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RESEARCH ARTICLE

# Facile Synthesis and Characterization of Chitosan Nanofibers by Oil/Water Emulsion Method

D. Ragupathy*[1], E. Muthusankar[1,2], D. Kamalakannan[3]

[1] Department of Nano Electro-Chemistry Lab (NEL), National Institute of Technology Puducherry, Karaikal 609-609, India
[1,2] Department of Electronics and Communication Engineering, National Institute of Technology Puducherry, Karaikal 609-609, India
[3] Department of Mechanical Engineering, Pondicherry Engineering College, Puducherry - 605 014, India

* Corresponding author email: ragu.nitpy@gmail.com; ragu@nitpy.ac.in

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## ABSTRACT

A facial approach for the synthesis of surfactant free, biodegradable and eco-friendly chitosan nanofibers (CS-NFS) was prepared by Oil/Water Emulsion method. The morphology of the CS-NFS was examined by field emission scanning electron microscopy (FE-SEM). FE-SEM images show the nanoscale chitosan nanofibers formation with sizes in the ranges of ~100-200 nm. Physiochemical characterizations of the CS-NFS were analyzed by Fourier transform infrared spectroscopy (FT-IR), UV-visible spectroscopy and thermogravimetric analysis. The CS-NFS are expected to be useful in electrical, optical and electrochemical devices.

**Keywords:** Chitosan. Surfactant free. Electrical. Nanofibers

## 1 Introduction

In research and scientific upgrading developments nanotechnology plays a historic task for the past numerous decades. Nanomaterials are materials with essential structural units, grains, particles, fibers or other constituent components smaller than 100 nm in at least one dimension and these nanomaterials can be made of metals, ceramics, polymers, organic materials and composite thereof, just like predictable or micron structured materials. Nanomaterials consist of nanoparticles, nano clusters, nanocrystals, nanotubes, Nanofibers, nanowires, nanorods, nanofilms, etc. So far abundant bottom-up and top-down nanofabrication technologies (such as electro spinning, phase separation, self–assembly process, chemical vapor deposition and chemical etching) are accessible to generate nanomaterials with controlled or random nanotopographies [1].

In topical material research studies, nanofiber technology was most important and precious object in the modern investigation [2]. Among the Nanomaterials, nanofibers have amazing characteristics such as exceptionally minute pore size with very outsized surface area-to-volume proportion, high porosity and diameters of the fiber was in nanometer sequence. So nanofibers can be capable materials for numerous biomedical and potential applications such as tissue templates, medical prostheses, artificial organ, wound dressings, drug delivery, pharmaceutical composition and filters, protective clothing, reinforcement in composite materials and sensor [3,4]. Owing to the current existence, there has been an increasing curiosity in edible and biodegradable fibers based on biopolymers. In these edible biopolymers, polysaccharide based multifunctional biomaterials find a continuous and growing interest. Among polysaccharides, glycosaminoglycans (chitosan) are particularly interesting for their bioactive properties [5,6]. The linear amino polysaccharide chitosan, are particularly attractive not only as abundant biomass resources but also as specialty biopolymers for preparing advanced functional materials.

Chitosan (CS) is the product of the partial deacetylation of the naturally occurring marine versatile biopolymer-chitin, which is found in the exoskeletons of insects and marine invertebrates [7]. Chitosan is composed primarily of glucosamine, 2-amino-2-deoxy-b-D-glucose, known as (1/4)-2-amino-2-deoxy-(D-glucose) [8-11]. The positive attributes of chitosan have admirable biocompatibility and excellent biodegradability with ecological safety and non-toxicity with versatile biological activities such as antimicrobial activity and low immunogenicity have provided generous opportunities for advance development. It has become of huge attention not only as an under-utilized resource but also as an efficient biomaterial of high potential in various fields, for example, biotechnology, wastewater treatment, cosmetics and biomedical areas as drug delivery vehicles, carriers of immobilized enzymes and cells, biosensors, artificial organs, orthopedic materials, surgical devices, biodegradable packaging, and in particular, temporary implants for fixations and supports in tissue regeneration [12-14]. Consequently, CS based nanomaterials finds various application and development for the future.

