Physicochemical Characterization and Drug Release of Thermosensitive Hydrogels Composed of a Hyaluronic Acid/Pluronic F127 Graft

Shu-Hui Hsu,[a] Yann-Lii Leu,[b] Jiuan-Wen Hu,[c] and Jia-You Fang*,[c]

[a] *Department of Pharmacy, Chia Nan University of Pharmacy and Science; Tainan County 717, Taiwan:* [b] *Natural Products Laboratory, Graduate Institute of Natural Products, Chang Gung University; and* [c] *Pharmaceutics Laboratory, Graduate Institute of Natural Products, Chang Gung University; Kweishan, Taoyuan 333, Taiwan.*

Received August 25, 2008; accepted February 9, 2009; published online February 10, 2009

Pluronic F127 (PF127) is a copolymer which forms thermosensitive hydrogels. Hyaluronic acid (HA) was grafted to PF127 to form a new hydrogel matrix (HP) for delivery of cisplatin and carboplatin. The physicochemical properties and drug delivery of the graft were examined in this study. HP system (20% HP copolymer in water) exhibited a similar sol–gel transition temperature (28.3 °C) as PF127 system (20% PF127 copolymer in water) but with a shorter gelling process. A stronger structure was obtained by HP system according to scanning electron microscopic (SEM) images and viscosity kinetics. *In vitro* release test showed the sustained-release characteristics of hydrogels entrapped with cisplatin and carboplatin. The drug release rate from HP hydrogel was slower than that from PF127 hydrogel. The reduction of drug release by HP system as compared to the control solution was more significant for cisplatin than for carboplatin. Such a thermosensitive hydrogel may be advantageous as an injectable therapeutic formulation for anticancer treatment.

Key words hyaluronic acid; Pluronic F127; thermosensitive hydrogel; cisplatin; carboplatin

Hydrogels are polymer-based systems that embrace numerous biomedical and pharmaceutical applications. Much interest has been focused on polymer systems that show a phase transition in response to temperature.[1)] Pluronic F127 (PF127) or Poloxamer 407 is non-ionic, polyoxyethylene–polyoxypropylene–polyoxyethylene tri-block copolymers which form micelles at low concentrations and clear, thermoreversible hydrogels at concentration above 20%.[2)] When injected into a body cavity, the gel preparation forms a solid artificial barrier and a sustained release depot.[3)] PF127 systems can prolong drug release which is one of the very few synthetic polymeric materials approved by the U.S. Food and Drug Administration for use as food additives and pharmaceutical ingredients. Potential drawbacks of PF127 hydrogels include their weak mechanical strength, rapid erosion, and the non-biodegradability.[4)] To circumvent these problems, PF127 can be chemically modified by other polymers. Chemical modifications of copolymers not only improve the properties of the polymers but also impart new and more attractive properties of them.

The use of natural polymers such as proteins and polysaccharides for biomedical application has attracted much investigation. Hyaluronic acid (HA) is an attractive building block for new biocompatible and biodegradable polymers with applications in drug delivery, tissue engineering, and also viscosupplementation to restore lubrication in osteoarthritic knee joints.[5)] It is a natural-occurring linear polysaccharide. In this study, HA grafted with PF127 was prepared as a thermosensitive material (HP) for heterogeneously-structured formulation composed of drug reservoir. There are two investigations which prepare the HP composite as the delivery vehicles of human growth hormone and ciprofloxacin.[6,7)] The synthetic method used was photo-polymerization or chemical reaction. Although the sol–gel temperature and drug release behaviors are examined previously, however, the physicochemical characterization of the hydrogels such as polarity, viscosity, and microstructural appearance are not fully explored. The aim of this study was to elucidate the correlation of theses physicochemical properties related to drug release. Two platinum drugs, cisplatin and carboplatin (Fig. 1), were used as the model drugs loaded in hydrogels because of their short half-lives and toxicity in human body.[8)]

Experimental

Materials Cisplatin, carboplatin, Nile red, and Pluronic F127 (PF127, polyoxyethylene–polyoxypropylene–polyoxyethylene tri-block copolymer, molecular weight=12.5 kDa, lot number 103K0058) were purchased from Sigma Chemical (St. Louis, MO, U.S.A.). 1-Ethyl-3-(3-dimethylaminopropyl)-carbodiimide (EDC, lot number A0219466) and *N*-hydroxylsuccinimide (NHS, lot number A0188987001) were obtained from Acros Organics (Geel, Belgium). Hyaluronic acid (HA) was produced from *Streptococcus zooepidemicus* by a method described by Armstrong and Johns.[9)] The molecular weight of HA was 1780 kDa (polydispersity=1.68) determined by size exclusion chromatography.[10)]

