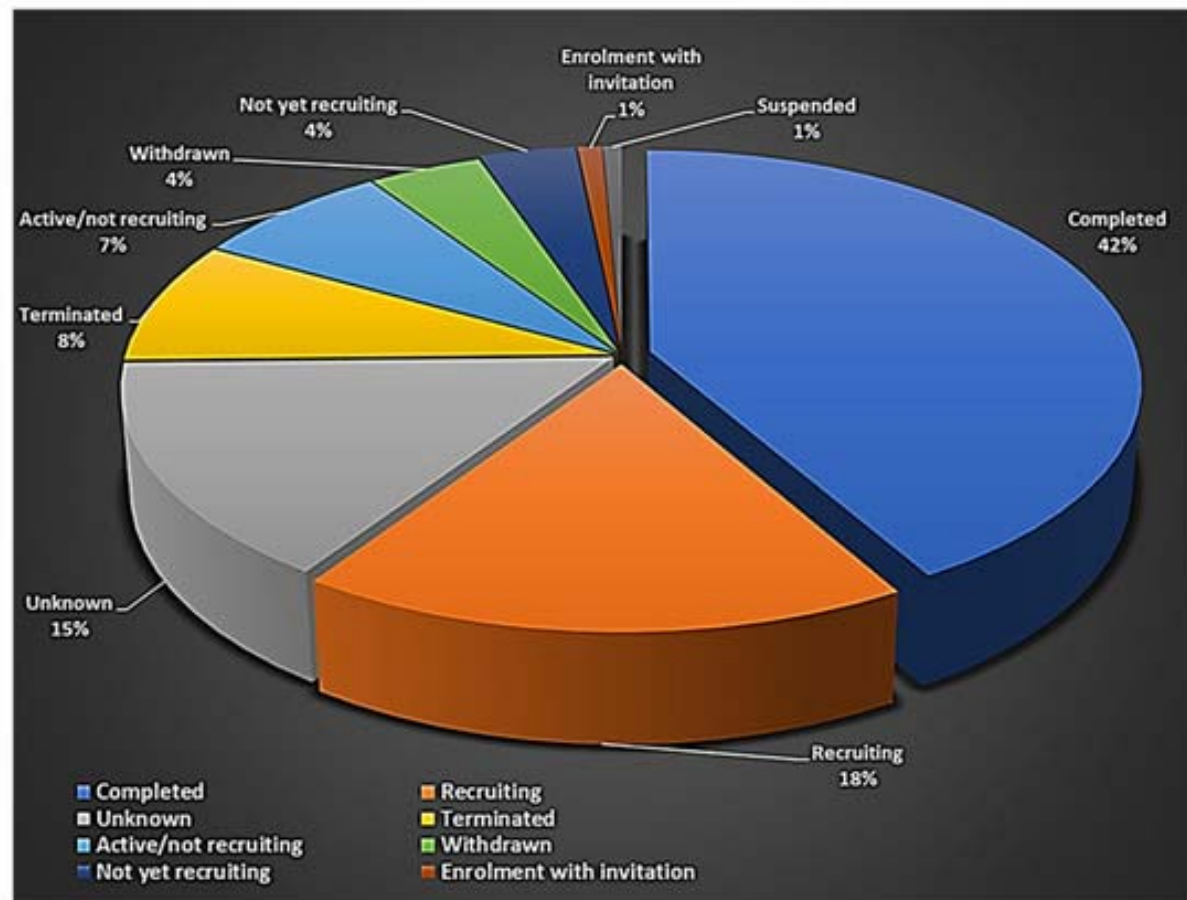
Κυτταρικές Θεραπείες με χρήση βλαστικών κυττάρων

- Βλαστικά κύτταρα του αιμοποιητικού συστήματος
 - Γενετικές ασθένειες, ανοσοανεπάρκειες, κακοήθειες
 - 30.000 αυτόλογες μετασχεύσεις (πολλαπλό μυέλωμα, non-Hodgkin's λέμφωμα) 15.000 αλλογενείς (οξείες λευχαιμίες)
 - Μεταμοσχεύσεις δέρματος σε εγκαυματίες
 - Επιδιόρθωση σοβαρών βλαβών του κερατοειδή χιτώνα του οφθαλμού



Κλινικές δοκιμές με βλαστικά κύτταρα





Neural stem cell transplantation can ameliorate the phenotype of a mouse model of spinal muscular atrophy

Stefania Corti,^{1,2} Monica Nizzardo,¹ Martina Nardini,¹ Chiara Donadoni,¹ Sabrina Salani,¹ Dario Ronchi,¹ Francesca Saladino,¹ Andreina Bordini,¹ Francesco Fortunato,¹ Roberto Del Bo,¹ Dimitra Papadimitriou,¹ Federica Locatelli,² Giorgia Menozzi,² Sandra Strazzer,² Nereo Bresolin,^{1,2,3} and Giacomo P. Comi^{1,2}

¹Cino Ferrari Centre, Department of Neurological Sciences, University of Milan and IRCCS Foundation Ospedale Maggiore Policlinico, Mangiagalli and Regina Elena, Milan, Italy; ²Centre of Excellence on Neurodegenerative Diseases, University of Milan, Milan, Italy; ³IRCCS Eugenio Medea, Brescia/Pavini, Lecco, Italy.

Spinal muscular atrophy (SMA), a motor neuron disease (MND) and one of the most common genetic causes of infant mortality, currently has no cure. Patients with SMA exhibit muscle weakness and hypotonia. Stem cell transplantation is a potential therapeutic strategy for SMA and other MNDs. In this study, we isolated spinal cord neural stem cells (NSCs) from mice expressing green fluorescent protein only in motor neurons and assessed their therapeutic effects on the phenotype of SMA mice. Intrathecally grafted NSCs migrated into the parenchyma and generated a small proportion of motor neurons. Treated SMA mice exhibited improved neuromuscular function, increased lifespan, and improved motor unit pathology. Global gene expression analysis of laser-capture-microdissected motor neurons from treated mice showed that the major effect of NSC transplantation was modification of the SMA phenotype toward the wild-type pattern, including changes in RNA metabolism proteins, cell cycle proteins, and actin-binding proteins. NSC transplantation positively affected the SMA disease phenotype, indicating that transplantation of NSCs may be a possible treatment for SMA.

Introduction

Spinal muscular atrophy (SMA) is an autosomal recessive motor neuron disease (MND) and is the second most common genetic disorder leading to death in childhood (1). SMA is caused by deletions or mutations in the telomeric copy of the survival motor neuron 1 gene (*SMN2*) (2), leading to the depletion of survival motor neuron (*SMN*) protein available for SMA, and using the mouse *Smn* gene and an additional *Smn* transgene to generate *SMN2^{+/+}SMN4^{+/+}* mice that exhibit a type 1 SMA phenotype (4). SMA animal models are useful for studying mechanisms of motor neuron loss and for testing potential therapeutic strategies.

