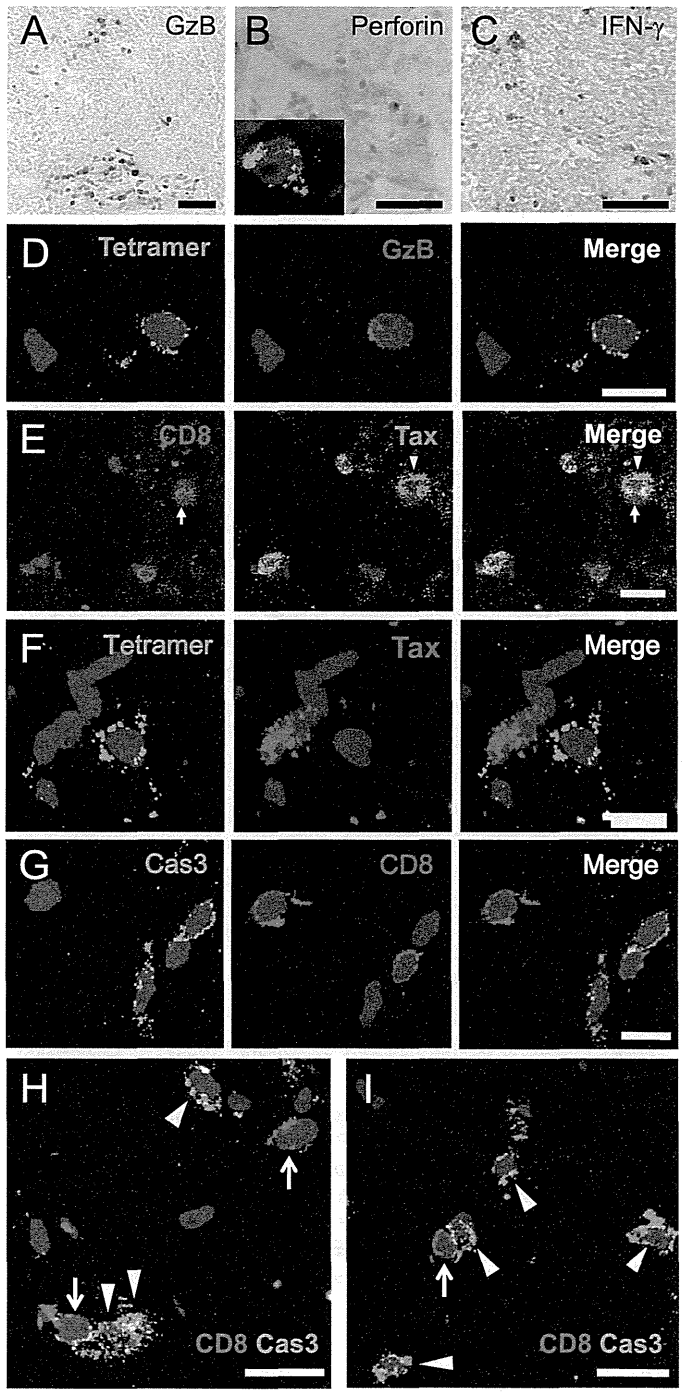


**FIGURE 5.** Cytotoxic T-lymphocyte (CTL) molecules in the CNS. **(A–C)** Immunohistochemistry shows granzyme B (GzB)–positive cells **(A)**, perforin-positive cells **(B)**, and interferon-γ (IFN-γ)–positive cells **(C)** in the perivascular area of the spinal cord of Patient 8624. Black bars indicate 100 μm. **(D)** Double staining with HLA-A*2402/Tax301-309-tetramer (green) and anti–granzyme B monoclonal antibody (mAb) (red) reveals a GzB-positive HTLV-1–specific CTL in the parenchyma of the spinal cord (Patient 6315). **(E)** A cell expressing HTLV-1 Tax protein is in contact with a CD8-positive cell in the spinal cord of Patient 6315. **(F)** An HTLV-1 Tax–specific CTL is next to the cell expressing Tax protein in the spinal cord of Patient 8614. **(G–I)** Double staining for active caspase-3 (Cas3) (green) and CD8 (red, arrows in **[H]** and **[I]**) reveals CD8-positive cells in contact with active caspase-3–positive cells in the spinal cord of Patient 8624. Nuclei were counterstained with DAPI. White bars indicate 10 μm.

