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FACULTEIT GENEESKUNDE EN LEVENSWETENSCHAPPEN
master in de revalidatiewetenschappen en de kinesitherapie

Masterproef deel 1

Effect of High-Intensity Interval Training on Mitochondrial Density in Skeletal Muscles of MS Patients

Promotor :
Prof. dr. Bert OP 'T EIJNDE

Copromotor :
dr. Inez WENS

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Eerste deel van het scriptie ingediend tot het behalen van de graad van master in de revalidatiewetenschappen en de kinesitherapie

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MP 1: Effect of High-Intensity Interval Training on Mitochondrial Density in Skeletal Muscles of MS Patients

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Research question: What is the effect of high-intensity interval training (HIIT) on mitochondrial density in skeletal muscles of MS patients (pwMS)?

Highlights:

- Multiple sclerosis patients have reduced skeletal muscle oxidative capacity and see a decline in skeletal muscle mitochondrial content and biogenesis.
- High-intensity interval training improves muscle oxidative capacity, mitochondrial content and mitochondrial biogenesis in healthy individuals.
- Key factors concerning mitochondrial biogenesis are peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), adenosine monophosphate-activated protein kinase (AMPK) and p38 mitogen-activated protein kinase (p38-MAPK)
- The impact of HIIT on mitochondrial biogenesis and mitochondrial density in pwMS has yet to be investigated.

Students: Severijns Tobias, Wijckmans Ferdy

Promotor: Prof.dr. Op 't Eijnde Bert

Co-promotor: Dr. Wens Inez

Research Framework

This literature study fits in the research domain of cardiorespiratory and internal diseases as well as the neurological subdomain of the rehabilitation sciences and physiotherapy department of Hasselt University and is constructed according to the central format.

The effect of high-intensity interval training (HIIT) on biomarkers of mitochondrial biogenesis and mitochondrial density in skeletal muscles of patients with multiple sclerosis (pwMS) was studied through a literature review. Outcome measures are: peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), p38 mitogen-activated protein kinase (p38-MAPK), adenosine monophosphate-activated protein kinase (AMPK), adenosine triphosphate (ATP), cytochrome c oxidase (COX) and citrate synthase (CS).

HIIT was the intervention of choice because of the small effect size of low to moderate intensity exercise. Here, higher intensity exercise might improve therapy outcomes, due to the major advantages of intense bursts of activity interspersed with fixed periods of less-intense activity, or even complete rest. Exercise duration of HIIT is short in comparison with endurance exercise. This makes it an interesting exercise modality for pwMS who often have a reduced exercise capacity. In the second part of this study, a research protocol is described.

The research topic was provided by our promotor prof. dr. Bert Op 't Eijnde and co-promotor dr. Inez Wens. The final research question, literature study and research protocol were specified and completed by master students Tobias Severijns and Ferdy Wijckmans in co-operation with the promotor and co-promotor.

Part two of this thesis will be done in Diepenbeek at the research centre REVAL of UHasselt.

This thesis is a duo-thesis and every part individually accomplished by both master students is clearly described in table 11.

Content

PART 2: RESEARCH PROTOCOL

1. Abstract

Background: Recent research, concerning skeletal muscle characteristics in patients with Multiple Sclerosis (pwMS), suggest that lower physical activity levels correlate with a reduction in skeletal muscle oxidative capacity and changes in muscle signalling cascades for mitochondrial and myofibrillar biogenesis. Therefore, exercise is a safe and efficient way in improving these deteriorations. The effect size of low to moderate intensity exercise, however, is small. Here, and similar to healthy controls and other disease populations, higher intensity (HIT) exercise might improve therapy outcome. Due to its short exercise duration, high-intensity interval training (HIIT) is an interesting exercise modality for pwMS, who often have a reduced exercise capacity.

Methods: PubMed and Web of Science (WoS) databases were scanned for literature on Multiple Sclerosis (MS), mitochondrial biogenesis and the effect of exercise training on both frameworks. Research was based on a wide array of MeSH-terms and general terms, which were selected following previous analysis concerning this subject-matter.

Results: HIIT protocols seem to improve muscle oxidative capacity, maximal activity of mitochondrial enzymes in skeletal muscle, mitochondrial content and also govern the activation of mitochondrial biogenesis in healthy individuals.

Discussion and conclusion: Exercise training has beneficial effects on biomarkers of mitochondrial biogenesis in healthy subjects. Furthermore, an increase in mitochondrial content is present. Namely low-volume HIIT programs seem to have a positive impact on these markers. Further research is needed to address the impact of HIIT on biomarkers of mitochondrial biogenesis and mitochondrial density in pwMS.

Operationalization: MS patients will be randomized in a sedentary control group and a high-intensity interval training group and will complete a six-week low-volume HIIT protocol. Pre- and post-measurements will consist of strength measurements of the quadriceps muscle, a body composition measurement, a cardiopulmonary exercise test, the physical activity scale for individuals with physical disabilities and muscle biopsies from the middle part of the vastus lateralis muscle of the weakest leg. Primary outcome measures will be: peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α) protein expression, adenosine monophosphate-activated protein kinase (AMPK), p38 mitogen-activated protein kinase (p38-MAPK) and mitochondrial density.

Keywords: AMPK, ATP, COX, CS, High-intensity interval training, Multiple sclerosis, Mitochondrial biogenesis, Mitochondrial density, PGC-1α, p38MAPK

2. Introduction

Multiple sclerosis (MS) is a progressive demyelinating disease of the central nervous system, affecting approximately 2 300 000 people worldwide. The early course of MS is marked by episodes of neurological dysfunction that generally recover. However, as the disease progresses, pathological changes become dominant and neurological degeneration is present. A wide range of heterogeneous neurological and peripheral symptoms characterizes MS. Most common symptoms being fatigue and muscle weakness, which, amongst others, lead to a decrease in functional capacity as well as a reduction in health related quality of life (HRQOL). Neurologically, metabolic changes within axons and demyelination of the central nervous system are present. Although MS primarily remains a neurological disease, it is evident that there are significant changes present within the musculoskeletal system. Evident is a downgrade in mean muscle fibre cross sectional area (CSA), muscle strength, muscle mass, a smaller type 1 and 2 skeletal muscle fibre diameter and complex-1 deficiency in skeletal muscle mitochondria.[1-7] However, the role of skeletal muscle mitochondria has only been sparsely investigated amongst pwMS.[5; 8-10] Thus, in conjunction with neurological deterioration, muscular adaptations are present, which need to be further investigated on cellular level.

Recent research, concerning skeletal muscle characteristics in pwMS, suggest that lower physical activity levels correlate with a reduction in skeletal muscle oxidative capacity and changes in muscle signalling cascades for mitochondrial and myofibrillar biogenesis.[1; 2] Mitochondrial biogenesis can be seen as the growth and division of pre-existing mitochondria and is influenced by environmental stress such as exercise.[11-13] Consequently, increasing physical activity is an interesting tool that safely and efficiently improves physical deconditioning in MS. Indeed, low-to moderate endurance training is well tolerated in pwMS and improves maximum aerobic capacity and HRQOL.[5; 14] The effect size of low to moderate intensity exercise, however, is small. Here and similar to healthy controls and other disease populations, higher intensity (HIT) exercise might improve therapy outcome.

To date, it is clear that HIT improves skeletal muscle oxidative capacity, maximal activity of mitochondrial enzymes in skeletal muscle, increases mitochondrial content and governs activation of mitochondrial biogenesis in healthy subjects. Which functionally lead to higher exercise performance rates, and an increase in HRQOL.[15-23] However, the impact of these HIT programs on mitochondrial biogenesis and mitochondrial density in pwMS has yet to be investigated.[6; 14]

Key factors, concerning mitochondrial biogenesis, investigated in this literature study are peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), which stimulates mitochondrial biogenesis in skeletal muscle, adenosine monophosphate-activated protein kinase (AMPK), which regulates intracellular energy metabolism as a response to an acute energy crisis and p38 mitogen-activated protein kinase (p38-MAPK), regulating exercise-induced mitochondrial biogenesis which leads to an increase in transcriptional activity of PGC-1α. Other key mitochondrial biomarkers are citrate synthase (CS) activity and the activity of cytochrome c oxidase (COX). CS activity is a validated biomarker for mitochondrial density in skeletal muscle, as well as a biochemical marker for skeletal

muscle oxidative adaptation to an exercise intervention. COX or complex IV is seen as a key regulation site for oxidative phosphorylation and catalyses the final step in the mitochondrial electron transfer chain. Further, an essential adaptation to endurance exercise is an increased capacity to supply adenosine triphosphate (ATP) through oxidative phosphorylation, marked by an increase in mitochondrial content and associated with enhanced exercise capacity. In conjunction with enzyme activity, mitochondrial density, consisting of mitochondrial number and fractional area, increases as well in response to endurance exercise.[11; 13; 15; 24-27]

The purpose of this literature study was to investigate the effect of HIIT on mitochondrial biogenesis and density in skeletal muscles of pwMS.

3. Method

3.1 Research question

What is the effect of high-intensity interval training on mitochondrial density in skeletal muscles of patients with multiple sclerosis?

3.2 Literature search

PubMed and Web of Science (WoS) databases were scanned for literature on this topic. A comprehensive study on MS, mitochondrial biogenesis and the effect of exercise training on both frameworks was executed. Based on this, a wide array of MeSH-terms was selected. Following MeSH-terms were used in PubMed: multiple sclerosis, demyelinating disease, immune system disease, neurodegenerative disease, mitochondria, mitochondria muscle, mitochondrial density, mitochondrial myopathies, mitochondria muscle/metabolism, mitochondria/enzymology, citrate (si)-synthase, mitochondrial ADP ATP translocases, exercise therapy, exercise, muscle skeletal/metabolism, muscle skeletal/pathology, muscle skeletal/physiopathology, muscle skeletal, AMP-activated protein kinases and p38 mitogen-activated protein kinases. In absence of MeSH-terms, general terms were selected based on previous research concerning this subject-matter, others were based on the research question. Following general terms were used: high intensity training, high intensity exercise, mitochondrial density and pgc-1 alpha mitochondrial. All these terms were combined with each other using “AND” or “OR” (Table 1). In case of more than 250 hits, the search term was specified, further combining them with other eligible terms. The titles of all hits were screened for relevance. Potential relevant articles were extracted by selecting them and sending them to EndNote X7 for further screening. However, few studies investigated the effect of exercise training on mitochondrial density in skeletal muscles of pwMS. The decision was made to include trials on healthy subjects. After completion of the literature search based on title screening, abstracts were assessed for usefulness. Non-eligible studies were excluded. In WoS, topic-related terms were used: multiple sclerosis, demyelinating disease, immune system disease, neurodegenerative disease, mitochondria, mitochondria muscle, mitochondrial density, mitochondrial myopathies, citrate synthase, mitochondrial ADP ATP translocases, exercise therapy, exercise high intensity exercise, high intensity training, muscle skeletal, peroxisome proliferator-activated receptor gamma coactivator 1-alpha, AMP-activated protein kinases and p38 mitogen-activated protein kinases (Table 2). Methods used for further examination were comparable with methods used in PubMed. Furthermore, for all articles, cited references were checked as well as the articles which cited them.

3.3 Selection criteria

Criteria for inclusion:	Criteria for exclusion
Humans	Animal studies
Multiple sclerosis	Review
Healthy subjects	Studies in languages other than Dutch,
Exercise training	English and French
Effect of exercise on muscle	No relevant population
metabolism	No relevant intervention
Mitochondria (skeletal muscle)	No relevant outcome
	Mitochondria (axonal)

3.4 Quality assessment

To assess the quality of all included articles, following checklists were used: the Cochrane checklist for RCT, the quality assessment standard for cross-over studies, a self-made checklist based on Downs and Black checklist (1998) and the STROBE statement checklist. The quality assessment was done by one author and controlled by the other.

- The Cochrane checklist for randomized controlled trials (RCT) reviewed three studies on 10 criteria with a maximal score of 10 (Table 5).

- The Quality assessment standard for a cross-over study examined three studies on nine items (Table 6).

- The self made checklist, based on Downs and Black checklist (1998) for quasi experimental studies without control group, evaluated 13 studies on 20 criteria with a maximal score of 20 (Table 7).

- The STROBE Statement Checklist for cross-sectional studies assessed seven studies on 22 criteria (Table 8).

3.5 Data extraction

Subsequently to completion of the literature search and quality assessment, 25 studies were included and used for data extraction. All eligible studies were processed and data was elaborated into three different tables (Table 9.1, 9.2 and 9.3). This study focusses on mitochondrial density in skeletal muscles of pwMS following exercise training. However, considering the low amount of research on this subject, articles in healthy subjects were included. Based on multiple reviews, concerning mitochondrial biogenesis, various parameters were admitted. The key parameter to be investigated being PGC-1α. Other important investigated parameters are p38-MAPK, AMPK, COX, CS and ATP.

4. Results

4.1 Results study selection

Due to the fairly recent nature of research regarding the effect of HIIT on mitochondrial density in skeletal muscles of pwMS, the study selection did not solely aim on articles concerning MS. Eligible trials on healthy subjects were included. Out of the 1000 hits on PubMed and WOS, concerning MS, we eventually excluded 939 articles (Figure 1). Out of the 2487 hits on PubMed and WOS, related to healthy subjects, we discarded 2469 articles (Figure 2). The most common reasons for exclusion were no relevant outcomes, intervention or population (Table 3, 4). Furthermore, for all articles, cited references were checked as well as the articles which cited them.

Thirteen articles, most of them being reviews, were included in the reference list for background purposes concerning multiple sclerosis, exercise, mitochondrial density, mitochondrial biogenesis and PGC-1α. These articles weren't used for the results of the data extraction.[5; 8; 10-14; 28-33].
Ultimately 25 articles were used for data extraction. Three randomized controlled trials, two randomized crossover designs, 13 quasi-experimental studies without control group, six cross sectional studies and one combination of a randomized crossover design and a cross sectional study design were included.

4.2 Results quality assessment

The quality assessment of all 25 included articles was completed with four different checklists. The lowest scores found in all checklists were 4/10 and 8/21. After discussion between both authors, the decision was made to not exclude articles with lower quality, because of the scarcity of eligible studies. Quality assessment of randomized controlled trials[6; 15; 34] varied from 4/10 to 6/10 (table 5). The Strobe checklist was used to assess the quality of studies with a cross sectional design[1; 2; 4; 7; 9; 25] (table 8). Most included studies used a quasi-experimental design without control group. For the assessment of these articles a self-made checklist was used, based on Downs and Black checklist (1998). Scores of these studies ranged between 8/20 and 16/20[16-20; 22; 23; 26; 35-39] (Table 7). Quality assessment standard for cross-over studies was used for three articles[21; 24; 25] (table 6).

In general, quality ranged between low and moderate and after consideration no studies were excluded.

4.3 Results data extraction

Twenty-five articles were included and used for data extraction. Various parameters were introduced based on multiple reviews regarding mitochondrial biogenesis, in agreement with the promotor of this study.[11; 13; 30]

The most important parameter to be investigated according to these reviews was peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α). A protein that induces and coordinates gene expression, which stimulates mitochondrial biogenesis in skeletal muscle through specific interaction with a variety of transcription factors.[11; 13; 15; 16; 19; 21-26; 29; 33; 35]

Another important regulator of mitochondrial biogenesis is adenosine monophosphate-activated protein kinase (AMPK), which regulates intracellular energy metabolism as a response to an acute energy crisis.[11; 13; 26]

P38 mitogen-activated protein kinase (p38-MAPK) is one more key regulator of exercise-induced mitochondrial biogenesis, which leads to an increase in transcriptional activity of PGC-1α.[11; 13; 24-26; 33]

Further, an essential adaptation to endurance exercise is an increased capacity to supply adenosine triphosphate (ATP) through oxidative phosphorylation, marked by an increase in mitochondrial content and associated with enhanced exercise capacity.[15; 26; 30; 36-38]

Other commonly used markers of mitochondrial content are citrate synthase (CS) activity and the activity of cytochrome c oxidase (COX). CS activity is a validated biomarker for mitochondrial density in skeletal muscle, as well as a biochemical marker for skeletal muscle oxidative adaptation to an exercise intervention. COX or complex IV is seen as a key regulation site for oxidative phosphorylation.[16; 22; 30; 34; 38]

Table 9.1 gives an overview of types of articles, subjects, used techniques and conclusion.
Table 9.2 gives an overview of patient populations, aims of the study and intervention.
Table 9.3 gives an overview of outcome measures and results.

4.3.1 The effect of exercise on **PGC-1α**

The outcomes of exercise on PGC-1α have been discussed in multiple studies amongst healthy participants. All of these studies took muscle biopsies from the vastus lateralis muscle pre- and post-exercise, with some taking additional biopsies two, three, 24, 48, 52 and/or 72 hours post-exercise. Western blotting was used to determine PGC-1α protein expression and PGC-1α mRNA.

Seven articles examined post-exercise PGC-1α mRNA in vastus lateralis muscle and found a significant increase compared to pre-exercise PGC-1α mRNA. With a rise of 400%[24; 25], 200%[26], 750%[19] and Stepto et al.[22] reporting a rise of 430% three hours post-training. One study found a 200-300% increase in PGC-1α mRNA immediately post-exercise, which further rose to 620% two hours post-training.[21] Edgett et al.[35] reported a difference between three different intensities, with the greatest increase in PGC-1α mRNA (790%) at a workrate of 100% of the peak rate of oxygen consumption ($VO2_{peak}$). After 24 hours of recovery PGC-1α mRNA returned to basal levels.[19; 21]

Nine articles discussed PGC-1α protein content after high intensity interval training. Five articles concluded there was no significant change in protein content three hours post-exercise.[19; 20; 23; 24; 26] However, one of these articles stated that there was a 57% increase at 24 hours of recovery noticeable.[19] The other four articles declared there was a significant increase in PGC-1α protein content after training.[15; 16; 21; 22]

To our knowledge, no research has been done into the effect of HIIT on PGC-1α in pwMS.

4.3.2 The effect of exercise on **AMPK**

The outcomes of exercise on AMPK have been discussed in multiple studies using healthy participants and in one study amongst pwMS. All of these studies took muscle biopsies from the vastus lateralis muscle pre- and post-exercise, with some taking additional biopsies three, 24 and/or 72 hours post-exercise. Western blotting was used to determine AMPK phosphorylation, phospho-AMPKα and acetyl-coenzyme A carboxylase (ACC) phosphorylation, which is a marker of AMPK activation.

Two studies examined the effect of exercise on AMPK phosphorylation. Bartlett et al.[24] investigated the difference between matched HIIT and CONT exercise protocols. Both training modalities produced a 1.5-fold increase of post-exercise AMPK phosphorylation, with no difference between conditions. AMPK phosphorylation returned to basal levels at three hours post-exercise. Gibala et al.[26] measured the effect of a low-volume, high intensity interval training protocol on AMPK phosphorylation after four 30 seconds “all out” exercise bouts, interspersed with four minutes of rest. A rise in phosphorylation of AMPK after bout four, compared with all other time points, was evident.

Hansen et al.[2] researched the effect of exercise on phospho-AMPKα in pwMS compared to healthy controls.

- Basal muscle phospho-AMPKα was significantly different between groups (1.46 ± 0.24 VS 1.09 ± 0.26 mg/mL in pwMS VS healthy subjects, respectively, observed statistical power α = 0.76).
- After endurance exercise, muscle phospho-AMPKα was significantly different between groups (1.66 ± 0.51 VS 1.12 ± 0.23 mg/mL in pwMS VS healthy subjects, respectively, observed statistical power α = 0.96).
- Within pwMS and healthy subjects muscle phospho-AMPKα did not significantly change. Relative change in muscle phospho-AMPKα (+19 ± 53% VS +4 ± 13% in pwMS VS healthy subjects, respectively) after endurance exercise was comparable between groups.

Three studies discussed ACC phosphorylation after exercise training. Two studies[19; 26] used a low-volume HIIT protocol to investigate the effect on ACC phosphorylation. HIIT resulted in an increase of ACC phosphorylation after bout one and four compared with the other bouts.[26] Little et al.[19] reported an immediate increase of cytosolic ACC phosphorylation.
One remaining article[25] studied the effect of both INT and CONT exercise. Both protocols elicited similar 2.5-fold increases of ACC serine-79.

4.3.3 The effect of exercise on **p38-MAPK**

Phosphorylation of p38-MAPK was studied in four articles[19; 24-26] amidst healthy subjects. All these articles discuss the acute effects of different HIIT protocols on skeletal muscle p38-MAPK phosphorylation.

