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Changes in muscle activation patterns in response to enhanced sensory input during treadmill stepping in infants born with myelomeningocele

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Abstract

Infants with myelomeningocele (MMC) increase step frequency in response to modifications to the treadmill surface. The aim was to investigate how these modifications impacted the electromyographic (EMG) patterns. We analyzed EMG from 19 infants aged 2–10 months, with MMC at the lumbosacral level. We supported infants upright on the treadmill for 12 trials, each 30 seconds long. Modifications included visual flow, unloading, weights, Velcro and Icriction. Surface electrodes recorded EMG from tibialis anterior, lateral gastrocnemius, rectus femoris and biceps femoris. We determined muscle bursts for each stride cycle and from these calculated various parameters. Results indicated that each of the five sensory conditions generated different motor patterns. Visual flow and friction which we previously reported increased step frequency impacted lateral gastrocnemius most. Weights, which significantly decreased step frequency increased burst duration and co-activity of the proximal muscles. We also observed an age effect, with all conditions increasing muscle activity in younger infants whereas in older infants visual flow and unloading stimulated most activity. In conclusion, we have demonstrated that infants with myelomeningocele at levels which impact the myotomes of major locomotor muscles find ways to respond and adapt their motor output to changes in sensory input.

Keywords

EMG; Infant; Muscle activation; Myelomeningocele; Sensory input; Stepping

1. Introduction

The stepping response is a cyclical patterned movement with alternating interlimb co-ordination that has been observed in neonates and fetuses (Barbu-Roth et al., 2009; Dominici et al., 2011; Forssberg, 1985; Thelen, Fisher, Ridley-Johnson, & Griffin, 1982; Thelen, Ulrich, & Niles, 1987). Researchers have also demonstrated that infants will step, when manually supported upright so their feet rest on a moving treadmill surface (Teulier et

al., 2009; Thelen, 1986; Thelen & Ulrich, 1991; Ulrich, Ulrich, Angulo-Kinzler, & Yun, 2001; Yang, Stephens, & Vishram, 1998). There are a range of theories underlying the occurrence of stepping responses. Forssberg proposed that steps are a manifestation of the central pattern generator (CPG) for walking, that is, a structural network of neurons in the spinal cord predisposed for producing coupled cyclical alternating contractions of the lower limb flexors and extensors with minimal input from supraspinal centers (Forssberg, 1985). An alternative concept, based on dynamical systems theory (DST), considers that stepping, at any point in development, is the product of many intrinsic and extrinsic subsystems converging to cause patterns that continually adapt but can also stabilize with sufficient repetition (Thelen & Smith, 1994; Ulrich, 2010). The fundamental difference between these two theories is that CPG theory assumes the innate existence of dedicated networks of neurons responsible for generation of a basic cyclical activation pattern for walking. By contrast, DST takes a more global approach to the neural system and stresses the necessity of sensory input to access and strengthen over time, from among a repertoire of available neural connections, synergies of neurons that fit the context and goal (Sporns & Edelman, 1993). From a therapeutic perspective, the two theories offer very different prognoses regarding development of stepping and walking, following an embryonic spinal cord lesion. In the event of a spinal cord lesion, such as myelomeningocele (MMC) impacting the primitive locomotor neurons, stepping could be lost, or display aberrant patterns according to classical CPG theory. However, DST postulates that behaviors fit the context and task, given the available resources and contexts. Repeated cycles of sensory input coupled with functional motor output drives the change in neuromotor networks producing a stepping response even in the presence of extensive spinal cord lesioning. Similarly, neuroscience research on the developmental organization of the brain and spinal cord emphasize the plasticity within the system, both in typical development and neurorehabilitation (Karmiloff-Smith, 2009; Kleim & Jones, 2008).

A factor known to affect step frequency and interlimb coordination is the infants' neural and physiological makeup, with fewer steps produced by infants with myelomeningocele (MMC) and Down syndrome (DS) compared to infants with typical development (TD) (Teulier et al., 2009; Ulrich, Ulrich, & Collier, 1992). In these infants, the feedforward and feedback loops organizing movement patterns are diminished, thus compromising the step response compared to infants with an intact nervous system. However, even in these clinical populations we have previously reported a change in the parameterization of their steps and increased step frequency in response to some forms of enhanced sensory inputs (Pantall, Teulier, Smith, Moerchen, & Ulrich, 2011; Ulrich, Ulrich, & Angulo-Kinzler, 1998). Visual flow increased treadmill step rate significantly in the older infants but not in younger infants. Specifically, enhanced sensory inputs via visual flow and friction on the treadmill belt significantly increased step rate, particularly for older (7–10 months) compared to younger (2–5 months) infants for whom only friction increased stepping (Pantall et al., 2011).

In this study we focus on the stepping response of infants with a lumbar or sacral myelomeningocele (MMC). MMC is the most common neural tube defect in the US (Bowman, Boshnjaku, & McLone, 2009), present in the spinal cords of approximately 1 in 3000 live births (Canfield et al., 2006). The relevance of studying this type of clinical population is to determine system adaptability, given that the link between the peripheral sensory and motor neurons, spinal interneurons and supraspinal components may be compromised, thereby disrupting neurophysiological and consequentially locomotor function. The clinical neurological picture presents as a combination of lesions of the lower and upper motor neurons, disturbed sensory pathways and compromise of the autonomic system at and distal to the level of the MMC. Commonly associated orthopaedic problems that may additionally impact gait include scoliosis, talipes equus, subluxated hip and muscle contractures (Iborra, Pages, & Cuxart, 1999; Norrlin, Strinnholm, Carlsson, & Dahl, 2003;

