Leading to the world of tomorrow **FIGURE 1**

Bio / Medical

• **Technical Review** Intelligent Particle for Mechanical Reinforcement and Suppressed Inflammation of Biodegradable Polymers

Materials / Systems

Technical Review
 Magnetic-Field-Controlled
 Reconfigurable Semiconductor
 Logic

Energy / Environment

Technical Review
 Environmental Nanobiosensors
 Based on Aptamer Technology

Korea Institute of Science and Technology

Contents

Foreword	
Bio / Medical	
Technical Review Intelligent Particle for Mechanical Reinforcement and Suppressed Inflammation of Biodegradable Polymers	
Feature Articles The New Era of Total Hip Joint Replacements Multi-Reaction Monitoring of 12 Peptides for Lowered Immunity Screening Stem Cell Recruitment and Angiogenesis of Neuropeptide Substance P Coupled with Self-Assembling Peptide Nanofiber in a Mouse Hind Limb Ischemia Model	12 19 24
Materials / Systems	
Technical Review Magnetic-Field-Controlled Reconfigurable Semiconductor Logic	
Feature Articles High-Strain Air-Operable Soft Transducers Produced from Nanostructured Block Copolymer Ionomer/Silicate/Ionic Liquid Nanocomposite Membranes Enhanced Oxygen Diffusion in Epitaxial Lanthanum Strontium Cobaltite Thin Film Cathodes for Micro Solid Oxide Fuel Cells Shape Control of Lanthanide Ion-Doped Inorganic Upconversion Nanophosphors	35 44 51
Energy / Environment	
Technical Review Environmental Nanobiosensors Based on Aptamer Technology	
Feature Articles Polycrystalline Nanowires of Gadolinium-Doped Ceria via Random Alignment Mediated by Supercritical Carbon Dioxide Facile Growth of Aligned WO ₃ Nanorods on FTO Substrate for Enhanced Photoanodic Water Oxidation Activity Supported Core@Shell Electrocatalysts for Fuel Cells: Close Encounter with Reality	69 75 81
Research Highlights	
KIST News	
A Sign of the Times	102
Interview	104

Foreword

Many parts of the world continued to experience extreme climate conditions this year. In Western Europe, where memories of the many human casualties resulting from the scorching summer of 2003 are still fresh, another summer of abnormally high temperatures caused immense damage in economic, environmental and human terms. Numerous places in China were hit with severe flooding due to a rainy season with substantially higher rainfall than average, while inland regions experienced severe drought during the same period.

Korea was no exception. We experienced record-breaking summer heat again this year, part of a trend that is causing average temperatures in our country to increase each year. As a result, Korea continues to confront national emergencies when the supply of electricity fails to meet

demand for cooling and industrial needs during the summer.

Key public institutions and government offices in Korea are taking aggressive steps to save energy in response to government policies for overcoming power shortages. Here at KIST, we are putting the utmost effort into using research equipment more efficiently and taking other measures to reach a goal of a 15-20% reduction in energy consumption from current levels.

This is just one of the ways that the entire staff and management at KIST are working to support President Park Geun-hye's policies to bring about improvements to the security and well-being of the people of this great nation. We take our social responsibilities very seriously, as befits the oldest national science and technology research institute in the country.

This issue of *KISToday* highlights just a few of the many important areas of research in which we are engaged. You'll see that we are constantly looking for innovative ways to respond to new realities, such as climate change. We appreciate your continued interest in KIST.

Dr. Kil-Choo Moon President of KIST [Technical Review]

Intelligent Particle for Mechanical Reinforcement and Suppressed Inflammation of Biodegradable Polymers



Chang Hun Kum Center for Biomaterials rmackdgjs @kist.re.kr



Yoon Ki Joung Center for Biomaterials ykjoung@kist.re.kr



Dong Keun Han Center for Biomaterials dkh@kist.re.kr

Synthetic biodegradable and biocompatible polyesters, such as poly(Llactide) (PLLA), polyglycolide (PGA), and poly(ɛ-caprolactone) (PCL), are useful materials for medical applications such as surgical implants, drug delivery, and tissue engineering. Although these polymers are suitable for a number of the above-mentioned applications because they are a wellprocessable, biocompatible, and biodegradable polymer, they have two critical shortcomings that restrict their applications including vulnerable inflammatory response and relatively poor mechanical property [1].

Especially, PLLA-based materials have frequently showed sustained inflammatory response upon implantation in the body. PLLA undergoes hydrolytic degradation via the bulk erosion mechanism by the random scission of the ester backbone. It degrades into lactic acid (LAc), a normal human metabolic by-product, which is broken down into water and carbon dioxide via the cycle of citric acid. Inflammatory response and cytotoxicity may be severe under the condition in which the acidic product accumulates and is concentrated. It becomes the cause of inflammation and cytotoxicity [2].

Herein, we propose to utilize magnesium hydroxide, Mg(OH)₂, as a reinforcing material for a composite of PLLA. Magnesium hydroxide is not only known to be a biodegradable and biocompatible material, but also can be grafted by oligolactide (OLA) on the surface through the hydroxyl group [3]. Therefore, the use of magnesium hydroxide with basic pH as a blending partner will be able to mechanically reinforce the PLLA matrix and, more



Figure 1. Schematic expression of suppressive effect of PLLA/Mg-OLA composite on inflammatory response caused by PLLA degradation when PLLA matrix is implanted into the body.

importantly, greatly suppress inflammatory response due to the pH neutralization effect on degraded acidic LAc from PLLA matrix

OLA-grafted magnesium hydroxide (Mg-OLA) was synthesized in order to: (1) neutralize acidic substances generated from hydrolysis of ester bond, and (2) enhance mechanical properties of Mg-OLA. We suggest for the first time a novel composite systems composed of PLLA and Mg-OLAs that can overcome the drawbacks caused by inflammatory response (Figure 1) and poor mechanical properties of a PLLA matrix.

As shown in Scheme 1, L-LA was polymerized on the surface of magnesium hydroxide through ROP resulting in the formation of Mg-OLAs. The PLLA composite films were prepared by blending PLLA matrix and the synthesized Mg-OLAs.

The FTIR spectra demonstrated the typical structural features of PLLA and Mg-OLAs (Figure 2). The sharp peak at 3693 cm⁻¹ indicates O-H stretching vibration in



Figure 2. FTIR spectra of Mg(OH)₂, PLLA, and Mg-OLAs.



Figure 3. Thermogravimetric diagram of Mg(OH)₂, PLLA, and Mg-OLAs.

the crystal structure of $Mg(OH)_2$. Strong absorption band at 1751 cm⁻¹ showed in Mg-OLAs, corresponding to the C=O stretching vibration of carbonyl groups in OLA. The



Scheme 1. Synthesis of Mg-OLAs by ring opening polymerization

Code	Mg20-OLA80	Mg50-OLA50	Mg60-OLA40	Mg70-OLA30	Mg80-OLA20
Feed amount of Mg(OH) ₂ (g)	20.0	50.0	60.0	70.0	80.0
Yield (%)	22.5	79.7	84.3	88.9	92.3
Amount of Mg (g)	44.5	62.7	71.1	78.7	86.6
Amount of PLLA ^[a] (g)	55.5	37.3	28.9	21.3	13.4
Amount of PLLA ^[b] (g)	58.4	32.1	26.4	19.4	10.2

Table 1. Preparation of OLA-Grafted Magnesium Hydroxides.

[a] OLLA or ODLA content (%) = [Total product (g) Used Mg(OH)2 (g)] / Total product (g) × 100

[b] Calculated from TGA data

C-O stretching vibrations from ester bond were observed at 1180, 1129, and 1082 cm⁻¹. The bands at 2996 and 2877 cm⁻¹ were assigned to the C-H asymmetric and symmetric stretching vibrations of CH₃ groups, which the bending vibration of the corresponding group was observed at 1454 cm⁻¹. The band at 2948 cm⁻¹ is attributed to the stretching vibration of C-H groups in PLLA, which the bending vibrations appear at 1382 and 1358 cm⁻¹. Mg-OLAs showed similar IR spectra to that of PLLA.¹⁶ Therefore, FTIR results demonstrate the grafting of OLA on the surface of Mg(OH)₂.

Thermal properties of Mg(OH)₂, PLLA, and Mg-OLAs were measured by TGA (Figure 3). It is known that the thermal decomposition of PLLA starts from below 300°C and is completed around 400°C, that a similar pattern was also observed in this study. The weight loss of Mg(OH)₂ showed at 404°C. The weight loss of Mg-OLAs started at 280 and 400°C, respectively. The first weight loss indicates the decomposition of OLA chains that were modified on the surface of magnesium hydroxide because pure PLLA decomposes at 270-280°C. As shown in TGA, the modification ratio of OLA in Mg-OLAs decreased as increasing the amount of Mg(OH)₂. The OLA contents on magnesium hydroxide could be determined and the results are listed in Table 1. The second weight loss indicates the decomposition of magnesium hydroxide around 400°C.

In addition, the hydrodynamic particle size and the distribution of Mg-OLAs were measured. Particle size distribution of Mg(OH)₂ and Mg-OLAs was shown in



Figure 4. Particle size distributions of Mg(OH)₂ and Mg-OLAs.

Figure 4. The average particle size of pure magnesium hydroxide was 583 ± 31.1 nm. Furthermore, the grafting of OLA made Mg-OLAs larger and the degree of the increment was in proportion to the OLA content (Mg80-OLA20; 634 ± 37.7 , Mg70-OLA30; 588 ± 47.1 , Mg60-OLA40; 608 ± 33.8 , Mg50-OLA50; 721 ± 41.2 , and Mg20-OLA80; 877 ± 57.6 nm).

Morphological changes of various Mg-OLAs were also observed in SEM image. It seems to be the Mg-OLAs were larger than magnesium hydroxide of the initial state (~50 nm) due to the aggregation of Mg-OLAs as shown in Figure 5. With increasing the amount of L-LA (from 0 to 80 wt%; Mg(OH)₂, Mg80-OLA20, Mg50-OLA50, and Mg20-OLA80), the surface of magnesium hydroxide with flat shape has become a corona-like structure (Figure 5a and 5d).



Figure 5. SEM images of magnesium hydroxides and Mg-OLAs: (a) $Mg(OH)_2$, (b) Mg80-OLA20, (c) Mg50-OLA50, and (d) Mg20-OLA80. The magnitude of images is $\times 100,000$.

Mechanical properties of PLLA films containing magnesium hydroxide or Mg80-OLA20 (from 0 to 20 wt%) were investigated. Mg80-OLA20 was shown significantly different mechanical changes to PLLA film than unmodified magnesium hydroxide. With increasing the amount of Mg80-OLA20 to 20 wt%, the tensile strength gradually increased from 26.4 to 39.3 MPa (Figure 6a). In contrast, the tensile strength of PLLA film containing magnesium hydroxide decreased to 11.7 MPa with increasing the amount of Mg(OH)₂. Compared to PLLA film without a Mg(OH)₂ particle (21.4 MPa), PLLA/ Mg80-OLA20 showed two-fold higher tensile strength, whereas PLLA/magnesium hydroxide showed two-fold lower value. As a result, incorporating 20 wt% of Mg80-OLA20 made the tensile strength four-folds higher than



Figure 6. (a) Tensile strength, (b) tensile modulus and (c) elongation of PLLA/Mg(OH)₂ and PLLA/Mg80-OLA20 composites as a function of the amount of Mg(OH)₂ and Mg80-OLA20, and (d) relative yield stress versus volume fraction of PLLA/Mg(OH)₂ (10 wt%) and PLLA/Mg80-OLA20 (10 wt%).

the same content of magnesium hydroxide. Similarly, the tensile modulus also increased ten-fold by blending Mg80-OLA20 with PLLA film (Figure 6b). Although the tensile modulus of PLLA/magnesium hydroxide increased by two-fold, the deviation of the tensile modulus between two materials was quite large. Therefore, these results demonstrate that Mg80-OLA20 significantly reinforces the mechanical properties of PLLA matrix, contrary to unmodified magnesium hydroxide, which means enhanced stiffness. This is thought to be a considerable increase in compatibility of PLLA and Mg-OLAs via the hydrophobic interaction.

However, the excess amount of Mg80-OLA20 weakened the mechanical properties of PLLA matrix in which the decrement in the tensile strength and modulus was measured at over 30 wt% of Mg80-OLA20 (data not shown). Elongation was strongly influenced by blending of magnesium hydroxide or Mg80-OLA20 (Figure 6c). However, there were nearly no differences between both materials in elongation at break. Incorporation of magnesium hydroxide or Mg80-OLA20 made PLLA matrix stiffer and decreased elasticity. Especially, the elongation of the PLLA/Mg(OH)₂ and PLLA/Mg80-OLA20 composite was similarly deteriorated at the above 10 wt%. The similar reductions in the elongation are mainly due to the inorganic property of magnesium hydroxide.³⁵ However, the elongation of matrix at below 10 wt% appeared highly more when it was blended with Mg80-OLA20 than when it was blended with Mg(OH)₂. It seems to be due to OLA modification on the surface of magnesium hydroxide.

The interfacial interaction parameter (B value) shows how much Mg80-OLA20 can reinforce the mechanical properties of PLLA matrix. Therefore, the interfacial interaction of PLLA containing typically 10 wt% of Mg80-OLA20 was calculated by the following Equation 1.

$$\sigma_{\rm c} = (1 - \varphi)/(1 + 2.5\varphi) \times \sigma_0 \times e^{B\varphi} \tag{1}$$

Here, σ_c is a yield stress with various contents of Mg80-OLA20 and σ_0 is a yield stress of the PLLA film.



Figure 7. pH changes in solutions of (a) Mg-OLAs (Mg20-OLA80, Mg50-OLA50, Mg60-OLA40, Mg70-OLA30 and Mg80-OLA20) and PLLA, and (b) PLLA/Mg80-OLA20 composites with different amounts of Mg80-OLA20 (0, 2, 10, 20, 30, 40, and 50 wt%) at a harsh condition (80°C and 100 rpm) for 21 days.

In addition, ϕ is a volume fraction of PLLA/Mg80-OLA20 film. An increment in the B value means that there exists a stronger interaction at the interfaces. As shown in Figure 6d, excess magnesium hydroxide content improved the relative yield stress of the composites. Based on this relationship, the B value of Mg80-OLA20 contained PLLA film (5.26) was remarkably higher than that of unmodified magnesium hydroxide contained PLLA film (0.09). This demonstrates that OLA-grafting on the surface of magnesium hydroxide enhances the interfacial interactions between PLLA matrix and Mg-OLA, and thereby reinforces mechanical properties of PLLA matrix.

Figure 7 shows pH changes of PLLA, Mg-OLAs,

and PLLA/Mg80-OLA20 composites for 21 days. The degradation of PLLA was accelerated under a harsh condition (80°C and 100 rpm), resulting in decrement of pH to 2.9 for 21 days. The decreased pH is due to the generation of LAc as a degradation by-product. However, all of Mg-OLAs presented a higher pH levels than pure PLLA, and the pH values increased with increasing the amount of magnesium hydroxide in Mg-OLA matrix (Figure 7a). This result indicates that magnesium hydroxide effectively neutralizes the acidic pH that was caused by surface-grafted OLA. Based on this result, the neutralizing effect of Mg80-OLA20 incorporated into the PLLA films was investigated at the different amounts of Mg80-OLA20 (Figure 7b). Increasing the portion of Mg80-OLA20 from 0 to 50 wt% resulted in increase in the pH level from 3.0 to 8.5, which was roughly in proportion to the portion of Mg80-OLA20. This can provide the information on the amount of Mg-OLA that is effective for neutralizing acidic pH caused by a known amount of PLLA matrix.

The cell viability and the expression levels of COX-2 and IL-6 of Mg80-OLA20 in PLLA matrix were investigated. In vitro HUVEC culture was carried out with LAc and Mg80-OLA20 to confirm each effect on cell viability (Figures 8a and 9a). Mg80-OLA20 seemed to have increasing effect on HUVEC viability in 0-30 wt% while LAc seemed to have considerable cytotoxic effect depending on the concentration. As compared to the control, the addition of LAc to HUVEC resulted in the significant decrements in a viability of over 50%. This means that LAc, a hydrolysate of PLLA, has severe cytotoxic effect on HUVEC. When Mg80-OLA20 and hydrolysate of PLLA were added to the media, the instant recovery of cell viability was observed. The addition of 10 wt% of Mg80-OLA20 significantly increased cell viability to 80%. For over 20 wt% of Mg80-OLA20, cell viability approached about 100%. This result demonstrates that Mg80-OLA20 can compensate the cytotoxic effect of LAc degraded from PLLA.

The expression level of COX-2 protein, an inflammatory marker, during HUVEC culture was

determined by Western blot analysis in order to confirm the correlation between the expression level of COX-2 and the amount of LAc (Figure 8b). When acrolein (ACR, positive control) was used to induce the inflammatory response of HUVEC, COX-2 protein level increased considerably as compared to the normal condition. This means that inflammation was definitely caused at the concentration of 25 μ M. Similarly, the addition of LAc to the culture media induced COX-2 protein expression. Although it was barely observed at the LAc concentration of 0.325 mM, it gradually increased from 0.65 to 2.6 mM. At the LAc concentration of 2.6 mM, the COX-2



Figure 8. (a) Cell viability and expression levels of (b) COX-2 and (c) IL-6 protein when various concentrations of LAc itself (mM) were added to the HUVEC culture media.

level was markedly elevated as compared to two times lower concentration. This indicates that the induction of inflammatory reponse in HUVEC culture depends on LAc concentration. However, the expression level of COX-2 protein dramatically decreased at the concentrations of 7.8 and 26.1 mM. This is probably attributed to significantly decreased cell viability as compared to lower concentrations (Figure 8a). Although COX-2 protein level was marginal due to decreased cell number, three-fold higher concentration (7.8 mM) of LAc might induce much lower expression of COX-2 protein, compared to 2.6 mM.

The expression level of COX-2 protein was determined at HUVEC culture media to which effluent from PLLA matrices embedded with different amounts of Mg80-OLA20 was added (Figure 9). The expression of COX-2 protein by ACR was also confirmed in this experiment. The expression of COX-2 protein was up-regulated by using 10-20 wt% of Mg80-OLA20. In contrast, COX-2 protein was almost not detected with 30-50 wt% portion of Mg80-OLA20. This result indicates that more than 30 wt% of Mg80-OLA20 can significantly suppress the expression of COX-2 protein from HUVEC. This correlates well with the pH neutralizing effect depending on the weight portion of Mg80-OLA20 (Figure 7b). More specifically, 10-30 wt% of Mg80-OLA20 presented lower pH values than physiological pH (7.4), whereas 40 and 50 wt% of Mg80-OLA20 revealed slightly higher pH levels than pH 7.4. In a literature, it was suggested that the use of COX-2 inhibitors is effective for preventing vascular inflammation.

Besides the anti-inflammatory effect of Mg-OLA caused by the PLLA hydrolysate, the expression level of IL-6 protein as a proinflammatory marker was also investigated by ELISA. The expression level of IL-6 protein showed similar tendency to the expression level of COX-2 protein. As a result, the expression level of IL-6 protein increased depending on the LAc concentration in the limited range, from 0.325 to 2.6 mM of LAc, however the expression level of IL-6 protein decreased at higher concentrations of LAc. This means that cell death was



Figure 9. (a) Cell viability and expression levels of (b) COX-2 and (c) IL-6 protein in the HUVEC culture after the addition of effluents from PLLA composites with different concentrations of Mg80-OLA20 (0-50 wt%).

rapid before IL-6 protein was expressed at concentrations above that of LAc 2.6 mM (Figure 8c). Similarly, adding Mg80-OLA20 and the hydrolysate of PLLA in the same media, the expression level of IL-6 protein decreased gradually at 20-50 wt% of Mg80-OLA20 (Figure 9c). However, the expression level of IL-6 protein from Mg80-OLA20 was lower than negative control. It seems that the expression level of IL-6 protein was suppressed by pH neutralization. We tried to investigate the cell viability and the expression levels of COX-2 and IL-6 on the PLLA/ Mg-OLA composites. It is suggested that the composite system composed of PLLA/Mg80-OLA20 (above 30 wt%) can overcome drawbacks caused by cytotoxicity and inflammatory response.

Consequently, the use of biodegradable Mg-OLAs as blending additive for PLLA matrix could reinforce the mechanical properties as well as suppress inflammatory response. Such hybrid composite system blending with oligomer-grafted magnesium hydroxide in biodegradable polymers would be a promising strategy for improving poor mechanical properties and preventing severe inflammation, in the use of biodegradable polyesters for biomedical applications including vascular stents, tissueengineered scaffolds and biodegradable implants.

References

- Anderson JM, Shive MS. Adv Drug Deliv Rev 1997; 28: 5-24.
- [2] van der Giessen WJ, Lincoff AM, Schwartz RS, van Beusekom HMM, Serruys PW, Holmes DR, Ellis SG, Topol EJ. Circulation 1996; 94: 1690-1697.
- [3] Giorgi R, Bozzi C, Dei L, Gabbiani C, Ninham BW, Baglioni P. Langmuir 2005; 21: 8495-8501.



Intelligent Biomedical Materials Laboratory

Feature Articles

The New Era of Total Hip Joint Replacements



Yu-Chan Kim Center for Biomaterials chany@kist.re.kr







Hyung-Seop Han Center for Biomaterials

hyuhan@kist.re.kr

Introduction

Human life expectancy has been significantly increased due, in part, to advancements in medical technology, so the short lifespan of implanted prosthetics has become a critical issue in the use of joint replacements. The useful life of total hip joint replacements (THRs) has typically been about 15 years for patients who receive the surgery before the age of 65 [1]. However, with the lengthening of the human lifespan and increasing numbers of younger patients affected by traumatic sports injuries and accidents, multiple revision surgeries are much more common; these surgeries are not only expensive but have a low success rate [2]. THRs consist of a polymerlined acetabular cup, a metallic femoral head, and a stem, as schematically depicted in Figure 1. Of these components, the polymer liner and metal head comprise a bearing couple for the prosthesis, and the friction and wear properties of the bearing are critical factors in the longevity of the implant [3]. The release of very small particles of wear debris from the bearing over a long period of time may cause osteolysis and inflammatory reactions. In addition, generation of this debris can ultimately result in the loosening and eventual failure of the implant [4].

In search of improvements to current prosthesis design, we developed a composite coating to overcome the drawbacks of previously developed coating materials. By creating hybrid materials from certain metals and ceramics, it was possible to make materials having both the hardness



Figure 1. Schematic description of a THR.

of ceramic and toughness of metal. Because these composite coatings combine the mechanical properties of hard ceramic and tough metal materials, wear properties and fracture toughness of the composite are superior to pure metals and alloys, such as Ti and the Co-28Cr-6Mo alloy. On the basis of both their biocompatibility test results and enhanced mechanical properties, the composite coatings developed in this study have great potential for use as bearing materials in THRs.

1. Fabrication of composite coating

A reactive plasma spraying method was selected to deposit the composite coatings. The plasma spraying process is known to be a reliable and cost-effective technique for depositing structural and functional coatings of various materials on a wide range of substrates [5]. The coating was Ti-based, but a Ti-TiN laminate composite was eventually formed by controlling atmospheric gases during the plasma spraying process. Three different coatings were deposited via plasma spraying on 304 stainless steel (SUS304) for microstructural surface analysis and a wear test. Nontoxic Ti powders (99.95%, LTS Chemical, Inc.) with an average particle size of approximately 35-43 µm (-325 mesh) were sprayed under a controlled atmosphere of either Ar (99.999%) or N₂ (99.999%) gas in a spraying chamber. During plasma spraying, the flow rate of N₂ gas in the chamber was controlled at 25 and 1000 L/min. The different processing conditions are summarized in Table 1. Prior to plasma spraying, the Ti powders were dried at 80°C for 30 min in a forced convection oven. Sand-blasting of the substrate surface was performed prior to plasma spraying to enhance coating adhesion. To compare the wear properties of the coatings with Co-Cr alloys, we produced the Co-28Cr-6Mo alloy, which is used most extensively, via arc melting.



Figure 2. XRD result of the plasma-sprayed coatings.

 Table 1. Types of Coatings and the Processing Conditions for Plasma Spraying.

Туре	atmospheric gas	atmospheric gas flow rate (L/min)	current (A)	plasma gas, Ar (L/min)	plasma gas, He (L/min)	spraying distance (mm)	chamber vacuum (torr)
1	Ar	1000	600	36	20	120	1 × 10 ⁻²
2	N ₂	25					
3	N ₂	1000					



Figure 3. (a-c) FE-SEM images of the cross-sectional images of Type 1-3 coatings. (d-f) FE-SEM surface images of Types 1-3 coatings.

Figure 2 shows the results of the XRD analyses of the three different coatings described in Table 1. As can be seen, Type 1 is mostly crystalline α -Ti, similar to the Ti powders, and Types 2 and 3 have both crystalline α -Ti and TiN peaks. In particular, the intensity of the TiN peaks is strongly enhanced in Type 3 as compared with Type 2, which demonstrates that the thickness of the TiN layer in the composite can be controlled by adjusting the amount of N₂ gas when other deposition conditions are fixed (Table 1).

Figures 3a-f show FE-SEM normal and backscattered electron (BSE) images that depict the cross-sectional microstructures and surface morphologies of the three types of coatings. In these coatings, two important microstructural characteristics should be noted: (1) under an N₂ atmosphere, lamellar structures are formed with alternating Ti and TiN layers; (2) Type 2 and 3 coatings are porous as compared with the Type 1 coating, and the pore density increases as the N₂ flow rate increases. The lamellar structures in the coatings were verified by the AES and EPMA elemental mapping results shown in Figure 4. The AES results in Figure 4a indicate that the thicknesses of the Ti and TiN layers in the lamellar structures of Type 2 are similar at approximately 2-5 µm. In Type 3, the thickness of the TiN layer is significantly increased to 10-20 µm (Figure 4b). The Ti layer in Type 3 is not as continuous as in Type 2 and seems to be embedded in the TiN layer. Figure 4b also shows that more pores and cracks are present in Type 3 than in Type 2. In most previously reported cases, titanium-nitride coatings applied using plasma spraying formed a coating layer with a small amount of a titanium-oxide phase. Notably, in the AES and EPMA results, small oxides are formed in the coatings. Figure 5a shows the HR-TEM bright-field image and selected area diffraction patterns (SADPs) of the interface and the adjacent materials of the Type 2 coating. According to Figure 5a, a sound interface with few defects is formed between the polycrystalline TiN (region A) and the polycrystalline Ti (region B). Images b and c in Figure 5 depict the HR-TEM images of the



Figure 4. AES element mapping results of the cross sections of (a) Type 2 and (b) Type 3 coatings.

nanostructured TiN phases in Types 2 and 3, respectively. The rapid cooling during the plasma spraying process resulted in the nano-sized grains observed in the coatings.

2. Mechanical properties and biocompatibility of the composite coatings

The surface properties of the three coatings are summarized and compared with those of the Co-1 28Cr-6Mo alloys in Table 2. According to the measured results reported in this table, the R_{rms} values of Types 1-3 are greater than the R_{rms} value of the Co-28Cr-6Mo alloy. In particular, the R_{rms} values of Types 2 and 3 are two to three orders of magnitude larger than the R_{rms} value of the Co-28Cr-6Mo alloy, as shown in Figure 3. As reported in Table 2, the hardness is greatly enhanced in the composite coatings as compared with the Co-28Cr-6Mo alloy and Type 1 coating, and hardness increases as the thickness of the TiN layer in



Figure 5. (a) HR TEM image and SADP of the interface and adjacent materials of Type 2. (b) A nano-size grain of TiN layers in Type 2 and the SADP from the grain. (c) A nano-size grain of TiN layers in Type 3.

the coating increases. However, the coating hardness standard deviations for Types 2 and 3 are also greater than those of the other two samples because of their lamellar structure and inhomogeneous pore distributions. The values of θ measured for Types 1-3 are smaller than that measured for the Co-28Cr-6Mo alloy, but there is a negligible difference among the θ values of the three coatings. The value of θ is a function of the surface area as well as the surface energy [6]. Because coating Types 1-3 are more porous than the Co-28Cr-6Mo alloy, the effective surface area that contacts the lubricant is increased. Based on these results and the results reported in Table 2, the coating surfaces of Types 1-3 appear to be more hydrophilic than the Co-28Cr-6Mo alloy because of the pores on their surfaces.

The wear test results (μ and k) are compared in Figure 6 and Table 2. Figure 6 presents the observed variations in μ as the sliding distance increases, and the average μ values are summarized in Table 2. According

samples	R _{rms} (nm)	hardness (StDev*) (Hv)	θ (deg)	μ (StDev*)	k (× 10 ⁻¹⁵ kg/Nm) (StDev*)
Co-28Cr-6Mo	3.0	365.6 (26.7)	73.8	0.027 (0.002)	21.2 (1.8)
Type 1	15.5	270.1 (28.1)	64.1	0.028 (0.001)	24.6 (2.4)
Type 2	51.9	756.4 (162.8)	63.6	0.035 (0.001)	18.3 (2.2)
Туре З	476.5	1098.5 (214.3)	61.4	0.041 (0.002)	16.7 (2.3)

Table 2. Surface Roughness (R_{ms}), Contact Angle (θ), Friction Coefficient (μ), and Wear Rate (k) of the Three Coatings and the Co–Cr–Mo Alloys. ((StDev*) is the standard deviation).

to Figure 6, the μ values of Types 2 and 3 are higher than those observed for the Co-28Cr-6Mo alloy and Type 1 throughout the applied sliding distance. In particular, the μ value of Type 3 is the highest among the tested samples and is larger than the μ value of the Co-Cr alloy by more than 35% (Table 2). Although the μ values of the Co-28Cr-6Mo alloy and Type 1 increased to 1000 m and then remained almost constant until the end of the test, the μ values of Types 2 and 3 slightly decreased



Figure 6. Friction coefficient with increasing sliding distance and biocompatibility test results for Types 1 and 3 coatings.

as the sliding distance increased. As shown in Table 2, the wear properties of the composite coatings (Types 2 and 3) are greatly enhanced as compared with those of the Co-28Cr-6Mo alloy and Type 1 coating. The value of k for Type 3 is the smallest among the samples and is approximately 25% smaller than the value of k for the Co-28Cr-6Mo alloy. A comparison of the three coatings, Types 1-3, reveals that k is inversely proportional to μ and decreases as the thickness of TiN layer increases although the pore density increases with this thickness.

The biocompatibility test results for coating Types 1 and 3 are summarized in Figure 6. The survival rates of the HeLa cells on Types 1 and 3 were higher than the control, which indicates excellent biocompatibility with no toxicity from the coatings. Type 1 shows a slight increase of cell viability when compared to the 100% control, whereas there was a significant increase of the cell viability in 48 h.

3. Mechanism of enhanced properties

In this study, we successfully developed Ti-TiN composite coatings with fine lamellar structures using reactive plasma spraying with atmospheric control. The plasma generated by high-voltage electrodes creates partially or fully melted Ti particles (droplets) that traverse the plasma jet. The nitride reaction occurs on the surface of the droplets during flight to the substrate under an N_2 atmosphere. Ti droplets with TiN on the surface are deposited on the substrate in a flattened shape, and they rapidly solidify to form an interlocking lamellae (or splats) structure. Figure 7 schematically depicts the plasma spraying and deposition processes. One of the most

beneficial properties of the composite coatings is that a compromise between hardness and toughness can be achieved. Using the plasma spraying process developed in this study, the mechanical properties of the coating can be tailored by varying the thickness of the TiN layer, which is controlled by the N₂ flow rate. As mentioned previously, ceramic coatings have enhanced hardness and wear properties as compared with metal implant materials, but the brittleness of the ceramics limits the application of the coatings. The improved fracture toughness of the composite coatings was also verified in that no fracture or delamination between the coating and the substrate was observed after the wear test. In real systems involving the accumulation of many solidifying droplets, porosity is an inherent problem and is believed to reduce useful properties such as strength and corrosion resistance [7]. The pore density is reduced because the liquid droplets have greater energy to penetrate into the pores. Their energy increases with velocity and droplet density. On the basis of the results presented in Figures 3,4, the coatings become more porous as the thickness of the TiN layer increases, although TiN has a greater density than Ti. This result occurs because the TiN formed on the outer surface of the flying droplets is almost solid because of the high melting point of TiN. As thick TiN is formed on the surface, the impinged droplets are too stiff to penetrate into the pores. However, the Type 3 coating, which is the most porous, and thus has the roughest surface, was found to have the best wear properties against the UHMWPE among the tested samples. UHMWPE wear depends not only on the surface roughness of the counterpart materials but also on the relationship between the lubricant and the pores [8]. The positive effects of the pores in coating Types 2 and 3 on the wear of UHMWPE are as follows: (i) the pores and cavities can act as bovine serum reservoirs, which prevent dry running at the start of the test and reduce the actual contact area and friction losses [9]; and (ii) as the metal surface becomes rougher, the temperature of the contact areas during the wear test can increase. This increased temperature



Figure 7. Schematic descriptions of (a) the plasma-spraying process and (b) the cross-sectional microstructure of the Ti-TiN composite coatings.

may cause denatured proteins from the bovine serum to accumulate more rapidly between the UHMWPEpin and the disk [10]. Ultimately, the accumulation of denatured protein leads to more extensive formation of a protein film adhering to the disk and thus protects the UHMWPE-pin from wear. Studies have shown that small pores worsen the lubricating conditions and multiply the direct metallic contacts in the friction zone [8, 9]. As seen in Figures 3,4, both the pore size and density in Type 3 are greater than those in Type 2. However, more extensive experimental studies are required to determine the optimum distribution, size, and density of pores for optimal wear properties. The chemical analyses in Figures 4,5 also reveal that the coatings are free of oxides and other contaminants that may produce undesirable side effects during operation. Biocompatibility is a critical parameter when considering medical device component material. Thus, the positive biocompatibility test results in Figure 6 indicate that the coatings produced in this study have great potential as bearing materials in THRs.