In two studies[19; 26] the participants performed four 30 seconds maximal intensity exercise bouts on an ergometer cycle, interspersed with four minutes of rest. Outcomes were a significant increase in p38-MAPK phosphorylation immediately after exercise, specific numbers were not mentioned. The research of Little et al.[19] indicated that p38-MAPK phosphorylation returned to basal levels after three hours of recovery, and once more significantly elevated after 24 hours of recovery (P = 0.004).

The other two articles compared a HIIT to a CONT protocol. J.D. Bartlett (2012)[24] compared a HIIT running protocol to CONT running and found no significant difference between these two protocols, which were matched for intensity, duration and distance. However, p38-MAPK phosphorylation increased 1.9 and 1.5 fold immediately following exercise for HIIT and CONT respectively. Cochran et al.[25] compared intermittent (HIIT) and continuous (CONT) high-intensity exercise to each other, performed on a cycle ergometer. There was an immediate increase of p38-MAPK phosphorylation after exercise by 300%, and no difference noticeable between treatments.

To our knowledge, no research has been done into the effect of HIIT on p38-MAPK in pwMS.

4.3.4 The effect of exercise on **CS**

The effect of CS in healthy subjects has been discussed in several studies. Nine studies demonstrated a significant increase of maximal CS activity following exercise training, although different exercise protocols were used. One study investigated the effect of low-volume "sprint" interval training (SIT) and endurance training (ET) on maximal activity of CS and marked an increase of 25% with no difference between interventions.[15] Fernstrom et al. assessed the difference between acute exercise and endurance training in a six-week training program, which showed an increase of CS maximal activity of 43% and 47% respectively[34]. Seven studies displayed a significant increase of CS maximal activity following HIIT, while different protocols of HIIT were used.

Following studies applied a low-volume HIIT protocol of one session[19], two weeks[20; 23] and six weeks[16; 18; 38], showing a significant increase of CS maximal activity in healthy subjects of 14%, 16%, 43%, 28%, 36%, and 26% respectively. One remaining study[39] marked an improvement of CS maximal activity of 30% following two weeks of training, four additional weeks of training resulted in further increases of 20%. Only one article[25], using a six week CONT exercise training protocol of 60 minutes at 65% of $VO2_{peak}$, displayed no significant difference in maximal activity of CS after training compared with pre-training.

In addition, CS protein content significantly increased by 30% 24 hours post-exercise[19] and 20%[20] after respectively one session or two weeks of low-volume high-intensity exercise training.

Furthermore, Stepto et al.[22] evaluated the effect of acute exercise or exercise training on CS mRNA, which remained unaltered after intervention.

To our knowledge, no research has been done into the effect of HIIT on CS in pwMS.

4.3.5 The effect of exercise on **COX**

The effect of exercise on maximal activity of COX was analysed in three studies.[17; 19; 20] All but one[19] concluded a significant increase of COX maximal activity. After one single session COX activity increased by 19% but did not reach statistical significance.
Two studies used a two week high-intensity low-volume interval training protocol, and found a significant improvement of COX maximal activity of 29%[20] no percentages were given in Jacobs et al.[17].

COX IV content was investigated in seven studies[16; 19; 20; 22; 25; 38; 39], all but one[22] found a significant increase. After one session[19], two weeks[20] and six weeks[16; 38; 39] of HIIT there was a significant increase of respectively 43% at three hours recovery and 30% at 24 hours of recovery[19], 38%[20], 36[16], 18%[38] and 30% following two weeks with a further increase of 20% after an additional four weeks of training.[39] Cochran et al.[25] used a CONT protocol consisting of six weeks of exercise training and demonstrated a significant increase of 20% in COX IV content.

The study of Stepto et al.[22] demonstrated a 1.6 fold rise of COX IV mRNA and a 1.5 fold increase of COX IV protein expression by training, however COX IV protein expression showed a 20% decrease by acute exercise pre- and post-training.

COX II protein content was assessed in two articles.[19; 20] In one after one session[19] and in the other study after two weeks[20] of high-intensity, low-volume interval training in healthy subjects. Respectively, an increase of 29% and 35% at 24 hours recovery was illustrated.

To our knowledge, no research had been done into the effect of HIIT on COX in pwMS.

4.3.6 The effect of exercise on **ATP**

The rates of ATP following HIIT has been investigated in five articles amongst healthy individuals.[15; 26; 36-38]

One of these studies[26] explored the effects of a brief intense interval exercise into human skeletal muscle. The training protocol consisted of four-30 seconds exercise bouts (maximum intensity), interspersed with four minutes of rest. The protocol was completed by six healthy young men. Muscle ATP was only lowered ($P \leq 0.05$) after bout four.

Burgomaster et al.[15] compared metabolic adaptations during exercise after low volume sprint interval and traditional endurance training, following a six weeks training programme. Endurance training (ET) consisted of 40 to 60 minutes of continuous cycling on a workload of 65% $VO2_{peak}$, five days a week. Sprint interval training (SIT) consisted of four to six repetitions of a 30 seconds Wingate test, interspersed with 4.5 minutes recovery between bouts, three days a week. Muscle ATP remained unchanged by acute exercise, but reduced ($P<0.05$) after six weeks of SIT compared to ET.

The other three articles used three different high intensity interval protocols. One[38] where eight untrained men performed six sessions of HIT in a two week period. Training consisted of four to six 30 seconds maximum intensity cycling bouts with four minutes of recovery between bouts. A second[37] where ten overweight untrained healthy subjects followed a six week (three times a week) exercise program consisting of five 60 seconds exercise bouts (at approximately 128% of the maximum load), interspersed by 90 seconds of active rest. The last study[36] where five males and three females attended a six week (three times a week) exercise program with ten four minute intervals at 90% $VO2_{peak}$ interspersed with two minutes of passive rest between intervals. The first and third article found no change in the unidirectional rate of ATP synthesis, as well as no change in the content of ATP in vastus lateralis muscle.[36; 38] The second study concluded that ATP production by each pathway was unchanged after the first session. Total ATP synthesis increased during a 24 seconds maximum voluntary contraction ($P< 0.001$), but remained the same across the three testing sessions ($P = 0.62$). Oxidative ATP synthesis (ATP_{OX}) increased with training ($P<0.001$). ATP provision from net

breakdown of phosphocreatine (ATPCK) and glycolysis (ATP_{GLY}) remained similar across the three testing sessions (P session ≥ 0.45).[37]

To our knowledge, no research has been done into the effect of HIIT on ATP in pwMS.

5 Discussion

5.1 Reflection of the quality assessment

The quality assessment of all 25 included articles was completed using four checklists due to different study designs. Of all included articles the highest level of evidence used in this literature search is randomized controlled trial. Three randomized controlled trials were admitted, two randomized crossover designs, six cross sectional studies, 13 quasi-experimental studies without control group and one combination of a randomized crossover design and a randomized controlled trial. For quality assessment of studies using a quasi-experimental study design without control group, a self-made checklist was used, based on the Downs and Black checklist (1998).

Further, nine reviews were included in the reference list for background purposes, these articles weren't used for the results of the data extraction. Quality of all articles used for data extraction ranged between low and moderate and after consideration no studies were excluded.

5.2 Reflection on findings related to the research question

The effect of exercise on PGC-1α

Since PGC-1α is one of the most important regulators of mitochondrial biogenesis, this metabolic regulator has been investigated in several studies. Seven articles examined the effect of exercise on PGC-1α mRNA and conclude a significant increase between 200%-750% regardless of exercise protocol modality.[19; 21; 22; 24-26; 35]

Bartlett et al.[24] and Cochran et al.[25] demonstrate a comparable increase of PGC-1α mRNA content in human skeletal muscle following both HIIT protocols and CONT running protocols. Previous research (Edgett, 2013) demonstrated a greater increase in PGC-1α mRNA following maximal (100%) compared to submaximal exercise (73%). The effect of supramaximal exercise (133%) was similar to that observed following submaximal exercise, so no further increase was seen after supramaximal exercise intensity.

Gibala et al.[26] demonstrates that a surprisingly small dose of very high intensity exercise (two minutes of all out cycling) is sufficient to increase PGC-1α mRNA.

Nine articles investigated the effect of PGC-1α after HIIT. The results described in these articles are contradictory. Five of these studies[19; 20; 23; 24; 26] concluded there was no significant increase in protein content following HIIT three hours post-exercise, while the four other articles declared a significant increase of PGC-1α following exercise training at high intensity.[15; 16; 21; 22]

Multiple studies describe that PGC-1α does not appear to increase as a reaction to an acute exercise stimulus, while in the contrary PGC-1α mRNA shows a rapid increase.[19; 20; 26]

All of these articles describe the effect of exercise training in a healthy population, no research has been done in pwMS.

The effect of exercise on AMPK

Five studies investigated the effect of exercise training on AMPK- and ACC-phosphorylation, which is a marker of AMPK activation, while AMPK is one of the proteins linked to PGC-1α and the regulation of mitochondrial biogenesis in skeletal muscle.

Two articles[24; 26] describe the effect of exercise on AMPK-phosphorylation, one study demonstrate that both HIIT and CONT exercise induce comparable increase in AMPK-phosphorylation[24]. Gibala et al.[26] also describes an increase of signalling through AMPK, following a low-volume, high intensity interval training protocol.

Three studies[19; 25; 26] investigate the effect of exercise training on ACC-phosphorylation and all show an increase in ACC phosphorylation regardless to the exercise protocol used, as well as Bartlet et al.[24] previously described an increase of AMPK-phosphorylation in both CONT and HIT protocols.

One single study[2] looked in to the effect of exercise training in pwMS. The basal muscle phospho-AMPKα was significantly higher in pwMS in comparison with healthy subjects, after endurance exercise there was no significant difference between both groups.

The effect of exercise on p38-MAPK

Phosphorylation of p38-MAPK, which is an important signalling cascade linked to PGC-1α and the regulation of mitochondrial biogenesis in skeletal muscle[26], had been investigated in four studies. p38-MAPK and AMPK are both activated in response to metabolic stress.[19] Studies investigating HIIT protocols[19; 24-26] conclude a significant increase following exercise regardless of the exercise volume. Gibala et al.[26] and Little et al.[19] both report a significant increase after one single session, following Little et al.[19], values returned to basal levels at three hours recovery and again a significant increase is shown after 24 hours recovery. Two studies[24; 25] investigated the difference between HIIT and CONT exercise protocols, a significant increase is seen with no difference between intervention protocols. Following Cochran et al.[25] the measurement of the gross phosphorylation of p38-MAPK in whole muscle may be less sensitive than the examination of subcellular localization of the same molecule.

All articles describe the effect of exercise training in a healthy population, to our knowledge no research has been done in pwMS.

The effect of exercise on CS

Nine out of ten studies[15; 16; 18-20; 23; 34; 38; 39] describe a significant increase in maximal activity of CS following exercise training in healthy subjects. Only Cochran et al.[25] reported unchanged results using a six-week CONT exercise training protocol. Two studies show an increase of CS protein content after one single session[19] and two weeks[20] of low-volume high-intensity interval training. All studies using a high-intensity interval protocol conclude significant increased results of both maximal activity as well as protein content of CS. The increase is probably regardless the volume of exercise because it is seen after one single session[19] as well as after two[20; 23] and six weeks[16; 18; 38; 39] of exercise training. This could be explained by a rapid increase of CS activity, even after one single session.

The effect of six weeks low-volume "sprint" interval training was compared with endurance exercise with both an increase of CS max activity with no difference between groups.[15]
Fernström et al.[34] investigated the difference between one single bout of acute exercise and six weeks of endurance exercise resulting both in a significant increase. The increase after one single session could be linked to the probable rapid increase of CS activity following exercise.

These findings suggest a high-intensity interval training protocol is a potential and time-efficient method to induce mitochondrial biogenesis in healthy subjects, even at low-volume exercise. No research has been done in pwMS.

The effect of exercise on COX

Only three studies investigated the effect of HIIT on the maximal activity of Cytochrome C Oxidase (COX). Their findings suggest a higher increase after high volume of HIIT, compared to low-volume. Two articles conclude a significant increase after two weeks of HIIT[17; 20] While one study[19] researched the effect of one single bout of HIIT and concluded a not significant increase of 19%.
The effect of HIIT on COX IV- and COX II-protein content was investigated in respectively seven[16; 19; 20; 22; 25; 38; 39] and two studies[19; 20]. HIIT induces an overall increase in both COX IV- and COX II protein content. Only one article[22] reported a 20% decrease in COX IV protein expression by acute exercise pre- and post-training.
Amongst previous research HIIT is an appropriate intervention for the improvement of COX maximal activity as well as the protein content. Previous research suggests a faster increase in COX IV than COX II following exercise buth further research is required.[19]
All articles report results of healthy subjects, no research has been done in patients with MS.

The effect of exercise on ATP

The effect of exercise on adenosine triphosphate (ATP) has been investigated in five articles. Endurance exercise leads to an increased capacity ATP supply through oxidative phosphorylation. Marked by an increase in mitochondrial content and associated with enhanced exercise capacity.[15; 26; 36-38] Three studies report a reduction of ATP content at rest following low-volume HIIT[15; 36] and SIT[37]. The lowered ATP content after exercise training was possibly due to the stress of chronic training or the acute residual effects of the last exercise performance (72 hours before muscle biopsy). Lowered concentration of ATP directly after exercise is due to high rates of ATP turnover which lead to higher concentration of adenosine monophosphate (AMP).
One article[38] described the effect of a high-volume HIIT protocol on resting ATP content with no significant difference following exercise. No research has been done in pwMS.

5.3 Reflection on strengths and weaknesses of literature search

PubMed and Web Of Science (WOS) databases were used for this literature search. In both databases a comprehensive study on MS, mitochondrial biogenesis and the effect of exercise on both

was executed thoroughly. All relevant MeSH-terms for this research question were used and in absence of MeSH-terms, general terms were selected based on previous research concerning this subject-matter and some were based on the research question. The use of this strategy delivered over thousand hits, In case of more than 250 hits, the search term was specified, further combining them with other eligible terms. However, few studies investigated the effect of exercise training on mitochondrial density in skeletal muscles of pwMS. The decision was made to include trials on healthy subjects. Furthermore, for all articles, cited references were checked as well as the articles which cited them. This broad search strategy is a strength of this study however no studies were excluded on basis of quality assessment which was kept in mind during data extraction and interpretation of results.

5.4 Recommendations for future research

After an extensive literature search as previously described, research regarding the effect of high-intensity interval training on mitochondrial density in pwMS, proved to be scarce. Further research in pwMS is required to evaluate the effect of this type of intervention on mitochondrial density and biogenesis. Momentarily conclusions can only be drawn from studies including healthy subjects. More studies addressing this subject are also needed to investigate the influence of different exercise protocols of HIIT and to conclude which type of exercise protocol is most effective in this particular population.

6 Conclusion

Exercise training of various modalities has beneficial effects on biomarkers of mitochondrial biogenesis such as PGC-1α, p38-MAPK and AMPK. Furthermore, an increase in mitochondrial content, as marked by CS enzyme activity, maximal COX activity and an increased rate of ATP synthesis, is present. Namely low-volume HIIT programs seem to have a positive impact on these markers. Further research is needed to address the impact of HIIT on biomarkers of mitochondrial biogenesis and mitochondrial density in pwMS.

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1. Introduction

Multiple sclerosis (MS) is a progressive demyelinating disease of the central nervous system, affecting approximately 2 300 000 people worldwide. It is characterised by a wide range of heterogeneous neurological and peripheral symptoms, such as fatigue and muscle weakness, which, amongst others, lead to a decrease in functional capacity as well as a reduction in health related quality of life (HRQOL). Neurologically, metabolic changes within axons and demyelination of the central nervous system are present. Increasing evidence indicates a role for mitochondria in the dysfunction and degeneration of neurons. Although MS remains primarily a neurological disease, it is evident that there are significant changes present within the musculoskeletal system. However, the role of skeletal muscle mitochondria has only been sparsely investigated amongst pwMS.[5; 8-10] Thus, in conjunction with neurological deterioration, muscular adaptations are present, which need to be further investigated on cellular level.

Recent research, concerning skeletal muscle characteristics in pwMS, suggest that lower physical activity levels correlate with a reduction in skeletal muscle oxidative capacity and changes in muscle signalling cascades for mitochondrial and myofibrillar biogenesis.[1; 2] Consequently, increasing physical activity is an interesting tool that safely and efficiently improves physical deconditioning in MS. Indeed, low-to moderate endurance training is well tolerated in pwMS and improves maximum aerobic capacity and HRQOL.[5; 14] The effect size of low to moderate intensity exercise, however, is small. Here and similar to healthy controls and other disease populations, higher intensity (HIT) exercise might improve therapy outcome.

To date, it is clear that HIT improves skeletal muscle oxidative capacity, maximal activity of mitochondrial enzymes in skeletal muscle, increases mitochondrial content and governs activation of mitochondrial biogenesis in healthy subjects. Which functionally lead to higher exercise performance rates, and an increase in HRQOL.[15-23] However, the impact of these HIT programs on mitochondrial biogenesis and mitochondrial density in pwMS has yet to be investigated.[6; 14] Key factors concerning mitochondrial biogenesis are peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), which stimulates mitochondrial biogenesis in skeletal muscle, adenosine monophosphate-activated protein kinase (AMPK), which regulates intracellular energy metabolism as a response to an acute energy crisis and p38 mitogen-activated protein kinase (p38-MAPK), regulating exercise-induced mitochondrial biogenesis which leads to an increase in transcriptional activity of PGC-1α. In conjunction with enzyme activity, mitochondrial density, consisting of mitochondrial number and fractional area, increases as well in response to endurance exercise.[11; 13; 15; 24-27]

In accordance with the above line of reasoning, we hypothesize that HIIT has beneficial effects on skeletal muscle characteristics of healthy subjects and pwMS. Specifically enhancing biomarkers of mitochondrial biogenesis such as PGC-1α protein expression, AMPK, p38-MAPK, as well as increasing mitochondrial density in skeletal muscle.

2. Research Goal

2.1 Research Question

What is the effect of high-intensity interval training on mitochondrial density in skeletal muscles of MS patients?

2.2 Hypothesis

We hypothesize that HIIT enhances biomarkers of mitochondrial biogenesis such as PGC-1α protein expression, AMPK, p38-MAPK as well as mitochondrial density in skeletal muscle in MS.

3. Method

3.1 Design

This research will be a randomized controlled trial consisting of pwMS randomized in a sedentary control group (SED, n=20) and an intervention group (HIIT, n=20) participating in a six week HIIT exercise intervention program.

3.2 Participants

A minimum of 40 MS patients with relapsing-remitting MS, diagnosed according to McDonald criteria (EDSS range 1-5), will be included in this study following a written informed consent. Subjects will be excluded when they have other disorders (cardiovascular, pulmonary, neurological, renal and/or cancer), are pregnant, are athletes or physically active adults, aren't able to perform high-intensity exercise, have had an acute MS exacerbation within six months prior to this study, are participating in another study and/or have any contra-indications to perform physical exercise. Inclusion criteria are: patients with relapsing-remitting MS, aged between 30-50 years. The participants are recruited from the MS clinic Overpelt (Belgium).

3.3 Medical Ethics

The study will be approved by the ethical committee of Jessa Hospital Hasselt (s1 protocol) and Hasselt University. All tests will be performed in accordance with the Declaration of Helsinki and public health.

3.4 Study Protocol

3.4.1 Pre-intervention measurements

Following study inclusion, baseline measurements will be performed. They include isometric muscle strength measurements of the quadriceps muscle (F_{quad}), a body composition measurement (BCM), a cardiopulmonary exercise test (CPET), the Physical Activity Scale for Individuals with Physical Disabilities (PASIPD) and muscle biopsies (Bergström needle technique) from the middle part of the vastus lateralis muscle of the weakest leg, according to isometric muscle strength measurements.

3.4.2 Intervention program

Hereafter, participants will be randomly assigned to a sedentary control group (SED) or an exercise intervention group (HIIT). Whereas SED subjects remain physically inactive during the study course, HIIT subjects will perform a six-week training program. Neither the researchers, nor the patients participating in this study will be blinded to group allocation.

HIIT = High Intensity Interval Training program

Subjects will engage in a six-week low-volume HIIT exercise program. Based on the literature search, six weeks of HIIT was chosen, being the most common intervention length in healthy subjects.[15; 16; 18; 25; 34; 38; 39] Multiple HIIT modalities are present. Most common used HIIT programs, to define progress in mitochondrial biogenesis, are low-volume HIIT protocols[15; 17-20; 23; 25; 26; 36; 37]. Particularly the Wingate

test, as a training protocol, is most often used. This is an anaerobic test performed on a cycle ergometer. Modified by means of a training program, it consists of four to six repeats of a 30 seconds Wingate test, interspersed with 4.5 minutes recoverytime (cycling at low cadence < 50 rpm against a light resistance of 30W) to decrease the onset of venous pooling and to reduce the change for a sudden blood pressure drop, nausea and light-headedness. Participants will perform these sessions five days per week (Monday-Friday) for six consecutive weeks. The first two weeks consist of four repeated Wingate tests, the next two weeks of five bouts, and the last two weeks of six bouts.