Swaroop & Dias, 2009). Infants with MMC who are capable of developing locomotor skills begin to walk on average 2 years later than typically developing (TD) infants (Iborra et al., 1999). Lesions above L2 carry a poor prognosis for independent walking compared to lesions at and caudal to S1 (Seitzberg, Lind, & Biering-Sørensen, 2008; Hoffer, Feiwel, Perry, Perry, & Bonnett, 1973; Iborra et al., 1999; Williams, Broughton, & Menelaus, 1999). The energy cost of locomotion is higher and lower limb muscle power is lower compared to TD children (Moore, Nejad, Moore, Robert, & Dias, 2001; Schoenmakers et al., 2009). The kinematics of gait in MMC children also differs from that of infants with TD, with more trunk rotation, anterior pelvic tilt and increased pelvic movements (Gutierrez, Bartonek, Haglund-Akerlind, & Saraste, 2003).

We have previously demonstrated in infants with MMC that there is a response in step frequency to enhanced sensory input indicating incompleteness of the spinal lesion permitting afferent and efferent neural signal transmission and thus a potential motor response to changes in sensory input (Pantall et al., 2011). The question we address here is how does enhancing the afferent input impact the underlying electromyographic (EMG) activity patterns during stepping in infants with MMC? The receptors stimulated by each of the enhanced sensory conditions differ in type, quantity and level of activation and may, thus, contribute to different muscle activity states during stepping. Can we identify different muscle activation patterns associated with each of the enhanced sensory conditions? In addition, we asked what effect age has on the response of the nervous system to each of the enhanced sensory conditions? Specifically, what effect does visual flow have on EMG in the younger infants even in the absence of increased stepping?

Temporal, kinematic and muscle activity have previously been investigated in the stepping response of infants with TD. The limited studies analyzing muscle activity have reported increased EMG amplitude in lateral gastrocnemius prior to foot contact in neonatal stepping (Forssberg, 1985; Thelen et al., 1982; Thelen et al., 1987). With increasing age, Okamoto et al. recorded a shift in pattern from co-activation and reversed reciprocal activity to reciprocal activation in infants (Okamoto, Okamoto, & Andrew, 2001; Okamoto, Okamoto, & Andrew, 2003). They further reported that during this period lateral gastrocnemius and quadriceps femoris became more active just before foot contact. A recent study by Dominici et al. (2011) suggested that the primitive patterns present in the stepping response is retained as independent walking is achieved and augmented by two additional motor patterns. There are no published studies on EMG in infants with MMC during the stepping response. However, a study investigating lower limb muscle activity in children with MMC aged 4–17 years during walking showed earlier and more prolonged activation of neurologically intact muscles and altered timings of neurologically impaired muscles (Park, Song, Vankoski, Moore, & Dias, 1997).

No previous study has examined the effect of enhanced sensory input on EMG in infants with or without MMC. Musselman and Yang (2008) investigated the effect of applying weights to the lower limbs of infants with TD. They reported that weights increased the extension phase during treadmill stepping. Barbu-Roth et al. observed that visual flow increased air stepping responses in neonates despite the nascent development of the visual system (Barbu-Roth et al., 2009; Gilmore, Hou, Pettet, & Norcia, 2007). Locomotor training for adults with spinal cord lesions stimulated changes in lower limb EMG patterns through increased and altered muscle activity (Erni & Colombo, 1998). Greater lower limb muscle activity has been associated with an increase in locomotor function in adults with paraplegia (Dietz, Colombo, & Jensen, 1994; Dietz, Colombo, Jensen, & Baumgartner, 1995).

Our goal was to investigate activation of four primary gait muscles bilaterally in younger infants (2–5 months) and older infants (7–10 months) with MMC when stepping on a

treadmill under five conditions of enhanced sensory input compared to baseline, on the treadmill. We examined patterns of activation for individual muscles, agonist-antagonist co-activation, and the distribution of muscle states (amount of overlap and lack of EMG activity during stride cycles). Muscle activation patterns are dynamic and constantly adapting to the sensory input, particularly in the infant population under investigation. In addition, we wanted to document for these infants with compromised neural tracts, the capacity of each of the primary gait muscles to activate during stepping (Chang, Kubo, Buzzi, & Ulrich, 2006). For infants this young, standard neurological testing of myotomes is not possible, thus, an activity measure like this, provides a measure of neuromotor functionality.

Our hypotheses were that unique muscle activation states would emerge based on sensory input as reflected in surface electromyography responses. We anticipated that the conditions of friction and visual flow that resulted in a statistically significant increase in step frequency would produce the most different muscle activation patterns compared to baseline. Specifically, our hypothesis was that these conditions would produce increased activity of the four gait muscles, particularly LG, in addition to reduced coactivity of muscle pairs. A further hypothesis was that younger infants would differ in their response to the enhanced sensory stimuli compared to older infants as a consequence of their less developed nervous system and limited ability to control muscles individually.

2. Methods

2.1. Participants

Twenty-seven infants with MMC enrolled in this study, eleven aged 2–5 months and sixteen aged 7–10 months (corrected age). All these infants produced ten steps or more across trials. We identified these two age ranges as ones that would reflect early development in the first year, prior to the typical emergence of trunk and leg functional skills, and later development, as more functional control began to emerge. We limited enrollment to infants with lesion levels identified as lumbar or sacral because this sample has a higher likelihood of walking than those with higher lesion levels. We also constrained the sample to infants with gestational age of >28 weeks, and no other neuromotor or cognitive anomalies not associated with spina bifida. We recruited infants via MMC hospital clinics and parent support groups in the SE Michigan, NW Ohio, and SE Wisconsin areas. Approval for this study was granted by the Institutional Review Board of the University of Michigan. Parents signed written informed consent documents prior to their child's participation and completed a medical status and history survey (including an indication of any complications and medical issues, level of spinal fusion surgery).