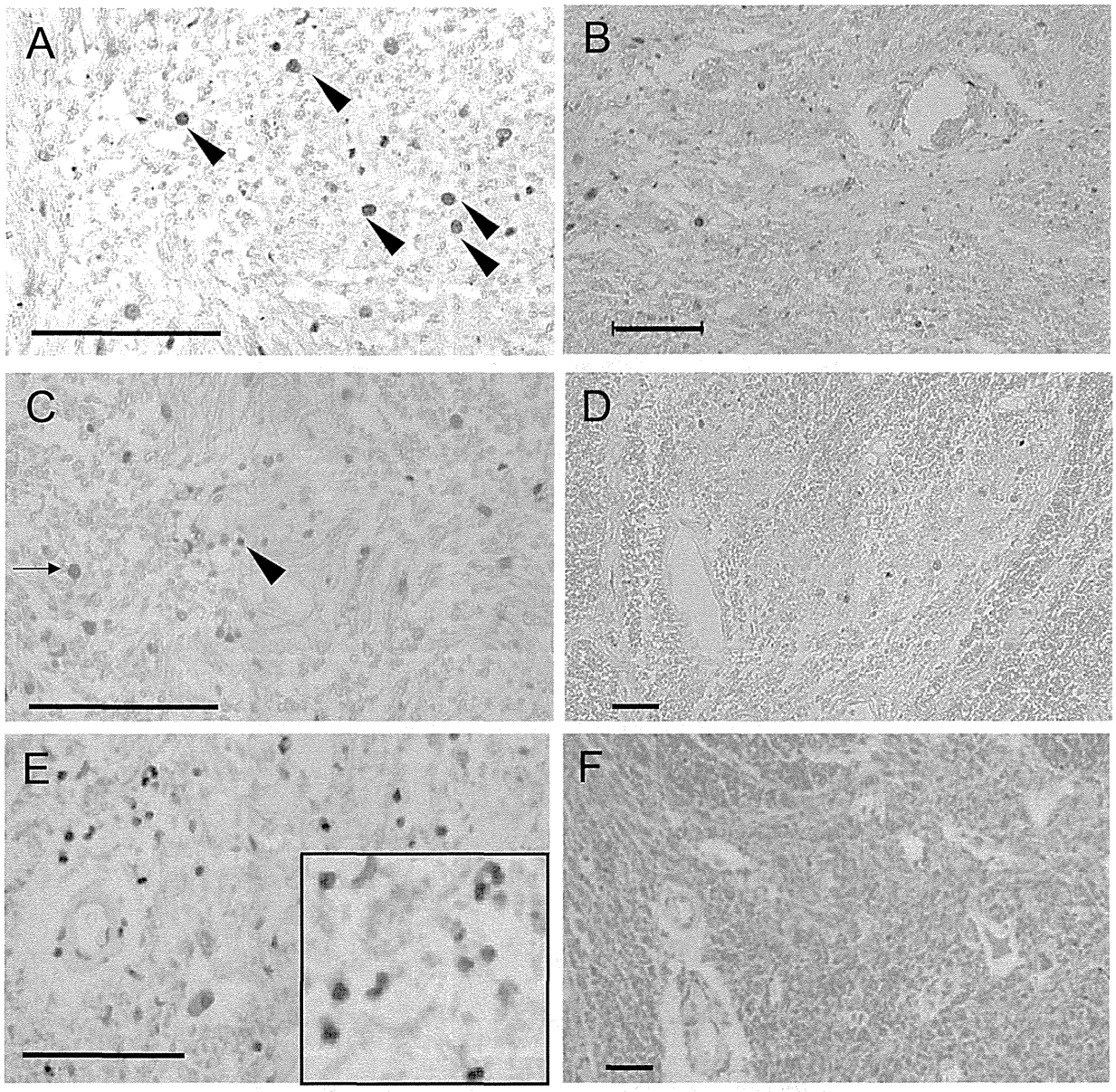


**FIGURE 6.** Detection of apoptotic cells in the CNS. **(A)** Some small cells are apoptotic (DAB; brown, arrowheads) detected by anti–active caspase-3 antibody (Ab). **(B–D)** TdT-mediated dUTP nick end labeling assay. **(B)** A number of apoptotic cells (DAB; brown) are detected in the spinal cord of a patient with HTLV-1–associated myelopathy/tropical spastic paraparesis (Patient 8624). **(C)** Some infiltrating small cells around a small vessel (arrowhead) and some relatively large cells in the parenchyma (arrow) are apoptotic. **(D)** The apoptotic cells are barely detectable in the control spinal cord from an HTLV-1–seronegative patient with hepatoma. **(E, F)** Anti-single-stranded DNA antibody staining. **(E)** Numerous apoptotic cells (AEC; red) are detected in the spinal cord (Patient 8624). A higher magnification picture in the inset shows apoptotic cells. **(F)** Apoptotic cells are barely detectable in the control patient spinal cord. Scale bar = 100 μm.

HTLV-1–infected cells could express viral antigens anywhere in the body of the infected individuals (14, 32), the expression of HTLV-1 proteins in vivo has remained elusive so far.

In this study, we succeeded in detecting HTLV-1 proteins in the CD4-positive T cells infiltrating the CNS. This is consistent with our previous reports in which HTLV-1–infected cells were determined to be CD4-positive lymphocytes in the CNS by in situ hybridization for HTLV-1 mRNA and in situ polymerase chain reaction for HTLV-1 DNA (33, 34). The infiltrating HTLV-1–infected CD4-positive cells may easily express the viral antigens in the CNS, which in turn facilitates the accumulation of HTLV-1–specific CTLs.

Human T-lymphotropic virus type-1 infection causes several organ-specific inflammatory diseases including HAM/TSP (2, 3). Previous reports demonstrating that HTLV-1 proviral loads are high in affected organs such as the muscles, lungs, and CNS suggest that HTLV-1–infected cells accumulate in the organs (13, 35). The pathogenesis model in which both

