Adaptation and aftereffects of split-belt walking in cerebellar lesion patients

Wouter Hoogkamer,1 Sjoerd M. Bruijn,1,2,3 Stefan Sunaert,4,5 Stephan P. Swinnen,1 Frank Van Calenbergh,6 and Jacques Duysens1,7

1Movement Control and Neuroplasticity Research Group, Department of Kinesiology, KU Leuven, Leuven, Belgium; 2Department of Orthopedics, First Affiliated Hospital of Fujian Medical University, Fuzhou, Fujian, People’s Republic of China; 3MOVE Research Institute, VU University Amsterdam, Amsterdam, The Netherlands; 4Department of Radiology, University Hospitals Leuven, Leuven, Belgium; 5Department of Imaging and Pathology, KU Leuven, Leuven, Belgium; 6Department of Neurosurgery, University Hospitals Leuven, Leuven, Belgium; and 7Biomechatronics Lab, Mechatronics Department, Escola Politécnica, University of São Paulo, São Paulo, Brazil

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Hoogkamer W, Bruijn SM, Sunaert S, Swinnen SP, Van Calenbergh F, Duysens J. Adaptation and aftereffects of split-belt walking in cerebellar lesion patients. J Neurophysiol 114: 1693–1704, 2015. First published July 22, 2015; doi:10.1152/jn.00936.2014.—To walk efficiently and stably on different surfaces under various constrained conditions, humans need to adapt their gait pattern substantially. Although the mechanisms behind locomotor adaptation are still not fully understood, the cerebellum is thought to play an important role. In this study, we aimed to address the specific localization of cerebellar involvement in split-belt adaptation by comparing performance in patients with stable focal lesions after cerebellar tumor resection and in healthy controls. We observed that changes in symmetry of those parameters that were most closely related to interlimb coordination (such as step length and relative double stance time) were similar between healthy controls and cerebellar patients during and after split-belt walking. In contrast, relative stance times (proportions of stance in the gait cycle) were more asymmetric for the patient group than for the control group during the early phase of the post-split-belt condition. Patients who walked with more asymmetric relative stance times were more likely to demonstrate lesions in vermal lobules VI and Crus II. These results confirm that deficits in gait adaptation vary with ataxia severity and between patients with different types of cerebellar damage.

ataxia; cerebellum; gait; locomotion; step length symmetry
between the cerebellum and split-belt adaptation (Jayaram et al. 2011, 2012). However, although cerebellar involvement in locomotor adaptation has been observed repeatedly, it is still under debate where exactly this involvement is localized within the cerebellum (Ilg et al. 2008; Morton and Bastian 2006). Morton and Bastian (2006) could not directly address localization due to the diffuse nature of the cerebellar damage in their patient group but suggested that the midline vermis and fastigial nuclei would be most important in split-belt adaptation. Alternatively, Ilg et al. (2008) observed that damage in the intermediate cerebellum, the interposed nuclei, and adjacent dentate nuclei was related to impaired locomotor adaptation. It should be noted, however, that they used a different paradigm (adaptation to added mass at the legs). Hence the question remains whether these results can be extrapolated to split-belt walking.

One part of the cerebellum that has received special attention with respect to locomotion is the vermis, along with the concomitant fastigial nucleus. Mori and colleagues (Mori 1987; Mori et al. 1998, 2004) have underlined that locomotion needs integration of neuronal subsystems involved in posture and locomotion. The fastigial nucleus receives input from a variety of sensory sources and projects to the reticulospinal, vestibulospinal, and fastigiospinal pathways. Stimulation of these pathways (at the midline region of the hook bundle of Russell) evokes a general increase in postural muscle tone in cats standing on a stationary surface (Asanome et al. 1998) and induces locomotion when the cat is placed on the surface of a moving treadmill (Mori et al. 1999). In humans, with the use of functional magnetic resonance imaging, it was shown that the same networks were activated during mental imagery of standing and walking (Jahn et al. 2008). It is also important to point out that there are strong connections between the cerebellar and the mesencephalic locomotor regions (e.g., the pedunculopontine nucleus). In recent years it was found that this connectivity is deficient in patients with Parkinson’s disease, especially those with freezing of gait (Fling et al. 2013). This has implications for split-belt walking, since it was observed that patients with freezing of gait and patients in the off dopaminergic state adapt less and slower to split-belt walking (Mohammadi et al. 2015; Nanhoe-Mabahier et al. 2013; Roemmich et al. 2014).