Closing remarks

The aim of this study was to develop new Ti-based composite coatings with enhanced wear resistance against a UHMWPE liner. Using a plasma spraying technique in a controlled N₂ gas atmosphere, Ti-TiN composite coatings with a lamellar structure of alternating Ti and TiN could be produced, and the relative composition of Ti and TiN could be controlled. Because the composite coatings have mechanical properties combining the properties of hard ceramic and tough metal materials, the combined wear properties and fracture toughness show a distinct improvement over pure metals and alloys, such as Ti and the Co-28Cr-6Mo alloy. Plasma-sprayed coatings generally have a porous structure that has previously been considered a disadvantage. However, our results clearly reveal that a certain level of porosity is highly beneficial to the friction and wear resistance by acting as a lubricant reservoir, and this porosity does not degrade the fracture resistance of the coatings. On the basis of both their biocompatibility test results and enhanced mechanical properties, the composite coatings developed in this study have great potential for use as bearing materials in THRs.

Note

This article and images are cited from "Multifunctional composite coating as a wear-resistant layer for the bearing in total hip joint replacement" in *ACS Appl Mater Interfaces*. 2013 Vol. 5(2), pp. 395-403.

References

- Geetha M, Singh AK, Asokamani R, Gogia AK. Prog. Mater. Sci. 2009; 54: 397–425.
- [2] Raimondi MT, Pietrabissa R. *Biomaterials* 2000; 21: 907–913.

- [3] Ryan G, Pandit A, Apatsidis DP. *Biomaterials* 2006; 27: 2651–2670.
- [4] Kanaga Karuppiah KS, Bruck AL, Sundararajan S, Wang J, Lin Z, Xu ZH, Li X. Acta Biomater. 2008; 4: 1401–1410.
- [5] Hunt JA, Callaghan JT, Sutcliffe CJ, Morgan RH, Halford B, Black RA. *Biomaterials* 2005; 26: 5890–5897.
- [6] Irene EA. Surfaces, Interfaces, And Thin Films for Microelectronics; John Wiley & Sons: New York, 2008.
- [7] Sanmarchi C, Liu H, Lavernia EJ, Rangel RH, Sickinger A, Muehlberger EJ. *Mater. Sci.* 1993; 28 (12): 3313–3321.
- [8] Takadoum J. Material and Surface Engineering in Tribology, John Wiley & Sons: New York, 2008.
- [9] Gradzka-Dahlke M, Dabrowski JR, Dabrowski B. Wear 2007; 263: 1023–1029.
- [10] Lu Z, McKellop H, Liao P, Benya PJ. *Biomed. Mater. Res.* 1999; 48: 458–464.



Integrated Biomaterial Process Laboratory

[Feature Articles]

Multi-Reaction Monitoring of 12 Peptides for Lowered Immunity Screening



Min-Jung Kang Molecular

Recognition Research Center, Future Convergence Research Division mjkang1@kist.re.kr

Oh-Seung Kwon

Doping Control

oskwon@kist.re.kr

Center





Byung-Hwa Jung Molecular Recognition Research Center, Future Convergence Research Division

jbhluck@kist.re.kr

Introduction

The immune system is known to be closely tied to aging, cancer, and infection. Endogenous peptides are known to have a regulatory role in the immune system. We selected 12 target peptides for further study that have previously been shown to be associated with inflammation, cardiac disease, kidney disease, and the elimination of infection. A multi-reaction monitoring method, using high performance liquid chromatography mass spectrometry, was developed for these 12 target peptides to determine endogenous peptide concentration from human serum. BNP (B-type natriuretic peptide), CCK (cholecystokinin), ACTH (7-38), SubP (substance P), NFMLP and VGPIPT were quantified in a concentration range of 0.1 to 50 ng/ml. ACTH (7-38) showed a significantly higher concentration in elderly people while NFMLP was significantly lower in young people (p < 0.0001). This result indicates a possible relationship between ACTH, NFMLP, and lowered immunity.

Body

Since peptidomics was introduced into systems biology by Schulz-Knappe in 1996 [1] and Liliane Schoofs in 2003 [2], its application has been



Figure 1. MRM chromatogram of 12 targeted peptides using the optimized HPLC gradient (gradient 4). The most intensive ion among fragments was selected as the monitoring MRM ion. 40 eV was used as collision energy for all peptides. Val5-ANG I (a), Neurotensin (b), CCK (c), VGPIPT (d), NFMLP (e), oxytocin (f), ANP (g), SubP (h), ACTH (1-17) (i), ACTH (7-38) (j), ACTH (18-39) (k), and BNP (47-76) (i) were measured using the MRM method.

Compound	Precursor ion	Monitoring fragment ion	Compound	Precursor ion	Monitoring fragment ion
[Val5]-Angiotensin	642.0	770.4	ANP(4-18)	532.0	768.3
Neurotensin	837.9	780.6	Substance P	674.3	656.5
CCK	1064.0	1047.0	ACTH (1-17)	524.3	615.3
VGPIPT	717.0	699.5	ACTH (7-38)	732.8	715.5
NFMLP	438.0	432.0	ACTH (18-39)	822.7	981.5
Oxytocin	1007.4	990.4	BNP (47-76)	578.6	654.4

Table 1. The Measured Precursor Ions and Monitoring Ions for Peptides Using LC-MS/MS.

expanded to the discovery of biomarkers, drug targets and lead candidates for novel drugs. Nonetheless, because of many difficulties associated with analysing endogenous peptides, particularly the selective extraction of peptides from complex biological samples, the biomarker discovery of endogenous peptides using peptide profiling is challenging. Therefore, the development of serum extraction methods has been the the main focus of peptidomics research to date, rather than endogenous peptide biomarker discovery. Our study targeted 12 peptides (Val5-ANG I, NT, CCK, VGPIPT, NFMLP, oxytocin, ANP(4-18), SubP, ACTH(1-17), ACTH(7-38), ACTH(18-39), BNP(47-76)) for quantitative analysis from blood (serum or plasma) of young and elderly subjects. Through the comparative analysis of serum samples from these donors, we expected to find regulatory peptides that could indicate lowered immunity during immunosenescence. We adapted liquid chromatography mass spectrometry (LC-MS/MS) for quantitative analysis of peptides because of its simultaneous detection capability. To replace immunoassay with the LC-MS/MS method, sensitivity and ionization efficiency were the main challenges faced. We optimized the extraction method and LC-MS/MS detection parameters and analyzed the young and elderly subjects using this method for quantitative comparison.

MRM method for the targeted 12 peptides

A LC-MS/MS method for the 12 targeted peptides was developed and optimized. Figure 1 shows the MRM chromatogram of the 12 peptides measured by optimized gradient. The fragment ion with the highest intensity was used as a monitoring fragment ion. The same collision energy (40 eV) was used for fragmentation for all peptides. The detected precursor ion and used fragment ion for MRM monitoring are summarized in Table 1. For the analysis of real samples, we analyzed MRM and collected fragmentation patterns of standard solutions at the same time.

The lower limit of detections (LOD) of the target peptides was lower than 10 ng/ml when we analyzed the 12 peptides using MRM mode in one bottle after extraction from the filtered plasma matrix. NFMLP and ACTH (18-39) could be detected at 1 ng/ml. When we analyzed standard solutions in the buffer, the peptides could be detected at a much lower concentration (LOD < 0.1 ng/ml). The mass resolutions were higher than 20,000 at LOD concentrations for all 12 peptides. The selectivity was confirmed for 6 peptides (Val5-ANG I, oxytocin, ACTH (1-17), ACTH (18-39), NT, ANP (4-18)) because these peptides existed below the LOD in young healthy subjects. In the case of the other 6 peptides, the selectivity could not be confirmed. A small amount of endogenous peptides were detected from plasma. The calibration curves for quantitative analysis were made using a standard solution, and therefore, precision and accuracy resulted only from pipetting and LC-MS/MS. The calibration curves made by standard spiked plasma gave an almost "0" slope in the range of 1 - 50 ng/ml of peptides. The endogenous peptide concentrations could be quantified only using calibration curves. For the peptide extraction, 10% formic acid (FA) gave the best results among acidic buffers. The SPE method was also tested using various pH dilution buffers. CCK gave higher recovery with basic pH (10 mM Tris and 0.1 M triethylamine) and BNP (47-76) gave higher recovery with acidic conditions (10, 0.2 % FA). 30% formic acid and 30% HCl were also tested but did not give a high recovery (< 10%). PBS dilution also gave a low recovery (< 10%) for all model peptides (data not shown). Fixed 10% formic acid was used as a dilution buffer and recovery was tested for all 12 target peptides using ultrafiltration (UF) and solid phase extraction (SPE). Some peptides gave better recovery with SPE (Val5-ANG II, NT, CCK, ANP (4-18), NFMLP). Higher recovery was obtained when the UF method was used for extraction of VGPIPT, oxytocin, SubP, ACTH (18-39), and BNP (47-76). ACTH (1-17) and ACTH (7-38) gave similar recovery with SPE and UF. An acidified SPE method was selected for further experiments because of its feasibility for enrichment and mass analysis.

Comparison of 12 endogenous peptides among young and elderly donors

Serum samples from 25 young (< 45 years old) and 23 elderly (> 65 years old) donors were measured for 12 peptide concentrations after acidified SPE extraction. Donor information is summarized in Table 2. The half peptides (BNP, CCK, NFMLP, ACTH (7-38), Sub P,



Figure 2. The 6-peptide profiling of young (25 samples) vs. elderly (23 samples) people using the optimized MRM method. 12 targeted peptides were tested, but 6 of these (BNP, CCK, NFMLP, ACTH (7-38), SubP, and VGPIPT) were detected at higher concentrations than LOQ.

VGPIPT) among the 12 targeted peptides were quantified clearly (higher than LOQ values). The qualitative fragment pattern of the precursor ion was checked for confirmation of each peptide. In the case of NFMLP, the elderly study participants showed concentrations that were 12 fold lower than the young participants. Similarly, the elderly showed a 5 fold higher concentration of ACTH (7-38) than the young. The t-test result (p value) was lower than 0.0001 in both cases. The endogenous peptide concentrations of the samples for the young and elderly donors are shown in Figure 2. The diagnostic criteria for aging was 8.17 (ng/ml) for ACTH and 0.092 (ng/ml) for NFMLP.

Table 2.	Study participant information and the measured concentration
of ACTH	(7-38) and NFMLP from healthy young and elderly participants.

Parameter	Young	Elderly
Number of subjects	25	23
Male/Female	11/14	9/14
Age [years old]	34.2 ± 4.6	70.2 ± 4.6
Conc. of ACTH (ng/ml)	3.50 ± 2.43	20.34 [*] ± 2.58
Conc. of NFMLP (ng/ml)	1.108 ± 0.88	$0.088^{\circ} \pm 0.003$

*P < 0.0001

Criteria for diagnosis is 8.17 for ACTH and 0.092 for NFMLP

We analyzed the different ACTH fragments such as ACTH (1-17) and ACTH (18-39). There were a few samples in which ACTH (1-17) or ACTH (18-39) was detected. ACTH (7-38) seems to be the active fragment released in serum during aging. ACTH is well known as a stress biomarker and NFMLP has been reported to activate the anti-inflammatory system [3,4]. These two marker candidates could be related to lowered immunity because we compared healthy subjects rather than patients with diagnosed illness, and inflammation is strongly related with immunosenescence. The developed MRM method for the analysis of the 12 target peptides was used for a comparison of healthy young and elderly subjects. The elderly subjects showed a lower concentration of NFMLP (p < 0.0001) and a significantly higher concentration of ACTH (7-38) (p < 0.0001) as compared to young subjects. This suggests that these two peptides could be utilized in lowered immunity screening. Further study of the mechanism of the relationship between ACTH stimulation or NFMLP suppression and aging will provide a deeper understanding of the immunosenescence process, but our research does indicate that peptides could possibly be used as screening markers of lowered age-related immunity.

Note

This article and images are cited from "Multi reaction monitoring of 12 peptides for lowered immunity screening" in *Anal. Bioanal. Chem.* 2012; 404: 2249-2258.

References

 Schulz-Knappe P, Raida M, Meyer M, Quellhorst EA, Forssmann WG. *European Journal of Medical Research* 1996; 1 (5): 223-236.

- [2] Baggerman G, Cerstiaens A, De Loof A, Schoofs L. The Journal of Biological Chemistry 2002; 277 (43): 40368-40374.
- [3] Manicone AM, Huizar I, McGuire JK. *The American Journal* of *Pathology* 2009; 175 (6): 2319-2331.
- [4] Trzonkowski P, Mysliwska J, Szmit E, Wieckiewicz J, Lukaszuk K, Brydak LB, Machala M, Mysliwski A. Vaccine 2003; 21 (25-26): 3826-3836.

Feature Articles

Stem Cell Recruitment and Angiogenesis of Neuropeptide Substance P Coupled with Self-Assembling Peptide Nanofiber in a Mouse Hind Limb Ischemia Model



Ji Hyun Kim Center for Biomaterials my3158@snu.ac.kr



Youngmee Jung Center for Biomaterials winnie97@kist.re.kr



Soo Hyun Kim Center for Biomaterials soohkim@kist.re.kr

For the successful treatment of ischemia, it is important to resupply sufficient blood into ischemic regions by inducing angiogenesis. Stem cell transplantation is currently generating a great deal of interest in the treatment of ischemic diseases. Many stem cell transplantation studies have been reported to enhance angiogenesis, especially those relating to mesenchymal stem cells (MSCs) [1]. However, cell transplantation often encounters problems, such as low rates of cell survival, tumorigenesis, and donor cell shortage [2]. Therefore, more efficient strategies to retrieve therapeutic MSCs from bone marrow are anticipated [3]. To overcome such obstacles in stem cell therapy, *in situ* tissue regeneration is another promising approach in the field of tissue engineering [2]. *In situ* tissue regeneration, which induces mobilization of host stem cells, does not include donor cell procurement and *in vitro* cell manipulation [4].

Recently, a new role for substance P has been reported, as an injuryinducible wound messenger that accelerates wound healing by mobilizing stromal-like cells [1]. Substance P is an 11–amino acid neuropeptide and mediates pain perceptions, neuroimmune modulation, and differentiation of endothelial cells (ECs) [1, 2]. In addition, substance P can accelerate wound healing by recruiting CD29+ stromal-like cells from the periphery to the site of injury [1]. Therefore, substance P is one of the factor candidates which can induce angiogenesis and tissue repair through an enhanced recruitment of autologous stem cells [5].

Self-assembling peptides are promising biomaterials in tissue

engineering, which are injectable, biodegradable, and non-immunogenic [6, 7]. They form fibers (< 10 nm) and assemble into a 3-dimensional (3D) scaffold in the physiological environment, and closely mimic the porosity and gross structure of extracellular matrices. Selfassembling peptides are composed of synthetic amino acids and can be easily designed and modified in a variety of ways [8].

In this study, we developed a novel bioactive peptide which has the ability to recruit MSCs by modifying selfassembling peptides using substance P sequence. The therapeutic effects of the bioactive peptides were evaluated in ischemic hind limb models by injecting them into ischemic regions. Their degradation rates as well as their sustained delivering ability of substance P and MSCs recruiting ability were investigated. In animal studies, the effects of ischemic tissue repair, neovascularizaion and tissue perfusion were estimated. Limb ischemia was produced in athymic mice, and 1% (wt/vol) peptides were injected into ischemic sites (n = 6 in each group): ischemia, substance P, RADA16-II, RADA16-II+substance P, and RADA16-II+RADA-SP (bioactive peptides)). The tissues were harvested for histological analysis and tissue perfusion measurement at 1, 3, 7, and 28 days after injection. By using bioactive peptides, we intended to improve MSC recruiting activity by increasing stability and long term release of substance P into ischemic sites. We postulated that the injection of suitable self-assembling peptides into ischemic regions would create 3D microenvironments, subsequently improve infiltration and attachment of recruited MSCs within them, and promote neovascularization.

By covalently immobilizing substance P sequences to self-assembling peptides, substance P was released over a longer term, and subsequently, MSCs were efficiently recruited into ischemic regions. Moreover, bioactive peptides provided 3D microenvironments by assembling nanofibrous structures. Therefore, bioactive peptides showed increased recruitment of MSCs, preventing fibrosis, and promoting neovascularization with high tissue



Figure 1. Mesenchymal stem cells (MSCs) recruitment of bioactive peptides. Bioactive peptides are effective for the recruitment of MSCs into ischemic regions. Representative images of ischemic regions from each group after (A) CD29-CD90 and (B) CD105-CD90 staining at 7 days after injection. Scale bars: 100 μm (x200).



Figure 2. Sustained delivery of substance P and degradation test of bioactive peptides. Injection of bioactive peptides can provide sustained delivery of substance P for 28 days. (A) Quantification of retained substance P of each group (μ m²/mm²). p > 0.05 with 1 day, p > 0.05 with 3 days, p > 0.05 with SP group. (B) Quantification of peptides' degradation rates for each group (μ m²/mm²). p < 0.05 over 1 day, p < 0.05 over 1 day, p < 0.05 over 3 days, p < 0.05 over 7 days.



Figure 3. Fibrotic tissue formation of ischemic limbs. Injection of bioactive peptides prevents fibrotic tissue formations 7 and 28 days after treatment. (A) and (B) Masson's Trichrome staining at 7 and 28 days, respectively. Scale bars: 100 μ m (x100). (C) Quantification of fibrosis (mm²/mm²). p > 0.05 with SP group, p < 0.05 with 7 days.

perfusion, which led to much improved therapeutic effects of the ischemic hind limb.

This study confirmed that bioactive peptides recruited MSCs efficiently *in vivo* (Figure 1). We confirmed that substance P promotes MSC recruitment into injected sites, even though substance P was physically adsorbed (R+SP) or covalently bonded (R+RSP). In particular, the

R+RSP group was the most effective in MSC recruitment due to the sustained delivery of substance P. In our study, bioactive peptides covalently immobilized substance P, increasing stability and diminishing the diffusion of substance P. Consequently, bioactive peptides released substance P over a longer term, which led to increased MSC recruitment (Figures 1 and 2). Also, injection of bioactive peptides was the most effective at prevention of fibrosis in the ischemic limb model (Figure 3). This is likely due to the structural properties of bioactive peptides which closely mimic the extracellular matrices and inhibit scar tissue formation [8].

The bioactive peptides developed in this study are one of the candidates which can induce angiogenesis through an enhanced recruitment of MSCs, with therapeutic potential for ischemia (Figure 4). In the R+RSP group, vWF⁺ cell density and α -SMA⁺ vessel density were 44.63 and 27.83 times greater than that of the IS group, respectively. Remarkably, this was the only group for which increased vascular density was maintained until 28 days with a high maturation index up to 95%. Moreover, tissue perfusion was tremendously improved and ischemic limbs were fully recovered in all mice, as demonstrated by the limb salvage score (Figure 5). The improvement in tissue perfusion and limb salvage score is due to fibrosis prevention as well as the recovery of blood flow with mature and functional vessels of bioactive peptides. We speculated that these beneficial effects of bioactive peptides result from (i) their MSCs' recruiting ability with angiogenic activity, (ii) 3D microenvironments provided by self-assembling peptides, and (iii) long-term release of substance P.

Substance P-immobilized bioactive peptides have the potential to recruit cells with angiogenic activities, including MSCs. Our results also confirmed that bioactive peptides were effective in the recruitment of MSCs which could induce angiogenesis. It is possible that recruited stem or progenitor cells and stimulated vascular cells by substance P-immobilized bioactive peptides proliferated or sometimes differentiated, and consequently, enhanced



Figure 4. Capillary density, arterial density and maturation index. Injection of bioactive peptides improves capillary density, arterial density and the maturation index. (A) and (B) Representative immunostaining of endothelial cells (ECs) stained with von Willebrand factor (WWF, green) and smooth muscle cells (SMCs) stained with α -smooth muscle actin (α -SMA, red) of each group 7 and 28 days after treatment, respectively. Scale bars: 100 µm (x200). (C) Quantification of vWF⁺ cell density (µm²/mm²). (D) Quantification of α -SMA⁺ vessel density (µm²/mm²). (E) Maturation index (%). Quantification of the percentage of α -SMA⁺ vessels to the total number of vessels. p > 0.05 with SP group, p > 0.05 with 7 days.

angiogenesis with recovery of blood flow. In addition, bioactive peptides provided 3D microenvironments which could promote neovascularization by infiltrating and attaching to vascular cells as well as MSCs. In this study, bioactive peptides could maintain increased vascular density by sustained delivery of substance P (Figure 2). As described previously, bioactive peptides which were covalently bonded with substance P, enabled substance P to long-term release, and that led to the enhanced recruitment of MSCs, extending their angiogenic activity. Therefore, to apply bioactive peptides by long-term delivery of substance P is one of the most powerful strategies to enhance angiogenesis for the treatment of ischemia.

In conclusion, we developed injectable bioactive peptides which could recruit MSCs, and their therapeutic potential was evaluated on an ischemic hind limb model. By applying bioactive peptides, substance P was released long-term and 3D microenvironments were provided into ischemic regions. Bioactive peptides could recruit MSCs successfully, prevent fibrosis, and promote neovascularization, with enhanced tissue perfusion that led to full recovery of ischemic hind limbs. Our results confirmed that bioactive peptides are one of the most powerful tools to treat ischemic disease without cell transplantation by recruiting autologous stem cells.



Figure 5. Laser Doppler perfusion image (LDPI) and limb salvage score. Injection of bioactive peptides enhances tissue perfusion that leads to full recovery of ischemic hind limb. (A) Representative pictures of ischemic limbs and LDPI from each group at 28 days, respectively. (B) Perfusion ratio of each group. (C) Limb salvage score of each group. p < 0.05 with IS group, p < 0.05 with SP group, p < 0.05 with R group, p < 0.05 with R group.

Note

This article and images are cited from "Stem cell recruitment and angiogenesis of neuropeptide substance P coupled with self-assembling peptide nanofiber in a mouse hind limb ischemia model' in *Biomaterials*, Vol. 34 (6), pp. 1657~68.

<u>References</u>

- [1] Hong HS, Lee J, Lee E, Kwon YS, Lee E, Ahn W, et al. *Nature Medicine*. 2009;15:425-35.
- [2] Kohara H, Tajima S, Yamamoto M, Tabata Y. *Biomaterials*. 2010;31:8617-25.
- [3] Hong HS, Kim DY, Yoon KJ, Son Y. Arch. Pharm. Res. 2011;34:2003-6.
- [4] Lee CH, Cook JL, Mendelson A, Moioli EK, Yao H, Mao JJ. Lancet. 2010;376:440-8.

- [5] Kim JH, Jung Y, Kim BS, Kim SH. *Biomaterials*. 2013;34: 1657-68.
- [6] Genove E, Shen C, Zhang S, Semino CE. *Biomaterials*. 2005;26:3341-51.
- [7] Horii A, Wang X, Gelain F, Zhang S. *PLoS One*. 2007;2:e190.
- [8] Kim JH, Jung Y, Kim SH, Sun K, Choi J, Kim HC, et al. Biomaterials. 2011;32:6080-8.



Biomedical Research Institute, Biomaterials Research Laboratory

[Technical Review]

Magnetic-Field-Controlled Reconfigurable Semiconductor Logic



Jin Dong Song Nano Photonics Research Center, KIST jdsong@kist.re.kr



Jinki Hong

Department of Display and Semiconductor Physics, Korea University



Joonyeon Chang

Spin Convergence Research Center, KIST presto@kist.re.kr

Mark Johnson

Naval Research Laboratory, Washington DC 20375, USA

Introduction

Companies producing Si-based chips have recently been struggling with the issue of how to reduce current leakage in devices using smaller and faster Si-based chips. This excessive leakage is caused by the extremely small size of the chips (where quantum physic donates), resulting in parasitic charges. Therefore, it is necessary to create more current in order to suppress parasitic charges and make the devices work effectively.

It is not easy to predict the future. However, considering the exponentially increasing power consumption by IT systems and the inadequacies of planned power provisions, a critical shortage of electric power is highly likely in the near future. One pessimistic study predicts that more than 44 nuclear power plants would be necessary to cope with anticipated power demands by 2020.

In this article we discuss magnetism-based logic devices that show promise for increasing computational efficiency while decreasing consumed power. They offer zero quiescent power and yet combine novel functions such as programmable logic operation and nonvolatile built-in memory [1].

Our approach to the development of a magnetic logic device is uniquely different from that taken for devices based on magnetoresistive effects that result from spin dependent transport of carriers. By exploiting the large magnetoresistance (MR) of non-magnetic semiconductors in high electric fields, our device shows a strong diode characteristic that is highly sensitive to both sign and magnitude of an external magnetic field, offering reversible



Figure 1. Magneto-conductance tunable by external voltage. (a) Schematic diagram and micrograph (bottom right inset) of an InSb device, Sample NP. (b) A family of plots of current versus magnetic field for Sample NP (B//Plane). For a given bias voltage, there is a transition to a high-current state at a characteristic on-set value of magnetic field. In the field region designated by a horizontal arrow (\leftrightarrow) the *I*-*B* characteristic resembles that of an electrical diode. (c) A family of I-B plots for B_Plane. (d) Schematic illustration of carrier transport processes in the channel. (Copied from [1])

change between two different characteristic states by applying a magnetic field.

This feature results from the magnetic control of carrier generation and recombination in an InSb p-n bilayer channel. Simple circuits combining such elementary devices were fabricated and tested by our team. Boolean logic functions including AND, OR, NAND and NOR were performed. They were programmed dynamically by external electric or magnetic signals, demonstrating a magnetic-field-controlled semiconductor reconfigurable logic at room temperature. This novel magnetic technology permits a new kind of spintronic device characterized as a current switch rather than a voltage switch and provides a simple and compact platform for non-volatile reconfigurable logic devices.

Research details

The device fabricated in our lab was based on n-InSb over p-InSb structures (herein referred to as a

NP device). Figure 1a shows the overall structure of the device. As shown in Figures 1b-d, its operation is based on two basic mechanisms: i) a magnetic-field-controlled avalanche process in low bandgap semiconductor materials (such as InSb); and ii) a magnetic lock effect stemming from the structural asymmetry caused by the p-n bilayer channel.

In Figures 1b and 1c, each current trace monotonically increases with increasing magnetic field, and there is an abrupt change in magnitude at a field value that represents the onset of the high current state. This behavior is similar to the characteristic of a p-n junction diode if the bias voltage is replaced by the magnetic field. This avalanche process is based on impact ionization. That is, an electron accelerated by external bias voltage frees the electron-hole pairs via collision with bound electrons. This happens easily with lower bias voltage in low bandgap materials such as InSb. The magnetic field hinders the acceleration of electrons, and as a result, the value for an abrupt increasing current of a specific bias voltage and magnetic field can be fixed. This is the basic



Figure 2. Programmable logic operation: AND/OR gate. Positive (\odot) or negative (\otimes) magnetic fields of -0.1 T and 0.1 T are applied to devices with B//Plane. (a) The circuit used for demonstration of reprogrammable Boolean logic. V1 and V2 are the voltage drops on Samples NP1 and NP2, respectively. Binary logic inputs are positive (negative) magnetic field, corresponding to binary "1" ("0"). Logic output is high (low) current, corresponding to binary "1" ("0"). The Boolean operation to be performed is programmed by setting the bias voltage V_{total} to 22.05 V for the AND operation or 22.20 V for the OR operation. (b) Current-voltage characteristics of NP1. The threshold voltage of NP1 and NP2 at the negative field, V_{\odot}, is 11.13 V, and that at the positive field, V_{\odot}, is 11.0 V. (c) Schematic explanation of AND gate operation, described in text. (d) Schematic explanation of OR gate operation, described in text. (e) Truth table summary of the operations described in c and d. Experimentally measured output values are included (all values were reproducible).

mechanism of the magnetic-field-controlled avalanche process.

In Figure 1c, the plot is symmetrical with respect to the magnetic field (perpendicular to the surface plane of the device). However, it is asymmetrical to the magnetic field in Figure 1b (parallel to the plane). The structural asymmetry caused by the p-n bilayer channel distinguishes the trend of magnetic field vs. current in our structure from other devices. Conductivity is sensitive to a carrier generation process induced by impact ionization, and transport is influenced by field-dependent recombination rates (Figure 1d). A large magnetic field along the negative z-axis deflects energetic carriers in the n-type layer towards the p-n interface by a Lorentz force. Enhanced recombination depletes the carrier population and diminishes the current. That is, the direction of the magnetic field can be distinguished in the device under the set-up where B is parallel to the plane (magnetic lock effect).

New functionalities for information processing are enabled by the unique properties of our device. We fabricated several circuits and demonstrated magneticfield-controlled reconfigurable Boolean logic gates at room temperature. A circuit of two identical devices (Figure 2a) demonstrates operation as a reconfigurable AND/ OR logic gate. The current I is the same for both devices and has the high-state value only if voltage drops V₁ and V₂ exceed the threshold voltages for each device, V₁>V₁₁ and V₂>V₁₂, where V₁₁ and V₁₂ can have values V₀ or V₀ as shown in Figure 2b. In any other case, I has the lowstate value. The circuit performs as an AND or OR gate depending on bias value V_{total}. In Figures 2c and 2d, the I_{output} axis represents circuit current, and bar plots for four different input configurations are presented. Vertical bars



Figure 3. Demonstration of various Boolean operations. A pair of NP and PN devices, designated by red dotted lines, receives a common input signal (magnetic field): One pair is NP3 and PN1 and the second pair is NP4 and PN2. NP and PN devices are driven by bias voltage V_{COPY} and V_{NOT} , respectively. The total current is a summation of the currents driven by these two voltage sources, and the logic output is determined by this total current. Experimentally measured output currents are listed in parentheses for various bias voltages and input values. The outputs of circuit a represent the Boolean logic function COPY or NOT, and those of circuit b represent AND, OR, NOR or NAND function. Thus, several elementary functions are possible with a single circuit system: two functions for circuit a and four functions for circuit b. Each function can be programmed by presetting the bias voltage values.

represent voltage drops across each of the two devices, red (blue) corresponding to a device in the presence of positive (negative) field. In the first configuration of Figure 2c, NP1 and NP2 are both in the negative field (inputs binary 0 and 0), V₁ and V₂ are both less than V_t= V₀, and the current is low. Only in the fourth configuration where NP1 and NP2 are in the positive field (the inputs are binary 1 and 1), both V1 and V2 are larger than V_t= V₀, and the circuit has high current. The input/output characteristics match the AND gate truth table. The OR gate of Figure 2d operates with a similar description. Figure 2e shows experimental results in truth table form.

This manner of reprogrammability can be extended to other Boolean logic functions. The magnetic field is a pseudovector and its sign changes when taking a mirror image of a physical system. Hence, a system is invariant under the substitution of its mirror image and reversal of the field direction. We introduced PN-type devices as mirror images of NP devices. NP devices have an n-type layer above a p-type layer, whereas PN devices have a reverse stack, an n-type beneath a p-type layer. Mirror reflection with respect to the p-n interface ensures the mirror image relation between these two types and a PN device can be replaced by an NP with reversed magnetic field direction. Equivalently stated, PNs in Figure 3 can be considered as NPs with logical negation at the inputs, because the Boolean negation for "1" (positive magnetic field) corresponds to "0" (negative magnetic field). The current was intentionally limited to 100 mA to protect the device from damage and considered as logic "1" for out. Current lower than 40 mA was considered logic "0" for out.

Experimental results for the two circuits are listed in the truth tables in Figure 3 and demonstrate that our reconfigurable logic gates provide the basic Boolean operations, with each logic function programmed by



Figure 4. Demonstration of programmable functionality by magnetic field. a) A circuit diagram whose logic function is changed by magnetic field. The direction of magnetic field on PN2 is the parameter that selects the Boolean operation: Θ for AND and \otimes for OR. V_{total} = 33.3 V.

presetting the bias voltages. Complex operations can be accomplished by building on these elementary functions.

