Stem Cells and Regenerative Medicine

Optimizing Stem Cells through hormones and neutraceuticals

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What is the name of our Specialty?

- Anti-Aging and Regenerative Medicine
- What are we doing about the Regenerative?
- Stem cell transplantation
  - Autologous
  - Allogeneic

Stem cells – Clinical Trials ClinicalTrials.gov

March 20, 2010

2863 Adult Stem Cell Clinical Trials

129 Umbilical Cord Stem cell Clinical Trials

Stem Cells
are the Cornerstone of
Regenerative Medicine

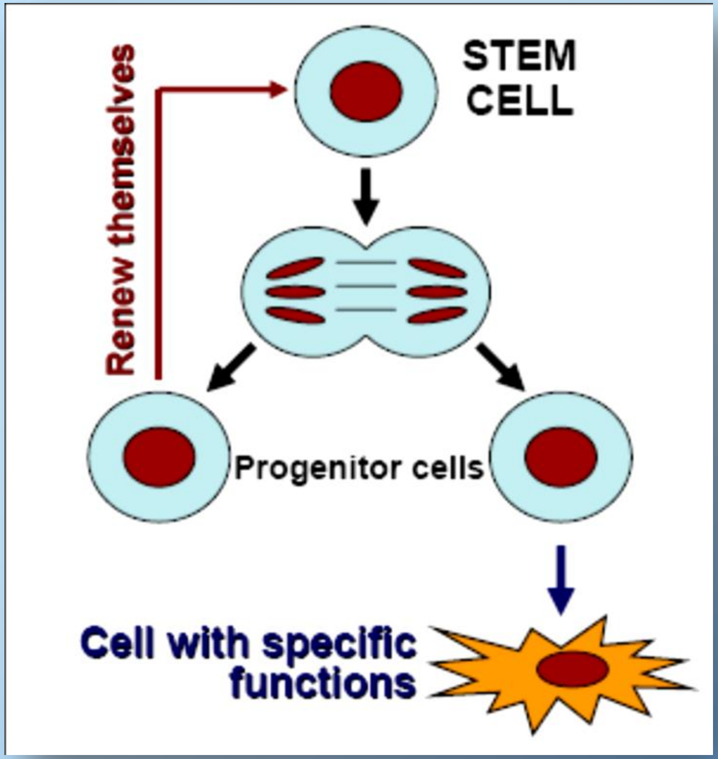
Stem Cell Therapy is
“Cure Directed”

Successful treatment with Stem Cells

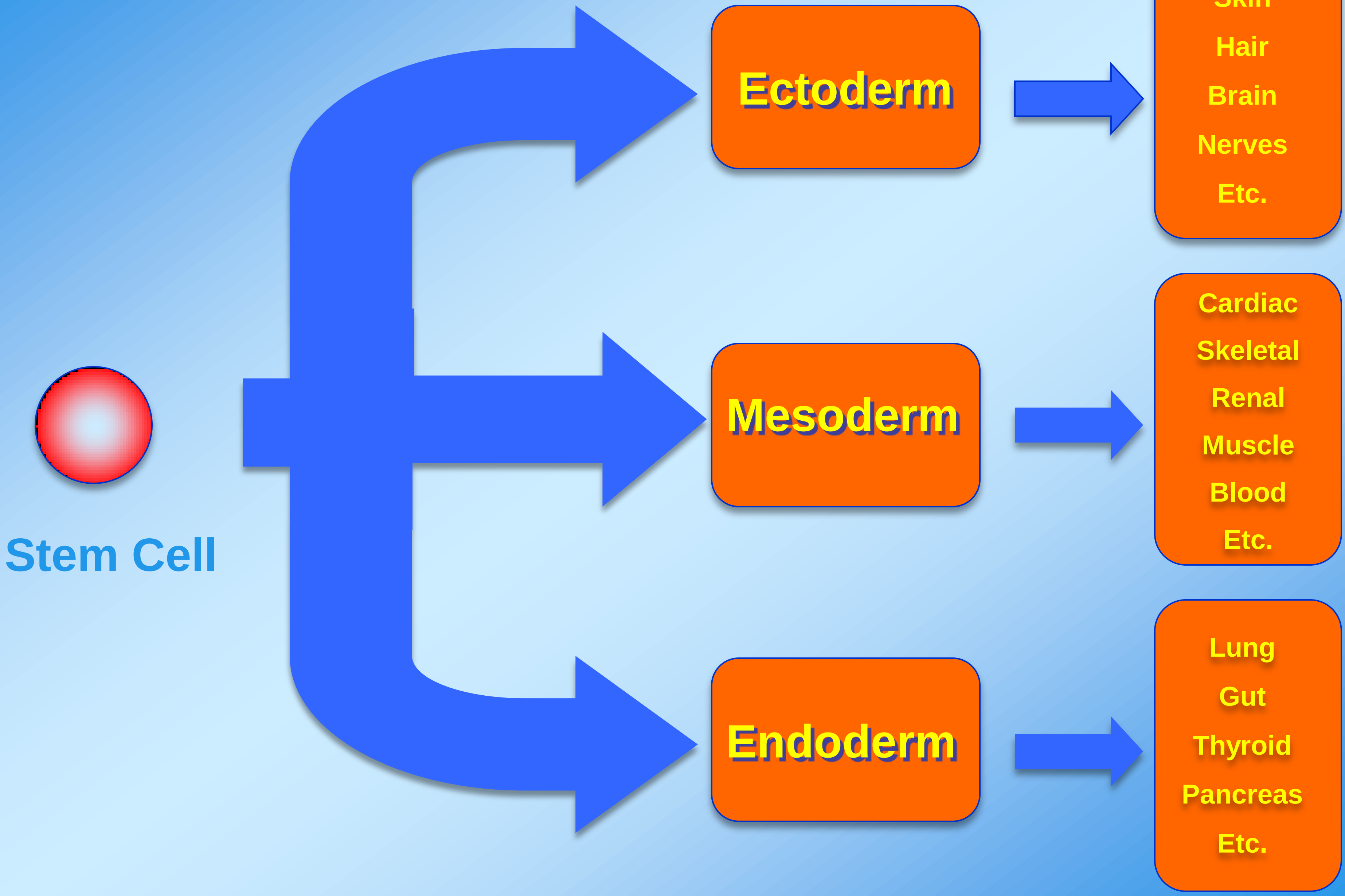
- Lupus
- MS
- Scleroderma
- Type 1 Diabetes
- Type 2 Diabetes
- Orthopedic Uses
- Peripheral Vascular Disease
- Cardiovascular uses
  - Acute MI
  - CHF
  - Severe Angina

What Are Stem Cells?

- UNSPECIALIZED, PRIMITIVE
- SELF-RENEWING
- CAN DIFFERENTIATE INTO CELLS WITH SPECIFIC FUNCTIONS



Stem Cells and Pluripotency

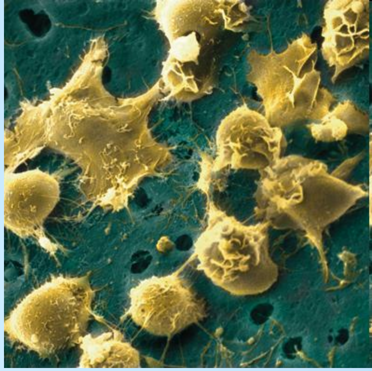


Controversial Stem Cells

- Embryonic Stem Cells
- Fetal Stem Cells

No Therapeutic use at this time

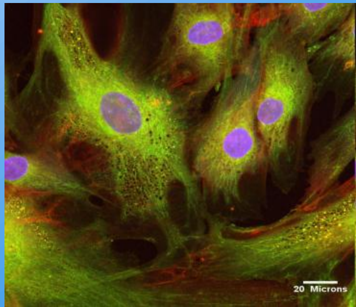
Important for research



Non-Controversial Adult Stem Cells

- Umbilical Cord Blood Stem Cells
- Bone Marrow Stem Cells
- Adult Peripheral Blood Stem Cells
- Adipose derived Mesenchymal Stem Cells

Therapeutic



Stem Cell Infusion (Transplant)

