GEN KÖK
Genom ve Kök Hücre Merkezi
Genome and Stem Cell Center

STEM CELL THERAPIES IN RETINAL DEGENERATIONS

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ERCİYES ÜNİVERSİTESİ
1978

I have no financial interest to disclose

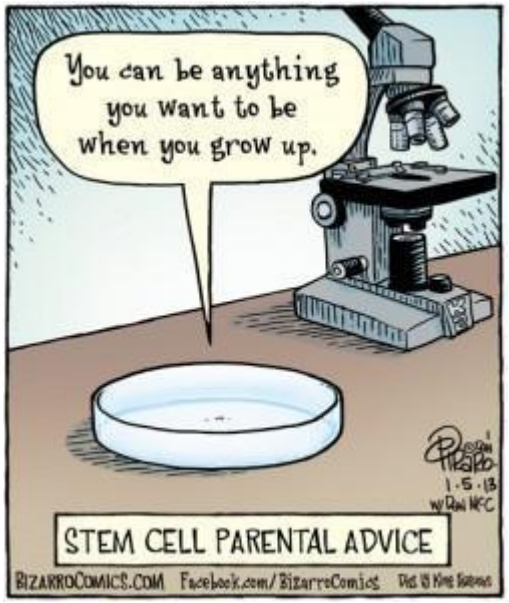
MAIN TOPICS

* What is stem cell?
* What are the types and properties of stem cell?
* Applications in retinal diseases?
* Our clinical phase 1 study?

WHAT IS STEM CELL?

Stem cell:

* Basic, pure, unspecialized form of cell.
* Has the potential to develop into many different cell types in the body.
* Repair damaged cells and tissues



History of Stem Cells

* **1981:** Discovery of embryonic stem cells from early mouse embryos.
* **1998:** Stem cells from human embryos are derived in the laboratory and called **human embryonic stem cells (hESCs)**
* **2006:** Researchers "reprogrammed" some adult cells to behave like an ESC (**induced pluripotent stem cells) (iPSCs)**,
* **2009:** With the permission of the FDA, the first hESC clinical trial was approved for spinal cord injury.

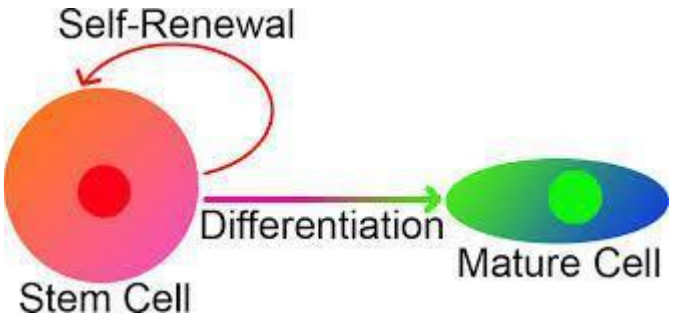
UNIQUE PROPERTIES OF SC

- Proliferation
- Self-renewal
- Differentiation

PROLIFERATION-SELF-RENEWAL

* **Proliferation:** Stem cells are capable of dividing and replicating themselves for long periods.

* **Self –renewal:** After division, it can continue as a stem cell, like the parent stem cells.



DIFFERENTIATION

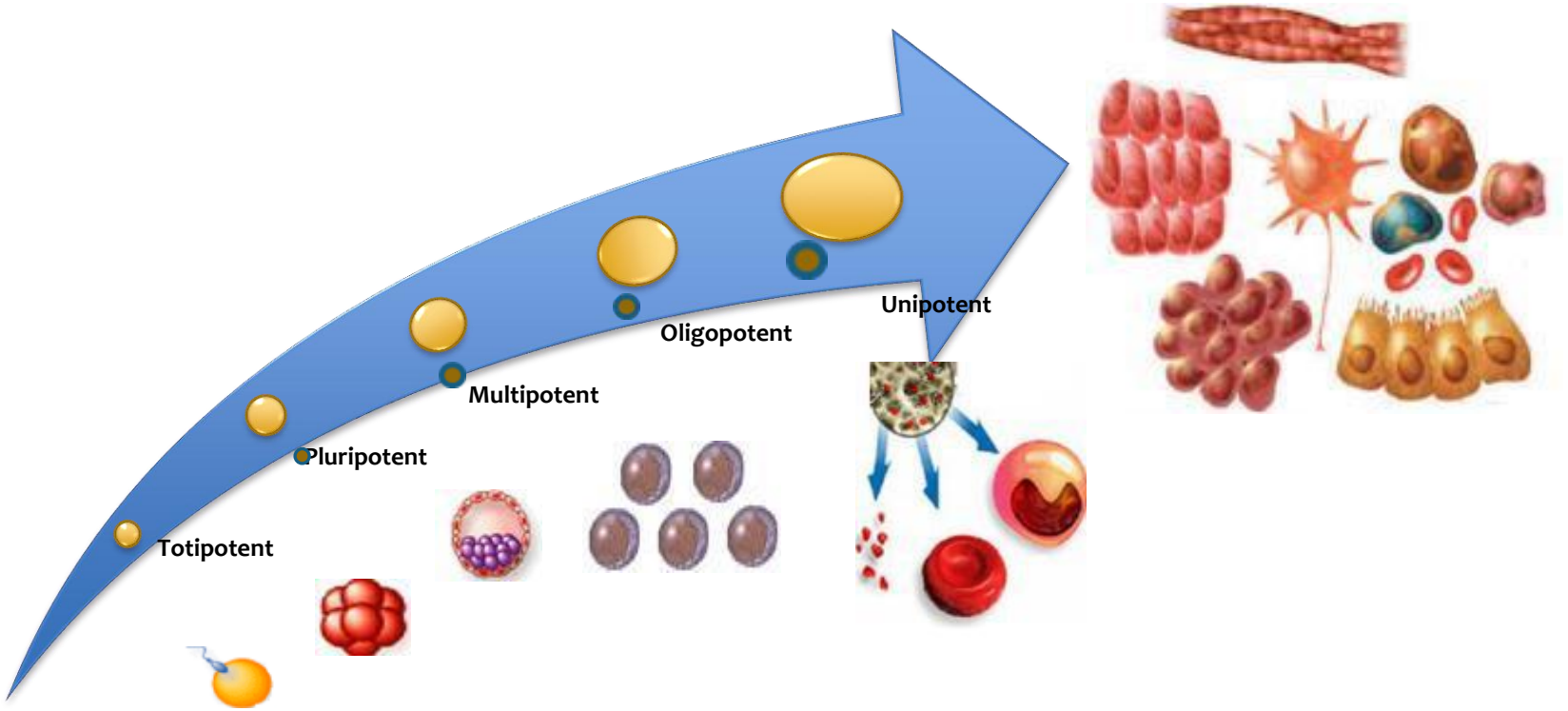
Differentiation: When unspecialized stem cells give rise to specialized cells, the process is called **differentiation.**

There are internal and external signals during differantiation.

The internal **signals** are controlled by a cell's **genes**

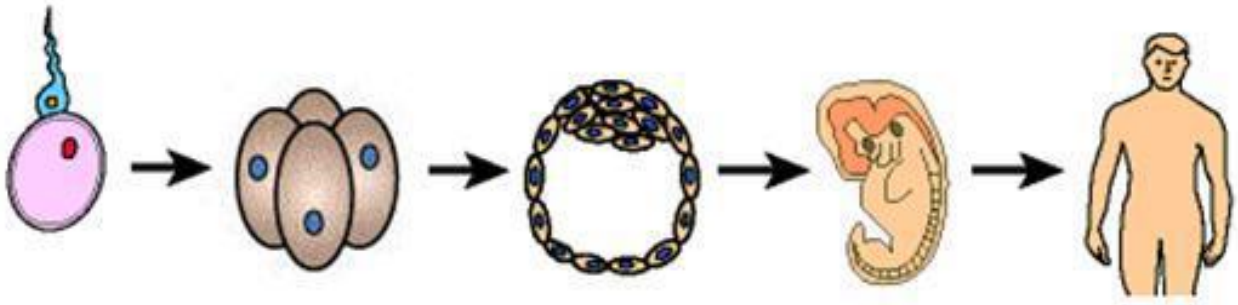
The external signals for cell differentiation include chemicals secreted by other cells, physical contact with neighboring cells, and certain molecules in the **microenvironment.**

POTENCY OF SC



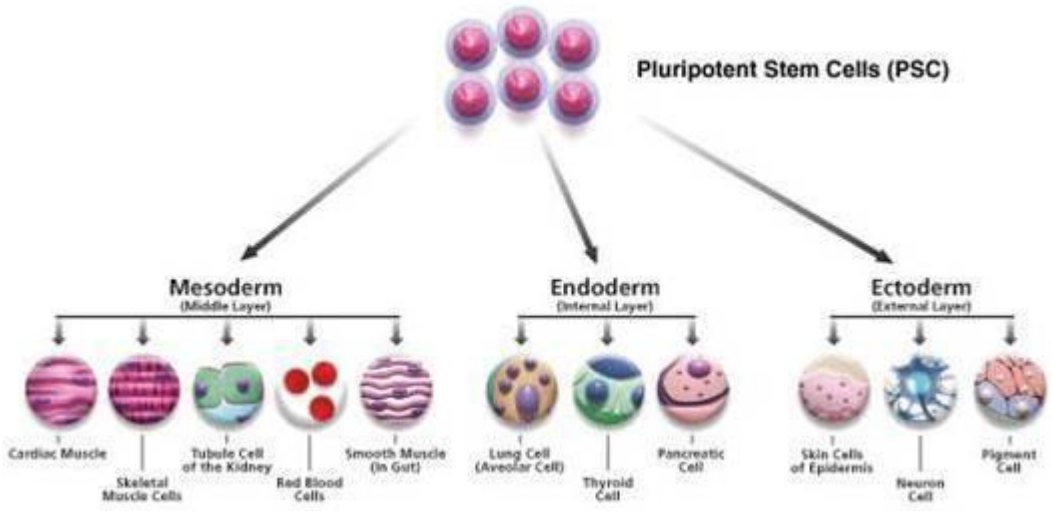
Totipotent:

- Unlimited proliferation capacity
- Are able to differentiate into embriyogenic and extra embriyogenic cells.



