Current Biology
Report

Visual Cortical Prosthesis with a Geomagnetic Compass Restores Spatial Navigation in Blind Rats

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http://dx.doi.org/10.1016/j.cub.2015.02.063

SUMMARY

Allocentric sense is one of the major components that underlie spatial navigation [1, 2]. In blind patients, the difficulty in spatial exploration is attributed, at least partly, to the deficit of absolute direction perception [3, 4]. In support of this notion, we announce that blind adult rats can perform spatial tasks normally when externally provided with real-time feedback of their head directions. Head-mountable microstimulators coupled with a digital geomagnetic compass were bilaterally implanted in the primary visual cortex of adult rats whose eyelids had been sutured. These "blind" rats were trained to seek food pellets in a T-shaped maze or a more complicated maze. Within tens of trials, they learned to manage the geomagnetic information source to solve the mazes. Their performance levels and navigation strategies were similar to those of normal sighted, intact rats. Thus, blind rats can recognize self-location through extrinsically provided stereotactic cues.

RESULTS AND DISCUSSION

Although the neocortex is segmented into functional subregions, such as the visual cortex and the auditory cortex [5], its anatomical laminar structure appears to be largely uniform across the subregions [6, 7]. Pioneering studies have demonstrated that the ferret auditory cortex can respond to visual stimuli and evokes visually guided behavior when retinal projections that were originally involved in the visual pathway are surgically rerouted to the auditory systems [8, 9]. Thus, the functional segregation of the neocortex is not fully genetically determined but is modifiable by the modality of sensory input. A recent study has also underpinned the latent flexibility of brain adaptation by showing that rats can perceive invisible infrared light using a neuroprosthetic intracortical stimulator with an infrared light sensor [10]; that is, neuroprosthetic devices can expand the perceptual range of the natural modality (here, concerning the visual light wavelength) of animals. However, it remains unknown whether, when the neocortex is provided externally with a new modality, animals can comprehend and leverage this concept for practical use.

To answer this question, we sought to provide eyelid-sutured adult rats with the information about their head directions through microstimulation of the neocortex. We developed a head-mountable device in which the output of a digital compass is connected to a microstimulator coupled to two electrodes (Figures 1A and 1B; Figures S1A–S1C). While the head direction of an animal remained within ±20° relative to the geomagnetic north (or the south) during exploration, the stimulator emitted electric pulse trains (50 pulses at 100 Hz) every 1 s through the right (or left) electrode (Figure 1C).

First, using normal sighted rats, we evaluated the device function. We implanted one stimulating electrode into the lateral hypothalamus, which constitutes a reward system in the brain [11], and we applied pulse trains when they faced north (Figure S1D). The rats were allowed to freely explore an open circular space for 10 min (φ = 750 mm). The observed ratio of time spent facing north was significantly greater than in a sham-operated group in which the devices did not stimulate the lateral hypothalamus (Figure S1E; *p = 0.034, t_5 = 2.90, Student's t test).

We next conducted a T-shaped maze task based on absolute orientation in which rats were trained to turn east at the T junction (Figure 2A). The rats were placed in a start box for 30 s and were then allowed to freely explore the T maze for up to 120 s until they found the pellets. During the entire session, the T maze was placed at the same location in the same testing room so that normal sighted rats could use visual cues in the room. However, in each trial, the orientation was randomly selected as northward or southward so that rats had to choose the right arm to obtain food pellets when they came from the south and the left arm when they came from the north. Twenty trials per day were conducted for 9 consecutive days. On day 1, the probability of intact rats choosing the correct arm as their first choice (success rate) corresponded to a chance level of ~50%. The rate increased gradually in the course of training and reached a steady state of approximately 80%–90% after 5–7 days (Figure S2A). This learning depended on their visual sense, as rats whose eyelids were sutured (referred to here as blind rats) exhibited no increase in success rate during a period of 9 days (Figure 2B).

We sought to rescue the spatial navigation deficit of the blind rats using a geomagnetic neuroprosthesis. We implanted the electrodes of the geomagnetic devices into the primary visual

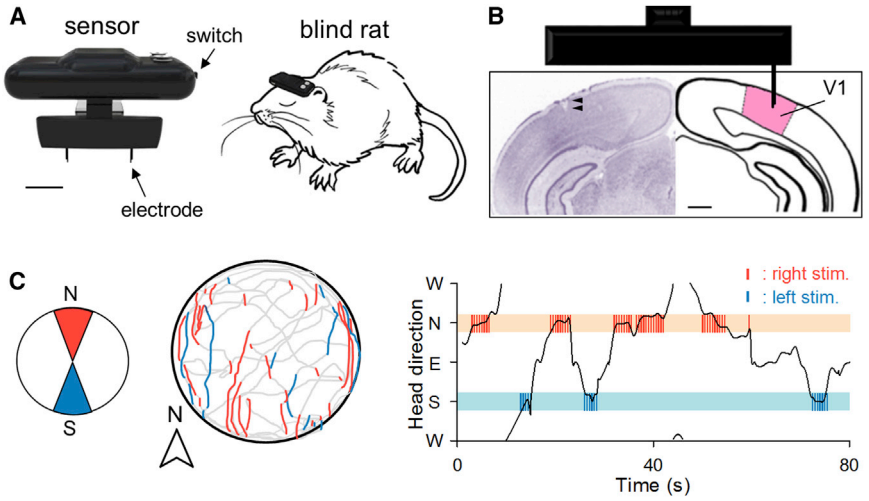

Figure 1. A Geomagnetic Sensor Is Head Mounted in an Adult Rat

(A) Left: the geomagnetic sensor device includes two stimulating electrodes and a sensor switch. The lithium battery lasts for 10 hr and is rechargeable. Right: the stimulating electrodes were bihemispherically implanted into the visual cortex of a blind rat whose eyelids had been sutured. The scale bar represents 5 mm.

