NEED FOR TISSUES

- 500,000+ procedures promote bone growth
- 500,000+ procedures repair cartilage
- 1 million+ patients need skin tissue
- 200 million+ teeth restorations
- 1 in 700 births: orofacial clefts
- 10-15% Americans periodontal tissue destruction

(numbers in USA per year)

CURRENT THERAPIES

- **Synthetic prosthesis**
- **Drug therapies**
- **Organ/tissue transplantation**

RAPID DRUG DESTRUCTION

- Insulin < 25 min.
- Growth hormone < 25 min.
- Parathyroid hormone < 15 min.
- Many small proteins sec.-min.

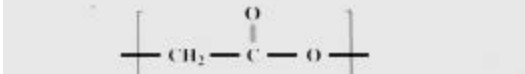
CONTROLLED RELEASE APPROACHES

- Polymers
- Pumps
- Gene therapy
- Immunoisolated cells

IMPROVED DRUG DELIVERY

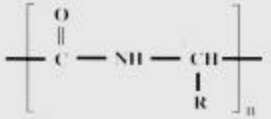
- Maintaining drug at desired level
- Minimize side effects
- Decrease amount of drug required
- Decrease doses, potentially less invasive
- Improve action of drugs which degrade quickly

Materials for 3-D Matrices

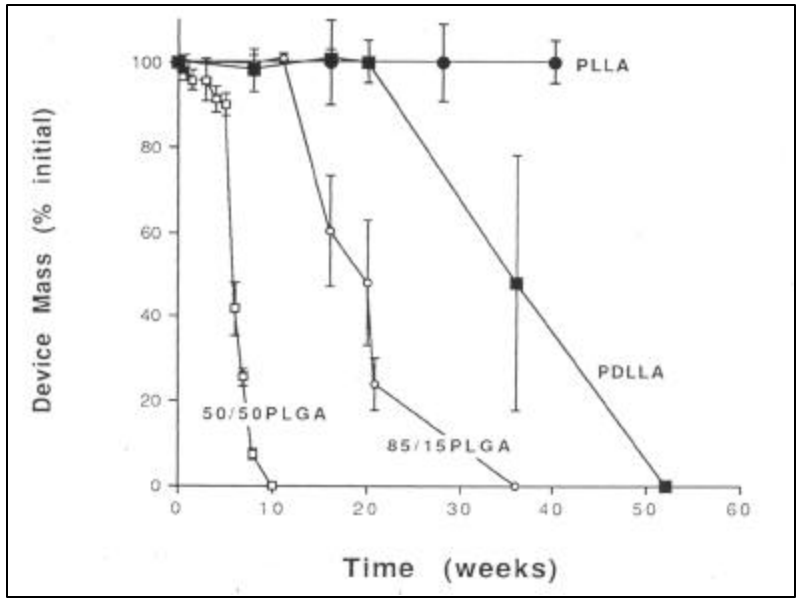


Poly(glycolic acid)

Poly(lactic-co-glycolic acid)



Collagen



Strategy for Inducing Blood Vessel Invasion

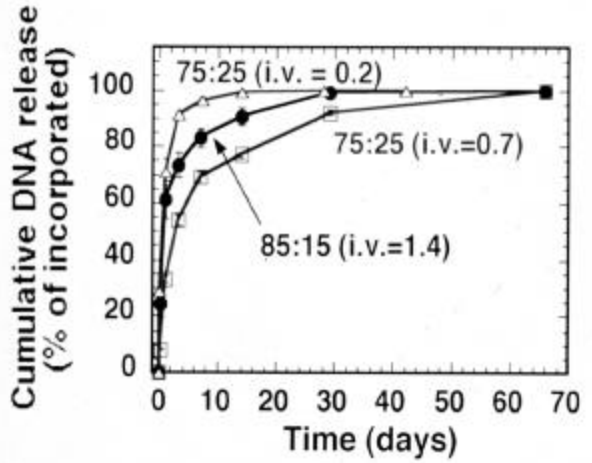
- Release pro-angiogenic growth factors (VEGF) from scaffold.
- Stimulate endothelial cells present in surrounding tissue to migrate into the matrix and differentiate into new vessels.