- Autologous = Self
- Allogeneic = Non–Self

Comparison of Adult Stem Cell Sources

	Typical Collection			Typical Dose (CD 34+)		
	Volume	TNC	CD34+	Diabetic Ulcers	Cardiac	Immune Reconst.
Mobilized perif. blood	300ml	28×10^9	1×10^8	5×10^6	1.6×10^7	1.2×10^6 / Kg
Cord Blood	75 ml	7.5×10^8	3.7×10^6			1.7×10^5 / Kg

References:

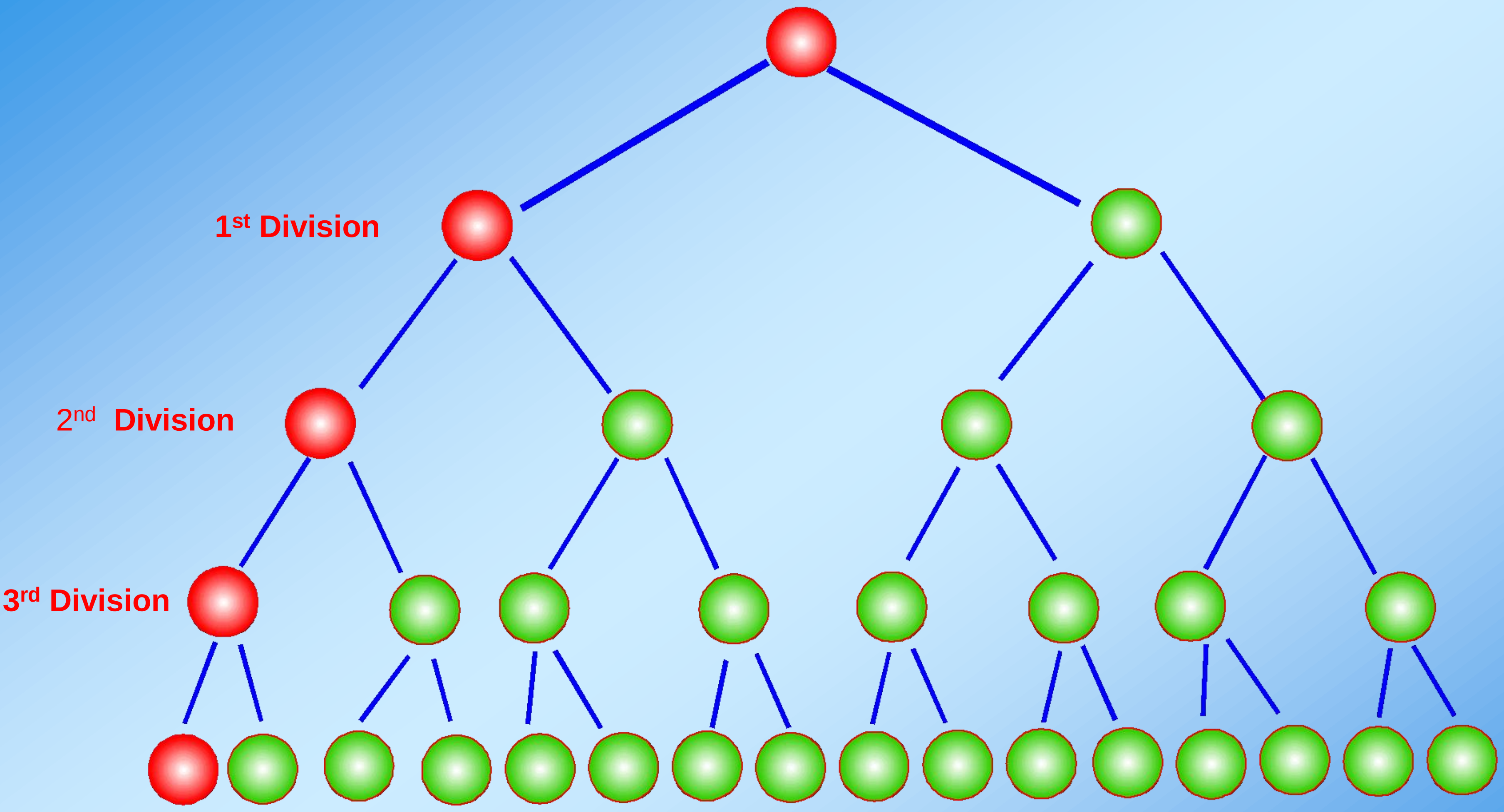
Badiavas & Falanga, *Arch. Derm* 139:510, 2003

Zeiher (Schachinger et al) *J. Amer. Coll. Cardiology* 44:1690, 2004

Zubair et al *Transfusion* 44:253, 2004

Schoemans et al *Bone Marrow Transplantation* 38:83, 2006

Asymetric Kinetics of Stem Cell Division



Mobilization Phase – Adult Autologous

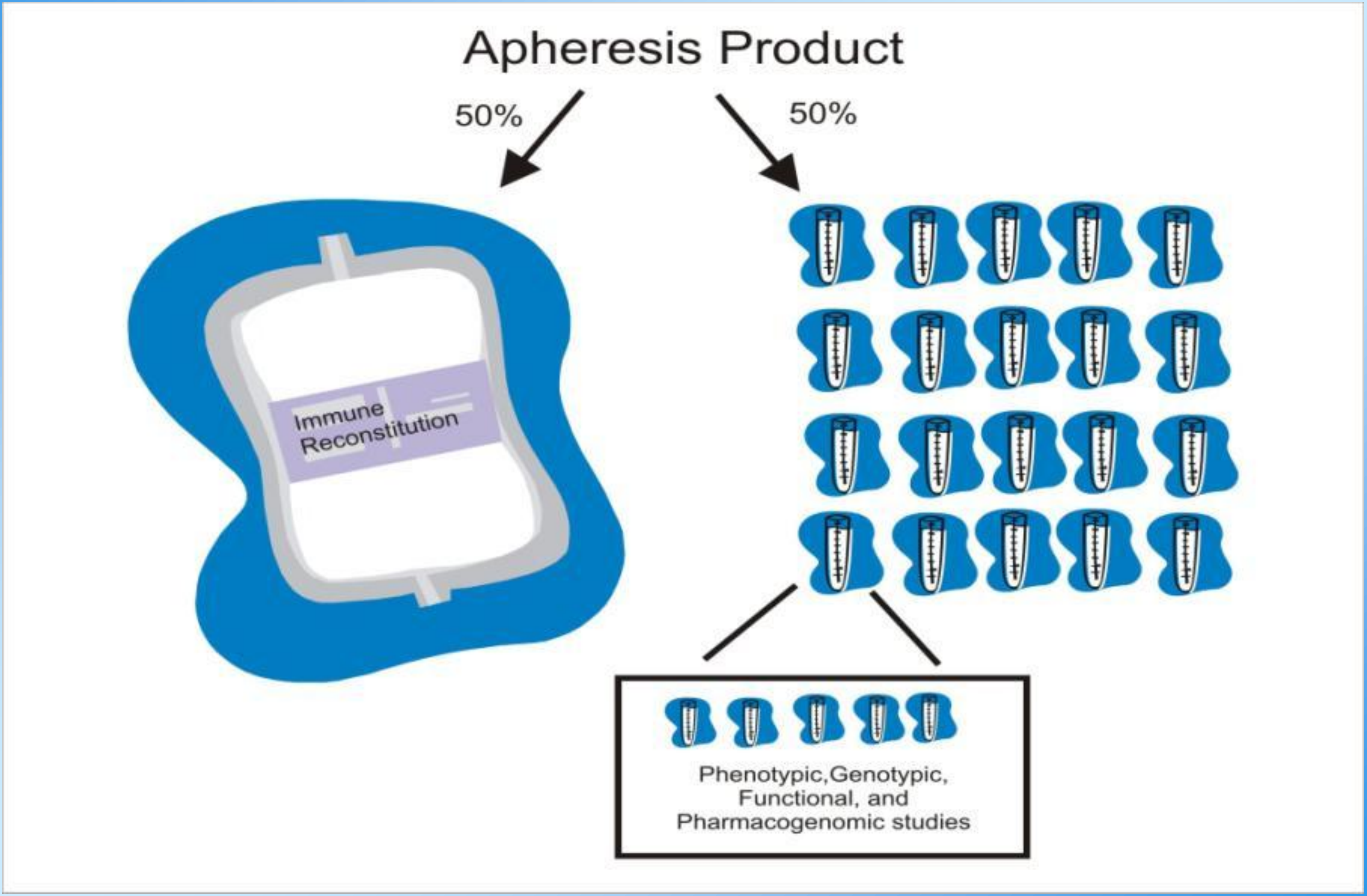
- Neutraceuticals to stimulate stem cell production and release
- After the patient has received his or her physical exam and is negative for the Infectious Disease Screen:
- Administration of SQ injection of G-CSF (for two consecutive days)
- The Adult Stem Cell Collection occurs on the third day
- The collection process usually takes under 3 hours and is safe and non-invasive

