

Adaptation reveals independent control networks for human walking

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Human walking is remarkably adaptable on short and long timescales. We can immediately transition between directions and gait patterns, and we can adaptively learn accurate calibrations for different walking contexts. Here we studied the degree to which different motor patterns can adapt independently. We used a split-belt treadmill to adapt the right and left legs to different speeds and in different directions (forward versus backward). To our surprise, adults could easily walk with their legs moving in opposite directions. Analysis of aftereffects showed that walking adaptations are stored independently for each leg and do not transfer across directions. Thus, there are separate functional networks controlling forward and backward walking in humans, and the circuits controlling the right and left legs can be trained individually. Such training could provide a new therapeutic approach for correcting various walking asymmetries.

Humans must constantly recalibrate their walking pattern to navigate different terrains and environments. Our locomotor system can react quickly to unpredictable conditions, but can also learn to make predictive adjustments in response to persistent perturbations^{1–3}. Adaptive learning mechanisms are therefore critical for optimizing coordination patterns so that individuals maintain walking stability and efficiency. People with cerebellar damage can make reactive changes normally during walking, but are impaired when attempting to learn predictive changes⁴. Individuals with cerebral damage from stroke, on the other hand, have a normal capacity to make both reactive and predictive locomotor adaptations during walking⁵. Hence, predictive control in human locomotion appears to depend specifically on cerebellar mechanisms that presumably modulate the output of spinal motor circuits that form the basic rhythmic limb movements during walking.

Which components of locomotor control circuits are adaptable? Little is known about the organization of motor circuits that generate walking rhythms and patterns in humans. Central pattern generators (CPGs) for locomotion have been shown to exist in the spinal cord of many vertebrates^{6,7}. Similar circuits likely exist in humans^{8,9}, although supraspinal structures must be important as a result of the additional demands of bipedal walking. CPGs are thought to be shared circuits that can be reconfigured and participate in several behaviors. It is presumed that the extent of circuit sharing between different behaviors depends on the similarities in the patterns of muscle activation or the resultant limb motion. In the case of forward versus backward walking, the temporal sequence of muscle activations and the organization of muscle synergies differ markedly, yet the limb motions (that is, kinematics) are remarkably similar¹⁰. If the adapted circuit encodes

muscular patterns, then learning might not be shared across locomotor directions; if it encodes kinematics, then learning might be shared. Another recent proposal is that a two-level CPG exists with a single rhythm generator that sets the phasing of a second set of neurons that generate the muscular patterns^{11,12}. This suggests that even locomotor forms requiring different motor patterns could be coordinated by a common rhythm generating circuitry, and would therefore be subject to crossadaptation. Support for a two-level CPG includes studies of human infants who can continuously transition between stepping directions without interruption of the stepping rhythm¹³ and maintain one-to-one coupling even when one leg steps forward and the other backward¹⁴.

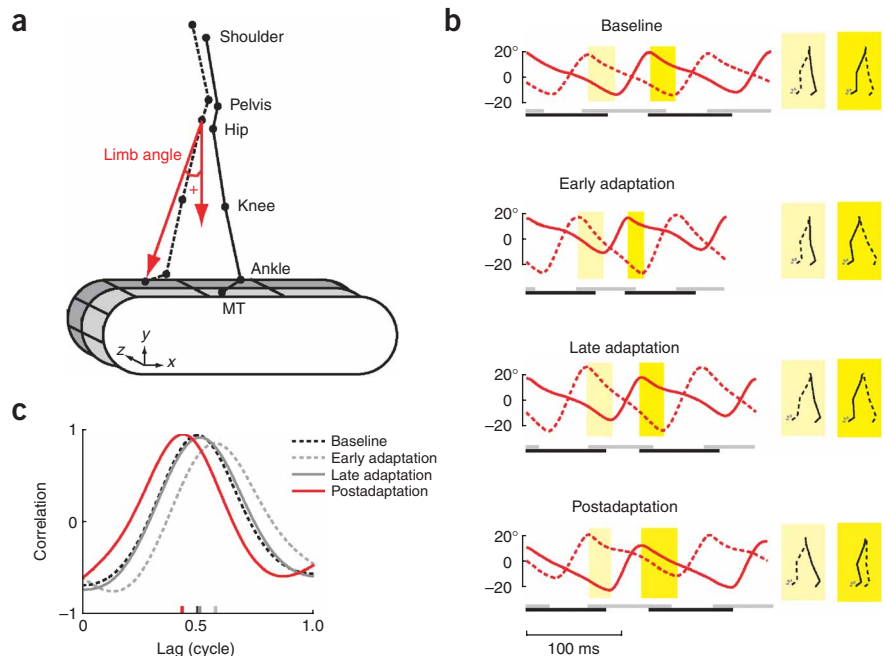
Here we investigated whether locomotor adaptation transfers between forward and backward walking. Adaptation was induced via a split-belt treadmill^{3,15}, where the legs are trained to walk at different speeds in the same or different directions. Human adults immediately react to the speed perturbations by scaling the stance and swing times of each leg independently to maintain an alternating walk pattern³. However, interlimb coordination is phase shifted and step lengths become asymmetric so that subjects appear to walk with a 'limp'. Adaptive mechanisms improve interlimb coordination (that is, phasing) over several minutes of training and reduce asymmetry; this eliminates the obvious limp and restores a symmetric gait pattern. We confirmed that the newly adapted motor pattern was stored by measuring the presence of an aftereffect when the legs returned to walking at the same speed. Our results demonstrate that humans can show aftereffects in both forward and backward walking. However, these effects do not transfer, nor do they interfere with each other (that is, no crossadaptation). Furthermore, by studying adaptation to hybrid

