

# The Building Process of the Water Exclusion Zone under the illumination of Torsion Field

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## Abstract

The water exclusion zone (EZ) developed between the hydrophilic surface and the bulk water are believed to be the common phenomenon near the cell membrane. Typically, the EZ width extended up to tens or hundreds of micrometers from the vicinity of the hydrophilic surfaces, it should cause tremendous biological effect when EZ water surrounds the cells and affect the ion channels on the cell membrane. The zone width had been found to be widened by the illumination of infrared light or torsion field generated by a fast speed rotator made of nickel-manganese alloy. The building process of the water exclusion zone were observed both from the top and lateral sides simultaneously with/without the illumination of the left-handed or right-handed torsion field. A lateral explosive expansion of structured water phase was observed, the structured water has higher refractive index and leads to the leak of light in the Nafion film waveguide and form a beautiful cross-shape light pattern.

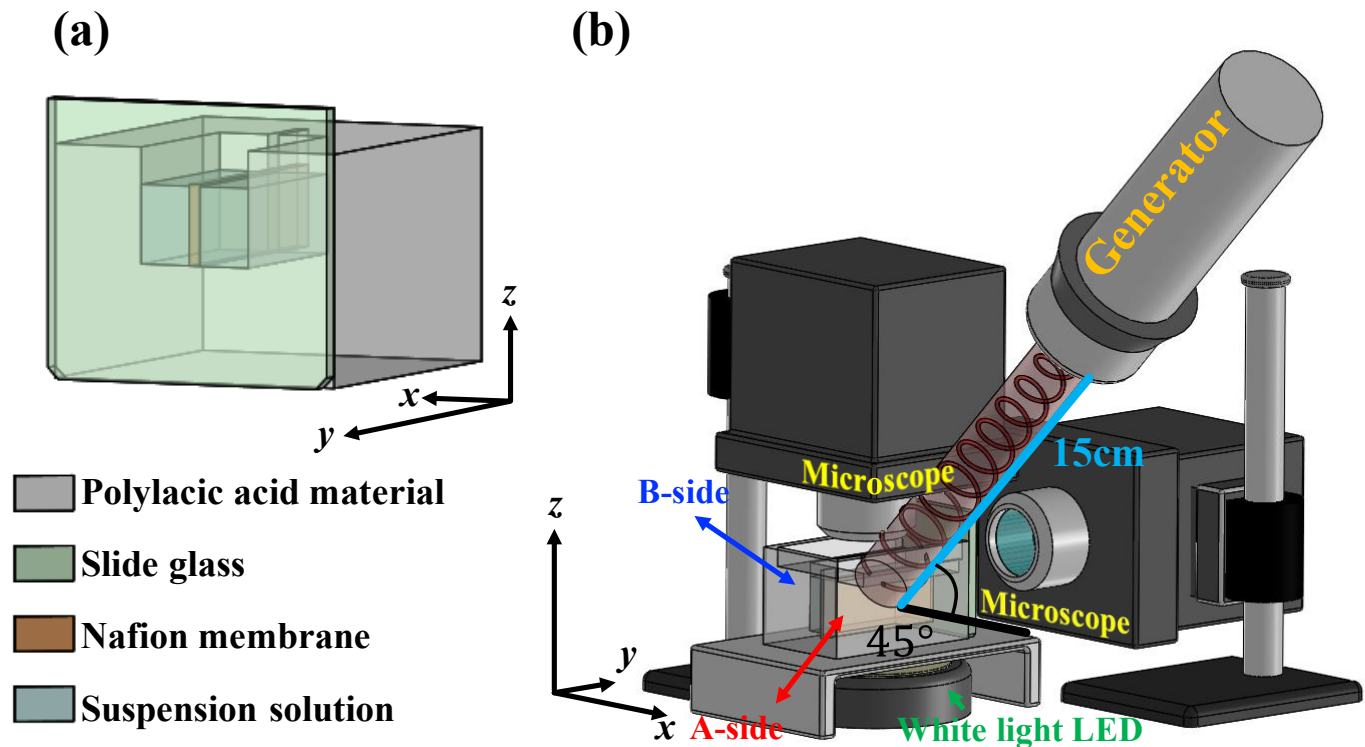
The water EZ width expansion ratio was measured at fixed time and found to be enlarged under the illumination of torsion field especially in the coming side of right-handed torsion field. This demonstrates that torsion field can indeed be detected by measuring the water EZ width ratio. It is expected that a torsion field display can be achieved in the future.

