

## Effect of Molecular Weight of Poly(Ethylene Glycol) Dicarboxylate on the Properties of Cross-Linked Hydrogel Film as an Anti-Adhesion Barrier

Haryanto, N. Aries Nurulita, E. Sundhani, P. Ho Huh & S. Cheol Kim

To cite this article: Haryanto, N. Aries Nurulita, E. Sundhani, P. Ho Huh & S. Cheol Kim (2017): Effect of Molecular Weight of Poly(Ethylene Glycol) Dicarboxylate on the Properties of Cross-Linked Hydrogel Film as an Anti-Adhesion Barrier, Polymer-Plastics Technology and Engineering, DOI: [10.1080/03602559.2017.1381254](https://doi.org/10.1080/03602559.2017.1381254)

To link to this article: <http://dx.doi.org/10.1080/03602559.2017.1381254>



Accepted author version posted online: 21 Sep 2017.



Submit your article to this journal [↗](#)



View related articles [↗](#)



View Crossmark data [↗](#)

# **Effect of molecular weight of poly(ethylene glycol) dicarboxylate on the properties of cross-linked hydrogel film as an anti-adhesion barrier**

Haryanto

Department of Chemical Engineering, Universitas Muhammadiyah Purwokerto, Central Java,  
Indonesia

N. Aries Nurulita

Department of Pharmacy, Universitas Muhammadiyah Purwokerto, Central Java, Indonesia

E. Sundhani

Department of Pharmacy, Universitas Muhammadiyah Purwokerto, Central Java, Indonesia

P. Ho Huh

Department of Polymer Science and Engineering, Pusan National University, Busan, Republic of  
Korea

S. Cheol Kim

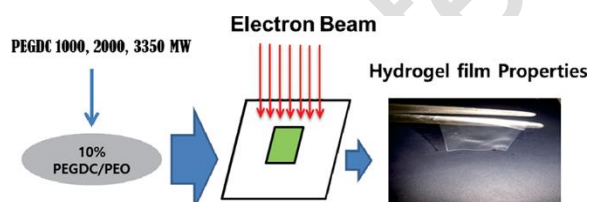
Department of Advanced Organic Materials Engineering, Yeungnam University, Gyongbuk,  
Republic of Korea

Address correspondence to Haryanto, Department of Chemical Engineering, Universitas Muhammadiyah Purwokerto, Central Java 53182, Indonesia. Tele: +62-8121531236. E-mail: harymsl@gmail.com

## ABSTRACT

Poly(ethylene glycol) dicarboxylate (PEGDC)/poly(ethylene oxide) (PEO) cross-linked hydrogel films were developed as an anti-adhesion barrier using an e-beam. The effects of the molecular weight of PEGDC on hydrogel properties were investigated. The decrease in molecular weight of PEGDC increased the gel fraction, and tissue adhesion, whereas the mechanical strength did not change considerably. On the other hand, the swelling ratio decreased rapidly with decreasing molecular weight of PEGDC. The cytotoxicity of PEGDC (2000 or 3000) was low, whereas that of PEGDC (1000) was higher. In animal studies, all hydrogels showed a better anti-adhesive effect compared to the control.

## GRAPHICAL ABSTRACT



**KEYWORDS:** carboxyl content, crosslinked hydrogel, electron beam, molecular weight, poly(ethylene glycol) dicarboxylate, tissue adhesion

## ARTICLE HISTORY

Received 20 July 2017

Accepted 14 September 2017.

# 1. Introduction

Adhesion occurs in almost all open abdominal surgical procedures. The most common site for adhesion after pelvic gynecological surgeries is the ovary.<sup>[1]</sup> Adhesions can result in critical complications, including small bowel obstruction, infertility, problems with next operations, and chronic debilitating pain.<sup>[2,3]</sup> Interceed, Sepra film, and Adept products have been approved and are now available for clinical use in the United States. Interceed is made of oxidized regenerated cellulose. Sepra film is a blend of modified hyaluronic acid and carboxy methyl cellulose. Adept is produced from an icodextrin solution that can be dispersed easily through out the abdomino pelvic cavity. All commercial anti adhesion products have different efficacies and do not fully satisfy the requirements for clinical implementation.<sup>[4]</sup>

A good adhesion-prevention product should be resolvable, non-reactive, easy to apply, and capable of being fixed. A previous study reported that polyethylene glycol dicarboxylate/ polyethylene oxide crosslinked hydrogel film have a good anti-adhesive effect compared to Guardix-SG<sup>®</sup> in an animal study.<sup>[5]</sup> The present paper describes the effects of the molecular weight of PEGDC on hydrogel films as an anti-adhesion barrier.

