

# A Facile Template-Free Approach to Magnetodriven, Multifunctional Artificial Cilia

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**ABSTRACT** Flexible and magnetic artificial cilia were grown on various substrates by a facile bottom-up approach based on template-free magnetic assembly. The magnetic cilia formed spontaneously from a suspension of micrometer-sized ferromagnetic particles and elastomeric polymer in a liquid solvent when dried in an external magnetic field. The cilia mimics were mechanically stable even in the absence of an external magnetic field and a solvent due to the polymer, which acted as “glue” holding the particles together and connecting the cilia to the substrate. The length of the magnetic cilia was in the millimeter range, that is, two to three orders of magnitude times the length of typical biological cilia. The aspect ratio reached values over 100 and was tunable with the magnetic field gradient and the size of the ferromagnetic particles. The cilia mimics responded to an external magnetic field by reversibly bending along the field. The bending actuation was sufficiently powerful to allow two functions: to translate macroscopic nonmagnetic objects placed over the cilia mimics and to mix liquids of even high viscosity. The mechanical properties of the magnetic cilia could be easily tuned by changing the impregnating polymer. The particularly simple template-free construction and fixation on various surfaces suggest applications as an externally controllable surface.

**KEYWORDS:** cilia • biomimetics • magnetic particles • actuator • mixer

## INTRODUCTION

Static topographically patterned surfaces have attracted considerable interest over recent years because they allow various functionalities (1). For example, arrays of cylindrical pores made by using block copolymer templates have been filled with magnetic material to make memory elements (2). Arrays of micrometer-sized pillars or vertical nanofibers have been constructed by using a multitude of approaches to allow various functionalities (1, 3–6). For example, inspired by water-repellent biological surfaces, man-made surfaces with properly treated vertical pillars, ridges, or nanofibers have been shown to have high contact angles for water and even for oils (7–12). Furthermore, vertical surface topographies such as carbon nanotube forests have been constructed to mimic biological adhesion (13–16). Topographical patterns are relevant also in tissue engineering (1, 17).

Beyond static topographical patterns, dynamic topographical patterns have been pursued very recently to allow for externally controllable surfaces. For example, mesoscale rods have been partly embedded in a thermoreversible polymer layer to make temperature-responsive surfaces for aqueous and humid conditions (18). Self-assembled mag-

netic needles composed of ferromagnetic particles on a substrate patterned with magnetic regions have been used in microfluidic channels as mixers (19). In addition to man-made surfaces, dynamic topographical patterns are ubiquitous also in nature; cilia and flagellae that cover protozoans and epithelial cells in biological organisms can make the organism motile, transport cargo on their surface, and sense external stimuli (20–22). The cilia and flagellae are very thin, in the range of 0.25  $\mu\text{m}$ , and their length can vary from several micrometers (cilia) (21) to even millimeters (flagellae) (23). A cilium can be considered to be a molecular motor driven by adenosine triphosphate (ATP). Microtubules, which are a part of the cilium, slide past each other because of the moment of the motor proteins, dyneins, which convert chemical energy of ATP into mechanical movement, resulting in bending of the cilium. Utilization of cilia for material transport in technological applications is attractive, but the complexity of these biological structures poses challenges. This has launched various approaches toward cilia-mimetic materials with simpler constructions and more practical actuation principles. For example, optically controlled cilia mimics have been recently demonstrated (24). In addition, the assembly of magnetic cilia mimics of less than 100  $\mu\text{m}$  in length with magnetically driven actuation capabilities has been demonstrated by using magnetic nanoparticles in template-based (25, 26) and template-free approaches (27). In one approach, a composite material consisting of ferrofluid and poly(dimethylsiloxane) was templated by polycarbonate track-etched membranes into rod arrays (25). In another approach, polystyrene beads were covered with