(B) Nissl-stained coronal hemispheric section of the primary visual cortex (V1) into which an electrode was inserted (arrowheads). The scale bar represents 1 mm.

(C) Right and left stimulation was applied for the north and south (±20°) directions, respectively. The center diagram indicates the locations where north and south stimulation (red and blue, respectively) were applied while a rat explored a circular space (φ = 750 mm). The right plot is a portion of the center diagram, indicating the time course as a function of the head direction of the rat and the times at which stimulation was applied.

cortices of both hemispheres in blind rats (Figure 1B) and conducted the same T-maze test. The switch of the device power supply was turned off on days 1–2 and 7–8 (sensor OFF) and turned on for days 3–6 and 9 (sensor ON). On day 4 (the second sensor-ON day), the success ratio reached 81.7% ± 2.9% (mean ± SEM of 11 rats), which was significantly higher than the chance level of 50% ($P < 10^{-15}$ versus chance, Z = 9.34, Z test for a proportion). The ratio dropped to near-chance levels on days 7 and 8 (sensor OFF) and returned to a significant high level on day 9 (sensor ON; $P < 10^{-15}$, Z = 10.5). In another 9-day training session (n = 6 rats), we applied the sensor-ON condition on days 3–9, during which period we reversed the paradigm on day 7 such that rats had to turn west on days 7–9. The rats acquired this reversal learning within the first 12 trials on day 7 (Figure 2C, gray symbols).

We also confirmed that blind rats could perform the T-maze task when the electrodes were implanted in the primary somatosensory cortices of both hemispheres (Figure S2B). These results suggest that blind rats can use the artificially provided head-direction information to solve spatial tasks that otherwise must depend on vision. However, it is still possible that the rats were guided simply by the hemispheric side of the instantaneous electric stimulation at the T junction rather than by the head-direction signal per se. Therefore, in rats that had been trained for 9 days in the Figure 2B experiments, we turned off the visual cortical sensors immediately after they exited the start box on day 10 so that the rats had to forage using geomagnetic orientation during the 30-s pretrial period in the start box and had to retain this information until the subsequent arm choice (Figure 3A, ON→OFF group). The four walls surrounding the start box were identical; thus, the blind rats could not identify the wall that would contain the maze entrance gate opening. Under these conditions, the success ratio of correct arm choices was still significantly higher than chance (Figure 3B; 80.0% ± 6.2%, $^{*}p = 4.8 \times 10^{-7}$ versus chance, Z = 4.90, Z test for a proportion, n = 7 rats), and the latency to find pellets in successful trials was significantly shorter compared with the performance observed under sensor-OFF conditions (Figure 3C; $^{*}p = 1.03 \times 10^{-4}$ versus sensor OFF, $D_{317} = 0.487$, Kolmogorov-Smirnov test); the same rats were tested alternately in sensor-ON→OFF and sensor-OFF trials on day 10. The performance level under the sensor-ON→OFF condition was almost equivalent to that scored by the same rats on the previous day (day 9; sensor ON in Figures 3B and 3C).

We next conducted tests in a more complicated, asymmetrical five-arm maze (Figure 4A). In this maze, five arms branched from a single main shaft; three arms were designated for the placement of start boxes (non-rewarded arms), while the two other arms were used as pellet stations (rewarded arms). The location and orientation of the maze and the rewarded and non-rewarded arms were unchanged across trials. The rats were placed in one of the start boxes for 30 s and were allowed to explore the maze for up to 90 s. Thirty trials per day were conducted for 2 consecutive days. The number of erroneous arm entries in which rats visited the non-rewarded arms (Figure 4B) and the latency to find two pellets (Figure S3B) were recorded for each trial.

In the first trial on day 1, no difference was observed in the number of erroneous arm entries (Figure 4B), latency (Figure S3B), or total number of arm entries (Figure S3A) between intact and blind (sensor OFF) rats (error: $^{ns}p = 0.06$, $t_{17} = 2.01$; latency: $^{ns}p = 0.96$, $t_{17} = 0.05$; total arm entry: $^{ns}p = 0.25$, $t_{17} = 1.18$, Student's t test), suggesting that blindness did not affect locomotion or motivation. However, intact rats learned the maze to a saturated level within the first 30 trials, whereas blind rats learned more slowly (Figures 4B and $S3B_1$; mean error: $^{*}p = 8.1 \times 10^{-9}$, $F_{2,22} = 48.2$, Bonferroni test after one-way ANOVA; latency: $^{*}p = 2.2 \times 10^{-16}$, $F_{2,427} = 401.5$, two-way ANOVA). Notably, in the blind rats, the latency to finding pellets changed little across the entire 60 trials ($S3B_1$). Therefore, this spatial task required visual cues regarding the maze. In contrast, when the geomagnetic devices were applied (sensor ON), blind rats performed as well as intact animals (Figures 4B and $S3B_1$; mean error: $^{ns}p = 0.25$ versus intact group, $F_{2,22} = 48.9$, Bonferroni test after one-way ANOVA; latency: $^{ns}p = 0.40$ versus intact group, $F_{1,290} = 0.70$, two-way ANOVA). They learned the locations of two baits, A and B, to similar extents ($S3B_2$).

