Emergent modular neural control drives coordinated motor actions

Stefan M. Lemke, Dhakshin S. Ramanathan, Ling Guo, Seok Joon Won and Karunesh Ganguly

A remarkable feature of motor control is the ability to coordinate movements across distinct body parts into a consistent, skilled action. To reach and grasp an object, ‘gross’ arm and ‘fine’ dexterous movements must be coordinated as a single action. How the nervous system achieves this coordination is currently unknown. One possibility is that, with training, gross and fine movements are co-optimized to produce a coordinated action; alternatively, gross and fine movements may be modularly refined to function together. To address this question, we recorded neural activity in the primary motor cortex and dorsolateral striatum during reach-to-grasp skill learning in rats. During learning, the refinement of fine and gross movements was behaviorally and neurally dissociable. Furthermore, inactivation of the primary motor cortex and dorsolateral striatum had distinct effects on skilled fine and gross movements. Our results indicate that skilled movement coordination is achieved through emergent modular neural control.

In this study, we report the emerging modular neural control of fine and gross movements as a coordinated motor action was learned. We recorded neural activity in the primary motor cortex (M1) and dorsolateral striatum (DLS), the primary striatal target of M1 (refs. 1,14), throughout the learning of a reach-to-grasp skill in rats. We observed emerging coordinated low-frequency activity across M1 and DLS that was linked to the emergence of fast and consistent gross movements. Surprisingly, the emergence of skilled fine movements was independent of this activity, evolved over a longer timescale and displayed a stronger cortical reliance. Consistent with these results, inactivation of M1 and DLS had distinct effects on skilled fine and gross movements. Together, our results indicate that skilled movement coordination can be achieved through emergent modular neural control.

Results

We recorded neural signals, including single-unit activity and local field potentials (LFPs) in M1 and DLS (Supplementary Figs. 1 and 2) as rats (n = 4 animals) learned a reach-to-grasp skill. Rats were trained for 8d in automated behavioral boxes, performing 75–150 trials each day16. Refinement of both ‘gross’ movements of the forearm, for an accurate reaching action, and ‘fine’ movements of the digits, to successfully grasp the pellet, are required to learn this skill (Fig. 1a). Consistent with past results14, over 8d of learning, the success rate increased and movements became faster and more consistent (Fig. 1b,c; reach duration: 874±203 ms on day 1 to 262±10 ms on day 8, mean ± s.e.m. across animals hereafter, mixed-effects model: t(913) = -16.6, P = 3.6×10⁻¹⁶; sub-move ment timing variability: 281±97 ms to 66±34 ms, mixed-effects model: t(913) = -4.4, P = 1.7×10⁻⁶; forearm trajectory consistency: 0.86±0.02 to 0.92±0.02 mean correlation value, mixed-effects model: t(516) = 4.4, P = 1.5×10⁻⁵; success rate: 25.2±9.9% to 51.4±10.3%, mixed-effects model: t(913) = 9.4, P = 5.1×10⁻²⁰).

1Neuroscience Graduate Program, University of California San Francisco, San Francisco, CA, USA. 2Department of Neurology, University of California San Francisco, San Francisco, CA, USA. 3Neurology Service, San Francisco Veterans Affairs Medical Center, San Francisco, CA, USA. 4Mental Health Service, San Francisco Veterans Affairs Medical Center, San Francisco, CA, USA. 5Department of Psychiatry, University of California San Francisco, San Francisco, CA, USA. 6Present address: Mental Health Service, San Diego Veterans Affairs Medical Center, San Diego, CA, USA. 7Present address: Department of Psychiatry, University of California San Diego, San Diego, CA, USA. 8e-mail: karunesh.ganguly@ucsf.edu
Refinement of skilled fine and gross movements is dissociable during reach-to-grasp skill learning. We first sought to determine how changes in success rate were related to changes in fine or gross movements. During learning, we observed that success rate and changes in gross forearm movements, measured by reach duration, sub-movement timing variability and forearm trajectory consistency, evolved on different timescales. While measures of gross forearm movements reached a plateau within 8 d, the success rate remained variable, resulting in a variable success rate. In fact, we observed that differences in forearm movements did not account for success on days 5 through 8 of learning, since we found no significant differences between reach duration, sub-movement timing variability or forelimb trajectory consistency for successful and unsuccessful trials on these days (Supplementary Fig. 3; reach duration: Pearson’s correlation $r = 0.11$, $P = 0.21$; sub-movement timing variability: Pearson’s correlation $r = 0.10$, $P = 0.26$).

Importantly, the control of skilled fine movements continued to evolve over a longer timescale. In a separate ‘extended training’ cohort ($n = 3$ animals), performing approximately 2,500 trials over 4 weeks, the average success rate reached a significantly higher rate than our ‘learning cohort’ reached in 8 d, while reach duration, sub-movement timing variability and forearm trajectory consistency were not significantly different between cohorts (Fig. 1c; reach duration: $262 \pm 10$ ms for the learning cohort and $279 \pm 39$ ms for the extended training cohort, mixed-effects model: $t(714) = 0.49$, $P = 0.62$; sub-movement timing variability: $66 \pm 34$ ms and $125 \pm 22$ ms, mixed-effects model: $t(135) = 1.5$, $P = 0.12$; forearm trajectory consistency: $0.92 \pm 0.02$ and $0.91 \pm 0.01$ mean correlation value, mixed-effects model: $t(714) = -0.001$, $P = 0.99$). Furthermore, success rate did not covary with measures of gross movements on these days (Supplementary Fig. 3; reach duration: Pearson’s correlation $r = 0.001$, $P = 0.29$; sub-movement timing variability: Pearson’s correlation $r = 0.001$, $P = 0.29$).
this indicated that the refinement of skilled fine and gross movements was dissociable during reach-to-grasp skill learning.

**Coordinated movement-related activity emerges across M1 and DLS during skill learning.** With learning, reaching sub-movements became consistently timed and the velocity profile of the forearm developed a multiphasic profile (Fig. 2a, top and middle).

Strikingly, we observed that coordinated low-frequency (approximately 3–6 Hz) activity emerged during movement across M1 and DLS that was closely related to the consistent timing of sub-movements and forearm muscle activity, which also displayed a similar low-frequency component (Fig. 2a, bottom).

The emergence of coordinated low-frequency activity across M1 and DLS was clearly observed in movement-related LFP signals. The
movement-related LFP power between 3 and 6 Hz increased from day 1 to day 8 in both M1 and DLS (Fig. 2b; M1: 1.0 ± 0.13 baseline-normalized power on day 1 to 1.74 ± 0.1 on day 8, mixed-effects model: \(t(146) = 9.1, P = 5.0 \times 10^{-14};\) DLS: 1.0 ± 0.7 to 1.67 ± 0.1, mixed-effects model: \(t(94) = 6.4, P = 5.1 \times 10^{-10}\)). Movement-related LFP coherence between M1 and DLS LFP also increased in the 3–6 Hz frequency range (Fig. 2c; 0.18 ± 0.03 coherence on day 1 to 0.24 ± 0.03 on day 8, mixed-effects model: \(t(870) = 9.1, P = 9 \times 10^{-11}\)). The movement-related 3–6 Hz LFP phase lag between high-coherence M1 and DLS channels was consistent with the connectivity between M1 and DLS and inconsistent with volume conducted signals (Supplementary Fig. 4). Additionally, increases in LFP power and coherence were not solely a by-product of faster and more consistent movements, since LFP power and coherence increased for behaviorally matched trials early and late during learning (Supplementary Fig. 5).