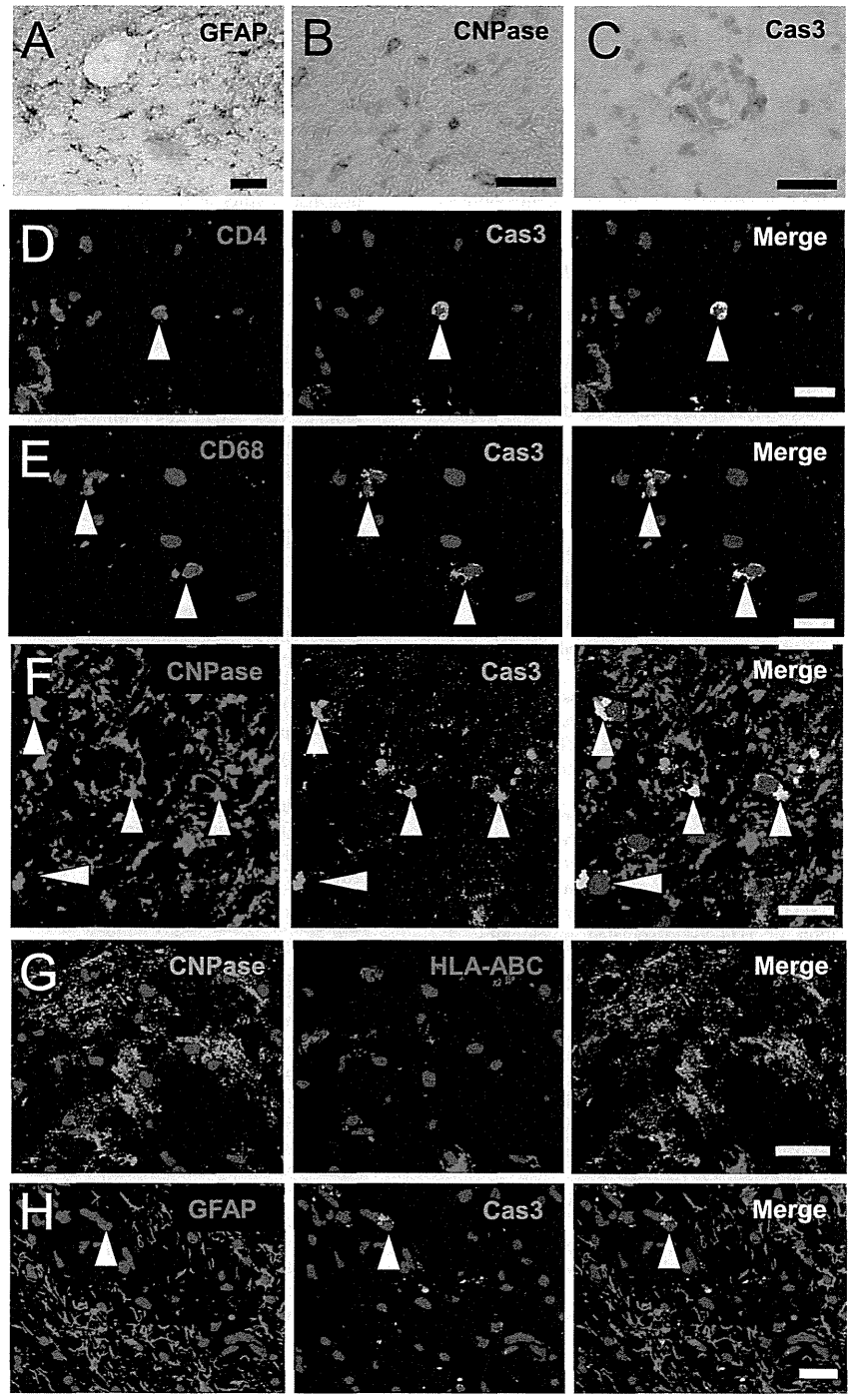


**FIGURE 7.** Cell identification of apoptotic cells. **(A–C)** Astrocytes **(A)**, oligodendrocytes **(B)**, and apoptotic cells **(C)** were stained with anti–glial fibrillary acidic protein (GFAP) antibody (Ab), anti–2′,3′-cyclic-nucleotide 3′-phosphodiesterase (CNPase) monoclonal antibody (mAb), or anti–active caspase-3 Ab, respectively, in the spinal cord of Patient 8624. Nuclei were counterstained with hematoxylin. **(D–F, H)** Double staining revealed that a CD4-positive cell (red, **D**), a CD68-positive cell (red, **E**), and some oligodendrocytes (red, **F**), but no astrocytes (red, **H**), were apoptotic (green) (arrowheads) in the spinal cord of Patient 8624. **(G)** Double staining with anti-CNPase mAb (green) and anti–HLA-ABC mAb (red) revealed that no oligodendrocyte expresses HLA-ABC. There is no double-positive signal (yellow) in the merged image. White bars indicate 20 μm. Cas3, active caspase-3.

HTLV-1–infected CD4-positive T cells and the virus-specific CD8-positive CTLs infiltrate the organs from the peripheral blood followed by bystander tissue damage may explain why HTLV-1 infection can cause several chronic inflammatory diseases in various organs. Further studies are needed to determine whether the similar immunopathologic model can be applied to HTLV-1–associated inflammatory diseases in other organs.

## ACKNOWLEDGMENT

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## REFERENCES

1. Uchiyama T, Yodoi J, Sagawa K, et al. Adult T-cell leukemia: Clinical and hematologic features of 16 cases. Blood 1977;50:481–92
2. Gessain A, Barin F, Vernant JC, et al. Antibodies to human T-lymphotropic virus type-I in patients with tropical spastic paraparesis. Lancet 1985;2: 407–10
3. Osame M, Usuku K, Izumo S, et al. HTLV-I–associated myelopathy: A new clinical entity. Lancet 1986;1:1031–32
4. Osame M, Matsumoto M, Usuku K, et al. Chronic progressive myelopathy associated with elevated antibodies to human T-lymphotropic virus type I and adult T-cell leukemialike cells. Ann Neurol 1987;21:117–22
5. Nakao K, Ohba N, Isashiki M, et al. Pigmentary retinal degeneration in patients with HTLV-I–associated myelopathy. Jap J Ophthalmol 1989; 33:383–91
6. Nishioka K, Maruyama I, Sato K, et al. Chronic inflammatory arthropathy associated with HTLV-I. Lancet 1989;1:441
7. Morgan OS, Rodgers-Johnson P, Mora C, et al. HTLV-1 and polymyositis in Jamaica. Lancet 1989;2:1184–87
8. Higuchi I, Nerenberg M, Yoshimine K, et al. Failure to detect HTLV-I by in situ hybridization in the biopsied muscles of viral carriers with polymyositis. Muscle Nerve 1992;15:43–47
9. Ozden S, Gessain A, Gout O, et al. Sporadic inclusion body myositis in a patient with human T cell leukemia virus type 1–associated myelopathy. Clin Infect Dis 2001;32:510–14
10. Matsuura E, Umehara F, Nose H, et al. Inclusion body myositis associated with human T-lymphotropic virus–type I infection: Eleven patients from an endemic area in Japan. J Neuropathol Exp Neurol 2008;67:41–49
11. Sugimoto M, Nakashima H, Watanabe S, et al. T-lymphocyte alveolitis in HTLV-I–associated myelopathy. Lancet 1987;2:1220
12. Nagai M, Usuku K, Matsumoto W, et al. Analysis of HTLV-I proviral load in 202 HAM/TSP patients and 243 asymptomatic HTLV-I carriers: High proviral load strongly predisposes to HAM/TSP. J Neurovirol 1998;4:586–93
13. Hayashi D, Kubota R, Takenouchi N, et al. Accumulation of human T-lymphotropic virus type I (HTLV-I)–infected cells in the cerebrospinal fluid during the exacerbation of HTLV-I–associated myelopathy. J Neurovirol 2008;14:459–63
14. Jacobson S, Gupta A, Mattson D, et al. Immunological studies in tropical spastic paraparesis. Ann Neurol 1990;27:149–56
15. Kubota R, Furukawa Y, Izumo S, et al. Degenerate specificity of HTLV-1–specific $CD8^+$ T cells during viral replication in patients with HTLV-1–associated myelopathy (HAM/TSP). Blood 2003;101:3074–81
16. Biddison WE, Kubota R, Kawanishi T, et al. Human T cell leukemia virus type I (HTLV-I)–specific $CD8^+$ CTL clones from patients with HTLV-I–associated neurologic disease secrete proinflammatory cytokines, chemokines, and matrix metalloproteinase. J Immunol 1997;159:2018–25
17. Kubota R, Kawanishi T, Matsubara H, et al. Demonstration of human T lymphotropic virus type I (HTLV-I) tax–specific $CD8^+$ lymphocytes directly in peripheral blood of HTLV-I–associated myelopathy/tropical spastic paraparesis patients by intracellular cytokine detection. J Immunol 1998;161:482–88
18. Greten TF, Slansky JE, Kubota R, et al. Direct visualization of antigen-specific T cells: HTLV-1 Tax11-19–specific CD8(+) T cells are activated in peripheral blood and accumulate in cerebrospinal fluid from HAM/TSP patients. Proc Natl Acad Sci USA 1998;95:7568–73
19. Levin MC, Lehky TJ, Flerlage AN, et al. Immunologic analysis of a spinal cord-biopsy specimen from a patient with human T-cell lymphotropic virus type I–associated neurologic disease. N Engl J Med 1997;336:839–45
20. Skinner PJ, Daniels MA, Schmidt CS, et al. Cutting edge: In situ tetramer staining of antigen-specific T cells in tissues. J Immunol 2000; 165:613–17
21. Ijichi S, Izumo S, Eiraku N, et al. An autoaggressive process against bystander tissues in HTLV-I–infected individuals: A possible pathomechanism of HAM/TSP. Med Hypoth 1993;41:542–47
22. Kozako T, Arima N, Toji S, et al. Reduced frequency, diversity, and function of human T cell leukemia virus type 1–specific $CD8^+$ T cell in adult T cell leukemia patients. J Immunol 2006;177:5718–26
23. Yashiki S, Fujiyoshi T, Arima N, et al. HLA-A*26, HLA-B*4002, HLA-B*4006, and HLA-B*4801 alleles predispose to adult T cell leukemia: The limited recognition of HTLV type 1 tax peptide anchor motifs and epitopes to generate anti-HTLV type 1 tax CD8(+) cytotoxic T lymphocytes. AIDS Res Hum Retroviruses 2001;17:1047–61
24. Bunce M, O'Neill CM, Barnardo MC, et al. Phototyping: Comprehensive DNA typing for HLA-A, B, C, DRB1, DRB3, DRB4, DRB5 & DQB1 by PCR with 144 primer mixes utilizing sequence-specific primers (PCR-SSP). Tissue Antigens 1995;46:355–67
25. Lee B, Tanaka Y, Tozawa H. Monoclonal antibody defining tax protein of human T-cell leukemia virus type-I. Tohoku J Exp Med 1989;157:1–11
26. Hayashi D, Kubota R, Takenouchi N, et al. Reduced Foxp3 expression with increased cytomegalovirus-specific CTL in HTLV-I–associated myelopathy. J Neuroimmunol 2008;200:115–24
27. Haring JS, Pewe LL, Perlman S. Bystander CD8 T cell–mediated demyelination after viral infection of the central nervous system. J Immunol 2002;169:1550–55
28. Fung-Leung WP, Kundig TM, Zinkernagel RM, et al. Immune response against lymphocytic choriomeningitis virus infection in mice without CD8 expression. J Exp Med 1991;174:1425–29
29. Marcondes MC, Burudi EM, Huitron-Resendiz S, et al. Highly activated CD8(+) T cells in the brain correlate with early central nervous system dysfunction in simian immunodeficiency virus infection. J Immunol 2001;167:5429–38
30. Kannagi M, Matsushita S, Shida H, et al. Cytotoxic T cell response and expression of the target antigen in HTLV-I infection. Leukemia 1994; 1(Suppl 8):S54–59
31. Hanon E, Asquith RE, Taylor GP, et al. High frequency of viral protein expression in human T cell lymphotropic virus type 1–infected peripheral blood mononuclear cells. AIDS Res Hum Retroviruses 2000; 16:1711–15
32. Nagasato K, Nakamura T, Ohishi K, et al. Active production of anti-human T-lymphotropic virus type I (HTLV-I) IgM antibody in HTLV-I–associated myelopathy. J Neuroimmunol 1991;32:105–9
33. Moritoyo T, Reinhart TA, Moritoyo H, et al. Human T-lymphotropic virus type I–associated myelopathy and tax gene expression in $CD4^+$ T lymphocytes. Ann Neurol 1996;40:84–90
34. Matsuoka E, Takenouchi N, Hashimoto K, et al. Perivascular T cells are infected with HTLV-I in the spinal cord lesions with HTLV-I–associated myelopathy/tropical spastic paraparesis: Double staining of immunohistochemistry and polymerase chain reaction in situ hybridization. Acta Neuropathol 1998;96:340–46
35. Seki M, Higashiyama Y, Mizokami A, et al. Up-regulation of human T lymphotropic virus type 1 (HTLV-1) tax/rex mRNA in infected lung tissues. Clin Exp Immunol 2000;120:488–98