The aim of the present study was to address the localization of the cerebellar involvement in split-belt adaptation by evaluating split-belt adaptation in patients with stable focal lesions following cerebellar tumor resection. On the basis of observations by Morton and Bastian (2006) and Ilg et al. (2008), we hypothesized that these patients would show several impairments during split-belt adaptation and that these impairments would be most pronounced in patients with lesions in the interposed nuclei. Specifically, with respect to interlimb coordination, we expected that patients would walk with a larger asymmetry in step lengths during the early phase of the split-belt condition and that this asymmetry would still be present during the late phase of the split-belt condition. Furthermore, we predicted that because of this reduced adaptation to split-belt walking, these patients would walk with more symmetric step lengths during the early phase of the postcondition. Finally, we hypothesized that patients with focal cerebellar lesions would show changes in intralimb gait parameters similar to those in healthy controls during the split-belt paradigm: no changes from the early to the late phase of the split-belt condition and no aftereffects during the postcondition.

**MATERIALS AND METHODS**

**Participants.** Fifteen patients with stable focal lesions after cerebellar tumor resection (age 23.0 ± 6.2 yr, mean ± SD; 10 women, 5 men; Table 1) and 13 healthy participants (age: 25.3 ± 4.6 yr; 9 women, 4 men) participated. All patients suffered from cerebellar tumors (pilocytic astrocytoma grade I, n = 6; astrocytoma grade II, n = 2; medulloblastoma, n = 5; Lhermitte-Duclos disease, n = 1; or hemangioblastoma, n = 1; Table 1). Seven patients received adjuvant radiotherapy; four of these patients received adjuvant chemotherapy (overview, Table 1; therapy details, Table 2). Extracerebellar damage, assessed on MRI images, was mainly limited to a residually enlarged supratentorial ventricular system or sequela due to a ventriculoperitoneal shunt in the right frontal lobe in some patients (Table 2). Patients were in a stable condition (>5 years postoperative, range 5 yr postoperative) for the present study.

**Table 1. Patients were mildly ataxic and in a stable condition (>5 yr postoperative)**

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age, yr</th>
<th>Time Postop, yr</th>
<th>Sex, M/F</th>
<th>Diagnosis</th>
<th>Interposed Nuclei</th>
<th>LeSION Volume, cm³</th>
<th>ICARS Total/100</th>
<th>P&amp;G/34</th>
<th>Kin Funct/52</th>
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<tbody>
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For patient 9 no MRI data were acquired. Postop, postoperative; F, female; M, male; RT, radiotherapy; CT, chemotherapy; Y, yes; ICARS, International Cooperative Ataxia Rating Scale; P&G, posture and gait subscore; Kin Funct, kinetic functions subscore.
ventriculocisternal shunt.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Diagnosis</th>
<th>Time Post-RT, yr</th>
<th>Target Areas Dose RT, Gy</th>
<th>Hypo-pituitarism</th>
<th>Time Post-CT, yr</th>
<th>Total Duration CT, mo</th>
<th>Scheme</th>
<th>VP Shunt</th>
<th>Extracerebellar sequela</th>
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Target areas: CSP, craniospinal; SP, spinal; FP, fossa posterior. Y, yes; VP, ventriculoperitoneal. CT schemes: HIT-2000, cisplatinum, vincristine, CCNU; HIT-91, ifosfamide, etoposide (VP16), metotrexate, ara-C, cisplatinum. Extracerebellar sequela: *Thalamic cavernous angioma, asymptomatic. †Hydrocephalus. ‡Cavernous angioma parietal white matter, asymptomatic; cavernous angioma intramedullary spinal cord, level D12, 1.8 × 2.6 mm, asymptomatic. §Ventriculocisternal shunt.

5.7–30.2 yr; Table 1) and were able to walk independently. All patients were able to walk on the treadmill without holding the hand railing. We rated severity of ataxia using the International Cooperative Ataxia Rating Scale (ICARS) (Trouillas et al. 1997). In this 100-point scale, a score of 0 indicates no deficits and increasing scores indicate more, or more severe, ataxic deficits. ICARS scores in our patient group ranged from 0 to 19, with only 3 patients scoring higher than 10 (Table 1). All participants gave written informed consent. The experiments were conducted in accordance with the Declaration of Helsinki and were approved by the local ethics committee.

**Experimental setup and protocol.** In general, procedures were similar to those of Buijn et al. (2012). On arrival of the participant in the lab, reflective markers were placed on the pelvis and lateral malleoli of the participants for movement registration with an optoelectronic system (Vicon Nexus; Oxford Metrics, Oxford, UK). Throughout all conditions, kinematics were sampled at 100 samples/s. In addition, during the walking trials on the treadmill, three-dimensional (3D) ground reaction forces and torques were sampled at 1,000 samples/s (instrumented dual-belt treadmill, custom built by ForceLink, Culemborg, The Netherlands).

