Available online at www.sciencedirect.com
SCIENCE DIRECT®
ELSEVIER
Biomaterials 27 (2006) 3031–3037
Biomaterials
www.elsevier.com/locate/biomaterials

Sustained release of dexamethasone from hydrophilic matrices using PLGA nanoparticles for neural drug delivery

Dong-Hwan Kim[a,d], David C. Martin[a,b,c,*]

[a]*Biomedical Engineering, University of Michigan, Michigan, USA*
[b]*Materials Science and Engineering, University of Michigan, Michigan, USA*
[c]*Macromolecular Science and Engineering at Michigan, University of Michigan, Michigan, USA*
[d]*Biomedical Engineering, Duke University, USA*

Received 22 September 2005; accepted 31 December 2005
Available online 27 January 2006

Abstract

The release of the anti-inflammatory agent dexamethasone (DEX) from nanoparticles of poly(lactic-*co*-glycolic acid) (PLGA) embedded in alginate hydrogel (HG) matrices was investigated. DEX-loaded PLGA nanoparticles were prepared using a solvent evaporation technique and were characterized for size, drug loading, and in-vitro release. The crosslinking density of the HG was studied and correlated with drug release kinetics. The amount of DEX loaded in the nanoparticles was estimated as ~13 wt%. The typical particle size ranged from 400 to 600 nm. The in-vitro release of DEX from NPs entrapped in the HG showed that 90% of the drug was released over 2 weeks. The impedance of the NP-loaded HG coatings on microfabricated neural probes was measured and found to be similar to the unmodified and uncoated probes. The in-vivo impedance of chronically implanted electrodes loaded with DEX was maintained at its initial level, while that of the control electrode increased by 3 times after about 2 weeks after implantation until it stabilized at approximately 3 MΩ. This improvement in performance is presumably due to the reduced amount of glial inflammation in the immediate vicinity of the DEX-modified neural probe.
© 2006 Published by Elsevier Ltd.

Keywords: Dexamethasone; Controlled delivery; Neural prosthetics; Hydrogel; PLGA nanoparticles

1. Introduction

For more than 2 decades, many different types of electrodes have been designed and used to monitor and stimulate neurons in the central and peripheral nervous system. A considerable amount of effort has been dedicated to understand the factors influencing biocompatibility and functionality of the inserted electrodes [1–6]. These neural electrodes include microwires, microfabricated microelectrode arrays (Utah probes) and micromachined planar electrodes (Michigan probes). While these devices generally work well in acute applications, there is excessive gliosis seen at the interface between the implant and neural component of interest in the brain resulting in loss of communication of the electrodes over extended periods of time [7,8]. This gliotic scar can be more than 50 μm thick, and is thought to play a direct role in increasing the barriers to communication between the neurons and the electrodes. The amplitude of neuronal peaks is likely to be significantly influenced by these connective tissue layers [9]. In addition, it has been found that there is also a significant decrease in the neuronal density around the electrode, in a region called the 'kill zone'. The kill zone has been defined as the region near the probe where the neuronal density is lower than the expected neuronal density by a 90% confidence [4]. It has been reported that neurons more than 50 μm apart from the recording sites can be hardly discriminated and neurons more than 140 μm apart are not recognizable [6]. Thus it is extremely important to reduce the size of the kill zone around the implant to achieve long term stable and

*Corresponding author. Department of Materials Science and Engineering, 2022 H.H. Dow Building, University of Michigan, Ann Arbor, MI 48109-2136, USA. Tel.: +1 734 936 3161; fax: +1 508 256 8352.
E-mail address: milty@umich.edu (D.C. Martin).

0142-9612/$ - see front matter © 2006 Published by Elsevier Ltd.
doi:10.1016/j.biomaterials.2005.12.021

analyzable neural recordings from the brain. In order to reduce the kill zone around the implant, a number of studies have been conducted by changing the insertion speed and method [1,3], the overall shape of the electrodes [1], or by using alternative materials such as polymers [5] or ceramics [2]. Among these efforts to understand the inflammation mechanisms and reduce tissue reactivity around the implant, the administration of anti-inflammatory drugs through either polymer coatings [10] or direct injection through microchannels incorporated into the electrodes [11] could be an effective method to limit the structural ensheathing layer caused by inflammation.