## Introduction

Exclusion Zone (EZ) — The fourth phase of water have been studied beyond a decade and arouse more

and more interest. It was proposed by Pollack at early 21st century.<sup>1</sup> The exclusion behavior of negatively charged nanobeads nearby the hydrophilic materials, i.e., Nafion film, have been discovered, the reordered water molecules built up at the interfacial layer between water and hydrophilic materials. The EZ could be observed in various hydrophilic surface, including ion-exchange beads<sup>2</sup>, hydrogels<sup>3</sup>, hydrophilic polymer<sup>4</sup>, and biological membranes.<sup>3</sup> Therefore, an interesting issue arises: what would EZ behave when it is surrounding the biological membranes. For example, phospholipid bilayer cytomembranes are ubiquitous in eukaryotic cell such as cell membranes, mitochondria, and karyotic cell. The EZ region is composed of structured water which excluded the other particles, i.e., nanobeads, up to tens to hundreds of micrometers. In addition, the zone was observed to develop whether the charges on both hydrophilic materials and nano-beads<sup>2</sup> are the same or opposite. This phenomenon indicated that the EZ water may develop everywhere. One of the important unsolved questions is “what is the biological effect of EZ region if it exists in cells?” because the hydrophilic interfaces are widely found in the basic units of organism, such as mitochondria, eukaryotic cell. Therefore, a lot of EZ researches were conducted recently to solve this problem.

Several factors influence the quality of the structured water, such as pH value of aqueous solution, concentration of suspension solvent, and the quality of hydrophilic surface. Furthermore, the electromagnetic radiation is one of the most important factors to affect the width of the structured EZ<sup>2,3</sup>, Near-infrared energy is the most capable one



**Figure 1.** The schematic diagram of (a) chamber for loading Nafion membrane and suspension solution. (b) two orthogonal microscopes were synchronized to observe the EZ water from top and side, the light source comes from the bottom of chamber. The left-handed or right-handed spiral torsion field generator was set along the  $45^\circ$  tilt angle with 15 cm distance between spintronic device and chamber, the side toward spintronic device (field coming side) was defined as A-side and the opposite side (field leaving side) was defined as B-side.

to converts ordinary bulk water into liquid crystalline water (EZ water).<sup>4</sup> The radiation provides the energy to enhance oxygen hydrogen (O-H) molecular bond vibration in water molecules, and help water molecules reordered in a hexamers ring structure which is the building block in the vicinity of hydrophilic material surface. On the other hand, quantum biology studies have shown that electron and nuclear spins play important roles in many life processes.<sup>5,6</sup> One of the significant example, electron spin entanglement and coherent spin transport are part of a possible explanation for the magnetic orientation of migratory birds.<sup>5,6</sup> In addition, the electron transfer processes is obviously affected by the double stranded DNA effect due to the spin filtration as demonstrated by Göhler *et al.*<sup>7</sup> These evidence indicated that the spins also play a role in a formation of structured water and demonstrated by He *et al.*<sup>8,9</sup> In the large scale space-time, the spin field is a necessary ingredient for the gravity theory discovered by Cartan<sup>10</sup> and embraced by Einstein.<sup>11</sup> It could be regarded as a field accompanying the rotation of the particle named torsion field.

