Use of a Turn Coil and Channel Above a GMR-SV Device to Observe and Measure the Properties of Deoxidized Red Blood Cells Coupled to Magnetic Beads

Jong-Gu Choi, Su-Hee Kim, and Sang-Suk Lee

Abstract

A giant magnetoresistance-spin valve (GMR-SV) having highly sensitive magnetic properties was prepared with a multilayer soft ferromagnetic/non-magnetic/pinned ferromagnetic/anti-ferromagnetic iridium manganese (IrMn) top layer (NiFe/CoFe/Cu/CoFe/IrMn) structure. Micron-sized magnetic beads (MBs) with a diameter of 1 μm immersed in a solution containing 50 mg of Co-Si-OH/ml were attached to red blood cells (RBCs) having the very low magnetization of hemoglobin (HEM) to detect a deoxidized RBC combined with MBs (RBC+MBs). The RBC+MB combinations can be captured on a single-turn μ-coil that maintains a magnetic field large enough to detect the MBs attached to the RBC. When the RBC+MBs passes through a PR μ-channel with a width of 10 μm, its stopping and starting are controlled by the electrical AC input signal applied to the single-turn μ-coil. The RBC+MBs combinations captured above the μ-device of the GMR-SV change the output signal and thus, can be used for detection. This observation implies that this device can be used as a biosensor to analyze the coupling force between the HEM and the MBs for RBCs with deformed membranes passing through a narrow capillary.

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1 Introduction

Hemoglobin (HEM) having an iron (Fe) is the protein in red blood cells (RBCs) that carries oxygen. HEM has the molecular formula $C_{3032}H_{4816}O_{872}N_{780}S_8Fe_4$ [1, 2]. Animals obtain oxygen through their lungs or gills, and that oxygen is leaved HEM of the blood when it reaches the terminal tissue and capillary. In HEM, 218 oxygen atoms combine with one atom of iron, and 1 μl of blood in vertebrates can contain approximately 4–5 × 10^6 blood cells; 2.8 million HEMs being found in a single RBC is not uncommon [3, 4].

This study observed the movements of RBCs combined with magnetic beads (MBs) (RBCs + MBs) in vitro status under a stereoscopic optical microscope. One physical property of RBCs+MBs is that the motion has magnetic characteristics. In this research, a nano-magnetic-field-sensing device, a giant magnetoresistance-spin valve (GMR-SV), was used to observe the motion of a RBC combined with micron-sized MBs in detail so as to further research and development efforts on DNA (deoxyribonucleic acid) chips [5, 6].

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The optical images of the motions of the RBCs combined with MBs were observed by using a metallurgical microscope [7]. The sensitivity of the MBs to an external magnetic field and the area limits of the fine lithography process could be used to change the magnetic resistance properties of the MBs. When MBs coupled to a RBC were trapped, a magnetic signal was detected [8]. Based on their basic physical and magnetic properties, we postulate the possibility of future observations of motility in humans, as well as biological molecules other than HEM. The MBs could effectively be applied as bio-magnetic labels having the dynamic range of biosensor assays [5].

2 GMR-SV Device and RBCs Combined with MBs

The GMR-SV film was prepared by using a dc magnetron sputtering deposition system as a Ta (5 nm)/NiFe (10 nm)/CoFe (5 nm)/Cu (2.5 nm)/CoFe (5 nm)/IrMn (8 nm)/Ta (5 nm) multilayer, in which the IrMn layer was antiferromagnetic and served as a pinned layer, and the NiFe layer was ferromagnetic and served as a free layer; the Cu layer was a spacer. The deposition rates of the Ta, NiFe, CoFe, Cu, CoFe, and IrMn thin films were 0.1 nm/s, 0.15 nm/s, 0.15 nm/s, 0.12 nm/s, 0.15 nm/s, and 0.1 nm/s, respectively.

In the manufacture of the GMR-SV device, a lithography process with electron cyclotron resonance (ECR) Ar-ion milling was used to etch a multilayer thin film so that it could be used as the patterned μ-device, with a size of $1 \times 18\ \mu m^2$, of the GMR-SV as shown in Fig. 1a. After the milling, the photo resistance (PR) was eliminated, and through another lithography process, a 2-probe Cr/Cu electrode with a width of 40 μm was formed by using the lift-off method, as shown in Fig. 1b. To observe the MR properties of the prepared sample, we determined the coercive force, the exchange coupling field (H_{ex}), and the MR ratio value by using the MR curves, which were measured with a two-probe MR measurement system at room temperature. The μ-turn coil was fabricated by using a Cu layer with a thickness of 50 nm and a width of 4 μm, as shown in Fig. 1c. To avoid the leakage effect, a thin (100-nm-thick) insulating film of SiO_2 above the μ-device of the GMR-SV was coated below the single-turn μ-coil [9]. We re-deposited the Cu paths of the μ-coil that were to be placed in the central part of the GMR-SV.

