Cardiovascular Response of Individuals with Spinal Cord Injury to Dynamic Functional Electrical Stimulation under Orthostatic Stress

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***Abstract*—In this pilot study, we examined how effectively functional electrical stimulation (FES) and passive stepping mitigated orthostatic hypotension in participants with chronic spinal cord injury (SCI). While being tilted head-up to 70 degrees from the supine position, the participants underwent four 10-minute conditions in a random sequence: 1) no intervention, 2) passive stepping, 3) isometric FES of leg muscles, and 4) FES of leg muscles combined with passive stepping. We found that FES and passive stepping independently mitigated a decrease in stroke volume and helped to maintain the mean blood pressure. The effects of FES on stroke volume and mean blood pressure were greater than those of passive stepping. When combined, FES and passive stepping did not interfere with each other, but they also did not synergistically increase stroke volume or mean blood pressure. Thus, the present study suggests that FES delivered to lower limbs can be used in individuals with SCI to help them withstand orthostatic stress. Additional studies are needed to confirm whether this use of FES is applicable to a larger population of individuals with SCI.**

***Index Terms*—Functional electrical stimulation, spinal cord injury, orthostatic hypotension, rehabilitation engineering.**

I. Introduction

Orthostatic hypotension is a condition characterized by symptoms of cerebral hypoperfusion, such as headache, lightheadedness, and dizziness [1,2]. It occurs due to gravitational venous pooling and a subsequent decrease in the circulating blood volume. Most of the pooling occurs within ten seconds of standing up from the supine position; after three to five minutes, the pooling is almost complete [3]. The pooling reduces venous return and increases the intra-capillary pressure, which causes the fluid to escape into the interstitial space [3,4]. If a person continues to stand still, the arterial pressure may eventually decrease enough to cause hypoperfusion in the brain. Normally, such orthostatic stress activates compensatory mechanisms that maintain arterial pressure. However, in individuals with spinal cord injury (SCI), impaired sympathetic cardiovascular control makes orthostatic hypotension more likely to occur. Furthermore, the symptoms of orthostatic hypotension can diminish mobility after SCI, especially in early stages of rehabilitation [5].

Pharmacological interventions against orthostatic hypotension use agents that either increase the sympathetic stimulation of the cardiovascular system or increase the blood volume in circulation, the latter of which is usually the first goal [6,7]. If an individual's orthostatic tolerance does not improve after replenishing the circulating blood volume, the use of sympathomimetic agents may be considered [6]. Some sympathomimetic agents can cause undesirable side effects, such as tachycardia and hypertension, and some agents have not demonstrated efficacy with adequate sample sizes [6,7]. In any case, mitigating orthostatic hypotension with a pharmacological agent requires planning before a person is subjected to orthostatic stress.

Careful planning of pharmacological interventions should also consider concurrent non-pharmacological ones. Non-pharmacological interventions include increased salt and fluid intake to increase the circulatory volume, applying external compression to reduce venous pooling in the abdomen or the lower extremities, applying functional electrical stimulation (FES) on the lower extremity muscles to induce venous return, and exercising, either as training to improve orthostatic tolerance or as an intervention during orthostatic stress [7,8]. Among these interventions, FES is unique because it can generate dynamic muscle contractions in the legs, much like the skeletal muscle pump in able-bodied individuals. Also, the stimulation parameters can be customized easily and immediately to optimize the effect of FES. Thus, an intervention with FES is worth investigating.

Isometrically stimulating the leg muscles can help maintain arterial pressure of individuals with SCI during passive standing or graded tilting [9-12]. Our group has found that, in able-bodied individuals, applying FES with passive leg movements is even better for improving orthostatic tolerance [13]. Furthermore, the combined intervention of FES and passive leg movements most probably induces greater venous return than isometric FES [14]. The benefit of leg movements has been suggested by other studies, which showed that passive

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or FES-induced leg cycling increased venous return or cardiac output in individuals with SCI [15-18]. To our knowledge, the combined effect of FES and passive leg movements on orthostatic hypotension has only been studied in able-bodied individuals. If we can achieve a similar synergy in individuals with SCI, FES intervention may become a viable means to treat orthostatic hypotension in this patient population.

We hypothesized that FES with passive leg movements should mitigate orthostatic hypotension in individuals with SCI by inducing venous return, thereby helping to maintain sufficient arterial pressure. Furthermore, we hypothesized that the combined intervention will be more effective than applying FES or passive leg movements separately. To test our hypothesis, we examined the cardiovascular responses of individuals with SCI during passive stepping and FES of the leg muscles (applied together or separately while the participants were tilted head-up to 70 degrees from the supine position).

TABLE I
DESCRIPTIONS OF STUDY PARTICIPANTS

Participant	Sex	Age (years)	Height (cm)	Weight (kg)	AIS	Level of injury	Years since injury
A	F	47	157	91	A	T6	6
B	F	38	170	77	B	T2	11
C	M	37	170	77	C	C4	13
D	M	57	183	110	A	C6	3
E	F	48	157	64	D	C5	1
F	M	35	183	81	A	T5-T7	17
G	M	27	191	133	B	C5	12
H	F	34	168	68	B	T4	2
I	M	59	171	84	A	T3-T4	5
J	M	53	172	85	A	T3	29
Mean	N/A	43.5	172	87.0	N/A	N/A	9.90
SD	N/A	10.8	11	20.6	N/A	N/A	8.56

AIS stands for American Spinal Injury Association (ASIA) Impairment Scale. The bottom two rows show the average values with standard deviations (SD). Females and males are indicated by the letters F and M, respectively.

II. MATERIALS AND METHODS

A. Participants

Ten individuals with chronic SCI participated in the experiment (TABLE I). Participants were recruited by convenience sampling from the population of outpatients at the Lyndhurst Centre of the Toronto Rehab. We approached all interested candidates with their consent, and their eligibility was determined by their physiatrists. As an inclusion criterion, the level of injury was specified as T6 or above. SCI at T6 or above would impair sympathetic vasoconstriction in the lower extremities [19]. Although SCI between T1 and T6 would leave some sympathetic stimulation of the heart intact, postural tachycardia does not protect individuals from orthostatic hypotension [20]. Also, without an increase in venous return, increased myocardial contraction would not be very effective in maintaining blood pressure. Thus, this criterion maximized the pool of potential participants while ensuring their predisposition to orthostatic hypotension. They did not have brittle diabetes, chronic renal failure, history of cardiovascular diseases, or peripheral nerve damage in the lower extremities. They were instructed not to consume caffeine, nicotine, or alcohol for eight hours before the experiment; they were also instructed not to consume fluids for two hours before the experiment. Seven participants consumed a light meal 2.6 ± 1.3 hours before the experiment. All experiments were conducted at a normal ambient temperature, between 9 a.m. and 5 p.m. Before the experiment, each participant gave written informed consent. The method of recruitment, the criteria for eligibility, and the experimental procedures were approved by the Research Ethics Board of the Toronto Rehabilitation Institute.

