Graphical Abstract:

$(\text{Zr}_{0.55}\text{Al}_{0.10}\text{Ni}_{0.05}\text{Cu}_{0.30})_{99}\text{Y}_1$ BMG

- Ar- or Ca-ion implantation
- 10 keV or 50 keV

**Surface Engineering of a Zr-based bulk metallic glass with low energy Ar- or Ca-ion
implantation**

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Abstract: In the present study, low energy ion implantation was employed to engineer the surface of a Zr-based bulk metallic glass (BMG), aiming at improving the biocompatibility and imparting bioactivity to the surface. Ca- or Ar-ions were implanted at 10 or 50 keV at a fluence of 8×10^{15} ions/cm² to (Zr_{0.55}Al_{0.10}Ni_{0.05}Cu_{0.30})₉₉Y₁ (at.%) BMG. The effects of ion implantation on materials properties and subsequent cellular responses were investigated. Both Ar- and Ca-ion implantations were suggested to induce atom displacements on the surfaces according to the Monte-Carlo simulation. The change of atomic environment of Zr in the surface regions as implied by the alteration in X-ray absorption measurements at Zr K-edge. X-ray photoelectron spectroscopy revealed that the ion implantation process has modified the surface chemical compositions and indicated the presence of Ca after Ca-ion implantation. The surface nanohardness has been enhanced by implantation of either ion species, with Ca-ion implantation showing more prominent effect. The BMG surfaces were altered to be more hydrophobic after ion implantation, which can be attributed to the reduced amount of hydroxyl groups on the implanted surfaces. Higher numbers of adherent cells were found on Ar- and Ca-ion implanted samples, while more pronounced cell adhesion were observed on Ca-ion implanted substrates. The low energy ion implantation resulted in concurrent modification in atomic structure, nanohardness, surface chemistry, hydrophobicity, and cell behavior on the surface of the Zr-based BMG, which were proposed to be mutually correlated with each other.

Keywords: Bulk metallic glass, Ion implantation, Bioactivity, Mechanical, Wettability.

1. Introduction

In the past decade, biomedical applications of bulk metallic glasses (BMGs) have garnered a large amount of scientific efforts [1-4]. Researches to exploit the biomedical potentials of BMGs as implants or other biomedical usages stemmed from their favorable properties, among which the low elastic modulus [4], high wear resistance [5, 6], good corrosion resistance [7, 8], and extraordinary thermal plastic processability [9, 10] usually topped the list.

Comparisons between BMGs and traditional crystalline biomedical alloys (i.e., Ti alloys, stainless steel, Co alloys, etc.) unveiled the advantages of BMGs as promising candidate biomaterials, especially as hard tissue prostheses [11]. BMGs excel in mechanical properties, substantiated by their combination of low elastic modulus and high wear resistance, which is beneficial to reduce the risk of some current medical complications (i.e., stress shielding and particle disease) [4, 11]. In the meantime, a number of BMGs were found to be stable in simulated body solutions, with corrosion resistance/rates comparable with those of highly corrosion resistant crystalline biomedical alloys [12, 13]. Moreover, their superplastic deformation in supercooled liquid region and small solidification shrinkage facilitate the accurate formation of complex medical devices and may reduce cost of post processing [3, 9].

A number of studies investigating the biocompatibility of BMGs, including *in vitro* cellular responses and *in vivo* tissue responses to BMGs with nondegradable or biodegradable characteristics have been reported, with findings suggesting general biocompatibility of this group of novel materials [1-4, 11, 14-16]. For long-term implant applications, Zr-based BMGs have been favorably considered due to high

1 biocompatibility of the base element Zr and the availability of highly glass-forming
2 compositions [15, 17]. However, the surfaces of Zr-based BMGs are typically bioinert,
3 which implies a lack of direct bonding between the tissue and the implant made from
4 such materials. For applications such as orthopedic implants, it is highly beneficial to
5 impart bioactivity into BMGs to facilitate biological bonding with bone tissues, so that
6 the long-term performance and success of the implants can be warranted.