Hydrogels are three-dimensional network structures that absorb and retain a large amount of water. Crosslinks must be present in a hydrogel to prevent dissolution of the hydrophilic polymer chains in an aqueous environment.<sup>[6]</sup> A hydrogel of many synthetic and natural polymers was produced with their end use mainly in pharmaceutical, and biomedical fields.<sup>[7]</sup> Owing to their high water absorption capacity and biocompatibility, they have been used in many medical applications, such as contact lenses,<sup>[8]</sup> wound dressings,<sup>[9]</sup> dental materials,<sup>[10]</sup> tissue engineering,<sup>[11]</sup> drug delivery,<sup>[12]</sup> and anti-adhesion barriers.<sup>[13,14]</sup>

Poly(ethylene oxide) (PEO) has been used in a range of biomedical applications. The material has several advantageous properties as a biomaterials, such as low toxicity, biocompatible, water soluble polymer, antithrombogenic polymer, and nonionic polymer, but it lacks contact with the tissue.<sup>[15,16]</sup> Poly(ethylene glycol) dicarboxylate (PEGDC) contains carboxyl groups that increase the adhesive strength of the film to the tissue.<sup>[17,18]</sup> A lower molecular weight of PEGDC might improve the mechanical strength and physical properties of hydrogel film due to the higher physical crosslink density. Furthermore, a lower molecular weight of PEGDC is expected to increase the adhesion of a hydrogel film on the desired site to ensure the prevention of a post-operative adhesion.

This study examined the effects of the molecular weight of PEGDC on the gel fraction, swelling ratio, crosslinking density, mechanical strength, tissue adhesion, and cytotoxicity of hydrogel films was investigated. The animal study was evaluated to examine the effects of the molecular weight of PEGDC on the postoperative adhesion formation in a rat model.

## **2. Experimental**

### **2.1. Materials**

Poly(ethylene oxide) (PEO) ( $M_n 6 \times 10^5$ ), poly(ethylene glycol) (PEG) ( $M_n 3350, 2000, 1000$ ), maleic anhydride ( $M_w 98.06$ ), Dulbecco's modified Eagle's medium (DMEM), 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT), fetal bovine serum, and penicillin-streptomycin were purchased from Sigma-Aldrich, St. Louis, USA. Mouse fibroblast cells (NIH3T3) were purchased from ATCC (Manassas, VA, USA) and the cytotoxicity kits were purchased from Life Technology, Invitrogen, USA.

## 2.2. *Synthesis of PEGDC*

Firstly, 6.7 g sample of PEG 3350 was dissolved in 50 ml toluene in a 2 necked flask. The solution was stirred for 30 minutes.<sup>[5]</sup> The stirred solution was dried at 130 °C by purging nitrogen for 1 hour. After cooling, 0.392 g of maleic anhydride was added and the reaction mixture was stirred for 18 hours at 85 °C under an N<sub>2</sub> atmosphere. The toluene was then removed by vacuum evaporation at 85 °C under reduced pressure. The crude product was dissolved in a small amount of methylene chloride and precipitated three times into diethyl ether to remove the unreacted maleic anhydride. The precipitate was then dried in an oven at 50 °C for 6 hours. The same procedure was followed using a different molecular weight of PEG (2000 and 1000) in the same mole composition.<sup>[19,20]</sup> For the next PEGDC, PEG 1000,2000 and 3350 are referred to as PEGDC (1000),(2000), and (3350), respectively. Selected IR data of Poly(ethylene glycol) dicarboxylate were as follows.

PEGDC (1000), IR (KBr):3479, 2883, 1727, 1448, 1371, 1105 cm<sup>-1</sup>.

PEGDC (2000), IR (KBr): 3500, 2863, 1720, 1625, 1460, 1359, 1073 cm<sup>-1</sup>.

PEGDC (3350), IR (KBr): 3482, 2862, 1728, 1626, 1468, 1385,1131 cm<sup>-1</sup>.