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THE SPINE JOURNAL

Clinical Study

# Voluntary driven exoskeleton as a new tool for rehabilitation in chronic spinal cord injury: a pilot study

Mirko Aach, MD[a,*], Oliver Cruciger, MD[a], Matthias Sczesny-Kaiser, MD[b], Oliver Höffken, MD[b], Renate Ch. Meindl, MD[a], Martin Tegenthoff, MD[b], Peter Schwenkreis, MD[b], Yoshiyuki Sankai, PhD[c], Thomas A. Schildhauer, MD[d]

[a]*Department of Spinal Cord Injuries, BG University Hospital Bergmannsheil, Bürkle-de-la-Camp-Platz 1, 44797, Bochum, Germany*
[b]*Department of Neurology, BG University Hospital Bergmannsheil, Bürkle-de-la-Camp-Platz 1, 44797, Bochum, Germany*
[c]*Faculty of Engineering, Information and Systems, University of Tsukuba, 1-1-1 Tennodai, Tsukuba, Ibaraki 305-0006, Japan*
[d]*Department of General and Trauma Surgery, BG University Hospital Bergmannsheil, Bürkle-de-la-Camp-Platz 1, 44797, Bochum, Germany*

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**Abstract**

**BACKGROUND CONTEXT:** Treadmill training after traumatic spinal cord injury (SCI) has become an established therapy to improve walking capabilities. The hybrid assistive limb (HAL) exoskeleton has been developed to support motor function and is tailored to the patients' voluntary drive.

**PURPOSE:** To determine whether locomotor training with the exoskeleton HAL is safe and can increase functional mobility in chronic paraplegic patients after SCI.

**DESIGN:** A single case experimental A-B (pre-post) design study by repeated assessments of the same patients. The subjects performed 90 days (five times per week) of HAL exoskeleton body weight supported treadmill training with variable gait speed and body weight support.

**PATIENT SAMPLE:** Eight patients with chronic SCI classified by the American Spinal Injury Association (ASIA) Impairment Scale (AIS) consisting of ASIA A (zones of partial preservation [ZPP] L3–S1), n=4; ASIA B (with motor ZPP L3–S1), n=1; and ASIA C/D, n=3, who received full rehabilitation in the acute and subacute phases of SCI.

**OUTCOME MEASURES:** Functional measures included treadmill-associated walking distance, speed, and time, with additional analysis of functional improvements using the 10-m walk test (10MWT), timed-up and go test (TUG test), 6-minute walk test (6MWT), and the walking index for SCI II (WISCI II) score. Secondary physiologic measures including the AIS with the lower extremity motor score (LEMS), the spinal spasticity (Ashworth scale), and the lower extremity circumferences.

FDA device/drug status: Not applicable.

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The disclosure key can be found on the Table of Contents and at www.TheSpineJournalOnline.com.

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Authors' contributions: MA and OC carried out the experiments and data analysis as well as drafting of the manuscript. RCM, PS and MT helped with the experimental set up. MS-K and OH contributed to the data analysis. TAS participated in study design and coordination of the study.

All authors read and approved the final manuscript.

YS is a founder, shareholder, and the CEO of Cyberdyne, Inc., which produces the HAL.