However, the torsion field was ignored for a long time because the field is too weak to detect except for the extremely large objects, like astronomical motion. Macroscopically, the field was canceled by the random distributions of the rotation directions. To investigate the field of spin, the amplified spin field is required. The theory proposed by Liang *et al.*<sup>12</sup> in our group showed that, the spin-curvature force (torsion field) could be greatly enhanced when the spin field is coupled with a global rotation. In this work, the EZ region was illuminated by two torsion field generators (TFGs), with opposite chiral polarized nickel- manganese ferrite rotator. The magnetic field around the rotating devices is ignorable, only less than 1 milli-gauss, it is much smaller than the geomagnetic field. Through the spin Aharonov-Bohm (AB) effect<sup>13</sup>, the torsion vector potential is created by the spin states in the nickel-manganese ferrite. Two orthogonal sets of optical microscope were adopted to record the development and characteristics of EZ region under the torsion field radiation. The optical investigations of the building process of EZ region are reported and

discussed. Considering that water is the cradle of life and the physical properties of torsion field is unknown, this study may shed light to both fields.

## Experimental Methods

In this study, the spintronic device was invented by Alexandr Shpilman, and the details of the inner configuration had been introduced by Xian He *at el.*<sup>9</sup> We used two spintronic devices with identical configuration but different spin states in the nickel-manganese ferrite polarized by two different chiral configurations, i.e., left-handed and right-handed directions, respectively. As the nickel-manganese ferrite rotates, a torsion vector potential was generated by the spin-spin contact interaction.<sup>12</sup>

The hydrophilic material we used here is DuPont Nafion-117 membrane with 200  $\mu\text{m}$  in thickness<sup>14</sup> in general. The membranes were cut into 5 mm  $\times$  20 mm squares, and then were cleaned in a standard wet procedure as follows. First, the membranes were immersed into 1%  $\text{H}_2\text{SO}_4$  solution and cleaned by ultrasonic bath for 1 hour. Next, the membranes were rinsed by deionized water 5 times. Finally, the membranes were cleaned again in ultrasonic bath for 1 hour in deionized water. After the standard wet procedure, Nafion membranes were stored in deionized water over one night to maintain the moisture condition. The influence of  $\text{H}_2\text{SO}_4$  concentration on the EZ formation was mainly due to the unexpected damages on membrane surface caused by the high concentration  $\text{H}_2\text{SO}_4$ . To eliminate the surface damages, they were cleaned by 1% concentration  $\text{H}_2\text{SO}_4$  and then stored in deionized water until the experiment.