3.4.3 Post- intervention measurements

Following HIIT exercise therapy or SED, post-intervention measurements will be performed identical to pre-intervention measurements.

Study design overview

Table 1: Study design overview of the HIIT protocol including pre- and post-intervention measurements.

Day 1	• F_{quad} • BCM • PASIPD
Day 3	• CPET
Day 7	• First biopsy taken from the vastus lateralis muscle at rest • 65% $VO2_{peak}$ cycling for 60 minutes • Second muscle biopsy taken immediately after exercise • Third muscle biopsy taken 3 hours post-exercise
Week 1 and 2	• Start HIIT protocol • 4 repeats of a 30s 'all out' Wingate test interspersed by 4.5min recovery
Week 3 and 4	• 5 repeats of a 30s 'all out' Wingate test interspersed by 4.5min recovery
Week 5 and 6	• 6 repeats of a 30s 'all out' Wingate test interspersed by 4.5min recovery
Week 6 + 1 day	• F_{quad} • BCM
Week 6 + 3 days	• CPET
Week 6 + 7 days	• First biopsy taken from the vastus lateralis muscle at rest • 65% $VO2_{peak}$ cycling for 60 minutes • Second muscle biopsy taken immediately after exercise • Third muscle biopsy taken 3 hours post-exercise • PASIPD

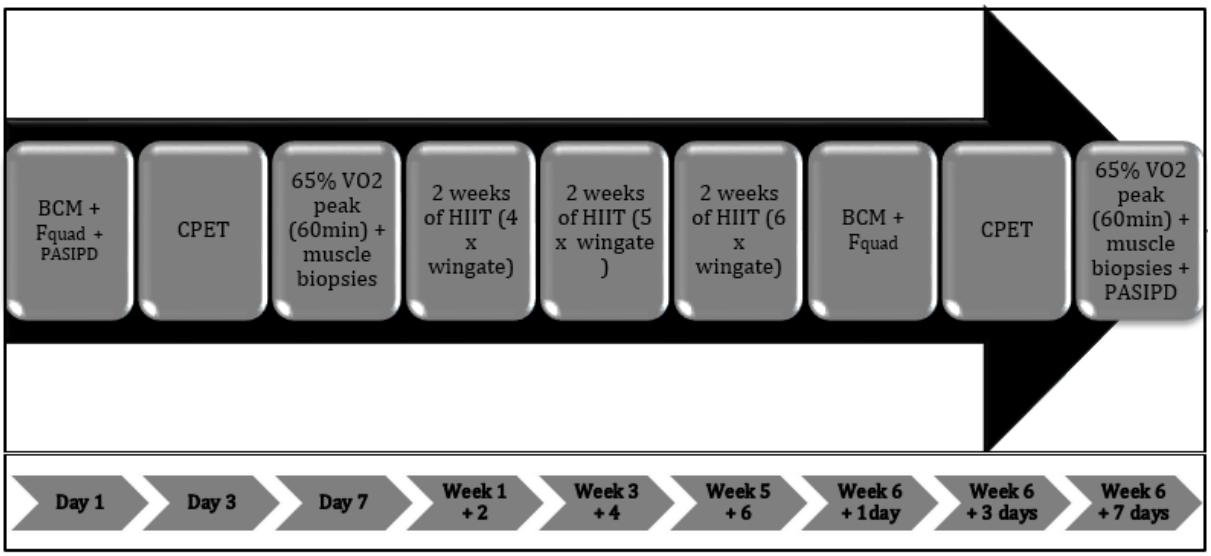

Figure 1. Graphical illustration of the study design overview of the HIIT protocol including pre- and post-intervention measurements.

3.4.4 Measurements

Muscle biopsy

To assess mitochondrial density in skeletal muscles of pwMS, muscle biopsies will be taken from the middle part of the vastus lateralis muscle (Bergström needle technique) of the weakest leg according to isometric muscle strength measurements, pre-exercise, directly post-exercise and three hours after exercise. During the three hour period between post-exercise measurements, subjects will remain seated, will refrain from physical exercise and will only perform light physical activities such as working on a computer, reading or watching television. During the biopsy an incision will be made under a local anaesthetic (2% lidocaine), then two small incisions will be made in the skin and overlying fascia. All muscle samples will be immediately frozen in liquid nitrogen and stored at minus 80 degrees for further analyses.

Mitochondrial density

For electron microscopy, specimens will be rinsed in 0.1 M phosphate buffer and post-fixed in the fixation buffer supplemented with 1% osmiumtetroxide for one hour, dehydrated through a graded ethanol series and embedded in epoxy resin. Appropriate locations and fibre longitudinal orientation will be evaluated in toluidine blue-stained semi-thin sections from the central region of each biopsy. Ultra-thin sections from the selected areas will be contrasted with uranyl acetate and lead citrate and viewed with an electron microscope. Using this staining protocol, mitochondria will appear as spherical round-shaped electron dense (dark grey or black) objects. Mitochondrial analysis will be based on the basic standards. Micrographs of randomly selected areas of central parts of muscle fibres will be obtained. Mitochondria will be identified based on their electron dense (dark grey or black) appearance, shape and subcellular location. These digitised micrographs will be analysed with an interactive image analysis system. Mitochondria will be outlined in an overlaying bitmap, which will be used to compute least diameter, perimeter and size (area). Subsequently, mitochondrial number will

be computed as the quantity of mitochondria in a given muscle fibre area. Mitochondrial fractional area will be computed as the percentile mitochondrial area fraction of total fibre area.[27; 40; 41]

PGC-1α protein expression, AMPK and P38-MAPK

PGC-1α protein expression will be investigated through immunoblotting. A 40- to 50-mg piece of frozen muscle will be added to 10 volumes of homogenizing buffer with 2 µl of protease inhibitor cocktail and homogenized on ice. The sample will be centrifuged at 13,000 rpm for five minutes, and the supernatant will be collected. The protein content of the supernatant will be determined using a bicinchoninic acid, and all samples will be subsequently diluted to a standard concentration using homogenizing buffer. Samples will be further diluted with 4µl Laemmli buffer and heated at 100°C for 5 min. For each blot, a standard and an internal control will be loaded along with 40 µl of each sample onto a 5% polyacrylamide stacking gel and will be separated using a 10% polyacrylamide separating gel of 1.5-mm thickness at 180 V with a running time of 45 minutes in Tris-glycine electrophoresis buffer. The gels will be electroblotted onto nitrocellulose membranes in transfer buffer for 90 min at 90 V at 4°C. Membranes will be incubated in Tris-buffered saline. Membranes will be incubated overnight at 4°C with primary antibodies. After incubation, the membranes will be washed and exposed to appropriate dilutions of anti-species horseradish peroxidase- conjugated secondary antibodies for one hour at room temperature. Membranes will then be rewashed before being exposed to a chemiluminescent liquid for two minutes. Membranes will be exposed using a Bio-Rad Chemi- Doc system, and band densities will be determined using image-analysis software.[19; 26]

Body composition

Body composition will be assessed by a Dual Energy X-ray Absorptiometry scan (GE Hologic Series Delphi-A, Vilvoorde, Belgium). Fat mass, lean tissue mass and bone density will be obtained for the whole body, as well as specific regions covering the legs. Fat mass of the limbs will be calculated. Measurements will be taken at baseline and after completion of the training program at the same time of day.

CPET

Subjects will perform a cardiopulmonary exercise test on an electronically braked cycle ergometer. They will be advised to refrain from any exercise one day in advance as well as the testing day itself, and to eat only a light meal two hours prior to testing. The participants will commence cycling at 60 - 90 r.p.m. for two minutes with an intensity of 40W. Hereafter workload will gradually increase by 20W/min using a ramp protocol (1W/3 sec) until volitional exhaustion (r.p.m. < 50), RER > 1.1 and/or a plateau in VO2 despite an increase in workload. During the cardiopulmonary exercise test respiratory exchange rate will be measured. This is the ratio of carbon dioxide output/oxygen uptake (Vco2/Vo2), measured by gas exchange at the mouth.

Isometric muscle strength of the quadriceps

Consecutive to a five minutes-warming-up period and after habituation, maximal voluntary isometric muscle strength of the knee extensors (45° and 90°) will be measured using an isokinetic

dynamometer (system 3, Biodex ENRAF-NONIUS, New York, USA) in all MS patients and healthy controls. Subjects will perform two maximal isometric extensions (four seconds), separated by 30 seconds of rest. The highest isometric extension peak torque (Nm) will be selected as the maximal isometric strength. The muscle strength of the weakest leg will be reported.

Blood Pressure

Blood pressure will be measured in rest, after the DEXA scan protocol at baseline and after six weeks following training.

Physical activity level

Before and after the HIIT program, subjects will report their physical activity level by using the Physical Activity Scale for Individuals with Physical Disabilities (PASIPD). Patients will be asked to report the amount of days and average hours in a day spent participating in 13 activities over the last seven days. Frequency responses range from 1 (never) to 4 (often), and duration responses range from 1 (less than one hour) to 4 (more than four hours). Total scores will be calculated as the product of the average hours spent in an activity daily and the metabolic equivalents (MET) summed over each item. Scores range from 0 (no activity) to over 100 MET*h/week (very high). At baseline all patients will need to be physical inactive, to be included in the study. Physical inactivity will be defined as < 30 MET*h/week.

3.5 Outcomes

Primary outcomes: PGC-1α protein expression, AMPK, p38-MAPK and mitochondrial density.
Secondary outcomes: $VO2_{peak}$, RER, body composition, isometric muscle strength of the quadriceps muscle, blood pressure and physical activity level.

3.6 Materials and methods

Muscle biopsies will be obtained from the middle part of the vastus lateralis muscle of the weakest leg (dynamometry test) using the Bergström needle technique. The collected muscle tissue will be freed from connective tissue and immediately embedded in Tissue-Tek, frozen in isopentane cooled with liquid nitrogen, and stored at minus 80 degrees until analysis

3.7 Data Analysis

The data analysis will be conducted using IBM SPSS statistics 23, whereas power analysis will be performed using G*Power (80% power set, 0,5 effect size). This indicated a sample size of 20 subjects per group. Between group differences will be investigated using a two-way repeated measures ANOVA. .

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APPENDICES

Table 1: List of search terms used in PubMed.

Search terms, which yielded over 250 hits, were further specified and underlined in following table. The literature study was conducted primarily in January and secondary in May as means of control. Based on literature search of January 2016, two extra search terms were added in May (#23 and #24).

	MESH-terms and keywords in PubMed	# January 2016	# May 2016
#1	"Multiple Sclerosis"[Mesh]	43511	47524
#2	"Demyelinating Diseases"[Mesh]	81577	83150
#3	"Immune System Diseases"[Mesh]	1253799	1270302
#4	"Neurodegenerative Diseases"[Mesh]	225750	230146
#5	"Mitochondria"[Mesh]	134177	136089
#6	"Mitochondria, Muscle"[Mesh]	19955	20135
#7	Mitochondrial Density	9186	9301
#8	"Mitochondrial Myopathies"[Mesh]	4262	4314
#9	"Mitochondria, Muscle/metabolism"[Mesh]	13977	14163
#10	"Mitochondria/enzymology"[Mesh]	28561	28659
#11	"Citrate (si)-Synthase"[Mesh]	3198	3236
#12	"Mitochondrial ADP, ATP Translocases"[Mesh]	1507	1519
#13	"Membrane Potential, Mitochondrial"[Mesh]	7603	7994
#14	pgc-1 alpha mitochondrial	131	131
#15	"Exercise Therapy"[Mesh]	34503	35538
#16	"Exercise"[Mesh]	133309	136818
#17	High intensity exercise	8842	9145
#18	"Muscle, Skeletal/metabolism"[Mesh]	51602	52827
#19	"Muscle, Skeletal/pathology"[Mesh]	23453	24140
#20	"Muscle, Skeletal/physiopathology"[Mesh]	28488	29150
#21	"Muscle, Skeletal"[Mesh]	220567	223771
#22	High intensity training	7113	7454
#23	"AMP-Activated Protein Kinases"[Mesh]	/	2938
#24	"p38 Mitogen-Activated Protein Kinases"[Mesh]	/	10251
#25	#1 AND #5	113	119
#26	#1 AND #6	4	4
#27	#1 AND #7	10	10
#28	#1 AND #8	8	8
#29	#1 AND #9	1	1
#30	#1 AND #10	9	9
#31	#1 AND #11	1	1
#32	#1 AND #12	0	0
#33	#1 AND #13	6	6

#34	#1 AND #14	0	0
#35	#1 AND #15	291	302
#36	#1 AND #16	575	600
#37	#1 AND #17	9	9
#38	#1 AND #18	21	20
#39	#1 AND #19	28	27
#40	#1 AND #20	194	196
#41	#1 AND #21	353	359
#42	#1 AND #22	8	9
#43	#5 AND #17	66	67
#44	#5 AND #22	46	47
#45	#7 AND #17	30	31
#46	#7 AND #22	28	28
#47	#1 AND #5 AND #16	1	1
#48	#1 AND #20 AND #16	28	28
#49	#1 AND #21 AND #16	44	45
#50	#1 AND #21 AND #5	4	4
#51	#2 OR #3 OR #4 AND #12	27	27
#52	#2 OR #3 OR #4 AND #14	9	9
#53	#2 OR #3 OR #4 AND #11	72	73
#54	#2 OR #3 OR #4 AND #5 AND #22	1	1
#55	#2 OR #3 OR #4 AND #5 AND #15	2	3
#56	#2 OR #3 OR #4 AND #5 AND #16	18	20
#57	#2 OR #3 OR #4 AND #6 AND #22	1	1
#58	#2 OR #3 OR #4 AND #6 AND #15	2	2
#59	#2 OR #3 OR #4 AND #6 AND #16	9	10
#60	#2 OR #3 OR #4 AND #7 AND #16	0	0
#61	#2 OR #3 OR #4 AND #8 AND #16	6	6
#62	#2 OR #3 OR #4 AND #9 AND #16	3	4
#63	#2 OR #3 OR #4 AND #10 AND #16	0	0
#64	#2 OR #3 OR #4 AND #13 AND #16	0	0
#65	#2 OR #3 OR #4 AND #18 AND #16	51	51
#66	#2 OR #3 OR #4 AND #19 AND #16	30	30
#67	#2 OR #3 OR #4 AND #20 AND #22	6	7
#68	#2 OR #3 OR #4 AND #20 AND #15	63	64
#69	#2 OR #3 OR #4 AND #20 AND #16	122	123
#70	#2 OR #3 OR #4 AND #21 AND #22	11	12
#71	#2 OR #3 OR #4 AND #21 AND #15	108	110
#72	#2 OR #3 OR #4 AND #21 AND #16	239	243

#73	#2 OR #3 OR #4 AND #21 AND #17	13	15
#74	#23 AND #1	/	3
#75	#23 AND #5	/	260
#76	#23 AND #6	/	30
#77	#23 AND #16	/	153
#78	#24 AND #1	/	11
#79	#24 AND #5	/	286
#80	#24 AND #6	/	10
#81	#24 AND #16	/	40

Table 2: List of search terms used in Web of Science (WoS).

Search terms, which yielded over 250 hits, were further specified and underlined in following table The literature study was conducted primarily in January and secondary in May as means of control. Based on literature search of January 2016, two extra search terms were added in May (#17 and #18).

	Keywords in WoS	**# January 2016**	**# May 2016**
#1	TOPIC: (multiple sclerosis)	95487	96124
#2	TOPIC: (demyelinating diseases)	10411	10472
#3	TOPIC: (immune system diseases)	54844	55256
#4	TOPIC: (neurodegenerative diseases)	55338	55834
#5	TOPIC: (mitochondria)	124882	125475
#6	TOPIC: (mitochondria, muscle)	12962	13023
#7	TOPIC: (mitochondrial density)	4483	4548
#8	TOPIC: (mitochondrial myopathies)	1235	1237
#9	TOPIC: (citrate synthase)	5492	5506
#10	TOPIC: (mitochondrial ADP, ATP translocases)	45	45
#11	TOPIC: (exercise therapy)	29609	29858
#12	TOPIC: (exercise)	324100	326168
#13	TOPIC: (high intensity exercise)	14614	14749
#14	TOPIC: (high intensity training)	10684	10774
#15	TOPIC: (muscle, skeletal)	171583	172181
#16	TOPIC: (peroxisome proliferator-activated receptor gamma coactivator 1-alpha)	1626	1657
#17	TOPIC: (AMP-Activated Protein Kinases)	/	7278
#18	TOPIC: (p38 Mitogen-Activated Protein Kinases)	/	16538
#19	#1 AND #11	213	220
#20	#1 AND #12	1286	1342
#21	#1 AND #13	30	28
#22	#1 AND #14	35	36
#23	#1 AND #16	6	6
#24	#5 AND #13	169	171
#25	#5 AND #14	90	92
#26	#7 AND #13	32	33
#27	#7 AND #14	26	26
#28	#16 AND #13	44	45
#29	#16 AND #14	30	31
#30	#1 AND #5 AND #12	5	3
#31	#1 AND #20 AND #12	0	0
#32	#1 AND #15 AND #12	71	72

#33	#1 AND #14 AND #5	17	18
#34	#2 OR #3 OR #4 AND #5	3541	3622
#35	#2 OR #3 OR #4 AND #6	264	265
#36	#2 OR #3 OR #4 AND #7	136	138
#37	#2 OR #3 OR #4 AND #8	48	48
#38	#2 OR #3 OR #4 AND #10	2	2
#39	#2 OR #3 OR #4 AND #14	1056	1062
#40	#2 OR #3 OR #4 AND #15	23	23
#41	#2 OR #3 OR #4 AND #11	172	174
#42	#2 OR #3 OR #4 AND #12	875	883
#43	#17 AND #1	/	25
#44	#17 AND #6	/	194
#45	#17 AND #12	/	763
#46	#17 AND #13	/	60
#47	#17 AND #14	/	21
#48	#18 AND #1	/	81
#49	#18 AND #6	/	40
#50	#18 AND #12	/	151
#51	#18 AND #13	/	12
#52	#18 AND #14	/	9

Figure 1: Flowchart illustrating the literature search of this study focussing on pwMS.

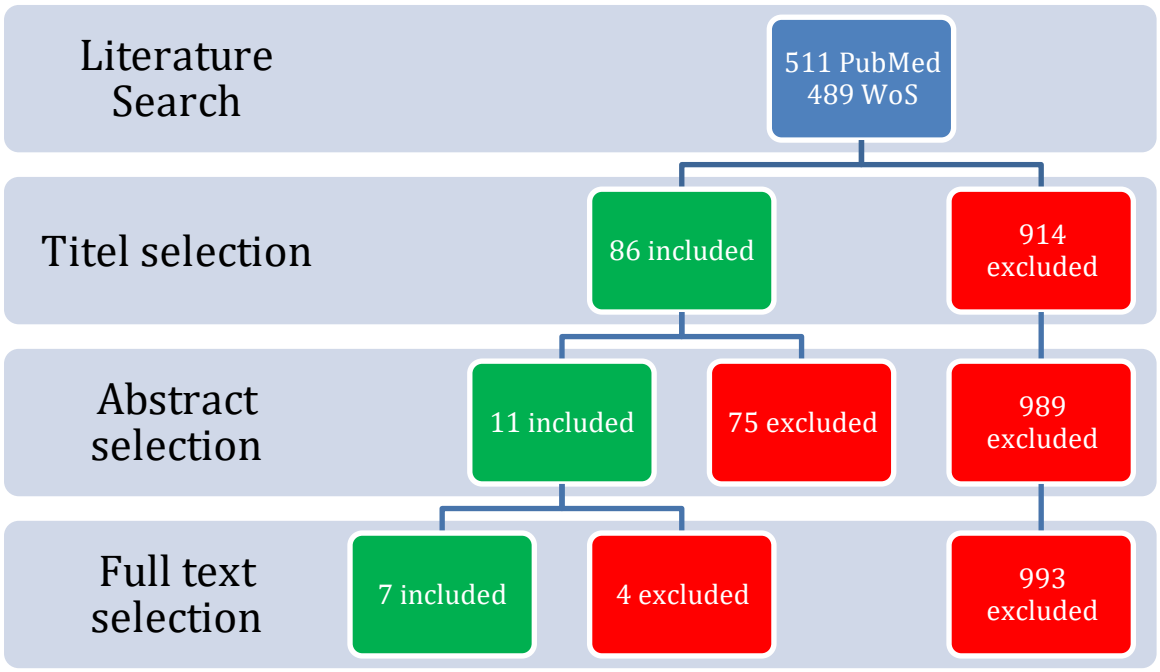

Figure 2: Flowchart illustrating the literature search of this study focussing on healthy subjects and pathologies similar to MS.

