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EFFECT OF GAIT IMAGERY TASKS ON LOWER LIMB MUSCLE ACTIVITY WITH RESPECT TO BODY POSTURE

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16 Summary.-(max 180 words)-

17 The objective of this study was to evaluate the effect of gait imagery tasks on lower limb muscle 18 activity with respect to body posture. The sitting and standing position and lower limb muscle activity 19 was evaluated in 27 healthy female students (24.4 ± 1.3 yrs, 167.2 ± 5.2 cm, 60.10 ± 6.4 kg). Surface 20 electromyography was assessed during rest and in three different experimental conditions using 21 mental imagery. These included; a rhythmic gait, rhythmic gait simultaneously with observation of a 22 model and rhythmic gait after performing rhythmic gait. The normalized rmsEMG values with respect 23 to corresponding rest position were compared using non-parametric statistics. Standing gait imagery 24 tasks had facilitatory effect on proximal lower limb muscle activity. However, EMG activity of distal 25 leg muscles decreased for all gait imagery tasks in the sitting position, when the proprioceptive 26 feedback was less appropriate. For subsequent gait motor imagery tasks the muscle activity decreased, 27 probably as result of habituation. In conclusion the effect of motor imagery on muscle activity appears 28 to depend on relative strength of facilitatory and inhibitory inputs.

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33 Motor imagery (MI) represents a pure cognitive process, which positively influences motor 34 performance in healthy subjects which has been shown for sport performance, e.g. gymnastics, ballet and tennis (Guillot, Di Rienzo, Macintyre, Moran, & Collet, 2012). In addition this has been shown 35 36 in patients following motor impairment and has been used in physical therapy during recovery of 37 function (Lotze & Halsband, 2006; Mizuguchi et al., 2012). Specifically walking skills in 38 neurological patients improved after motor imagery exercise (Dunsky, Dickstein, Marcovitz, Levy, 39 & Deutsch, 2008; Oostra, Oomen, Vanderstraeten, & Vingerhoets, 2015). Home-based motor 40 imagery gait training programs have been shown to improve gait parameters including; walking 41 speed, stride length, cadence, single and double support time in chronic post-stroke subjects (Dunsky, 42 Dickstein, Marcovitz, Levy, & Deutsch, 2008). Motor imagery training includes imagery of walking 43 tasks in combination with physical therapy has been suggested to be more effective for improving 44 gait velocity in sub-acute stroke patients then physical therapy alone (Oostra, Oomen, Vanderstraeten, 45 & Vingerhoets, 2015). In addition videotape-based locomotor imagery training together with regular 46 physical therapy has been shown to improve walking ability in post-stroke and people with 47 Parkinson's disease more than gait training alone (El-Wishy & Fayez, 2013; Hwang et al., 2010).

48 Motor imagery can be described as a conscious mental simulation of an action without actual 49 execution, is accompanied by activity in specific neural substrates (both supraspinal and spinal) 50 similar to those involved in the actual executed movement. Meta-analysis on effect of motor imagery 51 on brain structures conducted by Hetu, et al. (2013) provided evidence that motor imagery activates 52 motor related brain networks including large fronto-parietal and subcortical regions involved in motor execution. Several studies provided evidence that motor imagery increases excitability in 53 54 corticospinal tracts which projects directly to motoneurons and their interneurons controlling the 55 muscles (Clark, Mahato, Nakazawa, Law, & Thomas, 2014; Cowley, Clark, & Ploutz-Snyder, 2008; 56 Oku, Ishida, Okada, & Hiraoka, 2011; Roosink & Zijdewind, 2010). This has been shown to increase 57 the excitability of spinal reflexes (Li, Kamper, Stevens, & Rymer, 2004) and also in muscle 58 proprioceptive structures (muscle spindle Ia afferent fibers) (Bonnet, Decety, Jeannerod, & Requin, 59 1997). So it seems that the motoneuron pool of muscle involved in imaginary movement receives 60 summation of neural inputs via descending and ascending neural pathways in similar way as during 61 real movement. The possibility that mental imagery can have an effect on the muscles that create the 62 movement is supported by the positive influence of motor imagery training on muscle strength (Clark 63 et al., 2014; Yue & Cole, 1992). However the influence of motor imagery on electromyography 64 (EMG) measures is not clear yet. To date several studies have found no significant effect of motor imagery on electromyographic activity during imaginary pointing arm movement for upper limb 65 muscles (Demougeot & Papaxanthis, 2011; Gentili, Papaxanthis, & Pozzo, 2006) during imaginary 66 pointing arm movement for upper limb muscles including anterior deltoid, tricpes and biceps brachii, 67

68 pectoralis major. In addition, Ranganathan, Siemionow, Liu, Sahgal, & Yue (2004) found no increase 69 in activity of biceps brachii and finger abductor during imaginary isometric little finger abduction 70 and elbow flexion, and Lemos, Rodrigues, & Vargas (2014) who found no increase in activity of the 71 gastrocnemius lateralis during imaginary rising on tiptoes. However, Oku, Ishida, Okada, & Hiraoka 72 (2011) found increased EMG in extensor carpi radialis activity during imaginary wrist extension and 73 Guillot et al. (2007) and Dickstein, Gazit-Grunwald, Plax, Dunsky, & Marcovitz (2005) showed 74 increased EMG activity of nine upper limb muscles in agonists, synergists, fixators and antagonists 75 during imaginary lifting a weighted dumbbell and increased EMG activity of gastrocnemius medialis 76 and rectus femoris when performing imaginary rising on tiptoes respectively.