## 2.3. *Preparation of the hydrogel films*

PEO and PEGDC (with various molecular weight) were used to prepare the hydrogel films. A 5% aqueous solution of PEGDC/PEO (w/w) was prepared by stirring the solution at room temperature for 24 hours with a 10/100 (w/w) composition of PEGDC/PEO. The calculated amount of aqueous solution was poured into a petri dish for the formation of a dry PEGDC/PEO film with a thickness of 0.1 mm. The PEGDC/PEO solution was dried in an oven at 55 °C for 24

hours, and the remaining moisture was then removed from the vacuum oven for 6 hours at the same temperature. Dry PEGDC/PEO films were sealed in an evacuated polyethylene bag, followed by irradiation at 300 kGy using an EB with beam currents of 5 mA and 10 mA, and a voltage of 0.7 MeV.

#### **2.4. Cytotoxicity test of PEGDC**

A MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) assay was applied to check the metabolic activity of the cells on the PEGDC/PEO films and control-2D. The NIH3T3 cells of a mouse fibroblast cell line were grown in DMEM (Invitrogen, Carlsbad, CA) that contained 10% fetal bovine serum (FBS) (Invitrogen, Carlsbad, CA) and 1% penicillin–streptomycin (Invitrogen, Carlsbad, CA) at 37 °C in a humidified atmosphere containing 5% CO<sub>2</sub>. The cells (1 x 10<sup>4</sup>) were seeded overnight on a 24-well cell culture plate and incubated for an additional 1 day, 3 days, 5 days, and 7 days. Briefly, 500 µL of a MTT solution (5 mg/ml, Sigma Chemicals) was added to each well, which was followed by incubation for 4 h at 37 °C. The supernatant was aspirated, and the MTT-formazan crystals produced by the metabolically viable cells were dissolved in 1500 µl of DMSO. The absorbance was measured at a wavelength of 595 nm using an ELISA reader.<sup>[21]</sup>

#### **2.5. Determination of gel fraction**

Immediately after irradiation, the sealed film was opened and the weight of the dry film (2 cm x 2 cm) was measured accurately. The crosslinked hydrogel film was then placed in distilled water at 50 °C for 24 hours to remove the uncrosslinked soluble parts. The obtained insoluble gel was dried in an oven at 50 °C for 24 hours and then in a vacuum oven at 50 °C for 6 hours to obtain

a constant dry weight. The gel fraction was determined from the weight ratio between the weight of an insoluble dry gel and the initial weight of a dry film using the following formula:

$$\text{Gel fraction} = (W_c / W_o) \times 100 [\%]$$

where  $W_c$  and  $W_o$  are the weights of the insoluble dry gel and the initial weight of a dry film.<sup>[22]</sup>

## **2.6. Determination of swelling ratio**

The swelling ratio was determined by submerging the hydrogel film (1.5 x 1.5 cm) in a buffered solution at room temperature until the hydrogel reached the equilibrium weight. The weight of the equilibrium swollen gel was measured after removing the excess surface water using filter papers. The equilibrium swelling ratio ( $q$ ) was calculated using the following equation:

$$q = (W_t / W_o) \times 100 [\%]$$

where  $W_t$  and  $W_o$  are the weight of the swollen gel at equilibrium and that of the dry film, respectively.<sup>[23]</sup>

## **2.7. Mechanical properties**

The tensile strength and elongation at break of the crosslinked hydrogel films were measured using a tensile test machine (Instron 2710-105, USA) at a constant extension rate of 10 mm/min at room temperature.

## **2.8. Tissue adhesion**

A tissue adhesion test was performed using a universal tensile machine. A test strip of hydrogel film, measuring one inch in width and 7 inches in length, was prepared, bent back on

itself, and joined together with a half inch strip of masking tape. The specimen formed a tear drop-shaped loop. The taped end of the specimen loop was inserted into the upper grips. The test fixture was placed in the lower grip of the tensile tester. The tensile tester was activated so that the crosshead moves downward until the specimen loop is in complete contact with one square inch of moist bovine intestine for 5 minutes. The maximum force required to remove the specimen loop from the bovine intestine was recorded as a measure of a film's adherence to the intestine.<sup>[24]</sup>

## **2.9. Animal study**

This study was performed with assistance from the LPPT Gadjah Mada university. The animal care and use committee of Gadjah Mada university approved the methods described. Forty Sprague Dawley rats, weighing 240 to 325 g, were used for the study. All rats were housed in a 12-h-day and 12-h-night environment at standard temperature (22°C). They were fed standard laboratory food and tap water ad libitum. All rats were anesthetized with ketamine hydrochloride and xylazine hydrochloride intramuscularly.