After the Cu layer of the μ-coil, which had been evenly deposited on the center of the GMR-SV device, had been wound once, the electrode of the coil was placed in a location different from that of the GMR-SV electrode. Figure 1d presents the composite structure for the molecular device used to control the flow of a biomolecule with RBCs on a micron scale. The final lithography process to prepare the PR μ-channel with a thickness of 8 μm and a width of 10 μm was performed on the central part of the μ-device of the GMR-SV and a single turn μ-coil.

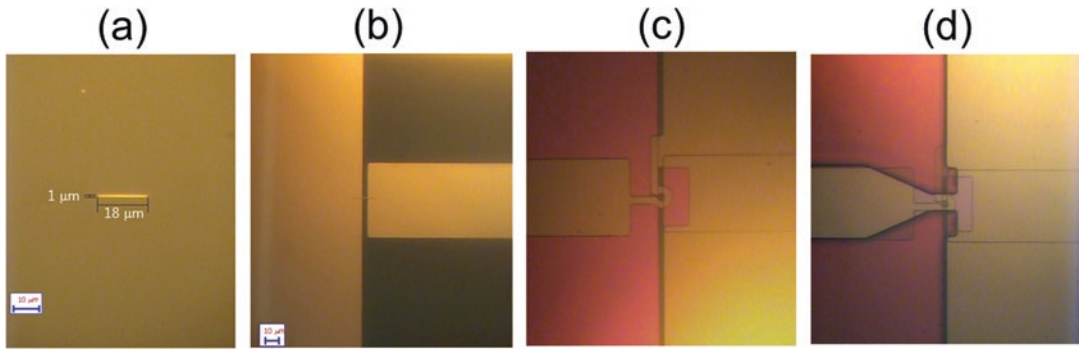

Fig. 1 Patterns for (**a**) the μ-device of the GMR-SV, (**b**) the 2-probe Cr/Cu electrode, (**c**) the single-turn μ-coil, and (**d**) the complex structure composed of the μ-coil, the μ-device of the GMR-SV, and the PR μ-channel

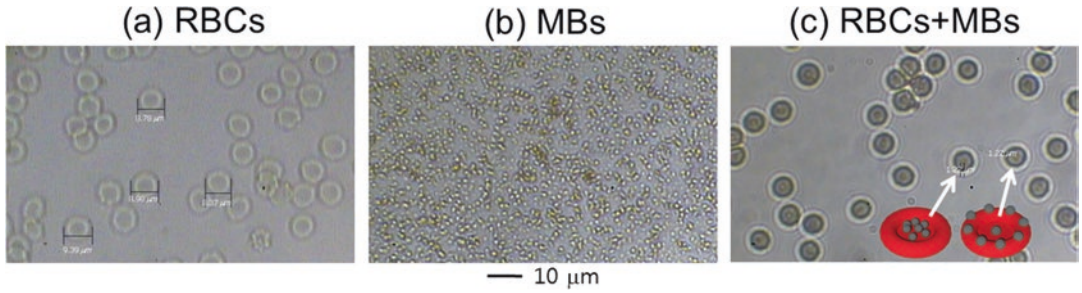

Fig. 2 Images of (**a**) human RBCs with diameters of about 8–9 μm in a solution of PBS, (**b**) MBs with diameters of about 1 μm in a solution of 50 mg of Co-Si-OH/ml, and (**c**) RBCs combined with MBs in such a way that 7–9 MBs were positioned in the center part or at the edge part of one RBC by organic chemical binding

