

ANAT 2341 Embryology Lab 10
8 Oct 2009

Therapeutic Use of Stem Cells

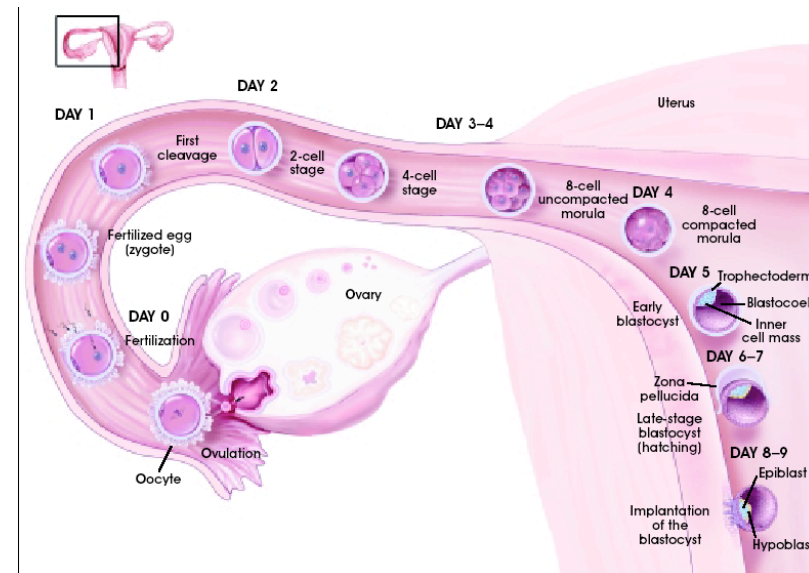
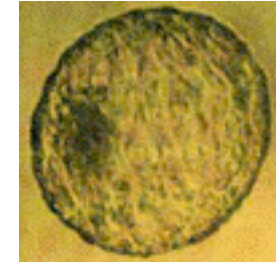
Practical Hurdles & Ethical Issues

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School of Medical Sciences, UNSW