A midline laparotomy incision measuring 4 cm was produced and the cecum was identified. The ventral side of the cecum was then abraded by thermal cautery until the burn area reached approximately 2 cm<sup>2</sup> (1 x 2 cm). The rats in the first group did not received the film on the burned area. The rats in the second, third, and fourth group received a 6 cm<sup>2</sup> (2 x 3cm) PEGDC/PEO films barrier with different molecular weights of PEGDC (1000, 2000 and 3350 g/mol), which was placed between the burned area and the peritoneum. The laparotomy incision was closed with 4/0 black silk suture. All rats were allowed to receive tap water and food ad libitum one day later until the 21<sup>th</sup> day.

All animals were euthanized by carbon dioxide inhalation on the 21<sup>st</sup> day postoperatively and examined for adhesion formation by two surgeons in a double-blinded manner. The adhesion criteria were classified according Avital et al.'s study (**Table 1**).<sup>[25]</sup>

### **3. Results and Discussion**

#### **3.1. Cytotoxicity of PEGDC/PEO hydrogel films**

In the first treatment, the hydrogel film from PEGDC (1000) showed the significant inhibition of cell viability compared to control-2D. The cell density also appears lower than that without treatment. The cell metabolic activity in PEGDC (2000) and (3350) appeared better and did not inhibit the cell viability. The cytotoxicity test showed that the hydrogel films of PEGDC (2000) and PEGDC (3350) do not show toxicity, whereas PEGDC 1000 showed some toxicity, albeit low. The cells were found to be viable and metabolically more active on the PEGDC/PEO hydrogel film with a higher molecular weight of PEGDC and the cell metabolic activity on all hydrogel films was better than the control-2D, as shown in **Figure 1a,b**.

#### **3.2. Effect of the molecular weight of PEGDC on the gel fraction and swelling properties**

**Figure 2a** shows the gel fraction of the PEGDC/PEO hydrogel films. The results showed that the gel fraction increased with decreasing molecular weight of PEGDC because of the increase in crosslinking density due to the higher double bond content in the lower molecular weight of PEGDC. **Figure 2b** shows the swelling ratio of the PEGDC/PEO hydrogel film with various molecular weights of PEGDC as a function of the swelling time. The swelling ratio decreased with increasing molecular weight of PEGDC from 651% in the PEGDC (3350)/PEO hydrogel film to

509% in the PEGDC (1000)/PEO film. The results indicate that the lower molecular weight of PEGDC decreases the water absorption capacity of the PEO/PEGDC hydrogel due to the increasing crosslinking of PEGDC together with radicals in the PEO backbones via radical coupling reaction.

### 3.3. *Mechanical properties*

**Figure 3a** shows the tensile strength of wet hydrogel film as a function of the molecular weight of PEGDC; there was no significant difference in the tensile strength and elongation at break of each hydrogel (**Figure 3b**), particularly at higher PEGDC contents. This indicates that all hydrogel films have good mechanical strength with 10% PEGDC for biomedical applications, such as an anti-adhesion barrier.

### 3.4. *Tissue adhesion*

The adhesion strength of a hydrogel film is important to maintain the position of the film and cover up the wound site to ensure the wound healing process and minimize the adhesion with neighboring tissues. The tissue adhesiveness of a wet hydrogel film was determined by measuring the peak detachment force required to separate the film from the bovine intestine. As shown in **Figure 4**, the force recorded for the tissue adhesiveness reached the minimum value of  $72.33 \pm 4.93\text{cN}$  at PEGDC (3350). The higher molecular weight of PEGDC (2000) increased the tissue adhesiveness to  $86.67 \pm 10.96\text{cN}$ . The highest peak detachment force ( $116.33 \pm 17.11\text{cN}$ ) was obtained at PEGDC (1000). This shows that the number of carboxyl groups enhanced the adherence of a crosslinked film.