7 Surface engineering techniques have been widely utilized to modulate surfaces of
8 traditional biomedical alloys to endow bioactivity [18-20]. Bearing the similar concept, a
9 number of surface engineering methods have been exploited to modify surface properties
10 of BMGs, including micro-arc [21], coating [22], ion implantation [23, 24], and so forth.
11 Among these surface modification techniques, ion implantation alters the original surface
12 of the substrate material and the modified layer is intimately integrated into the substrate,
13 providing a homogeneous and controllable doping of ions of interest [20, 25]. Such
14 versatility has propelled ion implantation as the choice of surface modification in our
15 studies to tune surface bioactivity of Zr-based BMGs. Previously, we investigated ion
16 implantation of a Zr-Al-Cu-Ag BMG with Ca- and Ar-ions [23]. The choice of Ca-ions is
17 inspired by the mineral phase of bone tissue, where the principal constituents are calcium,
18 phosphate, carbonate, and hydroxyl ions [26]. Furthermore, the effectiveness of Ca-ion
19 implantation has been demonstrated on traditional Ti alloys, which led to surface
20 enrichment of Ca and consequently improved cell/material interaction on Ti surfaces,
21 presumably due to the accelerated and increased precipitation of calcium phosphate that
22 enhanced the bioactivity of Ti surface [19, 20]. The use of inert element Ar for ion
23 implantation lies in its similarity in the atomic mass to that of Ca, thus allowing us to

differentiate the physical damage effect from chemically induced changes [23]. Additionally, Ar ion implantation has been reported to improve surface properties of BMGs and other metallic material, particularly surface hardness and corrosion resistance, which are advantageous for biomedical applications [20, 27]. Here, we are extending the use of ion implantation for BMG surface modification to a $(\text{Zr}_{0.55}\text{Al}_{0.10}\text{Ni}_{0.05}\text{Cu}_{0.30})_{99}\text{Y}_1$ (at.%) BMG substrate. Compared with the aforementioned Zr-Al-Cu-Ag BMG, this Y-containing Zr-based BMG exhibits higher corrosion resistance, which is much more favourable for applications where long-term implantation is desired [7]. The addition of Y to the $\text{Zr}_{55}\text{Al}_{10}\text{Ni}_5\text{Cu}_{30}$ BMG led to a significant improvement in corrosion resistance, as evidenced by the spontaneous passivation behaviour, comparable pitting resistance with medical grade stainless steel, and equivalently low corrosion penetration rate to Ti6Al4V alloys [7]. Moreover, the biocompatibility of this particular Y-containing Zr-based BMG was also well documented through *in vitro* cell culture studies up to 28 days [11]. The biosafety of the Zr-based BMG was initially revealed by studying bone-forming cell responses on the substrate, including adhesion, proliferation, and differentiation [11]. It is worth noting that the good corrosion resistance and biocompatibility of the Zr-Al-Ni-Cu-Y could be interconnected with each other, since the low toxicity could be partially attributed to the minute amounts of metal ions released from the corrosion resistant substrate.

The objective of the current study is to understand the effects of low energy ion implantation with Ca- and Ar-ions on the Y-containing Zr-based BMG. Material properties and responses of bone-forming cells on the ion irradiated Zr-based BMG

substrates are characterized, and the effects of ion implantation parameters including the type of ions and the acceleration energy are discussed.

2. Materials and Experimental Methods

2.1 Materials and ion implantation

Zr-based BMGs with a nominal composition of $(\text{Zr}_{0.55}\text{Al}_{0.10}\text{Ni}_{0.05}\text{Cu}_{0.30})_{99}\text{Y}_1$ were fabricated using arc melting and copper mold casting technique. Pure metals of the constituent elements were melted and mixed in high purity argon atmosphere. The ingots were flipped over and re-melted for multiple times to assure homogeneity. Master ingots were shattered and heated up to molten state using induction heating in quartz tubes and injected into copper molds to form plate samples with a geometry of $60 \times 10 \times 2 \text{ mm}^3$. The amorphous state of the resulting BMGs was verified using small angle synchrotron X-ray diffraction (Figure S1 in supplemental material) [11]. Testing samples ($5 \times 5 \times 2 \text{ mm}^3$) were cut from the as cast plates, manually ground to 1200-grit surface finish using SiC sandpapers, and polished with 1- μm alumina slurry using a VibroMet vibratory polisher (Buehler, USA). Samples were ultrasonically cleaned in acetone, ethanol, and distilled water for 5 min, respectively.

Ion implantation to the polished substrates was conducted using an Eaton NV-200 implanter. Ar- or Ca-ions were implanted at 10 keV and 50 keV with a fluence of $8 \times 10^{15} \text{ ions/cm}^2$. The aperture size of 13 cm^2 was chosen to allow implantation of multiple specimens simultaneously. BMG specimens were attached to a heavy metal sample holder, which allowed heat dissipation during the implantation process and restricted the temperature rise of the specimens and subsequent crystallization [23].

