ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017

Electro spun PLGA-PVA nano fibrous scaffolds: promising carriers for Curcumin

[1] P.Jhansi Lakshmi, [2] S.Sundar Manoharan, [3] Pravin walke
[1][2] Dept.of Nano technology, Karunya University ,Coimbatore
[3] Center for nano science and nanotechnology(NCNNUM),University of Mumbai, Mumbai

***Abstract*— Curcumin is a naturally occurring gist with innate antimicrobial and wound healing properties. Owing to its manifold mechanism of actions, curcumin is apt at the present time existing antibiotics to first-rate for unwilling bacteria. Curcumin's pitiable aqueous solubility and speedy degradation contour hinder its usage; nano enabled drug delivery system over comes this snare and facilitates unmitigated topical deliverance of Curcumin. The current cram is paying attention on the development of PLGA-PVA nano fibers loaded with Curcumin (Cur) by electro spinning technique. PLGA and PVA were blended in the ratios of 75:25, 65:35 and 50:50 correspondingly, for the fabrication of scaffold. Curcumin was weighed down with the sample containing equal amounts of PLGA and PVA. Process optimization was carried out by standardizing the Viscosity, Molecular weight of the polymer, Voltage, temperature, tip-collector distance, Solution flow rate for fiber formation. The alteration in fiber dimension and morphology of the electro spun nano fibers was obtained by field emission scanning electron microscopy (FE-SEM).In vitro release properties of the prepared scaffolds were observed by simulated fluid conditions of pH 7.4, 37°C incubation in rotary shaker incubator. The release profile from the drug loaded fibers showed varied release profiles with respect to their Lactidyl:Glycodyl ratio variations, which confirms the dissolution of the drug in the polymer fractions. On the whole, we hypothesize the suitable carrier material for Curcumin**

***Keyword*: PLGA, Curcumin, PVA, Electro spinning, nano carrier, In vitro degradation, Bio availability, nano fibers , biodegradable, wound healing.**

INTRODUCTION

Greater than the preceding century, Curcumin,a yellow crystalline composite and the dynamic ingredient of turmeric, which is an Asian spice, cosmetic and folk remedy has been found to own anti neoplastic (6), antimicrobial (7), anti-inflammatory (8), antioxidant (9) and wound healing activities (10). Particular to its copious therapeutic targets, Curcumin has been piloted in clinical trials for innumerable disease entities, with oral doses as high as 12gms per day, tolerated unharmed (11). Conversely, Curcumin's potential for therapeutic rendition has been held up by stumpy oral bioavailability, meager aqueous solubility, and hasty degradation, swift abolition restrict its clinical applicability (12).For these reasons, many studies have been carried out to improve the bioavailability of Curcumin using different drug carriers to obviate this.
Encapsulation of Curcumin in nano particle podium is a doable and lucrative means of enabling its delivery. By positive feature of their small size and high surface –to-volume ratio, nano particles can move across the skin barrier and intra cellularly (12), which in rank is superlative for topical drug delivery. Slow, sustained release of encapsulated stuffing and as a consequence limits toxicity as the theoretical maximum amount of drug is never in contact with skin at one time. Curcumin nano formulations have been urbanized for preclinical studies on cancer, inflammation, wound healing, and other conditions, which demonstrate superior therapeutic value with non-encapsulated Curcumin (13).For the encapsulation of drugs various FDA approved polymers were widely used;among them PLGA based nano carrier systems were safe and non immunogenic controlled delivery carriers. Depending on the ratio of Lactide:Glycolide used for the polymerization, different forms of PLGA can be obtained. These are usually identified in regard to the monomer ratio used (for example. PLGA 75:25 identifies a copolymer whose composition is 75% lactic acid and 25% glycolic acid). The degradation rates of the PLGAs are dependent on many factors including the molar ratio of lactide and glycolide acids in the polymer chain, molecular weight of the polymer, the degree of crystallinity, the Tg of the polymer and nature of the incubating media.The higher the content of glycolide units, the lower the time required for degradation.

