Temporal components of the motor patterns expressed by the human spinal cord reflect foot kinematics

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Running title: Locomotion encryptions in the human spinal cord

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What are the building blocks with which the human spinal cord constructs the motor patterns of locomotion? In principle they could correspond to each individual activity pattern in dozens of different muscles. Alternatively, there could exist a small set of constituent temporal components that are common to all activation patterns and reflect global kinematic goals. To address this issue, we studied patients with spinal injury trained to step on a treadmill with body-weight-support. Patients learned to produce foot kinematics similar to that of healthy subjects, but with activity patterns of individual muscles generally different from the control group. Hidden in the muscle patterns we found a basic set of five temporal components, whose flexible combination accounted for the wide range of muscle patterns recorded in both controls and patients. Furthermore, two of the components were systematically related to foot kinematics across different stepping speeds and loading conditions. We suggest that the components are related to control signals output by spinal pattern generators, normally under the influence of descending and afferent inputs.

**Keywords:**

Muscle synergy, locomotion, spinal cord injured patients, factor analysis, central pattern generator
Introduction

Recent evidence indicates that movement may not be controlled muscle-by-muscle, but the activity of many muscles is coordinated to produce global goals at the endpoint (hand or foot, Bizzi et al. 2000; Georgopoulos and Grillner 1989; Krebs et al. 1999; Lacquaniti et al. 1999). One hypothesis is that movements mediated by the spinal cord are based on the flexible combination of a small number of functional units, although the exact definition of such units remains controversial (Barbeau et al. 1999b; Bizzi et al. 2000; Grillner 1981; Hultborn 2001; Lacquaniti et al. 1999; Orlovsky et al. 1999; Stein et al. 2002). According to an influential scheme, locomotion is organized around a set of central pattern generators (Grillner 1981): unit burst generators would control a set of synergistic muscles acting around a particular joint under the modulatory influence of peripheral and descending inputs (Barbeau et al. 1999b; Grillner 1981, 2002; Orlovsky et al. 1999). Control of individual units could be coupled in many different ways to produce a wide range of behavior for the whole limb (as well as for inter-limb coordination, Grillner 1981; Orlovsky et al. 1999). Evidence for spinal circuitry underlying central pattern generation in humans is emerging from studies on spinal-cord-injured (SCI) patients (Barbeau et al. 1999b; Bussel et al. 1988; Calancie et al. 1994; Dietz 2002a; Harkema et al. 2000; Shapkova and Schomburg 2001; Wernig et al. 1995).

Also, there is growing evidence from animal studies that parameters related to endpoint motion might be encoded by populations of neurons at several different levels of integration in the CNS, including the spinal cord (Bizzi et al. 2000; Bosco and Poppele 2001; Georgopoulos and Grillner 1989). In lower vertebrates, control of the foot appears to be based on a few spinal inter-neuronal modules that can be flexibly
combined to generate global motor patterns (Bizzi et al. 1991, 2000; Kargo and Giszter 2000). A small set of basic muscle synergies has been recently identified by means of a computational analysis of the patterns of muscle activity (d’Avella et al. 2003; Tresch et al. 1999). In cats, position and direction of foot movement are encoded in dorsal spinocerebellar neurons of the spinal cord, independent of the specific combination of joint angles and level of muscle activity (Bosco and Poppele 2001). Spinalized cats can be trained to walk with normal kinematics (Belanger et al. 1996; de Leon et al. 1998). It has recently been shown that the human spinal cord interprets foot loading in locomotion (Harkema et al. 1997), but the kinematic determinants that can be expressed by the human spinal cord are poorly understood (Barbeau et al. 1999a; Dietz 2002a; Harkema et al. 2000).

Here we address the issue of what are the building blocks with which the human spinal cord constructs the motor patterns of locomotion. To this end, we studied patients with spinal injury trained to step on a treadmill with body-weight-support (BWS). We recorded kinematic and electromyographic (EMG) data in both these patients and healthy subjects. To unveil the common temporal components hidden in the profiles of muscle activity, we applied factor analysis to the simultaneous recordings of EMG activity of several limb and trunk muscles. In this analysis the basic temporal components are not specified a priori, but they are determined a posteriori by the statistical structure of the data waveforms (Glaser and Ruchkin 1976). The rationale behind this approach is that if all the muscles that are active during locomotion express combinations of the same basic temporal components, such components may be related to underlying control signals (including peripheral feedback) directed to the corresponding α-motoneuron pools. If, in addition, the basic temporal components
systematically correlate with foot kinematics, it may be hypothesized that the patterns of multiple muscle coordination reflect specific laws of kinematic control (Lacquaniti et al 1999).