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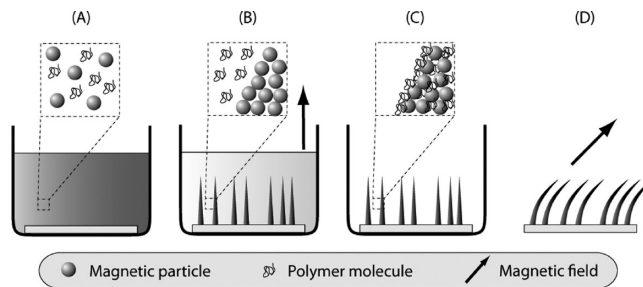
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magnetic nanoparticles, subsequently assembled into chains in an external magnetic field, and attached on a glass substrate by amidation chemistry (26). In a third template-free approach, cobalt nanoparticles coated with polymer brushes were shown to form cilia-mimetic chains connected with physical forces under an external magnetic field (27). The cilia mimics made by the latter two approaches are mechanically stable only in liquid medium, while those made by the first approach do not need a liquid medium. The new fabrication method presented in this paper combines some of the best features of these earlier approaches, namely, easy template-free synthesis and stability in the absence of a liquid medium. Furthermore, the cilia mimics made by the new approach can reach a length scale of 10  $\mu\text{m}$ , i.e., 2 orders of magnitude more than those made by the previous approaches.

Our magnetically driven cilia-like structures mimic biological cilia by their filamentous shape and actuating functionality. Similar to biological cilia where the dynein motors are located inside the cilia and the energy for the motion is supplied from the outside in the form of ATP, in the magnetic cilia mimics, the “motors”, i.e., the magnetic particles, are located inside the cilia, while the energy is supplied from outside via a magnetic field. The fabrication principle is closely related to the well-known magnetic-field-induced instability and the resulting spontaneous surface deformation of ferrofluids (28). In fact, a similar effect has been observed in films of colloidal magnetic nanocrystals deposited from a solution in a magnetic field (29) and in dielectric polymer films in an electric field (30). In all three cases, the unfavorable surface charge of the polarized film, either electric or magnetic, is reduced by undulation of the film surface or, in extreme cases, by division of the film into individual cylinders along the polarizing field. The induced structures can be periodic and exhibit hexagonal symmetry. An ideal ferrofluid with surface undulation relaxes back to a flat film when the polarizing external field is removed. The prerequisite is that the magnetic particles are not aggregated during the structure formation. In contrast to ferrofluids, films of magnetorheological fluids do not form well-defined structures in a polarizing magnetic field but instead large irregular solidlike protrusions. It is known that a dilute suspension of micrometer-sized ferromagnetic particles, which can be considered to be a dilute magnetorheological fluid, phase-separates in an external magnetic field into a liquid phase and a solid particle phase. We observed that when phase separation is done correctly, the particle phase is composed of individual conical structures that resemble the biological cilia by their aspect ratio. By adding a small amount of elastomeric polymer into the suspension before phase separation, we were able to make cilia mimics that are mechanically strong and flexible even after the carrier fluid is removed and the magnetic field is switched off. This extremely facile route gives cilia mimics that are convenient to handle. The orientation of the magnetic cilia mimics is controllable with an external magnetic field, and the resulting actuation force is capable of moving nonmagnetic ma-



**FIGURE 1.** Schematic illustration of the structure formation in a dilute magnetic suspension: (A) the initial mixed state (no magnetic field); (B) the phase-separated state (magnetic field perpendicular to the substrate); (C) the solvent removed state (no magnetic field); (D) the functional state (magnetic field in an arbitrary angle).

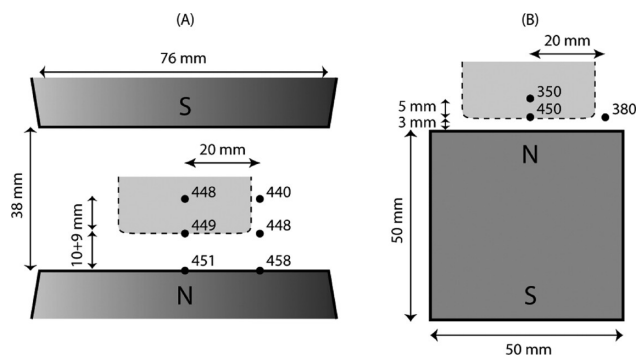
terial and mixing viscous fluids. Because of their robustness, our cilia mimics can find applications in microfluidics, in which cilia-like structures are currently being investigated for liquid mixing and pumping (31–33).