"Advances in nanotechnology-based delivery systems for Curcumin" illustrated about the potential pharmacological action(62) and advantages of the topical

ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017

applications , and enhancement of wound healing of the Curcumin for burn wounds (57,58,60) beneficial roles of Curcumin for skin diseases (59) Kumar et.al (61) explained about Conundrum and therapeutic potential of Curcumin in drug delivery and highlighted the importance of multifarious novel drug-delivery approaches, including micro emulsions, nano emulsions, liposomes, solid lipid nano particles, microspheres, solid dispersion, polymeric nano particles, and self-micro emulsifying drug-delivery systems which have been used to enhance the bioavailability and tissue-targeting ability of Curcumin.The emerging role of nano carriers (64) to increase the solubility and bioavailability of Curcumin studied by Mohanty et.al and the combined effect of PLGA and Curcumin on wound healing activity(65) by Chereddy et al, demonstrated the additive effect of lactic acid from PLGA and encapsulated Curcumin for the active healing of wounds.

In an endeavour to advance progressive Curcumin delivery, we utilized an innovative nano enabled drug delivery system to create PLGA-PVA composite nano fibers that slot in curcumin. The inventiveness of our nano fabrication allows for loading of diverse energetic ingredients, with therapeutic effectiveness when applied topically (locally)(15,16).We hypothesizes that this fabrication approach would permit for the encapsulation and incessant release of curcumin, with activity in the setting of infected burn wounds too.

MATERIALS AND METHODS:

1.1. Materials: Poly (D, L-lactide-co-glycolide) (50:50)Mw:30,000-60,000, Poly (D, L-lactide-co-glycolide) (65:35), Mw: 40,000-75,000,Poly (D, L-lactide-co-glycolide) (75:25) Mw: 66,000-107,000 and Curcumin was purchased from SIGMA, PVA (Poly (Vinyl alcohol)), Mw 85,000-124,000 was purchased from ALDRICH and Solvents, DCM (Di Chloro methane), Ethanol were of analytical grade and used without further purification. All preparations were done with Double distilled water.

1.2. Preparation of spinning dope solutions: (a).Preparation of 10% PVA solution: 100mg of 80,000Mw Poly Vinyl alcohol was dissolved in 10 ml of double distilled water. The solution was kept for magnetic stirring at 40°C temperature for 2 hrs. (b).PLGA-PVA blend: 300 mg of PLGA was dissolved in 10 ml of DCM (di chloro methane) at a temperature of 25°C with stirring for 30 min. To this solution 10% PVA solution was added drop by drop with magnetic stirring at 25°C temperature for 2 hrs. The final solution was ultra sonicated with probe sonicator for 3 min at 20mV.A uniform; homogeneous, viscous solution was obtained with translucent white colour.(c) Preparation of Curcumin solution: 300mg of Curcumin was dissolved in 10ml Ethanol.(d).PLGA-PVA-Curcumin blend: To the PLGA-PVA blend solution Curcumin solution was added drop by drop during sonication at 20mV for 3-5 min. The resultant solution was O/W emulsion (primary emulsion). To this primary emulsion 2ml of 10% PVA solution was added during sonication at 20 mV drop by drop to obtain a secondary O/W/O emulsion. The final blend (emulsion) was homogeneous, viscous blend with translucent yellow color.

1.3. Electro spinning process: Fabrication of ultra fine nano fibers were made with electro spinning set up (NaBond Technologies,Co.Limited, Hong kong).The PLGA-PVA-Curcumin blend solution loaded into a standard 10 mL plastic syringe attached to a spinneret containing an 8G stainless steel blunt ended needle. The syringe was then placed in a syringe pump at a flow rate of 1.0 ml/hr.The collector plate was covered with aluminum foil. The collector plate was placed at a distance of 14cm from the collector to needle tip. The electro spinning process was performed at standardized temperature and humidity conditions to ensure that solvent in the nano fiber mats. The mats were then dried in a vacuum dryer for 12 h at room temperature. (Fig.3)

Process optimization for electro spinning: In order to control the bead formation, one can change the applied voltage (12-22 Kv), the capillary tip to collector distance (8–15 cm) and Flow rate of solution (1.0-5ml/h). The optimum process parameters to be maintained during electro spinning for drug free and herbal drug loaded PLGA-PVA nano scaffold were finalized and they are as follows: Applied voltage. , Capillary tip to collector distance14cm and flow rate.