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Figure 1 Experimental setup and phase analysis. (a) Stick figure illustrates marker locations (dots) and limb angle. (b) Example limb trajectories during forward walking at baseline (tied belts), early and late adaptation (split belts), and early post-adaptation (tied belts). We will refer to the leg trained on fast belt during split-belts condition as the fast leg (dotted), and the leg trained on slow belt during split-belts conditions as the slow leg (solid). Horizontal bars below represent stance phases for the fast (gray) and slow leg (black). Light- and dark-yellow-shaded areas represent the periods from peak fast limb flexion to peak slow limb extension and the periods from peak slow limb flexion to peak fast limb extension, respectively. Stick figures on the right illustrate limb configurations corresponding to the beginning of each shaded time window (that is, a step with the fast or slow leg leading). Note the development of asymmetries (width of yellow boxes) and the step lengths (stick figures) during early adaptation. The asymmetries are reduced by late adaptation and reversed during postadaptation. (c) Examples of cross-correlation functions between limb trajectories, where interlimb phase is the lag time at peak correlation (tick marks on x axis). At baseline forward walking, the phase value was about 0.5 cycles, reflecting out-of-phase coordination. During split-belt adaptation, the fast leg was initially phase advanced (>0.5) relative to the slow leg and gradually phase shifted back to baseline (~ 0.5) by the end of the adaptation. In postadaptation tied-belts walking, the fast leg was phase lagged (<0.5) relative to baseline, indicating storage of aftereffects from split-belt training.



walking (that is, one leg stepping forward and one backward), we demonstrate that each leg shows independent aftereffects. Our findings indicate that plasticity associated with locomotor training is both leg and direction specific in human adults.

RESULTS

Subjects walked on a split-belt treadmill, with one belt moving twice as fast as the other during adaptation, and both belts moving at the same speed (tied-belts) during baseline and postadaptation. Split-belt training leads to locomotor adaptation that alters interlimb coordination³. We measured interlimb coordination by calculating the cross-correlation function between limb angle trajectories; interlimb phase was determined from the lag time at peak correlation (Fig. 1a–c). Motor adaptation was evident from the change in phasing from early to late split-belt adaptation, and the difference in phasing between baseline and postadaptation tied-belts conditions—that is, an aftereffect (Fig. 1c). In this study, we tested baseline walking in different directions. Split-belt training was done in one or more walking direction(s). After training, walking in different directions was tested again for aftereffects. Transfer to untrained walking directions would be evidenced by a phase change from baseline to postadaptation.

We hypothesized that if overlapping or shared neural circuits that control forward and backward walking were adapted, then training in forward walking should cause aftereffects in both forward and backward walking. Furthermore, washing out the adaptation in backward walking (that is, de-adaptation back to backward walking baseline) should simultaneously washout forward walking effects (Fig. 2a). However, if the neural circuits for forward and backward walking were functionally separated, then adaptation to forward walking should not transfer to backward walking, and backward walking should not washout forward walking adaptation (Fig. 2b). In experiment I, the legs moved at 0.5 cycles out of phase for both backward and forward

walking baselines on tied-belts. Subjects then walked forward on split-belts at a 2:1 speed ratio for 10 min. In a typical subject (Fig. 2c and **Supplementary Video 1** online), the fast leg was phase advanced relative to the slow leg for the initial steps in split-belts condition, and gradually phase shifted back to baseline range before the end of the adaptation period. After the adaptation, no aftereffects were observed in backward walking on tied-belts, indicating a lack of adaptation transfer. Despite 5 min of backward walking washout, robust aftereffects persisted in forward walking on tied-belts. **Supplementary Video 1** animates the performance of a single subject over the course of this experiment, illustrating the strong aftereffect of training on forward walking and the absence of this effect being transferred to backward walking.

Average interlimb phases were calculated for the following periods: baseline backward walking, baseline forward walking, early and late forward walking adaptation, early and late postadaptation backward walking, and early and late postadaptation forward walking. Group average (Fig. 2d) interlimb phase was significantly different across experimental periods ($P < 0.001$). Forward walking adaptation was indicated by a phase change from early to late split-belt forward walking adaptation ($P < 0.001$), and from baseline to early postadaptation forward walking on tied-belts ($P < 0.001$). Phase was not significantly different between baseline backward walking and early postadaptation backward walking on tied-belts ($P \geq 0.5$), indicating a lack of adaptation transfer from forward to backward walking. To confirm that backward walking did not partially wash out forward walking adaptation, we compared these results with those obtained without any backward walking washout (Fig. 2d). There was no difference between the group with and the group without backward walking washout (group effect, $P = 0.5$; group \times period interactions, $P = 0.06$), clearly demonstrating that backward walking did not washout forward walking aftereffects. Analysis of interlimb