Stem cell transplantation for MNDs such as SMA could improve the disease by multiple mechanisms: compensating for the loss of neurons, the delivery of neurotrophic factors (5), the reduction of

excitotoxicity (6), and the replacement of other, non-neuronal cells (7). However, the precise identity of these mechanisms has remained an open question. Transplantation of NSCs in SOD1G93A mice, an animal model of amyotrophic lateral sclerosis (ALS), modifies disease progression through both neurogenesis and growth factor release (8). We have previously described the isolation of a subset of self-

renewing NSCs from the spinal cord of *SMN2^{+/+}SMN4^{+/+}* mice and their transplantation into *SMN2^{+/+}SMN4^{+/+}* mice. We found that (a) ALDH⁺SSC⁺ NSCs generate a small amount of motor neurons, (b) transplantation improves motor unit survival of SMA mice; and (c) the pattern of endogenous gene expression in SMA mice is modified toward the wild-type profile. Our data indicate that NSC transplantation has a positive effect on

Nonstandard abbreviations

amyotrophic lateral sclerosis; BDNF, brain-derived neurotrophic factor; ChAT, choline acetyltransferase; GFAP, glial fibrillary acidic protein; MAP2, microtubule-associated protein-2; MND, motor neuron disease; NSC, neural stem cell; SSC, side scatter; SMA, spinal muscular atrophy; *Smn*, survival motor neuron; *SMN2^{+/+}SMN4^{+/+}*, *SMN2* cDNA lacking exon 7; TA, tibialis anterior.

Conflict of interest: The authors have declared that no conflict of interest exists.

Citation for this article: *J Clin Invest*. 118:3316–3330 (2008). doi:10.1172/JCI35432.

ALDH⁺SSC⁺ NSCs prolong the life and improve the disease phenotype of SMA mice. To examine the ability of NSCs to modify the disease progression of SMA mice, we used a population of these

Human Embryonic Germ Cell Derivatives Facilitate Motor Recovery of Rats with Diffuse Motor Neuron Injury

Douglas A. Kerr,¹ Jerònia Lladó,¹ Michael J. Shambloott,² Nicholas J. Maragakis,¹ David N. Irani,¹ Thomas O. Crawford,¹ Chitra Krishnan,¹ Sonny Dike,¹ John D. Gearhart,² and Jeffrey D. Rothstein¹

Departments of ¹Neurology and ²Gynecology and Obstetrics, Johns Hopkins University School of Medicine, Baltimore, Maryland 21287

We have investigated the potential of human pluripotent cells to restore function in rats paralyzed with a virus-induced motor neuronopathy. Cells derived from embryonic germ cells, termed embryoid body-derived (EBD) cells, introduced into the CSF were distributed extensively over the rostrocaudal length of the spinal cord and migrated into the spinal cord parenchyma in paralyzed, but not uninjured, animals. Some of the transplanted human cells expressed the neuroglial progenitor marker nestin, whereas others expressed immunohistochemical markers characteristic of astrocytes or mature neurons. Rare transplanted cells developed immunoreactivity to choline acetyltransferase (ChAT) and sent axons into the sciatic nerve as detected by retrograde labeling. Paralyzed animals transplanted with EBD cells partially recovered motor function 12 and 24 weeks after transplantation, whereas control animals remained paralyzed. Semi-quantitative analysis revealed that the efficiency of neuronal differentiation and extension of neurites could not account for the functional recovery. Rather, transplanted EBD cells protected host neurons from death and facilitated reafferentation of motor neuron cell bodies. *In vitro*, EBD cells secrete transforming growth factor- α (TGF- α) and brain-derived neurotrophic factor (BDNF). Neutralizing antibodies to TGF- α and to BDNF abrogated the ability of EBD-conditioned media to sustain motor neuron survival in culture, whereas neutralizing antibodies to BDNF eliminated the axonal outgrowth from spinal organotypics observed with direct coculture of EBD cells. We conclude that cells derived from human pluripotent stem cells have the capacity to restore neurologic function in animals with diffuse motor neuron disease via enhancement of host neuron survival and function.

Key words: motor neuron disease; Sindbis virus; stem cells; embryonic germ cell; trophic factors; embryoid body



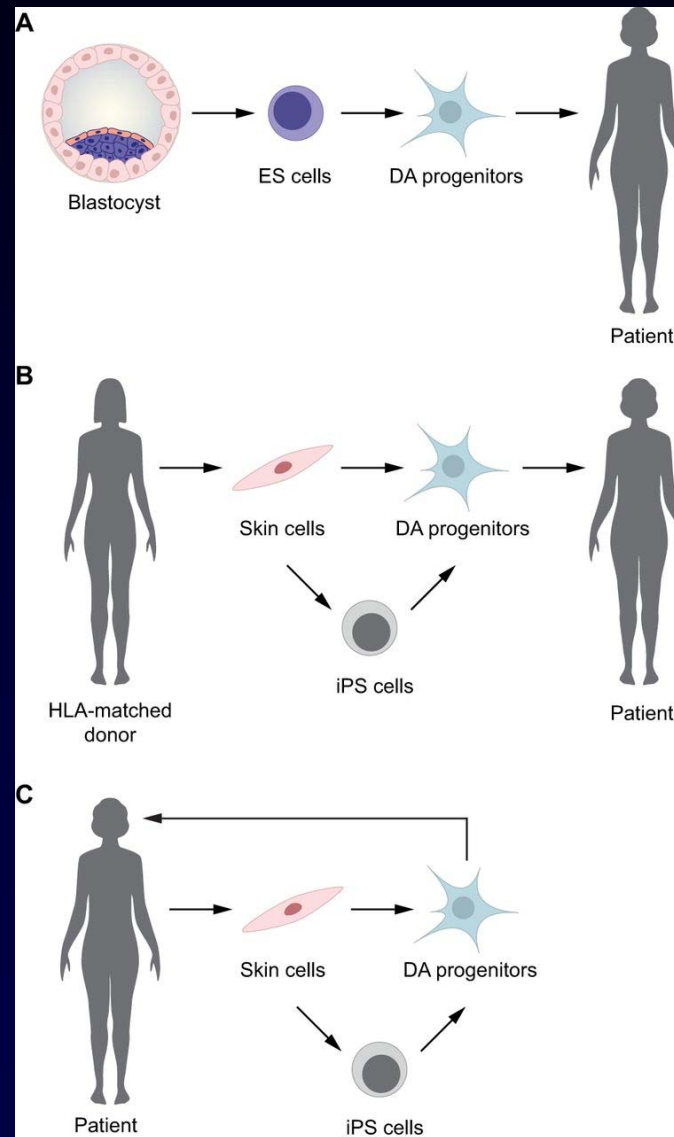
a rat paralyzed with the Sindbis virus.



paralyzed rat also 6 months after receiving a transplant.

Κλινικές εφαρμογές

Therapeutic cells for cell-based therapy.