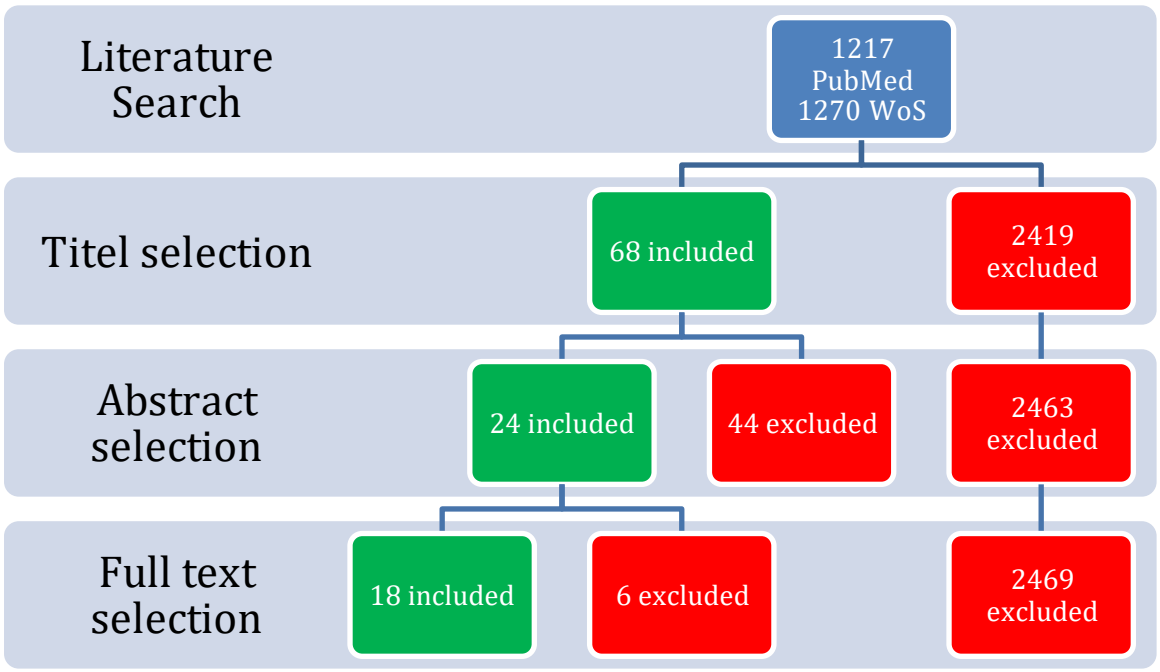

Table 3: Reasons of exclusion for excluded articles concerning pwMS.

Source	Title	Reason of exclusion
MULTIPLE SCLEROSIS		
Andrews, H.E., et al. (2005)	Mitochondrial dysfunction plays a key role in progressive axonal loss in Multiple Sclerosis	No relevant intervention/outcomes
Ban, M., et al. (2008)	Investigation of the role of mitochondrial DNA in multiple sclerosis susceptibility	No relevant intervention/outcomes
Bet, L., et al. (1994)	Multiple sclerosis and mitochondrial myopathy: an unusual combination of diseases	No relevant intervention/outcomes
Blokhin, A., et al. (2008)	Lack of mitochondrial DNA deletions in lesions of multiple sclerosis	No relevant intervention/outcomes
Bosnak-Guclu, M. (2012)	Comparison of functional exercise capacity, pulmonary function and respiratory muscle strength in patients with multiple sclerosis with different disability levels and healthy controls	No relevant intervention
Campbell, G.R. and D.J. Mahad (2012)	Clonal Expansion of Mitochondrial DNA Deletions and the Progression of Multiple Sclerosis	No relevant intervention/outcomes
Campbell, G.R., et al. (2012)	Mitochondrial changes within axons in multiple sclerosis: an update	No relevant intervention/outcomes
Campbell, G.R., J.T. Worrall, and D.J. Mahad (2014)	The central role of mitochondria in axonal degeneration in multiple sclerosis	No relevant intervention/outcomes
Cantalloube, S., et al. (2006)	Strength, postural and gait changes following rehabilitation in multiple sclerosis: a preliminary study	No relevant outcomes
Carter, A.M., et al. (2013)	Pragmatic exercise intervention in people with mild to moderate multiple sclerosis: a randomised controlled feasibility study	No relevant outcomes
Castellano, V., D.I. Patel, and L.J. White (2008)	Cytokine responses to acute and chronic exercise in multiple sclerosis	No relevant outcomes
Castellano, V. and L.J. White (2008)	Serum brain-derived neurotrophic factor response to aerobic exercise in multiple sclerosis	No relevant outcomes
Castro, M.J., et al.	Muscle fiber type-specific myofibrillar	No relevant intervention

(1998)	actomyosin Ca2+ ATPase activity in multiple sclerosis	
Chetta, A., et al. (2004)	Cardiorespiratory response to walk in multiple sclerosis patients	No relevant outcomes
Collet J. et al (2010)	Exercise for multiple sclerosis: a single-blind randomized trial comparing three exercise intensities	No relevant outcomes
Dalgas, U. (2011)	Multiple Sclerosis. Exercise and Chronic Disease: An Evidence-Based Approach	No relevant intervention/outcomes
Dalgas U. et al (2010)	Muscle fibre size increases following resistance training in multiple sclerosis	No relevant outcomes
Dalgas, U., et al. (2013)	Neural drive increases following resistance training in patients with multiple sclerosis	No relevant outcomes
Dawes, H., et al. (2014)	Delayed recovery of leg fatigue symptoms following a maximal exercise session in people with multiple sclerosis	No relevant outcomes
De Riccardis, L., et al. (2015)	Bioenergetics profile of CD4(+) T cells in relapsing remitting multiple sclerosis subjects	No relevant intervention/outcomes
Dutta, R., et al. (2006)	Mitochondrial dysfunction as a cause of axonal degeneration in multiple sclerosis patients	No relevant intervention/outcomes
Fimland, M.S., et al. (2010)	Enhanced neural drive after maximal strength training in multiple sclerosis patients	No relevant outcomes
Finsterer, J., et al. (2012)	Mimicry between mitochondrial disorder and multiple sclerosis	No relevant intervention/outcomes
Fukazawa, T., et al. (1996)	Serum carnitine and disabling fatigue in multiple sclerosis	No relevant intervention
Geurts, J.J. and J. van Horssen (2010)	The brake on neurodegeneration: Increased mitochondrial metabolism in the injured MS spinal cord	No relevant intervention/outcomes
Ghafourifar, P., et al. (2008)	Mitochondria in multiple sclerosis	No relevant intervention/outcomes
Giovannoni, G., et al. (1998)	The potential role of nitric oxide in multiple sclerosis	No relevant intervention/outcomes
Gironi, M., et al. (2014)	The peripheral network between oxidative stress and inflammation in multiple sclerosis	No relevant intervention/outcomes

Gironi, M., et al. (2014)	Oxidative Stress Is Differentially Present in Multiple Sclerosis Courses, Early Evident, and Unrelated to Treatment	No relevant intervention/outcomes
Haider, L., et al. (2011)	Oxidative damage in multiple sclerosis lesions	No relevant intervention/outcomes
Hayes, H., et al. (2008)	Effects of a high-intensity resistance training program on strength, mobility and fatigue in moderately severe individuals with multiple sclerosis	No relevant outcomes
Hayes, H., et al. (2008)	Safety and feasibility of a high-intensity resistance training program for individuals with multiple sclerosis	No relevant outcomes
Hayes, H.A., E. Gappmaier, and P.C. LaStayo (2011)	Effects of high-intensity resistance training on strength, mobility, balance, and fatigue in individuals with multiple sclerosis: a randomized controlled trial	No relevant outcomes
Hogancamp, W.E., M. Rodriguez, and B.G. Weinshenker (1997)	Identification of multiple sclerosis-associated genes	No relevant intervention/outcomes
Hu, J., et al. (2004)	Spectral pattern of total creatine and trimethyl ammonium in multiple sclerosis	No relevant intervention/outcomes
Ickmans, K., et al. (2014)	Recovery of peripheral muscle function from fatiguing exercise and daily physical activity level in patients with multiple sclerosis: a case-control study	No relevant intervention/outcomes
Iniguez, C., et al. (1998)	Mitochondrial respiratory chain deficiency may present as multiple sclerosis	No relevant intervention/outcomes
Inarrea, P., et al. (2014)	Mitochondrial complex enzyme activities and cytochrome C expression changes in multiple sclerosis	No relevant intervention
Kalman, B. (2006)	Role of mitochondria in multiple sclerosis	No relevant intervention/outcomes
Kalman, B., K. Laitinen, and S. Komoly (2007)	The involvement of mitochondria in the pathogenesis of multiple sclerosis	No relevant intervention/outcomes
Kalman, B. and T.P. Leist (2003)	A mitochondrial component of neurodegeneration in multiple sclerosis	No relevant intervention/outcomes
Kalman, B., F.D. Lublin, and H. Alder (1995)	Mitochondrial DNA mutations in multiple sclerosis	No relevant intervention/outcomes

Kentbraun, J.A., et al. (1994)	Postexercise phosphocreatine resynthesis is slowed in multiple sclerosis	No relevant intervention
Kiktenko, A.I., et al. (2005)	Structure of peripheral blood platelets surface in patients with amyotrophic lateral sclerosis and multiple sclerosis	No relevant intervention/outcomes
Koseoglu, B.F., et al. (2006)	Cardiopulmonary and metabolic functions, aerobic capacity, fatigue and quality of life in patients with multiple sclerosis	No relevant intervention
Kostic, M.S., et al. (2013)	Multiple sclerosis and oxidative stress-a clinical perspective	No relevant intervention/outcomes
Krupa, M., et al. (2015)	Increased platelet extracellular vesicle release and platelet mitochondrial bioenergetic changes in relapsing Multiple Sclerosis (MS)	No relevant intervention/outcomes
Lambert, C.P., et al. (2003)	Influence of creatine monohydrate ingestion on muscle metabolites and intense exercise capacity in individuals with multiple sclerosis	No relevant outcomes
Larson, R.D., et al. (2014)	Lower-limb performance disparities: implications for exercise prescription in multiple sclerosis	No relevant intervention/outcomes
Lassmann, H. (2013)	Pathology and disease mechanisms in different stages of multiple sclerosis	No relevant intervention/outcomes
Lassmann, H., J. van Horssen, and D. Mahad (2012)	Progressive multiple sclerosis: pathology and pathogenesis	No relevant intervention/outcomes
Latimer-Cheung, A. E, et al. (2013)	Effects of Exercise Training on Fitness, Mobility, Fatigue, and Health-Related Quality of Life Among Adults With Multiple Sclerosis	No relevant outcomes
Lu, F., et al. (2000)	Oxidative damage to mitochondrial DNA and activity of mitochondrial enzymes in chronic active lesions of multiple sclerosis	No relevant intervention/outcomes
Mahad, D.J., et al. (2008)	Mitochondrial defects in acute multiple sclerosis lesions	No relevant intervention/outcomes
Mahad, D.J., et al. (2009)	Mitochondrial changes within axons in multiple sclerosis	No relevant intervention/outcomes
Malagoni, A.M., et al. (2013)	Muscle oxygen consumption by NIRS and mobility in multiple sclerosis patients	No relevant intervention/outcomes
Malin, S.K., N.	Effect of creatine supplementation on	No relevant intervention

Cotugna, and C.S. Fang (2008)	muscle capacity in individuals with multiple sclerosis	
Mao, P. and P.H. Reddy (2010)	Is multiple sclerosis a mitochondrial disease?	No relevant intervention/outcomes
McLoughlin, J.V., et al. (2014)	Six minutes of walking leads to reduced lower limb strength and increased postural sway in people with Multiple Sclerosis	No relevant intervention
Medina-Perez, C., et al. (2014)	Effects of a resistance training program and subsequent detraining on muscle strength and muscle power in multiple sclerosis patients	No relevant outcomes
Nedeljkovic, U., et al. (2014)	Endurance and resistance training in rehabilitation of patients with multiple sclerosis	No relevant outcomes
Newman, M.A., et al. (2007)	Can aerobic treadmill training reduce the effort of walking and fatigue in people with multiple sclerosis: a pilot study	No relevant outcomes
Ng, A.V., et al. (2004)	Functional relationships of central and peripheral muscle alterations in multiple sclerosis	No relevant intervention/outcomes
Otaegui, D., et al. (2007)	UCP2 and mitochondrial haplogroups as a multiple sclerosis risk factor	No relevant intervention/outcomes
Pilutti, L.A., et al. (2011)	Effects of 12 weeks of supported treadmill training on functional ability and quality of life in progressive multiple sclerosis: a pilot study	No relevant outcomes
Poursadegh Zonouzi, A., et al. (2014)	Mitochondrial complex I gene variations; as a potential genetic risk factor in pathogenesis of multiple sclerosis	No relevant intervention/outcomes
Rietberg, M.B., et al. (2014)	Do Patients With Multiple Sclerosis Show Different Daily Physical Activity Patterns From Healthy Individuals?	No relevant outcomes
Robineau, S., et al. (2005)	Eccentric isokinetic strengthening in hamstrings of patients with multiple sclerosis	No relevant outcomes
Romberg, A., et al. (2004)	Exercise capacity, disability and leisure physical activity of subjects with multiple sclerosis	No relevant intervention/outcomes
Savci, S., et al. (2005)	Six-minute walk distance as a measure of functional exercise capacity in multiple	No relevant outcomes

	sclerosis	
Sawcer, S., P.N. Goodfellow, and A. Compston (1997)	The genetic analysis of multiple sclerosis	No relevant intervention/outcomes
Skjerbaek, A.G., et al. (2014)	Endurance training is feasible in severely disabled patients with progressive multiple sclerosis	No relevant outcomes
Slee, M., et al. (2011)	A novel mitochondrial DNA deletion producing progressive external ophthalmoplegia associated with multiple sclerosis	No relevant intervention/outcomes
Straudi, S., et al. (2014)	A task-oriented circuit training in multiple sclerosis: a feasibility study	No relevant outcomes
Su, K.G., et al. (2009)	Axonal degeneration in multiple sclerosis: the mitochondrial hypothesis	No relevant intervention/outcomes
Szolnoki, Z. (2010)	Common genetic variants of the mitochondrial trafficking system and mitochondrial uncoupling proteins affect the development of two slowly developing demyelinating disorders, leukoaraiosis and multiple sclerosis	No relevant intervention/outcomes
Szolnoki, Z., et al. (2009)	A homozygous genetic variant of mitochondrial uncoupling protein 4 exerts protection against the occurrence of multiple sclerosis	No relevant intervention/outcomes
Taylor, R.W., et al. (1998)	A novel mitochondrial DNA point mutation in the tRNA(Ile) gene: studies in a patient presenting with chronic progressive external ophthalmoplegia and multiple sclerosis	No relevant intervention/outcomes
van Horssen, J., M.E. Witte, and O. Ciccarelli (2012)	The role of mitochondria in axonal degeneration and tissue repair in MS	No relevant intervention/outcomes
Vyshkina, T., et al. (2005)	Genetic variants of Complex I in multiple sclerosis	No relevant intervention/outcomes
Wens, I., et al. (2015)	High intensity training may reverse the fibre type specific decline in myogenic stem cells in multiple sclerosis patients	No full text available
Wens, I., et al., (2013)	Impact of high intensity exercise on endurance capacity, muscle strength and	No full text available

	glucose tolerance in multiple sclerosis	
Witte, M.E., et al. (2014)	Mitochondrial dysfunction contributes to neurodegeneration in multiple sclerosis	No relevant intervention/outcomes
Witte, M.E., et al. (2013)	Reduced expression of PGC-1alpha partly underlies mitochondrial changes and correlates with neuronal loss in multiple sclerosis cortex	No relevant intervention/outcomes
Witte, M.E., et al. (2009)	Enhanced number and activity of mitochondria in multiple sclerosis lesions	No relevant intervention/outcomes
Zambonin, J.L., et al. (2011)	Increased mitochondrial content in remyelinated axons: implications for multiple sclerosis	No relevant intervention/outcomes

Table 4: Reasons of exclusion for excluded articles concerning healthy subjects and pathologies similar to MS.

Source	Title	Reason of exclusion
Healthy subjects		
Barsukova, A.G., D. Bourdette, and M. Forte (2011)	Mitochondrial calcium and its regulation in neurodegeneration induced by oxidative stress	No relevant intervention/outcomes
Bartlett, J.D., et al. (2011)	High-intensity interval running is perceived to be more enjoyable than moderate-intensity continuous exercise: Implications for exercise adherence	No relevant outcomes
Bengtsson, J., et al. (2001)	Mitochondrial transcription factor A and respiratory complex IV increase in response to exercise training in humans	No relevant outcomes
Bilberg, A., M. Ahlmen, and K. Mannerkorpi (2005)	Moderately intensive exercise in a temperate pool for patients with rheumatoid arthritis: a randomized controlled study	No relevant outcomes/population
Bishop, D.J., C. Granata, and N. Eynon (2014)	Can we optimise the exercise training prescription to maximise improvements in mitochondria function and content?	No relevant intervention/outcomes
Campbell, G.R. and D.J. Mahad (2012)	Mitochondrial changes associated with demyelination: consequences for axonal integrity	No relevant intervention/outcomes
Cettolo, V., et al. (2007)	Mitochondrial coupling in humans: assessment of the P/O2 ratio at the onset of calf exercise	No relevant intervention/outcomes
Chiu, S.Y. (2001)	Matching Mitochondria to Metabolic Needs at Nodes of Ranvier	No relevant intervention/outcomes
Ciolac, E.G. (2012)	High-intensity interval training and hypertension: maximizing the benefits of exercise?	No relevant outcomes
Ciolac, E.G., et al. (2010)	Effects of high-intensity aerobic interval training vs. moderate exercise on hemodynamic, metabolic and neuro-humoral abnormalities of young normotensive women at high familial risk for hypertension	No relevant outcomes
Cobley, J. N., Bartlett, J. D., et al. (2012).	PGC-1alpha transcriptional response and mitochondrial adaptation to acute exercise is maintained in skeletal muscle of sedentary	No relevant outcomes

	elderly males	
Da Cruz, S., et al. (2012)	Elevated PGC-1 alpha Activity Sustains Mitochondrial Biogenesis and Muscle Function without Extending Survival in a Mouse Model of Inherited ALS	No relevant population
Daussin, F.N., et al. (2008)	Training at high exercise intensity promotes qualitative adaptations of mitochondrial function in human skeletal muscle	No relevant outcomes
Dibble, L.E., et al. (2006)	High-intensity resistance training amplifies muscle hypertrophy and functional gains in persons with Parkinson's disease	No relevant outcomes
Edge, J., et al. (2013)	Altering the rest interval during high-intensity interval training does not affect muscle or performance adaptations	No relevant outcomes
Egan, B., et al. (2010)	Exercise intensity-dependent regulation of peroxisome proliferator-activated receptor. coactivator-1 alpha mRNA abundance is associated with differential activation of upstream signalling kinases in human skeletal muscle	No relevant intervention
Errea, O., et al. (2015)	The disruption of mitochondrial axonal transport is an early event in neuroinflammation	No relevant intervention/outcomes
Fang, C., D. Bourdette, and G. Banker (2012)	Oxidative stress inhibits axonal transport: implications for neurodegenerative diseases	No relevant intervention/outcomes
Forbes, S.C., J.M. Slade, and R.A. Meyer (2008)	Short-term high-intensity interval training improves phosphocreatine recovery kinetics following moderate-intensity exercise in humans	No relevant outcomes
Gerhart-Hines, Z., et al. (2007)	Metabolic control of muscle mitochondrial function and fatty acid oxidation through SIRT1/PGC-1 alpha	No relevant intervention
Ghiasi, P., et al. (2012)	Mitochondrial complex I deficiency and ATP/ADP ratio in lymphocytes of amyotrophic lateral sclerosis patients	No relevant population
Gibala, M. (2009)	Molecular responses to high-intensity interval exercise	No relevant outcomes
Gibala, M.J. and S.L. McGee (2008)	Metabolic adaptations to short-term high-intensity interval training: A little pain for a lot	No relevant outcomes

