Innovative Biomaterial for Orthopedic Tissue Disorder

(Plenary speech)

Biomaterials : **Technologies for Life and Society** (**Biomaterials-2019**),
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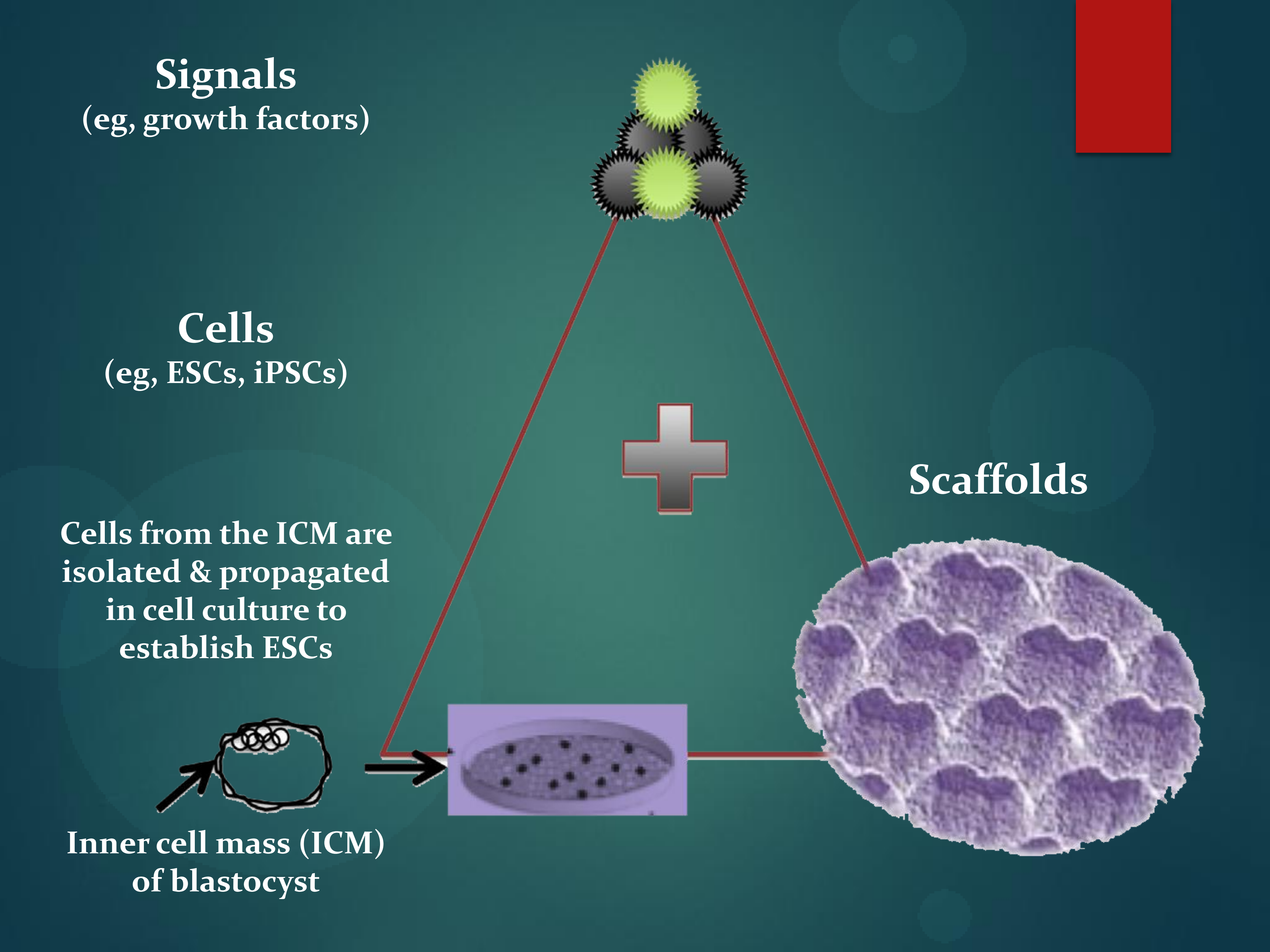
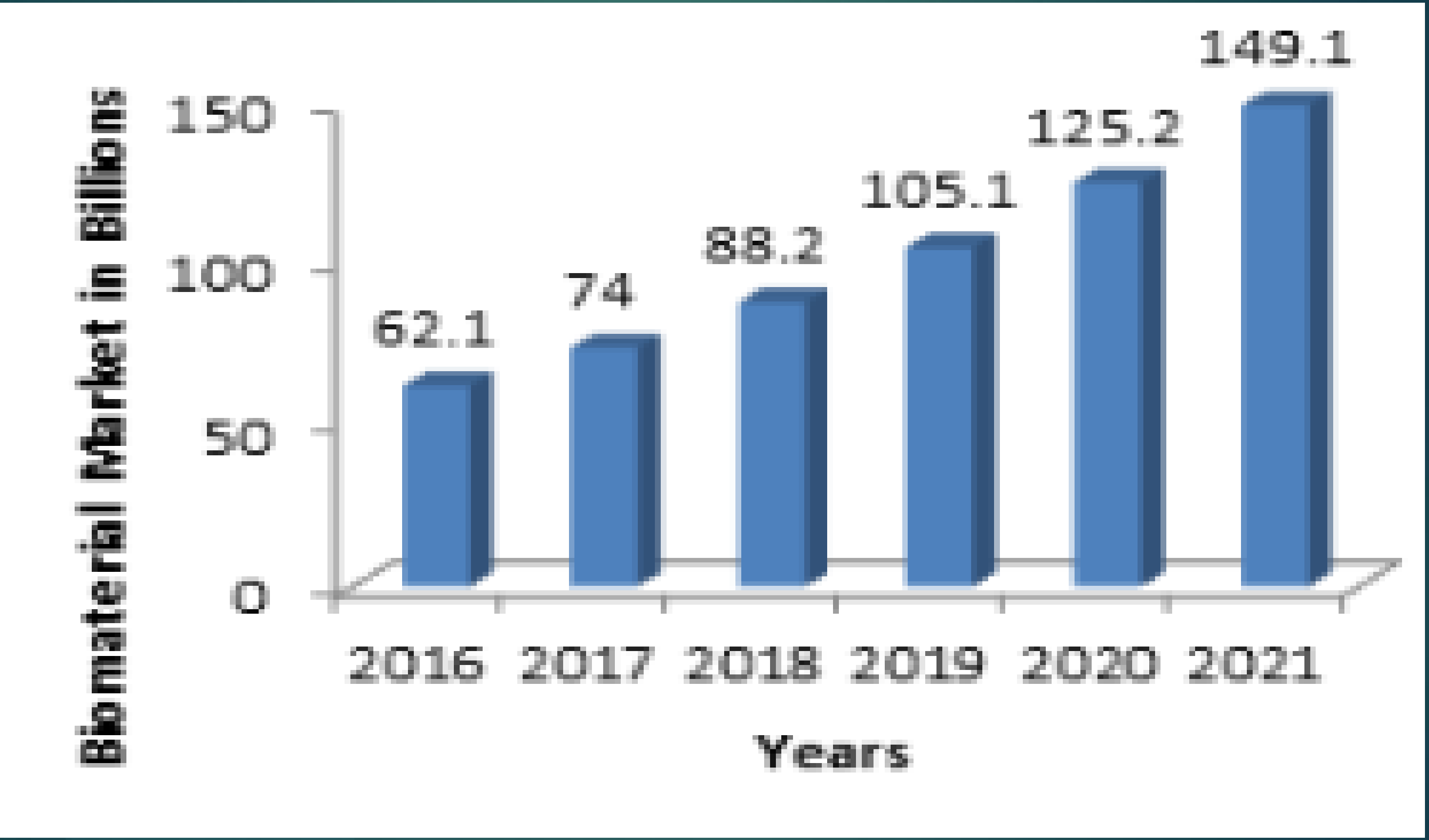
Department of Biotechnology & Medical Engineering


Signals
(eg, growth factors)
Cells
(eg, ESCs, iPSCs)
Scaffolds
Cells from the ICM are isolated & propagated in cell culture to establish ESCs
Inner cell mass (ICM) of blastocyst

Global Biomaterial Market



Biomaterials classification

I

- First generation
- Bioinert materials

II

- Second generation
- Bio active and biodegradable materials

III

- Third generation
- Materials designed to stimulate specific responses at molecular level

