

Seminar Title	: Development of a novel sodium alginate/chitosan based nano-composite three-dimensional (3D) printed scaffold for bone tissue regeneration
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Abstract	<p>The present work aims to develop sodium alginate (SA) and chitosan (CH) based nano-composite scaffold by 3D printing technique for bone tissue engineering (BTE). To this end, 3D printed SA/CH scaffolds with different compositions were fabricated. The scaffolds possess open pore microstructures and interconnected pores with appropriate pore size as evident from scanning electron microscopic image analysis. Fourier-transform-infrared spectroscopic analysis revealed polyelectrolyte complex formation when SA and CH were blended, that can provide superior scaffold surface for cell attachment, proliferation, and offers ideal microenvironment for neo tissue formation. Among the scaffolds, SA/CH with 60:40 ratio exhibited controlled swelling and degradation pattern, higher tensile strength (0.387 ± 0.015 MPa) and superior apatite layer deposition ability. Scaffolds are hydrophilic and biocompatible as evident from contact angle, protein adsorption, MTT and cell attachment assessment. Thus, 3D printed scaffold with SA/CH (60/40) is proven to be a suitable substrate for tissue engineering application. The biological property of the SA/CH scaffold was improved by blending with 0-15% (v/v) gelatin (GE) thereby promotes cell adhesion, proliferation and differentiation. The resulting tri-polymer complex was used to fabricate 3D printed SA/CH/GE matrices. The microfibrillar porous scaffolds having 383-419μm pore size were revealed by SEM study. X-ray diffraction (XRD) and FTIR analyses confirmed their amorphous nature and the strong electrostatic interactions among the polymer functional groups forming polyelectrolyte complexes that may improve mechanical property and structural stability during in vivo application. The scaffolds have controlled swelling and degradation pattern, hydrophilic characteristics favorable for bone tissue regeneration. An enhanced tensile strength was obtained due to increased stiffness of SA/CH scaffolds upon addition of GE. An enhanced protein adsorption and apatite layer formation confirmed the ability of SA/CH/GE scaffolds for higher cellular adhesion and bone like environment during tissue regeneration process. MTT assay, and confocal microscopy analysis exposed a significant enhancement in cell adhesion, metabolic activity, proliferation and biomineralization activity. Furthermore, SA/CH containing 15% GE (SA/CH/GE15) has shown superior performance indicating their suitability for bone tissue engineering application. To improve osteogenic property, Bioglass (Bg) and nMgO-loaded Bg nanoparticles were synthesized and characterised. The synthesized nBg was further introduced into SA/CH/GE15 polymeric network to achieve natural bone mimetic property containing the desired inorganic and organic phase. The osteogenic and other cell supportive property of the SA/CH/GE15 scaffold were enhanced by reinforcing nBg with different concentration (0.3%-0.5 %w/v) in the polymeric network resulting in composite bioinks which were used to fabricate 3D printed SA/CH/GE15/nBg. The nano-composite scaffold have microfibrillar open pore structure with pore size range of $419 \pm 102 \mu$m to $554 \pm 68 \mu$m. The hydrophilicity of the scaffold was improved on addition of nBg with decrease in contact angle. The scaffolds exhibited controlled swelling and degradation behavior desired for BTE and enhanced compressive strength with increased nBg content in the SA/CH/GE15 scaffold and the values were 1324.63 ± 32.71 kPa and 1942.33 ± 37.56 kPa for SA/CH/GE15/nBg0.4 and SA/CH/GE15/nBg0.5 which are desired for cancellous bone regeneration. An enhanced bioactivity and protein adsorption was achieved with nBg incorporated scaffolds. MTT assay with cultured bone osteosarcoma cells on the composite scaffolds showed that SA/CH/GE15/nBg scaffolds are cytocompatible. An improved cell supportive activity (cell attachment and proliferation) was shown by nBg loaded scaffold as evident from SEM and confocal image analysis. In comparison, a higher ALP activity representing higher osteogenic property was shown by SA/CH/GE15 containing nBg0.4. The effect of reinforcement of the synthesized nMgBg in the SA/CH/GE15 network on the osteogenic ability of the 3D printed scaffold was investigated. The reinforcement of nMgBg at 0.4% and 0.5% w/v concentration with SA/CH/GE15 polymeric network did not affect the hydrophilicity, swelling, degradation and protein adsorption activity of the scaffold. The presence of Mg promoted apatite layer formation over the scaffold as revealed by in-vitro bioactivity test and SA/CH/GE15/nMg1Bg0.4 showed the highest apatite formation. In-vitro cell studies suggested that low Mg containing scaffolds SA/CH/GE15/nMg1Bg0.4 is favorable for osteoblast proliferation. The scaffold showed superior ALP and ARS activity than the scaffolds containing nBg and nMg2Bg. Overall, SA/CH/GE15/nMg1Bg0.4 was demonstrated to be the most potential substrate that can pave the way for bone tissue regeneration in future.</p>