Materials and Methods

Subjects

Eleven SCI-patients (age 45 ±16 yr, mean±SD) and eleven healthy age-matched subjects were studied. The injuries of the patients were mainly at the thoracic level (on average T8 ± 4 segments), as a result of trauma or ischemia. At hospital admission, before and after training, they were submitted to neurological evaluation, routine radiological and neurophysiological tests. No signs of denervation were found in the leg muscles by EMG. Patients were classified according to the ASIA (American Spinal Injury Association) impairment scale (Maynard et al. 1997). In clinical rehabilitation, ASIA-A are defined as “sensory and motor complete”, ASIA-B as “sensory incomplete and motor complete”, ASIA-C and ASIA-D as “sensory and motor incomplete”. It should be noticed that the assessment of completeness of a spinal lesion is based on clinical, radiological and neurophysiological tests indicating the absence of motor and sensory function below the injury site, but it does not necessarily imply that there are no axons that cross the injury site.

On admission, patients were classified as follows: five patients as ASIA-A (complete paraplegia, no sensory or motor function below the neurological level including S4-S5 segments), two as ASIA-B (sensory but not motor function is preserved below the neurological level), and four as ASIA-C (motor function is preserved below the neurological level and the majority of key muscles below this level have a muscle
grade of less than 3/5, that is they cannot be actively contracted against gravity). At the end of training, two ASIA-C patients were re-classified as ASIA-D (the majority of key muscles below the neurological level have a muscle grade higher than 3/5, that is they can be actively contracted against gravity), whereas the classification of the other patients did not change. Written informed consent was obtained from each subject; the experiments and training procedures were approved by the Ethics Committee of IRCCS Fondazione Santa Lucia.

**Experimental setup**

General procedures are detailed in Ivanenko et al. (2002). Subjects stepped on a treadmill (EN-MILL 3446.527, Bonte Zwolle BV, Netherlands) at different controlled speeds. BWS was obtained by suspending the subjects in a harness connected to a pneumatic device that applied a controlled upward force at the waist (Gazzani et al 2000; Ivanenko et al. 2002). The overall constant error in the applied force and dynamic force fluctuations monitored by a load cell were less than 5% of body weight (Gazzani et al 2000). Subjects were asked to place the abducted arms on horizontal rollbars located at the side of the treadmill, at breast height. Three-dimensional motion of selected body points was recorded at 100 Hz by means either of the Optotrak system (Northern Digital, Waterloo, Ontario, ± 3SD-accuracy better than 0.2 mm for x,y,z coordinates) or of 9-TV cameras Vicon-612 system (1-mm-accuracy) during about 20-100 s depending on treadmill speed. Five markers were attached on the right side of the subject to the skin overlying the following landmarks: the midpoint between the anterior and the posterior superior iliac spine (ilium, IL), greater trochanter (GT), lateral femur epicondyle (LE), lateral malleolus (LM), and fifth metatarso-phalangeal joint (VM). In
all controls and 9 patients EMG activity was recorded by means of surface electrodes from tibialis anterior TA, lateral gastrocnemius GCL, biceps femoris long head BF, rectus femoris RF, vastus lateralis VL, gluteus maximus GM, rectus abdominis middle RAM and superior portions RAS, external oblique OE, internal oblique OI, latissimus dorsi LD, erector spinae ES recorded at L1-L2, trapezius TRAP, triceps brachii TRI, and biceps brachii BIC. In 6 controls and 3 patients EMG was also recorded from peroneus longus PER, semitendinosus SE, adductor longus AL, sartorius SA, tensor fasciae latae TF, and deltoid DEL. In 2 patients EMG was only recorded from leg muscles (TA, GCL, BF, RF, VL, GM). EMG signals were preconditioned at the recording site (active electrodes from BTS, Milan, Italy or DelSys, Boston, Massachusetts), digitized, transmitted to the remote amplifier (20-Hz-high-pass and 200-Hz-low-pass filters), and sampled at 500 or 1000 Hz (synchronized with kinematic sampling).