Apheresis

(Greek: "to take away")

Cobe “Spectra”

CryoPreservation & Storage



Cryopreservation

- The prepared cells are then cooled in a controlled-rate freezer and transferred to our liquid nitrogen tanks for long-term storage

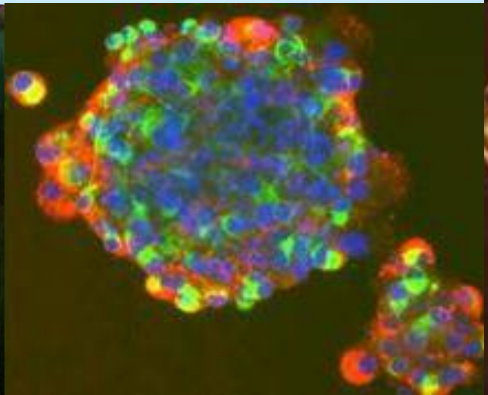
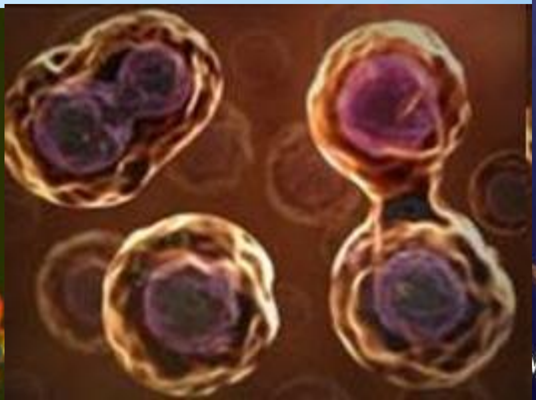
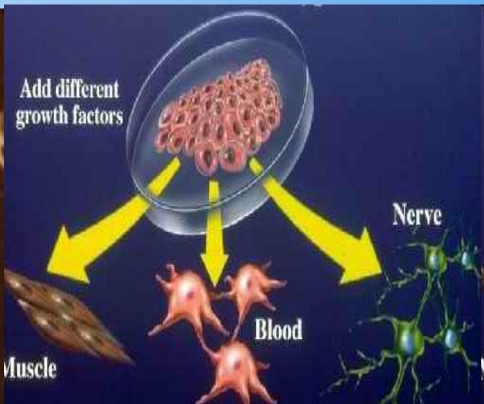
Long Term Viability of Stored Stem Cells

- No significant loss of cell viability in stem cells stored for up to ten years
- Viability may decrease at approx 0.3% per decade.

McCullough J, Clay M, Wagner JE. Cord blood stem cells. In Ball ED, Lister J, Law P, editors: *Hematopoietic Stem Cell Therapy*. Philadelphia: Churchill Livingstone, 2000: 287-297.

Umbilical cord stem cells

- No ethical controversy
- Full telomeres
- Low Infectious disease risk
- Matched/Related donor not necessary

Umbilical Cord Stem Cells

- Immediately after delivery, cord obtained and placed in ACD bag.
- Blood/Cells are pulled off and screening done on sample
- It is spun down and buffy coat obtained
- It is separated into progenitors and mesenchymals.
- The mesenchymals further differentiate based on growth factors that are added
- Placed in cryopreservation until needed

Why Anti-Aging medicine needs Stem Cell transplants now

- With the interventions of anti-aging medicine, many patients do well until about 80
- Very few live with full HealthSpan after 80-85
- Regenerative therapies needed to “rectangularize the HealthSpan cure
- Anti-aging patients are ready for regenerative therapies now
- Adult autologous and Umbilical allogeneic cord stem cells
  - Plasticity
  - Availability
  - Free from Political and Ethical Debate

What are adult stem cells?

- Adult stem cells are cells capable of dividing and replacing damaged tissue.
- Exist in tissues such as the bone marrow, brain, muscle and liver, adipose etc.
- Unlike their neighbors, which are already differentiated into specialized cell types, adult stem cells remain immature.
- If you are a stem cell you are either an embryo or an adult – no in between

What are adult stem cells?

- When tissue becomes damaged, the adult stem cells divide (self-renewal) to produce new cells.
- Some of the resulting cells divide, but unlike stem cells they mature to take over for the damaged cells.
- They become progenitors

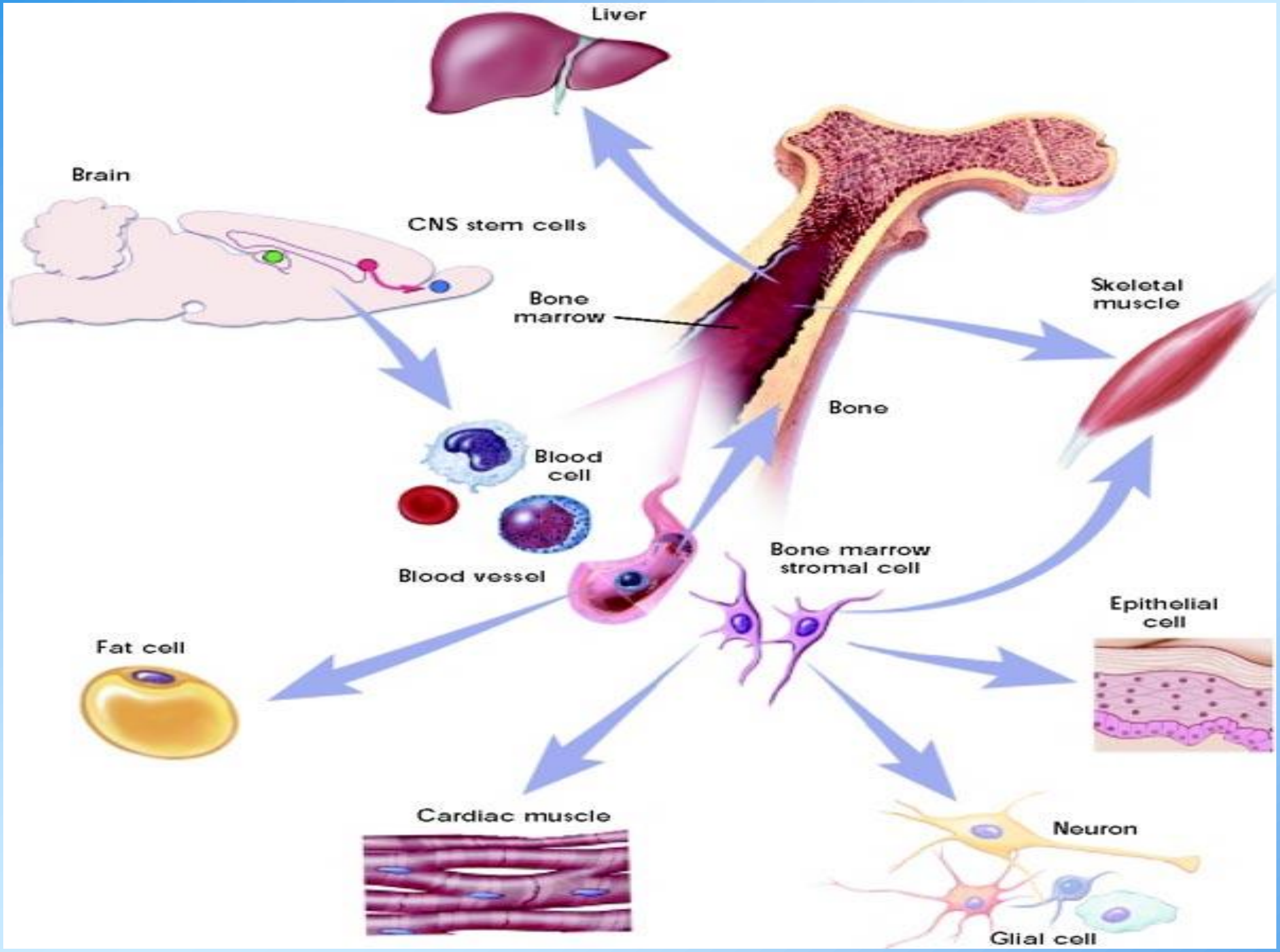
Hematopoietic stem cell Plasticity

- Bone marrow contains various stem and progenitor cells with a differentiation capability exceeding hematopoiesis – plasticity

- Stem cells in bone marrow are, mesenchymal stem cells, and multipotent progenitor cells.