.

1.4. Characterization of the prepared Scaffolds:

1.4.1. a. Morphology (a) Scanning Electron Microscopy: The morphology of the electro spun nano fibers was characterized using field emission scanning electron microscopy (FE-SEM) Inspect F50 make of FEI. The images were recorded at an operating voltage of 20kV, at different magnifications with a resolution of 1.2 nm at 30kV, and 3 nm at 1 kV. The diameter distribution was calculated manually.(Fig.1.1a,2a,3a,4a,5a,6a)

b. Atomic Force Microscopy (for phase behavior analysis): Atomic force microscopy (AFM) measurements were conducted using an SPM (Brucker),

ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017

model name –Dimension Icon, in the non contact mode. Freshly prepared samples were kept under high vacuum to avoid surface contamination and then stuck on a glass slide for imaging. Each sample was imaged at multiple locations for each image. All the samples imaging was done in air at room temperature and finally images were analyzed using Gwyddion (2.39) software .(Fig.1.2b,2c,3b,3c,4b,4c,5b,5c,6b,6c).

c. Dynamic Water Contact Angle measurement(WCA) (for Hydrophilic/Hydrophobic behavior analysis):
The degree of hydrophilic/hydrophobic of the PVA, PLGA-PVA, PLGA-PVA-Curcumin nano fibrous matrixes was measured by contact angle relaxation of water droplet by sessile drop method by using a contact angle goniometer (Rame-hart Inc., USA equipped with CCD camera and RHI2001 imaging software).The 1X1 cm freshly prepared electro spun meshes which were collected on glass slides was kept under vacuum for 24hrs to avoid surface contamination. In each measurement, a droplet of de-ionized water was allowed to drop onto the membrane surface and contact angle on the left and right sides of the drop was measured until equilibrium (i.e. no further change in contact angle).Images of the solution droplet were taken using a high speed digital camera and the values of contact angle were calculated by the soft ware. The average of ten angles was reported for each sample. The contact angle measurements were performed at room temperature.(Fig.9(a,b,c,d,e,f))

1.4.2. Structure & chemistry: (a) FTIR analysis: The structure of the nano sized fibers was examined using Fourier transform infrared (FTIR) spectrometer (IRPrestige-21, Schimadzu) in the region of 400-4000cm-1 with the resolution of 4 cm-1.Dried PVA, PLGA-PVA, PLGA-PVA-Curcumin nano fiber matrixes and Curcumin powder alone were mixed with spectroscopic grade KBr (Sigma-Aldrich)in an agate mortar. The KBr sample mixture was pressed into a pellet under a 10-ton load for 2-4 min and the spectrum were recorded immediately.(Fig.7)

(b) XRD analysis: The X-ray Powder Diffractometry of Curcumin powder and PVA,PLGA-PVA,PLGA(50:50)-PVA-Curcumin,PLGA(65:35)-PVA Curcumin, PLGA (75:25)- PVA-Curcumin nano fiber matrixes was performed on a XRD-6000 (Lab-X,Shimadzu) equipped with Cu X-ray source operating at 40kV and 30mA.Calibration of the goniometer was done with the special insert at 0° position and then verified with a quartz sample reference. Measurement was done from a scan range of 5 to 90°, drive axis Theta to 2 Theta, Scan speed 10 (deg/min), sampling pitch 0.1 (deg), with a preset time of 0.60(sec) in a continuous scan mode.(Fig.8)

1.4.3.In vitro drug release study: The release profile of Curcumin from the Curcumin-loaded PLGA-PVA nano fibers will be investigated in PBS at pH 7.4 containing 30% v/v ethanol. A piece of drug-containing certain amount of the sample (5 mg) was first placed in a vial filled with 10 mL of release medium. Drug release studies were carried out at 37º¬C and 100 rotation/min (rpm) in a thermostatically shaking incubator. The amount of Curcumin released at various time intervals, up to 24 h, was determined using an ultraviolet-visible (UV–Vis) spectrometer at excitation of 431 nm.With the aid of the calibration curve of Curcumin measured in the same condition, the percentage of Curcumin release was calculated and plotted versus time according to the equation.(Fig.10)