Protocols

Patients performed daily sessions of BWS-treadmill training for 1-3 months, starting from 1-6 months after the lesion. They were assisted to step as necessary by 2 physiotherapists. Under their guidance, patients underwent progressive training with increasing treadmill speed, decreasing BWS and decreasing manual assistance from the therapists. BWS was set at 75% of body weight at the beginning of the study and was subsequently decreased by 5%-steps according to the patient’s improvement. ASIA-C/D patients could reach 0%-BWS at the end of training, whereas ASIA-A and ASIA-B patients never went below 60-75%. Treadmill speed was set at 0.7, 1, 2, and 3 km/h. In control subjects, a higher speed (5 km/h) was also recorded. During training, each
physiotherapist held one patient’s leg at the ankle to assist with swing and foot placement. Kinematic and EMG data were collected during stepping attempts performed with minimal assistance, one day before and at the end of training. During the recording sessions, patients stepped by themselves, being helped by the physiotherapists only when they stumbled.

**Data analysis**

**Kinematics**

The body was modeled as an interconnected chain of rigid segments: IL-GT for the pelvis, GT-LE for the thigh, LE-LM for the shank, and LM-VM for the foot. The elevation angle of each segment in the sagittal plane corresponds to the angle between the projected segment and the vertical. These angles are positive in the forward direction (i.e., when the distal marker is located anterior to the proximal marker). The limb axis was defined as GT-LM. Gait cycle was defined as the time between two successive maxima of the elevation angle of the limb axis. The time of maximum and minimum elevation of the limb axis corresponds to heel-contact and toe-off (stance to swing transition), respectively, in normal subjects (Bianchi et al. 1998). These time markers were used to identify stance and swing phases. In previous experiments with healthy subjects in which a force platform (Kistler 9281B) was used to monitor the contact forces during ground walking, we found that this kinematic criterion predicts the onset and end of stance phase with an error smaller than 2% of the gait cycle duration (Borghese et al. 1996). This observation was confirmed in the present experiments in 4 control and 2 SCI subjects by monitoring in-shoe forces (PEDAR-mobile system, Novel, Germany). The insole contains 99 capacitive sensors interposed between the
subject’s foot and the shoe to measure the external vertical contact forces. Before each trial, the mean level of each sensor was measured while the foot was unloaded for a few seconds and this value was used as a zero level. Pressure threshold was 2 N/cm². We found that the resultant vertical force derived from the pressure sensors went above threshold (corresponding to foot landing on ground) and below threshold (foot take-off) in coincidence with the maximum and minimum elevation of the limb axis, respectively (with a precision of about 2% of the gait cycle). All data were time-interpolated over individual gait cycles on a time base with 200 points. In order to compare foot trajectories between patients and controls, we represented each VM trajectory as a time series of vectors and then compared the two resulting vector fields through a correlation measure. The directional correlation coefficient is given by the ratio of the co-variance of the times series and the product of their SDs (for a related approach, see Shadmehr and Mussa-Ivaldi 1994).

**EMG analysis**

EMG data were numerically rectified, low-pass filtered with a zero-lag Butterworth filter with cutoff at 3 Hz, and time-interpolated over a time base with 200 points for individual gait cycles. Filtering at 3 Hz was chosen in adherence with Winter’s (1991) recommendation to smooth the rectified EMG-data according to the biomechanics of movement of interest. Most kinetic (e.g., joint torques) and kinematic (e.g. joint angles) events of low-speed locomotion (such as that of the present study) have signal-power below 3 Hz. We used a zero-lag, two-pass (forward and backward) filter instead of a simple low-pass filter (that introduces frequency-dependent delays to mimic electrical-mechanical delays in force transmission, Winter 1991) because we did
not want to make any assumption about the temporal delays between the principal components extracted from the EMG patterns (see below) and the kinematics of the gait cycle. Note however that the temporal shift between the EMG-waveforms filtered with two-pass filter and those filtered with a simple low-pass filter would be less than 3% of the gait cycle in the present study. Mean EMG of ensemble averages were computed over all gait cycles.