With training, reaching sub-movements became precisely phase-locked to 3–6 Hz LFP signals in both M1 and DLS, consistent with what we would expect if this activity was involved in generating sub-movements (Fig. 2d; significant increase in inter-trial coherence (ITC) of the M1 LFP locked to movement onset: mixed-effects model: \(t(102) = 3.8, P = 2 \times 10^{-4}\); pellet touch: mixed-effects model: \(t(102) = 4.7, P = 1 \times 10^{-3}\); retraction onset: mixed-effects model: \(t(102) = 8.5, P = 2 \times 10^{-11}\); DLS LFP locked to movement onset: mixed-effects model: \(t(96) = 9.6, P = 1 \times 10^{-13}\); pellet touch: mixed-effects model: \(t(96) = 6.6, P = 3 \times 10^{-6}\); retraction onset: mixed-effects model: \(t(96) = 12.4, P = 1 \times 10^{-39}\). Additionally, the peak frequency of LFP coherence covaried with movement duration on day 8 (Supplementary Fig. 6), further suggesting that coordinated low-frequency activity across M1 and DLS was closely linked to skilled gross movements.

**Coordinated spiking activity emerges across M1 and DLS during skill learning.** The emergence of coordinated low-frequency activity across M1 and DLS was also clearly observed in movement-related spiking activity across M1 and DLS. Peri-event time histograms (PETHs) of M1 and DLS units displayed movement-related multiphasic activity locked to 3–6 Hz LFP activity (Fig. 3a). We quantified phase-locking of movement-related M1 and DLS spikes to low-frequency LFP signals by generating polar histograms of the LFP phase at which each spike occurred for a single unit and LFP channel (Fig. 3b). The non-uniformity of these histograms (indicating phase-locking) was quantified using a Raleigh test of circular non-uniformity. We compared all M1 and DLS units on day 1 and 8 to the same LFP channel in M1 and DLS and observed an increase in the percentage of M1 and DLS units phase-locked to both M1 and DLS LFP signals with training (Fig. 3c; the black vertical dashed lines correspond to the \(P = 0.05\) significance threshold of the natural log of the \(Z\) statistic; M1 unit-M1 LFP pairs: 40.2% day 1 to 76.3% day 8, \(P = 5 \times 10^{-4}\); Kolmogorov–Smirnov test; M1 unit-DLS LFP pairs: 38.9–59.6%, \(P = 6 \times 10^{-5}\); Kolmogorov–Smirnov test; DLS unit-M1 LFP pairs: 29.9–66.3%, \(P = 0.01\), Kolmogorov–Smirnov test; DLS unit-DLS LFP pairs: 37.0–66.0%, \(P = 0.03\), Kolmogorov–Smirnov test). We did not observe any clear timing differences in the average responses of phase-locked and non-phase-locked units (Supplementary Fig. 7i).

The percentage of units displaying movement-related multiphasic activity in the 3–6 Hz range also increased with learning (Fig. 3d). We classified units as ‘multiphasic’ based on the shape of each unit’s autocorrelation, providing an LFP-independent measure of movement-related low-frequency activity (47.1% of M1 multiphasic units were also phase-locked to M1 LFP on day 8, compared to 42.1% on day 1; 53.9% of DLS multiphasic units were also phase-locked to DLS LFP on day 8, compared to 50% on day 1). Strikingly, the mean cross-correlation of all M1 multiphasic units to all DLS multiphasic units displayed a short-latency peak consistent with the connectivity of M1 and DLS, as well as secondary peaks corresponding to a 3–6 Hz rhythm. This spiking relationship was not observed on day 1 (Fig. 3e). These results further suggest that coordinated low-frequency activity emerges across M1 and DLS during skill learning.

**Coordinated M1 and DLS activity is specifically linked to skilled gross movements.** If coordinated low-frequency activity across M1 and DLS is involved in generating skilled gross movements, we expect their emergence to coincide during learning. In fact, we found that increases in movement-related M1-DLS 3–6 Hz LFP coherence coincided with the transition to fast and consistent gross movements (Fig. 4a). Across animals, we observed a significant correlation between each day’s average movement-related 3–6 Hz M1-DLS LFP coherence and average reach duration, sub-movement timing variability and forelimb trajectory correlation (Fig. 4b; reach duration: Pearson’s \(r = -0.73, P = 3 \times 10^{-5}\); sub-movement timing variability: Pearson’s \(r = -0.58, P = 2 \times 10^{-5}\); forelimb trajectory correlation: Pearson’s \(r = 0.52, P = 6 \times 10^{-5}\); n = 25 sessions across 4 animals).

We next tested whether coordinated low-frequency activity across M1 and DLS was also related to changes in success rate. We compared movement-related 3–6 Hz M1-DLS LFP coherence between successful and unsuccessful trials after the stabilization of gross movements (that is, days 5 through 8 of learning; for example, Fig. 1b, D5–D8 in the gray box) and found no significant difference (Fig. 4a, c; 0.20 coherence ± 0.03 for successful trials and 0.21 ± 0.03 for unsuccessful trials, mixed-effects model: \((2,558) = 1.1, P = 0.28\). Since we attribute variability in success rate during this period to differences in fine movements, these results suggested that coordinated low-frequency activity across M1 and DLS was specifically linked to skilled gross movements.

M1 and DLS inactivation have differential effects on skilled fine and gross movements. To causally test the role of M1 and DLS activity in producing skilled fine and gross movements, we implanted a separate well-trained cohort (\(n = 5\) animals) with infusion cannulas in both M1 and DLS to acutely inactivate either M1 or DLS by muscimol infusion (Fig. 5a). To dissect impairments of either skilled fine or gross movements, we utilized a reach-to-grasp task design where the pellet was either placed at a ‘far’ position (same position used for training) or a ‘close’ position (Fig. 5b). The close position generated a reaching condition where the reliance on skilled gross movements for success was reduced, while skilled fine movements were still required to successfully grasp the pellet. Differential effects of region inactivation on success rate for the close and far position indicate differences in skilled fine and gross movement impairment.