77 Surface electromyographic measurements reflect, to some extent, the effort of neural system 78 for movement execution as EMG signal is usually proportional to the level of motor unit activity 79 (Richards, 2008). The muscle activity is altered by variations in the balance between inhibitory and 80 facilitatory input which go in parallel to the motoneuron pool, the terminal part of spinal afferent or efferent sensory/motor pathways (Daroff et al., 2012). So it might accepted that even during MI the 81 82 magnitude of EMG activity reflects the summation of facilitory and inhibitory inputs. This 83 assumption is supported by recent findings, which had shown that the increase of EMG activity during 84 MI mirrors a number of facilitatory inputs including mental effort related to e.g. characteristics of 85 imagined object, the heavier was the object lifted in imagination the showed a greater EMG signal 86 during MI (Bakker, Boschker, & Chung, 1996) and tends to be more pronounced in complex 87 functional movements (Bakker et al., 1996; Guillot et al., 2012; Guillot et al., 2007). The EMG signal 88 during motor imagery is classified mostly as subliminal (Guillot et al., 2012; Guillot et al., 2007) or 89 background muscle activity (Oku, Ishida, Okada, & Hiraoka, 2011) which indicates that detectable 90 muscle activity during MI does not have comparable magnitude and phasic pattern to real movement 91 execution. As the amount of increase in EMG amplitude during motor imagery is positively correlated 92 with the amount of corticospinal excitability (Oku, Ishida, Okada, & Hiraoka, 2011) and with respect 93 to previous findings that corticospinal excitability and brain activity during motor imagery is 94 enhanced with the real sensory feedback generated by holding an object which is imaginary 95 manipulated (Mizuguchi et al., 2012) we speculate that EMG activity during gait imagery may be 96 influenced by character of sensory feedback with respect to sitting (non-default position for walking) 97 or standing (default position for walking) body position during imagination.

With respect to imaginary training protocols in sport or in rehabilitation it has been suggested that simultaneously observing somebody doing the task during motor imagery further positively influences neural activity and enhances motor learning processes (Nedelko, Hassa, Hamzei, Schoenfeld, & Dettmers, 2012; Roosink & Zijdewind, 2010; Wright, Williams, & Holmes, 2014). In similar way with respect to motor learning even previous practice of imaginary movement facilitates neural activity more than imagery before practice, improves imagination ability of this movement
(Wriessnegger, Steyrl, Koschutnig, & Muller-Putz, 2014) and combination of imagination with real
practice is more effective for motor recovery then movement imagination or execution alone.
Therefore the simultaneous observing of imaginary movement will have facilitatory effect on muscle
activity.

It has also been previously suggested that the effectivity of the motor imagery training depends on individual's imaging ability (Gregg, Hall, & Butler, 2010). Subjects with a good motor imagery ability show a greater performance improvement following motor imagery training than do subjects with a poor imagery ability (Mizuguchi, Yamagishi, Nakata, & Kanosue, 2015).

112 The aim of the present study was to analyze the effect of gait imagery tasks from the first 113 person perspective on both proximal and distal lower limb muscle activity. Based on the prior finding 114 that motor imagery activates neural structures in similar way as movement execution and that muscle 115 activity reflects the summation of neural inputs coming to motoneuron pool via afferent and efferent 116 pathways we hypothesized that: (1) imagination of gait (which is considered as complex functional 117 task) modulates lower limb muscle activity, (2) the magnitude of muscle activity reflects character of 118 peripheral sensory inflow during imagination with respect to body posture and (3) the magnitude of 119 muscle activity is further influenced with respect to additional cognitive and motor task.

Therefore this study aimed to evaluate the electromyographic activity of proximal and distal lower limb muscles, which participate synergically on gait execution, during gait imagery tasks compare to rest conditions. This would potentially further our understanding of influence of gait imagery task on motor system and the effect of imagining or observing gait activity of lower limb muscles. This in turn provides important information for gait imagery rehabilitation protocols and could increase our understanding of gait control mechanisms.