Stem cells in Development

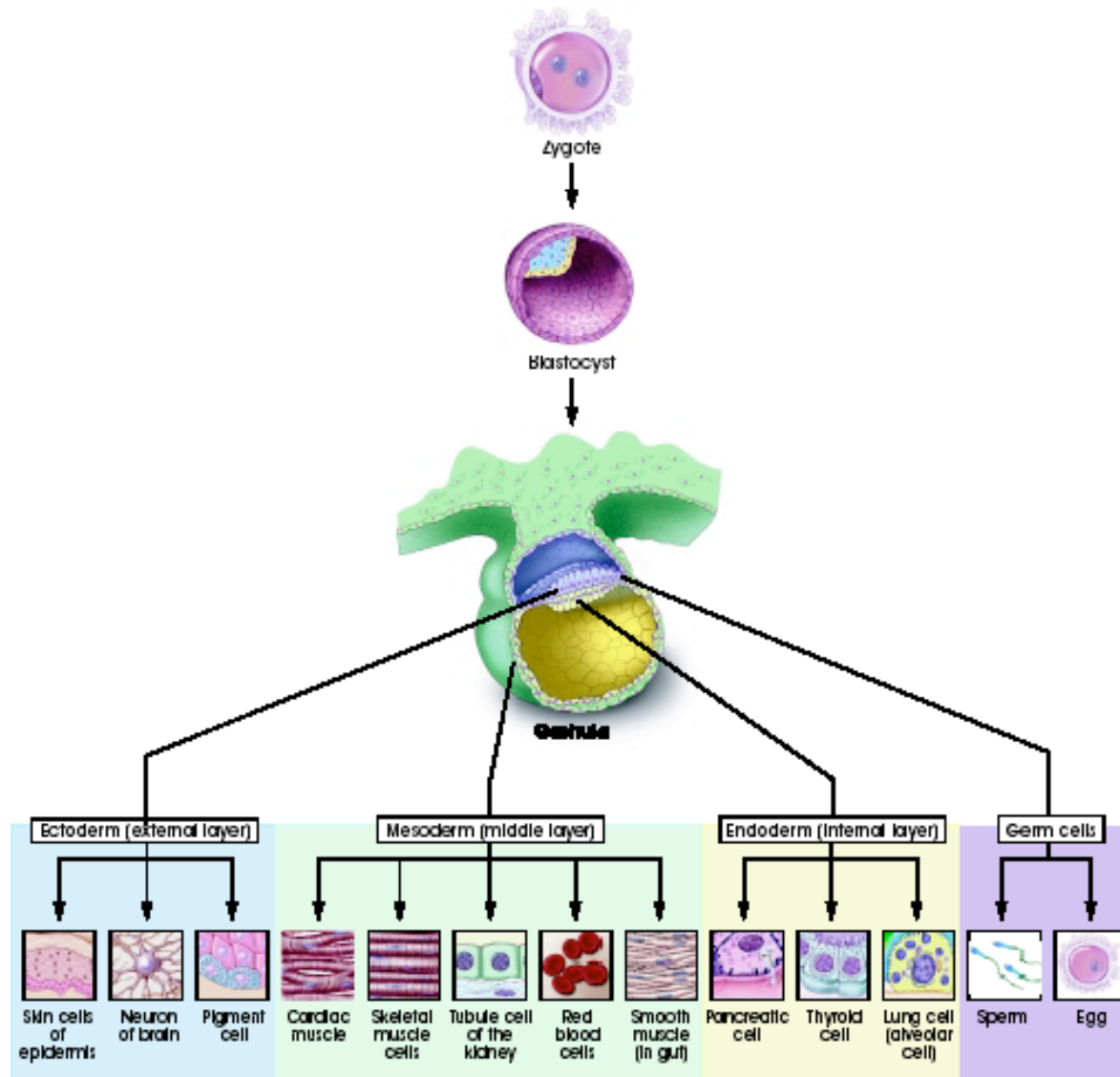
- Blastocyst
- Cord blood



UNSW Embryology

http://anatomy.med.unsw.edu.au/cbl/embryo/Notes/week2_10.htm

Pluripotent Stem Cells



What is a stem cell - Pluripotent

- Pluripotent

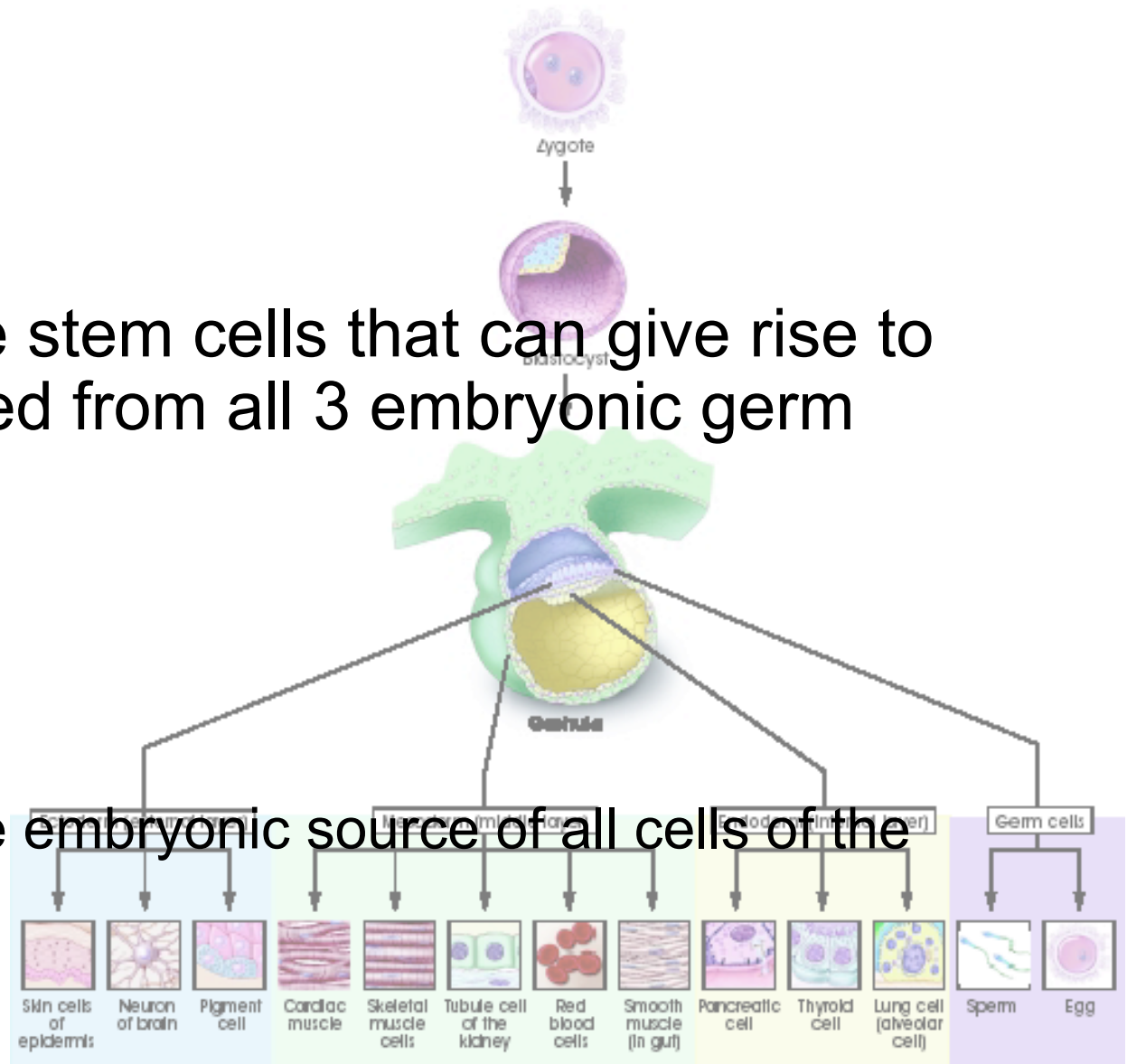
- to describe stem cells that can give rise to cells derived from all 3 embryonic germ layers

- Mesoderm

- Endoderm

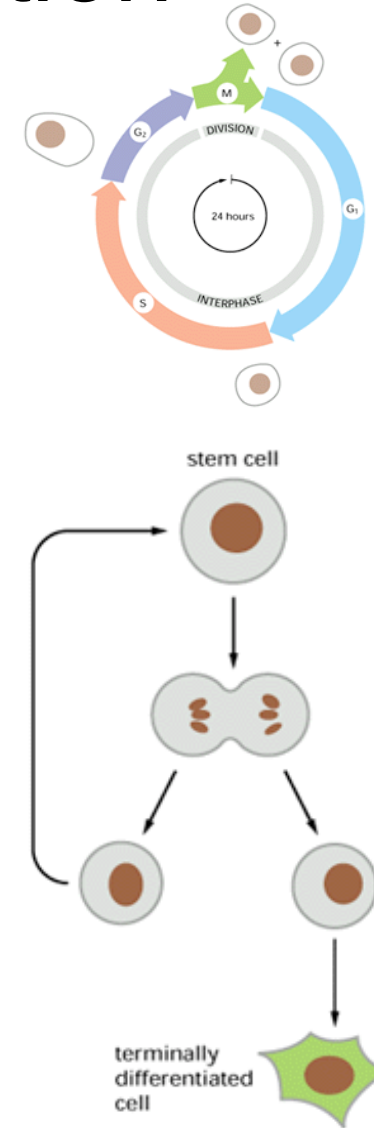
- Ectoderm

- layers are embryonic source of all cells of the body



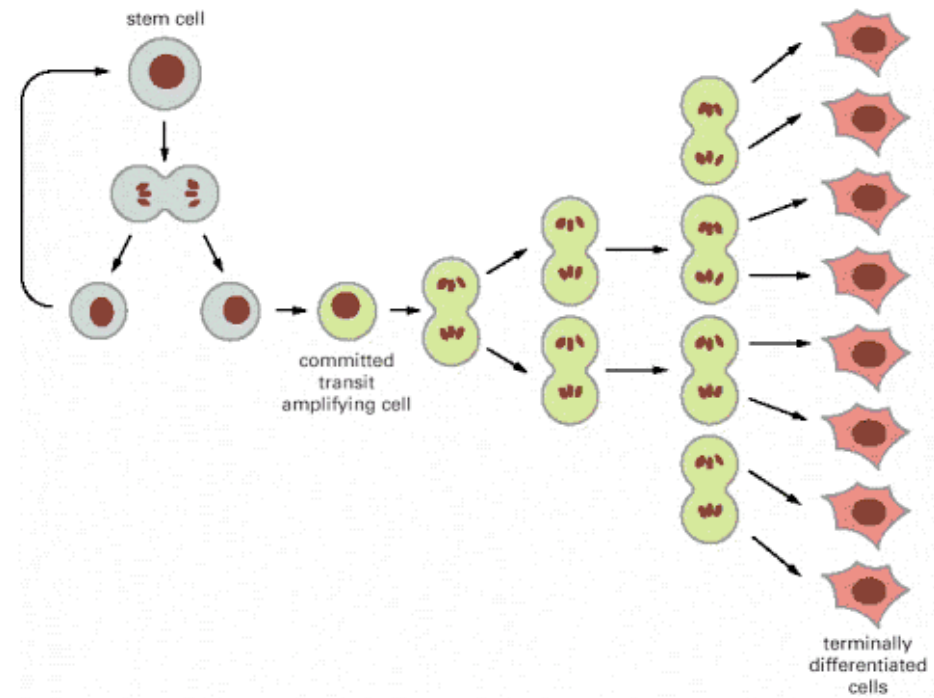
What is a stem cell- Definition

- Stem cell is a cell that has the ability to divide (self replicate) for indefinite periods
 - throughout life of organism
- Under the right conditions, or given the right signals, stem cells can differentiate to the many different cell types that make up the organism