**Factor Analysis.** The goal of factor analysis is to find a few orthogonal factors that adequately capture the underlying, hidden correlations among the original variables (Chau 2001), here represented by the recorded EMG profiles. Factor analysis therefore helps to understand multiple muscle coordination patterns. The steps involve calculation of the EMG correlation matrix, extraction of the initial principal components, application of the varimax rotation, calculation of factor scores and factor loadings, and interpretation of the results (Davis and Vaughan 1993; Merkle et al. 1998; Olree and Vaughan 1995; Poppele et al. 2002). The components are expressed using a varimax rotation in order to minimize the number of variables with high loadings on each component factor (Kaiser 1974). This has the effect of simplifying the interpretation since the waveforms of the rotated factors are more similar to those of the EMGs than are the initial principal components (Patla 1985; Wootten et al. 1990). While the first principal component in each case accounted for between 30% and 45% of the total waveform variance in the EMG-data set, the most significant rotated factors in each case explained between 20 and 30%. The appropriate application of factor analysis involves an initial estimate of a) the extent to which each original data waveform is composed of components common to other data waveforms, the communality, and b)
the extent to which activity is specific to each waveform alone, the uniqueness (Glaser and Ruchkin, 1976). In other words, we can think of EMG waveforms as being dependent on two aspects. First, there are some underlying common waveforms shared by other muscles. Second, each muscle also captures a unique aspect of activation that is not addressed by any other muscle.

In the present experiments, factor analysis was carried out on amplitude- and time-normalized EMG data of all recorded muscles over all steps of each session. Typically the data matrix included 360 columns (15 muscles x 8 gait cycles x 3 repetitions) by 200 rows of normalized gait cycle increments for each condition and subject. Eigenvalues and eigenvectors of the correlation matrix were computed. Finally, the eigenvectors were orthogonally rotated so that the variance explained by each rotated eigenvector was maximized for EMG-variables having similar profiles of activity and minimized for EMG-variables having dissimilar profiles. The resulting temporal components (factor scores) are independent of each other. The amount of variance explained by any given eigenvector was found by dividing its associated eigenvalue by the number of variables analyzed. We made a final ranking and selection of the number of basic components after the procedure of varimax rotation. Eigenvectors which explained only a small amount of variance (usually less than 5%) were excluded from further analysis. For each muscle, mean weighting coefficients (factor-loadings) of each component were obtained by averaging the values across all steps. Each weighting coefficient represents the correlation between an EMG-waveform and an EMG-component.

In addition to the components computed for each individual (control and SCI), we also computed a reference set of components from the ensemble of all controls, after
averaging across all gait cycles in each subject. To assess the predictive power of these reference components, the original EMG profiles of each patient were (least-squares) fitted as:

\[ \mathbf{m}(t) = \sum f_i(t)w_i \]  

(1)

where the vector \( \mathbf{m}(t) \) of time-varying muscle activation profiles of a patient is reconstructed as the linear combination of the first 5 reference time-varying components \( f_i(t) \) weighted by the vector \( w_i \) (weighting coefficients). Note that \( \mathbf{m}(t) \) corresponds to normalized EMG activity because of the normalization procedures of factor analysis (see above).

Statistics

All statistical comparisons between patients and controls were performed at matched values of BWS and treadmill speed using t-statistics. ANOVA designs were used when appropriate to test for the effect of different conditions on locomotor parameters. Reported results are considered significant for \( P<0.05 \). Statistics on correlation coefficients were performed on the normally distributed, Z-transformed values.

Results

Profiles of muscle activity

All patients could be trained to step with BWS. With training the extent of modulation of activity of limb and body muscles during the gait cycle increased. On average, the ratio of maximum to minimum of rectified EMG over the gait cycle in the last session was 3.24±1.13 higher (\( P<0.01 \)) than in the first session. In ASIA-C/D
patients, the mean amplitude of activity of leg muscles (TA, GCL, BF, RF, VL, GM) over the gait cycle in the last session did not differ significantly from controls, whereas the mean activity of axial muscles (RAM, RAS, OE, OI, LD, ES, TRAP) was significantly greater (by 3.19±1.59, P<0.01). In ASIA-A and ASIA-B patients, instead, the mean activity of leg muscles was significantly smaller than controls (0.29±0.24, P<0.001), and the mean activity of axial muscles was significantly greater (4.86±1.87, P<0.005).

At the end of training, the time course of changes of EMG activity of SCI-patients often remained poorly related to that of normal subjects (Fig. 1). Not only did EMG-waveforms of patients differ from controls, but they also could differ among patients. Thus, whereas controls activated reciprocally knee flexors and extensors (Fig. 1A,C), some patients co-activated knee flexors (BF) and extensors (RF,VL) throughout stance (e.g. D.S.), whereas others activated knee flexors briskly only in late swing (e.g. R.R. or O.M.). The correlation coefficient between the time series of activation of each muscle in a patient and the corresponding ensemble average in controls varied widely among patients but for most muscles was low (Fig. 1B, mean \( r = 0.13 \pm 0.36 \), range \(-0.63 / 0.89\)).