Biomaterials for Scaffolding

- **Natural biomaterials**
  - **Protein origin biomaterials**
    - Silk
    - Collagen
    - Fibrin
    - Gelatin
  - **Polysaccharides origin biomaterials**
    - Hyaluronan
    - Alginate
    - Agarose
    - Chitosan
- **Synthetic biomaterials**
  - **Polymer biomaterials**
    - Poly-ethylene glycol (PEG)
    - Polyglycolide (PGA)
    - Poly-lactic-co-glycolic acid (PLGA)
    - Poly-D, L-lactide (PDLLA)
    - Poly-e-caprolactone (PCL)
  - **Ceramic biomaterials**
    - Alumina
    - Zirconia
    - Sintered HA
    - α-or β tricalcium phosphate (α TCP, β -TCP)
    - Tetracalcium phosphate
    - Hydroxyapatite
    - Bioactive glass
    - Calcium phosphate

Orthopaedic TE Biomaterials

- Cartilage Tissue
- Bone Tissue
- Tendon Tissue
- Ligament Tissue
- Muscle Tissue

- Cartilage Tissue – Articular Cartilage
- Bone Tissue –Subchondral Bone
- Osteochodral

Advancement of Biomaterial Development

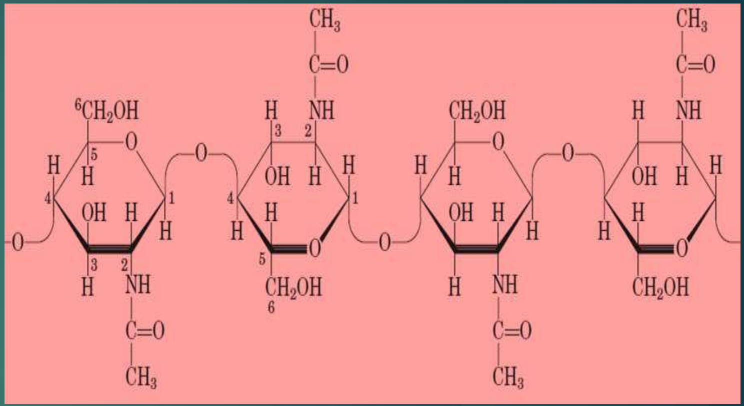
- Single Biopolymer
- Biopolymer Blend
- Bioplymer-Bioceramic composites (binary/ternary)
- Metal/metal oxide loaded biocomposites/nano composites

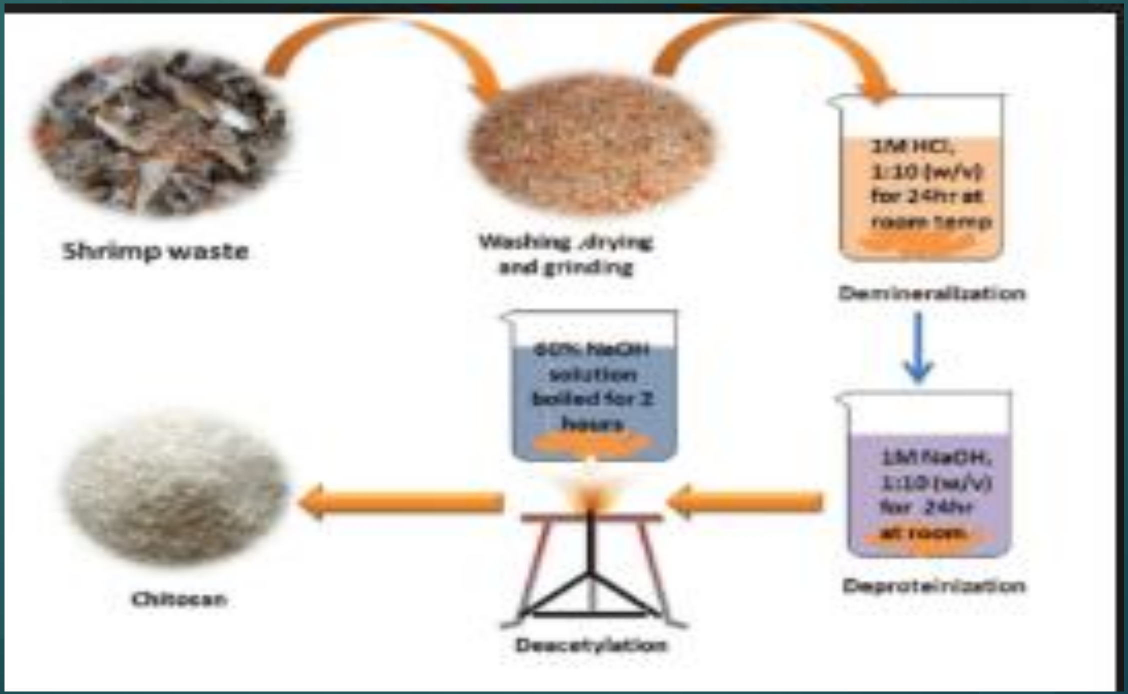
Ideal Natural Biopolymers

- Collagen-
- Chitosan-
- Silk Fibroin-
- CMC

Chitin

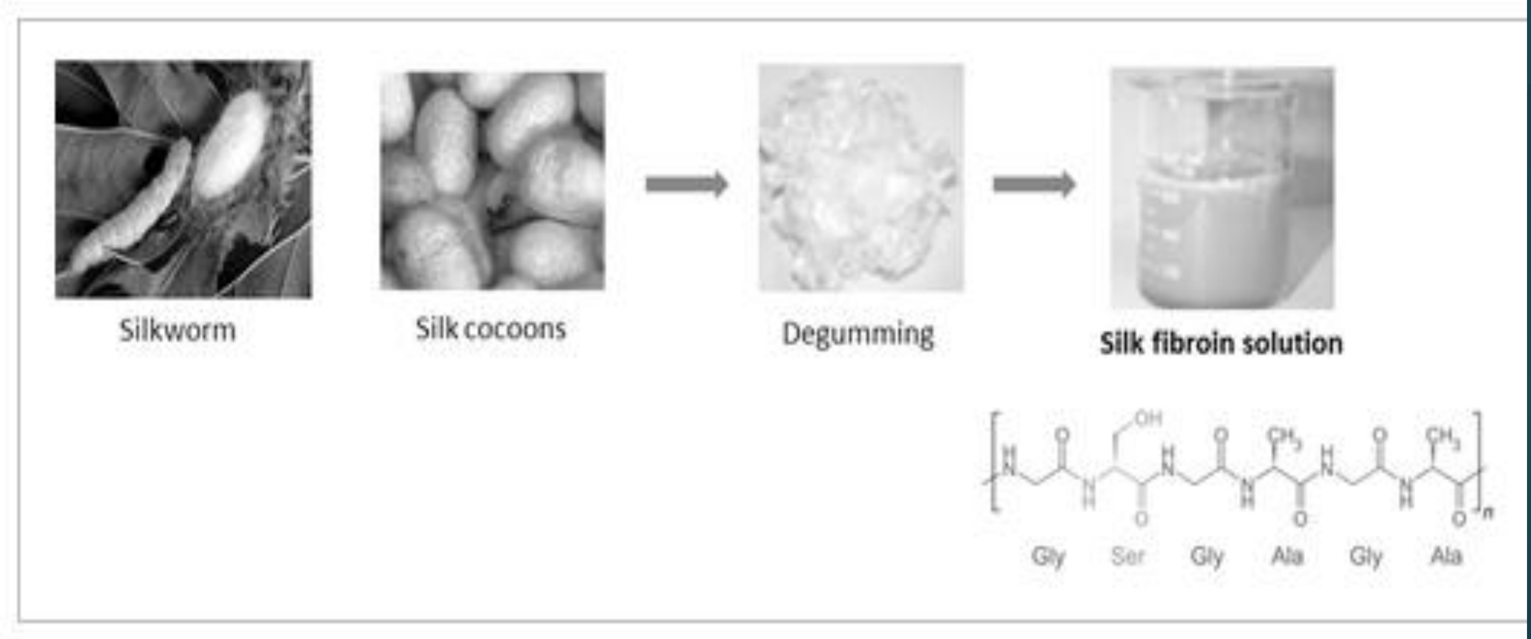
• Hydrophobic,
• Derived mostly from exoskeleton of Arthropods
• N-deacetylated derivative- CHITOSAN
• Wound healing, drug delivery, tissue engineering scaffolds