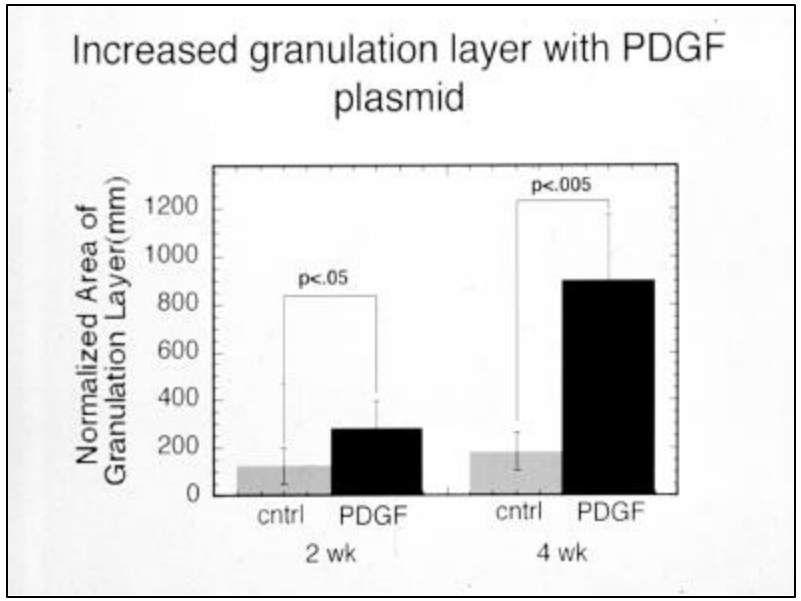
Mooney, D.J., Mikos, A.G., Scientific American, April 1999

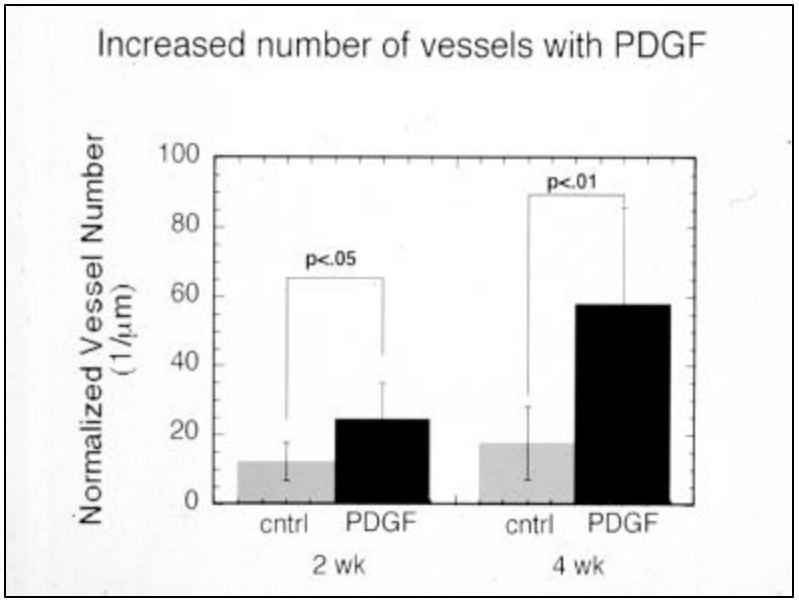
Method of Gene Delivery

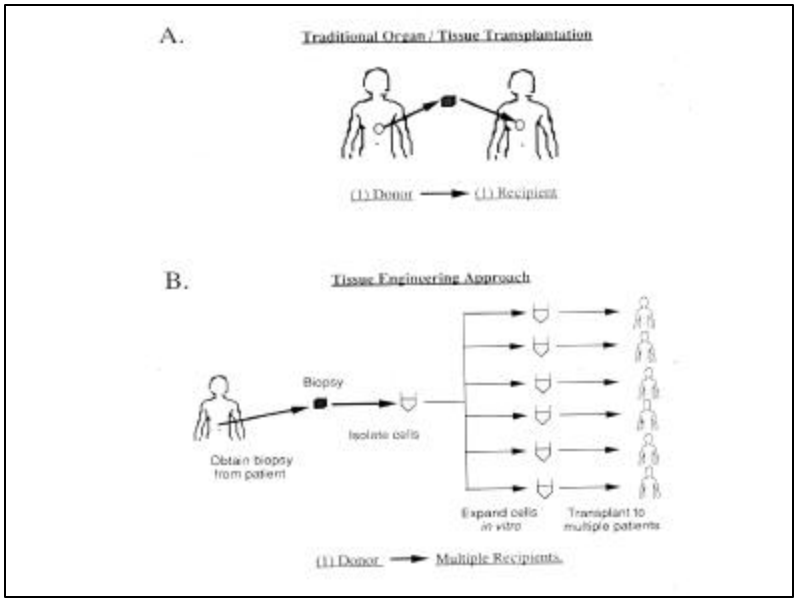
Viral	Non-Viral
• High expression levels	• Minimal safety concerns
• Potential long term expression	• Manufacturing cost
• Safety concerns	• Low expression levels