Synthesis of HA/PF127 Graft (HP) Graft copolymers were prepared by coupling mono amine-terminated PF127 (MATP) with a HA backbone using EDC and NHS as coupling agents. In the first step, PF127 (5 g) was reacted with 4-nitrophenyl chloroformate (80 mg) dissolved in 50 ml methylene chloride in the presence of triethylamine (0.15 ml) at room temperature for 4 h to yield an intermediate. This intermediate was recovered by extraction three times using petroleum ether. In the second step, the intermediate was reacted with diaminoethylene (1 ml) dissolved in methylene chloride (50 ml) at room temperature for 12 h. After reacting, the mixture was extracted three times with petroleum ether, then dialyzed against double-distilled water (ddH_2O) using a membrane with a molecular weight cutoff of

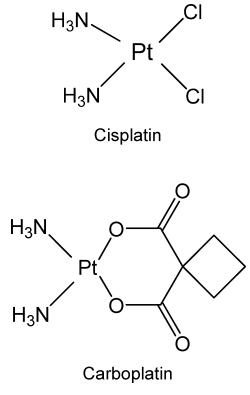

Fig. 1. Chemical Structures of Cisplatin and Carboplatin

* To whom correspondence should be addressed. e-mail: fajy@mail.cgu.edu.tw

3500 for 3 d, and then was freeze-dried to obtain the resulting product.

MATP (5 g) and HA (0.34 g) were dissolved in 50 ml ddH_2O. EDC (0.46 g) and NHS (1.38 g) were added to the above solution for 24 h. After reacting, the mixture was dialyzed for 3 d using a membrane with a molecular weight cutoff of 12000—14000, and then freeze-dried to obtain the HP copolymer.

Polarity of Copolymers The hydrophobic fluorescent marker, Nile red (2.5×10^{-5}%, w/w), was used as the model solute, and the molecular environment (polarity) was elucidated by fluorometric spectroscopy due to the solvatochromism of Nile red. Emission fluorescence spectra were determined with a Hitachi F-2500 fluorescence spectrophotometer. The spectra of the aqueous solutions of copolymers loaded with Nile red were recorded at room temperature with both slit widths set to 10 nm. The excitation wavelength was fixed at 546 nm, and emission spectra were recorded from 550 to 700 nm at a scanning speed of 300 nm/min.

Hydrogel Preparations The PF127 or HP copolymer was mixed with ddH_2O to a concentration of 20% (w/w). The resulting solution was left overnight in a refrigerator at 4 °C. A homogenous solution with a clear appearance was obtained. The mixtures were allowed to reach room temperature (less than 25 °C) before the experiments.

Sol–Gel Transition Temperature of Hydrogels Determined by Viscosity Viscosity measurements were used to assess the gelation behavior of the hydrogels. The viscosities of these systems were determined using a Brookfield RVDV-II viscometer (Middleboro, MA, U.S.A.). The spindle (s29, 40 mm) was rotated in the samples at a speed of 30 rpm, and the viscosity was determined from 22 to 31 °C.

Scanning Electron Microscopy (SEM) Examination of Freeze-Dried Hydrogels Hydrogels were frozen at −80 °C and then lyophilized by a freeze-dried method. Samples were fractured in liquid nitrogen and sputter-coated with gold. The resulting dried samples were examined using a Hitachi S3000N SEM (Tokyo, Japan).

Equilibrium Swelling Ratio of Hydrogels The method was modified from Van Tomme *et al.*[11] Hydrogels (1 g) were transferred into glass vials with a determined diameter and gently centrifuged (1000 rpm, 10 min). Subsequently the hydrogels were equilibrated at 4 °C for 12 h. The length of the hydrogels was measured (L_0) and 200 μl ddH_2O was added. The vials were incubated at 37 °C and at regular time intervals the length of the hydrogels was measured (L_t) to calculate the swelling ratio ($L_t/L_0\times100\%$).

Release of Cisplatin and Carboplatin from Hydrogels Drug release from the hydrogels was measured using a Franz diffusion cell. A cellulose membrane (Spectrapor® 3, with a molecular weight cutoff of 3500, Spectrum Laboratories, U.S.A.) was mounted between the donor and receptor compartments. The membrane was hydrated in ddH_2O for 12 h before the experiment. The donor medium consisted of 0.5 ml of a hydrogel formulation. The drug concentration was 2.6 mM. The receptor medium consisted of 5.5 ml of citrate–phosphate buffer at pH 7.4. The available diffusion area between cells was 1.13 cm^2. The stirring rate and temperature were maintained at 600 rpm and 37 °C, respectively. At appropriate intervals, 300-μl aliquots of the receptor medium were withdrawn and immediately replaced with an equal volume of fresh buffer. The amount of drugs was determined by measuring the absorbance by graphite furnace (flameless) atomic absorption spectrometry (Z5000, Hitachi, Tokyo, Japan).

