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3D Printed Microtransporters: Compound Micromachines for Spatiotemporally Controlled Delivery of Therapeutic Agents

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Biological systems are exquisitely sensitive to the location, dose, and timing of physiologic cues and pharmaceuticals. This spatiotemporal sensitivity indicates that diagnostic and

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Supporting Information

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therapeutic approaches with minimal off-target effects can be particularly efficacious. Targeted delivery of drugs, genetic material, and cells increases the effectiveness of therapies while minimizing side effects.^[1] Several micro- and nano-particles have been developed to safely carry these therapeutic payloads.^[2,3] Systemic injection of these particles results in their dilution and only a small fraction of the particles reaches the treatment region. Magnetically active particles can be guided directly to the treatment region, which decreases the delivery of their payload to other undesirable locations.^[4] They can be magnetically agitated to promote mixing after delivery, which increases mass transport into the target tissues compared to passive diffusion.^[5,6] Unfortunately, controlled navigation of individual nanoagents to target sites within a complex biological system is challenging due to the agent's small size and weak magnetization. Furthermore, the immune system, hostile environmental conditions, and physical barriers can neutralize them. Thus, a targeted delivery method is required that not only can release its contents at the target site in a dose-dependent manner, but also contain and protect the payload during transport.

Untethered, miniaturized robotic devices can enable us to perform minimally invasive operations in three-dimensional, complex microenvironments.^[7,8] These remotely actuated operations can perturb or investigate biological systems in a flexible and on-demand manner. A variety of simple micromachines have recently been developed including microstructures controlled by oscillating magnetic fields,^[9-11] helical swimmers demonstrating corkscrew motion,^[12] thermally or magnetically actuated microgrippers,^[13-16] self-propelled micromotors,^[17] electrostatic^[18] and impact-driven microactuators.^[19] These machines can interact with objects both through physical contact and fluid flow generated around their body.^[20-25] The diagnostic and therapeutic potential of robotics in these micro-biological contexts can be greatly enhanced with the development of compound micromachines that have multiple mechanisms working together to perform complicated tasks, such as the transport and release of therapeutic agents. We call these devices microtransporters, and they have uses beyond targeted delivery. For example, they can noninvasively collect biological samples from remote pathological sites for diagnostic purposes. Stimuli responsive mobile microcapsules have been introduced to achieve similar goals,^[15,26] but their payload is released en masse and they have neither an active loading nor mixing mechanism.

Here, we report a microtransporter that can actively collect, encapsulate, transport and controllably release micro and nanoagents. The fabrication method is based on three dimensional (3D) direct laser writing and selective physical vapor deposition of magnetic materials (Figure 1a). The two-photon induced photopolymerization process enables the manufacturing of custom shaped microparts,^[27-34] which we use to create a compound micromachine without requiring further assembly.^[35-37] As the integrated mechanical system contains parts that must move relative to each other, it is important to generate magnetic actuation only at specific locations. This is achieved through selective coating of magnetic materials. After printing the main assembly, a 3D sacrificial structure is printed to protect the non-actuated components during Ni/Ti metal deposition (Figure 1a(II)). Figure 1b shows SEM images of the micromachines along with the protective sacrificial structures. After the deposition process, these structures are removed using a sharp probe controlled by a micromanipulator. Application of an external magnetic field facilitates the detachment of

the micromachines from the substrate by inducing an attraction between the probe and the screw. Introducing a Ni/Ti coating on the printed microstructures does not induce cytotoxicity to mammalian cells after days of exposure, and the cells readily adhere and proliferate over the devices.^[38] We applied an additional polyethylene glycol (PEG) coating on the outer surface of the micromachine to hide the agent from the host's immune system and minimize protein accumulation, thus making them more compatible with bodily fluids and the tissue microenvironment.

The microtransporters comprises two nested components, a magnetically actuated main shaft with integrated parts embedded inside a passive cylinder (Figure 1c). The individual parts of the microtransporter are shown in Figure 1d. The shaft is free to rotate, and its translation is constrained at both ends of the cylinder by the circular bushings. We tried to keep the diameter of the shaft as small as possible to maximize the empty volume in the cylinder but a very thin structure would bend due to the torque applied by the screw. The minimum diameter of the shaft that did not lead to deformation was 6 μm . Depending on the laser intensity and optics, the radius of the volume pixel vary from 0.5 μm to 1.5 μm . We picked an inner diameter of 14 μm for the bearing to allow constraint without seizing. The magnetic external screw pulls the micromachine through the surrounding liquid, much like a corkscrew in a cork, when rotated with an externally applied uniform magnetic field generated by Helmholtz coils. This corkscrew motion results in the translation of the whole device for targeted therapy (see Figure S1). Figure 1e shows the relative motion of the main shaft inside the cylinder. Thus, the positioning of the microtransporter and its shaft's rotation can be controlled together using external magnetic signals. This customizable fabrication process allowed several different designs to be explored. Apart from the microtransporter described above, we also engineered microcapsules with removable caps and microsyringes with sliding plungers (Figure 1f). The cap of the microcapsule can be repeatedly closed and opened, and the plunger of the microsyringe can be moved back and forth by reversing the magnetic field's rotation.