Dexamethasone (DEX) is a glucocorticoid that is used clinically as an anti-inflammatory and immunosuppressive agent. The intra-arterial administration of DEX-containing nanoparticles has been demonstrated as effective for reducing restenosis in a rat model [12]. It was reported that even a single systemic injection of DEX at the time of implantation attenuated astrycytic responses, and a more pronounced effect was observed following six daily injections [10]. In order to minimize the exposure of the whole animal body to the high concentrations of drug needed to obtain an appropriate concentration around implants and to avoid the side effect of chronic use, the local delivery of steroidal drugs such as DEX is highly desired. It also has been shown that local delivery of DEX from poly(ethyl-vinyl) acetate (EVAc) can significantly attenuate astrocytic response at 1 week after implantation [10]. However, the advantage of local administration of DEX through a polymer matrix had faded after 6 weeks presumably because the release of DEX was not sustained for this period of time. Furthermore, the polymer used in this experiment was an electrically non-conductive material and thus cannot be used for coating neural prosthetic devices, since electrical conductivity is one of the most important parameters for achieving stable communication with neurons. Therefore, it is required to have alternative coating materials that will maintain the electrical properties of the electrodes and control the release of drugs for longer periods of time (>2 weeks) after implantation of the electrodes in the brain.

Much interest has been focused on preparation and characterization of biodegradable micro/nanoparticles as a delivery carrier of interesting pharmacological agents due to their potential usefulness in increasing efficacy [13], reducing enzymatic degradation [14] and controlling release rates [15]. The most commonly used materials for the preparation of these carriers are polylactide (PLA) and poly(lactide-*co*-glycolide) (PLGA) that are well known to be safe and biocompatible [16]. Although there are a large number of studies on the controlled release of various drugs from a PLGA/PLA polymer matrix, only a few studies have been performed on the controlled release of hydrophobic drugs through a hydrophilic hydrogel matrix. DEX is insoluble in water and has many pharmaceutically acceptable solvents. Alginate is a water-soluble, natural polysaccharide that can be gelled through ionic or covalent crosslinking [17]. Many alginate-based hydrogels have been used as drug delivery carriers. Hydrogel coatings on the neural probes should provide a mechanical buffer layer between the hard silicon-based probe and the soft brain tissue. In addition, the reswelling of dried hydrogel coatings should better anchor the position of the probe in the tissue post-implantation [18]. In this study, we are investigating (1) the formulation and in-vitro and in-vivo release characterization of DEX-loaded PLGA nanoparticles, (2) the characterization of several parameters of formulations which can affect the efficacy of drug loading and particle sizes, (3) the evaluation of in-vivo impedance of chronically implanted electrodes in the Central Nervous System of the Guinea pig.

2. Materials and methods

2.1. Materials

The sodium alginates were purchased from ProNova Biomedical (Norway). MVG alginate, a high G-containing alginate (M:G ratio 40:60 as specified by the manufacturer) was used in this study. Poly(DL-lactide-*co*-glycolide 50/50) of average molecular weight 40,000–75,000 g/mol and poly(vinyl alcohol) of Mw 15,000 g/mol were purchased from Sigma Aldrich. DEX (Mw 392.5 g/mol) was supplied by Alexis. Water-soluble DEX (W-DEX), containing approximately 65 mg DEX per gram with the balance 2-hydroxypropyl-β-cyclodextrin, was purchased from Sigma Aldrich. All other chemicals were purchased from Sigma (St. Louis, MO) unless otherwise stated. All chemicals were used without further purification.

2.2. Experimental

2.2.1. Nanoparticles preparation

PLGA nanoparticles loaded with DEX were prepared by a single oil-in-water (O/W) emulsion/solvent evaporation method [19–21]. In order to maximize drug loading in the nanoparticles, PLGA (800 mg) was dissolved in dichloromethane (15 ml) and then the DEX (200 mg) was dissolved in acetone (15 ml). The DEX in acetone was added to the PLGA in dichloromethane to form the oil phase. This oil phase was added dropwise to an aqueous solution containing 5% PVA. An oil-in-water (O/W) type emulsion was formed with a sonicator (Fisher 500 Sonic Dismembrator, Fisher Sci.) with a constant power output of 60 W for 10 min. The sample was kept on an ice bath to prevent overheating. The organic solvent was evaporated while being stirred gently at room temperature for 12 h. The unreacted drug and PVA residue were washed 3 times with DI water and the nanoparticles were collected using an ultracentrifuge at 35,000 RPM for 1 h (Beckman Coulter Optimal L-90K, Beckman, Arlington Heights, IL). A fine powder was obtained by lyophilization.

2.2.2. Preparation of alginate hydrogel loaded with PLGA NPs

The hydrogels were deposited by a dipping method, using a 1–3 wt% alginate solution that was gelled by ionic crosslinking with 0.5 M $CaCl_2$. The details of alginate hydrogel coating on the neural electrodes are described elsewhere [18]. In order to simplify observations of drug release profiles, we made phantom alginate spheres instead of coating cortical probes as a matrix through which DEX can be released. Sodium alginate solutions at concentrations of 1–3 wt% were freshly prepared by dissolving the alginate powder in double-distilled water, while mixing with a magnetic stirrer. Various amounts of NPs loaded with DEX were added to the alginate solution, and then the alginate solutions containing NPs were dropped using a syringe through 20-gauge needle into $CaCl_2$

solution (0.5 M) as a source of calcium ions. Ionically crosslinked alginate beads were formed and cured in the $CaCl_2$ solution for 30 min at room temperature. The alginate beads were then collected by filtration and gently washed twice with deionized water. The average diameter of the beads was 2.5 mm.