B. Protocol

Each participant underwent four experimental conditions: 1) passive head-up tilt (HUT) without any interventions, 2) passive stepping (STEP), 3) isometric FES (IFES), and 4) dynamic FES (DFES), which was a combination of FES and passive stepping. There were 24 possible sequences for applying four conditions without repetition. For each participant, a random sequence was selected using the random permutation function in a commercial numerical computing environment (MATLAB Version 7.10, The MathWorks, Inc., USA). The 10-minute supine rest between each condition should have reduced any residual effects of the previous experimental condition to a negligible level [21,22].

During each condition, participants were tilted head-up to 70 degrees from the supine position on a tilt table with a motorized stepper. This was done because 70-degree head-up tilt can frequently induce orthostatic hypotension in able-bodied individuals. Each condition lasted 10 minutes and was preceded by a 10 minutes of rest in the supine position. Tilting up before each condition took approximately 23 seconds, and tilting down after each condition took approximately 17 seconds. During the experiment, the participants were asked to report any symptoms of orthostatic hypotension, such as headache, dizziness, and lightheadedness. During the last minute of each condition, the inferior vena cava was imaged in the transverse plane. The electromyographic (EMG) signals of the leg muscles were recorded only during STEP because cyclic passive movements of the legs can induce rhythmical EMG activities. Throughout the experiment, beat-to-beat blood pressure was recorded non-invasively.

Tilt Table with a Motorized Stepper

We used a tilt table with a motorized stepper (Erigo, Hocoma AG, Switzerland). During STEP and DFES, the motorized mechanism of the table rotated the participants' hips in the sagittal plane. The resulting movement was rhythmical stepping that alternated between the left and right legs at 40 steps (or 20 cycles) per minute. During the first 50% of a cycle, the hip was flexed by 20 degrees from the neutral position (neutral position being the one where the hip was straight). During the next 50% of a cycle, the hip was returned to the neutral position. For more details on the tilt table, refer to [23] and [24].

Functional Electrical Stimulation

FES was applied on four muscle groups: 1) the tibialis anterior, 2) hamstring, 3) quadriceps femoris, and 4) triceps surae muscles of both legs. We used current-regulated programmable transcutaneous FES systems (Compex Motion, Compex SA, Switzerland). The stimulation was bipolar and biphasic, with a maximum pulse width of 300 μs and stimulation frequency of 40 Hz. The frequency of 40 Hz was chosen to ensure tetanic contractions (i.e., continuous and smooth muscle contractions) without early fatigue [25].

To determine the stimulation amplitude, we measured the motor threshold and the maximum contraction amplitude for each muscle group. The threshold was measured as the lowest amplitude that induced a visually detectible or palpable muscle contraction. The maximum contraction amplitude was measured as the lowest amplitude to induce maximum contraction or as the maximum tolerable stimulation amplitude. During the experiment, the muscles were stimulated at 70% of the maximum contraction amplitude to ensure muscle contraction (TABLE II). If 70% of the maximum amplitude was less than the threshold, the threshold was used for stimulation.

Two surface electrodes (ValueTrode, Axelgaard Manufacturing Co., Ltd., USA) were placed on the proximal and distal ends of each muscle group. We used 5-cm-by-5-cm electrodes for the tibialis anterior and 5-cm-by-9-cm electrodes for other muscle groups. Conductive gel (Spectra 360 Electrode Gel, Parker Laboratories, Inc., USA) was applied on the interface between the skin and each electrode before securing the electrodes with surgical tapes.

During IFES, stimulation alternated between the thigh and shank muscles. During stimulation, the pulse width was linearly increased from 0 to 300 μs in 0.3 seconds, held constant for 0.9 seconds, and linearly decreased to 0 μs in 0.3 seconds. Then, the stimulation ceased for 0.9 seconds before increasing the pulse width again. The periods of stimulation and no stimulation were staggered so that 0.9 seconds of a constant pulse width coincided with 0.9 seconds of no stimulation between the thigh and shank muscles. Similar protocols had been used in studies that investigated the efficacy of isometric FES against orthostatic hypotension in individuals with SCI [9-12].

During DFES, each muscle group was stimulated while the muscles shortened from passive stepping. To estimate the dynamic length of each muscle group during stepping, we simulated the movement using a commercial simulation environment (Simulink Version 7.4 and SimMechanics Version 3.1.1, The MathWorks, Inc., USA). The lower extremity was modeled as a series of rigid bodies connected by rotational joints (i.e., hip, knee, and ankle), whose axes were perpendicular to the sagittal plane. Passive stepping was simulated as a multi-joint movement, with the foot rotating about a fixed axis (also perpendicular to the sagittal plane) near the toes, and the hip joint fixed to the inertial reference [23,24]. The simulation calculated the dynamic ankle, knee, and hip angles during passive stepping. Then, these angles were used to estimate the dynamic lengths of the leg muscles [26]. We estimated that the tibialis anterior and the quadriceps femoris would shorten while the hip rotated toward the neutral position. Conversely, the hamstring and triceps surae muscles would shorten during hip flexion. Therefore, during DFES, the tibialis anterior and quadriceps were stimulated while the hip rotated toward the neutral position (50% of a cycle), with the same linear increase, maintenance, and linear decrease of the pulse width as those of IFES. During hip flexion (50% of a cycle), they were not stimulated. The on-off pattern of stimulation was reversed for the hamstring and triceps surae muscles.

TABLE II
MEAN STIMULATION AMPLITUDES FOR FES

Muscle	Left leg [mA]	Right leg [mA]
Triceps surae muscle	42.0 ± 8.1	46.4 ± 10.6
Tibialis anterior	42.4 ± 13.7	42.0 ± 12.9
Hamstring	44.1 ± 9.8	51.0 ± 11.7
Quadriceps	48.7 ± 13.3	50.9 ± 14.9

Blood Pressure Monitoring

We used a non-invasive cardiovascular monitoring system (Nexfin, BMEYE B.V., Netherlands) that measured the beat-to-beat pressure in the finger artery via an inflatable cuff, which was worn by the participants over the intermediate phalanx bone of the right middle finger. Using the volume-clamp method [27,28] and the physiologic calibration algorithm [29], the monitoring system reconstructed beat-to-beat brachial pressure from the finger artery pressure. The brachial pressure was measured in millimeters of mercury (mmHg) and expressed as systolic, diastolic, and mean blood pressures. The system also calculated the heart rate in beats per minute, stroke volume in mL, and systemic vascular resistance in dyn·s/cm^5.