Sustained, controlled release of plasmid DNA



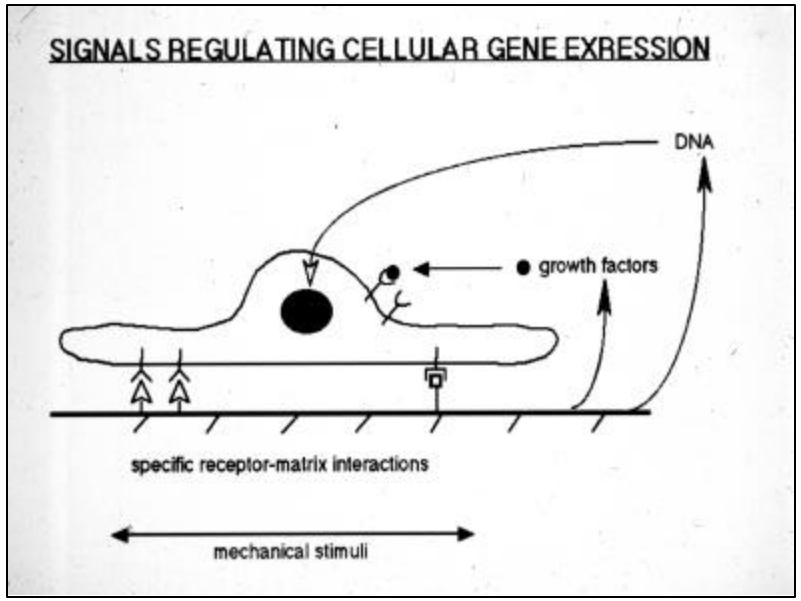

Increased granulation layer with PDGF plasmid
Normalized Area of Granulation Layer(mm)
1200
1000
800
600
400
200
0
p<.05
p<.005
cntrl
PDGF
cntrl
PDGF
2 wk
4 wk

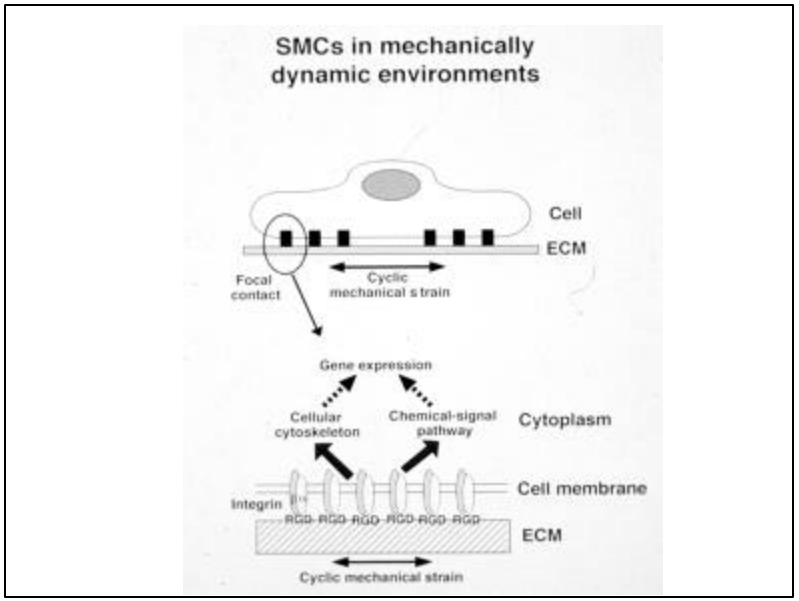

Increased number of vessels with PDGF
Normalized Vessel Number (1/μm)
100
80
60
40
20
0
p<.05
p<.01
cntrl
PDGF
cntrl
PDGF
2 wk
4 wk