### 3.5. *Animal study*

Three types of hydrogel films, PEGDC (1000)/PEO film, PEGDC (2000)/PEO film, and PEGDC (3350)/PEO film, and a control without a film were used in the animal study. No wound infection, sepsis, and dietary deficiency that would adversely affect the results were observed.

**Table 2** list the grade of adhesion and the percentage of injured cecal surface area for all animals. The frequency of adhesion severity with the PEGDC group was less than those of the control, as shown in **Figure 5**. The grade of adhesion was assigned according to the former research.<sup>[25]</sup> The hydrogel with higher molecular weight of PEGDC showed better *in vivo* results. The degree of adhesion depended on the molecular weight of PEGDC. The better adsorption of the crosslinked films induced less adhesion of the tissues with the cecum. On the other hand, the PEGDC (1000)/PEO showed worse results than that of the PEGDC(3330)/PEO hydrogel films. The increased postoperative adhesion could be related to the cytotoxicity of PEGDC (1000), which has a shorter poly(ethylene glycol) chain that should hide the inflammatory response against the carboxyl groups of PEGDC. The minimum tissue adhesion of PEGDC(3330)/PEO suggests that the cytotoxicity of the hydrogel films is more important than the adhesion of a gel film on the surface of a wound.

#### 4. Conclusions

This study examined the influence of the molecular weight of PEDC on the physical properties of hydrogel films and the anti-adhesive effect of hydrogel film in rat model as a continuation of previous work. The decrease in molecular weight of PEGDC increased the gel fraction of the hydrogel film significantly due to the increased number of double bonds in the 10% PEGDC/PEO hydrogels. The molecular weight had no significant effect on the tensile strength and elongation at break. The tissue adhesion increased considerably with decreasing molecular

weight of PEGDC probably due to the decreased cell compatibility with the MW of PEGDC. While the swelling ratio decreased rapidly with decreasing molecular weight of PEGDC. The cytotoxicity of the hydrogel films increased with decreasing MW of PEGDC. As a result, the anti-adhesive effect of the hydrogel films with PEGDC (3350)/PEO was better than the other gel films, even though the gel film has a weaker interaction with the cecum surface.

### Acknowledgements

This study was supported by the Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Science and Technology (20110008342).

### References

- [1] Diamond, M. P.; Linsky, C. B.; Cunningham, T. Adhesion Reformation: Reduction by the Use of Interceed (TC7) Plus Heparin. *J. Gynecol. Surg.* **1991**, *7*(1), 71–6.
- [2] Coleman, M. G.; McLain, A. D.; Moran, B. J. Impact of Previous Surgery on Time Taken for Incision and Division of Adhesions During Laparotomy. *Dis. Colon. Rectum.* **2000**, *43*, 1297–1299.
- [3] Van Der Krabben, A. A.; Dijkstra, F. R.; Nieuwenhuijzen, M.; Reijnen, M. M.; Schaapveld, M.; Van Goor, H. Morbidity and Mortality of Inadvertent Enterotomy During Adhesiotomy. *Br. J. Surg.* **2000**, *87*, 467–471.
- [4] Rajab, T. K.; Wallwiener, M.; Planck, C.; Brochhausen, C.; Kraemer, B.; Wallwiener, C. W. A Direct Comparison of Seprafilm, Adept, Intercoat, and Spray Gel for Adhesion Prophylaxis. *J. Surg. Res.* **2010**, *161*, 246–249.
- [5] Haryanto, Deepti, S.; Han, S. S.; Son, J. H.; Kim, S. C. Poly(Ethylene Glycol) Dicarboxylate/Poly(Ethylene Oxide) Hydrogel Film Co-Crosslinked by Electron Beam Irradiation as an Anti-Adhesion Barrier. *Mater. Sci. Eng. C* **2015**, *46*, 195–201.
- [6] Hoffman, A. S. Hydrogels for Biomedical Applications. *Adv. Drug Delivery Rev.* **2002**, *54*, 3–12.
- [7] Hoare, T. R.; Kohane, D. S. Hydrogels in Drug Delivery: Progress and Challenges. *Polymer* **2008**, *49*, 1993–2007.
- [8] Guillon, M. *Eye & Contact Lens.* **2013**, *39*, 86–92.