## EXPERIMENTAL METHODS

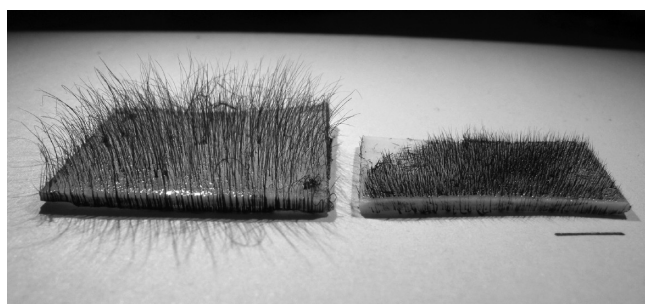
A magnetic suspension was prepared by mixing 20 mg of cobalt particles (average diameter 800 nm; OM Group, Kokkola, Finland) and 8 mL of toluene containing 30 mg of dissolved elastomeric poly(styrene-*block*-isoprene-*block*-styrene) (Kraton D1113BT). The suspension was vigorously ultrasonicated for 5 min with a Branson S-450 sonifier with a microtip and amplitude of 30% and poured into a 35-mm-wide poly(tetrafluoroethylene) (PTFE) dish with a flat substrate placed on the bottom (Figure 1A). The suspensions prepared in this way began to show aggregation approximately 10 s after ultrasonication, and for that reason the dish with the suspension was quickly placed in the magnetic field. When in the magnetic field, the particles instantaneously formed conical high-aspect-ratio structures, while the polymer molecules remained dissolved (Figure 1B). Toluene was allowed to evaporate completely at room temperature while keeping the dish in the magnetic field. During evaporation, the elastomeric polymer molecules covered the conical structures and penetrated the voids between the particles (Figure 1C). After evaporation, the cones were mechanically stable and bendable by a mechanical force or by reapplication of the magnetic field nonperpendicularly to the substrate (Figure 1D). These final structures were called magnetic cilia mimics to distinguish them from the corresponding mechanically unstable phase-separated structures that appeared if no polymer was used for impregnation. Glass, PTFE, and silicon substrates were used with no difference observed in the formed structures.

## RESULTS

The magnetic field gradient played an important role in the structure formation during phase separation. Its effect was studied by using two magnets to create a 450 mT vertical magnetic field with different vertical gradients on the substrate surface: an electromagnet with a circular-pole cross section (GMW 5403) and a permanent magnet with a square cross section (NdFeB, Neorem Magnets 495a). There was only a small field gradient along the axis of the electromagnet from the magnet center toward the poles (approximately 0.1 T/m; Figure 2A). More important was the transverse gradient (0.4 T/m), which in the central plane of the electromagnet (equal distances from the both poles) pointed toward the center and at the pole surfaces from the



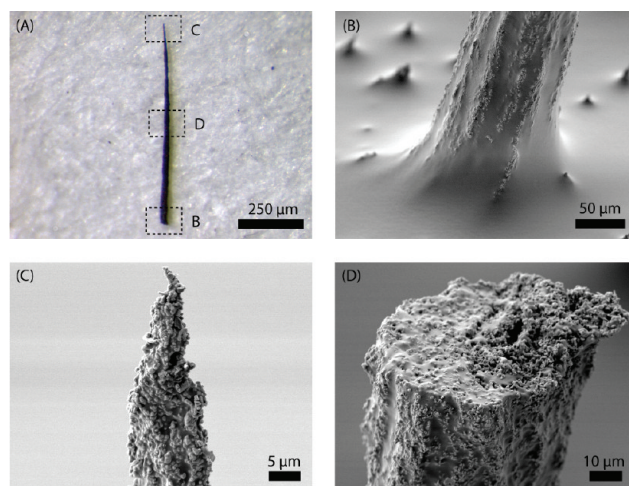
**FIGURE 2.** Illustrations of magnetic field configurations used in the experiment. Dots and adjacent numbers represent the measured vertical magnetic field strength (in mT). The light-gray area denotes the volume occupied by the magnetic suspension, and the horizontal dashed line indicates the location of the substrate: (A) a relatively homogeneous magnetic field created by the electromagnet with circular poles; (B) a strongly inhomogeneous field created by the cubic NdFeB magnet. Notice that the third dimension of the permanent magnet is the same as the other two and that the magnetic suspension is placed in the center of the upper square plane of the magnet.