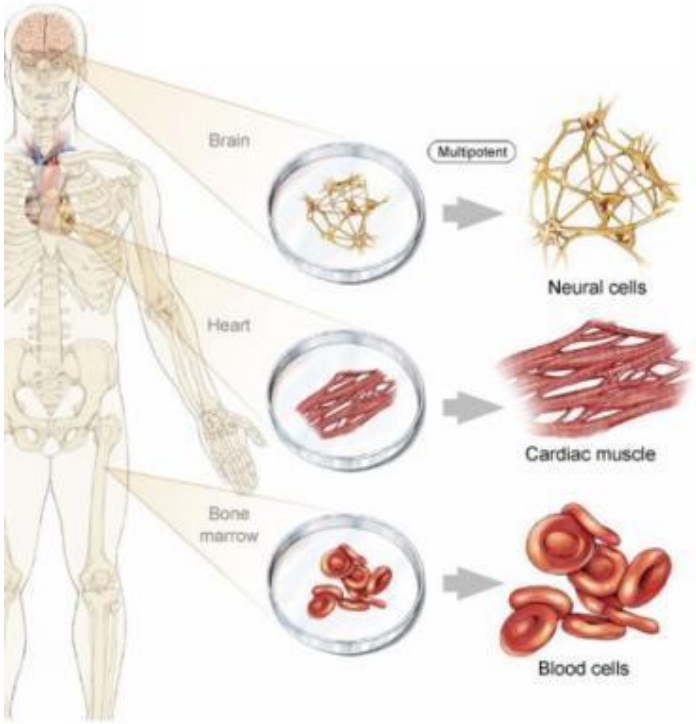
Pluripotent:

Capacity to differentiate into embriyogenic cell layers (endoderm, mesoderm, ectoderm)



Multipotent

Capacity to differentiate limited types of cell types.