There have been a number of earlier attempts at reviewing the region on chitosan fibers covering certain aspects of their importance, properties and applications. Chitosan fibers situate apart from all other biodegradable natural fibers in several inherent properties such as biocompatibility, non-toxicity, biodegradability, low immunogenic, non-toxicity, etc. These properties in combination with good mechanical properties make them good candidate materials for sutures that form the largest groups of material implants used in human body [15-17]. CS fibers have been found to be valuable useful in other medical textiles [18], wound dressing [19] and haemostatic materials [20] and several other prosthetic devices such as aemostatic clips, vascular and joint prostheses, mesh and knit abdominal thoracic wall replacements and as antimicrobial agents [21].

Earlier, the nanofibers were produced by several fabrication techniques, like electro spinning [22], melt-blown [23], phase separation [24], self-assembly [25] and template synthesis [26]. Compared to other methods, interfacial polymerization is a powerful, inexpensive and simple method for the preparation of polymer based nanofibers [27-29].

The present work investigates the potential use of CS nanofiberous scaffolds synthesize by simple Oil/Water Emulsion method. The low and high MW of chitosan nanofibrous matrix with different ratios was synthesized for comparative purposes. The morphology of the nanofibers was analyzed with an average diameter about ~ 100-200 nm. The structural characterization was analyzed using FTIR and UV-Visible spectroscopy. Themogravimetic analysis was exploited to determine the thermal property of the fibers. To the best of our knowledge, reports on the synthesis of CS nanofibers by Oil/Water Emulsion method are scarce.

## 2 Materials and Methods

### 2.1 Material

Chitosan (low molecular weight (LMW); $C_6H_{11}NO_4)_n$ & (high molecular weight HMW; $C_{12}H_{24}NO_4)_n$ were obtained from Wako. Petroleum ether (PE) was used as an organic solvent. Distilled water was used as aqueous phase. Ammonium persulphate (APS), hydrochloric acid and all other reagents were analytical grade and used without further purification.

### 2.2 Synthesis of Chitosan Nano Fibrous Scaffolds (CS-NFS)

Chitosan nano fibrous scaffolds (CS-NFS) were prepared by in the interface of two immiscible solvents (water/petroleum ether). The by-products of the result are easily separated owing to their solubility in the aqueous or the organic solvents. The condensed hydrocholoric acid (HCl, 3.0 mL) and ammonium persulphate (APS, 1.14g) were dissolved in 100 mL of distilled water in a 500 mL beaker, and then petroleum ether (PE, 100mL) was added to the surface of the water solution. After ten minutes later, LMW chitosan (20, 40, 60, 80 mM) mixed with 2% aqueous acetic acid was added carefully to the petroleum ether and then reaction was started at aqueous/organic interface of water and petroleum ether. The reaction was conceded out under room temperature for 3 days without stirring. The precipitate was centrifuged and washed with distilled water for several times to remove all other impurities. The consequential products were dried at room temperature for 24 hours. Thus, CS-NFS was synthesized. The procedure as mentioned above was adopted to prepare high MW chitosan NFS.

### 2.3 Instrumentation

The morphology of CS-NFS was investigated using field emission scanning electron microscopy (FE-SEM) (TEOL JSM-5600LV instrument with the operation of 25 kV). Fourier transform infrared spectrometer (FTIR) (Nicolet, USA) was used to record the spectra of CS-NFS using KBr pellets. Thermal stability (TG-DTG) (Perkin Elmer) of samples was made at a heating rate of 10°C/min under a nitrogen atmosphere over a temperature range of 30-800 °C. Shimadzu UV-1700 model UV-vis spectrophotometer was evaluated to analyze the fibers.