YS and Cyberdyne were neither involved in study funding, design, data collection, and analysis, nor in writing or submitting this article, therefore concluding in no specific influence on the trial. We certify that no party having a direct interest in the results of the research supporting this article has or will confer a benefit on us or on any organization with which we are associated.

YS and Cyberdyne as the manufacturer of the device provided exclusively technical and advisory support.

YS as the CEO of Cyberdyne has been involved exclusively in terms of an advisory capacity, regarding technical support and the limitations of the exoskeleton. Therefore, the inclusion and exclusion criteria have been modified (eg, body weight and contractures).

* Corresponding author. Department of Spinal Cord Injuries, BG University Hospital Bergmannsheil, Bürkle-de-la-Camp-Platz 1/44789, Bochum, Germany. Tel.: 81 29-853-2111.

*E-mail address*: bergmannsheil@bergmannsheil.de (M. Aach)


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**METHODS:** Subjects performed standardized functional testing before and after the 90 days of intervention.
**RESULTS:** Highly significant improvements of HAL-associated walking time, distance, and speed were noticed. Furthermore, significant improvements have been especially shown in the functional abilities without the exoskeleton for over-ground walking obtained in the 6MWT, TUG test, and the 10MWT, including an increase in the WISCI II score of three patients. Muscle strength (LEMS) increased in all patients accompanied by a gain of the lower limb circumferences. A conversion in the AIS was ascertained in one patient (ASIA B to ASIA C). One patient reported a decrease of spinal spasticity.
**CONCLUSIONS:** Hybrid assistive limb exoskeleton training results in improved over-ground walking and leads to the assumption of a beneficial effect on ambulatory mobility. However, evaluation in larger clinical trials is required. 

*Keywords:* Exoskeleton; Treadmill training; Rehabilitation; Paraplegia; Hybrid assistive limb; Spinal cord injury

## Introduction

About 1,200 people suffer a traumatic spinal cord injury (SCI) each year in Germany. Recent statistics indicate that more than 50% of these injured patients have a motor incomplete lesion [1]. In patients with initial motor incomplete SCI, at least 75% regain some kind of ambulatory function. Better functional outcome is associated with age, level of lesion, and the classification in the American Spinal Injury Association (ASIA) Impairment Scale [2]. In the first 2 months after initial SCI, approximately half of the recovery occurs. Within the following 4 months, a decreasing rate of recovery has been observed. One year after injury, neurologic recovery is assumed to be nearly complete [3]. Although conventional rehabilitation programs enhance the performance of functional tasks, the loss of strength and coordination substantially limit one's capacity for over-ground ambulation training [4]. In the past two decades, body weight supported treadmill training (BWSTT) has been proposed as a useful adjunct to enhance locomotor function after motor incomplete SCI [5]. In patients with incomplete or complete SCI, a bilateral leg muscle activation combined with coordinated stepping movements can be induced in partially unloaded patients, standing on a moving treadmill. Body weight supported treadmill training enables early initiation of gait training and integration of weight-bearing activities, stepping and balance, by the use of a task-specific approach and a systematic gait pattern [6]. To facilitate the delivery of BWSTT in SCI patients, the locomotor training evolved over the last 12 years and a motorized robotic driven gait orthosis (DGO) has been developed [7]. The advantages over conventional BWSTT methods are considered to be less effort for attending physiotherapists [8], longer duration, more physiologic and reproducible gait patterns, and the possibility to measure a patients' performance. Several studies pointed out that DGO training improves over-ground walking [9–13]. However, there was no reported difference in the outcome of DGO training compared with conventional training. A significant switch in the ASIA classification has not been found [10,14].

Over the last 5 years, exoskeletal systems became available for SCI patients. These systems offer different possibilities. Three exoskeletons (Ekso [EksoBionics, Richmond, CA, USA], Rex [Rex Bionics, Aukland, New Zealand] and Re-Walk [ARGO Medical Technolgies, Israel]) allow SCI patients to stand up, walk with a defined pattern, and even climbing stairs mainly on a basis of passive range of motion (ROM). The exoskeleton hybrid assistive limb (HAL; Cyberdyne, Inc., Japan) offers the possibility of getting connected with the SCI patient through electromyography electrodes on the skin at the extensor/flexor muscle region of the lower extremities. This allows voluntary machine supported ROM of incomplete SCI patients by using minimal bioelectrical signals, recorded and amplified from hip and knee flexors and extensors [15–17]. More recently, these various exoskeletal systems allow the patients mobilization outside the treadmill. A former study by Kawamoto et al. [18] concerning locomotion improvement using HAL in chronic stroke patients, emphasized the feasibility for rehabilitation of these particular patients.

The aim of this pilot study was to evaluate the possibilities of exoskeletal locomotor training (HAL; Cyberdyne, Inc.) under voluntary control and identify beneficial effects on functional mobility of the patients. The hypothesis was that exoskeleton treadmill training is feasible and safe in application and capable of improving ambulatory mobility in chronic SCI patients.

## Materials and methods

### *Patients*

We enrolled eight patients (two women, six men). The mean±standard deviation age at the time of enrollment was 48±9.43 years. All patients were in the chronic stage of traumatic SCI according to the time since injury of 1 to 19 years (97.2±88.4 months). Inclusion criteria were traumatic SCI with chronic incomplete (ASIA B/C/D) or complete paraplegia (ASIA A) after lesions of the conus medullaris/cauda equine with zones of partial preservation. Independent of ASIA classification, the enrolled patients

must present motor functions of hip and knee extensor and flexor muscle groups to be able to trigger the exoskeleton. Exclusion criteria were as follows: nontraumatic SCI, pressure sores, severe limitation of ROM regarding hip and knee joints, cognitive impairment, body weight more than 100 Kg, nonconsolidated fractures, and mild or severe heart insufficiency. Two patients suffered from an incomplete thoracic SCI (ASIA C/D) from 3 to 13 years. Two patients suffered from an incomplete lumbar SCI (ASIA B/C) from 12 to 13 months and four patients had a complete SCI with zones of partial preservation in L3–S1 after lesions of the conus medullaris. The classification according to the ASIA was carried out before the treadmill training was initiated. The study was approved by Ethical Board Committee of Bergmannsheil Hospital and the University of Bochum and followed strictly the declaration of Helsinki.

All patients provided written informed consent. The study design was a single case experimental A-B (pre-post) design by repeated assessments of the same patients (Table 1).

*Intervention*

During this study, the patients underwent a BWSTT five times per week using the HAL exoskeleton (Cyberdyne, Inc., Japan). The study was performed between June 2013 and September 2013 in the BG University Hospital Bergmannsheil, Bochum.

Neither adverse nor severe adverse events occurred during the intervention.