EMG Recording

We assessed the muscle activity of each muscle group during STEP to examine whether the induced stepping was purely passive or not. Surface EMG signals were recorded from the bellies of the medial gastrocnemius, tibialis anterior, biceps femoris, and rectus femoris during STEP (EMG_{STEP}) and during the supine rest (EMG_{REST}). Before placing the electrodes, hair was removed using a razor. Then, light abrasion was applied to remove dead skin cells, and the abraded skin was cleansed using alcohol wipes. We used disposable Ag-AgCl surface electrodes (BiPole, Bortec Biomedical Ltd., Canada). Each electrode had a diameter of 10 mm and was placed in a bipolar configuration for each muscle, with a center-to-center electrode distance of 18 mm. The signals from the electrodes were amplified by 5,000 using an EMG measurement system (AMT-8, Bortec Biomedical Ltd., Canada) with a frequency response of 10 to 1,000 Hz. The analog output of the EMG measurement system and the phase signal of the motorized stepper were digitized at 2,000 Hz using a data acquisition system (PowerLab/12SP, ADInstruments, Australia).

Ultrasound Imaging of the Inferior Vena Cava

We assessed the intravascular circulatory blood flow by

measuring the cross-sectional area of the inferior vena cava (CSA_{IVC}) with a diagnostic ultrasound system (ACUSON X150, Siemens AG, Germany). A transducer (CH5-2, Siemens AG, Germany) was placed on the subcostal area of the chest, approximately 2 cm inferior to the xiphoid process of the sternum. In this position, the inferior vena cava was viewed posteriorly to the liver at 5 MHz [14,30,31]. The same experimenter performed the ultrasound imaging for all participants. The video output of the ultrasound system was recorded onto the hard drive of a computer via a video capture device (Dazzle Video Creator Plus HD, Pinnacle Systems, USA) at 30 frames per second.

C. Data Analysis

The experimental data were processed in a commercial numerical computing environment (MATLAB Version 7.10, The MathWorks, Inc., USA) using a signal processing toolbox (Signal Processing Toolbox Version 6.13, The MathWorks, Inc., USA). Statistical analysis was performed using a statistics toolbox (Statistics Toolbox Version 7.3, The MathWorks, Inc., USA). The significance level for the statistical analysis was set to 5%.

Cardiovascular Responses

We conducted a series of statistical analyses using the binned differences (ΔSBP, ΔDBP, ΔMBP, ΔHR, ΔSV, and ΔSVR) of each participant (SBP is systolic blood pressure, DBP is diastolic blood pressure, MBP is mean blood pressure, HR is heart rate, SV is stroke volume, and SVR is systemic vascular resistance). First, baseline values were calculated as an average during the last minute of supine rest before each condition. Then, the cardiovascular parameters were binned by each minute of experimental conditions. Lastly, the binned parameters were expressed as a difference from the corresponding baseline value.

At each bin, we conducted two-tailed one-sample *t*-tests. The *t*-tests assessed if the cardiovascular parameters changed significantly from the baseline during experimental conditions.

To evaluate the effects of FES and passive stepping over time, we used three-way repeated-measures analysis of variance (ANOVA). The independent variables were 1) the presence or absence of FES in a condition, 2) the presence or absence of passive stepping in a condition, and 3) time (ten levels, one for each bin that represents each minute of an experimental condition).

To compare the cardiovascular responses between conditions, we conducted two-way repeated-measures ANOVA with Tukey's HSD post hoc tests. The independent variables were 1) the type of experimental condition (four levels) and 2) time (ten levels).

To assess whether participants experienced orthostatic hypotension during any of the experimental conditions, we averaged systolic and diastolic blood pressures (separately) every ten heartbeats during the first three minutes of each condition. If systolic blood pressure decreased by 20 mmHg or more from the baseline, or if diastolic blood pressure decreased by 10 mmHg or more from the baseline, the decrease qualified as orthostatic hypotension [1,5]. No statistical analysis was performed with the number of episodes.

EMG Signals of the Leg Muscles

To evaluate whether EMG_{STEP} indicated muscle activity, we conducted paired *t*-tests for each muscle. We calculated the sum of squared frequency components between 10 and 250 Hz for unprocessed EMG_{STEP} and EMG_{REST}. The sums excluded the frequency components between 59 and 61 Hz to eliminate the effect of electric hum. For each muscle, a paired *t*-test compared the group average of the sums between EMG_{STEP} and EMG_{REST}.

Cross-sectional Area of the Inferior Vena Cava (CSA_{IVC})

CSA_{IVC} was used as a surrogate measure of venous return. Due to the distensibility of the inferior vena cava, we assumed that CSA_{IVC} would change in proportion to the venous volumetric flow into the right atrium [14]. This assumption is supported by the strong correlation between the diameter of the inferior vena cava and the central venous pressure in anesthetized individuals undergoing elective cardiac surgery [32,33].

Using image-processing software (ImageJ, National Institute of Health, USA), instantaneous CSA_{IVC} was calculated by a blinded investigator, who did not know which experimental condition corresponded to each image sequence.

III. Results

A. Cardiovascular Responses

Comparing Cardiovascular Responses to the Baseline

Fig. 1 shows the average responses of each cardiovascular parameter and the results of the *t*-tests. The letters, 'H', 'S', 'I', and 'D', which respectively represent HUT, STEP, IFES, and DFES, indicate that the corresponding value was significantly different from the baseline. Heart rate was significantly higher than the baseline at the majority of bins during HUT, STEP, and DFES. Systolic blood pressure decreased significantly during HUT and increased significantly during DFES, especially toward the end. Diastolic blood pressure increased significantly during STEP and DFES. Mean blood pressure increased significantly only during DFES. Stroke volume was significantly lower than the baseline throughout HUT and STEP. Systemic vascular resistance did not change significantly during any of the conditions.

Main Effects and Interactions of FES, Passive Stepping, and Time

The results of the three-way ANOVA are shown in TABLE III and TABLE IV. FES significantly affected ΔHR, ΔSBP, ΔMBP, and ΔSV (changes in heart rate, systolic and mean blood pressures, and stroke volume). On average, heart rate increased less with FES than without FES. Also, on average, systolic and