## 3 Results and Discussion

### 3.1 Morphology

The FE-SEM images show the morphology of CS-NFS prepared with different CS concentrations. Fig.1 shows the FESEM images of low molecular weight CS-NFS at different concentration from 10 to 40 mM (Fig.1a – d). We could find a little fiber in fig.1a. Fig.1b shows the fine and uniform formation of CS-NF diameter in the range of 100 – 200 nm. Fig1.c and d show the aggregation of nanofibers to form bundled shaped fibers. While increasing the molar ratios of the chiotsan monomer the aggregation of the fibers tends to form bundles. As compared to other molar ratios of CS (10, 30 and 40 mM; Fig.1a, c and d), 20 mM shows the well orderly arranged nanofibers were found. While decreasing the lower concentration of chitosan to below 10 mM, we could find a film like morphology. Thus, from the FESEM image it is concluded that 20 mM is an excellent molar ratio for the CS-NFS formation.

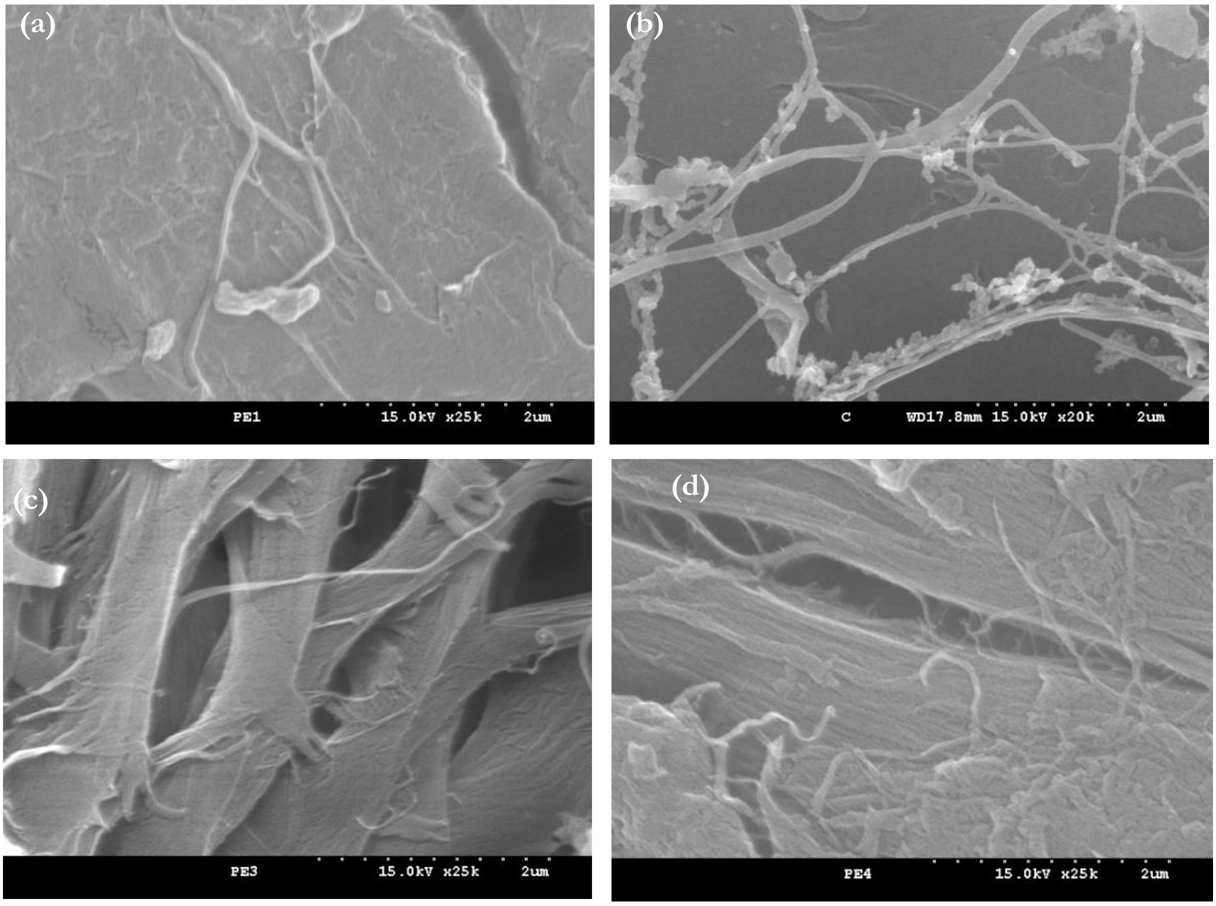

**Figure1:** *FESEM images of CS-NFS/LMW at different molar ratios* **(a)** *10* **(b)** *20* **(c)** *30 and* **(d)** *40*

Fig.2 presents the FESEM images CS-NFS/HMW at different