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Altering spinal cord excitability enables voluntary movements after chronic complete paralysis in humans

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Previously, we reported that one individual who had a motor complete, but sensory incomplete spinal cord injury regained voluntary movement after 7 months of epidural stimulation and stand training. We presumed that the residual sensory pathways were critical in this recovery. However, we now report in three more individuals voluntary movement occurred with epidural stimulation immediately after implant even in two who were diagnosed with a motor and sensory complete lesion. We demonstrate that neuromodulating the spinal circuitry with epidural stimulation, enables completely paralysed individuals to process conceptual, auditory and visual input to regain relatively fine voluntary control of paralysed muscles. We show that neuromodulation of the sub-threshold motor state of excitability of the lumbosacral spinal networks was the key to recovery of intentional movement in four of four individuals diagnosed as having complete paralysis of the legs. We have uncovered a fundamentally new intervention strategy that can dramatically affect recovery of voluntary movement in individuals with complete paralysis even years after injury.

Introduction

The clinical diagnosis of having a motor complete lesion commonly classified by the American Spinal Injury Association Impairment Scale (AIS) as grade A or B (Marino et al., 2003; Waring et al., 2010) is when there is no clinical evidence of volitional activation of any muscle below the lesion. Presently, the prognosis of recovery of any intentional control of movement below the injury level after clinically defined complete paralysis for over 2 years is negligible. This diagnosis has immediate and severe implications for the patient often limiting the rehabilitation and interventional strategies provided that are targeted for recovery (Waters et al., 1992, 1998; Ditunno, 1999; Burns et al., 2003; Calancie et al., 2004b). There is not currently an effective treatment that would result in regaining voluntary motor function for these individuals. The presumed solution ultimately has been thought to be to promote regeneration of axons across the lesion with the hope that functionally beneficial connections can be formed (Richardson et al., 1980; David and Aguayo, 1981; Zhao et al., 2013).

Table 1 Clinical characteristics of four research participants

Subject	Age (years)	Gender	Time since injury (years)	Injury level	Neuro level	AIS grade	SSEP		TMS
							Upper	Lower	
B07	23.8	Male	3.4	C7	T2	B	Normal	BD	NR
A45	24	Male	2.2	T5–T6	T4	A	Normal	NP	NR
B13	32.8	Male	4.2	C6–C7	C7	B	Normal	BD	Not tested
A53	27	Male	2.3	T5	T5	A	Normal	NP	NR

SSEP = somatosensory evoked potential; TMS = transcranial magnetic stimulation; C = cervical; T = thoracic; BD = bilateral delay; NP = not present; NR = no response.

However, we previously reported the return of control of movement in one individual with motor complete, but sensory incomplete spinal cord injury >2 years after complete paralysis after 7 months of intense stand training using epidural stimulation (Harkema et al., 2011a). These unexpected results led us to theorize that the residual sensory pathways were critical in mediating the voluntary movements elicited with epidural stimulation and specific intent by the individual. The intense stand training and repetitive stimulation may have driven neural plasticity that eventually resulted in the ability to voluntarily move the legs. In this study, we include three additional individuals with chronic spinal cord injury; one was AIS B (sensory incomplete) and two AIS A (sensory complete), who were also classified as motor complete using all currently available clinical and neurophysiological testing (Dimitrijevic et al., 1992a; McKay et al., 1997, 2005). We tested their ability to move voluntarily with epidural stimulation after implantation (before any training with stimulation) and then again after the intense stand training using epidural stimulation intervention (see Supplementary material for description of stand training) and after intense step training in combination with epidural stimulation. We hypothesized that the individuals with AIS A spinal cord injury would not elicit any voluntary movement using epidural stimulation even after training and that the individual with AIS B spinal cord injury would develop the ability to voluntarily move the legs only after training. These experiments were conducted as part of a larger ongoing study of stand and step training in combination with epidural stimulation in individuals with chronic motor complete spinal cord injury.

Materials and methods

Characteristics of subjects

An epidural spinal cord stimulation unit (Medtronics, RestoreADVANCED) and a 16-electrode array was implanted at vertebrae T11–T12 over spinal cord segments L1–S1 (Harkema et al., 2011a) in four individuals with motor complete spinal cord injury (Table 1 and Supplementary Fig. 1) using the following inclusion criteria: (i) stable medical condition without cardiopulmonary disease or dysautonomia that would contraindicate standing or stepping with body weight support training; (ii) no painful musculoskeletal dysfunction, unhealed fracture, contracture, pressure sore, or urinary tract infection that might interfere with stand or step training; (iii) no clinically significant depression or ongoing drug abuse; (iv) no current anti-spasticity medication regimen; (v) non-progressive spinal cord injury above T10; (vi) AIS A or B; (vii) no motor response present in leg muscles during transmagnetic stimulation; (viii) not present or bilateral delay of sensory evoked potentials; (ix) no volitional control during voluntary movement attempts in leg muscles as measured by EMG activity; (x) segmental reflexes remain functional below the lesion; (xi) brain influence on spinal reflexes is not observed as measured by EMG activity; (xii) must not have received Botox® injections in the previous 6 months; (xiii) be unable to stand or step independently; (xiv) at least 1-year post-injury; and (xv) must be at least 18 years of age

All four individuals implanted were male, at least 2 years post-date of injury and ranged in neurological level from C7–T5. The average age of all individuals was 26.9 (± 4) years at the time of implant. All individuals were unable to stand or walk independently or voluntarily move their lower extremities despite standard-of-care rehabilitation and additional intensive locomotor training (Harkema et al., 2011b). The research participants signed an informed consent for electrode implantation, stimulation, stand and stepping training interventions and physiological monitoring studies approved by the University of Louisville and the University of California, Los Angeles Institutional Review Boards.

Clinical and physiological status

Standard of care clinical evaluations were performed to characterize the injury. Two clinicians independently performed a physical exam and used the AIS to classify the injury clinically (Marino et al., 2003; Waring et al., 2010). Two individuals were classified and confirmed as AIS-B and two individuals were classified and confirmed as AIS-A before implantation and at the time of initial lumbosacral spinal cord epidural stimulation (Supplementary Fig. 1).

Upper and lower extremity somatosensory evoked potentials were assessed clinically before implantation and at multiple time points following implant in all participants (Perot and Vera, 1982; Dimitrijevic et al., 1983b) (Supplementary Fig. 1). All participants had normal somatosensory evoked potentials from upper extremity median nerve stimulation at the wrist. Both AIS-B individuals had bilateral cortical delays present from lower extremity stimulation at the posterior tibial nerve and ankle, whereas both AIS-A individuals had no response (Supplementary Fig. 1). Transcranial magnetic stimulation was used clinically to assess the functional integrity of the cortico-spinal tracts (Dimitrijevic et al., 1992a; McKay et al., 1997, 2005). No motor evoked potentials in the leg muscles were detected during transcranial magnetic stimulation of the motor cortex in tested individuals. A single pulse was delivered through a dual-cone coil placed slightly off centre from the scalp vertex to optimize left and right hemispheres. Pulses of 100-μs duration were delivered starting at 30% intensity increasing by 5% until 85% of maximum intensity was reached. Data for Patients A45 (post step training with epidural stimulation) and A53 (before
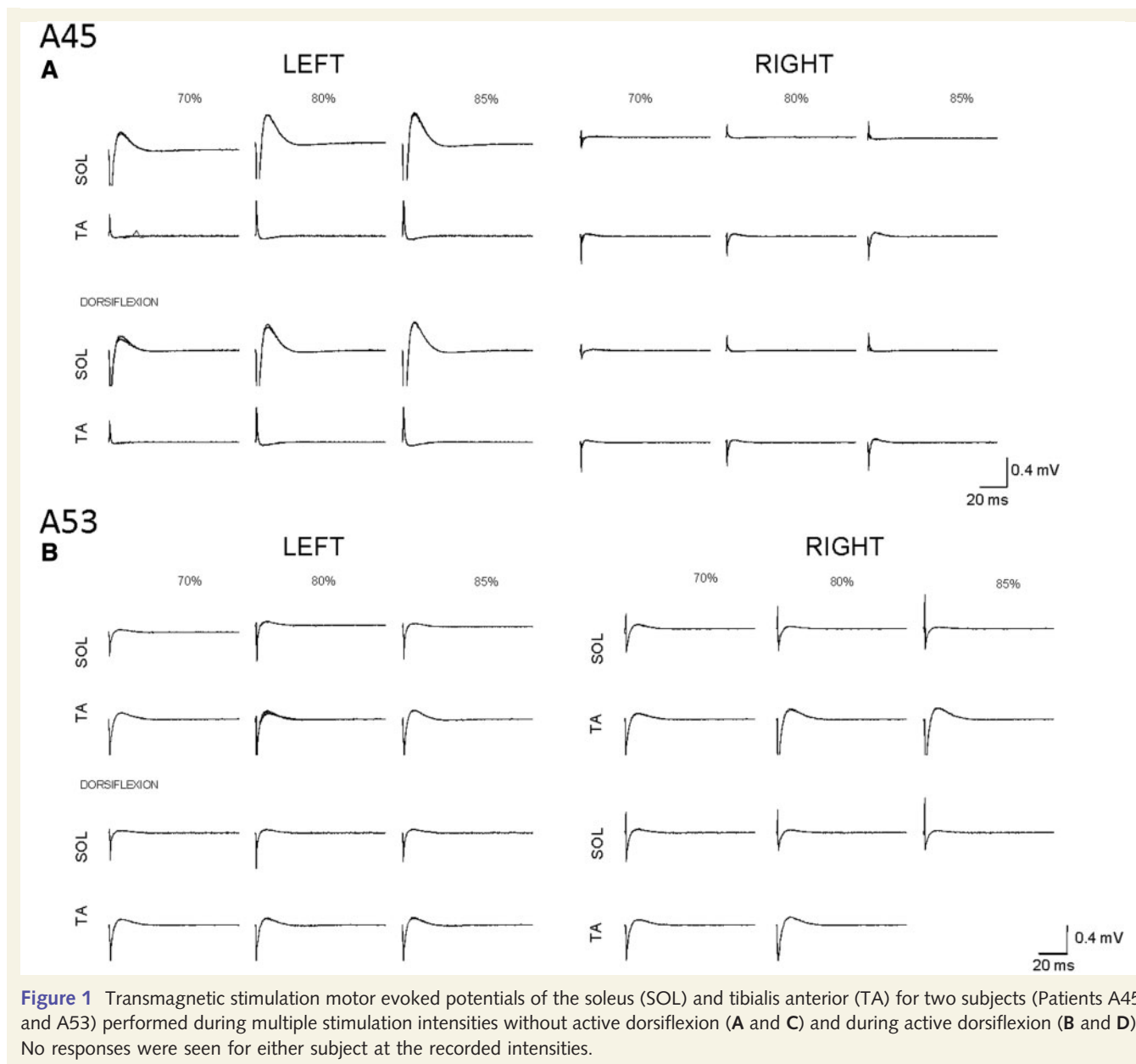
Figure 1 Transmagnetic stimulation motor evoked potentials of the soleus (SOL) and tibialis anterior (TA) for two subjects (Patients A45 and A53) performed during multiple stimulation intensities without active dorsiflexion (**A** and **C**) and during active dorsiflexion (**B** and **D**). No responses were seen for either subject at the recorded intensities.

implantation) are shown in Fig. 1A and C, respectively. Individuals were asked to attempt a sustained dorsiflexion during which transcranial magnetic stimulation was delivered following the same procedures described above. No motor evoked responses in leg muscles were detected during the combined voluntary attempt and transcranial magnetic stimulation and no movement resulted from the attempted action (Fig. 1B and D). Patient B13 was not tested before implantation. Patients A45 and A53 were classified as motor and sensory complete and Patients B07 and B13 as motor complete and sensory incomplete based on all clinical and neurophysiological measures described above.