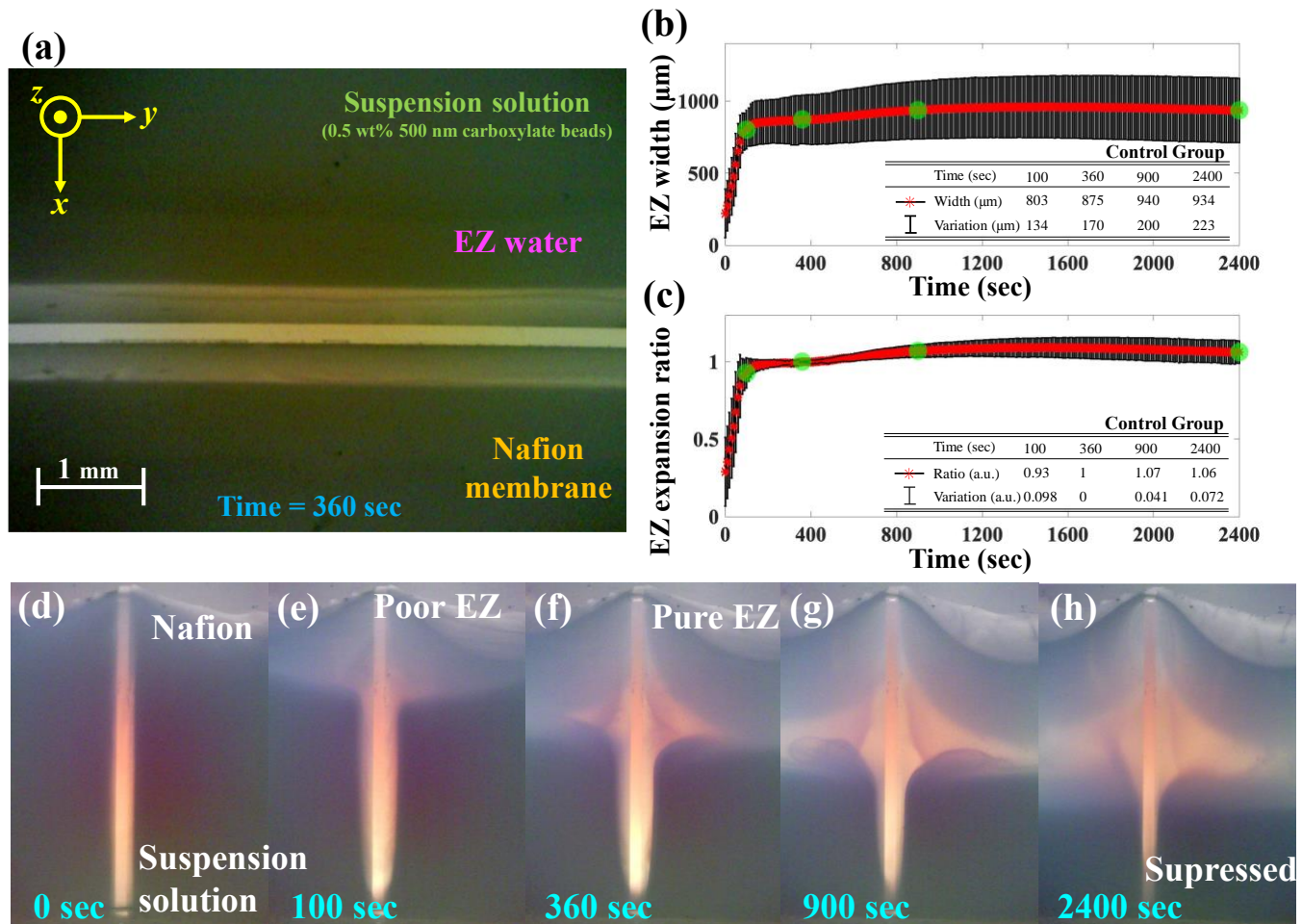
In order to observe the cross sections of EZ building processes and the lateral expansion phenomena, two microscopes with camera were installed both from side and top, they take photos simultaneously every 10 seconds. The chamber was made by white polylactic acid material and 3D printing technology. It was a cuboid of length 30 mm  $\times$  width 26 mm  $\times$  height 20 mm with a hollow rectangle and a ditch. The hollow rectangle size is length 16 mm  $\times$  width 10 mm  $\times$  height 10 mm and align with cuboid chamber in one side as shown in Fig. 1(a). Then a

slide glass was stuck on this side for side observation. On the opposite side, the size of ditch is length 6 mm  $\times$  width 1 mm  $\times$  height 10 mm to prevent the membrane from floating in solvent. The 640  $\mu\text{l}$  aqueous suspension of carboxylate nanospheres of 500 nm with 0.5wt% was injected into the hollow region. Immediately after, one side of prepared Nafion membrane was put against observation glass, and the other side was fixed inside the ditch. When the light came from the bottom of the chamber, it can barely pass through the suspending nanospheres, causing the dark region observed under top microscope. For the region inside the EZ, since particles were excluded by the EZ phase of water, the EZ region was bright under side microscope. In addition, the exclusion region and Nafion membranes are good waveguide material for light propagation. Finally, a great deal of light energy was trapped in the EZ region and observed in side view. This is why the contrast of side view was always higher than that in the top view.

In each experiment, the flux of light was fixed and the light was always turned on. During the first 360 secs (0 – 360 sec) of top view, the EZ water expanded rapidly and slowed down after 360 secs. Hence, the EZ width at 360 sec was defined as ratio 1 for normalization. Then the torsion field generator was placed along the 45° tilt angle at 15 cm distance away as shown in Fig. 1(b). The device turned on and off at the time 390 sec and 2190 sec, respectively. After the torsion field illumination, the EZ region was still observed for 210 secs (2190 – 2400 sec) continuously. The Nafion membrane and EZ region were observed in both side view and top view simultaneously by two microscopes synchronized with sampling rate at 1 picture per 10 seconds. The fluctuation of EZ region width and expansion ratio are measured to evaluate the measuring results.