Release (%) = (Release Curcumin/ Total loaded Curcumin) ×100(%)

In this case, 1.5 mL of sample was taken from the medium after appropriate intervals for about 24 hours and then the same volume of fresh release medium was added as replacement. A calibration curve was obtained for the drug concentration at a peak absorption wavelength of nm and a linear equation was derived by a curve-fitting method. In the assessment of drug release behavior, a cumulated amount of the released drug was calculated. The percentages of drug released from the nano fibers were plotted against time.

2.1. Morphology of electro spun nano fibers:
2.1.1. SEM analysis:
Morphology of electro spun PVA, PLGA-PVA, Cur-loaded PVA, Cur-loaded PLGA-PVA nano fiber mats and their diameter distributions were shown in (Fig.4.(1a,2a,3a,4a,5a,6a)). The incorporation of Cur into the PLGA-PVA nano fibers not only significantly varied their average diameter, but also narrowed the diameter distribution of the electro spun nano fibers. The diameter of the PLGA-PVA nano fibers was estimated to be in the distribution range of 30 to180 nm, while that of the Cur-loaded PLGA-PVA nano fibers was in the distribution range of 50 to 250 nm. This is probably due to the change in viscosity of the PLGA-PVA solution.

2.1.2. AFM for polymeric phase behavior analysis: The availability of multiple block co-polymers can cause

ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017

nano/micro scale phase separation within the bulk as well as surface of the electro spun polymeric micro/nano fibers. The presence of such separated phases can be visualized using AFM if the blended polymers are present in sufficient concentration. Such phase separation properties often result in deterioration of mechanical properties and hence can be undesirable.
The surface roughness of the PVA nano fibrous matrix was minimum, which is because of its elastic nature. From the topography image it is clear that the nano fibers surface was smooth throughout the matrix and there is no phase separation, which is depicted from the phase image.(Fig.1(1b-6b, 1c-6c).

2.1.3. Dynamic Water Contact Angle measurement (WCA): Poly (lactic acid-co-glycolic acid) exhibits a wide range of hydrophobic/hydrophilic properties depending on the LA:GA ratio.PVA is used in this study as a hydrophilic O/W surfactant .Owing to its visco-elastic nature PVA is used in this study as a blender, which was used to draw the PLGA fibers with uniform surface properties. Therefore in this study, contact angle measurements were performed to understand the influence of PVA enrichment on nano fiber surfaces and as a corollary on the whole hydrophilicity of the nano fiber meshes. Contact angle of water droplets on nano fiber meshes was recorded at 5s intervals from 0 to 50s.A significant change in contact angles was observed among drug loaded PLGA-PVA nano fibers and un doped fiber meshes. The chage in contact angle is shown in the Figure. On the whole, the result shows that the hydrophobic nature of PLGA is the suitable medium for the dissolution of Curcumin (hydrophobic material). The addition of Hydrophilic PVA to the PLGA modified the PLGA hydrophobicity and which in turn improved the hydrophilicity of the Curcumin.