Silk

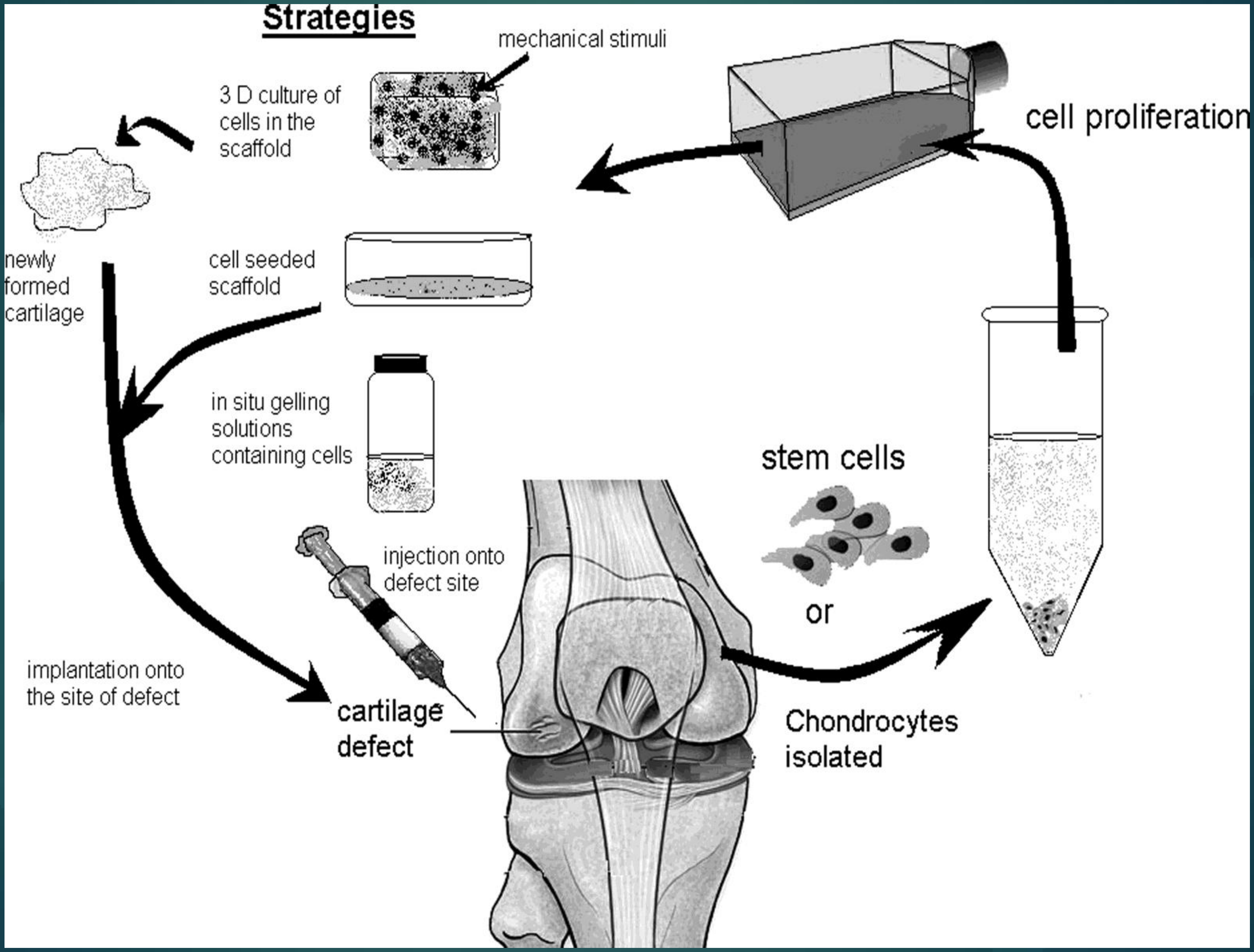
- Natural protein fibre, mainly composed of fibrinogen
- Produced by larvae of mulberry silkworm *Bombyx mori*
- Mostly in beta conformation
- Uses: Sutures, Scaffolds



Bio-ceramics

- Hydroxyapatite
- BTCP
- Bioactive Glass (Bioglass)

Biomaterial Development E

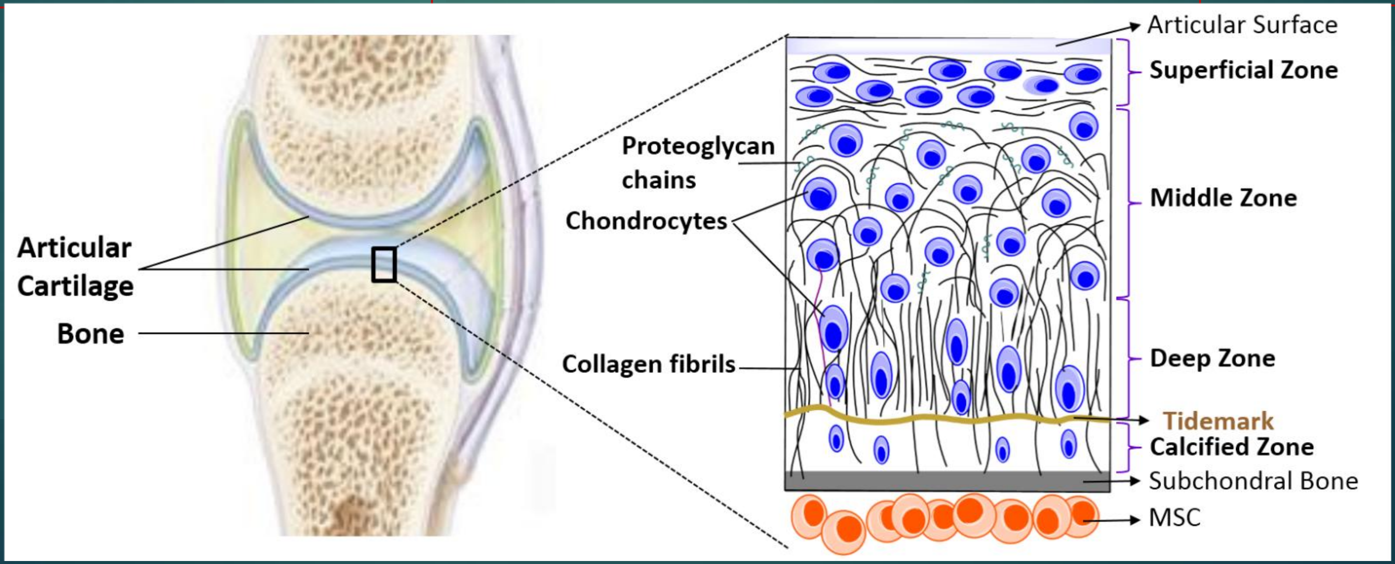


Cartilage Tissue

- Flexible connective tissue → Structural component
- Smooth surface → bones glide & move without friction, grinding or discomfort
- Avascular, aneural & alymphatic tissue
- At maturity has low metabolic activity → highly suited in its task but limits capacity of self-repair
- Composition → Chondrocytes and Matrix

Shock absorbing large polyionic aggregates, hydrophilic environment compressive strength