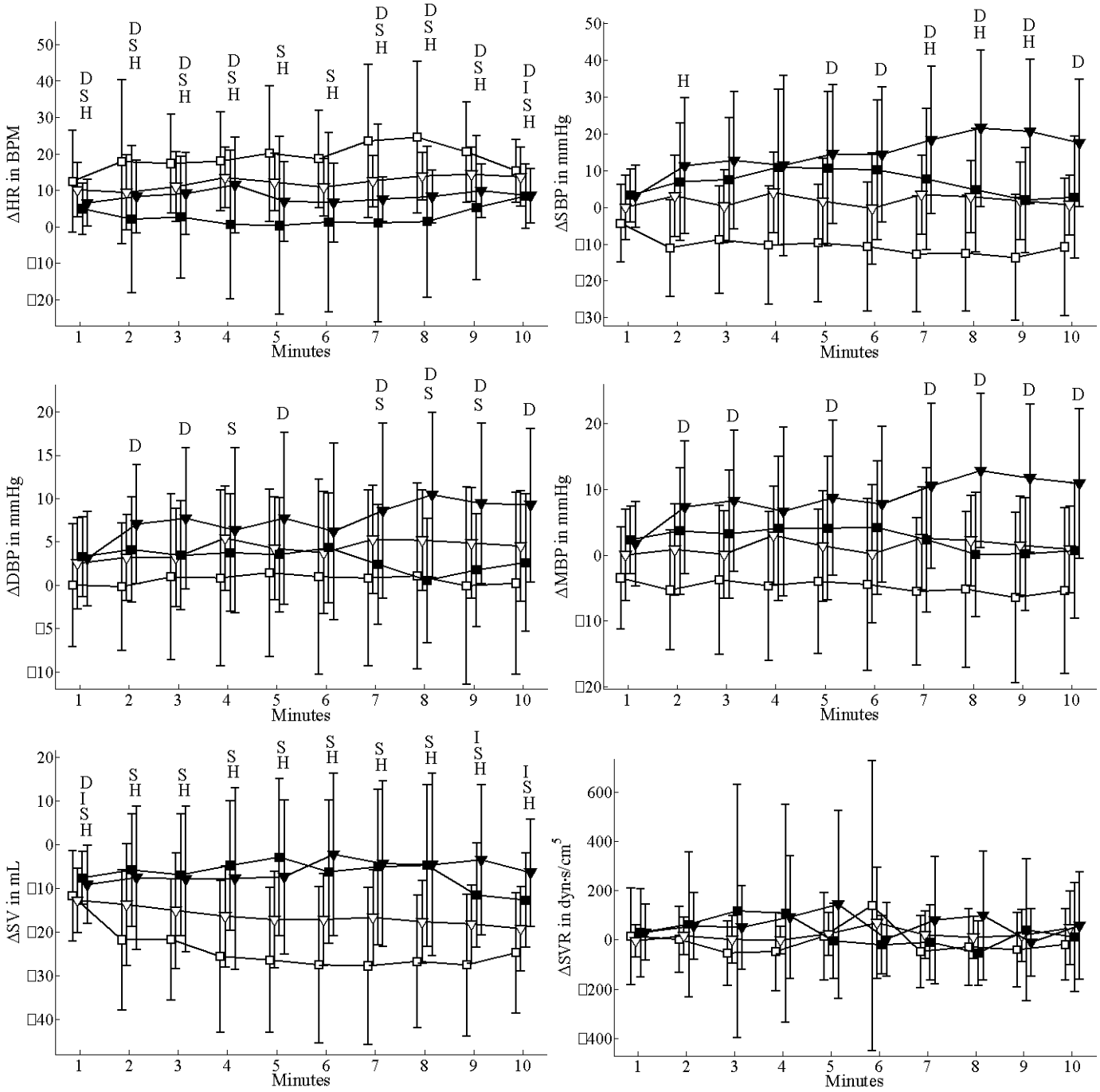

Fig. 1. Average cardiovascular responses. Positive values indicate an increased from the baseline. Error bars indicate the standard deviations between participants. □ represents HUT, ▽ represents STEP, ■ represents IFES, and ▼ represents DFES. The letters, 'H', 'S', 'I', and 'D', which respectively represent HUT, STEP, IFES, and DFES, indicate that the corresponding values were significantly different from the baseline. The line graphs have been staggered in the horizontal direction for better visualization. ΔHR, ΔSBP, ΔDBP, ΔMBP, ΔSV, and ΔSVR are changes in heart rate, systolic blood pressure, diastolic blood pressure, mean blood pressure, stroke volume, and systemic vascular resistance, respectively. BPM is beats per minute.

mean blood pressures increased more, and stroke volume decreased less with FES than without FES. Passive stepping significantly affected ΔSBP, ΔDBP, and ΔMBP (changes in systolic, diastolic, and mean blood pressures): on average, systolic, diastolic, and mean blood pressures increased more with passive stepping than without it. Time did not independently affect any of the cardiovascular parameters.

FES and passive stepping did not interact significantly. However, there was a significant interaction between FES and time for ΔSBP and ΔSV. Likewise, passive stepping and time interacted significantly for ΔSBP, ΔDBP, ΔMBP, and ΔSV.

Comparison between Experimental Conditions

TABLE V shows the results of the two-way ANOVA and post hoc tests with the cardiovascular parameters. There were significant differences between the conditions for all binned differences, except for ΔSVR (change in systemic vascular resistance). Compared to the other three conditions, HUT resulted in higher ΔHR and lower ΔSBP, ΔDBP, ΔMBP, and ΔSV. DFES had the highest ΔSBP, ΔDBP, and ΔMBP. The

TABLE III
MAIN EFFECTS OF FES, PASSIVE STEPPING, AND TIME ON CARDIOVASCULAR RESPONSES

Cardiovascular parameter	FES		Passive stepping		Time	
	$F_{(1,9)}$	p	$F_{(1,9)}$	p	$F_{(9,81)}$	p
ΔHR	7.07	0.026*	0.089	0.772	1.57	0.139
ΔSBP	14.3	0.004*	10.9	0.009*	0.607	0.788
ΔDBP	3.34	0.101	8.30	0.0182*	0.648	0.753
ΔMBP	12.5	0.006*	10.5	0.0102*	0.498	0.872
ΔSV	15.6	0.003*	2.35	0.159	1.59	0.131

mean cardiovascular parameters during each experimental condition can be compared in TABLE VI.

Observed Episodes of Orthostatic Hypotension

Based on the changes in blood pressure alone, we determined the number of participants that experienced orthostatic hypotension during the experiments: 6 during HUT, 5 during STEP, 4 during IFES, and 3 during DFES. Despite these findings, none of the participants reported perceived symptoms of orthostatic hypotension during the experiments.

B. EMG Signals of the Leg Muscles

The paired *t*-test failed to reject the null hypothesis for all muscles. In other words, for all muscles, the spectral power of EMG_{STEP} did not significantly differ from that of EMG_{REST} between 10 and 250 Hz.

C. Cross-sectional Area of the Inferior Vena Cava (CSA_{IVC})

Ultrasound imaging was successfully completed for 4 participants (*A*, *C*, *D*, and *I*). For the remaining 6 participants, the inferior vena cava could not be located while the participants were in the tilted position. Of the 4 participants, images for 2 participants (*D* and *I*) were clear enough to warrant further analysis.

For participant *D*, 53.9% of the images from HUT, 4.33% of the images from STEP, and 40.9% of the images from IFES could not be analyzed due to poor quality. Likewise, 25.4% of the images from HUT, 25.4% of the images from STEP, 30.3% of the images from IFES, and 41.3% of the images from DFES could not be analyzed for participant *I*. Based on the high percentages of images with poor quality, we did not conduct further analysis. Therefore, subsequent discussions regarding venous return are based on stroke volume, which was calculated by the beat-to-beat cardiovascular monitoring system. We did not make any inferences from the ultrasonography data.

IV. DISCUSSION

A. Cardiovascular Responses

Our results demonstrated that FES and passive stepping significantly affected the cardiovascular responses during a head-up tilt. During conditions with FES (i.e., IFES and DFES), on average, heart rate increased less, systolic and mean blood pressures increased more, and stroke volume decreased less (Fig. 1, TABLE III). During conditions with passive stepping (i.e., STEP and DFES), on average, systolic, diastolic, and mean blood pressures increased more (Fig. 1, TABLE III). These results imply that FES and passive stepping independently increased blood pressure, but FES also mitigated tachycardia and a decrease in stroke volume.