126

127 Method

128 Participants

Twenty seven healthy females participated in this study. Their mean $(\pm SD)$ age, height and 129 130 weight were 24.4 ± 1.3 yrs, 167.2 ± 5.2 cm and 60.10 ± 6.4 kg. All participants were recruited from students from a Physiotherapy department of Palacky University. All participants had good cognitive 131 132 function and communicative skills necessary for motor imagery and were able to generate gait motor 133 imagery. Only participants with at least moderate visual and kinesthetic imagery ability, evaluated by 134 Revised Movement Imagery Questionnaire (MIQ-R), were included in the study (Smith & Collins, 135 2004). MIO-R represents a reliable tool to assess motor imagery ability in healthy persons. MIO-R 136 consists of an eight-item self-report questionnaire using two 7-point scales to evaluate ability to form visual and kinaesthetic mental images (Hall & Martin, 1997). The exclusion criteria included 137

psychiatric, neurological or musculoskeletal disorders, balance or walking problems, the use of a walking aid, chronic pain, pregnancy, the use of medication affecting the level of vigilance and uncorrected visual impairments. The dominant lower limb was the right side in all participants, determined as preference for kicking a ball (Seeley, Umberger, & Shapiro, 2008). Testing occurred in a quiet room in one day. All participants signed an informed consent prior to participating in this study. The procedures, which were approved by the local ethics committee, were performed according to the ethical standards of the Declaration of Helsinki.

145 *Motor imagery ability measures*

146 When completing the MIQ-R, participants are asked to perform one of four movement tasks 147 and then rate the ease with which they form visual and kinaesthetic images of this movement (from 148 1 = "very hard to see/feel" to 7 = "very easy to see/feel"). In the study mean MIQ-R scores (SD) were 47.7 (5.9) for both subscales, 24.15 (2.94) for the visual subscale, and 23.15 (3.15) for the kinaesthetic 149 150 subscale. The MIO-R has demonstrated adequate internal consistency with Cronbach α coefficients 0.78 and 0.76 for visual and kinaesthetic subscales respectively. MIQ-R mean scores and consistency 151 152 were comparable to those observed in previous MI studies (Hall & Martin, 1997; Guillot, et al., 2012). 153 *Electromyography measures*

154 Muscle activity was measured using surface EMG using two self-adhesive electrodes (Ag-155 AgCl). The electrodes were placed in parallel to the muscle fibers in the midline over the muscle belly with an inter electrode distance of 2 cm. Prior to placing the EMG surface electrodes, the skin was 156 157 abraded and cleaned. EMG activity was recorded from biarticular lower limb muscles involved with gait execution by synergistic action (Chvatal & Ting, 2012). Three distal muscles of the dominant 158 159 lower limb: tibialis anterior (TA), gastrocnemius lateralis (GL), gastrocnemius medialis (GM), and 160 three proximal muscles of the dominant lower limb: biceps femoris (BF), semitendinosus (ST) and 161 rectus femoris (RF) were measured. The reference electrode was placed over the fibula head. EMG data were recorded at 1000 Hz using the wireless system TeleMyo 2400T G2 (Noraxon Co., USA) 162 163 with a system bandwidth was 20-1000 Hz. Real-time EMG signals were sent via telemetry at 1,000 164 Hz to an A-D converter (Noraxon Co., USA). The raw EMG signals were full wave rectified and the 165 root mean squared value of EMG (rmsEMG) signals was calculated using a time averaging period of 25 ms (Guillot, et al., 2007). The processing of the signal was performed by using the software 166 167 MyoResearch XP Master Edition 1.08.17 (Noraxon Co., USA). Raw EMG signal was visually 168 checked prior to processing and analysis to verify the absence of any artifacts.

169 Procedure

170 The test protocol was conducted with respect to previous findings such that the imagination 171 ability was enhanced when imagination was done from first person perspective, and is performed 172 with externally given auditive feedback (Guillot, et al., 2007; Heremans, et al., 2012; Koehler, et al.,

173 2012; Mizuguchi, *et al.*, 2012; Roosink & Zijdewind, 2010).

EMG data were initially collected in two default rest positions, sitting (non-default position for walking) and standing (default position for walking) without performing any voluntary activity or motor imagery, and then within six motor imagery experimental conditions in the following order:

177 1. gait imagery in the sitting position, gait imagery in the standing position,

- 2. gait imagery and simultaneous gait observation in the sitting position, gait imagery and
 simultaneous gait observation in the standing position,
- 180 3. gait imagery in the sitting position after gait execution, gait imagery in the standing position181 after gait execution.

182 Experimental conditions are illustratively demonstrated in Figure 1.

183 FIGURE 1

184 Figure 1 insert here

Default sitting or standing positions were standardized for all experimental conditions. In the sitting position, the participants were seated upright in a chair that leaned against the back and arm rest. In the standing position, the participants were standing upright with hands along their body. In both default positions, the feet were placed a pelvic width apart. In all experimental situations, the position of the feet was unchanged.

190 For every participant and for all tested conditions, the rhythm of gait was given to the 191 participants using a metronome set at 110 beats per minute, to replicate a normal gait cadence All 192 tested participants reported that they were able to imagine gait well at this step frequency. In the first 193 experimental imaginary gait conditions for sitting and standing, the participants were instructed to 194 imagine a rhythmic gait as vividly as possible, in the first person perspective, the instruction was "Imagine yourself walking on the pace of the metronome" without making any actual movements. In 195 196 second tested conditions, the participants observed the rhythmic gait of a second person in frontal 197 plane from posterior side on the projection screen (200 x 200 cm) placed 2 meters in front of them. 198 The participants were instructed to watch the gait and to simultaneously imagine a rhythmic gait as 199 if they were walking (the instruction was "Observe the woman on the screen walking at the pace of 200 the metronome and simultaneously imagine yourself walking at the same pace"). Next, real rhythmic 201 walking at the pace of the metronome in hospital corridor was performed by the participants for a few 202 minutes to enhance further rhythmic gait imagination ability (Wriessnegger, Stevrl, Koschutnig, & 203 Muller-Putz, 2014). Just after real rhythmic walking, third experimental conditions were performed, 204 the instruction within the gait imagery task after gait execution was the same task as in the first 205 experimental conditions "Imagine yourself walking on the pace of the metronome". Each gait imagery

task lasted for approximately 60 seconds. None of participants mentioned feelings of fatigue duringthe experimental session.