Figure 4 demonstrates that logic functionality is programmable not only by the external bias voltage, but also by the external magnetic field. This novel characteristic has several important advantages. (1) The logic function is programmed by a binary parameter, positive or negative magnetic field directions \odot and \otimes . (2) The binary nature avoids errors that might arise from narrow margins of control voltage and gives reliable performance. (3) The function programming is done in the same manner as the logical inputs, which provides compatibility between binary signals. (4) The magnetic method for function control can be adapted to use the fringe fields of integrated ferromagnetic elements and thereby can offer non-volatile applications by utilizing the element's bistable states.

Our room temperature experiments were performed using a magnetic field externally applied to individual devices in the circuit. For integrated circuit applications,



Figure 5. Schematic, cross-sectional sketch of an integrated, magnetic field controlled avalanche diode. The bilayer diode channel is at right. A patterned ferromagnetic sandwich structure is at left. Fringe magnetic field from the ferromagnet F provides appropriate magnetic field input to the diode, as discussed in the text.

a fully integrated cell is required and can be realized by fabricating ferromagnetic structures in proximity with the diode channels and using their fringe magnetic fields as the diode inputs. The basic idea is depicted in Figure 5. The ferromagnetic structure is a sandwich having a thin, pinned ferromagnetic layer (FP) on the bottom. The magnetization of FP always lies along the positive x axis. A thicker, switchable ferromagnetic layer (F) has a uniaxial anisotropy axis along the x axis with bistable orientations along + x or - x. Layer F is separated from FP by a thin nonmagnetic layer (NM), either a nonmagnetic metal or a thin tunnel barrier. Current pulses of positive or negative polarity can be driven through the sandwich, resulting in the flow of spin-polarized current from FP to F, with spin orientation either parallel or antiparallel with the x-axis. The pulses of polarized current can set the magnetization state of F to be parallel or antiparallel with the x-axis by a process called Spin Torgue Transfer. Ferromagnetic layer F has a dipolar magnetic fringe field, sketched in Figure 5 with dashed lines (layer FP is so thin that its fringe field can be neglected). For the geometry shown, the fringe field in the vicinity of the n-p interface is approximately parallel with the n-p interfacial plane of the bilayer channel. For distances on the order of 100 nm, the field can have magnitude on the order of tenths of a tesla.

The field orientation is reversed when the magnetization of F is reversed by a spin-torque-transfer current pulse. It follows that this localized magnetic field can provide an appropriate binary input for the field-controlled avalanche diode.

Our InSb avalanche diode, if reduced to channel length, width and thickness of roughly 100 nm, is expected to have a threshold voltage V_{th} of about 9 mV and output of 40 μ A in the low current state. Provided that output is limited to five times the low current value, the high current output is 0.2 mA. Unlike voltage-controlled semiconductor devices, magnetization switching is driven by current. Our device is a current switch, and the 0.2 mA output is sufficiently large to provide STT write current input to a subsequent device. Our device is therefore unique in the field of Spintronics because it is intrinsically adapted to device fanout, a requirement for digital logic applications.

It's important to note that magnetization orientation is maintained as a nonvolatile state because of the bistability of magnetic hysteresis. Our approach can be called nonvolatile reconfigurable logic. By adding a magnetic memory device to the cell, any circuit or sub-circuit will remember its most recent configuration and the latest logical result. Circuits, blocks of circuits, or entire chips can be powered off when not in use, followed by "instanton" performance when powered up for an operation. The quiescent power is zero and both the duty cycle and average power can be reduced substantially. The energy per operation is a characteristic parameter, rather than operating power. For a 2 nsec pulse duration, we estimate the largest dissipation in the channel (high current state) is 3.6 femtojoule and an STT write process dissipates 40 attojoule. As a final note, four of our magnetic diodes are adequate to perform the four Boolean functions in our reconfigurable gate (see Figure 4). This is more compact than a comparable cell in a Complementary Metal Oxide Semiconductor (CMOS) gate array, and may lead to higher packing density and faster operating speed. Of greater importance, our cell can be reprogrammed in a

single clock cycle and our architecture offers dynamic reconfigurability.

Note

The text and graphs of this report were reprinted and reused from reference [1] to be used only in KIST for academic and educational purposes.

References

 Joo SJ, Kim TY, Shin SH, Lim JY, Hong JK, Song JD, Chang JY, Lee HW, Rhie KW, Han SH, Shin KH, Johnson M. *Nature* 2013; 494: 72.



Authors of the article

Feature Articles

High-Strain Air-Operable Soft Transducers Produced from Nanostructured Block Copolymer Ionomer/Silicate/Ionic Liquid Nanocomposite Membranes



Jang-Woo Lee Center for Materials Architecturing 023215@kist.re.kr



Seunggun Yu Center for Materials Architecturing t11542@kist.re.kr



Soon Man Hong Center for Materials Architecturing smhong@kist.re.kr



Chong Min Koo Center for Materials Architecturing koo@kist.re.kr

Introduction

lonic polymer-metal composite (IPMC) transducers, which facilitate the conversion of electrical energy to mechanical energy, have been attracting much attention as potential candidates for nature-inspired polymer-based actuators, sensors, artificial muscles, and microrobots. This attraction has mainly been due to their large, fast, and soft bending actuation under fairly low operating voltages of $1 \sim 5$ V, which have not been realized by other electroactive materials, such as dielectric elastomers, ferroelectric polymers, and ceramic actuators [1,2].

IPMCs have a simple capacitor structure consisting of an ion-conducting polymer membrane incorporating mobile ions and inner solvent, and a pair of electrodes covering both surfaces of the membrane. The electromechanical energy conversion (electrical energy to mechanical energy) for the IPMC actuator is typically harnessed from the displacement and force produced through a bending motion in cantilever configuration. The bending mechanism refers to the transport of mobile ions triggered by an external electric stimulus, followed by the electro-osmotic drag of an inner solvent as a dissociation medium, typically water. The ion/solvent migration induces an asymmetric volume change between both sides of an IPMC, thus producing a bending deformation of the IPMC, mostly to the positively charged anodic direction of the electrodes [3,4].

Nafion from DuPont has been the most commonly used synthetic

ionomer membrane for IPMC actuators. The ionic passages clustered with sulfonic acid groups in Nafion on the scale of a few nanometers give high proton conductivity, and the tetrafluoroethylene backbone of the Nafion is responsible for thermal, chemical, and mechanical stabilities [5,6]. Despite the advantages of Nafion, a fast back relaxation and early dehydration have been fatal drawbacks in IPMC devices using a Nafion membrane [6-11]. Back relaxation behavior refers to the situation when a membrane bends back towards its initial position following its maximum forward displacement during the continuous application of dc voltage. It is known that a major cause of back relaxation is the diffusion of water back into the anode after full bending, which limits the frequency range and accuracy of the actuator's motion control [8,9]. The early loss of water out of IPMC during actuation leads to a rapid reduction of ionic flux followed by a decay of actuation performance in air operation. Thus, hydrated Nafion is limited to underwater applications, despite many potential applications in air-filled environments [6,7].

Many Nafion-based IPMCs incorporating organic solvents with high boiling temperatures or ionic liquids (ILs, ionic salts in the liquid state at room temperature) have been proposed to replace the hydrated IPMCs. However, their performance in air was far inferior to that of hydrated Nafion-based IPMCs because of the best dissociation ability and low viscosity in water, and consequently, the greatest proton conductivity in the Nafion membrane [12-14].

As a result of these issues with Nafion, a great deal of effort has been expended in the last decade on the development of new ionomer membranes for an airoperable IPMC that could replace Nafion [6-11,15-17]. These new polymers include sulfonated polystyrenes [7], sulfonated poly(vinyl alcohol)s [6], sulfonated poly(arylene ether sulfone)s [8], sulfonated ethylene vinyl alcohol copolymers [9], fluorinated acrylic copolymers [15], fluoropolymers grafted with sulfonated polystyrene [10], sulfonated poly(styrene-*alt*-maleimide)-incorporated poly(vinylidene fluoride) semi-interpenetrating polymer networks (semi-IPNs) [11], and many others [16,17]. Most of them, however, did not meet the desired ionic conductivities and IPMC actuation performance that would make them superior to Nafion.

In recent years, a handful of nanostructured block copolymer ionomers, such as sulfonated poly(styrene*b*-ethylene-*co*-butylene-*b*-styrene) (SSEBS) triblock copolymer and poly((t-butyl-styrene)-b-(ethylene-rpropylene)-b-(styrene-r-styrene sulfonate)-b-(ethylene-rpropylene)-b-(t-butyl-styrene)) (tBS-EP-SS-EP-tBS; SSPB) pentablock copolymer have been applied as alternatives to Nafion for IPMC applications. The nanostructured block copolymer ionomers were expected to form welldefined microphase-separated ionic domains on the scale of several tens of nanometers, which could work as more effective ion-conduction channels compared with ionic clusters of Nafion on the scale of a few nanometers. The size and microstructure of the ionic domains could also be well controlled by varying the fraction of each block unit and molecular weight of the block copolymer ionomers. Among the styrenic SSEBS and SSPB block copolymer ionomers, SSPB has performed much better in IPMC applications owing to its mechanical strength and toughness sustained even in a hydrated state, and its high ionic conductivity [18,21]. In addition, the hydrated SSPB/ sulfonated montmorillonite (s-MMT) nanocompositebased IPMCs, consisting of SSPB and s-MMT fillers, have revealed much larger bending displacements and blocking forces than the pristine SSPB, owing to strong interactions between mobile ions and sulfonated groups on the surface of s-MMT fillers [21].

Herein, we demonstrate that the nanostructured SSPB/s-MMT nanocomposite membranes, incorporating bulky imidazolium ionic liquids (IL), function as novel air-operable IPMC polymer electrolytes. The well defined microphase-separated ionic domains of the nanostructured SSPB ionomers on the scale of several tens of nanometers and the impregnation of sulfonated silicate fillers (s-MMT) into the nanostructured ionomers


Figure 1. (a) A schematic illustration of IPMCs made of SSPB/IL, SSPB/ s-MMT/IL, Nafion/IL membranes operating at dc potential. SSPB has a microphase-separated morphology. For simplicity, only a sphere symbol represents the mobile ions in the scheme, and X⁺ denotes mobile cations existing in the polyelectrolyte membrane, which can include not only the cation of the IL, but also the cation in the sulfonate group of SSPB. This scheme compares the SSPB and its s-MMT nanocomposites with Nafion. (b) Molecular structure of the SSPB copolymer. (c) Structure of the sulfonated silicate (s-MMT). (d) Molecular structure of the ILs used in this study. TEM images of: (e) SSPB, (f) SSPB/s-MMT, and (g) Nafion. (h) Wide angle X-ray diffraction (WAXD) patterns of MMT, s-MMT, SSPB/ MMT, and SSPB/s-MMT. The silicate content in the nanocomposites was 8 wt%.

resulted in not only an unexpectedly larger and faster bending actuation in an air environment without conventional IPMC drawbacks (including back relaxation and a sacrifice of mechanical strength after impregnation with IL), but also a larger energy efficient actuation than Nafion, as illustrated in Figure 1a.

Nanostructured block copolymer ionomer/silicate/ionic liquid membranes and their IPMCs

Three types of membranes, including SSPB, SSPB/

MMT, and SSPB/s-MMT, were prepared via solution blending and investigated. The chemical structure of the SSPB pentablock copolymer ionomer engaged in this study is shown in Figure 1b. The SSPB had a well-defined microphase-separated morphology in which the repeating domain spacing (d) composed of alternating styrene-rich regions (ionic phase) and aliphatic EP-rich regions (nonionic phase) was ca. 30 nm, and the average diameter of the ionic channel regions was ca. 20 nm. Nafion, however, had narrow ion clusters below 3.8 nm, as shown in Figures 1e and g. Two types of MMTs, pristine Na⁺-MMT (MMT) and the sulfonated MMT (s-MMT) (Figure 1c) were introduced to the SSPB ionomer with a fixed filler content of 8 wt%. MMT had an ion exchange capacity of 0.91 meq. per 1 g silicate and s-MMT had sulfonic acid anions of 0.33 mmol g⁻¹ on the basis of thermogravimetric analysis. In Figure 1h, the SSPB/s-MMT and SSPB/ MMT nanocomposites exhibited the characteristic layer spacing peak (d_{001}) of the layered silicates at smaller angles than s-MMT and MMT fillers. This indicates that the SSPB/s-MMT and SSPB/MMT nanocomposites had an intercalation morphology. The SSPB/s-MMT had a peak at a smaller angle than the SSPB/MMT, meaning that the s-MMT had a better dispersion state and a more developed intercalation than the MMT in the SSPB ionomer. In Figure 1f, the TEM image of SSPB/s-MMT also reveals that the SSPB/s-MMT achieved a welldeveloped intercalated morphology. It should be noted that silicate platelets were homogeneously distributed in the SSPB matrix and located over the several repeating domains in the SSPB nanostructured pentablock copolymer ionomer membrane.

SSPB/IL, SSPB/MMT/IL, and SSPB/s-MMT/IL IPMC actuators were fabricated via chemical deposition of platinum on both sides of each membrane, followed by impregnation with IL as the inner solvent in the IPMC [3,20]. The four imidazolium ionic liquids (C2, C4, C6, C8) introduced to each membrane are shown in Figure 1d. The four ILs have the same bis(trifluoromethylsulfonyl) imide (TFSI) anion but a different imidazolium cation with

different lengths of the linear hydrocarbon substituent on the N(1) position of the imidazolium cation. The number in the sample codes of ILs denotes the number of carbons in the hydrocarbon substituent on the imidazolium cation; thus, the bulkiness of the IL increases as the carbon number of the hydrocarbon substituent increases.

Characterization of membranes

Figure 2 shows the IL content in the SSPB/IL, SSPB/ MMT/IL, and SSPB/s-MMT/IL IPMCs after being fully impregnated with IL. The IL uptake by the bare SSPB membrane was much higher than that by a cast Nafion membrane, and steadily increased with the increasing length of the linear alkyl substituent on the imidazolium cation of IL from 81 to 126 wt%. The higher IL uptake level of the SSPB membrane was attributed not only to the larger concentration of sulfonic acid groups in the SSPB ionomer but also to the larger ionic channel size, compared with the Nafion counterpart [2,21]. The incorporation of MMT into the SSPB nanostructured block copolymer membrane augmented the uptake level, which



Figure 2. IL uptake of SSPB/IL, SSPB/MMT/IL, SSPB/s-MMT/IL, and Nafion/IL at equilibrium. The value of uptake was obtained by the following equation: uptake (wt%) = $(W_i - W_n)/W_n \times 100$, where W_n and W_i are the weights of the neat and IL- or water-impregnated membranes, respectively. The silicate content in the nanocomposites was 8 wt%.

further increased through the introduction of s-MMT with sulfonic acid groups. This result is reasonable considering the hygroscopic nature of MMT and s-MMT.

Figure 3a displays the ion conductivities of the SSPB/ IL, SSPB/MMT/IL, and SSPB/s-MMT/IL membranes. The ion conductivities of the SSPB/IL membranes decreased with increasing carbon number on the N(1) position of the imidazolium ring of IL in the range of 2.21 ~ 0.29 \times 10⁻⁴ S cm⁻¹, which were smaller than those of the respective neat ILs (8.02 ~ 2.38 × 10⁻³ S cm⁻¹). The SSPB/MMT/IL and SSPB/s-MMT/IL nanocomposite membranes also revealed the same tendency as the SSPB/IL membrane. The decay in ion conductivity observed from C2 to C8 was largely related to the increase in the bulkiness and viscosity of IL, both of which deteriorate ion transport in both cases of IL in the bulk state and inside the ionomer matrix [4,14]. The viscosities of C2, C4, C6, and C8 were 34, 52, 87.3, and 119.3 cP, respectively [22,23]. The SSPB/MMT/IL membranes had less ion conductivities than the SSPB/IL membranes at the same ILs. However, interestingly, the SSPB/s-MMT/IL membranes showed greater ion conductivities than the SSPB/IL membranes, regardless of ILs used. The ion conductivities of SSPB/ s-MMT/C2, SSPB/s-MMT/C4, SSPB/s-MMT/C6, and SSPB/s-MMT/C8 were found to be 2.60, 2.15, 0.62, and 0.32×10^{-4} S cm⁻¹, respectively. The incorporation of s-MMT into the SSPB matrix resulted in a well-dispersed intercalation morphology (i.e., sulfonated silicate platelets located over several repeating domains in the SSPB) due to the strong interaction between the sulfonic groups of SSPB and the sulfonic groups of s-MMT. Thus, the high ion conductivities in the SSPB/s-MMT/IL membranes could be attributed to the presence of sulfonic groups on the highly intercalated s-MMT platelets that facilitate ion transport between the adjacent wide ionic channels, like an ionic bridge in the nanostructured SSPB polymer matrix, as illustrated in Figure 1a. It is also worth noting that the ionic conductivities obtained from all the prepared SSPB-based membranes were higher than those of the Nafion counterpart, regardless of the ILs and silicate fillers



Figure 3. (a) Ion conductivity of SSPB/IL, SSPB/MMT/IL, and SSPB/ s-MMT/IL membranes with a series of ILs (C2, C4, C6, and C8). (b) Tensile stress-strain curves of the SSPB/IL, SSPB/MMT/IL, and SSPB/ s-MMT/IL membranes incorporated with C2 IL. The silicate content in the nanocomposites was 8 wt%.

used in this study.

The tensile stress-strain curves of SSPB/IL, SSPB/ MMT/IL, and SSPB/s-MMT/IL membranes are shown in Figure 3b. In spite of the enormous IL uptake, the SSPB/ IL, SSPB/MMT/IL and SSPB/s-MMT/IL membranes maintained the ultimate elongation levels of pristine SSPB of at least ca. 400%, while the Nafion/IL membranes revealed elongations at breakage of less than 45%. Generally, more IL uptakes in polymer membranes lead to larger ion conductivities [3,14,24]. An excessive IL uptake, however, decreases the mechanical strength of the membrane, and further IL uptake worsens even the bending actuation performance [6,12,21]. Fascinatingly, the strong swelling selectivity of IL on the microphaseseparated ionic domains of the SSPB copolymer could lead to excellent mechanical properties despite the high IL uptake level of up to 163 wt%. The selective swelling of IL prevents it from easily plasticizing the non-ionic phases responsible for the mechanical strength. The incorporation of layered silicates increased the tensile modulus of SSPB during the initial stage of tensile deformation. The SSPB/ s-MMT nanocomposites had larger tensile moduli than the SSPB/MMT because of their better intercalation states.

Electromechanical behavior of IPMCs

The electromechanical responses of IPMC actuators based on the SSPB/IL, SSPB/MMT/IL, and SSPB/ s-MMT/IL membranes were evaluated through measuring the horizontal displacement at the tip of the cantilever (bendable free length: 20 mm) under 2 V dc during 10min bending actuation. In Figure 4a, the tip displacement of the SSPB/IL IPMCs loaded with different ILs follows the order of C8 \approx C6 > C4 > C2. The tip displacements of the SSPB/MMT/IL and SSPB/s-MMT/IL IPMCs also showed the same tendency as that of the SSPB/IL IPMC. In addition, none of the examined IPMCs showed any evidence of back relaxation during the actuation period (10 min). Generally, the higher conductivity of an ionomer membrane leads to a larger bending deformation in the IPMC [16,17]. However, the actuation performance order of the SSPB-based IPMCs with the four imidazolium ILs (C8 \approx C6 > C4 > C2) was the reverse of the ion conductivity result, which was in the order of C2 > C4 >C6 > C8.

The tip displacements of the SSPB/C6, SSPB/MMT/ C6, and SSPB/s-MMT/C6 IPMCs incorporating C6 IL are exhibited in Figure 4b. Figure 4c is a visual representation of the time-dependent bending motion of the SSPB/



Figure 4. Tip displacement vs. time curves of IPMCs based on SSPB/ IL, SSPB/MMT/IL, SSPB/s-MMT/IL membranes under an applied voltage of 2 V dc: (a) SSPB/IL IPMCs with a series of ILs (C2, C4, C6, and C8); and (b) SSPB/IL, SSPB/MMT/IL, and SSPB/s-MMT/IL IPMCs incorporating C6 IL. The nanocomposite membranes as polymer electrolytes of the IPMCs had a silicate content of 8 wt%. (c) Visualized time-dependent bending motion of SSPB/s-MMT/C6 IPMC taken under an applied potential of 5 V dc.

s-MMT/C6 IPMC. The bending motion of the SSPBbased IPMCs was much larger and faster than that of the Nafion-based IPMC. The displacements and response rates followed the order of SSPB/s-MMT/C6 > SSPB/C6 > SSPB/MMT/C6 IPMCs, which was approximately the same as that of the ion conductivity results.

To the best of our knowledge, the maximum bending displacement of the SSPB/s-MMT/C6 IPMC, which was 10.2 mm under 2 V dc within the given 10 min, was superior to those of top-ranked IL-incorporated ionic polyelectrolytes [4,25-27]. Moreover, despite the use of bulky IL to give stable air-operation ability to the IPMC, the displacement and response rate of the SSPB/s-MMT/C6 IPMC were substantially superior to that of the SSPB/ s-MMT/water IPMC (SSPB/s-MMT/water IPMC: 4.6 mm after 2 min actuation at 2 V dc with the identical bendable free length of specimen) [21].

The largest and fastest bending performance of SSPB/s-MMT was attributed to the SSPB's large ionic domains as well as the strong interactions between the mobile ions and the sulfonic groups on the s-MMT. Although the IL molecules are much bigger than water molecules, the ionic domains of SSPB—much bigger than IL—facilitated the transport of the bulky mobile ions. The strong interactions between the mobile ions and the sulfonic groups on the s-MMT promote ion transport between the ionic domains in the SSPB matrix, resulting in the improvement of ion conductivity. That is, sulfonated silicates act as ionic bridges accelerating the transport of mobile ions between the ionic domains.

Nonetheless, the reverse order between the actuation performance of SSPB-based IPMCs with the four imidazolium ILs (C8 \approx C6 > C4 > C2) and the membrane ion conductivity (C2 > C4 > C6 > C8) has not yet been explained. To further understand this extraordinary phenomenon, the capacitance—another critical factor determining the bending deformation—was considered.

The capacitances of the SSPB/IL, SSPB/MMT/IL, and SSPB/s-MMT/IL IPMCs with respect to the area of electrode determined by cyclic voltammetry (CV) are shown in Figure 5a. ILs with shorter hydrocarbon substituents on the N(1) position of the imidazolium cation presented larger capacitances to the SSPB/IL IPMCs because the bulkiness of imidazolium cation sterically hinders charging at the interface of the membrane/ electrode [28]. Given this, it makes sense that the SSPBbased IPMCs possessing large ionic channels have capacitances larger than the Nafion-based IPMC because of a wide interface between the platinum electrode and the ionic domain of SSPB. In the meantime, the SSPB/ IL IPMCs had larger capacitances than both the SSPB/ MMT/IL and SSPB/s-MMT/IL IPMCs. This observation might be due to the same steric hindrance by the silicate fillers placed in the interface between the electrode and the ionic domain of SSPB.

The normalized charge-specific displacements for SSPB/IL, SSPB/MMT/IL, and SSPB/s-MMT/IL IPMC actuators are shown in Figure 5b. The charge-specific displacement reflecting the contribution of a mobile ion to the bending displacement was normalized based on the values of the C2-embedded sample of the respective SSPB, SSPB/MMT, and SSPB/s-MMT frameworks. The normalized charge-specific displacement increased with the increase in bulkiness of the IL, and saturation was observed at C6. In general, bulkier ions make larger contributions of mobile ions to the bending displacement because of their larger volume per unit ion. However, the normalized charge-specific displacement of SSPB-based IPMCs incorporating bulky IL must not be attributed only to the bulkiness of the mobile imidazolium cations. The SSPB/C6, SSPB/MMT/C6, and SSPB/s-MMT/C6 IPMCs measured normalized charge-specific displacements of 2.1, 2.6, and 3.0, respectively, which were much larger than the normalized molar volume of C6 of 1.3. The molar volumes of the ILs were calculated using the IL densities [29]. The behavior could be interpreted by an ion-pumping model [30]. The mobile ions, solvated with inner solvent, pump loosely-bound solvent molecules like a piston in the ionic channels under an external electric potential when the sizes of the solvated ion complex and the channel



Figure 5. (a) Capacitances of SSPB/IL, SSPB/MMT/IL, and SSPB/ s-MMT/IL IPMC actuators assessed through cyclic voltammetry (twoelectrode system, -2 ~ 2 V, 50 mV s⁻¹). (b) Normalized charge-specific displacements for SSPB/IL, SSPB/MMT/IL, and SSPB/s-MMT/IL IPMC actuators and normalized molar volumes of imidazolium ILs (C2, C4, C6, and C8). The charge-specific displacement, which is the maximum tip displacement (attained within 10 min under 2 V dc) divided by the capacitance, was normalized by that of each C2-impregnated IPMC with the identical framework. The molar volumes of ILs, which were calculated using the corresponding densities, were also normalized by that of C2. The nanocomposite membranes as polymer electrolytes of the IPMCs had a silicate content of 8 wt%.

match well with each other. In air-operable SSPB-based IPMC actuators, in addition to the dissociated mobile imidazolium cations of IL, the abundant associated ion pairs of IL, which are not a source of current and play as an inner solvent, were migrated by the ion-pumping effect [28]. This pumping effect caused larger bending displacements of IPMCs per unit charge. The pumping effect, depending on the viscosity of IL, indicated that bulky ILs gave stronger pumping effects than small ILs.

Despite a relatively significant mismatch between the sizes of the mobile ions (below 2 nm in length for all the ionic species including two ions from IL and a counter ion from SSPB) and the ionic channels (having diameters of approximately 20 nm), the pumping effect, evidently observed in our SSPB/IL system, was probably due to a loose size-matching between the large ion channel of SSPB and the highly viscous imidazolium-based ILs compared with water.

With the same IL, the normalized charge-specific displacements were in the order of SSPB/s-MMT/IL > SSPB/MMT/IL > SSPB/IL IPMCs. The largest normalized charge-specific displacement of SSPB/s-MMT could also be attributed to the strong ion-pumping effect of SSPB/s-MMT/IL IPMC caused by the strong interactions between the solvated mobile ions and the sulfonic acid groups on the s-MMT.

Conclusion

The nanostructured middle-block sulfonated styrenic pentablock copolymer ionomer (SSPB)/sulfonated silicate (s-MMT) nanocomposite membranes incorporating bulky imidazolium ionic liquids (IL) revealed a much larger and faster bending actuation while operating in air than the SSPB/IL and Nafion/IL membranes, without conventional IPMC drawbacks such as back relaxation and sacrifice of mechanical strength. Moreover, the airoperable IL-incorporated SSPB/s-MMT nanocomposite IPMC revealed a longer service life as well as a larger bending strain than the hydrated SSPB/s-MMT IPMC. The superior actuation performance of the SSPB/s-MMT/ IL IPMC when operating in air far surpassed that of its Nafion counterpart and is attributed to the attainment of high ion conductivity through the enormous IL uptake together with a capacity to maintain its inherent mechanical property. The effective transport of mobile ions for the bending motion is attributed to the welldefined microphase-separated large-sized ionic domain

morphology of the SSPB copolymer ionomer on the scale of several tens of nanometers and to the bridging effect originating from sulfonic groups on the s-MMT. Meanwhile, as the bulkiness of the IL increased, the ion conductivity decreased, whereas the charge-specific displacement of the SSPB/s-MMT/IL IPMC increased. This was caused by the bulky mobile ions of IL, producing strong pumping effects in the nanostructured SSPB ionomer membranes containing ionic domains larger than the mobile ions and solvent. We demonstrated a useful protocol to design a new generation of nanostructured ionomeric block copolymer nanocomposite membranes incorporating ion conduction-activating fillers for airoperable ionic transducers. We believe that this work can easily be extended to other applications with structural similarities, including fuel cells, capacitors, and a variety of other electronic devices.

Note

This article and images are cited from "Highstrain air-working soft transducers produced from nanostructured block copolymer ionomer/silicate/ionic liquid nanocomposite membranes" in *J. Mater. Chem. C*, Vol.1(24), pp. 3784~3793.

<u>References</u>

- Shahinpoor M, Bar-Cohen Y, Simpson JO. Smart Mater Struct. 1998; 7: R15-R30.
- [2] Shahinpoor M, Kim KJ. Smart Mater Struct. 2001; 10: 819-833.
- [3] Duncan AJ, Akle BJ, Long TE, Leo DJ. Smart Mater Struct. 2009; 18: 104005.
- [4] Lee JY, Wang HS, Yoon BR, Han MJ, Jho JY. Macromol Rapid Commun. 2010; 31: 1897-1902.
- [5] Shahinpoor M, Kim KJ, Leo DJ. *Polym Compos.* 2003; 24: 24-33.

- [6] Lee JW, Kim JH, Goo NS, Lee JY, Yoo YT. J Bionic Eng. 2010; 7: 19-28.
- [7] Luqman M, Lee JW, Moon KK, Yoo YT. J Ind Eng Chem. 2011; 17: 49-55.
- [8] Akle BJ, Leo DJ, Hickner MA, McGrath JE. J Mater Sci. 2005; 40: 3715-3724.
- [9] Phillips AK, Moore RB. Polymer. 2005; 46: 7788-7802.
- [10] Han MJ, Park JH, Lee JY, Jho JY. Macromol Rapid Commun. 2006; 27: 219-222.
- [11] Lu J, Kim SG, Lee S, Oh IK. Adv Funct Mater. 2008; 18: 1290-1298.
- [12] Lee JW, Vinh KN, Park SY, Yoo YT. J Korean Phys Soc. 2006; 48: 1594-1600.
- [13] Bennett MD, Leo DJ. Sens Actuators A. 2004; 115: 79-90.
- [14] Akle BJ, Bennett MD, Leo DJ. Sens Actuators A. 2006; 126: 173-181.
- [15] Jeong HM, Woo SM, Lee S, Cha GC, Mun MS. J Appl Polym Sci. 2005; 99: 1732-1739.
- [16] Tiwari R, Garcia E. Smart Mater Struct. 2011; 20: 083001.
- [17] Duncan AJ, Leo DJ, Long TE. *Macromolecules*. 2008; 41: 7765-7775.
- [18] Wang XL, Oh IK, Lu J, Ju J, Lee SW. *Mater Lett.* 2007; 61: 5117-5120.
- [19] Vargantwar PH, Shankar R, Krishnan AS, Ghosh TK, Spontak RJ. Soft Matter. 2011; 7: 1651-1655.
- [20] Vargantwar PH, Roskov KE, Ghosh TK, Spontak RJ. Macromol Rapid Commun. 2012; 33: 61-68.
- [21] Lee JW, Hong SM, Kim J, Koo CM. Sens Actuators B. 2012; 162: 369-376.
- [22] Bonhôte P, Dias AP, Papageorgiou N, Kalyanasundaram K, Grätzel M. *Inorg Chem.* 1996; 35: 1168-1178.
- [23] Zhang S, Sun N, He X, Lu X, Zhang X. J Phys Chem Ref Data. 2006; 35: 1475-1517.
- [24] Hoarfrost ML, Segalman RA. *Macromolecules*. 2011; 44: 5281-5288.
- [25] Vidal F, Plesse C, Teyssié D, Chevrot C. Synth Met. 2004; 142: 287-291.
- [26] Imaizumi S, Kokubo H, Watanabe M. *Macromolecules*. 2012; 45: 401-409.

- [27] Fukushima T, Asaka K, Kosaka A, Aida T. Angew Chem Int Ed. 2005; 44: 2410-2413.
- [28] Lee JW, Yoo YT. Sens Actuators B. 2009; 137: 539-546.
- [29] Ohlin CA, Dyson PJ, Laurenczy G. Chem Commun. 2004; 1070-1071.
- [30] Onishi K, Sewa S, Asaka K, Fujiwara N, Oguro K. Electrochim Acta. 2001; 46: 1233-1241.



Center for Materials Architecturing

[Feature Articles]

Enhanced Oxygen Diffusion in Epitaxial Lanthanum Strontium Cobaltite Thin Film Cathodes for Micro Solid Oxide Fuel Cells



Ho-II Ji High-Temperature Energy Materials Research Center earvb@kist.re.kr



Jong-Ho Lee High-Temperature Energy Materials Research Center jongho@kist.re.kr

Abstract

The chemical diffusion coefficient (D_{chem}) of LSC film was measured by employing a newly designed unique method. Remarkably, the D_{chem} of epitaxial LSC thin films was found to be higher than that of bulk LSC by up to two orders of magnitude, and was found to be enhanced further as the film became thinner. This remarkable discovery based on the enhanced oxygen transport property in the thin film cathode is very encouraging, providing clear insight into the development of thin film cathodes for high-performance μ -SOFCs. Therefore, the novel method to measure the bulk diffusion coefficient in thin films, which we demonstrate in this paper, represents great potential for the development of a highly efficient electrode for μ -SOFCs and also for other electrochemical devices, such as electrolysis cells, membrane reactors, and gas sensors.