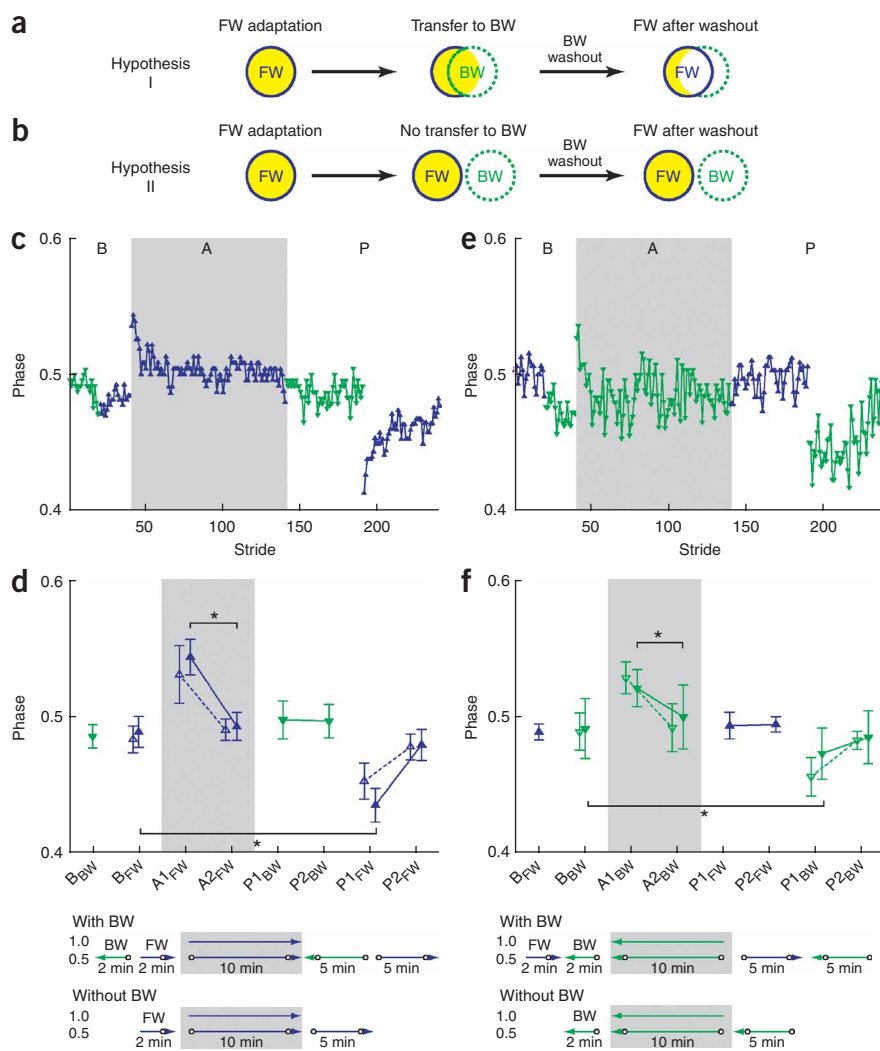


Figure 2 Adaptation does not transfer between forward and backward walking. **(a)** Hypothesis I: if forward walking (FW) and backward walking (BW) neural circuits overlap, then adapting FW will affect BW, and de-adapting BW will washout FW adaptation. **(b)** Hypothesis II: if the circuits are separated, then adapting FW will not affect BW, and BW will not washout FW adaptation. Yellow regions represent adapted circuits.

(c,d) Experiment I: we tested for transfer of adaptation from FW to BW. Stride-by-stride interlimb phase from a typical subject plotted for the first 20 strides of baseline BW (green) and FW (blue) on tied belts, the first 100 strides of FW adaptation on split-belts (shaded gray), and the first 50 strides of postadaptation BW and FW on tied belts is shown in **c**. Average interlimb phases of subjects who carried out experiment I ($n = 7$; solid line) and those who carried out the control experiment without BW washout are shown in **d** ($n = 7$; dotted line), calculated for B_{FW} (baseline FW), B_{BW} (baseline BW), $A1_{FW}$ (early adaptation FW), $A2_{FW}$ (late adaptation FW), $P1_{BW}$ (early postadaptation BW), $P2_{BW}$ (late postadaptation BW), $P1_{FW}$ (early postadaptation FW) and $P2_{FW}$ (late postadaptation FW). Error bars are ± 1 s.d. * indicates $P < 0.05$. Summary of behavioral procedures (bottom) showing the duration of each period, and the belts speed(s) and direction used. Single arrows represent tied belts and parallel arrows represent split belts. Circles represent time points at which average interlimb phase was calculated. **(e,f)** Results from experiment II, where we tested for transfer of adaptation from BW to FW.

adaptation). In experiment III, subjects adapted backward walking with, for example, the right leg fast and left leg slow on split-belts for 8 min. They then adapted forward walking in the opposite manner (that is, the right leg slow and the left leg fast) on split-belts for

coordination in terms of step length shows the same pattern of aftereffects (**Supplementary Data** online).

In experiment II, we tested whether the pattern of (or rather, the lack of) transfer was symmetric between forward and backward walking. Subjects adapting backward walking on split-belts did not show forward walking aftereffects, and forward walking did not washout backward walking aftereffects (**Fig. 2e,f** and **Supplementary Video 2** online). Note that backward walking was more variable, probably because this was a more unusual situation. Statistical analysis showed that interlimb phase changed ($P < 0.001$) over the course of the experiment; there was a phase change from early to late backward walking adaptation ($P < 0.005$) and a phase change from baseline to early postadaptation backward walking ($P < 0.01$), but no transfer to forward walking, as phase was not significantly different between baseline and early postadaptation forward walking ($P \geq 0.5$). Here again we ran a control experiment to determine backward walking aftereffects from split-belt training without forward walking washout, and found no difference between groups (group effect, $P = 0.5$; group \times period interactions, $P = 0.4$). Analysis of step length shows the same pattern of aftereffects (**Supplementary Data**).

If forward and backward walking were truly independent, then we would predict that concurrent and opposing adaptations in forward and backward walking should not interfere with one another (that is, dual

another eight minutes (**Fig. 3** and **Supplementary Video 3** online). The leg that was trained fast during backward adaptation was phase advanced initially, and gradually shifted back to baseline. The same leg trained slow during forward adaptation was phase lagged initially, and gradually phase shifted back to baseline before the end of forward adaptation. Post dual-adaptation, aftereffects were present in backward walking on tied-belts and were gradually washed out. Forward walking aftereffects persisted after backward walking aftereffects had been washed out. Statistical analysis revealed significant phase changes across periods ($P = 0.007$): phase changed from early to late backward walking adaptation ($P < 0.01$), from early to late forward walking adaptation ($P < 0.005$), between baseline and early postadaptation backward walking ($P < 0.005$), and between baseline and early postadaptation forward walking ($P < 0.005$). We controlled for possible interferences of adaptation to two different treadmill conditions by comparing the performance of subjects who carried out dual adaptation with the performance of those who carried out single adaptation. Phase changes for corresponding periods were not different between backward walking alone versus backward walking during dual adaptation (group effect, $P = 0.70$; group \times period, $P = 0.70$). Similarly, no differences were found between the groups that carried out forward walking alone versus forward walking during dual adaptation (group effect, $P = 0.90$; group \times period, $P = 0.90$). These data support the conclusion that

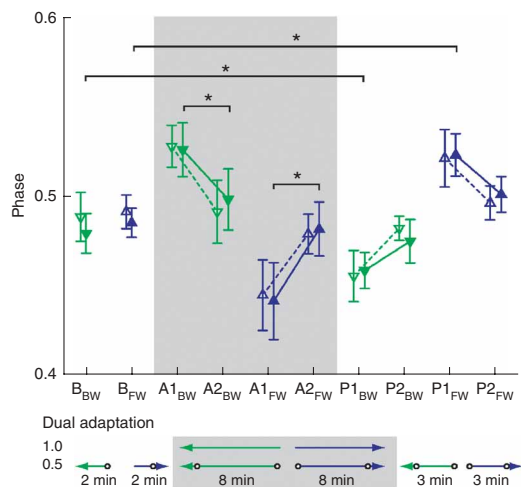


Figure 3 Dual storage of forward and backward aftereffects. Experiment III: we tested whether FW and BW are separately adaptable. Average interlimb phase for subjects who carried out dual-adaptation experiment ($n = 6$; solid blue and green lines), subjects who carried out the backward control experiment ($n = 5$; dotted green line), and subjects who carried out the forward control experiment ($n = 7$; dotted blue line) calculated for B_{BW} , B_{FW} , $A1_{BW}$ (early BW adaptation), $A2_{BW}$ (late BW adaptation), $A1_{FW}$, $A2_{FW}$, $P1_{BW}$, $P2_{BW}$, $P1_{FW}$ and $P2_{FW}$. Error bars are ± 1 s.d. * indicates $P < 0.05$. Summary of behavioral procedure (bottom) showing the duration of each period, and the belts speed(s) and direction used. Single arrows represent tied belts and parallel arrows represent split belts. Circles represent time points at which average interlimb phases were calculated.

forward and backward walking depend on separate neural circuits that are independently adaptable.