- [9] Haryanto, Kim, S. C.; Kim, J. H.; Kim, J. O.; Ku, S. K.; Cho, H.; Han, D. H.; Huh, P. Fabrication of Poly(Ethylene Oxide) Hydrogels for Wound Dressing Application Using e-Beam. *Macromol. Res.* **2014**, *22*, 131–138.
- [10] Cavalcantia, B. N.; Zeitlin, B. D.; Nör, J. E. A Hydrogel Scaffold that Maintains Viability and Supports Differentiation of Dental Pulp Stem Cells. *Dental Mater.* **2013**, *29*, 97–102.
- [11] Kuen, Y. L.; Mooney, D. J. Hydrogels for Tissue Engineering. *Chem. Rev.* **2001**, *101*(7), 1871.
- [12] Hoare, T. R.; Kohane, D. S. Hydrogels in Drug Delivery: Progress and Challenge. *Polymer* **2008**, *49*, 1993–2007.
- [13] Bing, Y.; Chang, Y. G.; Zhi, Y. Q.; Xia, Z.; Zheng, Y. L.; Xiao, R. Q.; Sheng, T. Z.; Qian Zhong, F. L.; Yu, Q. W. Prevention of post-Surgical Abdominal Adhesions by a Novel Biodegradable Thermosensitive PECE Hydrogel. *BMC Biotechnol.* **2010**, *10*, 65.
- [14] Ten Broek, R. P. G.; Kok-Krant, N.; Verhoeve, H. R.; Van Goor, H.; Bakkum, E. A. Efficacy of Polyethylene Glycol Adhesion Barrier After Gynecological Laparoscopic Surgery. *Gynecol. Surg.* **2012**, *92*, 9–35.
- [15] Zhang, Z.; Ni, J.; Chen, L.; Xu, J.; Ding, J. Biodegradable and Thermoreversible PCLA–PEG–PCLA Hydrogel as a Barrier for Prevention of Post-Operative Adhesion. *Biomaterials* **2011**, *32*, 4725–4736.
- [16] Torchilin, V. P. Polymeric Nanomaterials. In: *Nanomaterials for the Life Sciences*; C. S. S. R. Kumar, Ed.; 2011; Vol. 10, p 124.
- [17] Soltz, B. A.; DeVore, D. P.; DeVore, B. P.; Soltz, R.; Soltz, M. A. Composite Tissue Adhesive. US Patent 6939364B1, 2005.
- [18] Liu, L. S.; Berg, R. A. Adhesion Barriers of Carboxymethylcellulose and Polyethylene Oxide Composite Gels. *J. Biomed. Mater. Res.* **2002**, *63*, 326–332.
- [19] Bai, Y. Use of Carboxylated Polyethylene Glycol as Promoter for Platinum-Catalyzed Hydrosilylation of Alkenes. *Appl. Organomet. Chem.* **2011**, *25*, 400–405.
- [20] Haryanto, Deepti, S.; Han, S. S.; Son, J. H.; Kim, S. C. Poly(Ethylene Glycol) Dicarboxylate/Poly(Ethylene Oxide) Hydrogel Film Co-Crosslinked by Electron Beam Irradiation as an Anti-Adhesion Barrier. *Mater. Sci. Eng. C* **2015**, *46*, 195–201.
- [21] Way, T. D.; Hsieh, S. R.; Chang, C. J.; Hung, T. W.; Chiu, C. H. Preparation and characterization of branched polymers as postoperative anti-adhesion barriers. *Appl. Surf. Sci.* **2010**, *256*, 3330–3336.
- [22] Wu, M.; Bao, B.; Yoshii, F.; Makuuchi, K. Irradiation of Crosslinked, Poly (Vinyl Alcohol) Blended Hydrogel for Wound Dressing. *J. Radioanal. Nucl. Chem.* **2011**, *250*, 391–395.
- [23] Haryanto, Deepti, S.; Huh, P. H.; Kim, S. C. *Biomed. Mater. Res. Part A* **2015**, 1–9.
- [24] *Standard Test Methods for Loop Tack*; ASTM Standard D6195–03; ASTM International: West Conshohocken, 2011.

[25] Avital, S.; Bollinger, T. J.; Wilkinson J. D.; Marchetti, F.; Hellinger, M.; Sands, L. R. Preventing Intra-Abdominal Adhesions with Polylactic Acid Film: An Animal Study. *Dis. Colon Rectum*. **2005**, *48*, 153–157.