Introduction

Micro-solid oxide fuel cells (μ -SOFCs) have received great attention as novel power sources for mobile applications because of their many intriguing properties (high power and energy density, system efficiency, and fuel flexibility) [1]. However, the performance of μ -SOFCs can suffer, most likely as a result of electrode polarization, especially in relation to sluggish kinetics in cathode reactions [2,3]. Unfortunately, there is still a lack of basic understanding of the physicochemical properties of thin film cathodes. Thus far, most studies on the characterization of thin film cathodes have dealt with the measurement of their surface exchange coefficients (k_{chem}) rather than their bulk diffusion properties (D_{chem}), because the surface exchange reaction is thought to be a main rate-determining step in thin-film cathodes [4-6].

There have been only a few studies reporting oxygen diffusion kinetics in SOFC cathode thin films [7,8], primarily because the measurement of the bulk diffusion coefficient (equivalent to the oxygen diffusion coefficient in a thin film cathode) is a very intricate process due to the limited diffusion length in thin film cathodes. In a real μ -SOFC system, however, oxygen diffusion kinetics might become very important for cathode reactions because of the extremely complex cathode structure (e.g., gradient structured or multi-layered cathode based on composite materials) [2,9]. Hence, the transport properties of thin film cathodes should be thoroughly investigated and clarified in order to advance the development of a high performance cathode for μ -SOFCs.

Here we present the D_{chem} of La_{0.6}Sr_{0.4}CoO₃₋₈ (LSC) thin film, which has been measured in a rather straightforward manner by employing a newly designed conductivity relaxation technique. To the best of our knowledge, this is the first proposal for the viable measurement and interpretation of enhanced oxygen diffusion kinetics in thin film cathodes, and we believe that this study provides meaningful insight into the proper implementation of thin film cathodes for high-performance μ -SOFCs.

Experimental

60-nm- and 243-nm-thick LSC thin films were deposited on a (100) LaAlO_{3- δ} (LAO) substrate by pulsed laser deposition (PLD). Both LSC and LAO have the same pseudo-cubic perovskite structure and their lattice parameters are a \approx 3.84 Å [6,10] and a = 3.788 Å [11],

respectively, thus exhibiting a small lattice mismatch with each other. Figure 1 shows a transmission electron microscopy (TEM) cross-sectional image of the LSC/LAO interface and selected area electron diffraction patterns (SAED) of the LSC/LAO sample.

SAEDs of LAO (Figure 1b), LSC (Figure 1c), and the LSC/LAO interface (Figures 1d-e) clearly show that the LSC film was grown epitaxially on the LAO substrate. However, as can be seen in the {001} reflections in Figures 1d-e, LSC and LAO have different c lattice constants, whereas they have almost the same a (=b) lattice constants. This means that the LSC film was compressed along the lateral a and b directions due to constraint given



Figure 1. a) A TEM cross-sectional image of 60-nm-thick LSC film on LAO and b-e) selected area electron-diffraction patterns along the [110] axis of the LSC/LAO cross-section at: b) the LAO substrate, c) the LSC thin film, and d,e) the LSC/LAO interface.



Figure 2. Schematics of the D_{chem} measurement set-up.

by the LAO substrate, which has a smaller lattice constant than LSC, thereby inducing lattice expansion of the LSC film along the c direction as a counteraction to maintain the Poisson ratio. This anisotropic lattice distortion could be gradually eased with distance from the interface. The compressive strain given in the lateral direction of the LSC film is fairly released for the 243-nm-thick film, whereas that of the 60-nm-thick film largely remains, and thus results in a greater compressive state for the 60-nm-thick film than for the 243-nm-thick film. The compressive state would be expected to become higher in both LSC films at higher temperatures where the D_{chem} is measured because the thermal expansion coefficient (TEC) of LSC is larger than that of LAO (LSC: $22.3 \times 10^{-6} \text{ K}^{-1}$, LAO: $12.6 \times 10^{-6} \text{ K}^{-1}$) [12].

Results and discussion

The D_{chem} was measured using a conductivity relaxation technique via a DC four-probe method. As previously mentioned, oxygen diffusion inside the film is too fast because of the short diffusion length, which is limited by film thickness; therefore, oxygen transport in the cathode film is generally controlled by the surface exchange reaction [13]. Therefore, to accurately measure D_{chem} , a longer diffusion length should be secured by avoiding a situation in which the entire film surface is directly exposed to the atmosphere, thereby limiting the diffusion length to the thickness of the film. In this study, similar to a previously reported method [7,8], the



Figure 3. a) Conductivity relaxations of 60-nm- and 243-nm-thick films limited by oxygen diffusion inside the films. b) D_{chem} of 60-nm- and 243-nm-thick LSC thin films, and a LSC bulk. Several previously reported D_{chem} of LSC bulk samples [17,18] were also included for comparison despite some difference in their compositions (La_{0.5}Sr_{0.5}CoO₃).

surface of the LSC film was covered with an Al_2O_3 layer to block oxygen diffusion along the entire LSC surface in the vertical direction of the film (Figure 2), thereby only allowing oxygen diffusion along the lateral direction of the film. An additional LAO buffer layer was inserted between the LSC and Al_2O_3 layers to prevent interfacial reactions between those materials. All these covering layers were fully dense without any cracks and delamination. Significant leakage current was not observed through the LAO substrate [14] during the conductivity measurement because the conductance of the 60-nm-thick LSC film was already 500 times larger than that of the LAO substrate at a measurement temperature of 600°C.

Conductivity relaxation measurements were performed in a temperature range of 550°C to 650°C, where the reaction kinetics for the equilibration were sufficiently high, whilst avoiding any film morphology changes caused by thermal aging. The conductivity relaxation curves and their fitting results are shown in Figure 3a, in which it is shown that the relaxation curves fit well with Eq. 1 based on Fick's second law [15,16] :

$$\frac{\overline{\sigma}(t) - \sigma(0)}{\sigma(\infty) - \sigma(0)} = 1 - \sum_{n=0}^{\infty} \frac{8}{(2n+1)^2 \pi^2} \exp\left[-\frac{(2n+1)^2 \pi^2 D_{\text{chem}} t}{a^2}\right]$$
(1)

where $\sigma(t)$ is conductivity at time *t* and a is the blocking length of the film (4 mm in this study) by the Al₂O₃ layer.

The D_{chem} of the 60-nm- and 243-nm-thick LSC thin films are plotted as a function of temperature in Figure 3b and compared to those of a LSC bulk. The D_{chem} of the LSC bulk shown in this figure was also extracted from the conductivity relaxation technique, which was well matched to other reports [17,18]. Remarkably, the D_{chem} of the LSC thin films was greater than that of the LSC bulk by two orders of magnitude. From the definition of chemical diffusivity, the D_{chem} of oxygen in LSC is proportional to the self diffusivity of oxygen vacancy and electronic transference number, t_i (t_i is equal to 1 in LSC) and further enhanced by the thermodynamic factor. Therefore, even though the thermodynamic factor ($\partial \ln a_{0_2}/\partial \ln c_{V_0}$) of bulk LSC is known to be nearly constant over the oxygen partial pressure ($10^{-1.7} \sim 1$ atm) and temperature range (600~700°C) [17], the enhanced D_{chem} in LSC film can be attributed, in principle, to the increase in oxygen mobility and/or the increase in oxygen vacancy concentration (exactly the increased thermodynamic factor) in LSC thin films. Another interesting observation in Figure 3b was that the D_{chem} of the thinner film was larger than that of the thicker one. This increased D_{chem} of the thinner film can also be explained by the enhanced oxygen mobility and/ or the increased thermodynamic factor in thinner LSC

film.

In order to check which condition caused enhanced D_{chem} in thinner LSC film, we first tracked change in the oxygen vacancy concentration (equivalent to oxygen nonstoichiometry, δ) of LSC film. It is known that the oxygen non-stoichiometry (δ) of dense LSC films which have been estimated from impedance spectroscopy is smaller than that of bulk [19]. Recently, however, la O' et al. [6] also found from impedance spectroscopy that the oxygen non-stoichiometry (δ) of epitaxial LSC films was higher than that of bulk. These conflicting results regarding the extent of non-stoichiometry (δ) of LSC thin film and its relation to temperature and equilibrium oxygen partial pressure are not clearly explained yet.

In general, the valence state of cobalt ion varies between 2+ and 4+, while those of La(3+), Sr(2+), and O(2-) are fixed. Therefore, when a portion, x, of La³⁺ in LaCoO₃ is replaced by Sr²⁺ to form La_{1-x}Sr_xCoO_{3-δ}, electroneutrality is maintained by both a decrease in oxygen content and an increase in mean cobalt valence [20]. Hence, the oxygen non-stoichiometry, δ , of LSC film can be inferred from the mean cobalt valence, n, as follows;

$$\delta = 1/2 ((3+x)-n) = 1/2 (3.4-n) : (x=0.4)$$
 (2)

In order to evaluate the extent of oxygen nonstoichiometry, δ , from Eq. (2), the depth distribution of Co oxidation states in the LSC thin films was analyzed by Electron Energy Loss Spectroscopy (EELS) depth profiling. According to previous reporting [21], intensity ratios of L3 to L2 peaks of the L2, 3 edges in the EEL spectra of Co cation are known to carry information on the oxidation state of Co. Hence, from the traced depth profile of the Co oxidation state along the film thickness, the relative distribution of oxygen vacancies from the film surface to the LSC/LAO interface can be quantified because the abundance of Co cations with lower oxidation states reveals the existence of more oxygen vacancies for satisfying the charge neutrality criterion [20].



Figure 4. Depth profiling of L3 and L2 peaks of the L2, 3 edges in the EEL spectra of Co cation: a) in 60-nm-thick LSC film; and b) in 243-nm-thick LSC films. c) Comparison between thin films and bulk.

Figures 4a and b show EELS depth profiles of LSC films with the different thicknesses of 60 and 243 nm. As shown in these figures, each EEL spectrum kept a nearly equivalent intensity profile throughout the whole film thickness which indicated no variation in oxygen vacancy concentration with respect to the position along the film thickness, even though some anisotropic lattice distortion gradually emerged across the vertical direction from the interface. Furthermore, as shown in Figure 4c,

both 60 and 243 nm thick films showed no difference in Co L2 and L3 intensity and also showed nearly the same intensity profile as that of bulk LSC, indicating that the oxygen vacancy concentration of thin films is not much different than that of bulk and also has no thickness dependence. Consequently, if we are only concerned with oxygen vacancy concentration, the higher D_{chem} of the thinner film as compared to thicker or bulk LSC cannot be explained. These results are contradictory to our initial expectation that the oxygen vacancy concentration of LSC film would be different than that of bulk, as explained by Z. Cai et al. [22].

According to the first principles-based calculation, oxygen vacancy formation was facilitated by the tensile strain given to the LSC film by the lattice mismatch with the substrate. In our case, LSC film was compressed along the lateral direction due to the constraint given by the LAO substrate which has a smaller lattice constant than LSC, indicating that vacancy formation can't be easier in thin film than bulk. Nonetheless, since our EELS analysis showed the oxygen vacancy formation in LSC thin film was not affected by the constrained stress, we had to consider the possible change in thermodynamic factor with respect to the strained state of thin film.

Oxygen mobility, which can also be affected by the lattice strain, is another factor to be considered in explaining the difference in D_{chem} values between the 60nm- and 243-nm-thick films [16,23,24]. The different D_{chem} activation energies in each case can be circumstantial evidence for the change in oxygen mobility with respect to different thin film strain states. The strain effect on mobility, however, is still controversial and beyond the scope of the present investigation. This topic will be addressed in a forthcoming paper. Nevertheless, it is worth mentioning that the method used for measuring D_{chem} in the thin film cathode is a very unique and effective tool, which has never been used before. Furthermore, the distinctly observed enhanced D_{chem} of the thin film cathode is very encouraging, providing clear insight into the development of thin film cathodes for high-performance µ-SOFCs.

Summary

Chemical diffusion coefficients (D_{chem}) of LSC cathode thin films were measured by a modified conductivity relaxation technique which was uniquely designed to extend diffusion length. According to our measurements, the D_{chem} of epitaxial LSC thin films was found to be higher than that of bulk LSC by up to two orders of magnitude, and was further enhanced as the film became thinner. To the best of our knowledge, this is the first interpretation of enhanced oxygen diffusion kinetics in thin film cathodes that applies a viable measurement method. This remarkable discovery, based on enhanced oxygen transport in thin film cathodes, affords great potential for the development of highly efficient electrodes for μ -SOFCs as well as for other electrochemical devices, such as electrolysis cells, membrane reactors, and gas sensors.

Acknowledgments

This work was supported by the Fundamental R&D Program for Core Technology of Materials funded by the Ministry of Knowledge Economy, Republic of Korea, and the Institutional Research Program of the Korea Institute of Science and Technology (KIST).

Note

This article and images are cited from "Enhanced Oxygen Diffusion in Epitaxial Lanthanum Strontium Cobaltite Thin Film Cathodes for Micro Solid Oxide Fuel Cells" in *Energy and Environmental Science* 2013; 6: 116-120.

References

- Kwon CW, Son JW, Lee JH, Kim HM, Lee HW, Kim KB. Adv. Funct. Mater. 2011; 21: 1154. Su PC, Chao CC, Shim JH, Fasching R, Prinz FB. Nano Lett. 2008; 8: 2289. Evans A, Bieberle-Hutter A, Rupp JLM, Gauckler LJ. J. Power Sources 2009; 194: 119.
- [2] Adler S, Chem. Rev. 2004; 104: 4791.
- [3] Steele BCH, Heinzel A. Nature 2001; 414: 345. Minh NQ, J. Am. Ceram. Soc. 1993; 76: 563. Minh NQ, Singhal C, Williams M. ECS Trans. 2009; 17: 211. Fabbri E, Bi L, Pergolesi D, Traversa E. Adv. Mater. 2012; 24: 195.
- Sase M, Hermes F, Yashiro K, Sato K, Mizusaki J, Kawada T, Sakai N, Yokokawa H, *J. Electrochem. Soc.* 2008; 155:
 B793. Yang YML, Jacobson AJ, Chen CL, Luo GP, Ross KD, Chu CW. *Appl. Phys. Lett.* 2001; 79: 776.
- [5] Mutoro E, Crumlin EJ, Biegalski MD, Christen HM, Shao-Horn Y. *Energy Environ. Sci.* 2011; 4: 3689.
- [6] Ia O' GJ, Ahn SJ, Crumlin E, Orikasa Y, Biegalski MD, Christen HM, Shao-Horn Y. Angew. Chem. Int. Ed. 2010; 49: 5344.
- [7] Burriel M, Garcia G, Santiso J, Kilner JA, Richard JCC, Skinner SJ. *J. Mater. Chem.* 2008; 18: 416.
- [8] Wang L, Merkle R, Maier J, Acarturk T, Starke U. Appl. Phys. Lett. 2009; 94: 071908.
- [9] Prestat M, Infortuna A, Korrodi S, Rey-Mermet S, Muralt P, Gauckler LJ. J. Electroceram. 2007; 18: 111. Beckel D, Muecke UP, Gyger T, Florey G, Infortuna A, Gauckler LJ. Solid State Ionics 2007; 178: 407.
- [10] van Doorn RHE, Burggraaf AJ. Solid State Ionics 2000; 128: 65.
- [11] Geller S, Bala VB. Acta Cryst. 1956; 9: 1019.
- [12] Klenov DO, Donner W, Foran B, Stemmer S, *Appl. Phys. Lett.* 2003; 82: 3427.
- [13] Chen X, Wang S, Yang YL, Smith L, Wu NJ, Kim Bl, Perry SS, Jacobson AJ, Ignatiev A. *Solid State Ionics* 2002; 146: 405. Kim G, Wang S, Jacobson AJ, Chen CL. *Solid State Ionics* 2006; 177: 1461. Karthikeyan A, Ramanathan S. *Appl. Phys. Lett.* 2008; 92: 243109.

- [14] Kim HR, Kim JC, Lee KR, Ji HI, Lee HW, Lee JH, Son JW. *Phys. Chem. Chem. Phys.* 2011; 13: 6133.
- [15] Song CR, Yoo HI. Solid State Ionics 1999; 120: 141.
- [16] Song CR, Yoo HI. Phys. Rev. B 2000; 61: 3975.
- [17] Wang S, Verma A, Yang YL, Jacobson AJ, Abeles B. Solid State lonics 2001; 140: 125.
- [18] van der Haar M, den Otter MW, Morskate M, Bouwmeester HJM, Verweij H. J. Electrochem. Soc. 2002; 149: J41.
- [19] Kawada T, Suzuki J, Sase M, Kaimai A, Yashiro K, Nigara Y, Mizusaki J, Kawamura K and Yugami H. *Journal of the Electrochem. Soc.* 2002; 149: E252.
- [20] Mizusaki J, Mima Y, Yamauchi S, Fueki K, Tagawa H. J. Solid State Chem. 1989; 80: 102.
- [21] Kim YM, He J, Biegalski MD, Ambaye H, Lauter V, Christen HM, Pantelides ST, Pennycook SJ, Kalinin SV, Borisevich AY. *Nature Materials* 2012; 11(10): 888.
- [22] Cai ZH, Kuru Y, Han JW, Chen Y, Yildiz B. J. Am. Chem. Soc. 2011; 133: 17696.
- [23] Han JW, Yildiz B. J. Mater. Chem. 2011; 21: 18983.
- [24] De Souza RA, Ramadan A, Horner A. Energy Environ. Sci.
 2012; 5: 5545. Korte C, Peters A, Janek J, Hesse D, Zakharov N. Phys. Chem. Chem. Phys. 2008; 10: 4623. Schichtel N, Korte C, Hesse D, Janek J. Phys. Chem. Chem. Phys.
 2009; 11: 3043. Schichtel N, Korte C, Hesse D, Zakharov N, Buz B, Gerthsen D, Janek J. Phys. Chem. Chem. Phys.
 2010; 12: 14596.

Feature Articles

Shape Control of Lanthanide Ion-Doped Inorganic Upconversion Nanophosphors



Ho Seong Jang Molecular Recognition Research Center msekorea@kist.re.kr

Introduction

Phosphor is a material which converts external energies, such as electron beams and photons, to visible light. Depending on the type of external energy, there are a number of luminescence mechanisms, including cathodoluminescence for excitations by electron bombardment, photoluminescence for photon absorption, electroluminescence for electric fields, thermoluminescence for thermal energy, and chemiluminescence for chemical reaction [1]. Although most phosphors exhibit a downconversion process, that is, excitation energy is higher than the energy of emitted light, some materials show an upconversion process. In upconversion luminescence, the energy of emitted light from the phosphor is higher than the excitation energy. For example, infrared light can be converted into visible light by upconversion phosphors. The inorganic phosphors consist of a host material and dopant ions. When an inorganic crystal is doped with suitable dopant ions, it can show efficient upconversion luminescence. To date, β -NaYF₄ doped with Yb³⁺ and Er³⁺ (β -NaYF₄:Yb³⁺,Er³⁺) is known as the most efficient upconversion phosphor [2]. Unlike organic fluorophores and quantum dots (QDs) which show a two-photon process, upconversion phosphors show a much higher efficiency [3]. In cases of organic fluorophores and QDs, use of a high-power laser source is necessary because efficiency is very low due to the absence of a real energy level between ground and excited states. On the other hand, lanthanide iondoped upconversion phosphors have real intermediate energy levels between their ground and excited states, and thus, upconversion efficiency is significantly higher compared with the two-photon process. In this case, upconversion phosphors can show visible light under irradiation by using an inexpensive continuous wave (cw) diode laser whose power density is much weaker than a pulsed laser. When the size of upconversion phosphors is in a nanometer scale, these phosphors are potentially useful as bio-imaging probes due to their non-blinking and non-bleaching properties [4]. In addition, they can be applied to three-dimensional volumetric display devices [5]. For the past decade, a synthetic method for fabricating nanophosphors has been well developed. Recently, the synthesis of highly bright β-NaYF₄:Yb,Er upconversion nanophosphors (UCNPs) via a solution chemical route has been reported [3, 6]. During the growth stage after nucleation in a reaction solution, the morphology of these UCNPs can be specifically tailored. The morphology of nanocrystals (NCs) is of considerable interest because it can be rationally controlled and nanoparticles with different morphologies can exhibit various physical or chemical properties depending on the exposed crystallographic planes. Although there are many reports on morphology control of QDs and metal NPs, only a few studies have reported on the morphology control of β-NaYF₄:Yb,Er UCNPs. Recently, Murray's group reported on β-NaYF₄:Yb,Er upconversion nanorods (UCNRs) and hexagonal prisms [3]. However, in that study, each morphology resulted from specific synthetic conditions without predictable control or a detailed explanation of the mechanism for the morphology evolution of β-NaYF₄:Yb,Er UCNPs. Although NaYF₄:Yb,Er UCNRs with a high aspect ratio (AR) have been synthesized using oleic acid/trioctylphosphine/octadecene solvent, the detailed mechanism for shape evolution is not yet fully understood [6]. Therefore, achieving a controlled synthesis of β-NayF₄:Yb,Er UCNRs with predictable morphologies and tunable AR via a systematically controlled synthesis method remains a challenge. Here, we demonstrate the

morphology evolution of β -NaYF₄:Yb,Er UCNPs from sphere to rod and from rod to hexagonal prism/sphere by using a surfactant, an additive, and lanthanide doping. In addition, the applicability of the β -NaYF₄-based UCNPs in display devices is shown by preparing composites that incorporate the β -NaYF₄:Yb,Er/Tm green- and blue-emitting UCNRs into polydimethylsiloxane (PDMS) polymers.

Synthesis and characterization of upconversion nanophosphors

Hexagonal β -NaYF₄:Yb,Er upconversion nanophosphors were synthesized by a solution chemical route. To prepare the UCNR-PDMS composites, 0.2 ml of the UCNR solution was mixed with 10 ml SYLGARD 184 silicone elastomer 184 followed by mixing 1 ml of a curing agent (Dow Corning Corp.) Finally, the UCNR-PDMS composites were heat-treated at 80°C after overnight aging. The sizes of the synthesized UCNPs were characterized by using a Tecnai F20 G² transmission electron microscope at an accelerating voltage of 200 kV. The photoluminescence spectra of the UCNPs were obtained with a Hitachi F-7000 spectrophotometer coupled with a near infrared (NIR) diode laser (λ = 980 nm).

Morphology evolution of upconversion nanophosphors

In our experiments, the β -NaYF₄-based UCNPs were synthesized through the thermal decomposition of Ln(oleate)₃ [Ln = Y, Yb, Er/Tm] complexes and the heating of an oleic acid (OA) and 1-octadecene (ODE) solution containing Ln(oleate)₃, Na, and F precursors. The morphology of β -NaYF₄:Yb,Er UCNPs could be controlled by adjusting the quantity of capping ligand and a certain crystallographic plane of β -NaYF₄:Yb,Er UCNPs could be stabilized by preferential adsorption of the capping ligand. Figure 1 shows transmission electron microscopy (TEM) images showing the morphology evolution of the β-NaYF₄:Yb,Er UCNPs from spheres to rods. When total solvent volume was maintained at 21 ml and the ratio of OA to ODE was adjusted, spherical morphology of the β-NaYF₄:Yb,Er UCNPs was transformed into rod shape. As the ratio of OA to ODE increased, particle size decreased, and the shape of the β-NaYF₄:Yb,Er UCNPs transformed from sphere to rod with an increasing length to width aspect ratio (AR). The particle size of the UCNPs decreased as the OA/ OED ratio increased up to 6:15 because large quantities of OA provided less opportunity for the monomers to attach to the nucleated seeds than small quantities of OA. (Figure 1a and b). When the amount of OA equaled that of ODE, an anisotropic morphology with an AR of 1.28 was obtained (Figure 1c), which indicates that OA was preferentially adsorbed onto certain crystallographic planes. A further increase in the OA/ODE ratio resulted in a rod morphology (Figures d-e). The value of AR increased from 1.00 for spherical shapes to 2.80 for rod shapes. Previously, β-NaYF₄:Yb,Er UCNPs with an elliptical shape and a low AR were obtained with 10 ml of OA. However, in our study, nanorod (NR) morphology with higher AR was produced by using Ln(oleate)₃ precursors. In our experimental conditions, more effective quantities of oleate in the solution may have contributed to the stabilization of certain crystallographic planes, even though the reaction solution contained the same quantity of OA as reported in the literature. Consequently, for the first time, we achieved β-NaYF₄:Yb,Er UCNRs via thermal decomposition and a heating up method by manipulating only one synthetic parameter. When the OA/ ODE ratio was 19:2, β-NaYF₄:Yb,Er UCNRs with a high AR of 2.80 were obtained (Figure 1e). The synthesized β-NaYF₄:Yb,Er UCNRs were highly uniform in size and shape as shown in the low magnification TEM image of Figure 1f. Figures 1g and h show the HR-TEM image of single UCNRs. Highly clear lattice fringes and spotty fast Fourier transform patterns are observed, indicating high crystallinity and the single crystalline nature of the



Figure 1. TEM images of β -NaYF₄:Yb,Er UCNPs synthesized at various ratios of OA to OED of (a) 2:19, (b) 6:15, (c) 10.5:10.5, (d) 15:6, and (e) 19:2. (f) Low magnification TEM image of β -NaYF₄:Yb,Er UCNRs. (g) HR-TEM image of β -NaYF₄:Yb,Er UCNRs shown in (e). Inset shows FFT diffractogram of selected area. (h) Enlarged HR-TEM image of selected area in (g). Scale bars represent 20 nm for panels (a)-(e). Scale bar of panel (f) represents 200 nm. Scale bars of panels (g) and (h) indicate 5 nm.

synthesized UCNRs. The spacing between lattice fringes along the NRs is estimated to be 3.56 Å, which is in good accordance with the lattice spacing between the (0001) planes of β -NaYF₄, indicating that the UCNRs grow along the c-axis, i.e., the [0001] direction. In addition, the side faces of the UCNRs are bound by the {1010} planes, judging from lattice fringes with a spacing of 5.17 Å corresponding to the $d_{(1010)}$ of β -NaYF₄ doped with Yb and Er (JCPDS 28-1192). This result is attributed to the preferential adsorption of OA onto the {1010} planes.

Exposure with $\{10\overline{1}0\}$ planes of the β -NaYF₄:Yb,Er UCNRs is also verified in the TEM images shown in Figure 2. Highly monodispersed β -NaYF₄:Yb,Er UCNRs exhibit vertically aligned, ordered arrays. At first glance, the TEM image presented in Figure 3a indicates that mixed morphologies of one-dimensional (1D) NRs and zerodimensional (0D) particles can be doubted. However, a



Figure 2. (a) TEM image of vertically aligned β -NaYF₄:Yb,Er UCNRs. (b) TEM image taken at tilt angle of 37° of the area shown in (a). Scale bars represent 50 nm.

TEM image collected at a tilt angle of 37° indicates that the hexagonal images are ascribed to a vertical alignment of the UCNRs (Figure 2b). In addition, the TEM image in Figure 3a confirms that the cross-sectional shape of the β -NaYF₄:Yb,Er UCNRs is hexagonal, which is attributed to enclosure with {1010} planes.

Similarly, various morphologies of β -phase NaYF₄:Yb_{0.29}Tm_{0.01} (β-NaYF₄:Yb,Tm) UCNPs are shown in Figure 3. As with β -NaYF₄:Yb,Er, the spherical β-NaYF₄:Yb,Tm UCNPs changed to rods when the ratio of OA to ODE in the reaction solution was increased. From the XRD measurements, all samples were confirmed to be hexagonal. (Data are not shown here.) In addition, the HR-TEM images presented in Figure 3 show clear lattice fringes in the nanospheres and the NRs, which indicate a high degree of crystallinity of the synthesized β-NaYF₄:Yb,Tm UCNPs. The spherical β-NaYF₄:Yb,Tm UCNPs shown in Figure 3c were aligned with the zone axis along the [0001] direction. When the β -NaYF₄:Yb,Tm UCNPs were synthesized in a solvent containing 19 ml OA and 2 ml ODE, an NR morphology with a high AR was also successfully formed even though increasing the Yb concentration in the host lattice favors the formation of hexagonal plates [7]. By measuring the spacing between lattice fringes along the UCNRs, the β -NaYF₄:Yb,Tm UCNRs were found to grow along the *c*-axis.

Previously, Zhang et al. reported elliptical β -NaYF₄:Yb,Er synthesized from LnCl₃ precursors [8]. However, when we followed their synthesis method –



Figure 3. TEM and HR-TEM images of β -NaYF₄:Yb,Tm UCNPs synthesized at various ratios of OA to ODE of (a) 2:19, (b, c) 6:15, (d) 15:6, and (e, f) 19:2. Scale bars are 50 nm for panels a, b, d, and e. Scale bars represent 5 nm for panels c and f.



Figure 4. (a, b) Low and high magnification TEM images, (c) scanning electron microscopy (SEM), and (d) HR-TEM image of the β -NaYF₄:Yb,Er UCNPs synthesized with LnCl₃ instead of Ln(oleate)₃ as Ln precursors (Ln = Y, Yb, and Er). Inset shows FFT diffractogram for red frame of (d). Scale bars represent 100 nm for panels (a) and (c), 20 nm for panel (b), and 5 nm for panel (d).

lanthanide chloride precursors were directly dissolved in OA and ODE with an increased volume of OA (19 ml) – the NR morphology was not obtained. Under those conditions, hexagonal prisms were obtained as shown in Figure 4. Therefore, the use of Ln(oleate)₃ precursors rather than LnCl₃ appears critical for obtaining a nanorod morphology with a high aspect ratio.

Under our experimental conditions, preferential adsorption of OA to $\{10\overline{1}0\}$ planes led to faster growth along the c axis than lateral growth. Thus, a {1010} planestabilized rod with rounded ends was developed as shown in Figures 1 and 3, although a hexagonal plate bound with {1010} and {0001} planes is a thermodynamically stable morphology of β -NaYF₄ crystals [7]. However, hexagonal plate or prism morphology of the β -NaYF₄:Yb,Er UCNPs can be formed by adding appropriate additives to the reaction solution. The β -NaYF₄:Yb,Er hexagonal prism shown in Figure 4 may be attributed to Cl⁻ ions in the reaction solution because Cl⁻ ions are known as an efficient additive for stabilizing {0001} planes of some crystals [9]. To confirm the hypothesis, a methanolic solution of NaCl was added to the reaction solution as a Cl⁻ precursor. Although according to Choi et al., any MCI (M = Na, K) is an efficient additive for stabilizing the {001} or {0001} planes, NaCl was chosen to stabilize the $\{0001\}$ planes of the β -NaYF₄:Yb,Er UCNRs because the solubility of KCI in MeOH is considerably lower than that of NaCl and Na has little effect on the morphology of NaYF₄ [10-11]. Figures 5a-d present TEM images that show the effect of Cl⁻ ions on the morphology of the β -NaYF₄:Yb,Er UCNRs. When the amount of Cl⁻ ions was less than 2.5 mmol, hexagonal prism-shaped particles were not observed because the amount of Cl⁻ was insufficient to completely stabilize the {0001} planes of the β-NaYF₄:Yb,Er UCNRs. However, the {0001} planes were stabilized and hexagonal prisms were obtained, as shown in Figure 5b, when 2.5 mmol Cl⁻ ions were introduced. Highly monodisperse hexagonal prisms exhibited a two-dimensional (2D) self-assembled monolayer as shown in Figures 5b and c. An increase of Cl⁻ ions in the reaction solution decreased the AR of the β-NaYF₄:Yb,Er hexagonal prisms from 1.51 to 1.11. However, an excess amount of Cl⁻ ions led to the formation of small spherical and polyhedral NCs with a broad size distribution (12.0 \pm 4.9 nm) with no hexagonal prisms or rods observed.



Figure 5. TEM images of NaYF₄:Yb,Er UCNPs synthesized with varying NaCl amounts: (a) 0 mmol, (b) 2.5 mmol, (c) 2.7 mmol, and (d) 3 mmol. All scale bars represent 20 nm. (e) Corresponding XRD patterns of NaYF₄:Yb,Er UCNPs.

Although hexagonal-shaped particles were observed in Figure 5d, the HR-TEM analysis revealed that the distances between the lattice fringes of the hexagonalshaped particles were in accordance with the interplanar sapcing of the α -phase NaYF₄ (Figure is not shown here.). The formation of the α -phase NaYF₄:Yb,Er was also confirmed from the XRD pattern depicted in Figure 5e. However, the β -NaYF₄:Yb,Er hexagonal prisms exhibited the β -phase NaYF₄ diffraction pattern and an enhanced (002) peak which implies that the hexagonal prisms can be preferentially aligned with the {0001} planes parallel to the substrate. The abrupt change in morphology is attributed to a phase change from the β -phase to the α -phase. This result can be explained as follows: Sui et al. reported that the bonding of oleate ions to Y³⁺ ions is favorable and induces an ordered arrangement of Y³⁺ ions which reduces the energy barrier for phase transformation from the α - to β -phase [11]. Large quantities of Cl⁻ ions in the reaction solution are hypothesized to disturb bonding of oleate ions to the Y³⁺ ions. Instead, electrostatic interactions between the oleate and the {100} planes of the NaYF₄ result in the formation of α -phase NaYF₄ [12].