Before the split-belt trials, we assessed comfortable overground walking speed. Participants walked a distance of 6 m at their natural pace (Abellan van Kan et al. 2009; Buijn et al. 2012). This was repeated three times, and all walking trials were performed barefoot. Next, participants were familiarized to treadmill walking. During the treadmill trials, participants wore a safety harness attached to the ceiling and were not holding the hand railing. The split-belt paradigm started with 3-min walking with both belts at 1.0 m/s (baseline). Participants then performed a classic split-belt paradigm, consisting of 10 min of walking with one belt at 1.0 m/s and the other belt at 0.5 m/s, followed by 5 min with both belts at the same speed (e.g., Buijn et al. 2012; Choi and Bastian 2007; Morton and Bastian 2006). During the split-belt (‘split’) condition, patients walked with the most affected side on the fast belt, and healthy controls walked with their nondominant leg on the fast leg (Morton and Bastian 2006). In the subsequent tied-belt (‘post’) condition, both belts ran at 1.0 m/s (Buijn et al. 2012), close to the participants’ preferred overground walking speed but different from the study by Morton and Bastian (2006), where aftereffects were assessed at 0.5 m/s. Between conditions the treadmill was stopped for a maximum of 30 s, and during the start of all treadmill conditions the belts had an acceleration of 0.3 m/s² (Buijn et al. 2012).

**Data analyses.** Generally, data analysis procedures were similar to those of Buijn et al. (2012). We calculated overground walking speed as the mean forward velocity of the two posterior pelvis markers during the three overground walking trials (Hoogkamer et al. 2015b). We determined the instants of heel strike and toe-off based on the center of pressure trajectory (Roerdink et al. 2008). This method was validated using 3D kinematics. Nevertheless, all trials were visually inspected and manually corrected if needed, also using the 3D kinematic data of the ankle marker. All gait parameters calculated for the leg that was on the fast belt during the split condition are referred to as “fast” parameters, even for the baseline and post conditions (Reisman et al. 2005), and similarly for the “slow” leg. Gait parameters were calculated for each stride. Step length was calculated as the anterior-posterior distance between the ankle markers at heel contact (Reisman et al. 2005), with step length breath at the heel contact of the fast leg. In other words, step length is the anterior-posterior distance between the ankle marker of the fast leg and the ankle marker of the slow leg at the time of heel contact of the fast leg. Our main objective was to evaluate adaptation and aftereffects in step length symmetry (Choi et al. 2009):

\[ \text{Step length symmetry} = \frac{(\text{step length}_{\text{fast}} - \text{step length}_{\text{slow}})}{(\text{step length}_{\text{fast}} + \text{step length}_{\text{slow}})}. \]

Similarly to Morton and Bastian (2006), we also evaluated changes in symmetry of double support timing, limb excursion, and stance time. Double stance symmetry was calculated similarly to step length symmetry but based on the relative duration of the double stance phase. The relative duration of the double stance phase occurring at the end of the stance phase of the fast leg was referred to as double stance (Reisman et al. 2005).

Limb excursion was calculated as the distance traveled by the ankle marker in the anterior-posterior direction from heel contact to toe-off of one limb (Hoogkamer et al. 2014). It needs to be emphasized that this parameter reflects displacements during the stance phase (and not a step). Limb excursion symmetry was calculated analogous to step length symmetry. Finally, stance time symmetry was also determined in the same way, using the relative stance time of each leg. In other
words, the proportion of stance in the gait cycle was compared for both sides.

Baseline values were calculated over all steps/strides of the baseline condition. For statistical analyses we calculated values over the early and late phases of the split and post conditions (summing up to 5 “episodes” in total: baseline, early split, late split, early post, and late post). Early split and early post values were calculated as the mean value over the first five strides of the respective conditions. Late split and late post values were calculated as the mean value over the last 50 strides of the respective conditions, to obtain a more accurate plateau value.

**MRI data acquisition and processing.** Image acquisition and processing procedures were similar to those in Hoogkamer et al. (2015c). A Philips 3T Achieva MRI scanner (Philips, Best, The Netherlands) with a 32-channel matrix head coil was used for image acquisition. A 3D MPRAGE (magnetization-prepared rapid acquisition gradient echo) high-resolution T1-weighted image (repetition time 970 ms, echo time 4.60 ms, flip angle 8°, 230 1-mm slices, in-plane resolution 0.97 × 0.98, 384 × 384 matrix) was acquired for all patients except P9 (see Table 1).