Before electrode implantation, participants received a minimum of 80 locomotor training sessions (Harkema *et al.*, 2012*b*) using body weight support on a treadmill with manual facilitation. Average body weight support throughout all sessions for all participants was 43.6% (±4.5) walking at an average speed of 1.07 m/s (±0.04). Patients B07 and A45 had no significant EMG activity during stepping. Patients B13 and A53 had significant muscle activity during stepping.

There were no significant changes in the EMG activity during stepping following the stepping intervention before implantation in any of the four research participants (Harkema *et al.*, 2012*a*). Some neurophysiological measures can be used to detect residual descending input to the spinal circuitry even when there is no ability to voluntarily execute movement below the level of lesion and these observations have been used to reach a diagnosis of 'discomplete' (Dimitrijevic *et al.*, 1984; Sherwood *et al.*, 1992; Dimitrijevic, 1994; Calancie *et al.*, 2004*a*; McKay *et al.*, 2004). A functional neurophysiological assessment was performed at multiple time points before and after implantation (Fig. 2) to assess residual motor output present during various manoeuvres (Dimitrijevic, 1988; Dimitrijevic *et al.*, 1992*b*; McKay *et al.*, 2004, 2011; Li *et al.*, 2012). Supplementary Fig. 2 illustrates motor activity observed during 5-min relaxation, a reinforcement manoeuvre (Dimitrijevic *et al.*, 1983*a*, 1984) and attempts to actively move the ankle joint and hip joint at different time points before and after implantation. EMG activity during attempts to move the legs or during
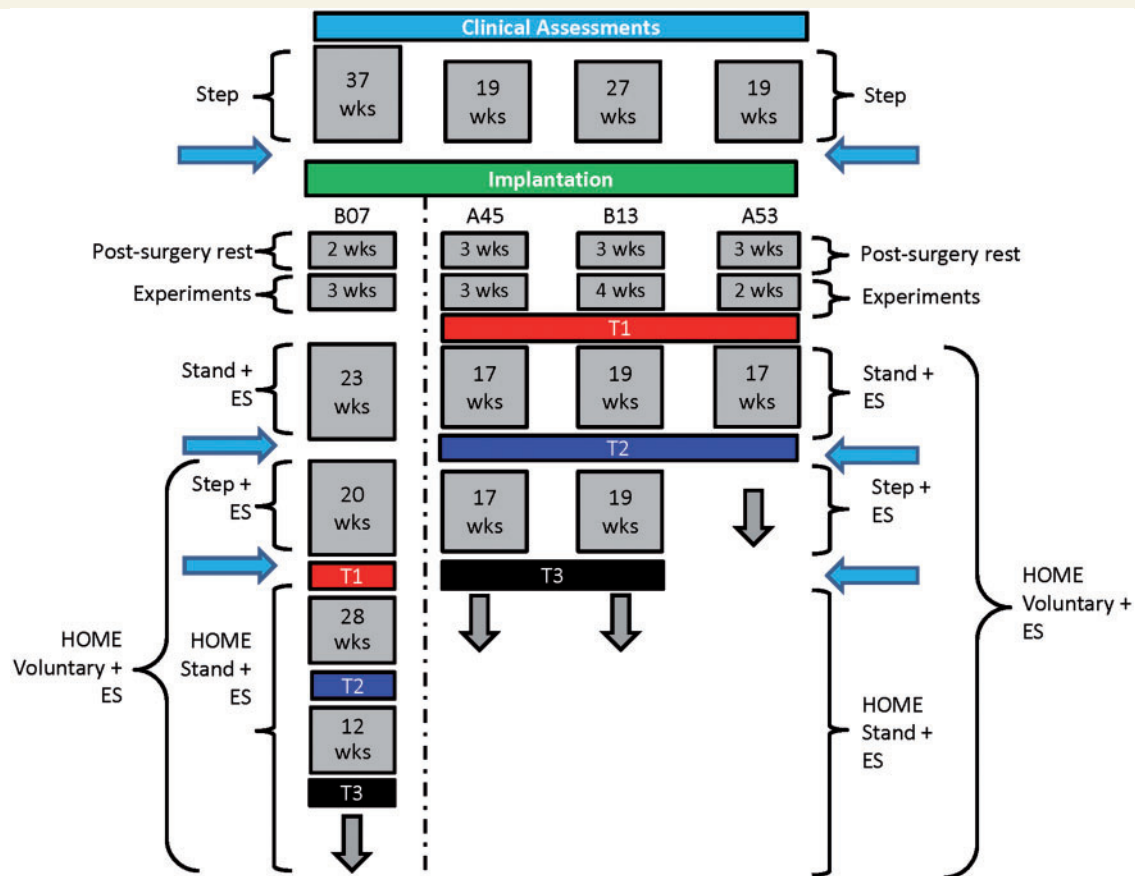
Figure 2 Timeline of implantation and experimental sessions for all participants. All individuals underwent a screening phase with at least 80 sessions of locomotor training before implantation. Patient B07 was the first subject implanted and voluntary activity was not found until the end of stand with epidural stimulation (ES). He was tested with EMG at this time point, however, T1 represents the first experimental session with force and fine motor control testing. Patients A45, B13 and A53 were tested with the same protocol post implantation and before the beginning of stand training with epidural stimulation. All participants initiated a home training programme for voluntary activity after the initial finding of their ability to move with epidural stimulation. Clinical evaluations including transcranial magnetic stimulation, Functional Neurophysiological Assessment, somatosensory evoked potentials and ASIA (American Spinal Injury Association) exams were performed before and after the 80 sessions of locomotor training during the screening phase and at the conclusion of stand and step training with epidural stimulation. Blue arrows show time points where clinical evaluations took place. Patient A53 is currently undergoing step training with epidural stimulation. See Supplementary Tables 1 and 2 for further details.

reinforcement manoeuvres was similar to the EMG seen during 5 min of relaxation in all individuals. In Patients B13 and A53 mean EMG amplitudes were higher during attempted relaxation compared with Patients B07 and A45 because of higher levels of resting excitability. In Patient A45 relaxation excitability increased post-stand training with epidural stimulation; however, no changes were seen in the ability to activate motor neurons below the level of the injury during active voluntary attempts. No functional connectivity between the supraspinal and spinal centres below the level of injury was detected with clinical or neurophysiological assessments in any of the four subjects.

Assessment of voluntary movement

Patient B07 was the first research participant to be implanted and we did not discover his ability to perform voluntary movements of the legs with epidural stimulation until the conclusion of the stand training intervention using epidural stimulation. We then designed and conducted the initial experiments to assess this individual's voluntary

function. Subsequently, the three additional research participants were implanted and before beginning any training interventions we conducted the experiments assessing voluntary movement (T1; see Fig. 2). As a result we have different time points for voluntary experiments in relation to training interventions for the first participant compared to the other three participants. Refer to the Supplementary Table 1 for a list of experimental sessions and testing time point used on each figure for each research participant.

We collected EMG, joint angles, and tensile force data at 2000 Hz using a 24-channel hard-wired AD board and custom-written acquisition software (LabView, National Instruments). Bilateral EMG (Motion Lab Systems) from the soleus, tibialis anterior, medial hamstrings, vastus lateralis, adductor magnus, gluteus maximus, and intercostal (sixth rib) muscles was recorded using bipolar surface electrodes with fixed inter-electrode distance (Harkema *et al.*, 1997). Bilateral EMG from the iliopsoas, extensor hallucis longus and extensor digitorum longus was recorded with fine wire electrodes. Two surface electrodes placed symmetrically lateral to the electrode array incision site over the

paraspinal muscles were used to record the stimulation artefact. Hip, knee, ankle and first toe joint angles were acquired using a high-speed optical motion capture system (Motion Analysis). EMG data were rectified and high-pass filtered at 32 Hz using Labview software customized by our laboratory. Tensile force was measured using a piezo-electric load cell (Kistler) mounted on a frame placed around the mat table.

Stimulation parameters were optimized for each leg and joint movement. Cathodes and anodes were selected to target primary motor pool activation areas that were key in the movement. Stimulation was constant and all participants achieved movement at optimal frequencies of either 25 Hz or 30 Hz. Voltage was typically tested through a wide range from sub-movement threshold to above optimal. Although initially configuration parameters were specific to a single joint movement and right or left leg, after training, participants were able to perform multiple movements on left and right legs using the same configuration (Supplementary Table 2).

The participants were in a supine position throughout all recordings. To measure force during extension of the first toe a ring was secured over the toe and attached to a force transducer by way of a non-elastic cable. Similarly the ring and cable were secured around the distal portion of the foot when recording force during ankle dorsiflexion. To record force during flexion of the knee and hip combined the cable was secured to the ankle. The cable had sufficient compliance to allow the leg to move from full extension to a position in which the knee and hip could reach a $<30°$ angle. The resistance to the movement was equivalent to the weight of the cable (0.16 N) and the friction force of the foot with the table in the early phase of the leg flexion. A computer monitor displayed a real time sine wave with frequency of 0.25 Hz as well as the force measured by the load cell as the research participants performed the requested action. Both signals were used to determine the degree to which the individual could translate the timing of the visual appearance of the sine wave to an analogous change in force. Although the sine wave was continuously displayed to the participants, they were instructed to initiate each effort with the rising phase of a sine wave of their choosing. There was no instruction to match the amplitude of force generation with the peak of the sine wave, and there was no verbal command given before the attempts.