We separated RBCs from human blood by using centrifugation, and an optical image of the results is shown in Fig. 2a. The dilution of RBCs in saline (phosphate buffer solution (PBS)) solution was 1.0%. The density of RBCs was reduced to allow easy capture, and a single RBC could be observed by using a micro capillary tube. The diameter of the capillary tube, which had been made by melting a glass tube of a few mm, used to capture pure RBCs was a few μm. The RBCs inside the capillary tube were dropped above the GMR-SV device. MBs with diameters of 1 μm in a solution containing 50 mg of (Co-Si-OH)/ml, as shown in Fig. 2b, usually exhibit superparamagnetic magnetization because when they lose their magnetic fields, those fields return in seconds. The MBs, which were to be magnetized at about 1 kOe (= 80 kA/m) for 2 h, were placed on a permanent neodymium-boron magnet. For adhesion, the RBCs were combined with MBs, in such a way that 7–9 MBs were positioned in the center part or at the edge part of one RBC by organic chemical binding, as shown in Fig. 2c. MBs coupled to the RBCs are seen to behave differently when an external magnetic field is applied [7].

3 Observation and Detection of Deoxidized RBC+MBs

The RBCs combined with MBs (RBCs+MBs) in normal saline are free to move along the path of the flowing liquid, as shown in Fig. 3a, b. A virtual cross section of the μ-channel with RBCs+MBs floating in saline is shown in Fig. 3c. Figure 3a, b present side views for two different flows of RBCs+MBs in saline over a complex structure composed of the single-turn μ-coil, the PR μ-channel, and the μ-device of the GMR-SV. The current shunting effect was avoided by depositing a passive insulating SiO_2 layer after the fabrication of each layer. After having passed through the μ-channel, the RBC+MBs was located in the center of the GMR-SV device between the two Cu electrodes.

The dilute saline solution with combinations of a RBC+MBs flows over a complex structure composed of the single-turn μ-coil, the PR μ-channel, and the μ-device of the GMR-SV due to a pressure difference induced by using a micro syringe. Those combinations then exit through to a valley in the 8-μm-thick, 10-μm-wide PR, as shown in Fig. 3c. Figure 4a–c show one of the RBC + MB combinations moving from (a) → (b) → (c) in time. The positions (a) and (c) are the left and the right positions, respectively, prior to the passage of the RBC+MB combination to the center of the μ-device of the GMR-SV.

Especially, at position (b), as shown in Fig. 4b, the RBC + MB combination is captured due to the inductive electromotive force caused by the AC input signal applied to the single-turn μ-coil. The flow mechanism depends on the fact that the deformation of the RBC's membrane can be controlled by using the flow velocity of the RBC + MBs. That is, when compared to the uniform feature and the flow velocity of a pancake-like normal RBC, those of a RBC with a deformed membrane are different due to the inductive electromotive forces being different.

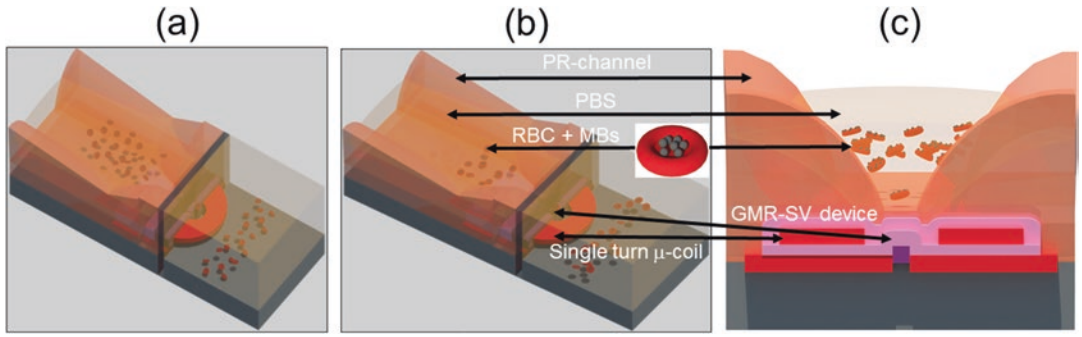

Fig. 3 (**a**) The initial status and (**b**) the final status to separate two different flows of RBCs+MBs immersed in saline (PBS) over a complex structure. (**c**) A virtual cross section of the μ-channel with RBCs+MBs floating in PBS