The damage (i.e., displacement per atom, dpa) and ion distribution in the BMG substrates were simulated using Stopping and Range of Ions in Solids (SRIM) package developed by Ziegler et al., which employs Monte-Carlo algorithms and a quantum mechanical treatment of ion-atom collisions [28]. In the full-cascade simulation, the displacement binding energy, surface binding energy, and lattice binding energy were set to be 40 eV, 2 eV, and 3 eV, respectively. The density of the Zr-based BMG applied in the simulation was experimentally measured to be 6.65 g/cm³ by Archimedes' principle. The experimental fluence of 8×10^{15} ions/cm² was plugged in to calculate the depth profiles of dpa and ion concentration.

2.2 Materials characterizations

Effect of ion implantation on the surface microstructure of the Zr-based BMGs was investigated using grazing incidence X-ray diffraction (GIXRD). Changes in atomic structure after ion irradiation was examined using X-ray absorption spectroscopy (XAS) analysis at Beamline 9-BM-B,C of the Advanced Photon Source at Argonne National Laboratory. The K edge of Zr was measured by detecting total electron yield signals, which constrained the sampling depth and collected the structural information within the near-surface region.

Surface morphology and the concentration of the constituent elements on the surface of the as cast and implanted Zr-based BMG samples was examined by the scanning electron microscopy (SEM) coupled with energy-dispersive X-ray (EDX) spectroscopy. To further examine the changes in surface morphology after ion implantation, atomic force microscopy (AFM) was employed under tapping mode. A more quantitative analysis of the surface chemistry of Ar- or Ca-ion implanted samples

1 was characterized using VersaProbe™ Scanning X-ray photoelectron spectroscopy (XPS)
2 Microprobe. Measurements were performed employing an Al K α monochromated X-ray
3 source and the results were analyzed using XPSPEAK 4.1 data processing software.
4 Shirley-type background was used during data analysis.

5 Nanoindentation experiments were performed at room temperature using a
6 Hysitron Triboindenter (Hysitron, Inc., USA) to characterize surface nanohardness. A
7 Berkovich diamond tip was used with an effective tip radius of 233 nm. For each
8 specimen, 15 indentations were carried out at 15 μ m intervals. A loading function
9 composed of a 5 s loading segment, 2 s holding segment and 5 s unloading segment was
10 applied with a peak load of 9 mN. The indentation depth was determined to cover both
11 modified layer and the substrate. Therefore, the nanohardness results are not solely
12 presenting the hardness of the modified layer, but providing evidence on the general
13 changes in surface hardness after ion implantation.

14 Surface wettability of the samples was determined by measuring the water contact
15 angle on the substrates using a CAM-Plus contact angle goniometer (Cheminstruments,
16 USA). About 5 μ L of deionized water (DI water) was placed on the samples. Water
17 contact angles were measured using the half-angle sessile-drop method (US Patent
18 5268733) at room temperature.

19 **2.3 Cell culture study**

20 Murine derived bone-forming MC3T3-E1 pre-osteoblasts (ATCC, USA) were employed
21 for *in vitro* cell culture studies to investigate cellular responses to the ion implanted
22 surfaces. The cells were maintained in growth medium composed of alpha-minimum
23 essential medium (α -MEM) supplemented with 10% fetal bovine serum and 1%

1 penicillin–streptomycin (Invitrogen, USA). Cells were harvested at confluency by
2 detaching with 0.25% trypsin with ethylene diamine tetraacetic acid (Invitrogen, USA)
3 for cell culture studies. Prior to cell seeding, the BMG substrates were sterilized by 30-
4 min ultraviolet light exposure.

5 Initial cell attachment and viability of the cells on the substrates were examined
6 using a LIVE/DEAD[®] Viability/Cytotoxicity Kit (Invitrogen, USA), which labels live
7 cells with calcein AM and dead cells with ethidium homodimer-1 (EthD-1). Cells at a
8 density of 1×10^4 cells/cm² were seeded on the as cast or implanted substrates and
9 incubated for 8 h. Samples were then rinsed with phosphate buffered saline (PBS) and
10 stained with the dye-containing PBS solution (2 μ M calcein AM and 4 μ M EthD-1) for
11 20 min at 37 °C. Fluorophore-labeled cells were viewed under a Zeiss Axio Observer A1
12 inverted fluorescent microscope (Zeiss, Germany). The number of adherent cells on each
13 substrate was quantified by counting live cells within each area of view of the fluorescent
14 images. More than 9 images for each experimental group were analyzed.