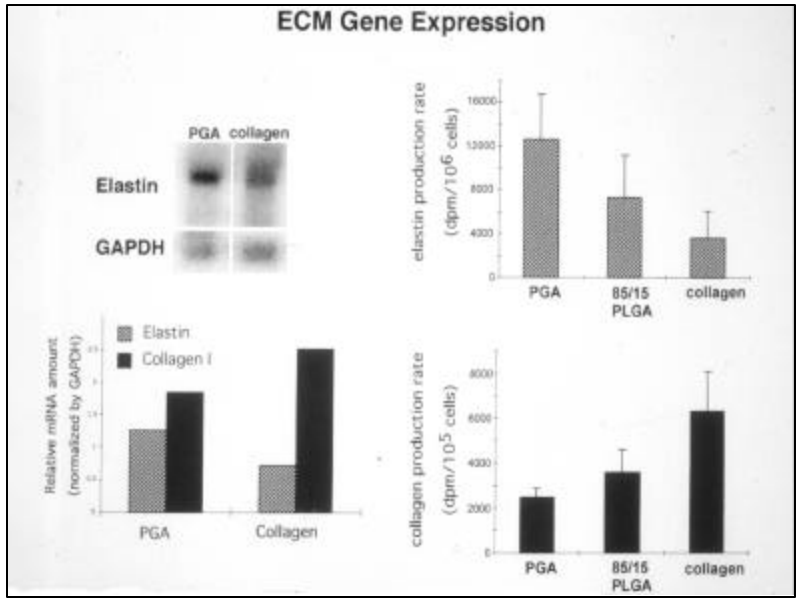


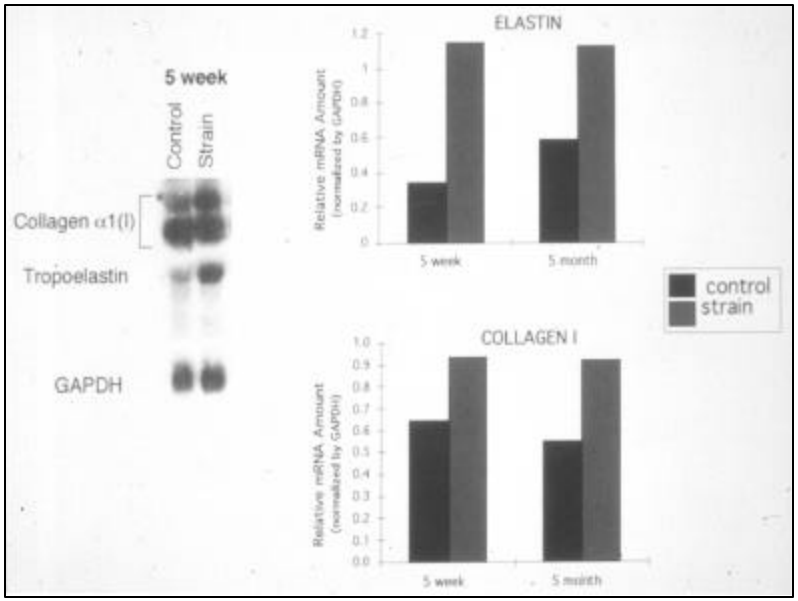
Roles of exogenous ECM in tissue engineering