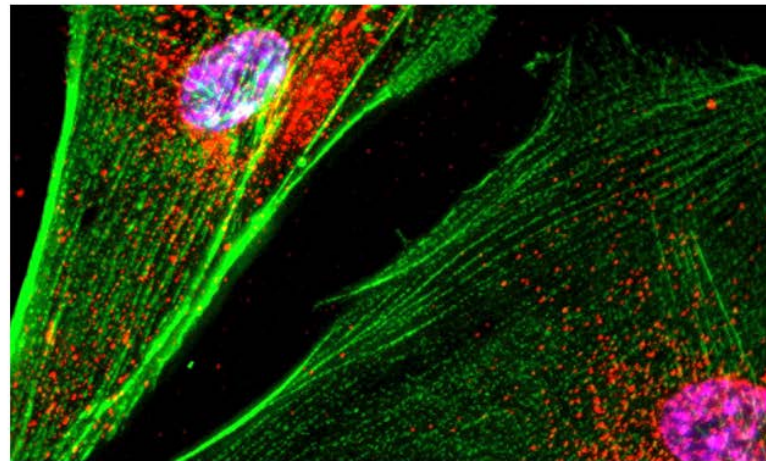
Malin Parmar Development 2018;145:dev156117

Reprogrammed cells relieve Parkinson's symptoms in trials

Monkeys implanted with neurons derived from stem cells showed sustained improvement after two years.

Ewen Callaway

30 August 2017



B. Bick, J. Pindexter, UT Med. School/SPL

A depletion of brain cells that produce dopamine is responsible for the mobility problems seen in people with Parkinson's disease.

Japanese researchers report promising results from an experimental therapy for Parkinson's disease that involves implanting neurons made from 'reprogrammed' stem cells into the brain. A trial conducted in monkeys with a version of the disease showed that the treatment improved their symptoms and seemed to be safe, according to a report published on 30 August in *Nature*¹.

The study's key finding — that the implanted cells survived in the brain for at least two years without causing any dangerous effects in the body — provides a major boost to researchers' hopes of testing stem-cell treatments for Parkinson's in humans, say scientists.

Jun Takahashi, a stem-cell scientist at Kyoto University in Japan who led the study, says that his team plans to begin transplanting neurons made from induced pluripotent stem (iPS) cells into people with Parkinson's in clinical trials soon.

The research is also likely to inform several other groups worldwide that are testing different approaches to treating Parkinson's using stem cells, with trials also slated to begin soon.



How iPS cells changed the world

Nature breaks down the latest research — and what it means for the future of stem-cell treatments.

<https://www.nature.com/news/reprogrammed-cells-relieve-parkinson-s-symptoms-in-trials-1.22531>

LETTER

doi:10.1038/nature23664

Human iPS cell-derived dopaminergic neurons function in a primate Parkinson's disease model

Tetsuhiro Kikuchi¹, Asuka Morizane¹, Daisuke Doi¹, Hiroaki Magotani¹, Hirotaka Onoe², Takuya Hayashi², Hiroshi Mizuma², Sayuki Takara², Ryosuke Takahashi³, Haruhisa Inoue⁴, Satoshi Morita⁵, Michio Yamamoto⁵, Keisuke Okita⁶, Masato Nakagawa⁶, Malin Parmar⁷ & Jun Takahashi^{1,8}

Box 1. Summary of pre-clinical assessments

Summary of key parameters measured in the three major steps for taking stem cell-derived dopaminergic neurons to clinical trial. These include: (A) monitoring of the cell product to ensure cells manufactured for clinical use are of sufficient quality and quantity, without any contaminating pluripotent cells and with minimal contamination of forebrain and hindbrain progenitors; (B) *in vivo* testing to ensure that cells used for grafting are safe; and (C) ensuring that cells have the ability to survive and mature *in vivo*. The most-important features are that they can mediate functional recovery in dopamine-depletion models and are able project long distances such that innervation is sufficient for use in humans.

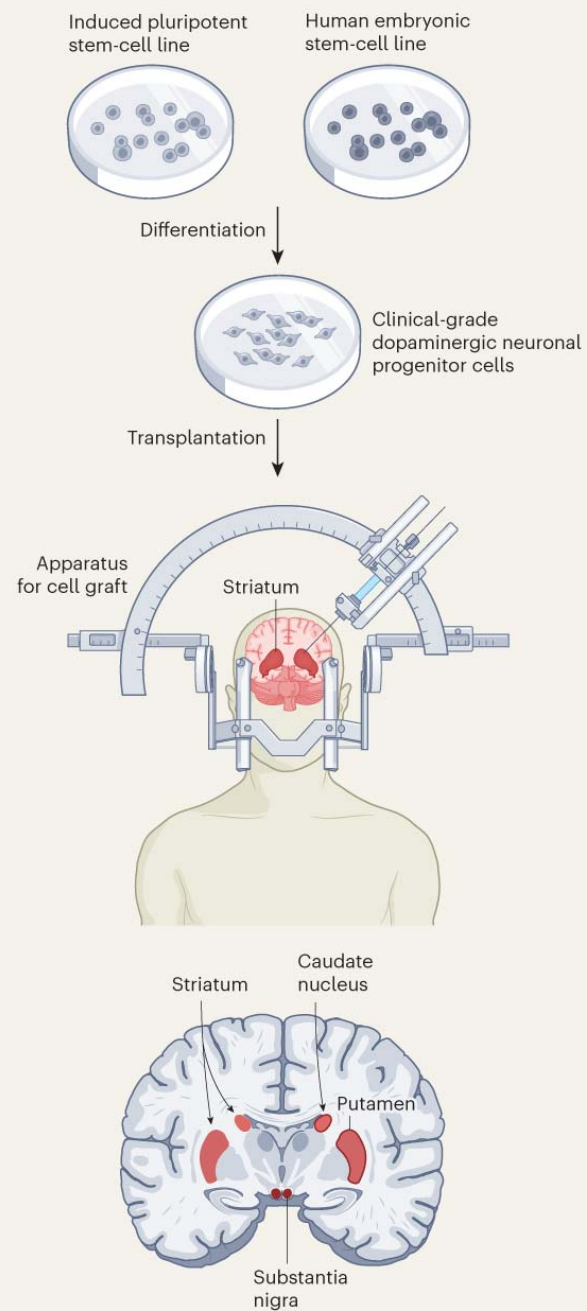
(A) Manufacturing process	(B) Safety	(C) Efficacy
• In process Quality Control (QC) and Release Criteria (RC) to ascertain quality of cells and comparability between batches Eighty percent of cells co-expressing midbrain markers such as FOXA2, LMX1A, EN1 and CORIN Absence of pluripotency markers Low expression of non-neural markers Low numbers of forebrain and hindbrain progenitors (i.e. PAX6⁺, FOXG1⁺, GBX2⁺) (Kirkeby et al., 2017b; Studer, 2017; Takahashi, 2017)	• Tumorigenicity Rule out pluripotent cells or other contaminants <i>in vitro</i> and ensure no tumor formation <i>in vivo</i> can be done in nude mice or rats at certified contract research organization • Biodistribution Standard <i>in vivo</i> testing at certified CRO • Toxicology Standard <i>in vivo</i> testing at certified CRO (Kirkeby et al., 2017b; Studer, 2017; Takahashi, 2017)	• Yield Estimate the number of mature dopaminergic neurons obtained per 100,000 grafted cells • Ability to restore motor function Full recovery of amphetamine-induced rotations in 6-hydroxydopamine lesion model • Long-range target specific innervation Intranigral grafting in rat model, or intrastriatal grafting in a large animal model (Barker et al., 2017; Grealish et al., 2014; Kikuchi et al., 2017)