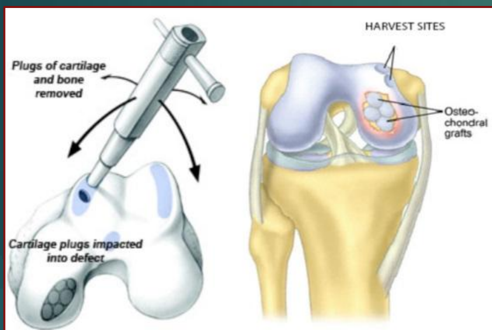
Mesh like framework tensile strength



Cartilage Defects Treatment

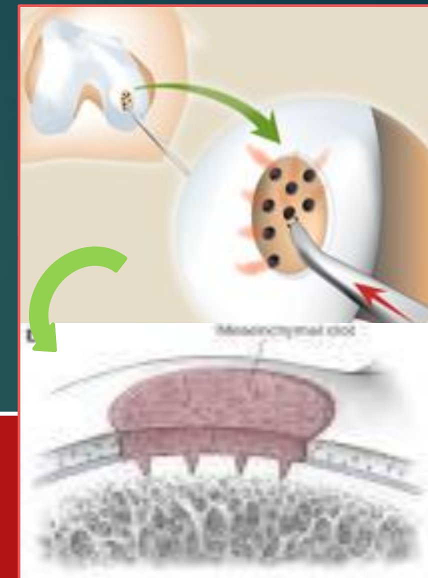
Symptom Management

Cartilage Restoration



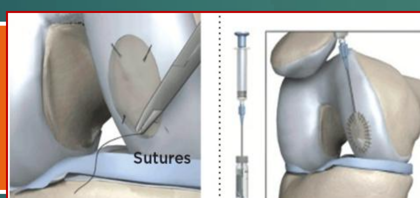
Osteochondral Grafting

Cartilage Regeneration

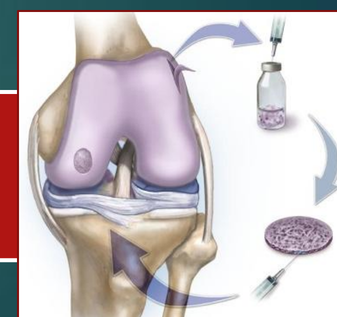


Marrow Stimulation

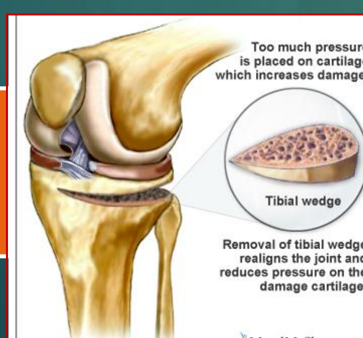
Resection



Autologous Chondrocyte Implantation

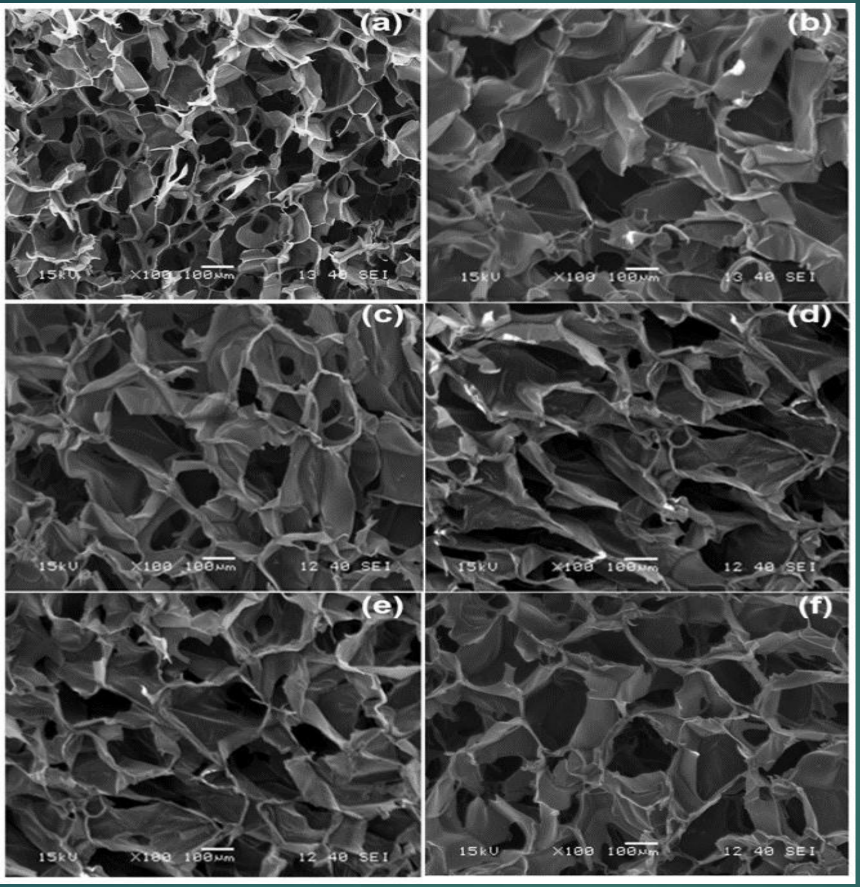


Osteotomy



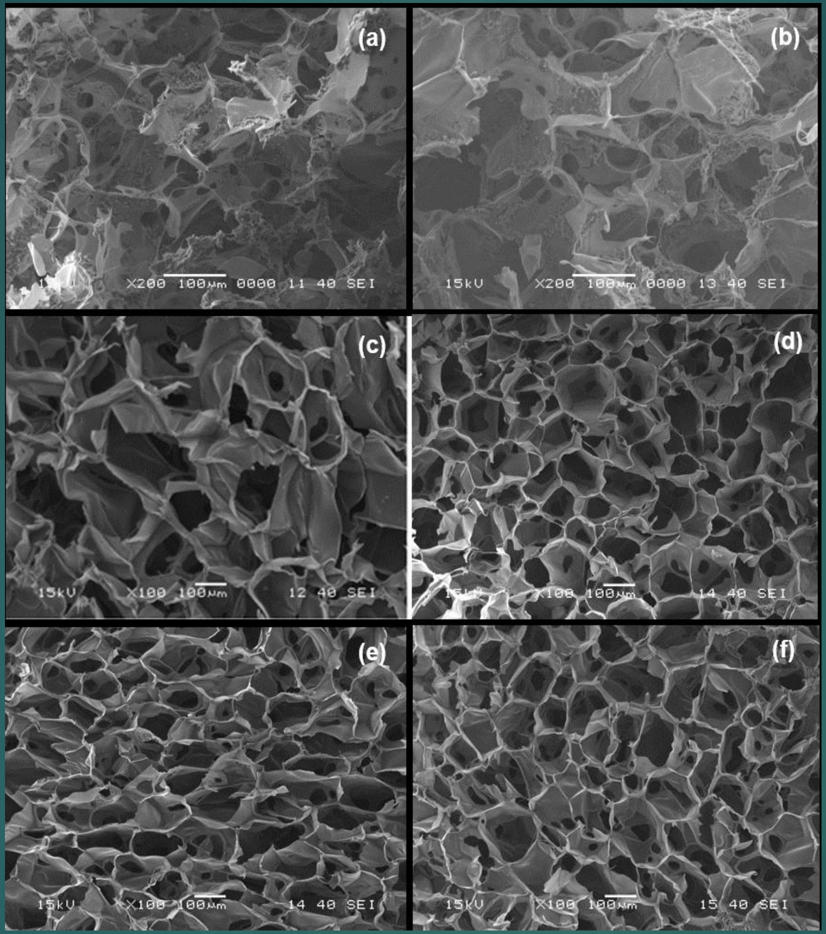
Tissue Engineered constructs

SF/CS Scaffold Biomaterial



Morphology of (a) pure SF, (b) SF/CS blend SF/CS 90 : 10, (c) SF/CS 80 : 20, (d) SF/CS 70 : 30 (e) SF/CS 60 : 40 and (f) SF/CS 50 : 50

SF/CS-Glycosaminoglycan porous scaffolds



Morphology of (a) 1% G in SF/CS(80:20) blend, (b) 2% G in SF/CS(80:20) blend, (c) SF/CS 80 : 20, (d)1% Rg G in SF/CS(80:20) blend, (e) 1.5% Rg G in SF/CS(80:20) blend and (f) 2% glycosaminoglycan in SF/CS(80:20) blend scaffolds

Conclusion

- UCB-hMSCs were successfully cultured on SF/CS-Ch porous scaffolds in spinner flask bioreactor.
- Culture condition was dominant over the effect of presence of Ch in the SF/CS scaffold particularly for promoting cell viability, metabolic activity and proliferation.
- sGAG and histology study- Ch is capable of inducing and promoting chondrocyte type matrix synthesis, irrespective of culture system.
- Immunofluorescence and qPCR study illustrated the beneficial effect of Ch in collagenous, proteoglycan matrix and gene expression in dynamic culture environment.