Statistical Analysis Statistical analysis of differences between the various treatments was performed using unpaired Student's *t*-test. A 0.05 level of probability ($p<0.05$) was taken as the level of significance. An analysis of variance (ANOVA) test was also used.

Results

Synthesis of HP Copolymer The HP copolymer was prepared by coupling MATP with HA by the EDC/NHS method. Amide bonds were formed through the reaction between amino groups of MATP and carboxyl ones of HA. The efficiency of grafting (%) was 92.7% which was calculated by the equation for measuring polymer grafting percentage[12]: $(W_{HP}-W_{HA})/W_{MATP}\times100\%$, where W_{HP} is the weight of the freeze-dried graft copolymer, and W_{HA} and W_{MATP} are the weights of HA and MATP in the feed, respectively. The information about synthesis of HP is summarized in Table 1. The yield of HP attained a high level of 93.3%. HA is a mixture with heterogeneous chain length thus composed of various molecular weight. The average molecular weight of HP was about 24.8×10^6 Da as determined by the end-group analysis.[12]

Table 1. The Efficiency of Grafting, Estimated Molecular Weight (MW), and Yield (%) of Hyaluronic Acid–Pluronic F127 (HP) Copolymer

Grafting parameter	Value
The efficiency of grafting (%)	92.7
Estimated MW ($Da\times10^6$)	24.8
Yield (%)	93.3

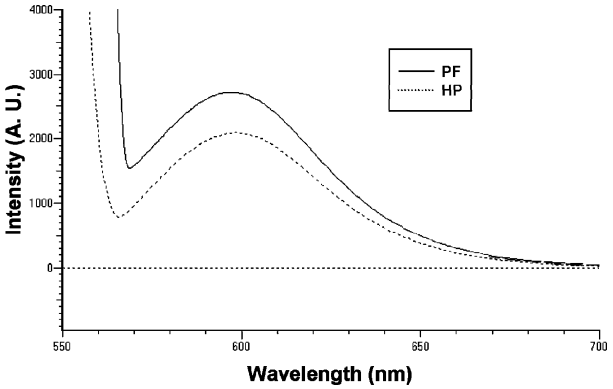

Fig. 2. Fluorescence Emission Spectra of Nile Red (2.5×10^{-5}%, w/w) in Pluronic F127 and HA/Pluronic F127 (HP) Graft Systems

Polarity of Copolymers Nile red is a dye whose absorption bands vary in shape, position, and intensity with the nature of the environment. The emission spectra of Nile red in copolymer solutions are shown in Fig. 2. Nile red is very soluble in organic solvents such as acetone and strongly fluorescent in a lipophilic environment.[14] The fluorescence of Nile red is quenched in a more-hydrophilic environment.[15] The emission maxima were found to be near 600 nm. Grafting of HA to PF127 exhibited a reduction in the fluorescence intensity at 604 nm. This indicates an increase in environmental polarity after conjugation.

Sol–Gel Transition Temperature of Hydrogels All copolymers tested in this study were soluble in water (20%, w/w) at room temperature. Gel formation was traced through the viscosity of the systems, which significantly increased during gelation process. Figure 3 illustrates the temperature-dependent gelation of PF127, which began to gel process at 26.4 °C (thin arrow). PF127 system showed an abrupt increase in viscosity until 28.2 °C (thick arrow). HA itself has a viscoelastic property. The HA solution at a concentration the same as the HA content in HP system (1.37%, w/w) was utilized to examine its viscosity. A constant viscosity of approximately 5300 cP was obtained for HA solution from 22 to 31 °C. The baseline of viscosity of HP system was similar to the average viscosity of HA system. The temperature at 27.2 °C marked the onset of the gelation process (thin arrow). Then a steep increase in viscosity was occurred at 28.3 °C (thick arrow).