TYPES OF STEM CELLS

1-EMBRYONIC STEM CELLS

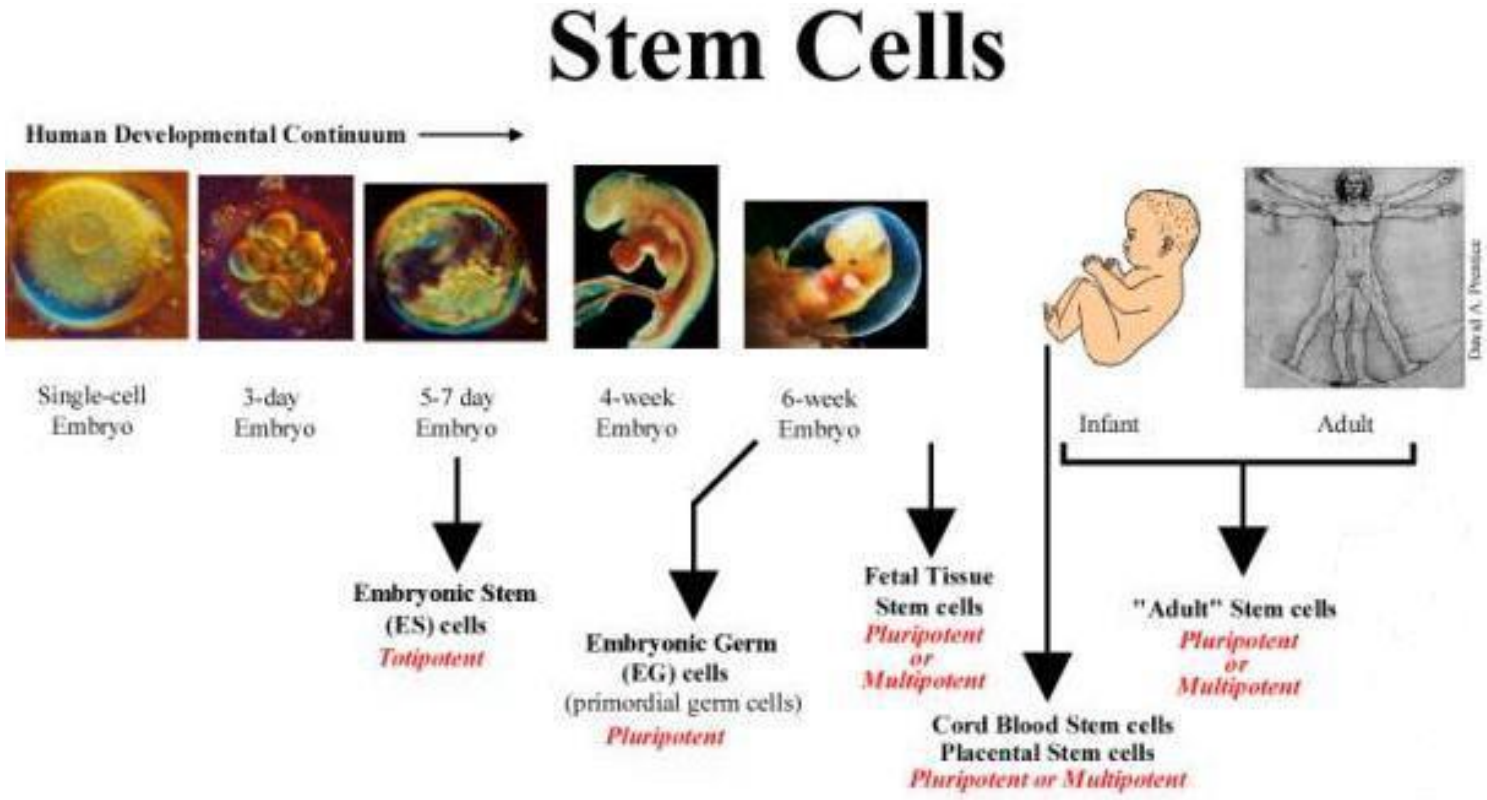
2- ADULT STEM CELLS

- Mesencymal SC
- İnduced Pluripotent SC

3-CORD BLOOD STEM CELLS

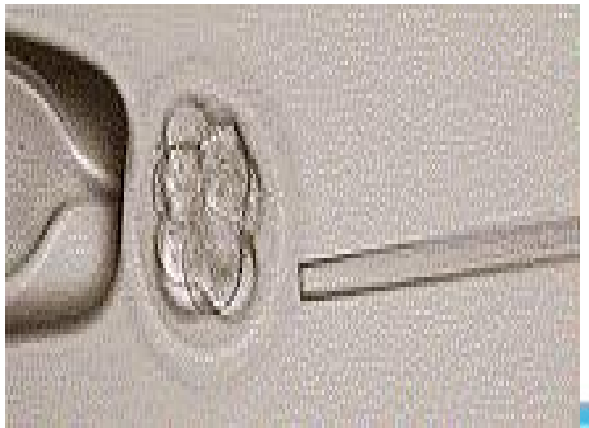
4- AMNIOTIC FLUID STEM CELLS

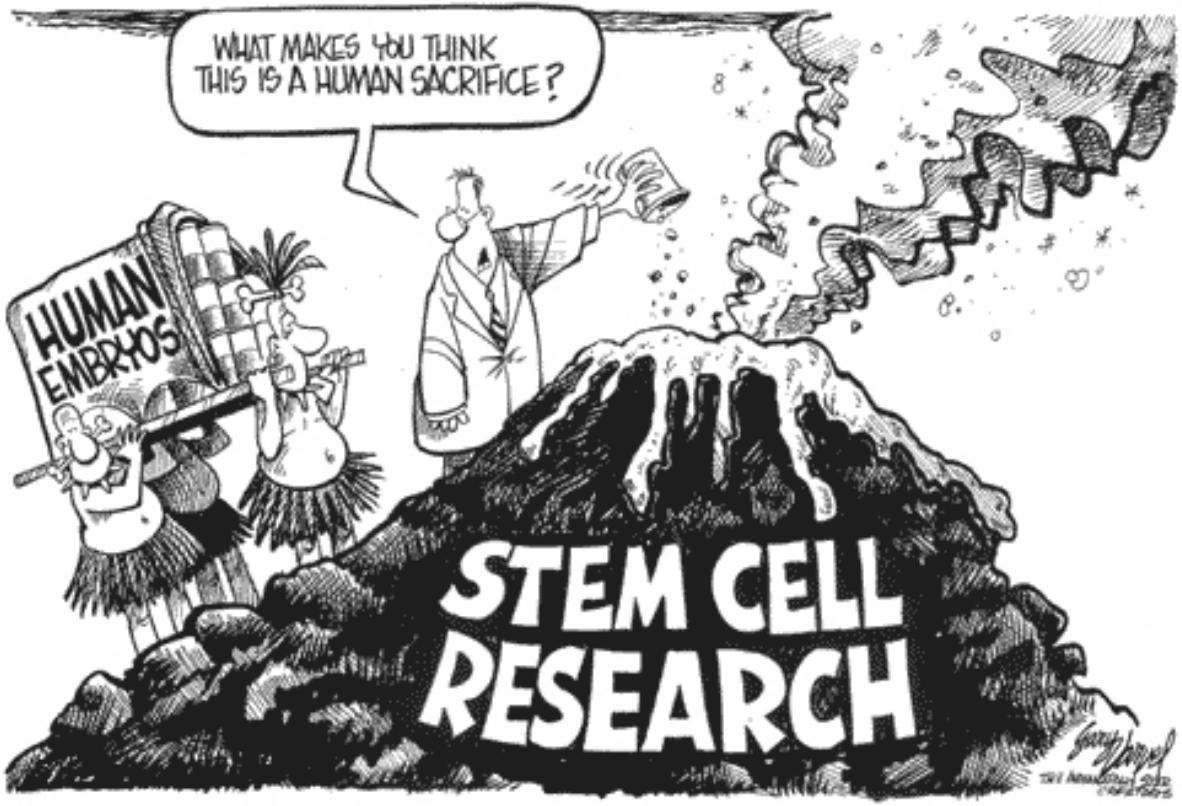
ORIGIN OF STEM CELLS



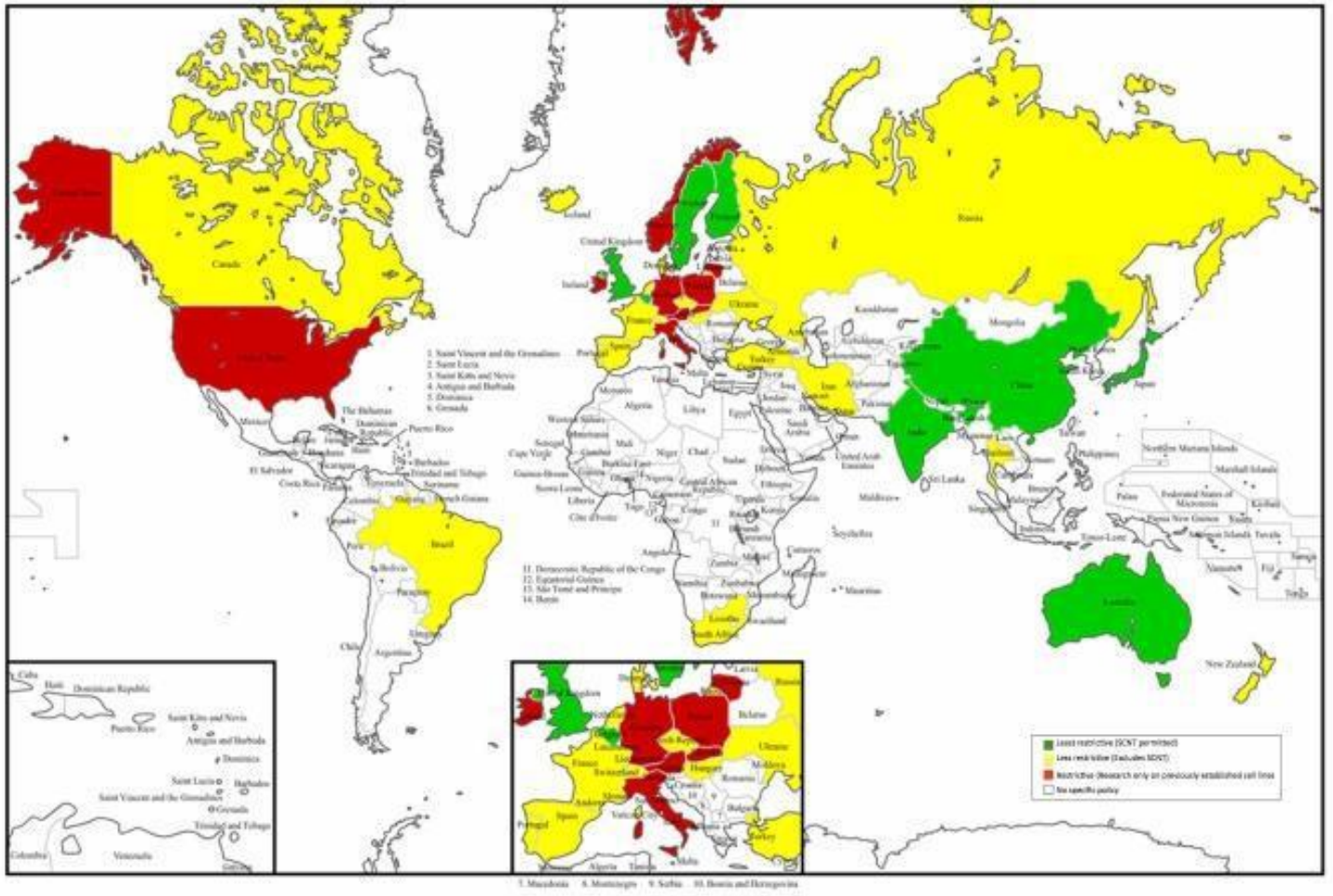
Embryonic (ES) SC and Foetal SC

* Derived from human embryos that are few days old or foetus
* Differentiate to any type of cell.
* Can be grown easily but difficult to manipulate.
* Tumor formation, hyperproliferation and possibility of rejection





ETHICAL ISSUES- EMBRYO IS DESTROYED



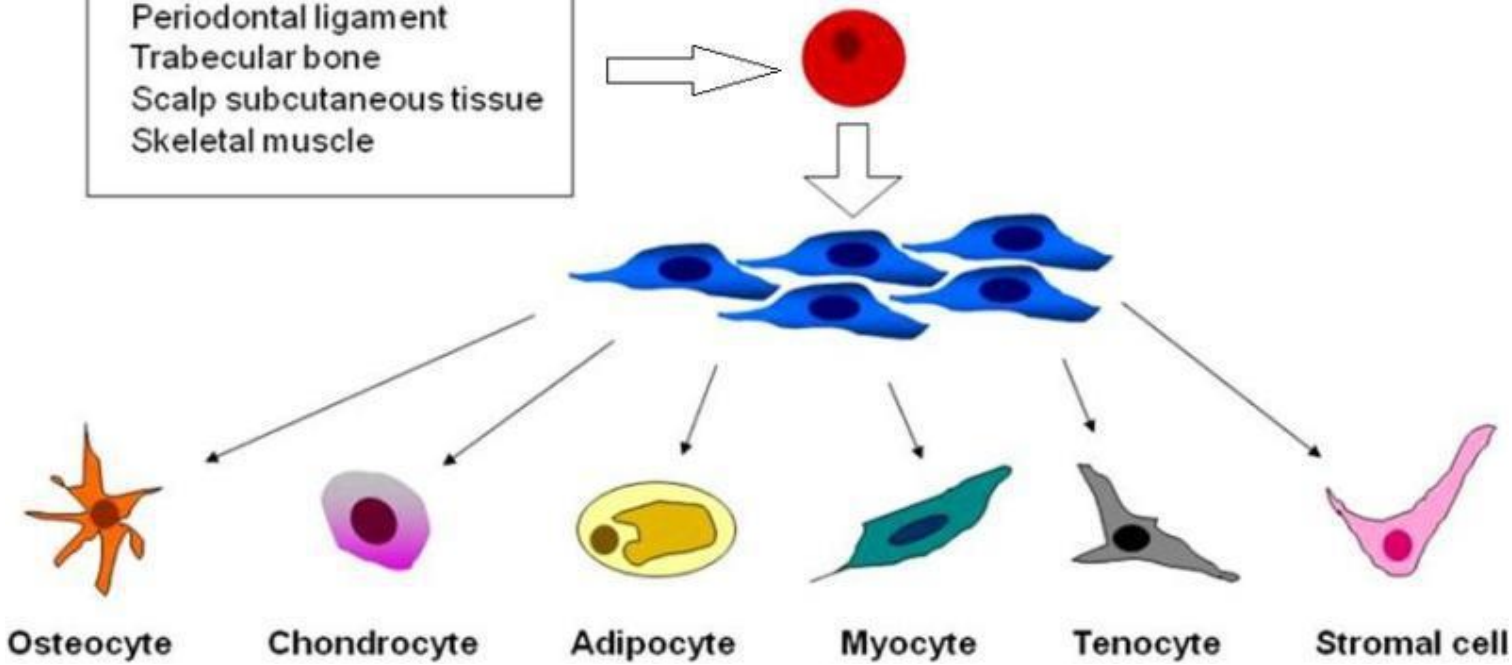
hESC MAP

Adult SC

* Found in all humans and can be obtained easily
* Differentiate to a limited number of cell types
* Maintain and repair the tissue in which they are found.
* Tumor formation and rejection is less likely

Adult tissues

- Bone marrow
- Deciduous teeth
- Fat
- Hair follicles
- Peripheral blood
- Periodontal ligament
- Trabecular bone
- Scalp subcutaneous tissue
- Skeletal muscle



Induced Pluripotent SC (IPSC)

* Adult somatic cells reprogramed to become like embryonic stem cells (**induced pluripotent stem cells, iPSCs**)
* Viruses are currently used to introduce the reprogramming factors (OCT4, SOX2, KLF4 and c-MYC) into adult cells.
* Limited availibility with difficult techniques.
* Cost is high.
* Tumor formation and rejection are the problems.