Both acute M1 and DLS inactivation disrupted gross movements, consistent with our conclusion that coordinated activity across both M1 and DLS is closely linked to skilled gross movements (Fig. 5c; M1 inactivation/far position: reach duration: 662 ± 63 ms baseline and 2.6 ± 10 ± 667 ms post-infusion, mixed-effects model: \((311) = 11.2, P = 2 \times 10^{-3};\) sub-movement timing variability: 121 ± 35 ms and 586 ± 228 ms, mixed-effects model: \((53) = 5.7, P = 5 \times 10^{-3};\) M1 inactivation/close position: reach duration: 662 ± 63 ms and 2.7 ± 10 ± 711 ms, mixed-effects model: \((312) = 14.8, P = 6 \times 10^{-5};\) sub-movement timing variability: 121 ± 35 ms and 641 ± 89 ms, mixed-effects model: \((54) = 7.0, P = 3 \times 10^{-3};\) M1 inactivation/far position: reach duration: 448 ± 63 ms baseline and 889 ± 279 ms post-infusion, mixed-effects model: \((725) = 7.5, P = 2 \times 10^{-5};\) sub-movement timing variability: 74 ± 8 ms and 237 ± 77 ms, mixed-effects model: \((136) = 3.7, P = 3 \times 10^{-2};\) DLS inactivation/close position: reach duration: 448 ± 63 ms and 995 ± 284 ms, mixed-effects model: \((735) = 10.0, P = 3 \times 10^{-2};\) sub-movement timing variability: 74 ± 8 ms and 195 ± 82 ms, mixed-effects model: \((138) = 4.2, P = 5 \times 10^{-3}\).
Fig. 3 | Coordinated spiking activity emerges across M1 and DLS during skill learning. a, Example PETHs from units in M1 (left) and DLS (right) displaying multiphasic activity locked to 3–6 Hz LFP activity. b, Diagram of spike-LFP phase-locking: Top: raster plot of example M1 unit spiking activity during movement aligned to movement onset. Middle: example unit PETH with M1 LFP activity overlaid and extracted 3–6 Hz LFP phase. Bottom: polar histogram of the LFP phases at which spikes occurred. c, Cumulative density functions (CDFs) of z-statistic for every unit–LFP pair across and within each region (the vertical dotted lines denote the significance threshold of the z-statistic at P < 0.05; the percentage of the respective unit–LFP pairs greater than the threshold are noted; the lighter color is day 1). n = 107 M1 unit–LFP pairs on day 1, n = 80 M1 unit–LFP pairs on day 8, n = 54 DLS unit–LFP pairs on day 1, n = 47 DLS unit–LFP pairs on day 8. P values derived using a Kolmogorov–Smirnov test. d, Left: PETHs from example unit displaying multiphasic activity and corresponding autocorrelations used for classifying multiphasic and non-multiphasic units (the n values derived using a Kolmogorov–Smirnov test. 47 DLS unit–LFP pairs on day 8, 80 M1 unit–LFP pairs on day 8, n = 107 M1 unit–LFP pairs on day 1, n = 219 M1 unit–LFP pairs on day 8, n = 336 M1 unit–LFP pairs on day 1). Threshold are noted; the lighter color is day 1). e, Mean cross-correlation between all multiphasic M1 and DLS units on day 1 (gray; arrows denote the ‘bumps’ used for classification; see Methods). Right: percentage of units in M1 and DLS on day 1 and day 8 that display multiphasic activity locked to 3–6 Hz LFP activity. Top: raster plot of example M1 unit spiking activity during movement aligned to movement onset. Middle: example unit PETH with M1 LFP activity overlaid and extracted 3–6 Hz LFP phase. Bottom: polar histogram of the LFP phases at which spikes occurred. c, Cumulative density functions (CDFs) of z-statistic for every unit–LFP pair across and within each region (the vertical dotted lines denote the significance threshold of the z-statistic at P < 0.05; the percentage of the respective unit–LFP pairs greater than the threshold are noted; the lighter color is day 1). n = 107 M1 unit–LFP pairs on day 1, n = 80 M1 unit–LFP pairs on day 8, n = 54 DLS unit–LFP pairs on day 1, n = 47 DLS unit–LFP pairs on day 8. P values derived using a Kolmogorov–Smirnov test. d, Left: PETHs from example unit displaying multiphasic activity and corresponding autocorrelations used for classifying multiphasic and non-multiphasic units (the n values derived using a Kolmogorov–Smirnov test. 47 DLS unit–LFP pairs on day 8, 80 M1 unit–LFP pairs on day 8, n = 107 M1 unit–LFP pairs on day 1, n = 219 M1 unit–LFP pairs on day 8, n = 336 M1 unit–LFP pairs on day 1). Threshold are noted; the lighter color is day 1). e, Mean cross-correlation between all multiphasic M1 and DLS units on day 1 (gray; n = 46 cross-correlations) and day 8 (blue; n = 104 cross-correlations). The gray and blue lines represent the mean deviation across cross-correlations; the width of the shaded region depicts the mean ± s.e.m.
different after DLS inactivation compared to pre-infusion baseline trials (Supplementary Fig. 8).

**Off-target effects of DLS inactivation.** To examine the possibility of off-target effects of DLS inactivation, we implanted infusion cannulas in the DLS and electrodes in M1 in a separate well-trained cohort (n = 3 animals) to acutely inactivate DLS by muscimol infusion and observed the effects on M1 activity (Fig. 6a). Movement-related 3–6 Hz M1 LFP power decreased with DLS inactivation (Fig. 6b) compared to saline control (P = 5 × 10−11; sub-movement timing variability: 10−5). This suggested that the DLS is required for movement-related low-frequency activity in M1. Importantly, this change was not attributable to a general suppression of M1 activity since movement-related firing rates in M1 were not changed with DLS inactivation (Fig. 6c; mixed-effects model: t(318) = 18.1, P = 5 × 10−11). Differences in movement-related M1 LFP power or firing rate were observed after saline infusion (LFP power, mixed-effects model: t(190) = 1.3, P = 0.20; movement-related firing rate, mixed-effects model: t(168) = 0.36, P = 0.72).

**Skilled movement impairments with chronic DLS lesion.** To test whether the acute off-target effects of DLS inactivation may be causing behavioral impairments, we performed excitotoxic lesions centered on the DLS that lesioned the DLS as well as small portions of the surrounding cortex (n = 3 animals). Consistent with previous work, we observed increased reach duration, sub-movement timing variability and success rate two weeks post-lesion (Supplementary Fig. 9; reach duration: 428 ± 4.3 ms, 63 ± 5.5 ms baseline; sub-movement timing variability: 100 ± 10 s, 106 ± 13 ms baseline; success rate: 63 ± 4.3% and 40.7 ± 2.9%, success rate: 63 ± 4.3% and 40.7 ± 2.9%, mixed-effects model: t(620) = 5.5, P = 5 × 10−5). This indicated that acute off-target effects do not fully account for the disruptions in skilled gross movements observed with acute DLS inactivation. Importantly, fine grasping movements appeared preserved two weeks post-lesion (Supplementary Video 1).

**Skilled movement impairments with chronic M1 lesion.** To further determine the role of M1 in skilled movements, we tested performance after chronic M1 lesions. In a well-trained cohort (n = 5 animals), we generated photothermal lesions centered on M1 (Fig. 7a). In the first training session post-lesion (within 8–13 d post-lesion), reach duration and sub-movement timing variability...
Fig. 6 | DLS inactivation decreases movement-related low-frequency M1 activity. a, Illustration of DLS muscimol infusion and M1 recording. b, Top: 3–6 Hz filtered LFP aligned to movement onset from the example M1 channel for trials before and after DLS inactivation; individual trials are overlaid with the mean signal. Bottom: difference in movement-related 3–6 Hz LFP power in M1 before and after DLS inactivation (n = 5 sessions across 3 animals). The gray lines represent the mean power values from individual sessions and the black lines represent the mean and s.e.m. across sessions. P values from mixed-effects model. c, Top: PETH from the example M1 unit for trials before and after DLS inactivation. Bottom: difference in movement-related firing rate before and after DLS inactivation (n = 5 sessions across 4 animals). The gray lines represent the mean firing rate across units from individual sessions and the black lines represent the mean and s.e.m. across sessions. P values from mixed-effects model.

were increased and success rate was decreased (Fig. 7b; reach duration: 339 ± 63 ms baseline and 756 ± 130 ms early post-lesion, mixed-effects model: t(806) = 14.5, P = 3 × 10−42; sub-movement timing variability: 183 ± 66 ms and 365 ± 63 ms, mixed-effects model: t(195) = 6.7, P = 2 × 10−16; success rate: 69.1 ± 2.5% and 19.4 ± 7.65%, mixed-effects model: t(808) = −15.84, P = 2 × 10−49). With training, gross movement metrics recovered while success rate remained disrupted. Comparing pre-lesion performance to performance once all behavioral performance measures had plateaued (within 15–73 d post-lesion), reach duration and sub-movement timing variability were not significantly different than pre-lesion performance, but success rate remained significantly decreased (Fig. 7b; reach duration: 339 ± 63 ms baseline and 394 ± 115 ms late post-lesion, mixed-effects model: t(927) = 1.9, P = 0.06; sub-movement timing variability: 183 ± 66 ms and 238 ± 117 ms, mixed-effects model: t(225) = 1.6, P = 0.12; success rate: 69.1 ± 2.5% and 43.9 ± 2.25%, mixed-effects model: t(929) = −8.5, P = 7 × 10−17).