15 Cell adhesion to the substrates was further investigated by immunofluorescent
16 staining of vinculin, a key protein highly expressed in focal adhesions formed by
17 adherent cells [29]. Cells were seeded on the substrates at a density of 5×10^3 cells/cm².
18 After 8 h of incubation, cells were fixed and stained following a protocol as previously
19 described [11]. Briefly, the cells were fixed with 4% paraformaldehyde, permeabilized
20 with 0.1% Triton X-100 (Sigma, USA) and blocked with 1% bovine serum albumin
21 (Sigma, USA). Vinculin was labeled with anti-vinculin monoclonal antibody (Santa Cruz
22 Biotechnology, USA) followed by secondary staining with Alexa Fluor 488 goat anti-
23 mouse IgG1 (Invitrogen, USA). Cell nuclei were dyed using 4,6-diamidino-2-

phenylindole (DAPI) (Chemicon, CA). After staining, samples were mounted on glass coverslips using fluoromount G (Southern Biotech, USA) antifade mounting solution and observed using the Zeiss fluorescent microscope.

3. Results

3.1 SRIM simulation

The simulation results of the ion irradiation induced disorder and implanted ion concentration are plotted in Figures 1a and b for Ar- and Ca-ion implantations, respectively. The calculated profiles for both types of ion implantations exhibited distorted bell shape, revealing the depths of the modified surface layer and the maxima of disorders and ion concentrations resulting from the ion bombardments. The thicknesses of the influenced region, which is characterized by the stopping ranges of the ions, were calculated to be around 25 nm and 80 nm for 10 keV and 50 keV ion implanted samples, respectively, regardless of the type of ions used. Slight differences were noted in the distributions of disorders and implanted ions after Ar- and Ca-ion implantation. The resultant peak values of disorders created by 10 keV and 50 keV Ar-ion collisions were 17 dpa and 21 dpa at 5 nm and 17 nm. Peak concentrations of Ar were 11.0 at.% and 3.3 at.% located at 7.5 nm and 30 nm for 10 keV and 50 keV implantations, respectively. Similar, although slightly higher, damages were produced by Ca-ion irradiations, with the disorder maxima identified to be about 18 dpa for 10 keV ions and 23 dpa for 50 keV ions. In terms of implanted ion concentration in the matrix, the highest concentrations of Ca were 11.2 at.% and 3.5 at.% at about 7.5 nm and 27 nm from the surface for the 10 keV and 50 keV Ca-ion implanted samples. For both types of ion irradiation, the distributions of implanted ions are much more dispersive for the 50 keV as compared to

the 10 keV implanted samples. With the increase of acceleration energy, ions were able to penetrate deeper and introduce more collision events to the substrate. Since the fluence of ions was fixed, higher acceleration energy was able to create a broader distribution of the implanted ions with a consequent reduction in the corresponding peak concentration.

3.2 Surface microstructure

The SRIM simulation implied that ion irradiation at low energies (10 or 50 keV) would produce displacements of atoms and subsequent atomic structure changes to the surface of the Zr-based BMGs. The microstructure of the surface region after ion implantation was examined using GIXRD. The representative pattern for 50 keV Ar-ion implanted sample is shown in Figure S2. The absence of crystalline peaks and the presence of the characteristic diffusive peak confirmed that the amorphous structure was maintained after ion implantations. In order to experimentally verify these ion irradiation induced changes, XAS was measured at the K-edge of the based element Zr. The complete XAS spectra of the as cast and ion irradiated Zr-based BMG substrates were presented in Figure 2 as a function of X-ray energy. A crystalline Zr metal foil was also included in the XAS study as the standard reference material. The overall general shapes of the absorption edges for all the BMG samples were distinctly different from the reference metal foil. A further comparison of the XAS spectra between the ion implanted BMGs and non-treated BMG revealed a clear change in terms of peak position and width, shown from the enlarged view in inset of Figure 2. We attribute these differences to the changes in the local atomic environment for Zr atoms as a result of ion implantation, which is consistent with the prediction by simulation. Nonetheless, the disparities in XAS spectra among the implanted BMGs were minute, necessitating further detailed measurements and analyses

1 to capture comprehensive information on the atomic structural changes induced by
2 different implantation conditions.