MRICron software (http://www.mccauslandcenter.sc.edu/mricro/mricron/index.html) was used to manually trace the lesions on the MPRAGE images. Lesion traces were spatially normalized to the atlas of the cerebellum (Diedrichsen et al. 2009, 2011) using the SUIT toolbox (http://www.icn.ucl.ac.uk/motorcontrol/imaging/suit.htm; Diedrichsen 2006) in SPM8 (http://www.fil.ion.ucl.ac.uk/spm/software/spm8/). In some cases (large lesions at the outer border of the cerebellum) spatial normalization with the SUIT toolbox was inaccurate. In those cases lesions were spatially normalized, based on the whole brain image, and the normalized lesions were manually corrected in atlas space when needed, based on the original image (Ilg et al. 2008). Lobules and nuclei with lesions were listed (Table 3), and patients with lesions in the interposed nuclei were identified (Table 1). For further analysis, all left-sided normalized lesions were flipped to the right along the midline (Ilg et al. 2013).

We hypothesized that patients with lesions in the interposed nuclei would show several impairments during split-belt adaptation: a reduced step length symmetry during early split and late split and a more symmetric step length during early post compared with the healthy controls. To test this, we performed statistics on these outcome measures for the patient subgroups with and without lesions in the interposed nuclei (lesion-based approach; see Statistical analyses of behavioral data). In addition, for voxel-based lesion-symptom mapping the patients were classified as “affected” or “unaffected” based on behavioral outcome measures, and then lesion locations between these subgroups were compared (symptom-based approach). This classification is commonly done with a cutoff threshold based on the behavioral data of the healthy controls, and this is not always straightforward (Hoogkamer and Meyns 2014). We applied a cutoff based on the 95% confidence interval of the values for the healthy controls. This was done for step length symmetry and in addition for stance time symmetry during early post. We used nonparametric mapping software within MRICron (http://www.mccauslandcenter.sc.edu/mricro/mricron/; Rorden et al. 2007) to perform the voxel-based lesion-symptom mapping analysis. We applied both statistical Liebermeister tests and subtraction analysis to identify lesion areas associated with deviant behavior (Christensen et al. 2014). For the Liebermeister test, significance threshold was set to $z = 1.65 (\alpha = 0.05)$, and only voxels damaged in at least two patients were considered (Ilg et al. 2013). Subtraction analysis was performed by subtracting the percentage of normally performing patients with a lesion in a specific voxel from the percentage of patients with deviant behavior with a lesion in that voxel (Christensen et al. 2014; Karnath et al. 2002). This was done for each lesioned voxel. We considered voxels that were at least 25% more likely to be lesioned in patients with deviant behavior (Christensen et al. 2014).

**Statistical analyses of behavioral data.** Student’s t-tests were used to compare overground walking speed and global gait parameters during baseline between healthy controls and the cerebellar lesion patient group. To evaluate changes over time and differences between groups in gait parameters during the split-belt paradigm, we performed two-factor repeated-measures ANOVAs with group and episode as factors. For each gait parameter we performed an ANOVA to compare early split and late split with baseline values. An additional ANOVA was performed to compare early post and late post with baseline values. Additionally, to compare gait parameters between the subgroups with and without lesions in the interposed nuclei, we performed a one-way ANOVA with three groups. For significant main and interaction effects we performed Tukey’s honestly significant difference post hoc analyses to identify significant differences between groups and/or episodes. Alongside significance, we report $t$ values of t-tests, $F$ values of ANOVAs, and $q$ values of Tukey’s post hoc tests, along with the corresponding degrees of freedom for each test. We used a traditional level of significance ($\alpha = 0.05$) for all statistical tests; when appropriate, this value was corrected for the number of analyses.
RESULTS

Self-selected overground walking speed was reduced in patients with cerebellar lesions compared with healthy controls [1.13 ± 0.13 vs. 1.35 ± 0.13 m/s, respectively; \( t(26) = 4.47; P < 0.001 \)]. During treadmill walking in the baseline condition (1.0 m/s, tied belts), gait parameters were similar between groups: stride time was 1.12 ± 0.06 vs. 1.12 ± 0.06 s \( [t(26) = 0.13; P = 0.90] \), stride time variability (SD) was 28 ± 8 vs. 28 ± 14 ms \( [t(26) = 0.01; P = 0.99] \), step length was 0.51 ± 0.03 vs. 0.51 ± 0.02 m \( [t(26) = 0.62; P = 0.54] \), and the relative stance time was 64.9 ± 0.7 vs. 65.0 ± 0.5% \( [t(26) = 0.58; P = 0.57] \) for the cerebellar lesion patients and the healthy controls, respectively. As such, these gait parameters confirmed that this group of patients with focal lesions in the cerebellum had only mild gait ataxia (see Table 1 for results of clinical evaluation).