concentration from 10 to 40 mM (Fig.1a – d). Relatively homogeneous, continuous and randomly oriented fiber marix were obtained under the optimized conditions for 10 mM (Fig. 2a), 20 mM (Fig. 2b), 30 mM (Fig. 2c) and 40 mM (Fig. 2d). As the solution concentration increased, the average fiber diameter linearly increased for all of the HMW chitosan samples. The diameter of the fiber was interrelated with chitosan ratios and solution concentrations. The ranges of the diameter distributions become fine for the HMW samples. While decreasing the lower concentration of chitosan to below 10 mM, higher quantity of fiber obtained compared to lower MW chitosan. These results indicate that the HMW chitosan were successfully fabricated into thinner and homogeneous fibers compared to those obtained using lower molecular weight chitosan. The ability of the fiber forming property was superior for the HMW chitosan than the lower molecular weight chitosan at very low concentrations.

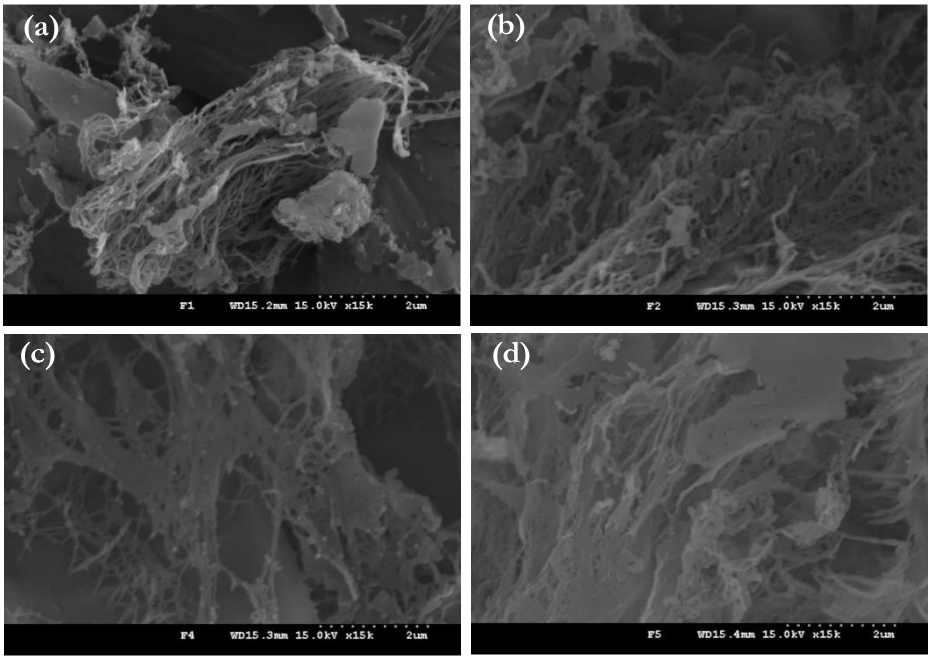

**Figure 2:** *FESEM images of CS-NFS/HMW at different molar ratios* **(a)** *10,* **(b)** *20,* **(c)** *30 and* **(d)** *40*