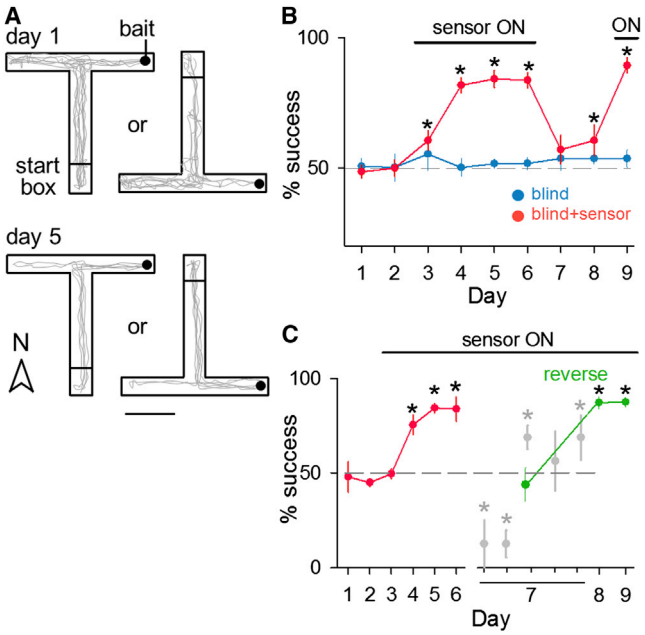

Figure 2. Blind Rats Perceive Geomagnetic Information through a Visual Cortical Prosthesis

(A) Diagrams in a T-shaped maze are superimposed with the tracks traversed by a sensor-carrying rat during representative successive five trials on days 1 (top) and 5 (bottom). The T maze was placed in two opposite orientations such that the start box was directed south (left) or north (right). In both cases, the rats had to enter the east arm to obtain bait. The scale bar represents 50 cm.

(B) The success rate of choosing the east arm was recorded (20 trials per day) in 11 blind rats with sensors (red) and 10 blind rats without sensors (blue; the sensors were turned off). The direction of the T maze was randomly chosen at each trial so that the chance of success was 50% (broken line).

(C) After training six blind rats with sensors for 6 days (red), the location of the bait was reversed to the west arm, and the training was continued (green). The gray plots on day 7 represent the averages for every four trials, indicating that rats acquired reverse learning within the first 12 trials. *$p < 0.01$ versus chance, Z test for a proportion. Error bars represent SEM.

Without spatial memory, rats might be able to solve this five-arm maze. For example, they could sequentially visit all pellet stations through an exploratory strategy regardless of the locations of the start boxes. To address this possibility, we focused on the turning probability at the first fork points. In blind rats, the probability of entering the main shaft at the first fork after exiting the start box was identical among the three start boxes, although the correct choices (i.e., whether rats should enter the main shaft or not) varied across the start boxes in this asymmetric maze (Figure 4D); that is, blind rats tended to go straight at the first corner wherever they started out. Thus, they solved the maze using the fixed foraging strategy. In contrast, intact rats adaptively changed their turning probability at the first fork, depending on the start box (Figure 4D). This flexible switching of the route choice suggests that they explored the maze using spatial navigation [12]. The behavioral patterns of blind rats with geomagnetic sensors resembled those of intact rats (Figure 4D), suggesting that blind rats could establish a spatially navigated strategy through the extrinsic head-direction signal.

On the next day (day 3), we conducted 30 additional trials in the same maze; however, in these trials, we tested intact rats in a dark (light OFF) room where visual cues were no longer available. We monitored their behaviors using an infrared camera. Under dark conditions, the intact rats took longer to find pellets in the first few trials, but within 30 trials, they performed the task as well as they did on day 1 (Figures 4C and S4). Likewise, when the head-direction cues were suddenly removed from blind rats with sensors (sensor OFF) on day 3, the performance deteriorated only in the first few trials and recovered within 30 trials (Figures 4C and S4). These learning results were better than those of continuously blinded rats on day 3 (Figures 4C and S4; light OFF versus blind: $p = 3.0 \times 10^{-4}$, $F_{1,70} = 14.5$; sensor OFF versus blind: $p = 6.9 \times 10^{-5}$, $F_{1,67} = 18.0$, two-way ANOVA) or on day 2 (light OFF versus blind: $p = 0.01$, $F_{1,107} = 6.3$; sensor OFF versus blind: $p = 7.9 \times 10^{-4}$, $F_{1,104} = 12.0$). These results suggest that blind rats that had previously explored the maze with real-time feedback of their head directions learned the maze shape and could thereby navigate the maze even under the sensor-OFF conditions. This notion is consistent with electrophysiological studies showing that once a spatial representation is formed in the hippocampus, visual input is dispensable to maintain the internal map system [13–17].