SEM of Freeze-Dried Hydrogels The morphology of the hydrogels in a cross-section manner was examined by SEM as shown in Fig. 4. The PF127 microstructure in hydrogels assembles into a transient polymeric network as exhibited in Fig. 4A (magnification×1000). The SEM image of

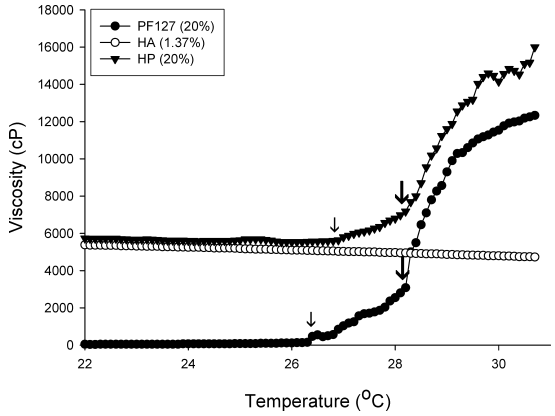

Fig. 3. Effect of Temperature on the Viscosities of Hydrogels with Pluronic F127, HA, or HA/Pluronic F127 (HP) Graft Hydrogels Determined by a Brookfield RVDV-II Viscometer

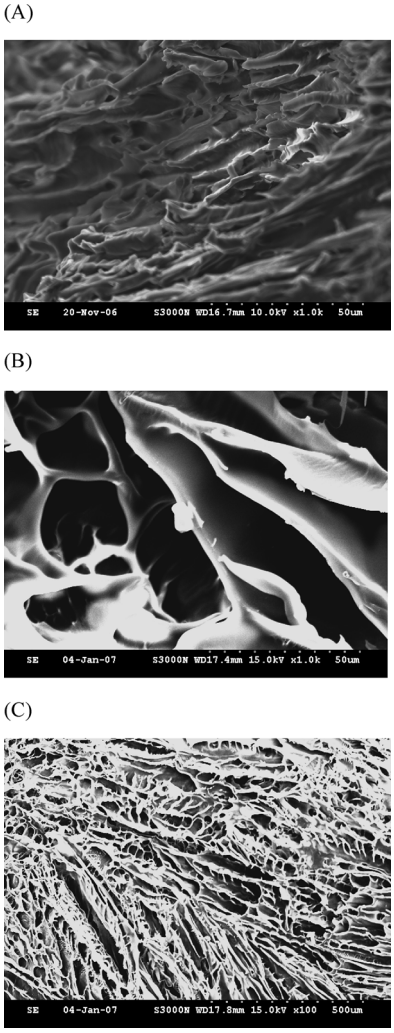

Fig. 4. Cross-Sectional Scanning Electron Microscopic (SEM) Images of Hydrogels after the Freeze-Drying Process Composed of Pluronic F127 (PF) with a Magnification of ×1000 (A), HA/Pluronic F127 (HP) Graft with a Magnification of ×1000 (B), and HA/Pluronic F127 (HP) Graft with a Magnification of ×100 (C) at a Concentration of 20% (w/w)

the freeze-dried HP hydrogel shows a regular texture with pores as shown in Fig. 4B (magnification×1000). One remarkable characteristic of the HP hydrogel was the deep pores. It can be seen that the pore size ranged between 20 and 30 μm. Enlarging the field of view in the image of the HP hydrogel (magnification×100) reveals a porous structure composed of sheets with interconnecting channels (Fig. 4C).

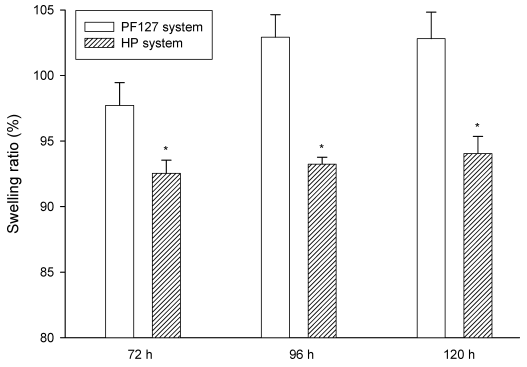

Fig. 5. Swelling Ratio of Pluronic F127 and HA/Pluronic F127 (HP) Graft Systems at Various Time Periods

Each value represents the mean±S.D. (n=4). *p<0.05 as compared to the value of PF127 system.

Equilibrium Swelling Ratio of Hydrogels The degradation of hydrogels composed of PF127 or HP was studied by swelling experiments at 37 °C. Figure 5 shows that there is almost no chemical disintegration of PF127 hydrogel produced by water during a 120-h period. HP hydrogel exhibited a more-significant degradation as compared to PF system (p<0.05). A less than 6% reduction of swelling ratio was observed for HP hydrogel at the end of experiment (120 h).