**FIGURE 3.** Photograph of two PTFE substrates covered with magnetic cilia mimics made in a magnetic field of 450 mT with a vertical gradient of 0.1 T/m using the electromagnet (left) and with a vertical gradient of 20 T/m using the permanent magnet (right). The scale bar is 5 mm.

center toward the pole edge. Approximately in the middle between the central plane and the pole planes was a region in which the transverse magnetic field gradient was negligible. If the substrate was placed on this plane, growth of the cilia mimics took place equally at all distances from the central axis of the magnet. On the other hand, if the substrate was placed on the lower pole, the transverse component pulled all of the formed structures to the edges of the dish, and if placed in the central plane, the structures migrated toward the axis of the magnet. In the case of the permanent magnet (Figure 2B), there was both a large vertical gradient (20 T/m) toward the magnet surface and a smaller gradient toward the central axis of the magnet (4 T/m). Being dominated by the vertical gradient, the magnetic structures did not show a tendency to migrate toward the central axis.

The length and aspect ratio of the magnetic cilia depended strongly on the vertical magnetic field gradient. The cilia mimics made under a small vertical gradient of 0.1 T/m (electromagnet) were approximately 6 mm long and had an aspect ratio of 120 on average (Figure 3, left). In contrast, under a large vertical gradient of 20 T/m (permanent magnet), they became only 1 mm long and had an aspect ratio

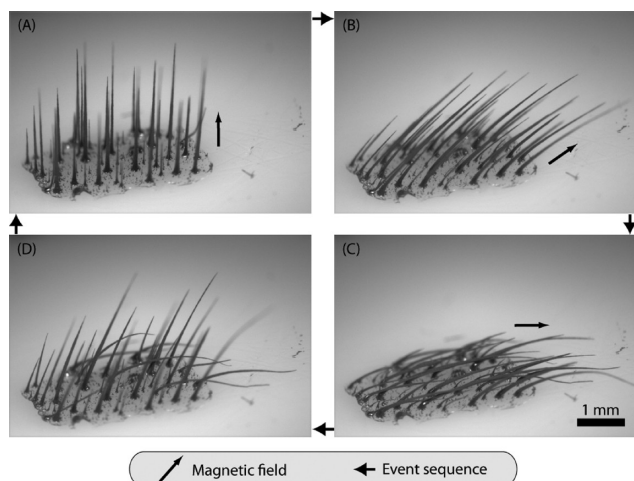


**FIGURE 4.** (A) Optical micrograph of a typical single magnetic cilium with an aspect ratio of 28. Marked areas (B–D) are shown magnified in the scanning electron microscope images: (B) the cilium base; (C) the cilium tip; (D) the cilium cross section.

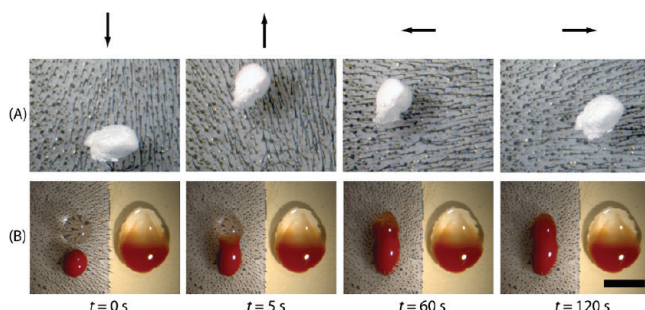
of 20 (Figure 3, right). These aspect ratios are of the same order of magnitude as those in typical biological cilia, but the cilia mimics are approximately 2 orders of magnitude longer. The number density of the cilia measured optically was  $400 \text{ cm}^{-2}$ . The magnetic cilia did not show periodic spatial ordering. The elastomer was present on the surfaces of the cilia (Figure 4B,C), in the voids between the ferromagnetic particles (Figure 4D), and on the substrate as a flat film. The cilia mimics were strongly attached to the substrate by the polymer. Thermogravimetric analysis (TGA) indicated that the cobalt content of the cilia mimics was 79 % of the mass or 30 % of the volume (see the Supporting Information).