208

209 Data processing

210 The rmsEMG [%] was calculated for every experimental condition in sitting or standing position and 211 then normalized to the rmsEMG of default sitting or standing rest positions. For the rest sitting and 212 standing positions the average rmsEMG values of all tested the muscles were calculated over a 20 213 seconds interval. These values calculated during the rest condition without any motor imaginary were 214 considered as reference values. For all rhythmic gait imagery tasks the mean rmsEMG values were 215 calculated over six gait cycles for the dominant lower limb. The duration of evaluated EMG period 216 was 6.6 seconds which was calculated from the metronome frequency where one gait cycle was 1.1 217 seconds. This period was selected from the middle part of the measured data for every experimental 218 condition with respect to adaptation on the imagery task. The mean rmsEMG values during 219 experimental gait imagery tasks were expressed as a percentage of reference value. Gait imaginary 220 experimental tasks conducted in sitting position were normalized to the respective reference value 221 obtained in rest sitting position and gait imaginary tasks conducted in standing position were 222 normalized to the respective reference value obtained in rest standing position for every participant 223 and tested muscle.

224

225 Statistical analysis

226 Data were tested to determine if they were normally distributed using Kolmogorov-Smirnov 227 test. All data were found not to be normally distributed, (p < 0.05), therefore non-parametric tests were 228 used throughout the analysis. For the statistical analysis the non-parametric Wilcoxon signed-rank 229 test was performed with the alpha value was set at p<0.05. This allowed the comparison of the reference values for sitting and standing positions and normalized EMG data for experimental 230 231 conditions in sitting and standing positions respectively (hypothesis 1). And the comparison of 232 normalized EMG data with respect to the default sitting and standing positions (hypothesis 2) alpha 233 value was set at p<0.05. The differences between each of the gait imagery conditions in the sitting or 234 standing position (hypothesis 3) were explored with Friedman tests with post-hoc Wilcoxon tests. As 235 normalizaed data for three experimental imagery conditions were compared and the alpha value was 236 calculated using Bonferroni's adjustment as 0.05/3 and set at p<0.017). In addition the effect size for 237 non-parametric data (Fritz, Morris, & Richler, 2012) Z values were computed. All statistical analysis 238 were performed using Statistica 9.0.

239

240 Results

241 For all tested muscles in rest default sitting and standing position the EMG activity was almost 242 silent, the mean and standard deviation reference rest electromyography data $[\mu V]$ are presented in 243 **Table 1**. All experimental gait imagery conditions were normalized as a percentage of the rest values 244 separately for each posture, muscle and participant, descriptive statistics of these data are presented 245 in **Table 2**. First gait imagery task in standing position had facilitatory effect on proximal lower limb 246 muscle activity (Table 2, Table 3). However, EMG activity of distal leg muscles decreased for all gait 247 imagery tasks in the sitting position, when the proprioceptive feedback was less appropriate. 248 249 TABLE 1 250 Table 1 insert here 251 TABLE 2 252 Table 2 insert here 253 *Gait imagery tasks vs. rest (Hypothesis 1)* 254 255 Conditions using rhythmic gait imagery mostly indicated an inhibitory effect on lower limb 256 muscle activity compared to the rest default positions (Table 3). In the sitting position this was 257 apparent for GM and GL and for TA in all experimental conditions, for BF and ST during gait imagery 258 and simultaneous gait observation and gait imagery after gait execution. 259 In the standing position significant inhibition was only present in GL for second gait imagery condition and in TA for second and third gait imagery condition. In the standing position, the first 260 261 gait imagery task in the proximal tested muscles (BF, RF) resulted in an increased EMG activity. 262 TABLE 3 263 *Table 3 insert here* 264 265 Standing vs. sitting position (Hypothesis 2) When comparing of the normalized EMG data between experimental conditions and between 266 267 the sitting and standing positions, muscle activity was mostly higher in the standing position (**Table** 268 3). This support the hypothesis that standing facilitates muscle activity in comparison to sitting. The difference were significant for GL (p<0.01, ES>0.3) and BF (p<0.05, ES>0.3) in every experimental 269

condition, for GM and TA (p<0.05, ES>0.3) in the first (SI1 × TI1) and third gait imagery condition (SI3 × TI3), for ST and RF (p<0.05, ES>0.3) in first gait imagery (SI1 × TI1) and imagery during gait observation (SI2 × TI2) conditions.