Release of Cisplatin and Carboplatin from Hydrogels Cisplatin was incorporated in the hydrogels and its *in vitro* release kinetics was characterized. The percentage of cisplatin released from the hydrogel formulations is plotted as a function of time in Fig. 6A. The composition of the hydrogels significantly affected the rate of cisplatin release. Cisplatin in ddH_2O at a dose of 2.6 mM was used as the control and a nearly complete drug release was observed in this aqueous control. The drug release of the control was virtually complete by 6 h. The entrapment of cisplatin in the hydrogels could significantly retard its *in vitro* release (p<0.05). The release profiles of the hydrogels demonstrate an initial burst release. Afterwards, the hydrogels steadily released cisplatin until the termination of the experiment. A Higuchi model (release percentage–$t^{1/2}$) was fitted for cisplatin release (Fig. 6B). Cisplatin from the HP system was released much more slowly than from the PF127 system (p<0.05). The direct mix of PF127 and HA (physical mixture) in the same ratio with the HP copolymer as the cisplatin vehicle did not produce drug release as low as that of the HP hydrogel. That is, the drug release from the physical mixture was significantly higher (p<0.05) as compared to that from the grafting copolymer. This suggests that the chemical grafting of PF127 and HA was necessary to further sustain cisplatin delivery.

The derivative of cisplatin, carboplatin, was also tested for its delivery from thermosensitive hydrogels. As shown in Fig. 7A, hydrogels of PF127, HP, and the PF/HA mixture can sustain the release of carboplatin with the trend the same as cisplatin. The reduction of drug release by HP system as compared to PF127 system was less significant for carboplatin than for cisplatin. Carboplatin release was also suitably fitted by Higuchi equation (Fig. 7B).

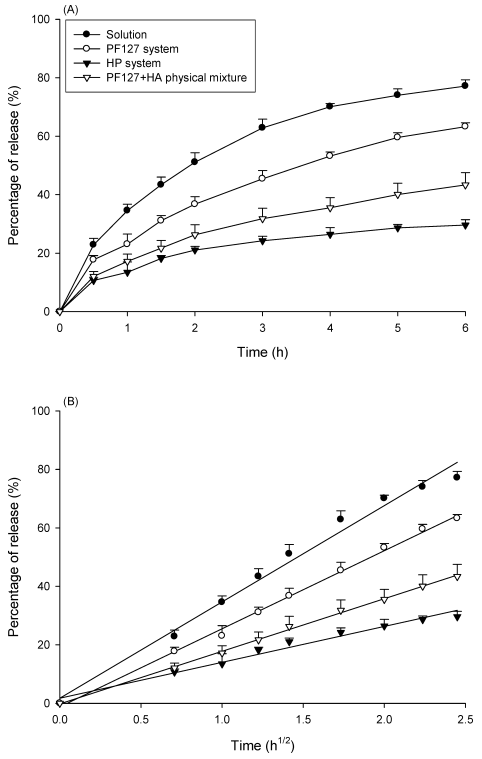

Fig. 6. Release Percentage (%)–Time Profile (A) and Release Percentage (%)–Time$^{1/2}$ Profile (B, Higuchi Model) of Cisplatin (Measured as Platinum by Atomic Absorbance) across a Cellulose Membrane from the Free Form (Control), a Pluronic F127 Hydrogel (PF), a HA/Pluronic F127 Graft Hydrogel (HP), and a Physical Mixture of HA and Pluronic F127

Each value represents the mean±S.D. (n=4).

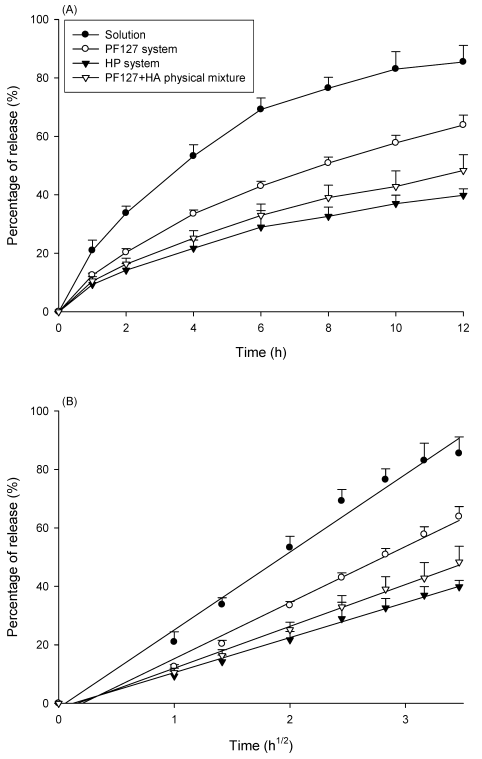

Fig. 7. Release Percentage (%)–Time Profile (A) and Release Percentage (%)–Time$^{1/2}$ Profile (B, Higuchi Model) of Carboplatin (Measured as Platinum by Atomic Absorbance) across a Cellulose Membrane from the Free Form (Control), a Pluronic F127 Hydrogel (PF), a HA/Pluronic F127 Graft Hydrogel (HP), and a Physical Mixture of HA and Pluronic F127

Each value represents the mean±S.D. (n=4).