- Generation of neuronal tissue, cardiomyocytes and functional hepatocytes in injured tissue from bone marrow stem cells

Kronenwett R et al. Differentiation potential of stem cells from bone marrow : *Med Klin* (Munich). 2006 Mar 22;101 Suppl 1:182-5.



•Kronenwett R et al. Differentiation potential of stem cells from bone marrow. Med Klin (Munich), 2006 Mar 22; 101 Suppl 1:182-5.

VSEL

- Express a multitude of pluripotent stem cell markers
- Genes found in skeletal muscles, heart, neural cells, liver, intestinal epithelium, skin epithelium and pancreas.
- Found in bone marrow and umbilical cord
- Embryonic stem cell-like characteristics:
  - capacity to form in vitro spheres resembling embryoid bodies which express the placental form of alkaline phosphatase
  - genes involved in early gastrulation.

VSEL

- Pluripotency has been confirmed by demonstrating the ability of VSELs to differentiate into all three germ-layer lineages, ectoderm, endoderm and mesoderm.
- VSELs have been shown to assist in cardiac repair after myocardial infarction.

Kucia M et al. Adult marrow-derived very small embryonic-like stem cells and tissue engineering. *Expert Opin Biol Ther*. 2007 Oct

Diseases Treatable with Stem Cells — Today

- Leukemias
- Lymphoma
- Multiple Myeloma
- Coronary Heart Disease
- Radiation Sickness
- Multiple Sclerosis
- Lupus
- Other Autoimmune Diseases
- Tissue Repair & Burns
- Orthopedics

CHF and Stem Cells

- Stem cell infusion during CABG
- EF 46% vs. 37% control.
- P<.001

Patel AN et al. Surgical treatment for congestive heart failure with autologous adult stem cell transplantation: a prospective randomized study. *J Thorac Cardiovasc Surg*. 2005 Dec;130(6):1631-8

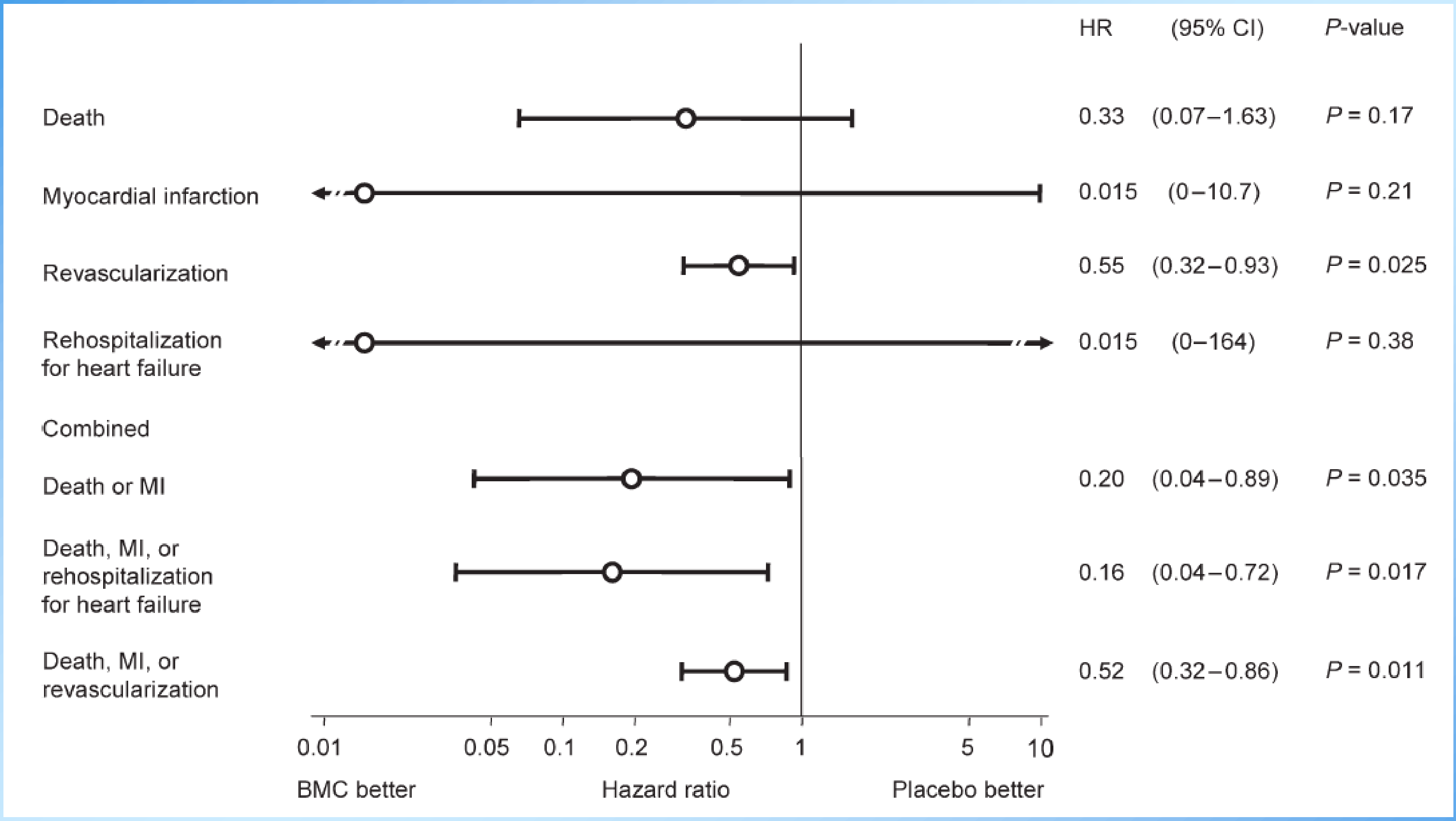
Intracoronary autologous stem cells - AMI

Intracoronary administration of autologous progenitor cells in patients with acute myocardial infarction