2.2. Procedure

Note that the infants involved in this study were part of a larger study, for which we reported previously the procedures for data collection and a subset of results (For additional details, please see Pantall et al., 2011). This was a collaborative study, thus, five infants were tested in the Pediatric Neuromotor Laboratory at the University of Wisconsin, Milwaukee; all others were tested either in their homes (SE Michigan, NW Ohio) or in the Developmental Neuromotor Control Laboratory at the University of Michigan. In order to increase the diversity of our sample and sample size, we developed a set-up that could be used in family homes; those families unable to drive to our lab could, thus, participate. Set-up was the same in the lab and homes except that our cameras could not be placed as far from the test session in all cases as in the lab (2.0 m) and the ambient lighting was less well-controlled.

The set-up consisted of a custom-designed pediatric treadmill (Carlin's Creations, Sturgis, MI) placed on top of a table in the center of the test area. Two digital camcorders (Panasonic PV-GS 35) were placed at 90° (*M*) angle to each other, facing the right side of the infant, at

chest level. We calibrated the test space prior to testing with a Peak Motus™ calibration frame and synchronized camcorders and EMG system with a mechanical trigger.

We prepared infants by removing their clothing and cleaning the skin surface for all markers and electrodes with alcohol wipes. We then placed spherical reflective markers (8 mm diameter) on the right iliac crest, greater trochanter, knee, ankle, and 3rd metatarsophalangeal joints and positioned preamplified bipolar (2 cm spacing) surface electrodes (Noraxon®, Dual electrode #272, Scottsdale, AZ) over the muscle bellies of the lateral gastrocnemius (LG), tibialis anterior (TA), rectus femoris (RF), and biceps femoris (BF) on both lower limbs. We selected these superficial muscles as they are considered primary gait muscles. Optimal sites for electrode placement were determined by first palpating anatomical landmarks to identify bony attachments of muscles, followed by locating the muscle belly. In infants, it is not possible to confirm optimal electrode placement by asking the participant to perform a maximum isometric voluntary contraction. A ground electrode (Huggables® ECG electrode, ConmedR, NY, USA) was attached to the skin overlying the right patella. We recorded EMG signals via an 8-channel Myosystem 1400A (Noraxon Inc., Scottsdale, AZ) unit at 1000 Hz, with an 8th order Butterworth low-pass filter, cut-off at 500 Hz and 2nd order high pass filter, cut-off at 10 Hz. The common mode rejection rate was a minimum 100 db at 50–60 Hz.

For treadmill test trials we held infants upright so their feet rested on the belt of the treadmill, with belt speed set at 0.16 m/s, for twelve 30-trials. Treadmill testing lasted approximately 6 min, with rest intervals interspersed among trials, as needed. Trials were presented in two sets of 6 conditions, with each set presented in random order:

- i. *Baseline* – smooth black belt surface.
- ii. *Visual flow* – black and white checker-board patterned belt used to elicit visual flow sensation.
- iii. *Unloading* – infant was held near the end of the treadmill, so their feet abruptly dropped off the surface, rapidly unloading the receptors in hip and ankle joints.
- iv. *Weights* – attached to the infant's shanks, tailored to their individual weight and equal to approximately 50% of shank mass. Our goal here was to enhance normal contribution of passive pendular forces in swing and to increase input to pressure receptors in the foot during stance.
- v. *Velcro* – infants wore socks with strips of Velcro sewn on and the treadmill was covered with a felt-like material. We predicted that this would increase duration of foot contact with the belt and increase muscle activity required to remove the foot from the belt.
- vi. *Friction* – belt made of Dycem (a non-slip surface) to limit infants' feet from sliding on the belt surface (a non-slip surface with a coefficient of friction of 1, compared to the baseline belt's coefficient of friction, of .2) to limit infants' feet from sliding on the belt surface.

2.3. Data reduction

Our first level of data reduction was to determine the frequency and type of steps taken by infants in these contexts. These results are published elsewhere (Pantall et al., 2011). We then processed the kinematic and EMG data. As a result of markers tracking incorrectly and excessive noise in the EMG signal, we were unable to process data for eight of the infants (this included all the infants from Wisconsin). We processed data for a total of nineteen infants, ten aged 2–5 months and nine aged 7–10 months. For the kinematic data we calculated segmental angles for the foot, shank and thigh. Foot segmental angle was defined

as the angle between the foot segment and the vertical axis; segmental angles for the shank and thigh were the angles between the axes of these respective segments and the right horizontal axis. We also calculated maximum vertical, anterior-posterior, and medial-lateral displacements of the toe marker during the stride cycle. All calculations were performed using custom programs written in Matlab (Natick, Massachusetts).

We selected a subset of steps per infant, prioritizing alternating steps and only selecting single steps when there were insufficient alternating steps. We preferentially selected alternating steps on the basis that these had a more regular pattern of motor activity. The number of steps available varied between conditions. We restricted the number of steps for each infant per condition to a maximum of five steps. In order to avoid bias, we only included the mean where infants produced multiple steps for a given condition. Table 1 lists the number and type of steps we analyzed for each infant per condition.