Discussion

The thermosensitive properties of hydrogels can provide a unique injectable hydrogel drug delivery system. The drug can be incorporated into the gel in a syringe at room temperature without any contact with organic solvents. The drug-loaded hydrogel then can be injected into a tissue under pressure because of the thixotropic property.[16] After restoration of the gelation, the hydrogel serves as a drug depot.

As measured by viscosity, the sol–gel transition temperature of PF127 and HP was lower than the body temperature, indicating the feasibility for clinical application. PF127 is surface active and forms micelles and liquid lyotropic crystalline phases. Packing of micelles and micellar entanglements is a possible mechanism of PF127 solution gelation with an increase in temperature.[4] As observed in Fig. 3, the attainment of complete gelling after initiation of gelation was slow for PF127 (26.4—28.2 °C). The viscosity of the HP system showed a quicker increase (27.2—28.3 °C) as compared to that of the PF127 system. It suggests that the presence of hydrophilic HA in the vicinity of PF127 may interfere the hydrophobic interaction of PF127. The drug loading into the structure may be limited by the slow gelation process. It would be attractive if the gelation of the thermosensitive hydrogel can be quickly attained in an aqueous environment.[16] HP may be applicable to achieve this aim.

HA is reported to form helical structures in solution due to inter- and intra-molecular interactions,[17] contributing a viscoelastic performance of HA. However, HA itself is unable to form gel in aqueous solution. This viscous property of HA gave a determined viscosity of HP system before gelling process. The final viscosity of HP system was also higher than that of PF127 system. Insufficient viscosity or gelation can be observed *in vivo* resulting in high initial drug release and the following toxicity.[4] A high viscosity of a thermosensitive vehicle can play an important role in maintaining a non-flowing organization in body tissues.

At higher temperature above the sol–gel transition, a phase of hexagonal-packed cylinders is formed for PF127 system. Thermal gelation occurs by micellar desolvation and swelling to form a pseudo-cross-linkage among the PF127 copolymer structures,[18] which was confirmed by the SEM images (Fig. 4A). The conjugation of HA to PF127 had produced a different microstructure as compared to the parent PF127 hydrogel. The HP system showed a reticular, sponge-like, and three-dimensional network (Figs. 4B, C). The formation or increase of cross-linkage can improve the stability of the network,[19] thus influence drug release.

In the process of hydration of a polymer material, water binds to the most-hydrophilic groups. The addition of a hydrophilic polymer can increase water uptake.[19] The water molecules could be absorbed in the pores of the HP hydrogel. The amide group which is rich in HP is still regarded as highly hydrated as compared to the carboxyl group.[20] An increase in water content in the hydrogels may result in a quicker chemical degradation of the hydrogels.[11] This degradation caused weight loss and decrease in gel volume of the HP system.

Compared to the aqueous control, entrapment of cisplatin

in hydrogels resulted in a prolongation of drug release. Around 30 to 80% of the cisplatin in the aqueous solution (control) and hydrogels was released to the receptor by the end of the experiment (5 h). There may be some drug molecules interacted to backbone of HP copolymer, which are not easy to escape from the structure. Another reason of the limited amount released may have been due to the use of the Franz diffusion assembly. Since a drug is released to the definitive space of receptor (5.5 ml) and diffusion area (1.13 cm^2), the drug loading in the receptor is limited. There is no longer a concentration gradient between the donor and receptor compartments. Nevertheless, this method is still useful for differentiating the release of various formulations, especially the free control. The results in Fig. 6 show that an initial burst release of cisplatin is observed for aqueous solution. HP provided a more-sustained drug release behavior as compared to PF and physical mixture. This suggests that the drug can be released from HP hydrogel for a prolonged time, which may reduce the administration frequency. Of course, it should be noticeable that the concentration of the released drug in the target tissue should attain the minimum pharmacologically effective concentration. Linear regression analysis of the release data of hydrogels was done to determine the proper order of release. Zero-, first-, and Higuchi model equations were applied to the release results. The cisplatin release was almost linear when using the Higuchi equation (release percentage–$t^{1/2}$) as shown in Fig. 6B. One can conclude that the drug was released from the hydrogels by a diffusion-controlled mechanism. Another observation is the lower drug release from physical mixture than from PF127 and control systems. This indicates that the addition of HA in PF127 hydrogel without chemical grafting can reduce drug delivery. A part of pathway filled by water was replaced by HA molecules in this physical mixture. The increase in hydrogel viscosity and prolongation of path length are the possible mechanisms for this release reduction.