Figure 6 shows the PL spectra and corresponding Commission Internationale de l'Eclairage (CIE) color coordinates of the β -NaYF₄:Yb,Er UCNPs with typical spherical, rod, hexagonal prism, and polyhedron morphologies (shown in Figures 1b, 1e, 5c, and 5d,



Figure 6. (a) PL spectra and (b) CIE color coordinates of NaYF₄:Yb,Er UCNPs with various morphologies including: (i) sphere (triangle), (ii) rods (circle), (iii) hexagonal prism (diamond), and (iv) small polyhedron (square) under the excitation of 980 nm NIR light. (c) PL spectra of β -NaYF₄:Yb,Tm UCNPs under 980 nm NIR light illumination. Insets of (b) and (c) illustrate digital camera images showing luminescence of the NaYF₄:Yb,Er/Tm UCNPs.

respectively) under excitation with 980 nm NIR light. The strong emission peaks at around 523 and 541 nm are attributed to the electronic transitions of $^2H_{11/2}$ and $^4S_{3/2} \rightarrow ^4I_{15/2}$ of Er³⁺ ions. Another emission peak at around

660 nm is attributed to the electronic transition of ${}^{4}F_{9/2} \rightarrow$ ⁴I_{15/2} of the Er³⁺ ions. Spherical, rod, and hexagonal prismshaped β-NaYF₄:Yb,Er UCNPs exhibited green emission, while small polyhedral-shaped UCNPs showed yellow emission (Figure 6b inset). The PL intensity and intensity ratio of green-to-red emission ($R_{\alpha/r}$) slightly increased when the morphology changed from sphere ($R_{g/r} = 3.6$) to rod ($R_{g/r} = 4.1$) and from rod to hexagonal prism ($R_{g/r} = 4.8$). This result is ascribed to the decrease of the surface-tovolume ratio of the UCNPs due to the increase in the NC size with the change in morphology, which is consistent with a previous result reported by Murray's group [3]. With the increase of the value of $R_{\alpha/r}$, the CIE color coordinates shifted from (0.2877, 0.6872) for the nanosphere to (0.2743, 0.7076) for the hexagonal prism (Figure 6b). In addition to the CIE color coordinates, the color purities of the UCNPs with different morphologies differed slightly from one another. The purity of color is expressed as the excitation purity and is defined as follows [13]:

$$P_{e} = \sqrt{\frac{(x - x_{w})^{2} + (y - y_{w})^{2}}{(x_{b} - x_{w})^{2} + (y_{b} - y_{w})^{2}}}$$
(1)

where x and y are the color coordinates of the UCNPs, $x_{\mbox{\tiny w}}$ and $y_{\mbox{\tiny w}}$ are the color coordinates of the standard white point, and x_b and y_b are the color coordinates of the boundary at which the line segment connecting the white point with the chromaticity of the UCNPs intersects the spectrum locus. According to Eq. (1), the excitation purities of the spherical, rod-, and hexagonal prismshaped UCNPs were calculated to be 95.4, 96.8, and 97.6%, respectively, when the standard illuminant C was considered. However, the small polyhedral UCNPs exhibited a low intensity ratio of green-to-red emission (R_{α} / $_r$ = 0.3). In this case, the low value of $R_{g/r}$ is attributed to the crystal structure of the small polyhedral NCs, that is, the formation of the α -phase rather than change in the morphology, because small β -phase UCNPs exhibit green emission with a high $R_{g/r}$ value [5, 14]. Consequently, an abrupt change in the emission color was observed and a yellow emission was generated from the small polyhedral UCNP solution, as shown in Figure 6b. In addition, the β -NaYF₄:Yb,Tm UCNPs exhibited blue upconversion luminescence attributed to the ${}^{1}G_{4} \rightarrow {}^{3}H_{6}$ transitions of the Tm³⁺ ions although the emission peak arising from the {}^{3}H_{4} \rightarrow {}^{3}H_{6} transition was the strongest (Figure 6c). The strong emission peak at approximately 808 nm did not contribute to the luminescent color considering the visual spectral sensitivity. Thus, blue light was observed from the β -NaYF₄:Yb,Tm under illumination with 980 nm NIR light.

In addition, the doping effect of Ln³⁺ ions on the morphology of the UCNRs was investigated. Figure 7 presents TEM images of the β-NaYF₄:Yb,Er,Gd UCNPs with varying quantities of Gd (Gd = 10, 15, and 20 mol%). The size of the β-NaYF₄:Yb,Er UCNRs decreased after doping with Gd. This reduction in size of the UCNR is attributed to the increase in electron charge density of the crystal surface by Gd doping, which slows the diffusion of F⁻ ions to the UCNR surface [5]. More importantly, a significant change in the shape of the β-NaYF₄:Yb,Er,Gd NCs was observed even when a small quantity of Gd was doped into the Y sites. When Gd was doped into the β-NaYF₄:Yb,Er, small UCNRs were expected. However, the AR of β -NaYF₄:Yb,Er,Gd decreased from 2.80 to nearly 1.00, and finally, spherical morphology was formed when β-NaYF₄:Yb,Er was doped with 0.2 mmol of Gd. A further increase in the Gd amount resulted in polydispersed NCs. Small particles coexisted with large ones, and spherical NCs were observed together with NRs. When the Gd concentration increases, Ostwald ripening of the NCs dominates and the monodispersity in the size distribution decreases [15]. Because the growth mechanism of NaGdF₄ differs from that of NaYF₄, NaGdF₄ NCs grow faster than NaYF₄ NCs [15]. Thus smaller quantities of OA should be used to synthesize monodispersed NaGdF₄ NCs. In this study, a high ratio of OA to ODE was used to develop rod shapes, which caused a decrease in the monodispersity of the NaYF4:Yb,Er,Gd under high Gddoping conditions. Therefore, under these experimental conditions for synthesizing β-NaYF₄:Yb,Er,Gd UCNRs,



Figure 7. TEM images of β -NaYF₄:Yb,Er,Gd UCNPs doped with various Gd amounts of (a) 0 mmol, (b) 0.1 mmol, (c) 0.15 mmol, (d) 0.2 mmol, (e) large area TEM image of β -NaYF₄:Yb,Er,Gd (0.2 mmol of Gd). Inset shows FFT diffractogram for the selected area marked with a white line in panel (e). Scale bars represent 20 nm for panels (a)-(d) and 200 nm for panel (e).

the quantity of Gd should be restricted to 0.15 mmol to maintain a uniform anisotropic morphology of the UCNRs. When the solvent was slowly evaporated from the β -NaYF₄:Yb,Er,Gd solution, a spherical UCNP superlattice monolayer was formed as shown in Figure 7d. Because the β -NaYF₄:Yb,Er,Gd UCNPs are highly monodispersed, a UCNP superlattice monolayer with a large area can be

obtained by more precise manipulation of the solvent evaporation rate and the UCNP solution concentration. In the case of β -NaYF₄:Yb,Er,Gd doped with 0.2 mmol of Gd, a large-area NC superlattice was formed, as shown in Figure 7e. The spotty pattern in the FFT diffractogram for the selected area in Figure 7e indicates that the β -NaYF₄:Yb,Er,Gd NCs are assembled into a closepacked hexagonal array (Figure 7e inset) [16].

Upconversion nanorods-PDMS composites

Small spherical UCNPs can reportedly be incorporated into the PDMS monolith [5]. However, the color purity of the spherical UCNP is less than that of the rod- and hexagonal prism-shaped UCNPs. Because the color of the light emitted from rod- and hexagonal prism-shaped UCNPs (upconversion hexagonal prisms, UCHPs) is closer to spectrally pure green than that of the spherical UCNPs, UCNR/UCHP-PDMS composites will show green light with higher color purity compared with previously reported UCNP-PDMS composites. To the best of our knowledge, large UCNPs with anisotropic morphology have not been incorporated into the PDMS monolith. Thus, in this study, the possibility of incorporating the greenand blue-emitting B-NaYF4:Yb,Er/Tm UCNRs into the PDMS monolith was investigated. Although the UCNRs are larger than the spherical UCNP particles, highly transparent UCNR-PDMS composites were prepared. The transmittance of NaYF₄:Yb,Er/Tm UCNR-PDMS composites was higher than 85% in a visible spectral range (Figure 8). Hexagonal prism-shaped β-NaYF₄:Yb,Er UCNPs were also incorporated into PDMS. Although the β-NaYF₄:Yb,Er UCHPs-PDMS composites were transparent to the naked eye, their transmittance (> 75% in visible region) was less than that of the UCNR-PDMS composites due to the scattering effect resulting from the large volume of the UCHPs [17]. The Figure 8 inset shows luminescence images of the β-NaYF₄:Yb,Er/Tm UCNR-PDMS composites under illumination with a 980 nm NIR



Figure 8. Transmittance spectra of UCNR-PDMS composites. Inset shows photographs of transparent UCNR-PDMS composites under room light (left) and NIR illuminated with cw laser (right). [(i, iii) β -NaYF4:Yb,Tm UCNR-PDMS and (ii, iv) β -NaYF4:Yb,Er UCNR-PDMS composites]

diode laser. The UCNR-PDMS composites exhibited homogeneous, bright green and blue light under NIR illumination, which implies that β-NaYF₄:Yb,Er/Tm UCNRs were well dispersed in the PDMS monolith. These results indicate that highly transparent UCNR-PDMS composites can be applied to a three-dimensional (3D) volumetric optical display [18]. More importantly, the higher color purity of the UCNRs compared to the spherical UCNPs allows the UCNR-PDMS composites to better embody spectrally pure green light in three-color 3D display.

Conclusion

The morphology of β -NaYF₄:Yb,Er UCNPs synthesized via a solution chemical route was rationally tailored using a surfactant, an additive, and lanthanide doping. The size and morphology of β -NaYF₄:Yb,Er/Tm were controlled by varying the quantity of the OA ligand. The size of the spherical UCNPs decreased when the ratio of OA to ODE was increased to 6:15, and the AR of the UCNPs increased upon a further increase of the OA/ODE ratio to 19:2. A rod shape was developed by stabilizing the {1010} planes by the OA ligand. To stabilize the {0001} planes,

NaCl was used as a Cl precursor and a hexagonal prism bound with {0001} and {1010} planes was formed. As the amount of Cl⁻ ions increased, the AR of the hexagonal prism decreased from 1.52 to 1.11. Additionally, doping with Gd led to a simultaneous change in the size and morphology of β-NaYF₄:Yb,Er UCNPs. The rod shape of the β -NaYF₄:Yb,Er UCNPs was transformed into a small spherical shape by doping with Gd because both the size and AR decreased. As a consequence, the morphology of the UCNPs could be systematically controlled from large spheres (37.9 nm) to rods, and then from rods to hexagonal prisms and small spheres (14.0 nm) by using a surfactant, an additive, and lanthanide doping, respectively. In addition, highly transparent β-NaYF₄:Yb,Er/ Tm UCNRs and PDMS composites were prepared and showed bright green and blue light under illumination with an NIR diode laser (980 nm). This result indicates that these UCNR-PDMS composites are promising for use in 3D volumetric displays.

Note

This article and its images are cited from "Rational morphology control of β -NaYF₄:Yb,Er/Tm upconversion nanophosphors using ligand, additive, and lanthanide doping" in *Nanoscale*, vol. 5 (10), pp. 4242-4251.

<u>References</u>

- Feldmann C, Jüstel T, Ronda CR, Schmidt PJ. Adv. Funct. Mater. 2003; 13 (7); 511-516.
- [2] Krämer KW, Biner D, Frei G, Güdel HU, Hehlen MP, Lüthi SR. *Chem. Mater.* 2004; 16 (7); 1244-1251.
- [3] Ye X, Collins JE, Kang Y, Chen J, Chen DTN, Yodh AG, Murray CB. *Proc. Natl. Acad. Sci. U.S.A.* 2010; 107 (52); 22430-22435.
- [4] Wu S, Han G, Milliron DJ, Aloni S, Altoe V, Talapin DV, Cohen BE, Schuck PJ. *Proc. Natl. Acad. Sci. U.S.A.* 2009; 106 (27); 10917-10921.

- [5] Wang F, Han Y, Lim CS, Lu Y, Wang J, Xu J, Chen H, Zhang C, Hong M, Liu X. *Nature* 2010; 463 (7284); 1061-1065.
- [6] Shan J, Ju Y. Appl. Phys. Lett. 2007; 91 (12); 123103.
- [7] Mai H-X, Zhang Y-W, Si R, Yan Z-G, Sun L-D, You L-P, Yan C-H. *J. Am. Chem. Soc.* 2006; 128 (19); 6426-6436.
- [8] Li Z, Zhang Y, Jiang S. Adv. Mater. 2008; 20 (24); 4765-4769.
- [9] Choi K-S, Jang HS, McShane CM, Read CG, Seabold JA. MRS Bull. 2010; 35 (10); 753-760.
- [10] Read CG, Steinmiller EMP, Choi K-S. J. Am. Chem. Soc. 2009; 131 (34); 12040-12041.
- [11] Sui Y, Tao K, Tian Q, Sun K. J. Phys. Chem. C 2012; 116
 (2); 1732-1739.
- [12] Ostrowski AD, Chan EM, Gargas DJ, Katz EM, Han G, Schuck PJ, Milliron DJ, Cohen BE. Acs Nano 2012; 6 (3); 2686-2692.
- [13] enwikipedia.org/wiki/Colorfulness (accessed June).
- [14] Jang HS, Lim K, Woo K. Opt. Express 2012; 20 (15); 17107-17118.
- [15] Johnson NJJ, Oakden W, Stanisz GJ, Prosser RS, van Veggel FCJM. *Chem. Mater.* 2011; 23 (16); 3714-3722.
- [16] Feng W, Sun L-D, Zhang Y-W, Yan C-H. Small 2009; 5 (18); 2057-2060.
- [17] Park HK, Oh JH, Do YR. *Opt. Express* 2012; 20 (9); 10218-12208.
- [18] Boyer JC, Johnson NJJ, van Veggel FCJM. *Chem. Mater.* 2009; 21 (10); 2010-2012.



Green Nano Materials Laboratory

[Technical Review]

Environmental Nanobiosensors Based on Aptamer Technology



Byoung Chan Kim Center for Environment, Health and Welfare Research bchankim@kist.re.kr



Jongsoo Jurng Center for Environment, Health and Welfare Research jongsoo@kist.re.kr

Introduction

Technical advances over the past decade in nanotechnology have spurred research efforts dedicated to the fabrication of nanomaterials and the manipulation of nanotechnology. One area of this research is the development of advanced environmental nanobiosensors for environmental monitoring, an increasingly important issue as pollution affects both human and ecological health. Fast, simple and more reliable identification and diagnosis of environmental analytes would greatly help in protecting the environment against pollution and would assist in the establishment of appropriate policies for pollution prevention. To aid in monitoring, biosensors are potentially useful tools meant to provide for the selective detection and identification, with high sensitivity and specificity, of hazardous materials in industrial products and environmental samples (e.g. air, soil, and water), as well as harmful microorganisms (bacteria or viruses) in biological matter [1-5].

Biosensors are composed of biological receptors (e.g. DNA, antibodies, enzymes, or whole cells) that recognize analytes via the epitopes of specific receptors and signal transducers that provide quantitative, semi-quantitative or qualitative information for converting interaction events between receptors and analytes to readable signals. The interaction of biological receptors with analytes produces various signals, such as electrical current, color, light emission, mass change, or fluorescence, depending on the design of the biosensors. These signals are converted to appropriate measurable and



Figure 1. Flow chart for the composition of environmental biosensors in environmental monitoring.

readable responses. The term "environmental biosensors," therefore, refers to biosensors that can diagnose and identify pollutants and substances in the environment for environmental monitoring purposes (Figure 1). Environmental biosensors should be seen as distinct from bioanalytical systems, which require additional processing steps such as reagent addition or sample preparation prior to detection, and as a result, are not as suitable for rapid on-site monitoring [3, 6]. The merits of environmental biosensors over conventional instrument analysis include the possibility of miniaturization, portability, rapid identification, minimal sample preparation, and minimal processing steps. Environmental biosensors can offer analyte information to determine not only specific chemicals, but also their biological effects, such as toxicity, depending on the specific receptors used in the biosensors. These considerations are important to the development of new types of environmental biosensors. To accurately detect environmental analytes found in complex environmental samples, environmental biosensors should have sufficient sensitivity and specificity. To enable this, samples should be concentrated and cleaned via filtration so that the recognition elements can identify the target analytes without hindrance by debris or an unknown matrix [7]. The specificity and sensitivity of environmental biosensors are determined

by the degree of interaction between recognition receptors and analytes and signal transduction translated during their binding event. Recently the development of environmental biosensors has focused on enhancing the degree of interaction between recognition receptors and analytes, and also on the sensitivity of receptors as they are combined with various nanoparticles (NPs) such as quantum dots [8, 9], gold NPs [10, 11], or dye-doped silica [12, 13]. As nanotechnology continues to develop, various NPs are being developed and used in fields such as energy, biotechnology, pharmaceutics, and electronics. Proponents of environmental monitoring have also been interested in using nanomaterials for the construction of environmental nanobiosensors. Usually, NP-based environmental nanobiosensors use a conjugation scheme between NPs and recognition receptors. The NPs in environmental nanobiosensors usually generate and enhance signals. In nanomaterial-based environmental nanobiosensors, the recognition receptors are immobilized in the surface of the nanomaterials, and the interaction of conjugate with analytes is monitored via a signal transduction mechanism. A variety of nanomaterial-embedded nanotechnologies is currently used in environmental nanobiosensor applications, and these nanomaterials influence both the type of recognition element employed and the signal transduction method utilized [7]. Many kinds of NPs that have unique properties for host materials and signal-enhancing elements have been developed as efficient supporters for the conjugation of receptors. One of the particularly advantageous features of using NPs is the ability to uniformly control size at the nanometer scale, resulting in higher surface area to volume ratios [14]. The uniform size of NPs and their compatibility in size with receptors used in environmental biosensors, together with high surface areas, offer new functions in the sensing process, such as a high loading capacity of receptors and the generation of high and stable signals. The capacity to control the surface of NPs with various functional groups and conjugation chemistry opens the door for the preparation of more sensitive and

stable nanobiosensors for environmental applications.

Regardless of what combination of receptors and NPs is applied, environmental nanobiosensors in the field should be able to discriminate between targets meant to be detected exactly and non-targets. They should have a broad detectable range to quantify the concentration of analytes as well as high stability during operation [7]. To apply environmental nanobiosensors in real situations, the biosensors should be tested under harsh environmental conditions. Many concepts for environmental biosensors using NPs have been suggested; however, most of them are suitable only under restricted laboratory conditions. Usually environmental nanobiosensors are considered for a single use only, and are unable to monitor analytes continuously on site. However, with appropriate system composition adjustment, biosensors can make continuous environmental monitoring possible. Despite such powerful advantages, the application of biosensors in the environmental field is still limited in comparison to medical or pharmaceutical applications, where most research and development has occurred. The majority of the systems developed are prototypes that still need to be validated before being commercialized or used extensively.

In order to develop suitable nanobiosensors for environmental applications, it is important to select appropriate receptors and nanomaterials. Among many candidates, we focus here on recent progress in the development of nanobiosensors using functional nucleic acid-based aptamers and nanomaterials for the detection of heavy metals or pathogens which are considered difficult to detect and identify on site using current technology, specifically in complex environmental samples. We note that an entire field related to environmental nanobiosensors is needed to develop methodologies for sample preparation. Our focus, however, is solely on recent "hot" bioreceptors, especially aptamers, and their combination with nanomaterials to detect heavy metals and pathogens.

The use of aptamers and nanomaterials for constructing environmental nanobiosensors

Metal NPs, such as gold NPs, have properties distinct from bulk metal particles, including size, surface functionality, and shape [15, 16]. In particular, the high stability and unique observable properties of gold NPs have made them good candidates for the enhancement of signal transduction and as carriers for bioreceptors. This has contributed to the development of nanobiosensors in a wide range of research fields including the quality monitoring of food, medical diagnosis and treatment, and environmental monitoring [17]. The most widely used method for synthesizing gold NPs of different sizes and shapes originated from a variation on the classic Turkevich-Frens citrate reduction route [18]; this modification allowed mass production of various gold NPs. In the citrate reduction route, a sodium citrate solution is added to a boiling solution of chloroauric acid (HAuCl₄) and after a few minutes, the color of solution changes from colorless to a deep red that suggests the formation of gold NPs. The cetyltrimethyl ammonium bromide (CTAB) gold NPs are frequently used as seeds for synthesizing monodispersed gold nanorods with diverse aspect ratios and facets for controlling the concentration of CTAB [19, 20]. Gold NPs as nanobiosensors using functional nucleic acids were introduced in 1996 by the Mirkin group [21]. In their report, the oligonucleotidetailed 13 nm gold NPs via thiol group capping and the complementary target DNA induced the aggregation of gold NPs. The aggregation of gold NPs induced the color change of solutions from red to blue. The reverse reaction, dehybridization of DNA (from blue to red), is possible when temperature is controlled. This simply observable and reversible process initiated the use of gold NPs as DNA detection agents. The selection of appropriate receptors and their combination with gold NPs is important to realize the sensing platforms that can be applied to environmental diagnosis with high rapidity and specificity through a colorimetric scheme. Colorimetric environmental nanobiosensors are useful because they can minimize the necessity for expensive instruments. Although the first application of gold NPs in a diagnosis scheme was based on DNA detection via complementary hybridization, many analytes can be detected in simple color changes visible to the naked eye, making the scheme preferable to DNA hybridization if the other receptors can be linked with gold NPs. A good candidate for linking with gold NPs is a functional nucleic acid, or aptamer. Aptamers are small nucleic acid sequences (usually 40-90 bp of DNAs or RNAs) that specifically bind to organic or inorganic molecules or to marcromolecules such as proteins with high affinity constants that lie in the micromolar to nanomolar ranges [22-25]. The nucleic acid aptamers fold into a three-dimensional configuration to their specific targets. Aptamer selection begins with a single-stranded random library of oligonucleotide (~10¹⁵ different single strands) and target substrate through a series of selection and amplification of oligonucleotides, known as the systematic evolution of ligands by the exponential enrichment (SELEX) method [23]. During the SELEX process, the few nucleic acids that have strong binding affinity are retained, while most in the rest of the library are removed. After their sequencing, selected aptamers can be synthesized in large quantities and are available for immobilization with other supports for modifying their 3' or 5' end. The SELEX process is simpler than antibody production; however, one limitation related to using aptamers in environmental nanobiosensors is nuclease that degrades DNA. Nevertheless, aptamers are widely accepted in finding appropriate receptors to monitor environmentally relevant chemicals, biomolecules, and pathogens because of their simple process and high affinity. Various aptamers for environmental analytes have been isolated, such as heavy metals [26, 27], pesticides [28], pathogens [29-31], toxins [32-34], and pharmaceuticals [35, 36]. The isolation of aptamers in various substrates and their application to environmental nanobiosensors continue to be widely reported (Figure 2).



Figure 2. Aptamer-based environmental nanobiosensors for various environmental analytes.

The combination of nucleic acid aptamers and gold NPs has allowed many analytes to be detected with simple strategies. Liu and Lu [37] aptly summarized the strategy of constructing aptamer and gold NP-mediated colorimetric sensors as follows. The aptamer-linked gold NPs are already aggregated and form a purple color. The purple-colored aggregates undergo fast disassembly to a red-colored dispersion of nanoparticles when the target analytes bind to their specific aptamers. The targets bound to aptamers overcome the aggregate interaction among gold NPs which releases aggregated gold NPs. This strategy is the opposite of the originally introduced method for fabricating gold NP-mediated colorimetric sensors. Either way, both methods demonstrated the useful application of gold NPs linked with DNA receptors as colorimetric environmental nanobiosensors.

Aptamer-based nanobiosensors for heavy metal detection

Among environmental analytes, heavy metal biosensors are the most applied, using a combination of aptamers and gold NPs in the colorimetry method. The detection of heavy metal ions in environmental samples is an important issue because these contaminants have severe effects on human health and environmental ecosystems. The accumulation of heavy metals in the human body cause damage to the brain, heart, lungs, kidneys, the central nervous system and the immune system [38, 39].

The highly selective and sensitive colorimetric detection of Hg²⁺ using aptamer-conjugated gold NPs in an aqueous environment has been demonstrated. Two different DNA probes conjugated with gold NPs were prepared (Probe A: A10-T-A10, Probe B: T10-T10) and the single mismatched T-T sites between the two different probes could be hybridized because Hg²⁺ selectively coordinates the T-T mismatch. The result of the coordination triggered by Hg2+ that selectively bound to the T-T sites led to the formation of aggregated gold NPs, changed the color of the solution from red to purple and raised the T_m (melting temperature) to dissociate aggregation [40]. Upon increasing the Hg²⁺ concentration in aqueous solutions, the T_m of aggregated gold NPs proportionally increased. This method provided a 100 nM limit of detection (LOD) in aqueous environments. Following that principle, a chip-based scanometric detection method for Hg²⁺ was suggested [41]. One probe (T10-T-T10) was immobilized on the plate, in this case glass, and the other probe (A10-T-A10) was conjugated with gold NPs. The presence of Hg²⁺ initiated the conjugation between probes immobilized on the plate, and the probes conjugated with gold NPs. After conjugation by Hg²⁺, the signals were amplified through the silver enhancement scheme. This chip could differentiate the Hg²⁺ in the presence of 15 other metal ions as well and had a 10 nM LOD.

A simple, rapid and colorimetric method for the detection of Hg^{2+} using electrostatic repulsion among aptamer-conjugated gold NPs has also been demonstrated [42]. In this method, a randomly-coiled poly T_n DNA aptamer adsorbed onto 13 nm gold NPs via electrostatic attraction. The gold NPs wrapped with DNA aptamers existed in the solution homogeneously because of electrostatic repulsion initiated by the high negative charges of aptamers in a specific salt condition. The presence of Hg^{2+} in this solution led to a folded structure of the poly T_n DNA aptamer because of the high affinity of Hg^{2+} to poly T_n DNA. The detachment of poly T_n DNA from gold NPs induced the loss of repulsion among gold NPs, and the color change of the solution from red to purple resulted in the aggregation of gold NPs. The LOD in this method was determined to be 250 nM. This strategy omits the conjugation step between aptamers and nanoparticles, reducing the time and cost of preparing the detection assay.

Another detection method for Hg²⁺ with aptamers and gold NPs introduced the use of a fluorescence resonance energy transfer (FRET) scheme [43]. The Hg²⁺-specific aptamer was linked with guantum dot (QD) NPs and the QD-conjugated aptamers were adsorbed onto gold NPs via electrostatic attraction. The superguenching effect of gold NPs reduces the fluorescence intensity of QDs in close proximity. However, the presence of Hg2+ induced the detachment of QD-conjugated aptamers from the gold NPs and led to the increase of fluorescence intensity of the QDs. The LOD of this method for Hg²⁺ was 500 nM. Another heavy metal ion, Pb2+, can also be detected by colorimetric sensors constructed in the combination of aptamers and gold NPs. Liu and Lu [44, 45] used a Pb²⁺-specific DNAzyme aptamer, or DNA molecules with catalytic activity to Pb2+ and isolated by the SELEX process. The colorimetric Pb2+ sensors consisted of a Pb²⁺-specific DNAzyme aptamer, its substrate strand DNA, and 5'-thiolmodified DNA (12-mer) attached on gold NPs. The substrate strand DNA had flanking regions which could hybridize with 12-mer DNA on gold NPs, complementary regions with DNAzymes aptamer and the cleavage site. The composites of these three components made gold NPs aggregate. The presence of Pb²⁺ in the DNAzyme aptamer catalyzed the hydrolytic cleavage of the substrate strand and prevented the aggregation of gold NPs, resulting in a transition to a red color from a purple color. Depending on the composition of selected DNAzyme aptamers, the sensitivity of the Pb²⁺ sensor can be controlled, and the LOD of this sensor to Pb2+ is in the sub-micromolar level. As demonstrated in the research efforts described above, if we isolate specific aptamers to different heavy metal ions, the combination of aptamers and gold NPs can offer effective detection of heavy metals in environmental samples.

Aptamer-based nanobiosensors for pathogen detection

While many aptamers have been isolated to proteins and chemicals, only a few have been isolated to bacterial cells. The direct real-time detection of living bacterial cells, especially pathogens, in environmental samples is important to prevent and control infectious diseases. Culturing methods and the PCR-based DNA amplification method are time-consuming and tedious, and their field application is limited because of the complexity of samples in the environment. It is essential to secure receptors that can recognize specific target bacteria in order to construct sensors that can detect bacterial cells in real time. The amplification of nucleic acid in bacteria using PCR or immunoassay has been commonly used for the detection of environmental pathogens. Recently, there have been demands for improvement of these methods for real-time detection of pathogens using alternative receptors that are more robust and selective than antibodies. Aptamers can be alternative receptors as they have many advantages over antibodies in detecting whole cells [46]. Despite the specificity of aptamers, very few aptamers for bacterial pathogens have been isolated and used for the purpose of biosensors. Bacterial aptamers have been isolated via the cell-SELEX method, which was developed to select aptamers for tumor cells [47-49]. The process of cell-SELEX is similar to the process for the isolation of chemicals or proteins; however, the immobilization of analytes can be omitted. The next few examples detail a good direction for environmental nanobiosensors using aptamers for real-time detection of bacterial pathogens in environmental samples.

Aptamers for non-pathogenic Escherichia coli (E. coli) DH5 α were isolated and loaded into biosensors composed of a single-wall carbon nanotube field effect transistor (SWCNT-FET). The specificity of the E. coli aptamer meant that it could recognize only E. coli in SWCNT-FET biosensors; another bacteria, S. typhimurium, didn't respond on the biosensor [50]. Although the sensitivity of SWCNT-FET was less than a two-order magnitude compared to the conventional MNP method, it had some merit for successfully detecting living bacterial cells. However, it was still ineffective at realtime detection of bacterial cells. Rius et al. developed a potentiometric biosensor composed of aptamer-SWCNT hybrid materials for detecting Salmonella Typhi (ST) in one single CFU level in real time [51]. The aptamer that could selectively recognize ST was modified with an amine group at the 3'-end and immobilized covalently with carboxylated SWCNTs that were already sprayed onto the surface of a glassy carbon rod that made contact with the potentiometer. The aptamer-SWCNT hybrid material generated a change of electromotive force when the target bacteria was present in samples, and acted as both the sensing and the transducing element for the biosensor. The recognition of target bacteria induced a conformational change of the aptamer attached onto the SWCNTs and this initiated a charge change to the SWCNTs. This nanobiosensor could be regenerated by dissociating the aptamers from bacteria in a specific salt condition, allowing new measurements. As an extension of this method, this type of biosensor was applied in complex samples to detect E. coli [52]. This report demonstrated that a particular bacterial strain can be detected in real samples that were pretreated. The most interesting part of this achievement is that the developed nanobiosensor was equipped with an on-line pretreatment system that removed the matrix in real samples. Various unknown matrices or compounds may interact with receptors non-specifically and induce non-specific signals in biosensors. Therefore, the pre-treatment of real samples is important to enhance the sensitivity and



Figure 3. Cover article by our KIST research group in Analytical Biochemistry. Reports on newly isolated aptamers targeting E. coli.



Figure 4. Binding saturation curve of selected ssDNA sequences to the target *E. coli* strain and estimation of their dissociation constants (K_d) by nonlinear regression analysis.

accuracy of biosensors in real field applications.

Recently, our group isolated four new aptamers with high affinity to *E. coli* originating in fecal samples [53] (Figure 3). We isolated and characterized single-stranded DNA (ssDNA) aptamers against *E. coli*. A total of 28 ssDNAs were isolated after 10 rounds of selection using a bacterial cell–SELEX process. Other bacterial species (*Klebsiella pneumoniae*, *Citrobacter freundii*, *Enterobacter aerogenes*, and *Staphylococcus epidermidis*) were



Figure 5. Fluorescence microscope images of *E. coli* (KCTC 2571) stained with fluorescein-labeled ssDNA aptamers (E1, and E2), and the fluorescein-labeled E4, which has no affinity to *E. coli* (scale bar: 50 µm).