The differential recovery of skilled fine and gross movements further suggested skilled fine movements have strong cortical reliance.

Skilled fine movement representation in M1. Lastly, we explored the representation of skilled fine movements in M1 and DLS. We used Gaussian-process factor analysis (GPFA) to find low-dimensional neural trajectory representations of population spiking activity in M1 and DLS on individual trials35 (Fig. 8a) and then compared trajectories for successful and unsuccessful trials on days 5 through 8, during the period of learning after gross movements had stabilized (for example, Fig. 1b, D5–D8). Since we attribute whether trials were successful during this period to the control of skilled fine movements of the digits, we expected to find a difference in movement-related neural signals between successful and unsuccessful trials only if a region encodes the control of skilled fine movement.

We observed a difference between trajectories for successful and unsuccessful trials in M1 but not DLS. To compare successful and unsuccessful trials we subtracted the mean neural trajectory for successful trials, that is, the ‘successful template’, from each individual trial’s neural trajectory (Fig. 8b) and calculated the mean absolute value of the deviation during each time point from 250 ms before movement onset until pellet touch. We focused on this period because it includes the fine movements involved in shaping the digits for contact with the pellet but does not include differences in retraction or reward between successful and unsuccessful trials. Since trials differed in the duration of this period, we interpolated trajectories such that they were all the same length. M1 neural
trajectories for unsuccessful trials had significantly higher deviation than successful trials starting after movement onset (Fig. 8c, top; *$P<0.05$, mixed-effects model with Bonferroni correction for multiple comparisons). DLS neural trajectories for successful and unsuccessful trials from 250 ms before movement onset to pellet touch, across animals. $n=10$ sessions across 4 animals. The width depicts the mean deviation across sessions ± s.e.m. *$P<0.05$; $P$ value from mixed-effects model with Bonferroni correction for multiple comparisons. AU, arbitrary unit.

**Discussion**

In summary, we found that skilled fine and gross movements were behaviorally and neurally differentiable during reach-to-grasp skill learning. Coordinated low-frequency activity emerged across M1 and DLS, linked to the emergence of skilled gross movements, while the emergence of skilled fine movements was independent of this activity, evolved over a longer timescale and displayed a stronger cortical reliance. Consistently, inactivation of either M1 or DLS disrupted gross movements, while only M1 inactivation disrupted fine movements. This work provides evidence that coordinated skills can emerge from the modular refinement of movements.

**The role of M1 in skill learning and execution.** M1 has been ascribed multiple roles in learning and executing motor skills. A critical determinant for the role of M1 is whether the skill involves dexterous movements. Our results indicate that distinct patterns of M1 activity emerge independently during skill learning related to the dexterous (fine) and non-dexterous (gross) components. This suggests that M1 plays dual, but separable, roles in learning a skill composed of both dexterous and non-dexterous components.

The motor cortex is known to play a critical role in the production of dexterous movements. Motor cortical projections to the brainstem and spinal cord are thought to mediate this role. Lesions to the cortex disrupt this pathway and lead to chronic impairments in dexterous movements. Interestingly, non-dexterous movements have been shown to recover after motor cortical lesion, suggesting a greater ability for gross movements to be generated subcortically. Our results are broadly consistent with this literature. While both acute and chronic lesions disrupted skilled fine movements, aspects of gross movements recovered with chronic M1 lesion. The disruption of skilled gross movements with acute M1 inactivation raises the possibility for off-target effects.

The role of the motor cortex in non-dexterous movements can evolve with learning. A recent study found that bilateral lesions of the premotor and motor cortex after learning did not impair the performance of complex motor skills composed of non-dexterous movements. However, similar lesions before learning prevented skill acquisition. This has led to the notion that non-dexterous skills are transferred to downstream structures and that the motor cortex plays a tutor role in skill learning, that is, it is required for learning but not to execute a motor skill. What may allow non-dexterous control in the absence of M1? Skill learning is commonly associated with plasticity in corticostratial projections; therefore, it has been theorized that the striatum and thalamostriatal inputs may be critical downstream producers of these learned skills. Our results suggest that corticostratial plasticity is indeed important to learn motor skills. Additionally, it is possible that the observed coordinated activity across M1 and DLS is a neurophysiological substrate for the motor cortical tutor signal, providing a mechanism through which M1 activity patterns induce long-term plasticity in the DLS. Modeling has shown that temporally patterned inputs to the striatum can drive inter-striatal plasticity.

Importantly, in our work, recovery of aspects of gross movements after motor cortical lesion occurred over time with training. This is different from Kawai et al., where full recovery was observed after a period of 10 d with no further training. This discrepancy may be due to differences in task (for example, the reach-to-grasp task contains both dexterous and non-dexterous components) or differences in training. In Kawai et al., the complex motor skill was learned over a long period of training (approximately 20,000 training trials over approximately 30 d); it remains possible that an action containing dexterous movements may be realized by downstream circuitry after cortical lesions with enough prior training.
The role of the DLS in skill learning and execution. The DLS's role in producing skilled movements is also multifaceted. Receiving widespread cortical innervation, the DLS is positioned to play a central role in motor function as is evidenced by the motor dysfunction caused by basal ganglia disorders\(^7\). Cortical innervation of the DLS projects back to the cortex via basal ganglia output nuclei and the thalamus, forming a corticostriatal 'loop'. The precise role of this loop in the production of motor skills remains unclear, although it has been shown that striatal activity can influence motor cortical activity with low latency\(^9\) (<200 ms). It has also been shown in a brain–machine interface task, where animals learn to modulate cortical activity to achieve reward, that plasticity in corticostriatal projections is required, suggesting that the cortex may require the DLS to generate stereotyped patterns of activity\(^9\). Consistent with this notion, we found disrupted movement-related LFP activity in M1 with DLS inactivation (Fig. 6). Altogether, this suggests that the DLS may play a role in modulating cortical activity during skill learning.

Recent work has also championed a role of the DLS in controlling movement vigor\(^11,13–19\). One such model focuses on the feedforward convergent pathway of motor cortical neurons, which project to 'premotor' brainstem regions, and their collaterals, which project to the dorsal striatum, that in turn projects to the same premotor brainstem regions through basal ganglia output nuclei\(^11\). This pathway through the DLS is theorized to play a role in controlling the gain of descending motor commands based on previous experience. One prediction of this 'history-dependent gain' model (for details, see Yitiri and Dudman\(^9\)) posits that the DLS is active during the acceleration and deceleration phases of a movement, which was confirmed in a joystick\(^11\) and locomotor\(^11\) task. Similarly, we found that striatal units were active throughout the reaching action (Figs. 2 and 3; Supplementary Fig. 7). Additionally, we found evidence that disrupting striatal activity affects movement vigor, since reach amplitude was decreased with DLS inactivation. Altogether, our results support the notion that the DLS may play a role in both modulating cortical activity and controlling movement vigor for skilled gross movements. Our finding that DLS activity is linked to skilled gross movements in the rodent is consistent with work in the monkey\(^10\) and human\(^11\) basal ganglia, demonstrating a greater representation of proximal, compared to distal, portions of the limb.

