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Maastricht University

Faculty of Medicine and Life Sciences
School for Life Sciences
Master of Biomedical Sciences

Masterthesis

Clinical profiling, manifestation and assessment of walking-related performance fatigability in persons with Multiple Sclerosis: a cross-sectional study

Hanne Bielen

Thesis presented in fulfillment of the requirements for the degree of Master of Biomedical Sciences, specialization Clinical Molecular Sciences

SUPERVISOR :

Prof. dr. Peter FEYS

MENTOR :

Mevrouw Fanny VAN GEEL

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Transnational University Limburg is a unique collaboration of two universities in two countries: the University of Hasselt and Maastricht University.

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Acknowledgement

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Table of contents

Abbreviations

6MWD	Six-minute walking distance
6MWT	Six-minute walking test
DWI_{6-1}	Distance walked index
EDSS	Expanded disability status scale
EDSS	Expanded disability system score
FES-I	Falls efficacy scale international
FSMC	Fatigue scale for motor and cognitive functions
FS	Functional system
FSS	Fatigue severity scale
HADS	Hospital anxiety and depression scale
HC	Healthy controls
ICC	Intra class correlation coefficient
IQR	Interquartile range
MAS	Modified ashworth scale
MFIS	Modified fatigue impact scale
MI	Motricity index
MS	Multiple Sclerosis
MSWS-12	Multiple Sclerosis walking scale
NHPT	Nine hole peg test
NWF	Non-walking fatigability
PASAT	Paced auditory serial addition test
PwMS	Persons with Multiple Sclerosis
RTI_{6-1}	Reaction time index
SCI	Sleep condition indicator
SD	Standard deviation
SDMT	Symbol digit modality test
SE	Standard Error
SEM	Standard error of measurement
SI	Symptom inventory
T25FW	Timed 25 foot walk
VAS	Visual analogue scale
WF	Walking fatigability

Abstract

Introduction: Fatigue and walking difficulties are the most common symptoms in persons with Multiple Sclerosis (PwMS). Previous research recommended the six-minute walking test (6MWT) as an objective assessment for walking fatigability (WF) by calculating differences in walking distance between the first and last minute. Our research group proposed a Distance Walk Index (DWI_{6-1}), with a decline of ≥15% as discriminative threshold. However, no control group was used to define this cut-off value and no psychometric properties were reported. Moreover, the manifestation of WF by other symptoms was still not well understood.

Materials and methods: 49 PwMS (EDSS 0-6) and 28 healthy controls (HC) performed a 6MWT twice (3-5 days in between). Short objective screening for spasticity, balance and strength was performed before, immediately after, and after 10, 20 and 30 minutes of the 6MWT. The severity of 11 common MS symptoms was rated on a 0-10 scale at the same time slots.

Results: By including a control group, it was shown that a $DWI_{6-1} \leq -10\%$ was already sufficient to state WF. Reliability analyses of the DWI_{6-1} indicated an intra class correlation coefficient of 0.76 and 0.60 for the total MS group and HC respectively. Only the last minutes of the 6MWT showed significant differences between the groups in walking distance decline compared to minute 1. Half of the pwMS showed WF, where it significantly manifested in greater subjective increases in gait impairments, spasticity and dizziness compared to the non-walking fatigability (NWF) group. Most symptoms returned to baseline 10 minutes after the 6MWT. At baseline, significantly more muscle weakness and gait impairment were reported in the WF group. Objectively, more balance problems and a lower muscle strength were significantly observed at baseline.

Discussion and conclusions: The DWI during the 6MWT of PwMS showed a good test-retest reliability in the total MS group. and is therefore a reliable measurement for WF. Long walking tests are recommended to differentiate between WF and non-walking fatigability (NWF) in terms of performance and perceived fatigability. Half of the MS patients showed WF, where it significantly manifested in gait impairments and muscle weakness. Further research is recommended to further investigate the clinical profile and underlying factors of WF with a larger sample size and better objective testing.

1. Introduction

Hundreds of millions of people worldwide suffer from a neurological disorder. **Multiple Sclerosis** (MS) is one of the most common ones, affecting approximately 2.3 million people worldwide in 2013 (1, 2). Moreover, MS is frequently diagnosed at a young age (25-32 years) and in 2013 the prevalence in Europe was 108 per 100 000 (2). It is an inflammatory auto-immune disease affecting the central nervous system. Auto-reactive immune cells attack the myelin sheath around the nerves, causing an impairment of the signal transduction of the nerve system throughout the body. Hereby, MS can lead to various symptoms, affecting different parts of the body with varying severities (2, 3).

Since there is currently no cure for MS, treatment mainly focusses on stabilizing the disease and diminishing the symptoms. A symptom that is often reported in many neurological conditions is **fatigue**. Pathological fatigue is for example prevalent in stroke patients, parkinson's disease, traumatic brain injuries,… and is characterized as one of the most common and first symptoms in persons with MS (pwMS). Even 40% of the pwMS indicate fatigue as their most disabling symptom (4).

1.1 Taxonomy of fatigue

Since the etiology and terminology of fatigue remains poorly understood and currently no effective treatment exists, there is a need for better conceptualization and shared frameworks. Throughout literature, many different terms and definitions have been used to describe fatigue, which makes this research field quite confusing. To acquire clear communication and scientific progress, our research group introduces a more profound **conceptualization of fatigue** based on the integration of recent taxonomy propositions (4-7). Firstly, fatigue can be divided into two main domains: **trait fatigue and state fatigue** (Figure 1). Trait fatigue refers to a general feeling of fatigue that is always present in an individual. It is therefore more a characteristic and does not fluctuate over time. State fatigue on the other hand, also known as fatigability, is a form of fatigue that changes according to tasks and circumstances. Trait fatigue and fatigability can be both subdivided into a **cognitive and motor domain**. In **fatigability,** both domains have a **perceived and a performance component** (Figure 2). Perceived fatigability is a subjective patient-reported change in physical and/or cognitive sensations of fatigue during and/or after activity, while performance fatigability is an objectively measured change in physical and/or cognitive parameters during and/or after activity.

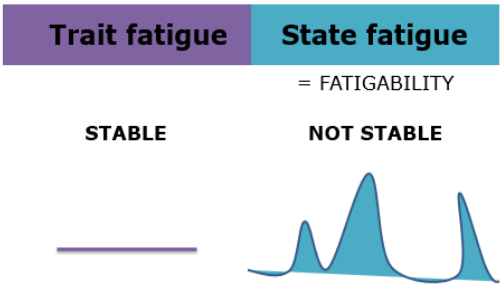

Figure 1: The two main domains of fatigue; trait fatigue and state fatigue.

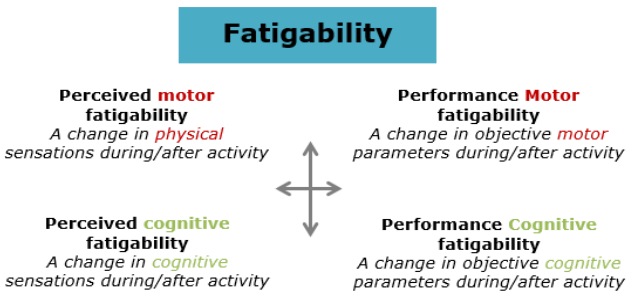

Figure 2: The motor and cognitive subdomains of fatigability; having both a perceived and performance. component.

1.2 Walking-related performance fatigability

Additionally, many pwMS experience **walking difficulties**, which together with fatigability, result in a decreased walking capacity over time (4, 6). This will have an impact on their daily life, both on activity and participation level, thereby reducing their quality of life (4, 6). Even though it is know that walking is one of the most important bodily functions, since it is an important factor contributing to the independence and ambulatory functions, only a few studies investigated the manifestation and assessment of **walking fatigability (WF)** (3, 5, 6, 8). Some studies reported for example a decrease in walking speed and distance, or other changes in spatiotemporal and kinematic factors during or after long walking tests (8-10). Moreover, The clinical manifestation other than gait changes over time are not well documented; For example WF may also manifest itself by changes in balance or coordination, or a decrease in attention thereby reducing the cognitive control of movement (11).

1.2.1 Assessment of walking-related performance fatigability

In order to further investigate the phenomenon of WF, it is in the first place of great importance to **objectively distinguish between patients with and without WF**. Nowadays, the assessment of fatigue and fatigability in pwMS is very subjective, since it mostly depends on self-reported questionnaires (e.g. the fatigue severity scale, multiple sclerosis walking scale) and scales (e.g. BORG scale), measuring trait fatigue and the perceived component of fatigability respectively, but not the performance component (5, 12).

Our research group conducted a **systematic review** (submission planned in June 2018) summarizing the objective measurement methods to assess walking-related performance fatigability in both healthy and diseased populations. The fact that WF is being investigated in such a wide range of pathologies and elderly, suggests its clinical importance. The aim of this review was to provide an overview of the methods currently used in clinical practice in order to conclude on an objective test and criteria to measure performance WF. The main findings were that in different populations similar test and formulas are used to measure WF, being mostly long walking test (e.g. six-minute walking test, 500m-walk) wherein comparisons are made between the beginning and the end of the test. Shorter walking tests (e.g. 10-meter walk, 2-minutes walking test) seemed not be sufficient enough to induce a clear deceleration (13). Longer walking tests on the other hand (e.g. ten-minute walking test) would be too hard and give bias towards drop-out (14). Besides, long walking test of a fixed number of minutes have shown to be more useful than walking tests with a fixed distance, since Schwid et al. (1999) indicated 60% of pwMS were not able to walk for 500m (15). However, discriminative data or cut-off values in any walking test are still scarce, thereby limiting its appliciation in experimental trials. In literature, also discussions arose about the use of treadmill walking or self-paced walking test for measuring WF. According to Schnelle et al. (2012), self-selected paced walking test using fatigability outcomes wherein the distance decline is normalized would be as standardized as treadmill walking (16). Moreover, treadmill walking would be more unsafe and not representative to daily live situations. Additionally, preferences towards self-paced walking test were made, as it can measure differences at spatiotemporal level, which is not the case in treadmill walking.

In general, it could be concluded that the **six-minute walking test (6MWT)** is recommended as objective assessment method to measure walking fatigability, by making comparisons between the first and the last minute of the walking task. This confirmed previous research of our research group, wherein a **Distance Walk Index (DWI)** was proposed with a walking distance decline of ≥15% as discriminative threshold to state WF (8). Based on this cut-off value, the prevalence of WF showed significant differences between low (1-4) and high (4.5-6.5) EDSS (Figure 3). However, no control group was used to define the cut-off score of -15% and no psychometric properties were reported. Moreover, the manifestation of WF by other symptoms was still not well understood.

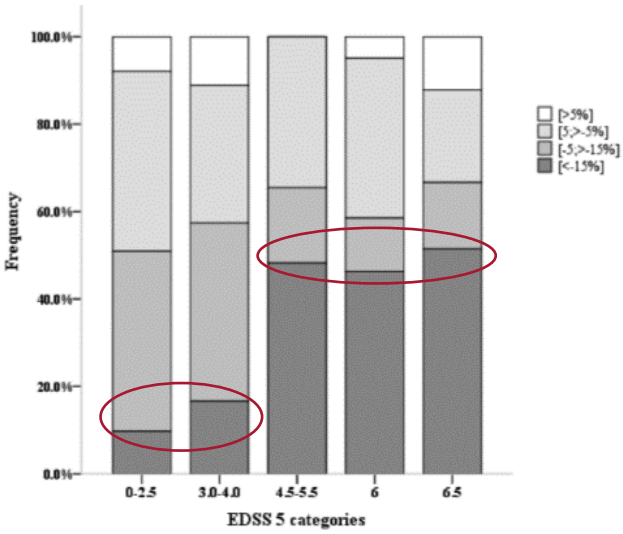

Figure 3: Frequency of different walking declines in five EDSS categories. Walking declines based on DWI_{6-1}: [(distance min1 – distance min6)/distance min1]x100. EDSS: expanded disability system score, DWI: distance walked index. (Leone et al., 2015)

1.2.2 Manifestation of walking-related performance fatigability

Additionally, there is a lack of research on the **underlying central and peripheral causes** of WF, together with its other **related and influencing factors**. Therefore, based on current literature (6, 7, 17, 18), our research group proposes the following taxonomy in terms of related and causal factors of WF (Figure 4).

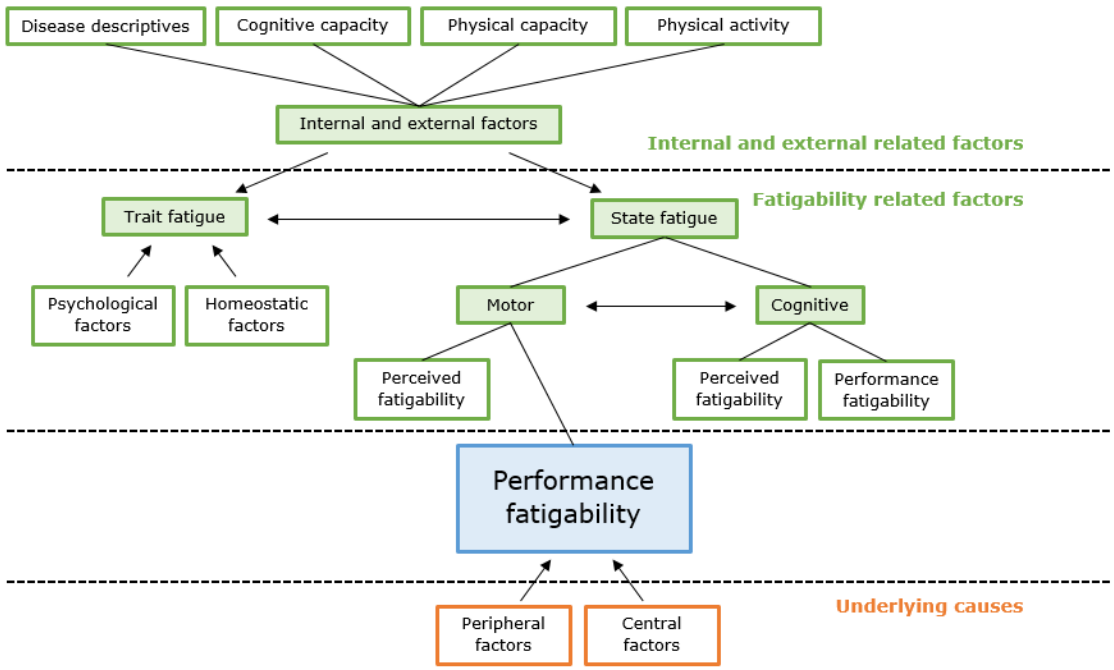

Figure 4: Schematic overview of the proposed fatigue taxonomy. Focus on performance motor fatigability, with its underlying causes and related factors.

Trait fatigue depends on the patient's psychological state and the physiological ability of their body to maintain homeostasis. **Central and peripheral impairments** are indicated as the contributors to performance motor fatigability (i.e. WF). Central factors could include neural integrity, voluntary central drive, conduction velocity and spinal motor excitability, while peripheral factors include muscle structures and contractile properties, as well as oxidative capacity and metabolite production (6, 7, 17, 18).

Figure 4 already gives an overview of the possible domains involved in the phenomenon of WF. This study will be a first screening in order to see what **symptoms and factors** seem to be **related to WF** and give a first indications towards a **clinical profile**, before going into more detailed objective testing. Literature states a strength decline as the most common indicator for motor fatigability, but it remains unclear whether other **manifestations** occur (4, 12, 19). Screening in this study will be based on subjective reporting of symptoms present in pwMS with and without WF. Muscle strength and gait pattern impairments can be questioned as part of the peripheral factors underlying performance WF, while spasticity, dizziness and balance problems for example either are indications for a central cause of WF. To overcome in a minimal way the subjectivity of the symptom reporting, short screening test could be executed for spasticity, balance and muscle strength. By doing only easy short subjective and objective screenings, is possible to repeat these measures at several timepoints after the fatiguing walking task to have an indication about the duration of WF and its related symptoms. Executing longer and objectively better tests after the walking task is not feasible, since currently it is not known how long walking fatigability persists. Baseline cognitive and physical capacity, as part of the internal and external factors influencing WF, can be examined via short standardized cognitive and motor test. Trait fatigue, with its underlying factors homeostatic and physiologic factors (e.g. depression, sleep problems), can be examined through various fatigue and fatigue-related questionnaires. Literature already describes that spasticity, sleep, physical activity, balance, gait pattern and strength are linked to fatigue, but the influence of fatigue and fatigability on these symptoms during a fatiguing task is still unclear (20-22).

Besides, some researchers state that every form of **fatigue manifests in a decrease in both motor and cognitive functions** (6). As walking is an elementary coordination pattern, characterized by alternated movements of different pairs of limbs, coordination and walking does require a cognitive control, which may fade over time if **cognitive fatigability** occurs (23). Therefore, a pilot study was additionally set up, in which we aimed to investigate the possible involvement of cognitive fatigability in WF and vice versa. Only a few studies investigated this association, wherein they found that a maximal exhaustion test on a treadmill influences the cognitive domain of 'alertness', resulting in reaction times after walking (11). Consequently, it can be hypothesized that not only physical, but also cognitive adaptations occur during a fatiguing walking task. As alertness is reported to be the best indication for cognitive fatigability, an auditory vigilance task could be recorded during the 6MWT in order to measure cognitive and motor fatigability at the same time. This is a cognitive task wherein participants need to responds as fast as possible to certain target letters of the alphabet. Moreover, previous research in our research group reported a very low dual task cost of 3% to perform this cognitive task while walking.

Insights in the clinical manifestation and underlying or related factors of WF are needed in order to propose a good **rehabilitation strategy.** So far, rehabilitation interventions have been targeting central (e.g. coordination or attention training through dance training) or peripheral (e.g. muscle strength and endurance training) factors, but were never investigated on their potential to impact on walking fatigability and related factors.

1.3 Research aims

The primary goal of this study is to get more insights in the phenomenon of WF, especially by investigating psychometric properties of the assessment method and examining its manifestation. In this study, the 6MWT will be used as fatiguing task, where after the $DWI_{6\text{-}1}$ will be calculated to measure WF. The following research questions will be addressed:

Research question 1: Is the $DWI_{6\text{-}1} \leq -15\%$ a good and reliable assessment method and criterium to objectively measure performance WF in pwMS?

Aim: With this research question, we aim to investigate test-retest reliability of the assessment method, as well as evidence for a good cut-off value to state WF.

Hypothesis: We hypothesize that the use of the 6MWT with calculations of the $DWI_{6\text{-}1}$ is a good and reliable measure to state WF, as a previously conducted systematic review on the assessment of WF recommended the 6MWT for measuring performance WF. The $DWI_{6\text{-}1}$ as assessment method will show discriminant validity to differentiate between WF and non-walking fatigability (NWF). In addition, Leone et al. (2015) reported the use of the $DWI_{6\text{-}1} \leq -15\%$ as criterium for WF. However, the correctness of the cut-off value of -15% will be further investigated in this study, as in the study of Leone et al. (2015) no control group was used (8).

Objectives: -1- Report test-retest reliability of the assessment methods by performing the walking test two times to investigate if the 6MWT with $DWI_{6\text{-}1}$ is a good and reliable measure to objectively assess WF and can therefore be used as an experimental outcome measure. -2- Investigate if the use of the $DWI_{6\text{-}1} \leq -15\%$ is a good criterium to state WF by comparing the outcome with the performance of healthy controls (HC) in order to see if clear distinctions in walking decline can be made between groups.

Research question 2: Does WF manifest itself in other MS-related symptoms and how long do these symptoms manifest?

Aim: With this research question, we aim to get preliminary insights about the manifestation and clinical profile of WF, as well as the duration of WF and its related symptoms.