Temporal components of motor patterns

Even though the profiles of activation of individual muscles in SCI-patients could differ substantially from the normal ones, they were decomposable in the same basic set of temporal components. Factor analysis was used to extract the common components that account for most of the variance of the EMG activity across all recorded muscles. On average, the variance-accounted-for (VAF) by the first 5 components was 79±12%.
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(mean±SD), 81±7%, and 78±12% in controls, ASIA-C/D, and ASIA-A/B patients, respectively. Higher-order components were much less systematic and VAF of each of them was usually less than 5%. The time course of these 5 components was similar in normal and injured subjects (Fig. 2A and 2B, respectively). The components have been ranked first through fifth in the order given by Olree and Vaughan (1995), because the present components closely resemble those found by these authors in normal locomotion (see also Davis and Vaughan 1993). On average, the correlation coefficient between the components of each control subject and the corresponding reference components derived from the normal ensemble (thick-line traces in Fig. 2) was 0.86±0.04, whereas the correlation between the components of each patient and the reference components was 0.69±0.12. All 5 normal components were present in most (9/11) patients. In 2 ASIA-A patients, 4 components were similar to the control, but one component (#4 or #2, depending on the patient) was absent.

The phasing of the main peaks of each component relative to that of all other components can be appreciated in the polar plots of Fig. 2C, in which the polar direction denotes the relative time over the gait cycle. The results in the patients are compared with those in normal subjects (the patients being grouped according to the severity of the lesion). The arrows point to the time of occurrence of the positive peak of each component. On average, the peak occurred at 9±2, 46±4, 56±3, 76±4, and 98±2% of the gait cycle for components #2, 1, 4, 5 and 3, respectively, in normals. The corresponding peaks in patients generally occurred very close to those in controls. Note that, based on the relative timing, one can identify three primary components (component #2, 1 and 5, see Fig. 2C) occurring about every 1/3 of the gait cycle. The other two components are essentially the same as the first two, offset by a half of a gait cycle. Thus, in the polar
plots of Fig. 2C, the component # 4 is 180°-out-of-phase with the component # 2 (it lags behind the latter by 47% of the gait cycle), whereas the component # 3 is 180°-out-of-phase with the component # 1 (it lags behind by 52% of the gait cycle).

We next modeled the generation of the profiles of muscle activity in each subject as linear combinations of the five reference components. To generate a specific EMG profile, each reference component was scaled in amplitude and then all components were summed together (eq. 1 in Methods). Fig. 1C shows that the essential features of measured EMG profiles are well reconstructed both in normal subjects, and in 3 representative patients even for the leg muscles, controlled by sublesional segments. On average, the correlation coefficient between the measured profiles of each patient and the reconstructed profiles was 0.89±0.07 (range 0.62 - 0.99, Fig. 1D).

It is important to consider the relative weight of each temporal component, in addition to its waveform. In fact the distribution of the weighting coefficients across muscles reflects the pattern of muscle synergies and inter-muscular coordination. In contrast to the waveform of the components, their weights could differ substantially between patients and controls, and among patients, as one would expect from the reported differences in the waveforms of the individual EMGs (Fig. 1A-C). Figure 3 shows the weighting coefficients plotted on a color-coded scale for the normal ensemble and the grouped data from all patients. In the controls, some muscles loaded highly on an individual component (TA on # 5 and GM on # 2), but most muscles loaded significantly on several components. Thus, VL and RF loaded significantly on # 2 and # 3, and BF loaded on most components. There was a gradual transition in the pattern of distribution of the weighting coefficients as a function of severity of the lesion in SCI-patients: the pattern in ASIA-C/D patients bore more resemblance with the normal
pattern than did the pattern in ASIA-A/B patients. GCL loaded highly on component #2 instead of (ASIA-A/B) or in addition to (ASIA-C/D) #1, and BF loaded highly on #3 in ASIA-A/B patients instead of being spread across several components as in ASIA-C/D patients and the controls. The trunk muscles typically loaded highly on components #4 and #5, especially in severe SCI-patients, much more than in the controls. An important observation in all patients was that a given component (e.g. #5) could load highly both in supra-lesional muscles (e.g. trunk muscles) and in sub-lesional muscles (e.g. TA).