To assess the ability to translate an auditory signal to an analogous change in force a signal from a tone generator (300 Hz tone frequency), modulated at 0.25 Hz was sent through headphones. Three different volumes (60, 70 and 80 dB) were presented in a random order. The research participant was instructed to modulate the force based on the modulation frequency of the sound and the given amplitude. A trial period was conducted so the participant could listen at the three different volumes and learn to discriminate between them. Full leg flexion was the only action tested with the auditory cues. Stimulation parameters used during these trials were matched to those found as optimal during the visual cue assessment for the same leg.

During experimental sessions continuous stimulation was provided with optimal parameters during all attempts. Stimulation was shut down only to change configurations and to assess the ability to move with no stimulation at the beginning of trials for each joint and side. Experimental sessions lasted for ~2 h and left/right leg, ankle and toe were tested starting by assessing threshold and optimal stimulation parameters using visual cues. Each joint was also assessed for fine motor control and accuracy including the modulation of three levels of force generation, fast oscillations and sustained force.

Voluntary movement training programme

A home-based stimulation protocol was developed to allow the individuals to stimulate for ~1 h while practicing intentional movement in the supine position. This protocol started immediately after the first assessment of successful voluntary movement was obtained (Fig. 2). All research participants were asked to practice daily (7 days a week) for 1 h. The stimulation protocol for the home-based programme was determined in the laboratory. The individuals modulated the voltage needed to optimize each movement after the stimulation configurations were selected for each individual to optimize the voluntary movement of the whole leg, ankle and toe (Supplementary videos 1-3).

Results

Four individuals diagnosed with clinically motor complete paralysis and implanted with a lumbrosacral spinal cord stimulator at least 2.2 years post injury (3.0 ± 0.95 years) were able to execute intentional movements of the legs in response to a verbal command. No motor activity was present when attempting to move without epidural stimulation following a verbal command (Patient B07) or a visual cue (Patients B13, A45 and A53) (Fig. 3A). However, all four individuals were able to generate EMG activity and movement during ankle dorsiflexion in the presence of epidural stimulation (Fig. 3B) during their first experimental session [T1 (before stand training): Patients A45, B13, A53 and post stand training: Patient B07, see Fig.2].

Appropriate activation and movement of ankle and toe muscles was achieved by all individuals when performing ankle dorsiflexion with epidural stimulation (Fig. 3B). In one subject (Patient B13) clonic-like activity in the toe extensors was present during the movement and sustained clonic activity remained when commanded to relax (Beres-Jones *et al*., 2003). However, this clonic activity did not prevent the movement. Patients A45 and A53 had tonic activity in the soleus before the attempt, showing a reduction in amplitude during the dorsiflexion effort. Reciprocal inhibition of antagonist muscles was also present in the execution of other movements such as leg flexion. These results demonstrate that humans diagnosed with complete motor paralysis can recover volitional motor drive which can drive coordinated, task-specific movements in the presence of lumbosacral spinal cord epidural stimulation.

Force and rate of modulation

Having observed some recovery of the ability to move intentionally by each of the four individuals, we began to assess the motor control fidelity. Graded levels of force were generated on command by three of four individuals (Fig. 4A and B). Three subjects (Patients B07, A45 and A53) generated forces that proportionately matched the percent of effort requested in flexing the entire leg (Fig. 4B). The level of activation of the iliopsoas muscle reflected the relative magnitude of force generated during leg flexion. The activation of the intercostal muscles (sixth rib), was closely synchronized with the initiation of force generation, generally reflecting the percent effort in stabilizing the rib cage (Fig. 4A).
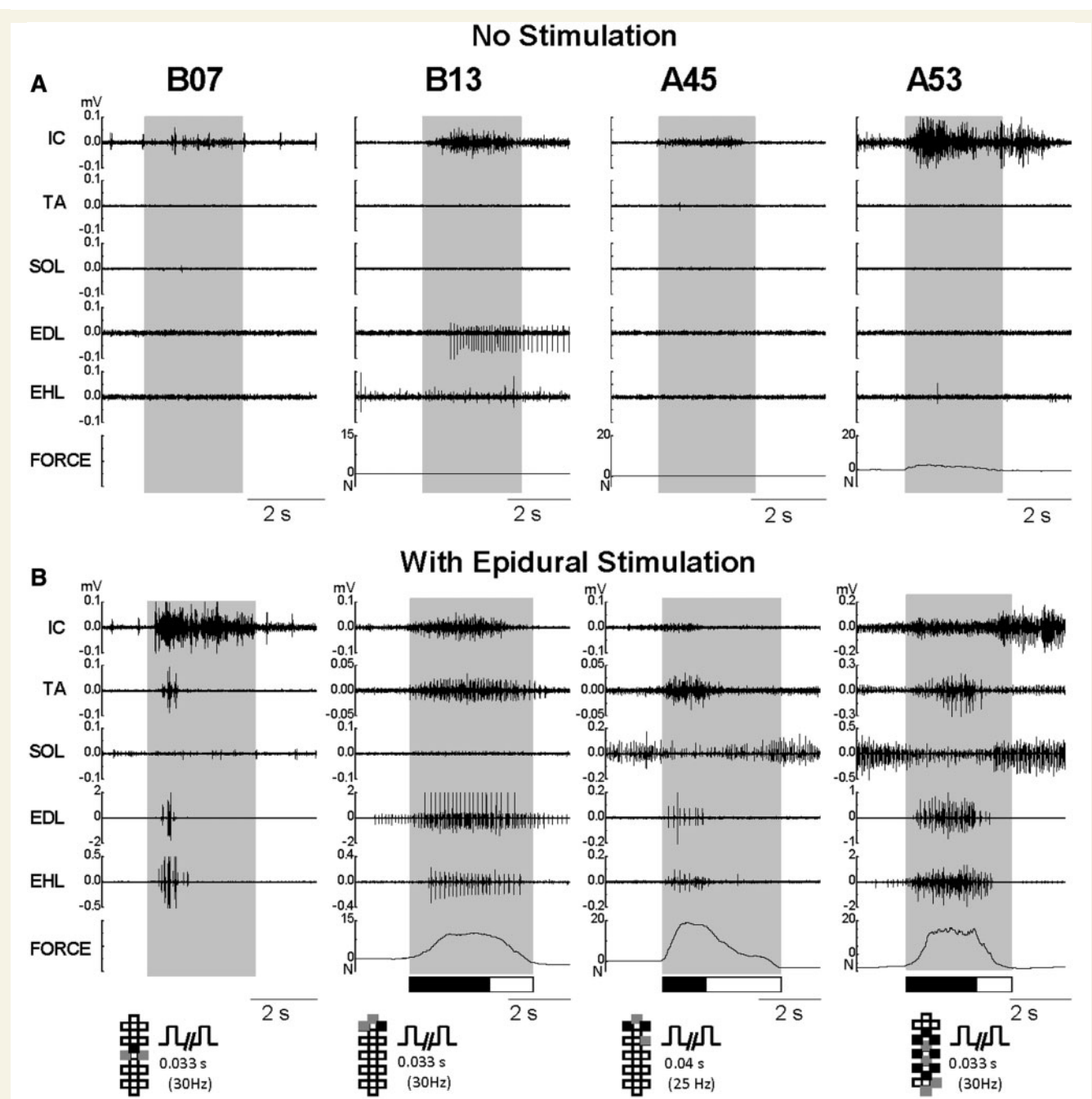
Figure 3 Lower extremity EMG activity during voluntary movement occurred only with epidural stimulation in four individuals with motor complete spinal cord injury. EMG activity during attempts of ankle dorsiflexion (**A**) without stimulation and (**B**) with stimulation. Force was not collected for Patient B07. Electrode representation for each subject denotes the stimulation configuration used. Although stimulation was applied throughout the time shown in **B**, in all four subjects EMG bursts were synchronized with the intent to move. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency varied from 25 to 30 Hz. Muscles, surface EMG: intercostal sixth rib (IC), tibialis anterior (TA), soleus (SOL); fine wire EMG: iliopsoas (IL), extensor digitorum longus (EDL), extensor hallucis longus (EHL). Black bars represent flexion as determined by peak force generation and white bars represent passive extension (from peak force to return to resting position). Although the subjects were instructed to flex and extend, they were unable to actively extend before training. Grey highlighted indicates the active 'flexion/extension' period.

Reciprocity in the EMG activity of extensors and flexors when flexing and extending the leg further indicates that the neural activation specificity to the lumbosacral motor pools was sufficient to generate a functionally coordinated movement (Fig. 4A and C).