2.2.3. Encapsulation efficiency

The amount of DEX entrapped within the NPs was determined by UV spectrophotometry. 10 mg of freeze-dried NP was taken in triplicate, and dissolved in 10 ml of mixture of CH_2Cl_2 and acetone and then quantified by measuring the UV absorbance at 242 nm. At this wavelength there was no interference in the absorbance reading from the PLGA. The amount of non-entrapped drug recovered in the supernatant was measured after ultracentrifugation of the NP at 35,000 rpm for 1 h.

2.2.4. Particle size

The particle size and morphological characteristics of NPs were observed and quantified with a Scanning Electron Microscope (SEM, Philips XL 30 FEG, Netherlands). A dilute suspension of NPs in deionized water was spread on the sticky surface of an aluminum stub then placed in the desiccator until completely dehydrated. The samples were coated with thin Au-alloy (~10 nm) prior to examination to reduce charging.

2.2.5. In-vitro release experiments

The in-vitro drug release from the hydrogels, the free NPs and the NPs entrapped in hydrogels was performed in sodium phosphate buffer solution (PBS, ph 7.4, 0.1 M). At regular time intervals, an aliquot (3 μl) was removed from stock solution of PBS with DEX in the hydrogels, DEX in the free NPs and DEX in the NPs entrapped in hydrogels and measured at 242 nm using UV spectrophotometry (Beckman Coulter, DU 800).

2.2.6. Impedance spectroscopy measurements

An Autolab FRA2 was used to obtain impedance spectra of the electrode sites. The apparatus was comprised of an Autolab model PGSTAT 12 Potentiostat/Galvanostat and an Autolab FRA 2 module. A solution of 0.1 M phosphate buffer solution was used as the electrolyte in a three-electrode cell. The electrode setup was the same as in previous studies [18]. An AC sinusoidal signal of 5 mV amplitude was used and the DC potential was set to 0 V. The value of the impedance was determined over a range of frequencies from 10 to 100,000 Hz.

2.2.7. Surgical procedure and in-vivo impedance measurement

Guinea pigs weighing between 300 and 500 g were used for this study and NIH guidelines for the care and use of laboratory animals (NIH Publication #85-23 Rev. 1085) have been observed. The animal was initially anesthetized with Ketamine (40 mg/kg) and Xylazine (5 mg/kg), with supplemental injections given regularly to maintain appropriate levels of anesthesia. At no time was the animal allowed to awaken from the anesthesia. The animal's head was shaved and the animal was placed on a heating blanket, with feedback, to prevent a drop in core body temperate. The surgical procedure consisted of a mid-line incision on the head of approximately 1.5 inches. The skull was fixed with a high degree of stability to a rigid bar by a bolt embedded in dental acrylic and anchored to the dorsal cranium with stainless steel screws. A craniotomy was performed to expose the auditory cortex or the cerebellum, the dura was removed, and the electrode inserted. A chamber was mounted to the skull, and surrounding the cranial defect, the chamber was filled with agar solution over which dental acrylic was placed. The skin was sutured and allowed to recover. During the recovery period, the University Committee on Use and Care of Animals (UCUCA) approved post-op monitoring procedures were followed. The impedance at 1 kHz of the each site of the DEX-loaded electrodes was measured over time with impedance apparatus.

3. Results and discussion

3.1. Preparation and characterization of DEX-loaded NPs

The main objectives of this work were to obtain DEX-loaded NPs, to embed the NPs into the hydrogel coatings on the microfabricated electrodes, and to evaluate their in-vivo release characteristics. Therefore, we prepared NPs using a single oil-in-water solvent evaporation method with modified recipe which was adjusted to this system due to the difference in solubility of PLGA and DEX in organic solvents. Several parameters including the ratio of polymer vs. solvent and DEX vs. solvent, the amount of emulsifier, and the output power of the sonicator were tested in order to obtain appropriate particle size and release profiles. It was observed that the concentration of the emulsifier (0.5%, 2.5%, 5.0%) in the water phase had little difference in the size of NPs. In order to maximize the amount of drug loading in the nanoparticles, PLGA (800 mg) and DEX (200 mg) were dissolved in dichloromethane (15 ml) and acetone (15 ml), respectively. For determining drug loading and efficiency, weighed NPs were extracted using a mixture of CH_2Cl_2 and acetone (1:1), and the extracts then were assayed for DEX content by UV spectrophotometry. A drug entrapping efficiency of 79 ± 5 wt% was observed with 13 ± 3 wt% drug loading.