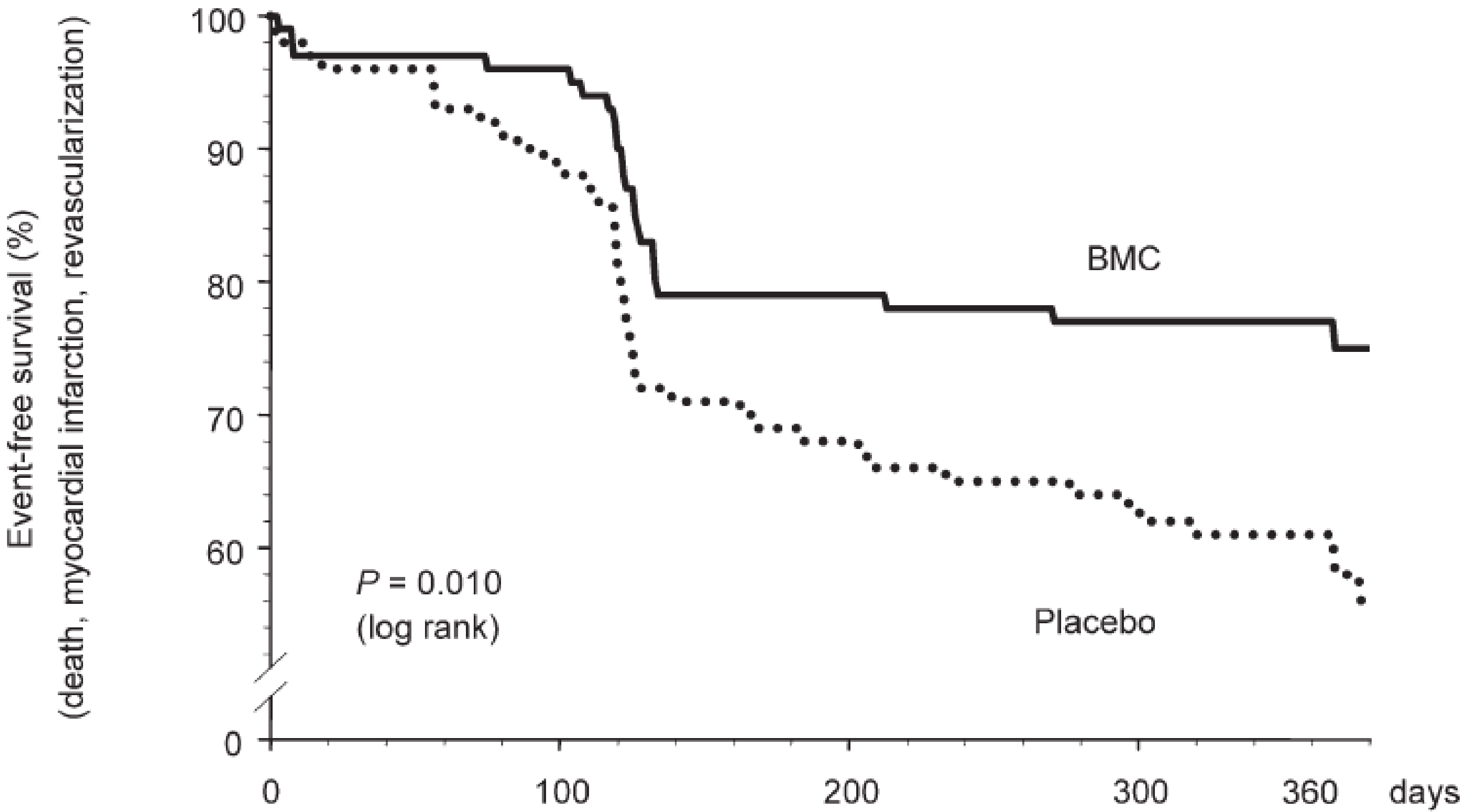
Double-blind, placebo-controlled, 204 pts

Successfully reperfused AMI, intracoronary infusion of bone-marrow-derived progenitor cells (BMCs) or placebo into the infarct artery 3-7 days after successful infarct reperfusion

Schachinner V et al. Improved clinical outcome after intracoronary administration of bone-marrow-derived progenitor cells in acute myocardial infarction: final 1-year results of the REPAIR-AMI trial. *Eur Heart J*. 2006 Dec;27(23):2775-83.



HR
(95% CI)
P-value
Death
0.33
(0.07–1.63)
P = 0.17
Myocardial infarction
0.015
(0–10.7)
P = 0.21
Revascularization
0.55
(0.32–0.93)
P = 0.025
Rehospitalization for heart failure
0.015
(0–164)
P = 0.38
Combined
Death or MI
0.20
(0.04–0.89)
P = 0.035
Death, MI, or rehospitalization for heart failure
0.16
(0.04–0.72)
P = 0.017
Death, MI, or revascularization
0.52
(0.32–0.86)
P = 0.011
0.01
0.05
0.1
0.2
0.5
1
5
10
BMC better
Hazard ratio
Placebo better



Event-free survival (%)
(death, myocardial infarction, revascularization)
100
90
80
70
60
0
0
100
200
300
360
days
BMC
Placebo
P = 0.010
(log rank)

Intracoronary autologous stem cells Acute Myocardial Infarct

- 12 months, - death, myocardial infarction, or necessity for revascularization was significantly reduced in the BMC group compared with placebo (P=0.009)
- 12 months - death, recurrence of myocardial infarction and rehospitalization for heart failure was significantly reduced (P = 0.006)
- Intracoronary administration of BMCs is associated with a significant reduction of the occurrence of major adverse cardiovascular events after AMI.

- Kim U et al. Homing of adipose-derived stem cells to radiofrequency catheter ablated canine atrium and differentiation into cardiomyocyte-like cells *Int J Cardiol*. 2009 Aug 14.
- Lee N et al. Repair of ischemic heart disease with novel bone marrow-derived multipotent stem cells *Cell Cycle*. 2005 Jul;4(7):861-4
- Lee RH et al. Intravenous hMSCs improve myocardial infarction in mice because cells embolized in lung are activated to secrete the anti-inflammatory protein TSG-6. *Cell Stem*. 2009 Jul 2;5(1):54-63.

- Goel RK et al. Effect of bone marrow-derived mononuclear cells on nerve regeneration in the transection model of the rat sciatic nerve. *J Clin Neurosci*. 2009 Sep;16(9):1211-7.
- Aslam M et al. Bone Marrow Stromal Cells Attenuate Lung Injury in a Murine Model of Neonatal Chronic Lung Disease. *Am J Respir Crit Care Med*. 2009 Aug 27.
- Fischer UM et al. Pulmonary passage is a major obstacle for intravenous stem cell delivery: the pulmonary first-pass effect. *Stem Cells Dev*. 2009 Jun;18(5):683-92.

- Negative study
- First pass effect

- Harting MT et al. Intravenous mesenchymal stem cell therapy for traumatic brain injury. *J Neurosurg.* 2009 Jun;110(6):1189-97.

Auto BMT for Treatment of Severe Autoimmune Disease

- Fassas (*J. Neurol*. 2002) Rx 188 pts with **MS** including 99 pts 2nd progressive, 19 primary progressive, 41 relapsing forms – 34 % sustained improvement
- Snowden (*J. Rheumatol*. 2004) All 78 **RA** pts showed significant improvement, 10% remained drug free, 73% easily controlled by drugs
- Burt (*JAMA* 2006) Rx 48 pts with severe **SLE**, 5 year survival 84%, 50% disease-free 5 yr survival & has treated >140 patients in the last 7 years
- Constantin G et al. Adipose-Derived Mesenchymal Stem Cells Ameliorate Chronic Experimental Autoimmune Encephalomyelitis. *Stem Cells*. 2009 Aug 12.

"Cure" of Type 1 DM

- 15 patients
- 7- to 36-month follow-up, 14 patients became insulin free

Voltarelli JC et al. Autologous nonmyeloablative hematopoietic stem cell transplantation in newly diagnosed type 1 diabetes mellitus. *JAMA*. 2007 Apr 11;297(14):1568-76.

“Cure” Type 1 DM continued

- 20 patients Type 1 DM
- Autologous nonmyeloablative hematopoetic stem cell transplantation
- 30 months later C-peptide increased significantly in all
- 12 remained insulin free 30 months

Couri CE et al. C-peptide levels and insulin independence following autologous nonmyeloablative hematopoietic stem cell transplantation in newly diagnosed type 1 diabetes mellitus. *JAMA*. 2009 Apr 15;301(15):1573-9

Stem cell therapy for type 1 diabetes

- Mesenchymal stem cells (MSCs) are multipotent non-hematopoietic progenitor cells with low immunogenic potential
- Generation of insulin-producing cells (IPCs) derived from MSCs
- Immunomodulatory effects

- Vija L et al. Mesenchymal stem cells: Stem cell therapy perspectives for type 1 diabetes. *Diabetes Metab*. 2009 Apr;35(2):85-93.

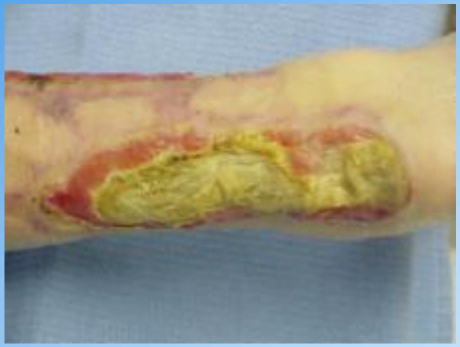
Tx of Chronic Wounds With Bone-Marrow Derived Cells

- Autologous bone marrow derived cells were used in the treatment of 3 patients with non-healing chronic wounds.
- Complete closure and evidence of dermal rebuilding was observed in all patients.
- Clinical and histologic evidence of reduced scarring was also observed.

Badiavas & Falanga, Arch.Dermatol. 139:510, 2003

- Kawamura A et al. Prevention of limb amputation in patients with limbs ulcers by autologous peripheral blood mononuclear cell implantation. *Ther Apher Dial*. 2005 Feb;9(1):59-63.