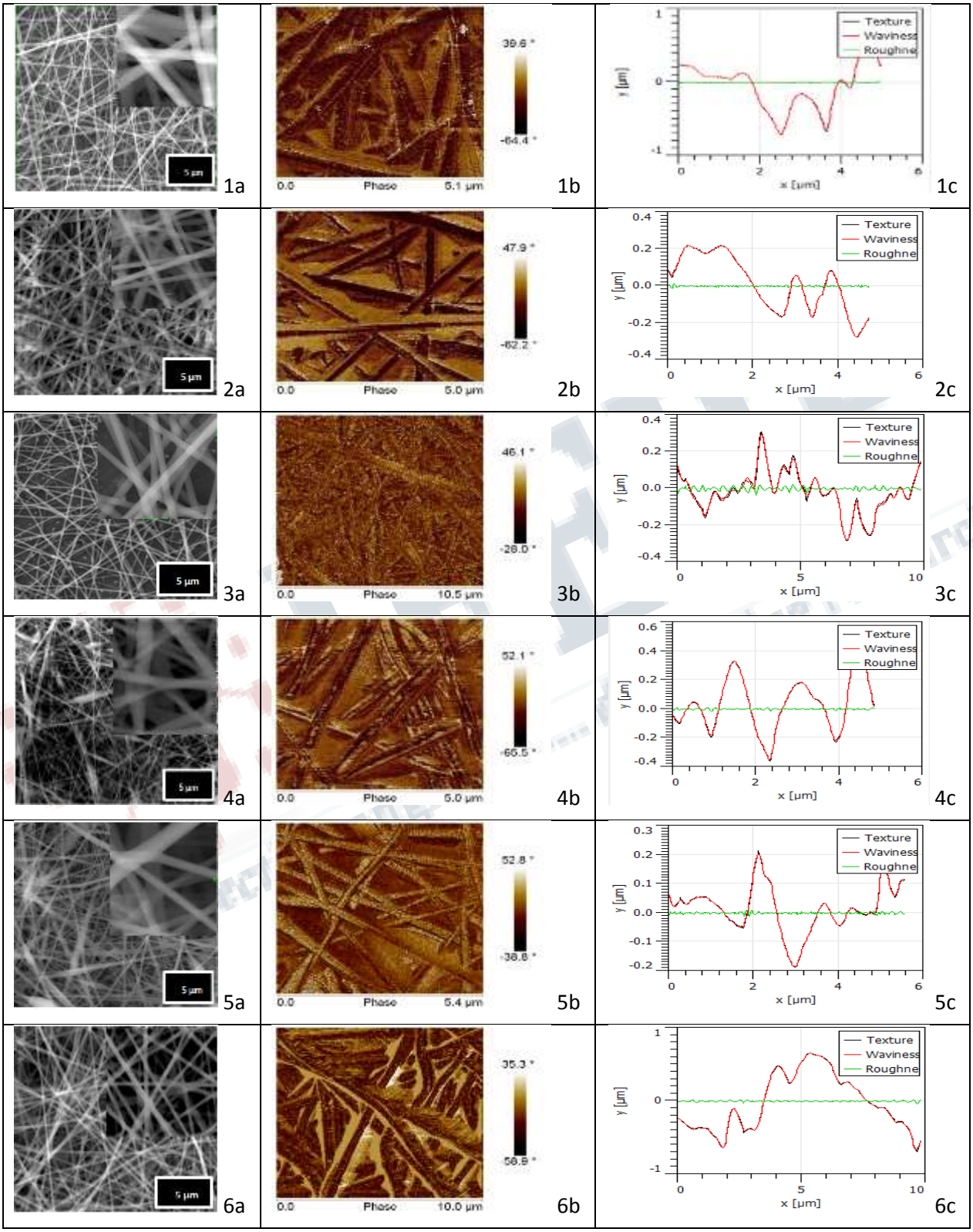
2.1.3. FTIR analysis: The FTIR spectra shows the characteristic peak of Curcumin at 3504cm-1(O-H) stretch.The bands at 1463 and 862 cm−1 are assigned to O–H in-plane and out-of-plane bending vibrations respectively. A strong band at 1759 cm−1 is due to the C-O stretching frequency of ester group and the bands at 3002 is attributed to C–H stretching frequency. The strong band at 3558 cm−1 is assigned to the O–H stretching frequency. This spectral study confirms1 the formation of PLGA, PVA and Curcumin copolymer. The FTIR spectrum shows the characteristic peak at 3500 cm−1 which can be attributed to O–H stretching vibration. The bands around 1500 and 1426 cm−1 are due to the stretching vibrations of C–C of benzene ring and bending vibration of C–H group bound to the benzene ring respectively. The peak at 865 cm−1 is due to the stretching vibration of C– O in –C–OCH3. A strong and sharp peak at 1750 cm−1 is attributed to carbonyl stretching (–C O). All major bands of both Curcumin and PLGA are not shifted to large extent but showing minor shift indicating that Curcumin and PLGA and PVA are bound together to form more stable nano fibers. From above data FTIR plot, it is difficult to conclude about Curcumin presence as there is not much change after varying the concentration of PLGA and PVA or by adding Curcumin. In order to confirm the presence of Curcumin; we have used OMINI software to subtract FTIR data of PLGA, PVA and Curcumin from PLGA and PVA. Hence it is confirmed from the peak around 1500-1700 cm−1 that Curcumin is embedded inside the co-polymer matrix.(Fig.7)