Fig. 1 shows optical images of hydrogel coatings on neural probes (a) without NPs, (b) with NPs in the hydrogel coating. The NPs in the hydrogel coatings were aggregated together to form chunks of NPs that were easily detected in the microscope and with an optical spectrometer. The addition of NPs to the alginate solution did not make uniform coatings difficult until the amount of NPs exceeded 10 wt%. Using UV spectrophotometry the release behavior of various drugs including water-soluble dexamethasone (W-DEX, a complex of DEX and cyclodextrin), water-insoluble dexamethasone (I-DEX) and naproxen (water soluble) from alginate hydrogels was measured. Regardless of the composition, the drugs were released within a very short time (less than 6 h) presumably because the characteristic pore size of an alginate hydrogel is much greater than the size of model drugs (Fig. 2).

Fig. 3 shows a typical SEM image of DEX-loaded NPs having a nominal diameter of 400–600 nm. The SEM morphologic observation of NPs loaded with DEX demonstrated uniform, rounded particles. Based on the smooth spherical surface of NPs, the presence of drug nanocrystals around the NPs was not observed. This precipitation can occur when the desired agents in the NPs are supersaturated in the process of preparation of NPs. In-vitro impedance spectroscopy of control (bare gold electrode, 1250 μm^2), hydrogel-coated electrodes and NP-loaded hydrogel-coated electrodes (10 wt% of NPs) was measured in the PBS buffer solution. Neither the drug- nor NP-loaded hydrogel-coated electrodes showed a significant increase of the impedance over a frequency range from 10 to 100,000 Hz as compared with the gold electrode

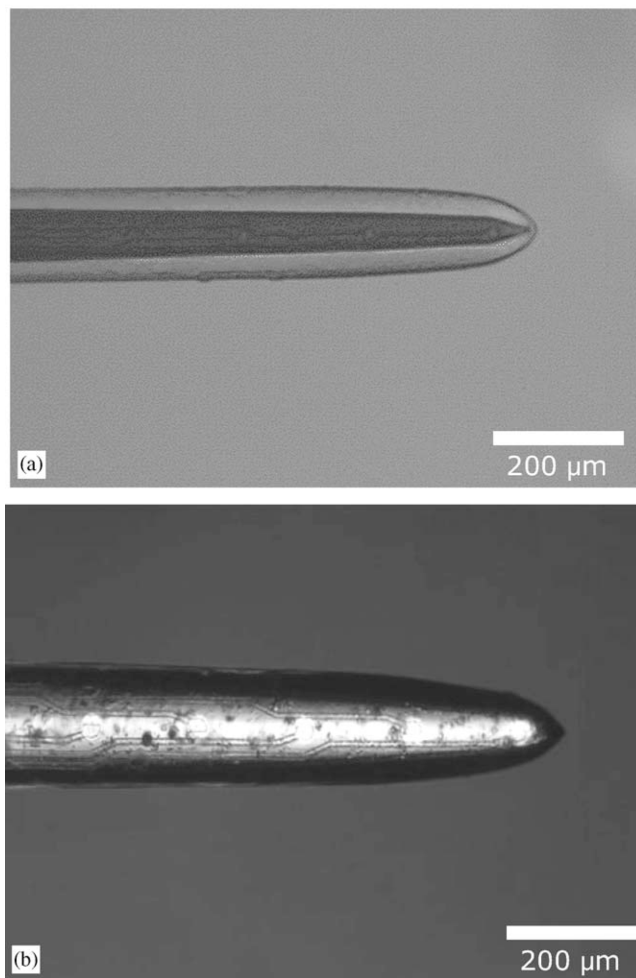

Fig. 1. Optical images of hydrogel-coated neural microelectrode arrays: (a) without NPs, (b) with NPs in the hydrogel coating.

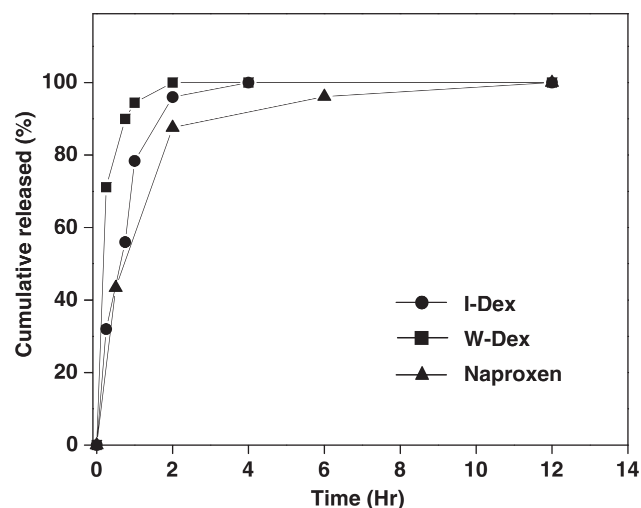

Fig. 2. The release behavior of various drugs such as water-soluble dexamethasone (W-DEX), water-insoluble dexamethasone (I-DEX) and naproxen from alginate hydrogels ($n = 3$).