*The exoskeleton*

The HAL robot suit (Cyberdyne, Inc., Japan) is an exoskeleton with a frame and robotic actuators that attach to the patients' legs. The joint movement is supported by electric motors. Voluntary initiated minimal bioelectrical signals recovered from extensor and flexor muscles of hip and knee are detected via electromyography electrodes (Fig. 1).

Through a cable connection between the exoskeleton and patient, this system allows voluntary robotic supported ROM (cybernic voluntary control mode). Also a passive, nonvoluntary ROM (cybernic autonomous control mode) is possible (Fig. 2).

*The treadmill*

The treadmill system (Woodway USA, Inc., Waukesha, WI, USA) includes a body weight support system with a harness. The speed can be adjusted from 0 Km/h to approximately 4.5 Km/h. During treatments, the velocity of the treadmill was set individually between comfortable and maximum speed tolerated by the patients. Approximately 50% of each patient's body weight needed to be supported by the harness system, individually reduced during the following sessions as tolerated without substantial knee buckling or toe drag.

EVIDENCE & METHODS

**Context**
The authors present a series of patients treated with an assistive exoskeleton developed to facilitate treadmill exercise in patients with spinal cord injury (SCI).

**Contribution**
In a series of eight patients with SCI graded ASIA A to C/D, improvements in walking time, distance and speed were noted after treatment with assistive exoskeleton.

**Implications**
This study is a case series of eight patients with heterogeneous clinical characteristics, including the severity of their spinal cord injury. The findings are limited to clinical contexts specific to these patients and clearly cannot be translated to the care of other individuals. This is simply a report that may show proof of concept. It should be noted that one of the authors reports a substantive conflict of interest (founder and shareholder of the company that produces the exoskeleton device).

*—The Editors*

*The training*

The patients underwent a 90-day period of HAL exoskeleton (Cyberdyne, Inc.) training (five per week), including a mean number of sessions of 51.75±5.6. The training was performed on a treadmill with individually adjustable body weight support and speed, recording walking speed, time, and distance. It included a 10-m walk test (10MWT) before and after each session and regular physiotherapy that lasted approximately 90 minutes. The training was supervised by a physiotherapist and a medical doctor.

*Measurements*

*Walking capabilities and neurologic status*

All patients were assessed on admission by medical doctors involved in this trial. The outcomes were assessed

Table 1
Subject demographics and clinical characteristics

| Case | Sex | Age (y) | Time since trauma, y | Etiology | Level | ASIA/ZPP | WISCI II | Ashworth |
|---|---|---|---|---|---|---|---|---|
| 1 | M | 40 | 13 | # T7/T8 | T8 | C | 13 | 4 |
| 2 | M | 63 | 1 | # T12 | L1 | B/L3 | 6 | 0 |
| 3 | M | 36 | 1.16 | # T11/T12 | T12 | A/L3 | 6 | 0 |
| 4 | F | 55 | 1.08 | # L1 | L1 | C | 13 | 0 |
| 5 | M | 42 | 16 | # L1 | L1 | A/L3 | 9 | 0 |
| 6 | M | 52 | 10 | # L3 | L2 | A/L3 | 6 | 0 |
| 7 | F | 40 | 19 | # L1 | T11 | A/S1 | 9 | 0 |
| 8 | M | 53 | 3 | # T12 | T12 | D | 18 | 0 |

M, male; F, female; #, fracture; ASIA, American Spinal Injury Association; ZPP, zones of partial preservation; WISCI, walking index for spinal cord injury; T, thoracic; L, lumbal; S, sacral.

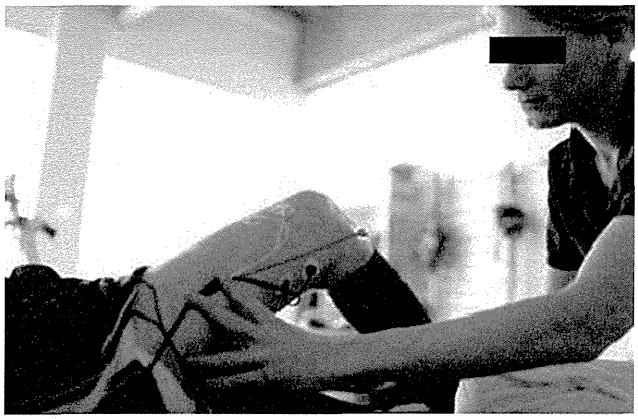

Fig. 1. Positioning of the electromyography electrodes on the knee extensor and flexor muscles.

by physiotherapists neither involved in the study design nor analysis after 45 days and on discharge from the training period. An assessment through the ASIA classification was already done on admission and on discharge from the SCI department, Bergmannsheil, Bochum, within the initial therapy after acute SCI. The 10MWT, done before and after each session, detected the needed time, the number of steps, and the required assistance to walk a 10 m distance [19,20]. The timed-up and go test (TUG test) describes the time and assistance required for standing up from the wheelchair, walk 3 m, turn around, walk back, and sit down. It was performed every 2 weeks. The 6-minute walk test (6MWT) was done at the beginning, at half time, and at the end if possible, depending on the patient. It evaluates the distance and assistance while walking for 6 minutes [21]. The main outcome was the functional motor assessment by the walking index for SCI II (WISCI II) [22,23]. The WISCI II score is a 20-item scale, measuring the walking capabilities of a patient based on the requirements of assistance because of walking aids, personal assistance, or braces. Grade 0 means that the patient has neither standing nor walking abilities. Grade 20 means that no assistance is needed to walk a distance of 10 m. The neurologic status was assessed using the ASIA Impairment Scale modified from the Frankel classification and classifies motor and sensory impairments that result from a SCI [3]. The lower extremity motor score (LEMS) acquired in this study was obtained by the addition of the impairment scores (0–5) of the lower extremity key muscles of both sides. Muscle volume was assessed by manual measurements, 20/10 cm above and 15 cm below inner knee gap.