Table 1. Main Feature Summary of GForce-PD Partners' Clinical Trials

	EUROPEAN STEM-PD ^a	NYSTEM-PD	CiRA Trial	Summit for PD Trial
Cell source	ESCs	ESCs	allogeneic iPSCs	autologous iPSCs
Cryopreserved cell product?	yes	yes	no	yes
Genetic testing of cell product	TBD	karyotype / + TBD	sequencing for certain genes	full genome sequencing
Cell delivery method	"Rehncrona" instrument previously used in fetal VM trials	MRI/Clearpoint system	purpose made needle	MRI/Clearpoint system
Dosing?	low dose, high dose	low dose, high dose	one dose	low dose, high dose
Immunosuppressive regime	yes, at least 12 months; probably CiclosporinA; Azathioprine; steroids	yes, 12 months; FK506; Basiliximab; TBD ± mycophenolate	yes, 1–2 years; FK506	none
Patient characteristics				
Age	<70 years old	40–70 years old	50–70 years old	45–70 years
Disease duration	<12 years	5–12 years	>5 years	>5 years
Significant LIDs?	no	no	no	no
L-dopa response?	>30%	>50%	>30%	>20%
Pre-transplant run-in period	>1 year	>1 year	TBD	>1 year
Follow-up period	indefinitely	at least 2 years	at least 2 years	at least 1 year
PET imaging	F-dopa; PE2i	F-dopa; PE2i; DPA 713	F-dopa; DAT-SPECT; FLT GE180	F-dopa; DAT-SPECT FLT GE180
Primary end point	adverse events	adverse events	adverse events	adverse events
Secondary clinical end points (changes in)	UPDRS motor 3 in defined "off"; PDQ39; Addenbrooke's Cognitive Examination (Revised)	UPDRS motor 3 in defined "off"; PDQ39; Montreal Cognitive Assessment	UPDRS motor 3 in defined "off"; "off" time period PDQ39; Mini Mental State Examination score	UPDRS motor 3 in defined "off"; PDQ39; Mini Mental State Examination Score
Date for planned first-in-human study	2019–2020	2018	2018	2019

^aThe outcome of NeuroStemCellRepair and TRANSEURO.

Out of the press!!!!



Article

Phase I trial of hES cell-derived dopaminergic neurons for Parkinson's disease

<https://doi.org/10.1038/s41586-025-08845-y>

Received: 26 July 2024

Accepted: 26 February 2025

Published online: 16 April 2025

Open access

 Check for updates

V. Tabar^{1,2,3}✉, H. Sarva⁴, A. M. Lozano^{5,6}, A. Fasano^{6,7,8}, S. K. Kalia^{5,6}, K. K. H. Yu¹, C. Brennan¹, Y. Ma^{9,10}, S. Peng⁹, D. Eidelberg^{9,10}, M. Tomishima¹¹, S. Irion¹¹, W. Stemple¹¹, N. Abid¹¹, A. Lampron¹¹, L. Studer^{2,12,14} & C. Henchcliffe^{13,14}

Parkinson's disease is a progressive neurodegenerative condition with a considerable health and economic burden¹. It is characterized by the loss of midbrain dopaminergic neurons and a diminished response to symptomatic medical or surgical therapy as the disease progresses². Cell therapy aims to replenish lost dopaminergic neurons and

Article

Phase I/II trial of iPS-cell-derived dopaminergic cells for Parkinson's disease

<https://doi.org/10.1038/s41586-025-08700-0>

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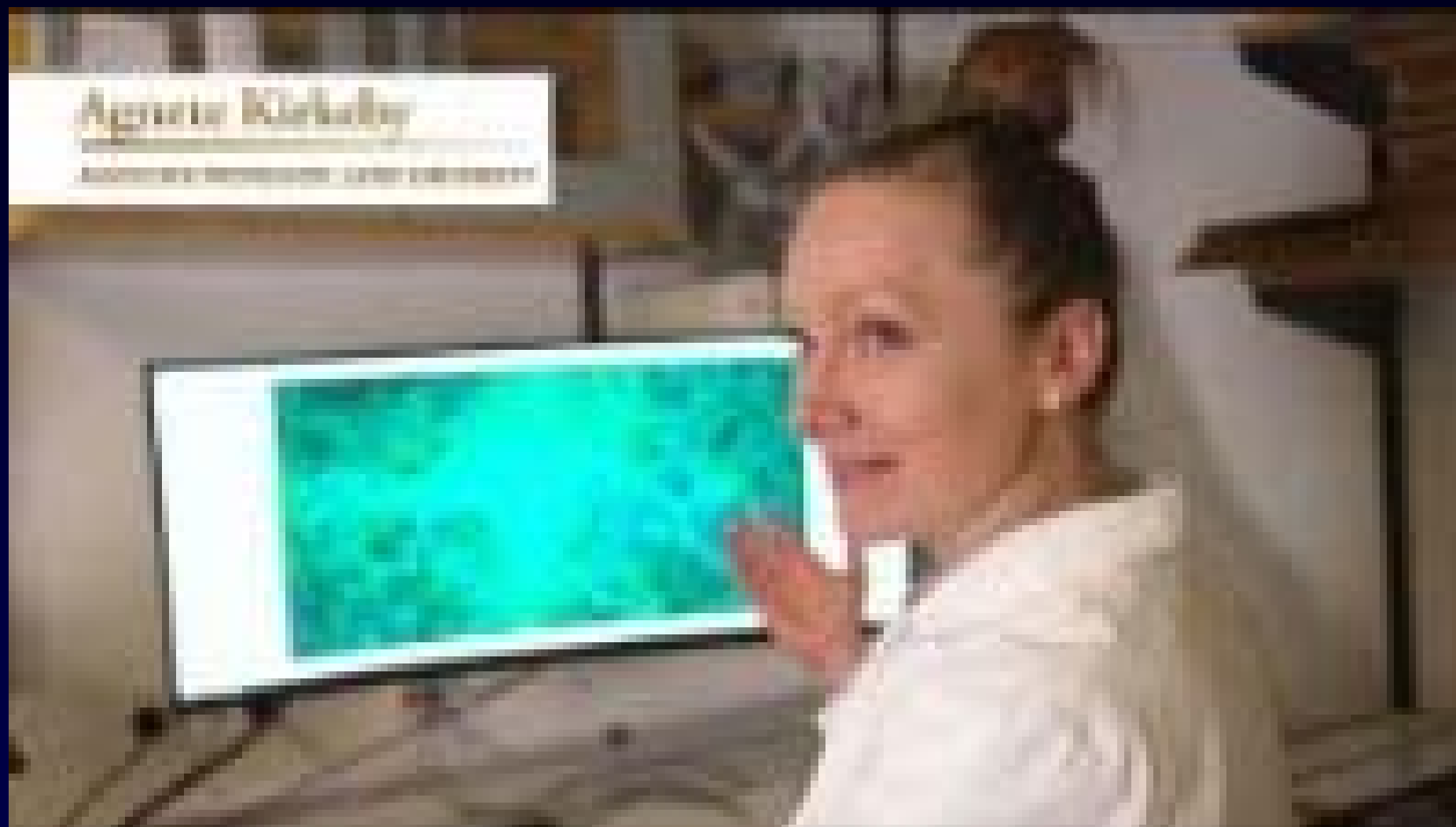
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Nobukatsu Sawamoto^{1,8}, Daisuke Doi^{2,8}, Etsuro Nakanishi^{1,8}, Masanori Sawamura^{1,8}, Takayuki Kikuchi³, Hodaka Yamakado¹, Yosuke Taruno¹, Atsushi Shima¹, Yasutaka Fushimi⁴, Tomohisa Okada⁴, Tetsuhiro Kikuchi², Asuka Morizane², Satoe Hiramatsu², Takayuki Anazawa⁵, Takero Shindo⁶, Kentaro Ueno⁷, Satoshi Morita⁷, Yoshiki Arakawa³, Yuji Nakamoto⁴, Susumu Miyamoto³, Ryosuke Takahashi¹✉ & Jun Takahashi²✉