### 3.2 FTIR Spectroscopy

The FTIR spectra of CS-NFS were displayed in Fig.3 (a-d) at different concentrations from 10, 20, 30 and 40 mM (chitosan monomer). The characteristic bands ~ 3200 and 3420 cm$^{-1}$ due to the stretching vibration of OH and $NH_2$ groups of chitosan and the bands at 2900 and 1540 cm$^{-1}$ represented the presence of –CH and –$CH_2$ group, 1444 cm$^{-1}$ peak is due to C–N axial deformation (amine group band). The typical band around 1085 cm$^{-1}$ peak represents the C–O groups of chitosan. The peak at 1570 cm$^{-1}$ is attributed to the N–H deformation of $NH_2$ group and 1029 cm$^{-1}$ stretching vibration of C–O–C in glucose circle and 1062–1010 cm$^{-1}$ band corresponds to CH–OH in cyclic compounds [30]. Hence, these spectral evidences further augment the existence of chitosan units in the nanofibrous scaffolds.

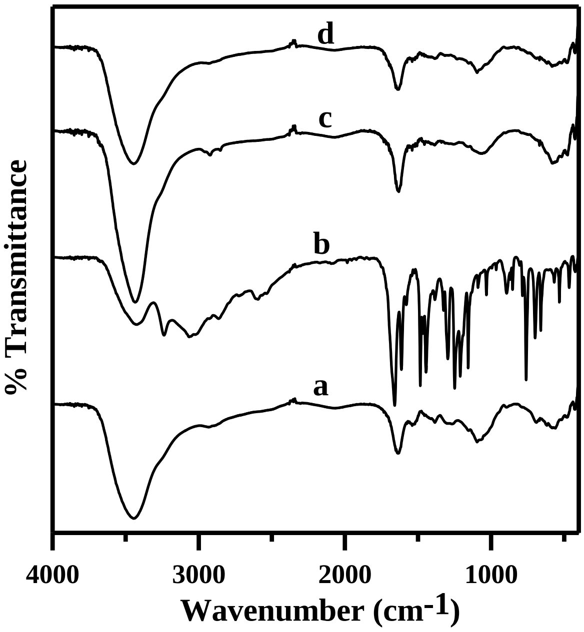

**Figure 3:** *FTIR spectra of CS-NFS/LMW at different molar ratios* **(a)** *10,* **(b)** *20,* **(c)** *30 and* **(d)** *40*

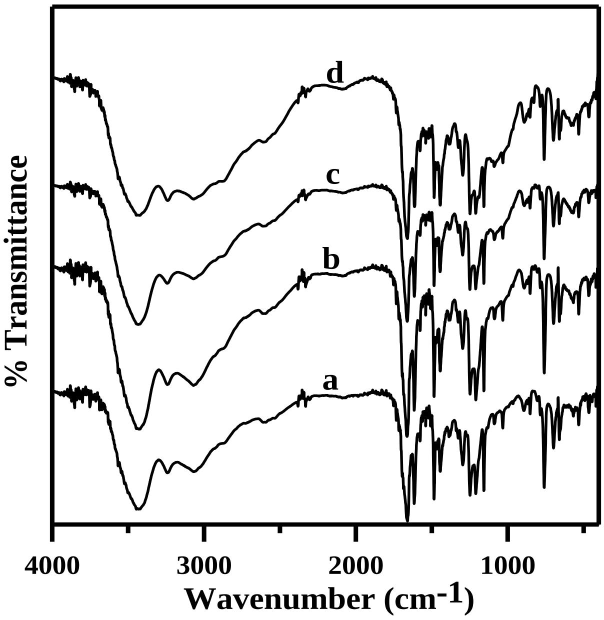

**Figure 4:** *FTIR spectra of CS-NFS/HMW at different molar ratios* **(a)** *10,* **(b)** *20,* **(c)** *30 and* **(d)** *40*