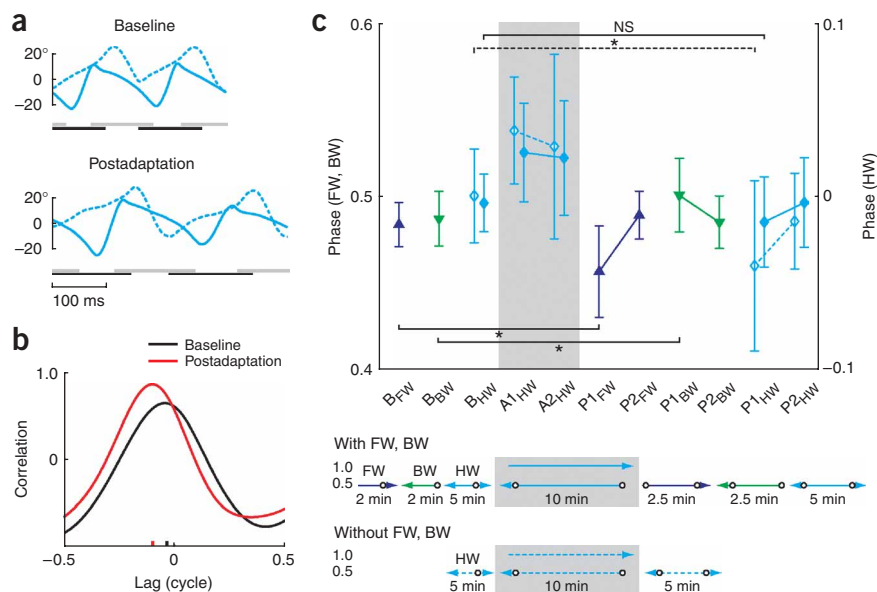
In experiment IV, we studied whether the left and right legs could be individually adapted. Training was done with the belts moving in opposite directions at different speeds, such that one leg stepped forward and the other stepped backward (hybrid walking). Like human infants¹⁴, adults could immediately carry out hybrid walking with no prior training. Limb movements were approximately in-phase at baseline (Fig. 4a,b), when the belts moved in opposite directions at the same speed. We adapted subjects to walk with the belts moving at different speeds (Supplementary Video 4 online). That the two legs can be adapted in opposite directions was confirmed by the presence of an aftereffect when the legs walked at the same speed during post-adaptation (Fig. 4a,b).

We first confirmed that hybrid walking *per se* did not induce aftereffects in forward or backward walking ($n = 7$, $P > 0.5$). We then examined whether hybrid walking adaptation transfers to forward

and backward walking (Fig. 4c and Supplementary Video 5 online) to determine individual leg effects. During adaptation, subjects were trained to step forward on the fast-belt and backward on the slow-belt during adaptation. After hybrid walking adaptation, forward walking showed aftereffects: the leg that was trained to step forward on the fast-belt was phase lagged relative to the other leg on tied-belts. Forward walking aftereffects were washed out and phase returned to baseline range by the end of the forward walking washout period. Subsequent backward walking showed aftereffects with a phase change in which the leg that was trained to step backward on the slow-belt was phase lagged relative to the other leg. Backward walking aftereffects were washed out before the end of the backward walking washout period. Subsequent hybrid walking on tied-belts did not show robust aftereffects.

Statistical analysis of experiment IV indicated that phase changes were significant between baseline and postadaptation for forward walking ($P < 0.001$) and for backward walking ($P < 0.01$), but were not significant between baseline and postadaptation for hybrid walking ($P = 0.1$). In contrast, hybrid walking training without forward and backward walking washout periods (control experiment) produced significant hybrid walking aftereffects in postadaptation relative to baseline ($P < 0.005$). Hybrid walking aftereffects were smaller in experiment IV compared with control experiment without intervening

Figure 4 Adaptation induces individual leg effects. (a–c) Experiment IV. Subjects adapted walking on belts moving in opposite directions at different speeds. During hybrid walking (HW), one leg stepped forward, while the other stepped backward. We tested for transfer of HW adaptation to both FW and BW to determine individual leg effects. Example limb angle trajectories for the forward stepping (solid) and backward stepping (dotted) legs during baseline and postadaptation HW are shown in a. Horizontal lines represent stance phases for the backward-stepping (black) and forward-stepping (gray) legs. Example cross-correlation functions for baseline and postadaptation HW (without washout) are shown in b. Baseline phase is close to 0 cycles, reflecting in-phase limb kinematics. Note that stance phases remained alternating. After adaptation, phase shifted relative to baseline. Average interlimb phases for subjects who completed experiment IV ($n = 15$; solid line), and for those who completed the control experiment without FW and BW washout are shown in c ($n = 10$; dotted line), calculated for B_{BW} , B_{FW} , B_{HW} (baseline HW), $A1_{HW}$ (early HW adaptation), $A2_{HW}$ (late HW adaptation), $P1_{BW}$, $P2_{BW}$, $P1_{FW}$, $P2_{FW}$, $P1_{HW}$ (early postadaptation HW) and $P2_{HW}$ (late postadaptation HW). Error bars are ± 1 s.d. * indicates $P < 0.05$; NS, not significant. Summary of behavioral procedure (bottom) showing the duration of each trial, and the belts speed(s) and direction used. Single arrows represent tied belts and parallel arrows represent split belts. Circles represent time points at which average interlimb phases were calculated.