STATUS IN OUR COUNTRY

* Human ESCs are restricted in our country (2005)
* Adult SCs and IPSC can be used with the approval of Local Ethical Committee and Ministry of Health

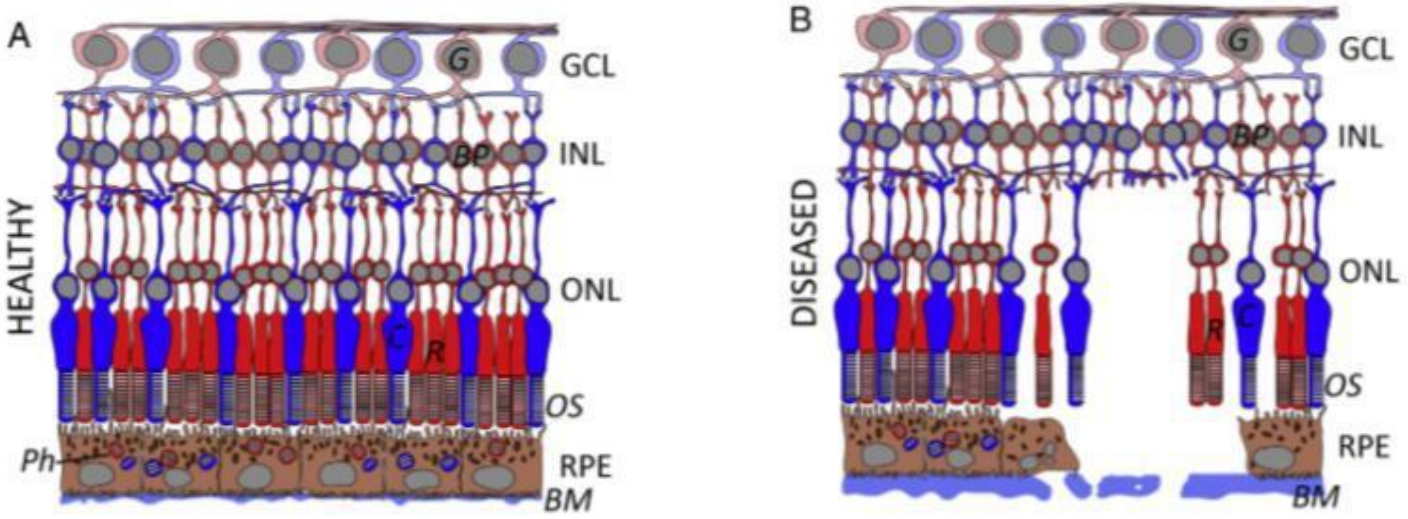
(Please read TCK: 90)

SC in OPHTALMOLOGY

CORNEA AND RETINA

SC in RETINAL DISEASES

* Very complex tissue
* Has 10 different layers and 9 types of cells connected with each other.
* An alteration or damage in any of these layers can impair the anatomy and functions of the retina.



Mechanisms of Stem Cell Therapy

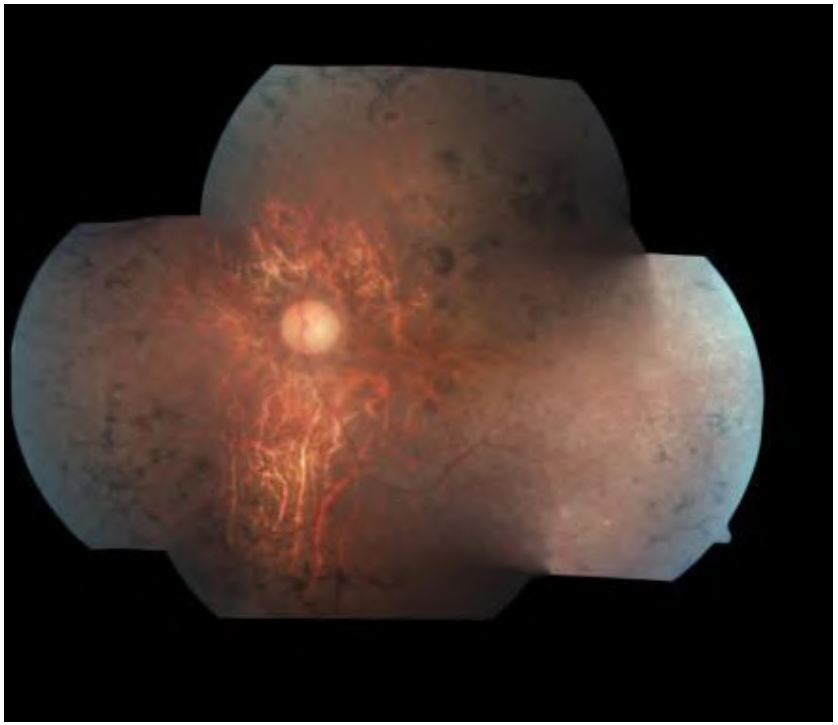
THERE ARE FOUR MECHANISMS:

* (1) Cell replacement: Healthy stem cells can replace degenerated cells
* (2) Nutritional support: Healthy stem cells secrete some trophic factors and promote the survival of surrounding cells.
* (3) Upregulating antiapoptotic genes and nutritional factors.
* (4) Promote new synaptic connections.

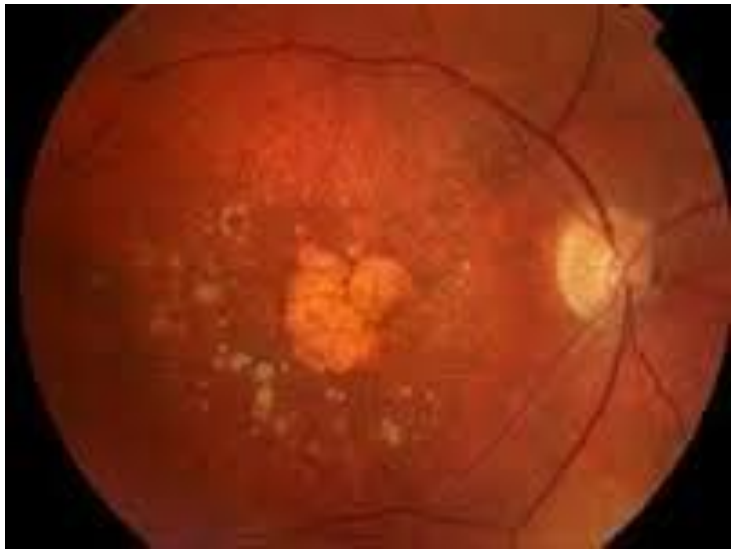
Advantages of SC Use in Eye

* Relatively small number of cells and low doses required
* Easy accesibility for surgery
* Direct visualization of grafts
* İmmun privilige of the eye
* One eye can be used as control in bilateral disease
* Rare extraocular distribution.

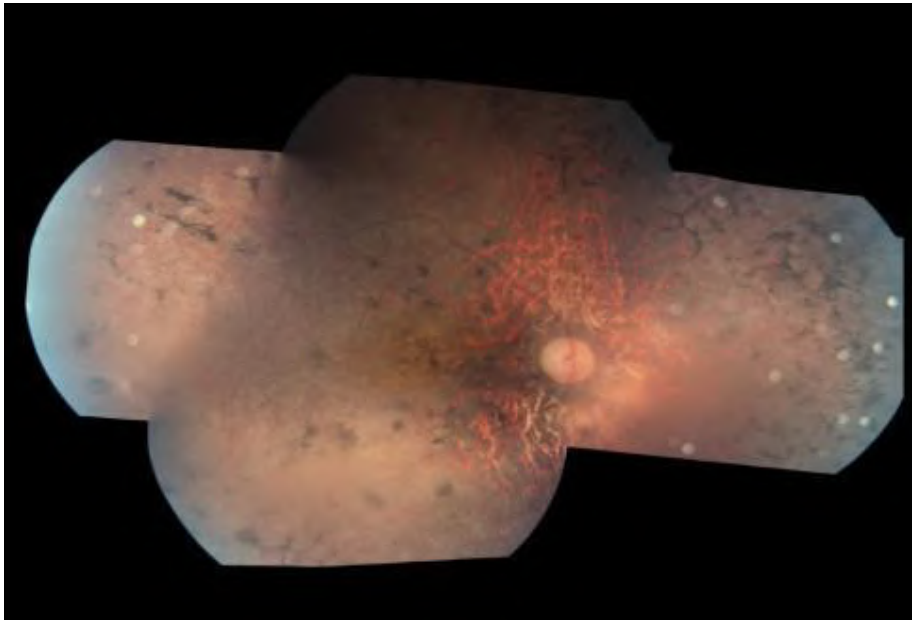
TARGET RETINAL DISEASES ARE THE DISEASES WITH NO DEFINITIVE TREATMENTS



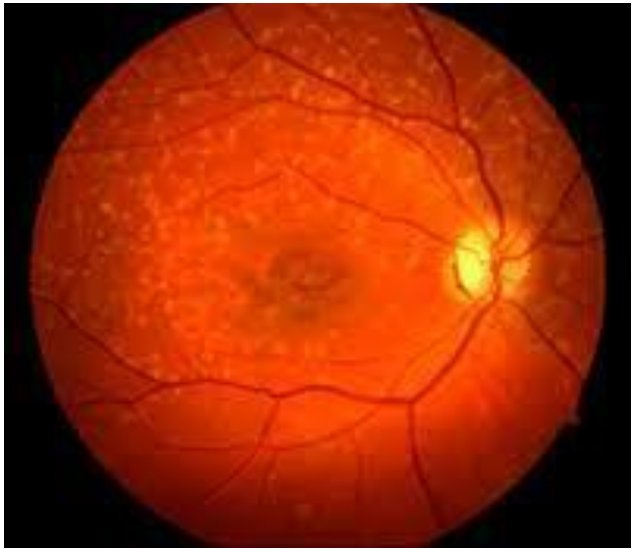
Age Related Macular Degeneration (AMD)