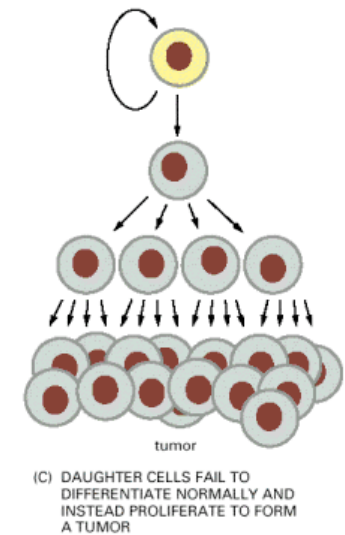
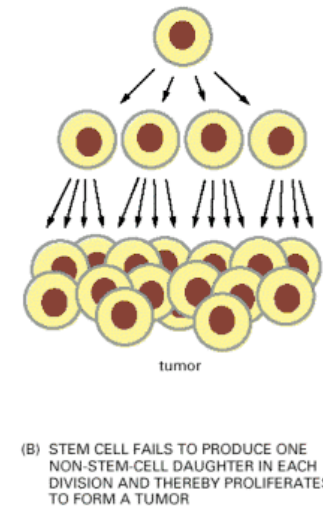
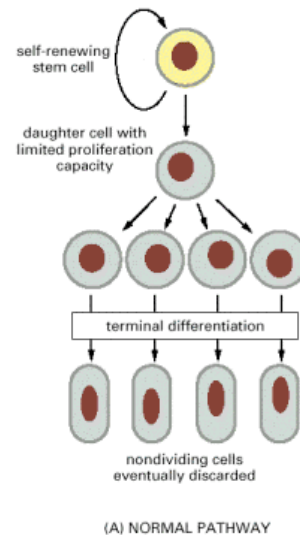
Amplifying Cells

- Stem cells in many tissues divide only rarely
 - give rise to transit amplifying cells
 - daughters committed to differentiation that go through a limited series of more rapid divisions before completing the process.
 - each stem cell division gives rise in this way to eight terminally differentiated progeny



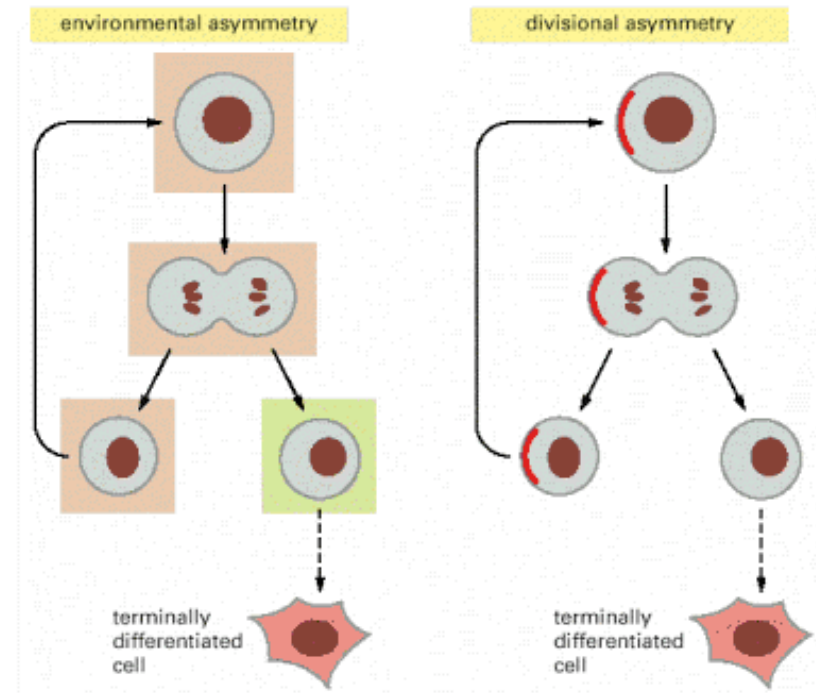
(Ab)Normal Stem Cell Production

- (A) normal strategy for producing new differentiated cells
- (B and C) 2 types of derangement that can give rise to unbridled proliferation characteristic of cancer



Stem Cell Daughter Fates

- environmental asymmetry
 - daughters are initially similar
 - directed into different pathways according to environmental influences that act on them after they are born
 - number of stem cells can be increased or reduced to fit niche available
- divisional asymmetry
 - stem cell has an internal asymmetry
 - divides in such a way that its two daughters are already endowed with different determinants at time of their birth



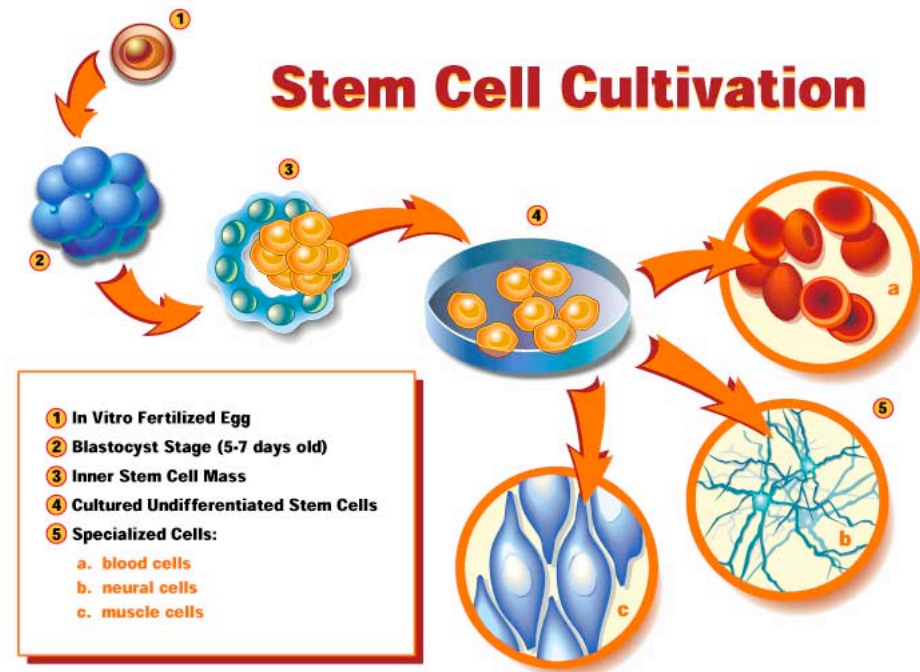
Modified Text/Image: MBoC Fig 22-5

Possible Therapeutic Uses

- Neural
 - Parkinson's, ALS, spinal cord injury.....
 - Cell Replacement
 - cell death, loss of function
 - Grafting
 - where host-graft rejection normally requires substantial ongoing immunosuppression
 - Repair
 - Spinal cord and brain injury
- Other Diseases
 - Diabetes, muscular dystrophies, cardiac, vital organs.....

