Indomethacin electrospun nanofibers for colonic drug delivery: in vitro dissolution studies

Article  (Accepted Version)


This version is available from Sussex Research Online: http://sro.sussex.ac.uk/id/eprint/67737/

This document is made available in accordance with publisher policies and may differ from the published version or from the version of record. If you wish to cite this item you are advised to consult the publisher’s version. Please see the URL above for details on accessing the published version.

Copyright and reuse:
Sussex Research Online is a digital repository of the research output of the University.

Copyright and all moral rights to the version of the paper presented here belong to the individual author(s) and/or other copyright owners. To the extent reasonable and practicable, the material made available in SRO has been checked for eligibility before being made available.

Copies of full text items generally can be reproduced, displayed or performed and given to third parties in any format or medium for personal research or study, educational, or not-for-profit purposes without prior permission or charge, provided that the authors, title and full bibliographic details are credited, a hyperlink and/or URL is given for the original metadata page and the content is not changed in any way.

http://sro.sussex.ac.uk
Accepted Manuscript

Title: Indomethacin Electrospun Nanofibers for Colonic Drug Delivery: In vitro dissolution studies

Author: Abbas Akhgari Zohreh Heshmati Hadi Afrasiabi Garekani Fatemeh Sadeghi Atena Sabbagh Behzad Sharif Makhmalzadeh Ali Nokhodchi

PII: S0927-7765(16)30872-4
Reference: COLSUB 8312

To appear in: Colloids and Surfaces B: Biointerfaces

Received date: 11-10-2016
Revised date: 20-12-2016
Accepted date: 21-12-2016

Please cite this article as: Abbas Akhgari, Zohreh Heshmati, Hadi Afrasiabi Garekani, Fatemeh Sadeghi, Atena Sabbagh, Behzad Sharif Makhmalzadeh, Ali Nokhodchi, Indomethacin Electrospun Nanofibers for Colonic Drug Delivery: In vitro dissolution studies, Colloids and Surfaces B: Biointerfaces http://dx.doi.org/10.1016/j.colsurfb.2016.12.035

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.
Indomethacin Electrospun Nanofibers for Colonic Drug Delivery: In vitro dissolution studies

Abbas Akhgari¹*, Zohreh Heshmati², Hadi Afrasiabi Garekani³, Fatemeh Sadeghi¹, Atena Sabbagh¹, Behzad Sharif Makhmalzadeh², Ali Nokhodchi⁴,*

¹Targeted Drug Delivery Research Center, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran

²Nanotechnology Research Center and School of Pharmacy, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran

³Pharmaceutical Research Center, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran

⁴Pharmaceutics Research Laboratory, School of Life Sciences, University of Sussex, Brighton, BN1 9QJ, UK

Number of words: 4754 (including references)

Number of Tables: 2

Number of Figures: 4

*Corresponding authors:

Ali Nokhodchi (a.nokhodchi@sussex.ac.uk; Tel: +44 1273872811)

Abbas Akhgari (akhgaria@mums.ac.ir; Tel: +98-5138823255)
Graphical abstract

Highlights

- Electrospun nanofibers of indomethacin were prepared
- Full factorial design was used for statistical optimization
- Dissolution of drug nanofibers can be modified using different polymers
- Optimum formulation exhibited appropriate drug release as a colon delivery system

Abstract

Generally, although the conventional drug delivery systems, such as using only pH-dependent polymers or time-dependent release systems are popular, the individuals’ variations of physiological conditions usually lead to premature or imperfect drug release from each of these systems. Therefore, a combination of pH- and time-dependent polymers could be more reliable for delivering drugs to the lower GI tract such as colon. To this end, electrospinning method was used as a fabrication approach for preparing electrospun nanofibers of indomethacin aimed for colon delivery. Formulations were prepared based on a $3^2$ full factorial design. Independent variables were the drug:polymer ratio (with the levels of 3:5, 4.5:5 and 6:5 w/w) and Eudragit S:Eudragit RS w/w ratio (20:80, 60:40 and 100:0). The evaluated responses were drug release at pH 1.2, 6.4, 6.8 and 7.4. Combinations of Eudragit S (ES), Eudragit RS (ERS) and drug based on factorial design were loaded in 10 ml syringes.
Electrospinning method was used to prepare electrospun nanofibers from electrospinning solutions. Conductivity and the viscosity of the solutions were analyzed prior to electrospinning. After collection, the nanofibers were evaluated in terms of morphology and drug release. It was shown that the ratio of drug:polymer and polymer:polymer were pivotal factors to control the drug release from nanofibers. A formulation containing Eudragit S:Eudragit RS (60:40) and drug:polymer ratio of 3:5 exhibited the most appropriate drug release as a colon delivery system with a minor release at pH 1.2, 6.4 and 6.8 and major release at pH 7.4. Nanofibers resulted from this formulation were also more uniform and contained fewer amounts of beads. It was demonstrated that the electrospinning could be regarded as a modern approach for the preparation of colon drug delivery systems leading to marketable products.