Individual microparticles can be trapped and transported using the microvortex generated by a rotating object.^[23-25] Although this does not extend well to multi-object transport and is very susceptible to environmental disturbances, it demonstrates that fluid vortices can be leveraged to transport particles without mechanical contact. Inspired by this mechanism, the microtransporter is fitted with an Archimedean screw-pump, created by engraving a second screw onto the main shaft (see Figure 1d). The rotating screw draws fluid and particles into the capsule while the piston slides forward to close the rear part of the capsule and trap the particles within (**Figure 2**). When actuated in reverse, the piston slides into the open configuration, and the trapped particles are gradually expelled with the reverse flow generated by the propeller. We engineered three microblades, forming an equilateral triangle in the front of the capsule, to form a sieve (see Figure 1d). The sieve, together with the apertures printed at the rear part of the capsule and on the piston (see Figure S2 and Supporting Information), function as filters to select particles within the desired size range while allowing the fluid flow through the body.

The schematic diagram in Figure 2a summarizes the remotely activated loading and releasing process (Supplementary Movie 1). The microparticles, drawn into the chamber by

the recirculating flow induced by the micropump (Figure 2b), stay inside the cylinder as the tail closed the rear side at this stage. As expected, the strongest pumping action is generated when the body is immobilized while driving the screws (Supplementary Movie 2). The screw is designed to rotate independently from the cylindrical capsule, but the capsule starts rotating sympathetically due to fluidic forces and part-to-part contacts during operation. On a planar surface, the undesired capsule rotation is limited due to non-specific surface adhesion. Further reduction of the undesired capsule rotation can be achieved by limiting the screw rotation to frequencies below 15Hz, because the fluidic coupling is proportional to the relative rotational speeds of the capsule and the shaft.

Monitoring the loading and unloading process is possible, because the capsules are optically transparent. Thus, the internal content can be visualized using brightfield or phase-contrast imaging. Figure 2c shows a loaded micromachine before, during, and after the release process. The machines were actuated with a rotating magnetic field of 9mT at several frequencies between 1Hz and 10Hz. The loading capacity was evaluated for two different polystyrene microspheres 3 μ m and 6 μ m in diameter (Figure 2d). The maximum numbers of beads that can be fit into these devices are 594 and 74 for 3 μ m and 6 μ m beads, respectively (see Supplementary Information). In practice, maximum packing efficiency is not achieved because the beads do not form a close-packed structure. We operated the microtransporter in the loading mode in a pool full of uniformly sized microbeads until there was no particle intake and fully emptied the cylinders into an empty pool. Processing images of such release events from several fully loaded micromachines (6 per condition) showed that the smaller beads were captured more efficiently (67% average) than larger beads (41% average). Larger beads occasionally clog the passage, disrupting the pumping mechanism.

At high enough concentrations, the trapped particles can jam causing the rotation of the capsule to couple directly to the rotation of the shaft and the pumping action to slow down or stop altogether. Although this synchronous motility impedes the active collection of the particles, the sieve minimizes the diffusion of particles from the capsule. Even in this undesired state, most of the microbeads remain inside the capsule throughout transport, which is desirable for therapeutic delivery and diagnostic recovery applications. Whether jammed or unjammed, the collected particles are released by reversing the flow direction by switching the rotational direction of the shaft. At a rotational rate of 10Hz, the microparticles are expelled at 9.2 pL/sec on average, which completely empties the capsule in less than 10 seconds. The flow rate, and therefore the delivered dose rate, can be adjusted through the rotation speed of the external magnetic field. As the process is reversible, the microtransporter can recollect some of the released particles for fine adjustment of the dose (See Supplementary Movie 3). We cannot completely decouple the reciprocal movement of the shaft from the overall motion of the transporter so delivery at a precise spot is challenging. However, by dynamically modulating the rotation frequency, the forward push of the pumping action and backward motion of the capsule can be balanced to minimize dispersion.