273

274 Subsequent gait imagery tasks (Hypothesis 3)

275 When comparing experimental conditions, in sitting position the EMG activity was lower 276 during the rhythmic gait imagery after rhythmic gait execution in comparison to the second gait 277 imagery condition for GM (Z=2.83, p=0.005, ES=0.36), GL (Z=3.24, p=0.001, ES=0.038), and TA 278 (Z=3.73, p<0.001, r=0.49) and in comparison to the first gait imagery condition (SI1 \times SI3) for GM 279 (Z=2.64, p=0.01, ES=0.39), GL (Z=2.79, p<0.001, ES=0.44), and TA (Z=3.63, p<0.001, ES=0.51). 280 In the standing position, the muscle activity was lower in the third tested condition compared to the 281 first tested condition for RF (Z=3.05, p<0.001, ES=0.42). For other comparisons the values did not 282 differ significantly.

283

284 Discussion

Guillot (2007) showed that MI was accompanied by subliminal EMG activity of muscles 285 286 participating on imagined movement execution. However the increase of lower limb muscle activity 287 during rhythmic gait imagery was not major finding in our study. Lower limb muscles mostly 288 decreased EMG activity during the experimental tasks using gait imagery compared to the rest 289 conditions, where EMG activity of all muscles was almost silent (Table 1). This was significant 290 especially for distal leg muscles in the sitting position (Table 2 and Table 3). The muscle activity 291 increase during MI compared to rest conditions was previously demonstrated mostly for upper limb 292 tasks (Bakker, et al., 1996; Guillot, Di Rienzo, et al., 2012; Guillot, et al., 2007; Solodkin, et al., 293 2004) or for non-gait foot tasks (Bakker, et al., 2007; Bonnet, et al., 1997). To follow on from the 294 results of Bakker et al. (2008) it could be suggested that during gait imagery compared to imagery of 295 non-gait or postural foot task supraspinal control is suppressed to some extent. Bakker et al. (2008) 296 compared corticospinal excitability within motor imagery of simple foot task (dorsiflexion) and MI 297 of gait measured by motor evoked potentials from task-related muscle m. tibialis anterior in sitting position. They found that motor evoked potentials areas increased during motor imagery of simple 298 299 foot task, however corticospinal excitability within gait imagery increased just in selected group of 300 subjects (5 from 16) who had larger increased during imagined foot dorsiflexion, so compared to the 301 majority of participants this simple task did not show and increase in muscle activity during gait 302 imagery.

As supraspinal control might be suppressed during imagery of postural task we speculate that the less expressed effect of gait imagery on muscle activity could be influenced by neural gait control mechanisms. Rhythmic complex patterns of synergistic muscle activity required for locomotion are to great extent under control of neural autonomy of CPG, neural networks located in lumbosacral spine connected with supraspinal motor regions and with lower limb afferent peripheral sensors (Solopova et al, 2015, Dietz, 2003, 2010; Chvatal & Ting, 2012; Dietz, 2003; MacKay-Lyons, 2002).

309 Motor imagery of lower-limb movements including gait relies mainly on the supplementary motor 310 area, cerebellum, putamen, and parietal regions (Hetu et al., 2013). Activity of these areas is required 311 more for gait planning with respect to changes of external environment rather than for stereotype 312 locomotion which has been shown to be more automatic (Hetu, et al., 2013; la Fougere, et al., 2010). 313 Activity of CPG might be modulated to a great extent by afferent sensory feedback from lower limb 314 receptors even with suppressed supraspinal control than has been previously demonstrated on spinal-315 cord-injured humans (Bussel et al, 1996, Dietz, 2003, 2010; Harkema, et al., 1997;) or in situations 316 without any extra demands on gait with respect to e.g. additional task or changes in the external 317 environment (Bussel, et al., 1996; Calancie, et al., 1994). Particularly phasic peripheral sensory 318 information associated with lower limb loading during walking evokes lower limb muscle activity 319 (Harkema et al., 1997). Harkema et al. (1997) found that by 70% unloaded body weight stepping (but 320 not 100% unloaded body weight stepping) movements induced by a driven gait orthosis on a treadmill 321 in healthy subjects elicited muscle activity of distal extensor lower limb muscles, namely 322 gastrocnemius medialis and soleus. So the EMG activity of distal lower limb muscles during the gait 323 is to a great extent dependent on phasic peripheral sensory information especially in situations when 324 no extra attention or demands on posture control are needed. The importance of proprioceptive 325 feedback for muscle activity during walking was suggested further McCrea (2001), who found that 326 feedback from extensor proprioceptors induces locomotor dependent reflexes that contribute 327 considerably to extensor muscle activity during real walking. So it is probable that especially distal 328 lower limb motor neurons don't receive enough facilitatory inputs to evoke muscle activity during 329 stereotype rhythmic gait imagery tasks in sitting position. Furthermore it seems that during the 330 imagining of gait in a position in which walking is impossible dominate inhibitory effect over possible 331 facilitatory on the muscle activity.

The emerging question from these current findings is not only why tested gait imagery conditions do not have facilitatory effect on muscle activity, which was the major focus in previous studies, but why gait imagery tasks resulted in decreased muscle activity compared to the rest condition in our experiment.