Hence, we have demonstrated that a geomagnetic neuroprosthesis can restore the spatial navigation deficits of blind animals.

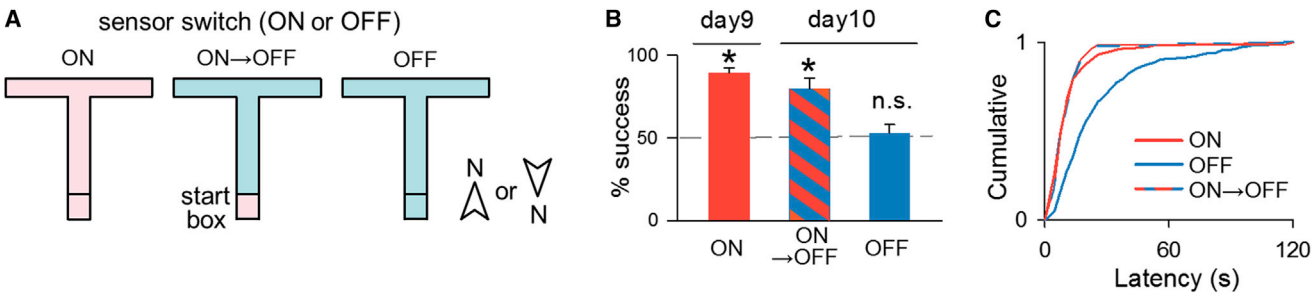

Figure 3. Rats Collect Geomagnetic Information to Solve a Spatial Maze

(A) Rats were allowed to use the geomagnetic sensor in the start box for 30 s before each trial but were forced to choose an arm with the sensor turned off (ON → OFF).

(B) Without receiving instantaneous geomagnetic information, the animals scored significantly higher success rates than by chance when they were allowed to use the sensors before the trials (hatched column). The success rates in the same animals on day 9 (with the sensor always ON) and in trials without the sensor on day 10 (with the sensor always OFF) are shown as controls. Error bars represent the SEM from seven rats each. *$p < 0.01$ versus chance, Z test for a proportion.

(C) The cumulative distribution shows the latency to finding bait in three groups. The data exclude failure trials.

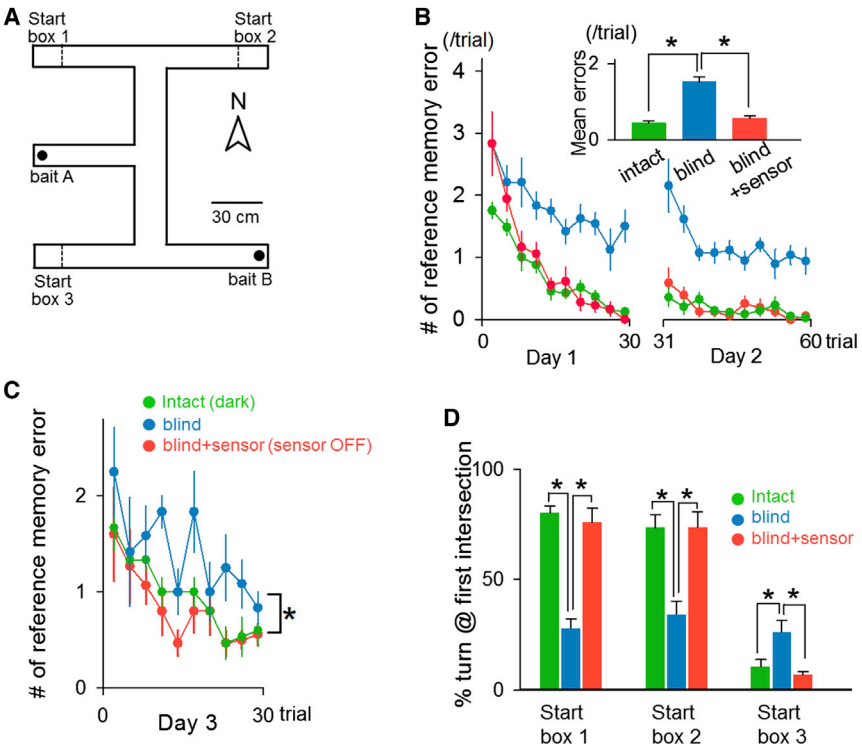

Figure 4. Blind Rats Link the Head-Direction Cues to Their Egocentric Conditions

(A) Rats were placed in one of three start boxes in an asymmetric five-arm maze and were allowed to search for bait in the ends of two arms. The orientation of the maze and the location of the bait were fixed throughout the sessions. Thirty trials per day were conducted on 2 successive days. The maximal exploration time was 90 s. The scale bar represents 30 cm.

(B) The number of entries into arms without bait (reference memory errors) was counted in 11 normal sighted, intact rats (green), six blind rats with sensors (red; sensor ON), and eight blind rats without sensors (blue; sensor OFF). The inset indicates the mean number of the errors across all trials. *$p < 0.01$, Bonferroni test after one-way ANOVA.