What kind of 'skill' is the reach-to-grasp task? The term 'motor skill' can describe a vast range of behaviors characterized by fast, accurate and consistent movements. One group of skills has been formalized using the speed-accuracy trade-off\(^2\). Learning such skills typically involves optimizing speed while maintaining accuracy. For example, one can play a series of notes on the piano consistently and accurately on the first day of learning if the movements are performed slowly enough. Therefore, learning involves optimizing speed. Other skills require optimizing accuracy without constraints on speed (for example, shooting a free throw). The rodent reach-to-grasp skill falls under this latter type of learning. Interestingly, while there were no explicit constraints on speed, we still observed a significant decrease in movement duration. This may be because the brain's motor network, in general, biases toward faster skills to increase rewards per unit time\(^20\). Another possibility is that coordinated low-frequency activity between M1 and DLS is critical for consistent and accurate movements, and that speed is simply a by-product of network dynamics. There is evidence that increases in speed are required to maximize efficiency or maintain consistency\(^24\).

Coordination across the motor network. It has been suggested that oscillatory or rhythmic activity is central to multi-region communication\(^45–47\). Intriguingly, a growing body of work has also proposed that transient oscillatory activity is central to motor function\(^11,12,37–40\). In fact, modeling has suggested that low-frequency activity may be an essential feature of neural activity that generates descending commands to muscles\(^9\). Our work suggests that coordinated low-frequency activity is present beyond the cortex in the motor network and that such rhythmic activity may allow for the coordination of activity across regions. Further work exploring multi-region interactions will be essential to understanding the interplay between cortex and striatum, and the greater motor network, during motor skill learning.

Online content
Any methods, additional references, Nature Research reporting summaries, source data, statements of code and data availability and associated accession codes are available at https://doi.org/10.1038/s41593-019-0407-2.

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References
1. Wang, X. et al. Deconstruction of corticospinal circuits for goal-directed motor skills. Cell 171, 440–455.e14 (2017).
2. Whishaw, I. Q. An endpoint, descriptive, and kinematic comparison of skilled reaching in mice (Mus musculus) with rats (Rattus norvegicus). Behav. Brain Res. 78, 101–111 (1996).
3. Diedrichsen, J., Shadmehr, R. & Ivry, R. B. The coordination of movement: optimal feedback control and beyond. Trends Cogn. Sci. 14, 31–39 (2010).
4. Todorov, E. & Jordan, M. I. Optimal feedback control as a theory of motor coordination. Nat. Neurosci. 5, 1226–1235 (2002).
5. Kargo, W. J. & Nitz, D. A. Improvements in the signal-to-noise ratio of motor cortex cells distinguish early versus late phases of motor skill learning. J. Neurosci. 24, 5560–5569 (2004).
6. Li, Q. et al. Refinement of learned skilled movement representation in motor cortex deep output layer. Nat. Commun. 8, 15834 (2017).
7. Ramanathan, D. S., Gulati, T. & Ganguly, K. Sleep-dependent reactivation of ensembles in motor cortex promotes skill consolidation. PLoS Biol. 13, e1002263 (2015).
8. Santos, F. J., Oliveira, R. F. J., Xin, X. & Costa, R. M. Corticostriatal dynamics encode the refinement of specific behavioral variability during skill learning. eLife 4, e09423 (2015).
9. Kawai, R. et al. Motor cortex is required for learning but not for executing a motor skill. Neuron 86, 800–812 (2015).
10. Costa, R. M., Cohen, D. & Nicollelis, M. A. L. Differential corticostriatal plasticity during fast and slow motor skill learning in mice. Curr. Biol. 14, 1124–1134 (2004).
11. Kupferschmidt, D. A., Juczewski, K., Cui, G., Johnson, K. A. & Lovinger, D. M. Parallel, but dissociable, processing in discrete corticostriatal inputs encodes skill learning. Neuron 96, 476–489.e5 (2017).
12. Yin, H. H. et al. Dynamic reorganization of striatal circuits during the acquisition and consolidation of a skill. Nat. Neurosci. 12, 333–341 (2009).
13. Rueda-Orozco, P. E. & Robbe, D. The striatum multiplexes contextual and kinematic information to constrain motor habits execution. Nat. Neurosci. 18, 453–460 (2015).
14. Hintiryan, H. et al. The mouse cortico-striatal projection. Nat. Neurosci. 19, 1100–1114 (2016).
15. Wong, C. C., Ramanathan, D. S., Gulati, T., Won, S. J. & Ganguly, K. An automated behavioral box to assess forelimb function in rats. J. Neurosci. Methods 246, 30–37 (2015).
16. Lalla, L., Rueda Orozco, P. E., Jurado-Parras, M. T., Brovelli, A. & Robbe, D. Local or not local: investigating the nature of striatal theta oscillations in behaving rats. eNeuro 4, ENEURO.0128-17.2017 (2017).
17. Hall, T. M., de Carvalho, F. & Jackson, A. A common structure underlies low-frequency cortical dynamics in movement, sleep, and sedation. Neuron 83, 1185–1199 (2014).
18. Riehle, A., Wirtzsohn, S., Grün, S. & Brochier, T. Mapping the spatiotemporal structure of motor cortical LFP and spiking activities during reach-to-grasp movements. Front. Neural Circuits 7, 48 (2013).
19. Turner, R. S. & Desmurget, M. Basal ganglia contributions to motor control: a vigorous tutor. Curr. Opin. Neurobiol. 20, 704–716 (2010).
20. Mazzoni, P., Hristova, A. & Krakauer, J. W. Why don't we move faster? Parkinson's disease, movement vigor, and implicit motivation. J. Neurosci. 27, 7105–7116 (2007).
21. Panigrahi, B. et al. Dopamine is required for the neural representation and control of movement vigor. Cell 162, 1418–1430 (2015).
22. Dudman, J. T. & Krakauer, J. W. The basal ganglia: from motor commands to the control of movement. Curr. Opin. Neurobiol. 37, 158–166 (2016).
23. Yttri, E. A. & Dudman, J. T. A proposed circuit computation in basal ganglia: history-dependent gain. Mov. Disord. 33, 704–716 (2018).

24. Whishaw, I. Q., Zeeb, E., Erickson, C. & McDonald, R. J. Neurotoxic lesions of the caudate-putamen on a reaching for food task in the rat: acute sensorimotor neglect and chronic qualitative motor impairment follow lateral lesions and improved success follows medial lesions. Neuroscience 146, 86–97 (2007).

25. Yu, B. M. et al. Gaussian-process factor analysis for low-dimensional single-trial analysis of neural population activity. J. Neurophysiol. 102, 614–635 (2009).

26. Peters, A. J., Liu, H. & Komiyama, T. Learning in the rodent motor cortex. Annu. Rev. Neurosci. 40, 77–97 (2017).

27. Alaverdashvili, M. & Whishaw, I. Q. Motor cortex stroke impairs individual digit movement in skilled reaching by the rat. Eur. J. Neurosci. 28, 311–322 (2008).

28. Guo, J. Z. et al. Cortex commands the performance of skilled movement. eLife 4, e10774 (2015).

29. Lemon, R. N. Descending pathways in motor control. Annu. Rev. Neurosci. 31, 195–218 (2008).

30. Miri, A. et al. Behaviorally selective engagement of short-latency effector pathways by motor cortex. Neuron 95, 683–696.e11 (2017).