	of gain?	
Granata, C., et al. (2016)	Training intensity modulates changes in PGC-1 alpha and p53 protein content and mitochondrial respiration, but not markers of mitochondrial content in human skeletal muscle	Full text unavailable
Harmer, A.R., et al. (2008)	Sprint training increases muscle oxidative metabolism during high-intensity exercise in patients with type 1 diabetes	No relevant population
Harmer, A.R., et al. (2014)	Effects of type 1 diabetes, sprint training and sex on skeletal muscle sarcoplasmic reticulum Ca2+ uptake and Ca2+-ATPase activity	No relevant population
Hogan, V., et al. (2009)	Increase in Mitochondrial Density Within Axons and Supporting Cells in Response to Demyelination in the Plp1 Mouse Model	No relevant population
Hood, M. S., et al. (2011)	Low-volume interval training improves muscle oxidative capacity in sedentary adults	Full text unavailable
Jacobs, R. A., et al. (2013). J Appl Physiol (1985)	Improvements in exercise performance with high-intensity interval training coincide with an increase in skeletal muscle mitochondrial content and function	No relevant outcomes
Kelly, N.A., et al. (2014)	Novel, high-intensity exercise prescription improves muscle mass, mitochondrial function, and physical capacity in individuals with Parkinson's disease	No relevant population
Kilpatrick, M.W., et al. (2015)	Impact of High-Intensity Interval Duration on Perceived Exertion. Medicine and Science in Sports and Exercise	No relevant outcomes
Kiryu-Seo, S., et al. (2010)	Demyelination Increases Axonal Stationary Mitochondrial Size and the Speed of Axonal Mitochondrial Transport	No relevant intervention/outcomes
Larsen, S., et al. (2012)	Biomarkers of mitochondrial content in skeletal muscle of healthy young human subjects	No relevant intervention
Lemmey, A.B., et al. (2009)	Effects of high-intensity resistance training in patients with rheumatoid arthritis: a randomized controlled trial	No relevant population
Lemmey, A.B., et al. (2012)	Are the benefits of a high-intensity progressive resistance training program sustained in rheumatoid arthritis patients? A 3-year	No relevant population

	followup study	
Little, J.P., et al. (2011)	Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes	No relevant population
MacKenzie, J.A. and R.M. Payne (2007)	Mitochondrial protein import and human health and disease	No relevant intervention
Marques-Aleixo, I., et al. (2012)	Physical exercise as a possible strategy for brain protection: evidence from mitochondrial-mediated mechanisms	No relevant intervention/outcomes
Morris, G. and M. Berk (2015)	The many roads to mitochondrial dysfunction in neuroimmune and neuropsychiatric disorders	No relevant intervention/outcomes
Ohno, N., et al. (2014)	Mitochondrial immobilization mediated by syntaphilin facilitates survival of demyelinated axons	No relevant intervention/outcomes
Palomo, G.M. and G. Manfredi (2015)	Exploring new pathways of neurodegeneration in ALS: The role of mitochondria quality control	No relevant intervention/outcomes/population
Picard, M., R.T. Hepple, and Y. Burelle (2012)	Mitochondrial functional specialization in glycolytic and oxidative muscle fibers: tailoring the organelle for optimal function	No relevant intervention/outcomes
Psilander, N., et al. (2010)	Mitochondrial gene expression in elite cyclists: effects of high-intensity interval exercise	No relevant population
Rasmussen, U.F., et al. (2001)	The effect of high-intensity exhaustive exercise studied in isolated mitochondria from human skeletal muscle	No relevant outcomes
Rezaee, A.R., et al. (2013)	Mitochondrial and nuclear genes as the cause of complex I deficiency	No relevant intervention/outcomes
Rose, M.H., et al. (2013)	Effects of training and weight support on muscle activation in Parkinson's disease	No relevant outcomes/population
Ryan, T.E., J.T. Brizendine, and K.K. McCully (2013)	A comparison of exercise type and intensity on the noninvasive assessment of skeletal muscle mitochondrial function using near-infrared spectroscopy	No relevant outcomes
Safdar, A., et al. (2011)	Exercise Increases Mitochondrial PGC-1 alpha Content and Promotes Nuclear-Mitochondrial Cross-talk to Coordinate Mitochondrial Biogenesis	No relevant intervention

Saft, C., et al. (2005)	Mitochondrial impairment in patients and asymptomatic mutation carriers of Huntington's disease	No relevant population
Sahlin, K., et al. (2007)	The potential for mitochondrial fat oxidation in human skeletal muscle influences whole body fat oxidation during low-intensity exercise	No relevant outcomes
Sahlin, K., et al. (2010)	Ultraendurance exercise increases the production of reactive oxygen species in isolated mitochondria from human skeletal muscle	No relevant outcomes
Schoenfeld, R., et al. (2010)	Oligodendroglial differentiation induces mitochondrial genes and inhibition of mitochondrial function represses oligodendroglial differentiation	No relevant intervention/outcomes
Simoneau, J.A., et al. (1985)	Human skeletal muscle fiber type alteration with high-intensity intermittent training	No relevant outcomes
Tadaishi, M., et al. (2011)	Skeletal muscle-specific expression of PGC-1alpha-b, an exercise-responsive isoform, increases exercise capacity and peak oxygen uptake	No relevant intervention
Toledo, F.G. and B.H. Goodpaster (2013)	The role of weight loss and exercise in correcting skeletal muscle mitochondrial abnormalities in obesity, diabetes and aging	No relevant intervention/population
Tollback, A., et al. (1999)	Effects of high resistance training in patients with myotonic dystrophy	No relevant intervention/outcomes/ population
Tonkonogi, M., et al. (2000)	Mitochondrial function and antioxidative defence in human muscle: effects of endurance training and oxidative stress	No relevant outcomes
Tonkonogi, M., et al. (1999)	Mitochondrial function in human skeletal muscle is not impaired by high intensity exercise	No relevant outcomes
Tuan, T.C., et al. (2008)	Deleterious effects of short-term, high-intensity exercise on immune function: evidence from leucocyte mitochondrial alterations and apoptosis	No relevant outcomes
Tucker, W.J. (2015)	Physiological responses to high-intensity interval exercise differing in interval duration	No relevant outcomes
Vielhaber, S., et al. (2000)	Mitochondrial DNA abnormalities in skeletal muscle of patients with sporadic amyotrophic	No relevant intervention/population

	lateral sclerosis	
Walsh, B., et al. (2001)	The role of phosphorylcreatine and creatine in the regulation of mitochondrial respiration in human skeletal muscle	No relevant intervention
Wright, D.C., et al. (2007	Exercise-induced mitochondrial biogenesis begins before the increase in muscle PGC-1 alpha expression	No relevant intervention
Yan, Z., et al. (2011)	Regulation of exercise-induced fiber type transformation, mitochondrial biogenesis, and angiogenesis in skeletal muscle	No relevant intervention
Zambonin, J., et al. (2010)	Identification and investigation of mitochondria lacking cytochrome c oxidase activity in axons	No relevant intervention
Zhang, C.L., et al. (2010)	Activity-Dependent Regulation of Mitochondrial Motility by Calcium and Na/K-ATPase at Nodes of Ranvier of Myelinated Nerves	No relevant intervention/outcomes
Zoll, J., et al. (2002)	Physical activity changes the regulation of mitochondrial respiration in human skeletal muscle	No relevant intervention/outcomes

Table 5: Quality assessment: Cochrane checklist for randomized controlled trials

Criteria:

1. Random allocation? (selection bias)
2. Allocation concealment? (selection bias)
3. Blinding of participants (performance bias)
4. Blinding of personnel (performance bias)
5. Blinding of outcome assessment (detection bias) (patient-reported outcomes)
6. Baseline characteristics different between groups? (selection bias)
7. Sufficient proportion of follow up of participants (attrition bias)
8. All participants analysed in the allocated group? (attrition bias)
9. All groups are treated the same despite the intervention?
10. Conclusion: are results valid and applicable?

Author	Criterion 1	Criterion 2	Criterion 3	Criterion 4	Criterion 5	Criterion 6	Criterion 7	Criterion 8	Criterion 9	Criterion 10	Score
Burgomaster, K. A., et al. (2008)	No	No	No	No	?	No	Yes	Yes	Yes	Yes	4/10
Fernstrom, M., et al. (2004)	?	?	No	No	?	No	Yes	Yes	Yes	Yes	5/10
Wens I., et al. (2015)	Yes	Yes	No	No	?	No	Yes	Yes	Yes and No	Yes	5-6/10

Yes and No: one sedentary control group with no intervention and 2 groups with 2 different interventions

Table 6: Quality assessment standard for a cross-over study

Matched work high-intensity interval and continuous running induce similar increases in PGC-1α mRNA, AMPK, p38, and p53 phosphorylation in human skeletal muscle

JD. Bartlett, CH. Joo, T-S. Jeong, J Louhelainen, AJ. Cochran, MJ. Gibala, W Gregson, GL. Close, B Drust, JP. Morton

Item	Description	Scoring	score
1. Appropriate cross-over design	Three points are considered: (1) the condition of the patients should be chronic and stable; (2) the intervention should not provide permanent change, but rather temporary relief; (3) the effect of the first intervention should not last into the second treatment period.	Low: all the three points are absolutely correct;	UNCLEAR
		Unclear: it hard to judge because some information was missing or ambiguous;	
		High: one or more points are incorrect.	
2. Randomized treatment order	The order of receiving treatments should be randomized adequately.	Low: the method is appropriate and clearly described;	UNCLEAR
		Unclear: it is described as “randomized”, but it is hard to judge whether the implementation was adequate because some information (method, etc.) was not provided;	
		High: the method is inappropriate, or no randomization is applied.	
3. Carry-over effect	The authors should evaluate the carry-over effect and provide relevant information clearly.	Low: carry-over effect was evaluated and the results showed no carry-over effect;	UNCLEAR
		Unclear: carry-over effect was not evaluated, and it is hard for evaluators to judge;	

		High: carry-over effect was evaluated and the results showed apparent carry-over effect, or indicated evidently from some other provided information.	
4. Unbiased data	That only first-period data are available is considered a risk of bias.	Low: data for every period are provided;	HIGH
		Unclear: data are unavailable for part of outcomes, or only analytical results are provided and it is hard to judge whether the results are 66nalysed based only on data from the first-period or every period.	
		High: only first-period data are available.	
5. Allocation concealment	The study should apply appropriate approaches to ensure the allocation sequence is concealed.	Low: allocation sequence was concealed adequately by appropriate methods;	UNCLEAR
		Unclear: concealment approaches were not described, or relevant information was ambiguous;	
		High: no approaches to allocation concealment were used, or concealed inadequately.	
6. Blinding	The study should apply a proper blinding method to prevent performance and detection bias. Those involved in blinding (participants, doctors, measurers, or analysts) depends on the particularity of the	Low: appropriate blinding method was applied; No blinding, but the outcome and the outcome measurement are not likely to be influenced by lack of blinding;	LOW
		Unclear: relevant information was not provided;	

	studies.	High: no blinding method was applied, or applied incorrectly, or ineffectively, which very likely affected the outcome.	
7. Incomplete outcome data	The authors should provide relevant information about the completeness of outcome data, including the level of incompleteness, reasons, and analytic method to tackle these data shortcomings, etc.	Low: no missing outcome data, or the reason is acceptable, or missing outcome data were appropriate 67nalysed;	LOW
		Unclear: it is hard to judge because some information was not provided;	
		High: missing outcome data existed and the reasons were unacceptable, and the analytic method was inappropriate.	
8. Selective outcome reporting	The authors should report all the outcomes fully. Selective reporting of part of outcomes or data for an outcome or subsets of the data or analyses using the same data and etc. should be avoided.	Low: fully reported;	LOW
		Unclear: it is hard to judge due to the unavailability of some original information;	
		High: the reports of the study suggest a high risk of selective outcome reporting.	
9. Other bias	Any other potential risk of bias that may affect the quality of cross-over studies.	Low: the study is apparently free of other problems;	UNCLEAR
		Unclear: whether certain problems existed and led to a risk of bias is uncertain;	
		High: high risk of bias existed due to evident problems.	

NOTE: the standard was summarized from the Cochrane Collaboration's tool for assessing risk of bias and the Cochrane handbook`s suggestions for assessing risk of bias in cross-over studies. The assessment of some items, especially items 5–8, are almost the same as that described in Cochrane Collaboration's tool for assessing the risk of bias.

Intermittent and continuous high-intensity exercise training induce similar acute but different chronic muscle adaptations (study 1: acute investigation)

AJ.R. Cochran, ME. Percival, S Tricarico, JP. Little, N Cermak, JB. Gillen, MA. Tarnopolsky, MJ. Gibala

Item	Description	Scoring	score
1. Appropriate cross-over design	Three points are considered: (1) the condition of the patients should be chronic and stable; (2) the intervention should not provide permanent change, but rather temporary relief; (3) the effect of the first intervention should not last into the second treatment period	Low: all the three points are absolutely correct;	HIGH
		Unclear: it hard to judge because some information was missing or ambiguous;	
		High: one or more points are incorrect.	
2. Randomized treatment order	The order of receiving treatments should be randomized adequately.	Low: the method is appropriate and clearly described;	HIGH
		Unclear: it is described as "randomized", but it is hard to judge whether the implementation was adequate because some information (method, etc.) was not provided;	
		High: the method is inappropriate, or no randomization is applied.	
3. Carry-over effect	The authors should evaluate the carry-over effect and provide relevant information clearly.	Low: carry-over effect was evaluated and the results showed no carry-over effect;	UNCLEAR
		Unclear: carry-over effect was not evaluated, and it is hard for evaluators to judge;	
		High: carry-over effect was evaluated and the results showed	

		apparent carry-over effect, or indicated evidently from some other provided information.	
4. Unbiased data	That only first-period data are available is considered a risk of bias.	Low: data for every period are provided;	LOW
		Unclear: data are unavailable for part of outcomes, or only analytical results are provided and it is hard to judge whether the results are 69nalysed based only on data from the first-period or every period.	
		High: only first-period data are available.	
5. Allocation concealment	The study should apply appropriate approaches to ensure the allocation sequence is concealed.	Low: allocation sequence was concealed adequately by appropriate methods;	HIGH
		Unclear: concealment approaches were not described, or relevant information was ambiguous;	
		High: no approaches to allocation concealment were used, or concealed inadequately.	
6. Blinding	The study should apply a proper blinding method to prevent performance and detection bias. Those involved in blinding (participants, doctors, measurers, or analysts) depends on the particularity of the studies.	Low: appropriate blinding method was applied; No blinding, but the outcome and the outcome measurement are not likely to be influenced by lack of blinding;	LOW
		Unclear: relevant information was not provided;	
		High: no blinding method was applied, or applied incorrectly, or ineffectively, which very likely affected the outcome.	

7. Incomplete outcome data	The authors should provide relevant information about the completeness of outcome data, including the level of incompleteness, reasons, and analytic method to tackle these data shortcomings, etc.	Low: no missing outcome data, or the reason is acceptable, or missing outcome data were appropriate 70nalysed;	LOW
		Unclear: it is hard to judge because some information was not provided;	
		High: missing outcome data existed and the reasons were unacceptable, and the analytic method was inappropriate.	
8. Selective outcome reporting	The authors should report all the outcomes fully. Selective reporting of part of outcomes or data for an outcome or subsets of the data or analyses using the same data and etc. should be avoided.	Low: fully reported;	LOW
		Unclear: it is hard to judge due to the unavailability of some original information;	
		High: the reports of the study suggest a high risk of selective outcome reporting.	
9. Other bias	Any other potential risk of bias that may affect the quality of cross-over studies.	Low: the study is apparently free of other problems;	UNCLEAR
		Unclear: whether certain problems existed and led to a risk of bias is uncertain;	
		High: high risk of bias existed due to evident problems.	

Rapid exercise-induced changes in PGC-1α mRNA and protein in human skeletal muscle

Anila S. Mathai, Arend Bonen, Carley R. Benton, D. L. Robinson, and Terry E. Graham

Item	Description	Scoring	score
1. Appropriate cross-over design	Three points are considered: (1) the condition of the patients should be chronic and stable; (2) the intervention should not provide permanent change, but rather temporary relief; (3) the effect of the first intervention should not last into the second treatment period	Low: all the three points are absolutely correct;	LOW
		Unclear: it hard to judge because some information was missing or ambiguous;	
		High: one or more points are incorrect.	
2. Randomized treatment order	The order of receiving treatments should be randomized adequately.	Low: the method is appropriate and clearly described;	LOW
		Unclear: it is described as "randomized", but it is hard to judge whether the implementation was adequate because some information (method, etc.) was not provided;	
		High: the method is inappropriate, or no randomization is applied.	
3. Carry-over effect	The authors should evaluate the carry-over effect and provide relevant information clearly.	Low: carry-over effect was evaluated and the results showed no carry-over effect;	UNCLEAR
		Unclear: carry-over effect was not evaluated, and it is hard for evaluators to judge;	
		High: carry-over effect was evaluated and the results showed apparent carry-over effect, or indicated evidently from some other	

		provided information.	
4. Unbiased data	That only first-period data are available is considered a risk of bias.	Low: data for every period are provided;	LOW
		Unclear: data are unavailable for part of outcomes, or only analytical results are provided and it is hard to judge whether the results are 72nalysed based only on data from the first-period or every period.	
		High: only first-period data are available.	
5. Allocation concealment	The study should apply appropriate approaches to ensure the allocation sequence is concealed.	Low: allocation sequence was concealed adequately by appropriate methods;	UNCLEAR
		Unclear: concealment approaches were not described, or relevant information was ambiguous;	
		High: no approaches to allocation concealment were used, or concealed inadequately.	
6. Blinding	The study should apply a proper blinding method to prevent performance and detection bias. Those involved in blinding (participants, doctors, measurers, or analysts) depends on the particularity of the studies.	Low: appropriate blinding method was applied; No blinding, but the outcome and the outcome measurement are not likely to be influenced by lack of blinding;	UNCLEAR
		Unclear: relevant information was not provided;	
		High: no blinding method was applied, or applied incorrectly, or ineffectively, which very likely affected the outcome.	

7. Incomplete outcome data	The authors should provide relevant information about the completeness of outcome data, including the level of incompleteness, reasons, and analytic method to tackle these data shortcomings, etc.	Low: no missing outcome data, or the reason is acceptable, or missing outcome data were appropriate 73nalysed;	LOW
		Unclear: it is hard to judge because some information was not provided;	
		High: missing outcome data existed and the reasons were unacceptable, and the analytic method was inappropriate.	
8. Selective outcome reporting	The authors should report all the outcomes fully. Selective reporting of part of outcomes or data for an outcome or subsets of the data or analyses using the same data and etc. should be avoided.	Low: fully reported;	LOW
		Unclear: it is hard to judge due to the unavailability of some original information;	
		High: the reports of the study suggest a high risk of selective outcome reporting.	
9. Other bias	Any other potential risk of bias that may affect the quality of cross-over studies.	Low: the study is apparently free of other problems;	UNCLEAR
		Unclear: whether certain problems existed and led to a risk of bias is uncertain;	
		High: high risk of bias existed due to evident problems.	

Table 7: Quality assessment: quasi experimental studies without control group: Self-made checklist (based on Downs and Black checklist 1998)

Criteria:

1. Is the hypothesis/aim/objective of the study clearly described?
2. Are the main outcomes to be measured clearly described in the introduction or methods section?
3. Are the characteristics of the individuals included in the study clearly described?
4. Are the interventions of interest clearly described?
5. Are the distribution of principal confounders in each group of subjects to be compared clearly described?
6. Are the main findings of the study clearly described?
7. Does the study provide estimates of the random variability in the data for the main outcomes?
8. Have all important adverse events that may be a consequence of the intervention been reported?
9. Have the characteristics of patients lost to follow-up been described?
10. Have actual probability values been reported (e.g. 0.035 rather than <0.05) for the main outcomes except where the probability value is less than 0.001?
11. Were the subjects asked to participate in the study representative of the entire population from which they were recruited?
12. Were those subjects who were prepared to participate representative of the entire population from which they were recruited?
13. Were the staff, places, and facilities where the patients were treated, representative of the treatment the majority of patients receive?
14. Was an attempt made to blind study subjects to the intervention they have received?
15. Was an attempt made to blind those measuring the main outcomes of the intervention?
16. In trials and cohort studies, do the analyses adjust for different lengths of follow-up of patients, or in case-control studies, is the time period between the intervention and outcome the same for cases and controls?
17. Were the statistical tests used to assess the main outcomes appropriate?
18. Was compliance with the intervention/s reliable?
19. Were the main outcome measures used accurate (valid and reliable)?
20. Were losses of patients to follow-up taken into account?