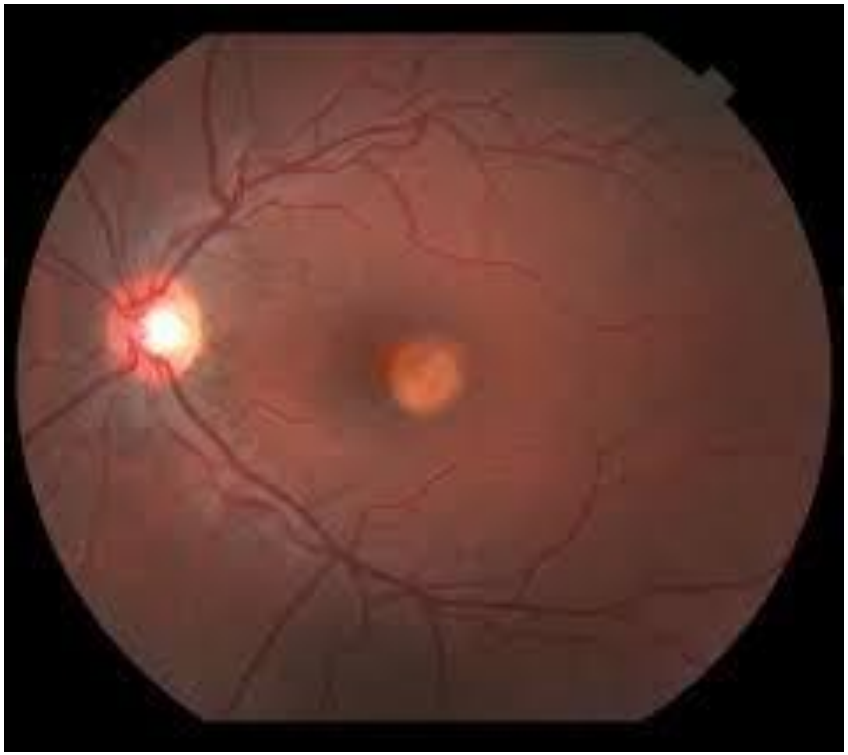
Retinitis Pigmentosa



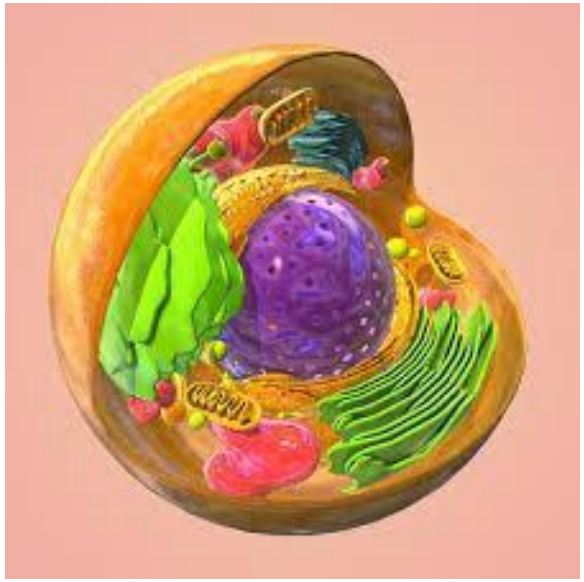
Stargardt's Macular Dystrophy



Best's Vitelliform Macular Distrophy



WHICH KIND OF STEM CELLS
CAN BE USED IN RETINAL DEGENERATIONS?



REGISTERED TRIALS WITH ESC clinicaltrials.gov

* (1) (NCT01469832) Safety and Tolerability of Sub-retinal Transplantation of hESC-RPE Cells in Patients with Stargardt's Macular Dystrophy (SMD)
* (2) (NCT01691261) A Study of Implantation of hESC-RPE Subjects with Acute Wet AMD and Recent Rapid Vision Decline
* (3) (NCT01674829) A Phase Ⅰ/Ⅱa, Open-Label, Single-Center, Prospective Study to Determine the Safety and Tolerability of Subretinal Transplantation of hESC-RPE (MA09hRPE) Cells in Patients With Advanced Dry AMD
* (4) (NCT02122159) Research With Retinal Cells Derived From Stem Cells for Myopic Macular Degeneration
* (5) (NCT01344993) Safety and Tolerability of Sub-retinal Transplantation of hESC Derived RPE (MA09-hRPE) Cells in Patients with Advanced Dry AMD
* (6) (NCT01345006) Sub-retinal Transplantation of hESC Derived RPE (MA09-hRPE) Cells in Patients with Stargardt's Macular Dystrophy;
* (7) (NCT01625559) Safety and Tolerability of MA09-hRPE Cells in Patients with SMD.

REGISTERED CLINICAL TRIALS WITH IPSC clinicaltrials.gov

* (1) (NCT02162953) Development of Induced Pluripotent Stem Cells from Patients with Best Disease and Other Inherited Retinal Degenerative Diseases

* (2) (NCT01432847) Generation of induced pluripotent stem (iPS) Cell Lines from Somatic Cells of Participants with Eye Diseases and from Somatic Cells of Matched Controls.

Registered Clinical Trials with BMDMSC

* 1) NCT01736059- Clinical Trial of Autologous Intravitreal Bone-marrow CD34+ Stem Cells for Retinopathy. Non-exudative AMD, Diabetic Retinopathy. RVO, RP, Hereditary Macular Degeneration.
* 2) NCT01560715- Autologous Bone Marrow-Derived Stem Cells Transplantation For RP **(RETICELL)**
* 3) NCT02280135- Clinical Trial of Intravitreal Injection of Autologous Bone Marrow Stem Cells in Patients With Retinitis Pigmentosa (TC/RP)
* 4)NCT01068561- Autologous Bone Marrow-Derived Stem Cells Transplantation For Retinitis Pigmentosa
* 5) NCT01914913- Clinical Study to Evaluate Safety and Efficacy of BMMNC in Retinitis Pigmentosa
* 6) NCT01531348-Feasibility and Safety of Adult Human Bone Marrow-derived Mesenchymal Stem Cells by Intravitreal Injection in Patients With Retinitis Pigmentosa
* 7) NCT01518127-Intravitreal Bone Marrow-Derived Stem Cells in Patients With Macular Degeneration (AMDCELL)

Registered Trials with ADMSC

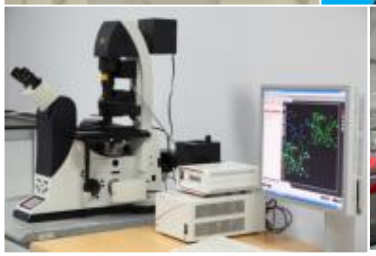
* (1) (NCT02024269) Study to Assess the Safety and Effects of Cells Injected Intravitreal in Dry Macular Degeneration

* (2) (NCT02144103) Effectiveness and Safety of Adipose-Derived Regenerative Cells for Treatment of Glaucomatous Neurodegeneration.

OUR CLINICAL STUDY

* Phase 1 prospective safety study.
* Approval of the Ethical Committee of University (March-2014)
* Approval of Ministry of Health (January-2015)
* 20 Volunteers with end-stage RP

GENKÖK



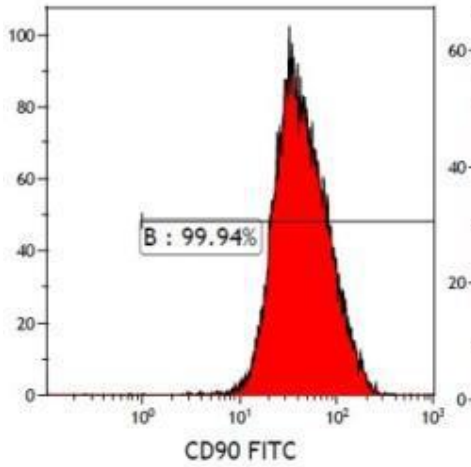
PATIENTS AND METHODS

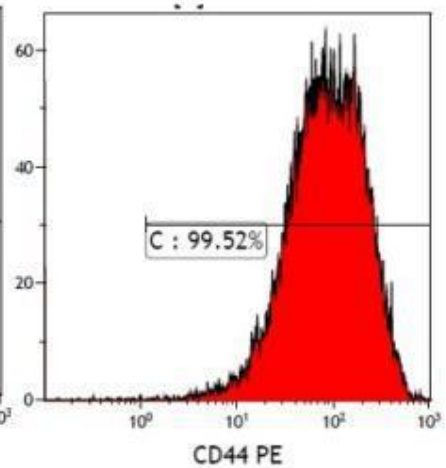
* 15 patients were included
* All had total visual field defect.
* % 50 of the patients were only p +
* The best VA was hand motion from 1 meter.
* All had undetectable ERG.
* The worse eye was operated.
* After total vitrectomy with 23 gauge, 1.000.000 SCs were injected subretinally.
* Adipose Tissue Derivated MSC s were used.