31. Ueno, M. et al. Corticospinal circuits from the sensory and motor cortices differentially regulate skilled movements through distinct spinal interneurons. Cell Rep. 23, 1286–1300.e7 (2018).

32. Lawrence, D. G. & Kuypers, H. G. The functional organization of the motor system in the monkey. I. The effects of bilateral pyramidal lesions. Brain 91, 1–14 (1968).

33. Lawrence, D. G. & Kuypers, H. G. The functional organization of the motor system in the monkey. II. The effects of lesions of the descending brain-stem pathways. Brain 91, 15–36 (1968).

34. Otchy, T. M. et al. Acute off-target effects of neural circuit manipulations. Nature 528, 358–363 (2015).

35. Díaz-Hernández, E. et al. Thalamostriatal projections contribute to the initiation and execution of a sequence of movements. Neuron 100, 739–752.e5 (2018).

36. Murray, J. M. & Escola, G. S. Learning multiple variable-speed sequences in striatum via cortical tutoring. eLife 6, e26084 (2017).

37. Dudman, J. T. & Gerfen, C. R. in The Rat Nervous System (ed. Paxinos, G.) Ch. 17, 391–410 (Academic Press, 2015).

38. Oldenburg, I. A. & Sabatini, B. L. Antagonistic but not symmetric regulation of primary motor cortex by basal ganglia direct and indirect pathways. Neuron 86, 1174–1181 (2015).

39. Koralek, A. C., Jin, X., Long, J. D. II, Costa, R. M. & Carmena, J. M. Corticostriatal plasticity is necessary for learning intentional neuroprosthetic skills. Nature 483, 331–335 (2012).

40. DeLong, M. R., Crutcher, M. D. & Georgopoulos, A. P. Primate globus pallidus and subthalamic nucleus: functional organization. J. Neurophysiol. 53, 530–543 (1985).

41. Baker, K. B. et al. Somatotopic organization in the internal segment of the globus pallidus in Parkinson’s disease. Exp. Neurol. 222, 219–225 (2010).

42. Shmuelof, L., Krakauer, J. W. & Mazzoni, P. How is a motor skill learned? Change and invariance at the levels of task success and trajectory control. J. Neurophysiol. 108, 578–594 (2012).

43. Hikosaka, O., Yamamoto, S., Yasuda, M. & Kim, H. F. Why skill matters. Trends Cogn. Sci. 17, 434–441 (2013).

44. Park, S.-W., Marino, H., Charles, S. K., Sternad, D. & Hogan, N. Moving slowly is hard for humans: limitations of dynamic primitives. J. Neurophysiol. 118, 69–83 (2017).

45. Harris, A. Z. & Gordon, J. A. Long-range neural synchrony in behavior. Annu. Rev. Neurosci. 38, 171–194 (2015).

46. Fries, P. A mechanism for cognitive dynamics: neuronal communication through neuronal coherence. Trends Cogn. Sci. 9, 474–480 (2005).

47. Fries, P. Rhythms for cognition: communication through coherence. Neuron 88, 220–235 (2015).

48. Ramanathan, D. S. et al. Low-frequency cortical activity is a neumeodulatory target that tracks recovery after stroke. Nat. Med. 24, 1257–1267 (2018).

49. Churchland, M. M. et al. Neural population dynamics during reaching. Nature 487, 51–56 (2012).

50. Sussillo, D., Churchland, M. M., Kaufman, M. T. & Shenoy, K. V. A neural network that finds a naturalistic solution for the production of muscle activity. Nat. Neurosci. 18, 1025–1033 (2015).

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Author contributions

S.M.L., D.S.R. and K.G. designed the experiments. S.M.L. and D.S.R. carried out the electrophysiology experiments. S.M.L. and D.S.R. carried out the acute inactivation experiments. S.M.L. and L.G. carried out the chronic lesion experiments. S.M.L. and S.J.W. performed the histology. S.M.L. carried out the analyses. S.M.L. and K.G. wrote the manuscript.

Competing interests

The authors declare no competing interests.

Additional information

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Correspondence and requests for materials should be addressed to K.G.

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Methods

Animal care and surgery. All procedures were in accordance with protocols approved by the Institutional Animal Care and Use Committee at the San Francisco Veterans Affairs Medical Center. Male Long-Evans rats between 3 and 4 months old weighing 250–300 g were used in this study. Animals were kept under controlled temperature and a 12-h light/12-h dark cycle with lights on at 6:00 a.m. All surgical procedures were performed using sterile techniques under 2–4% isoflurane. For electrode and/or cannula implantation, surgery involved exposure and cleaning of the skull, preparation of the skull surface (10 μg cyanoacrylate) and then implantation of skull screws for referencing and overall headstage stability. Reference screws were implanted posterior to lambda, contralateral to the neural recordings. Ground screws were implanted posterior to lambda, ipsilateral to the neural recordings. Cranioectomy and durectomy were performed, followed by implantation of neural probes or cannulas. Neural probes (32- or 64-channel 33 μm polyimide-coated tungsten microwire electrode arrays (Tucker-Davis Technologies)) or infusion cannulas (PlasticsOne) were implanted in the forelimb area of M1, centered at 3.5 mm lateral and 0.5 mm anterior to bregma and implanted in layer V at a depth of 1.5 mm, and the DLS, centered at 4 mm lateral and 0.5 mm anterior to bregma and implanted at a depth of 4.5 mm. The final location of the electrodes was confirmed by electrolytic lesion (Supplementary Fig. 1). Muscimol localization was performed by infusing a fluorescent muscimol (Supplementary Fig. 10; Invitrogen BODIPY TMR Conjugate) before perfusion and histology. The forearm was implanted with a pair of twisted electromyography (EMG) wires (0.007” single-stranded, Teflon-coated, stainless steel wire; A-M Systems). The EMG wires were fixed to the humeral epicondyle ball (J-B Weld Company) at one end, one end preceded by 1–2 mm of uncoated wire under the body. Wires were inserted into the muscle belly and pulled through until the ball came to rest on the belly. EMG wires were braided, tunneled under the skin to a scalp incision and soldered into headstage connectors. Fascia and skin incisions were closed with a suture. The postoperative recovery regimen included administration of buprenorphine at 0.02 mg kg$^{-1}$ and meloxicam at 0.2 mg kg$^{-1}$ per day. Dexamethasone (1 μg ul$^{-1}$ muscimol (Tocris Bioscience) in saline (0.9% sodium chloride) at a rate of 100 nl min$^{-1}$) was infused. After the 10 min infusion and a 5 min waiting period, a 100-trial block of the infusion protocol was the same except that a volume of 1 μl of muscimol was infused. After the 10 min infusion and a 5 min waiting period, a 200-trial block was performed at the ‘far’/normal pellet position to examine the effects of DLS inactivation on movement-related M1 activity.

Behavioral analysis. Learning was assessed using four metrics (Fig. 1): (1) reach duration defined as the time from movement onset to when the paw is fully retracted off the pellet tray (retract onset); (2) sub-movement timing variability defined as the standard deviation across trials of the duration less than 1 s from the paw touching the pellet (pellet touch) and when the paw is fully retracted off the pellet tray (retract onset); (3) success rate defined as the percentage of reaches that resulted in retrieval of the pellet into the box; and (4) forelimb trajectory consistency defined as the average correlation between each individual trial’s forelimb trajectory and the mean forelimb trajectory calculated over all trials in that session (computed separately in each of the two dimensions). These metrics were chosen because they measured changes in both gross movements of the forelimb involved in producing a consistent reach and fine movements of the fingers involved in successful grasping. For the scatterplots comparing changes in reach duration, sub-movement timing variability and forelimb trajectory consistency across learning to changes in movement-related 3–6 Hz M1–DLS LFP coherence (Fig. 4b), normalized values of reach duration, sub-movement timing variability and forelimb trajectory consistency were computed by z-scoring the 8 mean values corresponding to the 8 d of training for each animal separately, then combining the normalized values across animals. To determine body posture with DLS inactivation, we used a top camera and manually determined the body axis from nose to center of the body (Supplementary Fig. 8). We defined posture variability as the mean of the absolute value of the distance across all trials from the middle of the body to the average middle of the body position. We defined lateral bias as the mean distance in the x axis across all trials from the middle of the body to the average middle of the body position.