Hypothesis: We hypothesize that WF manifests itself in other MS-related symptoms, such as balance problems, muscle weakness and gait pattern impairments. The fatigability and its related symptoms will manifest longer in the WF group compared to the non-walking fatigability (NWF) group. Additionally, there will be a link between perceived and performance fatigability.

Objectives: -1- Investigate the walking distance decline in WF, NWF and HC minute per minute, as well as their perceived WF in order to examine differences between groups and associations between perceived and performance fatigability. -2- Examine the presence of other MS-related symptoms before and at multiple timepoints after the walking task, to have an indication about the manifestation and duration of WF by other symptoms. -3- Examine symptoms and functioning at baseline level to give a first indication of a clinical profile for WF.

Research question 3: Is there a relationship between motor and cognitive fatigability in pwMS?

Aim: Examine if pwMS showing WF, also show a decline in cognitive functioning at baseline or during a walking task.

Hypothesis: We hypothesize that there is an association between cognitive fatigability and walking fatigability and that cognitive functioning problems at baseline will be linked to presence of WF.

Objectives: -1- Performance of an alertness test at rest and during the performance of a walking task to examine the points of onset of possible cognitive and motor declines. -2- Examine baseline cognitive function to investigate correlations with WF.

2. Material and methods

A cross-sectional study was conducted with 52 pwMS and 30 HC, which all performed a 6MWT twice (3-5 days in between) to measure prevalence and test-retest reliability of WF. Possible symptoms were subjectively and objectively evaluated before and after the 6MWT, as well as every 10 minutes after the 6MWT until 30 minutes. This was done to have an indication about the manifestation and duration of WF in other symptoms. A third test session was set up as a pilot study, in which 30 pwMS and 15 HC agreed to participate, to investigate the relationship between cognitive and motor fatigability. During this third session, participants executed a cognitive auditory vigilance task for 6 minutes while seated and while walking.

2.1 Study population

Fifty-two PwMS and 30 age- and gender-matched HC got included on a voluntary basis. All participants were recruited and tested at the rehabilitation research institute of Hasselt University, National MS Center Melsbroek or Rehabilitation and MS Center Overpelt. The study complies with the Declaration of Helsinki. The ethical committees approved the protocol and written informed consent was obtained from all participating subjects. Adults until the age of 70, diagnosed with MS according to the Mc Donald criteria (24) and an Expanded Disability Status Scale (EDSS) ranging from [0-6] were included. Patients who met this inclusion criteria underwent further screening; They should be able to walk independently or with unilateral support (i.e. walking stick) for six minutes without rest and should have performed a 6MWT before to ensure familiarization. Participants were excluded in case of a MS relapse within the past three months or when suffering from any other condition possibly interfering with their walking capacity (e.g. cardiovascular or respiratory diseases, Parkinson, arthritis). This latter criterium also applied for HC.

2.2 Descriptive measures

Demographic and clinical characteristics were collected (i.e. age, gender, MS phenotype, disease duration, EDSS and functional system scores (FS)). Cognitive and motor functions of the PwMS were documented using the following descriptive measures; -1- The Timed 25 Foot Walk (T25FW) to assess mobility and functionality of the lower limbs. -2- The Nine Hole Peg Test (NHPT) assessing manual dexterity. -3- The Paced Auditory Serial Addition Test (PASAT) to assess cognitive attention. -4- The Symbol Digit Modality Test (SDMT) assessing processing speed. Completion of the SDMT and PASAT at 1/3, 2/3 and 3/3 of the test was noted to measure cognitive fatigability as the percentage difference between the last third and first third of the test. All test procedures are documented in appendix A.

2.2.1 Questionnaires

Additionally, PwMS completed questionnaires to rate cognitive and motor fatigue (Fatigue scale for motor and cognitive functions; FSMC), the severity of fatigue (Fatigue severity scale; FSS) and the impact of fatigue on physical, cognitive and psychosocial domains (Modified fatigue impact scale; MFIS). Their perceived walking ability was assessed through the Multiple Sclerosis Walking scale (MSWS-12) and their fear of falling was questioned via the Falls Efficacy Scale International (FES-I). Additionally, participants completed questionnaires evaluating sleep problems (Sleep condition indicator; SCI) and anxiety and depression (Hospital anxiety and depression scale: HADS). All questionnaires are documented in appendix B.

2.3 Study design and experimental outcome measures

Participants performed a 6MWT twice with 3-5 days in between to investigate test-retest reliability. There were four assessors in the study (3 master thesis students REKI and one master biomedical sciences), but per participant both test sessions were supervised and evaluated by the same researcher. Before (baseline) and immediately after (post) the 6MWT, short screening tests for spasticity, muscle strength and balance occurred in a randomized order. These measures were repeated every 10 minutes after the walking test for half an hour (post10, post20 and post30). Additionally, they had to fill in the symptom inventory (SI) at the same time slots after the objective screening tests, indicating on perceived symptom severity. If this period was insufficient for the participants to fully recover, they could return home and indicate afterwards when all symptoms returned to baseline (i.e. before the 6MWT).

For the third test session, participants performed a vigilance tasks while seated and while executing the 6MWT. The order of testing was randomized with a resting period of 5-10 minutes in between.

A schematic overview of the experimental protocol is given in figure 5. The case report forms used for testing are documented in appendix C. The experimental outcome measures are listed below.

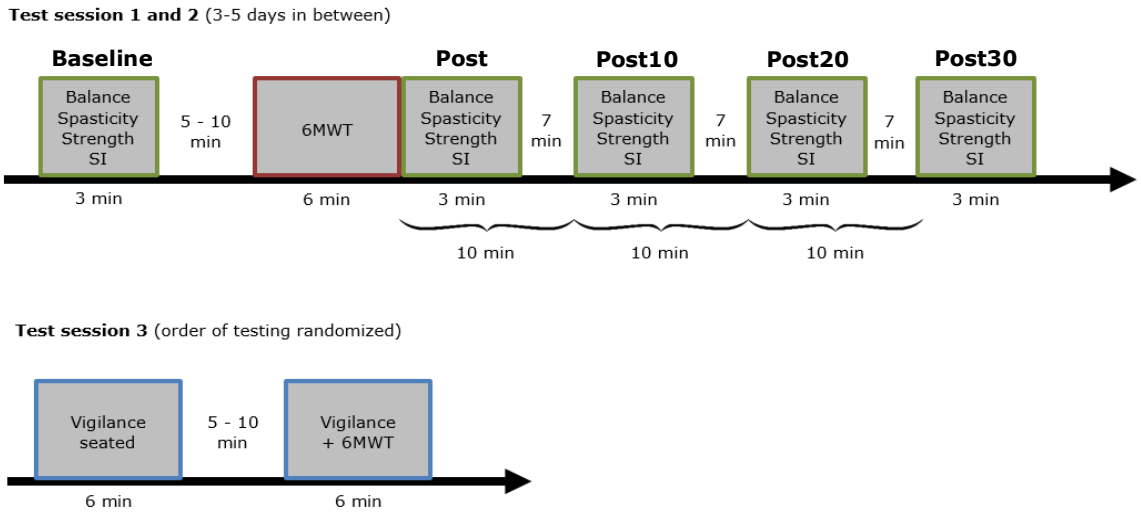

Figure 5: Schematic overview of the experimental protocol. 6MWT: six-minute walking test, SI: symptom inventory.

2.3.1 Six-minute walking test

Subjects performed the 6MWT indoors as fast as possible according to the protocol of Goldman et al (25). They had to walk back and forth in a 30m-hallway, marked every meter, and were allowed to use their (unilateral) assistive device if necessary. Their walking pattern was recorded using three sensors (APDM wearable technologies, Portland, United States of America), one on both feet and one at the level of lumbar region 2. The supplied Mobility Lab Software was used to analyse multiple gait parameters (e.g. stride length, cadence, double support, gait cycles and speed). However, the software was currently not able to provide minute per minute data and will therefore not be included in this data analysis. Their heart rate was recorded using a POLAR heart rate sensor (Polar Electro®, Dendermonde, Belgium), which was connected to a polar watch worn by the subjects. However, heart rate was not included in data analysis due to poor data quality.

Additionally, participants were informed about each expired minute without further encouragements. The distance covered every minute was noted. Additionally, the subjects indicated every minute their perceived walking fatigability on a scale ranging from 0-10 and read out loud their current heartbeat. As a primary outcome measure, the distance walk index (DWI) was calculated using the following formula based on Leone et al. (8):

$$DWI_{6\text{-}1} = \frac{\text{distance walked min 6} - \text{distance walked min 1}}{\text{distance walked min 1}} \text{ x } 100$$

2.3.2 Objective measures

The Romberg test per decade was performed to evaluate balance (Figure 6) (26). Balance problems were assumed when participants were not able to keep their balance for at least 10 seconds (ordinal scale: 0). When participants could keep their balance for 30 seconds, no balance problem was assumed (ordinal scale: 3). Keeping balance between 10 and 30 seconds was considered as a danger zone (ordinal scale: 2). Maximal muscle strength was assessed for ankle dorsiflexion, knee extension and hip flexion using the Motricity Index (MI) for the lower limbs (table 1). Evaluation for spasticity occurred in the m. quadriceps, hamstrings and triceps surae with the Modified Ashworth Scale (MAS) (table 2).

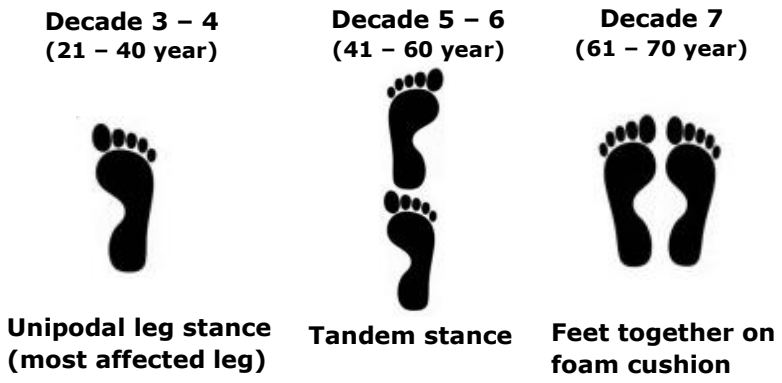

Figure 6: Romberg balance test per decade.

Table 1: Scoring of the MI for the lower limb. MI: motricity index.

MI	Ordinal	Explanation
0	1	No passive movement
9	2	Passive movement is palpable
14	3	Passive movement is visible, but not throughout the full movement range
19	4	Passive movement is possible throughout the full range, but not against resistance
25	5	Passive movement is possible against resistance throughout the full range, but is weaker than the unaffected side
33	6	Normal strength

Table 2: Scoring of the MAS. MAS: modified ashworth scale.

MAS	Ordinal	Explanation
0	1	No increase in muscle tone
1	2	Slight increase in muscle tone, manifested by a catch and release or by minimal resistance at the end of the range of motion when affected part(s) is moved in flexion or extension
1+	3	Slight increase in muscle tone, manifested by a catch and release or by minimal resistance throughout the remainder (less than half) of the range of motion
2	4	More marked increase in muscle tone through most of the range of motion, but affected part(s) is easily flexed
3	5	Considerable increase in muscle tone, passive movement is difficult
4	6	Affected part(s) is rigid in flexion or extension

2.3.3 Symptom Inventory

The SI is a standardized questionnaire based on Skjerbaek et al. consisting of ten possible clinical symptoms commonly present in pwMS; General fatigue, motor fatigability, muscle weakness, gait pattern impairments, balance disturbance, spasticity, visual impairment, sensory disturbance, pain and dizziness (27). 'Attention problems' was added to the questionnaire to have an indication of the involvement of cognitive problems in WF. They had to indicate the severity of these 11 symptoms on a visual analogue scale (VAS) ranging from zero to ten. Zero indicated that they did not experience the symptom, whereas ten indicated the most possible severity of the symptom.

2.3.4 Auditory vigilance task

A subsample of the subjects participated in a third test session, investigating cognitive fatigability. To measure cognitive fatigability, the participants had to perform an attention test for six minutes; the auditory vigilance alphabet test. This vigilance task was chosen because a previous study in our research group showed that performing this cognitive task together with a motor task for one minute is not considered as a dual task. In this study, the test was performed twice, while seated and while performing the 6MWT, in a random order with a 10 minute break in between (figure 1).

A tablet application (developed by PXL and EDM, UHasselt) was used to execute the vigilance test. Participants worn a headset with microphone connected to the tablet via a cable, through which they listened to random letters of the alphabet at a rate of one letter per 1.5 seconds. Each time they heard one of the two target letters (i.e. R and L), they had to respond with 'yes'. Their responses will be noted by the assessor on the tablet and will be recorded via the microphone, giving audio files per minute. Reaction times on the target letters were determined using Sonic Visualiser 3.0 (Queen Mary University, London). The primary outcome of this vigilance task was the average reaction time on the target letters each minute. Hereby, the reaction time index ($RTI_{6\text{-}1}$) could be calculated in the same way as the $DWI_{6\text{-}1}$;

$$RTI_{6\text{-}1} = \frac{\text{average reaction time min 6} - \text{average reaction time min 1}}{\text{average reaction time min 1}} \text{ x } 100$$

In total, three test conditions could be used to investigate on cognitive fatigability and its possible relationship with walking fatigability; -1- A single task condition in which the participants only performed the cognitive task while seated, -2- A single task condition in which the participants only performed the 6MWT and -3- A dual condition in which the participants performed the cognitive and walking task simultaneously (i.e. VigilanceWalk). For the single walk condition, the outcome of the first 6MWT session was used.

2.3 Data analysis

Data analysis was performed using SPSS Statistics 25 (IBM Analytics, Brussels, Belgium). Test-retest reliability of the 6MWD and $DWI_{6\text{-}1}$ was calculated using the intraclass correlation coefficient (ICC) and standard error of measurement (SEM) based on a mean-rating (k = 2), absolute agreement, 2-way mixed-effects model. ICC values between 0.00 and 0.25 were considered as no to poor correlations, between 0.25 and 0.50 as fair correlations, between 0.50 and 0.75 as moderate to good correlation, and between 0.75 and 1.00 as good to excellent correlations according to Terry and Mae (28). Test session 1 was randomly chosen to perform further data analysis. Normality was checked using the Shapiro-Wilk test for normality and evaluation of Q-Q plots. Data was also considered normally distributed for n<30.

The manifestation during and after the 6MWT in terms of perceived fatigability, performance fatigability and subjective symptom severity was evaluated using repeated measures ANOVA with post hoc Bonferroni corrections for groups, timepoints and the groups*timepoints interaction. To evaluate the manifestation of the objective measures (ordinal data), Friedman tests were used. Post hoc corrections for differences between timepoints occurred using Wilcoxon rank tests. Post hoc corrections for differences between groups occurred using Mann Whitney U tests. Pearson r correlation coefficients were calculated between the DWI_{6-1} and all secondary outcomes (i.e. subjective and objective symptoms, descriptive variables and questionnaires). Spearman rho correlations were calculated when data was not normally distributed. Correlations <0.30 were considered very low, between 0.30 and 0.49 low, between 0.50 and 0.69 moderate, between 0.70 and 0.89 high and >0.90 as very high correlations (29). Differences between all groups were evaluated using independent-samples t-tests if normality was assumed. Mann-Whitney U tests were used in case of no normal distribution.

3. Results

Test-retest reliability of the assessment method, and manifestation before, during and after the 6MWT will be presented in this section. PwMS were subdivided into a WF and NWF group, consisting of 24 and 25 patients respectively.

3.1 Study population

In total, 52 pwMS and 32 HC were enrolled in the study. However, one pwMS and one HC were excluded due to a secondary condition (Hashimoto syndrome and chronic fatigue respectively) interfering with their walking capacity. Another pwMS and a HC dropped out of the study after completion of only 1 session. Based on the differences between the two sessions, one pwMS and two HC were considered as outliers (Δ[session2-session1]>mean±1.96SD) and were therefore excluded (Figure 7 and 8). As a result, 49 pwMS and 28 HC were included for data-analysis (Figure 9).

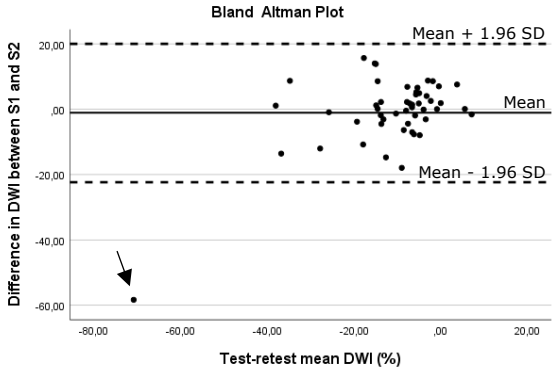

Figure 7: Bland Altman plot for the DWI of the two sessions in MS. Arrow indicates outlier. DWI: distance walked index, S1: session 1, S2: session 2, MS: multiple sclerosis.

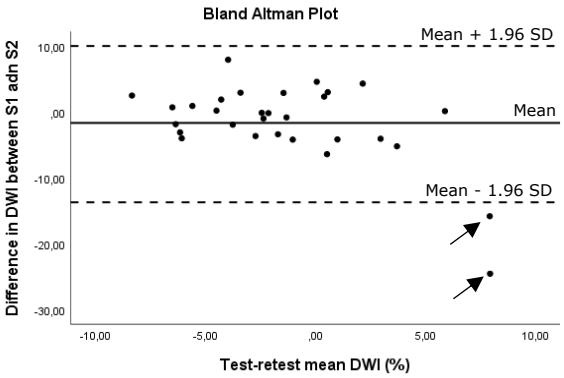

Figure 8: Bland Altman plot for the DWI of the two sessions in HC. Arrows indicate outliers. DWI: distance walked index, S1: session 1, S2: session 2, HC: healthy controls.

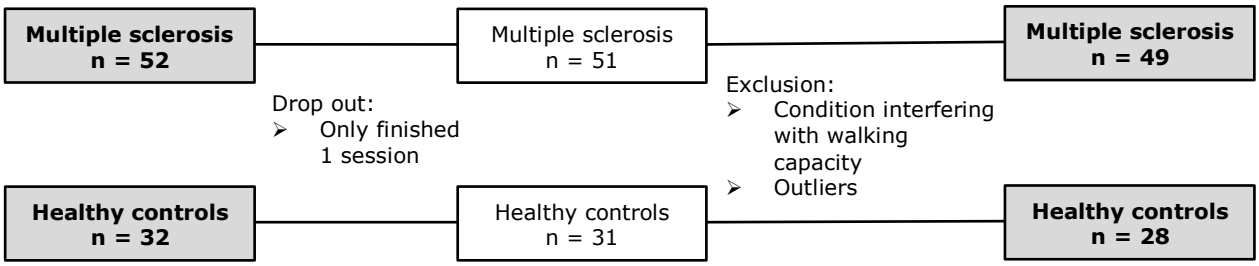

Figure 9: Flowchart exclusion participants for data analysis.

3.1.1 Demographic and clinical characteristics

Demographic characteristics for all groups are presented in table 3. No significant differences were found between any groups for age, gender and length. Only weight differed significantly for HC-MS and HC-WF.

Table 3: Demographic characteristics. Data is represented as mean±SD for total MS group, MS fatigability subgroups (WF and NWF) and HC. Corresponding p-values are noted to indicate significant differences between descriptives.