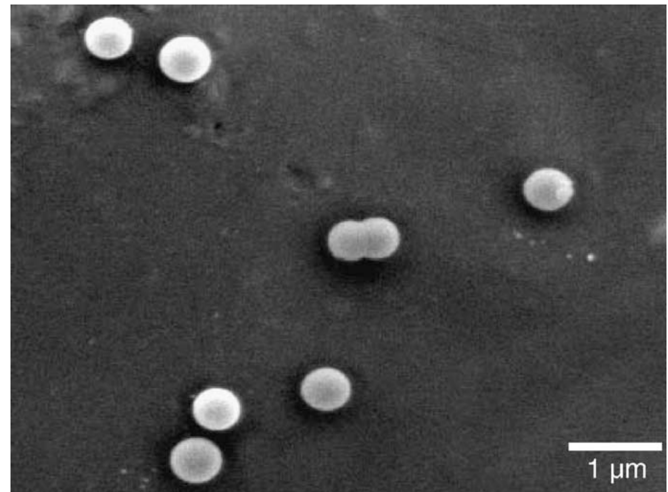

Fig. 3. A typical SEM image of DEX-loaded NPs having a nominal diameter of 400–600 nm.

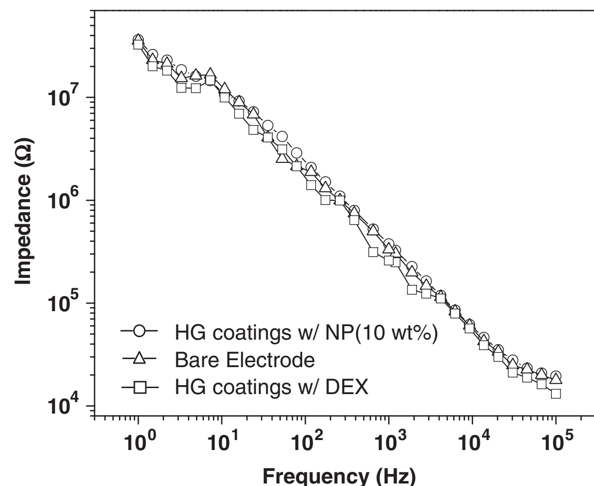

Fig. 4. In-vitro impedance spectroscopy of control (bare electrode), drug-loaded HG coating and NP-loaded HG coating (10 wt%) ($n = 3$).

controls, indicating that neither the NPs nor the drugs entrapped in the hydrogel coating significantly hindered electrical transport between the electrode sites and the buffer solution (Fig. 4).

3.2. In-vitro DEX release from NPs

The in-vitro release profiles of DEX either from free NPs in the PBS solution or NPs entrapped into hydrogels are shown in Fig. 5. Free NPs in PBS solution shows an initial burst of 60% during the first 2 days (a) followed by a gradual reduction of release while NPs entrapped in the hydrogels at both 3% (b) and 1% (c) of alginate concentration show only 20% release during the first day. DEX release from the NPs happens either by diffusion through the polymer barriers or by erosion of the polymer materials due to the hydrolytic degradation of ester linkages. Because of the hydrophobic characteristics of

DEX in a single emulsion formulation, it is likely that the encapsulated substance will agglomerate toward the NP surface during solvent evaporation. It is therefore expected that micro/nanoparticles prepared by emulsification solvent evaporation will present an initial burst release due to surface-located encapsulated substance. The NPs entrapped in the hydrogels show a relatively moderate initial burst presumably due to the buffering effect of the hydrogels as shown in Fig. 5. Note that this buffering effect of the hydrogel on the release of DEX (the extent of delayed time of DEX release through hydrogel) can be estimated to be less than 4 h as shown in the Fig. 2. However, the extent of the delayed time for DEX release from the NPs entrapped in the hydrogel is much greater than 4 h (Fig. 5). At 20% of the cumulative amount of DEX released the delayed time in the hydrogel was about 20 h, and at 40% the delayed time was about 100 h. DEX molecules can be preferably localized toward the surface of a nanoparticle due to increased hydrophobicity of hydrocarbon tails in a micelle while being solidified in the presence of emulsifier. Release of DEX from the micelle can thus take place as a form of molecular aggregates. This aggregation can delay particle-distribution, resulting in slower release from the hydrogel (Fig. 6). Therefore, DEX released from the nanoparticle takes a longer time to diffuse through the hydrogel than the system of DEX in free NPs. This can also be attributed to limited mobility of the NPs and the phase distribution process in the hydrogel. In the free NPs floating in the PBS solution, approximately 90% of DEX was released in 2 weeks whereas in the NPs entrapped in the hydrogel matrix, 80% of DEX was released in 3 weeks regardless of the concentration of the alginate hydrogel. The release profile of DEX also was measured at different temperatures, 25 and 37.5 °C (Fig. 7). At higher temperatures DEX was released somewhat faster; however, the difference was not large and both had the same degree of initial burst effect.