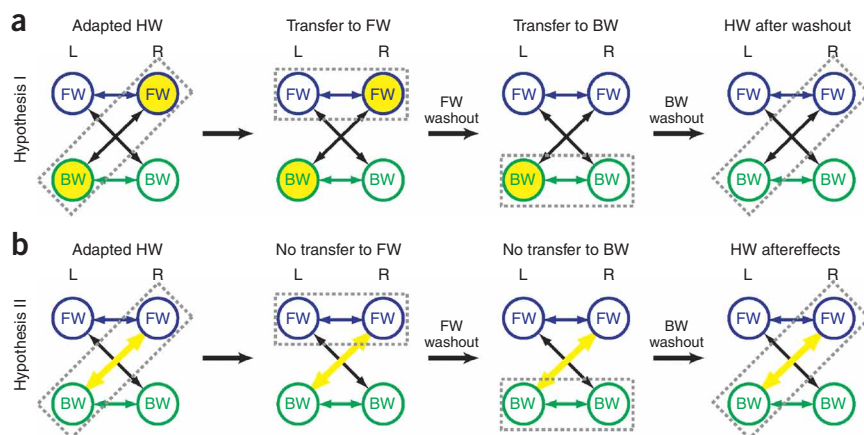


Figure 5 A schematic model of leg-specific adaptation. **(a)** FW and BW patterns are independently adaptable on each leg. Adaptive changes are represented by yellow-filled circles. The two legs can be flexibly coupled to one another and each leg can adapt independently. During HW, the right leg walking forward interacts with the left leg walking backward (dashed box). When the belts moved at different speeds, adaptive mechanisms induced changes in the properties of both legs. Following training, the adapted right leg walking forward interacted with the left counterpart to produce aftereffects in FW. Similarly, the adapted left leg walking backward interacted with the right counterpart to produce aftereffects in BW. The adaptive changes to each leg were washed out separately in the FW and BW washout periods. Subsequently, HW showed no aftereffects. **(b)** Alternatively, adaptation could affect bilateral connections that coordinate the left and right legs (yellow arrows). If this were the case, then adaptation should not transfer to FW or BW, and HW should show aftereffects even after FW and BW washouts. Our results do not support hypothesis II.

forward and backward walking washouts, but the difference only approached significance ($P = 0.10$). Analysis of step length also showed a significant transfer of hybrid walking adaptation to both forward and backward walking (**Supplementary Data**). Taken together, these results demonstrate that hybrid walking adaptation transfers to both forward and backward walking, and that forward and backward walking largely wash out hybrid walking aftereffects.

A schematic model that would explain our results is illustrated in **Figure 5a**. We propose that independently adaptable circuits (yellow circles) exist for forward and backward stepping on each leg. The two legs can be flexibly coupled to one another, and each can be adapted independently of its contralateral counterpart. During hybrid walking, the right leg walking forward interacts with the left leg walking backward (dashed box). When the belts move at different speeds, adaptive mechanisms induce changes in the properties of both legs. Following training, the adapted right leg walking forward interacts with the left counterpart to produce aftereffects in forward walking (dashed box). Similarly, the adapted left leg walking backward interacts with the right counterpart to produce aftereffects in backward walking. The adaptive changes to each leg revert separately in the forward and backward walking washout periods, and subsequently there is normal in-phase hybrid walking.

On the other hand, our results cannot be explained by the adaptation of neural elements (yellow arrows) mediating bilateral interactions between two pattern-generating circuits (**Fig. 5b**). That is, it is unlikely that adaptation influences the connections between leg- and direction-specific circuits. These would represent various types of commissural interneurons involved in left-right coordination¹⁶. There exist descending fibers that have specific modulatory effects on the crossed commissural interneuron connections¹⁷. These modulatory effects could potentially change the balance between crossed excitation and inhibition pathways, and thereby influence left-right coordination patterns. If that were the case, then hybrid walking

adaptation should not transfer to forward or backward walking, and hybrid walking should show aftereffects even after forward and backward walking washout periods. Our results show that this is not the case.

DISCUSSION

Our results indicate that walking patterns are direction and leg specific in humans. Adaptation to forward and backward walking are strictly independent in that they do not transfer between directions, nor interfere with one another. Furthermore, each leg can be adapted separately from its contralateral counterpart during hybrid walking, and the effects from adaptation are stored individually for each leg. These findings suggest that plasticity associated with locomotor adaptation in human is both leg and direction specific.

It is known that the context in which an individual trains affects whether adaptation transfers to untrained movements^{18,19}. Here, the direction that the treadmill belts move in and the form of locomotion (for example, backward) could be considered the 'context' in which adaptation occurs. Both forward and backward split-belt walking are strictly context dependent. Hybrid walking is a different and

completely novel context: the treadmill belts move in opposite directions, and this form of locomotion has in-phase, rather than anti-phase, kinematics. Despite the context cues that could have allowed hybrid walking to undergo independent adaptation, it instead transferred to both forward and backward walking. Thus, the hybrid walking experiment suggests that context cues can also be leg specific, with the forward walking context cue on one leg affecting only forward walking, and vice versa for backward walking. We interpret this direction and leg specificity as reflecting the basic organization of the underlying adapted components.

This pattern of generalization is not limited to the interlimb coordination studied here, but is also true when individuals learn to adjust walking to an environmental perturbation. Others have shown that when humans learn a constant effect in the environment, such as stepping onto a moving platform, they do not generalize the learned anticipatory adjustment from forward to backward walking²⁰. Consistent with our results, learning was the result of a specific direction-dependent mechanism, which may be joint- or muscle-specific²⁰.