The collection, transport, and delivery of cells have challenges beyond pharmaceutically functionalized microparticles. For example, the cells adhere to the interior surface of the capsule and to each other, creating agglomerates that are too large to be collected by the

microtransporter. Cell agglomeration can be avoided with surfactants, but this increases the morbidity during transport tasks, which can take minutes to hours. Also, the loading and unloading of the microtransporter can create high shear stresses in the cells, which also can contribute to cell death. By packaging the cells in protective biodegradable alginate microbeads, both of these challenges can be mitigated. Biodegradable alginate microbeads, formed using a sterile process with relatively mild pH and temperature conditions, allow for long-term and sustained delivery of both drugs and cells.^[39] We fabricated alginate microbeads with diameters ranging from 10 to 60 μm using a microfluidic cross-junction device (see Figure S3). To test the possibility of targeted cell-based therapeutics, live mammalian cells were encapsulated inside the microbeads (Figure 2f). The encapsulated cells stay viable for days under physiological conditions.^[15] To accommodate the larger payload dimensions, the compound micromachine is scaled-up and the number of turns in the magnetic screw is increased to generate stronger fluid flows (Figure 2e). The enlarged device can successfully encapsulate and transport several alginate beads with no observed sheering or agglomeration of the encapsulated cells.

Collection and dispersion of material is only useful if the device is in the correct location. For example, cell injection therapies are occasionally hampered due to the lack of control over their delivery location, because the infused cells are often trapped in the lungs or other organs.^[40] While antibody strategies partially address this problem, an active guidance and delivery platform could significantly improve the homing of cells to the target sites. In our previous work we showed that magnetic nanohelices can be used for targeted drug and gene delivery in vitro and in vivo.^[41-43] Swarms of functionalized helical swimmers could lead to more effective therapies due to their active transport capability.^[43] However, they still face the issues related to systemic dilution and active elimination when injected as a suspension.

Using the compartmentalization concept, we tested if microtransporters could be employed to transport smaller microswimmers (**Figure 3**). The helical structures were designed with left-handed chirality as opposed to the right-handed chirality of the microtransporter's screw to force helices and microtransporters to swim towards each other during loading and in the opposite direction during release. After loading, the microtransporters can successfully carry several microswimmers. Although the microhelices form agglomerates inside the confined volume of the capsule, the rotational motion of the shaft and the propeller in conjunction with the fluidic forces disentangle such clusters during the release process. To demonstrate the capabilities of this system, we fabricated a microfluidic channel with three branches to create an artificial capillary network. The loaded microtransporters were successfully guided into the main channel and the released tiny microswimmers were navigated further to the smaller branches (see Supplementary Movie 4). The process of targeted delivery with compartmentalization is demonstrated in Figure 3b-d. Due to their size and magnetization, the helical microstructures swim slower than compound micromachines (Figure 3e).

The frequency at which the maximum forward velocity is achieved is known as step-out frequency and the machine's velocity rapidly declines when operated above that level. The step-out occurs when the maximum magnetic torque, which is a function of the device size and magnetization, is not large enough to synchronously rotate the device due to environmental effects like friction. The microtransporters described here reach step-out

around 20Hz at 9mT, whereas the microswimmers used did not reach step-out until 100Hz. Because the difference in step-out frequency is so large between the devices, the frequency of rotating magnetic field can be modulated to control the microtransporter independently from the microswimmers once released.^[44] Magnetic attraction can induce the assembly of helices during collective swimming, whereas fluidic forces disassemble them or limit the size of the assembly.^[45] In our experiments, under rotating frequencies above 60Hz, fluidic forces among microhelices were strong enough to avoid formation of large clusters.

We introduce the design and fabrication of microtransporters for targeted and triggered delivery of particles, biological materials and smaller micromachines. Functional microparts were printed together to form a single device without the need for further assembly using direct laser writing and selective magnetic film deposition. A pumping mechanism based on an Archimedean screw-pump is engineered to load and release cargo in a dose-dependent manner. The presented approach is not limited to the transport of particles, cell-loaded microcarriers and magnetic nanohelices; other passive and active suspensions can be transported inside selected microfluidic systems. The fabrication method is compatible with the production of smaller microtransporters (up to tens of μm) and the biochemical properties of the machine's surface can be precisely controlled for providing selective adhesion or anti-fouling.^[46] Some of the parts can be printed using softer materials to minimize the risk of physical damage.^[47] The surface functionalization of micromachines with near-infrared fluorophores enables visualization and tracking *in vivo*.^[43] With these improvements, compound micromachines can be used to access remote places of the body such as the peritoneal cavity,^[43] hepatic arteries^[48] and the gastrointestinal tract^[13] for targeted delivery or to perform more complicated tasks such as mechanical removal of occlusions, collection of biological samples for diagnostic analysis and generation of local fluid flows for mixing. The released magnetic agents could be navigated to even smaller conduits in the body, allowing a modular approach to the problem of cell and drug delivery.