2.1.3. XRD analysis: The XRD spectrum shows the following features; Pure Curcumin exists in a crystalline state, displaying a number of characteristic reflections between 17° and 27° 2θ.PVA displays only a broad diffraction halo between 20°,22° and 25° ,typical of its semi crystalline nature. Where as in case of PVA-Curcumin nano fibers they have shown the sharp crystalline peaks at 78°, 65° and 44°. In case of PLGA-PVA nano fiber matrix, reflections were observed between 19°, 23° and 11° 2θ, where as in case of Curcumin loaded PLGA-PVA nano fiber matrixes there are no characteristic peaks of Curcumin in the spectrum. The observed phenomenon suggests that the formulation of PLGA-PVA-Curcumin involved the amorphization of Curcumin and the intermolecular interactions between Curcumin and PLGA-PVA, which is in accordance with the FTIR result. (Fig.8)

2.1.5. In vitro Curcumin release study:
Based on the calibration curve of Curcumin and absorbance of testing samples, released Curcumin concentration was calculated, and the release profile is shown in the figures. The percentage of Curcumin release was compared with the Curcumin release from cellulose acetate fiber mats reported by Suwantong et al (1), and Cur-loaded PLA nano fibers reported by T T T Mai et al (2). From the PVA-Curcumin scaffold, Curcumin showed burst release from the nano fibers during first 7 hrs, releasing almost nearly 40%of the loaded amount. This burst release may be attributed to the presence of Curcumin on or near the surface of the fibers. Then Curcumin release decreased slowly to 23%, till 24hrs.From that point, the amount of Curcumin release began to reduce gradually to 20% after 48hrs, 15% after 72hrs after, 10% after 96hrs, 5% after 120 hrs, and finally reached 1-2 % after 140hrs.

ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017



ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 2, Issue 8, August 2017

Figure: Morphology(Fig.1a-6a), Phase behaviour and surface properties (2b,2c – 6b,6c)