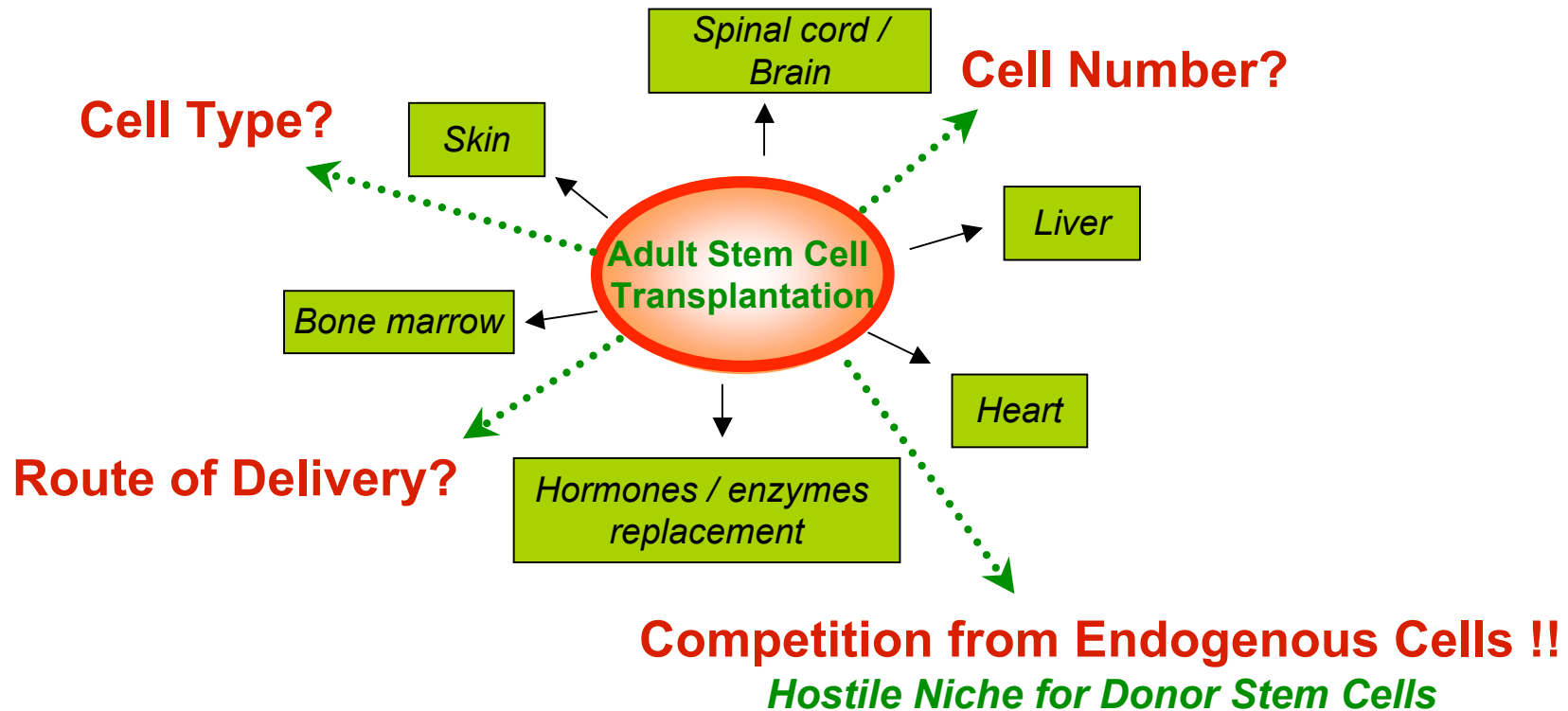
Current research on stem cells

- How to:
 - Isolate
 - Grow
 - Maintain, store
 - Differentiate
 - Therapeutic uses



Stem Cell Therapy :

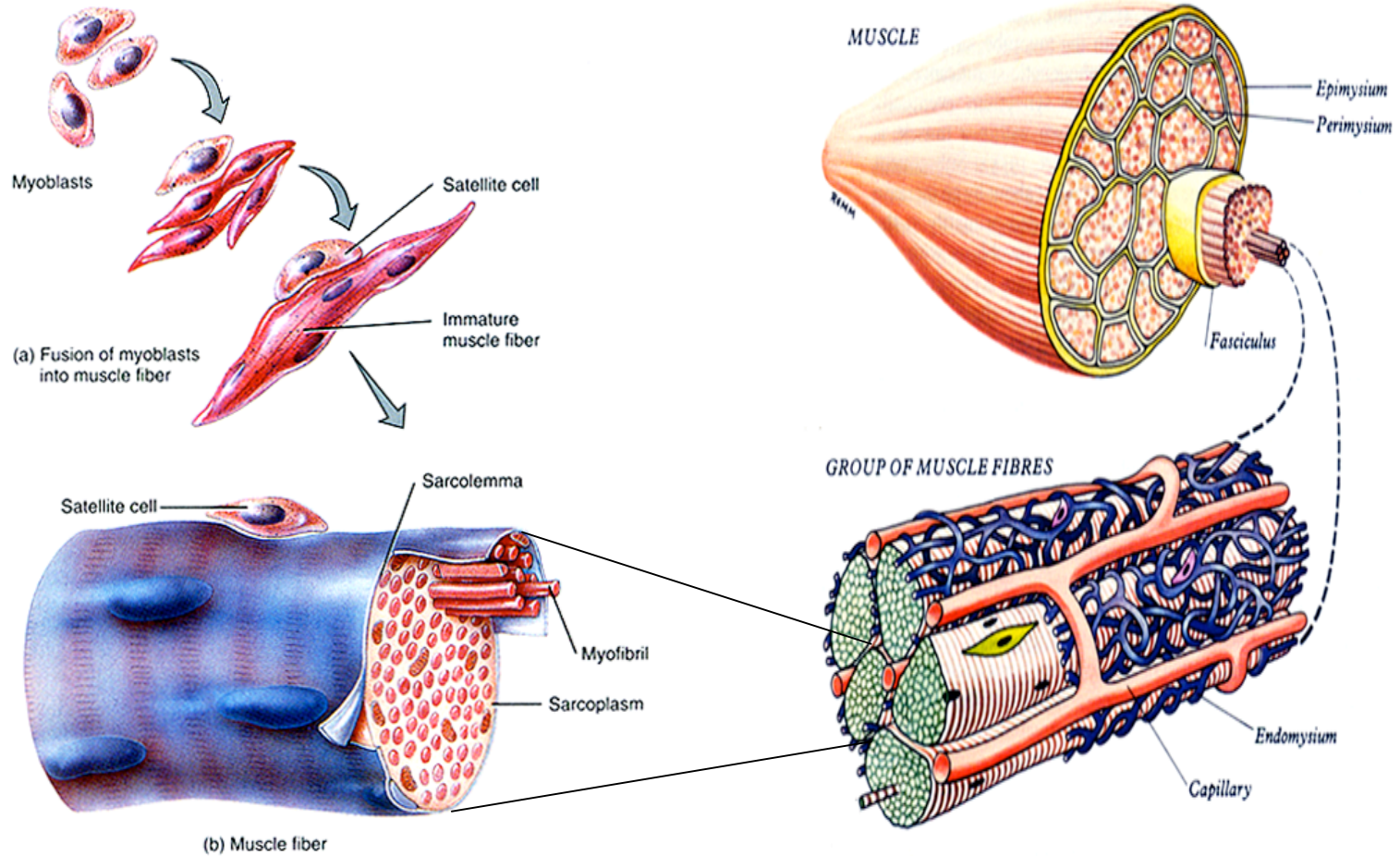
Current Limitations on Cell Transplantation