We applied a second order high pass (30 Hz) and eighth order low pass (400 Hz) forward-reverse Butterworth filter to the EMG data to remove movement artifact and high frequency thermal noise respectively. Additionally, due to high levels of electrical noise, we performed further filtering with a notch filter at 60 Hz and resonant frequencies up to 400 Hz. The band-passed EMG signal was then full-wave rectified and smoothed using a forward-reverse second order Butterworth filter with a window of 50 ms. We processed the smoothed rectified data into 'on' or 'off' activity. The threshold for 'on' was exceeded when the EMG amplitude exceeded two standard deviations above the mean EMG for the entire trial (Hodges & Bui, 1996). Activity above this threshold had to be maintained for a minimum consecutive time period of 5 ms for the muscle to be classified as 'on' which decreased the probability of incorrect classification of the EMG. If the time between consecutive muscle bursts was less than 50 ms, the individual muscle bursts were considered continuous. Time events (initial foot contact in stance, and foot off to begin swing) were then used to divide the muscle 'on-off' vector into individual stride cycles, which were then time normalized to 100 points.

We looked at the probability of the muscle being 'on' or 'off' by combining the stride cycles (maximum of five) for an infant for a given condition. The number of times the muscle was 'on' at a given point in the cycle was divided by the total number of stride cycles. A value of '0' therefore indicated that the muscle was never activated at that point in the stride cycle, a value of '0.5' that the muscle was 'on' in fifty percent of all cycles and a value of '1' that the muscle was always activated for that time-point of the cycle. We calculated the percentage of the gait cycle that a muscle was 'on' for each muscle recorded under the different sensory conditions. We also determined the mean burst duration and burst frequency for the entire trial and stride cycle for each of the 6 conditions. We defined the burst frequency for the stride cycle as the number of bursts during the stride cycle divided by the stride cycle duration.

We applied two methods to analyze activation of muscle combinations based on previous work in our laboratory (Chang et al., 2006). The first method reflected what combination of the four muscles were 'on' or 'off' during a frame. There were 16 possible combinations ranging from all 'off' [0000] to all 'on' [1111], where 0 represents 'off' and 1 represents 'on'. The second method determined the level of co-activation of ipsilateral agonist-antagonist muscle pairs, namely TA-LG, RF-BF and RF-LG. The algorithm we used was that developed by Chang et al. (2006), based on Winter's method (Winter, 1990) and defined as:

$$\text{Co-activation value} = 2 \times \frac{\text{No. of frames when both muscle A and B are 'on'}}{\text{No. of frames when muscle A is 'on'} + \text{No. of frames when muscle B is 'on'}}$$

2.4. Statistical analyses

We used SAS version 9.2 (SAS® Inc., Cary, NC) for statistical analyses. We applied the mixed model procedure to conduct analyses of variance (ANOVA) with repeated measures on enhanced sensory condition. We performed five (baseline versus enhanced sensory conditions) pairwise comparison tests to determine significant differences between parameters. We also applied a MANOVA with repeated measures on enhanced sensory condition, with the co-activation values for the three muscle pairs as dependent variables. For the nonparametric muscle states and kinematic data we applied Friedman's repeated measures ANOVA and Dunn's posthoc tests when the ANOVA resulted in significant differences between conditions. We set statistical significance at $p < .05$. We have presented statistics mainly for those results that achieved statistical significance and on occasion for those without statistical significance where we consider this lack of significance to be of interest.

The final analysis we undertook was principal component analysis (PCA). This multivariate analysis procedure allowed us to identify clusters of variables that distinguish among the enhanced sensory conditions and baseline. We combined data for each muscle's percentage of the cycle during which it was 'on', burst frequency and burst duration, along with the three values for co-activation states into a 6 (conditions) $\times$ 15 (EMG variables) matrix. We calculated PCA for younger infants, older infants and all infants combined. The Kaiser-Meyer-Olkin statistical sampling statistic was calculated for all 3 groupings which was greater than .5, therefore indicating an underlying factorial structure in the matrices and thus appropriate for PCA.

3. Results

3.1. Kinematics

In order to understand what movements the EMG patterns we report are producing, here we present endpoint trajectories of limb segments when our infants with MMC stepped on the treadmill. Fig. 1 shows foot, shank and thigh segmental angles for 3 stride cycles in Baseline for a typical infant in the older age group with a lower lumbar lesion; cycle events are labeled. As this figure illustrates, embedded within the ultimately appropriate flexions and extensions of swing and stance phases was high variability. This kinematic variability was evident in trajectories and in the frequent reversals of direction within a phase, for the same individual in addition to between infants.

The impact of enhanced sensory conditions on the mean peak displacement of the toe marker for all infant steps during swing, in vertical, anterior-posterior (AP), medial-lateral (ML) axes and cumulative 3D displacement is illustrated in Fig. 2. Standard deviation bars reflect similarly the high variability across cycles depicted in the segmental angles above. There was a significant difference in vertical displacement (Friedman's ANOVA $\chi^2(5) = 30.39$, $p < .001$) and AP displacement (Friedman's ANOVA $\chi^2(5) = 31.11$, $p < .001$). Velcro significantly increased vertical displacement ($p < .05$) compared to baseline. Unloading resulted in greater vertical displacement compared to baseline ($p < .05$) but decreased AP displacement ($p < .05$).

3.2. EMG – individual parameters

Fig. 3 illustrates the ultimate objective determination, via the application of our algorithm, of on-off periods of muscle activity. The trace is that of smoothed, rectified EMG for one 30 s trial in baseline, for the right TA muscle. The percentage of time a muscle was ‘on’ during stride cycles, across all infants and collapsed over conditions ranged from 28.1% (± 33.9) for TA to 40.7% (± 36.6) for BF. We found that for 50.2% of the cycle at least one of the 4 muscles was ‘on’ during baseline. However, in 24.4% of stride cycles we did not record any muscle activity. We did not find any significant condition effect on duration that a muscle was ‘on’.