We have previously shown that adaptive locomotor control is dependent on the cerebellum. Our results in humans⁴, and others' results in cats²¹, demonstrate that cerebellar lesions abolish the ability to adapt interlimb coordination without impairing the immediate reaction to split-belt treadmill perturbations. The cerebellum seems to be important in recalibrating predictive feedforward control—not only for locomotion⁴, but also for a host of other motor adaptation paradigms^{22–24}. This is presumably done through comparisons between predicted and actual movement outcomes²⁵. Afferent signals via dorsal spinocerebellar pathways inform the cerebellum about the actual orientation and loading of one or both legs^{26–29}, which could be compared with an internal prediction. The cerebellum also receives, from the ventral spinocerebellar tract, signals that reflect the activity of spinal networks that generate stepping³⁰. The cerebellum then influences walking via its efferent projections to descending brainstem

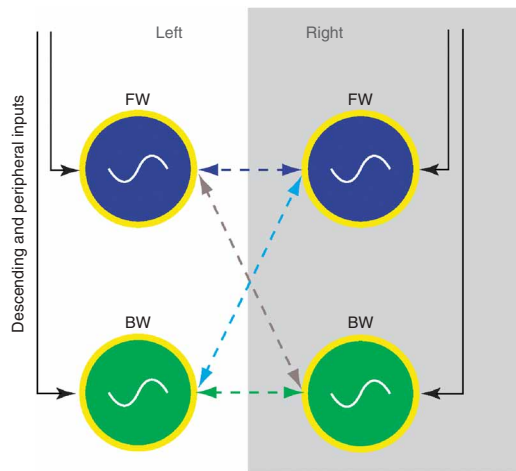


Figure 6 Sketch of proposed organization of adaptable locomotor networks in human. Basic locomotor networks are configured and modulated by descending signals as well as sensory feedback. The functional networks involved in motor pattern generation in FW and BW are largely nonoverlapping. Interlimb coordination is adapted through modulations to elements in individual oscillatory locomotor network for each leg (yellow circles), rather than through neural elements that coordinate a specific pair of oscillators (colored dotted lines).

motor tracts, including the vestibulospinal, reticulospinal and rubrospinal tracts^{31,32}. Our current results further suggest that these mechanisms can maintain accurate predictive calibrations that are distinct for forward and backward walking.

Little is known about the network structure of locomotor CPGs in humans compared with the extensive knowledge about other vertebrate systems. Studies of infant stepping have shown that the organizational principles in humans are similar to those of other terrestrial animals³³. Split-belt treadmill studies in human infants^{14,33} and cats³⁴ have reported the same interlimb pattern under similar conditions. Both species can take multiple steps on the fast belt during a single step cycle of the opposite leg on the slow belt. These observations provide compelling evidence that the basic pattern generator for each leg can operate independently of the other.

Our current findings add to the understanding of adaptive reorganization of locomotor circuits in human adults. The specificity of adaptations demonstrated here cannot be explained by changes to a common rhythm generator that acts on multiple motor pattern generators^{11,12}. Instead, it seems that changes affect more specialized circuits for a given leg and direction of walking. These circuits may control specific motor synergies involved in different forms of locomotion³⁵.

Adaptation also did not seem to affect specific circuit components used for left-right coordination. These components ensure out-of-phase coordination between legs during normal walking, or in-phase coordination, as seen in our hybrid walking or in jumping. Various types of commissural interneurons have been identified in the cat and rodent spinal cord that project to contralateral interneurons and motor neurons¹⁶. Out-of-phase and in-phase coordination are dependent on the balance between inhibitory and excitatory crossed connections³⁶, which are controlled by descending modulatory fibers¹⁷. We have shown here that human interlimb phasing is not adapted by changing the balance or strength of crossed connections, as adaptation of an in-phase pattern (hybrid walking) transferred to out-of-phase patterns (forward and backward walking).

We propose that independently adaptable locomotor networks exist for each leg in humans (Fig. 6). The basic locomotor networks are

configured and modulated by feedforward commands from descending brain signals as well as via sensory feedback from afferent inputs. Our results suggest that the functional networks involved in motor pattern generation in forward and backward walking are largely nonoverlapping in human. We do not suggest that they are completely spatially separated networks, but we think that they represent functional networks that can be altered independently. Interlimb coordination is controlled through descending modulation of elements in individual locomotor networks for each leg, rather than neural connections that couple a particular pair of oscillators.

Specialized locomotor adaptability has the advantage of context dependency and specificity. This means that our locomotor system can learn new patterns without compromising other related patterns. An increased understanding of the types of training procedures that show functional independence, along with an increased understanding of the sensitivity of motor adaptability to damage at various levels in the nervous system, are both critical for efficient design of effective rehabilitation protocols.

METHODS

Subjects. Forty healthy volunteers (19 males, 21 females; median age, 25 years) participated in this study. All subjects gave informed written consent before participating. Thirteen subjects participated in more than one experiment. The protocols were approved by the Johns Hopkins Institutional Review Board.

Experimental setup and design. We studied split-belt walking adaptation using a custom-built treadmill (Woodway) that had two separate belts driven by independent motors. Speed and direction commands for each belt were sent to the treadmill through a computer interface written in MATLAB (Math-Works). Subjects were positioned in the middle of the treadmill, with one foot on each belt. They held onto a front rail that was adjusted to elbow height, and wore a safety harness suspended from the ceiling, which did not support body weight. At the beginning of each trial, treadmill belts were stationary and subjects were informed about the direction in which the belts were going to move, but not the speeds. Once the belts started moving, subjects were instructed not to look down at the belts while walking on the treadmill. In the 'tied-belts' condition, both belts were set to move at 0.5 m s⁻¹; in the 'split-belts' condition, one belt was set at 0.5 m s⁻¹, while the other was set at 1.0 m s⁻¹. In both forward and backward walking, the two belts were set to move in the same direction; in hybrid walking, the right belt was set to forward, while the left belt was set to move backward.

Each experiment consisted of a baseline period, where the relevant types of walking (that is, forward, backward and/or hybrid walking) were tested with tied belts, an adaptation period, where one (or more) types of walking were exposed to split belts, and a postadaptation period, where the relevant types of walking were retested with tied belts to assess aftereffects. The postadaptation period was where we tested for transfer to untrained types of walking and any washout of aftereffects in the trained type of walking.

In Experiment I, we tested for transfer of split-belt forward walking training to backward walking ($n = 7$). Baseline forward and backward walking were 2 min each on tied belts, adaptation was 10 min of forward walking on split belts, and postadaptation was 5 min of backward walking followed by 5 min of forward walking on tied belts. Transfer to backward walking was evaluated by comparing baseline relative to postadaptation backward walking. We also assessed whether backward walking washed out forward walking aftereffects by comparing forward walking aftereffects from this experiment to those from a separate forward walking adaptation experiment with no backward walking washout period ($n = 7$).

In Experiment II, we tested for transfer of split-belt backward walking training to forward walking ($n = 7$). The procedure was identical to that in Experiment I, but with backward walking periods replacing forward walking periods, and vice versa. Backward walking was adapted, and transfer to forward walking was evaluated from aftereffects in forward walking relative to baseline. We also assessed whether forward walking washed out backward walking aftereffects by comparing backward walking aftereffects from this experiment

with those from a separate backward walking adaptation experiment with no forward walking washout period ($n = 5$).