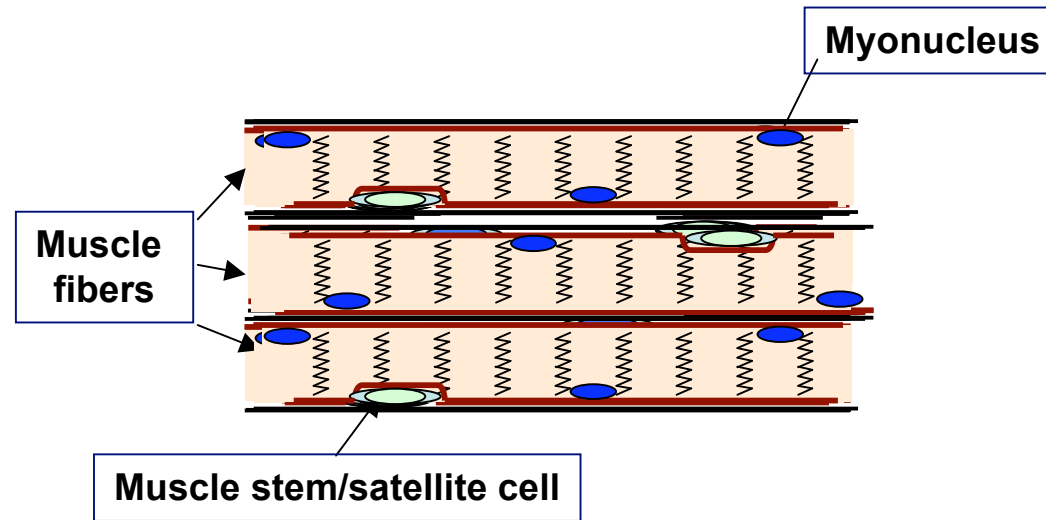
Enhancing Muscle Stem Cell Transplantation using Chemotherapeutic Drug Selection

- **Alkylating Chemotherapy + Drug Resistant Donor Cells -**
based on mechanisms established for Bone Marrow Transplantation
 - Efficient Elimination of Endogenous Cells
 - Creating Receptive & Favourable Niche for Donor Cells
 - Selective *in vivo* Expansion of the Protected Donor Cells
 - Feasibility in the Skeletal Muscle as a Solid Organ?

Skeletal Muscle Biology



Skeletal Muscle during Injury



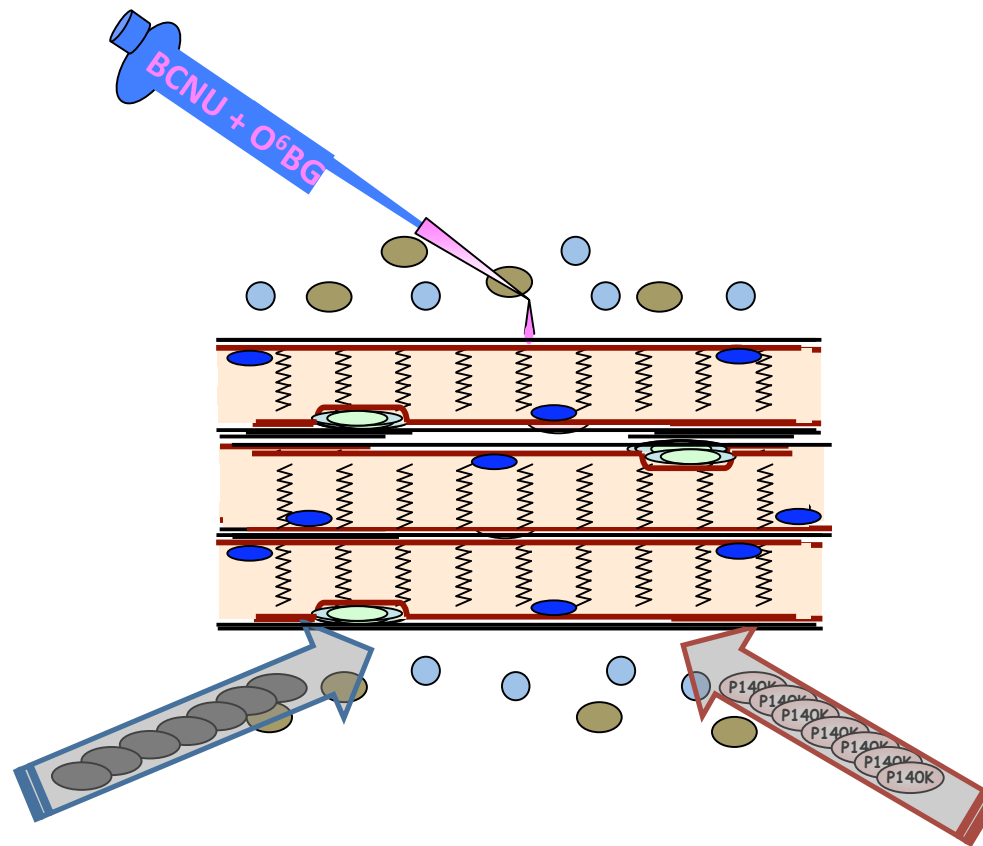
Normal Muscle

- Muscle fibers and myonuclei are post-mitotic
- Muscle stem / satellite cells remain quiescent

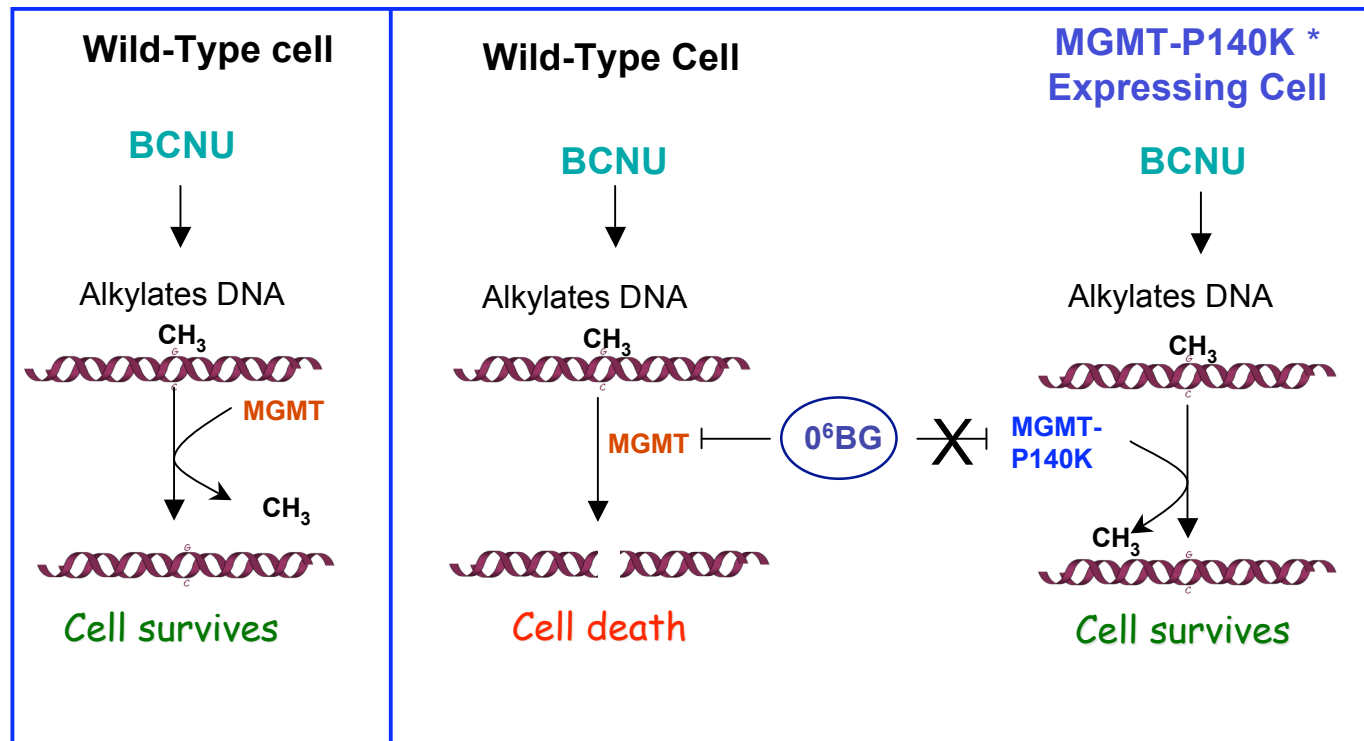
Injured Muscle

- Muscle stem / satellite cells are activated and rapidly proliferate
- Differentiated cells align and fuse to form new muscle fibres