HP could produce three-dimensional networks following water penetration into the structures. The release results clearly suggest that the cisplatin release was controlled by the stable and entangled networks as compared to PF127 hydrogel. The slow gelation process and insufficient viscosity of PF127 system also contributed to the quick cisplatin release. HP provided a fast gelling, thus could loaded more drug molecules into gel structure and lower their release from the system. As observed in the SEM images, PF system showed a soft and amorphous appearance. On the other hand, a rigid and cross-linkage structure was seen for HP hydrogel. This indicates that PF with a weaker mechanical strength may possess lower ability to retain the drug within the gel structure. Cisplatin is a platinum-chelated complex with four ligands: two ammonias and two chlorides (Fig. 1). According to previous studies,[21,22] the two chloride ligands are gradually substituted with H_2O molecules in water. Both $Pt(NH_3)_2Cl(OH_2)^+$ and $Pt(NH_3)_2Cl(OH_2)_2^{2+}$ can be formed by the aqueous complex based on the cisplatin dose used in this study. Since HA is a polyelectrolyte which is negatively charged, cisplatin was expected to form ionic interaction with HA on the HP copolymer backbone. Cisplatin interacts with HP not only by electrostatic force but also hydrophilic affinity. The ionic cisplatin complex is more hydrophilic than the cisplatin molecule alone, which may have a notable affinity to hydrophilic environment of HP hydrogel. Both interactions could contribute to the sustained release of cisplatin from HP hydrogel. The quicker degradation of HP system verified by swelling experiment may produce the release of cisplatin from this hydrogel. However, the gel degradation may not be the predominant factor governing drug delivery because PF127 even showed no degradation after water incorporation. Another observation is that further increment of incubation duration from 72 to 120 h did not degrade HP gel to a greater level. This suggests that HP system was degraded during a determined period. After which the degradation was almost stopped.

Carboplatin was selected mainly because of its lower non-hematological toxicity compared to cisplatin.[23] As shown in Fig. 7, carboplatin should attain its maximum release by a 12-h period, which was longer than cisplatin did. Carboplatin is more lipophilic than cisplatin.[24] Moreover, the formation of an aqueous complex may have further increased the hydrophilicity of cisplatin. Previous studies demonstrated that the release of a series of drug derivatives is a function of their hydrophilicities, with slower and more-sustained release occurring with more-lipophilic compounds.[2,5]

The reduction of carboplatin release after conjugating HA to PF127 was not as pronounced as compared to cisplatin release. HA in aqueous solution exhibits a random coil–coil structure with hydrophilic and hydrophobic strands.[5] Two platinum drugs may locate in the different sites with HP hydrogel structures. Carboplatin is produced by substituting chlorides in cisplatin with the ligand of 1,1-cyclobutane dicarboxylate (Fig. 1). Carboplatin cannot form the aqueous complex. Hence carboplatin was not expected to interact ionically with HP backbone. Based on the theory cited by Topp and colleagues,[24,25] lipophilic solutes may diffuse through the hydrophobic and hydrophilic pathways of the hydrated cross-linked polymer networks. On the other hand, it has been suggested that the major penetration pathways of solutes with a hydrophilic nature is only the hydrated portion of the hydrogel. Hence the release of cisplatin was limited to a greater degree as compared to carboplatin because of its higher hydrophilicity. This is why the release of hydrophilic cisplatin from HP hydrogel was limited. Further *in vivo* studies are planned. Terminal sterilization of products is a critical issue in the medical device. Traditional method for sterilization is heating. This method is not feasible for thermosensitive polymers. Sterilization under ambient temperature such as ultraviolet irradiation, high hydrostatic pressure, nanometer-scale filtration, and ethylene oxide treatment may be the methods for the thermosensitive systems.[26] Further experiment is needed to elucidate the suitable sterilization protocols.