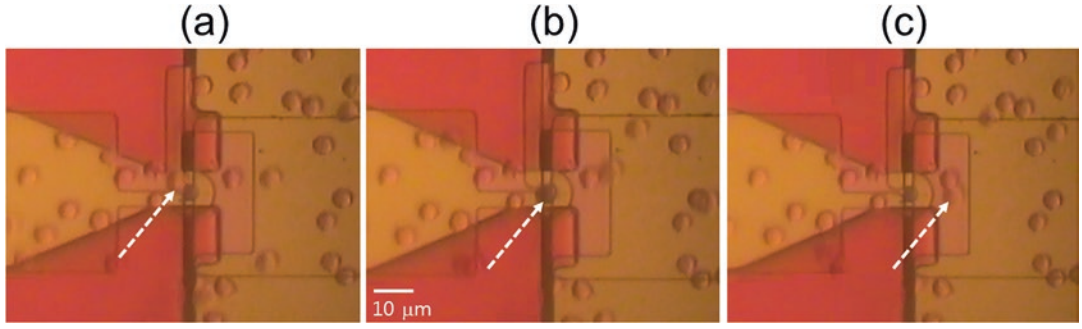

Fig. 4 Three visualization of the flow of RBCs+MBs passing through the PR μ-channel above the GMR-SV device: (**a**) left side, (**b**) center (stop status), and (**c**) right side of GMR-SV device. The dotted arrows are marked at each position of a single RBC+MBs at three different positions

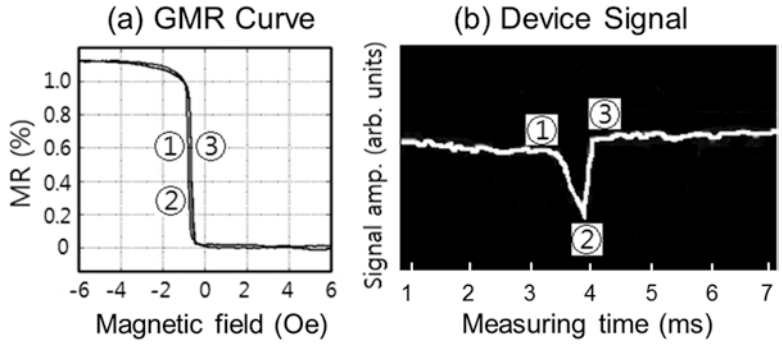

Fig. 5 (**a**) Minor MR curve of the GMR-SV device and (**b**) change in the output signal's amplitude with time [ms] when a RBC+MBs passes to the singe-turn μ-coil. The output signals (1), (2), and (3) of (**a**) and (**b**) correspond to before passing through, while stopping in, and after having passed through the center of the GMR-SV device

Figure 5a shows that a slight magnetic sensitivity (MS) of 1.04%/Oe at 0 Oe and an exchange coupling field (H_{ex}) of 0.80 Oe (1 Oe = 79.578 A/m) were measured from the MR curve. When a RBC+MBs passed to the singe-turn μ-coil, as shown in Fig. 4, the magnetoresistance of the GMR-SV element was kept at a constant value before the bound RBC+MBs was dropped. The output signals for the notations (1), (2), and (3) in Fig. 5b are the same as those of Fig. 5a and cor-

respond to before passing through, while stopping in, and after having passed through the center of the GMR-SV device.

The changes in the signals were caused by the change in the sensing position on the minor MR curve due to an abrupt variation in the sensing field in the presence of the RBC+MBs. The change in the MR in response to the presence of the RBC+MBs at 0 Oe was an increase of 0.4%. The detection of a deoxidized RBC+MBs passing through a single μ-coil and a PR μ-channel can be analyzed by using the output MR signal of the GMR-SV device. Therefore, detailed studies of hemoglobin are needed if it is to be used to determine those cases in which magnetic therapy treatments would be beneficial in the medical field.

4 Conclusions

To observe and measure the properties of deoxidized RBCs coupled to MBs, we used a turn coil and channel above a μ-device of a GMR-SV device fabricated by using photo-lithography, ECR Ar-ion milling, and a lift-off electrode process. A RBC+MBs can be captured on any single turn μ-coil that maintains a large enough magnetic field to detect the MBs attached to the RBC. When a RBC+MBs passes through a 10-μm channel, two states, no motion and motion, occur and can be controlled by using the electrical AC input signal applied to the single-turn μ-coil. The RBC captured above the GMR-SV device causes a change in the output signal that can be used for detection. This observation implies that this device can be used as a biosensor to analyze the coupling force between HEM and MBs and to identify any deformed features of the RBCs passing through a narrow capillary. Also, the process with a μ-device consisting of a GMR-SV, a single-turn μ-coil, and a μ-channel can be applied to analyze the properties of the deformed membrane of a RBC coupled to MBs.

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