**Keywords:** Colon delivery; Electrospinning; Nanofiber; Eudragit; Indomethacin, Release rate.

1. **Introduction**

In recent years, various methods have been examined for oral drug delivery to the colon. Among those techniques regarding drug delivery to the colon, pH-dependent and time-dependent drug delivery systems are popular [1]. Despite the popularity of the above drug delivery systems, it should be mentioned that individual variations of physiological conditions usually lead to premature or imperfect drug release from each of these systems [2]. Therefore, a combination of pH- and time-dependent polymers could be more reliable for delivering drugs to the lower GI tract and preferably to the terminal ileum and the first part of the colon which are the predominate sites of inflammation in colon diseases such as Crohn’s disease or ulcerative colitis [3].
Nowadays, nanofibers with diameters in the nanometer range have gained attention due to remarkable characteristics such as high porosity and large surface area to volume ratio which mostly improves the function of the incorporated materials [4, 5]. As a consequence, the application of these materials in biomedical usages such as tissue engineering [6, 7], wound dressing [8], and enzyme immobilization [9] has been widespread. Nanofibers have been used as drug carriers in the drug delivery system due to their high functional characteristics. Different controlled drug release profiles, such as immediate, sustained, and biphasic releases could be achieved by electrospun nanofibers. Moreover, fabrication of nanofibers using electrospinning is a simple and straightforward process. Polymer nanofibers with a wide range in size can be manufactured using electrospinning. The nanofibers have other remarkable characteristics such as high porosity and large surface area to volume ratio which mostly improves the function of the incorporated materials. Also, fibers have high encapsulation efficiency as there is no loss during the preparation [10-17].

In electrospinning, the induction of an electric charge in the surface of a polymeric solution gives an electric charge to the liquid. By extruding this charged polymeric solution through a nozzle the droplets of the solution are elongated by the electrostatic force operating between droplet and substrate. The nanofiber is formed by solvent evaporation from the droplet and the fabricated fibers can be collected in the collector plate [18-20].

Certain studies have been accomplished to produce an oral drug delivery system based on electrospun nanofibers [21-23]. Nevertheless, very limited work has been published on the application of these nano scale fibers in colonic drug delivery. In a study, Eudragit L 100-55 nanofibers containing diclofenac sodium were fabricated by electrospinning and they showed that nanofibers had a pH-dependent drug release profile and therefore have the potential as a colonic drug delivery system for diclofenac [24]. Similar results have been found for diclofenac-loaded Eudragit L100 nanofibers produced by a modified coaxial electrospinning
technology [25]. In addition, shellac nanofibers loaded with ferulic acid has been prepared in another investigation using a coaxial electrospinning process for colon delivery [26].

A colon drug delivery system using core/shell fibers prepared with Eudragit S100 and ethyl cellulose has already been reported [27] where, ethyl cellulose was used as a sustained release polymer in the core of nanofibers while Eudragit S as a pH-dependent polymer was in the shell. To best of our knowledge, the combination of pH- and time-dependent systems in the matrix of nanofibers has not been explored for colon delivery. Therefore, in the present study, electrospun nanofibers were fabricated using a combination of two different approaches i.e. pH- and time-dependent systems in the matrix of nanofibers providing a colon-specific drug delivery system. Eudragit S100 and Eudragit RS100 were used as pH-dependent and time-dependent polymers, respectively. Indomethacin was used as a model drug due to its potential treatment for colon cancer [28, 29].