Treatment of Chronic Wounds With Bone-Marrow Derived Cells



Pre Treatment

Stem Cell Prep

Debridement

1 Month Post TX

2 Month Post TX

6 Month Post TX

Evangelos V. Badiavas and Vincent Falanga
Arch. Dermatol. 139:510, 2003

Percutaneous Autologous Bone Marrow Grafting for Nonunion Fractures

Hernigou P et al. *J Bone & Joint Surg* 87A: 1430, 2005

1st stem cell transplant

- Don’t clamp and cut the cord too fast
- Umbilical cord blood contains various valuable stem cells such as hematopoietic stem cells, endothelial cell precursors, mesenchymal progenitors, and multipotent/pluripotent lineage stem cells

- Tolosa JN et al. Mankind's first natural stem cell transplant. *J Cell Mol Med*. 2010 Feb 5

- Meagher RC. Human umbilical cord blood cells: how useful are they for the clinician? *J Hematother Stem Cell Res*. 2002 Jun;11(3):445-8
- El-Badri, NS. Cord Blood Stem Cells: A Fountain of Youth for the Brain. *Stem Cells and Development*. June 2006: 293-294
- Schlechta, B et al. Ex-Vivo Expanded Umbilical Cord Blood Stem Cells Retain Capacity for Myocardial Regeneration. *Circ J* 2010; 74: 188-194.

Umbilical cells & revascularization

- UC-MSCs induced to differentiate into endothelial-like cells
- UC-MSCs expanded to functional EPC’s in 9 days
- Ischemia improved ($p<0.05$).
- UC-MSCs are well suited for revascularization therapies.
- XU, Y et al. Umbilical Cord Derived Mesenchymal Stem Cells Isolated by a Novel Explantation Technique Can Differentiate Into Functional Endothelial Cells and Promote Revascularization. *Stem Cells Dev*. 2010 Feb 19

UCB-SC's and Spinal Cord Injury

- Rats treated with UCB at 5 days were significantly improved
- Reverse the behavioral effects of spinal cord injury
- Migrate to and participate in the healing of neurological defects caused by traumatic assault
- Samuel Saporta et al. Human Umbilical Cord Blood Stem Cells Infusion in Spinal Cord Injury: Engraftment and Beneficial Influence on Behavior. *Journal of Hematotherapy & Stem Cell Research*. June 2003, 12(3): 271-278.

HUCBC Transplantation for Stroke

- Treatment at 24 hours after MCAO with HUCBC significantly improved functional recovery (P<0.05).
- In vitro, significant HUCBC migration activity was present at 24 hours after MCAO (P<0.01)
- Intravenously administered HUCBC enter brain, survive, migrate, and improve functional recovery after stroke

- Chen J et al. Intravenous administration of human umbilical cord blood reduces behavioral deficits after stroke in rats *Stroke*. 2001 Nov;32(11):2682-8.

Endothelial Progenitor cells (EPCs)

- Stem cells that reside in the adult bone marrow or circulate in the blood
- Differentiate and mature into endothelial cells.
- Identified by stem-cell markers (CD34, CD133)
- EPCs decrease with age and are a measurement of vascular senescence and a biomarker of aging

- Chen JF, et al. Relationship between aging and the number and function of bone marrow-derived endothelial progenitor cells in rats. *Zhonghua Xin Xue Guan Bing Za Zhi*. 2006 Nov;34(11):1026-8.
- Thum T, et al. Age-dependent impairment of endothelial progenitor cells is corrected by growth hormone mediated increase of insulin-like growth factor-1. *Circ Res*. 2007 Feb 16;100(3):434-43.

- Hoetzer GL, et al. Aging, Exercise and Endothelial Progenitor Cell Clonogenic and Migratory Capacity in Men. *J Appl Physiol*. 2006 Dec 7

- Imanishi T et al. Estrogen reduces endothelial progenitor cell senescence through augmentation of telomerase activity. *J Hypertens*. 2005 Sep;23(9):1699-706.

Exercise EPC’s

- Brehm M et al. Effects of exercise training on mobilization and functional activity of blood-derived progenitor cells in patients with acute myocardial infarction. . *Eur J Med Res*. 2009 Sep 1;14(9):393-405.

Stem cell mobilization – response to tissue injury

- AMI
- Stroke
- Long bone fractures
- Stem cells are mobilized to the site of damage

CD 34+ Mobilized with STEMI

- Brehm M et al. Enhanced mobilization of CD34(+) progenitor cells expressing cell adhesion molecules in patients with STEMI. *Clin Res Cardiol*. 2009 Aug;98(8):477-86.

Stroke recovery associated with CD 34+ mobilization

- Dunac A. et al. Neurological and functional recovery in human stroke are associated with peripheral blood CD34+ cell mobilization**.** *J Neurol*. 2007 Mar;254(3):327-32.

EPC promote fracture healing and fractures induce EPC mobilization

- Matsumoto T et al. Fracture induced mobilization and incorporation of bone marrow-derived endothelial progenitor cells for bone healing. *J Cell Physiol*. 2008 Apr;215(1):234-42

EPC's and Endothelial function

- Impact vascular health by modulating vascular repair and function.
- Repair endothelial injury and protect against atherothrombosis
- EPC number predict vascular events better than conventional risk factors
- Higher numbers associated with decreased risk of CV death

- Werner N et al. Endothelial progenitor cells correlate with endothelial function in patients with coronary artery disease. *Basic Res Cardiol*. 2007 Nov;102(6):565-71.
- Shantsila E et al. Endothelial progenitor cells in cardiovascular disorders. *J Am Coll Cardiol*. 2007 Feb 20;49(7):741-52.

EPCs and CV Outcomes

- Higher EPC's – 70 % less death from CV causes

Werner N et al. Circulating Endothelial Progenitor Cells and Cardiovascular Outcomes. *N Engl J Med* 353:999, September 8, 2005

GH/IGF-1 increases EPCs

- EPC number and function is restored with a GH-mediated increase in serum IGF-1.
- Modulation of the GH can prevent age-associated changes in the cardiovascular system

Devin JK et al. The effects of growth hormone and insulin-like growth factor-1 on the aging cardiovascular system and its progenitor cells. *Curr Opin Investig Drugs*. 2008 Sep;9(9):983-92.

Thum T et al. Age-dependent impairment of endothelial progenitor cells is corrected by growth-hormone-mediated increase of insulin-like growth-factor-1. *Circ Res*. 2007 Feb 16;100(3):434-43.

- Chakravarthy MV et al. Insulin-like growth factor-I extends in vitro replicative life span of skeletal muscle satellite cells by enhancing G1/S cell cycle progression via the activation of phosphatidylinositol 3'-kinase/Akt signaling pathway. *J Biol Chem*. 2000 Nov 17;275(46):35942-52.

Testosterone and satellite cells (stem cells)

- Dose-dependent increase in muscle fiber CSA and satellite cell number.
- Testosterone-induced skeletal muscle hypertrophy in older men is associated with increased satellite cell replication and activation.

- Sinha-Hikim I et al. Effects of testosterone supplementation on skeletal muscle fiber hypertrophy and satellite cells in community-dwelling older men. *J Clin Endocrinol Metab*. 2006 Aug;91(8):3024-33.

Testosterone Replacement Therapy Increases EPC's

- Hypogonadism – low EPC
- T gel 50 mg/day x 6 months
  - Normalized EPC's
  - Androgen receptor expressed on EPC's
- May be mechanism of T benefit in CV disease

- Foresta C. et al. Reduced Number of Circulating Endothelial Progenitor Cells in Hypogonadal Men. *Journal of Clinical Endocrinology & Metabolism* 91(11):4599–4602, Nov 2006

- Liu KQ et al. Treatment of acute myocardial infarction with autologous bone marrow stem cells mobilization combined with recombinant growth factor (GH) in rat. *Zhongguo Wei Zhong Bing Ji Jiu Yi Xue*. 2006 Aug;18(8):494-7.

- Iwakura A et al. . Estradiol enhances recovery after myocardial infarction by augmenting incorporation of bone marrow-derived endothelial progenitor cells into sites of ischemia-induced neovascularization via endothelial nitric oxide synthase-mediated activation of matrix metalloproteinase-9. *Circulation.* 2006 Mar 28;113(12):1605-14.