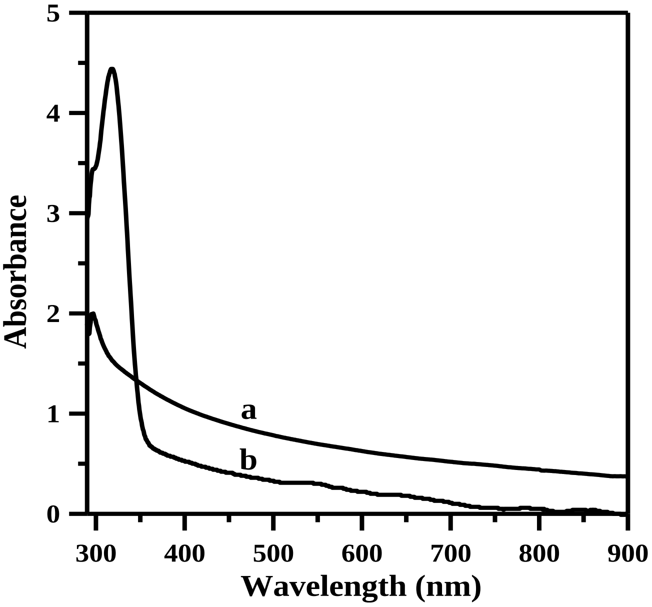

**Figure 5:** *UV-visible spectra of* **(a)** *CS-NFS/LMW and* **(b)** *CS-NFS/HMW*

Fig.4. displays the FTIR spectra of the CS-NFS/HMW. Through the exploit of a combination of the OH stretching band at 3440 cm$^{-1}$, amide I bands at 1654 and 1625 cm$^{-1}$, amide II band at 1570 cm$^{-1}$, C-H stretching band at 2871 cm$^{-1}$, the bridge oxygen stretching band at 1156 cm$^{-1}$, and the C-O stretching band at 1065 and 1032 cm$^{-1}$, the percent deacetylation can be determined [31].

### 3.3 UV-Visble Spectroscopy

Fig.5 presents the UV–visible spectrum of CS-NFS at low and high molecular weight. The absorption band seen at ~300 (Fig.5a) to 320 nm (Fig.5b) arising due to the π-π* transition of –C=O group of chitosan degradation [32]. Hence, this evidence confirms the existences of chitosan units in the nanofibrous matrix.

### 3.4 Thermal Studies

Thermal stability of the CS-NFS were evaluated with TG analysis (Fig.6 and 7 (a-d). TG analysis was carried out for all the NFM s from the temperature 25-800°C, ca. TG analysis of the CS-NFS confirms the presence of both polymer fractions in the fibers. The primary weight loss 100°C is due to loss of solvent and water from the chitosan nanofibers [33-35]. The second thermal degradation at 240°C is owing to the decomposition of chitosan nanofibers. Thus, this thermal evidence further confirms the chitosan units in the fibrous matrix.