## Results and discussion

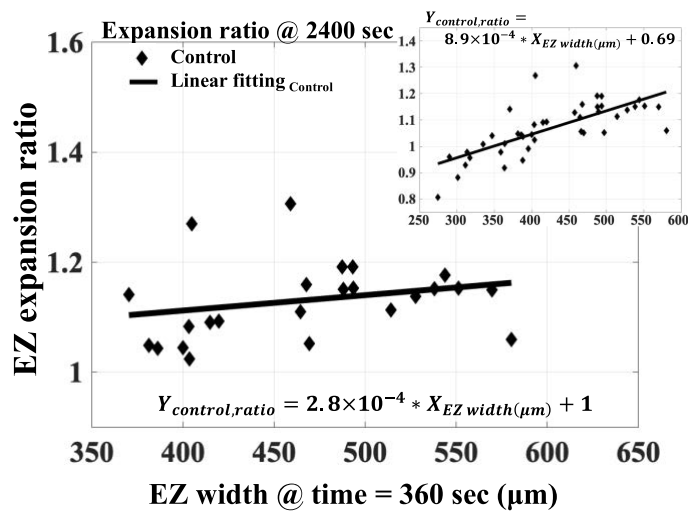
In the control group, 23 control experiments (without the illumination of torsion field) were measured and the width of EZ region was defined by the total average EZ region width divided by the length of nafion film times 200  $\mu\text{m}$  and there is one of top view image at time = 360 sec was shown in Fig. 2(a). The EZ width was calculated by the area of entire



**Figure 2.** The profile and analysis of control group. (a) The top view of EZ water squeezed between Nafion membrane and suspension solution at time = 360 sec. The EZ width was calculated by the area of entire exclude region divide by the horizontal length of Nafion membrane. The (b) expanding width and (c) normalized expansion ratio of 23 EZ width and variation were plot in red mark and black bar, respectively. The EZ width at time = 360 sec was defined as criteria width of EZ water in each experiment. (d), (e), (f), (g) and (h) were the side view of EZ water build up process at time = 0 sec, 100 sec, 360 sec, 900 sec, and 2400 sec, respectively. The orange long strip, gray region and dark area were Nafion, poor EZ water and suspension solution, respectively. The orange region appearing after time = 100 sec was pure and structured EZ water.

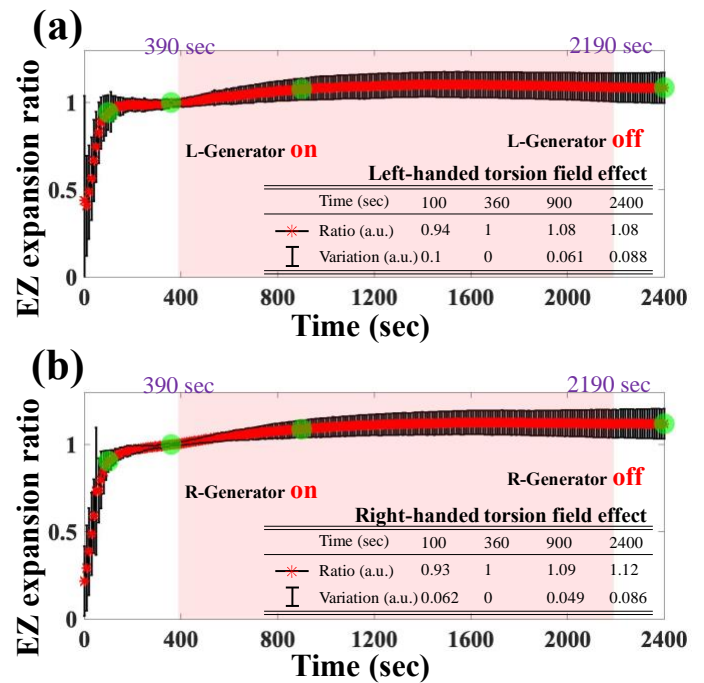
exclude region divide by the horizontal length of Nafion membrane. The statistical total EZ width of control group was shown in Fig. 2(b). After initial 360 secs, the width of EZ became stable and was identified by the obvious deceleration of expanding rate. Hence, the EZ width at time = 360 sec was utilized as the criteria ( $\equiv EZ_{width@360sec}$ ) of each experiment to eliminated the quality difference of Nafion membranes. The normalized statistical comparison was shown in Fig. 2(c). The EZ width still growth up in a very slightly expanding rate during 360 sec to 2400 sec. Here, the side view of EZ region was observed in order to understand the building process of EZ water. Five separated cross section pictures were shown in Fig. 2(d) – Fig. 2(h), and the corresponding ratio was labeled on Fig. 2(c)