As TABLE IV shows, we found significant interactions between FES and time for ΔSBP and ΔSV (changes in systolic blood pressure and stroke volume, respectively) and between passive stepping and time for ΔSBP, ΔDBP, ΔMBP, and ΔSV (changes in systolic, diastolic, and mean blood pressures, and stroke volume, respectively). These interactions probably indicate that the effects of FES and passive stepping on the respective binned differences became more pronounced over time (Fig. 1).

There was no significant interaction between FES and passive stepping (TABLE IV), implying that the combined effect of FES and passive stepping was not synergistic. This was contrary to our expectation that DFES would induce greater venous return than IFES alone by contracting the leg muscles in a more physiologically correct manner. Although passive stepping and FES independently mitigated a decrease in stroke volume (Fig. 1), they did not combine synergistically or even additively as DFES. The lack of synergy may have been caused by a ceiling effect.

TABLE IV
INTERACTIONS BETWEEN FES, PASSIVE STEPPING, AND TIME FOR CARDIOVASCULAR RESPONSES

Cardiovascular parameter	FES × Passive stepping		FES × Time		Passive stepping × Time		FES × Passive stepping × Time	
	$F_{(1,9)}$	p	$F_{(9,81)}$	p	$F_{(9,81)}$	p	$F_{(9,81)}$	p
ΔHR	2.34	0.161	1.63	0.120	0.601	0.792	0.967	0.474
ΔSBP	0.542	0.480	3.866	0.0004*	5.39	< 0.001*	1.65	0.115
ΔDBP	0.092	0.769	0.496	0.873	3.23	0.0022*	1.89	0.0644
ΔMBP	< 0.001	0.987	1.20	0.309	4.98	< 0.001*	1.73	0.0957
ΔSV	1.31	0.282	5.28	< 0.001*	2.59	0.0112*	1.27	0.265
ΔSVR	0.016	0.901	1.24	0.281	0.735	0.676	0.878	0.548

The asterisk indicates a statistically significant result. ΔHR, ΔSBP, ΔDBP, ΔMBP, ΔSV, and ΔSVR are changes in heart rate, systolic blood pressure, diastolic blood pressure, mean blood pressure, stroke volume, and systemic vascular resistance, respectively.

We found differences in cardiovascular responses between the conditions. Without any intervention (i.e., during HUT), heart rate significantly increased from the baseline and stroke volume significantly decreased from the baseline (Fig. 1). The increase in heart rate was probably due to reduced parasympathetic stimulation of the heart, and the decrease in

TABLE V
RESULTS OF TWO-WAY ANOVA AND TUKEY'S HSD POST HOC TESTS WITH CARDIOVASCULAR PARAMETERS

Cardiovascular parameter	$F_{(3,360)}$	p	Comparative relation
ΔHR	22.0	< 0.001*	H > (S, D, I) (S, D) > I
ΔSBP	42.9	< 0.001*	D > (I, S) > H
ΔDBP	13.2	< 0.001*	D > (S, I, H) S > H
ΔMBP	29.9	< 0.001*	D > (I, S) > H
ΔSV	33.9	< 0.001*	(D, I) > S > H
ΔSVR	1.53	0.208	N/A

The F and p are the results of the two-way ANOVA. The asterisk indicates a statistically significant result. 'Comparative relation' shows the results from the post hoc tests. Inequality signs indicate a significant difference between conditions. The letters, 'H', 'S', 'I', and 'D', represent HUT, STEP, IFES, and DFES, respectively. Parentheses indicate no significant difference between conditions.

TABLE VI
MEAN CHANGES IN CARDIOVASCULAR PARAMETERS DURING EXPERIMENTAL CONDITIONS †

Cardiovascular parameter	HUT		STEP		IFES		DFES	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
ΔHR [BPM]	18.9	3.57	12.2	1.73	2.88	2.59	8.40	1.54
ΔSBP [mmHg]	-10.4	2.59	1.78	1.55	6.69	3.33	14.6	5.43
ΔDBP [mmHg]	0.627	0.557	4.23	1.01	2.99	1.16	7.60	2.10
ΔMBP [mmHg]	-4.78	0.924	1.29	1.06	2.51	1.63	8.64	3.15
ΔSV [mL]	-24.1	4.91	-16.4	2.01	-6.77	3.09	-6.02	2.27
ΔSVR [dyn·s/cm^5]	-5.36	57.4	21.0	22.8	29.0	55.2	61.3	46.2

Positive values indicate an increase from the baseline. Standard deviations indicate the variation of the mean values during the conditions. ΔHR, ΔSBP, ΔDBP, ΔMBP, ΔSV, and ΔSVR are changes in heart rate, systolic blood pressure, diastolic blood pressure, mean blood pressure, stroke volume, and systemic vascular resistance, respectively. BPM is beats per minute. SD is

stroke volume was most likely due to gravitational pooling and the inadequacy of physiological responses to counteract it. Predominant responses would have comprised renin-angiotensin vasoconstrictor mechanism [4] and venoarteriolar reflex and myogenic response of the blood vessels [4,34,35]. Systemic vascular resistance increased by an average of 62.36 dyn·s/cm^5, but mean and systolic blood pressures decreased by an average of 4.781 mmHg and 10.44 mmHg, respectively. These results suggest that postural tachycardia and local vasoconstriction did not effectively maintain arterial pressure. Among the four conditions, HUT showed the largest increase in heart rate, the largest decrease in blood pressure, and the largest decrease in stroke volume (TABLE V). Because these unfavorable changes were most pronounced during HUT (i.e., without interventions), we can speculate that FES and passive stepping improved cardiovascular responses.

During STEP, heart rate significantly increased from the baseline and stroke volume significantly decreased from the baseline (Fig. 1). However, heart rate and stroke volume did not deviate from the baseline as much as they did during HUT (Fig. 1, TABLE V). Also, ΔSBP, ΔDBP, and ΔMBP (changes in systolic, diastolic, and mean blood pressures, respectively) were significantly higher during STEP than during HUT (TABLE V). These results suggest that passive stepping counteracted orthostatic stress to some extent and mitigated the decrease in arterial pressure by inducing greater venous return than HUT.