	MS				
	Total (n=49)	**WF (n=24)**	**NWF (n=25)**	**HC (n=28)**	**p-value**
Demographics					
Age (years)	47.48±10.17	49.00±11.47	46.85±9.70	49.19±11.71	NS[a,b]
Gender: female (n; %)	35; 71.43%	19; 79.17%	16; 64.00%	21; 75.00%	NS[d]
Length (cm)	166.95±25.55	162.24±34.90	171.67±8.38	167.70±9.11	NS[a,b]
Weight (kg)	75.32±14.00	75.92±13.87	74.72±14.38	69.07±14.87	<0.05[c]

[a] Mann-Whitney U test WF-NWF, [b] Independent-samples t-test MS-HC, [c] Mann-Whitney U test HC-MS and HC-WF, [d] Chi-square test between all groups.
MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability, HC: healthy controls, SD: standard deviation, NS: not significant (p>0.05).

The clinical MS characteristic are presented in table 4. For five pwMS, no MS specific data was available, except for the MS phenotypes of two of them. For another pwMS (NWF), no demographic data was available. Two pwMS were not able to complete the PASAT due to an inability to understand the task. The EDSS was significantly higher in WF compared to NWF. The measurements for motor function were significantly lower in the WF group. The FS scores, disease duration and cognitive function tests did not significantly differ between WF and NWF. For the questionnaires, significant higher scores were found in WF compared to NWF for the MFIS physical, HADS depression, MSWS-12 and FES-I. All the other questionnaires did not show any significant differences between both groups.

Table 4: Clinical MS characteristics. Data is represented as mean±SD for total MS group and MS fatigability subgroups (WF and NWF). Corresponding p-values are noted to indicate significant differences.

	MS			
	Total (n=49)	**WF (n=24)**	**NWF (n=25)**	**p-value**
MS characteristics				
MS-type (n)	RRMS: 41 PPMS: 2 SPMS: 3 Not known: 3	RRMS: 18 PPMS: 2 SPMS: 3 Not know: 1	RRMS: 23 Not known: 2	NS[c]
Disease duration (years)	12.13±7.47	13.73±7.56	10.58±7.20	NS[b]
EDSS	3.28±1.88	4.14±1.45	2.43±1.90	0.002[a]
FS scores				
– Bowel and bladder	1.15±1.20	1.39±1.34	0.87±0.99	NS[b]
– Brainstem	0.44±0.96	0.58±1.07	0.27±0.80	NS[b]
– Cerebellar	1.15±1.18	1.37±1.12	0.87±1.25	NS[b]
– Cerebral	1.09±1.11	1.26±1.20	0.87±0.99	NS[b]
– Pyramidal	1.91±1.29	2.26±1.05	1.47±1.46	NS[b]
– Sensory	1.79±1.43	2.05±1.31	1.47±1.36	NS[b]
– Visual	0.44±1.02	0.68±1.25	0.13±0.52	NS[b]
Motor function				
T25FW (m)	5.92±4.71	5.91±1.77	5.93±6.43	0.005[b]
NHPT dominant (s)	22.62±5.88	23.97±6.02	22.05±4.50	0.039[b]
NHPT non-dominant (s)	24.38±7.50	26.81±9.18	21.66±0.93	0.016[b]
Cognitive function				
PASAT (#correct)	45.13±13.87	41.48±14.72	48.63±12.31	NS[b]
SDMT (#correct)	47.78±12.77	44.67±12.28	50.76±12.75	NS[a]
Questionnaires				
MFIS total	38.31±18.01	43.04±18.14	33.58±16.94	NS[a]
MFIS physical	17.48±8.13	19.83±7.70	15.13±8.02	0.044[a]
MFIS cognitive	17.69±9.33	19.42±9.82	15.96±8.69	NS[a]
MFIS psychosocial	3.29±2.05	3.79±2.11	2.80±1.91	NS[b]
FSMC total	63.30±18.70	67.95±15.70	59.04±20.49	NS[b]
FSMC physical	33.17±9.34	36.04±6.18	30.42±11.04	NS[b]
FSMC mental	30.40±10.02	32.26±9.70	28.63±9.95	NS[b]
HADS total	11.87±8.50	14.17±8.92	9.67±7.62	NS[b]
HADS anxiety	6.21±4.45	7.09±4.94	5.38±3.81	NS[b]
HADS depression	5.66±4.83	7.09±4.86	4.29±4.49	0.046[b]
MSWS-12	34.92±14.48	41.17±11.94	28.67±14.31	0.004[b]
FSS average	4.75±1.39	5.07±1.29	4.42±1.44	NS[b]
SCI[a]	20.39±8.26	18.00±7.48	22.58±8.49	NS[a]
FES-I	28.81±11.85	34.63±12.14	23.00±8.31	0.001[b]

[a] independent samples t-test, [b] Mann-Whitney U test, [c] Chi-square test

MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability, RRMS: relapsing remitting multiple sclerosis, PPMS: primary progressive multiple sclerosis, SPMS: secondary progressive multiple sclerosis, EDSS: expanded disability system score, FS scores: functional system scores, T25FW: timed 25 foot walk, NHPT: nine hole peg test, PASAT: paced auditory vigilance addition test, SDMT: symbol digit modality test, MFIS: modified fatigue impact scale, FSMC, fatigue scale for motor and cognitive function, HADS: hospital anxiety and depression scale, MSWS-12: multiple sclerosis walking scale, FSS: fatigue severity scale, SCI: sleep condition indicator, FES-I: falls efficacy scale international, SD: standard deviation, NS: not significant ($p>0.05$).

3.2 Assessment of walking fatigability

To decide which DWI_{6-1} cut-off should be used to state WF, a control group was included in this study. The DWI_{6-1} of the HCs ranged from -9.57% to 6.32%. Therefore, the use of $DWI_{6-1} \leq -10\%$ was taken into consideration to state WF. Both cut-off values (i.e. -10% and -15%) were able to significantly differentiate between WF and NWF, as a continues walking distance decline can be seen in the WF group (Figure 10).

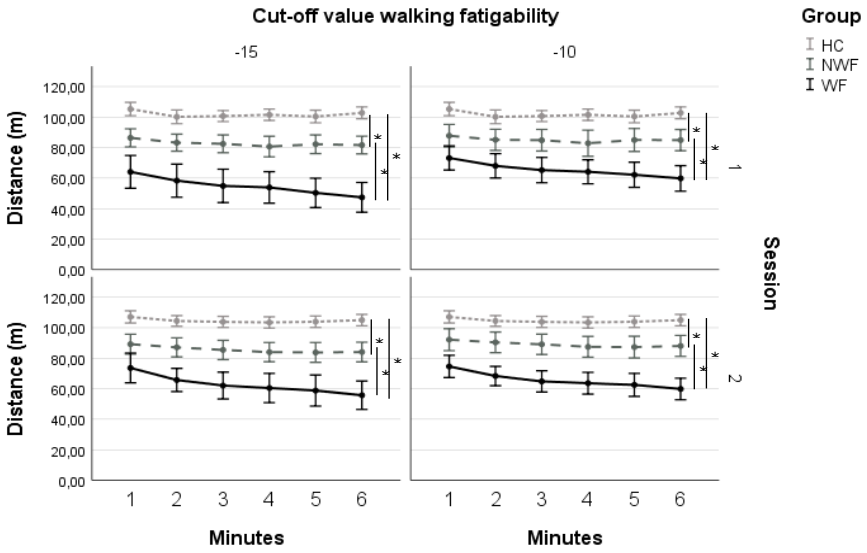

Figure 10: Distance walked every minute of the 6MWT for both MS (WF, NWF) and HC. Walking distance is represented as mean ± SEM. The 6MWT was executed two times (session 1 and 2; upper and lower graphs respectively). MS subgroups are based on a DWI_{6-1} cut-off value of -10% or -15% (right and left graphs respectively). Data is presented as mean±2SE. * indicates $p<0.05$. HC: healthy controls, MS: Multiple Sclerosis, NWF: non-walking fatigability, WF: walking fatigability, 6MWT: six-minute walking test, DWI: distance walked index, SEM: standard error of mean.

Beside the determination about which cut-off value to use, it is also important to consider if patients need to meet the criteria in both sessions in order to state WF. Therefore, the prevalence of both cut-off values was compared to the prevalence of WF in the study of Leone et al. (2015), conducted on a larger sample (table 5). The prevalence of WF in both session showed significant lower percentages compared the prevalence study of Leone et al. (2015).

Table 5: Prevalence of WF in lower and higher EDSS categories compared for different WF criteria. WF: walking fatigability, EDSS: expanded disability system score, DWI: distance walked index.

	Leone et al. $DWI_{6-1} \leq -15\%$	Both sessions $DWI_{6-1} \leq -15\%$	Both sessions $DWI_{6-1} \leq -10\%$	At least one session $DWI_{6-1} \leq -15\%$	At least one session $DWI_{6-1} \leq -10\%$
EDSS [1-4]	12.38%	0.00%	3.70%	11.10%	29.60%
EDSS [4.5-6]	47.13%	27.80%	61.10%	61.10%	77.80%

Therefore, based on comparisons between the prevalence of WF in our previous study and the fact that HC not exceed a decline of 10%, conclusion were made towards the use of the $DWI_{6-1} \leq -10\%$ in at least one of the two walking sessions as criterium to state WF. Moreover, if ICC values between the two sessions are good, it is not important to show the WF two times.

3.3 Test-retest reliability of the distance walked index

The 6MWD and DWI_{6-1} for both session, together with reliability measures are presented in table 6. Differences on the 6MWD and the DWI_{6-1} were both significant between all groups at both sessions. The difference between the two session (Δ[session1-session2]) in each group was not significantly different from zero, except for the NWF group. To evaluate if the walking task is performed good twice by the participants, test-retest reliability was calculated for the 6MWD. The ICC for 6MWD was considered excellent in all groups. The ICC for DWI_{6-1} was considered good in the total MS group, and moderate in the other groups. The standard error of measurement (SEM) provides an absolute index for the reliability, whereas the minimal detectable change (MDC) was calculated to determine whether a change in score can be considered without measurement error.

Table 6: Test-retest reliability of the 6MWD and DWI_{6-1} for MS and HC. Session 1: 20 WF and 29 NWF, session 2: 18 WF and 31 NWF. A $DWI_{6-1} \leq$-10% was used to state WF (NWF: DWI>-10%).

	MS			
	Total (n=49)	**WF** (n=24)	**NWF** (n=25)	**HC** (n=28)
6MWD				
Session 1[a]	462.43±126.05	391.46±103.85	530.56±107.55	610.86±60.42
Session 2[a]	482.80±124.56	413.50±102.73	549.32±107.27	627.75±57.24
Cronbach's Alpha	0.98	0.97	0.98	0.96
Pearson's r	0.96**	0.94**	0.96**	0.92**
ICC	0.95	0.92	0.94	0.89
DWI_{6-1}				
Session 1[a] (%)	-9.85±10.97	-17.68±10.12	-2.34±4.78	-2.20±3.61
Session 2[a] (%)	-10.19±9.89	-16.29±10.43	-4.33±4.30	-1.84±4.04
Δ(session1- session2)[a] (p-value)	0.34±7.26 (0.75)[d]	-1.39±9.27(0.47)[d]	2.00±4.14 (0.02)[d]	-0.36±3.45(0.59)[d]
Cronbach's Alpha	0.86	0.75	0.74	0.75
Pearson's r	0.76**	0.59**	0.59**	0.60**
ICC (95% CI)	0.76 (0.61-0.86)	0.60 (0.27-0.80)	0.54 (0.20-0.77)	0.60 (0.30-0.79)
SEM[b] (%)	5.10	6.73	3.07	2.42
MDC[c] (%)	14.12	18.04	8.50	6.70

MS: multiple sclerosis, HC: healthy controls, WF: walking fatigability, NWF, non-walking fatigability, 6MWD: six-minute walking distance, DWI_{6-1}: distance walked index, ICC: intraclass correlation coefficient, CI: confidence interval, SEM: standard error of measurement, MDC: minimal detectable change.

**p<0.01, [a]Mean±SD, [b]SEM = SD $\sqrt{1-ICC}$, [c]MDC = 1.96 SEM $\sqrt{2}$, [d]one sample t-test

3.4 Manifestation of walking fatigability during the 6MWT

The manifestation of WF during the 6MWT was evaluated in terms of distance and perceived fatigability every minute. Data was normally distributed according to Q-Q plots.

3.4.1 Performance walking fatigability

The course of the mean distance covered each minute of the 6MWT for WF, NWF and HC are shown in figure 11. Significant differences were seen between all groups at every minute of the 6MWT. Minute per minute analysis in each group showed only significant differences in distance between the first and the second minute in WF and HC. For the NWF, no significant differences were observed minute to minute. Data was normalized to minute 1 in order to visualize the decline in walking distance after the first minute (Figure 12). The mean walking distance in WF significantly declined every minute compared to minute 1. In the NWF group, significant declines to minute 1 were seen starting from minute 3. HC showed significant declines compared to minute 1 for all minutes, except for minute 4. Between groups, significant differences were observed between HC and WF starting from minute 3. Significant differences between WF and NWF were present from minute 2 until the end of the 6MWT. No significant differences were found between HC and NWF.

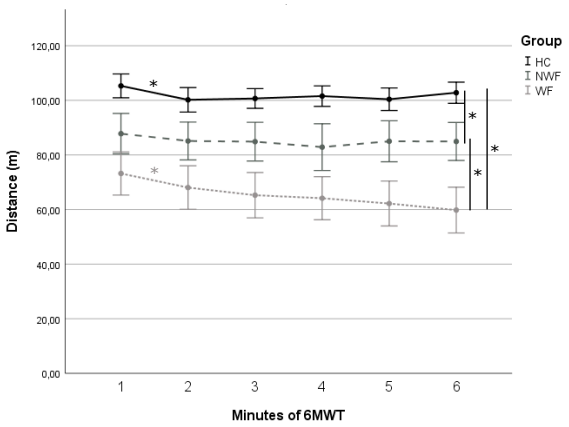

Figure 11: Distance walked every minute of the 6MWT for both MS groups (WF and NWF) and HC. Data is presented as mean±2SE. * indicates p<0.05. 6MWT: six-minute walking test, HC: healthy control, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability.

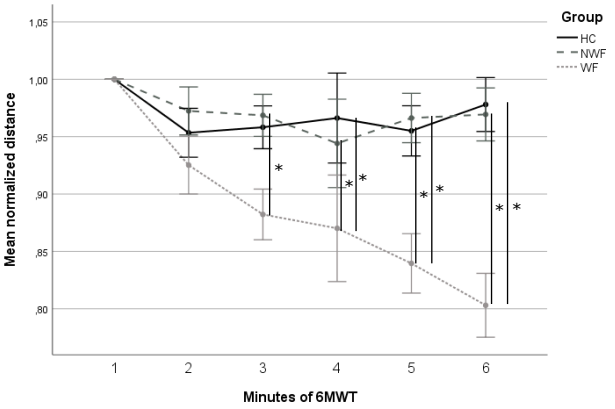

Figure 12: Normalized distance walked every minute of the 6MWT for both MS groups (WF and NWF) and HC. Data is presented as mean±2SE. * indicates p<0.05. 6MWT: six-minute walking test, HC: healthy control, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability.

3.4.2 Perceived walking fatigability

The course of the mean perceived fatigability (VAS score ranging from 0-10) before and during the 6MWT for WF, NWF and HC are shown in figure 13. Significant differences were seen every minute between NWF and WF compared HC. Between WF and NWF, significant differences were only present starting from minute 3 to 5. Minute per minute analysis in the WF group showed significant increases in perceived fatigability between all timepoints. The NWF group also showed significant increases in perceived fatigability after the first minute and all following minutes, except for min2-3. HC only showed significant increases in perceived fatigability after the first minute of walking and between the 5th and last minute of the 6MWT. After normalization of the perceived fatigability with baseline (pre VAS scores), no significant differences in increase of perceived fatigability were found between any of the groups (figure 14). In WF, significant increases in perceived fatigability compared to the pre VAS score were already seen after the first minute of the 6MWT. In NWF and HC, significant increases in perceived fatigability compared to pre VAS scores were seen after minute 2.

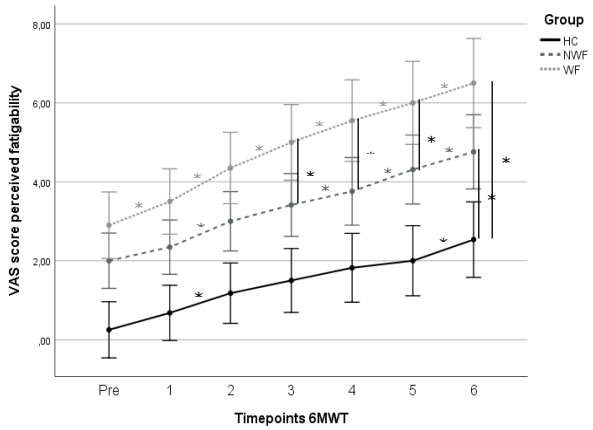

Figure 13: Perceived fatigability (0-10 VAS) before (pre) and after every minute of the 6MWT for both MS groups (WF and NWF) and HC. Data is presented as mean±2SE. * indicates p<0.05. VAS: visual analogue scale, 6MWT: six-minute walking test, HC: healthy controls, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability.

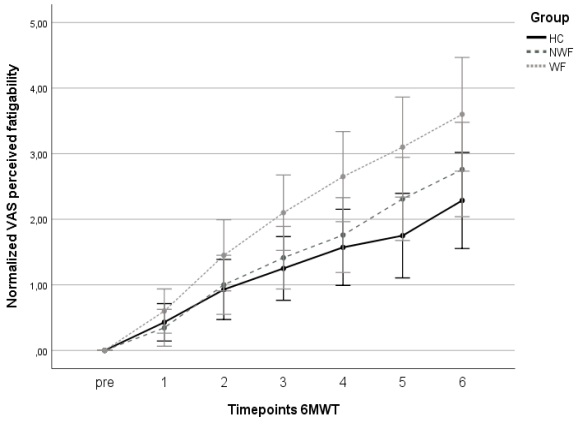

Figure 14: Normalized perceived fatigability (0-10 VAS) before (pre) and after every minute of the 6MWT for both MS groups (WF and NWF) and HC. Data is presented as mean±2SE. * indicates p<0.05. VAS: visual analogue scale, 6MWT: six-minute walking test, HC: healthy control, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability.

3.5 Manifestation of walking fatigability after the 6MWT

3.5.1 Subjective symptoms

The course of all the symptoms before and after the 6MWT for MS and HC are shown in figure 15.