2. Materials and methods

2.1 Materials

Indomethacin (Darupakhsh, Iran), Eudragit RS100 (ERS)(Rohm Pharma, Germany), Eudragit S100 (ES)(Rohm Pharma, Germany), KH₂PO₄ (Merck, Germany), NaOH (Merck, Germany), and hydrochloric acid (Merck, Germany) were obtained from the indicated sources.

2.2 Experimental design

A 3² full factorial design was used for the design of formulations. Independent variables were the ratio of Eudragit S: Eudragit RS (with the levels of 20:80, 60:40 and 100:0 w/w)(X₁) and drug:polymer ratio (with the levels of 3:5, 4.5:5 and 6:5 w/w)(X₂). Dependent variables (responses) were as follows:
Y₁: drug release at pH 1.2 for 2 h; Y₂: release at pH 6.4 for 1 h; Y₃: release at pH 6.8 for 2 h; and Y₄: release at pH 7.4 for 2 h. Although, the dissolution at pH 1.2 was performed for 4 h and in other pHs for 10 h, the selected dissolution times in the experimental design were on the basis of the residence time of particles in the GI tract. Table 1 depicts compositions of experimental formulations.

2.3 Preparation of spinning solutions

Solutions of a combination of ERS and ES (2.5% w/v) were prepared in ethanol as a good solvent. Separately, indomethacin (1.5% w/v) solution was prepared in ethanol. Afterward, the solutions were mixed with each other. The ratios of ES:ERS and drug:polymer solution were based on the full factorial design described earlier in the manuscript.

2.4 Characterization of spinning solutions

Prior to electrospinning, the conductivity (measured by a conductivity meter, 8301, AZ Instrument Corp., Taiwan) and viscosity (measured by Brookfield viscometer, R?S plus, Germany) of the prepared solutions were measured. The rheometer was equipped with a cone/plate accessory (spindle type CC3-25) which was used at a constant shear rate of 100 sec⁻¹ to measure the viscosity of the solutions at room temperature.

2.5 Electrospinning process

Electrospinning solutions containing different ratios of drug:polymers were loaded in 10 ml syringes. The feeding rate was controlled by a syringe pump (Cole-Pham®, USA) and was fixed at 2.0 mL/h. A high voltage supply fixed at 10-18 kV was applied, and a piece of aluminum foil was used to collect the ultrafine fibers with a horizontal distance of 15 cm from the needle tip. Electrospun nanofibers were collected and stored in a desiccator for further studies.
2.6 Scanning electron microscopy (SEM)

The surface morphologies of electrospun nanofibers were assessed using a scanning electron microscope (LEO-rp-1455). The samples were previously silver sputter-coated under argon to render them electrically conductive. The pictures were then taken at an excitation voltage of 15 kV.

2.7 *In vitro* dissolution studies

Dissolution studies were carried out by introducing a certain amount of each nanofiber formulation equivalent to 10 mg of indomethacin in the dissolution baskets (DT800, Erweka, Germany; USP apparatus I) at 37°C with a rotation speed of 100 rpm (100 mL of dissolution medium; n=3). The method was modified in a way that a smaller flask (500 ml) was set inside the main dissolution flasks, so that, the baskets could reach the bottom of the vessels containing 100 ml of the dissolution media. Dissolution tests were performed in 0.1 N HCl at pH 1.2 for 4 h. The dissolution of samples was also performed at different pHs (6.4, 6.8 and 7.4) for 10 h. At predetermined time intervals, aliquots of 1 mL were withdrawn for sampling and replaced by an equal volume of fresh dissolution medium to maintain a constant volume. Afterward, the sample solutions were assayed spectrophotometrically by a UV/Visible spectrophotometer (Biowave II, WPA, England) at a wavelength of 320 nm. A calibration curve was constructed and validated to measure the concentration of the samples. In case of high concentrations of the drug in the sample, dilution was made to reach the absorbance within the calibration curve set up.
2.8 Statistical analysis of data

The effects of the independent variables on each experimental response were modeled using a second-order polynomial equation:

\[ Y = C + b_1X_1 + b_2X_2 + b_3X_1^2 + b_4X_2^2 + b_5X_1X_2 \]  
Eq. 1

\( X_1 \) is the ratio of polymer 1:polymer 2 and \( X_2 \) is the ratio of drug:polymer. Models were simplified with a backward, stepwise linear regression technique. Only significant terms (\( P < 0.05 \)) were chosen for the final model. Modeling was performed using SPSS (version 15.0). Response surface plots and contour plots resulting from equations were obtained by Statgraphics XVI.