Figure 6. Fluorescence intensity of cell suspensions obtained by the interaction with single aptamer or aptamer cocktails.

used for counter selection to enhance the selectivity of ssDNA aptamers against *E. coli*. Finally, four ssDNA aptamers showed high affinity and selectivity to *E. coli*. The dissociation constants (K_d) of these four ssDNA aptamers to *E. coli* were estimated to range from 12.4 to 25.2 nM (Figure 4). These aptamers did not bind to other bacterial species, including four counter cells, but they showed affinity to different *E. coli* strains (Figure 5). Further, we examined the binding capacity of an aptamer mixture (aptamer cocktail) composed of various combinations of 3 different DNA aptamers isolated from *E. coli* and compared it to one of the single aptamers using fluorescence-tagged aptamers. The aptamer mixtures showed a higher fluorescence signal than did any single aptamer used, which suggests that the use of aptamer mixtures can enhance the sensitivity of the detection of microbial cells (Figure 6). Based on these high-affinity aptamers, our group is trying to develop a real-time *E. coli* monitoring system for field application.

Concluding remarks

The abundance of hazardous pollutants in environmental systems and the widespread usage of new chemicals and industrial pollutants make it hard to detect these substances using conventional approaches. Fortunately, functional nucleic acids-based aptamers that can be isolated from various substances with high affinity are showing considerable promise for identifying and monitoring target substances. The combination of newly isolated aptamers with nanomaterials will further enable the introduction of new types of nanobiosensors for environmental application. This article provides only a brief outline of several types of nanobiosensors using aptamers and nanomaterials. New strategies will be forthcoming to diagnose environmental analytes that cannot be identified with present technology.

References

- Vanderlaan M, Watkins BE, Stanker L. Environ. Sci. Technol. 1988; 22: 247-254.
- [2] Pancrazio JJ, Whelan JP, Borkholder DA, Ma W, Stenger DA. Ann. Biomed. Eng. 1999; 27: 697-711.
- [3] Palchetti I, Mascini M. Analyst. 2008; 133: 846-854.
- [4] Baeumner AJ. Anal. Bioanal. Chem. 2003; 377: 434-445.
- [5] Amine A, Mohammadi H, Bourais I, Palleschi G. *Biosens Bioelectron.* 2006; 21: 1405-1423.
- [6] Rodriguez-Mozaz S, de Alda MJL, Barcelo D. Anal. Bioanal. Chem. 2006; 386: 1025-1041.
- [7] Vikesland PJ, Wigginton KR. *Environ. Sci. Technol.* 2010; 44: 3656-3669.
- [8] Liu JW, Lee JH, Lu Y. Anal. Chem. 2007; 79: 4120-4125.

- [9] Hansen JA, Wang J, Kawde AN, Xiang Y, Gothelf KV, Collins G. J. Am. Chem. Soc. 2006; 128: 2228-2229.
- [10] Guo SJ, Dong SJ. Trac-Trends Anal. Chem. 2009; 28: 96-109.
- [11] Polsky R, Gill R, Kaganovsky L, Willner I. Anal. Chem. 2006; 78: 2268-2271.
- [12] Estevez MC, O'Donoghue MB, Chen XL, Tan WH. Nano Res. 2009; 2: 448-461.
- [13] Tallury P, Payton K, Santra S. *Nanomedicine*. 2008; 3: 579-592.
- [14] Kim JB, Grate JW, Wang P. *Trends Biotechnol.* 2008; 26: 639-646.
- [15] Daniel MC, Astruc D. Chem. Rev. 2004; 104: 293-346.
- [16] Sun YG, Xia YN. Science. 2002; 298: 2176-2179.
- [17] Rosi NL, Mirkin CA. Chem. Rev. 2005; 105: 1547-1562.
- [18] Frens G. Nature Physical Science. 1973; 241: 20-22.
- [19] Huang YZ, Wang WZ, Liang HY, Xu HX. *Cryst. Growth Des.* 2009; 9: 858-862.
- [20] Ming T, Feng W, Tang Q, Wang F, Sun LD, Wang JF, Yan CH. J. Am. Chem. Soc. 2009; 131: 16350-16351.
- [21] Mirkin CA, Letsinger RL, Mucic RC, Storhoff JJ. Nature. 1996; 382: 607-609.
- [22] Ellington AD, Szostak JW. Nature. 1992; 355: 850-852.
- [23] Tuerk C, Gold L. Science. 1990; 249: 505-510.
- [24] Willner I, Zayats M. Angew. Chem. Int. Edit. 2007; 46: 6408-6418.
- [25] Bock LC, Griffin LC, Latham JA, Vermaas EH, Toole JJ. Nature. 1992; 355: 564-566.
- [26] Smirnov IV, Kotch FW, Pickering IJ, Davis JT, Shafer RH. Biochemistry. 2002; 41: 12133-12139.
- [27] Kim M, Um HJ, Bang S, Lee SH, Oh SJ, Han JH, Kim KW, Min J, Kim YH. *Environ. Sci. Technol.* 2009; 43: 9335-9340.
- [28] He JA, Liu YA, Fan MT, Liu XJ. J. Agric. Food Chem. 2011; 59: 1582-1586.
- [29] Fischer N, Tarasow TM, Tok JBH. Curr. Opin. Chem. Biol. 2007; 11: 316-328.
- [30] Li H, Ding XH, Peng ZH, Deng L, Wang D, Chen H, He QZ. *Can. J. Microbiol.* 2011; 57: 453-459.
- [31] Stratis-Cullum DN, McMasters S, Pellegrino PM. Anal. Lett. 2009; 42: 2389-2402.

- [32] Bruno JG, Richarte AM, Carrillo MP, Edge A. Biosens. Bioelectron. 2012; 31: 240-243.
- [33] Wang LB, Ma WW, Chen W, Liu LQ, Ma W, Zhu YY, Xu LG, Kuang H, Xu CL. *Biosens. Bioelectron.* 2011; 26: 3059-3062.
- [34] Lamont EA, He LL, Warriner K, Labuza TP, Sreevatsan S. Analyst. 2011; 136: 3884-3895.
- [35] Hu XG, Mu L, Zhou QX, Wen JP, Pawliszyn J. Environ. Sci. Technol. 2011; 45: 4890-4895.
- [36] Song KM, Cho M, Jo H, Min K, Jeon SH, Kim T, Han MS, Ku JK, Ban C. Anal. Biochem. 2011; 415: 175-181.
- [37] Liu J, Lu Y. Nat. Protoc. 2006; 1: 246-252.
- [38] Jarup L. Br. Med. Bull. 2003; 68: 167-182.
- [39] Hu XG, Tulsieram KL, Zhou QX, Mu L, Wen JP. *Toxicol. Lett.* 2012; 208: 69-74.
- [40] Lee JS, Han MS, Mirkin CA. Angew. Chem. Int. Edit. 2007; 46: 4093-4096.
- [41] Lee JS, Mirkin CA. Anal. Chem. 2008; 80: 6805-6808.
- [42] Liu CW, Hsieh YT, Huang CC, Lin ZH, Chang HT. *Chem. Commun.* 2008; 2242-2244.
- [43] Kim YS, Jurng J. Analyst. 2011; 136: 3720-3724.
- [44] Liu JW, Lu Y. J. Am. Chem. Soc. 2003; 125: 6642-6643.
- [45] Liu JW, Lu Y. J. Am. Chem. Soc. 2004; 126: 12298-12305.
- [46] Ngundi MM, Kulagina NV, Anderson GP, Taitt CR. Expert Rev. Proteomics. 2006; 3: 511-524.
- [47] Daniels DA, Chen H, Hicke BJ, Swiderek KM, Gold L. Proc. Natl. Acad. Sci. USA. 2003; 100: 15416-15421.
- [48] Cerchia L, Duconge F, Pestourie C, Boulay J, Aissouni Y, Gombert K, Tavitian B, de Franciscis V, Libri D. *PLoS. Biol.* 2005; 3: 697-704.
- [49] Fang XH, Tan WH. Accounts Chem. Res. 2010; 43: 48-57.
- [50] So HM, Park DW, Jeon EK, Kim YH, Kim BS, Lee CK, Choi SY, Kim SC, Chang H, Lee JO. *Small.* 2008; 4: 197-201.
- [51] Zelada-Guillen GA, Riu J, Duzgun A, Rius FX. Angew. Chem. Int. Edit. 2009; 48: 7334-7337.
- [52] Zelada-Guillen GA, Bhosale SV, Riu J, Rius FX. Anal. Chem. 2010; 82: 9254-9260.
- [53] Kim YS, Song MY, Jurng J, Kim BC. *Anal. Biochem.* 2013; 436: 22-28.

Fusion Environmental Technology Lab, Center for Environment, Health and Welfare Research

Feature Articles

Polycrystalline Nanowires of Gadolinium-Doped Ceria via Random Alignment Mediated by Supercritical Carbon Dioxide



Sang Woo Kim Clean Energy Research Center swkim@kist.re.kr



Byung Hyuk Kim

Clean Energy Research Center 022804@kist.re.kr



Hee Chul Yeom Clean Energy Research Center chuleno1@naver. com



Jae-Pyoung Ahn Advanced Analysis Center jpahn@kist.re.kr

Introduction

Ceria (cerium oxide) has been extensively applied in fuel cells [1], solid media for chemical mechanical planarization [2], sensors [3], three-way catalysts for the elimination of toxic vehicular gases [4], and other catalysts [5]. Nanostructured CeO₂ has led to improvements in redox properties and transport properties due to high surface to volume ratio. Given such widespread applications and advantageous properties, much effort has been devoted to manufacturing CeO₂ nanomaterials.

Seeds [6-8] and organic templates [9, 10] are typically used to fabricate semiconducting nanowires [11], which are used in nanodevices [6, 11] photonic devices [12], nanosensors [13], solar cells [14], and catalysts [15, 16]. However, residual seeds/templates in the nanowires may act as contaminants [7, 9] and degrade the material properties. Hence, there is a pressing need to develop nanowire growth techniques that do not involve the use of seeds/templates, but instead, allow for easy manipulation of the material properties and dimensions and facilitate large-scale production. Methods based on supercritical fluid technology may be advantageous since they allow for deagglomeration [17] and result in defect-free nanowires in high yields with excellent chemical flexibility and synthetic tenability [18, 19]. Single-crystalline semiconducting nanowires [4, 7, 20-22] can grow via a metal-catalyzed [8], vapor-solid [23], oxide-assisted [6, 10], or self-assembly mechanism [10], and under supercritical fluid [17-19] conditions. Single-

crystalline nanowires in supercritical fluids are generally grown from catalyst seeds or by the oriented attachment of nanoparticles onto specific planes capped with dendrimers or hyperbranched molecules. We previously fabricated unique polycrystalline CeO₂ nanowires in a specific supercritical fluid region without using functional capping agents or catalyst seeds [24]. However, the mechanism underlying the nucleation and growth of such polycrystalline nanowires could not be explained by conventional theories [6, 10, 20-22]. In the present study, we fabricated randomly-oriented polycrystalline Gd-CeO₂ nanowires by a seed-free method using a supercritical CO₂-alcohol co-solvent system and revealed the growth mechanism on the basis of panoramic images obtained in detailed microscopy observations.

Experiments

Synthesis of Gd-CeO₂ nanomaterials

For solvothermal crystallization using supercritical carbon dioxide, 65 mL ethanolic solutions of cerium nitrate Ce(NO₃)₃·6H₂O, including 20 mol% gadolium nitrate Gd(NO₃)₃·6H₂O (concentration: 45–90 mg/mL) were fed to a 350-mL high-pressure vessel. The vessel was charged with 99.99% CO₂ at pressures of up to 7.5 MPa at 35°C and then heated for 15 min at 100–140°C and 5–35 MPa until the ethanolic solution and CO₂ reached the supercritical fluid state. Subsequently, the reaction solution was cooled to ambient temperature and the vessel was slowly depressurized. The as-synthesized nanomaterials were collected by sedimentation, centrifuged at 1,400 × *g* (3,000 rpm) for 10 min in a 15-mL conical centrifuge tube, repeatedly washed with ethanol three times, and dried at 60°C for 12 h.

Characterization

The morphology and microstructure of the nanomaterials were examined by scanning electron microscopy (SEM, Nova NanoSEM, FEI, USA) and transmission electron microscopy (TEM, Tecnai F20, FEI, USA), including high-resolution TEM (HR-TEM) and energy-dispersive X-ray spectrometry (EDS, EDAX). Cryo-TEM was adopted to investigate the actual dissociation state of the aggregates. In this method, the nanomaterial solution exiting the fluid chamber was cooled to cryogenic temperatures by the Vitrobot, and a 100-nm-thick vitrified film indicative of the near *in situ* state during the dissociation reaction was prepared. For observing the dissociation behavior of aggregates in particular, we prepared an *ex-situ* TEM sample by putting C-coated TEM grids on glass positioned at the center region of the chamber.

Results

Morphological evolution

The P-T map shown in Figure 1, which was experimentally classified by SEM and TEM observations, can be divided into three distinct regions-Regions 1, 2, and 3-corresponding to the morphology of the synthesized Gd-CeO₂ nanomaterials, on the basis of temperature and pressure. The SEM and TEM images corresponding to each region (Figures 2a-d) revealed the morphological evolution from nanoclusters (Region 1) to nanowires (Region 3) via the formation of aggregates (about 250 nm diameter) (Region 2). The nanoclusters formed in Region 1 were sphere-like and monodisperse, and their size and shape remained unchanged even at higher pressures and temperatures. The nanoclusters seemed to have a truncated octahedral shape in which the {111} and {220} planes were exposed, although it was difficult to define the shape because of their extremely small size. At a more detailed level, it was observed that the partially faceted planes of the nanoclusters corresponded to {111}, which is the lowest-energy plane in the ceria crystal, including {220}. In Regions 2 and 3, the nanoclusters assembled into spherical aggregates which were transformed into nanowire bundles. The aggregate-to-nanowire transition can be observed in Figure 2c.



Figure 1. P-T map showing the morphological evolution of nanomaterials under supercritical fluid condition. P-T map with a panoramic view of Gd-CeO₂ nanomaterial formation in the absence of functional capping agents or catalyst seeds.



Figure 2. SEM/TEM details on the morphological evolution of nanomaterials with variations of P and T. a) Nanoclusters (Region 1). b) Spheroidal aggregates (Region 2). c) Aggregates with nanowire sprouts (transition region between Regions 2 and 3). d) Nanowires (Region 3).

Formation of nanoclusters and aggregates

Figure 3 shows TEM images of the Gd-CeO₂ nanoclusters and single-crystalline aggregates observed in Regions 1 and 2, including the selected-area diffraction (SAD) pattern and fast Fourier transform (FFT) patterns in the inset. The nanoclusters include a stable Gd-CeO₂ phase with a face-centered cubic fluorite structure (average size: 3 nm), as shown by the dotted circles in Figure 3a, and are highly crystalline, as shown by the sharp ring pattern in the inset. The HRTEM image and FFT pattern in Figure 3b indicate that individual nanoclusters comprise a single-crystalline Gd-CeO₂ phase. TEM and HRTEM images (Figures 3c-f) acquired from specific regions on each aggregate revealed that the aggregates have many nanosized terraces at their surfaces. Although the aggregates comprised singlecrystalline CeO₂, they were loosely packed, as evident



Figure 3. TEM images and diffraction patterns of nanoclusters and single-crystalline structure of aggregates in Regions 1 and 2. a) HRTEM image of Gd-CeO₂ nanoclusters (average diameter: 3 ± 0.5 nm) with high crystallinity, as revealed by the sharp diffraction rings in the inset. b) HRTEM image and FFT pattern showing the fcc crystal structure of individual nanoclusters. c) Aggregates. d–f) Enlarged HRTEM images and FFT patterns. In Region 2, all the TEM observations indicate that the single-crystalline aggregates comprised loosely aligned nanoclusters.

from the broad spots in the FFT patterns (inset of Figures 3d, f). A unique contrast indicative of a 3-nm cluster was observed in the HRTEM images (marked by arrows in Figure 3d), strongly suggesting that the growth of seed-free single-crystalline aggregates in the supercritical fluid is preceded by coalescence of the nanoclusters, which leads to the morphological change from nanoclusters to a loose single-crystalline aggregate.

Dissociation of single-crystalline aggregates

Figure 4 shows the initial aggregate dissociation mechanism in the transition region (Region $2 \rightarrow 3$ in Figure 1). The internal microstructure of the aggregates (Figures 4a, b) revealed a loosely-packed single-crystalline structure comprising 3-nm nanoclusters (marked by arrows). However, the surface of the aggregates (Figure



Figure 4. HRTEM images of the interior and exterior of the dissociated aggregates at the early stage of the transition. a) Aggregates. b) Single crystal lattice image at the interior of the dissociated aggregates, with the clear contrast due to the nanoclusters (see arrows). c, d) HRTEM images of nanoclusters dissociating at the aggregate surface.

4c, d) had dissociated nanoclusters (marked by dotted circles), which served as building blocks for the growth and dissociation of aggregates in the supercritical fluid phase. During dissociation, nanoclusters could not be dissociated to atoms because of the inability of the large CO_2 molecules (3.87 Å in diameter) to diffuse into the cluster lattice (3.12 Å for the largest (111) d-spacing of CeO_2). Thus, HRTEM observations on the aggregate surfaces provided strong evidence for the dissociation-driven morphological transition from aggregates to nanowires between Regions 2 and 3 in the P-T map.

Random growth of polycrystalline nanowires

At high temperatures and pressures, efficient CO₂ permeance into the aggregates led to the formation of nanowire bundles (Figure 5). Nanowires were present inside the aggregates (indicated by white lines in the defocused TEM images (Figures 5a, b) but not outside the aggregates; well-developed nanowire bundles grew on the surface of the aggregates (Figures 6c, d). These observations confirmed that the nanowires begin to grow inside the aggregates and then propagate to the exterior.

The crystallographic images in Figure 6 illustrate how individual nanowire bundles sprout from an aggregate. A randomly-oriented polycrystalline structure comprising



Figure 5. TEM images of nanowires grown from aggregates at the final stage of the transition region. a, b) Gd-CeO₂ nanowires grown from aggregates. Defocused TEM image showing the growth of nanowires from the interior of the aggregates. c, d) Aggregates and well-developed nanowires. Nanowire bundles growing on the surface of the aggregates.

nanoclusters without any specific growth direction could be seen at the aggregate/nanowire sprout junction (Figures 6b, c). However, the region marked by rectangle d in Figure 6a (magnified image shown in Figure 6d) revealed the original single-crystal structure. The complete transformation of all single-crystalline aggregates to polycrystalline nanowire bundles (Figure 7) clearly implies that this unusual growth process is attributed to the dissociation and reconstruction of nanoclusters. The nanowire bundles were well separated into individual nanowires (diameter: ~20 nm; length: ~10 µm) by ultrasonic impact. Despite the shear force applied to the nanowires by the ultrasonic impact, they were mechanically stable. The nanowire bundles (Figures 7b, c) comprised several polycrystalline nanowires, each of which included randomly-aligned nanoclusters, the building blocks for nanowire assembly.

Discussion

Figure 8 schematically summarizes the mechanism underlying the unique nanoparticle-to-polycrystallinenanowire transition. While the formation mechanism for the nanoclusters and single-crystalline aggregates was similar to that previously reported [25], the mechanism


Figure 6. TEM images showing random-alignment-based growth mechanism of polycrystalline Gd-CeO₂ nanowires. a) A representative feature of an aggregate with nanowire sprouts. b) Individual nanocluster reconstructed into a polycrystalline nanowire with non-crystallographic alignment. c) Magnified image of b showing polycrystalline nanoclusters. d) Aggregate surface with single-crystalline structure during nanowire growth.



Figure 7. TEM images of polycrystalline nanowires. a, b) TEM images of nanowire bundles under different levels of magnification. c) A nanowire bundle clearly consisting of several polycrystalline nanowires with randomly-aligned 3-nm nanoclusters.

of polycrystalline nanowire growth beyond Region 2 was different. The nanoclusters formed by dissolution and precipitation of the precursors (Figure 8a) underwent collision and coalescence in the supercritical fluid to yield aggregates with intercluster interfaces. The nanoclusters in the aggregates aligned themselves into a single crystal, so that the interfacial energy was minimized, and eventually formed a loosely packed structure (similar to the mosaic structure, Region 2 in Figure 8b).

In the transition region and Region 3, it is important to consider the balance between the interfacial energy of

the nanoclusters in the aggregates and the permeance of CO₂. The interfacial energy decreases markedly with increases in pressure and temperature because the liquid/solid interfacial tension becomes almost zero under supercritical conditions [17]. Thus, the penetrating force is more dominant than the interfacial energy. Our results indicated that the single-crystalline aggregates dissociated into nanoclusters (Figure 8c) and reassembled to form polycrystalline nanowires inside and outside the aggregates (Figures 8d, e). Although we established the formation of polycrystalline nanowires by random alignment of nanoclusters in the absence of functional capping agents and catalyst seeds leading to selfaligned growth of nanowires, it is difficult to provide a thermodynamic explanation for the growth mechanism. The random orientation of the polycrystalline nanowires implies nonselective attachment of CO₂ molecules on arbitrary crystal planes of the nanoclusters in the metastable transition region. Fusion of the uncapped regions of the nanoclusters is energetically favorable as this reduces the difference in surface energy between the nanoclusters and facilitates the formation of polycrystalline nanowires (Figure 8f). Thus, supercritical CO₂ is confirmed to be the appropriate medium for the growth of



Figure 8. Schematic diagram showing random-alignment-based growth mechanism of polycrystalline Gd-CeO₂ nanowires. a–e) Schematic of the mechanism showing morphological change from nanoclusters to polycrystalline nanowires mediated by supercritical carbon dioxide. In the morphological evolutions, individual nanoclusters act as building blocks for nanomaterials.

polycrystalline nanowires.

Conclusion

In summary, the formation of Gd-CeO₂ nanomaterials such as nanoclusters, aggregates, and nanowires in a supercritical CO₂-ethanol system was analyzed on the basis of a P-T map divided into three regions. The single-crystalline aggregates comprised loosely-aligned nanoclusters for minimizing high interfacial energy between the nanoclusters under low CO₂ permeance. In the transition region with higher CO₂ permeance, the aggregates dissociated into nanoclusters partially covered by CO₂, which in turn attached to one another nonselectively to bring about polycrystalline nanowires. This type of reconstruction is significant in that it opens up a new nanowire fabrication route without the need for seeds or capping agents.

Note

This article and its images are cited from "Polycrystalline Nanowires of Gadolinium-Doped Ceria via Random Alignment Mediated by Supercritical Carbon Dioxide" in *Scientific Reports*, Vol. 3 [1606], pp. 1-5, and "Growth of nanostructured polycrystalline cerium oxide through a solvothermal precipitation using near-supercritical fluids" in *Journal of Nanoscience and Nanotechnolgy*, Vol.10, pp. 130-134.

<u>References</u>

- Chueh WC, Hao Y, Jung WC, Haile SM, *Nat. Mater.* 2011; 12: 1-7.
- [2] Wang YG, Zhang LC, Biddut, Wear 2011; 270: 312-316.
- [3] Moos R, Izu N, Rettig F, Reiß S, Shin W, Matsubara I. Sensors 2011; 11: 3439-3465.
- [4] Zhou K, Wang X, Sun X, Peng Q, Li Y, J. Catalysis 2005; 229: 206-212.

- [5] Georgi, N. et al. Nat. Mater. 2011; 10: 310-315.
- [6] Ji, Z. et al. ACS Nano 2012; 6: 5366-5380.
- [7] Yan RX, Gargas D, Yang PD, *Nat. Photon.* 2009; 3: 569-576.
- [8] Borgstrom MT, Immink G, Ketelaars B, Algra R, Bakkers EPAM *Nat. Nanotech.* 2007; 2: 541-544.
- [9] Yuan ZY, Vantomme A, Du GH, Su BL, *Langmuir* 2005; 21: 1132-1135.
- [10] Wang N, Cai Y, Zhang RQ. *Mater. Sci. Eng.* R. 2008; 60: 1-51.
- [11] Lu W, Lieber CM. Nat. Mater. 2007; 6: 841-850.
- [12] Law M. et al. Science 2004; 305: 1269-1273.
- [13] Hu YJ. et al. Nat. Nanotechnol. 2007; 2: 622-625.
- [14] Tian BZ. et al. Nature 2007; 449: 885-890.
- [15] Huang PX. et al. J. Phys. Chem. B. 2005; 109: 19169-19174.
- [16] Zhou K. et al. J. Catalysis 2005; 229: 206-212.
- [17] Shah PS. et al. J. Phys. Chem. B 2004; 108: 9574-9587.
- [18] Hanrath T, Korgel BA. Adv. Mater. 2003; 15: 437-440.
- [19] Holmes JD. et al. Science 2000; 287: 1471.
- [20] Wen JG. et al. Chem. Phys. Lett. 2003; 372: 717-722.
- [21] Lin KS, Chowdhury S. Int. J. Mol. Sci. 2010; 11: 3226-3251.
- [22] Yang P, Yan R, Fardy M. Nano Lett. 1010; 10: 1529-1536.
- [23] Lee JM, Ahn JP, Kim SW. J. Nanosci. Nanotech. 2010; 10: 130-134.
- [24] Cansell F, Aymonier C, Loppinet-Serani, Science 2003; 7: 331-340.



Clean Energy Research Center

Feature Articles

Facile Growth of Aligned WO₃ Nanorods on FTO Substrate for Enhanced Photoanodic Water Oxidation Activity



Sharanappa Kalanur Clean Energy Research Center shankarask@ gmail.com

Shankara



Yun Jeong Hwang Clean Energy Research Center yjhwang@kist.re.kr



Sang Youn Chae

Clean Energy Research Center 212009@kist.re.kr



Oh Shim Joo Clean Energy Research Center joocat@kist.re.kr

Introduction

Arguably the most challenging contemporary problem is the supply of safe, clean, and renewable energy sources. Worldwide energy consumption has continuously increased, and average power consumption reached 17 terawatts (TW) in 2012. However, more than 70% of that energy was still generated from fossil fuels, while nuclear power accounted for 13% and renewable energy sources the remainder [1]. Burning fossil fuels emits CO₂ and other pollutant gases, leading to global warming, as evidenced by recent climate change. Therefore, alternative energy resources should be carbon-neutral and capable of satisfying growing global energy demand. The recent Fukushima nuclear accident raised an alarm as to the wisdom of increasing dependence on nuclear power, and nuclear power is not capable of meeting increasing world consumption. Of the various renewable energy sources such as wind, hydroelectric, tidal, and biomass, the sun is by far the largest resource. Solar energy is an inexhaustible natural resource, and the amount of solar energy that arrives on the earth in 1 hour $(4.3 \times 10^{20} \text{ J})$ is comparable to annual global energy consumption $(4.74 \times 10^{20} \text{ J in } 2008)$ [1,2]. To utilize solar energy so that it is constantly available despite the daily and seasonal variability of sunlight, we have to develop ways to convert and store it to useful energy. The conversion of solar energy to chemical energy, such as hydrogen, has been suggested in a way that mimics photosynthesis in nature.

In 1972, Honda and Fujishima demonstrated that a semiconductor material can split water and produce hydrogen with TiO₂, which absorbs photons to generate excited electrons and holes to provide energy to break water molecules [3]. Various semiconductors have been studied in solar water splitting, especially ones containing metal oxides such as TiO₂, WO₃, and Fe₂O₃, because they remain stable in oxygen-evolving environments, they are easy to produce, and have reasonably high solar energy conversion efficiency. WO₃ has shown promise as a water splitting material due to its ability to capture approximately 12% of the solar spectrum (visible spectrum up to 500 nm) with an indirect band gap of 2.5-2.7 eV [4]. Moreover, the electrical properties of WO₃, such as inherently good electron mobility and decent hole diffusion length (~150 nm), can contribute to high quantum efficiency by increasing charge separation efficiency [5]. Although WO₃ does not possess a high enough conduction band minimum for spontaneous H⁺ reduction, it can be coupled with another semiconductor suited for solar H₂ production or a tandem device to achieve overall water splitting [6, 7].

In addition to material choice, the morphology of the semiconductor is critical for conversion efficiency since charges travel along the materials [6]. Much effort has focused on the development of nanostructure-based WO₃, such as nanorods, nanotubes, nanobelts, and nanoplatelets [8]. Among these options, one dimensional (1-D) semiconductor structures [9] offer the advantages of light absorption and direct electrical pathways for photogenerated charges to move along them while providing efficient charge separation across the diameter [10]. 1D nanostructure-based WO₃ photoanodes, such as nanorods, have a high interfacial contact area compared with bulk WO₃, a structure that can increase the density of redox-active sites and enhanced light harvesting and charge separation efficiency as well [8]. Therefore, developing a simple and one-step synthesis of 1-D WO₃ nanostructures on conducting substrates is a promising approach which can do efficient water photoelectrolysis [11].

The morphology, crystal phase, and catalytic activities of WO₃ are greatly influenced by the method used for its synthesis, and single-step synthesis of a 1D WO₃ nanostructure is still a challenging process for solar water splitting. Here, we demonstrate a facile hydrothermal synthesis of WO₃ nanorods directly aligned on FTO by using a stable aqueous precursor solution without the assistance of a seed layer or structure-directing agent. Depending on the annealing temperature, as-prepared orthorhombic hydrated WO₃ nanorods can be converted to hexagonal or monoclinic. We observed superior water oxidation performance with monoclinic WO₃ nanorods.

Synthetic Strategy of 1D WO₃ Nanorods

We designed a series of hydrothermal synthetic experiments with peroxoploytungstic acid (PTA) precursor solution to achieve 1D WO₃ nanorods on a transparent conducting oxide (TCO) substrate in a one-step process. Only a few reports have been published on the growth of WO₃ nanorods on FTO substrate, most of which used a seed layer, usually prepared by spin coating and annealed before growing the nanorods. Unfortunately, those syntheses resulted in a hexagonal WO₃ phase which is less active than monoclinic for water oxidation [10, 12]. Therefore, developing a simple solution-processed synthesis of nanostructured monoclinic WO₃ is required for wide photoanodic application.

In a typical synthesis, 1 g of ammonium paratungstate $\{(NH_4)_{10}(H_2W_{12}O_{42})\cdot 4H_2O\}$ was dissolved in 95 ml deionized (DI) water, and 1-10 ml of concentrated HCI was added (0.125-1.25 M). The solution initially turned to transparent light yellow, and then yellow gelatinous precipitate formed. 2~4 ml of H₂O₂ was added and stirred vigorously for 1 h to dissolve tungstic acid and form a stable and transparent solution of PTA. The prepared PTA solution was transferred to a Teflon-liner, and a clean FTO substrate was placed at an angle against the wall with the conducting side facing down. The FTO substrates were taken out and rinsed with DI water, and they were post-annealed for 1hr in air.

The morphologies, adhesion to the FTO substrate, and crystal structures of the WO₃ films varied by postannealing at different temperatures (300~700°C). WO₃ nanorod arrays were prepared at 160°C for 4h to observe the influence of the post-annealing. (Figure 1) The morphology remained unchanged when the samples were annealed up to 300°C, as observed in scanning tunnelling microscope (SEM) images. The morphology began to change starting at 400°C, and the rods first showed signs of splitting into twin rod-like morphology at 500°C. As post-annealing temperature increased, nanorods combined to make thicker flakes, and rodlike morphology disappeared eventually producing thick



Figure 1. SEM images of WO_3 nanorod arrays on FTO prepared at 160°C for 4 h before (a), and after annealing at: (b) 300; (c) 400; (d) 500; (e) 600; and (f) 700°C.

blocks reflecting mesoporous morphology at an annealing temperature of 700°C. In addition, the WO₃ films annealed at 600~700°C showed poor adhesion to the FTO surface.

The crystal phases of the WO₃ nanorods evolved from the WO₃·0.33H₂O orthorhombic phase to an anhydrous WO₃ hexagonal, and finally converted to stable monoclinic WO₃, according to X-ray diffraction patterns (Figure 2). The WO₃·0.33H₂O orthorhombic phase was obtained and maintained up to 300°C. However, when the sample was post-annealed at 400°C, the crystal phase was converted to an anhydrous hexagonal. The absence of water molecules in the anhydrous hexagonal phase of WO₃ was confirmed using FTIR. Further increases in annealing temperature > 500°C produced stable monoclinic WO₃. This phase transformation is an example of reconstructive transformation in which the mother phase framework is



Figure 2. XRD patterns of WO_3 nanorod arrays on FTO. (a) As-prepared (orthorhombic). Annealed at: (b) 300°C (orthorhombic hydrate); (c) 400°C (hexagonal); (d) 500°C (monoclinic); (e) 600°C (monoclinic); and (f) 700°C (monoclinic).



Figure 3. (a) TEM of WO₃ prepared at 160°C for 4 h (annealed at 500°C); insets show the electron diffraction pattern (top left) and a high resolution TEM image (bottom right). (b) UV-Vis absorption spectra of asprepared WO₃ $0.33H_2O$, and post annealed hexagonal and monoclinic WO₃.

broken, chemical composition changes, and new bonds are formed leading to a new stable structure [13].