IFES and DFES maintained stroke volume better than HUT or STEP: during IFES and DFES, stroke volume did not significantly decrease from the baseline most of the time (Fig. 1), and ΔSV (change in stroke volume) was significantly greater during IFES and DFES than during HUT or STEP (TABLE V). These results suggest that IFES and DFES induced greater venous return than STEP. Despite the common effect on venous return, IFES and DFES affected blood pressure differently. IFES maintained blood pressure as effectively as STEP (Fig. 1, TABLE V), whereas DFES resulted in significantly higher ΔSBP, ΔDBP, and ΔMBP (changes in systolic, diastolic, and mean blood pressures, respectively) than the other conditions (TABLE V). Also, blood pressure was significantly higher than the baseline during most of DFES (Fig. 1). Among the determinants of blood pressure (i.e., heart rate, stroke volume, and systemic vascular resistance), only ΔHR (change in heart rate) was significantly greater during DFES than during IFES (TABLE V). This discrepancy is partially explained by the perception of exertion (i.e., the perceived intensity of exercise) [36]. Because DFES involved passive leg movements, the participants may have perceived greater exertion during DFES. This could have reduced the parasympathetic stimulation of the heart and contributed to greater ΔHR, which increased the blood pressure (Fig. 1). The observed differences between DFES and IFES for changes in blood pressure suggest that DFES may be a more effective intervention against orthostatic hypotension. However, further investigation is required to confirm the relative efficacy of DFES and IFES.

In our previous study [14], the combined intervention of FES and passive stepping (DFES) maintained venous return much more effectively than IFES alone. In the present study, IFES and DFES did not significantly differ in their ability to induce venous return: ΔSV (change in stroke volume) did not differ significantly (TABLE V). This difference between our results and the previous findings may be due to discrepancies in methods. Several discrepancies in methods suggest that IFES in [14] may have been less effective compared to IFES in the present study. First, in [14], the gastrocnemius and quadriceps femoris were stimulated simultaneously during one half of a cycle, and the hamstring was stimulated during the other half. This pattern may not have created the squeezing effect that we intended with the protocol in the present study. Second, not stimulating the tibialis anterior would have reduced the number of veins compressed during contractions in [14]. Third, [14] used lower stimulation intensities. Although the observed difference between the two studies can be attributed to the above methodological discrepancies, further studies are needed to properly explain the difference.

B. EMG Signals of Leg Muscles

Passive leg movements in an upright standing position can induce rhythmic EMG signals in individuals with SCI [37]. These signals can have comparable amplitudes to those of able-bodied individuals during locomotion [38,39], and the signals are associated with observable changes in the oxygenation of the paralyzed muscles [40]. In this study, we

examined if passive stepping during STEP induced similar muscle activities that could have increased venous return.

The frequency analysis indicated that EMG signals during STEP were insignificant. We speculate that the lack of hip extension, insufficient weight bearing, and the slower rate of stepping caused the discrepancy between our results and the previously reported rhythmic EMG signals [37]. Passive stepping that we applied did not include hip extension, whereas the study that reported rhythmic EMG signals applied approximately 14 degrees of hip extension [37]. Our participants supported approximately 7.5 to 24% of their body weight on each foot, varying within each cycle; in the study that measured rhythmic EMG signals, the participants supported almost their entire body weight on the soles [37]. Lastly, we applied stepping at 20 cycles per minute, whereas rhythmical EMG signals were recorded at 60 cycles per minute [37].

The measured EMG signals suggested that passive stepping did not cause significant muscle activation. Therefore, STEP probably mitigated the decrease in stroke volume by stretching the veins, whose one-way valves caused venous return when the diameter of the veins decreased due to stretching.

C. Cross-sectional Area of the Inferior Vena Cava (CSA_{IVC})

Although we used the same experimenter and technique as in the previous study [14], we could not measure CSA_{IVC} with sufficient accuracy due to poor image quality. Based on the body mass index, the majority of our participants were overweight or obese (which is common after SCI), whereas none of the participants in [14] was overweight. This may have been partially responsible for our inability to measure CSA_{IVC}. Though not an independent determinant, obesity is strongly associated with poor image quality in ultrasonography, primarily due to an inadequate acoustic window for the region of interest [41]. Also, if two abdominal walls have the same muscle thickness, the one with a greater fat composition reduces the mean grey level and contrast more [42]. Further consideration is required before measuring CSA_{IVC} in future studies with SCI individuals.

D. D. Limitations of the Study

The present pilot study demonstrates the feasibility of using FES to mitigate orthostatic hypotension in individuals with SCI. However, the heterogeneity of our participants implies that larger-scale studies are needed to prove that this use of FES applies to the general SCI population. Although heterogeneity is common in any given local population of individuals with SCI, future studies should better control the extent of preservation of autonomic function. Using only the AIS classification, level of injury, and post-injury durations may be inadequate for assessing the preservation of autonomic function: the framework for a more appropriate assessment is described in [19]. With properly stratified participants, we can conduct more rigorous studies that will better explain how interventions like IFES, DFES, and STEP differently affect the orthostatic tolerance of individuals with SCI. For now, we conclude that cyclic FES to the leg muscles can effectively induce venous return, thereby helping to maintain the blood pressure of individuals with SCI under orthostatic stress.

E. Research towards Clinical Implementation

As stated above, this pilot study suggests that FES might enable individuals with SCI to withstand orthostatic stress for a prolonged period. If FES is successfully implemented, individuals with SCI may be allowed to receive treatments that are otherwise precluded due to orthostatic stress. This would be beneficial especially in early rehabilitation because patients would not have to merely wait for their blood pressure regulation to recover, and earlier rehabilitation will prevent prolonged inactivity and subsequent secondary complications such as muscle atrophy, pressure ulcers, and demineralization of the lower limb bones. For example, there are many potential clinical benefits of weight bearing in a standing position for individuals with SCI. Such weight bearing can mitigate hypercalciuria [43,44] and a decrease in bone mineral density [45]. It may also help to prevent urinary tract infections by increasing the bladder pressure [46]. Individuals with SCI have reported that standing (either passively or with assistive devices) improved many aspects of their health, including the perception of well-being, circulation (e.g., reduced swelling in the lower extremities), reflex activity (e.g., reduced muscle spasms), and bowel and bladder function [47]. By applying the FES intervention that this study presented, we may be able to place people with SCI in a standing posture with a mitigated risk of orthostatic hypotension. The FES intervention can also mitigate this risk during reaching, grasping, and trunk balance exercises.

Despite the potential benefit, additional investigation is required before FES can be implemented in a clinical setting. Specifically, we need to investigate whether a combination of FES and passive stepping can be improved to provide better outcomes than FES alone. The findings of such investigation will help us identify the best intervention against orthostatic hypotension in individuals with SCI.

V. Conclusion

Orthostatic hypotension occurs if venous pooling due to a postural change is not adequately counteracted by reflexive sympathetic cardiovascular stimulation. The present study suggests that, during a 70-degree head-up tilt, individuals with SCI at T6 or higher can better maintain their blood pressure if FES is applied cyclically to their leg muscles. FES achieves this function by inducing venous return. Passive stepping can also induce venous return, but it is less effective than FES. Consequently, passive stepping is less effective in mitigating a decrease in arterial pressure during a 70-degree head-up tilt. Thus, FES appears to be more effective than passive stepping as an intervention against orthostatic hypotension in individuals with SCI. When passive stepping was combined with FES, they

did not interfere with each other, but they also did not interact synergistically.