We have seen that the shape of the EMG-components generally changed little across conditions (speed and BWS changes). However their timing was systematically related to the speed of locomotion (Fig. 4). The peak of all 5 components shifted to successively earlier phases in the gait cycle as speed increased. These time-shifts suggest a linkage with specific speed-related events in the gait cycle (Bianchi et al. 1998; Grillner 1981; Winter 1991). Thus with increasing speed, the duration of stance as a percentage of gait cycle decreases and the foot is brought through faster during swing to place the heel for the subsequent stance earlier.

**Correlation with kinematics**

At the end of training, the foot path of the SCI-patients tended to recover the shape typical of normal stepping, with reduced step-by-step variability (Fig. 5A). Trajectory shape was compared between patients and controls by computing the correlation coefficient between the time series of foot-position-vectors in each patient and the ensemble average in controls. The mean correlation coefficient was 0.98±0.03. We also analyzed foot trajectory separately in the vertical direction (foot lift, VM_y) and
in the horizontal direction (foot translation, VM_x). It is well known that these two parameters are actively controlled in normal subjects at different speeds and BWS (Ivanenko et al. 2002; Winter 1991). The time series of VM_y and VM_x-values were compared between patients and controls by computing the corresponding correlation coefficients. In ASIA-C/D patients, the mean correlation coefficient was 0.94±0.04 and 0.99±0.01 for VM_y and VM_x, respectively. In ASIA-A and ASIA-B patients, mean correlation was 0.74±0.07 and 0.95±0.04 for VM_y and VM_x, respectively. The lower correlation in the vertical direction is due to foot-drop in paraplegics.

We found that EMG-components #5 and #3 were significantly correlated (P<0.001) with VM_y and VM_x, respectively, in both controls and patients, as shown by superposition (Fig. 5B) and cross-correlations (Fig. 5C). The time course of the components generally matched well that of the corresponding foot variable over wide portions of the gait cycle. In some patients (see R.R. in Fig. 5B), component #3 matched well VM_x during swing and heel strike, but poorly during mid-stance. Correlation between EMG components and foot kinematics also is shown by their in-phase shift with speed (Fig. 5D). When stepping is compared between 0.7 km/h and 2 km/h, all 4 variables shift by 9.7±1.4% of the gait cycle in controls and by 11.6±2.5% in patients.

**Discussion**

We reported that, with training, foot motion of SCI-patients tended to recover the shape and the step-by-step reproducibility that characterize normal gait. However the specific activation profiles of individual muscles often differed substantially from the normal ones, consistent with the hypothesis that recovery of foot kinematics may depend on learning new motor strategies to replace lost function (Barbeau et al.
1999a,b; Dietz 2002a; Edgerton et al. 2001; Harkema et al. 2000; Pearson 2000). A key point of the present study is that both the default motor strategies of healthy subjects and the new adaptive strategies developed by the patients are subserved by the same set of temporal components hidden in the patterns of muscle activity. Moreover, we showed that two of the identified components are systematically related to foot kinematics.

**Temporal components of motor patterns**

We used factor analysis to identify the temporal components that are common to the activation of multiple muscles during locomotion, and that therefore may reflect fundamental control signals directed to the corresponding α-motoneuron pools (Chau 2001; Davis and Vaughan 1993; Merkle et al. 1998; Olree and Vaughan 1995; Poppele et al. 2002). In this analysis the basic temporal components are not specified *a priori* as in a Fourier series expansion, but they are determined *a posteriori* by the statistical structure of the data waveforms (Glaser and Ruchkin 1976). Given the *a posteriori* statistical nature of the components, the finding that the same basic set of temporal components is extracted from a wide range of very different muscle activity profiles in both controls and SCI-patients is non-trivial. Five such components accounted for most of the variance of the EMG activity across all recorded muscles. Their shape generally was similar in both controls and patients, and across a variety of different loading and speed conditions (Fig. 2). One component was absent in 2 SCI-patients (a different one in each of them), further indicating the non-trivial nature of component identification.

The distribution of the weights of each component across muscles reflects the complex patterns of muscle synergies and inter-muscular coordination. The weights of the five basic temporal components often differed substantially between patients and
controls and among patients (Fig. 3). Thus, even though the fundamental signals expressed by the five temporal components are preserved after a spinal lesion, their distribution to the α-motorneurones pools is re-wired, presumably as a result of lesion-and training-induced plasticity (see below).