(C) On day 3 (the day following the 2-day sessions), blind rats were forced to find baits in the same maze but now under sensor-OFF conditions. During 30 trials, they performed more precisely and rapidly than blind rats (*$p = 6.9 \times 10^{-5}$, $F_{1,67} = 18.0$, two-way ANOVA). Intact rats under the dark (room light OFF) conditions also performed better than the blind group (*$p = 3.0 \times 10^{-4}$, $F_{1,70} = 14.5$, two-way ANOVA).

(D) The mean probability of turning to the main shaft at the first forked points after leaving the start boxes on days 1 and 2 is plotted for each start box indicated in (A) (*$p < 0.01$, Tukey's test after one-way ANOVA). Error bars represent SEM.

The fact that the blind rats did not develop a spatial navigation strategy shows that a visual signal helps to anchor egocentric information to the surrounding environment; however, our data indicate that without visual information, the externally provided head-direction information alone enabled animals to assign the self-locations in the maze. Because the visual cortex is usually not dedicated to head-direction processing, we speculate that receiving allocentric stimuli in any two neocortical loci is sufficient for the egocentric localization. The possibility still exists that the rats solved the tasks simply by associating each stimulated locus (or hemisphere) with a particular behavioral sequence. Although further investigations using more sophisticated tasks will allow for a firm conclusion, our findings suggest that the mammalian brain, even in adults, is adaptive enough to incorporate an externally provided modality into pre-existing information sources and expand the repertoire of available sensations in an experience-dependent manner. Because crossmodal prostheses may generate a vision-like sensation in humans [18], our findings shed light on a novel approach for alleviating spatial navigation deficits [19–21].

EXPERIMENTAL PROCEDURES

The experimental designs, methods, and analyses are described in the Supplemental Experimental Procedures.

SUPPLEMENTAL INFORMATION

Supplemental Information includes Supplemental Experimental Procedures and four figures and can be found with this article online at http://dx.doi.org/10.1016/j.cub.2015.02.063.

AUTHOR CONTRIBUTIONS

H.N. and Y.I. designed the study. H.N. conducted the experiments. H.N. and Y.I. analyzed the data. H.N. and Y.I. wrote the paper. The study was managed by Y.I.

ACKNOWLEDGMENTS

We are grateful to Mr. Mitsuhiro Edamura (Bio Research Center, Nagoya) for the geomagnetic sensor devices. This work is the result of the Strategic Research Program for Brain Sciences in the "Development of BMI Technologies for Clinical Application" and is partly supported by Grants-in-Aid for Scientific Research in Innovative Areas (#22115003, #25119004, and #26640003).

Received: June 23, 2014
Revised: February 9, 2015
Accepted: February 24, 2015
Published: April 2, 2015

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Supplemental Information

Visual Cortical Prosthesis with a Geomagnetic Compass Restores Spatial Navigation in Blind Rats

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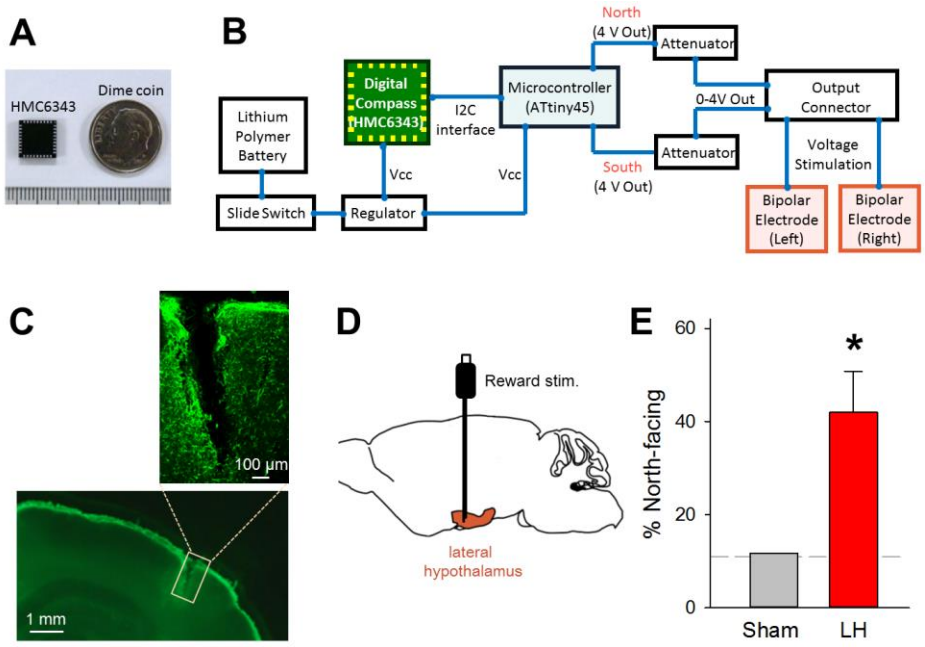
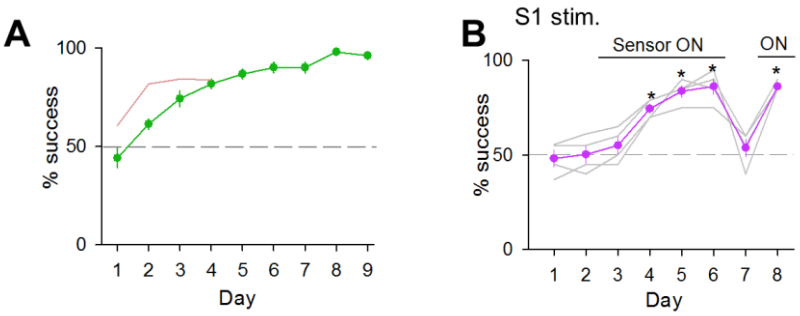
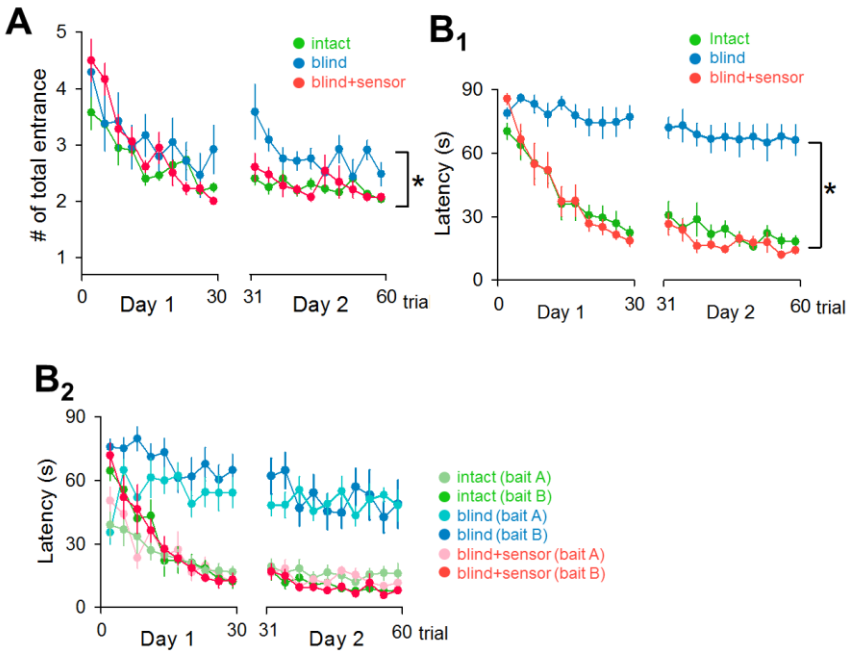