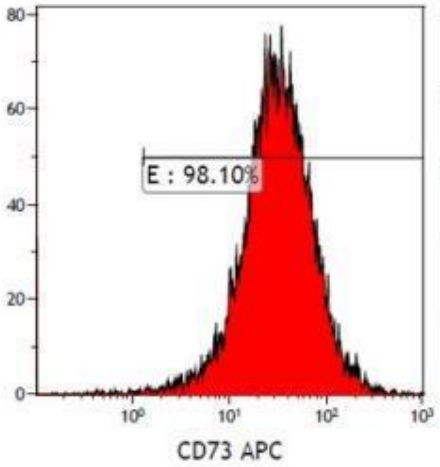
GMP (Good manufacturing practice)

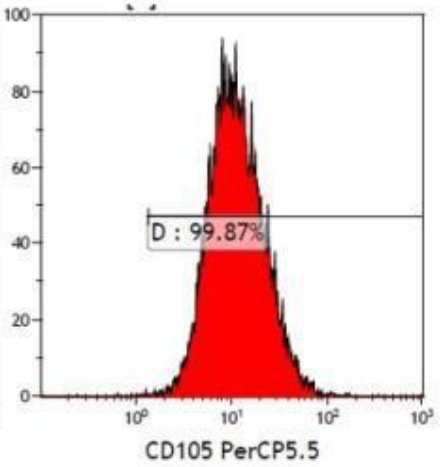
ADMSCs were prepared under strict environmental conditions with international standarts

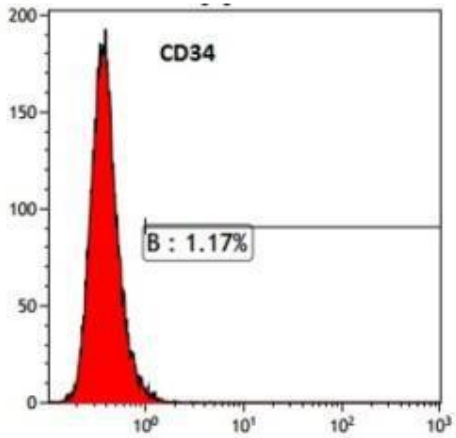
Allogenic human adipose derived MSCs flow cytometry results

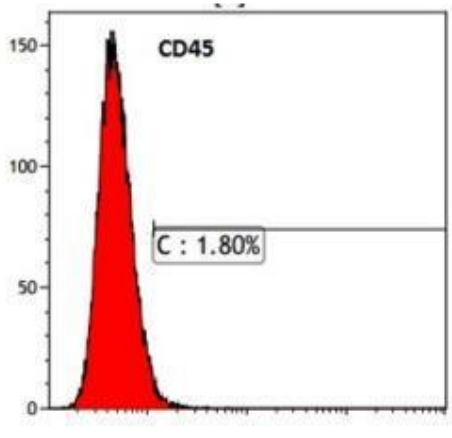


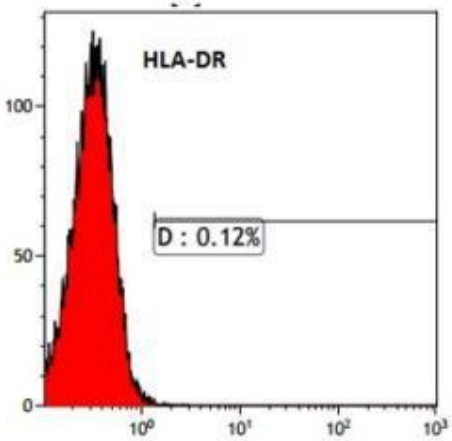


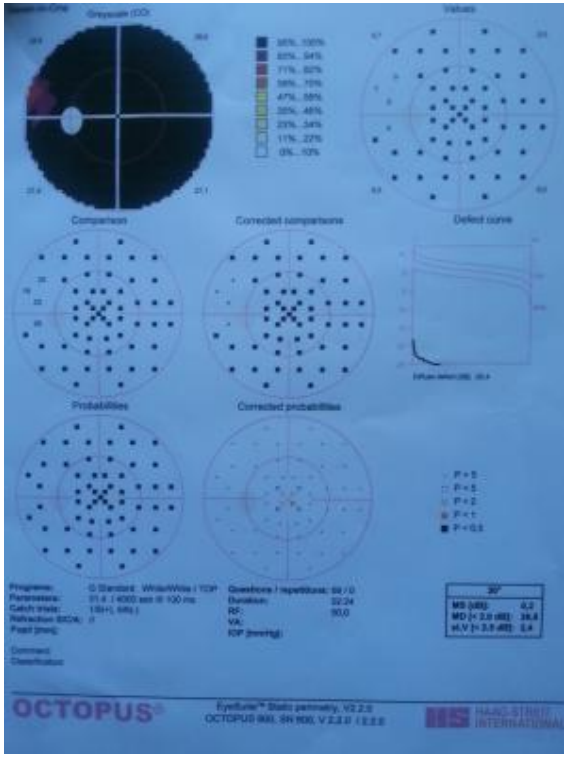


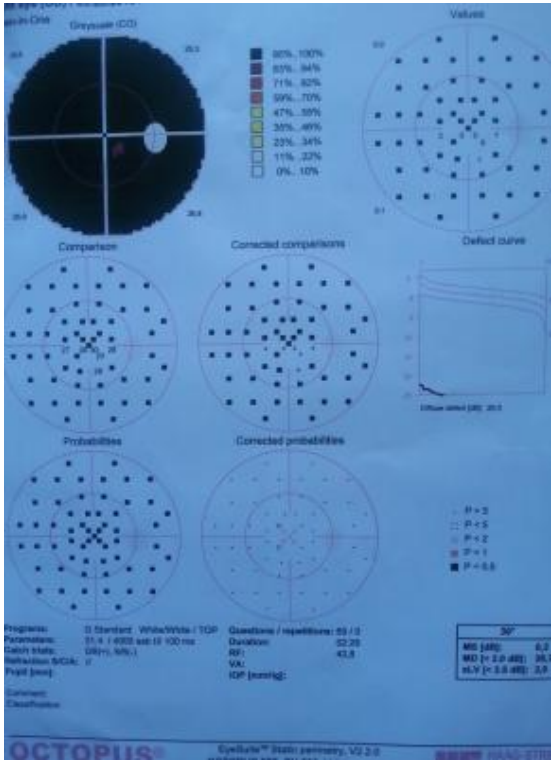




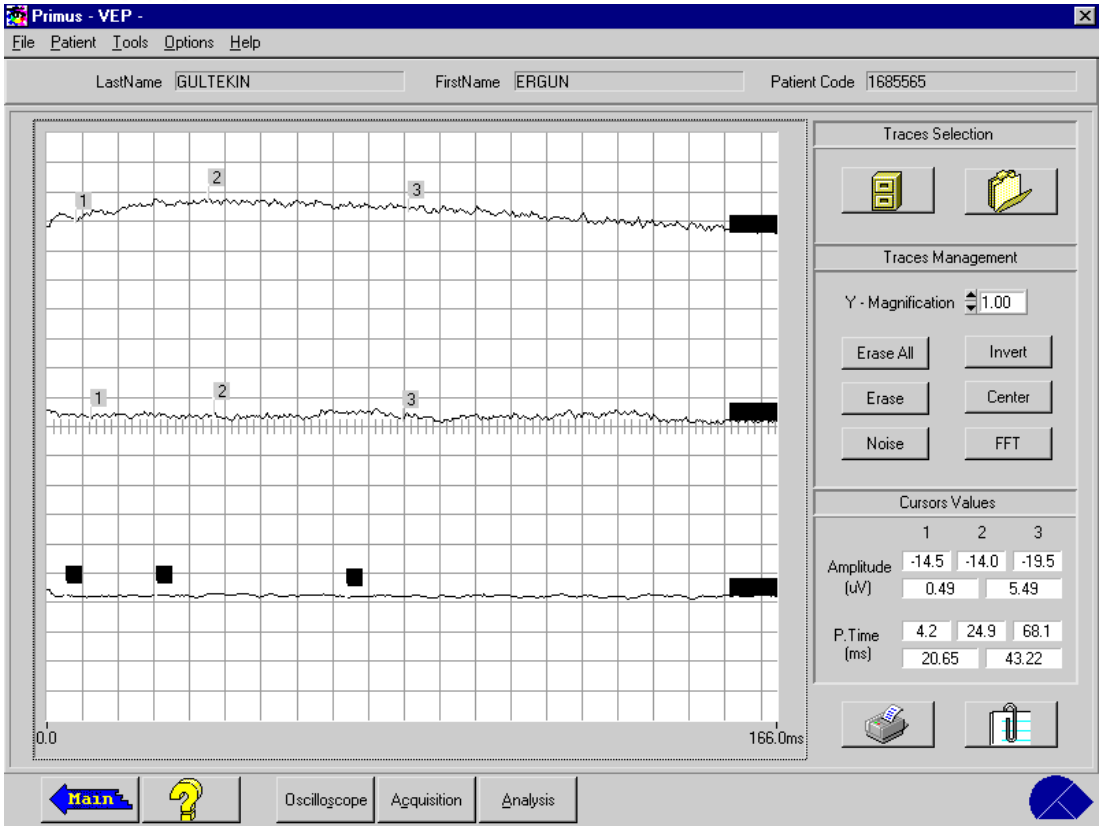








Two examples for visual field examination



An example for ERG

RESULTS

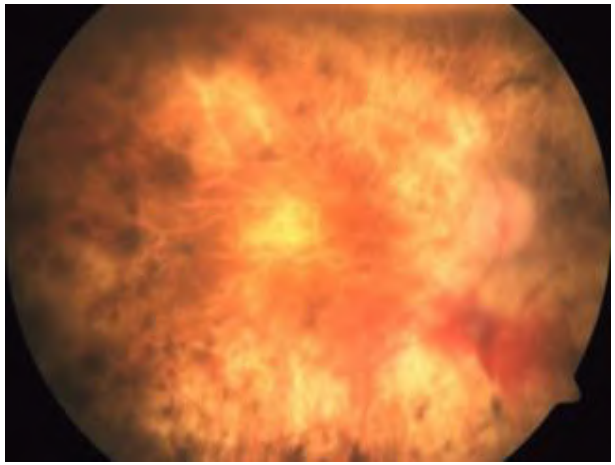
* 11 patients completed 12 month follow-up.
* None of them had systemic complications.
* 5 had no ocular complications.
* One had CNVM at the transplantation site and received intravitreal anti-VEGF drug for once.