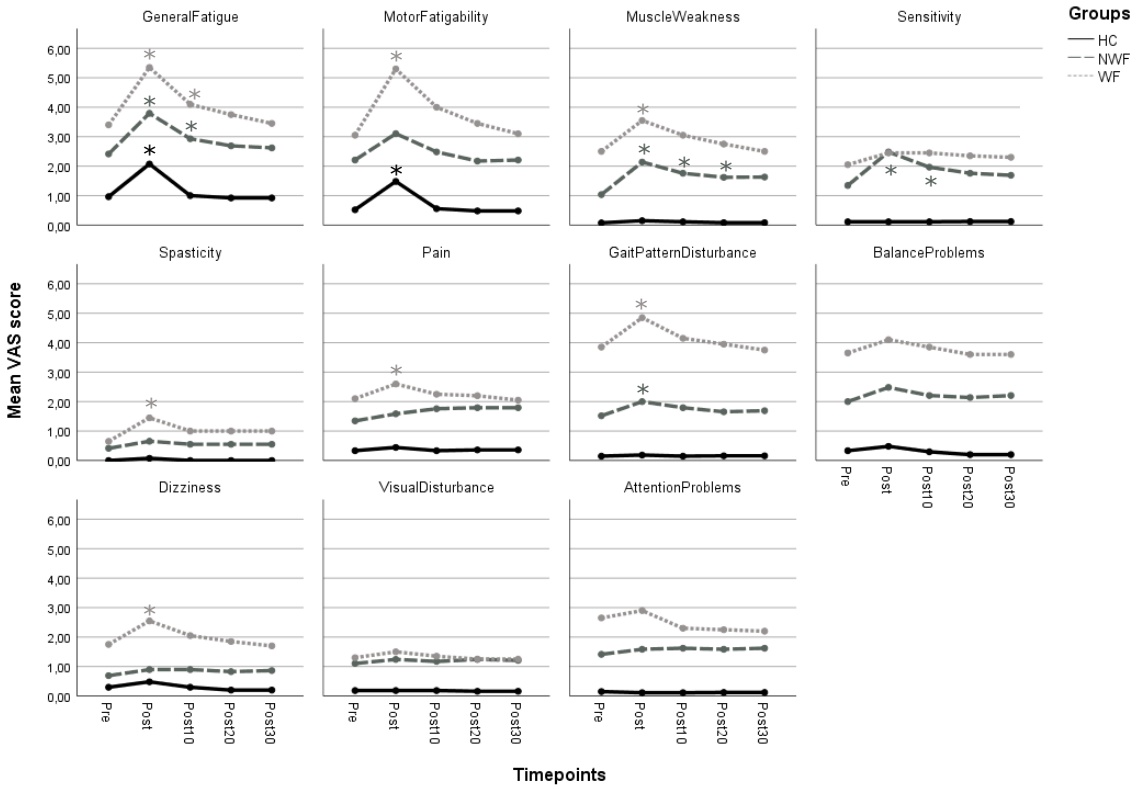

Figure 15: Mean VAS scores (range 0-10) for all symptoms of the symptoms inventory pre, post, 10, 20 and 30 minutes after the 6MWT for HC, WF and NWF. *Significant difference relative to VAS pre 6MWT ($p<0.05$). VAS: visual analogue scale, 6MWT: six-minute walking test, HC: healthy controls, WF: walking fatigability, NWF: non-walking fatigability.

General fatigue VAS scores increased significantly after the 6MWT in all groups. For the WF and NWF group, the significant differences with pre scores were present until post10. For the HC, only a significant difference post 6MWT was observed. **Motor fatigability** VAS scores increased significantly after the 6MWT in HC and WF, but not in NWF. For the WF group, the significant differences with baseline were present until post10.

Muscle weakness VAS scores increased significantly after the 6MWT in WF and NWF. For the NWF group, significant differences with baseline scores were present until post20. In the WF group, only a significant difference post 6MWT was observed. In HC, no significant difference post 6MWT was observed. **Gait pattern impairments** VAS scores showed significant increases in both MS groups, but at post10 no significant differences with VAS scores before the 6MWT were found in both MS groups. HC showed no significant increase in gait pattern impairments.

The VAS scores for **spasticity** and **pain** did only a show significant increase after the 6MWT in the WF group. Post10 there was no significant difference anymore with baseline VAS scores. In HC and NWF, no significant differences were found between baseline and post VAS scores.

Dizziness VAS scores showed no significant increase after the 6MWT for HC and NWF. For the WF group, a significant increase was observed post 6MWT, but post10 no significant difference with VAS scores before the 6MWT was found. **Sensitivity** VAS scores did only significantly increase in the NWF group. The significant difference with baseline was present until post10. The VAS scores for **attention problems**, **visual disturbance** and **balance problems** did not show significant differences between or within all the groups at any timepoint.

The post VAS scores were normalized to baseline VAS scores in each group to visualize differences in the increase of the symptoms in relation to their baseline experience (Figure 16). Data was not normally distributed according to the Shapiro-Wilk test for normality. Man-Whitney U tests between HC and NWF showed significant differences in VAS score increase for sensitivity, gait pattern impairments and muscle weakness. Between HC and WF, all symptoms were significantly more increased after the 6MWT, except for balance and pain. Comparisons between NWF and WF showed significant increases in VAS scores for motor fatigability, spasticity and dizziness. Differences between the groups at baseline level are presented in section 3.6.1 concerning the clinical profile of WF.

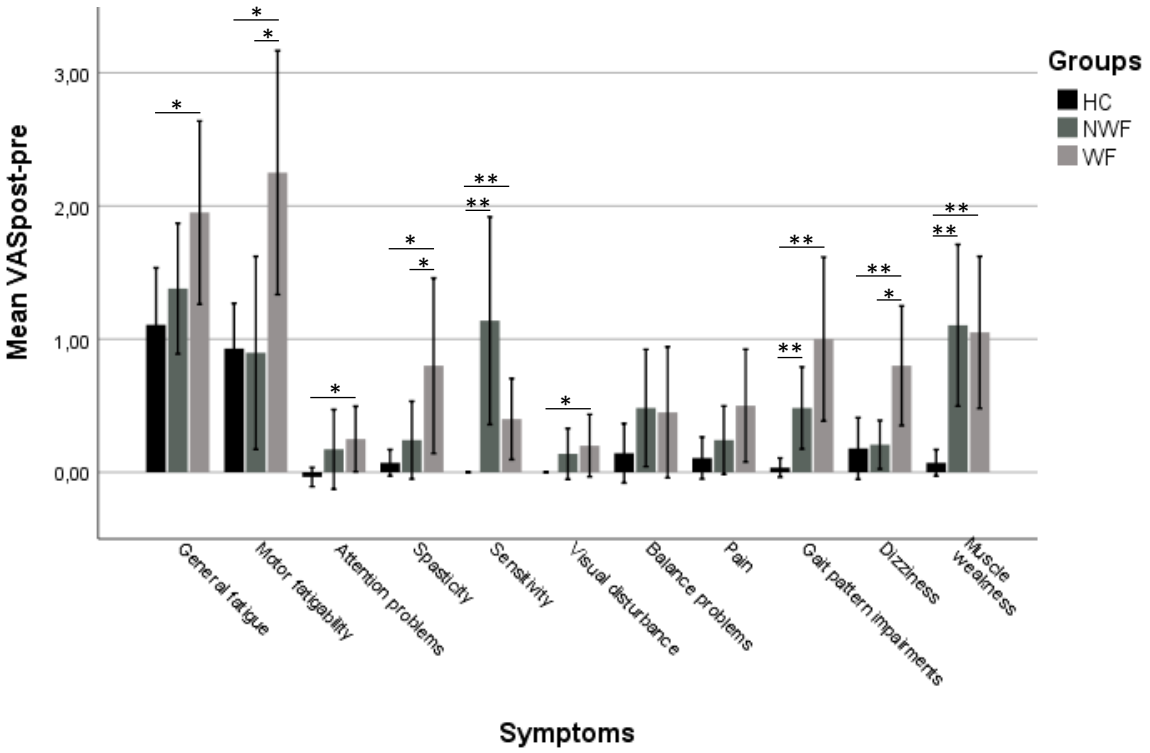

Figure 16: Difference in increase of subjective symptoms (0-10 VAS) pre-post 6MWT in WF, NWF and HC. Data is presented as mean±2SE. * indicates p-values <0.05, ** indicates p-values <0.01. VAS: visual analogue scale, 6MWT: six-minute walking test. WF: walking fatigability, NWF: non-walking fatigability, HC: healthy controls.

3.5.2 Objective symptoms

The median and interquartile range (IQR; Q1-Q3) of the objective symptoms at baseline (i.e. before the 6MWT) in each group are presented in table 7. In the NWF group, a significant decrease in the MI for right knee extension was seen post and post10 compared to baseline measures. For the MI of the left knee extension and the hip flexion of both legs, a significant decrease was seen post compared to baseline. In the WF group, only a significant decrease in the MI of the left knee extension was seen post compared to baseline measures. The MAS measurements showed no significant differences after the 6MWT compared to baseline in any group. For HC, a significant increase in balance was seen at post10, post20 and post30 compared to baseline. Moreover, no spasticity was found in HC, what could be expected.

Post (i.e. immediately after the 6MWT) significant differences between HC and NWF were found for the MAS of the right quadriceps and hamstrings (NWF>HC), the MI of the three tested muscles of both legs (NWF<HC) and balance (NWF<HC). Between HC and WF, significant lower post scores are seen in the WF group for the MI of all muscles and the Romberg. The MAS score were significantly higher in the WF group, except for the left hamstrings. Comparisons between WF and NWF, showed significant lower post scores for the MI of the right ankle dorsiflexion and hip flexion in the WF group. Balance was also considered as significantly lower in WF compared to NWF after the 6MWT. No significant differences in post MAS measurements were seen between WF and NWF.

Table 7: Objective measurements scores at baseline (i.e. before the 6MWT) for the MAS, MI and Romberg in WF, NWF and HC. Data is represented as median (Q1-Q3).

	MS		
	WF (n=20)	**NWF (n=29)**	**HC (n=28)**
Baseline MI			
Ankle dorsiflexion R	5.00 (4.00-6.00)	6.00 (5.50-6.00)	6.00 (6.00-6.00)
Ankle dorsiflexion L	6.00 (5.00-6.00)	6.00 (5.50-6.00)	6.00 (6.00-6.00)
Knee extension R	5.00 (5.00-6.00)	6.00 (5.50-6.00)	6.00 (6.00-6.00)
Knee extension L	6.00 (5.00-6.00)	6.00 (5.00-6.00)	6.00 (6.00-6.00)
Hip flexion R	5.00 (5.00-5.00)	6.00 (5.00-6.00)	6.00 (6.00-6.00)
Hip flexion L	5.50 (5.00-6.00)	6.00 (5.00-6.00)	6.00 (6.00-6.00)
Baseline MAS			
Quadriceps R	1.00 (1.00-1.75)	1 00 (1.00-1.00)	1.00 (1.00-1.00)
Quadriceps L	1.00 (1.00-1.75)	1.00 (1.00-1.00)	1.00 (1.00-1.00)
Hamstrings R	1.00 (1.00-1.75)	1.00 (1.00-1.00)	1.00 (1.00-1.00)
Hamstrings L	1.00 (1.00-1.00)	1.00 (1.00-1.00)	1.00 (1.00-1.00)
Triceps R	1.00 (1.00-1.00)	1.00 (1.00-1.00)	1.00 (1.00-1.00)
Triceps L	1.00 (1.00-2.00)	1.00 (1.00-1.00)	1.00 (1.00-1.00)
Baseline Romberg	1.00 (1.00-1.75)	2.00 (1.00-3.00)	3.00 (2.00-3.00)

6MWT: six-minute walking test, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability, HC: healthy controls, MAS: modified ashworth scale, MI: motricity index, Q1-Q3: interquartile range.

3.6 Clinical profile of walking fatigability

3.6.1 Subjective and objective symptoms

The VAS score of all the subjective symptoms at baseline in each group are presented in figure 17. Data was not normally distributed according to the Shapiro-Wilk test for normality. Mann-Whitney U test between HC and both NWF and WF showed significant higher VAS scores in the MS subgroups for all symptoms. Comparisons between NWF and WF showed significant higher VAS scores in WF for gait pattern impairments and muscle weakness.

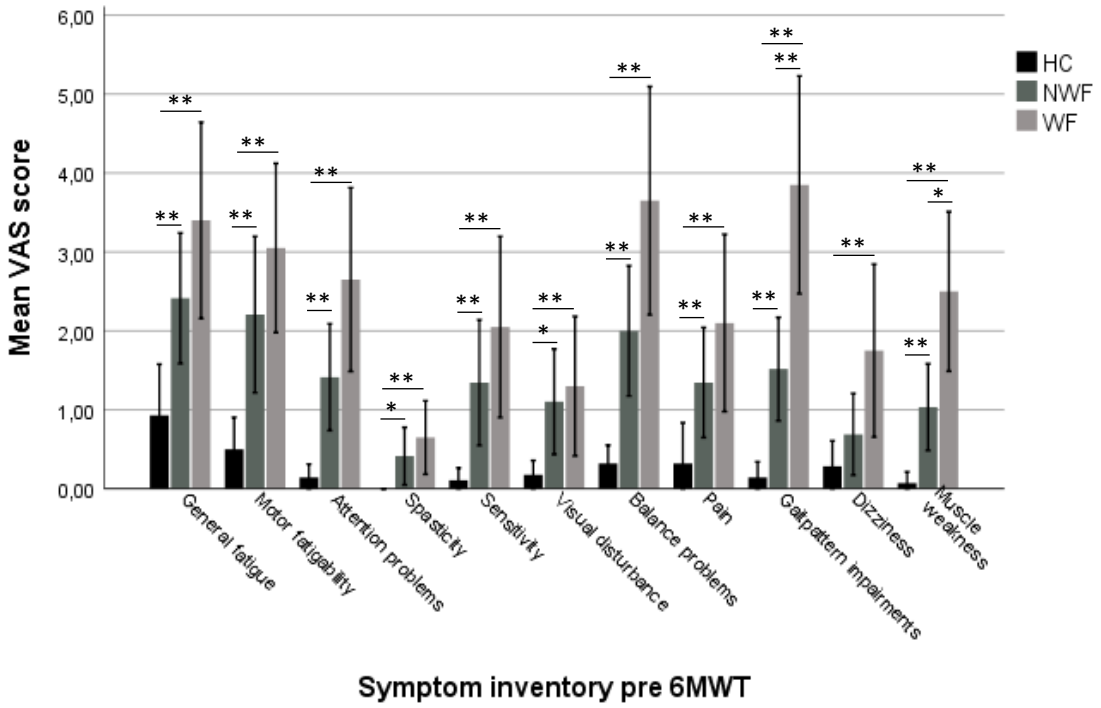

Figure 17: Difference in subjective symptoms (0-10 VAS) pre 6MWT between WF, NWF and HC. Data is presented as mean±2SE. * indicates p-values <0.05, ** indicates p-values <0.01. VAS: visual analogue scale, 6MWT: six-minute walking test. WF: walking fatigability, NWF: non-walking fatigability, HC: healthy controls.

The outcome measures of all objective symptoms at baseline in each group are presented in table 7. Data was not normally distributed according to the Shapiro-Wilk test for normality. Mann-Whitney U tests between HC and both NWF and WF showed a significant lower muscle strength in the WF and NWF group for ankle dorsiflexion, knee extension and hip flexion in both legs. Comparisons between NWF and WF showed only significant lower muscle strength in WF for all MIs of the right leg. The Romberg balance test at baseline showed significant differences between all groups (HC>NWF>WF). Significant differences in MAS were found between HC and NWF for the right quadriceps (HC<NWF) and between HC and WF for all MAS measurements (HC<WF), except for the left hamstrings. Between WF and NWF, spasticity was significantly higher in WF for the left triceps.

Besides, correlations of the subjective and objectives symptoms present at baseline (i.e. before the 6MWT) with the DWI_{6-1} were calculated and presented in table 8. The baseline VAS scores of the symptom inventory in MS showed low to moderate significant negative correlations for motor fatigability, sensitivity, gait pattern impairments, dizziness and muscle weakness (Table 8A). The objective measures at baseline only showed moderate significant positive correlations for MI of the right ankle dorsiflexion and hip flexion, as well as for the Romberg test (Table 8B).

Table 8: Correlations between DWI_{6-1} and symptoms pre 6MWT in MS. 8A) Pearson r correlations between the DWI_{6-1} and pre reported VAS scores for subjective symptoms. 8B) Spearman rho correlations between the DWI_{6-1} and objectively measures symptoms pre 6MWT. DWI: distance walked index, 6MWT: six-minute walking test, MS: multiple sclerosis.

A. Subjective symptoms pre	**Pearson r correlation with DWI_{6-1}**
General fatigue	-0.24
Motor fatigability	-0.28*
Attention problems	-0.28
Spasticity	-0.16
Visual disturbance	-0.11
Sensitivity	-0.32*
Balance problems	-0.25
Pain	-0.16
Gait pattern impairment	-0.47**
Dizziness	-0.29*
Muscle weakness	-0.39*

* p<0.05, ** p<0.001

B. Objective symptoms pre	**Spearman rho correlation with DWI_{6-1}**
Modified Ashworth Scale	
– Quadriceps R	-0.05
– Quadriceps L	-0.14
– Hamstrings R	-0.09
– Hamstrings L	-0.10
– Triceps R	-0.04
– Triceps L	-0.22
Motricity index	
– Ankle dorsiflexion R	0.40**
– Ankle dorsiflexion L	0.25
– Knee extension R	0.27
– Knee extension L	0.06
– Hip flexion R	0.35*
– Hip flexion L	0.14
Romberg	0.49**

* p<0.05, ** p<0.001

3.6.2 Clinical MS characteristics and questionnaires

As mentioned before, the EDSS in the WF group is significantly higher and the measurements for motor function were significantly lower in the WF compared to NWF. Pearson correlations between the DWI_{6-1} and the clinical MS characteristics showed only significant negative correlations with the EDSS and the NHPT (Table 8). The FS scores showed no significant correlations with DWI_{6-1}, nor significant differences between WF and NWF.

For the questionnaires, significant higher scores were found in WF compared to NWF for the MFIS physical, HADS depression, MSWS-12 and FES-I. Pearson correlations between the DWI_{6-1} and all the questionnaires showed low to moderate significant correlations with MSWS-12, FES-I, SCI and HADS (Table 9).

Table 8: Pearson r correlations between DWI_{6-1} and clinical MS characteristics.

Clinical characteristics	Pearson r correlation with DWI_{6-1}
Disease duration (years)	-0.17
EDSS	-0.43**
FS Bowel and bladder	-0.06
FS Brainstem	-0.17
FS Cerebellar	-0.21
FS Cerebral	-0.26
FS Pyramidal	-0.24
FS Sensory	-0.31
FS Visual	-0.24
T25FW (m)	-0.03
NHPT dominant (s)	-0.38**
NHPT non-dominant (s)	-0.36*
PASAT (#correct)	0.01
SDMT (#correct)	0.16

* p<0.05, ** p<0.001

DWI_{6-1}: distance walked index, MS: multiple sclerosis, EDSS: expanded disability system score, FS scores: functional system scores, T25FW: timed 25 foot walk, NHPT: nine hole peg test, PASAT: paced auditory vigilance addition test, SDMT: symbol digit modality test.

Table 9: Pearson r correlations between DWI_{6-1} and MS questionnaires.

Questionnaires	Pearson r correlation with DWI_{6-1}
MFIS total	-0.29*
MFIS physical	-0.36*
MFIS cognitive	-0.17
MFIS psychosocial	-0.23
FSMC total	-0.17
FSMC physical	-0.28
FSMC mental	-0.15
HADS total	-0.31*
HADS anxiety	-0.28
HADS depression	-0.32*
MSWS-12	-0.47**
FSS	-0.16
SCI	0.45**
FES-I	-0.56**

* p<0.05, ** p<0.001

DWI_{6-1}: distance walked index, MFIS: modified fatigue impact scale, FSMC, fatigue scale for motor and cognitive function, HADS: hospital anxiety and depression scale, MSWS-12: multiple sclerosis walking scale, FSS: fatigue severity scale, SCI: sleep condition indicator, FES-I: falls efficacy scale international.

3.7 Cognitive fatigability

Nineteen MS and ten HC participated in the third test session to investigate on cognitive fatigability and its relationship to WF. The RTI_{6-1} and the DWI_{6-1} for the three test conditions are summarized in table 10. The DWI_{6-1} was normally distributed in all groups and all conditions according to the Shapiro-Wilk test for normality, except for the DWI_{6-1} of the WF group in the VigilanceWalk condition. The RTI_{6-1} was normally distributed in all conditions and groups. Only significant differences were seen in the single walking condition, were the WF group showed a lower DWI_{6-1} compared to HC and NWF. A measure for cognitive fatigability was also assessed through the PASAT and SDMT declines between the first and the last 1/3 of the test (Table 10). PASAT data per 1/3 of the test was not available for two pwMS in the NWF group. SDMT data was not available for two pwMS in the WF group and one in the NWF group. No significant differences were found between WF and NWF for PASAT or SDMT decline.