3. Results and discussion

3.1 Fiber morphology

Based on our preliminary experiments, ethanol was chosen as a solvent for dissolving both ERS and ES. Also, the selection of various ratios of polymers to the drug was according to preformulation studies published elsewhere [30].

Scanning electron micrographs of drug-loaded nanofibers F1-F3 (Figure 1) showed that the increase in drug level in the composition of the formulations modified the morphology of nanofibers. For instance, formulation F3 with the high level of the drug showed some beads in its SEM image (Figure 1c). The changes in the morphology of nanofibers at high drug concentration was supported by previous studies [31, 32]. It has been reported that these changes in the morphology could be due to a reduction in the polymer solution viscosity following the addition of drug. This reduction can, in turn, increase the surface tension which leads to the morphology changes and the appearance of beads [33-34]. However, in the present study, the viscosity of spinning solutions did not show a significant decrease in viscosity when the level of drug was increased (Table 2). This may be due to having relatively low viscosity for the solution as a result of the low concentration of polymers used
in the formulations. Meanwhile, according to the results of the conductivity of the solutions (Table 2), the conductivity was lower in the solutions containing high amounts of the drug. The solution conductivity is a main effective parameter in the electrospinning method. In fact, repulsion of the charges on the surface of the droplet of the polymer solution is a key factor in stretching solution and the production of a uniform electrospun nanofiber. Therefore, higher solution conductivity creates more charge and subsequently bead-free uniform nanofibers [35]. This could be the main reason for the formation of more uniform fibers at a low concentration of the drug in the formulations compared to when a high concentration of indomethacin used.

According to SEM results (Fig. 1) formulation F1 produced nanofibers with some beads despite the highest degree of conductivity. This could be attributed to the high viscosity of this formulation compared with the other solutions (Table 2). When the viscosity of the solution gets high the formation of suitable nanofibers with acceptable characteristics is difficult. In another study focused on the effect of physical characteristics of poly (vinyl alcohol) solutions on nanofiber formation, Rosic et al., revealed that the effect of viscosity on the fiber structure and the shape of beads is relative and above a specific grade of viscosity, the effect would be inverted [36]. Nanofibers containing ES:ERS with the ratio of 60:40 (F4-F6) showed a uniform and smooth morphology in all ratios of drug:polymer (Fig. 1). This finding confirmed that a combination of ES and ERS in this ratio would be optimum for the preparation of favorable electrospun nanofibers.

### 3.2 In vitro release study

In vitro dissolutions were carried out in dissolution media with pH 1.2, 6.4, 6.8 and 7.4 to mimic various parts of the gastrointestinal tract. The drug release profiles of nanofibers in the
acid medium with pH 1.2 are shown in Figure 2A. As can be seen from Figure 2A, the drug release rate in the acid medium was very slow, with no more than 5% of the incorporated drug released from all the formulations within the first 2 h. Of all the electrospun nanofibers, formulations containing ES exhibited more resistance to drug release in the acidic media. For example, F7 containing 100% ES and the drug:polymer ratio of 3:5 showed the least drug release (less than 1% within 2 h) which can be ignored. It was also interesting to note that, although, in formulations F1-F3 the amount of ES was lower than the other nanofibers (1:4 relative to ERS) they still showed a low amount of drug released in acidic condition (less than 5% drug release). In fact, the presence of ES in the nanofiber structure even at the low levels caused drug protection in the simulated gastric medium. This finding was in agreement with the other investigations [37, 38] in which ES prevented drug release in acidic pH from electrospun nanofibers comprising pantoprazole and budesonide. In another study working on 5-fluorouracil (5-FU) loaded fibers with ES shell, a rapid release in the acidic medium was reported and explained by the high acid solubility of the drug and also the low molecular weight of 5-FU which enabled drug diffusion through the pores in the polymer coating [39]. Meanwhile, as pKa of indomethacin is 4.5, therefore, it is expected that its solubility should be low in acidic condition. On the basis of this fact, the protective effect of ES at pH 1.2 was the main factor for small drug release.