Experimental Section

Fabrication of microtransporters and microfluidic channels

The micromachines are fabricated in the horizontal manner with SU-8 photoresist using 3D direct laser writing (The Photonic Professional Laser Lithography System, Nanoscribe GmbH) as shown in Figure 1a. This technique works well with the Galvo (Layer by Layer) Scanning Writing Mode and offers a good adhesion to the surface of the substrate during development. After development, 300nm/5nm thickness Ni/Ti bilayers are deposited only on the outer propeller using Physical Vapor Deposition (PVD). The microfluidic channel and helical microswimmers were printed independently. A high concentration suspension of helical structures coated with 50nm/5nm thick Ni/Ti bilayers were formed using tip sonication.^[42] Just before the experiments, 0.1mL of Surface Active Agent (TEEN 20) was used to lubricate the junctions of mechanical components, and the solution was degassed for five minutes to release air bubbles trapped in the capsules. The micromachines were detached from the glass substrate and transported to the experimental chamber using a Tungsten probe with a sharp tip. In experiments with biological agents, the

microtransporters were kept in a 0.5 mg/mL Polyethylene glycol (PEG) solution (SuSoS AG, Duebendorf, Switzerland).

Magnetic manipulation system

Detailed information about the magnetic manipulation system can be found in our previous work.^[25] The system consists of three pairs of stationary electromagnetic Helmholtz coils and is capable of producing uniform magnetic fields up to 10 mT and at frequencies up to 200 Hz. The micromachines and channels are immersed with deionized (DI) water inside a plastic reservoir (35mm×35mm×3mm), which is always placed at the center of the magnetic coils. Using this setup, the motion of the micromachine can be controlled in 3D. The system also allows the use of a conical rotating field to minimize the hydrodynamically-induced wobbling.^[25]

Fabrication of the alginate microbeads

Sodium alginate was purchased from FMC Biopolymer (Princeton, NJ). Calcium carbonate nanoparticles (CalEssence(R) 70 PCC) were suspended in complete Dulbecco's Modified Eagle Media (Sigma-Aldrich, St. Louis, MO) and either combined with alginate alone or with a suspension of K562 leukemia cells (ATCC) in alginate to make empty alginate beads or cell-encapsulating alginate beads, respectively. The continuous phase was prepared by mixing 1% fluorosurfactant^[49] and sterile-filtered 0.31% acetic acid (EMD Chemicals, Gibbstown, NJ) in a fluorinated oil (3M™ Novec™ Engineering Fluid HFE-7500). The continuous and aqueous phases were injected into a cross-junction microfluidic for form a water-in-oil emulsion (see Figure S3). Emulsions were broken after a 30 minute incubation by the addition of 20% 1H,1H,2H,2H-perfluorooctanol (Alfa Aesar, Ward Hill, MA).

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Sawyers C. Nature. 2004; 432:294. [PubMed: 15549090]
2. Riehemann K, Schneider SW, Luger TA, Godin B, Ferrari M, Fuchs H. Angew. Chem. Int. Edit. 2009; 48:872.
3. Farokhzad OC, Langer R. ACS Nano. 2009; 3:16. [PubMed: 19206243]
4. Shapiro B, Kulkarni S, Nacev A, Sarwar A, Preciado D, Depireux DA. Annu. Rev. Biomed. Eng. 2014; 16:455. [PubMed: 25014789]
5. Cheng R, Huang W, Yang B, Mao L, Jin K, ZhuGe Q, Zhao Y. ACS Nano. 2014; 8:7746. [PubMed: 25006696]