Author	Criterion 1	Criterion 2	Criterion 3	Criterion 4	Criterion 5	Criterion 6	Criterion 7	Criterion 8	Criterion 9	Criterion 10	Criterion 11	Criterion 12	Criterion 13	Criterion 14	Criterion 15	Criterion 16	Criterion 17	Criterion 18	Criterion 19	Criterion 20	Score
Edgett B. A., et al. (2013)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	UTD	UTD	No	No	No	UTD	No	No	Yes	Yes	Yes	Yes	UTD	11/20
Gibala M. J., et al. (2009)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	UTD	UTD	Yes	Yes	Yes	UTD	No	No	Yes	Yes	Yes	Yes	UTD	14/20
Gurd B. J., et al. (2010)	Yes	Yes	Yes	Yes	Yes	yes	Yes	UTD	No	No	No	Yes	UTD	No	UTD	Yes	Yes	Yes	Yes	UTD	12/20
Jacobs R. A., et al. (2013)	Yes	Yes	Yes	Yes	Yes	Yes	No	No	No	Yes	UTD	UTD	UTD	No	No	Yes	Yes	Yes	Yes	UTD	11/20
Larsen S., et al. (2015)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	No	No	Yes	Yes	Yes	Yes	UTD	16/20
Larsen, R. G., (2013)	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	No	Yes	No	No	UTD	No	Yes	Yes	Yes	Yes	Yes	UTD	13/20
Larsen, R. G., (2014)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	UTD	Yes	UTD	UTD	Yes	Yes	Yes	Yes	UTD	15/20
Little J. P., et al. (2011)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	No	Yes	Yes	UTD	Yes	UTD	UTD	Yes	Yes	UTD	Yes	UTD	13/20
Little J. P., et al. (2010)	Yes	Yes	Yes	Yes	Yes	Yes	No	No	No	Yes	No	No	UTD	No	No	Yes	Yes	Yes	Yes	UTD	11/20

Perry C. G., et al. (2008)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	No	No	Yes	No	No	Yes	Yes	Yes	Yes	Yes	15/20
Stepto N. K., et al. (2012)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	UTD	UTD	Yes	No	No	UTD	No	No	Yes	Yes	Yes	Yes	UTD	12/20
Talanian, J. L. (2010)	Yes	Yes	Yes	Yes	No	Yes	No	No	No	No	No	No	UTD	No	No	Yes	Yes	UTD	Yes	No	8/21
Vincent G., et al. (2015)	Yes	Yes	Yes	Yes	Yes	Yes	Yes	UTD	No	No	No	No	UTD	No	No	Yes	Yes	Yes	Yes	UTD	11/20

UTD: unable to determine/not described

Table 8: STROBE 2007 (v4) Statement—Checklist of items that should be included in reports of cross-sectional studies

Multiple sclerosis affects skeletal muscle characteristics

I Wens, U Dalgas, F Vandenabeele, M Krekels, L Grevendonk, B O. Eijnde

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1	Yes
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	1	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	1	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses	1	Yes
Methods				
Study design	4	Present key elements of study design early in the paper	1-2	Yes
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	1-2	Yes
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	1-2	Yes
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	1-2	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	1-2	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	2	Yes

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	2	Yes
		(*b*) Describe any methods used to examine subgroups and interactions	2	Yes
		(*c*) Explain how missing data were addressed		No
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		No
		(*e*) Describe any sensitivity analyses	2	Yes
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	2-3	Yes
		(b) Give reasons for non-participation at each stage		/
		(c) Consider use of a flow diagram		No
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	3	Yes
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures		No
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	2	Yes
		(*b*) Report category boundaries when continuous variables were categorized		No
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	2-3	Yes
Discussion				
Key results	18	Summarise key results with reference to study objectives	3-4	Yes
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both	4	Yes

		direction and magnitude of any potential bias		
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	3-4	Yes
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	5	Yes

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Slowed exercise-onset VO2 kinetics during submaximal endurance exercise in subjects with multiple sclerosis
D Hansen, I Wens, L Kosten, K Verboven, B O. Eijnde

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	87	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	87	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses	87	Yes
Methods				
Study design	4	Present key elements of study design early in the paper	88	Yes
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	88-89	Yes
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	88	Yes
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	88-89	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	88-89	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why		No

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	89	Yes
		(*b*) Describe any methods used to examine subgroups and interactions	89	Yes
		(*c*) Explain how missing data were addressed		/
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		/
		(*e*) Describe any sensitivity analyses		No
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed		/
		(b) Give reasons for non-participation at each stage		/
		(c) Consider use of a flow diagram		/
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	90	Yes
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures		/
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	90-91	Yes
		(*b*) Report category boundaries when continuous variables were categorized	90-91	Yes
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	90-91	Yes
Discussion				
Key results	18	Summarise key results with reference to study objectives	93-94	Yes
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both		No

		direction and magnitude of any potential bias		
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	93	Yes
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	94	Yes

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Strength, skeletal muscle composition, and enzyme activity in multiple sclerosis.
Kent-Braun JA1, Ng AV, Castro M, Weiner MW, Gelinas D, Dudley GA, Miller RG.

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	1998	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	1998	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses	1998	Yes
Methods				
Study design	4	Present key elements of study design early in the paper		No
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	1998-2000	Yes
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants		No
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	1998-2000	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	1998-2000	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why		No

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	2000	Yes
		(*b*) Describe any methods used to examine subgroups and interactions	2000	Yes
		(*c*) Explain how missing data were addressed		/
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		No
		(*e*) Describe any sensitivity analyses		/
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	2000-2002	Yes
		(b) Give reasons for non-participation at each stage		No
		(c) Consider use of a flow diagram		No
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	1998	Yes
		(b) Indicate number of participants with missing data for each variable of interest		No
Outcome data	15*	Report numbers of outcome events or summary measures	2000-2002	Yes
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	2000-2002	Yes
		(*b*) Report category boundaries when continuous variables were categorized		/
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	2001	Yes
Discussion				
Key results	18	Summarise key results with reference to study objectives	2002-	Yes

			2003	
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias		No
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence		No
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	2003-2004	Yes

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	65	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	65-66	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses		No
Methods				
Study design	4	Present key elements of study design early in the paper		No
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection		No
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	66	Yes
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	66	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	66	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why		No

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	66	Yes
		(*b*) Describe any methods used to examine subgroups and interactions		No
		(*c*) Explain how missing data were addressed		/
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		/
		(*e*) Describe any sensitivity analyses		/
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed		No
		(b) Give reasons for non-participation at each stage		No
		(c) Consider use of a flow diagram		/
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders		No
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures	67	Yes
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	67	Yes
		(*b*) Report category boundaries when continuous variables were categorized		No
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses		/
Discussion				
Key results	18	Summarise key results with reference to study objectives	68	Yes
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both		No

		direction and magnitude of any potential bias		
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence		No
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based		No

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

No excess of mitochondrial DNA deletions within muscle in progressive multiple sclerosis
GR. Campbell, AK. Reeve, I Ziabreva, R Reynolds, DM. Turnbull, DJ. Mahad

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	1858	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	1858	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses		No
Methods				
Study design	4	Present key elements of study design early in the paper	1859	Yes
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection		No
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	1859	Yes
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	1859-1860	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	1859-1860	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	1859	Yes

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	1861	Yes
		(*b*) Describe any methods used to examine subgroups and interactions	1861	Yes
		(*c*) Explain how missing data were addressed		/
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		/
		(*e*) Describe any sensitivity analyses		/
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed		No
		(b) Give reasons for non-participation at each stage		/
		(c) Consider use of a flow diagram		/
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders		No
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures		
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	1961-1862	Yes
		(*b*) Report category boundaries when continuous variables were categorized		No
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses		No
Discussion				
Key results	18	Summarise key results with reference to study objectives	1862-	Yes

			1865	
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias		No
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	1862-1865	Yes
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	1865	Yes

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Altered signalling for mitochondrial and myofibrillar biogenesis in skeletal muscles of patients with multiple sclerosis
D Hansen, I Wens, F Vandenabeele, K Verboven, BO. Eijnde

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	70	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	70-71	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses	71	Yes
Methods				
Study design	4	Present key elements of study design early in the paper	71	Yes
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	72	Yes
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	72	Yes
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	72-73	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	72-73	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why		No

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	73	Yes
		(*b*) Describe any methods used to examine subgroups and interactions	73	Yes
		(*c*) Explain how missing data were addressed		/
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		/
		(*e*) Describe any sensitivity analyses	73	Yes
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	73-75	Yes
		(b) Give reasons for non-participation at each stage		/
		(c) Consider use of a flow diagram		/
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	74	Yes
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures	73-75	Yes
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	73-75	Yes
		(*b*) Report category boundaries when continuous variables were categorized	73-75	Yes
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	75	Yes
Discussion				
Key results	18	Summarise key results with reference to study objectives	75-77	Yes
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	77	Yes

Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	75-77	Yes
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based		No

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Intermittent and continuous high-intensity exercise training induce similar acute but different chronic muscle adaptations (study part 2)
AJ.R. Cochran, ME. Percival, S Tricarico, JP. Little, N Cermak, JB. Gillen, MA. Tarnopolsky, MJ. Gibala

Section/Topic	Item #	Recommendation	Page No.	Yes/No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract		No
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	782	Yes
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	783	Yes
Objectives	3	State specific objectives, including any prespecified hypotheses	783	Yes
Methods				
Study design	4	Present key elements of study design early in the paper	784	Yes
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	784-785	Yes
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants		No
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	785	Yes
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	785	Yes
Bias	9	Describe any efforts to address potential sources of bias		No
Study size	10	Explain how the study size was arrived at		No
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why		No

Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	786	Yes
		(*b*) Describe any methods used to examine subgroups and interactions		No
		(*c*) Explain how missing data were addressed		No
		(*d*) If applicable, describe analytical methods taking account of sampling strategy		/
		(*e*) Describe any sensitivity analyses		No
Results				
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed		No
		(b) Give reasons for non-participation at each stage		/
		(c) Consider use of a flow diagram		/
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders		No
		(b) Indicate number of participants with missing data for each variable of interest		/
Outcome data	15*	Report numbers of outcome events or summary measures	787	Yes
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	787	Yes
		(*b*) Report category boundaries when continuous variables were categorized	787	Yes
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period		/
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses		No
Discussion				
Key results	18	Summarise key results with reference to study objectives	788-790	Yes
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both	788-790	Yes

		direction and magnitude of any potential bias		
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	788-790	Yes
Generalisability	21	Discuss the generalisability (external validity) of the study results		No
Other information				
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	791	Yes

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Table 9.1: Overview included articles – type of article, subjects, used techniques and conclusion

Year	author	title	Type of article	subjects	Used techniques	conclusion
2012	Bartlett, J. D., et al.	Matched work high-intensity interval and continuous running induce similar increases in PGC-1alfa mRNA, AMPK, p38 and p53 phosphorylation in human skeletal muscle	Randomized crossover design	10 active men	Test the hypothesis that acute high-intensity interval running induces greater activation of signalling pathways associated with mitochondrial biogenesis compared to moderate-intensity continuous running. Muscle biopsies from vastus lateralis muscle were used.	Acute HIIT and CONT running induces similar activation of molecular signalling pathways associated with regulation of mitochondrial biogenesis.
2008	Burgomaster, K. A., et al.	Similar metabolic adaptations during exercise after low volume sprint interval and traditional endurance training in humans	Randomized controlled trial	20 healthy men and women (active but untrained)	Two experimental trials, before and after a 6 week exercise training programme. Needle biopsy samples of vastus lateralis muscle at rest and immediately after exercise. Heart rate	These data suggest that high-intensity interval training is a time-efficient strategy to increase skeletal muscle oxidative capacity and induce specific metabolic adaptations during exercise that are comparable to traditional ET.

					Expired gases were collected for the determination of VO2 ,VCO2 and respiratory exchange ratio (RER).	
2013	Campbell, G. R., et al.	No excess of mitochondrial DNA deletions within muscle in progressive multiple sclerosis	RCT (cross sectional study)	17 progressive MS cases + 15 controls	Studied muscle (paraspinal) and explored mitochondria in single fibres. Histochemistry, immunohistochemistry, laser microdissection, real-time polymerase chain reaction (PCR), long-range PCR and sequencing were used to resolve the single muscle fibres.	The findings do not provide support to the existence of a diffuse mitochondrial abnormality involving multiple systems in MS. Understanding the cause(s) of the CNS mitochondrial dysfunction in progressive MS remains a research priority.
2014	Cochran, A. J., et al.	Intermittent and continuous high-intensity exercise training induce similar acute but different chronic muscle adaptations	Study 1: Randomized crossover design	Study 1: 8 men	Study 1: examined whether the activation of signalling cascades linked to mitochondrial biogenesis was dependent on the manner in which an acute high-intensity exercise stimulus was	The intermittent nature of the stimulus is important for maximizing skeletal muscle adaptations to low-volume, all-out HIIT. Despite the lack of skeletal muscle mitochondrial adaptation, our data show that a training programme based on a brief bout of high-intensity exercise, which lasted <10min per

			Study 2: RCT (cross sectional study)	Study 2: 5 men and 4 women (young and healthy)	applied (INT or CONT). → AMPK, p38, PGC-1α mRNA expression Study 2: Investigated whether six weeks of a CONT protocol would increase skeletal muscle mitochondrial content to a similar extent as INT.	session, 3x/W for 6W, improved peak oxygen uptake in young healthy subjects.
2013	Edgett, B. A., et al.	Dissociation of increases in PGC-1alfa and its regulators from exercise intensity and muscle activation following acute exercise	Quasi experimental study without control group	8 lean healthy men	Muscle activation as well as changes in PGC-1α following high-intensity interval exercise.	Intensity-dependent increases in PGC-1α mRNA following submaximal exercise are largely due to increases in muscle recruitment. As well, the blunted response of PGC-1α mRNA expression following supramaximal exercise may indicate that signalling mediated activation of PGC-1α may also be blunted. We also identify that increases in PDK4, SIRT1, and RIP40 mRNA following acute exercise are dissociated from exercise intensity and muscle activation, while increases in EGR1 are augmented with supramaximal HIIE.

2003	Fernstrom, M., et al.	Effects of acute and chronic endurance exercise on mitochondrial uncoupling in human skeletal muscle	RCT	17 healthy subjects	Subjects were investigated with muscle biopsies before and after acute exercise (75 min of cycling at 70% of ˙VO2peak) or 6 weeks endurance training. Mitochondria were isolated and respiration measured in the absence (UCR or state 4) and presence of ADP (coupled respiration or state 3). Protein expression of UCP3 and ANT was measured with Western blotting.	UCP3 protein and UCR decrease after endurance training when related to mitochondrial volume. These changes may prevent excessive basal thermogenesis. Acute exercise enhances mitochondrial resistance to Ca2+ overload but does not influence UCR or protein expression of UCP3 and ANT. The increased Ca2+ resistance may prevent mitochondrial degradation and the mechanism needs to be further explored.
2008	Gibala, M. J., et al.	Brief intense interval exercise activates AMPK and p38 MAPK signalling and increases the expression of PGC-1alfa in human skeletal muscle	Quasi experimental study without control group	6 healthy active young men	We tested the hypothesis that an acute session of intense intermittent cycle exercise would activate signalling cascades linked to mitochondrial biogenesis in human skeletal muscle. → Muscle biopsies	Signalling through AMPK and p38 MAPK to PGC-1α may explain in part the metabolic remodelling induced by low-volume intense interval exercise, including mitochondrial biogenesis and an increased capacity for glucose and fatty acid oxidation.

2010	Gurd, B. J., et al.	High-intensity interval training increases SIRT1 activity in human skeletal muscle	Quasi experimental study without control group	9 subject (3 females, 6 males)	The effects of training on SIRT1 activity and protein in relationship to PGC-1α and mitochondrial content were determined in human skeletal muscle after six weeks of high-intensity interval training.	Increased maximal activities of mitochondrial enzymes in skeletal muscle and PGC-1α protein. Total muscle SIRT1 activity and activity per SIRT1 protein increased despite decreased SIRT1 protein. Exercise-induced mitochondrial biogenesis is accompanied by elevated SIRT1 activity in human skeletal muscle.
2013	Hansen, D., et al.	Slowed exercise-onset VO_2 kinetics during submaximal endurance exercise in subjects with multiple sclerosis	RCT (Cross sectional study)	38 MS patients and 16 healthy controls	Compared exercise-onset and –offset VO_2 kinetics, blood lactate, heart rate, expiratory volume and Borg rate of perceived exertion. Exercise at constant low to moderate intensity (below anaerobic threshold).	Exercise-onset VO_2 kinetics during submaximal endurance exercise are significantly slowed in MS patients → lowered skeletal muscle oxidative capacity.
2015	Hansen, D., et al.	Altered signalling for mitochondrial and myofibrillar biogenesis in skeletal muscles of patients with multiple sclerosis	RCT (cross sectional study)	10 patients with MS and 10 healthy controls (9 PWMS + 7 HC)	Endurance exercise bout → changes in muscle phospho-AMPKα and phospho-mTOR, body composition, muscle strength, exercise	Post-exercise skeletal muscle phospho-AMPKα and phospho-mTOR are increased in MS, and this increase is related to lowered exercise tolerance and greater level of disability.

					tolerance and muscle fibre type.	
2013	Jacobs, R. A., et al.	Improvements in exercise performance with high-intensity interval training coincide with an increase in skeletal muscle mitochondrial content and function	Quasi experimental study without control group	16 untrained adult males	HIIT 6 sessions high-intensity cycling Skeletal muscle respiratory capacity, mitochondrial content, skeletal muscle oxygenation, cardiac capacity, blood volumes and peripheral fatigue resistance assessed prior to and again following training.	Evidence indicating that an improved oxidative capacity of the skeletal muscle resulting from an increase in mitochondrial content may facilitate an improvement in tissue oxygenation, delayed peripheral fatigue, and explain the improvement in both maximal whole body exercise capacity as well as endurance performance following 6 sessions of HIT over a two week period in previously untrained adults.
1997	Kent-Braun, J. A., et al.	Strength, skeletal muscle composition, and enzyme activity in multiple sclerosis	RCT (cross sectional study)	17 subjects: *9 with MS (6 women, 3 men) → EDSS 2-6 (median 4) *8 healthy age-matched controls	Neurological evaluation before participation: EDSS, Ashworth scale of spasticity. Before collection of biopsy samples, symptomatic fatigue (Fatigue Severity Scale), muscle strength (dorsiflexor MVC) anterior compartment CSA (proton	The results of this study suggest that the inherent characteristics of skeletal muscle fibres per se and of skeletal muscle as a whole are altered in the direction of disuse in MS. They also suggest that changes in skeletal muscle in MS may significantly affect function.