3 ***3.3 Surface morphology and chemistry***

4 Prior to any ion implantation process, the Zr-based BMGs appeared mirror-like, typical
5 of the polishing regime adopted in this study. Representative SEM secondary electron
6 image collected for the surface of the Zr-based BMGs after 50 keV Ca-ion implantation
7 is presented in the inset of Figure 3. The surface appeared featureless, indicating no
8 observable morphological changes on the specimens after 50 keV Ca-ion implantation.
9 Similar smooth morphology was observed for other ion implanted groups (data not
10 shown). Moreover, no significant variations in surface topography were observed, as
11 shown in the AFM images (Figure S2). EDX elemental analysis (Figure 3 and Table S1)
12 of the average surface composition after ion implantation indicated that the surface
13 composition was close to the alloy nominal composition. However, considering the
14 sampling depth of EDX is within a few microns [23], the compositional data obtained
15 with this technique is inevitably obscured by the non-treated region beneath the ion-
16 implanted layer (less than 100 nm deep).

17 In order to focus solely on the ion-implanted region, we employed a more surface
18 sensitive chemical analysis method, namely XPS. The sampling depth of XPS is related
19 to the inelastic free mean path of the electrons, which ranges within a few nanometers
20 [30]. XPS scans for the elements present on the surface were recorded and the elemental
21 quantifications are shown in Figure 4. Using samples implanted with 50 keV Ca-ion as an
22 example, the narrow scans in Figure 4a displayed the peaks for O1s, Zr3d, Al2s, Cu2p3,
23 Y3d, and Ca2p3. Minute amount of Ni was also present on the surface indicated by the

Ni2p3 signals collected (data not shown). Quantification results of element concentrations for the as cast and ion implanted samples shown in the stacked bar graph (Figure 4b) manifest the dominance of O and Zr on the surface, which accounts for the major constituent of the surface oxide film. Oxygen was present in the forms of both metal oxide (marked as M_xO_y) and surface hydroxyl groups (marked as OH). The largest amount of hydroxyl group was found on the as cast sample, and a significant decrease was observed after implantation. Similar to the as cast surface characterized in our previous study [11], the surface of ion implanted Zr-Al-Ni-Cu-Y BMG was covered with a layer of passive film mainly composed of ZrO_2 as well as Al_2O_3 and Y_2O_3 (Figure 4). The concentration of Y_2O_3 has been slightly increased, while Ni and Cu were mostly depleted on the surface after implantation (Figure 4b). After Ca-ion implantation, peaks for Ca can be explicitly distinguished in an energy range between 342 eV and 357 eV, which corresponds to the Ca2p3 peaks. The presence of Ca on the surfaces after Ca-ion irradiation was evidenced in the forms of Ca and CaO, indicating that the bioactive element, Ca, has been successfully introduced to the bioinert Zr-based BMG surface through low energy Ca-ion implantation. The concentration of the Ca on the surface after 10 keV Ca-ion implantation (13.4 at.%) is slightly higher than that after 50 keV Ca-ion implantation (12.9 at.%).

3.4 Surface nanohardness

Surface nanohardness obtained from the indentation tests for each sample is summarized in Figure 5. The nanohardness for the as cast surface is around 6.45 GPa. After ion irradiation, the BMG surfaces were hardened, as shown by the increases in nanohardness. With an indentation depth larger than 250 nm, it was measured that 10 keV Ar ion

1 implantation led to a 1.6% increase of surface nanohardness to 6.55 GPa, and 50 keV Ar
2 ions gave rise to a 2.6% increase to 6.62 GPa in nanohardness. The surface nanohardness
3 increased by 5.7% to 6.82 GPa after 10 keV Ca implantation and 7.3% to 6.92 GPa after
4 50 keV Ca-ion implantations, both in comparison with the as cast sample.

5 ***3.5 Surface wettability***

6 Surface hydrophilicity for the Zr-based BMGs was varied after ion implantation, which
7 was demonstrated through water contact angle measurements using sessile drop method.
8 The histogram in Figure 6 depicted a general increase in water contact angle after ion
9 implantation, indicating that the surfaces were made less hydrophilic by ion implantation.
10 A more significant increase in water contact angle was found on the Zr-based BMGs
11 subjected to lower energy (10 keV) ion implantation, in comparison with those implanted
12 with the same ion but higher energy (50 keV). In the meantime, under the same
13 acceleration energy, the magnitude of surface wettability change was also dependent on
14 the type of ion implantation, with Ar-ion implantation showing a more pronounced effect
15 than Ca-ion implantation. The insets in Figure 6 present shadow images of the water
16 droplets, which provide a qualitative view of the wettability change of the substrates as a
17 result of ion implantation.