In Experiment III, we tested whether subjects can store opposite forward and backward walking aftereffects ($n = 6$). Baseline forward and backward walking were 2 min each on tied belts, followed by two consecutive adaptation periods: backward walking on split belts for 8 min and then forward walking on split belts for 8 min. The leg that was trained on the fast belt during backward walking was subsequently trained to walk on the slow belt during forward walking. After the two training periods on split belts, subjects carried out postadaptation trials, with 3 min of backward walking on tied belts, and then 3 min of forward walking on tied belts. We compared the aftereffects in forward and backward walking from dual adaptation with that from forward and backward walking adaptation alone without any washout (control experiments from I and II, see above).

In Experiment IV, we tested for transfer of split-belt hybrid walking training to both forward and backward walking ($n = 15$). Baseline forward and backward walking were 2 min each on tied belts. Baseline hybrid walking was also carried out on belts moving at the same speed, but in opposite directions. All subjects were able to step forward on one leg and backward on the other leg immediately after the belts moved in opposite directions. Baseline hybrid walking was 5 min to ensure that subjects felt comfortable with this unusual pattern. Adaptation for hybrid walking was 10 min, with the right belt moving forward twice as fast as the left belt moving backward. Postadaptation was 2.5 min of backward walking on tied belts, 2.5 min of forward walking on tied belts and then 2.5 min of hybrid walking on belts moving at the same speed in opposite directions. Transfer to forward and backward walking were evaluated from aftereffects in forward and backward walking relative to their baselines. We also assessed whether forward and backward walking washed out aftereffects in hybrid walking by comparing hybrid walking aftereffects with those from a separate hybrid walking adaptation experiment with no washout periods ($n = 10$). As a further control experiment, we tested whether hybrid walking alone (that is, without one leg moving faster than the other) caused aftereffects in forward and backward walking ($n = 7$).

Data collection. Kinematic data were collected at 100 Hz using Optotrak (Northern Digital). Infrared-emitting markers were placed bilaterally over the following joints (Fig. 1a): foot (fifth metatarsal head), ankle (lateral malleolus), knee (lateral femoral epicondyle), hip (greater trochanter), pelvis (iliac crest) and shoulder (acromion process). The coordinate system was aligned such that the x axis was parallel to the treadmill belts, the y axis was parallel to the vertical line, and the z axis was parallel to the horizontal line perpendicular to the x - y plane. Foot switches were used to record times of foot contact and lift off.

Data analysis. Limb angle was calculated as the angle between the vertical line and the vector from hip to foot on the x - y plane; it has positive values when the foot is in front of the hip (Fig. 1a). The coordination between two limb movements was measured by calculating the cross-correlation function (Signal Processing Toolbox, MATLAB) between the limb angle trajectories over one stride cycle. Interlimb phase was defined to be the lag time at peak correlation (Fig. 1b). For forward and backward walking, cross-correlation was estimated over lag range [0, 1] stride cycles and peak correlation lay around 0.5, which reflects out-of-phase coordination. For hybrid walking, cross-correlation was estimated over lag range [-0.5, 0.5] cycles and peak correlation lay around 0, which reflects in-phase coordination. By convention, positive lag times indicate a lead of the nonreference limb relative to the reference limb. The reference limb was defined to be the leg on the slow belt during split-belt training. For experiment III, with two split-belt training periods, the reference limb was defined to be the leg on the slow belt during backward walking split-belt training.

Statistical analysis. We used circular statistics to calculate mean phase and s.d. for the first five strides in each baseline period, the first and last five strides in each adaptation period (early and late adaptation, respectively), and the first and last five strides in each postadaptation period, (early and late postadaptation, respectively). Unit vectors with an angle of $2\pi\phi_i$ represented individual phase values, ϕ_i , such that phase value of one cycle corresponds to 2π . The resultant mean vector angle and angular deviation³⁷ divided by 2π corresponds to mean phase and s.d., respectively.

Aftereffects were assessed with a paired-sample tests for circular data³⁸ between baseline and early postadaptation for each type of locomotion. For the trained type(s) of locomotion, a paired-sample test was also carried out for early versus late adaptation. To determine whether aftereffects were washed out, the Watson's U^2 (two-sample) test for circular data³⁷ was used to compare the aftereffects from each experiment with their respective control experiment(s) with no washout trials. We used $\alpha = 0.05$ as the alpha level for each planned pair-wise comparison.

For experiments I, II and III, mean phase values did not span more than half a cycle. Therefore, we also ran separate repeated-measures analysis of variance in Statistica (StatSoft): one to test whether phase changed significantly across all time points, and a second one to test whether phase changes were different between experimental groups.

Note: Supplementary information is available on the Nature Neuroscience website.

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AUTHOR CONTRIBUTIONS

J.T.C. and A.J.B. designed the experiments and cowrote the manuscript. J.T.C. conducted the experiments and data analyses.

COMPETING INTERESTS STATEMENT

The authors declare no competing financial interests.