Drug release from all the formulations in the buffer medium (pH 6.4) was higher than the release in the acid medium (Figure 2B). This may be partly due to the better solubility of indomethacin at pH 6.4. Meanwhile, according to Figure 2B, formulations F8 and F9 showed unexpected highest drug release among the nanofibers despite the presence of ES in their compositions. The result showed that the presence of ERS in the structure of nanofiber is necessary to achieve a sustained drug release profile. Compositions of F8 and F9 include ES and drug in the ratios of 5:4.5 and 5:6, respectively. In this regard, the matrix formation of
pH-dependent polymer is incomplete and subsequently higher drug release from electrospun nanofibers will be expected compared to the nanofibers comprising ERS. This could be the reason for smaller drug release from F7 (the lower level of indomethacin in the formulation) compared to the two mentioned formulations. Moreover, as our previous investigation demonstrated, the inclusion of the drug in the nanofiber structure especially in formulations containing ERS caused a reduction in glass transition temperature of Eudragits [30] which in turn could complete the formation of polymeric matrix. According to Figure 2B, F4 comprising ES:ERS in the ratio of 60:40 and the high ratio of polymer:drug exhibited the lowest drug release at pH 6.4.

The in vitro release studies at pH 6.8 were approximately the same as the release at pH 6.4 (Figure 3A). As shown in Figure 3A, the highest drug release belonged to formulations containing ES alone (especially F8 and F9) whereas the addition of ERS up to 40% diminished the drug release at pH 6.8 in such a way that nanofibers of formulation F4 released the drug in the slowest manner.

Meanwhile, the drug release from formulations at pH 7.4 was dependent on the presence of ES (Figure 3B). As can be seen in Figure 3B, regardless of the amount of drug, fibers containing higher levels of ES exhibited faster drug release at pH 7.4.

Figure 4 depicts response surface plots for all the responses. Accordingly, drug release at pH 6.4 as well as pH 6.8 was dependent on the ratio of ES:ERS. Up to a specific ratio of ES:ERS (60% at pH 6.4 and 40% at pH 6.8) the increase in the level of ES resulted in a decrease in the drug release. However, the higher amounts of ES in the formulations enhanced drug release from nanofibers. This could be due to the presence of a more uniform structure of nanofibers containing higher levels of ERS which confirmed by SEM results (Fig. 1) and it may be the outcome of the complete polymeric matrix formation in the specific proportions of the two types of Eudragits, as mentioned before. Regarding the pH-independency of ERS
and low solubility of ES at pH values less than 7.0, the formation of matrix chains prevented a quick movement of the dissolution medium through the polymeric chains and thereupon delayed drug release from electrospun fibers. Nevertheless, considering a complete dissolution of ES at pH 7.4, the drug release was entirely dependent on the amount of ES in the formulation and with the increase of ES:ERS ratio, the release of indomethacin from nanofibers was amplified (Fig. 6).

3.3 Kinetics of drug release from nanofibers

The mechanism of release of indomethacin from fibers was evaluated by analyzing the drug release profiles using the zero-order, Higuchi, and Peppas models. According to Table S1 (see supplementary materials), the drug release at pH 6.4 was fitted well to Higuchi model ($R^2 > 0.955$ for all formulations). Therefore, the drug release occurred via diffusion-controlled mechanism at pH 6.4 [40, 41]. This can be explained by the insoluble characteristics of Eudragits at pH 6.4. At pH 6.8, the mechanism of drug release from fibers containing ERS except F4 was also fitted well to Higuchi model. However, the best fit for drug release from formulations F4, F7, F8 and F9 was found to be Peppas model ($Q = kt^n$) with the diffusion exponent $n$ equal to 0.9842, 0.5019, 0.8305, and 0.8416, respectively. Hence, the values of $n$ for these nanofibers fell between 0.5 and 1, indicating that non-Fickian diffusion mechanism may control the drug release i.e. release from the nanofibers was controlled via a combination of diffusion and erosion mechanisms [42]. Taking into account the presence of ES in the structure of mentioned fibers and the higher value of pH, some parts of the polymeric matrix could be erodible and therefore, the mechanism of erosion could also contribute to the drug release. Analyzing the drug release profiles at pH 7.4 verified this fact. Accordingly, the drug release from all the nanofibers at pH 7.4 was fitted to Peppas model. The value of $n$ for all the formulations was more than 0.5 except F1 and F2 which their $n$
values were 0.4104 and 0.3308, respectively, indicating that diffusion was a prominent mechanism of drug release from the two latter nanofibers.