For all groups, no significant differences were found in DWI_{6-1} or RTI_{6-1} between the two test conditions (i.e. single task vs dual task). Besides, no significant spearman's rho correlations were found between PASAT decline, SDMT decline and RTI_{6-1} and DWI_{6-1} of both dual and single task conditions in MS (Table 11).

Table 10: RTI_{6-1} and DWI_{6-1} for the dual and single task condition in WF, NWF and HC, as well as the PASAT and SDMT decline in each group. Data is represented as mean±SD. PASAT and SDMT declines were calculated based on the % decline in correct answers between the first 1/3 and the last 1/3 of the test.

	MS			
	WF (n=7)	**NWF (n=12)**	**HC (n=10)**	**p-value**
RTI_{6-1} (%)				
Single vigilance	-9.93±11.83	-1.04±13.71	-2.06±7.29	NS[b]
VigilanceWalk	-8.12±10.52	-1.09±16.58	4.02±24.07	NS[b]
DWI_{6-1} (%)				
Single walking	-21.94±10.99	-4.19±4.28	-2.03±3.42	<0.01[a]
VigilanceWalk	-9.83±16.49	-2.98±4.85	-0.60±7.77	NS[b]
Other cognitive fatigability measures				
PASAT % decline	-10.43±17.28	-11.08±25.83	/	NS[b]
SDMT % decline	4.31±23.57	-6.37±25.92	/	NS[b]

[a]Independent samples t-test HC-WF and WF-NWF, [b]Mann-Whitney U test or independent samples t-test between all groups. RTI_{6-1}: reaction time index, DWI_{6-1}: distance walked index, PASAT: paced auditory serial addition test, SDMT: symbol digit modality test, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability, HC: healthy controls.

Table 11: Spearman's rho correlation between several cognitive and motor fatigability outcomes in MS. All correlations were not significant.

	PASAT decline	**SDMT decline**	**DWI_{6-1} VigilanceWalk**	**RTI_{6-1} VigilanceWalk**
PASAT decline	1.00	-0.01	-0.05	-0.04
SDMT decline	-0.01	1.00	0.11	0.12
DWI_{6-1} VigilanceWalk	-0.05	0.11	1.00	0.10
RTI_{6-1} VigilanceWalk	-0.04	0.12	0.10	1.00
DWI_{6-1} single walking	0.20	-0.16	0.21	0.36
RTI_{6-1} Single vigilance	0.02	-0.48	-0.01	-0.08

RTI_{6-1}: reaction time index, DWI_{6-1}: distance walked index, PASAT: paced auditory serial addition test, SDMT: symbol digit modality test.

An overview of the walking distances each minute of the 6MWT in all groups is shown for both walking conditions, i.e. Single walking and VigilanceWalk, in figure 18. In the single walking condition, significant declines were seen in the WF group for minute 3, 5 and 6 compared to minute 1. No significant declines were found in any minute compared to minute 1 in the VigilanceWalk condition. For the NWF and HC group, no significant declines were found in any minute compared to minute 1 for both walking test conditions. In both test conditions, significant lower walking distances were observed in the WF group compared to HC at all minutes. In the single walking condition, significant lower walking distances were found in NWF compared to HC at minute 1, 2, 3 and 4. In the VigilanceWalk condition, only significant lower walking distances were found in the NWF group compared to HC at minute 1 and 3. Between WF and NWF, no significant differences were found at any minute in any walking condition. No significant differences between the two test conditions were observed at any minute in any group.

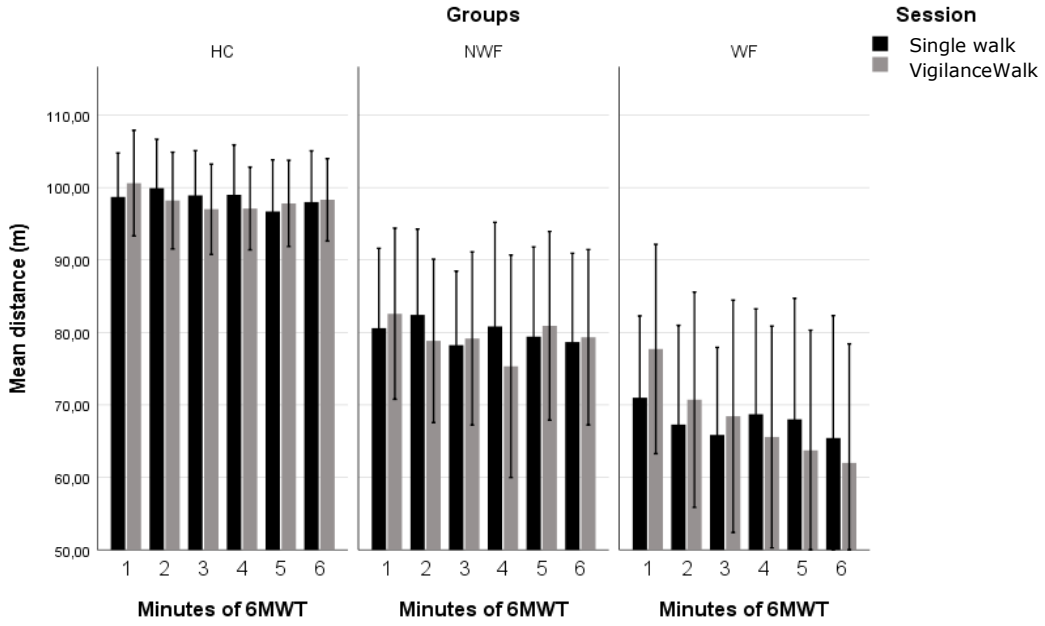

Figure 18: Walking distance each minute of the 6MWT in HC, WF and NWF for both walking conditions (i.e. VigilanceWalk and single walking). Data is represented as mean±2SE. HC: healthy controls, NWF: non-walking fatigability, WF: walking fatigability.

An overview of the reaction times each minute of the six-minute task in all groups is shown for both cognitive conditions, i.e. Single vigilance and VigilanceWalk, in figure 19. In both cognitive conditions, no significant differences were found for any minute compared to minute 1 in all groups. The only significant difference between the groups was found in the single vigilance condition between HC and NWF at minute 3. No significant differences between the two test conditions were observed at any minute in any group.

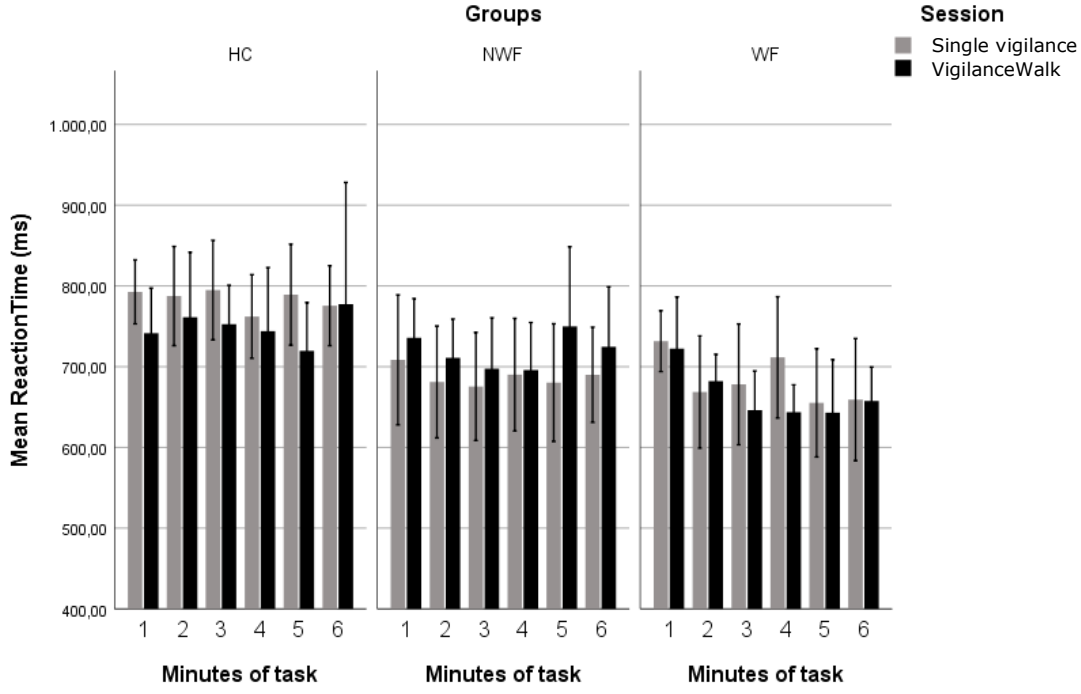

Figure 19: Reaction time each minute of the six-minute task in HC, WF and NWF for both cognitive conditions (i.e. VigilanceWalk and single vigilance). Data is represented as mean±2SE. HC: healthy controls, NWF: non-walking fatigability, WF: walking fatigability.

4. Discussion

In this cross-sectional observational study, the 6MWT was used for the assessment of WF in pwMS, wherein participants were asked to walk as fast as possible. WF was stated by calculating the DWI_{6-1} as percentage change from the first to the sixth minute with a walking distance decline of at least -10% to state WF. This cut-off value can be reported as a good cut-off value because of the inclusion of a control group. By performing the walking test at two different days, test-retest reliability could be investigated and was shown to be good in the total MS group. The symptom inventory and some objective measurements indicated that WF manifests in different symptoms such as gait impairments, balance problems and muscle weakness, but better objective measurements are necessary to confirm these findings.

An auditory vigilance task was performed in a single condition and simultaneously with walking. No significant differences were found in reaction times between the single vigilance task and the VigilanceWalk condition, indicating the vigilance is no dual task when performed simultaneously with walking. Besides, indications towards a protective effect of the vigilance task to declines in walking distance was shown in the WF group, indicating the vigilance task could be a facilitator to improve attention rather than being a disturbing factor.

4.1 Assessment of walking fatigability

WF was assessed through the execution of a 6MWT as fast as possible. Participants did not receive any verbal encouragements during the test, which could result in a submaximal performance. Moreover, some participants never reported VAS scores for perceived fatigability exceeding 5, suggesting an only moderate feeling of fatigue. However, providing no verbal encouragements during the test can also be considered as a strength, since this more represents real-life situations. Besides, the 6MWT is described as a submaximal endurance test and therefore moderate VAS scores are considered normal. Additionally, a study by Marinho et al. (2014) concluded that using verbal instructions during the 6MWT does not improve performance (30).

Testing was performed at three different locations, whereby the testing environment was not always standardized between participants. Disturbing factors were noise in the hallway, passage of personnel and possible obstructions on their walking path (i.e. smaller corridor, uneven surface). However, this possible disturbances were tried to be kept at a minimum. Moreover, as unforeseen circumstances always occur in clinical practice, the outcomes for the 6MWT with $DWI_{6-1} \leq -10\%$ should be reliable regardless of these circumstances in order to be implemented in clinical practice.

The DWI_{6-1} was calculated to measure WF. A previous study in our research group investigated the prevalence of WF and reported the use of a walking distance decline of at least 15% to state WF (8). However, the 6MWT was only performed once and no healthy control group was used as reference.

Because of the varying expressions of MS between days, some participants performed less one day, but better the other day, resulting in conflicting results about the presence of WF between both sessions. Around 10 patients (20%) switched between the WF and NWF group across the two sessions.

Therefore, one point of discussion was if patients needed to show the decline in walking distance during both sessions or not in order to state the presence of WF. Leone et al. (2015) showed that the prevalence of WF significantly differed among EDSS categories. EDSS 4.5 seemed to be a cut-off point in the prevalence of WF by a $DWI_{6\text{-}1} \leq$-15% (Figure 3). When comparing this prevalence to the one in our population, the prevalence of WF when reaching the cut-off in both test sessions was found to be much lower. Therefore, stating that they had to show the cut-off decline twice as criterium for WF, would be too strict (Table 5). On top, even if participants only showed the WF in one session, there can be concluded that they show certain problems with walking. Additionally, when implementing this measurement in clinical practice as a diagnostic assessment method for WF, only one execution of a test is desirable.

Besides, Leone did not state the prevalence of WF by a cut-off value of -10%. Since in the present study a control group was included, in which the $DWI_{6\text{-}1}$ never reached -10% , this lower walking distance decline cut-off was taken into consideration for the measurement of WF. When comparing both cut-off values during both session, there could be concluded that a $DWI_{6\text{-}1} \leq$-10% is sufficient to state WF. By lowering the cut-off value to -10%, there could still be found a significant difference between all the groups (Figure 10). The decline in walking distance every minute can still be clearly seen in the WF group, as well as a comparable course in walking distance every minute of the 6MWT between NWF and HC. Moreover, the SE in the WF was lower for the $DWI_{6\text{-}1} \leq$-10%, which is better. Additionally, the amount of people switching between the WF and NWF group between both sessions was similar for $DWI_{6\text{-}1} \leq$-15% and $DWI_{6\text{-}1} \leq$-1% (nine and ten pwMS respectively).

Extra evidence that the $DWI_{6\text{-}1} \leq$-10% is a good cut-off value for WF was shown by plotting the normalized distances each minute of the 6MWT (Figure 12). Hereby, a clear decline in normalized distance compared to minute one can be seen for the WF group. Moreover, the course in walking distance compared to minute one for each minute of the 6MWT in NWF and HC did not significantly differ, indicating again the cut-off value of -10% is correct.

Generally, it can therefore be concluded that the use of the $DWI_{6\text{-}1} \leq$-10%, present in at least one session, can be used as diagnostic criterium for WF.

4.2 Psychometric properties

Test-retest reliability of the DWI_{6-1} of the 6MWT was investigated in order to draw any conclusion about the use of this DWI_{6-1} as diagnostic assessment tool for WF in pwMS. Significant differences were seen for 6MWD between WF, NWF and HC (WF<NWF<HC). The ICC of the 6MWD was excellent, confirming the 6MWT was performed correctly and is therefore a good standardized test as suggested by Goldman et al. (2008) (25). Test-retest reliability of the DWI_{6-1} was good, as the ICC in the total MS group was high. As the ICC is sensitive to the total variance and spread in a group, the ICC of the DWI_{6-1} in the MS subgroups (i.e. WF and NWF) was lower. The SEM was calculated to indicate which difference in DWI_{6-1} is needed to be significantly changed. In general, after this study we could conclude that pwMS should improve their DWI_{6-1} with 5.10% after an intervention to be significant. However, the MDC is used to determine whether the change can be considered without measurement error(31). Hereby, it could be concluded that the MS patients needed to improve their DWI with 14.12% to be clinically relevant. The SEM (and therefore also the MDC) is based on the SD and ICC of the DWI_{6-1} and therefore lower in the NWF compared to the WF, as the SD in the NWF is lower. These lower SD in the NWF group compared to the WF group can be explained by the fact that the range of DWI_{6-1} percentages in the NWF is more limited, since even HC would not be able accelerate much (i.e. positive DWI_{6-1}), while the range of DWI_{6-1} percentages in WF can be much higher due to greater declines (i.e. more negative DWI_{6-1}) in more disabled patients. However, in general, larger sample sizes in the WF and NWF subgroup are required in order to minimize the variance and obtain better and more reliable results.

A review recently conducted in our research group (Van Geel et al., submission planned in June 2018) about the assessment of WF in any diseased population, reported the use of some other diagnostic formulas during the 6MWT to measure WF. In these formulas, the 6MWD was used as normalisation factor, while the DWI_{6-1} is normalised with the first minute of the 6MWT. Barbosa et al. (2016) calculated a deceleration index as follows: mean walking speed (MWS) over six minutes divided by the MWS in the first minute, divided by the total 6MWD and multiplied by 1000 (32). Murphy et al. used a formula based on Schnelle et al. wherein the MWS over six minutes is divided by the MWS over 2 minutes, divided by the total 6MWD and multiplied by 1000 (16, 33). However, both studies did not report a cut-off value for WF, nor any psychometric properties. Moreover, these studies were performed in elderly, not pwMS. Therefore, we opted for the use of the DWI_{6-1}, already used and investigated in previous research performed by our group.

4.3 Manifestation during 6MWT

4.3.1 Performance walking fatigability

As mentioned before, the $DWI_{6-1} \leq -10\%$ showed significant differences between WF, NWF and HC, indicating that the formula and cut-off value are good (Figure 11). Both WF and HC declined significantly between in the second minute of the 6MWT. The difference however is that HC will show a stagnation after the second minute and accelerate again in the last minute, while people with WF are not able to accelerate and will show a decline in walking distance every minute. This typical pattern of deceleration, stagnation and acceleration seen in the HC is normal according to literature. However, they do normally not fully return to their baseline walking distance of the first minute, which was also seen in this study as shown by the mostly slight negative DWI_{6-1} and is therefore a confirmation of proper implementation of the protocol.

Additionally, normalizing the distances every minute to minute one revealed longer walking test are necessary to measure WF. Significant differences in walking decline between NWF and HC compared to WF are only seen after the third and fourth minute (NWF and HC respectively) of the 6MWT (Figure 12). This is in line with findings in the beforementioned review of our research group about the assessment method for WF, wherein will be concluded long walking tests are preferred to differentiate between WF and NWF. Short walking tests are not sufficient as no clear deceleration will be found, whereas longer walking tests than 6 minutes or long walking test with a set distance would be too long and therefore give bias due to drop out (13-16). Additionally, Barbosa et al. stated that walking test at self-selected speed can give a lot variability in the first two minutes, suggesting again long walking test are necessary to measure performance fatigability (32).

4.3.2 Perceived walking fatigability

Significant differences were found for perceived fatigability assessed before and throughout the 6MWT between NWF and WF compared to HC (Figure 13). However, in order to differentiate between WF and NWF, three minutes of the walking test should be passed. This is again a confirmation for the need of long walking test for the patients to subjectively feel fatigued. Although, at minute six no significant difference was found between WF and NWF. This can be due to the large SE, suggesting larger sample sizes in order minimize variability.

Nevertheless, when normalizing the perceived fatigability VAS scores with baseline, no significant differences between all groups could be found. This indicates the increase in perceived fatigability throughout the walking test showed a similar pattern in WF, NWF and HC. Consequently, it can be concluded that de major differences in perceived fatigability are situated at baseline level, wherein the WF group experienced already more fatigue at rest.

Additionally, the differences found in perceived walking fatigability within and between the groups are not only significant, but also clinically relevant. Mean VAS scores increased from 0.25±0.65 at baseline to 2.5±1.93 at the end of the 6MWT in HC, which is considered as low.

In the NWF group, mean VAS scores increased from 2.00±1.95 to 4.76±2.87, which is considered moderate. The WF group showed an increase in experienced fatigue from 2.90±2.77 to 6.50±2.72, which is considered as more highly fatigued. These findings can be confirmed by a review of Loy et al. (2017), concluding that there is a significant relationship between perceived fatigue and fatigability in pwMS (5). However, both fatigue measures should be assessed independently as the correlations are considered medium and therefore probably not measuring the same construct.

4.4 Manifestation after 6MWT

To have an indication about the manifestation of WF by other symptoms, as well as the duration of these symptoms and therefore WF, some subjective and objective measures were assessed before and after the 6MWT and repeated every 10 minutes after the 6MWT until 30 minutes. It should be state that this testing procedure was not optimal, as performing the objective test (especially muscle strength assessment) every 10 minutes again, possibly no optimal recuperation could occur. However, these are rather simple and fast test which were needed in order to have an indication about the duration of the WF to subsequently know in the future which symptoms are related to WF and until how long after the inducement of the fatigability better test for related symptoms can be carried out.