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- Lam, T., Anderschitz, M. & Dietz, V. Contribution of feedback and feedforward strategies to locomotor adaptations. *J. Neurophysiol.* **95**, 766–773 (2006).
- Pearson, K.G. Neural adaptation in the generation of rhythmic behavior. *Annu. Rev. Physiol.* **62**, 723–753 (2000).
- Reisman, D.S., Block, H.J. & Bastian, A.J. Interlimb coordination during locomotion: what can be adapted and stored? *J. Neurophysiol.* **94**, 2403–2415 (2005).
- Morton, S.M. & Bastian, A.J. Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *J. Neurosci.* **26**, 9107–9116 (2006).
- Reisman, D.S., Wityk, R., Silver, K. & Bastian, A.J. Locomotor adaptation on a split-belt treadmill can improve walking symmetry post-stroke. *Brain*, published online 2 April 2007 (doi:10.1093/brain/awm035).
- Grillner, S. & Zangger, P. On the central generation of locomotion in the low spinal cat. *Exp. Brain Res.* **34**, 241–261 (1979).
- Martin, L.I. & Stein, P.S. Spinal cord segments containing key elements of the central pattern generators for three forms of scratch reflex in the turtle. *J. Neurosci.* **9**, 2285–2296 (1989).
- Calancie, B. *et al.* Involuntary stepping after chronic spinal cord injury. Evidence for a central rhythm generator for locomotion in man. *Brain* **117**, 1143–1159 (1994).
- Dimitrijevic, M.R., Gerasimenko, Y. & Pinter, M.M. Evidence for a spinal central pattern generator in humans. *Ann. NY Acad. Sci.* **860**, 360–376 (1998).
- Grasso, R., Bianchi, L. & Lacquaniti, F. Motor patterns for human gait: backward versus forward locomotion. *J. Neurophysiol.* **80**, 1868–1885 (1998).
- Lafreniere-Roula, M. & McCreary, D.A. Deletions of rhythmic motoneuron activity during fictive locomotion and scratch provide clues to the organization of the mammalian central pattern generator. *J. Neurophysiol.* **94**, 1120–1132 (2005).
- Rybak, I.A., Shevtsova, N.A., Lafreniere-Roula, M. & McCreary, D.A. Modelling spinal circuitry involved in locomotor pattern generation: insights from deletions during fictive locomotion. *J. Physiol. (Lond.)* **577**, 617–639 (2006).
- Lamb, T. & Yang, J.F. Could different directions of infant stepping be controlled by the same locomotor central pattern generator? *J. Neurophysiol.* **83**, 2814–2824 (2000).
- Yang, J.F., Lamont, E.V. & Pang, M.Y. Split-belt treadmill stepping in infants suggests autonomous pattern generators for the left and right leg in humans. *J. Neurosci.* **25**, 6869–6876 (2005).
- Dietz, V., Zijlstra, W. & Duysens, J. Human neuronal interlimb coordination during split-belt locomotion. *Exp. Brain Res.* **101**, 513–520 (1994).
- Kiehn, O. Locomotor circuits in the mammalian spinal cord. *Annu. Rev. Neurosci.* **29**, 279–306 (2006).
- Hammar, I., Bannatyne, B.A., Maxwell, D.J., Edgley, S.A. & Jankowska, E. The actions of monoamines and distribution of noradrenergic and serotonergic contacts on different subpopulations of commissural interneurons in the cat spinal cord. *Eur. J. Neurosci.* **19**, 1305–1316 (2004).
- Morton, S.M. & Bastian, A.J. Prism adaptation during walking generalizes to reaching and requires the cerebellum. *J. Neurophysiol.* **92**, 2497–2509 (2004).
- Krakauer, J.W., Mazzoni, P., Ghazizadeh, A., Ravindran, R. & Shadmehr, R. Generalization of motor learning depends on the history of prior action. *PLoS Biol.* **4**, e316 (2006).

20. Reynolds, R.F. & Bronstein, A.M. The moving platform aftereffect: limited generalization of a locomotor adaptation. *J. Neurophysiol.* **91**, 92–100 (2004).
21. Yanagihara, D. & Kondo, I. Nitric oxide plays a key role in adaptive control of locomotion in cat. *Proc. Natl. Acad. Sci. USA* **93**, 13292–13297 (1996).
22. Horak, F.B. & Diener, H.C. Cerebellar control of postural scaling and central set in stance. *J. Neurophysiol.* **72**, 479–493 (1994).
23. Lang, C.E. & Bastian, A.J. Cerebellar subjects show impaired adaptation of anticipatory EMG during catching. *J. Neurophysiol.* **82**, 2108–2119 (1999).
24. Smith, M.A. & Shadmehr, R. Intact ability to learn internal models of arm dynamics in Huntington's disease but not cerebellar degeneration. *J. Neurophysiol.* **93**, 2809–2821 (2005).
25. Bastian, A.J. Learning to predict the future: the cerebellum adapts feedforward movement control. *Curr. Opin. Neurobiol.* **16**, 645–649 (2006).
26. Bosco, G., Poppele, R.E. & Eian, J. Reference frames for spinal proprioception: limb-endpoint based or joint-level based? *J. Neurophysiol.* **83**, 2931–2945 (2000).
27. Bosco, G., Eian, J. & Poppele, R.E. Kinematic and nonkinematic signals transmitted to the cat cerebellum during passive treadmill stepping. *Exp. Brain Res.* **167**, 394–403 (2005).
28. Bosco, G., Eian, J. & Poppele, R.E. Phase-specific sensory representations in spinocerebellar activity during stepping: evidence for a hybrid kinematic/kinetic framework. *Exp. Brain Res.* **175**, 83–96 (2006).
29. Poppele, R.E., Rankin, A. & Eian, J. Dorsal spinocerebellar tract neurons respond to contralateral limb stepping. *Exp. Brain Res.* **149**, 361–370 (2003).
30. Arshavsky, Y.I., Gelfand, I.M., Orlovsky, G.N. & Pavlova, G.A. Messages conveyed by spinocerebellar pathways during scratching in the cat. II. Activity of neurons of the ventral spinocerebellar tract. *Brain Res.* **151**, 493–506 (1978).
31. Orlovsky, G.N. The effect of different descending systems on flexor and extensor activity during locomotion. *Brain Res.* **40**, 359–371 (1972).
32. Orlovsky, G.N. Activity of vestibulospinal neurons during locomotion. *Brain Res.* **46**, 85–98 (1972).
33. Yang, J.F. *et al.* Infant stepping: a window to the behaviour of the human pattern generator for walking. *Can. J. Physiol. Pharmacol.* **82**, 662–674 (2004).
34. Forssberg, H., Grillner, S., Halbertsma, J. & Rossignol, S. The locomotion of the low spinal cat. II. Interlimb coordination. *Acta Physiol. Scand.* **108**, 283–295 (1980).
35. Berkowitz, A. & Stein, P.S. Activity of descending propriospinal axons in the turtle hindlimb enlargement during two forms of fictive scratching: phase analyses. *J. Neurosci.* **14**, 5105–5119 (1994).
36. Kullander, K. *et al.* Role of EphA4 and EphrinB3 in local neuronal circuits that control walking. *Science* **299**, 1889–1892 (2003).
37. Batschelet, E. *Circular Statistics in Biology* (Academic Press, New York, 1981).
38. Zar, J.H. *Biostatistical Analysis* (Prentice Hall, Upper Saddle River, New Jersey, 1999).