18 ***3.6 Cell adhesion***

19 Live/Dead staining images of the bone-forming MC3T3-E1 cells growing on the as cast,
20 Ar-ion implanted, and Ca-ion implanted samples are presented in Figure 7. Cells
21 exhibited high viability on all the samples, as seen by the strong green fluorescence for
22 live cells and the absence of red signals for dead or membrane disrupted cells. The results
23 indicate that, similar to their as cast counterparts, the ion implanted samples also

supported initial cell attachment and exhibited no cytotoxicity to the cells. Furthermore, these images show no distinct differences in adherent cell morphology among these various substrates. Quantification of the live cell density on each substrate following 8 h of culture is summarized in the bar graph in Figure 7. On average, higher adherent cell densities were found on ion implanted samples, as compared with the cell density on the as cast sample.

Cell adhesion was further accessed by fluorescent staining of vinculin (Figure 8). Overall, cells exhibited a polygonal morphology on all the substrates. The concentrated green fluorescent signals observed mainly on the periphery of the cytoplasm of adherent cells suggest the formation of focal adhesion plaques at the corresponding locations. Both as cast and implanted samples were supportive of focal adhesion development, though qualitatively the number of focal adhesion plaques displayed by cells attached on the ion implanted substrates appeared to be higher than that on the as cast sample.

4. Discussion

The amorphous microstructure of BMGs was considered key to their unique properties. Therefore, low acceleration energies of ions were employed in our current study in order to maintain the amorphous structure on the surface. According to the simulation results, the maximum displacements per atom introduced by ion implantation were calculated to be smaller than 25 dpa. Such a low dpa is unlikely to introduce crystallization, which has been experimentally validated by GIXRD (Figure S2). This is in agreement with prior studies on Ar- or N-ion implantation to Zr- and Ti-based BMGs where the amorphous structure was observed after implantation using XRD and transmission electron microscopy (TEM) [24, 31]. Although the long range atomic ordering was not introduced

1 after ion implantation, the short range ordering on the BMG surface could be altered due
2 to the ion collision cascades. To probe this atomic structure changes, we employed the
3 nondestructive XAS technique, which requires no further sample preparation and
4 prevents potential artificial damage to the sample. The detections were limited within the
5 implanted surface regions by collecting total electron yield signals. XAS results
6 preliminarily revealed local changes in atomic structure after Ar- or Ca-ion implantation,
7 which could cause structural relaxation or change in free volume as suggested in a
8 previous report [23]. Further information on the atomic structure after ion implantation,
9 such as radial distances and coordination numbers of the neighboring atoms, can be
10 obtained by analyzing the extended x-ray absorption fine structure (the far-edge region of
11 XAS), which needs detailed data fitting and modeling in our future study.

12 Aside from the atomic structure modification, ion implantation to the Zr-based
13 BMG modified surface chemistry. The chemical changes to the implanted region were
14 predicted by SRIM simulation and the surface compositional variations were
15 substantiated by XPS analysis. Despite of the inert nature of Ar ions, Ar-ion implantation
16 modified the compositions of the surface layers on the Zr-based BMG by altering the
17 fractions of the constituent components (i.e., metal oxides, and hydroxyl groups) of the
18 ion implanted surfaces, as compared with the as cast surface. Ca-ion implantation
19 imparted more significant compositional alterations. According to the simulation, Ca ions
20 are more concentrated on the surface after 10 keV implantation, in contrast to a relatively
21 dispersive distribution of Ca over a broader region after 50 keV ion implantation. A
22 similar trend to the simulation results was revealed by XPS analysis, showing a higher
23 amount of Ca on 10 keV ion implanted surface than on 50 keV implanted one, although

1 the difference was not as significant as revealed by SRIM simulation. It is noted that
2 SRIM simulations were performed with nominal alloy composition of the Zr-based
3 BMG, whereas XPS measured the top surface covered by a layer of oxide film known as
4 passive film [11] of the Zr-based BMG. The discrepancy between SRIM simulations and
5 XPS measurements is mainly due to the fact that the composition of surface passive film
6 usually diverges from the underlying compositions.