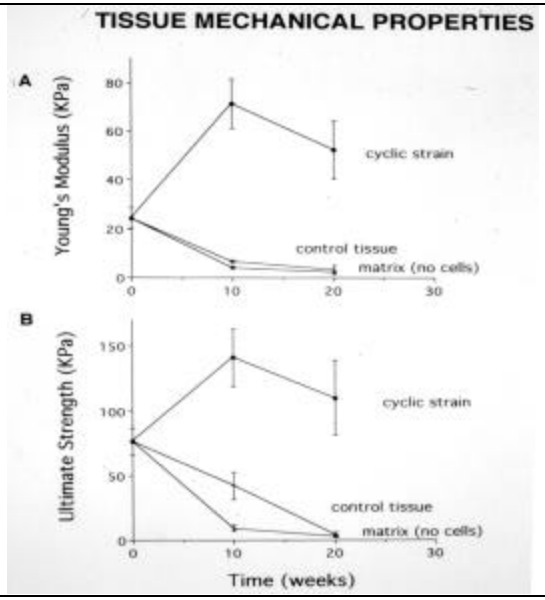
- Providing mechanical support and defining a potential space for tissue development
- Guiding new tissue regeneration with a pre-defined three-dimensional structure
- Delivering cells to desired sites in the body


SIGNALS REGULATING CELLULAR GENE EXRESSION
DNA
growth factors
specific receptor-matrix interactions
mechanical stimuli


SMCs in mechanically dynamic environments
Cell
ECM
Focal contact
Cyclic mechanical strain
Gene expression
Cellular cytoskeleton
Chemical-signal pathway
Cytoplasm
Cell membrane
Integrin
RGD RGD RGD RGD RGD RGD
ECM
Cyclic mechanical strain


ECM Gene Expression
PGA collagen
Elastin
GAPDH
elastin production rate
(dpm/10^6 cells)
PGA
85/15 PLGA
collagen
Relative mRNA amount
(normalized by GAPDH)
Elastin
Collagen I
PGA
Collagen
collagen production rate
(dpm/10^5 cells)
PGA
85/15 PLGA
collagen


5 week
Control
Strain
Collagen α1(I)
Tropoelastin
GAPDH
ELASTIN
Relative mRNA Amount
(normalized by GAPDH)
5 week
5 month
control
strain
COLLAGEN I
Relative mRNA Amount
(normalized by GAPDH)
5 week
5 month



CURRENT TISSUE ENGINEERING MATRICES

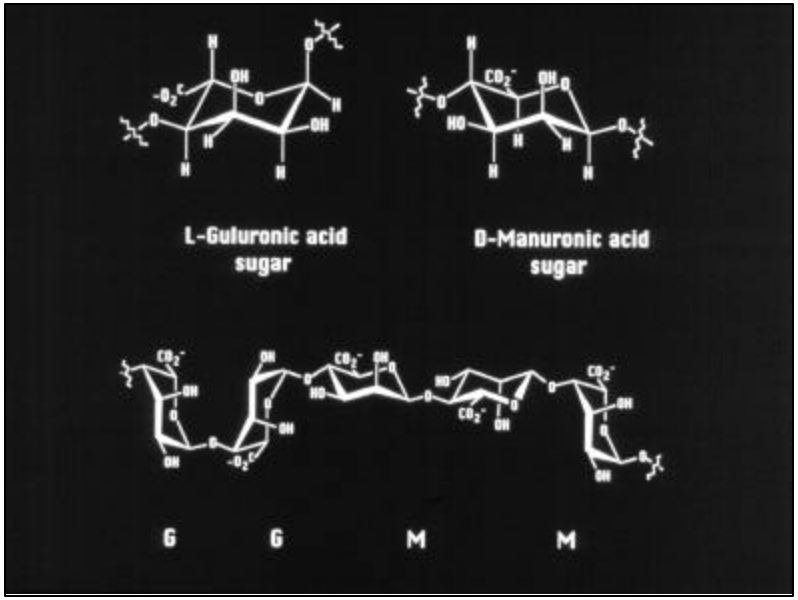
- **Can exploit default mechanism of cell adhesion**
- **CANNOT design mechanism of cell adhesion**