Fig. 4 shows that overall the probabilities of a muscle being ‘on’ were seldom above .5. Descriptively, sensory conditions did modify the muscle pattern, with the alteration depending on the age of the infants. For younger infants, baseline tended to produce less probability of activation in the TA and LG than nearly all enhanced sensory conditions; for the RF and BF baseline was more intermediate to the other conditions. For older infants, enhancing sensory input via visual flow and unloading tended to increase probabilities somewhat over baseline.

We found a significant condition effect for LG for both burst frequency and burst duration, $F(5,69) = 3.23$, $p = .011$, and $F(5,54) = 4.56$, $p = .002$, respectively. Weights decreased frequency from 0.83 bursts per second (± 0.68) in baseline to 0.41 bursts per second (± 0.54) for LG ($df = 69$, $t = 2.10$, $p = .039$) (see Fig. 5). Velcro increased burst durations from 0.48 s (± 0.51) in baseline to 1.20 s (± 1.06) ($df = 54$, $t = -3.57$, $p < .001$) (see Fig. 6). Fig. 7 presents graphically the percentage of the cycle during which each of the 16 muscle activation states occurred. The muscle states of no activation, single LG activation, single BF activation, LG/BF activation, LG/TA activation and all 4 muscles activated appeared to vary most between conditions. We therefore performed a sequence of Friedman’s ANOVAs for these 6 muscle states. However, we did not find any statistical significance for condition on muscle state. We observed that younger infants produced the ‘all on’ four muscle coactivity state for 8.9% of the time while stepping, compared to 2.3% of the stride cycle for older infants.

3.3. Principal component analysis of 15 EMG variables

In the previous results sections we analyzed muscle activity by separating various descriptive characteristics. However, none of these characteristics truly represents an aspect of the behavior that is unrelated to the others; for example frequency of activation is related to overall amount of activity. Additionally, muscles do not act in isolation but rather interact with each other in real time. PCA is a multivariate method that allows one to view the impact of a condition on a number of dependent variables simultaneously. An analogy can be drawn between listening to individual instrumental scores within an orchestral composition and hearing the entire orchestra play together. Each instrument will have some correlation with neighboring instruments but one cannot fully appreciate the music until one hears the entire orchestra playing. The dependent variables we entered into the PCA were the percentage of time a muscle was ‘on’ (BP), burst duration (BD) and burst frequency (BFR) for the four muscles. In addition we included values for co-activation of TA and LG (CO-TA/LG), co-activation of RF and BF (CO-RF/BF) and co-activation of RF and LG (CO-RF/LG).

Results showed that the relations among these 15 variables could be reduced to 4 principal components for younger infants and for all infants, and to 5 principal components for older infants. In order to increase the power of the PCA, we present data from the entire group of infants only. Table 2 contains the eigenvalues and loadings (standardized regression

coefficients), which show the correlation between each of the 15 EMG parameters and the first 4 principal components or factors for the combined group of infants. BP-LG, BFr-LG, BFr-RF load most, and positively, on the first factor while BD-RF and BD-BF have a high negative loading. For the second factor, BP-RF loads most positively whereas CO-LG/RF, BD-LG, BD-TA and CO-RF/BF load most negatively. Together, the first 2 factors accounted for 66.9% of the total variation between the 15 EMG parameters. The scores for each of the 5 enhanced sensory conditions and baseline for the first 4 factors are listed in Table 3. Visual flow and friction score most on the first factor, which have the heaviest loadings from BP-LG, BFr-LG and BFr-RF, whereas weights has a high negative score on the first factor. Because BD-RF and BD-BF also have negative loadings on this component there is a positive relationship between these EMG variables and Weights. Baseline has highest positive and Velcro highest negative scoring on the second factor. Baseline is therefore linked to an increase in BP-RF, whereas Velcro is associated with an increase in BD-TA, BD-LG, CO-LG/RF and CO-RF/BF. Unloading scores most on factor 3, associated with high positive loadings from BP-TA, BP-RF, BP-BF, BFr-BF and CO-RF/BF.

The data contained in Tables 2 and 3 can be represented graphically in a biplot. Fig. 8 shows a biplot of factors 1 and 2 with the 6 conditions labeled on the figure. In addition, vectors representing 10 EMG parameters with absolute loadings greater than .3 are superimposed, with the length and direction of the vectors indicating their loading on the first two factors. The angles between the sensory conditions indicate the size of the correlation, with smaller angles signifying greater correlation. Visual flow and friction clearly stand out as closely correlated conditions, whereas weights is negatively correlated with friction, specifically for the impact on BFr-LG, BFr-RF, BP-LG, BD-RF and BD-BF parameters.

4. Discussion

We previously showed that enhancing sensory input for infants with MMC, specifically visual flow and friction, during treadmill trials increased step responses over that of typical baseline treadmill conditions (Pantall et al., 2011). The purpose of this study was to investigate if and how enhancing sensory input modified muscle activation during the step cycle. The significance of investigating infants with MMC was that the myelomeningocele interrupts some of the neural pathways between receptors and effectors in addition to the spinal and supraspinal tracts. One of our aims was to investigate how adaptive damaged neural pathways are to sensory information when generating an organized movement pattern. Our results show that muscle activity was indeed altered, and often increased within stride cycles, with the effect varying depending on the infant's age, the muscle involved and the sensory condition. The kinematic data presented in Figs. 1 and 2 illustrate the high intra-subject and inter-subject variability, which is typically present in developing new motor patterns.