336 To date a decrease of EMG activity during imagination of movement execution task has not 337 been described. Decreased excitability of motor neural system during movement imagery compared 338 to rest condition, specifically decreased activity of corticospinal tract, has been previously reported 339 for imagination of muscle relaxation (Kato, Watanabe, Muraoka, & Kanosue, 2015) or during 340 imagination of suppressing movements (Sohn, Dang, & Hallett, 2003) for upper limb tasks. Few 341 studies found decreased corticospinal excitability during imagination of postural tasks in comparison 342 to rest conditions (Hiraoka, 2002; Oishi et al., 1994). Hiraoka (2002) suggested that imagination of stumbling in standing posture lead to decrease excitability of soleus H-reflex and Oishi (1994) found 343

that imaginary of skating motion in elite skate sprinters led to suppression of soleus H-reflex during
whole period of imaginary movement. All these finding are support the previous suggestion that
motor commands during motor imagery must be inhibited throughout the neural system to some
extent to prevent overt movement execution (Guillot, 2007; Jeanarod, 2001) as EMG activity (if
present) is just at subliminal intensity without tonic specific activity as during real movement (Guillot,
2007; Guillot, 2012; Jeanarod, 2001).

350 Inhibitory processes, which presumably propagate to the spinal motoneurons in parallel with 351 the excitatory inputs might have origin on the cortical, brainstem or either on spinal level (Jeannerod, 352 2006; Prut & Fetz, 1999). We speculate that the cause of EMG decrease, which occurred mostly in 353 sitting position during gait imagery tasks, presumably mostly took place on spinal level as sitting and 354 standing differs mostly by means of different proprioceptive input. It is probable that muscle spindle 355 afferents is gating the strength of Ia afferent synaptic input onto target motor neurons during gait 356 imagery in the same way as during gait execution (MacKay-Lyons, 2002). One of proposed 357 mechanisms of muscle activity inhibition is presynaptic inhibition according to a previous finding 358 that soleus H-reflex excitability as function of EMG level is decreased during gait (Stein & Capaday, 359 1998). Presynaptic inhibition reduces the amount of neurotransmitter released at the presynaptic terminal of the Ia axon which lead to decrease in EMG activity (Brooke et al., 1997; Bonnet et al. 360 361 1997). Furthermore we speculate that muscle activity decrease during gait imagery task might be 362 influenced by depression of afferent neuronal discharge as has been demonstrated during fictive 363 locomotion in the cat induced by mesencephalic locomotor region stimulation (Perreault et al., 1999). 364 Decrease of muscle and cutaneous afferent-evoked monosynaptic field potentials reflected a 365 reduction of depolarizing synaptic current into spinal neurons during fictive locomotion (Perreault et 366 al., 1999).

367

368 *The influence of posture*

369 For all tested muscles in most of experimental conditions was muscle activity during gait imaginary 370 tasks significantly lower in sitting position compared to muscle activity during gait imaginary tasks 371 in standing position (see Table 2, 3). Thus, the standing position compared to sitting position had an 372 excitatory effect on muscle activity during rhythmic gait imagery tasks. Standing posture is congruent 373 with walking and thus offer more appropriate somatosensory (tactile, proprioceptive and visual) 374 feedback compared to incongruent positions with walking such as sitting or lying. Presence of real 375 somatosensory feedback facilitates activity of neural structures within motor imagery and motor 376 observation (Mizuguchi et al., 2012; Vargas et al., 2004). Mizuguchi et al. (2012) found that 377 imagination of squeezing the ball and holding the real ball at the same time enhanced the MEPs in 378 comparison to the same situation just without the ball. Vargas et al. (2004) observed that corticospinal

379 excitability increased in situation when hand posture was compatible with the imagined task 380 compared to incompatible hand posture with the imagined task. Saimpont et al., 2012) proved that 381 posture might influence even accuracy of imagined movement, in their experiment the time duration 382 of gait motor imagery in standing posture (body posture congruent with walking) was more 383 comparable with real gait than gait motor imagery in sitting posture. It has been also previously 384 shown, that standing posture compared to supine posture (the one most used throughout the studies 385 concerning effect of gait observation or gait imagery) has excitatory effect on neural structures 386 (Nakazawa et al., 2003; Shimba et al., 2010). Nakazawa et al. (2003) demonstrated that both stretch 387 reflex and MEP elicited in tibialis anterior were significantly greater in standing compare to supine 388 posture (background EMG was silent in both conditions). Shimba et al. (2010) found that even passive 389 standing posture (accomplished by using gait orthosis) had higher impact on increased stretch reflex 390 of m. soleus compared to supine position. This might reflect facilitatory effect of standing position 391 on muscle spindle Ia afferent fibers. Facilitation of muscle spindle activity with respect to position 392 congruent with imaginary movement found also Bonnet et al. (1997). In their study they showed that 393 mental simulation of pressure on a pedal with the foot in reclined sitting position with their feet on 394 two pedals led to larger changes in T-reflex amplitude compared to H-reflex amplitude (activity of 395 muscle spindle Ia afferent fibres is elicited within the T-reflex, but not by H-reflex) in the leg involved 396 in the simulation. Even the extension of the hip in the standing position might have facilitatory effect 397 on muscle activity compared to sitting position, because also afferent input from hip joints is 398 important for the leg muscle activation during locomotion in dependence on hip position (Dietz and 399 Duysens, 2000; Dietz et al., 2002; Grillner & Rossignol, 1978). Grillner & Rossignol (1978) 400 previously proved that preventing the hip from extension in chronic spinal cats inhibits the flexors 401 muscle activity. As EMG activity depends on level of motoneuron pool excitation it is probable that 402 muscle proprioceptive (muscle spindle) afferents is gating the strength of Ia afferent synaptic insput 403 onto target motoneurons during gait imagery, same as during gait execution (MacKay-Lyons, 2002). 404 Then the level of proprioreceptors activation might be crucial for the the subtreshold activation of 405 target muscles during gait imagery tasks. This assumption is in accordance with previous studies the 406 appropriate propriceptive feedback (concretly posture congruent with imaginary task) provided 407 excitatory input to the motor system and facilitates muscle activity.