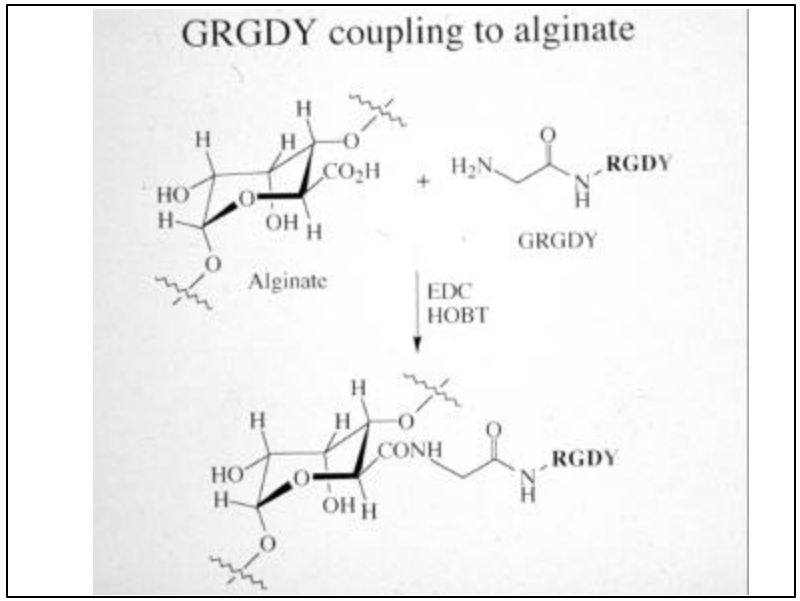
Hypothesis

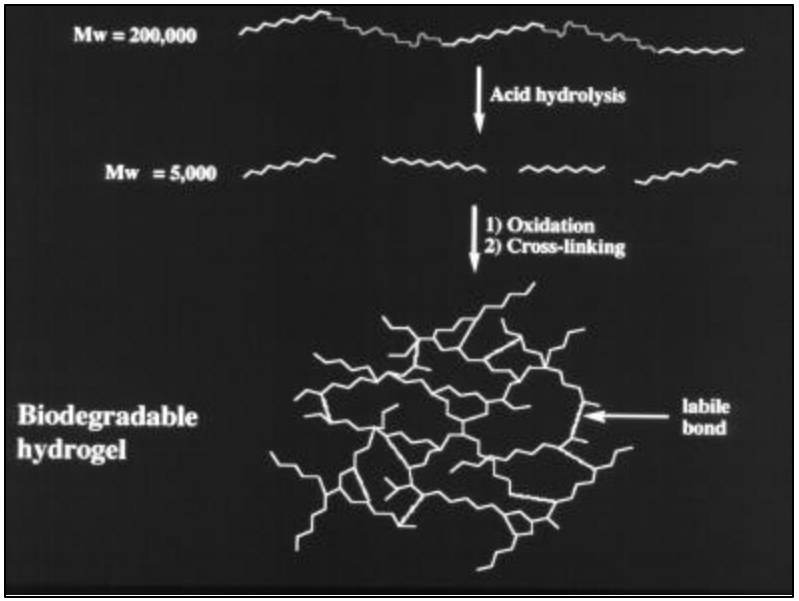
Cell gene expression within an engineered tissue can be controlled by:

1) the mechanism of cell adhesion to a matrix
   - regulated by adhesion ligand type
   - regulated by adhesion ligand density

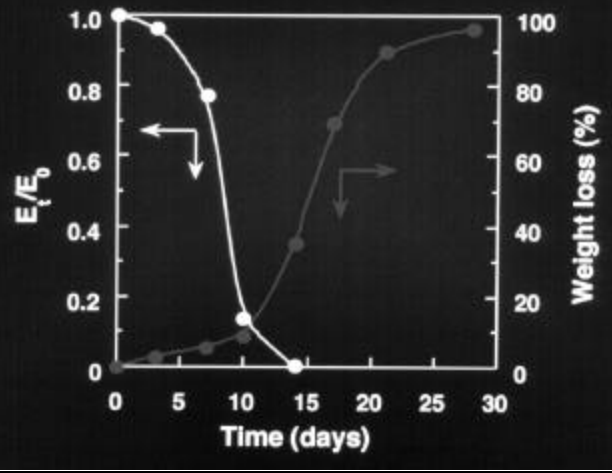
2) the subsequent interactions between the cells and the matrix
   - controlled by the mechanical properties of the matrix and its ability to resist cell-based tractional forces




GRGDY coupling to alginate
Alginate
GRGDY
RGDY
EDC
HOBT
CONH
RGDY


Mw = 200,000
Acid hydrolysis
Mw = 5,000
1) Oxidation
2) Cross-linking
Biodegradable
hydrogel
labile
bond

Hydrolytic degradation of hydrogels derived from alginate



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