EPC and ED

- Baumhäkel M et al. Circulating endothelial progenitor cells correlate with erectile function in patients with coronary heart disease. *European Heart Journal* 2006 27(18):2184-2188.

T and ED and EPC

- T improves ED and can resolve ED with PDE5 inhibitors when PDE5 inhibitors don’t work.
- T increases circulating endothelial progenitor cells from bone marrow which causes vascular repair.

- Caretta N et al. Erectile dysfunction in aging men: testosterone role in therapeutic protocols. *J Endocrinol Invest*. 2005;28 (11 Suppl Proceedings):108-11

Estrogen, telomerase, EPCs

- Imanishi T et al. Estrogen reduces endothelial progenitor cell senescence through augmentation of telomerase activity. *J Hypertens*. 2005 Sep;23(9):1699-706

Melatonin increase EPC's

- Melatonin has immunomodulatory effects
- Melatonin stimulates production of EPC's, natural killer cells and CD4 cells and inhibits CD 8 cells

Cardinale, D et al. Melatonin and the Immune System in aging. *NeuroImmuno Modulation* 2008;15:272-278.

Sub-Clinical (Mild) Hypothyroidism and EPC's

- EPC level with SCH low and improved with T4 treatment
- Shakoor SKA et al. Endothelial Progenitor Cells in Subclinical Hypothyroidism: The Effect of Thyroid Hormone Replacement Therapy. *J Clin Endocrinol Metab*, January 2010, 95(1):319–322

CRP and EPCs

- CRP
  - biomarker and participant in CV disease
  - Promotes endothelial dysfunction
  - Decreases endothelial NO synthase (eNOS)
  - Promotes LOX-1
  - Inhibits bone marrow derived EPC survival and differentiation

- Fujii H et al. C-Reactive Protein alters anti-oxidant defenses and promotes apoptosis in Endothelial Progenitor Cells. *Arterioslcer Thromb Vasc Biol* 2006;26:2476-2482

CRP

- Induces ROS formation in EPC's
- Alters expression of anti-oxidative enzymes ie glutathione
- Leads to decreased telomerase activity and EPC death
- Impairs endothelial repair and contributes to development and progression of atherosclerosis

Homocysteine and EPCs

- Homocysteine reduces EPC’s
- B6 and Folate reverses this effect

- Alam MM et al. Homocysteine reduces endothelial progenitor cells in stroke patients through apoptosis. *J Cereb Blood Flow Metab*. 2008 Sep 3.

How else can you grow your own stem cells beside hormone optimization?

- Many of the anti-aging measures that are the foundation of our specialty

Folate and Neuronal Stem Cells

- Neonatal rats
- Neuronal stem cells respond to folate with increased signaling and proliferation

- Zhang X et al. Effects of folate on notch signaling and cell proliferation in neural stem cells of neonatal rats in vitro *J Nutr Sci Vitaminol* (Tokyo). 2008 Oct;54(5):353-6.

Folic Acid and EPC genes

- van Oostrom O. et al Folic acid supplementation normalizes the endothelial progenitor cell transcriptome of patients with type 1 diabetes: a case-control pilot study. *Cardiovasc Diabetol*. 2009 Aug 25;8:47.

Thiamine, EPC, endothelium

- Diabetics have lower EPC count
- Increased thiamine intake increased EPC’s and FMD independent of other nutrients

Wong CY Daily intake of thiamine correlates with the circulating level of endothelial progenitor cells and the endothelial function in patients with type II diabetes. *Mol Nutr Food Res*. 2008 Dec;52(12):1421-7.

Antioxidants and EPCs

- Vitamin C and E prevent TNF-a induced apoptosis
- Exercise alone increased EPC's ($p<0.05$)
- Antioxidants plus moderated exercise amplified the increased the number of circulating EPC's ($p<0.001$)

Fiorito C et al. Antioxidants increase number of progenitor endothelial cells through multiple gene expression pathways. *Free Radic Res*. 2008 Aug;42(8):754-62.

Exercise and EPCs

- Sarto P et al. Effects of exercise training on endothelial progenitor cells in patients with chronic heart failure. *J Card Fail.* 2007 Nov;13(9):701-8.
- Van Craenenbroeck EM et al. A maximal exercise bout increases the number of circulating CD34+/KDR+ endothelial progenitor cells in healthy subjects. Relation with lipid profile. *J Appl Physiol*. 2008 Apr;104(4):1006-13

Red Wine

- Increase of EPC number ($P<0.05$) in mice after short-term supplementation with Red Wine (28 days) for sedentary group ? due to aortic SIRT 1 expression
- No statistical increase in EPC number in moderate exercise group

Balestrieri ML et al. Effect of red wine antioxidants and minor polyphenolic constituents on endothelial progenitor cells after physical training in mice. *Int J Cardiol.* 2008 May 23;126(2):295-7

Resveratrol and EPCs

- Red wine prevented TNF-a reduction of EPC number ($p<0.05$) as did pure resveratrol
- Red wine reduced p38 phosphorylation expression levels and increased NO levels

Xia L. et al. Resveratrol reduces endothelial progenitor cells senescence through augmentation of telomerase activity by Akt-dependent mechanisms. *Br J Pharmacol*. 2008 Oct;155(3):387-94.

MSC's, Sirt1, bone

- MSC's can differentiate to osteoblasts or adipocytes
- Sirt1 inhibits adipocyte development and promotes osteoblasts
- What turns on Sirt1?

- Backesjo CM. Activation of Sirt1 decreases adipocyte formation during osteoblast differentiation of mesenchymal stem cells. *Cells Tissues Organs*. 2009;189(1-4):93-7.

Resveratrol, D3, Stem cells, myeloma

- Resveratrol inhibits myeloma cells
- Mesenchymal stem cells → osteoblasts
- Resveratrol and D3 enhance above
- NFKB inhibited by Resveratrol inhibiting bone resorption and increasing bone formation via osteoblasts inhibiting plasma cell infiltration
- Resveratrol promotes expression of blast markers in stem cells and stimulates response to Vitamin D

- Boissy P et al. Resveratrol inhibits myeloma cell growth, prevents osteoclast formation, and promotes osteoblast differentiation. *Cancer Res.* 2005 Nov 1;65(21):9943-52.

D and K stimulate osteoblasts from stem

- Synergism D3 and K2
- Implies D3 and K2 needed for fracture healing from stem cells

Gigante A.Vitamin K and D association stimulates in vitro osteoblast differentiation of fracture site derived human mesenchymal stem cells. J *Biol Regul Homeost Agents*. 2008 Jan-Mar;22(1):35-44.

Resveratrol and EPC's

- Low concentration of resveratrol improved EPC function
- 10 mg/kg resveratrol in rats improved injured aorta endothelium and eNOS expression

JG, et al. Effects of resveratrol on endothelial progenitor cells and their contributions to reendothelialization in intima-injured rats. *J Cardiovasc Pharmacol*. 2006 May;47(5):711-21

L - arginine

- L-arginine potentiates the effects of moderate exercise on increasing number of EPC’s ($p<0.001$) and VEGF levels ($p<0.001$)

- Fiorito C et al. Effect of L-arginine on circulating endothelial progenitor cells and VEGF after moderate physical training in mice. *Int J Cardiol*. 2008 Jun 6;126(3):421-3.

Stem cells optimization through neutraceuticals?

- Blueberry
- Green tea
- Vitamin D3
- Carnosine

- Bickford PC et al. Neutraceuticals synergistically promote proliferation of human stem cells. *Stem Cells Dev*. 2006 Feb;15(1):118-23.

- Combinations of nutrients produce a synergistic effect to promote proliferation of human hematopoietic progenitors.

- Nutrients can act to promote healing via an interaction with stem cell populations.

Stem cells optimization through neutraceuticals?

- Approaches that stimulate endogenous stem cells to promote healing and regenerative medicine.
- A dose-related effect of blueberry, green tea, catechin, carnosine, and vitamin D(3) was observed on proliferation with human bone marrow as compared with human granulocyte-macrophage colony-stimulating factor (hGM-CSF).