Inactivation experiments. We performed two sets of inactivation experiments. For both experiments, rats were first tested for forelimb preference, then underwent either dual cannula surgery (M1 and DLS cannula implantation) or cannula and electrode implantation surgery (DLS cannula and M1 electrode implantation). After the recovery period, rats were trained for 10d (100 trials per day). Following this training, the inactivation experiments began. For each session, baseline performance was calculated from 100 trials performed before muscimol infusion. For the M1/DLS inactivation experiments (Fig. 5), the infusion consisted of anesthetizing the rat (with isoflurane) and infusing 250 nl of 1 ug ul$^{-1}$ muscimol (Tocris Bioscience) in saline (0.9% sodium chloride) at a rate of 100 nl min$^{-1}$ in either M1 or DLS. After the 2.5 min infusion and a 5 min waiting period with the infusion infusion inserted, the rat was taken off anesthesia and allowed to recover for 2 h. A 200-trial block was then performed alternating between 10 trials to the ‘close’ position (5 mm from the pellet center to the slot opening in the behavioral box) and 10 trials to the ‘far’ position (15 mm from the pellet center to the slot opening in the behavioral box). For the DLS inactivation experiments (Fig. 6), the infusion protocol was the same except that a volume of 1 μl of muscimol was infused. After the 10 min infusion and a 5 min waiting period, a 200-trial block was performed at the ‘far’/normal pellet position to examine the effects of DLS inactivation on movement-related M1 activity.

Lesion experiments. For the phototothrombotic lesion experiments (Fig. 7), rats were first tested for forelimb preference, then trained for 10d (100–150 trials per day). Pre-lesion performance was measured before animals underwent phototothrombotic lesion surgery. Post-lesion, animals began performing reaching trials at variable times, so ‘early’ lesion performance was defined as the performance during the first session that animals were completing trials (within 8–13 d post-lesion). Animals used to reach training units for performance plateaus was reached, which was defined as ‘late’ lesion performance (within 15–73 d). Phototothrombotic lesion size was determined with immunohistochemistry. For the excitorotic lesion experiments (Supplementary Fig. 11), rats were first tested for forelimb preference, then trained for 10d (100 trials per day). Pre-lesion baseline performance was then measured. Excitorotic lesions were then implemented with 500 nl infusions of 10 μg ul$^{-1}$ ibotenic acid (7.4 pH; Abcam) at an infusion rate of 100 nl min$^{-1}$ at the same DLS coordinates as referenced earlier. The animals recovered for two weeks, then performance post-lesion was measured. Excitorotic lesion position and size was determined with immunohistochemistry.

In vivo electrophysiology. Units, LFP and EMG activity were recorded using an RZ2 system (Tucker-Davis Technologies). Spike data were sampled at 24,414 Hz and LFP/EMG data at 1,017 Hz. ZIF clip-based analog headstages with a unity gain and high impedance (approximately 1 GΩ) were used. Behavior-related timestamps (that is, trial onset, trial completion) and video timestamps (that is, frame times) were sent to the RZ2 analog input channel using an Arduino digital board and synchronized to the neural data.

Neural data analysis. Analyses were conducted using a combination of custom-written scripts and functions in MATLAB 2015a/2017a (MathWorks), along with functions from the EEGLAB (http://sccn.ucsd.edu/eeglab/) and the Chronux (http://chronux.org/) toolboxes.

LFP analysis. The preprocessing steps for the LFP analysis included: artifact rejection (manually removing noisy or broken channels); z-scoring an entire
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LFP channel on day 1 and 8. These histograms were generated for each unit–LFP pairs with a significance threshold of \( P = 0.05 \) (Fig. 3c). A significantly non-uniform distribution signifies phase preference for spikes of a unit to an LFP signal. To characterize further the spiking activity of each unit, we computed the percentage of units that displayed low-frequency (3–6 Hz) multiphasic activity. To do this, we computed autocorrelations on each unit's PETH. If a unit's autocorrelation had a 'peak' between 166 and 333 ms time lags (corresponding to 3–6 Hz activity), the unit was considered 'multiphasic'. A 'peak' was defined as a higher average value between 166 and 333 ms time lags than between 100 and 166 ms (Fig. 3d).

To characterize spiking interactions between M1 and DLS, we calculated the mean cross-correlation of movement-related spiking across regions for all M1 and DLS multiphasic units on days 1 and 8 (Fig. 3e). To do this, we concatenated spiking activity for all trials between ~250 and 750 ms from movement onset for each M1 and DLS multiphasic unit and then computed the cross-correlation for each M1 and DLS multiphasic unit pair using the MATLAB function 'corrcoeff'.

To determine the effects of DLS inactivation on M1 spiking activity, we compared movement-related firing rates from pre-infusion baseline trials and post-infusion trials. Movement-related firing rates were calculated by averaging the firing rate from ~250 ms before movement onset to 500 ms after movement on each trial of the session (Fig. 6c).

To characterize single-trial representations of population spiking activity we used GPA\(^{-1}\) to find low-dimensional neural trajectories for each trial (Fig. 8 and Supplementary Fig. 12). GPA analyses were carried out using the MATLAB-based graphical user interface DataHigh (version 1.12)\(^\dag\), 25 ms time bins and a dimensionality of 5. The first two factors were used for analyzing PETHs.

To determine the effects of DLS inactivation on M1 spiking activity, we compared movement-related firing rates from pre-infusion baseline trials and post-infusion trials. Movement-related firing rates were calculated by averaging the firing rate from ~250 ms before movement onset to 500 ms after movement on each trial of the session.

Viral injection. We performed two sets of viral injections in separate cohorts of rats to label the anterograde projections from M1 and the retrograde projections from the DLS. To label the anterograde projections from M1, we injected 750 nl of AAV8-hsyn-JAWs-KGC-GFP-ER2 virus into two sites (1.5 mm anterior, 2.7 mm lateral to bregma, at a depth of 1.4 mm). We labeled DLS multiphasic units in the same way. Two weeks after injection, rats were anesthetized and transcardially perfused with 0.9% sodium chloride, followed by 4% formaldehyde. The harvested brains were postfixed for 24 h and immersed in 20% sucrose for 2 d. Coronal cryostat sections (40-μm thick) were then mounted and imaged with a fluorescent microscope.

Immunohistochemistry. Cryostat sections (40-μm thick) were preincubated with blocking buffer (2% goat serum, 0.1% bovine serum albumin and 0.3% Triton X-100 in 0.1 M phosphate buffer) at room temperature, and then incubated with mouse anti-NeuN antibody, clone A60 (1 mg/ml\(^-1\); catalog no. MAB377; Millipore) overnight. After washing, the sections were incubated with biotinylated anti-mouse immunoglobulin G secondary antibody (5 mg/ml\(^-1\); Vector laboratories) for 1 h. Sections were visualized with the 3,3'-diaminobenzidine (DAB) method, using the ABC reagents provided in a Vector ABC Kit (Vector laboratories) and DAB-peroxidase substrate solution (Vector laboratories). For fluorescence imaging, sections were incubated with Alexa Fluor 594-conjugated donkey anti-mouse IgG antibody (1:1,000; catalog no. A-21203; Thermof Fisher Scientific).