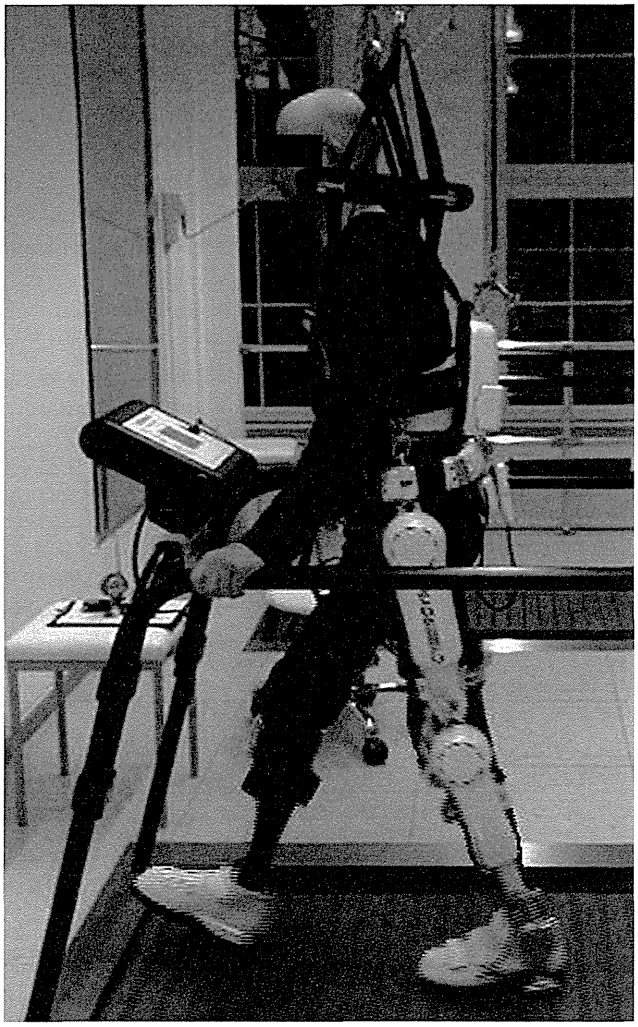

Fig. 2. Patient performing treadmill locomotion training with body weight support and hybrid assistive limb exoskeleton.

### *Statistical analysis*

Descriptive analysis of the demographic and injury characteristics was done using frequency distribution for categorical data and mean for continuous variables. Differences between pre- and posttraining sessions were assessed by a paired *t* test (for continuous variables). Treatment effects on functional performance as the WISCI II are all ordinal scales. Medians were used as descriptive statistics for these outcomes, and nonparametric tests were used to assess the relative effect of the treatments.

Results were considered statistically significant when the p value was $\leq .05$.

## Results

### *Treadmill associated results*

All patients improved in treadmill training by using HAL (Cyberdyne, Inc., Japan). The mean walking speed increased from $0.91 \pm 0.41$ m/s (0.5–1.8 m/s) in the first session up to $1.59 \pm 0.5$ m/s (0.8–2.1 m/s) in the last session after 3 months. The progress in speed after 6 weeks of training was lower than in the first weeks. The range was located between 0 km/h and 0.8 km/h. The mean walking time at the beginning was $12.37 \pm 4.55$ minutes. The average walking time at the

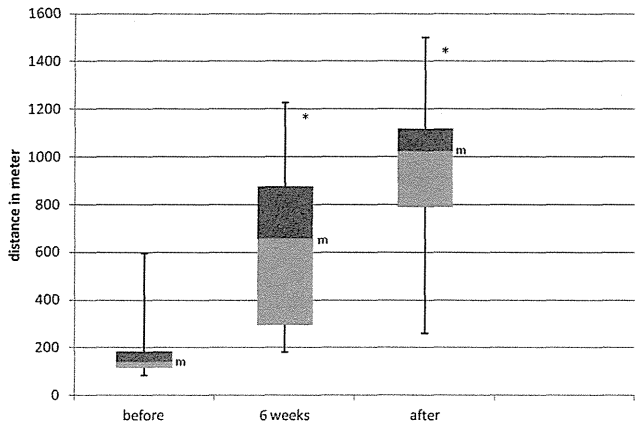


Fig. 3. Changes in treadmill-associated walking distance in pre-, mid-, and postevaluations. m, median. *pre-post difference, p<.05.

end was 31.97±9.45 minutes. The mean ambulated distance at the first session was 195.9±166.7 m and increased to 954.13±380.4 m on discharge (Fig. 3).

*Functional outcome*

Although the mean improvement concerning the WISCI II score was not statistically significant, three patients showed functional improvement in gait abilities. Two subjects needed braces, a walker, and support by a physiotherapist at the beginning and were able to walk after the training series only with a walker and braces (WISCI II score increased from 6 to 9). One patient increased from 9 to 12 and, therefore, was able to walk with two crutches and a brace compared with a walker and a brace before the training. At baseline, the mean WISCI II score was 10±4.3. At the end of the 90 days trial, the mean WISCI II was 11.13±6.68. Improvements in speed and endurance in over-ground gait assessments in all participants have been achieved. The 10MWT showed a significant increase in mean gait speed at the end of the training period compared with baseline (0.28±0.28 m/s vs. 0.50±0.34 m/s) (Fig. 4).

The improvement corresponded to a 44% faster walking than in initial evaluation. It also includes the reduction of

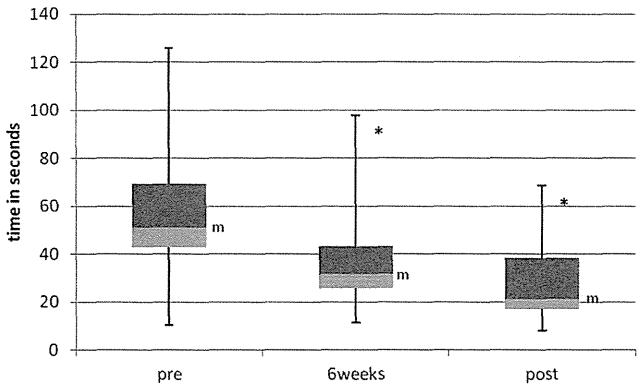


Fig. 4. Changes in 10-m walk test in pre-, mid-, and postintervention evaluations. m, median. *pre-post difference, p<.05.

Table 2
Comparison of pre- and postinterventions

| Outcome measurements | Before training | After training | n |
|---|---|---|---|
| 10MWT speed (m/s) | 0.28±0.28 | 0.5±0.34* | 8 |
| Number of steps | 29.88±7.85 | 19.38±3.16* | 8 |
| 6MWT distance (m) | 70.1±130 | 163.3±160.6* | 8 |
| TUG test (s) | 55.34±32.20 | 38.18±25.98* | 8 |
| Distance (m) | 195.88±166.71 | 954.13±380.35* | 8 |
| WISCI-II | 10±4.34 | 11.12±3.68 | 8 |

10MWT, 10-m walk test; 6MWT, 6-minute walk test; TUG, timed-up and go; WISCI, walking index for spinal cord injury.
Note: Values are means±standard deviation.
* Pre-post difference, p<.05.

support needed detected by the WISCI II score. The mean number of steps decreased from 29.8±7.85 to 19.4±3.16. We observed significant increase in gait speed from pre- to midtraining and from mid- to posttraining assessments. Similar results were detected for the TUG test. The mean time needed for the TUG test decreased from 55.34±32.2 seconds to 38.18±25.98 seconds. The 6MWT was done with a constant walking time of 6 minutes without any break. Only three patients were able to perform the 6MWT before the training with a mean walking distance of 187±162.2 m. The subjects in this subgroup improved their performance and increased the walking distance to 287.3±229.4 m. After completing the training, all eight patients could be evaluated, therefore the overall mean distance increased from 70.1±130 m to 163.3±160.6 m (Table 2).