7 Changes in microstructure and surface chemistry induced by ion implantation
8 may lead to or be directly correlated with the changes in surface properties (i.e., hardness,
9 wettability) and subsequent cellular responses. The enhancement in surface hardness by
10 ion implantation was demonstrated in the present study. When analyzing the
11 nanohardness of ion implanted samples, it is worth pointing out that the indentation depth
12 was approximately 250 nm, as shown in Figure S3, whereas the thickness of the modified
13 layer is 25 or 80 nm according to the simulation results. Therefore, each measurement of
14 hardness of the respective ion implanted sample was a result of a composite structure
15 composed of the ion implanted layer and the substrate material. In general, the
16 improvement in nanohardness after ion implantation may have been underestimated due
17 to the relatively large indentation depth. However, the improvement in nanohardness is
18 evident when comparing the Ar- and Ca-ion implanted samples with the as cast ones. Ion
19 bombardment generated surface hardening found in our study is in accordance with those
20 reported in previous research [23, 32]. The improvement in hardness may be attributed to
21 possible structural relaxation caused by irradiation, which was found to affect mechanical
22 properties of BMGs [23, 33]. On the other hand, the surface hardening resulting from Ca-
23 ion implantation was also found more considerable than from Ar-implantation, when

1 comparing the ion implanted samples with each other at the same acceleration energy.
2 The higher surface hardness resulting from Ca implantation may be attributed to
3 substantial chemical change on the surface due to the formation of ceramic-like CaO on
4 the surfaces and/or higher amount of metal oxides (i.e., ZrO_2 and Y_2O_3), as suggested by
5 the XPS analyses (Figure 4). The improved nanohardness by ion implantation implies a
6 higher wear resistance, which is beneficial for bone implant materials, in terms of
7 eliminating wear debris and subsequent particle disease.

8 Other than the mechanical changes, the change in hydrophobicity of the surface
9 was observed after implantation. It is acknowledged that the surfaces became more
10 hydrophobic after ion implantation, with Ar-ion implanted Zr-based BMGs being even
11 more hydrophobic than Ca-ion implanted substrates. The hydrophobicity change of the
12 tested specimens can be directly related to the amount of polar hydroxyl groups present
13 on the surfaces. Hydroxyl groups are hydrophilic groups due to their polarity, which
14 encourages their interaction with polar liquid (water). An increased amount of hydroxyl
15 groups was found to improve surface hydrophilicity [34, 35]. The most hydrophilic as
16 cast surface is related to the highest amount of hydroxyl group on the surface (Figure 4b).
17 Among the surface treated samples, those after 10 keV Ar-ion implantation held the
18 lowest amount of OH, hence highest hydrophobicity; whilst a relatively larger amount of
19 OH was observed on the less hydrophobic 50 keV Ca-ion implanted samples.

20 Corrosion resistance is a key feature of biomaterials, since the implant materials
21 will be facing the aggressive chemical environment of the body [36]. Corrosion
22 resistance of Zr-based BMGs was found to be dependent on surface properties (i.e.,
23 surface chemistry, wettability, etc). As discussed above, the depletion or reduction of Ni

1 and Cu on the alloy surface after implantation processes may lead to improved corrosion
2 resistance of the material [11]. Moreover, it was reported in a ZrTiCuNiBe BMG system
3 that a more hydrophobic surface could, to some extent, be related to higher corrosion
4 resistance [37]. The surface of our Zr-based BMG was altered to be more hydrophobic,
5 which also suggests a potential increase in corrosion resistance. In fact, an improvement
6 in corrosion resistance of a Zr-Al-Cu-Ag BMG after Ar- or Ca-ion implantation was
7 demonstrated in our previous study [23]. A detailed investigation on the effect of ion
8 implantation on corrosion resistance of BMGs is an interesting topic for future studies.

9 The change of hydrophobicity can also be correlated with cell attachment
10 behavior. It is well established that surface hydrophobicity can significantly influence the
11 protein adsorption behavior, which directly affect cell adhesion [38-40]. Ion implantation
12 was shown to make the surface more hydrophobic, and a relatively more hydrophobic
13 surface is considered to encourage protein adsorption [41]. Adsorption of proteins such as
14 fibronectin from the culture medium promotes cell adhesion [39]. Therefore, higher
15 numbers of adherent cells found on the ion implanted samples can be ascribed to the
16 change of surface wettability. In the meantime, surface chemistry is again considered an
17 influencing factor to cell adhesion. Promoted cell adhesion with the presence of Ca on the
18 surfaces in our study is in agreement with the higher cell adhesion and greater focal
19 adhesion formation due to Ca-ion implantation to Ti surface [42]. The synergistic effect
20 of wettability and chemistry changes lead to an equivalent improvement of cell adhesion
21 on Ca-ion implanted samples to that on Ar-ion implanted samples, although the
22 hydrophobicity change by Ca-ion implantation was not as prominent as by Ar-ion
23 implantation. In addition, the effect of chemical changes due to the presence of Ca was

1 reflected by the increased number and improved formation of focal adhesion on Ca
2 implanted samples. In fact, the mutual relationship among surface chemistry,
3 hydrophobicity, and cell adhesion was extensively studied and established for biomedical
4 alloys and polymers [42-45].