at time = 0 sec, 100 sec, 360 sec, 900 sec and 2400 sec, respectively. At the initial 100 secs of the experiment, the poor EZ water was build up by lateral explosive growth at the top of the suspension water. Then the poor structured water grow quickly and push down the suspension nanobeads and good structured water start to form which has larger refractive index. Orange light began to leak from the Nafion waveguide and formed a cross shape pattern as shown in Fig. 2(e). As times go on, the pure and structured EZ region which was bright orange color at Fig. 2(f) emerged at around 200-250 sec. The phenomenon could be explained by the expanding rate. First, poor excluded region with loose structured water was composed due to the fast EZ expanding rate, means there are several nanobeads



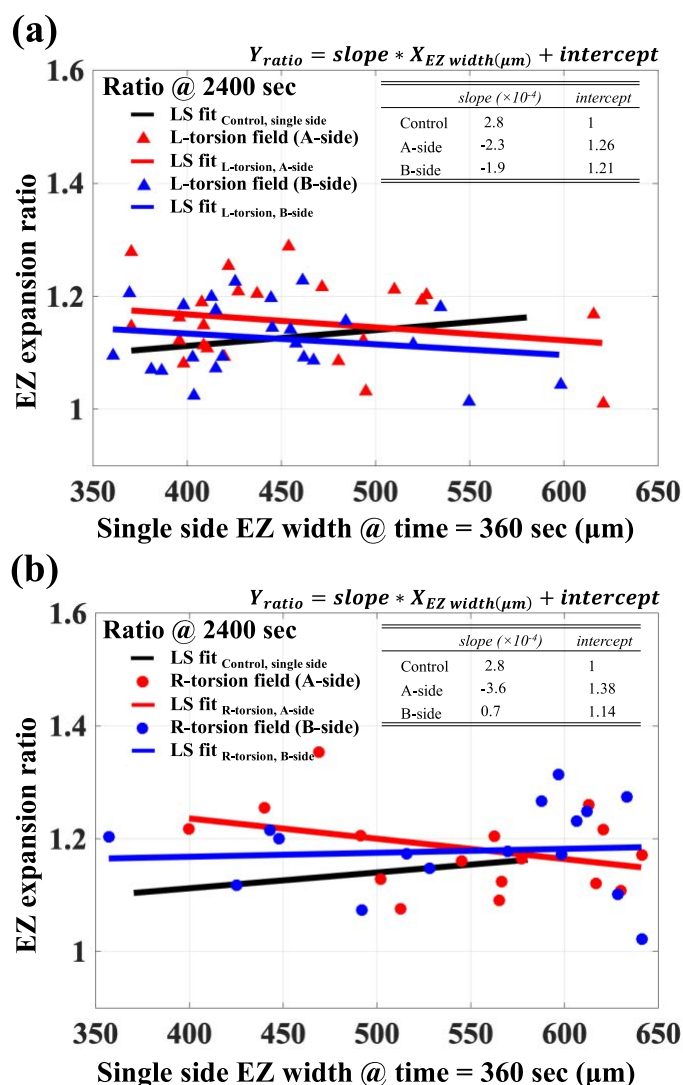
**Figure 3.** The expansion ratio of control group at time = 2400 sec in different criteria of EZ width.