**Functional significance**

The interpretation of the EMG-components rests on two key findings of the present study. First, SCI-patients demonstrated motor equivalence of the stepping behavior: foot kinematics similar to the normal one was generated by activity patterns of individual muscles and muscle synergies often different from the normal ones. Therefore, if spinal reflex mechanisms (such as the flexor reflex) are involved in step generation (Sherrington 1910), their contribution should be highly flexible and plastic. Also, invariant EMG-components underlying the generation of variable EMG activity cannot reflect local biomechanical variables of individual muscles, such as muscle forces, but might reflect more global parameters of limb motion, such as foot kinematics (Fig. 6). In locomotion, the control of foot position requires more global coordination than the control of a single joint. Foot kinematics depends on the spatio-temporal coordination of multiple muscles acting on several body and limb segments (Winter 1991). We found that two of the EMG-components systematically correlated with foot lift and translation, shifting in parallel with the latter variables when subjects incremented the speed of locomotion.

It is known that the kinematic variables of foot lift and translation are actively controlled during normal gait at different speeds and BWS (Ivanenko et al. 2002; Winter 1991). These variables are equivalent to parameters of endpoint position that are
encoded at different levels of the CNS, from motor cortical neurons (Georgopoulos and Grillner 1989) to spinal proprioceptors (Bosco and Poppele 2001). Thus foot translation is related to the direction of endpoint movement, and foot lift is related to the distance of the endpoint from the body. In locomotion these kinematic variables represent important signals to trigger stance-to-swing transition (Grillner 1981; Orlovsky et al. 1999), also seen in SCI-patients (Dietz et al. 2002a,b). The functional significance of the other temporal components remains to be determined by further studies, but they may reflect foot kinetics, such as loading and propulsion forces (Davis and Vaughan 1993; Olree and Vaughan 1995). Afferent inputs from load-receptors make essential contributions to the activation of leg muscles during locomotion also in SCI-patients (Dietz et al. 2002a,b; Harkema et al. 1997).

An intriguing question is how the complex re-organization of the pattern of motor activity can give rise to motor equivalent solutions. Temporally tuned patterns of muscle activity in proximal muscles may produce the desired kinematics of the foot by taking advantage of the biomechanical coupling of the angular motion of different limb and body segments (Bianchi et al. 1998; Lacquaniti et al. 1999). Coupled angular motions also generate sensory stimulation that can entrain caudal sublesional segments of the cord and result in patterned activity of leg muscles (Giszter et al 1998; Pearson 2001). These putative mechanisms are congruent with the current idea that long-term adaptive changes involved in functional restoration may be driven by alterations in the sensorimotor loops linking afferent feedback to ‘unit burst generators’ (Barbeau et al. 1999b; Dietz 2002a; Edgerton et al. 2001; Grillner 2002; Harkema et al. 2000; Pearson 2000).
Hypothetical organization of neural substrates

Although the elucidation of the neural substrates underlying the basic EMG-components probably must await invasive approaches in the animal, the present results do have a bearing on our ideas about the functional organization of locomotor pattern generators (Fig. 6). Because all the muscles that are active during locomotion express combinations of the same basic temporal components, such components may be related to the control signals directed to the muscles. Moreover, the observation that the components are present even in muscle activity originating from sublesional levels in SCI-patients with a clinically complete spinal transection suggests that these components are related to the control signals output by spinal pattern generators. However both descending and afferent inputs most likely contribute to their shaping.

In SCI-patients a given component could be expressed with a strong weight both in supra-lesional muscles and in sub-lesional muscles. This is coherent with the current idea that locomotor pattern generation may not be localized to discrete regions of the spinal cord, but may depend on a distributed network extending rostro-caudally across several spinal segments (Barbeau et al. 1999b; Dietz et al. 1999; Edgerton et al. 2001; Grillner 2002; Orlovsky et al. 1999; Yakovenko et al. 2002).

The basic temporal components might originate either from waves of activation propagating back and forth along the spinal cord or from abrupt switching between distinct generators with bursts of activity occurring at certain points in the step cycle (Kiehn et al. 1998; Orlovsky et al. 1999; Yakovenko et al. 2002). We found that three primary components occurred about every 1/3 of the gait cycle, while the other two components were half-cycle phase-shifted versions of two primary components (see Figs. 2C-E).
The basic components could be distributed with adjustable synaptic weights (corresponding to the weighting coefficients of Fig. 3) by propriospinal neurons that project to different pools of α-motorneurones across several spinal segments. The component weights generally differed substantially between patients and controls, and among patients. Component weights correspond to the connection strengths in a distributed network (Prentice et al. 1998). Lesion- and training-induced plasticity might lead to changes in the connections of the interneuronal network. These changes are adaptive and learnt (being specific to the trained task, de Leon et al. 1998) and involve a major redistribution of activity to different limb and body muscles (Pearson 2001), creating new muscle synergies (Barbeau et al. 1999b). The specific re-organization of the network probably depends on the level of the lesion (Dietz et al. 1999) and on the extent of plastic changes occurring in each individual (Harkema et al. 2000).