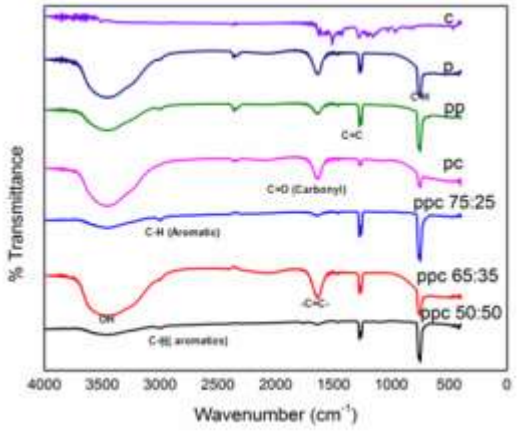


Fig.7.FTIR spectra

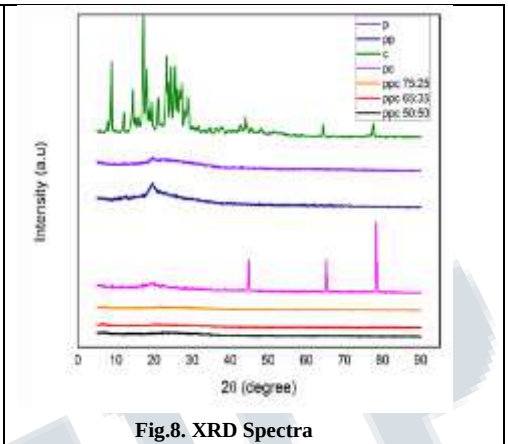


Fig.8. XRD Spectra

Fig.9.Dynamic water contact angle

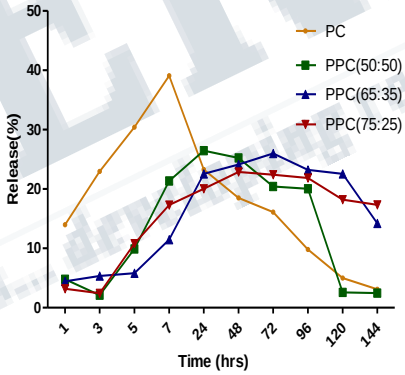


Fig.10. Drug release behaviour

C-Curcumin,P-PVA, PC-PVA-Curcumin, PPC- PLGA-PVA-Curcumin,PPC(%0:50)-PLGA(50:50)-PVA-Curcumin, PPC(65:35)-PLGA(65:35)-PVA-Curcumin, PPC(75:25)- PLGA(75:25)-PVA-Curcumin, PP-PLGA-PVA

SUMMARY & CONCLUSION:

Beginning with the morphology to until release studies PLGA (75:25) blend scaffolds showed finer excellence with reverence to PLGA (65:35), PLGA (50:50) scaffolds. Taken as a whole, from morphology, structural studies, Water contact angle measurements to anti bacterial activity testing and In vitro release study, PVA showed its it's poor carrying ability compared to PLGA-PVA blend. Hence it is clearly perceptible that PVA is not an appropriate carrier material for hydrophic materials like Curcumin.Hydrophilization of the synthetic copolymer blend (PLGA) showed improved loading and release profile of Curcumin. Among all molecular weight ratios studied sustained release for more than 7 days was observed in the case of PLGA (75:25)-PVA-Curcumin nano fibrous scaffold. Further studies by increasing the Curcumin concentration in the

said scaffold may give a path to success for activity testing. The concentration tested here was very low compared to the effective therapeutic conc. level of curcumin as per literature is (12g/day), owing to its poor bioavailability. With the help of nano strategies not only decrease in the conc. of Curcumin, we can deliver the Curcumin locally at the infection site, especially wound healing. As explained in the introduction the current PLGA-PVA scaffold can be a healthier carrier for the delivery of Curcumin to the burn wounds sites. Owing to the dual benefit given by the scaffold and Curcumin the recovery /regeneration process will speed up and skin will be scar free. Finally the nano fibrous scaffold not only acts as a good carrier, but also provides support for the skin which in turn will be useful for the cell adhesion and migration. Hydrophilization effect brought about by PVA is useful for the initial attachment of the scaffold to skin; further delivery of the material will be taken care of by the PLGA. All in all further standardization of solution , Curcumin conc. ,etc will give better quality fibers, with improved surface properties, which can be used not only for drug delivery for various biomedical applications , tissue engineering, etc. Last but not least the nano based advantage of surface area improvement can be a good advantage for the Curcumin delivery, which will take Curcumin from bench to bed side.

REFERENCES:

[1] Anand P, Sundaram C, Jhurani S, Kunnumakkara AB, Aggarwal BB. Curcumin and cancer: an "old-age" disease with an "age-old" solution. Cancer Lett 2008; 267(1):133-64.

[2] Gunes H, Gulen D, Mutlu R, Gumus A, Tas T, Eren Topkaya A. Antibacterial effects of curcumin: an in vitro minimum inhibitory concentration study. Toxicol Ind Health 2013.

[3] Sandur SK, Ichikawa H, Pandey MK, Kunnumakkara AB, Sung B, Sethi G, et al. Role of pro-oxidants and antioxidants in the anti-inflammatory and apoptotic effects of curcumin (diferuloylmethane). Free Radic Biol Med 2007;43(4):568-80)

[4] Somparn P, Phisalaphong C, Nakornchai S, Unchern S, Morales NP. Comparative antioxidant activities of curcumin and its demethoxy and hydrogenated derivatives. Biol Pharm Bull 2007;30(1):74-8.