were stuck in structured water. When the expanding rate became slower, the dense structured liquid water was ordered and the pure EZ region was formed. Then the region will continuous extend but in an unobvious expanding rate after 900 sec. Another noticeable that, the black region at bottom of image from Fig. 2(d) to Fig. 2(h) still shrink, it seems that the suspending solution was suppressed by a solid and structured EZ water. The different EZ region width observed from top view and side view in this work should be clarified by two major factors. One is the DOF length of objective len is only several tens to hundred micrometers in this work. The other is the observation plane different in two directions, the focal planes were fixed at the water-air interface and water-glass interface in the case of top view and side view, respectively. In addition, the effect of Nafion membrane quality and the final expansion ratio was illustrated and shown in Fig. 3. In the Fig.3 and the inset of Fig.3, the horizontal axis was the standard criteria of Nafion quality ( $EZ_{width@360sec}$ ), and the vertical axis was expansion ratio at time = 2400 sec. The larger EZ width was attributed to better Nafion quality as demonstrated in the inset of Fig. 3 by the linear least square fit (LS fit) of expansion ratio. The negative expansions ( $< 1$ ) at time = 2400 sec were observed when the initial EZ width is smaller than 350 μm which should be eliminated in this study due to the collapse of the poor-structured water during the measurement. In Fig. 3, the standard criteria for EZ width must be larger than 350 μm was chosen as a criterion to make comparison of torsion fields results with control.



**Figure 4.** The normalized expansion ratio of statistical EZ width and variation illuminated by (a) left-handed, (b) right-handed spiral magnetic field were plot in red mark and black bar, respectively. The control-similar EZ width at time = 360 sec was defined as criteria width of EZ water in each experiment.

To explore the effect of torsion field on the EZ water, the experiments under the chiral configurations (right-handed, R- and left-handed, L-) field illumination were repeated more than several tens-times. The time dependent statistical normalized EZ width expansion ratio of L-generator and R-generator were illustrated in Fig. 4(a) and Fig. 4(b), respectively. It is credible to normalized the ratio at time = 360 sec because the torsion field generator was turned on at time = 390 sec. In general, the profile of normalized EZ expansion ratios were similar to the control group except a little different of expansion ratios and variations. Once the ordered-structure water was build up, the region seems not to collapse even the generator was turned off after 2190 sec. The statistic results of final expansion ratio under the L- and R-torsion field illumination were 1.08 and 1.12, respective. Comparing with the expansion ratio of control group in Fig. 2(c), the EZ region would be enlarged by both left-handed and right-handed torsion field illumination. Different from the control group, the L-generator and R-generator were illuminated from one side of Nafion and the torsion fields were



**Figure 5.** The expansion ratio of (a) left-handed torsion field and (b) right-handed torsion field irradiation at time = 2400 sec in different criteria of EZ width. The red dots and red line in (a) and (b) were the expansion ratio in each experiment and the linear least square fit, respectively in A side. The opposite side (B-side) was labeled by blue dots and blue line.

leaving from the other side. To quantify the effect of R- and L-torsion fields in each experiment, the coming side and leaving side of torsion field were named as A-side and B-side, respectively. The relation of final expansion ratio (time = 2400 sec) and the criteria EZ width was discussed to investigate the torsion field effect on structured water. The different chiral configurations, left-handed and right-handed, torsion fields treated final EZ expansion ratio were shown in Fig. 5(a) and Fig. 5(b), respectively. The black line in Fig. 5(a) and Fig. 5(b) was extracted from single side of control group as a reference, the red line and blue