					MRI), basal muscle high-energy phosphate status (MRS) and level of physical activity (three-dimensional accelerometer) were assessed. Muscle biopsies of tibialis anterior muscle 1-2 weeks after preceding studies.	
2005	Kumleh, H. H., et al.	Complex 1 deficiency in Persian multiple sclerosis patients	RCT (cross sectional study)	10 MS patients + 11 control subjecs	Muscle specimens from quadriceps by open biopsy under local anaesthesia. Examine mitochondrial respiratory chain complex 1 → activity of it on isolated mitochondria derived from fresh skeletal muscle	Biochemical defect in complex 1activity may be involved in pathogenesis of MS.
2013	Larsen, R. G., et al.	High-intensity interval training increases in vivo oxidative capacity with no effect on P(i)-->ATP rate in resting	Quasi experimental study without control group	Eight healthy young males	V Pi→ATP and Vmax of the vastus lateralis muscle were measured through phosphorus magnetic resonance spectroscopy.	This novel analysis of resting and maximal high-energy phosphate kinetics in vivo in response to HIT provides evidence that distinct aspects of human skeletal muscle metabolism

		human muscle			Measurements were obtained at baseline, 15 hours after the first training session, and 15 hours after completion of the sixth session.	respond differently to this type of training.
2014	Larsen, R. G., et al.	High-intensity interval training alters ATP pathway flux during maximal muscle contractions in humans	Quasi experimental study without control group	8 young healthy men	6 sessions of repeated, 30-s “all-out” sprints on a cycle ergometer. Measures of muscle energetics were obtained at baseline, and after the first and sixth sessions.	Brief, maximal contractions are performed with increased support of oxidative ATP synthesis, and relatively less contribution from anaerobic ATP productions following training. 6 training sessions are sufficient to alter in vivo muscle energetics, which likely contributes to increased exercise capacity after short-term HIT.
2014	Larsen, S., et al.	The effect of high-intensity training on mitochondrial fat oxidation in skeletal muscle and subcutaneous adipose tissue	Quasi experimental study without control group	10 overweight untrained subjects	Effect of HIT on mitochondrial fat oxidation in skeletal muscle and adipose tissue. Mitochondrial oxidative phosphorylation capacity, mitochondrial substrate sensitivity and mitochondrial content → before and after 6	Six weeks of HIT increased VO2peak. Mitochondrial oxidative phosphorylation capacity were increased in skeletal muscle, but not in adipose tissue. Furthermore, mitochondrial fat oxidation was not improved in either skeletal muscle or adipose tissue.

					weeks of HIT	
2011	Little, J. P., et al.	An acute bout of high-intensity interval training increases the nuclear abundance of PGC-1alfa and activates mitochondrial biogenesis in human skeletal muscle	Quasi experimental study without control group	8 young men (habitual active)	Examine molecular processes involved in mitochondrial biogenesis in human skeletal muscle in response to an acute bout of HIT. → muscle biopsies	An acute bout of low-volume HIT activates mitochondrial biogenesis through a mechanism involving increased nuclear abundance of PGC-1α.
2010	Little, J. P., et al.	A practical model of low-volume high-intensity interval training induces mitochondrial biogenesis in human skeletal muscle: potential mechanisms	Quasi experimental study without control group	7 healthy young men	Determine the performance, metabolic and molecular adaptations to a more practical model of low-volume HIT → 6 sessions over 2 weeks → muscle biopsy (CS, COX, Tfam, PGC-1α, GLUT4	Demonstrates that a practical model of low volume HIT is a potent stimulus for increasing skeletal muscle mitochondrial capacity and improving exercise performance. Results suggest that increases in SIRT1, nuclear PGC-1α and Tfam may be involved in coordinating mitochondrial adaptations in response to HIT in human skeletal muscle.
2007	Mathai, A. S., et al.	Rapid exercise-induced changes in PGC-1α mRNA and protein in human skeletal muscle	Randomized crossover design	7 male subjects	Exercise to exhaustion followed by ingestion of either a high-carbohydrate or low-carbohydrate diet for 52 hours of recovery.	PGC-1α protein content increased in prolonged exercise and remained upregulated for 24 hours, but this could not have been predicted by the changes in mRNA.
2009	Perry, C. G., et al.	High-intensity aerobic	Quasi	8subjects (3	Six weeks of HIIT	Six weeks of this unique HIIIT protocol

		interval training increases fat and carbohydrate metabolic capacities in human skeletal muscle	experimental study without control group	females, 5 males)		increases whole body VO2peak, the maximal activities or protein content of five skeletal muscle mitochondrial enzymes, and the content of transport proteins for fatty acids, glucose and lactate as well as resting glycogen. HIIT increases the capacity for both fat and carbohydrate oxidation in muscle.
2012	Stepto, N. K., et al.	Short-term intensified cycle training alters acute and chronic responses of PGC1alfa and cytochrome C oxidase 4 to exercise in human skeletal muscle	Quasi experimental study without control group	9males (healthy and untrained)	Investigated changes in mitochondrial gene expression and protein abundance in response to the same acute exercise before and after 10-days of intensive cycle training. → muscle biopsies	Short-term intensified training promotes increased mitochondrial gene expression and protein abundance. Furthermore, acute indicators of exercise-induced mitochondrial adaptation appear to be blunted in response to exercise at the same absolute intensity following short-term training.
2010	Talanian, J. L., et al.	Exercise training increases sarcolemmal and mitochondrial fatty acid transport proteins in human skeletal muscle	Quasi experimental study without control group	10untrained females	Determined whether high-intensity interval training increased total skeletal muscle, sacrolemmal, and mitochondrial membrane fatty acid transport protein contents → muscle biopsies from	Increases in skeletal muscle fatty acid oxidation following training are related in part to changes in fatty acid transport protein content and localization.

					vastus lateralis (before training, and following 2 and 6 weeks of HIIT)	
2015	Vincent, G., et al.	Changes in mitochondrial function and mitochondria associated protein expression in response to 2-weeks of high intensity interval training	Quasi experimental study without control group	8males (moderately acitve)	Mitochondrial metabolism as well as mitochondrial-associated protein expression were tested in untrained participants performing HIT over a 2-week period.	Over only two weeks HIT significant increased mitochondrial function in skeletal muscle independently of detectable changes in mitochondrial-associated and mitogenic protein expression.
2015	Wens, I., et al.	High intensity exercise in multiple sclerosis: Effects on muscle contractile characteristics and exercise capacity, a randomised controlled trial	RCT	34MS patients	- sedentary control group - 12 weeks of high intensity interval and resistance training -12 weeks of high intensity continuous cardiovascular training + resistance training	12 weeks of high intensity cardiovascular exercise in combination with resistance training was safe, well tolerated and improved muscle contractile characteristics and endurance capacity, with interval training seemingly superior to continuous training.
2014	Wens, I., et al.	Multiple sclerosis affects skeletal muscle characteristics	RCT (Cross sectional study)	34MS patients and 18 healthy controls	- Muscle biopsy (vastus lat.) - DEXA-scan - Isokinetic dynamometer (Biodex)	MS has a negative influence on skeletal muscle fibre CSA, muscle strength and mass of lower limbs → Rehabilitation is needed for muscle preservation of the lower limbs.

Table 9.2: Overview included articles – population, aims and interventions

Authors & journal	population	Aims study	interventions
Bartlett, J. D., et al. J Appl Physiol	10 recreationally active men	Test the hypothesis that acute high-intensity interval running induces greater activation of signalling pathways associated with mitochondrial biogenesis compared with moderate-intensity continuous running matched for work done.	HIT: 6x3min at 90% VO_2max interspersed with 3min recovery periods at 50%VO_2max with 7min warm-up and cool-down period at 70% VO_2max CONT: 50min continuous running at 70% VO_2max
Burgomaster K. A., et al. The Journal of Physiology	20 healthy men and women (active but untrained)	Metabolic adaptations during exercise after traditional endurance (ET) training and high-intensity interval training (SIT).	*6 weeks training programma ET: 40-60 min continuous cycling on workload of 65% Vo2 peak, 5d/w • W1 - 2: 40 minutes • W3 - 4: 50 minutes • W5 - 6: 60 minutes SIT: 4-6 repeats of a 30s 'all out' Wingate test with 4.5min recovery (cycling at low cadence <50rpm against a light resistance of 30w) between bouts, 3d/w • W1 - 2: 4 Wingate tests • W3 - 4: 5 Wingate tests • W5 - 6: 6 Wingate tests
Campbell, G. R., et al. Multiple Sclerosis	17 progressive MS cases and 15 age matched controls	Investigated single fibres in paraspinal muscles from progressive MS cases, known to	Studied muscle (paraspinal) and explored mitochondria in single fibres. Histochemistry, immunohistochemistry, laser microdissection, real-

journal		harbour an excess of mtDNA deletions in the CNS, to determine whether the respiratory enzyme deficiency and mtDNA deletions were also present in excess of age-related changes within muscle.	time polymerase chain reaction (PRC), long range PCR and sequencing were used to resolve the single muscle fibres.
Cochran, A. J., et al. Experimental physiology	17 healthy subjects (8 subjects in study 1, 9 subjects in study 2)	Study 1: Examined whether the activation of signalling cascades linked to mitochondrial biogenesis was dependent on the manner in which an acute high-intensity exercise stimulus was applied. Study 2: Determined whether 6 weeks of the CONT protocol would increase skeletal muscle mitochondrial content to a similar extent to what we have previously reported after 6 weeks of INT	Study 1: Four 30sec Wingate tests interspersed with 4 minutes of rest (INT) or a bout of continuous exercise (CONT) that was matched for total work and which required ≈4min to complete as fast as possible. Study 2: 3 days per week for 6 weeks, for a total of 18 sessions Same as study 1
Edgett, B. A., et al. PLoS One	8 lean healthy men	Determine if increases in PGC-1α induced by higher intensities of submaximal exercise extend to supramaximal exercise by comparing changes in gene expression following three	Three separate sessions of high-intensity interval exercise targeting 73, 100, 133% of their peak aerobic power. These intensities were chosen so that a matched amount of external work would be achieved in 11, 8 and 6 intervals for each intensity, respectively.

		intensities of matched volume exercise. Determine the impact of exercise intensity on changes in gene expression of regulators of PGC-1α activity. Determine if greater increases in PGC-1α gene expression at higher intensities can be explained by intensity-dependent increases in muscle activation.	
Fernström M., et al. J Physiol	17 healthy subjects	The purpose of this study was (i) to investigate the effects of acute exercise and training on the protein expression of UCP3 and ANT, (ii) to investigate the effect of acute exercise and training on UCR in isolated mitochondria, and (iii) to investigate the effect of exercise on the vulnerability of mitochondria to Ca2+ overload.	Subjects were investigated with muscle biopsies before and after acute exercise (75 min of cycling at 70% of ˙VO2peak) or 6 weeks endurance training. Mitochondria were isolated and respiration measured in the absence (UCR or state 4) and presence of ADP (coupled respiration or state 3). Protein expression of UCP3 and ANT was measured with Western blotting.
Gibala, M. J., et al.	6 healthy active young men	Tested the hypothesis that an	Four 30-sec “all out” exercise bouts interspersed with

J Appl Physiol		acute session of brief, intense interval exercise would activate signalling cascades linked to PGC-1α, the transcriptional coactivator that functions as a regulator of mitochondrial biogenesis.	4 minutes of rest.
Gurd, B. J., et al. Applied Physiology nutrition and metabolism	9 healthy subjects: 3 females, 6 males	Examine the changes in SIRT1 activity and SIRT1 protein content in human skeletal muscle after 6 weeks of training and to compare with alterations in PGC-1α content and mitochondrial proliferation.	High-intensity interval training 10x4min intervals at 90%VO_2peak separated by 2 minutes of rest, 3 days per week.
Hansen, D., et al. Neurorehabilitation and Neural Repair	38 pwMS and 16 healthy subjects	Examined exercise-onset and –offset VO_2 kinetics in persons with mild MS when executing two subsequent endurance exercise bouts with similar intensity and duration, and compares these data with healthy subjects.	Exercise-onset and –offset VO_2 kinetics were determined during two 6-minute submaximal bouts of exercise separated by a 6-minute recovery interval.
Hansen, D., et al. Translational Research	Part 1: 14 pwMS compared with 10 healthy subjects Part 2: 9 pwMS compared with 7 healthy subjects	Studied skeletal muscle AMPKα and mammalian target of rapamycin phosphorylation (phospho-mTOR) in pwMS to explore the aetiology for the development of such muscle	Part 1: Assessment of body composition, muscle strength, exercise tolerance and muscle fibre type, muscle phospho-AMPKα and phospho-mTOR. part 2: Endurance exercise bout executed with assessment of changes in muscle phospho-AMPKα

		phenotype.	and phospho-mTOR.
Jacobs, R. A., et al. J Appl Physiol	16 untrained male adults	Perform a comprehensive evaluation of the physiological adaptations, ranging from cardiovascular to skeletal muscle properties, following 6 sessions of HIT in untrained young adults and determine the mechanisms explaining rapid improvements in exercise performance.	8-12 bouts at 100% peak power for 60sec, 75sec active rest between intervals, 2 weeks, 6 sessions.
Kent-Braun, J. A., et al. J Appl Physiol	17 subjects: *9 with MS (6 women, 3 men) → EDSS 2-6 (median 4) *8 healthy age-matched controls	This study examined functional, biochemical, and morphological characteristics of skeletal muscle in nine multiple sclerosis (MS) patients and eight healthy controls in an effort to ascertain whether intramuscular adaptations could account for excessive fatigue in this disease.	Not applicable
Kumleh, H. H., et al. Journal of the Neurological Sciences	10 patients with MS and 11 healthy controls	Because skeletal muscle and the nervous system are among the most energy-dependent tissues of the body with variable ATP demands, the most common forms of mitochondrial disorders are	Kinetic analysis of mitochondrial respiratory chain complex I enzyme was performed on intact mitochondria isolated from fresh skeletal muscle. mtDNA common deletion and deletions were also tested in pwMS.

		encephalomyopathies. Consequently mtDNA common deletion and deletions in MS patients was tested.	
Larsen, R. G., et al. Am J Physiol Regul Integr Comp Physiol	8 untrained men	High intensity interval training (HIT) would increase ATP flux in resting muscle (Vpi→ATP) in response to a single bout of exercise, whereas changes in the capacity for oxidative ATP production (Vmax) would require repeated bouts.	Eight untrained men performed six sessions of HIT in a 2 week period (4–6 x 30-s bouts of all-out cycling with 4-min recovery).
Larsen, R. G., et al. Acta Physiol	8 young healthy men	Examine the effects of HIT on ATP synthesis from net PCr breakdown (ATP_{CK}), oxidative phosphorylation (ATP_{OX}) and non-oxidative glycolysis (ATP_{GLY}) in vivo in vastus lateralis during a 24-s maximal voluntary contraction.	6 sessions of repeated, 30-s “all-out” sprints on a cycle ergometer. Measures of muscle energetics were obtained at baseline, and after the first and sixth sessions.
Larsen, S., et al. Scandinavian journal of medicine and science in sports	Ten overweight untrained healthy subjecs (2 females, 8 males)	Investigate the capacity for lipid oxidation in mitochondria and mitochondrial substrate sensitivity for medium- and long-chain fatty acids in skeletal muscle and subcutaneous adipose tissue after 6 weeks of HIT.	5 x 60seconds exercise bouts (approximately 128% of maximum load), 90 seconds active rest between intervals, after 2 weeks of training the load was increased by 10%, 6weeks, 3 x/week .

Little, J. P., et al. Am J Physiol Regul Integr Comp Physiol	Eight young, healthy and active man	Examine potential mechanisms controlling the adaptive response to HIT by determining the effects of an acute bout of HIT on levels of nuclear PGC-1α. Also obtain a more comprehensive characterization of the response to an acute bout of HIT by examining signalling pathways linked to PGC-1α activation, as well as changes in mRNA and protein expression of mitochondrial genes at selected time points throughout 24 hours of recovery.	4 bouts of 30 seconds all out maximal intensity (0.075kg/kg body mass), 4 minutes of rest between intervals, 1 session.
Little, J. P., et al. Journal physiology	Seven healthy young men	Examine the exercise performance and muscle metabolic adaptations to a more practical model of HIT that is nonetheless still time efficient. Also examined the effect of low-volume HIT on several proposed mediators of mitochondrial biogenesis and metabolic adaptations in skeletal muscle.	8-12 bouts of 60sec at 100% of peak power output separated by 75sec of active recovery, 6sessions, two weeks.

Mathai, A. S., et al. J Appl Physiol	7 recreationally active male subjects	Aim1: the effects of exhaustive exercise (2 hours) Aim 2: the effects of altered changes in muscle glycogen repletion (0–52 h) after exhaustive exercise on PGC-1α mRNA as well as PGC-1α protein in skeletal muscle of healthy men.	Prior to the experimental trials VO_{2max} tests on a stationary, electronically braked cycle ergometer Experimental protocol: 2 trials separated by 2 weeks in which the CHO consumption during the recovery period was assigned by randomized crossover design. Subjects cycled on a stationary cycle ergometer at 65% V˙ O2max until voluntary exhaustion repeated on the second test day, and subjects were instructed to cycle for the same length of time on both days → muscle biopsies vastus lateralis
Perry, C. G., et al. Appl Physiol Nutr Metab	8 untrained recreationally active individuals (5 males, 3 females)	Investigate the ability of 6weeks of HIIT to increase the capacity for skeletal muscle and whole-body carbohydrate and fat oxidation in recreationally active but untrained subjects.	10 x 4min intervals at 90% VO_2peak, 2 minutes of passive rest between intervals, 6 weeks, 3 days per week.
Stepto, N. K., et al. PLoS One	9 healthy untrained male volunteers	Firstly examine changes in expression of mitochondrial genes that are involved in the regulation	60 minutes bout of cycling exercise 10 days of cycling training which included four high-intensity interval training sessions (6x5min; 90-100%

		of mitochondrial biogenesis under exercise conditions that maximally highlighted training adaptions. Secondly, to investigate changes in protein abundance of mitochondrial biogenesis regulators and mitochondrial function including PGC-1α, RIP140 and the electron transport system proteins.	VO_2peak) and six prolongued moderate intensity sessions (45-90min; 75% VO_2peak).
Talanian, J. L., et al. Am J Physiol Endocrinol Metab	10 untrained females (untrained, light recreational physical activity)	Determine whether HIIT increased total skeletal muscle , sacrolemmal, and mitochondrial membrane fatty acid transport protein contents.	VO_{2peak} test: repeated at 2 and 6wk of HIIT 60min Cycling trial at 65% VO_{2peak}: blood samples at rest, 15, 30, 45 and 60min. Repeated at 2 and 6wk of HIIT. Muscle biopsies vastus lateralis 48h following the 60min cycling trails (PRE, 2, 6wk HIIT) 6 weeks of high-intensity interval training (48h following muscle biopsies) 3days/wk, 18sessions in 6wk ten 4-min cycling bouts at 90% VO_{2peak} separated by 2min of rest.
Vincent, G., et al. Frontiers in Physiology	8 male, moderately active participants	Investigate the changes in respiratory fluxes that underlie exercise performance responses to HIT using the permeabilized fibre	12 x 60s intervals at 120% of peak power with 90 seconds rest between intervals on cycle ergometer, 4 days/week, two weeks

		method, as well as the expression levels of proteins associated with mitochondrial function.	
Wens, I., et al. PLoS One	34 MS patients diagnosed according to McDonald criteria (EDSS range 1-5) , aged >18years	Investigate the impact of high intensity interval or continuous cardiovascular exercise, both in combination with resistance training, on muscle contractile characteristics, in terms of muscle fibre CSA/proportion, muscle strength and muscle mass and on endurance capacity in MS.	5 sessions per 2 weeks, sessions were interspersed by at least one day of rest. Each session started with endurance training, followed by resistance training, interspersed by a short resting period. - $H_{IT}R$ program: 5 minutes warming-up on cycle ergometer First 6 weeks duration gradually increased from 5 x1 minute to 5 x 2 minutes and 1 minute rest intervals. → 100% of maximal workload Second 6W: 5 x 2 minutes → 100-120% of max workload second part: moderate to high intensity resistance training. - $H_{CT}R$ program: 1x6min/session → 2x10min/session 80-90% HR_{max} second part: similar resistance training
Wens, I., et al. PLoS One	34 MS patients diagnosed according to the McDonald criteria (EDSS range 0 ,5-6) and 18 matched healthy controls, aged > 18years	Investigate the effect of MS on muscle fibre CSA and proportion, muscle strength and body composition in a larger group of MS patients, compared with HC.	*Muscle biopsies from the middle part of the m. Vastus lateralis (from MS + HC) → Bergström needle technique *Biopsies (MS) from weakest leg → isometric muscle strength test

			*DEXA scan 1-2 weeks before muscle biopsy *Following 5 minutes warming-up on a cycle ergometer and after habituation, the max voluntary isometric muscle strength of knee extensors were measured (45° and 90° knee angle) → isokinetic dynamometer. 2 max isometric extensions (4sec) separated by 30sec rest interval.