Both the flexibility of the cilia mimics and their response to the reapplied magnetic field improved as the aspect ratio increased. Tensile tests indicated that the elastic modulus of the cilia mimics was 2600 MPa, from which a bending stiffness of approximately  $40 \times 10^{-12} \text{ N m}^2$  was deduced for the high-aspect-ratio cilia. Even the low-aspect-ratio cilia mimics could be bent the complete range of  $90^\circ$  by applying a transverse magnetic field. To demonstrate this, we isolated a region of  $5 \text{ mm}^2$  containing magnetic cilia with an aspect ratio of 20 by removing all of the surrounding cilia mimics by cutting them off with a blade (Figure 5A). Initially, the magnetic field was applied normally to the substrate and the cilia mimics were totally upright. Gradual tilting of the magnetic field toward the substrate plane resulted in bending of the cilia mimics (Figure 5B,C). If the field was suddenly removed, the cilia mimics slowly relaxed toward the vertical orientation. The relaxation was never complete, and the magnetic cilia mimics remained in an intermediate stage (Figure 5D) between the vertical and tilted orientations until a vertical magnetic field was reapplied to erect the cilia mimics once again (Figure 5A). See also the Supporting Information for a video demonstration of the bending of the high-aspect-ratio cilia mimics.

Because the cilia mimics could be reversibly bent in any direction by changing the direction of the external magnetic



**FIGURE 5.** Magnetic-field-induced bending of the cilia mimics made in a 450 mT magnetic field with a 20 T/m vertical gradient. The magnetic field is (A) perpendicular, (B) at a 45° angle, and (C) parallel to the substrate surface. (D) After removal of the magnetic field, the cilia mimics relaxed toward the surface normally. By reapplication of the perpendicular field, the cilia mimics erected completely once again.



**FIGURE 6.** Optical micrograph sequences demonstrating actuation and mixing with the magnetic cilia. (A) Magnetic cilia as an actuator inducing movement to a 3 mm piece of expanded polystyrene. Arrows indicate the magnetic field direction. (B) Mixing of two drops of glycerol (one clear and one stained red) by periodic actuation of the magnetic cilia mimics at a rate of 1–2 beats/s (left) compared to diffusive mixing (right) at different time intervals. The scale bar is 5 mm. Videos are available in the Supporting Information.

field, the cilia mimics can be used to create an alternating actuating force that can act on materials and objects placed over the magnetic cilia. Note that one of the functions of biological cilia is to transport cargo. We demonstrated that the cilia mimics can be used to induce simple periodic motion in nonmagnetic objects by placing a piece of expanded polystyrene over the cilia mimics and by bending the cilia in a periodic fashion using a magnet placed under the substrate (Figure 6A). Controlled transport of material over larger distances would probably need the development of peristaltic progressing wavelike bending cycles of the cilia mimics. However, not all actuation processes require peristaltic motion: the mixing of two liquids can be realized by a simple back-and-forth motion of the cilia mimics, which is directly relevant for mixing on surfaces in miniaturized structures. The mixing capability of the cilia mimics was demonstrated for two viscous drops of glycerol that were placed on the magnetic cilia (Figure 6B). We selected a viscous liquid in order to demonstrate the strength of the magnetic cilia mimics as mixers. The time required for

mixing with the cilia mimics was significantly shorter than the time required for diffusive mixing. See also videos of actuation and mixing that are available in the Supporting Information.