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REFERENCES

[1] J. G. Bradley and K. A. Davis, "Orthostatic Hypotension," *American Family Physician*, vol. 68, no. 12, pp. 2393-2398, Dec. 2003.

[2] V. E. Claydon, J. D. Steeves, and A. Krassioukov, "Orthostatic hypotension following spinal cord injury: Understanding clinical pathophysiology," *Spinal Cord*, vol. 44, no. 6, pp. 341-351, Jun. 2006.

[3] A. A. J. Smit, J. R. Halliwill, P. A. Low, and W. Wieling, "Pathophysiological basis of orthostatic hypotension in autonomic failure," *Journal of Physiology*, vol. 519, no. 1, pp. 1-10, Aug. 1999.

[4] A. C. Guyton, *Textbook of Medical Physiology*, 11th ed. Philadelphia, Elsevier Saunders, 2006.

[5] A. Illman, K. Stiller, and M. Williams, "The prevalence of orthostatic hypotension during physiotherapy treatment in patients with an acute spinal cord injury," *Spinal Cord*, vol. 38, no. 12, pp. 741-747, Dec. 2000.

[6] R. Freeman, "Current pharmacologic treatment for orthostatic hypotension," *Clinical Autonomic Research*, vol. 18, no. SUPPL. 1, pp. 14-18, 2008.

[7] A. Krassioukov, J. J. Eng, D. E. Warburton, and R. Teasell, "A Systematic Review of the Management of Orthostatic Hypotension After Spinal Cord Injury," *Archives of Physical Medicine and Rehabilitation*, vol. 90, no. 5, pp. 876-885, May. 2009.

[8] D. J. Gillis, M. Wouda, and N. Hjeltnes, "Non-pharmacological management of orthostatic hypotension after spinal cord injury: A critical review of the literature," *Spinal Cord*, vol. 46, no. 10, pp. 652-659, 2008.

[9] A. S. Elokda, D. H. Nielsen, and R. K. Shields, "Effect of functional neuromuscular stimulation on postural related orthostatic stress in individuals with acute spinal cord injury," *Journal of Rehabilitation Research and Development*, vol. 37, no. 5, pp. 535-542, Sep./Oct. 2000.

[10] P. D. Faghri and J. Yount, "Electrically induced and voluntary activation of physiologic muscle pump: A comparison between spinal cord-injured and able-bodied individuals," *Clinical Rehabilitation*, vol. 16, no. 8, pp. 878-885, Aug. 2002.

[11] E. E. Sampson, R. S. Burnham, and B. J. Andrews, "Functional electrical stimulation effect on orthostatic hypotension after spinal cord injury," *Archives of Physical Medicine and Rehabilitation*, vol. 81, no. 2, pp. 139-143, Feb. 2000.

[12] C. Y. Chao and G. L. Cheing, "The effects of lower-extremity functional electric stimulation on the orthostatic responses of people with tetraplegia," *Archives of Physical Medicine and Rehabilitation*, vol. 86, no. 7, pp. 1427-1433, Jul. 2005.

[13] T. A. Thrasher, T. Keller, M. Lawrence, and M. R. Popovic, "Effects of isometric FES and dynamic FES on cardiovascular parameters on an active tilt-table stepper," *10th Annual Conference of the International FES Society*, Jul. 2005.

[14] L. Chi, K. Masani, M. Miyatani, T. Adam Thrasher, K. Wayne Johnston, A. Mardimae, C. Kessler, J. A. Fisher, and M. R. Popovic, "Cardiovascular response to functional electrical stimulation and dynamic tilt table therapy to improve orthostatic tolerance," *Journal of Electromyography and Kinesiology*, vol. 18, no. 6, pp. 900-907, Dec. 2008.

[15] S. Muraki, "Cardiovascular and respiratory responses to passive leg cycle exercise in people with spinal cord injuries," *European Journal of Applied Physiology and Occupational Physiology*, vol. 74, no. 1-2, pp. 23-28, Aug. 1996.

[16] S. Muraki, Y. Ehara, and M. Yamasaki, "Cardiovascular responses at the onset of passive leg cycle exercise in paraplegics with spinal cord injury," *European Journal of Applied Physiology*, vol. 81, no. 4, pp. 271-274, Mar. 2000.

[17] J. Raymond, G. M. Davis, and M. Van Der Plas, "Cardiovascular responses during submaximal electrical stimulation-induced leg cycling in individuals with paraplegia," *Clinical Physiology and Functional Imaging*, vol. 22, no. 2, pp. 92-98, Mar. 2002.

[18] S. P. Hooker, S. F. Figoni, R. M. Glaser, M. M. Rodgers, B. N. Ezenwa, and P. D. Faghri, "Physiologic responses to prolonged electrically stimulated leg-cycle exercise in the spinal cord injured," *Archives of Physical Medicine and Rehabilitation*, vol. 71, no. 11, pp. 863-869, Oct. 1990.

[19] M. S. Alexander, F. Biering-Sorensen, D. Bodner, N. L. Brackett, D. Cardenas, S. Charlifue, G. Creasey, V. Dietz, J. Ditunno, W. Donovan, S. L. Elliott, I. Estores, D. E. Graves, B. Green, A. Gousse, A. B. Jackson, M. Kennelly, A. -. Karlsson, A. Krassioukov, K. Krogh, T. Linsenmeyer, R. Marino, C. J. Mathias, I. Perkash, A. W. Sheel, G. Shilero, B. Schurch, J. Sonksen, S. Stiens, J. Wecht, L. A. Wuermser, and J. -. Wyndaele, "International standards to document remaining autonomic function after spinal cord injury," *Spinal Cord*, vol. 47, no. 1, pp. 36-43, Jan. 2009.

[20] J. L. Corbett, H. L. Frankel, and P. J. Harris, "Cardiovascular responses to tilting in tetraplegic man." *Journal of Physiology*, vol. 215, no. 2, pp. 411-431, Jun. 1971.

[21] K. Toska and L. Walløe, "Dynamic time course of hemodynamic responses after passive head-up tilt and tilt back to supine position," *Journal of Applied Physiology*, vol. 92, no. 4, pp. 1671-1676, Apr. 2002.

[22] P. Sundblad, Y. Haruna, B. Tedner, and D. Linnarsson, "Short-term cardiovascular responses to rapid whole-body tilting during exercise," *European Journal of Applied Physiology*, vol. 81, no. 4, pp. 259-270, Mar. 2000.

[23] D. Czell, R. Schreier, R. Rupp, S. Eberhard, G. Colombo, and V. Dietz, "Influence of passive leg movements on blood circulation on the tilt table in healthy adults," *Journal of NeuroEngineering and Rehabilitation*, vol. 1, Oct. 2004.

[24] G. Colombo, R. Schreier, A. Mayr, H. Plewa, and R. Rupp, "Novel tilt table with integrated robotic stepping mechanism: Design principles and clinical application," *Proceedings of the 2005 IEEE 9th International Conference on Rehabilitation Robotics*, vol. 2005, pp. 227-230, Jun.-Jul. 2005.