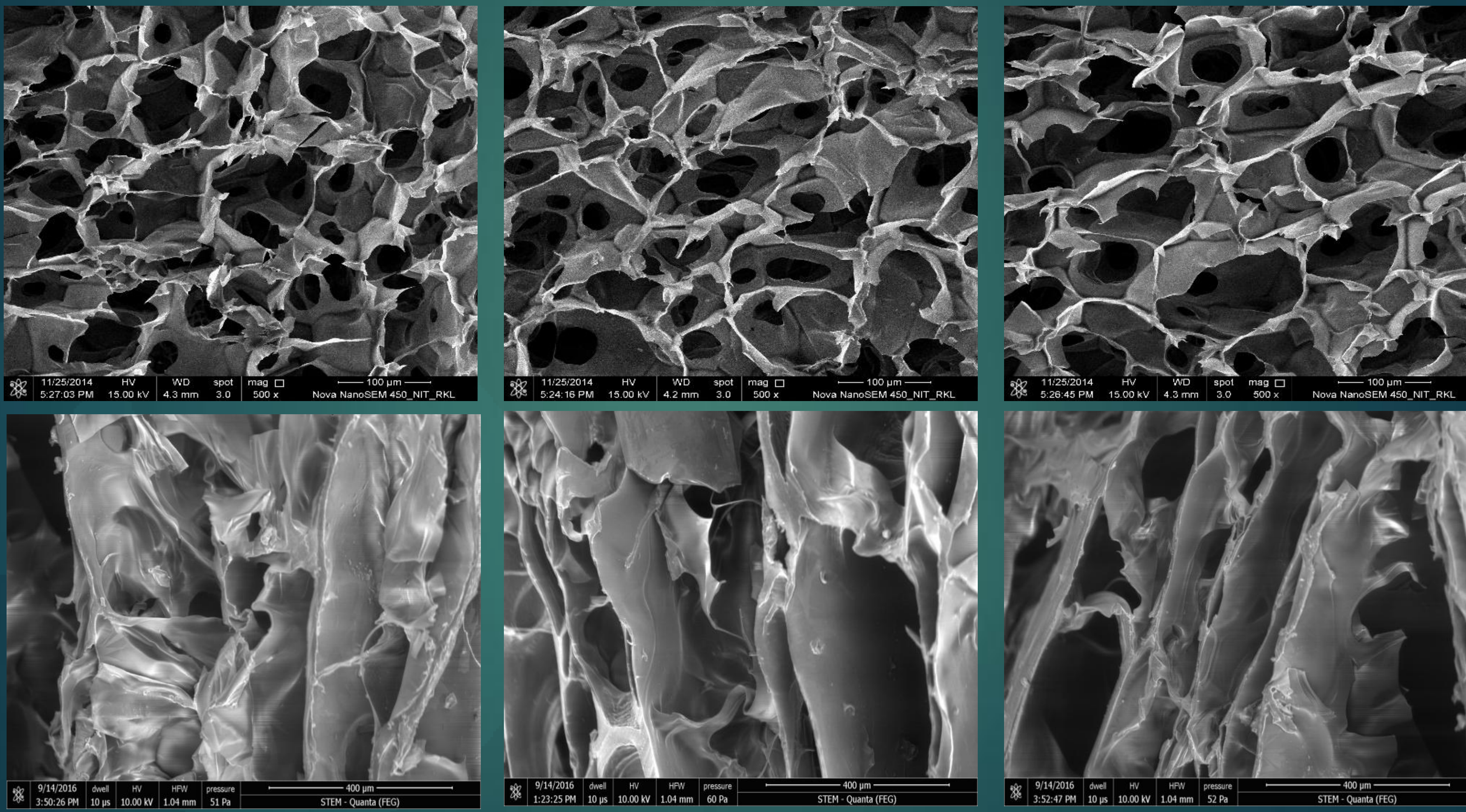
P. Agrawal et al. "Enhanced chondrogenesis of mesenchymal stem cells over silk fibroin/chitosan-chondroitin sulfate three dimensional scaffold in dynamic culture condition" Journal of Biomedical Materials Research Part B - Applied Biomaterials. 2018;106(7):2576–2587.

SEM micrograph

SF/CS-Gl-Ch0.5 | **SF/CS-Gl-Ch1.0** | **SF/CS-Gl-Ch1.5**

Transverse

Longitudinal



Conclusion

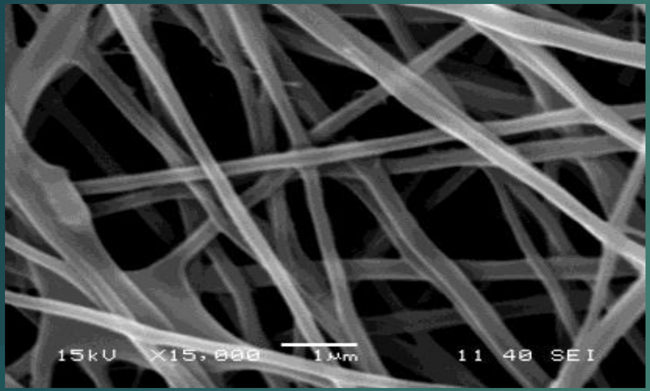
- Structural differences led to higher porosity and greater hydrophilicity in SF/CS-Gl-$Ch_{1.5}$ than 1.0 and 0.5% Ch scaffolds, facilitating cell attachment, infiltration and proliferation.
- Constructs provided an adequate cell density and metabolic activity; aid of dynamic culture facilitated sufficient nutrient distribution resulting into homogenous cartilage construct generation.
- Presence of GAG stimulating components, Gl and Ch in scaffolds, promoted differentiation and maintained chondrogenic phenotype of cells.
- Conventional pellet culture favored cell aggregation and proliferation but were less efficient in phenotype maintenance that limits their application in cartilage tissue engineering.

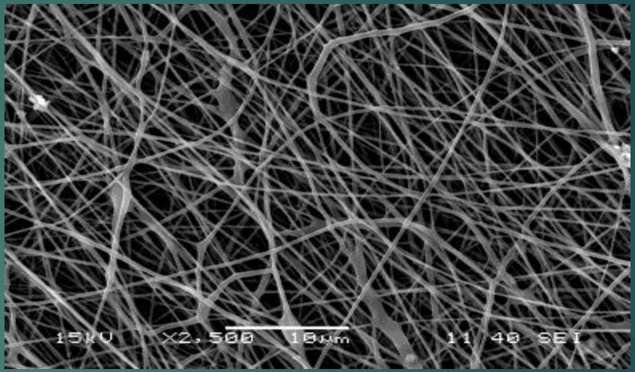
P. Agrawal *and K. Pramanik. "Fabrication of cartilage graft by differentiation of human mesenchymal stem cells over a novel glucosamine, chondroitin sulfate loaded silk fibroin/chitosan matrix under dynamic culture", Differentiation.*

Scaffold Biomaterial
Bone TE

- Electrospun nano fibrous SF/HAp nano composite
  (Eri-Tasar) Vs. Bombix Mouri
- SF/Chitosan/β-TCP composite Matrices
- Electrospun nano fibrous Nano bioglass SF/CMCcomposite

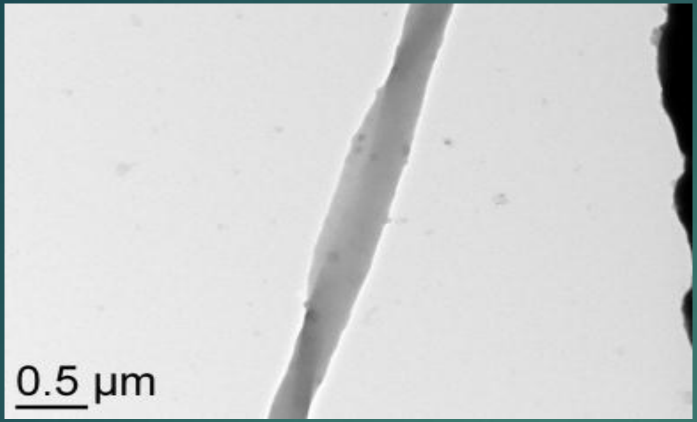
Electrospun nano fibrous SF/HAp nano composite