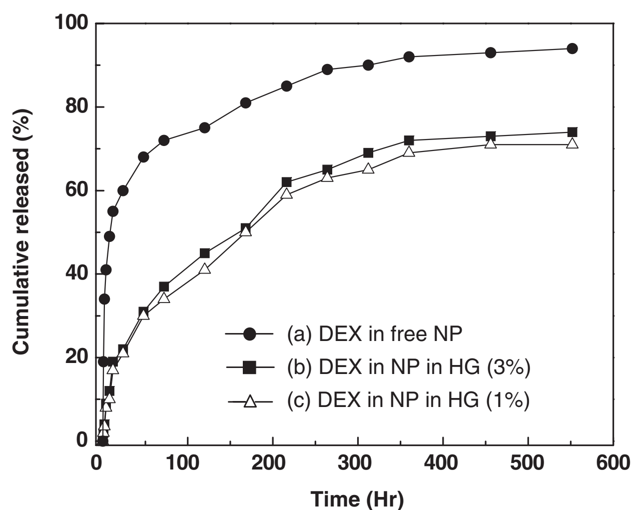

Fig. 5. The releases of dexamethasone from either (a) free NPs or NPs entrapped within hydrogels with different alginate concentrations ((b) 3% and (c) 1%, respectively) are illustrated ($n = 3$).

3.3. In-vivo impedance measurement

The in-vivo impedance measurements of implanted electrodes in the auditory cortex of the guinea pig are shown in Fig. 8. The impedance amplitude of DEX-loaded electrodes was maintained at about the level found in the initial stage of testing, while that of control electrode increased by 3 times about 2 weeks after implantation until it stabilized at approximately 3 MΩ. This value is similar to that routinely observed for chronically implanted bare electrodes [22,23]. This increase of impedance of the control electrodes is likely due to low conductivity of the ensheathing gliotic layers caused from inflammation around the implant. Our observations suggest that the

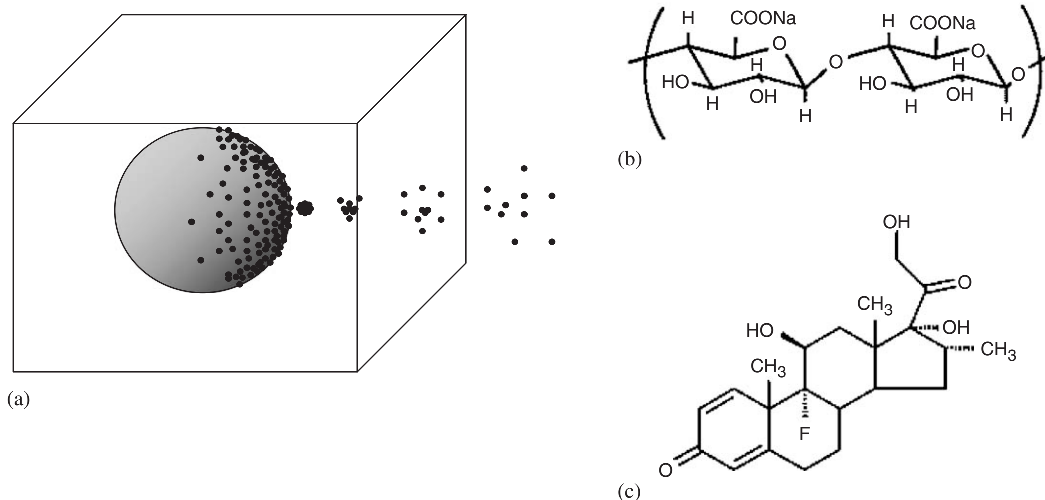

Fig. 6. (a) The schematic of release mechanism of encapsulated substance in the micro/NPs through the pore of hydrogel network (b) and chemical structure of sodium alginate which used for hydrogel matrix (c) and DEX used for releasing agent.