4.4.1 Subjective symptoms

WF manifested itself in a significant increase in perceived general fatigue, motor fatigability, muscle weakness, spasticity, pain, dizziness and gait pattern impairments after the 6MWT (Figure 15).

Sensitivity, balance problems, visual disturbance and attention problems did not significantly differed from baseline in the WF group. Most of the symptoms returned back to baseline 10 minutes after the 6MWT, except for the general feeling of fatigue, which showed no longer a significant difference with baseline after 20 minutes. However, general fatigue and muscle weakness were also significantly increased in the NWF group, wherein general fatigue also persisted until 10 minutes after the 6MWT and muscle weakness even showed a significant increase according to baseline until 20 minutes after 6MWT. However, it should be stated that the overall experience of all subjective symptoms immediately after the 6MWT was higher in the WF compared to the NWF, except for visual disturbance and sensitivity. HC only showed a significant increase in perceived general fatigue and motor fatigability after the 6MWT, which was moreover a very small not clinically relevant difference, as mean VAS scores did not exceed two. Besides, it should be kept in mind that only the mean VAS scores for general fatigue, motor fatigability and gait pattern disturbance reached a moderate level of severity in WF after the 6MWT. Additionally, our research group is currently submitting another study wherein the SI is evaluated before and after a single maximal endurance test. Another study, performed by Skjerbaek et al. (2012) showed significant increases in the severity of perceived symptoms after a 30-minute resistance versus endurance training in heat-sensitive pwMS (27).

Differences in the increase of subjective symptoms after the 6MWT between WF and NWF, showed significantly higher increases in WF for motor fatigability, spasticity and dizziness (Figure 16). However, also in the NWF significant higher increases in general fatigue, sensitivity, gait pattern impairments and dizziness were found compared to the HC, suggesting that pwMS without WF feel more fatigued and already experience some symptoms without showing an objective decline in performance that differed from the HC.

In elderly, Schnelle et al. (2012) and Murphy et al. (2017) normalized the differences in subjective fatigability VAS scores by dividing it with the total walking distance, thereby correcting for the effort made (16, 33). This was of no relevance for our study, since in this study no corrections were made with the 6MWD for measuring performance fatigability, as was done in the studies of Schnelle et al. (2012) and Murphy et al. (2017). In this study, only percentages changes were investigated compared to minute one instead of the overall performance.

Besides, it should be kept in mind that the SE for all symptoms in the MS groups is large, due to a relative small sample size in the first place, but also to the typical large variability of symptom presence and severity in-between MS individuals.

4.4.2 Objective symptoms

In the NWF group, more significant decreases were found in muscle strength after the 6MWT compared to the HC, who only showed a significant decrease in the left knee extension. All significant decreases in muscle strength in both MS groups disappeared 10 minutes after the 6MWT, except for the muscle strength in the right knee extension in NWF, which stayed significantly lower until 10 minutes after the 6MWT. Reporting of muscle weakness through the SI also showed significant increases in muscle weakness, which seemed to be also more and longer present in the NWF group (Figure 15). For the measurements of spasticity, no objective significant differences are found in any group after the 6MWT. However, spasticity was reported in the SI as significantly higher post 6MWT compared to baseline in the WF group (Figure 15). In HC, balance significantly increased after the 6MWT, suggesting they are more focussed after performing a motor task.

When comparing the presence of spasticity, muscle strength declines and balance problems after the 6MWT between all groups, significantly more objective problems could be found in NWF and WF compared to HC. This confirmed differences seen in the subjective reported increase in spasticity and muscle weakness after the 6MWT in WF and NWF compared to HC, except for subjective spasticity between HC and NWF. However, subjective reporting of spasticity between WF and NWF after the 6MWT was significantly increased in the WF group, but this was not seen in the objective measurement. Besides, literature also objectively describes significant changes in kinematics and declines in muscle strength after the performance of the 6MWT (20).

In general, it should be however be kept in mind that these objective measurements are rather simple screening test to have a first indication about the presence and duration of possible symptoms in WF. The test were physician-based since muscle strength and spasticity were not objectively

measures but scored subjectively by the researchers via the MI and MAS respectively. Moreover, four different assessors were involved in the measurement of the participants, probably resulting in some variability between raters and thus patients, and less reliability of the objectiveness of the measurements.

4.5 Clinical profile walking fatigability

In order to have an indication about a clinical profile for WF, symptoms, fatigue and fatigue-related questionnaires, and some MS characteristics at baseline level were compared between groups.

4.5.1 Subjective symptoms

PwMS with WF experienced significant higher gait pattern impairment and muscle weakness compared to pwMS without WF (figure 9). All other symptoms did differ significantly between WF and HC, but between WF and NWF, suggesting again varying symptoms and symptoms severities are present among MS individuals.

Correlations between the DWI and subjective symptoms were significant for motor fatigability, sensitivity, gait pattern impairments, dizziness and muscle weakness (table 4). However, correlations seemed rather low, but it should be kept in mind that these are correlations between subjective assessments and an objective outcome measure. Consequently, these correlations can be considered as relevant. Especially gait pattern impairments and muscle weakness increased significantly in lower $DWI_{6\text{-}1}$ (i.e. more negative outcomes and thus more decline in walking distance) and therefore probably in more disabled persons with WF.

4.5.2 Objective symptoms

Muscle strength was significantly lower at baseline in WF and NWF compared to HC. This confirmed the subjective muscle weakness findings. However, objective lower muscle strength between WF and NWF was only present in the right leg. The objective and subjective assessment of balance did significantly differ between all groups (HC>NWF>WF). Spasticity at baseline was significantly more reported in WF compared to NWF. These findings were only objectively confirmed in the left triceps, suggesting most of the patients experienced spasticity in their triceps. However, no significant correlations were found between MAS and $DWI_{6\text{-}1}$, suggesting pwMS showing a greater decline in walking distance do not have more spasticity. Positive moderate correlations observed between the $DWI_{6\text{-}1}$ and the MIs of the right ankle dorsiflexion and hip flexion at baseline suggests that pwMS showing a greater decline in walking distance also have lower muscle strength. However this was only the case for two of the six tested muscles. A moderate positive correlation was found between the $DWI_{6\text{-}1}$ and the Romberg at baseline, suggesting pwMS showing a greater decline in walking distance have more balance problems.

4.5.3 Descriptive variables

The DWI_{6-1} showed the greatest correlations with the subjective reported impact of MS on walking ability (assessed through the MSWS-12), falls (assessed via the FES-I) and sleep problems (assessed via the SCI) (table 5). Again, the correlations are not high due to comparisons between subjective and objective assessment of fatigability and its possible related factors.

EDSS and motor function test (i.e. T25FW and NHPT) showed significant higher outcomes in WF compared to NWF, again confirming the use of the 6MWT with $DWI_{6-1} \leq -10\%$ as a good diagnostic criteria for WF. The fact that significant differences were found in upper extremity function between WF and NWF, suggests a relationship between upper and lower limb fatigability . This relationship needs to be further investigated, since until now our research group only investigated upper limb fatigability and its relationship with fatigue or perceived fatigability (12, 19, 34). However, a study by Schwid et al. (1999) showed that pwMS were weaker in lower limb muscle strength but not in upper limb strength compared to HC. Moreover, no significant associations were found between strength in upper and lower muscles and fatigue (15).

FS scores, the sub scores of the EDSS, showed no significant differences between WF and NWF. This could be due to large variability in symptoms and their severities in the overall MS population. Therefore larger sample sized would be needed in order to find possible correlations between WF and FS scores.

4.6 Cognitive fatigability

The percentage decline in PASAT and SDMT were calculated for the subsample of pwMS who participated in the third test session, wherein no significant differences could be found between groups. When calculating the % decline for both measures in the whole study population, the same conclusion could be drawn (Table 12).

Table 12: PASAT and SDMT decline in the whole MS study population. Data is represented as mean±SD. PASAT and SDMT declines were calculated based on the % decline in correct answers between the first 1/3 and the last 1/3 of the test.

	Total MS (n=48)	WF (n=24)	NWF (n=24)	p-value
PASAT % decline	-16.09±27.29	-20.67±27.07	-11.50±27.29	NS[a]
SDMT % decline	-6.95±21.43	-5.85±23.62	-7.99±19.60	NS[a]

[a] Mann-Whitney U test.

PASAT: paced auditory serial addition test, SDMT: symbol digit modality test, MS: multiple sclerosis, WF: walking fatigability, NWF: non-walking fatigability.

Harrison et al. (2017) published a review on the measurement methods for cognitive fatigability in MS (35). They concluded that the assessment methods varied al lot between studies and that findings are inconsistent between and within measures. However, they suggested the PASAT to be possibly the best and mostly used measure for cognitive fatigability by making comparisons between parts of the test.

The overall difference with our testing procedure was that most studies performed the test multiple times, which showed significant decreases in performance across the trials, which could not be evaluated in our study, since the decline was only measures once. Giglio et al. (2015) investigated the use of the SDMT to measure cognitive fatigability by calculating the decline in correct answers between two consecutive measurements (36). They found that pwMS showed greater declines than HC. Unfortunately, we did not include a control group in this study and in addition did not performed the test two times. Moreover, the assessment of the SDMT in this study was conducted manually. It would have been better to execute the measurement orally to avoid bias for upper limb fatigability.

The RTI_{6-1} calculates the decline in reaction time and therefore the more negative the outcomes are, the faster the participants responded at the end of the test compared to the beginning (i.e. low reaction time = fast response). However, the RTI_{6-1} did not show any differences between the single and dual task condition in all groups, whereby can be concluded that there is no significant increase in reaction time when the cognitive (i.e. vigilance) and motor (i.e. walking) task are performed simultaneously. This indicates therefore that the vigilance is not considered as a dual task when performed simultaneously with walking.

The same conclusion could be made for the DWI_{6-1} in the single and dual task condition. No significant decrease in walking distance was observed when participants had to combine a cognitive and motor task. Besides, no significant differences could be found for RTI_{6-1} or DWI_{6-1} between the groups for all three conditions, except for the DWI_{6-1} in the single walk condition. Here significant differences in DWI_{6-1} were found for HC-WF and WF-NWF, which just confirms the findings already mentioned in the previously discussed manifestation of WF.

The percentage decline in correct answered in PASAT and SDMT did not significantly differ between WF and NWF, also suggesting there is no relationship between cognitive fatigability and WF. However, HC did not perform this cognitive fatigability measures and therefore no comparisons could be made between pwMS and a control group.

These findings were also confirmed by the fact that no correlations were found between the PASAT decline, SDMT decline and RTI_{6-1} and DWI_{6-1} of both dual and single task conditions in MS (table 10). However, a moderate, but not significant, negative correlation was observed between the decline in SDMT and RTI_{6-1} in the vigilance single task, suggesting that pwMS who perform better on the vigilance task (i.e. low RTI_{6-1}) show more decline in the SDMT (i.e. decline in correct answers between end and beginning of the test), which is contradictory. However, comparing SDMT and Vigilance is difficult, because it assesses a different aspect of cognitive fatigability, being processing speed and sustained attention respectively. Better correlations would be expected between the PASAT and the vigilance, since they both measure sustained attention, but this was not the case.

When looking in more detail at the pattern of the walking distances and reaction times in each minute, no significant differences could be observed in general between groups, timepoints or test conditions. However, some trends in the data can be observed and therefore further investigations in greater sample sizes are recommended. Differences observed in walking distance between the groups is normal, because of the walking difficulties present in MS.

Lower reaction times were observed in HC in the VigilanceWalk condition compared to the single vigilance condition, indicating they respond faster to the target letters when simultaneously walking. Besides, it did not seem that this was compensated in slower walking. The reaction times in the NWF group seem to be higher (i.e. slower response rate) when they had to perform the task when simultaneously walking. Additionally, also some interesting trends can also be seen in the WF group. Declines in walking distance can be observed in the single task condition, but these declines are not observed when the patients had to combine the walking task with the cognitive task, suggesting they are more able to sustain their walking capacity when doing a cognitive and motor task at the same time. In the first minutes of the VigilanceWalk condition, a decline in reaction time (i.e. faster response) can be seen, which will stagnate after minute 3. When comparing this with the fact that the WF group shows an increase in their walking distance at minute 4, we can see a trend indicating that in the middle of the test, patients with WF are able to increase their walking speed without getting slower in their responses. However in the last minute, their response rate became slower, which was accompanied with a decrease in walking distance, suggesting a possible decrease in the dual performance. Moreover, a lower walking distance was observed in the first minute in the Vigilance Walk condition compared to the single walking condition, suggesting they showed some kind of different pacing strategy. A familiarisation trial for the VigilanceWalk is therefore recommended to eliminate possible effect of starting more slowly because they did not know what to expect.

As no significant differences in reaction time were found between both conditions, the vigilance is not considered as a dual task when performed during six minutes of walking, supporting the evidence already found in our research group indicating that the vigilance task during one minute of walking is not considered as a dual task cost. Nevertheless, it should be kept in mind that above mentioned trends in the data are not significant and in order to make any conclusions a greater sample size is needed.

5. Conclusion and future perspectives

The purpose of this study was threefold: First, to investigate test-retest reliability of the DWI_{6-1} as objective measurement to state WF. Additionally, the manifestation of WF during walking, as well as the manifestation of WF by other symptoms before and after the walking test were investigated. Finally, this study aimed to provide first insights in a clinical profile for WF.

By including a control group in our study, a reconsideration of the previously stated cut-off value of -15% could be made. It could be concluded that a $DWI_{6-1} \leq -10\%$ was already enough to measure WF. This DWI_{6-1}, calculated by the performance of the 6MWT with a cut-off value of -10%, showed good test-retest reliability in the total MS group. However, larger sample sizes are recommended to confirm this in the WF and NWF subgroup, as the ICC in these subgroups was considered moderate.

Only the last minutes of the 6MWT showed significant differences between the groups in the walking distance decline compared to minute 1, which illustrates longer walking tests are recommended to measure WF. Additionally, these longer walking test are also needed to show significant differences in perceived fatigability between WF and NWF.

Half of the MS patients in our study population showed WF, where it significantly manifested in greater subjective increases in gait impairments, spasticity and dizziness compared to the NWF group. However, no significant difference was objectively observed for spasticity between WF and NWF after the 6MWT. Objectively, a lower muscle strength was observed in some muscle of patients with WF compared to patients without, but strength did not decline during the 6MWT. Most of the symptoms returned to baseline 10 minutes after the 6MWT.

Balance problems were objectively significantly more present at baseline in WF compared to NWF. Additionally pwMS who show WF have a lower baseline muscle strength compared to pwMS without WF. Spasticity at baseline was also considered higher in WF compared to NWF. Subjectively, significant more gait pattern impairments and muscle weakness was reported in the WF group compared to NWF. These subjective and objective symptoms showed low to moderate correlations with the DWI_{6-1}, which is considered relevant as comparisons are being made between subjective symptoms and an objectively measures outcome parameter.

In terms of the clinical profile, also significant differences were seen between WF and NWF for EDSS and upper and lower limb motor functions. The physical component of fatigue questionnaires (MFIS and FSMC) was significantly higher in WF, as well as the walking problems reported via the MSWS-12. Moreover, moderate significant correlations with the DWI were observed for the FES-I, SCI and MSWS-12, indicating a relationship of WF with self-reported falls, sleep and walking problems respectively.

Beside investigating the assessment and manifestation of WF, a pilot study was conducted to investigate a possible relationship between WF and cognitive fatigability. Indications towards a protective effect of the vigilance task to declines in walking distance were seen. Therefore, the cognitive task seems to be a facilitator to improve attention rather than being a disturbing factor. However, no significant conclusion could be drawn as the sample size was too small.

In general, future research is recommended in a larger sample size and with better objective testing in order to further investigate the clinical profile of WF, as well as its underlying and related factors. Balance problems, gait impairments and muscle weakness were indicated in the WF group and therefore require further investigation. The overall hypothesis is that both central and peripheral factors are involved in WF. Balance problems can be further investigated in terms of the central factor underlying WF, which can be done directly by measuring voluntary neural drive, motor neural integrity and conduction velocity, as it is known that demyelination in MS is associated with axonal loss (37-41). This measures will give more insights in the signal transduction throughout the nerves, as well as the ability to voluntary activate the muscles. Beside these central factor theory, also underlying peripheral factors could be involved in muscle weakness and therefore possibly WF. This can be further investigated by objectively measuring muscle strength by the static and dynamic fatigue index and a having a closer look into the muscle fibers by taking muscle biopsies (42). Gait impairments should be further investigated by using specialized treadmills or camera systems able to record and evaluate gait patterns in more detail (20). Related internal and external factors of WF can be investigated in more detail by examining the upper and lower limb functioning, thereby investigating associations between upper limb and lower limb fatigability. Using an activity tracker would be interesting to more objectively record their physical activity. More cognitive test assessing more cognitive domains (e.g. working memory, processing speed, sustained attention) should be included in further testing, to investigate a possible relation between WF and cognitive function. As ultimate goal, a rehabilitation program can be set up, focusing on either peripheral or central factors or both, in order to treat WF.

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Appendices

Appendix A: Descriptive measures

A.1 Expanded Disability System Score

Kurtzke Expanded Disability Status Scale (EDSS)

- ❑ 0.0 - Normal neurological exam (all grade 0 in all Functional System (FS) scores*).
- ❑ 1.0 - No disability, minimal signs in one FS* (i.e., grade 1).
- ❑ 1.5 - No disability, minimal signs in more than one FS* (more than 1 FS grade 1).
- ❑ 2.0 - Minimal disability in one FS (one FS grade 2, others 0 or 1).
- ❑ 2.5 - Minimal disability in two FS (two FS grade 2, others 0 or 1).
- ❑ 3.0 - Moderate disability in one FS (one FS grade 3, others 0 or 1) or mild disability in three or four FS (three or four FS grade 2, others 0 or 1) though fully ambulatory.
- ❑ 3.5 - Fully ambulatory but with moderate disability in one FS (one grade 3) and one or two FS grade 2; or two FS grade 3 (others 0 or 1) or five grade 2 (others 0 or 1).
- ❑ 4.0 - Fully ambulatory without aid, self-sufficient, up and about some 12 hours a day despite relatively severe disability consisting of one FS grade 4 (others 0 or 1), or combination of lesser grades exceeding limits of previous steps; able to walk without aid or rest some 500 meters.
- ❑ 4.5 - Fully ambulatory without aid, up and about much of the day, able to work a full day, may otherwise have some limitation of full activity or require minimal assistance; characterized by relatively severe disability usually consisting of one FS grade 4 (others or 1) or combinations of lesser grades exceeding limits of previous steps; able to walk without aid or rest some 300 meters.
- ❑ 5.0 - Ambulatory without aid or rest for about 200 meters; disability severe enough to impair full daily activities (e.g., to work a full day without special provisions); (Usual FS equivalents are one grade 5 alone, others 0 or 1; or combinations of lesser grades usually exceeding specifications for step 4.0).
- ❑ 5.5 - Ambulatory without aid for about 100 meters; disability severe enough to preclude full daily activities; (Usual FS equivalents are one grade 5 alone, others 0 or 1; or combination of lesser grades usually exceeding those for step 4.0).
- ❑ 6.0 - Intermittent or unilateral constant assistance (cane, crutch, brace) required to walk about 100 meters with or without resting; (Usual FS equivalents are combinations with more than two FS grade 3+).