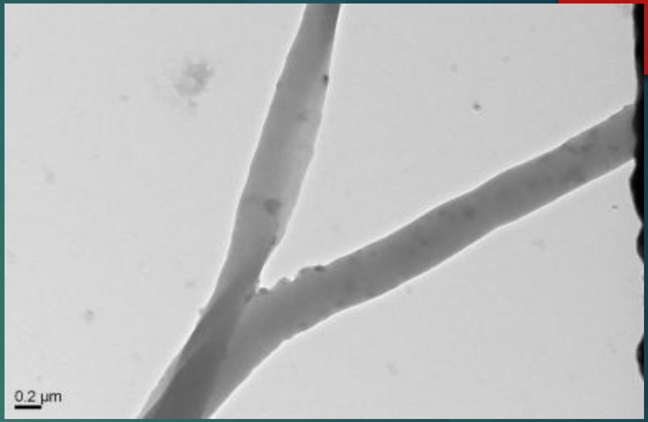


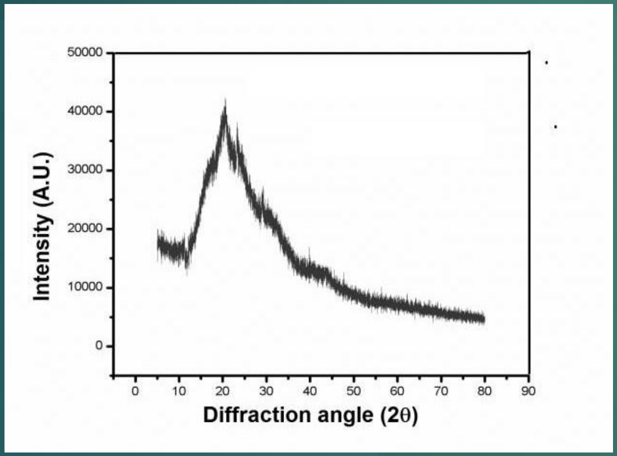


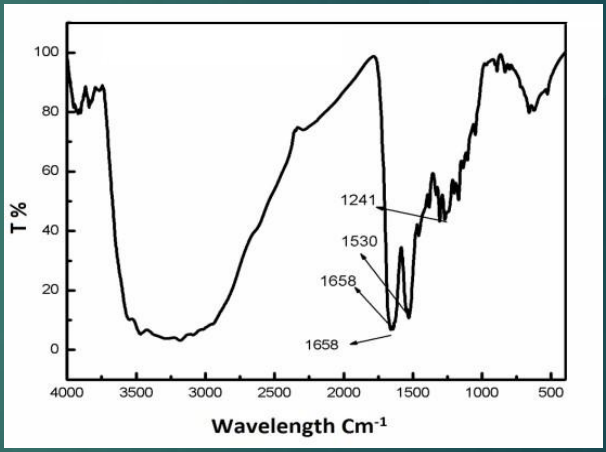
- *SEM images of eri-tasar SF blend nanofibrous mat with randomly oriented nano fibers with interconnected voids*
- ***AFM***
-

TEM images of a single nanofiber representing its shape and surface view



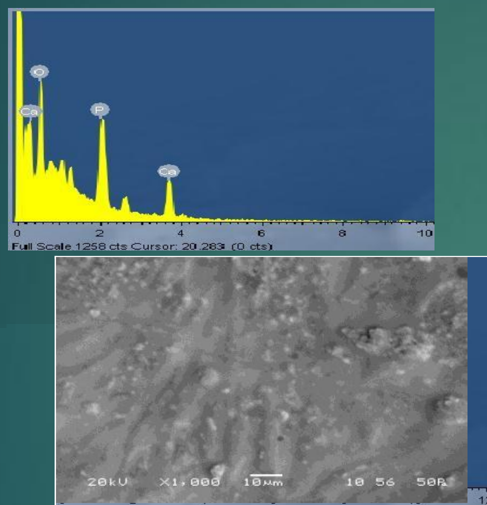




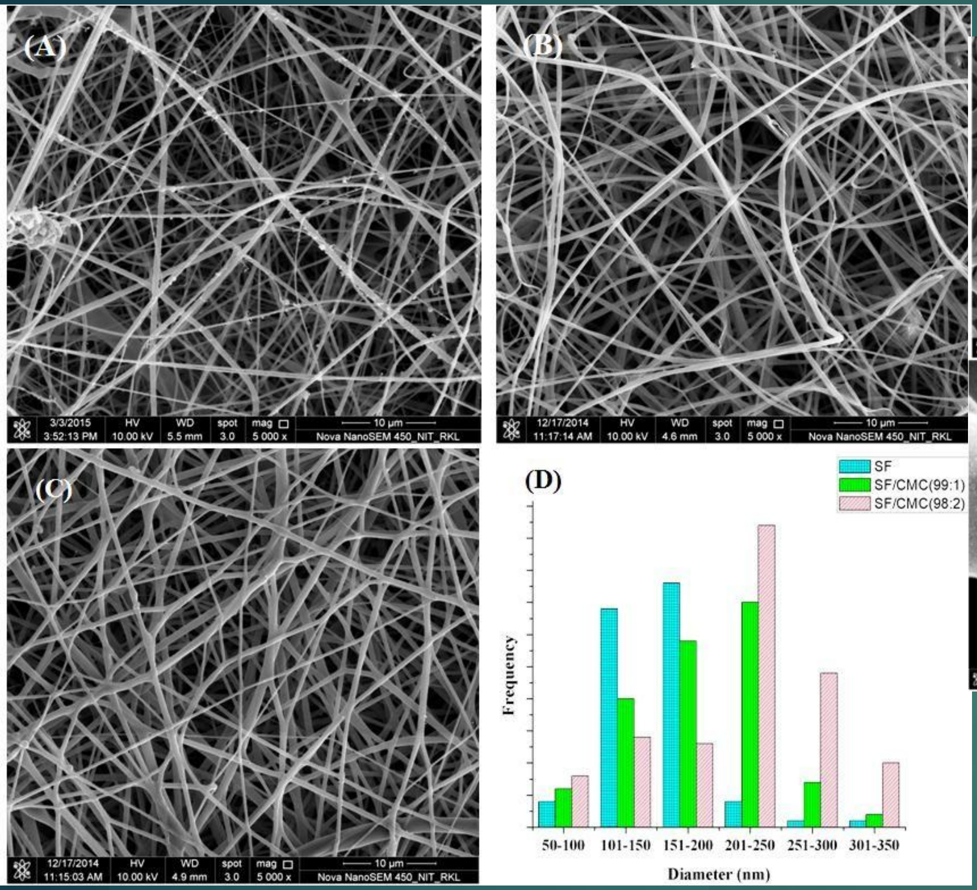


XRD & FTIR analysis of SF blend nanofiber

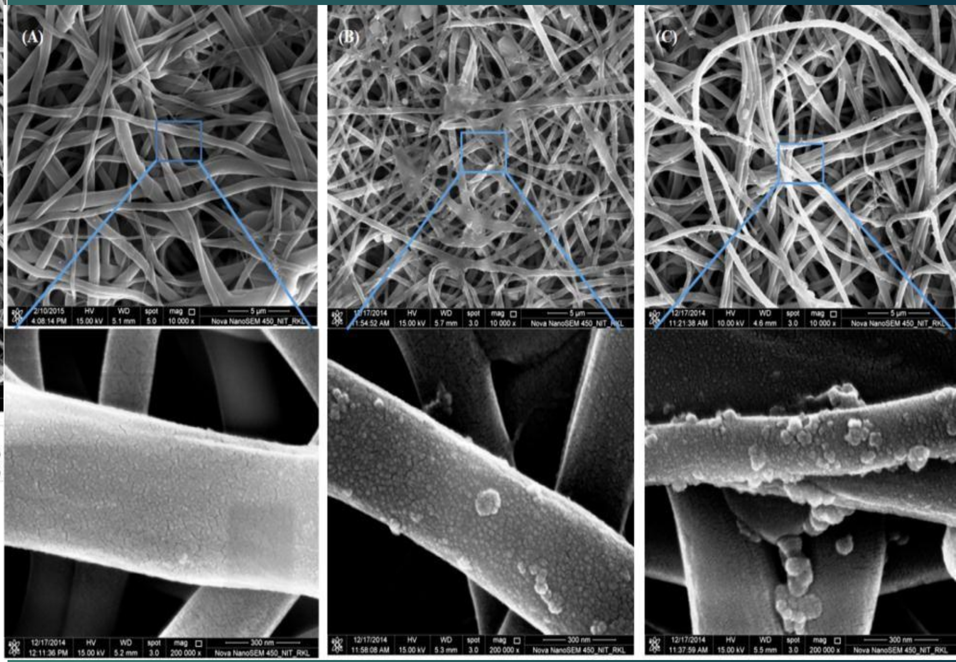
Figure 7: SEM images shows the deposition of HAp over (a) SF blend and (b) BM scaffolds and corresponding EDX figures after soaking in simulated body fluid for 14 days