The LEMS increased in all patients. The mean LEMS before the training increased significantly from 21.75±8.3 to 24.38±7.6 after the intervention (Fig. 5).

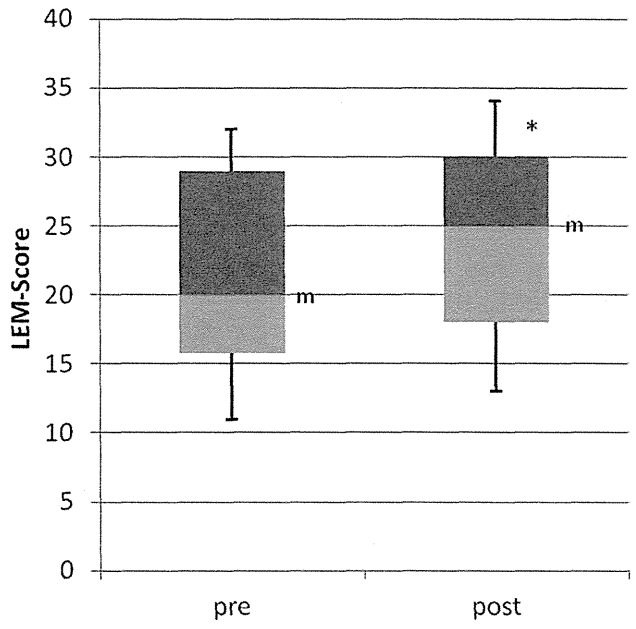


Fig. 5. LEMS in pre- and postevaluation. LEM, lower extremity motor score; m, median. *pre-post difference, p<.05.

One patient switched in the ASIA scale from ASIA B to C, he was at the beginning of the training 12 months posttrauma.

*Others*

To describe muscle volume, measurements of the circumferences 10/20 cm cranial of the inner knee joint gap and 15 cm distal of it have been done before and after the 90 days of training. Seven participants showed a gain of muscle circumference from 5 mm up to 50 mm. In one participant with edema in his lower legs, we observed a loss of circumference up to 25 mm. One patient suffering from a thoracic SCI presented a significant spinal spasticity. For spastic motor behaviors, we used the modified Ashworth scale to evaluate the involuntary resistance to passive stretch of the quadriceps muscle group. At pretraining evaluation, he showed an extensor spasm with high resistance to passive stretch according to Ashworth 4. After the training sessions, the resistance was reduced according to Ashworth 2. This level lasted for about 6 to 8 hours with a new maximum level at the next morning. All other patients showed no spastic motor behaviors.

## Discussion

The objective of the study was to determine whether locomotor training with the exoskeleton HAL is feasible and safe in application, improves functional mobility, and increases motor functions in chronic paraplegic patients after SCI. The results obtained revealed a highly significant improvement for over-ground walking abilities evaluated by the 10MWT, the 6MWT, and the TUG test and the partial reduction of physical assistance and walking aids in the WISCI II score. Muscle strength, measured with the LEMS increased in all patients.

The results acquired in this clinical trial imply that HAL-supported locomotion training can improve walking abilities in terms of speed, gait, and distance. Furthermore, it improves motor functions.

Thus far there is insufficient evidence and only a few articles addressing the main hypothesis of this study that locomotor training improves walking function for patients with SCI [24].

The present study is according to the knowledge of the authors the first to investigate the impact of HAL-supported locomotor training in chronic SCI patients, where referring to the current state of knowledge no further functional improvements are to be expected.

In the subject population consisting of eight patients including patients suffering from SCI from 1 to 19 years (8.03±7.4 years), all patients improved significantly regarding treadmill-associated walking distance and speed and functional improvement was detected in the over-ground walking tests.

Although no significant influence was seen on the requirements of assistance in the 10MWT, three patients attained improvement in walking abilities according to the WISCI II, under condition of a comfortable and stable gait. A further reduction of assistance was not forced because of more pathologic gait or higher risk of falling [24].

Although the evidence is still insufficient, the effectiveness of automated locomotor training using the DGO in patients with chronic SCI is being investigated and considered promising in several systematic reviews including a Cochrane review [25,26]. The results mentioned previously add to the wealth of that data presuming that HAL-assisted locomotion training is useful in terms of functional mobility and a safe adjunct to the treatment of patient with chronic SCI.

Our study had several limitations: the relatively small number of patients (n=8) and the mixture of complete and incomplete SCIs.

However, all the patients were treated in the same facility by the same multidisciplinary team, according to a standardized protocol.

In summary, our study provides the first data demonstrating the clinical potential of HAL-locomotor training based on voluntary drive in patients suffering from chronic SCI.

It was proven to be a safe device for locomotion therapy as neither adverse nor severe adverse events occurred.

However, continued research in the form of large randomized trials to compare the efficacy of HAL-assisted training with well established, conventional therapies is necessary.

## References

[1] Eng J, Teasell R, Miller W, et al. Spinal cord injury rehabilitation evidence: methods of the SCIRE systematic review. Top Spinal Cord Inj Rehabil 2007;13:1–10.

[2] Waters RL, Adkins R, Yakura J, Vigil D. Prediction of ambulatory performance based on motor scores derived from standards of the American Spinal Injury Association. Arch Phys Med Rehabil 1994;75:756–60.

[3] Piepmeier JM, Jenkins NR. Late neurological changes following traumatic spinal cord injury. J Neurosurg 1988;69:399–402.

[4] Gittler MS, McKinley WO, Stiens SA, et al. Spinal cord injury medicine. 3. Rehabilitation outcomes. Arch Phys Med Rehabil 2002;83:S65–71.

[5] Finch L, Barbeau H, Arsenault B. Influence of body weight support on normal human gait: development of a gait retraining strategy. Phys Ther 1991;71:842–55; discussion 855–6.

[6] Dietz V, Wirz M, Curt A, Colombo G. Locomotor pattern in paraplegic patients: training effects and recovery of spinal cord function. Spinal Cord 1998;36:380–90.

[7] Colombo G, Joerg M, Schreier R, Dietz V. Treadmill training of paraplegic patients with a robotic orthosis. J Rehabil Res Dev 2000;37:693–700.

[8] Winchester P, Querry R. Robotic orthosis for body weight-supported treadmill training. Phys Med Rehabil Clin N Am 2006;17:159–72.

[9] Colombo G, Wirz M, Dietz V. Driven gait orthosis for improvement of locomotor training in paraplegic patients. Spinal Cord 2001;39:252–5.

[10] Wirz M, Zemon DH, Rupp R, et al. Effectiveness of automated locomotor training in patients with chronic incomplete spinal cord