For the proximal tested muscles (BF and RF) the gait imagery task in the standing position was the only experimental condition when the muscle activity increased compared to the rest position. It has been previously suggested that the proximal leg muscles (e.g., BF) are mostly controlled by the monosynaptic corticospinal pathways compared to mostly polysynaptic corticospinal innervations of the distal leg muscles (e.g., GM) (Brouwer & Ashby, 1991; Cowan, Day, Marsden, & Rothwell, 1986). So presumably during the gait imagery task, the direct neural input from the cortex to the 414 motoneuron may enhance the ability of the cortex to control the proximal leg muscles (Brouwer & 415 Ashby, 1991). This assumption is in accordance with previous findings that during hand movements 416 dominates monosynaptic cortical-motoneuronal input (Nicolas et al., 2001) and mostly for upper limb 417 movements the presence of EMG activity during imagery tasks has been already demonstrated. It is 418 possible that motor imagery does not provide equivalent neural input to proximal and distal leg 419 muscles, but this has to be further explored. And still just biarticular lower limb muscles were 420 measured. To follow our results it is likely that the imagining of rhythmic gait provides inhibitory 421 input mostly to the distal leg muscles in the default sitting position. In accordance to previously 422 mentioned studies inhibition might reflect the summation of several factors including: decreased 423 supraspinal effort for stereotype gait imagery tasks, spinal inhibitory mechanisms (presynaptic 424 inhibition), different neural drive to the motoneurons of distal and proximal leg muscles, and default 425 sitting posture which does not provide appropriate feedback for real walking. However the results of 426 this study are limited to young woman population with good imagery ability, and to stereotype 427 rhythmic gait imagery task. Therefore, further research is required with respect to different genders 428 and populations.

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30 *Comparison of EMG activity during experimental conditions*

431 Combination of motor imagery and observation (Wright, Williams, & Holmes, 2014) or previous imagined movement execution (Wriessnegger, Steyrl, Koschutnig, & Muller-Putz, 2014) 432 433 enhances activity of neural structures and motor learning processes (Gomes, et al., 2014; Nedelko, 434 Hassa, Hamzei, Schoenfeld, & Dettmers, 2012) compared to motor imagery itself. Based on this 435 assumption we hypothesized, that both simultaneous motor imagery with motor observation and 436 previous execution of imagined movement would have further facilitatory effect on muscle activity 437 compared to gait imagery alone. So we added these "augmented" imagery conditions in given order to the experimental protocol. However in our experiment the second and the third experimental 438 439 condition mostly led to muscle activity decrease compared to the first tested situation. As the order 440 of first, second and third experimental conditions were not randomized we suggest that the decrease 441 in muscle activity within repeated tested motor imagery tasks in our experiment might reflect to some extent the gradual habituation effect. It has been previously described, that cortical activity is mostly 442 443 pronounced during initial trials of complex motor imagery tasks (imagery of volleyball spike attack) 444 compared to second and third motor imagery where the short-term habituation effect might be present 445 (Stecklow et al., 2010). None of tested participants reported feelings of tiredness during the 446 experiment the mental fatigue, which has been previously reported for prolonged imagery tasks 447 (Rozand et al., 2016), was not the reason of decreased muscle activity for subsequent imagery tasks.

- 448 We suggest here that more challenging imagery tasks as part of gait rehabilitation are required, then
- habituation effect might be avoided (Marchal-Crespo et al., 2014).
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The results of this study potentially further our understanding of influence of rhythmic gait imagination on lower limb muscles with respect to the body posture. This in turn provides important information for gait imagery rehabilitation protocols and could increase our understanding of gait

454 control mechanisms.