At the first level of analysis, simple activation at any level, we found that muscle activity was present in all infants for all muscles. We found that for 50.2% of the cycle at least one of the 4 muscles was 'on' during baseline condition, which was similar to the 52% reported from a longitudinal study involving infants with MMC (Sansom et al., submitted). Therefore, despite the major spinal trauma and subsequent surgery, these infants still retained some level of distributed neural function at and below their spinal level lesions. This indicates the presence of neuromuscular substrates that can be developed with repeated cycles of sensory input. These rates of activity are lower than the 68.8% recorded from muscles in infants with typical development during treadmill stepping (Teulier, Sansom, Muraszko, & Ulrich, in press). Lesioning of the nervous system early in gestation, which reduces transmission of neural impulses to the muscle fibres and impacts abnormal myogenesis may account for some of the differences between the two population groups

(Sival et al., 2008). These factors may also account for the lack of any activity recorded in a muscle, averaged across all trials, in 24.4% of stride cycles. However, in this study we monitored only four superficial gait muscles. Walking involves the recruitment of many additional muscles that we did not record.

At the other extreme, some muscles in a small number of infants displayed almost constant 'on' activity. One factor may be that lesioned motor pathways involving upper motor neurons resulted in reduced inhibition of the alpha motoneurons, thus increasing activation of the muscle fibers. Additionally, compensatory neuromuscular strategies may be emerging to control leg and pelvic movements, such as greater co-activation of muscles as a reactive strategy to the decreased leg and pelvic stability these infants have. This type of compensatory strategy has been recorded in older individuals with hemiplegia and spinal cord injuries (den Otter, Geurts, Mulder, & Duysens, 2006; Grasso et al., 2004; Lamontagne, Malouin, & Richards, 2001). The effect of increased co-activation is to increase the percentage of the stride cycle that the muscle is active. A further factor could be greater activity of synergists to compensate for neurologically impaired muscles within the same functional group (Maas et al., 2010). In infants with MMC, the myelomeningocele will result in irregular distribution of damage to the lower motor neuron cell bodies in the ventral horn, thus certain functionally related muscles are affected more than other muscles.

The more critical issue for this study was how the enhanced sensory conditions changed this underlying pattern of muscle activity. Our PCA offered insight into the impact of enhanced sensory conditions on fifteen different parameters collectively, rather than individually. Results showed that there was considerable correlation between parameters as they could be reduced from fifteen to just four factors for the entire infant group. The three parameters that had the greatest loadings on the first factor were burst frequency for LG and RF and burst duration for LG. The second factor had the highest loadings from burst duration of TA and LG and co-contraction of LG/RF and RF/BF. Four of the five enhanced sensory conditions and baseline occupied distinctly different quadrants in the PCA biplot, indicating different effects on muscle activation patterns. Visual flow and friction were located in the opposite quadrant to weights whereas Velcro was positioned opposite to baseline. The unloading condition was between baseline and the conditions of visual flow and friction. It is somewhat surprising that Visual flow and friction produce similar effects on muscle patterns given that their receptors are so different. As visual flow and friction scored most on the first factor, we can conclude that the greatest impact these two conditions make is on these three EMG parameters. Both visual flow and friction produced increased step response, as we reported previously (Pantall et al., 2011). Therefore our results would suggest that the early improvements in patterned step motions are linked to changes in activation of LG in particular. This hypothesis is supported by the observation that weights, which we previously reported was associated with a decrease in normalized step frequency compared to baseline, had opposite loadings on the first two factors compared to visual flow and friction (Pantall et al., 2011). Weights loaded negatively on the first factor and was associated with increased burst duration of RF and BF. Statistical analyses revealed that weights significantly reduced burst frequency of LG by over 50% compared to baseline. This unexpected result may reflect that the increased afferent input arising from the condition of weights had an inhibitory effect on the motor neurons to LG thereby decreasing burst frequency. Alternatively, the additional weight may simply have made the challenge to muscle strength too great to overcome. In fact, the increased burst duration of RF and BF suggests a strategy shift toward greater activity of more proximal and larger muscles to overcome the increased load on the leg. The reduced activity of LG and weak refinement of motor patterns in proximal muscles indicated by long burst durations may help explain our previous finding that weights reduced the stepping response. Velcro, although not represented highly on the first factor, had the highest absolute score on the second factor,

associated with increased burst duration of TA and LG and co-contraction of LG/RF and RF/BF. The effect of Velcro was therefore to increase the burst duration of the distal lower limb muscles in addition to increasing co-contraction of two of the three agonist-antagonist muscle pairs. These may have emerged as a solution to the need for a greater force to pull the foot away from the sticky surface of the treadmill. Velcro, although it did increase motor activity and vertical displacement, did not generate greater independent motor activation patterns which are associated with greater step response. A previous study on infants with Down syndrome reported that Velcro promoted the greatest increase in step frequency compared to baseline and other sensory enhancements (Ulrich et al., 1998). The difference between these studies' results may lie in the differences in underlying neurophysiology between the two groups of infants. The infants with Down syndrome were older and may have been stronger. Further, infants with Down syndrome, unlike infants with MMC, show no specific interruptions in peripheral neural signal transmission.