Muscle Stem Cell Transplantation - Improved Strategy



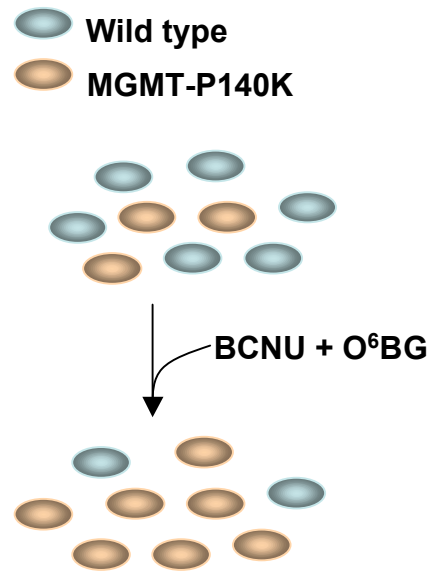
Selective Enrichment : The Mechanism



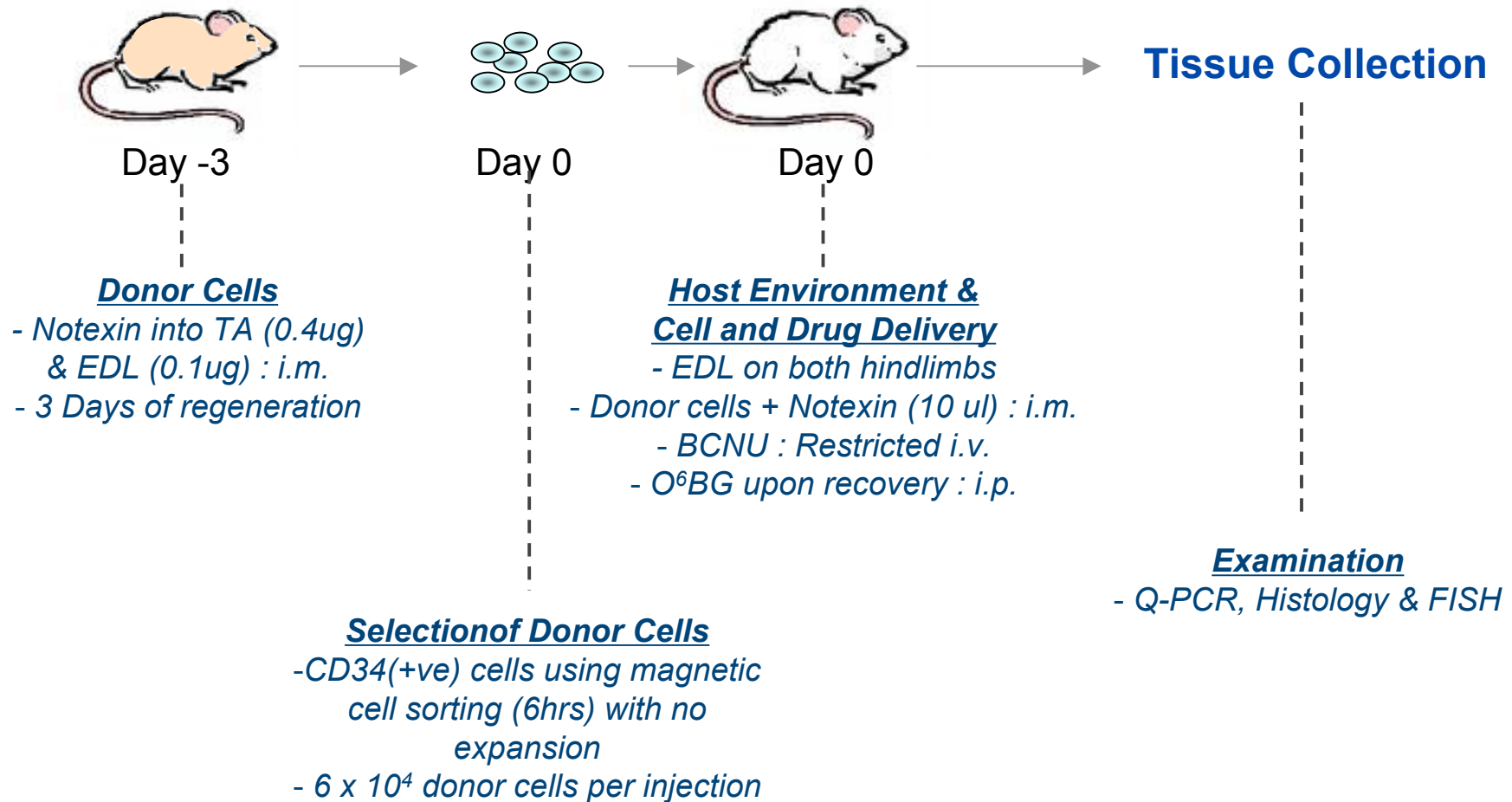
* Anthony Pegg

Selective Enrichment : The Mechanism

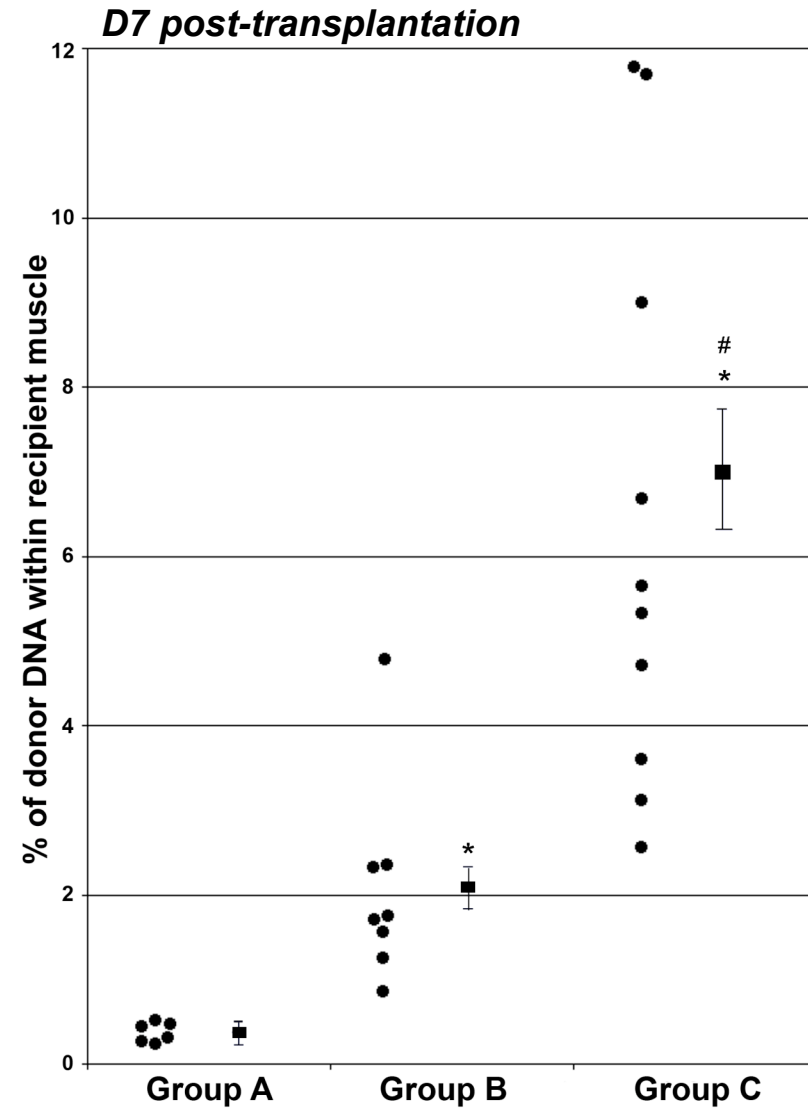
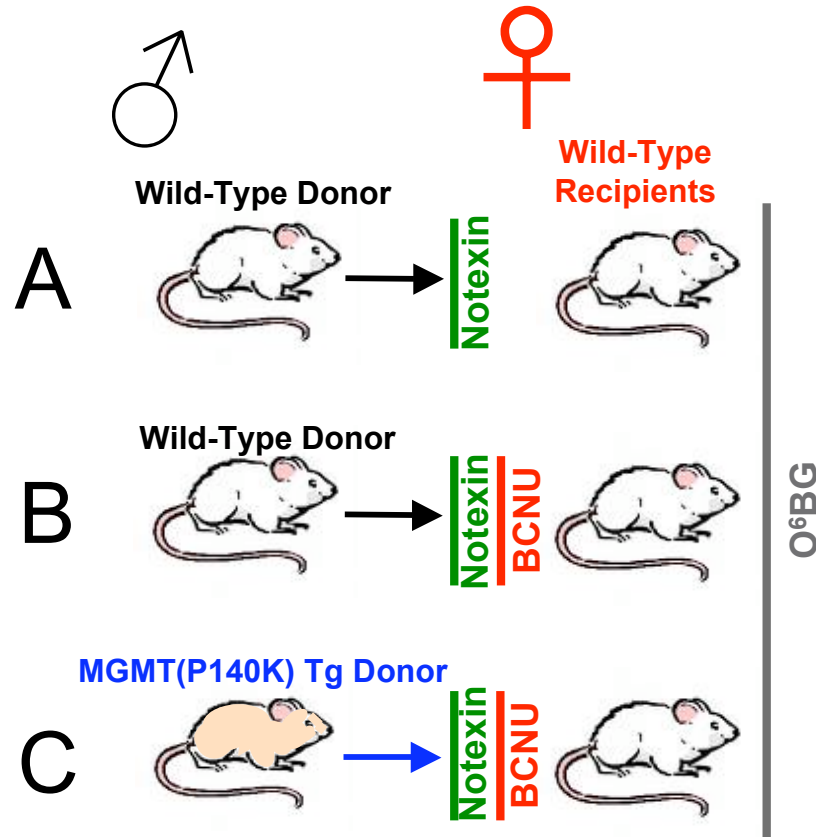
SELECTIVE ENRICHMENT