6. Wang Y, Zhe J, Chung BTF, Dutta P. *Microfluid Nanofluid.* 2008; 4:375.
7. Nelson BJ, Kaliakatsos IK, Abbott JJ. *Annu. Rev. Biomed. Eng.* 2010; 12:55. [PubMed: 20415589]
8. Sitti M, Ceylan H, Hu W, Giltinan J, Turan M, Yim S, Diller E. *Proc. IEEE.* 2015; 103:205.
9. Floyd S, Pawashe C, Sitti M. *IEEE T. Robot.* 2009; 25:1332.
10. Sakar MS, Steager EB, Kim DH, Kim MJ, Pappas GJ, Kumar V. *Appl. Phys. Lett.* 2010; 96:043705.
11. Jing W, Pagano N, Cappelleri DJ. *Appl. Phys. Lett.* 2009; 94:064107.
12. Peyer KE, Tottori S, Qiu F, Zhang L, Nelson BJ. *Chem. Eur. J.* 2013; 19:28. [PubMed: 23203403]
13. Gultepe E, Randhawa JS, Kadam S, Yamanaka S, Selaru FM, Shin EJ, Kalloo AN, Gracias DH. *Adv. Mater.* 2013; 25:514. [PubMed: 23047708]
14. Leong TG, Randall CL, Benson BR, Bassik N, Stern GM, Gracias DH. *Proc. Natl. Acad. Sci. USA.* 2009; 106:703. [PubMed: 19139411]
15. Fusco S, Sakar MS, Kennedy S, Peters C, Bottani R, Starsich F, Mao A, Sotiriou GA, Pane S, Pratsinis SE, Mooney D, Nelson BJ. *Adv. Mater.* 2014; 26:952. [PubMed: 24510666]
16. Diller E, Sitti M. *Adv. Funct. Mater.* 2014; 24:4397.
17. Patra D, Sengupta S, Duan W, Zhang H, Pavlick R, Sen A. *Nanoscale.* 2013; 5:1273. [PubMed: 23166050]
18. Donald BR, Levey CG, McGray C, Paprotny I, Rus D. *J. Microelectromech. Syst.* 2006; 15:1.
19. Frutiger DR, Vollmers K, Kratochvil BE, Nelson BJ. *Int. J. Rob. Res.* 2010; 29:613.
20. Zhang L, Peyer KE, Nelson BJ. *Lab Chip.* 2010; 10:2203. [PubMed: 20567752]
21. Hagiwara M, Kawahara T, Yamanishi Y, Masuda T, Feng L, Arai F. *Lab Chip.* 2011; 11:2049. [PubMed: 21562668]
22. Steager EB, Sakar MS, Magee C, Kennedy M, Cowley A, Kumar V. *Int. J. Rob. Res.* 2013; 32:346.
23. Tung H-W, Peyer KE, Sargent DF, Nelson BJ. *Appl. Phys. Lett.* 2013; 103:114101.
24. Ye Z, Sitti M. *Lab Chip.* 2014; 14:2177. [PubMed: 24663401]
25. Huang T-Y, Qiu F, Tung H-W, Chen X-B, Nelson BJ, Sakar MS. *Appl. Phys. Lett.* 2014; 105:114102.
26. Pedron S, van Lierop S, Horstman P, Penterman R, Broer DJ, Peeters E. *Adv. Funct. Mater.* 2011; 21:1624.
27. Maruo S, Ikuta K, Korogi H. *Appl. Phys. Lett.* 2003; 82:133.
28. Maruo S, Ikuta K, Korogi H. *J. Microelectromech. Syst.* 2003; 12:533.
29. Deubel M, Freymann GV, Wegener M, Pereira S, Busch K, Soukoulis CM. *Nat. Mater.* 2004; 3:444. [PubMed: 15195083]
30. LaFratta CN, Fourkas JT, Baldacchini T, Farrer RA. *Angew. Chem. Int. Ed.* 2007; 46:6238.
31. Xia H, Wang J, Tian Y, Chen Q-D, Du X-B, Zhang Y-L, He Y, Sun H-B. *Adv. Mater.* 2010; 22:3204. [PubMed: 20603886]
32. Buckmann T, Stenger N, Kadic M, Kaschke J, Frolich A, Kennerknecht T, Eberl C, Thiel M, Wegener M. *Adv. Mater.* 2012; 24:2710. [PubMed: 22495906]
33. Rohrig M, Thiel M, Worgull M, Holscher H. *Small.* 2012; 8:3009. [PubMed: 22778085]
34. Jang D, Meza LR, Greer F, Greer JR. *Nat. Mater.* 2013; 12:893. [PubMed: 23995324]
35. Galajda P, Ormos P. *Appl. Phys. Lett.* 2001; 78:249.
36. Maruo S, Inoue H. *Appl. Phys. Lett.* 2006; 89:144101.
37. Yang N, Quan Z, Zhang D, Tian Y. *Comput. Aided Des.* 2014; 56:11.
38. Tottori S, Zhang L, Qiu F, Krawczyk KK, Franco-Obregon A, Nelson BJ. *Adv. Mater.* 2012; 24:811. [PubMed: 22213276]
39. d'Ayala GG, Malinconico M, Laurienzo P. *Molecules.* 2008; 13:2069. [PubMed: 18830142]
40. Kean TJ, Lin P, Caplan AI, Dennis JE. *Stem Cells Int.* 2013:732742. [PubMed: 24000286]
41. Mhanna R, Qiu F, Zhang L, Ding Y, Sugihara K, Zenobi-Wong M, Nelson BJ. *Small.* 2014; 10:1953. [PubMed: 24616145]