Interlimb parameters. First, we evaluated the parameters that were expected to differ, namely, the interlimb gait parameters: step length symmetry and double stance symmetry. However, in both groups, these parameters increased similarly during the split-belt paradigm (Fig. 1). There was asymmetry during the early phase of the split condition, and values approached symmetry in the late phase of the split condition. The curve for step length symmetry suggests that the patients return to more symmetric values in late split; however, there was no significant main effect for group \( F(1,26) = 0.58; P = 0.45 \); Table 4 or group \( \times \) episode interaction effect \( F(1,26) = 0.21; P = 0.81 \). Double stance symmetry had not completely returned to baseline values during late split \( q(52) = 3.83; P = 0.024 \). During the post condition, both step length symmetry and double stance symmetry initially showed an overshoot in asymmetry and gradually returned to symmetric values.

Intralimb parameters. Second, we evaluated the parameters that were not expected to differ between groups, namely, the intralimb gait parameters: limb excursion symmetry and stance time symmetry. Both these parameters increased significantly during the split condition in both groups (Fig. 2; Table 4). Furthermore, during the post condition a larger overshoot in relative stance times was observed for the patient group.

Positive limb excursion symmetry values indicate that limb excursion of the fast leg was higher than that of the slow leg. Negative stance time symmetry values indicate that relative stance times of the fast leg were shorter than those of the slow leg. No significant main effects for group or group \( \times \) episode interaction effect were observed for limb excursion symmetry and stance time symmetry during split. During post, a significant asymmetric overshoot was observed during early post, returning to baseline values in late post, for both parameters. The asymmetry in relative stance times during early post was larger for the patient group (0.04 ± 0.03) than for the control group [0.00 ± 0.02; \( q(73) = 8.41; P = 0.001 \)].

Individual limbs. To further address the group difference in stance time symmetry, we evaluated the relative stance times of the individual limbs (Fig. 3A). In general, stance time symmetry changed simultaneously with the relative stance times of both the slow and the fast limb. However, in the overshoot in stance time symmetry during the early post phase, the changes in the relative stance time of the slow leg were more important than those of the fast leg.

During split, the between-limb difference in relative stance times became smaller, similarly in both groups. This occurred simultaneously with both an increase in the relative stance time of the fast leg \( q(52) = 3.54; P = 0.040 \) and a decrease in the relative stance time of the slow leg \( q(52) = 10.61; P < 0.001 \) from early to late split. During early post, the increased asymmetry in relative stance times was related to both an increased relative stance time of the fast leg compared with baseline \( q(52) = 9.21; P < 0.001 \) and a decreased relative stance time of the slow leg compared with baseline \( q(52) = 3.98; P = 0.019 \). During early post, the relative stance times were more asymmetric for the patient group than for the control group. When the fast and slow leg were evaluated separately during post, the group \( \times \) episode interaction effect was only significant for the slow leg \( F(2,52) = 4.41; P = 0.017 \), not for the fast leg \( F(2,52) = 1.66; P = 0.20 \). Post hoc analysis revealed that during early post, the relative stance time of the slow leg was lower in the patient group (62.7 ± 3.2%) than in the control group [65.3 ± 2.3%; \( q(63) = 7.73; P = 0.004 \)]. Furthermore, during early post, the relative

![Fig. 1. Interlimb gait parameters show similar changes for cerebellar lesion patients and healthy controls during split-belt walking. A: step length symmetry. B: double stance symmetry. Traces for cerebellar patients (gray) and healthy controls (black) during split (left) and post (middle) conditions. “Overview conditions” (right) summarizes values during the different phases and conditions. Shaded areas and error bars represent SE. Asterisks indicate values significantly different from baseline.](http://jn.physiology.org/)

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stance times of both legs were significantly correlated to the stance time symmetry (fast leg: $r = 0.67$; $P < 0.001$; Fig. 3B; slow leg: $r = -0.89$; $P < 0.001$; Fig. 3C). Multiple regression analysis revealed that the relative stance time of the slow leg explained a larger part (55%) of the variance in stance time symmetry than the relative stance time of the fast leg (21%).

**Stride times.** Along with the changes in the interlimb and intralimb gait parameters, changes in the stride time were observed (Fig. 3D; Table 4). In general, stride time changed similarly between groups, except during early post, when healthy controls walked with shorter stride times than the patient group and stride times were significantly correlated to stance time symmetry. Stride time increased from early split to late split and was reduced during early post compared with during baseline. Whereas stride time was similar between groups at baseline and during late post, during early post healthy controls walked with shorter stride times (0.98 ± 0.14 s) than the patient group [1.07 ± 0.09 s; $q(46) = 6.12; P = 0.041$]. To evaluate whether the group difference in stance time symmetry during early post was related to the group difference in stride time, we performed a regression analysis. This showed indeed a significant correlation between stride time and stance time symmetry ($r = 0.47; P = 0.011$; Fig. 3B).