Conclusions

The injectable, thermosensitive hydrogels were fabricated by chemically grafting HA and PF. A porous network of the cross-section of HP hydrogel was obtained. The complete gel-forming process could be shortened by HA incorporation. This hydrogel showed a higher mechanical strength and moderate degradation, thus the release of platinum drugs was sustained and prolonged. Since PF shows the disadvantages of weak mechanical strength and non-biodegradability, HP instead of PF can partially overcome these drawbacks. HP

hydrogel may serve as a rate-controlling barrier and be useful as drug vehicle to be administered intratumorally or intraperitoneally. Providing that similar release characteristics may occur *in vivo*, this result can be potentially important for developing formulations of cisplatin and carboplatin for cancer treatment. Further investigation about the *in vivo* animal study is needed in the near future.

References

1) Yoshizawa T., Shin-ya Y., Hong K. J., Kajiuchi T., *Eur. J. Pharm. Biopharm.*, **59**, 307—313 (2005).
2) Moore T., Croy S., Mallapragada S., Pandit N., *J. Controlled Release*, **67**, 191—202 (2000).
3) Escobar-Chávez J. J., López-Cervantes M., Naïk A., Kalia Y. N., Quintanar-Guerrero D., Ganem-Quintanar A., *J. Pharm. Pharmaceut. Sci.*, **9**, 339—358 (2006).
4) Ruel-Gariépy E., Leroux J.-C., *Eur. J. Pharm. Biopharm.*, **58**, 409—426 (2004).
5) Luo Y., Kirker K. R., Prestwich G. D., *J. Controlled Release*, **69**, 169—184 (2000).
6) Kim M. R., Park T. G., *J. Controlled Release*, **80**, 69—77 (2002).
7) Cho K. Y., Chung T. W., Kim B. C., Kim M. K., Lee J. H., Wee W. R., Cho C. S., *Int. J. Pharm.*, **260**, 83—91 (2003).
8) Konishi M., Tabata Y., Kariya M., Suzuki A., Mandai M., Nanbu K., Takakura K., Fujii S., *J. Controlled Release*, **92**, 301—313 (2003).
9) Armstrong D. C., Johns M. R., *Appl. Environ. Microbiol.*, **63**, 2759—2764 (1997).
10) Armstrong D. C., Johns M. R., *Biotechnol. Tech.*, **9**, 491—496 (1995).
11) Van Tomme S. R., van Nostrum C. F., de Smedt S. C., Hennink W. E., *Biomaterials*, **27**, 4141—4148 (2006).
12) Chen J. P., Cheng T. H., *Macromol. Biosci.*, **6**, 1026—1039 (2006).
13) Stevens M. P., "Polymer Chemistry," Chap. 2, ed. by Oxford University Press, New York, 1990, pp. 40—69.
14) Lombardi Borgia S., Regehly M., Sivaramakrishnan R., Mehnert W., Korting H. C., Danker K., Röder B., Kramer K. D., Schäfer-Korting M., *J. Controlled Release*, **110**, 151—163 (2005).
15) Jores K., Haberland A., Wartewig S., Mäder K., Mehnert W., *Pharm. Res.*, **22**, 1887—1897 (2005).
16) Li J., Ni X., Leong K. W., *J. Biomed. Mater. Res. A*, **65**, 196—202 (2003).
17) Jin Y., Yamanaka J., Sato S., Miyata I., Yomota C., Yonese M., *J. Controlled Release*, **73**, 173—181 (2001).
18) Guzmán M., García F. F., Molpeceres J., Aberturas M. R., *Int. J. Pharm.*, **80**, 119—127 (1992).
19) Kashyap N., Kumar N., Ravi Kumar M. N. V., *Crit. Rev. Ther. Drug Carrier Syst.*, **22**, 107—149 (2005).
20) Feil H., Bae Y. H., Feijen J., Kim S. W., *Macromolecules*, **26**, 2496—2500 (1993).
21) Yotsuyanagi T., Usami M., Noda Y., Nagata M., *Int. J. Pharm.*, **246**, 95—104 (2002).
22) Hwang T. L., Lee W. R., Hua S. C., Fang J. Y., *J. Dermatol. Sci.*, **46**, 11—20 (2007).
23) Desoize B., Madoulet C., *Crit. Rev. Oncol. Hematol.*, **42**, 317—325 (2002).
24) Joshi H. N., Topp E. M., *Int. J. Pharm.*, **80**, 213—221 (1992).
25) Sung K. C., Topp E. M., *J. Controlled Release*, **37**, 95—104 (1995).
26) Huebsch N., Gilbert M., Healy K. E., *J. Biomed. Mater. Res. Part B: Appl. Biomater.*, **74**, 440—447 (2005).