3.4 Optimization

Mathematical relationships were generated between the dependent and independent variables using the statistical package SPSS. The resulted second order polynomial equations for all of the responses are given below:

\[ Y_1 = 4.562 - 0.0003X_1X_1 \quad \text{Eq. 2} \]
\[ Y_2 = -11.352 - 0.858X_1 + 93.69X_2 + 0.004X_1X_1 - 60.432X_2X_2 + 0.499X_1X_2 \quad \text{Eq. 3} \]
\[ Y_3 = -58.353 - 2.245X_1 + 303.014X_2 + 0.019X_1X_1 - 181.481X_2X_2 + 0.681X_1X_2 \quad \text{Eq. 4} \]
\[ Y_4 = -76.871 + 0.648X_1 + 272.986X_2 - 0.005X_1X_1 - 171.914X_2X_2 + 0.705X_1X_2 \quad \text{Eq. 5} \]

The three-dimensional response surface plots and contour plots resulted from the above equations were drawn to find the best area for optimum formulation.

Optimization is one of the main objectives of using an experimental design for the production of formulations. To achieve a suitable optimization process the selection of the effective responses is the essential parameter. Since pH and the residence time of different parts of GI tract are substantial factors for the design of a colonic delivery system, they should be regarded as the main responses. All of the manufactured nanofibers showed minimum drug release and therefore optimum characteristics in the acid media. According to the relative constant residence time of materials in the small intestine [43] and the pH gradient of this part of GI tract, the residence time for dissolution media with pH 6.4, pH 6.8 and pH 7.4 were considered 1, 2 and 2 h, respectively. Drug release less than 5%, 10% and 20% was considered as optimum release at pH 1.2, 6.4 and 6.8, respectively. On the other hand, taking into account the fact that a colon delivery system based on pH- and time-dependent polymers
should release majority of the drug at the terminal ileum, and the authors believed that 2/3 of drug release which is around 65% could be as an indication of a good drug release in that region. It also ensures that the drug release is occurring. Therefore, the drug release more than 65% was considered as the best constraint at pH 7.4.

According to contour plots (Fig. S1) formulation containing ES:ERS (60:40) and drug:polymer ratio of 3:5 (F4) theoretically met the criteria for all the optimum responses. In order to check the validity of the process of optimization, a new batch of nanofibers with the predicted levels of independent variables was prepared and examined. The results in Table S2 showed that the observed responses were close to the predicted responses, and therefore the validation of experimental design for predicting the optimum formulation was confirmed. Therefore, as an overall consequence, nanofibers containing ES:ERS (60:40) and drug:polymer ratio of 3:5 exhibited the optimum drug release at various parts of GI tract with different pHs. This kind of nanofibers have the potential to release the major contents of the drug in the proximal colon.

Although, the scale up of the electrospinning technology might be a challenge for the pharmaceutical industry because of the low production rate of fibers [44], therefore, various attempts have been made to enhance the productivity of this technique. Recently, different processes such as multiple-jet electrospinning [45, 46], high speed electrospinning [47], edge electrospinning [48] and needleless electrospinning [49] have been developed. Also, downstream processing of nanofibers to produce tablet formulations from electrospun solid dispersions has been investigated [50]. The present study aimed to optimize the formulation to improve the potential for this technology to be scaled up for commercialization purpose.
4. Conclusion

According to the results of this study, electrospinning was an appropriate method for producing nanofibers of indomethacin aimed for colonic drug delivery. It was shown that selecting an appropriate experimental design and optimization technique can be successfully applied to the development of nanofibers based on Eudragit polymers to achieve colon drug delivery. According to optimization process, a formulation containing ES:ERS (60:40) and drug:polymer ratio of 3:5 exhibited suitable morphological characteristics and protected a major part of drug in the media simulating upper GI tract and therefore could be good excipients as a colonic drug delivery system for indomethacin.