**Lesion mapping of locomotor adaptation.** Eight patients had lesions in the interposed nuclei, six patients had no lesions in the interposed nuclei, and one patient was not included in the lesion-symptom mapping analyses because no MRI data were available (Table 1). In summary, we observed that lesion-based lesion-symptom mapping did not reveal any important group

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**Table 4. Summary of statistics**

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Values are means ± SE. BL, baseline; ES, early split; LS, late split. $P$ values in bold indicate significance.
differences. Alternatively, symptom-based lesion-symptom mapping could identify regions important for stance time symmetry, whereas lesion-symptom mapping based on step length symmetry was not feasible.

In the lesion-based approach, subgroups based on damage in the interposed nuclei were compared. During early split, no significant group effect on step length symmetry was observed \(F(2,24) = 0.05; P = 0.95\); Fig. 4A]. During late split, a significant group effect was observed \(F(2,24) = 3.50; P = 0.046\), Fig. 4B]. Post hoc intergroup comparisons suggest that the healthy controls had less symmetric step length than both patients with and patients without lesions in the interposed nuclei, but these differences were not significant \(q(24) = 5.38; P = 0.081\) and \(q(24) = 0.02; P = 0.118\), respectively]. Finally, during early post, no significant group effect was observed \(F(2,24) = 0.67; P = 0.52\), Fig. 4C].
To further explore potential relations between focal cerebellar damage and split-belt walking behavior, we applied voxel-based lesion-symptom mapping (see lesion overlap in Fig. 5A; Table 3). We evaluated both the step length symmetry during the episodes mentioned above and the stance time symmetry, which was observed to be different between groups during early post.

With regard to step length symmetry during split and post, patients were classified affected when symmetry values were below the 95% confidence interval of the values for the healthy controls (Fig. 4, A–C). Only one patient (P5; Table 1) could be classified to have a reduced step length symmetry during early split (Fig. 4A); during the other episodes, none of the patients displayed values lower than the cutoff threshold (Fig. 4, B and C). Therefore, voxel-based lesion-symptom mapping based on step length symmetry was not feasible. The disproportionately reduced step length symmetry during early split for P5 (Fig. 4A) was related to a negative step length during one of the first strides, where the foot of fast leg was placed posterior from the slow leg’s foot. This also occurred for the single healthy control whose step length symmetry was lower than the cutoff threshold (Fig. 4A).

Stance time symmetry during early post was higher in the patient group than in the control group (see above), and therefore patients were classified affected with values above the 95% confidence interval of the values for the healthy controls (Fig. 4D). Five patients had larger differences in relative stance times than the cutoff threshold (P4, P6, P8, P10, P12; Table 1). Subtraction analysis and the statistical Liebermeister test revealed importance for similar regions (Fig. 5, B and C). These were primarily in the posterior vermis; vermal lobules VI and Crus II, with, according to the subtraction analysis, extensions into vermal lobules VIIb and VIIIa. Interestingly, these affected patients walking with larger asymmetry in relative stance times during early post also showed increased values for step length symmetry during late split (0.01 ± 0.05) compared with the control group [−0.03 ± 0.03; t(16) = 2.65; P = 0.017]. Four of the 5 affected patients walked with positive step length symmetry values (range 0.010–0.089), overcompensating their asymmetric step lengths, as opposed to only 1 of the 13 controls (0.004).

**DISCUSSION**

The aim of the present study was to address the localization of the cerebellar involvement in split-belt adaptation. For this reason we included relatively mildly affected cerebellar patients having reasonably well-localized lesions. A major finding of the present study was that even mildly affected cerebellar patients show some changes in split-belt adaptation. Group differences were observed in stance time symmetry during early post: relative stance times were more asymmetric for the patient group than for the control group (Fig. 6). Specifically, affected cerebellar patients walked with shorter relative stance times during early post, placing the slow leg on the belt later than during baseline, whereas healthy controls placed their slow leg on the belt earlier than during baseline. Patients who walked with more asymmetric relative stance times were more likely to have lesions in vermal lobules VI and Crus II.