42. Qiu F, Fujita S, Mhanna R, Zhang L, Simona BR, Nelson BJ. *Adv. Funct. Mater.* 2015; 25:1666–1671.
43. Servant A, Qiu F, Mazza M, Kostarelos K, Nelson BJ. *Adv. Mater.* 2015; 27:2981. [PubMed: 25850420]
44. Mahoney AW, Nelson ND, Peyer KE, Nelson BJ, Abbott JJ. *Appl. Phys. Lett.* 2014; 104:144101.
45. Tottori S, Zhang L, Peyer KE, Nelson BJ. *Nano Lett.* 2013; 3:4263. [PubMed: 23947427]
46. Richter B, Pauloehrl T, Kaschke J, Fichtner D, Fischer J, Greiner AM, Wedlich D, Wegener M, Delaittre G, Barner-Kowollik C, Bastmeyer M. *Adv. Mater.* 2013; 25:6117. [PubMed: 24038437]
47. Xing J-F, Zheng M-L, Duan X-M. *Chem. Soc. Rev.* 2015; 44:5031. [PubMed: 25992492]
48. Pouponneau P, Leroux JC, Soulez G, Gaboury L, Martel S. *Biomaterials.* 2011; 32:3481. [PubMed: 21315445]
49. Holtze C, Rowat AC, Agresti JJ, Hutchison JB, Angile FE, Schmitz CHJ, Koster S, Duan H, Humphry KJ, Scanga RA, Johnson JS, Pisignano D, Weitz DA. *Lab Chip.* 2008; 8:1632. [PubMed: 18813384]

Figure 1.

Development of compound micromachines. a) Schematic description of the fabrication process: (I) Direct laser writing of microstructures and (II) Selective physical vapor deposition of magnetic material. b) A horizontal array of micromachines before the removal of sacrificial structures. The scale bars are 20 μm and 100 μm . c) The final product after the removal of the sacrificial structures. d) The cross-sectional view of the microtransporter illustrating different mechanical components. e) Two different modes of the reciprocating mechanism (closed and open). The main shaft translates due to the rotation of the magnetic

screw. f) 3D printed microcapsule (top) and microsyringe (bottom) functioning with the same reciprocating mechanism. The scale bars in (d)-(f) are 20 μm .

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Figure 2.

a) Schematic description of the loading and releasing mechanism. b) The remotely actuated rotation of the screw generates a fluid flow around the micromachine that goes through the capsule, which leads to the loading of polystyrene microbeads (3µm diameter). c) Sequence of images summarizing the release process. Time-lapse images showing a microtransporter going through release process; after the loading process, in the beginning of the release process and at the end of the release process. d) The loading capacities of the engineered microtransporters for 3µm and 6µm beads. 100% marks the total volume of the micromachine. e) An up-scaled prototype was fabricated to transport cell-loaded alginate microbeads. f) Cell encapsulation inside alginate microbeads. Fluorescence (green, top panel) and differential interference contrast (DIC) image (bottom panel) shows the microbeads and the encapsulated cells. All scale bars are 50µm.

Figure 3.

a) Different stages of the transport and delivery of tiny helical microstructures using the microtransporter: (I) Loading, (II) Transport, (III) Release, and (IV) Deployment. b) We have successfully transported helices inside microfluidic channels. c) Helices were released at the target location with reversal of the screw rotation. Due to change in chirality, the magnetic helices swam forward while the micromachine swam backwards. d) Swarms of microhelices were directed to move into the narrower branches. The inset shows an SEM

image of an array of helical microstructures. The scale bar is 2 μm . e) Velocity profile of the microtransporter and helical microswimmers with increasing rotating frequencies.