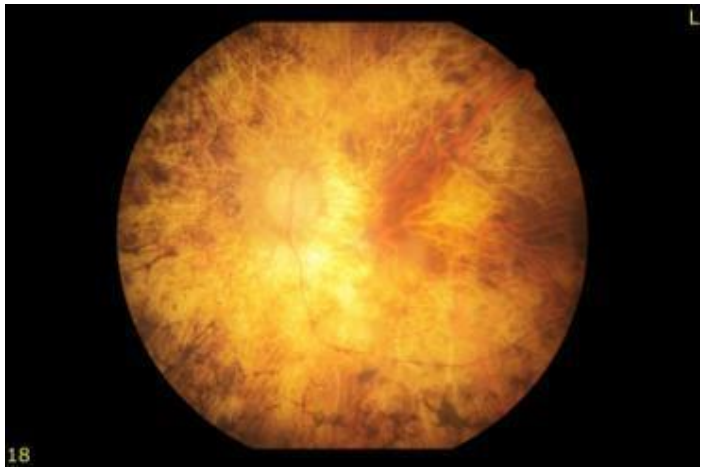
  (Surgical trauma to the Bruchs membrane)
* 5 had ERM around the transplantation area and at the periphery and received second PPV (vitreal reflux of the stem cells or inadvertent preretinal injection of SC)

RESULTS

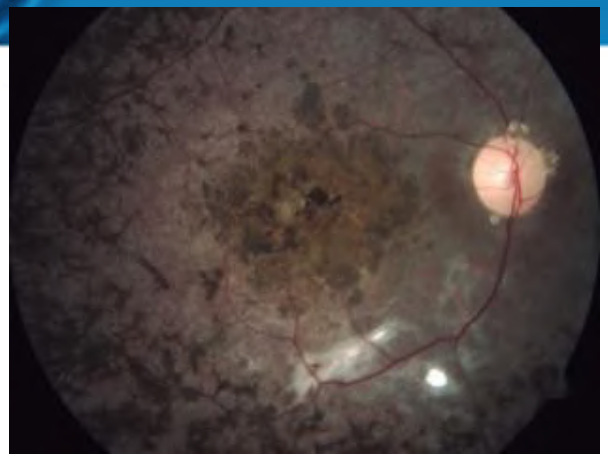
* Only 1 had visual acuity improvement (From 1 mhm to 0.05) and a slight ERG improvement .
* 3 mentioned that the light and some colors are brighter than before although there was not an improvement in VA.
* The other 7 had no VA improvement

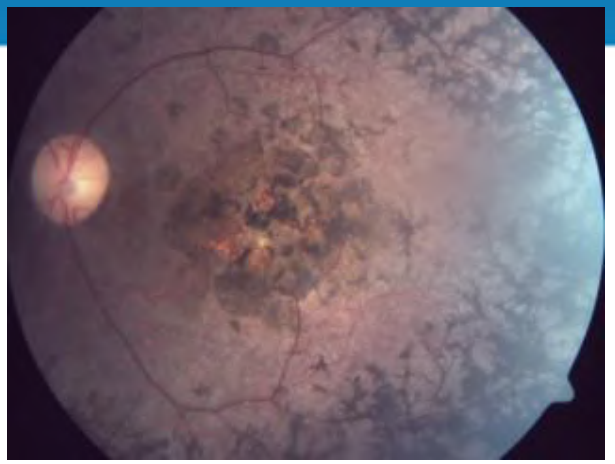
  (6 of them were p+)

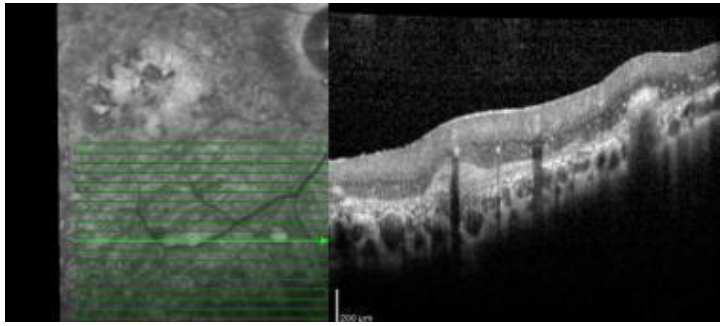


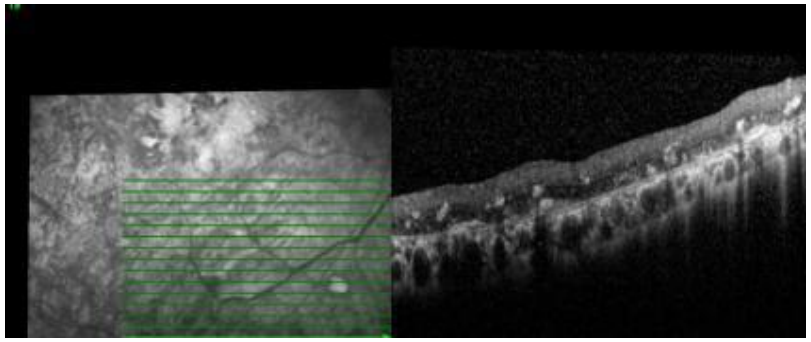


The same patient: Right eye 1 day after surgery and the left eye

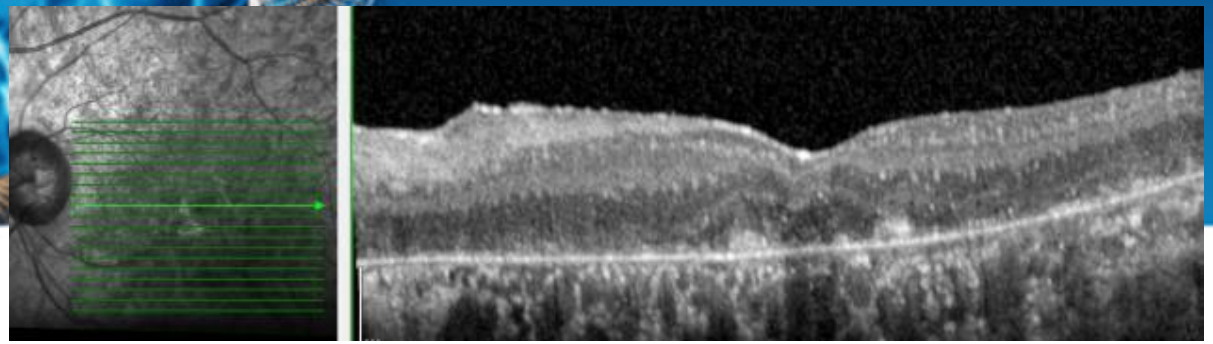




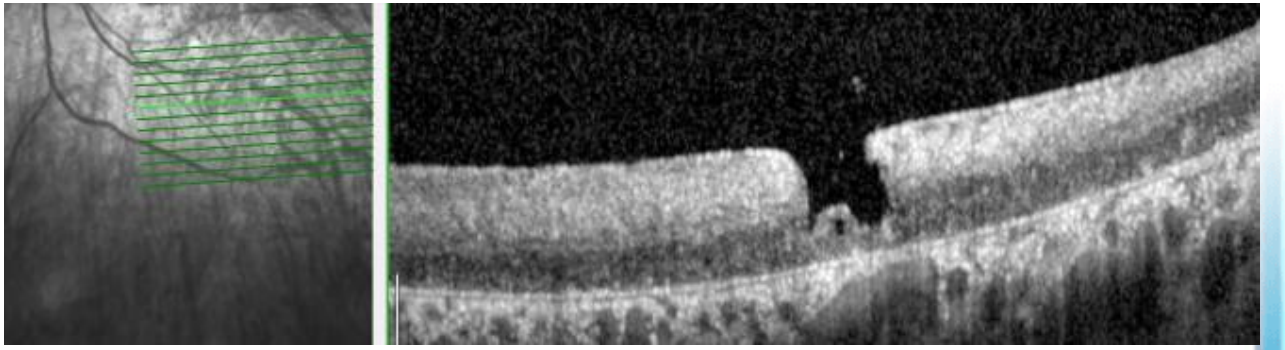




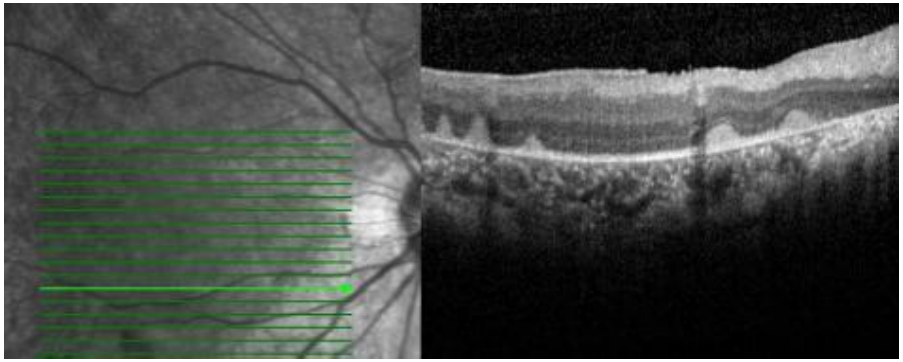
Fundus images of a patient at the sixth month
OCT at first week and at sixth month.
Right eye was treated, left eye was untreated.