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763 FIGURE 1

764 Illustration of tested experimental conditions
 Gait imagery conditions
 Gait imagery and observation conditions
 Image: Condition of tested experimental conditions
 Image: Condition of tes

766

767 TABLE 1

768 Mean EMG $[\mu V]$ reference values (±SD) for all tested muscles in default sitting and standing positions

	Gastrocnemius medialis		Gastrocnemius lateralis		Tibialis anterior		Biceps femoris		Semitendinosus		Rectus femoris	
	М	SD	М	SD	М	SD	М	SD	М	SD	М	SD
Sitting position	1.35	0.53	1.48	0.52	1.59	0.53	1.3	0.39	1.2	0.4	1.21	0.48
Standing position	6.17	3.72	3.65	1.79	2.45	0.96	2.6	2.57	2.82	3.6	1.72	1.4

769

771 TABLE 2

- Normalized elekctromyographic activity with respect to reference value for every muscle [%] during
- 773 gait imagery tasks in sitting and standing position

	Gait imagery			Gait imag	gery and observation	Gait imagery after gain execution				
		Med	IQR(Q1-Q3)	Med	IQR (Q1 - Q3)	Med	IQR (Q1 - Q3)			
Gastrocnemius	S	73.15	(58.31–97.48)	69.07	(54.05-92.82)	61.62	(45.73-84.55)			
lateralis	Т	95.33	(85.23-127.63)	87.31	(70.27–95.68)	89.85	(81.15-106.95)			
Gastrocnemius	S	80.64	(54.15-97.92)	79.13	(51.56–98.24)	60.22	(45.91–91.24)			
medialis	Т	97.19	(78.13–129.47)	84.53	(70.58–109.11)	91.09	(75.83–122.77)			
Tibialis anterior	S	75.24	(64.25–112.14)	77.7	(62.84–95)	59.53	(50.49-86.9)			
	Т	96.58	(75.73–119.36)	88.13	(82.11–99.03)	85.34	(70.78–103.78)			
Biceps femoris	S	117.9	(91.09-221.63)	101.49	(86.37–151.14)	104.77	(82.31-129.04)			
	Т	93.5	(88.57–103.43)	91.48	(82.49–102.03)	85.86	(78.97–98.64)			
Semitendinosus	S	92.26	(78.35–108.78)	88.40	(76.73–102.7)	87.33	(76.62–107.02)			
	Т	111.28	(89.03-158.43)	99.1	(87.29–129.14)	98.15	(71.07–148.37)			
Rectus femoris	S	91.32	(86.17-106.95)	90.33	(82.06–100.34)	90.83	(75.08–104.5)			
	Т	111.11	(93.8-270.79)	98.3	(84.09–156.77)	97.24	(78.19–154.44)			

S – sitting position, T – standing position, Med – median, $(Q1 – Q3) – (25^{th} – 75^{th})$ percentile of data)

782 TABLE 3

- 783 Results of statistical analysis (Wilcoxon signed rank test and Effect Size) of changes in the muscle
- 784 activity during gait imagery tasks

		gait imagery tasks in the sitting position compare to default sitting rest position			gait imagery tasks in the standing position compare to default standing rest position			gait imagery tasks in the sitting position compare to gait imagery tasks in the standing position		
		Wilcoxon's	n	Effect Size	Wilcoxon´s Z	р	Effect Size	Wilcoxon´s	n	Effect Size
		Ζ	р					Z p	Ρ	
Gastrocnemius medialis	I1	3.00	<.001	0.41	0.29	0.77	0.04	2.21	0.03	0.3
	I2	3.15	<.001	0.43	1.78	0.08	0.24	1.42	0.16	0.19
	I3	4.08	<.001	0.56	0.29	0.77	0.04	3.99	<.001	0.54
Gastrocnemeius lateralis	I1	3.29	<.001	0.45	0.65	0.52	-0.08	4.30	<.001	0.58
lateralis	I2	4.04	<.001	0.55	2.71	0.01	0.34	2.79	0.01	0.38
	I3	4.42	<.001	0.6	1.15	0.25	0.16	4.18	<.001	0.59
Tibialis anterior	I1	2.16	0.03	0.29	0.36	0.72	0.05	2.38	0.02	0.32
	I2	2.26	0.02	0.31	2.07	0.04	0.28	1.39	0.16	0.19
	13	3.89	<.001	0.53	2.81	<.001	0.38	3.08	<.001	0.42
Biceps Femoris	I1	1.71	0.09	0.23	2.16	0.03	-0.29	2.59	0.01	0.35
	I2	3.05	<.001	0.42	1.13	0.26	-0.15	2.64	0.01	0.36
	13	3.10	<.001	0.42	0.77	0.44	-0.11	1.99	0.05	0.271
Semitendinosus	I1	1.42	0.16	0.19	1.75	0.08	0.24	2.50	0.01	0.34
	I2	3.17	<.001	0.43	0.53	0.60	-0.07	1.80	0.07	0.26
	I3	2.09	0.04	0.28	0,22	0,83	-0.03	1,13	0.26	0.15
Rectus femoris	I1	1.18	0.24	0.16	2.45	0.01	-0.33	3.39	<.001	0.46
	I2	1.95	0.05	0.27	0.86	0.39	-0.12	2.35	0.02	0.32
	I3	1.49	0.14	0.2	0.26	0.79	-0.04	0.96	0.34	0.13

I1 - gait imagery, I2 - gait imagrey and observation, I3 - gait imagery after gait execution