The monoclinic WO₃ nanorods resulting from postannealing at 500°C had a single-crystalline nature and grew along a (002) direction. As seen in transmission electron microscopy (TEM) imaging, the measured lattice spacing was 0.38 nm, d-spacing of (002) planes of the monoclinic WO₃. The SAED pattern of a single nanorod showed regular diffraction spots of single crystalline (Figure



Figure 4. SEM images of WO_3 nanorods grown on FTO at 160°C for: (a) 1; (b) 2; (c) 3; and (d) 4 h. Insets in the figure represent corresponding cross-sectional images.

3a). Also, the optical properties of annealed samples improved to absorb up to almost 460 nm of visible light (Figure 3b). As-prepared WO₃ nanorod arrays absorbed light only in a UV region with wavelengths smaller than 370 nm. Meanwhile, monoclinic WO₃ absorbed more visible light compared to hexagonal WO₃. The measured E_g for WO₃ before and after annealing at 500°C was 3.06 and 2.61 eV, respectively.

Close observation of the WO₃ film in the early stage of growth suggests that a thin layer of WO₃ nanoparticles which formed in the first hour acts as a seed layer initiating nucleation and growth sites for WO₃ nanorods during later growth (Figure 4). Hydrothermal growth was carried out for 1, 2, 3, and 4 h at 160°C. Nanoparticles were first deposited on the FTO surface, then preferential growth occurred along the edges of the planes to form nanorods. The nanoparticle under-layer indicates that the nucleation of nanorods was relatively slow. The nanorod film achieved a maximum thickness of ~1 µm by 4 h of reaction time, but started to peel off from the FTO substrate when reaction time passed 4 h. This was due to dissolution of WO3 at the WO3/FTO interface. This in situ quasi seed layer likely assists the uniform distribution of the nanorods over the entire FTO surface, leaving no empty surface regions, unlike earlier studies reporting uneven or sparse distribution without a seed layer [14].

The resulting crystalline phase and morphology of hydrated tungsten oxide is dependent on nature and

the stability of the precursor solution. In our procedure, the H_2WO_4 precipitate was formed after adding HCl to ammonium paratungstate solution. In this stage, hydrated tungsten oxides (WO₃.nH₂O) began to form via condensation of H_2WO_4 .[15] WO₃.nH₂O turned out not to be suitable for the coating on FTO substrate, suggesting that coatings require stable colloidal precursor solutions by chelating ligands such as hydrogen peroxide [15]. The formed H_2WO_4 precipitate was dissolved by adding hydrogen peroxide-forming PTA and remained stable for 2~3 days [16]. This stable PTA solution was treated hydrothermally in the presence of FTO to obtain direct growth of WO₃ nanorods on FTO substrates.

Catalytic Activity in Solar Water Splitting

The photoelectrochemical (PEC) properties of the WO₃ nanorod arrays strongly depend on their postannealing temperature due to their phase transformation and morphology changes. The PEC properties of WO₃ nanorod arrays were investigated under simulated sunlight AM 1.5G at 100 mWcm⁻² in a 0.5 M Na₂SO₄ electrolyte. As-prepared orthorhombic WO₃ \cdot 0.33H₂O nanorod arrays showed negligible photocurrent values (<10 µA cm⁻²), and no change was oberved with a sample annealed at 300°C. Among all samples, those annealed at 500°C demonstrated the highest photocurrent, and it is worth noting that the photocurrent value (2.26 mA cm⁻² at 1.23 V versus RHE) is higher than other 1D WO₃ nanostructures reported by other groups.

The low photocurrent values observed below a 500°C annealing temperature is attributed to unfavourable phases. One of the reasons that monoclinic WO₃ shows a much higher photocurrent is its capability of visible light absorption. The wider bandgap of orthorhombic $WO_3 \cdot 0.33H_2O$ and hexagonal WO_3 limit their absorption in the visible region, resulting in low photocurrents. As previously reported [10, 12], nanorod- and nanoneedle-



Figure 5. (a) Plot of photocurrent density (at 1.23-V versus RHE) versus as-annealed WO₃ nanorod arrays at different temperatures. (b) I-V scans under chopped light at a scan rate of 10 mV/s. (c) IPCE under potential bias of 0.8-V versus Ag/AgCI.

like arrays on FTO are comprised of either hexagonal or orthorhombic phases, and hence show comparatively less photocurrent (less than 0.5 mAcm⁻²). In our research, photocurrent values decreased at higher post annealing temperatures (> 500°C), even with monoclinic phases. This might be due to changes of morphology, the reduced surface area of WO₃ nanorod arrays, or poor adhesion on the FTO substrate. The dramatic increase of photocurrent with monoclinic WO₃ indicates that the crystal phase of the WO₃ is important for PEC water oxidation activity. This also shows the advantage of the present hydrothermal method in providing the monoclinic crystal phase for photoanode application with high efficiency.

To further quantify the PEC performance of WO₃ nanorod arrays, the incident photon-to-current conversion efficiency (IPCE) was measured at a 0.8 V bias versus Ag/ AgCl using the following expression:

$$IPCE = \frac{1240 I_{ph}}{\lambda I_{power}}$$

in which, I_{ph} is a photocurrent density, λ is a wavelength of the incident light, and I_{power} is the measured power density of the incident irradiance. The onset of IPCE was observed at 460~470 nm, indicating 2.6~2.7 eV of bandgap, consistent with UV-Vis absorption

measurement. Monoclinic WO $_3$ nanorod arrays exhibit IPCE higher than 35% at 400 nm, and IPCE increases up to 90% at 350 nm.

Summary

Our group developed a simple and fast approach for the growth of oriented WO₃ nanorods on FTO. WO₃·0.33H₂O nanorod arrays were obtained hydrothermally using a stable aqueous solution of PTA without a seed layer or structure-directing agents. Orthorhombic WO₃·0.33H₂O converted to photocatalytically active monoclinic WO3 nanorod arrays through post-annealing at 500°C. Post-annealing decreased the band gap down to 2.6 eV and improved the photocurrent in water oxidation by more than two orders of magnitude. This significant increase of photocurrent is attributed to the direct pathway provided by the vertically aligned nanorod arrays on the conducting substrate coupled with a favorable crystal phase. The WO₃ nanorod arrays prepared using our proposed method can also be applied in the fields of gas sensing, electrochromic materials, and lithium ion batteries since the crystal phases of WO₃ nanorod arrays can be controlled by the post-annealing conditions, depending on the desired application.

Note

This article and images are cited from "Facile Growth of Aligned WO₃ Nanorods on FTO Substrate for Enhanced Photoanodic Water Oxidation Activity" in *J. Mater. Chem. A.* 2013; 1: 3479- 3488.

- [2] Lewis NS, Nocera DG. Proc. Natl. Acad. Sci. USA 2011; 103: 15729.
- [3] Fujishima A, Honda K. Nature 1972; 238; 37.
- [4] Gratian RB, Kazuhiroand S, Hironori A, J. Photochem. Photobiol. A: Chemistry 1999; 122: 175.
- [5] Van de Krol R, Liangand YQ, Schoonman J. J. Mater. Chem. 2008; 18: 2311.
- [6] Nozik AJ. Annu. Rev. Phys. Chem. 1978; 29: 189.
- [7] Jasonand AS, Choi KS. Chem. Mater. 2011; 2: 1105.
- [8] Xien L, Fengying W, Qing W. Phys. Chem. Chem. Phys. 2012; 14; 7894.
- [9] Gudiksen MS, Lauhon LJ, Wang J, Smithand DC, Lieber CM. *Nature* 2002; 415: 617.
- [10] Jinzhan S, Xinjian F, Jennifer DS, Liejin G, Craig AG. Nano Lett. 2011; 11: 203.
- [11] Santato C, Ulmann M, Augustynski J. *Adv. Mater.* 2001; 13: 511.
- [12] Jinzhan S, Liejin G, Ningzhong B, Craig AG. *Nano Lett*, 2011; 11: 1928.
- [13] Figlarz M. Mater. Sci. Forum 1994; 152-153: 55.
- [14] Jiao Y, Wenzhang L, Jie L, Dibo S, Qiyuan C. J. Mater. Chem. 2012; 22: 17744.
- [15] Livage J, Guzman G. Solid Stute Ionics 1996; 84: 205.
- [16] Pecquenard B, Lecacheux H, Livageand J, Julien C. J. Solid State Chem. 1998; 135: 159.



Photos of laboratory

<u>References</u>

[1] http://www.worldwatch.org/files/pdf/SOW09_chap3.pdf

Feature Articles

Supported Core@Shell Electrocatalysts for Fuel Cells: Close Encounter with Reality



Sung Jong Yoo Fuel Cell Research Center ysj@kist.re.kr

Introduction

Over the last few decades, advanced catalysts for low-temperature fuel cells have been extensively studied using theoretical design and experimental validation [1]. Nonetheless, there are still critical hurdles to overcome for electrocatalysis in proton exchange membrane fuel cells (PEMFCs), including: 1) overcoming the sluggish oxygen reduction reaction (ORR) kinetics with a minimum amount of Pt; 2) developing novel selective catalysts for the hydrogen oxidation reaction (HOR) for the shutdown/startup stability of the stack; and 3) enhancing the physical/chemical durability of the catalysts [2]. Despite the synthetic challenges, several notable studies have found that non-noble metal-alloys had significant activity and durability.

The advantage of core@shell nanostructures is high Pt utilization by interposition of the non-noble core, which further alters the electronic structure of Pt and stabilizes it by enhancing the interfacial bonding between the core and shell [3]. To date, reports on core@shell ORR catalysts can be classified by their synthetic approaches, which include the seed mediated sequential meth [4], the galvanic-replacement reaction [5], and structural rearrangement (de-alloying or segregation) [6]. However, studies of well-defined multi-metallic core@shells smaller than 10 nm are rare, particularly for the supported form that exhibits significant activity and durability and can potentially be used in real cells.

To achieve these performance goals at KIST, we computationally

screened potential core@shell combinations and devised a facile synthesis route for the proposed structures. We also characterized performance and durability at the single cell level. To determine the best core@shell combinations for ORR, we performed density functional theory (DFT) calculations using two descriptors. The first descriptor is the oxygen adsorption energy (OAE) on the Pt shell to measure the reactivity [2,7]. The other descriptor is the vacancy formation energy (VFE) of Pt in the Pt shell to measure the stability proposed in this study.

The computational screening of Ptcoated suitable core materials for high activity and durability

DFT calculations have become an important tool for understanding the properties of metal surfaces and their reactivity, including the effects of structure, alloying, and adsorption. This theoretical approach has begun to suggest the design of surfaces with specific catalytic properties of interest. In fuel cells, electrochemical reactions such as HOR and ORR have been explained by this theoretical approach. In the case of the ORR, it is truly needed to reduce a large overpotential even on Pt. Nørskov et al. have already shown the origin of the ORR [7]. Furthermore, they have suggested promising candidates with enhanced ORR rates and higher stability [1,2,7].

The computational screening of activity and durability using DFT calculations was performed on the overlayersubstrate system, which mimics core@shells, to avoid the complexity introduced from multiple atoms in the particles. The thickness of the Pt overlayer was fixed at 1 monolayer (ML) because thicker Pt layers exhibit bulk properties. The OAEs and VFEs of the pure elemental cores (Pd, Ir, Ni) are presented as open squares in Figure 1. The Δ OAE on the vertical scale is the difference between bulk Pt and core@shell structured model catalysts. The optimum Δ OAE value (the horizontal dashed line) was

referred from the work by Nørskov et al. [8]. The VFEs on the vertical scale are the averages which represent mixed property for the two symetrically distinct sites of "Up" and "Down". The OAEs of some metal@Pt were predicted to have better reactivity or stability than pure Pt. However, the reactivity was still far from the optimum value (the horizontal dashed line), which prompted us to extend the combinations to alloy cores (black circles). Though Pt₃Ni₁@Pt was predicted to be the most reactive, it was excluded due to its poor stability (i.e., its VFE was the second lowest), which would result in poor long-term operational performance. Pt₃Ni₁@Pt and Pd₃Fe₁@Pt were predicted to have weaker OAE and corresponded to the experimental results [2]. The better activity of Pt₃Ni₁@Pt reported from experimentation could be explained by the fact that the OAE of Pt₃Ni₁@Pt is closer to the optimum value than that of Pt. However, the Pd₃Fe₁@Pt, calculation showed a slight disagreement with the experiment; there was stronger OAE and better activity than that determined through the calculation. According to the experiment, the activity of Pd₃Fe₁@Pt was several times higher than that of Pt. This discrepancy might have originated from the uncertainty in the optimum value or computational accuracy. However, based on the relatively good stability and reactivity of Pd@Pt, we focused our attention on Pd-



Figure 1. The calculation was performed on the overlayer-substrate system with a fixed thickness of the Pt overlayer (1 monolayer). The optimum value of the reactivity is indicated in the horizontal dashed line.

based 3*d* transition metal alloy cores to minimize the cost/ activity ratio. As shown in Figure 1, Pd₃Cu₁@Pt has the most suitable OAE and VFE, implying significant activity and durability. In addition, we devised a facile and robust route to synthesize this core@shell [8].

Facile synthesis of carbonsupported core@shell electrocatalysts

Our novel two-step synthetic method is illustrated in Figure 2. For the production of the C-supported form, the shell generation is typically achieved by reduction of the precursor on the prepared cores/C using a mild reducing reagent with the proper surfactants [9] which can result in the simultaneous reduction of the precursor on the cores and carbon. This problem can be avoided by adding carbon in the final stage. However, this method still suffers from a weak metal-support interaction, an inability to precisely control the metal loading, and the need for additional treatment. Thus, selective reduction of the Pt shell on the core would be highly beneficial.

The preparation of a monodispersed core/C is a prerequisite for the uniform reduction of the shell because the reduction potential of the shell precursor varies with the core size. Although strong binding surfactants (i.e., amine reagents containing long alkyl chains and phosphine ligands) have traditionally been employed to synthesize the monodispersed metal nanoparticles, their residues may lead to deterioration of the activities of the nanoparticles. Thus, understanding the surface chemistry of colloidal nanoparticles is crucial for developing an efficient stabilizing system. We postulated that benzyl ether could be utilized as a solvent and weak coordinating surfactant for the synthesis of Pd-based cores. Its short alkyl chain and weak coordinating property compared to nitrogen or phosphorous eliminates the need for its removal by unwanted heat treatment. Based on these characteristics, the synthesis of Pd/C and Pd₃Cu₁/C was



Figure 2. Benzyl ether was utilized as a solvent as well as a weak coordinating surfactant for the synthesis of Pd-based cores without using surfactants. The Hantzsch ester is capable of reducing the shell precursors selectively on the core surface.

accomplished using benzyl ether and a mild borane *tert*butylamine complex as a reducing reagent. A TEM image showed that Pd/C and PdCu/C are mono-dispersed with particle sizes of 3.5 nm (Figures 3a and 3b). The lattice distances of Pd (2.41 Å) and Pd₃Cu₁ (2.12 Å) were also analyzed by HRTEM.

R. R. Adzic (Brookhaven Laboratory) showed examples of Pd@Pt or PdAu@Pt electrocatalysts using galvanic displacement with sacrificing Cu [10]. Though the displacement enabled the preferential deposition of Pt on the cores, the retention of Cu as an alloy core material may be difficult with their method. We have tried to find mild redox systems that are capable of reducing the precursor on the metal surface but not strong enough to reduce the precursor on the carbon. A naturally occurring hydrogenation redox process involving a combination of enzymes and hydride reduction cofactors is a good example. The Hantzsch ester, which was developed to perform the highly enantioselective transfer hydrogenation in organometallic chemistry, is a suitable reducing agent for the preferential deposition of the shell on the cores. To the best of our knowledge, this is the first study that uses the Hantzsch ester as a reducing agent for the synthesis of inorganic nanoparticles.

Core@shell characterizations

The TEM micrographs (Figures 3c and 3d) show well-dispersed spherical $Pd_3Cu_1@Pt/C$ and $Pd@Pd_4Ir_{6}/C$ particles that are larger than the cores (5 nm for

Pd₃Cu₁@Pt and 8 nm for Pd@Pd₄Ir₆). It should be noted that isolated Pt or Pd₄Ir₆ nanoparticles were not found on the carbon, implying the exclusive deposition of the shell material on the cores. Although Cu loss due to displacement with the Pt precursor occurred in the absence of the Hantzsch ester, no such loss occurred in its presence. The unique function of the Hantzsch ester was further verified by the failure to form a selective shell in the presence of ascorbic acid or sodium borohydride, which are common reducing agents for core@shell synthesis. Figures 3e and 3f depict the line-profile analysis using aberration-corrected STEM/energy dispersive spectrometry (EDS), revealing the distribution of the components in Pd@Pd₄Ir₆/C and Pd₃Cu₁@Pt/C. The lower Ir and Pt intensities and higher Pd and Cu intensities at the center clearly verify the formation of core@shell structures. The shell thicknesses and composition/ amounts of the core were easily controlled (note that the



Figure 3. (a) Pd/C NPs synthesized with borane *tert*-butylamine complex in benzyl ether. (b) Pd₃Cu₁/C NPs prepared in the same manner. (c) Pd@Pd₄lr₆/C and (d) Pd₃Cu₁@Pt/C core@shell electrocatalysts. HAADF-STEM images and their line profiles: (e) Pd@Pd₄lr₆ and (f) Pd₃Cu₁@Pt.

shell thickness of Pd₃Cu₁@Pt was approximately 1 nm; 2–3 monolayers of Pt).

Electrochemical performance: activity and stability

Figure 4a summarizes the ORR kinetics of the different alloys, core@shells, and JM 40 wt% Pt/C catalysts. The JM Pt/C catalyst has a particle size of 2-5 nm with a most probable size of 3 nm. The half-wave potential (HWP) exhibits the following trend: $Pd_3Cu_1@Pt/C > JM$ $Pt/C > Pd_3Cu_1/C > Pd/C > Pd@Pd_4Ir_6/C$. The kinetic current density of 0.9 V vs. RHE for Pd₃Cu₁@Pt/C was also much higher than that of the JM Pt/C. The Pt-massnormalized current density of Pd₃Cu₁@Pt with a 0.7 eq Pt shell content was 2.6 times higher than that of JM Pt/ C. In addition, the current density of the smaller Pt shell content (0.7 eq) was 1.7 times higher than that of the 1.5 eq Pt shell, which corresponds to a shell that is 1.8 times thicker, and the smaller Pt shell had a more positive HWP (by 15 mV); these results indicate the dependency of Pd₃Cu₁@Pt activity on the thickness of the Pt shell. For example, Pd₃Cu₁@Pt with a 45% Pd₃Cu₁/C core exhibited a 40-mV positive shift in the HWP compared to that of the 30% core.

Another interesting result is that while the Pt shell coating on Pd₃Cu₁ greatly enhanced the ORR activity, the Pd₄Ir₆ shell coating on Pd caused it to deteriorate, leading to an inactive ORR. However, the Pd@Pd₄Ir₆/C exhibited high activity for HOR, as shown in Figure 4b. The comparison of the mass-transfer-corrected Tafel plots and exchange current densities clearly showed that Pd@Pd₄Ir₆/C displayed a higher activity for HOR than JM Pt/C. The significant possibilities suggested by this result in combination with the results in Figure 4a can be summarized as follows: i) a consolidated synthetic method for making highly active Pt-free anode catalyst, and iii) a HOR-selective catalyst that prevents carbon corrosion during

the startup/shutdown processes [11]. Markovic et al. have suggested a chemical modification of the anode catalyst that would suppress the ORR [12]. However, our research suggests that the selectivity of the catalysts for ORR and HOR can easily be controlled by tuning the core@shell metals and fine-tuning the core size and shell thickness with our simple and novel synthetic method.

The single cell performances for both $Pd_3Cu_1@Pt/C$ and JM Pt/C as cathode catalysts (0.3 mg_{metal}/cm^2) are shown in Figure 4c under the following operating conditions: 10 cm² active area, 70°C, and atmospheric pressure. The MEA with $Pd_3Cu_1@Pt/C$ catalysts exhibited the best performance in the range of > 0.5 V, particularly in the high voltage area (0.9–0.6 V), where the currents were 1.6 and 2.0 times higher than those of Pt/C MEA at 0.7 V and 0.8 V, respectively. The mass activities (inset) were also compared in terms of Pt mass and Pt + Pd mass and showed a higher mass activity for $Pd_3Cu_1@Pt/C$ even



Figure 4. (a) ORR polarization curves for the Pd/C, Pd_3Cu_1/C , $Pd_3Cu_1@$ Pt/C, $Pd@Pd_4|r_6/C$, and Pt/C (room temperature, O_2 -saturated 0.1 M HClO₄ solution, 10 mV/s, 1600 rpm). (b) Tafel plot and the exchange current density (inset) for hydrogen oxidation reaction on Pt/C, Pd@lr/C, and Pd@Pdlr/C (H₂-saturated 0.5 M H₂SO₄ solution, 276 K, 20 mV/ s, 1000 to 3000 rpm). (c) The single cell polarization curves for both $Pd_3Cu_1@Pt/C$ and JM Pt/C as cathode catalysts (0.3 mg_{metal}/cm², 10 cm² active area, 70°C, atmospheric pressure). Inset: mass activities in terms of Pt mass and Pt + Pd mass. (d) The stability test of the Pd_3Cu_1@ Pt/C single cell (400 mA/cm², air-interruption method). Inset: the accelerated stability tests with potential cycling (OCP to 0.35 V, 10 mA/ cm²-sec, 70°C).

when based on the Pt + Pd mass. The stability test of the $Pd_3Cu_1@Pt/C$ single cell was performed at 400 mA/cm² (Figure 4d) and resulted in an approximately 8% (0.73 V to 0.67 V) decay rate, even after 4,000 h of operation. The inset of Figure 4d provides the results of the accelerated stability tests with potential cycling between open circuit voltage to 0.35 V and a scan rate of 10 mA/cm²·sec at 70°C. After 3,000 min of potential cycling, the Pt/C cell performance decreased to 65% and 45% of its initial performance at 0.6 V and 0.7 V, respectively. However, the degradation of the Pd₃Cu₁@Pt/C single cell was half of the Pt/C cell degradation, indicating the superior stability of $Pd_3Cu_1@Pt/C$.

Analysis and simulation of electronic structure in carbonsupported core@shell catalysts

The OAEs with various Pt thicknesses for Pd₃Cu₁@ Pt/C are presented in Figure 5b. The OAE for Pt 2 ML showed the closest value to the optimum and corresponded well to experimental results for 2 ML of Pt covering the Pd₃Cu core. However, from the viewpoint of durabily, Pt 1 ML showed the largest VFE, as shown in Figure 5a. This enhanced durability for Pt 1 ML was investigated using a Bader analysis of Pd₃Cu₁@Pt/C with Pt 1 ML compared to pure Pt layers. For pure Pt, there was a slight charge transfer from the subsurface to the surface by Friedel oscillations. The total charge at the surface Pt was estimated to be 10.045. The Bader charges of Pd and Cu in the bulk Pd₃Cu₁ phase were 10.10 and 10.70, respectively, implying a transfer of 0.1 electron from Cu to Pd. There was a significant charge transfer from Pd₃Cu to Pt (0.112 electron from Pd, and 0.06 electron from Cu). Because Pd had already gained electrons from Cu, it had a sufficient amount of potential to transfer electrons to Pt. In addition, Cu was expected to exhibit a small amount of charge transfer [13]. By increasing the Pt thickness to more than 2 ML, the charge



Figure 5. (a) Vacancy formation energy. (b) Oxygen adsorption energy with various thicknesses of Pt over-layer.

at the surface Pt atom approached the value of pure Pt (10.045), which predicted the same VFEs for Pt layers thicker than 2 ML.

In addition, the electron donation from the Pd_3Cu_1 sub-layer to Pt was verified by the X-ray absorption nearedge structure (XANES) on the Pt L_{III} edges of $Pd_3Cu_1@Pt/$ C by varying the Pt thickness (Figure 6a). The Pt *d*-band vacancy from the Pt L_{III} white lines verified the electron transfer between the core and shell. It is important to note that the intensity of the white line, the magnitude of which is a direct measure of *d*-band vacancies, decreases as the Pt shell thickness decreases. Based on studies by other researchers [14] and our previous study [15], these XANES data clearly indicate electron donation from the



Figure 6. (a) Pt L_{III}-edge XANES spectra and variation in unfilled *d*-states for samples having different shell thicknesses. Inset: enlargement of Pt L_{III}-edge XANES white line. (b) The distribution of the transferred charge from Pd₃Cu₁ in comparison with other Pt₃M (M=Ni, Co, Fe). (c) Charge accumulation (yellow) along the Pt and Pd₃Cu₁ interface and charge dissipation (cyan) in the Pd₃Cu₁ substrate.

Pd₃Cu₁ to Pt through strong metal-metal interactions [16].

For predicting stability, we plotted the difference in the charge density of $Pd_3Cu_1@Pt$ from its pure states of Pd_3Cu_1 and Pt (Figure 6b). Charge accumulation (yellow) along the Pt and Pd_3Cu_1 interface was obtained, while charge dissipation was found in the Pd_3Cu_1 substrate, implying charge transfer from the substrate to the Pt overlayer (Figure 6c). The distribution of the transferred charge from Pd_3Cu_1 differs from other Pt_3M systems. Compared to the triangular-shaped charge transfer for other Pt_3M systems, the transferred charge in the Pd_3Cu_1 system forms a hexagonal ring consisting of three Pd and three Pt atoms (Figure 6b), indicating that the active material for the charge transfer and interfacial bonding of $Pd_3Cu_1@Pt$ was Pd.

Conclusion

We have designed and developed highly active

and stable Pd₃Cu₁@Pt/C (ORR) and Pd@PdIr (HOR) electrocatalysts. The new core@shell combinations were designed using DFT calculations guided by the OAE and VFE. The surfactant-free synthesis of core/ C and the subsequent exclusive shell formations on the core using the Hantzsch ester as a reducing agent were demonstrated for the first time. Superior characteristics, such as high activity, HOR selectivity, and 4,000 h of long-term durability, were achieved with the novel core@ shell/C catalysts. The DFT calculations and XANES analysis strongly support the origin of the enhanced characteristics. The descriptors used in this study can be extended to screen other core@shell combinations.

Note

This article and images are cited from "Supported Core@Shell Electrocatalysts for Fuel Cells: Close Encounter with Reality" in *Sci. Rep.* 3, 1309.

References

- [1] Gasteiger HA, Markovic NM. *Science*. 2008; 324: 48.
- [2] Stamenkovic VR, Fowler B, Mun BS, Wang G, Ross PN, Lucas CA, Markovic NM. Science. 2007; 315: 493.
- [3] Yang H. Angew Chem Int Ed. 2011; 50: 2674.
- [4] Mazumder V, Chi M, More KL, Sun S. J Am Chem Soc. 2010; 132: 7848.
- [5] Gong K, Su D, Adzic RR. J Am Chem Soc. 2010; 132: 14364.
- [6] Strasser P, Koh S, Anniyev T, Greeley J, More K, Yu C, Liu Z, Kaya S, Nordlund D, Ogasawara H, Toney MF, Nilsson A. *Nat Chem.* 2010; 2: 454.
- [7] Nørskov JK, Rossmeisl J, Logadottir A, Lindqvist L, Kitchin JR, Bligaard T, Jónsson H. J Phys Chem B. 2004; 108: 17886.

- [8] Stephens IEL, Bondarenko AS, Perez-Alonso FJ, Calle-Vallejo F, Bech L, Johansson TP, Jepsen AK, Frydendal R, Knudsen BP, Rossmeisl J, Chorkendorff I. J Am Chem Soc 2011; 133: 5485.
- Habas SE, Lee H, Radmilovic V, Somorjai GA, Yang P. Nat Mater. 2007; 6: 692.
- [10] Sasaki K, Naohara H, Cai Y, Choi YM, Liu P, Vukmirovic MB, Wang JX, Adzic RR. Angew Chem Int Ed. 2010; 49: 8602.
- [11] Borup, R. et al. Chem Rev. 2007; 107: 3904.
- [12] Genorio B, Subbaraman R, Strmcnik D, Tripkovic D, Stamenkovic VR, Markovic NM. Angew Chem Int Ed. 2011; 50: 5468.
- [13] Tang W, Zhang L, Henkelman G. *J Phys Chem Lett.* 2011;2: 1328.
- [14] Ho VTT, Pan CJ, Rick J, Su WN, Hwang BJ. J Am Chem Soc. 2011; 133: 11716.
- [15] Yoo SJ, Hwang SJ, Lee JG, Lee SC, Lim TH, Sung YE, Wieckowski A, Kim SK. *Energy Environ Sci.* 2012; 5: 7521.
- [16] Rodriguez JA, Goodman DW. Science. 1992; 257: 897.

Heparin-conjugated pluronic nanogels as multi-drug nanocarriers for combination chemotherapy

Molecular Pharmaceutics 2012.12.28; 10(2): 685-693.

Yoon Ki Joung, Ji Young Jang, Jong Hoon Choi, Ki Dong Park ykjoung@kist.re.kr

Combination chemotherapy using more than two therapeutic agents with different modes of action is a promising strategy that can be used to enhance the therapeutic efficacy of cancer treatment, even though it is a complicated treatment modality. The current study describes a novel platform of a multi-drug nanocarrier that enables paclitaxel and DNase to be delivered to the intracellular sites of a cancer cell with each agent acting on its own as well as in combination. The multidrug nanocarrier was a nano-sized gel-like structure, called a nanogel, that was composed of heparin and Pluronic and that possessed both hydrogel-like and micellar properties. Our goal was to investigate whether the nanocarriers would show a synergistic cytotoxicity in combination with these agents which have different modes of action, and these findings could suggest novel multi-drug nanocarriers and efficient strategies for combination chemotherapy. The aim of this study was to investigate how a novel multi-drug nanocarrier could be effective for combination chemotherapy in vitro, and,



more specifically, whether combined agents with different modes of action and physico-chemical properties would show synergistic cytotoxicity with the use of this nanocarrier. A heparin-Pluronic (Hep-Pr) nanogel encapsulating both paclitaxel and DNase was shown to be efficient for intracellular delivery with respect to size, encapsulation efficiency, and intracellular uptake/fates. As a result of these properties, a Hep-Pr nanogel combined with paclitaxel and DNase exhibited a dose-dependent synergistic cytotoxicity compared to single drug and freedrug treatments, where combination indices were 0.93 and 0.45 at higher concentrations (250 and 500 μ g/mL). Therefore, Hep-Pr nanogels have the potential to deliver multi-therapeutic agents with different characteristics and thereby enhance the therapeutic efficacy of combination cancer chemotherapy.

Hair steroid profiling reveals racial differences in male pattern baldness between Korean and Caucasian populations

Journal of Investigative Dermatology 2013; 133: 822-824.

Man Ho Choi, Sun Ju Kim, Bark-Lynn Lew, Woo Young Sim, Bong Chul Chung mh_choi@kist.re.kr

Pathological and epidemiological studies show racial variations in the metabolic patterns of testosterone (T) and dihydrotestosterone (DHT) as well as the activity of their metabolic enzyme 5α -reductase. As the incidence of male-pattern baldness (MPB) varies between countries and ethnic groups, the concentrations of hair steroids in Koreans were compared with Caucasians with and without hair-loss. Twelve steroids in the vertex hair were examined by gas chromatography-mass spectrometry-based steroid signatures. The MPB related androgens



including T, DHT and Epi-T were up-regulated in both Korean groups, whereas pregnenolone was significantly higher in both Caucasian groups. Based on the higher incidence of MPB in Caucasians, the higher levels of DHT in the Korean populations suggest that hair-loss is not only affected by 5 α -reductase, but the affinity of androgen receptors. Regarding the metabolic ratios, 5 α -reductase and 3 β -hydroxysteroid dehydrogenase were up-regulated in Caucasians (P < 0.03) and Koreans (P < 0.006), respectively, and their activities were associated with androstenedione, which is the immediate precursor of T. This suggests racial differences within hair steroids and steroid metabolic enzymes can explain the clinically observed differences in the endocrinology of MPB between racial groups.

Thermoresponsive amphiphilic star block copolymer photosensitizer: smart BTEX remover

by sequential block copolymerization of styrene and N-isopropyl acrylamide from a photocatalytic porphyrin core containing eight chain transfer agent (CTA) arms, through a reversible addition fragmentation transfer (RAFT) process (PDI < 1.3).

copolymer with a porphyrin core was synthesized

These water soluble star block copolymers were then applied in a water purification system, which efficiently captured and removed environmentally toxic organic materials such as benzene, toluene, ethylbenzene, and xylene (BTEX) from the water by hydrophobichydrophobic interaction with the inner PS block segments and decomposed them by the core porphyrin induced photo-oxidation reaction under UV irradiation (benzene ~ ethyl benzene: $\sim 4.0\% > xylene: \sim 5\% > toluene: \sim 17\%$). Following the catalytic reaction, the star block copolymer photosensitizer was easily recycled by simple filtration because the hydrophilic PNIPAM exterior was thermoresponsive, which allowed for the star block copolymer photosensitizers to agglomerate in water by increasing temperature over its lower critical solution temperature (LCST) (~32 °C).