Accepted Manuscript

**Table 1.** Adhesion severity based on the adhesion grade and percent of injured cecal surface area.

Severity	Grade	% area involved
Absent	0	0
Moderate	1	1~100
	2	1~100
Severe	3	<100
	3	100

Grade

0 = no adhesions; 1 = thin filamentous, easily separated adhesions; 2 = thick adhesions, difficult to dissect, do not tear organ when separated; 3 = thick adhesions, not dissectible, tears organ when separated.

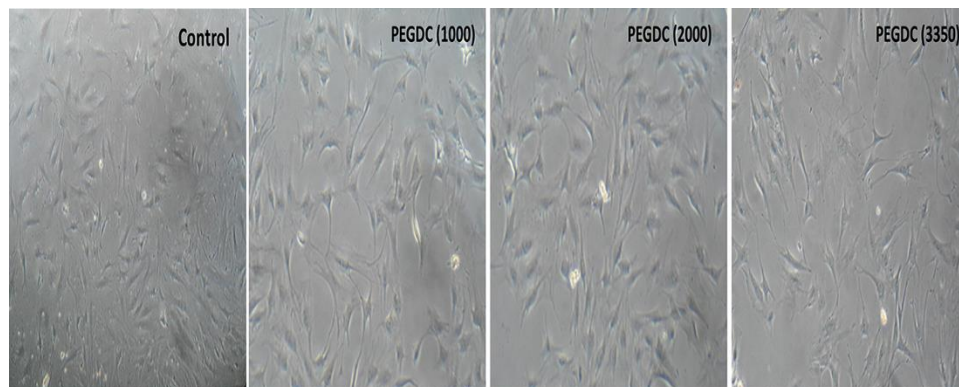
Accepted Manuscript

**Table 2.** The grade of adhesion and the percentage of injured cecal surface area.

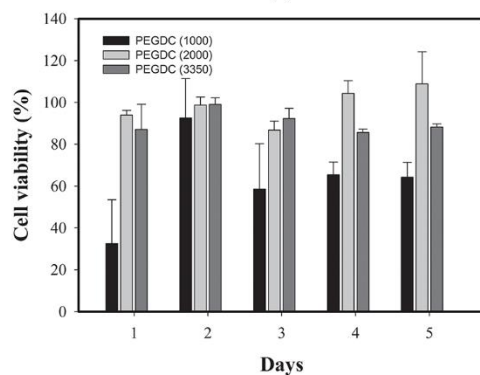
Rat		1	2	3	4	5	6	7	8	9	10
PEGDC- (1000)	Grade	2	2	2	2	2	2	2	0	0	1
	Area	3	4	3	3	3	3	3	0	0	2
PEGDC- (2000)	Grade	0	2	2	3	0	2	2	2	0	2
	area	0	3	3	4	0	3	3	3	0	3
PEGDC- (3350)	grade	0	1	2	1	2	2	0	0	0	1
	area	0	2	3	2	3	3	0	0	0	2
Control 10%	grade	0	0	3	3	2	2	2	3	2	3
PEGDC	area	0	0	4	3	4	3	2	4	4	4

Accepted Manuscript

**Figure 1.** (a). NIH3T3 cells after the 24 hours PEGDC/PEO hydrogel treatment. (b). Cell viability of the PEGDC/PEO hydrogels after the 24 hours treatment.

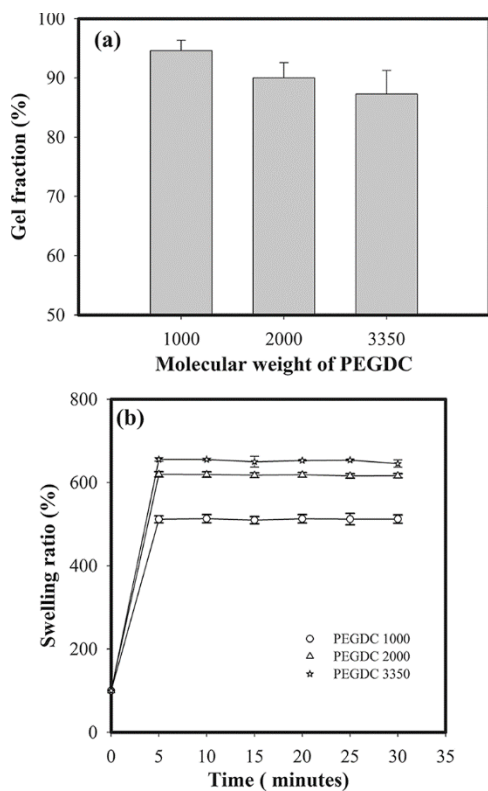


(a)