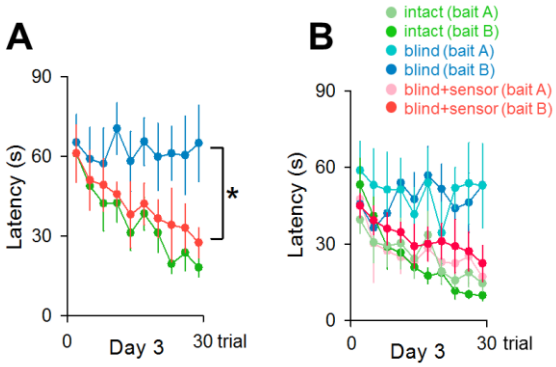
Supplementary Figure 1 | A Honeywell digital compass (HMC6343) is used in a geomagnetic neuroprosthetic device. (A) The digital compass is compared to a US dime coin. The scale unit is 1 mm. (B) The circuit diagram shows a digital compass coupled to a battery, a switch and two stimulators. (C) A coronal section of the electrode-implanted visual cortex was post-hoc immunolabeled against glial fibrillary acidic protein. Small gliosis was observed around the electrode tip. (D) A single stimulus electrode of a geomagnetic sensor was implanted into the lateral hypothalamus (LH) of the right hemisphere of an intact rat. Electric stimulation was delivered every 0.5 s while the rat directed its head toward the north ± 20°. (E) Rats without (Sham, n = 3 rats) and with electrodes in the lateral hypothalamus (LH, n = 4 rats) were allowed to freely explore an open circular space (φ = 750 mm). Rats with sensors spent more time in the north (±20°) direction. *P = 0.034, t_5 = 2.90, Student's t-test. Error bars represent SEM.



Supplementary Figure 2 | Normally sighted rats and S1-implanted rats learn the T-maze. (A) Intact rats were tested under the same T-maze paradigm indicated in Figure 2B. The pink line represents the data obtained from days 3−7 in blind rats with the sensor ON, as in Figure 2B. The blind rats learned faster than intact rats (P = 4.3×10^{-5}, $F_{1,60}$ = 19.5, two-way ANOVA). Error bars represent the SEM from 6 rats. (B) T-maze performance trained to use primary somatosensory cortex stimulation (*P< 0.01 versus chance, Z-test for a proportion). Error bars represent the SEM from 4 rats.



Supplementary Figure 3 | Performance in five-arm maze on days 1 and 2. (A) Time courses of the total numbers of arm entries in the tests in Figure 4B. $*P = 1.0\times10^{-3}$ (blind *versus* blind+sensor), $F_{1,227} = 11.12$, two-way ANOVA. (B) Time courses of the latency to obtaining the two bait sources shown in Figure 4A was compared between the groups (B_1). $*P = 2.2\times10^{-16}$, $F_{2,427} = 401.5$, two-way ANOVA. And time courses of latencies to find each bait (A or B) (B_2). Error bars represent the SEM of 11 intact rats (green), 6 blind rats with sensor ON (red) and 8 blind rats with sensor OFF (blue).