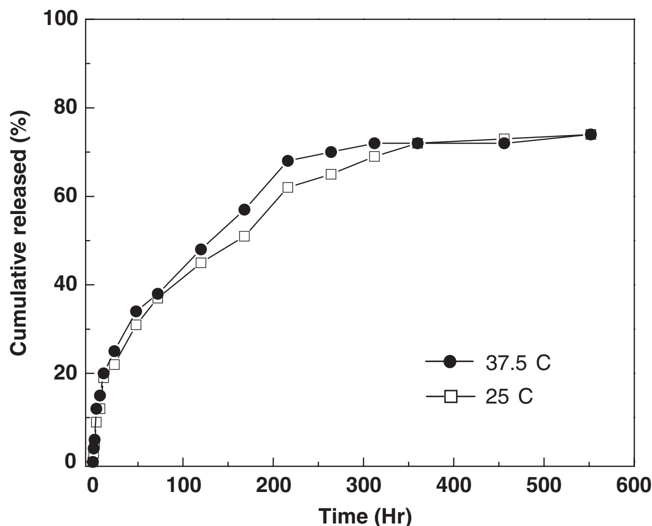

Fig. 7. The release of DEX from NPs entrapped within hydrogels with different surrounding temperatures is illustrated ($n = 3$).

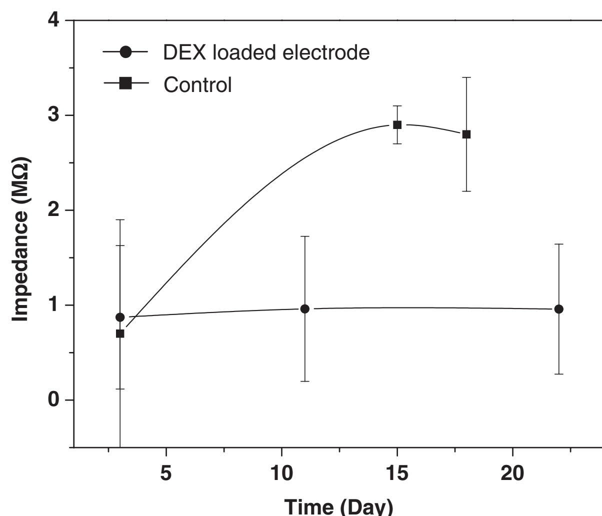

Fig. 8. In-vivo impedance spectroscopy of DEX-loaded hygdrogel coatings chronically implanted in the auditory cortex in the guinea pig as a function of time. The probe coated with a DEX-loaded hydrogel shows no significant change in impedance with time, whereas the control probe (no coating) shows a substantial increase in impedance. This response is due to the limited reactive response around the electrode in the presence of the anti-inflammatory agent, DEX.

limited reactive response around the DEX-loaded electrode has maintained the initially low impedance of the electrodes. Although we have not yet tested the electrical properties of hydrogel coatings alone in animals, we have been looking at the histology of unmodified hydrogel-coated electrodes in collaboration with Prof. Patrick Tresco at the University of Utah and this will be reported in near future. In these experiments, we found that the hydrogel coating itself reduces inflammatory reaction through mechanical buffering effect between stiff electrodes and soft brain tissue. We expect that the ability to minimize the impedance of the electrode will maximize the probability of obtaining high quality neural recordings during chronic recordings.

4. Conclusions

A PLGA-*co*-polymer nanoparticle formulation was developed that utilized an oil-in-water emulsion technique. The procedure efficiently incorporated hydrophobic molecules into nanoparticles, and these could be successfully embedded into the hydrogel coatings deposited the neural electrodes. NP-loaded hydrogel-coated electrodes did not show a significant increase of the impedance as compared with unmodified controls indicating that the NPs entrapped in the hydrogel did not hinder electrical transport and could be used for neural prosthetic applications. Sustained release has been observed from NPs loaded with DEX for up to 3 weeks, with a relatively mild initial burst in case of NPs entrapped in the hydrogel presumably due to buffering effect of hydrogel. The impedance amplitude of DEX-loaded electrodes was maintained about its initial level while that of the control electrode increases by 3 times about 2 weeks after implantation. Thus nanoparticles entrapped in the hydrogel coatings represent a means for the local administration of therapeutic agents for neural electrodes.

Acknowledgments

The authors would like to acknowledge Mr. Jim Wiler (Otolaryngology, University of Michigan) for the help provided with surgical procedures. This work was supported by the National Institute of Health-NINDS-N01-1-2338 and the National Science Foundation-DMR-0084304.