- ❑ 6.5 - Constant bilateral assistance (canes, crutches, braces) required to walk about 20 meters without resting; (Usual FS equivalents are combinations with more than two FS grade 3+).
- ❑ 7.0 - Unable to walk beyond approximately 5 meters even with aid, essentially restricted to wheelchair; wheels self in standard wheelchair and transfers alone; up and about in wheelchair some 12 hours a day; (Usual FS equivalents are combinations with more than one FS grade 4+; very rarely pyramidal grade 5 alone).
- ❑ 7.5 - Unable to take more than a few steps; restricted to wheelchair; may need aid in transfer; wheels self but cannot carry on in standard wheelchair a full day; May require motorized wheelchair; (Usual FS equivalents are combinations with more than one FS grade 4+).
- ❑ 8.0 - Essentially restricted to bed or chair or perambulated in wheelchair, but may be out of bed itself much of the day; retains many self-care functions; generally has effective use of arms; (Usual FS equivalents are combinations, generally grade 4+ in several systems).
- ❑ 8.5 - Essentially restricted to bed much of day; has some effective use of arm(s); retains some self-care functions; (Usual FS equivalents are combinations, generally 4+ in several systems).
- ❑ 9.0 - Helpless bed patient; can communicate and eat; (Usual FS equivalents are combinations, mostly grade 4+).
- ❑ 9.5 - Totally helpless bed patient; unable to communicate effectively or eat/swallow; (Usual FS equivalents are combinations, almost all grade 4+).
- ❑ 10.0 - Death due to MS.

*Excludes cerebral function grade 1.

A.2 Functional System Score

Kurtzke Functional Systems Scores (FSS)

Pyramidal Functions
0 - Normal
1 - Abnormal signs without disability
2 - Minimal disability
3 - Mild to moderate paraparesis or hemiparesis (detectable weakness but most function sustained for short periods, fatigue a problem); severe monoparesis (almost no function)
4 - Marked paraparesis or hemiparesis (function is difficult), moderate quadriparesis (function is decreased but can be sustained for short periods); or monoplegia
5 - Paraplegia, hemiplegia, or marked quadriparesis
6 - Quadriplegia
9 - (Unknown)

Cerebellar Functions
0 - Normal
1 - Abnormal signs without disability
2 - Mild ataxia (tremor or clumsy movements easily seen, minor interference with function)
3 - Moderate truncal or limb ataxia (tremor or clumsy movements interfere with function in all shpheres)
4 - Severe ataxia in all limbs (most function is very difficult)
5 - Unable to perform coordinated movements due to ataxia
9 - (Unknown)

Record #1 in small box when weakness (grade 3 or worse on pyramidal) interferes with testing.

Brainstem Functions
0 - Normal
1 - Signs only
2 - Moderate nystagmus or other mild disability
3 - Severe nystagmus, marked extraocular weakness, or moderate disability of other cranial nerves
4 - Marked dysarthria or other marked disability
5 - Inability to swallow or speak
9 - (Unknown)

Sensory Function
0 - Normal
1 - Vibration or figure-writing decrease only in one or two limbs
2 - Mild decrease in touch or pain or position sense, and/or moderate decrease in vibration in one or two limbs; or vibratory (c/s figure writing) decrease alone in three or four limbs
3 - Moderate decrease in touch or pain or position sense, and/or essentially lost vibration in one or two limbs; or mild decrease in touch or pain and/or moderate decrease in all proprioceptive tests in three or four limbs
4 - Marked decrease in touch or pain or loss of proprioception, alone or combined, in one or two limbs; or moderate decrease in touch or pain and/or severe proprioceptive decrease in more than two limbs
5 - Loss (essentially) of sensation in one or two limbs; or moderate decrease in touch or pain and/or loss of proprioception for most of the body below the head
6 - Sensation essentially lost below the head
9 - (Unknown)

Visual Function
0 - Normal
1 - Scotoma with visual acuity (corrected) better than 20/30
2 - Worse eye with scotoma with maximal visual acuity (corrected) of 20/30–20/59
3 - Worse eye with large scotoma, or moderate decrease in fields, but with maximal visual acuity (corrected) of 20/60–20/99
4 - Worse eye with marked decrease of fields and maximal visual acuity (corrected) of 20/100–20/200; grade 3 plus maximal acuity of better eye of 20/60 or less
5 - Worse eye with maximal visual acuity (corrected) less than 20/200; grade 4 plus maximal acuity of better eye of 20/60 or less
6 - Grade 5 plus maximal visual acuity of better eye of 20/60 or less
9 - (Unknown)

Record #1 in small box for presence of temporal pallor

Bowel and Bladder Function
(Rate on the basis of the worse function, either bowel or bladder)
0 - Normal
1 - Mild urinary hesitance, urgency, or retention
2 - Moderate hesitance, urgency, retention of bowel or bladder, or rare urinary incontinence (intermittent self-catheterization, manual compression to evacuate bladder, or finger evacuation of stool)
3 - Frequent urinary incontinence
4 - In need of almost constant catheterization (and constant use of measures to evacuate stool)
5 - Loss of bladder function
6 - Loss of bowel and bladder function
9 - (Unknown)

Cerebral (or Mental) Functions
0 - Normal
1 - Mood alteration only (does not affect EDSS score)
2 - Mild decrease in mentation
3 - Moderate decrease in mentation
4 - Marked decrease in mentation (chronic brain syndrome – moderate)
5 - Dementia or chronic brain syndrome – severe or incompetent
9 - (Unknown)

A.3 Timed 25 foot Walk

LOWER EXTREMITY FUNCTION: TIMED 25-FOOT WALK

Subject ID Number | Subject Initials | Visit Date: Day Month Year

TIMED 25-FOOT WALK

Did patient wear an AFO? ☐ Yes ☐ No

Was assistive device used? ☐ Yes ☐ No

Assistive device used *(mark one)*:
☐ Unilateral Assistance ☐ Cane ☐ Crutch
☐ Bilateral Assistance ☐ Cane ☐ Crutch ☐ Walker/Rollator

Trial 1

Time for 25-Foot Walk ☐☐☐☐☐ ☐ seconds

For a complete trial, record any circumstances that affected the patient's performance:

If trial was not completed *(mark one)*: Specify:
☐ Unable to complete trial due to physical limitations ➡
☐ Other ➡

Trial 2

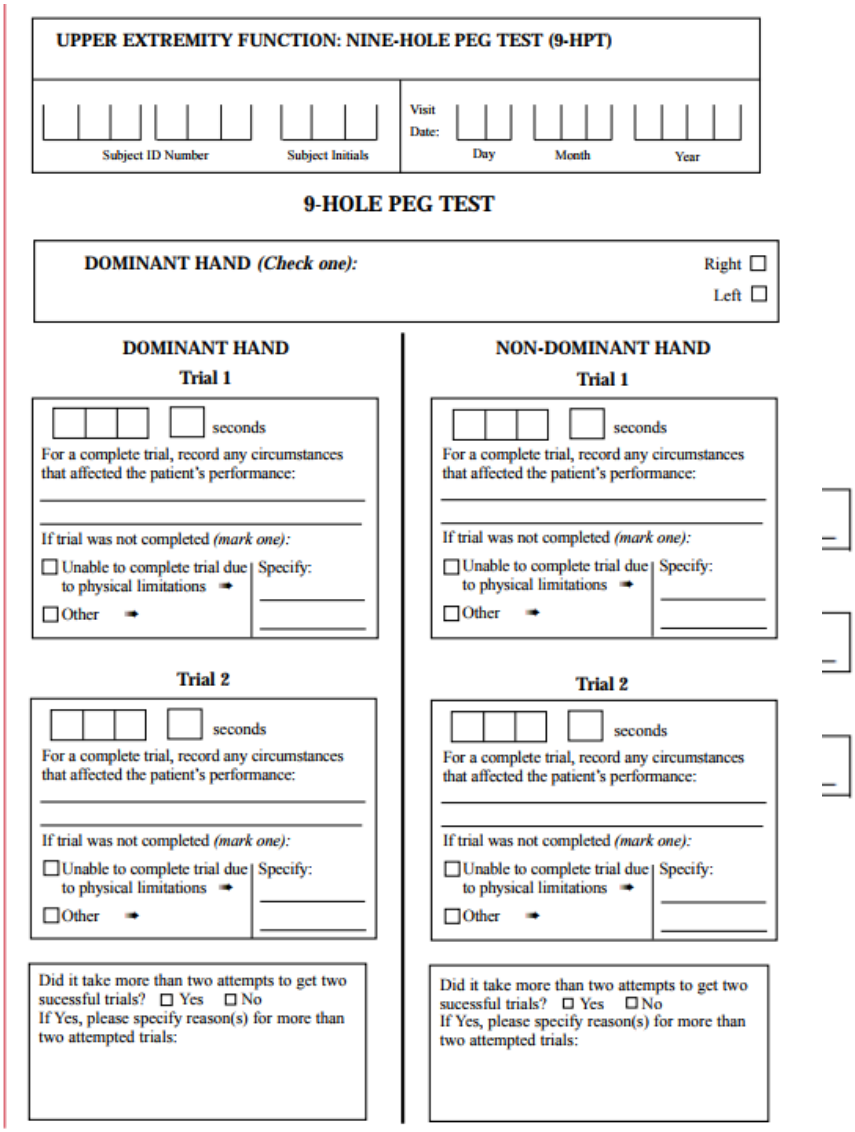
Time for 25-Foot Walk ☐☐☐☐☐ ☐ seconds

For a complete trial, record any circumstances that affected the patient's performance:

If trial was not completed *(mark one)*: Specify:
☐ Unable to complete trial due to physical limitations ➡
☐ Other ➡

Did it take more than two attempts to get two successful trials? ☐ Yes ☐ No
If yes, please specify reasons(s) for more than two attempted trials:

A.4 Nine Hole Peg Test



UPPER EXTREMITY FUNCTION: NINE-HOLE PEG TEST (9-HPT)

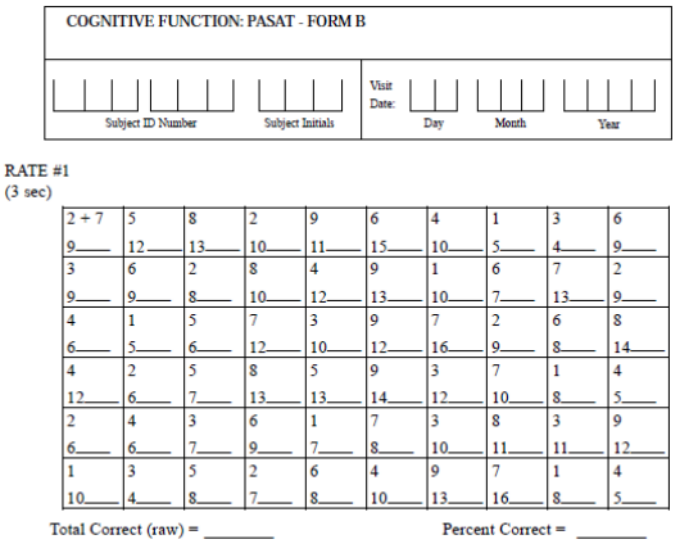
Subject ID Number | Subject Initials | Visit Date: Day | Month | Year

9-HOLE PEG TEST

DOMINANT HAND *(Check one):* Right ☐ Left ☐

DOMINANT HAND	NON-DOMINANT HAND
Trial 1	**Trial 1**
☐☐☐ ☐ seconds	☐☐☐ ☐ seconds
For a complete trial, record any circumstances that affected the patient's performance:	For a complete trial, record any circumstances that affected the patient's performance:
If trial was not completed *(mark one):*	If trial was not completed *(mark one):*
☐ Unable to complete trial due to physical limitations ➡ Specify:	☐ Unable to complete trial due to physical limitations ➡ Specify:
☐ Other ➡	☐ Other ➡
Trial 2	**Trial 2**
☐☐☐ ☐ seconds	☐☐☐ ☐ seconds
For a complete trial, record any circumstances that affected the patient's performance:	For a complete trial, record any circumstances that affected the patient's performance:
If trial was not completed *(mark one):*	If trial was not completed *(mark one):*
☐ Unable to complete trial due to physical limitations ➡ Specify:	☐ Unable to complete trial due to physical limitations ➡ Specify:
☐ Other ➡	☐ Other ➡
Did it take more than two attempts to get two sucessful trials? ☐ Yes ☐ No If Yes, please specify reason(s) for more than two attempted trials:	Did it take more than two attempts to get two sucessful trials? ☐ Yes ☐ No If Yes, please specify reason(s) for more than two attempted trials:

A.5 Paced Auditory Serial Addition test



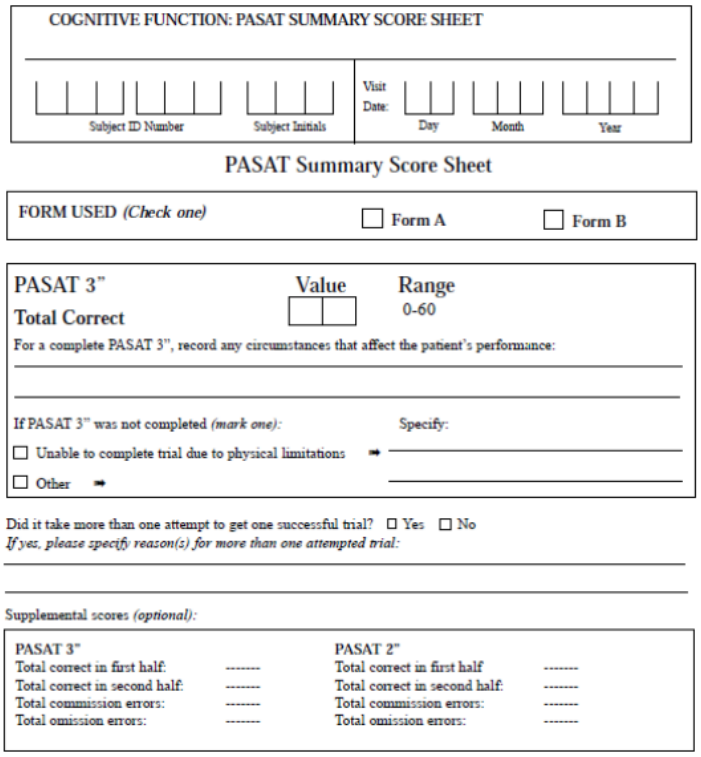
COGNITIVE FUNCTION: PASAT - FORM B

Subject ID Number | Subject Initials | Visit Date: Day | Month | Year

RATE #1
(3 sec)

2 + 7 9	5 12	8 13	2 10	9 11	6 15	4 10	1 5	3 4	6 9
3 9	6 9	2 8	8 10	4 12	9 13	1 10	6 7	7 13	2 9
4 6	1 5	5 6	7 12	3 10	9 12	7 16	2 9	6 8	8 14
4 12	2 6	5 7	8 13	5 13	9 14	3 12	7 10	1 8	4 5
2 6	4 6	3 7	6 9	1 7	7 8	3 10	8 11	3 11	9 12
1 10	3 4	5 8	2 7	6 8	4 10	9 13	7 16	1 8	4 5

Total Correct (raw) = _____ Percent Correct = _____



COGNITIVE FUNCTION: PASAT SUMMARY SCORE SHEET

Subject ID Number | Subject Initials | Visit Date: Day Month Year

PASAT Summary Score Sheet

FORM USED *(Check one)* ☐ Form A ☐ Form B

PASAT 3" Total Correct — Value ☐☐ Range 0-60

For a complete PASAT 3", record any circumstances that affect the patient's performance:

If PASAT 3" was not completed *(mark one)*: Specify:

☐ Unable to complete trial due to physical limitations ➡

☐ Other ➡

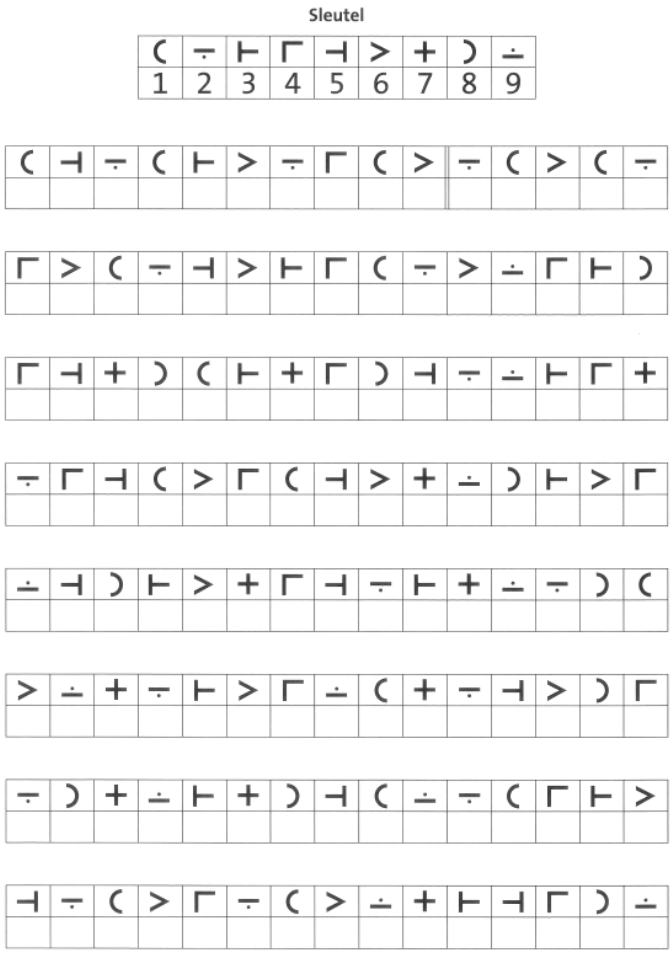
Did it take more than one attempt to get one successful trial? ☐ Yes ☐ No

If yes, please specify reason(s) for more than one attempted trial:

Supplemental scores *(optional)*:

PASAT 3"		PASAT 2"	
Total correct in first half:	-------	Total correct in first half	-------
Total correct in second half:	-------	Total correct in second half:	-------
Total commission errors:	-------	Total commission errors:	-------
Total omission errors:	-------	Total omission errors:	-------

A.6 Symbol Digit Modality Test



Appendix B: Questionnaires

B.1 Modified Fatigue Impact Scale

Dit is een korte vragenlijst (standaard 21-item vragenlijst) om de impact van algemene vermoeidheid in kaart te brengen. Deze vragenlijst beoordeelt de effecten van vermoeidheid op 3 verschillende niveaus: fysiek, cognitief en psychosociaal.

Score instructies:

- Nooit = 0
- Zelden = 1
- Soms = 2
- Vaak = 3
- Bijna altijd = 4

De scores worden bepaald door de punten van onderstaande vragen op te tellen:

- Fysieke subschaal (F) (0-36) = 4+6+7+10+13+14+17+20+21
- Cognitieve subschaal (C) (0-40) = 1+2+3+5+11+12+15+16+18+19
- Psychosociale subschaal (P) (0-8) = 8+9
- Totale score (0-84) = Som van alle punten

➔ **Evaluatie:** Totaalscore > 38 = MS-gerelateerde vermoeidheid

Omwille van mijn vermoeidheid (gedurende de laatste 4 weken) …		Nooit	Zelden	Soms	Vaak	Bijna altijd
C 1	ben ik minder aandachtig geweest					
C 2	heb ik moeite gehad om me lange tijd te concentreren					
C 3	ben ik niet in staat geweest om helder te denken					
F 4	ben ik onhandig geweest en had ik coördinatieproblemen					
C 5	ben ik vergeetachtig geweest					
F 6	heb ik mijn fysieke activiteiten trager moeten uitvoeren					
F 7	ben ik minder gemotiveerd geweest om fysieke activiteiten uit te voeren					
P 8	ben ik minder gemotiveerd geweest om aan sociale activiteiten deel te nemen					
P 9	ben ik beperkt geweest in de mogelijkheid om dingen buitenshuis te doen					
F 10	heb ik moeite gehad om fysieke inspanningen voor langere tijd vol te houden					
C 11	heb ik moeite gehad om beslissingen te nemen					
C 12	ben ik minder gemotiveerd geweest om iets te doen waarbij ik moest nadenken					
F 13	voelden mijn spieren zwak aan					
F 14	voelde ik mij fysiek niet goed					
C 15	heb ik moeite gehad om taken af te werken waarbij ik moest nadenken					
C 16	heb ik moeite gehad om mijn gedachten te ordenen bij taken thuis of op het werk					
F 17	ben ik minder in staat geweest om taken af te werken die fysieke inspanning vragen					
C 18	is mijn gedachtengang vertraagd geweest					
C 19	heb ik moeite gehad me te concentreren					
F 20	heb ik mijn fysieke activiteiten beperkt					
F 21	heb ik vaker of langer moeten rusten					

B.2 Fatigue Severity Scale

Dit is een korte vragenlijst om de ernst van algemene vermoeidheid in kaart te brengen.