Muscle Stem Cell Transplantation : Protocol



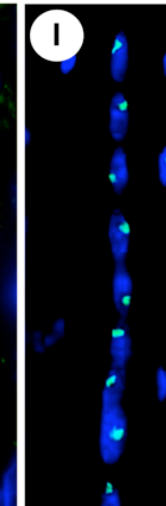
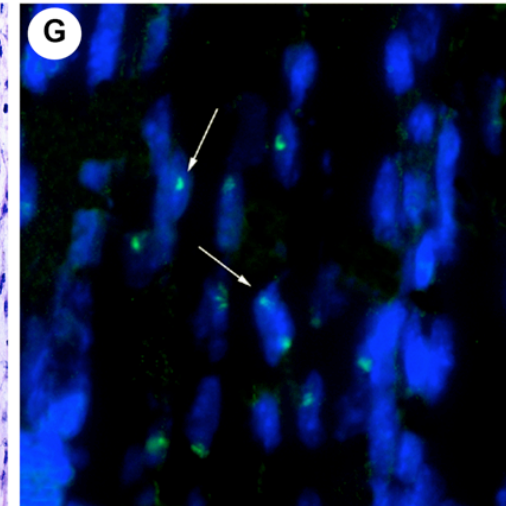
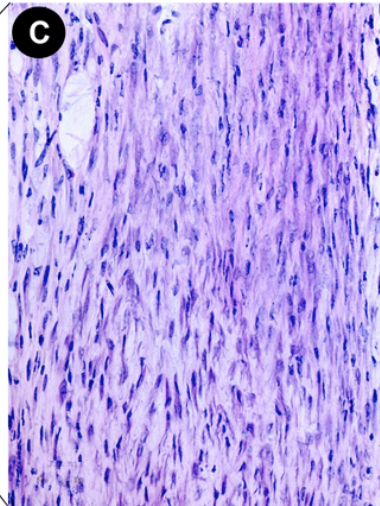
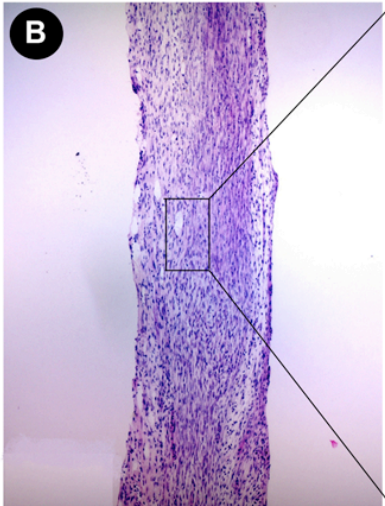
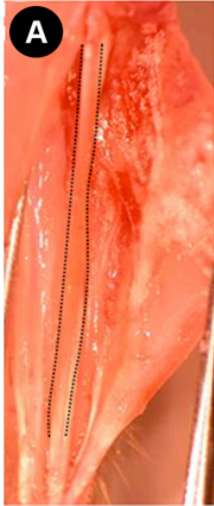
Higher Engraftment of MGMT(P140K)^{+ve} Donors in Chemo-Ablated Recipient Muscle Bed