line were linear LS fit of A-side and the B-side, respectively. In Fig. 5(a), the least square fitting curve of EZ water expansion ratio in both side were indicated that when the criteria of EZ width ( $EZ_{width@360sec}$ ) between 350  $\mu m$  and 500  $\mu m$ , the stronger interaction of left-handed torsion field and EZ width was demonstrated, especially in A-side. The EZ water is a good absorber of torsion field represented in the tendency of linear fit, which were slight go down when the criteria of EZ width became thicker in A- and B-side, this results was similar with the previous work.<sup>15</sup> The EZ water could be regarded as a container of torsion field which was revealed in the trend of A- and B-side EZ expansion ratio after the left-handed torsion field illumination. The larger expansion ratio by adopting smaller range of EZ width was attributed to the fact that the torsion field was absorbed by lesser EZ water (thinner), and on the opposite side, the expansion ratio almost maintained like original properties or even a little bit smaller was due to the separation of L-torsion field by the thicker EZ water plus the fluctuation of discrete statistical data. Overall the effect of left-handed torsion field was stronger in A-side (field coming side) of EZ water when the criteria width ( $EZ_{width@360sec}$ ) between 350  $\mu m$  to 500  $\mu m$ , and the average EZ water expansion ratio of control group, L-TF treated A-side and L-TF treated B-side were 11.9%, 16.2% and 12.9%, respectively. The effect of right-handed torsion field generator was shown in Fig. 5(b). It is clearly that, the EZ water was interact with the R-torsion field stronger than L-torsion, especially in A-side. When the criteria width ( $EZ_{width@360sec}$ ) between 350  $\mu m$  and 600  $\mu m$ , the average expansion ratio of control group, R-TF treated A-side and R-TF treated B-side were 15.1%, 21.4% and 17.3%, respectively. Compare with the L-torsion field treated EZ water, R-torsion field further promote the EZ water build up ability which was indicated on the larger final expansion ratio by statistics and linear least fit.

When the torsion field was illuminating, the interaction or resonance of the field and bulk water help more H<sub>2</sub>O molecular splitting in to negative ion (OH) and positive ion (H). Then the more negative ions constitute the larger building block of the EZ in the vicinity of Nafion membrane, it is why the enlarge efficiency was higher in the smaller EZ

width ( $EZ_{width@360sec}$ ). Owing to the penetration depth of torsion field on the EZ water, the field response was more obvious in the thinner EZ width. From the experimental results, it could ensure that the EZ water has different structured degree with different refractive index and the EZ growth rate could be influenced by the torsion fields. Another noticeable fact is that, both of L-torsion field and R-torsion field enlarge the EZ region no matter A-side or B-side when the EZ width is within a proper range. The effect of chiral configurations in L-polarized and R-polarized torsion fields were different on the EZ water by the expansion ratio and tendency in A-side and B-side. The effective working range of left-handed and right-handed torsion field treated EZ water was 350  $\mu\text{m}$  to 500  $\mu\text{m}$  and 350  $\mu\text{m}$  to 600  $\mu\text{m}$ , respectively. The right-handed torsion field treatment lead to stronger interaction in the A-side EZ water, the final enlargement ratio was up to 21.4% when the EZ width is between 350  $\mu\text{m}$  and 600  $\mu\text{m}$ . Incidentally, a method was developed to detect torsion field, which have been ignored by mainstream science for a long time.

## Conclusions

In summary, the building up process of water exclusion zone was studied by the observation from side view. The exclusion zone of water was affected by left-handed and right-handed torsion fields, which were generated by polarized nickel-manganese ferrite through the spin-spin contact interaction. The left-handed and right-handed torsion field enlarge the A-side EZ water more significantly than that of the B-side EZ water. A preliminary method to detect torsion field have been demonstrated. The effective detection range (EZ width at time = 360 sec) for left-handed torsion field and right-handed torsion field were 350 - 500  $\mu\text{m}$  and 350 - 600  $\mu\text{m}$ , respectively. Through the enlargement of EZ water, the spin field indeed plays a role on water molecular reconstruction. This work may play a potential role for future study on the quantum biology.

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