While our findings clearly demonstrate the effect of varying the sensory input on muscle activity patterns, the question is why, specifically, visual flow did not increase step frequency for young infants, or significantly change kinematic parameters for the whole infant group? Studies indicate that infants are able to transform visual information within an egocentric frame of reference early on (Brosseau-Lachaine, Casanova, & Faubert, 2008; Gilmore et al., 2007), with neonates demonstrating an increased air step frequency when presented with selected types of visual flow stimuli (Barbu-Roth et al., 2009). The mechanism for the earliest response to visual flow may involve subcortical circuitry projecting to the ventral horn cells rather than the visual cortical areas V5/MT⁺ which do not develop until 2–3 months of age (Banton & Bertenthal, 1997). Another reason, why specifically visual flow had a different impact between the two age groups, is that younger infants may have failed to perceive the implied motion or failed to attend to it. A previous study has demonstrated that infants, who looked at the belt more, stepped more frequently (Moerchen & Saeed, in press). Additionally, we may not have elicited greater step frequencies in response to visual flow in younger infants because control of the muscle most impacted by visual flow, LG, may simply not be sufficiently refined in younger infants to generate an organized response. This muscle in young infants was activated on average 42.3% of the cycle in young infants compared to only 16.2% in older infants. A further consideration is that air stepping requires less muscle force to raise the limb than stepping on a treadmill, where an increased anterior force is needed to counteract the posterior force applied to the foot by the moving treadmill. In summary, although younger infants were able to translate the visual input into a motor unit activity, they were unable to further develop the response into an organized patterned movement.

Our results also demonstrated that the impact of enhanced sensory input on muscle activity was modified by age. Similar changes as a consequence of intrinsic subsystem development over time have been observed in infants' treadmill stepping behavior with an increase in steps and more regular inter-limb coordination patterns as the infant develops (Teulier et al., 2009). Why do enhanced sensory conditions cause different patterns of muscle activity in younger versus older infants? One explanation is that older infants' muscles are stronger and sensory systems are better developed, for example the visual pathways. One muscle activity characteristic we observed between the two age groups was that for younger infants, the state of all four gait muscles being 'on' simultaneously was greater for younger infants. This finding has also been observed in young infants with TD (Teulier et al., in press). When muscles are weaker, a simple system response to affect movement may be to call on more muscles to create motion. Further, infants may activate many muscles in order to discover which combinations are most efficient or effective in producing the desired outcome.

A final observation is that changes in EMG patterns resulting from enhanced sensory input were not reflected by corresponding changes in kinematic patterns. This finding emphasizes the deeper level of information related to underlying neurophysiological mechanisms that can be gained from EMG analysis.

5. Conclusion

In this study we demonstrated that enhancing different sensory modalities for infants with MMC when supported on a treadmill elicited different muscle activation patterns to culminate in steps, supporting our first hypothesis. Our infants with myelomeningocele all had some level of disruption to the neural pathways modulating sensory and motor activity in the lower limbs. The significance of our results is that even in the presence of lumbar or sacral lesions, the infants' nervous systems still maintained the capacity to process sensory input and generate motor output, demonstrating the importance of extrinsic and intrinsic subsystems in moulding motor patterns. The lack of stereotypic muscle activation patterns is suggestive of a certain exploratory randomness in muscle recruitment, as proposed by DST. The variability indicates the numerous ways that infants of all ages have to access the same patterned movement, in this case, stepping. Exposure to repetitive sensory input linked to subsequent functionally relevant motor output in the form of treadmill training may therefore promote development of neural circuitry associated with stepping behavior at all levels. The type of muscle activity produced was also related to degree of change in step frequency. Visual flow and friction, which produced greatest increases in step frequency, promoted activity primarily of LG, as we stated in our hypotheses. In contrast, Weights, which previously we reported to decrease stepping response, increased activity of the proximal muscles, RF and BF. Further, in younger infants all conditions increased the probability of muscle activity compared to baseline, while for older infants visual flow and unloading most clearly increased probability compared to baseline. Velcro increased muscle activity, in particular the distal muscles, in addition to increasing co-activation. Weights increased burst duration of the proximal muscles. Thus, even with diminished sensorimotor properties, infants with MMC show tremendous and non-stereotypic adaptability to dynamic sensory motor contexts. In conclusion, results from our study reveal in this young neurologically impaired population at the most critical developmental period of ex utero life, the remarkable plasticity in the nervous system and consequences of different types of sensory input on motor pattern generation with significant clinical implications.

6. Limitations

In our subject group, correct positioning of the electrodes could not be confirmed by asking the subject to perform a contraction of the specific muscle nor was ultrasound applied to determine the underlying muscle structures. However, as we performed careful palpation of bony landmarks in addition to the muscle belly we are confident of achieving reasonable placement. Cross-talk is another issue which given the small size of the muscles we cannot overlook. The threshold we used to determine when a muscle was activated was two standard deviations above the mean. This threshold has been used in adults and there is some evidence for successful application in infants. Visual inspection of signals showed this threshold corresponded well to subjective determination of a muscle being active (see Fig. 2). Trials were only 30 s long, which arguably may not have provided sufficient time for the infant to adapt. However, the total time period that the infant was supported on the treadmill was six minutes, close to the time limit tolerated by the infant. Another consideration is that we only analyzed a few steps, varying in number between individuals and in type (alternating or single). This reflects the variability in the infants' development as well as the differing severity of the myelomeningocele on neurophysiological function. The study did not include a control group of TD infants and we were therefore unable to state how the

response to sensory input in our infant cohort corresponded to that which might be observed in infants with no neurological deficit. However, the aim of our study was to determine if infants with a dysfunction of the spinal cord can respond to enhanced sensory input which does not require a control group.

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Fig. 1. Exemplar, three strides for one 9 month-old infant with myelomeningocele. Cycle durations begin with toe off; vertical lines identify touchdown.