**Conclusions**

In summary, we reported that patients with spinal injury can learn to produce foot movements similar to that of healthy subjects. Hidden in the muscle patterns we found a basic set of five temporal components, whose flexible combination accounted for the wide range of muscle patterns recorded in both controls and patients. Because the temporal components correlate with global kinematic goals of locomotion, in the future they could be useful to drive neuroprostheses for SCI-people. Thus, appropriately weighted components could be distributed by neural network algorithms running on computers to a set of stimulators imbedded in the patient’s paralyzed muscles.
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Spatiotemporal activation of lumbosacral motoneurons in the locomotor step cycle.

Figure legends

Fig. 1. A: Distribution of activity across different limb and body muscles as a function of gait phase (ES early stance, MS1 mid-stance 1st half, MS2 mid-stance 2nd half, LS late stance, ESw early swing, LSw late swing). Rectified EMG was integrated over each epoch and plotted in color-scale. Data from a typical control, one ASIA-C-patient, and one ASIA-A-patient are plotted in the top, middle and bottom panels, respectively. B: Mean correlation coefficients of EMG activity between patients and controls. C: Comparison of normalized EMG profiles between the ensemble average of controls and one ASIA-C, one ASIA-A and one ASIA-B-patient (left to right). Thick traces superimposed on the raw data are the profiles reconstructed as weighted sum of the first 5 reference components derived from the normal ensemble. D: Mean correlation coefficients between the measured EMG profiles of each patient and the profiles reconstructed from the reference components. All data are derived from stepping at 0.7 km/h, 75%-BWS, and data from patients are at the end of training.

Fig. 2. Time course of the temporal components in controls (A) and patients (B) for stepping at 2 km/h, 0-75%-BWS. The components extracted by factor analysis from individual subjects (thin-line) are superimposed on the reference components derived from the normal ensemble (thick-line). Bottom-right panels in A and B: cumulative variance-accounted-for by the first 5 components. C: Polar plots of the components obtained from the normal ensemble, grouped data from all ASIA-C/D-patients, and grouped data from all ASIA-A/B-patients, from left to right. Polar direction denotes the relative time over the gait cycle (time progresses clockwise), and radius denotes the amplitude of the component.
Fig. 3. Weighting coefficients of the temporal components in individual activity patterns of 12 muscles for all subjects stepping at 2 km/h, 75%-BWS. In each panel, the coefficients derived from the grouped data of all ASIA-A/B-patients, ASIA-C/D-patients, and normal subjects are plotted in a color-coded scale in the left, middle and right columns, respectively.

Fig. 4. Effect of speed on the 5 components in one representative control (A) and one ASIA-B-patient (B).

Fig. 5. Correlation of the components #5 and #3 with foot kinematics. A: Shape and variability of foot trajectory in controls, two ASIA-C and two ASIA-A patients (left to right). The trajectories of the ilium and fifth metatarso-phalangeal joint over all consecutive unassisted step cycles (n=8) have been superimposed. B: The time course of component #5 (blue) is compared with that of foot lift (red), and component #3 is compared with foot translation, for the same subjects as above. C: Cross-correlograms between components and foot variables in all controls and patients. Correlation values are plotted as a function of time delay (expressed as percent of gait cycle). D: Phase shift of peak values of components (blue) and foot variables (red) as a function of speed. Shifts toward later (positive) or earlier (negative) phases of the gait cycle are plotted relative to the values at 2 km/h.

Fig. 6. Hypothetical scheme for limb control in locomotion. Basic temporal components are expressed by pattern generators under the influence of descending and afferent
inputs. Components are distributed with adjustable synaptic weights (including the weighting coefficients of Fig. 3) by an inter-neuronal network to different pools of α-motorneurones across several spinal segments.
Correlation with control EMG waveforms

A

D.S. patient

R.R. patient

B

Correlation with control EMG waveforms

C

averaged control
D.S. patient
O.M. patient
O.C. patient

D

Prediction from 5 reference components

Fig. 1
Fig. 2
Weighting coefficients

Fig. 3
Fig. 4
Fig. 5
Fig. 6