When Patient A45 was asked to generate a sustained contraction for 'as long as possible' the effort could be maintained for a few seconds followed by several oscillatory movements (Fig. 4C). There was alternating activity in the iliopsoas and vastus lateralis,
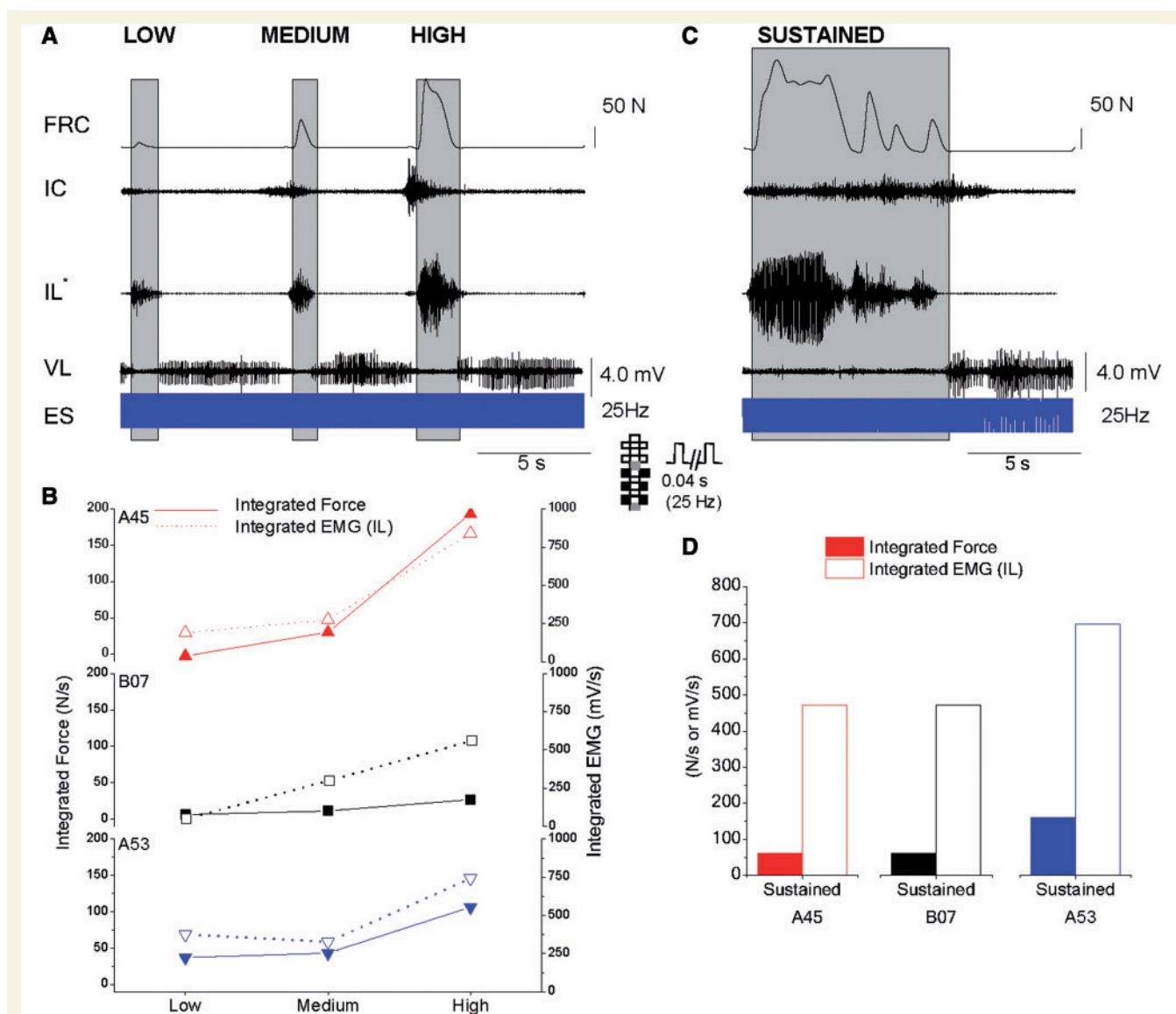
Figure 4 Force level and endurance of voluntary movements. (**A**) Left leg force and left iliopsoas, vastus lateralis and intercostals EMG activity generated during a low (20%), medium (60%) and high (100%) effort of hip/knee flexion with epidural stimulation from Patient A45. Grey shading indicates force duration. (**B**) Integrated leg force (left *y*-axis) and iliopsoas EMG activity (right *y*-axis) from Patients A45 (red), B07 (black) and A53 (blue) during hip/knee flexion with epidural stimulation. Solid symbols (with solid line) represent the integrated force for each attempt and open symbols (with dash line) represent the integrated EMG of the iliopsoas muscle for each attempt. (**C**) Left leg force and iliopsoas, vastus lateralis and intercostals EMG activity generated during a request to sustain voluntary flexion as long as possible from Patient A45. Shaded grey indicates duration of sustained force. (**D**) Integrated force (solid bar) and iliopsoas EMG (open bar) during sustained contraction from Patients A45, B07 and A53 as shown in **C**. Electrode representation denotes the stimulation configuration used by Patient A45. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency was 25 Hz. Muscles, surface EMG: intercostal sixth rib (IC), vastus lateralis (VL); fine wire EMG: iliopsoas (IL). Stimulation artefact recorded over paraspinal muscles at T12 [epidural stimulation (ES) = blue trace]. FRC = force measured through load cell and non-elastic wire.

with the vastus lateralis only becoming activated after flexor activity ended. Patients B07 and A53 were also able to perform sustained leg flexion. In all cases, the integrated force was closely linked to the iliopsoas EMG amplitude (Fig. 4D). Patient A53 demonstrated a similar pattern of alternating activity between the iliopsoas and vastus lateralis as shown by the other AIS-A individual (Patient A45). Patient B13 was unable to generate

graded levels of force or a sustained effort before training and could not consistently discriminate low and medium efforts following training.

To further assess the level of control of the newly acquired ability to move in the presence of epidural stimulation the individuals were then asked to oscillate as rapidly as possible. The cycle rate for the toe was ~1 Hz for Patient B07 (Fig. 5A–C) and
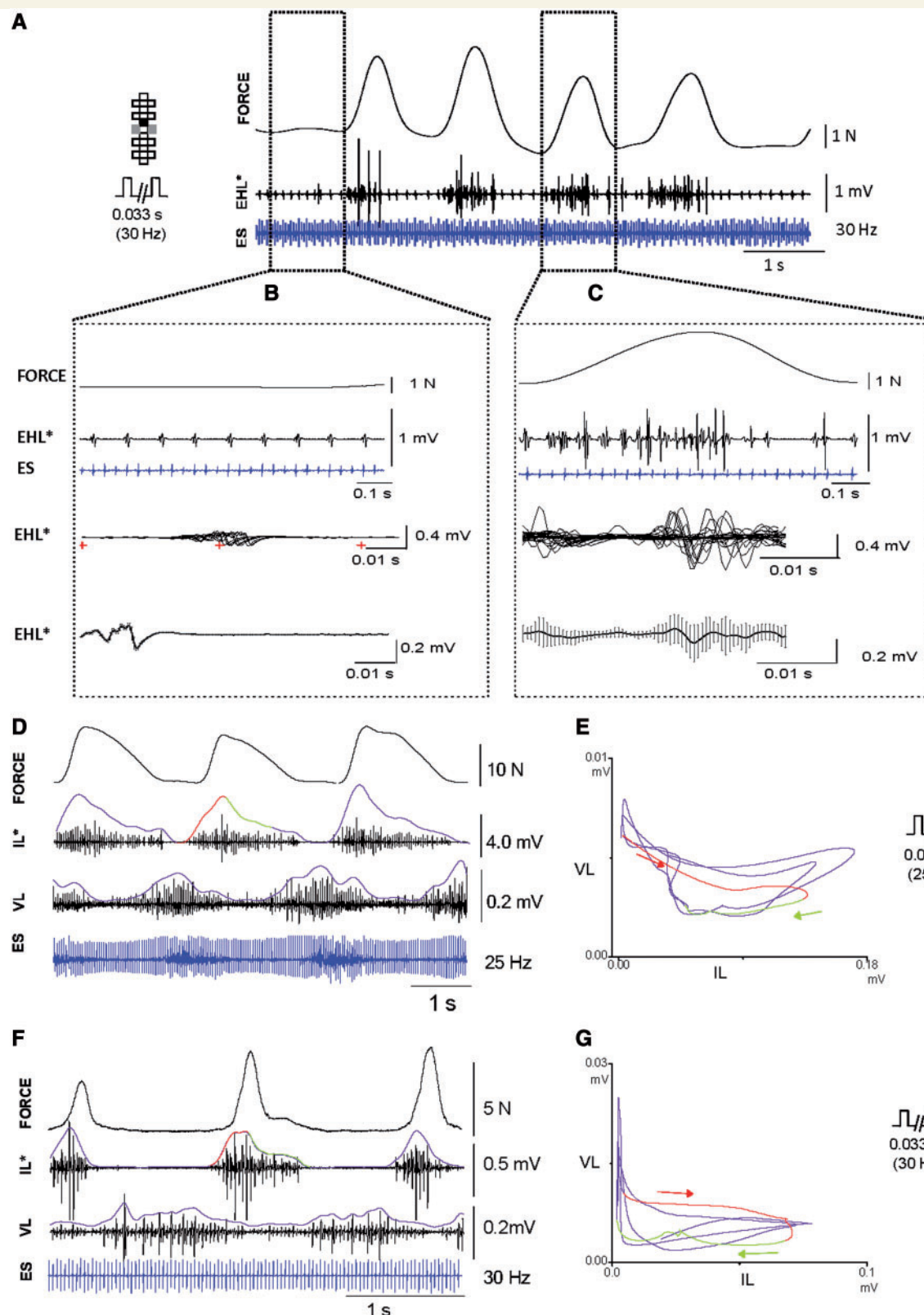
Figure 5 Rate of voluntary movements controlled by three individuals with motor complete spinal cord injury. (**A**) Force and extensor hallucis longus (EHL) EMG activity during fast voluntary first toe flexion/extension against a compliant resistance from Patient B07 from a single attempt. (**B**) A 1-s sweep from **A** before initiation of force generation encompassed by the first dashed vertical box. Ten 10 ms traces of EHL are overlaid every 0.1 s (*bottom* panel). The red crosses represent the timing of the stimulation artefact. (**C**) A 1-s period (**A**) during one cycle of force generation encompassed by the second dashed vertical box. Traces of EHL are shown as an overlay of 31 responses marked relative to the stimulation stimulus (*bottom* panel). (**D** and **F**) Force (black line) and iliopsoas and vastus lateralis EMG activity

(continued)

~0.4 Hz for Patient A45 and 0.85 Hz for Patient A53 when oscillating the whole leg (Fig. 5D–G, Supplementary Video 4). Clonic activity (6.5 Hz) of the extensor hallicus longus was not linked to the stimulation frequency (30 Hz) and occurred throughout the test in Patient B07 (Fig. 5B). EMG responses were initiated just before there was a rise in force (Fig. 5C). There was no periodic bursting of EMG activity within the timeframe of a single force effort (Fig. 5C). As noted earlier Patients A45 and A53 had reciprocity in the modulation of the EMG of the iliopsoas and the vastus lateralis during leg flexion and extension (Fig. 5D–G). In Fig. 5D, one of the oscillatory cycles has been marked in red and green, which denote the modulation of the amplitude of the EMG of the iliopsoas and vastus lateralis. Figure 5E shows the relative modulation of the hip flexor and knee extensor, and also highlights the one oscillation with flexion in red and extension in green (Patient A45). Patient A53 showed a similar modulation of the iliopsoas and vastus lateralis during multiple oscillatory cycles of leg flexion (Fig. 5F–G). After home-based training Patient B07 was able to generate similar reciprocal activation of flexors and extensors between the iliopsoas and vastus lateralis, but only during single attempts. Patient B13 was not able to generate different levels of force, oscillations or sustain the contractions upon request at his initial testing. His level of clonus and spasticity throughout the day was markedly higher than the other individuals.