5 **5. Conclusions**

6 Low energy Ar- or Ca-ion implantations were performed in the present study and the
7 effect of ion irradiation on the resultant properties were characterized. The ions were
8 implanted at a low energy to avoid crystallization on the surface of Zr-Al-Ni-Cu-Y BMG.
9 The irradiation was found to alter short-range atomic structure based on simulation and
10 XAS measurements. By imparting ions to the surfaces, surface chemistry was tuned and
11 surface nanohardness was improved. The water wettability of the Zr-based BMG surface
12 was shifted to be more hydrophobic. Cells exhibited high viability and higher adherent
13 cell densities on ion implanted samples. Cell adhesion behavior is related to surface
14 chemistry and hydrophobicity variations. Further investigations will focus on
15 comprehensive study of ion irradiation induced microstructural changes of BMGs, and
16 examination of long-term cell behaviour on ion implanted BMG substrates.

17 18 **Acknowledgement**

19 Our gratitude goes to Dr. T.G. Nieh from the University of Tennessee for his kind advice
20 on nanoindentation test and data interpretation. We are thankful to Drs. N. Dahotre and F.
21 Kuo of the University of North Texas for their kind help on XPS experiments. We
22 appreciate Dr. T.B. Bolin from Argonne National Laboratory and Mr. Z. An from the
23 University of Tennessee for their assistance on XAS and grazing incidence synchrotron

1 X-ray diffraction measurements. Use of the Advanced Photon Source, an Office of
2 Science User Facility operated for the U.S. Department of Energy (DOE) Office of
3 Science by Argonne National Laboratory, was supported by the U.S. DOE under
4 Contract No. DE-AC02-06CH11357. The grazing incidence X-ray diffraction experiment
5 was performed with the generous help of Dr. Maulik Patel from the University of
6 Tennessee under the support of General Infrastructure Grant of DOE-NEUP (DE-
7 NE0000693). WH, PKL, and LH acknowledge the financial support from National
8 Science Foundations (CMMI-1100080). PKL very much appreciates the support from the
9 Department of Energy (DOE), Office of Nuclear Energy's Nuclear Energy University
10 Program (NEUP, 00119262), and the DOE, Office of Fossil Energy, National Energy
11 Technology Laboratory (DE-FE-0008855) with R.O. Jensen, L. Tan, S. Lesica, and V.
12 Cedro as program directors.

13

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1 **Figure Captions**

2 Figure 1 SRIM simulation for the distributions of disorders and ion concentrations after
3 a) Ar-ion and b) Ca-ion implantation at 10 keV and 50 keV acceleration energies.

4 Figure 2 Normalized XAS spectra at Zr K-edge for Zr-based BMGs before and after ion
5 implantation and Zr foil. Inset shows the enlarged spectra at Zr K-edge.

6 Figure 3 EDX composition analysis of 50 keV Ca-ion implanted Zr-based BMGs
7 (dashed lines indicate nominal alloy composition of the respective element; inset: SEM
8 secondary electron image).

9 Figure 4 a) XPS narrow scans of O1s, Zr3d, Al2s, Cu2p, Y3d, and Ca2p3 for 50 keV
10 Ca-ion implanted Zr-based BMG, and b) quantifications of the element concentrations on
11 the surface of the as cast and ion implanted Zr-based BMGs.

12 Figure 5 Surface nanohardness of the Zr-based BMG before and after 10 or 50 keV ion
13 implantation with Ar- or Ca-ions.

14 Figure 6 Water contact angles on indicated as cast or ion implanted substrates (inset:
15 shadow images of water droplets on corresponding substrates)

16 Figure 7 Live/Dead staining and density of adherent MC3T3-E1 cells on indicated
17 substrates after 8 h incubation (scale bar: 200 μm)

18 Figure 8 Green fluorescence staining of vinculin for MC3T3-E1 cells on indicated
19 substrates after 8 h incubation showing focal adhesion plaques (scale bar: 50 μm). Blue
20 staining illustrates cell nuclei.

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1 **Figure 8**
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