SF/CMC/nBG composites

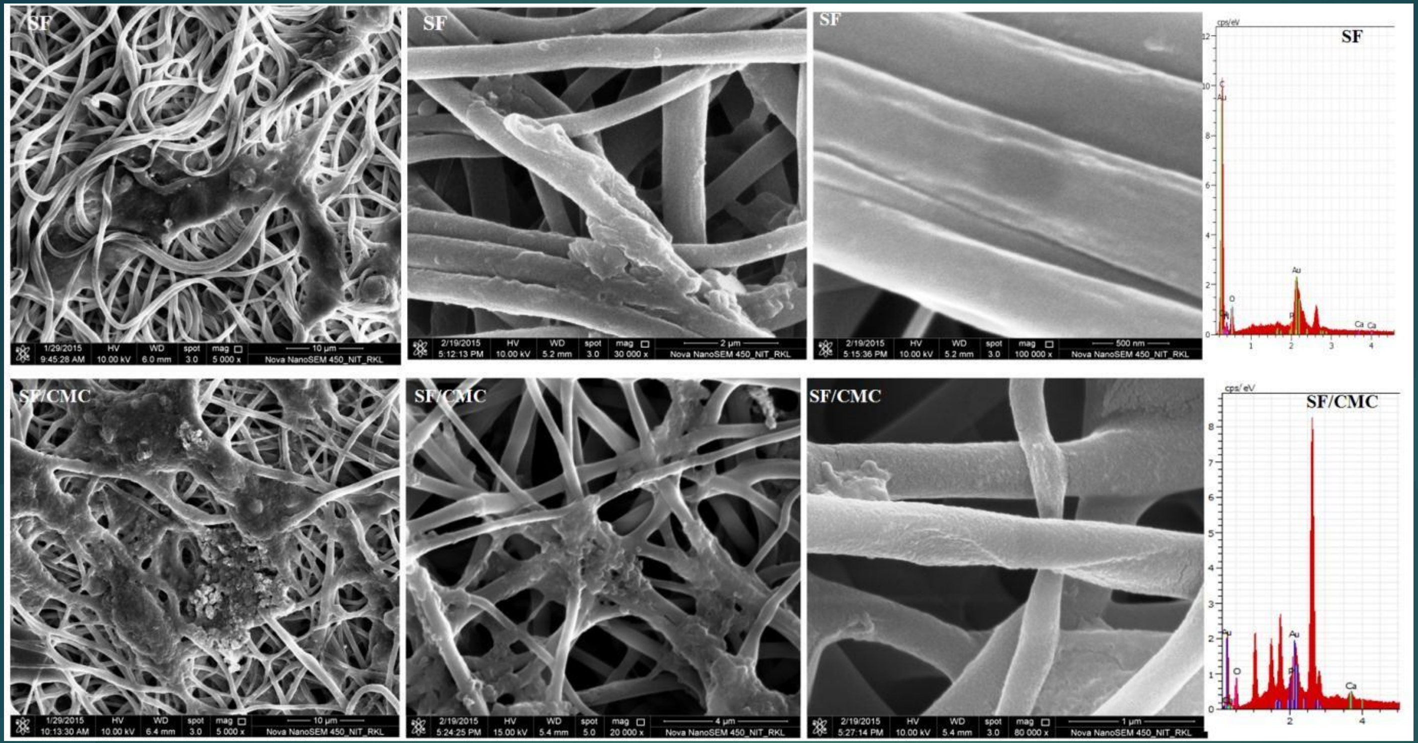


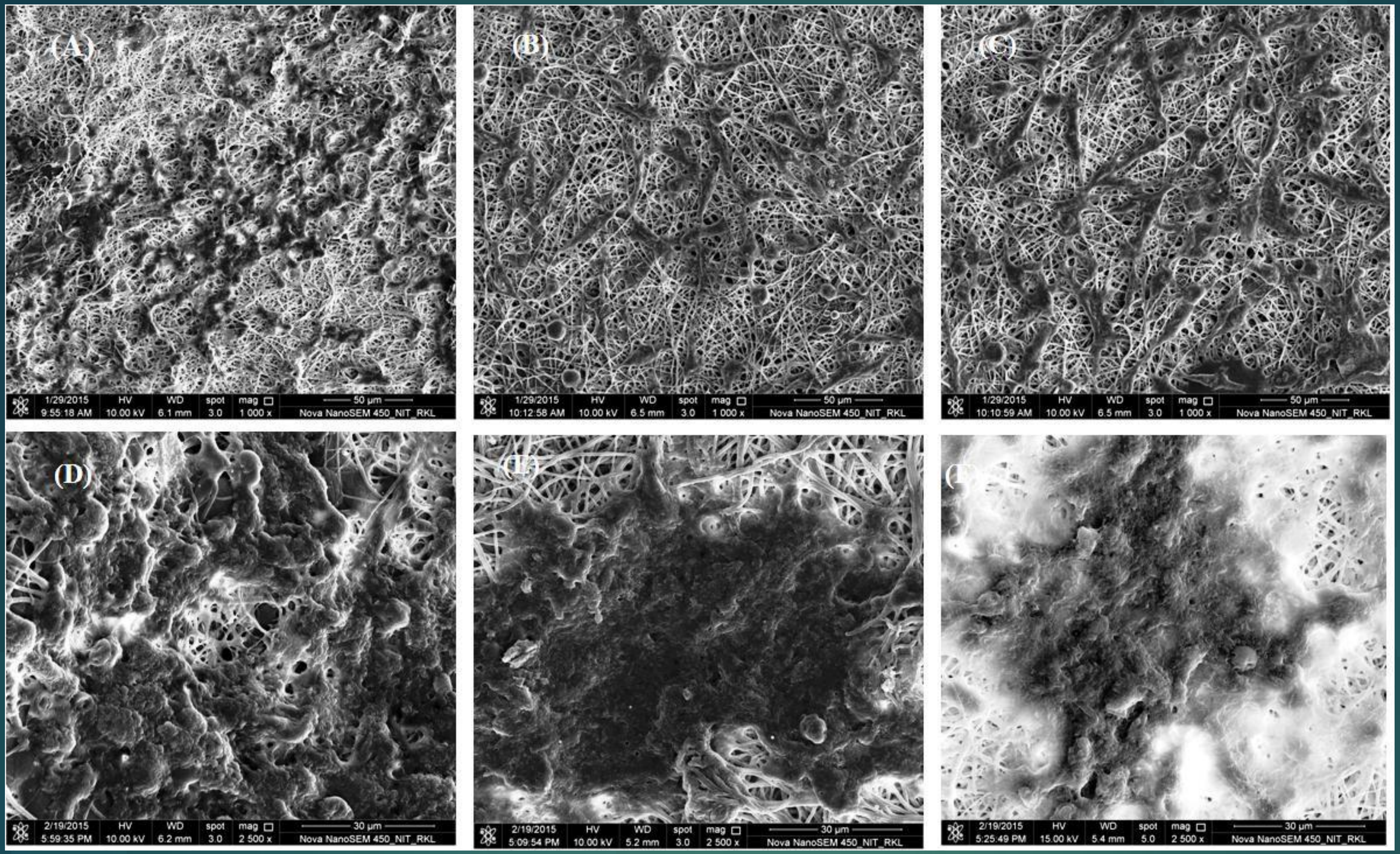
Field emission scanning electron micrographs of (A) SF, (B) SF/CMC (99:1), (C) SF/CMC (98:2), and (D) Fiber diameter distribution for SF, SF/CMC (99:1) & SF/CMC (98:2)



FESEM micrographs of scaffolds after incubation in SBF for 7 days (A) SF, (B) SF/CMC (99:1), (C) SF/CMC (98:2)

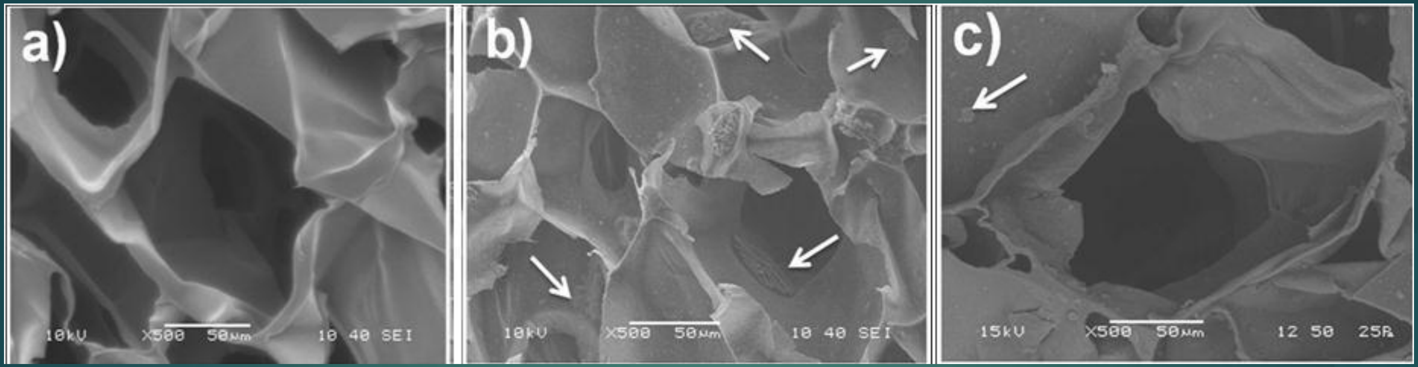
FESEM images and EDX spectra of mineral deposition of MSCs after day 7 of culture on SF and SF/CMC (98:2)



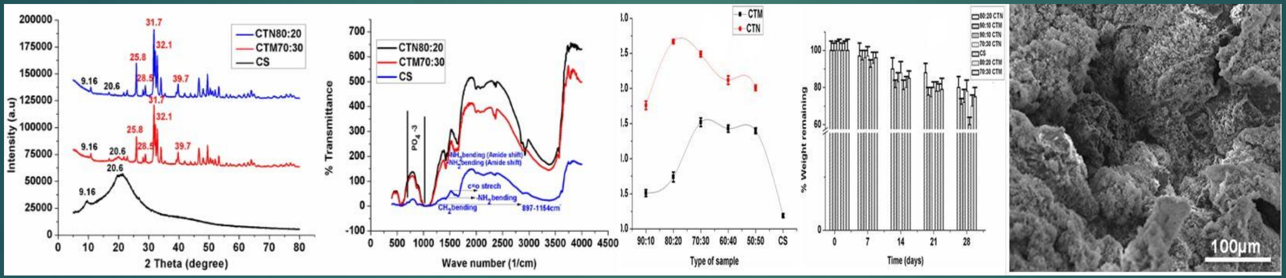


FESEM images for day 7 of cell cultured in gelatin (A), SF (B) and SF/CMC (98:2) (C).
Images for day 14 of cell cultured in gelatin (D), SF (E) and SF/CMC (98:2) (F).