In general, the differences between patients and controls were small, in line with the mild degree of the deficits in the patient group. The present observation that patients with focal cerebellar lesions did not show any deficits in adaptation of step length symmetry may appear to be in contrast with
observations of such deficits in patients with diffuse cerebellar damage (Morton and Bastian 2006). However, an important, often overlooked element is that Morton and Bastian (2006) only included severely ataxic patients (ICARS/H11022 > 30) in their main study. In additional analyses, they showed that adaptation impairments were related to severity of (posture and gait) ataxia. Patients in our study had less severe ataxia, with ICARS scores ranging from 0 to 19 and only three patients scoring higher than 10. We did not observe a significant correlation between ICARS posture and gait (P&G) subscore and step length symmetry during early post, but the patient with the highest subscore (P&G/H11005/10; P15; Table 1) did show the smallest asymmetry in step length of all participants. Furthermore, the patient that showed the lowest step length symmetry during early split (P5; Fig. 4A) also had a rather high P&G subscore (P&G/H11005/6; Table 1) compared with the other patients.

The mild degree of ataxia may also explain some of the differences observed with other studies. For example, we

Fig. 5. Lesion overlap and overview of areas related to an increased asymmetry in relative stance times during the early phase of the post condition. A: superposition of the regions of cerebellar lesions of all patients. Note that lesions were flipped to the right for analysis. Maximum overlap (10 patients) was within vermal lobule VIIIa and paravermal lobules VIIb and Crus II (color coding according to the heat index above cerebellar slices). B: vermal lobules VI and Crus II were significantly correlated to an increased asymmetry in relative stance times during early post. Regions with z values >1.65 (P < 0.05), resulting from the Liebermeister test are indicated (color coding according to the heat index above cerebellar slices). C: subtraction analysis identified the same regions; regions that were at least 25% more likely to be lesioned in patients with increased asymmetry in relative stance times are indicated (color coding according heat index above cerebellar slices).

Fig. 6. Differences in relative stance phases during the early phase of the post condition between healthy controls and “affected” cerebellar patients. Relative stance phases for healthy controls (black) and affected cerebellar patients (gray) during baseline (open bars) and during the early phase of the post condition (solid bars). Affected cerebellar patients walked with shorter relative stance times of the slow leg during early post, placing the slow leg on the belt later than during baseline, whereas healthy controls placed their slow leg on the belt earlier than during baseline. Error bars represent SE.
observed no differences in split-belt adaptation between patients with and patients without lesions in the interposed nuclei. This observation may appear in contrast with the observation that the interposed nuclei are important in adaptation of limb coordination, when walking with added mass at the shanks (Ilg et al. 2008). Again, one could argue that such differences were related to differences between patients. However, interestingly, ataxia severity of the patients with focal lesions in the latter study was similar to that of our patients (Ilg et al. 2008). This could suggest that adaptation to split-belt walking and adaptation to added-mass walking are rather different processes. The latter is impaired in mildly ataxic patients with focal lesions, specifically in patients with lesions in the interposed nuclei. In contrast, adaptation of interlimb parameters in split-belt walking was not observed to be impaired in mildly ataxic patients or to be dependent on the interposed nuclei. Split-belt adaptation appears to be more related to control of posture and gait (Morton and Bastian 2006), whereas added-mass adaptation appears to be more related to control of multi-joint movements (Ilg et al. 2008).

**Intralimb parameters.** Whereas most recent split-belt studies have focused mainly on changes in interlimb parameters such as step length, we observed significant aftereffects in intralimb parameters, as well. These aftereffects were most prominent in the patient group (Fig. 2) and were closely related to the relative stance time of the slow leg specifically. During the split condition, the relative stance times of the slow leg were higher than those of the fast leg. During the split trial, the participants reduced the relative stance time of the slow leg from the early to the late phase. In the patients, a larger part of this reduction was still present in the early phase of the post condition (storage and transfer), because they walked with shorter relative stance times in the slow leg than the controls. Although the larger aftereffects in intralimb parameters are not necessarily a deficit in adaptation, their presence suggests that the patients used a slightly different strategy to adapt their gait pattern to split-belt walking (see companion article, Hoogkamer et al. 2015a), reminiscent of the faster adjustments in swing times in elderly (Bruijn et al. 2012). This was confirmed when we compared the symmetry values of different gait parameters between the group of affected patients and the control group during split. Specifically, we observed that step length symmetry during the late phase of the split-belt condition was higher in the affected patients than in the control group. Four of these five patients showed overcompensation: during split, they first walked with smaller steps with the fast leg than with the slow leg; during split-belt walking, this asymmetry was not only reduced but overcompensated, resulting in asymmetry of opposite sign (smaller steps with the slow leg).