Visual and auditory processing

We also assessed whether volitional control could modulate the level of activation of the appropriate motor pools based on visual and/or auditory input. All individuals could modulate the motor tasks according to visual and auditory cues. The individuals were asked to synchronize the flexion of the leg, dorsiflexion of the ankle and extension of the toe according to the rise and fall of a sine wave displayed on a computer screen. The individuals were able to consistently activate the appropriate muscles for the specified action with temporally synchronized force generation (data not shown). We compared the ability of the four individuals to modulate the flexion of the leg to a visual cue during optimal stimulation to three volume levels (60, 70 and 80 dB) of a similar auditory cue (Fig. 6). Individuals were asked to generate leg flexion in response to the onset of a 0.25 Hz auditory signal with the level of force to be generated to correspond to the amplitude of the tone. Data shown in Fig. 6 are for the following time points, T3: Patients B07 and A45; T2: Patient B13; and T1: Patient A53 (Supplementary Table 1 and Fig. 2). Three of four individuals were able to discriminate between sound amplitudes, although the

differentiation between low and medium forces was not consistent in Patients B13 and B07. For Patients A45 and B07, EMG amplitude of the iliopsoas and the adductor for the high volume was comparable with the EMG amplitudes generated when the force was modulated by a visual cue using the same stimulation parameters. The results demonstrate that auditory and visual cues were processed by the sensorimotor cortex so that the appropriate spinal interneuronal systems below the level of injury enabling the subject to titrate the desired level of excitability of the correct motor pools for the intended movement.

Effects of repetitive training on voluntary performance

Daily training using epidural stimulation with stand training and home-based voluntary training with epidural stimulation resulted in the generation of voluntary efforts with higher forces and lower stimulation voltages to reach the thresholds that enabled voluntary motor responses in two individuals (Patients B07 and B13). After 28 weeks of home-based stand and voluntary training with epidural stimulation in Patient B07, the stimulation threshold for force generation was lower. The threshold intensity was further reduced after an additional 12 weeks of training (Fig. 7A). Similar results were observed in both legs and during toe extension for Patient B07 (data not shown). Following stand training and home-based voluntary training (Fig. 2), Patient B13 showed a similar trend of lower threshold for force generation even achieving movement with no stimulation. With continued home-based voluntary training and at the conclusion of step training, Patient B13 maintained the ability to perform leg flexion with no stimulation. The peak force to stimulation strength relationship for Patient B13 post step training was characteristic of a parabolic function. Stimulation amplitudes between 0.5 V and 1.5 V resulted in force values lower than those obtained when moving with no stimulation. From 1.5 V to 2.5 V, force increased linearly with increased intensity. In the case of Patient A45, although a force increase was not observed at lower stimulation voltages following training, improvements in the accuracy to match the oscilloscope signal were observed (Fig. 7B).

Figure 7B compares the ability of all subjects to match force generation during leg flexion to a visual cue at T1 (top panel), T2 (middle panel) and T3 (bottom panel). Patient A45 showed the greatest accuracy improvement matching the force generation to the visual cue (Fig. 7B). This accuracy was demonstrated both while responding to a visual cue, as well as to an auditory cue. Patient A53 is currently undergoing step training, therefore T3 data have not been collected. These results demonstrate the ability

Figure 5 Continued

during fast voluntary whole leg flexion/extension against a compliant resistance from Patients A45 and A53, respectively. The linear envelope of the EMG signals (purple line, filter: second-order Butterworth 500–100 Hz) is shown over the raw signal. (**E** and **G**) Plot of linear envelope of the vastus lateralis versus iliopsoas from **D** and **F**. Red over the linear envelopes represents the flexion phase while green represents the extension phase of one cycle of the movement. Electrode representation for each subject denotes the stimulation configuration used. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency varied from 25 to 30 Hz. Muscles, surface EMG: vastus lateralis (VL); *fine wire EMG: iliopsoas (IL), extensor hallucis longus (EHL). Stimulation artefact recorded over paraspinal muscles at T12 [epidural stimulation (ES) = blue trace].
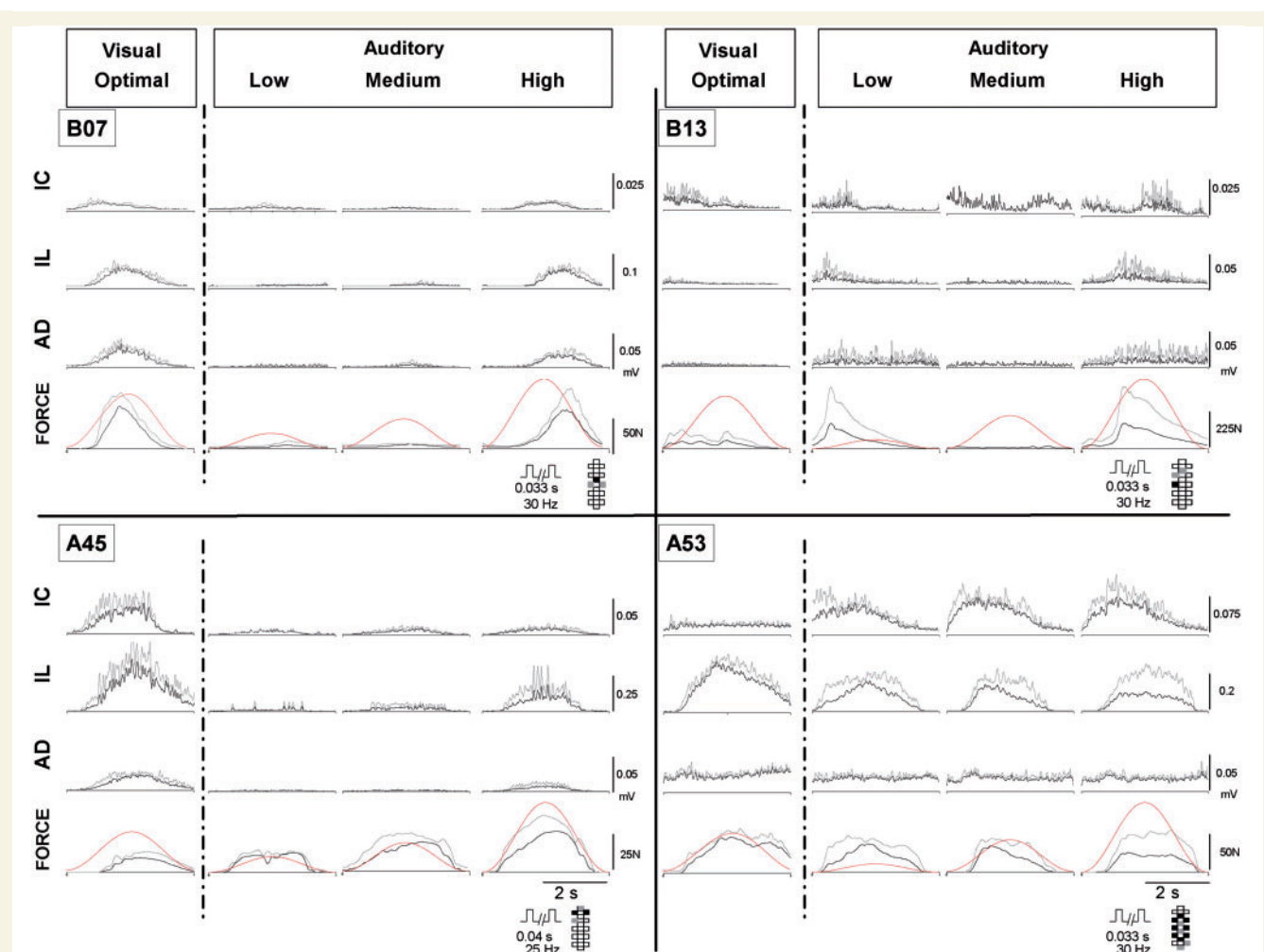
Figure 6 Voluntary movement with epidural stimulation performed in response to visual and auditory cues in four individuals with motor complete spinal cord injury. Averaged linear envelopes (filter: Winter Butterworth low-pass 2 Hz) of EMG activity and force (FORCE) generated from three trials. The left panel of each participant represents whole leg flexion/extension in response to a visual cue during optimal stimulation. Black line represents the mean signal and grey line indicates 1 SD (standard deviation) about the mean. The red line represents the oscilloscope signal which served as the visual cue. The right panel of each participant represents whole leg flexion in response three different volumes of an auditory cue during optimal stimulation. Black line represents the mean signal and grey line indicates one standard deviation about the mean. The red line represents the oscilloscope signal that matched the auditory volume cue. Stimulation parameters and voltages for the visual and auditory attempts were the same for each subject. Electrode representation for each subject denotes the stimulation configuration used. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency varied from 25 to 30 Hz. Muscles, surface EMG:intercostal sixth rib (IC), adductor magnus (AD); fine wire EMG: iliopsoas (IL).

of the spinal networks to learn with task-specific training and improve motor pool recruitment to promote force generation and accuracy. Another representation of the volitional contributions in a more task-specific activity was demonstrated by Patients A45 and A53 while stepping on the treadmill with body weight support and manual assistance, in the presence of epidural stimulation (Fig. 8). During stepping, Patient A45 was able to modulate the amount and pattern of EMG activity of lower extremity muscles when consciously thinking about stepping and moving the legs through the step cycle (Fig. 8A). Amplitude and burst duration of the flexors and extensors (bilaterally) were increased, extensors of the left knee and bilateral ankles were also modulated by the intent. Linear envelope plots of flexors and extensors show

