## Carbon nanofiber reinforced non-mulberry silk protein fibroin nanobiocomposite for tissue engineering applications

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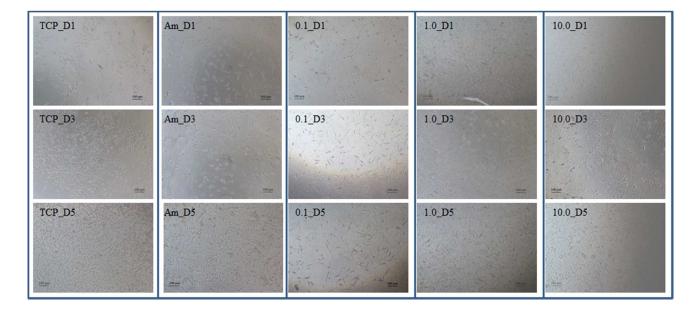
**Figure S1:** Phase contrast micrographs of L929 cells grown on TCP, AmF, AmF-0.1CNF, AmF-1.0CNF and AmF-10.0CNF coated wells for 1, 3 and 5 days. Healthy cells are observed on first four groups during the study course, whereas rounded morphology of cells is seen on the last group. Scale bar = 100  $\mu$ m.

 Table S1: Some background work on carbon nano tube (CNT)-silk fibroin based nanocomposite

 biomaterials

**Table S2:** Work already carried out on carbon nano fiber (CNF)-polymer based nanocomposite

 biomaterials for tissue engineering applications



## Supplemental figure

**Figure S1:** Phase contrast micrographs of L929 cells grown on TCP, AmF, AmF-0.1CNF, AmF-1.0CNF and AmF-10.0CNF coated wells for 1, 3 and 5 days. Healthy cells are observed on first four groups during the study course, whereas rounded morphology of cells is seen on the last group. Scale bar = 100  $\mu$ m.

SI	Method of CNT	Method of	Experimental design	Outcome/Result	Concluded applications	Refere
No	functionalization	reinforcement				nce
1.	Surfactant based functionalization	Dispersion and electrospinning of the blended solution (Solvent: sodium dodecyl benzene sulfonate)	Biophysical and cytocompatibility evaluation	Improved mechanical properties, MTT and SEM for lingua mucosa cells of Beagl dog and 3T3 cells	Matrices are cytocompatible	1
2.	As purchased	Dispersion and solvent evaporation of the blended solution (Solvent: water)	Human embryonic stem cell neuronal lineage differentiation	higher β-III tubulin and nestin expression suggest augmented neuronal differentiation	Matrices are suitable for nerve repair	2
3.	HNO <sub>3</sub> /H <sub>2</sub> SO <sub>4</sub> /HCl	/H <sub>2</sub> SO <sub>4</sub> /HCl Dispersion and solvent evaporation (Solvent: LiBr)		Cells adhered and proliferated on both the silk only and MWCNT- incorporated silk films	Matrices are cytocompatible	3
4.	As purchased	Dispersion directly in aqueous solution of silk fibroin	pH dependent stability of the dispersed blend solution	Sol-like behavior under basic (pH 12.0) and gel-like behavior under acidic (pH 4.0) conditions	Matrices may be useful for cancer detection and treatment	4
5.	High-pressure disproportionation synthesis; formic acid	Dispersion and electrospinning of the blended solution	FESEM, TEM, FTIR, RAMAN, microtensile testing	Young's modulus increases up to 460% for the reinforced matrices	More study required for proper dispersions and effects	5
6.	High-pressure carbon monoxide; formic acid	Dispersion and electrospinning of the blended solution	Effect of post spinning treatments using FTIR, RAMAN, WAXD.	7-fold increase in the strength and 35- fold increase in the modulus, more crystallinity, four times higher electrical conductivity	Matrices may be useful for load bearing applications	6
7.	As received; HNO <sub>3</sub> /HCl and formic acid	Dispersion and electrospinning of the blended solution	FESEM, TEM, RAMAN, tensile	Enhanced tensile property	Matrices may be useful for wound dressing applications	7
8.	HNO <sub>3</sub> /HCl; SDS, CTAB, TritonX100	Dispersion and stability	FESEM, TEM	Enhanced dispersion of CNT in fibroin solution	-	8
9.	HNO <sub>3</sub> /H <sub>2</sub> SO <sub>4</sub>	Dispersion and freeze drying for cryogel	FTIR, XRD, FESEM, TEM	MWCNTs induced a change in fibroin crystal structure.Mesopores and micropores are increased incryogels	-	9
10.	HNO <sub>3</sub> /HCl; Triton X100	Melt compression molding	Tensile strength, SEM.	Interfacial adhesion between PBS matrix and CNT-coated fiber improved in the composites.	-	10 11

**Table S1:** Some background work on carbon nano tube (CNT)-silk fibroin based nanocomposite biomaterials

**Table S2:** Work already carried out on carbon nano fiber (CNF)-polymer based nanocomposite biomaterials for tissue engineering applications

SI No	Method of CNF functionalization	Polymer used for base matrix	Method of reinforcement	Experimental design	Outcome/Result	Concluded applications	Reference
1.	As purchased	Polycarbonat e urethane (PCU)	Dispersion and solvent evaporation (Solvent: Chloroform [CHCl <sub>3</sub> ])	Cytocompatibility study using only fluorescent adherence count	Decreased muscle, fibroblast and chondrocyte adhesion and increased osteocyte adhesion	Matrices are cytocompatible	12
2.	As purchased	PCU	Dispersion and solvent evaporation (Solvent: CHCl <sub>3</sub> )	Mechanical and electric property. Cytocompatibility evaluations.	Support neural cell function and increased adhesion of osteoblasts.	May be useful for neural and bone tissue engineering	13
3.	As synthesized	PCU	Dispersion and solvent evaporation (Solvent: CHCl <sub>3</sub> )	Astrocytes (glial scar tissue- forming cells)	Limited astrocyte function leading to decreased scar tissue formation	Increased neuronal implant efficacy	14
4.	As purchased	PCU	Dispersion and solvent evaporation to make patterned film (Solvent: CHCl <sub>3</sub> )	Cytocompatibility evaluations	Selective adhesion and alignment of osteoblasts on CNF patterns	Electric stimulation may enhance the bone cell growth	15
5.	As purchased	Poly(lactic– co-glycolic acid)	Dispersion and solvent evaporation to make film (Solvent: Tetrahydrofuran and CHCl <sub>3</sub> )	Biophysical and cytocompatibility evaluation	Promoted cardiomyocyte and neuron cells adhesion and proliferation	Cardiovascular applicaions due to conductivity	16
6.	NaOH, water	None	Hydroxyapatite (HAp) synthesized on CNF using simulated body fluid. The purified powder sintered to make disc.	Biophysical characterizations	Strong interfacial bondings and high mechanical strength	Beneficial for load bearing tissue response	17
7.	As purchased	Polypropylen e with HAp nanorod filler	Melt compounding and injection molding of the blend powder	Biophysical and cytocompatibility evaluation	Enhanced osteoblast adhesion and proliferation by MTT study	Matrices are beneficial for bone tissue engineering	18
8.	As purchased and non-covalent surfactant absorption	Silk fibroin from <i>A.</i> <i>mylitta</i> silkworm	Dispersion and solvent evaporation to make film (Solvent: TritonX100 and sodium dodecyl sulfate solution)	Biophysical and cytocompatibility evaluation	Enhanced fibroblast adhesion and proliferation by MTT, SEM, live/dead and confocal studies	Applicable for conductive and load bearing tissue regeneration	This study

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