Parkinson's disease is caused by the loss of dopamine neurons, causing motor symptoms. Initial cell therapies using fetal tissues showed promise but had complications and

Agneta Kiebel

PhD in Psychology
Assistant Professor - University of North Carolina

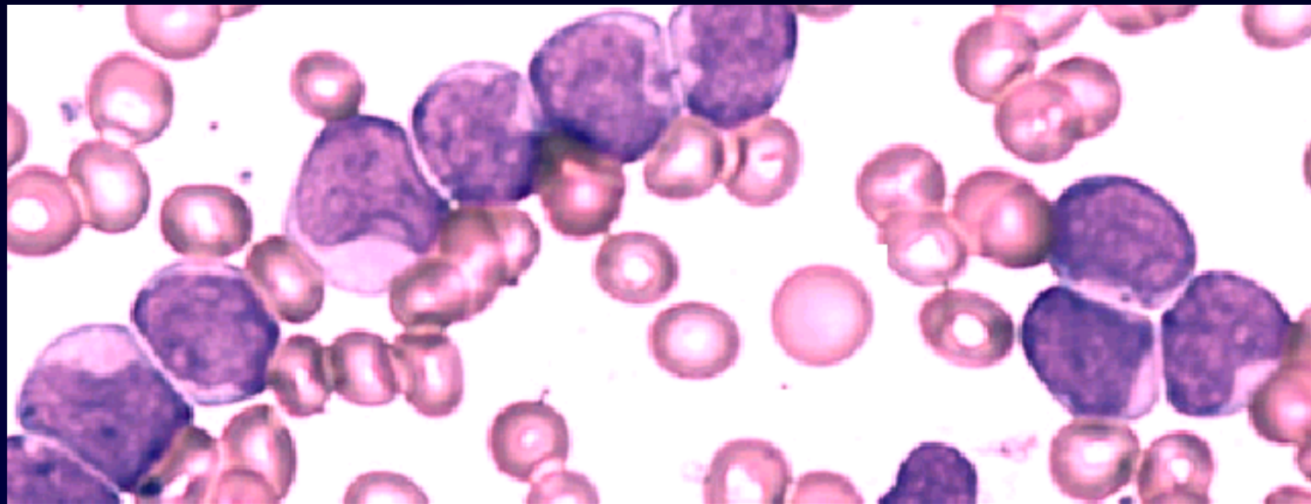


Καρκινικά βλαστικά κύτταρα

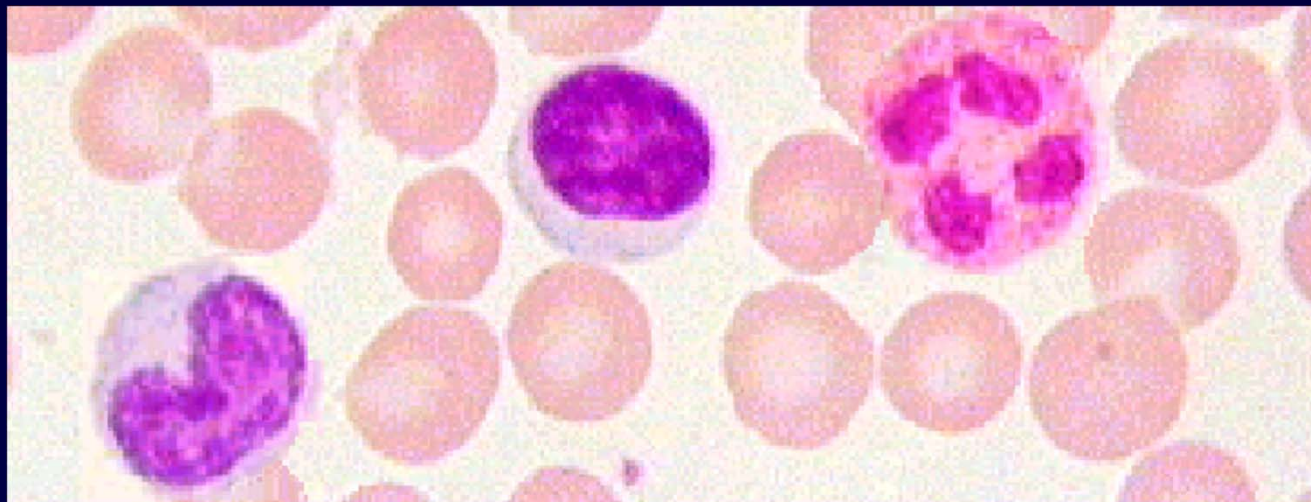
Καρκινικά βλαστικά κύτταρα (Cancer stem cells)

1937

With M. C. Kahn. The transmission of leukemia of mice with a single cell. *Am. J. Cancer* 31:276–82.