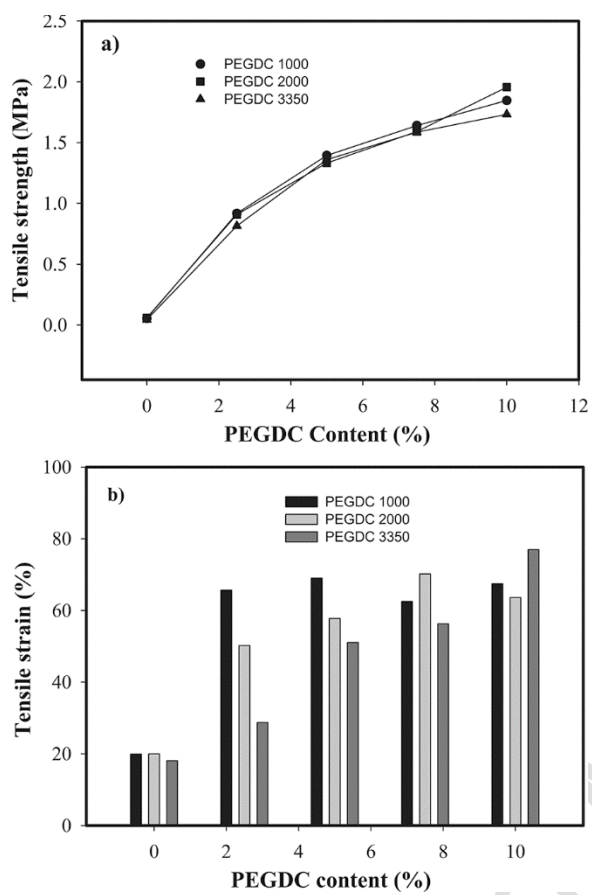


(b)

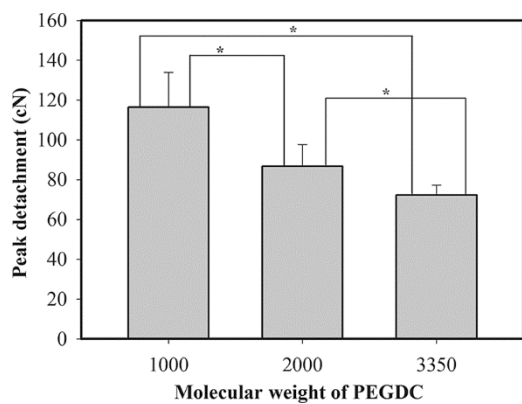
**Figure 2.** Effect of the molecular weight of PEGDC on (a) gel fraction, (b) on swelling ratio.



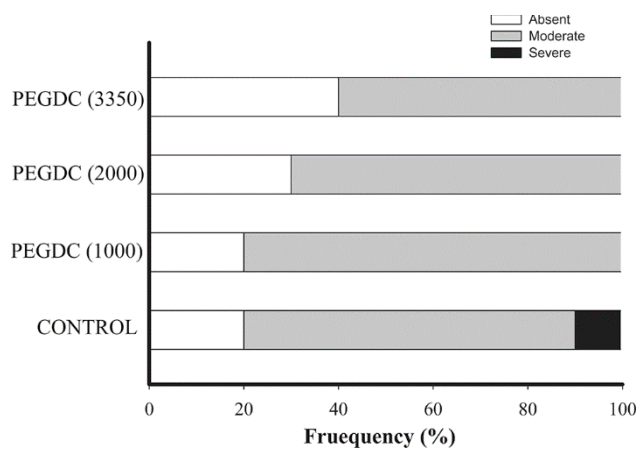
Accepted Manuscript

**Figure 3.** Effect of the PEGDC molecular weight on (a) tensile strength, (b) elongation break.

**Figure 4.** Peak Detachment Force of the PEGDC/PEO hydrogel film as a function of the PEGDC molecular weight.



**Figure 5.** Adhesion severity of various PEGDC/PEO hydrogels and the Control group.



Accepted Manuscript