5. Acknowledgments

This work was supported by a grant from research chancellor of Ahvaz Jundishapur University of Medical Sciences. The authors would like to thank Darupakhsh pharmaceutical co. for their collaboration and providing samples used in this paper.
References


Figure 1. SEM images of formulations; (a) F1, (b) F2, (c) F3, (d) F4, (e) F5, (f) F6, (g) F7, (h) F8, and (i) F9.
Figure 2. Dissolution profiles of formulations at (A) pH 1.2 and (B) pH 6.4 (n=3; error bars are standard deviation).
Figure 3. Dissolution profiles of formulations at (A) pH 6.8 and (B) at pH 7.4 (n=3; error bars are standard deviation).
Figure 4. Response surface plots: (a) $Y_2$, (b) $Y_3$, and (c) $Y_4$. 
Table 1. Composition of experimental formulations

<table>
<thead>
<tr>
<th>Variable factors</th>
<th>Components of nanofibers</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>X₁ (ES:ERS ratio)</td>
</tr>
<tr>
<td>F1</td>
<td>20:80</td>
</tr>
<tr>
<td>F2</td>
<td>20:80</td>
</tr>
<tr>
<td>F3</td>
<td>20:80</td>
</tr>
<tr>
<td>F4</td>
<td>60:40</td>
</tr>
<tr>
<td>F5</td>
<td>60:40</td>
</tr>
<tr>
<td>F6</td>
<td>60:40</td>
</tr>
<tr>
<td>F7</td>
<td>100:0</td>
</tr>
<tr>
<td>F8</td>
<td>100:0</td>
</tr>
<tr>
<td>F9</td>
<td>100:0</td>
</tr>
</tbody>
</table>
Table 2. Conductivity and viscosity of spinning solutions used for preparation of formulations (values are the mean and standard deviation of 3 determinations)

<table>
<thead>
<tr>
<th>Solutions</th>
<th>% ES (w/v)</th>
<th>% ERS (w/v)</th>
<th>% drug (w/v)</th>
<th>Conductivity (µs cm(^{-1}))</th>
<th>Viscosity (cP)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>0.25</td>
<td>1.00</td>
<td>0.75</td>
<td>44.1±0.2</td>
<td>27.2±0.1</td>
</tr>
<tr>
<td>F2</td>
<td>0.21</td>
<td>0.85</td>
<td>0.94</td>
<td>30.8±0.3</td>
<td>24.8±0.1</td>
</tr>
<tr>
<td>F3</td>
<td>0.18</td>
<td>0.73</td>
<td>1.09</td>
<td>26.4±0.2</td>
<td>25.1±0.0</td>
</tr>
<tr>
<td>F4</td>
<td>0.75</td>
<td>0.50</td>
<td>0.75</td>
<td>29.1±0.1</td>
<td>24.9±0.2</td>
</tr>
<tr>
<td>F5</td>
<td>0.64</td>
<td>0.42</td>
<td>0.94</td>
<td>23.5±0.1</td>
<td>25.4±0.0</td>
</tr>
<tr>
<td>F6</td>
<td>0.55</td>
<td>0.36</td>
<td>1.09</td>
<td>18.5±0.0</td>
<td>24.7±0.1</td>
</tr>
<tr>
<td>F7</td>
<td>1.25</td>
<td>0.00</td>
<td>0.75</td>
<td>14.0±0.1</td>
<td>25.3±0.1</td>
</tr>
<tr>
<td>F8</td>
<td>1.06</td>
<td>0.00</td>
<td>0.94</td>
<td>11.3±0.0</td>
<td>24.5±0.0</td>
</tr>
<tr>
<td>F9</td>
<td>0.91</td>
<td>0.00</td>
<td>1.09</td>
<td>11.0±0.1</td>
<td>24.8±0.0</td>
</tr>
</tbody>
</table>