Supplementary Figure 4 | Performance in five-arm maze on day 3. (A) Time courses of the latency to obtaining the two bait sources on day 3 was compared between the groups (*P = 1.1×10^{-4}, $F_{1,68}$ = 16.9, two-way ANOVA). Intact rats under the dark (room-light OFF) conditions also performed better than the blind group (*P = 1.2×10^{-7}, $F_{1,68}$ = 35.0, two-way ANOVA). (B) Latencies are plotted separately for each bait. Error bars represent SEM of 4-5 rats.

Supplemental Experimental Procedures

Animals

Experiments were performed with the approval of the animal experiment ethics committee of the University of Tokyo (approval number: P24-5) and according to the University of Tokyo guidelines for the care and use of laboratory animals. Male adult (7-to-10-wk-old) Sprague-Dawley rats weighing 330−450 g were housed under standard laboratory conditions (12-h light/dark cycle, free access to food and water). All efforts were made to minimize the animals' suffering and the number of animals used.

Pre-training

Rats were habituated to daily handling for 2−4 weeks. Following handling habituation, they were food deprived to reduce their body weights by 10−15% and were pretrained in a linear track (1,350 mm in length, 150 mm in width). The animals were deprived of food for 18 h before each training session. For training, food pellets (40 mg, Testdiet, SLC, Shizuoka, Japan) were placed alternately at either end of the track so that the animals ran back and forth. Twenty trials were conducted, with an intervening period of 3 min and were repeated twice per day until the rats met a performance criterion of showing 100% pellet visits. They were also habituated for 10 min per day in the spatial T-maze, as described below. Following behavioral habituation, the animals were allowed to recover their weight for at least 5 days before the surgical implantation of magnetic sensors. During the recovery period, the rats were allowed food *ad libitum*. Following >5 d of recovery from the surgery, they were again food deprived and were re-trained on the same linear track for 5 d.

Magnetic sensor

A head-mountable geomagnetic sensor device was designed to connect a digital compass (HMC6343, Honeywell, Morristown, NJ, USA) to two tungsten electrodes to allow intracortical stimulation through a microcontroller (ATtiny45, Atmel, San Jose, CA, USA) and attenuators (Supplementary Figure 1). Electrical power was supplied with a repeatedly chargeable lithium battery through a slide switch on the side surface of the device. The device was 10 mm in length, 25 mm in width, 9 mm in height and weighed 2.6 g. It generated electric pulse trains (monophasic 50 pulses at 100 Hz; pulse width of 100 μs, 4 V) every 1 s through the right and left stimulating electrodes while the direction of the device (i.e., the animal's head) remained within ±20° of geomagnetic north and south, respectively.

Sensor implantation

Rats in both the blind and blind+sensor groups were surgically implanted with the geomagnetic sensor devices, but the switches were always turned off in the blind group (sham operation). The rats were anesthetized with pentobarbital (50 mg/kg, i.p.) and xylazine (10 mg/kg, i.p.). In the experiment shown in supplementary figure 2, a single electrode was inserted into the lateral hypothalamus (AP: -2.8 mm, LM: 1.7 mm, DV: -8.8 mm relative to the bregma). In the other experiments, two electrodes were bilaterally inserted into the primary visual cortex (AP: -5.7 mm, LM: ±4.5 mm, DV: -1.6 mm relative to the bregma) or the primary somatosensory cortex (AP: -1.7 mm, LM: ±5.0 mm, DV: -3.0 mm relative to the bregma). The devices were fixed to the skull using a mixture of acrylic and dental cement. Then, the eyelids of each rat were sutured with a stapler designed for medical use (35R, JSS Osaka, Japan). The rats were allowed

a postoperative recovery period of at least 5 d and were then trained again in the linear track in two 20-trial sessions per day for 5 d. In these training sessions, all blind rats received geomagnetic stimulation.

Behavioral tests

All behavioral tests were conducted in the morning. The testing room was illuminated from the ceiling at 90 lux over the spatial mazes. The magnetic sensor was turned off when the rats remained in their home cages or a rest cage during the behavioral tests.

An open field test was conducted in a circular plastic arena (750 mm in diameter, 400 mm in depth). A rat was placed at the center of the field, and its position was recorded at 2 frames per second using a video camera installed above the field center. The locomotion was analyzed using custom-made ImageJ software (National Institutes of Health, Bethesda, MD, USA).

Spatial performance was tested in a T-maze and an asymmetric five-arm maze. To diminish the use of modalities other than vision and magnetic sense, we carefully minimized the possible sources of directional information in the mazes. To reduce environmental auditory cues, the behavioral testing room was soundproofed with double walls, and white noise was provided at 65 dB. The room temperature (at 22−24°C) was controlled via the air circulation through an underground duct. The circulation fans were located in a hub building, and conditioned air was supplied to the testing room through a duct in the center of the floor under the maze and exhausted from the room through a duct in the center of the ceiling. Therefore, the sound cues, odor cues and airflow from the air supply system were radially symmetric in the room. To reduce olfactory spatial cues in the mazes, all the inner walls of the mazes were scrubbed before every trial, first

using gauze impregnated with pellet-flavored solution and then with ethanol. As the blind rats exhibited a percentage of arm choices at the chance level throughout the entire session, it can be concluded that odor cues were reduced to a virtually negligible level.