Furthermore, these data revealed some lesion site dependencies. We identified five patients with larger differences in relative stance times than the healthy controls (Fig. 4D), and these patients were more likely to have lesions in the posterior vermis: vermal lobules VI and Crus II (Fig. 5, B and C). However, it should be noted that the identified regions were small and that accompanying z values ($z = 1.8$) and subtracted percentages were low ($<50\%$). Traditionally these regions have not been endowed with an important motor control function, but several observations from lesion and functional MRI studies suggest otherwise. From structural and functional connectivity studies, these regions appear to be mainly related to the limbic system (Stoodley and Schmahman 2010) and frontoparietal and dorsal attention networks (Buckner et al. 2011). In lesion studies, vermal lobule Crus II has been related to visuomotor adaptation, both in cerebellar stroke patients (chronic) and in patients with cerebellar degeneration (Donchin et al. 2012). In patients with acute and subacute stroke lesions, this relation was not observed (Burciu et al. 2014). Studies on force-field adaptation and added-mass walking did not observe a significant role for vermal lobules VI and Crus II (Burciu et al. 2014; Donchin et al. 2012; Ilg et al. 2008). It should be noted, however, that lesion overlap images from those studies suggest that very few patients had lesions in these regions. In animal studies, the vermis (and specifically the fastigial nucleus) has often been related to the control of posture and gait (for review, see Mori et al. 2004). In humans, the posterior vermis has been related to performance on tandem walking (Bastian et al. 1998), but all lesions in that patient sample included vermal lobule X, which can be expected to be more important in relation to balance (Stoodley and Schmahman 2010). In functional MRI studies, vermal lobule CII has been related to eye-hand coordination motor learning (Miall and Jenkinson 2005). Functional imaging studies from our group using bimanual coordination tasks have repeatedly observed involvement of cerebellar lobule VI, most often in the paravermal and hemispheric regions (Debaere et al. 2003; Heuninckx et al. 2005; Swinnen et al. 2010; Wenderoth et al. 2004, 2005) but also in vermal lobule VI (Beets et al. 2015; Debaere et al. 2004). Furthermore, both vermal lobules VI and Crus II have been observed to be enlarged in well-trained basketball players, based on MRI volumetric analyses (Park et al. 2009). In studies on cats, the importance of the vermis and associated nuclei (especially the fastigial nucleus) for posture and locomotion has been highlighted by Mori and colleagues (see Introduction). In addition, detailed studies are available on registration from the vermis and paravermis during cat locomotion, mostly from lobule V (Andersson and Armstrong 1987; Armstrong et al. 1982; Udo et al. 1981). A striking finding of one of these studies (on the lateral part of the vermis of lobule V) was that complex spikes were selectively generated when unexpected events occurred (such as during ladder stepping when a rung underwent an unexpected descent when stepped upon; Andersson and Armstrong 1987). This part of the vermis projects to the lateral vestibular nucleus.

**Limitations.** First, since our aim was to address the localization of the cerebellar involvement in split-belt adaptation, we included patients with focal cerebellar lesions. These patients are commonly only mildly ataxic, which turned out to be a limitation for our study, as argued above. Second, even though we aimed to include mainly patients who did not receive adjuvant radio- or chemotherapy, several patients displayed extracerebellar damage (Table 2). This is suboptimal for lesion symptom mapping analysis (Timmann et al. 2009), but it did not bias our results: subtraction analysis within the subset of patients who did not receive adjuvant therapies indicated importance for similar regions to those identified in our main analysis (vermal lobules VI and Crus II) and in addition for paravermal lobule IX. Another limitation of lesion analysis is that whereas it gives indications of how behavior changes when a specific region is dysfunctional, it does not prove that this region is involved under healthy conditions. Furthermore,
regions that are important for specific functions might not be identified, either because few, if any, of the patients have lesions in these regions or because other regions compensate for the deficits.

To further address the functional localization of locomotor adaptation, future studies could include patients with more severe forms of ataxia. Voxel-based morphometry could be used to localize deficits in cerebellar degeneration patients. Studies on severely ataxic patients with focal lesions (e.g., subacute or chronic stroke) could also provide useful insights. It should be mentioned, however, that this group is not easy to obtain because stroke lesions are seldom limited to the cerebellum. Furthermore, stroke patients are often at advanced age, which could confound behavioral outcomes (Bruijn et al. 2012). Finally, it should be mentioned that when studying severely ataxic patients, there is also the possibility that behavioral outcomes are confounded by other deficits (in balance or muscle coordination) or by compensation strategies to cope with those.

In summary, we observed that changes in symmetry of interlimb parameters, such as step length and relative double stance time during the split and post conditions, were similar between healthy controls and mildly ataxic patients with focal cerebellar lesions. Relative stance times were more asymmetric for the patient group than for the control group during the early phase of the post condition. Patients who walked with more asymmetric relative stance times were more likely to have lesions in vermal lobules VI and Crus II.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS


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