a general increase in amplitude although the coordination was not changed (Fig. 8B). Similarly, Patient A53 was able to modulate the amplitude of EMG of flexors and extensors during the step cycle (Fig. 8C). Both participants were able to have a greater modulation of the flexor groups as compared to the extensors. This is consistent with their voluntary activity practice as all participants regained the ability to perform flexion tasks with a greater difficulty performing active extension. This ability to modulate EMG with intent during stepping was only seen in two of three individuals. Because of the fact that this observation was performed after the completion of all laboratory training for Patient B07, we never tested his ability to modulate motor output during stepping. Patient B13 was not able to show this ability during stepping;
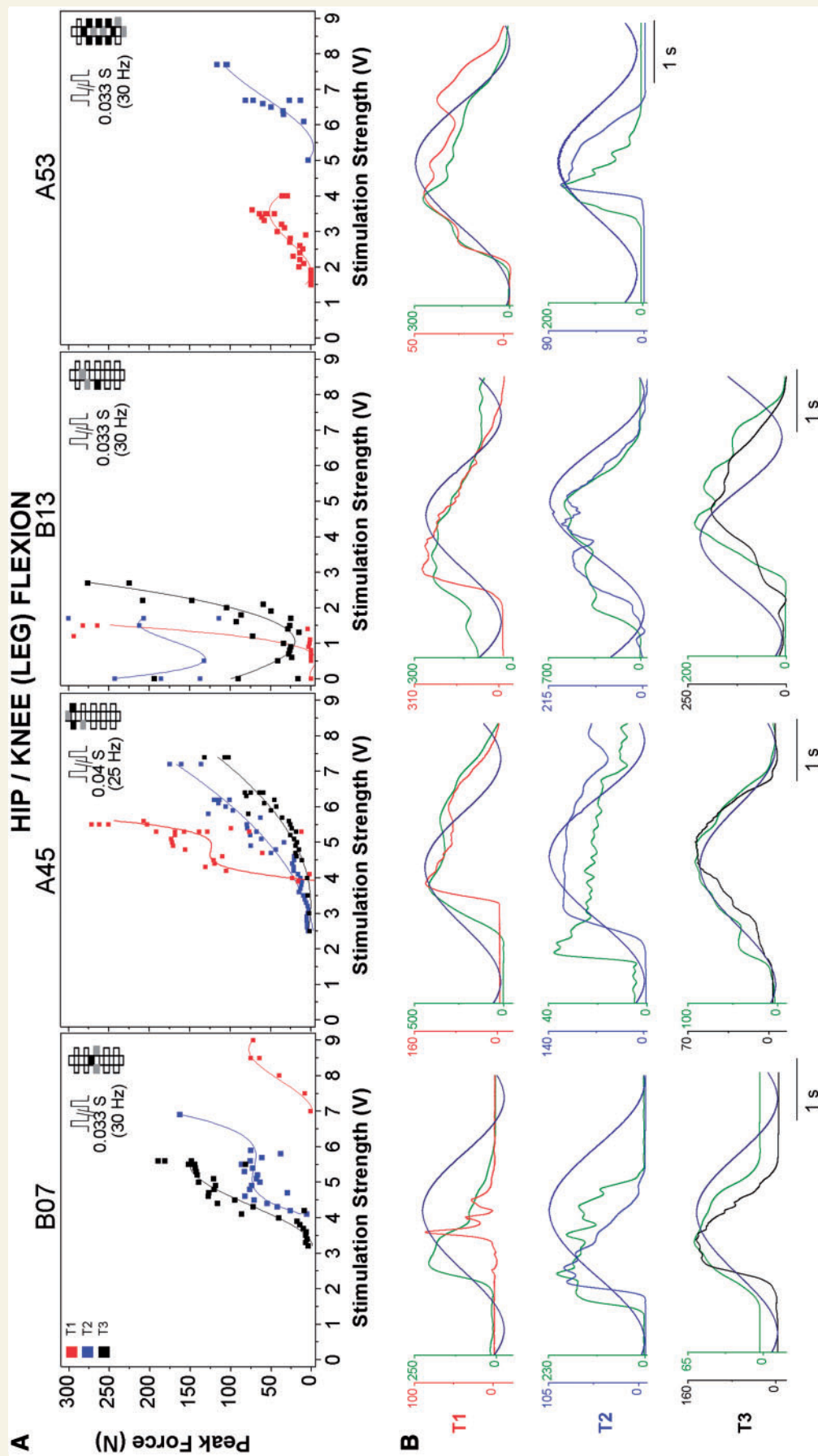
Figure 7 Stimulation voltage to force relationship during voluntary movement with epidural stimulation in three individuals with motor complete spinal cord injury. (**A**) Relationship between stimulation strength (V) and peak force (N) during hip/knee flexion for three time points in Patients B07, A45, B13 and A53 (only two time points tested). Cubic line of best fit is shown. Electrode representation for each subject denotes the stimulation configuration used. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency varied from 25 to 30 Hz. (**B**) *Top*: overlap of linear envelopes for IL (filter: Winter Butterworth low-pass 2 Hz) (green), force generation (red) and oscilloscope signal (dark blue) for first time point (T1 = red) performed at optimal voltage. *Middle*: overlap of linear envelopes (filter: Winter Butterworth low-pass 2 Hz) (green), force generation (black) and oscilloscope signal (dark blue) for post stand training (T2 = blue) performed at optimal voltage. *Bottom*: overlap of linear envelopes (filter: Winter Butterworth low-pass 2 Hz) (green), force generation (black) and oscilloscope signal (black) and oscilloscope signal (dark blue) for last time point (T3 = black) performed at optimal voltage. Attempts show improvements in accuracy of force generation during hip/knee flexion following a visual cue.
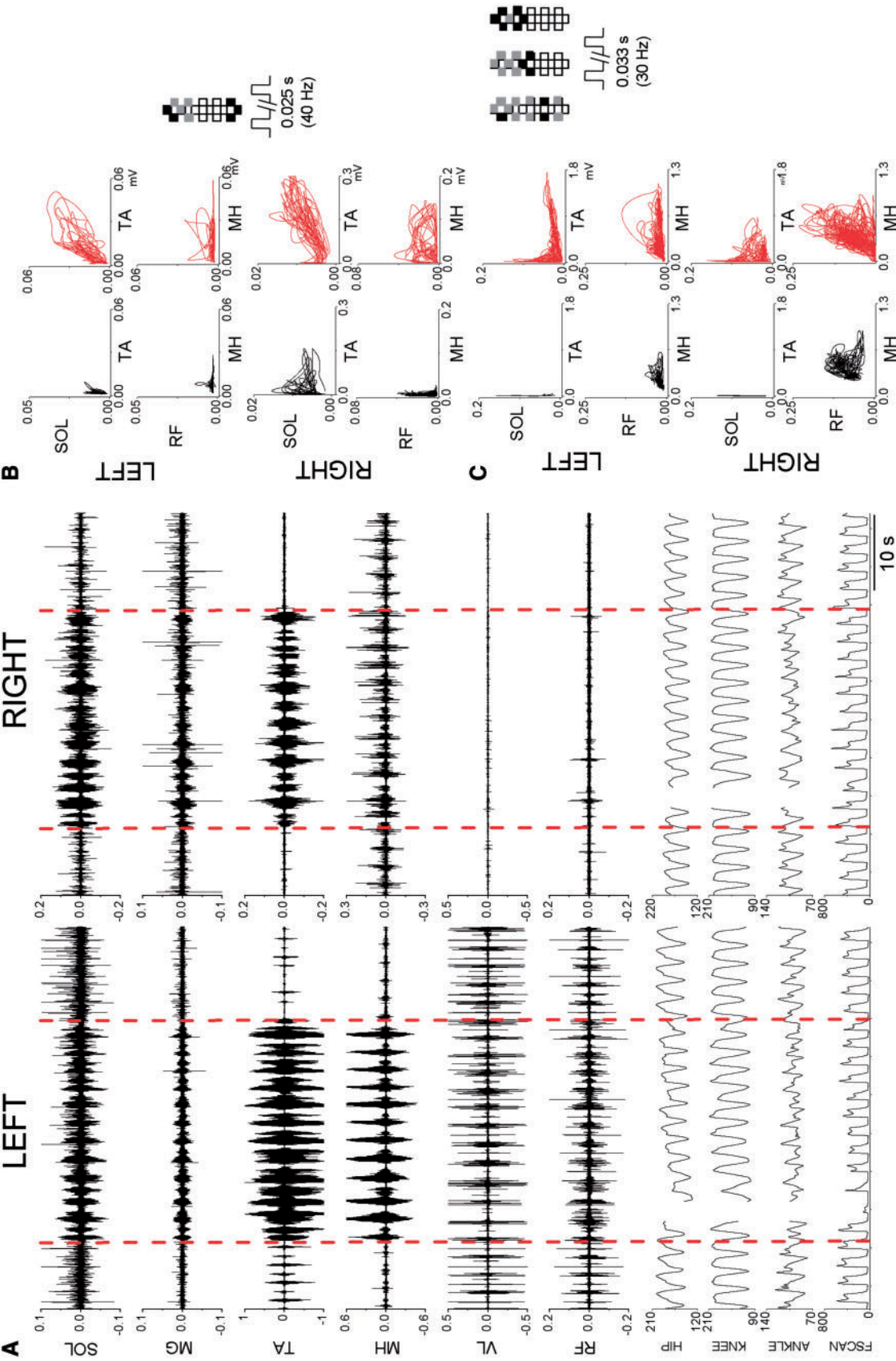
Figure 8 Modulation in EMG activity during volitional assistance during stepping with epidural stimulation by an individual with motor complete spinal cord. Coordination and amplitude of EMG was modified by the intent to step. (**A**) Left and right activity during continuous stepping (40% body weight support, 1.07 m/s) with epidural stimulation and manual assistance. Initial steps show EMG pattern while the subject (Patient A45) is not thinking about stepping. Section within the red dashed lines show the period of steps while the subject is consciously thinking about stepping and facilitating each step (with voluntary intent). (**B**) Plot of linear envelope of the EMG signals of the soleus (SOL) versus tibialis anterior (TA, *top*) and rectus femoris (RF) medial hamstrings (MH, *bottom*) (filter: second-order Butterworth 500–100 Hz). Black plots represent both the steps before and after the voluntary intent to assist; red plots represent all steps within the dashed lines during voluntary intent to assist from **A**. (**C**) Similar data from Patient A53 during continuous stepping (40% body weight support, 0.36 m/s) with epidural stimulation and manual assistance. Plot of linear envelope of the EMG signals of the soleus versus tibialis anterior (*top*) and rectus femoris versus medial hamstrings (*bottom*) (filter: second-order Butterworth 500–100 Hz). Black plots represent both the steps before and after the voluntary intent to assist, while red plots represent steps in which Patient A53 is consciously thinking about stepping and facilitating each step. Electrode representation denotes the stimulation configuration used. Grey boxes are cathodes and black boxes are anodes, white boxes are inactive electrodes. Stimulation frequency was 40 Hz for Patient A45 and 30 Hz for Patient A53. Note Patient A53 was using three interleaved configurations. Muscles: soleus (SOL), medial gastrocnemius (MG), tibialis anterior (TA), medial hamstrings (MH), vastus lateralis (VL), and rectus femoris (RF). EMG data are presented in mV. Hip, knee and ankle are sagittal joint angles (degrees). The break in kinematic data is the result of a brief interruption in the recording. Ground reaction forces (FSCAN) reflect stance and swing phase (N).

this might be the result of the higher sensitivity to stimulation and greater difficulty in performing multiple repetitions of volitional activity.