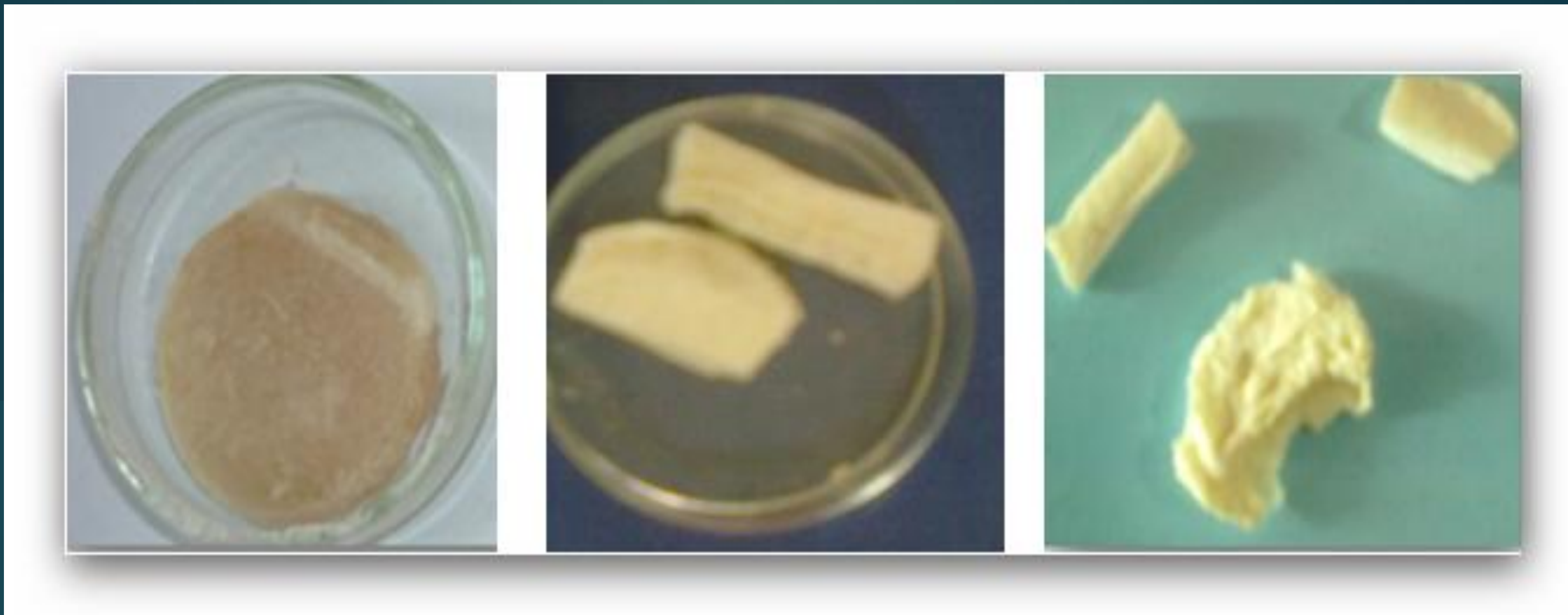
Chitosan/β-TCP composite



Morphology of CS,CS/micro β-TCP and CS/nano β-TCP composite scaffolds



Phase, structural, mechanical, degradation and bioactivity of CS, CS/micro β-TCP and CS/nano β-TCP composite scaffolds

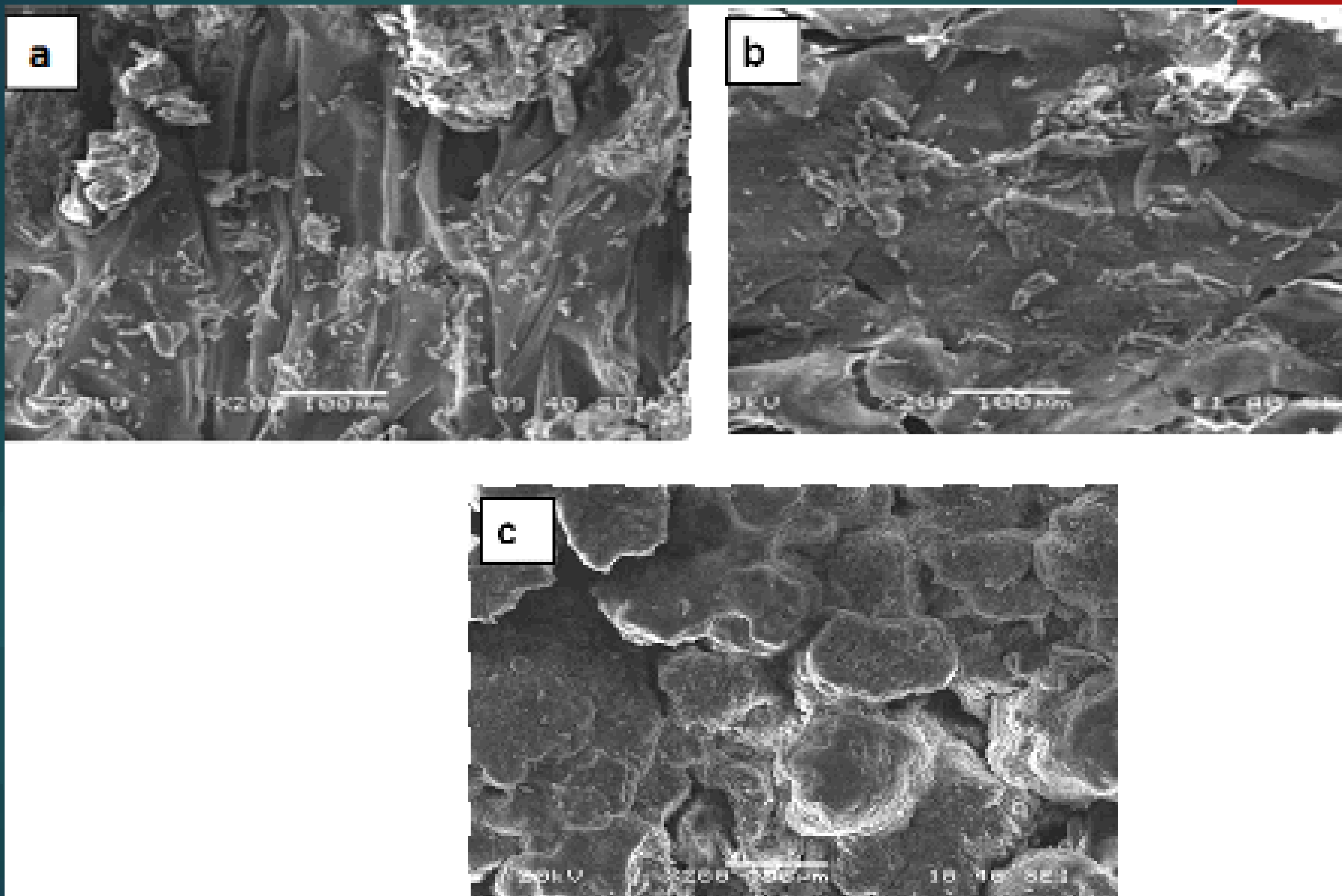


Chitosan scaffold

Cross linked Chitosan scaffold

Chitosan & β TCP composite scaffold

SEM images of a) Pure Chitosan b) Cross linked Chitosan c) Cross linked composite scaffold



Thank You