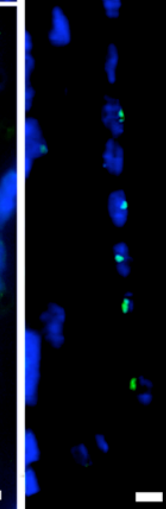
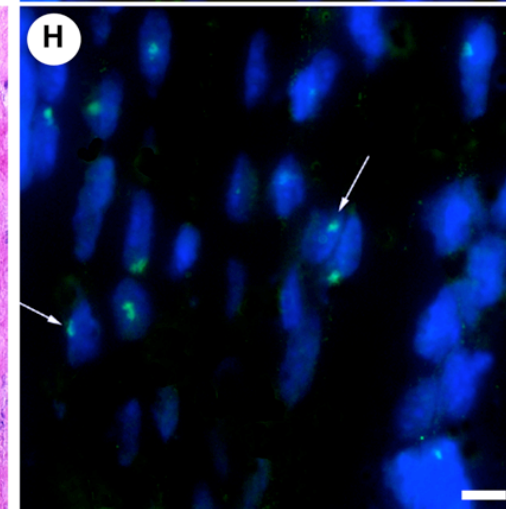
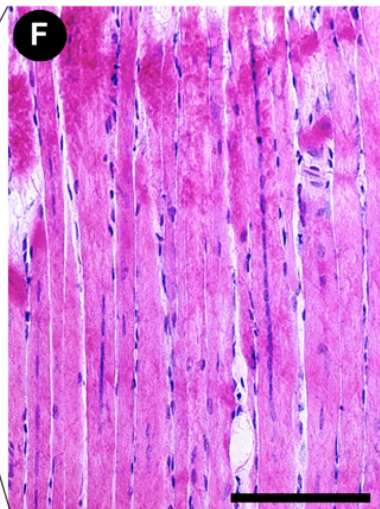
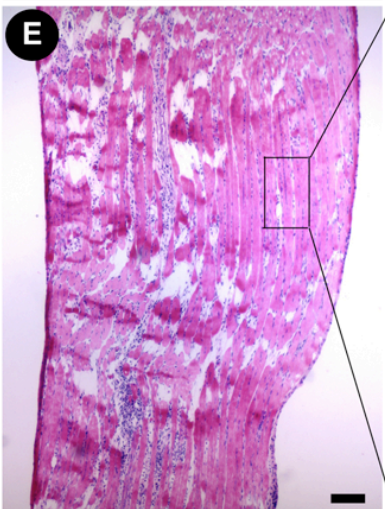
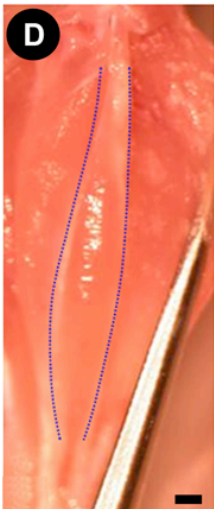
De Novo Muscle Fibre Formation by MGMT(P140K)^{+ve} Donors

D14 post-transplantation

Wild-Type Donor



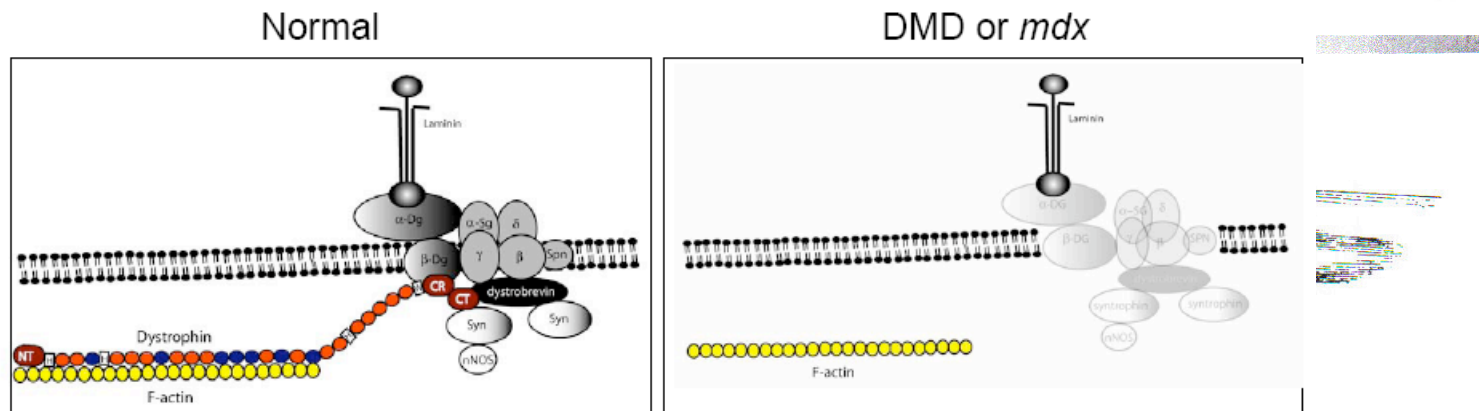
MGMT(P140K) Donor



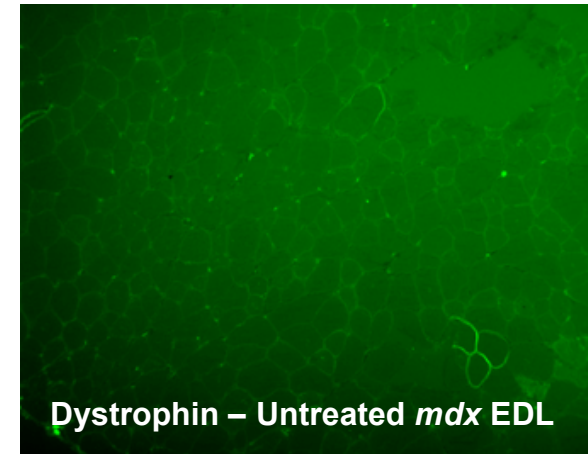
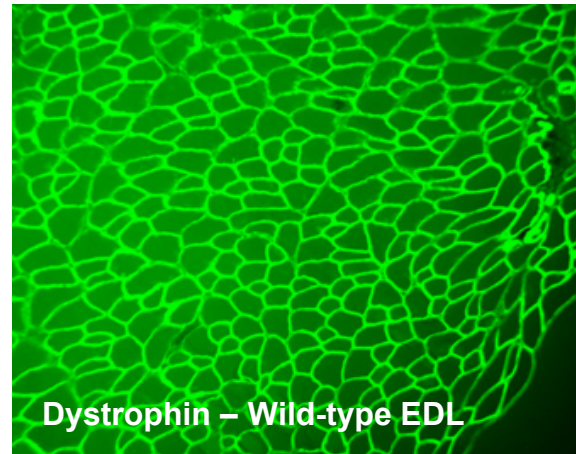
Y-Chromosome FISH / DAPI

Absence of Dystrophin in the Duchenne Muscular Dystrophy (DMD) Patients and *mdx* Mice

- **Duchenne Muscular Dystrophy (DMD)**
 - X-linked disorder with defects in Dystrophin gene
 - 1:3500 live Male Birth (20,000 babies / year)
 - Confined to wheelchair by 12 yrs and death by 30 yrs
 - Several mouse models exist including *mdx* mice (Dystrophin KO)



Restored Dystrophin Expression by Engrafted MGMT(P140K)^{+ve} Donors in the Recipient *mdx* Muscle



Notexin
+
BCNU
+
O⁶BG