References

[1] Szarowski DH, Andersen MD, Retterer S, Spence AJ, Isaacson M, Craighead HG, et al. Brain. Responses to micro-machined silicon devices. Brain Res 2003;983(1–2):23–35.
[2] Moxon KA, Leiser SC, Gerhardt GA, Barbee KA, Chapin JK. Ceramic-based multisite electrode arrays for chronic single-neuron recording. IEEE Trans Biomed Eng 2004;51(4):647–56.
[3] Turner JN, Shain W, Szarowski DH, Andersen M, Martins S, Isaacson M, et al. Cerebral astrocyte response to micromachined silicon implants. Exp Neurol 1999;156(1):33–49.
[4] Edell DJ, Toi VV, McNeil VM, Clark LD. Factors influencing the biocompatibility of insertable silicon microshafts in cerebral-cortex. IEEE Trans Biomed Eng 1992;39(6):635–43.
[5] Rousche PJ, Pellinen DS, Pivin DP, Williams JC, Vetter RJ, Kipke DR. Flexible polyimide-based intracortical electrode arrays with bioactive capability. IEEE Trans Biomed Eng 2001;48(3):361–71.
[6] Henze DA, Borhegyi Z, Csicsvari J, Mamiya A, Harris KD, Buzsaki G. Intracellular features predicted by extracellular recordings in the hippocampus in vivo. J Neurophysiol 2000;84(1):390–400.
[7] Zhong YH, Yu XJ, Gilbert R, Bellamkonda RV. Stabilizing electrode–host interfaces: a tissue engineering approach. J Rehab Res Dev 2001;38(6):627–32.
[8] Stensaas SS, Stensaas LJ. Histopathological evaluation of materials implanted in cerebral-cortex. Acta Neuropathol 1978;41(2):145–55.

[9] Biran R, Martin DC, Tresco PA. Neuronal cell loss accompanies the brain tissue response to chronically implanted silicon microelectrode arrays. Exp Neurol 2005:115–26.

[10] Shain W, Spataro L, Dilgen J, Haverstick K, Retterer S, Isaacson M, et al. Controlling cellular reactive responses around neural prosthetic devices using peripheral and local intervention strategies. IEEE Trans Neural Syst Rehab Eng 2003;11(2):186–8.

[11] Retterer ST, Smith KL, Bjornsson CS, Neeves KB, Spence AJH, Turner JN, et al. Model neural prostheses with integrated microfluidics: a potential intervention strategy for controlling reactive cell and tissue responses. IEEE Trans Biomed Eng 2004;51(11): 2063–73.

[12] Guzman LA, Labhasetwar V, Song CX, Jang YS, Lincoff AM, Levy R, et al. Local intraluminal infusion of biodegradable polymeric nanoparticles—a novel approach for prolonged drug delivery after balloon angioplasty. Circulation 1996;94(6):1441–8.

[13] Panyam J, Labhasetwar V. Sustained cytoplasmic delivery of drugs with intracellular receptors using biodegradable nanoparticles. Mol Pharmaceut 2004;1:77–84.

[14] Rosler A, Vandermeulen GWM, Klok HA. Advanced drug delivery devices via self-assembly of amphiphilic block copolymers. Adv Drug Deliv Rev 2001;53(1):95–108.

[15] Jain RA. The manufacturing techniques of various drug loaded biodegradable poly(lactide-*co*-glycolide) (PLGA) devices. Biomaterials 2000;21(23):2475–90.

[16] Anderson JM, Shive MS. Biodegradation and biocompatibility of PLA and PLGA microspheres. Adv Drug Deliv Rev 1997;28(1):5–24.

[17] Lee KY, Mooney DJ. Hydrogels for tissue engineering. Chem Rev 2001;101(7):1869–79.

[18] Kim D- H, Abidian M, Martin DC. Conducting polymers grown in hydrogel scaffolds coated on neural prosthetic devices. J Biomed Mater Res 2005;71A:577–85.

[19] Song CX, Labhasetwar V, Murphy H, Qu X, Humphrey WR, Shebuski RJ, et al. Formulation and characterization of biodegradable nanoparticles for intravascular local drug delivery. J Control Release 1997;43(2–3):197–212.

[20] Ogawa Y, Yamamoto M, Okada H, Yashiki T, Shimamoto T. A new technique to efficiently entrap leuprolide acetate into microcapsules of polylactic acid or copoly(lactic glycolic) acid. Chem Pharmaceut Bull 1988;36(3):1095–103.

[21] Fessi H, Puisieux F, Devissaguet J, Ammoury N, Benita S. Nanocapsule formation by interfacial deposition follwoing solvent displacement. Int J Pharmaceut 1989;55:1–4.

[22] Vetter RJ, Williams JC, Hetke JF, Nunamaker EA, Kipke DR. Chronic neural recording using silicon-substrate microelectrode arrays implanted in cerebral cortex. IEEE Trans Biomed Eng 2004;51(6):896–904.

[23] Cui XY, Wiler J, Dzaman M, Altschuler RA, Martin DC. In vivo studies of polypyrrole/peptide coated neural probes. Biomaterials 2003;24(5):777–87.