Neutraceuticals augment circulating EPC’s

- Green tea
- Astragalus
- Gogi berries
- Lactobacillus fermentum
- Ellagic acid
- Beta 1,3 glucan
- Vitamin D3

- Mikirova N et al. Nutraceutical augmentation of circulating endothelial progenitor cells and hematopoietic stem cells in human subjects *J Transl Med*. 2010 Apr 8;8:34.

Fish Oil and Stem Cells

- Promotes differentiation of neural stem cells into neurons
- Increased number of neurons in the dentate gyrus
- Promotes neurogenesis and this suggests it can modulate hippocampal function.
- Kawakita E et al. Docosahexaenoic acid promotes neurogenesis in vitro and in vivo. *Neuroscience* 2006:139:991-997

Ginkgo and EPCs

- Increases EPC number and function
- Prevents onset of EPC senescence through telomerase activity

- Dong XX et al. Ginkgo biloba extract reduces endothelial progenitor-cell senescence through augmentation of telomerase activity. *J Cardiovasc Pharmacol*. 2007 Feb;49(2):111-5.

Neoangiogenesis and Marine Algae

- Fucoidans from brown algae are Antithrombotic and Proangiogenic
- Cytokine stimulation triggers EPC precursors and mobilize to affected area.
- Improves membrane signaling
- Promotes revascularization of ischemic tissues.
- Boisson-Vidal, C et al. Neoangiogenesis Induced by Progenitor Endothelial Cells: effect of Fucoidan from Marine Algae. *Cardiovascular and Hematological Agents in Medicinal Chemistry*. 2007, 5, 67-77.

EPCs = Biomarker Aging

Increase

- GH/IGF-1
- E2 → Telomerase
- Testosterone
- Antioxidants → (ROS)
- Exercise
- Red Wine, Resveratrol
- L-Arginine
- Blueberries, Green Tea
- Carnosine
- Fish Oil
- Fucoidan
- Gingko → Telomerase

Decrease

- Inflammation
- Inflammatory Cytokines
- CRP
- ROS

Anti-Aging docs are already "stem cells docs"

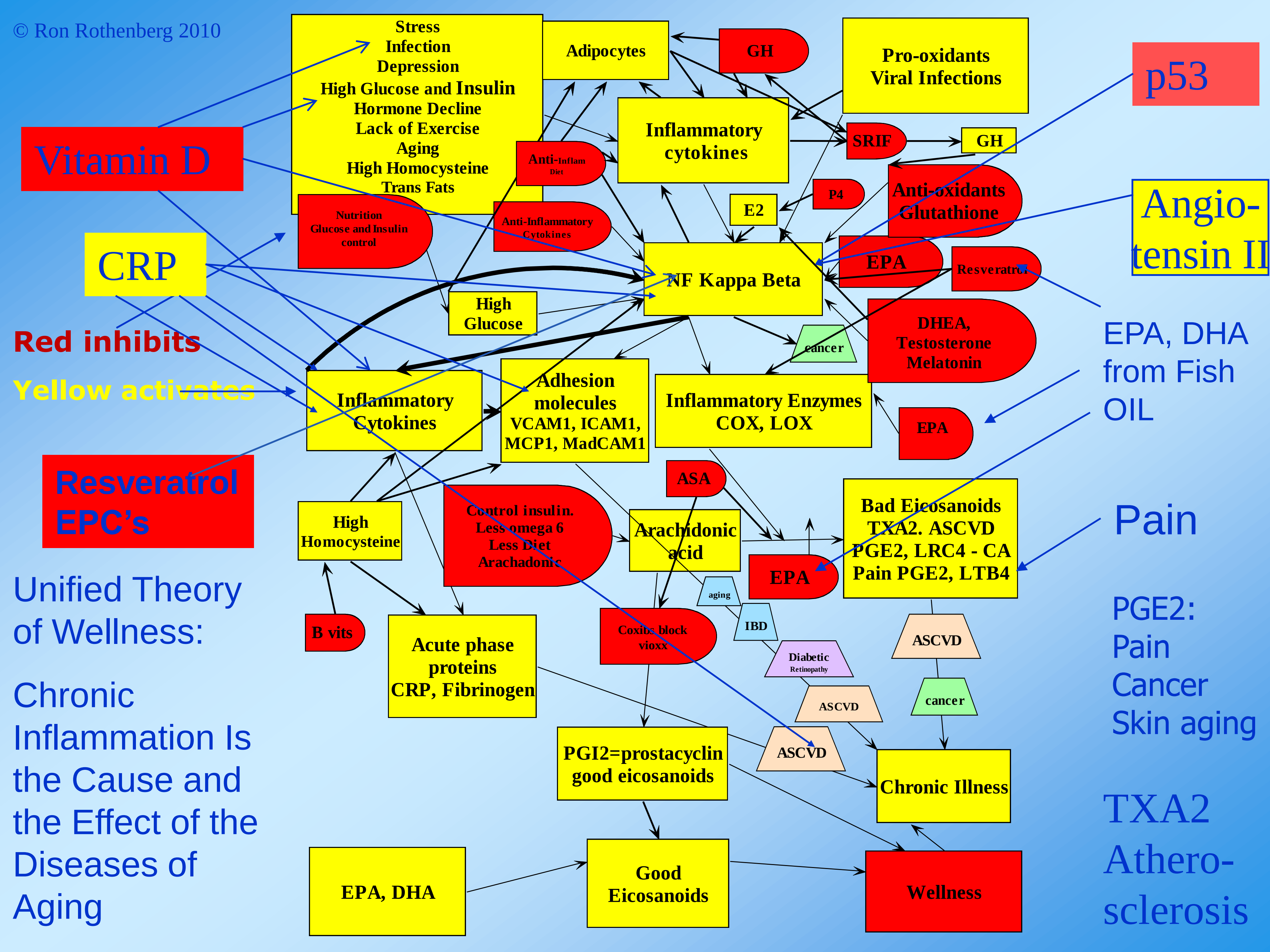
- Kawakita E et al. Docosahexaenoic acid promotes neurogenesis in vitro and in vivo. *Neuroscience* 2006:139:991-997

- Sinha-Hikim I et al. Effects of testosterone supplementation on skeletal muscle fiber hypertrophy and satellite cells in community-dwelling older men. *J Clin Endocrinol Metab*. 2006 Aug;91(8):3024-33

- Now you have the regenerative medicine technology to go to the next step in Anti-aging medicine
- You can answer that “What’s new” question,
- When should you bank your stem cells for “bio-insurance”?`
- Who should receive umbilical stem cells now?

Early adopter and Near Future Uses

- Immune system modulation
  - Auto immune
  - Cancer
- Brain Repair
- Damaged organs and tissues
  - Heart, Lung, Joints
- Rejuvenation tune ups?



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Stress
Infection
Depression
High Glucose and Insulin
Hormone Decline
Lack of Exercise
Aging
High Homocysteine
Trans Fats
Adipocytes
GH
Pro-oxidants
Viral Infections
p53
Vitamin D
Inflammatory cytokines
SRIF
GH
Anti-Inflam Diet
Anti-oxidants Glutathione
Angio-tensin II
Nutrition Glucose and Insulin control
Anti-Inflammatory Cytokines
E2
P4
CRP
NF Kappa Beta
EPA
Resveratrol
High Glucose
DHEA, Testosterone Melatonin
EPA, DHA from Fish OIL
Red inhibits
cancer
Yellow activates
Inflammatory Cytokines
Adhesion molecules VCAM1, ICAM1, MCP1, MadCAM1
Inflammatory Enzymes COX, LOX
EPA
Resveratrol EPC's
ASA
High Homocysteine
Control insulin. Less omega 6 Less Diet Arachadonic
Arachidonic acid
Bad Eicosanoids TXA2. ASCVD PGE2, LRC4 - CA Pain PGE2, LTB4
Pain
EPA
Unified Theory of Wellness:
aging
B vits
IBD
ASCVD
PGE2: Pain Cancer Skin aging
Acute phase proteins CRP, Fibrinogen
Coxibs block vioxx
Diabetic Retinopathy
ASCVD
cancer
Chronic Inflammation Is the Cause and the Effect of the Diseases of Aging
ASCVD
PGI2=prostacyclin good eicosanoids
Chronic Illness
TXA2 Athero-sclerosis
EPA, DHA
Good Eicosanoids
Wellness