[25] P. C. Eser, N. D. N. Donaldson, H. Knecht, and E. Stüssi, "Influence of different stimulation frequencies on power output and fatigue during FES-cycling in recently injured SCI people," *IEEE Transactions on Neural Systems and Rehabilitation Engineering*, vol. 11, no. 3, pp. 236-240, Sep. 2003.

[26] D. Hawkins and M. L. Hull, "A method for determining lower extremity muscle-tendon lengths during flexion/extension movements," *Journal of Biomechanics*, vol. 23, no. 5, pp. 487-494, Jan. 1990.

[27] G. P. Molhoek, K. H. Wesseling, and J. J. M. Settels, "Evaluation of the Penaz servo-plethysmo-manometer for the continuous, non-invasive measurement of finger blood pressure," *Basic Research in Cardiology*, vol. 79, no. 5, pp. 598-609, Sep. 1984.

[28] D. W. Eeftinck Schattenkerk, J. J. Van Lieshout, A. H. Van Den Meiracker, K. R. Wesseling, S. Blanc, W. Wieling, G. A. Van Montfrans, J. J. Settels, K. H. Wesseling, and B. E. Westerhof, "Nexfin noninvasive continuous blood pressure validated against Riva-Rocci/Korotkoff," *American Journal of Hypertension*, vol. 22, no. 4, pp. 378-383, Apr. 2009.

[29] J. R. Martina, B. E. Westerhof, J. Van Goudoever, N. De Jonge, J. J. Van Lieshout, J. R. Lahpor, and B. A. J. M. De Mol, "Noninvasive blood pressure measurement by the nexfin monitor during reduced arterial pulsatility: A feasibility study," *ASAIO Journal*, vol. 56, no. 3, pp. 221-227, May/Jun. 2010.

[30] Y. Ishizaki, H. Fukuoka, T. Ishizaki, M. Kino, H. Higashino, N. Ueda, Y. Fujii, and Y. Kobayashi, "Measurement of inferior vena cava diameter for evaluation of venous return in subjects on day 10 of a bed-rest

experiment," *Journal of Applied Physiology*, vol. 96, no. 6, pp. 2179-2186, Jun. 2004.

[31] M. Lyon, M. Blaivas, and L. Brannam, "Sonographic measurement of the inferior vena cava as a marker of blood loss," *American Journal of Emergency Medicine*, vol. 23, no. 1, pp. 45-50, Jan. 2005.

[32] S. Lorsomradee, S. Lorsomradee, S. Cromheecke, P. W. ten Broecke, and S. G. De Hert, "Inferior Vena Cava Diameter and Central Venous Pressure Correlation During Cardiac Surgery," *Journal of Cardiothoracic and Vascular Anesthesia*, vol. 21, no. 4, pp. 492-496, Aug. 2007.

[33] M. E. Arthur, C. Landolfo, M. Wade, and M. R. Castresana, "Inferior Vena Cava Diameter (IVCD) Measured with Transesophageal Echocardiography (TEE) can be used to derive the Central Venous Pressure (CVP) in anesthetized mechanically ventilated patients," *Echocardiography*, vol. 26, no. 2, pp. 140-149, Feb. 2009.

[34] M. Kooijman, M. De Hoog, G. A. Rongen, H. J. M. Van Kuppevelt, P. Smits, and M. T. E. Hopman, "Local vasoconstriction in spinal cord-injured and able-bodied individuals," *Journal of Applied Physiology*, vol. 103, no. 3, pp. 1070-1077, Sep. 2007.

[35] C. G. Crandall, M. Shibasaki, and T. C. Yen, "Evidence that the human cutaneous venoarteriolar response is not mediated by adrenergic mechanisms," *Journal of Physiology*, vol. 538, no. 2, pp. 599-605, Jan. 2002.

[36] J. W. Williamson, R. McColl, D. Mathews, J. H. Mitchell, P. B. Raven, and W. P. Morgan, "Brain activation by central command during actual and imagined handgrip under hypnosis," *Journal of Applied Physiology*, vol. 92, no. 3, pp. 1317-1324, Mar. 2002.

[37] N. Kawashima, D. Nozaki, M. O. Abe, M. Akai, and K. Nakazawa, "Alternate leg movement amplifies locomotor-like muscle activity in spinal cord injured persons," *Journal of Neurophysiology*, vol. 93, no. 2, pp. 777-785, Feb. 2005.

[38] J. F. Yang and D. A. Winter, "Surface EMG profiles during different walking cadences in humans," *Electroencephalography and Clinical Neurophysiology*, vol. 60, no. 6, pp. 485-491, Jun. 1985.

[39] D. A. Winter and H. J. Yack, "EMG profiles during normal human walking: Stride-to-stride and inter-subject variability," *Electroencephalography and Clinical Neurophysiology*, vol. 67, no. 5, pp. 402-411, Nov. 1987.

[40] N. Kawashima, K. Nakazawa, and M. Akai, "Muscle oxygenation of the paralyzed lower limb in spinal cord-injured persons," *Medicine and Science in Sports and Exercise*, vol. 37, no. 6, pp. 915-921, Jun. 2005.

[41] A. Shmulewitz, S. A. Teefey, and B. S. Robinson, "Factors affecting image quality and diagnostic efficacy in abdominal sonography: A prospective study of 140 patients," *Journal of Clinical Ultrasound*, vol. 21, no. 9, pp. 623-630, Nov. 1993.

[42] U. Haberkorn, G. Layer, V. Rudat, I. Zuna, A. Lorenz, and G. Van Kaick, "Ultrasound image properties influenced by abdominal wall thickness and composition," *Journal of Clinical Ultrasound*, vol. 21, no. 7, pp. 423-429, Sep. 1993.

[43] L. W. Freeman, "The metabolism of calcium in patients with spinal cord injuries," *Annals of Surgery*, vol. 129, no. 2, pp. 177-184, Feb. 1949.

[44] P. E. Kaplan, W. Roden, and E. Gilbert, "Reduction of hypercalciuria in tetraplegia after weight-bearing and strengthening exercises," *Paraplegia*, vol. 19, no. 5, pp. 289-293, Oct. 1981.

[45] V. Alekna, M. Tamulaitiene, T. Sinevicius, and A. Juocevicius, "Effect of weight-bearing activities on bone mineral density in spinal cord injured patients during the period of the first two years," *Spinal Cord*, vol. 46, no. 11, pp. 727-732, 2008.

[46] D. W. Gould, A. C. L. Hsieh, and L. F. Tinckler, "The effects of posture on bladder pressure," *The Journal of Physiology*, vol. 129, no. 3, pp. 448-453, Sep. 1955.

[47] J. J. Eng, S. M. Levins, A. F. Townson, D. Mah-Jones, J. Bremner, and G. Huston, "Use of prolonged standing for individuals with spinal cord injuries," *Physical Therapy*, vol. 81, no. 8, pp. 1392-1399, Aug. 2001.