Score instructies:

- 7-puntenschaal van 1 (helemaal oneens) tot 7 (helemaal eens)
- Totaalscore wordt berekend door de scores per item op te tellen

➔ **Evaluatie:** Totaalscore delen door 9 → Hoe hoger de score des te groter is de vermoeidheid/de impact van vermoeidheid op het dagelijks leven.

Score ≥ 4 = matige tot hoge vermoeidheid

Fatigue Severity Scale (FSS)

1. Als ik moe ben, ben ik minder gemotiveerd.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

2. Mijn vermoeidheid wordt opgeroepen door lichaamsbeweging.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

3. Ik ben snel vermoeid.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

4. Moeheid belemmert mijn lichamelijk functioneren.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

5. Mijn moeheid zorgt vaak voor problemen.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

6. Langdurig lichamelijk inspannen kan ik niet door de vermoeidheid.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

7. Moeheid belemmert mij bij het uitvoeren van bepaalde taken en verantwoordelijkheden.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

8. Van de klachten die mij het meest hinderen, is vermoeidheid één van de drie ergste.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

9. Bij mijn werk, gezinsleven of sociale contacten word ik belemmerd door mijn vermoeidheid.

Helemaal mee oneens ☐☐☐☐☐☐☐ Helemaal mee eens

B.3 Fatigue Scale for Motor and Cognitive functions

Dit is een korte vragenlijst die dieper ingaat op cognitieve en motorische vermoeibaarheid.

Score instructies:

- 5-puntenschaal van 1 (helemaal niet toepasselijk) tot 5 (volledig toepasselijk)

De scores worden bepaald door de punten van onderstaande vragen op te tellen:

- Mentale subschaal = 1+4+7+8+11+13+15+17+18+20
- Fysieke subschaal = 2+3+5+6+9+10+12+14+16+19

➔ Evaluatie:

FSMC totaal	≥ 43	light vermoeidheid
	≥ 53	gematigde vermoeidheid
	≥ 63	ernstige vermoeidheid
FSMC cognitief	≥ 22	Milde cognitieve vermoeidheid
	≥ 28	Matige Cognitive Vermoeidheid
	≥ 34	Ernstige cognitieve vermoeidheid
FSMC motor	≥ 22	Lichte motor Vermoeidheid
	≥ 27	Matige motor Vermoeidheid
	≥ 32	Zware motor Vermoeidheid

FSMC

Datum:_____

Fatigue schaal voor motorische en cognitieve functies ID:_____

Initialen: _____

Leeftijd: _____ Geslacht: m ○ v ○

Instructies

In de volgende vragenlijst gaat het om alledaagse problemen, die in direct verband staan met een extreme vorm van vermoeidheid (fatigue). Onder deze extreme vorm van vermoeidheid verstaan wij een overweldigende lethargie, uitputting, een gebrek aan energie, een toestand die plots optreedt, onafhankelijk van duidelijke externe oorzaken. Daarmee worden niet de afzonderlijke perioden van vermoeidheid bedoeld die elk mens ervaart in de loop van de dag, na inspanning of een slapeloze nacht.

Lees elke uitspraak zorgvuldig door en beslis dan, *in hoeverre* die bepaalde uitspraak op u en uw alledaagse leven toepasselijk is. Probeer uw antwoord onafhankelijk van uw huidige toestand te geven, maar probeer ons een beeld te geven van uw toestand zoals u die dag in dag uit beleeft. Kruis daartoe de bijbehorende cirkel aan (slechts één kruisje per uitspraak, graag).

	Helemaal niet toepasselijk	Niet erg toepasselijk	Enigszins toepasselijk	Erg toepasselijk	Volledig toepasselijk
1. Wanneer ik me langere tijd concentreer, raak ik sneller uitgeput dan andere mensen van mijn leeftijd.	○	○	○	○	○
2. Wanneer ik mij uitgeput voel, worden mijn bewegingen duidelijk onhandiger en minder gecoördineerd.	○	○	○	○	○
3. Vanwege mijn uitputtingstoestanden heb ik nu vaker een pauze en/of een langere pauze nodig dan vroeger tijdens lichamelijke activiteit.	○	○	○	○	○
4. Wanneer ik mij uitgeput voel, ben ik niet in staat beslissingen te nemen.	○	○	○	○	○
5. Wanneer ik met stresssituaties geconfronteerd word, voel ik me nu lichamelijk sneller uitgeput dan vroeger.	○	○	○	○	○
6. Vanwege mijn uitputtingstoestanden heb ik nu aanzienlijk minder sociale contacten dan vroeger.	○	○	○	○	○
7. Vanwege mijn uitputtingstoestanden valt het me nu moeilijker iets nieuws te leren dan vroeger.	○	○	○	○	○

Zie ommezijde

SMC

	Helemaal niet toepasselijk	Niet erg toepasselijk	Enigszins toepasselijk	Erg toepasselijk	Volledig toepasselijk
8. De eisen van mijn werk putten me geestelijk sneller uit dan vroeger.	○	○	○	○	○
9. Ik voel mijn uitputting vooral sterk in mijn spieren.	○	○	○	○	○
10. Ik heb niet langer het uithoudingsvermogen van voorheen voor langdurige lichamelijke inspanning.	○	○	○	○	○
11. Bij stress neemt mijn concentratievermogen aanzienlijk af.	○	○	○	○	○
12. Wanneer ik mij uitgeput voel, ben ik minder gemotiveerd dan andere mensen om activiteiten aan te pakken waaraan lichamelijke inspanning verbonden is.	○	○	○	○	○
13. Wanneer het heet is, vertraagt mijn denkvermogen steeds verder.	○	○	○	○	○
14. Wanneer ik mij uitgeput voel, worden mijn bewegingen duidelijk trager	○	○	○	○	○
15. Vanwege de uitputtingstoestanden heb ik minder zin dan vroeger om iets te doen, waar ik mijn aandacht bij moet houden.	○	○	○	○	○
16. Wanneer ik in een uitputtingstoestand raak, ben ik gewoon niet meer in staat snel te reageren.	○	○	○	○	○
17. Wanneer ik mij uitgeput voel, kan ik niet op bepaalde woorden komen.	○	○	○	○	○
18. Wanneer ik mij uitgeput voel, verslapt mijn aandacht aanzienlijk sneller dan vroeger.	○	○	○	○	○
19. Wanneer het heet is, voel ik me vooral lichamelijk extreem zwak en futloos.	○	○	○	○	○
20. Wanneer ik mij uitgeput voel, neemt mijn vergeetachtigheid duidelijk toe.	○	○	○	○	○

Controleer alstublieft of u uw initialen, uw leeftijd en uw geslacht op pagina 1 opgegeven heeft en dat u bij elke uitspraak een kruisje geplaatst heeft. Hartelijk dank.

B.4 Multiple Sclerosis Walking Scale

Dit is een korte vragenlijst om de algemene wandelproblemen in het dagelijkse leven in kaart te brengen.

Score-instructies:

- Scores per vraag optellen

Gedurende de afgelopen twee weken, in welke mate heeft de MS …..	Helemaal niet	Een beetje	Matig	Tamelijk veel	Heel erg
1. U beperkt in uw mogelijkheid te lopen?	1	2	3	4	5
2. U beperkt in uw mogelijkheden te rennen?	1	2	3	4	5
3. U beperkt in uw mogelijkheden de trap op en af te gaan?	1	2	3	4	5
4. Het u moeilijker gemaakt om te staan terwijl u dingen deed?	1	2	3	4	5
5. U beperkt in uw balans als u stond of liep?	1	2	3	4	5
6. U beperkt in hoe ver u kon lopen?	1	2	3	4	5
7. Ervoor gezorgd dat lopen u meer moeite kostte?	1	2	3	4	5
8. Het noodzakelijk gemaakt dat u steun gebruikte bij het lopen in huis (b.v. vastpakken van meubels of gebruik van een stok, etc)?	1	2	3	4	5
9. Het noodzakelijk gemaakt dat u steun gebruikte bij het lopen buitenshuis (b.v. gebruik van een stok of looprekje, etc)?	1	2	3	4	5
10. Ervoor gezorgd dat u langzamer ging lopen?	1	2	3	4	5
11. Invloed gehad op hoe soepel u liep?	1	2	3	4	5
12. Ervoor gezorgd dat u zich moest concentreren op het lopen?	1	2	3	4	5

Controleert u alstublieft of u bij **ALLE** vragen **ÉÉN** cijfer heeft omcirkeld.

B.5 Hospital Anxiety And Depression Scale

Dit is een korte vragenlijst om de symptomen van angst en depressie in het dagelijkse leven in kaart te brengen. Het meet de gevoelens en klachten die bij de patiënt de afgelopen week het meest aanwezig zijn geweest. De vragenlijst kan de aanwezigheid van depressie en angst uitsluiten, maar niet vaststellen.

Score instructies:

- 4-puntenschaal van 0 tot 3

De scores worden bepaald door de punten van onderstaande vragen op te tellen:

- Depressie schaal = 2+4+6+8+10+12+14
- Angst schaal = 1+3+5+7+9+11+13

➔ **Evaluatie:** score ≥ 11 = vermoedelijke angst- of depressiestoornis; score [8-10] = mogelijke angst- of depressiestoornis (alertheid)

Vragen:		
1. Ik voel me gespannen	3 ☐ 2 ☐ 1 ☐ 0 ☐	meestal vaak af en toe, soms helemaal niet
2. Ik geniet nog steeds van de dingen waar ik vroeger van genoot	0 ☐ 1 ☐ 2 ☐ 3 ☐	zeker zo veel wel wat minder duidelijk minder eigenlijk nauwelijks nog
3. Ik heb een soort angstgevoel alsof er iets vreselijks zal gebeuren	3 ☐ 2 ☐ 1 ☐ 0 ☐	jazeker, en vrij erg ja, maar niet zo erg een beetje, maar het hindert me niet helemaal niet
4. Ik kan best lachen en de dingen van de vrolijke kant zien	0 ☐ 1 ☐ 2 ☐ 3 ☐	net zoveel als vroeger nu wel wat minder duidelijk minder helemaal niet
5. Ik maak me ongerust	3 ☐ 2 ☐ 1 ☐ 0 ☐	heel erg vaak vaak af en toe, maar niet zo vaak heel soms
6. Ik voel me opgewekt	3 ☐ 2 ☐ 1 ☐ 0 ☐	helemaal niet heel af en toe soms meestal
7. Ik kan best rustig zitten en me ontspannen	0 ☐ 1 ☐ 2 ☐ 3 ☐	jazeker meestal af en toe helemaal niet
8. Ik heb het gevoel dat alles moeizaam gaat	3 ☐ 2 ☐ 1 ☐ 0 ☐	bijna altijd heel vaak soms helemaal niet
9. Ik heb een soort angstig, gespannen gevoel in mijn buik	0 ☐ 1 ☐ 2 ☐ 3 ☐	helemaal niet soms vrij vaak heel vaak
10. Het interesseert me niet meer hoe ik eruit zie	3 ☐ 2 ☐ 1 ☐ 0 ☐	inderdaad, helemaal niet meer niet meer zoveel als eigenlijk zou moeten het interesseert me wel, maar minder dan vroeger het interesseert me nog net zoveel als vroeger
11. Ik ben onrustig en voel dat ik iets te doen moet hebben	3 ☐ 2 ☐ 1 ☐ 0 ☐	inderdaad, heel duidelijk duidelijk enigszins helemaal niet
12. Ik verheug me van tevoren op dingen die komen gaan	0 ☐ 1 ☐ 2 ☐ 3 ☐	net zoveel als vroeger een beetje minder dan vroeger veel minder dan vroeger bijna nooit
13. Ik raak plotseling in paniek	3 ☐ 2 ☐ 1 ☐ 0 ☐	inderdaad, zeer vaak tamelijk vaak soms helemaal nooit
14. Ik kan van een goed boek genieten, of van een radio- en televisieprogramma	0 ☐ 1 ☐ 2 ☐ 3 ☐	vaak tamelijk vaak af en toe heel zelden

B.6 The Sleep Condition Indicator

Dit is een korte vragenlijst die dieper ingaat op het slaappatroon en eventuele slaapproblemen.

Score instructies:

- 5-puntenschaal van 0 tot 4
- Totale SCI = som van de scores per vraag

➔ Evaluatie:

- Scores kunnen gedeeld worden door 3.2 om een score van 0 tot 10 te bekomen. → Hogere score = betere slaap
- Item scores in grijze zone = threshold voor insomnia (slapeloosheid)

	Score				
Item	4	3	2	1	0
Thinking about a typical night in the last month … 1. … how long does it take you to fall asleep?	0 – 15 min	16 – 30 min	31 – 45 min	46 – 60 min	≥ 61 min
2. … if you then wake up during the night … how long are you awake for in total? (add all the wakenings up)	0 – 15 min	16 – 30 min	31 – 45 min	46 – 60 min	≥ 61 min
3. … how many nights a week do you have a problem with your sleep?	0 - 1	2	3	4	5 - 7
4. … how would you rate your sleep quality?	Very good	Good	Average	Poor	Very poor
Thinking about the past month, to what extent has poor sleep … 5. … affected your mood, energy, or relationships?	Not at all	A little	Somewhat	Much	Very much
6. … affected your concentration, productivity, or ability to stay awake	Not at all	A little	Somewhat	Much	Very much
7. … troubled you in general	Not at all	A little	Somewhat	Much	Very much
Finally … 8. … how long have you had a problem with your sleep?	I don't have a problem / < 1 mo	1 – 2 mo	3 – 6 mo	7 – 12 mo	> 1 yr

B.7 Falls Efficacy Scale International

Dit is een korte vragenlijst die betrekking heeft op valincidenten en eventuele bezorgdheid hierover.

Score instructies:

- 4-puntenschaal → scores optellen → Hoge score = grote valangst

→ **Evaluatie:**

- Score 16-19: Personen zijn weinig bezorgd om te vallen
- Score 20-27: Personen zijn gemiddeld bezorgd om te vallen
- Score 28-64: Personen zijn zeer bezorgd om te vallen

Instructies:

- we willen u graag enkele vragen stellen over hoe bezorgd u bent dat u zou kunnen vallen bij het uitvoeren van een bepaalde activiteit
- het gaat er hierbij om hoe u **gewoonlijk** deze activiteit uitvoert
- als u tegenwoordig deze activiteit **niet doet** willen we u vragen of dit zo is uit *bezorgdheid om (opnieuw) te vallen* of om een *andere reden*

Hoe bezorgd bent u dat u zou kunnen vallen bij…	Helemaal niet bezorgd	Een beetje bezorgd	Tamelijk bezorgd	Erg bezorgd	(*) Deze activiteit wordt niet uitgevoerd uit bezorgdheid om te vallen
1. Het schoonmaken in huis (zoals vegen, stofzuigen of afstoffen)	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
2. Het aan- of uitkleden	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
3. Het klaarmaken van eenvoudige maaltijden	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
4. Het nemen van een bad of douche	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
5. Het doen van boodschappen	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
6. Het in of uit een stoel komen	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
7. Het op- of aflopen van een trap	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
8. Het maken van een wandeling in de buurt	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
9. Het reiken naar iets boven uw hoofd of naar iets op de grond	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
10. Het beantwoorden van de telefoon voordat deze ophoudt met overgaan	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
11. Het lopen op een gladde ondergrond (bijvoorbeeld nat of bevroren)	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
12. Het bezoeken van een vriend(in), kennis of familielid	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
13. Het lopen op een plek waar veel mensen zijn	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
14. Het lopen op oneffen ondergrond (zoals kinderkopjes of slecht onderhouden trottoir)	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
15. Het op- of aflopen van een helling	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:
16. Het bezoeken van een sociale gelegenheid (zoals kerkdienst, familiebijeenkomst of verenigingsactiviteit)	1☐	2☐	3☐	4☐	- ja ☐ neen ☐ - andere reden:

(*) Redenen voor restrictie activiteit toegevoegd door de wetenschappelijke werkgroep "Uniforme Aanpak Valpreventie Vlaanderen"

Appendix C: Case report forms

C.1 Test session one and two

Name patient: Patient code: Date:

Name researcher: Test session:

Test session _

1. 6MWT

	Pre	Min. 1	Min. 2	Min. 3	Min. 4	Min. 5	Min. 6
VAS fatigability							
Heart rate							
Distance	X						

Remarks:

Page **1** of **4**

Name patient: Patient code: Date:

Name researcher: Test session:

2. Objective measures

Spasticity – Modified Ashworth Scale (0 - 1 - 1+ - 2 - 3 - 4)

	Pre		Post		+10 min.		+20 min.		+30min.	
	Right	Left	Right	Left	Right	Left	Right	Left	Right	Left
Quadriceps										
Hamstrings										
Triceps surae										

Muscle strength – Motricity index (0 - 9 - 14 - 19 - 25 - 33)

	Pre		Post		+10 min.		+20 min.		+30min.	
	Right	Left	Right	Left	Right	Left	Right	Left	Right	Left
Ankle dorsiflexion										
Knee extension										
Hip flexion										

Balance – Romberg test

Tandem – Foam – Single leg (circle the one performed)

	Pre	Post	+10 min.	+20 min.	+30min.
Trial 1					
Trial 2					
Trial 3					

Name patient: Patient code: Date:

Name researcher: Test session:

3. Symptom inventory

Geef een score tussen 0 en 10 voor de volgende symptomen:

	Voor wandeltest	Na wandeltest	10 min. na wandeltest	20 min. na wandeltest	30 min. na wandeltest	Tijd na wandeltest tot baseline
Algemene vermoeidheid						
Motorische vermoeibaarheid						
Aandachtsproblemen						
Spasticiteit						
Sensitiviteit/gevoelsstoornis						
Visuele problemen						
Balansstoornissen						
Pijn						
Negatieve veranderingen in gangpatroon						
Duizelig						
Spierzwakte						

0 1 2 3 4 5 6 7 8 9 10

Neen/geen Middelmatig Extreem

C.2 Test session three

Name patient: Patient code: Date:

Name researcher: Test session:

Test session _

1. 6MWT

	Min. 1	Min. 2	Min. 3	Min. 4	Min. 5	Min. 6
Distance						

Order of testing: ☐ **Vigilance None**
Indicate with 1 and 2
☐ **Vigilance Walk**

Remarks:

Name patient: Patient code: Date:

Name researcher: Test session:

3. Symptom inventory

Geef een score tussen 0 en 10 voor de volgende symptomen:

	Voor wandeltest	Na wandeltest
Algemene vermoeidheid		
Motorische vermoeibaarheid		
Aandachtsproblemen		
Spasticiteit		
Sensitiviteit/gevoelsstoornis		
Visuele problemen		
Balansstoornissen		
Pijn		
Negatieve veranderingen in gangpatroon		
Duizelig		
Spierzwakte		

0 1 2 3 4 5 6 7 8 9 10

Neen/geen Middelmatig Extreem

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Jaar: **2018**

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Bielen, Hanne

Datum: **7/06/2018**