Leukemia



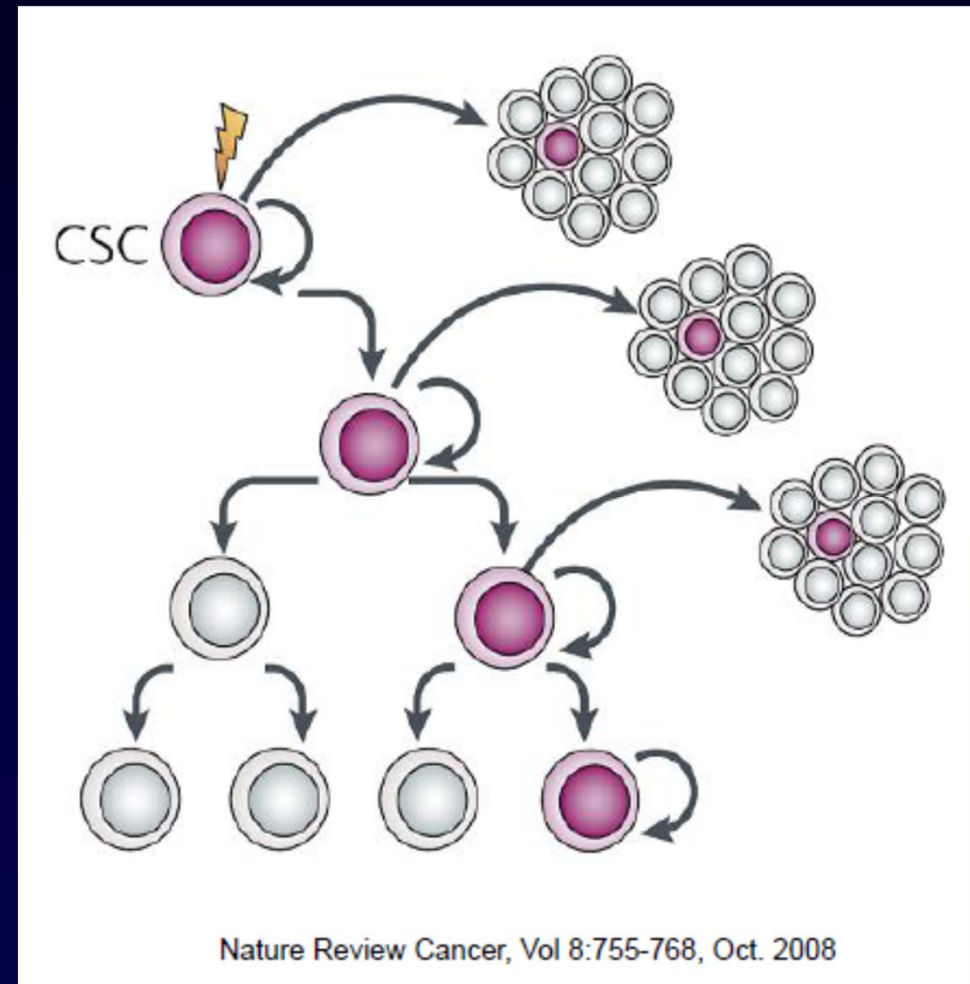
Normal

Τι είναι τα Καρκινικά βλαστικά κύτταρα

- είναι διαφορετικά από τα φυσιολογικά βλαστικά κύτταρα τα οποία γίνονται καρκινικά

- **Cancer Stem Cells (CSCs):**

- Υποπληθυσμός των κυττάρων ενός όγκου ο οποίος έχει την ικανότητα αυτοανανέωσης και μπορεί να δώσει γένεση σε διαφορετικούς κυτταρικούς τύπου που αποτελούν τον όγκο
- Οι ομοιότητες των καρκινικών βλαστικών κυττάρων αντιπροσωπεύουν την ομοιότητα τους με τα καρκινικά βλαστικά κύτταρα και της ικανότητάς τους να διατηρήσουν τον όγκο



Καρκινικά βλαστικά κύτταρα είναι κύτταρα τα οποία ευθύνονται για την έναρξη του όγκου και έχουν χαρακτηριστικά βλαστικών κυττάρων