Fig. 2.

Mean displacement of toe marker in swing, across enhanced sensory conditions, in (a) vertical, (b) anterior-posterior, (c) medial-lateral, and (d) cumulative displacement in 3D. BL – Baseline; Vis – Visual Flow; UL – Unloading; W – Weights; Vel – Velcro; F – Friction.

Fig. 3.

Example of smoothed, rectified EMG trace for one 30 s test trial in baseline for the tibialis anterior muscle of a 9 month-old infant. Objectively determined (via our algorithm) on-off bursts and also stride cycle events are superimposed. TO – toe-off; TD – toe down.

Fig. 4. Probabilities of tibialis anterior (TA), gastrocnemius (LG), rectus femoris (RF) and biceps femoris (BF) being 'on' during a stride cycle for the younger age group and older age group. TO – toe-off; TD – toe down; ES – end-stance.

Fig. 5. Mean burst frequency during a stride cycle across all infants by condition. Significant difference compared to baseline ($p < .05$).

Fig. 6. Mean burst duration during a stride cycle across all infants by condition. Significant difference compared to baseline ($p < .05$).

Fig. 7. Percent of activation of each of the 16 possible muscle states during stride cycles, collapsed over babies, by condition, and by age: (a) 2–5-month olds and (b) 7–10-month olds.

Fig. 8. Biplot of the First Principal Component and Second Principal Component for infants aged 2–10 months for Baseline and the 5 enhanced sensory conditions with superimposed loadings with absolute values $>.3$ from EMG parameters for tibialis anterior (TA), lateral gastrocnemius (LG), rectus femoris (RF) and biceps femoris (BF).

Table 1

Number and type of steps analyzed for each infant per condition.

Infant	Baseline	Visual Flow	Unloading	Weights	Velcro	Friction
1	5 (2as + 3s)	5 (5s)	5 (2as + 3s)	4 (2as + 2s)	5 (5s)	5 (5as)
2	3 (3s)	3 (3s)	1 (1s)	0	2 (2s)	1 (1s)
3	2 (2s)	3 (3s)	4 (2as + 2s)	4 (4s)	4 (4s)	3 (3s)
4	2 (2s)	2 (2s)	2 (2s)	2 (2s)	1 (1s)	3 (3s)
5	2 (2s)	3 (3s)	1 (1s)	3 (2as + 1s)	0	4 (4s)
6	4 (4s)	1 (1s)	3 (3s)	1 (1s)	2 (2s)	4 (4s)
7	1 (1s)	2 (2s)	0	1 (1s)	2 (2s)	3 (3s)
8	5(5as)	4 (4s)	5 (5s)	5 (5s)	5 (5s)	5 (5s)
9	1 (1s)	3 (3s)	1 (1s)	1 (1s)	0	2 (2s)
10	5 (5s)	0	0	0	2 (2s)	5 (5s)
11	2 (2s)	2 (2s)	3 (3s)	5 (5s)	5 (5as)	5 (4as + 1s)
12	5 (5as)	5 (3as + 2s)	2 (2as)	2 (2s)	4 (4as)	5 (5s)
13	0	0	3 (3s)	0	3 (3s)	0
14	4 (4s)	5 (5s)	3 (3s)	5 (5s)	3 (2as + as)	1 (1s)
15	5 (5as)	5 (5as)	4 (4as)	5 (5as)	5 (5as)	5 (5as)
16	2 (2s)	1 (1s)	5 (5s)	1 (1s)	2 (2s)	0
17	0	4 (4as)	0	0	4 (4s)	5 (5as)
18	5 (5as)	5 (5as)	4 (4as)	5 (5as)	5 (5as)	5 (5as)
19	0	1 (1s)	0	2 (2as)	0	3 (2as + 1s)

as – alternating step; s – single step.

Table 2

Loading and eigenvalues of factors 1–4. Shaded values indicate significant loading values.

	Factor 1	Factor 2	Factor 3	Factor 4
BP-TA	-.10	-.29	.49	-.03
BP-LG	.38	-.09	-.02	-.15
BP-RF	.07	.34	.39	.24
BP-BF	-.19	.15	.41	-.37
BFr-TA	.23	-.19	-.01	.47
BFr-LG	.39	.05	-.06	.06
BFr-RF	.39	-.01	.01	-.10
BFr-BF	.26	-.07	.46	-.07
BD-TA	.14	-.35	-.06	-.46
BD-LG	-.20	-.44	.02	-.02
BD-RF	-.35	.00	-.05	.21
BD-BF	-.34	-.10	-.21	.11
CO-RF/BF	-.22	-.33	.33	.07
CO-TA/LG	.16	-.26	.16	.52
CO-LG/RF	.11	-.47	-.19	-.06
Eigenvalue (%)	6.3 (42.1%)	3.7 (24.8%)	2.5 (16.6%)	1.8 (12.0%)

BP – burst percent ‘on’; BFr – burst frequency; BD – burst duration.

TA – tibialis anterior; LG – lateral gastrocnemius; RF – rectus femoris; BF – biceps femoris.

Table 3

Representation of enhanced sensory conditions in Principal Component space. Shaded values indicate significant loading values.

	Factor 1	Factor 2	Factor 3	Factor 4
Baseline	−0.61	2.59	−0.65	−1.14
Visual flow	2.12	−0.98	−0.39	−1.91
Unloading	1.22	1.01	2.89	0.68
Weights	−4.16	0.36	−0.46	0.52
Velcro	−1.14	−3.13	0.39	0.03
Friction	2.57	0.15	−1.78	1.82