Table 9.3: Overview included articles – outcome measures and results

Authors & journal	Outcome measures	results
Bartlett, J. D., et al. J Appl Physiol	Heart rate, rate of perceived exertion and blood lactate	Significantly greater in HIT compared with CONT
	Muscle glycogen	Decreased 30% in both conditions
	Plasma glucose concentration	Increased in both protocols
	Plasma NEFA and glycerol	Significantly greater in the CONT trial
	AMPK phosphorylation	Increased 1.5 fold post-exercise and returned to basal levels at 3h post-exercise
	P38MAPK phosphorylation	Increased 1.9 and 1.5 fold immediately following exercise for HIT and CONT respectively, no significant difference between both protocols
	PGC-1α mRNA	Increased fourfold at 3h following exercise
	PGC-1α protein content	Unchanged in both protocols
	P53 phosphorylation	Tended to increase immediately post-exercise but not significant, 3h following exercise increased 2.7 and 2.1 fold in HIT and CONT trials respectively
	HSP72 and MnSOD mRNA	3- to 4-fold increase immediately post-exercise but not significant
Burgomaster K. A., et al. The Journal of Physiology	Vo2 peak	Increased, no difference between groups
	Peak power output	Increased, no difference between groups
	oxygen uptake	similar before and after training
	mean heart rate and ventilation	decreased, no difference between groups
	mean RER	decreased, no difference between groups
	calculated rates of whole-body fat	increased, no difference between groups
	carbohydrate oxidation rates	decreased, no difference between groups
	CS	increased after training, no difference between groups
	b- HAD	increased after training, no difference between groups

	PGC-1a protein content	increased after training, no difference between groups
	Muscle glycogen content	higher at 60 min of exercise post-training compared to pretraining, no difference between groups
	Net muscle glycogenolysis	reduced after training, no difference between groups
	Muscle PCr content	Higher at 60 min of exercise post-training, no difference between groups
	Muscle ATP	Unchanged by acute exercise, but reduced after 6 weeks op SIT compared to ET
	Lactate accumulation	No significant effects
Campbell, G. R., et al. Multiple Sclerosis journal	Density of respiratory enzyme-deficient fibres	Not significantly different in MS compared with controls
	mtDNA deletion level	Not significantly different in MS compared with controls
	percentage of muscle fibres harbouring high levels of mtDNA deletions	Not significantly different in MS compared with controls
Cochran, A. J., et al. Experimental physiology	Total work	Unchanged
	Ratings of perceived exertion	Unchanged
	Peak power output and mean power output	Higher than the respective values calculated for the CONT trial
	Total exercise duration	In the CONT trial approximately double that of the INT trial
	Muscle glycogen content	Reduced 25%
	Muscle lactate content	Elevated 10-fold
	P38 MAPK phosphorylation	Increased immediately after exercise by 3-fold
	ACC serine-79	Increased immediately after exercise by 2.5-fold
	PGC-1α mRNA	Increased 4-fold from rest after 3h of recovery
	Max activity of CS	Unchanged
	Cytochrome c oxidase subunit IV	Increase of 20%

	GLUT4, MCT1 and MCT4	Unchanged
	VO_2peak	Increased 6%
Edgett, B. A., et al. PLoS One	Peak EMG	Sign. Higher in the 100 and 133% conditions compared to the 73% condition
	PGC-1α mRNA	Elevated after all three conditions, with greater increase following 100% condition
	PDK4	Significant main effect of time with exercise, but no differences between intensity conditions
	SIRT1mRNA	Elevated after all three conditions
	GCN5	Unchanged (negative regulator of PGC-1α activity)
	RIP140 mRNA	Significant main effect of time
Fernström M., et al. J Physiol	VO2 peak + heart rate	Acute: 10% increase Endurance: 24% increase
	Muscle CS activity	Acute: increase with 43% + elevated in recovery state Endurance: increase with 47%
	ADP-stimulated mitochondrial respiration	Acute: coupled respiration (state 3) and UCR were unchanged Endurance: state 3 respiration remained unchanged, whereas state 4 respiration (UCR) decreased by 18%
	Mitochondrial proteins	Acute: UCP3 and ANT remained unchanged Endurance: ANT was increased, UCP3 was not significantly changed
Gibala, M. J., et al. J Appl Physiol	Peak power, mean power and total work	Progressively decreased
	Muscle glycogen content	Lower after exercise and recovery compared with the rest
	Muscle lactate and creatine	Higher compared with the rest after bouts 1 and 4 but not different after recovery
	Muscle phosphocreatine	Lower after bouts 1 and 4 compared with the rest
	Muscle ATP	Only lower after bout 4

	Phosphorylation of AMPK	Higher after bout 4 compared with all other time points
	ACC phosphorylation	Higher after bouts 1 and 4 compared with the rest
	Phosphorylation of p38 MAPK	Higher after bout 4 compared with the rest
	CaMKII phosphorylation	Tended to be higher after bout 4 compared with the rest but not significant
	PGC-1α mRNA expression	Increased approximately twofold above rest after 3h of recovery
	PGC-1α protein content	unchanged
Gurd, B. J., et al. Applied Physiology nutrition and metabolism	VO_2peak	Increased 11%
	Citrate synthase max activity (CS)	Increased 28%
	B-hydroxyacyl-coenzyme A dehydrogenase max activity (β-HAD)	Increased 28%
	COX-IV content	Increased 36%
	SIRT1 activity	Increased 31%
	SIRT1 protein content	Decreased 20%
	PGC-1α	Increased 16%
	Mitochondrial transcription factor A (Tfam)	No change
Hansen, D., et al. Neurorehabilitation and Neural Repair	Total body mass%	Greater in MS
	Leg adipose tissue mass %	Greater in MS
	Exercise cycling power output	Significantly higher in healthy subjects
	HR, % of predicted max HR, expiratory volume and blood lactate content	Similar between groups
	Borg RPEs	Significantly elevated during both exercise bouts in MS patients
	Exercise-onset mean response time	Significantly slower in MS

	Exercise-offset mean response time	Not different between groups or was independently related to having MS
Hansen, D., et al. Translational Research	Subject characteristics (part 1)	Between groups subjects were comparable except for type IIa muscle fibre CSA and VO_{2peak}
	AMPKα and mTOR phosphorylation	Significantly higher in pwMS as opposed to healthy subjects
	Subject characteristics (part2)	Between groups body height, VO_{2peak} and fat mass were different
	W_{max}	Significantly higher in healthy controls
	Exercise intensity, ratings of perceived exertion and caloric expenditure	Comparable between groups
	AMPKα and mTOR phosphorylation	Significantly different between groups before and after endurance exercise Increased in pwMS
Jacobs, R. A., et al. J Appl Physiol	VO_2peak	Increased 7,9%
	Peak power output	Increased 7%
	Time trial performance	Improved
	COX activity	Increased
Kent-Braun, J. A., et al. J Appl Physiol	Biopsies of the tibialis anterior muscle	*fewer type I fibres (66 +- 6 vs. 76 +- 6%) in MS but higher percent of type 2a fibres *fibres of all types were smaller (average <26%) in MS *lower succinic dehydrogenase (SDH average <40%) and SDH/a-glycerol-phosphate dehydrogenase (GPDH) in MS
	Maximal voluntary isometric force for dorsiflexion	associated with both average fibre cross-sectional area (r = 0.71, P = 0.005) and muscle fat-free cross-sectional area by magnetic resonance imaging (r= 50.80, P = 0.001)
	Physical activity, assessed by accelerometer,	was associated with average fibre SDH/GPDH (r= 5 0.78, P =0.008). There was a tendency for symptomatic fatigue to be inversely associated with average fibre SDH activity (r=20.57, P =0.068).

Kumleh, H. H., et al. Journal of the Neurological Sciences	NADH-ferricyanide reductase activities	Significant lower in patients than in control subjects
	Complex I activities	Significant reduced in patients compared with control
Larsen, R. G., et al Am J Physiol Regul Integr Comp Physiol	kPCr (rate of PCr recovery) and Vmax (max. capacity for oxidative phosphorylation)	* single session of HIT: unchanged * completion of six training sessions: 14% increase in muscle oxidative capacity
	VPi→ATP (estimation of unidirectional rate of ATP synthesis)	* single session of HIT: unchanged * completion of six training sessions: unchanged
Larsen, R. G., et al. Acta Physiol	VO_{2peak}	Increased 10%
	Peak force	No main effects of short-term HIT
		[PCr] decreased and [Pi] increased during each contraction [PCr] was lower and [Pi] was higher during the 24-s contraction after training
	Concentration ADP	increased during the contraction with no effect of training
	Concentration ATP	[ATP] at the end of 24-s MVC was not different from [ATP] in resting muscle While ATP production by each pathway was unchanged after the first session, 6 sessions increased the relative contribution of ATP_{OX} , and lowered the relative contribution from both ATP_{CK} and ATP_{GLY}.
Larsen, S., et al.	VO_2 peak	Increased
	Blood pressure	Unchanged
	HbA1c	Decreased

Scandinavian journal of medicine and science in sports	CS activity	Increased 36%
	HAD activity	Unchanged
	mtDNA	Unchanged
	Mitochondrial OXPHOS capacity	Unchanged
	Body weight	Tended to increase
	Fat %	Unchanged
	Lean body mass	Increased
	Fasting blood glucose concentration	Unchanged
	HSL concentration	Increased
	FABPm expression	Tended to increase
	CD36, DGAT, DGAT2, LPL, PLIN5, ATGL	Unchanged
	Mitochondrial lipid OXPHOS capacity	Unchanged
	Complex I-linked OXPHOS capacity	Unchanged
	Complex I + II-linked OXPHOS capacity	Increased
	COX flux	Increased
Little, J. P., et al. Am J Physiol Regul Integr Comp Physiol	Nuclear PGC-1α	Unchanged immediately after exercise, increased 66% at 3h recovery and not sign different from baseline at 24h recovery.
	Whole muscle PGC-1α	Unchanged immediately and 3h after exercise but increased by 57% at 24h of recovery
	PGC-1α mRNA	Unchanged immediately after exercise, increased 750% at 3h recovery and returned to basal levels at 24h
	CS protein content	Unchanged immediately and 3h after exercise but increased 30% at 24h
	COX II protein content	Unchanged immediately and 3h after exercise but increased 29% at 24h

	COX IV protein content	Increased 43% at 3h recovery and 30% at 24h
	P38 MAPK	Higher immediately after exercise, returned to basal levels at 3h recovery and again sign elevated at 24h recovery
	Cytosolic ACC phosphorylation	Increased immediately after exercise
	CS maximal activity	Increased 14% at 24h recovery
	COX maximal activity	Increased 19% at 24h recovery but did not reach statistical significance
Little, J. P., et al. Journal physiology	Time to complete time trials (50kJ and 750kJ)	Improved 11% and 9% respectively
	COX max activity	Increased 29%
	COX II protein content	Increased 35%
	COX IV protein content	Increased 38%
	CS max activity	Increased 16%
	CS protein content	Increased 20%
	PGC-1α in nuclear fractions	Increased 24%
	Whole muscle PGC-1α	Unchanged
	Total SIRT1 content	Increased 56%
	Tfam total protein content	Increased 37%
	NRF-1 protein content	Unchanged
	GLUT4 protein content	Increased 119%
	Resting muscle glycogen	Increased 17%
Mathai, A. S., et al. J Appl Physiol	Blood glucose concentration	Exercise resulted in similar concentrations during both trials. Ingestion of 1 g CHO/kg body wt at the start of recovery and again 1 h later resulted in significant increases in blood glucose and insulin concentrations ($P < 0.05$) within 30 min and throughout the first 2 h of recovery relative to Exh.

	Serum fatty free acids concentration	increased to similar concentrations during the two exercise sessions
	Plasma insuline concentration	Exercise resulted in similar concentrations during both trials. Ingestion of 1 g CHO/kg body wt at the start of recovery and again 1 h later resulted in significant increases in blood glucose and insulin concentrations ($P < 0.05$) within 30 min and throughout the first 2 h of recovery relative to Exh.
	PGC-1α mRNA	- Exhaustive exercise induced a two- to threefold increase in PGC-1_ mRNA relative to rest - PGC-1α mRNA abundance continued to increase to 6.2-fold by 2 h of recovery - By 24 h of recovery, PGC-1α mRNA abundance was repressed and corresponded to levels observed at rest
	PGC-1α protein concentration	- Increased significantly at the end of exhaustive exercise, and it remained elevated at 2 h of recovery, despite the large increase in mRNA at this time. - At 24 h after exercise, the PGC-1α protein was still 16 ± 9% greater (P _ 0.05) than at rest
Perry, C. G., et al. Appl Physiol Nutr Metab	Mean power output	Increased 21%
	Body mass	Unchanged
	VO_2peak	Increased 9%
	Resting concentrations of Pcr, ATP, ADP_f and AMP_f	Unchanged
	CS max activity	Increased 26%
	COX-IV protein content	Increased 18%
	B-HAD max activity	Increased 29%
	FAT/CD36	Increased 16%
	FABPpm	Increased 30%
	FATP4	Unchanged

	m-AspAT and PDHt max activities	Increased 26%
	resting glycogen content	Increased 59%
	GLUT4	Increased 21%
	MCT1 protein content	Increased 14%
	MCT4 protein content	Increased 16%
	PHOSb	Unchanged
	Time to exhaustion	Improved 111%
	Muscle glycogenolysis	Decreased 32%
	Lactate accumulation	Lower
	Substrate phosphorylation	Reduced 20%
	Content ATP	Unchanged
	Muscle pH	Higher
	PDH activity	Unchanged
	lactate	decreased
Stepto, N. K., et al. PLoS One	PGC-1α mRNA	Increased
	PGC-1β mRNA	Decreased
	PRC mRNA	Increased
	Tfam mRNA	Tended to increase by 10% in response to exercise pre-training
	NRF2 mRNA	Unchanged
	Resting cytochrome C mRNA	Increased 26%
	CS mRNA	Unchanged
	COX IV mRNA	Only increased by exercise training
	B-HAD mRNA	Tended to decrease
	PGC-1α protein expression	Increased
	Co-repressor RIP140 expression	increased

	COX I expression	Decreased
	COX IV expression	Decreased
Talanian, J. L., et al. Am J Physiol Endocrinol Metab	Training power output	Increased every week throughout training
	VO_{2peak}	Increased significantly by 11 and 18% following 2 and 6wk, respectively
	RER	Significant lower at all time points following both 2 and 6wk
	Blood lactate concentrations	Increased during pre exercise trial, but not observed following 2, 6wk HIIT
	Whole muscle maximal activity of CPT I	Significantly increased by 30% following 2wk of training, four additional weeks of training resulted of in further increases of 20%
	Whole muscle maximal activity of β-HAD	Significantly increased by 30% following 2wk of training, four additional weeks of training resulted of in further increases of 20%
	Whole muscle maximal activity of CS	Significantly increased by 30% following 2wk of training, four additional weeks of training resulted of in further increases of 20%
	Whole muscle maximal activity of mAspAT	Significantly increased by 30% following 2wk of training, four additional weeks of training resulted of in further increases of 20%
	COX-IV content	Significantly increased by 30% following 2wk of training, four additional weeks of training resulted of in further increases of 20%
	Whole muscle HSL content	Increased with training
	Total muscle and sarcolemmal GLUT4	Increased significantly following 2wk (30%) and increased further following 6wk (50%) of training.
	Whole body fat oxidation	Higher throughout training
	Whole body carbohydrate oxidation	20% decrease following training
Vincent, G., et al. Frontiers in	Peak power	Increased 22%
	Time to fatigue	Increased 33%
	VO_2peak	No significant increase
	CS activity	Increased 43%

Physiology	CYT-C	Increased 21%
	PGC-1α protein expression	No significant effect
Wens, I., et al. PLoS One	Muscle fibre CSA and proportion	Unchanged in SED Mean CSA increased in $H_{IT}R$ and $H_{CT}R$ Muscle fibre type I CSA increased in $H_{CT}R$ whereas muscle fibre type II and IIa increased in $H_{IT}R$ Fibre type IIx CSA did not change No changes in fibre type proportion observed
	Isometric muscle strength	Remained stable in SED Knee flexion and extension strength of the weakest leg improved in $H_{IT}R$ compared to SED Only hamstring strength of the strongest leg improved with $H_{IT}R$ $H_{CT}R$ flexion and extension strength improved in weakest leg, unchanged in strongest leg
	Endurance capacity	Unchanged in SED and $H_{CT}R$ W_{max}, test duration and VO_{2max} significantly improved in $H_{IT}R$
	Body composition	Body weight remained stable Body fat % tended to decrease within $H_{IT}R$ and $H_{CT}R$ Lean tissue mass significantly increased within $H_{IT}R$, unchanged in $H_{CT}R$ and SED
	Physical activity level	Compared to SED, PA level of $H_{IT}R$ and $H_{CT}R$ significantly increased Unchanged in SED

Wens, I., et al. PLoS One	skeletal muscle fibre cross sectional area and fibre type proportion	Compared to HC, mean muscle fibre CSA, as well as CSA of type I,II an IIa fibres were significantly smaller in MS patients ($p<0.05$), whereas muscle fibre CSA of type IIx was comparable between both groups. Type II fibres experienced a larger atrophy, compared to type I fibres in MS ($p<0.05$). Compared to women, men had higher CSA for almost all fibre types in both HC and MS. Compared to HC, fibre type I proportion tended to be lower in MS ($p=0.1$), whereas type IIa proportion tended to be higher ($p=0.1$).
	Body composition	There were no differences between total body weight, adipose and lean tissue mass of MS and HC. Compared to HC, the lower limb, of which the muscle tissue was collected tended to have a higher fat percentage ($p=0.1$) and a lower lean mass ($p=0.06$) in MS.
	Isometric muscle strength of the quadriceps	Compared to HC, MS patients showed reduced isometric muscle strength of the quadriceps of the biopsied leg (-22%, $p<0.05$).

Table 10: list of abbreviations

ACC	acetyl-coenzyme A carboxylase
AMP	adenosine monophosphate
AMPK	adenosine monophosphate-activated protein kinase
ATP	adenosine triphosphate
CONT	continuous
COX	cytochrome c oxidase
CS	citrate synthase
CSA	cross sectional area
HIIT	High intensity interval training
HIT	High intensity training
HRQOL	health related quality of life
INT	interval
MS	Multiple sclerosis
p38-MAPK	p38 mitogen-activated protein kinase
PGC-1α	peroxisome proliferator-activated receptor gamma coactivator 1-alpha
Phospho-AMPKα	activated protein kinase phosphorylation
pwMS	Patients with multiple sclerosis
RCT	Randomized controlled trial
RER	Respiratory exchange ratio
$VO2_{peak}$	peak rate of oxygen consumption
WOS	Web Of Science

Table 11: individual accomplishments

	Tobias Severijns	**Ferdy Wijckmans**
Period 1: (21/09/2015 - 08/11/2015)	- Composing search strategy - Searching articles	- Composing search strategy - Searching articles
Period 2: (09/11/2015 - 03/01/2016)	- Composing search strategy - Searching articles	- Composing search strategy - Searching articles
Period 3: (04/01/2016 - 14/02/2016)	- Composing search strategy - Searching articles - Completion first search	- Composing search strategy - Searching articles - Completion first search - Start writing “Method”
Period 4: (15/04/2016 - 10/04/2016)	/	/
Period 5: (11/04/2016 - 22/05/2016)	- Start writing “Results” - Double check method, tables (1, 2, 3, 4) and figures (1, 2). - Table 5: Cochrane checklist for randomized controlled trials - Table 6: Quality assessment standard for a cross-over study - Table 7: Self-made checklist for quasi experimental studies without control group - Table 8: Checklist for cross-sectional studies	- Repeat search strategy - Finish writing “Method” - Start writing “Introduction” - Table 1: List of search terms used in PubMed - Table 2: List of search terms used in Web of Knowledge (WOK) - Figure 1: Flowchart in- and excluded articles of pwMS - Figure 2: Flowchart in- and excluded articles of healthy subjects and pathologies similar to MS - Table 3: Reasons of exclusion concerning pwMS - Table 4: Reasons of exclusion concerning healthy subjects and pathologies similar to MS - Double check tables (5, 6, 7, 8)
Period 6: (23/05/2016 - 30/06/2016)	- Finish writing “Results”. (50%) - Double check “Introduction” - Table 9.1 (75%): Overview included articles - type of article, subjects, used	- Finish writing “Results”. (50%) - Finish writing “Introduction” - Start writing “Research protocol” (part 2)

	techniques and conclusion - Table 9.2 (75%): Overview included articles - population, aims and interventions - Table 9.3 (75%): Overview included articles - outcome measures and results	- Table 9.1 (25%): Overview included articles - type of article, subjects, used techniques and conclusion - Table 9.2 (25%): Overview included articles - population, aims and interventions - Table 9.3 (25%): Overview included articles - outcome measures and results
Period 7: (01/07/2016 - 24/07/2016)	- Writing "Discussion" - Writing "Abstract" - Writing "Research framework" - Table 10: list of abbreviations - Double check "Research protocol" (part 2)	- Double check "Discussion", "Abstract" and "Research framework" - Writing "Highlights" - References part 1 and 2 - Finish writing "Research protocol" (part 2) - Table 11: individual accomplishments

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Table 12: Progress form

DATUM	INHOUD OVERLEG	HANDTEKENINGEN
16/10/2015	* Kennismaking promotor * Afspraken omtrent masterproef met promotor * Tekenen contract masterproef	Promotor: Prof. Dr. Bert Op 't Eijnde Student(e): Ferdy Wijckmans Student(e): Tobias Severijns
03/11/2015	* Kennismaking copromotor * Afspraken omtrent uitwerking masterproef met copromotor	Copromotor: Dr. Wens Inez Student(e): Ferdy Wijckmans Student(e): Tobias Severijns
08/02/2016	* Presentatie literatuurstudie masterproef * Bespreking voortgang * Vastleggen verdere afspraken	Promotor: Prof. Dr. Bert Op 't Eijnde Copromotor: Dr. Wens Inez Student(e): Ferdy Wijckmans Student(e): Tobias Severijns
24/06/2016	* Bespreking voortgang * Laatste afspraken * Vastleggen Deadlines	Promotor: Prof. Dr. Bert Op 't Eijnde Copromotor: Dr. Wens Inez Student(e): Ferdy Wijckmans Student(e): Tobias Severijns

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Ik/wij verlenen het wereldwijde auteursrecht voor de ingediende eindverhandeling:
Effect of High-Intensity Interval Training on Mitochondrial Density in Skeletal Muscles of MS Patients

Richting: **master in de revalidatiewetenschappen en de kinesitherapie-revalidatiewetenschappen en kinesitherapie bij neurologische aandoeningen**
Jaar: **2016**

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