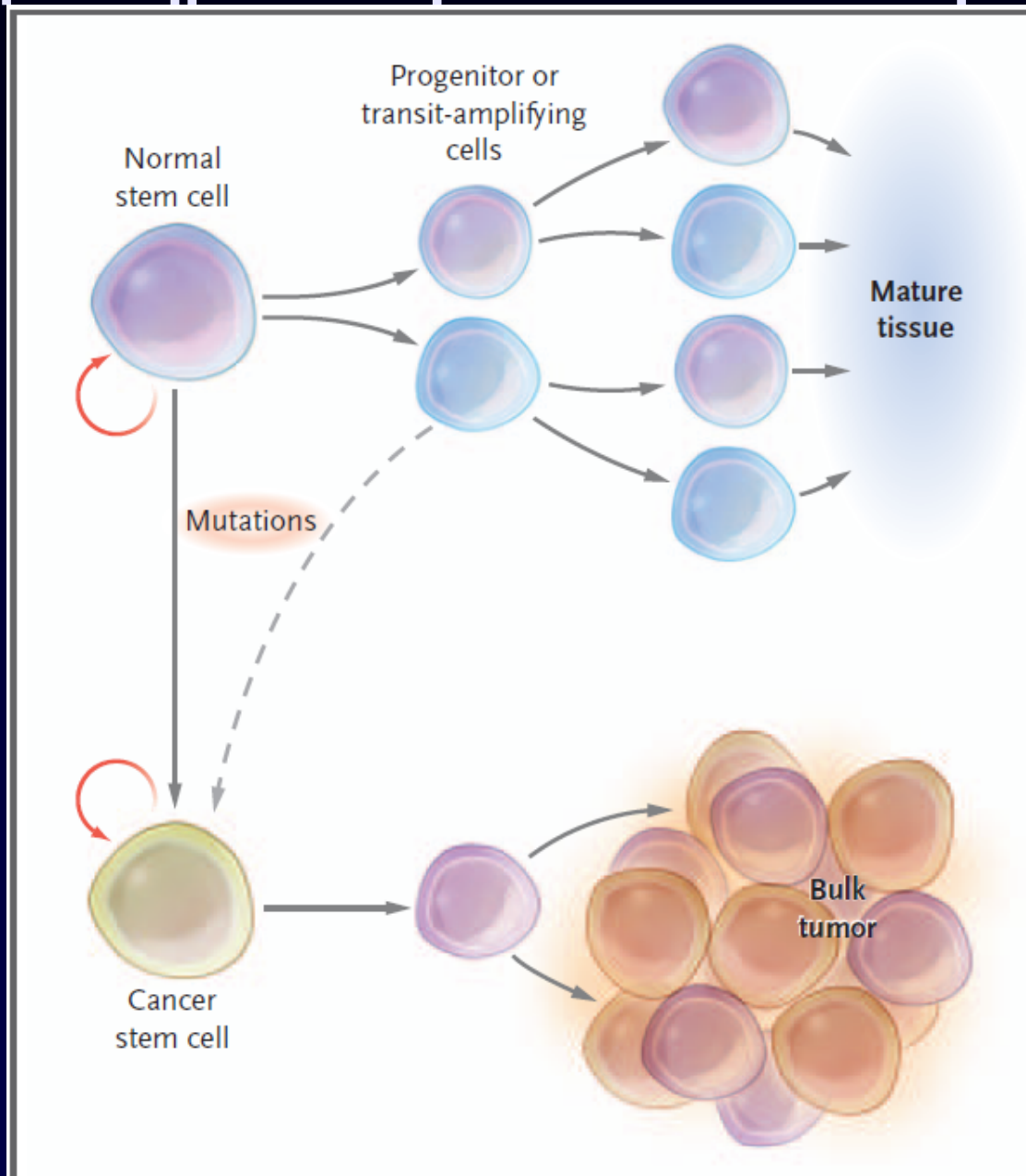


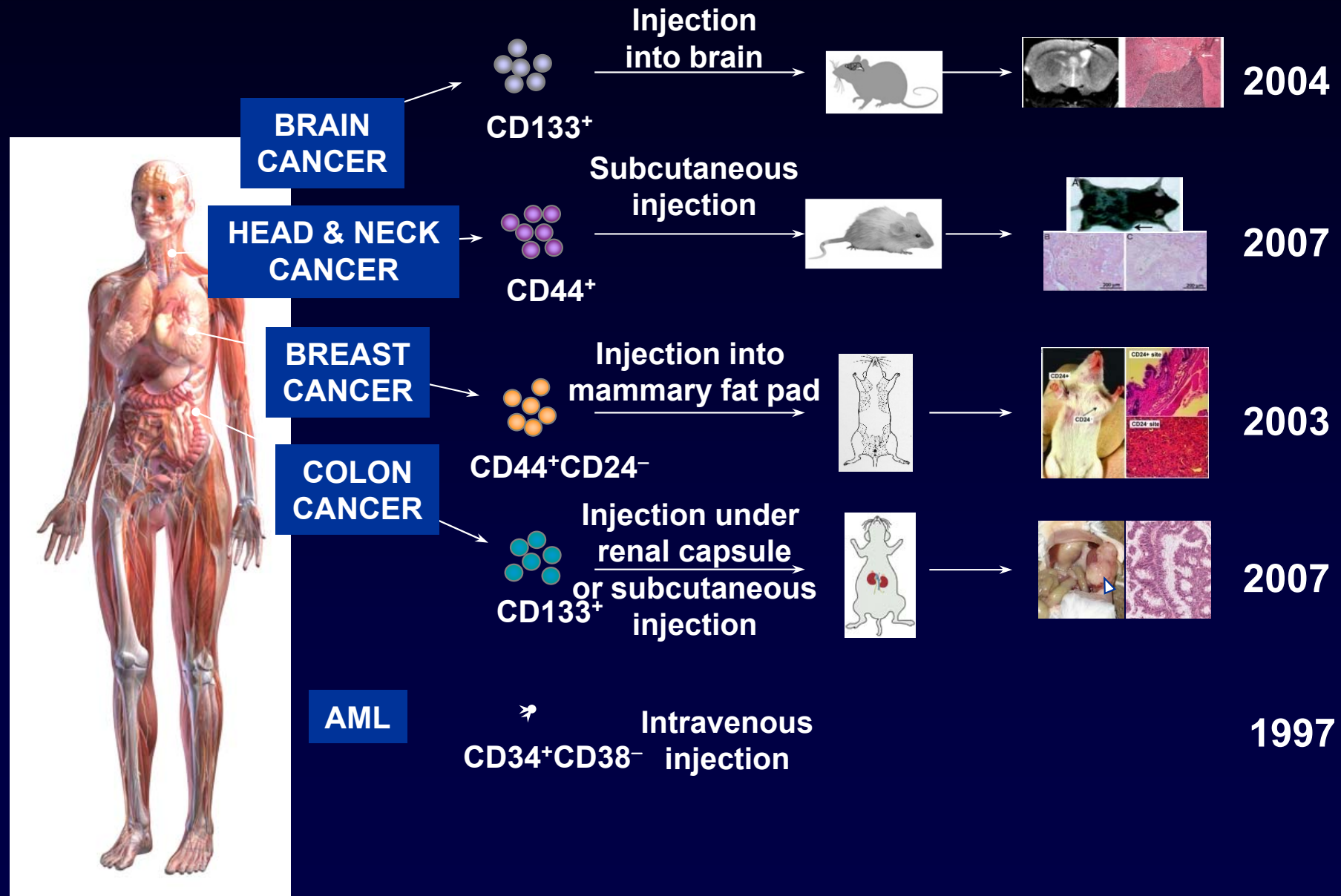
Figure 2. Stem-Cell Systems.

TABLE 1. Properties of neural stem cells and cancer stem cells^a

	Neural stem cells	Cancer stem cells
Cell surface marker	CD133	CD133; A2B5
Self-renewal	Unlimited	Unlimited
Proliferation	Low	Variable
Location	SVZ, SGZ of hippocampus	Variable
Proximity to vessel	Adjacent	Adjacent
Signaling pathway regulating fate	EGF, bFGF, Wnt, Shh, TGF β	EGF, bFGF, PDGF, Wnt, Shh, TGF β
Chemosensitivity	Sensitive	Variable with some resistance
Radiation	Sensitive	Variable with some resistance

^a SVZ, subventricular zone; SGZ, subgranular zone; EGF, epidermal growth factor; bFGF, basic fibroblast growth factor; Shh, sonic hedgehog; TGF, transforming growth factor.

Ταυτοποίηση καρκινικών βλαστικών κυττάρων που επάγουν τον όγκο (cancer- initiating cells)



Stem Cell Therapy? - διαφοροποίηση

Differentiation Therapy

Immature cells

(self renewal, unspecialized)

Differentiation

Mature cells

(end stage, specialized)

Cancer

(self renewal, unspecialized)