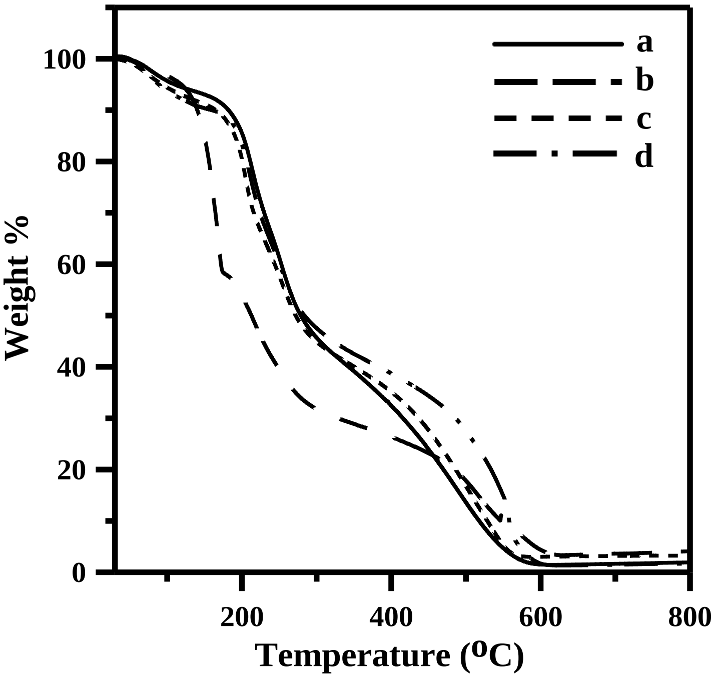

**Figure 6:** *TGA curves of CS-NFS/LMW at different molar ratios* **(a)** *10,* **(b)** *20,* **(c)** *30 and* **(d)** *40*

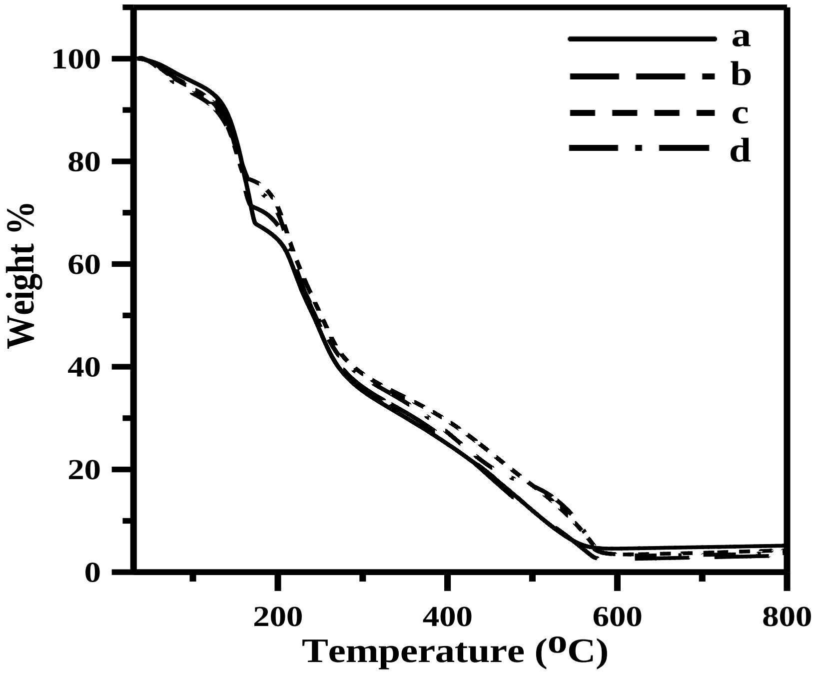

**Figure 7:** *TGA curves of CS-NFS/HMW at different molar ratios* **(a)** *10,* **(b)** *20,* **(c)** *30 and* **(d)** *40*

## 4 Conclusions

It was able to obtain bead-free, continuous, randomly oriented nanofibrous matrix through Oil/Water Emulsion method from various CS molecular weights (low and high). The fiber diameter and molar ratio of CS are correlated. The morphology of the nanofibrous matrix was examined using field emission scanning electron microscopy. The diameter of the fiber is in the range of ~100-200 nm. The structural features were characterized by UV-visible and FTIR spectroscopy. The thermal properties of the fibers also discussed. Our technology for effective production of the chitosan nanofibrous matrix will contribute to the advanced functional materials and anticipated to be useful for biosensors, tissue engineering and membrane science.

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