Statistics. Linear mixed-effects models were used to test the significance of differences across both behavioral and neural measures. Using these models accounts for the fact that units, channels or trials from the same animal are more tightly correlated than those from different animals; thus, it is more stringent than computing statistical significance over all units, channels or trials\(^\dag\). For example, to test for a delay in peak delays during the period of training, we regressed the delay of each unit on trial number using a linear mixed-effects model (using MATLAB 'fitlm') with random intercepts/effects for each rat (\( n = 4 \)) and reported the P values for the regression coefficients associated with day 1 and 8. Similar models were used to test for changes in other behavioral or neural measures, including LFP power, LFP coherence and LFP/sub-movement \( P = \times 10^{-12} \). Two-sample Kolmogorov–Smirnov tests were used to test whether spike-LFP phase-locking values on day 1 and 8 of learning came from the same distribution. Pearson's correlation coefficient \( r \) was then used to calculate the percentage of significantly non-uniform distributions across unit–LFP pairs with a significance threshold of \( P = 0.05 \) (Fig. 3c). A significantly non-uniform distribution signifies phase preference for spikes of a unit to an LFP signal. To characterize further the spiking activity of each unit, we computed the percentage of units that displayed low-frequency (3–6 Hz) multiphasic activity. To do this, we computed autocorrelations on each unit's PETH. If a unit's autocorrelation had a 'peak' between 166 and 333 ms time lags (corresponding to 3–6 Hz activity), the unit was considered 'multiphasic'. A 'peak' was defined as a higher average value between 166 and 333 ms time lags than between 100 and 166 ms (Fig. 3d).
behavioral and neural measures. No normality tests were carried out, but individual data points have been included in the figures to display distributions.

**Reporting Summary.** Further information on research design is available in the Nature Research Reporting Summary linked to this article.

**Data availability**
The data used for the analyses that support the findings of this study are available from the corresponding author upon reasonable request.

**Code availability**
The code used for the analyses that support the findings of this study are available from the corresponding author upon reasonable request.

**References**
51. Delorme, A. & Makeig, S. EEGLAB: an open source toolbox for analysis of single-trial EEG dynamics including independent component analysis. *J. Neurosci. Methods* **134**, 9–21 (2004).
52. Bokil, H., Andrews, P., Kulkarni, J. E., Mehta, S. & Mitra, P. P. Chronux: a platform for analyzing neural signals. *J. Neurosci. Methods* **192**, 146–151 (2010).
53. Cowley, B. R. et al. DataHigh: graphical user interface for visualizing and interacting with high-dimensional neural activity. *J. Neural Eng.* **10**, 066012 (2013).
54. Aarts, E., Verhage, M., Veenwiel, J. V., Dolan, C. V. & van der Sluis, S. A solution to dependency: using multilevel analysis to accommodate nested data. *Nat. Neurosci.* **17**, 491–496 (2014).
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Software and code

Policy information about availability of computer code

Data collection

TDT OpenEx Software Suite (Version 2.30) was used for data collection. Custom MATLAB scripts (MATLAB R2017a) were used to control Arduino microcontrollers and cameras for automated behavioral boxes (as reported in Wong et al., 2015). All MATLAB scripts are available upon request.

Data analysis

Custom MATLAB scripts (MATLAB R2017a) including standard toolbox functions as well as EEGLab (https://sccn.ucsd.edu/eeglab/) and Chronux (http://chronux.org/) functions were used for data analysis. All MATLAB scripts are available upon request. Plexon OfflineSorter (v4.3.0) was used for spike sorting. Zeiss Zen 2 (version 2.0.0.0) was used for imaging.

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The data and corresponding code used for analyses that support the findings of this study are available from the corresponding author upon reasonable request.
Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

**Sample size**
Sample sizes were chosen based on previous experiments characterizing changes in neural activity during skill learning and previous experiments characterizing behavioral impairments with lesions. Behavioral (movement duration, success rate, etc.) and physiological (single unit activity, LFP power/coherence, etc.) changes with learning and with acute and chronic lesions were large enough that 3-5 animals per group were sufficient to perform relevant analyses.

**Data exclusions**
No animals or sessions were excluded from analyses. Individual broken or high-noise channels were excluded from analyses.

**Replication**
All data collected have been reported in the paper. All learning and lesion effects were investigated with multiple animals (n = 3-5) as well as multiple sessions (for lesion experiments; up to two sessions in each animal for each condition) to ensure replicability. Often, our results are consistent with previous work observing behavioral changes with learning and lesions, we have noted such replicability in the text (e.g., behavioral improvements with reach-to-grasp learning, behavioral impairments with chronic DLS lesions).

**Randomization**
Randomization was not performed in the current study. As we collected data during natural learning and inactivation experiments with automated behavioral boxes, this work did not require the random allocation of experimental subjects.

**Blinding**
Collection of learning data was not blinded, as a single experimenter collected and analyzed the data for each animal. As behavioral data was collected by automated behavioral boxes, we do not believe this introduced experimental bias. Similarly, behavioral scoring was not blinded. However, we validated such scoring by having multiple, different scorers score the same sessions.

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### Materials & experimental systems

| n/a | Involved in the study |
|-----|-----------------------|
| ☒   | Antibodies           |
| ☒   | Eukaryotic cell lines |
| ☒   | Palaeontology        |
| ☒   | Animals and other organisms |
| ☒   | Human research participants |
| ☒   | Clinical data        |

### Methods

| n/a | Involved in the study |
|-----|-----------------------|
| ☒   | ChIP-seq              |
| ☒   | Flow cytometry        |
| ☒   | MRI-based neuroimaging |

### Antibodies

**Antibodies used**
Three antibodies were used in the study: mouse anti-NeuN (1 mg/ml; Millipore, Temecula, CA), biotinylated anti-mouse IgG secondary antibody (5 mg/ml; Vector laboratories, Burlingame, CA), and Alexa Fluor 594-conjugated donkey anti-mouse IgG (Life Technologies Corporation, Grand Island, NY; 1:1000).

**Validation**
Validation for the primary antibody used (mouse anti-NeuN; 1 mg/ml; Millipore, Temecula, CA) can be found at: http://www.emdmillipore.com/Web-CA-Site/en_CA/-/CAD/ShowDocument-File?ProductSKU=MM_NF-MAB377&DocumentUID=5656845&DocumentType=COA&Language=EN&Country=US&ProductBatchNo=2424507&Origin=PDP

### Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research.

**Laboratory animals**
Male Long-Evans rats were used in this study. There were housed in a 12h:12h light/dark cycle room. Animals were received as 3-4 months old adults between 250 - 300 grams and used for the study at between 4-6 months old and 300 - 400 grams.
| Category               | Description                                                                                                                                 |
|-----------------------|---------------------------------------------------------------------------------------------------------------------------------------------|
| Wild animals          | No wild animals were used in the study.                                                                                                        |
| Field-collected samples| No field-collected samples were used in the study.                                                                                           |
| Ethics oversight      | All procedures were in accordance with protocols approved by the Institutional Animal Care and Use Committee at the San Francisco Veterans Affairs Medical Center |

Note that full information on the approval of the study protocol must also be provided in the manuscript.