The T-maze consisted of a single main-shaft track (1,350 mm in length) and two arm tracks (each 600 mm in length) branching perpendicularly from one end of the shaft track as a T junction. The other end of the shaft track contained a start box (170 × 150 mm) with a removable door. All the tracks were 150 mm in width. The start box faced either south or north, and the direction was randomly chosen in each trial. Unless otherwise specified, food pellets (40 mg, Testdiet, SLC, Shizuoka, Japan) were placed in a dimpled dish at the end of one arm so that rats had to enter the east arm at the T junction to obtain bait, irrespective of the maze orientation. The rats were first allowed to move freely in the start box for 30 s before each trial, and the door was opened. The maximum exploration time was 120 s. When the rats reached the pellets within 120 s, they were removed from the maze and left in a rest cage for 3 min. Otherwise, at 120 s, the rats were manually guided to the end of the east arm and were transferred to the rest cage. In this case, the latency was scored as 120 s. Therefore, the latency could range from 0 to 120 s. On each test day, the animals performed 20 run trials, which were interspersed with a 3-min rest period. The sessions were repeated for 9 consecutive days. Throughout the sessions, the T-maze was fixed at the same location in the same behavior testing room, although its orientation was rotated north or south so that normally sighted, intact rats could utilize distal visual cues in the room to solve the maze.

The asymmetric five-arm maze consisted of a single main-shaft track (1,350 mm in length) and five arm tracks (each 600 mm in length) branching perpendicularly from

the two ends and the middle of the shaft track. The ends of three arms contained start boxes (150 × 150 mm) with removable doors, and the ends of the other arms were used as food stations. All tracks were 150 mm in width. For habituation, the rats were allowed to freely explore the five-arm maze without pellets for 15 min on two days. In the test sessions, the rats were placed in one of three start boxes in a random manner and were allowed to move freely for 30 s before each trial. After the door opened, the maximum exploration time was 90 s. The trials were interspersed with a rest period of 3 min and were repeated 30 times per day. These sessions were repeated for two or three consecutive days. When the rats found the bait within 90 s, they were immediately moved to a rest cage. Otherwise, after 90 s, they were sequentially guided by hand to the two reward arms and then transferred to the rest cage. In these cases, the search time was scored as 90 s.

Histology

After all of the behavioral sessions, the animals were deeply anesthetized and subjected to intracardiac perfusion with 75 ml of ice-cold phosphate-buffered saline (pH 7.4), followed by 75 ml of 4% paraformaldehyde in phosphate-buffered saline. Their brains were removed, post-fixed in 4% paraformaldehyde overnight at 4°C and then treated with 20% or 30% sucrose overnight at 4°C. The tissues were subsequently rapidly frozen using dry ice for 2 min and then coronally sectioned at a thickness of 40 μm using a cryostat (HM520 Microm, Thermo Fisher Scientific, Waltham, MA, USA). Every tenth section was arranged on glass slides. The sections were air dried and stored at -80°C until use. To confirm the position of the tip of each electrode, the sections were stained with cresyl violet or fluorescent green Nissl stain (NeuroTrace Green, Molecular Probes, Eugene, OR,

USA) and photographed using a macrozoom microscope (MVX10 MacroView, Olympus, Tokyo, Japan) or a Nipkow-disk confocal microscope (CV1000, Yokogawa, Tokyo, Japan). Behavioral data were excluded from the analyses unless the electrode tips were positioned in the lateral thalamus or the primary visual (or somatosensory) cortex. For immunostaining against glial fibrillary acidic protein (GFAP), rats were anesthetized with diethyl ether and perfused transcardially with cold phosphate-buffered saline. The brain samples were post-fixed with 4% paraformaldehyde for 3 h at 4°C and were sectioned coronally at a thickness of 300 μm. The sections were incubated with a primary rabbit monoclonal anti-GFAP antibody (1:500, Dako, Glostrup, Denmark or Sigma-Aldrich) overnight at 4°C after blocking with 5-10% goat serum and 0.1-0.3% Triton X-100 for 60 min. Then, they were labeled with a secondary anti-rabbit IgG Alexa-594 (1:400; Invitrogen) and were imaged using a confocal microscope (IX83 with FV 1200; Olympus) with water-immersion objective lens (10×) and an Ar or a green He-Ne laser.

Report

Current Biology

Visual Cortical Prosthesis with a Geomagnetic Compass Restores Spatial Navigation in Blind Rats

Highlights

- Blind adult rats were informed of their head directions via geomagnetic prosthetics
- Blind rats learned to use head-direction information to solve spatial tasks
- Thus, rats can identify their location via the externally provided direction signal

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In Brief

Norimoto and Ikegaya implanted head-mountable microstimulators coupled with digital geomagnetic compasses into the visual cortices of adult rats whose eyelids had been sutured. The rats learned to seek food pellets in spatial mazes using the head-direction signals. These data indicate that blind rats can recognize self-location through extrinsically provided stereotactic cues.

Norimoto & Ikegaya, 2015, Current Biology *25*, 1091–1095
April 20, 2015
http://dx.doi.org/10.1016/j.cub.2015.02.063

CellPress