ώριμα / εξειδικευμένα



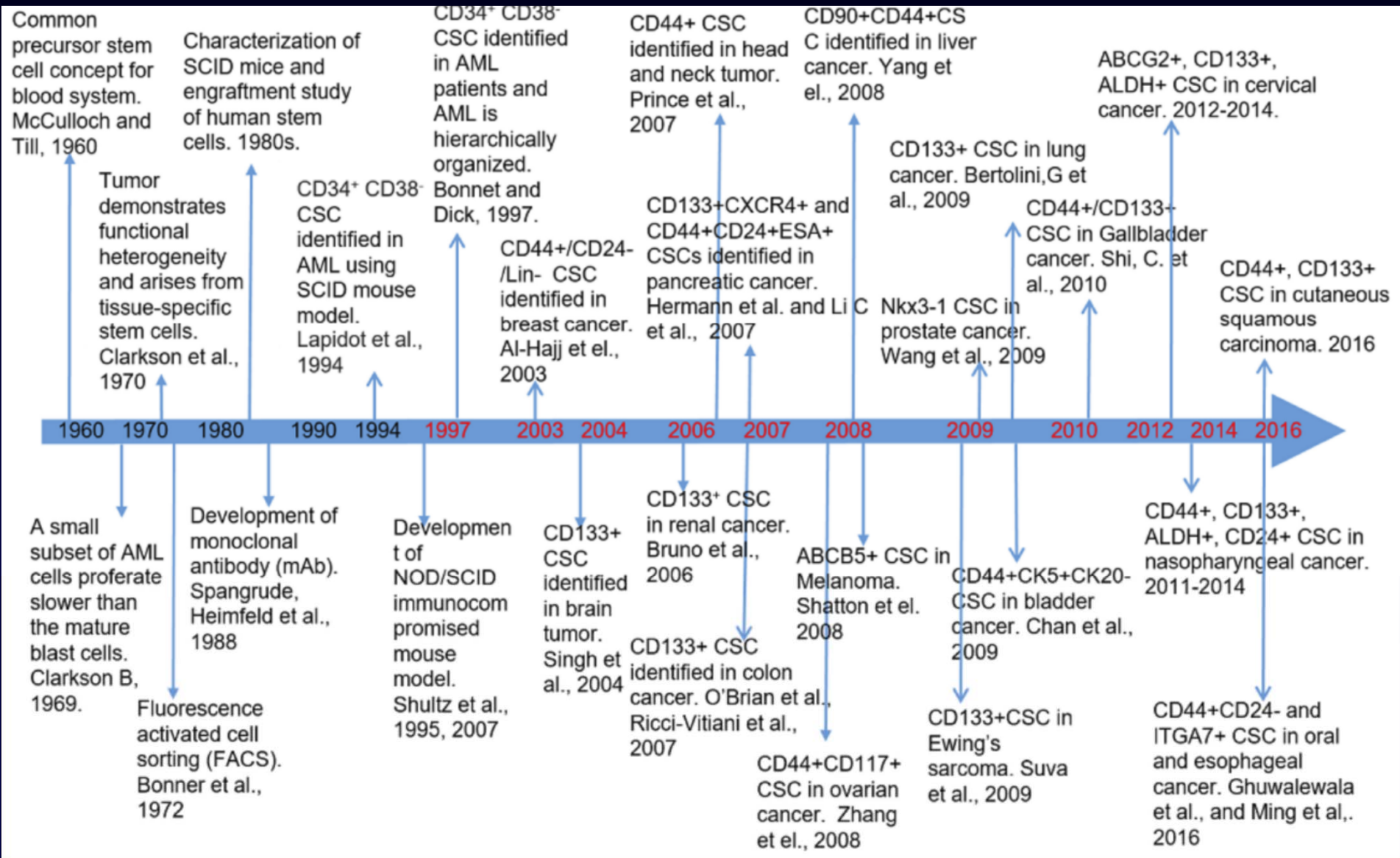
ανώριμα / αδιαφοροποίητα

↓ κυτταρική
διαίρεση

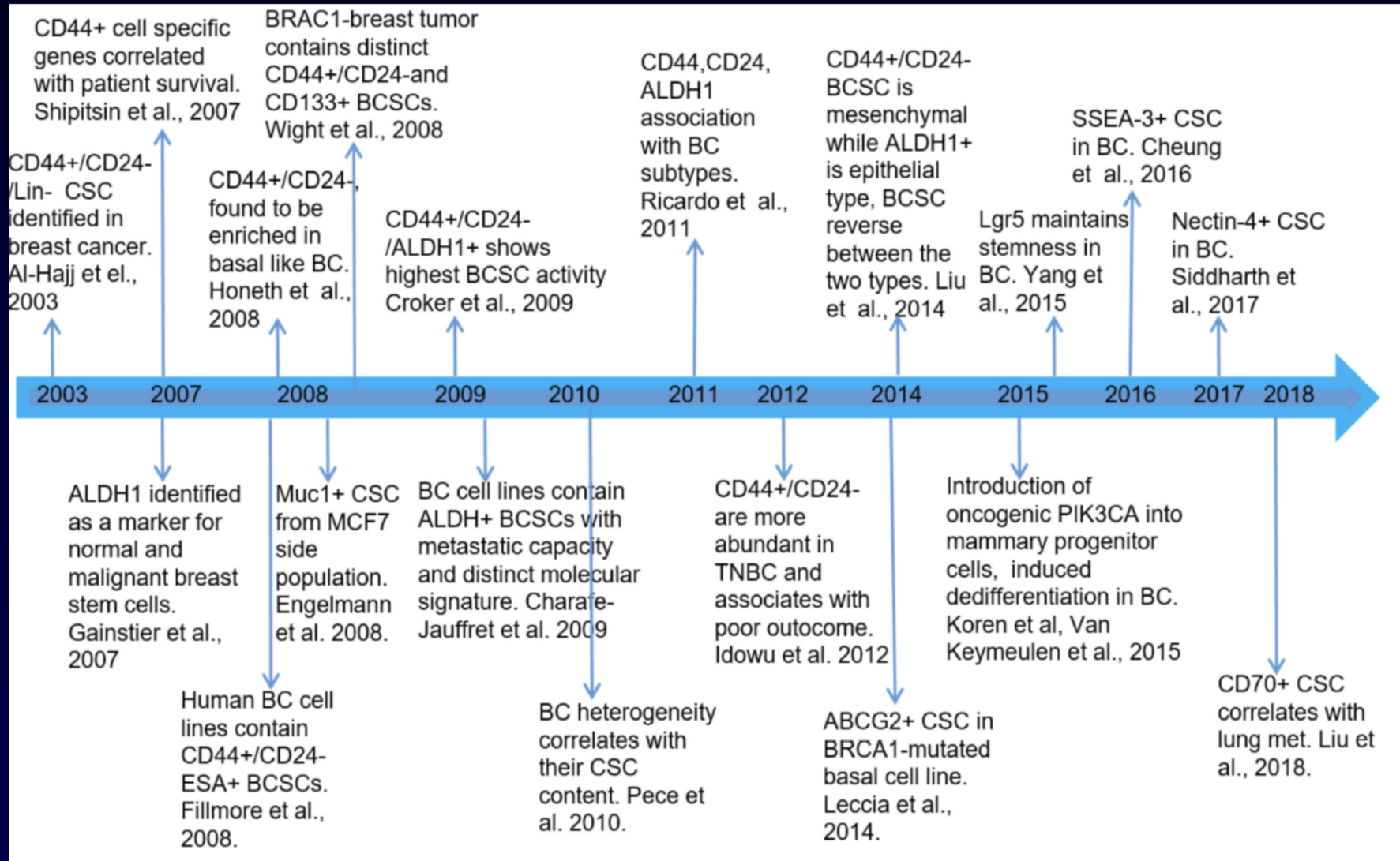
Καρκινικά βλαστικά κύτταρα – το μέλλον

- νέες θεραπευτικές εξελίξεις θα εξαρτηθούν από την ικανότητα της ανίχνευσης των καρκινικών βλαστικών κυττάρων
- νέα φάρμακα θα προσδιοριστούν από την ικανότητά τους να στοχεύσουν και να καταστρέψουν τα καρκινικά κύτταρα τα οποία προάγουν τον όγκο

AML



Breast cancer



■ ΤΕΛΟΣ