Discussion

This study demonstrates the ability of four individuals with chronic complete motor paralysis to execute voluntary tasks with selectivity of appropriate motor pools in the presence of epidural stimulation. High fidelity sensorimotor translation of visual and auditory signals were processed to control the timing and amount of force generated during the movements. In three of four individuals, we observed the recovery of voluntary movement with epidural stimulation soon after implantation, two of whom had complete loss of both motor and sensory function (Supplementary Videos 1–3). This shows that by neuromodulating the spinal circuitry at sub-threshold motor levels with epidural stimulation, chronically complete paralysed individuals can process conceptual, auditory, and visual input to regain specific voluntary control of paralysed muscles. We have uncovered a fundamentally new intervention strategy that can dramatically affect recovery of voluntary movement in individuals with complete paralysis even years after injury.

The results in the three individuals who were tested after implantation, but before repetitive training, suggests that descending connections to the spinal cord circuitry may have existed since the time of injury. Patient B07 did not show voluntary ability until after 7 months of epidural stimulation and stand training, from an attempt he initiated. The initial study protocol did not include attempted voluntary movements so we cannot verify whether movement was possible before training. However, Patients A45, B13 and A53 were able to voluntarily execute movements after 11, 4 and 7 days of epidural stimulation, respectively (Supplementary Table 1). Anatomical connections may have persisted after the injury that were previously 'silent' because of loss of conduction as a result of disruption of myelin or the ionic channels of the neurons (Waxman, 1989; Fehlings and Nashmi, 1996; Shi and Blight, 1997; Sinha *et al.*, 2006; Coggan *et al.*, 2011). In our study, these individuals initially were able to elicit these intentional movements only with the epidural stimulation indicating that the limited influence of the available supraspinal connections was not sufficient to activate motor pools. As demonstrated in Fig. 1B and D, transcranial magnetic stimulation did not show any changes in excitation even when the individuals were requested to attempt to actively dorsiflex the ankle at the time the transcranial magnetic stimulation pulse was delivered. This suggests that the alteration of the spinal cord circuitry with epidural stimulation was enhancing the central excitatory drive to the motor neurons. Conceivable through activated lumbosacral interneurons, since before the intent of the movement the motor pools were not active (Angel *et al.*, 1996, 2005; Djouhri and Jankowska, 1998; Edgley *et al.*, 2004; Cabaj *et al.*, 2006; Bannatyne *et al.*, 2009). The somatosensory evoked potentials results in the two AIS-B individuals showing a shorter latency of responses after training may suggest improved somatosensory transmission promoted by repetitive epidural stimulation.

The resting general excitability state of the spinal circuitry was different among the four individuals that participated in this study (Supplementary Fig. 2). There were differences in stimulation intensity thresholds seen across individuals for the generation of movement (Fig. 7). Patients B07 and A45 required higher levels of stimulation to initiate movement and had relatively low levels of clonus and spasticity throughout the day. In contrast, Patients B13 and A53, the more spontaneously active individuals who reported more clonus and spasticity, could initiate movement at lower thresholds of stimulation. We propose that the functional state of spinal network excitability of interneurons (Bannatyne *et al.*, 2009) and motor neurons was modulated by the epidural stimulation, presumably driving them closer to their appropriate activation threshold, enabling intentional movement (Quevedo *et al.*, 2005; Berg *et al.*, 2007; Yarom and Hounsgaard, 2011). Thus, loss of voluntary control of movement may be attributed to not only a physical disruption of descending connections, but also to a physiological alteration of the central state of excitability of the spinal circuitry (Edgerton *et al.*, 1997; de Leon *et al.*, 1999; Tillakaratne *et al.*, 2002). These findings have important implications regarding the significance of sub-motor threshold modulation of spinal circuitry as an important factor in motor control in the uninjured as well as injured spinal cord.

The recovery of the volitional motor drive in four individuals diagnosed with complete motor paralysis (Fig. 3) could have been facilitated by lumbosacral spinal cord epidural stimulation through transmission of rostro-caudal signal propagation through the propriospinal interneuronal projections (Zaporozhets *et al.*, 2006, 2011; Courtine *et al.*, 2008; Cowley *et al.*, 2008; Flynn *et al.*, 2011). We assessed the individuals' ability to execute a movement on command by comparing the EMG and force patterns with a sine wave pattern on a computer screen (Fig. 7B). In general, at the first time point tested, all individuals demonstrated a delay in the initiation of the force relative to the rise of the sine wave and the force decline was earlier than the decline of the position cursor. Regardless of these time differences in initiation and termination of the effort, the patients could clearly execute the movements on command. The delay in the motor response related to the intention to move observed in each individual could be consistent with the voluntarily controlled motor commands being mediated more indirectly through descending propriospinal pathways. Although we cannot rule out a delay of processing the auditory and visual cues, there is precedence in the rat model that tonic intraspinal stimulation immediately caudal to the injury resulted in functional improvements comparable with those seen following long-distance axon regeneration (Yakovenko *et al.*, 2007). Cowley *et al.* (2010) showed that even in the absence of long direct transmission, propriospinal pathways through commissural projections can transmit a locomotor command to the lumbrosacral spinal cord with 27% success. Thus, the stimulation may facilitate excitation of propriospinal neurons which supports propagation of the voluntary command to the lumbosacral spinal cord.

It is also possible that reticulospinal neurons may mediate ipsilateral corticospinal tract relay through commissural interneurons. Studies have shown that commissural interneurons activated by reticulospinal neurons have direct contacts with interneurons mediating reflex actions from Group Ib tendon organ afferents and

Group II muscle spindle afferents and modulate actions of these interneurons. (Edgley *et al.*, 2004; Cabaj *et al.*, 2006).The strengthening of reticulospinal connections to motor neurons was demonstrated in monkeys following a unilateral corticospinal tract lesion (Zaaimi *et al.*, 2012). Future studies are needed to probe pathways including propriospinal, corticospinal, reticulospinal and vestibulospinal, that have residual connectivity and can support the propagation of signals.

In all four research participants, their ability to voluntarily move improved over time with daily epidural stimulation and voluntary training while also receiving stand or step training. Figure 7 shows the effect of training on the ability to generate higher forces with similar stimulation intensities as well as the ability to more accurately match a visual cue. These results demonstrate the ability of the spinal networks to learn with task-specific training and improve motor pool recruitment to promote force generation and accuracy. Conceivably, after repetitive epidural stimulation and training, plasticity of these disrupted pathways could have resulted in a more functional state. The newly established functional connectivity presumably involves multiple, novel neuronal pathways and synapses (Raineteau *et al.*, 2002). Exercise training after spinal cord injury in a rodent model has been shown to enhance corticospinal tract sprouting and increased functional connections to spinal neurons (Bareyre *et al.*, 2004; Engesser-Cesar *et al.*, 2007; Courtine *et al.*, 2008; Goldshmit *et al.*, 2008; Flynn *et al.*, 2013). Similarly it is conceivable that sprouting or growth of axons across the lesion occurred in response to repetitive epidural stimulation and/or stand training. Axonal sprouting is unlikely as the mechanism for early execution of the voluntary movement in three of four individuals studied here.

This study demonstrates that individuals diagnosed as clinically motor complete can develop functional connectivity across the lesion in the presence of epidural stimulation. This emphasizes the importance of resolving the uncertainty associated with classifying a patient as clinically complete and then determining that no recovery is possible (Waters *et al.*, 1998; Ditunno, Jr, 1999; Burns *et al.*, 2003; Calancie *et al.*, 2004a; Kakulas, 2004). Although the exact mechanisms that enabled these surprising results cannot be definitively identified, the possibility of re-establishing functionally meaningful voluntary control in the presence of epidural stimulation places a high priority on reconsideration of the mechanisms contributing to paralysis in humans. These results indicate that epidural stimulation with activity-dependent plasticity can be used to develop effective therapeutic interventions for recovery of movement in individuals with chronic complete motor paralysis.

The acquired ability to have fine motor control and fidelity of movement in the presence of epidural stimulation currently has limitations when attempting to translate functional benefit to daily life. The lack of a usable interactive patient interface with embedded control algorithms makes the movements as used before injury limited. However, all four of these individuals have found unique ways to incorporate their ability to move their trunk and legs into daily activities. For example, they use their trunk and leg epidural stimulation configurations during physical workouts and facets of their daily life allowing them to be more stable, pursue activities for longer periods of time and they report greater strength and less fatigue. We are following these reported activities to guide the design of the control algorithms and patient interfaces. Future experiments with improved technology are needed expediently to take the most advantage functionally of these neurophysiological findings in people after severe spinal cord injury.

Supplementary material

Supplementary material is available at *Brain* online.

References

Angel MJ, Guertin P, Jimenez I, McCrea DA. Group I extensor afferents evoke disynaptic EPSPs in cat hindlimb extensor motorneurones during fictive locomotion. J Physiol 1996; 494: 851–61.

Angel MJ, Jankowska E, McCrea DA. Candidate interneurones mediating group I disynaptic EPSPs in extensor motoneurones during fictive locomotion in the cat. J Physiol 2005; 563: 597–610.

Bannatyne BA, Liu TT, Hammar I, Stecina K, Jankowska E, Maxwell DJ. Excitatory and inhibitory intermediate zone interneurons in pathways from feline group I and II afferents: differences in axonal projections and input. J Physiol 2009; 587: 379–99.

Bareyre FM, Kerschensteiner M, Raineteau O, Mettenleiter TC, Weinmann O, Schwab ME. The injured spinal cord spontaneously forms a new intraspinal circuit in adult rats. Nat Neurosci 2004; 7: 269–77.

Beres-Jones JA, Johnson TD, Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. Exp Brain Res 2003; 149: 222–36.

Berg RW, Alaburda A, Hounsgaard J. Balanced inhibition and excitation drive spike activity in spinal half-centers. Science 2007; 315: 390–3.

Burns AS, Lee BS, Ditunno JF Jr, Tessler A. Patient selection for clinical trials: the reliability of the early spinal cord injury examination. J Neurotrauma 2003; 20: 477–82.

Cabaj A, Stecina K, Jankowska E. Same spinal interneurons mediate reflex actions of group Ib and group II afferents and crossed reticulospinal actions. J Neurophysiol 2006; 95: 3911–22.

Calancie B, Molano MR, Broton JG. Abductor hallucis for monitoring lower-limb recovery after spinal cord injury in man. Spinal Cord 2004a; 42: 573–80.

Calancie B, Molano MR, Broton JG. EMG for assessing the recovery of voluntary movement after acute spinal cord injury in man. Clin Neurophysiol 2004b; 115: 1748–59.

Coggan JS, Ocker GK, Sejnowski TJ, Prescott SA. Explaining pathological changes in axonal excitability through dynamical analysis of conductance-based models. J Neural Eng 2011; 8: 065002.