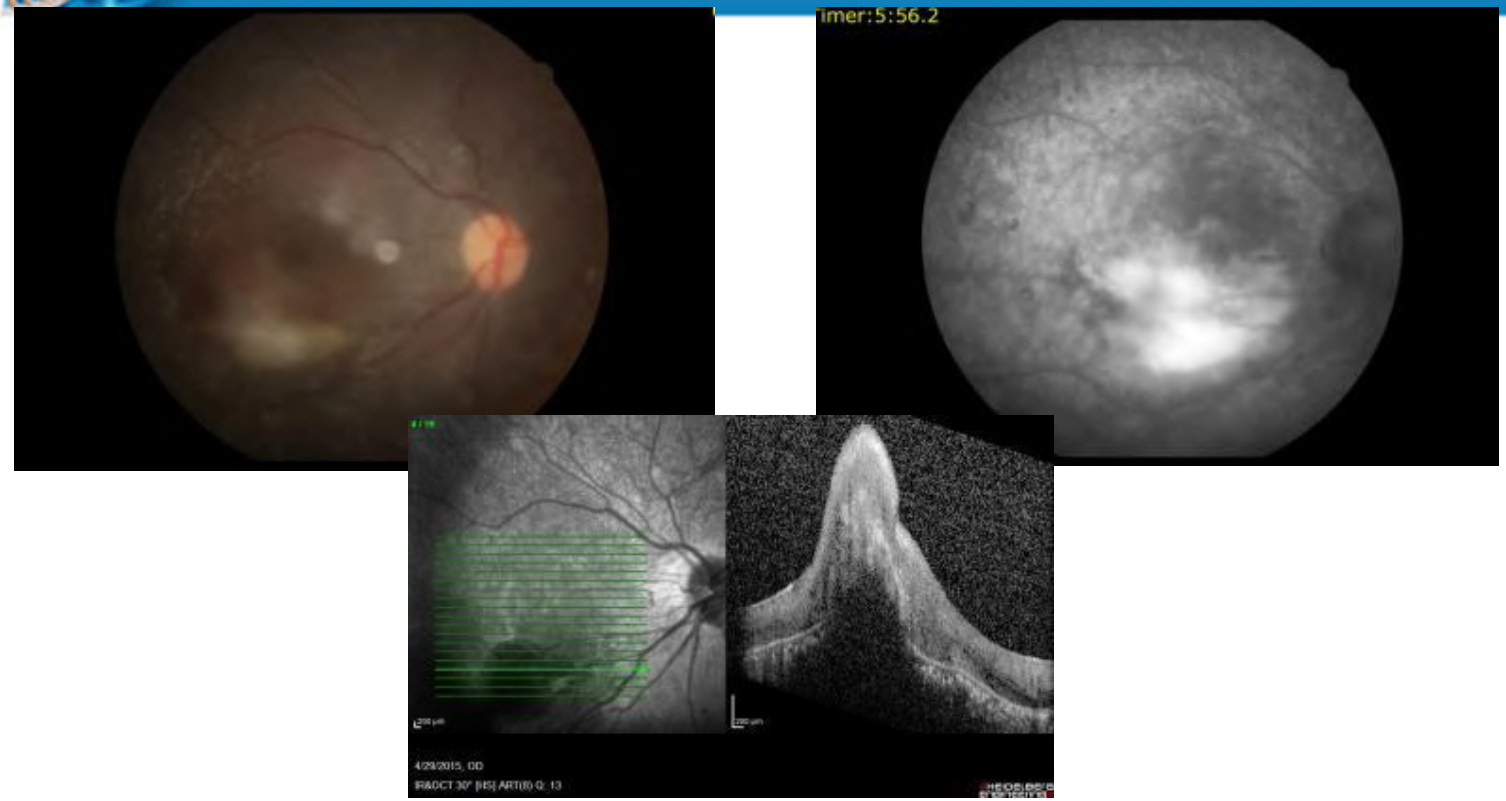
OCT image of a patient at sixth month



Transplantation area on OCT



OCT of another patient after 2 weeks



The patient with CNVM: Fundus image, angiography and OCT

CONCERNS ABOUT SC

SAFETY- Tumor Formation?

* Despite all the advantages of SCs, (especially ESCs) tumor formation is considered an important risk for the clinical application of these stem cells.

* In clinical studies there is no reported tumor formation yet.

* Cui L, et al. WNT signaling determines tumorigenicity and function of ESC-derived retinal progenitors. J Clin Invest 2013; 123: 1647-1661.

IMMUN REACTION ?

- SCs are known to be immunsupressive.
- Subretinal region is belived to be immune privileged because of the presence of the blood–brain and blood–retinal barriers.
- Immunosuppression is not necessary as long as the blood–retinal barrier was not damaged by the transplantation procedure.
- Some researchers recommend immunsupressive drugs for human.

- Hambright, D.; et al. Long-term survival and differentiation of retinal neurons derived from human embryonic stem cell lines in un-immunosuppressed mouse retina. Mol. Vis. 2012, 18, 920–936.

Low Rate of Cell Survival and Migration

* But a low rate of transplanted cell survival is a major problem in stem cell therapy.
* The diseases are progressive and stem cells could be affected from the disease.
* OLM may presents a physical barrier to integration of stem cells into the retina.

* Chen, L.F.; et al. Localization and developmental expression patterns of CSPG-cs56 (aggrecan) in normal and dystrophic retinas in two rat strains. Exp. Neurol. 2012, 234, 488–498.
* Tucker, B.A.; et al. Transplantation of adult mouse iPS cell-derived photoreceptor precursors restores retinal structure and function in degenerative mice. PLoS One 2011, 6, e18992. 114.
* Pearson, R.A.; et al. Targeted disruption of outer limiting membrane junctional proteins (Crb1 and ZO-1) increases integration of transplanted photoreceptor precursors into the adult wild-type and degenerating retina. Cell Transpl. 2010, 19, 487–503.
* Jiang, C.; et al. Laser injury promotes migration and integration of retinal progenitor cells into host retina. Mol. Vis. 2010, 16, 983–990.
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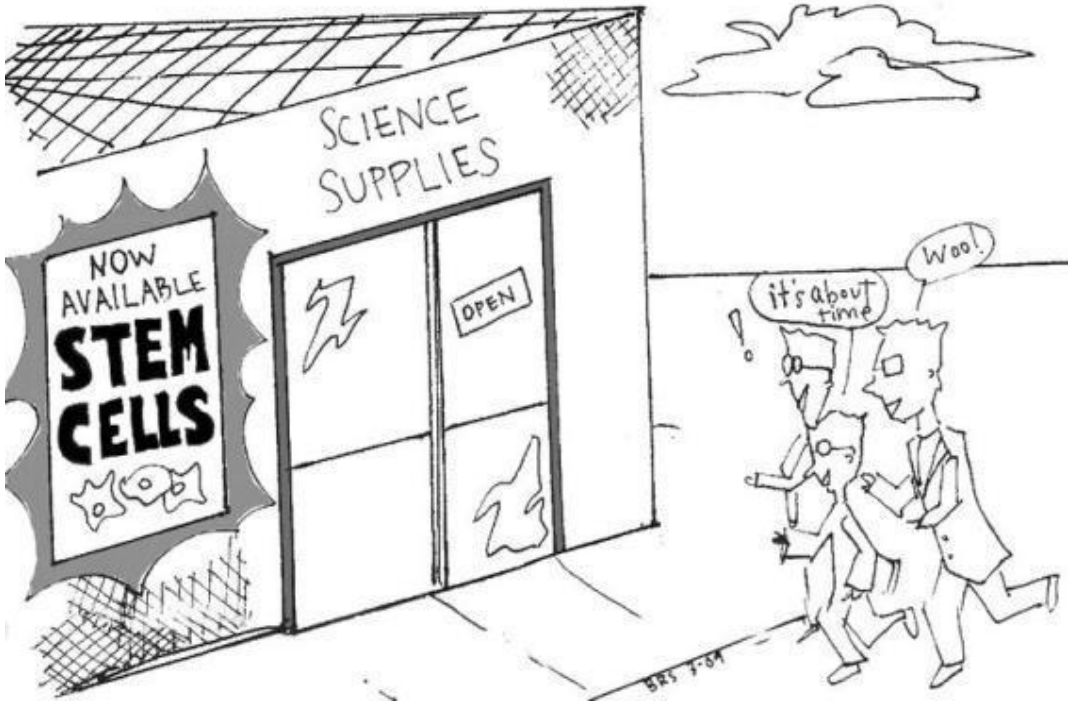
STANDART PROTOCOL?

* Which kind of stem cell is most suitable for stem cell therapy?
* What is the perfect dosage?
* What is the most suitable way ? Subretinal, intravitreal?
* What is the most suitable stage for patients to received the cells?

IN THE FUTURE



THANK YOU FOR YOUR İNTEREST