## DISCUSSION

Magnetic-field-induced structure formation in dilute magnetic suspensions is both similar and dissimilar to its analogues observed in ferrofluids. Ferrofluids are composed of at least two phases that form a stable colloidal dispersion that will not phase-separate even in a magnetic field. In contrast to ferrofluids, dilute suspensions of micrometer-sized ferromagnetic particles are two-phase dispersions that slowly phase-separate by sedimentation even in the absence of a magnetic field. This phase separation is greatly enhanced in a magnetic field and even more if the field is inhomogeneous. Because of the tendency to phase-separate, kinetics plays a crucial role in the structure formation. If the suspension is allowed some time to develop particle aggregates prior to application of the magnetic field, the obtained phase-separated structures are different from those obtained from a nonaggregated suspension. In general, the regularity of the structures, in terms of length, diameter, and spacing, is decreased and cannot be improved, for example, by annealing because the strong physical interaction between particles essentially works as a kinetic trap; once the particles come into contact with each other, they can be separated only by agitation much stronger than random thermal fluctuations.

If the kinetic effects are not taken into account, the equilibrium geometry of the cilia mimics is determined by the magnetostatic interaction between the cilia mimics (favoring their spatial separation), the demagnetizing effect of individual cilia (favoring a high aspect ratio), and the interaction of the cilia mimics with the external magnetic field (favoring accumulation of the particles to the regions where the external field is strongest). The differences in the length and aspect ratio of the cilia mimics made in the homogeneous and inhomogeneous fields observed in this work are manifestations of the interaction between the ferromagnetic particles and the external field. The interaction results in a stronger accumulation of the particles toward the substrate in the case of the inhomogeneous field compared to the homogeneous field, decreasing the length and aspect ratio of the cilia mimics. In a similar fashion, the final magnetic cilia mimics bend in an external magnetic field because it leads to a decrease in the magnetic interaction energy between the particles and the external field and between the particles themselves. The counteracting elastic force comes from the polymer, resisting deviation from the original vertical alignment of the cilia mimics. The energy balance of magnetic cilialike structures has been discussed in depth elsewhere (25).

Controlling the length, aspect ratio, and density of the cilia mimics can be important for applications. We have tuned the aspect ratio by the magnetic field gradient and also by the size of the cobalt particles (see the Supporting Information). Particles with smaller diameter make cilia mimics with

lower aspect ratio. The length of the cilia mimics was shown to be tunable by the amount of magnetic particles used. However, independent of the particle size, field gradient, and amount of cobalt used, the number density of the cilia mimics remains approximately the same.

Our template-free method has the advantage of being general and applicable to various combinations of solvents, polymers, substrates, and magnetic particles. By selecting different polymers, one can tune the mechanical properties of the cilia mimics. For example, using a glassy polymer instead of an elastomer yields rigid cilia mimics that do not bend in a magnetic field. The main prerequisites for the method to work are that the polymer is soluble and that no major sedimentation of the particles takes place before magnetic field is applied.

## CONCLUSIONS

The formation of magnetic cilia mimics from a dilute suspension of micrometer-sized magnetic particles and polymer in an external magnetic field was demonstrated. The aspect ratio of the magnetic cilia mimics was found to be tunable with the magnetic field gradient and particle size. The magnetic cilia mimics were mechanically stable even in the absence of a carrier fluid and magnetic field. Furthermore, the mechanical properties could be tuned by selecting mechanically different polymers. The high-aspect-ratio cilia mimics were flexible and responded to the external magnetic field by reversibly bending along the field. Magnetic actuation and fluid mixing were demonstrated with the cilia mimics. The bottom-up fabrication method demonstrated here is very general and can be applied to various combinations of solvents, polymers, substrates, and magnetic particles. It is also a demonstration of direct bottom-up fabrication of a functional macroscopic (>1 mm) externally controlled device from micrometer- and submicrometer-sized constituents.

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**Supporting Information Available:** MPEG4 videos demonstrating bending of the high-aspect-ratio cilia mimics in a magnetic field and demonstrations of moving a piece of expanded polystyrene and mixing two drops of glycerol by movement of the low-aspect-ratio cilia mimics, and a PDF file containing the TGA curve of high-aspect-ratio cilia mimics, optical images of cilia mimics made of 800, 300, and 80 nm cobalt particles, and the stress–strain curve of a

single magnetic cilium. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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