[5] Kulac M, Aktas C, Tulubas F, Uygur R, Kanter M, Erboga M, et al. The effects of topical treatment with curcumin on burn wound healing in rats. J Mol Histol 2013;44(1):83-90.

[6] Gupta SC, Patchva S, Aggarwal BB. Therapeutic roles of curcumin: lessons learned from clinical trials. AAPS J 2013;15(1):195-218.

[7] Flora G, Gupta D, Tiwari A. Nano curcumin: a promising therapeutic advancement over native curcumin. Crit Rev Ther Drug Carrier Syst 2013;30(4):331-68.

[8] Chereddy KK, Coco R, Memvanga PB, Ucakar B, des Rieux A, Vandermeulen G, et al. Combined effect of PLGA and curcumin on wound healing activity. J Control Release 2013;171(2):208-15.

[9] Mustafa Kulac, Cevat Aktas, Feti Tulubas, Ramazan Uygur, Mehmet Kanter, Mustafa Erboga, Mehmet Ceber, Birol Topcu, Oguz Aslan Ozen , The effects of topical treatment with curcumin on burn wound healing in rats, Journal of Molecular Histology February 2013, Volume 44, Issue 1, pp 83-90.

[10] Sidhu GS, Singh AK, Thaloor D, Banaudha KK, Patnaik GK, Srimal RC, Maheshwari RK. Enhancement of wound healing by curcumin in animals, Wound Repair Regen. 1998 Mar-Apr; 6(2):167-77.

[11] Thangapazham RL, Sharma A, Maheshwari RK. Beneficial role of curcumin in skin diseases, Adv Exp Med Biol. 2007; 595:343-57.

[12] Manikandan Panchatcharam, Sumitra Miriyala, Vinaya Subramani Gayathri, Lonchin Suguna, Curcumin improves wound healing by modulating collagen and decreasing reactive oxygen species, Molecular and Cellular Biochemistry October 2006, Volume 290, Issue 1-2, pp 87-96

[13] Thangapazham RL, Sharad S, Maheshwari RK. Skin regenerative potentials of curcumin, Biofactors.2013 Jan-Feb;39(1):141-9.

[14] Kumar A, Ahuja A, Ali J, Baboota S, Conundrum and therapeutic potential of curcumin in drug delivery. Crit Rev Ther Drug Carrier Syst.2010;27(4):279-312.

[15] Sun M, Su X, Ding B, He X, Liu X, Yu A, Lou H, Zhai G. Advances in nanotechnology-based delivery systems for curcumin. Nano medicine (Lond). 2012 Jul; 7(7):1085-100.

[16] Liu J, Chen S, Lv L, Song L, Guo S, Huang S. Recent progress in studying curcumin and its nano-preparations for cancer therapy. Curr Pharm Des. 2013; 19(11):1974-93.

[17] Mohanty C, Das M, Sahoo SK. Emerging role of nano carriers to increase the solubility and bioavailability of curcumin.Expert Opin Drug Deliv. 2012 Nov;9(11):1347-64.

[18] Chereddy KK, Coco R, Memvanga PB, Ucakar B, des Rieux A, Vandermeulen G, Préat V,Combined effect of PLGA and Curcumin on wound healing activity, J Control Release. 2013 Oct 28; 171(2):208-15.

[19] Stevens MM, George JH. Exploring and engineering the cell surface interface. Science 2005 Nov 18; 310(5751):1135-1138)

[20] Maedeh Zamani1 Molamma P Prabhakaran2 Seeram Ramakrishna1,Advances in drug delivery via electro spun and electro sprayed nano materials, International Journal of Nano medicine, 2013:8.

[21] Martinez LR, Han G, ChackoM,Mihu MR, JacobsonM, Gialanella P, et al. Antimicrobial and healing efficacy of sustained release nitric oxide nano particles against Staphylococcus aureus skin infection.J.Invest Dermatol .2009;129(10):2463-9.,

[22] Nacharaju P, Tuckman-Vernon C, Maier KE, Chouake J, Friedman A, Cabrales P, et al. A nano particle delivery vehicle for S-nitroso-N-acetyl cysteine: sustained vascular response. Nitric Oxide 2012;27(3):150-60.