Courtine G, Song B, Roy RR, Zhong H, Herrmann JE, Ao Y, et al. Recovery of supraspinal control of stepping via indirect propriospinal relay connections after spinal cord injury. Nat Med 2008; 14: 69–74.

Cowley KC, Zaporozhets E, Schmidt BJ. Propriospinal neurons are sufficient for bulbospinal transmission of the locomotor command signal in the neonatal rat spinal cord. J Physiol 2008; 586: 1623–35.

Cowley KC, Zaporozhets E, Schmidt BJ. Propriospinal transmission of the locomotor command signal in the neonatal rat. Ann N Y Acad Sci 2010; 1198: 42–53.

David S, Aguayo A. Axonal elongation into peripheral nervous system "bridges" after central nervous system injury in adult rats. Science 1981; 214: 931–3.

de Leon RD, Tamaki H, Hodgson JA, Roy RR, Edgerton VR. Hindlimb locomotor and postural training modulates glycinergic inhibition in the spinal cord of the adult spinal cat. J Neurophysiol 1999; 82: 359–69.

Dimitrijevic MR. Residual motor functions in spinal cord injury. Adv Neurol 1988; 47: 138–55.

Dimitrijevic MR. Motor control in chronic spinal cord injury patients. Scand J Rehabil Med Suppl 1994; 30: 53–62.

Dimitrijevic MR, Dimitrijevic MM, Faganel J, Sherwood AM. Suprasegmentally induced motor unit activity in paralyzed muscles of patients with established spinal cord injury. Ann Neurol 1984; 16: 216–21.

Dimitrijevic MR, Faganel J, Lehmkuhl D, Sherwood A. Motor control in man after partial or complete spinal cord injury. Adv Neurol 1983a; 39: 915–26.

Dimitrijevic MR, Hsu CY, McKay WB. Neurophysiological assessment of spinal cord and head injury. J Neurotrauma 1992b; 9: S293–300.

Dimitrijevic MR, Kofler M, McKay WB, Sherwood AM, Van der LC, Lissens MA. Early and late lower limb motor evoked potentials elicited by transcranial magnetic motor cortex stimulation. Electroencephalogr Clin Neurophysiol 1992a; 85: 365–73.

Dimitrijevic MR, Prevec TS, Sherwood AM. Somatosensory perception and cortical evoked potentials in established paraplegia. J Neurol Sci 1983b; 60: 253–65.

Ditunno JF Jr. The John Stanley Coulter lecture. Predicting recovery after spinal cord injury: a rehabilitation imperative. Arch Phys Med Rehabil 1999; 80: 361–4.

Djouhri L, Jankowska E. Indications for coupling between feline spinocervical tract neurones and midlumbar interneurones. Exp Brain Res 1998; 119: 39–46.

Edgerton VR, de Leon RD, Tillakaratne N, Recktenwald M R, Hodgson J A, Roy RR. Use-dependent plasticity in spinal stepping and standing. In: Seil FJ, editor. Advances in neurology. Vol. 72. Philadelphia: Lippincott-Raven Publishers; 1997. p. 233–47.

Edgley SA, Jankowska E, Hammar I. Ipsilateral actions of feline corticospinal tract neurons on limb motoneurons. J Neurosci 2004; 24: 7804–13.

Engesser-Cesar C, Ichiyama RM, Nefas AL, Hill MA, Edgerton VR, Cotman CW, et al. Wheel running following spinal cord injury improves locomotor recovery and stimulates serotonergic fiber growth. Eur J Neurosci 2007; 25: 1931–9.

Fehlings MG, Nashmi R. Changes in pharmacological sensitivity of the spinal cord to potassium channel blockers following acute spinal cord injury. Brain Res 1996; 736: 135–45.

Flynn JR, Dunn LR, Galea MP, Callister R, Callister RJ, Rank MM. Exercise training after spinal cord injury selectively alters synaptic properties in neurons in adult mouse spinal cord. J Neurotrauma 2013; 30: 891–6.

Flynn JR, Graham BA, Galea MP, Callister RJ. The role of propriospinal interneurons in recovery from spinal cord injury. Neuropharmacology 2011; 60: 809–22.

Goldshmit Y, Lythgo N, Galea MP, Turnley AM. Treadmill training after spinal cord hemisection in mice promotes axonal sprouting and synapse formation and improves motor recovery. J Neurotrauma 2008; 25: 449–65.

Harkema S, Gerasimenko Y, Hodes J, Burdick J, Angeli C, Chen Y, et al. Effect of epidural stimulation of the lumbosacral spinal cord on voluntary movement, standing, and assisted stepping after motor complete paraplegia: a case study. Lancet 2011a; 377: 1938–47.

Harkema SJ, Behrman AL, Barbeau H. Locomotor training: principles and practice. New York: Oxford University Press; 2011b.

Harkema SJ, Hillyer J, Schmidt-Read M, Ardolino E, Sisto S, Behrman A. Locomotor training: as a treatment of spinal cord injury and in the progression of neurological rehabilitation. Arch Phys Med Rehabil 2012a; 93: 1588–97.

Harkema SJ, Hurley SL, Patel UK, Requejo PS, Dobkin BH, Edgerton VR. Human lumbosacral spinal cord interprets loading during stepping. J Neurophysiol 1997; 77: 797–811.

Harkema SJ, Schmidt-Read M, Lorenz DJ, Edgerton VR, Behrman AL. Balance and ambulation improvements in individuals with chronic incomplete spinal cord injury using locomotor training-based rehabilitation. Arch Phys Med Rehabil 2012b; 93: 1508–17.

Kakulas BA. Neuropathology: the foundation for new treatments in spinal cord injury. Spinal Cord 2004; 42: 549–63.

Li K, Atkinson D, Boakye M, Tolfo CZ, Aslan S, Green M, et al. Quantitative and sensitive assessment of neurophysiological status after human spinal cord injury. J Neurosurg Spine 2012; 17: 77–86.

Marino RJ, Barros T, Biering-Sorensen F, Burns SP, Donovan WH, Graves DE, et al. International standards for neurological classification of spinal cord injury. J Spinal Cord Med 2003; 26: S50–6.

McKay WB, Lee DC, Lim HK, Holmes SA, Sherwood AM. Neurophysiological examination of the corticospinal system and voluntary motor control in motor-incomplete human spinal cord injury. Exp Brain Res 2005; 163: 379–87.

McKay WB, Lim HK, Priebe MM, Stokic DS, Sherwood AM. Clinical neurophysiological assessment of residual motor control in post-spinal cord injury paralysis. Neurorehabil Neural Repair 2004; 18: 144–53.

McKay WB, Ovechkin AV, Vitaz TW, Terson de Paleville DG, Harkema SJ. Neurophysiological characterization of motor recovery in acute spinal cord injury. Spinal Cord 2011; 49: 421–9.

McKay WB, Stokic DS, Dimitrijevic MR. Assessment of corticospinal function in spinal cord injury using transcranial motor cotrex stimulation: a review. J Neurotrauma 1997; 14: 539–48.

Perot PL, Vera CL. Scalp-recorded somatosensory evoked potentials to stimulation of nerves in the lower extremities and evaluation of patients with spinal cord trauma. Ann N Y Acad Sci 1982; 388: 359–68.

Quevedo J, Stecina K, Gosgnach S, McCrea DA. Stumbling corrective reaction during fictive locomotion in the cat. J Neurophysiol 2005; 94: 2045–52.

Raineteau O, Fouad K, Bareyre FM, Schwab ME. Reorganization of descending motor tracts in the rat spinal cord. Eur J Neurosci 2002; 16: 1761–71.

Richardson PM, McGuinnes UM, Aguayo AJ. Axons from CNS neurones regenerate into PNS grafts. Nature 1980; 284: 264–5.

Sherwood AM, Dimitrijevic MR, McKay WB. Evidence of subclinical brain influence in clinically complete spinal cord injury: discomplete SCI. J Neurol Sci 1992; 110: 90–8.

Shi R, Blight AR. Differential effects of low and high concentrations of 4-aminopyridine on axonal conduction in normal and injured spinal cord. Neuroscience 1997; 77: 55–62.

Sinha K, Karimi-Abdolrezaee S, Velumian AA, Fehlings MG. Functional changes in genetically dysmyelinated spinal cord axons of shiverer mice: role of juxtaparanodal Kv1 family K+ channels. J Neurophysiol 2006; 95: 1683–95.

Tillakaratne NJ, de Leon RD, Hoang TX, Roy RR, Edgerton VR, Tobin AJ. Use-dependent modulation of inhibitory capacity in the feline lumbar spinal cord. J Neurosci 2002; 22: 3130–43.

Waring WP, Biering-Sorensen F, Burns S, Donovan W, Graves D, Jha A, et al. 2009 review and revisions of the international standards for the neurological classification of spinal cord injury. J Spinal Cord Med 2010; 33: 346–52.

Waters RL, Adkins R, Yakura J, Sie I. Donal Munro Lecture: functional and neurologic recovery following acute SCI. J Spinal Cord Med 1998; 21: 195–9.

Waters RL, Yakura JS, Adkins RH, Sie IH. Recovery following complete paraplegia. Arch Phys Med Rehabil 1992; 73: 784–9.

Waxman SG. Demyelination in spinal cord injury. J Neurol Sci 1989; 91: 1–14.

Yakovenko S, Kowalczewski J, Prochazka A. Intraspinal stimulation caudal to spinal cord transections in rats. Testing the propriospinal hypothesis. J Neurophysiol 2007; 97: 2570–4.

Yarom Y, Hounsgaard J. Voltage fluctuations in neurons: signal or noise? Physiol Rev 2011; 91: 917–29.

Zaaimi B, Edgley SA, Soteropoulos DS, Baker SN. Chages in descending motor pathway connectivity after corticospinal tract lesion in macaque monkey. Brain 2012; 135: 2277–89.

Zaporozhets E, Cowley KC, Schmidt BJ. Propriospinal neurons contribute to bulbospinal transmission of the locomotor command signal in the neonatal rat spinal cord. J Physiol 2006; 572: 443–58.

Zaporozhets E, Cowley KC, Schmidt BJ. Neurochemical excitation of propriospinal neurons facilitates locomotor command signal transmission in the lesioned spinal cord. J Neurophysiol 2011; 105: 2818–29.

Zhao RR, Andrews MR, Wang D, Warren P, Gullo M, Schnell L, et al. Combination treatment with anti-Nogo-A and chondroitinase ABC is more effective than single treatments at enhancing functional recovery after spinal cord injury. Eur J Neurosci 2013; 2946–61.