Polymer Chemistry

2013; 4: 2400-2405.

Kie Yong Cho, Jae-Woo Choi, Sang-Hyup Lee, Seung Sang Hwang, and Kyung-Youl Baek* baek@kist.re.kr

Thermosensitive amphiphilic PNIPAM-PS star block

Efficient catalytic conversion of ammonia borane to borazine and its use for hexagonal boron nitride (white graphene)

Journal of Materials Chemistry A 2013; 1: 1976-1981.

Sung-Kwan Kim, Hyunjin Cho, Myung Jong Kim*, Hee-Jun Lee, Jin-hyung Park, Young-Boo Lee, Hwan Chul Kim, Chang Won Yoon, Suk Woo Name and Sang Ook Kang* myung@kist.re.kr

White graphene (hexagonal boron nitride) is a new type of material where carbon atoms in graphene are exchanged with boron and nitride alternatively. Due to an atomically flat surface and dielectric properties, white graphene is useful for various applications, including nanoelectronics, thermal management, and functional coatings. Here, we report methods from precursor synthesis to layer-controlled white graphene synthesis.

Nickel nanoparticles (NiNPs) prepared in tetraglyme (TG) efficiently catalyzed the conversion of ammonia borane (AB, NH₃BH₃) to borazine (B₃N₃H₆). Under the optimized conditions, 3 mol% of the NiNPs were introduced into a 1.5 M AB solution in TG and held at 80°C for 6 h under a dynamic vacuum that was maintained at 30 torr. Borazine was isolated through a series of -45°C, -78°C, and -196°C traps to give (-78°C trap) pure borazine in 53% yield. The borazine produced was then utilized as a molecular precursor for high-



quality h-BN (white graphene) and large-area h-BN sheets were prepared by applying low pressure chemical vapor deposition (LPCVD). Ultra-thin (single to a few layers) h-BN was synthesized on Ni foil at the optimal ratio between borazine and NH_3 , and the number of layers was tuned by varying the NH_3 partial pressure.

lonic-liquid catalyzed decarboxylation of glycerol carbonate to glycidol

Journal of Catalysis 2013; 297: 248-255.

Ji Sik Choi, Fidelis Stefanus Hubertson Simanjuntak, Ji Young Oh, Gun Im Lee, Sang Deuk Lee, Minserk Cheong, Hoon Sik Kim, Hyunjoo Lee hjlee@kist.re.kr

Glycidol is a chemical compound that is suitable for use as a plastics modifier, surfactant, fire retardant, etc. Conventionally, glycidol is produced by oxidation of allyl alcohol using hydrogen peroxide as the oxidant in the presence of a tungsten oxide-based catalyst or through the reaction of epichlorohydrin with bases. These processes have several drawbacks, such as the high cost of raw materials and/or the generation of waste byproducts. In this study, we synthesized glycidol from direct decarboxylation of glycerol carbonate (GLC) in the presence of various kinds of ionic liquids (ILs) as catalyst. This process makes economic and environmental sense because glycerol carbonate can be obtained from the derivatization of glycerol, a co-product of biodiesel production. Our results showed that ILs bearing an anion with medium hydrogen bond basicity, such as NO₃⁻ and I-, exhibited higher glycidol yields than those having an anion with a low or strong hydrogen bond. The presence of zinc salt with a medium Lewis acidity improved the yield of glycidol. Carrying out the reaction in a continuous system containing a high boiling point solvent, such as DMPEG



350 or dibenzyl ether, enhanced the selectivity of glycidol due to less contact between the IL and glycidol.

Decarboxylation of glycerol carbonate was catalyzed by ionic liquids.

Computation calculation revealed the first step is the anion-assisted ring-opening of glycerol carbonate followed by the ring closure, resulting in the formation of glycidol.

Impacts of solid retention time, nitrification and microbial activity on the attenuation of pharmaceuticals and estrogens in membrane bioreactors

Water Research

2013; 47: 3151-3162.

Sung Kyu Maeng, Byeong Gyu Choi, Kyu Tae Lee, Kyung Guen Song kgsong@kist.re.kr

This study investigated the effects of solid retention time (SRT), nitrification and microbial activity on the attenuation of pharmaceuticals, estrogens and total estrogenic activity, using identical bench-scale membrane bioreactors. Phenacetine, acetaminophen, pentoxyifylline and caffeine, bezafibrate, ibuprofen, fenoprofen, 17β-estradiol and estrone were effectively attenuated even at a short SRT (8 d). However, the attenuation efficiencies of gemfibrozil, ketoprofen, clofibric acid and 17α-ethynylestradiol (synthetic estrogen) were dependent upon SRTs (20 and 80 d). Some acidic pharmaceuticals (diclofenac, gemfibrozil, bezafibrate and ketoprofen) and 17α -ethynylestradiol were influenced by nitrification. Relatively high removal efficiencies were observed for 17β-estradiol and estrone (natural estrogens) compared to 17α -ethynylestradiol (synthetic estrogen) when nitrification was inhibited. Most of the selected pharmaceuticals were not significantly attenuated under presumably abiotic conditions created by adding sodium azide, except phenacetine, acetaminophen and caffeine. In this study, carbamazepine was found to be recalcitrant to biological wastewater treatment using membrane bioreactors, regardless of SRT length or microbial activity.



Apparatus and method for manufacturing manganese oxide-titania catalyst

US 8476185 (2013.07.02)

Jongsoo Jung / jongsoo@kist.re.kr

Disclosed are an apparatus and method for preparing a manganese oxide-titania catalyst. The apparatus for preparing a manganese oxide-titania catalyst includes: a vaporizer vaporizing a manganese precursor and a titanium precursor; a carrier gas supply line supplying a carrier gas; (which carries precursor vapors vaporized by the vaporizer to a reactor); to the vaporizer; an oxygen supply line supplying an oxygen source to the reactor; the reactor reacting the precursor vapors with the oxygen source to synthesize a manganese oxidetitania catalyst; and a collector condensing and collecting the manganese oxide-titania catalyst synthesized in the reactor. The method for preparing a manganese oxide-titania catalyst includes: 1) vaporizing a manganese precursor and a titanium precursor; 2) carrying precursor vapors (vapors of the manganese precursor and the titanium precursor) and an oxygen source to a reactor; 3) reacting the precursor vapors and the oxygen source to synthesize a manganese oxide-titania catalyst; and 4) condensing and collecting the manganese oxide-titania catalyst. According to the present disclosure, mass production of manganese oxide-titania catalysts with high decomposition efficiency of organic compounds can be prepared through fewer and continuous processes.



Remote control apparatus with dialing scheme providing haptic sensations

US 8471686 (2013.06.25) Laehyun Kim / laehyunk@kist.re.kr

A remote control apparatus that provides haptic sensations. The remote control apparatus includes a dial, a motor, a rotary encoder, an encoder counter, a control unit and a motor controller. The remote control apparatus further includes a storage unit, a movement detection unit and a display unit. When a user selects an icon displayed on the display unit by rotating the dial, the control unit interprets the user instruction through the rotary encoder and the encoder counter to control the motor to generate torque for causing a haptic sensation corresponding to the interpreted instruction via the motor controller. While the haptic sensation is conveyed to the user through the dial, the control unit controls an appliance to perform the interpreted instruction thereon.



Resistance switchable conductive filler for ReRAM and its preparation method

US 8450712 (2013.05.28)

Sang-Soo Lee / s-slee@kist.re.kr

Disclosed are a resistive random-access memory (ReRAM) based on resistive switching using a resistance-switchable conductive filler and a method for preparing the same. When a resistance-switchable conductive filler (prepared by coating a conductive filler with a material whose resistance is changeable) is mixed with a dielectric material, the dielectric material is given resistive switching characteristics without losing its inherent properties. Therefore, various resistance-switchable materials having various properties can be prepared by mixing the resistance-switchable conductive filler with different dielectric materials. The resulting resistance-switchable material shows resistive switching characteristics comparable to those of the existing metal oxide film-based resistance-switchable materials. Accordingly, a ReRAM device having the inherent properties of a dielectric material can be prepared using the resistanceswitchable conductive filler.



Apparatus and method for balancing and damping control in whole body coordination framework for biped humanoid robot

US 8447429 (2013.05.21)

Yong Hwan Oh / oyh@kist.re.kr

Apparatus and a method for balancing and damping control in whole body coordination framework for a biped humanoid robot. The method comprises the steps of: (a) damping the structural vibration of the main body of the robot caused when the robot walks; (b) compensating for the trajectories of the zero moment position (ZMP) and the center of mass (COM) of the robot which changes in accordance with the damping of the structural vibration; and (c) compensating for the body orientation of the robot which changes in accordance with the damping of the structural vibration and the trajectory of the COM



UPnP control point and UPnP device based on the UPnP network and connecting method using the same

US 8443123 (2013.05.14)

Sang Chul Ahn / asc@kist.re.kr

Disclosed herein are a UPnP control point and a UPnP device based on the UPnP network and a connecting method using the same, in which a service connection is performed. The service connection is restrictedly performed using a configuration file defined to allow or deny a connection with a specific device on the UPnP network, so that only devices specified by a user can be connected.



Photostimulation apparatus

US 8348986 (2013.01.08)

Jee Hyun Choi / jeechoi@kist.re.kr

A photostimulation apparatus may include: a membrane for insertion into a living body, and at least one cell disposed on the membrane. Each cell may include a first light source for irradiating light to a photosensitive material in the living body. Further, a photostimulation apparatus may include: a membrane for insertion into a living body, and at least one first light source disposed on the membrane for irradiating light to a photosensitive material in the living body. Since the photostimulation apparatus is placed on the surface of cortex or dura, it may minimize damage of the brain tissue and may activate and/or inhibit a large area simultaneously using light.



Coupling device for coupling a rotating shaft and a rotating member, and rotating device using the same

US 8366560 (2013.02.05) Woo Sub Lee / robot@kist.re.kr

A coupling device has a flexible member that is vertically connected to a rotating shaft and whose length can be varied in an inward or outward direction with respect to the rotating shaft, an elastic member that is connected to the flexible member and causes the flexible member to receive an elastic force in the outward direction with respect to the rotating shaft, and a cam member that is rotatably connected to the rotating shaft and delivers the rotating force of the rotating shaft to a rotating member. The cam member has a through-hole having an initial-position portion in which the flexible member is positioned at the initial stage and a displaced-position portion which has a smaller diameter than the initial-position portion and of which the diameter is constant about the rotating shaft, and another through-hole having an initial-position portion in which the flexible member is positioned and a return-position portion of which the radius decreases from the initial-position portion.



Method for preparing metal oxide sol, method for preparing metal oxide thin film using said sol, and solid oxide fuel cell comprising said thin film

US 8383286 (2013.02.26)

Hae-Weon Lee / hwlee@kist.re.kr

A metal oxide thin film structure for a solid oxide fuel cell, prepared by a method comprising the dispersal of a metal oxide nanopowder in a metal oxide salt solution and subsequent coating of the resulting metal oxide powder dispersed sol and the metal oxide salt solution on a porous substrate, has excellent gas impermeability, excellent phase stability, and is devoid of cracks or pinholes.



Light-emitting material comprising photoactive group-bonded polysilsesquioxane having a ladder structure, thin film using the same and organic electronic device comprising the same

US 8441005 (2013.05.14)

Kyung Youl Baek / baek@kist.re.kr

Disclosed is a light-emitting material including a polysilsesquioxane having a ladder structure with photoactive groups bonded to a siloxane backbone. In addition to superior heat resistance and mechanical property, the light-emitting material provides improved coatability and film property when prepared into a thin film. Further, it provides higher luminous efficiency than existing organic-based light-emitting materials.



Celebrating the 47th anniversary of KIST Feb 7, 2013

On February 7, 2013, KIST celebrated its 47th birthday with a ceremony held on KIST's Seoul campus.

President Kil Choo Moon, in his commemorative address, stated, "The science and technology sector and government-invested research institutes in Korea are confronted with enormous changes this year," and, "We must have an institutional revival and become born again as a research institute loved by all the people of Korea. Further, we must create new growth engines by taking up the challenges of future technologies based on selftransition and innovation, making a contribution towards creating new jobs, which is one of the major pending issues in Korea, and through resolving national social issues."

At the commemorative ceremony, the KIST Indonesia



ODA Project Team received the Grand Prize for their efforts in designing, establishing and operating an on-site pilot plant in Serre-Pon, Indonesia, that can produce bioethanol using agricultural wastes. This was recognized as a new ODA model for developing countries. In addition, Dr. Seong Moo Cho of the Carbon Convergence Material Research Center at KIST received the Songgok Science and Technology Award for having published several research papers in Korea and overseas on the development of nano-fiber technology.

KIST Launches "K-Club" to support SMEs Mar 8, 2013

KIST joined the Korea Industrial Technology Association (KOITA) and the Korea Trade Investment Promotion Agency (KOTRA) in launching the K-Club, a program designed to support promising small and medium enterprises (SMEs) by sharing KIST's accumulated expertise involving both manpower and technology. Member SMEs of the K-Club receive intensive consultation to help them grow into successful mediumand large-scale companies.

This first year, a total of 31 SMEs became members of the K-Club. KIST plans to select 10-20 SMEs each subsequent year after a thorough review process, which will include evaluation of applications and onsite visits. Candidates will be evaluated based on the quality of their company's technology, its marketing potential, previous patents, and current financial status.

KIST plans to strengthen all parts of the program, including technology commercialization, marketing, management, financial consultation, research staff support, and inter-industry networking, by providing experts who will work closely with member SMEs. "The launching of the K-Club not only complies with the policies of the new administration on vitalizing a creative economy, but it will also become an exemplary case for supporting SMEs by government-sponsored research institutes," said KIST President Kil Choo Moon.





Generous Donation by Former and Current Scientists of KIST

Mar 12, 2013

On March 12, 2013, Director Jong Oh Park (The Robot Research Center of Chonnam National University, Former Senior Researcher at KIST), Professor Byeong Gyu Kim (The Aerospace and Mechanical Engineering Department of the Korea Aerospace University, Former Senior Researcher at KIST) and Dr. Tae Song Kim (Current Senior Researcher at the Bio-micro System Research Group of KIST) together donated 150 million Korean Won to KIST's Science Sharing Fund. Their donation represented a portion of royalties received from transferring a patented technology overseas. These scientists developed a robotic colorectal endoscopy technology that moves freely and smoothly, even inside severely undulated and curved large intestines, by applying the movement principle of an inchworm. This technology was subsequently transferred to an Italian medical equipment and supply company, ERA Endoscopy, for 1 million Euros (approximately 1.5 billion Korean Won) in February 2012. This robotic colorectal endoscopy technology markedly reduces the pain and discomfort experienced with current colorectal endoscopy equipment and reduces the risk of puncturing the intestinal lining during procedures.

KIST established the Science Sharing Fund to support community social welfare activities by sharing profits generated through homegrown technology transfers. The fund is composed of voluntary donations from KIST



98 KIST NEWS

employees as well as outside contributions and is used for engineering scholarships, support to developing countries and social volunteering services. It also helps fund special initiatives of KIST such as support of outstanding academic researchers and the expansion of research areas. Since its inception in April 2012, a total of 460 people have participated and donated over 400 million Korean Won, with an additional 1.5 billion Won promised as of the end of February 2013. KIST has set a goal of raising 11.8 billion Won by 2015.

KIST Develops New Process for Manufacturing Nano-Substances Apr 8, 2013





Dr. Sang Woo Kim

Dr. Jae Pyeong Ahn

Researchers at KIST have developed a manufacturing process for nano-substances that is easier, more economical and more environmentally friendly than existing procedures.

Dr. Sang Woo Kim of the Clean Energy Research Center and Dr. Jae Pyeong Ahn of the Property Analysis Center succeeded in developing technology for manufacturing multi-dimensional semiconductor nanosubstances (nano-particles, nano-sheets, and nano-wire) by freely utilizing nano-particles as though assembling Lego blocks. Their research was published in the recent issue of *Scientific Reports* (the April issue), a sister magazine of *Nature*.

Multi-dimensional semiconductor nano-substances are now widely used in all areas of industry in catalysts, semiconductors, nano-elements, sensors and solar cells, but impurities such as mold (templates) or metallic growth catalyst arising from existing manufacturing procedures





necessitate their later removal at significant expense and complexity.

KIST's joint research team tackled both these issues by developing a technology for manufacturing nano-crystalloid nucleons into a higher-dimensional configuration by combining and disassembling them just like Lego blocks into a lower-dimensional configuration. This was achieved by reacting Gadolinium (Gd) and Cerium (Ce) within the mixed fluid of supercritical carbon dioxide-ethanol by using supercritical fluid technology without a template or growth catalyst.

It is highly anticipated that this technology will allow manufacturing of multi-dimensional nano-substances in a diverse range of materials by freely assembling them. It will now be possible to easily manufacture semiconductor nano-substances of high purity without a cleaning process, which considerably reduces environmental pollution. In addition, the joint research team presented the possibility of nano-substance control using the supercritical fluid process, which sheds new light on the mechanism of generation and growth through microanalysis of multi-dimensional nano-substances.

Celebrating the 10th Anniversary of KIST's Gangreung Institute May 9, 2013

The Gangreung Institute, KIST's first branch, celebrated its 10th anniversary on May 9, 2013. At that time, a new vision was announced, "A Better Life for Mankind through Natural Products" and a fresh goal established, "Development of Source Technology in Research Focusing on a Long Healthy Life."

Since its establishment, the Institute has used

natural ingredients that are abundant in the regions of the Taebaek Mountain Range and the East Sea in its research to develop a wide range of products, including soap, cosmetics, beauty products, functional foods and anti-cancer drugs. The Institute has presented over 450 research papers and 162 applications for patents, resulting in 71 patents registered by the end of 2012.

The Institute's focus on natural resources reflects a worldwide demand for natural products. Products made from natural substances can be consumed for prolonged periods of time, and their efficacy and safety has already been verified through extensive clinical testing of their ingredients. They also have the advantage of producing fewer side effects than synthetically produced foods and medicines and have relatively higher success rates and lower development costs. Natural product development





also has a very high investment efficiency, achieving maximum outcomes with minimum investment. It is more lucrative than developing a new synthetic medicine since the size of the investment and the time it takes for development can be reduced substantially.

In 2004, the Gangreung Institute carried out its first research project which involved using medicinal plants to fight cancer. Since then, the Institute has conducted important research projects into cancer, diabetes, obesity, high blood pressure and dementia in order to find natural substances for the treatment of such diseases.

Among its notable research achievements, the Institute has commercialized a functional health food to improve liver function using the extract from *Youngia denticulatum*, a plant native to Korea. It has also established a treatment system for acidic coal mine drainage using a neutralizing agent, a soil contamination restoration system, and developed materials for soap using the antibiotic effects of medicinal herbs from Mongolia. Additionally, the Institute succeeded in developing basic materials for high-grade cosmetics using the water obtained from boiling the Japanese sea cucumber that thrives in the East Sea and transferred that technology to a company that farms sea cucumbers.

In order to distinguish its research on natural substances, the Gangreung Institute plans to sharply

increase its capacity by 2020 in order to achieve three major strategies: (1) the establishment of a KGI Library with the most detailed database on natural substances in Korea; (2) the establishment and operation of the High Throughput Screening (HTS) system on natural substances; and (3) the pursuit of the "K-Farm Project," which is a large-scale research project on natural substances that most companies or universities cannot attempt due to its size, cost, and complex management requirements.

Initiation of the Hongneung Hakdang Lecture Program

May 10, 2013

The first Hongneung Hakdang lecture series event took place on the KIST campus with the theme, "Convergence of Humanity, Science and Art."

Hongneung Hakdang is a special lecture program where speakers from universities and research institutes in Hongneung share important ideas in the humanities, arts and sciences with the public in an easy-to-understand way. Since Hakdang was historically a local education center that cultivated talented individuals and shared knowledge within the community, Hongneung Hakdang was developed to play a similar role in Hongneung and its surrounding area.

Lecturers in the First Hongneung Hakdang included Seong Wook Hong, Fellow at the Korea Institute of





Advanced Study (Sciences: Rationality and Imagination, Its History and Variations), Jong Hoe Kim, Professor at Kyunghee University (Humanities: Creative Imagination and Humanistic Cognition), and In Soo Cho, Professor at the Korea National University of Arts (Arts: Visual Joy of Endless Imagination). They made their respective presentations as keynote speakers on why convergence is so important today.



KIST Europe Forschungsgesellschaft mbH

KIST Europe was founded as an overseas branch of KIST in Saarbrücken, Germany, on February 10th, 1996, with three main objectives:

- to expand Korea's expertise in the field of science and technology by conducting research activities in Europe (EU On-Site Research);
- to establish a base for international technology exchange and joint research with Europe, as well as Germany (Korea-EU Cooperation Hub); and
- to support Korean companies looking to expand overseas into Europe (Industry Support).

Initially, 14 employees worked in leased office and laboratory space at the University of Saarland until it was decided that the institute should establish its own buildings. In 2000, the first institute building was constructed on the university campus. It joined other prestigious research institutes already located on campus, including the Leibniz Institute for New Materials (INM), the German Research Center for Artificial Intelligence (DFKI), the Max-Planck Institute for Computer Science (MPI) and the Fraunhofer Institute for Nondestructive Testing (IZFP). In 2010 a second building was completed to serve as an industry and research cooperation center. The potential use of this building was maximized by the installation of a bio-lab with high biosafety standards. Other Korean Institutes such as KETEP, KAERI and KICT, as well as the German company, Ursapharm, have successfully installed branch offices and labs in this second building.

Despite the many challenges associated with its establishment, KIST Europe has been able to achieve real success in environmental and bioscience research. Currently, 18 national and international patents have been registered and 67 patent applications are pending. In addition, KIST Europe has enabled collaboration between Korea and the EU in the form of joint research, the hosting of international meetings and conferences, and the establishment of international programs. Participation and support of several international collaboration programs (EU FP7 KORRIDOR, KESTCAP), the organization of workshops (Bubble Tech to Bio App Lab on a Chip 2011, Germany-Korea Nano Bio Material Expert Workshop 2013), and cooperation with the German Federal Government on several projects (AiF Project, BMWi) are examples of the cooperation profile of KIST Europe.

To support Korean industry, KIST Europe has provided registration services for Korean companies to meet EU chemical regulations known as REACH (Registration, Evaluation, Authorization and Restriction of Chemicals), and to date, has acted as the sole representative for 25 of these companies. Due to our experience and expertise in this area, KIST has provided the Korean government with valuable input as it establishes similar chemical regulations, K-REACH, on a national scale.

Under the direction of the current Director, Professor Dr. Ho Seong Lee, and the Head of Research, Professor Dr. Andreas Manz, KIST Europe today employs a diverse staff of about 70 employees from 12 countries. Current research areas range from microfluidics, cellular immunotherapy,

o **2000-2003** >>>



Completion of the 1st Building (April 10, 2000)



Inauguration Ceremony of the 2nd Director (Dr. Oh Kwan Kwon) (June 27, 2001)



Inauguration Ceremony of the 3rd Director (Dr. Jun Keun Lee) (January 27, 2003)

nanomaterials for medical treatments, and mixture toxicity. The institute effectively supports coordination of S&T activities between Korea and the EU and provides a convenient, well-equipped base for joint research. Based on these many ongoing activities, KIST Europe is poised to become a center of excellence in environmental and integrative nano-bio-engineering research while continuing to promote effective S&T collaboration between Korea and the EU.

• 2006-2009 >>>



Celebration of the 10th Anniversary of KIST Europe (April 27, 2006)



Korea-EU Joint Workshop (December 3, 2007)



Groundbreaking for the Construction of the 2nd Building (May 7, 2008)



KIST – KIST Europe Joint Workshop (October 1, 2008)



Recruitment of Professor Andreas Manz as KIST Europe's R&D Director (September 28, 2009)

• 2012 >>>



High-Level Round Table Meeting (July 25, 2012)



Inauguration Ceremony of the 6th Director (Professor Ho Seong Lee) (September 3, 2012)



Korea-EU Cooperation Workshop (December 3, 2012)

• 2013 >>>



Excursion of KIST Europe Staff Members (May 29, 2013)



Expo on EU Programs at the Max Planck Institute of Computer Science – The Visit of the Minister President of Saarland at KIST Europe's booth (July 3, 2013)

At the Forefront of Brain Science Research: KIST's World Class Institute

KIST's World Class Institute (WCI) was initiated in 2010 with the support of Korea's Ministry of Education, Science and Technology. The purpose of the institute is to develop patents on a global level through joint research in which outstanding international researchers participate in and manage projects of international importance in conjunction with Korean scientists.

Within the WCI, the Center for Functional Connectomics conducts research in diverse areas of brain science. For example, the Center developed technology for imaging the brain using a light that reveals previous secrets of how the brain acquires spatial information. Papers associated with these findings were published last year in several renowned science magazines, including *Cell* and *Nature Methods*. In 2012 this finding was selected as the leading news item in *Science and Technology News of Korea*, published by the Korean Federation of Science and Technology Societies.

In addition, the Center has succeeded in investigating the principle of how the brain acquires spatial information and the diverse roles of suppressive nerve cells by using a treadmill training device for experimental animals in conjunction with a brain circuitry analysis technology, known as optogenetics. This research was conducted jointly with the Janelia Farm Research Campus of the Howard Hughes Medical Institute in April of last year. It is highly anticipated that this research will be instrumental in discovering the principles of spatial memory within the hippocampus, which could represent a breakthrough in the treatment of brain disease associated with the hippocampus, such as epilepsy, Alzheimer's, and others.

Moreover, the Center has developed technology for visually observing living synapses, and is currently developing source technology for the treatment of various diseases, including dementia, Parkinson's disease, epilepsy, insomnia, depression, etc., that originate in nonneuronal brain cells that are not yet well understood.

Attracting Global Talent

Currently, the WCI consists of just over 90 personnel. Professor George Augustine of the Neurobiology Department of Duke University (USA), the foremost authority in optogenetics, serves as Director of the WCI. Professor Augustine has published over 100 research papers in renowned journals including *Nature, Science* and *Cell*, and he was the first person to prove the relevance between synapse plasticity and calcium.

Following its invitation to Professor Augustine to head the institute, KIST took steps to recruit outstanding researchers and scientists and ultimately selected about 20 researchers internationally. All of the candidates offered a position accepted. This 100% acceptance rate seems to have resulted from WCI's promise to guarantee full support and to create a research environment that would eliminate any distractions from administrative red tape.



Dr. Chang Joon Lee

Dr. Chang Joon Lee, Assistant Director of the WCI, holds a Ph.D. in functional connectomics and is a founding member of the WCI. He has the principle responsibility for purchasing equipment and setting up laboratories for the Center to be fully operational when the international researchers arrive. In addition, KIST offers exceptional terms and conditions for the recruits as well as shouldering the investment in setting up fully equipped facilities, which include a number of experimental laboratories including: the Neuron-Network Imaging Research Lab; the Glial Cell Research Lab; the Neurocircuit Cell Biology Research Lab; the Hippocampus Network Dynamics Research Lab; and the Post-Synaptic Regulations Research Lab.

Currently, the Center is involved in brain circuitry

mapping using the optogenetics technique, which is the most advanced technology yet found. It selectively regulates the activities of neurons by using optical stimulation with a specific wavelength that transcends existing anatomical brain analysis methods. Using this technology, the Center is hoping to make significant advancements in better understanding the causes of cerebral diseases, discovering treatment targets and developing treatment options.

Dr. Chang Joon Lee, Assistant Director of WCI, states, "We have been able to smoothly carry out our active research since WCI projects have meshed well with the long-term goals of KIST and the timing has been just right. Our Center will expend considerable future effort to ensure outstanding research results in the area of cerebral disease, thus becoming an excellent example for others and a household name for brain mapping."

A Research Environment That Encourages Creative Thought

"I wanted to pursue research if I were given a good research environment, including a favorable research system and support. I decided to come to Korea because the terms and conditions offered by the WCI were excellent in comparison to advanced countries, and because I wanted to share the benefits I was receiving." (Dr. Jinny Kim)

While in the U.S., Dr. Jinny Kim heard about the establishment of the WCI and decided to come to Korea. She was attracted to the fact that she could work with people with a wide range of experience in an international setting and was also drawn by the offer of excellent research conditions. In her application she stated, "In fact, I, as a Korean, also had a strong desire to give back some of the benefits which I have received from Korea in hopes that it will be passed down to the next generation."

Dr. Kim is very pleased with the international atmosphere of the WCI, especially its system of encouraging free expression and opinions among staff, regardless of position or seniority, She states, "This setting allows members to argue and challenge opinions in order to carry out research and development creatively. Since the WCI is so international, we all speak mostly in English, which has the advantage of allowing the members to express creative and great ideas without hesitation or reservation."

Last year Dr. Kim developed a new neuron network mapping technique. After this achievement, she commented, "I want to improve research outcomes by sharing my experience and philosophy with other members of the laboratory." She added, "Although the field of neuroscience is enormously extensive, I am working to pursue research that produces solid information. Just as strong bricks should be used when building a house, our research should help build a solid foundation for neuroscience."

"Optogenetics technology is the most advanced technology in the field of optical science. I decided to join the WCI because I wanted to carry out research at the Center where the foremost authority and pioneer in this area was the new director." (Dr. Ryuichi Nakajima)



Dr. Jinny Kim



Dr. Ryuichi Nakajima

Dr. Ryuichi heard of the WCI while working as a researcher at Dalhousie University. He looked forward to working at an institution that would truly allow his skills to flourish. In particular, he was thrilled at the opportunity to work with Professor George Augustine, a pioneer and foremost authority in optogenetics, the most advanced technology in the field of neuroscience.

Dr. Ryuichi states, "I found the WCI attractive because it has a state-of-the-art infrastructure and research is facilitated with top-level equipment. In addition, the Center provides full support to its researchers. Working at the WCI doing research in optogenetics technology will most definitely be a great benefit and asset to my research career and I will surely strengthen my research capability, as well."

Working jointly with KAIST and Korea University, Dr. Ryuichi utilized optogenics technology to discover a particular neuron circuitry which is involved in drug addiction and feeling of compensation. In applying the results of this research, Dr. Ryuichi says that he plans to devote himself to research that aims to unravel the mystery of neurons related to fear and depression.

"I visited Korea for the first time for my interview for the WCI. I had a good first impression of the research facility and I felt enormous energy from the people at the Institute." (Dr. Bradley Baker)

Dr. Bradley Baker is pursuing research at the WCI on the development of technology that observes neurons through probes.



Dr. Bradley Baker

Although he did not have much knowledge of Korea prior to his arrival at the Center, he distinctly remembers his first impression of Korea. He said, "I arrived in Korea for the first time for an interview at the Center, and right away, I felt good energy at KIST and in Seoul. My personal research capability has been reinforced and strengthened since I've been here."

Last year Dr. Baker was promoted to senior researcher at KIST, along with four other WCI researchers, in recognition of his research performance on WCI projects. "I am thankful that I was able to have the opportunity to pursue research here," he said, and added, "Currently, a lot of amazing research is being carried out at the Center. I have decided to stay at KIST in order to continue my research since going elsewhere would interrupt my career."



Cover image : Silver nanoparticle/ion-decorated micrometer-sized magnetic hybrid colloids bite away bacteria and chemisorb viruses using their surface structure. After use, they can be collected using a magnet.

[J. Mat. Chem. B., 2013; 1: 2701.] - Reproduced by permission of The Royal Society of Chemistry

Editorial Information

Editor-in-Chief Byung Gwon Lee

Editorial Board Members Hae-Young Koh Hyun Kwang Seok Il Ki Han Joonyeon Chang Youngsoon Um Seok Won Hong Jongjoo Kim Seung Yun

Managing Editor

Sang-A Chang sarachang@kist.re.kr Editorial Office Telephone +82-2-958-6039 Web Address www.kist.re.kr/en

English Advisory Services

Anne Charlton The Final Word Editing Services the_final_word@live.com



Korea Institute of Science and Technology

Hwarangno 14-gil 5, Seongbuk-gu, Seoul 136-791, Republic of Korea Tel +82-2-958-6313 www.kist.re.kr/en E-mail kiwihyang@kist.re.kr

Korea Institute of Science and Technology

Biomedical Research Institute



Developing Biomedical Technologies to Improve Human Health and Well-Being

The Biomedical Research Institute is responsible for developing cognitive and motor rehabilitation technology for the elderly and disabled. It also conducts research for cutting-edge devices and materials to replace human tissues and organs as well as innovative medical technology (personalized medicine) for more effective diagnosis and treatment of different diseases. The Biomedical Research Institute is at the forefront of Korea's health and medical welfare research.

